A Phase 2/3 Study for the Evaluation of Safety and Efficacy of Humacyte's Human Acellular Vessel for Vascular Replacement or Reconstruction in Patients with Life or Limb-threatening Vascular Trauma

Study No.: CLN-PRO-V005

Investigational Product: Human Acellular Vessel (HAV)

Study No.: CLN-PRO-V005

IND No.: 15263

Version Date: 24May2023

Version: 4.0

Sponsor: Humacyte Global, Inc.
2525 East NC Highway 54
Durham, NC 27713

Sponsor Approval:

Shamik Parikh Digitally signed by Shamik Parikh
Date: 2023.05.24 17:11:31 -04'00'

Signature Date (dd/mmm/yyyy):
Printed Name: Shamik Parikh, MD
Title: Chief Medical Officer, Humacyte Global, Inc.
Telephone: 1 (919) 313-9633 ext 130
email: sparikh@humacyte.com

Confidentiality Statement

REVISION HISTORY

Protocol Version	Active Region(s)	Date Finalized
Original Protocol	USA	29 December 2015
Version 2.0	USA	24 October 2016
Version 3.0	USA, Israel	18 July 2018
Version 3.1	For potential Poland sites – not implemented.	22 June 2021
Version 3.2	USA	18 August 2021
Version 3.3	USA (added pediatric enrollment)	12 August 2022
Version 4.0	USA (Master protocol) **Rationale for key revisions:** - Incorporate modifications requested by the Agency during the March 2022 meeting (March 30, 2022 (Reference IND 16746 CRMTS #13939 Meeting Summary March 30, 2022 Type B OIND Teleconference) and during the March 2023 Type C pre-BLA meeting (reference): o Separate analyses for the group of patients with non-iatrogenic arterial trauma of the extremities, and the rest of the patients enrolled (patients with iatrogenic and torso injuries). o Inclusion of pediatric patients and clarification of category of eligible pediatric patients o Modification of statistical analysis section to include While-on-Treatment analysis methodology and define the estimands. - Serious Adverse Event definition updated to align with industry standards and FDA guidance	24 May 2023

STATEMENT OF COMPLIANCE

This trial will be conducted in compliance with the protocol and the following regulatory requirements:

- Declaration of Helsinki adopted by the 18th World Medical Assembly in Helsinki, Finland, in 1964, as last amended by the World Medical Assembly in 2013
- International Conference on Harmonization of Technical Requirements for Registration of Pharmaceuticals for Human Use (ICH), E6 Good Clinical Practice: Consolidated Guidance (ICH E6)
- ICH E2A Clinical Safety Data Management: Definitions and Standards for Expedited Reporting
- ICH E8 Guidance on General Considerations for Clinical Trials
- Applicable sections of United States Food and Drug Administration (FDA) Code of Federal Regulations (CFR), including:
    - 21 CFR Part 11, Electronic Records; Electronic Signatures
    - 21 CFR Part 50, Protection of Human Subjects
    - 21 CFR Part 54, Financial Disclosure by Clinical Investigators
    - 21 CFR Part 56, Institutional Review Boards
    - 21 CFR Part 312, Investigational New Drug Application

STUDY SITE PRINCIPAL INVESTIGATOR AGREEMENT

Protocol Title: **A Phase 2/3 Study for the Evaluation of Safety and Efficacy of Humacyte's Human Acellular Vessel for Vascular Replacement or Reconstruction in Patients with Life or Limb-threatening Vascular Trauma**

Study No.: CLN-PRO-V005

Version: 4.0

I agree:

- To assume responsibility for the proper conduct of the study at this site, and to conduct the study in compliance with this protocol, any future amendments, and with any other study conduct procedures provided by Humacyte Global, Inc. (Humacyte) or its authorized representatives.
- Not to implement any deviations from or changes to the protocol (including protocol amendments) without agreement from Humacyte and prior review and written approval from the Institutional Review Board (IRB) and relevant regulatory authorities, if applicable, except where necessary to eliminate an immediate hazard to the patient(s), or for administrative aspects of the study (where permitted by all applicable regulatory requirements).
- That I am familiar with the appropriate use of the investigational product, as described in this protocol and any other information provided by Humacyte including, but not limited to, the current Investigator Brochure (IB) or equivalent document.
- To ensure that all persons assisting me with the study are adequately informed about the investigational product and trained on all study-related duties and functions.
- That I have been informed that certain regulatory authorities require Humacyte to obtain and supply details about the investigator's ownership interest in Humacyte or the investigational product (IP), and more generally about his/her financial ties with Humacyte.
- That I have been informed that Humacyte will use and disclose the information solely for the purpose of complying with regulatory requirements.

Principal Investigator: _____ _____
Signature Date

Printed Name and Title: _____

Institution Name: _____

Institution Address: _____

_____

PROTOCOL SYNOPSIS

Title of Study:	A Phase 2/3 Study for the Evaluation of Safety and Efficacy of Humacyte's Human Acellular Vessel for Vascular Replacement or Reconstruction in Patients with Life or Limb-threatening Vascular Trauma
Protocol Number:	CLN-PRO-V005
Study Phase:	Phase 2
Name of Sponsor/Company:	Humacyte Global, Inc.
Planned Study Sites:	Up to 35 sites in the United States (US), Israel, and Ukraine will be recruited to enroll patient.
Planned Sample Size:	Up to 100 patients
Study Population:	Patient with vascular trauma to size appropriate vessels in the limb or torso, requiring replacement or reconstruction.
Inclusion Criteria:	1. Patient with life or limb threatening traumatic injury to an arterial vessel in the limb or torso, other than the heart, which requires replacement or reconstruction. 2. Preoperative imaging or clinical examination indicates the damaged vessel has a defect length of ≤ 38cm and is appropriately size matched to the 6mm Human Acellular Vessel (HAV) per the judgment of the treating surgeon taking into account vasoconstriction and situational inflow and outflow considerations. 3. Autologous vein graft is either not feasible in the judgment of the treating surgeon (e.g., because of lack of availability of suitable conduit, presence of severe venous insufficiency) or is not desirable because of the urgency of revascularization. 4. Adults aged 18 to 85 inclusive (all sites) and adolescents who have achieved Tanner Sexual Maturity Rating Stage V (US sites Only). 5. Able to communicate meaningfully with investigative staff and able to comply with study procedures. If the patient is unconscious, then information from a reliable witness indicates that the patient would normally be able to understand and comply with study procedures. 6. Patient or legal representative is able, willing and competent to give informed consent. 7. Life expectancy of at least 1 year.
Exclusion Criteria	1. Mangled Extremity Severity Score (MESS) of ≥ 7. 2. Limb at high risk of amputation despite vascular reconstruction (e.g., because of crush injury). 3. Catastrophic injuries that make survival unlikely (e.g., Abbreviated Injury Scale (AIS) > 5 or Injury Severity Score (ISS) >60). 4. HAV may not be used for coronary artery repair. 5. Women known to be pregnant. 6. Known medical condition which would preclude long term antiplatelet therapy after resolution of acute injuries. 7. Any other condition which in the judgment of the investigator would preclude adequate evaluation of the safety and efficacy of the Humacyte Human Acellular Vessel (HAV).

<table>
<tr><td></td><td>8. Previous exposure to HAV.
9. Known participation in any investigational study within the last 30 days.
10. Employees of the sponsor or patients who are employees or relatives of the investigator.</td></tr>
<tr><td>Expected Enrollment Start</td><td>3Q2018</td></tr>
<tr><td>Accrual Period:</td><td>Enrollment will close on or before December 31, 2024.</td></tr>
<tr><td>Study Duration</td><td>The active study duration for each study participant will be 36 months from HAV implantation. All patients will be followed for the initial 12 months. Beyond 12 months (Long-Term Follow Up), only patients with a patent HAV will be followed out to a total of 36 months from HAV implantation. The total expected duration of the clinical study is 61 months.</td></tr>
<tr><td>Study Design</td><td>Prospective, multicenter, non-randomized study</td></tr>
<tr><td>Investigational Medicinal Product/Intervention Description</td><td>Patients will be implanted with a Humacyte Human Acellular Vessel (HAV) as an interposition replacement or bypass using standard vascular surgical techniques.</td></tr>
<tr><td>Primary Objectives and Endpoints</td><td>
<table>
<tr><th colspan="2">Primary Safety</th></tr>
<tr><th>Objectives</th><th>Endpoints</th></tr>
<tr><td>To determine infection rates of the HAV</td><td>HAV Infection</td></tr>
<tr><td>To evaluate the safety and tolerability of the Humacyte HAV in vascular trauma patients following surgery for vascular replacement or reconstruction due to life- or limb-threatening trauma of the extremities</td><td>Adverse Events</td></tr>
<tr><th colspan="2">Primary Efficacy</th></tr>
<tr><th>Objectives</th><th>Endpoints</th></tr>
<tr><td>To determine the primary patency of the HAV at 30 days in vascular trauma patients following surgery for vascular replacement or reconstruction due to life- or limb-threatening trauma of the extremities</td><td>Primary HAV patency for at least 30 days after implantation</td></tr>
</table>
</td></tr>
</table>

Secondary Safety Objectives and Endpoints

Key Secondary Safety	
Objectives	**Endpoints**
To determine mechanical stability of the HAV based on freedom from aneurysmal degeneration, anastomotic bleeding or spontaneous rupture, infection, or significant stenosis.	Adverse events of special interest HAV infection
To determine the durability of the HAV repair in terms of freedom from interventions needed to maintain or restore HAV patency.	Adverse events HAV interventions
To determine amputation rate	Amputations
To determine mortality	Death
To determine the rate of HAV removal	Partial or complete removal of HAV
To determine the rate of post-operative surgical site infection	Post-operative surgical site infection

Secondary Efficacy Objectives and Endpoints

Key Secondary Efficacy	
Objectives	**Endpoints**
To determine the patency of the HAV at least 30 days, regardless of interventions	Patency (Secondary) for at least 30 days after implant
To determine the ability of the HAV to remain infection free for 30 days	Conduit infection in 30 days after implant
To determine the rate of limb salvage for 30 days	Amputation in 30 days after implant
Secondary Efficacy	
To determine the long-term limb salvage	Amputation
To determine the long-term patency of the HAV, regardless of interventions	Patency (Secondary)
To determine the long-term ability of HAV to stay infection free	HAV infection
To determine the rates of interventions needed to maintain/restore patency in the HAV	HAV interventions to maintain/restore patency
To determine patient survival	Death
To evaluate remodeling of the HAV	Histopathology of any clinical explants

Protocol Approval	Version 4.0 Approved 24May2023

Figure 1 Study Schematic

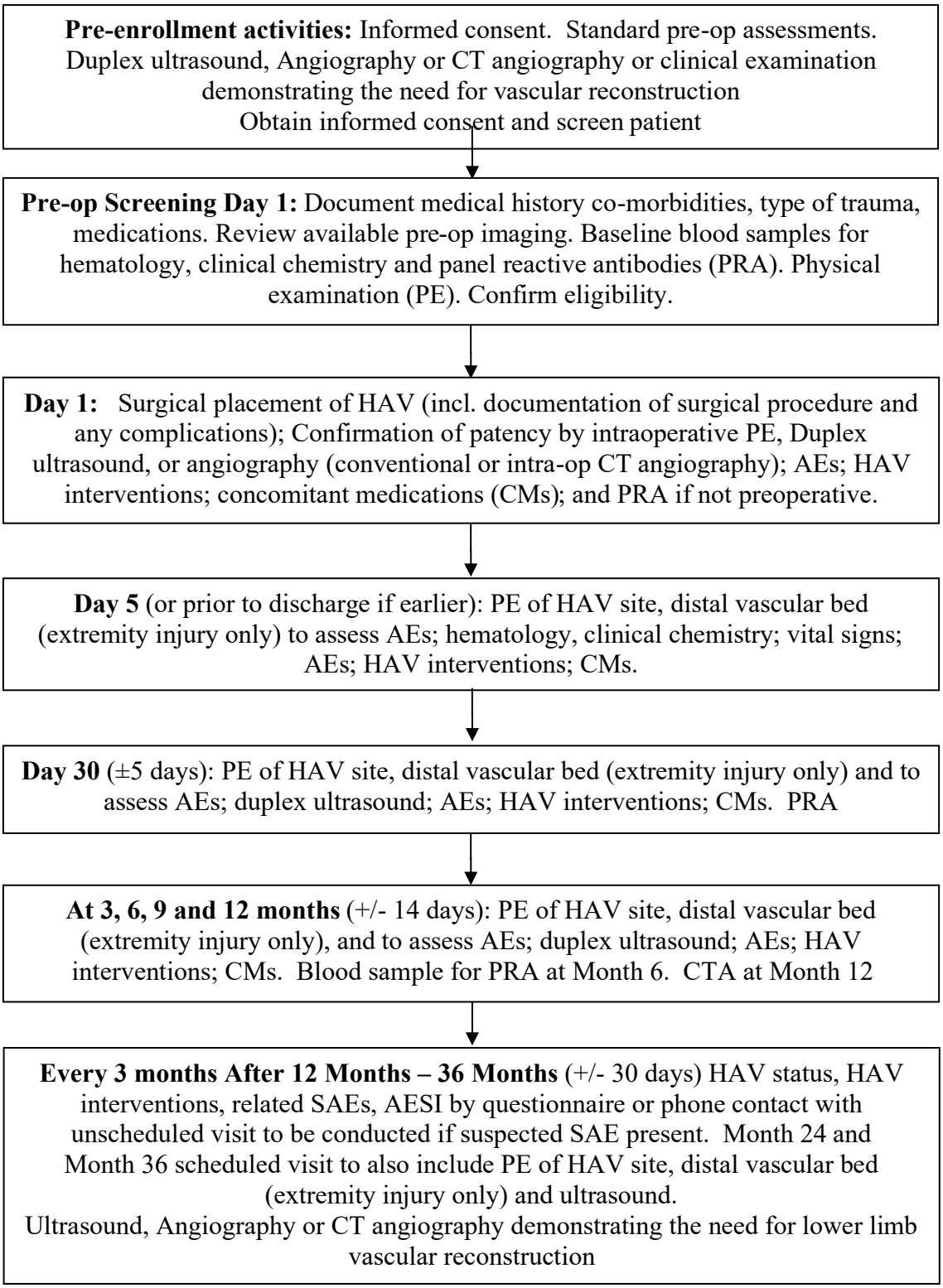

Table 1 Schedule of Visits and Assessments

	Pre-op screening D1	D 1	D 5 or prior to d/c	D 30 ± 5 days	M3 ± 14 days	M 6 ± 14 days	M 9 ± 14 days	M12/ET† ± 14 days	M15-M36 †± 30 days
Informed consent	X								
Medical history and nature of trauma	X								
Concomitant medication	X	X	X	X	X	X	X	X	
Physical exam [1]	X	X	X	X	X	X	X	X	X [7]
Pre-op Ultrasound or CT angiography [2]	X								
Vital signs			X						
Eligibility (inclusion/exclusion criteria)	X								
HAV implantation and intraoperative confirmation of patency [3]		X							
Documentation of surgery and any complications		X							
Clinical chemistry	X [5]		X						
Hematology	X [5]		X						
PRA	X [5]			X		X		X [6]	
Duplex Ultrasound [4]				X	X	X	X	X	X [7]
CT angiography								X	
AEs		X	X	X	X	X	X	X	X [8]
Documentation of HAV interventions		X	X	X	X	X	X	X	X[8]

Abbreviations: AEs, adverse events; D, day; d/c, discharge; ET, early termination; HAV, human acellular vessel; M, month; PRA = panel reactive antibodies PRA = panel reactive antibodies.

1. Physical examination includes clinical exam of the operative limb and HAV at all post-operative visits (incl. patency assessment on D1) and physical exam to evaluate AEs; include distal vascular bed (extremity injury only)

2. Pre-op imaging is at the discretion of the investigator based on the clinical condition of the patient. It may be omitted if not deemed necessary in order to proceed to vascular surgical repair.
3. Determination of intraoperative HAV patency can be done by physical exam, Doppler, angiography or ultrasound at the discretion of the investigator.
4. An alternative imaging method (CTA, MRI, etc.) may be substituted for duplex ultrasound at the discretion of the investigator if it is medically appropriate and in the best interest of the patient.
5. Measured at preoperative screening when possible; if PRA titer not obtained preoperatively, it must be completed within 12 hours after HAV implantation.
6. PRA only collected at ET visit if the visit occurs before Month 6 collection.
7. Visits at month 24 and month 36 to be conducted in person with physical exam of the HAV site and duplex ultrasound imaging of HAV.
8. The status of the patient and HAV will be ascertained every 3 months post Month 12 until 36 months after HAV implantation by telephone contact with the patient and/or his physician. Only related SAEs and all AESI will be reported after 12 months. If a suspected SAE related to HAV is discovered an unscheduled visit should be conducted to investigate.

† Patients withdrawn before Month 12 will perform ET visit that correlates with the procedures at Month 12. Patients withdrawn after Month 12 and prior to Month 36 should complete an ET visit that correlates with procedures post Month 12 through Month 36.

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LIST OF ABBREVIATIONS AND DEFINITIONS OF TERMS

Abbreviation	Definition
AE	Adverse Event
FDA	Food and Drug Administration
GCP	Good Clinical Practice
GLP	Good Laboratory Practice
HAV	Human acellular vessel
HIV	Human immunodeficiency virus
IB	Investigator Brochure
ICF	Informed consent form
ICH	International Conference on Harmonization of Technical Requirements for Registration of Pharmaceuticals for Human Use
IgG	Immunoglobulin G
IHC	Immunohistochemistry
IM	Intramuscular
IMP	Investigational medicinal product
INR	International normalized ratio
IRB	Institutional Review Board
ISO	International Organization for Standardization
IU	International unit
IV	Intravenous
MedDRA	Medical Dictionary for Regulatory Activities
M	Month
NYHA	New York Heart Association
OTC	Over-the-counter
PAD	Peripheral arterial disease
PE	Physical examination
PHI	Protected health information
PI	Principal Investigator
PRA	Panel reactive antibodies
PT	Prothrombin time
PTFE	Polytetrafluoroethylene
QA	Quality Assurance

Abbreviation	Definition
QC	Quality Control
RRT	Renal replacement therapy
SAE	Serious adverse event
SFA	Superficial Femoral Artery
SOP	Standard operating procedure
SVS WIfI	Society for Vascular Surgery: Wound, Ischemia, and foot Infection
US	Ultrasound
USA	United States of America
WFI	Water for injection
WBC	White blood cell(s)
WHO	World Health Organization

1. STUDY PERSONNEL

An updated list of all study personnel will be maintained by the CRO.

Protocol amendments will not be required for staff changes at Humacyte, the CRO or the sites (except change of Principal Investigator at a site).

Key Study Management Personnel

CRO Safety Representative:

Todd Fisher
Drug Safety Manager
Atlantic Research Group
2421 Ivy Road
Charlottesville, VA 22903

Email: safety@atlanticresearchgroup.com
Phone: 888-619-3216

Medical Monitor:

Jennifer Prichard, MD
Medical Monitor
Atlantic Research Group
2421 Ivy Road
Charlottesville, VA 22903

Email: jprichard@atlanticresearchgroup.com
Mobile: 434-249-5530
Phone: 888-619-3216

2. BACKGROUND INFORMATION AND SCIENTIFIC RATIONALE

2.1. Background Information

In the civilian population, traumatic vascular injuries are mainly concentrated to the limbs and torso (abdomen, retroperitoneum, thorax and thoracic outlet). According to the PROspective Observational Vascular Injury Treatment (PROOVIT) database, designed to collect vascular trauma injuries from 24 Level I and Level II trauma centers in the United States, vascular injuries in the lower limb, torso, upper limb and neck have a distribution of 41%, 30%, 22%, and 6%, respectively (Dubose, 2015; Faulconer, 2018). These reports incorporate a significant percentage of relevant venous injuries in addition to arterial. Additionally, in the civilian population, lower extremity bone fractures with associated arterial injuries are common, due to motor vehicle accidents, gunshot wounds, dog bites, and other situations resulting in blunt or penetrating trauma (Akingba, A. George, 2012; Akingba, A. G., 2013; Andrikopoulos, 1995; Helfet, 1990). In fact, the incidence of this type of vascular injury has increased considerably in the past 50 years (Andrikopoulos, 1995). Although this type of injury represents less than 1% of all civilian injuries, fractures with associated vascular damage require special attention because of their potentially severe complications, including limb necrosis and amputation.

Currently, in order to attempt to salvage the injured limb or end-organ in the distal vascular bed and to prevent life threatening hemorrhage the vascular component of these injuries is treated with interposition or bypass grafting. This type of reconstruction is performed by using either autologous vein from the patient (typically great saphenous vein), or by using a synthetic graft, such as ePTFE or Dacron. However, the use of these grafts in the civilian population is not always possible or without additional risks. The patient may not have adequate autologous vein for harvest, and in many of these cases, such as dog bite injuries, the wound is “dirty” and a synthetic vascular substitute is contraindicated due to the risk of infection (Akingba, A. George, 2012; Akingba, A. G., 2013). Thus, civilians would benefit from a vascular graft that does not contain infection-prone synthetic material and has similar properties as human tissue but does not require time consuming or high morbidity procedures to harvest vessels from the patient.

Military personnel could also benefit from an off-the-shelf biologic vascular graft. In modern combat, the incidence of vascular injury is much greater than in previous wars. The rate of vascular injury in the Vietnam War was 2-3%. But between 2002-2009, the rate of vascular injury was over 12% in a study of over 13,000 battlefield injuries. In combat scenarios vascular injuries occur in all locations of the body, but injuries to the lower extremities are the most common, followed by vascular injuries to the upper extremity, and then neck, followed by torso. (Rasmussen, 2018). Improvised Explosive Devices, or IED’s, caused more than 3,000 casualties per year during recent conflicts (Cordesman and Greenough, 2010; Cordesman and Lemieux, 2010). Arterial damage, laceration and thrombosis can require vascular reconstruction to save tissues from ischemia, necrosis, and further amputation. Harvesting autologous vein for vascular reconstruction is problematic because IED casualties often have multi-limb injuries, making harvest of autologous vein highly risky or impossible (Holcomb, 2010). Synthetic vascular reconstruction using synthetic vascular grafts made from Teflon (ePTFE)/Dacron is relatively

contraindicated, since IED wounds are always "dirty", and bacteria in the wound can colonize the synthetic graft, causing abscesses and sepsis.

Thus, there is a significant unmet medical need for alternative grafts, which can be used in situations where autologous vein is unavailable or undesirable to use and which more closely mimic human vascular tissue to avoid or reduce the infection complications associated with ePTFE and Dacron.

2.2. Scientific Rationale

Humacyte has developed an acellular, human tissue engineered vascular conduit, the human acellular vessel (HAV, to provide an alternative to synthetic materials and to autologous grafts in the repair of traumatic vascular damage. Because this product mimics native vascular tissue, it may possess the advantages of an autologous graft; it also has the benefits of synthetic grafts in that it is available off-the-shelf. Use of an off-the-shelf product avoids the surgical morbidity associated with vein graft harvest and most importantly allows vessel bypass surgery in patients who have no suitable vessels available. In addition, the HAV may not exhibit the compliance mismatch associated with synthetic alternatives. In addition, pre-clinical studies in pigs, canines and primates have shown that the HAVs resist intimal hyperplasia at the anastomoses (Dahl, 2011; Prichard, 2011; Quint, 2011). Upon implantation, the collagen matrix comprising the HAV is infiltrated with host cells and remodeled by the host. This could result in a vascular structure more similar to the histological composition of the native vascular tissue; this remodeling may improve bypass longevity and make the HAV less likely to become infected. The latter potential advantage is of critical importance in the repair of peripheral vascular trauma where most wounds are heavily contaminated.

2.3. Summary of Nonclinical Information

The nonclinical testing program was designed to comprehensively address:

- local and systemic effects of the product in multiple in vivo animal models both acutely and chronically,
- functional aspects of product implanted into animal models as an arteriovenous conduit, and
- biocompatibility of the HAV material in standardized in vitro and in vivo test protocols.

Overall, the results of these studies indicated that the HAV extracellular matrix material was nontoxic, well-tolerated, and met standards for biocompatibility. Generally, the HAVs functioned as intended and maintained patency during the implantation period. See the HAV Investigator Brochure for a detailed summary of nonclinical data.

Prior to implantation, the HAV has mechanical properties (burst pressure and suture retention strength) comparable with native human arteries and veins (Table 2) There were no observations indicative of deterioration of HAV strength after long-term implantation into baboons.

Table 2 Summary of Mechanical Properties of Explanted Acellular Vessels

Test Material	Burst Pressure (mm Hg)	Suture Strength (g)
Pre-Implant Humacyte HAVs	3415 ± 1011 (n=4)	180 ± 44 (n=12)
Post-Explant Humacyte HAVs	3669 ± 1305 (n=5)	276 ± 84 (n=11)
Human Saphenous Vein	1,680 – 2,273[a]	196 ± 2 (n=7)[a]
Human Artery	2,031 – 4,225[a]	200 ± 119 (n=9) [a]

a From L'Heureux et al, Nature Medicine, 2006. (L'heureux, 2006)

In the chronic animal studies, Humacyte vessels equivalent to the HAV but produced using canine cells were implanted into 12 dogs (canine acellular vascular graft, CAVG) in a variety of anatomical locations. The human HAV was similarly implanted into 14 baboons. In general, the Humacyte vessels were safe and well tolerated, and functioned as intended.

Mechanical failure was not observed in any HAV. Calcification was not observed in any CAVG or HAV. No graft exhibited hemodynamically significant intimal hyperplasia. Unlike with ePTFE graft implantation, no evidence of systemic infection attributable to implantation of HAV was observed in any of the animals. One HAV developed an aneurysm that was resected and did not harm the animal. The HAV material showed no evidence of toxicity in hematology, clinical chemistry, or necropsy data. The HAVs could be accessed by venipuncture and hemostasis was achieved after needle puncture.

Under microscopic analysis, the HAVs were found to be well-integrated into the host tissue. Overall, the cellular host response to the HAVs demonstrated smooth muscle actin-positive cells within the vessel wall, endothelial cells lining the lumen, and an adventitial-like outer layer adjacent to the vessel. These findings indicate that implanted HAVs were populated with cell types that are characteristic of healthy native vasculature. Examination of the anastomotic sections showed that the HAVs were well integrated with adjoining vasculature, with minimal intimal hyperplasia observed. Furthermore, immunohistochemistry (IHC) was employed to identify $CD68^+$ macrophages in the venous intimal tissue. Studies have shown a substantial macrophage population has been observed within venous intimal tissue adjacent to inflammatory ePTFE arteriovenous grafts (Kelly, 2002; Roy-Chaudhury, 2001). Only sparse $CD68^+$ macrophages were observed, indicating that the degree and the aggressiveness of the intimal hyperplasia associated with the HAV were less than that typically associated with ePTFE grafts (Prichard, 2011).

Over time, the organization and composition of extracellular matrix (ECM) components indicated that, aided by infiltration of host vascular cells, HAVs were remodeled in vivo in a manner that mimicked the dynamic remodeling process of native blood vessels. Given the difficulties associated with the baboon animal model, where mismatches in vein vs graft diameter were encountered and animals perturbed their wounds postoperatively, an overall assisted patency rate of approximately 80% (11/14) was achieved. In a xenogeneic transplant model that did not employ immunosuppression, the HAV material did not elicit biologically significant cellular or delayed-type hypersensitivity (DTH) immune responses. All animals

developed immunoglobulin G (IgG) titers to the HAV material that did not appear to detrimentally impact vessel function.

These data collectively support the safety of the HAV for the proposed clinical investigation.

2.4. Summary of HAV Clinical Studies

Human studies are ongoing in the United States (US), Europe, and Israel. In all, eight clinical studies were initiated to evaluate three clinical indications for use of the HAV. Seven studies completed their planned follow-up times with three continuing to follow patients long-term (CLN-PRO-V001, CLN-PRO-V002, CLN-PRO-V004, and CLN-PRO-V007). One clinical study NCT03005418 (EudraCT #2020-003383-12) or CLN-PRO-V005 (i.e., V005) that is evaluating HAV for repair of vascular trauma is continuing to enroll patients. As of 10 April 2022, a total of 374 patients with ESRD, 35 patients with PAD, and 51 patients with vascular trauma have had the HAV implanted. In addition, an HAV has been implanted in over 30 cases under an Expanded Access program. The first implant for hemodialysis was performed in December 2012, the first for peripheral arterial bypass in October 2013, and the first implant for trauma in September 2018. Across all clinical studies the total treatment exposure to HAV is approximately 1,005 patient-years, with approximately 830 patient-years, 119 patient-years, and 56 patient-years in ESRD, PAD, and vascular trauma patients, respectively.

More information on the clinical profile of the HAV in these ongoing studies is provided in the Investigators Brochure.

2.4.1. Experience in Patients Undergoing Peripheral Arterial Bypass

Humacyte has two phase 2 studies to assess the safety and efficacy of the HAV when used as an above-knee arterial bypass graft. The first study, CLN-PRO-V002, is a single group uncontrolled study conducted at 3 sites in Poland that is fully enrolled and in long-term follow up. Eligible patients required a femoro-popliteal bypass graft for the management of symptomatic peripheral arterial disease. Pre-operative imaging (conventional or CT angiography) must have demonstrated at least two below knee vessels patent to the ankle with good runoff. The proximal anastomosis was expected to be below the inguinal ligament and the distal anastomosis above the knee. Autologous vein grafts must not have been suitable or feasible (e.g., because of severe venous disease or prior use of leg veins for other bypass surgery or there is a clinical need to preserve those veins for future bypass surgery in the coronary or peripheral circulation).

The HAV was implanted using standard vascular surgical techniques and the patency of the bypass confirmed by intraoperative angiography (conventional or intra-op CT angiography) or ultrasound. The patient was then followed up at study visits at 15 days, 6 weeks and 3, 6, 12, 18 and 24 months. At each visit safety was assessed by clinical examination and adverse events, and the HAV was examined using duplex ultrasound to visualize the entire length to confirm patency, flow and to detect stenosis, aneurysm development or dilatation.

The primary objectives of the study are to evaluate the safety and tolerability of the Humacyte HAV in PAD patients undergoing above-knee femoro-popliteal bypass surgery and to determine the patency (primary, primary assisted and secondary) rate of the Humacyte HAV at 24 months.

Secondary objectives include assessment of the panel reactive antibodies (PRA)) and IgG response to the HAV and to assess patency (primary, primary assisted and secondary) at 6, 12 and 18 months, to determine the rates of interventions needed to maintain / restore patency in the HAV, to assess any effect of implantation on claudication, rest pain and ischemic ulcers and to assess any effect on ankle-brachial index (ABI).

The second PAD study of similar design, CLN-PRO-V004, is being conducted in the US and long-term follow-up is currently ongoing.

CLN-PRO-V002 Study Results (72 M)

Recruitment began in October 2013 and was completed in June 2014 with 20 patients implanted. Thirteen patients completed the 2-year follow up visit. Of the seven patients terminating the study early, three died and four were withdrawn after occlusion of the HAV. None of the deaths were considered related to the HAV or implantation procedure.

Kaplan-Meier (K-M) analyses in which deaths were censored revealed primary, primary assisted, and secondary patency probability rates of 79.2%, 79.0%, and 89.5% at Week 26, 63.3%, 63.2%, and 84.2% at Month 12, 63.3%, 63.2%, and 79.0% at Month 18, and 58.1%, 57.9%, and 73.7% at Month 24.

Six patients (30%) required at least 1 graft intervention to maintain or restore HAV patency during the study. Four patients required 1 intervention and 1 patient each required 3 and 4 interventions. Most interventions successfully restored patency. However, in 1 patient the graft patency could not be restored and the HAV was replaced with an alternative bypass graft. Two patients, who had previously undergone successful interventions, developed a recurrent thrombosis which was not treated and the HAV was left occluded. Two patients experienced HAV thrombosis with no or minimal symptoms and refused interventions on the HAV.

All 20 patients experienced AEs (a total of 92 events). Thirty-one of these events in 13 patients were considered serious. The most frequent AEs reported included graft thrombosis (35% of patients), anastomotic stenosis (20% of patients), lymphocele (20% of patients), and local swelling (15% of patients). Those SAEs reported by at least 2 patients were graft thrombosis (6 patients, 30%) and anastomotic stenosis (2 patients, 10%).

No patient showed an increase in PRA levels. Two patients had a significant (>2 fold) increase from baseline in IgG levels. One of these patients experienced a thrombosis of the HAV between 3 and 6 months after implantation, while the other patient has had no HAV-related AEs and continues to have primary patency. Neither patient has had any evidence of dilatation or structural degeneration of the HAV.

At six years (72 months), six patients have completed follow-up, with five of six patients retaining HAV patency (i.e., primary patency = 4 and secondary patency = 1), which was confirmed by Doppler ultrasound. Three of the long-term patients died during follow-up; one from cancer at 71 months and two from other non-HAV-related causes at 54 months and approximately 67 months post-implant (Gutowski, 2023). Also, one patient voluntarily withdrew from the study at 61 months post-implant. During long-term follow-up (i.e., post 24 months), one patient had two thrombectomies performed. For all remaining patients, no interventions and no infections were documented. Overall, 7 deaths were recorded at the 72-month endpoint, for a

mortality rate of 35% in this patient cohort (i.e., severe PAD with 7-year follow-up). No deaths were attributable to the HAV.

In conclusions, this study demonstrated that

- Humacyte HAV was safe and well tolerated in patients with PAD.
- The HAV is able to withstand long term use in a high pressure, high outflow resistance arterial circuit.
- Patency rates for the HAV are within the ranges of patency rates of synthetic and autologous grafts presented in the literature.
- The HAV was not immunogenic.

2.4.2. Experience in Patients Undergoing Hemodialysis

Two phase 2 trials, one in Poland (CLN-PRO-V001) and one in the US (CLN-PRO-V003) have completed enrollment. Both recruited patients requiring hemodialysis access for end-stage renal disease who were not suitable for creation of an AVF. Most patients had undergone previous vascular access procedures, in many cases multiple attempts including both AVFs and synthetic grafts. Initial results from these Phase 2 studies are discussed below.

The primary objectives of these two studies are to evaluate both the safety of HAV and its efficacy in terms of primary and secondary patency at 6 months. Secondary objectives include measurement of a panel of reactive antibodies (PRA) response, development of IgG antibodies to the extracellular matrix material in the HAV, the evaluation of patency 2 years after implantation, and an assessment of the need for interventions to maintain or restore patency. Follow-up has now been extended up to 120 months.

A Phase 3 randomized study comparing HAV with ePTFE grafts (CLN-PRO-V006) has completed enrollment in the US, Europe, and Israel. A second Phase 3 randomized study (CLN-PRO-V007) comparing HAV with AVF has also completed enrollment in the US. As the sponsor is blinded to treatment allocation, no efficacy information currently available for these Phase 3 studies; however, blinded safety data is presented in the Investigator Brochure.

CLN-PRO-V001 and CLN-PRO-V003 Study Results

All patients (n=60) have now completed at least 24 months since implantation (or had a censoring event). The first patients recruited are now beyond 60 months after HAV implantation, some with functioning HAV for hemodialysis access. Together these two trials provide more than 150 years of follow up during which the HAV has been used for more than 15,000 hemodialysis sessions.

- When HAV thrombosis has occurred, it has almost always been managed successfully, often allowing immediate resumption of dialysis without the need for the placement of a dialysis catheter.
- One non-serious arteriovenous graft aneurysm was reported in Study CLN PRO V001 (moderate in intensity, considered possibly related to the HAV and considered not related

to the implantation procedure – this patient died before the Sponsor could complete the follow up of this event).

- An expected number of small pseudoaneurysms have been observed, which is consistent with all surgically-created hemodialysis access. Most have resolved spontaneously, with only 2 cases requiring surgical intervention. Flow rates through the HAV were more than sufficient to allow for effective dialysis.

In both studies, the product has generally been well tolerated and blood chemistry, hematology and coagulation data are not indicative of any HAV-associated toxicity. Immunogenic response to the HAV material has not been observed as demonstrated by a general lack of HAV-related change in PRA levels (Class I or II). Three patients had elevations in their PRA levels: all 3 patients had experienced one or more renal transplant failures; one patient recently; one patient developed septic shock about a month before the elevated value; and the third patient, who was severely debilitated with a decubitus ulcer, died approximately a month after HAV abandonment.

IgG titers increased in 5 patients; in 4 cases the IgG titer increased and then decreased while the HAV remained functional with no clinical evidence of an inflammatory response; in one case the IgG titer increase occurred in a patient who maintained primary patency.

Adverse events (AEs) related to the HAV / access site (excluding thrombotic events) were few; there have been only three access-site infections, of which only one required removal of part of the HAV. There have been:

- 1 transplant (known to be functioning well at 12 months post-transplant)
- 15 deaths, all after abandonment or during follow-up; none of the deaths were considered related to the presence of the HAV

Patency data for the two studies in dialysis access has been pooled for a combined K-M survival analysis (Lawson, 2016). Based on these K-M plots the patency at 6, 12 and 24 months is estimated to be 60%, 26% and 15% (primary patency) and 97%, 91% and 77% (secondary patency).

2.5. Human Acellular Vessel Host Response and Remodeling Data

Humacyte has been able to assess the general host response to the HAV in 8 human participants; this was accomplished through the microscopic examination of explanted HAV and adjoining tissue samples that were obtained during surgical revision procedures (Kirkton, 2018; Kirkton, 2019; Lawson, 2016). The analysis (mostly of a section close to the venous anastomosis) included assessments of:

- Cellular infiltration of histotypic, inflammatory and immunological populations.
- Extracellular remodeling processes, including neosynthesis and reorganization of ECM components representative of those typically occurring in native blood vessels.

In these cases, small segments of the HAV and adjacent vascular tissue were explanted, fixed in formalin solution and shipped to Humacyte for analysis. Implant duration ranged from 16 to 55 weeks (median: 37 weeks).

In humans, the HAV remodeled in a manner consistent with that observed in primate studies. There was infiltration of cell populations that are normally associated with angiogenesis, vascular organization, and structure; namely, cells with endothelial, smooth muscle and fibroblastic phenotypic characteristics were observed. Endothelial cells formed a monolayer on the luminal surface of the HAV. Migration of actin-positive smooth muscle cells into the wall of the HAV was consistently observed. A well-vascularized adventitial layer of non-constricting fibrous tissue formed around the HAV implant. Infiltration of the graft material by inflammatory and immunoreactive cell populations was either not evident or was mild and generally unremarkable. Degradation or breakdown of the implant was not observed.

Histotypic neosynthesis and reorganization of the ECM was observed in patterns indicative of integration of the HAV into the host. An increase in the density of collagen type I, the main type of collagen found in the wall of native blood vessels, was apparent in the majority of HAV explant specimens. The structure of collagen type I in these specimens exhibited a mature, organized pattern, with distinct fibers and a prominent circumferential alignment evident in explanted samples compared to pre-implant specimens. In some specimens, the fibrillar staining pattern of collagen III became more prominent and more organized, with a circumferential orientation. Fibronectin levels and staining patterns remained unchanged.

Cannulation sites within the HAV appeared to be repaired by the host in a fashion similar to wound repair in the body (Figure 2). In one case, an explanted specimen was tested for suture retention strength at the time of explant and exhibited a substantial increase over the pre-implant level.

Based on this evidence, the HAVs were remodeled by the host after implantation to form a vascular-like structure similar to the histological appearance of native vasculature. The HAVs were repopulated by cell types that are characteristic of healthy native vasculature. Evidence of ECM remodeling processes, including neosynthesis and reorganization of ECM components of a type that typically occurs in native blood vessels were observed in the explanted HAV. The cellular infiltration and ECM remodeling patterns were indicative of the integration of the HAV into the host physiology.

Figure 2 Images of Mid-Vessel Segment Explanted at 11 Months After HAV Implantation

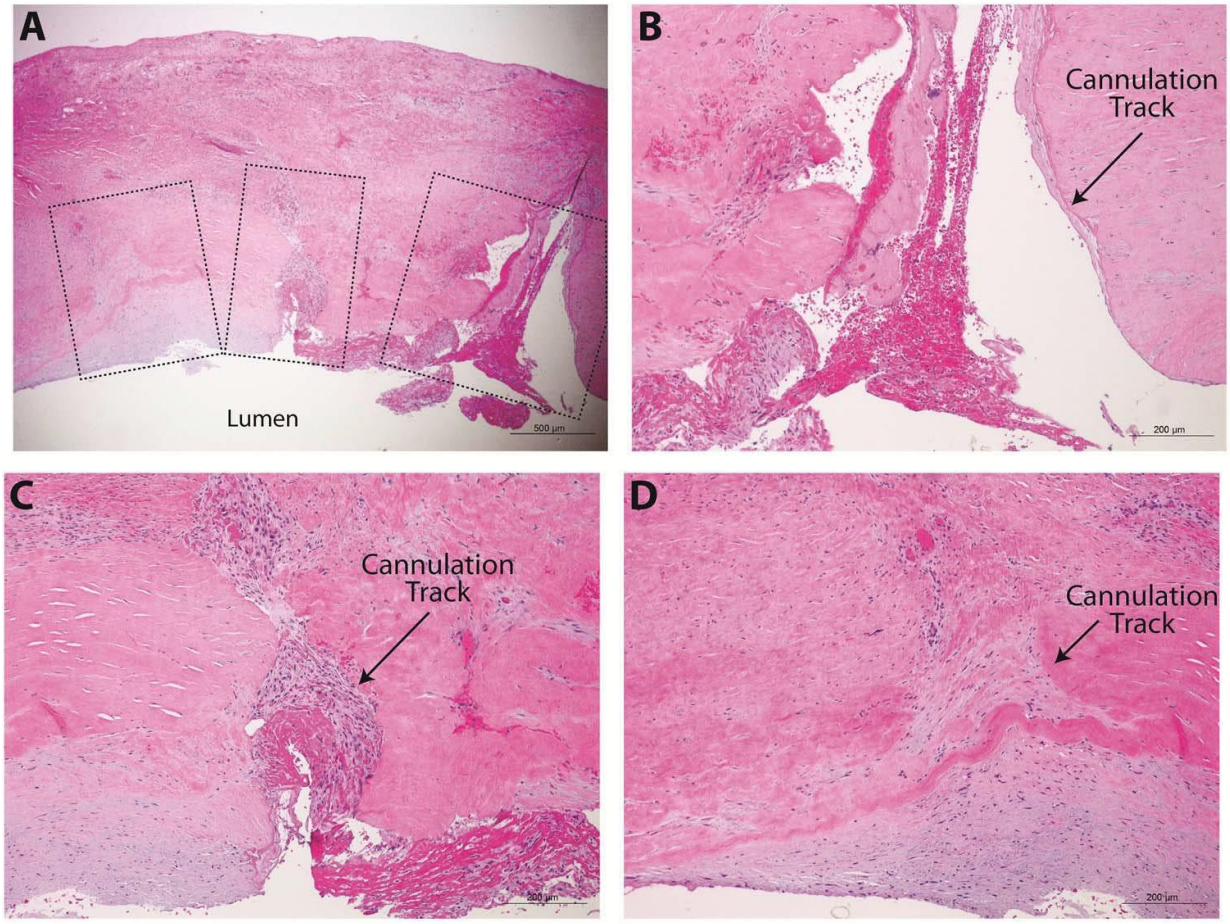

Figure 2. Histopathology images of mid-vessel HAV segment explanted 11 months after HAV implantation. Panel A: Low magnification showing 3 cannulation sites (in dashed boxes) with several prior cannulation tracts from dialysis access; Panel B: Fresh cannulation track with a fresh clot extending into the tract from the lumen; Panel C: Cannulation track during remodeling; and Panel D: shows an older cannulation track that has been repaired. Panels C and D both show partially healed cannulation tracts, with evidence of cellular repopulation extending in from the lumen. Remodeled cannulation tracks contain new collagen and a few micro-conduits (Kirkton, 2019; Lawson, 2016).

Conclusions

Clinical experience indicates that the HAV remains mechanically strong over implantation periods of more than 60 months with no evidence of dilatation. During more than 200 patient-years of follow up across the three phase 2 studies only one case of infection of the HAV material itself has been reported. The serious adverse event (SAE) profile has been typical of that expected in the dialysis and PAD populations. In hemodialysis populations, secondary patency of the HAVs is substantially higher than the historical data for both ePTFE and AVF (accounting for non-maturation). In PAD, patency is in line with historical ePTFE and autologous conduit for above knee bypass. No evidence of immunogenicity of the HAV has been found and the HAV remains mechanically robust even after repeated puncture for hemodialysis and under high pressure, high outflow resistance in arterial reconstruction.

These data support the use of HAV in future Phase 2 and Phase 3 studies for vascular replacement and reconstruction in diseased or damaged (trauma) vessels.

2.6. Potential Risks and Benefits

This is a first-in-human study in which the HAV will be used to repair vascular injuries within the torso (thorax, thoracic outlet, abdomen, and retroperitoneum) as well as the extremities (upper and lower limbs).

Risks associated with HAV use in vascular repair specific to these locations have not been specifically characterized. However, these risks are not expected to be significantly different than those experienced/reported when used in the upper or lower extremity for AV access or arterial reconstruction.

The Data Monitoring Committee (DMC) will review safety data of the torso injuries on the earlier of when the first torso patient reaches 3 months post-implantation or when the first 2 torso patients have both reached 30 days post-implantation. Overall recruitment will be restricted to a maximum of 100 patients who receive implants.

This is also the first time the HAV will be used to repair vascular injuries in a pediatric population. Because enrollment in this study is limited to older adolescents, who have reached pubertal development Tanner Stage V, these risks are not expected to be significantly different than those experienced/reported in adults.

2.6.1. Potential Risks

It is anticipated that patients participating in the study will be exposed to similar risks to those associated with other arterial conduits. Risks associated with the study investigational product may include but are not limited to:

- Thrombosis/occlusion of the conduit or host vessels, with consequent limb ischemia
- Embolism from a thrombosed conduit
- Bleeding and hematoma formation at the surgical site
- Infection – at the surgical site or systemically
- Stenosis of the conduit or its anastomoses
- Aneurysm or pseudoaneurysm formation
- Swelling of the limb
- Failure/injury to the target end-organ
- Bleeding/hemorrhage in the peritoneum or retroperitoneum

The HAV is grown using donor human aortic smooth muscle cells; the vessel is decellularized during manufacturing and thus consists of human extracellular matrix proteins. It is possible that the HAV may provoke an immune response which may lead to damage to the HAV and possible cross reactivity against host proteins. Possible antibody formation will be assessed by

determining and analyzing concentrations of panel-reactive antibodies (PRA) at various times during the study.

2.6.2. Potential Benefits

Patients who undergo implantation of the Humacyte HAV may benefit from improved patency and a reduced number of interventions versus a conventional ePTFE or Dacron graft.

- This may result from a decreased propensity for anastomotic and downstream neointimal hyperplasia, which often leads to graft occlusion with synthetic grafts.
- In addition, risks of infection typically encountered with conventional synthetic grafts may be decreased with the Humacyte HAV.
- Finally, the longevity (secondary patency) of the Humacyte HAV may be greater than that of conventional synthetic grafts.

Patients may also benefit from reduced morbidity secondary to harvest of autologous vessels for vascular reconstruction. Harvest of autologous vascular conduit requires additional time to perform and in the urgent/emergent trauma scenario, time is of the essence and is key to positive patient outcomes. Use of saphenous, or other autologous vessels for purposes of vascular repair in young, otherwise healthy patients prevents its future potential use for other indications (e.g., coronary bypass, etc.) in the future.

Vascular reconstruction using synthetic vascular grafts (ePTFE, or woven materials like Dacron) may be contraindicated for use in trauma cases, as the wounds are often contaminated and allow bacteria in the wound to colonize the synthetic graft. Such colonization may potentially lead to abscesses, hemorrhage secondary to anastomotic blow out, blood stream infection, and sepsis. Unlike other readily available synthetic vascular grafts made from ePTFE or Dacron, HAVs are comprised of human proteins, resulting in grafts that may be less prone to infection than synthetic materials, as shown in preclinical studies (Kirkton, 2018).

2.6.3. Risk-Benefit Rationale

The risks anticipated in this study are similar to those associated with currently marketed prosthetic grafts used for vascular repair. The potential advantages of the HAV compared to currently marketed grafts may lead to a lower complication rate and reduced need for surgical intervention and graft replacement as well as the potential for reduced secondary complications associated with autologous vessel harvest.

This is the first in man study in which the HAV will be used to repair vascular injuries within the torso (thorax, thoracic outlet, abdomen, and retroperitoneum) and so risks related to those locations have not been specifically characterized. However, these risks are not expected to be significantly different than those experienced or reported when used in the upper or lower extremity for AV access or arterial reconstruction.

This is the first in man study in which the HAV will be used to repair vascular injuries in a pediatric population. Because enrollment in this study is limited to older adolescents who have reached pubertal development Tanner Stage 5, these risks are not expected to be significantly different than those experienced/reported in adults.

3. STUDY OBJECTIVES

3.1. Primary Objectives and Endpoints

The primary objectives and the corresponding endpoints of this study are provided in Table 3.

Table 3 Primary Objectives and Endpoints

Primary Safety	
Objectives	**Endpoints**
To determine infection rates of the HAV	HAV Infection
To evaluate the safety and tolerability of the Humacyte HAV in vascular trauma patients following surgery for vascular replacement or reconstruction due to life- or limb-threatening trauma of the extremities	Adverse Events
Primary Efficacy	
Objectives	**Endpoints**
To determine the primary patency of the HAV at 30 days in vascular trauma patients following surgery for vascular replacement or reconstruction due to life- or limb-threatening trauma of the extremities	Primary HAV patency for at least 30 days after implantation

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3.2. Secondary Objectives and Endpoints

The secondary objectives for Safety and the corresponding endpoints of this study are provided in Table 4.

Table 4 Secondary Safety Objectives and Endpoints

Secondary Safety	
Objectives	**Endpoints**
To determine mechanical stability of the HAV based on freedom from aneurysmal degeneration, anastomotic bleeding or spontaneous rupture, infection, or significant stenosis	Adverse events of special interest HAV infection
To determine the durability of the HAV repair in terms of freedom from interventions needed to maintain or restore HAV patency	Adverse events HAV interventions
To determine amputation rate	Amputation

To determine mortality	Death
To determine the rate of HAV removal	Partial or complete removal of HAV
To determine the rate of post-operative surgical site infection	Post-operative surgical site infection

The secondary objectives for Efficacy and the corresponding endpoints of this study are provided in Table 5.

Table 5 Secondary Efficacy Objectives and Endpoints

Key Secondary Efficacy	
Objectives	**Endpoints**
To determine the patency of the HAV at least 30 days, regardless of interventions	Patency (Secondary) for at least 30 days after implant
To determine the ability of the HAV to remain infection free for 30 days	Conduit infection in 30 days after implant
To determine the rate of limb salvage for 30 days	Amputation in 30 days after implant
Secondary Efficacy	
To determine the long-term limb salvage	Amputation
To determine the long-term patency of the HAV, regardless of interventions	Patency (Secondary
To determine the long-term ability of HAV to stay infection free	HAV infection
To determine the rates of interventions needed to maintain/restore patency in the HAV	HAV interventions to maintain/restore patency
To determine patient survival	Death
To evaluate remodeling of the HAV	Histopathology of any clinical explants

4. STUDY DESIGN

4.1. Description of the Study Design

This is a prospective, open-label, non-randomized, multicenter Phase 2/3 study. No formal hypothesis testing is planned.

Injuries to both the limbs and torso have been permitted in the study. Moving forward after approval of this version, patients with extremity injuries will be the main target for enrollment.

Vessels of the heart are excluded. Examples of size-appropriate vessels include (but are not limited to):

- Axillary
- Brachial
- Basilic
- Popliteal
- Femoral
- Subclavian
- Brachiocephalic/innominate
- Celiac
- Hepatic
- Splenic
- Superior mesenteric
- Renal
- Iliac

4.2. Overall Structure of the Study

A schematic illustrating the overall study structure as well as the sequence and timing of planned assessments is provided in Figure 1; the corresponding schedule of assessments is provided in Table 1.

4.3. Study Endpoints

Endpoints will be assessed for up to 36 months after HAV implantation. The primary analysis of the study will be conducted when the earlier of these two milestones are reached:

a) when the last patient enrolled reaches 30 days after implantation, or

b) all patients enrolled in the initial 24-month accrual period have reached 30 days after HAV implantation.

The objectives, endpoints and the corresponding estimands for this study have been updated for this version of the protocol as described in the corresponding Statistical Analysis Plan (SAP).

- The primary Safety and Efficacy endpoints of this study are provided in Table 3.
- The secondary Safety endpoints of this study are provided in Table 4.
- The secondary Efficacy endpoints of this study are provided in Table 5.

4.4. Duration of Study Participation

The active study duration for each study participant will be 36 months from HAV implantation. All patients will be followed for the initial 12 months. Beyond 12 months (Long-Term Follow-up), only patients with a patent HAV will be followed out to a total of 36 months from the date of HAV implantation. The total expected duration of the clinical study is 61 months.

5. STUDY POPULATION

5.1. Description of the Study Population

The study population will consist of patients with vascular trauma to size appropriate vessels in the limb or torso, requiring replacement or reconstruction.

Overall recruitment will be restricted to a maximum of 100 patients who receive an HAV implant.

5.1.1. Patient Inclusion Criteria

Patients must satisfy all of these criteria to be eligible for enrollment into the study:

1. Patients with life or limb threatening traumatic injury to an arterial vessel in the limb or torso, other than the heart, which requires replacement or reconstruction.
2. Preoperative imaging or clinical examination indicates the damaged vessel has a defect length of ≤ 38 cm and is appropriately size matched to the 6 mm diameter of the HAV per the judgment of the treating surgeon, taking into account vasoconstriction and situational inflow and outflow considerations.
3. Autologous vein graft is either not feasible in the judgment of the treating surgeon (e.g., because of lack of availability of suitable conduit, presence of severe venous insufficiency) or is not desirable because of the urgency of revascularization.
4. Adults aged 18 to 85 inclusive (all sites) and adolescents who have achieved Tanner Stage V Sexual Maturity Rating (US sites only).
5. Able to communicate meaningfully with investigative staff, and able to comply with entire study procedures. If the patient is unconscious, then information from a reliable witness indicates that the patient would normally be able to comply with study procedures.
6. Patient or legal representative is able, willing and competent to give informed consent.
7. Life expectancy of at least 1 year.

5.1.2. Patient Exclusion Criteria

Patients satisfying any of these criteria will not be eligible to enroll into the study:

1. Mangled Extremity Severity Score (MESS) of ≥ 7.
2. Affected limb is at high risk of amputation despite vascular reconstruction (e.g., because of crush injury).

3. Catastrophic injuries that make survival unlikely (e.g., Abbreviated Injury Scale (AIS) > 5 or Injury Severity Score (ISS) > 60).
4. HAV may not be used for coronary artery repair.
5. Women known to be pregnant.
6. Known medical condition which would preclude long-term antiplatelet therapy after resolution of acute injuries.
7. Any other condition which in the judgment of the investigator would preclude adequate evaluation of the safety and efficacy of the HAV.
8. Previous exposure to HAV.
9. Known participation in any investigational study within the last 30 days.
10. Employees of the Sponsor or patients who are employees or relatives of the Investigator.

5.1.3. Enrollment of Adolescents who have achieved Tanner Stage V Sexual Maturity Rating (US sites only)

The study population includes patients who are assessed to have reached Tanner Stage V by the health care provider. As in adults, vascular injuries in this pediatric population can be treated with the HAV only if the vascular trauma involves size-appropriate vessels, per the judgement of the treating surgeon.

6. INVESTIGATIONAL MEDICINAL PRODUCT

6.1. Product Description

The Investigational Medicinal Product (IMP) is the Humacyte Human Acellular Vessel (HAV), which is a tissue-engineered vascular prosthesis for vascular bypass or reconstruction in patients with peripheral vascular disease or peripheral vascular trauma.

- The HAV is a sterile, non-pyrogenic, acellular tubular vessel composed of human collagen Type I and Type III, along with other extracellular matrix proteins.
- The HAV is 6 mm in diameter and approximately 42 cm in length.
- The product is supplied on a silicone mandrel immersed in sterile phosphate-buffered saline (PBS) in a sealed and labeled plastic container.
- There is no placebo control or comparator group in this study.

6.2. Manufacturer of the Human Acellular Vessel

Initially, the HAV was manufactured for Humacyte by:

Allosource
6278 S. Troy Circle
Centennial, CO 80111 USA

Since 01July2021, all HAV are now manufactured by:

Humacyte Global, Inc. (previously known as Humacyte, Inc.)
2525 East NC Highway 54
Durham, NC 27713

Traceability of the HAVs during and after the clinical investigation will be achieved by the assignment of lot numbers. A unique identifying lot number is assigned to each vessel.

6.3. Packaging, Storage, and Labeling

Packaging: Each HAV is contained in a sealed, flexible plastic primary container closure system that was developed by Humacyte. The system meets container/closure requirements to maintain sterility as well as product and fluid integrity. The vessel is contained inside the system in a fixed manner, immersed in a sterile, phosphate buffered saline. The total volume of the storage solution is approximately 300 mL.

Storage: The product is shipped under controlled conditions to maintain temperature at 4ºC (range: 2 – 8ºC). The product should be stored in a refrigerator that maintains this temperature range. The HAV must NOT be allowed to freeze.

Labeling: The HAV will be labeled according to applicable guidelines and relevant regulatory agency requirements. A tamper-resistant label affixed to the secondary container will be used to ensure that the product is not compromised prior to use.

6.4. Implantation of the Human Acellular Vessel (HAV)

The HAV is implanted using standard vascular surgical techniques that are similar to placement of autologous or synthetic peripheral vascular prostheses (see the V005 Study Manual for details).

Tunneling for implantation of the HAV, if required, must be performed using a sheathed tunneler. After inserting the assembled tunneler into the tissue, the inner mandrel of the tunneler should be removed from the sheath. The sheath is lubricated with saline and then with the silicone mandrel in place; the HAV can be easily pushed through the sheath without the need to tie to the inner mandrel and is then pulled through the tunneler (see the V005 Study Manual for details).

After placement, HAV patency and integrity are checked by pressurizing the conduit. Prior to completion of surgery, HAV patency is confirmed by physical examination, Doppler ultrasound evaluation, angiography (conventional or intra-operative CT angiography), or ultrasound. The surgical site is closed using standard techniques.

Implantation of an HAV will be undertaken by qualified vascular surgeons experienced in peripheral vascular surgery.

6.5. HAV Accountability Procedures

Documentation of receipt, dispensing, and return of all HAVs must be maintained by the Principal Investigator or his/her designee. It is the Principal Investigator's responsibility to

ensure that all HAVs are kept in a secure location, with access limited to individuals authorized by the Investigator. The product will be shipped with the HAV Shipment Confirmation Form. Once signed, the form should be returned to Humacyte or authorized designee, and the original will be maintained in the Investigator's Files. The HAV Accountability Log will be used to account for all IMP received, dispensed, and returned and must be maintained by the site until the conclusion of the study. Following accountability of the HAV by Humacyte or their authorized designee, all unused HAVs will be returned to Humacyte.

7. OTHER TREATMENTS AND MEDICATIONS

7.1. Prior and Concomitant Medications

7.1.1. Medication Data to be Collected

For each prior or concomitant reportable medication taken by or administered to the patient, the following information must be collected:

- Medication generic name / components of combination products
- Dose
- Route of administration
- Frequency of administration
- Date started
- Date stopped
- Indication for use

7.1.2. Prior Medications

Prior medications are defined as all prescription and over the counter (OTC) medications taken within 7 days before the initial trauma (i.e., prior to Day 1) whether use of that medication was continuing during the study or not.

- All prior and concomitant medications (including immediately pre-surgery and post-surgery medications) must be listed in the patient's medical record.
- Drugs used during anesthesia should be recorded in the anesthesia records but should not be transcribed into the eCRF.
- Drugs used to manage standard issues that may arise during any surgical procedure should not be transcribed into the eCRF, unless they are specifically listed in the reportable medications listed in Table 6.

7.1.3. Concomitant Medications

Concomitant medications are defined as medications taken after the initial trauma, even if this is prior to HAV implantation. Only the concomitant medications described in Table 6 must be recorded on the eCRF.

After HAV implantation, patients should be questioned at each study visit concerning any new medications or changes in current medications. Particular attention should be made to identify the use of antithrombotic or antiplatelet agents.

Table 6 Concomitant Medications That Must Be Recorded in the Patient eCRF

Drug Class	Includes (but is not limited to) These Drugs
Any medications used to manage an adverse event that was considered to be related to the HAV	Any medication satisfying the criterion
Any medications used to manage an adverse event that was considered to be a Serious Adverse Event (SAE)	Any medication satisfying the criterion
Any medications used to manage an adverse event that was considered to be an Adverse Event of Special Interest	Any medication satisfying the criterion
Systemic anti-infective medications (excludes topical, intraocular, local oral treatments such as troche/mouthwash)	Any antibiotic, antifungal, or antiviral medication
Anticoagulant, antiplatelet, or thrombolytic medications	Heparin, warfarin, aspirin, clopidogrel, prasugrel, epoprostenol, dabigatran, rivaroxaban, apixaban, edoxaban, betrixaban, alteplase, direct thrombin inhibitors, factor X_a inhibitors, or vitamin K antagonists
Non-steroidal anti-inflammatory drugs (NSAIDS)	Any medication satisfying the criterion
Systemic glucocorticoids	Any medication satisfying the criterion (excludes those administered by inhalation and those administered by intranasal, topical, or intraocular routes).
Immunomodulatory drugs	Any medication satisfying the criterion
Blood products	Packed red blood cells (RBCs), platelets, fresh frozen plasma, cryoprecipitate, albumin, immunoglobulin
Chemotherapy or Radiation used to treat malignancy	Any medication satisfying the criterion
Lipid lowering agents	Statins, niacin, gemfibrozil, fenofibrate

7.2. Essential, Precautionary and Prohibited Medications

7.2.1. Essential Medications

All patients should receive both antibiotic and antithrombotic prophylaxis in conjunction with HAV implantation in accordance with local hospital guidelines.

Antibiotic prophylaxis: All patients must have at least 1 day of antibiotic prophylaxis initiated on the same day as surgery. Longer antibiotic prophylaxis is at the discretion of the investigator.

Antithrombotic prophylaxis:

- Intraoperative heparin: the doses of heparin to be used during surgery will be determined by the investigator.
- Further measures to prevent venous thromboembolism are at the discretion of the investigator and may include low molecular weight heparin (LMWH).
- If antiplatelet therapy was not ongoing at the time of surgery, it should be commenced as soon as medically appropriate post operatively.
    - Recommended antiplatelet therapy (aspirin 81-325 mg and/or clopidogrel 75 mg daily) is at the discretion of the investigator and should continue long term while the HAV is in place.
    - If the patient is unable to tolerate aspirin and/or clopidogrel the choice of antiplatelet regimen is at the investigator's discretion.

7.2.2. Restricted and Prohibited Medications

Vitamin K antagonists, antiplatelet agents other than aspirin and clopidogrel, direct thrombin inhibitors and factor X_a inhibitors (e.g., dabigatran, apixaban and rivaroxaban) should be avoided unless essential for treatment of a medical condition arising postoperatively. In that case consideration should be given to modification or cessation of antiplatelet therapy. Antiplatelet therapy should be restarted on cessation of these anticoagulant drugs.

8. STUDY PROCEDURES AND EVALUATIONS

8.1. Clinical Evaluations Through Month 12

- Medical History: pre-operatively, from patient / legal representative interview and medical records covering relevant past medical history.
- Smoking History
- Medication History: prescription and OTC medication from Day -7 onwards (see Section 6.7). Particular attention should be paid to the identification of OTC medications containing aspirin.
- Physical Exam: full exam (as far as possible) at pre-operative screening, at the Month 12 Visit, or at the final study visit for early termination (ET). Clinical examination of the operative site and HAV at all post-operative visits; exam of distal vascular bed (extremity injury only); physical exam for lymphadenopathy; additional clinical exam as needed to evaluate adverse events.
- Vital Signs: heart rate, blood pressure and temperature should be obtained at Day 5 after implantation.

- Clinical Laboratories: Blood samples for hematology, clinical chemistry at pre-operative screening and Day 5. Samples for PRA titers should be obtained at pre-operative screening (or within 12 hours of HAV implantation), and at the Day 30, Month 6, and Month 12 Visits.
- Pre-operative Imaging (ultrasound or angiography) is at the discretion of the investigator based on the clinical condition of the patient. It may be omitted if not deemed necessary in order to proceed to vascular surgical repair.
- Adverse Events – post-operatively on Day 1 and at all post-operative visits, the patient will be asked a general question about his/her health and for any HAV problems since the previous visit.
- Post-operative Imaging: Intraoperative HAV bypass or interposition repair exam to assess anastomotic anatomy, patency and runoff. This may include physical exam, Doppler exam, angiography (conventional or intra-operative CT angiography) or ultrasound at the investigator's discretion.
- Duplex ultrasound: clinical assessment must be performed at all postoperative visits from postoperative Day 30 through the Month 12 Visit; HAV patency, mid HAV diameter, and HAV stenosis must be documented. The full length of the HAV should be imaged at each assessment to monitor for aneurysm development.
- Documentation of HAV interventions, surgical procedures, and any complications that occur from the immediate postoperative interval through the Month 12 Visit.

8.2. Clinical Evaluations in Long Term Follow Up (Post Month 12 to Month 36)

The status of the patient and HAV will be ascertained every 3 months after the Month 12 Visit until the Month 36 Visit. Patient status may be ascertained via telephone contact with the patient and/or his physician. If a suspected SAE related to HAV is discovered an unscheduled visit should be conducted to investigate.

Visits at Month 24 and Month 36 are to be conducted in person with a physical exam of the HAV site and duplex ultrasound imaging of the HAV.

Fatal AEs (Deaths) will be reported through the Month 36 Visit.

Only related SAEs and all AESI will be reported.

8.3. Laboratory Evaluations

8.3.1. Clinical and Research Laboratory Evaluations and Specimen Collection

The following parameters will be measured wherever possible at pre-operative screening and all should be measured at Day 5:

- Hematology: hemoglobin, hematocrit, RBC, white blood cells (WBC) with differential, platelet count

- Clinical chemistry: sodium, potassium, calcium, blood urea nitrogen, creatinine, albumin, total bilirubin, glucose (non-fasting)
- PRA titers will be measured at pre-operative screening (or within 12 hours of HAV implantation), at the Day 30 Visit, the Month 6 Visit, and the Month 12 Visit.

All laboratory tests (except assay of PRA) will be conducted at certified hospital laboratories. Routine monitoring, maintenance or calibration of laboratory equipment is required per local site procedures. Samples for PRA analysis will be shipped to LabConnect for storage until they are sent for analysis at a central laboratory. Details concerning sample collection and processing can be found in the Study Manual.

8.4. Imaging Evaluations

8.4.1. CT Angiography and Conventional Angiography

CT angiography or conventional angiography will be conducted as pre-operative screening when feasible. Pre-operative imaging is at the discretion of the investigator, based on the clinical condition of the patient. It may be omitted if not deemed necessary in order to proceed to vascular surgical repair.

8.4.2. Duplex Ultrasound

Duplex ultrasound examinations will be performed at Day 30, 3, 6, 9,12, 24, and 36 months and follow standard bypass graft imaging protocols, including B-mode, power Doppler and color duplex ultrasound imaging of the HAV with velocity spectral waveform analysis. The purpose of this duplex ultrasound surveillance is to detect HAV stenosis and aneurysm development. An alternative imaging method (e.g., CTA, MRI, etc.) may be substituted for duplex ultrasound at the discretion of the investigator if it is medically appropriate and in the best interest of the patient.

Determination of intraoperative HAV patency on Day 1 is required by physical examination, Doppler exam, angiography (conventional or intra-operative CT angiography), or ultrasound at the discretion of the investigator.

8.5. Study Schedule

8.5.1. Preoperative Screening (Day 1)

Potential study participants who are being considered for surgical repair of vascular injury appropriate for inclusion into the study will be informed about the study and invited to participate. After explanation of the potential risks and benefits of the HAV and of the study procedures, written informed consent will be obtained. No study specific procedures may be performed prior to patient consent. If the patient is unable to give informed consent, then this may be sought from the patient's legal representative (usually a close relative). Standard of care procedures such as laboratory evaluations conducted prior to screening may be used rather than repeating the test.

The following assessments will be performed, as far as possible, prior to surgery (Day 1):

- Informed consent
- Medical history
- Prior and concomitant medication
- Full physical examination
- Evaluation of inclusion/exclusion criteria
- Reasons for not using an autologous venous conduit
- Laboratory testing (or standard pre-op lab profile for the institution)
    - Hematology: full blood count and differential
    - Clinical chemistry; sodium, potassium, calcium, blood urea nitrogen, creatinine, albumin, total bilirubin, glucose (non-fasting)
    - PRA titer (if not obtained pre-operatively may be obtained within 12 hr. of surgery)

Ultrasound or CT angiography (CTA) (pre-op imaging is at the discretion of the investigator based on the clinical condition of the patient. It may be omitted if not deemed necessary in order to proceed to vascular surgical repair

8.5.2. Enrollment – Day 1 (HAV Implantation)

The HAV will be implanted as an interposition replacement or bypass in the required location using standard vascular surgical techniques. Details of the surgical anatomy and any complications will be documented. Operative procedures note and surgical diagram will be uploaded into the EDC for medical monitor review. Determination of intraoperative HAV patency is required by physical examination, Doppler exam, angiography (conventional or intra-operative CT angiography), or ultrasound at the discretion of the investigator.

8.5.3. Follow-up Visits Day 5 through Month 12

Day 5 (or prior to hospital discharge if earlier)

The following assessments should be completed:

- Concomitant Medication
- Physical exam including the surgical site
- Vital signs (heart rate, blood pressure and temperature)
- Documentation of any HAV interventions
- Adverse events
- Laboratory assessments (clinical chemistry, hematology)

Day 30 (± 5 days)

The following assessments should be completed:

- Concomitant medication

- Physical exam including the surgical site
- Duplex ultrasound of the HAV
- Documentation of any HAV interventions
- Adverse events
- PRA sample collection

Months 3, 6 and 9 (± 14 days)

The following assessments should be completed:

- Concomitant Medication
- Physical exam including the surgical site
- Duplex ultrasound of the HAV
- Documentation of any HAV interventions
- Adverse events
- PRA sample collection (Month 6 only)

Month 12 (± 14 days) and Early Termination

The following assessments should be completed:

- Concomitant Medication
- Full physical exam including the surgical site
- Duplex ultrasound of the HAV
- Documentation of any HAV interventions
- Adverse events
- PRA (only at early termination if prior to the Month 6 PRA sample collection)
- CT angiography

8.5.4. Long Term Follow-up Post Month 12 through Month 36 (± 30 days)

The status of the patient and HAV will be ascertained every 3 months after the Month 12 Visit up to the Month 36 Visit. The following assessments should be completed:

Quarterly questionnaire covering the status of the patient via a telephone contact with the patient and/or physician. If a suspected SAE related to HAV is discovered an unscheduled visit should be conducted to investigate.

- Documentation of any HAV interventions
- Adverse events (all AESI and related SAEs to be reported)
- Physical exam including surgical site at Month 24 and Month 36

- Duplex ultrasound at Month 24 and Month 36

8.5.5. Early Termination Visit

The patient may withdraw from the study at any time at his/her own discretion. The treating physician may also withdraw the patient for safety reasons. If withdrawal occurs before 12 months, the patient will be asked to complete an early termination visit at which all assessments normally performed at the Month 12 Visit will be completed. PRA will be collected at ET visit if the visit occurs before Month 6 collection of the sample. If withdrawal occurs after the Month 12 Visit and prior to the Month 36 Visit, the patient will be asked to complete an early termination visit at which all assessments normally conducted during the long-term follow-up visits will be completed.

The reasons for early termination should be recorded in the eCRF.

With the exception of patients who withdraw, all patients will be followed for 12 months from HAV implantation (or until HAV removal or death if earlier). The patient should be withdrawn from the study if the HAV is completely removed or the HAV becomes permanently occluded (loss of secondary patency) after Month 12.

8.5.6. Unscheduled Visits

If necessary to evaluate adverse events or HAV complications additional visits may be scheduled at the discretion of the investigator. At a minimum HAV status on clinical examination and Duplex ultrasound and adverse events will be recorded.

If, at any of the scheduled visits, duplex ultrasound surveillance suggests the development of a ≥ 50% stenosis within the HAV but immediate intervention is not required closer follow up should be considered. Intervention to manage any such stenosis is at the discretion of the investigator taking into account the degree and rate of progression of the stenosis.

8.6. Medical Care during the Study and upon Study Termination

Optimal medical therapy should be continued during the study. This should include antiplatelet therapy (see Section 7.2.1)

After the final study visit at Month 36 patients will not receive any further study-specific treatment. They will be treated by their medical doctor in a way that is appropriate for them.

8.7. Histological Examination of Resected HAV Material

If all or part of the HAV is resected it should, wherever possible, be retained for future histological examination. Instructions for preservation, storage and shipping of this material will be provided separately in a procedures manual. If a patient dies with an HAV in situ and it is feasible to obtain a fresh postmortem sample of the bypass this should be attempted in accordance with local regulations.

9. SAFETY ASSESSMENTS AND ADVERSE EVENTS

Safety of the HAV will be assessed in terms of:

- Aneurysm formation
- Pseudoaneurysm formation
- Anastomotic bleeding or spontaneous rupture
- HAV infection
- Need for HAV removal
- Inflammation at the implantation site
- Adverse events
- Increase from baseline in PRA titers

9.1. Adverse Event Definition

An AE is any untoward medical occurrence in a patient administered an Investigational Medicinal Product (IMP) and which does not necessarily have a causal relationship with the IMP. An AE can, therefore, be any unfavorable and unintended sign, symptom, or disease temporally associated with the use of the HAV, whether or not the AE is considered to be related to the HAV. Any worsening of the patient's disease under study or other medical conditions will also be considered to be an AE, unless it is within the normal range of disease fluctuation for that patient.

9.2. Serious Adverse Event Definition

An AE is considered to be a Serious Adverse Event (SAE) if it results in any of the following outcomes:

- Death
- Is life-threatening
    - An adverse event is considered "life-threatening" if, in the view of either the investigator or sponsor, its occurrence places the subject or patient at immediate risk of death. It does not include an adverse event or suspected adverse reaction that, had it occurred in a more severe form, might have caused death.
- Inpatient hospitalization or prolongation of existing hospitalization.
    - Note that hospitalization for the surgery to implant the HAV is not an SAE. However, prolongation of the initial hospitalization due to an AE will be considered an SAE.
- Persistent or significant incapacity or substantial disruption of the ability to conduct normal life functions.
- Congenital anomaly or birth defect
- Important medical events that may not result in death, be life-threatening, or require

hospitalization may be considered serious when, based upon appropriate medical judgment, they may jeopardize the subject or patient and may require medical or surgical intervention to prevent one of the outcomes listed in this definition.

9.3. Suspected Unexpected Serious Adverse Reaction

A suspected unexpected serious adverse reaction (SUSAR) is any adverse drug reaction that is serious (as defined in Section 9.2), unexpected (is not listed in the IB or is not listed at the specificity or severity that has been observed) and suspected (meaning there is a reasonable possibility that the HAV caused the adverse event).

9.4. Adverse Events of Special Interest

Adverse Events of Special Interest (AESI) are:

- HAV occlusion (thrombosis)
- HAV spontaneous rupture
- HAV infection
- HAV abandonment
- HAV aneurysm
- HAV pseudoaneurysm
- HAV Excision (partial or complete)

Note that AEs associated with iatrogenic injuries are not considered to be AESI; instead, they should be reported as AEs.

9.5. Reporting of Adverse Events

At each evaluation, the investigator will determine whether any AEs have occurred. The patient will be questioned in a general way and no specific symptoms will be suggested. If any AEs have occurred, they should be documented in the patient's medical chart and recorded on the AE pages of the eCRF. If known, the diagnosis should be recorded in preference to the listing of individual signs and symptoms. All SAEs should be reported to the Safety CRO within 24 hours from the time the investigator or study personnel first become aware of the event.

AE reporting begins from time of anesthesia induction for implantation of the HAV and ends at the conclusion of the Month 12 Visit or the Early Termination Visit, unless an unresolved AE is still being followed.

During the long term follow up period after the Month 12 Visit through the Month 36 Visit, only the following will be reported by the investigator:

- All SAEs considered related to the HAV
- All Events of Special Interest (Section 9.4)
- All Fatal AEs (deaths)

9.5.1. Causal Relationship to the HAV and the Index Surgical Procedure

The criteria for determining the causal relationship of an AE to the HAV itself, as well as a separate assessment of causal relationship of an AE to the index surgical procedure are required (Table 7). Note that causal relationship to procedure only refers to the index surgical procedure in which the HAV was initially implanted.

The sponsor will make the final determination of causality for the purposes of reporting to the regulatory authorities and to the Principal Investigators.

Table 7 Criteria for Determining Causal Relationship to the HAV or the Index Procedure

Relatedness	Criteria for Relatedness Category
Definitely Related	There is clear evidence to suggest a causal relationship, and other possible contributing factors can be ruled out. The clinical event, including an abnormal laboratory test result, occurs in a plausible time relationship to surgical placement of the HAV and cannot be explained by concurrent disease or other devices, drugs, or chemicals.
Possibly Related	There is some evidence to suggest a causal relationship (e.g., the event occurred within a reasonable time after the surgical placement of the HAV). However, the influence of other factors may have contributed to the event (e.g., the patient's clinical condition, other concomitant medications). Although an adverse event may rate only as "possible" soon after discovery, it can be flagged as requiring more information and later be upgraded to certain as appropriate.
Unlikely Related	A clinical event, including an abnormal laboratory test result, whose temporal relationship makes a causal relationship improbable (e.g., the event did not occur within a reasonable time after surgical placement of the HAV) and in which other drugs or chemicals or underlying disease provide plausible explanations (e.g., the patient's clinical condition, other concomitant treatments).
Not Related	A clinical event, including an abnormal laboratory test result, which occurs when the HAV was not implanted; or, another cause is obvious and in which there is sufficient information that the etiology of the event is not related to the HAV.

9.5.2. Criteria for Defining the Severity of an Adverse Event

The severity of adverse events, including abnormal clinical laboratory values, will be assessed according to the criteria below and entered in the eCRF:

1-Mild: Events require minimal or no treatment and do not interfere with the patient's daily activities.

2-Moderate: Events result in a low level of inconvenience or concern with the therapeutic measures. Moderate events may cause some interference with functioning.

3-Severe: Events interrupt a patient's usual daily activity and may require systemic drug therapy or other treatment. Severe events are usually incapacitating.

4-Life-threatening: Any adverse event that places the patient or participant, in the view of the investigator, at immediate risk of death from the reaction as it occurred, i.e., it does not include a reaction that, had it occurred in a more severe form, might have caused death.

5-Death: Death was related to the AE.

9.5.3. Reporting of Action Taken to Resolve AE

Five categories of actions taken to manage the AE are available for entry in the eCRF:

- None
- Lab tests / further evaluation
- Treatment required (specify if hospitalized)
- Patient withdrawn from study
- Other (specify)

9.5.4. Reporting the Outcome of the AE

Five categories of AE outcomes are available for entry in the eCRF:

- Recovered, with sequelae
- Recovered, without sequelae
- Ongoing
- Death
- Lost to follow-up

9.5.5. Reporting Serious Adverse Events

The urgency for reporting SAEs is 4-fold: (1) to facilitate discussion (and implementation, if necessary) by the sponsor and the investigator of appropriate follow-up measures, (2) to facilitate investigator reporting of unanticipated problems involving risk to human patients to the institutional review board (IRB), (3) to facilitate the sponsor's rapid dissemination of information regarding AEs to other investigators/sites in a multi-center study, and (4) to enable the sponsor to fulfill the reporting requirements to the appropriate regulatory authority.

- Any SAE that occurs from postoperative Day 1 through the Month 12 Visit (whether or not causally related to the HAV) must be reported by the investigator or designee to the Safety CRO within 24 hours of learning of its occurrence.

- Any SAE that occurs during Long-term Follow-up (after the Month 12 Visit through the Month 36 Visit) that is considered causally related to the HAV must be reported by the investigator or designee to the Safety CRO within 24 hours of learning of its occurrence.
- Any SAE resulting in patient death (regardless of relatedness attribution) that occurs during Long-term Follow-up (after the Month 12 Visit through the Month 36 Visit) must be reported by the investigator or designee to the Safety CRO within 24 hours of learning of its occurrence.

Information about an SAE will be collected and recorded on the SAE Report Form. The investigator must assess the relationship to the investigational product and any relevant procedure.

The investigators must complete the SAE Report Form in English, and send the completed, signed form by email to the Safety Monitor (see below) IMMEDIATELY (at latest within 24 hours) after becoming aware of the SAE.

Atlantic Research Group, Inc.
Drug Safety Department
Email: Safety@atlanticresearchgroup.com
Telephone: +1-888-619-3216

The investigator will be requested to supply as much detailed information as possible regarding the SAE that is available at the time of the initial contact. The investigator should also complete missing or requested information and submit follow-up reports until the SAE has resolved or, in the case of permanent impairment, until the SAE has stabilized.

Copies of relevant portions of medical records (e.g., admission and/or discharge summary, laboratory reports and autopsy report), may also be submitted with the SAE form to clarify the circumstances surrounding the SAE(s). The entire medical records should NOT be sent with the SAE form.

It is the responsibility of each Principal Investigator to promptly notify his/her IRB of all SAEs that are received by the Sponsor or designee and that occur at his/her institution in accordance with institutional practices.

The Safety CRO will inform the sponsor about all SAEs within 1 business day after receipt of the respective report from the investigator.

9.5.6. Reporting of Events of Special Interest

Events of Special Interest are defined in Section 9.4 and should be reported to the Safety CRO within 24 hours of learning of its occurrence. For each of these events detailed surgical notes (with illustrative diagram), including reason for and outcome of any intervention or abandonment, should be completed within 48 hours and uploaded to the clinical database.

Detailed information about the occurrence and treatment/intervention for these events will be collected throughout the study up to 3 years post HAV implant. This information will include the following:

- Summarized surgical notes, including a simplified anatomical diagram showing where angioplasties, stents, or revisions have been performed (using intervention worksheet provided)
- Need for antibiotics (in the case of infections)

9.5.7. Follow-Up of Adverse Events

If any AEs are present when a patient completes 1 year after implantation (i.e., the Month 12 Visit) or the Early Termination Visit (if earlier), or if a patient is withdrawn from the study, the patient will be re-evaluated within an appropriate period of time.

At the investigator's discretion, minor AEs can be re-evaluated via telephone and documented. If the AE has still not resolved, additional follow-up will be performed as appropriate. The investigator or his designee should make every effort to contact the patient until the AE has resolved or stabilized or the medical monitor and investigator agree that further follow-up is not necessary. This should be documented in the patient's medical records.

