



## Research Article

# The Oral Health Status in relation to Salivary Antimicrobial Peptide in Pregnant Women

Saja Ali Hussein\*, Athraa Alwaheb

Department of Pediatric and Preventive Dentistry, College of Dentistry, University of Baghdad, Baghdad, Iraq

\* Corresponding author's email: [sajaalihussein92@gmail.com](mailto:sajaalihussein92@gmail.com)

## ABSTRACT

### Article history:

Received 1 October 2022

Accepted 15 November 2022

Available online 30 April 2023

<https://doi.org/10.47723/kcmj.v19i1.905>

**Keywords:** Dental caries, Salivary human,  $\beta$ -Defensin 2, Pregnant women.



This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license <http://creativecommons.org/licenses/by/4.0/>

**Background:** Oral health is a mirror of general health. During pregnancy, oral health is affected by changes in saliva and oral hygiene measures which may lead to more dental caries. Objective: Assess oral health status in relation to salivary antimicrobial peptides in pregnant women.

**Subjects and Method:** This cross sectional study was carried out in different primary health care centers in Rusafa sectors/ Baghdad city. The total sample included was 80 women (their age range from 21-30). The study group consisted of 40 pregnant women: half of them were in the first trimester and others were in third trimester, while the control group included 40 non-pregnant married females. Dental Plaque was recorded according to simplified oral hygiene index. Dental caries were diagnosed by using WHO (2013) criteria. Unstimulated salivary samples were collected. Chemical analysis of salivary samples was performed for the detection of human  $\beta$ -Defensin 2.

**Results:** The current study revealed that dental caries experiences (decayed surfaces(DS), missed surfaces (MS) and decayed, missing, filled surfaces(DMFS)) were higher among pregnant than non-pregnant females especially in the third trimester, add percentage with no significant differences, while filled surfaces (FS) were higher in the control group with also no significant difference. Regarding plaque index, results revealed that pregnant women had higher plaque index than non-pregnant women, with significant higher per-centage (53.3%) in the third trimester, P value = 0.027. The salivary human  $\beta$ -Defensin 2 was higher among the pregnant group in the third trimester with statistically no significant difference.

**Conclusion:** More plaque accumulation during pregnancy may explain the higher prevalence of dental caries as the dental plaque considered to be the chief contributing factor in dental caries.

## Introduction

Oral health is an important predictor of general health, well-being and quality of life which involves a variety of diseases and conditions that include dental caries, periodontal (gum) disease, tooth loss, oral cancer, and other less prevalent oral diseases (1). One of the most serious oral health issues is dental caries. Dental caries is

a biofilm-induced disease, caused by interactions between bacteria, host factors, and food (sugars), all of which influence the dynamic production of biofilms on tooth surfaces (2). It results in localized chemical breakdown of a tooth surface as a result of metabolic activity in microbial aggregates (dental plaque) (3). According to the

World Health Organization (WHO), over 2.4 billion individuals worldwide suffered from dental caries (4). However, caries onset and activity are complex. Dental plaque is considered to be the primary causative factor of dental caries. It is an oral microbial biofilm that is found on exposed tooth surfaces in the mouth (5). The levels of oral microorganisms in dental plaque and saliva, such as mutans streptococci, could be predicting for both caries activity and the transmission risk of the mutans streptococci which is an important etiological factor of caries (6).

Saliva, on the other hand, is a primary modifying factor (7), as it contains proteins and peptides for the protection and maintenance of oral health dynamics (8,9). Salivary human  $\beta$ -Defensin 2 is a powerful antimicrobial peptide formed by epithelial cells that plays a chief role in establishing a native immune response against bacteria and plays a few roles in maintaining a balance between health and disease (10).  $\beta$ -Defensin 2 is effective against a diverse range of bacteria, including Gram-positive, Gram-negative, and fungus, such as *S. mutans*, *Lactobacillus acidophilus*, *Actinobacillus actinomycetemcomitans* and *Porphyromonas gingivalis* (11).

Pregnancy is a distinct stage in a woman's life. Many complex physiologic changes occur during pregnancy, which might have an unfavorable effect on oral health (12). Oral health is affected by many systemic conditions. It is influenced by female steroid sex hormones during pregnancy (13). Pregnancy causes alterations in the salivary flow rate, pH of saliva, and biochemical composition (14). Accompanied by changes in food and oral cleanliness practices, which lead to an rise in dental caries (15).

The aim of current study was to assess oral health status in relation to salivary antimicrobial peptide in pregnant women.

