QUALITY CONTROL AND SAFETY OF THE PROCUREMENT, PROCESSING, STORAGE, AND ADMINISTRATION OF MSCs FOR CELLULAR THERAPY

De Santis GC[1,2], Prata KL[1,2], Orellana MD[1,2], Candido KS[1], Caruso SR[1,2], Fernandes TR[1], Palma PVB[1],Kashima S[1], 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

The infusion of multipotent mesenchymal stromal cells (MSCs) is a promising therapy option to prevent or treat different diseases. It may be the first cell type able to be used as an "off-the-shelf" therapeutic product.

OBJECTIVES

The aim of this study is to describe our experience of 127 MSC infusions in 34 different patients done between June 2007 and August 2011.

MATERIAL AND METHODS

Bone marrow (BM) MSC cultures were derived from BM-aspirates collected from 18 donors selected according the current protocol for BM transplantation. For unrelated allogeneic use of the cells, the standards for blood donation were used. Clinical-grade MSCs were generated under good manufacturing practice conditions. BM mononuclear cells (MNC-BM) were obtained by Ficoll-HypaqueTM gradient centrifugation. MSCs were isolated by plastic adherence and expanded ex vivo by culturing in culture flasks (175 cm^2) with α-MEM enriched with 15% of fetal bovine serum (FBS). At confluence, MSCs were harvested, washed and resuspended in saline solution with albumin and infused through a membrane filter (170µm). Microbial contamination was assessed in each passage and immediately before the infusion.

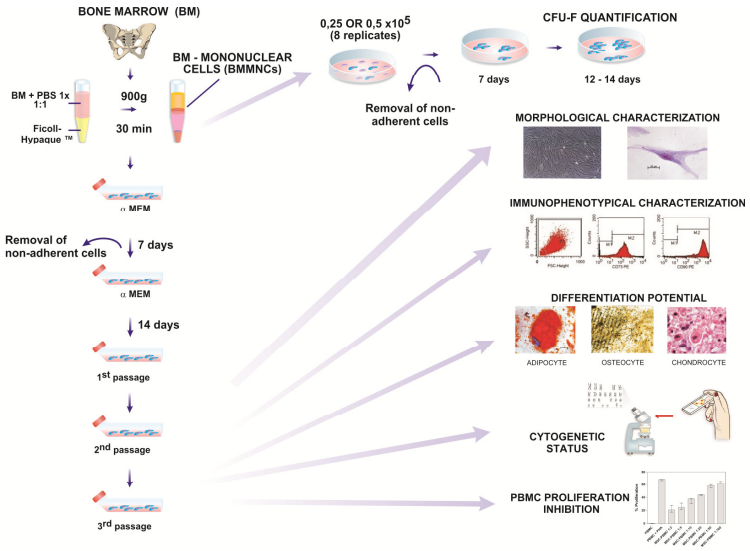

Figure 1. Methodology overview.

RESULTS

All cultured cells showed characteristic morphology and immunophenotype of normal MSCs and were capable of differentiating into adipocytes and osteocytes. MSCs were derived from either family [N=57 (45%) infusions] or 3rd party donors [n=70 (65%) infusions].

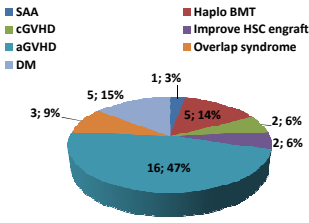

Figure 2: Indication of use

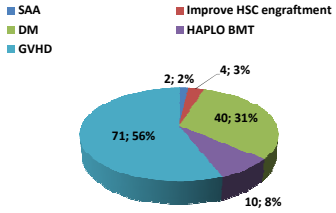

Figure 3: Infusion per disease

Table 1: Some parameters assessed during the processing of the cells.

PARAMETER	N	MEDIAN	MINIMUM	MAXIMUM
BM volume (mL)	18	108	21	266
MNCs-BM/flask	18	1.0×10^8	0.68×10^8	1.67×10^8
MSC/cm^2	18	5.76×10^5	5.14×10^5	9.54×10^5
CFU-F (per 10^6 MNC-BM)	11	76	36	128
Doubling time (hours)	18	71	32	365
Final cell expansion (fold)	18	4.33	2.22	11.52
Culture days	18	32	12	54
Passage number	18	3.5	2	5
PBMC proliferation inhibition [MSC:PBMC 1:2 + PHA] (%)	14	90.37	78.04	93.96

Table 2: Some parameters assessed for the MSCs preparations.

PARAMETER	N	MEAN	SD
Cell dose infused (10^6)	127	130.1	± 60.7
Cell dose infused (10^6/kg)	127	2.6	± 1.1
Cell (10^6) concentration/mL	127	1.5	± 0.7
Infusion duration (min)	110	18	± 9
Bag volume (mL)	127	85	± 23
Cell viability (%)	119	89.4	± 5.3

Cytogenetic status was evaluated (N=13) at the 2nd or 3rd passages and no evidence of clonal proliferation was observed. No severe adverse events were observed.

CONCLUSIONS

Based on these results, we considered that MSCs infusions are feasible and safe. However, the process of isolation and expansion of the cells takes time, which can cause some delay on the treatment for emergencies with freshly isolated cells. One possible solution to obtain the cells in a short time period could be the establishment of a process of cryopreservation of the MSCs in bags. These bags would be thawed and infused at any time needed. Moreover we recommend the use of a membrane filter (170µm) to avoid infusion of cell aggregates and more severe reactions like pulmonary embolism.

Apoio: