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Research Report

Comparison of salivary alpha-amylase levels in gingivitis and periodontitis

Dyah Nindita Carolina, Yanti Rusyanti, and **Agus Susanto**
Department of Periodontics
Faculty of Dentistry, University of Padjajaran
Bandung – Indonesia

ABSTRACT

Background: *The development of periodontal disease is influenced by bacteria-plaque, while there are also several factors modifying the host's response, one of which is psychological stress. Alpha-amylase as a biomarker is also associated with periodontal inflammatory disease.* **Purpose:** *The purpose of this study was to examine the difference of alpha-amylase level between gingivitis and periodontitis.* **Methods:** *This research constitutes a descriptive study involving 44 subjects, divided into two groups: one of 22 gingivitis subjects and the other of 22 periodontitis subjects. These individuals completed a PSS-14 questionnaire before their levels of alpha salivary amylase were measured by Cocorometer. Data was analyzed by means of a paired T test and a Mann Whitney test with p < 0.05.* **Results:** *There were significant differences between the alpha-amylase levels of gingivitis and periodontitis. However, no significant contrast existed in the PSS-14 scores of the two periodontal disease groups.* **Conclusion:** *In conclusion alpha-amylase levels in the periodontitis group were higher than those in the gingivitis group and could be used as marker indicators of stress.*

Keywords: *alpha-amylase level; gingivitis; periodontitis; saliva; stress*

Correspondence: Dyah Nindita Carolina, Department of Periodontics, Universitas Padjajaran, Jl. Sekeloa Selatan I, Bandung 40132, Indonesia. E-mail: dyah.nindita@unpad.ac.id

INTRODUCTION

The oral cavity can be regarded as one site of systemic diseases because several such ailments can be identified through its overall condition. The parameters of oral examination in diagnosing disease may comprise the clinical condition of the cavity supported by analysis of, for example, the blood, gingival crevicular fluids and saliva. Blood and saliva can be used to monitor systemic conditions indicating the general state of oral health.[1,2] Saliva has achieved rapid adoption as a biomarker for more than a decade, especially in the fields of psychology and biomedical research.[3] The analysis of saliva has several advantages including: rapid and easy collection of samples, the lack of need for special equipment and/or operators, non-invasive and painless processes, as well as the absence of stress caused to the patient. Saliva contains several biomarkers such as cystatins, albumin, alpha-amylase, IgA, and cortisol. Certain studies argue that alpha-amylase as a biomarker is also associated with periodontal inflammatory disease.[4,5]

Psychological stress is one of the important risk factors in periodontitis. One study highlighted the significant numbers of people suffering from psychosocial or occupational stress (17.05%) and the complex mechanisms through which such stress could affect their periodontal condition. It suggested that one possible cause involves modification of undesirable behavior such as smoking and poorly maintained oral hygiene.[6] The measurement of psychological stress can be viewed subjectively based on individual feelings and thoughts regarding recent developments, one of the most commonly used measuring tools being perceived stress scale-14 (PSS-14).[7] Stress is one factor evident in physiological changes manifested by an individual and an analysis of saliva can determine the associated level of stress.[8]

Periodontal diseases, most notably gingivitis and periodontitis, are defined as inflammation caused by pathogenic microflora making up the biofilm surrounding the teeth. Gingivitis is an inflammatory condition that affects the surface of the gingiva and is reversible in the absence of alveolar bone destruction, while periodontitis is a disease resulting in damage to the tooth-supporting tissues that can cause tooth loss.[9,10]

This study used alpha-amylase enzyme as a parameter of the severity of periodontal disease and a marker of psychological stress in the patient. Within this study, the selection of alpha-amylase was based on the assertion of Ackali *et al*. that it is one of the main saliva-based markers serving as a stress indicator that has been investigated in the hope of confirming the interrelationship between psychological stress and periodontal disease.[9] It is also associated with other studies suggesting that alpha-amylase levels in saliva tend to increase in individuals with periodontal disease and in others who are psychologically stressed.[5,7,8] Tanaka *et al*. identified a practical, Japanese-made tool (the Nipro Cocorometer) for measuring stress based on alpha-amylase levels in saliva.[11] Using a Cocorometer, the present study measured alpha-amylase levels in patients afflicted with gingivitis and periodontitis. Furthermore, these levels are used as stress parameters when observing the relationship between periodontal disease and psychological stress. The authors were interested in conducting research into the measurement of salivary alpha-amylase levels in patients with gingivitis and periodontitis. The measurement results for alpha-amylase levels were also linked since psychological stress parameters and psychological stress were often linked as a risk factor influencing the occurrence of periodontitis.

