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Evaluation of salivary Aspartate Aminotransferase Enzyme level in Smoker Patients with Peptic Ulcer in Relation to Periodontal Condition.

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ABSTRACT

Periodontal diseases are inflammatory condition result from interaction between pathological bacteria and the host defense system that cause tissue damage while, peptic ulcer is one of gastrointestinal diseases which involve ulceration in the epithelium lining of the stomach or duodenum or both. Smoking is a great problem that cause severe damage and complications in the human body such as periodontium and gastrointestinal system. Aspartate aminotransferase (AST) enzyme is frequently called liver enzyme and its activity increase in many diseases such as diabetic mellitus, peptic ulcer, chronic periodontitis, and others. Evaluate and compare the level of salivary AST enzyme and the periodontal health status by measuring the clinical periodontal parameters (plaque index (PLI), gingival index (GI), bleeding on probing (BOP), probing pocket depth (PPD), clinical attachment level (CAL) and tooth loss(TL)), at the study groups (group of smoker patients with peptic ulcer, group of non smoker patients with peptic ulcer, group of non smoker patients and without peptic ulcer (each group subdivided into gingivitis and chronic periodontitis), and control group), and correlate between the level of AST salivary enzyme with clinical periodontal parameters. Salivary AST enzyme level and clinical periodontal parameters (PLI, GI, BOP, PPD, CAL, and TL) were recorded from 140 males, age range (25-40) years old, that divided into study groups which include ; group of 40 smoker patients with peptic ulcer group, group of 40 non smoker patients with peptic ulcer group and group of 40 non smoker patients and without peptic ulcer , then each of these groups subdivided into(20 had gingivitis and 20 had chronic periodontitis),also control group which consists of 20 subjects without peptic ulcer and non smoker with healthy periodontium. The results showed that the highest median value of plaque index was in patients with gingivitis ,peptic ulcer and smoker, while the median values of gingival index and bleeding on probing were highest among patients with gingivitis, peptic ulcer and non -smoker. The highest median values of probing pocket depth ,clinical attachment level and tooth loss were recorded in patients with chronic periodontitis ,peptic ulcer and smoker. The median value of salivary enzyme AST level (39.25) was found to be highest in patients with chronic periodontitis, peptic ulcer and smoker. All of the clinical periodontal parameters as well as salivary AST enzyme level demonstrated highly significant differences in the comparisons among the groups and subgroups. The correlations between the level of salivary AST enzyme with clinical periodontal parameters was almost statistically non significant for the study subgroups and control group**.** It can be concluded that the susceptibility for and the severity of periodontal diseases were significantly increase in patients with peptic ulcer or smoker through demonstrating more periodontal tissue destruction, and the level of salivary AST enzyme increase with the increase in the severity of periodontal diseases as well as the presence of peptic ulcer and smoking; accordingly, these results can suggest that salivary AST enzyme was considered as a good biochemical marker of tissue destruction and this provide better chance in diagnosis, monitoring the efficacy of the management of periodontal diseases and peptic ulcer.

Keywords: Periodontal diseases, peptic ulcer, smoking, AST salivary enzyme.

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INTRODUCTION

Periodontal diseases are the inflammation which are affecting the supporting tissues of the tooth, result from a shift in the microbial consistency within the oral cavity and may lead to loss of teeth **(1).** Dental plaque is the main etiological factor of the periodontal diseases while dental calculus it is one of the retentive factor of plaque **.**There are two main types of periodontal diseases: gingivitis and periodontitis. Gingivitis is the first and more stable type of periodontal disease. It is characterized by reversible inflammation of soft tissue which surrounded the teeth without loss of attachment and without pocket formation **.** Periodontitis is consider irreversible chronic inflammation of supporting tissues of the teeth caused by specific types of microorganisms mainly anaerobic, gram negative strains form subgingivally in the periodontal pockets**(2)**. Periodontitis characterized by loss of attachment and alveolar bone loss with or without gingival recession**(3).** Peptic ulcer is the ulceration that take place in the epithelium lining of the stomach (gastric ulcer) or duodenum (duodenal ulcer)**(4) .** Peptic ulcer will develop when there is imbalance between gastric acid secretion and gastroduodenal mucosal defense because of some aggressive factors such as non- steroidal anti-inflammatory drugs(NSAIDs), *Helicobacter pylori* bacteria (*H. pylori*), alcohol, smoking, bile salt, gastric acid, and pepsin. Many methods used in diagnosis of peptic ulcer divided into specific and non-specific, but the most specific method is gastrointestinal endoscope **(5)**. Smoking have a great effect on both periodontium and gastrointestinal system. It increased periodontal tissues destruction causing alveolar bone loss , gingival recession, increase mobility and furcation involvement because it interfere with vascular and immunological functions which impair the defensive functions of the periodontal tissues**(6)** . In gastrointestinal system , smoking participate in development and maintaining peptic ulcer by decrease gastric mucosal blood flow and constrict small blood vessels in the stomach**(7)**. Saliva is an exocrine secretion produced by salivary glands , considered a good biomarker for health and disease state **(8)** . Aspartate aminotransferase enzyme (AST) is named liver enzyme. In medicine, it is a good biomarker for diagnosis of many clinical conditions such as hepatic disease, diabetic mellitus, myocardial infraction , and others. The AST liberate from dead cells into extracellular fluid and evaluated in serum, saliva, tears, and gingival crevicular fluid (GCF) . The level of AST enzyme elevated in periodontal disease**(9 ,10)** as well as in patients with peptic ulcer **(11)** . The activity of this enzyme also increased in smokers when it is compared with non- smokers **(12) .** Hence, there was no pervious study concerning the correlation between periodontal health status, peptic ulcer, smoking and the level of AST salivary enzyme among men so, this study was designed and conducted**.**

MATERIALS AND METHODS

The human samples included 140 males age from (25-40) years old. They were selected from the Department of Periodontology at the Teaching Hospital of College of Dentistry, University of Tikrit and from Baghdad/ Digestive System and Liver Teaching Hospital**.** Inclision criteria included ;20 teeth present for each patient, patients with peptic ulcer (PU) diagnosed by gastrointestinal specialist via endoscope and smoker patients who smoke >10 cig\day for the last five years **(13)**. While the exclusion criteria were females, alcoholism, patients receive medication for treatment of peptic ulcer, had other systemic disease, undergone periodontal treatment or course of anti-inflammatory, anti microbial or other medications in the 3 months before the study. subject's acceptance documented by signing on specially designed informed consent. When the subjects had been selected , the whole un stimulated saliva collected after that the examination of clinical periodontal parameters (plaque index PLI **(14)**, gingival index GI **(15)**, bleeding on probing BOP**(16),** probing pocket depth PPD **(16),** clinical attachment level CAL**(17),** and tooth loss TL**(18)** were carried by using the Michigan O periodontal probe on four surfaces (mesial, distal, lingual/palatal, buccal/labial) of all teeth except third molars that were excluded . Subjects shared in the study divided into four study groups:

1- **Group (no.=40)** : consisted of patients with peptic ulcer and smoker.
2- **Group(no.=40)** : consisted of patients with peptic ulcer and non smoker.
3- **Group(no.=40)** : consisted of patients without peptic ulcer and non smoker.

Each group subdivided according to periodontal health status into:
Gingivitis (Ging.) subgroup(no.=20).
Chronic periodontitis (CP) subgroup(no.=20).
Gingivitis: : was defined by the presence of signs and symptoms of gingival inflammation **(15),** and without periodontal pocket or clinical attachment loss.

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Chronic periodontitis: was defined by the presence of at least four sites with PPD≥4mm and clinical attachment loss of (1-2mm) or more, this made according to the system of international classification for periodontal disease (PD) **(19) .**

4-**Control group(no.=20)** : consisted of subjects without peptic ucer and non smoker with clinically healthy periodontium.

Healthy periodontium: was defined by the absence of any signs and symptoms of gingival inflammation and without periodontal pocket or clinical attachment loss.

The whole unstimulated saliva samples about 3ml were collected**(20)**. Samples were subjected to centrifuged at 1000 rpm for 10 minutes then the supernatant saliva stored at -20 °C (freezer) until biochemical analysis of AST enzyme. The statistical analysis included were descriptive (median and percentage (%)) and inferential (Kruskal-Wallis H test , Mann-Whitney U test , and Simple person's correlation coefficients (r). In the statistical tests, the levels of significant(S) when 0.05≥P≥0.01, non significant (NS) when P>0.05, while highly significant (HS) when P≤ 0.01.We certify that this study involving human subjects is in accordance with the Helsinki declaration of 1975 as revised in 2013 and that it has been approved by the relevant institutional Ethical Committee**(21)**.

RESULTS

Table (1) showed that the highest median value of PLI (3.53) was at Ging.+PU+Smoker, while the highest median values of GI and BOP (3.39, 30.7) respectively were at Ging.+PU+Non-smoker. The highest median values of PPD, CAL, and TL (6.62, 6.87, 18.19) respectively were at CP+PU+Smoker ; hence, highly significant differences were revealed among the study subgroups for all clinical periodontal parameters . Table (2) showed the comparisons of median values of PLI and GI between each pair of the study subgroups which detected almost highly significant differences **.** However for GI significant differences showed between CP subgroup with both Ging.+PU+Smoker and CP+PU+Smoker. The comparisons in table (3) of the median percentages of BOP score 1 and TL between each pair of the study subgroups demonstrated almost highly significant and significant differences. Table (4) showed the comparisons of median values of PPD and CAL between each pair of study subgroups which were highly significant differences, while a significant difference revealed between CP+PU+Non-smoker with CP for PPD. The highest median value of salivary AST enzyme (39.25) was at CP+PU+Smoker , demonstrated in table (5) with highly significant difference among the study subgroups and control group. Table (6) revealed the comparisons of the median values of salivary AST enzyme between each pair of study subgroups and control group, which detected almost highly significant differences. Almost non-significant weak correlations between AST enzyme with each one of the clinical periodontal parameters were demonstrated, however, significant moderate positive correlation with BOP at CP+PU+Smoker hence, Tooth Loss revealed moderate negative correlation at Ging.+PU+Smoker and CP but it was significant moderate positive at CP+PU+Non-smoker while, significant strong positive correlation was detected at Ging.+PU+Non-smoker, as showed in table (7).

Table 1: Statistical analysis of the clinical periodontal parameters of the study subgroups and control group.

Groups and subgroups	PLI	GI	BOP score 1	PPD	CAL	TL
	Median value	Median value	Median %	Median value	Median value	Median %
CP+PU+Smoker	2.05	1.52	17.95	6.62	6.87	18.19
Ging.+PU+Smoker	3.53	2.56	23.25	-	-	1.603
CP+PU+Non-smoker	1.58	1.91	25.84	5.32	5.42	11.75
Ging.+PU+Non-smoker	3.06	3.39	30.7	-	-	1.06
CP	1.06	1.09	18.55	4.12	4.47	3.73
Ging.	2.55	2.02	28.95	-	-	0.35
Control group	0.39	0.35	-	-	-	0
Kruskal –wallis H test without control group	115.77	112.86	106.64	40.06	39.84	31.75
P-value	0.000	0.000	0.000	0.000	0.000	0.000
Sig.	HS	HS	HS	HS	HS	HS

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Table 2: Comparisons of the median values of PLI and GI parameters between all pairs of the study subgroups.

Subgroups		Mann-whitney U test for PLI	P-value	Sig.	Mann-Whitney U test for GI	P-value	Sig.
Ging.+PU+Smoker	CP+PU+Smoker	66.04	0.000	HS	58.45	0.000	HS
	Ging.+PU+Non-smoker	24.52	0.06	NS	22.19	0.078	NS
	CP+PU+Non-smoker	43.05	0.004	HS	46.06	0.004	HS
	Ging.	40.45	0.000	HS	39.3	0.007	HS
	CP	38.32	0.003	HS	21.65	0.05	S
CP+PU+Smoker	Ging.+PU+Non-smoker	81.62	0.000	HS	77.5	0.000	HS
	CP+PU+Non-smoker	21.4	0.069	NS	17.5	0.119	NS
	Ging.	104.5	0.000	HS	97.5	0.000	HS
	CP	54.6	0.004	HS	37.7	0.015	S
Ging.+PU+Non-smoker	CP+PU+Non-smoker	63.5	0.000	HS	60	0.000	HS
	Ging.	28.9	0.069	NS	19	0.078	NS
	CP	40	0.000	HS	40.55	0.005	HS
CP+PU+Non-smoker	Ging.	88.33	0.000	HS	81	0.000	HS
	CP	25.7	0.06	NS	22.8	0.067	NS
Ging.	CP	67.2	0.000	HS	75.9	0.000	HS

Table 3: Comparisons of the median values of BOP score 1 and Tooth Loss parameters between all pairs of the study subgroups.

