THE LEVELS OF TNF-A IN GINGIVAL CREVICULAR FLUID (GCF) OF OSING TRIBE WOMEN WITH OCCLUSAL DISHARMONY

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INTRODUCTION

The Osing tribal region is demographically adjacent to the island of Java, Madura and Bali. The proximity of this demographic regions affects multiple systems of their organization, culture, and arts. The Osing's familial and societal patterns are similar to the other Javanese tribes, including housing, food, lifestyle and health. Osing tribe is frequently compared to Balinese culture, such as traditional clothing, wedding dresses, and others [1]. One of the cultures associated with the teeth is Pangur or asa i.e. reducing/flattening the occlusal surfaces of the teeth especially for Osing bride women. In addition, there is also a tradition of Osing women called Nginang or betel chewing [2]. This situation may lead to an imbalance of dental occlusion and disruption of the normal flora in the oral cavity alleging effect on oral health

Dental and oral health can be achieved if there is a relationship of harmony and balance between the organs and tissues in the oral cavity. This is because the oral cavity is an integral stomatognatic complex system.

Stomatognatic system is unitary organ that has mutual-related functions. These organs include jaw bones, temporo mandibular joint (TMJ), teeth, and other supporting structures e.g. mastication muscles, facial muscles, head and neck. This stomatognatic system plays an active role in the process of mastication. In normal conditions, it occurs a harmonious relationship of all components of the masticatory system i.e. teeth, muscles, TMJ, lips, cheeks, palate, tongue and salivary secretion [3]. The conditions of normal tooth anatomy and well-structured on curved teeth will put both condyle joints at the center of the articular disc. This situation will create an effective mastication process [4].

Abnormal condition of tooth structure will cause malocclusion [3]. Occlusion is an interocclusal relation (intercups) between the teeth of each jaw in accordance with the position and movement of the mandible [5]. Occlusion pathological state is known as occlusal disharmony. Occlusal disharmony can be caused by several factors such as the loss of teeth, caries, attrition, tooth anomalies, bruxism, and the habit of chewing on one side [6].

Occlusal disharmony is suspected to cause inflammation in periodontal tissues. Periodontal tissue disease is one of the dental and oral disease that most commonly occurs. Periodontal diseases can be caused by bacteria or mechanical the overloads. The overloads can be caused by the occlusal disharmony. Thus, occlusal disharmony is suspected to cause inflammation in the periodontal tissues. Abnormal oocclusion condition in this occlusion disharmony leads into occlusal trauma which will increase the load received by the periodontal tissue [7]. If this condition is persistent, it will cause periodontal tissue inflammation. Inflammation undergoing locally to the periodontal tissues will induce the immune system to increase the activity of the inflammatory cytokine secretion, one of which is TNF- α (tumor necrosis factor-alpha). TNF- α will play a role in activating osteoclasts, which can result in the destruction of the alveolar bone [8]. This inflammatory cytokines can be found in gingival crevicular fluid (GCF), which are inflammatory exudates collected in the gingival sulcus [9].

TNF- α is a cytokine that plays a role in the occurrence of periodontal disease through its activities, which triggers proliferation, differentiation and activity of osteoclasts resulting in bone resorption, and inducing production of proteinase in the cells of the mesenchyme that is responsible in hond tissue destruction. In patients with periodontitis it occurs increased levels of TNF- α , which will be decreased after periodontal treatment. Based on the description above, it is necessary to know the levels of TNF- α in GCF Osing women as an indicator of periodontal tissue inflammation due to occlusion disharmony. It is important for prevention and treatment of abnormalities of occlusion disharmony that will improve oral health and quality of life, especially in the Osing community in Kemiren village, Glagah District, Banyuwangi Regency.

