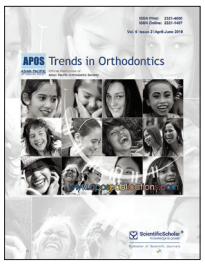
ScientificScholar®
Knowledge is power
Publisher of Scientific Journals

www.apospublications.com

APOS Trends in Orthodontics



Original Article

Salivary oxidative stress during and after rapid maxillary expansion

Sila Caglayan Topal[1], Nurdan Ozmeric[2], Serenay Elgun[3], Burcu Baloş Tuncer[1]

Departments of [1]Orthodontics, [2]Periodontology, Gazi University Faculty of Dentistry, [3]Department of Medical Biochemistry, Ankara University Faculty of Medicine, Ankara, Turkey.

***Corresponding author:**
Burcu Baloş Tuncer,
Department of Orthodontics,
Gazi University Faculty of
Dentistry, Ankara, Turkey.

burcubalostuncer@yahoo.com

Received: 25 September 2021
Accepted: 06 May 2022
Epub Ahead of Print: 16 June 2022
Published: 05 August 2022

DOI
10.25259/APOS_137_2021

Quick Response Code:

ABSTRACT

Objectives: The objectives of the study were to evaluate oxidative stress biomarkers during a rapid maxillary expansion (RME).

Material and Methods: Fourteen patients were treated with an acrylic RME device, and after treatment, all were followed for 3 months. Saliva samples were collected before activation (baseline), 1st, 10th days after the first activation, and after retention. Periodontal indexes were recorded at baseline and after retention. Nitric oxide (NO) and malondialdehyde (MDA) levels were evaluated.

Results: NO levels were elevated on the 10th day compared to baseline ($P < 0.01$), revealing a decrease after retention ($P < 0.01$). MDA levels were increased on the 10th day and after retention periods compared to baseline ($P < 0.01$, respectively). Both plaque and gingival indexes increased after retention relative to baseline ($P < 0.01$, respectively).

Conclusion: Initial stages of orthopedic force increased salivary oxidative stress biomarkers. Long-term results showed decreased levels of NO, but still high MDA levels. The initially increased NO expression decreased after retention, despite the increase in microbial load at this period which might show the effect of mechanical stimuli to be more effective than the microbial load. MDA levels remained at high levels during the post-retention period, arising a possible consideration for the impact of material properties or deficiencies of oral hygiene. Future long-term evaluations for oxidative stress status and orthodontic appliances would be useful.

Keywords: Rapid maxillary expansion, Oxidative stress, Nitric oxide, Malondialdehyde, Retention

INTRODUCTION

Orthodontically induced tooth movement is achieved by damage/repair process with inflammation-like reactions, activities of leukocytes, macrophages, and synthesis of pro-inflammatory cytokines in the periodontal structures.[1] Oxidative stress is directly involved in the inflammatory process and is mediated by reactive oxygen species (ROS) and ROS-derived radicals, which have damaging effects on cellular components of the human body.[2] In this respect, nitric oxide (NO) is a free radical related to the defensive response to inflammation by affecting bone remodeling.[3,4] Oxidative stress index has been evaluated previously in patients with periodontitis, by estimating malondialdehyde (MDA) levels to measure the oxidative damage of the lipids occurring from free radicals.[5]

Orthodontic forces comprise optimal force application to provide the desired tooth movements without unwanted reactions to the periodontal structures. However, orthopedic forces, such as

rapid maxillary expansion (RME), are more powerful and act on the basal parts of the jaws.

RME induces orthodontic and orthopedic treatment through the expansion of the mid-palatal suture, leading to the correction of posterior crossbites,[6,7] the coordination of the dental arches before orthopedic or functional treatment,[8] and to increase in the arch perimeter.[9] The previous histological data presented that the stage of skeletal maturation at the mid-palatal suture is important for the treatment of maxillary transverse deficiency using an orthopedic device, therefore, the literature search provides studies of the growth and maturation of the intermaxillary sutural system.[10,11] Mechanical loading creates cellular reactions induced by an aseptic inflammation in periodontal tissues. Tooth movement is created by cellular mechanisms. There are few studies in the literature related to oxidative stress levels during fixed orthodontic treatment.[12] We believe that an issue that has not yet been adequately assessed in the literature is the effect of an orthopedic therapeutic procedure such as RME on the role of oxidative stresses. Therefore, patients who needed a transversal maxillary expansion were observed in this study during orthopedic maxillary expansion. RME has been used to stimulate sutural growth and to correct the transverse maxillary deficiency and it has been declared that orthopedic expansion stimulates the synthesis of structural proteins, and triggers DNA synthesis, cell proliferation, and bone deposition.[1] RME involves an active stage in which the orthopedic forces are applied and treatment is followed by a retention period to provide bone remodeling.

