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Effects of Simultaneous Liquid or Gel Sodium Hypochlorite Irrigation on the Cyclic Fatigue of Two Single-File Nickel-Titanium Instruments

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Featured Application: The proposed customized device could represent a way to standardize cyclic fatigue tests of nickel-titanium instruments in the presence of irrigant solutions. Future studies could pursue laboratory investigations on nickel-titanium instrument surface exposed to irrigant solutions under these standardized methodological conditions.

Abstract: To evaluate the effect of simultaneous liquid or gel sodium hypochlorite (NaOCl) irrigation on cyclic fatigue of F6 SkyTaper (F6ST) and OneCurve (OC) single files, 180 new 25/0.06 F6ST and OC files were divided into 6 groups ($n = 15$) for each brand. Groups 1 and 4 included new instruments not exposed to NaOCl at 20 °C and 37 °C, respectively. Groups 2 and 5 included files activated with liquid NaOCl at 20 °C and 37 °C, respectively. Groups 3 and 6 consisted of instruments tested with NaOCl gel at 20 °C and 37 °C, respectively. Instruments were subjected to a fatigue test using a novel customized device. Data were expressed as time to fracture (TtF) and statistically analyzed ($p < 0.05$) after checking their normality through the Shapiro–Wilk test. Because they were normally distributed, 2-way analyses of variance (ANOVA) and the Tukey multiple comparison post-hoc test were used. Time to fracture of all tested instruments decreased at 37 °C ($p < 0.05$). At 20 °C, NaOCl improved TtF of F6ST and OC ($p < 0.05$). NaOCl liquid increased TtF of F6ST ($p < 0.05$) in comparison with gel, while there was no difference between the two formulations for OC. At 37 °C, both NaOCl formulations had no significant influence on TtF for F6ST, while they increased TtF of OC ($p < 0.05$). NaOCl improved the cyclic fatigue resistance of OC, independently of the temperature, while for F6ST the negative impact of higher temperature reduced the irrigant benefits.

Keywords: cyclic fatigue resistance; F6 SkyTaper; gel; OneCurve; sodium hypochlorite; temperature

1. Introduction

Nickel-titanium (NiTi) alloy is widely employed in endodontics due to its properties of superelasticity and shape memory [1]. Nevertheless, NiTi instruments are subjected to an unexpected separation, due to the cyclic fatigue and torsional fracture, during the clinical use [2,3]. Because of this, producers have tried to propose new designs, manufacturing processes as heat treatments and protocols to reduce the fracture risk [4]. Heat treatments change the transitional temperature of martensitic

transformation, favoring a high percentage of the martensitic phase to increase the flexibility of heat-treated NiTi [5,6]. Moreover, the surrounding temperature influences the crystalline phase of NiTi alloy [6–9]. Single-file systems have been presented to facilitate the instrumentation protocols and minimize mechanical stresses [10]. In addition, the shaping phase with a single-file system may be more rapid in comparison with traditional multi-file sequences [11,12]. F6 SkyTaper (F6ST; Komet Dental, Gebr. Brasseler, Lemgo, Germany) is an example of a single-file system made by traditional NiTi alloy available in four different sizes, including 25 size. All instruments have a continuous taper of 0.06 and an S-shaped cross-section [13]. Another single-file system is the OneCurve endodontic file (OC; Micro Méga, Besançon, France), made from a NiTi alloy that receives a patent-protected heat treatment (C-Wire) which provides to the file a shape-memory effect. OC files also present a tip size of 25 and a constant taper (6%) with a variable cross-section [12].

