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MINIREVIEWS

Current applications of adipose-derived stem cells and their future perspectives

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Abstract

Adult stem cells have a great potential to treat various diseases. For these cell-based therapies, adipose-derived stem cells (ADSCs) are one of the most promising stem cell types, including embryonic stem cells (ESCs) and induced pluripotent stem cells (iPSCs). ESCs and iPSCs have taken center stage due to their pluripotency. However, ESCs and iPSCs have limitations in ethical issues and in identification of characteristics, respectively. Unlike ESCs and iPSCs, ADSCs do not have such limitations and are not only easily obtained but also uniquely expandable. ADSCs can differentiate into adipocytes, osteoblasts, chondrocytes, myocytes and neurons under specific differentiation conditions, and these kinds of differentiation potential of ADSCs could be applied in regenerative medicine *e.g.,* skin reconstruction, bone and cartilage formation, *etc.* In this review, the current status of ADSC isolation, differentiation and their therapeutic applications are discussed.

Key words: Adipose-derived stem cells; Isolation; Differentiation; Transplantation; Cell-based therapy

Core tip: Adult stem cells have a great potential for reconstructive and regenerative medicine. Particularly, adipose-derived stem cells (ADSCs) are a promising useful cell source for cell-based therapy because of their capability of expansion and differentiation into special cell types. In this review, the current status of ADSC isolation, differentiation and their therapeutic applications are discussed.

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INTRODUCTION

Stem cells include embryonic stem cells (ESCs), induced pluripotent stem cells (iPSCs) and postnatal adult stem cells. ESCs are capable of self-renewal and differentiation into any cell type in the body. Induced PSCs are genetically reprogrammed somatic cells and have characteristics of ESCs but it is still unknown what the differences between ESCs and iPSCs are. Because of ethical and political concerns, it is difficult to apply ESCs in clinical research and practice but iPSCs and postnatal adult stem cells do not have such problems. Among postnatal adult stem cells, adipose-derived stem cells (ADSCs) are one of the most promising stem cell types. They can be easily obtained from liposuction aspirates or subcutaneous adipose tissue fragments and expanded *in vitro* and there are no ethical concerns like human ESCs for their use in diverse clinical applications.

ADSCs are found in any type of white adipose tissue, including subcutaneous and omental fat[1]. To obtain adipose tissue for ADSC isolation, liposuction is a safe

process with a low complication rate[2]. Shiffman *et al*[3] reported that 90%-100% of adipocytes from lipoaspirate are intact after autologous fat transplantation. The isolated ADSCs can be expanded vigorously until they enter into the differentiation process to specific cell lineages. ADSCs are capable of differentiating into adipocytes, osteoblasts, chondrocytes, myocytes *etc*., *in vitro* and genetically stable in long-term culture. Thus, ADSCs would be a valuable stem cell source for clinical use, with fewer restrictions compared to other cell sources.

ISOLATION AND CULTURE OF ADSCs

Stem cells derived from adipose tissue show higher yields compared with other stem cell sources. Currently, ADSCs could be isolated not only manually but also automatically using automatic centrifuge for cell isolation specialized in cells from adipose tissue.

To isolate stem cells from adipose tissue, current methods rely on a collagenase digestion followed by centrifugal separation. They display a fibroblast-like morphology and lack intracellular lipid droplets seen in adipocytes. Isolated ADSCs are typically expanded in a monolayer on standard tissue culture plastics with a basal medium containing 10% fetal bovine serum[4].

DIFFERENTIATION POTENTIAL

ADSCs are multipotent and can differentiate into adipocytes[5-7], osteoblasts[5,8], chondrocytes[5], myocytes[5,9] and neuronal cells[10].

For induction of adipogenic differentiation, dexamethasone, insulin and isobutylmethylxanthine are needed[5]. Adipogenic differentiation status can be evaluated by Oil Red O staining.

Differentiation into osteoblasts can be induced by dexamethasone, ascorbic acid and glycerophosphate and identified using Alizarin red which stains calcified extracellular matrix in the osteoblasts or alkaline phosphatase[11].