Subgroups		Mann-Whitney U test for BOP score 1	P-value	Sig.	Mann-Whitney U test for TL	P-value	Sig.
Ging.+PU+Smoker	CP+PU+Smoker	22.79	0.006	HS	24.91	0.003	HS
	Ging.+PU+Non-smoker	15.34	0.004	HS	5.25	0.600	NS
	CP+PU+Non-smoker	11.054	0.000	HS	12.61	0.05	S
	Ging.	14.605	0.000	HS	9.25	0.450	NS
	CP	15.821	0.000	HS	5.36	0.966	NS
CP+PU+Smoker	Ging.+PU+Non-smoker	45.53	0.006	HS	17.86	0.025	S
	CP+PU+Non-smoker	23.007	0.000	HS	12.30	0.017	S
	Ging.	75.004	0.000	HS	34.16	0.028	HS
	CP	3.87	0.423	NS	24.55	0.001	HS
Ging.+PU+Non-smoker	CP+PU+Non-smoker	24.842	0.000	HS	30.16	0.000	HS
	Ging.	22.56	0.000	HS	4	0.744	NS
	CP	12.83	0.002	HS	5.61	0.509	NS
CP+PU+Non-smoker	Ging.	16.76	0.000	HS	21.86	0.04	S
	CP	9.94	0.03	S	12.25	0.04	S
Ging.	CP	16.747	0.000	HS	9.61	0.385	NS

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Table 4: Comparisons of the median values of PPD and CAL parameters between all pairs of the study subgroups.

Subgroups		Mann-Whitney U test for PPD	P-value	Sig.	Mann-Whitney U test for CAL	P-value	Sig.
CP+PU+Smoker	CP+PU+Non-smoker	18.95	0.001	HS	15.75	0.012	HS
	CP	33.85	0.000	HS	34.50	0.000	HS
CP+PU+Non-smoker	CP	14.9	0.016	S	18.75	0.002	HS

Table 5: Statistical analysis of the salivary AST enzyme concentration (IU/L) of the study subgroups and control group.

Groups and subgroups	Median	Kruskal-Wallis H test	P-value	Sig.
CP+PU+Smoker	39.25	133.362	0.000	HS
Ging.+PU+Smoker	25			
CP+PU+Non-smoker	36.25			
Ging.+PU+Non-smoker	21.5			
CP	25.15			
Ging.	18.2			
Control group	12.85			

Table 6: Comparisons of the median values of salivary AST enzyme between all pairs of the study subgroups and control group

Groups and subgroups		Mann-Whitney U test	P-value	Sig.
Ging.+PU+Smoker	CP+PU+Smoker	50.4	0.002	HS
	Ging.+PU+Non-smoker	30.0	0.402	NS
	CP+PU+Non-smoker	31.3	0.015	S
	Ging.	48.1	0.004	HS
	CP	1.5	0.907	NS
	Control group	69.1	0.000	HS
CP+PU+Smoker	Ging.+PU+Non-smoker	80.4	0.000	HS
	CP+PU+Non-smoker	19.1	0.136	NS
	Ging.	98.5	0.000	HS
	CP	48.9	0.000	HS
	Control group	119.5	0.000	HS
Ging.+PU+Non-smoker	CP+PU+Non-smoker	61.3	0.000	HS
	Ging.	18.1	0.157	NS
	CP	31.5	0.019	S
	Control group	39.1	0.002	HS
CP+PU+Non-smoker	Ging.	79.4	0.000	HS
	CP	29.8	0.02	S
	Control group	100.4	0.000	HS
Ging.	CP	49.6	0.002	HS
	Control group	21	0.100	NS
Control group	CP	70.6	0.000	HS

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Table 7: Correlation between the levels of salivary AST enzyme with the clinical periodontal parameters at each study subgroups and control group

