Recent Advances of Adipose-Derived Stem Cell-Based Therapies

Yanhua Qi

Abstract: Adipose-derived stem cells (ADSCs), which were initially isolated from the aspirate of human fat by Zuk et al., are considered as an attractive cell source of mesenchymal stem cells (MSCs) for cell-based therapies. Due to their easily accessible, broader sources of multipotent cells and stronger ability for amplification in vitro, ADSCs have attracted considerable attention and curiosity from both scientific and clinical communities for their potential mechanisms in future stem cell treatments. Furthermore, their promising therapeutic characteristics were the unique ability of immunosuppression, secretory function and the potential for multi-directional differentiation. With more research efforts made to understand their effects on both scientific and clinical fields, ADSCs possess great potential as a future therapeutic strategy in treatment for widely various diseases. Therefore, this review intends to lay stress on the clinical application of ADSCs and their advancement in clinical trials across a variety of medical disciplines. Due to their advantages in various fields, ADSCs have a strong potential for application in future stem cell treatments and are beneficial for allogenic transplantation treatments. ADSCs may be considered as an alternative in cell transplantation and tissue engineering clinical treatments.

Keywords: ADSCs; immunoregulatory; paracrine; differentiation capacity; cell transplantation

1. Introduction

Adipose-derived stem cells (ADSCs) are generally isolated from the stromal vascular fraction (SVF) of homogenized adipose tissues [1]. Initially, ADSCs were isolated from the aspirate of human fat by Zuk et al. [2]. Because of their abundance and easy accessibility, ADSCs have obtained great attention and curiosity for their promising therapeutic applications since its discovery. ADSCs are one of the most potential stem cell types, since they are easy to be obtained from liposuction aspirates or subcutaneous adipose tissue fragments [3]. Stem cells refer to cells capable of self-renewal and differentiation into various phenotypes, and they can be divided into embryonic stem cells (ESCs) and adult stem cells (ASCs) [4]. It is also known that ADSCs belong to mesenchymal stem cell (MSC), a kind of ASCs, which are applied in cell-based therapies and are regenerative medicine, such as neurodegenerative disease, bone repair therapy and so on [5,6]. In accordance with the standard criteria established by the Mesenchymal and Tissue Stem Cell Committee of the International Society for Cellular Therapy, MSC must meet the following criteria: (i) plastic adherent in standard culture conditions; (ii) express CD105, CD73 and CD90, but negative for the surface expression of CD45, CD34, CD14, CD79 and HLA-DR; and (iii) able to differentiate into osteoblasts, adipocytes and chondroblasts in vitro [7]. MSCs are multipotent stromal cells that are found in some tissues and that have ability to differentiation into tissues of mesenchymal origin, including bone marrow, lung and adipose tissue [8–10].

In comparison to BMSCs, ADSCs hold a longer incubation time and the stronger ability of proliferation so that the adipose tissue becomes a good source of autologous stem cells [11]. It is reported that ADSCs could be induced to differentiate into adipocytes, cardiac myocytes and osteoblasts [12,13]. The conditioned medium derived from ADSCs is also a therapeutic choice and has been indicated to play protective roles in chondrocytes, such as the reduction of oxidative and inflammatory stress [14]. As such, ADSCs are now a widely recognized source for stem cells in regenerative medicine and has been attached great attention to a lot of preclinical and clinical studies geared towards numerous applications [15]. In many aspects, ADSCs are applied in cell transplantation therapy, above all for avoiding the ethical issues and tumorigenic complications that are generally correlated with embryos or induced pluripotent stem cells [11]. It is no doubt that ADSCs serve as an attractive cell source for therapeutic applications, including neurodegenerative disorders [16], arthritis [17], wound healing [18], and plastic surgery [19]. Over past decades since the discovery of ADSCs, this paper aims at offering an appropriate review of the development of ADSCs in clinical applications. The review emphasizes the clinical application of ADSCs and its advancement in a variety of clinical studies, as well as a prospect of the ADSC therapy field.

2. Immunoregulatory Functions of ADSCs

The current studies have proved that ADSCs are like BMSCs in the ability of immunosuppression, while the recent studies indicated that there still are some differences between them. Dendritic cells (DCs) are the most potent antigen-presenting cells (APCs), capable of activating both naive and memory immune responses, and maintaining the delicate
balance between immunity and tolerance [20]. MSCs could inhibit monocyte CD14+ to differentiate to DC. But for differentiatied DC, MSCs can activate the expression level of costimulatory receptor CD83 by downregulating its T cells so that it can transform the mature state to immature state. In addition, because MSCs could downregulate HLA-DR, costimulatory receptor and decrease the level of IL-12, resulting in the ability of DC stimulating primary T cells deceased. In conclusion, MSCs could affect the antigen-presenting function of DC so that the suppression of immune response could be achieved. Ivanova-Todorova E made a comparative study in immunosuppression of ADSCs and BMSCs. The results indicated that ADSCs can also suppress monocyte to differentiate into DC, and inhibit co-stimulating factors CD80 and CD86 expressed by DC. Moreover, the immunosuppressive effect of ADSCs was stronger than that of BMSC. And the ability of increasing level of immunosuppressive factor IL-10 by simulating DC is much stronger. The findings of in vitro experiments showed that compared with BMSCs, the immunosuppression of ADSCs was stronger [21]. However, the accurate mechanism for this kind of difference has not been explained, maybe it was related to different immunophenotypes in ADSCs and BMSCs.

DCs modulate B-cell survival and differentiation, mainly through production of growth factors such as B lymphocyte stimulator [22]. B cells, also known as B lymphocytes, are a type of white blood cell of the lymphocyte subtype [23]. B cells regulate various functions that affect immune and inflammatory responses in autoimmune diseases, including rheumatoid arthritis (RA) [24]. They function in the humoral immune component of the adaptive immune system by secreting antibodies. Some reports have proved that the modulation of B lymphocyte by MSCs was determined by status and species origin of MSCs. Krampera et al. indicated that primary culture of MSCs would not influence the proliferation of B lymphocytes, but if BMSCs were treated with interferon-gamma (IFN-γ) in vitro culture, BMSCs would affect the proliferation of B lymphocytes. There were some studies demonstrated that it would make a stop for cell proliferation in phase G0/G1 by secreting soluble factors, not touching with the cells. Bochev et al. constructed a co-culture system with peripheral blood mononuclear cells (PBMC) and with ADSC or BMSC respectively. Next, pokeweed mitogen (PWM) was added into the system for incubation for 7 days respectively, while the systems without PWM served as control. Enzyme-linked immunosorbent assay (ELISA) was performed to detect the immunoglobulin concentration of culture supernatant. The results indicated that MSCs significantly inhibit immunoglobulin through stimulation of monocyte by PWM. The immunosuppression in ADSC group was stronger than that in the BMSC group [25]. Moreover, it could suppress the secretion of immunoglobulin by B lymphocytes, and interfere chemotaxis of B lymphocytes by downregulating the expression of chemokine receptor on the surface of B lymphocytes.

3. Paracrine Function of ADSCs

Paracrine function of MSCs plays a crucial role in the field of regenerative medicine. Renata I. Dmitrieva et al isolated hADSCs and hBMSCs from human adipose tissues and from myeloid tissues respectively. Then, hADSCs and hBMSCs were cultured and their secretions were analyzed. The results showed that there were no differences in IL-6 level between hADSCs and hBMSCs, while the levels of vascular endothelial growth factor (VEGF), SDF1, monocyte chemoattractant protein 1 (MCP1) and TGFβ1 in hADSC were significantly lower than those in hBMSCs [26]. Quantitative real-time polymerase chain reaction (qRT-PCR) and ELISA were performed to evaluate hADSCs and hBMSCs from five volunteers. The findings indicated that mRNA levels of brain-derived neurotrophic factor (BDNF), VEGF and hepatocyte growth factor (HGF) expressed by hADSCs were significantly higher than those expressed by hBMSCs. The results of ELISA further revealed that the concentration of BDNF and VEGF in hADSCs was evidently higher than that in hBMSCs [27]. Above experimental data were acquired from human MSCs. Gene technology was adopted by Chiaki Nakanishi to analyze the gene expression level in ADSCs and BMSC31009. The results indicated that the expression levels of 571 genes in ADSCs were three times as much as those in BMSCs. Meanwhile, the expression levels of 571 genes in BMSCs were three times as much as those in ADSCs. Through classification analysis, the results showed that the expression levels of those genes that are related to cell differentiation, such as related protein of WNT-1α secreted by hADSCs from human or mouse were differentiated into neurons. Basic fibroblast growth factor (bFGF) and epidermal growth factor (EGF) were added during the process of neural induction of hADSCs. After seven days, the results of immunohistochemistry indicated that the expression levels of NeuN and nestin significantly increased [29]. After treated with bFGF and EGF, respectively, hADSCs were formed into neurospheres, which would proliferate and be induced and differentiated into schwann cells.
and glial cells [30]. Zhang et al. compared the differentiation capacity between hADSCs and hBMSCs, which were extracted from adipose and marrow of a volunteer. Firstly, hADSCs and hBMSCs were incubated in neuronal medium containing bFGF, EGF and B27 so that the neurospheres were formed. Next, the neurospheres were transferred into medium containing all-trans retinoic acid (RA), bovine serum albumin (BSA), horse serum and N2 additive agent. After several days of incubation, the findings showed that the level of nestin expressed by hADSCs was significantly higher than that by hBMSCs. During the further process of neural induction, the levels of neuron and glial cell markers expressed by hADSCs were evidently higher than those by hBMSCs [31]. These results indicated that hADSCs possesses differentiation capacity under some condition, and it was stronger than that of hBMSCs. It was widely recognized that hADSCs could be differentiated into schwann cells. hADSCs was differentiated into schwann cells induced by β-Mercaptoethanol, RA, forskolin, bFGF and platelet-derived growth factor (PDGF) [32,33].

