



Fat obtained from plastic surgery procedures—stem cells derived from adipose tissue and their potential in technological innovation: a narrative literature review and perspective on dissociative methods

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Abstract

Background Throughout its illustrious history, plastic surgery has searched for novel regenerative therapies and procedures. Recently, interest has emerged in using adipose tissue-derived stem cells (ASCs) in an ethical, easy, and reproducible manner. ASCs are generally not administered alone but as a constituent of the stromal vascular fraction (SVF) in clinical practice. Herein, we searched for innovative fat collection and ASC isolation technologies and applications and evaluated each study's relevance to plastic surgery.

Methods A narrative literature review was carried out using the MEDLINE/PubMed databases. Studies published from January 1993 to August 2020 and written in English, Portuguese, or Spanish were considered.

Results The selection process yielded 33 articles for subsequent review, involving exploratory, selective, and interpretive reading, material choice, and text analysis. Twenty-three articles employed enzymatic dissociation methods to isolate ASCs, and 25 employed liposuction as the plastic surgery technique. Moreover, articles describing new devices ($n=2$), techniques ($n=4$), computational models ($n=1$), tissue scaffolds ($n=21$), and therapies and/or treatments ($n=5$) were identified.

Conclusions Given the importance of fat tissue for plastic surgery purposes, innovative ASC isolation and liposuction technologies could change how the surgeon conducts surgeries and improve surgical outcomes. Furthermore, many articles investigating tissue scaffolds demonstrate the importance of this area of research and development in plastic surgery and regenerative medicine. Continued efforts in the identified research areas will eventually bring in vivo human plastic surgery applications and regenerative medicine into the operating room. Level of evidence: Not gradable.

Keywords Adipose tissue-derived stem cells · Innovation-technology · Plastic surgery · Dissociative methods

Introduction

Fat tissue, like bone marrow, is derived from the mesenchyme and contains a stroma that is easily isolated, thus making it a potential source of regenerative cells [1, 2]. It has been shown that the lipoaspirate is naturally a rich source of mesenchymal and stromal cells, exhibiting stable growth and kinetic proliferation in specific culture media [1, 3]. When processed, these cells tend to concentrate in the stromal vascular fraction (SVF), which is the infranant of the lipoaspirate [1]. When cultured in vitro, ASCs

can differentiate into osteogenic, adipogenic, myogenic, and chondrogenic lineages when subjected to specific lineage factors [1]. On the other hand, the non-cultured heterogeneous population of cells, known as stromal cells (SCs) or SVF cells, contain ASCs, hematopoietic stem cells, progenitor cells, endothelial cells, red blood cells (RBCs), fibroblasts, lymphocytes, monocytes/macrophages, pericytes, and other cell types [4].

In clinical practice, adipose tissue-derived mesenchymal cells are generally not administered alone but as a constituent of the SVF, a heterogeneous mixture of cells resulting from aspirated adipose tissue processing [1]. Notably, the adipose tissue SVF has become the focus of mesenchymal cell research, regenerative medicine, and fat grafting, with new processing equipment and methods being developed worldwide. Indeed, the engineering associated with SVF cells represents an exciting field of

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research for different diseases, including degenerative, congenital, or traumatic conditions and bone, joint, and soft tissue defects. These cells are primarily used in plastic surgery to supplement fat grafts, improving graft retention and long-term results [5–7].

The SVF can be dissociated and isolated from the adipose tissue using enzymatic and nonenzymatic methods, manually, or in an automated closed system. The most used isolation protocol involves washing the lipoaspirate, digesting it with collagenase enzyme, centrifugation, and RBC lysis [1]. It should be pointed out that despite the frequent use of this technique, there are challenges and barriers associated with collagenase, making it currently only appropriate for in vitro and in vivo animal studies. This observation opens up the perspective for more innovative techniques that can be utilized in humans.

In 2009, the concept of nonenzymatic dissociation was proposed and investigated to circumvent these collagenase-related obstacles [8]. However, in 2013, Shah et al. (2013) observed that cells acquired from a nonenzymatic method proliferated more slowly in culture [9]. Several authors have demonstrated that the composition of cell populations recovered by simple centrifugation and other nonenzymatic methods contains more significant proportions of peripheral blood mononuclear cells (PBMCs) and substantially fewer progenitor cells [7, 9–11]. Despite this apparent drawback, unique devices have been created to separate and concentrate the ASC and SVF cells from the adipose tissue stroma. These systems circumvent the need for manual and external manipulation, consequently facilitating fat incorporation and improving grafting results [12, 13]. Currently, there are several devices at various stages of development for the isolation of SVF cells. Each device is distinct in terms of complexity, automation, cost, efficiency, and effectiveness [14].

Considering that an invention is a discovery or a new way of doing something, identifying innovation can transform the invention into a product. Some examples of innovation

include using new devices in existing procedures, introducing new procedures that use new devices, and using existing devices in new procedures [15]. When an innovation changes the way people usually perform a procedure, it can become disruptive.

Technical innovation is essential for plastic surgeons because it stimulates basic and clinical research to develop novel procedures and treatments. Unique plastic surgery niches that have significantly benefited from innovations and inventions include fat transfer, microsurgery, muscle flaps, tissue expansion, craniofacial surgery, transplantation, liposuction, and laser technologies [16]. Indeed, innovation makes plastic surgery different and distinct and is vital for the specialty's survival [15].

Even though innovation has constantly fueled the plastic surgery field, studies evaluating the innovative and potentially disruptive technologies in this specialty are scarce. Herein, we conducted a narrative review to inform plastic surgeons about emerging and potentially disruptive technologies involving adipose tissue-derived SVF cell-based therapies (i.e., ASCs and the SVF). We hope this article will open up new frontiers, develop ideas, and raise the awareness of physicians that want to utilize these cells in clinical practice [14].

Methods

Study design

This study consisted of a narrative literature review. Data collection was performed using works published from January 1993 to August 2020, in English, Portuguese, and Spanish, in the MEDLINE/PubMed databases. The search strategies to identify relevant studies describing innovative plastic surgery procedures and technological applications employed five search strategies, summarized in Table 1. Strategy #1 used the following terms or combinations

Table 1 Search strategy using the MEDLINE/PubMed database

Strategy	Terms or combination of terms used
#1	"mesenchymal stem cells"[MeSH Terms] OR ("mesenchymal"[All Fields] AND "stem"[All Fields] AND "cells"[All Fields]) OR "mesenchymal stem cells"[All Fields]
#2	"stromal cells"[MeSH Terms] OR ("stromal"[All Fields] AND "cells"[All Fields]) OR "stromal cells"[All Fields]
#3	bodyweight [tw] OR obesity OR skinfold thickness [tw] OR bariatrics OR lipectomy OR lipoaspiration OR plastic surgery)
#4	Cloud computer OR Machine learning OR Big data OR Bussiness Intelligence Virtual reality OR Augmented reality OR Artificial intelligence OR Robotics
#5	diffusion of innovation OR inventions OR Knowledge Management OR information dissemination OR change management OR culturally appropriate technology OR patents OR patents as topic OR intellectual property OR tecnolog* OR economic development OR cost–benefit analysis (#1 OR #2) AND #3 AND (#4 OR #5)

MH, mesh terms; applied filters, human, English, Portuguese, and Spanish

of terms "mesenchymal stem cells"[MeSH Terms] OR ("mesenchymal"[All Fields] AND "stem"[All Fields] AND "cells"[All Fields]) OR "mesenchymal stem cells"[All Fields]; Strategy #2 used "stromal cells"[MeSH Terms] OR ("stromal"[All Fields] AND "cells"[All Fields]) OR "stromal cells"[All Fields]; Strategy #3 used (bodyweight [tw] OR obesity OR skinfold thickness [tw] OR bariatrics OR lipectomy OR lipoaspiration OR plastic surgery); Strategy #4 used Cloud computer OR Machine learning OR Big data OR Business Intelligence Virtual reality OR Augmented reality OR Artificial intelligence OR Robotics; and Strategy #5 used diffusion of innovation OR inventions OR Knowledge Management OR information dissemination OR change management OR culturally appropriate technology OR patents OR patents as topic OR intellectual property OR technolog* OR economic development OR cost–benefit analysis. Additionally, we utilized a combination of Strategies #1 OR #2 AND #3 AND #4 OR #5.

Eligibility criteria

Review articles and articles that did not use human cells were not considered. The following eligibility criteria were used for the inclusion of articles:

1. Obtained adipose-derived cells (ADCs)
2. Obtained ASCs and SVF cells
3. Used plastic surgery procedures that remove and collect fat
4. Demonstrated potential for technological innovation in the processing or use of ASCs.
5. Original article or review

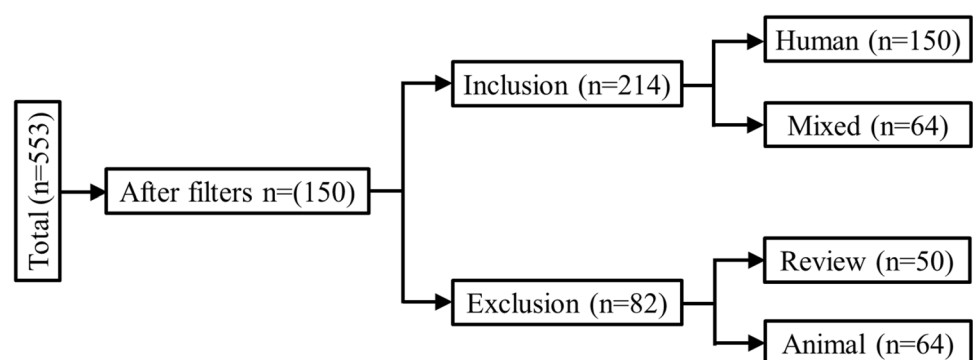
After selecting articles, an exploratory reading was carried out based on the titles and abstracts, and then a full reading of the selected article, contemplating the objectives of this study and interpretation of the text, was performed.

Results

The MEDLINE/PubMed database search was conducted on September 21, 2020, and retrieved 553 potential articles published between 1993 and 2020. Applying filters and reducing the timeframe only to include articles published from 2001 to 2020 reduced the sample to 296 articles. The organization chart in Fig. 1 shows the number of articles included and excluded from the study. Separating the 214 tentatively selected articles based on whether it was a human or mixed study, further subdividing these articles as plastic surgery-related or other, and then evaluating each article demonstrated that a total of 33 articles (17 human and 16 mixed) presented innovation in their area of research and development. Figure 2 displays the number and distribution of the identified articles. A summary of the selected articles for this study is presented in alphabetical order in Table 2.

