Abstract

Aim: The study was aimed to evaluate the effect of 5.25% NaOCl and 17% ethylenediaminetetraacetic acid (EDTA) Solutions on Surface Topography of ESX, TruShape, and ProTaper Gold nickel-titanium (NiTi) Rotary Instruments using Atomic Force Microscope (AFM).

Materials and Methodology: A total of 27 each of three commercially available brands of endodontic NiTi instruments were analyzed, which were divided into three groups as follows: Group I: ESX (Brasseler, USA) Group II: Tru shape (Dentsply Tulsa, USA) Group III: Pro Taper Gold (Dentsply Tulsa, USA). These files were further divided into three subgroups containing nine files each, no immersion, immersion in 5.25% NaOCl for 5 min, and immersion in 17% EDTA for 5 min. Surface areas along 3 mm sections at the tip of the files (perfect squares of 10 μm × 10 μm) were analyzed using AFM operating in contact mode under ambient conditions.

Statistical Analysis: Data were analyzed using two-way ANOVA and pairwise comparison of three main groups with respect to Ra and RMS by Tukey’s multiple posthoc procedures.

Results: Three-dimensional AFM images of the surface of all the rotary NiTi instruments, including new and those immersed in 5.25% NaOCl and 17% EDTA solutions, revealed topographic irregularities at the nanometric scale. Ra and RMS values of instruments treated with 5.25% NaOCl and 17% EDTA solutions were statistically higher than that of the new ones (P < 0.05).

Conclusion: Using AFM for analysis indicated that short-term contact between 5.25% NaOCl and 17% EDTA solutions and NiTi instruments caused alterations in the topography of instruments.

Keywords: Atomic force microscope; nickel-titanium rotary instruments; sodium hypochlorite; surface roughness

INTRODUCTION

Over the past few years, mechanical instrumentation of root canals has been carried out conventionally with the help of carbon steel instruments, which were later replaced by stainless steel instruments due to their poor corrosion resistance. However, stainless steel instruments have advantages such as better cutting action and superior corrosion resistance, but its modulus of elasticity was found to be high, resulting in the lack of flexibility, which led to the advent of rotary nickel-titanium instruments (NiTi).[1] In recent years, the popularity of NiTi instruments has increased because of their superior elasticity and resistance to torsional fracture compared to stainless steel hand files. Despite these advantages of NiTi instruments, they were prone to fracture during their clinical performance.[2,3] Manufacturers of rotary NiTi...
instruments generally claim a single use, but there are many studies that suggest and support the multiple uses of rotary NiTi instruments.\[^{12}\]

The NiTi alloys are generally made up of 56\% by wt nickel and 44\% by wt titanium. Some amount of nickel generally disappears from the surface due to its unstable oxide resulting in the highly irregular long island like structures indicating their selective dissolution. This tendency during mechanical usage and autoclaving cycles results in the unequal dissolution of nickel and titanium, thus leading to an easy predilection for surface irregularities.\[^{14}\]

Due to the pseudoelastic nature of these alloys, NiTi instruments are manufactured by the machining process rather than twisting like stainless steel hand files. Grinding of rotary NiTi instruments during the manufacturing process often results in a surface that is irregular, stressed, and showing cracks, milling grooves, pits, and fissures and regions of metal rollover (Alapati et al. 2005).\[^{9}\] These surface irregularities may act as a nidus for the crack propagation whenever there is an increased fatigue load on the instrument (Kuhn et al. 2001).\[^{10}\] Usually, endodontic instruments during endodontic treatment are used in the presence of lubricants or irrigants, which are mainly chlorine-containing compounds, which tend to cause pitting corrosion due to the localized attack on passive metals.\[^{6,7}\] Although direct evidence regarding the failure due to pitting corrosion and manufacturing defects were not reported, these can alter the fracture mechanisms during their usage leading to instrument separation.\[^{7,8}\]

The field emission Scanning Electron Microscopic (SEM) images and Energy Dispersive X-Ray Spectroscopy clearly revealed the presence of non-corroded zones and of corroded areas where the localized attack had created pitting and cracks close to the fracture surface on the NiTi instruments. The phenomenon observed may be attributed to galvanic corrosion, also known as “dissimilar metal corrosion.”\[^{11}\]

Hence, various developments were made in the metallurgical aspects of NiTi instruments, to improve the properties, which made them more resistant to corrosion and fractures. Therefore, various surface treatment techniques have been tried to create smoother surfaces to enhance the wear resistance of rotary NiTi instruments.\[^{10}\]