9.6. Reporting of Pregnancy

If a study participant becomes pregnant during study participation, basic information about the pregnancy will be recorded in the Pregnancy eCRF and the Pregnancy Outcome and Report Form, and submitted to the Safety CRO. If there are complications during the pregnancy, the complications are recorded as AEs. The participant will be asked to report the outcome of the pregnancy and the site should submit the information to the Safety CRO within 30 days after the outcome of the pregnancy. If there is a congenital anomaly in the infant, this will be recorded as a SAE in the data forms for the mother (i.e., the study participant).

Partner pregnancies do not need to be reported.

9.7. Data Monitoring Committee

A Data Monitoring Committee (DMC) will review safety on an ongoing basis and provide recommendations about stopping, continuing or otherwise modifying the study. The DMC consists of individuals who are not directly involved in the conduct of the study. A charter describes the roles and responsibilities of the DMC. Responsibilities of the DMC will include review of aggregate safety data from other studies in the HAV clinical development program.

The DMC will at a minimum meet every 6 months from the date of initial enrollment of the first patient. Additionally, the DMC will review the safety data of the torso injuries on the earlier of when the first torso patient reaches 3 months post-implantation or when the first 2 torso patients have both reached 30 days post-implantation.

10. STATISTICAL CONSIDERATIONS

This is a prospective, open label, nonrandomized multicenter Phase 2/3 study to evaluate the safety and efficacy of the HAV in patients undergoing vascular replacement or reconstruction. There is no formal hypothesis testing planned.

The primary analysis will be conducted when approximately 50 patients complete the 30-day follow-up. Specific details of data handling and analysis will be described in the SAP.

10.1. Analysis Sets

Three Sets of patients will be examined during analysis of study data:

- The **Extremity Set** will include patients who underwent arterial vascular repair with an HAV in an extremity. This population was previously referred to as the "Eligible" population in the Interim Analysis.
- The **All HAV Set** will include all patients who have received an HAV, regardless of the location of the vessel repaired and the type of injury. This population was previously referred to as the "Total" population or the "Safety" population in the Interim Analysis.
- The **Torso+Iatrogenic Set** will include all patients who received an HAV in a location in the Torso as well as any patient who received an HAV to repair an iatrogenic injury. This population was previously referred to as the "Non-Eligible" population in the Interim Analysis.

10.2. Safety Analyses

Safety analyses will be performed on all patients in the Extremity Set (as defined in Section 10.1).

10.2.1. Primary Safety Analyses

Primary Safety Analyses will include the following:

- Rate of HAV infection
- Frequency of Adverse Events of each severity grade.

10.2.2. Secondary Safety Analyses

Secondary safety analyses may focus on evaluating the mechanical stability of the HAV, based on freedom from aneurysmal degeneration, anastomotic bleeding, spontaneous rupture, infection, or significant stenosis. Safety data evaluated to support these analyses may include:

- Adverse Events of Special Interest (AESI) and
- Occurrences of HAV infection.

To determine the durability of the HAV in arterial vascular repair, the rates of interventions needed to maintain or restore HAV patency will be assessed.

In addition, other aspects of HAV function may be assessed for safety, data permitting:

- Amputation rate for the implanted limb
- Patient mortality
- Rate of HAV removal

- Rate of postoperative surgical site infection

Adverse events will be coded using Medical Dictionary for Regulatory Activities (MedDRA) terms.

HAV complications will be listed in terms of incidence, severity, and (where appropriate) time to onset and duration. Additional details will be provided in the SAP.

10.3. Efficacy Analyses

Following discussions with the FDA on 30th March 2022 and in April 2023, the primary Set of patients for analysis of efficacy will be defined as the "Extremity Set", formerly known in the Interim Analysis as the "Eligible population", as defined in Section 10.1.

The sections below provide concise descriptions of the efficacy analyses and endpoints. Specific details will be provided in the SAP.

Primary patency is defined as functional access patency until any type of intervention; primary assisted patency is defined as an HAV still working without complete thrombosis, with or without interventional or surgical procedures to maintain patency; secondary patency is defined as functional HAV patency, with or without preceding successful interventional or surgical procedures to maintain or reestablish patency, until the HAV is abandoned.

10.3.1. Primary Efficacy Analyses

Response will be defined as loss of primary patency before day 30 or maintained primary patency for at least 30 days. The proportion of patients with patency at 30-day will be estimated along with 95% confidence interval. The SAP will describe the estimand approach, including the main estimator and sensitivity analyses.

10.3.2. Secondary Efficacy Analyses

The key secondary efficacy endpoints include secondary patency at 30 days, HAV infection-free rate, and limb salvage rate. For each of these endpoints, the proportion of patients will be estimated along with 95% confidence interval.

Other secondary endpoints include long-term secondary patency, long-term HAV infection-free rate, intervention rates, patient survival and HAV remodeling. Details will be described in the SAP.

10.3.3. Other Analyses

Details of other analysis will be described in the SAP. .

10.4. Sample Size Rationale

Up to 100 patients will be enrolled in the study. The primary efficacy analysis will be performed after approximately 50 patients in the Extremity Set complete the 30-day follow-up. Sample size rationale for primary efficacy analysis will be provided in the SAP.

10.5. Interim Analyses

An interim analysis (IA) was performed using a data cutoff of 22April2022. In this IA, a total of 52 patients were included, of which 34 were in the Extremity Set.

The primary efficacy analysis will be performed after approximately 50 patients in the Extremity Set complete the 30-day follow-up.

11. STUDY MANAGEMENT AND DATA COLLECTION

11.1. Ethical Conduct of the Trial

This study will be conducted according to the protocol; 21 CFR Parts 11, 50, 54, 56, and 312; the World Medical Association Declaration of Helsinki and Good Clinical Practice (GCP). Each Investigator will conduct the trial according to applicable local or regional regulatory requirements.

11.2. Institutional Review Board

IRBs must be constituted according to the applicable state and federal requirements, including ICH GCP.

It is the responsibility of each investigator to submit the protocol, Investigator's Brochure, patient informed consent, patient recruitment materials (if applicable), and other documentation as required by the IRB to his/her IRB for review and approval. A copy of the written approval must be provided to the contract research organization (CRO). The documentation should clearly mention the approval/favorable opinion of the protocol, the patient informed consent form, and patient recruitment materials (if applicable), including respective version dates. The written approval and a list of members, their titles or occupations, and their institutional affiliations may be obtained from the IRBs if available, and provided to the CRO prior to the release of clinical study supplies to the investigational site and commencement of the study. If any member of the IRB has direct participation in this trial, written notification regarding his or her abstinence from voting must also be obtained.

Each investigator must adhere to all requirements stipulated by his/her respective IRB. This includes notification to the IRB regarding protocol amendments, updates to the patient informed consent, recruitment materials intended for viewing by patients, investigational new drug safety reports, SAEs and unexpected AEs, reports and updates regarding the ongoing review of the trial at intervals specified by the respective IRB, and submission of final study reports and summaries to the IRB.

11.3. Patient Informed Consent

Prior to any study procedures being performed, patients and persons conducting the consent discussion will be required to sign and date the IRB-approved informed consent, and each patient will be given a copy. In addition, this information should be recorded in the patient's medical record (i.e., source document). If the patient is unable to give informed consent, then this may be sought from the patient's legal representative, usually a close relative.

The written consent document will embody the elements of informed consent as described in the World Medical Association Declaration of Helsinki, 21 CFR Part 50.25, ICH E6 guideline (GCP), and in accordance with any local regulations. The investigator is responsible for the preparation, content, and IRB approval of the informed consent document. The consent form must be approved by the site's IRB and be acceptable to Humacyte.

The consent form must be written in a language fully comprehensible to the prospective patient. The investigator or designee shall give the patient adequate opportunity to read it before it is signed and dated. Information should be given in both oral and written form whenever possible and in the manner deemed appropriate by the IRB. Patients must be given ample opportunity to inquire about details of the study.

Pediatric enrollment: Adolescents who have reached Tanner Stage V Sexual Maturity Rating may be enrolled if the vessel to be repaired is size-appropriate (US sites only); this requires obtaining informed consent from the minors legally authorized representative (parental consent or surrogate consent).

11.4. Amendments to the Protocol

An amendment must be agreed to in writing by Humacyte and submitted to the FDA (or other Competent Authorities, if applicable) and approved by the IRBs before the amendment can be implemented. Written approval of a protocol amendment is not required prior to implementation of changes to the protocol which eliminate an immediate hazard to the study patient; however, approval must be obtained as soon as possible thereafter. Any agreed amendments must also be signed by the investigator.

11.5. Study Initiation

The investigator must not enroll any patients prior to attendance at the Investigator Meeting or the completion of a formal site initiation visit conducted by the CRO. These meetings will include a detailed review of the study protocol and eCRF pages. The investigator will not be supplied with IMP until all necessary pre-study requirements have been completed and essential signed documents provided to the CRO.

11.6. Study Monitoring

It is the responsibility of the investigator to ensure that the study is conducted in accordance with the protocol, GCP, applicable regulatory requirements, and the currently approved Declaration of Helsinki, and that valid data are entered into the eCRF.

To achieve this objective, the monitor’s duties are to ensure the maintenance of complete, legible, well-organized, and easily retrievable data. The monitor will review the protocol with the investigator. In addition, the monitor will explain the investigator’s reporting responsibilities and all applicable regulations concerning the clinical evaluation of the IMP.

The investigator will permit representatives of Humacyte and the CRO to monitor the study as frequently as Humacyte or the CRO deem necessary to determine that data recording and protocol adherence are satisfactory. The eCRF data and related source documents will be reviewed in detail by the monitor at each visit, in accordance with relevant SOPs and ICH GCP regulations. This includes results of tests performed as a requirement for participation in this study and any other medical records required to confirm information contained in the eCRF such as past medical history and secondary diagnoses. The investigator and his/her staff will be expected to cooperate with the monitor and provide any missing information whenever possible.

All monitoring activities will be reported and archived. In addition, monitoring visits will be documented at the investigational site by signature and date on the study-specific monitoring log.

11.7. Case Report Form

An electronic CRF will be used for this study. The data will be entered into the eCRF in a timely manner on an ongoing basis.

The investigator is responsible for ensuring that data are properly recorded on each patient's eCRF and related documents. An investigator who has signed the protocol signature page should personally sign the eCRFs in accordance with the procedure described in the eCRF completion guidelines to ensure that the observations and findings are correct and complete.

For data handled by the CRO, eCRF data and some or all of the study-related data will be managed and stored electronically in the CRO's database system. Validated data will subsequently be transferred to the sponsor.

11.8. Verification Procedures

It is the investigator's obligation to ensure documentation of all relevant data in the patient's medical record. The patient's medical record will be considered the source document. The eCRF should not be used as the source for study information.

The investigator will maintain a patient identification code list to enable unambiguous identification of the patients (patient names and corresponding patient numbers). The patient identification code list is an essential document and as such should be maintained according to the ICH GCP guidelines.

11.9. Retention of Records

All documentation pertaining to the study will be kept by Humacyte or their designee in accordance with ICH guidelines, US FDA regulations, or applicable country regulations).

The investigator will maintain a study file, which should be used to file the Investigator's Brochure, protocol, and IMP records; correspondence with the IRB and Humacyte; and other study-related documents.

The investigator agrees to keep records and those documents that include (but are not limited to) the identification of all participating patients, medical records, study-specific source documents, source worksheets, all original signed and dated informed consent forms, query responses, and detailed records of drug disposition to enable evaluations or audits from regulatory authorities and Humacyte or its designees.

The investigator shall retain records required to be maintained under this part for a period of 2 years following the date a marketing application is approved for the IMP for the indication for which it is being investigated; or, if no application is to be filed or if the application is not approved for such indication, until 5 years after the investigation is discontinued. However, these documents should be retained for a longer period if required by the applicable regulatory requirement(s) or if needed by the sponsor. In addition, the investigator must make provision for

the patient's medical records to be kept for the same period of time. No data should be destroyed without the agreement of Humacyte. Humacyte will inform the investigator in writing when the trial-related records are no longer needed. Patient medical records and other original data will be archived in accordance with the archiving regulations or facilities of the study site.

11.10. Protocol Deviations

A protocol deviation is any noncompliance with the protocol or GCP requirements. The noncompliance may be either on the part of the participant, the investigator, or the study site staff. As a result of deviations, corrective actions are to be developed by the site and implemented promptly. Although in principle, protocol deviations are not permitted, under emergency circumstances, deviations may proceed without prior approval of the sponsor and the IRB to protect the rights, safety, and well-being of human patients.

All protocol deviations will be documented and reported by the CRO during the course of the study in the Monitoring Reports. All deviations will be reported to the sponsor who will agree on the necessary actions to be taken.

If required per their guidelines, reports about protocol deviations must be reported to the local IRB.

11.11. Insurance and Indemnity

Insurance coverage for damages emerging from the study will be provided according to applicable legal requirements. During the informed consent procedure, the investigator must inform the patient accordingly.

11.12. Audits

It is the responsibility of CRO and Humacyte to perform auditing (if applicable) as part of implementing quality assurance. The purpose of an audit, which is independent of and separate from routine monitoring or quality control functions, is to evaluate trial conduct and compliance with the protocol, SOPs, GCPs, and the applicable regulatory requirements. The auditor and regulatory authorities will require authority from the investigator to have direct access to the patient's medical records.

12. REPORTING

Following completion of follow-up of all patients to the 12-month endpoint, the results will be evaluated by Humacyte or a designee for clinically meaningful findings. A clinical study report will be generated, including a summary of all available data, statistical measures, tabulated results, graphical results and interpretations. This report will be submitted to regulatory authorities in a timely manner. An addendum to the report will be generated to include data up to 36 months follow-up. This addendum will be submitted to regulatory authorities in a timely manner.

13. QUALITY CONTROL AND QUALITY ASSURANCE

Following written standard operating procedures, the monitors will verify that the clinical trial is conducted and data are generated, documented (recorded), and reported in compliance with the protocol, GCP, and the applicable regulatory requirements. Reports of monitoring activities will be submitted to Humacyte in a timely manner.

The investigational site will provide direct access to all trial related areas, source data/documents, and reports for the purpose of monitoring and auditing by the sponsor, and inspection by local and regulatory authorities.

Quality control procedures will be implemented for data entry and the generation of data quality control checks and will be run on the database. Any missing data or data anomalies will be communicated to the site(s) for clarification/resolution.

14. RESPONSIBILITIES OF THE PRINCIPAL INVESTIGATOR

The role of the principal investigator is to implement and manage the day-to-day conduct of the clinical investigation as well as to ensure data integrity and the rights, safety, and well-being of the patients involved in the clinical investigation.

14.1. Informed Consent

The principal investigator shall ensure that the process for obtaining informed consent

- Includes all aspects of the clinical investigation that are relevant to the patient's decision to participate throughout the clinical investigation,
- Avoids any coercion or undue improper influence on, or inducement of, the patient to participate,
- Does not waive or appear to waive the patient's legal rights,
- Uses native non-technical language that is understandable to the patient,
- Provides ample time for the patient to read and understand the informed consent form and to consider participation in the clinical investigation, and
- Provides the patient with a copy of the signed and dated informed consent form and any other written information.

The principal investigator shall ensure and document appropriate training if an authorized designee is appointed to conduct the informed consent process.

14.2. Compliance with the Protocol

The principal investigator shall:

- Indicate his/her acceptance of the protocol in writing.
- Conduct the clinical investigation in compliance with the protocol.

- Create and maintain source documents throughout the clinical investigation and make them available as requested during monitoring visits or audits.
- Ensure that the IMP is used solely by authorized users, and in accordance with the protocol and instructions for use.
- Propose to the sponsor any appropriate modification(s) of the protocol.
- Refrain from implementing any modifications to the protocol without agreement from the sponsor, IRB, and, if required, regulatory authorities.
- Document and explain any deviation from the approved protocol that occurred during the course of the clinical investigation.
- Ensure that an adequate investigation site team and facilities exist and are maintained and documented during the clinical investigation.
- Ensure that maintenance and calibration of the equipment relevant for the assessment of the clinical investigation is appropriately performed and documented, where applicable.
- Ensure the accuracy, completeness, legibility, and timeliness of the data reported to the sponsor in the eCRFs and in all required reports.
- Maintain the clinical trial material accountability records.
- Allow and support the sponsor to perform monitoring and auditing activities.
- Be accessible to the monitor and respond to questions during monitoring visits.
- Allow and support regulatory authorities and the IRB when performing auditing activities.
- Ensure that all clinical-investigation-related records are retained as specified in this protocol.

14.3. Medical Care of Patients

The principal investigator shall:

- Provide adequate medical care to a patient during and after a patient's participation in a clinical investigation in the case of AEs.
- Inform the patient of the nature and possible cause of any adverse events experienced.
- Inform the patient of any new significant findings occurring during the clinical investigation, including the need for additional medical care that may be required.
- Provide the patient with well-defined procedures for possible emergency situations related to the clinical investigation, and make the necessary arrangements for emergency treatment.
- Ensure that clinical records are clearly marked to indicate that the patient is enrolled in a particular clinical investigation.

- Inform, with the patient's approval or when required by national regulations, the patient's personal physician about the patient's participation in the clinical investigation.
- Make all reasonable efforts to ascertain the reason(s) for a patient's premature withdrawal from the clinical investigation while fully respecting the patient's rights.

14.4. Safety Reporting

The principal investigator shall:

- Record every adverse event together with an assessment, in accordance with Section 9 of this protocol,
- Report to the sponsor, without unjustified delay, all serious adverse events and events of special interest as specified in Section 9 of this protocol, and
- Supply the sponsor, upon sponsor's request, with any additional information related to the safety reporting of a particular event.

15. SUSPENSION OR PREMATURE TERMINATION OF THE CLINICAL INVESTIGATION

The sponsor may suspend or prematurely terminate either a clinical investigation in an individual investigation site or the entire clinical investigation for significant and documented reasons.

A principal investigator, IRB, or regulatory authority may suspend or prematurely terminate participation in a clinical investigation at the investigation sites for which they are responsible.

If suspicion of an unacceptable risk to patients arises during the clinical investigation, or when so instructed by the IRB or regulatory authorities, the sponsor shall suspend the clinical investigation while the risk is assessed. The sponsor shall terminate the clinical investigation if an unacceptable risk is confirmed.

The sponsor shall consider terminating or suspending the participation of a particular investigation site or investigator in the clinical investigation if monitoring or auditing identifies serious or repeated deviations on the part of an investigator.

If suspension or premature termination occurs, the terminating party shall justify its decision in writing and promptly inform the other parties with whom they are in direct communication.

If, for any reason, the sponsor suspends or prematurely terminates the investigation at an individual investigation site, the sponsor shall inform the responsible regulatory authority if required and ensure that the IRB is notified. If the suspension or premature termination was in the interest of safety, the sponsor shall inform all other principal investigators.

If suspension or premature termination occurs,

1. The sponsor shall remain responsible for providing resources to fulfill the obligations from the protocol and existing agreements for following up the patients enrolled in the clinical investigation, and

2. The principal investigator or authorized designee shall promptly inform the enrolled patients at his/her investigation site, if appropriate.

In the event that the study is discontinued, the reasons for discontinuation will be explained to the investigators and may be disclosed to the study participants. Humacyte will provide all information needed by the investigator to ensure the safety and well-being of the study participants.

16. PUBLICATION POLICY

A Publication Committee comprising the Principal Investigator from each participating site and a representative of Humacyte will oversee all publication of data from this study. Prior to submitting for publication, presenting, using for instructional purposes, or otherwise disclosing the results of the study, the investigator agrees to allow the Publication Committee and Humacyte a period of at least 30 days (or, for abstracts, at least 5 calendar days) to review the proposed publication or disclosure prior to its submission for publication or other disclosure. Publications or disclosures of study results shall not include other confidential information belonging to Humacyte. If the proposed publication/disclosure risks Humacyte's ability to patent any invention related to the study, the publication or disclosure will be modified or delayed, at the investigator's option, a sufficient time to allow Humacyte to seek patent protection of the invention. For multicenter studies, the first publication or disclosure shall be a complete, joint multicenter publication or disclosure. This statement does not give Humacyte any editorial rights over the content of a publication or disclosure, other than to restrict the disclosure of Humacyte's confidential information. If a written contract for the conduct of the study is executed which includes publication provisions inconsistent with this statement, then that contract's publication provisions shall apply rather than this statement.

17. REFERENCES

Akingba, A.G., Rapp, B., Tsai, A., Jester, A., Fajardo, A., Motaganahali, R., Dalsing, M. and Murphy, M. (2012). "Surgical Management of Vascular Trauma From Dog Bites: The 10-year Experience of a Level I Trauma Center." Journal of Vascular Surgery **56**: 1190.

Akingba, A.G., Robinson, E.A., Jester, A.L., Rapp, B.M., Tsai, A., Motaganahalli, R.L., Dalsing, M.C. and Murphy, M.P. (2013). "Management of vascular trauma from dog bites." J Vasc Surg **58**(5): 1346-1352.

Andrikopoulos, V., Antoniou, I. and Panoussis, P. (1995). "Arterial injuries associated with lower-extremity fractures." Cardiovasc Surg **3**(1): 15-18.

Cordesman, A.H. and Greenough, N.B. (2010). "Shape, Clear, Hold, Build, and Transfer: The Metrics of the Afghan War. Center for Strategic & International Studies. www.csis.org/burke/reports, Center for Strategic & International Studies**:** 185.

Cordesman, A.H. and Lemieux, J. (2010). Improvised Explosive Devices (IED) Metrics for Afghanistan January 2004-May 2010. Center for Strategic & International Studies, https://csis-website-prod.s3.amazonaws.com/s3fs-public/legacy_files/files/publication/100112_AfghanLngMetrics.pdf.

Dahl, S.L., Kypson, A.P., Lawson, J.H., Blum, J.L., Strader, J.T., Li, Y., Manson, R.J., Tente, W.E., Dibernardo, L. and Hensley, M.T. (2011). "Readily available tissue-engineered vascular grafts." Sci Transl Med **3**.

Dubose, J.J., Savage, S.A., Fabian, T.C., Menaker, J., Scalea, T., Holcomb, J.B., Skarupa, D., Poulin , N., Chourliaras, K., Inaba, K. and Rasmussen, T.E. (2015). "The American Association for the Surgery of Trauma PROspective Observational Vascular Injury Treatment (PROOVIT) registry: multicenter data on modern vascular injury diagnosis, management, and outcomes." Journal of Trauma and Acute Care Surgery **78**(2): 215-223.

Faulconer, E.R., Branco, B.C., Loja, M.N., Grayson, K., Sampson, J., Fabian, T.C., Holcomb, J.B., Scalea, T., Skarupa, D., Inaba, K., Poulin, N., Rasmussen, T.E. and Dubose, J.J. (2018). "Use of open and endovascular surgical techniques to manage vascular injuries in the trauma setting: A review of the American Association for the Surgery of Trauma PROspective Observational Vascular Injury Trial registry." J Trauma Acute Care Surg **84**(3): 411-417.

Gutowski, P., Guziewicz, M., Ilzecki, M., Kazimierczak, A., Lawson, J.H., Prichard, H.L., Przywara, S., Samad, R., Tente, W., Turek, J., Witkiewicz, W., Zapotoczny, N., Zubilewicz, T. and Niklason, L.E. (2023). "Six-year outcomes of a phase II study of human-tissue engineered blood vessels for peripheral arterial bypass." JVS Vasc Sci **4**: 100092.

Helfet, D., Howey, T., Sanders, R. and Johansen, K. (1990). "Limb salvage versus amputation. Preliminary results of the mangled extremity severity score." Clin Orthop Relat Res **256**.

Holcomb, J. (2010). Devastating dismounted IED injuries in OEF. Increasing amputation and genital injury rates admitted to LRMC 2009–2010. Presentation to the defense health board.

Kelly, B.S., Heffelfinger, S.C., Whiting, J.F., Miller, M.A., Reaves, A., Armstrong, J., Narayana, A. and Roy-Chaudhury, P. (2002). "Aggressive venous neointimal hyperplasia in a pig model of arteriovenous graft stenosis." Kidney Int **62**(6): 2272-2280.

Kirkton, R.D., Prichard, H.L., Santiago-Maysonet, M., Niklason, L.E., Lawson, J.H. and Dahl, S.L.M. (2018). "Susceptibility of ePTFE vascular grafts and bioengineered human acellular vessels to infection." J Surg Res **221**: 143-151.

Kirkton, R.D., Santiago-Maysonet, M., Lawson, J.H., Tente, W.E., Dahl, S.L.M., Niklason, L.E. and Prichard, H.L. (2019). "Bioengineered human acellular vessels recellularize and evolve into living blood vessels after human implantation." Sci Transl Med **11**(485).

L'heureux, N., Dusserre, N., Konig, G., Victor, B., Keire, P., Wight, T.N., Chronos, N.A., Kyles, A.E., Gregory, C.R. and Hoyt, G. (2006). "Human tissue-engineered blood vessels for adult arterial revascularization." Nat Med **12**.

Lawson, J.H., Glickman, M.H., Ilzecki, M., Jakimowicz, T., Jaroszynski, A., Peden, E.K., Pilgrim, A.J., Prichard, H.L., Guziewicz, M., Przywara, S., Szmidt, J., Turek, J., Witkiewicz, W., Zapotoczny, N., Zubilewicz, T. and Niklason, L.E. (2016). "Bioengineered human acellular vessels for dialysis access in patients with end-stage renal disease: two phase 2 single-arm trials." Lancet **387**(10032): 2026-2034.

Prichard, H.L., Manson, R.J., Dibernardo, L., Niklason, L.E., Lawson, J.H. and Dahl, s.L.M. (2011). "An Early Study on the Mechanisms that Allow Tissue-Engineered Vascular Grafts to Resist Intimal Hyperplasia." J. of Cardiovasc. Trans. Res. **4**: 674-682.

Quint, C., Kondo, Y., Manson, R.J., Lawson, J.H., Dardik, A. and Niklason, L.E. (2011). "Decellularized tissue-engineered blood vessel as an arterial conduit." Proc Natl Acad Sci U S A **108**(22): 9214-9219.

Rasmussen, T., Stockinger, Z., Antevil, J., White, C., Fernandez, N., White, J. and White, P. (2018). "Wartime Vascular Injury." Military Medicine **183**(suppl_2): 101-104.

Roy-Chaudhury, P., Kelly, B.S., Miller, M.A., Reaves, A., Armstrong, J., Nanayakkara, N. and Heffelfinger, S.C. (2001). "Venous neointimal hyperplasia in polytetrafluoroethylene dialysis grafts." Kidney Int **59**(6): 2325-2334.

A Phase 2/3 Study for the Evaluation of Safety and Efficacy of Humacyte's Human Acellular Vessel for Vascular Replacement or Reconstruction in Patients with Life or Limb-threatening Vascular Trauma

Study No.: CLN-PRO-V005

Investigational Product: Human Acellular Vessel (HAV)

Study No.: CLN-PRO-V005

IND No.: 15263

Version Date: **24**May2023

Version: Protocol 4.0 Summary of Changes

Sponsor: Humacyte Global, Inc.
2525 East NC Highway 54
Durham, NC 27713

Sponsor Approval:

Shamik Parikh Digitally signed by Shamik Parikh
Date: 2023.05.24 17:16:01 -04'00'

Signature Date (dd/mmm/yyyy):

Printed Name: Shamik Parikh, MD
Title: Chief Medical Officer, Humacyte Global, Inc.
Telephone: 1 (919) 313-9633 ext 130
email: sparikh@humacyte.com

Confidentiality Statement

PROTOCOL REVISION HISTORY

Protocol Version	Active Region(s)	Date Finalized
Original Protocol	USA	29 December 2015
Version 2.0	USA	24 October 2016
Version 3.0	USA, Israel	18 July 2018
Version 3.1	For potential Poland sites – not implemented.	22 June 2021
Version 3.2	USA	18 August 2021
Version 3.3	USA (added pediatric enrollment)	12 August 2022
Version 4.0	USA (Master protocol) **Rationale for key revisions:** Incorporate modifications requested by the Agency during the March 2022 meeting (March 30, 2022 (Reference IND 16746 CRMTS #13939 Meeting Summary March 30, 2022 Type B OIND Teleconference) and during the March 2023 Type C pre-BLA meeting (reference): o Separate analyses for the group of patients with non-iatrogenic arterial trauma of the extremities, and the rest of the patients enrolled (patients with iatrogenic and torso injuries). o Inclusion of pediatric patients and clarification of category of eligible pediatric patients o Modification of statistical analysis section to include While-on-Treatment analysis methodology and define the estimands. - Serious Adverse Event definition updated to align with industry standards and FDA guidance	24 May 2023

SUMMARY OF CHANGES FOR AMENDMENT VERSION 4.0

SUMMARY OF CHANGES to PROTOCOL CLN-PRO-V005 to produce Version 4.0		
Affected Section	**Original Text**	**Amended Text (new or revised text is in bold)**
page headers (all)	Humacyte, Inc ... Confidential ..,.05 August 2022 Study No. CLN-PRO-V005 ...Version 3.3	Humacyte **Global,** Inc. ... **12May2023** Study No. CLN-PRO-V005 ... **Protocol Version 4.0**
throughout document	Section headers, text, and tables all in Arial font	Change Section headers, text, and tables to **Times New Roman** to conform to other Humacyte documents.
throughout document	In-text reference citations were in blue font but were not active hyperlinks.	**Recreated in-text reference citations using** the HAV EndNote Library **including hyperlink to the bibliography in Section 17**
throughout document		Reformatting for improved readability (example: replaced "Postimplantation" with "**after implantation**")
throughout document	Use of the regulatory technical terminology "IMP" for "Investigational Medicinal Product" is not needed in most sections, since this is typically used to distinguish between an investigational treatment and other approved/SOC treatment being given in the study - direct reference to the HASV is preferred, as it is the only treatment being offered.	Replace "IMP" with "**HAV**" where appropriate to improve clarity.
throughout document		Use of the term 'subject' is replaced with '**patient'** for consistency with Humacyte standards
page footers (all)	1 of XX	**CONFIDENTIAL** **Page 3** of XX
Title Page. Signature page, and Synopsis	A Phase 2 Study for the Evaluation of Safety and Efficacy of Humacyte's Human Acellular Vessel for Vascular Replacement or Reconstruction in Patients with Life or Limb-threatening Vascular Trauma	A **Phase 2/3** Study for the Evaluation of Safety and Efficacy of Humacyte's Human Acellular Vessel for Vascular Replacement or Reconstruction in Patients with Life or Limb-threatening Vascular Trauma
Title page	Reverted to standard Title page format and moved CRO name and address to Section 1. STUDY	Reverted to standard Title page format and moved CRO name and address to Section 1. STUDY PERSONNEL. Moved CMO signature from page 4 to the Title Page under "Sponsor Approval"

SUMMARY OF CHANGES to PROTOCOL CLN-PRO-V005 to produce Version 4.0		
Affected Section	**Original Text**	**Amended Text (new or revised text is in bold)**
	PERSONNEL, moved CMO signature to Title Page under "Sponsor Approval"	
Title page	No revision history	Added Table showing Revision History to follow the Title page
page 4	Not applicable	Added new section " SUMMARY OF CHANGES FOR AMENDMENT VERSION 4.0"
page XX	Protocol Approval	Deleted, replaced by Amendment 4 Summary of Changes; protocol approval signature (CMO) now moved to the title page.
Study Synopsis, pg. 13	Formerly called the Protocol Summary	Moved to follow the Summary of Changes (was previously on page 13) renamed to the more standard "**Protocol Synopsis**".
Protocol Synopsis	Planned Study Sites: Up to 25 sites in the United States (US) and Israel	Planned Study Sites: **Up to 35 sites** in the United States **(US), Israel, and Ukraine will be recruited to enroll patients**.
Protocol Synopsis and Section 5.1.1	Inclusion Criteria: 4. Aged 14 to 85 years old, inclusive.	4. **Adults aged 18 to 85 inclusive (all sites) and adolescents who have achieved Tanner Sexual Maturity Rating Stage V (US sites only).**
Protocol Synopsis and Section 5.1.2	Exclusion #5. "Known pregnant women"	Corrected grammar and syntax to more standard English construction: "**Women known to be pregnant**"
Protocol Synopsis	Accrual Period: 24 Months	Accrual Period: **enrollment will close on or before December 31, 2024.**
Protocol Synopsis Primary Objectives	Minor clarification: Efficacy: To determine the rate of primary patency of the HAV at 30 days	Replaced original text with **Table 3, Primary Objectives and Endpoints for Safety and Efficacy**
Protocol Synopsis Secondary Objectives	Safety • To determine mechanical stability of the HAV based on freedom from aneurysmal degeneration, anastomotic bleeding or spontaneous rupture, infection, or significant stenosis • To determine HAV durability in terms of freedom from HAV removal or replacement	Replaced original text with **Table 4 Secondary Objectives and Endpoints for Safety**

SUMMARY OF CHANGES to PROTOCOL CLN-PRO-V005 to produce Version 4.0		
Affected Section	**Original Text**	**Amended Text (new or revised text is in bold)**
Protocol Synopsis Secondary Objectives	Efficacy: • To determine the patency of the HAV (primary, primary assisted and secondary) • To determine the rates of interventions needed to maintain / restore patency in the HAV • To determine the rate of limb salvage (extremity injuries only) • To determine subject survival • To evaluate remodeling of HAV	Replaced original text with **Table 5, Secondary Objectives and Endpoints for Efficacy** Replaced the Objective and Endpoint of **"To determine the primary assisted patency of the HAV at 30 days/Primary assisted patency"** by **Objective and Endpoint "To determine the long-term limb salvage/Amputation" in Secondary Endpoints**
Protocol Synopsis	Study Design Prospective, multicenter, open-label, study	Study Design Prospective, multicenter, open-label, **non-randomized study**
Figure 1, text for Day 1 box	Day 1: Surgical placement of HAV (incl. documentation of surgical procedure and any complications); Confirmation of patency by intraoperative PE, Duplex ultrasound, or angiography (conventional or intra-op CT angiography); AEs; HAV interventions; concomitant medications (CMs).	Day 1: Surgical placement of HAV (incl. documentation of surgical procedure and any complications); Confirmation of patency by intraoperative PE, Duplex ultrasound, or angiography (conventional or intra-op CT angiography); AEs; HAV interventions; concomitant medications (CMs)**; and PRA if not preoperative.**
Table 1 Schedule of Visits and Assessments	Schedule of Assessments was previously provided in the Appendix "APPENDIX 1: HAV CLINICAL VISIT SCHEDULE"	Moved the Schedule of Assessments table up from the Appendix to now follow the Synopsis. Renamed the table to the more commonly-used "**Schedule of Visits and Assessments**"
Table 1 Schedule of Visits and Assessments	Abbreviations	Added "**PRA = panel reactive antibodies**"
Table 1 Schedule of Visits and Assessments	Footnotes - clarified the need to record and report fatal AES out to the Month 36 Visit (was formerly not specified).	i The status of the **patient** and HAV will be ascertained every 3 months after Month 12 until 36 months after HAV implantation. Status may be ascertained via telephone contact with the **patient** and/or his physician. **Fatal AEs (Deaths) will be followed and reported up to the Month 36 Visit.** Only related SAEs and all AESI will be reported after 12 months. If a suspected SAE related

SUMMARY OF CHANGES to PROTOCOL CLN-PRO-V005 to produce Version 4.0		
Affected Section	**Original Text**	**Amended Text (new or revised text is in bold)**
		to HAV is discovered, an unscheduled visit should be conducted to investigate.
Table 1 Schedule of Visits and Assessments, Footnote 5	X for PRA in preoperative screening column 5. Measured at preoperative screening when possible.	Superscript 5 added to X for PRA in preoperative screening column 5. Measured at preoperative screening when possible**; if PRA titer not obtained preoperatively, it must be completed within 12 hours after HAV implantation.**
1. STUDY PERSONNEL	The original Table listed personnel that do not need to be specified in the protocol	Renamed the embedded table to " Key Study Management Personnel" and deleted the ~~Sponsor Medical Representative, CRO Project Manager, Sponsor Regulatory representative, Sponsor Clinical Operations Representative,~~ **leaving the CRO Safety Representative and the Medical Monitor as Key personnel.**
2.4 Summary of Clinical Studies	The HAV clinical development program currently includes 7 clinical studies: 4 in patients with end-stage renal disease receiving hemodialysis (CLN PRO V001, CLN PRO V003, CLN PRO V006 and CLN PRO V007),.....and one phase 3 study (CLN-PRO-V007) are open for enrollment. As of 10 April 2018, 272 patients (244 hemodialysis access patients and 28 PAD patients) have received an HAV. The first implant for hemodialysis was performed in December 2012, and the first peripheral arterial bypass in October 2013. Overall, the total treatment exposure is approximately 329 patient years in the hemodialysis access population and 55 patient years in the PAD population.	Updated the content of the paragraph to reflect recent data: "**Human studies are ongoing in the United States (US), Europe, and Israel. In all, eight clinical studies were initiated to evaluate three clinical indications for use of the HAV. Seven studies completed their planned follow-up times with three continuing to follow patients long-term (CLN-PRO-V001, CLN-PRO-V002, CLN-PRO-V004, and CLN-PRO-V007). One clinical study NCT03005418 (EudraCT #2020-003383-12) or CLN-PRO-V005 (i.e., V005) that is evaluating HAV for repair of vascular trauma is continuing to enroll patients. As of 10 April 2022, a total of 374 patients with ESRD, 35 patients with PAD, and 51 patients with vascular trauma have had the HAV implanted. In addition, an HAV has been implanted in over 30 cases under an Expanded Access program. The first implant for hemodialysis was performed in December 2012, the first for peripheral arterial bypass in October 2013, and the first implant for trauma in September 2018. Across all clinical studies the total treatment exposure to HAV is approximately 1,005 patient-years, with approximately 830 patient-years, 119**

SUMMARY OF CHANGES to PROTOCOL CLN-PRO-V005 to produce Version 4.0		
Affected Section	**Original Text**	**Amended Text (new or revised text is in bold)**
		patient-years, and 56 patient-years in ESRD, PAD, and vascular trauma patients, respectively."
2.4.1 Experience in Patients Undergoing Peripheral Arterial Bypass	The second PAD study of similar design, CLN-PRO-V004, is being conducted in the US with enrollment ongoing.	Updated the sentence to reflect recent data: **"The second PAD study of similar design, CLN-PRO-V004, is being conducted in the US and long-term follow-up is ongoing."**
2.4.1 Experience in Patients Undergoing Peripheral Arterial Bypass	In subsection CLN-PRO-V002 Study Results (24 M, 72 M), added the long-term data.	Added paragraph to report recent data at 72 months: **"At six years (72 months), six patients completed follow-up, with five of six patients retaining HAV patency (i.e., primary patency = 4 and secondary patency = 1), which was confirmed by Doppler ultrasound. Three of the long-term patients died during follow-up; one from cancer at 71 months and two from other non-HAV-related causes at 54 months and approximately 67 months post-implant (Gutowski, 2022). Also, one patient voluntarily withdrew from the study at 61 months post-implant. During long-term follow-up (i.e., post 24 months), one patient had two thrombectomies performed. For all remaining patients, no interventions and no infections were documented. Overall, 7 deaths were recorded at the 72-month endpoint, for a mortality rate of 35% in this patient cohort (i.e., severe PAD with 7-year follow-up). No deaths were attributable to the HAV."**
2.6.3. Risk-Benefit Rationale	Added new text - 2nd paragraph, and deleted statement about DMC which is covered in Section 9.7 and the maximum enrollment which is covered in Section XX	The risks anticipated in this study are similar to those associated with currently marketed prosthetic grafts used for vascular repair. The potential advantages of the HAV compared to currently marketed grafts may lead to a lower complication rate and reduced need for surgical intervention and graft replacement as well as the potential for reduced secondary complications associated with autologous vessel harvest.

SUMMARY OF CHANGES to PROTOCOL CLN-PRO-V005 to produce Version 4.0		
Affected Section	**Original Text**	**Amended Text (new or revised text is in bold)**
		This is the first in man study in which the HAV will be used to repair vascular injuries within the torso (thorax, thoracic outlet, abdomen, and retroperitoneum) and so risks related to those locations have not been specifically characterized. However, these risks are not expected to be significantly different than those experienced or reported when used in the upper or lower extremity for AV access or arterial reconstruction. ~~The DMC will review safety data of the torso injuries on the earlier of when the first torso subject reaches 3 months post-implantation or when the first 2 torso subjects have both reached 30 days post-implantation. Overall recruitment will be restricted to a maximum of 100 subjects who receive implants.~~
3.2 Secondary Objectives and Endpoints	Efficacy: • To determine the patency of the HAV (primary, primary assisted and secondary) • To determine the rates of interventions needed to maintain / restore patency in the HAV • To determine the rate of limb salvage (extremity injuries only) • To determine subject survival • To evaluate remodeling of HAV	Replaced original text with **Table 5, Secondary Objectives and Endpoints for Efficacy** Replaced the Objective and Endpoint of **"To determine the primary assisted patency of the HAV at 30 days/Primary assisted patency"** by **Objective and Endpoint "To determine the long-term limb salvage/Amputation" in Secondary Endpoints**
4.1 Description of the Study Design	Removed a redundant sentence:	"~~The DMC will review the earlier of when the first torso patient reaches 3 months post-implantation or when the first 2 torso patients have both reached 30 days post-implantation.~~"
Section 4.3	Section replaced to conform to new plans for primary and secondary analyses; original text reads as follows: Safety:	**Endpoints will be assessed for up to 36 months after HAV implantation. The primary analysis of the study will be conducted when the earlier of these two milestones are reached:**

SUMMARY OF CHANGES to PROTOCOL CLN-PRO-V005 to produce Version 4.0		
Affected Section	**Original Text**	**Amended Text (new or revised text is in bold)**

	• Frequency of adverse events of special interest:
	  • HAV occlusion (thrombosis)
	  • HAV spontaneous rupture
	  • Iatrogenic injuries are not an Event of Special Interest and should be reported as an AE
	  • HAV infection
	  • HAV abandonment
	  • HAV aneurysm
	  • HAV pseudoaneurysm
	HAV Excision (partial or complete)
		a) when the last patient enrolled reaches 30 days post-implantation or
		b) all patients enrolled in the initial 24-month accrual period have reached 30 days after HAV implantation.
		The objectives, endpoints and the corresponding estimands for this study have been updated for this version of the protocol as described in the corresponding Statistical Analysis Plan (SAP).
		• **The primary Safety and Efficacy endpoints of this study are provided in Table 3.**
		• **The secondary Safety endpoints of this study are provided in Table 4.**
		• **The secondary Efficacy endpoints of this study are provided in Table 5.**

5.1. Description of the Study Population	Original text: The study population will consist of patients with vascular trauma to size appropriate vessels in the limb or torso, requiring replacement or reconstruction. Added sample size of 100 patients enrolled	The study population will consist of **patients** with vascular trauma to size appropriate vessels in the limb or torso, requiring replacement or reconstruction. **Overall recruitment will be restricted to a maximum of 100 patients who receive an HAV implant.**
5.1.3.	5.1.3. Enrollment of Patients Aged 14 Through < 18 Years of Age	5.1.3. Enrollment of **Adolescents who have achieved Tanner Stage V Sexual Maturity Rating (US sites only)**
6.2. Manufacturer of the Human Acellular Vessel	Only the current manufacturer (Humacyte) is listed – the information is thus not complete. March 8 2021 is the date the Humacyte manufacturing site was approved.	Details of previous manufacturer and date of transition to manufacturing by Humacyte should be presented for clarity and completeness: **Initially, the HAV was manufactured for Humacyte by:** **Allosource** **6278 S. Troy Circle** **Centennial, CO 80111 USA** **Since 01July2021, all HAVs are now manufactured by:**

SUMMARY OF CHANGES to PROTOCOL CLN-PRO-V005 to produce Version 4.0		
Affected Section	**Original Text**	**Amended Text (new or revised text is in bold)**
		Humacyte Global, Inc. **(previously known as Humacyte, Inc.)** 2525 East NC Highway 54 Durham, NC 27713
7.1.1. Medication Data to be Collected	Original section deleted and moved up to 7.1.1 and edited for clarity	**For each prior or concomitant reportable medication taken by or administered to the patient, the following information must be collected:** • **Medication generic name / components of combination products** • **Dose** • **Route of administration** • **Frequency of administration** • **Date started** • **Date stopped** • **Indication for use**
7.1.2. Prior Medications	Original section revised per reviewer comments and moved up to 7.1.1	Prior medications are defined as all prescription and over the counter (OTC) medications **taken within 7 days before the initial trauma (i.e., prior to Day 1) whether use of that medication was continuing during the study or not.** • **All prior and concomitant medications (including immediately pre-surgery and post-surgery medications) must be listed in the patient's medical record.** • **Drugs used during anesthesia should be recorded in the anesthesia records but should not be transcribed into the eCRF.** • **Drugs used to manage standard issues that may arise during any surgical procedure should not be transcribed into the eCRF, unless they are specifically listed in the reportable medications listed in Table 3**.

SUMMARY OF CHANGES to PROTOCOL CLN-PRO-V005 to produce Version 4.0		
Affected Section	**Original Text**	**Amended Text (new or revised text is in bold)**
7.1.3. Concomitant Medications	Original section replaced per reviewer comments and Table 3 added for clarity	**Concomitant medications are defined as medications taken after the initial trauma, even if this is prior to HAV implantation. Only the concomitant medications described in Table 3 must be recorded on the eCRF.** **After HAV implantation, patients should be questioned at each study visit concerning any new medications or changes in current medications. Particular attention should be made to identify the use of antithrombotic or antiplatelet agents.**
8.1. Clinical Evaluations Through Month 12	Clinical Laboratories: Blood samples for hematology, clinical chemistry at pre-operative screening and Day 5 and PRA at pre-operative screening, Day 30 and the Month 6 Visit.	• Clinical Laboratories: Blood samples for hematology, clinical chemistry at pre-operative screening and Day 5**. Where applicable per country regulations, samples for** PRA **titers should be obtained at pre-operative screening or within 12 hours of HAV implantation, and at the Day 30, Month 6, and Month 12 Visits.**
8.2. Clinical Evaluations in Long Term Follow Up (Post Month 12 to Month 36)	First paragraph	Clarification added to improve sentence structure: " The status of the patient and HAV will be ascertained every 3 months after the Month 12 Visit until the Month 36 Visit. **Patient status may be ascertained via** telephone contact with the patient and/or his physician. "
8.2. Clinical Evaluations in Long Term Follow Up (Post Month 12 to Month 36)	After second paragraph	Added statement from the Synopsis: " **Fatal AEs (Deaths) will be reported through the Month 36 Visit** "
8.3.1. Clinical and Research Laboratory Evaluations and Specimen Collection	3rd bullet: • PRA will be measured at pre-operative screening, Day 30, and the Month 6 Visit.	3rd bullet: • **Where applicable per country regulations,** PRA **titers** will be measured at pre-operative screening **(within 12 hours of HAV implantation), at the** Day 30 Visit, the Month 6 Visit**, and the Month 12 Visit**.
8.5.1. Preoperative Screening (Day 1)	7th bullet:	7th bullet:

SUMMARY OF CHANGES to PROTOCOL CLN-PRO-V005 to produce Version 4.0		
Affected Section	**Original Text**	**Amended Text (new or revised text is in bold)**
	• Laboratory testing (or standard pre-op lab profile for the institution) o Hematology: full blood count and differential o Clinical chemistry; sodium, potassium, calcium, blood urea nitrogen, creatinine, albumin, total bilirubin, glucose (non-fasting) o PRA	• Laboratory testing (or standard pre-op lab profile for the institution) o Hematology: full blood count and differential o Clinical chemistry; sodium, potassium, calcium, blood urea nitrogen, creatinine, albumin, total bilirubin, glucose (non-fasting) o PRA **titer (if not obtained pre-operatively may be obtained within 12 hr. of HAV implantation)**
9.1. Adverse Event Definition	Minor clarifications to grammar and syntax the text were made	"An AE is any untoward medical occurrence in a **patient** administered an **Investigational Medicinal Product (IMP)** and which does not necessarily have a causal relationship with the **HAV**. An AE can, therefore, be any unfavorable and unintended sign, symptom, or disease temporally associated with the use of **the HAV**, whether or not **the AE is considered to be related** to **the HAV**. Any worsening of the patient's disease under study or other medical conditions will also be considered to be an AE, unless it is within the normal range of disease fluctuation for that patient."
9.2. Serious Adverse Event Definition	Entire section replaced	9.2. Serious Adverse Event Definition An AE is considered to be a Serious Adverse Event (SAE) **if it results in any of the following outcomes:** • **Death** • **Is life-threatening** o **An adverse event is considered "life-threatening" if, in the view of either the investigator or sponsor, its occurrence places the subject or patient at immediate risk of death. It does not include an adverse event or suspected adverse reaction that, had it occurred in a more severe form, might have caused death.**

SUMMARY OF CHANGES to PROTOCOL CLN-PRO-V005 to produce Version 4.0		
Affected Section	**Original Text**	**Amended Text (new or revised text is in bold)**
		• **Inpatient hospitalization or prolongation of existing hospitalization.** o **Note that hospitalization for the surgery to implant the HAV is not an SAE. However, prolongation of the initial hospitalization due to an AE will be considered an SAE.** • **Persistent or significant incapacity or substantial disruption of the ability to conduct normal life functions.** • **Congenital anomaly or birth defect** • **Important medical events that may not result in death, be life-threatening, or require hospitalization may be considered serious when, based upon appropriate medical judgment, they may jeopardize the subject or patient and may require medical or surgical intervention to prevent one of the outcomes listed in this definition.**
9.5 Adverse Events of Special Interest	9.5 Adverse Events of Special Interest	9.5. **Adverse** Events of Special Interest
9.5.6 Reporting of ESIs		Removed bullet "need for hospitalization (number of nights)"
9.6. Reporting of Adverse Events	Added 3rd bullet to the list of things to be reported out to the Month 36 Visit	During the long term follow up period after the Month 12 Visit through the Month 36 Visit, only the following will be reported by the investigator: • All SAEs considered related to the HAV • All Events of Special Interest (Section 9.5) • **All Fatal AEs (deaths)**
9.6.5. Reporting Serious Adverse Events	Added clarification in 3rd bullet	"**• Any SAE resulting in patient death (regardless of relatedness attribution) that occurs during Long term Follow up (after the Month 12 Visit through the Month 36 Visit) must be reported by the investigator or designee to the Safety CRO within 24 hours of learning of its occurrence.**"

SUMMARY OF CHANGES to PROTOCOL CLN-PRO-V005 to produce Version 4.0		
Affected Section	**Original Text**	**Amended Text (new or revised text is in bold)**
10. STATISTICAL CONSIDERATIONS	This is a prospective, open label, multicenter pilot study to evaluate the safety and efficacy of the HAV in patients undergoing vascular replacement or reconstruction. The primary objective of this study is to evaluate the safety and tolerability of the HAV in these patients and to determine the primary patency of the HAV at 30 days post-implantation. The secondary objectives of this study are to further assess safety in terms of adverse events of special interest, to determine the rate interventions required to keep the HAV patent, and to further characterize efficacy in terms of secondary patency and limb salvage. There is no formal hypothesis testing planned. Endpoints will be assessed over a period of up to 36 months after HAV implantation. The primary analysis of the study will be conducted on the earlier of a) when the last subject enrolled reaches 30 days post-implant or b) all subjects enrolled in the initial 24 month accrual period have reached 30 days post-implant. Details of data handling and planned descriptive statistics are given in the Statistical Analysis Plan.	**This is a prospective, open label, nonrandomized multicenter Phase 2/ 3 study to evaluate the safety and efficacy of the HAV in patients undergoing vascular replacement or reconstruction. There is no formal hypothesis testing planned. The primary analysis will be conducted when approximately 50 patients complete the 30-day follow-up. Specific details of data handling and analysis will be described in the SAP.**
10.1. Analysis Populations	Section revised to conform to the terminology used in the V005 Statistical Analysis Plan (SAP). Original Text: All patients who receive an HAV will be included in the analyses. For discontinued or withdrawn patients, all available data will be included in the safety and efficacy analyses.	**Three groups of patient will be examined during analysis of study data:** • **The Extremity Group will include patients who underwent arterial vascular repair with an HAV in an extremity. This population was previously referred to as the "Eligible" Group in the Interim Analysis.**

SUMMARY OF CHANGES to PROTOCOL CLN-PRO-V005 to produce Version 4.0		
Affected Section	**Original Text**	**Amended Text (new or revised text is in bold)**
		• **The All HAV Group will include all patients who have received an HAV, regardless of the location of the vessel repaired and the type of injury. This population was previously referred to as the "Total" population or the "Safety" Population" in the Interim Analysis.** • **The Torso+Iatrogenic Group will include all patients who received an HAV in a location in the Torso as well as any patient who received an HAV to repair an iatrogenic injury. This population was previously referred to as the "Non-Eligible" Group in the Interim Analysis.**
10.2. Safety Analyses	Section revised to conform to the terminology used in the V005 SAP for Version 4 Original Text: Safety analyses will be performed on all patients who have had an HAV implanted. The incidence of aneurysm formation, anastomotic bleeding or spontaneous rupture, HAV removal, HAV infection, and inflammation at the implantation site will be tabulated by visit and overall. Adverse events will be coded using Medical Dictionary for Regulatory Activities (MedDRA) terms. Adverse events will be listed and summarized by body system, incidence, severity, and duration. HAV complications will be listed in terms of incidence, severity, and (where appropriate) time to onset and duration. Serious adverse events will be summarized separately.	**10.2. Safety Analyses** **Safety analyses will be performed on all patients in the Extremity Group (as defined in Section 10.1). Similar analyses may be performed in the All HAV Group and the Torso+Iatrogenic Group if the amount and quality of data available permits.** **10.2.1. Primary Safety Analyses** **Primary Safety Analyses for the Extremity Group will include the following:** • **Rate of HAV infection** • **Frequency of Adverse Events of each severity grade.**

SUMMARY OF CHANGES to PROTOCOL CLN-PRO-V005 to produce Version 4.0		
Affected Section	**Original Text**	**Amended Text (new or revised text is in bold)**
	Any premature discontinuations due to adverse events and deaths will be listed and summarized. PRA data will be listed and summarized using appropriate descriptive statistics for the change from baseline values.	
10.2. Safety Analyses	Section revised to conform to the terminology used in the V005 SAP for Version 4 Original Text: Safety analyses will be performed on all patients who have had an HAV implanted. The incidence of aneurysm formation, anastomotic bleeding or spontaneous rupture, HAV removal, HAV infection, and inflammation at the implantation site will be tabulated by visit and overall. Adverse events will be coded using Medical Dictionary for Regulatory Activities (MedDRA) terms. Adverse events will be listed and summarized by body system, incidence, severity, and duration. HAV complications will be listed in terms of incidence, severity, and (where appropriate) time to onset and duration. Serious adverse events will be summarized separately. Any premature discontinuations due to adverse events and deaths will be listed and summarized. PRA data will be listed and summarized using appropriate descriptive statistics for the change from baseline values.	**10.2.2. Secondary Safety Analyses** **Secondary safety analyses may focus on evaluating the mechanical stability of the HAV, based on freedom from aneurysmal degeneration, anastomotic bleeding, spontaneous rupture, infection, or significant stenosis. Safety data evaluated to support these analyses may include:** • **Adverse Events of Special Interest (AESI) and** • **Occurrences of HAV infection.** **To determine the durability of the HAV in arterial vascular repair, the rates of interventions needed to maintain or restore HAV patency will be assessed.** **In addition, other aspects of HAV function may be assessed for safety, data permitting:** • **Amputation rate for the implanted limb** • **Patient mortality** • **Rate of HAV removal** • **Rate of postoperative surgical site infection**
10.3. Primary Efficacy Analyses	Section replaced to conform to the new SAP for Version 4	**10.3. Efficacy Analyses**

SUMMARY OF CHANGES to PROTOCOL CLN-PRO-V005 to produce Version 4.0		
Affected Section	**Original Text**	**Amended Text (new or revised text is in bold)**
		Following discussions with the FDA on 30th March 2022 and in April 2023, the primary group for analysis of efficacy will be defined as the "Extremity Group", formerly known in the Interim Analysis as the "Eligible Group", as defined in Section 10.1 **The sections below provide concise descriptions of the efficacy analyses and endpoints. Specific details will be provided in the SAP.** **Primary patency is defined as functional access patency until any type of intervention; primary assisted patency is defined as an HAV still working without complete thrombosis, with or without interventional or surgical procedures to maintain patency; secondary patency is defined as functional HAV patency, with or without preceding successful interventional or surgical procedures to maintain or reestablish patency, until the HAV is abandoned.**
10.3.1. Primary Efficacy Analyses	10.3.1. Primary Efficacy Analyses The primary efficacy analysis will be the rate of primary patency at 30 days after HAV implantation. Primary, primary assisted, and secondary patency rates of the HAV at 12 months post-implantation and at all other post-implantation visits..... regarding the efficacy parameters employed in this study are consistent with current clinical practice and are meaningful to the research community. Every attempt has been made to minimize the variability on the part of the surgeon when using this product.	**10.3.1. Primary Efficacy Analyses** **Response will be defined as loss of primary patency before day 30 or maintained primary patency for at least 30 days. The proportion of patients with patency at 30-day will be estimated along with 95% confidence interval. The SAP will describe the estimand approach, including the main estimator and sensitivity analyses.**
		Replaced original text with **Table 5, Secondary Objectives and Endpoints for Efficacy**

SUMMARY OF CHANGES to PROTOCOL CLN-PRO-V005 to produce Version 4.0		
Affected Section	**Original Text**	**Amended Text (new or revised text is in bold)**
		Replaced the Objective and Endpoint of **"To determine the primary assisted patency of the HAV at 30 days/Primary assisted patency"** by **Objective and Endpoint "To determine the long-term limb salvage/Amputation" in Secondary Endpoints**
10.3.3 Other Analyses	Added a section "Other Analyses"	**10.3.3 Other Analyses** **Details of other analysis will be described in the SAP.**
10.4 Sample Size Rationale	Up to 100 subjects will be recruited into the study, enrolled from up to 35 sites in the US, Israel, and Ukraine. As this Phase 2/Phase 3 study is the first human study of the HAV for vascular trauma, the study was designed to provide preliminary evidence of safety and efficacy.	**10.4 Other Analyses** **Up to 100 patients will be enrolled in the study. The primary efficacy analysis will be performed after approximately 50 patients in the Extremity Group complete the 30-day follow-up. Sample size rationale for primary efficacy analysis will be provided in the SAP.**
10.5. Interim Analyses	~~Not Applicable~~	**An interim analysis (IA) was performed using a data cutoff of 22April2022. In this IA, a total of 52 patients were included, of which 34 were in the Extremity group.** **The primary efficacy analysis will be performed after approximately 50 patients in the Extremity group complete the 30-day follow-up.**
11.3 Patient Informed Consent	Pediatric enrollment (ages 14 through <18) requires obtaining informed consent from the minors legally authorized representative (parental consent or surrogate consent).	Pediatric enrollment**: Adolescents who have achieved Tanner Stage V Sexual Maturity Rating may be enrolled if the vessel to be repaired is size-appropriate (US sites only);** this requires obtaining informed consent from the minors legally authorized representative (parental consent or surrogate consent).

SUMMARY OF CHANGES to PROTOCOL CLN-PRO-V005 to produce Version 4.0		
Affected Section	**Original Text**	**Amended Text (new or revised text is in bold)**
11.4. Amendments to the Protocol	An amendment must be agreed to in writing by Humacyte and submitted to the FDA and approved by the IRBs....	An amendment must be agreed to in writing by Humacyte and submitted to the FDA **(or other Competent Authorities, if applicable)** and approved by the IRBs....
11.9. Retention of Records	All documentation pertaining to the study will be kept by Humacyte or their designee in accordance with ICH guidelines and US FDA regulations.	All documentation pertaining to the study will be kept by Humacyte or their designee in accordance with ICH guidelines**, US FDA regulations, or applicable country regulations)**.
14.4 Safety Reporting		Replaced "medically significant events" with "Events of Special Interest"
18. Appendix	18. Appendix - previously contained the Schedule of Assessments Table.	**Deleted** as the Schedule of Assessments Table was moved up to follow the Protocol Synopsis

A Phase 2 Study for the Evaluation of Safety and Efficacy of Humacyte's Human Acellular Vessel for Vascular Replacement or Reconstruction in Patients with Life or Limb-threatening Vascular Trauma

Medicinal Product:	Humacyte Human Acellular Vessel (Humacyte HAV)
Study No.:	CLN-PRO-V005
Sponsor:	Humacyte, Inc
Address:	2525 East NC Highway 54 Durham, NC 27713
Phone:	919-313-9633
CRO:	Atlantic Research Group 2421 Ivy Road, Suite 200 Charlottesville, VA 22903
Version:	3.3 (12 August 2022) This Protocol Amendment is Specific to the United States Only (Amendment to version 3.2 dated 18 August 2021)

Confidentiality Statement

STATEMENT OF COMPLIANCE

This trial will be conducted in compliance with the protocol and the following regulatory requirements:

- Declaration of Helsinki adopted by the 18th World Medical Assembly in Helsinki, Finland, in 1964, as last amended by the World Medical Assembly in 2013
- International Conference on Harmonization of Technical Requirements for Registration of Pharmaceuticals for Human Use (ICH), E6 Good Clinical Practice: Consolidated Guidance (ICH E6)
- ICH E2A Clinical Safety Data Management: Definitions and Standards for Expedited Reporting
- ICH E8 Guidance on General Considerations for Clinical Trials
- Applicable sections of United States Food and Drug Administration (FDA) Code of Federal Regulations (CFR), including:
    - 21 CFR Part 11, Electronic Records; Electronic Signatures
    - 21 CFR Part 50, Protection of Human Subjects
    - 21 CFR Part 54, Financial Disclosure by Clinical Investigators
    - 21 CFR Part 56, Institutional Review Boards
    - 21 CFR Part 312, Investigational New Drug Application

PRINCIPAL INVESTIGATOR AGREEMENT PAGE FOR THE PROTOCOL

I agree:

- To assume responsibility for the proper conduct of the study at this site, and to conduct the study in compliance with this protocol, any future amendments, and with any other study conduct procedures provided by the sponsor, Humacyte, Incorporated (Humacyte), or their authorized representatives.

- Not to implement any deviations from or changes to the protocol (including protocol amendments) without agreement from the sponsor and prior review and written approval from the Institutional Review Board (and FDA, if applicable) except where necessary to eliminate an immediate hazard to the patient(s), or for administrative aspects of the study (where permitted by all applicable regulatory requirements).

- That I am familiar with the appropriate use of the investigational medicinal product, as described in this protocol and any other information provided by the sponsor including, but not limited to the current Investigator's Brochure or equivalent document provided by Humacyte.

- To ensure that all persons assisting me with the study are adequately informed about the investigational medicinal product and of their study-related duties and functions.

- That I have been informed that certain regulatory authorities require the sponsor to obtain and supply details about the investigator's ownership interest in the sponsor or the Investigational Medicinal Product, and more generally about his/her financial ties with the sponsor. Humacyte will use and disclose the information solely for the purpose of complying with regulatory requirements.

Principal Investigator: _____

Name and Title

Signed: _____ Date: _____

PROTOCOL APPROVAL

Sponsor Medical Approval: Shamik Parikh, MD, Chief Medical Officer, Humacyte

Signed: Shamik Parikh Date: Aug 12, 2022

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LIST OF ABBREVIATIONS

AE	Adverse event
AIDS	Acquired immunodeficiency syndrome
ALT	Alanine aminotransferase
aPTT	Activated partial thromboplastin time
ASA	Acetylsalicylic acid (aspirin)
ABI	Ankle brachial index
AST	Aspartate aminotransferase
AV	Arteriovenous
AVF	Autologous arteriovenous fistula
BP	Blood pressure
CAVG	Canine acellular vascular graft
CBC	Complete blood count
CKD	Chronic kidney disease
CTA	Computed tomography angiography
CM	Concomitant medication
eCRF	Electronic case report form
CRO	Contract research organization
DMC	Data Monitoring Committee
DTH	Delayed-type hypersensitivity
ECG	Electrocardiogram
ECM	Extracellular Matrix
ePTFE	Expanded polytetrafluoroethylene
ESRD	End-stage renal disease
ET	Early termination

LIST OF ABBREVIATIONS

FDA	Food and Drug Administration
GCP	Good Clinical Practice
GLP	Good Laboratory Practice
HAV	Human acellular vessel
HIV	Human immunodeficiency virus
IB	Investigator Brochure
ICF	Informed consent form
ICH	International Conference on Harmonization of Technical Requirements for Registration of Pharmaceuticals for Human Use
IgG	Immunoglobulin G
IHC	Immunohistochemistry
IM	Intramuscular
IMP	Investigational medicinal product
INR	International normalized ratio
IRB	Institutional Review Board
ISO	International Organization for Standardization
IU	International unit
IV	Intravenous
MedDRA	Medical Dictionary for Regulatory Activities
M	Month
N	Number (typically refers to participants)
NYHA	New York Heart Association
OTC	Over-the-counter
PAD	Peripheral arterial disease

LIST OF ABBREVIATIONS

PE	Physical examination
PHI	Protected health information
PI	Principal Investigator
PRA	Panel reactive antibodies
PT	Prothrombin time
PTFE	Polytetrafluoroethylene
QA	Quality Assurance
QC	Quality Control
RRT	Renal replacement therapy
SAE	Serious adverse event
SFA	Superficial Femoral Artery
SOP	Standard operating procedure
SVS WIfI	Society for Vascular Surgery: Wound, Ischemia, and foot Infection
US	Ultrasound
USA	United States of America
WFI	Water for injection
WBC	White blood cell(s)
WHO	World Health Organization

PROTOCOL SUMMARY

Full Title	A Phase 2 Study for the Evaluation of Safety and Efficacy of Humacyte's Human Acellular Vessel for Vascular Replacement or Reconstruction in Patients with Life or Limb-threatening Vascular Trauma
Clinical Trial Phase	Phase 2
Sponsor	Humacyte, Inc.
Planned Study Sites	Up to 25 sites in the United States and Israel
Sample Size	Up to 100 subjects
Study Population	Patients with vascular trauma to size appropriate vessels in the limb or torso, requiring replacement or reconstruction.
Inclusion Criteria	1. Patients with life or limb threatening traumatic injury to an arterial vessel in the limb or torso, other than the heart, which requires replacement or reconstruction. 2. Preoperative imaging or clinical examination indicates the damaged vessel has a defect length of ≤ 38cm and is appropriately size matched to the 6mm Human Acellular Vessel (HAV) per the judgment of the treating surgeon taking into account vasoconstriction and situational inflow and outflow considerations. 3. Autologous vein graft is either not feasible in the judgment of the treating surgeon (e.g. because of lack of availability of suitable conduit, presence of severe venous insufficiency) or is not desirable because of the urgency of revascularization 4. Aged 14 to 85 years old, inclusive 5. Able to communicate meaningfully with investigative staff and able to comply with study procedures. If the patient is unconscious then information from a reliable witness indicates that the patient would normally be able to understand and comply with study procedures 6. Patient or legal representative is able, willing and competent to give informed consent 7. Life expectancy of at least 1 year

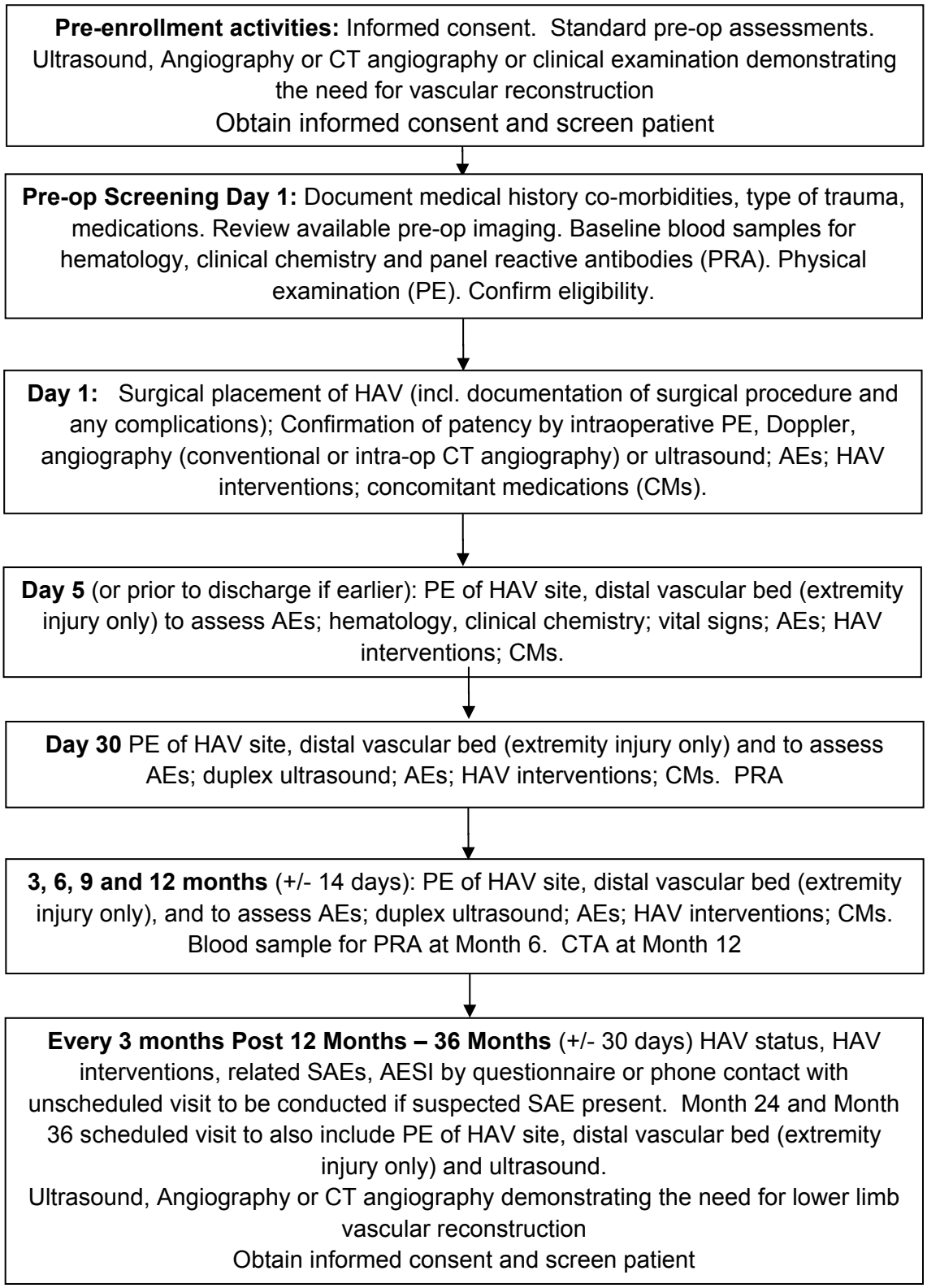
Exclusion Criteria	1. Mangled Extremity Severity Score (MESS) of ≥ 7. 2. Limb at high risk of amputation despite vascular reconstruction (e.g., because of crush injury) 3. Catastrophic injuries that make survival unlikely (e.g. Abbreviated Injury Scale (AIS) > 5 or Injury Severity Score (ISS) >60) 4. HAV may not be used for coronary artery repair. 5. Known pregnant women 6. Known medical condition which would preclude long term antiplatelet therapy after resolution of acute injuries 7. Any other condition which in the judgment of the investigator would preclude adequate evaluation of the safety and efficacy of the Humacyte Human Acellular Vessel (HAV) 8. Previous exposure to HAV 9. Known participation in any investigational study within the last 30 days 10. Employees of the sponsor or patients who are employees or relatives of the investigator
Expected Enrollment Start	3Q 2018
Accrual Period	24 months
Study Duration	The active study duration for each study participant will be 36 months from HAV implantation. All subjects will be followed for the initial 12 months. Beyond 12 months (Long-Term Follow Up), only subjects with a patent HAV will be followed out to a total of 36 months from HAV implantation. The total expected duration of the clinical study is 61 months.
Study Design	Prospective, multicenter, non-randomized study
Investigational Medicinal Product/Intervention Description	Patients will be implanted with a Humacyte Human Acellular Vessel (HAV) as an interposition replacement or bypass using standard vascular surgical techniques.

Primary Objectives	**Safety** To evaluate the safety and tolerability of the Humacyte HAV in vascular trauma patients undergoing surgery for vascular replacement or reconstruction **Efficacy** To determine the rate of primary patency at 30 days
Secondary Objectives	**Safety** • To determine mechanical stability of the HAV based on freedom from aneurysmal degeneration, anastomotic bleeding or spontaneous rupture, infection, or significant stenosis • To determine HAV durability in terms of freedom from HAV removal or replacement **Efficacy** • To determine the patency of the HAV (primary, primary assisted and secondary) • To determine the rates of interventions needed to maintain / restore patency in the HAV • To determine the rate of limb salvage • To determine patient survival • To evaluate remodeling of HAV
Endpoints	Endpoints will be assessed for up to 36 months after HAV implantation. The primary analysis of the study will be conducted on the earlier of a) when the last subject enrolled reaches 30 days post-implant or b) all subjects enrolled in the initial 24 month accrual period have reached 30 days post-implant. **Primary Endpoints:** *Safety:* • Frequency and severity of adverse events *Efficacy:* • HAV primary patency at 30 days

	Secondary Endpoints:
	Safety:
	• Frequency of adverse events of special interest:
	  ○ HAV occlusion (thrombosis)
	  ○ HAV spontaneous rupture
	  ○ Iatrogenic injuries are not an Event of Special Interest and should be reported as an AE
	  ○ HAV infection
	  ○ HAV abandonment
	  ○ HAV aneurysm
	  ○ HAV pseudoaneurysm
	  ○ HAV Excision (partial or complete)
	• HAV partial or complete removal
	Efficacy:
	• HAV primary patency
	• HAV primary assisted patency
	• HAV secondary patency
	• Rate of HAV interventions
	• Limb salvage (extremity injury only)
	• Patient survival
	• HAV remodeling as shown by histopathology of any clinical explants

Protocol Approval (Version and Date)	Version 3.3 (US Only) 12 August 2022

Schematic of Study Design:



1. STUDY PERSONNEL

An updated list of all study personnel will be maintained by the CRO. Protocol amendments will not be required for staff changes at Humacyte, the CRO or the sites (except change of Principal Investigator at a site).

CRO Project Manager:

Brad Whitlow
Project Manager
Atlantic Research Group
2421 Ivy Road, Suite 200
Charlottesville, VA 22903

Email: bwhitlow@atlanticresearchgroup.com
Mobile: 828-442-0526
Office: 434-202-4537
Fax: 434-202-4537

Sponsor Medical Representative:

Shamik Parikh, MD
Chief Medical Officer
Humacyte, Inc.
2525 East NC Highway 54
Durham, NC 27713, USA

Email: sparikh@humacyte.com
Office: 919-313-9633

Sponsor Regulatory Representative:

William Tente
Chief Regulatory Officer
Humacyte, Inc.
2525 East NC Highway 54
Durham, NC 27713, USA

Email: tente@humacyte.com
Mobile: 401-714-4151
Office: 919-313-9633

Sponsor Clinical Operations Representative:

Elizabeth Taylor
Director, Clinical Operations
Humacyte, Inc.
2525 East NC Highway 54
Durham, NC 27713, USA

Email: etaylor@humacyte.com
Office: 919-313-9633

CRO Safety Representative:

Todd Fisher
Sr. Drug Safety Specialist
Atlantic Research Group
2421 Ivy Road
Charlottesville, VA 22903

Email: safety@atlanticresearchgroup.com
Phone: 888-619-3216
Fax: 866-851-9318

Medical Monitor:

Jennifer Prichard, MD
Medical Monitor
Atlantic Research Group
2421 Ivy Road
Charlottesville, VA 22903

Email: jprichard@atlanticresearchgroup.com
Mobile: 434-249-5530
Phone: 888-619-3216
Fax: 866-851-9318

2. BACKGROUND INFORMATION AND SCIENTIFIC RATIONALE

2.1. Background Information

In the civilian population, traumatic vascular injuries are mainly concentrated to the limbs and torso (abdomen, retroperitoneum, thorax and thoracic outlet). According to the PROspective Observational Vascular Injury Treatment (PROOVIT) database, designed to collect vascular trauma injuries from 24 Level I and Level II trauma centers in the United States, vascular injuries in the lower limb, torso, upper limb and neck have a distribution of 41%, 30%, 22%, and 6%, respectively. (DuBose, 2015, Faulconer, 2017) These reports incorporate a significant percentage of relevant venous injuries in addition to arterial. Additionally, in the civilian population, lower extremity bone fractures with associated arterial injuries are common, due to motor vehicle accidents, gunshot wounds, dog bites, and other situations resulting in blunt or penetrating trauma. (Helfet, 1990, Andrikopoulos, 1995, Akingba, 2012) In fact, the incidence of this type of vascular injury has increased considerably in the past 50 years. (Andrikopoulos, 1995) Although this type of injury represents less than 1% of all civilian injuries, fractures with associated vascular damage require special attention because of their potentially severe complications, including limb necrosis and amputation.

Currently, in order to attempt to salvage the injured limb or end-organ in the distal vascular bed and to prevent life threatening hemorrhage the vascular component of these injuries are treated with interposition or bypass grafting. This type of reconstruction is performed by using either autologous vein from the patient (typically great saphenous vein), or by using a synthetic graft, such as ePTFE or Dacron. However, the use of these grafts in the civilian population is not always possible or without additional risks. The patient may not have adequate autologous vein for harvest, and in many of these cases, such as dog bite injuries, the wound is “dirty” and a synthetic vascular substitute is contraindicated due to the risk of infection. (Akingba, 2012) Thus, civilians would benefit from a vascular graft that does not contain infection-prone synthetic material and has similar properties as human tissue, but does not require time consuming or high morbidity procedures to harvest vessels from the patient.

Warfighters could also benefit from an off-the-shelf biologic vascular graft. In modern combat, the incidence of vascular injury is much greater than in previous wars. The rate of vascular injury in the Vietnam War was 2-3%. But between 2002-2009, the rate of vascular injury was over 12% in a study of over 13,000 battlefield injuries. In combat scenarios vascular injuries occur in all locations of the body, but injuries to the lower extremities are the most common, followed by vascular injuries to the upper extremity, and then neck, followed by torso. (Rasmussen, 2010) Improvised Explosive Devices, or IED’s, caused more than 3,000 casualties per year during recent conflicts. (Cordesman, 2010) Arterial damage, laceration and thrombosis can require vascular reconstruction to save tissues from ischemia, necrosis, and further amputation. Harvesting autologous vein for vascular reconstruction is problematic because IED casualties often have multi-limb injuries, making harvest of autologous vein highly risky or impossible.

(Holcomb, 2011) Synthetic vascular reconstruction using synthetic vascular grafts made from Teflon (ePTFE)/Dacron is relatively contraindicated, since IED wounds are always “dirty”, and bacteria in the wound can colonize the synthetic graft, causing abscesses and sepsis.

Thus, there is a significant unmet medical need for alternative grafts, which can be used in situations where autologous vein is unavailable or undesirable to use and which more closely mimic human vascular tissue to avoid or reduce the infection complications associated with ePTFE and Dacron.

2.2. Scientific Rationale

Humacyte has developed an acellular, human tissue engineered vascular conduit, the human acellular vessel (HAV, to provide an alternative to synthetic materials and to autologous grafts in the repair of traumatic vascular damage. Because this product mimics native vascular tissue, it may possess the advantages of an autologous graft; it also has the benefits of synthetic grafts in that it is available off-the-shelf. Use of an off-the-shelf product avoids the surgical morbidity associated with vein graft harvest and most importantly allows vessel bypass surgery in patients who have no suitable vessels available. Because the product mimics a native vessel, it may not have the compliance mismatch associated with synthetic alternatives. In addition, pre-clinical studies in pigs, canines and primates have shown that the HAVs resist intimal hyperplasia at the anastomoses. (Quint, 2011, Prichard, 2011, Dahl, 2011) Upon implantation, the collagen matrix comprising the HAV is infiltrated with host cells and remodeled by the host. This could result in a vascular structure more similar to the histological composition of the native vascular tissue that may improve bypass longevity and be less likely to become infected. The latter potential advantage is of high importance in the repair of peripheral vascular trauma where most wounds are heavily contaminated.

2.3. Summary of Nonclinical Information

The non-clinical testing program was designed to comprehensively address:

- local and systemic effects of the product in multiple in vivo animal models acutely and chronically,
- functional aspects of product implanted into animal models as an arteriovenous conduit
- biocompatibility of the HAV material in standardized in vitro and in vivo test protocols.

Overall, the results of these studies indicated that the HAV extracellular matrix material was non-toxic, well tolerated, and met standards for biocompatibility. Generally, the HAVs functioned as intended and maintained patency during the implantation period. (See the Investigator Brochure for a detailed summary of non-clinical data.)

Pre-implantation, the HAV has mechanical properties (burst pressure and suture retention strength) comparable with native human artery and vein. (Table 1.) There was no evidence that HAV strength deteriorated after long-term implantation into baboons.

Table 1: Summary of Mechanical Properties of Explanted Acellular Vessels

Test Material	Burst Pressure (mm Hg)	Suture Strength (g)
Pre-Implant Humacyte HAVs	3415 +/- 1011 (n=4)	180 +/- 44 (n=12)
Post-Explant Humacyte HAVs	3669 +/- 1305 (n=5)	276 +/- 84 (n=11)
Human Saphenous Vein	1,680 – 2,273[a]	196 +/- 2 (n=7)[a]
Human Artery	2,031 – 4,225[a]	200 +/- 119 (n=9) [a]

[a] From L'Heureux et al, *Nature Medicine*, 2006. (L'Heureux, 2006)

In the chronic animal testing Humacyte vessels produced using canine cells were implanted into 12 dogs (canine acellular vascular graft, CAVG) and 14 baboons (human acellular vessel -HAV) in a variety of anatomical locations. In general, the Humacyte vessels were safe and well tolerated, and functioned as intended.

Mechanical failure was not observed in any HAV. Calcification was not observed in any CAVG or HAV. No graft exhibited hemodynamically significant intimal hyperplasia. Unlike with ePTFE graft implantation, no evidence of systemic infection attributable to implantation of HAV was observed in any of the animals. One HAV developed an aneurysm that was resected and did not harm the animal. The HAV material showed no evidence of toxicity in hematology, clinical chemistry, and necropsy data. The HAVs could be accessed by venipuncture and hemostasis was achieved following needle puncture.

On microscopic analysis, the HAVs were found to be well integrated into the host tissue. Overall, the cellular host response to the HAVs demonstrated smooth muscle actin-positive cells within the vessel wall, endothelial cells lining the lumen, and an adventitial-like outer layer adjacent to the vessel. These findings indicate that implanted HAVs were populated with cell types that are characteristic of healthy native vasculature. Examination of the anastomotic sections showed that the HAVs were well integrated with adjoining vasculature with minimal intimal hyperplasia observed. Furthermore, IHC was employed to identify CD-68 positive macrophages in the venous intimal tissue. Studies have shown a substantial macrophage population has been observed within venous intimal tissue adjacent to inflammatory ePTFE arteriovenous grafts. (Kelly, 2002, Roy-Chaudhury, 2001) Only sparse CD-68 positive macrophages were observed, indicating that the degree and the aggressiveness of the intimal hyperplasia associated with the HAV were less than that typically associated with ePTFE grafts. (Prichard, 2011)

Over time, the organization and composition of extracellular matrix (ECM) components indicated that, aided by infiltration of host vascular cells, HAVs were remodeled in vivo in a manner that mimicked the dynamic remodeling process of native blood vessels. Given the difficulties associated with the baboon animal model, where mismatches in vein vs graft diameter were encountered and animals perturbed their wounds postoperatively, an overall assisted patency rate of approximately 80% (11/14) was achieved. In a xenogeneic transplant model that did not employ immunosuppression, the HAV material did not elicit biologically significant cellular or

delayed-type hypersensitivity (DTH) immune responses. All animals developed immunoglobulin G (IgG) titers to the HAV material that did not appear to detrimentally impact vessel function.

