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Intravenous administration of umbilical cord lining stem cells in left ventricular assist device recipient: Rationale and design of the uSTOP LVAD BLEED pilot study

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ABSTRACT

Background: Left ventricular assist device (LVAD) implantation provides a robust survival advantage, however despite improvements in mortality, the adverse event burden of durable mechanical circulatory support remains high. Bleeding complications are one such significant complication. The uSTOP LVAD BLEED (Utilization of umbilical cord lining Stem cells TO Prevent LVAD associated angiodysplastic BLEEDing) pilot study is designed to evaluate the safety and tolerability of escalating doses of umbilical cord lining stem cells (ULSCs) in LVAD recipients to ameliorate the dysregulation of angiogenic factors seen in this population.

Design: This Phase Ia single-ascending dose pilot study will evaluate the IV administration of ULSCs in stable outpatients supported with an LVAD. In a 3 + 3 design, a maximum of 18 patients will receive an IV infusion of ULSCs.

Main outcome measures: The primary endpoints are safety and tolerability, secondary exploratory endpoints will include biomarker evaluation of angiogenic dysregulation.

Summary: This represents a novel cell type and route of administration in this population, while collecting initial data regarding the magnitude and duration of effects of cell therapy, and assessing the possibility of decreasing bleeding by a strategy of vascular stabilization.

Clinical trial registration: ClinicalTrials.gov Identifier: NCT04811261. https://clinicaltrials.gov/ct2/show/NCT04811261.

1. Introduction

Heart failure (HF) affects more than 6.5 million people in the U.S. and remains the leading cause of hospitalization for patients >65 years of age [1]. Due to an aging population, the projected prevalence of HF is expected to increase, and by 2030, it is estimated that 1 in every 33 individuals in the U.S. will have HF [2]. This will lead to a growing number of individuals with advanced HF given the disease's chronic and progressive nature. Heart transplantation remains the gold standard treatment for advanced HF, however due to the challenges of organ donor recipient matching, as well as the limited resource of organs, the waitlist for heart transplantation continues to grow at more than twice the rate of increase in organ availability [3]. Furthermore, transplantation is only offered to patients until the age of 65–70, depending on institution specific protocols, and comorbid conditions such as prior malignancy, diabetes, and pulmonary hypertension render many individuals not candidates for heart transplantation. As this number of patients increases, there will remain an even greater shortage of donor organs and also a growing number of patients identified as poor candidates for transplantation due to age and comorbid conditions. The development of left ventricular assist devices (LVADs), initially as a bridge to transplant and more recently with the continuing improvement in technology of the devices resulting in LVADs as destination therapy has opened a new treatment option for a significant proportion of the sickest HF patients. For patients with New York Heart Association (NYHA) Class IV HF, continuous-flow LVADs have become increasingly available, and provide patients with improved functional capacity, survival and quality of life [4]. However, despite providing increasingly impressive survival advantages, LVADs remain associated with significant adverse events. The most common adverse event is LVAD-associated gastrointestinal bleeding (GIB), with an incidence of up to 39% [5,6]. This adverse event often leads to hospitalization and invasive

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diagnostic procedures, which markedly increase the morbidity and diminish the cost-effectiveness of this life-saving therapy. Although newer generation LVADs, such as the HeartMate 3, have demonstrated better biocompatibility, this device is accompanied by a high incidence of GIB (27%), which is only marginally improved in comparison to earlier generation devices [7]. Furthermore, GIB places a significant toll on the health care system. It is the second leading cause for 30-day readmissions after LVAD implantation, and the most frequent cause for any post-implant hospitalization [8,9]. Indeed, LVAD-attributable major adverse events can cost up to average of ~$46,000 per hospitalization [10].

The pathophysiology of GIB in LVAD patients is complex due to the perturbations caused by the implantation of mechanical circulatory support combined with a failing heart and the medical therapy required to manage these patients. The resultant mucosal bleeding involves both an acquired von Willebrand syndrome (avWS) with platelet/coagulation dysfunction, as well as pathologic vascular weakness related to angiodysplasia and endothelial barrier dysfunction. The avWS is secondary to the supraphysiological shear stresses exerted on blood within LVADs. These shear stresses lead to a conformational change of von Willebrand factor (vWF) and exposure of the ADAMTS13 cleavage site of vWF, resulting in a loss of the high molecular weight multimers (HMWMs) of vWF and a decrease in platelet adhesion [11–13]. While some newer generation LVADs, specifically centrifugal flow devices such as the HVAD and HeartMate 3, are characterized by lower internal shear stresses and thereby shown to produce different patterns of avWS, this has not correlated to better platelet aggregation nor a decrease in bleeding events [14,15].