Groups and subgroups	Statistical Analysis	PLI	GI	BOP Score1	PPD	CAL	Tooth Loss
CP+PU+Smoker	r	-0.317	0.156	0.511	0.063	0.076	-0.078
	p	0.173	0.513	0.021	0.790	0.790	0.782
	Sig.	NS	NS	S	NS	NS	NS
Ging.+PU+Smoker	r	-0.279	0.011	-0.135	.	.	-0.544
	P	0.234	0.862	0.569	.	.	0.456
	Sig.	NS	NS	NS	.	.	NS
CP+PU+Non-smoker	r	-0.051	-0.297	-0.040	0.217	0.188	0.583
	p	0.832	0.204	0.889	0.356	0.429	0.023
	Sig.	NS	NS	NS	NS	NS	S
Ging.+PU+Non-smoker	r	-0.068	-0.177	-0.290	.	.	0.943
	P	0.783	0.489	0.215	.	.	0.057
	Sig.	NS	NS	NS	.	.	S
CP	r	0.029	0.269	-0.165	0.043	-0.034	-0.455
	P	0.902	0.252	0.483	0.857	0.886	0.219
	Sig.	NS	NS	NS	NS	NS	NS
Ging.	r	-0.143	-0.097	0.245	.	.	.
	P	0.548	0.683	0.297	.	.	.
	Sig.	NS	NS	NS	.		.
Control group	r	-0.171	0.223-		.		
	P	0.471	0.725	.	.		
	Sig.	NS	NS		.		

DISCUSSION

The results showed that the median value of the PLI was highest at Ging.+PU+Smoker subgroup with a highly significant difference among the study subgroups. This may be due to the leading role of dental plaque in the pathogenesis of periodontal diseases, as the primary etiological factor of gingivitis is poor or ineffective oral hygiene, which leads to the accumulation of dental plaque. Smoking was considered a major risk factor for development and progression of periodontal diseases**(22),** and found that smokers had significantly higher mean values of PLI as compared with non smokers **(23)**. The *H. pylori bacteria* was detected in high percentage at supragingival plaque in peptic ulcer patients **(24)**. This study showed highly significant differences among the study subgroups, although the increased gingival inflammation represented by highest median values of GI and BOP sites score (1) were found among Ging.+PU+Non-smoker subgroup. The absence of good oral hygiene lead to accumulation of dental plaque and subsequent gingival inflammation with development of gingivitis; however, gingival inflammation may be linked to systemic inflammation such as gastric ulcer**(25)**. The oral biofilm consists of different microbiota, and one of the microorganisms present in dental plaque is *H. pylori* hence, the mouth considered the first extra-gastric reservoir for this bacteria that lead to reinfection of stomach **(26)**. There was at least one strain of *H. pylori* isolated from the plaque was genetically closely related to identical strain from the stomach and this bacteria in the dental plaque had the potential for gastric infection **(27)** . The patients with peptic ulcer (gastric and duodenal ulcers) had higher periodontal index including bleeding on probing when compared with controls **(28)** . Nicotine in cigarette cause vasoconstriction in gingival blood vessels which decreased bleeding on probing hence, reduced inflammatory response had been attributed to the alterations in gingival microvasculature and gingival epithelium in smokers, so there was decrease in the number of bleeding sites in smokers as compared with non smokers **(29)** . The median values of PPD and CAL were found to be highest at CP+PU+Smoker subgroup than other subgroups with statistically highly significant differences. Patients with peptic ulcer demonstrated increase in PPD and CAL associated with gingival recession when compared with non peptic ulcer group and those patients had PPD>4mm and loss of clinical attachment **(30)** ; however, it was revealed that patients with PPD≥5mm had higher quantity of *H. pylori* bacteria in subgingival plaque that cause reinfection of the stomach which lead to development of peptic ulcer also showed that patients with CP harbor a number of *H. pylori* in subgingival plaque which significantly higher than subjects without periodontitis**(3)** . Smoking is one of the major risk factor that contribute in the pathogenesis of peptic ulcer and periodontal diseases. In peptic ulcer , smoking impair the therapeutic effect of histamine 2-antagonist, stimulate the pepsin secretion, promote

