Comparison of the Incidence of Postoperative Pain after Using 2 Reciprocating Systems and a Continuous Rotary System: A Prospective Randomized Clinical Trial

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Abstract

Introduction: The objective of the present study was to clinically compare the incidence of postoperative pain and the intake of analgesic medication (frequency and quantity) after endodontic treatment of posterior teeth using 2 reciprocating systems and a continuous rotary system. Methods: In a prospective randomized clinical study, 210 patients with vital teeth indicated for conventional endodontic treatment were treated by 5 specialists according to a pre-established protocol. The teeth were randomly assigned to 1 of 3 groups (n = 70) according to the instrumentation system used: ProTaper Next (Dentsply Tulsa Dental Specialties, Johnson City, TN), WaveOne (Dentsply Tulsa Dental Specialties), or Reciproc (VDW, Munich, Germany). Treatments were performed in a single visit. After the visit, the patients were given a prescription for ibuprofen 400 mg to be taken every 6 hours if they experienced pain. Participants were asked to rate the intensity of the postoperative pain on a visual analog scale according to 4 classes (no pain, mild pain, moderate pain, and severe pain) after 24 hours, 48 hours, 72 hours, and 7 days. Patients were also asked to record the number of prescribed analgesic medication tablets taken at these time points. Results: No statistically significant difference was found among the 3 groups in relation to postoperative pain or analgesic medication intake at the 4 time points assessed (P > .05, Kruskal-Wallis test). Conclusions: The reciprocating systems and the continuous rotary system were found to be equivalent in regard to the incidence of postoperative pain and intake of analgesic medication at the time points assessed. (J Endod 2016;42:171–176)

Key Words

Nickel-titanium instruments, postoperative pain, ProTaper Next, Reciproc, WaveOne

P postoperative pain is defined as the sensation of discomfort after endodontic intervention and is reported by 25%–40% of patients irrespective of pulp and periradicular status (1–3). According to the 2011 systematic review of Pak and White (4), the prevalence of pain in the first 24 hours is 40%, falling to 11% after 7 days. Dentinal debris, pulp tissue, microorganisms, and irrigants can be conveyed to the periradicular tissues during root canal preparation, and such extrusion of debris can lead to postoperative complications, such as flare-ups. Thus, adequate control of the working length (WL) can reduce the extrusion of material through the apical foramen but cannot prevent this completely (5). According to the literature, the incidence of flare-ups during endodontic treatment ranges from 1.4%–16% (6–9). Major advances in rotary instrumentation and metallurgy have led to the introduction of numerous systems with innovative designs in recent years. Nonetheless, all the preparation techniques and instruments available to date are still associated with some degree of extrusion of debris (10–12).

The concept of single-file canal preparation was introduced in endodontics (13) with the launch of Reciproc (VDW, Munich, Germany) and WaveOne (Dentsply Tulsa Dental Specialties, Johnson City, TN) instrumentation systems. These instruments are fabricated with a nickel-titanium alloy called M-Wire using an innovative thermal treatment process (14). The reciprocating motion involves an initial rotation of the instrument in a counterclockwise direction, during which the instrument penetrates and cuts the dentin, and then a rotation in the opposite direction, during which the instrument is released.

The Reciproc system consists of 25.08, 40.06, and 50.05 instruments characterized by an “S”-shaped cross section, spiral flutes with high cutting efficiency, and a gradually decreasing taper after the apical 3 mm. WaveOne system files are available in sizes 21.06, 25.08, and 40.08. The 21.06 (Small) instrument has a constant 6% taper along the entire length of its working part, whereas the 25.08 (Primary) and 40.08 (Large) instruments have an 8% taper from D1 to D3, decreasing progressively from D4 to D16. The WaveOne files have 2 different cross-sectional designs: a modified convex triangular shape from D1 to D8 and a convex triangular shape from D9 to D16.

The ProTaper Next files (Dentsply Tulsa Dental Specialties) operate in continuous rotary motion, and their center of mass or center of rotation is positioned off-center.
relative to the instrument’s central axis of rotation. During rotation, the files of this design produce a mechanical wave of motion, which travels along the length of the working part of the instrument, minimizing the contact between the file and dentin. According to the manufacturer, the offset design of this instrument also improves debris removal and flexibility in the working part of the file (15). To our knowledge, the implications of shifting the center of mass and/or rotation of the central axis of the instruments on debris extrusion and incidence of postoperative pain have not been assessed by clinical trials in the literature. Furthermore, reciprocating instruments have not been thoroughly and clinically compared with those of continuous clockwise rotary motion in regard to the incidence of postoperative pain.

The purpose of the present study was to clinically compare the incidence of postoperative pain after the root canal preparation of posterior teeth using 2 reciprocating systems (Reciproc and WaveOne) and a continuous rotary system (ProTaper Next) through a prospective randomized trial. Analgesic medication intake by patients was also studied. The null hypothesis tested was that there is no difference in the incidence of postoperative pain or intake of analgesic medication after using any of the 3 instrumentation systems.

**Materials and Methods**

This study was approved by the local research ethics committee (069127/2014). Five endodontists experienced in the techniques, materials, and technologies analyzed took part in the study. All specialists followed a pre-established protocol for the ProTaper Next, WaveOne, and Reciproc instrument systems.

**Patient Selection**

A total of 210 patients (133 women and 77 men) aged 19–73 years were included in this study. Sample size calculation was performed using Cochran’s method (1986). Based on a type I error of 0.05 and a power of 80%, a minimum sample size of 43 would be required to detect differences between the 3 study groups. Therefore, the 70 teeth assigned to each group were enough to ensure a representative sample. All participants had maxillary or mandibular molar or premolar teeth indicated for conventional endodontic treatment for prosthodontic purposes diagnosed with vital pulps. Patients with non-vital teeth and cases of apical periodontitis, endodontic retreatment or symptomatic/asymptomatic irreversible pulpitis, root resorption, immature/open apex, or a root canal in which patency of the apical foramen could not be established were all excluded from the study. Patients refusing to participate in the study, whose teeth had issues precluding single-visit treatment, those using some type of medication preoperatively such as analgesics or nonsteroidal or steroidal anti-inflammatory drugs, and patients with any uncontrolled systemic disease were also excluded.

Patients were referred for treatment at 1 of the private dental clinics of the participating endodontists over a 6-month period spanning from June to December 2014. The diagnosis of vital pulp was confirmed by collecting dental history and performing periapical digital radiography, periodontal evaluation, percussion, and cold test (EndoIce; Coltene/Whaledent Inc, Cayahoga Falls, OH). The diagnostic findings were checked by comparing the tooth’s response against that of an adjacent tooth with a vital pulp. The treatment protocol was explained to patients, and all selected patients signed an informed consent form. Although patients had a general notion of the type of instruments to be used, they were not told which specific system would be used in their particular case.

**Random Selection of Instrumentation System**

Of the total sample of 210 teeth, 70 were designated to each of the 3 instrumentation systems. Because the study design included 5 specialists, each professional prepared 42 teeth, 14 per system. The following method was used to ensure random selection: at the outset of the investigation, each endodontist had a dark box at their dental practice containing 14 red, 14 blue, and 14 green tokens; each color represented 1 of the 3 systems investigated in the study. At the beginning of the treatment visit, the clinical assistant randomly determined the instrumentation system to be used for the patient by blindly drawing a colored token from the dark box, without returning it until all the patients had been assigned to 1 of the 3 systems. The groups were allocated as follows: group PTN (red token), preparation using ProTaper Next; group WO (blue token), preparation using WaveOne; and group R (green token), preparation using Reciproc.

**Treatment Protocol**

Local anesthesia was administered and consisted of 2% lidocaine with epinephrine 1:100,000 (Xylocaine; Dentsply Pharmaceutical, York, PA).

After gaining access, the canals were explored with #06, #08, #10, and #15 K-type hand files (Dentsply Maillefer, Ballaigues, Switzerland) according to the initial diameter of the foramen, its degree of flattening, and its canal curvature using a watch-winding motion. The entire procedure was performed under a dental operating microscope (OPMI PICO; Carl Zeiss, Göttingen, Germany).

The WL was established by introducing a #10 K-file up to the apical foramen as determined by a Root ZX II apex locator (J Morita Corp, Kyoto, Japan), and then by withdrawing the file and subtracting 0.5 mm from the length, which was measured with the aid of an endodontic ruler. The WL was confirmed radiographically. All instruments were driven by an electric micromotor with limited torque (VDW Silver Reciproc Motor, VDW). Torque limits and rotation speed were set individually for each file system used. WaveOne and Reciproc instruments were used in a reciprocating mode (WaveOne All and Reciproc All). Debris was removed from the instrument using alcohol-soaked gauze, either immediately after each instrument change (ProTaper Next system) or after 3 in-and-out (pecking) motions (WaveOne and Reciproc systems) according to the manufacturer’s recommendations. Irrigation with 2 mL 2.5% sodium hypochlorite (NaOCl) was performed using a 24-G needle (Max-I-Probe; Dentsply Tulsa Dental, York, PA) during access and a 31-G NaviTip needle (Ultradent Products Inc, South Jordan, UT) when reaching the WL after each file insertion.

The instrumentation sequence used during the treatments in each group followed the procedure recommended by the respective manufacturer.

**Group PTN.** For the PTN group, SX files (originals from the ProTaper Universal system; Dentsply Maillefer, Ballaigues, Switzerland) were used for preflaring of the first two thirds using X1 and X2 (25.06) for preparation of narrow and curved canals and X3 and X4 (40.06) for preparation of large canals up to the WL. The files were worked using a continuous rotary brushing motion at a speed of 300 rpm and a torque of 2 Ncm.

**Group WO.** For the WO group, the Primary file (25.08) was used to prepare narrow and curved canals, and the Large file (40.08) was used for large canals. Three in-and-out motions were applied with stroke lengths not exceeding 3 mm in the cervical, middle, and apical thirds until attaining the established WL.

**Group R.** For the Reciproc group, R25 files (25.08) were used in narrow and curved canals, and R40 files (40.08) were used in large canals. Three in-and-out motions were applied with stroke lengths not
exceeding 3 mm in the cervical, middle, and apical thirds until attaining the established WL.

All the files were used in only 1 tooth (single use) and then discarded. Patency of the apical foramen was maintained during all the techniques by introducing a #10 or #15 K-type file (Dentsply) to a point 1 mm beyond the WL at each instrument change. The preparations for all the groups were finished using a #25 file for narrow or curved canals and a #40 file for wide canals.

After concluding the instrumentation, the coronal chamber was flushed with 1 ml 2.5% NaOCl, and the solution was agitated ultrasonically with a #20/25 Irrisafe tip (Satelec Acteon Group, Merignac, France) placed inside the canal up to 2 mm short of the WL for 1 minute per canal followed by irrigation with 5 ml 17% EDTA solution and agitated ultrasonically for 1 minute to remove the smear layer. Afterward, irrigation was repeated with 5 ml 2.5% NaOCl, finishing the procedure by irrigation with 5 ml 0.9% physiological saline solution. Final aspiration was performed using a capillary tip (Ultradent). An average volume of 40 ml of irrigant was used for all the teeth during preparation. All the teeth were then dried using ProTaper Next and WaveOne absorbent paper points (Dentsply Maillefer) for groups PTN and WO and Reciproc paper points (VDW) for group R.

The canals were subsequently filled with the gutta-percha cones of the respective systems and AH-plus sealer (Dentsply Maillefer) using the continuous wave of condensation technique. The treatment phase was concluded by sealing the coronal access cavity with a dentinal adhesive and composite resin (P60; 3M Dental Products, St Paul, MN). After completing the endodontic treatment procedure, all patients were given postoperative instructions to take analgesics (400 mg ibuprofen) in the event of any other type of emergency.

Patients were instructed to contact the clinic or the dentist in charge in the event of any other type of emergency. The Kruskal-Wallis nonparametric test was applied to compare the incidence of postoperative pain, and the likelihood ratio test was used to compare analgesic medication intake for the 3 groups and the 4 time points assessed. The level of significance adopted was 5% \( (P < .05) \).

### Results

The baseline demographic and clinical features of the study groups are summarized in Table 1. The mean age of the 210 patients enrolled in this study was 47 years. All the patients who underwent endodontic treatment answered the questionnaire satisfactorily at all the time points assessed (24 hours, 48 hours, 72 hours, and 7 days). There was no statistically significant difference \( (P > .05) \) among the ProTaper Next, WaveOne, and Reciproc systems in regard to the incidence of postoperative pain at any of the 4 time points assessed (Table 2). The highest mean postoperative pain scores were observed 24 hours after treatment in all the instrumentation groups with a significant decline thereafter (Fig. 1).

