

# Impact of orthodontic movement on clinical, microbiological and tomographic parameters in patients with a history of periodontitis: a pilot study

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## Abstract

**Objective:** The purpose of this study was to evaluate the impact of orthodontic movement on clinical, microbiological and tomographic parameters in patients with a history of periodontitis. **Materials and Methods:** Twelve individuals (2 males and 10 females, mean age 42.1 years) who had undergone treatment for generalized chronic periodontitis were selected from the Periodontal Clinic of Guarulhos University (Brazil). The patients undergoing orthodontic treatment were submitted to a clinical examination, and cone beam computed tomography scans were performed at baseline and after 12 months of treatment. Subgingival biofilm samples were analyzed by checkerboard DNA-DNA hybridization. Statistical analysis was performed by a Wilcoxon test. **Results:** With regard to the periodontal clinical parameters, when comparing baseline and 12-month evaluations, statistically significant reductions were found in Probing Depth and Clinical Attachment Level (CAL). There was a statistically significant increase in relation to the number of sites that had lost CAL. There were no statistically significant tomographic differences, and no loss in the buccal alveolar bone thickness was detected. **Conclusions:** Considering clinical periodontal parameters, a reduction was observed in the number and percentage of CAL. There was a reduction in *A. gerencseriae* and an increase in *F. nuc. nucleatum*, *S. constellatus* and *T. Forsythia* species. Regarding the tomographic parameters, no statistically significant differences were observed.

**Keywords:** Orthodontics. Periodontal disease. Microbiology. Tomography.

## Introduction

In the last two decades, there has been an increase in the adult population's interest in oral health and aesthetics. Although this is a positive trend, some precautions should be taken so that the aesthetic demand is coupled with a broader scope, including periodontal monitoring of these individuals, since periodontitis is a common occurrence in the adult population (Melsen *et al.*, 1989; Zachrisson, 1996; Ong & Wang, 2002; Reichert *et al.*, 2011; Han *et al.*, 2019). Reduced clinical attachment level (CAL), bone defects, pathological tooth migration, tooth losses and reduced vertical dimension are common sequela associated with periodontitis. Thus, any aesthetic treatment in patients with a history of periodontitis would require interdisciplinary therapeutic intervention, which may include orthodontic treatment (Williams *et al.*, 1982; Mathews & Kokich, 1997; Ong *et al.*, 1998; Gkantidis *et al.*, 2010; Roccuzzo *et al.*, 2018).

Clinical studies have shown that it is feasible to perform orthodontic treatment in patients with reduced periodontal support, after they have been treated and are engaged in

a periodontal maintenance program (Melsen *et al.*, 1989; van Gastel *et al.*, 2011). Moreover, it is only after reaching this stage of disease control that the orthodontic treatment plan can be defined (Boyd *et al.*, 1989; Petti *et al.*, 1997; Corrente *et al.*, 2003). Throughout the orthodontic treatment, other points related to these patients should be also addressed, such as the changes in oral microbiota due to the use of bonded brackets (Bergamo *et al.*, 2019). Some authors also consider orthodontic treatment as a two-pronged approach, in which the periodontal condition can sometimes be improved (Melsen, 1991; Zachrisson, 1996), while in other situations, orthodontic treatment might lead to periodontal complications (Naranjo *et al.*, 2006; Verrusio *et al.*, 2018; Reichardt *et al.*, 2019).

Several studies have confirmed that the gingival and periodontal changes occurring during orthodontic treatment are temporary, and usually do not result in permanent periodontal loss (Zachrisson & Zachrisson, 1972; van Gastel *et al.*, 2011; Bergamo *et al.*, 2019). However, some authors have suggested increased loss of clinical attachment occurring during orthodontic treatment (Zachrisson & Alnaes, 1973; Zachrisson, 1976; Boyd *et al.*, 1989; Melsen, 1991; Corrente *et al.*, 2003; Papageorgiou *et al.*, 2018).

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There is a lack of well-designed clinical studies with an interdisciplinary approach to Orthodontics/Periodontics, which would make it possible to establish better levels of planning, treatment and retention (Papageorgiou *et al.*, 2018, Martin *et al.*, 2022). To date, no clinical studies has evaluated the clinical outcomes, subgingival biofilm composition, and cone beam computed tomography (CBCT) images of adult patients who have undergone periodontal treatment and are receiving orthodontic treatment. Therefore, the aim of the present study was to evaluate the effects of bonded full-fixed orthodontic appliances and orthodontic treatment on the periodontium of adult individuals who had been treated for periodontitis and were enrolled in a maintenance program, over a 12-month interval. This evaluation included clinical, microbiological, and tomographic assessments.

## Materials and Methods

Adult individuals, who attended the Dental Clinic at Universidade Guarulhos (Guarulhos, SP, Brazil) were consecutively selected in the period between March 2011 and July 2012, to participate in the present study. A single trained professional (Maurilo de Mello Lemos, MML) selected the participants. The 12 volunteers (2 males and 10 females) with a mean age of 42.1 years (35–49 years) underwent treatment for generalized periodontitis (Stages III/IV) and received periodontal maintenance. The individuals were informed of the study objectives, risks and benefits, and of the therapies to which they would be submitted. The individuals who agreed to participate in the study signed a term of free and informed consent, answered a health questionnaire/anamnesis, and received periodontal maintenance therapy and orthodontic treatment, in accordance with the guidelines and rules of the National Health Council (Resolution No. 466/2012). The project was approved by the Human Research Ethics Committee of the Universidade of Guarulhos (N0156 / 2007).

## Inclusion criteria

- » Age equal to or older than 30 years.
- » Minimum of 20 teeth, excluding third molars.
- » Angle Class I, II or III malocclusion needing corrective orthodontic therapy, and presenting light to moderate crowding.
- » Patients with a history of generalized periodontitis meeting the following parameters at baseline:  $\geq 30\%$  of sites with probing depth (PD) and CAL  $\geq 4\text{mm}$ , and a minimum of six teeth with at least one non-contiguous interproximal site with concomitant PD and CAL  $\geq 5\text{mm}$ , preferably distributed in different sextants (see section “Clinical Evaluation”).
- » Patients should have completed active periodontal treatment at least six months before entering

the study, and should be enrolled in periodontal maintenance program. These individuals were treated at Guarulhos University clinics.

## Exclusion criteria

- » Smokers and ex-smokers who quit less than five years ago
- » Individuals with systemic diseases that could compromise the host's response, bone metabolism and the course of periodontal diseases (e.g. diabetes mellitus, immune deficiencies, osteoporosis, rheumatoid arthritis, etc.).
- » Pregnant or lactating women.
- » Antibiotic therapy and use of anti-inflammatory drugs in the last three months.
- » Continued use of mouthwashes in the last three months.
- » History of previous orthodontic treatment.
- » Caries lesions.
- » Mouth breathers.
- » Xerostomia and/or reduced salivary flow.
- » Continual use of medications that influence salivary flow.
- » Extensive prosthetic rehabilitation.

## Experimental design

At the beginning of the study, all individuals underwent anamnesis and periodontal clinical examination, in accordance with the inclusion criteria described above. They also underwent supragingival scaling/polishing, and received general dental hygiene instructions. Patients were asked not to use any other oral care products, such as mouthwashes, during the period of the study. Clinical, microbiological and tomographic parameters were evaluated at baseline and at 12 months after orthodontic treatment. At 12 months, clinical parameters were assessed and microbiological samples were collected before periodontal maintenance procedures.

It is worth mentioning that orthodontic treatments were performed until their completion, with the placement of retainers, even in patients who needed a period longer than 12 months.

## Periodontal maintenance therapy

In the six months prior to the study and throughout the study, subjects received periodontal maintenance. Every three months, individuals received supragingival scaling, prophylaxis, reinforcement of oral hygiene instructions and root planing of sites with periodontal pockets  $\geq 4\text{mm}$ . Scaling was conducted with Gracey curettes 5/6, 7/8, 11/12 and 13/14 (Hu-Friedy, Chicago, USA), performed in a single session. When additional dental treatment needs were observed, patients were referred to the other specialties.

## Orthodontic treatment description

After periodontal preparation, conventional straight-wire metal orthodontic brackets (0.022 x 0.028-in slot, Roth Max, Morelli – Sorocaba/SP, Brazil) were bonded to both maxillary and mandibular teeth, using standardized light-cured composite (Transbond XT; 3M Unitek, Monrovia, California, USA). In the leveling phase, nickel-titanium (NiTi) (Thermo-Plus, Morelli – Sorocaba/SP, Brazil), copper nickel-titanium (CuNiTi) (Thermo-Active Copper NiTi, Ormco Corp. Thermodynamic – Orange, CA, USA) and chrome-nickel (CrNi) (Arco intraoral CrNi, Morelli – Sorocaba/SP, Brazil) archwires were used. Wire progression was performed at each appointment, with four to five-week intervals, according to the following sequence: 0.014-in NiTi, 0.016-in NiTi, 0.018-in NiTi, 0.019 x 0.025-in 40°C CuNiTi, and 0.019 x 0.025-in CrNi.

## Clinical evaluation

Two examiners were trained and calibrated (Araujo *et al.*, 2003) for evaluation of the following clinical parameters: visible plaque (VP); gingival bleeding (GB); bleeding on probing (BoP); suppuration (SUP); PD and CAL.

Measurements were performed at six sites per tooth (mesiobuccal, buccal, distobuccal, distolingual, lingual and mesiolingual) for all teeth, with the exception of the third molars (Ainamo and Bay, 1975).

## Microbiological evaluation

One calibrated examiner collected the samples in nine non-contiguous interproximal sites, uniformly distributed among the four quadrants. Samples of subgingival plaque were collected using individual sterile mini-Gracey curettes (#11/12, Millennium, Golgran, São Caetano do Sul/SP, Brazil) and these were immediately transferred to sterile plastic tubes (Eppendorf AG Barkhausenweg, Hamburg, Germany), containing 0.15mL of TE (10mmol/L Tris-hydrochloric acid, 1mmol/L EDTA, pH 7.6). Afterwards, 0.10mL of 0.5M sodium hydroxide were added to each tube, and the samples were dispersed using a vortex mixer. All samples were stored at -20°C. The checkerboard DNA-DNA hybridization technique was performed by an experienced biologist. This technique was originally designed to examine the microbial composition of supra and subgingival biofilm in healthy and periodontally compromised patients (Socransky and Haffajee, 1994). The technique uses 40 DNA probes for bacterial species associated with periodontal health or disease (Socransky and Haffajee, 1994).