As shown in Table 3, the most used method for isolating ASCs was enzymatic dissociation ($n=21$), followed by mixed methodologies ($n=3$), using manual isolation by enzymatic and mechanical dissociation in the same work. Furthermore, one study used a mechanical technique, and six studies did not report the ASC isolation method. Concerning the preferred plastic surgery procedure, most of the studies used liposuction ($n=22$) or abdominoplasty ($n=4$). Four studies used more than one technique, and three did not report the surgical procedure (Table 3). Notably, the most observed innovative technologies were associated with scaffold development. Additionally, studies utilized and/or developed innovative equipment, such as a mechanical system for cell separation, 3D printers, bioreactors, and other procedures, for tissue collection, grafting, and therapeutic purposes (Table 3).

Fig. 1 Distribution of results from the MEDLINE/PubMed database. The organization chart shows the number of articles found in each research phase divided between inclusion and exclusion. The filters used were human, English, Portuguese, and Spanish



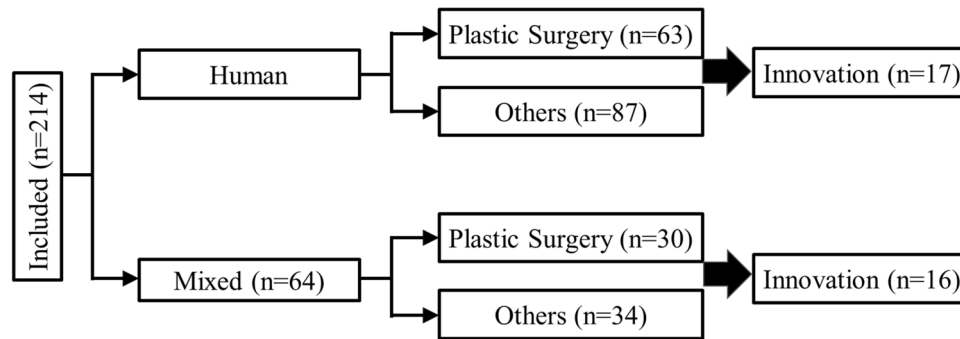


Fig. 2 Distribution of results included in the study. The organization chart shows the number of articles included in the study and later divided into human (used stem cells derived from adipose tissue) and

mixed (used stem cells derived from adipose tissue associated with nonhuman models). The groups were subdivided to locate innovative plastic surgery procedures

Discussion

We are just now experiencing the beginning of a scientific revolution. Recent scientific advances have facilitated the sequencing of the human and other genomes and the creation of genetically modified plants and animals. The rapid growth in scientific research and development resulting from innovative and sometimes disruptive technologies has changed how we function at work and as a society.

In medicine, disruptive technologies are currently driven by biotechnology and digital processing. Indeed, bridging science and technology to convert signs and symptoms into information for Big Data and Machine Learning could improve health and society [17]. In plastic surgery, tissue manipulation, especially adipose tissue and adipose-derived cells (ADCs), has demonstrated regenerative potential, bringing innovation and technology to the daily life of plastic surgeons. Therefore, the field has been forced to embrace biomolecular medicine and tissue engineering, which sometimes requires the surgeon to bring the laboratory into the operating room.

Technology-based innovation leads to the development of new designs, materials and products, and/or procedures. It can include equipment, components, and/or processes that introduce novel techniques, describe new layouts, or improve existing procedures or methods. Indeed, innovative processes facilitate the efficient production of quality products at the lowest possible cost [18]. The main characteristic of innovation is its novelty and the possibility of different interpretations [19–21]. Several scholars have recognized that the criterion “novelty” cannot be the only criterion for innovation because inventions or ideas only become innovative as they are transformed and applied in practice [22–24]. In its simplest sense, innovation is an ongoing creative process that occurs with continuous research and development [25].

Another important aspect related to innovation is the creation of a niche. It involves a genuinely trivial change

in the technology, which impacts production systems. The technical knowledge associated with this type of innovation is incremental, creating disruptive technology and opening up new market categories and applicability [26]. Thus, technological innovations and/or cutting-edge products in the cell and tissue biotechnology and bioengineering fields that directly involve plastic surgery bring plastic surgeons into this environment.

Plastic surgeons actively obtain adipose tissue in the form of lipoaspirate during liposuction, tissue block, or abdominoplasty procedures. Thus, innovations that significantly improve any physical equipment, technique, and organizational system, including information technology, hardware, and software, associated with these processes could change how plastic surgeons work, improving patient outcomes and quality of life.

The present review revealed that the most frequently performed plastic surgery procedure in the selected articles was liposuction ($n = 22$) followed by an abdominoplasty ($n = 4$). Liposuction can also be combined with abdominoplasty or other procedures for body contouring, as in a few articles [27]. This observation is not entirely unexpected because liposuction and lipoaspirate are the best sources for obtaining ADCs. Additionally, liposuction is a mechanical maneuver that dissociates the adipose stroma from the ADCs, partially breaking the extracellular matrix (ECM) that strongly binds adipocytes to each other and maintains the intimacy of these cells in this compartment. In addition to its usefulness for purely aesthetic purposes, liposuction is an essential adjuvant in reconstructive surgery, during which the collected fat is reinjected (autologous) into other regions of the body, like the breasts, buttocks, or face [28]. Plastic surgeons cannot ignore that liposuction is a mechanical dissociative method that precedes chemical and/or nonenzymatic dissociation. Thus, liposuction-related innovations represent a niche for potentially disruptive technologies in the plastic surgery field. Abdominoplasty is used to restore

Table 2 Selected studies

Study number	Reference
1	Aronowitz JA, Lockhart RA, Hakakian CS, Birnbaum ZE. Adipose stromal vascular fraction isolation: a head-to-head comparison of 4 cell separation systems #2. <i>Annals of plastic surgery</i> . 2016;77(3):354–62
2	Bi HS, Zhang C, Nie FF, Pan BL, Xiao E. Basic and clinical evidence of an alternative method to produce Vivo nanofat. <i>Chinese medical journal</i> . 2018;131(5):588–93
3	Chen J, Ren S, Duscher D, Kang Y, Liu Y, Wang C, et al. Exosomes from human adipose-derived stem cells promote sciatic nerve regeneration via optimizing Schwann cell function. <i>Journal of cellular physiology</i> . 2019;234(12):23,097–110
4	Chen L, Mou S, Li F, Zeng Y, Sun Y, Horch RE, et al. Self-assembled human adipose-derived stem cell-derived extracellular vesicle-functionalized biotin-doped polypyrrole titanium with long-term stability and potential osteoinductive ability ACS applied materials & interfaces. 2019;11(49):46,183–96
5	Güven S, Karagianni M, Schwalbe M, Schreiner S, Farhadi J, Bula S, et al. Validation of an automated procedure to isolate human adipose tissue-derived cells by using the Sepax® technology. <i>Tissue engineering Part C, Methods</i> . 2012;18(8):575–82
6	Haskett DG, Saleh KS, Lorentz KL, Josowitz AD, Luketich SK, Weinbaum JS, et al. An exploratory study on the preparation and evaluation of a “same-day” adipose stem cell-based tissue-engineered vascular graft. <i>The Journal of thoracic and cardiovascular surgery</i> . 2018;156(5):1814–22.e3
7	Hu L, Yang G, Hägg D, Sun G, Ahn JM, Jiang N, et al. IGF1 Promotes adipogenesis by a lineage bias of endogenous adipose stem/progenitor cells. <i>Stem Cells</i> . 2015;33(8):2483–95
8	Kiiskinen J, Merivaara A, Hakkarainen T, Kääriäinen M, Miettinen S, Yliperttula M, et al. Nanofibrillar cellulose wound dressing supports the growth and characteristics of human mesenchymal stem/stromal cells without cell adhesion coatings. <i>Stem cell research & therapy</i> . 2019;10(1):292
9	Kim KJ, Joe YA, Kim MK, Lee SJ, Ryu YH, Cho DW, et al. Silica nanoparticles increase human adipose tissue-derived stem cell proliferation through ERK1/2 activation. <i>International journal of nanomedicine</i> . 2015;10:2261–72
10	Krawiec JT, Liao HT, Kwan LL, D’Amore A, Weinbaum JS, Rubin JP, et al. Evaluation of the stromal vascular fraction of adipose tissue as the basis for a stem cell-based tissue-engineered vascular graft. <i>Journal of vascular surgery</i> . 2017;66(3):883–90.e1
11	Li S, Poche JN, Liu Y, Scherr T, McCann J, Forghani A, et al. Hybrid synthetic-biological hydrogel system for adipose tissue regeneration. <i>Macromolecular bioscience</i> . 2018;18(11):e1800122
12	Lin YC, Brayfield CA, Gerlach JC, Rubin JP, Marra KG. Peptide modification of polyethersulfone surfaces to improve adipose-derived stem cell adhesion. <i>Acta biomaterialia</i> . 2009;5(5):1416–24
13	McMaster R, Hoefner C, Hrynevich A, Blum C, Wiesner M, Wittmann K, et al. Tailored melt electrowritten scaffolds for the generation of sheet-like tissue constructs from multicellular spheroids. <i>Advanced healthcare materials</i> . 2019;8(7):e1801326
14	Meyers CA, Xu J, Zhang L, Asatrian G, Ding C, Yan N, et al. Early immunomodulatory effects of implanted human perivascular stromal cells during bone formation. <i>Tissue engineering Part A</i> . 2018;24(5–6):448–57
15	Meyers CA, Xu J, Asatrian G, Ding C, Shen J, Broderick K, et al. WISP-1 drives bone formation at the expense of fat formation in human perivascular stem cells. <i>Scientific reports</i> . 2018;8(1):15,618
16	Mineda K, Feng J, Ishimine H, Takada H, Doi K, Kuno S, et al. Therapeutic potential of human adipose-derived stem/stromal cell microspheroids prepared by three-dimensional culture in non-cross-linked hyaluronic acid gel. <i>Stem cells translational medicine</i> . 2015;4(12):1511–22
17	Mou S, Zhou M, Li Y, Wang J, Yuan Q, Xiao P, et al. Extracellular vesicles from human adipose-derived stem cells for the improvement of angiogenesis and fat-grafting application. <i>Plastic and reconstructive surgery</i> . 2019;144(4):869–80
18	Nyberg E, Farris A, O’Sullivan A, Rodriguez R, Grayson W. Comparison of stromal vascular fraction and passaged adipose-derived stromal/stem cells as point-of-care agents for bone regeneration. <i>Tissue engineering Part A</i> . 2019;25(21–22):1459–69
1	Park IS, Rhie JW, Kim SH. A novel three-dimensional adipose-derived stem cell cluster for vascular regeneration in ischemic tissue. <i>Cytotherapy</i> . 2014;16(4):508–22
20	Pati F, Jang J, Ha DH, Won Kim S, Rhie JW, Shim JH, et al. Printing three-dimensional tissue analogues with decellularized extracellular matrix bioink. <i>Nature communications</i> 2014;5:3935
21	Rath SN, Brandl A, Hiller D, Hoppe A, Gbureck U, Horch RE, et al. Bioactive copper-doped glass scaffolds can stimulate endothelial cells in coculture in combination with mesenchymal stem cells. <i>PloS one</i> . 2014;9(12):e113319
22	Schellenberg A, Ross R, Abagnale G, Jousen S, Schuster P, Arshi A, et al. 3D non-woven polyvinylidene fluoride scaffolds: fibre cross section and texturizing patterns have impact on growth of mesenchymal stromal cells. <i>PloS one</i> . 2014;9(4):e94353
23	Schellenberg A, Jousen S, Moser K, Hampe N, Hersch N, Hemeda H, et al. Matrix elasticity, replicative senescence and DNA methylation patterns of mesenchymal stem cells. <i>Biomaterials</i> . 2014;35(24):6351–8
24	Sesé B, Sanmartín JM, Ortega B, Matas-Palau A, Llull R. Nanofat cell aggregates: a nearly constitutive stromal cell inoculum for regenerative site-specific therapies. <i>Plastic and reconstructive surgery</i> . 2019;144(5):1079–88
25	Tang H, Husch JFA, Zhang Y, Jansen JA, Yang F, van den Beucken J. Coculture with monocytes/macrophages modulates osteogenic differentiation of adipose-derived mesenchymal stromal cells on poly(lactic-co-glycolic) acid/polycaprolactone scaffolds. <i>Journal of tissue engineering and regenerative medicine</i> . 2019;13(5):785–98