In addition to avWS, angiodysplasia manifested as arteriovenous malformations (AVMs) that develop throughout the body is a key driver of bleeding events in the LVAD population [16]. This has been postulated to be due to systemic inflammation and abnormal angiogenesis created by systemic dysregulation of angiogenic factors, specifically angiopoietin-1 and angiopoietin-2 (Ang-1 and Ang-2). Ang-1 is normally synthesized by perivascular cells and binds as an agonist to the receptor tyrosine kinase Tie-2 on the surface of endothelial cells, resulting in intracellular signaling that promotes vascular maturity and stability, and supports normal endothelial survival in conjunction with vascular endothelial factor (VEGF) [17]. Ang-2 is synthesized by endothelial cells and following release, it is available as an autocrine factor to bind Tie-2 cell surface receptors in a competitive fashion with Ang-1, antagonizing Ang-1 signaling and promoting endothelial barrier permeability [17,18].

Such dysregulation in the physiological balance of Ang-1/Ang-2 levels has been shown to lead to vascular destabilization, inflammation, vascular malformations and mucosal angiodysplasia in multiple in vitro and in vivo animal models [19–23]. Recent studies of factors modulating angiogenesis and vascular remodeling in an LVAD population have demonstrated that Ang-2 is overexpressed, while Ang-1 is reduced in patients with LVADs compared with age-matched controls, and that those patients experiencing GIB are characterized by lower serum levels of Ang-1 compared to those free of GI bleeding ($p = 0.03$) [24]. This resultant 'angiogenic switch' is further demonstrated with a markedly abnormal ratio of Ang-1/Ang-2 in LVAD patients with GIB at 0.5, compared with an Ang-1/Ang-2 ratio of 7 in patients free from GIB [24]. These data strongly suggest that a phenotype of vascular instability promoted by a systemic deficit in Ang-1 relative to Ang-2 contributes to the occurrence of non-surgical bleeding in patients with LVADs.

Historically it was felt that the only methods to improve GIB rates would be improvement in LVAD technology to decrease shear stress, and vigilant monitoring of anticoagulation. Contrary to this hypothesis, recent data have suggested that a cellular based therapy may offer a treatment that can reduce GIB in LVAD patients [25]. Our proposed Phase Ia clinical study will expand upon those findings in several novel ways. We will evaluate a novel cell type and delivery method that would make this type of treatment practical and scalable. In addition to determining the feasibility and safety of intravenous administration of ULSCs in LVAD recipients, this study will obtain initial information about the bioactivity of ULSCs and their ability to ameliorate the dysregulation of angiogenic factors seen in this population. The establishment of feasibility and safety will in turn provide a foundation for a future randomized clinical trial evaluating the effect of adjunctive administration of ULSCs in LVAD patients to provide a clinically meaningful reduction in GIB. This reduction would address a serious, unmet medical need, improve on quality of life, and markedly reduce the costs of healthcare delivery in this population.

2. Methods

2.1. Study design

The study population will consist of ambulatory patients with chronic systolic heart failure who met criteria for LVAD implantation and subsequently were implanted with a centrifugal flow LVAD either as bridge-to-transplant (BTT), bridge-to-candidacy (BTC), or destination therapy (DT). Patients will have been receiving stable medical regimen for at least two weeks, including angiotensin converting enzyme inhibitor, angiotensin receptor blocker, or angiotensin receptor-neprilysin inhibitor (ACE/ARB/ARNI), unless there is a documented contraindication to their use. Patients will also be on anti-platelet and anti-coagulation per institutional protocol. Inclusion and exclusion criteria are listed in Table 1.

This Phase Ia, single ascending dose, first in human, open-label, non-controlled, dose- escalation trial will investigate intravenous infusions of UCLSC in LVAD patients. This study will recruit patients with recently implanted patients, as well as patients on chronic long-term support, with the goal of enrolling up to 18 subjects to participate in the study. The participation of each subject in the study includes informed consent, screening, one administration of the study treatment (ULSC infusion), and follow-up for a duration of 12 months.

Subjects will be entered into a 3 + 3 dose escalation study [Fig. 1], a standard design for initial trials to evaluate tolerability of a therapeutic agent and identify a maximal tolerated dose if indeed toxicities are identified in the pre-specified dosing range [26]. The first 3 subjects' patients will be administered 50×10^6 ULSCs reconstituted in HTS-FRS

Table 1
Key Inclusion and exclusion criteria.

Inclusion
≥ 18 years of age
Have heart failure with reduced ejection fraction and a durable centrifugal flow LVAD
Be on a stable regimen of heart failure medications for at least two weeks, including ACEi/ARB/ARNi unless there is a documented contraindication to their use.
Exclusion
Durable biventricular support
An axial flow LVAD
History of Crohn's Disease, Ulcerative Colitis, or other Inflammatory Bowel Disease on active treatment
LVAD implantation within the last 30 days
Anticipated need for non-cardiac surgery within the next 12 months
Evidence of active systemic infection at time of study product delivery
Evidence of infectious diseases such as hepatitis B, hepatitis C and HIV
Prior heart transplant recipients
Active cancer (or prior diagnosis of cancer within the past 2 years)
Recent (<14 days) or active use of immunosuppressive drugs
Chronic auto-immune or auto-inflammatory disease
Known hypo- or hyper-coagulable state such as disseminated intravascular coagulation and heparin induced thrombocytopenia (HIT)
Platelet count <100 K
Inability to maintain an INR of 2–3

LVAD = left ventricular assist device, ACE = angiotensin converting enzyme inhibitor, ARB = angiotensin receptor blocker, ARNi = angiotensin receptor-neprilysin inhibitor, INR = international normalizing ratio.

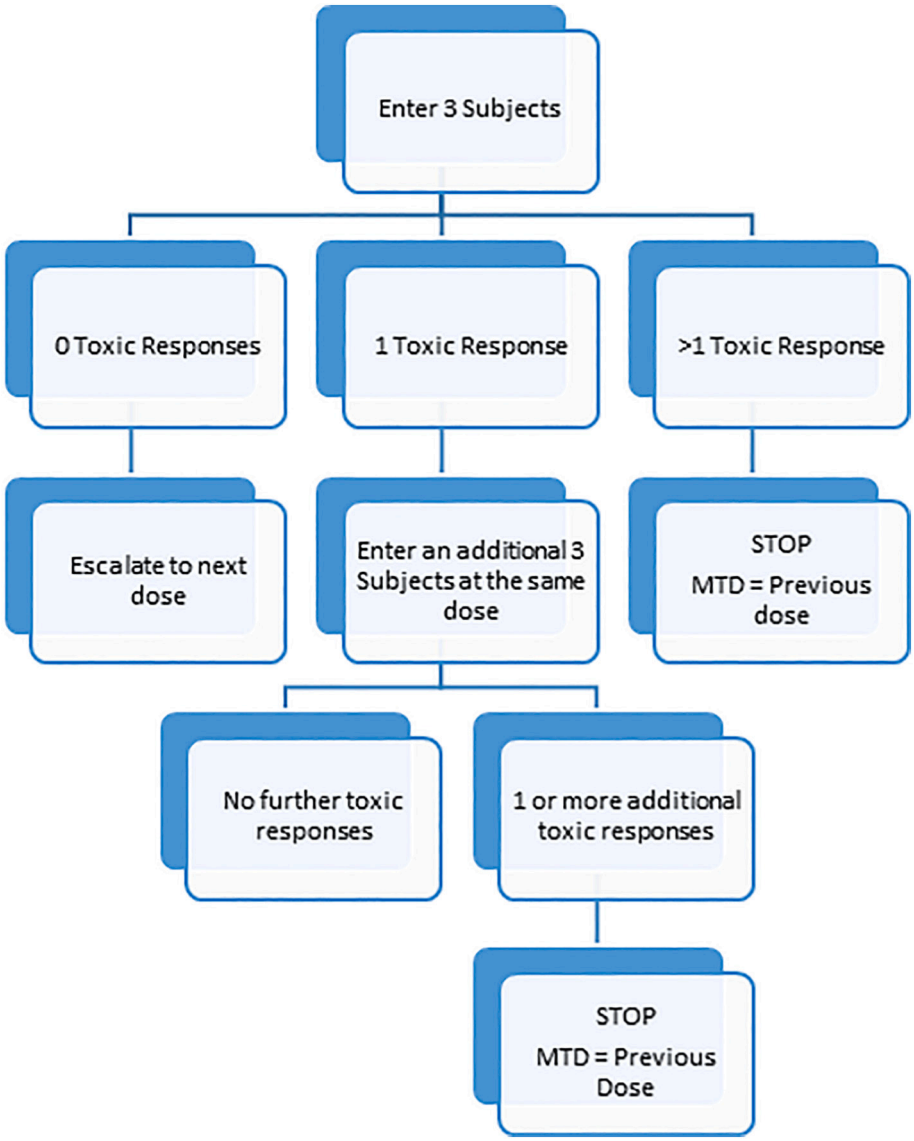

Fig. 1. 3 × 3 Ascending dose escalation design.
MTD = maximum tolerated dose.

(Biolife Solutions) in a volume of 250 ml, and monitored for adverse events or toxicities, immediately following dosing, and again at 7 and 30 days. Safety endpoints will be reviewed by the DSMB and depending on the response, the dose will be escalated to 100×10^6 and subsequently 200×10^6 ULSC in a constant 250 ml volume. Adverse events (AEs) and dose limiting toxicities (DLTs) will be recorded and used to direct enrollment. An independent DSMB, comprised of two cardiologists and one statistician with authority to suspend enrollment, will conduct ongoing review of the nature, frequency, and severity of the safety data. We will list all the incidence of DLTs for each dose evaluation cohort. DLTs will be recorded and used to direct enrollment as follows:

- If 0 out of 3 enrolled subjects at the low dose have a DLT after 1 month of monitoring, and review of safety data, then we will proceed to initiate enrollment in the intermediate dose cohort and enroll 3 subjects. If 0 out of these first 3 enrolled subjects treated with an intermediate dose have a DLT, we will enroll another 3 subjects in the high dose group.
- If 1 out of 3 subjects have a DLT at either dose, then we will add 3 more subjects at the same dose level. Outcomes will be evaluated within each dosing group and treatment arm.
- If 2 or 3 subjects in a dosing group experience a DLT, then we plan to pause the trial to obtain DSMB input as to the risk/benefit ratio in the exact circumstance encountered. The maximum tolerated dose (MTD) will be defined as the dose level that does not cause this frequency of DLT events.

Study participants will be followed-up for 12 months through a total of 8 scheduled clinic visits and/or phone calls (Clinic visits at day 7, 1 month, 3 months, 6 months and 12 months; phone calls at day 3, 2 months, and 4 months SPI).

2.2. Study objective & endpoints

This study will attempt to answer the following research questions regarding ULSC as an investigational medicinal product and its use: (1) Is the IV delivery of ULSCs safe and well-tolerated in patients with LVADs? (2) What is the tolerability of ascending doses of ULSCs? (3) What is the magnitude and duration of the effect of ULSCs delivery on

surrogate biomarkers of inflammation and angiodysplasia?

The primary endpoint of this study will focus on safety based in 3 domains; tolerability of infusion assessed at time of infusion, provocation of thrombosis or hemolysis during the study period, and immune sensitization during the study period. Secondary safety endpoints include changes in complete blood count (CBC), renal function, and liver function from baseline to 1, 3, 6, and 12 months. After the enrollment of each cohort of 3 patients, safety data will be reviewed by assessing any immediate adverse effects, as well as at 7 and 30 days. These data will be reviewed by the DSMB and further enrollment will be halted until the DSMB has cleared ongoing recruitment and dose for the subsequent cohort of 3. Exploratory endpoints will assess for any evidence of early efficacy in providing vascular stability or improvement in a composite of clinical outcomes. The details of all endpoints are listed in Table 2.

2.3. Statistical considerations

We will conduct interim analysis for 1, 3, and 6 month and final analysis after all data have been collected. The biomarkers Ang-1, Ang-2, soluble Tie2 (sTie2), VEGF will be evaluated as continuous variables by ANOVA to assess changes from time zero (pre-infusion) in comparison to 1 month, 3 month, 6 month and 12 month levels. This approach will also be used for qFIT. As this is a Phase I study, we anticipate that we may not have power to detect changes in these variables, and are treating these, as well as the exploratory clinical outcome of the cumulative incidence of angiodysplastic bleeding, as exploratory analyses useful to demonstrate feasibility in this patient population.

We will compute summary statistics such as mean, standard deviation, minimum, median, and maximum for continuous variables. We will compute frequency and percentage for categorical variables. For time to event variable, we will analyze with Kaplan-Meier product limit method.

3. Discussion

Durable mechanical support has not only revolutionized our approach to advanced heart failure, it continues to evolve and provide greater survival advantages to LVAD recipients, with the most recent registry data demonstrating a median survival of greater than 4 years regardless of pump type or indication for implantation [27]. However, due to the ongoing burden of adverse events and potential for rehospitalization as a result, LVADs remain only a moderately cost-effective therapy with wide ranging estimates of incremental cost-effectiveness ratio (ICER) per quality adjusted life year (QALY), none of which have been better than $84,963 [28–31]. The most recent of these analyses suggested that ICER/QALY could be augmented by nearly 67% with streamlining of out-patient costs and a reduction in readmissions by 1 event per-patient year [29]. Therefore, considerable effort should be placed in creating management strategies to diminish both the incidence of angiodysplastic bleeding as well as its impact as it is the largest driver of readmissions and a significant contributor to the cost and intensity of out-patient care of LVAD patients.