It is suggested that root canal preparation be performed with sodium hypochlorite (NaOCl) due to its antimicrobial activity and lubricity (in liquid or in the most recent gel formulation), which facilitates the mechanical action of endodontic files [14,15]. Nevertheless, the contact of NiTi files with NaOCl during instrumentation raises doubt about the consequence of such short-term contact on the integrity of the instruments [14]. A previous study evaluated the effects of liquid and paste-type lubricants on torque values during simulated rotary root canal instrumentation, but in that case ethylenediamine tetraacetic acid (EDTA) effects were evaluated [16]. In particular, it was reported that irrigation solutions were generally more efficacious than the paste-type chelator in reducing torque. Nevertheless, the study did not completely mirror a clinical situation; moreover, the results obtained were limited to the type of lubricant used and instrument tested [16]. In addition, the potentially corrosive effect of NaOCl has been investigated performing the cyclic fatigue test after the files' exposure to NaOCl or when the instruments were immersed in the irrigant solution [7,14,17]. Nevertheless, neither case allowed an irrigant flow simultaneous to the fatigue test.

To date, no studies have investigated the effects of different NaOCl formulations on the cyclic fatigue resistance of single-file instruments at body temperature. Thus, the aim of this study was to evaluate the influence of liquid and gel sodium hypochlorite on the cyclic fatigue of F6 SkyTaper and OneCurve single-file systems at body temperature. For this purpose, a new customized testing machine, which allowed us to inject the irrigant solutions (liquid or gel) simultaneously to the fatigue test, was used. The null hypothesis was that there would be no significant differences among the cyclic fatigue resistance of the NiTi files which were exposed to different NaOCl formulations at different temperatures.

2. Materials and Methods

2.1. Study Design

A pilot study was performed to establish the sample size calculation. The minimum sample size was fixed at 15 instruments for each group ($n = 15$) to ensure a test power of 0.80 (G*Power 3.1.9.2 software, Heinrich-Heine-Universität Düsseldorf, Düsseldorf, Germany) with $\alpha = 0.05$ and $\beta = 0.95$. Therefore, 180 new 25/0.06 F6ST and OC files of 25-mm length were selected. All instruments were previously examined through an optical stereomicroscope with 20 magnification (SZR-10; Optika, Ponteranica, Bergamo, Italy) for any evidence of visible deformation. No instrument was discarded. Instruments, all from the same production lot, were randomly allocated into 6 groups ($n = 15$) for each brand. Groups 1 and 4 (the control groups) included new instruments that were tested without NaOCl contact at room (20 °C) or body (37 °C) temperature, respectively. Groups 2 and 5 were composed of instruments tested with liquid NaOCl (Chloraxid 5.25%, PPH CERKAMED, Stalowa, Poland) at 20 °C or 37 °C, respectively. The remaining groups (groups 3 and 6) received instruments that were tested with NaOCl gel (Chloraxid 5.25% GEL, PPH CERKAMED, Stalowa, Poland) at 20 °C or 37 °C, respectively. According to the manufacturer, the two formulations present the same composition

(i.e., sodium hypochlorite (5.25% of active chlorine), purified water European Pharmacopoeia) except for the gel base (ethoxylated behenyl alcohol, glycol solvent) present in the Chloraxid 5.25% GEL [18,19].

2.2. Cyclic Fatigue Testing

A customized static cyclic fatigue testing device was used [20] (Figure 1). The device ensured a standardized instrument insertion through a fixed block and the possibility to test the files at different inclinations. For this study, all instruments were tested at the standard position (0°).

