Effects of Temperature and Hypochlorite Concentration on the Rate of Collagen Dissolution

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Abstract

Introduction: The aim of this research was to quantify the effects of temperature on the rate of collagen dissolution in different concentrations of sodium hypochlorite solution. **Methods:** A customized laboratory apparatus was used to immerse 23 samples of reticulated collagen matrices and 20 samples of non-reticulated collagen matrices in a continuously agitated solution of 1%–5% (m/v) sodium hypochlorite at 20°C–35°C. The time needed for complete dissolution of each sample was used to generate mathematical equations that described the effects of temperature and hypochlorite concentration on the dissolution of collagen. **Results:** Both temperature and concentration of sodium hypochlorite were positively correlated with the rate of collagen dissolution. The same speed of dissolution was obtained for 5% sodium hypochlorite at 20°C, 4% solution at 20.8°C, 3% solution at 23.5°C, 2% solution at 26.9°C, and 1% solution at 36°C. **Conclusions:** Treatment protocols that involve sodium hypochlorite, which is the preferred root canal irrigant, should consider the important effects caused by variations in the use of different irrigant temperatures and concentrations. Further investigation is required to establish the optimal concentration and temperature of sodium hypochlorite solution for root canal irrigation. *(J Endod 2015;41:903–906)*

Key Words

Collagen, root canal irrigant, sodium hypochlorite, speed of dissolution, tissue dissolution

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The aim of root canal treatment is to obtain germ-free and compact-filled canals to prevent diseases such as apical periodontitis (1). To reach this goal, the soft organic tissue content of the canal must be removed. This is achieved by irrigation of the parts of the canal that cannot be reached by mechanical means. The tissue-dissolving ability of an irrigant is important because it enables the removal of pulp and pulp remnants from the root canal (2).

Sodium hypochlorite solution has long been advocated as the preferred irrigant for root canal irrigation owing to its tissue-dissolving and antimicrobial activities. To date, there has been no suggestion in the literature about which temperature and concentration of sodium hypochlorite solution are optimal for root canal irrigation. Many authors have demonstrated that sodium hypochlorite dissolves pulp tissue (3). Others have shown that increasing the concentration or temperature of the solution appears to enhance the solvent effect on pulp tissue (4–6). The pH of the solution does not influence the tissue-dissolving ability of the sodium hypochlorite solution (7). Immersing organic tissue samples in pots filled with sodium hypochlorite solution and a large specimen contact surface increase the ability of sodium hypochlorite to dissolve organic tissue (3, 8).

Collagen is abundant in the dental pulp and in the vicinity of the canal wall (9). Because collagen is one of the most compact proteins found within soft tissues (10), its use in experimental settings provides a worst-case scenario on which to test various parameters. Accordingly, collagen has been used as a substitute for dental pulp in studies to investigate the dissolution effect of sodium hypochlorite (5, 11, 12). Dissolution of collagen samples appears to occur in 2 major stages as follows:

1. The surface stage, which involves fragmentation of collagen in fibrils, followed by their liberation into the liquid phase
2. The liquid phase, which involves hydrolysis of the fibrils into amino acids and their neutralization by conversion to salts and chloramines (13)

All of the studies that have investigated the variables that affect the dissolution of collagen by sodium hypochlorite have investigated the effects of each factor in isolation. Few of the aforementioned studies have used agitated sodium hypochlorite solution to represent the solution flow in the root canal (8). The relationship between temperature and sodium hypochlorite concentration that affects the rate of pulp dissolution has yet to be established. The aim of the present study was to obtain a mathematical equation that described the dependence of the rate of tissue dissolution on the temperature and concentration of agitated sodium hypochlorite solution. Knowledge of the correlation between these factors will enable us to choose the optimal correlation between temperature and concentration of sodium hypochlorite solution that will maximize the rate of collagen dissolution.

Materials and Methods

Reticulated collagen matrix (RCG) and non-reticulated collagen matrix (NRCG) samples were obtained from Bucharest Leather Research Institute (Bucharest, Romania). The RCG samples had 25 ± 3 μm fiber diameter, and the NRCG samples had 23 ± 4 μm fiber diameter. The porosity was 0.87 m^3/1 m^3 for RCG and 0.88 m^3/1 m^3 for NRCG. The RCG samples contained 3% water and 1% impurities, whereas NRCG samples had 4% water and 3% impurities. All specimens were 3 mm thick.

Sodium hypochlorite aqueous solutions at 1%–5% concentrations were prepared from 10% sodium hypochlorite solution (Merck KGaA, Darmstadt, Germany). All other

