UNIVERSITA DEGLI STUDI DI MILANO

XXXII CYCLE DOCTORAL RESEARCH IN ORAL SCIENCES

Effects of the innovative minimally invasive method of orthodontic treatment acceleration on the periodontal conditions. INTERIM RESULTS

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Table of contents

I Overview of Ortho-Perio interactions

Introduction

Thanks to the increasing demand in appearance, orthodontic treatment is being more and more adopted in the adult population. As adult orthodontic patients may also have restorative and periodontal needs, the interaction between different specialties becomes even more important. That means orthodontic therapy of adult patients has become an essential part of the complex dental treatment, that was also described in the previous issue. As we know the aim of orthodontic therapy is the correction of teeth position with appropriate interrelationship of dental arches providing better dental and facial esthetics and function. It might be a self-sufficient treatment or it may be part of the pre-prosthetic preparation, when favorable occlusal conditions are needed to provide a satisfactory prosthetic rehabilitation. Although adult orthodontics requires a slightly different treatment approach and does have some limitations when compared to adolescents orthodontics, it may be successfully applied in many clinical scenarios. For instance it can be used to create a space for implant placement or it may prevent excessive tooth preparation (in case of tipped teeth) ameliorating long-term prognosis for dentition due to the balanced occlusal loading.

Patients affected by periodontal disease may present with pathological tooth migration or other deformities where orthodontics may represent an important part of their treatment. These patients may need orthodontic interventions to correct spacing, proclination of incisors, rotation and tipping. As a consequence of the high prevalence of periodontal disease among adults aged ≥30 years (1), modern orthodontists may often meet patients affected by periodontal disease and other preexisting conditions. Tooth malposition is also a risk factor for periodontal disease progression due to root proximity and to the reduced effectiveness of hygiene procedures contributing to the periodontal inflammation (2). Occlusal forces not regularly distributed act not along the tooth axis and may create an occlusal trauma, affecting periodontal health (3). When treatment planning complex clinical cases with malpositioned periodontally compromised teeth, orthodontic treatment should be considered, which may positively influence the long-term prognosis of the maintained periodontally compromised teeth (4). Both periodontists and orthodontists should understand each other's perspective and cooperate in clinical practice to deliver the best possible result to their patients.

The amount of publications dedicated to the interdisciplinary dental treatment including orthodontics and periodontology has lately noticeably increased. Articles about interactions between these two fields are mostly focused on observations in limited topics, or reports of applied treatment approaches in managing complex dental problems. Technological progress in the last 30 years has significantly changed the world of dentistry in a way that specialists tend to be highly specified in a narrow dental field and prefer to restrict the area of their competence. For this reason, we would like to give a more comprehensive description of the complex interaction between these two different disciplines.

At first, we will introduce the basic mechanisms and cell-signalling pathways involved with tooth movement, that are of paramount importance for both orthodontists and periodontists. A large number of publications in both fields has deeply investigated these mechanisms, and the first chapter will attempt to provide a description of the aforementioned processes, which may be useful in both specialties' points of view.

Another area where orthodontics and periodontics have important connections is the evaluation of the influence on periodontal parameters and microbiological changes during the therapy. It is interesting to notice the impact that both disciplines may have on the oral ecosystem and how complex these interactions can be. Microbiological changes determined by fixed orthodontic appliances wearing, plaque accumulation and the response of periodontal tissues to such changes will be discussed.

After first chapters dedicated to micro-changes, we will observe how it may be realised on macro-level. Next chapter is dedicated to the use of TADs and dental implants to obtain absolute anchorage, the response of periodontal tissues to bone modification agents, the possible adoption of orthodontic therapies to prevent or correct tissue deformities or to provide a valid treatment alternative.

Orthodontic tooth movement causes reorientation and remodelling of supporting periodontal tissues. When teeth are moved into a new positions it causes changes in soft tissue and in underlying bone. In the following two chapters will be extensively described the possible effects of orthodontic treatment on periodontal tissues: how to prevent a deterioration of periodontal conditions, how to treat patient if one of adverse effects had place, when it is the best time to interact with colleagues.

Finally - while active treatment often plays the leading role in symposia, meetings and in the available literature - maintenance of patients' health, function and aesthetics following active therapy must be a priority in the management of both specialties' populations.

Knowing that there are many prejudices about unfavourable influence of the orthodontic therapy on periodontal status we invite you to get acquainted with the evidence and to summarise if orthodontic treatment may have a detrimental effect on periodontal tissues, when it may happen and how to avoid it. Since both periodontists and orthodontists work primarily on tooth supporting structures, soft and hard periodontal tissues' changes during tooth movement and inflammatory disease must be fully understood from both specialties' perspectives.

We hope, that it will help you to run your clinical practice in a better way, to avoid possible mistakes and to provide the best possible treatment options to your patients.

1. Biological aspects of orthodontic tooth movement

Every orthodontic tooth movement (OTM) causes a variety of microscopic and macroscopic changes. When applying mechanical forces to the tooth supporting structure cell-signalling pathways are activated through numerous cascade reactions, which stimulate periodontal ligament (PDL) turnover and bone turnover. It is well-known, that the inflammation is a complex biological process that occurs in response to infection and/or other triggers results in tissue damage (5). Certainly, besides common basic mechanisms of inflammation that are present during both OTM and periodontal disease there are some noticeable differences: after the application of the orthodontic forced a strain and localised hypoxia induce an aseptic inflammation devoid of bacteria that results in bone and PDL remodelling.

Tooth movement induced by orthodontic force is the result of placing controlled forces on teeth. The applied force causes remodelling changes in the dental and periodontal tissues. Orthodontic force application results in compression of the alveolar bone and the periodontal ligament on one side while the periodontal ligament is stretched on the opposite side. The bone is selectively resorbed on the compressed side and deposited on the tension side (6).

In the “pressure-tension theory” the PDL senses a change in mechanical forces or stresses. The theory proposes that PDL progenitor cells differentiate into compression-associated osteoclasts and tension-associated osteoblasts, causing bone resorption and apposition, respectively (7). The following biological processes are proposed on the compression side: disturbance of blood flow in the compressed PDL, cell death in the compressed area of the PDL (hyalinization), resorption of the hyalinized tissue by macrophages, and undermining bone resorption by osteoclasts beside the hyalinized tissue. It is proposed that tooth movement will not take place until the necrosed tissue is removed by the cells on the compression side.

On the tension side, it is proposed that the periodontium, including the PDL, alveolar bone and cementum remodels and undergoes bone apposition. Osteoblasts differentiate from mesenchymal stem/progenitor cells. Mature osteoblasts form the osteoid or type I collagen matrix, which is later undergo mineralization (8). Endothelial nitric oxide synthase (eNOS) mediates bone formation on the tension side of orthodontic forces (9).

Moreover, levels of ALP during OTM, are noted to be significantly greater in sites of tension than in sites of compression, indicating the differences of the remodelling processes (10). On the compression side expression of bone resorptive mediators nuclear factor-kappa (RANK) and transforming growth factor ß1(TGF-ß1) increase and expression of a bone-forming mediator osteoprotegerin (OPG) decreased (11).

In 1932 Schwarz correlated the tissue response to the magnitude of force with capillary blood pressure. Light orthodontic force, i.e., force less than capillary blood pressure, causes periodontal ligament ischemia with simultaneous bone resorption and deposition resulting in continuous tooth movement. Moderate orthodontic forces, i.e., forces exceeding capillary blood pressure lead to periodontal ligament strangulation resulting in delayed bone resorption. Strong/heavy orthodontic forces, i.e., forces far exceeding capillary blood pressure, cause ischemia and degeneration of the periodontal ligament on the compressed side resulting in hyalinization with more delay in tooth movement (12). If the force exceeds the pressure (20–25 g/cm2 of root surface), tissue necrosis can occur due to the strangulated periodontium (13).

The intensity of applied forces correlates with the amount of the released inflammatory mediators: the stronger the mechanical force is applied the higher the levels of mediators are found in GCF. For example levels of MMP-8 and MMP-9 that take part in collagen remodelling, after application of mechanical forces were found significantly increase (14, 15). This may be interpreted in two different ways: as a negative factor predisposing to periodontal breakdown, and also represent a marker for increased tissue metabolism anticipating a faster response to the applied force.

Although inflammation during orthodontic treatment leads to an increase of levels of numerous inflammatory mediators such as (interleukins (IL), beta Glucuronidase (bG), alkaline phosphatase (ALP), lactate dehydrogenase (LDG), prostaglandin E2 (PGE2), endothelial nitric oxide synthase (eNOS) and Ca2+ independent nitric oxide synthase (iNOS), Matrix metalloproteinase 8 (MMP-8), Matrix metalloproteinase 9 (MMP-9), it has a different mechanism if compared with periodontitis (14-19).

Periodontitis has deal with pro-inflammatory events involving both cells and mediators from innate and adaptive immunity as a defence against invading organisms. The adaptive immune response is activated with involvement of the cellular and non-cellular mechanisms of acquired immunity. Immune mechanisms play further roles in the resolution of inflammation and in the healing process, including the repair and the regeneration of lost or damaged tissues.

2. Orthodontic treatment and environmental changes

Often, periodontal inflammation in patients undergoing orthodontic treatment could be associated not only with mechanical stress in PDL or disturbances in tissue turnover, but also with the presence of dental plaque accumulation both supragingivally and subgingivally. In periodontal patients undergoing orthodontic therapy plaque control has to be closely monitored.

Several aspects may influence plaque accumulation during OTM: the selected type of appliance (fixed or removable), bracket material, type of ligation and type of retainer may affect the patients' ability to maintain a good level of oral hygiene. Plaque Index (PI) in patients with fixed appliances appears to be greater than that in patients wearing removable appliances (20), in its turn, fixed orthodontic appliances may perform differently. Stainless steel brackets appeared to harvest significantly bigger amount of plaque when compared to ceramic and polycarbonate brackets. The greatest amount of plaque in ceramic brackets accumulated on occlusal and gingival surfaces, while plaque accumulation was higher on mesial and distal surfaces of stainless steel brackets (21, 22). Also material of ligatures is important: gingival inflammation was shown to be higher when using elastomeric rings for archwire ligation compared to ligature wires (23, 24).

After active phase of OTM the treatment does not finish, continuing with the phase of retention. The presence of fixed retainers may be associated with a risk of higher level of plaque accumulation, gingival recession, and presence of bleeding on probing due to more complicated maintenance (25). Patients with multi-strand wire retainers exhibited more plaque accumulation on the distal surfaces of the lower anterior teeth in comparison with a single span, round wire retainers (26). Since the choice of orthodontic devices and materials is aways up to the clinician, this aspects are likely to be considered.

