

## ENAMEL-DENTIN AND DENTIN-CEMENTUM COMPLEX STRUCTURE FORMATION IN TOOTH-TISSUE REGENERATION

M. Honda<sup>1</sup>, Y. Sumita<sup>1</sup>, H. Kagami<sup>1</sup>, I. Asahina<sup>2</sup>, M. Ueda<sup>1</sup>

<sup>1</sup>*Stem Cell Engineering, The Institute of Medical Science, The University of Tokyo*

<sup>2</sup>*Nagasaki University, School of Dentistry*

**INTRODUCTION:** Previous studies have demonstrated tooth-tissue regeneration with porcine tooth cells in the early stage of crown formation seeded onto scaffolds [1, 2]. However, the formation of a tooth with a normal shape was infrequent as most tissue-engineered teeth exhibited a round enamel-dentin complex or a stick-shaped dentin-cementum complex. This study aimed to investigate the mechanism underlying tooth-tissue reconstitution to identify why only enamel-dentin or dentin-cementum structures were formed in the scaffold.

**METHODS:** Tooth buds from 6-month-old pigs were separated into six compartments at the early stage of crown formation: 1) enamel organ without cervical loop region, 2) cervical loop region, 3) dental pulp horn within dental cusp, 4) dental pulp cone in apical side of pulp, 5) dental follicle upper enamel organ, and 6) dental follicle beneath dental papilla. These segment tissues were dissociated into single cells by enzyme digestion. Single-cell populations were recombined with other single-cell populations on a collagen sponge scaffold then implanted into the omentum of immunodeficient rats. Specimens were collected and examined histologically 15 weeks after transplantation. In addition, semi-quantitative RT-PCR was performed to examine the characteristics of cell populations in their segment tissues.

**RESULTS:** Enamel-dentin complex structures were reconstituted by the cervical loop region cells (CLC) in recombination with dental pulp cone cell population (PCC; Figure 1A, Table 1). In contrast, dentin-cementum complex structures were reconstituted by cervical loop region cells in recombination with the dental pulp horn cell population (PHC) within the dental cusp (Figure 1B, Table 1). A complete tooth was not observed using this recombination technique.

In addition, RT-PCR analysis revealed different characteristics between dental pulp horn and dental pulp cone cells. The dental pulp

cone cell population was more immature than the dental pulp horn cell population.

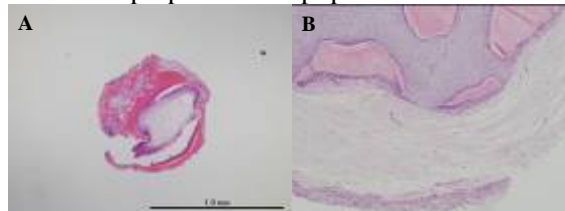


Figure 1: A. Enamel-dentin complex structures. B. Dentin-cementum complex structures.

Table 1:

Relative amounts of complex structures in reconstituted tooth tissue.

	CLC and PHC	and	CLC and PCC
Enamel-dentin	None		High
Dentin-cementum	All		Low

**DISCUSSION & CONCLUSIONS:** These results suggest that cervical loop cells play a key role in the reconstitution of tooth structures. In addition, dental pulp cells have diverse abilities depending on their location in the reconstituting tooth-tissue. We have also identified the mechanism by which enamel-dentin and dentin-cementum tissues form in the scaffold using recombinant methods. However, further investigation is required to identify the mechanism behind complete normal tooth regeneration.

### REFERENCES:

[1] Honda MJ, Sumita Y, Kagami H, Ueda M. Histological and immunohistochemical studies of tissue engineered odontogenesis. Arch Histol Cytol 2005; 68: 89-101.

[2] Honda MJ, Tsuchiya S, Sumita Y, Sagara H, Ueda M. The sequential seeding of epithelial and mesenchymal cells for tissue-engineered tooth regeneration. Biomaterials 2007; 28: 680-9.

### ACKNOWLEDGEMENTS:

This study was supported by Grant-in-aid for Scientific Research from the Japanese Society for the Promotion of Science.