**INTRODUCTION**

During preparation of the root canal, dentine debris, remaining pulp tissue, microorganisms, and irrigation solutions can be extruded beyond the apical foramen, coming into contact with the periapical tissues (1, 2). This debris can result in inflammation of the apical region, causing postoperative pain, acute inflammatory response, or even the exacerbation of chronic inflammatory processes, known as “flare-ups,” which occur in 1.5%–16% of cases. Moreover, such extrusion may cause a delay in periapical repair (3-6).

Apically extruded debris varies in quantity and may differ according to several factors related to both the tooth and the technique used. These factors include the type of canal, the diameter of the foramen, the type of technique, the apical preparation limit, the irrigation technique, and the irrigation solution used. However, there is still no ideal technique that prevents the extrusion of debris (7, 8).
Sodium hypochlorite (NaOCl), often used at concentrations ranging between 0.5% and 6%, is the most popular irrigation fluid used in endodontics. In addition to its proven bactericidal action, it exhibits excellent dissolution activity of organic matter (7). Because its improved biocompatibility, in addition to its antimicrobial properties, chlorhexidine (CHX) has been recommended as an adjunct irrigant. It can be used in solution or gel form and at concentrations ranging from 0.2% to 2%. The gel form of CHX has different physical properties from the solution due to its viscosity and ability to eliminate much of the smear layer (9).

It is important to emphasize that despite the excellent antibacterial action of NaOCl and CHX, anatomical variations and remnants of obturator materials, in cases of retreatment, interfere with disinfection of the root canal system because they limit the access of the irrigation solution to the remaining pulp tissues (10).

During endodontic treatment, irrigants, such as NaOCl and CHX, may extrude to the periapical tissue. In vitro methods used to collect apically extruded debris frequently use distilled water in the preparation of specimens to avoid any possible crystalization of NaOCl that could alter dentine debris (11). However, the effect of NaOCl crystals on debris weight is unclear; furthermore, NaOCl may play an important role in the dissolution of organic dentine tissue (12).

The aim of the present study was to evaluate the influence of different irrigation chemical solutions on the amount of extruded residues apically, varying the instrumentation technique in manual, continuous rotation, or reciprocation motions. The null hypothesis was that different irrigation solutions do not influence the amount of apical extruded detritus, independently of the instrumentation technique used.

MATERIALS AND METHODS

Specimen preparation
The present study was approved by the local ethics committee (no. 129.690). A total of 90 recently extracted single rooted mandibular premolars with single canals were investigated. All teeth were clinically inspected and radiographed in the buccolingual and mesiodistal directions. All teeth with single canals, mature root apices, with no root fractures, no root resorption, and tissue (10).

Preparation of root canals
Pre-enlargement of the cervical and middle third of all teeth selected for the study was performed using the Gates–Gladen drill was removed, cleaned with the aid of a brush in running water, dried in an oven at 170 °C for 10 min, and inspected before being reused. The patency of the canals was then confirmed using a #10 K-file (Dentsply Maillefer). Instrumentation was performed by a single endodontic specialist.

Manual instrumentation (MI): Preparation of the canals was performed with Flexofile instruments (Dentsply Maillefer), using the crown-down technique, with 1/4 turn kinematics. The preparation was initiated with file size #50 in the coronal aspect with file sizes reducing progressively in the apical direction, ending finally with file size #30.

ProTaper Universal (PTU): Preparation was initiated with S1 and S2 PTU instruments (Dentsply Maillefer) progressively in the apical direction and with slight pressure until resistance was encountered and finalized with F1, F2, and F3 instruments, up to the working length. The X-Smart Plus motor (Dentsply Maillefer) was used with the PTU function in each instrument. The file was then withdrawn, cleaned, and inspected before being reused.

WaveOne Gold (WO): Preparation was performed using the WO primary single-file reciprocation system (25/08) (Dentsply Maille-
coupled with the X-Smart Plus motor. The function WO ALL was used to perform pecking movements until the working length was reached. After the pecking motions, the file was withdrawn, then cleaned, and inspected before being reused.

Irrigation parameters

In all groups, a 5 mL syringe and a 24G three-quarter inch, 0.55 mm×20 mm needle were used, which was introduced in the middle third of the roots 5 mm short of the working length. A constant apex–crown movement (1–2 mm) was maintained during irrigation. Aspiration was performed using a disposable cannula positioned at the entrance of the canal.