The microbiological imbalance derived from orthodontic therapy can be associated not only with the elevated quantity of plaque accumulation. Bacterial composition of dental plaque changes with the presence of orthodontic appliances. Orthodontic bracket-systems appeared to be niches suitable for colonization by gram-negative periodontal pathogens of the orange and red complexes, described by Socransky, 1998 (27). Bacterial species from these groups have been more closely correlated with gingival inflammation and chronic periodontal disease. All brackets of patients who didn't apply any antimicrobial agents were heavily contaminated by at least 1 of the 16 gram-negative periodontal pathogenic bacterial species evaluated (28). For example, young patients wearing fixed orthodontic appliances were found to harbour Actinobacillus actinomycetemcomitans (Aa) with a statistically significant greater frequency of detection than appliance-free individuals (29, 30). Thornberg et al analyzed subgingival microflora for anaerobic or facultative anaerobic bacteria before, during and after OTM and found out, that pathogen counts for 6 of the 8 pathogens increased significantly after being wearing appliances for 6 months. On the other hand after 12 month of fixed orthodontic appliances wearing no pathogen level was significantly higher compared to pretreatment scores due to the improved plaque control following teeth alignment and probably also thanks to a newly established equilibrium of oral ecosystem (31).

After debonding periodontal and microbiological parameters continue to change. Sallum et al reported decreased levels of periodontopathogens 30 days after removal of fixed orthodontic appliances, prophylaxis, and oral hygiene instructions: an approximately 50% reduction of A. actinomycetemcomitans levels with a significant reduction in the proportion of sites positive for B. forsythus. This reduction was compatible with clinical signs of periodontal health after orthodontic appliance removal (32). Periodontal probing depths (PPD), the number of proximal sites that showed bleeding on probing (BOP) and GCF flow increase significantly between baseline and end of treatment on fixed appliances for both the banded and the bonded sites. After three month follow-up PPD and GCF flow appeared to be reduced but still remain significantly higher than at baseline. After removal of fixed orthodontic appliances banded sites may still show a significantly higher number of sites with BOP than before treatment.

The supragingival colony-forming units (CFU) ratio normalised after three months, possibly because these parts used to be more available to the oral hygiene. The subgingival CFU ratio three months after bracket removal remained significantly lower compared with baseline, indicating that changes induced by orthodontic treatment are partially irreversible. Periodontal parameters tended to normalise after debonding, but most values three month after treatment remained significantly elevated, representing some grade of risk for a periodontal health (33).

However clinical changes in periodontal conditions after OTM on fixed appliances may not be necessarily related to worsening periodontal conditions, because one of the most important factors influencing the presence of inflammation in periodontal tissues is a susceptibility to periodontal disease (34, 35). Individual susceptibility is independent from the presence of a well-known retentive plaque factor, i.e. orthodontic appliances and/or bands. Visible plaque index (VPI), gingival bleeding index (GBI), bleeding on probing (BOP), periodontal pocket depth (PPD), clinical attachment level (CAL) may show no statistically significant differences.

The severity and speed of progression of periodontitis depends upon the balance of a number of factors: the number and type of bacteria present, how strong the individual's defence mechanisms are, and the presence or absence of certain risk factors. Important risk factors include inherited or genetic susceptibility, smoking, lack of adequate home care, age, diet, health history, and medications.

To reduce risks of periodontal breakdown during OTM clinicians should pay more attention to periodontal examination and periodontal risk assessment before starting the orthodontic therapy. Also more attention should be paid to the bracket material characteristics and type of ligation. Periodontal status in orthodontically treated patients might be assessed not only during therapy and after debonding, but likely also during follow-ups in retention period.

Professional interactions between periodontology and orthodontics during multidisciplinary dental treatment could be divided in two big groups, depending if any of these two disciplines plays the leading or complimentary role. There are clinical situations, when periodontal health and long-term prognosis may benefit from the correction of the teeth position in the dental arch and there are situations, when periodontist may be useful to a specialist before during and after OT.

3. Anchorage

Anchorage is a very important part of orthodontic treatment and it represents another area of interaction between orthodontics and periodontology. An increasing interest in the orthodontic field has recently arisen in the use of Temporary Anchorage Devices (TADs) to obtain absolute anchorage intraorally. As TADs closely resemble and have a lot in common with dental implants, the interaction between periodontists and orthodontists have been even closer.

Anchorage is defined as the resistance to unwanted tooth movements. Control of anchorage is one of the most important issues in orthodontics and is considered a prerequisite for orthodontic treatment of dental and skeletal malocclusions. Various techniques of anchorage reinforcement have been devised and successfully used in orthodontic practice. Before temporary anchorage devices orthodontists have used teeth, intraoral appliances, and extraoral appliances, to control anchorage. Recently TAD-provided skeletal anchorage is progressively being incorporated into treatment modalities of orthodontic practice. Cope et al indicated a number of characteristics for ideal TADs: simple to use, inexpensive, immediately loadable, immobile, does not require (patient) compliance, biocompatible, and provides clinically equivalent or superior results compared to conventional systems (36).

Absolute anchorage, defined as no movement of the anchorage unit as a consequence to the reaction forces applied to move teeth, may be provided by using ankylosed teeth or implants. Implant anchorage may be obtained with conventional dental implants, palatal implants, mini-plates, and mini screw implants. Conventional dental implants and palatal implants may provide maximum anchorage reinforcement, but they require time to be osteointegrated before loading, increasing the duration of treatment. Hence mini screw implants and plates have become more frequently used in practice.

Sugavara et al observing literature about miniplates' characteristics concluded that mini plates appeared to be the most reliable, predictable, and effective modality (37). Moreover it showed best results in providing stable anchorage independently of the thickness of the cortical bone, probably due to the possible adoption of multiple fixation screws. However mini plates placement requires open flap surgery with more related postoperative complications.

The placement of mini screw implants has demonstrated to be a dependable way of anchorage reinforcement. Titanium mini implants, which are surgically inserted in the bone are small, relatively uncomplicated to place and could be successfully used as intraoral extradental anchorage. The mini implants' stability was suggested to be influenced by direction of the orthodontic force, angle of insertion, shape of thread, insertion torque, length of screws, bone density and thickness of the cortical bone (38-42). The benefit of mini screws application may be affected by patient related factors as systemic diseases, smoking, habits and hygiene level, which should be evaluated while planning in order to avoid undesirable aftereffects. Cone Beam Computer Tomography (CBCT) may be used during treatment in order to better understand individual patient anatomy or it may be used after insertion to better control the placement of the TADs (43). In some cases providing molar distalization using TADs possible to achieve teeth alignment without tooth extraction.

The other possible way to enhance anchorage during OTM is to influence the bone turnover by pharmacological means. Modifying osteoclastic activity we can influence anchorage stability during the active orthodontic treatment and retention. A single low-dose application of bisphosphonates (zoledronate, risedronate) as well as Bis-enoxacin (bisphosphonate derivative of enoxacin) can maintain the stability of mini implants over time by regulation of bone resorption through inhibiting activity of osteoclasts (44-48). Osteoprotegerin-Fc (OPG-Fc) being an inhibitor of osteoclastic activity may affect bone maturation during orthodontic tooth movement. As a consequence, local administration of OPG-Fc may lead to decrease of post orthodontic relapse and probably may reinforce anchorage (49).

Anchorage planning may benefit from the coordinated work of orthodontists and periodontists. Using TADs, oral hygiene should be closely monitored as plaque accumulation near the TAD gingival margin can cause perimucositis, which may lead to breakdown of bone around implants and eventually implant loss (50). After TAD placement, the surrounding soft tissues should be carefully maintained to ensure durability of the implant, thus patients require instructions on daily plaque control at home and periodic professional care, similar to regular periodontal maintenance.

4. Gingival invagination

Orthodontic treatments that include extraction of dental units and movements of adjacent teeth in to the extraction sites can lead to gingival invaginations (GI). Gingival ingrowth was defined by Robinson et al (51) as a linear invagination of the interproximal tissue with mesial and distal orientation and a probing depth of at least 1 mm (Fig. 1). The frequency of GI is reported to be high (approximately 35%) and may be observed more often in the lower jaw (51-53). They vary from mild fissures in keratinized gingiva to deep clefts in the alveolar bone crossing interdental papilla either buccally or lingually through the alveolar bone, penetrating the alveolar ridge in 25% of cases (53). Since these gingival invaginations could serve as potential sites for dental plaque accumulation, it has been considered as a potential risk factor for initiation of periodontal disorders during the course of orthodontic treatment (51, 54). Furthermore GI may be associated with the increased marginal bone loss in the spaces between neighbouring teeth, a reduction in interdental bone height, and increased time necessary for the orthodontic space to close (55).

Investigating the effects of a timely versus a prolonged space closure after tooth extraction on the incidence and severity of gingival invaginations, significantly more GI were reported when there was a delay in space closure and orthodontic treatment was initiated late after tooth extraction (52, 56) therefore proper communication between specialists is particularly important.

According to the GI severity treatment strategies may vary. When GI is located in soft tissues only it may be treated using a cold blade or the electric cautery with no significant difference between the two gingivectomy techniques (57). Soft tissue diode laser in the management of mucogingival problems may present some advantages as minimal post operative pain and no need for suturing (58). In contrast to previously described surgical procedures for the correction of gingival invaginations, a preventive approach was proposed.

In order to prevent GI formation during orthodontic treatment with extractions Tiefengraber et al. used a socket preservation technique, applying e-PTFE membranes after premolar extractions (59). No gingival invaginations appeared with the subsequent translational tooth movement in the membrane-treated areas, whereas gingival invaginations was apparent in almost all the untreated sites.

Another preventive measure, demonstrated in the literature was guided bone regeneration (GBR) with bone replacement materials (60). However the success of tooth movement previously augmented with a synthetic material seems to be strongly influenced by the material properties. For example, the use of non-resorbable hydroxyapatite ceramic showed negative results (61). Meanwhile, using synthetic nanocrystalline hydroxyapatite, which is able to resorb very fast, Reichert et al. obtained significantly lower probing depths of the adjacent teeth and a reduced number of sites with communicating buccal and lingual invaginations (62).

GBR applied to the post extraction area was suggested to be a good approach to prevent GI formation during OTM, however the best timing for to initiate tooth approximation after surgery is still under discussion and sufficient information from controlled clinical studies is missing.

Fig.1. Intraoral view of the patient after completed orthodontic treatment. Treatment plan included mandibular premolars' extraction. There is a gingival invagination within soft periodontal tissues formed by the end of OT. (Courtesy of Dr. Michele Izzo)

5. Gingival enlargement

GE is a very common condition in orthodontic population that is characterised by an increase in the size of gingiva usually resulting in pseudo-pocketing without attachment loss. As it may involve the anterior region it may have an impact on oral health-related quality of life

(63). Traditionally GE was considered as a plaque-related inflammatory reaction consecutive to bacterial plaque accumulation in a difficult-to-access tooth surfaces (64).

Despite this inflammatory factor, other causal factors have been proposed to explain the association between orthodontic treatment and GE.

Chemical irritation produced by materials used for banding, mechanical irritation by bands, and food impaction have been suggested to explain the pathogenesis of GE (65). However already in 1972 Zachrisson and Zachrisson (66) had reported gingival enlargement in patients maintaining excellent oral hygiene. More recently orthodontic patients with good dental hygiene exhibiting GE without any clinical signs of gingival inflammation were studied (15). These patients exhibited elevated MMP-8 and MMP-9 levels in GCF. The authors suggested that during orthodontic treatment, the mechanical stress appeared to be one of the key-factors determining the increase of MMP-9 in GCF and the onset of GE.