Table 2
uSTOP LVAD BLEED Endpoints.

Primary Safety Endpoints
Fever, pain at infusion site, evidence of allergic reaction, dyspnea or hypoxia during infusion
Adjudicated clinical LVAD thromboembolic events
Changes in LDH levels
Changes in PRA
Secondary Safety Endpoints
Changes in CBC
Changes in AST, ALT, serum bilirubin
Changes in BMP
Clinical evidence of tumors
Exploratory Endpoints
Change in biomarkers (Ang-1, Ang-2, soluble Tie2, VEGF)
Angiodysplastic bleeding (occult GIB as evidenced by positive qFIT, $\geq$ 2 g drop in HgB with no identifiable bleeding source, epistaxis which requires transfusion or results in $\geq$2 g drop in HgB)
Hospitalization for bleeding
Need for any blood transfusion

LVAD = left ventricular assist device, DH = lactose dehydrogenase, PRA = panel reactive antibody, CBC = complete blood count, AST = aspartate aminotransferase, ALT = alanine transaminase, BMP = basic metabolic panel, Ang-1 = angiopoetin 1, ANG-2 = angiopoetin 2, VEGF = vascular endothelial growth factor, GIB = gastrointestinal bleeding, qFIT = Quantitative Faecal Immunochemical Test, HgB = hemoglobin.

Cell therapy represents an attractive target to accomplish these goals due to the mechanism of angiodysplastic bleeding in this population; with significant contributions from vascular instability, pro-inflammatory cytokines and dysregulation of angiogenic factors; coupled with the unique properties of mesenchymal stem cells (MSCs). Our overarching hypothesis is that the administration of MSCs, which are known to down-modulate pro-inflammatory cytokines in a wide range of inflammatory states, and to promote vascular stabilization by paracrine mechanisms including the secretion of Ang-1 will provide a useful therapeutic approach to reduce mucosal bleeding in patients with LVADs [32].

Furthermore, umbilical cord derived MSCs have been demonstrated to enhance vascular remodeling that promotes vascular integrity. Recent data in two ischemic brain rodent models show that administration of UC-MSC lead to decreased circulating Ang-2 levels while increasing both the level of circulating Ang-1 and the expression of Tie2 cell-surface receptor, which combine to provide vascular stability as well as improved functional outcomes [33,34]. A key role for secretion of Ang-1 was highlighted by studies in a murine lung model in which unmodified MSCs promoted vascular stability, while MSCs overexpressing Ang-1 were found to even more potently decrease pulmonary vascular leak as well as proinflammatory cytokine levels [35]. Mechanisms underlying improvement in endothelial layer integrity were recently described in human lung microvascular endothelium cells as including a transfer of Ang-1 mRNA to the damaged endothelium, resulting in an ongoing increase in Ang-1 secretion [36]. Furthermore, Ang-1 has also been demonstrated to prevent bleeding complications associated with anticoagulation in multiple pre-clinical models by augmenting vascular stability and preventing hemorrhage despite a persistent anticoagulant state [37]. These mechanistic findings suggest that administration of MSCs would directly address key systemic factors that result in mucosal bleeding in LVAD recipients.

The above pathophysiologic considerations are bolstered by clinical data demonstrating an improvement in GIB in LVAD patients with cell therapy. Intramyocardial injections of allogeneic MSCs at the time of LVAD implantation was shown to significantly decrease the rate of major GIB events by 76%, and the cumulative incidence by 48% [25]. This confirmed data seen in a 30-patient pilot demonstrating a longer time to first hospitalization due to major GIB. This has led the FDA to grant orphan drug designation to rexlemestrocel-L, the cell therapy utilized in the cited trial [38]. That this data was obtained in a non-contemporary axial flow LVAD cohort, with a negative primary endpoint, and via an invasive delivery platform, informs our decision to perform this more focused investigation.

Additional benefits of the uSTOP LVAD BLEED pilot include dose definition as there is not yet a standard dosing regimen for cell therapy in heart failure patients. The aforementioned LVAD trial provided patients with 1.5×10^8 cells. The RIMECARD trial provided umbilical cord-derived MSCs for patients with heart failure via intravenous infusions of 1×10^6 cells/kg [39]. Similarly, another Phase IIa trial of MSCs in HFrEF used 1.5×10^6 cells/kg [40]. While both of these studies demonstrated the safety of IV administration of stem cells in heart

failure patients, we hope to define a dosing range as well as magnitude and duration of affect based upon dose which may inform future investigations. Importantly, neither study demonstrated any allosensitization affects of cell therapy, an important consideration for patients who may be considered for transplantation.