No statistically significant difference was found among the 3 groups assessed in the study in terms of frequency and quantity of analgesic medication intake \( (P > .05, \text{Table 3}) \). In general, analgesic intake was confined to the first 48 hours after treatment in all the groups assessed. None of the 210 participants reported severe pain or flare-ups during the period of the study.

### Discussion

One of the major obstacles to assessing postoperative pain encountered in clinical studies conducted for this purpose is the subjective nature of this evaluation and the inherent difficulty in measuring pain \( (17) \). Therefore, designing the most adequate questionnaire to be applied is a critical step in these studies. The questionnaires must be fully understood by patients and lend themselves to straightforward interpretation \( (17) \). In the current study, the VAS was selected based on its confirmed reliability for pain assessment \( (10, 18, 19) \).

Several factors are involved in the sensation of postoperative pain. This makes clinical investigations that associate pain incidence with possible causes even more challenging \( (20) \). Although mild discomfort is generally expected after undergoing endodontic treatment \( (21) \), the incidence of postoperative pain and flare-up as reported in the literature is estimated as ranging from 3%–58% \( (22) \). Mechanical, chemical, or microbial injuries to periapical tissues are the leading causes of acute periapical inflammation \( (23) \).

Preoperative pain is 1 of the strongest predictors of postoperative pain \( (18) \). Therefore, only teeth with vital pulp indicated for endodontic treatment because of prosthodontic reasons were selected for the present study. Teeth with a nonvital pulp response and symptomatic/asymptomatic apical periodontitis, endodontic retreatment, or symptomatic/asymptomatic irreversible pulpitis were all excluded in an effort to isolate potential factors of postoperative pain from those strictly related to the instrumentation technique. In addition, all the teeth were treated

### Table 1. Baseline Demographic and Clinical Features of Patients in the Study Groups

<table>
<thead>
<tr>
<th>Baseline demographic and clinical features</th>
<th>ProTaper Next, ( n (%) ) ( (n = 70) )</th>
<th>WaveOne, ( n (%) ) ( (n = 70) )</th>
<th>Reciproc, ( n (%) ) ( (n = 70) )</th>
<th>Total ( (n = 210) )</th>
</tr>
</thead>
<tbody>
<tr>
<td>Male</td>
<td>26 (33.77)</td>
<td>24 (31.17)</td>
<td>27 (35.06)</td>
<td>77</td>
</tr>
<tr>
<td>Female</td>
<td>44 (33.08)</td>
<td>46 (34.59)</td>
<td>43 (32.33)</td>
<td>133</td>
</tr>
<tr>
<td>Maxillary teeth</td>
<td>39 (32.23)</td>
<td>42 (34.71)</td>
<td>40 (33.06)</td>
<td>121</td>
</tr>
<tr>
<td>Mandibular teeth</td>
<td>31 (34.83)</td>
<td>28 (31.46)</td>
<td>30 (33.71)</td>
<td>89</td>
</tr>
<tr>
<td>Premolar</td>
<td>12 (31.58)</td>
<td>14 (36.84)</td>
<td>12 (31.58)</td>
<td>38</td>
</tr>
<tr>
<td>Molar</td>
<td>58 (33.72)</td>
<td>56 (32.56)</td>
<td>58 (33.72)</td>
<td>172</td>
</tr>
</tbody>
</table>

SPSS software (Statistical Package for Social Sciences, v. 22.0; IBM Corp, Chicago, IL) was used for the statistical treatment of results. The baseline demographic and clinical features

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in a single visit to rule out the potential influence of intracanal medication or other factors triggering pain.

Nonsteroidal anti-inflammatory drugs have been recommended as first-choice medication for postoperative pain management after endodontic treatment, and ibuprofen has been included in numerous investigations on the effect of different techniques and medications on pain relief after root canal treatment (24, 25). Therefore, ibuprofen was used in the present study as the nonsteroidal anti-inflammatory drug of choice for controlling postoperative pain.

