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Effect of A Clinical-Replicable Cooling Protocol on the Cyclic Fatigue Resistance of Heat-Treated Nickel-Titanium Instruments

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Introduction: The aim of this study was to evaluate the bending and cyclic fatigue resistance of Wave One Gold (WOG) and X1 Blue (X1B) instruments when tested at body temperature (36°C ± 1°C) with and without subjected to an alloy cooling protocol. **Materials and Methods**: A total of sixty instruments (n=30) were tested. Forty instruments (n=20) were randomly selected and divided into two groups: body temperature (BT; n=20) and body temperature with cooling protocol (CP; n=20). Cyclic fatigue test was performed until fracture in a conventional stainless-steel device with water bath equipment to simulate body temperature. CP group instruments were subjected to 5 seconds of spray cooling every 30 seconds. Time to fracture was recorded in seconds. Resistance to bending at an angle of 45 degrees was evaluated using twenty instruments (n=10). Fractured surfaces were examined under scanning electron microscopy (SEM). Statistical analysis was performed at a 5% significance level. Results: There was no difference in the cyclic fatigue resistance between instruments in BT groups (P>0.05). Cooling protocol significantly increased the cyclic fatigue resistance of X1B instruments (P=0.0003) and WOG instruments (P=0.0003). **Results:** WOG instruments had a significantly lower cyclic fatigue resistance compared to X1B instruments in CP group (P=0.0001). There were no significant differences between the values of resistance increase presented by the instruments after cooling (P>0.05). Bending test presented no statistically significant differences between the tested instruments (P>0.05). Both instruments in both groups showed typical features of cyclic fatigue behavior under SEM. **Conclusions**: X1 Blue #25.06 and WaveOne Gold #25.07 instruments presented similar cyclic fatigue resistance. The investigated clinical-replicable cooling protocol improved the cyclic fatigue resistance of the tested instruments, with X1 Blue #25.06 presenting a greater cyclic fatigue resistance after cooling. Both instruments presented a similar bending capacity.

Keywords: Body Temperature; Cooling; Cyclic Fatigue; Heat Treatment; Nickel-Titanium Alloy

Introduction

The fracture of endodontic instruments mainly occurs during the preparation of curved and constricted root canals [1]. However, despite having better mechanical properties when compared to stainless steel instruments, nickel-titanium (NiTi) instruments can also fracture due to torsional stress or cyclic fatigue. The latter mainly ensues in curved canals when the instrument is subjected to repetitive tension and compression cycles at the point of maximum flexure [1, 2], occurring unexpectedly and not presenting deformation prior to fracture [3].

To enhance mechanical properties, improvements have been proposed by manufacturers with the intent of providing greater safety and efficiency of these instruments for clinicians [4]. Technologies *e.g* the heat treatments of NiTi alloy seek to increase the resistance to cyclic fatigue [5-7] and the instrument's bending capacity [8].

The heat treatment of NiTi alloys is performed through temperature variations during the manufacturing process of the endodontic instrument(s), achieving a different microstructural arrangement and promoting greater flexibility [9-11] .

Recently, it has been found that a decrease in the instrument temperature has an effect on its mechanical properties, proportionally increasing the resistance to cyclic fatigue as the temperature of alloy decreases, while it is drastically reduced when the temperature of the allow increases [12-14]. However, up to date, none of the investigation tested a cooling protocol which could be replicated in clinical situations.

Therefore, the purpose of the present study was to evaluate a clinical-replicable cooling protocol to increase the resistance to cyclic fatigue. Two heat-treated reciprocating NiTi instruments were tested. (i) Wave One Gold (WOG) Primary (Dentsply Sirona, Ballaigues, Switzerland), a #25 tip size and 0.07 taper at its first three millimeters, presenting a Gold treatment and a parallelogram cross-section design [6]; and (ii) X1 Blue (X1B) 25.06 (MK Life Medical and Dental Products, Porto Alegre, Brazil), a #25 tip size and 0.06 taper at its first three millimeters, presenting a heat treatment similar to the Blue treatment and a convex triangular cross-section[6].

