Chapter I

General introduction
**Optimizing the chemical aspect of root canal irrigation**

**General Introduction**

Endodontology is concerned with the study of the form, function and health of the dentinal-pulp complex and periradicular region, in addition to injuries to and diseases of it. It also aids the prevention and treatment of apical periodontitis caused by root canal infection (Loest 2006). The aim of a root canal treatment is to completely remove inflamed and infected tissue present in the root canal system and to efficiently prevent the entrance of new microorganisms or nutrients in order to maintain or create a healthy environment around the root (Haapasalo et al. 2011). Recent evidence shows that endodontic therapy may not always result in complete healing of apical periodontitis (Liang et al. 2013). These results highlight the importance of optimization of root canal cleaning procedures to face the following challenges:

1. The root canal anatomy is complex and highly variable. The root canals are often long, multi-curved and oval shaped (Wu et al. 2000). The root structure consists of dentin which is a porous material containing tubules with a typical diameter of 0.6-3.2 µm and length of 1-2 mm and which are accessible for microorganisms (Haapasalo et al. 2011). Accessory root canal morphology such as lateral canals, fins and isthmi are unreachable by instrumentation (Peters et al. 2001) and difficult to clean with syringe-based irrigation (Verhaagen et al. 2013) consequently, persistent infection in those regions may lead to endodontic failure (Wu & Wesselink 2005; Ricucci et al. 2013). *In this thesis we propose the use of root canal models of various complexities to study the occurrence of transient cavitation and streaming in sonic and ultrasonic activated irrigation (Chapter IV).*

2. In infected root canals microorganisms grow attached to the wall as biofilm (Ricucci et al. 2009), which is to a large extent resistant to mechanical and chemical attacks imposed by cleaning procedures and disinfectants. It is reported in the literature that up to 90% of the biofilm structure is composed by an extracellular polymeric matrix (EPS) which gives the biofilm its viscoelastic properties and capacity to deform and adapt under mechanical stresses (Körstgens et al. 2001). *In this thesis we introduce a novel hydrogel model that mimics the viscoelastic properties of a biofilm and allows for the visualization and quantification of the cleaning ability of several cleaning methods and irrigants in artificial accessory root canal morphology (Chapter V).*

3. Mechanical instrumentation can remove the main bulk of infected tissue and its nutritional supply, facilitate delivery/refreshment of irrigants through out the root canal system and allow proper placement of a root canal filling (Hülsmann et al. 2005). Its contribution for the cleaning of the root canal system is questionable (Attin et al. 2002) as it cannot adequately reduce viable microorganisms in the infected root canal system (Grahnen & Krasse 1963; Byström & Sundqvist 1981), nor can it prevent the formation of smear layer or dentin debris (Robinson et al. 2012). Furthermore, even the most advanced instrumentation systems failed to
prepare the majority of the main root canal (Metzger et al. 2010) and may con-
tribute negatively to the outcome of the endodontic treatment by instrumenta-
tion failures, weakening of the root structure and apical crack formation (Gorni
& Gagliani 2004; Wu et al. 2004; Shemesh et al. 2010). The optimization of the
chemical aspect of disinfection aims to contribute for a future more efficient and
safer non-instrumentation technique however it is still necessary to create space in
the root canal system with instruments to be able to apply disinfection solutions or
medicaments.

4. Smear layer, produced during root canal instrumentation procedures, is strongly
attached to the root canal wall and penetrates deep into the dentinal tubules and
accessory root canal morphology. Both smear layer and dentin debris will inacti-
vate root canal antimicrobials and irrigants and hinder their access to the biofilm
(Haapasalo et al. 2007). Recently, it was shown that the production of dentin de-
bris and the subsequent blockage of isthmi may be a larger problem than antici-
pated (Paqué et al. 2011). Its removal is challenging and plays a crucial role in the
disinfection process. This debate is addressed in multiple studies in this thesis where
we investigate the effect of dentin in the reaction rate of different NaOCl solutions,
including its buffering capacity (Chapters II and III).

Root Canal Irrigation

Root canal irrigation can be defined as the procedure to deliver a liquid or irrigant in the
root canal system before, during and after instrumentation of the root canal. The aims of
this procedure are the mechanical detachment of pulp tissue; dentin debris and smear lay-
er (instrumentation products); micro-organisms (planktonic or biofilm) and their products
(all together hereafter named substrate) from the root canal wall, their removal out of the
root canal system and their chemical dissolution or disruption. For that, root canal irrigation
should induce a flow of irrigant to the full extent of the root canal system, in order to come
in close contact with the substrate, carry away the substrate and provide lubrication for the
instruments. Such flow should ensure an adequate delivery throughout the root canal sys-
tem, refreshment and mixing of the irrigant, in order to retain an effective concentration of
the active chemical component(s) and compensate for its eventual inactivation. Its delivery
should be targeted towards the root canal wall, in order to detach/disrupt the substrate by wall
shear stress, meanwhile it should remain restricted to the constraints of the root canal, thus
preventing irrigant extrusion towards the periapical tissues.

