



## Original Article

# Oral Findings, Salivary Bicarbonate, and Phosphate Levels in Patients with Gastroesophageal Reflux Disease

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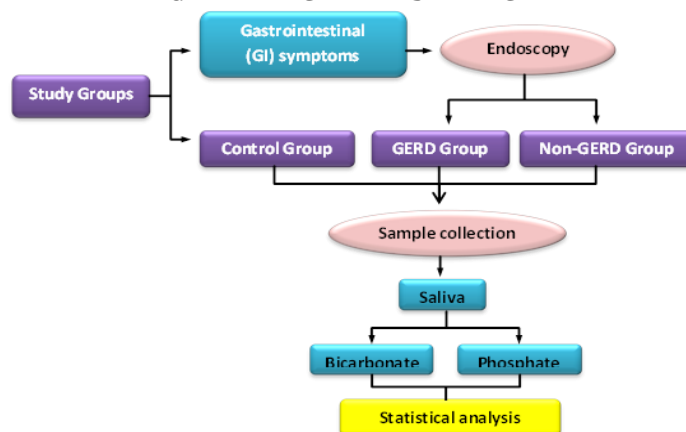
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## ABSTRACT

The neutralization of stomach's acidic contents and protection of esophageal mucous membrane during reflux episodes is the primary function of the major buffering components of saliva, bicarbonate, and phosphate. The concentrations of inorganic compounds in the saliva of subjects who have Gastroesophageal reflux disease versus healthy individuals are poorly understood. In this work, the aim of study was to assess the occurrence of soft tissues alterations and symptoms in oral cavity and analysis of bicarbonate and phosphate to evaluate their levels in saliva of patients with GERD and compare with control individuals. A case-control study included 25 GERD patients diagnosed both clinically and endoscopically (GERD Group), 25 patients with upper gastrointestinal symptoms with no endoscopic signs of GERD (non GERD group) and 25 healthy subjects (control group) have been evaluated through visual examination of the oral mucosa, oral symptoms, non-stimulated salivary flow rate, swallowing function, in addition to the estimation of salivary concentration of bicarbonate and phosphate. Study result was significantly higher salivary bicarbonate in GERD patients ( $p < 0.05$ ), significantly lower Salivary flow rate and swallowing frequency compared to non-GERD group and control group. Among the patients with GERD, 56% of the cases suffering from oral dryness, 40% halitosis, 28% acid taste, 20% burning sensation, 20% palatal erythema. Salivary bicarbonate, oral symptom and mucosal alteration were shown to be significantly related to GERD. Therefore, GERD should be taken into account by dentists as a possible risk factor for oral health

## GRAPHICAL ABSTRACT



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## Introduction

The Montreal Consensus defined gastroesophageal reflux disease (GERD) as a chronic condition that result from passage of stomach contents into the esophagus and toward the oral cavity, and lungs to manifests as symptoms and/or complications that interfere with daily activities [1]. GERD remains a common disease around the world and the estimates of GERD prevalence vary greatly, the variety of prevalence percentages that found between studies may be due to distinct geographic variations, variations in study's design, and measuring methods [2]. Obesity, tobacco smoking, meals with high level of fat, and specific types of food such as chocolate and spicy foods are all thought to be risk factors that contribute to the onset and exacerbation of GERD symptoms. Therefore, changes in lifestyle are regarded as the initial line of GERD therapy [3]. Within many symptoms of GERD heartburn and regurgitation consider as an esophageal symptoms that associated with reflux. Also, there are extraesophageal symptoms related to acid reflux such as chronic cough and dental erosion, while complications vary from esophagitis to Barrett's esophagus and esophageal adenocarcinoma [1]. Many studies documented oral symptoms of GERD that can be include dental caries, dry mouth, feeling of acid/burning sensation, halitosis, erythema of the oral mucosa especially palatal mucosa, and uvula [4]. Another study demonstrates microscopic changes in the palatal mucosa of GERD patients represented by mucosal atrophy and increase in fibroblast [5]. Acid clearance from the esophagus occurs mechanically via primary peristalsis of esophagus and chemically via swallowing saliva and neutralizes residual acid [6]. Thus, the integrity of esophageal epithelium depends upon balance between excitation factors (such as gastric acid and pepsin) and protective systems. Saliva's volume and neutralizing ability (via bicarbonate and phosphate mainly) act as pre-epithelial protective factor during esophageal exposure to gastric acid. The most important neutralizing component is bicarbonate; as bicarbonate concentration rises, salivary pH rises as well,

further buffering acidity [7-9]. The aim of this study was to evaluate oral findings in GERD patients include mucosal lesion, salivary flow rate, and swallowing function in those patients with assessment of most common complaint. Another aim of this study was to determine the level of bicarbonate and phosphate in saliva from GERD patients and compare concentrations with non-GERD patients and control individuals.

