DOI: 10.7860/JCDR/2025/76466.21138

Original Article

Dentistry Section

Assessment of Salivary and GCF pH in Periodontally Healthy and Stage II, Grade B Periodontitis Subjects: An In-vivo Study

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ABSTRACT

Introduction: Oral fluids can be used for the diagnosis of periodontal disease, as they can be easily collected. Saliva is a complex fluid that impacts oral health. Gingival Crevicular Fluid (GCF) is a physiological fluid as well as an inflammatory exudate present in the dentogingival space. It is established that every inflammatory change, along with resultant damage to tissues, leads to altered pH values of these fluids. Periodontal disease is a chronic inflammatory and infectious condition that affects the pH levels of saliva. Furthermore, it is understood that periodontal pathogens grow at acidic pH levels and the growth of these bacteria further contributes to changes in pH levels.

Aim: To assess and compare the pH values of saliva and GCF in periodontally healthy subjects and those with chronic periodontitis.

Materials and Methods: This in-vivo study was carried out at Outpatient Department of Periodontology, Bharati Vidyapeeth (Deemed To Be) University Dental College and Hospital, Pune, Maharashtra, India, over a period of three months, from August 2024 to October 2024. A total of 30 subjects visiting the department of periodontology underwent detailed periodontal examinations and were categorised into two groups: healthy periodontium (Group I, n=15) and generalised stage II, grade B periodontitis (Group II, n=15). Saliva and GCF samples were collected and analysed for pH levels. An independent sample t-test was applied. The analysis was conducted using Statistical Package for the Social Sciences (SPSS) version 25.0. A p-value of less than 0.05 was considered statistically significant.

Results: A total of 30 subjects were studied, of which 20 were females and 10 were males, with a mean age of 32.5 years. In periodontally healthy subjects (Group I), the mean salivary pH was found to be 7.05±0.01, whereas, in stage II, grade B periodontitis subjects (Group II), the mean salivary pH was 6.14±0.60 (p=0.038). The mean GCF pH was 6.73±0.14 in Group I and 8.19±0.29 in Group II (p=0.041). Thus, in chronic periodontitis patients, the salivary pH was acidic and the GCF pH was alkaline compared to periodontally healthy subjects.

Conclusion: The present study indicates that subjects with Stage II, Grade B periodontitis have an acidic salivary pH and a more alkaline GCF pH compared to periodontally healthy subjects.

Keywords: Acidic pH, Alkaline pH, Biomarker, Chairside test, Gingival crevicular fluid, Saliva

INTRODUCTION

Periodontal disease is a chronic inflammatory condition characterised by the breakdown of bone and connective tissue due to bacteria, dental plaque and their byproducts. It is known that any inflammatory changes and the subsequent tissue damage lead to altered pH values in bodily fluids [1]. Therefore, saliva and GCF play an important role in accurately understanding periodontal health status in order to obtain data that mimic periodontitis [2]. The biochemical composition of saliva and GCF, including their pH values and enzyme content, determines oral health and the occurrence of oral diseases [2].

The GCF arises from the gingival vascular plexus within the gingival bulb and is located beneath the epithelium, extending along the gingival cavity. Waerhaug J demonstrated the composition and flow of GCF in 1952 [3,4]. Under healthy conditions, the pH of GCF typically ranges between 6.5 and 7.5. However, in states of periodontal disease, alterations in GCF pH may occur due to dysbiosis, tissue inflammation and the breakdown of periodontal tissues [5]. Later, in 1974, Alfano presented two mechanisms underlying the origin of GCF, including the development of a permanent concentration differential and the induction of classical oedema [6]. Barros SP et al., emphasised the role of GCF as a source of biomarkers for periodontal disease and examined the implementation of crevicular fluid analysis as a diagnostic marker for periodontal disease [7].

Previous literature has shown that GCF is directly related to periodontitis. In the periodontal pocket, its composition changes depending on the presence of inflammation [8]. Bickel M and Cimasoni G, in 1985, measured the pH of GCF and demonstrated that pH values increased and became alkaline with increasing gingival inflammation [9]. In contrast, saliva plays a crucial role in diagnosing specific systemic diseases and evaluating the risk of developing various conditions [10]. Salivary pH is normally maintained between 6.7 and 7.3 [11]. Numerous studies have examined salivary pH and buffering capacity to determine an individual's vulnerability to caries [12-14]. Compared to the clinically healthy group, individuals with chronic generalised periodontitis exhibited more acidic saliva pH levels [15]. Thus, determining the pH of saliva and GCF is considered a potential method for identifying periodontal disease activity.

Hence, the aim of the present study was to assess the pH of saliva and GCF and understand its relationship to periodontitis.

MATERIALS AND METHODS

The present in-vivo clinical study was conducted at the Outpatient Department (OPD) of Periodontology at Bharati (Deemed to be) University Dental College and Hospital, Pune, Maharashtra, India. The research included groups of patients who reported to the OPD. The study was conducted over a period of three months from August 2024 to October 2024 and was approved by the Institutional Ethics Committee (IEC) of Bharati (Deemed to be) University Dental College and Hospital, Pune (Registration No. EC/NEW/INST/2021/MH/0029).