Both instruments were tested in simulated body temperature with and without being subjected to a cooling protocol. Moreover, the bending capacity of both instruments were evaluated. The null hypotheses of the study were: (*i*) there were no differences in the cyclic fatigue resistance between the tested instruments when subjected or not to alloy cooling and (*ii*) there were no differences between the bending capacities of both instruments.

Materials and Methods

Sample size calculation

The sample size calculation was performed using G*Power v3.1 for Mac (Heinrich Heine, Universität Düsseldorf, Düsseldorf, Germany) by selecting the Student *t* test. The data obtained in a previous study [6] was used and the effect size in the present study was established (=1.70). Furthermore, an alpha type error of 0.05 and beta power of 0.95 were stipulated. A total of 9 samples per group was indicated as the ideal size required for observing significant differences. However, 10 instruments per groups were used to compensate for possible sample losses.

A total of 60 instruments were used. For the cyclic fatigue test, 40 instruments were selected for the cyclic fatigue test. The instruments were divided into two groups (n=20): (*i*) BT-tested at body temperature (36°C±1°C); (*ii*) CP-tested at body temperature (36°±1°C) subjected to a cooling protocol with ice spray (Endo-Frost, Roeko, Langenau, Germany) for 5 seconds on the instruments surface every 30 seconds. For the bending test, 20 instruments were selected (n=10). For all tests, all instruments were previously inspected under a stereomicroscope (Carl Zeiss, LLC, USA) at x16 magnification to detect possible defects or deformities before the mechanical testing [15].

Cyclic fatigue test

This test was performed using a custom-made device with a 60º angle and a 5-mm radius of curvature, which located 5mm from their tips, that simulated an artificial canal made of stainless steel; reproducing the size and taper of instruments. The curvature of the artificial canal was fitted onto a cylindrical guide made of the same material. An outer arch had a 1-mm-deep groove, serving as a guide path for the instruments, which kept the instruments reciprocating freely on the curvature during the entire test. This device allows an accurate and reproducible position of the curvature to be established for all instruments, as described in a previous study [16].

The cyclic fatigue test was performed at simulated body temperatures (36°C±1°C) using a histology water bath equipment (Leica HI 1210; Nussloch, Germany), allowing the control of temperature [16]. A total of 600 mL of water was used to fill the equipment container to the desired level, allowing the simulated canal to be submerged on water. The temperature was controlled using a digital thermometer of the equipment and infrared thermometer throughout the test. The instruments were operated by a 6:1 reduction handpiece (Sirona Dental Systems, GmbH, Bensheim, Germany) powered by an endodontic motor (Silver Reciproc, VDW, Munich, Germany) set in “WaveOne All” program, at a speed of 350 rotations per minute (rpm) and a torque of 2 Newtons per centimeter (N.cm).

In CP group, after every 30 seconds of the instrument’s activation, the device was turned off and removed from water for the application of cooling spray, during 5 seconds, aiming directly at the tip of the instrument. For the application of the cooling spray, the instrument was positioned perpendicular to the cooling spray. Immediately after the cooling protocol, the device was re-inserted into water and turned back on. In this part of the experiment, the timer was paused during cooling and started again when the handpiece was activated.

All instruments were operated until a fracture occurrds. A digital chronometer measured the time to fracture, in seconds. Video recordings were made during all the tests, to precisely determine the time and moment of fracture.

Bending resistance test

This test was performed using a torsion machine (Analógica, Belo Horizonte, Brazil) adapted to the ISO 3630-1 specifications, as previously described [17]. A total of 10 instruments of each manufacturer was used to evaluate the flexibility and maximum force required to bend the instrument at 45° angular deflection.