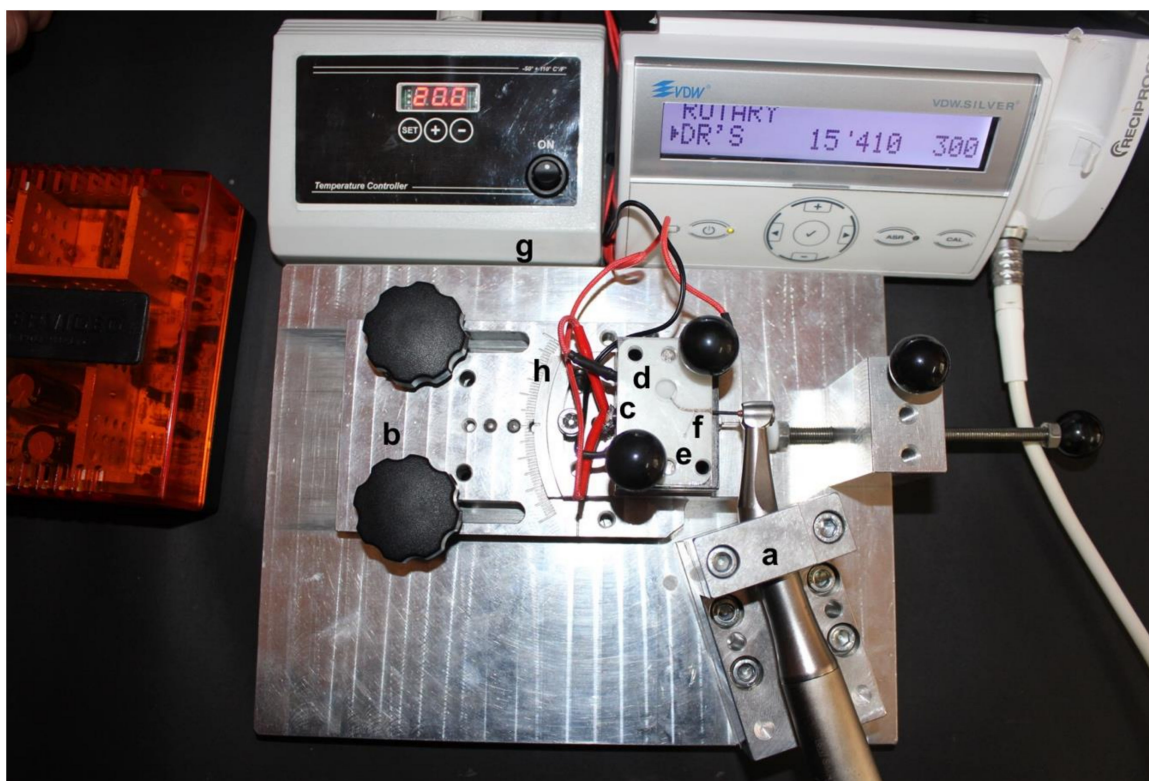


Figure 1. The custom-made apparatus for cyclic fatigue tests with an OneCurve (OC) file and a preset temperature of 20°C . (a) The fixed block in which the electric handpiece is inserted and (b) the mobile support on rails to allow the insertion/withdrawal of the file; (c) the ceramic canal with a 60° angle and a 5-mm radius covered by the superimposition of two Plexiglas slides; (d) the hole made on the cover near the corresponding canal apex to permit the outflow of the solution during the test through an aspiration system; (e) the insertion point of the irrigant tip and (f) the channel built inside the cover to permit the irrigant flow during the test; (g) the thermostat which allowed the temperature adjustment through to a thermocouple (h) applied to the artificial canal.

A simulated 16-mm-long ceramic canal was made by replicating the instrument's size and taper. It presented a curvature with a 5-mm radius (calculated at the internal concave surface of the artificial canal), a 60° angle of curvature assessed using the Schneider's method [21], a center of curvature 5 mm from the end of the canal and a curved segment of 5 mm in length [14]. The ceramic artificial canal was milled in a zirconium oxide disc (Dental Zirconia Pre-Shaded Blank, Liaoning Upcera Co., Ltd., Shenzhen, Guangdong, China) using the milling machine Roland DWX-51D (Roland DG Mid Europe, AP, Italy). To permit the injection/expulsion of the liquid or gel sodium hypochlorite from the artificial canal, a cover formed by the superimposition of two Plexiglas slides was employed. A 1-mm diameter channel was built inside this cover in correspondence with the artificial canal to permit the irrigant flow in the system, ensuring the absence of contact between the syringe and the instrument or the zirconium canal. The insertion point of the irrigant syringe was placed at the beginning of this channel.

