Evaluation of the Salivary level of Sphingosine _ 1 phosphate in periodontitis

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Abstract

The reactive signaling molecule sphingosine-1-phosphate (S1P) plays a vital function in many biological processes that encompass cell development. In addition, it has been linked to bone resorption and formation. Based on its biological effects on osteoclastic and osteoblastic cells and immune cells, S1P has recently been discovered as a mediator and biomarker in inflammatory bone disorders such as osteoporosis and inflammatory osteolysis. Sphingosine-1-phosphate (S1P) may play a role in developing periodontitis, an inflammatory disorder that destroys bone. This study aims to evaluate the salivary level of S1P in periodontitis. The study sample consisted of 70 participants, both males and females. It was divided into three groups: the first group, the Healthy Control group (20 Subjects); the second group, Periodontitis Stage II (25 Subjects); and the third group, Periodontitis Stage III (25 Subjects). Clinical periodontal characteristics were evaluated after entire, unstimulated saliva samples were collected from all individuals (plaque index, probing pocket depth, bleeding on probing, and clinical attachment level). As a biomarker, the amount of S1P in the saliva was measured using an enzyme-linked immunosorbent assay (ELISA). This study demonstrated a statistically significant correlation between higher mean S1P levels and higher periodontitis severity. The study demonstrated that the salivary S1P level could help to monitor periodontal disease progression.

Keywords: Sphingosine -1 phosphate, Periodontitis, Saliva.

Introduction

Initiated by bacterial biofilm and further modified by some risk factors, periodontal diseases are among the most widespread health issues impacting modern humans [1]. These conditions affect the teeth' supporting structures (gingiva, bone, and periodontal ligament), potentially resulting in tooth loss. The commensal oral microbiota interacts with the host's immune responses, leading to inflammation and periodontal disease [2]. In most of the world's adult populations, periodontal disease, especially its mild and moderate forms, is quite common, with prevalence rates of approximately 50%. [3], while its severe form is more common in the third and fourth decades of life, with a global prevalence of approximately 10% [4]. Periodontitis is an inflammatory condition brought on by bacteria and other factors.[5], it is affecting the periodontal ligament and alveolar bone [6]. Loss of alveolar bone, which supports teeth, is an irreversible consequence of periodontitis because it triggers osteoclastogenesis [7].

Sphingosine-1-phosphate (S1P) is a bioactive sphingolipid generated through the phosphorylation of Sphingosine 1-kinase (SPHK) that affects a variety of cellular processes [8]. Van Brooklyn et al. research .'s indicates that S1P serves as both an external first messenger through its cell surface receptor and an intracellular second messenger to promote cell proliferation and inhibit apoptosis. [9]. Sphingosine 1 _phosphate has been linked

to osteoblast and osteoclast differentiation during bone remodeling [10]. Researchers have shown that Sphingosine 1 _phosphate plays a role in inflammatory illnesses like rheumatoid arthritis and bone homeostasis [11]; moreover, it plays a role in maintaining bone tissue homeostasis by initiating chemotaxis and controlling the migration of osteoclast precursors [12]. However, its function in periodontitis-related osteoclastogenesis is still not well understood.

Saliva as a diagnostic tool has gained popularity in recent years. Patient saliva collection is less stressful than collecting any other bodily fluid and its low-cost storage [13]. Particular attention has been paid to salivary components as a possible indicator of periodontal disease [14]. Oral disorders such as periodontitis, peri-implantitis, primary Sjogren's syndrome, oral lichen planus, oral leukoplakia, and medication-related osteonecrosis of the jaw can all be diagnosed and their severity predicted by analyzing the patient's saliva [15].

Because clinical studies evaluate Sphingosine 1_ phosphate in individual periodontium in health and disease are insufficient, throughout this study, we aimed to evaluate the salivary level of Sphingosine _1 phosphate in periodontitis.

