

COMPARATIVE ANALYSIS OF MSCs OBTAINED FROM DIFFERENT FETAL ADNEXA TISSUES

Prata KL^{1,2}, De Santis GC^{1,2}, Orellana MD^{1,2}, Fernandes TR¹, Candido KS¹, Caruso SR^{1,2}, Silva ACG¹, Gonçalves NC¹, Bonaldo CCO¹, Santos DFD¹, Wagatsuma VMD¹, Palma PVB¹, Kashima S¹, Covas DT^{1,2}

¹ National Institute of Science and Technology in Stem Cell and Cell Therapy, Center for Cell Therapy and Regional Blood Center, Ribeirão Preto, Brazil

² Department of Internal Medicine, School of Medicine, University of São Paulo, Ribeirão Preto, Brazil

INTRODUCTION

In the last years, multipotent mesenchymal stromal cells (MSCs) have been considered as a therapeutic toll for the treatment of several diseases. In most cases, they are isolated from the bone marrow and cultured in an animal protein supplemented medium. This procedure causes discomfort to the donor and risks to the receptor, which can be avoided by using alternatives sources of MSCs cultivated in xenofree conditions. Fetal adnexa tissues may be the ideal source of MSCs to be cultivated for clinical purposes. So far, neither are there systematic studies comparing MSCs isolated from different tissues of fetal adnexa nor human protein supplements aiming receptor safety.

OBJECTIVES

The objectives of this study are to establish the best fetal adnexa source of MSCs and the best human protein supplement to expand MSCs for clinical purposes.

MATERIAL AND METHODS

The MSCs were isolated from amniotic (AM) and chorionic membranes (CM), placental cotyledons (PC) and umbilical cord (UC), and cultured in parallel in a medium supplemented with fetal bovine serum (FBS), human plasma converted in serum (HP) or platelet lysate (PL). Comparative assays with cells isolated from 4 sources and cultured in 3 different protein supplements were performed. Furthermore, the methods used to obtain the human protein supplements were established, as well as the process validation for cryopreservation of MSCs in vials and bags.

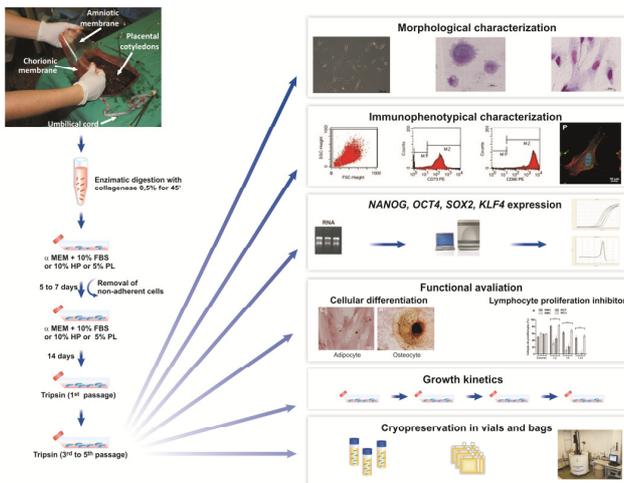


Figure 1. Methodology overview.

RESULTS

The cells showed typical MSCs morphology and immunophenotype, and were capable of differentiating into osteocytes and adipocytes. The transfusion residual risk of the medium supplemented with 5%LP was at least 2-fold higher than the one supplemented with 10%HP.

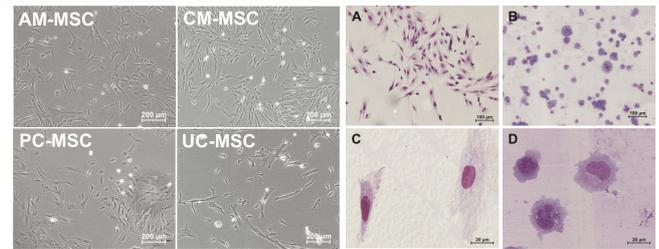


Figure 2: MSCs morphology.

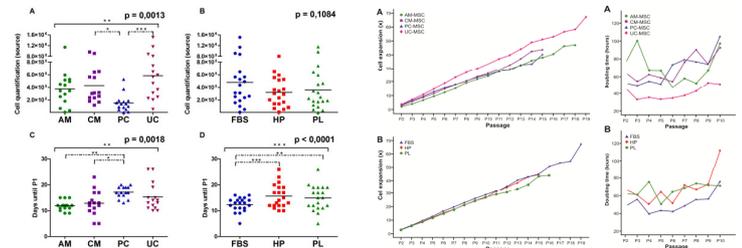


Figure 3: Isolation method efficiency and growth kinetics.

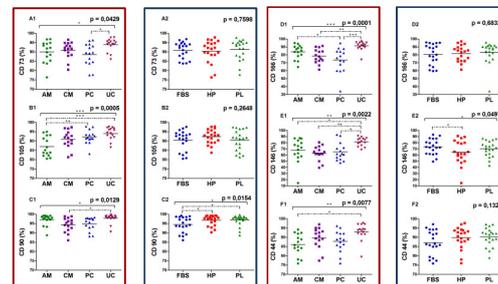


Figure 5: MSC immunophenotype.

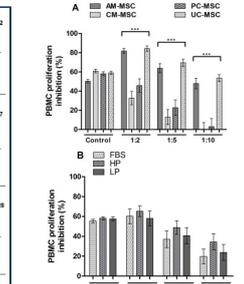


Figure 4: Lymphocyte proliferation inhibition

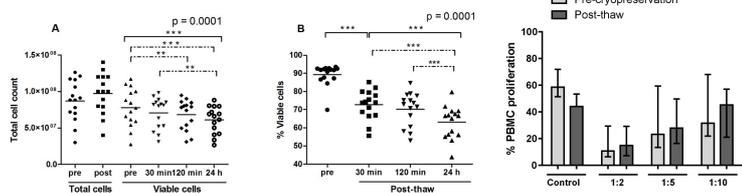


Figure 6: Cell viability and lymphocyte proliferation inhibition pre-cryopreservation and post-thaw.

CONCLUSIONS

It was possible to expand efficiently the MSCs in xenofree conditions without changing their characteristics. Considering transfusional residual risk and stock availability of raw materials necessary to obtain protein supplement, the HP was superior to the LP. Among the sources, the umbilical cord was the best one because the MSCs isolated from it showed an ideal association among the evaluated parameters such as cell yield, expansion potential, cell viability, purity and potency. Additionally, they presented a very good cell recuperation, viability and potency after the cryopreservation and thaw process.

Apoio: