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Review

MEIGE SYNDROME: A NARRATIVE REVIEW

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ABSTRACT

A particular combination of oromandibular dystonia combined with blepharospasm is known as Meige's syndrome. In order to avoid any misunderstandings, diagnosis is primarily clinical and necessitates a high index of suspicion. Striatal dopaminergic predominance is the most frequently accepted theory because the exact mechanism is unknown. Unfortunately, there is no cure, but injections of botulinum toxin and other medication classes, including anticholinergics, tetrabenazine, baclofen, and benzodiazepines, provide relief.

KEYWORDS: *Meige syndrome, spasm, dystonia, eye, muscle*

INTRODUCTION

Meige syndrome is one of the focused dystonic movement diseases, along with oromandibular dystonia and blepharospasm. Dystonia is characterized by abnormal, involuntary postures or motions of the body brought on by constant muscle contractions. Typically, neurological and medical issues are to blame. Meige syndrome comes under rare neurological movement disease characterized by uncontrollable, frequently startling contractions of the jaw and tongue muscles. French neurologist Henry Meige discovered aberrant midline facial muscle contractions in some patients in the early 20th century. He first referred to this as "spasm facial median." The clinical manifestation in the jaw and also oropharynx muscles of these patients was likewise comparable. In order to describe individuals with facial muscular spasms, notably blepharospasm and dystonia of the oromandibular muscles, Dr. George Paulson originally coined the name "Meige syndrome" in 1972 (1).

Meige's syndrome often manifests between the ages of 30 and 70. Women experience it twice as frequently as males. The typical symptoms include difficulty opening the mouth, tightness of the lips, jaw deviation, and teeth grinding. Some potential symptoms are blepharospasm, sensitivity to light, excessive squinting, and a higher blink rate. The masseter, temporalis, and platysma muscles are frequently involved. The actions of talking, chewing, and biting can exacerbate Meige's syndrome symptoms. Gum chewing, applying pressure to the chin, and sleeping help to relieve it. Numerous theories have been put up for its possible causes because the specific aetiology is unknown. The theory pointing to

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dopaminergic and cholinergic hyperactivity is the most frequently recognized, aside from idiopathic Meige's syndrome. The aetiology has also been linked to a defect in the basal ganglia region (2).

Typical and more recent atypical antipsychotics have been observed to cause acute and tardive Meige's syndrome, which promptly improves with antipsychotic withdrawal. Several pharmacological studies have proposed central dopaminergic predominance as a potential biochemical cause of Meige's illness. The finding further corroborates that it becomes better when dopamine-depleting medication tetrabenazine is used (3).

Meige's syndrome cannot be cured by any medical procedure. The most beneficial treatment to date is considered to be botulinum injections into the facial muscles. Other successful treatment approaches for Meige's syndrome include deep brain stimulation of the *globus pallidus internus* and electroconvulsive therapy. Only oral medications are utilised to treat most patients, and Meige's syndrome is often treated with various medications (4).

Epidemiology

The impacts of race and ethnicity have been implicated in the significant variation in incidence rates of segmental dystonia and blepharospasm. The epidemiology of primary dystonia from 14 chosen studies was examined by Defazio and colleagues. Segmental dystonia was present in 2-20% of people. Defazio et al. estimated that Segmental dystonia had a crude frequency of 59 per million (5).

In another Italian research focusing on 9 blepharospasm patients. The average age of onset for Meige's syndrome is in the sixth decade of life, and age seems to have been an independent risk factor in its development. Meige's syndrome affects more women than men, with a roughly 2:1 female-to-male ratio (6).

Marsden also noted that in all three forms of craniocervical dystonia, women were affected more frequently than men (7).

In research on 100 patients having blepharospasm and oro-facial dystonia, Jankovic and Orman showed a sizable woman:man ratio (women 62, men 38) (8). In addition, according to Duane (9), some oestrogen receptors that affect involuntary motor function put women at higher risk than men (10).

Pathophysiology

As previously mentioned, the most widely recognized theory for the pathophysiology of Meige syndrome is dopaminergic and cholinergic hyperactivity. However, it could also be brought on by the cortex's inhibitory neurons' (such as GABAergic neurons') diminished activity. Additionally, the patient is more susceptible to craniocervical dystonia due to a number of environmental and genetic variables. Positron emission tomography scans of the patients have revealed reduced blood flow towards the sensorimotor area in relation to lower facial vibrations, leading some researchers to hypothesize that these individuals have faulty sensorimotor functioning. In patients with Meige syndrome with isolated blepharospasm, silent "functional magnetic resonance imaging" has revealed reduced activation of the primary motor cortex, including the premotor cortex in the mouth region. It could be brought on by aberrant basal ganglia control over cranial nerve nuclei inside the brain stem. Individuals with craniocervical dystonia had reduced grey matter volume in the insular cortex, cerebellum, superior frontal gyrus, and calcarine fissure, as per brain imaging (11).

Additionally, genetics plays a role in the etiopathogenesis of focal dystonias. For example, the torsinA gene mutation seems to disrupt the flow of vesicles in and out of the nucleus at the cellular level. Dysregulation of transcription is the result of this. Other main focal dystonias are caused by a similar mechanism, such as CNS transcriptional factors (TAF1, THAP1). Studies using animal and human models provide evidence for the above-mentioned genetic abnormalities that may cause the neural network's growth to be altered (3).

Emotional stress may have a role in 33 percent of Meige syndrome patients, whose main symptoms include anxiety, sadness, and sleep issues (11).

Evaluation

It can be challenging to know where to begin the diagnostic process. For many, it starts at a primary care physician's office, an urgent care facility, or an emergency hospital. A diagnosis can become evident through these encounters, or it might take specialized testing and referrals. Dystonic anatomical classification and diagnosis continue to be clinical tasks greatly impacted by training and experience. Task-specific, subtle, and intermittent regional engagement is possible.

Additionally, many muscles have origins and insertions in many anatomical areas of the body. As a result, it may be challenging for many patients to distinguish between "focal" and "segmental" dystonia. For example, electromyographically-demonstrable minor participation of the upper thoracic paraspinous muscle is common in cervical dystonia patients having retrocollis (12).

Patients referred for neuropsychiatric evaluations and who had not responded to therapy in 49 cases were recorded over the course of multiple sessions. In the context of blepharospasm and oromandibular dystonia, Meige's syndrome was identified. The orbicularis oculi muscles' short, chronic, repetitive contractions and the facial, masticatory, labial, lingual, and mandibular muscles' dystonic involuntary motions were carefully observed. In addition, patients' medical histories and records were examined to weed out those who had taken neuroleptic medications. None of these individuals had ever used an antipsychotic drug, a neuroleptic, or anything else that could have caused parkinsonism or other uncontrollable movements. The results of the neurological and neuropsychological tests showed no intellectual, sensory, or pyramidal impairments. Similar negative results came from neuroradiological investigations, particularly brain computed tomography and magnetic resonance imaging (13).

Meeting with a doctor frequently during the diagnosis procedure may be beneficial and required. During the diagnostic phase, a primary care physician and specialist may suggest treatment alternatives to control symptoms. Doctors can also connect the patient with regional assistance programs, mental health services, and research opportunities.

Treatment

Meige syndrome is treated according to the distinctive symptoms that each patient shows. Botulinum A toxin (Botox) injections and medication therapy are the two main forms of treatment. After a injection, about 70% of people report improved chewing, swallowing, and spasm reduction (4).

Oral medicines are used to treat about a third of those affected (drug therapy). Unfortunately, the effects of these pharmacological therapies are typically mild or disappointing and frequently transient. No medication seems to work consistently well. Meige syndrome has been treated with clonazepam, trihexyphenidyl, diazepam, and baclofen (14).

The Food and Drug Administration has authorized the orphan drug botulinum A toxin as a therapeutic for blepharospasm, which has since become the main type of treatment. First, small doses of botulinum toxin are injected into the orbicularis oculi, paralyzing these muscles for many months. The process then has to be repeated. Many patients with blepharospasm have found relief from botulinum toxin injections, although not everyone does (4).

In rare circumstances, making particular motions, often referred to as "sensory tricks", may help people feel better. These motions include speaking, chewing gum, conversing, and lightly caressing the chin or lips. As a result, spasms may be reduced, the range of motion may be improved, and unaffected muscles may be strengthened through speech and swallowing treatment (15).

Various interventions fall within the categories of medicine and surgery as part of treatment. The effectiveness of anticholinergics, dopamine antagonists, and GABA receptor agonists for such individuals can be clearly understood once the process of Meige syndrome is understood. Antiepileptics, such as valproic acid, and a number of psychotropic pharmaceuticals are among the other medications. The particular GABA receptor complex components omega-1 and omega-2 are where eszopiclone and nitrazepam operate to reduce eyelid spasming. Some case studies claim that zolpidem is beneficial in treating these people because it is very selective for a GABA omega-1 receptor (16).

Eyelid spasms brought on by prolonged use of psychoactive substances can intensify through the use of olanzapine; however, they have been recorded with other antipsychotics as well.

Deep brain stimulation

In rare patients with Meige syndrome, deep brain stimulation (DBS) can result in a consistent, long-lasting relief of the symptoms of dystonia. DBS entails implanting a computerized pulse generator. The pulse is a thin metal electrode connected to generator as the brain's pacemaker. During office visits, a doctor programs settings that can be changed to best regulate symptoms. DBS functions by obstructing aberrant brain activity patterns (17).

Botulinum toxin and other conventional therapy approaches have not successfully treated patients when deep brain stimulation of the *globus pallidus interna* is used instead. The positioning of electrodes should be planned conceptually since the *globus pallidus interna's* ventral and posterior regions correspond topographically to the face, whereas the cervicofacial region is more anterior (18).

CONCLUSION

Meige syndrome is a rare spasm disease that must be taken in mind by clinicians, must be recognized, and has a different treatment protocol to be delivered.

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Case Report

CONGENITALLY MISSING MAXILLARY LATERAL INCISORS. SPACE CLOSURE AND DIGITAL WORKFLOW: A CASE REPORT

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ABSTRACT

There are two major treatment approaches in cases with missing maxillary lateral incisors: space closure and space open. Closing the spaces by mesializing the posterior teeth and substituting missing incisors with cuspids seems to be the favorable treatment goal for both periodontal and aesthetic reasons. However, anterior anchorage control is crucial.

The aim of this case report was to describe the orthodontic treatment in a young patient with congenitally missing maxillary lateral incisors. The use of skeletal anchorage was necessary to gain space closure and to achieve proper occlusion and better esthetics.

KEYWORDS: *lateral incisors, missing tooth, dental implants*

INTRODUCTION

The upper lateral incisor is the third most common congenitally missing tooth after third molars and lower second premolars, representing about 20% of all tooth agenesis (1). The most appropriate treatment option depends on the type of malocclusion (2), alveolar process characteristics (3), patient's age, dental and gingival display at smile (4, 5), and color and shape of maxillary cuspids (6–8). The two main treatment options are space closure or space opening. Both options were challenging to achieve the desired aesthetic and functional outcomes.

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Single-tooth dental implants in the maxillary anterior region have the highest risk of esthetic complications from infra-positioning due to the continuing growth and eruption of the adjacent teeth (7).

Furthermore, when the canine substitution option is chosen, the cuspids need to be mesially moved to replace the missing lateral incisors. In this case, the anchorage is provided by the maxillary incisors (i.e., "pulling" mechanics) and/or the posterior teeth (i.e., "pushing" mechanics); when the sagittal position of the incisors needs to be maintained, mesial movement of the cuspids can be difficult. It follows that using skeletal anchorage provides effective treatment solutions (8).

Nowadays, there is no consensus among specialists regarding the optimal treatment plan for patients with missing upper lateral incisors, despite the guidelines by the Angle Society of Europe (9).

However, more authors suggest that proper orthodontic space closure is well accepted by patients and can be completed as soon as the dentition is complete. Thus, a second orthodontic treatment to upright tipped adjacent roots before insertion of a dental implant can be avoided. Moreover, canine substitution does not produce a higher risk for temporomandibular disorders (TMD) and is safer than prosthetic replacements from a periodontal standpoint (10–12). Finally, it can be accomplished with optimal aesthetic outcomes by performing tooth reshaping and positioning, bleaching, and direct and indirect final esthetic restorations.

In 2013, Ludwig and collaborators proposed a skeletally anchored mesializer, "Mesialslider," to meet anchorage requirements in canine substitution, trying to overcome the disadvantages of this treatment option (6, 7).

This paper aims to describe a clinical case of a young patient characterized by bilateral congenitally missing upper lateral incisors treated with Mesialslider and fixed orthodontic appliances.

CASE REPORT

Diagnosis

The patient, a 12-year-old girl, came with their parents to the Orthodontic Program with the father's complaint to improve her smile.

She was in late mixed dentition with a dento-skeletal class II tendency, a hypodivergent pattern, and unilateral crossbite.

The intraoral examination showed the canine erupted in the incisor lateral place along with the persistence of the deciduous one, 2.3 crossbite, lower midline deviation of 1.5 mm on the left, normal overjet and overbite, bilateral end-to-end molar relationship, and mild crowding in the lower arch.

The extraoral examination showed a symmetrical face with a slightly reduced lower third of the face and an irregular smile arch. The profile was slightly convex, with a lack of chin projection.

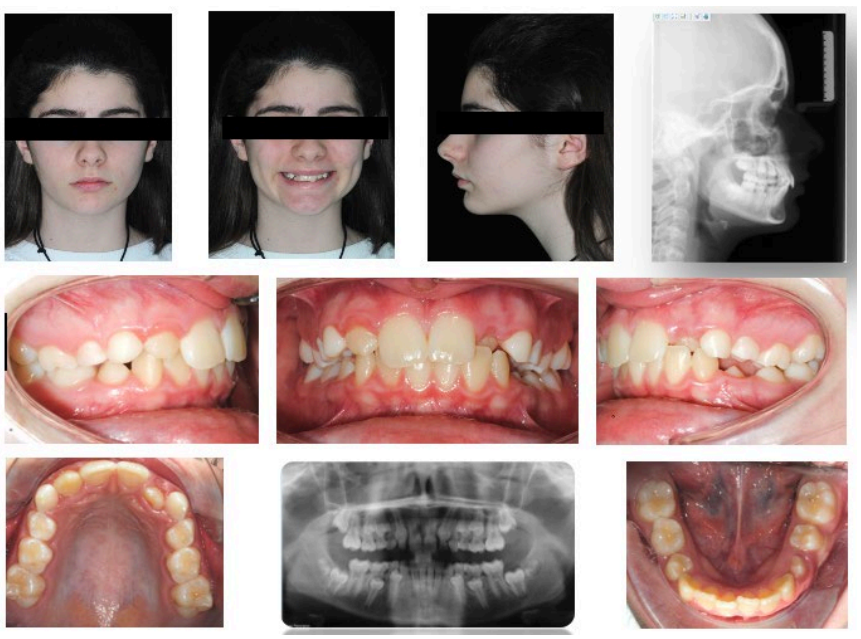

Fig. 1. *Initial records*

The panoramic X-ray highlighted the presence of all permanent teeth except the upper lateral incisors and the persistence of the deciduous canine on both sides.

The cephalometric analysis revealed a skeletal class I to class II tendency (ANB: 4°; AOBO: 1.5 mm) with a hypodivergent growth pattern (SN^GoMe: 27°; FMA: 18°) and incisors proclination (I^SN: 106°; IMPA: 102°). From the cervical vertebral maturation study, the patient was in stage 4, which means that the mandibular growth peak has already started (Fig. 1).

Finally, the intraoral scans were performed, and thus the digital casts of both arches were obtained. The patient did not receive any previous dental or orthodontic treatment. The medical history did not report any oral habits, temporomandibular disorders, or systemic pathologies.

Treatment objectives

The first objective was to correct the anterior crossbite during the transition. For that reason, a removable acrylic splint with an active coil was placed to push the 2.3 buccally. At this point, the progress of the orthodontic treatment could be performed with full permanent dentition. The space closure was planned according to the patient's characteristics, which may influence the final smile esthetics. The patient presented a class II tendency malocclusion, cuspids close to the central incisors and with homogeneous color, and good gingival and dental displays at smile. For these reasons, space closure was considered the best option, with no need for implant rehabilitation and less treatment time.

After extracting the deciduous cuspids, the appliances used were a 7-7 self-ligating multibracket fixed appliance in the upper and lower arch (0.022x0.028" Roth prescription), class II and cusp seating elastics, and a mini-screw supported Mesializer to close the spaces. This phase of treatment lasted 2 years.

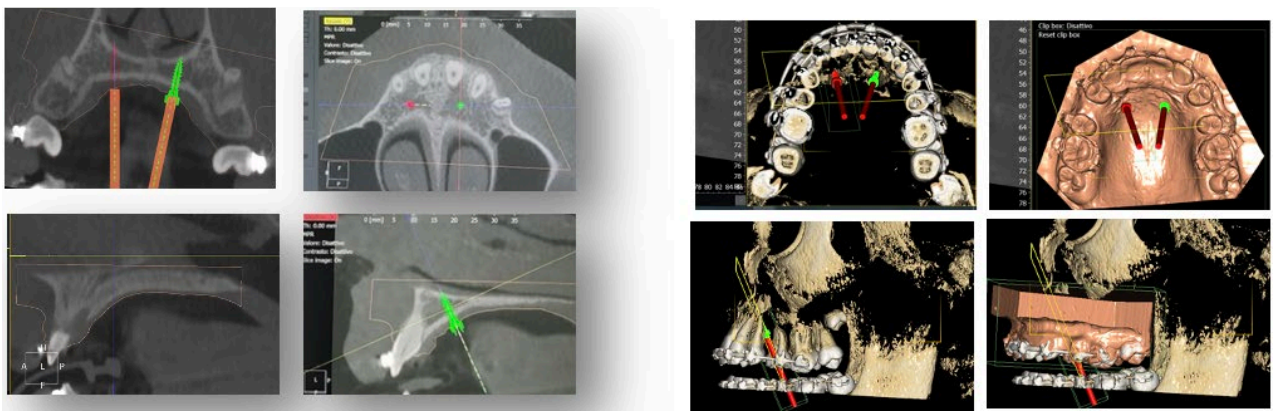

Fig. 2. *Digital planning with Easy driver protocol*

Fig. 3. *Surgical guide and Mesialslider*

The retention phase included a Hawley retainer appliance in the upper arch and a cuspid-to-cuspid fixed retainer in the lower arch.

Treatment progress

The first phase of treatment consisted of an acrylic removable splint used to correct the upper left cuspid crossbite with a pushing coil activated until the crossbite was solved. After 12 months of treatment, a re-evaluation was performed, and a more regular smile was appreciated thanks to the left canine sitting in place.

The second phase of treatment started at 13 years and 2 months after the extraction of the deciduous cuspids was performed. The self-ligating brackets with Roth prescription were applied in both arches.

The Mesialslider was used for anchorage control, which is crucial in biomechanics to maintain the central incisor's position. It includes two coupled mini-implants, stabilized by a plate (Beneplate) with 1.1 mm wire in place, fixed on the implants by two small fixing screws. Moreover, it comprises two NiTi coil springs, two activation locks, and two sliding hooks inserted into the molars' lingual tubes. The mesialization force is delivered by NiTi closing springs (200 g) attached to the activation locks. Finally, the Mesialslider is activated by pushing the activation locks mesially. Follow-up controls are scheduled every 4–6 weeks.

The Mesialslider was realized following the Easy Driver protocol. This process allowed the insertion of the appliance during the same appointment. In the beginning, the digital cast STL file was superimposed on a CBCT image to identify optimal sites for mini-implant (2mm × 9mm, Benefit system) placement in the anterior palate (Fig. 2). Once the anatomical positions of the implants were identified, the surgical insertion guide was generated, and thus Mini-implants were inserted through the surgical guide, using a contra-angle-screwdriver (Fig. 3). The device was fabricated on the same cast used for the virtual planning of the surgical guide.

Once the upper arch alignment and the leveling were achieved, the appliance was inserted, and the mesializing mechanics started (Fig. 4).

Sixteen months after the beginning of the treatment, the bilateral spaces were closed, the planned mesial movement of the maxillary molars was achieved, and the Mesialslider was removed.

Then, the treatment continued with the refinement phase as follows:

molar tubes with zero offset to permit a mesial rotation of the molar to attain an optimal cusp-to-fossa relationship;

lateral incisors brackets were bonded on the cuspids, with a strategic position: more gingival to extrude and to move down the gingival margin;

the canine cusps were grounded, and the mesio-distal width was reduced;

positive torque was added to reduce cuspid root prominence and tip forward bend to upright the root.

In these stages, 019x025 superelastic NiTi archwires were used with an extra torque in the anterior region.

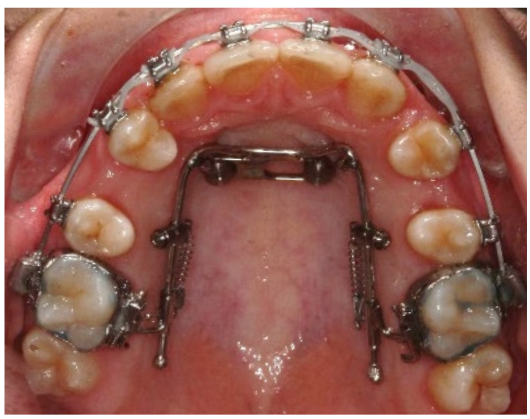

Fig. 4. *Mesialslider placed in upper jaw*

When micro aesthetics goals were achieved, multistranded archwires were used. The treatment ended after 24 months. At that point, the patient was waiting for a gingivectomy of the first bicuspids to regularize the gingival contours and the final esthetic restorations of the cuspids.

Treatment results

The treatment goals were achieved (Fig. 5). The occlusal, functional, and esthetic results were satisfactory. The outcome was rewarding for the clinicians and appreciated by the patient and her parents. A class II relationship on both sides was obtained with a correct overjet and overbite, a centered midline, and a good alignment. Extraoral records showed nice esthetics with a full pleasant smile. The panoramic x-ray showed the optimized space closure with bodily mesialization of posterior teeth and good root parallelism on both arches. The skeletal values were controlled, improving the upper and lower incisor inclination.

The key points determining the success of the treatment were good interdisciplinary cooperation (orthodontist, periodontist, and esthetic dentistry specialist) and the patient's and her parents' collaboration. It was important to begin the orthodontic treatment as soon as possible to improve the quality of the patient's present and future life.

DISCUSSION

In the dental and medical fields, there are always several therapeutic approaches to solving diagnosed problems (13, 14) temporomandibular joint (TMJ. The two alternative treatment approaches for congenitally missing upper lateral incisors are space closure and space opening. Both show advantages and disadvantages. Due to the continuing growth and the eruption of the adjacent teeth, there is a high risk of esthetic complications from the infra-positioning of single-tooth dental implants in the long term. Furthermore, implant insertion has several potential problems (15–20)temporomandibular joint (TMJ.

Moreover, in young patients, a temporary post-orthodontic retention plan needs to be adopted until the patient reaches the appropriate age for implant placement (5); this significantly postpones the final restoration, especially if pre-treatment is needed, which could not be an option for many patients. Particular attention should be paid to patients with an excess of the gingival display, such as in gummy smile situations. All these reasons have encouraged the space closure option in recent years. Canine substitutions can be accomplished with good aesthetic outcomes by tooth reshaping and positioning, bleaching, and direct or indirect restorations. To facilitate upper space closure, a mini-screw-supported device seems to be a reasonable option to provide maximum anchorage. The anterior palate proved to be a suitable insertion site, where mini-implants with larger dimensions and higher stability can be placed in a region of high bone quality, thin overlying soft tissue, and nearly negligible risk of interference with teeth or potential root damage.

The achievement of successful results in canine substitution cases depends not only on the efficiency of the space closure but also on completing all the necessary adjustments in tooth position and morphology during and after the orthodontic treatment.

Canine substitution in patients with missing lateral incisors is a well-accepted treatment option for clinicians and laypeople and has several advantages compared to single-tooth implants and tooth-supported restorations.

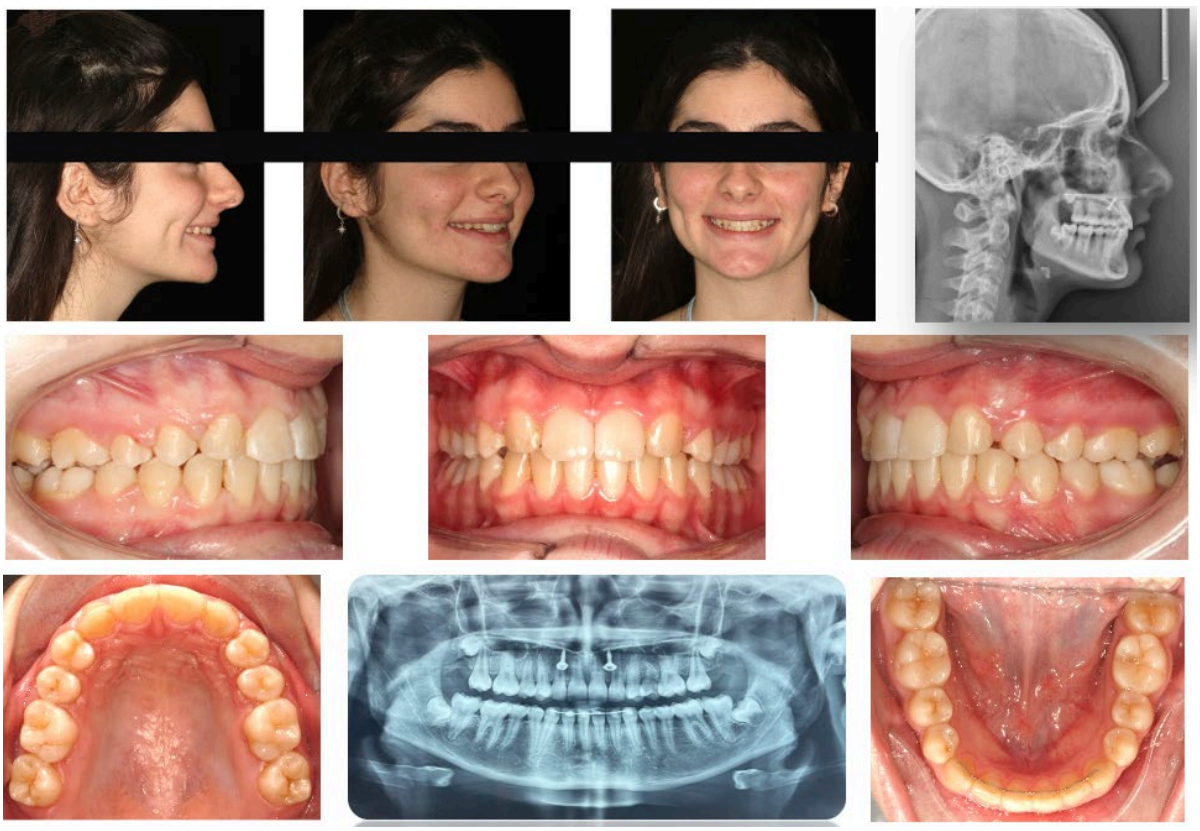

Fig. 5. *Final records*

CONCLUSIONS

Treatment of adolescent patients with bilateral agenesis of lateral incisors is among the most difficult challenges for orthodontists. Space closure is the treatment alternative that scientific evidence has proven to be the most predictable long-term. Among the advantages stand the faster outcomes achievement in young patients since the whole treatment can be completed after the orthodontic intervention, and the long-term adaptations of the teeth and supporting structures appear natural.

With palatal miniscrew-supported appliances, the clinical cases previously considered difficult or impossible to treat without oral surgery can now be successfully approached in a reasonable time since many teeth can be moved mesially simultaneously with other tooth movements in both arches. This case report is a clear example of this new treatment completed in a proper and successful manner.

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Basic Science Research

COMPARISON BETWEEN KINESIOGRAPHY AND MRI

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ABSTRACT

Background. Today, kinesiography (KN) is the most complete and advanced method for analyzing movement of the stomatognathic system. It allows to measure and graphically show mandibular movement. In the present study, magnetic resonance imaging (MRI) is used to assess the diagnostic reliability of KN in patients affected by temporomandibular (TM) disorders.

Methods. Thirty-four patients (30 females and 4 males) aged between 20 and 78 years (median 42 years) were enrolled. A KN examination using the integrated "BioPAK" system was carried out. In each patient the mandibular movements of maximal opening and fast closing were evaluated.

MRI exams were performed a few days after KN. The images of the 68 TM joints were compared with the 34 KN traces in order to assess a concordance of results.

Results. When KN shows an anomaly then MRI confirms the presence of an anatomical change. Instead, in case of normal KN, MRI can discover impairment of the TM joint.

Conclusions. This study showed that KN, despite being a useful method in recording mandibular kinetics, is not very sensitive in detecting TM disorder especially when traces are normal, or if one of the two joints is particularly compromised.

KEYWORDS: *joint disorders, jaw, mandible, record*

INTRODUCTION

Today, kinesiography (KN) is the most complete and advanced method for a functional investigation of stomatognathic system. It allows to measure and graphically show alterations of mandibular movement. Qualitative variations can be classified into: (a) kinetic, (b) vector, (c) dynamic and (d) centric anomalies (1, 2).

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A comparative evaluation of axiography and magnetic resonance imaging (MRI) was made by Rozencweig (3) and by Moritz M et al. (4). The use of both methods was suggested to increase the accuracy of diagnosis. Kuwahara T et al. (5) showed a connection between different types of TM disorders and specific impairments during chewing.

Today, MRI is considered the gold standard in TM joint imaging thanks to its ability to highlight intra-articular soft tissues and the functional alterations with static (6) and dynamic sequences (7), respectively.

In the present study, MRI is used to assess the diagnostic reliability of KN in a series of 34 patients affected by TM disorders.

MATERIALS AND METHODS

Thirty-four patients (30 females and 4 males) aged between 20 and 78 years (average age of 42 years) were enrolled. Diagnostic, and treatment, as well as the consensus for the scientific use of data, was obtained from all patients. The study was performed with respect to the Declaration of Helsinki of 2013.

A KN examination using the integrated "BioPAK" system was carried out. In each patient, the mandibular movements of maximal opening and fast closing were evaluated.

Both extents of the maximal opening on the sagittal plane measured in mm and the presence of deflections (i.e., lateral displacement on the frontal plane of the opening path of the mandibular movement which then re-centers) or deviations (i.e., lateral displacement on the frontal plane of the opening path of the mandibular movement which remains deviated from the median line) on the frontal and axial plane were recorded. KN alterations on the frontal plane were classified as mild or severe as they were smaller or greater than 3 mm. Patients were classified into 5 groups based on KN semeiotics:

1) six patients with normal tracing;
2) three patients had deflection (1 left and 2 right);
3) eleven patients had mild deviation (3 left and 8 right);
4) eight patients had severe deviation (4 left and 4 right);
5) six patients had limited mouth opening.

Patients with two or more functional alterations (i.e., limitation of mouth opening, deviation and deflection) were included in the group of severe dysfunctions.

MRI was performed using a 1.5 Tesla superconducting magnet and a dedicated surface coil. Parasagittal scans of the TM joint with closed and open mouth were carried out. Scans were centered, using an axial scout, on the head of the condyle. In addition, scans were inclined perpendicular to the axis of the condyle. TSE T2 weighted sequences (TR 2000, TE 105) and PD weighted sequences (TR 2000, TE 15) were performed, with FoV 160, 256 x 256 matrix, 3 mm thickness, 0.2 acquisition gap. MRI detected the following pathological variations: reducible disc dislocation (RDD), not reducible disc dislocation (NRDD), lock, and osteoarthrosis. MRI exams were performed a few days after the KN. The images of the 68 TM joints were compared with the 34 KN traces to assess if there was a concordance between the results.

RESULTS

In the group of 6 patients with normal traces (Table I), MRI showed 2 patients with both normal TMJs (Fig. 1); 2 subjects with bilateral RDD, and 2 patients with bilateral NRDD (Fig. 2).

In the group of 3 patients with deflection, the first patient had a joint lock whereas the other TMJ was normal, the second had a dysmorphism of the TMJ ipsilateral on the side of the deflection and the opposite TMJ with an RDD, the third patient had bilateral NRDD.

In the group of 11 patients who presented a slight deviation in the frontal plane, 4 patients had an NRDD ipsilateral to the deviation, being the opposite TMJ normal in 2 cases and with an NRDD in other 2 cases; 4 patients had an RDD in the TMJ ipsilateral to deviation while the opposite TMJ had NRDD in 1 case and RDD in other 3 cases; 2 patients had normal TMJ on the side of deviation while the opposite joint had NRDD in one case and dimorphism of the condyle in the other case; the last had a dimorphism of the condyle on the side of deviation and a normal TMJ contralaterally.

In the group of 8 patients with severe deviation, 2 subjects had TMJ lock ipsilateral to deviation and the opposite joint was normal in one case and with RDD in another; 2 patients had NRDD in both TMJs; 3 subjects had a bilateral RDD, and the last patient had a dimorphic condyle on the side of deviation and an RDD in the opposite joint.

In the group of 6 patients with limited mouth opening, a bilateral lock was present in one case; one patient had an RDD in the right TMJ and an NRDD in the left TMJ; the remaining 4 patients had bilateral NRDD.

Table I. *Results of patients with normal traces.*

N°	Kinesiographic trace	MRI of Right TMJ	MRI of left TMJ
1	Normal	Normal	Normal
2	Normal	RDD	RDD
3	Normal	Normal	Normal
4	Normal	NRDD	NRDD
5	Normal	RDD	RDD
6	Normal	NRDD	NRDD
7	Right deflection	Lock	Normal
8	Right deflection	NRDD	NRDD
9	Left deflection	RDD	osteoarthrosis
10	Mild right deviation	NRDD	Normal
11	Mild right deviation	RDD	RDD
12	Mild right deviation	NRDD	Normal
13	Mild right deviation	NRDD	NRDD
14	Mild right deviation	osteoarthrosis	Normal
15	Mild right deviation	Normal	NRDD
16	Mild right deviation	RDD	RDD
17	Mild right deviation	NRDD	NRDD
18	Mild left deviation	osteoarthrosis	Normal
19	Mild left deviation	RDD	RDD
20	Mild left deviation	NRDD	RDD
21	Severe right deviation	Lock	Normal
22	Severe right deviation	Lock	RDD
23	Severe right deviation	osteoarthrosis	RDD
24	Severe right deviation	NRDD	NRDD
25	Severe left deviation	RDD	RDD
26	Severe left deviation	RDD	RDD
27	Severe left deviation	RDD	RDD
28	Severe left deviation	NRDD	NRDD
29	Mouth opening limitation	NRDD	NRDD
30	Mouth opening limitation	NRDD	NRDD
31	Mouth opening limitation	RDD	NRDD
32	Mouth opening limitation	NRDD	NRDD
33	Mouth opening limitation	NRDD	NRDD
34	Mouth opening limitation	Lock	Lock

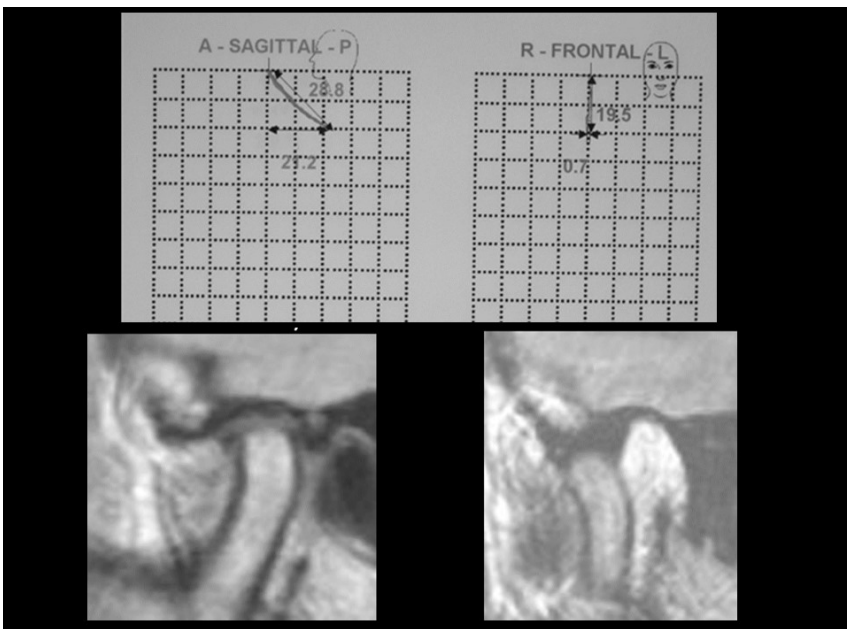

Fig. 1. *Kinesiography, fast opening and closing traces on the sagittal and frontal planes. MRI TSE T2 weighted with mouth closed and open in the sagittal plane. Regular kinesiographic trace but limited mouth opening. MRI shows a normal relationship between condyle and disc.*

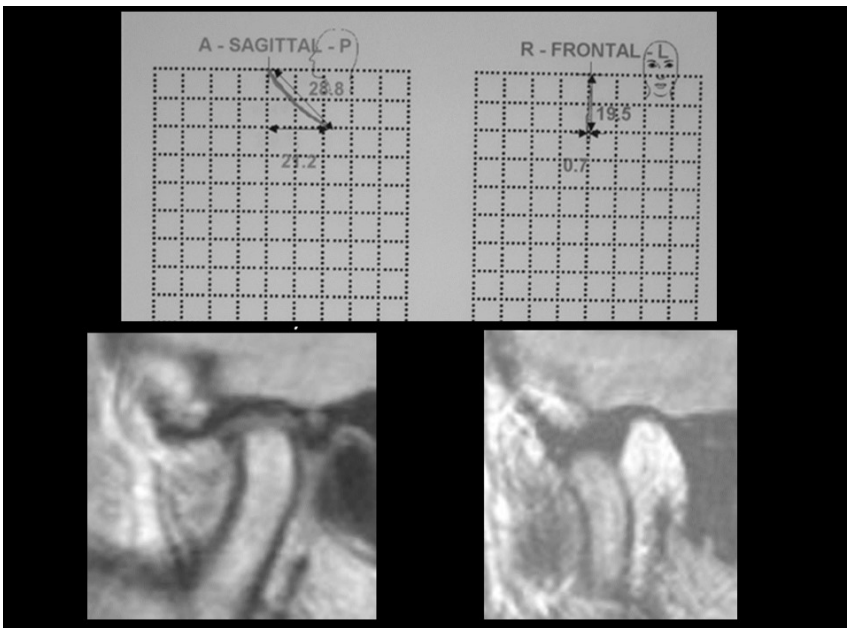

Fig. 2. *Kinesiography, fast opening and closing traces on the sagittal and frontal planes. MRI TSE T2 weighted in closed and opened mouth position in the sagittal plane. Regular kinesiographic trace but limited mouth opening while the MRI shows a bilateral NRDD.*

DISCUSSION

When the KN examination shows an evident anomaly of the traces, the MRI confirms the presence of an alteration compatible with KN, while when KN is normal MRI can confirm the diagnosis or detect anomalies.

Our data confirms what Rozencweig (3) and Moritz et al. (4) reported: there is not a bi-univocal relation between MRI and KN traces. In fact, NRDDs can determine both a deviation of the tracing towards the sick TM joint as well as a straight sign similar to those of a normal TM joint. This is due to the fact that KN records the movement of the jaw and not of a single TM joint. So KN is ideal for studying chewing cycles, pre-contacts, and freeway space but in the case of high TM joint dysfunction, KN records the same side anomaly but hardly highlights a dysfunction present in TMJ of the opposite side (7-9). Our results are in agreement with those of a recent systematic review of the literature (10-12).

Despite the intrinsic limitations of the exam, KN is useful in patients who cannot undergo to MRI due to severe claustrophobia or carrying a prosthesis that produces ferromagnetic artifacts.

This study shows that kinesiography, despite being a useful method in recording mandibular kinetics, is not very sensitive in the diagnosis of TMD especially when traces are normal or if one of the two TMJs is highly compromised.

Contributions

DM: acquisition of clinical and imaging data and interpretation of data; FC: drafting the manuscript; MDG: revision of the manuscript; LB: final approval of the published version.

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Case series

MINIMALLY INVASIVE SINUS CRESTAL LIFT: A CASE SERIES

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ABSTRACT

In cases where the residual height of the maxillary alveolar process does not allow implant placement, a regenerative technique is needed. This case series aims to suggest a minimally invasive technique to achieve controlled sinus floor elevation and the concomitant insertion of implants, minimizing the patient's postoperative discomfort. This procedure is combined with "compacting drills" for implant site preparation to make an osteotomy that compacts the alveolar bone toward the most apical part and laterally to the prepared site. Radiographic control was performed before each surgical re-entry and showed slight radiopacity apical and lateral to the implants, suggesting bone neo-formation, compared with the baseline radiograph.

KEYWORDS: *sinus crestal lift, implant, implant rehabilitation, bone regeneration*

INTRODUCTION

Prosthetic rehabilitation of single or multiple edentulism of the posterior maxillary region using osseointegrated implants is challenging in case of inadequate bone volume. In addition, the remodelling process following tooth removal may result in bone defects, which may jeopardize the correct three-dimensional positioning of the implant and the biological outcomes of the final implant-supported rehabilitation (1-3). In cases where the residual height of the maxillary alveolar process does not allow implant placement, therapeutic solutions include, depending on individual clinical indications, removable prostheses or traditional tooth-supported fixed prostheses rehabilitation, short implant placement or implants in the mesial and distal sinus region (4-6). As early as the 1980s, Boyne and James (7) began documenting cases of alveolar process atrophy treated by surgical augmentation of the available bone height. Their technique involved lateral access to the Schneiderian membrane, elevation, and filling the space created by its lift using biomaterial for bone regeneration (7). This therapeutic solution has been widely used and reviewed over the years and subsequently modified by Tatum in 1986 (8). The revised technique aimed for a less invasive procedure, replacing the lateral approach with a crestal access to the maxillary sinus (8). The same concept was later adopted by Summers in 1994 using osteotomes (9).

Given the frequent adverse outcomes, such as dizziness and headache, from sinus lift procedure with Summers' technique, the significant fluctuations in the procedure success depending on the surgeon's skills and experience, and the difficulty of controlling the forces during sinus floor elevation with osteotomes, several authors have worked to find a

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different procedure to achieve the same result while reducing the traumatic nature of the procedure (10-15).
This case series aims to suggest a minimally invasive technique (Sinus Easy-Fluid, FMD, Roma, Italy) aimed to achieve controlled sinus floor elevation and minimize the patient's postoperative discomfort.

MATERIALS AND METHODS

Four healthy patients, aged 36-47 years, had single or a edentulism in the posterior maxillary area. The treatment plan discussed and accepted by the patients included the placement of implants in the edentulous area. A radiographic investigation is prescribed for a three-dimensional assessment of the area to be treated. CBCT scan (Cone Beam Computer Tomography) showed adequate bone thickness in the buccal-lingual aspect but insufficient residual bone ridge height for implant placement. The treatment plan included a minimally invasive technique (Sinus Easy-Fluid) for crestal sinus lift and concomitant implant placement.

This procedure is combined with "compacting drills" for implant site preparation (Model Compact Drill, FMD, Rome, Italy) to make an osteotomy that compacts the alveolar bone toward the most apical part and laterally to the prepared site; this generates dense and compact bone tissue resulting in increased primary implant stability.

Before the surgery, the patients signed an informed consent form. The procedure began with the patient rinsing with 0.12% chlorhexidine mouthwash for 1 minute. Next, Mepivacain 20mg/ml with adrenaline 1:100000 (Optocain) was administered as local anaesthesia.

In all surgeries, a crestal incision was made, and a full-thickness flap was raised slightly more on the buccal side and less on the palatal side to properly visualize the bony ridge of the edentulous area. Buccal 45° bevelled releasing incisions may be performed at the clinician's discretion (Fig. 1).

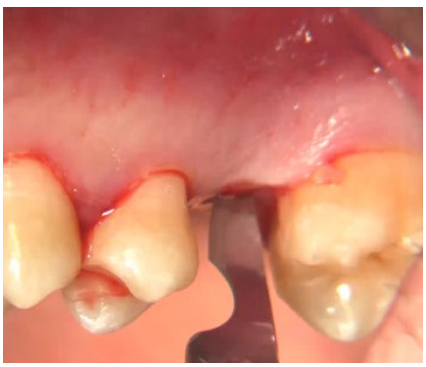


Fig. 1. *A crestal incision was performed to raise a full-thickness flap on the buccal and palatal site of the edentulous area*

However, adequate passivation of the buccal flap is recommended to allow its coronal slide tension-free during the suturing phases to achieve primary closure and submerge the implant, promoting osseointegration and apical bone neoformation.

Once the full-thickness palatal flap has been raised, a small sterile gauze is inserted between the palatal flap and the bony ridge to keep it diverged during the surgical steps. Based on radiographic evaluation or a surgical template, the first drilling phase is made with a lancet bur to create the implant site preparation. At this point, the threading steps begin (Fig. 2).

In this case, the compacting drills kit (Model Compact Drill, FMD, Rome, Italy) must be used under copious irrigation and at high RPM (1200 rpm). As mentioned earlier, the special quality of these drills is a cutting capacity when used in reverse mode; when used clockwise (forward), due to their ergonomics, they subtract bone laterally and push it apically toward the tip of the drill, which is bevelled and not sharp. This particular drill pattern is specifically designed to minimize the risk of Schneiderian membrane perforation when used near the maxillary sinus and to convey all the laterally subtracted bone to the more apical portion. Depending on the bone quality, the clinician can decide whether to use drills in a cutting or compacting mode. In the presence of extremely tight cortical bone, the clinician can perform reverse drilling for the first few millimetres and when drills are near the maxillary sinus, switch to forward mode to compact the bone apically. Taking the measurement that separates the alveolar surface from the maxillary sinus, apical stops are inserted into the drill for more security.

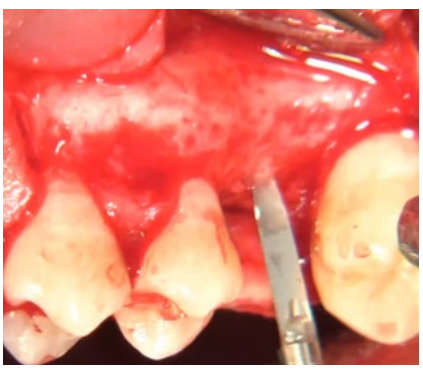
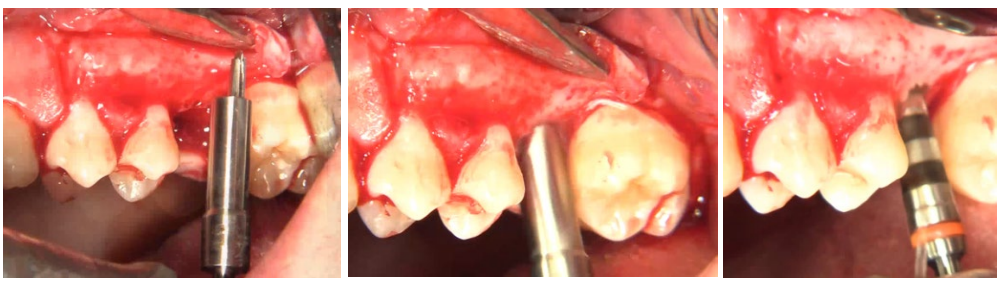


Fig. 2. *A lancet bur is used to the implant site preparation*

The first drill has an apical diameter of 1.5mm and a coronal diameter of 2.3mm (conical shape) (Fig. 3). According to the implant diameter to be placed, it is suggested to use them sequentially until the drill has a slightly smaller apical diameter than the implant. In the case illustrated, an apical diameter of 3.4 mm and a coronal diameter of 4.2 mm is used as the last compacting drill to place an implant with a diameter of 4.2 mm and a length of 10 mm. As previously described, drills are associated with apical stops to get more control over the working height. Once in the proximity of the apical bone crest, the button probe can be gently inserted to check the integrity of the Schneiderian membrane: slight mobility in the apical direction will be appreciated but with adequate resistance due to the apically compacted bone. Once the sinus floor

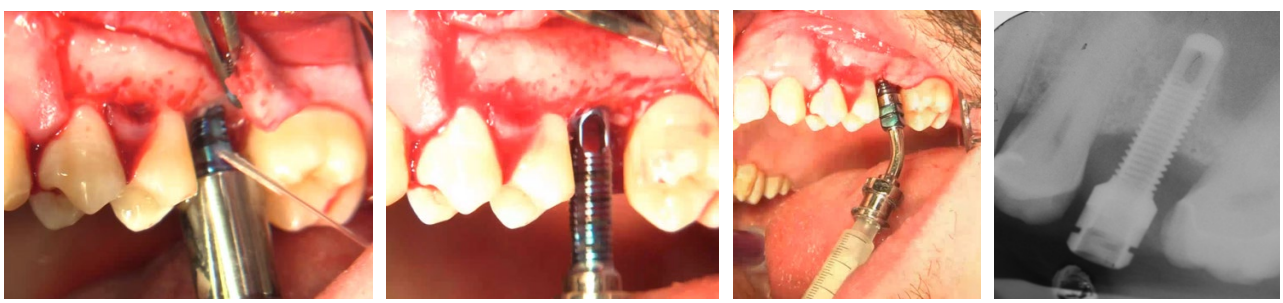
discontinuity is created, the Schneider membrane elevation kit (Sinus Easy-Fluid, FMD, Rome, Italy) is used (Fig. 4). The profiler regularizes the preparation to facilitate the placement, and insertion is performed at low rpm. The millimetre notches allow the clinician meticulous control of the elevation being performed. The profiler and dispenser have different diameters, which will be selected based on the diameter of the implant chosen to be inserted. The dispenser then begins the steps of the membrane's detachment by having a completely smooth and rounded tip. Through the notches patterned on the drill, the clinician proceeds with insertion according to the required height and length of the selected implant.

Once the desired height and good stability of the dispenser drill are achieved, the 30° curved connector is put into the dispenser and allows the Sinus Easy Fluid to inject the biomaterial into the sinus. A collagen matrix (Type I and III) loaded 60% by volume with micronized porcine bone (Gel 40 Osteobiol Tecnoss® s.r.l., Giaveno, TO, Italy) was the biomaterial used. The material leaks out of the apical lateral slots and releases the biomaterial into the space created between the Schneiderian membrane and the sinus floor in a gradual and controlled pattern. The material will then act as a space maintainer for the newly formed bone. Once this stage is completed, the implant and healing screw are inserted (Fig. 5).



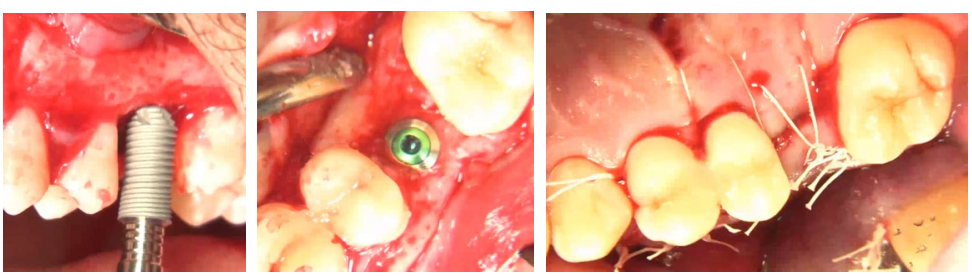
A B C

Fig. 3. ***A)*** *The first drill, with an apical diameter of 1.5 mm and a coronal diameter of 2.3 mm, is inserted inside the first hole;* ***B)*** *The apical stop allows to control the working length;* ***C)*** *Planning to place a 4.2mm-diameter implant, the last drill used for implant site preparation has an apical diameter of 3.4mm and a coronal diameter of 4.2mm.*



A B C D

Fig. 4. ***A)*** *The profiling bur is inserted inside the implant bed in order to adjust the preparation and make ready for the dosing bur.* ***B)*** *The dispenser bur with the lateral apical holes.* ***C)*** *The 30° curved connector is grafted into the dispenser and allows the Sinus Easy Fluid to inject the biomaterial into the sinus.* ***D)*** *Intraoperative radiographic control of correct dispenser bur placement.*



A B C

Fig. 5. ***A)*** *Implant placement.* ***B)*** *Occlusal-view of the implant placement and its healing screw. C) A complete closure of the flaps by primary intention with Gatlow's suture and simple detached stitches.*

It is important to check that primary stability is achieved. The first surgical phase ends with a suture with Gatlow's grooved stitches and additional simple detached stitches to achieve complete closure of the flaps by primary intention and leave the implant submerged. A second surgical phase for implant uncovering then follows after 9 months. A partial-thickness flap was raised to remove the healing screw and, following placement, a transmucosal abutment. In some cases, it was necessary to increase the keratinized tissue band by using connective grafts to ensure good stability of the peri-implant soft tissue over the years.

Antibiotic therapy with Amoxicillin 1 g every 12 hours for 6 days, rinses with chlorhexidine 0.12% for 10 days starting the day before the surgery and ibuprofen 600 mg after the surgery and every 12 hours if needed were given to patients.

RESULTS

Clinical findings analyzed during the second surgical phase (to uncover the fixture) showed implant stability and no signs of inflammation. Radiographic control was performed before each surgical re-entry and was repeated after 6 months. Slight radiopacity apical and lateral to the implants is appreciated, suggesting bone neo-formation, compared with the baseline radiograph (Fig. 6); the patient did not complain of pain or show signs of oedema, bruising or bleeding.

DISCUSSION

A sinus lift procedure is a surgical intervention aimed at increasing the height of residual bone in the posterior maxilla by repositioning the floor of the maxillary sinus in an upward direction, creating an appropriate bone height that can adequately accommodate functional dental implant placement (16). Sinus lift surgery is usually followed by bone grafting to fill the compartment created between the osseous floor of the maxillary sinus and the Schneiderian membrane (16). Recent anatomical and surgical progress in understanding and performing sinus lift surgery are reported (16-19).

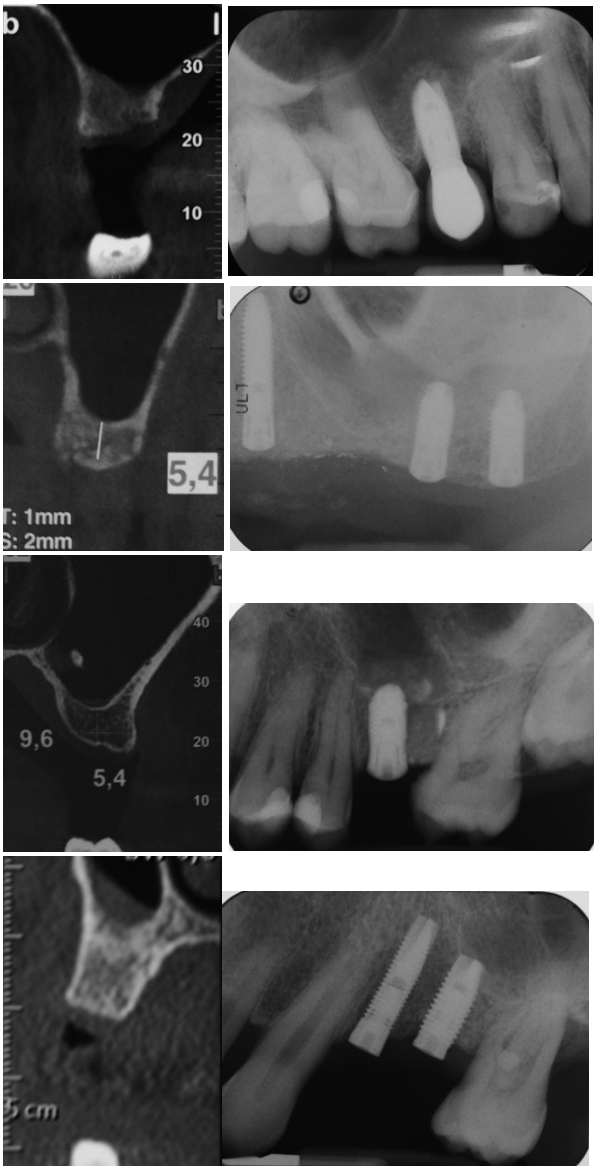


Fig. 6. *The baseline CBCT and its 9-month radiographic follow-up after the first surgical stage of patients treated with crestal sinus lift.*

Within due anatomical considerations, the clinician can use the described technique to achieve vertical bone augmentation and concomitant implant placement while reducing morbidity to the patient.

The Sinus Easy Fluid kit was used in the crestal sinus lift technique. This kit allowed the injection of biomaterial in a calibrated and predictable pattern, allowing the simultaneous disconnection of the sinus mucosa and filling of the sub-Schneiderian space thus created. A domed elevation of the sinus membrane is thus formed just apically to the implant. The benefits of the proposed technique include adequate elevation of the sinus floor achieved in a short time due to the simultaneous lifting and filling operated by the biomaterial.

The considerable advantage of approaching the crestal level also reduces both the trauma and the morbidity of the surgery. This technique, aimed only at sinus floor elevation, can be combined with the more common implant tunnel preparation methods implemented in crestal elevation, whether using rotary or piezoelectric instruments. Other studies document that hydraulic sinus lift is an effective technique (20, 21). Indeed, Sisti et al. (21) document 100% success and an increased vertical dimension maintained at 18 months.

Another issue highly debated in the literature concerns the use of biomaterials or bone substitutes during the sinus lift technique, arguing that it does not affect the success of the treatment (22). The systematic review by Del Fabbro et al. (22), conducted on 19 studies with 1822 patients and 3131 implants, shows no significant difference in survival rate when biomaterials were used and when they were not. This systematic review also showed that residual bone height was the most important prognostic factor compared to biomaterials. Thus regardless of the use of biomaterials or bone substitutes, the best survival rate was gained with a mean residual height of at least 5 mm. (22)

Some controlled clinical studies evaluate its effectiveness in maintaining at the apical level the new space obtained through trans-crestal elevation (23, 24). It seems that clinicians prefer the application of bone substitutes when performing trans-crestal elevation techniques with osteotomes (23, 24). Periapical bone maturation at the implant by the trans-crestal technique demonstrated more significant bone tissue formation when biomaterials were applied, compared with sites without bone substitutes. In addition, radiographically, the more significant bone gain was appreciated for a longer time compared with sites without biomaterials; this supports the need to perform the trans-crestal sinus lift technique simultaneously with bone substitutes (24). Precisely for these reasons, the profiler drill is used to regularize the preparation and begins to cleave and lift the Schneider's membrane, creating an apical space to the future implant; conversely, the dispenser drill, with its smooth, rounded tip, completes the desired and calibrated lift thanks to the millimetre notches. Lateral slots allow the injection of the biomaterial into the space created between Schneider's membrane and the sinus floor.

In our series, a collagen matrix (Type I and III) loaded 60% by volume with micronized porcine bone was used as the biomaterial (Gel 40 Osteobiol Tecnoss® s.r.l., Giaveno (TO), Italy). The choice of material is justified by the clinician's need to employ an osteoconductive material that facilitates the formation of a primary blood clot and the subsequent infiltration of repairing and regenerative cells; moreover, the cortico-cancellous component provides the necessary scaffold function. The size of the bone substitute, which in this case is up to 300 μm, must also be selected by the clinician according to the diameter of the side holes of the dispenser drill.

An additional important aspect of a successful crestal sinus lift is to achieve good primary stability that ensures osseointegration. For this reason, compact drills can also positively influence implant stability by preventing bone subtraction and lateral compaction. This is because compact drills are based on osteo-modelling: their use does not prepare the osteotomy by subtraction of bone (which occurs with helical drills) but by displacing it at the apex and laterally to the prepared hole. Thus, dense and compact bone tissue is generated, resulting in increased primary implant stability.

The main complication of this technique is the perforation of Schneider's membrane. In addition, the existence of intra-sinus trabeculae may increase this complication. Small perforations can be resolved with a fibrin sponge inserted in the channel before fixture placement; however, large perforations require a lateral approach using membranes (25, 26).

Regardless of the limitations associated with this case series, 100% success was found in treated patients. Other studies report 100% success at 3-year follow-up undergoing implant therapy with crestal sinus lift (22, 27).

A study (28) recently assessed the success rate of implants placed after sinus lift by the crestal approach. A total of 1344 implants rehabilitated with single crowns were followed for 121.1 months (range 60-229 months). Overall success was 98.8%. Onehundredandninety implants of 6 mm, were followed during an average period of 109.2 months and had 97.5% success. Eleven implants, 7 mm long, were followed during an average period of 218.5 months and had a success rate of 100%. Onethousandandninetyfour implants, 8 mm long, were followed for 112.3 months and had a success rate of 98.9%. Forty-nine implants 9 mm in length, followed by an average of 212.1 months, had a 100% success rate. The authors concluded that this technique is safe and predictable.

CONCLUSIONS

Due to the continuous innovation of techniques and the increasing research of surgical instrumentation, the implantologist's operative work is simplified. As a result, operative times are shortened considerably as well as the postoperative discomfort for the patient. It is recommended to have a minimum residual alveolar ridge height of 5 mm for greater success predictability. It is also recommended to use bone substitutes following the creation of the sub-Schneiderian space, the grain size and fluidity of which will be key factors to reach a successful therapy.

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Evaluation Study

EVALUATION OF ORTHODONTIC MOVEMENT WITH THE USE OF PROPEL® OSTEOPERFORATIONS IN CASES TREATED WITH ALIGNERS

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ABSTRACT

The aim of this study was to examine the effects of the association of osteoperforations performed with Propel® and a new generation of aligners (F22®) on orthodontic movement. Seven Caucasian adults were selected for this study. The patients presented a malocclusion with symmetrical dental positions and/or orthodontic movements to be performed bilaterally in a symmetrical way. Osteoperforations were performed on one side of the mouth, and after surgery, they started orthodontic treatment with aligners. The dental movent was evaluated by examining scans of dental arches and photographic examination. The results showed significant differences in the rate of tooth movement for all three variables: tip, torque and rotation. Alveolar decortication can accelerate the orthodontic movement, but it cannot be proposed today to reduce treatment times because it is highly unpredictable.

KEYWORDS: *orthodontic movement, osteoperforations, aligners*

INTRODUCTION

In recent years the number of adult patients requiring orthodontic treatment to improve their smile has increased (1). This trend can be justified by developing methods like clear aligners or lingual orthodontics. The last one was found to be a suitable method to correct various malocclusions as the technique evolved (2). Even the clear aligner treatment was effective in complex cases such as a dentoalveolar openbite (3).

In order to have faster, more effective and more efficient orthodontic movements, many protocols are tested to facilitate orthodontic treatment while avoiding excessive forces in any case, particularly in the anterior region (4). Among

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these research areas is alveolar decortication surgery (corticotomy) (5-10). With corticotomy, we mean any intentional surgical injury of the alveolar bone to induce a metabolic response known as RAP reaction (Regional Accelerated Phenomenon) (11). After corticotomy, we have an increase in vascular and cellular supply and a temporary decrease in the hard component (osteopenia), allowing for faster tissue remodeling (2 to 10 times) than normal healing processes. In the presence of orthodontic movement, RAP should reduce the resistance of the orthodontic forces of the tooth and facilitate orthodontic movement.

Among the numerous protocols described for performing corticotomy, one of those that have received the most significant clinical feedback is osteoperforations. Osteoperforations are performed with an instrument called PROPEL, which allows trans-mucosal surgical insults of different depths around teeth whose movement is to be accelerated (12).

This study aims to evaluate whether there is a facilitation and acceleration of dental movement in adult patients by associating the RAP reaction deriving from osteoperforations with PROPEL to new generation aligners (F22R).

MATERIALS AND METHODS

Seven Caucasian adults were selected, 3 males and 4 females, at the Department of Orthodontics of the University of Ferrara. Patients required aesthetic orthodontic treatment to improve their smiles. The patients presented a malocclusion with symmetrical dental positions and/or orthodontic movements to be performed bilaterally in a symmetrical way.

The teeth examined were: incisors, canines, and upper and lower premolars. These teeth must change tip, torque and rotation. The osteoperforations were performed on one side of the mouth; the other side was used as a control. Inclusion criteria:

- patients at the end of the growth aged between 18 and 30 years old;
- healthy;
- adequate oral hygiene;
- complete dentition up to the second molar;
- absence of bone loss at the radiographic level;
- absence of prosthetic rehabilitation;
- presence of moderate to severe crowding.

Patients were treated with aligners both in the upper and lower arch. The aligners have been in polyurethane with a thickness of 0,75 mm and a grip point to increase stability. All teeth were stripped of 0.2 mm mesially and distally at T0 (before the surgical procedure) and T1 (after 4 weeks) to eliminate movement interference as programmed in the set-up. Stripping was performed with abrasive DC Orthofile® mounted on Synea WH contra-angle. Two sets of aligners were given to all patients every month. Each aligner was changed every 2 weeks. All patients were instructed to wear the aligners no less than 22 hours daily (excluding meals and oral hygiene). Osteoperforations were produced at T0 on all patients on a random side.

Surgical procedure

The surgical procedure was performed with the use of PROPEL®. The Propel© is a manual, sterile, single-patient device that produces trans-mucosal micro-osteoperforations. The device has a steel tip with a diameter of 1.6 mm which, when rotated, produces osteoperforations up to 7 mm in depth.

The osteoperforation was done directly through the keratinized gum. An anaesthetic gel (30% benzocaine) and local anaesthetic (3% carbocaine) were used to anaesthetize the surgical area. The same operator always performed the experimental side mesially and distally at a depth of 3 mm and the level of the middle third of the root only on the buccal surface adjacent to the teeth under examination. No surgical procedure was performed on the control side. Aligners were given to the patients, and they received postoperative instructions.

Assessment method

The following tests were performed on each patient at T0 (before the surgical procedure), at T1 (4 weeks), and at T2 (8 weeks) to evaluate tooth movement:

- examination of scans of dental arches;

- photographic examination.

Dental arch scans were performed with a standard TRIOS intraoral scanner (Trios @ 3shape). The teeth examined were premolars, canines and incisors in both the upper and lower arches. Each scan was then converted into a Standard Triangulation Language (STL) file. The STL files were then imported into a measurement program and converted to TOM files.

VAM software (Vectra, Canfield Scientific, Fairfield, NJ, USA) was used to measure the TOM files by entering 100 points for each arch following this protocol:

- 6 points on each tooth for incisors and canines;
- 8 points on each tooth for premolars and molars.

When all points up to the second molars have been entered, the coordinates (XYZ) have been exported. Data were then imported into an Excel file (Microsoft Excel, Microsoft, Redmond, WA, USA) and to obtain the angular measurements, they were finally transferred to a customized Excel file.

All measurements were performed twice, always by the same operator. The analysis of the TOM models included 3 angular variables: tip, torque and rotation. In addition, the intraoral photos in frontal, right lateral, left lateral, superior occlusal and inferior occlusal projections were taken at T0, T1 and T2.

Statistic analysis

The analyzes were distinguished by tip, torque and rotation. The average and standard deviation of each variable at each T were calculated. The model was estimated with the longitudinal multilevel model approach (9). The lme4 and package of the R statistical software were used. The significance level was set at $p < 0.05$.

RESULTS

The averages and standard deviations for the three variables considered are shown in Table I. Standard deviations are high compared to the mean values; this can be attributed to the sample being quantitatively small. There is no significant interaction effect (measures do not change differently over time between treated and untreated groups). On the other hand, significant differences emerge between the treated and untreated groups at the global mean level (the surgical procedure influenced the tooth movement).

The results show significant differences in the rate of tooth movement for all three variables, tip, torque, and rotation. On the experimental side, at T1 and T2, there is an increase in tooth movement compared to the control side. However, the temporal effect, understood as the variation of the measurements over time is only significant for the torque variable.

For the rotation variable, the statistical evidence shows that a statistically significant difference is present only in the global mean tooth movement with a p-value of 0.015. There is more tooth movement on the experimental side than on the control side at both T1 and T2. There is no time and interaction effect (the time dynamics do not change between the experimental and control sides) (Fig. 1).

Table I. *Descriptive statistics of the rotation, tip and torque*

Rotation, descriptive statistics (bivariate)					Tip, descriptive statistics (bivariate)					Torque, descriptive statistics (bivariate)				
Treated SN	Measurement	mu	sd	num	Treated SN	Measurement	mu	sd	num	Treated SN	Measurement	mu	sd	num
NT	0	54.64	15.02	10	NT	0	-0.3	6.081	2	NT	0	-11.19	12.2	17
T	0	51.51	21.39	10	T	0	0.15	7.283	2	T	0	-8.294	13.71	17
NT	1	54.76	15.16	10	NT	1	-0.5	5.515	2	NT	1	-10.34	11.59	17
T	1	50.74	20.54	10	T	1	-1.1	8.344	2	T	1	-7.594	12.9	17
NT	2	55.03	15.58	10	NT	2	-0.7	5.233	2	NT	2	-9.776	11.45	17
T	2	49.91	20.25	10	T	2	-1.8	8.202	2	T	2	-6.829	12.15	17

Also, for the tip variable, there is a significant difference only in the global tooth movement with a p-value of 0.042. The teeth of the experimental side had significant movement, but there was no time effect (Fig. 2). However, it must be remembered that the sample for this variable is not a significant number of teeth.

Considering torque, the temporal effect was poor but significant, with a tendency to increase over time (p-value 0.069). At T1, as the graph shows, the lines are almost parallel; this demonstrates no excessive increase in the rate of tooth movement, while at T2, there was an increasing trend (Fig. 3).

Rotation, type III Anova Test

	num DF	den DF	F-value	p-value
(Intercept)	1	44	116	0.000000000000066
Measurement	2	44	0.037	0.96
Treated SN	1	44	6.4	0.015
Measurement: treated SN	2	44	0.1	0.9

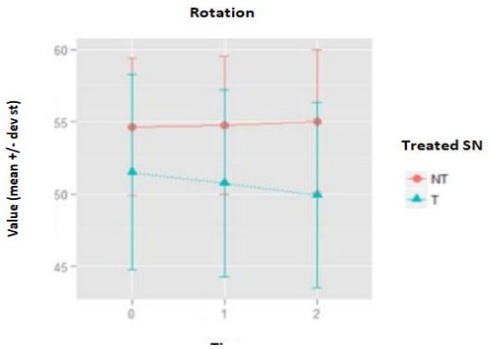

Fig. 1. *Rotation, type III Anova Test. Graph shows the movement change at T0, T1 and T2. T = treated side. NT = Non-treated side.*

Tip, type III Anova Test

	num DF	den DF	F-value	p-value
(Intercept)	1	4	0.48	0.53
Measurement	2	4	5.4	0.072
Treated SN	1	4	8.7	0.042
Measurement: treated SN	2	4	2.4	0.21

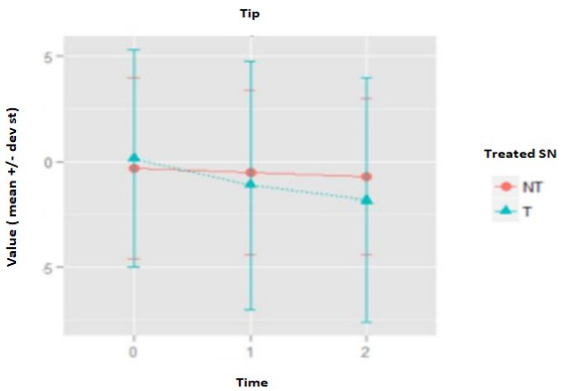

Fig. 2. *Tip, type III Anova Test. Graph shows the movement change at T0, T1 and T2.*

Torque, type III Anova Test

	num DF	den DF	F-value	p-value
(Intercept)	1	79	11	0.0012
Measurement	2	79	2.8	0.069
Treated SN	1	79	31	0.00000038
Measurement: treated SN	2	79	0.015	0.98

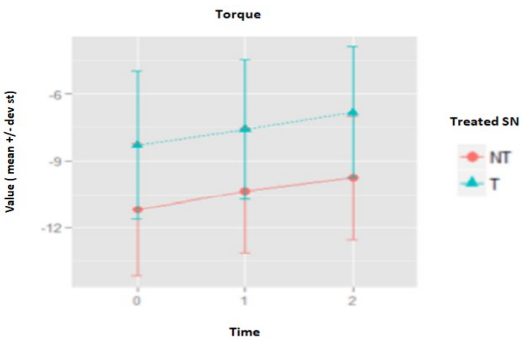

Fig. 3. *Torque, type III Anova Test. Graph shows the movement change at T0, T1 and T2*

DISCUSSION

Alveolar decortication has always been associated with orthodontic treatment to accelerate tooth movement and reduce treatment times (8, 10, 16, 17). In the present split-mouth study, we found that both at T1 (after the first month) and T2 (after the second month), the rate of tooth movement was increased on the experimental side compared to the control side. The Wilcko brothers (10) argue that following the surgical insult on the alveolar bone, there is a RAP response with osteopenia and vascular and cellular increase. Corticotomy seems to cause a RAP reaction that accelerates the intervention of macrophages responsible for the rapid removal of the hyaline tissue and increases the catabolic osteoclastic activity that allows the rapid resorption of the cortical lamina dura of the alveolus, and induces bone turnover. All these changes would result in easier and faster movement of the tooth. Our results agree with those of Murphy et al. (18), Yu et al. (19), Ren et al. (20), Mostafa et al. (21), and Hou et al. (22), who, in splitmouth studies, demonstrated that the velocity of movement of the teeth on the experimental side is greater than that on the control side.

In 2001, Aboul-Ela (23), in a study of 13 adult patients, showed that the retraction rate of maxillary canines on the corticotomised side doubled during the first two months, decreasing by 1.6 from the third month and 1.06 (i.e. similar on both sides) from the fourth month. This finding is coherent with the transitory nature of the RAP. In our study, the aligners were applied immediately after the osteoperforations were performed with the PROPEL. Osteoperforations were performed at the beginning of the treatment because the most extensive and complex movements (especially if to be obtained with aligners) generally occur during alignment and levelling. This treatment phase usually lasts even longer (about 6 months); this is in agreement with what has been argued by various authors (24-26).

Unfortunately, the evaluations only extended to the second month of treatment. The study will continue extending them up to the sixth month of treatment to establish how long the RAP effect can last and be effective on the orthodontic movement.

Combining orthodontic treatment with fixed appliances to corticotomise makes it possible to complete the therapies in 4-9 months (8, 10), while with conventional orthodontics alone, in accordance with the American Association of Orthodontists, it would take an average of 18 to 30 months. Timing greatly depends on the difficulty of the treatment, the clinician's experience and the subject's individual characteristics (AAO, 2007) (26).

Murphy et al. (18) argue that by maintaining constant mechanical stimulation, the state of transient osteopenia and the period during which the teeth can be moved can be prolonged more quickly; it is recommended to see patients more frequently, every 2 weeks, and keep the applied orthodontic forces active to achieve this effect. There is no clinical or scientific evidence of this possible extension of the "Golden State of RAP". Other authors have proposed to repeat the surgical insult several times during the treatment to increase the effect of RAP.

Sanjideh et al. (27) showed that the movement of the maxillary teeth reaches a peak between 22 and 25 days in a split-mouth study in mice, where the extractive spaces of the premolars were closed after corticotomy on one side only. At the peak, the tooth movement speed on the experimental side is greater than 85%, and the entire tooth movement is twice that of the control side. In the mandible, where a second corticotomy was performed on one side on day 28, most of the tooth movement was prolonged, but the differences were so minimal that they did not justify a second surgery. However, it is still to be determined if a second surgical insult on the alveolar bone, performed at a greater distance from the first, could be as effective as the first. In our study, no further osteoperforations were planned because of the short duration, but they could be justified in particularly long and complex treatments.

Spena et al. (28), in two studies carried out on 12 adult patients with Class II malocclusions treated with distalization of the upper molars, showed how the upper molars can be distalized in a bodily way with simple vestibular mechanics and no anterior anchoring. Furthermore, the corticotomy was performed only on the teeth that needed to be moved, reducing their resistance to distal forces and the need for anchorage. Therefore, Oliveira et al. (29) agree with what Spena demonstrated, corticotomies, generally indicated to reduce orthodontic treatment times, should be used to facilitate biomechanical difficulties of orthodontic movements.

In our study, we wanted to evaluate whether the surgical insult can facilitate dental movements both in the mesio-distal and vestibular direction after applying orthodontic forces with aligners. The results show how the association of osteoperforations performed with Propel to treatment with aligners facilitates the movements both in the mesio-distal direction and the vestibular direction. However, temporal dynamics were found only for torque movement; in this variable,

the rate of tooth movement increased more from T1 to T2 than T0 to T1; this may have been due to the surgical procedure. Unlike mesio-distal movements, buccal movements may be subject to greater resistance from the denser cortical bone. The study will have to be continued to verify the changes in the times following those examined by us, establish when the effect of the osteoperforations is present and, finally, evaluate the possible need and benefit of further subsequent osteoperforations.

CONCLUSIONS

The present study examined the effects of the association of osteoperforations performed with Propel® and the new generation of aligners (F22®) on orthodontic movement. The results showed significant differences in the rate of tooth movement for all three variables examined: tip, torque and rotation. On the experimental side, there was an increase in tooth movement compared to the control side at both T1 and T2. However, the temporal effect, understood as the variation of the measurements over time was found to be significant only for the torque value.

The association of osteoperforations performed using Propel, and new generation aligners (F22) has facilitated and accelerated the orthodontic tooth movement. However, despite the positive results obtained from the study, it must be remembered that the number of patients and teeth examined was limited. Therefore, expanding the sample to confirm these results is necessary.

Alveolar decortication can accelerate the orthodontic movement, but it cannot be proposed today to reduce treatment times because it is highly unpredictable.

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Evaluation Study

VITAMIN D ENHANCEMENT OF VIABILITY AND ANTIMICROBIAL PROPERTIES OF HUMAN GINGIVAL FIBROBLASTS AND PERIODONTAL LIGAMENT CELLS

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ABSTRACT

The better-known function of vitamin D is to support calcium homeostasis. However, the presence of vitamin D receptors in many tissues not involved in skeletal health suggested additional effects. Furthermore, vitamin D seems to be also involved in response to infection. Therefore, the aim of our study was to investigate the *in vitro* effects of the exogenous active form of vitamin D3 (VD) on human gingival fibroblasts (hGF) and human periodontal ligament cells (hPDLc) viability and on cellular mediated antibacterial effect against *Porphyromonas gingivalis* and *Streptococcus pyogenes*. Primary hGF and hPDLc pretreated or not by VD (10^{-8}, 10^{-9}, 10^{-10} mol/L) were exposed to *P. gingivalis* and S. *pyogenes* for 24h. MTT-test evaluated cell viability. CFU counting assessed bacterial growth inhibition at 8, 12 and 24 h. When infected by *P. gingivalis* and *S. pyogenes*, VD treatment hugely increased the viability in hGF (50.2% to 89.1%, 47.9% to 86.6%) and hPDLc (60.8% to 94.8% and 59.5% to 85.9%). Furthermore, treatment with VD decreased bacterial growth by approximately 30%. The obtained results support the hypothesis of a protecting role of VD on periodontal cells exposed to bacterial infection, increasing cell viability and enhancing their antibacterial effect.

KEYWORDS: *vitamin D, cell viability, gingival fibroblasts, periodontal ligament cells, antibacterial activity*

INTRODUCTION

The better-understood function of vitamin D is to support calcium homeostasis. Vitamin D regulates serum calcium and phosphate in bone by increasing absorption and, via parathormone, decreasing the release of calcium from bone and

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stimulating the reabsorption of calcium in the distal renal tubule (1, 2). However, the presence of vitamin D receptors (VDR) in many tissues not involved in skeletal health suggested that vitamin D could have additional effects (3). All the immune system cells have been shown to express VDR and can convert vitamin D to its active form (4). Moreover, vitamin D is a potent stimulator of antimicrobial peptides (cathelicidin and some defensins) in innate immunity (5). Antimicrobial peptides may have antibacterial and LPS-neutralizing activity against periodontapathogens (6). Human periodontal ligament cells (hPDLc) have been proven to take part not only in the tooth movement (7) and the maintenance of periodontal tissues but also in the regulations of periodontal inflammation secreting pro-inflammatory cytokines (8, 9) in response to *Porphyromonas gingivalis* and its component. A previous study found that hPDLc expresses vitamin D receptors (VDR) (10). Human gingival fibroblasts (hGF) are a key cellular component of gingival connective tissue, highly involved in tissue reactions to the first microbiological challenges in the gingival sulcus, being part both in signalling, responding and reparative functions (11) VDR was demonstrated to be present also on hGF (12).

However, the role vitamin D exerts is still unclear through hGF and hPDLc and if there is any effect on the antimicrobial defence exerted in the periodontium's first layers by the connective tissues. De Filippis et al. (13) previously demonstrated the decrease of *P. gingivalis* growth when the infection targeted gingival epithelial cells and periodontal cells treated with exogenous Vitamin D.

P. gingivalis (Pg), a Gram-negative anaerobic, black-pigmented rod, has been implicated as a crucial etiologic agent in the initiation and progression of periodontitis. *P. gingivalis* produces several virulence factors that stimulate hPDLc to secrete inflammatory mediators that further activate the periodontal cells to mount excessive host inflammatory responses, resulting in disease progression (14). We previously showed that the production of cytokines by hGF and hPDL cells in response to *P. gingivalis* and *S. pyogenes* could be modulated by treating the cells with vitamin D, inducing a cumulative reduction of the pro-inflammatory and an increase of the anti-inflammatory mediators (15). *P. gingivalis* requires a preformed streptococcal substratum to incorporate into a biofilm (16).

Streptococcus pyogenes (group A streptococci, GAS) is a common coloniser of the oral mucosa, nose, and pharynx. *S. pyogenes* has been shown to enter and survive intracellularly in macrophages (17), neutrophils (18, 19), and epithelial cells (20, 21) and shares with *P. gingivalis* the production of a cysteine proteinase that has been implicated in periodontal pathogenicity (22). One shared feature of the two pathogens is the ability to inhibit or evade innate host responses (23).

The aim of the current study was to examine *in vitro* the effects of an exogenous active form of vitamin D3 ($1a,25(OH)_2D_3$) on hGF and hPDL cells with respect to cellular viability and antibacterial activity against *P. gingivalis* and *S. pyogenes*.

MATERIALS AND METHODS

Reagents and bacterial strains

Active Vitamin D3 ($1\alpha,25(OH)_2D_3$) (further referred to as "VD") was obtained from Elifab s.r.l. Naples, Italy. VD was dissolved in 0.1% of absolute ethyl alcohol at 10^{-10} mol/l, 10^{-9} mol/l and 10^{-8} mol/l.

P. gingivalis from the American Type Culture Collection (ATCC 33277) was anaerobically grown at 37 °C for 2 to 3 days in Trypticase soy (TS) broth (30 g/l) containing 1 g/l yeast extract, 1 g/l glucose, 0.5 g/l potassium nitrate, 1 ml/l sodium lactate (L-1375), 0.5 g/l sodium succinate and 1 g/l sodium fumarate; after autoclaving, filter-sterilised supplements were added [0.4 g/l sodium carbonate; 0.005 g/l hemin (H-2250); 0.4 g/l cysteine; and 0.001 g/l vitamin K (M-5625)]. Late logarithmic-phase cells were employed; bacteria were washed twice in sterile PBS.

S. pyogenes was isolated from the sample from a patient with pharyngotonsillitis. *S. pyogenes* cultures were grown overnight at 37°C in fresh sterile Muller-Hinton broth, and after incubation, bacteria were washed twice in sterile PBS.

Following the final resuspension, *P. gingivalis* and *S. pyogenes* were diluted to an OD600 nm in sterile PBS, which corresponded approximately to10^7CFU/ml, to obtain multiplicities of infection (MOI) of 100. From this, stock bacteria were diluted 10^6 to obtain the MOI of 50.

Cell culture

Twelve periodontally healthy premolars and molars were obtained from young individuals (aged 22 to 36) starting

orthodontic extractive treatments after informed consent.

Teeth were washed twice in sterile PBS supplemented with antibiotics (100 U/ml penicillin, 125 ug/ml amphotericin B). Gingival tissue was isolated from the extracted teeth as described in detail by De Filippis et al. (13). Primary hPDL cells were scraped from the middle parts of the roots and treated to be employed for the experiments described below. hPDL cells from the second to fifth passages were used.

Vitamin D treatment and MTT cell viability assay

In order to evaluate the concentration effect of VD on hGF and hPDLc viability, each cell line was seeded in 0.1 ml at a concentration of 3.0 x 10^4 cells per well in a 96-well micro-titration plate without antibiotics in the cell medium. The medium was replaced with fresh 10% FBS–DMEM containing VD at a concentration of 10^{-10} mol/l, 10^{-9} mol/l and 10^{-8} mol/l and incubated for 12 h at 37°C. Control cultures received fresh 10% FBS–DMEM without VD. The optical density of the wells was determined using a microplate reader, and viability was calculated by measuring the increase in absorbance at 570 nm and was expressed as a percentage of the control value. Control was represented by hGF and hPDLc not treated with vitamin D in the medium. The MTT assay data were confirmed by counting VD-treated and VD-untreated cells in a Bürker chamber. The experiments were repeated at least three times, and the data were expressed as the mean ±SD.

Vitamin D treatment and MTT viability assay of infected cells

Cell viability was then evaluated in hGF and hPDLc exposed or not to VD at a concentration of 10^{-8} mol/l as described above and infected 100 µl of *P. gingivalis*, (MOI=50) and 100 µl of *S. pyogenes* (MOI =50). Control cultures received medium without bacteria.

Unbound bacteria were removed to prevent overgrowth by washing the plates with a medium at 2 h post-infection. At 8, 12 and 24 h post-treatment, the supernatant (100 µl) was replaced with RPMI (without phenol red) and 50 µg of 3-(4.5-dimethyl-2.5 thiazolyl)-2.5 diphenyl tetrazolium bromide was added to each well and incubated for 2 h at 37°C, as described by Nagy et al. (24). The medium was removed, and isopropanol was added. The optical density was measured as described above. The experiments were repeated at least three times, and the data were expressed as the mean ±SD.

For the following experimental models, hGF and hPDLc exposed to 10^{-8} mol/l of VD and incubated for 12 h at 37°C as described above will be reported as VD-treated cells.

Inhibition assay of Vitamin D on P. gingivalis and S. pyogenes growth in hGF and hPDLc

hGF and hPDLc were seeded in 96-well plates at a density of 1×10^6 cells/ml and were pretreated with VD at 10^{-8} mol/l for 12 h as described above. Washed and resuspended in PBS, *P. gingivalis* and *S. pyogenes* cells (MOI=50) were added and incubated in 5% CO_2 at 37 °C for 8, 12 and 24h.

After incubation, the cells were washed and lysed in cold distilled water. Bacteria were serially diluted and spread on plates for viable counts. The CFU of *P. gingivalis* and *S. pyogenes* were counted after suitable dilutions of the lysates were plated on TS agar and brain heart agar, respectively and incubated for 24-48 h at 37°C. All serial dilutions were in a cell-culture medium. Bacteria were tested in triplicate, and the assay was repeated three times. Results were expressed as a mean of the assay ±SD.

Statistical analysis

The significance of the differences in the results of each test compared to the relative control values was determined with the Student's t-test. Values of $P<0.05$ were considered statistically significant. The data are presented as means ±SD of three independent experiments.

RESULTS

MTT cell viability assay

For the dose-response experiments, hGF and hPDLc were cultured for 12 h with 10^{-10} mol/l, 10^{-9} mol/l and 10^{-8} mol/l of VD. Treatment with VD significantly increased hGF and hPDLc (Fig.1). The highest viability rate was observed with

a VD concentration of 10^{-8} mol/l and exceeded the values of VD-untreated hGF and hPDLc by 28.1% ± 3.9% and 30.3% ± 4.7% respectively.

MTT viability assay of infected cells

In Fig. 2, we show the relationship between viability rates and time exposure to *P. gingivalis* or *S. pyogenes* on 10^{-8} mol/l VD-treated cells. In comparison to the control (no exposure to VD, no contact with bacteria) considered 100%, exposure to *P. gingivalis* and *S. pyogenes* leads to a reduction of the hGF viability rate (Fig. 2A). The treatment with VD increased the viability of hGF after *P. gingivalis* and *S. pyogenes* infection from 50.2% ± 3.9% to 89.1% ± 4.8% and from 47.9% ± 3.5% to 86.6% ± 4.5%, respectively.

In hPDLc, treatment with vitamin D significantly increased the viability in comparison to untreated cells after *P. gingivalis* and *S. pyogenes* exposure time of 24 h (Fig. 2B). In particular, the treatment with VD increased hPDLc viability after *P. gingivalis* and *S. pyogenes* infection from 60.8% ± 4.2% to 94.8% ± 4.6% and from 59.5% ± 3.2% to 85.9% ± 3.8% respectively.

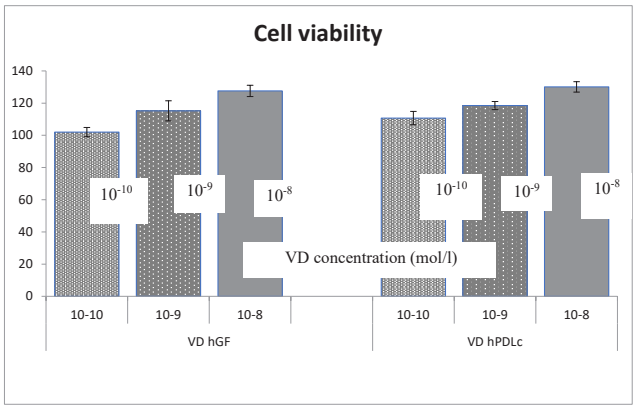

Fig. 1. *Effect of vitamin D on human gingival fibroblasts and human periodontal ligament cells viability. Dose-dependent effect of Vitamin D3 10-10 mol/l , 10-9 mol/l and 10-8 mol/l on hGF and hPDLc viability (MTT viability assay). Viability rates in percent of VD treated hGF and hPDLc, considering 100% the viability of VD untreated cell. VD hGF= human gingival fibroblasts treated with VD. VD hPDLc= Human periodontal ligament cells treated with VD. Data are means ± SD of three independent experiments. The asterisk indicates a statistically significant difference between VD treated and VD-untreated cells; *P < 0.05.*

Inhibition assay of Vitamin D on P. gingivalis and S. pyogenes growth in hGF and hPDLc

VD-Treated hGF and hPDLc cultures exhibited a significant increase in antibacterial activity (Fig 3). Vitamin D treatment showed strong time-dependent activity against both bacteria. In particular, when VD-treated hGF were infected with *P. gingivalis* and *S. pyogenes* for 24 h, the growth inhibition exerted was 27.6% ± 3.0% and 29.9% ± 2.8%, respectively, versus VD-untreated cells (9.6% ± 2.5% and 10.2% ± 2.3% respectively) (Fig. 3A).

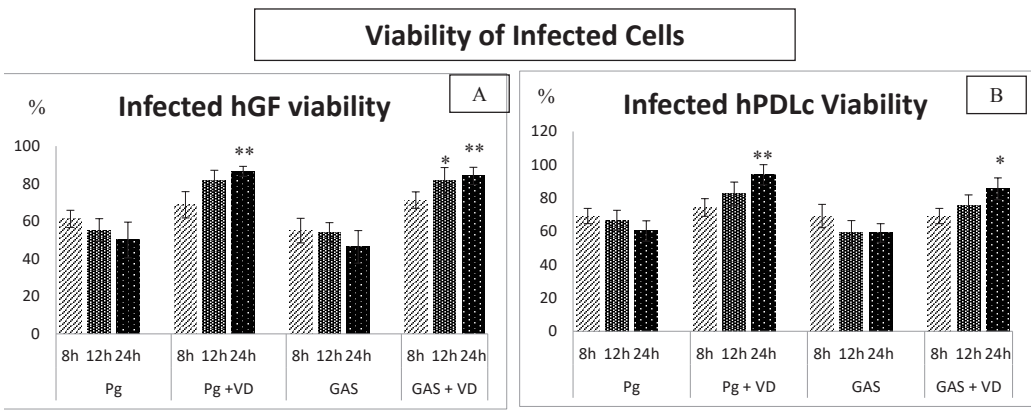

Fig. 2. *Effect of vitamin D on infected human gingival fibroblasts and human periodontal ligament cells viability. Kinetics of viability of human gingival fibroblasts (hGF) (A) and human periodontal ligament cells (hPDLc) (B) treated with VD (10-8 mol/l) and exposed to P. gingivalis (Pg) and S. pyogenes (GAS), in percent considering 100% the viability of VD-untreated not infected cells. Pg = VD-untreated hGF or hPDLc exposed to P gingivalis infection; GAS = VD-untreated hGF or hPDLc exposed to S. pyogenes infection. Data are means ± SD of three independent experiments. The asterisk indicates a statistically significant difference between VD-treated and VD-untreated infected cells. *p < 0.05. **p < 0.01*

In VD-treated hPDLc infected with *P. gingivalis* and *S. pyogenes* for 24 h, the growth inhibition was 29.7% ± 2.5% and 32.8% ± 3.4%, respectively, compared to the 8.7% ± 2.7% and 9.5% ± 2.4% of the corresponding VD-untreated cells (Fig. 3B).

DISCUSSION

In this study, we investigated the effects of vitamin D on cell viability using primary cultures of periodontal cells, both from the gingival tissue, hGF and from the periodontal ligament, hPDLc. First, we evaluated the effect of infections with *S. pyogenes* and *P. gingivalis* on the same cell types, and finally, we evaluated the antibacterial effect of vitamin D on the two bacterial strains. Since no established cell lines of hGF or hPDL are available, the cells used in the present study were all obtained from primary culture. In a previous preliminary study by our group (15), we investigated the effect of vitamin D supplementation on the viability of gingival epithelium cells and periodontal ligament cells from a single extracted tooth. Even if the samples came from a periodontally healthy subject, we considered it necessary to involve different sources of individuals, so we harvested the samples from twelve different teeth of 8 periodontally healthy young individuals. The improvement of viability of hPDL cells was consistent with the previous experiment. In the present study, vitamin D promoted hGF or hPDLc homeostasis through increased viability, as demonstrated by MTT. The results demonstrated that vitamin D increased hGF and hPDLc viability, per De Filippis et al. (13). These cells are considered of pivotal importance in inflammatory tissue response and in tissue reparative processes.

Periodontal tissue homeostasis could be considered a delicate balance between the periodontal microbiota and the host response, with occasional microbial challenges readily subdued by immune defences (24). The onset of periodontitis requires a shift in microbiological composition and a susceptible host, which exerts a complex reaction involving innate and adaptative elements that ultimately cause collateral damage to periodontal tissues (25, 26). A key factor seems to be the host's ability to manage the presence of potentially harmful microorganisms in the oral cavity through the local immune system and, on the other hand, the bacterial ability to either inhibit or evade innate host responses (27). In this view, vitamin D has a role in an antimicrobial function, thus, increasing the effects of innate immune processes, as we showed with our experimental design. These outcomes are consistent with De Filippis et al. (13) findings demonstrating

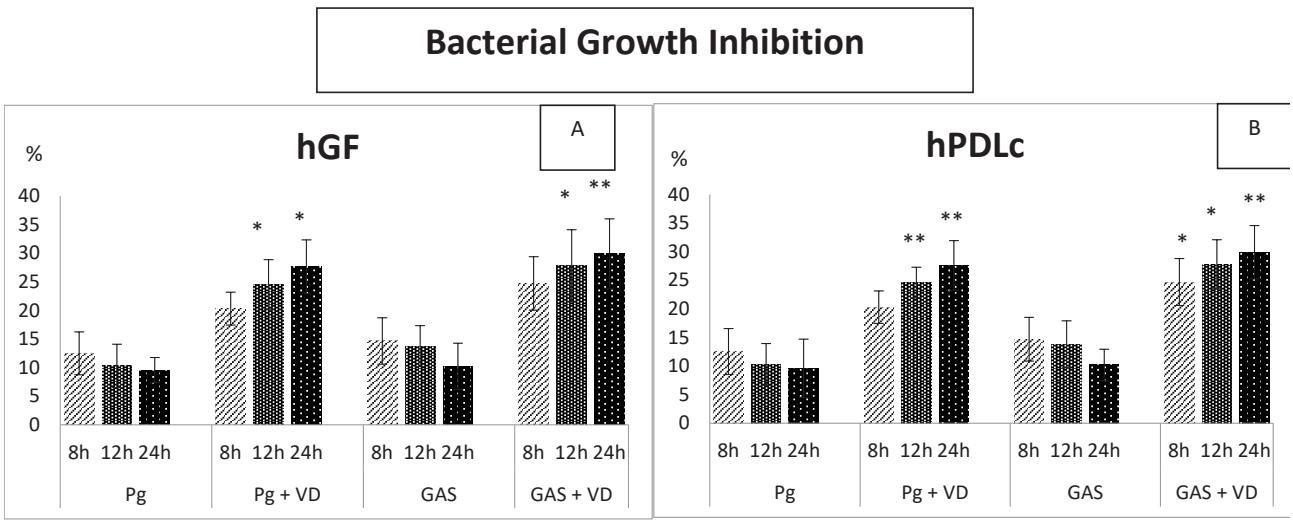

Fig. 3. *Effect of vitamin D on P. gingivalis and S. pyogenes growth inhibition. P. gingivalis (Pg)and S. pyogenes (GAS) growth inhibition (%) exerted by VD-treated and VD-untreated hGF (A) and hPDL cells (B) at 8, 12 and 24 h. Pg = VD-untreated cells infected by Pg; GAS= VD-untreated cells infected by GAS; Pg+VD= VD-treated cells infected by Pg; GAS+VD= VD-treated cells infected by GAS. Data are means ± SD of three independent experiments. The asterisks indicate a statistically significant difference in bacterial growth inhibition between the VD-treated and VD-untreated infected cells; *P < 0.05 and ** P < 0.01.*

a reduced growth of *Porphyromonas gingivalis* CFU with vitamin D on gingival epithelial cells and hPL cells. The finding of the previously published study was that vitamin D induces an antimicrobial response, and the production of the antimicrobial peptide HBD-3 was upregulated (15). The same antibacterial effect of CFU count was demonstrated by our work with *P. gingivalis* and *S. pyogenes* and exerted on hGF.

Exogenous vitamin D inhibited *P. gingivalis* and *S. pyogenes* growth on hGF and hPDLc. The exogenous form of vitamin D, thus, enhanced the innate antimicrobial properties of the cells involved in periodontal homeostasis. Recently, it was reported that vitamin D might be metabolised by hGF and hPDLc. The expression, activity and functionality of 1α-hydroxylase were detected in human gingival fibroblasts and periodontal ligament cells, raising the possibility that vitamin D acts in an autocrine/paracrine manner in these cells (28). A function of vitamin D on cytokine modulation was also reported, with a demonstrated effect on the increase of IL-10 and decrease of IL- 8 and IL-12 production by hGF and hPDL cells when stimulated by a bacterial challenge (16). Future studies may focus on this research field in several clinical applications in different dental fields (29). Clinical and experimental observations support the hypothesis that a deficit of vitamin D may lead to the deregulation of the human immune response and may be an underlying cause of infectious diseases and immune disorders acting on several molecular and cellular levels (30). The finding that vitamin D can directly modify hGF and hPDL cells could open a new perspective on the functions and significance of vitamin D deficiency.

CONCLUSION

In conclusion, our observation that vitamin D can enhance the antibacterial properties of human gingival fibroblasts and human periodontal ligament cells suggests that this pathway can be useful for the treatment or prevention of infectious diseases in the oral cavity. The enhanced hGF and hPDLc viability could promote better homeostasis and increased periodontal tissue repair. Vitamin D administration can affect the host immune responses at local and systemic levels and may represent a strategy for preventing colonisation and proliferation of oral pathogens.

Further studies are needed to elucidate the molecular mechanisms involved in these effects and to verify the potential therapeutic role of vitamin D supplementation in periodontal treatments.

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COMPARISON BETWEEN KINESIOGRAPHY AND MRI

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ABSTRACT

Today, kinesiography (KN) is the most complete and advanced method for analyzing movement of the stomatognathic system. It allows to measure and graphically show mandibular movement. In the present study, magnetic resonance imaging (MRI) is used to assess the diagnostic reliability of KN in patients affected by temporomandibular (TM) disorders. Thirty-four patients (30 females and 4 males) aged between 20 and 78 years (median 42 years) were enrolled. A KN examination using the integrated "BioPAK" system was carried out. In each patient the mandibular movements of maximal opening and fast closing were evaluated. MRI exams were performed a few days after KN. The images of the 68 TM joints were compared with the 34 KN traces in order to assess a concordance of results. When KN shows an anomaly then MRI confirms the presence of an anatomical change. Instead, in case of normal KN, MRI can discover impairment of the TM joint. This study showed that KN, despite being a useful method in recording mandibular kinetics, is not very sensitive in detecting TM disorder especially when traces are normal, or if one of the two joints is particularly compromised.

KEYWORDS: *joint disorders, jaw, mandible, record*

INTRODUCTION

Today, kinesiography (KN) is the most complete and advanced method for a functional investigation of stomatognathic system. It allows to measure and graphically show alterations of mandibular movement. Qualitative variations can be classified into: (a) kinetic, (b) vector, (c) dynamic and (d) centric anomalies (1, 2).

A comparative evaluation of axiography and magnetic resonance imaging (MRI) was made by Rozencweig (3) and by Moritz M et al. (4). The use of both methods was suggested to increase the accuracy of diagnosis. Kuwahara T et al. (5) showed a connection between different types of TM disorders and specific impairments during chewing.

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Today, MRI is considered the gold standard in TM joint imaging thanks to its ability to highlight intra-articular soft tissues and the functional alterations with static (6) and dynamic sequences (7), respectively.

In the present study, MRI is used to assess the diagnostic reliability of KN in a series of 34 patients affected by TM disorders.

MATERIALS AND METHODS

Thirty-four patients (30 females and 4 males) aged between 20 and 78 years (average age of 42 years) were enrolled. Diagnostic, and treatment, as well as the consensus for the scientific use of data, was obtained from all patients. The study was performed with respect to the Declaration of Helsinki of 2013.

A KN examination using the integrated "BioPAK" system was carried out. In each patient, the mandibular movements of maximal opening and fast closing were evaluated.

Both extents of the maximal opening on the sagittal plane measured in mm and the presence of deflections (i.e., lateral displacement on the frontal plane of the opening path of the mandibular movement which then re-centers) or deviations (i.e., lateral displacement on the frontal plane of the opening path of the mandibular movement which remains deviated from the median line) on the frontal and axial plane were recorded. KN alterations on the frontal plane were classified as mild or severe as they were smaller or greater than 3 mm. Patients were classified into 5 groups based on KN semeiotics:

1) six patients with normal tracing;
2) three patients had deflection (1 left and 2 right);
3) eleven patients had mild deviation (3 left and 8 right);
4) eight patients had severe deviation (4 left and 4 right);
5) six patients had limited mouth opening.

Patients with two or more functional alterations (i.e., limitation of mouth opening, deviation and deflection) were included in the group of severe dysfunctions.

MRI was performed using a 1.5 Tesla superconducting magnet and a dedicated surface coil. Parasagittal scans of the TM joint with closed and open mouth were carried out. Scans were centered, using an axial scout, on the head of the condyle. In addition, scans were inclined perpendicular to the axis of the condyle. TSE T2 weighted sequences (TR 2000, TE 105) and PD weighted sequences (TR 2000, TE 15) were performed, with FoV 160, 256 x 256 matrix, 3 mm thickness, 0.2 acquisition gap. MRI detected the following pathological variations: reducible disc dislocation (RDD), not reducible disc dislocation (NRDD), lock, and osteoarthrosis. MRI exams were performed a few days after the KN. The images of the 68 TM joints were compared with the 34 KN traces to assess if there was a concordance between the results.

RESULTS

In the group of 6 patients with normal traces (Table I), MRI showed 2 patients with both normal TMJs (Fig. 1); 2 subjects with bilateral RDD, and 2 patients with bilateral NRDD (Fig. 2).

In the group of 3 patients with deflection, the first patient had a joint lock whereas the other TMJ was normal, the second had a dysmorphism of the TMJ ipsilateral on the side of the deflection and the opposite TMJ with an RDD, the third patient had bilateral NRDD.

In the group of 11 patients who presented a slight deviation in the frontal plane, 4 patients had an NRDD ipsilateral to the deviation, being the opposite TMJ normal in 2 cases and with an NRDD in other 2 cases; 4 patients had an RDD in the TMJ ipsilateral to deviation while the opposite TMJ had NRDD in 1 case and RDD in other 3 cases; 2 patients had normal TMJ on the side of deviation while the opposite joint had NRDD in one case and dimorphism of the condyle in the other case; the last had a dimorphism of the condyle on the side of deviation and a normal TMJ contralaterally.

In the group of 8 patients with severe deviation, 2 subjects had TMJ lock ipsilateral to deviation and the opposite joint was normal in one case and with RDD in another; 2 patients had NRDD in both TMJs; 3 subjects had a bilateral RDD, and the last patient had a dimorphic condyle on the side of deviation and an RDD in the opposite joint.

In the group of 6 patients with limited mouth opening, a bilateral lock was present in one case; one patient had an RDD in the right TMJ and an NRDD in the left TMJ; the remaining 4 patients had bilateral NRDD.

Table I. *Results of patients with normal traces.*

N°	Kinesiographic trace	MRI of Right TMJ	MRI of left TMJ
1	Normal	Normal	Normal
2	Normal	RDD	RDD
3	Normal	Normal	Normal
4	Normal	NRDD	NRDD
5	Normal	RDD	RDD
6	Normal	NRDD	NRDD
7	Right deflection	Lock	Normal
8	Right deflection	NRDD	NRDD
9	Left deflection	RDD	osteoarthrosis
10	Mild right deviation	NRDD	Normal
11	Mild right deviation	RDD	RDD
12	Mild right deviation	NRDD	Normal
13	Mild right deviation	NRDD	NRDD
14	Mild right deviation	osteoarthrosis	Normal
15	Mild right deviation	Normal	NRDD
16	Mild right deviation	RDD	RDD
17	Mild right deviation	NRDD	NRDD
18	Mild left deviation	osteoarthrosis	Normal
19	Mild left deviation	RDD	RDD
20	Mild left deviation	NRDD	RDD
21	Severe right deviation	Lock	Normal
22	Severe right deviation	Lock	RDD
23	Severe right deviation	osteoarthrosis	RDD
24	Severe right deviation	NRDD	NRDD
25	Severe left deviation	RDD	RDD
26	Severe left deviation	RDD	RDD
27	Severe left deviation	RDD	RDD
28	Severe left deviation	NRDD	NRDD
29	Mouth opening limitation	NRDD	NRDD
30	Mouth opening limitation	NRDD	NRDD
31	Mouth opening limitation	RDD	NRDD
32	Mouth opening limitation	NRDD	NRDD
33	Mouth opening limitation	NRDD	NRDD
34	Mouth opening limitation	Lock	Lock

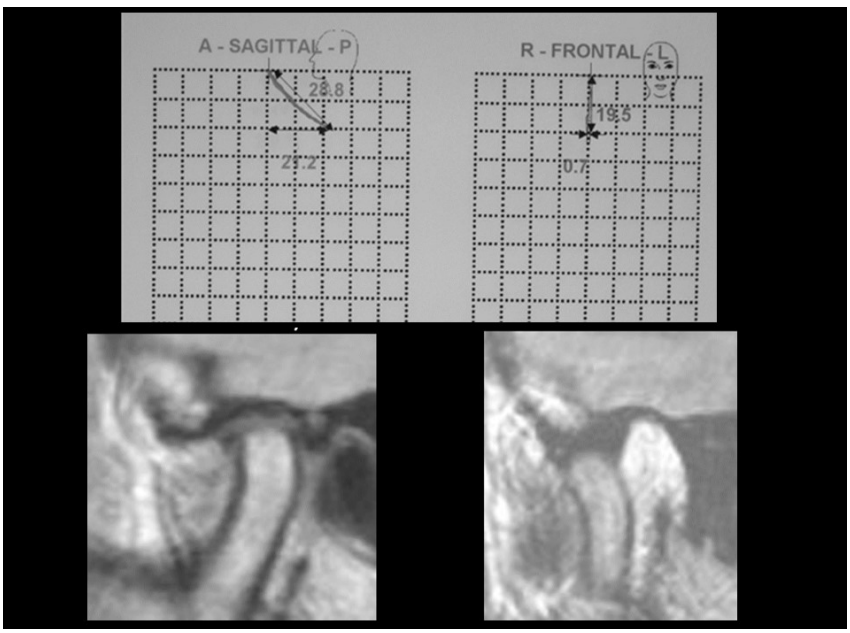

Fig. 1. *Kinesiography, fast opening and closing traces on the sagittal and frontal planes. MRI TSE T2 weighted with mouth closed and open in the sagittal plane. Regular kinesiographic trace but limited mouth opening. MRI shows a normal relationship between condyle and disc.*

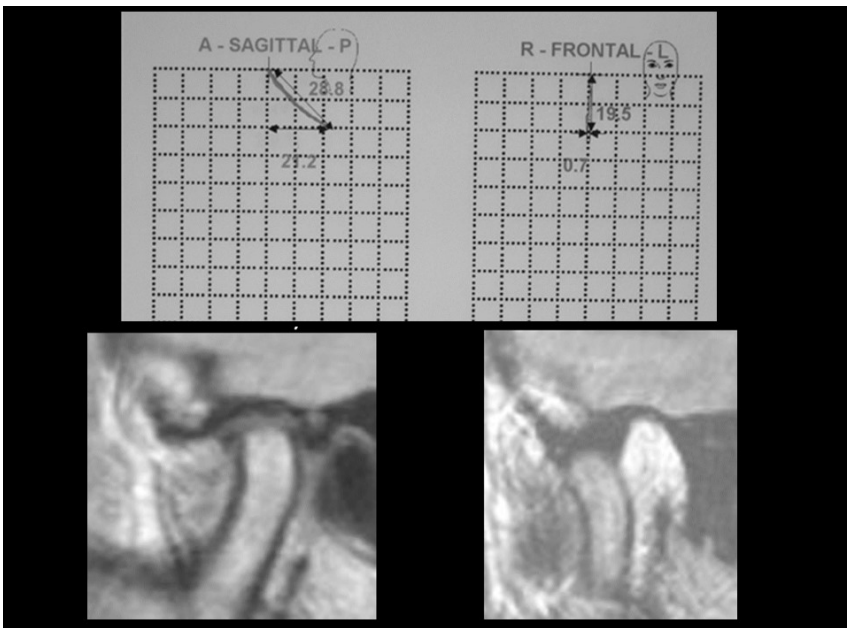

Fig. 2. *Kinesiography, fast opening and closing traces on the sagittal and frontal planes. MRI TSE T2 weighted in closed and opened mouth position in the sagittal plane. Regular kinesiographic trace but limited mouth opening while the MRI shows a bilateral NRDD.*

DISCUSSION

When the KN examination shows an evident anomaly of the traces, the MRI confirms the presence of an alteration compatible with KN, while when KN is normal MRI can confirm the diagnosis or detect anomalies.

Our data confirms what Rozencweig (3) and Moritz et al. (4) reported: there is not a bi-univocal relation between MRI and KN traces. In fact, NRDDs can determine both a deviation of the tracing towards the sick TM joint as well as a straight sign similar to those of a normal TM joint. This is due to the fact that KN records the movement of the jaw and not of a single TM joint. So KN is ideal for studying chewing cycles, pre-contacts, and freeway space but in the case of high TM joint dysfunction, KN records the same side anomaly but hardly highlights a dysfunction present in TMJ of the opposite side (7-9). Our results are in agreement with those of a recent systematic review of the literature (10-12).

Despite the intrinsic limitations of the exam, KN is useful in patients who cannot undergo to MRI due to severe claustrophobia or carrying a prosthesis that produces ferromagnetic artifacts.

This study shows that kinesiography, despite being a useful method in recording mandibular kinetics, is not very sensitive in the diagnosis of TMD especially when traces are normal or if one of the two TMJs is highly compromised.

Contributions

DM: acquisition of clinical and imaging data and interpretation of data; FC: drafting the manuscript; MDG: revision of the manuscript; LB: final approval of the published version.

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