Materials and Methods

Seventy male and female participants aged 30-50 years old who met the study's inclusion and exclusion criteria were used in this observational case-control study. The periodontics department of the college of dentistry at the university of Baghdad provided the samples. Using a form approved by the ethical council of the College of Dentistry at the University of Baghdad, we gained each participant's informed consent after providing them with comprehensive information about the study and all procedures.

A minimum of 20 teeth were required for inclusion, and so was systemic health and a lack of recent anti-inflammatory or antibacterial medication use. Patients who have had or are currently undergoing effective periodontal treatment; patients who have been on a course of anti-inflammatory or antibacterial therapy within the past three months; patients who are smokers or heavy drinkers; patients who have chronic systemic disease; patients who are immunocompromised; pregnant, pill-taking, or lactating women; patients with diseases of the soft and hard palate and mucosa; patients who wear orthodontic appliances, removable dentures, implant, crown, and bridge were omitted.

The study's subjects were categorized into three groups: the control group with healthy intact periodontium (n=20) [16], and the periodontitis patients (cases) divided into two groups, including stage II and III periodontitis patients (n=50) [17]. The control group and the periodontitis patients were as follows:

- Control healthy intact periodontium (20 subjects): no probing attachment loss, Probing pocket depths ≤3 Mm, Bleeding on probing <10%, and no Radiological bone loss (Chapple et al., 2018).
- Stage II periodontitis (25 patients): (bone loss involving Coronal 1/3 of the root).
- Stage III periodontitis (25 patients): (bone loss involving the middle 1/3 of the root).

All subjects had their clinical periodontal parameters measured by the same person. Using a periodontal probing tool (the University of Michigan O probe marked by Williams at 1, 2, 3, 5, 7, 8, 9, and 10 mm), we determined periodontal parameters (PLI, BOP, PPD, and CAL). Each tooth was looked at from six different angles (mesiobuccal, buccal, distobuccal, mesiolingual, lingual, distolingual), except for the plaque scores, which only looked at four of these angles (mesial, buccal, distal, lingual). The patient's wisdom teeth did not participate in the study.

The salivary level of S1P was measured by collecting samples from patients and healthy

controls. Donors were asked to sit comfortably with their legs propped up, avoid eating or drinking (apart from water) for at least one to two hours before saliva collection, and throw out any samples that showed signs of blood. Between 9 and 12 a.m., saliva samples were taken. Five milliliters of the whole, unstimulated saliva were collected from the participant after he swished the water in his mouth several times to eliminate any debris or contaminated material before saliva collection. For the experiment, 3 ml of saliva was centrifuged at 2500 rpm for 20 minutes, and the resultant supernatant was kept in Eppendorf tubes at -20 C.

Using the ELISA technique, the biomarker analysis of salivary S1P was done using a kit manufactured by SUNLONG Biotech Co., Ltd, China

This information was characterized, analyzed, and shown using SPSS version 21 (Chicago, USA, Illinois). Nominal variables were examined using the mean and standard deviation. In contrast, quantitative variables were checked for normality with the Shapiro-Wilk and D'Agostino-Pearson tests and One-Way Analysis of Variance (ANOVA) with the Games-Howell posthoc test and the Tukey Kramer-Kruskal-Wallis (K-W) test (S.D.).

Results

The S1P descriptive statistics demonstrate that the healthy periodontal control group had the lowest mean value. As shown in Table 1, and figure 1 this variable rose markedly across all groups as periodontitis severity rose.

Table 1: Descriptive and statistical test of S1P (ng/L) among groups using One Way (ANOVA).

Groups	Mean	±SD	±SE	Minimum	Maximum	F	P value
Control	141.53 (ng/L)	29.15	7.52	92.79	183.92	5.27	0.007 **Sig.**
Stage-II	170.165(ng/L)	37.71	6.88	101.96	242.82		
Stage-III	187.16 (ng/L)	55.55	10.14	103.97	317.59		

*=significant at p≤0.05.

