



International Journal of Current Research Vol. 10, Issue, 06, pp.70174-70178, June, 2018

## **RESEARCH ARTICLE**

# ASSESSMENT OF SALIVARY pH, URIC ACID AND PERIODONTAL STATUS IN VEGETARIAN AND NON-VEGETARIAN SUBJECTS

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#### **ARTICLE INFO**

#### Article History:

Received 10<sup>th</sup> March, 2018 Received in revised form 12<sup>th</sup> April, 2018 Accepted 20<sup>th</sup> May, 2018 Published online 28<sup>th</sup> June, 2018

#### Key words:

Vegetarian, Non Vegetarian, Antioxidant, Saliva.

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#### **ABSTRACT**

Aim: This study was conducted to estimate and compare the salivary pH, uric acid and periodontal status in vegetarians and non vegetarians. Material and Methods: The study consisted of 90 systemically healthy subjects in the age group of 18 years or above which was further divided into two groups: vegetarian and non vegetarian group. Plaque index, gingival index, sulcus bleeding index, probing pocket depth, clinical attachment level were recorded. Estimation of salivary pH was done using pH meter and salivary uric acid was estimated using Uric acid Kit in a semi automatic biochemical analyser. Statistical analysis was performed using independent t test with a level of significance 5% and power 80% and median effect size of 0.6. Results: A total of 90 patients were recruited for the study (21 females and 24 males in the vegetarian group and 25 females and 20 males in the non vegetarian group) The results revealed that comparison of uric acid between the non vegetarian and vegetarian group shows a statistical significant difference (p-value=0.003). There was significant difference between the groups on comparing gingival index and sulcus bleeding index with a p value of 0.041 and 0.045. Conclusion: This study has shown that vegetarian subjects had a statistically significant higher uric acid level (antioxidant) and reduced inflammatory signs.

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Citation: Dr. Rajesh K.S., Dr. Vino Tito V Kurien, Dr. Shashikanth Hegde and Dr. Arun Kumar, M.S. 2018. "Assessment of salivary ph, uric acid and periodontal status in vegetarian and non-vegetarian subjects", *International Journal of Current Research*, 10, (06), 70174-70178.

## INTRODUCTION

Gingivits and periodontitis are chronic inflammatory diseases causing destruction of the tooth supporting structures due to combination of many factors. The causative agent associated is bacterial infection. The bacterial biofilm consist of diverse group of communities which causes oral diseases (gingivitis and periodontitis) which results in tooth loss (Pihlstrom et al., 2005). Periodontitis is one of the most health debilitating conditions in the world (Petersen et al., 2005). Both gingivitis and periodontitis cause inflammatory condition in oral cavity and also it relates to the systemic health (Tonetti et al., 2007). There is an increase in evidence that periodontitis and systemic diseases associate with each other (Kinane, 2001). The role of systemic diseases in initiating periodontal diseases is clearly mutual (Kinane, 2008). Periodontal health is also influenced by a numerous other factors like oral hygiene, genetic factors, epigenetic factors and nutrition (Najeeb et al., 2016). The relationship between periodontal disease and nutritional factors is not clear. Nutrition is a process of providing food for fitness, growth, metabolism and repair.

Nutritional aspects are dissimilar from dietary. A diet is special course of food that a person consumes for health. Nutrition refers to sum total of biologic process that relates to dietary intake (Hujoel, 2017). The role of diet in the course of oral disease is less clearly accepted (Schifferle, 2005). Recent studies have reported that people tend to lose their teeth due to poor nutritional diet (Marshall, 2002; Hung, 2003). A person's diet can display different oral and systemic illness in their body and its tissues (Hujoel, 2017). Food occupies the supreme position in the hierarchical needs of man (Swami Nathan, 1990). The source of food is either from plant or animal origin. Vegetarianism is the implementation of having only plant based foods and restraining from all meat products (Tiegen, 1981). Lacto vegetarians consume plant food along with dairy products and lacto ovo vegetarians add eggs to their diet (Herman et al., 2011). Vegetarians also consume large quantity of antioxidants mainly from fruits and vegetables (Linkosalo, 1988). Several studies also revealed that less alcohol and tobacco consumption and they are more physically active and have lower body mass index compared to non-vegetarians (Larsson et al., 2002; Davey et al., 2003).

It has been shown that in all phases of life a vegan diet is nutritionally sufficient and provides healthier lifestyle in the protection of many diseases (Key *et al.*, 1999). Therefore aim of the present study was to estimate and compare uric acid, salivary pH of unstimulated saliva and the periodontal health status in vegetarians and non vegetarians.

## **MATERIALS AND METHODS**

This study was conducted in Department of Periodontology Yenepoya University after the due approval from the institutional Ethical committee. All the participants were provided with the verbal explanation of the nature of the study, and informed consent was obtained. The sample size was calculated at 80% of power and 5% significance level. The study population consisted of 90 systemically healthy subjects above the age group of 18 years that was further categorized into two groups comprising 45 in each group.

• Group I: Vegetarian subjects.

(Lacto-vegetarians: only consume dairy products in addition to vegetables)

• Group II: Non vegetarian subjects. (Mixed diet)

[Consume all types of animal-based food products, including red meat, poultry and seafood. They also consume eggs, dairy products and plant-based food in their meals]

Subjects with any systemic diseases such as diabetes, hepatic disease, renal disease and malignant diseases and those with any history of long term medication for the past six months, smokers, pregnant, lactating women, subjects who had received any periodontal treatment during the past 6 months, subjects with <20 natural teeth, were excluded from the study. A complete medical and dental history was obtained. Periodontal examination included gingival index, sulcus bleeding index, probing pocket depth, and clinical attachment loss.

Saliva collection and estimation of pH and uric acid: Human whole unstimulated was collected by spitting method without swallowing, with the subject seated in an upright position after refraining from food intake 2 hours before saliva collection. Approximately, 5 mL of saliva was collected and stored in to graduated saliva collecting vials and refrigerated at 4°C until it is further processed. The samples were centrifuged at 2800 rpm for 10 min and the supernatant was separated from the substrate and stored at -20°C. Next the sample was defrosted at room temperature. The supernatant was analysed for uric acid analysis. Salivary pH was estimated electrometrically with the help of a pH meter. A pair of electrodes were dipped in saliva, whereby an electric potential was developed across the thin glass of the bulb (of glass electrode). Variations of pH with electromotive force (E) was recorded directly on the potentiometer scale graduated to read pH directly. Uric acid concentrations were estimated using Uric acid Kit in a semi automatic biochemical analyzer.

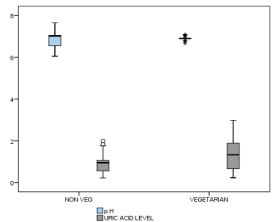
**Statistical Analysis:** Data is expressed in median and interquartile range (IQR).Mann-Whitney U test is used to compare the salivary parameters and periodontal status

between the groups.p<0.05 is considered significant. Statistical analysis was done using SPSS Software version 22.

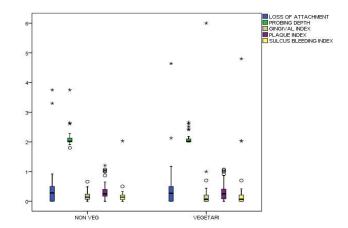
#### RESULTS

A total of 90 patients were recruited for the study (21 females and 24 males in the vegetarian group and 25 females and 20 males in the non vegetarian group). Table number 1 showing the comparison of salivary parameters between the groups. The median value of pH of non vegetarian was (7.01) and vegetarian group (6.90). The median values of uric acid in non vegetarian group is 0.95 and vegetarian group is 1.33. Comparison of uric acid between the non vegetarian and vegetarian group shows a statistical significant difference (p-value=0.003). There was significant difference between the groups on comparing gingival index and sulcus bleeding index with a p value of 0.041 and 0.045.

Box plot showing comparison in pH and uric acid level in vegetarian and non vegetarian.



It was observed that there is a significant difference in uric acid level between the groups (p value =0.003).



Box plot showing comparison in clinical parameters between the groups. It was observed there is significant difference between the gingival index and sulcus bleeding index between the groups (p value=0.041,0.045).

