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ORIGINAL ARTICLE

In Vitro Study of Tooth Surfaces After Interproximal Enamel Reduction: Extraoral Scanner and SEM Analysis

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ABSTRACT

Objective: To perform a morphological evaluation concerning the extent of interproximal enamel reduction (IPR) with different manual instruments in different types of teeth and a qualitative analysis of enamel surface characteristics at the contact point before and after IPR. **Material and Methods:** 40 freshly extracted, caries-free, and intact human teeth were used for the study (20 bicuspids and 20 incisors) and performed IPR just on the mesial surface. The morphological variation of contact point was evaluated by superimposed the stl file, obtained thanks to an extraoral scanner, at T0 and T1 for each tooth. Two types of strip were used, Intensiv Manual Ortho Strips Coarse/Medium and Steelcarbo Horico Strips. Teeth were then cut lengthwise, removed the most apical root portion and the mesial and distal halves were gilded and observed at different magnifications. **Results:** The morphological variation following stripping mainly depends on the extent of the stripping, while the diameter, the type of strip and the shape of the tooth itself do not appear to be relevant. The 500X and 1500X magnifications allowed to appreciate better the characteristics of the surface of the stripped enamel and the differences with the intact enamel. All teeth treated, independently from the kind of strip used, shows deep marks and grooves in the direction of stripping. In both cases, the enamel appears significantly damaged at great magnifications. **Conclusion:** Stripping always and inevitably leads to a change in the shape of the contact point and is directly correlated to the amount of stripping performed. The use of polishing after the removal of enamel interproximal is necessary in all cases.

Keywords: Dentition; Orthodontics; Malocclusion; Dental Arch.

Introduction

Stripping can be considered as the mechanical removal of interproximal enamel (IPR) with a reduction of the mesio-distal width of teeth. It was initially proposed by Ballard in 1944, who first thought that the discrepancies of tooth size could be solved by undergoing lower anterior teeth to this procedure [1]. The need of stripping was further reiterated by Bolton in his study on the association between disharmony of dental dimensions and malocclusion [2].

In 1970, Peck and Peck [3] developed an index indicating the relationship between dental morphology and alignment, facilitating the treatment plan. It uses the ratio between mesio-distal and buccal-lingual size (MD/VL ratio) to indicate the alignment of lower incisors. They suggest that the ideal MD/VL ratio to achieve good alignment should be 88-92% for central incisors and 90-95% for lateral incisors.

Nowadays, the use of IPR to recover space has significantly increased and represents a common clinical procedure in orthodontic therapy. The need for extraction in case of crowding might be eliminated by interdental stripping [4,5]. Reshaping the proximal contacts, solving Bolton discrepancy problems, treating mild or moderate crowding, and stabilizing the dental arch are the main clinical indications to interproximal enamel reduction [6,7], especially when lingual appliances are used and labial inclination of incisor could be not easily obtainable [8].

Even though many beneficial outcomes have been well documented, the effects on the enamel surfaces have also been an issue of debate, and the ultramorphology, surface roughness, and microhardness after various stripping methods have already been studied [9-11].

The most common IPR technique investigated are air-rotor stripping (ARS), diamond discs, manual or mechanical finishing abrasive strips. The contact point must remain 4.5-5 mm from the bone crest to avoid the worsening of black triangles or the increase of periodontal pocket, if closer [12].

There are different statements to be found in the literature regarding the amount of enamel reduction. For example, Chudasama and Sheridan [13] reported that only 1 mm of enamel should be removed from each posterior contact point. This corresponds to 0.5 mm per interproximal surface. Since enamel is thinner at upper lateral and lower incisors, only 0.5 mm of enamel should be removed at this level.

Fillion, for example, recommended that reduction not exceed 0.3 mm of the surface in the upper incisors, 0.6 mm in upper premolars and molars, 0.2 mm in the lower incisors, and 0.6 mm in the lower premolars and molars [14].

An important aspect to keep in mind is no relationship between tooth width and enamel thickness. Therefore, larger teeth do not necessarily have one greater amount of enamel. Furthermore, there is no correlation between shape and enamel thickness [15]. Therefore, although it's well known that IPR determines a change in the shape of the overall tooth itself, its analysis must be studied in more detail [10,16,17].

Therefore, the current work aims to perform a morphological evaluation concerning the extent of IPR with different manual instruments in different types of extracted teeth. Furthermore, a qualitative analysis of enamel surfaces at the contact point before and after IPR was performed.

Material and Methods

Study Design and Sample

In this in vitro study, 40 freshly extracted, caries-free, intact human teeth were used: 20 bicuspids and 20 incisors. They were all divided into eight series, of 5 teeth each. Therefore, there are four incisors series

(called 1I, 2I, 3I, 4I) and four bicuspid series (called 1P, 2P, 3P, 4P). Within each, every single tooth was indicated with a letter (a, b, c, d, e).

Sample Preparation

The mesial and distal faces of each tooth, together with the most prominent mesial and distal points, were identified and marked. Furthermore, 3 reference points were created in the buccal surface with a small ball-shaped diamond bur and then superimposed the .stl files at T0 and T1. At T1, teeth were all stripped at the distal surface until the amount of stripping planned was reached: enamel reduction was performed by wetting teeth with saline to simulate the moist condition of oral cavity.

Two types of strip were used: Intensiv Manual Ortho Strips Coarse/Medium (Intensiv SA, Montagnola, Switzerland) and Steelcarbo Horico Strips (Horico Dental, Berlin, Germany) to compare strips designed specifically for the IPR execution and generic strips, which can also be used for this purpose. The Intensiv Manual Ortho Strips Coarse/Medium with grain size 40/80 microns were chosen, using just the 80 microns grain. Instead, Steelcarbo Horico strips of medium blue grain (80-100 microns) were used because they were more like the other strips. They are all abrasive on just one side and have a height of 4 mm. It was performed according to Fillion [14], who recommended removing less than 0.3 mm from upper incisors, 0.6 mm from molars and bicuspids and less than 0.3 from lower incisors.

Morphological Teeth Evaluation

Superimposition of .stl files was performed using 3shape Ortho Analyzer Software (3Shape, Copenhagen, Denmark) at T0 and T1, using 3 landmarks made on each tooth's buccal surface ensure greater accuracy. Two different operators observed and evaluated the overlaps relating to all teeth. Thanks to a "dynamic" evaluation, a score on shape's change according to the morphological scale (Table 1) was given. Regarding incisors, a preliminary assessment was also made about the shape (triangular, rectangular, and ovoid). Finally, the cross-section of each tooth was carried out at the equator level to precisely measure the amount of IPR performed.

Table 1. Scale of morphological evaluation of contact points.

Alteration of Enamel Surface: Quantitative Scale	
Grade 0	No Changes
Grade 1	Little Shape Change
Grade 2	Medium Shape Change
Grade 3	High Shape Change

Qualitative Analysis of Enamel Surface

One tooth was randomly selected for each group (1Ib, 1Pd, 2Ib, 2Pd, 3Ic, 3Pb, 4Ib, 4Pd) to be observed at scanning electron microscope – Carl Zeiss EVO 40 (Carl Zeiss AG, Oberkochen, Germany). All teeth were cut in half lengthwise, removed the most apical root portion and the mesial and distal halves were gilded and observed at different magnifications (50X, 500X, 1500X) (Figure 1).

Two different operators evaluated the quality and regularity of enamel surface. Differences between the intact mesial face and the stripped distal face were observed. In particular, they independently observed the photos and expressed their judgement on the superficial enamel alteration at 500X magnification using the above qualitative alteration scale (Table 2).

Figure 1. All samples prepared before SEM observation.

Table 2. Scale of qualitative evaluation of enamel surface.

Alteration of Enamel Surface: Qualitative Scale	
Grade 0	Smooth Surface: No Scratches
Grade 1	Acceptable Surface: Presence of Scratches
Grade 2	Rough Surface: Spaced Deep Furrows
Grade 3	Rough Surface: Broad, Deep and Irregular Furrows

Long-term stability is enhanced by IPR of the contact point to transform it in a surface contact, moving it in a more gingival position. This is particularly important at incisors level, where long-term stability is more complex to reach. Therefore, the contact surface obtained after IPR was measured only on incisors. It was evaluated at T1 to see how it changes in relation to different IPR quantity (Figure 2). The contact surface (rhomboid) is equal to major diagonal x minor diagonal / 2.

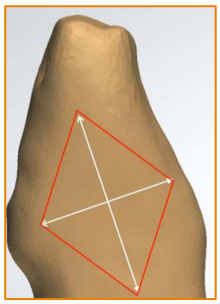

Figure 2. Rhomboid surface contact. The Greater diagonal was related to the occlusion-gingival width and the smaller diagonal to the vestibular-lingual portion.

Statistical Analysis

Two independent operators, based on the observations of the photos obtained by SEM, have expressed a judgment on the superficial alteration of the enamel ad 500X magnification using the qualitative scale reported previously (Table 2).

The retrospectively minimum effect size (f2) detectable for the regression analyses, taking into account the number of observations (n=20), the type I error level (α=0.05) and the usual statistical power threshold (1-β=0.8) is:

1. 0.938 for Incisors (2,5) models (5 independent variables);
2. 0.698 for Incisors (1,4) and bicuspid models (3 independent variables).

The regression model multivariate (OLS) has the objective of analyze if a change in the shape of the tooth contact point depends on the diameter of the same before stripping, the extent of the stripping, the type of stripe and the shape of the tooth for incisors (triangular, oval and rectangular).

Results

The stripping performed was analyzed using the 3shape Ortho analyzer software and the measurements obtained were confirmed by calculating the T0 and T1 diameter differences using the micrometer. Generally, the average quantity of IPR performed on incisors appears to be consistent with what was planned, except for 4I group, which exceeded the programmed range of 0.024 mm. Instead, the average IPR performed on bicuspids is consistent with what was planned.

Morphological Teeth Evaluation

The dynamic evaluation of file superimposition at T0 and T1 of each tooth was performed by two operators considering all tooth surfaces (mesial, distal, occlusal, vestibular and lingual). The morphological variations were expressed according to the scale reported in Table 1. All results were then collected and for incisors, the tooth shape was considered, as we see from Table 3 (T: triangular, R: rectangular, O: Oval).

Furthermore, from the superimposition analysis, the area subjected to IPR has an occlusion-gingival extension wider than the height of the strip used. This means that the amount of enamel removed on the distal surface of each tooth shows significant differences whether one moves occlusal or gingival to the point where the micrometer was positioned.

Table 3. Morphological variations for incisors.

Incisors	Operator 1	Operator 2
1Ia	T2	T2
1Ib	T1	T1
1Ic	T2	T1
1Id	R1	R2
1Ie	R0	R0
2Ia	T1	T2
2Ib	O1	O1
2Ic	O1	O2
2Id	O2	O1
2Ie	O1	O2
3Ia	R1	R2
3Ib	T2	T2
3Ic	T2	T2
3Id	T3	T2
3Ie	T2	T3
4Ia	T3	T3
4Ib	T1	T2
4Ic	R1	R1
4Id	R2	R2
4Ie	R1	R1

Qualitative Analysis of Enamel Surface

On SEM observation (50X, 500X, 1500X magnifications) the intact enamel of the mesial surface appears quite smooth, except for slight furrows related to function, chewing and brushing (Figure 3).

If the distal surface underwent an interproximal reduction is considered. Images at 50X magnification effectively show the flattening of the stripped surface and the increase in roughness both in incisors and premolars (Figures 4 and 5).

The 500X and 1500X magnifications allowed to better appreciate the characteristics of the surface of the stripped enamel and the differences with the intact enamel. In addition, the transition areas between intact enamel and treated enamel can be clearly visualized at all magnifications considered (Figure 6).

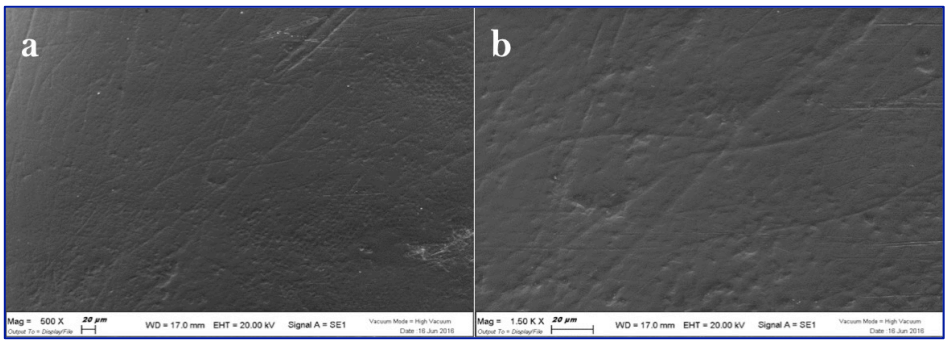

Figure 3. (a) Intact enamel at 500X; (b) Intact enamel at 1500X.

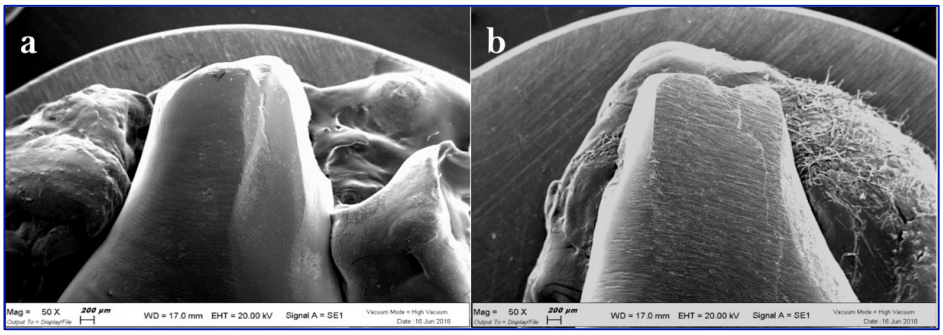

Figure 4. (a) Mesial surface of an incisor; (b) Distal surface of the same incisor (where we performed IPR) at 50X.

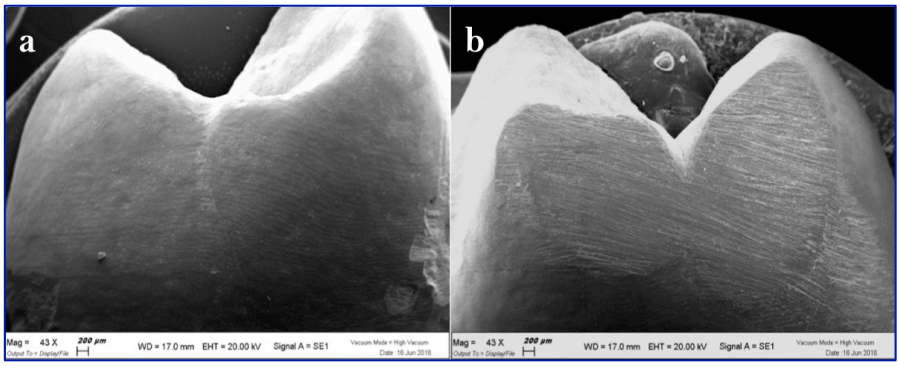

Figure 5. (a) Mesial surface of a bicuspid; (b) Distal surface of the same bicuspid (where we performed IPR) at 50X.

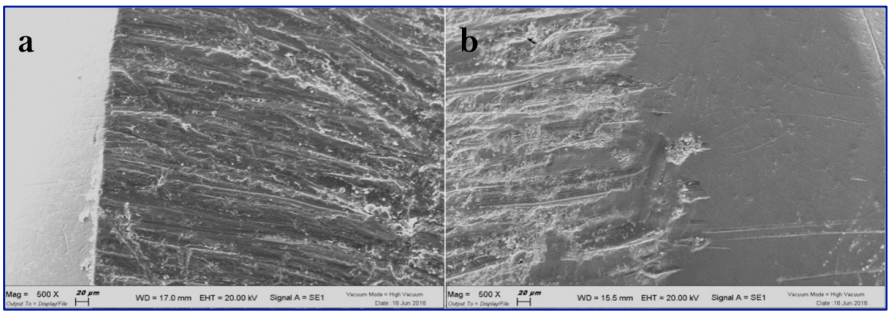

Figure 6. (a) Transition zone between intact enamel and treated with Horico at 500X magnification; (b) Transition zone between intact enamel and treated with Ortho Strip at 500X magnification.

All teeth treated, independently from the kind of strip used, shows deep marks and grooves in the direction of stripping. In both cases, the enamel appears significantly damaged at great magnifications (Figures 7 and 8).

