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Modified protocol for improvement of differentiation potential of menstrual blood-derived stem cells into adipogenic lineage

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Abstract

Objectives: To characterize potency of menstrual blood-derived stem cells (MenSCs) for future cell therapies, we examined differentiation potential of MenSCs into adipocytes.

Materials and methods: Differentiation potential of MenSCs in comparison to bone marrow stem cells (BMSCs) was assessed in conventional culture medium. Differentiation potential of MenSCs into adipocytes was improved using different combinations of growth factors and hormones.

Results: First, we demonstrated that MenSCs preserve their appearance and karyotypic stability during passages. Although these cells express mesenchymal stem cells markers, they cannot simply be classified as mesenchymal stem cells due to expression of embryonic stem cells marker, OCT-4. Oil red O staining showed that differentiated MenSCs in conventional medium with/without retinoic acid (protocols 1 and 2) did not attain adipocyte characteristics, whereas differentiated BMSCs in conventional medium accumulated oil vacuoles typically. Nevertheless, real-time RT-PCR results showed that LPL gene expression was up-regulated in both protocols 1 and 2, whereas LEPR was up-regulated only in protocol 2 (fortified with retinoic acid). Surprisingly, protocol 3 (including rosiglitazone) had odd influence on mRNA expression of all genes (LEPR, LPL and PPAR-c). Oil red O staining confirmed fat-producing ability of MenSCs under protocol 3.

Conclusions: Presented data suggest an efficient differentiation protocol for in vitro production of MenSC-derived adipocytes. These cells are suggested to be an apt alternative to BMSCs for future stem cell therapy of soft tissue injuries.

Introduction

A large percentage of plastic and reconstructive surgical procedures are performed each year to restore soft tissue defects resulting from serious burns, tumour resection, and genetic and congenital defects (1). Strategies to restore soft tissue injuries consist of use of adipose tissue as an implant and filler (2). However, grafting adipose tissue has problems, including tissue resorption, immunological rejection and more (3). Due to these limitations, in recent decades, characteristics of stem cells such as self-renewal potential and trans-differentiation into other lineages (including adipocytes) have impelled scientists to take advantage of stem cell therapy as a novel therapeutic approach for soft tissue healing (4). Nevertheless, the main challenge in everyday clinical practice is to achieve high cell density from an easily accessible and safe stem cell resource.

Numerous ethical considerations surrounding use of embryonic stem cells (ESCs) have triggered the scientist's interest in adult stem cells (5). Their most conventional source has been bone marrow, cell obtained from the iliac crest. Trans-differentiation ability of bone marrow-derived stem cells (BMSCs) into adipocytes consistent with differentiation into other lineages such as

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potential of MenSCs into adipocytes has been compared to BMSCs using cytochemical and molecular experiment. To find an appropriate stimulus to trigger adipo-