A small circular hole was made on the cover near the corresponding canal apex to ensure the outflow of the solution during the test by means of an aspiration system (Figure 2). Moreover, a thermostat was associated to the customized device to allow the temperature adjustment in the artificial canal. The temperature was kept constant during the test using a thermocouple applied to the artificial canal and which activated or deactivated the thermostatic resistance when the temperature decreased or reached the preset one respectively. In particular, the instruments were tested at room temperature ($20\text{ }^{\circ}\text{C} \pm 1\text{ }^{\circ}\text{C}$) and body temperature ($37\text{ }^{\circ}\text{C} \pm 1\text{ }^{\circ}\text{C}$).



Figure 2. A detail of device with a F6 SkyTaper (F6ST) instrument exposed to NaOCl gel. (a) Irrigant tip insertion; (b) channel made inside the cover; (c) aspiration system allocated near to the canal apex.

A 6:1 reduction handpiece (Sirona Dental Systems GmbH, Bensheim, Germany) activated by a torque-controlled motor (Silver Reciproc, VDW, Munich, Germany) was used to turn on 15 instruments of each system in continuous rotation (300 rpm) as indicated by the manufacturer. Torque was fixed at the maximum value provided (4.1 Ncm). Each instrument was located in the contra-angle handpiece and inserted into the canal at the same length (16 mm). The rotation started at the preset temperature, and this temperature was kept constant for the whole duration of the test. For groups 1 and 4 (control groups), the friction of the file on the artificial canal walls was reduced by employing a special high-flow synthetic oil (Super Oil; Singer Co Ltd., Elizabeth, NJ, USA) projected for lubrication of mechanical parts. Experimental protocols (for liquid and gel solutions) included a continuous and constant injection of irrigant for 10 s every 30 s of rotation of the instruments. The solution injection was able to pass through the irrigant syringe (2 mL) thanks to a suitable tip provided by the manufacturer, which was placed in correspondence with the insertion point at the beginning of the artificial channel. The flow rate of irrigant was constantly registered and maintained constant for all the experimental procedures. A control device similar to the ones used in the previous studies [22,23] was employed to

guarantee a constant axial velocity of 8.6 m/s, which is congruous with a clinically realistic irrigant flow rate of 0.26 mL/s provided by a 30-G needle [23]. In both protocols, the aspiration system was activated during the test to avoid the permanence of the irrigant in the system beyond the set times. Cyclic fatigue resistance was expressed as time to fracture in seconds (TtF), which was recorded from the start of the test until the visual and audible point of breakage by a chronometer presenting an accuracy of 0.1 s. To exclude human error, a video recording was made simultaneously, and the recordings have been controlled to cross-check the time of file fracture. A digital microcaliper (Mitutoyo Italiana srl, Lainate, Italy) was used to assess the length of the broken file tip. The fractured fragments were investigated by a field-emission scanning electron microscope (Nova NanoSEM 450; FEI, Eindhoven, the Netherlands) with magnification from 480x to 600x to evaluate the morphology of the fracture surface.

2.3. Statistical Analysis

Data were subjected to the Shapiro–Wilk test to characterize their normality. Because they were normally distributed, data were expressed as means and standard deviations and statistically evaluated using 2-way analyses of variance (ANOVA) and the Tukey multiple comparison post-hoc test (Prism 7.0; GraphPad Software, Inc, La Jolla, CA, USA) with the significance level established at 5% ($p < 0.05$).

3. Results

The means and standard deviations of TtF are presented in Table 1.

Table 1. Time to Fracture in Seconds of Each Group as the Mean $\pm$ Standard Deviation.