The instruments were fixed at 3mm from the tip and perpendicularly to motor axis. The bending angle (45°) was measured and controlled by a resistive angular transducer connected to a process controller. The force required to bend the instruments was automatically measured by the load cell and then recorded by a machine's specific program (MicroTorque; Analógica), as previously described [17]

Scanning electron microscopy evaluation

All instruments subjected to the cyclic fatigue tests were examined under scanning electron microscope (SEM - JEOL, JSM-TLLOA, Tokyo, Japan) to determine the topographic features of the fractured surface of the instruments submitted to the cyclic fatigue. Before SEM evaluation, the instruments were ultrasonically cleaned (L100, Schuster, Santa Maria, RS, Brazil) in saline solution for 3 minutes. Instruments were examined at ×150 magnification.

Statistical analysis

The mean values and standard deviations of time to fracture and bending resistance were calculated for each system. Shapiro-Wilk test was performed to verify the presence or absence of normality. Data was subjected to two-way ANOVA (time to fracture), Mann-Whitney Test (percentage of resistance increase) and Unpaired T test (bending resistance). The Prism 6.0 software (GraphPad Software Inc., La Jolla, CA, USA) was used as the analytical tool, and the level of significance was set at 5%.

Results

The values (mean and standard deviations) of the time to fracture at body temperature with and without cooling are shown in Table 1. There was no statistical difference on the cyclic fatigue resistance of both instruments in the BT groups ($P>0.05$). A significant increased resistance was observed for the X1B instruments ($P=0.0003$) and WOG instruments ($P=0.0003$) in the CP groups compared to the BT groups. WOG instruments had a significant lower time to fracture than the X1B instruments in the CP ($P=0.0001$) groups. No differences were verified between the values of resistance increase presented by the instruments ($P>0.05$).

Table 1. Mean (SD) of the time to fracture (TTF) in seconds of the instruments subjected to the cyclic fatigue test in body temperature without cooling (BT) and body temperature with cooling protocol (CP) groups

	BT group	**CP group**
Instrument	TTF (sec)	TTF (sec)
X1 Blue	204.78 ^{aA} (91.8)	467.70 ^{aB} (159.78)
Wave One Gold	109.02 ^{bA} (21.38)	214.08 ^{bB} (34.96)

Different superscript lowercase letters in columns represent significant differences amongst the instruments ($P<0.05$); Different superscript uppercase letters in rows represent significant differences of the instruments in different temperatures ($P<0.05$)

Table 2 shows the median and values of 25th and 75th percentiles of resistance increase after cooling for both instruments and mean and standard deviation of bending test (N.cm). There were no statistical differences of the resistance values among groups ($P>0.05$). The bending test showed that there were no differences between the instruments tested ($P>0.05$).

The fracture surfaces at different temperatures observed by SEM showed typical features of cyclic fatigue, such as ductile morphologic characteristics, signs of cracks, fatigue striations and dimple fracture zone in the fracture location, without plastic deformation on their helical shafts. (Figure 1).

Discussion

NiTi alloys has an interesting property of promoting their change of phase in response to temperature changes or applied mechanical stress. Regarding the responses to temperatures, when the alloy is subjected to cold, reaching temperatures below the martensite finish temperature, the NiTi alloy is more ductile and flexible, presenting a greater bending capability and the shape memory effect (martensitic phase); meanwhile, when subjected to heat, and reaching temperatures above the austenite finish temperature, the NiTi alloy returns to its original shape, is stiffer, harder, and presents superelastic properties (austenitic phase) [18].

For this reason, this study aimed to develop a clinical-replicable cooling protocol of the NiTi alloy, allowing an increase of the cyclic fatigue resistance of the instruments, especially when used in severely curved canals.

Throughout this method, an ice gas spray was used to induce temperature drops in the NiTi alloy, being easily replicable in clinical situations. According to the manufacturer, the temperature at which the Endo-Frost (Roeko) gas exits the spray bottle is of -50°C. In a study that tested ice gas sprays, when Endo-Frost (Roeko, Langenau, Germany) was applied to a cotton swab, the temperature dropped to -28.0°C±11.2°C [19]. Thus, this is

Table 2. Values of increase in resistance (%) after cooling protocol, and mean and standard deviations after bending test (N.cm)