## Martials and Methods

During period from January 2022 to May 2022, this case-control study was carried out in the Department of Gastroenterology, Al-Ramadi Teaching Hospital, Al-Anbar, Iraq. Seventy-five individuals were enrolled in this study. They were attending the gastroenterology unit for various symptoms. The clinical and endoscopic examination was the basis for the diagnosis of GERD under supervision of physician and surgeon specialists. They were (38) males and (37) females; with an age range (18-50) years old, classified as 25 patients with upper gastrointestinal symptoms whom endoscopic evidences of GERD were confirmed (GERD group) and 25 patients with upper gastrointestinal symptoms who had no endoscopic signs of GERD (Non-GERD group). In addition to 25 healthy controls that have no symptoms or medical history of gastrointestinal system diseases, with no signs and symptoms of any other systemic diseases as control group. Exclusion criteria was patients with previous esophageal or gastric surgery history, any condition of the salivary glands known to affect salivation, pregnant women, individuals with history of underlying systemic diseases affecting esophageal motor activity, patients on antacids or PPIs therapy for 2 weeks before taking samples. The purpose of the study was explained to each subject. The ethical approval was obtained from the collage of Dentistry University of Baghdad and informed consent was obtained from all participants. Case sheet was contained the information about name, gender, age, medical history, family history, dietary history and some oral manifestation that noticed in GERD patients (oral dryness, acid taste, bitter taste, halitosis, and pharyngeal discomfort).

Patients were questioned about six GERD predictors using the GERD Q score [10] before having an oesophago-gastroduodenoscopy (OGD) procedure. All the patients have been examined by single examiner, under standardized conditions; the oral cavity has been examined by artificial light using a mouth mirror. The procedure of examination of oral tissue was done in sequence according to direction suggested by WHO (2013) [11]. In case of oral mucosal lesion; following scoring system was used: score 0 (no inflammation), 1(redness), and 2 (ulceration) [12].

Fasting un-stimulated whole saliva was obtained between (9-12) a.m. Patients were instructed to not brush their teeth before the collection and did not spit up any mucus during the saliva collection. The individuals were asked to expectorate saliva into graduated tube every one minute for a total of 5 minutes, and then measured to determine salivary flow rate (ml/minutes) [13]. The collected saliva was stored at -20 °C until laboratory analysis. Swallowing function was assessed by the repetitive saliva swallowing test (RSST) [14] which evaluates the potential to swallow saliva. It was performed by asking the patient to swallow their own saliva and by vision or palpating the hyoid, counts the number of completed swallow in 30 second as possible, and also record the time to onset of first swallow. RSST was carried out under the same circumstances for each individual. The Diagnostic cobas C311 analyzer from Roche was used to evaluate bicarbonate and phosphate level in saliva. It was automated, software controlled analyzer for clinical chemistry analysis. It is designed for both

quantitative and qualitative determination via perform photometric assays and ion selective electrode measurements.

Statistical analysis

Data analysis was carried out using Statistical Packages for Social Sciences-version 28 (SPSS-28). The significance of difference of different means (quantitative data) was tested using students-t-test for difference between two independent means or ANOVA test for difference among more than two independent means. Statistical significance was considered whenever p-value was equal or less than 0.05.

Results and Discussion

Clinical findings

This study showed that the mean age and standard deviation of GERD patients was 34.6±8.6 years old, non-GERD patients was 38.2±6.8 years old, and for control subjects was 32.9±8.1 years old. Statistical analysis tests showed no significant differences between the ages of study groups (Table 1).

The number of female GERD patients was14 (56.0%) and the number of male GERD patients was11 (44.0%). The percentage of female patients with GERD was higher than the percentage of male patients, with no significant difference between study groups, as listed in (Table 2).

Using point 8 as a cut-off value, in total GerdQ score the current study showed that five GERD patients (20%) had a total GerdQ score below 8 and 20 GERD patients (80%) had a total score ≥8. A significant difference between study groups was found in relation to GerdQ score (Table 3).

Table 1: The number of patients/subjects and mean age of study groups with their statistical analysis

		GERD		Non-GERD		Control		P-value
		NO	%	NO	%	NO	%	
Age (years old)	< 30 years old	7	28.0	4	16.0	13	40.0	0.080 NS
	30-39	12	48.0	7	28.0	12	32.0	
	40-49	6	24.0	14	56.0	7	28.0	
	Mean± SD (range)	34.7±8.6 (19-49)		38.2±6.8 (22-49)		32.9±8.1 (20-48)		0.059 NS

NS: Non-significant p > 0.05.