NaOCl and distilled water: Teeth in the MI and PTU groups were irrigated with 1 mL of the irrigation solution every time the instruments were changed. Irrigation with 1 mL of the irrigation solution was performed after three pecking movements in the WO group. At the end of the preparation, each group used 5 mL of the irrigation solution.

CHX gel: Teeth in the MI and PTU groups were irrigated with 0.25 mL of 2% CHX gel deposited in the pulp chamber every time the instruments were changed. The gel was then washed with 0.75 mL of distilled water. The same procedure was performed on teeth in the WO group; however, a change of instruments was performed after every three pecking movements. At the end of the preparation, a total of 5 mL of the irrigation solution were used in each group.

Debris collection

After instrumentation, the rubber stopper with the tooth and the needle was removed, and the debris adhering to the outer surface of the root was collected by washing with 1 mL of distilled water to deposit in the glass vial. The vials were placed in an oven at 140 °C for 5 h so that the mixture would be totally evaporated water before weighing the dry debris (14). The debris was weighed using an analytical balance and recorded as the average of three consecutive measurements, subtracting the mass of the empty vials, which was previously determined.

Statistical analysis

All statistical tests were performed using SPSS version 23 (IBM, Armonk, NY, USA). In the first test, data were summarized in tabular form and analyzed using the Kolmogorov–Smirnov, two-way ANOVA, Levene variance homogeneity, and Games–Howell multiparameter comparison tests for heterogeneous variances. In the second test, the one-way ANOVA and Tukey tests were used to compare the mass of the residues of the irrigation solution alone. The level of significance was set at 0.05 (P=0.05).

RESULTS

Apical extrusion of debris occurred in all groups. The mean and standard deviation of the values according to each group are shown in Table 1. There were significant differences in apically extruded debris among the irrigation solutions and techniques used (P<0.05). Of the irrigation solutions used, 2% CHX resulted in the most debris formation when root canal preparation was performed using PTU (P<0.05). The groups associated with distilled water (P<0.05) combined with the reciprocal technique had the least extruded debris (P<0.05), regardless of the technique used (Table 2). Moreover, the results shown in Table 2 indicate that there was a statistical difference regarding the formation of crystals of the irrigation solutions used (P<0.05). Therefore, the crystals formed only by the evaporation of the irrigation solutions differed when 2.5% NaOCl, 2% CHX, and distilled water were used. The 2.5% NaOCl solution produced the greatest amount of residues compared with other irrigation solutions (P<0.05), whereas distilled water resulted in the lowest amount of residue (P<0.05).

DISCUSSION

All instrumentation methods, whether manual, continuous rotation or reciprocation motions led to the extrusion of apical debris, regardless of the irrigation solution used, thus corroborating the results obtained in previous studies (15, 16). The null hypothesis was rejected on the assumption that the irrigation solutions used had no influence on the amount of debris extruded.

The irrigation solutions were weighed to verify the amount of residue that would remain after drying the mounts. This second test was included because distilled water is usually recommended in apically extruded debris methodology (11, 13, 16); however, water has not been proposed during biomechanical preparation because it is not able to promote tissue dissolution or kill microorganisms. In this in vitro study, the amount of NaOCl crystals was higher than those of CHX and distilled water (P<0.05). On the other hand, the effects of NaOCl, as used in

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### TABLE 1. Mean mass (g) of apical debris generated according to instrumentation technique and irrigation solution

| Instrumentation/irrigation | n | Mean | SD |
|----------------------------|---|------|----|
| PTU/CHX                    | 10| 0.211<sup>a</sup> | 0.045 |
| PTU/NaOCl                  | 10| 0.105<sup>b</sup> | 0.057 |
| MI/CHX                     | 10| 0.120<sup>b</sup> | 0.064 |
| MI/NaOCl                   | 10| 0.103<sup>b</sup> | 0.038 |
| WO/CHX                     | 10| 0.080<sup>b</sup> | 0.056 |
| WO/NaOCl                   | 10| 0.012<sup>c</sup> | 0.011 |
| PTU/distilled water        | 10| 0.001<sup>c</sup> | 0.001 |
| MI/distilled water         | 10| 0.000<sup>c</sup> | 0.000 |
| WO/distilled water         | 10| 0.000<sup>c</sup> | 0.000 |

Superscript letters represent statistically significant differences between values.