		X1 Blue	**Wave One Gold**	***P*-value**
Percentage of resistance increase	**Median**	127.5%	92.15%	0.5787
	Percentile 25	13.58%	65.70%	
	Percentile 75	289.8%	134.8%	
Bending		1.33 (0.24)	1.37 (0.15)	0.6729

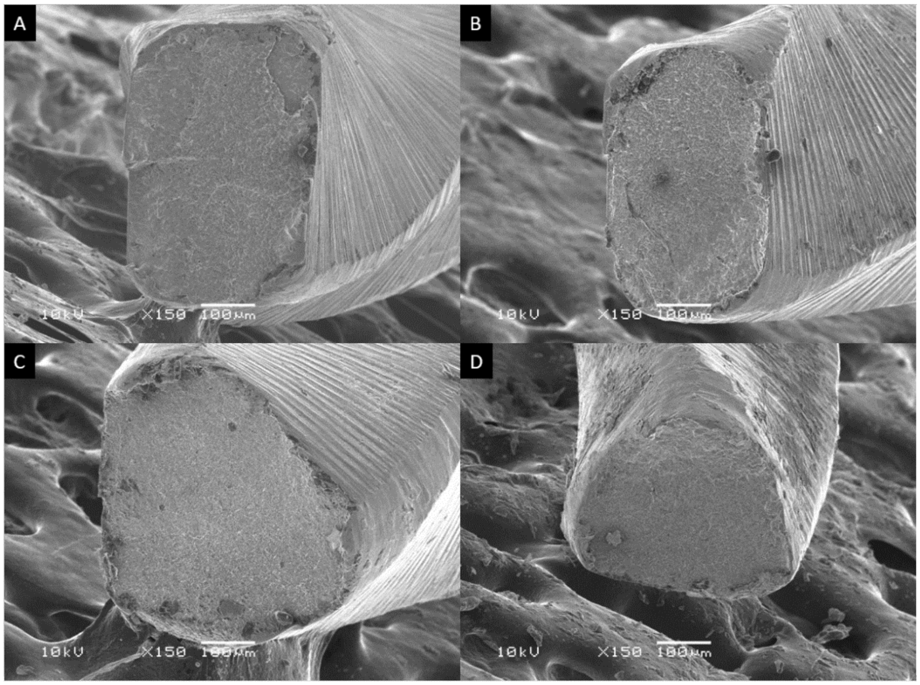

Figure 1. Scanning electron microscopy images of the fractured surfaces of; *A)* Wave One Gold 25.07 at 36°C±1°C; *B)* After cooling protocol; C) X1 Blue 25.06 at 36°±1°C; D) After cooling protocol

assumed to be the temperature of the instruments, after ice gas spraying. However, it is not possible to determine the exact temperature presented by the instruments after cooling, thus being a limitation of the present study.

Another limitation of this study is that when the cooled instrument makes contact with the heated water, the temperature gradually increases, therefore, the exact temperature in which the instruments were activated for the cyclic fatigue test is also not possible to be determined. However, despite the described limitations, the results of this study showed that the investigated cooling protocol increased the resistance to cyclic fatigue of the tested NiTi instruments, thus, the null hypothesis was rejected.

To discuss the present results is a difficult task, since the tested instruments vary in many aspects, such as the heat treatment, cross-sectional design and taper size, which directly influence the instruments' mechanical properties. Regarding the effects of the variations of temperatures in the instruments alloy, our results are in accordance to previous studies [6, 11-14] in which the resistance to cyclic fatigue increased, when decreasing temperature. Controversially, another study presented that the immersion of Reciproc Blue instruments in higher temperatures (60ºC) for 5 minutes positively affected its resistance to cyclic fatigue [20]. This contradiction can be explained by the variation of the adopted methodologies. While in our study the tests were performed in 36°±1°C all the time, in this study the instruments were immersed during 5 minutes in heated liquids but the tests were performed at room temperature. Another factor is that the temperature in which Reciproc Blue instruments had greater results was of 60ºC, a condition that does not reflect the temperature in which the instrument operates during clinical practice.