Table 2 (continued)

Study number	Reference
26	Tawonsawatruk T, West CC, Murray IR, Soo C, Péault B, Simpson AH. Adipose derived pericytes rescue fractures from a failure of healing–non-union. <i>Scientific reports</i> . 2016;6:22,779
27	Tong J, Mou S, Xiong L, Wang Z, Wang R, Weigand A, et al. Adipose-derived mesenchymal stem cells formed acinar-like structure when stimulated with breast epithelial cells in three-dimensional culture. <i>PloS one</i> . 2018;13(10):e0204077
28	Vezzani B, Gomez-Salazar M, Casamitjana J, Tremolada C, Péault B. Human adipose tissue micro-fragmentation for cell phenotyping and aecretome characterization. <i>Journal of visualized experiments: JoVE</i> . 2019(152)
29	Vindigni V, Tonello C, Lancerotto L, Abatangelo G, Cortivo R, Zavan B, et al. Preliminary report of in vitro reconstruction of a vascularized tendonlike structure: a novel application for adipose-derived stem cells. <i>Annals of plastic surgery</i> . 2013;71(6):664–70
30	Wang CC, Wang CH, Chen HC, Cherng JH, Chang SJ, Wang YW, et al. Combination of resveratrol-containing collagen with adipose stem cells for craniofacial tissue-engineering applications. <i>Int Wound J</i> . 2018;15(4):660–72
31	Wang Y, Zhao L, Hantash BM. Support of human adipose-derived mesenchymal stem cell multipotency by a poloxamer-octapeptide hybrid hydrogel. <i>Biomaterials</i> . 2010;31(19):5122–30
32	Weyand B, Israelowitz M, Kramer J, Bodmer C, Noehre M, Strauss S, et al. Three-dimensional modelling inside a differential pressure laminar flow bioreactor filled with porous media. <i>BioMed research international</i> . 2015;2015:320,280
33	Yu Z, Cai Y, Deng M, Li D, Wang X, Zheng H, et al. Fat extract promotes angiogenesis in a murine model of limb ischemia: a novel cell-free therapeutic strategy. <i>Stem cell research & therapy</i> . 2018;9(1):294

the abdomen's contour and shape, tighten the skin, correct rectus abdominis diastasis, and remove adiposity [27]. When used for obtaining ADCs, abdominoplasty produces a compact block of adipose tissue, making dissociation difficult, usually requiring an enzymatic and chemical treatment to obtain a liquefied material before starting dissociation.

Adipose tissue, like bone marrow, is derived from the mesenchyme and contains a stroma, thus making it a potential source of mesenchymal stem cells (MSC). Therefore, human lipoaspirate is an easy to isolate source of adult stem cells that exhibit stable growth and kinetic proliferation in specific culture media [1]. A previous in vitro study showed that ASCs and bone marrow MSCs can differentiate into osteogenic, adipogenic, myogenic, and chondrogenic lineages when subjected to specific lineage factors [29]. Due to the ease of collecting and processing ASCs, these cells have exciting potential in regenerative medicine. Indeed, fresh adipose tissue containing ASCs increases vascularization and graft take [5]. Moreover, ASCs have vast application potential in cell therapies and tissue engineering. For example, wound healing, reconstruction of bone defects in the calvaria, tissue regeneration with an anti-inflammatory effect, homeostasis, immunosuppression treatment in transplant-versus-host and transplant diseases, and neovascularization have all been reported following ASC administration [30]. Notably, to utilize ASCs for healing or reconstructive purposes, the adipose tissue must be processed, representing another plastic surgery niche that could produce innovative and perhaps disruptive technologies.

In the present review, we found that most of the selected studies used enzymatic dissociation, making it the most used method for adipose tissue processing and ASC isolation. However, most of the studies were conducted in the

laboratory using in vitro or in vivo experimental animals. These ASC collection protocols require enzymatically digesting (i.e., chemical dissociation) the adipose tissue to separate the cellular components of that tissue. Zuk et al. (2001) introduced chemical dissociation as the gold standard method for isolating ASCs [1]. However, the methods for this procedure typically employ collagenase type I, limiting the therapeutic potential of ASCs in humans. Collagenase type I isolated from mammalian tissue components [31] increases the risk of patient contamination because some lots of this enzyme may contain infectious pathogens (e.g., prions), thus inhibiting the immediate use of autologous ASCs in fat grafting.

The US Food and Drug Administration (FDA) has set up contrasting regulatory pathways for “human cells, tissues, or cellular or tissue-based products” (HCT/P). An HCT/P that meets specific criteria of the regulatory requirements 21 CFR 1271 Sect. 361, including being minimally manipulated, intended for homologous use only, does not involve the combination of the cells or tissues with other substances; besides, water, crystalloids, or a sterilizing, preserving, or storage agents, are considered “361” and do not require FDA approval for release. On the other hand, other HCT/Ps that do not meet the regulatory requirements are classified as “351” and must be approved as a Biologics License Application (BLA). In most cases, mechanical and nonenzymatic dissociation methods that produce SVF or ADSCs are classified as “361,” while chemical dissociation methods are typically considered “351” because of the use of collagenase to digest the tissue.

After dissociation, ASCs are then collected in a pellet, known as SVF cells. The SVF contains a heterogeneous population of cells, including ASCs, endothelial progenitor

Table 3 Brief description of the general objective and conclusion of the selected articles

Study number	Relevance	Surgery	Isolation method	Model	Study summary	Innovation
1	Devices	Liposuction	Enzymatic	In vitro	<p>Compare four enzymatic cell isolation systems: PNC MultiStation International, the LipoKit of MediKhan, the GID SVF-2 platform of GID Europe Ltd., and the StemSource 900 system/MB of Cytori Therapeutics, Inc. Processing times ranged from about 1.5 to 2 h. The Celution System yielded the highest mean number of viable nucleated cells. The Celution System also yielded significantly more endothelial cells, CD34/CD31 cells, and adipose-derived stem cells, with less residual enzyme levels. Significant variability exists concerning the recovered viable cells among systems</p>	Enzymatic dissociation device
5	Devices	Liposuction	Enzymatic	In vitro	<p>Validate a newly developed, automated procedure to isolate adipose-derived mesenchymal stem/stromal cells (ASCs) from adult human lipospi- rates in a closed and clinical-grade device, based on the Sepax® technology. Compared to the manual process, automation increased the number of nucleated cells and clonogenic progenitors per mL of lipospi- rate and attenuated yield vari- ability. The two methods produced similar cytofluorimetric profiles and in vitro differentiation capacities into mesenchymal lineages. The new Sepax-based process thus allows effi- cient isolation of ASCs with higher and more reproducible yields than the standard manual procedure, along with minimal operator intervention</p>	Automated device for ASCs isolation

Table 3 (continued)

Study number	Relevance	Surgery	Isolation method	Model	Study summary	Innovation
29	Techniques	Liposuction	x	In vitro	<p>Explored a method for differentiating ADSCs by applying an external mechanical stimulus to create a reconstructed tendon-like structure with a microcapillary network in vitro</p> <p>ADSCs seeded onto Hyalocort adhered and differentiated along the biomaterial's entire surface and infiltrated within its structure. Additionally, endothelial cells formed a capillary in the new extracellular matrix. This technique produced a vascularized tendon equivalent that could be implanted. The biodegradable hyaluronic acid-based (HYAFF-11) scaffold was shown to be suitable for autologous extracellular matrix deposition, a critical event for ADSC differentiation</p>	Vascularized tendon-like scaffolds can be developed in bioreactors
24	Techniques	Abdominoplasty liposuction	Mechanical enzymatic	In vitro	<p>Evaluated the cell yield obtained from nanofat generation and compared the results to traditional enzymatic dissociation methods. Nanofat samples exhibited a cell yield of 6.63 million cells/g lipoaspirate, whereas stromal vascular fraction (SVF) preparations resulted in only 0.68 million cells/g lipoaspirate. Cell viability was not altered in the nanofat samples compared to the SVF samples. Notably, nanofat preparations contained roughly the same number of cells (120–125 million cells) in about one-tenth the volume. Mechanical dissociation yields a better cell inoculum over conventional enzymatic dissociation methods, using less fat tissue and delivering higher cell yields</p>	Method/procedure comparison. Mechanical dissociation provided better cell yield than chemical methods