In internationally recognized in vitro and in vivo International Organization for Standardization (ISO) test protocols, the HAV material met criteria for biocompatibility required of medical devices.

These data collectively support the safety of the HAV for the proposed clinical investigation.

2.4. Summary of Clinical Studies

2.4.1. Overview

The HAV clinical development program currently includes 7 clinical studies: 4 in patients with end-stage renal disease receiving hemodialysis (CLN-PRO-V001, CLN-PRO-V003, CLN-PRO-V006 and CLN-PRO-V007), 2 in patients with peripheral arterial disease (CLN-PRO-V002 and CLN-PRO-V004) and 1 in patients with vascular trauma (CLN-PRO-V005). Three Phase 1/2 studies have completed primary analysis with long-term follow-up ongoing (CLN-PRO-V001, CLN-PRO-V002 and CLN-PRO-V003), 1 phase 3 study completed enrollment and follow-up is ongoing (CLN-PRO-V006), 2 phase 2 studies (CLN-PRO-V004, CLN-PRO-V005) and one phase 3 study (CLN-PRO-V007) are open for enrollment.

As of 10 April 2018, 272 patients (244 hemodialysis access patients and 28 PAD patients) have received a HAV. The first implant for hemodialysis was performed in December 2012, and the first peripheral arterial bypass in October 2013. Overall, the total treatment exposure is approximately 329 patient years in the hemodialysis access population and 55 patient years in the PAD population. More information on the clinical profile of the HAV in these ongoing studies is provided in the Investigator Brochure.

2.4.2. Experience in Peripheral Arterial Bypass Patients

Humacyte has two phase 2 studies to assess the safety and efficacy of the HAV when used as an above-knee arterial bypass graft. The first study, CLN-PRO-V002, is a single group uncontrolled study conducted at 3 sites in Poland that is fully enrolled and in long-term follow up. Eligible patients required a femoro-popliteal bypass graft for the management of symptomatic peripheral arterial disease. Pre-operative imaging (conventional or CT angiography) must have demonstrated at least two below knee vessels patent to the ankle with good runoff. The proximal anastomosis was expected to be below the inguinal ligament and the distal anastomosis above the knee. Autologous vein grafts must not have been suitable or feasible (e.g., because of severe venous disease or prior use of leg veins for other bypass surgery or there is a clinical need to preserve those veins for future bypass surgery in the coronary or peripheral circulation).

The HAV was implanted using standard vascular surgical techniques and the patency of the bypass confirmed by intraoperative angiography (conventional or intra-op CT angiography) or ultrasound. The patient was then followed up at study visits at 15 days, 6 weeks and 3, 6, 12, 18

and 24 months. At each visit safety was assessed by clinical examination and adverse events, and the HAV was examined using duplex ultrasound to visualize the entire length to confirm patency, flow and to detect stenosis, aneurysm development or dilatation.

The primary objectives of the study are to evaluate the safety and tolerability of the Humacyte HAV in PAD patients undergoing above-knee femoro-popliteal bypass surgery and to determine the patency (primary, primary assisted and secondary) rate of the Humacyte HAV at 24 months. Secondary objectives include assessment of the panel reactive antibodies (PRA)) and IgG response to the HAV and to assess patency (primary, primary assisted and secondary) at 6, 12 and 18 months, to determine the rates of interventions needed to maintain / restore patency in the HAV, to assess any effect of implantation on claudication, rest pain and ischemic ulcers and to assess any effect on ankle-brachial index (ABI).

The second PAD study of similar design, CLN-PRO-V004, is being conducted in the US with enrollment ongoing.

2.4.2.1. CLN-PRO-V002 Study Results (24 M)

Recruitment began in October 2013 and was completed in June 2014 with 20 patients implanted. Thirteen patients completed the 2 year follow up visit. Of the seven patients terminating the study early, three died and four were withdrawn after occlusion of the HAV. None of the deaths were considered related to the investigational device or procedure.

Kaplan-Meier analyses in which deaths were censored revealed primary, primary assisted, and secondary patency probability rates of 79.2%, 79.0%, and 89.5% at Week 26, 63.3%, 63.2%, and 84.2% at Month 12, 63.3%, 63.2%, and 79.0% at Month 18, and 58.1%, 57.9%, and 73.7% at Month 24.

Six patients (30%) required at least 1 graft intervention to maintain or restore HAV patency during the study. Four patients required 1 intervention and 1 patient each required 3 and 4 interventions. Most interventions successfully restored patency. However, in 1 patient the graft patency could not be restored and the HAV was replaced with an alternative bypass graft. Two patients, who had previously undergone successful interventions, developed a recurrent thrombosis which was not treated and the HAV was left occluded. Two patients experienced HAV thrombosis with no or minimal symptoms and refused interventions on the HAV.

All 20 patients experienced AEs (a total of 92 events). Thirty-one of these events in 13 patients were considered serious. The most frequent AEs reported included graft thrombosis (35% of patients), anastomotic stenosis (20% of patients), lymphocele (20% of patients), and local swelling (15% of patient). Those SAEs reported by at least 2 patients were graft thrombosis (6 patients, 30%) and anastomotic stenosis (2 patients, 10%).

No patient showed an increase in PRA levels. Two patients had a significant (>2 fold) increase from baseline in IgG levels. One of these patients experienced a thrombosis of the HAV between 3 and 6 months after implantation, while the other patient has had no HAV-related AEs and

continues to have primary patency. Neither patient has had any evidence of dilatation or structural degeneration of the HAV.

CLN-PRO-V002 Conclusions:

- Humacyte HAV was safe and well tolerated in PAD patients.
- The HAV is able to withstand long term use in a high pressure, high outflow resistance arterial circuit.
- Patency rates for the HAV are within the ranges of patency rates of synthetic and autologous grafts presented in the literature.
- Humacyte HAV was not immunogenic.

2.4.3. Experience in Hemodialysis Patients

Two phase 2 trials, one in Poland (CLN-PRO-V001) and one in the US (CLN-PRO-V003) have completed enrollment. Both recruited subjects requiring hemodialysis access for end-stage renal disease whom were not suitable for creation of an AVF. Most subjects had undergone previous vascular access procedures, in many cases multiple attempts including both AVFs and synthetic grafts. Initial results from these phase 2 studies are discussed below.

The primary objectives of these two studies are to evaluate both the safety of HAV and its efficacy in terms of primary and secondary patency at 6 months. Secondary objectives include measurement of a panel of reactive antibodies (PRA) response, development of IgG antibodies to the extracellular matrix material in the HAV and a 2-year evaluation of patency and an assessment of the need for interventions to maintain/restore patency. Follow up has now been extended up to 120 months.

A phase 3 randomized study comparing HAV with ePTFE grafts (CLN-PRO-V006) has completed enrollment in the US, Europe, and Israel. A second phase 3 randomized study (CLN-PRO-V007) comparing HAV with AVF is currently enrolling in the US. As the sponsor is blinded, no efficacy information currently available for the phase 3 studies; however, blinded safety data is presented in the Investigator Brochure.

2.4.3.1. CLN-PRO-V001 and CLN-PRO-V003 Study Results (24 M)

All subjects (n=60) have now completed at least 24 months since implantation (or had a censoring event). The first subjects recruited are now beyond 60 months after HAV implantation, some with functioning HAV for hemodialysis access. Together these two trials provide more than 150 years of follow up during which the HAV has been used for more than 15,000 hemodialysis sessions.

When HAV thrombosis has occurred it has almost always been managed successfully, often allowing immediate resumption of dialysis without the need for the placement of a dialysis catheter. One non-serious arteriovenous graft aneurysm was reported in Study CLN PRO V001 (moderate in intensity, considered possibly related to IMP and considered not related to procedure – this patient died before the Sponsor could complete the follow up of this event). An expected

number of small pseudoaneurysms have been observed, which is consistent with all surgically-created hemodialysis access. Most have resolved spontaneously with only 2 cases requiring surgical intervention. Flow rates through the HAV were more than sufficient to allow for effective dialysis.

In both studies, the product has generally been well tolerated and blood chemistry, hematology and coagulation data are not indicative of any HAV-associated toxicity. Immunogenic response to the HAV material has not been observed as demonstrated by a general lack of HAV-related change in PRA levels (Class I or II). Three subjects had elevations in their PRA levels: all 3 subjects had experienced one or more renal transplant failures; one subject recently; one subject developed septic shock about a month before the elevated value; and the third subject, who was severely debilitated with a decubitus ulcer, died approximately a month after HAV abandonment.

IgG titers increased in 5 subjects; in 4 cases the IgG titer increased and then decreased while the HAV remained functional with no clinical evidence of an inflammatory response; in one case the IgG titer increase occurred in a subject who maintained primary patency.

Adverse events (AEs) related to the HAV / access site (excluding thrombotic events) were few; there have been only three access-site infections, of which only one required removal of part of the HAV. There have been:

- 1 transplant (known to be functioning well at 12 months post-transplant)
- 15 deaths, all after abandonment or during follow-up; none of the deaths were considered related to the presence of the HAV

Patency data for the two studies in dialysis access has been pooled for a combined Kaplan Meier analysis (Lawson, 2016). Based on these K-M plots the patency at 6, 12 and 24 months is estimated to be 60%, 26% and 15% (primary patency) and 97%, 91% and 77% (secondary patency).

2.4.4. Human Acellular Vessel Host Response and Remodeling Data

Humacyte has been able to assess the general host response to the HAV in a number of human participants; this was accomplished through the microscopic examination of explanted HAV and adjoining tissue samples obtained during surgical revision procedures in eight cases. The analysis (mostly of a section close to the venous anastomosis) included assessments of:

- Cellular infiltration of histotypic, inflammatory and immunological populations.
- Extracellular remodeling processes, including neo-synthesis and reorganization of ECM components that typically occur in native blood vessels.

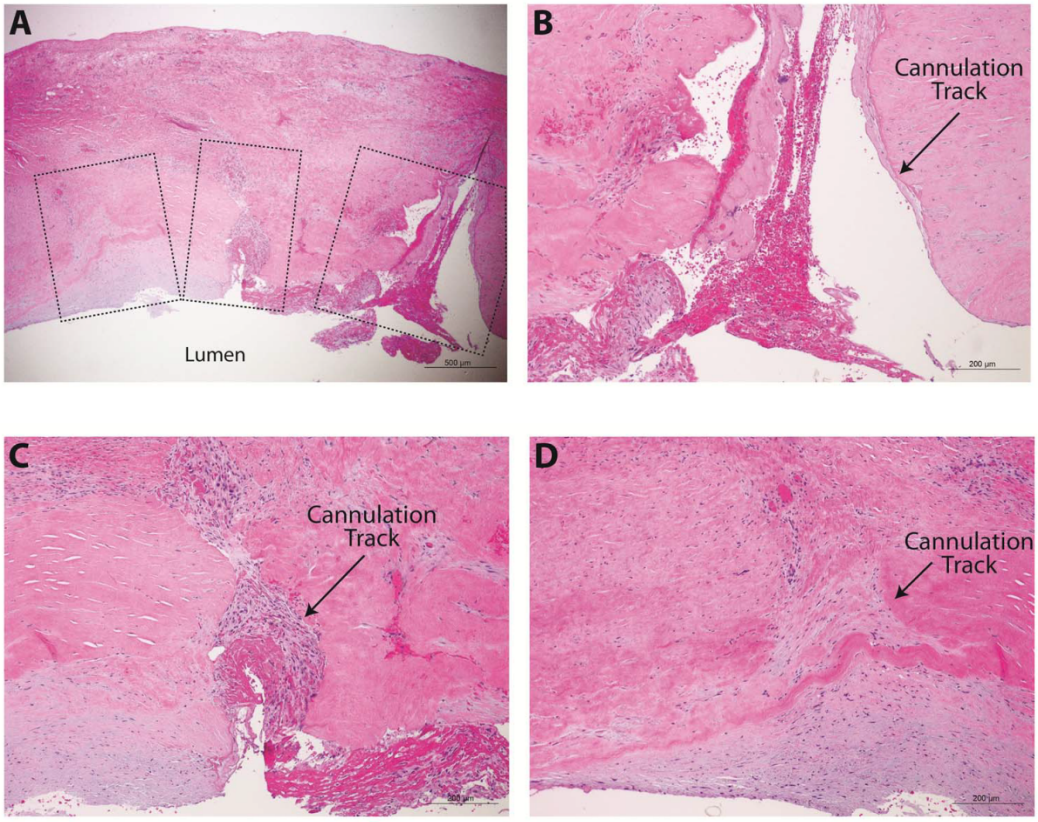
In these cases, small segments of the HAV and adjacent vascular tissue were explanted, fixed in formalin solution and shipped to Humacyte for analysis. Implant duration ranged from 16 to 55 weeks (median: 37 weeks).

In man, the HAV remodeled in a manner consistent with that observed in primate studies. There was infiltration of cell populations that are normally associated with angiogenesis and vascular organization and structure; namely, those with endothelial, smooth muscle and fibroblastic phenotypic characteristics were observed. Endothelial cells formed a monolayer on the luminal surface of the HAV. Migration of actin-positive smooth muscle cells into the wall of the HAV was consistently observed. A well-vascularized adventitial layer of non-constrictive fibrous tissue formed around HAV. Infiltration of the graft material by inflammatory and immunoreactive cell populations was either not evident or was mild and generally unremarkable. Degradation or breakdown of the implant was not observed.

Histotypic neo-synthesis and reorganization of the ECM was observed in patterns indicative of integration of the HAV into the host. An increase in the density of collagen type I, the main type of collagen found in the wall of native blood vessels, was apparent in the majority of HAV explant specimens. The structure of collagen type I in these specimens exhibited a more mature, organized pattern, with distinct fibers and a prominent circumferential alignment evident in explanted samples in comparison with pre-implant specimens. In some specimens, the fibrillar staining pattern of collagen III became more prominent and more organized, with a circumferential orientation. Fibronectin levels and staining patterns remained unchanged.

Cannulation sites within the HAV appeared to be repaired by the host in a fashion similar to wound repair in the body (Figure 1). In one case, an explanted specimen was tested for suture retention strength at the time of explant and exhibited a substantial increase over the pre-implant level.

Figure 1: Images of Mid-Vessel Segment Explanted at 11-Months Post-Implant



A: Low magnification showing 3 cannulation sites (in dashed boxes),

B: Fresh cannulation track,

C: Cannulation track during remodeling

D: Older cannulation track that has been repaired.

The images above show a mid-vessel segment explanted at 11 months post-implant, and shows several prior cannulation tracts from dialysis access. Section B shows a very recent cannulation site with fresh clot extending into the tract from the lumen. Sections C and D show partially healed cannulation tracts, with evidence of cellular repopulation extending in from the lumen. Remodeled cannulation tracks contain new collagen and a few micro-conduits.

In conclusion, the HAVs were remodeled by the host to form a vascular-like structure more similar to the histological appearance of native vasculature. The HAVs were repopulated by cell types that are characteristic of healthy native vasculature. Evidence of ECM remodeling processes, including neo-synthesis and reorganization of ECM components that typically occur in native blood vessels, were observed. The cellular infiltration and ECM remodeling patterns were indicative of the integration of the HAV into the host.

2.4.5. Conclusions

Clinical experience indicates that the HAV remains mechanically strong over implantation periods of more than 60 months with no evidence of dilatation. During more than 200 patient-years of follow up across the three phase 2 studies only one case of infection of the HAV material itself has been reported. The serious adverse event (SAE) profile has been typical of that expected in the dialysis and PAD populations. In hemodialysis populations, secondary patency of the HAVs is substantially higher than the historical data for both ePTFE and AVF (accounting for non-maturation). In PAD, patency is in line with historical ePTFE and autologous conduit for above knee bypass. No evidence of immunogenicity of the HAV has been found and the HAV remains mechanically robust even after repeated puncture for hemodialysis and under high pressure, high outflow resistance in arterial reconstruction.

These data support the use of HAV in future phase 2 and phase 3 studies for vascular replacement and reconstruction in diseased or damaged (trauma) vessels.

2.5. Potential Risks and Benefits

2.5.1. Potential Risks

It is anticipated that subjects participating in the study will be exposed to similar risks to those associated with other arterial conduits. Risks associated with the study investigational product may include but are not limited to:

- Thrombosis/occlusion of the conduit or host vessels, with consequent limb ischemia
- Embolism from a thrombosed conduit
- Bleeding and hematoma formation at the surgical site
- Infection – at the surgical site or systemic
- Stenosis of the conduit or its anastomoses
- Aneurysm or pseudoaneurysm formation
- Swelling of the limb
- Failure/injury to the target end-organ
- Bleeding/hemorrhage in the peritoneum or retroperitoneum

Regular clinical examination of the HAV implantation site and assessment of the patency, blood flow and diameter using ultrasound during the study should allow early detection of complications and permit appropriate intervention including HAV explantation.

The HAV is grown using donor human aortic smooth muscle cells. The vessel is decellularized during manufacturing and thus consists of human extracellular matrix proteins. It is possible that the HAV may provoke an immune response which may lead to damage to the HAV and possible

cross reactivity against host proteins. Possible antibody formation will be assessed by analyzing PRA.

2.5.2. Potential Benefits

Patients who undergo implantation of the Humacyte HAV may benefit from improved patency and a reduced number of interventions versus a conventional ePTFE or Dacron graft. This may result from a decreased propensity for anastomotic and downstream neointimal hyperplasia, which often leads to graft occlusion with synthetic grafts. In addition, risks of infection typically encountered with conventional synthetic grafts may be decreased with the Humacyte HAV. Finally, the longevity (secondary patency) of the Humacyte HAV may be greater than that of conventional synthetic grafts.

Patients may also benefit from reduced morbidity secondary to harvest of autologous vessels for vascular reconstruction. Harvest of autologous vascular conduit requires additional time to perform and in the urgent/emergent trauma scenario, time is of the essence and is key to positive patient outcomes. Use of saphenous, or other autologous vessels for purposes of vascular repair in young, otherwise healthy patients prevents its future potential use for other indications (e.g. coronary bypass, etc.). Vascular reconstruction using synthetic vascular grafts (ePTFE, or woven materials like Dacron) may be contraindicated for use in trauma scenarios since the wounds are often contaminated allowing bacteria in the wound to colonize the synthetic graft potentially leading to abscesses, hemorrhage secondary to anastomotic blow out, blood stream infection, and sepsis. Unlike other readily available synthetic vascular grafts made from ePTFE or Dacron, HAVs are comprised of human proteins, resulting in grafts that may be less prone to infection than synthetic materials, as shown in pre-clinical studies. (Kirkton, 2018)

2.5.3. Risk-Benefit Rationale

The risks anticipated in this study are similar to those associated with currently marketed prosthetic grafts used for vascular repair. The potential advantages of the HAV compared to currently marketed grafts may lead to a lower complication rate and reduced need for surgical intervention and graft replacement as well as the potential for reduced secondary complications associated with autologous vessel harvest.

This is the first in man study in which the HAV will be used to repair vascular injuries within the torso (thorax, thoracic outlet, abdomen, and retroperitoneum) and so risks specific to those locations have not been specifically characterized. However, these risks are not expected to be significantly different than those experienced/reported when used in the upper or lower extremity for AV access or arterial reconstruction. The DMC will review safety data of the torso injuries on the earlier of when the first torso subject reaches 3 months post-implantation or when the first 2 torso subjects have both reached 30 days post-implantation. Overall recruitment will be restricted to a maximum of 100 subjects who receive implants.

This is the first in man study in which the HAV will be used to repair vascular injuries in a pediatric population. Because enrollment in this study is limited to older adolescents who have reached pubertal development (Tanner Stage 5), these risks are not expected to be significantly different than those experienced/reported in adults.

3. STUDY OBJECTIVES

3.1. Primary Objectives

This is an open label phase 2 study. There is no formal hypothesis testing.

Safety:

- To evaluate the safety and tolerability of the Humacyte HAV in vascular trauma patients undergoing surgery for vascular replacement or reconstruction

Efficacy:

- To determine the rate of primary patency at 30 days

3.2. Secondary Objectives

Safety:

- To determine mechanical stability of the HAV based on freedom from aneurysmal degeneration, anastomotic bleeding or spontaneous rupture, infection, or significant stenosis
- To determine HAV durability in terms of freedom from HAV removal or replacement

Efficacy:

- To determine the patency of the HAV (primary, primary assisted and secondary)
- To determine the rates of interventions needed to maintain / restore patency in the HAV
- To determine the rate of limb salvage (extremity injuries only)
- To determine patient survival
- To evaluate remodeling of HAV

4. STUDY DESIGN

4.1. Description of the Study Design

Prospective, multicenter, non-randomized phase 2 study.

Injuries to both the limbs and torso will be permitted in the study. Vessels of the heart are excluded. Example of size appropriate vessels include (but are not limited to):

- Axillary
- Brachial
- Basilic
- Popliteal
- the femoral vessels
- subclavian
- brachiocephalic/innominate
- celiac
- hepatic
- splenic
- superior mesenteric
- renal
- the iliac vessels

The DMC will review the earlier of when the first torso subject reaches 3 months post-implantation or when the first 2 torso subjects have both reached 30 days post-implantation.

4.2. Study Endpoints

Endpoints will be assessed for up to 36 months after HAV implantation. The primary analysis of the study will be conducted on the earlier of a) when the last subject enrolled reaches 30 days post-implant or b) all subjects enrolled in the initial 24 month accrual period have reached 30 days post-implant.

4.2.1. Primary Endpoints

Safety:

- Frequency and severity of adverse events

Efficacy:

- HAV primary patency at 30 days

4.2.2. Secondary Endpoints

Safety:

- Frequency of adverse events of special interest:
  - HAV occlusion (thrombosis)
  - HAV spontaneous rupture
  - Iatrogenic injuries are not an Event of Special Interest and should be reported as an AE
  - HAV infection
  - HAV abandonment
  - HAV aneurysm
  - HAV pseudoaneurysm
  - HAV Excision (partial or complete)

Efficacy:

- HAV primary patency
- HAV primary assisted patency
- HAV secondary patency
- Rate of HAV interventions
- Limb salvage (extremity injury only)
- Patient survival
- HAV remodeling as shown by histopathology of any clinical explants

4.3. Duration of Study Participation

The active study duration for each study participant will be 36 months from HAV implantation. All subjects will be followed for the initial 12 months. Beyond 12 months (Long-Term Follow Up), only subjects with a patent HAV will be followed out to a total of 36 months from HAV implantation. The total expected duration of the clinical study is 61 months.

5. STUDY POPULATION

5.1. Description of the Study Population

The study population will consist of patients with vascular trauma to size appropriate vessels in the limb or torso, requiring replacement or reconstruction.

5.1.1. Patient Inclusion Criteria

1. Patients with life or limb threatening traumatic injury to an arterial vessel in the limb or torso, other than the heart, which requires replacement or reconstruction
2. Preoperative imaging or clinical examination indicates the damaged vessel has a defect length of ≤ 38cm and is appropriately size matched to the 6mm Human Acellular Vessel (HAV) per the judgment of the treating surgeon taking into account vasoconstriction and situational inflow and outflow considerations.
3. Autologous vein graft is either not feasible in the judgment of the treating surgeon (e.g. because of lack of availability of suitable conduit, presence of severe venous insufficiency) or is not desirable because of the urgency of revascularization
4. Aged 14 to 85 years old, inclusive
5. Able to communicate meaningfully with investigative staff, and able to comply with entire study procedures. If the patient is unconscious, then information from a reliable witness indicates that the patient would normally be able to comply with study procedures
6. Patient or legal representative is able, willing and competent to give informed consent
7. Life expectancy of at least 1 year

5.1.2. Patient Exclusion Criteria

1. Mangled Extremity Severity Score (MESS) of ≥ 7
2. Limb at high risk of amputation despite vascular reconstruction (e.g., because of crush injury)
3. Catastrophic injuries that make survival unlikely (e.g. Abbreviated Injury Scale (AIS) > 5 or Injury Severity Score (ISS) >60)
4. HAV may not be used for coronary artery repair
5. Known pregnant women
6. Known medical condition which would preclude long term antiplatelet therapy after resolution of acute injuries
7. Any other condition which in the judgment of the investigator would preclude adequate evaluation of the safety and efficacy of the HAV
8. Previous exposure to HAV
9. Known participation in any investigational study within the last 30 days

10. Employees of the sponsor or patients who are employees or relatives of the investigator

5.1.3. Enrollment of Patients Aged 14 through 17

The study population includes patients 14 through 17 years of age, who are assessed to be Tanner Stage V by the health care provider. As in adults, vascular injuries can be treated with the HAV only if they occur to size appropriate vessels per the judgement of the treating surgeon.

6. INVESTIGATIONAL MEDICINAL PRODUCT

Additional information on the manufacturing process and testing of the investigational medicinal product (IMP)is provided in the Investigator Brochure.

6.1. Product Description

The IMP is a Humacyte Human Acellular Vessel (HAV), which is a tissue-engineered vascular prosthesis for vascular bypass or reconstruction in patients with peripheral vascular disease or peripheral vascular trauma. It is a sterile, non-pyrogenic acellular tubular vessel composed of human collagen types I and III and other extracellular matrix proteins, including fibronectin and vitronectin. The HAV is 6 mm in diameter and approximately 42 cm in length. The product is supplied on a silicone mandrel immersed in sterile phosphate buffered saline in a sealed and labeled plastic container.

There is no placebo or comparator control group in this study.

6.2. Manufacturer of the IMP –

The HAV is manufactured by:

Humacyte Global, Inc.

2525 NC Highway 54

Durham, NC 27713

Traceability of the HAV during and after the clinical investigation will be achieved by the assignment of lot numbers. A unique identifying lot number will be assigned to each vessel.

6.3. Packaging, Storage, and Labeling

Packaging: Each HAV is contained in a sealed, flexible plastic primary container closure system that was developed by Humacyte. The system meets container/closure requirements to maintain sterility as well as product and fluid integrity. The vessel is contained inside the system in a fixed manner, immersed in a sterile, phosphate buffered saline. The total volume of the storage solution is approximately 300 mL.

Storage: The product is shipped under controlled conditions to maintain temperature at 4˚C (range: 2 – 8˚C). The product should be stored in a refrigerator that maintains this temperature range. The HAV **MUST NOT** be allowed to freeze.

Labeling: The IMP will be labeled according to applicable guidelines and relevant regulatory agency requirements. A tamper resistant label affixed to the secondary container will be used to ensure that the product is not compromised prior to use.

6.4. Implantation of the Humacyte Human Acellular Vessel (HAV)

The Humacyte HAV is implanted using standard vascular surgical techniques similar to placement of predicate peripheral vascular prostheses (see study manual for details).

Tunneling of the HAV, if required, must be performed using a sheathed tunneler. After inserting the assembled tunneler into the tissue, the inner mandrel of the tunneler should be removed from the sheath. The sheath is lubricated with saline and then with the silicone mandrel in place, the HAV can be easily pushed through the sheath without the need to tie to the inner mandrel and pulled through the tunneler (see study manual for details).

After placement, HAV patency and integrity are checked by pressurizing the conduit. Prior to completion of surgery, HAV patency is confirmed by physical exam, Doppler, angiography (conventional or intra-op CT angiography) or ultrasound. The surgical site is closed using standard techniques.

Implantation of the HAV will be undertaken by qualified vascular surgeons experienced in peripheral vascular surgery.

6.5. IMP Accountability Procedures

Documentation of receipt, dispensing, and return of all IMP must be maintained by the Principal Investigator or his/her designee. It is the Principal Investigator's responsibility to ensure that all IMPs are kept in a secure location, with access limited to individuals authorized by the Investigator. The product will be shipped with the IMP Shipment Confirmation Form. Once signed, the form should be returned to Humacyte or authorized designee, and the original will be maintained in the Investigator's Files. The IMP Accountability Log will be used to account for all IMP received, dispensed, and returned and must be maintained by the site until the conclusion of the study. Following accountability of the IMP by Humacyte or their authorized designee, all unused IMP will be returned to Humacyte.

7. OTHER TREATMENTS AND MEDICATIONS

7.1. Prior and Concomitant Medications

Prior medications are defined as all prescription and over the counter (OTC) medications taken within 7 days (whether continuing or not) prior to Day 1. All prior and concomitant medications (including immediately pre-surgery and post-surgery medications) must be listed in the patient's medical record and recorded on the eCRF. Drugs used during anesthesia should be recorded in the anesthesia records but should not be transcribed into the eCRF. Patients should be questioned at each study visit concerning any new medications or changes in current medications. **Note: particular attention should be made to identify the use of antithrombotic or antiplatelet agents (e.g., aspirin, clopidogrel, prasugrel, direct thrombin inhibitors, factor Xa inhibitors, or vitamin K antagonists).**

For each medication taken, the following information will be collected:

- Medication generic name / components of combination product
- Dose
- Route of administration
- Frequency of administration
- Date started
- Date stopped
- Indication for use

7.2. Essential, Precautionary and Prohibited Medications

7.2.1. Essential Medications

All patients should receive both antibiotic and antithrombotic prophylaxis in conjunction with HAV implantation:

Antibiotic prophylaxis:

- All patients must have at least 1 day of antibiotic prophylaxis the same day as surgery in accordance with local hospital guidelines. Longer antibiotic prophylaxis is at the discretion of the investigator.

Antithrombotic prophylaxis:

- Intraoperative heparin: the doses of heparin to be used during surgery will be determined by the investigator.
- Further measures to prevent venous thromboembolism are at the discretion of the investigator and may include LMWH.

- If antiplatelet therapy was not ongoing at the time of surgery it should be commenced as soon as medically appropriate post operatively. Recommended antiplatelet therapy (aspirin 81-325 mg and/or clopidogrel 75 mg daily) is at the discretion of the investigator and should continue long term while the HAV is in place. If the patient is unable to tolerate aspirin and/or clopidogrel the choice of antiplatelet regimen is at the investigator's discretion.

7.2.2. Restricted Medications

Vitamin K antagonists, antiplatelet agents other than aspirin and clopidogrel, direct thrombin inhibitors and factor Xa inhibitors (e.g., dabigatran, apixaban and rivaroxaban) should be avoided unless essential for treatment of a medical condition arising postoperatively. In that case consideration should be given to modification or cessation of antiplatelet therapy. Antiplatelet therapy should be restarted on cessation of these anticoagulant drugs.

8. STUDY PROCEDURES / EVALUATIONS

8.1. Clinical Evaluations Through Month 12

- Medical History – pre-operatively, from patient / legal representative interview and medical records covering relevant past medical history.
- Smoking history
- Medication History – prescription and OTC medication from Day -7 onwards (see Section 6.7). Particular attention should be paid to the identification of OTC medications containing aspirin.
- Physical Exam – full exam (as far as possible) at pre-operative screening, 12 month visit or final study visit for early termination (ET). Clinical examination of the operative site and HAV at all post-operative visits; exam of distal vascular bed (extremity injury only); physical exam for lymphadenopathy; additional clinical exam as needed to evaluate adverse events
- Vital signs (heart rate, blood pressure and temperature) – at D5
- Blood samples for hematology, clinical chemistry at pre-operative screening and Day 5 and PRA at pre-operative screening, Day 30 and M6
- Pre-operative imaging (ultrasound or angiography) is at the discretion of the investigator based on the clinical condition of the patient. It may be omitted if not deemed necessary in order to proceed to vascular surgical repair
- Adverse events – post-operatively on Day 1 and at all post-operative visits, the patient will be asked a general question about his/her health and for any HAV problems since the previous visit
- Intraoperative HAV bypass or interposition repair exam to assess anastomotic anatomy, patency and runoff. This may include physical exam, Doppler exam, angiography (conventional or intra-op CT angiography) or ultrasound at the investigator's discretion.
- Duplex ultrasound – clinical assessment at all postoperative visits from day 30 thru M12, to assess HAV patency, mid HAV diameter and stenosis. The full length of the HAV should be imaged at each assessment to monitor for aneurysm development
- Documentation of HAV interventions, surgical procedures and any complications immediately postoperatively through Month 12

8.2. Clinical Evaluations in Long Term Follow Up (Post Month 12 to Month 36)

- The status of the patient and HAV will be ascertained every 3 months from post Month 12 until 36 months after HAV implantation by telephone contact with the patient and/or

his physician. If a suspected SAE related to HAV is discovered an unscheduled visit should be conducted to investigate.

- Only related SAEs and all AESI will be reported.
- Visits at Month 24 and Month 36 are to be conducted in person with a physical exam of the HAV site and duplex ultrasound imaging of the HAV.

8.3. Laboratory Evaluations

8.3.1. Clinical and Research Laboratory Evaluations and Specimen Collection

The following parameters will be measured wherever possible at pre-operative screening and all should be measured at Day 5

- Hematology: hemoglobin, hematocrit, RBC, white blood cells (WBC) with differential, platelet count
- Clinical chemistry: sodium, potassium, calcium, blood urea nitrogen, creatinine, albumin, total bilirubin, glucose (non-fasting)
- PRA will be measured at pre-operative screening, Day 30, and Month 6.

All laboratory tests (except assay of PRA) will be conducted at certified hospital laboratories. Routine monitoring, maintenance or calibration of laboratory equipment is required per local site procedures. Samples for PRA analysis will be shipped to LabConnect for storage until they are sent for analysis at a central laboratory. Details concerning sample collection and processing can be found in the Study Manual.

8.4. Imaging Evaluations

8.4.1. CT Angiography and Conventional Angiography

CT angiography or conventional angiography will be conducted as pre-operative screening when feasible. Pre-operative imaging is at the discretion of the investigator based on the clinical condition of the patient. It may be omitted if not deemed necessary in order to proceed to vascular surgical repair.

8.4.2. Duplex Ultrasound

Duplex ultrasound examinations will be performed at Day 30, 3, 6, 9,12, 24, and 36 months and follow standard bypass graft imaging protocols, including B-mode, power Doppler and color duplex ultrasound imaging of the HAV with velocity spectral waveform analysis. The purpose of this duplex ultrasound surveillance is to detect HAV stenosis and aneurysm development. An alternative imaging method (e.g. CTA, MRI, etc.) may be substituted for duplex ultrasound at the discretion of the investigator if it is medically appropriate and in the best interest of the patient.

Determination of intraoperative HAV patency on Day 1 is required by physical examination, Doppler exam, angiography (conventional or intra-op CT angiography), or ultrasound at the discretion of the investigator.

8.5. Study Schedule

8.5.1. Pre-operative Screening (Day 1)

Potential study participants who are being considered for surgical repair of vascular injury appropriate for inclusion into the study will be informed about the study and invited to participate. After explanation of the potential risks and benefits of the HAV and of the study procedures, written informed consent will be obtained. No study specific procedures may be performed prior to patient consent. If the patient is unable to give informed consent then this may be sought from the patient's legal representative, usually a close relative. Standard of care procedures such as laboratory evaluations conducted prior to screening may be used rather than repeating the test.

The following assessments will be performed, as far as possible, prior to surgery (Day 1):

- Informed consent
- Medical history
- Prior and concomitant medication
- Full physical examination
- Evaluation of inclusion/exclusion criteria
- Reasons for not using an autologous venous conduit
- Laboratory testing (or standard pre-op lab profile for the institution)
    - Hematology: full blood count and differential
    - Clinical chemistry ; sodium, potassium, calcium, blood urea nitrogen, creatinine, albumin, total bilirubin, glucose (non-fasting)
    - PRA
- Ultrasound or CT angiography (CTA) (pre-op imaging is at the discretion of the investigator based on the clinical condition of the patient. It may be omitted if not deemed necessary in order to proceed to vascular surgical repair

8.5.2. Enrollment – Day 1 (HAV Implantation)

The HAV will be implanted as an interposition replacement or bypass in the required location using standard vascular surgical techniques. Details of the surgical anatomy and any complications will be documented. Operative procedures note and surgical diagram will be uploaded into the EDC for medical monitor review. Determination of intraoperative HAV patency

is required by physical examination, Doppler exam, angiography (conventional or intra-op CT angiography) , or ultrasound at the discretion of the investigator.

8.5.3. Follow-up Visits Day 5 through Month 12

Day 5 (or prior to hospital discharge if earlier)

- Concomitant Medication
- Physical exam including surgical site
- Vital signs (heart rate, blood pressure and temperature)
- Documentation of any HAV interventions
- Adverse events
- Laboratory assessments (clinical chemistry, hematology)

Day 30 (+ 5 days)

- Concomitant medication
- Physical exam including surgical site
- Duplex ultrasound of the HAV
- Documentation of any HAV interventions
- Adverse events
- PRA

Months 3, 6 and 9 (+/- 14 days)

- Concomitant Medication
- Physical exam including surgical site
- Duplex ultrasound of the HAV
- Documentation of any HAV interventions
- Adverse events
- PRA (Month 6 only)

Month 12 (+/-14 days) and Early Termination

- Concomitant Medication
- Full physical exam including surgical site
- Duplex ultrasound of the HAV

- Documentation of any HAV interventions
- Adverse events
- PRA (only at early termination if prior to 6 month PRA collection)
- CT angiography

8.5.4. Long Term Follow-up Post Month 12 through Month 36 (+/- 30 days)

The status of the patient and HAV will be ascertained every 3 months from post Month 12 through 36 months after HAV implantation.

- Quarterly questionnaire covering status of the patient via a telephone contact with the patient and/or physician. If a suspected SAE related to HAV is discovered an unscheduled visit should be conducted to investigate.
- Documentation of any HAV interventions
- Adverse events (all AESI and related SAEs to be reported)
- Physical exam including surgical site at Month 24 and Month 36
- Duplex ultrasound at Month 24 and Month 36

8.5.5. Early Termination Visit

The subject may withdraw from the study at any time at his/her own discretion. The treating physician may also withdraw the subject for safety reasons. If withdrawal occurs before 12 months the subject will be asked to complete an early termination visit at which all assessments normally performed at 12 months will be completed. PRA will be collected at ET visit if the visit occurs before Month 6 collection of the sample. If withdrawal occurs after Month 12 and prior to Month 36 the patient will be asked to complete an early termination visit at which all assessments normally conducted during the long term follow up visits will be completed.

The reasons for early termination should be recorded in the eCRF.

With the exception of patient withdraw, all subjects will be followed for 12 months from HAV implantation (or until HAV removal or death if earlier). The subject should be withdrawn from the study if the HAV is completely removed or the HAV becomes permanently occluded (loss of secondary patency) after Month 12.

8.5.6. Unscheduled Visits

If necessary to evaluate adverse events or HAV complications additional visits may be scheduled at the discretion of the investigator. At a minimum HAV status on clinical examination and Duplex ultrasound and adverse events will be recorded.

If, at any of the scheduled visits, duplex ultrasound surveillance suggests the development of a ≥ 50% stenosis within the HAV but immediate intervention is not required closer follow up should be considered. Intervention to manage any such stenosis is at the discretion of the investigator taking into account the degree and rate of progression of the stenosis.

8.6. Medical Care during the Study and upon Study Termination

Optimal medical therapy should be continued during the study. This should include:

- Antiplatelet therapy (see section 7.2.1)

After the final study visit at month 36 patients will not receive any further study-specific treatment. They will be treated by their medical doctor in a way that is appropriate for them.

8.7. Histological Examination of Resected HAV Material

If all or part of the HAV is resected it should, wherever possible, be retained for future histological examination. Instructions for preservation, storage and shipping of this material will be provided separately in a procedures manual. If a patient dies with an HAV in situ and it is feasible to obtain a fresh post mortem sample of the bypass this should be attempted in accordance with local regulations.

9. SAFETY ASSESSMENTS AND ADVERSE EVENTS

Safety of the HAV will be assessed in terms of:

- Aneurysm formation
- Pseudoaneurysm formation
- Anastomotic bleeding or spontaneous rupture
- HAV infection
- Need for HAV removal
- Inflammation at the implantation site
- Other adverse events
- Increase from baseline in PRA

9.1. Adverse Event Definition

An AE is any untoward medical occurrence in a patient administered an IMP and which does not necessarily have a causal relationship with the IMP. An AE can, therefore, be any unfavorable and unintended sign, symptom, or disease temporally associated with the use of an IMP, whether or not related to the IMP. Any worsening of the patient's disease under study or other medical conditions will also be considered to be an AE, unless it is within the normal range of disease fluctuation for that patient.

9.2. Serious Adverse Event Definition

An AE is considered "serious" if, in the view of either the investigator or Sponsor, it:

- Results in death
- Is life-threatening
    - The term "life-threatening" in the definition of "serious" refers to an event in which the patient was at immediate risk of death at the time of the event; it does not include an adverse event that had it occurred in a more severe form, might have caused death.
- Requires patient hospitalization or prolongation of existing hospitalization
    - This is defined as the patient being hospitalized for 24 hours or more or the patient's hospital stay being prolonged for at least an additional overnight stay.
- Requires intervention to prevent permanent damage
- Results in persistent or significant disability/incapacity
- Is a congenital anomaly/birth defect

- Important Medical Events
    - For the purpose of this study, this includes any event involving the HAV that results in a surgical or endovascular radiological intervention. The event(s) which caused the procedure should be reported as an SAE. For example: in the event of HAV thrombosis, the thrombosis would be considered the SAE; any associated stenoses (or other associated findings) that are present would be considered AEs.

Medical and scientific judgment should be exercised in deciding whether expedited reporting to the sponsor is appropriate in other situations, such as important medical events that may not be immediately life-threatening or result in death or hospitalization but may jeopardize the patient or may require medical or surgical intervention to prevent one of the other outcomes listed in the definition above. These should also be considered to be SAEs.

Note: Hospitalization for the surgery to implant the HAV is not a SAE. However, prolongation of the initial hospitalization due to an AE will be considered a SAE.

9.3. Suspected Unexpected Serious Adverse Reaction

A suspected unexpected serious adverse reaction (SUSAR) is any adverse drug reaction that is serious (as defined in 9.2), unexpected (is not listed in the IB or is not listed at the specificity or severity that has been observed) and suspected (meaning there is a reasonable possibility that the IMP caused the adverse event).

9.4. Events of Special Interest

Events of Special Interest are:

- HAV occlusion (thrombosis)
- HAV spontaneous rupture
    - Iatrogenic injuries are not an Event of Special Interest and should be reported as an AE
- HAV infection
- HAV abandonment
- HAV aneurysm
- HAV pseudoaneurysm
- HAV Excision (partial or complete)

9.5. Reporting of Adverse Events

At each evaluation, the investigator will determine whether any AEs have occurred. The patient will be questioned in a general way and no specific symptoms will be suggested. If any AEs have

occurred they should be documented in the patient's medical chart and recorded on the AE pages of the eCRF. If known, the diagnosis should be recorded in preference to the listing of individual signs and symptoms. All SAEs, should be reported to the Safety CRO within 24 hours from the time the investigator or study personnel first become aware of the event.

AE reporting begins from time of anesthesia induction for implantation of the HAV and ends at the conclusion of the Month 12 visit or ET visit, unless an unresolved AE is still being followed.

During the long term follow up period from post Month 12 through Month 36, only the following will be reported by the investigator:

- All SAEs considered related to the HAV
- All Events of Special Interest (Section 9.4)

9.5.1. Criteria for Determining Causal Relationship to the HAV and Criteria for Determining Causal Relationship to the Index Surgical Procedure

The criteria for determining the causal relationship of an AE with the HAV are presented in the table below. A separate assessment of causal relationship of an AE to the index surgical procedure is required as well using the same criteria and definitions presented in the table below. Please note that causal relationship to procedure only refers to the index surgical procedure in which the HAV was initially implanted.

Causal Relationship to the IP	**Criteria for Determining Causal Relationship**
Definitely Related	There is clear evidence to suggest a causal relationship, and other possible contributing factors can be ruled out. The clinical event, including an abnormal laboratory test result, occurs in a plausible time relationship to surgical placement of the HAV and cannot be explained by concurrent disease or other devices, drugs, or chemicals.
Possibly Related	There is some evidence to suggest a causal relationship (e.g., the event occurred within a reasonable time after the surgical placement of the HAV). However, the influence of other factors may have contributed to the event (e.g., the subject's clinical condition, other concomitant medications). Although an adverse event may rate only as "possible" soon after discovery, it can be flagged as requiring more information and later be upgraded to Definitely Related as appropriate.
Unlikely Related	A clinical event, including an abnormal laboratory test result, whose temporal relationship makes a causal relationship improbable (e.g., the event did not occur within a reasonable time after surgical placement of

	the HAV) and in which other drugs or chemicals or underlying disease provide plausible explanations (e.g., the subject's clinical condition, other concomitant treatments).
Not Related	A clinical event, including an abnormal laboratory test result, which occurs when the HAV was not implanted; or, another cause is obvious and in which there is sufficient information that the etiology of the event is not related to the HAV.

The sponsor will make the final determination of causality for the purposes of reporting to the regulatory authorities and to the Principal Investigators.

9.5.2. Criteria for Defining the Severity of an Adverse Event

Severity of adverse events, including abnormal clinical laboratory values, will be assessed according to the criteria below and entered in the eCRF:

Grade	**Severity Assessment Standard**
1-Mild	Events require minimal or no treatment and do not interfere with the subject's daily activities.
2-Moderate	Events result in a low level of inconvenience or concern with the therapeutic measures. Moderate events may cause some interference with functioning.
3-Severe	Events interrupt a subject's usual daily activity and may require systemic drug therapy or other treatment. Severe events are usually incapacitating.
4-Life-threatening	Any adverse event that places the subject or participant, in the view of the investigator, at immediate risk of death from the reaction as it occurred, i.e., it does not include a reaction that, had it occurred in a more severe form, might have caused death.
5-Death	Death related to AE.

9.5.3. Reporting of Action Taken to Resolve AE

- None
- Lab tests / further evaluation
- Treatment required (specify if hospitalized)

- Patient withdrawn from study
- Other (specify)

9.5.4. Reporting the Outcome of the AE

- Recovered, with sequelae
- Recovered, without sequelae
- Ongoing
- Death
- Lost to follow-up

9.5.5. Reporting Serious Adverse Events

The urgency for reporting SAEs is 4-fold: (1) to facilitate discussion (and implementation, if necessary) by the sponsor and the investigator of appropriate follow-up measures, (2) to facilitate investigator reporting of unanticipated problems involving risk to human subjects to the institutional review board (IRB), (3) to facilitate the sponsor's rapid dissemination of information regarding AEs to other investigators/sites in a multi-center study, and (4) to enable the sponsor to fulfill the reporting requirements to the appropriate regulatory authority.

Any SAE that occurs through Month 12, whether or not causally related to the IMP, must be reported by the investigator or designee to the Safety CRO within 24 hours of learning of its occurrence. This applies also to any AE that could affect the safety of the study participants or the conduct of the study. Any SAE that occurs during long term follow up post Month 12 through Month 36 that is causally related to the IMP, must be reported by the investigator or designee to the Safety CRO within 24 hours of learning of its occurrence.

Information about an SAE will be collected and recorded on the SAE Report Form. The investigator must assess the relationship to the investigational product and any relevant procedure.

The investigators must complete the SAE Report Form in English, and **send the completed, signed form by fax or email (see below) IMMEDIATELY (at latest within 24 hours) after becoming aware of the SAE.**

Copies of relevant de-identified medical records (e.g., admission and/or discharge summary, laboratory reports and autopsy report), may also be submitted with the SAE form to clarify the circumstances surrounding the SAE(s). The entire medical records should **NOT** be sent with the SAE form.

Atlantic Research Group, Inc.

Drug Safety Department

Fax: +1-866-851-9318

Email: Safety@atlanticresearchgroup.com

Phone: +1-888-619-3216

The investigator will be requested to supply as much detailed information as possible regarding the SAE that is available at the time of the initial contact. The investigator should also complete missing or requested information and submit follow-up reports until the SAE has resolved or, in the case of permanent impairment, until the SAE has stabilized.

It is the responsibility of each Principal Investigator to promptly notify his/her IRB of all SAEs that are received by the Sponsor or designee and that occur at his/her institution in accordance with institutional practices.

The Safety CRO will inform the sponsor about all SAEs within 1 business day after receipt of the respective report from the investigator.

9.5.6. Reporting of Events of Special Interest

Events of Special Interest are defined in Section 9.4 and should be reported to the Safety CRO within 24 hours of learning of its occurrence. For each of these events detailed surgical notes (with illustrative diagram), including reason for and outcome of any intervention or abandonment, should be completed within 48 hours and uploaded to the clinical database.

Detailed information about the occurrence and treatment/intervention for these events will be collected throughout the study up to 3 years post HAV implant. This information will include the following:

- Summarized surgical notes, including a simplified anatomical diagram showing where angioplasties, stents, or revisions have been performed (using intervention worksheet provided)
- Need for hospitalization (number of nights)
- Need for antibiotics (in the case of HAV-related infections)

9.5.7. Follow-Up of Adverse Events

If any AEs are present when a subject completes 1 year post implant (Month 12) or ET, if earlier, or if a subject is withdrawn from the study, the subject will be re-evaluated within an appropriate period of time. At the investigator's discretion, minor AEs can be re-evaluated via telephone and documented. If the AE has still not resolved, additional follow-up will be performed as appropriate. The investigator or his designee should make every effort to contact the subject until the AE has

resolved or stabilized or the medical monitor and investigator agree that further follow-up is not necessary. This should be documented in the subject's medical records.

9.6. Reporting of Pregnancy

If a study participant becomes pregnant during study participation, basic information about the pregnancy will be recorded in the Pregnancy eCRF and the Pregnancy Outcome and Report Form, and submitted to the Safety CRO. If there are complications during the pregnancy, the complications are recorded as AEs. The participant will be asked to report the outcome of the pregnancy and the site should submit the information to the Safety CRO within 30 days after the outcome of the pregnancy. If there is a congenital anomaly in the infant, this will be recorded as a SAE in the data forms for the mother (i.e., the study participant).

Partner pregnancies do not need to be reported.

9.7. Data Monitoring Committee

A Data Monitoring Committee (DMC) will review safety on an ongoing basis and provide recommendations about stopping, continuing or otherwise modifying the study. The DMC consists of individuals who are not directly involved in the conduct of the study. A charter describes the roles and responsibilities of the DMC. Responsibilities of the DMC will include review of aggregate safety data from other studies in the HAV clinical development program.

The DMC will at a minimum meet every 6 months from the date of initial enrollment of the first subject. Additionally, the DMC will review the safety data of the torso injuries on the earlier of when the first torso subject reaches 3 months post-implantation or when the first 2 torso subjects have both reached 30 days post-implantation.

9.8. Interim Analysis and Stopping Criteria

This is a phase 2 study with no formal interim analysis. Periodic reviews of safety data will be undertaken by the DMC with particular attention to events that might indicate structural failure of the HAV. Events that might have implications for already implanted HAVs and their possible removal - such as aneurysm formation (true or pseudo) or spontaneous rupture -would trigger an urgent review of the safety data for DMC review.

The DMC may recommend modification or early termination of the study for safety reasons.

10. STATISTICAL CONSIDERATIONS

This is a prospective, open label, multicenter pilot study to evaluate the safety and efficacy of the HAV in patients undergoing vascular replacement or reconstruction. The primary objective of this study is to evaluate the safety and tolerability of the HAV in these patients and to determine the primary patency of the HAV at 30 days post-implantation. The secondary objectives of this study are to further assess safety in terms of adverse events of special interest, to determine the rate interventions required to keep the HAV patent, and to further characterize efficacy in terms of secondary patency and limb salvage. There is no formal hypothesis testing planned.

Endpoints will be assessed over a period of up to 36 months after HAV implantation. The primary analysis of the study will be conducted on the earlier of a) when the last subject enrolled reaches 30 days post-implant or b) all subjects enrolled in the initial 24 month accrual period have reached 30 days post-implant. Details of data handling and planned descriptive statistics are given in the Statistical Analysis Plan.

10.1. Analysis Population

All patients who receive an HAV will be included in the analyses. For discontinued or withdrawn patients, all available data will be included in the safety and efficacy analyses.

10.2. Safety Analyses

Safety analyses will be performed on all patients who have an HAV implanted.

The incidence of aneurysm formation, anastomotic bleeding or spontaneous rupture, HAV removal, HAV infection, and inflammation at the implantation site will be tabulated by visit and overall.

Adverse events will be coded using Medical Dictionary for Regulatory Activities (MedDRA) terms. Adverse events will be listed and summarized by body system, incidence, severity, and duration. HAV complications will be listed in terms of incidence, severity, and (where appropriate) time to onset and duration. Serious adverse events will be summarized separately. Any premature discontinuations due to adverse events and deaths will be listed and summarized.

PRA data will be listed and summarized using appropriate descriptive statistics for the change from baseline values.

10.3. Efficacy Analyses

The primary efficacy analysis will be the rate of primary patency at 30 days after HAV implantation. Primary, primary assisted, and secondary patency rates of the HAV at 12 months and at all other post-surgery visits with evaluation of patency will be described. The rate of limb salvage and patient survival at 12 months will also be described.

Primary patency is defined as the functional access patency until any type of intervention; primary assisted patency is defined as an HAV still working without thrombosis; secondary patency is

defined as the functional HAV patency, with or without preceding successful interventional or surgical procedures to maintain or reestablish patency, until the HAV is abandoned. Early discontinued patients prior to the visit of interest will be determined as being non-patent irrespective of the reason for discontinuation.

The rate and type of interventions needed to maintain / restore patency in the HAV will be descriptively tabulated.

The absolute change from baseline (Day 1) values to all post-surgery visits of duplex ultrasound parameters will be summarized. Summary statistics will also be provided at each time point.

The methods and endpoints regarding the efficacy parameters employed in this study are consistent with current clinical practice and are meaningful to the research community. Every attempt has been made to minimize the variability on the part of the surgeon when using this product.

10.4. Other Analyses

All clinical parameters will be listed for all patients treated at each study visit. Descriptive statistics will be summarized for continuous outcomes such as age and BMI. If necessary, number and percentage of patients will be reported for categorical outcomes.

10.5. Sample Size Rationale

Up to 100 subjects will be recruited into the study. As this phase 2 study is the first human study of the HAV for vascular trauma, the study was designed to provide preliminary evidence of safety and efficacy.

10.6. Interim analyses

There is no formal interim analysis.

11. STUDY MANAGEMENT AND DATA COLLECTION

11.1. Ethical Conduct of the Trial

This study will be conducted according to the protocol; 21 CFR Parts 11, 50, 54, 56, and 312; the World Medical Association Declaration of Helsinki and Good Clinical Practice (GCP). Each Investigator will conduct the trial according to applicable local or regional regulatory requirements.

11.2. Institutional Review Board

IRBs must be constituted according to the applicable state and federal requirements, including ICH GCP.

It is the responsibility of each investigator to submit the protocol, Investigator's Brochure, subject informed consent, subject recruitment materials (if applicable), and other documentation as required by the IRB to his/her IRB for review and approval. A copy of the written approval must be provided to the contract research organization (CRO). The documentation should clearly mention the approval/favorable opinion of the protocol, the subject informed consent form, and subject recruitment materials (if applicable), including respective version dates. The written approval and a list of members, their titles or occupations, and their institutional affiliations may be obtained from the IRBs if available, and provided to the CRO prior to the release of clinical study supplies to the investigational site and commencement of the study. If any member of the IRB has direct participation in this trial, written notification regarding his or her abstinence from voting must also be obtained.

Each investigator must adhere to all requirements stipulated by his/her respective IRB. This includes notification to the IRB regarding protocol amendments, updates to the subject informed consent, recruitment materials intended for viewing by subjects, investigational new drug safety reports, SAEs and unexpected AEs, reports and updates regarding the ongoing review of the trial at intervals specified by the respective IRB, and submission of final study reports and summaries to the IRB.

11.3. Subject Informed Consent

Prior to any study procedures being performed, subjects and persons conducting the consent discussion will be required to sign and date the IRB-approved informed consent, and each subject will be given a copy. In addition, this information should be recorded in the subject's medical record (i.e., source document). If the patient is unable to give informed consent then this may be sought from the patient's legal representative, usually a close relative.

The written consent document will embody the elements of informed consent as described in the World Medical Association Declaration of Helsinki, 21 CFR Part 50.25, ICH E6 guideline (GCP), and in accordance with any local regulations. The investigator is responsible for the preparation, content, and IRB approval of the informed consent document. The consent form must be approved by the site's IRB and be acceptable to Humacyte.

The consent form must be written in a language fully comprehensible to the prospective subject. The investigator or designee shall give the subject adequate opportunity to read it before it is signed and dated. Information should be given in both oral and written form whenever possible and in the manner deemed appropriate by the IRB. Subjects must be given ample opportunity to inquire about details of the study.

11.3.1. Informed Consent for Pediatrics

Pediatric enrollment (ages 14 through 17) requires obtaining informed consent from the minor's legally authorized representative (parental consent or surrogate consent).

11.4. Amendments to the Protocol

An amendment must be agreed to in writing by Humacyte and submitted to the FDA and approved by the IRBs before the amendment can be implemented. Written approval of a protocol amendment is not required prior to implementation of changes to the protocol which eliminate an immediate hazard to the study patient; however, approval must be obtained as soon as possible thereafter. Any agreed amendments must also be signed by the investigator.

11.5. Study Initiation

The investigator must not enroll any patients prior to attendance at the Investigator Meeting or the completion of a formal site initiation visit conducted by the CRO. These meetings will include a detailed review of the study protocol and eCRF pages. The investigator will not be supplied with IMP until all necessary pre-study requirements have been completed and essential signed documents provided to the CRO.

11.6. Study Monitoring

It is the responsibility of the investigator to ensure that the study is conducted in accordance with the protocol, GCP, applicable regulatory requirements, and the currently approved Declaration of Helsinki, and that valid data are entered into the eCRF.

To achieve this objective, the monitor's duties are to ensure the maintenance of complete, legible, well-organized, and easily retrievable data. The monitor will review the protocol with the investigator. In addition, the monitor will explain the investigator's reporting responsibilities and all applicable regulations concerning the clinical evaluation of the IMP.

The investigator will permit representatives of Humacyte and the CRO to monitor the study as frequently as Humacyte or the CRO deem necessary to determine that data recording and protocol adherence are satisfactory. The eCRF data and related source documents will be reviewed in detail by the monitor at each visit, in accordance with relevant SOPs and ICH GCP regulations. This includes results of tests performed as a requirement for participation in this study and any other medical records required to confirm information contained in the eCRF such as

past medical history and secondary diagnoses. The investigator and his/her staff will be expected to cooperate with the monitor and provide any missing information whenever possible.

All monitoring activities will be reported and archived. In addition, monitoring visits will be documented at the investigational site by signature and date on the study-specific monitoring log.

11.7. Case Report Form

An electronic CRF will be used for this study. The data will be entered into the eCRF in a timely manner on an ongoing basis.

The investigator is responsible for ensuring that data are properly recorded on each patient's eCRF and related documents. An investigator who has signed the protocol signature page should personally sign the eCRFs in accordance with the procedure described in the eCRF completion guidelines to ensure that the observations and findings are correct and complete.

For data handled by the CRO, eCRF data and some or all of the study-related data will be managed and stored electronically in the CRO's database system. Validated data will subsequently be transferred to the sponsor.

11.8. Verification Procedures

It is the investigator's obligation to ensure documentation of all relevant data in the subject's medical record. The subject's medical record will be considered the source document. The eCRF should not be used as the source for study information.

The investigator will maintain a subject identification code list to enable unambiguous identification of the subjects (subject names and corresponding subject numbers). The subject identification code list is an essential document and as such should be maintained according to the ICH GCP guidelines.

11.9. Retention of Records

All documentation pertaining to the study will be kept by Humacyte or their designee in accordance with ICH guidelines and US FDA regulations.

The investigator will maintain a study file, which should be used to file the Investigator's Brochure, protocol, and IMP records; correspondence with the IRB and Humacyte; and other study-related documents.

The investigator agrees to keep records and those documents that include (but are not limited to) the identification of all participating subjects, medical records, study-specific source documents, source worksheets, all original signed and dated informed consent forms, query responses, and detailed records of drug disposition to enable evaluations or audits from regulatory authorities and Humacyte or its designees.

The investigator shall retain records required to be maintained under this part for a period of 2 years following the date a marketing application is approved for the IMP for the indication for

which it is being investigated; or, if no application is to be filed or if the application is not approved for such indication, until 5 years after the investigation is discontinued. However, these documents should be retained for a longer period if required by the applicable regulatory requirement(s) or if needed by the sponsor. In addition, the investigator must make provision for the subject's medical records to be kept for the same period of time. No data should be destroyed without the agreement of Humacyte. Humacyte will inform the investigator in writing when the trial-related records are no longer needed. Subject's medical records and other original data will be archived in accordance with the archiving regulations or facilities of the study site.

11.10. Protocol Deviations

A protocol deviation is any noncompliance with the protocol or GCP requirements. The noncompliance may be either on the part of the participant, the investigator, or the study site staff. As a result of deviations, corrective actions are to be developed by the site and implemented promptly. Although in principle, protocol deviations are not permitted, under emergency circumstances, deviations may proceed without prior approval of the sponsor and the IRB to protect the rights, safety, and well-being of human subjects.

All protocol deviations will be documented and reported by the CRO during the course of the study in the Monitoring Reports. All deviations will be reported to the sponsor who will agree on the necessary actions to be taken.

If required per their guidelines, reports about protocol deviations must be reported to the local IRB.

11.11. Insurance and Indemnity

Insurance coverage for damages emerging from the study will be provided according to applicable legal requirements. During the informed consent procedure, the investigator must inform the patient accordingly.

11.12. Audit

It is the responsibility of CRO and Humacyte to perform auditing (if applicable) as part of implementing quality assurance. The purpose of an audit, which is independent of and separate from routine monitoring or quality control functions, is to evaluate trial conduct and compliance with the protocol, SOPs, GCPs, and the applicable regulatory requirements. The auditor and regulatory authorities will require authority from the investigator to have direct access to the subject's medical records.

12. REPORTING

Following completion of follow-up of all patients to the 12-month endpoint, the results will be evaluated by Humacyte or a designee for clinically meaningful findings. A clinical study report will be generated, including a summary of all available data, statistical measures, tabulated results, graphical results and interpretations. This report will be submitted to regulatory authorities in a timely manner. An addendum to the report will be generated to include data up to 36 months follow-up. This addendum will be submitted to regulatory authorities in a timely manner.

13. QUALITY CONTROL AND QUALITY ASSURANCE

Following written standard operating procedures, the monitors will verify that the clinical trial is conducted and data are generated, documented (recorded), and reported in compliance with the protocol, GCP, and the applicable regulatory requirements. Reports of monitoring activities will be submitted to Humacyte in a timely manner.

The investigational site will provide direct access to all trial related areas, source data/documents, and reports for the purpose of monitoring and auditing by the sponsor, and inspection by local and regulatory authorities.

Quality control procedures will be implemented for data entry and the generation of data quality control checks and will be run on the database. Any missing data or data anomalies will be communicated to the site(s) for clarification/resolution.

14. RESPONSIBILITIES OF THE PRINCIPAL INVESTIGATOR

The role of the principal investigator is to implement and manage the day-to-day conduct of the clinical investigation as well as to ensure data integrity and the rights, safety, and well-being of the patients involved in the clinical investigation.

14.1. Informed Consent

The principal investigator shall ensure that the process for obtaining informed consent

- includes all aspects of the clinical investigation that are relevant to the patient's decision to participate throughout the clinical investigation,
- avoids any coercion or undue improper influence on, or inducement of, the patient to participate,
- does not waive or appear to waive the patient's legal rights,
- uses native non-technical language that is understandable to the patient,
- provides ample time for the patient to read and understand the informed consent form and to consider participation in the clinical investigation,
- provides the patient with a copy of the signed and dated informed consent form and any other written information.

The principal investigator shall ensure and document appropriate training if an authorized designee is appointed to conduct the informed consent process.

14.2. Compliance with the Protocol

The principal investigator shall:

- indicate his/her acceptance of the protocol in writing
- conduct the clinical investigation in compliance with the protocol
- create and maintain source documents throughout the clinical investigation and make them available as requested during monitoring visits or audits
- ensure that the IMP is used solely by authorized users, and in accordance with the protocol and instructions for use
- propose to the sponsor any appropriate modification(s) of the protocol
- refrain from implementing any modifications to the protocol without agreement from the sponsor, IRB, and, if required, regulatory authorities
- document and explain any deviation from the approved protocol that occurred during the course of the clinical investigation

- ensure that an adequate investigation site team and facilities exist and are maintained and documented during the clinical investigation
- ensure that maintenance and calibration of the equipment relevant for the assessment of the clinical investigation is appropriately performed and documented, where applicable
- ensure the accuracy, completeness, legibility, and timeliness of the data reported to the sponsor in the eCRFs and in all required reports
- maintain the clinical trial material accountability records
- allow and support the sponsor to perform monitoring and auditing activities
- be accessible to the monitor and respond to questions during monitoring visits
- allow and support regulatory authorities and the IRB when performing auditing activities
- ensure that all clinical-investigation-related records are retained as specified in this protocol.

14.3. Medical Care of Patients

The principal investigator shall:

- provide adequate medical care to a patient during and after a patient's participation in a clinical investigation in the case of AEs
- inform the patient of the nature and possible cause of any adverse events experienced
- inform the patient of any new significant findings occurring during the clinical investigation, including the need for additional medical care that may be required
- provide the patient with well-defined procedures for possible emergency situations related to the clinical investigation, and make the necessary arrangements for emergency treatment,
- ensure that clinical records are clearly marked to indicate that the patient is enrolled in a particular clinical investigation
- inform, with the patient’s approval or when required by national regulations, the patient's personal physician about the patient's participation in the clinical investigation
- make all reasonable efforts to ascertain the reason(s) for a patient's premature withdrawal from the clinical investigation while fully respecting the patient's rights.

14.4. Safety Reporting

The principal investigator shall:

- record every adverse event together with an assessment, in accordance with Section 9 of this protocol,
- report to the sponsor, without unjustified delay, all serious adverse events and medically significant events as specified in Section 9 of this protocol,
- supply the sponsor, upon sponsor's request, with any additional information related to the safety reporting of a particular event.

15. SUSPENSION OR PREMATURE TERMINATION OF THE CLINICAL INVESTIGATION

The sponsor may suspend or prematurely terminate either a clinical investigation in an individual investigation site or the entire clinical investigation for significant and documented reasons.

A principal investigator, IRB, or regulatory authority may suspend or prematurely terminate participation in a clinical investigation at the investigation sites for which they are responsible.

If suspicion of an unacceptable risk to patients arises during the clinical investigation, or when so instructed by the IRB or regulatory authorities, the sponsor shall suspend the clinical investigation while the risk is assessed. The sponsor shall terminate the clinical investigation if an unacceptable risk is confirmed.

The sponsor shall consider terminating or suspending the participation of a particular investigation site or investigator in the clinical investigation if monitoring or auditing identifies serious or repeated deviations on the part of an investigator.

If suspension or premature termination occurs, the terminating party shall justify its decision in writing and promptly inform the other parties with whom they are in direct communication.

If, for any reason, the sponsor suspends or prematurely terminates the investigation at an individual investigation site, the sponsor shall inform the responsible regulatory authority if required and ensure that the IRB is notified. If the suspension or premature termination was in the interest of safety, the sponsor shall inform all other principal investigators.

If suspension or premature termination occurs,

1. the sponsor shall remain responsible for providing resources to fulfill the obligations from the protocol and existing agreements for following up the patients enrolled in the clinical investigation, and
2. the principal investigator or authorized designee shall promptly inform the enrolled patients at his/her investigation site, if appropriate.

In the event that the study is discontinued, the reasons for discontinuation will be explained to the investigators and may be disclosed to the study participants. Humacyte will provide all information needed by the investigator to ensure the safety and well-being of the study participants.

16. PUBLICATION POLICY

A Publication Committee comprising the Principal Investigator from each participating site and a representative of Humacyte will oversee all publication of data from this study. Prior to submitting for publication, presenting, using for instructional purposes, or otherwise disclosing the results of the study, the investigator agrees to allow the Publication Committee and Humacyte a period of at least 30 days (or, for abstracts, at least 5 calendar days) to review the proposed publication or disclosure prior to its submission for publication or other disclosure. Publications or disclosures of study results shall not include other confidential information belonging to Humacyte. If the proposed publication/disclosure risks Humacyte's ability to patent any invention related to the study, the publication or disclosure will be modified or delayed, at the investigator's option, a sufficient time to allow Humacyte to seek patent protection of the invention. For multicenter studies, the first publication or disclosure shall be a complete, joint multicenter publication or disclosure. This statement does not give Humacyte any editorial rights over the content of a publication or disclosure, other than to restrict the disclosure of Humacyte's confidential information. If a written contract for the conduct of the study is executed which includes publication provisions inconsistent with this statement, then that contract's publication provisions shall apply rather than this statement.

17. REFERENCES

Akingba G, Rapp BM, Tsai A, Jester AL, Fajardo A, Motaganahali R, et al. Surgical Management of Vascular Trauma From Dog Bites: The 10- year Experience of a Level I Trauma Center. Journal of Vascular Surgery. 2012;56(4):1190.

Andrikopoulos V, Antoniou I, Panoussis P. Arterial injuries associated with lower-extremity fractures. Cardiovascular Surgery. 1995;3(1):15-8.

Cordesman AH, Lemieux, J. IED Metrics for Afghanistan. January 2004-May 2010. Center for Strategic & International Studies, 2010.

Dahl, SL, Kypson, AP, Lawson, JH et al. Readily available tissue-engineered vascular grafts. Sci Transl Med. 2011: 68ra9

Dixon BS, Beck GJ, Vazquez MA et al. Effect of dipyridamole plus aspirin on hemodialysis graft patency. N Engl J Med 2009;360:2191-201.

DuBose JJ, Savage SA, Fabian TC, Menaker J, Scalea T, Holcomb JB, Skarupa D, Poulin N, Chourliaras K, Inaba K, Rasmussen TE. The American Association for the Surgery of Trauma PROspective Observational Vascular Injury Treatment (PROOVIT) registry: multicenter data on modern vascular injury diagnosis, management, and outcomes. Journal of Trauma and Acute Care Surgery. 2015 Feb 1;78(2):215-23.

Faulconer ER, Branco BC, Loja MN, Grayson K, Sampson J, Fabian TC, Holcomb JB, Scalea T, Skarupa D, Inaba K, Poulin N. Use of open and endovascular surgical techniques to manage vascular injuries in the trauma setting: A review of the American Association for the Surgery of Trauma PROspective Observational Vascular Injury Trial registry. Journal of Trauma and Acute Care Surgery. 2018 Mar 1;84(3):411-7.

Helfet DL, Howey T, Sanders R, Johansen K. Limb Salvage Versus Amputation: Preliminary Results of the Mangled Extremity Severity Score. Clinical Orthopaedics & Related Research. 1990;256:80-6.

Holcomb J. Devastating Dismounted IED Injuries in OEF. Increasing amputation and genital injury rates admitted to LRMC 2009-2010. 2011. Presentation to the Defence Health Board, (http://www.health.mil/Reference-Center/Presentations/2011/03/08/Devastating-Dismounted-Improvised-Explosive-Device-Injuries-in-Operation-Enduring-Freedom)

Kelly BS, Heffelfinger SC, Whiting JF, et al. Aggressive venous neointimal hyperplasia in a pig model of arteriovenous graft stenosis. Kindey Int. 2002;62(6):2272-2280.

Kirkton RD, Prichard HL, Santiago-Maysonet M, Niklason LE, Lawson JH, Dahl SL. Susceptibility of ePTFE vascular grafts and bioengineered human acellular vessels to infection. Journal of Surgical Research. 2018 Jan 1;221:143-51.

L'Heureux N, Dusserre N, Konig G, et al. Human tissue-engineered blood vessels for adult arterial revascularization. Nat Med 2006; 12(3):361-365.

Mills JL et al on behalf of the Society for Vascular Surgery Lower Extremity Guidelines Committee. The Society for Vascular Surgery Lower Extremity Threatened Limb Classification System: Risk stratification based on Wound, Ischemia, and foot Infection (WIfI). J Vasc Surg 2013,

Morgan MBF, Crawford T, Murrin B and Fraser SCA. Developing the vascular quality of life questionnaire: a new disease specific quality of life measure for use in lower limb ischemia. J Vasc Surg 2001; 33: 679 - 687

Prichard, H.L, Manson, R.J.,Dibernado, L. et al. An early study on the mechanisms that allow tissue-engineered vascular grafts to resist vascular hyperplasia. J Cardiovasc Transl Res 2011, 4, 674-682

Quint C, Kondo Y, Manson RJ,et al. Decellularized tissue-engineering blood vessel as an arterial conduit. Proc Natl Acad Sci USA. 108(22):9214-9, 2011.

Rasmussen TE. Vascular Injury Rates in the Wars in Iraq and Afghanistan: How are they different from the past? United States Army Institute of Surgical Research, 2010.

Roy-Chaudhury P, Kelly BS, Miller MA, et al. Venous neointimal hyperplasia in polytetrafluoroethylene dialysis grafts. Kidney Int. 2001;59(6):2325-2334.

Rutherford RB1, Baker JD, Ernst C, et al Recommended standards for reports dealing with lower extremity ischemia: revised version. J Vasc Surg. 1997 Sep;26(3):517-38.

APPENDIX 1: HAV CLINICAL VISIT SCHEDULE

	Pre-op screening D1	**D 1**	**D 5** or prior to d/c	**D 30** + 5 days	**M3** ± 14 days	**M 6** ± 14 days	**M 9±** 14 days	**M12 / ET†** ± 14 days	**M15-M36** †+/- 30 days
Informed consent	X								
Medical history and nature of trauma	X								
Concomitant medication	X	X	X	X	X	X	X	X	
Physical exam [1]	X	X	X	X	X	X	X	X	X [7]
Pre-op Ultrasound or CT angiography [2]	X								
Vital signs			X						
Eligibility (inclusion/exclusion criteria)	X								
HAV implantation and intraoperative confirmation of patency [3]		X							
Documentation of surgery and any complications		X							
Clinical chemistry	X [5]		X						
Hematology	X [5]		X						
PRA	X			X		X		X [6]	
Duplex ultrasound [4]				X	X	X	X	X	X [7]
CT angiography								X	
AEs		X	X	X	X	X	X	X	X [8]

Documentation of HAV interventions		X	X	X	X	X	X	X	X[8]

Abbreviations: AEs, adverse events; D, day; d/c, discharge; ET, early termination; HAV, human acellular vessel; M, month

1. Physical examination includes clinical exam of the operative limb and HAV at all post-operative visits (incl. patency assessment on D1) and physical exam to evaluate AEs; include distal vascular bed (extremity injury only)
2. Pre-op imaging is at the discretion of the investigator based on the clinical condition of the patient. It may be omitted if not deemed necessary in order to proceed to vascular surgical repair
3. Determination of intraoperative HAV patency can be done by physical exam, Doppler, angiography or ultrasound at the discretion of the investigator
4. An alternative imaging method (CTA, MRI, etc.) may be substituted for duplex ultrasound at the discretion of the investigator if it is medically appropriate and in the best interest of the patient.
5. Measured at preoperative screening when possible
6. PRA only collected at ET visit if the visit occurs before Month 6 collection
7. Visits at month 24 and month 36 to be conducted in person with physical exam of the HAV site and duplex ultrasound imaging of HAV.
8. The status of the patient and HAV will be ascertained every 3 months post Month 12 until 36 months after HAV implantation by telephone contact with the patient and/or his physician. Only related SAEs and all AESI will be reported after 12 months. If a suspected SAE related to HAV is discovered an unscheduled visit should be conducted to investigate.

† Patients withdrawn before Month 12 will perform ET visit that correlates with the procedures at Month 12. Patients withdrawn after Month 12 and prior to Month 36 should complete an ET visit that correlates with procedures post Month 12 through Month 36.

SUMMARY OF PROTOCOL AND STATISTICAL ANALYSIS PLAN CHANGES

All changes since version 3.2 of protocol CLN-PRO-V005 incorporated in the current version (version 3.3) are briefly described below. Each change is also categorized as either related to safety or efficacy, changes to statistical plan, or administrative in nature. Please note that no change was made due to a safety concern associated with the trial or with the investigational product.

Changes Incorporated in Protocol Version 3.3 (12 August 2022)	
Change and Reason for Change	**Section(s) Changed**
Changes Related to Safety and/or Efficacy	
Change: Expanded the age limit to include pediatrics of 14 - 17 years of age who have reached Tanner Stage 5. Reason: To comply with FDA suggestion to include pediatrics of appropriate age.	Protocol Summary Inclusion/Exclusion Section 2.5.3
Change: Updated quantity of planned study sites from “up to 10 sites in the United States” to “Approximately 25 sites in US and Israel.” Reason: To enable faster enrollment and utilize international sites, which participated in prior clinical evaluations of the HAV.	Protocol Summary SAP Section 1.2
Change: Language describing the delineation of torso and extremity were modified. Reason: The delineation between torso and extremity does not impact patient care and is a subgroup definition, which will be described in the SAP.	Section 4.1

Changes Incorporated in Protocol Version 3.3 (12 August 2022)	
Change and Reason for Change	**Section(s) Changed**
Change: Description of the events of special interest were aligned. Reason: To make the protocol internally consistent across sections regarding the events of special interest.	Protocol Summary Section 4.2.2
Administrative Changes and Clarifications	
Change: Clarified Version 3.3 of the CLN-PRO-V005 is specific to the United States. Reason: To provide clarification that this protocol version, allowing pediatric enrollment, is specific to the United States.	Cover Page Protocol Summary
Change: Updated Sponsor Medical Approver and Clinical Operations Representative and provided updated contact information. Reason: To reflect staff responsibility changes within Humacyte, Inc.	Protocol Approval Page and Section 1.0-Study Personnel
Change: Changed storage temperature from 2 – 10C to 2 – 8C. Reason: This was previously clarified in a clarification memo, but has now been updated in the protocol.	Section 6.3
Change: Updated manufacturer information to reflect Humacyte Global, Inc. as the manufacturer Reason: Clarification to reflect change in manufacture site	Section 6.2

A Phase 2 Study for the Evaluation of Safety and Efficacy of Humacyte's Human Acellular Vessel for Vascular Replacement or Reconstruction in Patients with Life or Limb-threatening Vascular Trauma

Medicinal Product:	Humacyte Human Acellular Vessel (Humacyte HAV)
Study No.:	CLN-PRO-V005
Sponsor:	Humacyte, Inc
Address:	2525 East NC Highway 54 Durham, NC 27713
Phone:	919-313-9633
CRO:	Atlantic Research Group 2421 Ivy Road, Suite 200 Charlottesville, VA 22903
Version:	3.2 (Amendment to version 3.2 dated 18 July 2018) 18 August 2021

Confidentiality Statement

STATEMENT OF COMPLIANCE

This trial will be conducted in compliance with the protocol and the following regulatory requirements:

- Declaration of Helsinki adopted by the 18th World Medical Assembly in Helsinki, Finland, in 1964, as last amended by the World Medical Assembly in 2013
- International Conference on Harmonization of Technical Requirements for Registration of Pharmaceuticals for Human Use (ICH), E6 Good Clinical Practice: Consolidated Guidance (ICH E6)
- ICH E2A Clinical Safety Data Management: Definitions and Standards for Expedited Reporting
- ICH E8 Guidance on General Considerations for Clinical Trials
- Applicable sections of United States Food and Drug Administration (FDA) Code of Federal Regulations (CFR), including:
    - 21 CFR Part 11, Electronic Records; Electronic Signatures
    - 21 CFR Part 50, Protection of Human Subjects
    - 21 CFR Part 54, Financial Disclosure by Clinical Investigators
    - 21 CFR Part 56, Institutional Review Boards
    - 21 CFR Part 312, Investigational New Drug Application

PRINCIPAL INVESTIGATOR AGREEMENT PAGE FOR THE PROTOCOL

I agree:

- To assume responsibility for the proper conduct of the study at this site, and to conduct the study in compliance with this protocol, any future amendments, and with any other study conduct procedures provided by the sponsor, Humacyte, Incorporated (Humacyte), or their authorized representatives.

- Not to implement any deviations from or changes to the protocol (including protocol amendments) without agreement from the sponsor and prior review and written approval from the Institutional Review Board (and FDA, if applicable) except where necessary to eliminate an immediate hazard to the patient(s), or for administrative aspects of the study (where permitted by all applicable regulatory requirements).

- That I am familiar with the appropriate use of the investigational medicinal product, as described in this protocol and any other information provided by the sponsor including, but not limited to the current Investigator's Brochure or equivalent document provided by Humacyte.

- To ensure that all persons assisting me with the study are adequately informed about the investigational medicinal product and of their study-related duties and functions.

- That I have been informed that certain regulatory authorities require the sponsor to obtain and supply details about the investigator's ownership interest in the sponsor or the Investigational Medicinal Product, and more generally about his/her financial ties with the sponsor. Humacyte will use and disclose the information solely for the purpose of complying with regulatory requirements.