MATERIALS AND METHODS

The research was conducted on 44 patients of the Periodontic Department, Dental Hospital, Universitas Padjadjaran, selected according to the inclusion criteria of there being 22 gingivitis patients and 22 chronic periodontitis patients. The inclusion criteria of subjects included the following: all were aged 17–55 years old, all examined teeth afflicted by gingivitis had probing depths of Ý 3 mm, those with periodontitis had a probing depth of š 4 mm and a loss of clinical attachment of š4 mm. As confirmed by screening conducted by an internist, no subjects suffered systemic disease: diabetes mellitus (DM), rheumatoid arthritis or kidney disease. None had undergone periodontal therapy or taken antibiotics or contraceptive pills during the preceding three months, nor were any pregnant or lactating. This study used a non-random technique, selecting samples by determining which subjects met the research criteria included in the study within a certain time period.[5]

The tools and research materials utilized included: a Cocorometer, Nipro Co., Japan,[10] glass mouth, explorer, Williams probe, gloves and mask, mouth rinse and a cotton roll. The research was descriptive in nature, comparing the level of alpha-amylase in the specific forms of periodontal disease, namely; gingivitis and periodontitis. The required ethical clearance was issued by the Commission of Dentistry Ethics, Faculty of Dentistry, Universitas Padjadjaran. Clinical examination was performed on the oral cavities of patients suffering from gingivitis and periodontitis who attended the Department of Periodontics, Dental Hospital, Universitas Padjadjaran. The objective of the PSS-14 questionnaire completed by patients upon termination of the oral examination was to measure psychological stress based on individuals' subjective thoughts and feelings about their ongoing situation. The stress level criteria relating to PSS-14 scores were as follows: low: 15–19, medium: 20–24, high: 25–29, very high: > 30.[6] Alpha-amylase measurement was conducted using a Cocorometer (Nipro, Japan) tool to monitor an individual's physiological changes, particularly by determining the associated stress level through the analysis of saliva. The end of a measuring stick was placed at the base of the tongue for one minute until it had become wet (Figure 1A). It was then inserted into the Cocorometer tool (Figure 1B).[11] The results of alpha-amylase levels were recorded on the Cocorometer's monitor which operates on the basis that a rise in norepinephrine will affect the level of alpha-amylase in human saliva (Figure 1C). Alpha-amylase reacts faster than cortisol and norepinephrine to acute stress. Therefore, the instant a person feels stressed, it will appear in the results. The criteria for stress levels based on the alpha amylase level (KU/l) are as follows:[10] low: 0–30, medium: 30–45, height: 46–60, very high: > 61.

Figure 1. (A) Measurement of alpha-amylase content using Nipro Japan cocorometer tool. (B) The sticks were included in the Nipro Japan cocorometer tool. (C) Record the results of the alpha-amylase levels listed on the Japanese Nipro Cocorometer monitor.

Table 1.   Characteristic of subjects

Characteristic	Groups	
	Gingivitis (n = 22)	Periodontitis (n = 22)
Gender		
Male	11	10
Female	11	12
Age		
X (SD)	23.9 (7.1)	40.9 (6.9)
Median	21	39
Range	17-47	32-55

Table 2.   Examination of alpha-amylase levels and PSS-14 score in the groups of gingivitis and periodontitis

Characteristic	Groups	
	Gingivitis (n = 22)	Periodontitis (n = 22)
Alpha-amylase		
X (SD)	82.4 (52.7)	147.6 (101.4)
Median	74	118
Range	7–193	10–436
Score PSS-14		
X (SD)	23.1 (4.0)	22.7 (4.4)
Median	22	23
Range	18-31	15-31

The measurements of alpha-amylase levels in groups of individuals suffering from gingivitis were compared with those afflicted with periodontitis.

The research data was processed by calculating the mean, standard deviation, median and range. The average difference in alpha amylase levels was analyzed by means of a Mann Whitney test, while the PSS Score was calculated using a t-test.

RESULTS

The research reported here was conducted on 44 subjects all of whom were patients of the Department of Periodontics, Dental Hospital, Universitas Padjadjaran. The inclusion criteria of 22 individuals suffering from gingivitis and 22 afflicted with chronic periodontitis were fully met. Their demographic characteristics were shown in Table 1. The alpha amylase levels and PSS-14 score of sufferers of periodontal disease (gingivitis and periodontitis) can be seen in Table 2.

Table 1 contains data on the general characteristics of the study sample with regard to gender and age. In terms of gender, the gingivitis group contained 11 males and 11 females, while in the periodontitis group there were 10 males and 12 females. When tested statistically, the latter did not show any significant differences. As for the age of the subjects, the mean and median of the periodontitis group

were both higher than those of the gingivitis group.