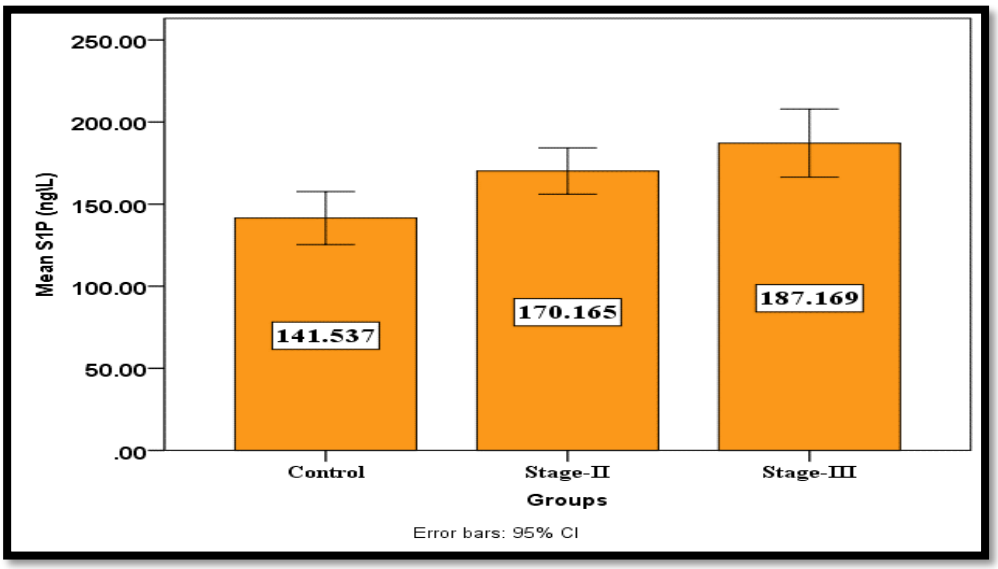

Figure 1: The bar chart represents the mean value of salivary S1P (ng/L) among groups.

Discussion

Data from the S1P biomarker study showed that the average level of S1P was lowest in the healthy periodontal control group and increased significantly with increasing stages of periodontitis. The loss of endothelial barrier integrity and the accompanying increase in vascular tone may be attributed to the hyperpermeability of the endothelial barrier induced by high concentrations of S1P. This mimics what happens in periodontitis, where inflammatory chemicals in the bloodstream penetrate the epithelium separating the two surfaces of the gingiva [10, 18, 19].

Yu et al. found that periodontitis is an inflammatory disease that causes bone loss due to germs. When oral pathogens like A. actinomycetemcomitans, the pathogen linked to localized aggressive periodontitis, invade healthy tissue, they set off a cascade of events that ultimately results in periodontal soft tissue destruction, alveolar bone resorption, and tooth loss. This research shows that the oral pathogen Actinomycetemcomitans stimulated the production of S1P by activating SPHK-1 (an enzyme that converted Sphingosine into S1P). An absence of SPHK-1 significantly reduced the chemotactic response of monocytes and macrophages to A. actinomycetemcomitans in vitro. Supporting previous research, this study demonstrates that SPHK1 and S1P are critical for controlling the inflammatory bone loss response triggered by the oral pathogen A. actinomycetemcomitans [11].

Researchers Moritz et al. observed that S1P levels in the blood were significantly higher in those with moderate to severe periodontitis than in those with healthy gums and teeth. Increased SPHK-1 expression in macrophages and related immune cells may cause locally raised S1P levels, which were observed to correlate with many periodontitis variables measuring the severity or extent of the present (PPD) or long-term (CAL) periodontal disease [20].

Conclusion

Increases in salivary S1P levels have been observed with increasing periodontitis severity, suggesting that S1P can be utilized to track the development of periodontal disease.

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Conflicts of Interest:

As stated by the authors, there is no bias or conflict of interest.

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