The study population consisted of patients who reported to the OPD and were divided into two groups. Group I comprised 15 subjects with healthy gingiva, while Group II consisted of 15 subjects with Stage II, Grade B periodontitis. The total study population included 30 patients.

Sample size calculation: Sample size was estimated using Opera epi software (Version 3.04) with an alpha (type I error) of 5% and 80% power [16].

Inclusion criteria: In the present study, a total of 30 subjects were included. Group I comprised 15 subjects with healthy gingiva, whereas Group II consisted of 15 subjects with Stage II, Grade B periodontitis (according to the 2017 classification) [17]. The study volunteers were informed about the aim of the study and informed consent was obtained from all participants (as per the Helsinki Declaration). A thorough clinical examination and a detailed history of the subjects were recorded.

Exclusion criteria: The study excluded patients with a history of systemic disorders and/or any conditions that could negatively impact periodontal health, saliva, or GCF composition, such as kidney disease, diabetes, cancer, fungal infections, or respiratory infections. Patients with a current or past history of smoking or tobacco chewing, mouth breathing, malocclusion, or local pathological factors were also excluded. Additionally, patients who were completely edentulous or had a history of medication or hospitalisation within the past six months were excluded from the study.

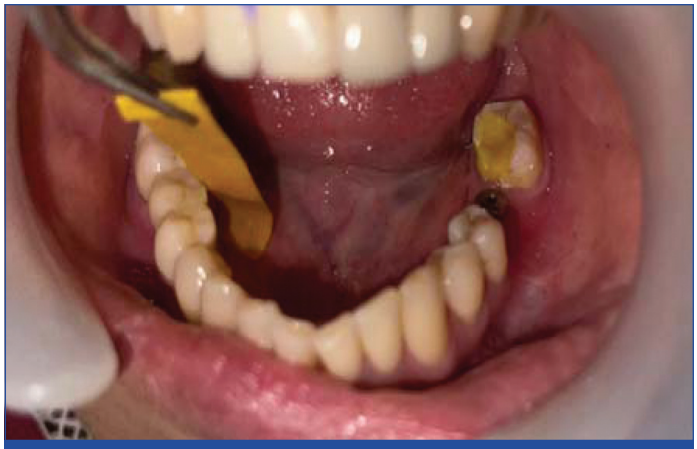
Study Procedure

For each patient, gingival and periodontal findings such as Clinical Attachment Level (CAL) and Probing Depth (PD) were noted. Patients in the control group had clinically healthy gingiva with Probing Depth (PPD) of up to three millimetres, while patients in the test group had clinical attachment loss with pocket depths of ≥5 mm in at least 30% of sites [17].

The procedure used to assess saliva was based on the "Common Minimal Technical Standards and Protocols" guideline provided by the International Agency for Research on Cancer and the World Health Organisation [15]. After an overnight fast during which the individuals were instructed to consume only water, saliva samples were collected in the morning. Saliva was allowed to pool in the buccal and lingual vestibule for five minutes following the chewing of a piece of sugar-free xylitol gum for one minute. The individuals were also instructed not to cough up mucus. The salivary pH was measured by placing litmus paper in the lingual lower front area of the mouth. The salivary pH was then measured using pH paper (Merck pH Indicator Paper) [Table/Fig-1,2].

[Table/Fig-1]: MERCK pH Indicator paper.

For GCF sampling, a Gracey curette was used to remove supragingival plaque prior to GCF collection. Cotton rolls were used to isolate the area and a brief air blast directed straight through the contact helped to dry it (but not into the sulcus/pocket). The pH paper strip was placed into the sulcus for 30 seconds [Table/Fig-3]. Strips that were contaminated with blood were discarded and the colour change was quickly compared to the manufacturer's colour-coded chart.



[Table/Fig-2]: Salivary pH analysis using a pH paper.

[Table/Fig-3]: Assessment of GCF pH.

STATISTICAL ANALYSIS

The study data were entered into Microsoft Excel 2007 and SPSS version 25.0 was used for data analysis. All quantitative data were tabulated using means and standard deviations. Comparisons between the groups were conducted using an unpaired Student's t-test (if the data were parametric) and the Mann-Whitney U test (if the data were non parametric). A p-value less than or equal to 0.05 was considered statistically significant.

RESULTS

In the present study, a total of 30 subjects were included, of which 20 were female and 10 were male, with a mean age of 32.5 years (range: 20 to 55 years).

A comparison of salivary pH levels between Group I (periodontally healthy) and Group II (periodontitis patients) was conducted as shown in [Table/Fig-4]. It was found that the mean salivary pH values for Group I and Group II were 7.05±0.01 and 6.14±0.60, respectively, with a mean difference of 0.91±0.59. The results of the study concluded that the salivary pH levels of patients with chronic periodontitis were more acidic compared to those of healthy subjects and this difference was statistically significant (p-value <0.05).