Principal Investigator: _____

Name and Title

Signed: _____ Date: _____

PROTOCOL APPROVAL

Sponsor Medical Approval: Kiernan DeAngelis, M.D. Chief Medical Officer, Humacyte

Signed: Kiernan DeAngelis Digitally signed by Kiernan DeAngelis Date: 2021.08.18 14:24:54 -04'00' Date: _____

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LIST OF FIGURES

LIST OF ABBREVIATIONS

AE	Adverse event
AIDS	Acquired immunodeficiency syndrome
ALT	Alanine aminotransferase
aPTT	Activated partial thromboplastin time
ASA	Acetylsalicylic acid (aspirin)
ABI	Ankle brachial index
AST	Aspartate aminotransferase
AV	Arteriovenous
AVF	Autologous arteriovenous fistula
BP	Blood pressure
CAVG	Canine acellular vascular graft
CBC	Complete blood count
CKD	Chronic kidney disease
CTA	Computed tomography angiography
CM	Concomitant medication
eCRF	Electronic case report form
CRO	Contract research organization
DMC	Data Monitoring Committee
DTH	Delayed-type hypersensitivity
ECG	Electrocardiogram
ECM	Extracellular Matrix
ePTFE	Expanded polytetrafluoroethylene
ESRD	End-stage renal disease
ET	Early termination

LIST OF ABBREVIATIONS

FDA	Food and Drug Administration
GCP	Good Clinical Practice
GLP	Good Laboratory Practice
HAV	Human acellular vessel
HIV	Human immunodeficiency virus
IB	Investigator Brochure
ICF	Informed consent form
ICH	International Conference on Harmonization of Technical Requirements for Registration of Pharmaceuticals for Human Use
IgG	Immunoglobulin G
IHC	Immunohistochemistry
IM	Intramuscular
IMP	Investigational medicinal product
INR	International normalized ratio
IRB	Institutional Review Board
ISO	International Organization for Standardization
IU	International unit
IV	Intravenous
MedDRA	Medical Dictionary for Regulatory Activities
M	Month
N	Number (typically refers to participants)
NYHA	New York Heart Association
OTC	Over-the-counter
PAD	Peripheral arterial disease

LIST OF ABBREVIATIONS

PE	Physical examination
PHI	Protected health information
PI	Principal Investigator
PRA	Panel reactive antibodies
PT	Prothrombin time
PTFE	Polytetrafluoroethylene
QA	Quality Assurance
QC	Quality Control
RRT	Renal replacement therapy
SAE	Serious adverse event
SFA	Superficial Femoral Artery
SOP	Standard operating procedure
SVS WIfI	Society for Vascular Surgery: Wound, Ischemia, and foot Infection
US	Ultrasound
USA	United States of America
WFI	Water for injection
WBC	White blood cell(s)
WHO	World Health Organization

PROTOCOL SUMMARY

Full Title	A Phase 2 Study for the Evaluation of Safety and Efficacy of Humacyte's Human Acellular Vessel for Vascular Replacement or Reconstruction in Patients with Life or Limb-threatening Vascular Trauma
Clinical Trial Phase	Phase 2
Sponsor	Humacyte, Inc.
Planned Study Sites	Up to 10 sites in the United States
Sample Size	Up to 100 subjects
Study Population	Patients with vascular trauma to size appropriate vessels in the limb or torso, requiring replacement or reconstruction.
Inclusion Criteria	1. Patients with life or limb threatening traumatic injury to an arterial vessel in the limb or torso, other than the heart, which requires replacement or reconstruction. 2. Preoperative imaging or clinical examination indicates the damaged vessel has a defect length of ≤ 38cm and is appropriately size matched to the 6mm Human Acellular Vessel (HAV) per the judgment of the treating surgeon taking into account vasoconstriction and situational inflow and outflow considerations. 3. Autologous vein graft is either not feasible in the judgment of the treating surgeon (e.g. because of lack of availability of suitable conduit, presence of severe venous insufficiency) or is not desirable because of the urgency of revascularization 4. Aged 18 to 85 years old, inclusive 5. Able to communicate meaningfully with investigative staff and able to comply with study procedures. If the patient is unconscious then information from a reliable witness indicates that the patient would normally be able to understand and comply with study procedures 6. Patient or legal representative is able, willing and competent to give informed consent 7. Life expectancy of at least 1 year

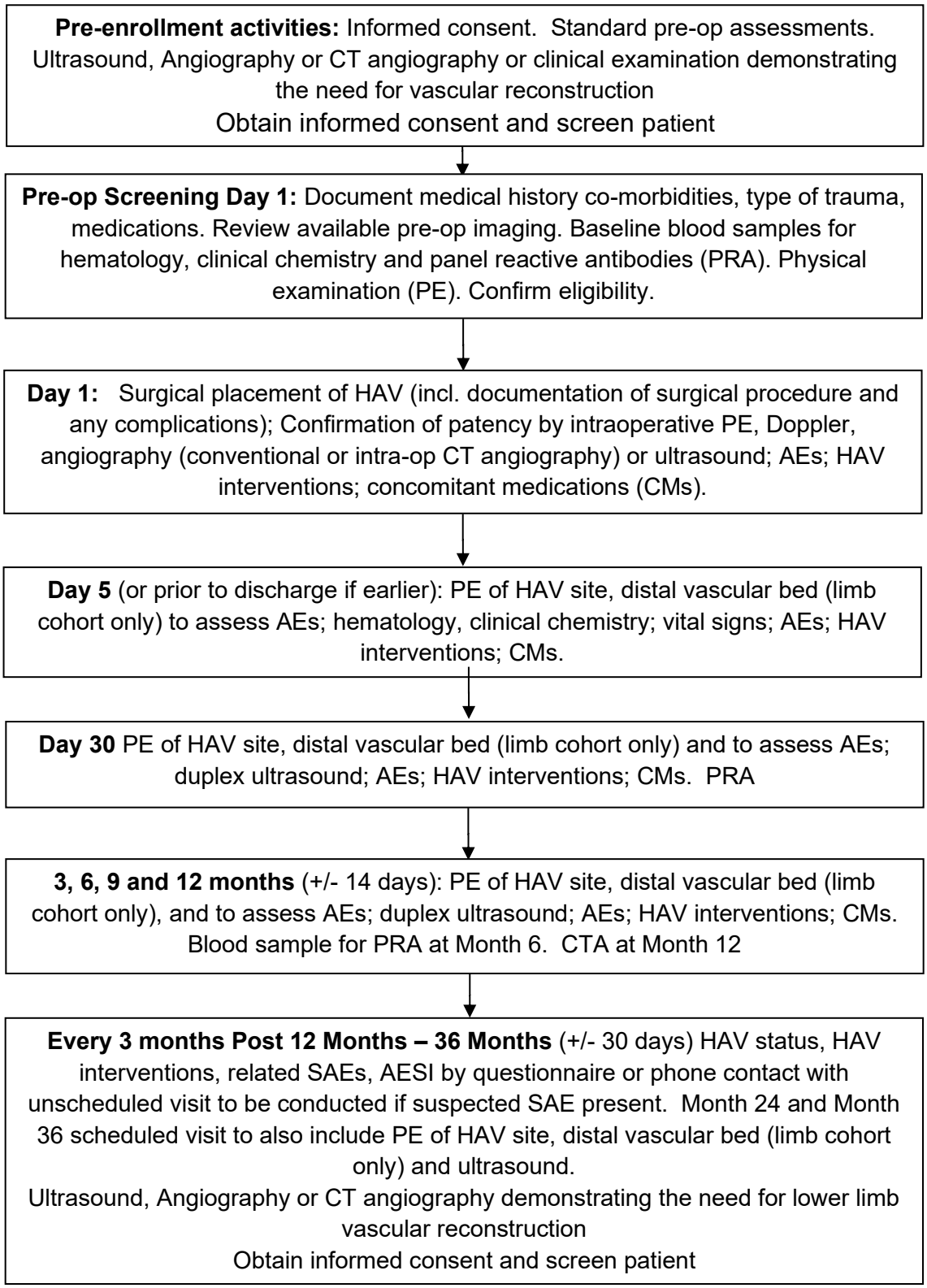
Exclusion Criteria	1. Mangled Extremity Severity Score (MESS) of ≥ 7. 2. Limb at high risk of amputation despite vascular reconstruction (e.g., because of crush injury) 3. Catastrophic injuries that make survival unlikely (e.g. Abbreviated Injury Scale (AIS) > 5 or Injury Severity Score (ISS) >60) 4. HAV may not be used for coronary artery repair. 5. Known pregnant women 6. Known medical condition which would preclude long term antiplatelet therapy after resolution of acute injuries 7. Any other condition which in the judgment of the investigator would preclude adequate evaluation of the safety and efficacy of the Humacyte Human Acellular Vessel (HAV) 8. Previous exposure to HAV 9. Known participation in any investigational study within the last 30 days 10. Employees of the sponsor or patients who are employees or relatives of the investigator
Expected Enrollment Start	3Q 2018
Accrual Period	24 months
Study Duration	The active study duration for each study participant will be 36 months from HAV implantation. All subjects will be followed for the initial 12 months. Beyond 12 months (Long-Term Follow Up), only subjects with a patent HAV will be followed out to a total of 36 months from HAV implantation. The total expected duration of the clinical study is 61 months.
Study Design	Prospective, multicenter, multi-cohort, non-randomized study There will be a limb cohort and a torso cohort.
Investigational Device/Intervention Description	Patients will be implanted with a Humacyte Human Acellular Vessel (HAV) as an interposition replacement or bypass using standard vascular surgical techniques.

Primary Objectives	**Safety** To evaluate the safety and tolerability of the Humacyte HAV in vascular trauma patients undergoing surgery for vascular replacement or reconstruction **Efficacy** To determine the rate of primary patency at 30 days
Secondary Objectives	**Safety** • To determine mechanical stability of the HAV based on freedom from aneurysmal degeneration, anastomotic bleeding or spontaneous rupture, infection, or significant stenosis • To determine HAV durability in terms of freedom from HAV removal or replacement **Efficacy** • To determine the patency of the HAV (primary, primary assisted and secondary) • To determine the rates of interventions needed to maintain / restore patency in the HAV • To determine the rate of limb salvage • To determine patient survival • To evaluate remodeling of HAV
Endpoints	Endpoints will be assessed for up to 36 months after HAV implantation. The primary analysis of the study will be conducted on the earlier of a) when the last subject enrolled reaches 30 days post-implant or b) all subjects enrolled in the initial 24 month accrual period have reached 30 days post-implant. **Primary Endpoints:** *Safety:* • Frequency and severity of adverse events *Efficacy:* • HAV primary patency at 30 days **Secondary Endpoints:**

	Safety:
	• Frequency of adverse events of special interest:
	  ○ Anastomotic bleeding or spontaneous rupture
	  ○ HAV infection
	  ○ Thrombosis
	  ○ Pseudoaneurysm formation
	  ○ Aneurysm formation
	  ○ Hemodynamically significant stenosis (>70% by duplex ultrasound criteria)
	• HAV partial or complete removal
	Efficacy:
	• HAV primary patency
	• HAV primary assisted patency
	• HAV secondary patency
	• Rate of HAV interventions
	• Limb salvage (limb cohort only)
	• Patient survival
	• HAV remodeling as shown by histopathology of any clinical explants

Protocol Approval (Version and Date)	Version 3.2 18 August 2021

Schematic of Study Design:



1. STUDY PERSONNEL

An updated list of all study personnel will be maintained by the CRO. Protocol amendments will not be required for staff changes at Humacyte, the CRO or the sites (except change of Principal Investigator at a site).

CRO Project Manager:

Brad Whitlow
Project Manager
Atlantic Research Group
2421 Ivy Road, Suite 200
Charlottesville, VA 22903

Email: bwhitlow@atlanticresearchgroup.com
Mobile: 828-442-0526
Office: 434-202-4537
Fax: 434-202-4537

Sponsor Medical Representative:

Kiernan DeAngelis, MD
Chief Medical Officer
Humacyte, Inc.
2525 East NC Highway 54
Durham, NC 27713, USA

Email: kdeangelis@humacyte.com
Office: 919-313-9633

Sponsor Regulatory Representative:

William Tente
Chief Regulatory Officer
Humacyte, Inc.
2525 East NC Highway 54
Durham, NC 27713, USA

Email: tente@humacyte.com
Mobile: 401-714-4151
Office: 919-313-9633

Sponsor Clinical Operations Representative:

Karthi Natarajan
Sr. Director, Clinical Development
Humacyte, Inc.
2525 East NC Highway 54
Durham, NC 27713, USA

Email: knatarajan@humacyte.com
Office: 919-313-9633

CRO Safety Representative:

Todd Fisher
Email: safety@atlanticresearchgroup.com
Sr. Drug Safety Specialist
Phone: 888-619-3216
Atlantic Research Group
Fax: 866-851-9318
2421 Ivy Road
Charlottesville, VA 22903

Medical Monitor:

Jennifer Prichard, MD
Email: jprichard@atlanticresearchgroup.com
Medical Monitor
Mobile: 434-249-5530
Atlantic Research Group
Phone: 888-619-3216
2421 Ivy Road
Fax: 866-851-9318
Charlottesville, VA 22903

2. BACKGROUND INFORMATION AND SCIENTIFIC RATIONALE

2.1. Background Information

In the civilian population, traumatic vascular injuries are mainly concentrated to the limbs and torso (abdomen, retroperitoneum, thorax and thoracic outlet). According to the PROspective Observational Vascular Injury Treatment (PROOVIT) database, designed to collect vascular trauma injuries from 24 Level I and Level II trauma centers in the United States, vascular injuries in the lower limb, torso, upper limb and neck have a distribution of 41%, 30%, 22%, and 6%, respectively. (DuBose, 2015, Faulconer, 2017) These reports incorporate a significant percentage of relevant venous injuries in addition to arterial. Additionally, in the civilian population, lower extremity bone fractures with associated arterial injuries are common, due to motor vehicle accidents, gunshot wounds, dog bites, and other situations resulting in blunt or penetrating trauma. (Helfet, 1990, Andrikopoulos, 1995, Akingba, 2012) In fact, the incidence of this type of vascular injury has increased considerably in the past 50 years. (Andrikopoulos, 1995) Although this type of injury represents less than 1% of all civilian injuries, fractures with associated vascular damage require special attention because of their potentially severe complications, including limb necrosis and amputation.

Currently, in order to attempt to salvage the injured limb or end-organ in the distal vascular bed and to prevent life threatening hemorrhage the vascular component of these injuries are treated with interposition or bypass grafting. This type of reconstruction is performed by using either autologous vein from the patient (typically great saphenous vein), or by using a synthetic graft, such as ePTFE or Dacron. However, the use of these grafts in the civilian population is not always possible or without additional risks. The patient may not have adequate autologous vein for harvest, and in many of these cases, such as dog bite injuries, the wound is “dirty” and a synthetic vascular substitute is contraindicated due to the risk of infection. (Akingba, 2012) Thus, civilians would benefit from a vascular graft that does not contain infection-prone synthetic material and has similar properties as human tissue, but does not require time consuming or high morbidity procedures to harvest vessels from the patient.

Warfighters could also benefit from an off-the-shelf biologic vascular graft. In modern combat, the incidence of vascular injury is much greater than in previous wars. The rate of vascular injury in the Vietnam War was 2-3%. But between 2002-2009, the rate of vascular injury was over 12% in a study of over 13,000 battlefield injuries. In combat scenarios vascular injuries occur in all locations of the body, but injuries to the lower extremities are the most common, followed by vascular injuries to the upper extremity, and then neck, followed by torso. (Rasmussen, 2010) Improvised Explosive Devices, or IED’s, caused more than 3,000 casualties per year during recent conflicts. (Cordesman, 2010) Arterial damage, laceration and thrombosis can require vascular reconstruction to save tissues from ischemia, necrosis, and further amputation. Harvesting autologous vein for vascular reconstruction is problematic

because IED casualties often have multi-limb injuries, making harvest of autologous vein highly risky or impossible. (Holcomb, 2011) Synthetic vascular reconstruction using synthetic vascular grafts made from Teflon (ePTFE)/Dacron is relatively contraindicated, since IED wounds are always “dirty”, and bacteria in the wound can colonize the synthetic graft, causing abscesses and sepsis.

Thus, there is a significant unmet medical need for alternative grafts, which can be used in situations where autologous vein is unavailable or undesirable to use and which more closely mimic human vascular tissue to avoid or reduce the infection complications associated with ePTFE and Dacron.

2.2. Scientific Rationale

Humacyte has developed an acellular, human tissue engineered vascular conduit, the human acellular vessel (HAV, to provide an alternative to synthetic materials and to autologous grafts in the repair of traumatic vascular damage. Because this product mimics native vascular tissue, it may possess the advantages of an autologous graft; it also has the benefits of synthetic grafts in that it is available off-the-shelf. Use of an off-the-shelf product avoids the surgical morbidity associated with vein graft harvest and most importantly allows vessel bypass surgery in patients who have no suitable vessels available. Because the product mimics a native vessel, it may not have the compliance mismatch associated with synthetic alternatives. In addition, pre-clinical studies in pigs, canines and primates have shown that the HAVs resist intimal hyperplasia at the anastomoses. (Quint, 2011, Prichard, 2011, Dahl, 2011) Upon implantation, the collagen matrix comprising the HAV is infiltrated with host cells and remodeled by the host. This could result in a vascular structure more similar to the histological composition of the native vascular tissue that may improve bypass longevity and be less likely to become infected. The latter potential advantage is of high importance in the repair of peripheral vascular trauma where most wounds are heavily contaminated.

2.3. Summary of Nonclinical Information

The non-clinical testing program was designed to comprehensively address:

- local and systemic effects of the product in multiple in vivo animal models acutely and chronically,
- functional aspects of product implanted into animal models as an arteriovenous conduit
- biocompatibility of the HAV material in standardized in vitro and in vivo test protocols.

Overall, the results of these studies indicated that the HAV extracellular matrix material was non-toxic, well tolerated, and met standards for biocompatibility. Generally, the HAVs functioned

as intended and maintained patency during the implantation period. (See the Investigator Brochure for a detailed summary of non-clinical data.)

Pre-implantation, the HAV has mechanical properties (burst pressure and suture retention strength) comparable with native human artery and vein. (Table 1.) There was no evidence that HAV strength deteriorated after long-term implantation into baboons.

Table 1: Summary of Mechanical Properties of Explanted Acellular Vessels

Test Material	Burst Pressure (mm Hg)	Suture Strength (g)
Pre-Implant Humacyte HAVs	3415 +/- 1011 (n=4)	180 +/- 44 (n=12)
Post-Explant Humacyte HAVs	3669 +/- 1305 (n=5)	276 +/- 84 (n=11)
Human Saphenous Vein	1,680 – 2,273[a]	196 +/- 2 (n=7)[a]
Human Artery	2,031 – 4,225[a]	200 +/- 119 (n=9) [a]

[a] From L'Heureux et al, *Nature Medicine*, 2006. (L'Heureux, 2006)

In the chronic animal testing Humacyte vessels produced using canine cells were implanted into 12 dogs (canine acellular vascular graft, CAVG) and 14 baboons (human acellular vessel -HAV) in a variety of anatomical locations. In general, the Humacyte vessels were safe and well tolerated, and functioned as intended.

Mechanical failure was not observed in any HAV. Calcification was not observed in any CAVG or HAV. No graft exhibited hemodynamically significant intimal hyperplasia. Unlike with ePTFE graft implantation, no evidence of systemic infection attributable to implantation of HAV was observed in any of the animals. One HAV developed an aneurysm that was resected and did not harm the animal. The HAV material showed no evidence of toxicity in hematology, clinical chemistry, and necropsy data. The HAVs could be accessed by venipuncture and hemostasis was achieved following needle puncture.

On microscopic analysis, the HAVs were found to be well integrated into the host tissue. Overall, the cellular host response to the HAVs demonstrated smooth muscle actin-positive cells within the vessel wall, endothelial cells lining the lumen, and an adventitial-like outer layer adjacent to the vessel. These findings indicate that implanted HAVs were populated with cell types that are characteristic of healthy native vasculature. Examination of the anastomotic sections showed that the HAVs were well integrated with adjoining vasculature with minimal intimal hyperplasia observed. Furthermore, IHC was employed to identify CD-68 positive macrophages in the venous intimal tissue. Studies have shown a substantial macrophage population has been observed within venous intimal tissue adjacent to inflammatory ePTFE arteriovenous grafts. (Kelly, 2002, Roy-Chaudhury, 2001) Only sparse CD-68 positive macrophages were observed, indicating that the degree and the aggressiveness of the intimal hyperplasia associated with the HAV were less than that typically associated with ePTFE grafts. (Prichard, 2011)

Over time, the organization and composition of extracellular matrix (ECM) components indicated that, aided by infiltration of host vascular cells, HAVs were remodeled in vivo in a manner that mimicked the dynamic remodeling process of native blood vessels. Given the difficulties associated with the baboon animal model, where mismatches in vein vs graft diameter were encountered and animals perturbed their wounds postoperatively, an overall assisted patency rate of approximately 80% (11/14) was achieved. In a xenogeneic transplant model that did not employ immunosuppression, the HAV material did not elicit biologically significant cellular or delayed-type hypersensitivity (DTH) immune responses. All animals developed immunoglobulin G (IgG) titers to the HAV material that did not appear to detrimentally impact vessel function.

In internationally recognized in vitro and in vivo International Organization for Standardization (ISO) test protocols, the HAV material met criteria for biocompatibility required of medical devices.

These data collectively support the safety of the HAV for the proposed clinical investigation.

2.4. Summary of Clinical Studies

2.4.1. Overview

The HAV clinical development program currently includes 7 clinical studies: 4 in patients with end-stage renal disease receiving hemodialysis (CLN-PRO-V001, CLN-PRO-V003, CLN-PRO-V006 and CLN-PRO-V007), 2 in patients with peripheral arterial disease (CLN-PRO-V002 and CLN-PRO-V004) and 1 in patients with vascular trauma (CLN-PRO-V005). Three Phase 1/2 studies have completed primary analysis with long-term follow-up ongoing (CLN-PRO-V001, CLN-PRO-V002 and CLN-PRO-V003), 1 phase 3 study completed enrollment and follow-up is ongoing (CLN-PRO-V006), 2 phase 2 studies (CLN-PRO-V004, CLN-PRO-V005) and one phase 3 study (CLN-PRO-V007) are open for enrollment.

As of 10 April 2018, 272 patients (244 hemodialysis access patients and 28 PAD patients) have received a HAV. The first implant for hemodialysis was performed in December 2012, and the first peripheral arterial bypass in October 2013. Overall, the total treatment exposure is approximately 329 patient years in the hemodialysis access population and 55 patient years in the PAD population. More information on the clinical profile of the HAV in these ongoing studies is provided in the Investigator Brochure.

2.4.2. Experience in Peripheral Arterial Bypass Patients

Humacyte has two phase 2 studies to assess the safety and efficacy of the HAV when used as an above-knee arterial bypass graft. The first study, CLN-PRO-V002, is a single group uncontrolled study conducted at 3 sites in Poland that is fully enrolled and in long-term follow up. Eligible patients required a femoro-popliteal bypass graft for the management of

symptomatic peripheral arterial disease. Pre-operative imaging (conventional or CT angiography) must have demonstrated at least two below knee vessels patent to the ankle with good runoff. The proximal anastomosis was expected to be below the inguinal ligament and the distal anastomosis above the knee. Autologous vein grafts must not have been suitable or feasible (e.g., because of severe venous disease or prior use of leg veins for other bypass surgery or there is a clinical need to preserve those veins for future bypass surgery in the coronary or peripheral circulation).

The HAV was implanted using standard vascular surgical techniques and the patency of the bypass confirmed by intraoperative angiography (conventional or intra-op CT angiography) or ultrasound. The patient was then followed up at study visits at 15 days, 6 weeks and 3, 6, 12, 18 and 24 months. At each visit safety was assessed by clinical examination and adverse events, and the HAV was examined using duplex ultrasound to visualize the entire length to confirm patency, flow and to detect stenosis, aneurysm development or dilatation.

The primary objectives of the study are to evaluate the safety and tolerability of the Humacyte HAV in PAD patients undergoing above-knee femoro-popliteal bypass surgery and to determine the patency (primary, primary assisted and secondary) rate of the Humacyte HAV at 24 months. Secondary objectives include assessment of the panel reactive antibodies (PRA)) and IgG response to the HAV and to assess patency (primary, primary assisted and secondary) at 6, 12 and 18 months, to determine the rates of interventions needed to maintain / restore patency in the HAV, to assess any effect of implantation on claudication, rest pain and ischemic ulcers and to assess any effect on ankle-brachial index (ABI).

The second PAD study of similar design, CLN-PRO-V004, is being conducted in the US with enrollment ongoing.

2.4.2.1. CLN-PRO-V002 Study Results (24 M)

Recruitment began in October 2013 and was completed in June 2014 with 20 patients implanted. Thirteen patients completed the 2 year follow up visit. Of the seven patients terminating the study early, three died and four were withdrawn after occlusion of the HAV. None of the deaths were considered related to the investigational device or procedure.

Kaplan-Meier analyses in which deaths were censored revealed primary, primary assisted, and secondary patency probability rates of 79.2%, 79.0%, and 89.5% at Week 26, 63.3%, 63.2%, and 84.2% at Month 12, 63.3%, 63.2%, and 79.0% at Month 18, and 58.1%, 57.9%, and 73.7% at Month 24.

Six patients (30%) required at least 1 graft intervention to maintain or restore HAV patency during the study. Four patients required 1 intervention and 1 patient each required 3 and 4 interventions. Most interventions successfully restored patency. However, in 1 patient the graft patency could not be restored and the HAV was replaced with an alternative bypass graft. Two patients, who had previously undergone successful interventions, developed a recurrent

thrombosis which was not treated and the HAV was left occluded. Two patients experienced HAV thrombosis with no or minimal symptoms and refused interventions on the HAV.

All 20 patients experienced AEs (a total of 92 events). Thirty-one of these events in 13 patients were considered serious. The most frequent AEs reported included graft thrombosis (35% of patients), anastomotic stenosis (20% of patients), lymphocele (20% of patients), and local swelling (15% of patient). Those SAEs reported by at least 2 patients were graft thrombosis (6 patients, 30%) and anastomotic stenosis (2 patients, 10%).

No patient showed an increase in PRA levels. Two patients had a significant (>2 fold) increase from baseline in IgG levels. One of these patients experienced a thrombosis of the HAV between 3 and 6 months after implantation, while the other patient has had no HAV-related AEs and continues to have primary patency. Neither patient has had any evidence of dilatation or structural degeneration of the HAV.

CLN-PRO-V002 Conclusions:

- Humacyte HAV was safe and well tolerated in PAD patients.
- The HAV is able to withstand long term use in a high pressure, high outflow resistance arterial circuit.
- Patency rates for the HAV are within the ranges of patency rates of synthetic and autologous grafts presented in the literature.
- Humacyte HAV was not immunogenic.

2.4.3. Experience in Hemodialysis Patients

Two phase 2 trials, one in Poland (CLN-PRO-V001) and one in the US (CLN-PRO-V003) have completed enrollment. Both recruited subjects requiring hemodialysis access for end-stage renal disease whom were not suitable for creation of an AVF. Most subjects had undergone previous vascular access procedures, in many cases multiple attempts including both AVFs and synthetic grafts. Initial results from these phase 2 studies are discussed below.

The primary objectives of these two studies are to evaluate both the safety of HAV and its efficacy in terms of primary and secondary patency at 6 months. Secondary objectives include measurement of a panel of reactive antibodies (PRA) response, development of IgG antibodies to the extracellular matrix material in the HAV and a 2-year evaluation of patency and an assessment of the need for interventions to maintain/restore patency. Follow up has now been extended up to 120 months.

A phase 3 randomized study comparing HAV with ePTFE grafts (CLN-PRO-V006) has completed enrollment in the US, Europe, and Israel. A second phase 3 randomized study (CLN-PRO-V007) comparing HAV with AVF is currently enrolling in the US. As the sponsor is blinded, no efficacy information currently available for the phase 3 studies; however, blinded safety data is presented in the Investigator Brochure.

2.4.3.1. CLN-PRO-V001 and CLN-PRO-V003 Study Results (24 M)

All subjects (n=60) have now completed at least 24 months since implantation (or had a censoring event). The first subjects recruited are now beyond 60 months after HAV implantation, some with functioning HAV for hemodialysis access. Together these two trials provide more than 150 years of follow up during which the HAV has been used for more than 15,000 hemodialysis sessions.

When HAV thrombosis has occurred it has almost always been managed successfully, often allowing immediate resumption of dialysis without the need for the placement of a dialysis catheter. One non-serious arteriovenous graft aneurysm was reported in Study CLN PRO V001 (moderate in intensity, considered possibly related to IMP and considered not related to procedure – this patient died before the Sponsor could complete the follow up of this event). An expected number of small pseudoaneurysms have been observed, which is consistent with all surgically-created hemodialysis access. Most have resolved spontaneously with only 2 cases requiring surgical intervention. Flow rates through the HAV were more than sufficient to allow for effective dialysis.

In both studies, the product has generally been well tolerated and blood chemistry, hematology and coagulation data are not indicative of any HAV-associated toxicity. Immunogenic response to the HAV material has not been observed as demonstrated by a general lack of HAV-related change in PRA levels (Class I or II). Three subjects had elevations in their PRA levels: all 3 subjects had experienced one or more renal transplant failures; one subject recently; one subject developed septic shock about a month before the elevated value; and the third subject, who was severely debilitated with a decubitus ulcer, died approximately a month after HAV abandonment.

IgG titers increased in 5 subjects; in 4 cases the IgG titer increased and then decreased while the HAV remained functional with no clinical evidence of an inflammatory response; in one case the IgG titer increase occurred in a subject who maintained primary patency.

Adverse events (AEs) related to the HAV / access site (excluding thrombotic events) were few; there have been only three access-site infections, of which only one required removal of part of the HAV. There have been:

- 1 transplant (known to be functioning well at 12 months post-transplant)
- 15 deaths, all after abandonment or during follow-up; none of the deaths were considered related to the presence of the HAV

Patency data for the two studies in dialysis access has been pooled for a combined Kaplan Meier analysis (Lawson, 2016). Based on these K-M plots the patency at 6, 12 and 24 months is estimated to be 60%, 26% and 15% (primary patency) and 97%, 91% and 77% (secondary patency).

2.4.4. Human Acellular Vessel Host Response and Remodeling Data

Humacyte has been able to assess the general host response to the HAV in a number of human participants; this was accomplished through the microscopic examination of explanted HAV and adjoining tissue samples obtained during surgical revision procedures in eight cases. The analysis (mostly of a section close to the venous anastomosis) included assessments of:

- Cellular infiltration of histotypic, inflammatory and immunological populations.
- Extracellular remodeling processes, including neo-synthesis and reorganization of ECM components that typically occur in native blood vessels.

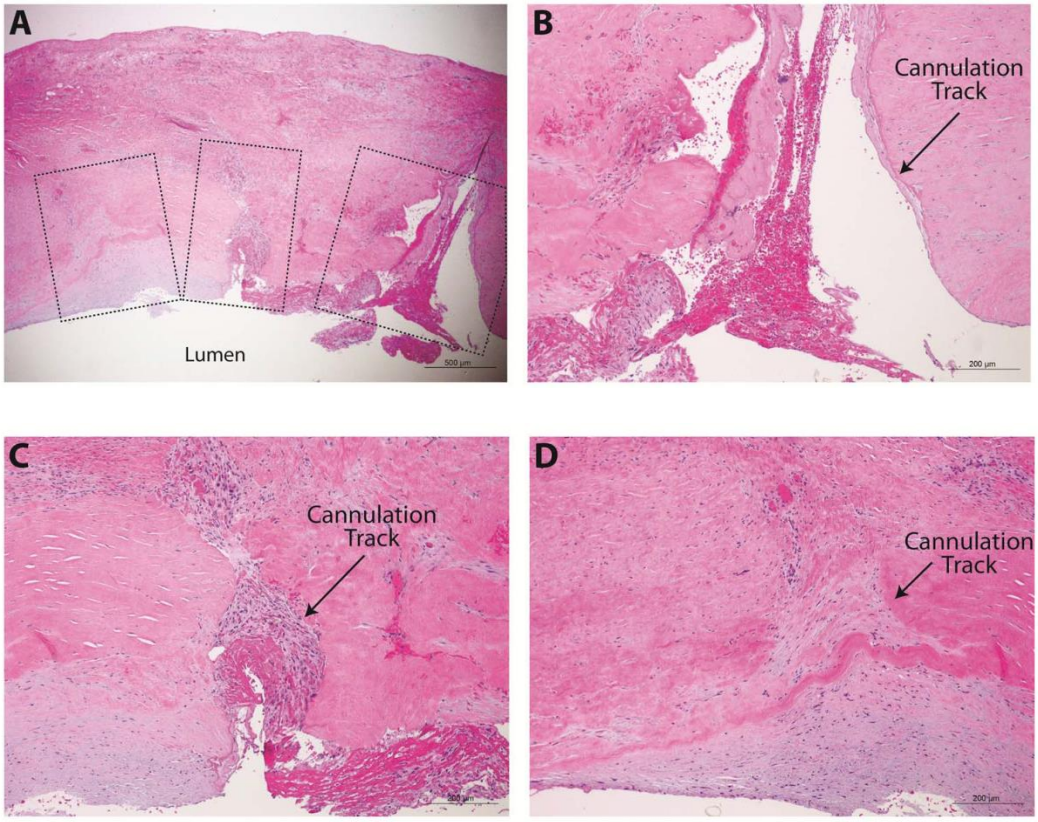
In these cases, small segments of the HAV and adjacent vascular tissue were explanted, fixed in formalin solution and shipped to Humacyte for analysis. Implant duration ranged from 16 to 55 weeks (median: 37 weeks).

In man, the HAV remodeled in a manner consistent with that observed in primate studies. There was infiltration of cell populations that are normally associated with angiogenesis and vascular organization and structure; namely, those with endothelial, smooth muscle and fibroblastic phenotypic characteristics were observed. Endothelial cells formed a monolayer on the luminal surface of the HAV. Migration of actin-positive smooth muscle cells into the wall of the HAV was consistently observed. A well-vascularized adventitial layer of non-constrictive fibrous tissue formed around HAV. Infiltration of the graft material by inflammatory and immunoreactive cell populations was either not evident or was mild and generally unremarkable. Degradation or breakdown of the implant was not observed.

Histotypic neo-synthesis and reorganization of the ECM was observed in patterns indicative of integration of the HAV into the host. An increase in the density of collagen type I, the main type of collagen found in the wall of native blood vessels, was apparent in the majority of HAV explant specimens. The structure of collagen type I in these specimens exhibited a more mature, organized pattern, with distinct fibers and a prominent circumferential alignment evident in explanted samples in comparison with pre-implant specimens. In some specimens, the fibrillar staining pattern of collagen III became more prominent and more organized, with a circumferential orientation. Fibronectin levels and staining patterns remained unchanged.

Cannulation sites within the HAV appeared to be repaired by the host in a fashion similar to wound repair in the body (Figure 1). In one case, an explanted specimen was tested for suture retention strength at the time of explant and exhibited a substantial increase over the pre-implant level.

Figure 1 Images of Mid-Vessel Segment Explanted at 11-Months Post-Implant



A: Low magnification showing 3 cannulation sites (in dashed boxes),

B: Fresh cannulation track,

C: Cannulation track during remodeling

D: Older cannulation track that has been repaired.

The images above show a mid-vessel segment explanted at 11 months post-implant, and shows several prior cannulation tracts from dialysis access. Section B shows a very recent cannulation site with fresh clot extending into the tract from the lumen. Sections C and D show partially healed cannulation tracts, with evidence of cellular repopulation extending in from the lumen. Remodeled cannulation tracks contain new collagen and a few micro-conduits.

In conclusion, the HAVs were remodeled by the host to form a vascular-like structure more similar to the histological appearance of native vasculature. The HAVs were repopulated by cell types that are characteristic of healthy native vasculature. Evidence of ECM remodeling processes, including neo-synthesis and reorganization of ECM components that typically occur in native blood vessels, were observed. The cellular infiltration and ECM remodeling patterns were indicative of the integration of the HAV into the host.

2.4.5. Conclusions

Clinical experience indicates that the HAV remains mechanically strong over implantation periods of more than 60 months with no evidence of dilatation. During more than 200 patient-years of follow up across the three phase 2 studies only one case of infection of the HAV material itself has been reported. The serious adverse event (SAE) profile has been typical of that expected in the dialysis and PAD populations. In hemodialysis populations, secondary patency of the HAVs is substantially higher than the historical data for both ePTFE and AVF (accounting for non-maturation). In PAD, patency is in line with historical ePTFE and autologous conduit for above knee bypass. No evidence of immunogenicity of the HAV has been found and the HAV remains mechanically robust even after repeated puncture for hemodialysis and under high pressure, high outflow resistance in arterial reconstruction.

These data support the use of HAV in future phase 2 and phase 3 studies for vascular replacement and reconstruction in diseased or damaged (trauma) vessels.

2.5. Potential Risks and Benefits

2.5.1. Potential Risks

It is anticipated that subjects participating in the study will be exposed to similar risks to those associated with other arterial conduits. Risks associated with the study investigational product may include but are not limited to:

- Thrombosis/occlusion of the conduit or host vessels, with consequent limb ischemia
- Embolism from a thrombosed conduit
- Bleeding and hematoma formation at the surgical site
- Infection – at the surgical site or systemic
- Stenosis of the conduit or its anastomoses
- Aneurysm or pseudoaneurysm formation
- Swelling of the limb
- Failure/injury to the target end-organ
- Bleeding/hemorrhage in the peritoneum or retroperitoneum

Regular clinical examination of the HAV implantation site and assessment of the patency, blood flow and diameter using ultrasound during the study should allow early detection of complications and permit appropriate intervention including HAV explantation.

The HAV is grown using donor human aortic smooth muscle cells. The vessel is decellularized during manufacturing and thus consists of human extracellular matrix proteins. It is possible that the HAV may provoke an immune response which may lead to damage to the HAV and possible

cross reactivity against host proteins. Possible antibody formation will be assessed by analyzing PRA.

2.5.2. Potential Benefits

Patients who undergo implantation of the Humacyte HAV may benefit from improved patency and a reduced number of interventions versus a conventional ePTFE or Dacron graft. This may result from a decreased propensity for anastomotic and downstream neointimal hyperplasia, which often leads to graft occlusion with synthetic grafts. In addition, risks of infection typically encountered with conventional synthetic grafts may be decreased with the Humacyte HAV. Finally, the longevity (secondary patency) of the Humacyte HAV may be greater than that of conventional synthetic grafts.

Patients may also benefit from reduced morbidity secondary to harvest of autologous vessels for vascular reconstruction. Harvest of autologous vascular conduit requires additional time to perform and in the urgent/emergent trauma scenario, time is of the essence and is key to positive patient outcomes. Use of saphenous, or other autologous vessels for purposes of vascular repair in young, otherwise healthy patients prevents its future potential use for other indications (e.g. coronary bypass, etc.). Vascular reconstruction using synthetic vascular grafts (ePTFE, or woven materials like Dacron) may be contraindicated for use in trauma scenarios since the wounds are often contaminated allowing bacteria in the wound to colonize the synthetic graft potentially leading to abscesses, hemorrhage secondary to anastomotic blow out, blood stream infection, and sepsis. Unlike other readily available synthetic vascular grafts made from ePTFE or Dacron, HAVs are comprised of human proteins, resulting in grafts that may be less prone to infection than synthetic materials, as shown in pre-clinical studies. (Kirkton, 2018)

2.5.3. Risk-Benefit Rationale

The risks anticipated in this study are similar to those associated with currently marketed prosthetic grafts used for vascular repair. The potential advantages of the HAV compared to currently marketed grafts may lead to a lower complication rate and reduced need for surgical intervention and graft replacement as well as the potential for reduced secondary complications associated with autologous vessel harvest.

This is the first in man study in which the HAV will be used to repair vascular injuries within the torso (thorax, thoracic outlet, abdomen, and retroperitoneum) and so risks specific to those locations have not been specifically characterized. However, these risks are not expected to be significantly different than those experienced/reported when used in the upper or lower extremity for AV access or arterial reconstruction. The DMC will review safety data of the torso cohort on the earlier of when the first torso subject reaches 3 months post-implantation or when the first 2 torso subjects have both reached 30 days post-implantation. Overall recruitment will be restricted to a maximum of 100 subjects who receive implants.

3. STUDY OBJECTIVES

3.1. Primary Objectives

This is an open label phase 2 study. There is no formal hypothesis testing.

Safety:

- To evaluate the safety and tolerability of the Humacyte HAV in vascular trauma patients undergoing surgery for vascular replacement or reconstruction

Efficacy:

- To determine the rate of primary patency at 30 days

3.2. Secondary Objectives

Safety:

- To determine mechanical stability of the HAV based on freedom from aneurysmal degeneration, anastomotic bleeding or spontaneous rupture, infection, or significant stenosis
- To determine HAV durability in terms of freedom from HAV removal or replacement

Efficacy:

- To determine the patency of the HAV (primary, primary assisted and secondary)
- To determine the rates of interventions needed to maintain / restore patency in the HAV
- To determine the rate of limb salvage
- To determine patient survival
- To evaluate remodeling of HAV

4. STUDY DESIGN

4.1. Description of the Study Design

Prospective, multicenter, multi-cohort, non-randomized phase 2 study.

There will be a limb cohort and a torso cohort. The limb cohort will include patients who require repair of a vessel contained to the upper or lower extremity. Upper extremity vasculature is defined as the axillary and more distal vessels. The lower extremity vasculature is defined as the common femoral and more distal vessels. Examples of size appropriate vessels within the extremities include (but are not limited to):

- Axillary
- Brachial
- Basilic
- Popliteal
- the femoral vessels

The torso cohort includes patients who require repair of vessels within the thorax (excluding the heart), abdomen, and retroperitoneum. A vascular repair in which any portion originates or terminates in the torso will be considered a patient in the torso cohort even if the other end of the repair resides in an extremity. End to side anastomotic takeoff from a larger caliber vessel (e.g. aorta, vena cava) is acceptable for enrollment within this study. Examples of size appropriate vessels within the torso include (but are not limited to):

- subclavian
- brachiocephalic/innominate
- celiac
- hepatic
- splenic
- superior mesenteric
- renal
- the iliac vessels

The DMC will review the earlier of when the first torso subject reaches 3 months post-implantation or when the first 2 torso subjects have both reached 30 days post-implantation.

4.2. Study Endpoints

Endpoints will be assessed for up to 36 months after HAV implantation. The primary analysis of the study will be conducted on the earlier of a) when the last subject enrolled reaches 30 days post-implant or b) all subjects enrolled in the initial 24 month accrual period have reached 30 days post-implant.

4.2.1. Primary Endpoints

Safety:

- Frequency and severity of adverse events

Efficacy:

- HAV primary patency at 30 days

4.2.2. Secondary Endpoints

Safety:

- Frequency of adverse events of special interest:
  - Anastomotic bleeding or spontaneous rupture
  - HAV infection
  - Thrombosis
  - Pseudo-aneurysm formation
  - Aneurysm formation
  - Hemodynamically significant stenosis (>70% by duplex ultrasound criteria)
- Frequency of study conduit removal

Efficacy:

- HAV primary patency
- HAV primary assisted patency
- HAV secondary patency
- Rate of HAV interventions
- Limb salvage (limb cohort only)
- Patient survival
- HAV remodeling as shown by histopathology of any clinical explants

4.3. Duration of Study Participation

The active study duration for each study participant will be 36 months from HAV implantation. All subjects will be followed for the initial 12 months. Beyond 12 months (Long-Term Follow Up), only subjects with a patent HAV will be followed out to a total of 36 months from HAV implantation. The total expected duration of the clinical study is 61 months.

5. STUDY POPULATION

5.1. Description of the Study Population

The study population will consist of patients with vascular trauma to size appropriate vessels in the limb or torso, requiring replacement or reconstruction.

5.1.1. Patient Inclusion Criteria

1. Patients with life or limb threatening traumatic injury to an arterial vessel in the limb or torso, other than the heart, which requires replacement or reconstruction
2. Preoperative imaging or clinical examination indicates the damaged vessel has a defect length of ≤ 38cm and is appropriately size matched to the 6mm Human Acellular Vessel (HAV) per the judgment of the treating surgeon taking into account vasoconstriction and situational inflow and outflow considerations.
3. Autologous vein graft is either not feasible in the judgment of the treating surgeon (e.g. because of lack of availability of suitable conduit, presence of severe venous insufficiency) or is not desirable because of the urgency of revascularization
4. Aged 18 to 85 years old, inclusive
5. Able to communicate meaningfully with investigative staff, and able to comply with entire study procedures. If the patient is unconscious, then information from a reliable witness indicates that the patient would normally be able to comply with study procedures
6. Patient or relative is able, willing and competent to give informed consent
7. Life expectancy of at least 1 year

5.1.2. Patient Exclusion Criteria

1. Mangled Extremity Severity Score (MESS) of ≥ 7
2. Limb at high risk of amputation despite vascular reconstruction (e.g., because of crush injury)
3. Catastrophic injuries that make survival unlikely (e.g. Abbreviated Injury Scale (AIS) > 5 or Injury Severity Score (ISS) >60)
4. HAV may not be used for coronary artery repair
5. Known pregnant women
6. Known medical condition which would preclude long term antiplatelet therapy after resolution of acute injuries
7. Any other condition which in the judgment of the investigator would preclude adequate evaluation of the safety and efficacy of the HAV
8. Previous exposure to HAV

9. Known participation in any investigational study within the last 30 days
10. Employees of the sponsor or patients who are employees or relatives of the investigator

6. INVESTIGATIONAL MEDICINAL PRODUCT

Additional information on the manufacturing process and testing of the investigational medicinal product (IMP)is provided in the Investigator Brochure.

6.1. Product Description

The IMP is a Humacyte Human Acellular Vessel (HAV), which is a tissue-engineered vascular prosthesis for vascular bypass or reconstruction in patients with peripheral vascular disease or peripheral vascular trauma. It is a sterile, non-pyrogenic acellular tubular vessel composed of human collagen types I and III and other extracellular matrix proteins, including fibronectin and vitronectin. The HAV is 6 mm in diameter and approximately 42 cm in length. The product is supplied on a silicone mandrel immersed in sterile phosphate buffered saline in a sealed and labeled plastic container.

There is no placebo or comparator control group in this study.

6.2. Manufacturer of the IMP

The HAV is manufactured by:

AlloSource

6278 S. Troy Circle

Centennial, CO 80111 USA

Traceability of the HAV during and after the clinical investigation will be achieved by the assignment of lot numbers. A unique identifying lot number will be assigned to each vessel.

6.3. Packaging, Storage, and Labeling

Packaging: Each HAV is contained in a sealed, flexible plastic primary container closure system that was developed by Humacyte. The system meets container/closure requirements to maintain sterility as well as product and fluid integrity. The vessel is contained inside the system in a fixed manner, immersed in a sterile, phosphate buffered saline. The total volume of the storage solution is approximately 300 mL.

Storage: The product is shipped under controlled conditions to maintain temperature at 4˚C (range: 2 – 10˚C). The product should be stored in a refrigerator that maintains this temperature range. The HAV **MUST NOT** be allowed to freeze.

Labeling: The IMP will be labeled according to applicable guidelines and relevant regulatory agency requirements. A tamper resistant label affixed to the secondary container will be used to ensure that the product is not compromised prior to use.

6.4. Implantation of the Humacyte Human Acellular Vessel (HAV)

The Humacyte HAV is implanted using standard vascular surgical techniques similar to placement of predicate peripheral vascular prostheses (see study manual for details).

Tunneling of the HAV, if required, must be performed using a sheathed tunneler. After inserting the assembled tunneler into the tissue, the inner mandrel of the tunneler should be removed from the sheath. The sheath is lubricated with saline and then with the silicone mandrel in place, the HAV can be easily pushed through the sheath without the need to tie to the inner mandrel and pulled through the tunneler (see study manual for details).

After placement, HAV patency and integrity are checked by pressurizing the conduit. Prior to completion of surgery, HAV patency is confirmed by physical exam, Doppler, angiography (conventional or intra-op CT angiography) or ultrasound. The surgical site is closed using standard techniques.

Implantation of the HAV will be undertaken by qualified vascular surgeons experienced in peripheral vascular surgery.

6.5. IMP Accountability Procedures

Documentation of receipt, dispensing, and return of all IMP must be maintained by the Principal Investigator or his/her designee. It is the Principal Investigator's responsibility to ensure that all IMPs are kept in a secure location, with access limited to individuals authorized by the Investigator. The product will be shipped with the IMP Shipment Confirmation Form. Once signed, the form should be returned to Humacyte or authorized designee, and the original will be maintained in the Investigator's Files. The IMP Accountability Log will be used to account for all IMP received, dispensed, and returned and must be maintained by the site until the conclusion of the study. Following accountability of the IMP by Humacyte or their authorized designee, all unused IMP will be returned to Humacyte.

7. OTHER TREATMENTS AND MEDICATIONS

7.1. Prior and Concomitant Medications

Prior medications are defined as all prescription and over the counter (OTC) medications taken within 7 days (whether continuing or not) prior to Day 1. All prior and concomitant medications (including immediately pre-surgery and post-surgery medications) must be listed in the patient's medical record and recorded on the eCRF. Drugs used during anesthesia should be recorded in the anesthesia records but should not be transcribed into the eCRF. Patients should be questioned at each study visit concerning any new medications or changes in current medications. **Note: particular attention should be made to identify the use of antithrombotic or antiplatelet agents (e.g., aspirin, clopidogrel, prasugrel, direct thrombin inhibitors, factor Xa inhibitors, or vitamin K antagonists).**

For each medication taken, the following information will be collected:

- Medication generic name / components of combination product
- Dose
- Route of administration
- Frequency of administration
- Date started
- Date stopped
- Indication for use

7.2. Essential, Precautionary and Prohibited Medications

7.2.1. Essential Medications

All patients should receive both antibiotic and antithrombotic prophylaxis in conjunction with HAV implantation:

Antibiotic prophylaxis:

- All patients must have at least 1 day of antibiotic prophylaxis the same day as surgery in accordance with local hospital guidelines. Longer antibiotic prophylaxis is at the discretion of the investigator.

Antithrombotic prophylaxis:

- Intraoperative heparin: the doses of heparin to be used during surgery will be determined by the investigator.

- Further measures to prevent venous thromboembolism are at the discretion of the investigator and may include LMWH.
- If antiplatelet therapy was not ongoing at the time of surgery it should be commenced as soon as medically appropriate post operatively. Recommended antiplatelet therapy (aspirin 81-325 mg and/or clopidogrel 75 mg daily) is at the discretion of the investigator and should continue long term while the HAV is in place. If the patient is unable to tolerate aspirin and/or clopidogrel the choice of antiplatelet regimen is at the investigator's discretion.

7.2.2. Restricted Medications

Vitamin K antagonists, antiplatelet agents other than aspirin and clopidogrel, direct thrombin inhibitors and factor Xa inhibitors (e.g., dabigatran, apixaban and rivaroxaban) should be avoided unless essential for treatment of a medical condition arising postoperatively. In that case consideration should be given to modification or cessation of antiplatelet therapy. Antiplatelet therapy should be restarted on cessation of these anticoagulant drugs.

8. STUDY PROCEDURES / EVALUATIONS

8.1. Clinical Evaluations Through Month 12

- Medical History – pre-operatively, from patient / legal representative interview and medical records covering relevant past medical history.
- Smoking history
- Medication History – prescription and OTC medication from Day -7 onwards (see Section 6.7). Particular attention should be paid to the identification of OTC medications containing aspirin.
- Physical Exam – full exam (as far as possible) at pre-operative screening, 12 month visit or final study visit for early termination (ET). Clinical examination of the operative site and HAV at all post-operative visits; exam of distal vascular bed (limb cohort only); physical exam for lymphadenopathy; additional clinical exam as needed to evaluate adverse events
- Vital signs (heart rate, blood pressure and temperature) – at D5
- Blood samples for hematology, clinical chemistry at pre-operative screening and Day 5 and PRA at pre-operative screening, Day 30 and M6
- Pre-operative imaging (ultrasound or angiography) is at the discretion of the investigator based on the clinical condition of the patient. It may be omitted if not deemed necessary in order to proceed to vascular surgical repair
- Adverse events – post-operatively on Day 1 and at all post-operative visits, the patient will be asked a general question about his/her health and for any HAV problems since the previous visit
- Intraoperative HAV bypass or interposition repair exam to assess anastomotic anatomy, patency and runoff. This may include physical exam, Doppler exam, angiography (conventional or intra-op CT angiography) or ultrasound at the investigator's discretion.
- Duplex ultrasound – clinical assessment at all postoperative visits from day 30 thru M12, to assess HAV patency, mid HAV diameter and stenosis. The full length of the HAV should be imaged at each assessment to monitor for aneurysm development
- Documentation of HAV interventions, surgical procedures and any complications immediately postoperatively through Month 12

8.2. Clinical Evaluations in Long Term Follow Up (Post Month 12 to Month 36)

- The status of the patient and HAV will be ascertained every 3 months from post Month 12 until 36 months after HAV implantation by telephone contact with the patient and/or his physician. If a suspected SAE related to HAV is discovered an unscheduled visit should be conducted to investigate.
- Only related SAEs and all AESI will be reported.
- Visits at Month 24 and Month 36 are to be conducted in person with a physical exam of the HAV site and duplex ultrasound imaging of the HAV.

8.3. Laboratory Evaluations

8.3.1. Clinical and Research Laboratory Evaluations and Specimen Collection

The following parameters will be measured wherever possible at pre-operative screening and all should be measured at Day 5

- Hematology: hemoglobin, hematocrit, RBC, white blood cells (WBC) with differential, platelet count
- Clinical chemistry: sodium, potassium, calcium, blood urea nitrogen, creatinine, albumin, total bilirubin, glucose (non-fasting)
- PRA will be measured at pre-operative screening, Day 30, and Month 6.

All laboratory tests (except assay of PRA) will be conducted at certified hospital laboratories. Routine monitoring, maintenance or calibration of laboratory equipment is required per local site procedures. Samples for PRA analysis will be shipped to LabConnect for storage until they are sent for analysis at a central laboratory. Details concerning sample collection and processing can be found in the Study Manual.

8.4. Imaging Evaluations

8.4.1. CT Angiography and Conventional Angiography

CT angiography or conventional angiography will be conducted as pre-operative screening when feasible. Pre-operative imaging is at the discretion of the investigator based on the clinical condition of the patient. It may be omitted if not deemed necessary in order to proceed to vascular surgical repair.

8.4.2. Duplex Ultrasound

Duplex ultrasound examinations will be performed at Day 30, 3, 6, 9,12, 24, and 36 months and follow standard bypass graft imaging protocols, including B-mode, power Doppler and color duplex ultrasound imaging of the HAV with velocity spectral waveform analysis. The purpose of this duplex ultrasound surveillance is to detect HAV stenosis and aneurysm development. An alternative imaging method (e.g. CTA, MRI, etc.) may be substituted for duplex ultrasound at the discretion of the investigator if it is medically appropriate and in the best interest of the patient.

Determination of intraoperative HAV patency on Day 1 is required by physical examination, Doppler exam, angiography (conventional or intra-op CT angiography), or ultrasound at the discretion of the investigator.

8.5. Study Schedule

8.5.1. Pre-operative Screening (Day 1)

Potential study participants who are being considered for surgical repair of vascular injury appropriate for inclusion into the study will be informed about the study and invited to participate. After explanation of the potential risks and benefits of the HAV and of the study procedures, written informed consent will be obtained. No study specific procedures may be performed prior to patient consent. If the patient is unable to give informed consent then this may be sought from the patient's legal representative, usually a close relative. Standard of care procedures such as laboratory evaluations conducted prior to screening may be used rather than repeating the test.

The following assessments will be performed, as far as possible, prior to surgery (Day 1):

- Informed consent
- Medical history
- Prior and concomitant medication
- Full physical examination
- Evaluation of inclusion/exclusion criteria
- Reasons for not using an autologous venous conduit
- Laboratory testing (or standard pre-op lab profile for the institution)
    - Hematology: full blood count and differential
    - Clinical chemistry ; sodium, potassium, calcium, blood urea nitrogen, creatinine, albumin, total bilirubin, glucose (non-fasting)
    - PRA

- Ultrasound or CT angiography (CTA) (pre-op imaging is at the discretion of the investigator based on the clinical condition of the patient. It may be omitted if not deemed necessary in order to proceed to vascular surgical repair

8.5.2. Enrollment – Day 1 (HAV Implantation)

The HAV will be implanted as an interposition replacement or bypass in the required location using standard vascular surgical techniques. Details of the surgical anatomy and any complications will be documented. Operative procedures note and surgical diagram will be uploaded into the EDC for medical monitor review. Determination of intraoperative HAV patency is required by physical examination, Doppler exam, angiography (conventional or intra-op CT angiography) , or ultrasound at the discretion of the investigator.

8.5.3. Follow-up Visits Day 5 through Month 12

Day 5 (or prior to hospital discharge if earlier)

- Concomitant Medication
- Physical exam including surgical site
- Vital signs (heart rate, blood pressure and temperature)
- Documentation of any HAV interventions
- Adverse events
- Laboratory assessments (clinical chemistry, hematology)

Day 30 (+ 5 days)

- Concomitant medication
- Physical exam including surgical site
- Duplex ultrasound of the HAV
- Documentation of any HAV interventions
- Adverse events
- PRA

Months 3, 6 and 9 (+/- 14 days)

- Concomitant Medication
- Physical exam including surgical site
- Duplex ultrasound of the HAV
- Documentation of any HAV interventions

- Adverse events
- PRA (Month 6 only)

Month 12 (+/-14 days) and Early Termination

- Concomitant Medication
- Full physical exam including surgical site
- Duplex ultrasound of the HAV
- Documentation of any HAV interventions
- Adverse events
- PRA (only at early termination if prior to 6 month PRA collection)
- CT angiography

8.5.4. Long Term Follow-up Post Month 12 through Month 36 (+/- 30 days)

The status of the patient and HAV will be ascertained every 3 months from post Month 12 through 36 months after HAV implantation.

- Quarterly questionnaire covering status of the patient via a telephone contact with the patient and/or physician. If a suspected SAE related to HAV is discovered an unscheduled visit should be conducted to investigate.
- Documentation of any HAV interventions
- Adverse events (all AESI and related SAEs to be reported)
- Physical exam including surgical site at Month 24 and Month 36
- Duplex ultrasound at Month 24 and Month 36

8.5.5. Early Termination Visit

The subject may withdraw from the study at any time at his/her own discretion. The treating physician may also withdraw the subject for safety reasons. If withdrawal occurs before 12 months the subject will be asked to complete an early termination visit at which all assessments normally performed at 12 months will be completed. PRA will be collected at ET visit if the visit occurs before Month 6 collection of the sample. If withdrawal occurs after Month 12 and prior to Month 36 the patient will be asked to complete an early termination visit at which all assessments normally conducted during the long term follow up visits will be completed.

The reasons for early termination should be recorded in the eCRF.

With the exception of patient withdraw, all subjects will be followed for 12 months from HAV implantation (or until HAV removal or death if earlier). The subject should be withdrawn from the

study if the HAV is completely removed or the HAV becomes permanently occluded (loss of secondary patency) after Month 12.

8.5.6. Unscheduled Visits

If necessary to evaluate adverse events or HAV complications additional visits may be scheduled at the discretion of the investigator. At a minimum HAV status on clinical examination and Duplex ultrasound and adverse events will be recorded.

If, at any of the scheduled visits, duplex ultrasound surveillance suggests the development of a ≥ 50% stenosis within the HAV but immediate intervention is not required closer follow up should be considered. Intervention to manage any such stenosis is at the discretion of the investigator taking into account the degree and rate of progression of the stenosis.

8.6. Medical Care during the Study and upon Study Termination

Optimal medical therapy should be continued during the study. This should include:

- Antiplatelet therapy (see section 7.2.1)

After the final study visit at month 36 patients will not receive any further study-specific treatment. They will be treated by their medical doctor in a way that is appropriate for them.

8.7. Histological Examination of Resected HAV Material

If all or part of the HAV is resected it should, wherever possible, be retained for future histological examination. Instructions for preservation, storage and shipping of this material will be provided separately in a procedures manual. If a patient dies with an HAV in situ and it is feasible to obtain a fresh post mortem sample of the bypass this should be attempted in accordance with local regulations.

9. SAFETY ASSESSMENTS AND ADVERSE EVENTS

Safety of the HAV will be assessed in terms of:

- Aneurysm formation
- Pseudoaneurysm formation
- Anastomotic bleeding or spontaneous rupture
- HAV infection
- Need for HAV removal
- Inflammation at the implantation site
- Other adverse events
- Increase from baseline in PRA

9.1. Adverse Event Definition

An AE is any untoward medical occurrence in a patient administered an IMP and which does not necessarily have a causal relationship with the IMP. An AE can, therefore, be any unfavorable and unintended sign, symptom, or disease temporally associated with the use of an IMP, whether or not related to the IMP. Any worsening of the patient's disease under study or other medical conditions will also be considered to be an AE, unless it is within the normal range of disease fluctuation for that patient.

9.2. Serious Adverse Event Definition

An AE is considered "serious" if, in the view of either the investigator or Sponsor, it:

- Results in death
- Is life-threatening
    - The term "life-threatening" in the definition of "serious" refers to an event in which the patient was at immediate risk of death at the time of the event; it does not include an adverse event that had it occurred in a more severe form, might have caused death.
- Requires patient hospitalization or prolongation of existing hospitalization
    - This is defined as the patient being hospitalized for 24 hours or more or the patient's hospital stay being prolonged for at least an additional overnight stay.
- Requires intervention to prevent permanent damage
- Results in persistent or significant disability/incapacity

- Is a congenital anomaly/birth defect
- Important Medical Events
    - For the purpose of this study, this includes any event involving the HAV that results in a surgical or endovascular radiological intervention. The event(s) which caused the procedure should be reported as an SAE. For example: in the event of HAV thrombosis, the thrombosis would be considered the SAE; any associated stenoses (or other associated findings) that are present would be considered AEs.

Medical and scientific judgment should be exercised in deciding whether expedited reporting to the sponsor is appropriate in other situations, such as important medical events that may not be immediately life-threatening or result in death or hospitalization but may jeopardize the patient or may require medical or surgical intervention to prevent one of the other outcomes listed in the definition above. These should also be considered to be SAEs.

Note: Hospitalization for the surgery to implant the HAV is not a SAE. However, prolongation of the initial hospitalization due to an AE will be considered a SAE.

9.3. Suspected Unexpected Serious Adverse Reaction

A suspected unexpected serious adverse reaction (SUSAR) is any adverse drug reaction that is serious (as defined in 9.2), unexpected (is not listed in the IB or is not listed at the specificity or severity that has been observed) and suspected (meaning there is a reasonable possibility that the IMP caused the adverse event).

9.4. Events of Special Interest

Events of Special Interest are:

- HAV occlusion (thrombosis)
- HAV spontaneous rupture
    - Iatrogenic injuries are not an Event of Special Interest and should be reported as an AE
- HAV infection
- HAV abandonment
- HAV aneurysm
- HAV pseudoaneurysm
- HAV Excision (partial or complete)

9.5. Reporting of Adverse Events

At each evaluation, the investigator will determine whether any AEs have occurred. The patient will be questioned in a general way and no specific symptoms will be suggested. If any AEs have occurred they should be documented in the patient's medical chart and recorded on the AE pages of the eCRF. If known, the diagnosis should be recorded in preference to the listing of individual signs and symptoms. All SAEs, should be reported to the Safety CRO within 24 hours from the time the investigator or study personnel first become aware of the event.

AE reporting begins from time of anesthesia induction for implantation of the HAV and ends at the conclusion of the Month 12 visit or ET visit, unless an unresolved AE is still being followed.

During the long term follow up period from post Month 12 through Month 36, only the following will be reported by the investigator:

- All SAEs considered related to the HAV
- All Events of Special Interest (Section 9.4)

9.5.1. Criteria for Determining Causal Relationship to the HAV and Criteria for Determining Causal Relationship to the Index Surgical Procedure

The criteria for determining the causal relationship of an AE with the HAV are presented in the table below. A separate assessment of causal relationship of an AE to the index surgical procedure is required as well using the same criteria and definitions presented in the table below. Please note that causal relationship to procedure only refers to the index surgical procedure in which the HAV was initially implanted.

Causal Relationship to the IP	**Criteria for Determining Causal Relationship**
Definitely Related	There is clear evidence to suggest a causal relationship, and other possible contributing factors can be ruled out. The clinical event, including an abnormal laboratory test result, occurs in a plausible time relationship to surgical placement of the HAV and cannot be explained by concurrent disease or other devices, drugs, or chemicals.
Possibly Related	There is some evidence to suggest a causal relationship (e.g., the event occurred within a reasonable time after the surgical placement of the HAV). However, the influence of other factors may have contributed to the event (e.g., the subject's clinical condition, other concomitant medications). Although an adverse event may rate only as "possible" soon after discovery, it can be flagged as requiring more information and later be upgraded to certain as appropriate.

Unlikely Related	A clinical event, including an abnormal laboratory test result, whose temporal relationship makes a causal relationship improbable (e.g., the event did not occur within a reasonable time after surgical placement of the HAV) and in which other drugs or chemicals or underlying disease provide plausible explanations (e.g., the subject's clinical condition, other concomitant treatments).
Not Related	A clinical event, including an abnormal laboratory test result, which occurs when the HAV was not implanted; or, another cause is obvious and in which there is sufficient information that the etiology of the event is not related to the HAV.

The sponsor will make the final determination of causality for the purposes of reporting to the regulatory authorities and to the Principal Investigators.

9.5.2. Criteria for Defining the Severity of an Adverse Event

Severity of adverse events, including abnormal clinical laboratory values, will be assessed according to the criteria below and entered in the eCRF:

Grade	**Severity Assessment Standard**
1-Mild	Events require minimal or no treatment and do not interfere with the subject's daily activities.
2-Moderate	Events result in a low level of inconvenience or concern with the therapeutic measures. Moderate events may cause some interference with functioning.
3-Severe	Events interrupt a subject's usual daily activity and may require systemic drug therapy or other treatment. Severe events are usually incapacitating.
4-Life-threatening	Any adverse event that places the subject or participant, in the view of the investigator, at immediate risk of death from the reaction as it occurred, i.e., it does not include a reaction that, had it occurred in a more severe form, might have caused death.
5-Death	Death related to AE.

9.5.3. Reporting of Action Taken to Resolve AE

- None

- Lab tests / further evaluation
- Treatment required (specify if hospitalized)
- Patient withdrawn from study
- Other (specify)

9.5.4. Reporting the Outcome of the AE

- Recovered, with sequelae
- Recovered, without sequelae
- Ongoing
- Death
- Lost to follow-up

9.5.5. Reporting Serious Adverse Events

The urgency for reporting SAEs is 4-fold: (1) to facilitate discussion (and implementation, if necessary) by the sponsor and the investigator of appropriate follow-up measures, (2) to facilitate investigator reporting of unanticipated problems involving risk to human subjects to the institutional review board (IRB), (3) to facilitate the sponsor's rapid dissemination of information regarding AEs to other investigators/sites in a multi-center study, and (4) to enable the sponsor to fulfill the reporting requirements to the appropriate regulatory authority.

Any SAE that occurs through Month 12, whether or not causally related to the IMP, must be reported by the investigator or designee to the Safety CRO within 24 hours of learning of its occurrence. This applies also to any AE that could affect the safety of the study participants or the conduct of the study. Any SAE that occurs during long term follow up post Month 12 through Month 36 that is causally related to the IMP, must be reported by the investigator or designee to the Safety CRO within 24 hours of learning of its occurrence.

Information about an SAE will be collected and recorded on the SAE Report Form. The investigator must assess the relationship to the investigational product and any relevant procedure.

The investigators must complete the SAE Report Form in English, and **send the completed, signed form by fax or email (see below) IMMEDIATELY (at latest within 24 hours) after becoming aware of the SAE.**

Copies of relevant medical records (e.g., admission and/or discharge summary, laboratory reports and autopsy report), may also be submitted with the SAE form to clarify the circumstances surrounding the SAE(s). The entire medical records should **NOT** be sent with the SAE form.

Atlantic Research Group, Inc.

Drug Safety Department

Fax: +1-866-851-9318

Email: Safety@atlanticresearchgroup.com

Phone: +1-888-619-3216

The investigator will be requested to supply as much detailed information as possible regarding the SAE that is available at the time of the initial contact. The investigator should also complete missing or requested information and submit follow-up reports until the SAE has resolved or, in the case of permanent impairment, until the SAE has stabilized.

It is the responsibility of each Principal Investigator to promptly notify his/her IRB of all SAEs that are received by the Sponsor or designee and that occur at his/her institution in accordance with institutional practices.

The Safety CRO will inform the sponsor about all SAEs within 1 business day after receipt of the respective report from the investigator.

9.5.6. Reporting of Events of Special Interest

Events of Special Interest are defined in Section 8.4 and should be reported to the Safety CRO within 24 hours of learning of its occurrence. For each of these events detailed surgical notes (with illustrative diagram), including reason for and outcome of any intervention or abandonment, should be completed within 48 hours and uploaded to the clinical database.

Detailed information about the occurrence and treatment/intervention for these events will be collected throughout the study up to 3 years post HAV implant. This information will include the following:

- Summarized surgical notes, including a simplified anatomical diagram showing where angioplasties, stents, or revisions have been performed (using intervention worksheet provided)
- Need for hospitalization (number of nights)
- Need for antibiotics (in the case of HAV-related infections)

9.5.7. Follow-Up of Adverse Events

If any AEs are present when a subject completes 1 year post implant (Month 12) or ET, if earlier, or if a subject is withdrawn from the study, the subject will be re-evaluated within an appropriate period of time. At the investigator's discretion, minor AEs can be re-evaluated via telephone and documented. If the AE has still not resolved, additional follow-up will be performed as appropriate. The investigator or his designee should make every effort to contact the subject until the AE has resolved or stabilized or the medical monitor and investigator agree

that further follow-up is not necessary. This should be documented in the subject's medical records.

9.6. Reporting of Pregnancy

If a study participant becomes pregnant during study participation, basic information about the pregnancy will be recorded in the Pregnancy eCRF and the Pregnancy Outcome and Report Form, and submitted to the Safety CRO. If there are complications during the pregnancy, the complications are recorded as AEs. The participant will be asked to report the outcome of the pregnancy and the site should submit the information to the Safety CRO within 30 days after the outcome of the pregnancy. If there is a congenital anomaly in the infant, this will be recorded as a SAE in the data forms for the mother (i.e., the study participant).

Partner pregnancies do not need to be reported.

9.7. Data Monitoring Committee

A Data Monitoring Committee (DMC) will review safety on an ongoing basis and provide recommendations about stopping, continuing or otherwise modifying the study. The DMC consists of individuals who are not directly involved in the conduct of the study. A charter describes the roles and responsibilities of the DMC. Responsibilities of the DMC will include review of aggregate safety data from other studies in the HAV clinical development program.

The DMC will at a minimum meet every 6 months from the date of initial enrollment of the first subject. Additionally, the DMC will review the safety data of the torso cohort on the earlier of when the first torso subject reaches 3 months post-implantation or when the first 2 torso subjects have both reached 30 days post-implantation.

9.8. Interim Analysis and Stopping Criteria

This is a phase 2 study with no formal interim analysis. Periodic reviews of safety data will be undertaken by the DMC with particular attention to events that might indicate structural failure of the HAV. Events that might have implications for already implanted HAVs and their possible removal - such as aneurysm formation (true or pseudo) or spontaneous rupture -would trigger an urgent review of the safety data for DMC review.

The DMC may recommend modification or early termination of the study for safety reasons.

10. STATISTICAL CONSIDERATIONS

This is a prospective, open label, multi-cohort, multicenter pilot study to evaluate the safety and efficacy of the HAV in patients undergoing vascular replacement or reconstruction. The primary objective of this study is to evaluate the safety and tolerability of the HAV in these patients and to determine the primary patency of the HAV at 30 days post-implantation. The secondary objectives of this study are to further assess safety in terms of adverse events of special interest, to determine the rate interventions required to keep the HAV patent, and to further characterize efficacy in terms of secondary patency and limb salvage. There is no formal hypothesis testing planned.

Endpoints will be assessed over a period of up to 36 months after HAV implantation. The primary analysis of the study will be conducted on the earlier of a) when the last subject enrolled reaches 30 days post-implant or b) all subjects enrolled in the initial 24 month accrual period have reached 30 days post-implant. Details of data handling and planned descriptive statistics are given in the Statistical Analysis Plan.

10.1. Analysis Population

All patients who receive an HAV will be included in the analyses. For discontinued or withdrawn patients, all available data will be included in the safety and efficacy analyses.

10.2. Safety Analyses

Safety analyses will be performed on all patients who have an HAV implanted.

The incidence of aneurysm formation, anastomotic bleeding or spontaneous rupture, HAV removal, HAV infection, and inflammation at the implantation site will be tabulated by visit and overall.

Adverse events will be coded using Medical Dictionary for Regulatory Activities (MedDRA) terms. Adverse events will be listed and summarized by body system, incidence, severity, and duration. HAV complications will be listed in terms of incidence, severity, and (where appropriate) time to onset and duration. Serious adverse events will be summarized separately. Any premature discontinuations due to adverse events and deaths will be listed and summarized.

PRA data will be listed and summarized using appropriate descriptive statistics for the change from baseline values.

10.3. Efficacy Analyses

The primary efficacy analysis will be the rate of primary patency at 30 days after HAV implantation. Primary, primary assisted, and secondary patency rates of the HAV at 12 months and at all other post-surgery visits with evaluation of patency will be described. The rate of limb salvage and patient survival at 12 months will also be described.

Primary patency is defined as the functional access patency until any type of intervention; primary assisted patency is defined as an HAV still working without thrombosis; secondary patency is defined as the functional HAV patency, with or without preceding successful interventional or surgical procedures to maintain or reestablish patency, until the HAV is abandoned. Early discontinued patients prior to the visit of interest will be determined as being non-patent irrespective of the reason for discontinuation.

The rate and type of interventions needed to maintain / restore patency in the HAV will be descriptively tabulated.

The absolute change from baseline (Day 1) values to all post-surgery visits of duplex ultrasound parameters will be summarized. Summary statistics will also be provided at each time point.

The methods and endpoints regarding the efficacy parameters employed in this study are consistent with current clinical practice and are meaningful to the research community. Every attempt has been made to minimize the variability on the part of the surgeon when using this product.

10.4. Other Analyses

All clinical parameters will be listed for all patients treated at each study visit. Descriptive statistics will be summarized for continuous outcomes such as age and BMI. If necessary, number and percentage of patients will be reported for categorical outcomes.

10.5. Sample Size Rationale

Up to 100 subjects will be recruited into the study. As this phase 2 study is the first human study of the HAV for vascular trauma, the study was designed to provide preliminary evidence of safety and efficacy.

10.6. Interim analyses

There is no formal interim analysis.

11. STUDY MANAGEMENT AND DATA COLLECTION

11.1. Ethical Conduct of the Trial

This study will be conducted according to the protocol; 21 CFR Parts 11, 50, 54, 56, and 312; the World Medical Association Declaration of Helsinki and Good Clinical Practice (GCP). Each Investigator will conduct the trial according to applicable local or regional regulatory requirements.

11.2. Institutional Review Board

IRBs must be constituted according to the applicable state and federal requirements, including ICH GCP.

It is the responsibility of each investigator to submit the protocol, Investigator's Brochure, subject informed consent, subject recruitment materials (if applicable), and other documentation as required by the IRB to his/her IRB for review and approval. A copy of the written approval must be provided to the contract research organization (CRO). The documentation should clearly mention the approval/favorable opinion of the protocol, the subject informed consent form, and subject recruitment materials (if applicable), including respective version dates. The written approval and a list of members, their titles or occupations, and their institutional affiliations may be obtained from the IRBs if available, and provided to the CRO prior to the release of clinical study supplies to the investigational site and commencement of the study. If any member of the IRB has direct participation in this trial, written notification regarding his or her abstinence from voting must also be obtained.

Each investigator must adhere to all requirements stipulated by his/her respective IRB. This includes notification to the IRB regarding protocol amendments, updates to the subject informed consent, recruitment materials intended for viewing by subjects, investigational new drug safety reports, SAEs and unexpected AEs, reports and updates regarding the ongoing review of the trial at intervals specified by the respective IRB, and submission of final study reports and summaries to the IRB.

11.3. Subject Informed Consent

Prior to any study procedures being performed, subjects and persons conducting the consent discussion will be required to sign and date the IRB-approved informed consent, and each subject will be given a copy. In addition, this information should be recorded in the subject's medical record (i.e., source document). If the patient is unable to give informed consent then this may be sought from the patient's legal representative, usually a close relative.

The written consent document will embody the elements of informed consent as described in the World Medical Association Declaration of Helsinki, 21 CFR Part 50.25, ICH E6 guideline (GCP), and in accordance with any local regulations. The investigator is responsible for the

preparation, content, and IRB approval of the informed consent document. The consent form must be approved by the site's IRB and be acceptable to Humacyte.

The consent form must be written in a language fully comprehensible to the prospective subject. The investigator or designee shall give the subject adequate opportunity to read it before it is signed and dated. Information should be given in both oral and written form whenever possible and in the manner deemed appropriate by the IRB. Subjects must be given ample opportunity to inquire about details of the study.

11.4. Amendments to the Protocol

An amendment must be agreed to in writing by Humacyte and submitted to the FDA and approved by t IRBs before the amendment can be implemented. Written approval of a protocol amendment is not required prior to implementation of changes to the protocol which eliminate an immediate hazard to the study patient; however, approval must be obtained as soon as possible thereafter. Any agreed amendments must also be signed by the investigator.

11.5. Study Initiation

The investigator must not enroll any patients prior to attendance at the Investigator Meeting or the completion of a formal site initiation visit conducted by the CRO. These meetings will include a detailed review of the study protocol and eCRF pages. The investigator will not be supplied with IMP until all necessary pre-study requirements have been completed and essential signed documents provided to the CRO.

11.6. Study Monitoring

It is the responsibility of the investigator to ensure that the study is conducted in accordance with the protocol, GCP, applicable regulatory requirements, and the currently approved Declaration of Helsinki, and that valid data are entered into the eCRF.

To achieve this objective, the monitor's duties are to ensure the maintenance of complete, legible, well-organized, and easily retrievable data. The monitor will review the protocol with the investigator. In addition, the monitor will explain the investigator's reporting responsibilities and all applicable regulations concerning the clinical evaluation of the IMP.

The investigator will permit representatives of Humacyte and the CRO to monitor the study as frequently as Humacyte or the CRO deem necessary to determine that data recording and protocol adherence are satisfactory. The eCRF data and related source documents will be reviewed in detail by the monitor at each visit, in accordance with relevant SOPs and ICH GCP regulations. This includes results of tests performed as a requirement for participation in this study and any other medical records required to confirm information contained in the eCRF such as past medical history and secondary diagnoses. The investigator and his/her staff will be expected to cooperate with the monitor and provide any missing information whenever possible.

All monitoring activities will be reported and archived. In addition, monitoring visits will be documented at the investigational site by signature and date on the study-specific monitoring log.

11.7. Case Report Form

An electronic CRF will be used for this study. The data will be entered into the eCRF in a timely manner on an ongoing basis.

The investigator is responsible for ensuring that data are properly recorded on each patient's eCRF and related documents. An investigator who has signed the protocol signature page should personally sign the eCRFs in accordance with the procedure described in the eCRF completion guidelines to ensure that the observations and findings are correct and complete.

For data handled by the CRO, eCRF data and some or all of the study-related data will be managed and stored electronically in the CRO's database system. Validated data will subsequently be transferred to the sponsor.

11.8. Verification Procedures

It is the investigator's obligation to ensure documentation of all relevant data in the subject's medical record. The subject's medical record will be considered the source document. The eCRF should not be used as the source for study information.

The investigator will maintain a subject identification code list to enable unambiguous identification of the subjects (subject names and corresponding subject numbers). The subject identification code list is an essential document and as such should be maintained according to the ICH GCP guidelines.

11.9. Retention of Records

All documentation pertaining to the study will be kept by Humacyte or their designee in accordance with ICH guidelines and US FDA regulations.

The investigator will maintain a study file, which should be used to file the Investigator's Brochure, protocol, and IMP records; correspondence with the IRB and Humacyte; and other study-related documents.

The investigator agrees to keep records and those documents that include (but are not limited to) the identification of all participating subjects, medical records, study-specific source documents, source worksheets, all original signed and dated informed consent forms, query responses, and detailed records of drug disposition to enable evaluations or audits from regulatory authorities and Humacyte or its designees.

The investigator shall retain records required to be maintained under this part for a period of 2 years following the date a marketing application is approved for the IMP for the indication for which it is being investigated; or, if no application is to be filed or if the application is not

approved for such indication, until 5 years after the investigation is discontinued. However, these documents should be retained for a longer period if required by the applicable regulatory requirement(s) or if needed by the sponsor. In addition, the investigator must make provision for the subject's medical records to be kept for the same period of time. No data should be destroyed without the agreement of Humacyte. Humacyte will inform the investigator in writing when the trial-related records are no longer needed. Subject's medical records and other original data will be archived in accordance with the archiving regulations or facilities of the study site.

11.10. Protocol Deviations

A protocol deviation is any noncompliance with the protocol or GCP requirements. The noncompliance may be either on the part of the participant, the investigator, or the study site staff. As a result of deviations, corrective actions are to be developed by the site and implemented promptly. Although in principle, protocol deviations are not permitted, under emergency circumstances, deviations may proceed without prior approval of the sponsor and the IRB to protect the rights, safety, and well-being of human subjects.

All protocol deviations will be documented and reported by the CRO during the course of the study in the Monitoring Reports. All deviations will be reported to the sponsor who will agree on the necessary actions to be taken.

If required per their guidelines, reports about protocol deviations must be reported to the local IRB.

11.11. Insurance and Indemnity

Insurance coverage for damages emerging from the study will be provided according to applicable legal requirements. During the informed consent procedure, the investigator must inform the patient accordingly.

11.12. Audit

It is the responsibility of CRO and Humacyte to perform auditing (if applicable) as part of implementing quality assurance. The purpose of an audit, which is independent of and separate from routine monitoring or quality control functions, is to evaluate trial conduct and compliance with the protocol, SOPs, GCPs, and the applicable regulatory requirements. The auditor and regulatory authorities will require authority from the investigator to have direct access to the subject's medical records.

12. REPORTING

Following completion of follow-up of all patients to the 12-month endpoint, the results will be evaluated by Humacyte or a designee for clinically meaningful findings. A clinical study report will be generated, including a summary of all available data, statistical measures, tabulated results, graphical results and interpretations. This report will be submitted to regulatory authorities in a timely manner. An addendum to the report will be generated to include data up to 36 months follow-up. This addendum will be submitted to regulatory authorities in a timely manner.

13. QUALITY CONTROL AND QUALITY ASSURANCE

Following written standard operating procedures, the monitors will verify that the clinical trial is conducted and data are generated, documented (recorded), and reported in compliance with the protocol, GCP, and the applicable regulatory requirements. Reports of monitoring activities will be submitted to Humacyte in a timely manner.

The investigational site will provide direct access to all trial related areas, source data/documents, and reports for the purpose of monitoring and auditing by the sponsor, and inspection by local and regulatory authorities.

Quality control procedures will be implemented for data entry and the generation of data quality control checks and will be run on the database. Any missing data or data anomalies will be communicated to the site(s) for clarification/resolution.

14. RESPONSIBILITIES OF THE PRINCIPAL INVESTIGATOR

The role of the principal investigator is to implement and manage the day-to-day conduct of the clinical investigation as well as to ensure data integrity and the rights, safety, and well-being of the patients involved in the clinical investigation.

14.1. Informed Consent

The principal investigator shall ensure that the process for obtaining informed consent

- includes all aspects of the clinical investigation that are relevant to the patient's decision to participate throughout the clinical investigation,
- avoids any coercion or undue improper influence on, or inducement of, the patient to participate,
- does not waive or appear to waive the patient's legal rights,
- uses native non-technical language that is understandable to the patient,
- provides ample time for the patient to read and understand the informed consent form and to consider participation in the clinical investigation,
- provides the patient with a copy of the signed and dated informed consent form and any other written information.

The principal investigator shall ensure and document appropriate training if an authorized designee is appointed to conduct the informed consent process.