5. Applications of ADSCs in Cell Transplantation

5.1. ADSC Transplantation in CNS Lesion

Previous studies have explored the beneficial effects of MSCs transplantation in CNS injuries, including traumatic brain injury (TBI), stroke and spinal cord injury animal models [34]. A recent study reported by Ji and his team has shown protective effect of brain-derived neurotrophic factor and neurotrophin-3 overexpression by ADSCs combined with silk fibroin/chitosan scaffold in spinal cord injury [35]. Besides, ADSCs expressing the neurogenin-2 promote functional recovery after spinal cord injury in rat [36]. Intracarotid transplantation of autologous ADSCs was shown to significantly improve neurological deficits in rats following ischemia/reperfusion injury caused by middle cerebral artery occlusion [37]. Ryu et al. prepared adult dog models of compression spinal cord injury (SCI). In the 1st week after injury, ADSCs were transplanted. The results showed that in the 2nd week after injury, recovery of motor function in model group was better than that in the control group until the 8th week. The finding of spinal cord evoked potential (SCEP) revealed that transplantation of ADSCs could significantly improve the spinal function of experimental animals. After 9 weeks, the dogs were sacrificed, the results of immunohistochemistry showed that transplantation of ADSCs could differentiate into GFAP, β-tubulin (Tuj) and NF160 positive cells. The author suggested that neural differentiation of ADSCs was a potential mechanism for repairing animals in SCI. I remain cautious for this research finding, especially ADSC transplantation and differentiation into neuronal cells. The results of our study indicated that hADSC transplantation could repair rats in SCI. In the 4th week after transplantation, the survival rate of hADSC was very low. ADSC differentiation into positive cells, such as GFAP, Tuj, NF200, S100, CNPase, was not detected. The reasons for this difference might be that seed cells ADSCs in this study were from human beings, and the time of transplantation is transplanting immediately after injury, which were different from the last study. However, it was widely discussed whether MSCs could differentiate into neuronal cells or not, which needs to be further studied [38]. Kang et al. utilized ADSC in rats to induce and differentiate into oligodendrocyte precursor cells (OPCs) and vein transplantation into SCI rats. In this study, after 4–5 weeks for transplantation, there were about 30% cells survived, and the cells migrated to injured parts. Some cells were differentiated into neurons and oligodendrocytes. The author indicated that the transplanted cells were found in kidney, brain, lung and liver tissues after vein transplantation. The findings of praxeology showed that cell transplantation could promote the recovery of rat motor function. It needs to be noticed that vein transplantation of ADSCs was adopted in this study, and the transplanted stem cells were found in brain tissues and so on [39]. When Ikeyame et al. studied that vein transplantation of ADSCs and BMSCs could repair ischemic stroke rat model, they indicated that after 24 h for transplantation, the transplanted cells were found in lung tissues rather than brain tissues, while ADSC transplantation could significantly increase the expression levels of VEGF and HGF in brain tissues of a rat model of stroke and decrease infarct size to promote the recovery of cerebral function [40]. Kolar et al. investigated the therapeutic effects of ADSCs on axonal regeneration following transplantation into an injured rat model of cervical spinal cord. The results of this study revealed that ADSCs do not affect rat astrocytes in culture and can enhance neurite outgrowth across the co-cultured astrocytes. After transplantation, ADSCs still expressed BDNF, VEGF, and fibroblast growth factor-2 for 3 weeks when rats were treated with cyclosporine A (CsA). The extensive ingrowth of 5HT-positive raphaeospinal axons was stimulated into the trauma zone by transplanted ADSCs. Moreover, transplanted cells caused the change of the structure of the lesion scar with an ample amount of astrocytic processes extended into the middle of the trauma zone in a chain-like pattern and closely related to regenerating axons. In addition, ADSCs can decrease the reactivity of astrocyte and microglial cells. The results indicated that ADSC transplantation can modify the structure of the glial scar and stimulate axonal sprouting to exert beneficial influences on rats with SCI. [41]. Leu has done the same research that ADSC transplantation repairs rats with ischemic cerebral infarction. He reported that 21 days after ADSC transplantation, transplanted cells were found in rats’ brain. At that time, some ADSCs have differentiated into endothelial cells. The results of his study showed that ADSCs could downregulate inflammation and apoptosis around the infarction and promote vascular proliferation around the injury. There was great difference in cell survival data between these two subjects. The reasons for this difference might be caused by the different number of transplanted cells, the former was 1 × 10^5 cells, while the later was 2 × 10^6 cells. However, it remains controversial that transplanted cells were detected in
brain after vein transplantation, which needs to be further studied. Moreover, ADSCs could exert greatly positive influence in the treatment for hemorrhagic stroke [42]. Furthermore, Tang et al. investigated the roles of ADSCs overexpressing the Ngn2 transgene (Ngn2-ADSCs) in the improvement of functional recovery in a rat model of SCI. After transplantation, an ample amount an ample amount of ADSCs was found around the center of the injury spinal cord at 1 and 4 weeks, which improved retention of tissue at the lesion site. In addition, the findings of immunohistochemistry with glial fibrillary acidic protein revealed that transplantation of Ngn2-ADSCs increased the trophic factors, and repressed the glial scar formation. Thus, these major findings implied that Ngn2-overexpressed ADSC transplantation promote the functional recovery from SCI, and improve the local microenvironment of injured cord in a more efficient way than that with ADSCs alone [36]. Kim et al. made a research that rat models of cerebral hemorrhage were treated with vein transplantation of ADSCs, which revealed that ADSC transplantation downregulate the terminal transferase uridyl nick end labelling (TUNEL)* the number of apoptotic cells and myeloperoxidase (MPO)* the number of inflammatory cells. Four weeks after cell transplantation, the recovery of cerebral function of rats in the transplanted group was better than that of rats in the control group. The results of immunohistochemistry showed that in the 6th week after cell transplantation, an ample amount of ADSCs were found around the brain edema. These cells could express marker of epidermal cells von willebrand factor (vWF), but not express markers of neuronal and glial cells. This study found that ADSC transplantation repairing rats with cerebral ischemic stroke could decrease inflammation and apoptosis in the acute stage, and alleviate cerebral atrophy in chronic-phase to promote the recovery of rats’ long-term function. The role of ADSCs in brain trauma was not proved in the current study, which has limitations on effects of pure ADSC transplantation, but if ADSCs were induced into schwann cells before transplantation, they could significantly downregulate the activity of glial cells, and decrease the secretion of glial inhibitory factor to promote the recovery of cerebral function in rats with cerebral injury [43].

Tomita et al. proved that after ADSCs inducing into schwann cells, the expression levels of BDNF, GDNF, and NGF much more significantly increased secreted by ADSC than before inducement. After transplantation of ADSCs into rats with cerebral injury, the survival rate of induced ADSCs was much higher, and the rate of intramedullary pin reformation and non-induced ADSCs significantly increased. In the last paragraph, we concluded the immunoregulatory functions of ADSCs, and in vivo, it has been demonstrated that ADSC transplantation could also regulate immunologic functions [44]. Constantin et al. found that vein transplantation of ADSCs before inducing animal models of encephalomyelitis could evidently decrease experimental animals with encephalomyelitis and demyelinating diseases. Moreover, the mechanisms of ADSCs were suppressing autoimmune responses in the early stage of disease and inducing neuroprotective effect s of endogenous neural stem cells [45].