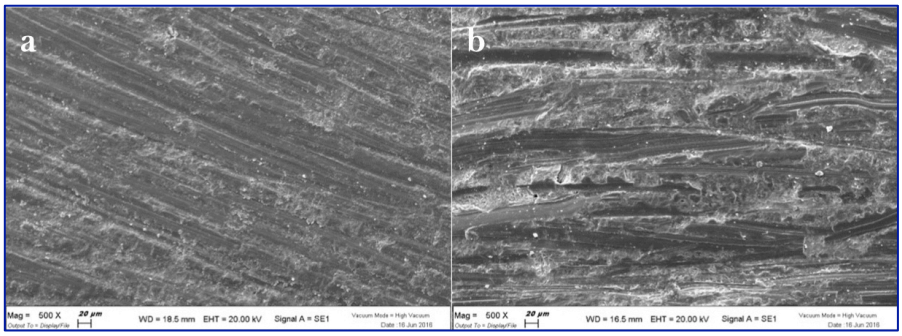

Figure 7. (a) Enamel treated with Horico at 500X; (b) Enamel treated with Ortho Strip at 500X.

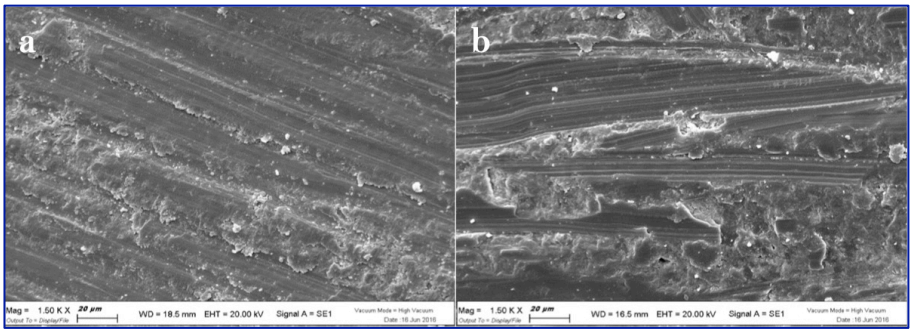

Figure 8. (a) Enamel treated with Horico at 1500X; (b) Enamel treated with Ortho Strip at 1500X.

The variation of the enamel surface is greater with the Ortho Strip Intensive rather than with Horico (Table 4). With both types of strips, a correlation between the entity of stripping and the degree of surface enamel alteration was not recorded.

Table 4. Descriptive analysis.

Series	Strip	Quantity of IPR		Morphological Alteration		Roughness Alteration	
		Mean (mm)	SD	Operator 1	Operator 2	Operator 1	Operator 2
1I	Horico	0.154	0.027	1.2	1.2	2	2
2I	Horico	0.162	0.053	1.2	1.6	3	2
3I	Ortho Strip Intensiv	0.272	0.065	2.0	2.2	3	3
4I	Ortho Strip Intensiv	0.324	0.121	1.6	1.8	3	3
1P	Horico	0.352	0.033	2.0	1.2	3	3
2P	Horico	0.360	0.034	2.0	2.4	3	3
3P	Ortho Strip Intensiv	0.470	0.084	2.6	2.8	2	2
4P	Ortho Strip Intensiv	0.420	0.041	2.0	2.2	3	3

The contact surface area does not seem to grow in a directly proportional manner as the extent of stripping increases (in the incisors). However, if we consider the distinction by shape (rectangular, triangular, oval incisors), it appears that a greater area of the contact zone is created in the rectangular teeth.

If the standard deviation is considered (Table 4), bicuspids show a lower variability compared to the incisors, with an average value equal to 0.048, while the incisors show a total standard deviation of approximately 0.066. What comes out is that the variations increase as the IPR increases. In particular, when

the IPR programmed for the incisors is between 0.1 and 0.2 mm, the average variation is 1.2 with Horico strip and 1.4 with Ortho Strip Intensiv. The change is small and slightly more pronounced for Ortho Strip Intensiv. While when the IPR range programmed for incisors is greater, for example, 0.2-0.3 mm, the average morphological change is equal to 2.1 with Horico strip and 1.7 with Ortho Strip Intensiv. Therefore, the opinions of the operators tend to highlight an average morphological variation greater for bicuspids treated with Horico strip (Figure 9). Although the operators are independent, no significant difference emerged from the comparison between the judge collected.