Table 3 (continued)

Study number	Relevance	Surgery	Isolation method	Model	Study summary	Innovation
28	Techniques	Liposuction	Enzymatic	In vitro	<p>Characterized a closed system for harvesting and processing human adipose tissue. Adipose tissue was collected by liposuction, washed, emulsified, rinsed, and minced mechanically into 0.3 to 0.8 mm cell clusters. The micro-fragmented adipose tissue contained small intact vessels within the adipocyte clusters, indicating that the perivascular niche is undisturbed. The clusters were enriched in perivascular cells, increasing the release of growth factors and cytokines involved in tissue repair and regeneration in vitro compared to enzymatically derived MSCs. It is plausible that the improved therapeutic potential of microfragmented adipose tissue is due to a higher frequency of presumptive MSCs and augmented secretion. This clinically approved procedure allows presumptive MSC translation without cell expansion or enzymatic treatment, simplifying the use and reducing the costs of cell-based therapies</p>	Human liposariate mechanically fragmented phenotyping and characterization
2	Techniques	Liposuction	Enzymatic and mechanical	Mixed	<p>Develop a new method to produce viable adipocytes, progenitors, and stromal stem cells using collagenase I digestion and centrifugation. Vivo nanofat. Injected Vivo nanofat and nanofat into mice. Vivo nanofat contained many colony-forming cells, expressing MSC markers and displaying multi-differentiative potential. Vivo nanofat presented greater preservation of the injected volume in the animal model</p>	Alternative dissociation method

Table 3 (continued)

Study number	Relevance	Surgery	Isolation method	Model	Study summary	Innovation
32	Computational models	Abdominoplasty	Not reported	In vitro	<p>Developed a three-dimensional computational fluid dynamics model based on a differential pressure laminar flow bioreactor prototype and evaluated its performance under different culture conditions. Cell growth inside the scaffolds was simulated by decreasing intrinsic permeability values in pressure buildup in the upper culture chamber. Pressure release allowed culturing to continue. The specific shape of the bioreactor culture vessel supported a homogenous flow profile and mass flux at the scaffold level at various scaffold permeabilities. Increased oxygen concentrations were detected inside a collagen scaffold seeded with human mesenchymal stem cells when cultured in the perfusion bioreactor compared to static culture in a Petri dish. This model's computational fluid simulation helps facilitate the design of bioreactor systems for tissue engineering applications</p>	Computational fluid simulation and bioreactors in tissue engineering

Table 3 (continued)

Study number	Relevance	Surgery	Isolation method	Model	Study summary	Innovation
25	Scaffold	Abdominoplasty	Enzymatic	In vitro	<p>Evaluated the effect of monocytes/macrophages on the osteogenic differentiation of adipose-derived mesenchymal stromal cells (ADM-SCs) in three-dimensional (3D) cocultures. THP-1 monocytes, M1 macrophages, or M2 macrophages were cocultured with ADMSCs on 3D poly(lactic-co-glycolic) acid / polycaprolactone scaffolds. ADMSC-mediated osteogenic differentiation was inhibited by monocytes and M1 and M2 macrophages in the 3D scaffolds. The secretion of oncostatin M (OSM) and bone morphogenetic protein 2 (BMP-2) was inhibited in cocultures of monocytes/macrophages and ADMSCs, and the expression of osteogenic markers alkaline phosphatase (ALP), bone sialoprotein (BSP), and runt-related transcription factor 2 (RUNX2) was downregulated. Monocytes significantly inhibited osteogenic differentiation of ADMSCs compared to cocultures with either macrophage subtype. It appears as though there are mutual interactions between monocytes/macrophages and ADMSCs that negatively affect MSC osteogenic differentiation and consequently bone healing capacity. This study highlights the importance of the microenvironment when employing cell-based constructs to treat bone defects. Resolving inflammation before treatment could potentially improve osteogenic differentiation</p>	Inflammatory environment negatively affects the drive of ASCs in osteogenic lineage
13	Scaffold	Liposuction	Not reported	In vitro	<p>Used melt electrowriting (MEW) to seed multicellular adipose-derived stromal cell (ASCs) spheroids, yielding an easy to handle, single, sheet-like tissue-scaffold construct. Poly(ε-caprolactone) processed via MEW into scaffolds was seeded with ASCs. Maintained high cell viability during a 2-week culture period. Induced ASC differentiation the adipogenic lineage</p>	MEW and spheroids/ASCs scaffolds for 3D culture

Table 3 (continued)

Study number	Relevance	Surgery	Isolation method	Model	Study summary	Innovation
20	Scaffold	Liposuction	Not reported	Mixed	Developed a method for the bioprinting of cell-laden constructs with novel decellularized extracellular matrix (dECM) bioink capable of optimizing the microenvironment conducive to the growth of three-dimensional structured tissue. Demonstrated a bioprinting process that can utilize different tissue-specific dECM bioinks (e.g., adipose, cartilage, and heart tissues) with high cell viability. We achieve high cell viability and functionality of the printed dECM structures using our bioprinting method	Bioprinting tissue and bioengineering laden cells incorporated into dME
21	Scaffold	Liposuction	Enzymatic	In vitro	Investigated the in vitro biocompatibility and bioactivity of Cu2+-doped bioglass (BG)-derived scaffolds seeded with cultured bone-marrow-derived mesenchymal stem cells (BMSCs) or a co-culture of BMSC and human dermal microvascular endothelial cells (HDMSCs). While direct osteoinduction of BMSCs was not detected, there was an increase in vascular endothelial growth factor (VEGF) expression. The scaffolds were not toxic to BMSCs. The authors found that Cu2+-doped BG scaffolds with BMSCs exhibited increased VEGF expression, endothelial markers, and typical tubular structures in culture plastics. Cu2+ stimulates BMSCs to secrete VEGF	Bioglass as a scaffold in bone tissue engineering

Table 3 (continued)

Study number	Relevance	Surgery	Isolation method	Model	Study summary	Innovation
27	Scaffold	Liposuction	Enzymatic	In vitro	Investigated the effect of mammary epithelial cells on human ASCs in 3D culture. ASCs were cultivated on matrigel using conditioned medium prepared from a human breast epithelial cell line (HBL-100). After stimulation with breast epithelial cells, the ASCs formed KRT18-positive acini-like structures and displayed upregulated expression of epithelial genes (CDH1 and KRT18) and downregulated expression of mesenchymal specific genes (CDH2 and VIM), the stemness marker (CD29), and angiogenic factors (CD31 and VEGF). Conditioned medium obtained from HBL-100 enhanced ASC migration and inhibited adipogenic differentiation. ASCs can transform into epithelial-like cells when cultured with mammary epithelial cells, suggesting ASCs positively affect lipotransfer due to growth factor secretion and their direct participation in new breast tissue formation	ASCs differentiate into epithelial cells when cultured with breast epithelial cells
3	Scaffold	Skin Flap	Enzymatic	Mixed	Investigate the effect of human ASCs derived exosomes (ASC-Exos) on peripheral nerve regeneration in vitro and in vivo. In vitro, ASC-Exos promoted Schwann cell (SC) proliferation, migration, myelination, and secretion of neurotrophic factors by upregulating gene expression after being internalized. ASC-Exos stimulated axon regeneration and myelination and restored muscle atrophy denervation in a rat sciatic nerve transection model. ASC-Exos effectively promote peripheral nerve regeneration via optimizing SC function	Treatment for regenerative medicine and nerve tissue engineering

Table 3 (continued)

Study number	Relevance	Surgery	Isolation method	Model	Study summary	Innovation
19	Scaffold	Abdominoplasty	Enzymatic	Mixed	Describe an innovative three-dimensional cell mass (3DCM) culture that is based on cell adhesion. hASCs were cultured on a substrate with immobilized fibroblast growth factor-2 to form the 3DCMs. The 3DCMs released various angiogenic factors and differentiated into vascular cells. They regenerated blood vessels and tissues regeneration and slowed limb necrosis in vivo. However, half of the mice suffered from limb loss within 28 days. 3DCM culture promotes the efficient vascular differentiation of stem cells	Paracritical effects of SVF in 3D model using innovative culture of cell mass three-dimensional (3DCM) polystyrene (PS)
30	Scaffold	x	Enzymatic Mixed	Not reported	Evaluated the effect of collagen and resveratrol (RSV) (collagen/RSV) scaffolds for their wound healing and bone regeneration potential both in vitro and in vivo. It was shown that hASCs adhere to and proliferate on collagen only and collagen/RSV scaffolds. Oral mucosal lesion experiments showed that the collagen/RSV scaffold improves wound closure and contraction compared to the collagen scaffold. In defects covered with hASCs on collagen/RSV scaffolds, regenerating bone is more visible than with hASCs on collagen scaffolds. The results suggest that collagen/RSV scaffolds could stimulate craniofacial tissue formation through biological signals	Collagen biomaterials containing RSV can be more effective than collagen scaffolds by increasing epithelial and osteogenic differentiation of ASCs

Table 3 (continued)

Study number	Relevance	Surgery	Isolation method	Model	Study summary	Innovation
17	Scaffold	Liposuction	Enzymatic	Mixed	<p>Evaluated whether adipose-derived mesenchymal stem cell-derived extracellular vesicles improved vascularization of fat grafts and increased their retention rate. Adipose-derived mesenchymal stem cell-derived extracellular vesicles were isolated from the supernatant of cultured human adipose-derived mesenchymal stem cells and incubated with human umbilical vein endothelial cells <i>in vitro</i>. The vesicles were subcutaneously co-transplanted with fat into nude mice, and the experimental group exhibited improved migration and tube formation, larger volume, fewer cysts and vacuoles, less fibrosis, and more neovessels. Adipose-derived mesenchymal stem cell-derived extracellular vesicles improve volume retention by enhancing vascularization and regulating the inflammatory response</p>	ADSC-derived EVs increase vascularization and fat graft retention
12	Scaffold	Abdominoplasty	Enzymatic	In vitro	<p>Modified polyethersulfone (PES) surfaces with fibronectin-based peptide sequences, such as Arg-Gly-Asp (RGD), Arg-Gly-Asp-Ser, and Gly-Arg-Gly-Asp-Ser and evaluated adipose-derived stem cell (ASC) adhesion. RGD-treated surfaces resulted in a higher number of attached ASCs. PES membranes modified with the RGD peptide sequence exhibit enhanced ASC attachment</p>	ASC attachment optimization in 3D cell culture models