Type of Irrigation	Instrument			
	F6 SkyTaper		One Curve	
	20 °C $\pm$ 1 °C	37 °C $\pm$ 1 °C	20 °C $\pm$ 1 °C	37 °C $\pm$ 1 °C
Without NaOCl	120 ^{a1} $\pm$ 15	90 ^{b1} $\pm$ 14	180 ^{c1} $\pm$ 14	146 ^{d1} $\pm$ 18
Liquid NaOCl 5.25%	186 ^{a2} $\pm$ 18	98 ^{b1} $\pm$ 11	279 ^{c2} $\pm$ 20	225 ^{d2} $\pm$ 21
Gel NaOCl 5.25%	147 ^{a3} $\pm$ 13	92 ^{b1} $\pm$ 11	269 ^{c2} $\pm$ 16	215 ^{d2} $\pm$ 17

Different superscript letters show significant differences between the same instruments at different temperatures as well as between the two brands in the same irrigation group ($p < 0.05$). Different superscript numbers show significant differences between irrigation groups in the same brand of files ($p < 0.05$).

The inferential analysis revealed statistically significant differences between the different types of irrigation, considering the temperature as the independent variable (two-way ANOVA, $p < 0.05$; interaction < 0.05). Moreover, there were statistically significant differences between the temperatures considering the type of irrigation as the independent variable (two-way ANOVA, $p < 0.05$). Post-hoc analysis revealed that the cyclic fatigue resistance of all instruments, independently from the testing conditions, decreased at 37 °C ($p < 0.05$). At 20 °C, NaOCl improved cyclic fatigue resistance of F6ST and OC compared to the control group ($p < 0.05$). However, liquid irrigant significantly improved the TtF of F6ST compared to the gel one ($p < 0.05$), while no difference was observed between the two formulations for OC ($p > 0.05$). At 37 °C, no difference was observed among the control and experimental groups for F6ST ($p > 0.05$). For OC, groups treated with hypochlorite revealed significantly higher TtF than the control group ($p < 0.05$), with no difference between liquid and gel NaOCl ($p > 0.05$). Independently of the temperature and irrigant use, OC reported significant higher TtF than F6ST ($p < 0.05$).

The length of the fractured file fragments was not statistically different among the tested instruments (mean = 5.1 mm) ($p > 0.05$). Field-emission scanning electron microscopic analysis of the fractured surfaces exhibited the representative signs of cyclic fatigue failure for both instruments. The crack initiation areas, the presence of fatigue striations and the overload fast fracture zone induced by cyclic fatigue are visible in the fracture surface of all instruments (Figure 3A–L). More specifically, crack initiation was generated at the cutting edges of the fracture cross sections, with microscopic

dimples present on the separation area. None of the tested files showed pitting or crevice corrosion with or without 5.25% NaOCl as assessed through field scanning electron microscopy.

