

Aspartate Aminotransferase Activity in Gingival Crevicular Fluid during Orthodontic Tooth Movement

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ABSTRACT

Introduction: Movement of teeth during orthodontic treatment takes place as a result of remodeling in periodontium and certain chemical mediators in the form of biomarkers are expressed in gingival crevicular fluid. **Objective:** To evaluate change of Aspartate Aminotransferase level in gingival crevicular fluid during orthodontic tooth movement. **Study design:** Randomized control trial; a split mouth design. **Setting:** Department of Orthodontics, Dental Section, Punjab Medical College Faisal Abad. **Duration of study:** Six months 01-01-2016 to 30-06-2016. **Sample size:** Thirty selected patients. **Sampling technique:** Non –Probability purposive sampling. **Data collection procedure:** Full mouth plaque score and full mouth bleeding score were assessed at base line and 28th day. Patients undergoing orthodontic treatment at initial stage of alignment were evaluated where right upper 1st premolars were taken as experimental and left 1st premolar kept as control. A paper point inserted into gingival sulcus for one minute, removed then stored in normal saline. Level of Aspartate Aminotransferase was determined by spectrophotometric automatic apparatus. Data was analyzed using SPSS version 18 and tabulated. **Results:** Full mouth plaque score slightly increased while full mouth bleeding score showed slight decrease. Aspartate Aminotransferase level in both groups showed insignificant difference at base line and 1 hour, however significant difference was found at 1st, 2nd, 3rd and 4th week with highest at the end of 1st week. **Conclusion:** Aspartate Aminotransferase level in gingival crevicular fluid changes with orthodontic tooth movement and may be used as biomarker during orthodontic treatment.

Keywords: Aspartate Aminotransferase, gingival crevicular fluid, orthodontic tooth movement

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INTRODUCTION

Tooth movement due to the application of orthodontic force is based on remodeling changes in the periodontal and alveolar tissues.¹ Two processes engaged in orthodontic tooth movement are bending of bone and periodontal tissues remodeling, including the periodontal ligament (PDL), gingiva and alveolar bone. The functional force causes the compression of the PDL and alveolar bone through pressure and on the opposite side there is a tension which stretches the PDL.²

Orthodontic forces modify the vascularity of periodontal tissue resulting in the synthesis of many metabolites and signaling molecules. The liberated molecules produce cellular response around the teeth for bone remodeling. Orthodontic tooth movement also involves the damage of epithelial cells of the pocket lining as well as the degradation of connective tissues.^{3,4}

Mechanical loading also alters vascularity of periodontal tissue and blood flow, resulting in the

synthesis and secretion of many molecules, such as cytokines, enzymes, growth factors and neurotransmitters. Due to the release of many molecules which are biologically active, a number of biomarkers are proposed to take part in orthodontic tooth movement for improving treatment and reducing side effects.⁵

Expression of these observable products can be found in the gingival crevicular fluid (GCF) of moving teeth, with significant increase in the level of its components like neurotransmitters, cytokines, growth factors, and cytosolic enzymes etc.^{6,7,8} Attention is focused on composition of GCF and its changes found during orthodontic tooth movement (OTM). Many biomarkers of GCF are reported representing remodeling of bone during orthodontic tooth movement.^{9,10}

Aspartate Aminotransferase (AST) is one of these biomarkers and variation in its level shows the biological activity in the periodontium during orthodontic treatment. It is a cell cytosolic enzyme released in blood after necrosis of cell.¹¹ Aspartate

Aminotransferase would be expected to pass from periodontal tissues as inflammatory exudate in to gingival crevicular fluid. Increased level of this enzyme may provide evidence of cell death within the periodontal tissues.^{12, 13} Elevated level of AST in GCF was associated with active sites of tissue damage as a result of inflammation or orthodontic tooth movement.^{14, 15} It was hypothesized that change in AST levels in GCF during orthodontic tooth movement is greater as compared to control.

Objective:

To find out the changes in Aspartate Aminotransferase level in gingival crevicular fluid during orthodontic tooth movement and compare with control.

METHODOLOGY

Study design: Randomized control trial. A split mouth design

Setting: Department of Orthodontics, Dental Section, Punjab Medical College Faisal Abad.

Duration of study: Six months 01-01-2016 to 30-06-2016.

Sample size: Thirty selected patients

Sampling technique: Non –Probability purposive sampling

Inclusion Criteria:

- Both sexes
- Age range 9-15 years
- Patient's with good general and periodontal health
- Full mouth plaque score (FMPS) less than 20%
- Full mouth bleeding score (FMBS) less than 20%
- Periodontal pocket depth less than 4mm

Exclusion Criteria:

- Patients with any history of disease
- Patients taking anti-inflammatory drugs at least one month before and during study

Data collection procedure:

Thirty patients according to inclusion criteria were included. Informed consent was taken from parents or guardian of patients. Patients underwent a session of supra and sub gingival scaling one week prior to baseline examination. Assessment of FMPS and FMBS was done on base line and end of 4th week. Brackets (Ortho organizers® Roth prescription 0.022x 0.028) were bonded on all teeth between right 1st molar to left 1st molar skipping left upper 1st premolar. Ni-Ti 0.014 wire ligated into the brackets and patients undergoing orthodontic

treatment in the initial stage of alignment were enrolled. A split mouth study design was selected with right upper 1st premolars chosen as experimental while left upper 1st premolars as control. These teeth chosen for data collection along with their gingiva were dried with light pressure of oil free air and the field was isolated by cotton rolls. A paper point was then inserted to a depth of 1mm into the gingival sulcus of the test and control teeth, held in place for 1min then removed and stored in test-tube containing 2ml of normal saline. Three samples were taken from each tooth and paper points without blood contamination used as test sample. Samples were taken on base line then after one hour, end of 1st, 2nd, 3rd and finally 4th week of activation.

Paper points were incubated in the test tubes for 15min, centrifuged and then thrown out. Level of AST was determined by a spectrophotometric automatic apparatus (Roche Hitachi 912®) and all values recorded.

Data Analysis:

Data was analyzed using SPSS version 18. Mean \pm S.D was calculated for quantitative variables like age, AST level in GCF for test and control group at baseline, after 1hour, 1st, 2nd, 3rd and 4th week while FMPS and FMBS level at baseline and 28th day. Independent sample t- test was used to analyze AST levels in test and control group. Paired sample t-test was applied to compare mean AST level at baseline, after one hour, end of 1st week, 2nd week, 3rd week and finally 4th week of activation. p-value <0.05 was considered significant.

RESULTS

Age statistics are presented in table 1. The mean \pm SD with minimum and maximum values at baseline and 28th day for FMPS and FMBS are mentioned in table 2 and 3.

Mean \pm SD of AST levels for test and control groups at base line then after one hour, end of 1st week, 2nd week, 3rd week and finally 4th week of activation given in table 4. Comparison of AST level between test and control group with level of significance tabulated in table 5. The difference between test and control groups at baseline was statistically insignificant (p-value 0.865). The difference of mean AST levels after 1 hour between the two groups was also statistically insignificant (p- value 0.852).

The difference of mean AST levels at the end of 1st week, 2nd week, 3rd week and 4th week between the test and control groups was statistically significant (p- value of 0.001).

Table 1: Age of the patients

	N	Minimum	Maximum	Mean	Standard deviation
Age of patients	30	9	15	11.93	2.08

Table2: Minimum, maximum, mean and standard deviation in FMPS

FMPS	N	Minimum	Maximum	Mean	Standard deviation
Baseline	30	1	20	9.43	6.00
28 th day	30	1	20	10.73	6.34

Table3: Minimum, maximum, mean and standard deviation in FMBS

FMBS	N	Minimum	Maximum	Mean	Standard deviation
Baseline	30	1	20	10.47	5.96
28 th day	30	1	20	9.60	6.05

Table 4: Comparison of mean change in AST level between test and control group

AST (mU/s)in GCF	Mean± SD in test group	Mean± SD in control group	P value
Baseline	16.659±3.383	16.510±3.384	0.865
1hr	16.979±3.383	16.533 ± 3.378	0.852
1 st week	35.63 ±3.164	16.587 ± 3.433	0.0001
2 nd week	31.527 ±2.586	16.570 ± 3.421	0.0001
3 rd week	27.650 ±3.122	16.600 ± 3.431	0.0001
4 th week	23.080 ±2.240	16.604 ± 3.435	0.0001

p-value < 0.05 = Significant

DISCUSSION

Orthodontic tooth movement is an inflammatory mechanism characterized by cell death, osteoclastogenesis and osteogenesis. AST is an intracellular enzyme that is used to mark cell death. Its potential to be used as a biomarker has not been explored extensively although its activity had been noted to increase with increasing force.¹⁶

According to our study there was slight increase in the baseline FMPS reading as compared to the reading taken at 28th day. On the other hand, the FMBS reading showed slight decrease from baseline to reading taken at 28th day.

It is reported that FMPS and FMBS are used to evaluate the periodontal status and oral hygiene. A study showed that an increase in FMBS and FMPS decreases the salivary pH and buffering capacity, thus reducing the salivary protective

effect.¹⁷ Another study assessed the FMPS, and FMBS at the start of treatment and after 12 months then found that scores of FMBS and FMPS were significantly decreased in the group treated with Invisalign as compared to the group treated with fixed brackets.¹⁸

Comparison of AST level in GCF for test and control groups showed insignificant difference between the two groups at base line and 1 hour samples. The findings were significant at the end of first, 2nd, 3rd, and 4th week and found highest at the end of 1st week. Subsequently levels started to decrease after that but were still significantly greater than their control counterparts. Levels in the control group remained constant throughout the study.

In this study, the activity of AST in GCF of the test teeth increased significantly and noted at its highest level by the end of the first week as compared to

control group. Subsequently the next three weeks showed that the activity of the enzyme gradually reduced although level was still significantly higher than the control group.

In the study conducted by Pernitti et al, the mesial and distal aspect of teeth showed different activities due to the distally directed force.¹⁹ Present study concentrated on the alignment stage where the tipping force doesn't have specific direction. In another similar study conducted to find AST assays in GCF during leveling and alignment stages, the results obtained correlated with the present study. They went a step further and compared the results between adolescents and adults and found non-significant difference between the two groups.²⁰ In another study conducted by the same authors on AST assays during bodily movement where they retracted canines with separator method; the results of that study were however different as level increased at the end of first week and kept on increasing till the fourth week.²¹ This difference in results can be explained by the fact that in their case, application of greater force had caused the inflammatory process to continue until the fourth week. The authors of all these studies came to the same conclusion that AST does depict the degree of orthodontic tooth movement and definitely has the potential to serve as a biomarker for this purpose.

Our study is in line with another study who reported that the level of AST in GCF is significantly increased in both compression and tension sites at days 7 and 14. This rise may be due to the controlled trauma, which produces cell death as a result of mechanical force applied on PDL and alveolar bone.²⁰ Present study is also comparable with another study that was conducted to evaluate AST level in GCF during canine distalization where similarly, level increased on compression site.²² An increase in AST level showed the exertion of orthodontic force on teeth and is directly linked with the site of compression during orthodontic tooth movement.^{16, 17}

Therefore, these findings showed that the increase in activity of AST in the GCF had reflected the force levels applied in four weeks. The pattern of activity of AST can also be explained by the usual practice of reactivating the appliance every 4-6 weeks as it is believed that the force becomes too low to induce anymore tooth movement beyond this period of time.

CONCLUSION

It was concluded that Aspartate Aminotransferase level increase with increasing force during orthodontic treatment; therefore level of this enzyme in gingival crevicular fluid may be used as a

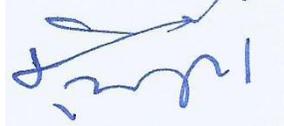
biomarker to find out changes in periodontium during orthodontic tooth movement.

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