Irrigant of choice: sodium hypochlorite.

Sodium hypochlorite (NaOCl) is widely used as the primary root canal irrigant (Dutner et
al. 2012) due to its unparalleled action against microorganisms (McDonnell & Russell 1999)
and biofilm (Arias-Moliz et al. 2009; Bryce et al. 2009) and its unique capacity to dissolve pulp
tissue (Sirtes et al. 2005) and organic components of the smear layer (Baumgartner & Mader
1987). Once delivered inside the root canal, NaOCl reacts with its organic content such as
pulp, biofilm or dentin (canal wall, smear layer or debris) causing depletion of the free available
chlorine (Baker 1947), resulting in protein degradation, rise of temperature (Baker 1947) and changes in pH (Jungbluth et al. 2011).

In spite of its low clinical toxicity (Zehnder 2006), NaOCl has been shown to be extremely caustic when in contact with organic tissue in vitro (Pashley et al. 1985), even at concentrations lower than 0.1% (Chang et al. 2001; Heling et al. 2001; Barnhart et al. 2005). The choice of the NaOCl concentration to be used is generally considered a trade-off between cleaning efficiency and tissue damage in the case of inadvertent extrusion (Spencer et al. 2007).

Different strategies to optimize the efficiency of NaOCl solutions will be evaluated in this thesis:

1. Concentration
   The amount of free chlorine available per volume of solution defines the concentration of NaOCl solutions (%). Clinically, the use of concentrations between 0.5% and 6% are suggested, however, the optimal clinical concentration is still a subject of controversy (Zehnder 2006). The cleaning efficiency of NaOCl (antimicrobial capacity and tissue dissolution effect) increases with its concentration (Moorer & Wesselink 1982; Arias-Moliz et al. 2009; Retamozo et al. 2010). However, higher concentrations help to dissolve more tooth structure (Sim et al. 2001); or could be more harmful when inadvertently extruded into the periapical tissues (Boutsioukis et al. 2013). In this thesis the impact of concentration on the reaction rate of NaOCl (Chapter II) and its capacity to compensate for the buffer capacity of dentin have been investigated (Chapter III).

2. Refreshment
   Refreshment has been suggested to be an effective method to compensate the loss of chemical efficiency of a lower concentration of NaOCl solutions (Moorer & Wesselink 1982; Zehnder 2006), but this has never been proven. Refreshment may also help sustaining the irrigant concentration at the interface with root canal wall, enhancing diffusion of NaOCl into tubules and lateral canals (Verhaagen et al. 2013). The Intermittent Flush Method (IntFM) (Cameron 1988) combines refreshment with ultrasonic activated irrigation (UAI), in a multiple refreshment/activation cycle protocol. In this thesis we evaluate the influence of IntFM on the reaction rate of NaOCl (Chapter II) and on the removal of dentin debris from an artificially created groove in a root canal wall (Chapter V).

3. pH
   The pH of the NaOCl solution determines the equilibrium (Figure 1) of the free available chlorine, the hypochlorite ion (OCl⁻) and the hypochlorous acid (HOCl) (Baker 1947). The biological effect of NaOCl, which can be defined as its tissue-dissolving capacity and antimicrobial effect, will be influenced by this equilibrium. In alkaline solutions (pH > 7.5), OCl⁻ prevails, which has a powerful oxidative effect and therefore a higher tissue dissolving capacity than HOCl (Baker 1947). On the other hand, HOCl prevails in acidic solutions (3 < pH < 7.5). It has a powerful bactericidal effect probably because it is a smaller uncharged molecule, which can relatively easily penetrate the bacterial membrane. After penetration, it can result in protein degradation (Winter et al. 2008). In this thesis we evaluate the buffer capacity of dentin in NaOCl solutions with different pH. We also characterize the short-term stability of the NaOCl at low pH and its reaction rate with dentin (Chapter III).
4. Temperature
Temperature is the only clinical strategy that has proven to be effective in compensating a reduction in concentration. (Sirtes et al. 2005) showed that a 1% NaOCl solution heated to 45°C equals the tissue dissolution capacity of a 5.25% NaOCl solution at 20°C; at 60°C the tissue dissolution capacity is even higher. However no evidence existed in the ability to keep the temperature high inside the root canal for the entire cleaning process. In this thesis we monitor the temperature evolution of pre-heated irrigant inside a root canal and introduce and validate a numerical model to study the thermodynamics of irrigation procedure (Chapter III).

