

EFFECTS OF EGCG ON THE DIFFERENTIATION ABILITY OF HUMAN DENTAL PULP CELLS IN VITRO

DH. Lee¹, JW. Park²

¹Dept of Oriental Medicinal Materials & Processing, College of Life Science, Kyung Hee Univ, Gyeonggi-do, ² Dept of Periodontology, College of Dentistry, Kyung Pook National Univ, , Daegu, South Korea

INTRODUCTION: Recent attempts to engineer dental pulp in vitro and in animal models by using modern tissue engineering technologies have shed light on pulp regeneration. Regenerated pulp tissue should be functionally competent, e.g., capable of forming dentin to repair lost structure. (-)-epigallocatechin gallate (EGCG) is one of the major polyphenolic flavonoids present in red wine, which has been shown to have antioxidant properties in different biological systems. Some reports have showed the evidence for a possibly beneficial influence of EGCG in some cases of osteogenesis imperfecta clinically. In the present study, we examined the effect of EGCG on the differentiation of human dental pulp cells. **METHODS:** Human pulp tissue was obtained from three healthy premolars. The teeth were immediately cracked open, and the coronal pulp was removed, minced into small pieces (1 mm × 1 mm × 2 mm) and placed in 6-wells containing α -MEM, supplemented with 10% FBS and antibiotic solution in a humidified atmosphere of 95% air and 5% CO₂ at 37°C. To investigate the influence of EGCG on pulp cells, we assessed cellular viability, alkaline phosphatase (ALP) activity, dentin sialophosphoprotein (dspp)/osteocalcin (OCN) gene expression and nodule formation of dental pulp cells after treatment with EGCG.

RESULTS: EGCG (<15 μ M) did not significantly affect both the viability and proliferation of pulp cells compared with the control over the course of 10 day. EGCG-treatment of pulp cells stimulated the ALP activity in a time-course and dose-dependence. DSPP mRNA expression, which was first detectable after 1 days of all culture conditions before 14 days of EGCG-treatment, was markedly stimulated by EGCG-treatment after 15 days. OCN mRNA expression was also markedly induced by EGCG-treatment after 15 days. To determine the degree of mineralization in pulp cell cultures, cultures were stained with Alizarin Red S. Consistent with the result of

ALP assay, pulp cell cultures treated with EGCG also showed increased numbers of nodules as indicated by the intense Alizarin Red S staining (Fig. 1). Pulp cells treated in the presence of 9 μ M of EGCG for 25 days exhibited a significant increase ($p < 0.001$) in mineralization of the extracellular matrix compared with the control (DM only, Fig. 1).

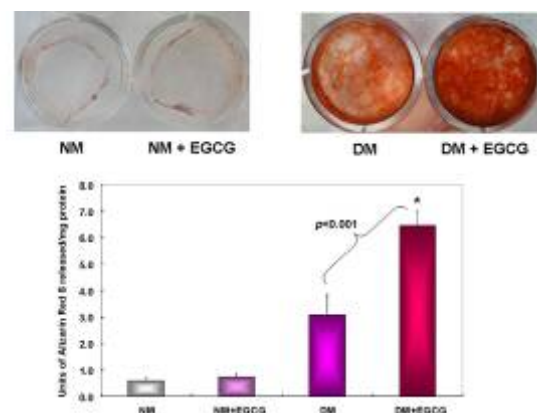


Fig. 1: Extent of matrix mineralization in pulp cell cultures treated with EGCG.

DISCUSSION & CONCLUSIONS: When the cells were treated with EGCG at the concentrations below 10 μ M, pulp cells displayed a significant increase in ALP activity and mineralized bone matrix. Treatment with EGCG also stimulated the expression of dspp & OCN mRNA in dental pulp cell cultures. Our data indicate that the induction of osteogenic/odontogenic differentiation and mineralization in EGCG-treated pulp cell cultures may result in the application for odontogenic agent of pulp tissue engineering/regeneration

REFERENCES: ¹ O. Cortes O, C. Garcia, L. Perez, et al (2006) *Eur Arch Paediatr Dent* 7:96-9. ² B. Vali, L.G. Rao, A. El-Soheymy (2007) *J Nutr Biochem* 18:341-7.