It seems that neuroprotective effect of ADSCs was not restricted to CNS, and it also plays a positive role in repairing peripheral nervous system. Di Summa et al prepared animal models of sciatic nerve injury and repairing injury with technology for tissue engineering. The findings showed that axonal regeneration of animals with ADSC transplantation was better than that of animals in the control group, which indicated the great effects on repairing peripheral nerve, but this study did not discuss the mechanism of neural regeneration [46]. Lopatina et al. further studied the repairing effects of ADSCs on brachial plexus injury, which showed that ADSC transplantation could significantly increase the expression level of BDNF. The results of immunohistochemistry indicated that axonal regeneration of brachial plexus showed a marked increase. However, if BDNF was eliminated by BDNF antibody, the axonal regeneration would disappear. This study pointed out that the mechanism of ADSCs repairing brachial plexus injury was correlated with the elevation of BDNF secretion [47]. However, although ADSCs have been adopted in various animal models to aid the recovery, at present, the effect of the ADSCs on peripheral nerve injury is still required to be discussed. Furthermore, the optimum timings and methods of insertion of ADSC have yet to be confirmed. The preliminary results have indicated a future role in peripheral nerve regeneration, but future studies are need towards further optimization and safety of ADSCs in peripheral nerve regeneration [48].

5.2. ADSC Transplantation in Vascular Lesion

Moon et al indicated that in vitro, hADSCs could differentiate into endothelial cells and express vWF. The conditioned medium from hADSCs could increase the arterial endothelial cell proliferation and inhibit apoptosis. Transplanting hADSCs into nude mouse models of limb ischemia could significantly improve the muscle function of ischemic limb and increase the new vessels. hADSCs were great cell sources for treating ischemic diseases, which is consistent with the results of ADSC repairing rats with ischemic stroke [49]. According to the findings of this study, Kim et al. underwent a deep research the mechanism of hADSCs repairing angiogenesis and compared hADSCs with hBMSCs using the same rat models. The results showed that the expression levels of matrix metallopeptidase 3 (MMP3) and matrix metallopeptidase 9 (MMP9) secreted by hADSCs in vitro were higher than that by hBMSC, and the ability of vascular cavity formation of hADSCs was much better than that of hBMSC. Downregulating the expression levels of hADSC MMP3 and MMP9 by RNA interference technology could suppress the ability of hADSCs promoting RNA interference technology. After ADSC transplantation, laser Doppler showed that the blood flow of experimental animal limb significantly elevated, which referred that hADSCs were better sources of angiogenesis than DMSC 9 [50]. Bhang et al
indicated that hADSCs could improve limb ischemia injury. But the findings showed that the expression level of SDF-1α significantly increased and the ability of ADSCs secreting HGF, VEGF and FGF also increased by changing the culture condition into hypoxia to simulating ADSCs through microsphere culture. After transplanting ADSCs into ischemic limb muscles, compared with ADSCs cultured by the single cell, the survival rate of ADSC that cultured by microsphere and the expression level of VEGF were higher, the number of new blood vessels was more, and the treatment effect was better. The results of ADSC promoting angiogenesis in different studying group were similar, and a lot of studies demonstrated that ADSC could differentiate into vascular endothelial cells (VEC) in vivo and in vitro [51]. Furthermore, Ning and Konno cultured ADSC with medium for VEC differentiation. The conditions of ADSCs differentiating into VECs were analyzed in accordance with adding different factors, such as bFGF, EGF, VEGF, IGF-I, hydrocortisone, heparin and Vitamin C. The results showed that removing VEGF, EGF, and IGF-I does not affect ADSC differentiation, while removing bFGF could significantly downregulate the ability of ADSCs increasing low density lipoprotein (LDL) and expression levels of endothelial cell markers, which showed that bFGF plays an important role in ADSC differentiating into VEC [52,53].

5.3. ADSC Transplantation in Myocardial Ischemia

Because the ability of ADSC promoting angiogenesis is strong, and ADSCs exert great effects on peripheral vascular injury, it is naturally that researchers focused on the role of ADSC transplantation in animal models of myocardial ischemia. Valina et al. firstly discussed this subject, and compared it with BMSCs. They prepared experimental animal models of acute myocardial infarction (AMI). After 30 days, ADSCs and AMSCs were transplanted into coronary artery. The results indicated that compared with injury control group, left ventricular ejection fraction was increased by 11.39 ± 4.62% and 9.59 ± 7.95% in the ADSC and BMSC groups respectively. The transplanted MSCs could differentiate into endothelial cells and smooth muscle cells. From the functional aspect, it seems that there was no difference in repairing effects of ADSCs and BMSCs. However, according to calculation of infarct zone/non- infarct zone, it revealed that ADSC transplantation could improve ventricular remodeling of experimental animals, which showed that ADSC exerts notable impacts on AMI [54]. However, van der Bogt et al. different opinions about ADSC repairing AMI. They suggested that both ADSCs and BMSCs showed an ample amount of MSC death within 4 - 5 weeks after transplantation, which has no evidently positive effects on the recovery of cardiac function. It may be related to the low survival rate of cells [55]. The two Research groups differs in experimental animals (the former was pig, and the later was rat), transplantation method (the former was coronary artery, and the later was caudal vein) and detection means (the former was iconography and immunochemistry, and the later was reporter-gene imaging with fluorescence and ultrasonic). Although the effect of ADSC transplantation in repairing AMI is controversial, ADSC cardiomyocyte differentiation is recognized. Planet-Benard and his teamwork found that primary cultured hADSC could differentiate into cells that are like myocardial cells in the morphology, molecular and function. These cells could express markers of myocardial cells, including Gata4, Nkx2.5, Mlc-2v, and Mi-2a, and spontaneous action potential and evoked action potential would occur under current-clamp [56]. Furthermore, Rasmussen et al. compared BMSCs with ADSCs from an elderly ischemic patient in the treatment of myocardial infarction (MI) with a fully grown non-immune-compromised rat model. After 1 week following induction of MI, rats treated with intramyocardial injections of ADSCs, BMSCs or phosphate-buffered saline. The results showed that left ventricular ejection fraction (LVEF) was accelerated in the ADSC group, and scar wall thickness was greater compared with the saline group. Cardiac function was recovered by ADSCs from a human ischemic patient preserved following MI, while the effects of BMSCs in cardiac function could not be proved, with only ADSCs exerting great impacts on improvement of LVEF [57]. However, there were few studies discussing whether ADSCs could differentiate into myocardial cells in the host body or not. Therefore, the role of ADSCs in myocardial infarction models needs to be further developed.

5.4. ADSC Transplantation in Liver Damage

Liver is one of the sources of MSCs, but what is the effect of ADSCs in the treatment for liver failure? Recently, Paik and his teamwork has proposed novel antifibrotic strategy utilizing conditioned media obtained from miR-150-transfected ADSCs in an animal model of liver fibrosis [58]. Banas et al. discussed ADSCs repairing liver damage. The findings of experiments indicated that ADSC transplantation could improve the liver function of animal with liver failure. For further developing the mechanism of this effect, the factors secreted by ADSCs were extracted and compared with BMSCs, which shows that the concentration of interleukin receptors secreted by ADSCs, such as 1α, IL-6, IL-8, granulocyte colony-stimulating factor (G-CSF), granulocyte macrophage colony stimulating factor, monocyte chemoattractant protein 1 (MCP 1), NGF and HGF was higher than the concentration of interleukin receptors secreted by BMSC [59]. Later, they proved that ADSCs could differentiate into liver cells, which possess the function of primary cultured liver cells. After transplanting ADSCs into rat models of liver failure, it could downregulate alanine aminotransferase, aspartate aminotransferase etc. Its repairing effect was better than pure ADSC transplantation [60]. Above-mentioned studies demonstrated the repairing effects of ADSCs on animals with liver failure. Meanwhile, ADSCs could also exert protective effects on liver injury induced by ischemia/reperfusion. Sum et al. proved that after rats with
liver injury induced by ischemia/reperfusion, ADSC transplantation could significantly downregulate liver oxidative stress, cell apoptosis and inflammatory reaction of rats to improve liver function of rats [61]. Pan et al. isolated ADSCs from the adipose tissues of rats, and ADSCs were transplanted into the liver of high-fat-diet-induced non-alcoholic fatty liver disease (NAFLD) rats through the portal vein to decrease the disease progression of NAFLD. The results demonstrated that after ADSC transplantation, the serum levels of alanine aminotransferase, total bilirubin, total cholesterol, triglycerides and fatty acids, and the content of malondialdehyde in the liver homogenates showed significant reduction. However, superoxide dismutase activity significantly increased after ADSC transplantation. These findings revealed that the ADSC transplantation enhanced the liver function, and decreased lipid metabolism and oxidative stress. Moreover, the serum levels of IL-6 and TNF-α, which are predominantly secreted by the kuffer cells in the liver and actively lead to the acute inflammatory responses, were notably dropped at the 4th week post-ADSC transplantation in comparison to the NAFLD rats. ADSC transplantation selectively reduced the secretion of pro-inflammatory cytokines, such as TNF-α and IL-6, attenuating the progression of NAFLD. In conclusion, ADSC transplantation decreases the disease progression of high-fat-diet induced NAFLD [62]. These studies indicated that the repairing effects of ADSCs on liver are not different, though the types of damage are different.