### **DISCUSSION**

Vegetarian diet is diverse in composition and it refers to absence of meat. Different variants of vegetarian diet comprise of lacto-vegetarians (includes dairy products to vegan diet) and lacto-ovo-vegetarians (adds egg and dairy products to their diet).

Table 1. Showing the comparison of salivary parameters between the groups

	Non-Vegetarian			Vegetarian			Mann-Whitney Test	
	Median	IQR		Median	IQR		Z statistic	p-value
		Q1	Q3		Q1	Q3		
P.H	7.01	6.54	7.06	6.90	6.90	6.90	-0.801	0.423
URIC ACID LEVEL	.95	.56	1.07	1.33	.66	1.90	-2.959	0.003*

<sup>\*</sup>indicates statistical significance between the groups

Table 2. Showing comparison of clinical parameters between the groups

Clinical Parameters	Non-Vegetarian		Vegetarian			Mann-Whitney Test		
	Median	ΙÇ	)R	Median	ΙÇ	)R	Z statistic	p-value
		Q1	Q3		Q1	Q3		
LOSS OF ATTACHMENT	.28	.00	.50	.26	.00	.52	-0.435	0.664
PROBING DEPTH	2.02	2.00	2.14	2.01	2.00	2.11	-0.004	0.997
GINGIVAL INDEX	.14	.07	.25	.07	.00	.22	-2.04	$0.041^{*}$
PLAQUE INDEX	.25	.16	.41	.25	.08	.44	-1.188	0.235
SULCUS BLEEDING INDEX	.14	.06	.22	.07	.00	.22	-1.563	$0.045^{*}$

<sup>\*</sup>indicates significant difference between the groups

Table Number 3. The results of the questionnaire is given

Questions	Vegetarian group	Non vegetarian group		
1) Number of meals per day?				
1 3	Thrice (44.4%)	Thrice (40.0%)		
	More than thrice (55.6)	More than thrice (60.0%)		
2) Do you take fizzy drinks?	Yes (37.8%)	Yes (68.9%)		
	No (62.2%)	No (31.1%)		
3) Frequency of brushing?	Once (11.1%)	Once (51.1%)		
-	Twice (75.5%)	Twice (48.9%)		
	Thrice (13.3%)	Thrice (0%)		
	More than thrice (0%)	More than thrice (0%)		
4) Type of brush used?	Soft (20.0%)	Soft (24.4%)		
•	Medium (80.0%)	Medium (73.3%)		
	Hard (0%)	Hard (0%) Not applicable (1.1		
5) Type of brushing?	Horizontal (64.4%)	Horizontal (57.7%)		
	Rotatory (0%)	Rotatory (0%)		
	Vertical (22.2%)	Vertical (6.7)		
	Scrub (13.3%)	Scrub (35.6%)		
6) Do you use any other oral hygiene aids other	Yes (6.4%)	Yes (4.0%)		
than tooth brush?	No (93.6%)	No (96.6%)		
7) Frequency of dental visit per year	Once (77.8%)	Once (71.1%)		
	Twice or more (22.2%)	Twice or more (28.9%)		

Various authors came to the conclusion that systemic diseases like coronary heart disease, type 2 diabetes mellitus, and cancer are comparatively less in vegetarian population (McEvoy, 2012). Research on the effect of nutrition status and its role on saliva cover a wide range of factors (Lingström, 2003). Saliva is a combination of proteins, enzymes along with some other molecules including electrolytes, cells of epithelium, cells of immune system and microorganisms and their products. Saliva consists of various enzymes, molecules and therefore it acts as important antioxidant system. 70% of the total antioxidant system in saliva is represented by uric acid (Akgul *et al.*, 2013).

The ease of sampling saliva has made it a useful source of various biomarkers. Its biochemical composition can reflect the relation between oral health and various systemic diseases (Amirmozafari *et al.*, 2013). Salivary pH also changes dramatically in mixed diet people. It is usually acidic in nature. For vegetarian subjects pH is usually neutral (Nihlani *et al.*, 2011). The normal pH of a healthy subject ranges from 6.2-7.6. pH is maintained between 6.7 and 7.3 in the oral cavity and its resting pH is 6.3 (Baliga *et al.*, 2013). Only few studies are available in the literature investigating periodontal status in vegetarian subjects. So this study aimed to investigate the effects of salivary pH and uric acid level and the clinical parameters in two groups was compared with a appropriate sample size (n=90).