Parameters	Group	Mean±SD	Mean difference	p-value
Salivary pH	I (Periodontally healthy subjects)	7.05±0.01	0.91±0.59	0.038*
	II (Chronic periodontitis)	6.14±0.60		

[Table/Fig-4]: The comparison of salivary pH levels between Group I (periodontally healthy) and Group II (periodontitis patients).
p<0.05* Statistically significant

The intergroup comparison of GCF pH level values between periodontally healthy subjects and periodontitis patients is displayed in [Table/Fig-5].

Parameters	Group	Mean±SD	Mean difference	p-value
GCF pH	I (Periodontally healthy subjects)	6.73±0.14	-1.46±0.15	0.041*
	II (Chronic periodontitis)	8.19±0.29		

[Table/Fig-5]: The group comparison of GCF pH level values between periodontally healthy and periodontitis patients.
p<0.05* Statistically significant

DISCUSSION

The present study assessed the salivary and GCF pH in periodontally healthy subjects and those with Stage II, Grade B periodontitis. It was found that the mean salivary pH in the diseased group was more acidic compared to that in the periodontally healthy group, while the mean GCF pH in the diseased group was more alkaline.

Saliva is primarily composed of water, accounting for more than 99% of its content. Whole saliva collected from the mouth is a complex blend of various substances and is a thick secretion from both major and minor salivary glands. It also helps maintain the pH of the oral cavity by neutralising the acids produced by bacteria when they metabolise carbohydrates. Salivary pH is normally maintained between 6.7 and 7.3 [18]. Salivary secretion may be quiescent (not stimulated), with contributions of 25% from the parotid gland, 60% from the submandibular gland, 7-8% from the sublingual gland and the remaining 7-8% from the minor salivary glands. The stimulus determines the amount, composition and pH of saliva, with normal saliva flow ranging from 800 to 1500 mL per day [2]. Takahashi N et al., indicated that different periodontal pathogens thrive at different pH levels (*P. gingivalis* grows at a pH of 6.5-7.0, *P. intermedia* grows at a pH of 5.0-7.0 and *F. nucleatum* grows at a pH of 5.5-7.0). The growth of these bacteria contributes to the progression of periodontal disease [19]. Koppolu P, in 2022, studied the correlation between blood and salivary pH levels in healthy individuals, those with gingivitis and patients with periodontitis before and after non surgical periodontal therapy. They found that saliva from subjects with periodontitis had a more acidic pH compared to the healthy group and subjects with gingivitis.

Korte DL and Kinney J explored the potential of using saliva as a diagnostic tool for periodontal disease [20]. Fujikawa K et al., stated that in deep periodontal pockets or in cases of extensive gingival inflammation, pH levels decrease [21].

The GCF is a physiological fluid and an inflammatory exudate [22]. It can help in assessing the severity of gingival diseases, the effectiveness of periodontal therapy and oral hygiene. GCF pH is alkaline (pH 7.5-8.7) in periodontally healthy subjects [23]. In the present study, GCF pH was found to be more alkaline than that in subjects with a healthy periodontium (8.8-9). Furthermore, Bickel M and Cimasoni G measured the pH of GCF and demonstrated that pH values increased and became alkaline with greater gingival inflammation [9]. They further reported that it was unclear whether this increase was due to the bacterial flora or to any metabolic activity in the crevice area.

Limitation(s)

The present study had a few limitations. In the field of periodontics, understanding the buffering capacity of GCF is important because it helps to maintain a stable pH in the periodontal pocket, even when acidic by-products are produced by bacteria. However, pH paper does not measure buffering capacity, meaning that important aspects of periodontal health or disease dynamics are overlooked.

CONCLUSION(S)

Saliva and GCF are easily accessible fluids that contain indicators of periodontal infection, making them potential tools for the prognostic assessment of periodontitis. This in-vivo study demonstrated significant differences in the pH levels of saliva and GCF between periodontally healthy individuals and those with Stage II, Grade B periodontitis. The findings suggest that periodontal disease is associated with altered pH values in both saliva and GCF, highlighting the potential role of pH as a biomarker in the progression and management of periodontal disease. Further research is needed to explore the clinical implications of pH alterations in these fluids for diagnostic and therapeutic strategies in periodontitis.

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PLAGIARISM CHECKING METHODS: [Jain H et al.]
- Plagiarism X-checker: Oct 24, 2024
- Manual Googling: Feb 26, 2025
- iThenticate Software: Mar 15, 2025 (16%)

ETYMOLOGY: Author Origin

EMENDATIONS: 8

AUTHOR DECLARATION:
- Financial or Other Competing Interests: None
- Was Ethics Committee Approval obtained for this study? Yes
- Was informed consent obtained from the subjects involved in the study? Yes
- For any images presented appropriate consent has been obtained from the subjects. NA

Date of Submission: **Oct 23, 2024**
Date of Peer Review: **Jan 06, 2025**
Date of Acceptance: **Mar 18, 2025**
Date of Publishing: **Jun 01, 2025**