The small sample size of this study will preclude any definitive conclusions regarding the effects of ULSCs in LVAD patients. This, however, is appropriate for a Phase Ia study to evaluate safety and tolerability, while potentially informing early mechanistic explorations. Other limitations include the possible heterogeneity in medical therapy which impacts the Ang-1 and Ang-2 pathway, specifically ACEi/ARB/ARNi and digoxin, which we have attempted to mitigate with our protocol.

4. Conclusion

This manuscript delineates a pilot investigation of ULSCs in a selected and stable LVAD patient population with the objective to evaluate the safety and tolerability of IV infusion of ULSCs in this, while exploring early markers of efficacy in potentially ameliorating markers of vascular stability. The hope is that results will inform a larger late-phase study powered to demonstrate a change in the clinical outcome of angiodysplastic bleeding.

CRediT authorship contribution statement

Mustafa M. Ahmed: Conception and design, manuscript writing, final approval of manuscript.

Lauren E. Meece: Administrative support, manuscript writing, final approval of manuscript.

Eileen M. Handberg: Conception and design, manuscript writing, final approval of manuscript.

Carl J. Pepine: Conception and design, manuscript writing, final approval of manuscript.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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References

[1] E.J. Benjamin, M.J. Blaha, S.E. Chiuve, et al., Heart disease and stroke Statistics-2017 update: a report from the American Heart Association, Circulation 135 (10) (2017) e146–e603, https://doi.org/10.1161/CIR.0000000000000485.

[2] P.A. Heidenreich, N.M. Albert, L.A. Allen, et al., Forecasting the impact of heart failure in the United States: a policy statement from the American Heart Association, Circ. Heart Fail. 6 (3) (2013) 606–619, https://doi.org/10.1161/HHF.0b013e318291329a.

[3] Annual Report of the U.S. Organ Procurement and Transplantation Network and the Scientific Registry of Transplant Recipients: Transplant Data 1994-2003, Department of Health and Human Services, Health Resources and Services Administration, Healthcare Systems Bureau, Division of Transplantation, Rockville, MD, 2017. United Network for Organ Sharing, Richmond, VA; University Renal Research and Education Association, Ann Arbor, MI.

[4] J.G. Rogers, K.D. Aaronson, A.J. Boyle, et al., Continuous flow left ventricular assist device improves functional capacity and quality of life of advanced heart failure patients, J. Am. Coll. Cardiol. 55 (17) (2010) 1826–1834, https://doi.org/10.1016/j.jacc.2009.12.052.

[5] Z.T. Demirozu, R. Radovancevic, L.F. Hochman, et al., Arteriovenous malformation and gastrointestinal bleeding in patients with the HeartMate II left ventricular assist device, J. Heart Lung Transplant. 30 (8) (2011) 849–853, https://doi.org/10.1016/j.healun.2011.03.008.

[6] H.R. Jabbar, A. Abbas, M. Ahmed, et al., The incidence, predictors and outcomes of gastrointestinal bleeding in patients with left ventricular assist device (LVAD), Dig. Dis. Sci. 60 (12) (2015) 3697–3706, https://doi.org/10.1007/s10620-015-3743-4.

[7] M.R. Mehra, D.J. Goldstein, N. Uriel, et al., Two-year outcomes with a magnetically levitated cardiac pump in heart failure, N. Engl. J. Med. 378 (15) (2018) 1386–1395, https://doi.org/10.1056/NEJMoa1800866.

[8] S. Agrawal, L. Garg, M. Shah, et al., Thirty-day readmissions after left ventricular assist device implantation in the United States: insights from the Nationwide readmissions database, Circ. Heart Fail. 11 (3) (2018), e004628, https://doi.org/10.1161/CIRCHEARTFAILURE.117.004628.

[9] S.A. Akhter, A. Badami, M. Murray, et al., Hospital readmissions after continuous-flow left ventricular assist device implantation: incidence, causes, and cost analysis, Ann. Thorac. Surg. 100 (3) (2015) 884–889, https://doi.org/10.1016/j.athoracsur.2015.03.010.

[10] M.R. Mehra, C. Salerno, J.C. Cleveland, et al., Healthcare resource use and cost implications in the MOMENTUM 3 long-term outcome study, Circulation 138 (18) (2018) 1923–1934, https://doi.org/10.1161/CIRCULATIONAHA.118.035722.

[11] U. Geisen, C. Heilmann, F. Beyersdorf, et al., Non-surgical bleeding in patients with ventricular assist devices could be explained by acquired von willebrand disease, Eur. J. Cardiothorac. Surg. 33 (4) (2008) 679–684, https://doi.org/10.1016/j.ejcts.2007.12.047.