5.5. ADSC Transplantation in Bone, Joint, and Muscle System
ADSCs originate from cells in mesoderm, and its ability of differentiating into bone and cartilage is strong. The experiments on ADSC transplantation repairing bone defect were earlier, and the experimental results were consistent. Chang et al. demonstrated that ADSCs suppress osteoclastogenesis and bone erosion in collagen-induced arthritis [63]. In addition, secreted factors and EV-miRNAs orchestrate the healing capacity of ADSCs in treating knee osteoarthritis [64]. Cowan et al. demonstrated that the effects of ADSC transplantation repairing skull defect was great, and there were no needs for any treatment for ADSCs in vitro or adding any simulating factors. The results of chromosome examination showed that 84–89% new bone was formed via cell transplantation [65]. Di Bella treated ADSCs with osteoinduction, and combined with polylactide (PLA) scaffolds, which would increase the ability of ADSC repairing skull defect. Except for skull, ADSCs could promote lumbar spinal posterolateral fusion [66]. Lopez et al. established rat models of transverse process of lumbar vertebra and transplanted ADSCs on a biomaterial scaffold composed of tricalcium phosphate and collagen I into local damage. The results indicated that ADSCs composite scaffold significantly promote lumbar spinal posterolateral fusion, while 8 weeks after damage, the animals in the scaffold group without cells had no effects on lumbar spinal posterolateral fusion [67]. Peterson discussed genetic modification of ADSCs, overexpression of bone morphogenetic protein 2 (BMP-2), exerts great effects on repairing femoral defect in athymic rats [68]. Shoji demonstrated that hADSC transplantation could repair immunodeficient rats with femoral non-union fracture. The results of immunohistochemistry and qRT-PCR indicated that in the 2nd week after transplantation, hADSCs could differentiate into bone cells and endothelial cells. The laser Doppler showed that hADSC transplantation improve the blood volume of non-union fracture, and the histopathology and iconography proved that bone healing of rats in the ADSC transplantation group was better than that in the control group [69]. These experimental results showed that the potentials of ADSCs repairing bone defect were affirmed. Intrararticular injection of ADSCs could notably improve the on promoting joint regeneration and cartilage protection of experimental animals with chronic osteoarthritis [70]. Vein transplantation of ADSCs could decrease autoinflammatory response and autoimmune response of animals with rheumatoid arthritis [71]. At present, ADSCs were applied in repairing models of tendon injury. Uysal et al. established rabbit models of tendon injury, and treated rabbit models with local transplantation of ADSCs. The findings showed that ADSC transplantation could promote tendon repair. The results of histopathology indicated that ADSCs could differentiate into cells and endothelial cells and increase local angiogenic growth factors [72].

5.6. ADSC Transplantation in Plastic Surgery
Because of the ability of self-renewal, secretion of trophic factors and differentiation into different cell types, stem cells have provided more customizable treatment options, and ADSCs are being adopted for various clinical applications in plastic surgery [19]. Due to the unpredictability and low survival rates related to lipotransfer, researchers have tried to raise the vascularization of the graft with some products, including insulin, VEGF, and enriching fat with ADSCs [73]. Cell assisted lipotransfer (CAL), an autologous fat transfer enriched with stromal cells, is a surgical method that is presently adopted sparingly in the plastic and reconstructive surgery [74]. In the CAL method, fat is enriched with ADSCs, contained in the SVF obtained after enzymatic digestion of fat or after cell culture to improve the fat survival rate [73]. Autologous fat transplantation is often applied for various cosmetic and reconstructive indications which were not restricted to posttraumatic defects of the face and body, involutional disorders including hemifacial atrophy, and many aesthetic applications such as lip and facial augmentation and wrinkle therapy [75]. In addition, Eva Koellensperger et al. explored the effects of ADSCs on breast cancer cell lines (BRCAs) regarding the safety of cell-assisted lipotransfers for breast reconstruction. In this study, ADSCs were co-cultured with different human BRCAs, such as MCF-7, MDA-MB-231, SK-BR-3, ZR-75-30, and EVSA-T. The results of qRT-PCR indicated significant changes in the expression of multiple tumor-associated genes in co-culture in comparison to monocultures of both ADSCs and BRCAs. Co-culture did
not significantly affect cellular proliferation of either ADSCs or BRCAs. The migration of MCF-7 and MDA-MB-231 BRCAs was notably increased in co-culture with ADSCs. Co-culture with MDA-MB-231, SK-BR-3, and EVSA-T BRCAs showed evident increases in the ADSC invasion. An in vitro angiogenesis assay revealed that compared with the respective monocultures, an increased tube formation of conditioned media was found in co-cultured BRCAs and ADSCs. However, a potential oncological risk should not be neglected when considering a clinical application of CAL in breast reconstruction [76]. Presently, significant translational barriers remain including regulatory challenges and ethical considerations [1]. Therefore, further technical, outcome standardization and rigorous randomized controlled trials are required to delineate long-term clinical efficacy and safety.

5.7. ADSC Transplantation in Wound Healing

Wound healing is a normal process in response to soft tissue injury, which involves a highly organized cascade of events. These phases include hemostasis, inflammation, proliferation, and remodeling. Over the last few years, stem cell therapy has provided promising treatment options for various diseases including wound repair and tissue regeneration. [77]. Because ADSCs not only differentiate into keratinocytes, fibroblasts, and endothelial cells, as evidenced by their morphology, expression of cell surface markers, and gene expression, but also secrete several soluble factors, which positively contribute to wound healing in a paracrine manner. Clinical trials have been conducted using autologous ADSCs with great success [78]. Alternatively, ADSCs may be immediately administered without in vitro differentiation in culture. The extraordinarily high cell yield from lipoaspirate, compares with the bone marrow aspiration, making ADSCs a particularly appealing cell source for the acute wound setting [79]. Not only that, the effects of ADSCs have been proved in some preclinical trials on wound healing and have been demonstrated on significantly enhancing cutaneous wound healing and promoting blood vessel formation [80,81]. An et al. demonstrated that ADSC isolated from diabetic mice improve cutaneous wound healing in streptozotocin-induced diabetic mice [82]. Yoshida et al. treated hindlimbs of Sprague-Dawley rats with 20-Gy x-ray irradiation and surgical occlusion. The results of Laser tissue blood flow showed a notable elevation in the combined treatment of flap and ADSCs in the 1st and the 2nd week, while no significant differences were observed between the treatment groups treated with flaps alone and those treated with ADSCs alone. Wound healing showed a significant increase following combined treatment in the 1st week. The number of vessels evidently increased in the combined treatment group in the 1st and the 2nd week. In the cell tracking group, in the 2nd week, the green fluorescent protein–tagged ADSCs were markedly more positive in the no-flap group than in the flap group. From this study, it could be found that ADSCs can be a promising cell source in irradiated and occluded limbs by promoting tissue blood flow and blood vessel density, and exerted a great effect on some difficult ischemic conditions in terms of wound healing [83]. Kuo et al. proved that ADSCs enhance diabetic wound healing via the induction of autocrine and paracrine effects. Dorsal full-thickness skin wound defects (6 × 5 cm) were created in a rodent model of streptozotocin (STZ)-induced diabetes. Results indicated that complete wound healing time statistically decreased in the ADSC-treated group in comparison to the controls group. Histological examination revealed that compared with the control group, proinflammatory reaction significantly reduced, while the levels of EGF, VEGF, rPH, and Ki-67 expression notably elevated in ADSC-treated group. The populations of green fluorescent protein (GFP)^+-ADSCs in circulating blood significantly increased after ADSC injection in comparison to those in the control group. The findings of immunofluorescence staining revealed that GFP^+-ADSCs evidently increased in the subdermal layer of the wound margin and elevated angiogenesis through vWF and VEGF expression after injection. Therefore, ADSCs significantly enhanced diabetic wound healing, engrafted into the local wound tissue, and implanted into circulating blood [84]. However, the clinical application of ADSC transplantation in wound healing is still at its preliminary stage and the number of relevant documents remains limited, further studies with larger sample size are needed to explore the effect of ADSC transplantation on wound healing.