The literature search reveals a limited amount of knowledge considering the levels of oxidative stress markers during orthodontic treatment. In a previous study, Atuğ Özcan *et al*.[12] examined the oxidative damage during fixed orthodontic treatment. However, the effect of orthopedic forces concerning oxidative damage is not clear. We hypothesized that mechanical forces through RME therapy might create oxidative stress and further lead to changes in dental and periodontal tissues. For this aim, to understand the potential impact of mechanical loading on oxidative stress, this study planned to evaluate the levels of salivary oxidative stress biomarkers during active and retention periods of RME. We aimed to measure the levels of biochemical parameters indicating current oxidative stress changes due to RME therapy to elucidate the pathogenesis of orthopedic forces exerted on palatal tissues. The correlations between clinical periodontal parameters and oxidative stress levels were also assessed.

MATERIAL AND METHODS

This study was approved by the Ethics Committee of Gazi University (25901600-5565). All patients were informed regarding their treatments and consent forms were obtained from all patients. The sample size was calculated to detect significant differences at the 0.05 significance level, and the number of patients was required to be a minimum of 12. The immunological properties were evaluated previously,[13] and the study comprised eight female and six male patients, with a mean age of 12.9 ± 0.6 years, who had been seeking orthodontic treatment at the Department of Orthodontics, Faculty of Dentistry, University of Gazi. The inclusion criteria contained that all patients required RME treatment due to the existence of transverse maxillary arch deficiency. Importance was given to the presence of good periodontal health without any evidence of bone loss, no signs of gingival inflammation, and no previous periodontal or orthodontic treatment. All patients were systemically healthy and did not take any anti-inflammatory agents, antibiotics, or immunosuppressants in the past 6 months. Saliva samples were collected before activation (baseline), 1st and 10th days after the first activation, and after a retention period of 3 months. Again, all received only RME treatment during the study with no additional braces or any other orthodontic appliances. All patients were informed to activate the appliance on the same predetermined days.

Periodontal evaluation

The study protocol is presented in [Figure 1]. All patients initially received periodontal prophylaxis with scaling and polishing. They were informed about the importance of oral hygiene and instructed to brush their teeth twice a day for a minimum of 3 min. No oral antiseptic solutions or mouthwashes were used, but motivations for maintaining oral hygiene were repeated throughout the study. After 2 weeks from the first visit, the status of the periodontal tissues was evaluated by clinical periodontal assessments including plaque index (PI),[14] gingival index (GI),[15] probing depth (PD), bleeding on probing (BOP),[16] and clinical attachment level (CAL) which were recorded as baseline measurements. All measurements were repeated at the end of retention by the same researcher (SÇT). PD was measured with a periodontal sond (Hu Friedy, Chicago, IL, USA) positioned parallel to the long axis of the tooth with no pressure. BOP was measured as the percentage of sites with bleeding. Recordings were performed around each tooth from the mesiobuccal, buccal, distobuccal, mesiopalatal, palatal, and distopalatal surfaces. Average scores of each parameter were assessed.

RME appliance

At the baseline visit, the RME appliance was placed and the first activation has been performed. The acrylic bonded RME device consisted of a Hyrax screw (Leone orthodontic products, Sesto Fiorentino, Firenze, Italy), two 0.045″ stainless steel wires extending to the palatal surfaces of the premolars

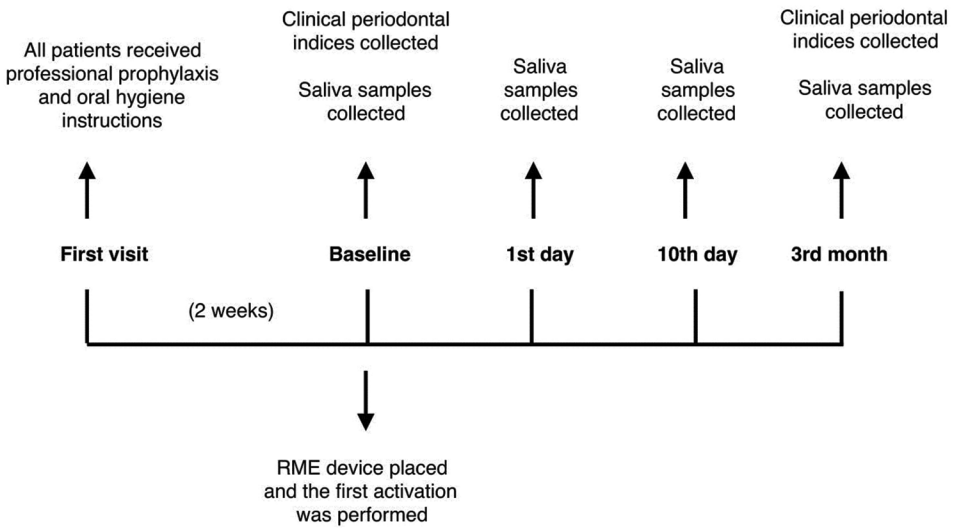


Figure 1: The protocol used in the study.

and molars, and an acrylic covering the occlusal surfaces of posterior teeth extending 2–3 mm away from the gingiva. Care was taken to ensure that the arms of the screw were parallel to the palatal mucosa. The Hyrax screw was activated twice per day with a one-quarter turn in the morning and in the evening until sufficient expansion and overcorrection were obtained. After the active phase, the screw was fixed and the appliance was left in place as a retainer for 3 months. All patients received only RME treatment during the study with no additional braces or any other orthodontic appliances. All patients were informed to activate the appliance on the same predetermined days. The expansion time was 18–21 days, with 8 ± 1.2 mm of expansion.

Collection of saliva samples

Each sampling was held in the morning, between 9.00 am and noon to avoid diurnal variation, and the participants refrained from eating, drinking, and oral hygiene procedures for 2 h before saliva collection. The participants rinsed their mouths thoroughly several times with distilled water and were rested for 3 min. Unstimulated whole saliva (3 ml) was collected concerning the spitting method.[17] Each participant spitted the accumulated saliva into an ice-chilled sterilized test tube every 60 s. Samples were checked for a possible blood leakage into saliva, not to affect the diagnostic use of saliva. Samples were centrifuged for 10 min at 15,000× *g* at 4°C to remove any particulate matter. The supernatant was frozen at −20°C until analyzed.

NO assay method

The quantitative measurement of NO was performed using a colorimetric non-enzymatic assay. Metallic cadmium was used for quantitative conversion of nitrate to nitrite before quantitation of nitrate using Griess reagent. Following the completion of the reaction, the absorbance of the colored azo dye product of the Griess reaction was measured at 540 nm and the results were expressed as μmol/mg of protein.[18]

MDA assay method

The Salivary MDA level, which provides information on lipid peroxidation, was determined by a method based on reaction with thiobarbituric acid (TBA). MDA, an end product of fatty acid peroxidation, reacts with TBA to form a colored complex, and absorbance was measured at 532 nm. Results were expressed as nmol/mg protein.[19] The analytical imprecision (i.e., the coefficient of variation expressed as a percentage, CV%) of NO and MDA analyses were determined. Within-run and day-to-day NO analyses yielded 4.3% and 5.7%, MDA analyses 4.5% and 6.0%, respectively. The analyses were performed by the same operator. The limit of detections of both analyses has been determined to be 1.0 μM.

Statistical analysis

The Statistical Package for the Social Sciences SPSS version 20.0 (USA) was used for data analysis. Sample size estimation was carried out to achieve an 80% probability of power to detect a difference of 3.2° of maxillary molar angulation concerning a previous report with a significance level of 0.05.[20] Numerical variables were presented as mean and standard deviation (SD). The normality of data distribution was verified by the Shapiro–Wilk test. The differences between the time intervals within each group were analyzed with Mann–Whitney U-test, for variables that were not normally distributed. When the distribution was normal, paired t-test was used. The correlation

between changes in the levels of oxidative stress biomarkers and changes in clinical periodontal parameters was assessed by Spearman's correlation analysis. $P < 0.05$ was considered to be statistically significant.

RESULTS

The expansion screw was activated twice a day with a mean activation period of 20 days until the required overcorrection. The mean expansion was 8 mm. After the activation period, the appliances were passively kept for 3 months in the mouth for retention. The descriptive measurements (mean, median, SD, minimum, and maximum values) of clinical periodontal parameters are shown in [Table 1]. Results demonstrated that PI and GI were significantly increased at the end of the retention period relative to baseline ($P < 0.01$, respectively).

The mean and SD of salivary oxidative stress markers are presented in [Table 2]. NO levels declared a significant increase on the 10th day when compared to baseline ($P < 0.01$). However, there was a significant decrease in the 3rd month relative to the 10th day ($P < 0.01$). MDA levels demonstrated an increase on the 10th day concerning baseline at a significance level of < 0.01. The highest level was found in the 3rd month concerning baseline ($P < 0.01$) [Figure 2].

Correlation analysis revealed no significant interaction between changes in the clinical periodontal parameters with NO and MDA levels ($P > 0.05$) [Table 3].

DISCUSSION

Orthodontic or orthopedic loading creates an aseptic inflammation in periodontal tissues and induces localized cell reactions by the release of mediators, prostaglandins, and/or cytokines to stimulate bone remodeling. These cellular mechanisms may raise questions about whether these forces induce oxidative damage in the supporting tissues. In this respect, the present study aimed to evaluate the effects of RME on salivary oxidative stress levels. We wanted to investigate early responses by detecting results on the 1st and 10th days after initial force loading, and long-term responses during the following retention period of 3 months. As far as we know, there are a limited number of studies relative to oxidative stress levels in patients with fixed orthodontic treatment.[12,21] Our findings revealed changes in salivary oxidative stress levels with orthopedic loading. The clinical contribution that can be obtained from this study demonstrates the importance of the reduction of NO levels after the retention period and that with the removal of the orthopedic force, oxidative stress levels decreased to baseline levels and remained within physiological limits. On the other hand, the high MDA levels obtained throughout the study also highlighted that orthopedic force and materials such as polymethyl methacrylate (PMMA) resins can cause oxidative damage to the lipid components of tissues in the first 3 months of RME treatment. A sampling of saliva provides easy, non-invasive, and easily tolerated evaluations, and also the whole saliva reflects a pooled sample containing variable amounts of substances from all sites, which can be used as a diagnostic tool for periodontal diseases.[22,23] During our study, oral hygiene motivations were periodically given for effective hygiene; however, we found significant increases in PI and GI scores at the end of retention, which might be linked with the long duration of the study design and/or the existence of bonded appliances. Despite the finding that none

Table 1: Clinical periodontal parameters at baseline and post-retention, and comparison between periods.

	Baseline (n=14)			Post-retention (n=14)			P-value
	X (SD)	Median	Min-Max	X (SD)	Median	Min-Max	
Plaque index	0.70 (0.41)	0.60	0.24–1.33	0.80 (0.39)	0.70	0.35–1.38	0.001
Gingival index	0.90 (0.48)	1.10	0.26–1.48	1.00 (0.47)	1.20	0.39–1.53	0.004
Probing depth (mm)	2.10 (0.47)	2.00	1.46–2.86	2.10 (0.49)	2.00	1.46–2.90	0.302
Bleeding on probing (%)	33.36 (14.12)	28.12	16.20–52.00	33.40 (15.07)	29.18	16.09–53.00	1.108

X: Mean; SD: Standard deviation; *P<0.05; **P<0.01; ***P<0.001

Table 2: Levels of NO and MDA and comparison between periods.

	Baseline		1st day		10th day		3rd month	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD
NO (μmol/mg)	44.96	24.87	49.91	25.14	56.88*	22.01	36.27†	18.48
MDA (nmol/mg)	1.50	0.92	2.17	0.93	2.33*	0.93	2.53‡	3.46

NO: Nitric oxide; MDA: Malondialdehyde, SD: Standard deviation; *P<0.05; **P<0.01; ***P<0.001. (*) Indicates significant difference for the 10th day relative to baseline at a significance level of P<0.01. (†) Indicates significant difference for the 3rd month relative to the 10th day at a significance level of P<0.01. (‡) Indicates significant difference for the 3rd month relative to baseline at a significance level of P<0.01

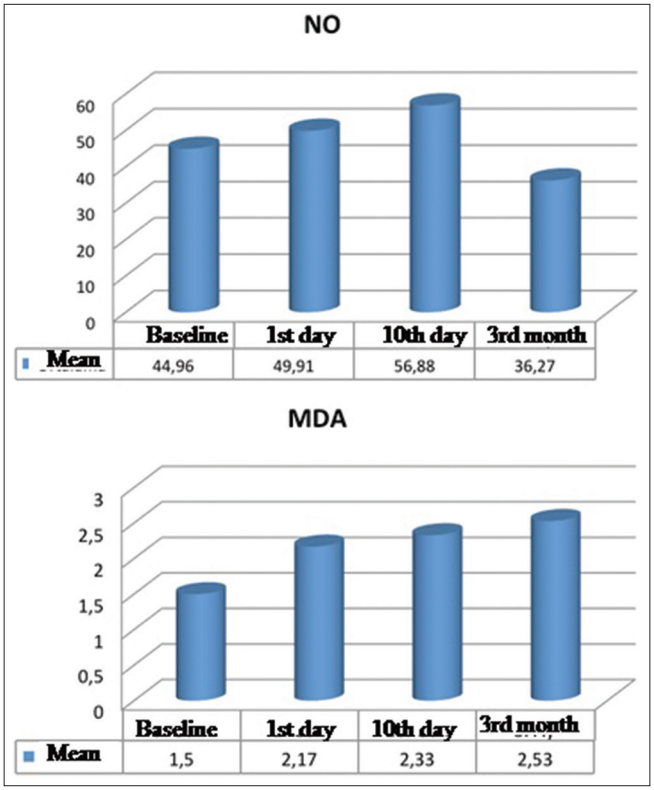


Figure 2: Levels of NO and MDA at observation periods.

Table 3: Correlation coefficients between clinical periodontal parameters with NO and MDA levels.

	NO levels	MDA levels
Plaque index		
r	0.420	0.204
P	0.135	0.483
Gingival index		
r	0.267	−0.165
P	0.356	0.572
Bleeding on probing		
r	0.114	−0.305
P	0.697	0.288

NO: Nitric oxide, MDA: Malondialdehyde; *P*>0.05, non-significant

of the patients had bone or attachment loss at the end of this period, the increased periodontal index scores declared the importance of effective tooth cleaning in the presence of such orthodontic appliances.

The active expansion period of RME includes heavy forces, while the retention period involves a certain duration to achieve adequate bone remodeling. One of our major findings was the significant elevations of NO levels on the 10th day after force loading. A previous study examined the gingival response by examining endothelial nitric oxide synthase (eNOS) and inducible nitric oxide synthase (iNOS) levels during the distalization of canines in humans, and they found their apparent role during the initial stages of orthodontic treatment, similar to our finding.[24] Again, another study reported that human pulps subjected to orthodontic force were inflamed in the first 15 days with high levels of iNOS and low levels of eNOS, followed by a decrease of iNOS and increase of eNOS levels after 30 days. This result ascertains the differential response of NOS.[25] Inline, the present results declared a significant decrease in the 3rd month relative to the 10th day. Since mechanical loading influences NO production, this finding might be related to the decreased loading and/or inflammatory response after the retention period.[25,26] Accordingly, Nilforoushan and Manolson[27] found that the high levels of NO isoforms returned to baseline levels during the later stages of orthodontic tooth movement in rats. They concluded that PDL cells respond to early orthodontic tooth movement through the production of NO by different expressions at the tension and pressure sides. The differential response of NOS has also been stated by another study, declaring the importance of iNOS and eNOS in bone remodeling during orthodontic treatment.[28] Han *et al*.[29] stated an association with the high levels of salivary NO and severe periodontitis and suggested that salivary NO might provide beneficial knowledge for monitoring periodontitis. In our study, plaque and GIs were increased; however, NO levels were decreased at the end of retention. This result showed that a decrease in NO levels occurred despite the increase in microbial load at this period. This interesting finding might point out the early influence of the orthopedic force to be more effective than the microbial load. The previous studies also suggested that human PDL cells produce NO through eNOS which is activated by cyclic tension forces[30] and that the magnitude of the orthodontic force enhances NO production.[31]

As mentioned in the literature, the oxidative damage of the lipids leads to the production of MDA, which is the most often measured evaluation for lipid peroxidation.[22] In this study, the high levels of MDA were found on the 10th day, and the highest level was evident in the 3rd month relative to baseline. In contrast to our findings, Atuğ Özcan *et al*.[12] could not detect the presence of a significant difference in levels of MDA and NO during fixed orthodontic treatment. They declared that orthodontic forces and the fixed appliances did not expose oxidative damage in the first 6 months of orthodontic tooth movement. Recently, Portelli *et al*.[21] also stated that fixed orthodontic appliances did not affect oxidative stress during the first 10 weeks of treatment. The differences between our results and previous studies might be due to the differences in force amounts, the appliance designs, and the materials.

Analysis of data suggested that higher salivary MDA levels were found in patients with periodontitis compared to healthy subjects,[32] and a correlation was found between high MDA levels and the clinical periodontal status.[33] In

contrast, Portelli *et al*.[21] did not find any correlation between oxidative stress and dental hygiene level. The high levels of MDA in the present study might be linked with the status of periodontal health, but similar to Portelli *et al*.'s results,[21] we could not find a correlation between periodontal parameters and oxidative stress markers. In addition, no evidence of periodontitis was found in patients at the end of retention. This arises a possible consideration for the impact of the material properties used for the RME device on the increased MDA levels. However, the possible impact of oral hygiene should also be taken into account when interpreting the results.

The previous studies have examined the relations between oxidative stress and the metal ions released from several types of brackets.[33,34] Buljan *et al*.[33] mentioned orthodontic brackets to be a source of oxidative stress *in vitro*, regardless of the materials. An acrylic device was applied in this study. From this viewpoint, further reasons for the high levels of MDA in our study might be related to the cytotoxicity of PMMA dentin resins, which are usually used in such intraoral appliances. These materials have potentially adverse effects on oral epithelial cells and may induce apoptosis. Methyl methacrylate has been reported to destroy cellular components and oxidative stress.[35] The close contact between oral epithelial tissues and the resins may cause an inflammatory reaction that conceives clinical relevance.[36] The changes observed in oxidative stress biomarkers could be also the result of tooth movement and remodeling of nearby alveolar bone and the periodontal tissues since saliva reflects all the changes in biological events that happened in the oral cavity. However, this could not be the case in the present study since periodontal tissues involved in RME therapy were healthy at baseline and through the evaluation period.

Taken together, the imbalance between the production of free radicals and antioxidants in favor of oxidants causes oxidative stress. Therefore, the antioxidant capability of saliva is essential for the maintenance of oral cavity balance. However, we did not evaluate the protective antioxidant enzymes present in saliva in this study. We believe that this requires further long-term evaluations concerning the effects of orthodontic appliances on the periodontal and oral environment. The potential limitations of this study included a small sample size and a lack of evaluation of the antioxidant markers together with the oxidative stress biomarkers. Another limitation of the present study is that the interpretation of the results in the molecular level could not be possible since the histological evaluation of the tissues could not be performed due to ethical issues.

The future studies would provide more beneficial data regarding the oxidative stress status with orthodontic appliances, especially after long periods of observation that may allow for repair processes.

CONCLUSION

Significant elevations in oxidative stress biomarkers were found at the early stages of RME treatment. NO may serve as an indicator of inflammation and/or bone response to mechanical loading, so the decreased levels after the retention period despite the increase in microbial load might be interpreted as a possible effect of the orthopedic force to be more effective than the microbial load. MDA levels were elevated from the early periods until the end of retention, declaring a possible adverse effect of the acrylic resin properties, but the possibility of inadequate oral hygiene should also be considered.

Declaration of patient consent

Patient's consent not required as there are no patients in this study.

Financial support and sponsorship

Nil.

Conflicts of interest

There are no conflicts of interest.

REFERENCES

1. Thilander B, Rygh P, Reitan K. Tissue reactions in orthodontics. In: Graber TM, Vanarsdall RL, Vig KW, editors. Orthodontics: Current Principles and Techniques. 4th ed. St Louis, Missouri: Elsevier; 2005. p. 145-219.
2. Chapple IL, Matthews JB. The role of reactive oxygen and antioxidant species in periodontal tissue destruction. Periodontol 2000 2007;43:160-232.
3. Uğar-Cankal D, Ozmeric N. A multifaceted molecule, nitric oxide in oral and periodontal diseases. Clin Chim Acta 2006;366:90-100.
4. Hayashi K, Igarashi K, Miyoshi K, Shinoda H, Mitani H. Involvement of nitric oxide in orthodontic tooth movement in rats. Am J Orthod Dentofacial Orthop 2002;122:306-9.
5. Baltacıoğlu E, Yuva P, Aydın G, Alver A, Kahraman C, Karabulut E, *et al.* Lipid peroxidation levels and total oxidant/antioxidant status in serum and saliva from patients with chronic and aggressive periodontitis. Oxidative stress index: A new biomarker for periodontal disease? J Periodontol 2014;85:1432-41.
6. Hesse KL, Artun J, Joondeph DR, Kennedy DB. Changes in condylar postition and occlusion associated with maxillary expansion for correction of functional unilateral posterior crossbite. Am J Orthod Dentofacial Orthop 1997;111:410-8.
7. Baccetti T, McGill JS, Franchi L, McNamara JA Jr., Tollaro I. Skeletal effects of early treatment of Class III malocclusion with maxillary expansion and face-mask therapy. Am J Orthod Dentofacial Orthop 1998;113:333-43.
8. Tollaro I, Baccetti T, Franchi L, Tanasescu CD. Role of

posterior transverse interarch discrepancy in Class II, division 1 malocclusion during the mixed dentition phase. Am J Orthod Dentofacial Orthop 1996;110:417-22.
9. Adkins MD, Nanda RS, Currier GF. Arch perimeter changes on rapid palatal expansion. Am J Orthod Dentofacial Orthop 1990;97:194-9.
10. Melsen B. Palatal growth studied on human autopsy material. A histologic microradiographic study. Am J Orthod 1975;68:42-54.
11. Melsen B, Melsen F. The postnatal development of the palatomaxillary region studied on human autopsy material. Am J Orthod 1982;82:329-42.
12. Atuğ Özcan SS, Ceylan I, Ozcan E, Kurt N, Dağsuyu IM, Canakçi CF. Evaluation of oxidative stress biomarkers in patients with fixed orthodontic appliances. Dis Markers 2014;2014:597892.
13. Topal SÇ, Tuncer BB, Elgun S, Erguder I, Ozmeric N. Levels of cytokines in gingival crevicular fluid during rapid maxillary expansion and the subsequent retention period. J Clin Pediatr Dent 2019;43:137-43.
14. Silness J, Loe H. Periodontal disease in pregnancy. II. Correlation between oral hygiene and periodontal condtion. Acta Odontol Scand 1964;22:121-35.
15. Loe H, Silness J. Periodontal disease in pregnancy. I. Prevalence and severity. Acta Odontol Scand 1963;21:533-51.
16. Ainamo J, Bay I. Problems and proposals for recording gingivitis and plaque. Int Dent J 1975;25:229-35.
17. Navazesh M. Methods for collecting saliva. Ann N Y Acad Sci 1993;694:72-7.
18. Ridnour LA, Sim JE, Hayward MA, Wink DA, Martin SM, Buettner GR, *et al.* A spectrophotometric method for the direct detection and quantitation of nitric oxide, nitrite, and nitrate in cell culture media. Anal Biochem 2000;281:223-9.
19. Agha-Hosseini F, Mirzaii-Dizgah I, Farmanbar N, Abdollahi M. Oxidative stress status and DNA damage in saliva of human subjects with oral lichen planus and oral squamous cell carcinoma. J Oral Pathol Med 2012;41:736-40.
20. Pangrazio-Kulbersh V, Wine P, Haughey M, Pajtas B, Kaczynski R. Cone beam computed tomography evaluation of changes in the naso-maxillary complex associated with two types of maxillary expanders. Angle Orthod 2012;82:448-57.
21. Portelli M, Militi A, Cervino G, Lauritano F, Sambataro S, Mainardi A, *et al.* Oxidative stress evaluation in patients treated with orthodontic self-ligating multibracket appliances: An *in vivo* case-control study. Open Dent J 2017;11:257-65.
22. Khalili J, Biloklytska HF. Salivary malondialdehyde levels in clinically healthy and periodontal diseased individuals. Oral Dis 2008;14:754-60.
23. Ozmeric N. Advances in periodontal disease markers. Clin Chim Acta 2004;343:1-16.
24. D'Attillio M, Di Maio F, D'Arcangela C, Filippi MR, Felaco M, Lohinai Z, *et al.* Gingival endothelial and inducible nitric oxide synthase levels during orthodontic treatment: A cross-sectional study. Angle Orthod 2004;74:851-8.
25. D'Attilio M, De Angelis F, Vadini M, Rodolfino D, Trubiani O, Di Nardo Di Maio F, *et al.* Endodontic-orthodontic relationships: Expression of no synthase in human dental pulp during orthodontic tooth movement. J Biol Regul Homeost Agents 2012;26:35-43.
26. Watarai H, Warita H, Soma K. Effect of nitric oxide on the recovery of the hypofunctional periodontal ligament. J Dent Res 2004;83:338-42.
27. Nilforoushan D, Manolson MF. Expression of nitric oxide synthases in orthodontic tooth movement. Angle Orthod 2009;79:502-8.
28. Tan SD, Xie R, Klein-Nulend J, van Rheden RE, Bronckers AL, Kuijpers-Jagtman AM, *et al.* Orthodontic force stimulates eNOS and iNOS in rat osteocytes. J Dent Res 2009;88:255-60.
29. Han DH, Kim MS, Shin HS, Park KP, Kim HD. Association between periodontitis and salivary nitric oxide metabolites among community elderly Koreans. J Periodontol 2013;84:776-84.
30. Kikuiri T, Hasegawa T, Yoshimura Y, Shirakawa T, Oguchi H. Cyclic tension force activates nitric oxide production in cultured human periodontal ligament cells. J Periodontol 2000;71:533-9.
31. Nakago-Matsuo C, Matsuo T, Nakago T. Basal nitric oxide production is enhanced by hydraulic pressure in cultured human periodontal ligament fibroblasts. Am J Orthod Dentofacial Orthop 2000;117:474-8.
32. Tsai CC, Chen HS, Chen SL, Ho YP, Ho KY, Wu YM, *et al.* Lipid peroxidation: A possible role in the induction and progression of chronic periodontitis. J Periodontal Res 2005;40:378-84.
33. Buljan ZI, Ribaric SP, Abram M, Ivankovic A, Spalj S. *In vitro* oxidative stress induced by conventional and self-ligating brackets. Angle Orthod 2012;82:340-5.
34. Faccioni F, Franceschetti P, Cerpelloni M, Fracasso ME. *In vivo* study on metal release from fixed orthodontic appliances and DNA damage in oral mucosa cells. Am J Orthod Dentofacial Orthop 2003;124:687-93.
35. Yamada M, Ogawa T. Chemodynamics underlying N-acetyl cysteine-mediated bone cement monomer detoxification. Acta Biomater 2009;5:2963-73.
36. Nishimiya H, Yamada M, Ueda T, Sakurai K. N-acetyl cysteine alleviates inflammatory reaction of oral epithelial cells to poly (methyl methacrylate) extract. Acta Odontol Scand 2015;73:616-25.

How to cite this article: Caglayan Topal S, Ozmeric N, Elgun S, Baloş Tuncer B. Salivary oxidative stress during and after rapid maxillary expansion. APOS Trends Orthod 2022;12:94-100.