reflux of duodenal content into the stomach , increase the risk of harmful effect of *H. pylori,* and increase the production of free radicals and platelet activating factor, also smoking caused delay of healing of peptic ulcer via decrease gastric mucosal blood flow**(31)** . On the other hand, smoking increase the risk for periodontal disease progression because it interferes with vascular and immunological functions which impair the defensive functions of the periodontal tissues by weaken the inflammatory and immune responses to periodontal pathogen, cause defect in the neutrophil function so that smoking increase pocket formation and clinical attachment loss as well as alveolar bone loss **(32)** .There were significant differences with higher mean values of the clinical periodontal parameters(PLI, PPD, and CAL) in smokers group compared to non smokers both with CP **(33)** .The highest median percentage value for TL detected in CP+PU+smoker subgroup. Patients with peptic ulcer had number of missing teeth about five or more teeth **(18)** however, men who smoked cigarette had increase in risk of TL , and this risk decreased upon smoking cessation, hence, the risk of TL is greater among cigarette smokers with CP than among CP non-smoker patients , in addition they observed that current male smokers had more teeth with calculus, tooth mobility, probing pocket depth ≥4mm, filled and decayed teeth than non smokers **(34)**. The results of the current study showed that the median value of salivary AST enzyme was highest at CP+PU+Smoker with a highly significant difference among the study subgroups and control group. The level of AST salivary enzyme was significantly increased in patients with chronic periodontitis when compared with control group **(35)**. The level of AST salivary enzyme used among the important biochemical marker that release in response to some systemic diseases hence, the level of this enzyme was significantly increased in patients with peptic ulcer as compared with non peptic ulcer subjects **(36)** . The level of this enzyme also increased in smokers **(37)** . The AST enzyme is an intracellular enzyme included in the metabolic processes of the cells and it is present in the cells of soft tissues. The higher activity of this enzyme is indicator of higher events of cellular damage and the increased level in GCF and saliva is a consequence of its increased release from the damaged cells of the soft tissues of the periodontium, and is a reflection of the metabolic changes in the inflamed gingiva and dead of cells of periodontium **(35)** as well as in peptic ulcer, sores develop in the epithelium lining of the stomach so that in response to damaged cells the liberation of this enzyme will increase in the extracellular fluid such as saliva**(11)**.Smoking also cause a major destruction to the liver cells that lead to release of this enzyme from damaged cells and subsequent increase its level in fluid **(37)** . In the present study the statistical analysis revealed almost non-significant correlations, however, significant moderate positive correlation with BOP at CP+PU+Smoker hence, TL demonstrated moderate correlation which were significant positive at CP+PU+Non-smoker but non-significant negative at both Ging.+PU+Smoker and CP while, significant strong positive correlation was detected at Ging.+PU+Non-smoker. The non-significant correlations detected in this study, may be due to the presence of more inactive sites during the saliva collection or due to small human sample size. Abdul-Hadi J Mustafa and Alsafi A Khulood, 2009**(38)** found that there were non-significant correlations between salivary AST enzyme activity with GI and BOP in patients with CP . A study carried out by Totan *et al*., 2006 **(39)** revealed that there were statistically significant correlations between BOP and PPD with AST level in saliva at patients had CP .In conclusion, salivary AST enzyme is consider as a good biochemical marker of periodontal tissue destruction and measuring the level of this enzyme is usually easy, time saving and non-invasive method for screening and early diagnosis of CP and to evaluate the effect of peptic ulcer and smoking on periodontal health status.

REFERENCES

[1] Mattew R Boylan **,** Hamed Kalili, Edward S , Huang , Dominiques S, Micchaud , Jacques Lazard, Kanmndi J, Joshipure, Andrew T, Chan. 2014**.** A prospective study of periodontal disease and risk of gastric and duodenal ulcer in male health professional . Clinical and Translation Gastroenterology Feb.13, vol. 5(2):49.

[2] Carranza**,** Newman Taki, and Klokevold. 2015**.** Carranza's clinical periodontology. Elsvier Saunders, 12th edition.

[3] Sophia Marbiax , Assem Soudied , Maya Romani, Gnillauma Compard , Gills Amador, Zahahi Padrann. 2013. Helicobacter Pylori and periodontal disease: An update and proposal of a multidisciplinary clinical protocol. Open Journal of stomatology, 13 June, 3:318-322.