Induction of chondrocyte differentiation is carried out by the addition of insulin, transforming growth factor beta 1 and ascorbic acid. The chondrocyte differentiation can be assessed by safranin O or toluidine blue staining.

ADSCs differentiate into myocytes in media supplemented with hydrocortisone and dexamethasone usually. ADSCs can also differentiate in a medium which is composed of control medium supplemented with horse serum and hydrocortisone and expresses myoD1 and myosin heavy chain[12,13]. The differentiated cells form myotubules and express myosin light chain kinase in addition to other markers characteristic of the myocyte lineage[13].

Recently, ADSCs have also been induced to differentiate into neuronal cells. The composition of the neuronal induction medium is basal medium with butylated hydroxyanisole, retinoic acid, epidermal growth factor and basic fibroblast growth factor. The differentiated cells express neuronal markers for immature and mature neurons, such as βⅢ-tubulin, microtubule-associated protein 2, neuron specific enolase, synaptophysin and TAU[10].

CELL-BASED THERAPEUTIC APPLICATIONS OF ADSCs

Due to multipotency of the ADSCs, they can be used widely in various clinical applications. Unlike ESCs, ADSCs lack the ability to form all tissues or organs of the body and regenerate an entire living organism. Inducing differentiation of ADSCs requires potent doses of growth factors *in vitro*. ADSCs do not easily transform to mature cell types without strong signaling and they tend to resist differentiation *in situ*. The mechanisms for signaling ADSC differentiation to a mature adipocyte within a native adipose deposit are not well understood[14,15].

Adipocytes derived from ADSCs have uses in soft tissue defects, postmastectomy repair, lipodystrophy and soft tissue cosmetic applications, like anti-contour defects and anti-wrinkles. For soft tissue regeneration, autologous fat grafts have been widely used; however, several limitations still remain. One of the limitations is the poor long-term graft retention. The transplanted fat grafts can lose its volume over time due to tissue resorption that can results in the loss of 20%-90% of the original transplanted grafts volume[16]. The soft tissue regeneration would be more effective if the defect volume is filled. To fill the soft tissue defects, vasculature for supplying nutrition to the grafted tissue is needed. ADSCs could help the neovascularization by vascular endothelial growth factor (VEGF) secretion and adipocyte and fibroblast regeneration by their differentiation potential.

Chondrocytes differentiated from ADSCs express extracellular matrix components which are localized in cartilage and maintain their phenotype *in vivo*[17]. Chondrocyte derived ADSCs show practical possibilities for applications in repair of articular cartilage defects, such as osteoarthritis, in the future.

For bone repair, isolated ADSCs can be induced to differentiate into osteoblasts which are able to mineralize their extracellular matrix and express genes and proteins associated with a bone phenotype[18]. Osteoblasts or precursors of osteoblasts derived from ADSCs are able to be applied not only as cell materials, but also in combination with scaffold to a bone defect site[19].

Various cardiovascular diseases are the leading causes of mortality worldwide. Growing evidence indicates that injection of ADSCs improves cardiac function *via* the differentiation into cardiomyocytes and vascular cells and through paracrine pathways. Paracrine factors secreted by injected ADSCs enhance angiogenesis, reduce cell apoptosis rates, and promote neuron sprouts in damaged myocardium[20-23]. Danoviz *et al*[22] showed the effects of co-injecting ADSCs with biopolymers on cell cardiac retention, ventricular morphometry and performance in a rat model of myocardial infarction. They could confirm that intramyocardial injection of ADSCs mitigates the negative cardiac remodeling and preserves ventricular function post myocardial infarction. These findings sug-

gest important implications for the design of future cell therapy strategies for cardiac repair.

For treatment of neurodegenerative diseases, various stem cell types are under investigation. Stem cells are able to differentiate into neurons[24,25] and glial cells[26,27]. Similarly to other stem cell types, ADSCs have been known to have a differentiation potential into neuronal and glial cells[28,29] and are capable of promoting neuronal healing by secretion of some nerve growth factors. ADSCs express a significantly high proportion of nestin, which is a marker for neural progenitor cells[30]. ADSCs can secrete angiogenetic factors such as VEGF[31] and some neuroprotective factors such as insulin-like growth factor 1, the major factor that mediates protection against serum and potassium deprivation-induced apoptosis of cerebellar granule neurons[32]. Limitations in protocols to establish homogeneous populations of neural progenitors and stem cells still need to be resolved for effective therapy for neurodegenerative diseases like Parkinson's disease, multiple sclerosis and Alzheimer's disease[33].

