CHAPTER 1

Introduction

1.1 Background and rationale

Regenerative endodontic procedures (REPs) have become alternative approaches for treating immature permanent teeth with pulpal necrosis. Current clinical outcomes exhibiting the signs of healing and radiographic root maturation have been continuously reported (1, 2). However, some histological data have revealed that the tissue formed inside the root canal is not exactly dental pulp (3, 4). Therefore, many efforts are being constantly made to explore the gap of knowledge regarding REPs, in order to improve the ultimate treatment outcomes.

Stem cells from apical papilla (SCAPs) have been raised as one of the key factors that may participate in the regenerative process after REPs. The potential of pulpal reparation depends on 1) the potential of multi-lineage differentiation, 2) the capability to survive during infection, and 3) the possibility of these cells to be recruited into the root canal system (5-7). Thus, promoting initial attachment of cells inside the root should offer a superior situation for tissue generation.

Several studies have focused on dentin pre-conditioning techniques that can be used during routine treatment protocols (8, 9). Several studies have reported that when EDTA is used as a final irrigant, cell attachment to the dentin surface increases (10, 11). Moreover, calcium hydroxide (CH) medication improves the ability of APCs to attach to the medicated root dentin (12). Interestingly, Galler and colleagues have demonstrated that the transforming growth factor beta-1 (TGF-β1) is expressed on the dentin surface after CH and EDTA treatment (13). Addition of 0.12% CHX in dentin conditioning for

five minutes promotes TGF-β1 release more than EDTA alone. Thus, to develop the dentin pre-conditioning step for clinical use, a variety of solutions and protocols must be evaluated.

To date, a variety of dynamic irrigation protocols are available including needle irrigation (NI), EndoActivator (EA), and Passive Ultrasonic Irrigation (PUI). NI provides activation by manual work in a short up-and-down motion. EndoActivator (Dentsply Tulsa Dental Specialties, Tulsa, OK, USA), a sonically activated irrigation device, offers forceful streaming at a specific frequency (190 Hz). PUI possesses 30 kHz of oscillation frequency providing acoustic streaming and cavitation surrounding the activated file (14-16). This dynamic movement causes pressure and shear stress on the dentin surface which results in well-debrided root canal walls (17-19). Moreover, the release of growth factors from dentin has been observed after the root canal is irrigated with EDTA using PUI (20). Therefore, the use of these dynamic irrigation techniques might improve the dentin pre-conditioning steps in REPs. However, in respect of regenerative endodontics, these techniques would be applied to immature teeth with wide root canals and open apices. Then, the most appropriate, effective techniques that cause the least damage should be elucidated.

This study aims to evaluate the effects of various dynamic irrigation techniques: NI, EA, and PUI, incorporating a variety of irrigating solutions, on APC attachment in an *ex-vivo*, immature tooth model.

1.2 Research questions

- Does dynamic irrigation affect APC attachment to human root canal dentin in an *ex-vivo* immature tooth model?

- Does dynamic irrigation affect the morphological features of human root canal dentin in an *ex-vivo* immature tooth model?

1.3 Objectives of the study

- To determine the effects of dynamic irrigation on APC attachment to human root canal dentin in an *ex-vivo* immature tooth model, by means of counting the number of fibronectin-positive cells per microscopic field.

- To determine the effects of dynamic irrigation on the morphological features of human root canal dentin in an *ex-vivo* immature tooth model, by means of scanning electron microscopic observation.

1.4 Research hypotheses

- Dynamic irrigation increases APC attachment to human root canal dentin in an *ex-vivo* immature tooth model, measured by fibronectin positive cell count.

- Dynamic irrigation improves the surface cleanliness of human root canal dentin in an *ex-vivo* immature tooth model, measured by scanning electron microscopy.