Table 3 (continued)

Study number	Relevance	Surgery	Isolation method	Model	Study summary	Innovation
7	Scaffold	Lipectomy, Liposuction	Enzymatic	In vitro	<p>Demonstrated that CD31 (-)/34(+)/146(-) cells, a subpopulation of the stromal vascular fraction (SVF) of human adipose tissue, were robustly adipogenic. Insulin growth factor-1 (IGF1) promoted a lineage bias towards CD31 (-)/34(+)/146(-) cells at the expense of CD31 (-)/34(+)/146(+) cells. IGF1, microencapsulated in poly(lactic-co-glycolic acid) scaffolds and implanted in the inguinal fat pad of mice, induced adipogenesis <i>in vivo</i>. The authors found that the Axin2/PPARγ axis contributes to a lineage bias towards CD31 (-)/34(+)/146(-) cells during adipogenesis</p>	Therapy/treatment—Obesity/Adipose regeneration
9	Scaffold	Liposuction	Enzymatic	Mixed	<p>Examined the effect of different sized particles on growth and mitogen-activated protein kinase signaling in hADSCs. hADSCs were incubated with silica nanoparticles (NPs; <220 nm) or 3 μm silica microparticles (MPs). Silica NPs entered endocytosed vacuoles in the cytosol of hADSCs, whereas silica MPs did not. Silica NPs increased the hADSC proliferation, induced slight apoptosis, increased extracellular signal-related kinase (ERK)1/2 phosphorylation. Silica MPs increased p38 phosphorylation. Pretreatment with PD98059, a MEK inhibitor, prevented the ERK1/2 phosphorylation and silica NP-induced proliferation. Scaffolds containing silicon dioxide NPs for tissue engineering may enhance ERK1/2 activation and consequently cell growth</p>	Scaffold and ASC optimization through silicon dioxide NP

Table 3 (continued)

Study number	Relevance	Surgery	Isolation method	Model	Study summary	Innovation
23	Scaffold	Liposuction	Enzymatic	In vitro	Continuously culture-expanded MSCs on tissue culture plastic (TCP) or polydimethylsiloxane (PDMS) gels of different elasticity. The maximal number of cumulative population doublings was not affected by matrix elasticity. Differentiation towards an adipogenic and osteogenic lineage was increased on soft and rigid biomaterials, respectively. The results show that matrix elasticity influences cellular behavior, while the cells reside on the substrate	Insights into cell behavior and matrix elasticity in tissue engineering and biomaterials
22	Scaffold	Liposuction	Enzymatic	In vitro	Developed polyvinylidene fluoride (PVDF) nonwoven scaffolds with different patterns (e.g., round, trilobal, or snowflake) and fiber crimp patterns (e.g., 10, 16, or 28 needles per inch). Seeded the scaffolds with adipose tissue-derived human mesenchymal stromal cells (MSCs). Initial cell adhesion was increased using nonwoven scaffolds with round fibers. All non-woven PVDF scaffolds facilitated cell growth. Proliferation was enhanced on nonwoven, round, or trilobal fiber scaffolds. In general, proliferation increased in broader, less dense networks. MSCs aligned along the fibers and formed cellular layers spanning over the pores. The 3D PVDF nonwoven scaffolds support MSC growth	Demonstrated that scaffold properties (e.g., fiber morphology and mesh size) are relevant to cell adhesion and proliferation
11	Scaffold	Not reported	Not reported	Mixed	Incorporated varying concentrations of decellularized adipose tissue, with a thiol-acrylate fraction that is polymerized to produce hydrogels via a Michael addition reaction. Collagen I is the major component of the isolated adipose-derived extracellular matrix (ECM). The mechanical properties of ECM polyethylene glycol (PEG) are not negatively affected by ECM incorporation. Human adipose-derived stem cells (hASCs) encapsulated in the ECM PEG hydrogel with varying ECM concentrations showed that 1% ECM PEG hydrogel maintained the highest viability and proliferation rate. Enhanced adipose regeneration was also associated with 1% ECM PEG in vivo	Adipose tissue regeneration applications through hydrogel scaffolds

Table 3 (continued)

Study number	Relevance	Surgery	Isolation method	Model	Study summary	Innovation
31	Scaffold	Liposuction	x	x	<p>Characterized a hybrid hydrogel scaffold composed of poloxamer 407 (PO) and the self-assembling oligopeptide EFK8 in vitro and in vivo. The storage modulus of EFK8-PO increased compared to that of EFK8 alone, indicating that EFK8-PO integrates PO's mechanical strength and integrity with the bioactivity of EFK8. Culturing human adipose-derived mesenchymal stem cells (hAMSCs) in PO resulted in severe aggregation, but almost no aggregation was observed in EFK8 or EFK8-PO. The viability of hAMSC in the three hydrogels remained above 80% after 2 weeks of culturing. Moreover, EFK8 and EFK8-PO significantly increased hAMSC proliferation rates differentiated into adipocytes or osteoblasts, suggesting EFK8 supports hAMSC multipotency in vitro. Only EFK8-PO supported hAMSC engraftment and adipogenic differentiation posttransplantation in a mouse model. This study showed that EFK8-PO improved mechanical properties and bioactivity, representing a potentially valuable stem cell scaffold for soft tissue engineering</p>	<p>EFK8-PO supports the promising potential to be used as a stem cell framework in soft tissue engineering</p>
8	Scaffold	Not reported	Enzymatic	In vitro	<p>Evaluate the potential use of wood-derived nanofibrillar cellulose (NFC) wound dressing as a cell scaffold material for hASCs and develop a cell transplantation method free from animal-derived components for wound healing. hASCs adhered to the NFC dressing, surviving for 2 weeks without cell adhesion coatings or adverse effects</p>	<p>Wound healing dressing incorporating ASCs</p>

Table 3 (continued)

Study number	Relevance	Surgery	Isolation method	Model	Study summary	Innovation
18	Scaffold_Materials (SVF vs. ASC)	Liposuction	Enzymatic	Mixed	<p>Compared the bone-forming potential of freshly isolated and passaged stromal vascular fraction (SVF) cells obtained from human liposyrate. Isogenic SVF and ASCs were suspended in fibrin hydrogels and seeded in three-dimensional-printed osteoinductive scaffolds of decellularized bone matrix and polycaprolactone. In vitro, both cell populations successfully mineralized the scaffold, revealing the bone formation properties of SVF. Both cell-laden interventions resulted in improved bone healing.</p>	<p>3D printed scaffold An SVF and ASCs minimally processed in bone regeneration</p>
16	Scaffold	Liposuction	Enzymatic	In vitro	<p>Compared spheroids prepared with cultured human adipose-derived stem/stromal cells (hASCs) in a non-cross-linked hyaluronic acid (HA) gel, dissociated hASCs and hASC spheroids prepared using a nonadherent dish. Showed that 4% HA gel effectively formed hASC spheroids with a relatively consistent size. Spheroids were positive for pluripotency markers, and nearly half were SSEA-3-positive, a marker of the multilineage differentiating stress enduring or Muse cell. Dissociated ASCs displayed increased cytokine excretion (e.g., hepatocyte growth factor) when cultured under hypoxia. ASC spheroids also exhibited upregulation of some pluripotency markers and downregulation of genes related to the mitotic cell cycle. After an ischemia–reperfusion injury to the fat pad in SCID mice, local injection of hASC spheroids promoted tissue repair and reduced the atrophy compared to dissociated hASCs or vehicle. Some administered hASCs differentiated into vascular endothelial cells. ASC spheroids prepared in a HA gel contain undifferentiated cells that can promote angiogenesis and tissue regeneration after damage</p>	<p>HA as a promising material for 3D cell culture and vehicle for stem cell injectable therapy</p>

Table 3 (continued)

Study number	Relevance	Surgery	Isolation method	Model	Study summary	Innovation
6	Scaffold_Vascular Grafts	Liposuction abdominoplasty panniculectomy	Enzymatic	Mixed	Determine if stromal vascular fraction (SVF) seeded tissue-engineered vascular grafts (TEVG) remain patent and remodel, allowing for “same-day” TEVG fabrication and implantation. SVF was seeded onto poly(ester urethane)urea bilayered scaffolds using a customized rotational vacuum seeding device and then surgically implanted as abdominal aortic interposition grafts in rats. Patency was observed in 5/7 implanted scaffolds after 8 weeks, with neotissue formation and remodeling also being detected	Bioengineering incorporating ASCs into vascular scaffolds
10	Scaffold	Liposuction	Enzymatic	Mixed	Evaluated the use of the raw stromal vascular fraction (SVF) obtained after digestion of human liposuction aspirates for tissue-engineered vascular grafts (TEVGs). Human SVF cells performed the same functions as AD-MSCs, including differentiating into SMCs and secreting promigratory factors. The SVF cells were seeded into a biodegradable, elastomeric, porous scaffold that produces patent TEVGs populated with primary vascular components	Bioengineer stem cell-based vascular graft improvement

Table 3 (continued)

Study number	Relevance	Surgery	Isolation method	Model	Study summary	Innovation
14	Therapy/Treatment	Liposuction	Not reported	Mixed	Examined differences in the early innate immune response to human perivascular stem/stromal cells (PSCs) or unpurified stromal vascular fraction (SVF) during <i>in vivo</i> bone formation. Showed significantly greater neutrophilic and macrophage infiltrates within and around SVF implants. Differences in early postoperative inflammation among SVF-laden implants were associated with reduced osteogenic differentiation and bone formation. Similar findings were recapitulated with PSC implantation in immunocompetent mice. Exaggerated postoperative inflammation was associated with increased IL-1 α , IL-1 β , IFN- γ , and TNF- α gene expression among SVF samples and conversely increased IL-6 and IL-10 expression among PSC samples. These data document a robust immunomodulatory effect of implanted PSC and an inverse correlation between host inflammatory cell infiltration and stromal progenitor cell-mediated ossification	Immunomodulatory effect of SVF on bone healing
15	Therapy treatment	Liposuction	Not reported	Mixed	Observed high WISP-1 expression in human perivascular stem/stromal cells (PSCs) after purification and upon transplantation to a bone microenvironment <i>in vivo</i> . <i>In vitro</i> results showed that WISP-1 displays pro-osteogenic/anti-adipocytic effects in human PSCs, and BMP signaling activity regulation may modulate these effects. Thus, WISP-1 contributes to the regulation of human stem cell differentiation within the perivascular niche	Therapy/treatment ASCs signaling optimizing bone healing