PERSPECTIVES

Stem cells would be a useful tool for cell-based therapies for diverse diseases. A number of challenges still remain for cell-based therapies using stem cells. Safety issues in clinical use of stem cells expanded *in vitro*, development of differentiation protocol and *in vivo* delivery method, and problems of immune response in allogeneic transplantation are some to be overcome.

Because ADSCs can be harvested in large numbers and have shown evidence of safety and efficacy, their use is currently increasing in clinical fields. For ADSC culture, a whole adipose-derived stromal vascular fraction is usually used which is a heterogeneous mixture of various cell populations, including ADSCs. However, suitable cell surface markers can identify an ADSC population and the positively marked ADSCs can be separated by a cell sorting experiment. ADSCs purified by the specific cell surface markers would differentiate more efficiently into targeted cell types and make it easier to evaluate their influences on the therapeutic effects. Development of culture media compositions without animal origins is also an important aspect. This problem could be resolved by technology of recombinant proteins and cryopreservation methods of ADSCs over long time periods would be also useful.

Recently, many of these aspects have been considered and investigated and the progress to overcome such limitations would lead to applying stem cells, including ADSCs, widely in clinical practice in the future.

REFERENCES

1 **Locke M**, Windsor J, Dunbar PR. Human adipose-derived stem cells: isolation, characterization and applications in surgery. *ANZ J Surg* 2009; **79**: 235-244 [PMID: 19432707 DOI: 10.1111/j.1445-2197.2009.04852.x]

2 **Housman TS**, Lawrence N, Mellen BG, George MN, Filippo JS, Cerveny KA, DeMarco M, Feldman SR, Fleischer AB. The safety of liposuction: results of a national survey. *Dermatol Surg* 2002; **28**: 971-978 [PMID: 12460288 DOI: 10.1046/j.1524-4725.2002.02081.x]

3 **Shiffman MA**, Mirrafati S. Fat transfer techniques: the effect of harvest and transfer methods on adipocyte viability and review of the literature. *Dermatol Surg* 2001; **27**: 819-826 [PMID: 11553171 DOI: 10.1046/j.1524-4725.2001.01062.x]

4 **Sterodimas A**, de Faria J, Nicaretta B, Pitanguy I. Tissue engineering with adipose-derived stem cells (ADSCs): current and future applications. *J Plast Reconstr Aesthet Surg* 2010; **63**: 1886-1892 [PMID: 19969517 DOI: 10.1016/j.bjps.2009.10.028]

5 **Zuk PA**, Zhu M, Mizuno H, Huang J, Futrell JW, Katz AJ, Benhaim P, Lorenz HP, Hedrick MH. Multilineage cells from human adipose tissue: implications for cell-based therapies. *Tissue Eng* 2001; **7**: 211-228 [PMID: 11304456 DOI: 10.1089/107632701300062859]

6 **Hong L**, Peptan IA, Colpan A, Daw JL. Adipose tissue engineering by human adipose-derived stromal cells. *Cells Tissues Organs* 2006; **183**: 133-140 [PMID: 17108684 DOI: 10.1159/000095987]

7 **De Ugarte DA**, Morizono K, Elbarbary A, Alfonso Z, Zuk PA, Zhu M, Dragoo JL, Ashjian P, Thomas B, Benhaim P, Chen I, Fraser J, Hedrick MH. Comparison of multi-lineage cells from human adipose tissue and bone marrow. *Cells Tissues Organs* 2003; **174**: 101-109 [PMID: 12835573 DOI: 10.1159/000071150]

8 **Seong JM**, Kim BC, Park JH, Kwon IK, Mantalaris A, Hwang YS. Stem cells in bone tissue engineering. *Biomed Mater* 2010; **5**: 062001 [PMID: 20924139 DOI: 10.1088/1748-6041/5/6/062001]