5.8. ADSC Transplantation in Diabetes Mellitus

Replacement of pancreatic beta cells by islet-like cells has the potential to cure diabetes mellitus. The current studies proved that ADSCs would be seed cells. Chandra successfully induced ADSCs into islet-like cells that could secrete insulin, glucagon and somatostatin after 10 days of ADSC induction. Rats with diabetes mellitus were treated with intraperitoneal injection of islet-like cells. After two weeks, blood sugar level was reduced to the normal range [85]. At the same time, Kajiyama et al. adopted gene transduction for pancreatic duodenal homeobox gene-1 (Pdx1) overexpression. The results of ELISA in vitro showed that after treated with stimulation of high glucose, the number of ADSCs secreting insulin that expressing Pdx1 significantly increased. ADSCS carrying Pdx1 were transplanted into rats with diabetes mellitus, which could evidently decrease blood sugar of rats and increase sugar tolerance. The results of histopathology indicated that transplanted cells formed tissue-like structure and express insulin [86]. Lin et al. elucidated the effects of ADSCs in type 1 diabetes mellitus (T1DM), which is a type of early onset diabetes mellitus, whose main feature is the autoimmune destruction of insulin-producing cells (IPCIs), leading to hyperglycemia and abnormal glucose metabolism. Therapeutic advantages and clinical applicability of ADSCs have been demonstrated in T1DM, ensuring their suitability for transplantation therapy. In addition, some previous reports have highlighted the therapeutic potential
of ADSCs for congenital causes or acquired factors of diabetes. [11]. In addition, there is a previous study has discussed the ADSC infusion on rats with type 2 diabetes mellitus (T2DM). Compared with diabetic control group, ADSC infusion improved hyperglycemia in ADSC group in the 2nd week and maintained for about 6 weeks, and significantly increased plasma concentrations of insulin and C-peptide. Number of islet β cells and concentration of vWF in islets in ADSC group showed an elevation, while activity of caspase-3 in islets presented a reduction. Furthermore, concentrations of TNF-α, IL-6 and IL-1β in ADSC group evidently reduced. The expression of GLUT4, IRS1, and phosphorylation of insulin signaling molecules in insulin target tissues were effectively improved. ADSC infusion could aid in T2DM through recovery of islet β cells and improvement of insulin sensitivity [87]. However, these studies indicate that therapeutic ability in partial recovery of syndromes of diabetes with a short effective period was limited by transplantation of undifferentiated ADSCs

5.9. ADSC Transplantation in Alzheimer's Disease (AD)

It is reported that ADSCs are a promising cell source for regenerative therapy. Katsuda et al. described a method by which to evaluate the therapeutic potential of hADSC-derived EVs in the treatment for AD from aspect of their β-amyloid peptide (Aβ)-degrading capacity. They found that exosomes carrying enzymatically active nεpilisin secreted by hADSCs is the most significant Aβ-degrading enzyme in the brain [88]. Yan et al. demonstrated that ADSC transplantation promotes adult neurogenesis in the brains of AD. In this study, ADSCs was transplanted into the hippocampi of APP/PS1 transgenic mouse model of AD. The results of immunofluorescence staining revealed that ADSC transplantation significantly increased the number of newly generated (BrDU+) cells in the subgranular zone of the dentate gyrus in the hippocampus, which was evidently higher in AD mice, and the number of BrDU+/DCX+ neuroblasts significantly elevated in mice. Moreover, ADSC transplantation enhanced neurogenic activity in the subventricular zone and decreased oxidative stress and attenuated cognitive impairment in mice. The findings of this study elucidated that ADSC transplantation promotes endogenous neurogenesis in both the subgranular and subventricular zones in APP/PS1 transgenic AD mice so that can enhance functional recovery [89]. Ma et al. demonstrated that intracerebral transplantation of ADSCs alternatively activates microglia and enhances neuropathological deficits in AD mice. They proved the roles of intracerebral ADSC transplantation in AD pathology and spatial learning/memory of APP/PS1 double transgenic mouse model of AD. Results showed that after ADSC transplantation, Aβ peptide deposition showed a significant reduction and the learning/memory function in APP/PS1 transgenic mice was notably restored. After ADSC transplantation, more activated microglia in both regions of the hippocampus and the cortex t were preferentially surrounded and infiltrated into plaques. The activated microglia exhibited an alternatively activated phenotype, as indicated by their decreased expression levels of pro-inflammatory factors and elevated expression levels of alternative activation markers, as well as Aβ-degrading enzymes. In conclusion, ADSC transplantation could regulate microglial activation in AD mice, mitigate AD symptoms, and reduce cognitive decline, all of which indicate ADSC transplantation serving as a potential target for AD therapy [90].

5.10. ADSC Transplantation in Parkinson's Disease (PD)

MSC promote neuroprotection and neurogenesis, which had greatly beneficial effects on neurodegenerative disorders, such as Parkinson's disease. Soon et al. explored therapeutic potentials of ADSCs on the mouse model of PD. The consistency and high reliability of the experimental results confirmed by animal models are a critical factor in the stability of stem cell transplantation for PD. The hADSCs were intravenously injected into the tail vein of a mouse model of PD induced by 6-hydroxydopamine. The results showed that the behavioral performances were significantly improved in the 3rd week after the injection of hADSCs. Additionally, dopaminergic neurons were rescued, the number of structure-modified mitochondria was decreased, and mitochondrial complex I activity was recovered in the brains of the hADSC-injected PD mouse model. Overall, this study demonstrated that intravenous transplantation of hADSC may have promising strategy for PD by recovering mitochondrial functions [91]. Schwerk et al. demonstrated that ADSCs induce long-term neurogenic and anti-inflammatory effects and ameliorate cognitive in a rat model of PD. ADSCs were transplanted one week after 6-hydroxydopamine lesioning. The findings of this study revealed that ADSCs were found around the substantia nigra and the arachnoid mater, pericyte and endothelial markers were expressed. In addition, ADSC upregulated neurogenesis in hippocampal and subventricular regions, and promoted memory functioning. The results showed that the effects of ADSC transplantation could stay over 6 months, resulting in improvements on cognitive performance, elevated TH levels, subventricular and hippocampal neurogenesis and increased the content of anti-inflammatory factors, suggesting ADSCs exerted a neuroprotective impact on PD rat models, along with the potential of inducing endogenous repair programs in both neurogenic niches of the adult brain, potentially resulting in advancement in nonmotor symptoms of PD, which affect patients early and hence represent a potential disease-modifying target. Considering that hyposmia and loss of memory function are two major non-motor symptoms in PD, transplants with modulatory influences on the hippocampus and subventricular zone could provide a promising therapy [92].
5.11. ADSC Transplantation in Melanoma

ADSCs act as attractive tools for cancer gene therapy because of their intrinsic tropism to the tumor environment. Jing et al. explored that ADSCs could accelerate tumor necrosis factor-related apoptosis-inducing ligand (TRAIL) expression in melanoma treatment in vitro. This study indicated the ability of ADSC-harborred human TRAIL cDNA to enhance TRAIL expression and induce A375 melanoma cell apoptosis. The findings of transwell co-culture system demonstrated that TRAIL-ADSCs could induce A375 cell apoptosis in a dose-dependent manner. In conclusion, the finding from this study proved that preclinical support of ADSCs promoting TRAIL expression in the treatment for melanoma. However, further studies are required to discuss and confirm the in vivo ability of TRAIL-ADSCs in therapy of melanoma [93]. V Bahrambeigi et al. investigated that genetically modified ADSCs (GM-ADSCs) expressing interleukin-2 enhances b16f10 melanoma cell proliferation. IL2 is regarded as a critical regulatory molecule, which promotes the activity and growth of the immune effector cell function. It is reported that high-dose IL2 therapy is a choice for treatment of malignant melanoma but has some adverse effects. Before vivo studies, they proved that IL2 expressed by GM-ADSCs may serve as a growth factor for melanoma cells because of the elevated viability and decreased apoptosis of melanoma cells after in vitro treatment. Subcutaneous co-injection of IL2-expressing ADSCs with melanoma cells significantly increased the melanoma tumor growth. Moreover, in models of pulmonary metastases, melanoma cells were injected intravenously and 10 days later mice were treated by systematical injection of GM-ADSCs. Intravenously injected IL2-ADSCs transplanted into melanoma lung tumors but were unable to decrease melanoma lung metastases. Therefore, this study showed that IL2-expressing ADSCs can enhance the ability of B16F10 melanoma cell proliferation. However, the details of its antitumor action should be discussed in the future studies [94]. In addition, Lee et al. explored that the effects of ADSC-conditioned medium (ADSC-CM) on the proliferation and migration of B16 melanoma cells. The results of MTT assay revealed that ADSC-CM evidently decreased the proliferation of B16 melanoma cells and cell cycle analysis indicated that ADSC-CM notably increased the number of cells in G1 phase while the number of cells in the S and G2/M phases significantly decreased. Moreover, a wound migration model demonstrated that ADSC-CM treatment significantly decreased the migration ability of B16 melanoma cells. In addition, C57BL/6 mice were administered with a single intratumoral injection of ADSC-CM, daily or every other day, and a significant reduction in the volume of the tumor mass was observed in comparison to that of the control group. Thus, the findings of the present study indicated that ADSC-CM has an anti-tumorigenic effect on B16 melanoma cells in vitro and in vivo, and may potentially be used to support the treatment of melanoma in the future. However, the mechanism of this ADSC translation therapy can exert adverse effects locally and consistently. Further studies particularly combined with the use of clinical tumor samples are required [95].