[12] H.M. Tsai, I.I. Sussman, R.L. Nagel, Shear stress enhances the proteolysis of von Willebrand factor in normal plasma, Blood 83 (8) (1994) 2171–2179, https://doi.org/10.1182/blood.V83.8.2171.2171.

[13] N. Uriel, S.W. Pak, U.P. Jorde, et al., Acquired von Willebrand syndrome after continuous- flow mechanical device support contributes to a high prevalence of bleeding during long- term support and at the time of transplantation, J. Am. Coll. Cardiol. 56 (15) (2010) 1207–1213, https://doi.org/10.1016/j.jacc.2010.05.016.

[14] A.L. Meyer, D. Malehsa, U. Budde, C. Bara, A. Haverich, M. Strueber, Acquired von Willebrand syndrome in patients with a centrifugal or axial continuous flow left ventricular assist device, JACC Heart Fail. 2 (2) (2014) 141–145, https://doi.org/10.1016/j.jchf.2013.10.008.

[15] I. Netuka, T. Kvasnicka, J. Kvasnicka, et al., Evaluation of von willebrand factor with a fully magnetically levitated centrifugal continuous-flow left ventricular assist device in advanced heart failure, J. Heart Lung Transplant. 35 (7) (2016) 860–867, https://doi.org/10.1016/j.healun.2016.05.019.

[16] S.R. Patel, S. Madan, O. Saeed, et al., Association of nasal mucosal vascular alterations, gastrointestinal arteriovenous malformations, and bleeding in patients with continuous-flow left ventricular assist devices, JACC Heart Fail. 4 (12) (2016) 962–970, https://doi.org/10.1016/j.jchf.2016.08.005.

[17] T. Cascone, J.V. Heymach, Targeting the angiopoietin/Tie2 pathway: cutting tumor vessels with a double-edged sword? J. Clin. Oncol. 30 (4) (2012) 441–444, https://doi.org/10.1200/JCO.2011.38.7621.

[18] D.J. Metcalf, T.D. Nightingale, H.L. Zenner, W.W. Lui-Roberts, D.F. Cutler, Formation and function of Weibel-Palade bodies, J. Cell Sci. 121 (Pt 1) (2008) 19–27, https://doi.org/10.1242/jcs.03494.

[19] G. Holleran, B. Hall, M. O'Regan, S. Smith, D. McNamara, Expression of angiogenic factors in patients with sporadic small bowel angiodysplasia, J. Clin. Gastroenterol. 49 (10) (2015) 831–836, https://doi.org/10.1097/MCG.0000000000000260.

[20] R.D. Liersch, W.E. Berdel, T. Kessler, Angiogenesis inhibition, Springer, Heidelberg, Germany, 2010.

[21] A.M. Randi, M.A. Laffan, R.D. Starke, Von willebrand factor, angiodysplasia and angiogenesis, Mediterr. J. Hematol. Infect. Dis. 5 (1) (2013), e2013060, https://doi.org/10.4084/MJHID.2013.060.

[22] P. Redondo, L. Aguado, M. Marquina, et al., Angiogenic and prothrombotic markers in extensive slow-flow vascular malformations: implications for antiangiogenic/ antithrombotic strategies, Br. J. Dermatol. 162 (2) (2010) 350–356, https://doi.org/10.1111/j.1365-2133.2009.09513.x.

[23] T. Ziegler, J. Horstkotte, C. Schwab, et al., Angiopoietin 2 mediates microvascular and hemodynamic alterations in sepsis, J. Clin. Invest. 123 (8) (2013) 3436–3445, https://doi.org/10.1172/jci66549.

[24] A.L. Edwards, C.M. Wilcox, S.V. Pamboukian, P. Mannon, S. Peter, Gastrointestinal bleeding is associated with a shift towards a pro-angiogenic phenotype after continuous flow left ventricular assist device implantation, J. Card. Fail. 24 (8 Suppl) (2018) S22–S23, https://doi.org/10.1016/j.cardfail.2018.07.066.

[25] T.M. Yau, F.D. Pagani, D.M. Mancini, et al., Intramyocardial injection of mesenchymal precursor cells and successful temporary weaning from left ventricular assist device support in patients with advanced heart failure: a randomized clinical trial, JAMA 321 (12) (2019) 1176–1186, https://doi.org/10.1001/jama.2019.2341.

[26] B.E. Storer, Design and analysis of phase I clinical trials, Biometrics 45 (3) (1989) 925–937.