Table 3 (continued)

Study number	Relevance	Surgery	Isolation method	Model	Study summary	Innovation
4	Therapy/treatment	Liposuction	Enzymatic	Mixed	Evaluate biofunction of self-assembling biotinylated mesenchymal stem cell-derived extracellular vesicles (MSC-EVs) immobilized onto the surface of biotin-doped polypyrrole titanium (Bio-Ppy-Ti) in vitro and in vivo. This method increased the amount of human adipose-derived stem cell-EVs (hASC-EVs) anchored onto the Bio-Ppy-Ti surface, remaining stable for 14 days at 4 °C. EV-Bio-Ppy-Ti increased cell compatibility and osteoinductivity for osteoblasts in vitro and displayed anti-apoptosis potential. Several osteoinductive microRNAs (miRNAs) were encapsulated in hASC-EVs, possibly accounting for the enhanced bone regeneration of EV-Bio-Ppy-Ti. Thus, this MSC-EV biotin-immobilized method appears to be highly efficient and has long-term stability for bone graft bioactive modification	MSC -EVs and titanium/biotin complex scaffolds. Application for osteogenic differentiation
26	Therapy/treatment	Liposuction	Enzymatic	In vitro	Tested the ability of human bone marrow-derived mesenchymal stem cells (MSC) and human adipose-derived pericytes (the native ancestor of the MSC) to prevent the formation of atrophic nonunion in rats. Animals in the cell treatment groups displayed augmented bone healing compared to the control group. Radiographic parameters were significantly improved, and bone bridges at the fracture gap were observed in the MSC – and pericyte-treated groups. Bone quality and its biomechanical properties were enhanced in both treatment groups. MSC and pericytes stimulate bone regeneration potential in an atrophic non-union model. These cells may help prevent atrophic nonunions, representing an alternative treatment of fractures that can potentially develop nonunion	Promising therapy with pericytes preventing and treating atrophic nonunion

Table 3 (continued)

Study number	Relevance	Surgery	Isolation method	Model	Study summary	Innovation
33	Therapy/treatment_fat extract	Liposuction	Mechanical	Mixed	Produced an aqueous cell-free extract directly from human fat tissue [fat extract (FE)] and evaluated its therapeutic efficacy using the ischemic hindlimb model of nude mice. FE was prepared and characterized, and intramuscular injection reduced severe limb loss and increased the ischemic tissue's blood flow and capillary density. The FE contained high levels of various growth factors. Interestingly, when added as a cell culture supplement, FE promoted HUVEC proliferation, migration, and tube formation in a dose-dependent manner. Additionally, subcutaneous injections of Matrigel infused with FE improved vascular formation. The results suggest that FE could be used to treat ischemic disorders	New cell-free therapeutic agent FE, as a bio-product from the human lipospi-rate using simply physical methods, showing potential as a therapeutic agent for ischemic disorders

cells, growth factor-producing cells, leukocytes, red blood cells (RBCs), macrophages, fibroblasts, and vascular smooth muscle cells [29]. Thus, innovations that provide alternatives or improvements to existing protocols for isolating these cells for human use would significantly impact the research and development (R&D) in the regeneration field and be of great interest to plastic surgeons.

As an alternative to chemical dissociation, mechanical methods, including topics related to the isolation of MSCs within the operating room, were reported publicly from 2006 to 2011. These methods rely on subjecting the tissue to disruptive dissociative forces, resulting in the release of adipocytes and ADCs from the ECM without the need for collagenase, minimizing contamination risks and providing a safer alternative to chemical dissociation, all within the operating room setting.

This protocol described by Lamblet is divided into two parts: shear force and pellet collection. The shear force portion involves collecting the lipoaspirated tissue in 50 mL syringes and subjecting it to mechanical dissociation via a Luer-Lock transpacer (Grams Medical, Costa Mesa, CA, USA) that provides a 0.5-inch channel firmly connected to another empty 50-mL syringe, allowing the connection between the two lumens of the syringes and facilitating material collection. The lipoaspirated tissue is subjected to shear forces that liberate the ADCs from the adipocytes and ECM from the tissue cluster when passed through the straight channel between the two syringes. The final product contains digested adipocytes, ADCs, and connective tissue that could be applied directly to the patient for regenerative purposes. In tissue augmentation, this product could be incorporated into the fresh lipoaspirate to be grafted, increasing retention.

The second part of the protocol involves collecting the mechanically digested material into a pellet to isolate all the ADCs. The digested fat is washed with PBS and then centrifuged at $1100 \times g$. The infranatant is collected and washed one more time with PBS and centrifuged a second time at $1100 \times g$. The infranatant is collected and passed through a 100-nm filter that retains cells and debris greater than this diameter. The filtrate is collected and centrifuged a third time. The button of the centrifuge tube contains a pellet with all ADCs, including RBCs. At this point, it should be pointed out that this product could be used in patients by incorporating this pellet into the fresh adipose tissue to be grafted for autologous fat tissue augmentation purposes. Alternatively, ASCs can be isolated from this heterogeneous mixture of cells using a 100-nm filter followed by a 20-nm filter and RBC lysis buffer, as in the chemical dissociative method, and following the appropriate sorting and expansion protocols. However, despite not using collagenase in this final process, the presence of the RBC lysis buffer limits the use of this product to in vitro or in vivo laboratory animal studies.

Notably, the author presented the shear force part of the protocol at conferences since 2006 but was published and referred to as nanofat by different authors [32]. However, the nanofat study only described the mechanical part of this process and never developed the second part (Pellet collection), which was never presented publicly. This omission consequently generated confusion in the field, especially since the author did not disclose Lamblet's protocol or that this was an alternative mechanical method to chemical dissociation, probably in an attempt to not correlate it with the protocol described above.

Since then, few protocols using mechanical dissociation for obtaining and isolating cells have been established compared to chemical methods [28]. From Romanov et al. 2005 to Gentile et al. 2015, the alternative methods to chemical dissociation were based on washing, vibration, shaking and filtration, or combined with centrifugation and were not even mentioned as mechanical methods. This approach is sufficient for isolating some progenitor cells but is not adequate when comparing the results to chemical methods.

Of the 33 articles identified in this review, only one used only mechanical dissociation [33], and three used a combination of mechanical and enzymatic dissociation techniques [31, 32, 34]. The lack of nonenzymatic methods and their previous credibility is partially attributed to nonenzymatic dissociation processes yielding significantly fewer nucleated cells than chemical methods [7, 11]. These methods generally involve washing and agitating the liposuction product or combining these processes followed by filtration or centrifugation to concentrate the SVF ADCs. It has been reported that the composition of cell populations recovered by simple centrifugation and other nonenzymatic methods contains higher amounts of peripheral blood mononuclear cells (PBMCs) and lower amounts of progenitor cells than obtained with chemical dissociation [7, 9, 10]. It has been suggested that ASCs are concentrated in small and medium adipose tissue vascular structures. Without enzymatic degradation of the ECM, many progenitor cells remain trapped in the vascular endothelial layers and connective tissue fragments of the liposuction product.

Indeed, immunohistochemical and immunofluorescent experiments revealed that ASCs and pericytes are primarily located in perivascular niches [35, 36]. Since there is no ECM disruption, some nonenzymatic isolation methods fail to release the progenitor cells from the perivascular spaces, leaving many desired cells trapped in larger tissue fragments and eventually discarded. Notably, it has been reported that reduced progenitor cell populations and CD34+ expression are the contributing factors for cultures requiring longer incubation times to reach 80–90% confluence [9]. Furthermore, these nonenzymatic dissociation studies are based on one or two methods using an apparatus or laboratory device used separately, without synergy or considering the intimacy

between the ADCs and collected adipose tissue in human lipoaspirate (HLA).

The HLA comprises fat tissue clusters (FTCs), and ADCs are released during the liposuction maneuver, a mechanical dissociative force. The FTCs contain adipose tissue matrix, adipose cells, mature adipocytes, blood vessels, smooth vascular cells, and trapped ADCs, including adult stem cells (ASCs), growth factor producing cells (GFPC), endothelial progenitor cells, lymphocytes, macrophages, red blood cells (RBCs), fibroblasts, plasma, and derivatives.

Unlike chemical dissociation, the beauty of mechanical dissociative force, as in Lamblet's protocol, is that when you apply shear stress forces to the FTC, it releases and at the same time retains substantial amounts of cytokines and GFPCs in the dissociated tissue, which are subsequently grafted along with the ADCs and fresh collected tissue, which is essential for the graft to take in vivo. Interestingly, when grafted together, these released cells create an environment that promotes repair and regeneration through multiple mechanisms, known as the immunomodulatory response [30]. Notably, in the last year, several clinical trials evaluating the efficacy of ADCs as a COVID-19 therapy have also emerged.

It is important to point out that TGF- β has been shown to stimulate collagen production, and vitamins B1, B2, and B3 stimulate fibroblast collagen and glycosaminoglycan production, increase fibronectin synthesis, inhibit matrix degradation, and facilitate cell chemotaxis [30]. Additionally, PDGF stimulates collagenase glycosaminoglycan fibroblast production, angiogenesis, and wound contraction and facilitates cell chemotaxis. GM-CSF improves white blood cell function, activates neutrophils, eosinophils, and monocytes/macrophages, and stimulates the proliferation and differentiation of hematopoietic cell lines [30]. Furthermore, it is well-known that interleukins (ILs) regulate cell homeostasis. For example, IL-3 and IL-6 stimulate the proliferation and differentiation of hematopoietic cell lines, and IL-7 and IL-8 augment neutrophil functions, act as anti-inflammatory agents and stimulate wound healing [30]. These effects are easily detected by performing a complete blood count, platelets, and coagulogram 1 week after a fat graft is performed. However, the cells cannot be washed or separated after the mechanical dissociative process, as publicly described by Lamblet in 2011.