## Subjects and Methods

This study was carried out in different primary health care centers in Rusafa sectors/ Baghdad city and during the period from the beginning of March,2022 till mid of May. The protocol of the study had been approved by ethical committee (No. 487, 19/ 1/ 2022). The total sample included was 80 women (females with an age range from 21-30). The whole sample involved was 80 females (ranging in age from 21 to 30 years). Forty pregnant women made up the study group, 20 of whom were in the first trimester and 20 in the third, while 40 married women who were not pregnant made up the control group. This study conducted on healthy looking, non-smoker and non-medicated women to avoid any confounding factors resulting from disease or its medicament which impact the accuracy of the results. So exclusion criteria were any women had systemic disease, take medication or smoker.

The collection of unstimulated whole saliva had been accomplished under resting situations in the morning (around 9 to 11 am) by spitting passively for 5 min into saliva collecting tubes (16). After saliva collection, salivary samples were centrifuged for 15 minutes at 3000 rpm and kept at -20 °C until conduct assessment. Salivary samples were then taken to the lab for a biochemical evaluation. Concentration of salivary human  $\beta$ -Defensin 2 was measured using ELISA kit. The simplified oral hygiene index stated by Greene and Vermillion was used to record dental plaque (17). Dental caries were

recorded by using Decay, Missing and Filled index (DMF) according to the criteria of WHO (18).

Analysis, interpretation, and presentation of the data were done using Statistical Package for social Science (SPSS version -22, Chicago, Illinois, USA). Results were presented as mean, standard error and the significance level was set at  $p < 0.05$ . Pearson's correlation coefficient ( $r$ ) was used to evaluate and compare correlations among groups.

## Results

Table 1 shows the mean values and standard errors of caries experience. Results showed that control group had higher filled surfaces than other two trimesters groups while in other caries experiences components (DS, MS, DMFS), the 3rd trimester group had higher values than the other groups with no significant findings.

**Table 1:** Descriptive and statistical test of caries experience among groups

Variables	Control		1st		F	P-value
	Mean	$\pm$ SE	Mean	$\pm$ SE		
DS	2.274	7.9	1.222	0.301 <sup>^</sup>	0.663	0.518 <sup>^</sup>
MS	1.471	5.5	2.631	0.014		
FS	0.578	1.4	0.363	2.857		
DMFS	2.82		19.6	2.947		

<sup>^</sup>=not significant at  $p > 0.05$

DS=Decayed surfaces; MS=Missed surfaces; FS=Filled surfaces; DMFS=Decayed, missing, filled surfaces

Table 2 presents the mean values of plaque index (PII) among groups and showed that it was higher in the 1st trimester than other two groups and lower in control group with significant difference among groups.

**Table 2:** Descriptive and statistical test of PII among groups

Variables	Control		1st		3rd		F	P-value
	Mean	$\pm$ SE	Mean	$\pm$ SE	Mean	$\pm$ SE		
PII	0.445	0.05	0.701	0.07	0.534	0.07	3.849	0.027*

\*=significant at  $p < 0.05$

PII=plaque index

Table 3 shows that the mean value of salivary  $\beta$ -Defensin 2 was lowest in first trimester followed by control while it was highest in the third trimester but with no significant difference.

**Table 3:** Descriptive and statistical test of hBD-2 among groups

Variables	1st		3rd		F	P-value		
	Mean	$\pm$ SE	Mean	$\pm$ SE				
HBD-2	0.832	0.04	0.78	0.052	0.885	0.052	0.997	0.375 <sup>^</sup>

<sup>^</sup>=not significant at  $p > 0.05$

Table 4 illustrates the relation of dental caries parameters with PII among groups. Results revealed that there was a positive weak not significant correlation between PII and caries experience in the control group while in the 1st trimester PII had a strong positive with DS and DMFS. In the 3rd trimester, there was a positive strong correlation between PII with DS and positive weak significance with DMFS.

Table 4: Correlation between caries experience by surfaces and teeth with PII among groups

Groups	PII					
	Control		1st		3rd	
	r	p	r	p	r	p
DS	0.215	0.183	0.671	0.001*	0.684	0.001*
MS	0.063	0.698	0.269	0.252	0.052	0.829
FS	0.283	0.077	0.215	0.363	0.242	0.303
DMFS	0.148	0.361	0.569	0.009*	0.449	0.047*

\*=significant at p<0.05

Table 5 demonstrates correlation between caries experience by surfaces and teeth with  $\beta$ -Defensin 2 among groups. There were either positive or negative weak not significant correlations between  $\beta$ -Defensin 2 and caries experience by teeth and surfaces except for MT, MS ,DMFS with hDB-2 in the 3rd trimester. The result of MT and MS with hDB-2 was a strong negative significant correlation. A negative weak significant correlation between DMFS and hDB-2 was recorded.