The systems selected for assessment in this study were based on their close match in terms of tip size of the instruments used in different systems with the nature of the prepared root canals in which the final preparations of narrow and curved canals were standardized with #25 files (ProTaper Next X2 25.06, WaveOne Primary 25.08, and Reciproc R25.08) and those of large canals were standardized with #40 files (ProTaper Next X4 40.06, WaveOne Large 40.08, and Reciproc R40.06).

According to the conditions established for this study, there was no statistically significant difference among the instrumentation systems assessed; therefore, the null hypothesis was accepted. This finding contrasts with the results from a randomized clinical trial conducted by Nekooifar et al (26), who found postoperative pain to be significantly lower in patients treated with the ProTaper Universal rotary system than in those treated with the WaveOne reciprocating system. This disparity between the studies may be explained by the differences in the inclusion criteria (diagnosis of irreversible pulpitis versus vital teeth), irrigating solution (chlorhexidine vs NaOCl), and the kinematics adopted during reciprocating instrumentation.

The greatest mean pain in all 3 study groups occurred in the first 24 hours, with a significant reduction in pain ratings at the subsequent observation time points of 48 hours, 72 hours, and 7 days. Similar findings were observed in a systematic review conducted by Pak and White (4) in 2011 in which pain incidence in the first 24 hours was 40%, declining sharply thereafter, particularly over the first 2 days, and reaching levels of 11% at 7 days. The 2004 study by Ng et al (27) also used a VAS and found that 40.2% of the patients had pain 48 hours after obturation, but less than 12% experienced severe pain. Postoperative pain and edema after treatment are more commonly associated with preparation procedures, arising from an immune response to the irrigant, microorganisms present in extruded debris, overinstrumentation, or foreign body reactions to filling materials.

All instrumentation techniques produce some extrusion of debris, but the quantity of material extruded can differ depending on the preparation technique and instrument design of the system (28). Wider-ranging variability in debris extrusion between different mechanical systems has been reported in the literature (29). Bürklein and Schäfer (11) compared debris extrusion in vitro produced by 2 rotary systems (Mtwo [VDW, Munich, Germany] and ProTaper Universal) and 2 reciprocating systems. The authors concluded that full-sequence rotary instrumentation was associated with less extruded debris. Conversely, in an ex vivo assessment of the apical extrusion of bacteria produced using reciprocating files of the Reciproc and WaveOne systems and rotary files of the BioRace system (BioRace, FKG Dentaire, Lachaux-de-Fonds, Switzerland), Tinoco et al (30) observed less extrusion of bacteria when single-file systems were used. According to the same

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**TABLE 2.** Descriptive Statistics and Kruskal-Wallis Test Applied to the Postoperative Pain Results for the Groups Instrumented with ProTaper Next, WaveOne, and Reciproc Systems