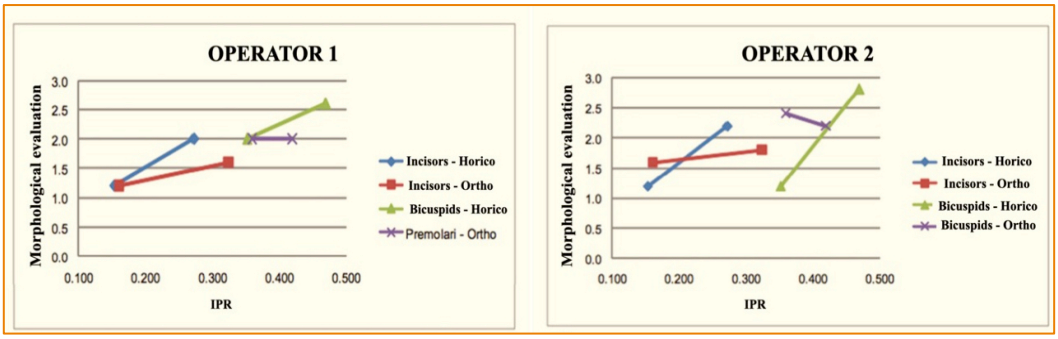

Figure 9. Relationship between IPR quantity and morphological change by type of tooth, strip and operator.

Regarding the variation in roughness, from the descriptive statistics reported in Table 4, IPR modifies the enamel surface causing the formation of furrows and marks. More generally, premolars show the greater roughness alteration. Both operators show that increasing bicuspid stripping with Horico, the roughness alteration seems to decrease (Figure 10).

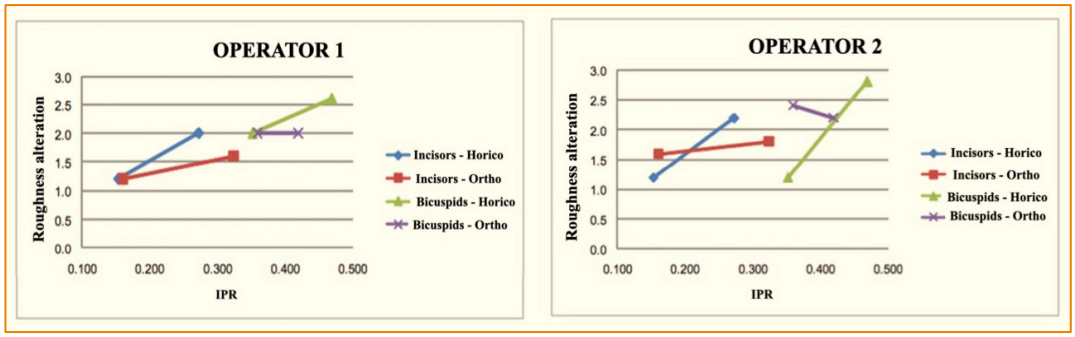

Figure 10. Relationship between IPR quantity and roughness alteration by type of tooth, strip and operator.

A further analysis is conducted to define whether the shape of the incisors (triangular, oval, or rectangular) assumes a relevant role in the morphological variation. The triangular shape always shows a medium morphological variation higher than the other two shapes, although the rectangular and oval incisors show an average variation always greater than 1, too (Figure 11).

Regardless of the type of tooth, the morphological variation following stripping mainly depends on the extent of the stripping, while the diameter, the type of strip and the shape of the tooth itself do not appear to be relevant (Table 5).

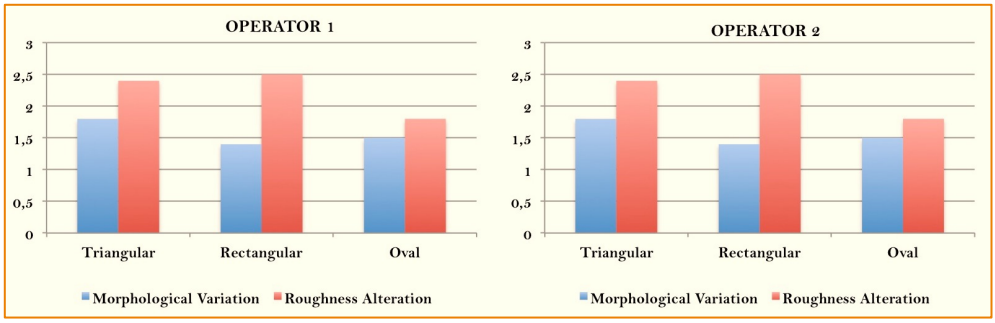

Figure 11. Morphological and roughness alteration concerning the incisor's shape.