Table 5: Correlation between caries experience with hDB-2 among groups

Groups	HDB-2					
	Control		2nd		3rd	
	r	p	r	p	r	p
DS	-0.002	0.99	0.183	0.439	0.01	0.967
MS	-0.02	0.902	0.049	0.839	-0.544	0.013*
FS	-0.052	0.752	-0.299	0.2	0.286	0.222
DMFS	-0.023	0.889	0.091	0.701	-0.444	0.049*

\*=significant at p<0.05

## Discussion

During pregnancy, a variety of localized effects impacting the oral cavity were also observed. The effects of pregnancy on the hard and soft tissues of the mouth cavity had been well established (19). Dental caries is a common oral health condition in pregnancy because pregnant women are more prone to caries than non-pregnant women (15). The same results found in the current study which revealed that dental caries experience (DS, MS and DMFS) was higher among pregnant than non-pregnant women (especially in the third trimester) with non-significant differences. This is probably due to alteration in salivary physiochemical properties, and biochemical content, as well as variances in dietary and dental hygiene practices (20). In addition it has been reported that the number of certain salivary cariogenic microorganism such as streptococcus mutans increases as pregnancy progresses which is connected to a higher incidence of caries in third trimester (21). This accounted for the greater prevalence of dental caries in third trimester in present study. Additionally, there are some

psychological changes that take place during pregnancy, including mood swings and worry for the unborn child in the third trimester that will influence how pregnant women behave with regard to dental hygiene (22). The results of the present study were consistent with the other studies (23,24). Opposite results reported higher caries prevalence during the 2nd trimester with significant differences (25,26). Regarding FS, control group was higher than pregnant groups with significant differences. This finding was in line with Amin and Shetty (2014) study (27). This is due to the fact that the attitude of non-pregnant women is better than the pregnant women about the treatment of carious teeth, oral hygiene measures and attending dental services. In the present study, plaque index was significantly higher among pregnant than non-pregnant women among all groups. According to trimester, the current study indicated that dental plaque was highest in the first trimester, which is consistent with the findings of Al-Zaidi (2007) (25), while other Iraqi studies found highest values in the third trimester (28,29). This could be explained by the fact that pregnancy is a stressful state (30) that is accompanied with multiple physiological and psychological events that lead to higher self-neglect (31). A positive correlations of plaque with DS and DMFS in first and third trimesters were recorded in this study, as dental caries is a bio-film-associated disease (32).

Human  $\beta$ -Defensin 2 is a powerful antimicrobial peptide tends to be up-regulated by a variety of inflammatory stimuli including bacteria, bacterial products, and inflammatory cytokines (33). During pregnancy, human  $\beta$ -Defensin 2 levels in saliva relate to the exaggerated inflammatory response (34). In the current study, the salivary concentration of  $\beta$ -Defensin 2 found to be highest in third trimester with no significant difference. An increased in  $\beta$ -Defensin 2 may be attributed to higher prevalence of dental caries in third trimester. As streptococcus mutans is main bacteria that causes dental caries, a tendency in the rise of  $\beta$ -Defensin 2 level along with the increase in the overall content of bacteria from the mutans streptococci group was noticed (35). This immune response plays important role to protect the tooth structure from caries (36) this fact is supported by the result of present study in which positive correlation was found between DS and  $\beta$ -Defensin 2. A study of Barrera et al. (37) reported positive significant correlation between dental caries and  $\beta$ -Defensin 2 and concluded that high salivary levels and expression of  $\beta$ -Defensin 2 may represent a biological response of oral tissue to caries. While Al-Ali et al. (38) revealed a significant negative correlation between  $\beta$ -Defensin 2 with caries experience. A strong negative significant correlation between MS with  $\beta$ -Defensin 2 in the 3rd trimester was recorded in this study, as more teeth lose in the third trimester may reduce antigenic stimulation and subsequently  $\beta$ -Defensin 2 production. Since the secretion of  $\beta$ -Defensin 2 from epithelial cells is dependent on infectious or inflammatory stimuli (39). This consistent with Ribeiro et al.(40), who found decreased in  $\beta$ -Defensin 2 that correlated with lower average number of teeth.

