# In Vivo Response of Osteoblast-like and Odontoblast-like Cells in Intraperitoneal Diffusion Chamber

**Andrea Paola Rodriguez[1)], Hidetsugu Tsujigiwa[1)], Hitoshi Nagatsuka[1)], Kazuo Ichikawa[1)], Atsuhisa Minonishi[2)], Missana Liliana[3)], C. L. Qin[4)], Noriyuki Nagai[1)]**

[1)] *Okayama University, Graduate School of Medicine, Dentistry and Pharmaceutical Sciences*
[2)] *Japan Institute for Advanced Dentistry, Osaka*
[3)] *Tucuman University, Argentina*
[4)] *Texas University, U.S.A*

Abstract: Preliminary analysis of cells before using them in tissue engineering is mandatory. Thus, to clarify the behavior of cells in an appropriate experimental model, we evaluated the characteristic of MDPC-23 cells and KUSA/A1 cells in vitro and in vivo seeded in diffusion chamber. Our results indicated that KUSA/A1 cells differentiated into osteoblast-like cells and induced bone tissue inside the chamber. Whereas, MDPC-23 cells were odontoblast-like cells but with low ability to induce dentin formation. This study suggests that MDPC-23 cells are special cells, which possess morphological and functional character of odontoblast-like cells, expressing DSPP only in vivo with low capacity to induce mineralized dentin matrix.

Keywords: diffusion chamber, MDPC-23 cells, KUSA/A1 cells.

## Introduction

Preliminary analysis of the odontogenic and osteogenic-like cells before using them in tissue engineering is mandatory. We believe that host microenvironment play an important biological role in cell differentiation. Because of this, we evaluated specific protein expression of odontogenic-like (MDPC-23)[1)] and osteogenic-like (KUSA/A1)[2)] cells seeded in diffusion chamber[3)] and implanted in intraperitoneal cavity. Interestingly, our previous gene analysis of MDPC-23 cells has not expressed DSPP in vitro.

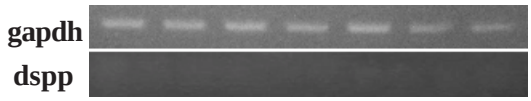
## Materials and Methods

In this study, diffusion chamber (Millipore Corporation, MA, filter type nitrocellulose membrane with the size of 0.22 μm), MDPC-23 (kindly provided by Dr. Hanks, University of Michigan) and KUSA/A1 (kindly provided by Dr. Umezawa, Center of Human Development) cells were used. The odontogenic and osteogenic-like cells were seeded in diffusion chamber and implanted in intraperitoneal cavity of mice. The transplants were stained with routine hematoxylin and eosin, Von Kossa staining, immunohistochemistry and also subjected to transmission electron microscopy after 2, 4 and 6 weeks of implantation.

## Results and Discussion

Interestingly, our previous gene analysis of MDPC-23 cells seeded in a-medium and a-medium with β-glycerophosphate (β-GP) has not expressed dspp (Fig.1). In contrast, when MDPC-23 cells were implanted in intraperitonial diffusion chamber, the cells were capable to express dspp within the membrane of the chamber at 2, 4 and 6 weeks (Fig.4). In contrast, KUSA/A1 cells did not express dspp (Fig.7). Histological analysis of MDPC-23 cells showed excessive cellular proliferation at 2 week, irregular polarized odontoblast-like cells at 4 weeks (Fig.2) and

Fig. 1 Gene expression of MDPC-23 cells in vitro



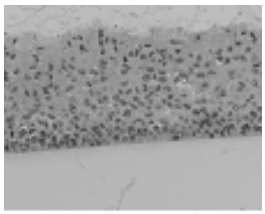



Fig.2 MDPC-23 4 weeks, HE

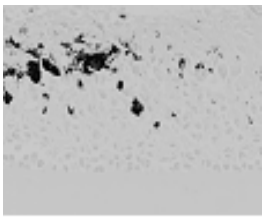


Fig.3 MDPC-23 6 weeks, Von Kossa

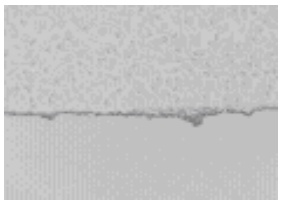


Fig.4 MDPC-23 4 weeks, dspp

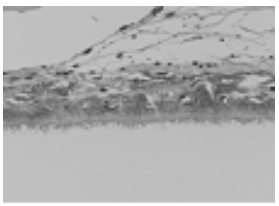


Fig.5 KUSA/A1 4 weeks, HE

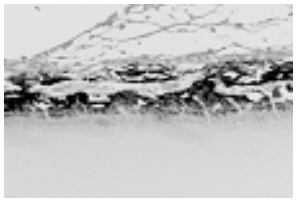


Fig.6 KUSA/A1 4weeks, Von Kossa

Fig.7 KUSA/A1 4weeks, dspp

degenerative cells at 6 weeks. In KUSA/A1 cells at 2 and 4 weeks (Fig.5), revealed new bone and protein secretion inside the porous of the membrane of the chamber and degenerative cells and bone tissues at 6 weeks. Calcification analysis of MDPC-23 demonstrated presence of large areas of dystrophic calcification at 6 weeks (Fig.3). KUSA/A1 cells showed calcium deposition in the bone matrix at 2, 4 (Fig.6) and 6 weeks.This study suggests that the diffusion chamber is a good alternative to analyze gene and protein expression of implanted cells in vivo. It seems that there is an alternate signaling pathway for the expression of DSPP in MDPC-23 cells distinct from the usual epithelial-mesenchymal interaction of the normal tooth development. KUSA/A1 and MDPC23 might be considered as a good candidate for dentin and bone regeneration in basic research.

**References**

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