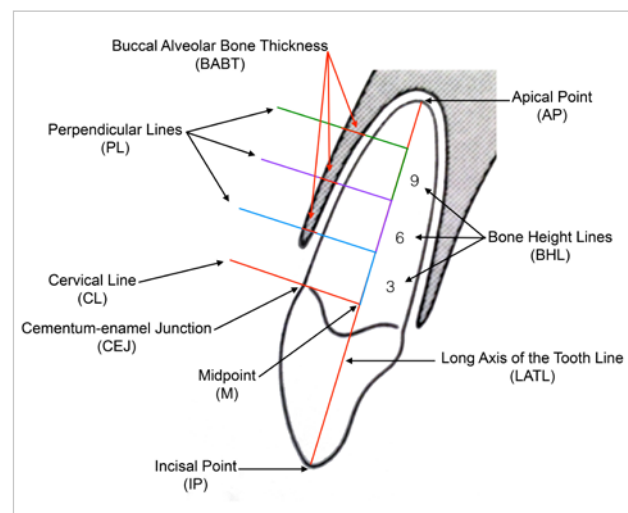
Subsequently, the cells were lysed, and the resultant denatured DNAs were fixed in individual lanes on a nylon membrane (Boehringer Mannheim, Indianapolis, Ind, USA) by using the checkerboard Minislot blot device (Minislot 30; Immunetics, Cambridge, Mass, USA). Digoxigenin-labeled genomic DNA probes (Roche

Applied Science, Indianapolis, Ind, USA) were then hybridized perpendicularly to the lanes of the clinical samples by using a Miniblotter 45 apparatus (Immunetics). Bound probes were detected by using phosphatase-conjugated antibody to digoxigenin (Roche Applied Science) and chemiluminescence. Signals were evaluated visually by comparison with the standards at  $10^5$  and  $10^6$  bacterial cells for the test species on the same membrane. They were recorded as 0 (not detected), 1 ( $<10^5$  cells), 2 (approximately  $10^5$  cells), 3 ( $10^5$  to  $10^6$  cells), 4 (approximately  $10^6$  cells), or 5 ( $>10^6$  cells) (Feres *et al.*, 2004).

## Tomographic evaluation (CBCT)

All images used in this investigation were obtained by the i-CAT Imaging System (Imaging Sciences International, Hatfield, PA, USA), and the imaging acquisition parameters were: 5 mA, 120 kV, field of view (FOV) of 13-cm height/16-cm diameter, and 20-second exposure time, which generated an isotropic voxel size of 0.2 mm (Morais *et al.*, 2018). The files were acquired in Digital Imaging and Communications in Medicine (DICOM) format, and the images were processed and reconstructed using Mimics v. 11 software (Materialise, Lenven, Belgium). Maxillary and mandibular volumetric rendering was performed by one calibrated examiner (MML), and bucco-central slices of teeth (#11, #21, #31, #41, #15, #25, #35, #45, #16, #26, #36 and #46) were generated, according to Cattaneo *et al.* (2011).

The measurements (Fig 1) were obtained by a calibrated examiner using the Mimics software, who was a different professional from the examiner who performed the clinical examination.



**Figure 1. Illustration of measurement of the maxillary central incisor, with all reference points and lines represented. Incisal point (IP); Apical point (AP); Buccal alveolar bone thickness (BAPT); Perpendicular lines (PL); Cervical line (CL); Cementum-enamel junction (CEJ); Midpoint (M); Long axis of the tooth line (LATL) and Bone height lines (BHL).**

Initially, for each tooth, a line was drawn from the long axis of the tooth (LATL) up to the axial section selected, passing through the incisal point (IP), crossing the center of the root to the apical point (AP) (Fig 2). This line can be drawn irrespective of the angulation/rotation of the tooth relative to the alveolar process, or the presence of crowding. On these images, the buccal alveolar bone thickness (BABT) values were assessed at 3, 6 and 9mm from the cervical line (CL) at the cementum-enamel junction (CEJ) (Lemos *et al.*, 2020).