Table 5. Regression model multivariate (OLS) on morphological variations.

Variables	Operator 1			Operator 2		
	Incisors		Bicuspids	Incisors		Bicuspids
	(1)	(2)	(3)	(4)	(5)	(6)
Tooth Diameter	-0.115	-0.054	-0.706	-0.189	-0.166	-0.421
	(0.111)	(0.079)	(0.443)	(0.130)	(0.177)	(0.506)
Stripping Quantity	4.804***	5.093***	7.007***	2.845***	3.067***	6.875***
	(0.783)	(0.942)	(1.537)	(1.184)	(1.394)	(1.419)
Ortho Strip	-0.306	-0.396	-0.145	-0.04	-0.113	0.459
	(0.253)	(0.306)	(0.249)	(0.307)	(0.350)	(0.329)
Incisor Form: Rectangular		-0.511			-0.248	
		(0.332)			(0.540)	
Incisor Form: Ovoid		0.152			0.150	
		(0.344)			(0.549)	
Constant	1.284	0.998*	4.814	2.259*	2.145*	2.394
	(0.762)	(0.503)	(3.164)	(0.858)	(1.080)	(3.843)
Observations	20	20	20	20	20	20

***p<0.01; **p<0.05, *p<0.1

The roughness alteration shows two different results depending on the type of tooth. In the case of incisors, it seems to be more sensitive to the type of strip used to carry out the stripping rather than to the extent of the stripping itself, without no distinction for form. Conversely, the roughness of the premolars depends on both the extent of stripping, with a negative sign, and the strip used (Table 6). One element, which is in common between incisors and premolars, is the non-significance of the diameter of the previous tooth stripping in the consequent alteration of roughness.

Table 6. Regression model multivariate (OLS) on roughness changes.

Variables	Operator 1			Operator 2		
	Incisors		Bicuspids	Incisors		Bicuspids
	(1)	(2)	(3)	(4)	(5)	(6)
Tooth Diameter	0.002	-0.021	-0.001	-0.018	-0.028	0.035
	(0.062)	(0.079)	(0.138)	(0.090)	(0.078)	(0.139)
Stripping Quantity	1.564	1.798	-3.369***	3.596***	3.232***	-3.421***
	(1.007)	(1.176)	(0.771)	(1.190)	(1.146)	(0.748)
Ortho Strip	1.453***	0.358	0.429***	-0.104	0.029	0.427***
	(0.160)	(0.205)	(0.140)	(0.162)	(0.188)	(0.139)
Incisor Form: Rectangular		0.07			0.248	
		(0.241)			(0.236)	
Incisor Form: Ovoid		0.238			-0.299	
		(0.275)			(0.252)	
Constant	2.157***	2.241***	3.394***	1.846**	1.908***	3.637***
	(0.502)	(0.560)	(1.093)	(0.592)	(0.589)	(1.120)
Observations	20	20	20	20	20	20

***p<0.01; **p<0.05, *p<0.1.

Discussion

In all teeth analyzed, the treated area has a gingival extension, which is wider than the height of the strip, both in the case of using a generic one or a dedicated one. Indeed, from the digital measurements of the stripping performed, it emerges that the quantity of enamel removed in the same tooth is not equal if different sections are compared.

The amount of enamel that is removed is equal to the amount programmed in correspondence with the measurement point. Instead, it changes if you move more occlusal or more gingival to the point where the micrometer measurement was performed. This is related to the way in which stripping is performed, not only in vestibular-lingual sense but also in the occlusion-gingival sense. If the same amount of enamel were removed at each height, no changes in the shape of the contact point would occur [18]. This is not possible because there are no strips with features complementary to the interproximal surface of the teeth.

In general, bicuspids show a greater morphological variation than incisors. This may be related to the fact that the stripping entity performed in bicuspids was greater. This choice was made because several authors in the literature agree that the amount of enamel that can be removed at the level of bicuspids are greater than that which is advisable to remove from the incisors [15]. Posterior teeth are characterized by a greater quantity of enamel at the interproximal level interproximal than anterior teeth. [17,19,20]. Another reason why the morphological variation of bicuspids is greater could depend on the fact that incisors have a flatter interproximal surface compared to bicuspids, which is more convex.