6. Perspective

Because ADSCs can be collected in large numbers and their safety and efficacy have been demonstrated, their applications have made advancements in the clinical field. However, the accumulating evidence of ADSC therapy in safety and efficacy is restricted to anecdotal studies or phase I/II clinical trials.

Although ADSC therapy acts as a potential strategy to treatment of various diseases, it is important that some risks of malignancies are required to be assessed before engaging in a novel ADSC-based therapy, which is a bottleneck that restricts its clinical development. Recent studies have shown that MSCs enhance cancer formation, increase tumor volume, and promote new blood vessel formation and cancer invasion, though some previous studies indicated the roles of MSCs in anti-carcinogenesis, suppressing proliferation and promoting apoptosis. Therefore, their applications in cancer treatment were limited by the pro-tumorigenic effects of MSCs [96]. Previous studies on have investigated that many cytokines were expressed by MSCs to promote cancer cell growth, proliferation, metastasis to other tissues, etc. [97]. It has been suggested that the properties of proangiogenic, anti-apoptotic and immunomodulatory may act together as tumor promoters, which raising significant safety concerns [98]. Few articles specifically address the effects of ADSCs on tumor microenvironment. For example, there are gaps in the available papers regarding potential hypoxic mechanisms in association with ADSCs, as well as little discussion addressing the role of ADSCs in gynecologic malignancies, such as endometrial cancer. Because adipose tissue plays a key role in endometrial cancer development and has not been comprehensively evaluated in patients with endometrial cancer [98]. Therefore, it is still pending to make a definite conclusion on effect of ADSCs on pro-tumorigenesis. Furthermore, hADSCs can potentially lead to cancer recurrence by activating residual breast cancer cells persisting in tumor bed after mastectomy through promoting angiogenesis. For instance, chemokine ligand 1 (CXCL1) and chemokine ligand 8 (CXCL8) secreted by hADSCs could promote breast tumor growth by enhancing angiogenesis [99]. The clinical studies with ADSCs have not confirmed the potential risk of ADSCs in promoting tumor. Nevertheless, until now, because the follow-up periods in many of the published clinical studies are considered relatively lack, it may be too early to judge the safety of ADSCs. Therefore, future studies should make efforts to bridge this gap and identify potential mechanisms of ADSC transplantation in the occurrence, progression, growth and metastasis of cancer.

In addition, cell therapies with ADSCs in cosmetic and reconstructive surgery are already widely applied in clinical treatments as they provide more satisfied results, and the regenerative and therapeutic secretory properties of ADSCs are
promising in attenuating symptoms of chronic diseases. However, although positive effects of ADSCs have been reported, we still lack a complete set of clinical practice guidelines that includes the effect of postoperative evaluation criteria, so it is too early to evaluate the efficacy of cell-based with ADSCs. To date, as ADSC-based therapies are at the preliminary stage of clinical treatments, it seems that there are a few more years before ADSCs are considered as a standard strategy to treat some human diseases. Therefore, with more research efforts will be made into the study of ADSCs in both scientific and clinical settings, ADSCs may play critical roles in treating a wide variety of diseases.

**Funding:** None.

**Conflicts of Interest:** The author declares no conflict of interest.

**Copyright Statement**

©2020 the authors. This article is an open access article licensed under the terms and conditions of the [CREATIVE COMMONS ATTRIBUTION (CC BY) LICENSE](http://creativecommons.org/licenses/by/4.0/).

**References**

1. Arshad Z, Halioua-Haubold CL, Roberts M, Urso-Baiarda F, Branford OA, et al. Adipose-derived stem cells in aesthetic surgery: a mixed methods evaluation of the current clinical trial, intellectual property, and regulatory landscape. *Aesthetic Surgery Journal*, 2018, 38: 199–210.

2. Wang JM, Gu Y, Pan CJ, Yin LR. Isolation, culture and identification of human adipose-derived stem cells. *Experimental and Therapeutic Medicine*, 2017, 13: 1039–1043.

3. Kim EH, Heo CY. Current applications of adipose-derived stem cells and their future perspectives. *World Journal of Stem Cells*, 2014, 6: 65.

4. Suzuki E, Fujita D, Takahashi M, Oba S, Nishimatsu H. Adipose tissue-derived stem cells as a therapeutic tool for cardiovascular disease. *World Journal of Cardiology*, 2015, 7: 454.

5. Capelli C, Zaccara E, Cipriani P, Di Benedetto P, Maglion W, et al. Phenotypical and functional characteristics of in vitro-expanded adipose-derived mesenchymal stromal cells from patients with systemic sclerosis. *Cell Transplantation*, 2017, 26: 841–854.

6. Ardestishirajami A, Mossahebi - Mohammad M, Vakilian S, et al. Comparison of osteogenic differentiation potential of human adult stem cells loaded on bioceramic-coated electrospun poly (L-lactide) nanofibres. *Cell Proliferation*, 2015, 48: 47–58.

7. Lim MH, Ong WK, Sugii S. The current landscape of adipose-derived stem cells in clinical applications. *Expert Reviews in Molecular Medicine*, 2014, 16.

8. Li C, Wu X, Tong J, et al. Comparative analysis of human mesenchymal stem cells from bone marrow and adipose tissue under xeno-free conditions for cell therapy. *Stem Cell Research & Therapy*, 2015, 6: 55.

9. Hosseinpur Z, Hashemi SM, Salehi E, Ghazanfari T. Comparison of TGF-β1 and NO production by mesenchymal stem cells isolated from murine lung and adipose tissues. *Immunopharmacology and Immunotoxicology*, 2016, 38: 214–220.

10. Sheykhhasan M, Qomi RT, Ghiasi M. Fibrin scaffolds designing in order to human adipose-derived mesenchymal stem cells differentiation to chondrocytes in the presence of TGF-β3. *International Journal of Stem Cells*, 2015, 8: 219.

11. Lin HP, Chan TM, Fu RH, Chuu CP, Chiu SC, et al. Applicability of adipose-derived stem cells in type 1 diabetes mellitus. *Cell Transplantation*, 2015, 24: 521–532.

12. Wang B, Ma X, Zhao L, Zhou X, Ma Y, et al. Injection of basic fibroblast growth factor together with adipose-derived stem cell transplantation: improved cardiac remodeling and function in myocardial infarction. *Clinical and Experimental Medicine*, 2016, 16: 539–550.

13. Mohammadi Z, Afshari JT, Keramati MR, Alamdari DH, Ganjibakhsh M, et al. Differentiation of adipocytes and osteocytes from human adipose and placental mesenchymal stem cells. *Iranian Journal of Basic Medical Sciences*, 2015, 18: 259.

14. Platas J, Guillén ML, del Caz MDP, Gomar F, Castejón MA, et al. Paracrine effects of human adipose-derived mesenchymal stem cells in inflammatory stress-induced senescence features of osteoarthritic chondrocytes. *Aging* (Albany NY), 2016, 8: 1703.

15. Minter EM, Marra KG, Rubin JP. Adipose stem cells: biology, safety, regulation, and regenerative potential. *Clinics in Plastic Surgery*, 2015, 42: 169–179.

16. Chan TM, Chen JYR, Ho LI, Lin HP, Hsueh KW, et al. ADSC therapy in neurodegenerative disorders. *Cell Transplantation*, 2014, 23: 549–557.

17. ter Huurme M, Schelbergen R, Blattes R, Blom A, de Munter W, et al. Antiinflammatory and chondroprotective effects of intraarticular injection of adipose-derived stem cells in experimental osteoarthritis. *Arthritis & Rheumatism*, 2012, 64: 3604–3613.

18. McLaughlin MM, Marra KG. The use of adipose-derived stem cells as sheets for wound healing. *Organogenesis*, 2013, 9: 79–81.

19. Banyard DA, Salibian AA, Widgerow AD, Evan GRD. Implications for human adipose-derived stem cells in plastic surgery. *Journal of Cellular and Molecular Medicine*, 2015, 19: 21–30.