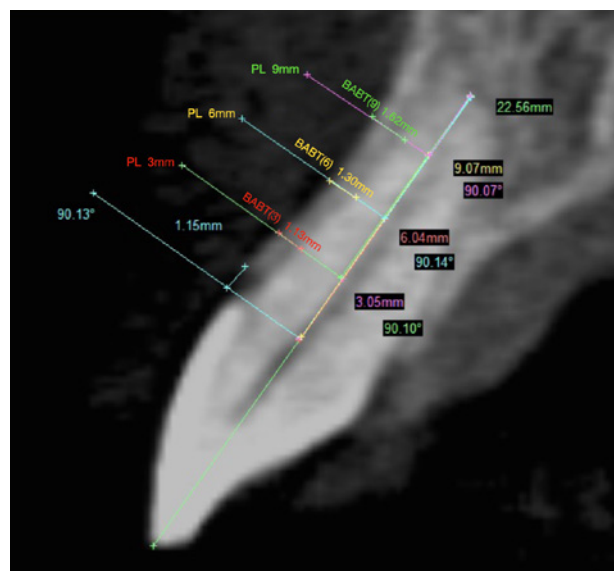
### Statistical analyses

The percentage of sites with VP, GB, BoP and SUP, and the mean values for PD and CAL, number and percentage of sites that gained or lost CAL ( $\geq 2$ mm) between baseline and 12 months were computed for each patient and then averaged across patients. Similarly, changes in bone level height and bone thickness at 3mm, 6mm and 9mm and mean counts ( $\times 10^5$ ) and proportions (%) of individual bacterial species, as well as of the microbial complexes (Socransky *et al.*, 1998), were averaged within each patient and then averaged across patients.

The significance of differences between time points for the clinical, tomographic and microbiological parameters was assessed with the Wilcoxon test. The analyses were made using a statistical program developed by Socransky *et al.* (1998) (Forsyth, USA). The level of significance was set at 5%. For the tomographic measurements, the error of the method was also calculated using the Dahlberg formula (Cattaneo *et al.*, 2011). The data were statistically analyzed using SPSS v. 17.0 (Chicago, IL, USA).

### Results

The clinical characteristics and mean periodontal clinical parameters evaluated at baseline and at 12 months of orthodontic treatment are shown in Table 1.



**Figure 2.** The CBCT measurements of buccal alveolar bone thickness (BABT) of the maxillary central incisor, with all reference points and lines represented. In these three perpendicular lines (PL), the measures at three segments of buccal alveolar bone thickness. BABT(3) = 1.13mm, BABT(6) = 1.30mm and BABT(9) = 1.62mm, respectively, are represented at different predetermined heights: PL 3mm, PL 6mm, and PL 9mm (Mimics 11 software - Materialise, Leven, Belgium).

Twelve individuals were included in this study. The mean age was 42.1 years (ranging from 35 to 49 years). Frequency distribution of malocclusion, according to the Angle classification, was: 25.0% for Class I, 58.3% for Class II, and 16.7% for Class III. There were no subject or site dropouts during the study period.

Statistically significant reductions between baseline and 12 months were observed for PD ( $3.0 \pm 0.8$ mm to  $2.8 \pm 0.5$ mm,  $p = 0.001$ ) and CAL ( $3.6 \pm 1.1$ mm to  $3.2 \pm 0.7$ mm;  $p = 0.001$ ). There was also a statistically significant

**Table 1.** Clinical characteristics, means ( $\pm$  SD) of periodontal clinical parameters, and confidence intervals (CI) at baseline and after 12 months of orthodontic treatment.

| VARIABLE                       | TIME      | means $\pm$ SD    | CI (95%)    |
|--------------------------------|-----------|-------------------|-------------|
| Probing depth (mm)             | Baseline  | $3.0 \pm 0.8^a$   | 2.5 - 3,5   |
|                                | 12 months | $2.8 \pm 0.5^a$   | 2.5 - 3,1   |
| Clinical attachment level (mm) | Baseline  | $3.6 \pm 1.1^a$   | 3.0 - 4.2   |
|                                | 12 months | $3.2 \pm 0.7^b$   | 2.8 - 3.6   |
| Percentage of sites with       |           |                   |             |
| Visible plaque                 | Baseline  | $20.4 \pm 17.8^a$ | 10.3 - 30.5 |
|                                | 12 months | $35.6 \pm 18.0^a$ | 25.4 - 45.8 |
| Gingival bleeding              | Baseline  | $4.6 \pm 11.5^a$  | 0 - 11.1    |
|                                | 12 months | $3.4 \pm 3.8^a$   | 1.2 - 5.6   |
| Bleeding on probing            | Baseline  | $7.5 \pm 4.5^a$   | 5 - 10      |
|                                | 12 months | $14.6 \pm 23.5^a$ | 1.3 - 27.9  |
| Suppuration                    | Baseline  | $0.2 \pm 0.6^a$   | 0 - 0.5     |
|                                | 12 months | $0.1 \pm 0.2^a$   | 0 - 0.2     |

increase in the percentage of sites with VP ( $20.4 \pm 17.8\%$  to  $35.6 \pm 18.0\%$ ,  $p = 0.003$ ) and BoP ( $7.5 \pm 4.5\%$  to  $14.6 \pm 23.5\%$ ,  $p = 0.003$ ), and a significant reduction in GB ( $4.6 \pm 11.5\%$  to  $3.4 \pm 3.8\%$ ,  $p = 0.003$ ) after the 12-month orthodontic treatment interval.

Table 2 shows a more detailed analysis of the variable CAL, in relation to the number and percentage of sites that gained or lost ( $\geq 2$  mm) attachment or remained stable (variation between -1 mm and 1 mm) throughout orthodontic therapy. It was possible to observe that 6% of the sites lost attachment, but 94% of the remaining sites gained attachment (19%) or remained stable (75%).