Expanding a single-cell type requires first isolating, characterizing, and expanding it in a specific culture media. On the other hand, cells and cell aggregates produced with mechanical dissociation do not need to be isolated. Thus, if we use a mechanical maneuver that pays attention to the breakdown of the ECM and the release of this strong adhesion between adipocytes and the ADCs, a final count of nucleated cells is very similar or even higher than chemical dissociation will likely yield. Notably, the

therapeutic potential has been reported to be optimized when cell conglomerates and/or aggregates are transferred together. When discussing cell conglomerates, we refer to all the cells in the collected tissue, including RBCs, sometimes considered contaminants in ADC transfer. Indeed, these cells are contaminants when attempting to isolate a single cell (i.e., ASCs) in the laboratory. However, when transferring the pool of cells, the RBCs can contribute to wound healing and, in some cases, compensate for blood loss in more extensive liposuction cases, where the RBCs are transferred to the third space, functioning as an auto hemotransfusion process. We hope that our patent-pending surgical device will offer the convenience and yield required to make this process a reality.

As mentioned, 25 articles employed liposuction as the plastic surgery technique, and 23 used enzymatic dissociation methods to isolate ASCs. Moreover, articles describing new devices ($n = 2$), techniques ($n = 4$), computational models ($n = 1$), tissue scaffolds ($n = 21$), and therapies and/or treatments ($n = 5$) were major areas of research and development identified in the present study. In the following paragraphs, these innovative studies and their possible clinical applicability are described. The potential applications of these inventions in terms of ASC procedures employed during plastic surgery are also discussed. While six articles did not report the cell isolation method (enzymatic or mechanical), innovation-related observations and possible clinical applicability are described. A summary of the characteristics and relevance of these studies are presented in Table 3. We will begin with innovative devices designed for ASC isolation.

Devices

Aronowitz et al. (2016) compared the performance of four SVF cell isolation systems [11]. The authors found that despite the innovation and technology of these devices, their clinical applicability still bumps into the regulatory barrier with collagenase (see above). Furthermore, the cost of the devices and associated disposables makes their large-scale clinical applicability unfeasible.

Güven et al. (2012) developed an automated procedure to isolate ASCs from adult human liposuctions in a closed clinical-grade device based on Sepax technology [37]. However, the Sepax system does not do all the processing in a closed system since the digestion is conducted outside the device with an adapted kit. Although the device's value should be lower compared to existing ones, the cost–benefit was not declared. In addition to devices, our search also identified alternative techniques that could improve downstream applications using ASCs.

Techniques

For example, in a different type of study, Vindigni et al. (2013) combined the great facility of ASCs to differentiate by applying an external mechanical stimulus that successfully creates a tendon-like structure reconstructed in vitro with a microcapillary network [38]. This study shows that physical factors influence the activity of ASCs, and mechanical stimuli, generated in vitro by bioreactors, can produce a transplantable vascularized tendon, representing an inexhaustible source of possibilities in tissue repair clinically.

Sesé et al. (2019) evaluated cell yields obtained from the generation of nanofat particles compared to traditional enzymatic dissociation methods [39]. Comparing the two methods showed that mechanical disaggregation provided a better cell inoculum than conventional enzymatic dissociation methods, using ten times less adipose tissue as starting material and providing a higher cell yield. The technology and products developed by Tulip Medical allow researchers and medical doctors to mechanically break down the adipose tissue into small fat particles, known as nanofat, immediately after the fat tissue is removed from the patient. The study compared chemical dissociation with mechanical but had two significant limitations. The first was that the authors used different fat tissue collection processes for each dissociation method. For more reliable and acceptable results, the same collection procedure should have been performed. The second concerns mechanical digestion, which discarded fluids after the liposuction process, eliminating a substantial percentage of the cell precipitate.

Vezzani et al. (2019) [34] performed cellular phenotyping and characterized mechanically fragmented human liposuction material. The authors found that mechanical dissociation of liposuctions resulted in the production of micro-fragmented adipose tissue (MAT), consisting of adipocyte aggregates with a microvascular network. They found that the mechanical micro-fragmentation process did not affect the perivascular cell compartment due to the presence of perivascular cells, both pericytes and adventitia, which were confirmed by flow cytometry. Furthermore, cytokines and angiogenic factors produced by both MAT and the SVF were more abundant in the MAT supernatants. Consequently, MAT digested with collagenase and placed in culture produced a secretome similar to conventional SVF. These results demonstrate that MAT is therapeutically beneficial and amenable to phenotypic and functional investigations. Additionally, the small size of the MAT clusters allows researchers to measure culture secretory activity, which is more challenging with larger adipose tissue pieces. It is important to point out that identifying specific cells in the fragmented lipoaspirate promoted the development of this innovative technology.

The study of Bi et al. (2018) compared mechanical dissociation (i.e., nanofat) with slight chemical dissociation (0.2 mg/mL collagenase type I for 15 min), referred to as *Vivo* nanofat [40]. The size of the transplanted nanofat graft was smaller than *Vivo* nanofat. The authors also observed that the number of viable adipocytes, colony formation, and MSCs expression was more remarkable in *Vivo* nanofat. However, the authors reduced the collagenase concentration and incubation period (0.2 mg/ml for 15 min) for the chemical dissociation compared to standard methods for isolating MSCs from adipose tissue (0.075% for 30 min). Notably, the experiments should have been limited to the animal model and not used in humans since even small concentrations of collagenase could harm the patient. Moreover, only the supernatant is collected after digestion, and the infranatant is apparently discarded. As previously mentioned, ASCs are concentrated in the SVF, which is the infranatant following enzymatic digestion.

Computational model

Our search also identified a 3D computational fluid dynamics model based on a differential pressure laminar flow bioreactor prototype developed to examine the performance of constant changes in the culture environment [41]. This model characterizes the flow and pressure distribution within a perfusion bioreactor prototype, uniting biotechnology with computational Big Data, resulting in more precise control of the culture media. Next, we will discuss articles that reported innovative approaches for incorporating ASCs into tissue scaffolds, a theme observed in most of the selected articles.

Tissue scaffolds

In the study by Tang et al. (2019), the effect of monocytes/macrophages on the osteogenic differentiation of adipose tissue-derived mesenchymal stromal cells (MSCs) in three dimensional (3D) cocultures was evaluated [42]. The authors showed that monocytes and macrophage subtypes inhibit the osteogenic differentiation of ASCs in 3D PLGA/PCL structures. More specifically, the cocultured monocytes/macrophages decreased the expression of osteogenic markers such as ALP, BSP, and RUNX2. These results highlight the overlooked fact that inflammation can regulate osteoblasts from MSC-based bone constructs within the bone microenvironment. In this sense, the tight control of inflammation may be necessary to create an anabolic environment and improve cell-based bone construct performance.

McMaster et al. (2019) demonstrated that melt electrowriting (MEW) could be adapted for seeding multicellular spheroids [43]. The authors manufactured this scaffold in sheet form and produced spheroids containing 1000–2000 ASC aggregates inside each pore. The cultures are easy

to handle and can be transferred to other sites for mixed implants containing living organic and inorganic elements, augmenting implant grip and adaptation. This purely innovative and applied technology was due to the utility of 3D printers and the new electro-engraving by fusion technique.

In the study by Pati et al. (2014), the authors developed a method of biological imprinting with laden cells incorporated into a decellularized extracellular matrix (dECM) [44]. The bioink provides a favorable microenvironment for 3D tissue growth. The ability to print analog tissue structures by providing living cells with the appropriate material in a defined and organized way, in the right place, in sufficient numbers, and in the right environment, is critical for many emerging technologies. The concept of tissue and organ printing or bioprinting is performed in a liquid medium, making it useful for *in vivo* tissue engineering experiments and *in vitro* experiments with drugs and tissue models and tumor growth.

In the study by Rath et al. (2016), stem cells from ASCs or bone marrow MSCs were tested for their ability to differentiate into highly porous 3D 45S5 Bioglass® scaffolds [45]. Interestingly, after five passages, ASCs differentiate into a bioactive glass, even without any means of differentiation. This technology, based on Bioglass scaffolds, opens up new possibilities for use in bone tissue engineering.

The study by Tong et al. (2018) was the first to evaluate the effect of breast epithelial cells on human ASCs in 3D culture [46]. This study revealed that ASCs form structures similar to acinar cells and exhibit characteristics of epithelial differentiation when stimulated by the HBL-100 mammary epithelial cell lineage in 3D. In the clinical context, the findings show that ASC characteristics are beneficial for cell-assisted lipotransfer for breast reconstruction since they can promote mammary gland growth. However, care must be taken when ASCs are cultured and expanded *in vitro* and then transplanted because little is known about the interactions between exogenous ASC and the breast epithelium.

An innovative 3D cell mass (3DCM) based on cell adhesion was described by Park et al. (2014). This study evaluated the therapeutic potential of 3DCMs composed of ASCs [47]. The 3DCM culture promoted efficient vascular stem cell differentiation. Additionally, 3DCM transplantation resulted in direct vascular regeneration of the injected cells and improved therapeutic efficacy. The authors also showed that 3D cell aggregates prevent cell apoptosis and promote cell stabilization. In ischemia models, stem cell spheroids improved therapeutic efficacy through enhanced cell viability and paracrine effects.

Other tissue scaffold-related articles described specific supplements that could serve as therapies or improve surgical outcomes. For example, the effects of exosomes derived from human ASC (ASC-Exos) on the regeneration of peripheral nerves *in vitro* and *in vivo* were investigated

by Chen et al. (2019) [48]. The authors observed that human ASC-Exos promoted the axonal growth of neurons. This study represents a potential treatment for nerve regeneration and nerve tissue engineering.

In the study by Wang et al. (2018), collagen scaffolds and ASCs were combined for bone regeneration of the oral and calvaria mucosa using resveratrol (RSV) [49], which affects the differentiation of mesenchymal stem cells. The experiment proves to be innovative since it aimed to identify active substances like RSV that can activate the cellular potential of ASCs.

The study by Mou et al. (2019) showed that extracellular vesicles from adipose tissue-derived MSCs improve the vascularization of fat grafts and increase their retention rate [50]. The innovative aspect of this study is that the authors used adipose tissue MSCs and the extracellular vesicles, a by-product of the cell culture. Furthermore, using tissue engineering in which a Matrigel scaffold was colonized with umbilical vein endothelial cells supplemented with extracellular vesicles, the authors evaluated the angiogenic retention regenerative potentials of the transplanted adipose tissue. This complex but simultaneously simple and innovative process has great potential for clinical applicability.

In the study by Lin et al. (2009), it was hypothesized that polyethersulfone (PES) surfaces modified with the fibronectin-based Arg-Gly-Asp (RGD) peptide sequence would increase ASC adhesion compared to the unmodified cells [51]. By evaluating ASC fixation and proliferation on PES surfaces modified with different fibronectin-derived peptide sequences and using cutting-edge biotechnology to evaluate RGD-treated surfaces to attach a more significant number of ASCs, the authors were able to develop a model that bridges bioreactor technology to *in vitro* 3D cell cultures and prospective studies of 3D tissues with fluid mechanics.