9 **Gwak SJ**, Bhang SH, Yang HS, Kim SS, Lee DH, Lee SH, Kim BS. In vitro cardiomyogenic differentiation of adipose-derived stromal cells using transforming growth factor-beta1. *Cell Biochem Funct* 2009; **27**: 148-154 [PMID: 19319827 DOI: 10.1002/cbf.1547]

10 **Cardozo AJ**, Gómez DE, Argibay PF. Neurogenic differentiation of human adipose-derived stem cells: relevance of different signaling molecules, transcription factors, and key marker genes. *Gene* 2012; **511**: 427-436 [PMID: 23000064 DOI: 10.1016/j.gene.2012.09.038]

11 **Abdel-Aal MM**, Tholpady S, Helm GA, Ogle RC. Bone differentiation from human lipoaspirate. *Egypt J Plast Reconstr Surg* 2008; **32**: 285-291

12 **Mizuno H**. The myogenic potential of human processed lipoaspirates - Part I: Morphological, immunohistochemical analysis and gene expression. *J Japan Soc Plast Reconstr Surg* 2001; **21**: 427-436

13 **Mizuno H**, Zuk PA, Zhu M, Lorenz HP, Benhaim P, Hedrick MH. Myogenic differentiation by human processed lipoaspirate cells. *Plast Reconstr Surg* 2002; **109**: 199-209; discussion 210-211 [PMID: 11786812 DOI: 10.1097/00006534-200201000-00030]

14 **Gimble JM**. Adipose tissue-derived therapeutics. *Expert Opin Biol Ther* 2003; **3**: 705-713 [PMID: 12880371 DOI: 10.1517/14712598.3.5.705]

15 **Brayfield C**, Marra K, Rubin JP. Adipose stem cells for soft tissue regeneration. *Handchir Mikrochir Plast Chir* 2010; **42**: 124-128 [PMID: 20352575 DOI: 10.1055/s-0030-1248269]

16 **Tobita M**, Orbay H, Mizuno H. Adipose-derived stem cells: current findings and future perspectives. *Discov Med* 2011; **11**: 160-170 [PMID: 21356171]

17 **Erickson GR**, Gimble JM, Franklin DM, Rice HE, Awad H, Guilak F. Chondrogenic potential of adipose tissue-derived stromal cells in vitro and in vivo. *Biochem Biophys Res Commun* 2002; **290**: 763-769 [PMID: 11785965 DOI: 10.1006/bbrc.2001.6270]

18 **Halvorsen YD**, Franklin D, Bond AL, Hitt DC, Auchter C, Boskey AL, Paschalis EP, Wilkison WO, Gimble JM. Extracellular matrix mineralization and osteoblast gene expression by human adipose tissue-derived stromal cells. *Tissue Eng* 2001; **7**: 729-741 [PMID: 11749730 DOI: 10.1089/10763270175