20. Sabado RL, Bhaward N. Directing dendritic cell immunotherapy towards successful cancer treatment. *Immunotherapy*, 2010, 2: 37–56.

21. Ivanova-Todorova E, Bochev I, Moudrjeva M, Dimitrov R, Bukarev D, et al. Adipose tissue-derived mesenchymal stem cells are more potent suppressors of dendritic cells differentiation compared to bone marrow-derived mesenchymal stem cells. *Immunology*
Letters, 2009, 126: 37–42.
22. Chagnon-Choquet J, Gauvin J, Roger J, Fontaine J, Poudrier J, et al. HIV Nef promotes expression of B-lymphocyte stimulator by blood dendritic cells during HIV infection in humans. The Journal of Infectious Diseases, 2015, 211: 1229–1240.
23. Kono M, Takagi Y, Kawauchi S, Wada A, Morikawa T, et al. Non-activated T and B lymphocytes become morphologically distinguishable after detergent treatment. Cytometry Part A, 2013, 83: 396–402.
24. Maseda D, Bonami RH, Crofford LJ. Regulation of B lymphocytes and plasma cells by innate immune mechanisms and stromal cells in rheumatoid arthritis. Expert Review of Clinical Immunology, 2014, 10: 747–762.
25. Bochev I, Elmadjian G, Kyurkchiev D, Tzvetanov L, Altankova I, et al. Mesenchymal stem cells from human bone marrow or adipose tissue differently modulate mitogen-stimulated B-cell immunoglobulin production in vitro. Cell Biology International, 2008, 32: 384–393.
26. Dmitrieva RI, Minullina IR, Bilibina AA, Tarasova OV, Anisimov SV, et al. Bone marrow-and subcutaneous adipose tissue-derived mesenchymal stem cells: differences and similarities. Cell Cycle, 2012, 11: 377–383.
27. Zhou Z, Chen Y, Zhang H, Min S, Yu B, et al. Comparison of mesenchymal stromal cells from human bone marrow and adipose tissue for the treatment of spinal cord injury. Cytoscape, 2013, 15: 434–444.
28. Nakanishi C, Nagaya N, Ohnishi S, Yamahara K, Takabatake S, et al. Gene and protein expression analysis of mesenchymal stem cells derived from rat adipose tissue and bone marrow. Circulation Journal, 2011, 11: 107061312–1107061312.
29. Safford KM, Hickok KC, Safford SD, Halvorsen YDC, Wilkison WO, et al. Neurogenic differentiation of murine and human adipose-derived stromal cells. Biochemical and Biophysical Research Communications, 2002, 294: 371–379.
30. Zavan B, Michelotto L, Lancerotto L, Puppa AD, D’Avella D, et al. Neural potential of a stem cell population in the adipose and cutaneous tissues. Neurological Research, 2010, 32: 47–54.
31. Zhang HT, Liu ZL, Yao XQ, Yang JZ, Xu RX. Neural differentiation ability of mesenchymal stromal cells from bone marrow and adipose tissue: a comparative study. Cytoscape, 2012, 14: 1203–1214.
32. Ko MS, Jung JY, Shin IS, Choi EW, Kim JH, et al. Effects of expanded human adipose tissue-derived mesenchymal stem cells on the viability of cryopreserved fat grafts in the nude mouse. International Journal of Medical Sciences, 2011, 8: 231.
33. Tomita K, Madura T, Sakai Y, Yano K, Terengh G, et al. Glial differentiation of human adipose-derived stem cells: Implications for cell-based transplantation therapy. Neuroscience, 2013, 236: 55–65.
34. Zhang R, Liu Y, Yan K, Chen L, Chen XR, et al. Anti-inflammatory and immunomodulatory mechanisms of mesenchymal stem cell transplantation in experimental traumatic brain injury. Journal of Neuroinflammation, 2013, 10: 871.
35. Ji WC, Li M, Jiang WT, Ma X, Li J. Protective effect of brain-derived neurotrophic factor and neurotrophin-3 overexpression by adipose-derived stem cells combined with silk fibroin/chitosan scaffold in spinal cord injury. Neurological Research, 2020: 1–11.
36. Tang L, Lu X, Zhu R, Qian T, Tao Y, et al. Adipose-derived stem cells expressing the neurogenin-2 promote functional recovery after spinal cord injury in rat. Cellular and Molecular Neurobiology, 2016, 36(5): 657–667.
37. Jiang W, Liang G, Li X, Li Z, Gao X, et al. Intracarotid transplantation of autologous adipose-derived mesenchymal stem cells significantly improves neurological deficits in rats after MCAo. Journal of Materials Science: Materials in Medicine, 2014, 25: 1357–1366.
38. Ryu HH, Kang BJ, Park SS, Kim Y, Sung GI, et al. Comparison of mesenchymal stem cells derived from fat, bone marrow, Wharton’s jelly, and umbilical cord blood for treating spinal cord injuries in dogs. Journal of Veterinary Medical Science, 2012: 12-0065.
39. Kang SK, Shin MJ, Jung JS, Kim YG, Kim CH. Autologous adipose tissue-derived stromal cells for treatment of spinal cord injury. Stem Cells and Development, 2006, 15: 583–594.
40. Ikegame Y, Yamashita K, Hayashi SI, Mizuno H, Tawada M, et al. Comparison of mesenchymal stem cells from adipose tissue and bone marrow for ischemic stroke therapy. Cytoscape, 2011, 13: 675–685.
41. Kolar MK, Kingham PJ, Novikova LN, Wiberg M, Novikov LN. The therapeutic effects of human adipose-derived stem cells in a rat cerebral spinal cord injury model. Stem Cells and Development, 2014, 23: 1659–1674.
42. Leu S, Lin YC, Yuen CM, Yen CH, Kao YH, et al. Adipose-derived mesenchymal stem cells markedly attenuate brain infarct size and improve neurological function in rats. Journal of Translational Medicine, 2010, 8: 63.
43. Kim JM, Lee ST, Chu K, Jung KH, Song EC, et al. Systemic transplantation of human adipose stem cells attenuated cerebral inflammation and degeneration in a hemorrhagic stroke model. Brain Research, 2007, 1183: 43–50.
44. Yang L, Fang JS, Wang W, Chen RK, Shen CF. Transplantation of Schwann cells differentiated from adipose-derived stem cells modifies reactive gliosis after contusion brain injury in rats. Journal of International Medical Research, 2011, 39: 1344–1357.
45. Constantin G, Marconi S, Rossi B, Angiari S, Calderan L, et al. Adipose-derived mesenchymal stem cells ameliorate chronic experimental autoimmune encephalomyelitis. Stem Cells, 2009, 27: 2624–2635.
46. di Summa PG, Kingham PJ, Raffoul W, Wiberg M, Terenghi G, et al. Adipose-derived stem cells enhance peripheral nerve regeneration. Journal of Plastic, Reconstructive & Aesthetic Surgery, 2010, 63: 1544–1552.
47. Lopatina T, Kalinina N, Karagyaur M, Stambolsky D, Rubina K, et al. Adipose-derived stem cells stimulate regeneration of peripheral nerves: BDNF secreted by these cells promotes nerve healing and axon growth de novo. PloS One, 2011, 6: e17899.
48. Zack-Williams DL, Butler PE, Kalaskar DM. Current progress in use of adipose derived stem cells in peripheral nerve regeneration. World Journal of Stem Cells, 2015, 7: 51.
49. Moon MH, Kim SY, Kim YJ, Kim SJ, Lee JB, et al. Human adipose tissue-derived mesenchymal stem cells improve postnatal
neovascularization in a mouse model of hindlimb ischemia. *Cellular Physiology and Biochemistry*, 2006, 17: 279–290.

50. Kim YJ, Kim HK, Cho HH, Bae YC, Suh KT, et al. Direct comparison of human mesenchymal stem cells derived from adipose tissues and bone marrow in mediating neovascularization in response to vascular ischemia. *Cellular Physiology and Biochemistry*, 2007, 20: 867–876.

51. Bhang SH, Cho SW, La WG, Lee TJ, Yang HS, et al. Angiogenesis in ischemic tissue produced by spheroid grafting of human adipose-derived stromal cells. *Biomaterials*, 2011, 32: 2734–2747.

52. Ning H, Liu G, Lin G, Yang R, Lu T, et al. Fibroblast growth factor 2 promotes endothelial differentiation of adipose tissue-derived stem cells. *The Journal of Sexual Medicine*, 2009, 6: 967–979.

53. Konno M, Hamazaki TS, Fukuda S, Tokuhara M, Uchiyama H, et al. Efficiently differentiating vascular endothelial cells from adipose tissue-derived mesenchymal stem cells in serum-free culture. *Biochemical and Biophysical Research Communications*, 2010, 4: 461–465.