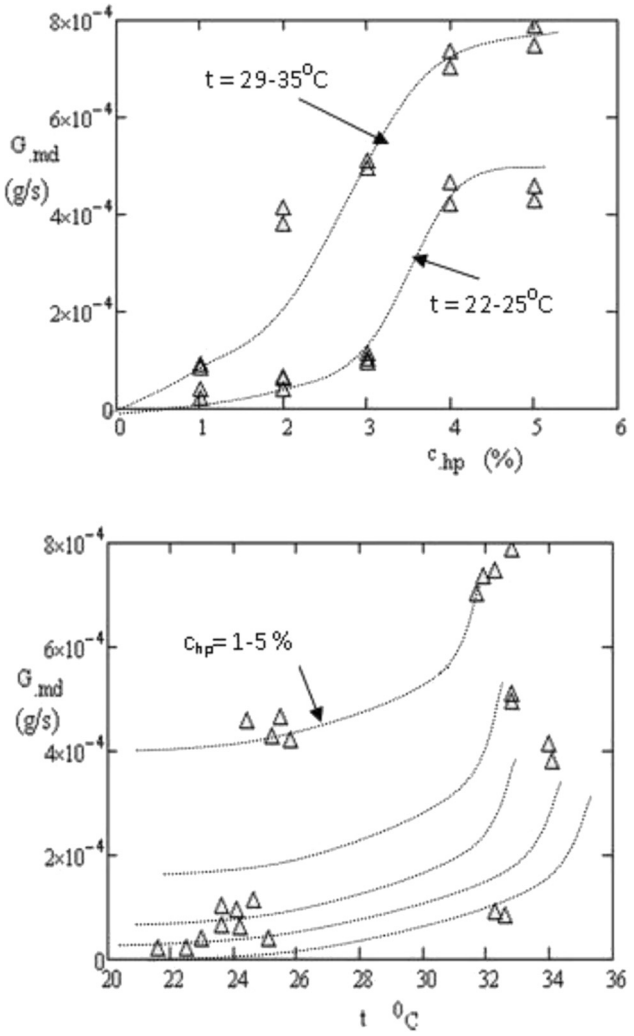

Figure 1. Evolution of RCG dissolving rate (G_{md}) with hypochlorite concentration (c_{hp}) and temperature (t).

reagents used for iodometric concentration determination were of analytical quality. Sodium hypochlorite solution was agitated and heated by using a Falc Heidolf magnetic agitator (model AG2; Nitech, Bucharest, Romania) that had a built-in thermostat. The pH and chloride concentration were determined by using a Jenway model 3310 pH meter with incorporated ionometer (Fisher Scientific, Loughborough, UK), which was equipped with specific electrodes (FB61546 for pH and PH-905-014P for chloride ion concentration) and a temperature probe.

All the samples were weighed precisely by using a Kern & Sohn analytical balance (model ACS/ACJ; Balingen, Germany).

Twenty-three samples of RCG were divided into 5 groups, 3 containing 5 samples and 2 containing 4 samples. Whereas 5 samples were used for each of the 1% (10 g/L), 2%, and 3% sodium hypochlorite treatments, 4 samples were used for each of the 4% and 5% sodium hypochlorite treatments.

The 20 samples of NRCG were divided into 5 groups, each of which contained 4 samples dissolved into concentrations of 1%, 2%, 3%, 4%, or 5% sodium hypochlorite.

For all of the dissolved samples, the temperature of the sodium hypochlorite solution varied from 21.9°C to 34.1°C. The pH of the solution varied with a range of 12.9–13.9. The solution was heated. The temperature and concentration of the sodium hypochlorite solution and the mass of the samples were recorded before dissolving each sample.

The method used to dissolve each sample was as follows:

1. The collagen matrix samples were cut to dimensions of 5 mm long, 3 mm wide, and 2 mm thick.
2. Samples were weighed and the mass was recorded.
3. 250 mL sodium hypochlorite solution was prepared at the desired dilution, placed in a glass beaker on the Falc Heidolf magnetic heater agitator, and the pH ionometer temperature probe was placed in the solution.
4. The temperature was set, and the solution was agitated to the speed of 1 m/s.
5. The samples were placed on a glass probe, immersed in solution, and maintained in position.
6. The chronometer was started and watched until the sample completely disappeared.
7. The time, pH of the solution, and voltage of the ionic chloride probe were recorded. After the dissolution of each group of samples, the concentration of the solution was tested by iodometric titration. The test results were recorded in tables and represented in Figure 2 and Figure 3.

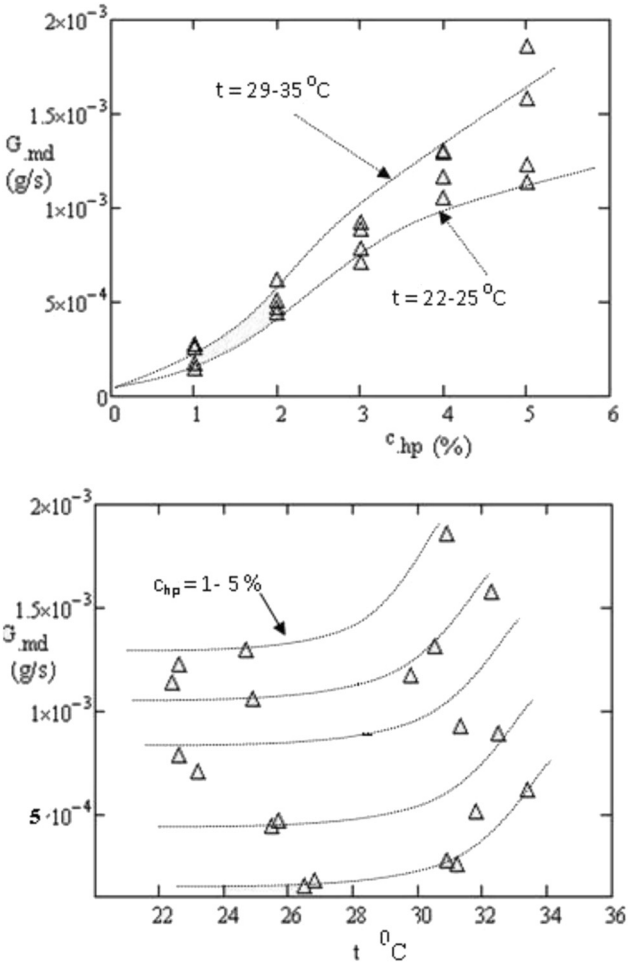

Figure 2. Evolution of NRCG dissolving rate (G_{md}) with hypochlorite concentration (c_{hp}) and temperature (t).