Table 2: The number and percentage of both genders of each group

Gender		GERD		Non-GERD		Control		P-value
		NO	%	NO	%	NO	%	
	Male	11	44.0	14	56.0	13	52.0	0.688 NS
	Female	14	56.0	11	44.0	12	48.0	
	Total	25	100	25	100	25	100	

NS: Non-significant  $p > 0.05$ .

Table 3: Total GerDQ scores for study groups with statistical analysis

		GERD		Non-GERD		Control		P-value
	Total score	No	%	No	%	No	%	
GERD questionnaire	0	-	-	-	-	25	100.0	0.0001*
	2	-	-	3	12.0	-	-	
	3	-	-	3	12.0	-	-	
	4	1	4.0	7	28.0	-	-	
	5	1	4.0	4	16.0	-	-	
	6	-	-	5	20.0	-	-	
	7	3	12.0	3	12.0	-	-	
	8	2	8.0	-	-	-	-	
	9	5	20.0	-	-	-	-	
	10	2	8.0	-	-	-	-	
	11	2	8.0	-	-	-	-	
	12	4	16.0	-	-	-	-	
	13	3	12.0	-	-	-	-	
	14	2	8.0	-	-	-	-	

\*Significant difference between percentages using Pearson Chi-square test ( $\chi^2$ -test) at 0.05 level

This study showed that the number of patients who feel Oral dryness was 56% of GERD group, 12% of non GERD group and there was no oral dryness in control subjects. Statistically, the number of subjects with oral dryness was significantly higher in GERD patients than in non-GERD and control subjects (Table 4). There was no acid taste felt in both non GERD patients and control subjects, while 28% of GERD patients were with acid taste feeling in oral cavity. This study also showed that the percentage of GERD patients who feel bitter taste was 36%, for non-GERD patients was 12%. The statistical analysis showed significant differences in acid taste and also in bitter taste between study groups (Table 4). This study showed that 40% of GERD patients felt bad breath (halitosis), while 20% of non-GRED patients were reported such feeling. There was statistical significant difference in bad breath

between study groups. Burning sensation reported in 20% of GERD patients and 4% of non-GERD patients. Statistical analysis showed significant differences between study groups (Table 4). The current study, 16% of GERD patients and 4% of non-GERD patients reported pharyngeal discomfort. Statistical analysis showed no significant differences between study groups (Table 4). The results of the study showed that 20% of GERD patients had erythematous area in oral buccal mucosa, 8% for non GERD patients. In palatal mucosa, 20% of GERD patients showed erythematous area. No oral ulceration found in all study groups. Statistical analysis showed significant differences between study groups for both buccal and palatal mucosal lesion ( $p < 0.05$ ), as provided in (Table 5).

**Table 4:** The percentage of patients/subjects that complain oral finding in each group with statistical analysis

Presence of following problems		GERD		Non-GERD		Control		P-value
		No	%	No	%	No	%	
Oral dryness	Yes	14	56.0	3	12.0	-	-	0.0001*
	No	11	44.0	22	88.0	25	100.0	
Acid taste	Yes	7	28.0	-	-	-	-	0.0001*
	No	18	72.0	25	100.0	25	100.0	
Bitter taste	Yes	9	36.0	3	12.0	-	-	0.002*
	No	16	64.0	22	88.0	25	100.0	
Bad breath (halitosis)	Yes	10	40.0	5	20.0	-	-	0.002*
	No	15	60.0	20	80.0	25	100.0	
Burning sensation	Yes	5	20.0	1	4.0	-	-	0.022*
	No	20	80.0	24	96.0	25	100.0	
Pharyngeal discomfort	Yes	4	16.0	1	4.0	-	-	0.062 NS
	No	21	84.0	24	96.0	25	100.0	

NS: Non-significant  $p > 0.05$ .

\*Significant difference between percentages using Pearson Chi-square test ( $\chi^2$ -test) at 0.05 level.

**Table 5:** The number and percentage of patients/subjects that had mucosal lesion and their oral site in each group with statistical analysis

Mucosal lesion		GERD		Non-GERD		Control		P-value
		No	%	No	%	No	%	
Palatal mucosa	No	20	80.0	25	100.0	25	100.0	0.005*
	Redness	5	20.0	-	-	-	-	
	Ulceration	-	-	-	-	-	-	
Buccal mucosa	No	20	80.0	23	92.0	25	100.0	0.050*
	Redness	5	20.0	2	8.0	-	-	
	Ulceration	-	-	-	-	-	-	

\*Significant difference between percentages using Pearson Chi-square test ( $\chi^2$ -test) at 0.05 level.