In recent years, it has been suggested that a continuing low dose of nickel released to the epithelium from corrosion of orthodontic appliances may be the initiating factor of GE in orthodontic patients.

Some authors also evaluated the possible role of an allergic reaction to nickel, which is commonly used in orthodontic appliances (67-70). In vitro and in vivo studies suggest that released nickel ions may cause an exposure time dependent allergic reaction characterised by an up-regulated proliferation of keratinocytes and increased epithelial cell proliferation (71, 72). It may be therefore important to treat patients with nickel-free appliances and to adopt questionnaires to evaluate previous history of allergies to metals as they have been linked to an increased frequency of GE (73-75).

The time under fixed orthodontic treatment strongly influenced the extension of GE, with more severe manifestations of this condition among patients using orthodontic appliances for longer periods of time). According to location, enlargements could be marginal, papillary or diffuse. Enlargement of interdental papillae and accumulation of gingival tissue may appear due to the application of compressive or retraction forces at the site of extraction space closure. In orthodontic treatment the extraction of teeth may be required. Most commonly, first or second premolars are extracted in both dental arches. Along with GI, an accumulation of gingival tissue may appear on teeth adjacent to the extraction site during the space closure (76).

Based on the extent and severity, these enlargements may lead to functional disturbances like altered speech, difficulty in mastication and aesthetic and psychological problems. The prime treatment modalities involve obtaining a detailed medical history and non–surgical periodontal therapy. In the management of gingival enlargement, patient motivation to maintain meticulous self-care oral hygiene with adjunctive use of mouth rinses is the first line of treatment. Nonsurgical periodontal treatment (including oral hygiene instructions, scaling, root planing, and prophylaxis) is the conventional management approach for gingival enlargement but it may not always be sufficient when gingival enlargement is extensive and self-care is compromised (77).

The surgical approach for management of gingival enlargement was proposed. Although surgical treatment is considered to be invasive and may not be effective if self-care oral hygiene practices remain poor. There are some evidence that nonsurgical periodontal treatment with the adjunct use of laser therapy can be effective in the management of gingival health problems in patients with fixed orthodontic appliances. Diode laser gingivectomy was shown as a valuable tool for obtaining quicker and greater improvement in gingival health, suggesting its beneficial use for orthodontic patients especially when oral hygiene was not sufficient to achieve normal healthy gum (78).

6. Gingival recessions

OT is always accompanied by changes in tissues surrounding teeth. Tooth movement may promote GR formation as well as can possibly improve soft tissue conditions (79-84). As you know, GR appears to be a reason of root sensitivity, increased susceptibility to caries, tooth abrasion, following difficulties in maintenance of oral hygiene and unsatisfactory esthetics. The incidence of GR formation among the orthodontic patients was reported to be up to 10-12% (79, 85). The prevalence of gingival recession during orthodontic treatment was shown to increase during the treatment with the further raise during the long-term post-treatment period. Thus, 98.9% of the orthodontically treated patients had at least 1 recession, when 85.2% - had at least 1 recession 10 to 15 years post-treatment (86).

Studies dedicated to association between orthodontic treatment and the development of GR have conflicting results, probably because of the differences in the study designs and lengths of the observation periods. However, the systematic by Bollen et al confirms a positive association between gingival recessions and orthodontic treatment (87).

Although the etiology of gingival recessions remains unclear, several predisposing factors have been suggested. One of the main reasons for GR development is believed to be a continuous mechanical trauma by toothbrush (88, 89), but Matthews (90) and Rajapakse et al (91) suggested that there is no good evidence of direct link between toothbrushing and appearance of GR. When talking about tooth brushing, attention should be paid to the possible variables of the procedure, such as: brushing force, tooth-brushing technique, duration of toothbrushing, frequency of changing the toothbrush, brush /bristle hardness. During fixed orthodontic therapy the way of brushing has to be changed because to the obstacles presented by braces, elastics, arches and wires. In this way the efficacy of the brushing may decrease and the possibility of the trauma with the domestic hygiene aids increases. Periodontal inflammation encompasses gradual bone loss also might lead to apical gingival migration and root exposure. Difficulty with tooth brushing and plaque accumulation during orthodontic treatment and fixed retention thus can contribute to inflammation-related recession formation (92).

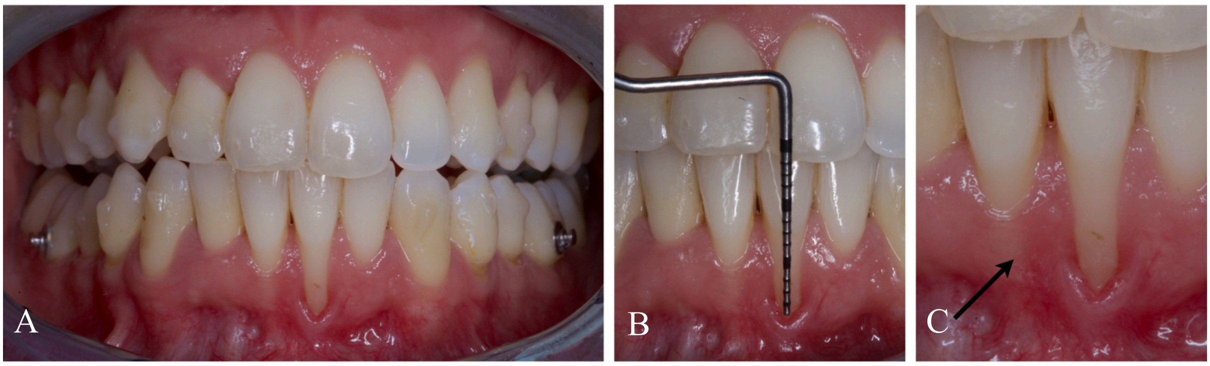

Fig 2. Intraoral view after orthodontic treatment provided by clear aligners.

A: Patient represents multiple gingival recessions in low and upper jaws; B: The most pronounced soft tissue defect shows 3.1: gingival recession of 5 mm and additional 3 mm of periodontal probing; tartar can be easily detected on the root surface of 3.1; C: The transparency of thin periodontal phenotype in the area makes it possible to observe subgingival tartar on the root surface of the neighbouring tooth.

(Courtesy of Dr. Michele Izzo)

Also several anatomical and morphological characteristics were suggested to play a role in GR formation. During OTM alveolar bone dehiscences may occur when tooth roots move through the alveolar cortical bone (93-95). More often this condition is performed in patients with a small alveolar process, thin buccal or lingual bone plates, eccentric position of teeth, basally extended maxillary sinus and progressive alveolar bone loss (96). However if the tooth is moved within the envelope of the alveolar bone, the risk of harmful side-effect on the marginal soft tissue is minimal (93,94). The direction of applied orthodontic forces may also have an impact on soft tissues. Some studies suggested that controlled proclination of mandibular incisors could be carried out in orthodontic patients without a risk of periodontal breakdown if good level of dental hygiene is provided (79, 85, 97, 98]. However according to the latest study (99) pro-inclination orthodontic movement may be significantly associated with a reduction of the keratinized tissue width. Facial tooth movement may result in decreased bucco-lingual tissue thickness and may reduce the height of the free gingiva and as consequence may lead to development of GR. A recent systematic review concluded that the direction of the tooth movement and the bucco-lingual thickness of the gingiva may play important roles in soft tissue alteration during orthodontic treatment (100).

Periodontal phenotype also has been suggested to be an important factor in GR development. It includes gingival thickness and keratinized tissue width, bone morphotype and tooth dimension (101). Patients with thin phenotype have significantly more chances to develop GR during orthodontic treatment than those ones with the thick one (99).

A strong correlation was found between thin phenotype and pro-inclination orthodontic movement in terms of GR depth and keratinized tissue width. Thin periodontal phenotype and amount of attached gingiva were found to be significantly related to labial plate thickness, alveolar crest position and gingival architecture. This type of periodontium has decreased resistance to mechanical stress or inflammation and may correlate with development of GR (80, 97, 102, 103). An accurate evaluation of gingival thickness before to start OTM is recommended (99). In clinical situations where tooth movement is planned in the areas with < 2 mm of keratinised mucosa, it was suggested to apply a preventive gingival augmentation before the initiation of orthodontic treatment to reduce the risk of gingival alteration during the orthodontic stage.

Every clinical case may include a combination of different predisposing factors that can affect the treatment outcome, therefore it is important to evaluate risk factors while planning orthodontic treatment in order to avoid or prevent GR formation during orthodontic therapy.

7. OT of periodontally affected patients

Today many adult patients are actively seeking orthodontic treatment as an adjunctive treatment to facilitate other dental treatment (104). It is well-documented that there is a high prevalence of periodontal disease among adults of 30 years of age or more (105). It is expected that orthodontists will treat patients affected by periodontal disease more frequently; the number of adult patients with periodontal problems attending orthodontic practices keeps increasing (106).

More than a half of periodontally affected patients suffer from pathological tooth migration (107). There are several etiologic factors may contribute to pathological tooth migration: periodontal attachment loss, pressure from inflamed tissues, occlusal factors, oral habits such as tongue thrusting and bruxism, loss of teeth without replacement, gingival enlargement, periodontal inflammation and eruptive forces. Due to the inflammation of the supporting tissues and a tooth loss, in the presence of reduced periodontal support under occlusal overloading, patients may develop teeth extrusion, rotation, teeth flaring or diastema formation.

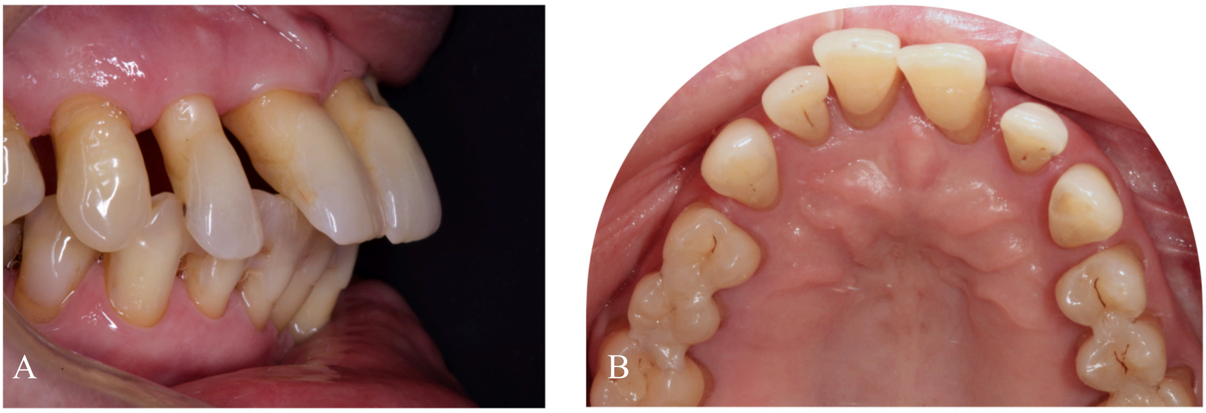

Fig. 3. A, B: Intraoral view of the denture in patient with pronounced periodontal disease. Teeth flaring was developed during the periodontal inflammation progression.