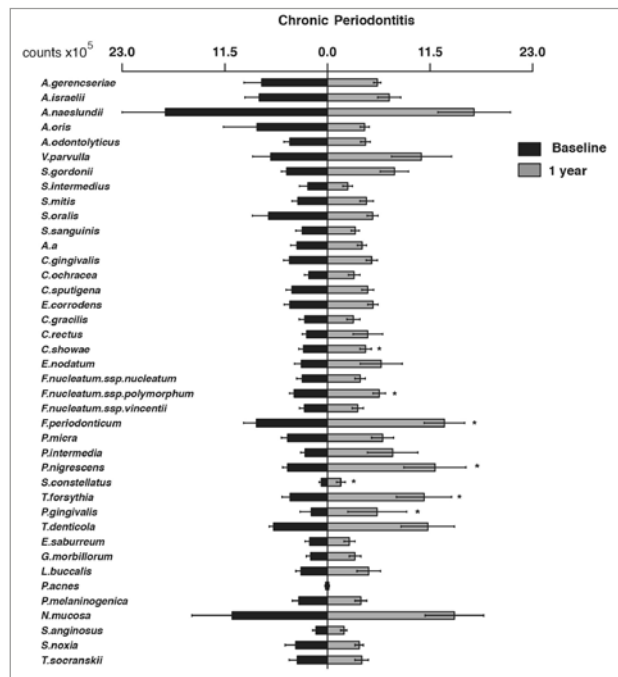
At baseline, microbiological profiles (average count  $\times 10^5$  of the 40 bacterial species) were compatible with periodontal health (Fig 3). The overall levels of species considered beneficial remained stable at 12 months, especially

*Actinomyces* sp., while levels of some periodontal pathogens increased, such as *Campylobacter showae*, *Fusobacterium nucleatum polymorphum*, *Fusobacterium periodonticum*, *Prevotella nigrescens*, *Streptococcus constellatus*, *Tannerella forsythia* and *Porphyromonas gingivalis*. When the proportions of the 40 individual species were evaluated, it was observed that one species (*Actinomyces gerencseriae*) decreased (Fig 4), while there was an increase in the proportions of *F. nuc. nucleatum*, *S. constellatus*, and *T. forsythia* at the end of 12 months of orthodontic treatment.

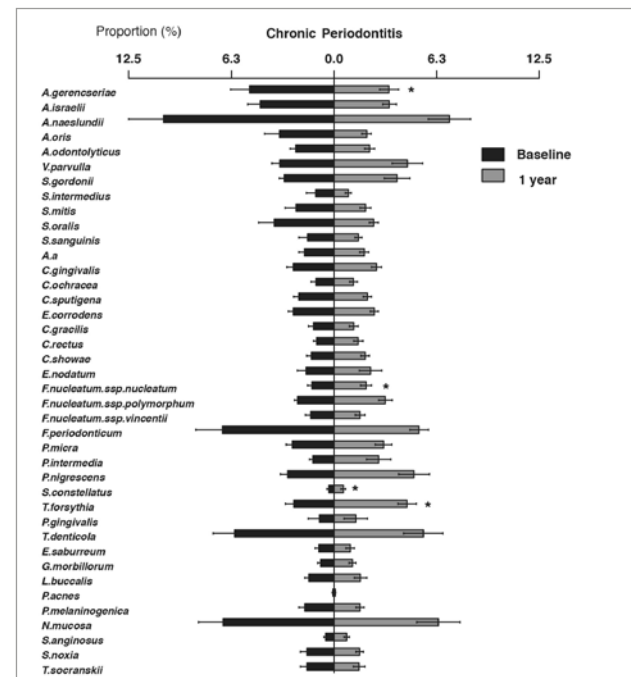
No statistically significant changes were observed for the microbial complex over the course of the study (Fig 5). There were no statistically significant differences in tomographic data between treatment periods, i.e., the analysis at 12 months showed no loss of structure in the buccal alveolar bone thickness (Table 3).

**Table 2.** Mean ( $\pm$  SD) number (n) and percentage (%) of sites that gained ( $\geq 2$  mm) or lost ( $<2$  mm) clinical attachment (CA) or remained stable (ranging from -1 mm to 1 mm) during the 12 months of orthodontic therapy.

| Variable                   | Status          | Mean $\pm$ SD    |
|----------------------------|-----------------|------------------|
| Number (n)<br>of sites     | Gained CA       | 26.8 $\pm$ 20.3  |
|                            | Lost CA         | 9.5 $\pm$ 4.9    |
|                            | Remained stable | 109.5 $\pm$ 24.1 |
| Percentage (%)<br>of sites | Gained CA       | 19.0 $\pm$ 14.0  |
|                            | Lost CA         | 6.0 $\pm$ 3.0    |
|                            | Remained stable | 75.0 $\pm$ 14.0  |