The result of this study showed a statistically significant decrease in swallowing frequency, while the time to onset of first swallow was significantly longer in GERD patients (Table 6). Salivary flow rate was significantly decreased in GERD group than in the other groups with a mean and SD of  $0.596 \pm 0.162$  ml/min, the mean and SD of non GERD was  $0.824 \pm 0.148$  ml/min, while mean and SD for the control subjects was  $1.024 \pm 0.171$  ml/min (Table 7).

Laboratory findings

Salivary bicarbonate showed a significant difference between three study groups. Salivary bicarbonate was significantly higher in GERD patients with mean and SD of  $9.048$  mmol/L  $\pm 3.125$ . The mean and SD for the non GERD patients was  $6.212$  mmol/L  $\pm 4.745$ , while for

control subjects was  $4.548$  mmol/L  $\pm 1.848$ . No significant difference in salivary phosphate between GERD group, non GERD group, and control group ( $p > 0.05$ ), as indicated in (Table 8). The aim of the current study was to evaluate bicarbonate and phosphate levels in saliva of GERD patients and to compare oral findings in those patients with patients who had upper gastrointestinal symptoms but who did not have endoscopic evidence of GERD (non-GERD) and with healthy participants. In the current study and compared with the groups of non-GERD patients and control subjects, GerdQ questionnaire score was significantly higher in GERD patients. Use of GERD Q in this study was supported by another study which found that GerdQ can diagnose GERD with the same accuracy as gastroenterologist [10].



Table 6: The swallowing onset and frequency in each group with statistical analysis

		GERD		Non-GERD		Control		P-value
		No	%	No	%	No	%	
Time to onset of first swallowing (second)	<1.0	-	-	2	8.0	8	32.0	0.0001*
	1.0-1.4	-	-	9	36.0	10	40.0	
	1.5-1.9	12	48.0	12	48.0	7	28.0	
	≥ 2.0	13	52.0	2	8.0	-	-	
	Mean ±SD (Range)	2.0±0.3 (1.66-2.45)		1.5±0.3 (0.90-2.05)		1.2±0.3 (0.82-1.94)		0.0001^
Swallowing frequency per 30 second	4	3	12.0	-	-	-	-	0.0001*
	5	4	16.0	-	-	-	-	
	6	11	44.0	2	8.0	5	20.0	
	7	6	24.0	10	40.0	7	28.0	
	8	1	4.0	9	36.0	7	28.0	
	9	-	-	4	16.0	4	16.0	
	10	-	-	-	-	2	8.0	

\*Significant difference between percentages using Pearson Chi-square test ( $\chi^2$ -test) at 0.05 level.

^Significant difference among more than two independent means using ANOVA-test at 0.05 level.

Table 7: Statistical analysis of salivary flow rate between study groups

Salivary flow rate (ml/min) Mean ±SD (Range)	GERD	Non-GERD	Control	P-value
	0.596±0.162 (0.3-0.9)	0.824±0.148 (0.6-1.2)	1.024±0.171 (0.8-1.4)	0.0001^

^Significant difference among more than two independent means using ANOVA-test at 0.05 level.

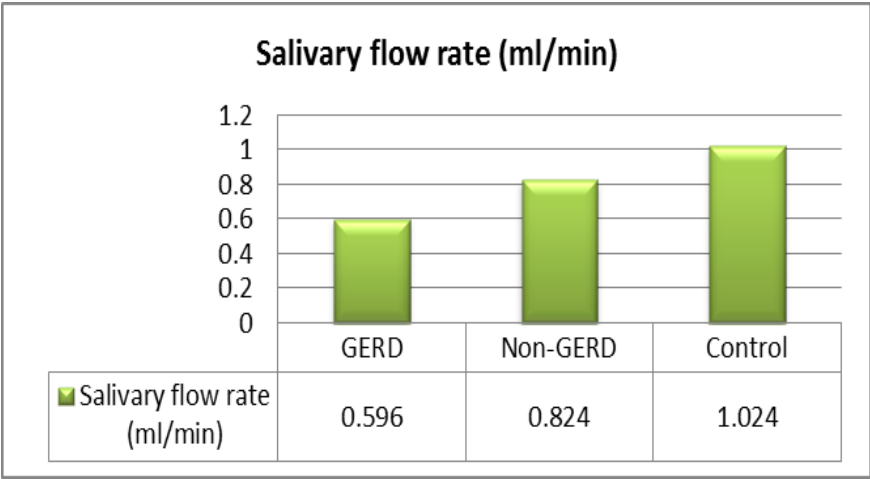


Figure 1: The mean of salivary flow rate for the study groups

Table 8: Statistical analysis of salivary bicarbonate and phosphate between study groups