Fig. 4. Clinical case 1.

A Patient successfully treated with interdisciplinary approach. Before the treatment patient presented with periodontal disease Stage 4 Grade B.

A, D: Smile of the patient before and after perio-orto-implant dental rehabilitation.

B, E: Intraoral front view before and after the treatment.

C, F: Intraoral lateral left side in occlusion. Before and after treatment.

Fig. 5. Surgical phase of periodontal therapy. The infrabony defect is filled with Enamel Matrix Derivative (EMD) and deproteinised bovine bone (DBBM). Preoperative examination. A: the loss of clinical attachment at the mesial side of 23. Intraopertive steps. B: Visualization of the infraosseus component. Access to the infrabony defect realised by the single flap approach (SFA). C: biomaterials placement D: wound closure. Re-evaluation 6 month after the intervention. E: soft tissue healing 6 month after surgery. F: 12 month after periodontal regenerative surgery. G: Apical intraoral Rx before the surgery H: one year after full dental rehabilitation. (Courtesy of prof. Giulio Rasperini)

Nonetheless some grade of pathological tooth migration may be self-corrected after elimination of the inflammation. Non-surgical or/and surgical periodontal therapy may lead to the so-called «reactive repositioning» - the movement leading to spontaneous teeth realignment (108-110).

Our knowledge of the extent of spontaneous repositioning of pathologically migrated teeth that we expect after periodontal therapy is limited. Gaumet et. al. reported a full diastema closure in 50% of cases, demonstrating the correspondence between the gap size and repositioning: the bigger is the gap - the less chances of spontaneous realignment after periodontal therapy (111). When small to moderate diastema were considered, the frequency of complete repositioning appeared to be up to 78%. Authors stated, that if a recently formed diastema of anterior teeth is associated with periodontal disease was ≤1 mm in dimension, the closure was highly predictable. In addition, in 20% of patients experienced a further improvement of the teeth position after surgical phase of periodontal therapy.

However, in another 50% of cases, to align teeth after periodontal treatment patient need to undergo orthodontic treatment. Prior to OT, periodontal inflammation must be eliminated, and the orthodontist must make sure the patient is capable of performing adequate oral hygiene care. Also, it is better to inform the patient before the beginning of OT, that non-adherence to the oral hygiene protocol can result in high risks of periodontal tissue damage, so the discontinuation of the orthodontic treatment should be expected (112). On the other hand, if patient presents with reduced, but healthy periodontium, without infra bony defects and has a good level of dental hygiene - it is safe to perform the treatment both on fixed and
More attention should be paid when planning periodontally compromised patients in which periodontal inflammation resulted in vertical bony defects.

Orthodontic extrusion may lead to clinical and radiographic improvements of infrabony defects. This type of movement may provide levelling of isolated infrabony defects, repositioning of the gingival margin, and increased amount of attached gingiva and bone (113, 114). Forced orthodontic eruption may be combined also with periodontal regenerative procedures, resulting in soft and hard periodontal tissue improvements (115). Teeth can be moved into an alveolar ridge area previously augmented with biomaterials, as we mentioned above, talking about GTR used for prevention of GI formation. Periodontal regeneration before orthodontic therapy aims to prevent a down-growth of the junctional epithelium that may appear during tooth movements (115, 116). Periodontal regeneration may be combined with any kind of orthodontic tooth movement: extrusion, intrusion and sagittal tooth movements and in some cases may improve the prognosis of the teeth with furcation involvement (117, 118). There are few studies describing orthodontic treatment after periodontal regeneration procedures (119-123) and only one study reporting a long-term result 10 years after combined GTR-ortho treatment (121). In all mentioned publications authors stated that this interdisciplinary approach is valid and safe, when periodontal infection is under control and low continuous mechanical forces are applied.

The question of when the orthodontic phase should be initiated is still open. To the date, the optimal time to begin orthodontic movement after GTR has never been analyzed in a randomized clinical controlled study. Different researchers describe the initiation of orthodontic treatment from 10 days to 1 year following regeneration surgery (119-126). In presence of limited scientific evidence it was suggested to wait at least 6 months after the completion of periodontal therapy with regenerated materials in order to obtain the movement in fully healed sites (127).

Given that biological and biomechanical conditions are different in periodontally compromised patients as compared to a patient with good periodontal health, due to the reduced periodontal ligament - crown-root length ratio is increased, and the centre of resistance is moved apically. This means, that a greater moments during force application arise and it should be fully understood by orthodontists (119). An interdisciplinary periodontal-orthodontic treatment could be beneficial even in a case that seems hopeless, but these patients should be seen frequently for periodontal maintenance and minimal orthodontic forces should be applied. (128, 129). An appropriate force system should be used to treat adult patients with periodontal disease like segmental arches and skeletal anchorage (130). Also the retention phase after the active might be different from a conventional one. Often a permanent or long term retention is required. It should be kept in mind that orthodontic retainers, both removable and fixed, are potentially plaque retentive devices and as such have potential risk for periodontal breakdown (131, 132).

Clinical case 2.

Fig. 6-12. Periodontally affected patient, 22 y.o. Periodontitis Stage 3, Grade A.

Fig. 6 Patients ortomantamography before the treatment.

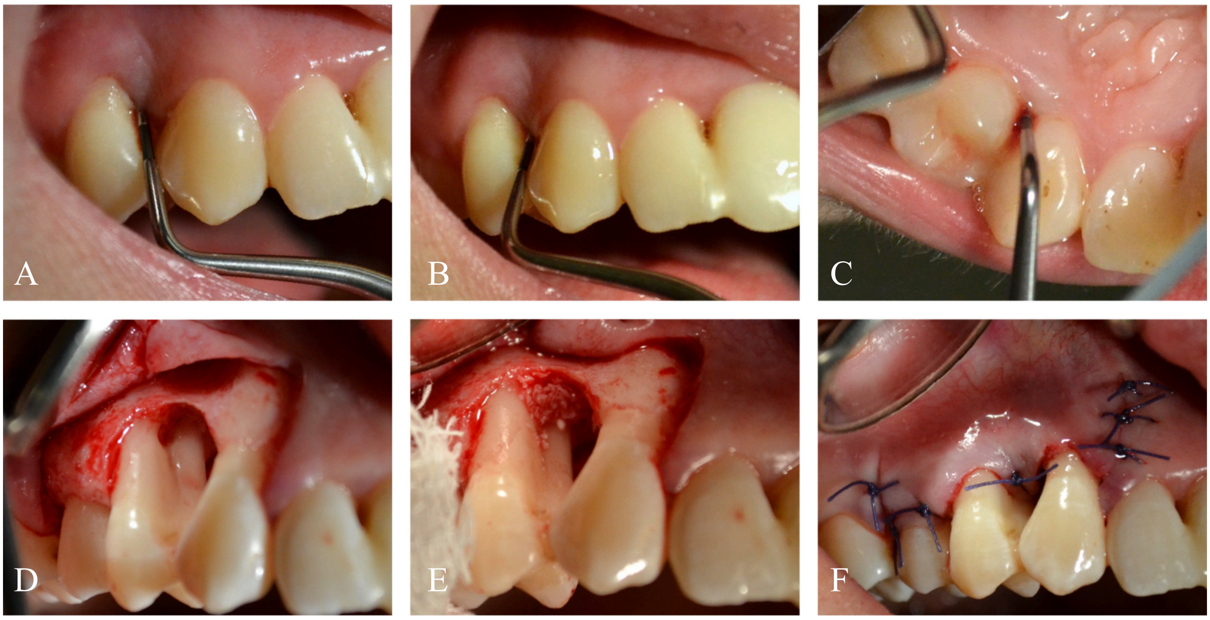

Fig. 7. Periodontal surgery of the pathological site with the deepest bony defect. A-C:intraoral view of probing; D-E: Intraoperative view of the defect. D: Infrabony defect; E: vertical component of the defect is filled with biomaterial; F: flap repositioning and sutures.

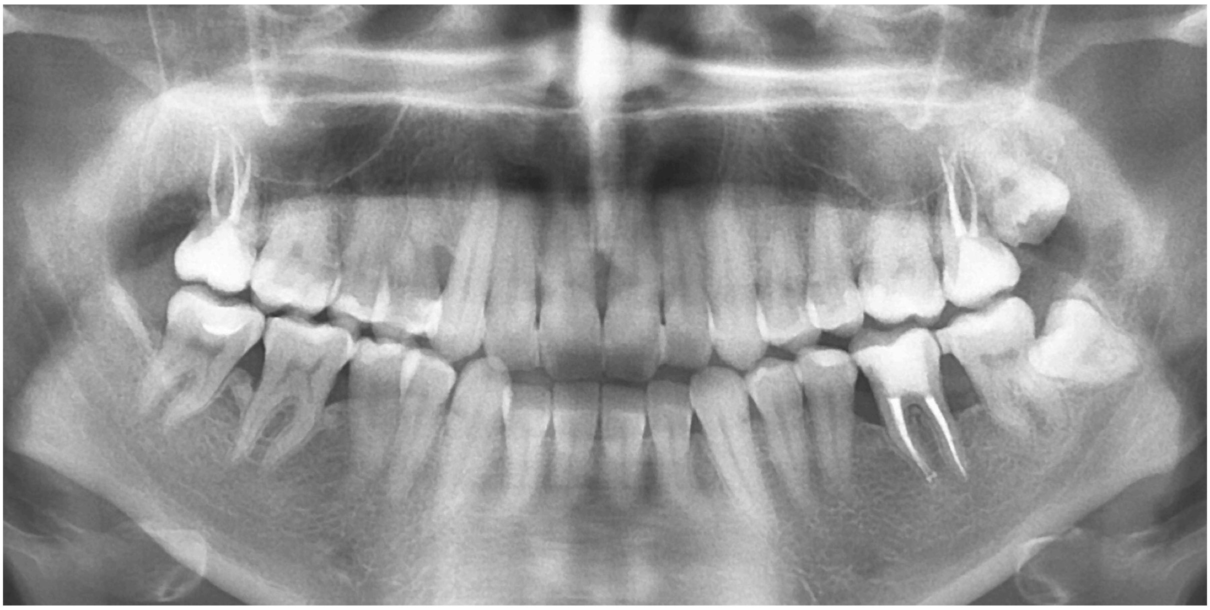

Fig. 8. Ortopantomography of the patient one year after periodontal surgeries.

Fig. 9. Intraoral view of the patient 1 year after periodontal surgery. Periodontal tissues are healthy and stable. No pathological probing, no bleeding on probing and a very good oral hygiene with the perfect adherence to the periodontal maintenance recall program.

Fig. 10. A, B: Intraoral view during orthodontic therapy; a fixed orthodontic appliance was used for OT with minor force application; during the treatment patient showed a good compliance and regularly showed up for periodontal maintenance visits every 3 month.
Some teeth have reduced but healthy periodontal support, however this condition can not be considered a contraindication for the orthodontic treatment.

Fig. 11. Intraoral view at the end of the ortho-perio treatment; after an active phase of the orthodontic therapy was placed a fixed retainer on the lingual surfaces of the lower and upper anterior teeth from 1.3 to 2.3 and from 3.3 to 4.3; this retainers do not impede domiciliar routine hygiene manoeuvres of the patient.