<table>
<thead>
<tr>
<th>Instrumentation system</th>
<th>n</th>
<th>Mean</th>
<th>Standard deviation</th>
<th>Minimum</th>
<th>Maximum</th>
<th>25th percentile</th>
<th>50th percentile</th>
<th>75th percentile</th>
<th>P value*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pain after 24 h</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>ProTaper Next</td>
<td>70</td>
<td>0.57</td>
<td>0.80</td>
<td>0.00</td>
<td>2.00</td>
<td>0.00</td>
<td>0.00</td>
<td>1.00</td>
<td>.143</td>
</tr>
<tr>
<td>WaveOne</td>
<td>70</td>
<td>0.63</td>
<td>0.79</td>
<td>0.00</td>
<td>3.00</td>
<td>0.00</td>
<td>0.00</td>
<td>1.00</td>
<td></td>
</tr>
<tr>
<td>Reciproc</td>
<td>70</td>
<td>0.87</td>
<td>0.96</td>
<td>0.00</td>
<td>4.00</td>
<td>0.00</td>
<td>1.00</td>
<td>2.00</td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>210</td>
<td>0.69</td>
<td>0.86</td>
<td>0.00</td>
<td>4.00</td>
<td>0.00</td>
<td>0.00</td>
<td>1.00</td>
<td></td>
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<tr>
<td>Pain after 48 h</td>
<td></td>
<td></td>
<td></td>
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<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>ProTaper Next</td>
<td>70</td>
<td>0.24</td>
<td>0.43</td>
<td>0.00</td>
<td>1.00</td>
<td>0.00</td>
<td>0.00</td>
<td>0.25</td>
<td>.400</td>
</tr>
<tr>
<td>WaveOne</td>
<td>70</td>
<td>0.23</td>
<td>0.42</td>
<td>0.00</td>
<td>1.00</td>
<td>0.00</td>
<td>0.00</td>
<td>0.00</td>
<td></td>
</tr>
<tr>
<td>Reciproc</td>
<td>70</td>
<td>0.21</td>
<td>0.59</td>
<td>0.00</td>
<td>3.00</td>
<td>0.00</td>
<td>0.00</td>
<td>0.00</td>
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</tr>
<tr>
<td>Total</td>
<td>210</td>
<td>0.23</td>
<td>0.48</td>
<td>0.00</td>
<td>3.00</td>
<td>0.00</td>
<td>0.00</td>
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<td></td>
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<tr>
<td>Pain after 72 h</td>
<td></td>
<td></td>
<td></td>
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<td></td>
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<td></td>
<td></td>
</tr>
<tr>
<td>ProTaper Next</td>
<td>70</td>
<td>0.00</td>
<td>0.00</td>
<td>0.00</td>
<td>0.00</td>
<td>0.00</td>
<td>0.00</td>
<td>0.00</td>
<td>.363</td>
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<tr>
<td>WaveOne</td>
<td>70</td>
<td>0.03</td>
<td>0.17</td>
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<tr>
<td>Reciproc</td>
<td>70</td>
<td>0.04</td>
<td>0.27</td>
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<tr>
<td>Total</td>
<td>210</td>
<td>0.02</td>
<td>0.18</td>
<td>0.00</td>
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<td>0.00</td>
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<tr>
<td>Pain after 7 days</td>
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<td></td>
<td></td>
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<td></td>
<td></td>
</tr>
<tr>
<td>ProTaper Next</td>
<td>70</td>
<td>0.00</td>
<td>0.00</td>
<td>0.00</td>
<td>0.00</td>
<td>0.00</td>
<td>0.00</td>
<td>0.00</td>
<td>.368</td>
</tr>
<tr>
<td>WaveOne</td>
<td>70</td>
<td>0.00</td>
<td>0.00</td>
<td>0.00</td>
<td>0.00</td>
<td>0.00</td>
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<td>Reciproc</td>
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<td>0.01</td>
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<td>Total</td>
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<td>0.07</td>
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<td>1.00</td>
<td>0.00</td>
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<td>0.00</td>
<td></td>
</tr>
</tbody>
</table>

*Kruskal-Wallis test, P < .05.

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**Figure 1.** Mean postoperative pain scores after instrumentation with ProTaper Next, WaveOne, and Reciproc systems at the 4 time points assessed (24 hours, 48 hours, 72 hours, and 7 days).
authors, the variation observed could be attributed to differences in the cross section, cutting-edge design, taper, tip type, configuration, use concept, flexibility, alloy type, number of files used, kinematics, or cutting efficacy.

In the present study, we sought to minimize the variation in the manipulation method by establishing the protocols based on recommendations by the manufacturers. Instrumentation was performed on root thirds using 3 in-and-out motions with stroke lengths not exceeding 3 mm for cases in which reciprocating systems were used and using a brushing action for cases in which the rotary system was used. These protocols were adopted in view of the lack of specific recommendations in the related literature for motion amplitude and number of repetitions to be used during instrumentation.

In *in vitro* studies have shown that reciprocating systems can lead to a greater amount of extruded debris (11) or debris remnants in the root canal (31) than rotary systems, possibly as a result of the reverse motion of the reciprocating instrument. Although it may be hypothesized that a greater amount of extruded debris could be related to a higher incidence of pain after treatment, the differences in instrument design and preparation technique among the systems examined in the current clinical study seemed to have no influence on the incidence of postoperative pain.

The standardized and controlled kinematics used in this study may also have contributed to minimizing debris remnants or extrusion thereof, thereby reducing the possible contribution of this factor to the occurrence of postoperative pain.

In conclusion, the incidence of postoperative pain and the intake (frequency and quantity) of analgesic medication prescribed for all the postoperative time points were similar across all 3 types of instrumentation systems assessed in this study.

## Acknowledgments

The authors deny any conflicts of interest related to this study.

## References