14.2. Compliance with the Protocol

The principal investigator shall:

- indicate his/her acceptance of the protocol in writing
- conduct the clinical investigation in compliance with the protocol
- create and maintain source documents throughout the clinical investigation and make them available as requested during monitoring visits or audits
- ensure that the IMP is used solely by authorized users, and in accordance with the protocol and instructions for use
- propose to the sponsor any appropriate modification(s) of the protocol
- refrain from implementing any modifications to the protocol without agreement from the sponsor, IRB, and, if required, regulatory authorities
- document and explain any deviation from the approved protocol that occurred during the course of the clinical investigation

- ensure that an adequate investigation site team and facilities exist and are maintained and documented during the clinical investigation
- ensure that maintenance and calibration of the equipment relevant for the assessment of the clinical investigation is appropriately performed and documented, where applicable
- ensure the accuracy, completeness, legibility, and timeliness of the data reported to the sponsor in the eCRFs and in all required reports
- maintain the clinical trial material accountability records
- allow and support the sponsor to perform monitoring and auditing activities
- be accessible to the monitor and respond to questions during monitoring visits
- allow and support regulatory authorities and the IRB when performing auditing activities
- ensure that all clinical-investigation-related records are retained as specified in this protocol.

14.3. Medical Care of Patients

The principal investigator shall:

- provide adequate medical care to a patient during and after a patient's participation in a clinical investigation in the case of AEs
- inform the patient of the nature and possible cause of any adverse events experienced
- inform the patient of any new significant findings occurring during the clinical investigation, including the need for additional medical care that may be required
- provide the patient with well-defined procedures for possible emergency situations related to the clinical investigation, and make the necessary arrangements for emergency treatment,
- ensure that clinical records are clearly marked to indicate that the patient is enrolled in a particular clinical investigation
- inform, with the patient’s approval or when required by national regulations, the patient's personal physician about the patient's participation in the clinical investigation
- make all reasonable efforts to ascertain the reason(s) for a patient's premature withdrawal from the clinical investigation while fully respecting the patient's rights.

14.4. Safety Reporting

The principal investigator shall:

- record every adverse event together with an assessment, in accordance with Section 9 of this protocol,
- report to the sponsor, without unjustified delay, all serious adverse events and medically significant events as specified in Section 9 of this protocol,
- supply the sponsor, upon sponsor's request, with any additional information related to the safety reporting of a particular event.

15. SUSPENSION OR PREMATURE TERMINATION OF THE CLINICAL INVESTIGATION

The sponsor may suspend or prematurely terminate either a clinical investigation in an individual investigation site or the entire clinical investigation for significant and documented reasons.

A principal investigator, IRB, or regulatory authority may suspend or prematurely terminate participation in a clinical investigation at the investigation sites for which they are responsible.

If suspicion of an unacceptable risk to patients arises during the clinical investigation, or when so instructed by the IRB or regulatory authorities, the sponsor shall suspend the clinical investigation while the risk is assessed. The sponsor shall terminate the clinical investigation if an unacceptable risk is confirmed.

The sponsor shall consider terminating or suspending the participation of a particular investigation site or investigator in the clinical investigation if monitoring or auditing identifies serious or repeated deviations on the part of an investigator.

If suspension or premature termination occurs, the terminating party shall justify its decision in writing and promptly inform the other parties with whom they are in direct communication.

If, for any reason, the sponsor suspends or prematurely terminates the investigation at an individual investigation site, the sponsor shall inform the responsible regulatory authority if required and ensure that the IRB is notified. If the suspension or premature termination was in the interest of safety, the sponsor shall inform all other principal investigators.

If suspension or premature termination occurs,

1. the sponsor shall remain responsible for providing resources to fulfill the obligations from the protocol and existing agreements for following up the patients enrolled in the clinical investigation, and
2. the principal investigator or authorized designee shall promptly inform the enrolled patients at his/her investigation site, if appropriate.

In the event that the study is discontinued, the reasons for discontinuation will be explained to the investigators and may be disclosed to the study participants. Humacyte will provide all information needed by the investigator to ensure the safety and well-being of the study participants.

16. PUBLICATION POLICY

A Publication Committee comprising the Principal Investigator from each participating site and a representative of Humacyte will oversee all publication of data from this study. Prior to submitting for publication, presenting, using for instructional purposes, or otherwise disclosing the results of the study, the investigator agrees to allow the Publication Committee and Humacyte a period of at least 30 days (or, for abstracts, at least 5 calendar days) to review the proposed publication or disclosure prior to its submission for publication or other disclosure. Publications or disclosures of study results shall not include other confidential information belonging to Humacyte. If the proposed publication/disclosure risks Humacyte's ability to patent any invention related to the study, the publication or disclosure will be modified or delayed, at the investigator's option, a sufficient time to allow Humacyte to seek patent protection of the invention. For multicenter studies, the first publication or disclosure shall be a complete, joint multicenter publication or disclosure. This statement does not give Humacyte any editorial rights over the content of a publication or disclosure, other than to restrict the disclosure of Humacyte's confidential information. If a written contract for the conduct of the study is executed which includes publication provisions inconsistent with this statement, then that contract's publication provisions shall apply rather than this statement.

17. REFERENCES

Akingba G, Rapp BM, Tsai A, Jester AL, Fajardo A, Motaganahali R, et al. Surgical Management of Vascular Trauma From Dog Bites: The 10- year Experience of a Level I Trauma Center. Journal of Vascular Surgery. 2012;56(4):1190.

Andrikopoulos V, Antoniou I, Panoussis P. Arterial injuries associated with lower-extremity fractures. Cardiovascular Surgery. 1995;3(1):15-8.

Cordesman AH, Lemieux, J. IED Metrics for Afghanistan. January 2004-May 2010. Center for Strategic & International Studies, 2010.

Dahl, SL, Kypson, AP, Lawson, JH et al. Readily available tissue-engineered vascular grafts. Sci Transl Med. 2011: 68ra9

Dixon BS, Beck GJ, Vazquez MA et al. Effect of dipyridamole plus aspirin on hemodialysis graft patency. N Engl J Med 2009;360:2191-201.

DuBose JJ, Savage SA, Fabian TC, Menaker J, Scalea T, Holcomb JB, Skarupa D, Poulin N, Chourliaras K, Inaba K, Rasmussen TE. The American Association for the Surgery of Trauma PROspective Observational Vascular Injury Treatment (PROOVIT) registry: multicenter data on modern vascular injury diagnosis, management, and outcomes. Journal of Trauma and Acute Care Surgery. 2015 Feb 1;78(2):215-23.

Faulconer ER, Branco BC, Loja MN, Grayson K, Sampson J, Fabian TC, Holcomb JB, Scalea T, Skarupa D, Inaba K, Poulin N. Use of open and endovascular surgical techniques to manage vascular injuries in the trauma setting: A review of the American Association for the Surgery of Trauma PROspective Observational Vascular Injury Trial registry. Journal of Trauma and Acute Care Surgery. 2018 Mar 1;84(3):411-7.

Helfet DL, Howey T, Sanders R, Johansen K. Limb Salvage Versus Amputation: Preliminary Results of the Mangled Extremity Severity Score. Clinical Orthopaedics & Related Research. 1990;256:80-6.

Holcomb J. Devastating Dismounted IED Injuries in OEF. Increasing amputation and genital injury rates admitted to LRMC 2009-2010. 2011. Presentation to the Defence Health Board, (http://www.health.mil/Reference-Center/Presentations/2011/03/08/Devastating-Dismounted-Improvised-Explosive-Device-Injuries-in-Operation-Enduring-Freedom)

Kelly BS, Heffelfinger SC, Whiting JF, et al. Aggressive venous neointimal hyperplasia in a pig model of arteriovenous graft stenosis. Kindey Int. 2002;62(6):2272-2280.

Kirkton RD, Prichard HL, Santiago-Maysonet M, Niklason LE, Lawson JH, Dahl SL. Susceptibility of ePTFE vascular grafts and bioengineered human acellular vessels to infection. Journal of Surgical Research. 2018 Jan 1;221:143-51.

L'Heureux N, Dusserre N, Konig G, et al. Human tissue-engineered blood vessels for adult arterial revascularization. Nat Med 2006; 12(3):361-365.

Mills JL et al on behalf of the Society for Vascular Surgery Lower Extremity Guidelines Committee. The Society for Vascular Surgery Lower Extremity Threatened Limb Classification System: Risk stratification based on Wound, Ischemia, and foot Infection (WIfI). J Vasc Surg 2013,

Morgan MBF, Crawford T, Murrin B and Fraser SCA. Developing the vascular quality of life questionnaire: a new disease specific quality of life measure for use in lower limb ischemia. J Vasc Surg 2001; 33: 679 - 687

Prichard, H.L, Manson, R.J.,Dibernado, L. et al. An early study on the mechanisms that allow tissue-engineered vascular grafts to resist vascular hyperplasia. J Cardiovasc Transl Res 2011, 4, 674-682

Quint C, Kondo Y, Manson RJ,et al. Decellularized tissue-engineering blood vessel as an arterial conduit. Proc Natl Acad Sci USA. 108(22):9214-9, 2011.

Rasmussen TE. Vascular Injury Rates in the Wars in Iraq and Afghanistan: How are they different from the past? United States Army Institute of Surgical Research, 2010.

Roy-Chaudhury P, Kelly BS, Miller MA, et al. Venous neointimal hyperplasia in polytetrafluoroethylene dialysis grafts. Kidney Int. 2001;59(6):2325-2334.

Rutherford RB1, Baker JD, Ernst C, et al Recommended standards for reports dealing with lower extremity ischemia: revised version. J Vasc Surg. 1997 Sep;26(3):517-38.

APPENDIX 1: HAV CLINICAL VISIT SCHEDULE

	Pre-op screening D1	**D 1**	**D 5** or prior to d/c	**D 30** + 5 days	**M3** ± 14 days	**M 6** ± 14 days	**M 9±** 14 days	**M12 / ET†** ± 14 days	**M15-M36 †**+/- 30 days
Informed consent	X								
Medical history and nature of trauma	X								
Concomitant medication	X	X	X	X	X	X	X	X	
Physical exam [1]	X	X	X	X	X	X	X	X	X [7]
Pre-op Ultrasound or CT angiography [2]	X								
Vital signs			X						
Eligibility (inclusion/exclusion criteria)	X								
HAV implantation and intraoperative confirmation of patency [3]		X							
Documentation of surgery and any complications		X							
Clinical chemistry	X [5]		X						
Hematology	X [5]		X						
PRA	X			X		X		X [6]	
Duplex ultrasound [4]				X	X	X	X	X	X [7]
CT angiography								X	
AEs		X	X	X	X	X	X	X	X [8]
Documentation of HAV interventions		X	X	X	X	X	X	X	X[8]

Abbreviations: AEs, adverse events; D, day; d/c, discharge; ET, early termination; HAV, human acellular vessel; M, month

1. Physical examination includes clinical exam of the operative limb and HAV at all post-operative visits (incl. patency assessment on D1) and physical exam to evaluate AEs; include distal vascular bed (limb cohort only)
2. Pre-op imaging is at the discretion of the investigator based on the clinical condition of the patient. It may be omitted if not deemed necessary in order to proceed to vascular surgical repair
3. Determination of intraoperative HAV patency can be done by physical exam, Doppler, angiography or ultrasound at the discretion of the investigator
4. An alternative imaging method (CTA, MRI, etc.) may be substituted for duplex ultrasound at the discretion of the investigator if it is medically appropriate and in the best interest of the patient.
5. Measured at preoperative screening when possible
6. PRA only collected at ET visit if the visit occurs before Month 6 collection
7. Visits at month 24 and month 36 to be conducted in person with physical exam of the HAV site and duplex ultrasound imaging of HAV.
8. The status of the patient and HAV will be ascertained every 3 months post Month 12 until 36 months after HAV implantation by telephone contact with the patient and/or his physician. Only related SAEs and all AESI will be reported after 12 months. If a suspected SAE related to HAV is discovered an unscheduled visit should be conducted to investigate.

† Patients withdrawn before Month 12 will perform ET visit that correlates with the procedures at Month 12. Patients withdrawn after Month 12 and prior to Month 36 should complete an ET visit that correlates with procedures post Month 12 through Month 36.

SUMMARY OF PROTOCOL CHANGES

All changes since version 3.0 of the protocol and incorporated in the current version (version 3.2) are briefly described below. Each change is also categorized as either related to safety or efficacy (thus requiring IRB/EC review) or administrative in nature.

Changes Incorporated in Protocol Version 3.2 (18 Aug 2021)	
Change and Reason for Change	**Section(s) Changed**
Changes Related to Safety and/or Efficacy	
Change: The sample size was increased from 40 subjects to 100 subjects. Reason: To permit enrollment of a sufficient number of subjects to power for statistical analysis of safety and efficacy per FDA recommendation (see associated note to file from Sponsor)	Study Synopsis, Section 10.5 Sample Size Rationale
Administrative Changes	
Change: Changes to personnel Sponsor Medical Advisor is now Kiernan DeAngelis, MD Sponsor Clinical Operations Representative is now Karthi Natarajan Reason: Administrative change	Sponsor's Authorized Medical Advisor, Study Personnel

A Phase 2 Study for the Evaluation of Safety and Efficacy of Humacyte's Human Acellular Vessel for Vascular Replacement or Reconstruction in Patients with Life or Limb-threatening Vascular Trauma

Medicinal Product:	Humacyte Human Acellular Vessel (Humacyte HAV)
Study No.:	CLN-PRO-V005
Sponsor:	Humacyte, Inc
Address:	2525 East NC Highway 54 Durham, NC 27713
Phone:	919-313-9633
CRO:	Atlantic Research Group 2421 Ivy Road, Suite 200 Charlottesville, VA 22903
Version:	3.0 (Amendment to version 2.0 dated 24 October 2016) 18 July 2018

Confidentiality Statement

STATEMENT OF COMPLIANCE

This trial will be conducted in compliance with the protocol and the following regulatory requirements:

- Declaration of Helsinki adopted by the 18th World Medical Assembly in Helsinki, Finland, in 1964, as last amended by the World Medical Assembly in 2013
- International Conference on Harmonization of Technical Requirements for Registration of Pharmaceuticals for Human Use (ICH), E6 Good Clinical Practice: Consolidated Guidance (ICH E6)
- ICH E2A Clinical Safety Data Management: Definitions and Standards for Expedited Reporting
- ICH E8 Guidance on General Considerations for Clinical Trials
- Applicable sections of United States Food and Drug Administration (FDA) Code of Federal Regulations (CFR), including:
    - 21 CFR Part 11, Electronic Records; Electronic Signatures
    - 21 CFR Part 50, Protection of Human Subjects
    - 21 CFR Part 54, Financial Disclosure by Clinical Investigators
    - 21 CFR Part 56, Institutional Review Boards
    - 21 CFR Part 312, Investigational New Drug Application

PRINCIPAL INVESTIGATOR AGREEMENT PAGE FOR THE PROTOCOL

I agree:

- To assume responsibility for the proper conduct of the study at this site, and to conduct the study in compliance with this protocol, any future amendments, and with any other study conduct procedures provided by the sponsor, Humacyte, Incorporated (Humacyte), or their authorized representatives.

- Not to implement any deviations from or changes to the protocol (including protocol amendments) without agreement from the sponsor and prior review and written approval from the Institutional Review Board (and FDA, if applicable) except where necessary to eliminate an immediate hazard to the patient(s), or for administrative aspects of the study (where permitted by all applicable regulatory requirements).

- That I am familiar with the appropriate use of the investigational medicinal product, as described in this protocol and any other information provided by the sponsor including, but not limited to the current Investigator's Brochure or equivalent document provided by Humacyte.

- To ensure that all persons assisting me with the study are adequately informed about the investigational medicinal product and of their study-related duties and functions.

- That I have been informed that certain regulatory authorities require the sponsor to obtain and supply details about the investigator's ownership interest in the sponsor or the Investigational Medicinal Product, and more generally about his/her financial ties with the sponsor. Humacyte will use and disclose the information solely for the purpose of complying with regulatory requirements.

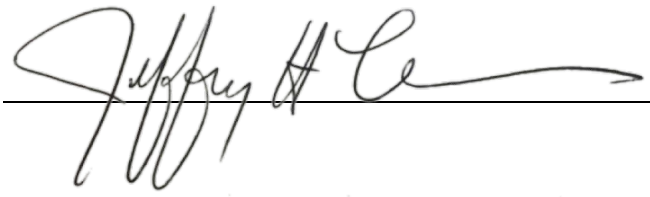
Principal Investigator: _____

Name and Title

Signed: _____ Date: _____

Protocol Approval

Sponsor Medical Approval: Jeffrey H. Lawson, M.D. Ph.D. Chief Medical Officer, Humacyte

Signed:  Date: 19JUL2018

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LIST OF ABBREVIATIONS

AE	Adverse event
AIDS	Acquired immunodeficiency syndrome
ALT	Alanine aminotransferase
aPTT	Activated partial thromboplastin time
ASA	Acetylsalicylic acid (aspirin)
ABI	Ankle brachial index
AST	Aspartate aminotransferase
AV	Arteriovenous
AVF	Autologous arteriovenous fistula
BP	Blood pressure
CAVG	Canine acellular vascular graft
CBC	Complete blood count
CKD	Chronic kidney disease
CTA	Computed tomography angiography
CM	Concomitant medication
eCRF	Electronic case report form
CRO	Contract research organization
DMC	Data Monitoring Committee
DTH	Delayed-type hypersensitivity
ECG	Electrocardiogram
ECM	Extracellular Matrix
ePTFE	Expanded polytetrafluoroethylene
ESRD	End-stage renal disease
ET	Early termination

LIST OF ABBREVIATIONS

FDA	Food and Drug Administration
GCP	Good Clinical Practice
GLP	Good Laboratory Practice
HAV	Human acellular vessel
HIV	Human immunodeficiency virus
IB	Investigator Brochure
ICF	Informed consent form
ICH	International Conference on Harmonization of Technical Requirements for Registration of Pharmaceuticals for Human Use
IgG	Immunoglobulin G
IHC	Immunohistochemistry
IM	Intramuscular
IMP	Investigational medicinal product
INR	International normalized ratio
IRB	Institutional Review Board
ISO	International Organization for Standardization
IU	International unit
IV	Intravenous
MedDRA	Medical Dictionary for Regulatory Activities
M	Month
N	Number (typically refers to participants)
NYHA	New York Heart Association
OTC	Over-the-counter
PAD	Peripheral arterial disease

LIST OF ABBREVIATIONS

PE	Physical examination
PHI	Protected health information
PI	Principal Investigator
PRA	Panel reactive antibodies
PT	Prothrombin time
PTFE	Polytetrafluoroethylene
QA	Quality Assurance
QC	Quality Control
RRT	Renal replacement therapy
SAE	Serious adverse event
SFA	Superficial Femoral Artery
SOP	Standard operating procedure
SVS WIfI	Society for Vascular Surgery: Wound, Ischemia, and foot Infection
US	Ultrasound
USA	United States of America
WFI	Water for injection
WBC	White blood cell(s)
WHO	World Health Organization

PROTOCOL SUMMARY

Full Title	A Phase 2 Study for the Evaluation of Safety and Efficacy of Humacyte's Human Acellular Vessel for Vascular Replacement or Reconstruction in Patients with Life or Limb-threatening Vascular Trauma
Clinical Trial Phase	Phase 2
Sponsor	Humacyte, Inc.
Planned Study Sites	Up to 10 sites in the United States
Sample Size	Up to 40 subjects with a minimum of 10 subjects in the limb cohort and 10 subjects in the torso cohort
Study Population	Patients with vascular trauma to size appropriate vessels in the limb or torso, requiring replacement or reconstruction.
Inclusion Criteria	1. Patients with life or limb threatening traumatic injury to an arterial vessel in the limb or torso, other than the heart, which requires replacement or reconstruction. 2. Preoperative imaging or clinical examination indicates the damaged vessel has a defect length of ≤ 38cm and is appropriately size matched to the 6mm Human Acellular Vessel (HAV) per the judgment of the treating surgeon taking into account vasoconstriction and situational inflow and outflow considerations. 3. Autologous vein graft is either not feasible in the judgment of the treating surgeon (e.g. because of lack of availability of suitable conduit, presence of severe venous insufficiency) or is not desirable because of the urgency of revascularization 4. Aged 18 to 85 years old, inclusive 5. Able to communicate meaningfully with investigative staff and able to comply with study procedures. If the patient is unconscious then information from a reliable witness indicates that the patient would normally be able to understand and comply with study procedures 6. Patient or legal representative is able, willing and competent to give informed consent 7. Life expectancy of at least 1 year

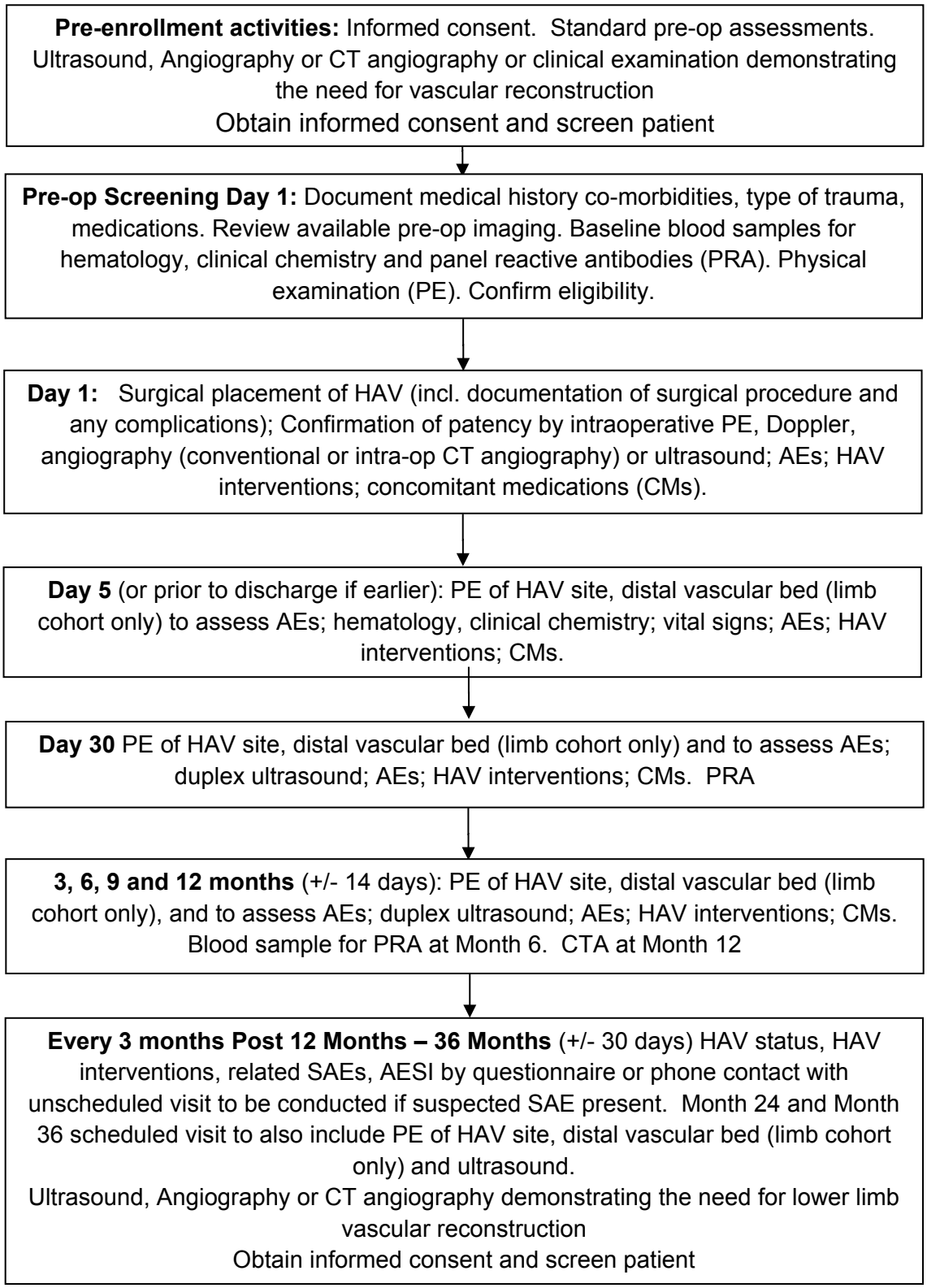
Exclusion Criteria	1. Mangled Extremity Severity Score (MESS) of ≥ 7. 2. Limb at high risk of amputation despite vascular reconstruction (e.g., because of crush injury) 3. Catastrophic injuries that make survival unlikely (e.g. Abbreviated Injury Scale (AIS) > 5 or Injury Severity Score (ISS) >60) 4. HAV may not be used for coronary artery repair. 5. Known pregnant women 6. Known medical condition which would preclude long term antiplatelet therapy after resolution of acute injuries 7. Any other condition which in the judgment of the investigator would preclude adequate evaluation of the safety and efficacy of the Humacyte Human Acellular Vessel (HAV) 8. Previous exposure to HAV 9. Known participation in any investigational study within the last 30 days 10. Employees of the sponsor or patients who are employees or relatives of the investigator
Expected Enrollment Start	3Q 2018
Accrual Period	24 months
Study Duration	The active study duration for each study participant will be 36 months from HAV implantation. All subjects will be followed for the initial 12 months. Beyond 12 months (Long-Term Follow Up), only subjects with a patent HAV will be followed out to a total of 36 months from HAV implantation. The total expected duration of the clinical study is 61 months.
Study Design	Prospective, multicenter, multi-cohort, non-randomized study There will be a limb cohort and a torso cohort.
Investigational Device/Intervention Description	Patients will be implanted with a Humacyte Human Acellular Vessel (HAV) as an interposition replacement or bypass using standard vascular surgical techniques.

Primary Objectives	**Safety** To evaluate the safety and tolerability of the Humacyte HAV in vascular trauma patients undergoing surgery for vascular replacement or reconstruction **Efficacy** To determine the rate of primary patency at 30 days
Secondary Objectives	**Safety** • To determine mechanical stability of the HAV based on freedom from aneurysmal degeneration, anastomotic bleeding or spontaneous rupture, infection, or significant stenosis • To determine HAV durability in terms of freedom from HAV removal or replacement **Efficacy** • To determine the patency of the HAV (primary, primary assisted and secondary) • To determine the rates of interventions needed to maintain / restore patency in the HAV • To determine the rate of limb salvage • To determine patient survival • To evaluate remodeling of HAV
Endpoints	Endpoints will be assessed for up to 36 months after HAV implantation. The primary analysis of the study will be conducted on the earlier of a) when the last subject enrolled reaches 30 days post-implant or b) all subjects enrolled in the initial 24 month accrual period have reached 30 days post-implant. **Primary Endpoints:** *Safety:* • Frequency and severity of adverse events *Efficacy:* • HAV primary patency at 30 days **Secondary Endpoints:**

	Safety:
	• Frequency of adverse events of special interest:
	  ○ Anastomotic bleeding or spontaneous rupture
	  ○ HAV infection
	  ○ Thrombosis
	  ○ Pseudoaneurysm formation
	  ○ Aneurysm formation
	  ○ Hemodynamically significant stenosis (>70% by duplex ultrasound criteria)
	• HAV partial or complete removal
	Efficacy:
	• HAV primary patency
	• HAV primary assisted patency
	• HAV secondary patency
	• Rate of HAV interventions
	• Limb salvage (limb cohort only)
	• Patient survival
	• HAV remodeling as shown by histopathology of any clinical explants

Protocol Approval (Version and Date)	Version 3.0 18 July 2018

Schematic of Study Design:



1. STUDY PERSONNEL

An updated list of all study personnel will be maintained by the CRO. Protocol amendments will not be required for staff changes at Humacyte, the CRO or the sites (except change of Principal Investigator at a site).

CRO Project Manager:

Brad Whitlow
Project Manager
Atlantic Research Group
2421 Ivy Road, Suite 200
Charlottesville, VA 22903

Email: bwhitlow@atlanticresearchgroup.com
Mobile: 828-442-0526
Office: 434-202-4537
Fax: 434-202-4537

Sponsor Medical Representative:

Jeffrey H. Lawson, MD, PhD
Chief Medical Officer
Humacyte, Inc.
2525 East NC Highway 54
Durham, NC 27713, USA

Email: lawson@humacyte.commailto:
Mobile: 919-630-7851
Office: 919-313-9633

Sponsor Regulatory Representative:

William Tente
Chief Regulatory Officer
Humacyte, Inc.
2525 East NC Highway 54
Durham, NC 27713, USA

Email: tente@humacyte.com
Mobile: 401-714-4151
Office: 919-313-9633

Sponsor Clinical Operations Representative:

Angela Rose
Sr. Director, Clinical Development
Humacyte, Inc.
2525 East NC Highway 54
Durham, NC 27713, USA

Email: rose@humacyte.com
Mobile: 919-667-3507
Office: 919-313-9633

CRO Safety Representative:

Todd Fisher
Sr. Drug Safety Specialist
Atlantic Research Group
2421 Ivy Road
Charlottesville, VA 22903

Email: safety@atlanticresearchgroup.com
Phone: 888-619-3216
Fax: 866-851-9318

Medical Monitor:

Jennifer Prichard, MD
Medical Monitor
Atlantic Research Group
2421 Ivy Road
Charlottesville, VA 22903

Email: jprichard@atlanticresearchgroup.com
Mobile: 434-249-5530
Phone: 888-619-3216
Fax: 866-851-9318

2. BACKGROUND INFORMATION AND SCIENTIFIC RATIONALE

2.1. Background Information

In the civilian population, traumatic vascular injuries are mainly concentrated to the limbs and torso (abdomen, retroperitoneum, thorax and thoracic outlet). According to the PROspective Observational Vascular Injury Treatment (PROOVIT) database, designed to collect vascular trauma injuries from 24 Level I and Level II trauma centers in the United States, vascular injuries in the lower limb, torso, upper limb and neck have a distribution of 41%, 30%, 22%, and 6%, respectively. (DuBose, 2015, Faulconer, 2017) These reports incorporate a significant percentage of relevant venous injuries in addition to arterial. Additionally, in the civilian population, lower extremity bone fractures with associated arterial injuries are common, due to motor vehicle accidents, gunshot wounds, dog bites, and other situations resulting in blunt or penetrating trauma. (Helfet, 1990, Andrikopoulos, 1995, Akingba, 2012) In fact, the incidence of this type of vascular injury has increased considerably in the past 50 years. (Andrikopoulos, 1995) Although this type of injury represents less than 1% of all civilian injuries, fractures with associated vascular damage require special attention because of their potentially severe complications, including limb necrosis and amputation.

Currently, in order to attempt to salvage the injured limb or end-organ in the distal vascular bed and to prevent life threatening hemorrhage the vascular component of these injuries are treated with interposition or bypass grafting. This type of reconstruction is performed by using either autologous vein from the patient (typically great saphenous vein), or by using a synthetic graft, such as ePTFE or Dacron. However, the use of these grafts in the civilian population is not always possible or without additional risks. The patient may not have adequate autologous vein for harvest, and in many of these cases, such as dog bite injuries, the wound is “dirty” and a synthetic vascular substitute is contraindicated due to the risk of infection. (Akingba, 2012) Thus, civilians would benefit from a vascular graft that does not contain infection-prone synthetic material and has similar properties as human tissue, but does not require time consuming or high morbidity procedures to harvest vessels from the patient.

Warfighters could also benefit from an off-the-shelf biologic vascular graft. In modern combat, the incidence of vascular injury is much greater than in previous wars. The rate of vascular injury in the Vietnam War was 2-3%. But between 2002-2009, the rate of vascular injury was over 12% in a study of over 13,000 battlefield injuries. In combat scenarios vascular injuries occur in all locations of the body, but injuries to the lower extremities are the most common, followed by vascular injuries to the upper extremity, and then neck, followed by torso. (Rasmussen, 2010) Improvised Explosive Devices, or IED’s, caused more than 3,000 casualties per year during recent conflicts. (Cordesman, 2010) Arterial damage, laceration and thrombosis can require vascular reconstruction to save tissues from ischemia, necrosis, and further amputation. Harvesting autologous vein for vascular reconstruction is problematic

because IED casualties often have multi-limb injuries, making harvest of autologous vein highly risky or impossible. (Holcomb, 2011) Synthetic vascular reconstruction using synthetic vascular grafts made from Teflon (ePTFE)/Dacron is relatively contraindicated, since IED wounds are always “dirty”, and bacteria in the wound can colonize the synthetic graft, causing abscesses and sepsis.

Thus, there is a significant unmet medical need for alternative grafts, which can be used in situations where autologous vein is unavailable or undesirable to use and which more closely mimic human vascular tissue to avoid or reduce the infection complications associated with ePTFE and Dacron.

2.2. Scientific Rationale

Humacyte has developed an acellular, human tissue engineered vascular conduit, the human acellular vessel (HAV, to provide an alternative to synthetic materials and to autologous grafts in the repair of traumatic vascular damage. Because this product mimics native vascular tissue, it may possess the advantages of an autologous graft; it also has the benefits of synthetic grafts in that it is available off-the-shelf. Use of an off-the-shelf product avoids the surgical morbidity associated with vein graft harvest and most importantly allows vessel bypass surgery in patients who have no suitable vessels available. Because the product mimics a native vessel, it may not have the compliance mismatch associated with synthetic alternatives. In addition, pre-clinical studies in pigs, canines and primates have shown that the HAVs resist intimal hyperplasia at the anastomoses. (Quint, 2011, Prichard, 2011, Dahl, 2011) Upon implantation, the collagen matrix comprising the HAV is infiltrated with host cells and remodeled by the host. This could result in a vascular structure more similar to the histological composition of the native vascular tissue that may improve bypass longevity and be less likely to become infected. The latter potential advantage is of high importance in the repair of peripheral vascular trauma where most wounds are heavily contaminated.

2.3. Summary of Nonclinical Information

The non-clinical testing program was designed to comprehensively address:

- local and systemic effects of the product in multiple in vivo animal models acutely and chronically,
- functional aspects of product implanted into animal models as an arteriovenous conduit
- biocompatibility of the HAV material in standardized in vitro and in vivo test protocols.

Overall, the results of these studies indicated that the HAV extracellular matrix material was non-toxic, well tolerated, and met standards for biocompatibility. Generally, the HAVs functioned

as intended and maintained patency during the implantation period. (See the Investigator Brochure for a detailed summary of non-clinical data.)

Pre-implantation, the HAV has mechanical properties (burst pressure and suture retention strength) comparable with native human artery and vein. (Table 1.) There was no evidence that HAV strength deteriorated after long-term implantation into baboons.

Table 1: Summary of Mechanical Properties of Explanted Acellular Vessels

Test Material	Burst Pressure (mm Hg)	Suture Strength (g)
Pre-Implant Humacyte HAVs	3415 +/- 1011 (n=4)	180 +/- 44 (n=12)
Post-Explant Humacyte HAVs	3669 +/- 1305 (n=5)	276 +/- 84 (n=11)
Human Saphenous Vein	1,680 – 2,273[a]	196 +/- 2 (n=7)[a]
Human Artery	2,031 – 4,225[a]	200 +/- 119 (n=9) [a]

[a] From L'Heureux et al, *Nature Medicine*, 2006. (L'Heureux, 2006)

In the chronic animal testing Humacyte vessels produced using canine cells were implanted into 12 dogs (canine acellular vascular graft, CAVG) and 14 baboons (human acellular vessel -HAV) in a variety of anatomical locations. In general, the Humacyte vessels were safe and well tolerated, and functioned as intended.

Mechanical failure was not observed in any HAV. Calcification was not observed in any CAVG or HAV. No graft exhibited hemodynamically significant intimal hyperplasia. Unlike with ePTFE graft implantation, no evidence of systemic infection attributable to implantation of HAV was observed in any of the animals. One HAV developed an aneurysm that was resected and did not harm the animal. The HAV material showed no evidence of toxicity in hematology, clinical chemistry, and necropsy data. The HAVs could be accessed by venipuncture and hemostasis was achieved following needle puncture.

On microscopic analysis, the HAVs were found to be well integrated into the host tissue. Overall, the cellular host response to the HAVs demonstrated smooth muscle actin-positive cells within the vessel wall, endothelial cells lining the lumen, and an adventitial-like outer layer adjacent to the vessel. These findings indicate that implanted HAVs were populated with cell types that are characteristic of healthy native vasculature. Examination of the anastomotic sections showed that the HAVs were well integrated with adjoining vasculature with minimal intimal hyperplasia observed. Furthermore, IHC was employed to identify CD-68 positive macrophages in the venous intimal tissue. Studies have shown a substantial macrophage population has been observed within venous intimal tissue adjacent to inflammatory ePTFE arteriovenous grafts. (Kelly, 2002, Roy-Chaudhury, 2001) Only sparse CD-68 positive macrophages were observed, indicating that the degree and the aggressiveness of the intimal hyperplasia associated with the HAV were less than that typically associated with ePTFE grafts. (Prichard, 2011)

Over time, the organization and composition of extracellular matrix (ECM) components indicated that, aided by infiltration of host vascular cells, HAVs were remodeled in vivo in a manner that mimicked the dynamic remodeling process of native blood vessels. Given the difficulties associated with the baboon animal model, where mismatches in vein vs graft diameter were encountered and animals perturbed their wounds postoperatively, an overall assisted patency rate of approximately 80% (11/14) was achieved. In a xenogeneic transplant model that did not employ immunosuppression, the HAV material did not elicit biologically significant cellular or delayed-type hypersensitivity (DTH) immune responses. All animals developed immunoglobulin G (IgG) titers to the HAV material that did not appear to detrimentally impact vessel function.

In internationally recognized in vitro and in vivo International Organization for Standardization (ISO) test protocols, the HAV material met criteria for biocompatibility required of medical devices.

These data collectively support the safety of the HAV for the proposed clinical investigation.

2.4. Summary of Clinical Studies

2.4.1. Overview

The HAV clinical development program currently includes 7 clinical studies: 4 in patients with end-stage renal disease receiving hemodialysis (CLN-PRO-V001, CLN-PRO-V003, CLN-PRO-V006 and CLN-PRO-V007), 2 in patients with peripheral arterial disease (CLN-PRO-V002 and CLN-PRO-V004) and 1 in patients with vascular trauma (CLN-PRO-V005). Three Phase 1/2 studies have completed primary analysis with long-term follow-up ongoing (CLN-PRO-V001, CLN-PRO-V002 and CLN-PRO-V003), 1 phase 3 study completed enrollment and follow-up is ongoing (CLN-PRO-V006), 2 phase 2 studies (CLN-PRO-V004, CLN-PRO-V005) and one phase 3 study (CLN-PRO-V007) are open for enrollment.

As of 10 April 2018, 272 patients (244 hemodialysis access patients and 28 PAD patients) have received a HAV. The first implant for hemodialysis was performed in December 2012, and the first peripheral arterial bypass in October 2013. Overall, the total treatment exposure is approximately 329 patient years in the hemodialysis access population and 55 patient years in the PAD population. More information on the clinical profile of the HAV in these ongoing studies is provided in the Investigator Brochure.

2.4.2. Experience in Peripheral Arterial Bypass Patients

Humacyte has two phase 2 studies to assess the safety and efficacy of the HAV when used as an above-knee arterial bypass graft. The first study, CLN-PRO-V002, is a single group uncontrolled study conducted at 3 sites in Poland that is fully enrolled and in long-term follow up. Eligible patients required a femoro-popliteal bypass graft for the management of

symptomatic peripheral arterial disease. Pre-operative imaging (conventional or CT angiography) must have demonstrated at least two below knee vessels patent to the ankle with good runoff. The proximal anastomosis was expected to be below the inguinal ligament and the distal anastomosis above the knee. Autologous vein grafts must not have been suitable or feasible (e.g., because of severe venous disease or prior use of leg veins for other bypass surgery or there is a clinical need to preserve those veins for future bypass surgery in the coronary or peripheral circulation).

The HAV was implanted using standard vascular surgical techniques and the patency of the bypass confirmed by intraoperative angiography (conventional or intra-op CT angiography) or ultrasound. The patient was then followed up at study visits at 15 days, 6 weeks and 3, 6, 12, 18 and 24 months. At each visit safety was assessed by clinical examination and adverse events, and the HAV was examined using duplex ultrasound to visualize the entire length to confirm patency, flow and to detect stenosis, aneurysm development or dilatation.

The primary objectives of the study are to evaluate the safety and tolerability of the Humacyte HAV in PAD patients undergoing above-knee femoro-popliteal bypass surgery and to determine the patency (primary, primary assisted and secondary) rate of the Humacyte HAV at 24 months. Secondary objectives include assessment of the panel reactive antibodies (PRA)) and IgG response to the HAV and to assess patency (primary, primary assisted and secondary) at 6, 12 and 18 months, to determine the rates of interventions needed to maintain / restore patency in the HAV, to assess any effect of implantation on claudication, rest pain and ischemic ulcers and to assess any effect on ankle-brachial index (ABI).

The second PAD study of similar design, CLN-PRO-V004, is being conducted in the US with enrollment ongoing.

2.4.2.1. CLN-PRO-V002 Study Results (24 M)

Recruitment began in October 2013 and was completed in June 2014 with 20 patients implanted. Thirteen patients completed the 2 year follow up visit. Of the seven patients terminating the study early, three died and four were withdrawn after occlusion of the HAV. None of the deaths were considered related to the investigational device or procedure.

Kaplan-Meier analyses in which deaths were censored revealed primary, primary assisted, and secondary patency probability rates of 79.2%, 79.0%, and 89.5% at Week 26, 63.3%, 63.2%, and 84.2% at Month 12, 63.3%, 63.2%, and 79.0% at Month 18, and 58.1%, 57.9%, and 73.7% at Month 24.

Six patients (30%) required at least 1 graft intervention to maintain or restore HAV patency during the study. Four patients required 1 intervention and 1 patient each required 3 and 4 interventions. Most interventions successfully restored patency. However, in 1 patient the graft patency could not be restored and the HAV was replaced with an alternative bypass graft. Two patients, who had previously undergone successful interventions, developed a recurrent

thrombosis which was not treated and the HAV was left occluded. Two patients experienced HAV thrombosis with no or minimal symptoms and refused interventions on the HAV.

All 20 patients experienced AEs (a total of 92 events). Thirty-one of these events in 13 patients were considered serious. The most frequent AEs reported included graft thrombosis (35% of patients), anastomotic stenosis (20% of patients), lymphocele (20% of patients), and local swelling (15% of patient). Those SAEs reported by at least 2 patients were graft thrombosis (6 patients, 30%) and anastomotic stenosis (2 patients, 10%).

No patient showed an increase in PRA levels. Two patients had a significant (>2 fold) increase from baseline in IgG levels. One of these patients experienced a thrombosis of the HAV between 3 and 6 months after implantation, while the other patient has had no HAV-related AEs and continues to have primary patency. Neither patient has had any evidence of dilatation or structural degeneration of the HAV.

CLN-PRO-V002 Conclusions:

- Humacyte HAV was safe and well tolerated in PAD patients.
- The HAV is able to withstand long term use in a high pressure, high outflow resistance arterial circuit.
- Patency rates for the HAV are within the ranges of patency rates of synthetic and autologous grafts presented in the literature.
- Humacyte HAV was not immunogenic.

2.4.3. Experience in Hemodialysis Patients

Two phase 2 trials, one in Poland (CLN-PRO-V001) and one in the US (CLN-PRO-V003) have completed enrollment. Both recruited subjects requiring hemodialysis access for end-stage renal disease whom were not suitable for creation of an AVF. Most subjects had undergone previous vascular access procedures, in many cases multiple attempts including both AVFs and synthetic grafts. Initial results from these phase 2 studies are discussed below.

The primary objectives of these two studies are to evaluate both the safety of HAV and its efficacy in terms of primary and secondary patency at 6 months. Secondary objectives include measurement of a panel of reactive antibodies (PRA) response, development of IgG antibodies to the extracellular matrix material in the HAV and a 2-year evaluation of patency and an assessment of the need for interventions to maintain/restore patency. Follow up has now been extended up to 120 months.

A phase 3 randomized study comparing HAV with ePTFE grafts (CLN-PRO-V006) has completed enrollment in the US, Europe, and Israel. A second phase 3 randomized study (CLN-PRO-V007) comparing HAV with AVF is currently enrolling in the US. As the sponsor is blinded, no efficacy information currently available for the phase 3 studies; however, blinded safety data is presented in the Investigator Brochure.

2.4.3.1. CLN-PRO-V001 and CLN-PRO-V003 Study Results (24 M)

All subjects (n=60) have now completed at least 24 months since implantation (or had a censoring event). The first subjects recruited are now beyond 60 months after HAV implantation, some with functioning HAV for hemodialysis access. Together these two trials provide more than 150 years of follow up during which the HAV has been used for more than 15,000 hemodialysis sessions.

When HAV thrombosis has occurred it has almost always been managed successfully, often allowing immediate resumption of dialysis without the need for the placement of a dialysis catheter. One non-serious arteriovenous graft aneurysm was reported in Study CLN PRO V001 (moderate in intensity, considered possibly related to IMP and considered not related to procedure – this patient died before the Sponsor could complete the follow up of this event). An expected number of small pseudoaneurysms have been observed, which is consistent with all surgically-created hemodialysis access. Most have resolved spontaneously with only 2 cases requiring surgical intervention. Flow rates through the HAV were more than sufficient to allow for effective dialysis.

In both studies, the product has generally been well tolerated and blood chemistry, hematology and coagulation data are not indicative of any HAV-associated toxicity. Immunogenic response to the HAV material has not been observed as demonstrated by a general lack of HAV-related change in PRA levels (Class I or II). Three subjects had elevations in their PRA levels: all 3 subjects had experienced one or more renal transplant failures; one subject recently; one subject developed septic shock about a month before the elevated value; and the third subject, who was severely debilitated with a decubitus ulcer, died approximately a month after HAV abandonment.

IgG titers increased in 5 subjects; in 4 cases the IgG titer increased and then decreased while the HAV remained functional with no clinical evidence of an inflammatory response; in one case the IgG titer increase occurred in a subject who maintained primary patency.

Adverse events (AEs) related to the HAV / access site (excluding thrombotic events) were few; there have been only three access-site infections, of which only one required removal of part of the HAV. There have been:

- 1 transplant (known to be functioning well at 12 months post-transplant)
- 15 deaths, all after abandonment or during follow-up; none of the deaths were considered related to the presence of the HAV

Patency data for the two studies in dialysis access has been pooled for a combined Kaplan Meier analysis (Lawson, 2016). Based on these K-M plots the patency at 6, 12 and 24 months is estimated to be 60%, 26% and 15% (primary patency) and 97%, 91% and 77% (secondary patency).

2.4.4. Human Acellular Vessel Host Response and Remodeling Data

Humacyte has been able to assess the general host response to the HAV in a number of human participants; this was accomplished through the microscopic examination of explanted HAV and adjoining tissue samples obtained during surgical revision procedures in eight cases. The analysis (mostly of a section close to the venous anastomosis) included assessments of:

- Cellular infiltration of histotypic, inflammatory and immunological populations.
- Extracellular remodeling processes, including neo-synthesis and reorganization of ECM components that typically occur in native blood vessels.

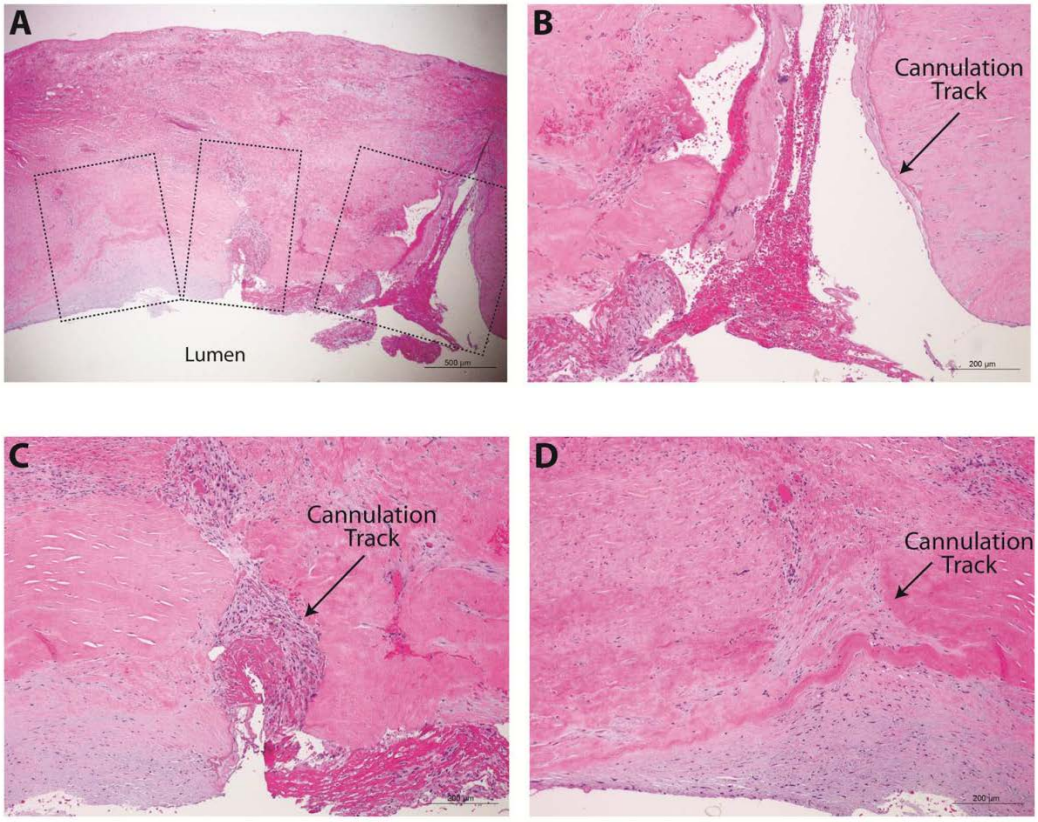
In these cases, small segments of the HAV and adjacent vascular tissue were explanted, fixed in formalin solution and shipped to Humacyte for analysis. Implant duration ranged from 16 to 55 weeks (median: 37 weeks).

In man, the HAV remodeled in a manner consistent with that observed in primate studies. There was infiltration of cell populations that are normally associated with angiogenesis and vascular organization and structure; namely, those with endothelial, smooth muscle and fibroblastic phenotypic characteristics were observed. Endothelial cells formed a monolayer on the luminal surface of the HAV. Migration of actin-positive smooth muscle cells into the wall of the HAV was consistently observed. A well-vascularized adventitial layer of non-constrictive fibrous tissue formed around HAV. Infiltration of the graft material by inflammatory and immunoreactive cell populations was either not evident or was mild and generally unremarkable. Degradation or breakdown of the implant was not observed.

Histotypic neo-synthesis and reorganization of the ECM was observed in patterns indicative of integration of the HAV into the host. An increase in the density of collagen type I, the main type of collagen found in the wall of native blood vessels, was apparent in the majority of HAV explant specimens. The structure of collagen type I in these specimens exhibited a more mature, organized pattern, with distinct fibers and a prominent circumferential alignment evident in explanted samples in comparison with pre-implant specimens. In some specimens, the fibrillar staining pattern of collagen III became more prominent and more organized, with a circumferential orientation. Fibronectin levels and staining patterns remained unchanged.

Cannulation sites within the HAV appeared to be repaired by the host in a fashion similar to wound repair in the body (Figure 1). In one case, an explanted specimen was tested for suture retention strength at the time of explant and exhibited a substantial increase over the pre-implant level.

Figure 1 Images of Mid-Vessel Segment Explanted at 11-Months Post-Implant



A: Low magnification showing 3 cannulation sites (in dashed boxes),

B: Fresh cannulation track,

C: Cannulation track during remodeling

D: Older cannulation track that has been repaired.

The images above show a mid-vessel segment explanted at 11 months post-implant, and shows several prior cannulation tracts from dialysis access. Section B shows a very recent cannulation site with fresh clot extending into the tract from the lumen. Sections C and D show partially healed cannulation tracts, with evidence of cellular repopulation extending in from the lumen. Remodeled cannulation tracks contain new collagen and a few micro-conduits.

In conclusion, the HAVs were remodeled by the host to form a vascular-like structure more similar to the histological appearance of native vasculature. The HAVs were repopulated by cell types that are characteristic of healthy native vasculature. Evidence of ECM remodeling processes, including neo-synthesis and reorganization of ECM components that typically occur in native blood vessels, were observed. The cellular infiltration and ECM remodeling patterns were indicative of the integration of the HAV into the host.

2.4.5. Conclusions

Clinical experience indicates that the HAV remains mechanically strong over implantation periods of more than 60 months with no evidence of dilatation. During more than 200 patient-years of follow up across the three phase 2 studies only one case of infection of the HAV material itself has been reported. The serious adverse event (SAE) profile has been typical of that expected in the dialysis and PAD populations. In hemodialysis populations, secondary patency of the HAVs is substantially higher than the historical data for both ePTFE and AVF (accounting for non-maturation). In PAD, patency is in line with historical ePTFE and autologous conduit for above knee bypass. No evidence of immunogenicity of the HAV has been found and the HAV remains mechanically robust even after repeated puncture for hemodialysis and under high pressure, high outflow resistance in arterial reconstruction.

These data support the use of HAV in future phase 2 and phase 3 studies for vascular replacement and reconstruction in diseased or damaged (trauma) vessels.

2.5. Potential Risks and Benefits

2.5.1. Potential Risks

It is anticipated that subjects participating in the study will be exposed to similar risks to those associated with other arterial conduits. Risks associated with the study investigational product may include but are not limited to:

- Thrombosis/occlusion of the conduit or host vessels, with consequent limb ischemia
- Embolism from a thrombosed conduit
- Bleeding and hematoma formation at the surgical site
- Infection – at the surgical site or systemic
- Stenosis of the conduit or its anastomoses
- Aneurysm or pseudoaneurysm formation
- Swelling of the limb
- Failure/injury to the target end-organ
- Bleeding/hemorrhage in the peritoneum or retroperitoneum

Regular clinical examination of the HAV implantation site and assessment of the patency, blood flow and diameter using ultrasound during the study should allow early detection of complications and permit appropriate intervention including HAV explantation.

The HAV is grown using donor human aortic smooth muscle cells. The vessel is decellularized during manufacturing and thus consists of human extracellular matrix proteins. It is possible that the HAV may provoke an immune response which may lead to damage to the HAV and possible

cross reactivity against host proteins. Possible antibody formation will be assessed by analyzing PRA.

2.5.2. Potential Benefits

Patients who undergo implantation of the Humacyte HAV may benefit from improved patency and a reduced number of interventions versus a conventional ePTFE or Dacron graft. This may result from a decreased propensity for anastomotic and downstream neointimal hyperplasia, which often leads to graft occlusion with synthetic grafts. In addition, risks of infection typically encountered with conventional synthetic grafts may be decreased with the Humacyte HAV. Finally, the longevity (secondary patency) of the Humacyte HAV may be greater than that of conventional synthetic grafts.

Patients may also benefit from reduced morbidity secondary to harvest of autologous vessels for vascular reconstruction. Harvest of autologous vascular conduit requires additional time to perform and in the urgent/emergent trauma scenario, time is of the essence and is key to positive patient outcomes. Use of saphenous, or other autologous vessels for purposes of vascular repair in young, otherwise healthy patients prevents its future potential use for other indications (e.g. coronary bypass, etc.). Vascular reconstruction using synthetic vascular grafts (ePTFE, or woven materials like Dacron) may be contraindicated for use in trauma scenarios since the wounds are often contaminated allowing bacteria in the wound to colonize the synthetic graft potentially leading to abscesses, hemorrhage secondary to anastomotic blow out, blood stream infection, and sepsis. Unlike other readily available synthetic vascular grafts made from ePTFE or Dacron, HAVs are comprised of human proteins, resulting in grafts that may be less prone to infection than synthetic materials, as shown in pre-clinical studies. (Kirkton, 2018)

2.5.3. Risk-Benefit Rationale

The risks anticipated in this study are similar to those associated with currently marketed prosthetic grafts used for vascular repair. The potential advantages of the HAV compared to currently marketed grafts may lead to a lower complication rate and reduced need for surgical intervention and graft replacement as well as the potential for reduced secondary complications associated with autologous vessel harvest.

This is the first in man study in which the HAV will be used to repair vascular injuries within the torso (thorax, thoracic outlet, abdomen, and retroperitoneum) and so risks specific to those locations have not been specifically characterized. However, these risks are not expected to be significantly different than those experienced/reported when used in the upper or lower extremity for AV access or arterial reconstruction. The DMC will review safety data of the torso cohort on the earlier of when the first torso subject reaches 3 months post-implantation or when the first 2 torso subjects have both reached 30 days post-implantation. Overall recruitment will be restricted to a maximum of 40 subjects who receive implants.

3. STUDY OBJECTIVES

3.1. Primary Objectives

This is an open label phase 2 study. There is no formal hypothesis testing.

Safety:

- To evaluate the safety and tolerability of the Humacyte HAV in vascular trauma patients undergoing surgery for vascular replacement or reconstruction

Efficacy:

- To determine the rate of primary patency at 30 days

3.2. Secondary Objectives

Safety:

- To determine mechanical stability of the HAV based on freedom from aneurysmal degeneration, anastomotic bleeding or spontaneous rupture, infection, or significant stenosis
- To determine HAV durability in terms of freedom from HAV removal or replacement

Efficacy:

- To determine the patency of the HAV (primary, primary assisted and secondary)
- To determine the rates of interventions needed to maintain / restore patency in the HAV
- To determine the rate of limb salvage
- To determine patient survival
- To evaluate remodeling of HAV

4. STUDY DESIGN

4.1. Description of the Study Design

Prospective, multicenter, multi-cohort, non-randomized phase 2 study.

There will be a limb cohort and a torso cohort. The limb cohort will include patients who require repair of a vessel contained to the upper or lower extremity. Upper extremity vasculature is defined as the axillary and more distal vessels. The lower extremity vasculature is defined as the common femoral and more distal vessels. Examples of size appropriate vessels within the extremities include (but are not limited to):

- Axillary
- Brachial
- Basilic
- Popliteal
- the femoral vessels

The torso cohort includes patients who require repair of vessels within the thorax (excluding the heart), abdomen, and retroperitoneum. A vascular repair in which any portion originates or terminates in the torso will be considered a patient in the torso cohort even if the other end of the repair resides in an extremity. End to side anastomotic takeoff from a larger caliber vessel (e.g. aorta, vena cava) is acceptable for enrollment within this study. Examples of size appropriate vessels within the torso include (but are not limited to):

- subclavian
- brachiocephalic/innominate
- celiac
- hepatic
- splenic
- superior mesenteric
- renal
- the iliac vessels

The DMC will review the earlier of when the first torso subject reaches 3 months post-implantation or when the first 2 torso subjects have both reached 30 days post-implantation.

4.2. Study Endpoints

Endpoints will be assessed for up to 36 months after HAV implantation. The primary analysis of the study will be conducted on the earlier of a) when the last subject enrolled reaches 30 days post-implant or b) all subjects enrolled in the initial 24 month accrual period have reached 30 days post-implant.

4.2.1. Primary Endpoints

Safety:

- Frequency and severity of adverse events

Efficacy:

- HAV primary patency at 30 days

4.2.2. Secondary Endpoints

Safety:

- Frequency of adverse events of special interest:
  - Anastomotic bleeding or spontaneous rupture
  - HAV infection
  - Thrombosis
  - Pseudo-aneurysm formation
  - Aneurysm formation
  - Hemodynamically significant stenosis (>70% by duplex ultrasound criteria)
- Frequency of study conduit removal

Efficacy:

- HAV primary patency
- HAV primary assisted patency
- HAV secondary patency
- Rate of HAV interventions
- Limb salvage (limb cohort only)
- Patient survival
- HAV remodeling as shown by histopathology of any clinical explants

4.3. Duration of Study Participation

The active study duration for each study participant will be 36 months from HAV implantation. All subjects will be followed for the initial 12 months. Beyond 12 months (Long-Term Follow Up), only subjects with a patent HAV will be followed out to a total of 36 months from HAV implantation. The total expected duration of the clinical study is 61 months.

5. STUDY POPULATION

5.1. Description of the Study Population

The study population will consist of patients with vascular trauma to size appropriate vessels in the limb or torso, requiring replacement or reconstruction.

5.1.1. Patient Inclusion Criteria

1. Patients with life or limb threatening traumatic injury to an arterial vessel in the limb or torso, other than the heart, which requires replacement or reconstruction
2. Preoperative imaging or clinical examination indicates the damaged vessel has a defect length of ≤ 38cm and is appropriately size matched to the 6mm Human Acellular Vessel (HAV) per the judgment of the treating surgeon taking into account vasoconstriction and situational inflow and outflow considerations.
3. Autologous vein graft is either not feasible in the judgment of the treating surgeon (e.g. because of lack of availability of suitable conduit, presence of severe venous insufficiency) or is not desirable because of the urgency of revascularization
4. Aged 18 to 85 years old, inclusive
5. Able to communicate meaningfully with investigative staff, and able to comply with entire study procedures. If the patient is unconscious, then information from a reliable witness indicates that the patient would normally be able to comply with study procedures
6. Patient or relative is able, willing and competent to give informed consent
7. Life expectancy of at least 1 year

5.1.2. Patient Exclusion Criteria

1. Mangled Extremity Severity Score (MESS) of ≥ 7
2. Limb at high risk of amputation despite vascular reconstruction (e.g., because of crush injury)
3. Catastrophic injuries that make survival unlikely (e.g. Abbreviated Injury Scale (AIS) > 5 or Injury Severity Score (ISS) >60)
4. HAV may not be used for coronary artery repair
5. Known pregnant women
6. Known medical condition which would preclude long term antiplatelet therapy after resolution of acute injuries
7. Any other condition which in the judgment of the investigator would preclude adequate evaluation of the safety and efficacy of the HAV
8. Previous exposure to HAV

9. Known participation in any investigational study within the last 30 days
10. Employees of the sponsor or patients who are employees or relatives of the investigator

6. INVESTIGATIONAL MEDICINAL PRODUCT

Additional information on the manufacturing process and testing of the investigational medicinal product (IMP)is provided in the Investigator Brochure.

6.1. Product Description

The IMP is a Humacyte Human Acellular Vessel (HAV), which is a tissue-engineered vascular prosthesis for vascular bypass or reconstruction in patients with peripheral vascular disease or peripheral vascular trauma. It is a sterile, non-pyrogenic acellular tubular vessel composed of human collagen types I and III and other extracellular matrix proteins, including fibronectin and vitronectin. The HAV is 6 mm in diameter and approximately 42 cm in length. The product is supplied on a silicone mandrel immersed in sterile phosphate buffered saline in a sealed and labeled plastic container.

There is no placebo or comparator control group in this study.

6.2. Manufacturer of the IMP

The HAV is manufactured by:

AlloSource

6278 S. Troy Circle

Centennial, CO 80111 USA

Traceability of the HAV during and after the clinical investigation will be achieved by the assignment of lot numbers. A unique identifying lot number will be assigned to each vessel.

6.3. Packaging, Storage, and Labeling

Packaging: Each HAV is contained in a sealed, flexible plastic primary container closure system that was developed by Humacyte. The system meets container/closure requirements to maintain sterility as well as product and fluid integrity. The vessel is contained inside the system in a fixed manner, immersed in a sterile, phosphate buffered saline. The total volume of the storage solution is approximately 300 mL.

Storage: The product is shipped under controlled conditions to maintain temperature at 4˚C (range: 2 – 10˚C). The product should be stored in a refrigerator that maintains this temperature range. The HAV **MUST NOT** be allowed to freeze.

Labeling: The IMP will be labeled according to applicable guidelines and relevant regulatory agency requirements. A tamper resistant label affixed to the secondary container will be used to ensure that the product is not compromised prior to use.

6.4. Implantation of the Humacyte Human Acellular Vessel (HAV)

The Humacyte HAV is implanted using standard vascular surgical techniques similar to placement of predicate peripheral vascular prostheses (see study manual for details).

Tunneling of the HAV, if required, must be performed using a sheathed tunneler. After inserting the assembled tunneler into the tissue, the inner mandrel of the tunneler should be removed from the sheath. The sheath is lubricated with saline and then with the silicone mandrel in place, the HAV can be easily pushed through the sheath without the need to tie to the inner mandrel and pulled through the tunneler (see study manual for details).

After placement, HAV patency and integrity are checked by pressurizing the conduit. Prior to completion of surgery, HAV patency is confirmed by physical exam, Doppler, angiography (conventional or intra-op CT angiography) or ultrasound. The surgical site is closed using standard techniques.

Implantation of the HAV will be undertaken by qualified vascular surgeons experienced in peripheral vascular surgery.

6.5. IMP Accountability Procedures

Documentation of receipt, dispensing, and return of all IMP must be maintained by the Principal Investigator or his/her designee. It is the Principal Investigator's responsibility to ensure that all IMPs are kept in a secure location, with access limited to individuals authorized by the Investigator. The product will be shipped with the IMP Shipment Confirmation Form. Once signed, the form should be returned to Humacyte or authorized designee, and the original will be maintained in the Investigator's Files. The IMP Accountability Log will be used to account for all IMP received, dispensed, and returned and must be maintained by the site until the conclusion of the study. Following accountability of the IMP by Humacyte or their authorized designee, all unused IMP will be returned to Humacyte.

7. OTHER TREATMENTS AND MEDICATIONS

7.1. Prior and Concomitant Medications

Prior medications are defined as all prescription and over the counter (OTC) medications taken within 7 days (whether continuing or not) prior to Day 1. All prior and concomitant medications (including immediately pre-surgery and post-surgery medications) must be listed in the patient's medical record and recorded on the eCRF. Drugs used during anesthesia should be recorded in the anesthesia records but should not be transcribed into the eCRF. Patients should be questioned at each study visit concerning any new medications or changes in current medications. **Note: particular attention should be made to identify the use of antithrombotic or antiplatelet agents (e.g., aspirin, clopidogrel, prasugrel, direct thrombin inhibitors, factor Xa inhibitors, or vitamin K antagonists).**

For each medication taken, the following information will be collected:

- Medication generic name / components of combination product
- Dose
- Route of administration
- Frequency of administration
- Date started
- Date stopped
- Indication for use

7.2. Essential, Precautionary and Prohibited Medications

7.2.1. Essential Medications

All patients should receive both antibiotic and antithrombotic prophylaxis in conjunction with HAV implantation:

Antibiotic prophylaxis:

- All patients must have at least 1 day of antibiotic prophylaxis the same day as surgery in accordance with local hospital guidelines. Longer antibiotic prophylaxis is at the discretion of the investigator.

Antithrombotic prophylaxis:

- Intraoperative heparin: the doses of heparin to be used during surgery will be determined by the investigator.

- Further measures to prevent venous thromboembolism are at the discretion of the investigator and may include LMWH.
- If antiplatelet therapy was not ongoing at the time of surgery it should be commenced as soon as medically appropriate post operatively. Recommended antiplatelet therapy (aspirin 81-325 mg and/or clopidogrel 75 mg daily) is at the discretion of the investigator and should continue long term while the HAV is in place. If the patient is unable to tolerate aspirin and/or clopidogrel the choice of antiplatelet regimen is at the investigator's discretion.

7.2.2. Restricted Medications

Vitamin K antagonists, antiplatelet agents other than aspirin and clopidogrel, direct thrombin inhibitors and factor Xa inhibitors (e.g., dabigatran, apixaban and rivaroxaban) should be avoided unless essential for treatment of a medical condition arising postoperatively. In that case consideration should be given to modification or cessation of antiplatelet therapy. Antiplatelet therapy should be restarted on cessation of these anticoagulant drugs.

8. STUDY PROCEDURES / EVALUATIONS

8.1. Clinical Evaluations Through Month 12

- Medical History – pre-operatively, from patient / legal representative interview and medical records covering relevant past medical history.
- Smoking history
- Medication History – prescription and OTC medication from Day -7 onwards (see Section 6.7). Particular attention should be paid to the identification of OTC medications containing aspirin.
- Physical Exam – full exam (as far as possible) at pre-operative screening, 12 month visit or final study visit for early termination (ET). Clinical examination of the operative site and HAV at all post-operative visits; exam of distal vascular bed (limb cohort only); physical exam for lymphadenopathy; additional clinical exam as needed to evaluate adverse events
- Vital signs (heart rate, blood pressure and temperature) – at D5
- Blood samples for hematology, clinical chemistry at pre-operative screening and Day 5 and PRA at pre-operative screening, Day 30 and M6
- Pre-operative imaging (ultrasound or angiography) is at the discretion of the investigator based on the clinical condition of the patient. It may be omitted if not deemed necessary in order to proceed to vascular surgical repair
- Adverse events – post-operatively on Day 1 and at all post-operative visits, the patient will be asked a general question about his/her health and for any HAV problems since the previous visit
- Intraoperative HAV bypass or interposition repair exam to assess anastomotic anatomy, patency and runoff. This may include physical exam, Doppler exam, angiography (conventional or intra-op CT angiography) or ultrasound at the investigator's discretion.
- Duplex ultrasound – clinical assessment at all postoperative visits from day 30 thru M12, to assess HAV patency, mid HAV diameter and stenosis. The full length of the HAV should be imaged at each assessment to monitor for aneurysm development
- Documentation of HAV interventions, surgical procedures and any complications immediately postoperatively through Month 12

8.2. Clinical Evaluations in Long Term Follow Up (Post Month 12 to Month 36)

- The status of the patient and HAV will be ascertained every 3 months from post Month 12 until 36 months after HAV implantation by telephone contact with the patient and/or his physician. If a suspected SAE related to HAV is discovered an unscheduled visit should be conducted to investigate.
- Only related SAEs and all AESI will be reported.
- Visits at Month 24 and Month 36 are to be conducted in person with a physical exam of the HAV site and duplex ultrasound imaging of the HAV.

8.3. Laboratory Evaluations

8.3.1. Clinical and Research Laboratory Evaluations and Specimen Collection

The following parameters will be measured wherever possible at pre-operative screening and all should be measured at Day 5

- Hematology: hemoglobin, hematocrit, RBC, white blood cells (WBC) with differential, platelet count
- Clinical chemistry: sodium, potassium, calcium, blood urea nitrogen, creatinine, albumin, total bilirubin, glucose (non-fasting)
- PRA will be measured at pre-operative screening, Day 30, and Month 6.

All laboratory tests (except assay of PRA) will be conducted at certified hospital laboratories. Routine monitoring, maintenance or calibration of laboratory equipment is required per local site procedures. Samples for PRA analysis will be shipped to LabConnect for storage until they are sent for analysis at a central laboratory. Details concerning sample collection and processing can be found in the Study Manual.

8.4. Imaging Evaluations

8.4.1. CT Angiography and Conventional Angiography

CT angiography or conventional angiography will be conducted as pre-operative screening when feasible. Pre-operative imaging is at the discretion of the investigator based on the clinical condition of the patient. It may be omitted if not deemed necessary in order to proceed to vascular surgical repair.

8.4.2. Duplex Ultrasound

Duplex ultrasound examinations will be performed at Day 30, 3, 6, 9,12, 24, and 36 months and follow standard bypass graft imaging protocols, including B-mode, power Doppler and color duplex ultrasound imaging of the HAV with velocity spectral waveform analysis. The purpose of this duplex ultrasound surveillance is to detect HAV stenosis and aneurysm development. An alternative imaging method (e.g. CTA, MRI, etc.) may be substituted for duplex ultrasound at the discretion of the investigator if it is medically appropriate and in the best interest of the patient.

Determination of intraoperative HAV patency on Day 1 is required by physical examination, Doppler exam, angiography (conventional or intra-op CT angiography), or ultrasound at the discretion of the investigator.

8.5. Study Schedule

8.5.1. Pre-operative Screening (Day 1)

Potential study participants who are being considered for surgical repair of vascular injury appropriate for inclusion into the study will be informed about the study and invited to participate. After explanation of the potential risks and benefits of the HAV and of the study procedures, written informed consent will be obtained. No study specific procedures may be performed prior to patient consent. If the patient is unable to give informed consent then this may be sought from the patient's legal representative, usually a close relative. Standard of care procedures such as laboratory evaluations conducted prior to screening may be used rather than repeating the test.

The following assessments will be performed, as far as possible, prior to surgery (Day 1):

- Informed consent
- Medical history
- Prior and concomitant medication
- Full physical examination
- Evaluation of inclusion/exclusion criteria
- Reasons for not using an autologous venous conduit
- Laboratory testing (or standard pre-op lab profile for the institution)
    - Hematology: full blood count and differential
    - Clinical chemistry ; sodium, potassium, calcium, blood urea nitrogen, creatinine, albumin, total bilirubin, glucose (non-fasting)
    - PRA

- Ultrasound or CT angiography (CTA) (pre-op imaging is at the discretion of the investigator based on the clinical condition of the patient. It may be omitted if not deemed necessary in order to proceed to vascular surgical repair

8.5.2. Enrollment – Day 1 (HAV Implantation)

The HAV will be implanted as an interposition replacement or bypass in the required location using standard vascular surgical techniques. Details of the surgical anatomy and any complications will be documented. Operative procedures note and surgical diagram will be uploaded into the EDC for medical monitor review. Determination of intraoperative HAV patency is required by physical examination, Doppler exam, angiography (conventional or intra-op CT angiography) , or ultrasound at the discretion of the investigator.

8.5.3. Follow-up Visits Day 5 through Month 12

Day 5 (or prior to hospital discharge if earlier)

- Concomitant Medication
- Physical exam including surgical site
- Vital signs (heart rate, blood pressure and temperature)
- Documentation of any HAV interventions
- Adverse events
- Laboratory assessments (clinical chemistry, hematology)

Day 30 (+ 5 days)

- Concomitant medication
- Physical exam including surgical site
- Duplex ultrasound of the HAV
- Documentation of any HAV interventions
- Adverse events
- PRA

Months 3, 6 and 9 (+/- 14 days)

- Concomitant Medication
- Physical exam including surgical site
- Duplex ultrasound of the HAV
- Documentation of any HAV interventions

- Adverse events
- PRA (Month 6 only)

Month 12 (+/-14 days) and Early Termination

- Concomitant Medication
- Full physical exam including surgical site
- Duplex ultrasound of the HAV
- Documentation of any HAV interventions
- Adverse events
- PRA (only at early termination if prior to 6 month PRA collection)
- CT angiography

8.5.4. Long Term Follow-up Post Month 12 through Month 36 (+/- 30 days)

The status of the patient and HAV will be ascertained every 3 months from post Month 12 through 36 months after HAV implantation.

- Quarterly questionnaire covering status of the patient via a telephone contact with the patient and/or physician. If a suspected SAE related to HAV is discovered an unscheduled visit should be conducted to investigate.
- Documentation of any HAV interventions
- Adverse events (all AESI and related SAEs to be reported)
- Physical exam including surgical site at Month 24 and Month 36
- Duplex ultrasound at Month 24 and Month 36

8.5.5. Early Termination Visit

The subject may withdraw from the study at any time at his/her own discretion. The treating physician may also withdraw the subject for safety reasons. If withdrawal occurs before 12 months the subject will be asked to complete an early termination visit at which all assessments normally performed at 12 months will be completed. PRA will be collected at ET visit if the visit occurs before Month 6 collection of the sample. If withdrawal occurs after Month 12 and prior to Month 36 the patient will be asked to complete an early termination visit at which all assessments normally conducted during the long term follow up visits will be completed.

The reasons for early termination should be recorded in the eCRF.

With the exception of patient withdraw, all subjects will be followed for 12 months from HAV implantation (or until HAV removal or death if earlier). The subject should be withdrawn from the

study if the HAV is completely removed or the HAV becomes permanently occluded (loss of secondary patency) after Month 12.

8.5.6. Unscheduled Visits

If necessary to evaluate adverse events or HAV complications additional visits may be scheduled at the discretion of the investigator. At a minimum HAV status on clinical examination and Duplex ultrasound and adverse events will be recorded.

If, at any of the scheduled visits, duplex ultrasound surveillance suggests the development of a ≥ 50% stenosis within the HAV but immediate intervention is not required closer follow up should be considered. Intervention to manage any such stenosis is at the discretion of the investigator taking into account the degree and rate of progression of the stenosis.

8.6. Medical Care during the Study and upon Study Termination

Optimal medical therapy should be continued during the study. This should include:

- Antiplatelet therapy (see section 7.2.1)

After the final study visit at month 36 patients will not receive any further study-specific treatment. They will be treated by their medical doctor in a way that is appropriate for them.

8.7. Histological Examination of Resected HAV Material

If all or part of the HAV is resected it should, wherever possible, be retained for future histological examination. Instructions for preservation, storage and shipping of this material will be provided separately in a procedures manual. If a patient dies with an HAV in situ and it is feasible to obtain a fresh post mortem sample of the bypass this should be attempted in accordance with local regulations.

9. SAFETY ASSESSMENTS AND ADVERSE EVENTS

Safety of the HAV will be assessed in terms of:

- Aneurysm formation
- Pseudoaneurysm formation
- Anastomotic bleeding or spontaneous rupture
- HAV infection
- Need for HAV removal
- Inflammation at the implantation site
- Other adverse events
- Increase from baseline in PRA

9.1. Adverse Event Definition

An AE is any untoward medical occurrence in a patient administered an IMP and which does not necessarily have a causal relationship with the IMP. An AE can, therefore, be any unfavorable and unintended sign, symptom, or disease temporally associated with the use of an IMP, whether or not related to the IMP. Any worsening of the patient's disease under study or other medical conditions will also be considered to be an AE, unless it is within the normal range of disease fluctuation for that patient.

9.2. Serious Adverse Event Definition

An AE is considered "serious" if, in the view of either the investigator or Sponsor, it:

- Results in death
- Is life-threatening
    - The term "life-threatening" in the definition of "serious" refers to an event in which the patient was at immediate risk of death at the time of the event; it does not include an adverse event that had it occurred in a more severe form, might have caused death.
- Requires patient hospitalization or prolongation of existing hospitalization
    - This is defined as the patient being hospitalized for 24 hours or more or the patient's hospital stay being prolonged for at least an additional overnight stay.
- Requires intervention to prevent permanent damage
- Results in persistent or significant disability/incapacity

- Is a congenital anomaly/birth defect
- Important Medical Events
    - For the purpose of this study, this includes any event involving the HAV that results in a surgical or endovascular radiological intervention. The event(s) which caused the procedure should be reported as an SAE. For example: in the event of HAV thrombosis, the thrombosis would be considered the SAE; any associated stenoses (or other associated findings) that are present would be considered AEs.

Medical and scientific judgment should be exercised in deciding whether expedited reporting to the sponsor is appropriate in other situations, such as important medical events that may not be immediately life-threatening or result in death or hospitalization but may jeopardize the patient or may require medical or surgical intervention to prevent one of the other outcomes listed in the definition above. These should also be considered to be SAEs.

Note: Hospitalization for the surgery to implant the HAV is not a SAE. However, prolongation of the initial hospitalization due to an AE will be considered a SAE.

9.3. Suspected Unexpected Serious Adverse Reaction

A suspected unexpected serious adverse reaction (SUSAR) is any adverse drug reaction that is serious (as defined in 9.2), unexpected (is not listed in the IB or is not listed at the specificity or severity that has been observed) and suspected (meaning there is a reasonable possibility that the IMP caused the adverse event).

9.4. Events of Special Interest

Events of Special Interest are:

- HAV occlusion (thrombosis)
- HAV spontaneous rupture
    - Iatrogenic injuries are not an Event of Special Interest and should be reported as an AE
- HAV infection
- HAV abandonment
- HAV aneurysm
- HAV pseudoaneurysm
- HAV Excision (partial or complete)

9.5. Reporting of Adverse Events

At each evaluation, the investigator will determine whether any AEs have occurred. The patient will be questioned in a general way and no specific symptoms will be suggested. If any AEs have occurred they should be documented in the patient's medical chart and recorded on the AE pages of the eCRF. If known, the diagnosis should be recorded in preference to the listing of individual signs and symptoms. All SAEs, should be reported to the Safety CRO within 24 hours from the time the investigator or study personnel first become aware of the event.

AE reporting begins from time of anesthesia induction for implantation of the HAV and ends at the conclusion of the Month 12 visit or ET visit, unless an unresolved AE is still being followed.

During the long term follow up period from post Month 12 through Month 36, only the following will be reported by the investigator:

- All SAEs considered related to the HAV
- All Events of Special Interest (Section 9.4)

9.5.1. Criteria for Determining Causal Relationship to the HAV and Criteria for Determining Causal Relationship to the Index Surgical Procedure

The criteria for determining the causal relationship of an AE with the HAV are presented in the table below. A separate assessment of causal relationship of an AE to the index surgical procedure is required as well using the same criteria and definitions presented in the table below. Please note that causal relationship to procedure only refers to the index surgical procedure in which the HAV was initially implanted.

Causal Relationship to the IP	**Criteria for Determining Causal Relationship**
Definitely Related	There is clear evidence to suggest a causal relationship, and other possible contributing factors can be ruled out. The clinical event, including an abnormal laboratory test result, occurs in a plausible time relationship to surgical placement of the HAV and cannot be explained by concurrent disease or other devices, drugs, or chemicals.
Possibly Related	There is some evidence to suggest a causal relationship (e.g., the event occurred within a reasonable time after the surgical placement of the HAV). However, the influence of other factors may have contributed to the event (e.g., the subject's clinical condition, other concomitant medications). Although an adverse event may rate only as "possible" soon after discovery, it can be flagged as requiring more information and later be upgraded to certain as appropriate.

Unlikely Related	A clinical event, including an abnormal laboratory test result, whose temporal relationship makes a causal relationship improbable (e.g., the event did not occur within a reasonable time after surgical placement of the HAV) and in which other drugs or chemicals or underlying disease provide plausible explanations (e.g., the subject's clinical condition, other concomitant treatments).
Not Related	A clinical event, including an abnormal laboratory test result, which occurs when the HAV was not implanted; or, another cause is obvious and in which there is sufficient information that the etiology of the event is not related to the HAV.

The sponsor will make the final determination of causality for the purposes of reporting to the regulatory authorities and to the Principal Investigators.

9.5.2. Criteria for Defining the Severity of an Adverse Event

Severity of adverse events, including abnormal clinical laboratory values, will be assessed according to the criteria below and entered in the eCRF:

Grade	**Severity Assessment Standard**
1-Mild	Events require minimal or no treatment and do not interfere with the subject's daily activities.
2-Moderate	Events result in a low level of inconvenience or concern with the therapeutic measures. Moderate events may cause some interference with functioning.
3-Severe	Events interrupt a subject's usual daily activity and may require systemic drug therapy or other treatment. Severe events are usually incapacitating.
4-Life-threatening	Any adverse event that places the subject or participant, in the view of the investigator, at immediate risk of death from the reaction as it occurred, i.e., it does not include a reaction that, had it occurred in a more severe form, might have caused death.
5-Death	Death related to AE.

9.5.3. Reporting of Action Taken to Resolve AE

- None

- Lab tests / further evaluation
- Treatment required (specify if hospitalized)
- Patient withdrawn from study
- Other (specify)

9.5.4. Reporting the Outcome of the AE

- Recovered, with sequelae
- Recovered, without sequelae
- Ongoing
- Death
- Lost to follow-up

9.5.5. Reporting Serious Adverse Events

The urgency for reporting SAEs is 4-fold: (1) to facilitate discussion (and implementation, if necessary) by the sponsor and the investigator of appropriate follow-up measures, (2) to facilitate investigator reporting of unanticipated problems involving risk to human subjects to the institutional review board (IRB), (3) to facilitate the sponsor's rapid dissemination of information regarding AEs to other investigators/sites in a multi-center study, and (4) to enable the sponsor to fulfill the reporting requirements to the appropriate regulatory authority.