333768]
19 **Hicok KC**, Du Laney TV, Zhou YS, Halvorsen YD, Hitt DC, Cooper LF, Gimble JM. Human adipose-derived adult stem cells produce osteoid in vivo. *Tissue Eng* 2004; **10**: 371-380 [PMID: 15165454 DOI: 10.1089/107632704323061735]
20 **Bai X**, Alt E. Myocardial regeneration potential of adipose tissue-derived stem cells. *Biochem Biophys Res Commun* 2010; **401**: 321-326 [PMID: 20833143 DOI: 10.1016/j.bbrc.2010.09.012]
21 **Cai L**, Johnstone BH, Cook TG, Tan J, Fishbein MC, Chen PS, March KL. IFATS collection: Human adipose tissue-derived stem cells induce angiogenesis and nerve sprouting following myocardial infarction, in conjunction with potent preservation of cardiac function. *Stem Cells* 2009; **27**: 230-237 [PMID: 18772313 DOI: 10.1634/stemcells.2008-0273]
22 **Danoviz ME**, Nakamuta JS, Marques FL, dos Santos L, Alvarenga EC, dos Santos AA, Antonio EL, Schettert IT, Tucci PJ, Krieger JE. Rat adipose tissue-derived stem cells transplantation attenuates cardiac dysfunction post infarction and biopolymers enhance cell retention. *PLoS One* 2010; **5**: e12077 [PMID: 20711471 DOI: 10.1371/journal.pone.0012077]
23 **van der Bogt KE**, Schrepfer S, Yu J, Sheikh AY, Hoyt G, Govaert JA, Velotta JB, Contag CH, Robbins RC, Wu JC. Comparison of transplantation of adipose tissue- and bone marrow-derived mesenchymal stem cells in the infarcted heart. *Transplantation* 2009; **87**: 642-652 [PMID: 19295307 DOI: 10.1097/TP.0b013e31819609d9]
24 **Dezawa M**, Takahashi I, Esaki M, Takano M, Sawada H. Sciatic nerve regeneration in rats induced by transplantation of in vitro differentiated bone-marrow stromal cells. *Eur J Neurosci* 2001; **14**: 1771-1776 [PMID: 11860471 DOI: 10.1046/j.0953-816x.2001.01814.x]
25 **Cho MS**, Lee YE, Kim JY, Chung S, Cho YH, Kim DS, Kang SM, Lee H, Kim MH, Kim JH, Leem JW, Oh SK, Choi YM, Hwang DY, Chang JW, Kim DW. Highly efficient and large-scale generation of functional dopamine neurons from human embryonic stem cells. *Proc Natl Acad Sci USA* 2008; **105**: 3392-3397 [PMID: 18305158 DOI: 10.1073/pnas.0712359105]
26 **Chi GF**, Kim MR, Kim DW, Jiang MH, Son Y. Schwann cells differentiated from spheroid-forming cells of rat subcutaneous fat tissue myelinate axons in the spinal cord injury. *Exp Neurol* 2010; **222**: 304-317 [PMID: 20083105 DOI: 10.1016/j.expneurol.2010.01.008]
27 **Kang SM**, Cho MS, Seo H, Yoon CJ, Oh SK, Choi YM, Kim DW. Efficient induction of oligodendrocytes from human embryonic stem cells. *Stem Cells* 2007; **25**: 419-424 [PMID: 17053214 DOI: 10.1634/stemcells.2005-0482]
28 **Jiang L**, Zhu JK, Liu XL, Xiang P, Hu J, Yu WH. Differentiation of rat adipose tissue-derived stem cells into Schwann-like cells in vitro. *Neuroreport* 2008; **19**: 1015-1019 [PMID: 18580571 DOI: 10.1097/WNR.0b013e3283040efc]
29 **Ryu HH**, Lim JH, Byeon YE, Park JR, Seo MS, Lee YW, Kim WH, Kang KS, Kweon OK. Functional recovery and neural differentiation after transplantation of allogenic adipose-derived stem cells in a canine model of acute spinal cord injury. *J Vet Sci* 2009; **10**: 273-284 [PMID: 19934591 DOI: 10.4142/jvs.2009.10.4.273]
30 **Safford KM**, Hicok KC, Safford SD, Halvorsen YD, Wilkison WO, Gimble JM, Rice HE. Neurogenic differentiation of murine and human adipose-derived stromal cells. *Biochem Biophys Res Commun* 2002; **294**: 371-379 [PMID: 12051722 DOI: 10.1016/s0006291x02004692]
31 **Rehman J**, Traktuev D, Li J, Merfeld-Clauss S, Temm-Grove CJ, Bovenkerk JE, Pell CL, Johnstone BH, Considine RV, March KL. Secretion of angiogenic and antiapoptotic factors by human adipose stromal cells. *Circulation* 2004; **109**: 1292-1298 [PMID: 14993122 DOI: 10.1161/01.CIR.0000121425.42966.F1]
32 **Zavan B**, Vindigni V, Gardin C, D'Avella D, Della Puppa A, Abatangelo G, Cortivo R. Neural potential of adipose stem cells. *Discov Med* 2010; **10**: 37-43 [PMID: 20670597]
33 **Taupin P**. Adult neurogenesis, neural stem cells and Alzheimer's disease: developments, limitations, problems and promises. *Curr Alzheimer Res* 2009; **6**: 461-470 [PMID: 19747153 DOI: 10.2174/156720509790147151]

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