To date, the surfaces of NiTi rotary instruments have been analyzed using a variety of different techniques. SEM was widely used to evaluate the surface topographic characteristics of different types of materials. Recently, the atomic force microscope (AFM) for imaging the three-dimensional surfaces of different materials has become increasingly popular and has been recommended as a valuable research tool for investigating the topography of various endodontic instruments.\[^{3,8}\] This study involves the testing of three new commercially available NiTi instruments, i.e., ESX (Brasseler, USA), TruShape (TS) (Dentsply Tulsa, USA) and ProTaper Gold (PTG) (Dentsply Tulsa, USA), which were recently introduced with improved mechanical properties and to date, there appear to be no studies that evaluated the effect of 5.25% NaOCl and 17% ethylenediaminetetraacetic acid (EDTA) solutions on the topography of these files. Hence, the aim of this study was to evaluate and compare the effects of 5.25% NaOCl and 17% EDTA solutions on the surface nanostructure of three rotary NiTi instruments with different manufacturing processes using AFM analysis.

**MATERIALS AND METHODOLOGY**

This study was conducted in the Department of Conservative Dentistry and Endodontics, Sibar Institute of Dental Sciences, Guntur. The purpose of this study was to evaluate the effect of 5.25% NaOCl and 17% EDTA on the surface of three different rotary NiTi instruments. A total of 81 NiTi rotary endodontic files, of 27 each of ESX (Brasseler, USA), TS (Dentsply Tulsa, USA) and PTG (Dentsply Tulsa, USA) rotary instruments were used for this study.

These files were divided into three groups as follows:

- **Group I**: ESX (Brasseler, USA) rotary NiTi instruments
- **Group II**: TS (Dentsply Tulsa, USA) rotary NiTi instruments
- **Group III**: ProTaper Gold (Dentsply Tulsa, USA) rotary NiTi instruments.

These files were further divided into three subgroups containing nine files each: (a) new un-immersed instruments were kept as controls, (b) instruments immersed in 5.25% NaOCl for 5 min, and (c) instruments immersed in 17% EDTA for 5 min.

With the fine orthodontic pliers, the active part of these instruments, i.e., (apical 3 mm) was separated. Nine active parts from each group were kept as controls, the other nine active parts were immersed in 5.25% NaOCl for 5 min, and another nine active parts were immersed in 17% EDTA for 5 min, respectively. After the stipulated duration of immersion, these samples were dried, and then, un-immersed and immersed instruments were attached to a silicon wafer with the help of rapid setting cyanoacrylate glue.

**Image analysis**

These samples were subjected to atomic force microscopic (3D standalone, Asylum Research, IIT Mumbai) analysis. For each sample, the surfaces were analyzed on a 3-mm section starting at the tip of the file. The AFM images were recorded in contact mode operation under ambient conditions. Atomic force
microscopy probes (curvature radius <20 nm) mounted on cantilevers (250 μm), with a spring constant of 0.1 Nm², were used. Scanned areas were perfect squares (10 μm × 10 μm). This was possible by visualizing a monitor displaying an image of the specimen at 500x magnification, providing data on the topography of the analyzed area in nanometers (nm). Three-dimensional images (256 lines × 256 lines) were processed with C-AFM (3D standalone, Asylum Research, IIT Mumbai). For comparison, Roughness (Ra) and Root Mean Square (RMS) values were chosen to investigate the surface features of endodontic files. The Ra and RMS values of the scanned surface profiles were recorded [Figure 1]. An increase in Ra and RMS values indicates alterations of NiTi instruments surface caused by the 5.25% NaOCl and 17% EDTA.

Statistical analysis
Comparison of three main groups (ESX, TS, PTG) and three subgroups (Control, NaOCl, EDTA) with respect to Roughness values and RMS values was made by two-way ANOVA, and pairwise comparison of three main groups (ESX, TS, PTG) and three subgroups (control, NaOCl, EDTA) with respect to RMS values and roughness average (Ra) values by Tukey’s multiple posthoc procedures. The level of statistical significance was set at P < 0.05.

RESULTS
There was a significant difference between the main groups and between the subgroups. Controls of ESX, TS, and PTG instruments had significantly lower Ra and RMS values when compared to instruments immersed in 5.25% NaOCl and 17% EDTA, where values of ESX were highest followed by PTG and least for TS. For instruments immersed in 5.25% NaOCl for 5 min, Ra and RMS values were highest for TS, followed by PTG and least for ESX. For instruments immersed in 17% EDTA for 5 min, RA and RMS values were highest for ESX, followed by PTG and least for TS [Figures 1-3].