Any SAE that occurs through Month 12, whether or not causally related to the IMP, must be reported by the investigator or designee to the Safety CRO within 24 hours of learning of its occurrence. This applies also to any AE that could affect the safety of the study participants or the conduct of the study. Any SAE that occurs during long term follow up post Month 12 through Month 36 that is causally related to the IMP, must be reported by the investigator or designee to the Safety CRO within 24 hours of learning of its occurrence.

Information about an SAE will be collected and recorded on the SAE Report Form. The investigator must assess the relationship to the investigational product and any relevant procedure.

The investigators must complete the SAE Report Form in English, and **send the completed, signed form by fax or email (see below) IMMEDIATELY (at latest within 24 hours) after becoming aware of the SAE.**

Copies of relevant medical records (e.g., admission and/or discharge summary, laboratory reports and autopsy report), may also be submitted with the SAE form to clarify the circumstances surrounding the SAE(s). The entire medical records should **NOT** be sent with the SAE form.

Atlantic Research Group, Inc.

Drug Safety Department

Fax: +1-866-851-9318

Email: Safety@atlanticresearchgroup.com

Phone: +1-888-619-3216

The investigator will be requested to supply as much detailed information as possible regarding the SAE that is available at the time of the initial contact. The investigator should also complete missing or requested information and submit follow-up reports until the SAE has resolved or, in the case of permanent impairment, until the SAE has stabilized.

It is the responsibility of each Principal Investigator to promptly notify his/her IRB of all SAEs that are received by the Sponsor or designee and that occur at his/her institution in accordance with institutional practices.

The Safety CRO will inform the sponsor about all SAEs within 1 business day after receipt of the respective report from the investigator.

9.5.6. Reporting of Events of Special Interest

Events of Special Interest are defined in Section 8.4 and should be reported to the Safety CRO within 24 hours of learning of its occurrence. For each of these events detailed surgical notes (with illustrative diagram), including reason for and outcome of any intervention or abandonment, should be completed within 48 hours and uploaded to the clinical database.

Detailed information about the occurrence and treatment/intervention for these events will be collected throughout the study up to 3 years post HAV implant. This information will include the following:

- Summarized surgical notes, including a simplified anatomical diagram showing where angioplasties, stents, or revisions have been performed (using intervention worksheet provided)
- Need for hospitalization (number of nights)
- Need for antibiotics (in the case of HAV-related infections)

9.5.7. Follow-Up of Adverse Events

If any AEs are present when a subject completes 1 year post implant (Month 12) or ET, if earlier, or if a subject is withdrawn from the study, the subject will be re-evaluated within an appropriate period of time. At the investigator's discretion, minor AEs can be re-evaluated via telephone and documented. If the AE has still not resolved, additional follow-up will be performed as appropriate. The investigator or his designee should make every effort to contact the subject until the AE has resolved or stabilized or the medical monitor and investigator agree

that further follow-up is not necessary. This should be documented in the subject's medical records.

9.6. Reporting of Pregnancy

If a study participant becomes pregnant during study participation, basic information about the pregnancy will be recorded in the Pregnancy eCRF and the Pregnancy Outcome and Report Form, and submitted to the Safety CRO. If there are complications during the pregnancy, the complications are recorded as AEs. The participant will be asked to report the outcome of the pregnancy and the site should submit the information to the Safety CRO within 30 days after the outcome of the pregnancy. If there is a congenital anomaly in the infant, this will be recorded as a SAE in the data forms for the mother (i.e., the study participant).

Partner pregnancies do not need to be reported.

9.7. Data Monitoring Committee

A Data Monitoring Committee (DMC) will review safety on an ongoing basis and provide recommendations about stopping, continuing or otherwise modifying the study. The DMC consists of individuals who are not directly involved in the conduct of the study. A charter describes the roles and responsibilities of the DMC. Responsibilities of the DMC will include review of aggregate safety data from other studies in the HAV clinical development program.

The DMC will at a minimum meet every 6 months from the date of initial enrollment of the first subject. Additionally, the DMC will review the safety data of the torso cohort on the earlier of when the first torso subject reaches 3 months post-implantation or when the first 2 torso subjects have both reached 30 days post-implantation.

9.8. Interim Analysis and Stopping Criteria

This is a phase 2 study with no formal interim analysis. Periodic reviews of safety data will be undertaken by the DMC with particular attention to events that might indicate structural failure of the HAV. Events that might have implications for already implanted HAVs and their possible removal - such as aneurysm formation (true or pseudo) or spontaneous rupture -would trigger an urgent review of the safety data for DMC review.

The DMC may recommend modification or early termination of the study for safety reasons.

10. STATISTICAL CONSIDERATIONS

This is a prospective, open label, multi-cohort, multicenter pilot study to evaluate the safety and efficacy of the HAV in patients undergoing vascular replacement or reconstruction. The primary objective of this study is to evaluate the safety and tolerability of the HAV in these patients and to determine the primary patency of the HAV at 30 days post-implantation. The secondary objectives of this study are to further assess safety in terms of adverse events of special interest, to determine the rate interventions required to keep the HAV patent, and to further characterize efficacy in terms of secondary patency and limb salvage. There is no formal hypothesis testing planned.

Endpoints will be assessed over a period of up to 36 months after HAV implantation. The primary analysis of the study will be conducted on the earlier of a) when the last subject enrolled reaches 30 days post-implant or b) all subjects enrolled in the initial 24 month accrual period have reached 30 days post-implant. Details of data handling and planned descriptive statistics are given in the Statistical Analysis Plan.

10.1. Analysis Population

All patients who receive an HAV will be included in the analyses. For discontinued or withdrawn patients, all available data will be included in the safety and efficacy analyses.

10.2. Safety Analyses

Safety analyses will be performed on all patients who have an HAV implanted.

The incidence of aneurysm formation, anastomotic bleeding or spontaneous rupture, HAV removal, HAV infection, and inflammation at the implantation site will be tabulated by visit and overall.

Adverse events will be coded using Medical Dictionary for Regulatory Activities (MedDRA) terms. Adverse events will be listed and summarized by body system, incidence, severity, and duration. HAV complications will be listed in terms of incidence, severity, and (where appropriate) time to onset and duration. Serious adverse events will be summarized separately. Any premature discontinuations due to adverse events and deaths will be listed and summarized.

PRA data will be listed and summarized using appropriate descriptive statistics for the change from baseline values.

10.3. Efficacy Analyses

The primary efficacy analysis will be the rate of primary patency at 30 days after HAV implantation. Primary, primary assisted, and secondary patency rates of the HAV at 12 months and at all other post-surgery visits with evaluation of patency will be described. The rate of limb salvage and patient survival at 12 months will also be described.

Primary patency is defined as the functional access patency until any type of intervention; primary assisted patency is defined as an HAV still working without thrombosis; secondary patency is defined as the functional HAV patency, with or without preceding successful interventional or surgical procedures to maintain or reestablish patency, until the HAV is abandoned. Early discontinued patients prior to the visit of interest will be determined as being non-patent irrespective of the reason for discontinuation.

The rate and type of interventions needed to maintain / restore patency in the HAV will be descriptively tabulated.

The absolute change from baseline (Day 1) values to all post-surgery visits of duplex ultrasound parameters will be summarized. Summary statistics will also be provided at each time point.

The methods and endpoints regarding the efficacy parameters employed in this study are consistent with current clinical practice and are meaningful to the research community. Every attempt has been made to minimize the variability on the part of the surgeon when using this product.

10.4. Other Analyses

All clinical parameters will be listed for all patients treated at each study visit. Descriptive statistics will be summarized for continuous outcomes such as age and BMI. If necessary, number and percentage of patients will be reported for categorical outcomes.

10.5. Sample Size Rationale

Up to 40 subjects will be recruited into the study. As this phase 2 study is the first human study of the HAV for vascular trauma, the study was designed to provide preliminary evidence of safety and efficacy.

10.6. Interim analyses

There is no formal interim analysis.

11. STUDY MANAGEMENT AND DATA COLLECTION

11.1. Ethical Conduct of the Trial

This study will be conducted according to the protocol; 21 CFR Parts 11, 50, 54, 56, and 312; the World Medical Association Declaration of Helsinki and Good Clinical Practice (GCP). Each Investigator will conduct the trial according to applicable local or regional regulatory requirements.

11.2. Institutional Review Board

IRBs must be constituted according to the applicable state and federal requirements, including ICH GCP.

It is the responsibility of each investigator to submit the protocol, Investigator's Brochure, subject informed consent, subject recruitment materials (if applicable), and other documentation as required by the IRB to his/her IRB for review and approval. A copy of the written approval must be provided to the contract research organization (CRO). The documentation should clearly mention the approval/favorable opinion of the protocol, the subject informed consent form, and subject recruitment materials (if applicable), including respective version dates. The written approval and a list of members, their titles or occupations, and their institutional affiliations may be obtained from the IRBs if available, and provided to the CRO prior to the release of clinical study supplies to the investigational site and commencement of the study. If any member of the IRB has direct participation in this trial, written notification regarding his or her abstinence from voting must also be obtained.

Each investigator must adhere to all requirements stipulated by his/her respective IRB. This includes notification to the IRB regarding protocol amendments, updates to the subject informed consent, recruitment materials intended for viewing by subjects, investigational new drug safety reports, SAEs and unexpected AEs, reports and updates regarding the ongoing review of the trial at intervals specified by the respective IRB, and submission of final study reports and summaries to the IRB.

11.3. Subject Informed Consent

Prior to any study procedures being performed, subjects and persons conducting the consent discussion will be required to sign and date the IRB-approved informed consent, and each subject will be given a copy. In addition, this information should be recorded in the subject's medical record (i.e., source document). If the patient is unable to give informed consent then this may be sought from the patient's legal representative, usually a close relative.

The written consent document will embody the elements of informed consent as described in the World Medical Association Declaration of Helsinki, 21 CFR Part 50.25, ICH E6 guideline (GCP), and in accordance with any local regulations. The investigator is responsible for the

preparation, content, and IRB approval of the informed consent document. The consent form must be approved by the site's IRB and be acceptable to Humacyte.

The consent form must be written in a language fully comprehensible to the prospective subject. The investigator or designee shall give the subject adequate opportunity to read it before it is signed and dated. Information should be given in both oral and written form whenever possible and in the manner deemed appropriate by the IRB. Subjects must be given ample opportunity to inquire about details of the study.

11.4. Amendments to the Protocol

An amendment must be agreed to in writing by Humacyte and submitted to the FDA and approved by t IRBs before the amendment can be implemented. Written approval of a protocol amendment is not required prior to implementation of changes to the protocol which eliminate an immediate hazard to the study patient; however, approval must be obtained as soon as possible thereafter. Any agreed amendments must also be signed by the investigator.

11.5. Study Initiation

The investigator must not enroll any patients prior to attendance at the Investigator Meeting or the completion of a formal site initiation visit conducted by the CRO. These meetings will include a detailed review of the study protocol and eCRF pages. The investigator will not be supplied with IMP until all necessary pre-study requirements have been completed and essential signed documents provided to the CRO.

11.6. Study Monitoring

It is the responsibility of the investigator to ensure that the study is conducted in accordance with the protocol, GCP, applicable regulatory requirements, and the currently approved Declaration of Helsinki, and that valid data are entered into the eCRF.

To achieve this objective, the monitor's duties are to ensure the maintenance of complete, legible, well-organized, and easily retrievable data. The monitor will review the protocol with the investigator. In addition, the monitor will explain the investigator's reporting responsibilities and all applicable regulations concerning the clinical evaluation of the IMP.

The investigator will permit representatives of Humacyte and the CRO to monitor the study as frequently as Humacyte or the CRO deem necessary to determine that data recording and protocol adherence are satisfactory. The eCRF data and related source documents will be reviewed in detail by the monitor at each visit, in accordance with relevant SOPs and ICH GCP regulations. This includes results of tests performed as a requirement for participation in this study and any other medical records required to confirm information contained in the eCRF such as past medical history and secondary diagnoses. The investigator and his/her staff will be expected to cooperate with the monitor and provide any missing information whenever possible.

All monitoring activities will be reported and archived. In addition, monitoring visits will be documented at the investigational site by signature and date on the study-specific monitoring log.

11.7. Case Report Form

An electronic CRF will be used for this study. The data will be entered into the eCRF in a timely manner on an ongoing basis.

The investigator is responsible for ensuring that data are properly recorded on each patient's eCRF and related documents. An investigator who has signed the protocol signature page should personally sign the eCRFs in accordance with the procedure described in the eCRF completion guidelines to ensure that the observations and findings are correct and complete.

For data handled by the CRO, eCRF data and some or all of the study-related data will be managed and stored electronically in the CRO's database system. Validated data will subsequently be transferred to the sponsor.

11.8. Verification Procedures

It is the investigator's obligation to ensure documentation of all relevant data in the subject's medical record. The subject's medical record will be considered the source document. The eCRF should not be used as the source for study information.

The investigator will maintain a subject identification code list to enable unambiguous identification of the subjects (subject names and corresponding subject numbers). The subject identification code list is an essential document and as such should be maintained according to the ICH GCP guidelines.

11.9. Retention of Records

All documentation pertaining to the study will be kept by Humacyte or their designee in accordance with ICH guidelines and US FDA regulations.

The investigator will maintain a study file, which should be used to file the Investigator's Brochure, protocol, and IMP records; correspondence with the IRB and Humacyte; and other study-related documents.

The investigator agrees to keep records and those documents that include (but are not limited to) the identification of all participating subjects, medical records, study-specific source documents, source worksheets, all original signed and dated informed consent forms, query responses, and detailed records of drug disposition to enable evaluations or audits from regulatory authorities and Humacyte or its designees.

The investigator shall retain records required to be maintained under this part for a period of 2 years following the date a marketing application is approved for the IMP for the indication for which it is being investigated; or, if no application is to be filed or if the application is not

approved for such indication, until 5 years after the investigation is discontinued. However, these documents should be retained for a longer period if required by the applicable regulatory requirement(s) or if needed by the sponsor. In addition, the investigator must make provision for the subject's medical records to be kept for the same period of time. No data should be destroyed without the agreement of Humacyte. Humacyte will inform the investigator in writing when the trial-related records are no longer needed. Subject's medical records and other original data will be archived in accordance with the archiving regulations or facilities of the study site.

11.10. Protocol Deviations

A protocol deviation is any noncompliance with the protocol or GCP requirements. The noncompliance may be either on the part of the participant, the investigator, or the study site staff. As a result of deviations, corrective actions are to be developed by the site and implemented promptly. Although in principle, protocol deviations are not permitted, under emergency circumstances, deviations may proceed without prior approval of the sponsor and the IRB to protect the rights, safety, and well-being of human subjects.

All protocol deviations will be documented and reported by the CRO during the course of the study in the Monitoring Reports. All deviations will be reported to the sponsor who will agree on the necessary actions to be taken.

If required per their guidelines, reports about protocol deviations must be reported to the local IRB.

11.11. Insurance and Indemnity

Insurance coverage for damages emerging from the study will be provided according to applicable legal requirements. During the informed consent procedure, the investigator must inform the patient accordingly.

11.12. Audit

It is the responsibility of CRO and Humacyte to perform auditing (if applicable) as part of implementing quality assurance. The purpose of an audit, which is independent of and separate from routine monitoring or quality control functions, is to evaluate trial conduct and compliance with the protocol, SOPs, GCPs, and the applicable regulatory requirements. The auditor and regulatory authorities will require authority from the investigator to have direct access to the subject's medical records.

12. REPORTING

Following completion of follow-up of all patients to the 12-month endpoint, the results will be evaluated by Humacyte or a designee for clinically meaningful findings. A clinical study report will be generated, including a summary of all available data, statistical measures, tabulated results, graphical results and interpretations. This report will be submitted to regulatory authorities in a timely manner. An addendum to the report will be generated to include data up to 36 months follow-up. This addendum will be submitted to regulatory authorities in a timely manner.

13. QUALITY CONTROL AND QUALITY ASSURANCE

Following written standard operating procedures, the monitors will verify that the clinical trial is conducted and data are generated, documented (recorded), and reported in compliance with the protocol, GCP, and the applicable regulatory requirements. Reports of monitoring activities will be submitted to Humacyte in a timely manner.

The investigational site will provide direct access to all trial related areas, source data/documents, and reports for the purpose of monitoring and auditing by the sponsor, and inspection by local and regulatory authorities.

Quality control procedures will be implemented for data entry and the generation of data quality control checks and will be run on the database. Any missing data or data anomalies will be communicated to the site(s) for clarification/resolution.

14. RESPONSIBILITIES OF THE PRINCIPAL INVESTIGATOR

The role of the principal investigator is to implement and manage the day-to-day conduct of the clinical investigation as well as to ensure data integrity and the rights, safety, and well-being of the patients involved in the clinical investigation.

14.1. Informed Consent

The principal investigator shall ensure that the process for obtaining informed consent

- includes all aspects of the clinical investigation that are relevant to the patient's decision to participate throughout the clinical investigation,
- avoids any coercion or undue improper influence on, or inducement of, the patient to participate,
- does not waive or appear to waive the patient's legal rights,
- uses native non-technical language that is understandable to the patient,
- provides ample time for the patient to read and understand the informed consent form and to consider participation in the clinical investigation,
- provides the patient with a copy of the signed and dated informed consent form and any other written information.

The principal investigator shall ensure and document appropriate training if an authorized designee is appointed to conduct the informed consent process.

14.2. Compliance with the Protocol

The principal investigator shall:

- indicate his/her acceptance of the protocol in writing
- conduct the clinical investigation in compliance with the protocol
- create and maintain source documents throughout the clinical investigation and make them available as requested during monitoring visits or audits
- ensure that the IMP is used solely by authorized users, and in accordance with the protocol and instructions for use
- propose to the sponsor any appropriate modification(s) of the protocol
- refrain from implementing any modifications to the protocol without agreement from the sponsor, IRB, and, if required, regulatory authorities
- document and explain any deviation from the approved protocol that occurred during the course of the clinical investigation

- ensure that an adequate investigation site team and facilities exist and are maintained and documented during the clinical investigation
- ensure that maintenance and calibration of the equipment relevant for the assessment of the clinical investigation is appropriately performed and documented, where applicable
- ensure the accuracy, completeness, legibility, and timeliness of the data reported to the sponsor in the eCRFs and in all required reports
- maintain the clinical trial material accountability records
- allow and support the sponsor to perform monitoring and auditing activities
- be accessible to the monitor and respond to questions during monitoring visits
- allow and support regulatory authorities and the IRB when performing auditing activities
- ensure that all clinical-investigation-related records are retained as specified in this protocol.

14.3. Medical Care of Patients

The principal investigator shall:

- provide adequate medical care to a patient during and after a patient's participation in a clinical investigation in the case of AEs
- inform the patient of the nature and possible cause of any adverse events experienced
- inform the patient of any new significant findings occurring during the clinical investigation, including the need for additional medical care that may be required
- provide the patient with well-defined procedures for possible emergency situations related to the clinical investigation, and make the necessary arrangements for emergency treatment,
- ensure that clinical records are clearly marked to indicate that the patient is enrolled in a particular clinical investigation
- inform, with the patient’s approval or when required by national regulations, the patient's personal physician about the patient's participation in the clinical investigation
- make all reasonable efforts to ascertain the reason(s) for a patient's premature withdrawal from the clinical investigation while fully respecting the patient's rights.

14.4. Safety Reporting

The principal investigator shall:

- record every adverse event together with an assessment, in accordance with Section 9 of this protocol,
- report to the sponsor, without unjustified delay, all serious adverse events and medically significant events as specified in Section 9 of this protocol,
- supply the sponsor, upon sponsor's request, with any additional information related to the safety reporting of a particular event.

15. SUSPENSION OR PREMATURE TERMINATION OF THE CLINICAL INVESTIGATION

The sponsor may suspend or prematurely terminate either a clinical investigation in an individual investigation site or the entire clinical investigation for significant and documented reasons.

A principal investigator, IRB, or regulatory authority may suspend or prematurely terminate participation in a clinical investigation at the investigation sites for which they are responsible.

If suspicion of an unacceptable risk to patients arises during the clinical investigation, or when so instructed by the IRB or regulatory authorities, the sponsor shall suspend the clinical investigation while the risk is assessed. The sponsor shall terminate the clinical investigation if an unacceptable risk is confirmed.

The sponsor shall consider terminating or suspending the participation of a particular investigation site or investigator in the clinical investigation if monitoring or auditing identifies serious or repeated deviations on the part of an investigator.

If suspension or premature termination occurs, the terminating party shall justify its decision in writing and promptly inform the other parties with whom they are in direct communication.

If, for any reason, the sponsor suspends or prematurely terminates the investigation at an individual investigation site, the sponsor shall inform the responsible regulatory authority if required and ensure that the IRB is notified. If the suspension or premature termination was in the interest of safety, the sponsor shall inform all other principal investigators.

If suspension or premature termination occurs,

1. the sponsor shall remain responsible for providing resources to fulfill the obligations from the protocol and existing agreements for following up the patients enrolled in the clinical investigation, and
2. the principal investigator or authorized designee shall promptly inform the enrolled patients at his/her investigation site, if appropriate.

In the event that the study is discontinued, the reasons for discontinuation will be explained to the investigators and may be disclosed to the study participants. Humacyte will provide all information needed by the investigator to ensure the safety and well-being of the study participants.

16. PUBLICATION POLICY

A Publication Committee comprising the Principal Investigator from each participating site and a representative of Humacyte will oversee all publication of data from this study. Prior to submitting for publication, presenting, using for instructional purposes, or otherwise disclosing the results of the study, the investigator agrees to allow the Publication Committee and Humacyte a period of at least 30 days (or, for abstracts, at least 5 calendar days) to review the proposed publication or disclosure prior to its submission for publication or other disclosure. Publications or disclosures of study results shall not include other confidential information belonging to Humacyte. If the proposed publication/disclosure risks Humacyte's ability to patent any invention related to the study, the publication or disclosure will be modified or delayed, at the investigator's option, a sufficient time to allow Humacyte to seek patent protection of the invention. For multicenter studies, the first publication or disclosure shall be a complete, joint multicenter publication or disclosure. This statement does not give Humacyte any editorial rights over the content of a publication or disclosure, other than to restrict the disclosure of Humacyte's confidential information. If a written contract for the conduct of the study is executed which includes publication provisions inconsistent with this statement, then that contract's publication provisions shall apply rather than this statement.

17. REFERENCES

Akingba G, Rapp BM, Tsai A, Jester AL, Fajardo A, Motaganahali R, et al. Surgical Management of Vascular Trauma From Dog Bites: The 10- year Experience of a Level I Trauma Center. Journal of Vascular Surgery. 2012;56(4):1190.

Andrikopoulos V, Antoniou I, Panoussis P. Arterial injuries associated with lower-extremity fractures. Cardiovascular Surgery. 1995;3(1):15-8.

Cordesman AH, Lemieux, J. IED Metrics for Afghanistan. January 2004-May 2010. Center for Strategic & International Studies, 2010.

Dahl, SL, Kypson, AP, Lawson, JH et al. Readily available tissue-engineered vascular grafts. Sci Transl Med. 2011: 68ra9

Dixon BS, Beck GJ, Vazquez MA et al. Effect of dipyridamole plus aspirin on hemodialysis graft patency. N Engl J Med 2009;360:2191-201.

DuBose JJ, Savage SA, Fabian TC, Menaker J, Scalea T, Holcomb JB, Skarupa D, Poulin N, Chourliaras K, Inaba K, Rasmussen TE. The American Association for the Surgery of Trauma PROspective Observational Vascular Injury Treatment (PROOVIT) registry: multicenter data on modern vascular injury diagnosis, management, and outcomes. Journal of Trauma and Acute Care Surgery. 2015 Feb 1;78(2):215-23.

Faulconer ER, Branco BC, Loja MN, Grayson K, Sampson J, Fabian TC, Holcomb JB, Scalea T, Skarupa D, Inaba K, Poulin N. Use of open and endovascular surgical techniques to manage vascular injuries in the trauma setting: A review of the American Association for the Surgery of Trauma PROspective Observational Vascular Injury Trial registry. Journal of Trauma and Acute Care Surgery. 2018 Mar 1;84(3):411-7.

Helfet DL, Howey T, Sanders R, Johansen K. Limb Salvage Versus Amputation: Preliminary Results of the Mangled Extremity Severity Score. Clinical Orthopaedics & Related Research. 1990;256:80-6.

Holcomb J. Devastating Dismounted IED Injuries in OEF. Increasing amputation and genital injury rates admitted to LRMC 2009-2010. 2011. Presentation to the Defence Health Board, (http://www.health.mil/Reference-Center/Presentations/2011/03/08/Devastating-Dismounted-Improvised-Explosive-Device-Injuries-in-Operation-Enduring-Freedom)

Kelly BS, Heffelfinger SC, Whiting JF, et al. Aggressive venous neointimal hyperplasia in a pig model of arteriovenous graft stenosis. Kindey Int. 2002;62(6):2272-2280.

Kirkton RD, Prichard HL, Santiago-Maysonet M, Niklason LE, Lawson JH, Dahl SL. Susceptibility of ePTFE vascular grafts and bioengineered human acellular vessels to infection. Journal of Surgical Research. 2018 Jan 1;221:143-51.

L'Heureux N, Dusserre N, Konig G, et al. Human tissue-engineered blood vessels for adult arterial revascularization. Nat Med 2006; 12(3):361-365.

Mills JL et al on behalf of the Society for Vascular Surgery Lower Extremity Guidelines Committee. The Society for Vascular Surgery Lower Extremity Threatened Limb Classification System: Risk stratification based on Wound, Ischemia, and foot Infection (WIfI). J Vasc Surg 2013,

Morgan MBF, Crawford T, Murrin B and Fraser SCA. Developing the vascular quality of life questionnaire: a new disease specific quality of life measure for use in lower limb ischemia. J Vasc Surg 2001; 33: 679 - 687

Prichard, H.L, Manson, R.J.,Dibernado, L. et al. An early study on the mechanisms that allow tissue-engineered vascular grafts to resist vascular hyperplasia. J Cardiovasc Transl Res 2011, 4, 674-682

Quint C, Kondo Y, Manson RJ,et al. Decellularized tissue-engineering blood vessel as an arterial conduit. Proc Natl Acad Sci USA. 108(22):9214-9, 2011.

Rasmussen TE. Vascular Injury Rates in the Wars in Iraq and Afghanistan: How are they different from the past? United States Army Institute of Surgical Research, 2010.

Roy-Chaudhury P, Kelly BS, Miller MA, et al. Venous neointimal hyperplasia in polytetrafluoroethylene dialysis grafts. Kidney Int. 2001;59(6):2325-2334.

Rutherford RB1, Baker JD, Ernst C, et al Recommended standards for reports dealing with lower extremity ischemia: revised version. J Vasc Surg. 1997 Sep;26(3):517-38.

APPENDIX 1: HAV CLINICAL VISIT SCHEDULE

	Pre-op screening D1	D 1	D 5 or prior to d/c	D 30 + 5 days	M3 ± 14 days	M 6 ± 14 days	M 9± 14 days	M12 / ET† ± 14 days	M15-M36 †+/- 30 days
Informed consent	X								
Medical history and nature of trauma	X								
Concomitant medication	X	X	X	X	X	X	X	X	
Physical exam [1]	X	X	X	X	X	X	X	X	X [7]
Pre-op Ultrasound or CT angiography [2]	X								
Vital signs			X						
Eligibility (inclusion/exclusion criteria)	X								
HAV implantation and intraoperative confirmation of patency [3]		X							
Documentation of surgery and any complications		X							
Clinical chemistry	X [5]		X						
Hematology	X [5]		X						
PRA	X			X		X		X [6]	
Duplex ultrasound [4]				X	X	X	X	X	X [7]
CT angiography								X	
AEs		X	X	X	X	X	X	X	X [8]
Documentation of HAV interventions		X	X	X	X	X	X	X	X[8]

Abbreviations: AEs, adverse events; D, day; d/c, discharge; ET, early termination; HAV, human acellular vessel; M, month

1. Physical examination includes clinical exam of the operative limb and HAV at all post-operative visits (incl. patency assessment on D1) and physical exam to evaluate AEs; include distal vascular bed (limb cohort only)
2. Pre-op imaging is at the discretion of the investigator based on the clinical condition of the patient. It may be omitted if not deemed necessary in order to proceed to vascular surgical repair
3. Determination of intraoperative HAV patency can be done by physical exam, Doppler, angiography or ultrasound at the discretion of the investigator
4. An alternative imaging method (CTA, MRI, etc.) may be substituted for duplex ultrasound at the discretion of the investigator if it is medically appropriate and in the best interest of the patient.
5. Measured at preoperative screening when possible
6. PRA only collected at ET visit if the visit occurs before Month 6 collection
7. Visits at month 24 and month 36 to be conducted in person with physical exam of the HAV site and duplex ultrasound imaging of HAV.
8. The status of the patient and HAV will be ascertained every 3 months post Month 12 until 36 months after HAV implantation by telephone contact with the patient and/or his physician. Only related SAEs and all AESI will be reported after 12 months. If a suspected SAE related to HAV is discovered an unscheduled visit should be conducted to investigate.

† Patients withdrawn before Month 12 will perform ET visit that correlates with the procedures at Month 12. Patients withdrawn after Month 12 and prior to Month 36 should complete an ET visit that correlates with procedures post Month 12 through Month 36.

SUMMARY OF PROTOCOL CHANGES

All changes since version 2.0 of the protocol and incorporated in the current version (version 3.0) are briefly described below. Each change is also categorized as either related to safety or efficacy (thus requiring IRB/EC review) or administrative in nature.

Important note: No subjects were enrolled under Version 1.0, or 2.0. Version 1.0 was the initial protocol approved by the FDA.

Changes Incorporated in Protocol Version 3.0 (16 Jul 2018)	
Change and Reason for Change	**Section(s) Changed**
Changes Related to Safety and/or Efficacy	
Change: Additional potential risks of failure/injury to target end-organ and bleeding/hemorrhage in the peritoneum or retroperitoneum were added. In addition, possible antibody formation will be assessed by analyzing PRA. Reason: Risk added to account for additional anatomic locations added to the protocol.	Potential Risks, Section 2.5.1
Change: Discussion of potential benefits was revised and updated with current references. Discussion of additional cohorts was added as follows: “The DMC will review safety data of the torso cohort on the earlier of when the first torso subject reaches 3 months post-implantation or when the first 2 torso subjects have both reached 30 days post-implantation. Overall recruitment will be restricted to a maximum of 40 subjects who receive implants.” Reason: To be consistent with change in study design; expanded enrollment in the protocol allows for additional anatomic locations for size appropriate vessels.	Potential Benefits and Risk-Benefit Rationale, Sections 2.5.2 and 2.5.3, respectively

Changes Incorporated in Protocol Version 3.0 (16 Jul 2018)	
Change and Reason for Change	**Section(s) Changed**
Changes: Primary safety and efficacy objectives revised as follows: Previous Primary Objectives: **Safety:** To evaluate the safety and tolerability of the Humacyte HAV in patients with vascular trauma in the lower limb undergoing vascular reconstructive surgery **Efficacy:** ● To determine the patency (primary, primary assisted and secondary) rate of the Humacyte HAV at 12 months ● To determine the rate of limb salvage New Primary Objectives: **Safety:** To evaluate the safety and tolerability of the Humacyte HAV in vascular trauma patients undergoing surgery for vascular replacement or reconstruction **Efficacy:** To determine the rate of primary patency at 30 days Reason: Objectives were revised based on new limb and torso cohort study design.	Study Synopsis and Primary Objectives, Section 3.1
Changes: Secondary safety and efficacy objectives were revised as follows: **Safety:** ● Mechanical stability of the HAV will now be based on freedom from aneurysmal degeneration, anastomotic bleeding or spontaneous rupture, infection, or significant stenosis (previously based on duplex ultrasound) ● HAV durability will now be determined based on HAV removal or replacement (previously based on need for bypass conduit or replacement due to infection, bleeding, or conduit degeneration) **Efficacy:** ● Objectives were revised to reflect longer Accrual Period ● Additional objectives were added as follows: ○ To determine the rate of limb salvage ○ To determine patient survival ○ To evaluate remodeling of HAV Reason: Objectives were revised based on new study design.	Study Synopsis and Secondary Objectives, Section 3.2

Changes Incorporated in Protocol Version 3.0 (16 Jul 2018)	
Change and Reason for Change	**Section(s) Changed**
Change: Study design changed from a single-arm study to a multi-cohort study (limb cohort and torso cohort added). Details were added to Section 4.1 to define cohorts. Reason: Expanded enrollment in the protocol allows for additional anatomic locations for size appropriate vessels.	Study Synopsis and Description of Study Design, Section 4.1
Change: Extended expected duration of active study participation to approximately 61 months (from 24) and duration of study (Accrual Period) from 12 months to 24 months. Additional data on patient and HAV status will be collected at 3-month intervals through 36 months after HAV implantation. Reason: Increased duration more accurately reflects the study timeline with the addition of new limb and torso cohorts.	Study Synopsis and Duration of Study Participation, Section 4.3
Change: As a result of extending the Accrual Period from 12 to 24 months, a new section was added to outline the evaluations to be performed during the Long Term Follow Up: ● The status of the patient and HAV will be ascertained every 3 months from post Month 12 until 36 months after HAV implantation by telephone contact with the patient and/or his physician. If a suspected SAE related to HAV is discovered an unscheduled visit should be conducted to investigate. ● Only related SAEs and all AESI will be reported. ● Visits at Month 24 and Month 36 are to be conducted in person with a physical exam of the HAV site and duplex ultrasound imaging of the HAV. Reason: Due to the extended duration of the Accrual Period (from 12 months to 24 months), follow up was also extended.	Clinical Evaluations in Long Term Follow Up (Post Month 12 to Month 36), Section 8.2 (new section)
Change: PRA now measured at pre-operative screening, Day 30, and Month 6 (previously only pre-operative screening and Month 6). Reason: To increase the chance of a post-implantation PRA sample being obtained in a potentially difficult to follow trauma study population.	Clinical and Research Laboratory Evaluations and Specimen Collection, Section 8.3.1

Changes Incorporated in Protocol Version 3.0 (16 Jul 2018)	
Change and Reason for Change	**Section(s) Changed**
Change: Pseudoaneurysm formation was added as an assessment and anastomotic bleeding or rupture clarified as "spontaneous" rupture. "Irritation" removed from "irritation/inflammation at the implantation site." Laboratory parameters were removed as a safety assessment, and replaced with increase from baseline in PRA. Reason: To ensure that all aneurysm data is collected in order to capture adequate safety data; to clarify that irritation should not be reported as an AESI.	Safety Assessments and Adverse Events, Section 9
Administrative Changes	
Change: Revised title of study from "A Phase 2 Study for the Evaluation of Safety and Efficacy of Humacyte's Human Acellular Vessel for Use as a Vascular Bypass or Interposition Replacement in Patients with Limb-threatening Peripheral Arterial Trauma" to "A Phase 2 Study for the Evaluation of Safety and Efficacy of Humacyte's Human Acellular Vessel for Vascular Replacement or Reconstruction in Patients with Life or Limb-threatening Vascular Trauma." Reason: To be consistent with change in study design.	Title page and Study Synopsis
Change: Updated sponsor office and personnel addresses. Reason: Sponsor has officially changed office location since last protocol.	Title page and Study Personnel, Section 1
Change: Revised "up to 3 sites" to "up to 10 sites." Reason: Additional sites are enrolling patients.	Study Synopsis
Change: Updated background information with current references. Reason: More current information regarding traumatic vascular injuries made available.	Background Information, Section 2.1; Scientific Rationale, Section 2.2

Changes Incorporated in Protocol Version 3.0 (16 Jul 2018)	
Change and Reason for Change	**Section(s) Changed**
Change: Updated overview of clinical development program. At the time the previous version was approved, 3 Phase 2 clinical studies with the HAV were ongoing, and 80 patients had received a HAV. At the time of this version, 7 clinical studies comprised the clinical development program and (as of 10 Apr 2018), 272 patients have received a HAV. Reason: Clinical development program has progressed since last version.	Summary of Clinical Studies, Section 2.4
Change: Updated overview of studies in peripheral arterial bypass patients (CLN-PRO-V002 and CLN-PRO-V004). Added presentation of preliminary results (as of 2-year follow up) and conclusions for CLN-PRO-V002. Reason: Clinical development program has progressed since last version and new data obtained since approval of last version.	Peripheral Arterial Bypass Patients, Section 2.4.2 and CLN-PRO-V002 Study Results (24 M), Section 2.4.2.1 (new section)
Change: Updated overview of studies in hemodialysis patients (CLN-PRO-V001, CLN-PRO-V003, and CLN-PRO-V006 have completed enrollment and CLN-PRO-V007 is currently enrolling patients). Added presentation of preliminary results (as of 2-year follow up) and conclusions for CLN-PRO-V001 and CLN-PRO-V003. Reason: Clinical development program has progressed since last version and new data obtained since approval of last version.	Experience in Hemodialysis Patients, Section 2.4.3; and CLN-PRO-V001 and CLN-PRO-V003 Study Results (24 M), Section 2.4.3.1 (new section)
Change: Added discussion of assessment of general host response to the HAV in study participants. The analysis (mostly of a section close to the venous anastomosis) included assessments of: ● Cellular infiltration of histotypic, inflammatory and immunological populations. ● Extracellular remodeling processes, including neo-synthesis and reorganization of ECM components that typically occur in native blood vessels. Reason: New data obtained since approval of last version.	Human Acellular Vessel Host Response and Remodeling Data, Section 2.4.4 (new section)

Changes Incorporated in Protocol Version 3.0 (16 Jul 2018)	
Change and Reason for Change	**Section(s) Changed**
Change: Updated clinical experience to date with HAV. Reason: Clinical development program has progressed since last version and new data obtained since approval of last version.	Conclusions, Section 2.4.5
Changes: Primary safety and efficacy endpoints were revised as follows: **Safety:** Incidence of aneurysm, anastomotic bleeding or rupture, HAV infection, HAV removal and irritation/inflammation at the implantation site were removed as the primary endpoint (and are now presented as a secondary endpoint, as adverse events of special interest). In addition, the following were added as adverse events of special interest: thrombosis, pseudo-aneurysm formation, and hemodynamically significant stenosis (>70% by duplex ultrasound criteria). The secondary safety endpoint of "Frequency of study conduit removal" was replaced with "HAV partial or complete removal." **Efficacy:** Incidence of limb salvage, HAV primary patency rate, HAV primary assisted patency rate, HAV secondary patency rate, and rate of HAV interventions were removed as primary endpoints (and are now presented as secondary endpoints) and replaced with HAV primary patency at 30 days. Patient survival and HAV remodeling as shown by histopathology of any clinical explants were added as secondary endpoints. Reason: Modified to account for the additional anatomic locations added to the protocol enrollment.	Study Synopsis and Study Endpoints, Section 4.2; Primary Endpoints, Section 4.2.1, Secondary Endpoints, Section 4.2.2
Change: The study population was revised to include patients with vascular trauma to size appropriate vessels in the limb or torso, requiring replacement or reconstruction (from patients with limb-threatening damage to the superficial femoral or popliteal artery who require surgical repair with an interposition bypass). Reason: Modified to account for the additional anatomic locations added to the protocol enrollment.	Study Synopsis and Description of Study Population, Section 5.1

Changes Incorporated in Protocol Version 3.0 (16 Jul 2018)	
Change and Reason for Change	**Section(s) Changed**
Change: Revision to patients with life or limb threatening traumatic injury to a vessel in the limb or torso, other than the heart, which requires replacement or reconstruction in Inclusion Criterion #1. Reason: Modified to account for the additional anatomic locations added to the protocol enrollment.	Study Synopsis and Patient Inclusion Criteria, Section 5.1.1
Change: Requirement for patient to have perioperative ultrasound or angiography or CT angiography changed to "imaging" and requirement for patient to have damage to superficial femoral artery or popliteal artery changed to "damaged vessel has a defect length of ≤ 38cm and is appropriately size matched to the 6mm HAV per the judgment of the treating surgeon taking into account vasoconstriction and situational inflow and outflow considerations" in Inclusion Criterion #2. Reason: To be consistent with change in study design and clarification of inclusion criterion.	Study Synopsis and Patient Inclusion Criteria, Section 5.1.1
Change: Inclusion Criteria #3 and #4 removed: 3. Proximal anastomosis is expected to be to the common femoral artery below the inguinal ligament or to the superficial femoral artery or popliteal artery 4. Distal anastomosis is expected to be to the SFA, popliteal artery or the tibio-peroneal trunk Reason: To be consistent with change in study design.	Study Synopsis and Patient Inclusion Criteria, Section 5.1.1
Change: Exclusion Criterion #1 was added: 1. Mangled Extremity Severity Score (MESS) of ≥ 7 Reason: To be consistent with change in study design.	Study Synopsis and Patient Exclusion Criteria, Section 5.1.2
Change: Exclusion Criterion #3 was added: 3. Catastrophic injuries that make survival unlikely (e.g. Abbreviated Injury Scale [AIS] > 5 or Injury Severity Score (ISS) >60) Reason: To be consistent with change in study design.	Study Synopsis and Patient Exclusion Criteria, Section 5.1.2

Changes Incorporated in Protocol Version 3.0 (16 Jul 2018)	
Change and Reason for Change	**Section(s) Changed**
Change: Exclusion Criterion #4 was added: 4. HAV may not be used for coronary artery repair Reason: To be consistent with change in study design.	Study Synopsis and Patient Exclusion Criteria, Section 5.1.2
Change: Exclusion Criteria #2 and #3 were deleted: 2. Life threatening head, chest, or abdominal injuries that make survival unlikely 3. Distal anastomosis planned to a tibial or pedal artery Reason: To be consistent with change in study design.	Study Synopsis and Patient Exclusion Criteria, Section 5.1.2
Change: Exclusion Criterion #8 was revised from "Previous enrollment in this study" to "Previous exposure to HAV," and #9 was added: 9. Known participation in any investigational study within the last 30 days Reason: Clarification of exclusion criteria and to align with other Humacyte study protocols.	Study Synopsis and Patient Exclusion Criteria, Section 5.1.2
Change: Description of assessment of HAV patency and integrity was revised. Previous version stated "Prior to completion of surgery, angiography is performed to confirm adequacy of the bypass anastomoses, HAV patency and peripheral runoff. New version states "Prior to completion of surgery, HAV patency is confirmed by physical exam, Doppler, angiography (conventional or intra-op CT angiography) or ultrasound. Reason: To be consistent with change in study design.	Implantation of the Humacyte Human Acellular Vessel, Section 6.4
Change: Section removed. Reason: To align with other Humacyte study protocols.	Assessment of Patient Compliance with IMP, Section 6.6

Changes Incorporated in Protocol Version 3.0 (16 Jul 2018)	
Change and Reason for Change	**Section(s) Changed**
Change: As a result of extending the Accrual Period from 12 to 24 months, the timings of evaluations in this section were clarified to be through Month 12. Revisions were made to address new cohorts. Revisions were made to imaging techniques to assess anastomotic anatomy. Reason: Due to the extended duration of the Accrual Period (from 12 months to 36 months) and changes in study design.	Clinical Evaluations Through Month 12, Section 8.1
Change: Section was removed. Reason: To align with other Humacyte study protocols. Details concerning sample collection and processing can be found in the Study Manual.	Specimen Preparation, Handling and Shipping, Section 7.2.2
Change: Section header was removed; text moved to Section 8.3.1 (Clinical and Research Laboratory Evaluations and Specimen Collection). Reason: To align with other Humacyte study protocols. Details concerning sample collection and processing can be found in the Study Manual.	Monitoring, maintenance and calibration of equipment, Section 7.2.3
Changes: Imaging criteria were revised to remove the specific requirements for conventional angiography or ultrasound. The following statements were added: ● An alternative imaging method (e.g. CTA, MRI, etc.) may be substituted for duplex ultrasound at the discretion of the investigator if it is medically appropriate and in the best interest of the patient. ● Determination of intraoperative HAV patency on Day 1 is required by physical examination, Doppler exam, angiography (conventional or intra-op CT angiography), or ultrasound at the discretion of the investigator. Duplex ultrasound examination time points were revised (now Day 30 and 3, 6, 9, 12, 24, and 36 months). Reason: To be consistent with change in study design.	Imaging Evaluations, Section 8.4

Changes Incorporated in Protocol Version 3.0 (16 Jul 2018)	
Change and Reason for Change	**Section(s) Changed**
Changes made to the Study Schedule are as follows: ● Revisions in each subsection to address change in study participants considered suitable for the study: “surgical repair of vascular injury appropriate for inclusion into the study” (previously “surgical repair of superficial femoral or popliteal artery”). ● Enrollment - Day 1 (HAV Implantation): Revision to address imaging requirement for HAV patency: “by physical examination, Doppler exam, angiography (conventional or intra-op CT angiography), or ultrasound at the discretion of the investigator” (previously by “conventional angiography or ultrasound”) ● Follow-up Visit timing revised. ● Evaluations were added to Month 12 and Early Termination: o PRA (only at early termination if prior to 6 month PRA collection) o CT angiography ● Long Term Follow-up Post Month 12 through Month 36 evaluations were added (status of patient and HAV ascertained every 3 months from post Month 12 through 36 months after HAV implantation): o Quarterly questionnaire covering status of the patient via a telephone contact with the patient and/or physician. If a suspected SAE related to HAV is discovered an unscheduled visit should be conducted to investigate. o Documentation of any HAV interventions o Adverse events (all AESI and related SAEs to be reported) o Physical exam including surgical site at Month 24 and Month 36 o Duplex ultrasound at Month 24 and Month 36 Reason: To be consistent with change in study design and due to the extended duration of the Accrual Period (from 12 months to 36 months), follow up was also extended.	Study Schedule, Section 8.5

Changes Incorporated in Protocol Version 3.0 (16 Jul 2018)	
Change and Reason for Change	**Section(s) Changed**
Changes: ● Early Termination Visit section revised to clarify that a treating physician may also withdraw subjects for safety reasons. Additionally, if withdrawal occurs after Month 12 and prior to Month 36, the patient will be asked to complete an early termination visit at which all assessments normally conducted during the long term follow up visits will be completed. With the exception of patient withdrawal, all subjects will be followed for 12 months from HAV implantation (or until HAV removal or death if earlier). ● Unscheduled Visits section revised to clarify that, if duplex ultrasound surveillance suggests the development of a ≥ 50% stenosis within the HAV, then immediate intervention is not required closer follow up should be considered. Reason: To be consistent with change in study design and due to the extended duration of the Accrual Period (from 12 months to 36 months), follow up was also extended.	Early Termination Visit, Section 8.5.5; Unscheduled Visits, Section 8.5.6
Change: Additional section was added to provide clarification of when an AE is considered serious. Reason: To align with other Humacyte study protocols.	Serious Adverse Event Definition, Section 9.2
Change: Additional section was added to provide clarification of when an AE should be considered a SUSAR. Reason: To align with other Humacyte study protocols.	Suspected Unexpected Serious Adverse Reaction, Section 9.3
Change: Additional section was added to provide definitions of events of special interest. Reason: To align with other Humacyte study protocols.	Events of Special Interest, Section 9.4

Changes Incorporated in Protocol Version 3.0 (16 Jul 2018)	
Change and Reason for Change	**Section(s) Changed**
Change: Clarifications were made to address AE reporting during the Long Term Follow Up Period post Month 12 through Month 36. The following was added to the long term follow up: ● All SAEs considered related to the HAV ● All Events of Special Interest (Section 9.4) Reason: Due to the extended duration of the Accrual Period (from 12 months to 24 months), follow up was also extended.	Reporting of Adverse Events, Section 9.5
Change: Additional section was added to provide guidance on determining causality of event to the HAV and to the index surgical procedure. Reason: To align with other Humacyte study protocols.	Criteria for Determining Causal Relationship to the HAV and Criteria for Determining Causal Relationship to the Index Surgical Procedure, Section 9.5.1
Change: Additional section was added to provide guidance on determining severity of events. Reason: To align with other Humacyte study protocols.	Criteria for Defining the Severity of an Adverse Event, Section 9.5.2
Change: Additional section was added to provide guidance on reporting of action taken with regard to AEs. Reason: To align with other Humacyte study protocols.	Reporting of Action Taken to Resolve AE, Section 9.5.3
Change: Additional section was added to provide guidance on reporting of outcomes with regard to AEs. Reason: To align with other Humacyte study protocols.	Reporting of the Outcome of the AE, Section 9.5.4

Changes Incorporated in Protocol Version 3.0 (16 Jul 2018)	
Change and Reason for Change	**Section(s) Changed**
Change: Additional section was added to provide guidance on reporting SAEs and to define reporting requirements for SAEs that occur through Month 12 and during long term follow up. Reason: To align with other Humacyte study protocols.	Reporting Serious Adverse Events, Section 9.5.5
Change: Additional section was added to provide guidance on reporting events of special interest. Reason: To align with other Humacyte study protocols.	Reporting of Events of Special Interest, Section 9.5.6
Change: Revisions were performed to provide guidance on follow up of adverse events after 1 year post implant. Reason: To align with other Humacyte study protocols.	Follow-up of Adverse Events, Section 9.5.7
Change: Updated to state that a Data Monitoring Committee (DMC) (previously Data Safety Monitoring Board) will review safety on an ongoing basis and provide recommendations about stopping, continuing or otherwise modifying the study. Details regarding responsibilities of the DMC and meeting schedule were added. Reason: To align with other Humacyte study protocols.	Data Monitoring Committee, Section 9.7

Changes Incorporated in Protocol Version 3.0 (16 Jul 2018)	
Change and Reason for Change	**Section(s) Changed**
Change: Revisions were incorporated to address change in study design from a single treatment arm study to a multi-cohort study (and subsequent changes in endpoints). In addition, the following were added to the Statistical Considerations: ● Endpoints will be assessed over a period of up to 36 months after HAV implantation. The primary analysis of the study will be conducted on the earlier of a) when the last subject enrolled reaches 30 days post-implant or b) all subjects enrolled in the initial 24 month accrual period have reached 30 days post-implant. ● PRA data will be listed and summarized using appropriate descriptive statistics for the change from baseline values. ● Sample size was increased from 20 to 40 subjects. Reason: To be consistent with change in study design; expanded enrollment in the protocol allows for additional anatomic locations for size appropriate vessels.	Study Synopsis and Statistical Considerations, Section 10
Change: Minor text clarifications were made. Reason: To improve clarity and reduce typographical errors	Throughout

A Phase 2 Study for the Evaluation of Safety and Efficacy of Humacyte's Human Acellular Vessel for Use as a Vascular Bypass or Interposition Vessel in Patients with Limb-threatening Peripheral Arterial Trauma

Medicinal Product:	Humacyte Human Acellular Vessel (Humacyte HAV)
Study No.:	CLN-PRO-V005

Sponsor:	Humacyte, Inc
Address:	7020 Kit Creek Road
	PO Box 12695
	Research Triangle Park, NC 27560
Phone:	919-313-9633
Fax:	919-313-9634

CRO:	Atlantic Research Group
	2421 Ivy Road, Suite 200
	Charlottesville, VA 22903

Version:	2.0
	24 October 2016

Confidentiality Statement

Statement of Compliance

This trial will be conducted in compliance with the protocol and the following regulatory requirements:

- Declaration of Helsinki adopted by the 18th World Medical Assembly in Helsinki, Finland, in 1964, as last amended by the World Medical Assembly in 2013
- International Conference on Harmonization of Technical Requirements for Registration of Pharmaceuticals for Human Use (ICH), E6 Good Clinical Practice: Consolidated Guidance (ICH E6)
- ICH E2A Clinical Safety Data Management: Definitions and Standards for Expedited Reporting
- ICH E8 Guidance on General Considerations for Clinical Trials
- Applicable sections of United States Food and Drug Administration (FDA) Code of Federal Regulations (CFR), including:
    - 21 CFR Part 11, Electronic Records; Electronic Signatures
    - 21 CFR Part 50, Protection of Human Subjects
    - 21 CFR Part 54, Financial Disclosure by Clinical Investigators
    - 21 CFR Part 56, Institutional Review Boards
    - 21 CFR Part 312, Investigational New Drug Application

Principal Investigator Agreement Page for the Protocol

I agree:

- To assume responsibility for the proper conduct of the study at this site, and to conduct the study in compliance with this protocol, any future amendments, and with any other study conduct procedures provided by the sponsor, Humacyte, Incorporated (Humacyte), or their authorized representatives.

- Not to implement any deviations from or changes to the protocol (including protocol amendments) without agreement from the sponsor and prior review and written approval from the Institutional Review Board (and FDA, if applicable) except where necessary to eliminate an immediate hazard to the patient(s), or for administrative aspects of the study (where permitted by all applicable regulatory requirements).

- That I am familiar with the appropriate use of the investigational medicinal product, as described in this protocol and any other information provided by the sponsor including, but not limited to the current Investigator's Brochure or equivalent document provided by Humacyte.

- To ensure that all persons assisting me with the study are adequately informed about the investigational medicinal product and of their study-related duties and functions.

- That I have been informed that certain regulatory authorities require the sponsor to obtain and supply details about the investigator's ownership interest in the sponsor or the Investigational Medicinal Product, and more generally about his/her financial ties with the sponsor. Humacyte will use and disclose the information solely for the purpose of complying with regulatory requirements.

Principal Investigator: _____

Name and Title

Signed: _____ Date: _____

Protocol Approval

Sponsor Medical Approval: Jeffrey H. Lawson, M.D. Ph.D. Chief Medical Officer, Humacyte

Name and Title

Signed:  Date: 26-OCT-2016

Table of Contents

List of In-text Tables

List of Abbreviations

AE	Adverse event
AIDS	Acquired immunodeficiency syndrome
ALT	Alanine aminotransferase
aPTT	Activated partial thromboplastin time
ASA	Acetylsalicylic acid (aspirin)
ABI	Ankle brachial index
AST	Aspartate aminotransferase
AV	Arteriovenous
AVF	Autologous arteriovenous fistula
BP	Blood pressure
CAVG	Canine acellular vascular graft
CBC	Complete blood count
CKD	Chronic kidney disease
CTA	Computed tomography angiography
CM	Concomitant medication
eCRF	Electronic case report form
CRO	Contract research organization
DSMB	Data Safety Monitoring Board
DTH	Delayed-type hypersensitivity
ECG	Electrocardiogram
ECM	Extracellular Matrix
ePTFE	Expanded polytetrafluoroethylene
ESRD	End-stage renal disease
ET	Early termination
FDA	Food and Drug Administration
GCP	Good Clinical Practice
GGT	Gamma glutamyl transpeptidase
GLP	Good Laboratory Practice
HAV	Human acellular vessel
HIV	Human immunodeficiency virus
IB	Investigator Brochure
ICF	Informed consent form
ICH	International Conference on Harmonization of Technical Requirements for Registration of Pharmaceuticals for Human Use
IgG	Immunoglobulin G

List of Abbreviations

IHC	Immunohistochemistry
IM	Intramuscular
IMP	Investigational medicinal product
INR	International normalized ratio
IRB	Institutional Review Board
ISO	International Organization for Standardization
IU	International unit
IV	Intravenous
MedDRA	Medical Dictionary for Regulatory Activities
M	Month
N	Number (typically refers to participants)
NYHA	New York Heart Association
OTC	Over-the-counter
PAD	Peripheral arterial disease
PE	Physical examination
PHI	Protected health information
PI	Principal Investigator
PRA	Panel reactive antibodies
PT	Prothrombin time
PTFE	Polytetrafluoroethylene
QA	Quality Assurance
QC	Quality Control
RRT	Renal replacement therapy
SAE	Serious adverse event
SFA	Superficial Femoral Artery
SOP	Standard operating procedure
SVS WIfI	Society for Vascular Surgery: Wound, Ischemia, and foot Infection
US	Ultrasound
USA	United States of America
WFI	Water for injection
WBC	White blood cell(s)
WHO	World Health Organization

Protocol Summary

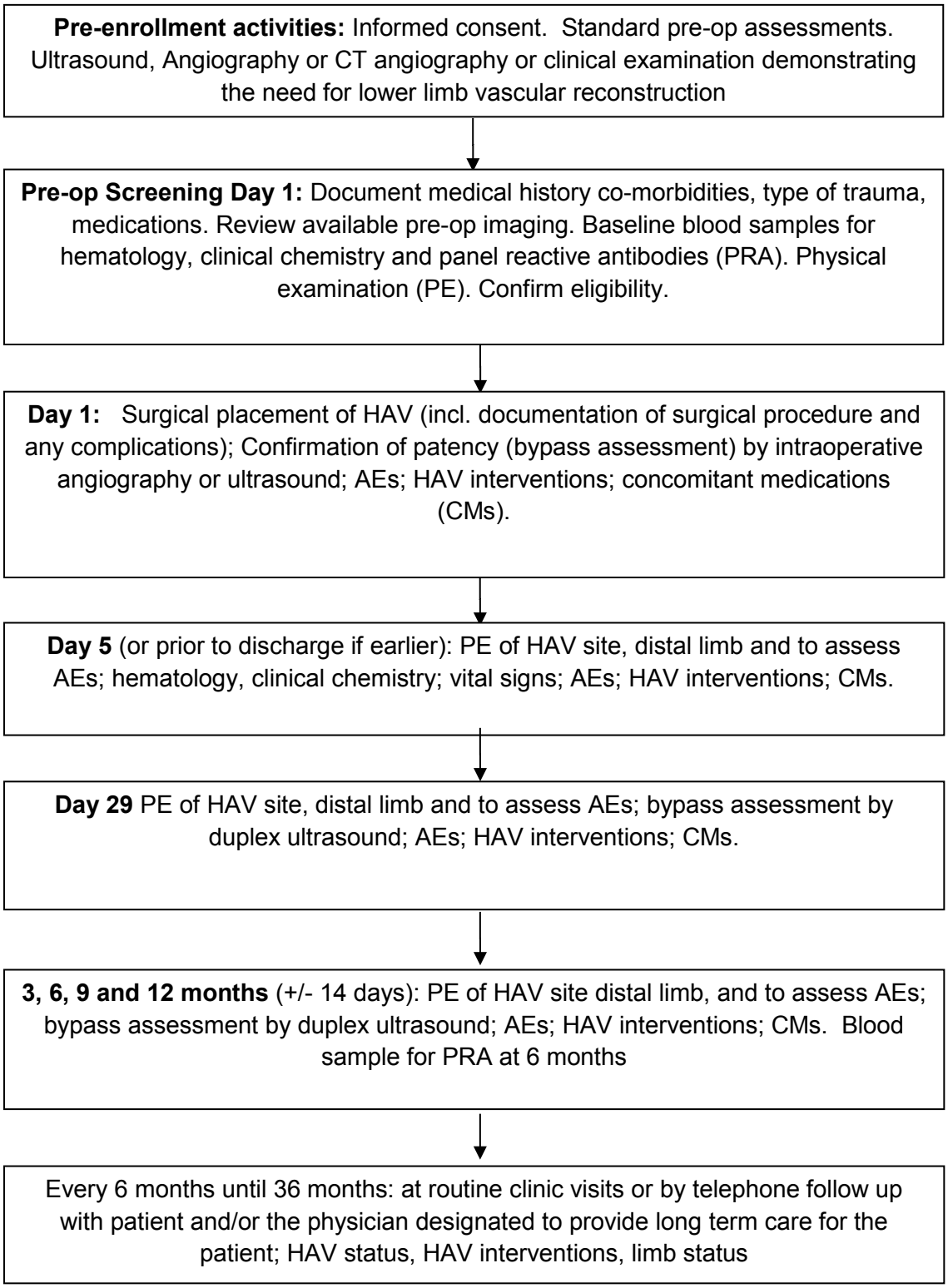
Full Title	A Phase 2 Study for the Evaluation of Safety and Efficacy of Humacyte's Human Acellular Vessel for Use as a Vascular Bypass or Interposition Vessel in Patients with Limb-threatening Peripheral Arterial Trauma
Clinical Trial Phase	Phase 2
Sponsor	Humacyte, Inc.
Planned Study Sites	Up to 3 sites to include: Baltimore Shock Trauma, Baltimore, MD Johns Hopkins, Baltimore, MD Harborview Medical Center, Seattle, WA
Sample Size	Up to 20 patients
Study Population	Patients with peripheral arterial trauma
Inclusion Criteria	1. Patients with lower limb vascular trauma which threatens the viability of the leg and who require reconstruction of the superficial femoral or popliteal artery 2. Preoperative ultrasound or angiography or CT angiography or clinical examination indicates damage to the superficial femoral artery (SFA) or popliteal artery requiring reconstruction with an interposition bypass vessel AND required bypass length of ≤ 38cm 3. Proximal anastomosis is expected to be to the common femoral artery below the inguinal ligament or to the superficial femoral artery or popliteal artery 4. Distal anastomosis is expected to be to the SFA, popliteal artery or the tibio-peroneal trunk 5. Autologous vein graft is either not feasible in the judgment of the treating surgeon (e.g. because of lack of availability of suitable conduit, presence of severe venous insufficiency) or is not desirable because of the urgency of revascularization 6. Aged 18 to 85 years old, inclusive 7. Able to communicate meaningfully with investigative staff and

	able to comply with study procedures. If the patient is unconscious then information from a reliable witness indicates that the patient would normally be able to understand and comply with study procedures 8. Patient or legal representative is able, willing and competent to give informed consent 9. Life expectancy of at least 1 year
Exclusion Criteria	1. Limb at high risk of amputation despite vascular reconstruction e.g., because of crush injury 2. Life threatening head, chest, or abdominal injuries that make survival unlikely 3. Distal anastomosis planned to a tibial or pedal artery 4. Pregnant women 5. Known medical condition which would preclude long term dual antiplatelet therapy after resolution of acute injuries 6. Any other condition which in the judgment of the investigator would preclude adequate evaluation of the safety and efficacy of the Humacyte Human Acellular Vessel (HAV) 7. Previous enrollment in this study 8. Employees of the sponsor or patients who are employees or relatives of the investigator
Expected Enrollment Start	2Q 2016
Accrual Period	12 months
Study Duration	The active study duration for each study participant will be 12 months from HAV implantation or until HAV failure/ removal/ death if earlier. Additional follow up at 6 month intervals until 36 months will involve the capture of information on assessments performed at “standard of care” routine clinic visits or by telephone follow up with the patient or his/her physician. There are no study specific visits or procedures after 12 months. The total expected duration of the clinical study is 24 months (plus 2 years additional follow up at routine clinical visits or via telephone contact with the patient and/or the physician designated to continue long term care of the patient).

Study Design	Prospective, multicenter, single arm, non-randomized study
Investigational Device/Intervention Description	Patients will be implanted with a Humacyte Human Acellular Vessel (HAV) as an interposition vessel or bypass using standard vascular surgical techniques.
Primary Objectives	**Safety** To evaluate the safety and tolerability of the Humacyte HAV in patients with vascular trauma in the lower limb who are undergoing vascular reconstructive surgery **Efficacy** To determine the patency (primary, primary assisted and secondary) rate of the Humacyte HAV at 12 months To determine the rate of limb salvage
Secondary Objectives	**Safety** • To determine mechanical stability of the HAV based on freedom from aneurysmal degeneration on duplex ultrasound • To determine HAV durability in terms of freedom from need for HAV explantation or replacement due to infection, bleeding, or conduit degeneration **Efficacy** • To determine the patency of the HAV (primary, primary assisted and secondary) at 3, 6 and 9 months • To determine the rates of interventions needed to maintain / restore patency in the HAV over 12 months
Endpoints	All endpoints will be assessed over a period of up to 12 months after HAV implantation. The primary analysis of the study will be based on the 12 month follow up of all patients who receive an HAV.

	Safety Endpoints: • Incidence of aneurysm formation, anastomotic bleeding or rupture, HAV infection, HAV removal, and irritation/inflammation at the HAV implantation site • Frequency and severity of adverse events Efficacy Endpoints: • Incidence of limb salvage • HAV patency rates (i.e., primary, primary assisted, and secondary) • HAV interventions Long term Endpoints (Every 6 months until 36 months): • Limb viability • HAV survival • Patient survival
Protocol Approval (Version and Date)	Version 2.0 24 October 2016

Schematic of Study Design:



1. STUDY PERSONNEL

An updated list of all study personnel will be maintained by the CRO. Protocol amendments will not be required for staff changes at Humacyte, the CRO or the sites (except change of Principal Investigator at a site).

CRO Project Manager:

Brad Whitlow
Project Manager
Atlantic Research Group
2421 Ivy Road, Suite 200
Charlottesville, VA 22903

Email: bwhitlow@atlanticresearchgroup.com
Mobile: 828-442-0526
Office: 434-202-4537
Fax: 434-202-4537

Sponsor Medical Representative:

Jeffrey H. Lawson, MD, PhD
Chief Medical Officer
Humacyte, Inc.
PO Box 12695
7020 Kit Creek Road
Research Triangle Park, NC 27709, USA

Email: lawson@humacyte.com
Mobile: 919-630-7851
Office: 919-313-9633 ext. 130

Sponsor Regulatory Representative:

William Tente
Vice President Regulatory Affairs
Humacyte, Inc.
PO Box 12695
7020 Kit Creek Road
Research Triangle Park, NC 27709, USA

Email: tente@humacyte.com
Mobile: 401-714-4151
Office: 919-313-9633 ext. 236

Sponsor Clinical Operations Representative:

Angela Rose
Director, Clinical Development
Humacyte, Inc.
PO Box 12695
7020 Kit Creek Road
Research Triangle Park, NC 27709, USA

Email: rose@humacyte.com
Mobile: 919-667-3507
Office: 919-313-9633 ext. 185

CRO Safety Representative:

Audra Rodrigues
Sr. Drug Safety Specialist
Atlantic Research Group
2421 Ivy Road
Charlottesville, VA 22903

Email: safety@atlanticresearchgroup.com
Phone: 888-619-3216
Fax: 866-851-9318

Medical Monitor:

Jennifer Prichard, MD
Medical Monitor
Atlantic Research Group
2421 Ivy Road
Charlottesville, VA 22903

Email: jprichard@atlanticresearchgroup.com
Mobile: 434-249-5530
Phone: 888-619-3216
Fax: 866-851-9318

2. BACKGROUND INFORMATION AND SCIENTIFIC RATIONALE

2.1 Background Information

In modern combat, the incidence of vascular injury is much greater than in previous wars. The rate of vascular injury in the Vietnam War was 2-3%. But between 2002-2009, the rate of vascular injury was over 12% in a study of over 13,000 battlefield injuries. (Rasmussen, 2010] Improvised Explosive Devices, or IED's, cause more than 3,000 casualties per year – and this number is rising. (Cordesman, 2010) Arterial damage, laceration and thrombosis can require vascular reconstruction to save tissues from ischemia, necrosis, and further amputation. There are two current methods for vascular reconstruction: harvesting of autologous vein, or using synthetic graft materials, and each has important limitations. Harvesting autologous vein for vascular reconstruction is problematic because IED casualties often have multi-limb injuries, making harvest of autologous vein highly risky or impossible. (Holcomb, 2011) Synthetic vascular reconstruction using synthetic vascular grafts made from Teflon (ePTFE)/Dacron is relatively contraindicated, since IED wounds are always "dirty", and bacteria in the wound can colonize the synthetic graft, causing abscesses and sepsis.

In the civilian population, lower extremity bone fractures with associated arterial injuries are common, due to motor vehicle accidents, gunshot wounds, dog bites, and other situations resulting in blunt or penetrating trauma. (Helfet, 1990, Andrikopoulos, 1995, Akingba, 2012) In fact, the incidence of this type of vascular injury has increased considerably in the past 50 years. (Andrikopoulos, 1995) Although this type of injury represents less than 1% of all civilian injuries, fractures with associated vascular damage require special attention because of their potentially severe complications, including limb necrosis and amputation. Currently, in order to attempt to salvage the injured limb, the vascular component of these injuries are treated with interposition or bypass grafting, and these grafts are usually created either using the patient's saphenous vein or a synthetic ePTFE graft. However, the use of these grafts in the civilian population has many of the same downsides that accompany their use in combat injuries. The patient may not have adequate saphenous vein for harvest, and in many of these cases, such as dog bite injuries, the wound is "dirty" and a synthetic vascular substitute such as ePTFE is contraindicated due to the risk of infection. (Akingba, 2012) Thus, civilians would also benefit from a vascular graft that is available "off-the-shelf" – one that does not contain synthetic material and that has the same properties as human tissue, but does not require harvesting of vessel or cells from the patient.

There is thus a need for alternative grafts, which more closely mimic human vascular tissue that may avoid or reduce the complications associated with ePTFE and Dacron.

2.2 Scientific Rationale

Humacyte, Inc. (Humacyte) has developed an acellular, human collagen-based vascular conduit (human acellular vessel – HAV) to provide an alternative to synthetic materials and to autologous grafts in the creation of vascular access for dialysis and for use in peripheral vascular bypass surgery or as an interposition bypass to repair traumatic arterial damage. Because this product mimics native vascular tissue, it possesses all of the advantages of an autologous graft; it also has the benefits of synthetic grafts in that it is available off-the-shelf. Use of an off-the-shelf product avoids the surgical morbidity associated with vein graft harvest and most importantly allows vessel bypass surgery in patients who have no suitable veins available. Because the product mimics native vessel, it does not have the compliance mismatch associated with synthetic alternatives. In addition, pre-clinical studies in pigs, canines and primates have shown that the HAVs resist intimal hyperplasia at the anastomoses. (Quint, 2011, Prichard, 2011, Dahl, 2011) Upon implantation, it is anticipated (based on pre-clinical studies) that the collagen matrix comprising the HAV will be infiltrated with host cells and remodeled by the host. This will result in a vascular structure more similar to the histological composition of the native vascular tissue that may improve bypass longevity and be less likely to become infected. The latter potential advantage is of high importance in the repair of peripheral vascular trauma where most wounds are heavily contaminated.

2.3 Summary of Nonclinical Information

The non-clinical testing program was designed to comprehensively address:

- local and systemic effects of the product in multiple in vivo animal models acutely and chronically,
- functional aspects of product implanted into animal models as an arteriovenous conduit
- biocompatibility of the HAV material in standardized in vitro and in vivo test protocols.

Overall, the results of these studies indicated that the HAV extracellular matrix material was non-toxic, well tolerated, and met standards for biocompatibility. Generally, the HAVs functioned as intended and maintained patency during the implantation period. (See the Investigator Brochure for a detailed summary of non-clinical data.)

Pre-implantation, the HAV has mechanical properties (burst pressure and suture retention strength) comparable with native human artery and vein. (**Table 1**.) There was no evidence that HAV strength deteriorated after long-term implantation into baboons.

Table 1: Summary of Mechanical Properties of Explanted Acellular Vessels

Test Material	Burst Pressure (mm Hg)	Suture Strength (g)
Pre-Implant Humacyte HAVs	3415 +/- 1011 (n=4)	180 +/- 44 (n=12)
Post-Explant Humacyte HAVs	3669 +/- 1305 (n=5)	276 +/- 84 (n=11)
Human Saphenous Vein	1,680 – 2,273[a]	196 +/- 2 (n=7)[a]
Human Artery	2,031 – 4,225[a]	200 +/- 119 (n=9)[a]

[a] From L'Heureux et al, *Nature Medicine*, 2006. (L'Heureux, 2006)

In the chronic animal testing Humacyte vessels produced using canine cells were implanted into 12 dogs (canine acellular vascular graft, CAVG) and 14 baboons (human acellular vessel -HAV) in a variety of anatomical locations. In general, the Humacyte vessels were safe and well tolerated, and functioned as intended.

Mechanical failure was not observed in any HAV. Calcification was not observed in any CAVG or HAV. No graft exhibited hemodynamically significant intimal hyperplasia. Unlike with ePTFE graft implantation, no evidence of systemic infection attributable to implantation of HAV was observed in any of the animals. One HAV developed an aneurysm that was resected and did not harm the animal. The HAV material showed no evidence of toxicity in hematology, clinical chemistry, and necropsy data. The HAVs could be accessed by venipuncture and hemostasis was achieved following needle puncture.

On microscopic analysis, the HAVs were found to be well integrated into the host tissue. Overall, the cellular host response to the HAVs demonstrated smooth muscle actin-positive cells within the vessel wall, endothelial cells lining the lumen, and an adventitial-like outer layer adjacent to the vessel. These findings indicate that implanted HAVs were populated with cell types that are characteristic of healthy native vasculature. Examination of the anastomotic sections showed that the HAVs were well integrated with adjoining vasculature with minimal intimal hyperplasia observed. Furthermore, IHC was employed to identify CD-68 positive macrophages in the venous intimal tissue. Studies have shown a substantial macrophage population has been observed within venous intimal tissue adjacent to inflammatory ePTFE arteriovenous grafts. (Kelly, 2002, Roy-Chaudhury, 2001) Only sparse CD-68 positive macrophages were observed, indicating that the degree and the aggressiveness of the intimal hyperplasia associated with the HAV were less than that typically associated with ePTFE grafts. (Prichard, 2011)

Over time, the organization and composition of extracellular matrix (ECM) components indicated that, aided by infiltration of host vascular cells, HAVs were remodeled in vivo in a manner that mimicked the dynamic remodeling process of native blood vessels. Given the difficulties associated with the baboon animal model, where mismatches in vein vs graft diameter were encountered and animals perturbed their wounds postoperatively, an overall assisted patency rate of approximately 80% (11/14) was achieved. In a xenogeneic transplant model that did not employ immunosuppression, the HAV material did not elicit biologically significant cellular or delayed-type hypersensitivity (DTH) immune responses. All animals developed immunoglobulin G (IgG) titers to the HAV material that did not appear to detrimentally impact vessel function.

In internationally recognized in vitro and in vivo International Organization for Standardization (ISO) test protocols, the HAV material met criteria for biocompatibility required of medical devices.

These data collectively support the safety of the HAV for the proposed clinical investigation.

2.4 Summary of Clinical Studies

2.4.1 Overview

Three phase 2 clinical studies using the HAV have completed enrollment with available results, two in vascular access for hemodialysis and one in peripheral arterial bypass surgery. Eighty patients have been implanted with an HAV with the longest follow up being more than 42 months. More information on the clinical profile of the HAV in these ongoing studies is provided in the Investigator Brochure.

Currently enrolling is a phase 3, prospective, multicenter, multinational, open-label, randomized, two-arm study comparing the HAV with ePTFE grafts in subjects with end-stage renal disease who require hemodialysis and are targeted for implantation of an AV graft for dialysis access. The primary endpoint is time to loss of secondary patency from implantation, and the primary analysis will be conducted when all subjects remaining on study have completed Month 12. Enrollment commenced in May 2016 with a target enrollment of approximately 350 subjects across all study sites in the United States, Israel, United Kingdom, Poland, Germany, and Portugal.

An additional phase 2, prospective, multicenter, single-arm, open-label study evaluating the HAV as a vascular prosthesis for femoro-popliteal bypass in up to 20 patients with peripheral arterial disease in the United States is expected to begin enrolling patients in Q4 2016.

2.4.2 Peripheral Arterial Bypass

A study is ongoing in Poland that is evaluating the HAV as femoro-popliteal bypass in patients with peripheral arterial disease (PAD). Recruitment began in October 2013 and was completed in June 2014 with 20 patients implanted. Follow up of patients continues in patients with patent HAVs up to five years post-implantation.

This is a single group uncontrolled study being conducted at 3 sites in Poland. Eligible patients require a femoro-popliteal bypass graft for the management of symptomatic peripheral arterial disease. Pre-operative imaging (angiography or CT angiography) must have demonstrated at least two below knee vessels patent to the ankle with good runoff. The proximal anastomosis was expected to be below the inguinal ligament and the distal anastomosis above the knee. Autologous vein grafts must not have been suitable or feasible (e.g., because of severe venous

disease or prior use of leg veins for other bypass surgery or there is a clinical need to preserve those veins for future bypass surgery in the coronary or peripheral circulation).

The HAV was implanted using standard vascular surgical techniques and the patency of the bypass confirmed by intraoperative angiography or ultrasound. The patient was then followed up at study visits at 15 days, 6 weeks and 3, 6, 12, 18 and 24 months. Dual antiplatelet therapy with aspirin and clopidogrel is continued while the HAV is in situ. From 24 to 60 months there are no study-specific visits but the status of the patient and the HAV status is ascertained yearly at routine clinical visits or by phone calls to the patients or their primary care physician. At each visit safety is assessed by clinical examination and adverse events, and the HAV is examined using duplex ultrasound to visualize the entire length to confirm patency, flow and to detect stenosis, aneurysm development or dilatation.

The primary objectives of the study are to evaluate the safety and tolerability of the Humacyte HAV in PAD patients undergoing above-knee femoro-popliteal bypass surgery and to determine the patency (primary, primary assisted and secondary) rate of the Humacyte HAV at 24 months. Secondary objectives include assessment of the panel reactive antibodies (PRA)) and IgG response to the HAV and to assess patency (primary, primary assisted and secondary) at 6, 12 and 18 months, to determine the rates of interventions needed to maintain / restore patency in the HAV, to assess any effect of implantation on claudication, rest pain and ischemic ulcers and to assess any effect on ankle-brachial index (ABI).

An interim analysis of the study was conducted in September 2015. At that time, the duration of follow up ranged from 14 to 22 months with a total of about 26 patient years of follow up.

The majority of the patients recruited into the study (65%) are male and all are Caucasian. The mean age is 66 years (range 54 to 79 years) and 45% have a history of diabetes while 50% have a history of atherosclerotic disease in the coronary and/or carotid circulation. Five patients (25%) are obese with a BMI >30Kg/M^2.

There has been one death during the study, which was considered to be unrelated to the HAV. This 68 year old male with a history of peripheral arterial disease (since 2013), diabetes, asthma, hypertension and ulcerative colitis, underwent implantation of an HAV as a left femoro-popliteal arterial bypass on 22 May 2014. The patient was discharged from hospital in good condition and returned for the 15 day study visit on 9 June. At this visit the patient was in good general health with no fever or signs of systemic infection. Inspection of the surgical wound

revealed a local superficial infection at the lower end of the surgical incision which did not appear to extend down to the HAV. The HAV was patent on ultrasound examination with good flow. At this visit the patient had a white cell count of 12.56G/l (normal range 4-10). The patient refused immediate hospital admission for local wound management and for control of a very elevated blood glucose (value unknown) for personal reasons but agreed to return to the hospital within the next few days for wound cleansing and VAC therapy. The following day (10 June 2014) the patient was found dead at home. The death was reported to the investigator by the patient's sister on 7 July 2014. The cause of death was reported as cardiorespiratory collapse by the patient's primary care physician, but no autopsy was done.

Seven patients have experienced adverse events, which have affected HAV patency. Two of these patients were found to have occluded HAVs at routine study visits (at 12 and 26 weeks respectively). Non-compliance with dual antiplatelet therapy may have contributed to one of these occlusions. Because these patients did not report significant symptoms associated with the HAV occlusions they have been managed conservatively and the HAVs are considered to have lost both primary and secondary patency.

Five other patients experienced serious adverse events (SAEs) involving loss of primary patency of the HAV (4 thromboses and 1 pseudoaneurysm). In four of these cases (three thrombosis and one pseudoaneurysm) the patient was managed surgically or endovascularly and the HAVs still have secondary patency. The treated thromboses occurred at 20, 26, and 48 weeks after implantation and in each case a stenosis of the HAV or the HAV / artery anastomosis was identified and treated by angioplasty. The pseudoaneurysm was identified at the 12 week visit and was managed by replacement of a short section of the HAV with an interposition ePTFE graft. Histology of the resected HAV did not show any evidence of inflammation or infection and the hole in the HAV wall may have resulted from iatrogenic damage at a previous thrombectomy attempt with a Fogerty catheter. At that thrombectomy attempt – performed during surgical management of a post-operative hematoma at the site of a femoral endarterectomy -no thrombus was found in the HAV. In the fifth case the HAV was found to be occluded at the 12 month visit and the HAV was abandoned and replaced with an ePTFE graft.

Kaplan-Meier analysis of patency indicates a primary patency of 74% at 6 months and 63% at 12 months. Secondary patency is 89% at both 6 and 12 months. These patency rates are comparable to those reported with synthetic grafts used for femoro-popliteal bypass.

2.4.3 Vascular Access for Dialysis

The HAV is currently under evaluation as a vascular access for dialysis in two phase 2 clinical studies in patients with end-stage renal disease (ESRD), one in Poland and one in the United States (US). These studies are of very similar design, including patients with ESRD who were not suitable candidates for creation of an arteriovenous fistula (AVF), usually because of unsuitable vasculature or failure of previous attempts to create an AVF. The HAV was placed as an upper arm conduit. Patients were followed up with monthly visits and ultrasound surveillance of the HAV for the first 6 months and then less frequently out to two years. Provided that the HAV was functioning well and the surgical wounds healed, the HAV was used for dialysis from 4 - 8 weeks after implantation. Dialysis was initiated with small needles (as for a new AVF) with gradual increase in needle size over the first few weeks of use. The primary objective of the studies is to evaluate safety of the HAV and to assess patency at 6 months. Secondary objectives include measurement of any panel reactive antibody (PRA) response and evaluation of patency and need for interventions to maintain or restore that patency over 2 years.

The US study is being performed under IND 15,263. Enrollment started in June 2013 and was completed in June 2014 with 20 patients implanted.

The Polish study was initiated in December 2012 and the final (40^{th}) patient was implanted in April 2014.

An interim analysis of the combined data from these studies was performed in May 2015. A total of 60 patients (40 in Poland and 20 in the US) had been implanted with HAVs for dialysis access. Baseline characteristics of the two study populations were similar though the proportion of female patients was higher in the US (65% as compared to 45% in Poland), and the incidence of hypertension, diabetes, and cardiovascular disease was higher in the US population. In the US, 65% of patients were African-American, while all of the patients in Poland were Caucasian. In both studies, patients had multiple prior accesses for hemodialysis, ranging from 1-9 in the Poland, and from 1-6 in the US.

There have been 6 deaths in the two studies, three during ongoing follow up, one of a patient who withdrew from the study having decided to discontinue dialysis and two within 1 month after HAV abandonment. None of the deaths were considered related to the HAV and the death rate during the study is comparable with that observed generally in the ESRD population. One

patient underwent a renal transplant during the study and the renal allograft is functioning well 12 months after transplantation.

The HAV rates of primary, primary assisted, and secondary patency at 6 months were 63%, 72%, and 97%, respectively. At 12 months, patency rates were 28%, 38%, and 89%, respectively. While primary and primary assisted patency rates are comparable to those reported for ePTFE in recent randomized trials, secondary patency of 97% at 6 months and 89% at one year is greater than prior reported ePTFE values, which range from 55-65%. (CDRH 510K Gore Acuseal, 2013, Dixon, 2009) Mean blood flow rates in the HAVs as measured by ultrasound were typically over 1.0 L/min and did not tend to decrease over time. Mean diameters of HAVs, as measured by ultrasound, remained overall stable, without evidence of substantial dilatation or narrowing. Dialysis centers reported that the HAVs were straightforward to cannulate using standard techniques.