## Conclusion

Prevalence of dental caries was the highest among pregnant group compared to control group. This could be due to poor oral hygiene status (more plaque buildup) along with alteration in salivary parameters which leads to increase prevalence of dental caries during pregnancy. The salivary concentration of  $\beta$ -Defensin 2

found to be highest in third trimester, this may explain as immune response to increased dental caries in third trimester.

The main limitations of the study were small sample size that affects the significances of the results and generalization. Additionally, due to the cross-sectional design, it was not possible to analyze the dynamics changes in pregnant women's dental health from first to third trimesters.

#### Funding

This research did not receive any specific fund.

#### Conflict of Interest

No conflict of interest

#### References

- [1] World Health Organisation. Oral health. Available at: [https://www.who.int/health-topics/oral-health#tab=tab\\_1](https://www.who.int/health-topics/oral-health#tab=tab_1) . 2022.
- [2] Gupta P, Gupta N, Singh HP. Prevalence of dental caries in relation to body mass index, daily sugar intake, and oral hygiene status in 12-year-old school children in Mathura city: A pilot study. *Int J Pediatr*. 2014.
- [3] Kidd EA, Fejerskov O. Essentials of dental caries. Oxford University Press; 2016.
- [4] Kassebaum NJ, Bernabé E, Dahiya M, Bhandari B, Murray CJ, Marcenes W. Global burden of untreated caries: a systematic review and metaregression. *J Dent Res*. 2015 ;94(5):650-8.
- [5] Frencken JE, Sharma P, Stenhouse L, Green D, Laverty D, Dietrich T. Global epidemiology of dental caries and severe periodontitis—a comprehensive review. *J Clin Periodontol*. 2017 ;44:S94-105.
- [6] Flayyih AS, Hassani HH, Wali MH. Identification of Strepto-coccus mutans from human dental plaque and dental caries using 16srrna gene. *Iraqi J Sci*. 2016;57(1C):552-7.
- [7] Ritter AV. Sturdevant's art & science of operative dentistry-e-book. Elsevier Health Sciences; 2017.
- [8] Khurshid Z, Naseem M, Sheikh Z, Najeeb S, Shahab S, Zafar MS. Oral antimicrobial peptides: Types and role in the oral cavity. *Saudi Pharm J* . 2016;24(5):515-24.
- [9] Khurshid Z, Najeeb S, Mali M, Moin SF, Raza SQ, Zohaib S, Sefat F, Zafar MS. Histatin peptides: Pharmacological functions and their applications in dentistry. *Saudi Pharm J*. 2017 ;25(1):25-31.
- [10] Li X, Duan D, Yang J, Wang P, Han B, Zhao L, Jepsen S, Dommisch H, Winter J, Xu Y. The expression of human  $\beta$ -defensins (hBD-1, hBD-2, hBD-3, hBD-4) in gingival epithelia. *Archives of Oral Biology*. 2016;66:15-21.
- [11] Lee SH, Baek DH. Antibacterial and neutralizing effect of human  $\beta$ -defensins on *Enterococcus faecalis* and *Enterococcus faecalis* lipoteichoic acid. *J Endod*. 2012;38(3):351-6.
- [12] Geevarghese A, Baskaradoss JK, Sarma PS. Oral health-related quality of life and periodontal status of pregnant women. *Matern Child Health J*. 2017;21(8):1634-42.
- [13] Wu M, Chen SW, Su WL, Zhu HY, Ouyang SY, Cao YT, Jiang SY. Sex hormones enhance gingival inflammation without affecting IL-1 $\beta$  and TNF- $\alpha$  in periodontally healthy women during pregnancy. *Mediators Inflamm*. 2016.
- [14] Silk H, Douglass AB, Douglass JM, Silk L. Oral health during pregnancy. *Am Fam Physician*. 2008;77(8):1139-44.
- [15] Martínez-Beneyto Y, Vera-Delgado MV, Pérez L, Maurandi A. Self-reported oral health and hygiene habits, dental decay, and periodontal condition among pregnant European women. *Int J Gynaecol Obstet*. 2011;114(1):18-22.
- [16] Navazesh M, Kumar SK. Xerostomia: prevalence, diagnosis, and management. *Compend Contin Educ Dent* . 2009: 8-326 (6) 30.
- [17] Greene JG, Vermillion JR. The simplified oral hygiene index. *J Am Dent Assoc*. 1964;68(1):7-13.
- [18] World Health Organization. Oral health surveys: basic methods. World Health Organization. 2013.
- [19] Gupta R, Acharya AK. Oral health status and treatment needs among pregnant women of Raichur District, India: A population based cross-sectional study. *Scientifica*. 2016.
- [20] Sonbul H, Ashi H, Aljahdali E, Campus G, Lingström P. The influence of pregnancy on sweet taste perception and plaque acidogenicity. *Matern Child Health J*. 2017;21(5):1037-46.
- [21] Yousefi M, Parvaie P, Riahi SM. Salivary factors related to caries in pregnancy: a systematic review and meta-analysis. *J Am Dent Assoc*. 2020;151(8):576-88.
- [22] Srivastava VK. Modern pediatric dentistry. 1st ed. Jaypee Brothers Medical Publishers Ltd.; 2011.
- [23] Issa ZM, El-Samarrai SK. Oral health status among a group of pregnancy and lactating women in relation to salivary constituents and physical properties (A comparative study). *J Baghdad Coll Dent*. 2012;24(2):155.
- [24] Africa CW, Turton M. Oral health status and treatment needs of pregnant women attending antenatal clinics in KwaZulu-Natal, South Africa. *Int Dent*. 2019.
- [25] Al Zaidi W. Oral immune proteins and salivary constituents to oral health status among pregnant women (Doctoral dissertation, PHD. Thesis submitted to the College of Dentistry, University of Baghdad). Department of Preventive Dentistry .2007.
- [26] Mutlak NQ, Yas BA. Dental caries severity in relation to selected salivary variables among a group of pregnant women in Baghdad city/Iraq. *J Baghdad Coll Dent*. 2017;29(2):115-21.
- [27] Amin R, Shetty P. Oral health status during pregnancy in Mangalore. *J health Allied Sci Nu*. 2014;4(02):114-7.
- [28] Al Najjar SN, Hussein B. Oxidative status among a group of pregnant women in relation to gingival health condition. *J Baghdad Coll Dent*. 2019;31(4).
- [29] Al-Zaidi WH, AbdulAziz MA. Salivary sex hormones & its relation to periodontal status among pregnant & non pregnant women. *Mustansiria Dent J*. 2011;8(3):343-8.
- [30] Hassan IS, Laylani LA. Oxidative stress state during pregnancy period. *Iraqi J Med Sc*. 2017;58(2C):984-7.
- [31] Tedjosasonko U, Anggraeni F, Wen ML, Kuntari S, Puteri MM. Prevalence of caries and periodontal disease