Fig. 12. Ortopantomography of the patient 1 year after completion of the orto-perio treatment.

8. Maintenance

All patients undergoing orthodontic treatment have difficulties to maintain a good level of oral hygiene, because orthodontic constructions and accessories may hinder conventional brushing and flossing. The same applies to post-orthodontic fixed retainers. Patient's compliance, motivation, and oral hygiene maintenance are well-known to have a significant impact on periodontal status. This treatment phase is also extremely important during orthodontic treatment for maintaining healthy periodontal condition. Oral hygiene education supplemented with positive motivation should be started at the initial stages of the treatment strategy in order to obtain predictable outcomes. Prior to treatment, the orthodontist must make sure the patient is capable of performing adequate oral hygiene measurements and aware, that in case of non-compliance and unsatisfactory level of domestic hygiene active orthodontic treatment will be postponed until satisfactory plaque control is achieved.

The first part of maintenance consists of a daily oral hygiene program that should be individually tailored for each patient. It must be clear and achievable.

There are many studies dedicated to different types of toothbrushes (133). All toothbrushes can be decided on two big groups: manual (special orthodontic settle, mono-settle, ionic) and powered (oscillating-rotating, ultrasonic, sonic). There is a Cochrane review from 2011, that was updated in 2014, based on huge number of observations, concluding that powered toothbrushes provide a significant benefit when compared with manual toothbrushes (134). However, if patient has never been familiar with powered tooth brush before, a separate visit dedicated to a brushing technique is recommended. In orthodontic patients oscillating-rotating toothbrushes has also performed higher effectiveness of in dental plaque removal and gingivitis reduction when compared to manual brushes (135, 136).

The additional use of chemical agents as adjuvants to mechanical biofilm control (brushing and flossing), may result in additional advantages compared to mechanical plaque control alone (137). For patients who might have a higher risk of periodontal breakdown, such as periodontally affected patients undergoing orthodontic treatment, the use of it additional oral antiseptics is recommended. Chlorhexidine is one of the most studied oral antiseptics. Mouthwashes containing chlorhexidine (CHX) shows high anti-plaque and anti-gingivitis efficacy (mean reduction of 40% and 28%, respectively) (138). However long-term use of CHX-based products results in tooth and soft tissue staining. For this reason CHX contained mouthiness should be indicated only for short periods of time, especially in cases of periodontal inflammation. Among the daily-use agents, essential oils are recommended as the first option, because of their proved efficacy (139, 140).

During orthodontic treatment of periodontally healthy subjects, clinical periodontal evaluation (including periodontal charting) should be performed every 6 months. A finding of pathological pocket will require further radiographic evaluation and referral to a periodontist. While treating orthodontically periodontally affected patients, periodontal recall appointments and professional hygiene maintenance should be performed with shorter intervals, once every 3 months during active OT, and they are essential to be continued after completion of orthodontic treatment. In other words periodontal maintenance should be provided from the beginning of periodontal therapy, through all the steps of periodontal treatment, it should be even more closely monitored during orthodontic treatment and later should be continued throughout the life-time of the patient.

II Periodontally accelerated orthodontics

In order to shorten the duration of active orthodontic therapy was proposed the use of orthodontic teeth movement acceleration which is based on stimulation of local bone metabolism by different means. Acceleration methods may be divided on three groups: surgically assisted, drug-induced, and physically/mechanically stimulated.

1. Non-surgical methods of acceleration of orthodontic tooth movement

Drug-induced acceleration

The approach of local drug-administration is based on the ability influence bone remodelling activity. Such substances as prostaglandins, corticosteroid hormones, vitamin D3, parathyroid hormone, thyroxin and some others were proposed for this purpose (141, 142).

Keeping in mind, that these drugs may have systemic effects on body metabolism, therefore they are difficult to apply for orthodontic tooth movement and to control their absorption in the blood stream. For this reason daily systemic administration or daily local injection should be provided to obtain the constant concentration in the interested area. Repetitive frequent injections may evoke fear in patients and cause problems in medical treatment. For these reasons, routine clinical use of drug-induced orthodontic acceleration still requires further investigations, to determine the correct dosage, frequency of administration and, especially, the possible local and side effects of its long term use (143).

All the drugs reviewed have therapeutic effects as well as side effects that influence the cells targeted by orthodontic forces. The value of a thorough medical history is increasingly significant as young and old alike are exposed to a greater range of therapeutic agents. Therefore, it is imperative that the orthodontists need to pay attention to drug consumption and history of each and every patient, before and during the course of orthodontic treatment, so that the best treatment strategy (including force control and appointment intervals) can be selected for each case. Acetaminophen, which does not have significant influence on the rate of tooth movement, can be recommended for controlling pain during orthodontic treatment.

Physically/mechanically stimulated

Several non-surgical adjuncts were suggested to promote tooth movement via physical or mechanical stimuli, electric current, pulsed electromagnetic fields and photobiomodulation. Low Intensity Pulsed Ultrasound (LIPUS) is widely used in the field of medicine for diagnostic and therapeutic purpose. Since LIPUS stimulation is being utilised effectively as therapeutic modality for bone regeneration and fracture healing it was suggested that it may accelerate bone remodelling during orthodontic treatment. The same was suggested for electric field application (144). However, up to date, electric and pulsed electromagnetic field has no robust evidence to be an effective modality for OTM acceleration (145). Low level laser therapy (LLLT) is also known as photobiomodulation or biostimulation, that involves the use of near infrared or low levels of red light LLLT, represents another adjuvant therapy for orthodontic treatment (146, 147). There are some evidence of accelerating effect during OT, however there is no set protocol found for the procedures yet, and more studies with a focus on the characteristics of used laser on a high number of teeth are necessary to confirm positive results (148, 149). Moreover, the use of physically induced orthodontic acceleration is limited because of the necessity of performance by disciplined clinicians. The use of supplemental vibrational force has been advocated as a method of speeding up orthodontic tooth movement. This involves the application of low-level vibration directly to the dentition as it is subjected to orthodontic force (150). The basic underlying principle of this type of acceleration is the ability of alveolar bone to respond with remodelling after the application of external force. However prospective randomized studies were not able to support its efficacy (151, 152).

Despite the number of papers dedicated to the topic, it is impossible to determine if there is a positive effect of non-surgical adjunctive interventions, because of the limited clinical data concerning the effectiveness of non-surgical interventions (153).

2. Surgical-assisted methods of acceleration of orthodontic tooth movement

Surgically assisted orthodontic tooth movement has been used since the 1800s. Corticotomy-facilitated tooth movement was first described by L.C. Bryan in 1893. However it was first introduced in 1959 by Kole and his «bony-block technique» in 1959. He believed that the main resistance to tooth movement was due to the cortical bone and by disrupting its integrity, tooth movement could be accelerated. Kole's procedure involves the reflection of full thickness flaps to expose buccal and lingual alveolar bone, followed by interdental cuts through the cortical bone and barely penetrating the medullary bone.

Since that time various surgical techniques were proposed and they can be divided on corticotomy-assisted orthodontic treatment (CAOT) and periodontally accelerated osteogenic orthodontics (PAOO) developed a patented by Wilcko brothers. Both approaches are based on "regional acceleratory phenomenon" (RAP) described by Frost in 1983, which is characterised by increased bone turnover in response to noxious stimuli (154). It was shown to cause 10-50 times faster bone turnover in the affected zone (155). Apoptotic osteocytes in damaged area produce signals that attract vital osteocytes and make them migrate to the damaged area surrounding the focal injury. Later this population of osteocytes produces cytokines, including RANKL and VEGF, that induce the recruitment of osteoclasts, that, in turn, provide bone resorption that is necessary for tooth movement (156, 157).

The aim of CAOT was to create an extent area of corticotomy cuts to physically disrupt the continuity of the cortical plates and induce the RAP in the particular area to the jaw bone. Meanwhile PAOO has a different concept: this technique is similar to conventional corticotomy except that selective decortication in the form of lines and dots is performed on the surface of the cortical plate in the area of the teeth that are to be moved. In addition, a selective decortications is supplemented by bone augmentation i.e it allows to change the volume of the alveolar bone.

Later, when new piezosurgical instruments were introduced into the dental world they were successfully applied also for corticotomies, thats how the term «corticision» was born. Corticision may be translated as a «cortical bone incision». In its turn, when piezosurgical instruments are applied for the procedure- it is called «piezocision».

Clinical case 3. Patient with sceletal malocclusion Class III has refused maxillo-facial surgery. In the upper jaw after the fixed orthodontic appliance placement was performed a piezo-corticotomy. In the lower jaw the treatment plan included two premolars' extraction and the following orthodontic therapy.

A: Intraoral frontal view after upper jaw bonding.;

B: Occlusal aspect of the upper jaw anfter bonding;

C, D: intraoral intraoperative view after mucogingival flap reflection, when a piezo surgical corticotomy was performed. Before the surgery the arch wire was removed;

E: the flap is positioned and sutured back; the orthodontic arch wire is bonded selectively for the soft tissue healing period;

F: completed alignment of the upper jaw;

G: a five-strand twisted steel retainer was fixed on the palatal aspect of the anterior upper segment.

(Courtesy of Dr Andrea Podesta)

Minimally invasive techniques

Recently minimally invasive techniques without muco-periosteal flap elevation have been developed: piezocision and micro-osteoperforations (159, 160). Piezocision technique still requires the use of the microsurgical blade to make gingival incisions in order to create an access for piezoelectrical knife tip for the cortical alveolar bone cuts. It was first described in 2009 by Dibart who combined the advantage of grafting offered by PAOO of Wilcko and Wilcko with the flapless approach adopted by Park. The surgery is performed 1 week after the placement of the fixed orthodontic appliance under local anesthesia. Vertical interproximal incisions are made below the interdental papilla on the buccal aspect of the elements planned for major movement. The incisions are kept minimal except in the areas requiring bone grafting and go through the periosteum to the alveolar bone. Corticotomies are performed with a Piezoelectrical surgical knife (BS1 insert, Piezotome, Satelec Acteon Group, Merignac, France) through the gingival incision in the bone with a depth of 3 mm to pass the cortical plate. Where bone grafting is needed a small periosteal elevator is used to create a tunnel which will accomodate the bone graft. Suturing is recommended only in areas that receive graft material and is not necessary if only corticotomy is performed. Besides the advantages of this flapless approach there are also some adverse effects like a scar tissue formation on the mucosa, that can compromise the aesthetic outcomes of dental treatment.

Micro-osteoperforations represent a novel minimally invasive approach for OT acceleration. The microosteoperforation technique was developed based on previous animal studies showing that small and shallow perforations in alveolar bone increased the rate of tooth movement without the need for flaps, bone grafting, or suturing. Alveolar bone perforations activated the cytokine cascade and subsequently increased osteoclast activity, allowing for enhanced bone remodelling after orthodontic force application. In the area where accelerated bone turnover is needed, between the roots of neighbouring teeth 1-3 transgingival MOP's of the cortical bone are placed. The diameter of the tip is 1,6 mm and insertion of self-drilling tip may be provided up to 7 mm in deep, depending on the clinicians' decision. Insertion of self-drilling tip leads to direct bone injure and also creates an area of bone compression holding numerous microcracks in the alveolar bone tissue leading to RAP (160). During the normal skeletal function microcracks usually form as a result of the bone fatigue damage performing the important part of the normal bone remodelling (161). From advantages of this method there is no need in extra time after procedure for healing and orthodontic forces may be applied immediately after procedure.

