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## **ORIGINAL ARTICLE**

# A COMPARATIVE EVALUATION OF SALIVARY FLOW RATE, pH, BUFFERING CAPACITY, CALCIUM AND TOTAL PROTEIN LEVELS IN PREGNANT AND NON PREGNANT WOMEN

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#### ABSTRACT:

**Background:** Hormonal changes in females may affect physiology of the entire body including the oral cavity. The present study aimed to evaluate salivary flow rate, ph, buffering capacity, calcium and total protein levels in pregnant and non pregnant women. **Materials and Methods:** A cross-sectional study was designed and thirty pregnant and 30 non pregnant women women aged between 19-34 years in the third trimester were included in the study. The salivary samples were collected between 9- 11.30 a.m in both the study and control group. The salivary flow, pH, and buffering capacity was measured using "Saliva-check buffer kit. **Results:** Salivary flow rate, pH has decreased in pregnancy than non pregnant women. DMFT index showed a strong correlation with pH in pregnant women (P = 0.002) and non-pregnant women (P = 0.01). Calcium and protein level decreases in pregnant women in comparison to non pregnant women but difference was statistically non significant. **Conclusion:** Even though the salivary parameters were observed in normal range but there were definitive alterations.

**Key words:** Buffering capacity, Calcium, pH, Pregancy.

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### NTRODUCTION:

Saliva is a biologic and diagnostic fluid in the oral cavity, composed of a mixture of secretary products from the major and minor salivary glands. Saliva plays key roles in lubrication, mastication, taste perception, prevention of oral infection and dental caries.

Normal salivary gland function is essential for the maintenance of oral health. Oral health is affected by

many systemic conditions. A decrease in the salivary secretion rate promotes caries and gingival inflammation.<sup>3</sup>

Changes in salivary composition and flow rate may compromise the integrity of the soft tissues in oral cavity. Hormonal changes in females may affect physiology of the entire body including the oral cavity. Besides the direct effect on metabolism of periodontal tissue, pregnancy may induce short term changes in flow rate, buffering capacity and biological composition of slaiva.<sup>4</sup>

Pregnancy is accompanied by profound physiologic changes due to complex hormonal interactions which may lead to an increase in the incidence of dental caries,<sup>5</sup>

Female sex hormones (estrogen, progesterone & human gonadotropin) are secreted primarily by the placenta. These hormones are responsible for most of the physiologic changes during pregnancy. The main salivary changes in pregnancy involve its flow, composition, pH and hormone levels.

Pregnancy increases the propensity to gingival inflammation known as pregnancy gingivitis, with an enhanced gingival bleeding tendency without specific plaque association; periodontal pocket formation and dental caries can increase during pregnancy. These changes are reversible after delivery and the exact etiology for this is still unclear.<sup>6</sup>

Many studies discussed about salivary flow rate, pH, buffer capacity and total protein in relation to dental caries, but there are differences in obtained results between the studies in different regions.

The present study is undertaken to evaluate salivary flow rate, ph, buffering capacity, calcium and total protein levels in pregnant and non pregnant women.

#### MATERIALS AND METHODS:

Ethical clearance was taken from Institutions Ethical Committee before the commencement of study. A written consent from all the subjects were taken who were involved in the study.

A cross-sectional study was conducted among thirty pregnant and 30 non pregnant women women aged between 19-34 years in the third trimester who visited the clinics of Oral Medicine and Radiology Department at Triveni Institute of Dental Sciences, Hospital and Research Centre, Bilaspur, Chhatisgarh. None of the healthy, non-pregnant women were taking oral contraceptive pills.

Subjects with salivary gland disorders and with any systemic illness were excluded from study group.

A questionnaire was structured to acquire data on oral hygiene habits (tooth brushing frequency and method, use of interproximal brushes, dental floss, tongue cleaning habit). Information regarding pregnancy trimester and use of medication was procured from the subject's medical charts. The intraoral examination was conducted by a single examiner under favorable lighting conditions using a sterile mouth mirror, diagnostic probe, and explorer. The

clinical findings were recorded in the study proforma. The Decayed-Missing-Filled Teeth (DMFT) index, an irreversible index, was obtained to determine the prevalence of dental caries.

The saliva sample collection procedure was standardized prior to the study. The salivary samples were collected between 9- 11.30 a.m in both the study and control group. The salivary flow, pH, and buffering capacity was measured using "Saliva-check BUFFER kit" (In Vitro test for pH and Saliva Buffering Capacity) manufactured by GC Corporation. The kit is provided with a pH strips which measures the pH between 5-8, saliva collection cups, paraffin wax for saliva stimulation, saliva dispensing pipette and buffer test strips.

The participants were refrained from smoking, eating and drinking for a minimum of 1 h prior to saliva collection. The subjects were asked to sit comfortably with head tilted slightly forward and expectorate the saliva accumulated in the floor of the mouth into disposable plastic containers for duration of 5 min. The resting salivary flow rate is measured as ml/min. The stimulated salivary flow was assessed by asking the patient to chew a piece of paraffin wax. After 30 second, the patient was asked to expectorate into the spittoon. The patient was instructed to continue chewing the wax for 5 minutes and the saliva was collected in a collection cup with ml marking. The pH of unstimulated saliva was determined by using a pH strip provided in the kit and placing it in the collected sample of resting saliva for 10 seconds. The color change of the strip was compared with the testing chart available with the kit and recorded.

The buffering capacity of the unstimulated saliva was measured by using a buffer strip provided in the kit. Using pipette sufficient saliva from the collection cup was dispensed on to the test pad. At the end of 2 min the test pad would change its color, comparing the change in color with the chart provided in the kit the buffering capacity was scored and recorded.