DISCUSSION
Over the years, various attempts to enhance the surface characteristics of NiTi instruments were made by manufacturers to minimize or eliminate inherent defects, increase surface hardness, flexibility, and to improve resistance to cyclic fatigue, corrosion, and cutting efficiency of endodontic instruments. Some of these techniques, such as plasma immersion ion implantation, thermal nitridation, cryogenic treatment, and electropolishing, have improved the properties of NiTi instruments.[10]

Till date, a variety of different techniques have been used to analyze the surface topographic proprieties, such as SEM However, SEM produces two-dimensional images of NiTi rotary instruments but cannot directly provide any quantitative data. On the contrary, AFM provides both qualitative and quantitative information by scanning the surface topography.[3] Therefore, in the present study, topographic surface changes in all NiTi rotary instruments were evaluated using AFM that were immersed in NaOCl and EDTA solution. AFM records the data of samples in digital form as sets of x, y, and z values. These sets can be analyzed with digital software to give all the data pertaining to the examined surface in quantitative form by using vertical topographic parameters. Ra and RMS values of instrument surfaces were analyzed. These values are also known as the quadratic mean, which is a statistical measurement of the magnitude of a variable quantity in nanometers (nm), of the surface topography and the area in square micrometers (μm²).[11]

In the present study, both of the solutions, i.e., 5.25% NaOCl and 17% EDTA caused significant deterioration of all instrument surfaces resulting in an increase in RMS and Ra values compared to their controls. The surface irregularities of the controls may be due to the industrial process (i.e., manufacturing) with the highest Ra and RMS values recorded for ESX, followed by PTG and lowest for TS. These nanostructure alterations may have a considerable impact on resistance to fracture of endodontic files and probably make them more prone to corrosion.[8]

The results for instruments immersed in 5.25% NaOCl for 5 min, Ra and RMS values were highest for TS followed by PTG and lowest for ESX. According to Topuz et al. (2008) it was shown that immersion in 5.25% NaOCl for 5 min caused localized surface pitting and cracks that modify the integrity and resistance to fracture of NiTi instruments.[12]
In a study, conducted by Saglam et al. Protaper Rotary NiTi Instruments treated with 2.5% NaOCl demonstrated less surface alterations compared to the group treated with 5% NaOCl and concluded that the lower concentration demonstrated less surface alterations, and the use of 2.5% NaOCl may be recommended to avoid weakening of the ProTaper file. Sodium Hypochlorite is corrosive to metals involving the selective removal of nickel from the surface, creating micro pitting. It was evident that microstructural defects can lead to areas of stress concentration and crack formation, that weakens the structure of the instrument.

According to a study conducted by Pedulla et al. 2014, 5.25% NaOCl did not influence the cyclic fatigue of all instruments tested, instead 17% EDTA reduced the cyclic fatigue resistance of files after 3 min immersion. According to Haikel et al., neither the mechanical properties of NiTi instruments were affected by NaOCl nor was the cutting efficiency.

PTG files (Dentsply Tulsa, USA) were developed using gold Wire technology, which is an advanced metallurgical process that makes files significantly more flexible and resistant to cyclic fatigue. In the present study, PTG showed an increase in Ra and RMS values when exposed to irrigating solutions than controls.

In this study, the ESX file, which was evaluated, had electropolished finish surface. Electropolishing (EP) is a process in which surface titanium of NiTi instruments is oxidized to titanium dioxide (TiO2), which protects the underlying material from further corrosion. The electro-polished finish of ESX files did not inhibit the development of micro-fractures on its surface and showed increased RMS and Ra values when immersed in irrigating solutions when compared to the control group. According to an SEM study done by Herold et al. Electropolishing did not inhibit the development of microfractures in endosequence rotary instruments.

There is not much literature available regarding surface finish and topography of TS files. The lower RMS and Ra values for TS immersed in EDTA could be due to smoother surface finish compared to ESX and PTG which could be explained by Reinhard et al. 1992 as that the larger molecules of EDTA, have greater difficulty in concentrating and orienting within small pits to increase the acidity to adequate values that could trigger corrosion.

The possible limitation of the present study could be the variable time of contact with irrigant, which is more compared to the clinical scenario during cleaning and shaping. On the other hand, we have evaluated topography in a stagnant irrigant solution, but while performing cleaning and shaping, the instrument will be continuously rotating with irrigant solution surrounding it. This situation may result in a different alteration of surface topography and it has to be evaluated in further studies.

**CONCLUSION**

Within the limitations of this study, short-term contact of 5.25% NaOCl and 17% EDTA solutions with ESX, TS, and PTG Rotary Files showed an increase in their surface roughness values indicating surface deterioration. AFM proved to be a practical method to directly characterize the endodontic file surface, allowing enhanced knowledge of the manufacturing quality of NiTi rotary instruments.
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Conflicts of interest
There are no conflicts of interest.

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