In this study uric acid was analysed because it is the predominant antioxidant found in saliva (Akgul et al., 2015). The salivary pH was significantly lower in vegetarian group. 66.7% of the subjects in vegetarians group had pH less than <7 than in non vegetarians (33.9%). Similar results in favour of pH reported in a study by Linkosalo et al and Laffranchi et al. where they compared periodontal health and salivary condition in lacto vegetarians (Linkosalo et al., 1985; Laffranchi et al., 2010). According to Bergenholtz et al. fruits and vegetables have been regarded as major foods affecting salivary flow and pH (Bergenholtz et al., 1967). Slightly higher pH in vegetarians was reported in a study by Johansson et al. without reaching a statistical significance (Johansson, 1995). According to Linkosalo et al the duration of the diet influenced the pH and buffer capacity in vegetarians. Fruits and vegetables affected the salivary flow and pH because stimulate large amount of alkaline saliva in the oral cavity and exerts a cleansing action on tooth surfaces (Linkosalo, 1985). A Finnish study reported that 76% of lactovegetarians had dental erosion and salivary pH was lower in vegetarians. On the contrary Laffranchi et al reported a pH between 5 and 6 in vegetarian population and observed visible precarious lesions (Laffranchi et al., 2010). In our study we observed 66.7% of vegetarians showed pH of less than 7 and 33.9% of non vegetarians showed pH of less than 7. Uric acid appears to be the predominant antioxidant present in saliva and displays a concentration homogenous to that of serum (Sculley, 2002).

So in our study we analysed the uric acid level between the two groups. Our data indicate a higher uric acid level among the vegetarians. Amirmozafari et al. compared the antibacterial and antioxidant activity of saliva in vegetarian and non vegetarians and found that the there is a significant reduction in protecting ability of saliva in vegetarians (Amirmozafari et al., 2013). The periodontal status in our study showed less bleeding on probing and inflammatory signs in vegetarian group. Linkosalo et al. revealed a lower bleeding on probing with vegetarians compared scores in the group 14 Vegetarian show more physical activity and they consume higher amount of antioxidants which improves their immune response and lead to less inflammatory signs (Tonstad et al., 2009). Other parameters such as probing depth, loss of attachment and plaque scores was not statistically significant in our study. Staufenbiel et al. (2013) and Rahamatulla et al. (1990) reported a lesser periodontal damage in vegetarian group. The possible reasons for less periodontal damage was due to lower body mass index, higher consumption of antioxidants and better hygiene index (Staufenbiel et al., 2013). Whereas no statistical significance difference were found in Sedgley et al between the groups (Sedgley et al., 1996). The questionnaire revealed higher frequency of twice oral hygiene in vegetarians (75.5%) than non vegetarians (48.9%) which might have contributed to less plaque scores and inflammatory signs in vegetarian group. Similar results in favour of oral hygiene were also reported by Staufenbiel et al. (1990). The amount of consumption of fizzy drinks were higher in non vegetarian group (68.9%). Dlaigan et al.33 and Herman et al. (2011) reported a relationship between acidic food and drink consumption and tooth erosion. The higher percentage of scrub technique of tooth brushing performed by non vegetarians (35.6%) than vegetarian group (13.3%), could have contributed for more periodontal damage in non vegetarians.

#### Conclusion

In conclusion, one of the famous quotes by Hippocrates "Let food be thy medicine, and medicine be thy food", which highlight the importance of our daily food choices and nutrition plays a prominent role for our underlying health. Based on the present evidence all dietry practices should aim to meet current nutritional guidelines to reduce chronic disease development.<sup>34</sup> A vegetarian diet has some favourable effect on periodontal health. It has been shown that at all periods of human life vegetarian diet is nutritionally adequate and provides a healthier life style protecting from various diseases. This study has shown that vegetarian subjects had a statistically significant higher uric acid level (antioxidant) and reduced inflammatory signs. Diet can affect several salivary variables. Therefore more controlled studies with definite information on lifestyle and dietary habits are paramount to clarify the role of diet on periodontal health.

## Conflict of interest statement: None

Funding Statement: None

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