54. Valina C, Pinkernell K, Song YH, Bai X, Sadat S, et al. Intracoronary administration of autologous adipose tissue-derived stem cells improves left ventricular function, perfusion, and remodeling after acute myocardial infarction. *European Heart Journal*, 2007, 28: 2667–2677.

55. van der Bogt KEA, Schreper S, Yu J, Sheikh AY, Hoyt G, et al. Comparison of transplantation of adipose tissue-and bone marrow-derived mesenchymal stem cells in the infarcted heart. *Transplantation*, 2009, 87: 642.

56. Planat-Benard V, Menard C, Andrè M, et al. Spontaneous cardiomyocyte differentiation from adipose tissue stroma cells. *Circulation Research*, 2004, 94: 223–229.

57. Rasmussen JG, Frøbert O, Holst-Hansen C, Kastrup J, Baandrup U, et al. Comparison of human adipose-derived stem cells and bone marrow-derived stem cells in a myocardial infarction model. *Cell Transplantation*, 2014, 23: 195–206.

58. Paik KY, Kim KH, Park JH, Lee JI, Kim OH, et al. A novel antifibrotic strategy utilizing conditioned media obtained from miR-150-transfected adipose-derived stem cells: validation of an animal model of liver fibrosis. *Experimental & Molecular Medicine*, 2020, 1–12.

59. Banas A, Teratani T, Yamamoto Y, Tokuhara M, Takeshita F, et al. IFATS collection: in vivo therapeutic potential of human adipose tissue mesenchymal stem cells in the rat. *Stem Cells*, 2008, 26: 2705–2712.

60. Banas A, Teratani T, Yamamoto Y, Tokuhara M, Takeshita T, et al. Rapid hepatic fate specification of adipose-derived stem cells and their therapeutic potential for liver failure. *Journal of Gastroenterology and Hepatology*, 2009, 24: 70–77.

61. Sun CK, Chang CL, Lin YC, Kao, YH, Chang, LT, et al. Systemic administration of autologous adipose-derived mesenchymal stem cells alleviates hepatic ischemia–reperfusion injury in rats. *Critical Care Medicine*, 2012, 40: 1279–1290.

62. Pan F, Liao N, Zheng Y, Wang Y, Gao Y, et al. Intrahepatic transplantation of adipose-derived stem cells attenuates the progression of non-alcoholic fatty liver disease in rats. *Molecular Medicine Reports*, 2015, 12: 3725–3733.

63. Chang Q, Li C, Lu Y, Geng R, Wei J, et al. Adipose-derived Mesenchymal Stromal Cells Suppress Osteoarthritis and Bone Erosion in Collagen-induced Arthritis. *Scandinavian Journal of Immunology*, 2020, e12877.

64. Ragni E, Perucca Orfei C, De Luca P, Colombini A, Viganò M, et al. Secreted Factors and EV-miRNAs Orchestrate the Healing Capacity of Adipose Mesenchymal Stem Cells for the Treatment of Knee Osteoarthritis. *International Journal of Molecular Sciences*, 2020, 21: 1582.

65. Cowan CM, Shi YY, Aalam OI, Chou YF, Mari C, et al. Adipose-derived adult stromal cells heal critical-size mouse calvarial defects. *Nature Biotechnology*, 2004, 22: 560–567.

66. Di Bella C, Farlie P, Penington AJ. Bone regeneration in a rabbit critical-sized skull defect using autologous adipose-derived cells. *Tissue Engineering Part A*, 2008, 14: 483–490.

67. Lopez MJ, McIntosh KR, Spencer ND, Borneman JN, Horswell R, et al. Acceleration of spinal fusion using syngeneic and allogeneic adult adipose derived stem cells in a rat model. *Journal of Orthopaedic Research*, 2009, 27: 366–373.

68. Peterson B, Zhang J, Iglesias R, Kabo M, Hedrick M, et al. Healing of critically sized femoral defects, using genetically modified mesenchymal stem cells from human adipose tissue. *Tissue Engineering*, 2005, 11: 120–129.

69. Shoji T, Li M, Mifune Y, Matsumoto T, Kawamoto A, et al. Local transplantation of human multipotent adipose-derived stem cells accelerates fracture healing via enhanced osteogenesis and angiogenesis. *Laboratory Investigation*, 2010, 90: 637–649.

70. Li M, Luo X, Lv X, Liu Y, Zhao G, et al. In vivo human adipose-derived mesenchymal stem cell tracking after intra-articular delivery in a rat osteoarthritis model. *Stem Cell Research & Therapy*, 2016, 7: 160.

71. González MA, Gonzalez-Rey E, Rico L, Büscher D, Delgado M. Treatment of experimental arthritis by inducing immune tolerance with human adipose-derived mesenchymal stem cells. *Arthritis & Rheumatism: Official Journal of the American College of Rheumatology*, 2009, 60: 1006–1019.

72. Uysal AC, Mizuno H. Differentiation of adipose-derived stem cells for tendon repair. In *Adipose-Derived Stem Cells*. Humana Press, Totowa, NJ, USA, 2011, 702: 443–451.

73. Lalozé J, Varin A, Bertheuil N, Groelle JL, Vaysse C, et al. Cell-assisted lipotransfer: Current concepts. *Annales de Chirurgie Plastique Esthétique*, 2017, 62: 609–616.

74. Arshad Z, Karmen L, Choudhary R, Smith JA, Branford OA, et al. Cell assisted lipotransfer in breast augmentation and reconstruction: A systematic review of safety, efficacy, use of patient reported outcomes and study quality. *JPRAS Open*, 2016, 10: 5–20.

75. Moseley TA, Zhu M, Hedrick MH. Adipose-derived stem and progenitor cells as fillers in plastic and reconstructive surgery.
99. Koelensperger E, Bonnert LC, Zoernig I, Marme F, Sandmann S, et al. The impact of human adipose tissue-derived stem cells on breast cancer cells: Implications for cell-assisted lipotransfers in breast reconstruction. Stem Cell Research & Therapy, 2017, 8: 121.

76. Hassan WU, Greiser U, Wang W. Role of adipose-derived stem cells in wound healing. Wound Repair and Regeneration, 2014, 22: 313–325.

82. Shingyoci Y, Orbay H, Mizuno H. Adipose-derived stem cells for wound repair and regeneration. Expert Opinion on Biological Therapy, 2015, 15: 1285–1292.

97. Duscher D, Barrera J, Wong VW, Marme F, Sandmann S, et al. Stem cells in wound healing: the future of regenerative medicine? A mini-review. Gerontology, 2016, 62: 216–225.

84. Garg RK, Rennert RC, Duscher D, Sorkin M, Kosaraju R, et al. Capillary force seeding of hydrogels for adipose-derived stem cell delivery in wounds. Stem Cells Translational Medicine, 2014, 3: 1079–1089.

78. Hassan WU, Greiser U, Wang W. Role of adipose-derived stem cells in wound healing. Wound Repair and Regeneration, 2014, 22: 313–325.

88. Torsvik A, Bjerkvig T. Adipose-derived mesenchymal stem cells alternatively induce long-term inflammatory effects and improve cognitive but not motor performance in a rat model of Parkinson's disease. Regenerative Medicine, 2015, 10: 431–446.

70. Cheng JT, Kao GS, Chiang YC, et al. Adipose-derived stem cells accelerate diabetic wound healing through the induction of autocrine and paracrine effects. Cell Transplantation, 2016, 25: 71–81.

83. Choi HS, Kim HJ, Oh JH, Park HG, Ra JC, et al. Therapeutic potentials of human adipose-derived stem cells on the mouse model of Parkinson’s disease. Neurobiology of Aging, 2015, 36: 2885–2892.

85. Schwerk A, Altschuler J, Roeh M, Gossen M, Winter C, et al. Adipose-derived human mesenchymal stem cells induce long-term neurogenic and anti-inflammatory effects and improve cognitive but not motor performance in a rat model of Parkinson's disease. Regenerative Medicine, 2015, 10: 431–446.

86. Teng M, Huang Y, Zhang H. Application of stem cells in wound healing - an update. Wound Repair and Regeneration, 2014, 22: 151–160.

75. Koellensperger E, Bonnert LC, Zoernig I, Marme F, Sandmann S, et al. The impact of human adipose tissue-derived stem cells on breast cancer cells: Implications for cell-assisted lipotransfers in breast reconstruction. Stem Cell Research & Therapy, 2017, 8: 121.

77. Hassan WU, Greiser U, Wang W. Role of adipose-derived stem cells in wound healing. Wound Repair and Regeneration, 2014, 22: 313–325.