Incisors can have different shapes: triangular, rectangular, oval. At all stripping ranges, the triangular incisors show morphological variation greater than the other 2 forms. This is quite predictable as one of the indications of stripping is precisely the remodeling of triangular teeth. However, how emerges from the statistical analysis, the number of different shapes (triangular, rectangular, oval) is very different in the 4 groups of the sample. Although teeth have been assigned to various groups randomly, the analysis could be affected by this aspect. However, on average, there are no morphological variations equal to 3, in the stripping ranges considered. It is probably related to the fact that the stripping was performed in ranges considered acceptable from literature.

Regarding the observation at the SEM, it confirms that the IPR significantly changes the surface of enamel, causing the formation of deep grooves and marks. After having performed stripping with Ortho Strip Intensives, almost in all cases, we found out one alteration of the enamel surface equal to 3 (which is the maximum degree of the scale used). Horico strips, on the other hand, cause a significant change in the surface of the enamel but with slightly less alteration. This could be linked to the greater effectiveness in removing enamel with Ortho Strip Intensiv. Instead, common strips Horico are made of steel and have no holes and are subject to wear more quickly than Ortho Strip Intensiv. It can therefore be confirmed that the removal of the enamel must necessarily be followed by polishing [9,21,22].

Finally, a relationship between the extent of the variation in the contact point shape and alteration of the enamel surface characteristics was not observed. Indeed, in case of bicuspids treated with Horico in the wider range (0.4-0.5 mm), a minor superficial alteration of the enamel was observed (grade 2).

Arman et al. [9] carried out SEM observations, at 500X and 1500X magnifications, of intact enamel and enamel treated with diamond strips. In the study performed, SEM observations were made at the same magnifications. Indeed, images of intact enamel and the one treated with the Ortho-Strip are similar to those published previously [9].

According to Arman et al. [9], microscope analysis can only provide a visualization of the surface characteristics of enamel, but other methods are needed to establish quantitatively the extent of the superficial changes. Therefore, these authors did use the profilometer and the microhardness test to obtain data quantities. In any case, they claim that the measurements are consistent with the SEM results.

Grippaudo et al. [16] performed SEM observations of stripped molars with Horico and Komet strips (which are fenestrated diamond strips more similar to the Ortho-Strip Intensiv). Different magnifications were used, including 503X (therefore, a very similar magnification to one of those considered for the study of this thesis). The authors found that treated enamel does not look very different: the number and the depth of the grooves left by the Horico are comparable to those created by the Komet. Also, in the current study, the differences between the SEM images related to the enamel treated with Horico and those related to the enamel treated with Ortho Strip Intensiv are comparable. In addition, Grippaudo et al. [16] pointed out that a clear transition line is observed between the intact enamel area and that of the stripped enamel. The same type of image was obtained in this study from SEM analysis with both types of strips considered (Horico and Ortho Strip Intensiv).

The study by Danesh et al. [10] also compared different stripping methods and evaluated the results in terms of roughness of the enamel surface. According to these authors, the main limitation of the use of SEM is the subjectivity of the observer; moreover, it is not possible to measure the roughness [23,24]. In addition to this, another great limitation of this study is related to the experimental conditions and the fact that it was not possible to recreate the conditions of when we perform IPR in-vivo.

Conclusion

The morphological variation of enamel surfaces depends on IPR extension: for higher ranges, there is a more marked morphological variation in bicuspids for incisors. On the other hand, the type of strip (generic Horico or specific Ortho Strip Intensiv) doesn't seem to have a statistically significant impact on the morphological variation of the teeth. This can be attributed to the fact that more enamel was removed from bicuspid, according to the literature. Based on SEM evaluations, treated enamel appears remarkably damaged regardless of the extent of stripping performed. Ortho Strip Intensiv alters the enamel surface characteristics to a slightly greater extent. Therefore, the use of polishing after removing enamel interproximal is necessary and essential in all cases, as already stated extensively from the literature.

Authors' Contributions

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All authors declare that they contributed to critical review of intellectual content and approval of the final version to be published.

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Conflict of Interest

The authors declare no conflicts of interest.

Data Availability

The data used to support the findings of this study can be made available upon request to the corresponding author.

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