- among In-donesian pregnant women. *Pesqui Bras Odontopediatria Clin Integr.* 2019;19: 4533.
- [32] Uadav K, Prakash S. Dental Caries: A Microbiological Ap-proach. *J Clin Infect Dis Pract.* 2017;2(1):1-5.
- [33] Lai Y, Gallo RL. AMPed up immunity: how antimicrobial peptides have multiple roles in immune defense. *Trends in immunology.* 2009;30(3):131-41.
- [34] Gürsoy M, Gürsoy UK, Liukkonen A, Kauko T, Penkkala S, Könönen E. Salivary antimicrobial defensins in pregnancy. *J Clin Periodontol.* 2016;43(10):807-15.
- [35] Phattarataratip E, Olson B, Broffitt B, Qian F, Brogden KA, Drake DR, Levy SM, Banas JA. *Streptococcus mutans* strains recovered from caries- active or caries- free individuals differ in sensitivity to host antimicrobial peptides. *Mol. Oral Microbiol.* 2011;26(3):187-99.
- [36] Dale BA, Tao R, Kimball JR, Jurevic RJ. Oral antimicrobial peptides and biological control of caries. *BMC oral health.* 2006;6(1):1-7.
- [37] Barrera J, Tortolero S, Rivas A, Flores C, Gonzales E. Increased expression and levels of human  $\beta$  defensins (hBD2 and hBD4) in adults with dental caries. *J. Health Sci.* 2013;3(2):88-97.
- [38] Al-Ali GM, Jafar ZJ, AL-Ghurabi BH. The relation of salivary cathelicidin and beta-defensin with dental caries of school-children. *J Res Med Dent Sci.* 2021;9(4):30-5.
- [39] Gomes PD, Fernandes MH. Defensins in the oral cavity: dis-tribution and biological role. *J Oral Pathol Med.* 2010;39(1):1-9.
- [40] Ribeiro AE, Lourenço AG, Motta AC. Influence of periodontal condition on levels of human beta defensins 1 and 2 in saliva. *J Microbiol Exp.* 2018;6(2):00186.

**To cite this article:** Hussein SA, Alwaheb A. The Oral Health Status in relation to Salivary Antimicrobial Peptide in Pregnant Women . *Al-Kindy College Medical Journal.* 2023;19(1):81–5.