[4] Hayder M Iddan, Fawaz D Abdul-Razaq. 2011. Oral manifestation , Microbiological study and enzymes analysis in patients with peptic ulcer . J Baghdad collage of Dentistry, vol.23 (2).

[5] Anand MD.2017. Peptic ulcer disease. Gasteroenterology. American association for the study of liver disease. American gasteroenterological association , American society for Gastro intestinal Endoscopy. Jan. 29, p:1-6

[6] Tajinder Kaur Saggu, KMK Masthan, Mahesh Pundaleek Dudanakar, Shams UI Nisa, Shankargouda Patil. 2012. Evaluation of salivary antioxidant enzymes among smoker and non- smoker. World Journal of Dentistry, March; 3(1):18-21.

[7] Gareth A. Thomas, John Rhodes, John R .2005. Mechanisms of disease : nicotine. A review of it is action in the context of gastrointestinal disease . Nat Clin Pract. Gastroenterol Hepatol,2(11): 536-544.

[8] Simranjit Singh, Venkatapathy Ramesh, Nirima Oza, Pemagaram, Dhanasekaran Balamurali, Karthikshree, Vishun Prashad and Premalatha, Balakrishnan. 2014. Evaluation of serum and salivary lipid profil: A correlation study. J oral and maxillofacial pathology, April; 18(1):4-8.

[9] Akashi Dewan Pankaj Bhatia.2011. Evaluation of aspartate aminotransferase enzyme level in saliva and gingival crevicular fluid with periodontal disease progression. A pilot study.J clin periodontal, May, vol. 3, P.1-6.

[10] Praveen Kudya, Neha Sainia, Hema Kudya and Varun Saini. 2014. Estimation salivary aspartate aminotransferase levels in chronic periodontitis and chronic gingivitis prior and following non-surgical periodontal therapy : A clinico –biochemical study. J Indian soc periodontal, Feb. ; 18(1):53-58.

[11] Moslem Boghori , Mahmoudreza Aghamaal, Reyhaneh Sariri, Fahime Mohamadpour, Hosein Ghafour. 2014. Salivary enzyme and flow rate : Markers of peptic ulcer . Journal of oral biology and craniofacial research (4): 24-29.

[12] Wamamethee SG, Shaper AG.2010. Cigarette smoking and serum liver enzyme , the role of alcohol and inflammation . Ann clin Biochemistry , July, 47 (4):321-6.

[13] Maffei G Brouwer N. Dolman KM, Van der velden U. Roos D. Loose BG. 2005. Plasma levels of mannan-binding lectin in relation to periodontitis and smoking. J periodontal , 76: 1881-9.

[14] Sliness J and Löe H. 1964. Periodontal disease in pregnancy II. Acta. Odonto Scand, 24:747-749.

[15] Löe H .1967. Gingival Index , the plaque Index and the retention index system. J Clin periodontal 38:610-616.

[16] Carranza, Newman Taki, and Klokevold. 2012. Carranza's clinical periodontology. Elsevier Saunders, 11th edition (chapter 4, P 41-42) and (chapter 27, P 305-308).

[17] American Academy of periodontology (AAP).1999. Parameters on chronic periodontitis with slight to moderate loss of periodontal support (supplement). J periodontal.volume 71 no. 5, P 853-855.

[18] Chei-Kaneto, Satoshi Toyokawa, Kazio Inone, Marko Inone, Toshihiko Senba, Yasuo Suyama, Yuji Myoshi, Yauski Kobayashi. 2012. Association between periodontal disease and peptic ulcers among Japanese workers: My health up study . Global Journal of health science, March, vol. 4, no. 2 , P:42-50.

[19] Lang NP, Bartold PM, Cullinam M et al.1999. International classification workshop. Consensus report: chronic periodontitis. Annals of periodontology, 4:53.

[20] Bradely Stephen Henson and David T Wong.2010.Collection, storage, and processing of saliva samples for downstream molecular application. Spring science business media,26 July, vol.666,P. 21-33.

[21] World Medical Association .2013. Declaration of Helsinki Ethical Principles for Medical Research Involving Human Subjects. JAMA.310(20):2191

[22] Ali H. Omer. 2015. The effect of cigarette smoking on salivary IgA and periodontal disease. Journal of Baghdad collage of dentistry, vol. 27, issue 3, p. 116-119.