Hu et al. (2015) microencapsulated insulin growth factor-1 (IGF1) in poly(lactic-co-glycolic acid) scaffolds and implanted them into mice. The authors found that IGF1 positively regulates Axin2 and PPAR α and simultaneously attenuates Wnt/b-catenin under adipogenic conditions, promoting a lineage bias towards CD31(-)/34(+)/146(-) cells at the expense of CD31(-)/34(+)/146(+) cells and leading to adipogenesis *in vitro* and *in vivo* [52]. This innovative study has direct implications related to obesity and adipose regeneration.

In the study by Kim et al. (2015), the effect of different sized silicon dioxide particles on the growth of and mitogen-activated protein kinase signaling in ASCs was verified [53]. This study demonstrated that only nanoparticles (NPs) with a 50–120 nm diameter are beneficial, while microparticles can induce apoptosis. Silica-based scaffolds and coated plates have hydrophilic properties, and it was also shown that only silica NPs entered cells, and some were clustered

in vesicles, suggesting that silica NPs enter cells partly by endocytosis.

The effect of continuously expanding MSCs in parallel culture on tissue culture plastic (TCP) or polydimethylsiloxane gels (PDMS) of different elasticity was reported by Schellenberg et al. (2014) [54]. In this study, the authors showed that PDMS elasticity had no sustained effect on replicative senescence, intrinsic cell lineage impairment, or mRNA profiles of cells continuously cultured on these substrates.

Schellenberg et al. (2014) developed scaffolds of polyvinylidene fluoride (PVDF) non-tissue with round, trilobal, or snowflake fiber cross section and different fiber crimp patterns (10, 16, or 28 needles per inch) to evaluate the effect of the biomaterial's 3D scaffold architecture on cell behavior and fate [55]. Due to their complex 3D conformation, they can provide interesting perspectives for some surgical interventions. For example, non-tissue PVDF scaffolds can be used as meshes in hernia repair. Notably, MSCs mediate and promote the wound healing process, supporting vascularization and differentiation into many cell types. The *in vitro* study of interactions between PVDF and MSCs could stimulate implant cellularization *in vivo*. The biotechnology used to fabricate non-tissue scaffolds with polymeric PVDF granules involves a completely labor-intensive and innovative method with three main processing steps: texturing, web forming, and gluing. It is plausible that additional innovations could be developed specifically for each step.

Li et al. (2018) decellularized adipose tissue and combined it, in variable concentrations, with a fraction of thiol-acrylate to produce hydrogels [56]. Incorporating bioactive molecules into a hydrogel system can increase the cell proliferation rate and improve the adipogenic differentiation performance of stromal stem cells. However, this approach suffers from high costs and can also cause cytotoxicity due to the light/heat used for curing (i.e., polymerization). The innovation associated with this study comes from applying polyethylene glycol (PEG) MEC without modifying it and encapsulating ASCs at various hydrogel/PEG/MEC concentrations. The authors reported that 1% is the ideal concentration for stimulating adipogenesis. Bioengineering and biotechnology were fundamental for fabricating ECM PEG hydrogel scaffolds and the encapsulation of the human ASCs.

In the study by Wang et al. (2010), the characteristics of a hybrid hydrogel framework composed of poloxamer 407 and the self-assembly oligopeptide EFK8 *in vitro* and *in vivo* were examined [57]. The hybrid hydrogel containing ASCs has great promise as a 3D scaffold for stem cell-based soft tissue engineering.

Interestingly, one article reported the identification of a new scaffolding material. Kiiskinen et al. (2019) demonstrated the potential use of wood-derived nanofibrillar

cellulose (NFC) as a wound dressing and cell scaffolding material for ASCs. The authors developed an animal-derived component-free cell transplantation method for wound care [58]. This study proved to be innovative in developing a biotechnological dressing that allowed the adhesion of MSCs and did not interfere with their cell viability and function.

Nyberg et al. (2019) explored the bone formation potential of clinically relevant SVF cells obtained from human liposuction samples [59]. This study harvested the SVF, combined it with 3D-printed osteoinductive supports, and implanted it in the bone defect to stimulate regeneration in the same surgical procedure. This process reduces the number of operations required for the patient and eliminates the cost and practical limitations of handling *ex vivo* cells. Moreover, it leverages the regulatory feasibility of an intraoperative versus multi-operative procedure and enables administering the SVF intraoperatively.

Mineda et al. (2015) showed the therapeutic potential of stromal/stem cell microspheroids derived from human adipose tissue prepared by 3D culture in non-cross-linked hyaluronic acid gel [60]. The delivery system from cells to organs and tissues is promising, and the involved technology and subsequent innovation have been continuously improved. Furthermore, hyaluronic acid, which is already being used on a large scale in plastic surgery and other specialties as fillers, is a perfect candidate for this purpose.

In the study by Haskett et al. (2018), the tissue-engineered vascular graft (TEVG) models were seeded with SVF for 8 weeks *in vivo* [61]. The approach shows that bioengineering combined with surgery and methods for obtaining and isolating SVF from adipose tissue allows its later use in the laboratory and immediate use *in vivo*.

Krawiec et al. (2017) evaluated the use of the SVF from adipose tissue as a vascular graft developed based on stem cells in producing a urethane polyester scaffold that had been previously shown to be effective with other cell types [33]. This study demonstrated the possibility of using freshly obtained stromal SVFs for seeding polyester urethane scaffolds rather than adipose tissue-derived stem cells that required culturing for 2–3 days. It brought innovation, demonstrated clinical applicability of a tissue bioengineering technology, and opened the door to new studies in vascular graft fabrication in tissue engineering with these polyester urethane scaffolds.

Therapies/treatments

Beyond describing and discussing some innovative and exciting emerging technologies in the field, other articles provided insights into novel therapies and treatments. For example, the immunomodulatory effect of purified human perivascular stromal cells (PSCs) implanted during bone formation was reported by Meyers et al. (2018) [34]. The study

demonstrated that differentiated cells from the perivascular region of the adipose stroma have a different phenotypic differentiation from the total cells of the SVF. These observations are particularly relevant to bone induction and differentiation and open up several lines of research investigating PSC versus SVF bone differentiation.

In another study by Meyers et al. (2018), high WISP-1 expression was observed in human PSCs *in vivo* after purification and transplantation into a bone defect [62]. Previously, WISP-1 was defined for its role as a bone matrix protein and its upregulation of osteoblastogenic differentiation into other types of osteoblastic cells. However, this study demonstrated a new role for WISP-1 signaling in stromal progenitor cells in the perivascular niche of human adipose tissue. Moreover, the authors implicated WISP-1 in promoting angiogenesis, especially tumor-associated angiogenesis. This study complemented the previous one and built upon it by showing the relationship of perivascular adipose stromal cells with the WISP-1 activity of the osteogenic marker, implicating these cells as progenitors of osteogenic activity and opening up several research paths with these differentiated cells that do not need expansion.

In the study by Chen et al. (2019), a new method which is immobilization method by self-assembly of biotinylated MSC-EVs on the surface of biotin-doped polypyrrole titanium (Bio-Ppy-Ti) was reported to improve *in vitro* and *in vivo* biofunction that could be applied to bone regeneration [63]. In this study, human fat ASC-derived exosomes were stably anchored in a scaffold produced by the electropolymerization of a biotin-doped polypyrrole film on titanium EV-Bio-Ppy-Ti and incorporated into the differentiation process of cultured osteoblasts. The authors assessed osteoinduction and found it to be 185 times greater than the control cells [48]. This technology is particularly relevant to individuals requiring metallic titanium implants.

Tawonsawatruk et al. (2016) investigated the capacity of human bone marrow-derived MSCs and human adipose tissue-derived pericytes percutaneously administered to the bone fracture gap to prevent pseudarthrosis [64]. In this study, innovation and technology came together to create a method for measuring bone healing of non-atrophic fracture union, from mathematical methods for measuring bone callus to the use of Micro-CT to assess bone density and thickness of trabeculae, histological evaluations and mechanical tests to determine final load and tension and ending with radioisotope cell tracking tests to track and display the contribution of transplanted cells to repair and remodeling. Thus, it shows the preparation and scientific rigor in detecting functional evidence using state-of-the-art technology from different technological sectors. It also brought an immense contribution to clinical applicability.

A novel physical approach was also identified to produce a cell-free aqueous extract of human adipose tissue [65]. The therapeutic potential of fat extract (FE) was investigated in the hindlimb of nude mice following ischemia. This study is extraordinarily innovative and uses a different mechanical dissociation from those previously demonstrated. The *in vitro* and *in vivo* experiments showed that endothelial cells assume a tubular formation and are induced with and without FE when combined with Matrigel scaffold formation. A significant advantage of this technology is that no immunological rejection should occur when using cell-free FE, meaning that FE could be marketed as an “off-the-shelf” product for treating ischemic disorders. This study, in particular, supports the idea that no fluid should be discarded from the chemical or mechanical dissociation of adipose tissue without fully understanding its properties since it is possible that these “waste products” contain valuable material with regenerative potential. The innovative adipose tissue dissociative methods and all of the currently available information could attenuate the waste of cells and derivatives and generate different products from the human lipoaspirate.

Based on the results of this review article, it is clear that biotechnology, Big Data, and technological disruption have entered the plastic surgeon’s world and have started to change our way of thinking, guiding us to a horizon of new ideas and innovation.

Conclusions

Innovation and technology are linked to plastic surgery and procedures that use adipose tissue as a source of ASCs and the SVF. Of the 33 selected articles, the isolation of ASCs was primarily performed using an enzymatic dissociation method. It was also determined that liposuction was the most performed plastic surgery procedure, while most innovative technologies were tissue scaffold related. These reoccurring themes possess enormous potential in innovative areas and introduce plastic surgeons to regenerative medicine, transforming them from a supporting role to the leading actor in this scenario of disruptive technological innovation. We conclude that these experimental research and development areas could facilitate utilizing ASCs for *in vivo* human plastic surgery and regenerative medicine applications.

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Declarations

Ethics approval This study was a review article and did not require ethical approval.

Conflict of interest Hebert Lamblet and Lydia Masako Ferreira declare no conflict of interest.

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