5. Surface of contact
Sodium hypochlorite acts by direct contact between free available chlorine molecules and organic matter (Moorer & Wesselink 1982). Increasing the movement of molecules in the root canal system increases the contact of active chlorine molecules and organic matter and therefore the tissue dissolution capacity/ antimicrobial effect of the irrigant. The study of the chemical and mechanical processes, interactions or impediments occurring at this interface is fundamental to understand the cleaning process. In this thesis we look at the interface NaOCl with a novel hydrogel, confined in accessory lateral canals and isthmi, using high-speed imaging (Chapter V). We also highlight the importance of respecting a clinically representative ratio volume of irrigant to the subtract surface to be dissolved in invitro chemical assays and introduce a new dentin model (Chapter III).

6. Activation
Irrigants are often delivered in root canals by a syringe with a needle. Once in the canal, they can be activated by different means such as sonic, ultrasonic or laser agitation/activation, in order to refresh consumed irrigant at the reaction site, to enhance wall shear stresses and to displace irrigant into the accessory root canal anatomy. The last two methods are the most efficient methods to mechanically clean confined areas of root canal systems (de Groot et al. 2009; Jiang et al. 2010). Ultrasonic Activated Irrigation (UAI) increases tissue dissolution (Moorer & Wesselink 1982), especially when NaOCl solutions are used (van der Sluis et al. 2010). Microstreaming and hydrodynamic cavitation (the formation and implosion of vapor bubbles) have been suggested as their working mechanisms (de Groot et al. 2009; Ruddle 2009; Jiang et al. 2010). However, the occurrence of cavitation inside the root canal has been questioned due to space restrictions. Various forms of irrigant activation are addressed in this thesis. We investigate the influence of Laser and Ultrasonic Activated Irrigation in the reaction rate of NaOCl (Chapter II); detect, quantify and visualize the occurrence of cavitation in sonic and ultrasonic activated irrigation for clinically realistic settings (Chapters IV); and investigate various irrigant solutions in their capacity to remove dentin debris from the root canal during UAI (Chapter V).

7. Volume and time
Irrigation time and volume are key clinical aspects for which guiding evidence is scarce (Zehnder 2006). Currently, clinical protocols on irrigation (Liang et al. 2013) are governed by reported evidence on the mechanical aspect of irrigation (van der Sluis et al. 2010). To take into account the chemical aspect, further research into the optimal irrigation time and volume at given concentration, pH and temperature is required. However, the highly variable clinical situation in terms of pathology, morphology, microbiology and the quantity and
quality of tissue to be dissolved make its study a great challenge. With this in mind, the results presented in this thesis contribute to a better understanding of the optimum irrigant delivery time at given temperature and the cooling rate of irrigant in the syringe which should guide the pre-heated syringe turn-over (Chapter III).

Objectives of the thesis

The aim of this thesis was to suggest new models and methodologies to measure, image and compare the cleaning efficiency of various irrigation methods and solutions and to optimize the chemical aspect of root canal irrigation.

Outline of the thesis

In chapter II the optimization of the chemical efficiency of NaOCl in contact with dentin is under investigation. In a bovine tooth model, the effect of multiple factors such as concentration, temperature, pH, laser and ultrasonic activated irrigation, irrigant refreshment and exposure time in the reaction rate of NaOCl with dentine is analyzed. The characterization of the chemical reaction, ultrasonic power and the dentin model in comparison with a human size tooth, are discussed.

In chapter III new insights on the evolution of temperature and pH of irrigants during root canal therapy are presented. Clinically representative models are proposed and characterized. A numerical model simulating the thermodynamics of root canal irrigation was introduced and validated for endodontic research.

In chapter IV, the occurrence of transient cavitation (growing and implosion of bubbles) around sonic or ultrasonic activated endodontic instruments is investigated. The effect of power, confinement, tip design, taper and size in the occurrence of cavitation is analyzed. Combining sensitive analytical sonochemical techniques with powerful imaging tools, this chapter gives new detailed insights into the cleaning mechanisms of irrigant activation techniques.

In chapter V, the cleaning efficiency and evolution of different irrigation methods and solutions in confined accessory root canal anatomy, is assessed. A novel hydrogel model that mimics biofilm viscoelastic properties is introduced in endodontic research.
References


