VOLUME-9, ISSUE-6, JUNE-2020 • PRINT ISSN No. 2277 - 8160 • DOI : 10.36106/gjra

Original Research Paper

Dental Science

THE RELATIONSHIP OF ROOT RESORPTION AND SALIVARY CALCIUM LEVELS IN ORTHODONTIC PATIENTS

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ABSTRACT

Background: Root resorption is a progressive loss of dentin and cementum through continuous action of osteoclastic cells and is an unintended consequence of orthodontic mechanotherapy. **Objective:** To connect the level of root resorption and salivary calcium levels in patients after fixed orthodontic treatment. **Methods:** The research sample was 52 people. Sampling was collected through the purposive sampling technique. The measurement of root resorption in tooth 11 and 21 used the ImageJ application. The saliva of patients was collected using the draining method, and calcium levels were measured using the Glory Diagnostic Calcium Kit reagent and spectrophotometer. **Results:** Differences in the mean of external apical root resorption in tooth 11 (1.9 ± 4.6 mm) women experienced greater resorption than men, but in tooth 21 (2.9 ± 3.8 mm) men experienced greater resorption. The mean root resorption of tooth 11 (1.6 ± 4.2 mm) and tooth 21 (1.2 ± 2.3 mm). The mean calcium of patients after fixed orthodontic treatment (2.4 ± 0.8 mmol/L) and control (3 ± 0.9 mmol/L). The mean pH of patients' saliva after fixed orthodontic treatment (7 ± 0.9) and control (6.4 ± 1.0). There was a positive correlation between root resorption of tooth 11 and salivary calcium level ($r = 0.096$, $p > 0.05$), whereas there was a negative correlation between root resorption of tooth 21 and salivary calcium level ($r = 0.040$, $p > 0.05$). **Conclusion:** There was no significant difference between EARR of male and female in cases with or without premolar extraction. The pH of the patients' saliva after fixed orthodontic treatment was higher than the control, whereas the salivary calcium level after fixed orthodontic treatment was lower than the control, but both increased above the normal limits.

KEYWORDS : Resorption, Root, Calcium, Saliva

INTRODUCTION

Tooth movement is the key to any orthodontic treatment. Orthodontic tooth movement is a biological response to mechanical forces.[1] However, if the force is too large and exceeds the reparative cementum capacity, root resorption will occur.[2] The excessive pressure can suppress the periodontal ligaments. The pressure on these ligaments is known to cause the hyaline areas and damage the cementoblast layer, which can cause resorption[3]. Furthermore, it can increase the production of receptor activator of nuclear factor k-B ligand (RANKL), which stimulates the development of osteoclasts, that also play a role in root resorption.[4]

Root resorption is a progressive loss of dentin and cementum through continuous action of osteoclastic cells. Root resorption has been an unintended consequence of orthodontic mechanotherapy and has been a concern for clinicians and patients since Ottolengui first reported it in 1914.[5]

The prevalence of root resorption during orthodontic treatment varies from 4% to 91%, and Matsuda reported that 78% of patients with root resorption occurred at the end of the orthodontic treatment.[6] Marques et al. also found that 14.5% of 1,049 patients experienced severe root resorption after being treated with the edgewise method.[7] Abbas and Hartsfield reported that approximately 1 in 20 patients undergoing the orthodontic treatment were prone to experiencing a root shortening at least five mm.[5]

Root resorption has a relationship with systemic calcium levels. Low serum calcium levels can stimulate root and bone resorption.[8] Low calcium levels can stimulate the release of parathyroid hormone (PTH), which will increase osteoclast activity and induce resorption[9,10] to increase systemic calcium levels.[11] Research by Bairwa et al. showed that systemic calcium levels in serum have a positive correlation with salivary calcium levels.[12]

Fixed orthodontic appliances are also known to increase salivary calcium levels. Bhavsar et al. found that the salivary calcium level of orthodontic patients on day 45 was higher than that before treatment. The increased calcium was thought to occur due to tooth demineralization and fixed orthodontic treatment. Moreover, the saliva pH of the patients after fixed orthodontic treatment decreased in the study.[13] Research by Corega et al. found that calcium levels of orthodontic patients at week-16 were higher than at the start of the treatment. The increase in calcium levels was likely to have implications for the periodontal condition of the patients.[14]

Root resorption due to orthodontic treatment can be evaluated by conventional radiography, electron microscopy, and histopathology.[15] Ahuja et al. compared the use of periapical and panoramic radiographic techniques for measuring root resorption. Although periapical radiography is generally considered to be more efficient, there are no significant differences between the use of the two techniques for measuring root resorption in the maxillary incisors and canine.[16] However, the histopathological examination and electron microscopy were not clinically applicable.

MATERIALS AND METHODS

This study was an analytic observational study with a cross-sectional design, in which observations of calcium levels and root resorption were done in orthodontic patients. The study was conducted at the PPDGS Orthodontic clinic of Faculty of Dentistry at University of Sumatera Utara and the Integrated Laboratory of the Faculty of Medicine, University of Sumatera Utara. The study was carried out from 2019 to 2020. The total sample was 26 patients who had completed orthodontic treatment. The inclusion criteria in this study were patients who had completed orthodontic treatment and were aged 18 to 30 years. Other inclusion criteria were patients with no history of orthognathic surgery beforehand, patients with +7 years of fixed orthodontic treatment, and good sharpness and contrasted panoramic radiography. On the other hand, the

exclusion criteria were patients who refused to participate in the study and in functional care. Furthermore, the independent variable was root resorption, as seen from panoramic photographs before and after treatment. The dependent variable was the calcium levels in the patient's saliva measured by the spectrophotometer test. The controlled variables were patients aged 18 to 30 years, research procedures, saliva sampling techniques, the same panoramic instrument, and gender. The uncontrollable variables were etiology, nutrition, orthodontic mechano therapy, and Anchorage loss. The tools and materials were image J software j (NIH, Maryland, USA), Macbook Air, salivary container, digital pH meter, atomic absorption spectrophotometer (SSA), Mask, Handscoon, icebox, Deep Freezer-800C, Tripod Excell UFO 260, Canon 300D Camera, Tracing box, Black cardboard, scissors, tape, and Panoramic photos before and after treatment. The materials used in this study were panoramic radiography in patients before and after orthodontic treatment and salivary retrieval in patients after orthodontic treatment. 26 patients who had never used a bracket, their saliva were taken as a control in this study.

The procedures of this study, including the measurement of root length before and after treatment, were analyzed using ImageJ software. The steps in the analysis of ImageJ research were the researchers conducted a calibration by measuring mesiodistal tooth 11on one of the study sample models. From the measurement results, the obtained mesiodistal tooth size 11 was 8.5 mm. Furthermore, panoramic photographs of patients that have been distalized were opened through an ImageJ software application. The researchers measured mesiodistal tooth 11 on panoramic photographs using the straight-line feature (Figure 1a), followed by analyze and set scale commands. The known distance column was filled by the measurement results on the model, which was 8.5 mm. Moreover, the unit of length was filled with millimeters (mm) as the unit of measurement. The global column was checked so that the scale of measurement remained the same for all photos analyzed (Figure 1b).

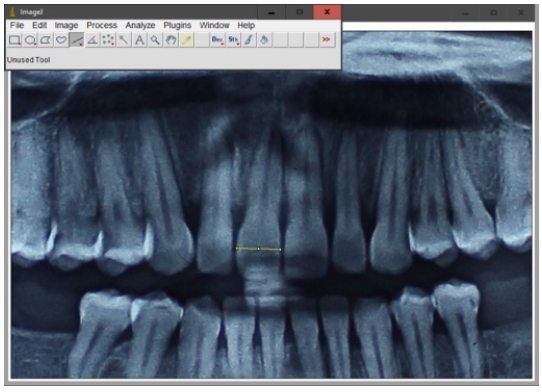

Figure1a

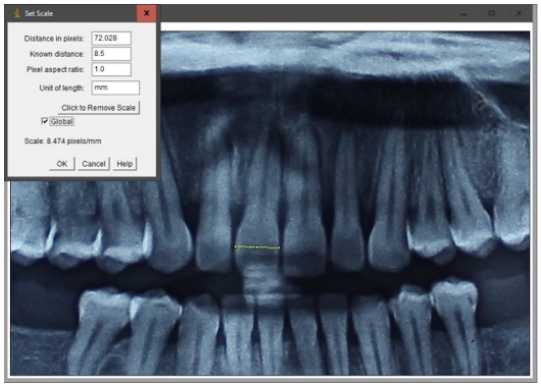

Figure 1b

Figure 1. The calibration process of *ImageJ software*.
(a) The use of the straight-line feature for the mesiodistal measurement of tooth 11 on panoramic photographs.
(b) The set of the measurement scale in *ImageJ software*.

Next, the location of the cementoenamel junction (CEJ) and tooth length axis using the straight-line feature were determined (Figure 2)

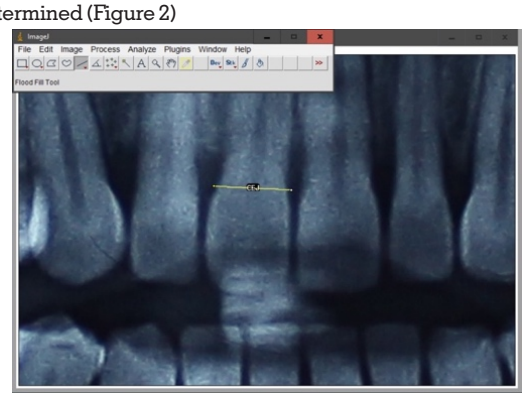

Figure 2

After the CEJ determination, the length of the crown (distance from the incisal midpoint to the CEJ midpoint, Figure 3a) and root length (distance from the CEJ midpoint to the apex, Figure 3b) with the feature of the straight line *plugins analyze measure and label* (Figure 3c). Furthermore, the measurement results of the crown and root lengths before (C, R1) and after fixed orthodontic treatment (C2, R2) were recorded on the examination sheet.

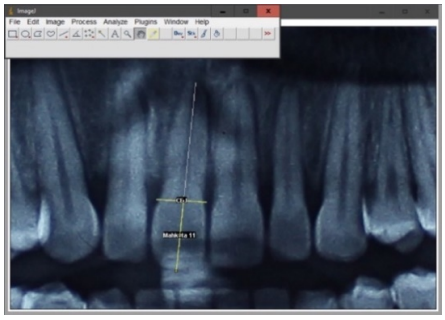

Figure 3a

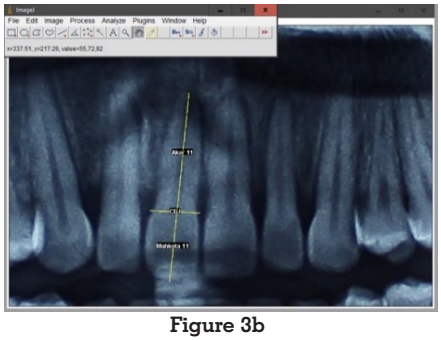

Figure 3b

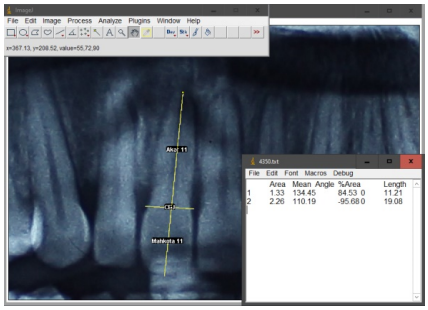

Figure 3c

External apical root resorption was calculated using the following two formulas to avoid enlargement errors on the radiographs:

EARR (mm) = R1 – (R2 x C1/C2)

$$EARR\ (\%) = \frac{R2 : C2}{R1 : C1} \times 100\%$$

Note :
R1 = root length before treatment
R2 = root length after treatment

C1 = crown length before treatment
C2 = crown length after treatment

Furthermore, the severity of resorption was categorized based on the Malmgren and Levander indices.

Saliva collection. Saliva, which was collected at 09.00-11.00 WIB, was carried out in a resting condition and was unstimulated by the draining method. All respondents were instructed not to eat, drink and smoke at least one hour before saliva was collected and were asked to rinse their mouth with water to avoid salivary contamination by local factors such as debris and tobacco. Respondents were asked to collect saliva in the mouth and drain it into a saliva pot for 5 minutes. The sample was then labeled according to the respondent's number. After that, the sample was isolated and put into a cooling box that has been filled with a cooling pack. Saliva samples were immediately taken to the Research Laboratory of the Faculty of Pharmacy, the University of Sumatera Utara, for pH and calcium concentration measurements.

Measurement of salivary pH. Salivary pH measurements were carried out with a digital pH meter. Measurements were made by inserting a digital pH meter into the saliva pot until the electrode sensor was submerged in saliva, then allowed a few seconds to show the pH level of the saliva. The measurement results were recorded on the sheet provided. After the measurement was complete, the pH meter must be cleaned and calibrated in a buffer solution before being used for further measurements.

Measurement of calcium levels. Saliva was inserted into a 250 mL microtube using a syringe and centrifuged. Then, 200 mL of reagent was placed on a blank microplate to the well of the microplate. Next, 200 mL of the reagent with the sample was mixed and allowed to stand for 2 minutes at a temperature space, then the wavelength of absorbance was read. The absorbance of the sample and the standard was subtracted with the absorbance of the blank. Calculation of salivary calcium ion levels in this study used the formula:

$$\frac{Asampel}{Astandard} \times CStandard\ mg/dL\ total\ calcium$$

The measurement results that have been obtained were recorded and analyzed.

RESULTS

Research data were processed and analyzed using a computer system.

Table 4.1 Distribution of root resorption degree of tooth 11 and 21 based on gender

Gender	N (%)	Resorption	Tooth					Total
			11		21			
			N	%	N	%	N	%
male	6 (23.1)	0	3	50	1	16.7	4	33.3
		1	1	16.7	3	50	4	33.3
		2	1	16.7	0	0	1	8.3
		3	1	16.7	0	0	1	8.3
		4	0	0	2	33.3	2	16.7
Female	20 (76.9)	0	4	20	6	30	10	25
		1	6	30	8	40	14	35
		2	1	5	3	15	4	10
		3	7	35	3	15	10	25
		4	2	10	0	0	2	5

*Mann-Whitney test

Table 4.2 Mean of External Apical Roots Resorption of Tooth 11 and 21 Based on Gender

Tooth	Gender	EARR (mean±SD)	p-value
11	Men	0.7±2.6 mm	0.429
	Women	1.9±4.6 mm	
21	Men	2.9±3.8 mm	0.201
	Women	0.7±1.5 mm	

*Mann-Whitney test

Table 4.3 Distribution of Degree and Mean of External Apical Roots Resorption of Tooth 11 and 21

Tooth	EARR (mean±SD)	Resorption Degree										Total		p-value
		0		1		2		3		4				
		N	%	N	%	N	%	n	%	n	%	n	%	
11	1.6±4.2 mm	7	26.9	7	26.9	2	7.7	8	30.8	2	7.7	26	100	0.448
21	1.2±2.3 mm	7	26.9	11	42.3	3	11.5	3	11.5	2	7.7	26	100	

*Mann-Whitney test

Table 4.4 Differences in the Mean of External Apical Roots Resorption in Patients with and without Extraction

Tooth	Case	N	EARR (mean±SD)	p-value
11	Without Extraction	15	0.6±3 mm	0.246
	With Extraction	11	2.4±4.8 mm	
21	Without Extraction	15	0.3±0.6 mm	0.169
	With Extraction	11	1.9±2.9 mm	
Total	Without Extraction	30	2.1±3.9 mm	0.101
	With Extraction	22	0.4±2.1 mm	

*Mann-Whitney test

Table 4.5 Differences in the Calcium of Patients' Saliva after Fixed Orthodontic Treatment and Control

Research Subject Group	n	Salivary Calcium Level (mean±SD)	p-value
Fixed Orthodontic Patients	26	2.4 ± 0.8 mmol/L	0.008
Control	26	3 ± 0.9 mmol/L	

*t-independent test

Table 4.6 Differences in the Average pH of Saliva in Fixed Orthodontic Patients and Control

Research Subject Group	N	the pH of Saliva (mean±SD)	p-value
Fixed orthodontic patients	26	7 ± 0.9	0.02
Control	26	6.4 ± 1.0	

*Mann-Whitney test

Table 4.7 Differences in the Average of Salivary Calcium Levels Based on Severity of External Apical Roots Resorption in Tooth 11 and 21

Tooth	Resorption Degree	Salivary Calcium Levels (Mean±SD)	p-value
11	0	2.3±0.5 mmol/L	0,779
	1	2.4±0.7 mmol/L	
	2	1.9 ± 0.2 mmol/L	

	3	2.4±1 mmol/L	
	4	3±1 mmol/L	
21	0	2.7±0.8 mmol/L	0,284
	1	2±0.7 mmol/L	
	2	1.9±0.2 mmol/L	
	3	2.2±0.6 mmol/L	
	4	2.8±1 mmol/L	

*One-Way ANOVA test

Table 4.8 Correlation between EARR (mm) and Salivary Calcium Levels

Tooth	EARR (Mean±SD)	Salivary Calcium Levels (Mean±SD)	Statistical Analysis Results	
			R	P
11	1.6±4.2 mm	2.4 ± 0.8 mmol/L	0.096	0.641
21	1.2±2.3 mm	2.4 ± 0.8 mmol/L	-0.040	0.845

*Spearman test

DISCUSSION

In this study, the average EARR that occurred in the right upper central incisor was 1.6 ± 4.2 mm, whereas the left upper central incisor was 1.2 ± 2.3 mm. The prevalence of severe resorption (grade 4) in each tooth was 7.7%.

The EARR assessment in this study was carried out with the Malmgren and Levander grading system through the calculation of changes in root length (distance from the middle of the CEJ to the most apical point of the tooth root before and after treatment) in millimeters (mm) and percent (%) using radiography panoramic. The difference in magnification between panoramic photographs before and after treatment was corrected by crown length registration.[55,56]

Research on root resorption in fixed orthodontic treatment has been carried out using a variety of different mechanotherapy.[7] Marques et al. have conducted studies to investigate root resorption in patients treated with the edgewise method. The results showed that 14.5% of patients experienced severe resorption (more than 1/3 the length of the initial root).[7] The study results by Zawawi et al. showed that root resorption in the use of the *preadjusted Roth* method was only in the mild stage (62.5%) and moderate (10%), and no severe resorption was found in the study.[57] Both results of the study were supported by Budiman's research which showed a significant difference in root resorption between the use of edgewise and preadjusted brackets, whereas root resorption in the use of edgewise brackets (2.35 ± 0.699 mm) was higher than preadjusted (1.00 ± 0.75 mm) with a p-value < 0.05.[58] A research by Reukers et al. showed no significant difference in root resorption that occurred due to the use of conventional and Roth edgewise methods (p = 0.1). However, in that study, the degree of root resorption occurred in the use of the conventional edgewise method (8.2 ± 6.4%) was greater than the Roth method (7.5 ± 7.6%) although the prevalence of root resorption in the use of the edgewise method conventional (55%) was lower than the Roth method (55%).[58]

Several studies have shown that root resorption in cases involving tooth extraction is higher than without extraction.[7,31] This is likely due to greater movement and retraction to close the extraction chamber and the longer duration of treatment.[7] In this study, there was no difference in root resorption in patients with and without premolar extraction (p > 0.05).

Moreover, various studies have also been conducted to compare differences in root resorption in fixed orthodontic treatment based on gender. Budiman's research showed that women had a higher risk of root resorption than men (p = 0.314).[59] In this study, male or female gender did not affect root resorption (p > 0.05). In line with the results of this study, Pastro et al. found no effect of gender on the severity of root resorption (p = 0.235).[61]

A study by Lindawati et al. showed that the pH and calcium of fixed orthodontic patients' saliva were higher than those of samples that did not use fixed orthodontic appliances (p = 0.001). An increase in pH in patients using fixed orthodontic appliances occurred as the body's physiological response to the presence of a bracket that is considered a foreign object.[62] The study results showed a significant difference in the salivary pH between patients after fixed orthodontic treatment and control, where the salivary pH of fixed orthodontic patients was higher (7 ± 0.9) than controls (6.4 ± 1.0) with a p-value < 0.05.[64]

Meanwhile, the research results indicated a significant difference between the patient's salivary calcium levels after fixed orthodontic treatment and control (p < 0.05). The average salivary calcium level of patients after fixed orthodontic treatment was 2.4 ± 0.8 mmol/L while the average salivary calcium level of the control was 3 ± 0.9 mmol/L; both of which were higher than the standard reference values of 1-2 mmol/L.[47] Higher calcium levels in control might be due to a decrease in pH which resulted in demineralization of the teeth.[13] However, an increase in the patient's salivary calcium levels after fixed orthodontic treatment might not be due to tooth demineralization, as described by Bhavsar et al. because the pH of the patient's saliva after fixed orthodontic treatment was high in this study, so it does not support tooth demineralization. We hypothesized that an increase in salivary calcium in patients after fixed orthodontic treatment in this study was due to root resorption initiated by cementum demineralization. To date, there have been no studies that directly link root resorption due to orthodontic treatment with changes in salivary calcium levels. However, there are several studies that might support the researcher's hypothesis, such as research on the relationship between root resorption and systemic calcium and a positive correlation between systemic calcium and salivary calcium.

Bairwa et al. have conducted studies to see whether changes in salivary calcium levels are the same as changes in calcium levels that occur systemically in serum. The results showed a positive correlation between systemic calcium levels (serum) and salivary calcium levels (r = 0.726). The study explained that bone resorption due to osteoporosis would cause the release of calcium into the serum. The level of calcium in saliva, which is an ultrafiltrate from plasma, apparently also increases so that the measurement of calcium saliva can be used as a diagnostic tool that is non-invasive, easy, and fast compared to measurement through serum.[12]

In this study, a correlation test was performed to see the relationship between root resorption levels (in mm) in tooth 11 and 21 with salivary calcium levels. Although researchers found a positive correlation between EARR on tooth 11 and salivary calcium levels, the statistical test results did not show a significant relationship between the magnitude of EARR with salivary calcium levels (p > 0.00).

CONCLUSION

1. The severity of apical root resorption in men was 23.1%, whereas the severity of apical root resorption in women was 76.9%.
2. The average pH of patients' saliva after fixed orthodontic treatment was 7 ± 0.9 higher than the control, which was 6.4 ± 1.0.
3. Based on the distribution of the severity of external apical root resorption in tooth 11, women experienced greater resorption (1.9 ± 4.6 mm) than men, whereas in tooth 21, men experienced greater resorption (2.9 ± 3.8 mm) compared to women.

4. The mean external of apical root resorption in patients with and without extraction was not significantly different between external apical root resorption in patients with and without extraction ($p > 0.05$)

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