Three access-associated infections have been reported. In only one case was the HAV material demonstrated to be infected and required partial resection. In this ESRD patient methicillin-resistant Staphylococcus aureus was isolated from a segment of resected HAV. In two other ESRD patients infection associated with the access has been reported but did not appear to involve the HAV. In one patient, a perigraft hematoma became infected but the HAV was salvaged with use of parenteral antibiotics. In the second patient, the infection involved an ePTFE jump graft (inserted during revision of the venous anastomosis after thrombosis of the HAV) but not the remaining HAV. HAV

Overall, HAVs have been abandoned in ten patients. Two HAV abandonments were associated with over-dilation with 8-mm high-pressure balloons, which disrupted the 6-mm diameter HAV material. In one case, the HAV was ligated and resected to manage a suspected case of ischemic monomelic neuropathy. In the other seven instances, the HAVs were abandoned due to stenosis in the run-off vein or the venous anastomosis, or due to recurrent thromboses. None of the HAVs were abandoned due to infection.

Biochemical analysis of the HAV indicates that it contains trace or non-detectable levels of cellular proteins such as beta-actin and human leukocyte antigen I and is thus potentially suitable for use in any patient without the need for tissue matching. To confirm this lack of immunogenicity blood samples for measurement of PRA are being collected from all patients in the pilot studies. At six months after implantation of the HAV only three patients showed any increase in PRA values. All three patients had histories of previous failed renal transplants. In two cases the rise in PRA occurred shortly after discontinuation of immunosuppressive therapy.

In the third case the patient was an infirm elderly lady with multiple medical problems and generalized debility. In none of these patients was there evidence of an inflammatory response to the HAV or any complication with the HAV coinciding with the PRA increases.

2.4.4 Conclusions

Early clinical experience with the HAV indicates that it functions as intended and remains mechanically strong (even after repeated puncture for hemodialysis) over implantation periods of more than 42 months with no evidence of dilatation. The safety profile has been favorable with no serious, unexpected adverse events attributable to the HAV in the hemodialysis and peripheral arterial disease populations. In more than 90 patient years of follow up across the three pilot studies (ESRD and PAD) only one case of infection of the HAV material itself has been reported, suggesting that the HAV may be less prone to infection compared to currently available ePTFE grafts. Primary patency of the HAV appears to be comparable to that observed with synthetic grafts, and secondary patency is higher than historically observed rates for ePTFE. No evidence of immunogenicity of the HAV has been observed.

These data support the use of the HAV in this proposed phase 2 study in peripheral vascular trauma.

2.5 Potential Risks and Benefits

2.5.1 Potential Risks

It is anticipated that subjects participating in the study will be exposed to similar risks to those associated with other arterial conduits. Risks associated with the study investigational product may include but are not limited to:

- Thrombosis/occlusion of the conduit or host vessels, with consequent limb ischemia
- Embolism from a thrombosed conduit
- Bleeding and hematoma formation at the surgical site
- Infection – at the surgical site or systemic
- Stenosis of the conduit or its anastomoses
- Aneurysm or pseudoaneurysm formation
- Swelling of the limb

Regular clinical examination of the HAV implantation site and assessment of the patency, blood flow and diameter using ultrasound during the study should allow early detection of complications and permit appropriate intervention including HAV explantation.

The HAV is grown using donor human aortic smooth muscle cells. The vessel is decellularized during manufacturing and thus consists of human extracellular matrix proteins. It is possible that the HAV may provoke an immune response which may lead to damage to the HAV and possible cross reactivity against host proteins.

2.5.2 Potential Benefits

Patients who undergo implantation of the Humacyte HAV may benefit from improved patency resulting in a reduced number of interventions versus a conventional expanded PTFE (ePTFE) or a Dacron graft. This may result from a decreased propensity for anastomotic and downstream neointimal hyperplasia, which often leads to graft occlusion with synthetic grafts. In addition, the risks such as infection listed in section 2.5.1 typically encountered with conventional synthetic grafts may be decreased with the Humacyte HAV. Finally, the longevity of the Humacyte HAV may be greater than that of conventional synthetic grafts.

2.5.3 Risk-Benefit Rationale

The risks anticipated in this study are similar to those associated with currently marketed prosthetic grafts used for peripheral vascular repair. The potential advantages of the HAV compared to currently marketed grafts may lead to a lower complication rate and reduced need for surgical intervention and graft replacement.

Recruitment will be restricted to a maximum of 20 subjects who receive implants to provide safety data prior to the planning and initiation of further studies

There is no formal hypothesis testing in this study but data from the study will be compared with historical data on synthetic peripheral bypass grafts to assess the safety and efficacy of the HAV prior to initiation of larger studies.

3. STUDY OBJECTIVES

3.1 Primary Objectives

This is an open label phase 2 study. There is no formal hypothesis testing.

Safety:

- To evaluate the safety and tolerability of the Humacyte HAV in patients with vascular trauma in the lower limb undergoing vascular reconstructive surgery

Efficacy:

- To determine the patency (primary, primary assisted and secondary) rate of the Humacyte HAV at 12 months
- To determine the rate of limb salvage

3.2 Secondary Objectives

Safety:

- To determine mechanical stability of the HAV based on freedom from aneurysmal degeneration on duplex ultrasound
- To determine HAV durability in terms of freedom from need for bypass conduit explantation or replacement due to infection, bleeding, or conduit degeneration

Efficacy:

- To determine the patency of the HAV (primary, primary assisted and secondary) at 3, 6 and 9 months
- To determine the rates of interventions needed to maintain / restore patency in the vessel over 12 months

4. STUDY DESIGN

4.1 Description of the Study Design

Prospective, multicenter, single arm, non-randomized phase 2 study

4.2 Study Endpoints

All endpoints will be evaluated at multiple time points over 12 months after HAV implantation. However, the main analysis of the study will be based on the 12-month follow up of all patients who receive a Humacyte HAV.

4.2.1 Endpoints

Safety:

- Incidence of aneurysm formation, anastomotic bleeding or rupture, HAV infection, HAV removal and irritation/inflammation at the implantation site
- Frequency and severity of adverse events

Efficacy:

- Incidence of limb salvage
- Primary patency rate
- Primary assisted patency rate
- Secondary patency rate
- HAV interventions

4.2.2 Long Term Endpoints

To be evaluated at “standard of care” routine clinical visits or by telephone contact with the patient and/or physician every 6 months until 36 months

- Limb viability
- HAV survival

- Patient survival

4.3 Duration of Study Participation

For an individual subject, the expected duration of study participation is approximately 12 months. Enrollment (accrual) is expected to occur over 12 months. Additional data on patient and HAV status will be collected at routine clinical visits (standard of care for peripheral vascular surgical patients) at 6 month intervals up until 36 months after HAV implantation but there are no study-specific visits or procedures after 12 months. If the patient is not scheduled to return to the investigational site for these visits (e.g. for geographical reasons) then the post 12 month follow up may be conducted by telephone contact with the patient and/or the physician designated to continue long term care of the patient.

5. STUDY POPULATION

5.1 Description of the Study Population

The study population will consist of patients with limb-threatening damage to the superficial femoral or popliteal artery who require surgical repair with an interposition bypass.

5.1.1 Patient Inclusion Criteria

1. Patients with lower limb vascular trauma which threatens the viability of the leg and who require reconstruction of the superficial femoral or popliteal artery
2. Preoperative ultrasound or angiography or CT angiography or clinical examination indicates damage to the superficial femoral artery (SFA) or popliteal artery requiring reconstruction with an interposition bypass vessel AND required bypass length of ≤ 38cm
3. Proximal anastomosis is expected to be to the common femoral artery below the inguinal ligament or to the superficial femoral artery or popliteal artery
4. Distal anastomosis is expected to be to the SFA, popliteal artery or the tibio-peroneal trunk
5. Autologous vein graft is either not feasible in the judgment of the treating surgeon (e.g. because of lack of availability of suitable conduit, presence of severe venous insufficiency) or is not desirable because of the urgency of revascularization
6. Aged 18 to 85 years old, inclusive
7. Able to communicate meaningfully with investigative staff, and able to comply with entire study procedures. If the patient is unconscious then information from a reliable witness indicates that the patient would normally be able to comply with study procedures
8. Patient or relative is able, willing and competent to give informed consent
9. Life expectancy of at least 1 year

5.1.2 Patient Exclusion Criteria

1. Limb at high risk of amputation despite vascular reconstruction e.g., because of crush injury
2. Life threatening head, chest, or abdominal injuries that make survival unlikely
3. Distal anastomosis planned to a tibial or pedal artery
4. Known pregnant women
5. Known medical condition which would preclude long term dual antiplatelet therapy after resolution of acute injuries

6. Any other condition which in the judgment of the investigator would preclude adequate evaluation of the safety and efficacy of the HAV
7. Previous enrollment in this study
8. Employees of the sponsor or patients who are employees or relatives of the investigator

6. INVESTIGATIONAL MEDICINAL PRODUCT

Additional information on the manufacturing process and testing of the IMP is provided in the Investigator Brochure.

6.1 Product Description

The investigational medicinal product (IMP) is a Humacyte Human Acellular Vessel (HAV), which is a tissue-engineered vascular prosthesis for arterial bypass or reconstruction in patients with peripheral arterial disease or peripheral arterial trauma. It is a sterile, non-pyrogenic acellular tubular vessel composed of human collagen types I and III and other extracellular matrix proteins, including fibronectin and vitronectin. The vessel is 6 mm in diameter and approximately 42 cm in length. The product is supplied on a silicone mandrel immersed in sterile phosphate buffered saline in a sealed and labeled plastic container.

There is no placebo or comparator control group in this study.

6.2 Manufacturer of the IMP

The HAV is manufactured by:

AlloSource
6278 S. Troy Circle
Centennial, CO 80111 USA

Traceability of the HAV during and after the clinical investigation will be achieved by the assignment of lot numbers. A unique identifying lot number will be assigned to each vessel.

6.3 Packaging, Storage, and Labeling

Packaging: Each HAV is contained in a sealed, flexible plastic primary container closure system that was developed by Humacyte. The system meets container/closure requirements to maintain sterility as well as product and fluid integrity. The vessel is contained inside the system in a fixed manner, immersed in a sterile, phosphate buffered saline. The total volume of the storage solution is approximately 300 mL.

Storage: The product is shipped under controlled conditions to maintain temperature at 4°C (range: 2 – 10°C). The product should be stored in a refrigerator that maintains this temperature range. The HAV MUST NOT be allowed to freeze.

Labeling: The IMP will be labeled according to applicable guidelines and relevant regulatory agency requirements. A tamper resistant label affixed to the secondary container will be used to ensure that the product is not compromised prior to use.

6.4 Implantation of the Humacyte Human Acellular Vessel (HAV)

The Humacyte HAV is implanted using standard vascular surgical techniques similar to placement of predicate peripheral vascular prostheses (see study manual for details).

Tunneling of the HAV, if required, must be performed using a sheathed tunneler. After inserting the assembled tunneler into the tissue, the inner mandrel of the tunneler should be removed from the sheath. The sheath is lubricated with saline and then with the silicone mandrel in place, the HAV can be easily pushed through the sheath without the need to tie to the inner mandrel and pulled through the tunneler (see study manual for details).

After placement, HAV patency and integrity are checked by pressurizing the conduit. Prior to completion of surgery, angiography is performed to confirm adequacy of the bypass anastomoses, HAV patency and peripheral runoff. The surgical site is closed using standard techniques.

Implantation of the HAV will be undertaken by qualified vascular surgeons experienced in peripheral vascular surgery.

6.5 IMP Accountability Procedures

Documentation of receipt, dispensing, and return of all IMP must be maintained by the Principal Investigator or his/her designee. It is the Principal Investigator's responsibility to ensure that all IMPs are kept in a secure location, with access limited to individuals authorized by the Investigator. The product will be shipped with the IMP Shipment Confirmation Form. Once signed, the form should be returned to Humacyte or authorized designee, and the original will be maintained in the Investigator's Files. The IMP Accountability Log will be used to account for all IMP received, dispensed, and returned and must be maintained by the site until the

conclusion of the study. Following accountability of the IMP by Humacyte or their authorized designee, all unused IMP will be returned to Humacyte.

6.6 Assessment of Patient Compliance with IMP

Not applicable.

6.7 Prior and Concomitant Medications

Prior medications are defined as all prescription and over the counter (OTC) medications taken within 7 days (whether continuing or not) prior to Day 1. All prior and concomitant medications (including immediately pre-surgery and post-surgery medications) must be listed in the patient's medical record and recorded on the eCRF. Drugs used during anesthesia should be recorded in the anesthesia records but should not be transcribed into the eCRF. Patients should be questioned at each study visit concerning any new medications or changes in current medications. **Note: particular attention should be made to identify the use of antithrombotic or antiplatelet agents (e.g., aspirin, clopidogrel, prasugrel, direct thrombin inhibitors, factor Xa inhibitors, or vitamin K antagonists).**

For each medication taken, the following information will be collected:

- Medication generic name / components of combination product
- Dose
- Route of administration
- Frequency of administration
- Date started
- Date stopped
- Indication for use

6.8 Essential, Precautionary and Prohibited Medications

6.8.1 Essential Medications

All patients should receive both antibiotic and antithrombotic prophylaxis in conjunction with HAV implantation:

Antibiotic prophylaxis:

- All patients must have at least 1 day of antibiotic prophylaxis the same day as surgery in accordance with local hospital guidelines. Longer antibiotic prophylaxis is at the discretion of the investigator.

Antithrombotic prophylaxis:

- Intraoperative heparin: The doses of heparin to be used during surgery will be determined by the investigator.
- Further measures to prevent venous thromboembolism are at the discretion of the investigator and may include LMWH.
- If antiplatelet therapy was not ongoing at the time of surgery it should be commenced as soon as medically appropriate post operatively. Antiplatelet therapy (ideally dual therapy with aspirin 81-325 mg and clopidogrel 75 mg daily) should continue long term while the HAV is in place. If the patient is unable to tolerate aspiring and/or clopidogrel the choice of antiplatelet regimen is at the investigator's discretion.

6.8.2 Restricted Medications

Vitamin K antagonists, antiplatelet agents other than aspirin and clopidogrel, direct thrombin inhibitors and factor Xa inhibitors (e.g., dabigatran, apixaban and rivaroxaban) should be avoided unless essential for treatment of a medical condition arising postoperatively. In that case consideration should be given to modification or cessation of antiplatelet therapy. Antiplatelet therapy should be restarted on cessation of these anticoagulant drugs.

7. STUDY PROCEDURES / EVALUATIONS

7.1 Clinical Evaluations

- Medical History – pre-operatively, from patient / legal representative interview and medical records covering relevant past medical history.
- Smoking history
- Medication History – prescription and OTC medication from Day -7 onwards (see Section 6.7). Particular attention should be paid to the identification of OTC medications containing aspirin.
- Physical Exam – full exam (as far as possible) at pre-operative screening, 12 month visit or final study visit for early termination (ET). Clinical examination of the operative limb and HAV at all post-operative visits; physical exam for lymphadenopathy; additional clinical exam as needed to evaluate adverse events
- Vital signs (heart rate, blood pressure and temperature) – at D5
- Blood samples for hematology, clinical chemistry at pre-operative screening and Day 5 and PRA at pre-operative screening and M6
- Pre-operative imaging (ultrasound or angiography) is at the discretion of the investigator based on the clinical condition of the patient. It may be omitted if not deemed necessary in order to proceed to vascular surgical repair
- Adverse events – at all post-operative visits, the patient will be asked a general question about his/her health and for any HAV problems since the previous visit
- Intraoperative angiography or ultrasound to assess anastomotic anatomy, patency and runoff.
- Duplex ultrasound – clinical assessment at all postoperative visits from day 29 thru M12, to assess HAV patency, mid HAV diameter and flow rate. The full length of the HAV should be imaged at each assessment to monitor for aneurysm development
- Documentation of surgical procedure and any complications immediately postoperatively
- Capture of data on patient status, HAV patency and any intervention or complications at routine “standard of care” 6 monthly clinic visits from 18 to 36 months post implantation or by telephone contact with the patient or the patient’s physician

7.2 Laboratory Evaluations

7.2.1 Clinical and Research Laboratory Evaluations and Specimen Collection

The following parameters will be measured wherever possible at pre-operative screening and all should be measured at Day 5

- Hematology: hemoglobin, hematocrit, RBC, white blood cells (WBC) with differential, platelet count
- Clinical chemistry: sodium, potassium, calcium, blood urea nitrogen, creatinine, albumin, total bilirubin, glucose (non-fasting)

PRA will be measured at pre-operative screening and Month 6.

7.2.2 Specimen Preparation, Handling and Shipping

Biochemistry (no additives) and Hematology (EDTA) Tubes – collect and transport to the appropriate local clinical laboratory according to institutional procedures. To avoid cross-contamination of additives between tubes, the order of draw is as follows:

FIRST: Draw two biochemistry tubes (no additives) – the first is for clinical chemistry, the second is for PRA [See below for processing of this sample.]

SECOND: Draw one hematology tube - (EDTA tube) for hematology studies

Second Non-Additive Red Top Tube for PRA: Allow sufficient time for the blood to clot and then centrifuge the tube to separate the serum from clotted material. Transfer at least 0.5 ml of serum into a pre-labeled screw cap vial (provided by sponsor) for PRA testing. Transfer this vial to a secure, monitored freezer (≤ -20°C). PRA samples will be shipped to Humacyte, Inc. in batches.

7.2.3 Monitoring, Maintenance and Calibration of Equipment

All laboratory tests (except assay of PRA) will be conducted at certified hospital laboratories. Routine monitoring, maintenance or calibration of laboratory equipment is required per local site procedures. Samples for PRA analysis will be shipped to Humacyte for analysis at a central laboratory.

7.3 Imaging Evaluations

7.3.1 CT Angiography and Conventional Angiography

CT angiography or conventional angiography will be conducted as pre-operative screening when feasible. Pre-operative imaging is at the discretion of the investigator based on the clinical condition of the patient. It may be omitted if not deemed necessary in order to proceed to vascular surgical repair. Determination of intraoperative HAV patency on Day 1 can be done by conventional angiography or ultrasound at the discretion of the investigator.

7.3.2 Duplex Ultrasound

Clinical duplex ultrasound examinations will be performed at Day 29, 3, 6, 9 and 12 months and follow standard bypass graft imaging protocols, including B-mode, power Doppler and color duplex ultrasound imaging of the HAV with velocity spectral waveform analysis. The purpose of this clinical duplex ultrasound surveillance is to detect HAV stenosis and aneurysm development. Determination of intraoperative HAV patency on Day 1 can be done by conventional angiography or ultrasound at the discretion of the investigator.

7.4 Study Schedule

7.4.1 Pre-operative Screening (Day 1)

Potential study participants who are being considered for surgical repair of the superficial femoral or popliteal artery will be informed about the study and invited to participate. After explanation of the potential risks and benefits of the HAV and of the study procedures, written informed consent will be obtained. No study specific procedures may be performed prior to patient consent. If the patient is unable to give informed consent then this may be sought from the patient's legal representative, usually a close relative. Standard of care procedures such as laboratory evaluations conducted prior to screening may be used rather than repeating the test.

The following assessments will be performed, as far as possible, prior to surgery (Day 1):

- Informed consent
- Medical history
- Prior and concomitant medication
- Full physical examination
- Evaluation of inclusion/exclusion criteria

- Reasons for not using an autologous venous conduit
- Laboratory testing (or standard pre-op lab profile for the institution)
  - Hematology: full blood count and differential
  - Clinical chemistry ; sodium, potassium, calcium, blood urea nitrogen, creatinine, albumin, total bilirubin, glucose (non-fasting)
  - PRA
- Ultrasound or CT angiography (CTA) (pre-op imaging is at the discretion of the investigator based on the clinical condition of the patient. It may be omitted if not deemed necessary in order to proceed to vascular surgical repair

7.4.2 Enrollment – Day 1 (HAV Implantation)

The HAV will be implanted as an interposition replacement or bypass vessel in the superficial femoral or popliteal artery using standard vascular surgical techniques. Details of the surgical anatomy and any complications will be documented. Determination of intraoperative HAV patency can be done by conventional angiography or ultrasound at the discretion of the investigator.

7.4.3 Follow-up Visits

Day 5 (or prior to hospital discharge if earlier)

- Concomitant Medication
- Physical exam including surgical site (and HAV patency) and to evaluate any AEs
- Vital signs (heart rate, blood pressure and temperature)
- Documentation of any HAV interventions
- Adverse events
- Laboratory assessments (clinical chemistry, hematology)

Day 29 (+/- 4 days)

- Concomitant medication
- Physical exam including surgical site (and HAV patency), and to evaluate any AEs
- Clinical duplex ultrasound of the HAV
- Documentation of any HAV interventions
- Adverse events

Months 3, 6 and 9 (+/- 14 days)

- Concomitant Medication
- Physical exam including surgical site (and HAV patency) and to evaluate any AEs
- Clinical duplex ultrasound of the HAV
- Documentation of any HAV interventions
- Adverse events
- PRA (Month 6 only)

Month 12 (+/-14 days) and Early Termination

- Concomitant Medication
- Documentation of HAV interventions
- Adverse events
- Clinical duplex ultrasound of the HAV
- Full physical exam including surgical site (and HAV patency) and to evaluate any AEs

7.4.4 Final Study Visit

The final study visit is at 12 months.

7.4.5 Early Termination Visit

The patient may withdraw from the study at any time at his/her own or his/her physician's discretion. If withdrawal occurs before 12 months the patient will be asked to complete an early termination visit at which all assessments normally performed at 12 months will be completed. PRA will be collected at ET visit if the visit occurs before Month 6 collection of the sample.

The reasons for early termination should be recorded in the eCRF.

The patient should be withdrawn from the study prior to 12 months if the HAV is removed or becomes permanently occluded (loss of secondary patency).

7.4.6 Unscheduled Visits

If necessary to evaluate adverse events or HAV complications additional visits may be scheduled at the discretion of the investigator. At a minimum HAV status on clinical examination and Duplex ultrasound and adverse events will be recorded.

If, at any of the scheduled visits, duplex ultrasound surveillance suggests the development of a ≥ 50% stenosis within the HAV the patient should be asked to return for an additional visit 6 weeks later for a repeat duplex ultrasound study. Intervention to manage any such stenosis is at the discretion of the investigator taking into account the degree and rate of progression of the stenosis.

7.4.7 Follow Up from Months 18-36 (+/- 30 days)

The final study visit is at Month 12. Thereafter the status of the patient and HAV will be ascertained every 6 months until 3 years after HAV implantation at routine 6 month interval clinical visits or by telephone contact with the patient and/or his physician. The investigator will complete a brief questionnaire covering the status of the patient, known patency of the HAV and any complication, interventions or other vascular procedures on the operative leg. This information will be entered onto a form, which will be returned to the CRO / sponsor after each follow up visit or call. These data will not be verified against source documents, and the investigational sites will not undergo additional monitoring now that the main study is complete.

The patient should be withdrawn from the study if the HAV is removed or becomes permanently occluded (loss of secondary patency).

There are no study specific visits or procedures after 12 months.

7.5 Medical Care during the Study and upon Study Termination

Optimal medical therapy should be continued during the study. This should include:

- Dual antiplatelet therapy (see section 6.8.1)

After the final study visit at month 12 patients will not receive any further study-specific treatment. They will be treated by their medical doctor in a way that is appropriate for them.

7.6 Histological Examination of Resected HAV Material

If all or part of the HAV is resected it should, wherever possible, be retained for future histological examination. Instructions for preservation, storage and shipping of this material will be provided separately in a procedures manual. If a patient dies with an HAV in situ and it is feasible to obtain a fresh post mortem sample of the bypass this should be attempted in accordance with local regulations.

8. ASSESSMENT OF SAFETY

Safety of the HAV will be assessed in terms of:

- Aneurysm formation
- Anastomotic bleeding or rupture
- HAV infection
- Need for HAV removal
- Irritation/Inflammation at the implantation site
- Other adverse events
- Laboratory parameters (clinical chemistry, hematology)

8.1 Adverse Event Definition

An AE is any untoward medical occurrence in a patient administered an IMP and which does not necessarily have a causal relationship with the IMP. An AE can, therefore, be any unfavorable and unintended sign, symptom, or disease temporally associated with the use of an IMP, whether or not related to the IMP. Any worsening of the patient's disease under study or other medical conditions will also be considered to be an AE, unless it is within the normal range of disease fluctuation for that patient.

8.2 Reporting of Adverse Events

At each evaluation, the investigator will determine whether any AEs have occurred. The patient will be questioned in a general way and no specific symptoms will be suggested. If any AEs have occurred they should be documented in the patient's medical chart and recorded on the AE pages of the eCRF. If known, the diagnosis should be recorded in preference to the listing of individual signs and symptoms. All SAEs (see Section 9), whether or not considered to be related to study treatment, should be reported to the study pharmacovigilance officer within 24 hours of identification.

AE reporting begins from time of anesthesia induction for implantation of the HAV and ends at the conclusion of the post-treatment follow-up period (i.e., Month 12 visit) unless an unresolved AE is still being followed (covered in further detail in Section 8.3).

Adverse events will be graded as follows:

- Grade 1 (Mild): Events require minimal or no treatment and do not interfere with the patient's daily activities.

- Grade 2 (Moderate): Events result in a low level of inconvenience or concern with the therapeutic measures. Moderate events may cause some interference with functioning.

- Grade 3 (Severe): Events interrupt a patient's usual daily activity and may require systemic drug therapy or other treatment. Severe events are usually incapacitating.

- Grade 4 (Life threatening): Events posing an immediate risk to the patient's life and requiring urgent intervention

- Grade 5 (Fatal): Events resulting in death

Relationship assessment of AEs/SAEs to the IMP and to the procedure should be made by the investigator. (NOTE: Relationship assessment is not a factor in determining what is or is not reported in the study.)

- Definitely Related: There is clear evidence to suggest a causal relationship, and other possible contributing factors can be ruled out. The clinical event, including an abnormal laboratory test result, occurs in a plausible time relationship to placement of the IMP and cannot be explained by concurrent disease or other devices, drugs, or chemicals.

- Possibly Related: There is some evidence to suggest a causal relationship (e.g., the event occurred within a reasonable time after the placement of the IMP). However, the influence of other factors may have contributed to the event (e.g., the patient's clinical condition, other concomitant events). Although an adverse event may rate only as "possible" soon after discovery, it can be flagged as requiring more information and later be upgraded to definitely as appropriate.

- Unlikely Related: A clinical event, including an abnormal laboratory test results, whose temporal relationship makes a causal relationship improbable (e.g., the event did not occur within a reasonable time after placement of the IMP) and in which other drugs or chemicals or underlying disease provide plausible explanations (e.g., the subject's clinical condition, other concomitant treatments).

- Not Related: The AE is, in the opinion of the investigator, independent of the placement of the investigational product, and/or evidence exists that the event is definitely related to another etiology. There must be an alternative, definitive etiology documented by the clinician.

If sponsor and reporting principal investigator disagree on the seriousness of an event or on its relationship to the IMP, the sponsor will make the final determination of causality that will be communicated to the relevant authorities and involved principal investigators.

8.3 Reporting of Pregnancy

If a study participant becomes pregnant during study participation, basic information about the pregnancy will be recorded in the Pregnancy eCRF and the Pregnancy Outcome and Report Form, and submitted to the Safety CRO. If there are complications during the pregnancy, the complications are recorded as AEs. The participant will be asked to report the outcome of the pregnancy and the site should submit the information to the Safety CRO within 30 days after the outcome of the pregnancy. If there is a congenital anomaly in the infant, this will be recorded as a SAE in the data forms for the mother (i.e., the study participant).

Partner pregnancies do not need to be reported.

8.4 Follow-Up of Adverse Events

If any SAEs are present when a patient completes the study or is withdrawn from the study, the patient will be re-evaluated within an appropriate period of time. If the SAE has still not resolved, additional follow-up will be performed as appropriate. Every effort should be made by the investigator or delegate to contact the patient until the SAE has resolved or stabilized or the medical monitor and investigator agree that further follow-up is not necessary. This should be documented in the patient's medical records.

9. SERIOUS ADVERSE EVENTS

A SAE is any untoward medical occurrence that at any dose:

- Results in death
- Is life-threatening
- Requires patient hospitalization or prolongation of existing hospitalization
- Results in persistent or significant disability/incapacity
- Is a congenital anomaly/birth defect

Medical and scientific judgment should be exercised in deciding whether expedited reporting to the sponsor is appropriate in other situations, such as important medical events that may not be immediately life-threatening or result in death or hospitalization but may jeopardize the patient or may require intervention to prevent one of the other outcomes listed in the definition above. These should also be considered to be SAEs.

9.1 Life-Threatening Adverse Event

The term “life-threatening” in the definition of “serious” refers to an event in which the patient was at risk of death at the time of the event; it does not refer to an event, which, hypothetically, might have caused death if it were more severe.

9.2 Hospitalization

This is defined as the patient being hospitalized overnight or the patient’s hospital stay being prolonged for at least an additional overnight stay, excluding pre-planned or elective procedures.

9.3 Unexpected Adverse Reaction

An unexpected adverse reaction is defined as an adverse reaction to the IMP, the nature or severity of which is not consistent with Investigator’s Brochure for the IMP.

9.4 Reporting Serious Adverse Events

The urgency for reporting SAEs is 4-fold: (1) to facilitate discussion (and implementation, if necessary) by the sponsor and the Investigator of appropriate follow-up measures, (2) to

facilitate Investigator reporting of unanticipated problems involving risk to human subjects to the institutional review board (IRB), (3) to facilitate the sponsor's rapid dissemination of information regarding AEs to other Investigators/sites in a multi-center study, and (4) to enable the sponsor to fulfill the reporting requirements to the appropriate regulatory authority.

Any SAE that occurs during the course of this study from the time of HAV implantation until the 12 month follow up, whether or not causally related to the IMP, must be reported by the investigator or designee to the Safety CRO within 24 hours of learning of its occurrence. This applies also to any AE that could affect the safety of the study participants or the conduct of the study. SAEs which involve the HAV (including thrombosis, aneurysm formation or infection) which are reported to the investigator during the long term follow up period should also be reported to the Safety CRO within 24 hours.

Information about an SAE will be collected and recorded on the SAE Report Form. The investigator must assess the relationship to the investigational product and any relevant procedure.

The investigators complete the SAE Report Form in English, and **send the completed, signed form by fax or email (see below) IMMEDIATELY (at latest within 24 hours) after becoming aware of the SAE.**

Copies of relevant medical records (e.g., admission and/or discharge summary, laboratory reports and autopsy report), may also be submitted with the SAE form to clarify the circumstances surrounding the SAE(s). The entire medical records should **NOT** be sent with the SAE form.

Atlantic Research Group, Inc

Drug Safety Department

Fax: +1-866-851-9318

Email: Safety@atlanticresearchgroup.com

Phone: +1-888-619-3216

The investigator will be requested to supply as much detailed information as possible regarding the SAE that is available at the time of the initial contact. The investigator should also complete missing or requested information and submit follow-up reports on a new SAE Form until the SAE has resolved or, in the case of permanent impairment, until the SAE has stabilized.

It is the responsibility of each Investigator to promptly notify his/her IRB of all SAEs that are received by the Sponsor or designee and that occur at his/her institution in accordance with institutional practices.

The Safety CRO will inform the sponsor about all SAEs within 1 business day after receipt of the respective report from the investigator.

9.5 Medically Significant Events

Any adverse event indicating a clinically significant impairment of HAV function or involving an intervention on the HAV, **even if it does not meet any criteria for an SAE**, should be reported to the pharmacovigilance officer as described in section 9.4 above.

9.6 Data Safety Monitoring Board

A Data Safety Monitoring Board (DSMB) will be convened by the Sponsor to review safety data on an ongoing basis for this study. The DSMB will consist of individuals who are not directly involved in the conduct of the study. A separate charter will be established that will describe the roles and responsibilities of the DSMB.

9.7 Interim Analysis and Stopping Criteria

This is a phase 2 study with no formal interim analysis. Periodic reviews of safety data will be undertaken by the sponsor (and DSMB) with particular attention to events that might indicate structural failure of the HAV. Events that might have implications for already implanted HAVs and their possible removal - such as aneurysms or rupture -would trigger an urgent review of the safety data and in the interim no new patients would be implanted. Any serious deep space infection necessitating surgical resection of the HAV would also trigger a suspension of recruitment pending review.

10. STATISTICAL CONSIDERATIONS

This is a prospective, open label, single treatment arm, multicenter pilot study to evaluate the safety and efficacy of the HAV in patients undergoing peripheral vascular repair. The primary objective of this study is to evaluate the safety and tolerability of the HAV in these patients and to determine the patency of the Humacyte HAV and the rate of limb salvage at 12 months post-implantation. The secondary objectives of this study are to further assess safety in terms of adverse events, and laboratory parameters and to determine the rates of bypass interventions required to keep the HAV patent. There is no formal hypothesis testing planned; the study involves only a single, open-label treatment group.

Details of data handling and planned descriptive statistics are given in the Statistical Analysis Plan.

10.1 Analysis Population

All patients who receive an HAV will be included in the analyses. For discontinued or withdrawn patients, all available data will be included in the safety and efficacy analyses.

10.2 Safety Analyses

Safety analyses will be performed on all patients who have an HAV implanted.

The incidence of aneurysm formation, anastomotic bleeding or rupture, HAV removal, HAV infection, and irritation/inflammation at the implantation site will be tabulated by visit and overall.

Adverse events will be coded using Medical Dictionary for Regulatory Activities (MedDRA) terms. Adverse events will be listed and summarized by body system, incidence, severity, and duration. HAV complications will be listed in terms of incidence, severity, and (where appropriate) time to onset and duration. Serious adverse events will be summarized separately. Any premature discontinuations due to adverse events and deaths will be listed and summarized.

Laboratory data, including PRA, will be listed and summarized using appropriate descriptive statistics for the absolute change from baseline values for all post-surgery visits. The closest non-missing values prior to surgery on Day 1 will be used as baseline values.

10.3 Efficacy Analyses

Primary, primary assisted, and secondary patency rates of the HAV at 12 months and at all other post-surgery visits with evaluation of patency will be described. The rate of limb salvage at 12 months will also be described.

Primary patency is defined as the functional access patency until any type of intervention; primary assisted patency is defined as an HAV still working without thrombosis; secondary patency is defined as the functional HAV patency, with or without preceding successful interventional or surgical procedures to maintain or reestablish patency, until the HAV is abandoned. Early discontinued patients prior to the visit of interest will be determined as being non-patent irrespective of the reason for discontinuation.

The rate and type of interventions needed to maintain / restore patency in the HAV will be descriptively tabulated.

The absolute change from baseline (Day 1) values to all post-surgery visits of duplex ultrasound parameters will be summarized. Summary statistics will also be provided at each time point.

The methods and endpoints regarding the efficacy parameters employed in this study are consistent with current clinical practice and are meaningful to the research community. Every attempt has been made to minimize the variability on the part of the surgeon when using this product. The results of this study will be used to determine the sample size of subsequent clinical studies.

10.4 Other Analyses

All clinical parameters will be listed for all patients treated at each study visit. Descriptive statistics will be summarized for continuous outcomes such as age and BMI. If necessary, number and percentage of patients will be reported for categorical outcomes.

10.5 Sample Size Rationale

Up to 20 patients will be recruited into the study. As this phase II study is the first study of the HAV in humans for vascular trauma, the number of patients was chosen in order to provide sufficient safety information on this bioengineered vessel to allow the initiation of further trials in larger numbers of patients.

The study is not powered to assess the efficacy of the HAV.

10.6 Interim analyses

There is no formal interim analysis.

11. STUDY MANAGEMENT AND DATA COLLECTION

11.1 Ethical Conduct of the Trial

This study will be conducted according to the protocol; 21 CFR Parts 11, 50, 54, 56, and 312; the World Medical Association Declaration of Helsinki and Good Clinical Practice (GCP). Each Investigator will conduct the trial according to applicable local or regional regulatory requirements.

11.2 Institutional Review Board

IRBs must be constituted according to the applicable state and federal requirements, including ICH GCP.

It is the responsibility of each investigator to submit the protocol, Investigator's Brochure, subject informed consent, subject recruitment materials (if applicable), and other documentation as required by the IRB to his/her IRB for review and approval. A copy of the written approval must be provided to the contract research organization (CRO). The documentation should clearly mention the approval/favorable opinion of the protocol, the subject informed consent form, and subject recruitment materials (if applicable), including respective version dates. The written approval and a list of members, their titles or occupations, and their institutional affiliations may be obtained from the IRBs if available, and provided to the CRO prior to the release of clinical study supplies to the investigational site and commencement of the study. If any member of the IRB has direct participation in this trial, written notification regarding his or her abstinence from voting must also be obtained.

Each investigator must adhere to all requirements stipulated by his/her respective IRB. This includes notification to the IRB regarding protocol amendments, updates to the subject informed consent, recruitment materials intended for viewing by subjects, investigational new drug safety reports, SAEs and unexpected AEs, reports and updates regarding the ongoing review of the trial at intervals specified by the respective IRB, and submission of final study reports and summaries to the IRB.

11.3 Subject Informed Consent

Prior to any study procedures being performed, subjects and persons conducting the consent discussion will be required to sign and date the IRB-approved informed consent, and each subject will be given a copy. In addition, this information should be recorded in the subject's medical record (i.e., source document). If the patient is unable to give informed consent then this may be sought from the patient's legal representative, usually a close relative.

The written consent document will embody the elements of informed consent as described in the World Medical Association Declaration of Helsinki, 21 CFR Part 50.25, ICH E6 guideline

(GCP), and in accordance with any local regulations. The investigator is responsible for the preparation, content, and IRB approval of the informed consent document. The consent form must be approved by the site's IRB and be acceptable to Humacyte.

The consent form must be written in a language fully comprehensible to the prospective subject. The investigator or designee shall give the subject adequate opportunity to read it before it is signed and dated. Information should be given in both oral and written form whenever possible and in the manner deemed appropriate by the IRB. Subjects must be given ample opportunity to inquire about details of the study.

11.4 Amendments to the Protocol

An amendment must be agreed to in writing by Humacyte and submitted to and approved by the respective Regulatory Authority and IRB before the amendment can be implemented. Written approval of a protocol amendment is not required prior to implementation of changes to the protocol which eliminate an immediate hazard to the study patient; however, approval must be obtained as soon as possible thereafter. Any agreed amendments must also be signed by the investigator.

11.5 Study Initiation

The investigator must not enroll any patients prior to attendance at the Investigator Meeting or the completion of a formal site initiation visit conducted by the CRO. These meetings will include a detailed review of the study protocol and eCRF pages. The investigator will not be supplied with IMP until all necessary pre-study requirements have been completed and essential signed documents provided to the CRO.

11.6 Study Monitoring

It is the responsibility of the investigator to ensure that the study is conducted in accordance with the protocol, GCP, applicable regulatory requirements, and the currently approved Declaration of Helsinki, and that valid data are entered into the eCRF.

To achieve this objective, the monitor's duties are to ensure the maintenance of complete, legible, well-organized, and easily retrievable data. The monitor will review the protocol with the investigator. In addition, the monitor will explain the investigator's reporting responsibilities and all applicable regulations concerning the clinical evaluation of the IMP.

The investigator will permit representatives of Humacyte and the CRO to monitor the study as frequently as Humacyte or the CRO deem necessary to determine that data recording and protocol adherence are satisfactory. The eCRF data and related source documents will be reviewed in detail by the monitor at each visit, in accordance with relevant SOPs and ICH GCP

regulations. This includes results of tests performed as a requirement for participation in this study and any other medical records required to confirm information contained in the eCRF such as past medical history and secondary diagnoses. The investigator and his/her staff will be expected to cooperate with the monitor and provide any missing information whenever possible.

All monitoring activities will be reported and archived. In addition, monitoring visits will be documented at the investigational site by signature and date on the study-specific monitoring log.

11.7 Case Report Form

An electronic CRF will be used for this study. The data will be entered into the eCRF in a timely manner on an ongoing basis.

The investigator is responsible for ensuring that data are properly recorded on each patient's eCRF and related documents. An investigator who has signed the protocol signature page should personally sign the eCRFs in accordance with the procedure described in the eCRF completion guidelines to ensure that the observations and findings are correct and complete.

For data handled by the CRO, eCRF data and some or all of the study-related data will be managed and stored electronically in the CRO's database system. Validated data will subsequently be transferred to the sponsor.

11.8 Verification Procedures

It is the investigator's obligation to ensure documentation of all relevant data in the subject's medical record. The subject's medical record will be considered the source document. The eCRF should not be used as the source for study information.

The investigator will maintain a subject identification code list to enable unambiguous identification of the subjects (subject names and corresponding subject numbers). The subject identification code list is an essential document and as such should be maintained according to the ICH GCP guidelines.

11.9 Retention of Records

All documentation pertaining to the study will be kept by Humacyte or their designee in accordance with ICH guidelines and US FDA regulations.

The investigator will maintain a study file, which should be used to file the Investigator's Brochure, protocol, and IMP records; correspondence with the IRB and Humacyte; and other study-related documents.

The investigator agrees to keep records and those documents that include (but are not limited to) the identification of all participating subjects, medical records, study-specific source documents, source worksheets, all original signed and dated informed consent forms, query responses, and detailed records of drug disposition to enable evaluations or audits from regulatory authorities and Humacyte or its designees.

The investigator shall retain records required to be maintained under this part for a period of 2 years following the date a marketing application is approved for the IMP for the indication for which it is being investigated; or, if no application is to be filed or if the application is not approved for such indication, until 5 years after the investigation is discontinued. However, these documents should be retained for a longer period if required by the applicable regulatory requirement(s) or if needed by the sponsor. In addition, the investigator must make provision for the subject's medical records to be kept for the same period of time. No data should be destroyed without the agreement of Humacyte. Humacyte will inform the investigator in writing when the trial-related records are no longer needed. Subject's medical records and other original data will be archived in accordance with the archiving regulations or facilities of the study site.

11.10 Protocol Deviations

A protocol deviation is any noncompliance with the protocol or GCP requirements. The noncompliance may be either on the part of the participant, the investigator, or the study site staff. As a result of deviations, corrective actions are to be developed by the site and implemented promptly. Although in principle, protocol deviations are not permitted, under emergency circumstances, deviations may proceed without prior approval of the sponsor and the IRB to protect the rights, safety, and well-being of human subjects.

All protocol deviations will be documented and reported by the CRO during the course of the study in the Monitoring Reports. All deviations will be reported to the sponsor who will agree on the necessary actions to be taken.

If required per their guidelines, reports about protocol deviations must be reported to the local IRB.

11.11 Insurance and Indemnity

Insurance coverage for damages emerging from the study will be provided according to applicable legal requirements. During the informed consent procedure, the investigator must inform the patient accordingly.

11.12 Audit

It is the responsibility of CRO and Humacyte to perform auditing (if applicable) as part of implementing quality assurance. The purpose of an audit, which is independent of and separate from routine monitoring or quality control functions, is to evaluate trial conduct and compliance with the protocol, SOPs, GCPs, and the applicable regulatory requirements. The auditor and regulatory authorities will require authority from the investigator to have direct access to the subject's medical records.

12. REPORTING

Following completion of follow-up of all patients to the 12-month endpoint, the results will be evaluated by Humacyte or a designee for clinically meaningful findings. A clinical study report will be generated, including a summary of all available data, statistical measures, tabulated results, graphical results and interpretations. This report will be submitted to regulatory authorities in a timely manner. An addendum to the report will be generated to include data up to 36 months follow-up. This addendum will be submitted to regulatory authorities in a timely manner.

13. QUALITY CONTROL AND QUALITY ASSURANCE

Following written standard operating procedures, the monitors will verify that the clinical trial is conducted and data are generated, documented (recorded), and reported in compliance with the protocol, GCP, and the applicable regulatory requirements. Reports of monitoring activities will be submitted to Humacyte in a timely manner.

The investigational site will provide direct access to all trial related areas, source data/documents, and reports for the purpose of monitoring and auditing by the sponsor, and inspection by local and regulatory authorities.

Quality control procedures will be implemented for data entry and the generation of data quality control checks and will be run on the database. Any missing data or data anomalies will be communicated to the site(s) for clarification/resolution.

14. RESPONSIBILITIES OF THE PRINCIPAL INVESTIGATOR

The role of the principal investigator is to implement and manage the day-to-day conduct of the clinical investigation as well as to ensure data integrity and the rights, safety, and well-being of the patients involved in the clinical investigation.

14.1 Informed Consent

The principal investigator shall ensure that the process for obtaining informed consent

- includes all aspects of the clinical investigation that are relevant to the patient's decision to participate throughout the clinical investigation,
- avoids any coercion or undue improper influence on, or inducement of, the patient to participate,
- does not waive or appear to waive the patient's legal rights,
- uses native non-technical language that is understandable to the patient,
- provides ample time for the patient to read and understand the informed consent form and to consider participation in the clinical investigation,
- provides the patient with a copy of the signed and dated informed consent form and any other written information.

The principal investigator shall ensure and document appropriate training if an authorized designee is appointed to conduct the informed consent process.

14.2 Compliance with the Protocol

The principal investigator shall:

- indicate his/her acceptance of the protocol in writing
- conduct the clinical investigation in compliance with the protocol
- create and maintain source documents throughout the clinical investigation and make them available as requested during monitoring visits or audits
- ensure that the IMP is used solely by authorized users, and in accordance with the protocol and instructions for use

- propose to the sponsor any appropriate modification(s) of the protocol
- refrain from implementing any modifications to the protocol without agreement from the sponsor, IRB, and, if required, regulatory authorities
- document and explain any deviation from the approved protocol that occurred during the course of the clinical investigation
- ensure that an adequate investigation site team and facilities exist and are maintained and documented during the clinical investigation
- ensure that maintenance and calibration of the equipment relevant for the assessment of the clinical investigation is appropriately performed and documented, where applicable
- ensure the accuracy, completeness, legibility, and timeliness of the data reported to the sponsor in the eCRFs and in all required reports
- maintain the clinical trial material accountability records
- allow and support the sponsor to perform monitoring and auditing activities
- be accessible to the monitor and respond to questions during monitoring visits
- allow and support regulatory authorities and the IRB when performing auditing activities
- ensure that all clinical-investigation-related records are retained as specified in this protocol.

14.3 Medical Care of Patients

The principal investigator shall:

- provide adequate medical care to a patient during and after a patient's participation in a clinical investigation in the case of AEs
- inform the patient of the nature and possible cause of any adverse events experienced
- inform the patient of any new significant findings occurring during the clinical investigation, including the need for additional medical care that may be required
- provide the patient with well-defined procedures for possible emergency situations related to the clinical investigation, and make the necessary arrangements for emergency treatment,

- ensure that clinical records are clearly marked to indicate that the patient is enrolled in a particular clinical investigation
- inform, with the patient's approval or when required by national regulations, the patient's personal physician about the patient's participation in the clinical investigation
- make all reasonable efforts to ascertain the reason(s) for a patient's premature withdrawal from the clinical investigation while fully respecting the patient's rights.

14.4 Safety Reporting

The principal investigator shall:

- record every adverse event together with an assessment, in accordance with Sections 8 and 9 of this protocol,
- report to the sponsor, without unjustified delay, all serious adverse events and medically significant events as specified in Sections **8** and **9** of this protocol,
- supply the sponsor, upon sponsor's request, with any additional information related to the safety reporting of a particular event.

15. SUSPENSION OR PREMATURE TERMINATION OF THE CLINICAL INVESTIGATION

The sponsor may suspend or prematurely terminate either a clinical investigation in an individual investigation site or the entire clinical investigation for significant and documented reasons.

A principal investigator, IRB, or regulatory authority may suspend or prematurely terminate participation in a clinical investigation at the investigation sites for which they are responsible.

If suspicion of an unacceptable risk to patients arises during the clinical investigation, or when so instructed by the IRB or regulatory authorities, the sponsor shall suspend the clinical investigation while the risk is assessed. The sponsor shall terminate the clinical investigation if an unacceptable risk is confirmed.

The sponsor shall consider terminating or suspending the participation of a particular investigation site or investigator in the clinical investigation if monitoring or auditing identifies serious or repeated deviations on the part of an investigator.

If suspension or premature termination occurs, the terminating party shall justify its decision in writing and promptly inform the other parties with whom they are in direct communication.

If, for any reason, the sponsor suspends or prematurely terminates the investigation at an individual investigation site, the sponsor shall inform the responsible regulatory authority if required and ensure that the IRB is notified. If the suspension or premature termination was in the interest of safety, the sponsor shall inform all other principal investigators.

If suspension or premature termination occurs,

1. the sponsor shall remain responsible for providing resources to fulfill the obligations from the protocol and existing agreements for following up the patients enrolled in the clinical investigation, and

2. the principal investigator or authorized designee shall promptly inform the enrolled patients at his/her investigation site, if appropriate.

In the event that the study is discontinued, the reasons for discontinuation will be explained to the investigators and may be disclosed to the study participants. Humacyte will provide all information needed by the investigator to ensure the safety and well-being of the study participants.

16. PUBLICATION POLICY

A Publication Committee comprising the Principal Investigator from each participating site and a representative of Humacyte will oversee all publication of data from this study. Prior to submitting for publication, presenting, using for instructional purposes, or otherwise disclosing the results of the study, the investigator agrees to allow the Publication Committee and Humacyte a period of at least 30 days (or, for abstracts, at least 5 calendar days) to review the proposed publication or disclosure prior to its submission for publication or other disclosure. Publications or disclosures of study results shall not include other confidential information belonging to Humacyte. If the proposed publication/disclosure risks Humacyte's ability to patent any invention related to the study, the publication or disclosure will be modified or delayed, at the investigator's option, a sufficient time to allow Humacyte to seek patent protection of the invention. For multicenter studies, the first publication or disclosure shall be a complete, joint multicenter publication or disclosure. This statement does not give Humacyte any editorial rights over the content of a publication or disclosure, other than to restrict the disclosure of Humacyte's confidential information. If a written contract for the conduct of the study is executed which includes publication provisions inconsistent with this statement, then that contract's publication provisions shall apply rather than this statement.

17. REFERENCES

Akingba G, Rapp BM, Tsai A, Jester AL, Fajardo A, Motaganahali R, et al. Surgical Management of Vascular Trauma From Dog Bites: The 10- year Experience of a Level I Trauma Center. Journal of Vascular Surgery. 2012;56(4):1190.

Andrikopoulos V, Antoniou I, Panoussis P. Arterial injuries associated with lower-extremity fractures. Cardiovascular Surgery. 1995;3(1):15-8.

Cordesman AH, Lemieux, J. IED Metrics for Afghanistan. January 2004-May 2010. Center for Strategic & International Studies, 2010.

Dahl, SL, Kypson, AP, Lawson, JH et al. Readily available tissue-engineered vascular grafts. Sci Transl Med. 2011: 68ra9

Dixon BS, Beck GJ, Vazquez MA et al. Effect of dipyridamole plus aspirin on hemodialysis graft patency. N Engl J Med 2009;360:2191-201.

Helfet DL, Howey T, Sanders R, Johansen K. Limb Salvage Versus Amputation: Preliminary Results of the Mangled Extremity Severity Score. Clinical Orthopaedics & Related Research. 1990;256:80-6.

Holcomb J. Devastating Dismounted IED Injuries in OEF. Increasing amputation and genital injury rates admitted to LRMC 2009-2010. 2011. Presentation to the Defence Health Board, (http://www.health.mil/Reference-Center/Presentations/2011/03/08/Devastating-Dismounted-Improvised-Explosive-Device-Injuries-in-Operation-Enduring-Freedom)

Kelly BS, Heffelfinger SC, Whiting JF, et al. Aggressive venous neointimal hyperplasia in a pig model of arteriovenous graft stenosis. Kindey Int. 2002;62(6):2272-2280.

L'Heureux N, Dusserre N, Konig G, et al. Human tissue-engineered blood vessels for adult arterial revascularization. Nat Med 2006; 12(3):361-365.

Mills JL et al on behalf of the Society for Vascular Surgery Lower Extremity Guidelines Committee. The Society for Vascular Surgery Lower Extremity Threatened Limb Classification System: Risk stratification based on Wound, Ischemia, and foot Infection (WIfI). J Vasc Surg 2013,

Morgan MBF, Crawford T, Murrin B and Fraser SCA. Developing the vascular quality of life questionnaire: a new disease specific quality of life measure for use in lower limb ischemia. J Vasc Surg 2001; 33: 679 - 687

Prichard, H.L, Manson, R.J.,Dibernado, L. et al. An early study on the mechanisms that allow tissue-engineered vascular grafts to resist vascular hyperplasia. J Cardiovasc Transl Res 2011, 4, 674-682

Quint C, Kondo Y, Manson RJ,et al. Decellularized tissue-engineering blood vessel as an arterial conduit. Proc Natl Acad Sci USA. 108(22):9214-9, 2011.

Rasmussen TE. Vascular Injury Rates in the Wars in Iraq and Afghanistan: How are they different from the past? United States Army Institute of Surgical Research, 2010.

Roy-Chaudhury P, Kelly BS, Miller MA, et al. Venous neointimal hyperplasia in polytetrafluoroethylene dialysis grafts. Kidney Int. 2001;59(6):2325-2334.

Rutherford RB1, Baker JD, Ernst C, et al Recommended standards for reports dealing with lower extremity ischemia: revised version. J Vasc Surg. 1997 Sep;26(3):517-38.

APPENDIX 1: HAV CLINICAL VISIT SCHEDULE

	Pre-op screening D1	**D 1**	**D 5** or prior to d/c	**D 29** ± 7 days	**M3** ± 14 days	**M 6** ± 14 days	**M 9**± 14 days	**M12 / ET†** ± 14 days	**M18-M36** Routine SOC Visits
Informed consent	X								
Medical history and nature of trauma	X								
Concomitant medication	X	X	X	X	X	X	X	X	
Physical exam	X	X[1]	X[1]	X[1]	X[1]	X[1]	X[1]	X	
Ultrasound or (CT) angiography[2]	X								
Vital signs			X						
Eligibility (inclusion/exclusion criteria)	X								
HAV implantation and intraoperative angiography or ultrasound to confirm patency[3]		X							
Documentation of surgery and any complications		X							
Clinical chemistry	X[4]		X						
Hematology	X[4]		X						
PRA	X					X		X[5]	
Clinical duplex ultrasound				X	X	X	X	X	X[6]
AEs		X	X	X	X	X	X	X	
Documentation of HAV interventions		X	X	X	X	X	X	X	

Abbreviations: AEs, adverse events; D, day; d/c, discharge; ET, early termination; HAV, human acellular vessel; M, month

1. Physical examination includes clinical exam of the operative limb and HAV at all post-operative visits (incl. patency assessment on D1) and physical exam to evaluate AEs
2. Pre-op imaging is at the discretion of the investigator based on the clinical condition of the patient. It may be omitted if not deemed necessary in order to proceed to vascular surgical repair
3. Determination of intraoperative HAV patency can be done by conventional angiography or ultrasound at the discretion of the investigator
4. Measured at preoperative screening when possible
5. PRA only collected at ET visit if the visit occurs before Month 6 collection
6. The status of the patient and HAV will be ascertained every 6 months until 3 years after HAV implantation at routine 6 month interval clinical visits or by telephone contact with the patient and/or his physician. The investigator or designee will complete a brief questionnaire covering the status of the patient, known patency of the HAV and any complication, interventions or other vascular procedures on the operative leg. The results of the clinical ultrasound surveillance if conducted will be collected.

† Patients withdrawn before Month 12 will perform ET visit

SUMMARY OF PROTOCOL CHANGES

All changes since Version 1.0 of the protocol and incorporated into the current protocol version (Version 2.0) are summarized below. Each change is also categorized as either related to safety and/or efficacy, which would require IRB/EC review, or administrative in nature, which does not require review by the IRB/EC.

Important note: No subjects were enrolled under Version 1.0, which was the initial protocol approved by the FDA.

Changes Incorporated in Protocol Version 2.0 (24 Oct 2016)	
Changes Related to SAFETY and/or EFFICACY	
Change and Reason for Change	**Section(s) Changed**
SAFETY CHANGE: Updated protocol to consistently reference use of the investigational HAV medicinal product as a "Bypass" or interposition vessel. Original protocol did not consistently reference use as a Bypass vessel (i.e., only interposition use was consistently referenced). **REASON:** Added additional consistency to protocol (i.e., for use as Bypass). The original protocol intended to specify use of the investigational HAV medicinal product as both an Interposition and Bypass vessel but did not consistently reference use as a bypass vessel. This change is highlighted under SAFETY and/or EFFICACY since certain reviewers of the protocol may not have understood that the investigational HAV medicinal product may be used to bypass vasculature in addition to an interposition bypass; therefore, these reviewers may not have considered risks associated with Bypass use.	Changes throughout protocol, including protocol title.
SAFETY/EFFICACY CHANGE: Modified Inclusion Criterion #2 to reduce the maximum required interposition bypass length for a damaged SFA or popliteal artery from 40 cm to 38 cm. **REASON:** Based on the available length of the current HAV (i.e., approximately 42 cm) the maximum length for interposition bypass is 38 cm or less to allow the HAV to be adequately prepared and positioned for surgical placement.	Protocol Summary Table – Inclusion Criterion # 2 5.1.1 – Patient Inclusion Criteria

Changes Incorporated in Protocol Version 2.0 (24 Oct 2016)	
Changes Related to SAFETY and/or EFFICACY	
Change and Reason for Change	**Section(s) Changed**
EFFICACY CHANGE: Reduced the total duration of the clinical study after completion of the final 12-month follow-up from 4 additional years to 2 additional years. Additionally, opened the two-year additional follow-up to allow "telephone contact with the patient and/or the physician designated to continue long-term care of the patient." **REASON:** Based on additional clinical trial experience with the patient population associated with this study, patients do not return to for additional follow-up once the defined follow-up period was achieved. Follow-up via telephone contact increases the likelihood to obtain longer-term follow-up information. The defined final endpoint follow-up duration remains 12 months.	Protocol Summary Table – Study Duration
SAFETY/EFFICACY CHANGE: Added requirement to obtain Panel Reactive Antibodies (PRA) Panel from pre-screening blood sample and at 6-month follow-up. **REASON:** To be consistent with changes to protocol. A baseline PRA will be obtained from all subjects to allow assessment of any immunologic reactive changes at 6 months following implantation of the investigational HAV medicinal product.	Schematic of Study Design
SAFETY CHANGE: Removed requirement to obtain subjects vital signs at pre-screening visit and during most follow-up visits. **REASON:** To be consistent with changes to protocol, which no longer requires initial documentation of subject patient vital signs due to potential challenges associated with trauma patients (e.g., shock, etc.).	Schematic of Study Design
SAFETY/EFFICACY CHANGE: Updated status/results regarding ongoing study to assess use of HAV for Vascular Access during Dialysis. **REASON:** To update protocol to include results and status of the HAV for vascular access during dialysis investigation. All clinical events were summarized, which included infection of the HAV by MRSA, two infections not involving the HAV and perigraft hematoma. None of the events referenced raised new concerns regarding safety or effectiveness.	2.4 – Summary of Clinical Studies, 2.4.3 – Vascular Access for Dialysis

Changes Incorporated in Protocol Version 2.0 (24 Oct 2016)	
Changes Related to SAFETY and/or EFFICACY	
Change and Reason for Change	**Section(s) Changed**
SAFETY/EFFICACY CHANGE: Updated conclusion to summarize new information added to Section 2.4. **REASON:** To update clinical studies conclusion section to be consistent with all new information added to Section 2.4. Conclusion indicates that the new information does not raise new concerns regarding safety or effectiveness of the investigational HAV medicinal product.	2.4 – Summary of Clinical Studies, 2.4.4 – Conclusion
SAFETY/EFFICACY CHANGE: Updated conclusion to summarize new information added to Section 2.4. **REASON:** To update clinical studies conclusion section to be consistent with all new information added to Section 2.4. Conclusion indicates that the new information does not raise new concerns regarding safety or effectiveness of the investigational HAV medicinal product.	6.8 – Essential, Precautionary and Prohibited Medications 6.8.1 – Essential Medications
SAFETY CHANGE: Updated: "All patients must have at least 1 day of antibiotic prophylaxis in accordance with local hospital guidelines." To: All patients must have at least 1 day of antibiotic prophylaxis *the same day as surgery* in accordance with local hospital guidelines." **REASON:** To improve alignment with other Humacyte protocols and to the established clinical site procedures.	6.8 – Essential, Precautionary and Prohibited Medications 6.8.1 – Essential Medications (Antibiotic Prophylaxis)
SAFETY CHANGE: Updated: "Intraoperative heparin: up to 150 IU/kg unfractionated heparin during surgery." To: "The doses of heparin to be used during surgery will be determined by the investigator." **REASON:** To improve alignment with other Humacyte protocols and to the established clinical site procedures.	6.8 – Essential, Precautionary and Prohibited Medications 6.8.1 – Essential Medications (Antithrombotic Prophylaxis)

Changes Incorporated in Protocol Version 2.0 (24 Oct 2016)	
Changes Related to SAFETY and/or EFFICACY	
Change and Reason for Change	**Section(s) Changed**
SAFETY CHANGE: Updated: "If antiplatelet therapy was not ongoing at the time of surgery it should be commenced as soon as possible post operatively. Antiplatelet therapy (ideally dual therapy with aspirin 75-325 mg and clopidogrel 75 mg daily should be continue long term while the graft is in place." To: "If antiplatelet therapy was not ongoing at the time of surgery it should be commenced as soon as *medically appropriate* post operatively. Antiplatelet therapy (ideally dual therapy with aspirin *81*-325 mg and clopidogrel 75 mg daily should be continue long term while the *HAV* is in place." **REASON:** To improve alignment with other Humacyte protocols and to the established clinical site procedures.	6.8 – Essential, Precautionary and Prohibited Medications 6.8.1 – Essential Medications (Antithrombotic Prophylaxis)
SAFETY CHANGE: Added the following information: • Blood samples for hematology, clinical chemistry at pre-operative screening and Day 5 and PRA at pre-operative screening and M6 • Pre-operative imaging (ultrasound or angiography) is at the discretion of the investigator based on the clinical condition of the patient. It may be omitted if not deemed necessary in order to proceed to vascular surgical repair **REASON:** To improve alignment with other Humacyte protocols and to the established clinical site procedures.	7 – STUDY PROCEDURES/ EVALUATIONS 7.1 – Clinical Evaluations
SAFETY CHANGE: Updating information regarding obtaining Vital Signs data, From: Vital signs (heart rate and blood pressure) - at screening, D1, D5, D29 and M3, M6, M9 and M12 or ET. To: Vital signs (heart rate, blood pressure and temperature) – at D5. **REASON:** To align with changes to protocol regarding collection of Vital Signs data.	7 – STUDY PROCEDURES/ EVALUATIONS 7.1 – Clinical Evaluations

Changes Incorporated in Protocol Version 2.0 (24 Oct 2016)	
Changes Related to SAFETY and/or EFFICACY	
Change and Reason for Change	**Section(s) Changed**
SAFETY CHANGE: Updating requirements for patient informed consent to allow a potential patient's Legal Representative to provide consent on behalf of the patient. **REASON:** To align with changes to protocol and other Humacyte protocols.	11.3 – Informed Consent

Changes Incorporated in Protocol Version 2.0 (24 Oct 2016)	
ADMINISTRATIVE Changes	
Change and Reason for Change	**Section(s) Changed**
CHANGE: Updated protocol to consistently characterize the investigational HAV medicinal product more specifically as a "vessel' rather than a "graft," which is a more general term. **REASON:** To improve specificity within the protocol. The term "vessel" more specifically defines the investigational Human Acellular Vessel (HAV) medicinal product's name and use.	Throughout protocol, including Protocol Title, where the investigational HAV medicinal/product is characterized.
CHANGE: Added name and title of Sponsor Medical Approver, who is Jeffrey H. Lawson, MD, PhD, - Chief Medical Officer, Humacyte, to protocol. **REASON:** To clearly identify who should sign the protocol on behalf of the Sponsor and for convenience (i.e., approver does not need to hand print their name and title.)	Protocol Approval Page
CHANGE: Removed Medical Monitor Approval signature block from Protocol Approval Page. **REASON:** The third-party Medical Monitor is not required to approve the clinical protocol. The requirements of the Medical Monitor are defined in a separate clinical contract that includes signature approvals.	Protocol Approval Page
CHANGE: Added City/State information (i.e., Baltimore, MD) to two of the three sites listed in the "Planned Study Sites" section. **REASON:** To assure consistency and accuracy. The Seattle, WA clinical site included city/state information, while the two Baltimore sites (i.e., Baltimore Shock Trauma & Johns Hopkins) did not include city/state information since their locations are well-known.	Protocol Summary Table – Planned Study Sites
CHANGE: Modified Inclusion Criterion #2 to define SFA as "Superficial Femoral Artery," replaced the term "graft" with "vessel" and added reference to "bypass." **REASON:** To add specificity and define acronyms within body of protocol.	Protocol Summary Table – Inclusion Criterion # 2 5.1.1 – Patient Inclusion Criteria

Changes Incorporated in Protocol Version 2.0 (24 Oct 2016)	
ADMINISTRATIVE Changes	
Change and Reason for Change	**Section(s) Changed**
CHANGE: Removed reference to "graft" when referring to anastomosis. **REASON:** A qualifier for the type of anastomosis was not required, and "graft" is not used to describe the HAV investigational medicinal product, which is more specifically described as a vessel.	Protocol Summary Table – Inclusion Criterion # 3
CHANGE: Added the term "intervals" to the 6-month follow-up times and removed reference to "vessel" before "removal" to define end of study. **REASON:** To improve clarity and specificity about the follow-up times occurring every 6 months (i.e., not 6 occurring monthly). The term "vessel" was removed since it was redundant since HAV was already identified.	Protocol Summary Table – Study Duration
CHANGE: Changed the sequence of endpoint categories, so Safety Endpoints are listed first. No endpoint was modified. Updated reference to "vessel" to state "HAV." **REASON:** To align with other Humacyte protocols and add specificity.	Protocol Summary Table – Study Duration
CHANGE: Updated references to "graft" or "vessel" to "HAV" and changed the acronym for US to ultrasound. **REASON:** To add specificity and clarity within protocol.	Schematic of Study Design
CHANGE: Updated the description of the investigational HAV medicinal product during follow-up assessments to state "bypass." **REASON:** To add specificity and alignment within protocol.	Schematic of Study Design
CHANGE: Updated name and contact information of Sponsor Clinical Operations Representative to "Angela Rose." **REASON:** Sponsor representative has changed and now reflects name and contact information for current representative.	Section 1 – STUDY PERSONNEL

Changes Incorporated in Protocol Version 2.0 (24 Oct 2016)	
ADMINISTRATIVE Changes	
Change and Reason for Change	**Section(s) Changed**
CHANGE: Updated name of CRO Safety Representative to "Audra Rodrigues." **REASON:** Safety representative has changed and now reflects name of current representative.	Section 1 – STUDY PERSONNEL
CHANGE: Corrected one minor typographical error and included references to "bypass" when "interposition" was mentioned. Also updated references to "graft" to state "conduit," as appropriate. **REASON:** To add specificity and alignment within protocol.	2.1 - Background Information 2.2 – Scientific Rationale
CHANGE: Updated Clinical Summary Overview to include updated status of current Phase 2 studies, which included a follow-up extending from 30 to 42 months, information about a new multi-center Phase 3 study to compare HAV to ePTFE grafts, and an additional multicenter Phase 2 study evaluating HAV as a vascular prosthesis for femoropopliteal bypass. **REASON:** To update protocol to summarize all current clinical investigations.	2.4 – Summary of Clinical Studies, 2.4.1 - Overview
CHANGE: Updated status/results regarding ongoing study to assess use of HAV for femoropopliteal bypass. **REASON:** To update protocol to include result and status of the HAV for femoropopliteal bypass investigation. None of the results raised issues regarding safety or efficacy of the HAV.	2.4 – Summary of Clinical Studies, 2.4.2 – Peripheral Arterial Bypass
CHANGE: Updated reference to "graft" to state "bypass conduit." **REASON:** To add specificity and consistency within protocol.	3.2 - Secondary Objective - Safety
CHANGE: Updated reference to "vessel" to state "HAV." **REASON:** To add specificity and consistency within protocol.	3.2 - Secondary Objective - Efficacy
CHANGE: Updated to specify follow-up endpoint to occur every 6 months until 36 months. **REASON:** To align with clarification changes to the protocol.	4.2.2 - Long Term Endpoints

Changes Incorporated in Protocol Version 2.0 (24 Oct 2016)	
ADMINISTRATIVE Changes	
Change and Reason for Change	**Section(s) Changed**
CHANGE: Updated criterion #4 from "Pregnant Women" to "Known Pregnant Women." **REASON:** To align with other Humacyte clinical protocols.	5.1.2 – Patient Exclusion Criteria
CHANGE: Updated instructions in the protocol for implanting the HAV to state: "Tunneling of the HAV, if required, must be performed using a sheathed tunneler. After inserting the assembled tunneler into the tissue, the inner mandrel of the tunneler should be removed from the sheath. The sheath is lubricated with saline and then with the silicone mandrel in place, the HAV can be easily pushed through the sheath without the need to tie to the inner mandrel and pulled through the tunneler (see study manual for details). **REASON:** Additional details were added to describe the tunneling procedure required to implant the HAV.	6.4 – Implantation of the Humacyte Human Acellular Vessel (HAV)
CHANGE: Updated name of "IMP Receipt Form" to "IMP Shipment Confirmation Form." **REASON:** To increase specificity and align with changes to the protocol.	6.5 – IMP Accountability Procedures
CHANGE: Added information to allow the signed IMP form to also be returned to an authorized designee of Humacyte. Also specified that the Original signed document shall be maintained in the Investigator's Files. **REASON:** To align with other Humacyte clinical protocols and procedures.	6.5 – IMP Accountability Procedures
CHANGE: Removed the following instructions regarding the IMP Accountability Log: "at which time the original will be retrieved by Humacyte or their authorized designee and a copy kept at the site. **REASON:** This information was redundant and already included in Humacyte-established procedures.	6.5 – IMP Accountability Procedures
CHANGE: Added "prescription and over the counter (OTC)" before the word "medications" to add specificity. **REASON:** To align with other Humacyte clinical protocols.	6.7 – Prior and Concomitant Medications

Changes Incorporated in Protocol Version 2.0 (24 Oct 2016)	
ADMINISTRATIVE Changes	
Change and Reason for Change	**Section(s) Changed**
CHANGE: Medication History section From: "Medication History – prescription and non-prescription medication from Day -7 onwards (see 6.5 above). Particular attention should be paid to the identification of OTC medications containing aspirin." To: "Medication History – prescription and *OTC* medication from Day -7 onwards (see *Section* 6.*7*). Particular attention should be paid to the identification of OTC medications containing aspirin. **REASON:** To provide additional clarity and update subsection number from 6.5 to 6.7.	7 – STUDY PROCEDURES/ EVALUATIONS 7.1 – Clinical Evaluations
CHANGE: Updated information regarding imaging by ultrasound, From: **"**● Intraoperative angiography to assess anastomotic anatomy, patency and runoff. ● Duplex ultrasound – clinical assessment at all postoperative visits from day 29 onwards, to assess HAV patency, mid HAV diameter and flow rate. The full length of the HAV should be imaged at each assessment to monitor for aneurysm development" To: **"**● Intraoperative angiography *or ultrasound* to assess anastomotic anatomy, patency and runoff. ● Duplex ultrasound – clinical assessment at all postoperative visits from day 29 *thru M12*, to assess HAV patency, mid HAV diameter and flow rate. The full length of the HAV should be imaged at each assessment to monitor for aneurysm development" "Medication History – prescription and *OTC* medication from Day -7 onwards (see *Section* 6.7). Particular attention should be paid to the identification of OTC medications containing aspirin. **REASON:** To provide additional clarity. Use of ultrasound was an established imaging modality in Version 1 of the protocol.	7 – STUDY PROCEDURES/ EVALUATIONS 7.1 – Clinical Evaluations

Changes Incorporated in Protocol Version 2.0 (24 Oct 2016)	
ADMINISTRATIVE Changes	
Change and Reason for Change	**Section(s) Changed**
CHANGE: Added "pre-operative" before "screening." Updated section related to Clinical Chemistry From: "Clinical chemistry: sodium, potassium, calcium, blood urea nitrogen, creatinine, albumin, total bilirubin, ~~alanine aminotransferase (ALT), aspartate aminotransferase (AST), gamma glutamyl transferase, alkaline phosphatase~~, glucose (non-fasting)." To: "Clinical chemistry: sodium, potassium, calcium, blood urea nitrogen, creatinine, albumin, total bilirubin, glucose (non-fasting). *PRA will be measured at pre-operative screening and Month 6*" **REASON**: To provide additional clarity and to align with Humacyte and clinical site protocols.	7.2.1 – Clinical and Research Laboratory Evaluations and Specimen Collection
CHANGE: Updated protocol instructions regarding processing specimens, From: **"**Biochemistry and Hematology Tubes – transport to the appropriate local clinical laboratory according to institutional procedures." To: "Biochemistry *(no additives)* and Hematology *(EDTA)* Tubes – *collect and* transport to the appropriate local clinical laboratory according to institutional procedures. *To avoid cross-contamination of additives between tubes, the order of draw is as follows:* *FIRST: Draw two biochemistry tubes (no additives) – the first is for clinical chemistry, the second is for PRA [See below for processing of this sample.]* *SECOND: Draw one hematology tube - (EDTA tube) for hematology studies* *Second Non-Additive Red Top Tube for PRA: Allow sufficient time for the blood to clot and then centrifuge the tube to separate the serum from clotted material. Transfer at least 0.5 ml of serum into a pre-labeled screw cap vial (provided by sponsor) for PRA testing. Transfer this vial to a secure, monitored freezer (≤ -20°C). PRA samples will be shipped to Humacyte, Inc. in batches.* **REASON**: To provide additional clarity and to align with Humacyte and clinical site protocols.	7.2.2 – Specimen Preparation, Handling and Shipping

Changes Incorporated in Protocol Version 2.0 (24 Oct 2016)	
ADMINISTRATIVE Changes	
Change and Reason for Change	**Section(s) Changed**
CHANGE: Updated protocol instructions regarding test equipment to exclude PRA testing. From: "All laboratory tests will be conducted at certified hospital laboratories. Routine monitoring, maintenance or calibration of laboratory equipment is required per local site procedures." To: "All laboratory tests *(except assay of PRA)* will be conducted at certified hospital laboratories. Routine monitoring, maintenance or calibration of laboratory equipment is required per local site procedures. *Samples for PRA analysis will be shipped to Humacyte for analysis at a central laboratory.*" **REASON**: To align with protocol changes and provide additional clarity regarding samples for PRA analysis since testing is outsourced.	7.2.3 – Monitoring, Maintenance, and Calibration of Equipment
CHANGE: Added Section 7.3.1 to describe evaluation by CT or conventional angiography. The following information was added: "7.3.1 **CT Angiography and Conventional Angiography** CT angiography or conventional angiography will be conducted as pre-operative screening when feasible. Pre-operative imaging is at the discretion of the investigator based on the clinical condition of the patient. It may be omitted if not deemed necessary in order to proceed to vascular surgical repair. Determination of intraoperative HAV patency on Day 1 can be done by conventional angiography or ultrasound at the discretion of the investigator." **REASON**: To align with protocol changes and provide additional clarity regarding requirements for pre-operative imaging.	7.3 – Imaging Evaluation

Changes Incorporated in Protocol Version 2.0 (24 Oct 2016)	
ADMINISTRATIVE Changes	
Change and Reason for Change	**Section(s) Changed**
CHANGE: Shifted information regarding use of Duplex Ultrasound to section 7.3.2, updated 1-month timepoint to "Day 29," and added the following additional information: "*Determination of intraoperative HAV patency on Day 1 can be done by conventional angiography or ultrasound at the discretion of the investigator*." **REASON**: To align with protocol changes and provide additional clarity regarding requirements for pre-operative imaging.	7.3.2 – Duplex Ultrasound (previously Section 7.3.1)
CHANGE: Added the following additional information: *"Standard of care procedures such as laboratory evaluations conducted prior to screening may be used rather than repeating the test.*" +Added requirement to perform PRA +Added following information regarding imaging: "*Ultrasound or CT angiography (CTA) (pre-op imaging is at the discretion of the investigator based on the clinical condition of the patient. It may be omitted if not deemed necessary in order to proceed to vascular surgical repair."* -Removed requirement for obtaining vital signs prior to surgery (Day 1). -Removed requirement to obtain the following values for blood chemistry testing: Alkaline Phosphatase, AST, and ALT. **REASON**: To provide additional clarity and align with other Humacyte protocols.	7.4.1 – Pre-Operative Screening (Day 1)

Changes Incorporated in Protocol Version 2.0 (24 Oct 2016)	
ADMINISTRATIVE Changes	
Change and Reason for Change	**Section(s) Changed**
CHANGE: Updated Day 1 enrollment information, From: "The HAV will be implanted as an interposition graft in the superficial femoral or popliteal artery using standard vascular surgical techniques. Details of the surgical anatomy and any complications will be documented." To: "The HAV will be implanted as an interposition *replacement or bypass vessel* in the superficial femoral or popliteal artery using standard vascular surgical techniques. Details of the surgical anatomy and any complications will be documented. *Determination of intraoperative HAV patency can be done by conventional angiography or ultrasound at the discretion of the investigator.*" **REASON**: To provide additional clarity and align with other Humacyte protocols.	7.4.2 – Enrollment - Day 1 (HAV Implantation)
CHANGE: Changed the following information regarding Follow-up Visits: Day 5: Added detail regarding the specific Vital Signs to collect (i.e., "heart rate, blood pressure, and temperature)" Day 29: Added "Any HAV" before "interventions" Day 29: Removed requirement to collect Vital Signs Months 3, 6, & 9: Added "Any HAV" before "interventions" Months 3, 6, & 9: Added "PRA (Month 6 only)" Month 12 and Early Termination: Added "HAV" before "interventions" **REASON**: To provide additional clarity and align with other Humacyte protocols.	7.4.3 – Follow-up Visits
CHANGE: Added the following information regarding early termination visit: "PRA will be collected at ET visit if the visit occurs before Month 6 collection of the sample." **REASON**: To provide additional clarity and align with other Humacyte protocols.	7.4.5 – Early Termination Visit

Changes Incorporated in Protocol Version 2.0 (24 Oct 2016)	
ADMINISTRATIVE Changes	
Change and Reason for Change	**Section(s) Changed**
CHANGE: Added the following information regarding Follow-up from Months 18-36: "This information will be entered onto a form, which will be returned to the CRO / sponsor after each follow up visit or call. These data will not be verified against source documents, and the investigational sites will not undergo additional monitoring now that the main study is complete. The patient should be withdrawn from the study if the HAV is removed or becomes permanently occluded (loss of secondary patency)." **REASON:** To provide additional clarity and align with other Humacyte protocols.	7.4.7 – Follow-up from Months 18-36
CHANGE: Added the following additional specificity regarding when AE reporting process begins under the protocol: AE reporting begins from *"time of anesthesia induction for*" implantation of the HAV. **REASON:** To provide additional clarity and align with other Humacyte protocols.	8.2 – Reporting of Adverse Events
CHANGE: Added the following additional specificity to the risk assessment regarding likelihood that an AE is related to the IMP: "*Unlikely Related: A clinical event, including an abnormal laboratory test results, whose temporal relationship makes a causal relationship improbable (e.g., the event did not occur within a reasonable time after placement of the IMP) and in which other drugs or chemicals or underlying disease provide plausible explanations (e.g., the subject's clinical condition, other concomitant treatments).* **REASON:** To provide additional clarity and align with other Humacyte protocols and Risk Management.	8.2 – Reporting of Adverse Events

Changes Incorporated in Protocol Version 2.0 (24 Oct 2016)	
ADMINISTRATIVE Changes	
Change and Reason for Change	**Section(s) Changed**
CHANGE: Modified instructions to indicate that the SPONSOR makes the final determination regarding the causality/relationship of an event to the IMP. **REASON:** To align with other Humacyte protocols and Investigator Contracts.	8.2 – Reporting of Adverse Events
CHANGE: Added reference to complete the "Pregnancy Outcome and Report Form" to document a pregnancy during the study. **REASON:** To align with other Humacyte protocols.	8.3 – Reporting of Pregnancy
CHANGE: Added the following information to better define when a hospitalization is considered to have occurred under the study, "*excluding pre-planned or elective procedures*." **REASON:** To add specificity and align with other Humacyte protocols.	9.2 – Hospitalization
CHANGE: Removed all references to "Drug" and added reference to "IMP." **REASON**: To align with other Humacyte protocols and the investigation article is an investigational medicinal product.	9.3 – Unexpected Adverse Reactions (previously, Unexpected Adverse Drug Reactions)
CHANGE: Added instruction for investigators indicating that any follow-up reports should be completed and submitted "on a new SAE Form." **REASON**: To align with other Humacyte protocols.	9.4 – Reporting Serious Adverse Events

Changes Incorporated in Protocol Version 2.0 (24 Oct 2016)	
ADMINISTRATIVE Changes	
Change and Reason for Change	**Section(s) Changed**
CHANGE: Updated entire section, From: A DSMB will be convened by the Sponsor to review data ~~from a randomized phase 3 study using the HAV. This DSMB will also have safety oversight of this study and any other ongoing phase 2 studies using the HAV.~~ To: A *Data Safety Monitoring Board (*DSMB) will be convened by the Sponsor to review *safety* data *on an ongoing basis for this study*. *The DSMB will consist of individuals who are not directly involved in the conduct of the study. A separate charter will be established that will describe the roles and responsibilities of the DSMB.* **REASON**: To align with other Humacyte protocols and the Humacyte DSMB process.	9.6 – Data Safety Monitoring Board
CHANGE: Updated IRB requirements to indicate that the list of IRB members, their titles or occupations, and their affiliations "may be obtained, if available," rather than "must" be obtained. **REASON**: To align with other Humacyte protocols and IRB policies/capabilities.	11.2 – Institutional Review Board
CHANGE: Updated Clinical Visit Summary Table to reflect all changes to the clinical protocol. Representative changes include: Added column to table for "M18 - M36 Routine SOC Visit" Added row to table to indicate visits for testing PRA (i.e., Pre-Op, M6, and M12). Added the following footnotes: 3. Determination of intraoperative HAV patency can be done by conventional angiography or ultrasound at the discretion of the investigator 4. Measured at preoperative screening when possible 5. PRA only collected at ET visit if the visit occurs before Month 6 collection 6. The status of the patient and HAV will be ascertained every 6 months until 3 years after HAV implantation at routine 6 month interval clinical visits or by telephone contact with the patient and/or his physician. The investigator or designee will complete a brief questionnaire covering the status of the patient, known patency of the HAV and any complication, interventions or other vascular procedures on the operative leg. The results of the clinical ultrasound surveillance if conducted will be collected. **REASON:** To align with changes to the protocol.	APPENDIX 1 – HAV CLINICAL VISIT SCHEDULE

Changes Incorporated in Protocol Version 2.0 (24 Oct 2016)	
ADMINISTRATIVE Changes	
Change and Reason for Change	**Section(s) Changed**
CHANGE: Minor text clarifications were made. **REASON**: To improve clarity and reduce typographical errors	Throughout entire protocol

A Phase 2 Study for the Evaluation of Safety and Efficacy of Humacyte's Human Acellular Vessel for Use as a Vascular Interposition Graft in Patients with Limb-threatening Peripheral Arterial Trauma

Medicinal Product:	Humacyte Human Acellular Vessel (Humacyte HAV)
Study No.:	CLN-PRO-V005

Sponsor:	Humacyte, Inc
Address:	7020 Kit Creek Road
	PO Box 12695
	Research Triangle Park, NC 27560
Phone:	919-313-9633
Fax:	919-313-9634

CRO:	Atlantic Research Group
	2421 Ivy Road, Suite 200
	Charlottesville, VA 22903

Version:	1.0
	29 December 2015

Confidentiality Statement

Statement of Compliance

This trial will be conducted in compliance with the protocol and the following regulatory requirements:

- Declaration of Helsinki adopted by the 18th World Medical Assembly in Helsinki, Finland, in 1964, as last amended by the World Medical Assembly in 2013
- International Conference on Harmonization of Technical Requirements for Registration of Pharmaceuticals for Human Use (ICH), E6 Good Clinical Practice: Consolidated Guidance (ICH E6)
- ICH E2A Clinical Safety Data Management: Definitions and Standards for Expedited Reporting
- ICH E8 Guidance on General Considerations for Clinical Trials
- Applicable sections of United States Food and Drug Administration (FDA) Code of Federal Regulations (CFR), including:
    - 21 CFR Part 11, Electronic Records; Electronic Signatures
    - 21 CFR Part 50, Protection of Human Subjects
    - 21 CFR Part 54, Financial Disclosure by Clinical Investigators
    - 21 CFR Part 56, Institutional Review Boards
    - 21 CFR Part 312, Investigational New Drug Application

Principal Investigator Agreement Page for the Protocol

I agree:

- To assume responsibility for the proper conduct of the study at this site, and to conduct the study in compliance with this protocol, any future amendments, and with any other study conduct procedures provided by the sponsor, Humacyte, Incorporated (Humacyte), or their authorized representatives.

