# EFFECT OF SINGLE PULSED ELECTROMAGNETIC FIELDS STIMULATION ON THE PROLIFERATION OF MESENCHYMAL STEM CELLS

Ming-Tzu Tsai, Walter H. Chang, Ru-Jyuan Hou, Dai-Wei Wu

Bone Tissue Engineering Research Center, Chung Yuan Christian University, TAIWAN, E-mail: g9102504@cycu.edu.tw

### INTRODUCTION

Mesenchymal stem cells (MSCs), which have the capacity of self-renewal and multipotent potential of differentiation, is the promising tool for bone tissue engineering, such as bone repair and reconstruction [1]. However, the origin and number of MSCs are still the problems in clinics. Based on the few population of MSCs contained from adult bone marrows, the main objective of this research was to enhance the recruitment of MSCs in vitro by using physical stimuli, single pulsed electromagnetic fields (sPEMF) stimulation. The activity of MSCs was estimated by MTT assay as well as the histomorphological stain.

### MATERIAL AND METHOD

Bone marrow MSCs primary culture. MSCs were isolated from bone marrows of bilateral femora and tibiae acquired from 4-6-week-old male Wistar rats. Isolated MSCs were resuspended in  $\alpha$ -MEM containing 10% FBS [2].

*sPEMF system.* sPEMF was well-established in our lab for many years. The parameters of sPEMF were: single pulse, 7.5 Hz, 1.3, 2.4, and 3.2 Gauss of magnetic intensities. The stimulation period of sPEMF was two hours a day for 14 days. *Experimental design.* Isolated bone marrow MSCs were cultured in 8-well chamber slides for two days, and then cells were exposed to sPEMF stimulation with specific parameters for 2 hr/day for 14 days. Conditioned medium and cells were collected and assayed at day 0, 3, 6, 9, 12, and 14.

*Proliferation and differentiation assay.* The proliferation and of MSCs was determined by MTT assay. The differentiation of osteoblasts and adipocytes from MSCs were evaluated by von Kossa stain and Oil Red stain.

## **RESULT AND DISCUSSION**

Fig. 1-2 showed von Kossa stain of osteoblasts and oil-red O stain of adipocytes differentiated from bone marrow MSCs. It indicated that bone marrow MSCs still had multipotent ability of differentiation after appropriate PEMF stimulation with specific parameters. Fig. 3-5 showed the proliferation of MSCs with three original MSCs densities with/without PEMF stimulation with different intensities. PEMF-50, PEMF-500, and PEMF-1000 maintained higher proliferation than their controls during 14-day period (Fig. 3). The effect of PEMF stimulation with 2.4 G and 3.2 G inhibited cell viability of MSCs, especially 3.2 G of magnetic intensity. The original cell densities of MSCs also had dose-dependent relationship with their growth rate with or without PEMF exposure.

### CONCLUSION

The purpose of this study was to estimate the ability of PEMF stimulation on MSCs proliferation in vitro. It indicated that physical stimuli might be a promising physical stimulus for MSCs expression in the future.

## ACKNOWLEDGEMENT

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## REFERENCE

- [1] Long MW, "Osteogenesis and Bone-Marrow-Derived Cells," *Blood Cells, Molecules, and Diseases*, **27(3)**: 677-691, 2001
- [2] Meirelles LDS and Nardi NB, "Murine Marrow-Derived Mesenchymal Stem Cell: Isolation, in vitro Expansion, and Characterization," *British Journal of Haematology*, 123: 702-711, 2003



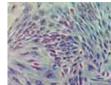


Fig. 1. Von Kossa stain of osteoblasts.

Fig. 2. Oil-red O stain of adipocytes.

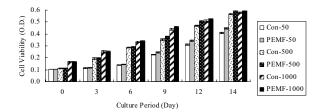


Fig. 3. Proliferation of MSCs by MTT assay (PEMF = 1.3 Gauss).

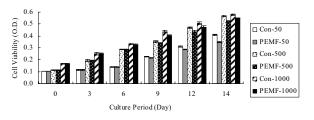


Fig. 4. Proliferation of MSCs by MTT assay (PEMF = 2.4 Gauss).

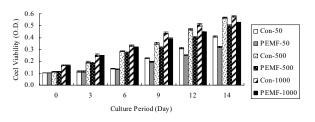


Fig. 5. Proliferation of MSCs by MTT assay (PEMF = 3.2 Gauss).