In other study [21] that tested the ProTaper Universal (Dentsply Maillefer, Ballaigues, Switzerland) and ProTaper Gold (Dentsply Maillefer) in different temperatures, it was reported that the latter did not suffer significant differences in the cyclic fatigue resistance in thermal variations between temperatures of 20°±1°C and 35°±1°C. However, in this study there was no water immersion of the instrument during cyclic fatigue tests.

Throughout this study we were cautious to adjust the instrument to the artificial canal so that it could uniformly reciprocate. It has been described that when the instrument freely rotates inside the device, it can potentially follow a more severe curvature or not, depending on the stiffness of the instrument, which explains the wide variety of related fatigues across reported findings [3].

There is an increasing number of studies comparing resistance to cyclic fatigue at body temperature [6, 11, 13, 14, 20-25]. Since the cyclic fatigue resistance of instruments is affected by temperature variations, to test such instruments at temperature near to those found in the patient's body can present more reliable results [12, 26].

In this study, two instruments presenting different heat treatments, Gold and Blue, were tested in order to verify if the effects would vary according the instruments NiTi alloy. According to the presented results, X1 Blue instruments have not demonstrated significantly greater resistance to cyclic fatigue when compared to Wave One Gold instruments in the body temperature group. These results are consistent with those obtained by a previous study [6] that did not observe differences between X1B and WOG cyclic fatigue resistance in body temperature.

Additionally, a percentage point increase in resistance was evidenced in this study when the instrument underwent the cooling process in both groups (Table 2). Our results are aligned with several other studies that also observed an increase in resistance at lower temperatures [6, 11-14].

In this study, although testing a clinical-replicable protocol, the static cyclic fatigue model was selected. This model allows a better comprehension of the influence of specific factors on the instrument's performance [16], such as the influence of the changes caused by the temperature variations after the ice gas spraying. Also, the static model minimizes biases like speed and amplitude movements, possible to be reproduced in the dynamic model, but that are operator-dependent in clinical situations [27].

Complementarily, the flexibility of the instruments was verified through the bending test. To test the bending capacity of NiTi instruments is important since this property is related to the instrument's performance in preparing curved root canals [28]. In the current study, there was no difference in the bending resistance between X1 and WOG instruments. A possible explanation would be that these instruments present a greater flexibility due to their heat treatments, which allows reduction of the load exerted on the instrument's cutting blades in a curved canal, reducing the tension on the instrument and the risk of fracture, despite the differences in the taper size of the instruments. A previous study [29] that tested Gold and Blue alloys had provided similar results. Additionally, the absence of differences between the tested instruments may be related to their cross-section design and to the capacity to distribute mechanical stress. A previous study had shown that the cross-section design presents a greater impact than taper or size of the instrument on the stress development, and also reported that instruments presenting triangular convex and parallelogram cross-section design can distribute stress in similar patterns [30], possibly explaining the present results.

The SEM analysis of the instruments subjected to the cyclic fatigue test demonstrated similar characteristics for both groups. SEM mainly showed crack initiation areas, propagating over a single or in multiple planes and the presence of numerous dimples with varied forms, agreeing with a previous study [16].

The purpose of this study was to investigate a cooling protocol to be safely reproduced in clinical situations in order to increase the instrument's cyclic fatigue resistance, especially when used in severely curved canals. Throughout the presented results, it is possible to conclude that this cooling protocol presented benefits regarding cyclic fatigue on the tested instruments. However, other aspects related to this cooling protocol, such as the shaping ability and life span of the cooled instruments, should be verified previously to the clinical insertion of this protocol. Also, in this study, only two instruments were tested, therefore, more studies evaluating this protocol are necessary.

Conclusion

Within the limitations of this study, it is possible to conclude that X1 Blue #25.06 and WaveOne Gold #25.07 instruments presented similar results on their cyclic fatigue resistance. However, the investigated clinical-replicable cooling protocol improved the cyclic fatigue resistance of the tested instruments, with X1 Blue #25.06 presenting a greater cyclic fatigue resistance after the cooling protocol. And finally, both instruments presented a similar bending capacity.

Conflict of Interest: 'None declared'.

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