To date there is a lack of evidence in changes of periodontal parameters and its efficacy in reducing of the total treatment time.

Alikhani et al. reported promising results in a study where canine retraction was performed with combination of MOPs, TADs and fixed orthodontic devices (1). He reported a higher rate of tooth movements in the test group and significant reduction. However more well-designed prospective studies are needed to draw a conclusion about minimally-invasive procedures for orthodontic acceleration.

III Clinical study

Introduction

Dentofacial appearance plays a big role in psychosocial well-being and self-esteem of people and was suggested to be the strongest motivating factor for orthodontic patients to begin the treatment (162). It was suggested that the most frequently mentioned factors that influence patients′ satisfaction with orthodontic treatment (OT) are: duration of the treatment and pain during the therapy (163). There is a high demand in decreasing of orthodontic treatment time among patients and orthodontists (164).

The reduction of treatment time may result not only in a higher appreciation from patients, preventing patients' burnout of cooperation, but also leads to a better clinical outcomes. As reported previously, OT provided by fixed or removable orthodontic appliances is able to affect the equilibrium of oral microflora and that the duration of orthodontic devices wearing may correlate with microbiological changes in oral cavity (165-167). During OT may be observed a decrease of colony forming units ratio (CFU aerobes/CFU anaerobes), accompanied by growing number of bacteria ascribed to the red and orange complexes (Socransky et.al.) performing a risk of periodontal disease occurrence (168-170). The incidence of white spot lesions is significantly associated with the duration of the OT (171). Furthermore it was claimed that external root resorption in teeth undergoing orthodontic displacement may depend on the duration of the mechanical forces' application (172). Thus, reduction of total time of the orthodontic appliances wearing may diminish the risk of harmful side effects on oral health.

Various techniques and methods for orthodontic treatment acceleration were invented and proposed. Still, the most effective and for this reason called «the gold standard» of accelerated orthodontics, is corticotomy. Technically, this procedure requires an extensive full-thickness flap elevation and a massive bone resection that can be considered as an extremely aggressive and invasive intervention leading to significant postoperative consequences. The acceptance of corticotomy-assisted orthodontics as a treatment option appeared to be low among orthodontic patients, probably because of the high anxiety rate of dental patients when they hear about invasive surgical dental treatment (173, 174). In consideration of all these facts cited above it is of interest of contemporary dentistry to explore minimally invasive approaches, that will be less aggressive, but still effective in terms of dental movement acceleration.

Aim and hypothesis

The aim of this project is to evaluate effects of the innovative micro-osteoperforations (MOPs) method of orthodontic treatment acceleration on periodontal tissues and particularly will provide an answer to the question «if it is possible to change the Invisalign trays every 3 days after applying MOPs (instead of changing trays every 7 days) without any detrimental effect on periodontium».

Null hypothesis: Periodontal parameters of teeth after orthodontic alignment in control and test group will not show statistically significant difference.

Materials and Methods

Study Design

This is a multicenter, parallel-arm, randomized controlled clinical trial.
The study was conducted in the following centres:

Department of Periodontal diseases, University of Milan, Italy;
Dental department of the Military hospital, Milan, Italy.

All procedures and materials in the present study were approved by the ethical committee at the University of Milan (Italy) on 16 December 2016.

Orthodontic patients with Class I and II malocclusion with diastema or with mild to moderate teeth irregularity (1-6 grade of Little's index) of the lower incisors in a permanent dentition, in absence of craniofacial anomalies were recruited. Study subjects were randomly allocated in one of the following groups: Control Group will receive conventional Invisalign treatment changing trays every 7 days, Test Group receiving MOPs and changing trays every 3 days.

Patient entry

Inclusion criteria for participants:

- Male and female with no systemic condition that generally precludes surgical therapy or that could influence the outcome of therapy
- Women not pregnant
- 18 < Age < 70
- Good oral hygiene with full mouth plaque score (FMPS) and full mouth bleeding score (FMBS) < 20% at baseline
- No untreated caries
- No history of orthodontic treatment
- Class I and II malocclusion with diastema or mild to moderate Little's index(1-6 mm), in a permanent dentition and without craniofacial anomalies
- No smoking

Exclusion criteria for participants:

- Systemic condition that generally precludes surgical therapy or that could influence the outcome of therapy (e.g. Diabetes with HbA1c > 7%, INR > 3 etc.)
- Non-compensated systemic disease
- Smoking
- Long-term use of antibiotics, phenytoin, cyclosporine, anti-inflammatory drugs, systemic corticosteroids, and calcium channel blockers
- Poor oral hygiene with full mouth plaque score (FMPS) and full mouth bleeding score (FMBS) > 20% at baseline
- Radiographic evidence of bone loss
- History of orthodontic treatment

Subject protection

Written, informed consent was obtained from each patient prior to performing any study procedure or assessment. Prior to enrolling a subject into this study, the Investigator explained the study protocol, procedures and objectives to the. When the subject understoods and was willing to participate in the clinical trial, he/she signed and date the IRB-approved Informed Consent Form (ICF). The ICF described the study and potential discomforts, risks and benefits of participating. One copy of the consent form was be provided to the subject, and one copy will was maintained with the subject's permanent medical records.

Randomization

Patients were randomly allocated to test or control groups.

A balanced random permuted block approach was used to prepare the randomisation tables in order to avoid unequal balance between the 2 onset groups. In order to reduce the chance of unfavourable splits between test and control teeth in terms of key prognostic factors, the randomisation process of accelerated and non-accelerated orthodontic treatment took into account the type of malalignment, gingival phenotype and gender.

Onset assignment was noted in the registration form that was sent to the central registrar. Randomization was performed after the patient enrolment into the study and after signing the informed consent. After receiving the number of the envelope from the central registrar the investigator opened the randomisation envelope and disclosed the patients' group assignment before the virtual orthodontic treatment planning.

Pretreatment phase

After randomization, during two weeks before the initiation of the orthodontic treatment a full mouth professional prophylaxis appointment was scheduled for each subject. Oscillating-rotating powered toothbrush with a pressure sensor and soft bristles were assigned to each participant to avoid influence of gingival trauma during manual brushing and diminish its possible influence on final gingival margin position. Patients were given a detailed hygiene instructions along with the brushing technique demonstration and motivation. Additionally during the treatment period, patients were enrolled in an oral hygiene maintenance program with monthly recall appointments. These visits included motivation, oral hygiene checks and teeth polishing with Glycine powder, if needed.

Orthodontic treatment

For diagnosis and treatment planning all subjects were examined clinically and radiographically (CBCT) before including into the study. Patients of the test group started the orthodontic treatment with Invisalign immediately after MOPs performance changing Invisalign trays every 3 days, while patients of the control group underwent conventional orthodontic treatment with Invisalign changing trays every 7 days.

Different types of orthodontic movements were performed according to the clinical need of realigning the dental arch.

Invisalign treatment provided a series of nearly invisible appliances, (Invisalign Align Technology, Santa Clara, Calif.), to progressively move teeth from their initial position to their final straightened position. Additionally, to avoid/reduce the need of refinement, the treatment plan was performed using ClearTPS®.

Clinical measurement (gingival phenotype, PPD, REC, FMPS and FMBS) were performed at baseline, every 10 aligners and after the end of active orthodontic treatment.

The clinician selected an appropriate fixed retention device after active therapy, in order to prevent relapse, to decrease tooth mobility and to improve chewing comfort.

MOP mapping and procedure

After Invisalign treatment plan optimising ClearTPS® provided so-called Propel map- an individual recommendations for the MOPs localisation and drilling depth. The experimental group received MOPs provided by Propel device (Propel Orthodontics, Milpitas, Calif) the same day of the first Invisalign tray application. Before receiving MOPs each patient in the test group was asked to rinse the mouth with chlorhexidine 0.20% for 2 minutes.

An isolation of the surgical field from saliva was provided with cotton rolls and cheek retractors. Placement of cheek retractors followed by an application of the pre-cut, non-woven sponge gauze strips with topical, containing 12,5% Lidocaine, on the soft tissue covering the affected gingiva and mucosa to be treated. Using a timer, the topical was left on the tissue for four minutes. After a thoroughly rinsing off the oral cavity with water the operator waited 10 minutes before placing the MOPs. If discomfort occurred during treatment, an injection of local anaesthetic (Articaine 4% with epinephrine 1:100,000; Ubistesin, 3M ESPE) was provided. Then 2-3 consecutive micro-osteoperforations were performed between teeth suggested to more rapid orthodontic movement. The device's tip was inserted into the tissues at least for 3 mm using the depth control tool.

After making MOPs a light pressure with a wet gauze for several minutes was provided to control the bleeding. No suturing was needed.

Non-steroidal anti-inflammatory drugs and antibiotics were prescribed.

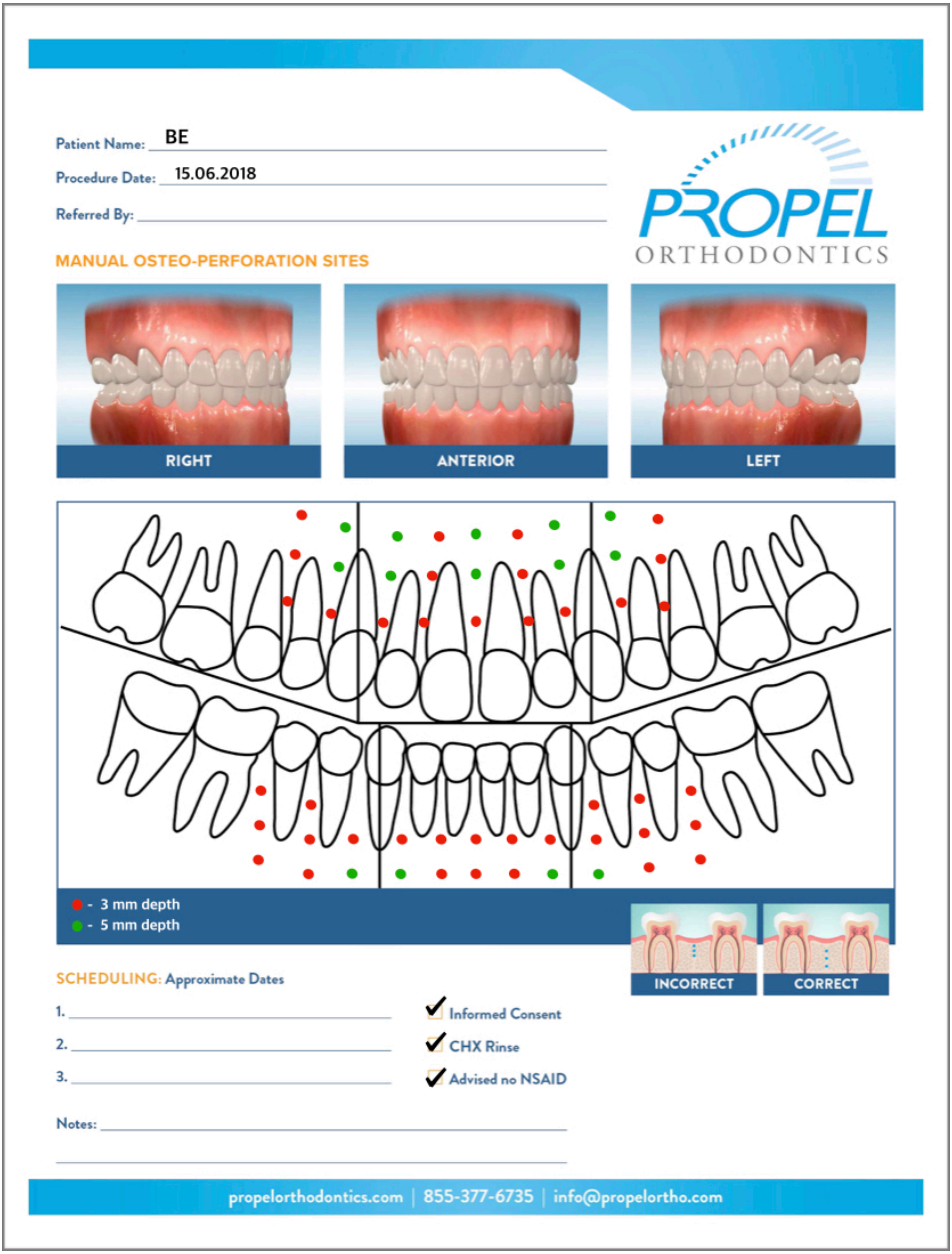

Fig.13. An example of the MOP map.

Measurements

On 30 of October 2017 there was a calibration meeting for examiner (the same for two clinical centres) and operators.

Clinical measurements are:

- Probing pocket depth (PPD): distance from the gingival margin to the bottom of the probing pocket. measured at 6 points for each tooth (3 vestibular and 3 palatal/ lingual).
- Gingival Recession (REC): distance from the CEJ to the gingival margin. Measured in the midline of the vestibular surface of each tooth.
- Full Mouth Plaque Score (FMPS)
- Full Mouth Bleeding Score (FMBS)
- Periodontal phenotype evaluation: Periodontal phenotype evaluation using HU-FRIEDY COLORVUE® BIOTYPE PROBE should be performed before the randomization. First, the White tip of the probe should be inserted in the gingival sulcus with < 30 g of pressure. If the color is visible, meaning if you can see the color through the gingival tissue, the phenotype is thin. If the white is not visible the Green probe should be applied in the same manner. If the color is visible the phenotype can be classified as medium thickness. If the Green is not visible through the gingival tissue, the Blue probe should be used. If blue is the only color visible through the gingival tissue, the phenotype is thick. If the blue tip is also not visible, the gingival tissue can be classified as very thick. Registered in the midline of the vestibular surface of each tooth.

PPD, REC, FMPS and FMBS, were recorded at baseline (T0), every 10 aligners (T10, T20, T30) and after the complete orthodontic treatment.

Additionally, after the completion of orthodontic treatment all patients received a VAS questionnaire of 100 mm

Subject code: _ I _ _ I _ _ _ **Iniziali del paziente _ _**

Questionario di valutazione del trattamento ortodontico

Mettere una "X" sulla linea

- Il trattamento ricevuto è stato doloroso?

Assolutamente no |——|——| Molto

- Il tempo dedicato alla terapia è stato adeguato?

Troppo breve |——|——| Troppo lungo

- Le piacerebbe ottenere lo stesso risultato nella metà del tempo?

No |——|——| Si

- Quanto sarebbe disposto a pagare di più per questa soluzione?

0 € |——|——| 2000 €

- E' contento del trattamento ricevuto?

Per niente soddisfacente |——|——| Pienamente soddisfacente

- Il trattamento proposto era in linea con le Sue aspettative?

Per niente soddisfacente |——|——| Pienamente soddisfacente

- Se tornasse indietro ripercorrerebbe il trattamento ricevuto?

No |——|——| Si

Consegnando questo questionario autorizzo l'utilizzo dei dati a scopi di ricerca_________________

Subject code: _ I _ _ I _ _ _ **Iniziali del paziente _ _**

Questionario di valutazione del trattamento ortodontico

Mettere una "X" sulla linea

- Il trattamento ricevuto è stato doloroso?

Assolutamente no |——|——| Molto

- Il tempo dedicato alla terapia è stato adeguato?

Troppo breve |——|——| Troppo lungo

- Quale sarebbe il suo grado di soddisfazione se ci avesse messo il doppio del tempo?

Per niente soddisfatto |——|——| Pienamente soddisfatto

- E' contento del trattamento ricevuto?

Per niente soddisfacente |——|——| Pienamente soddisfacente

- Il trattamento proposto era in linea con le Sue aspettative?

Per niente soddisfacente |——|——| Pienamente soddisfacente

- Se tornasse indietro ripercorrerebbe il trattamento ricevuto?

No |——|——| Si

Consegnando questo questionario autorizzo l'utilizzo dei dati a scopi di ricerca__________________

Statistics and data analyses

Sample size calculation

Assumptions:

- Acceptable max difference is +/- less than 1 mm (Delta) - DS = 1 mm
- Power 80% (z (1 - beta / 2) = z (0.90) = 1.28
- Z (1 - alpha) = z (0.975) = 1.96

Number of subjects per group = 2 x (1 ^ 2) / (1 ^ 2) x (1.96 + 1.28) ^ 2 = 21

Taking into account the drop-out rate (20%), it was sufficient to recruit at least 26 subjects for each group in this study.

To balance four periodontal phenotypes a total of 28 patients were recruited for each group (7 patients per phenotype in each group).

Confidence interval

We assumed that the average difference between the two treatment modalities of REC is zero. ES (difference) = square-root (1 x (1/21 + 1/21)) (we used the number of patients calculated above - 21).

IC 95% of difference: from - 0.6 to + 0.6, which is within the maximum acceptable difference of +/- 1 mm.

Assuming that the two treatments are equal, that the DS is the REC = 1 mm, a power of 80% (z = 1.28), and an error of the two-tailed alpha 0.05 (z = 1.96), would be sufficient 21 patients per group. Assuming conservatively that lost to follow-up can be 20%, it is expected to enrol 26 subjects per group. [38]

Data analysis

Data analysis was performed using JMP software (version 5.01a, SAS). Descriptive statistics of baseline characteristics were calculated and expressed as mean values ± standard deviation (SD).

Further, statistical analysis of qualitative and quantitative variables will be carried out by t-Student and X^2 tests, while Anova test will be used for data analyses in different timepoints. An α-error of 0.05 was accepted as a statistically significant difference.

Results

This manuscript reports *ad interim* results of the RCT.

In this prospective clinical study all subjects were selected from patients received in the Clinical centres mentioned above between June 2018 and September 2019 and were treated from September 2018 till the moment. A total of 10 patients were enrolled, however during the experimental period, 1 patient was lost because of an interruption of the orthodontic treatment, thus a total of 9 subjects (5 males and 4 females; mean age 30.6±10.4 years;) were included in the present study, distributed among two clinical centres as shown in the Table 1. Two patients represented with diastema and seven with teeth crowding. Five patients were assigned to the test group (4 males and 1 females, mean age 33.2±12.9 years) and four were in the control group (1 male and 3 females, 27.5±6.6 years). Table 2 reports the distribution of types of malocclusion and gender in study groups at baseline and the mean age for the test and control group.

After the MOP placement all treated sites healed uneventfully in all subjects assigned to the test group.

Given a low number of the patients recruited in this study at the moment, no inferential statistical tests may be performed.

FMPS and FMBS described for each patient for every time point from T0 to T30 are described in Table 3 and Table 5 respectively.

Mean values of FMPS and FMBS of experimental groups at different time points are summarized in Table 4 and Table 6 respectively. All patients presented with indices lower than 20%, performing acceptable level of patients compliance and from low to moderate risk for the periodontal breakdown.

Probing depth never exceeded 4 mm in both groups from T0 to T30 and only few sites were deeper than 3mm. Table 7 reflects Mean PD±SD for test and experimental groups.

Total amount of gingival recessions at all timepoints and their distribution are described in Table 8 and Table 9.

In two patients (1 from Clinical centre 1 and 1 from clinical centre 2) after uneventful healing of the mucosa and orthodontic forces application we observed multiple exostosis formation in the area of the MOPs' placement (Fig.14). Patients who experienced this unwanted effect reported some grade of discomfort, that diminished in time. During further two month of the accelerated orthodontic therapy following the study protocol patients experienced a graduate reduction of exostoses.

Fig. 14. Intraoral view of the patient with multiple exostoses. 2 weeks after MOP procedure.

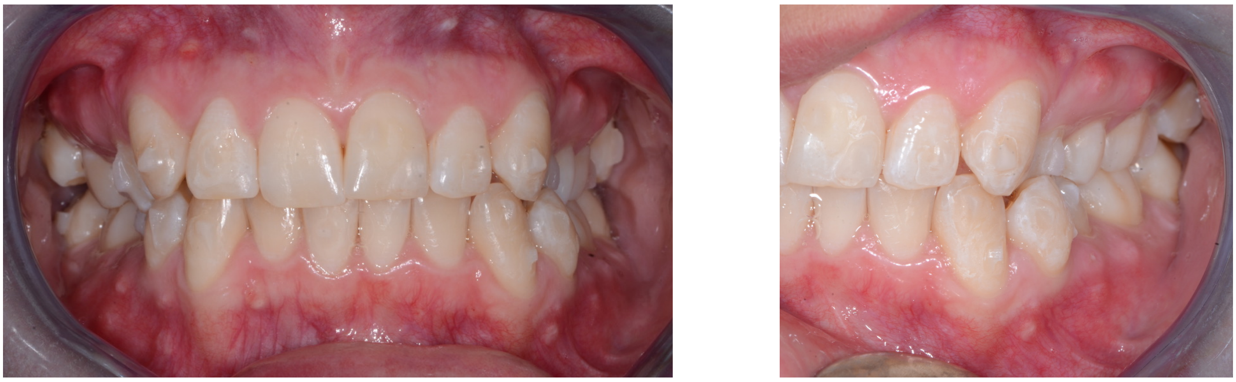

Table 1. Demographic data of subjects included into the study.

Patient	Experimental group Test (T)/Control C)	Clinical center	Gender (M/F)	Age	Type of malocclusion
1	T	2	M	34	Diastema
2	C	2	M	29	Crowding
3	T	2	M	53	Crowding
4	C	2	F	35	Crowding
5	C	2	F	19	Crowding
6	C	2	F	27	Crowding
7	T	2	M	34	Diastema
8	T	1	M	18	Crowding
9	T	1	F	27	Crowding

Table 2. Type of malocclusion and demographic data of groups.

	CONTROL		TEST	
MEAN AGE	33,2 ± 12,87		27,5 ± 6,6	
	count	proportion	count	proportion
F	3	75,%	1	20,%
M	1	25,%	4	80,%
TYPE OF MALOCCLUSION				
CROWDING	3	75,%	4	80,%
DIASTEMA	1	25,%	1	20,%

Table 3. FMBS of subjects at different timepoints of the study.

Patient	T0	T10	T20	T30
1	8	9,2	11	20
2	5	5	3	4,8
3	4	0	0	0,8
4	12	8	0	3
5	0	0	4	0
6	6	4	2	18
7	0,7	2	7	20
8	5	5	20	19
9	4	15	14	20

Table 4. Mean FMBS of experimental groups at different timepoints of the study.

	Test				Control			
	Mean (%)	SD (±)	min (%)	max (%)	Mean (%)	SD (±)	min (%)	max (%)
T0	5,8	4,49	0	12	3,93	2,3	0,7	6
T10	4,44	4,33	0	9,2	6,5	5,8	2	15
T20	3,6	4,5	0	11	10,75	7,89	2	18
T30	5,72	8,2	0	20	19,25	0,96	10	20

Table 5. FMPS of subjects at different timepoints of the study.

Patient	T0	T10	T20	T30
1	15	10	18	20
2	5	0	0	0
3	5	0	0	3,5
4	16	8	0	4
5	0	1	0	0
6	2	9	2	16
7	9	4	3	20
8	6	18	16	7
9	10	14	20	5

Table 6. Mean FMPS of experimental groups at different timepoints of the study.

	Test				Control			
	Mean (%)	SD (±)	min (%)	max (%)	Mean (%)	SD (±)	min (%)	max (%)
T0	8,2	6,98	0	16	6,75	3,59	2	10
T10	3,8	4,82	0	10	11,25	6,08	4	18
T20	3,6	8,05	0	18	10,25	9,11	2	20
T30	5,5	8,32	0	20	12	7,16	5	20

Table 7. MEAN PD for subjects in different time points.

	T0		T10		T20		T30	
Patient	Mean PD (mm)	SD (±)	Mean PD (mm)	SD (±)	Mean PD (mm)	SD (±)	Mean PD (mm)	SD (±)
1	1,40	0,76	1,59	0,67	1,68	0,67	2,07	0,62
2	1,40	0,70	1,40	0,91	1,67	0,82	1,93	0,73
3	1,51	0,59	1,47	0,60	1,41	0,55	1,09	0,31
4	2,42	0,50	1,80	0,58	1,73	0,63	1,54	0,62
5	1,15	0,36	1,49	0,58	1,61	0,61	1,63	0,61
6	1,35	0,51	1,73	0,65	1,42	0,52	1,67	0,65
7	1,65	0,69	1,54	0,65	1,56	0,70	1,93	0,73
8	1,49	0,60	1,65	0,62	1,96	0,84	2,08	0,69
9	1,35	0,58	1,46	0,60	1,58	0,60	1,89	1,09

Table 8. Counts of gingival recessions at different timepoints.

Patient No	T0	T10	T20	T30
1	3	3	3	3
2	13	14	13	14
3	4	4	2	3
4	0	0	0	0
5	0	0	0	0
Total count Test group	20	21	18	20
6	0	0	0	0
7	0	0	0	0
8	1	0	0	0
9	1	0	5	0
Total count Control group	2	0	5	0

Table 9. Gingival recession distribution in Test and Control group (signed in red).

tooth	T0	T10	T20	T30
17	1	1	2	1
16	2	2	3	2
14	1	1	1	1
13	1	1	1	1
23	1	1	1	1
24	-	-	1	-
25	1	1	1	1
26	3	2	1	3
27	2	2	2	2
36	1	-	1	1
35	1 | 5	1	1	
34	-	-	1	-
33	-	1	1	-
41	-	-	1	-
42	1	1	1	1
43	-	1	1	-
44	-	-	1	-
46	-	-	1	-

Fig. 15. Example of the patient treated as test.

A-E: Intraoral view of the dental arches before orthodontic treatment. A- Right side, the intraoral view of the occlusion. B- Frontal view in occlusion. c- Left side, the intraoral view of the occlusion. D- Occlusal view of the upper dental arch. E- Occlusal view of the lower dental arch.

F-H: Micro-osteoperforation procedure. F, H- MOPs' placement. The tip of the Propel device is set up on 5 mm depth.

G- Intraoral frontal view immediately after procedure, with clear aligners on place.

Fig. 16. During orthodontic treatment.

Intraoral view of the dental arches after first 20 aligners changed every 3 days. A- Right side, the intraoral view of the occlusion. B- Frontal view in occlusion. C- Left side, the intraoral view of the occlusion. D- Occlusal view of the upper dental arch. E- Occlusal view of the lower dental arch.

Fig. 17. After orthodontic treatment.

Intraoral view of the dental arches after periodontally accelerated orthodontic treatment.

A- Right side, the intraoral view of the occlusion. B- Frontal view in occlusion. C- Left side, the intraoral view of the occlusion. D- Occlusal view of the lower dental arch. E- Occlusal view of the upper dental arch.

Discussion

The first publications about micro-osteoperforations is dated by 2010 and the majority of the papers dedicated to the topic were published from 2016 to 2019, it represents an area of interest for contemporary dentistry and may be legitimately called a «novel technique». The results of preclinical studies suggest that MOPs may activate cell proliferation and apoptosis of periodontal ligament cells leading to tooth movement acceleration and increases the number of osteoclasts in effected area (175-177). Cramer et al in his study on beagle dogs stated that MOPs placed 3 mm away from teeth do not increase tooth movements which is in line with the results of van Gemert et al, who determined that the principal effects extend only ~1.5 mm from the MOP site (178,179). Another study showed a 1.86-fold increase in the rate of tooth movement after MOP placement in rats, without increased risk for root resorption (177).

Meanwhile results from clinical studies remain controversial. There are evidence of a significant increase of the rate of the tooth movement, a significant raise of the levels of inflammatory markers and a significant reduction of the overall treatment time with no harm for periodontal health (160,180-182). However, another author did not find the ability of MOPs to accelerate the rate of canine retraction, but claimed that it seemed to facilitate root movement. With respect to the treatment time, the canine retraction is the most time consuming phase of the OT and the study design based on acceleration of the canine retraction after extraction of the premolars is the most prevalent in the literature. The only study that did not use canine retraction to assess the ability of MOPs to accelerate tooth movement - is the study of Bansal et al. who applied MOPs for mandibular anterior teeth alignment (182) and reported that the completion of cases in experimental group was 43.93% times faster than in the control group. In our study we focused on clinical cases with mild malocclusions (crowding and diastema), because we believe that orthodontic patient who do not require canine retraction in their treatment may benefit from periodontal assisted acceleration. At the same time premolars' extraction prior to canine retraction may represent a source of bias while evaluating orthodontic acceleration. Since the trauma from tooth extraction leads to the inflammatory markers activity elevation, it may result in additional tooth movement acceleration. In the current study only non-extraction cases were treated and this possible interference was minimised to zero.

It is well known that the timing of treatment and the age of patients have a significant role in orthodontic treatment. To reduce the influence of age on the rate of tooth movement, we chose adults between 18 and 55 years. In this age range, the development is almost or completely finished (183).

Gender of the patients is another factor that affects the treatment results due to the hormone differences in males and females. Similar to most of the studies, our study could not decrease this problem due to the limitation in patients' numbers and it might be assigned to the limits of this research (184, 185). Also, it must be noted that at the moment test and control groups show an inverse gender distribution that is expected to become balanced further on.

This is the first study, where periodontally assisted orthodontic acceleration provided by MOPS is combined with clear-aligner treatment in contrast to the studies mentioned above, where fixed orthodontic appliances were used with or without TADs. Knowing about limited duration of RAP, we found beneficial the possibility of virtual planning and treatment progress modelling with MOPs and clear aligners. Furthermore, clear aligners helped to avoid occlusion interferences that once were shown to affect significantly the rate of tooth movements (186). To rule out the effect of occlusion in this study, we also selected patients with similar severities of malocclusion, as was mentioned above. Moreover, being a removable construction, clear aligners provide some advantages over fixed appliances such as major aesthetics, comfort, and convenience for oral hygiene, representing less risks for periodontal health (187-190).

Another argument that has been always studied along with any type of surgical tooth movement acceleration - an external root resorption. External apical root resorption (EARR) is a frequent, unpredictable, and unavoidable clinical complication secondary to orthodontic tooth movement mediated by odontoclasts/cementoclasts originating from circulating precursor cells in the periodontal ligament. Due to the accelerated bone and periodontal ligament turnover after MOPs application there is always a question if some root damage can appear. At the present time it is almost impossible to establish any reliable estimate of the incidence, prevalence, or degree of severity of orthodontically induced apical external root resorption. A number of studies showed no significant difference in root resorption between conventionally treated and rapidly moved teeth (182, 191), however several studies showed a greater extent of root resorption associated with micro-osteoperforations. Still, the clinical significance of this findings is debatable.

Dental hygiene is crucial for maintaining periodontal health. Preexisted periodontal inflammation after orthodontic force application may cause a periodontal attachment loss and might have an impact on the rate of the tooth movement. Therefore we payed a great attention to this part of this study. Only periodontally healthy patients, with a good compliance were selected. Furthermore, the oral hygiene for all subjects included into the study was standardised. Patients were provided with an oscillating rotating toothbrush with a built-in pressure-sensor, soft bristle heads, and a strict hygiene instructions in order to obtain a good plaque control and eliminate the risk of the soft tissue trauma. Since gingival recession has a multifactorial nature, we strived to eliminate any external effect potentially contributing to the gingival recession formation.

Finally, patient's satisfaction and discomfort were investigated in a number of studies. From moderate pain that rapidly faded away after 1 week to no discomfort was reported by patients in different studies (160, 191, 192-194). Level of satisfaction was high, which is consistent with the feedbacks of the patients from the test group of the current study. A significant percentage of patients were willing to repeat the procedure and recommend it to others (192).

Multiple exostoses formation occurred during orthodontic treatment following MOPs' placement were never described before in the literature dedicated to the topic. Agrawal et. al. in her study reported a significant increase of the buccal bone thickness after performing MOPs. These events are probably legated to the RAP amplified by orthodontic force application. Further investigations are necessary to confirm these findings (193).

At the moment the number of patients is limited and the data showed above represent a preliminary results. Since only one patient has completed the orthodontic treatment it is early now to draw the conclusions. This study should be extended further to increase the sample size and the follow-up period.

Conclusions and recommendation for future research.

MOPs can be considered as a new minimally invasive, easy-to-use and efficient technique for acceleration of orthodontic tooth movement with improved patient acceptance. If the oral hygiene is kept under control, and timing of the tooth acceleration after procedure is respected- MOPs' do not lead to the periodontal breakdown. It is important to investigate if there is a correlation between the incidence of gingival recession and periodontal phenotype features in orthodontic patients treated with or without periodontally assisted acceleration, therefore, it is necessary to augment the number of participants in order to validate the statistics. Further studies are required to evaluate the effect of different numbers of MOPs on the rate of tooth movements and to find the ideal timing and frequency of the MOP application in order to achieve optimum tooth movement acceleration.

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