- Not to implement any deviations from or changes to the protocol (including protocol amendments) without agreement from the sponsor and prior review and written approval from the Institutional Review Board (and FDA, if applicable) except where necessary to eliminate an immediate hazard to the patient(s), or for administrative aspects of the study (where permitted by all applicable regulatory requirements).

- That I am familiar with the appropriate use of the investigational medicinal product, as described in this protocol and any other information provided by the sponsor including, but not limited to the current Investigator's Brochure or equivalent document provided by Humacyte.

- To ensure that all persons assisting me with the study are adequately informed about the investigational medicinal product and of their study-related duties and functions.

- That I have been informed that certain regulatory authorities require the sponsor to obtain and supply details about the investigator's ownership interest in the sponsor or the Investigational Medicinal Product, and more generally about his/her financial ties with the sponsor. Humacyte will use and disclose the information solely for the purpose of complying with regulatory requirements.

Principal Investigator: _____

Name and Title

Signed: _____ Date: _____

Protocol Approval

Sponsor Medical Approval: JEFFREY H. LAWSON, MD. Ph.D, Chief Medical Officer, Humacyte

Name and Title

Signed: _____ Date: 29-Dec-2015

Table of Contents

List of In-text Tables

List of Abbreviations

AE	Adverse event
AIDS	Acquired immunodeficiency syndrome
ALT	Alanine aminotransferase
aPTT	Activated partial thromboplastin time
ASA	Acetylsalicylic acid (aspirin)
ABI	Ankle brachial index
AST	Aspartate aminotransferase
AV	Arteriovenous
AVF	Autologous arteriovenous fistula
BP	Blood pressure
CAVG	Canine acellular vascular graft
CBC	Complete blood count
CKD	Chronic kidney disease
CT	Computed tomography
CM	Concomitant medication
eCRF	Electronic case report form
CRO	Contract research organization
DSMB	Data Safety Monitoring Board
DTH	Delayed-type hypersensitivity
ECG	Electrocardiogram
ECM	Extracellular Matrix
ePTFE	Expanded polytetrafluoroethylene
ESRD	End-stage renal disease
ET	Early termination
FDA	Food and Drug Administration
GCP	Good Clinical Practice
GGT	Gamma glutamyl transpeptidase
GLP	Good Laboratory Practice
HAV	Human acellular vessel
HIV	Human immunodeficiency virus
IB	Investigator Brochure
ICF	Informed consent form
ICH	International Conference on Harmonization of Technical Requirements for Registration of Pharmaceuticals for Human Use
IgG	Immunoglobulin G

List of Abbreviations

IHC	Immunohistochemistry
IM	Intramuscular
IMP	Investigational medicinal product
INR	International normalized ratio
IRB	Institutional Review Board
ISO	International Organization for Standardization
IU	International unit
IV	Intravenous
MedDRA	Medical Dictionary for Regulatory Activities
M	Month
N	Number (typically refers to participants)
NYHA	New York Heart Association
OTC	Over-the-counter
PAD	Peripheral arterial disease
PE	Physical examination
PHI	Protected health information
PI	Principal Investigator
PRA	Panel reactive antibodies
PT	Prothrombin time
PTFE	Polytetrafluoroethylene
QA	Quality Assurance
QC	Quality Control
RRT	Renal replacement therapy
SAE	Serious adverse event
SFA	Superficial Femoral Artery
SOP	Standard operating procedure
SVS WIfI	Society for Vascular Surgery: Wound, Ischemia, and foot Infection
US	Ultrasound
USA	United States of America
WFI	Water for injection
WBC	White blood cell(s)
WHO	World Health Organization

Protocol Summary

Full Title	A Phase 2 Study for the Evaluation of Safety and Efficacy of Humacyte's Human Acellular Vessel for Use as a Vascular Interposition Graft in Patients with Limb-threatening Peripheral Arterial Trauma
Clinical Trial Phase	Phase 2
Sponsor	Humacyte, Inc.
Planned Study Sites	Up to 3 sites to include: Baltimore Shock Trauma Johns Hopkins Harborview Medical Center, Seattle, WA
Sample Size	Up to 20 patients

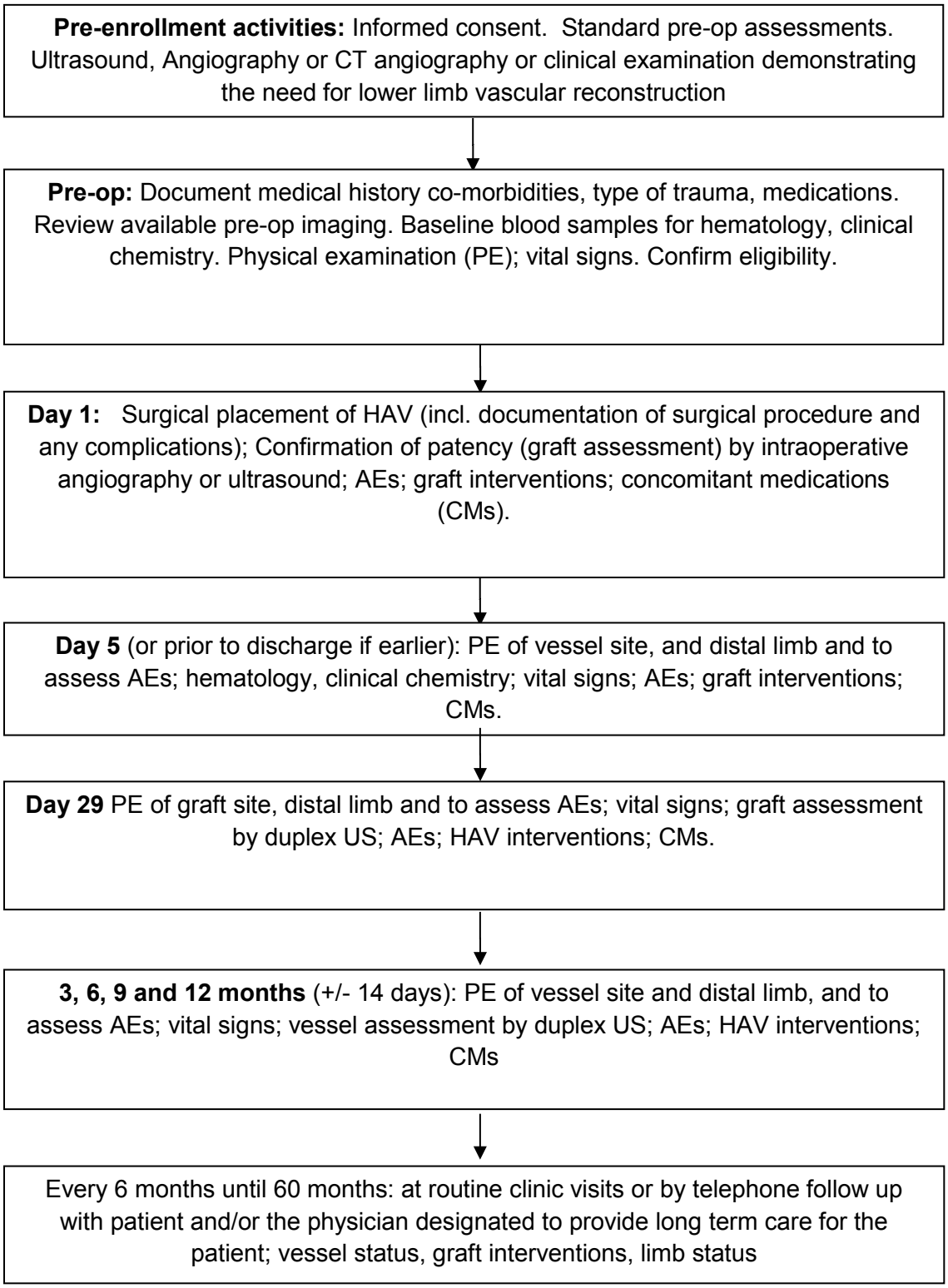
Study Population	Patients with peripheral arterial trauma
Inclusion Criteria	1. Patients with lower limb vascular trauma which threatens the viability of the leg and who require reconstruction of the superficial femoral or popliteal artery 2. Preoperative ultrasound or angiography or CT angiography or clinical examination indicates damage to the SFA or popliteal artery requiring reconstruction with interposition of a vascular graft AND required graft length of ≤ 40cm 3. Proximal graft anastomosis is expected to be to the common femoral artery below the inguinal ligament or to the superficial femoral artery or popliteal artery 4. Distal anastomosis is expected to be to the SFA, popliteal artery or the tibio-peroneal trunk 5. Autologous vein graft is either not feasible in the judgment of the treating surgeon (e.g. because of lack of availability of suitable conduit, presence of severe venous insufficiency) or is not desirable because of the urgency of revascularization 6. Aged 18 to 85 years old, inclusive 7. Able to communicate meaningfully with investigative staff and able to comply with study procedures. If the patient is unconscious then information from a reliable witness indicates that the patient would normally be able to understand and comply with study procedures 8. Patient or legal representative is able, willing and competent to give informed consent 9. Life expectancy of at least 1 year
Exclusion Criteria	1. Limb at high risk of amputation despite vascular reconstruction e.g., because of crush injury 2. Life threatening head, chest, or abdominal injuries that make survival unlikely 3. Distal anastomosis planned to a tibial or pedal artery 4. Pregnant women 5. Known medical condition which would preclude long term dual antiplatelet therapy after resolution of acute injuries 6. Any other condition which in the judgment of the investigator would preclude adequate evaluation of the safety and efficacy of the Humacyte Human Acellular Vessel (HAV)

	7. Previous enrollment in this study 8. Employees of the sponsor or patients who are employees or relatives of the investigator
Expected Enrollment Start	2Q 2016
Accrual Period	12 months
Study Duration	The active study duration for each study participant will be 12 months from HAV implantation or until HAV failure/ vessel removal/ death if earlier. Additional follow up 6 monthly until 36 months will involve the capture of information on assessments performed at “standard of care” routine clinic visits or by telephone follow up with the patient or his/her physician. There are no study specific visits or procedures after 12 months. The total expected duration of the clinical study is 24 months (plus 4 years additional follow up at routine clinical visits).
Study Design	Prospective, multicenter, single arm, non-randomized study
Investigational Device/Intervention Description	Patients will be implanted with a Humacyte Human Acellular Vessel (HAV) as an interposition graft using standard vascular surgical techniques.

Primary Objectives	**Safety** To evaluate the safety and tolerability of the Humacyte HAV in patients with vascular trauma in the lower limb who are undergoing vascular reconstructive surgery **Efficacy** To determine the patency (primary, primary assisted and secondary) rate of the Humacyte HAV at 12 months To determine the rate of limb salvage
Secondary Objectives	**Safety** • To determine mechanical stability of the HAV based on freedom from aneurysmal degeneration on duplex ultrasound • To determine HAV durability in terms of freedom from need for graft explantation or replacement due to infection, bleeding, or conduit degeneration **Efficacy** • To determine the patency of the HAV (primary, primary assisted and secondary) at 3, 6 and 9 months • To determine the rates of interventions needed to maintain / restore patency in the HAV over 12 months
Endpoints	All endpoints will be assessed over a period of up to 12 months after vessel implantation. The primary analysis of the study will be based on the 12 month follow up of all patients who receive an HAV. Efficacy Endpoints: • Incidence of limb salvage • Vessel patency rates (i.e., primary, primary assisted, and secondary) • HAV interventions

	Safety Endpoints: • Incidence of aneurysm formation, anastomotic bleeding or rupture, HAV infection, HAV removal, and irritation/inflammation at the HAV implantation site • Frequency and severity of adverse events Long term Endpoints (Every 6 months until 36 months): • Limb viability • HAV survival • Patient survival
Protocol Approval (Version and Date)	Version 1.0 29 December 2015

Schematic of Study Design:



1. STUDY PERSONNEL

An updated list of all study personnel will be maintained by the CRO. Protocol amendments will not be required for staff changes at Humacyte, the CRO or the sites (except change of Principal Investigator at a site).

CRO Project Manager:

Brad Whitlow
Project Manager
Atlantic Research Group
2421 Ivy Road, Suite 200
Charlottesville, VA 22903

Email: bwhitlow@atlanticresearchgroup.com
Mobile: 828-442-0526
Office: 434-202-4537
Fax: 866-851-9318

Sponsor Medical Representative:

Jeffrey H. Lawson, MD, PhD
Chief Medical Officer
Humacyte, Inc.
PO Box 12695
7020 Kit Creek Road
Research Triangle Park, NC 27709, USA

Email: lawson@humacyte.commailto:
Mobile: 919-630-7851
Office: 919-313-9633 ext. 130

Sponsor Regulatory Representative:

William Tente
Vice President Regulatory Affairs
Humacyte, Inc.
PO Box 12695
7020 Kit Creek Road
Research Triangle Park, NC 27709, USA

Email: tente@humacyte.com
Mobile: 401-714-4151
Office: 919-313-9633 ext. 236

Sponsor Clinical Operations Representative:

John M. Adams, PharmD
Sr. Director, Clinical Development
Humacyte, Inc.
PO Box 12695
7020 Kit Creek Road
Research Triangle Park, NC 27709, USA

Email: adams@humacyte.com
Mobile: 919-605-5713
Office: 919-313-9633 ext. 276

CRO Safety Representative:

Samuel Bittar
Sr. Drug Safety Specialist
Atlantic Research Group
2421 Ivy Road
Charlottesville, VA 22903

Email: safety@atlanticresearchgroup.com
Phone: 888-619-3216
Fax: 866-851-9318

Medical Monitor:

Jennifer Prichard, MD
Medical Monitor
Atlantic Research Group
2421 Ivy Road
Charlottesville, VA 22903

Email: jprichard@atlanticresearchgroup.com
Mobile: 434-249-5530
Phone: 888-619-3216
Fax: 866-851-9318

2. BACKGROUND INFORMATION AND SCIENTIFIC RATIONALE

2.1 Background Information

In modern combat, the incidence of vascular injury is much greater than in previous wars. The rate of vascular injury in the Vietnam War was 2-3%. But between 2002-2009, the rate of vascular injury was over 12% in a study of over 13,000 battlefield injuries. (Rasmussen, 2010] Improvised Explosive Devices, or IED's, cause more than 3,000 casualties per year – and this number is rising. (Cordesman, 2010) Arterial damage, laceration and thrombosis can require vascular reconstruction to save tissues from ischemia, necrosis, and further amputation. There are two current methods for vascular reconstruction: harvesting of autologous vein, or using synthetic graft materials, and each has important limitations. Harvesting autologous vein for vascular reconstruction is problematic because IED casualties often have multi-limb injuries, making harvest of autologous vein highly risky or impossible. (Holcomb, 2011) Synthetic vascular reconstruction using synthetic vascular grafts made from Teflon (ePTFE)/Dacron is relatively contraindicated, since IED wounds are always "dirty", and bacteria in the wound can colonize the synthetic graft, causing abscesses and sepsis.

In the civilian population, lower extremity bone fractures with associated arterial injuries are common, due to from motor vehicle accidents, gunshot wounds, dog bites, and other situations resulting in blunt or penetrating trauma. (Helfet, 1990, Andrikopoulos, 1995, Akingba, 2012) In fact, the incidence of this type of vascular injury has increased considerably in the past 50 years. (Andrikopoulos, 1995) Although this type of injury represents less than 1% of all civilian injuries, fractures with associated vascular damage require special attention because of their potentially severe complications, including limb necrosis and amputation. Currently, in order to attempt to salvage the injured limb, the vascular component of these injuries are treated with interposition grafting, and these grafts are usually created either using the patient's saphenous vein or a synthetic ePTFE graft. However, the use of these grafts in the civilian population has many of the same downsides that accompany their use in combat injuries. The patient may not have adequate saphenous vein for harvest, and in many of these cases, such as dog bite injuries, the wound is "dirty" and a synthetic vascular substitute such as ePTFE is contraindicated due to the risk of infection. (Akingba, 2012) Thus, civilians would also benefit from a vascular graft that is available "off-the-shelf" – one that does not contain synthetic material and that has the same properties as human tissue, but does not require harvesting of vessel or cells from the patient.

There is thus a need for alternative grafts, which more closely mimic human vascular tissue that may avoid or reduce the complications associated with ePTFE and Dacron.

2.2 Scientific Rationale

Humacyte, Inc. (Humacyte) has developed an acellular, human collagen-based vascular graft (human acellular vessel – HAV) to provide an alternative to synthetic materials and to autologous grafts in the creation of vascular access for dialysis and for use in peripheral vascular bypass surgery or as an interposition graft to repair traumatic arterial damage. Because this product mimics native vascular tissue, it possesses all of the advantages of an autologous graft; it also has the benefits of synthetic grafts in that it is available off-the-shelf. Use of an off-the-shelf product avoids the surgical morbidity associated with vein graft harvest and most importantly allows vessel bypass surgery in patients who have no suitable veins available. Because the product mimics native vessel, it does not have the compliance mismatch associated with synthetic alternatives. In addition, pre-clinical studies in pigs, canines and primates have shown that the HAVs resist intimal hyperplasia at the anastomoses. (Quint, 2011, Prichard, 2011, Dahl, 2011) Upon implantation, it is anticipated (based on pre-clinical studies) that the collagen matrix comprising the HAV will be infiltrated with host cells and remodeled by the host. This will result in a vascular structure more similar to the histological composition of the native vascular tissue that may improve graft longevity and be less likely to become infected. The latter potential advantage is of high importance in the repair of peripheral vascular trauma where most wounds are heavily contaminated.

2.3 Summary of Nonclinical Information

The non-clinical testing program was designed to comprehensively address:

- local and systemic effects of the product in multiple in vivo animal models acutely and chronically,
- functional aspects of product implanted into animal models as an arteriovenous conduit
- biocompatibility of the HAV material in standardized in vitro and in vivo test protocols.

Overall, the results of these studies indicated that the HAV extracellular matrix material was non-toxic, well tolerated, and met standards for biocompatibility. Generally, the HAVs functioned as intended and maintained patency during the implantation period. (See the Investigator Brochure for a detailed summary of non-clinical data.)

Pre-implantation, the HAV has mechanical properties (burst pressure and suture retention strength) comparable with native human artery and vein. (**Table 1**.) There was no evidence that HAV strength deteriorated after long-term implantation into baboons.

Table 1: Summary of Mechanical Properties of Explanted Acellular Vessels

Test Material	Burst Pressure (mm Hg)	Suture Strength (g)
Pre-Implant Humacyte HAVs	3415 +/- 1011 (n=4)	180 +/- 44 (n=12)
Post-Explant Humacyte HAVs	3669 +/- 1305 (n=5)	276 +/- 84 (n=11)
Human Saphenous Vein	1,680 – 2,273[a]	196 +/- 2 (n=7)[a]
Human Artery	2,031 – 4,225[a]	200 +/- 119 (n=9)[a]

[a] From L'Heureux et al, *Nature Medicine*, 2006. (L'Heureux, 2006)

In the chronic animal testing Humacyte vessels produced using canine cells were implanted into 12 dogs (canine acellular vascular graft, CAVG) and 14 baboons (human acellular vessel -HAV) in a variety of anatomical locations. In general, the Humacyte vessels were safe and well tolerated, and functioned as intended.

Mechanical failure was not observed in any HAV. Calcification was not observed in any CAVG or HAV. No graft exhibited hemodynamically significant intimal hyperplasia. Unlike with ePTFE graft implantation, no evidence of systemic infection attributable to implantation of HAV was observed in any of the animals. One HAV developed an aneurysm that was resected and did not harm the animal. The HAV material showed no evidence of toxicity in hematology, clinical chemistry, and necropsy data. The HAVs could be accessed by venipuncture and hemostasis was achieved following needle puncture.

On microscopic analysis, the HAVs were found to be well integrated into the host tissue. Overall, the cellular host response to the HAVs demonstrated smooth muscle actin-positive cells within the vessel wall, endothelial cells lining the lumen, and an adventitial-like outer layer adjacent to the vessel. These findings indicate that implanted HAVs were populated with cell types that are characteristic of healthy native vasculature. Examination of the anastomotic sections showed that the HAVs were well integrated with adjoining vasculature with minimal intimal hyperplasia observed. Furthermore, IHC was employed to identify CD-68 positive macrophages in the venous intimal tissue. Studies have shown a substantial macrophage population has been observed within venous intimal tissue adjacent to inflammatory ePTFE arteriovenous grafts. (Kelly, 2002, Roy-Chaudhury, 2001) Only sparse CD-68 positive macrophages were observed, indicating that the degree and the aggressiveness of the intimal hyperplasia associated with the HAV were less than that typically associated with ePTFE grafts. (Prichard, 2011)

Over time, the organization and composition of extracellular matrix (ECM) components indicated that, aided by infiltration of host vascular cells, HAVs were remodeled in vivo in a manner that mimicked the dynamic remodeling process of native blood vessels. Given the difficulties associated with the baboon animal model, where mismatches in vein vs graft diameter were encountered and animals perturbed their wounds postoperatively, an overall assisted patency rate of approximately 80% (11/14) was achieved. In a xenogeneic transplant model that did not employ immunosuppression, the HAV material did not elicit biologically significant cellular or delayed-type hypersensitivity (DTH) immune responses. All animals developed immunoglobulin G (IgG) titers to the HAV material that did not appear to detrimentally impact vessel function.

In internationally recognized in vitro and in vivo International Organization for Standardization (ISO) test protocols, the HAV material met criteria for biocompatibility required of medical devices.

These data collectively support the safety of the HAV for the proposed clinical investigation.

2.4 Summary of Clinical Studies

2.4.1 Overview

Three phase 2 clinical studies using the HAV are currently ongoing, two in vascular access for dialysis and one in peripheral arterial bypass surgery. Eighty patients have so far been implanted with an HAV with the longest follow up being more than 30 months. More information on the clinical profile of the HAV in these ongoing studies is provided in the Investigator Brochure.

2.4.2 Peripheral Arterial Bypass

A study is ongoing in Poland that is evaluating the HAV as femoro-popliteal bypass in patients with peripheral arterial disease (PAD). Recruitment began in October 2013 and was completed in June 2014 with 20 patients implanted.

This is a single group uncontrolled study being conducted at 3 sites in Poland. Eligible patients require a femoro-popliteal bypass graft for the management of symptomatic peripheral arterial disease. Pre-operative imaging (angiography or CT angiography) must have demonstrated at least two below knee vessels patent to the ankle with good runoff. The proximal anastomosis must be expected to be below the inguinal ligament and the distal anastomosis above the knee. Autologous vein grafts must not suitable or feasible (e.g., because of severe venous disease or prior use of leg veins for other bypass surgery or there is a clinical need to preserve those veins for future bypass surgery in the coronary or peripheral circulation).

The HAV is implanted using standard vascular surgical techniques and the patency of the graft confirmed by intraoperative angiography or ultrasound. The patient is then followed up at study visits at 15 days, 6 weeks and 3, 6, 12, 18 and 24 months. Dual antiplatelet therapy with aspirin and clopidogrel is continued while the HAV is in situ. From 24 to 60 months there are no study-specific visits but the status of the patient and the HAV status will be ascertained yearly at routine clinical visits or by phone calls to the patients or his/her primary care physician. At each visit safety is assessed by clinical examination and adverse events, and the HAV is examined using duplex ultrasound to visualize the entire length to confirm patency, flow and to detect stenosis, aneurysm development or dilatation.

The primary objectives of the study are to evaluate the safety and tolerability of the Humacyte HAV in PAD patients undergoing above-knee femoro-popliteal bypass surgery and to determine the patency (primary, primary assisted and secondary) rate of the Humacyte HAV at 24 months. Secondary objectives include assessment of the panel reactive antibodies (PRA)) and IgG response to the HAV and to assess patency (primary, primary assisted and secondary) at 6, 12 and 18 months, to determine the rates of interventions needed to maintain / restore patency in the HAV, to assess any effect of implantation on claudication, rest pain and ischemic ulcers and to assess any effect on ankle-brachial index (ABI).

An interim analysis of the study was conducted in September 2015. At that time, the duration of follow up ranged from 14 to 22 months with a total of about 26 patient years of follow up.

The majority of the patients recruited into the study (65%) are male and all are Caucasian. The mean age is 66 years (range 54 to 79 years) and 45% have a history of diabetes while 50% have a history of atherosclerotic disease in the coronary and/or carotid circulation. Five patients (25%) are obese with a BMI >30Kg/M^2.

There has been one death during the study, which was considered to be unrelated to the HAV. This 68 year old male with a history of peripheral arterial disease (since 2013), diabetes, asthma, hypertension and ulcerative colitis, underwent implantation of an HAV as a left femoro-popliteal arterial bypass on 22 May 2014. The patient was discharged from hospital in good condition and returned for the 15 day study visit on 9 June. At this visit the patient was in good general health with no fever or signs of systemic infection. Inspection of the surgical wound revealed a local superficial infection at the lower end of the surgical incision which did not appear to extend down to the HAV. The HAV was patent on ultrasound examination with good flow. At this visit the patient had a white cell count of 12.56G/l (normal range 4-10). The patient refused immediate hospital admission for local wound management and for control of a very elevated blood glucose (value unknown) for personal reasons but agreed to return to the hospital within the next few days for wound cleansing and VAC therapy. The following day (10 June 2014) the patient was found dead at home. The death was reported to the investigator by the patient's sister on 7 July 2014. The cause of death was reported as cardiorespiratory collapse by the patient's primary care physician, but no autopsy was done.

Seven patients have experienced adverse events, which have affected HAV patency. Two of these patients were found to have occluded HAVs at routine study visits (at 12 and 26 weeks respectively). Non-compliance with dual antiplatelet therapy may have contributed to one of

these occlusions. Because these patients did not report significant symptoms associated with the HAV occlusions they have been managed conservatively and the HAVs are considered to have lost both primary and secondary patency.

Five other patients experienced serious adverse events (SAEs) involving loss of primary patency of the HAV (4 thromboses and 1 pseudoaneurysm). In four of these cases (three thrombosis and one pseudoaneurysm) the patient was managed surgically or endovascularly and the HAVs still have secondary patency. The treated thromboses occurred at 20, 26, and 48 weeks after implantation and in each case a stenosis of the HAV or the HAV / artery anastomosis was identified and treated by angioplasty. The pseudoaneurysm was identified at the 12 week visit and was managed by replacement of a short section of the HAV with an ePTFE graft. Histology of the resected HAV did not show any evidence of inflammation or infection and the hole in the HAV wall may have resulted from iatrogenic damage at a previous thrombectomy attempt with a Fogerty catheter. At that thrombectomy attempt – performed during surgical management of a post-operative hematoma at the site of an iliac endarterectomy -no thrombus was found in the HAV.. In the fifth case the HAV was found to be occluded at the 12 month visit and the HAV was abandoned and replaced with a Vascutek graft.

Kaplan-Meier analysis of patency indicates a primary patency of 74% at 6 months and 63% at 12 months. Secondary patency is 89% at both 6 and 12 months. These patency rates are comparable to those reported with synthetic grafts used for femoro-popliteal bypass.

2.4.3 Vascular Access for Dialysis

The HAV is currently under evaluation as a vascular access for dialysis in two clinical studies in patients with end-stage renal disease (ESRD), one in Poland and one in the United States (US). These pilot studies are of very similar design, recruiting patients with ESRD who are not suitable for creation of an arteriovenous fistula (AVF), usually because of unsuitable vasculature or failure of previous attempts to create an AVF. The HAV is placed as an upper arm conduit. Patients are followed up with monthly visits and ultrasound surveillance of the HAV for the first 6 months and then less frequently out to two years. Provided that the HAV is functioning well and the surgical wounds have healed, the HAV can be used for dialysis from 4 - 8 weeks after implantation. Dialysis is initiated with small needles (as for a new AVF) with gradual increase in needle size over the first few weeks of use. The primary objective of the studies is to evaluate

safety of the HAV and to assess patency at 6 months. Secondary objectives include measurement of any panel reactive antibody (PRA) response and evaluation of patency and need for interventions to maintain or restore that patency over 2 years.

The US study is being performed under IND 15,263 and was initiated in June 2013. Recruitment started in June 2013 and was completed in June 2014 with 20 patients implanted.

The Polish study was initiated in December 2012 and the final (40^{th}) patient was implanted in April 2014

An interim analysis of the combined data set of 60 patients was performed in May 2015. A total of 60 patients (40 in Poland and 20 in the US) had been implanted with HAVs for dialysis access. Baseline characteristics of the two study populations were similar though the proportion of female patients was higher in the US (65% as compared to 45% in Poland), and the incidence of hypertension, diabetes, and cardiovascular disease was higher in the US population. In the US, 65% of patients were African-American, while all of the patients in Poland were Caucasian. In both studies, patients had multiple prior accesses for hemodialysis, ranging from 1-9 in the Poland, and from 1-6 in the US.

There have been 6 deaths in the two studies, three during ongoing follow up, one of a patient who withdrew from the study having decided to discontinue dialysis and two within 1 month after HAV abandonment. None of the deaths were considered related to the HAV and the death rate during the study is comparable with that observed generally in the ESRD population. One patient underwent a renal transplant during the study and the renal allograft is functioning well 12 months after transplantation.

The HAV rates of primary, primary assisted, and secondary patency at 6 months were 63%, 72%, and 97%, respectively. At 12 months, patency rates were 28%, 38%, and 89%, respectively. While primary and primary assisted patency rates are comparable to those reported for ePTFE in recent randomized trials, secondary patency of 97% at 6 months and 89% at one year is substantially greater than prior reported values, which range from 55-65%. (CDRH 510K Gore Acuseal, 2013, Dixon, 2009) Mean blood flow rates in the HAVs as measured by ultrasound were typically over 1.0 L/min and did not tend to decrease over time. Mean diameters of HAVs, as measured by ultrasound, remained overall stable, without

evidence of substantial dilatation or narrowing. Dialysis centers reported that the HAVs were straightforward to cannulate using standard techniques.

Three access-associated infections have been reported. In only one case was the HAV material demonstrated to be infected and required partial resection. The remaining cases involved an infected perigraft hematoma and an infected ePTFE jump graft inserted during revision of the venous anastomosis after thrombosis of the graft.

Overall, HAVs have been abandoned in ten patients. Two HAV abandonments were associated with over-dilation with 8-mm high-pressure balloons, which disrupted the 6-mm diameter HAV material. In one case, the HAV was ligated and resected to manage a suspected case of ischemic monomelic neuropathy. In the other seven instances, the HAVs were abandoned due to stenosis in the run-off vein or the venous anastomosis, or due to recurrent thromboses. None of the HAVs were abandoned due to infection.

Biochemical analysis of the HAV indicates that it contains trace or non-detectable levels of cellular proteins such as beta-actin and human leukocyte antigen I and is thus potentially suitable for use in any patient without the need for tissue matching. To confirm this lack of immunogenicity blood samples for measurement of PRA are being collected from all patients in the pilot studies. At six months after implantation of the HAV only three patients showed any increase in PRA values. All three patients had histories of previous failed renal transplants. In two cases the rise in PRA occurred shortly after discontinuation of immunosuppressive therapy. In the third case the patient was an infirm elderly lady with multiple medical problems and generalized debility. In none of these patients was there evidence of an inflammatory response to the HAV or any complication with the HAV coinciding with the PRA increases.

2.4.4 Conclusions

In conclusion, the early clinical experience with the HAV indicates that it remains mechanically strong over implantation periods of more than 30 months with no evidence of dilatation. In more than 90 patient years of follow up across the three pilot studies (ESRD and PAD) only one case of infection of the HAV material itself has been reported. In this ESRD case methicillin-resistant Staphylococcus aureus was isolated from a segment of resected HAV. In two further ESRD patients infection associated with the access has been reported but did not appear to involve the HAV - in one patient a perigraft hematoma became infected but the graft was salvaged with use of parenteral antibiotics and in the second the infection involved an ePTFE jump graft but not the remaining HAV. This suggests that the HAV may offer improved resistance to infection

compared to currently available ePTFE grafts. The SAE profile has been typical of that expected in the dialysis and peripheral arterial disease populations. Patency of the HAVs is comparable with that observed historically with synthetic grafts and the secondary patency is substantially higher than historical data for ePTFE. No evidence of immunogenicity of the HAV has been found and the HAV remains mechanically robust even after repeated puncture for hemodialysis.

These data support the use of the HAV in this proposed phase 2 study in peripheral vascular trauma.

2.5 Potential Risks and Benefits

2.5.1 Potential Risks

It is anticipated that subjects participating in the study will be exposed to similar risks to those associated with other arterial conduits. Risks associated with the study investigational product may include but are not limited to:

- Thrombosis/occlusion of the conduit or host vessels, with consequent limb ischemia
- Embolism from a thrombosed conduit
- Bleeding and hematoma formation at the surgical site
- Infection – at the surgical site or systemic
- Stenosis of the conduit or its anastomoses
- Aneurysm or pseudoaneurysm formation
- Swelling of the limb

Regular clinical examination of the HAV implantation site and assessment of the patency, blood flow and diameter using ultrasound during the study should allow early detection of complications and permit appropriate intervention including HAV explantation.

The HAV is grown using donor human aortic smooth muscle cells. The graft is decellularized during manufacturing and thus consists of human extracellular matrix proteins. It is possible that the HAV may provoke an immune response which may lead to damage to the HAV and possible cross reactivity against host proteins.

2.5.2 Potential Benefits

Patients who undergo implantation of the Humacyte HAV may benefit from improved patency resulting in a reduced number of interventions versus a conventional expanded PTFE (ePTFE) or a Dacron graft. This may result from a decreased propensity for anastomotic and downstream neointimal hyperplasia, which often leads to graft occlusion with synthetic grafts. In addition, the risks such as infection listed in section 2.6.1 typically encountered with conventional synthetic grafts may be decreased with the Humacyte HAV. Finally, the longevity of the Humacyte HAV may be greater than that of conventional synthetic grafts.

2.5.3 Risk-Benefit Rationale

The risks anticipated in this study are similar to those associated with currently marketed prosthetic grafts used for peripheral vascular repair. The potential advantages of the HAV compared to currently marketed grafts may lead to a lower complication rate and reduced need for surgical intervention and graft replacement.

Recruitment will be restricted to a maximum of 20 subjects who receive implants to provide safety data prior to the planning and initiation of further studies

There is no formal hypothesis testing in this study but data from the study will be compared with historical data on synthetic peripheral bypass grafts to assess the safety and efficacy of the HAV prior to initiation of larger studies.

3. STUDY OBJECTIVES

3.1 Primary Objectives

This is an open label phase 2 study. There is no formal hypothesis testing.

Safety:

- To evaluate the safety and tolerability of the Humacyte HAV in patients with vascular trauma in the lower limb undergoing vascular reconstructive surgery

Efficacy:

- To determine the patency (primary, primary assisted and secondary) rate of the Humacyte HAV at 12 months
- To determine the rate of limb salvage

3.2 Secondary Objectives

Safety:

- To determine mechanical stability of the HAV based on freedom from aneurysmal degeneration on duplex ultrasound
- To determine HAV durability in terms of freedom from need for graft explantation or replacement due to infection, bleeding, or conduit degeneration

Efficacy:

- To determine the patency of the vessel (primary, primary assisted and secondary) at 3, 6 and 9 months
- To determine the rates of interventions needed to maintain / restore patency in the vessel over 12 months

4. STUDY DESIGN

4.1 Description of the Study Design

Prospective, multicenter, single arm, non-randomized phase 2 study

4.2 Study Endpoints

All endpoints will be evaluated at multiple time points over 12 months after HAV implantation. However, the main analysis of the study will be based on the 12-month follow up of all patients who receive a Humacyte HAV.

4.2.1 Endpoints

Safety:

- Incidence of aneurysm formation, anastomotic bleeding or rupture, HAV infection, HAV removal and irritation/inflammation at the implantation site
- Frequency and severity of adverse events

Efficacy:

- Incidence of limb salvage
- Primary patency rate
- Primary assisted patency rate
- Secondary patency rate
- HAV interventions

4.2.2 Long Term Endpoints

– to be evaluated at “standard of care” routine clinical visits or by telephone contact with the patient and/or physician

- Limb viability

- HAV survival
- Patient survival

4.3 Duration of Study Participation

For an individual subject, the expected duration of study participation is approximately 13 months. Enrollment (accrual) is expected to occur over 12 months. Additional data on patient and HAV status will be collected at routine clinical visits (standard of care for peripheral vascular surgical patients) at 6 month intervals up until 36 months after HAV implantation but there are no study-specific visits or procedures after 12 months. If the patient is not scheduled to return to the investigational site for these visits (e.g. for geographical reasons) then the post 12 month follow up may be conducted by telephone contact with the patient and/or the physician designated to continue long term care of the patient.

5. STUDY POPULATION

5.1 Description of the Study Population

The study population will consist of patients with limb-threatening damage to the superficial femoral or popliteal artery who require surgical repair with an interposition graft.

5.1.1 Patient Inclusion Criteria

1. Patients with lower limb vascular trauma which threatens the viability of the leg and who require reconstruction of the superficial femoral or popliteal artery
2. Preoperative ultrasound or angiography or CT angiography or clinical examination indicates damage to the SFA or popliteal artery requiring reconstruction with interposition of a vascular graft AND required graft length of ≤ 40cm.
3. Proximal graft anastomosis is expected to be to the common femoral artery below the inguinal ligament or to the superficial femoral artery or popliteal artery
4. Distal anastomosis is expected to be to the SFA, popliteal artery or the tibio-peroneal trunk
5. Autologous vein graft is either not feasible in the judgment of the treating surgeon (e.g. because of lack of availability of suitable conduit, presence of severe venous insufficiency) or is not desirable because of the urgency of revascularization
6. Aged 18 to 85 years old, inclusive
7. Able to communicate meaningfully with investigative staff, and able to comply with entire study procedures. If the patient is unconscious then information from a reliable witness indicates that the patient would normally be able to comply with study procedures
8. Patient or relative is able, willing and competent to give informed consent
9. Life expectancy of at least 1 year

5.1.2 Patient Exclusion Criteria

1. Limb at high risk of amputation despite vascular reconstruction e.g., because of crush injury
2. Life threatening head, chest, or abdominal injuries that make survival unlikely
3. Distal anastomosis planned to a tibial or pedal artery
4. Pregnant women
5. Known medical condition which would preclude long term dual antiplatelet therapy after resolution of acute injuries

6. Any other condition which in the judgment of the investigator would preclude adequate evaluation of the safety and efficacy of the Humacyte Human Acellular Vessel (HAV)
7. Previous enrollment in this study
8. Employees of the sponsor or patients who are employees or relatives of the investigator

6. INVESTIGATIONAL MEDICINAL PRODUCT

Additional information on the manufacturing process and testing of the IMP is provided in the Investigator Brochure.

6.1 Product Description

The investigational medicinal product (IMP) is a Humacyte Human Acellular Vessel (HAV), which is a tissue-engineered vascular prosthesis for arterial bypass or reconstruction in patients with peripheral arterial disease or peripheral arterial trauma. It is a sterile, non-pyrogenic acellular tubular vessel composed of human collagen types I and III and other extracellular matrix proteins, including fibronectin and vitronectin. The vessel is 6 mm in diameter and approximately 42 cm in length. The product is supplied on a silicone mandrel immersed in sterile phosphate buffered saline in a sealed and labeled plastic container.

There is no placebo or comparator control group in this study.

6.2 Manufacturer of the IMP

The HAV is manufactured by:

AlloSource
6278 S. Troy Circle
Centennial, CO 80111 USA

Traceability of the HAV during and after the clinical investigation will be achieved by the assignment of lot numbers. A unique identifying lot number will be assigned to each vessel.

6.3 Packaging, Storage, and Labeling

Packaging: Each HAV is contained in a sealed, flexible plastic primary container closure system that was developed by Humacyte. The system meets container/closure requirements to

maintain sterility as well as product and fluid integrity. The vessel is contained inside the system in a fixed manner, immersed in a sterile, phosphate buffered saline. The total volume of the storage solution is approximately 300 mL.

Storage: The product is shipped under controlled conditions to maintain temperature at 4°C (range: 2 – 10°C). The product should be stored in a refrigerator that maintains this temperature range. The HAV MUST NOT be allowed to freeze.

Labeling: The IMP will be labeled according to applicable guidelines and relevant regulatory agency requirements. A tamper resistant label affixed to the secondary container will be used to ensure that the product is not compromised prior to use.

6.4 Implantation of the Humacyte Human Acellular Vessel (HAV)

The Humacyte HAV is implanted using standard vascular surgical techniques similar to placement of predicate peripheral vascular prostheses (see study manual for details).

Tunneling of the graft, if needed, must be performed using a sheathed tunneler. During tunneling, the graft should be handled by pulling on the Dacron cuff (see study manual for details).

After placement, graft patency and integrity are checked by pressurizing the graft. Prior to completion of surgery angiography is performed to confirm adequacy of the graft anastomoses, HAV patency and peripheral runoff. The surgical site is closed using standard techniques.

Implantation of the HAV will be undertaken by qualified vascular surgeons experienced in peripheral vascular surgery.

6.5 IMP Accountability Procedures

Documentation of receipt, dispensing, and return of all IMP must be maintained by the Principal Investigator or his/her designee. It is the Principal Investigator's responsibility to ensure that all IMPs are kept in a secure location, with access limited to individuals authorized by the Investigator. The product will be shipped with the IMP Receipt Form. Once signed, the original IMP Form should be returned to Humacyte, and a copy will be maintained in the Investigator's Files. The IMP Accountability Log will be used to account for all IMP received, dispensed, and

returned and must be maintained by the site until the conclusion of the study, at which time the original will be retrieved by Humacyte or their authorized designee and a copy kept at the site. Following accountability of the IMP by Humacyte or their authorized designee, all unused IMP will be returned to Humacyte.

6.6 Assessment of Patient Compliance with IMP

Not applicable.

6.7 Prior and Concomitant Medications

Prior medications are defined as all medications taken within 7 days (whether continuing or not) prior to Day 1. All prior and concomitant medications (including immediately pre-surgery and post-surgery medications) must be listed in the patient's medical record and recorded on the eCRF. Drugs used during anesthesia should be recorded in the anesthesia records but should not be transcribed into the eCRF. Patients should be questioned at each study visit concerning any new medications or changes in current medications. **Note: particular attention should be made to identify the use of antithrombotic or antiplatelet agents (e.g., aspirin, clopidogrel, prasugrel, direct thrombin inhibitors, factor Xa inhibitors, or vitamin K antagonists).**

For each medication taken, the following information will be collected:

- Medication generic name / components of combination product
- Dose
- Route of administration
- Frequency of administration
- Date started
- Date stopped
- Indication for use

6.8 Essential, Precautionary and Prohibited Medications

6.8.1 Essential Medications

All patients should receive both antibiotic and antithrombotic prophylaxis in conjunction with graft implantation:

Antibiotic prophylaxis:

- All patients must have at least 1 day of antibiotic prophylaxis in accordance with local hospital guidelines. Longer antibiotic prophylaxis is at the discretion of the investigator.

Antithrombotic prophylaxis:

- Intraoperative heparin: up to 150 IU / kg unfractionated heparin during surgery.
- Further measures to prevent venous thromboembolism are at the discretion of the investigator and may include LMWH.
- If antiplatelet therapy was not ongoing at the time of surgery it should be commenced as soon as possible post operatively. Antiplatelet therapy (ideally dual therapy with aspirin 75-325 mg and clopidogrel 75 mg daily) should continue long term while the graft is in place. If the patient is unable to tolerate aspiring and/or clopidogrel the choice of antiplatelet regimen is at the investigator's discretion.

6.8.2 Restricted Medications

Vitamin K antagonists, antiplatelet agents other than aspirin and clopidogrel, direct thrombin inhibitors and factor Xa inhibitors (e.g., dabigatran, apixaban and rivaroxaban) should be avoided unless essential for treatment of a medical condition arising postoperatively. In that case consideration should be given to modification or cessation of antiplatelet therapy. Antiplatelet therapy should be restarted on cessation of these anticoagulant drugs.

7. STUDY PROCEDURES / EVALUATIONS

7.1 Clinical Evaluations

- Medical History – pre-operatively, from patient / legal representative interview and medical records covering relevant past medical history.
- Smoking history
- Medication History – prescription and non-prescription medication from Day -7 onwards (see 6.5 above). Particular attention should be paid to the identification of OTC medications containing aspirin.
- Physical Exam – full exam (as far as possible) at screening, 12 month visit or final study visit for early termination (ET). Clinical examination of the operative limb and graft at all post-operative visits; physical exam for lymphadenopathy; additional clinical exam as needed to evaluate adverse events
- Vital signs (heart rate and blood pressure) – at screening, D1, D5, D29 and M3, M6, M9 and M12 or ET
- Adverse events – at all post-operative visits, the patient will be asked a general question about his/her health and for any graft problems since the previous visit
- Intraoperative angiography to assess anastomotic anatomy, patency and runoff.
- Duplex ultrasound – clinical assessment at all postoperative visits from day 29 onwards, to assess HAV patency, mid HAV diameter and flow rate. The full length of the HAV should be imaged at each assessment to monitor for aneurysm development
- Documentation of surgical procedure and any complications immediately postoperatively
- Capture of data on patient status, HAV patency and any intervention or complications at routine “standard of care” 6 monthly clinic visits from 18 to 36 months post implantation or by telephone contact with the patient or the patient’s physician

7.2 Laboratory Evaluations

7.2.1 Clinical and Research Laboratory Evaluations and Specimen Collection

The following parameters will be measured wherever possible at screening and all should be measured at Day 5

- Hematology: hemoglobin, hematocrit, RBC, white blood cells (WBC) with differential, platelet count
- Clinical chemistry: sodium, potassium, calcium, blood urea nitrogen, creatinine, albumin, total bilirubin, alanine aminotransferase (ALT), aspartate aminotransferase (AST), gamma glutamyl transferase, alkaline phosphatase, glucose (non-fasting)

7.2.2 Specimen Preparation, Handling and Shipping

Biochemistry and Hematology Tubes – transport to the appropriate local clinical laboratory according to institutional procedures.

7.2.3 Monitoring, maintenance and calibration of equipment

All laboratory tests will be conducted at certified hospital laboratories. Routine monitoring, maintenance or calibration of laboratory equipment is required per local site procedures.

7.3 Imaging Evaluations

7.3.1 Duplex Ultrasound

Clinical duplex ultrasound examinations will be performed at 1, 3, 6, 9 and 12 months and follow standard bypass graft imaging protocols, including B-mode, power duplex and color duplex ultrasound imaging of the graft with velocity spectral waveform analysis. The purpose of this clinical duplex ultrasound surveillance is to detect HAV stenosis and aneurysm development.

7.4 Study Schedule

7.4.1 Pre-operative Screening (Day 1)

Potential study participants who are being considered for surgical repair of the superficial femoral or popliteal artery will be informed about the study and invited to participate. After explanation of the potential risks and benefits of the HAV and of the study procedures, written informed consent will be obtained. No study specific procedures may be performed prior to

patient consent. If the patient is unable to give informed consent then this may be sought from the patient's legal representative, usually a close relative.

The following assessments will be performed, as far as possible, prior to surgery (Day 1):

- Informed consent
- Medical history
- Prior and concomitant medication
- Full physical examination
- Evaluation of inclusion/exclusion criteria
- Reasons for not using an autologous venous conduit
- Vital signs (blood pressure, heart rate, temperature – axillary or tympanic)
- Laboratory testing (or standard pre-op lab profile for the institution)
  - Hematology: full blood count and differential
  - Clinical chemistry ; sodium, potassium, calcium, blood urea nitrogen, creatinine, albumin, total bilirubin, alkaline phosphatase,, AST and ALT, glucose (non-fasting)

7.4.2 Enrollment – Day 1 (HAV Implantation)

The HAV will be implanted as an interposition graft in the superficial femoral or popliteal artery using standard vascular surgical techniques. Details of the surgical anatomy and any complications will be documented.

7.4.3 Follow-up Visits

Day 5 (or prior to hospital discharge if earlier)

- Concomitant Medication
- Physical exam including surgical site (and graft patency) and to evaluate any AEs
- Vital signs
- Documentation of any HAV interventions
- Adverse events
- Laboratory assessments (clinical chemistry, hematology)

Day 29 (+/- 4 days)

- Concomitant medication
- Physical exam including surgical site (and HAV patency), and to evaluate any AEs
- Clinical duplex ultrasound of the HAV
- Documentation of interventions
- Adverse events
- Vital signs

Months 3, 6 and 9 (+/- 14 days)

- Concomitant Medication
- Physical exam including surgical site (and HAV patency) and to evaluate any AEs
- Clinical duplex ultrasound of the HAV
- Documentation of interventions
- Adverse events

Month 12 (+/-14 days) and Early Termination

- Concomitant Medication
- Documentation of interventions
- Adverse events
- Clinical duplex ultrasound of the HAV
- Full physical exam including surgical site (and HAV patency) and to evaluate any AEs

7.4.4 Final Study Visit

The final study visit is at 12 months.

7.4.5 Early Termination Visit

The patient may withdraw from the study at any time at his/her own or his/her physician's discretion. If withdrawal occurs before 12 months the patient will be asked to complete an early termination visit at which all assessments normally performed at 12 months will be completed.

The reasons for early termination should be recorded in the eCRF.

The patient should be withdrawn from the study prior to 12 months if the HAV is removed or becomes permanently occluded (loss of secondary patency).

7.4.6 Unscheduled Visits

If necessary to evaluate adverse events or HAV complications additional visits may be scheduled at the discretion of the investigator. At a minimum HAV status on clinical examination and Duplex ultrasound and adverse events will be recorded.

If, at any of the scheduled visits, duplex ultrasound surveillance suggests the development of a ≥ 50% stenosis within the HAV the patient should be asked to return for an additional visit 6 weeks later for a repeat duplex ultrasound study. Intervention to manage any such stenosis is at the discretion of the investigator taking into account the degree and rate of progression of the stenosis.

7.4.7 Follow up from Months 18-36 (+/- 30 days)

The final study visit is at Month 12. Thereafter the status of the patient and HAV will be ascertained every 6 months until 3 years after graft implantation at routine 6 monthly clinical visits or by telephone contact with the patient and his physician. The investigator will complete a brief questionnaire covering the status of the patient, known patency of the HAV and any complication, interventions or other vascular procedures on the operative leg. There are no study specific visits or procedures after 12 months.

7.5 Medical Care during the Study and upon Study Termination

Optimal medical therapy should be continued during the study. This should include:

- Dual antiplatelet therapy (see section 6.8.1)

After the final study visit at month 12 patients will not receive any further study-specific treatment. They will be treated by their medical doctor in a way that is appropriate for them.

7.6 Histological Examination of Resected HAV Material

If all or part of the HAV is resected it should, wherever possible, be retained for future histological examination. Instructions for preservation, storage and shipping of this material will be provided separately in a procedures manual. If a patient dies with an HAV in situ and it is feasible to obtain a fresh post mortem sample of the graft this should be attempted in accordance with local regulations.

8. ASSESSMENT OF SAFETY

Safety of the HAV will be assessed in terms of:

- Aneurysm formation
- Anastomotic bleeding or rupture
- HAV infection
- Need for HAV removal
- Irritation/Inflammation at the implantation site
- Other adverse events
- Laboratory parameters (clinical chemistry, hematology)

8.1 Adverse Event Definition

An AE is any untoward medical occurrence in a patient administered an IMP and which does not necessarily have a causal relationship with the IMP. An AE can, therefore, be any unfavorable and unintended sign, symptom, or disease temporally associated with the use of an IMP, whether or not related to the IMP. Any worsening of the patient's disease under study or other medical conditions will also be considered to be an AE, unless it is within the normal range of disease fluctuation for that patient.

8.2 Reporting of Adverse Events

At each evaluation, the investigator will determine whether any AEs have occurred. The patient will be questioned in a general way and no specific symptoms will be suggested. If any AEs have occurred they should be documented in the patient's medical chart and recorded on the AE pages of the eCRF. If known, the diagnosis should be recorded in preference to the listing of individual signs and symptoms. All SAEs (see Section 9), whether or not considered to be related to study treatment, should be reported to the study pharmacovigilance officer within 24 hours of identification.

AE reporting begins from implantation of the HAV and ends at the conclusion of the post-treatment follow-up period (i.e., Month 12 visit) unless an unresolved AE is still being followed (covered in further detail in Section 8.3).

Adverse events will be graded as follows:

- Grade 1 (Mild): Events require minimal or no treatment and do not interfere with the patient's daily activities.

- Grade 2 (Moderate): Events result in a low level of inconvenience or concern with the therapeutic measures. Moderate events may cause some interference with functioning.

- Grade 3 (Severe): Events interrupt a patient's usual daily activity and may require systemic drug therapy or other treatment. Severe events are usually incapacitating.

- Grade 4 (Life threatening): Events posing an immediate risk to the patient's life and requiring urgent intervention

- Grade 5 (Fatal): Events resulting in death

Relationship assessment of AEs/SAEs to the IMP and to the procedure should be made by the investigator. (NOTE: Relationship assessment is not a factor in determining what is or is not reported in the study.)

- Definitely Related: There is clear evidence to suggest a causal relationship, and other possible contributing factors can be ruled out. The clinical event, including an abnormal laboratory test result, occurs in a plausible time relationship to placement of the IMP and cannot be explained by concurrent disease or other devices, drugs, or chemicals.

- Possibly Related: There is some evidence to suggest a causal relationship (e.g., the event occurred within a reasonable time after the placement of the IMP). However, the influence of other factors may have contributed to the event (e.g., the patient's clinical condition, other concomitant events). Although an adverse event may rate only as "possible" soon after discovery, it can be flagged as requiring more information and later be upgraded to probable or certain as appropriate.

- Not Related: The AE is, in the opinion of the investigator, independent of the placement of the investigational product, and/or evidence exists that the event is definitely related to another etiology. There must be an alternative, definitive etiology documented by the clinician.

If sponsor and reporting principal investigator disagree on the seriousness of an event or on its relationship to the IMP, both opinions will be communicated to the relevant authorities and involved principal investigators.

8.3 Reporting of Pregnancy

If a study participant becomes pregnant during study participation, basic information about the pregnancy will be recorded in the Pregnancy eCRF and submitted to the Safety CRO. If there are complications during the pregnancy, the complications are recorded as AEs. The participant will be asked to report the outcome of the pregnancy and the site should submit the information to the Safety CRO within 30 days after the outcome of the pregnancy. If there is a congenital anomaly in the infant, this will be recorded as a SAE in the data forms for the mother (i.e., the study participant).

Partner pregnancies do not need to be reported.

8.4 Follow-Up of Adverse Events

If any SAEs are present when a patient completes the study or is withdrawn from the study, the patient will be re-evaluated within an appropriate period of time. If the SAE has still not resolved, additional follow-up will be performed as appropriate. Every effort should be made by the investigator or delegate to contact the patient until the SAE has resolved or stabilized or the medical monitor and investigator agree that further follow-up is not necessary. This should be documented in the patient's medical records.

9. SERIOUS ADVERSE EVENTS

A SAE is any untoward medical occurrence that at any dose:

- Results in death
- Is life-threatening
- Requires patient hospitalization or prolongation of existing hospitalization
- Results in persistent or significant disability/incapacity
- Is a congenital anomaly/birth defect

Medical and scientific judgment should be exercised in deciding whether expedited reporting to the sponsor is appropriate in other situations, such as important medical events that may not be immediately life-threatening or result in death or hospitalization but may jeopardize the patient or may require intervention to prevent one of the other outcomes listed in the definition above. These should also be considered to be SAEs.

9.1 Life-Threatening Adverse Event

The term “life-threatening” in the definition of “serious” refers to an event in which the patient was at risk of death at the time of the event; it does not refer to an event, which, hypothetically, might have caused death if it were more severe.

9.2 Hospitalization

This is defined as the patient being hospitalized overnight or the patient’s hospital stay being prolonged for at least an additional overnight stay.

9.3 Unexpected Adverse Drug Reaction

An unexpected adverse drug reaction is defined as an adverse drug reaction, the nature or severity of which is not consistent with Investigator’s Brochure for the IMP.

9.4 Reporting Serious Adverse Events

The urgency for reporting SAEs is 4-fold: (1) to facilitate discussion (and implementation, if necessary) by the sponsor and the Investigator of appropriate follow-up measures, (2) to facilitate Investigator reporting of unanticipated problems involving risk to human subjects to the institutional review board (IRB), (3) to facilitate the sponsor’s rapid dissemination of information regarding AEs to other Investigators/sites in a multi-center study, and (4) to enable the sponsor to fulfill the reporting requirements to the appropriate regulatory authority.

Any SAE that occurs during the course of this study from the time of HAV implantation until the 12 month follow up, whether or not causally related to the IMP, must be reported by the investigator or designee to the Safety CRO within 24 hours of learning of its occurrence. This applies also to any AE that could affect the safety of the study participants or the conduct of the study. SAEs which involve the HAV (including thrombosis, aneurysm formation or infection) which are reported to the investigator during the long term follow up period should also be reported to the Safety CRO within 24 hours.

Information about an SAE will be collected and recorded on the SAE Report Form. The investigator must assess the relationship to the investigational product and any relevant procedure.

The investigators complete the SAE Report Form in English, and **send the completed, signed form by fax or email (see below) IMMEDIATELY (at latest within 24 hours) after becoming aware of the SAE.**

Copies of relevant medical records (e.g., admission and/or discharge summary, laboratory reports and autopsy report), may also be submitted with the SAE form to clarify the circumstances surrounding the SAE(s). The entire medical records shout **NOT** be sent with the SAE form.

Atlantic Research Group, Inc

Drug Safety Department

Fax: +1-866-851-9318

Email: Safety@atlanticresearchgroup.com

Phone: +1-888-619-3216

The investigator will be requested to supply as much detailed information as possible regarding the SAE that is available at the time of the initial contact. The investigator should also complete missing or requested information and submit follow-up reports until the SAE has resolved or, in the case of permanent impairment, until the SAE has stabilized.

It is the responsibility of each Investigator to promptly notify his/her IRB of all SAEs that are received by the Sponsor or designee and that occur at his/her institution in accordance with institutional practices.

The Safety CRO will inform the sponsor about all SAEs within 1 business day after receipt of the respective report from the investigator.

9.5 Medically Significant Events

Any adverse event indicating a clinically significant impairment of HAV function or involving an intervention on the HAV, **even if it does not meet any criteria for an SAE**, should be reported to the pharmacovigilance officer as described in section 9.4 above.

9.6 Data Safety Monitoring Board

A DSMB will be convened by the Sponsor to review data from a randomized phase 3 study using the HAV. This DSMB will also have safety oversight of this study and any other ongoing phase 2 studies using the HAV.

9.7 Interim Analysis and Stopping Criteria

This is a phase 2 study with no formal interim analysis. Periodic reviews of safety data will be undertaken by the sponsor (and DSMB) with particular attention to events that might indicate structural failure of the HAV. Events that might have implications for already implanted HAVs and their possible removal - such as aneurysms or rupture -would trigger an urgent review of the safety data and in the interim no new patients would be implanted. Any serious deep space infection necessitating surgical resection of the HAV would also trigger a suspension of recruitment pending review.

10. STATISTICAL CONSIDERATIONS

This is a prospective, open label, single treatment arm, multicenter pilot study to evaluate the safety and efficacy of the HAV in patients undergoing peripheral vascular repair. The primary objective of this study is to evaluate the safety and tolerability of the HAV in these patients and to determine the patency of the Humacyte HAV and the rate of limb salvage at 12 months post-implantation. The secondary objectives of this study are to further assess safety in terms of adverse events, and laboratory parameters and to determine the rates of graft interventions required to keep the graft patent. There is no formal hypothesis testing planned; the study involves only a single, open-label treatment group.

Details of data handling and planned descriptive statistics are given in the Statistical Analysis Plan.

10.1 Analysis Population

All patients who receive an HAV will be included in the analyses. For discontinued or withdrawn patients, all available data will be included in the safety and efficacy analyses.

10.2 Safety Analyses

Safety analyses will be performed on all patients who have an HAV implanted.

The incidence of aneurysm formation, anastomotic bleeding or rupture, HAV removal, HAV infection, and irritation/inflammation at the implantation site will be tabulated by visit and overall.

Adverse events will be coded using Medical Dictionary for Regulatory Activities (MedDRA) terms. Adverse events will be listed and summarized by body system, incidence, severity, and duration. HAV complications will be listed in terms of incidence, severity, and (where

appropriate) time to onset and duration. Serious adverse events will be summarized separately. Any premature discontinuations due to adverse events and deaths will be listed and summarized.

Laboratory data, including PRA, will be listed and summarized using appropriate descriptive statistics for the absolute change from baseline values for all post-surgery visits. The closest non-missing values prior to surgery on Day 1 will be used as baseline values.

10.3 Efficacy Analyses

Primary, primary assisted, and secondary patency rates of the HAV at 12 months and at all other post-surgery visits with evaluation of patency will be described. The rate of limb salvage at 12 months at 12 months will also be described.

Primary patency is defined as the functional access patency until any type of intervention; primary assisted patency is defined as an HAV still working without thrombosis; secondary patency is defined as the functional HAV patency, with or without preceding successful interventional or surgical procedures to maintain or reestablish patency, until the HAV is abandoned. Early discontinued patients prior to the visit of interest will be determined as being non-patent irrespective of the reason for discontinuation.

The rate and type of interventions needed to maintain / restore patency in the graft will be descriptively tabulated.

The absolute change from baseline (Day 1) values to all post-surgery visits of duplex ultrasound parameters will be summarized. Summary statistics will also be provided at each time point.

The methods and endpoints regarding the efficacy parameters employed in this study are consistent with current clinical practice and are meaningful to the research community. Every attempt has been made to minimize the variability on the part of the surgeon when using this product. The results of this study will be used to determine the sample size of subsequent clinical studies.

10.4 Other Analyses

All clinical parameters will be listed for all patients treated at each study visit. Descriptive statistics will be summarized for continuous outcomes such as age and BMI. If necessary, number and percentage of patients will be reported for categorical outcomes.

10.5 Sample Size Rationale

Up to 20 patients will be recruited into the study. As this phase II study is the first study of the HAV in humans for vascular trauma, the number of patients was chosen in order to provide sufficient safety information on this graft to allow the initiation of further trials in larger numbers of patients.

The study is not powered to assess the efficacy of the HAV.

10.6 Interim analyses

There is no formal interim analysis.

11. STUDY MANAGEMENT AND DATA COLLECTION

11.1 Ethical Conduct of the Trial

This study will be conducted according to the protocol; 21 CFR Parts 11, 50, 54, 56, and 312; the World Medical Association Declaration of Helsinki (Appendix II) and Good Clinical Practice (GCP). Each Investigator will conduct the trial according to applicable local or regional regulatory requirements.

11.2 Institutional Review Board

IRBs must be constituted according to the applicable state and federal requirements, including ICH GCP.

It is the responsibility of each investigator to submit the protocol, Investigator's Brochure, subject informed consent, subject recruitment materials (if applicable), and other documentation as required by the IRB to his/her IRB for review and approval. A copy of the written approval must be provided to the contract research organization (CRO). The documentation should clearly mention the approval/favorable opinion of the protocol, the subject informed consent form, and subject recruitment materials (if applicable), including respective version dates. The written approval and a list of members, their titles or occupations, and their institutional affiliations must be obtained from the IRBs and provided to the CRO prior to the release of clinical study supplies to the investigational site and commencement of the study. If any member of the IRB has direct participation in this trial, written notification regarding his or her abstinence from voting must also be obtained.

Each investigator must adhere to all requirements stipulated by his/her respective IRB. This includes notification to the IRB regarding protocol amendments, updates to the subject informed consent, recruitment materials intended for viewing by subjects, investigational new drug safety

reports, SAEs and unexpected AEs, reports and updates regarding the ongoing review of the trial at intervals specified by the respective IRB, and submission of final study reports and summaries to the IRB.

11.3 Subject Informed Consent

Prior to any study procedures being performed, subjects and persons conducting the consent discussion will be required to sign and date the IRB-approved informed consent, and each subject will be given a copy. In addition, this information should be recorded in the subject's medical record (i.e., source document).

The written consent document will embody the elements of informed consent as described in the World Medical Association Declaration of Helsinki, 21 CFR Part 50.25, ICH E6 guideline (GCP), and in accordance with any local regulations. The investigator is responsible for the preparation, content, and IRB approval of the informed consent document. The consent form must be approved by the site's IRB and be acceptable to Humacyte.

The consent form must be written in a language fully comprehensible to the prospective subject. The investigator or designee shall give the subject adequate opportunity to read it before it is signed and dated. Information should be given in both oral and written form whenever possible and in the manner deemed appropriate by the IRB. Subjects must be given ample opportunity to inquire about details of the study.

11.4 Amendments to the Protocol

An amendment must be agreed to in writing by Humacyte and submitted to and approved by the respective Regulatory Authority and IRB before the amendment can be implemented. Written approval of a protocol amendment is not required prior to implementation of changes to the protocol which eliminate an immediate hazard to the study patient; however, approval must be obtained as soon as possible thereafter. Any agreed amendments must also be signed by the investigator.

11.5 Study Initiation

The investigator must not enroll any patients prior to attendance at the Investigator Meeting or the completion of a formal site initiation visit conducted by the CRO. These meetings will include a detailed review of the study protocol and eCRF pages. The investigator will not be supplied with IMP until all necessary pre-study requirements have been completed and essential signed documents provided to the CRO.

11.6 Study Monitoring

It is the responsibility of the investigator to ensure that the study is conducted in accordance with the protocol, GCP, applicable regulatory requirements, and the currently approved Declaration of Helsinki, and that valid data are entered into the eCRF.

To achieve this objective, the monitor's duties are to ensure the maintenance of complete, legible, well-organized, and easily retrievable data. The monitor will review the protocol with the investigator. In addition, the monitor will explain the investigator's reporting responsibilities and all applicable regulations concerning the clinical evaluation of the IMP.

The investigator will permit representatives of Humacyte and the CRO to monitor the study as frequently as Humacyte or the CRO deem necessary to determine that data recording and protocol adherence are satisfactory. The eCRF data and related source documents will be reviewed in detail by the monitor at each visit, in accordance with relevant SOPs and ICH GCP regulations. This includes results of tests performed as a requirement for participation in this study and any other medical records required to confirm information contained in the eCRF such as past medical history and secondary diagnoses. The investigator and his/her staff will be expected to cooperate with the monitor and provide any missing information whenever possible.

All monitoring activities will be reported and archived. In addition, monitoring visits will be documented at the investigational site by signature and date on the study-specific monitoring log.

11.7 Case Report Form

An electronic CRF will be used for this study. The data will be entered into the eCRF in a timely manner on an ongoing basis.

The investigator is responsible for ensuring that data are properly recorded on each patient's eCRF and related documents. An investigator who has signed the protocol signature page should personally sign the eCRFs in accordance with the procedure described in the eCRF completion guidelines to ensure that the observations and findings are correct and complete.

For data handled by the CRO, eCRF data and some or all of the study-related data will be managed and stored electronically in the CRO's database system. Validated data will subsequently be transferred to the sponsor.

11.8 Verification Procedures

It is the investigator's obligation to ensure documentation of all relevant data in the subject's medical record. The subject's medical record will be considered the source document. The eCRF should not be used as the source for study information.

The investigator will maintain a subject identification code list to enable unambiguous identification of the subjects (subject names and corresponding subject numbers). The subject identification code list is an essential document and as such should be maintained according to the ICH GCP guidelines.

11.9 Retention of Records

All documentation pertaining to the study will be kept by Humacyte or their designee in accordance with ICH guidelines and US FDA regulations.

The investigator will maintain a study file, which should be used to file the Investigator's Brochure, protocol, and IMP records; correspondence with the IRB and Humacyte; and other study-related documents.

The investigator agrees to keep records and those documents that include (but are not limited to) the identification of all participating subjects, medical records, study-specific source documents, source worksheets, all original signed and dated informed consent forms, query responses, and detailed records of drug disposition to enable evaluations or audits from regulatory authorities and Humacyte or its designees.

The investigator shall retain records required to be maintained under this part for a period of 2 years following the date a marketing application is approved for the IMP for the indication for which it is being investigated; or, if no application is to be filed or if the application is not approved for such indication, until 5 years after the investigation is discontinued. However, these documents should be retained for a longer period if required by the applicable regulatory requirement(s) or if needed by the sponsor. In addition, the investigator must make provision for the subject's medical records to be kept for the same period of time. No data should be destroyed without the agreement of Humacyte. Humacyte will inform the investigator in writing when the trial-related records are no longer needed. Subject's medical records and other original data will be archived in accordance with the archiving regulations or facilities of the study site.

11.10 Protocol Deviations

A protocol deviation is any noncompliance with the protocol or GCP requirements. The noncompliance may be either on the part of the participant, the investigator, or the study site

staff. As a result of deviations, corrective actions are to be developed by the site and implemented promptly. Although in principle protocol deviations are not permitted, under emergency circumstances, deviations may proceed without prior approval of the sponsor and the IRB to protect the rights, safety, and well-being of human subjects.

All protocol deviations will be documented and reported by the CRO during the course of the study in the Monitoring Reports. All deviations will be reported to the sponsor who will agree on the necessary actions to be taken.

If required per their guidelines, reports about protocol deviations must be reported to the local IRB.

11.11 Insurance and Indemnity

Insurance coverage for damages emerging from the study will be provided according to applicable legal requirements. During the informed consent procedure, the investigator must inform the patient accordingly.

11.12 Audit

It is the responsibility of CRO and Humacyte to perform auditing (if applicable) as part of implementing quality assurance. The purpose of an audit, which is independent of and separate from routine monitoring or quality control functions, is to evaluate trial conduct and compliance with the protocol, SOPs, GCPs, and the applicable regulatory requirements. The auditor and regulatory authorities will require authority from the investigator to have direct access to the subject's medical records.

12. REPORTING

Following completion of follow-up of all patients to the 12-month endpoint, the results will be evaluated by Humacyte or a designee for clinically meaningful findings. A clinical study report will be generated, including a summary of all available data, statistical measures, tabulated results, graphical results and interpretations. This report will be submitted to regulatory authorities in a timely manner. An addendum to the report will be generated to include data up to 36 months follow-up. This addendum will be submitted to regulatory authorities in a timely manner.

13. QUALITY CONTROL AND QUALITY ASSURANCE

Following written standard operating procedures, the monitors will verify that the clinical trial is conducted and data are generated, documented (recorded), and reported in compliance with

the protocol, GCP, and the applicable regulatory requirements. Reports of monitoring activities will be submitted to Humacyte in a timely manner.

The investigational site will provide direct access to all trial related areas, source data/documents, and reports for the purpose of monitoring and auditing by the sponsor, and inspection by local and regulatory authorities.

Quality control procedures will be implemented for data entry and the generation of data quality control checks and will be run on the database. Any missing data or data anomalies will be communicated to the site(s) for clarification/resolution.

14. RESPONSIBILITIES OF THE PRINCIPAL INVESTIGATOR

The role of the principal investigator is to implement and manage the day-to-day conduct of the clinical investigation as well as to ensure data integrity and the rights, safety, and well-being of the patients involved in the clinical investigation.

14.1 Informed Consent

The principal investigator shall ensure that the process for obtaining informed consent

- includes all aspects of the clinical investigation that are relevant to the patient's decision to participate throughout the clinical investigation,
- avoids any coercion or undue improper influence on, or inducement of, the patient to participate,
- does not waive or appear to waive the patient's legal rights,
- uses native non-technical language that is understandable to the patient,
- provides ample time for the patient to read and understand the informed consent form and to consider participation in the clinical investigation,
- provides the patient with a copy of the signed and dated informed consent form and any other written information.

The principal investigator shall ensure and document appropriate training if an authorized designee is appointed to conduct the informed consent process.

14.2 Compliance with the Protocol

The principal investigator shall:

- indicate his/her acceptance of the protocol in writing
- conduct the clinical investigation in compliance with the protocol
- create and maintain source documents throughout the clinical investigation and make them available as requested during monitoring visits or audits
- ensure that the IMP is used solely by authorized users, and in accordance with the protocol and instructions for use
- propose to the sponsor any appropriate modification(s) of the protocol
- refrain from implementing any modifications to the protocol without agreement from the sponsor, IRB, and, if required, regulatory authorities
- document and explain any deviation from the approved protocol that occurred during the course of the clinical investigation
- ensure that an adequate investigation site team and facilities exist and are maintained and documented during the clinical investigation
- ensure that maintenance and calibration of the equipment relevant for the assessment of the clinical investigation is appropriately performed and documented, where applicable
- ensure the accuracy, completeness, legibility, and timeliness of the data reported to the sponsor in the eCRFs and in all required reports
- maintain the clinical trial material accountability records
- allow and support the sponsor to perform monitoring and auditing activities
- be accessible to the monitor and respond to questions during monitoring visits
- allow and support regulatory authorities and the IRB when performing auditing activities
- ensure that all clinical-investigation-related records are retained as specified in this protocol.

14.3 Medical Care of Patients

The principal investigator shall:

- provide adequate medical care to a patient during and after a patient's participation in a clinical investigation in the case of AEs
- inform the patient of the nature and possible cause of any adverse events experienced
- inform the patient of any new significant findings occurring during the clinical investigation, including the need for additional medical care that may be required
- provide the patient with well-defined procedures for possible emergency situations related to the clinical investigation, and make the necessary arrangements for emergency treatment,
- ensure that clinical records are clearly marked to indicate that the patient is enrolled in a particular clinical investigation
- inform, with the patient's approval or when required by national regulations, the patient's personal physician about the patient's participation in the clinical investigation
- make all reasonable efforts to ascertain the reason(s) for a patient's premature withdrawal from the clinical investigation while fully respecting the patient's rights.

14.4 Safety Reporting

The principal investigator shall:

- record every adverse event together with an assessment, in accordance with Sections 8 and 9 of this protocol,
- report to the sponsor, without unjustified delay, all serious adverse events and medically significant events as specified in Sections 8 and 9 of this protocol,
- supply the sponsor, upon sponsor's request, with any additional information related to the safety reporting of a particular event.

15. SUSPENSION OR PREMATURE TERMINATION OF THE CLINICAL INVESTIGATION

The sponsor may suspend or prematurely terminate either a clinical investigation in an individual investigation site or the entire clinical investigation for significant and documented reasons.

A principal investigator, IRB, or regulatory authority may suspend or prematurely terminate participation in a clinical investigation at the investigation sites for which they are responsible.

If suspicion of an unacceptable risk to patients arises during the clinical investigation, or when so instructed by the IRB or regulatory authorities, the sponsor shall suspend the clinical investigation while the risk is assessed. The sponsor shall terminate the clinical investigation if an unacceptable risk is confirmed.

The sponsor shall consider terminating or suspending the participation of a particular investigation site or investigator in the clinical investigation if monitoring or auditing identifies serious or repeated deviations on the part of an investigator.

If suspension or premature termination occurs, the terminating party shall justify its decision in writing and promptly inform the other parties with whom they are in direct communication.

If, for any reason, the sponsor suspends or prematurely terminates the investigation at an individual investigation site, the sponsor shall inform the responsible regulatory authority if required and ensure that the IRB is notified. If the suspension or premature termination was in the interest of safety, the sponsor shall inform all other principal investigators.

If suspension or premature termination occurs,

1. the sponsor shall remain responsible for providing resources to fulfill the obligations from the protocol and existing agreements for following up the patients enrolled in the clinical investigation, and

2. the principal investigator or authorized designee shall promptly inform the enrolled patients at his/her investigation site, if appropriate.

In the event that the study is discontinued, the reasons for discontinuation will be explained to the investigators and may be disclosed to the study participants. Humacyte will provide all information needed by the investigator to ensure the safety and well-being of the study participants.

16. PUBLICATION POLICY

A Publication Committee comprising the Principal Investigator from each participating site and a representative of Humacyte will oversee all publication of data from this study. Prior to submitting for publication, presenting, using for instructional purposes, or otherwise disclosing the results of the study, the investigator agrees to allow the Publication Committee and Humacyte a period of at least 30 days (or, for abstracts, at least 5 calendar days) to review the proposed publication or disclosure prior to its submission for publication or other disclosure. Publications or disclosures of study results shall not include other confidential information belonging to Humacyte. If the proposed publication/disclosure risks Humacyte's ability to patent any invention related to the study, the publication or disclosure will be modified or delayed, at the investigator's option, a sufficient time to allow Humacyte to seek patent protection of the invention. For multicenter studies, the first publication or disclosure shall be a complete, joint multicenter publication or disclosure. This statement does not give Humacyte any editorial rights over the content of a publication or disclosure, other than to restrict the disclosure of Humacyte's confidential information. If a written contract for the conduct of the study is executed which includes publication provisions inconsistent with this statement, then that contract's publication provisions shall apply rather than this statement.

17. REFERENCES

Akingba G, Rapp BM, Tsai A, Jester AL, Fajardo A, Motaganahali R, et al. Surgical Management of Vascular Trauma From Dog Bites: The 10- year Experience of a Level I Trauma Center. Journal of Vascular Surgery. 2012;56(4):1190.

Andrikopoulos V, Antoniou I, Panoussis P. Arterial injuries associated with lower-extremity fractures. Cardiovascular Surgery. 1995;3(1):15-8.

Cordesman AH, Lemieux, J. IED Metrics for Afghanistan. January 2004-May 2010. Center for Strategic & International Studies, 2010.

Dahl, SL, Kypson, AP, Lawson, JH et al. Readily available tissue-engineered vascular grafts. Sci Transl Med. 2011: 68ra9

Dixon BS, Beck GJ, Vazquez MA et al. Effect of dipyridamole plus aspirin on hemodialysis graft patency. N Engl J Med 2009;360:2191-201.

Helfet DL, Howey T, Sanders R, Johansen K. Limb Salvage Versus Amputation: Preliminary Results of the Mangled Extremity Severity Score. Clinical Orthopaedics & Related Research. 1990;256:80-6.

Holcomb J. Devastating Dismounted IED Injuries in OEF. Increasing amputation and genital injury rates admitted to LRMC 2009-2010. 2011. Presentation to the Defence Health Board,

(http://www.health.mil/Reference-Center/Presentations/2011/03/08/Devastating-Dismounted-Improvised-Explosive-Device-Injuries-in-Operation-Enduring-Freedom)

Kelly BS, Heffelfinger SC, Whiting JF, et al. Aggressive venous neointimal hyperplasia in a pig model of arteriovenous graft stenosis. Kindey Int. 2002;62(6):2272-2280.

L'Heureux N, Dusserre N, Konig G, et al. Human tissue-engineered blood vessels for adult arterial revascularization. Nat Med 2006; 12(3):361-365.

Mills JL et al on behalf of the Society for Vascular Surgery Lower Extremity Guidelines Committee. The Society for Vascular Surgery Lower Extremity Threatened Limb Classification System: Risk stratification based on Wound, Ischemia, and foot Infection (WIfI). J Vasc Surg 2013,

Morgan MBF, Crawford T, Murrin B and Fraser SCA. Developing the vascular quality of life questionnaire: a new disease specific quality of life measure for use in lower limb ischemia. J Vasc Surg 2001; 33: 679 - 687

Prichard, H.L, Manson, R.J.,Dibernado, L. et al. An early study on the mechanisms that allow tissue-engineered vascular grafts to resist vascular hyperplasia. J Cardiovasc Transl Res 2011, 4, 674-682

Quint C, Kondo Y, Manson RJ,et al. Decellularized tissue-engineering blood vessel as an arterial conduit. Proc Natl Acad Sci USA. 108(22):9214-9, 2011.

Rasmussen TE. Vascular Injury Rates in the Wars in Iraq and Afghanistan: How are they different from the past? United States Army Institute of Surgical Research, 2010.

Roy-Chaudhury P, Kelly BS, Miller MA, et al. Venous neointimal hyperplasia in polytetrafluoroethylene dialysis grafts. Kidney Int. 2001;59(6):2325-2334.

Rutherford RB1, Baker JD, Ernst C, et al Recommended standards for reports dealing with lower extremity ischemia: revised version. J Vasc Surg. 1997 Sep;26(3):517-38.

APPENDIX 1: HAV CLINICAL VISIT SCHEDULE

	Preop	D 1	D 5	D 29	M3	M 6	M 9	M12 / ET†
Informed consent	X							
Medical history and nature of trauma	X							
Prior or concomitant medication	X	X	X	X	X	X	X	X
Physical exam	X	X*	X*	X*	X*	X*	X*	X
Ultrasound or (CT) angiography **	X							
Vital signs	X		X	X				
Eligibility (inclusion/exclusion criteria)	X							
HAV implantation and angiography or US to confirm patency		X						
Documentation of surgery and any complications		X						
Clinical chemistry	X		X					
Hematology	X		X					
Clinical duplex ultrasound				X	X	X	X	X
AEs		X	X	X	X	X	X	X
Documentation of vessel interventions		X	X	X	X	X	X	X

Abbreviations: AEs, adverse events; D, day; ET, early termination; HAV, human acellular vessel; M, month; W, week.

Note: Visits should be performed using the following time windows: Day 5 or prior to discharge, D29 ± 7 days; Months 3, 6, 9, 12 ± 14 days

*Physical examination includes clinical exam of graft site and distal limb (incl. patency assessment on D1) and physical exam to evaluate AEs.

** Pre-op imaging is at the discretion of the investigator based on the clinical condition of the patient. It may be omitted if not deemed necessary in order to proceed to vascular surgical repair

† Patients withdrawn before Month 12 will perform ET visit