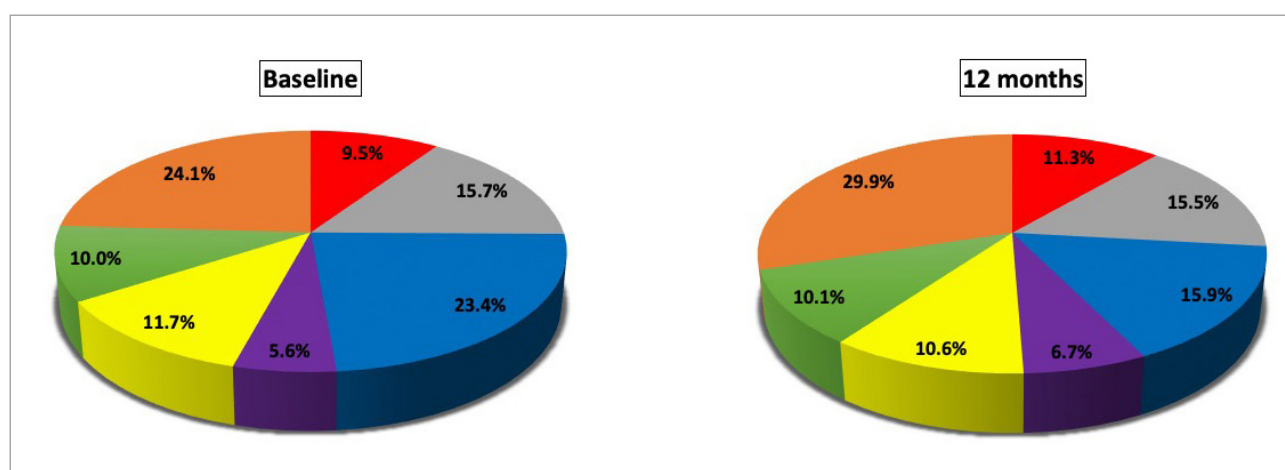


**Figure 3.** Levels (average count  $\times 10^5$ ) of the 40 bacterial species evaluated at baseline and after 12 months of orthodontic treatment (\*Wilcoxon test,  $p < 0.05$  indicates differences between baseline and 12 months).



**Figure 4.** Proportions of the 40 bacterial species evaluated at baseline and after 12 months of orthodontic treatment (\*Wilcoxon test,  $p < 0.05$  indicates differences between baseline and 12 months).





**Figure 5.** The different colors are the mean proportions of microbial complexes (Socransky *et al.*, 1998) of 40 bacterial species evaluated in subgingival biofilm samples collected at baseline and at 12 months after orthodontic treatment (\*Wilcoxon test, differences between baseline and 12 months,  $p < 0.05$ ).

**Table 3.** Mean ( $\pm$  SD) of the measurements obtained by tomography at baseline and after 12 months of orthodontic treatment.

| Variable  | Time      | Mean $\pm$ SD              |
|-----------|-----------|----------------------------|
| Thickness |           |                            |
| 3mm       | Baseline  | 1.2 $\pm$ 0.4 <sup>a</sup> |
|           | 12 months | 1.1 $\pm$ 0.2 <sup>a</sup> |
| 6mm       | Baseline  | 1.2 $\pm$ 0.3 <sup>a</sup> |
|           | 12 months | 1.2 $\pm$ 0.2 <sup>a</sup> |
| 9mm       | Baseline  | 1.4 $\pm$ 0.3 <sup>a</sup> |
|           | 12 months | 1.5 $\pm$ 0.3 <sup>a</sup> |

Equal superscript letters indicate absence of difference between the times within the group (Wilcoxon test).

## Discussion

The results of this study showed that orthodontic therapy in individuals with history of severe generalized periodontitis and under periodontal maintenance does not affect periodontal stability within a period of 12 months. Although some sites lost clinical attachment, there was a mean full-mouth gain in clinical attachment at 12 months after orthodontic treatment. However, a slight negative influence was observed in the subgingival microbiota, with an increase in the levels of some periodontal pathogens.

The impact of orthodontic treatment on the periodontium remains a controversial topic. Although there seems to be no reliable evidence on the actual effects of orthodontic therapy on patients during the periodontal maintenance phase (Bollen *et al.*, 2008), there is no formal contraindication for orthodontic treatment in adults who have had severe periodontal disease. This is provided that they have been properly treated and are in a stable condition before beginning orthodontic treatment (Williams *et al.*, 1982; Boyd *et al.*, 1989; Melsen *et al.*, 1989; Melsen, 1991; Mathews and Kokich, 1997; Ong *et al.*, 1998; Cardaropoli *et al.*, 2001; Ong & Wang, 2002; Corrente *et al.*, 2003; Gkantidis *et al.*, 2010).

Previous studies have investigated the impact of treatment with full-fixed orthodontic appliances on periodontal clinical parameters (Melsen *et al.*, 1989; Ong and Wang, 2002; Corrente *et al.*, 2003; Re *et al.*, 2004; Speer *et al.*, 2004; Cirelli *et al.*, 2006; Reichert *et al.*, 2011; Agarwal *et al.*, 2014) and microbiological parameters (Diamanti-Kipioti *et al.*, 1987; Petti *et al.*, 1997; Paoloantonio *et al.*, 1999; Sallum *et al.*, 2004; Speer *et al.*, 2004; van Gastel *et al.*, 2007; Lo Bue *et al.*, 2008; van Gastel *et al.*, 2008; Thornberg *et al.*, 2009; Rego *et al.*, 2010; van Gastel *et al.*, 2011; Baka *et al.*, 2013). However, there is still lack of reliable information for the actual effect of orthodontic treatment in the biofilm composition and in adults, as several studies focused on young patients (Artun and Urbye, 1988; Boyd *et al.*, 1989; Corrente *et al.*, 2003; Sallum *et al.*, 2004; Speer *et al.*, 2004; Lo Bue *et al.*, 2008).