[27] E.J. Molina, P. Shah, M.S. Kiernan, et al., The Society of Thoracic Surgeons intermacs 2020 annual report, Ann. Thorac. Surg. 111 (3) (2021) 778–792, https://doi.org/10.1016/j.athoracsur.2020.12.038.

[28] A. Clarke, R. Pulikottil-Jacob, M. Connock, et al., Cost-effectiveness of left ventricular assist devices (LVADs) for patients with advanced heart failure: analysis of the british NHS bridge to transplant (BTT) program, Int. J. Cardiol. 171 (3) (2014) 338–345, https://doi.org/10.1016/j.ijcard.2013.12.015.

[29] J.Baras Shreibati, J.D. Goldhaber-Fiebert, D. Banerjee, et al., Cost-effectiveness of left ventricular assist devices in ambulatory patients with advanced heart failure, JACC Heart Fail. 5 (2) (2017) 110–119, https://doi.org/10.1016/j.jchf.2016.09.008.

[30] D.S. Chew, B. Manns, R.J.H. Miller, et al., Economic evaluation of left ventricular assist devices for patients with end stage heart failure who are ineligible for cardiac

transplantation, Can. J. Cardiol. 33 (10) (2017) 1283–1291, https://doi.org/10.1016/j.cjca.2017.07.012.
[31] J.G. Rogers, R.R. Bostic, K.B. Tong, et al., Cost-effectiveness analysis of continuous-flow left ventricular assist devices as destination therapy, Circ. Heart Fail. 5 (1) (2012) 10–16, https://doi.org/10.1161/CIRCHEARTFAILURE.111.962951.
[32] J. Rehman, D. Traktuev, J. Li, et al., Secretion of angiogenic and antiapoptotic factors by human adipose stromal cells, Circulation 109 (10) (2004) 1292–1298, https://doi.org/10.1161/01.CIR.0000121425.42966.F1.
[33] Q. Zhao, J. Hu, J. Xiang, et al., Intranasal administration of human umbilical cord mesenchymal stem cells-conditioned medium enhances vascular remodeling after stroke, Brain Res. 1624 (2015) 489–496, https://doi.org/10.1016/j.brainres.2015.08.003.
[34] X. Cui, M. Chopp, A. Zacharek, et al., Combination treatment of stroke with sub-therapeutic doses of simvastatin and human umbilical cord blood cells enhances vascular remodeling and improves functional outcome, Neuroscience 227 (2012) 223–231, https://doi.org/10.1016/j.neuroscience.2012.09.066.
[35] S.H. Mei, S.D. McCarter, Y. Deng, C.H. Parker, W.C. Liles, D.J. Stewart, Prevention of LPS-induced acute lung injury in mice by mesenchymal stem cells overexpressing angiopoietin 1, PLoS Med. 4 (9) (2007), e269, https://doi.org/10.1371/journal.pmed.0040269.
[36] S. Hu, J. Park, A. Liu, et al., Mesenchymal stem cell microvesicles restore protein permeability across primary cultures of injured human lung microvascular endothelial cells, Stem Cells Transl. Med. 7 (8) (2018) 615–624, https://doi.org/10.1002/sctm.17-0278.
[37] M. Jerebtsova, J.R. Das, P. Tang, E. Wong, P.E. Ray, Angiopoietin-1 prevents severe bleeding complications induced by heparin-like drugs and fibroblast growth factor-2 in mice, Am. J. Physiol. Heart Circ. Physiol. 309 (8) (2015) H1314–H1325, https://doi.org/10.1152/ajpheart.00373.2015.
[38] Mesoblast Ltd, Mesoblast heart failure cell therapy receives orphan drug designation from FDA for prevention of gastrointestinal bleeding in patients with left ventricular assist devices, Available at, investorsmedia.mesoblast.com/static-files/c08cebeb-456b-49d4-8e0d-cfc5df110d9e. (Accessed 24 June 2019).
[39] J. Bartolucci, F.J. Verdugo, P.L. Gonzalez, et al., Safety and efficacy of the intravenous infusion of umbilical cord mesenchymal stem cells in patients with heart failure: a phase 1/2 randomized controlled trial (RIMECARD trial [Randomized clinical trial of intravenous infusion umbilical cord mesenchymal stem cells on Cardiopathy]), Circ. Res. 121 (10) (2017) 1192–1204, https://doi.org/10.1161/CIRCRESAHA.117.310712.
[40] J. Butler, S.E. Epstein, S.J. Greene, et al., Intravenous allogeneic mesenchymal stem cells for nonischemic cardiomyopathy: safety and efficacy results of a phase II-a randomized trial, Circ. Res. 120 (2) (2017) 332–340, https://doi.org/10.1161/CIRCRESAHA.116.309717.