The alpha-amylase levels and PSS-14 scores in those individuals suffering from periodontal disease (gingivitis and periodontitis) can be seen in Table 2. The mean alpha-amylase level of 82.4 KU/l in gingivitis patients compared to one of 147.6 KU/l in members of the periodontitis group. The median of the periodontitis group was greater than that of the gingivitis group. An examination of the scores on PSS-14 revealed that those of the periodontitis group ranged from 15 to 30 with an average of 22.7, while in the gingivitis group they ranged from 18 to 31 with an average of 23.1.

DISCUSSION

The progress of periodontal disease remains unclear, although it is influenced by differences in the respective susceptibility of individuals. The progression of periodontal disease can be more rapid (aggressive) given the presence of systemic or environmental factors such as diabetes mellitus, smoking, or stress.[2,3,12] that influence the host's response to plaque. Stress reduces salivary flow and supports bacterial plaque formation, while emotional stress modifies pH levels and salivary composition.[13] Saliva contains several proteins that can be used as markers of stress, one such indicator being alpha amylase.[9] The results of this study confirmed that there was an average difference in alpha-amylase levels in gingivitis patients and periodontitis patients. In this study, the alpha-amylase levels present in cases of periodontitis were higher, compared to those in gingivitis patients. This finding was consistent with that of research conducted by Sanchez *et al.*[5] which suggested that periodontitis may stimulate the increased production of proteins in saliva, including mucin and amylase. The concentration of these two types of protein is increased in the sufferers of periodontitis moderate to severe in severity. The increasing alpha-amylase levels in periodontitis were associated with a study by Papacosta which stated that salivary alpha-amylase is the first line of defense. According to Rohleder, this enzyme prevents pathogens entering the body through the mucosal surface and could be considered as the best indicator of mucosal immunity in the oral cavity by inhibiting attachment of bacterial growth.[14,15]

The stress levels of patients suffering from gingivitis and periodontitis which were measured in this study by PSS-14 confirmed there to be no difference in stress levels between members of either group. These were supposedly related to the level of subjectivity of each individual under stress recorded by the measuring tool which used a PSS-14. Measures the feelings and thoughts of individuals related to situations that occurred within the previous month. Research subjects were asked to choose the statement that best suitable of his condition.[7] The perceived stress scale (PSS) was not a diagnostic tool, as there was no standard reference value that measures stress levels. However,

from these measurements, comparative values in groups of research samples could be observed. Stress was part of being human which occurs to varying degrees and produces different effects on one's health.[16] Several questionnaires such as the Perceived Stress Scale have been developed to better identify and classify stress, but this questionnaire had deficiencies in the standardization of psychological scales for the quantity and definition of stress.[13] A more thorough questionnaire that includes lifestyles and other powerful influences such as gender, age and characteristic traits would be better at clarifying a person's stress levels relating to the developments that occur in his or her life.[9]

There are certain factors, conditions or occurrences, that cause stress. These are often referred to as the stressor. The response or reaction to something that can cause stress and the processes that actively occur affect its impact on the individual. This phenomenon is called the locus of control and constitutes a measure of the extent to which an individual believes he has control over his life and where such control comes from.[17,18]

Levels of alpha-amylase measured by means of a Cocorometer were used as a marker of stress. When the levels were higher than normal, they were expressed as stress (> 60 KU/l).[11] The group suffering from more severe periodontal disease (periodontitis) demonstrated a tendency towards higher levels of stress. Such elevated levels were in accordance with the findings of research conducted by Shende which suggested that psychosocial factors such as severe stress, personality type and individual coping strategies to deal with a stress environment can modify the immune response. These result in an individual being more sensitive to unhealthy conditions which also affects the state of periodontal tissue.[19] Patients suffering from psychiatric disorders demonstrated a level of susceptibility to periodontitis greater than that of individuals free of such conditions.[20]

This study shows that the objective measurement of alpha-amylase levels in the periodontitis group was higher compared to that of subjects with gingivitis. However, in the assessment of PSS-14, there was no difference in stress levels between the gingivitis and periodontitis groups. This was influenced by individual subjectivity in completing the questionnaire. Stress is a non-observable abstract condition that cannot be precisely measured by the completing of such a survey alone. Other forms of measurements, including interviews and observations from the experts concerned, were required. Their absence must, therefore, be regarded as representing a major limitation on the rigor of this study. In conclusion alpha-amylase levels in the periondontitis group were higher than those in the gingivitis group and could be used as marker indicators of stress.

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