Whereas previous studies found some clinical attachment loss during orthodontic treatment (Zachrisson & Alnaes, 1973; Zachrisson, 1976; Boyd *et al.*, 1989), in our study we found that patients undergoing periodontal maintenance experienced a gain in the CAL ( $3.6 \pm 1.1$ mm to  $3.2 \pm 0.7$ mm). The most plausible hypothesis for this finding is that all patients in our study

were treated with adjunctive metronidazole and amoxicillin, a protocol that has shown to be the most effective one to treat severe periodontitis (Borges *et al.* 2017, Teughels *et al.* 2020). During the 12-month period of orthodontic treatment, only two teeth (molars, in two patients) were indicated for extraction. It is important to emphasize that, at the baseline, these teeth had PD exceeding 5 mm, in addition to furcation involvement.

The microbiological analysis assessed the impact of orthodontic therapy on the levels and proportions of 40 bacterial species, which have been shown to be good biological markers for periodontal dysbiosis/homeostasis (Feres *et al.*, 2020). At baseline, patients harbored low proportions of red complex ( $9.4 \pm 5.8\%$ ) and high proportions of *Actinomyces* *sp* group, yellow and green complexes, which were compatible to periodontal health (Feres *et al.*, 2004). After 12 months, we observed higher levels of *C. showae*, *F. nuc. polymorphum*, *F. periodonticum*, *P. nigrescens*, *S. constellatus*, *T. forsythia* and *P. gingivalis*. Moreover, there was reduction in *A. gerencseriae* and an increase of *F. nuc. nucleatum*, *S. constellatus*, *T. forsythia*. A slight increase in specific periodontal pathogens during orthodontic treatment have also been reported by other authors (Naranjo *et al.*, 2006, Thornberg *et al.*, 2009, Torlakovic *et al.* 2013). Despite the increase in levels of individual periodontal pathogens after treatment, the proportions of red complex remained stable at 12 months (Socransky *et al.*, 1998), and no statistically significant changes were observed in any other the microbial complexes. These data suggest that the orthodontic treatment did not result in significant inflammatory changes in periodontal patients under periodontal maintenance.

The specific changes observed in the subgingival biofilm during orthodontic treatment may be explained by the brackets design and their irregular surface, which may create retentive areas for biofilm formation (Speer *et al.*, 2004; van Gastel *et al.*, 2007; Nelson-Filho *et al.*, 2012; Gujar *et al.*, 2020). Indeed, many authors agree that gingivitis is more likely to occur during the use of orthodontic appliances (Re *et al.*, 2004; Speer *et al.*, 2004; Lo Bue *et al.*, 2008; van Gastel *et al.*, 2011; Baka *et al.*, 2013; Torlakovic *et al.*, 2013; Agarwal *et al.*, 2014; Ghijselings *et al.*, 2014; Ireland *et al.*, 2014; Liu *et al.*, 2014; Yáñez-Vico *et al.*, 2015). Therefore, rigorous periodic control of oral hygiene plus instructions and motivation before during the orthodontic treatment is crucial for treatment success with low impact in the periodontium (Melsen *et al.*, 1988; Melsen *et al.*, 1989; Melsen, 1991; Ong *et al.*, 1998; Ong and Wang, 2002; Re *et al.*, 2004; Bergamo *et al.*, 2019; Reichardt *et al.*, 2019).

We utilized tomographic examination (CBCT) in this study because it is considered the gold standard diagnostic tool in Orthodontics. Although routine use of CBCT in daily clinical practice is not always feasible due to its high cost, in this research, it was the only method

available to determine the maxillary buccal alveolar bone thickness and height with high resolution (Morais *et al.*, 2018). In the present study, after 12 months of orthodontic treatment, no significant bone loss was detected by CBCT. A few previous studies using radiographs (Kloehn and Pfeifer, 1974; Bondemark, 1998) revealed no areas of permanent alveolar crest resorption during orthodontic treatment, which is in agreement with our data. In a previous study with adults in good oral health, Suomi *et al.* (1971) found that the maintenance of these good oral conditions made it possible to restrain the rate of alveolar bone loss. The only previous study using CBCT scans in one group of adult patients under periodontal maintenance and a second one with normal periodontal tissues, after fixed orthodontic treatment, reported significant bone loss (Ma *et al.*, 2015). Although these results were in contrast to ours, it is essential to consider that they measured bone density and we measured bone thickness. Furthermore, Ma *et al.* (2015) found no significant differences in bone height between the two groups under orthodontic treatment. Our results are consistent with this finding.