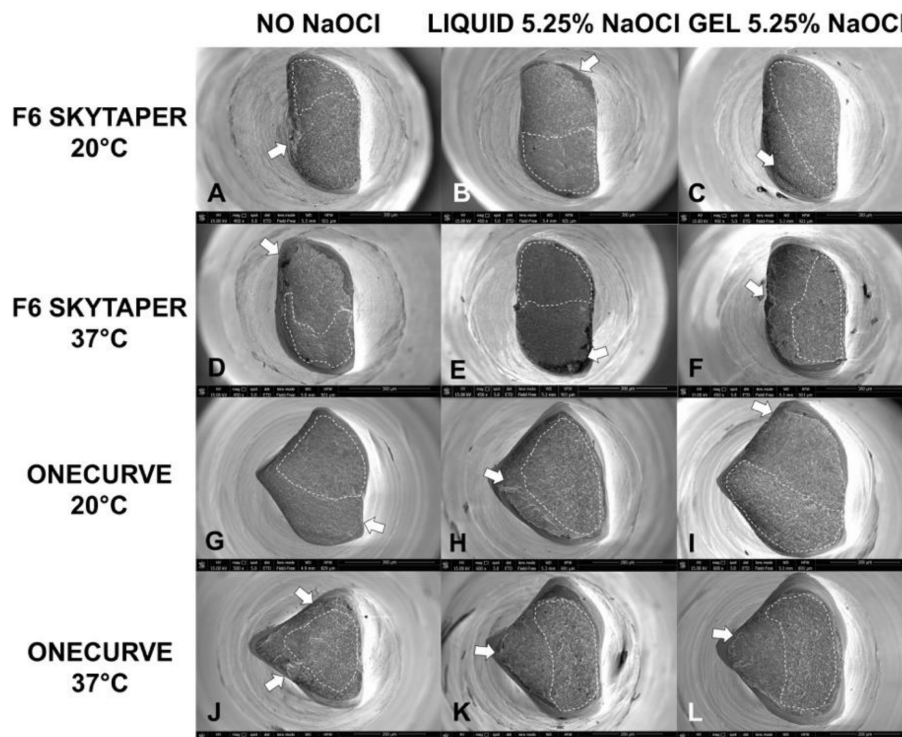


Figure 3. (A–L) Scanning electron micrographs of the fracture surface of F6 SkyTaper and OneCurve fragments in the axial view ($450 \times -600 \times$ magnification) after the cyclic fatigue tests in the different experimental conditions. The white arrows indicate the origins of the crack, and the dotted area indicates the final abrupt breakage. The typical surface pattern of the fibrous fatigue zone showed dimples and cones in all of the fracture surfaces.

4. Discussion

On the basis of the results obtained, increased temperature negatively affected the cyclic fatigue resistance of the tested instruments, as previously reported [6,8,9,14]. Consequently, the null hypothesis was rejected. The phase constitution of NiTi files at a specific temperature assumes a key role in defining their mechanical properties [24]. At higher temperatures than the transformation level, the NiTi alloy exhibits an austenite structure which is more rigid, whereas it possesses a martensite structure, which is more flexible, at lower temperatures [5]. Therefore, the progressive reduction in lifespan of the NiTi instruments tested at higher temperatures could be caused by the progressive transition to an austenite phase [6]. At 20 °C, NaOCl increased cyclic fatigue resistance of F6ST and OC files, with liquid formulation performing significantly better than gel solution for F6ST. It is possible to hypothesize that this result is due to wetting properties of the irrigant solution, which could reduce flexural stress of NiTi files; further research is needed to confirm these hypotheses. The outcomes obtained are in contrast with those of Alfawaz et al. who reported that NaOCl negatively affected the fatigue behavior of NiTi instruments [7]. These differences are probably due to the different experimental procedures. Indeed, in that study, the block containing the artificial canal was fixed inside a recipient that was filled with 2.5% NaOCl or 5.25% NaOCl. This condition is different from our methodological set-up, in which NaOCl flow occurs in the artificial canal simultaneously to the fatigue test. Moreover, an artificial metallic canal was used, while in the current study a ceramic canal, which reduces the risk of corrosive phenomena, was employed. In addition, a different type of

instrument (i.e., ProTaper Gold) with metallurgical properties different from those of F6ST and OC was chosen. On the other hand, Huang et al. reported 5.25% NaOCl did not significantly influence the cyclic fatigue resistance of NiTi files. In a similar way to the present study, Huang et al. used an artificial ceramic canal, avoiding the potential corrosion of the test models made by metallic parts [14]. In addition, it is reasonable to think that liquid solutions are more effective than gel probably due to the wetting properties and penetration ability of liquid formulation [25]. Indeed, the modification of the consistency solution modifies its resistance to deformation (the so-called viscosity), wettability and surface tension. Specifically, gel components provide high viscosity to the formulation, reducing its capacity to flow in comparison with liquid [25,26]. However, no significant difference was observed between liquid and gel formulation for OC at either temperature. These results are probably due to the differences in time to fracture between the two files. Indeed, OC instruments showed higher TtF in comparison with F6ST ones, at both temperatures, probably due to the advantages provided by its heat treatment [6,12,27]. Consequently, OC files remain active for more time; it is plausible to suppose that the potential differences between gel and liquid NaOCl formulations are not such as to generate significant variations for prolonged periods.