### TABLE 2. Mean mass (g) of debris according to instrumentation technique, regardless of the irrigation solution, and in relation to the irrigation solution, regardless of the instrumentation technique

| Irrigation        | n | Mean | SD |
|-------------------|---|------|----|
| PTU               | 30| 0.105<sup>a</sup> | 0.096 |
| MI                | 30| 0.074<sup>b</sup> | 0.067 |
| WO                | 30| 0.031<sup>c</sup> | 0.047 |
| CHX               | 30| 0.137<sup>a</sup> | 0.077 |
| NaOCl             | 30| 0.073<sup>d</sup> | 0.058 |
| Distilled Water   | 30| 0.000<sup>e</sup> | 0.000 |
| Crystals of the irrigation solutions (g) | | | |
| NaOCl             | 10| 0.106<sup>f</sup> | 0.001 |
| CHX               | 10| 0.034<sup>g</sup> | 0.000 |
| Distilled Water   | 10| 0.001<sup>h</sup> | 0.000 |

Values with different superscript letters for instrumentation, irrigation, and residues of the irrigation solution are statistically different.
endodontics, on dentine are different because of its small size. The hypochlorite anion is capable of infiltrating mineralized collagen and destroying collagen fibrils (12), and this reaction occurs during the use of endodontic instrumentation and irrigation (17).

After instrumentation and irrigation, some bacteria may remain in the canal space. Colonization of Enterococcus faecalis, which is closely associated with failure of endodontic treatment, may produce more biofilm mass and protein in response to high pH conditions or antimicrobial sub-inhibitors, thus increasing the amount of debris (18). The risk for extrusion of debris could be decreased when the activation of irrigations is performed and produces a synergistic effect in the debridement of smear layer accumulated in non-instrumented areas (10).

In the present study, the technique that resulted in the most debris extrusion was the PTU system, as verified in other previous studies (4, 15, 19-22). This can be explained by the higher number of instruments that produce the largest quantity of residues (19), regardless of the irrigation solution used.

Different outcomes have been reported in some studies using an apical debris collection methodology. These outcomes demonstrated no significant differences between the amount of debris extruded between continuous rotation (ProTaper Next system, Dentsply Tulsa Dental, Tulsa, OK, EUA) and reciprocation file systems (WaveOne system, Dentsply Tulsa Dental, Tulsa, OK, EUA) (11). In contrast, another study showed that reciprocation system (Reciproc system, VDW, Munich, Germany) leads to more extrusion of debris than the rotational system (ProTaper Universal system, Dentsply Maillefer, Ballaigues, Switzerland) (23). This difference may be related to the applied method, mode of debris collection, type of tooth, and/or instruments used in the tests (7).

Gel-based CHX 2% solution exhibits a rheological action, which maintains debris in suspension (9). In the present study, the amount of debris was significantly more when it was associated with 2% CHX gel. This high apical extrusion can be attributed to the thixotropic property of the gel (24). An increase in fluidity occurs as it is agitated. This physical property of the gel may also explain the increase in the quantity of debris generated when it was associated with the rotational system compared with the reciprocal system. Furthermore, the 2% CHX gel exhibited a different behaviour because it is unable to dissolve organic matter, which remains in suspension and is eventually pushed to the periapical area (9, 25).

The 2.5% NaOCl solution generated a larger amount of residue when only the irrigation solution was weighed. When the instrumentation technique was associated with the irrigation solution, 2.5% NaOCl generated a smaller amount of debris than CHX gel, presenting a different behaviour. It is assumed that the amount of hypochlorite crystals, although statistically different from the other substances tested when weighed separately, is directly associated with the endodontic technique used.

Distilled water has been consistently used in these types of studies because it does not generate any residue that could influence the final weight of debris (8, 9). However, the results generated in studies using distilled water cannot be translated clinically because, in contrast to NaOCl and CHX gel, it is not used as an irrigation fluid alone.

The three groups with distilled water produced less debris compared to other irrigation solutions, with no significantly different quantities of debris observed. This suggests that residues of other solutions increase debris formation (8). In the present study, the weighing of irrigation solutions alone also corroborates this hypothesis, because the quantity of residue generated by drying distilled water is not statistically significant when compared with residues from 2.5% NaOCl and 2% CHX gel. Among the solutions tested, distilled water exerted less influence on the final mass of apically extruded debris. However, it is not recommended as a primary irrigation solution in routine endodontic practice owing to the lack of antimicrobial properties and low dissolution capacity of organic matter.

CONCLUSION
Different irrigation solutions influenced the amount of apically extruded debris during the preparation of the canal using different instrumentation techniques. The PTU instrument used with 2% CHX resulted in the highest extrusion of debris.

Disclosures
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