



Harvesting, Isolation and Culture of Adipose Derived Stem Cells

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DESCRIPTION

Current methods for harvesting Adipose Derived Stem Cells (ADSCs) include aspiration, liposuction, and direct excision. The most commonly used method for the collection of adipose tissue is Coleman's aspiration technique, which relies on the minor negative pressure with a syringe. Also, the negative pressure of liposuction-related methods by motor could harvest a huge volumetric adipose tissue. Liposuction-related methods contain conventional, ultrasound-assisted liposuction, power-assisted liposuction, and laser-assisted. And direct removal could harvest a piece of adipose tissue, and the attained fragments need mincing into tiny particles with the use of surgical blades.

According to multiple variables the yield and properties of ADSCs may differ, such as the the adipose tissue depot, harvesting method, medical comorbidities of the patient, Body Mass Index (BMI), and age. There is proof that harvesting adipose tissue by aspiration splits the concentration of ADSCs compared with harvesting by excision. The yield and biological characteristics of viable ADSCs found by excision are expressively developed when compared with those obtained through liposuction. The gene expression formation and the tendency toward differentiation into a detailed germ layer can also be affected by the harvesting method. ADSCs collected through direct excision tend toward mesodermal and ectodermal differentiation, whereas those obtained by liposuction are more likely to differentiate into endoderm.

The location of collection also affects the yield and differentiation capability of ADSCs. There is some evidence presenting that the thigh can provide a better yield of ADSCs than the abdomen, waist, and inner knee. In contrast, there is no major difference in cell viability among the donor areas. ADSC yields and distinction potential are also stated to be higher in subcutaneous tissue than in instinctual depots. Moreover, the differentiation ability of ADSCs also depends on the characteristics of the donor, such as age, gender, and metabolic index. Older age, high BMI (>30 kg/m²), suffering from diabetes mellitus, or exposure to radiotherapy and endocrine therapy will decrease the proliferative and differentiation potential of ADSCs.

However, further research is required to define whether the *in vitro* and *in vivo* findings translate into clinically significant differences.

The most widely utilized method for isolating ADSCs was first proposed by Zuk and colleagues. This method includes extensive washing with Phosphate Buffered Saline (PBS) and digestion of lipoaspirate with 0.075% collagenase to release the Stromal Vascular Fraction (SVF) of cells. The SVF is incubated in the medium overnight at 37°C in an atmosphere with 5% CO₂ after a series of washes and centrifugation steps. Following incubation, the plates are extensively washed with PBS to eradicate residual, nonadherent red blood cells. The resulting cells are considered to be ADSCs.

Collagenase digestion remains the gold standard among the currently used methods for isolating ADSCs, although other enzymes, such as trypsin, clostripain, and dispase, can also be used. A latest study proposed that, even though trypsin-digested and collagenase-digested ADSCs current similar adipogenic differentiation and proliferative capability, the osteogenic differentiation potential of the trypsin-treated cells is up to sevenfold higher. Despite the widespread use of the above-mentioned methods for isolating ADSCs, enzymatic digestion-based methods have many disadvantages. The use of enzymes may change or disrupt cell viability and surface antigens, which may decrease ADSC regenerative potential, while question marks also remain regarding whether residual enzyme activity can affect safety. So, an increasing number of studies have explored economical enzyme-free methods for ADSC isolation, including new mechanical techniques or methods that do not rely on enzymatic activity or centrifugation.

CONCLUSION

The emergence of ADSC therapy offers a different means for tissue regeneration. Numerous clinical and preclinical studies have established the vital role of ADSCs in recreating and repairing target organs, such as bone, cartilage, myocardium, liver, nervous system, and skin. However, many safety issues need to be urgently addressed, from the preparation of ADSCs to their application.

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