At 37 °C, no significant difference was observed among the control and experimental groups for F6ST. However, hypochlorite significantly improved TtF of OC files. It is possible to speculate that the different behavior of the two files depends on the new proprietary thermal treatment employed to manufacture the OC files (C-Wire). Indeed, F6ST files are made from a conventional austenite 55-nickel-titanium alloy which shows an austenite finish temperature (A_f) below the body temperature [28,29], while OC files are constituted of a supposed control memory NiTi wire exhibiting an A_f between 40 °C and 50 °C [6]. Therefore, the gradual transition to the austenitic phase is more pronounced at 37 °C for F6ST files in comparison with OC ones. The negative impact of increased temperature could overcome the irrigant benefits for F6ST instruments [30]. Differently from our outcomes, a previous study reported that at body temperature, NaOCl did not significantly improve the cyclic fatigue resistance of OC [31]. This difference is likely a consequence of dissimilar methodological conditions, including the NaOCl concentration tested (2.5% vs. 5.25%) and experimental apparatus (stainless steel block immersed in 2.5% NaOCl solution vs. continuous NaOCl flow).

It is difficult to compare NiTi instruments with different features (geometrical and metallurgical). Nevertheless, in agreement with previous findings, the fatigue life of heat-treated NiTi files (OC) was longer than non-heat-treated files (F6ST) in all testing conditions. This behavior is probably due to the greater flexibility of OC files caused by the heat treatment [6,32,33].

The principal novelty and strength of the apparatus described is the possibility to activate instruments simultaneously to the irrigant flow. Even if the previous studies investigated the corrosive behavior of sodium hypochlorite on NiTi files, methodological conditions did not allow a good simulation of the real clinical situations. Indeed, fatigue tests were performed after a previous exposure of NiTi files to NaOCl for non-standardized time periods [17,34,35] or while the instruments were immersed in low NaOCl concentrations [36,37]. Both scenarios could induce a prolonged or reduced contact between instrument and NaOCl solution. Moreover, the new customized apparatus ensured standardized methodological conditions, thus allowing the reproducibility of tests performed. In addition, a zirconium artificial canal was adopted for cyclic fatigue tests. Zirconia appears to be a suitable material because it possesses mechanical properties similar to stainless steel but avoids the potential corrosion phenomena of the metallic parts used [14,38,39]. Furthermore, the present study provides new relevant information about influence of irrigants on the cyclic fatigue resistance of F6ST and OC instruments, which are both single-file systems widely used in clinical practice. Despite these advantages, the clinician should apply these results in clinical condition with attention. First of all, in vitro conditions vary from intracanal instrumentation, in which the fracture is the result of numerous factors such as torsional stress [40], anatomic canal configuration and the kind of instrument used [20]. Although methodology ensured a standard approach, instruments could be subjected to different friction levels due to the configuration of their blades during clinical use [41]. In addition, a possible

comparison between the two files requires caution because cyclic fatigue resistance is influenced by several factors [6,12,20], including geometrical variables such as cross section (a constant S-shaped for F6ST and triangular for OC at 5 mm from the tip) [12,28], pitch width and rake angle as well as metallurgical features. Lastly, the type of irrigant used as well as its concentration and usage conditions (such as temperature and flow rate) may influence results obtained. Further studies are necessary to confirm these in vitro results; moreover, supplementary examinations to evaluate different kind of irrigants and instruments are auspicious, and more sophisticated laboratory analysis of the surface of NiTi instruments exposed to irrigants' action under these methodological conditions could be performed.

5. Conclusions

Within the limits of the present in vitro study, the use of hypochlorite solutions improved the cyclic fatigue resistance of OC, independently from the temperature, while for F6ST the negative impact of higher temperature reduced the solutions' irrigant benefits.

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