	GERD	Non-GERD	Control	P-value
Salivary Bicarbonate (mmol/L)	9.048±3.125 (2.1-17.4)	6.212±4.745 (2.6-27.5)	4.548±1.848 (2.2-11.8)	0.0001^
Salivary Phosphate (mg/dL)	8.488±4.061 (4.9-22.8)	8.824±3.975 (5.4-19.3)	8.384±2.965 (4.7-20.7)	0.908 NS

^Significant difference among more than two independent means using ANOVA-test at 0.05 level.

NS: Non-significant  $p > 0.05$ .

GERD was diagnosed by endoscopic examination, and the Los Angeles classification system was used to evaluate the severity of the disease. Los Angeles classification system considered as the most reliable method for assessing GERD erosions [15, 16]. Most of the patients were in the third decade of life with no significant differences between the ages of GERD patients and ages of subjects in another study groups. Some studies were agreed with this finding [17, 18]. The other studies were disagreed with our result and found an association between GERD and age [19, 20]. This difference in conclusion can result from differences in the populations studied or difference in the methods employed to diagnose GERD. In regarding to gender, no significant differences in gender between the GERD patients and another study group this agreed with some studies [21, 22]. Females have a greater incidence of GERD, according to some research findings that indicate GERD is more common in women due to more anxious behavioral state and stress life [23]. Eusebi *et al.* suggested that the prevalence of GERD and their symptoms differs according to the geographical area, even when same terms were employed to describe their presence [24]. In the present study, low salivary flow rate, oral dryness (56%), halitosis (40%), bitter taste, (36%) acid taste (28%) and burning sensation (20%) were found in the GERD patients group. The results are agreed with the other previous studies that found 57.5% of GERD patients felt dry mouth and 48.3% suffer from burning sensation [25, 26] and according to Campisi *et al.*, these findings was caused by reduce in salivary secretion and inadequate gastric acid clearance manifesting as a sensation of dry mouth associated with burning sensation in the oral cavity [26]. In this study, inflammation was observed in oral mucosal area, including the buccal and palatal mucosa with significant difference between GRED group and another study groups for both sites. Di Fede *et al.* in their study, oral mucosal tissues examination in 200 patients with GERD was performed and they found that 21.5% of patients with GERD showed red soft tissue lesions on soft and hard palate mucosa and uvula. They also linked these tissue

changes to direct exposure of tissues to acid [27]. Microscopic analysis of the palatal mucosa in GERD patients showed some microscopic changes include palatal atrophy and increase in the number of fibroblast cells [28]. Some studies reported that buccal mucosa was the most common oral site showed erythematous lesion result from GERD. Study results showed less swallowing frequency and longer duration for first swallow onset in the GERD patients compared with another groups. Same result has been shown by other studies [29].

Salivary bicarbonate was significantly higher in GERD patients than in other study groups. These results were agreed with other studies that found there was significant elevation in bicarbonate levels in patients with GERD group than healthy subjects [30]. Another study showed that salivary bicarbonate secretion was stimulated by esophageal acid perfusion in dose-dependent manner and this response protects the esophageal mucosa from acid injury [31]. Bicarbonate level influences salivary pH and pH increase as bicarbonate concentration increase, but non-stimulated saliva insufficiently buffered because bicarbonate level is too low to be effective [32]. Based on the present results, no significant difference was found in salivary phosphate between established groups and this result was agreed with other studies [33].

## Conclusion

In this research, oral findings as well as bicarbonate and phosphate content in the saliva samples of patients with GERD were described. Oral dryness, acid and bitter taste sensation, halitosis, buccal and palatal erythema, and burning sensation were more prevalent in subjects with GERD. These finding also were associated with lower salivary flow rate. Likewise, the elevated salivary bicarbonate levels were found in the GERD patients with no difference in the salivary phosphate concentration. Hence, routine oral examinations for GERD patients to avoid worsening of the oral manifestations, was advised to be performed.

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## Authors' contributions

All authors contributed to data analysis, drafting, and revising of the paper and agreed to be responsible for all the aspects of this work.

## Conflict of Interest

There are no conflicts of interest in this study.

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