The protein content of saliva samples was measured using Bradford method (Nicholas, 2009). Saliva total calcium concentration was measured using spectrophotometric method (Pars Azmoon Kit, Iran). Statistical analysis was done using SPSS software version 15.0 using Student's *t*-test. The correlations between DMFT index and saliva parameters were measured using Pearson's correlation coefficients. P value <0.05 was considered statistically significant.

#### **RESULTS:**

Salivary flow rate has decreased in pregnancy (0.62) than non pregnant women having values of 0.86. (Table 1)There was a reduction in the pH and buffering capacity in the study group with a mean pH and buffering capacity of 6.34 and 7.80 respectively. The control group had a mean pH of 6.85 and the buffering capacity of 9.86. (Table 1)

DMFT index showed a strong correlation with pH in pregnant women (P = 0.002) and non-pregnant women (P = 0.01). The co-relation coefficients were negative for pregnant and non pregnant women groups, indicating that caries incidence increases with decrease in pH and buffering capacity of saliva. (Table 2)

Calcium and protein level decreases in pregnant women in comparison to non pregnant women but difference was statistically non significant. (Table 1)

#### **DISCUSSION:**

About 600ml of saliva containing minerals, electrolytes, buffers, enzymes, growth factors enzyme inhibitors and immunoglobulin's, cytokines, mucin and other glycoproteins is produced by the salivary gland every day. Saliva possesses antimicrobial

components and buffering agents that act to maintain oral tissue. Many studies have shown that saliva has intricate relationship with the serum parameters, hence it can be used in detecting physiological and pathological changes in the body. Hence, it plays a major role in maintenance of oral health. <sup>8</sup>

Present study revealed that the salivary flow rate and pH of saliva decreased in pregnant women than in non-pregnant women. Similar results has also been obtained by Rockenbach M, et al<sup>9</sup> who did a cross sectional study in 2006 to compare the salivary flow rate, pH on 22 pregnant and non-pregnant women in Obstertics and Gyneocology clinics in Sao Lucas Hospital in Brazil. The study concluded that the pregnant women had lower pH than the non-pregnant women but the salivary flow rate was not so satisfactorily different. The salivary composition and flow rate are altered during pregnancy, menstruation, and menopause due to changes in steroid hormone levels. These alterations may be due to the hormonal changes that take place during pregnancy. Saliva composition and secretion is modulated by many hormones but the exact mechanism how these hormones bring about these changes is poorly understood.

**Table 1:** Salivary flow rate, pH, DMFT index, buffering capacity, calcium and protein level in pregnant and non pregnant women

Variables	Pregnant women	Non pregnant	t value	P value			
		women					
Salivary flow rate	$0.62 \pm 0.24$	0.86±0.28	3.42	0.001 H.S.			
pН	6.34±0.38	$6.85 \pm 0.25$	5.565	0.000 H.S.			
DMFT index	7.97±3.42	8.23±4.46	2.86	0.68 N.S			
Buffering	7.80±1.18	9.86±1.56	5.886	0.000 H.S.			
capacity							
Calcium	$0.876 \pm 0.09$	1.020±0.06	1.14	0.074 N.S			
(mmol/L)							
Protein	7.74±1.6	9.94±3.6	3.44	0.062 N.S			
(mg/ml)							

Table 2: Correlation between salivary flow rate and pH of saliva for pregnant and non-pregnant groups

Pearson	Salivary flow rate		pН		DMFT Index	
co-	Pregnant	Non	Pregnant	Non	Pregnant	Non Pregnant
relation	women	Pregnant	women	Pregnant	women	women
		women		women		
p	0.26	0.05	0.002	0.02	0.002	0.01
r	-0.24	0.28	-0.56	-0.44	-0.36	-0.24

It has been hypothesized that increased estrogen during pregnancy increases blood flow of salivary glands which may be associated with variation in salivary secretions.<sup>10</sup>

Studies conducted by Hugoson<sup>11</sup> and González et al<sup>12</sup> also showed similar findings of decreased salivary flow rate in pregnant women. Salivary flow rate has strong influence on the pH of saliva. At higher flow rate, there is an increased level of salivary pH because the bicarbonate concentration increases with elevated flow rate. In present study, the salivary pH levels for both the groups were above the critical pH of 5.5. Critical pH is the point where the hydroxyapatite crystals begin to dissolve causing demineralization of teeth resulting in cavitation or dental caries.

Laine et al<sup>13</sup> reported that the salivary pH decreased toward late pregnancy, followed by a rapid increase after childbirth. The most important protein of saliva is  $\alpha$ -amylase which is secreted by parotid gland. Increasing trend of this enzyme activity may lead to increased microorganism substitution and reduced pH of saliva. It was found that  $\alpha$ - amylase activity increase during 10 and 21 weeks of gestation

Eliasson et al<sup>10</sup> suggested that the inorganic and protein composition of saliva changes during the course of pregnancy but our study revealed that Calcium and protein level decreases in pregnant women in comparison to non pregnant women but difference was statistically non significant. Similar results have also been obtained by Olatunbosun et al<sup>14</sup> who evaluated Serum calcium, phosphorus and magnesium levels in pregnant and non-pregnant Nigerians. Orosz et al. suggested that a positive correlation exists between pregnancy and caries. But in contrary to these results our study revealed no any significant correlation between them. Similar results have also been obtained by Russell et al who suggested that no linear relationship exists between salivary secretion rate, DMFT/DMFS index, and caries activity. Mandel and Russell et alreported that only weak or no association was found between salivary secretion rate and caries incidence.<sup>15</sup>

#### **CONCLUSION:**

Saliva is one of the most important factors in prevention of dental caries. Salivary components are altered during pregnancy. Further studies are required to assess salivary parameters in medically compromised women with high-risk pregnancy with larger sample size.

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