Other studies using radiographs showed that patients with reduced periodontium undergoing orthodontic treatment can present minimal or no bone loss during the orthodontic movement (Artun & Urbye, 1988; Melsen *et al.*, 1989; Cardaropoli *et al.*, 2001; Corrente *et al.*, 2003; Bollen *et al.*, 2008) and, in some cases, bone gain (Artun and Urbye, 1988; Boyd *et al.*, 1989; Melsen, 1991; Corrente *et al.*, 2003). Relative to data obtained by CBCT analysis, only Morais *et al.* (2018) analyzed the changes in the buccal alveolar bone thickness in periodontally healthy patients undergoing orthodontic treatment with self-ligating brackets. They found reduction in bone thickness after treatment, which contradicts our results. Some aspects that may have contributed to the maintenance of bone thickness in our study are good oral hygiene and absence of gingivitis (Melsen *et al.*, 1988; Melsen *et al.*, 1989; Melsen, 1991; Ong *et al.*, 1998; Ong and Wang, 2002; Re *et al.*, 2004). In contrast to our study, Fuhrmann (1996) also applied the straight wire technique associated with continuous archwires. They observed a reduction in the buccal alveolar bone thickness when a system of forces not controlled and rectangular wires was applied, which led to uncontrolled forces in some cases, resulting in gingival recession. These results may probably be associated with individual variability of buccal alveolar bone thickness, CAL, biomechanics, and the straight wire technique. On other studies using the same straight wire technique with continuous archwires (Edwards, 1976), and the biomechanics focused on light and continuous forces, the researchers found the bone level stabled (Edwards, 1976; Artun and Urbye, 1988; Melsen *et al.*, 1988; Ogaard, 1988; Melsen *et al.*, 1989; Melsen, 1991; Ong and Wang, 2002;

Corrente *et al.*, 2003; Leung *et al.*, 2008; Gkantidis *et al.*, 2010). It should be noted that, to achieve the required levels of force, a special Thermo-Active Copper NiTi wire (Ormco Corp. Thermodynamic) should be used. This has specific characteristics using transitional temperature 40°C, providing the necessary minimum and the most constant level of force (5 to 15g) per tooth (Melsen *et al.*, 1989; Melsen, 1991; Cardaropoli *et al.*, 2001; Corrente *et al.*, 2003; Dalstra and Melsen, 2004; Sakima *et al.*, 2006; Gkantidis *et al.*, 2010; Agarwal *et al.*, 2014). Moreover, the study of Morais *et al.* (2018) was conducted with the use of self-ligating brackets, as opposed to ours, in which straight wire brackets were used. Another potential reason for controversy could be our patients' ages, which ranged between 30 and 57 years. In the cited study (Morais *et al.*, 2018), the patients were between 11 and 17 years old. Most importantly, we cannot compare these two different groups of patients, since in the cited study (Morais *et al.*, 2018), the patients showed no signs of periodontal problems.

In our study, the maxillary incisors of Angle Class II patients (58.3%) were more buccally inclined, as expected, due to the tooth pathology (Artun & Urbye, 1988; Melsen *et al.*, 1989; Ong *et al.*, 1998; Cardaropoli *et al.*, 2001; Corrente *et al.*, 2003; Gkantidis *et al.*, 2010; Reichert *et al.*, 2011; Agarwal *et al.*, 2014). In such cases, lingual movements are considered beneficial. Therefore, in accordance with our results, the alveolar bone thickness could be maintained by bone remodeling in order to compensate for this lingual orthodontic movement, by means of mechanisms of bone apposition and resorption, thereby maintaining the stability of buccal alveolar bone (Edwards, 1976).

The limitation of this study was the small sample size, consisting of only 12 subjects, and the absence of

a control group. However, the main strengths included the inclusion of adult subjects, a robust microbiological evaluation, and a follow-up period of 12 months. Another strength was that a single professional (MML) conducted the entire orthodontic treatment, following the same protocol for all patients. In the majority of controlled clinical trials with approaches similar to ours, there were multiple professionals performing the orthodontic treatment (Boyd *et al.*, 1989; Speer *et al.*, 2004; Naranjo *et al.*, 2006; Morais *et al.*, 2018). Additionally, the three-dimensional analysis provided by CBCT is the most reliable tool for planning, monitoring treatment, and documenting treatment progress for periodontal patients (Farman and Scarfe, 2006; Misch *et al.*, 2006; Cattaneo & Melsen, 2008; Harrell, 2009; Lund *et al.*, 2010; Kapila *et al.*, 2011).

Finally, from our perspective of future studies, chair-side tests designed to evaluate the oral ecology would be helpful to identify stable patients that could safely undergo orthodontic treatment. Future studies should also address longer-term clinical, microbiological, immunological and tomographic evaluations in adult patients with different severities of periodontitis undergoing periodontal maintenance phase.

## Conclusions

Patients with history of severe periodontitis enrolled in a regular maintenance program may be successfully treated by orthodontic therapy without negative consequences to periodontal stability. However, the treatment may have a slight negative impact on the subgingival biofilm, underscoring the importance of maintaining strict periodontal maintenance for these patients during and after orthodontic therapy.

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