Effect of Different Cleaning and Sterilization Methods on the Surface Morphology of Mini-implants

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The objective of this study was to analyse morphological and surface topography variations of two types of mini-implants after using different chemical and physical cleaning methods and autoclaved sterilization. One hundred mini-implants from two different manufacturers were used in this study. The mini-implants from each manufacturer were divided in five groups, each consisting of ten samples: G0 new, unused, G1 ultrasonically cleaned, G2 chemically cleaned, G3 sandblasted, G4 cleaned with distilled water. SEM analyses of the mini-implants were performed. Only procedures used in samples from group G2 and G3 removed the tissue remains from the mini-implants surface.

Keywords: mini-implants, scanning electron microscopy, sterilization, morphology

The temporary anchorage devices, also known as mini-implants became popular in clinical orthodontics, due to their numerous advantages: atraumatic insertion, different applications, immediate loading and small dimensions which results in minimal anatomical limitations [1]. Other advantages include their high success rate and versatility which results in minimal anatomical limitations [1]. During the orthodontic treatment, relocation of the biomechanics in developing effective orthodontic advantages include their high success rate and versatility which results in minimal anatomical limitations [1]. Other applications, immediate loading and small dimensions of the mini-implants is sometimes needed, especially because mini-implants inserted between the tooth roots may interfere with the tooth movement [2]. Re-using the temporary implant devices might be considered both for economic reasons and environmental conservation, but it involves ethical considerations. Some authors [3, 4] agreed re-using these devices, in the same patient, after sterilization. However, sterilization may contribute to surface topography alteration that changes the mechanical properties [5-7]. Eliades, Zinelis, Papadopoulos and Eliades [8] reported that used titanium alloy mini-implants have both surface and morphological modifications. Worn surfaces and scratch marks were observed by Mattos, Ruellas and Elias [9] on the autoclaved and retrieved implants.

Different cleaning and sterilization methods are available, including steam autoclaving, gamma irradiation, chemical cleaning and sandblasting [10]. Therefore, the objective of this study was to analyse morphological and surface topography variations of two commercially available mini-implants after using different chemical and physical pre-sterilization cleaning methods and autoclaved sterilization.

Experimental part

A total number of 100 mini-implants from two different manufacturers (Link from MIS™, MIS Implants Distribution, Bucharest and Yesanchor from Orlus™, Seoul, Korea) were tested in this study. These two manufacturers were chosen because their mini-implants are the most used and popular in Romania. On the other hand, one of the mini-implants was cylindrical and the other conical, both having the same dimension 1.6 x 8 mm. The implants from each manufacturer were randomly divided in five groups from G0 to G4. The G0 group included ten new, unused mini-implants as control group. The G1 group consisted of ten mini-implants inserted in pig mandibular bone and removed to reproduce the clinical conditions of insertion and removal from jaw bones. The removed mini-implants were subjected to ultrasonic cleaning, at a frequency of 40 kHz and a temperature of 25°C in an ultrasonic washer (Digital Ultrasonic Cleaner, CD 4820, Codyson) completely immersed in detergent, in order to remove the organic debris from their surface. After the 20 min cleaning cycle in detergent solution, the mini-implants were removed, rinsed with distilled water and cleaned ultrasonically once again for 15 min in distilled water. This procedure was followed by autoclave sterilization at 121°C, at a pressure of 1.03 bar for 20 min (Vakuclav 31B, Melag™, Berlin), according to the recommendations of the manufacturers. The G2 group underwent to the same insertion and removal protocol as group G1 followed by chemical cleaning. Chemical cleaning consisted of fully coverage with phosphoric (H3PO4) acid gel, 37% (Ultra-Etch, Ultradent) and then immersion in 1mL of the same acid for 10 min. The samples were irrigated, dried and

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immersed in 10 mL sodium hypochlorite 5.25% (NaOCl) for 30 min. After rinsing with distilled water, they were packed in sealed bags and sterilized in autoclave (same protocol as G1).

The G3 group underwent to the same insertion protocol as group G1 followed by ultrasonic cleaning in detergent solution for 8 min and rinsing with distilled water. Then, sandblasting was performed with Al₂O₃ particles with a dimension of 90µm, at a pressure of 4.14 bar, from a distance of 10 mm. The samples were cleaned, once again in ultrasonic bath for 20 min, followed by autoclave sterilization.

The G4 group consisted of ten mini-implants from each manufacturer with the same insertion and removal protocol of group G1 followed by rinsing with distilled water and autoclave sterilization.

Scanning electron microscopy (SEM) (JEOL 100, JEOL Ltd., Tokyo, Japan) of the mini-implants was performed at an acceleration voltage of 30 kV and a magnification of 30x-100x. Representative micrographs were taken from each sample group. SEM analyses were done at the head, neck, body and tip of the implants.

Results and discussions

SEM images of the new, unused mini-implants from group G0 are shown in figures 1a and 1b. On the surface of the Link mini-implants, noticeable uniform and unidirectional striations are more obvious, when compared with the Yesanchor mini-implants of the same category. These striations are likely to be the results of machining procedure. However, the surface of the new Yesanchor mini-implant presented more structural defects such as grooves.

The Link mini-implants from G1 group showed rough surface deposits on the tip of the device, visible at 50x magnification (fig. 2a).

