

UC Irvine

UC Irvine Previously Published Works

Title

Marrow Stromal Cell (MSC) Growth from Long Term Cryopreserved Bone Marrow.

Permalink

<https://escholarship.org/uc/item/50x6q0kr>

Journal

Blood, 108(11)

ISSN

0006-4971

Authors

Lane, Thomas A
Garls, Davina
Mackintosh, Ellen
et al.

Publication Date

2006-11-16

DOI

10.1182/blood.v108.11.5227.5227

Copyright Information

This work is made available under the terms of a Creative Commons Attribution License, available at <https://creativecommons.org/licenses/by/4.0/>

Peer reviewed



Marrow Stromal Cell (MSC) Growth from Long Term Cryopreserved Bone Marrow.

Thomas A. Lane, Davina Garls, Ellen Mackintosh, and Steven C. Cramer

Blood 2006 108:5227;

Article

Info & Metrics

Abstract

Autologous MSC are under active investigation for a variety of potential clinical indications in regenerative medicine and gene therapy. The potential to derive autologous MSC from long-term cryopreserved bone marrow has not been fully elucidated. We report here the growth characteristics of MSC from bone marrow that was cryopreserved for more than 10 years.

Methods: We identified 3 deceased patients whose cryopreserved autologous bone marrow that was originally intended for autologous transplant, was to be discarded after fulfilling all accepted criteria for discard. The pts were age 46, 47, and 60 at the time of harvest, one had high risk and 2 had metastatic breast cancer (both to bone) and all had had multiple chemotherapy regimens prior to harvest. The marrow was cryopreserved between 1994–1995 in 10% DMSO using a controlled-rate freezer and stored in the vapor phase of liquid nitrogen until June, 2006 (years). The marrow was thawed in a 37 C waterbath, diluted with an equal volume of PBS × 3 in PBS, resuspended in culture medium with 20% fetal bovine serum and plated at 5 × 10⁶

mononuclear cells (mnc)/cm² in 2 – 4 T175 flasks. Culture medium was changed in 48 hours and the plates were observed during subsequent culture medium changes every 3–4 days.

Results: At 2 – 4 days only scattered round cells were observed, but all samples grew scattered spindle-shaped cells were between days 4 – 12. Six of 8 primary cultures became > 70% confluent between days 11 and 21; 2 remained at < 40% confluence until replated (then achieving > 70% confluence at day 31). First passage cultures (n=12) became > 70% confluent in 4 – 10 days (median = 7) and 2nd passage cultures were > 70% confluent in 7 – 10 days (median = 9). Second or 3rd passage cultures yielded 2.1 – 4.2 × 10⁶ MSC/T175 (n= 14; median 3.15) and were 89 – 96 % viable. Flow cytometry (n = 3) confirmed that 3rd passage cells stained positive for CD105 and CD73 and negative for CD14 and CD45. Additional characterization of the differentiation potential of these presumptive MSC are in progress.

Conclusion: These studies indicate that cells typical for MSC can be readily grown from marrow that has been cryopreserved for > 10 years and suggest that even long-term stored bone marrow may serve as a source of cells for regenerative medicine.



Take **notes**, share and follow articles, make **comments**, and collaborate with peers!



DISCUSS



ADD TO LIST



FOLLOW



SHARE



COMMENT



NOTE

2006, The American Society of Hematology

Potential Articles of Interest

Marrow Stromal Cell (MSC) Growth from Long Term Cryopreserved Bone Marrow.

Thomas A. Lane et al., Blood, 2006

Feasibility of Rapidly Generating Sufficient Autologous Human Marrow Stromal Cells for Cellular Therapy.

Thomas A. Lane et al., Blood, 2007

Large-Scale, Bioreactor-Generated Expansion of Umbilical Cord Blood Tissue-Derived Mesenchymal Stromal Cells for Clinical Use

Lucila Nassif Kerbauy et al., Blood, 2017

Clinical-Scale Expansion of Human Bone Marrow-Derived Mesenchymal Stromal Cells to Treat Patients After Ischemic Stroke.

Patrick J Hanley et al., Blood, 2012

Regulation of glutamine synthetase in cultured 3T3-L1 cells by insulin, hydrocortisone, and dibutyryl cyclic AMP.

R E Miller et al., Proc Natl Acad Sci U S A, 1978

Murine bone marrow cell line producing colony-stimulating factor.

K Harigaya et al., Proc Natl Acad Sci U S A, 1981

Differential expression of SM1 and SM2 myosin isoforms in cultured vascular smooth muscle

P. Babij et al., American Journal of Physiology - Cell Physiology, 1992

Aged Human Multipotent Mesenchymal Stromal Cells Can Be Rejuvenated by Neuron-Derived Neurotrophic Factor and Improve Heart Function After Injury

Xiao-Yan Zhai et al., JACC: Basic To Translational Science, 2017