[23] Wali H zainab and Mahmoud H Faud. 2002. Tobacco exposure and clinical attachment loss and gingival recession among the fifth year dental students in Iraq. Journal of Baghdad collage of Dentistry, Vol. 13, no.8, P. 60-66.

[24] Gebara, EC, Faria CM, Pannuti, C, Chehter, L, Mayer, MP, and Lima, LA. 2006. Persistence of *H. pylori* in the oral cavity after systemic eradication therapy. Journal of clinical periodontology, 33: 329-333.

[25] Loose BG, Cranndijk J, Hoek FJ, Paulein ME, Wertheim-van Dillen PM and van der Velden U. 2000. Elevation of systemic markers related to cardiovascular diseases in the peripheral blood of periodontitis patients. J periodontal ; 71(10):1528-1534

[26] Isabel Adler, Andrea Muino, Silva Aghas, Laura Harada, Mariana Diaz, Adriana Lence , Mario Labborozzi, Juan Manual , Boris Elsner, Alejandra Avagnine , Valeria Denninghoff. 2014. Helicobacter Pylori and oral pathology relationship with the gastric infection. World Journal of Gastrenerology , Aug. 7, 20(29): 9922-9935.

[27] Desai HG, Gill HH, ShankranK, Mehta PR, Prabhu SR.1991. Dental plaque: a permanent reservoir of Helicobacter pylori. Scand J Gastroenterol, Nov.26 (11) p.1205-1208.

[28] Namiote DE, Namiot Z, Kemona A, Golebiewska M.2006. Peptic ulcer and oral health status. Adv Med Sci., 51:153-5.

[29] Jalayer Naderi N, Semyari H, Elahinia. 2015. The impact of smoking on gingiva: a Histopathological study. Iran J patho, Summer;10(3):214-20.

[30] Khader YS**,** Umeda , M, Kobayashi, H, Takenchi, Y, Hayashi, J, Morotome-Hayashi, Y, Yano, K, Aoki, A, Ohkusa, T and Ishikawa. 2003. High prevalence of Helicobacter pylori detected by PCR in the oral cavities of periodontitis patients. Journal of periodontology, 74:129-134.

[31] Li Luf, Chan RL, LuL, Shen J, Zhang L, WnWk, Wang L, Hut, Li Mx, Cho CH.2014. Cigarette smoking and gastrointestinal disease: The causal relationship and underlying molecular mechanisms. Int. J Mol Med, Aug.; 34(2): 72-80.

[32] Underner M, Maes I, Urban T, Mentice JC. 2009**.** Effect of smoking on the periodontal disease. US National Library of Medicine, National institute of health, December; 26(1): 1-8.

[33] Ali G Basima and Ali H Omer. 2012. Detection of salivary flow rate and minerals in smokers and non smokers with chronic periodontitis (clinical and biochemical study). Journal of Baghdad collage of Dentistry, vol. 24, issue 1 P.:68-7.

[34] Krall EA, Garvey AJ, Garcia RI. 1999. Alveolar bone loss and tooth loss in male cigar and pipe smokers. Journal AM Association, Jan. 130(1): 57-64.

[35] Dabra S**,** China K, Kaushik A.2012. Salivary enzymes as diagnostic markers for detection of gingival /periodontal disease and their correlation with the severity of the disease. J India Soc. Periodontal, 16(3)358-364.

[36] Barkun A., Leotiadis G.2010. Systemic review of the symptoms burdea, quality of life impairment and costs association with peptic ulcer disease. Am J Med, 123(358-366).

[37] Elameen K Mohammed and Abdrabo A Abdul-Elkarim**.** 2013. Comparative study of liver enzymes activities in smokers and diabetic sudanneses patients. Asian Journal of biomedical and pharmacentical science ,Jan. 3(27):39-41.

[38] Abdul- Hadi J Mustafa and Alsafi A Khulood. 2010. Evaluation of salivary enzymes activities among patients with chronic periodontitis. Journal of Baghdad collage of Dentistry ,vol. 22(1):1-3.

[39] Totan A, Greabu M, Totan C, Spinu T.2006. Salivary aspartate aminotransferase, alanine aminotransferase, and alkaline phosphtase possible marker in periodontal diseases. Clin Chem. Lab med; 44(5); 612-5.