Figure 2b shows the surface morphology at 50x magnification of the Yesanchor mini-implants from the G1 group. Very few organic tissue remains are visible and a smoother surface on the threads and scratch marks on the head were observed.

Figure 3a shows the surface morphology of the chemically cleaned and sterilised Link mini-implants (G2 group). The SEM image does not show corrosion signs and no residual tissue remains are obvious. The sharpness of the tip of the device is comparable with the unused mini-implants. On the other hand, pitting corrosion is obvious on the surface of the Yesanchor mini-implants of the same group (fig. 3b).

Sandblasting modified the surface topography of both type of mini-implants for the G3 group. Their surface appeared rougher but no tissue remains were detected (figure 4a). However, the threads of the Yesanchor mini-implants presented some defects such as grooves, probably due to reinsertion (fig. 4b).

The SEM images of the autoclaved mini-implants of the G4 group showed numerous surface deposits, especially on the outer border of the flutes (fig. 5a and 5b).

Re-using a mini-implant in the same patient is possible during different phases of an orthodontic treatment [3]. However, there are numerous problems regarding the
reused mini-implant's structural integrity and surface modifications. It is also important to assure effectiveness of the cleaning and sterilization methods, in order to avoid infections.

In our study, two different types (one cylindrical and one tapered) orthodontic mini-implants were subjected to several cleaning methods, followed by autoclaved sterilization. The G1 group of mini-implants were ultrasonically cleaned in order to determine whether or not this method completely decontaminates and cleans the mini-implants surface. Some studies have demonstrated [11-13] that sonication cannot be sufficient to remove the proteinaceous biofilm from the contaminated instruments. We can also state that sonication did not cleaned the implants from the G1 group. Surface deposits were more visible on the Link mini-implants. On the other hand, chemical cleaning removed the organic tissues from the surface of both type of mini-implants from the G3 group. This method was suggested by Noorollahian, Alavi and Monirifard [14]. In their study [14], the used mini-implants were immersed in 37% phosphoric acid for 10 min, followed by sodium hypochlorite 5.25% for 30 min to reduce the tissue debris to a comparable level to that of unused mini-implants. The phosphoric acid has the advantages of being easily available and at low risk during manipulation [14]. The sodium hypochlorite can dissolve organic tissues and it is also cheap [14]. Their properties include a corrosion potential, although Noorollahian et al. stated that neither sodium hypochlorite, nor phosphoric acid harm the titanium surface at room temperature [14]. However, we observed pitting corrosion on the surface of the Yesanchor mini-implants. Some authors [15, 21,22] stated that titanium alloys used to fabricate mini-implants are less resistant to corrosion due to the manufacturing process which creates surface defects. These surface defects represent discontinuities in the corrosion resistant passive titanium oxide layer. Insertion of the mini-implant in the oral environment determines the corrosion of the device. The corrosion on the studied implant surface from our study was not determined by the local conditions because the mini-implants were inserted and then immediately removed from the pig bone. It is more likely that the chemical factors modified the surface of the mini-implants.

Eliades et al. [16] found surface and morphologic structural modifications in intraorally used mini-implants. However, Mattos et al. [9] found no pores, cracks or corrosion on the retrieved mini-implants.

Several studies [17,18] have demonstrated that autoclaving alone does not completely decontaminate infected devices or instruments, especially if it is not cleaned before sterilization. The mini-implants from the G4 group, tested in our study, showed significant tissue remains, which might determine an inflammatory response and have deleterious effects on bone response. Surface roughness is an important factor in implants integration in bone. Hansson and Norton [19] demonstrated that surface texturing or treating the implant surface improve bone apposition. Surface roughness created by sandblasting also determines a better biological response [20]. On the other hand, the importance of removing loose Al₂O₃ particles from mini-implants surface was emphasized [20, 24, 25]. These remaining particles might disturb bone differentiation and deposition, so ultrasonic cleaning is indicated after sandblasting [20]. In our study, the sandblasted mini-implants from the G3 group had a significantly increased surface roughness, but no remaining organic deposits were observed.

Regarding the two types of mini-implants used in our study, one cylindrical and one tapered, it can be stated that no significantly differences were observed on the implant's surfaces during the cleaning and sterilization processes. On the other hand, the new, unused mini-implants had some differences due to the fabrication process. The Link mini-implants had noticeable uniform and unidirectional striations, while the Yesanchor mini-implants presented more structural defects such as grooves. In their study regarding different titanium alloy implants surfaces, Burmann, Ruschel, Vargas, De Verney and Kramer [23] stated that the orthodontic mini-implants exhibited significant differences in the design of their parts and surface irregularities were found on their surfaces.

Conclusions
Reusing mini-implants is only recommended if pre-sterilization cleaning and sterilization is properly done. Sandblasting with aluminum oxide and chemical cleaning with phosphoric acid and sodium hypochlorite followed by
autoclaved sterilization removed the tissue remains from the mini-implants surface. Ultrasonic cleaning followed by autoclave sterilization and only autoclave sterilization did not properly remove the organic debris from the mini-implants surface.

Acknowledgments: This work was supported by the University of Medicine and Pharmacy of Tîrgu Mures, Research Grant number 17800/13/22/12/2015.

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Manuscript received: 26.03.2017