METHODS

Prior to this research, an ethical feasibility study was conducted by the Ethics Committee. The subjects of the study was obtained from the female population Osing tribe in Kemiren Village, Glagah District of Banyuwangi Regency elected by incidental random sampling for groups with occlusion disharmony, and the group without any disharmony occlusion (control group) in accordance with the criteria. The criteria of the study subjects were Osing women aged 30-65 years old, suffering from occlusal disharmony characterized by the presence of one or more disorders (tooth loss in posterior area, attrition of teeth, bruxism, wide caries, anomalies location / position of the teeth, habit of chewing on one side), not having ever smoked, not suffering from systemic diseases e.g diabetes mellitus, hypertension, renal rheumatoid arthritis disease, and other inflammation disesases, not using mouthwash at least 6 months and not being in the periodontal treatment for last 6 months at least. The research subjects were given an explanation of the purpose and procedures of GCF-taking and had filled informed consent. The normal group was obtained from Osing woman population who did not suffer from occlusal disharmony or normal. All of the subjects filled out a questionnaire of oral health.

GCF on molar or premolar of right and left maxilla was taken from each group. Prior to taking the GCF, the area around the tooth in which the fluid would be taken was cleaned and isolated with a cotton roll to avoid contamination from saliva, plaque, calculus and blood. GCF was taken using paper points sized # 20 inserted into the proximal area of the gingival sulcus and allowed to stand for 30 seconds. Then the paper points were inserted into 0.5 mL of eppendorf. Then stored in a deep freezer with a temperature of -30 ° C up to be measured levels of TNF- α .

Measurement of TNF- α levels used techniques sandwich enzyme linked immunosorbent assay (ELISA). Furthermore, the data resulted from the measurement of Independent T-test with a significance of p <0.05.

RESULT AND DISCUSSION

Geographically, *Osing* tribe inhabites the area in the Kemiren Village, Glagah District of Banyuwangi Regencey. The presence of other tribes in Banyuwangi e.g. Java, Madura and Bugis, does not alter the general view including *Osing* people themselves that the so-called Banyuwangi society is the society of *Osing*.

Residents in this village is a group of people who have distinctive customs and culture as one tribe known as the Osing. It has a special place to implement their custom events known as Rumah Osing. They earn their mostly as farmers.

Based on the interviews and the questionnaire, it is known that they have the same diet like the general population i.e consuming the staple food e.g rice, vegetables and side dishes. Their insight on oral health is almost as well as other communities like brushing teeth at least once during the evening bath. A visit to the dentist in the clinic is conducted if they have tooth pains. Most of occlusal disharmonies occur in *Osing* women caused by tooth loss and attrition.

Levels of TNF- α produced from the GCF specimen of *Osing* women with occlusal disharmony and those with normal occlusion are shown in Table 1.

Table 1. The Results of TNF- α Levels Measurement on GCF (nmol/L)

Groups	Mean ± SD
Occlusal Disharmony	83,22 ± 16,95
Normal	24,40 ± 5,45



Figure 1. Mean of TNF- α **Levels on GCF Subjects** Based on the Table 1 and Figure 1, shown that TNF- α levels were higher in subjects with occlusal disharmony (83.2 nmol/L). Furthermore, the statistical analysis with Independent T-test showed significant differences between groups of subjects with occlusal disharmony and normal occlusion. Levels of TNF- α in GCF of occlusal disharmony subjects was higher than that of normal subjects (see at table 2).

Table 2. The Results of Independent T-test of TNF- α Levels on GCF

Groups	Sig. (2-tailed)
Occlusal Disharmony	- 0.000*
Normal	
Note :	

* : significant diffrences

TNF- α is a cytokine that plays a role in the occurrence of periodontal disease through its activities including proliferation trigger, differentiation and activity of osteoclasts that results in bone resorption, and induce production of proteinase in the mesenchyme cells that are responsible in bond tissue destruction. In patients with periodontitis levels of TNF- α are increased and will be decreased after periodontal treatment. The

measurement of periodontal tisues results showed that 45% of *Osing* women that became the subject of research suffered from periodontal disease in the early stages of periodontitis .

The results also show that increased levels of TNF- α in periodontal tissues were detected in GCF. Periodontal tissue abnormalities can be caused by bacteria or because of the overload [10]. The overloads can be caused by the occlusal disharmony. Occlusal disharmony can cause inflammation in the periodontal tissues. Abnormal oocclusion condition in the occlusal disharmony became occlusal trauma which would increase the load received by the periodontal tissue [7]. If this condition is persistent it will cause periodontal tissue inflammation.

The results of this study indicate that TNF- α levels were increased in GCF of Osing women suffering from occlusal disharmony. It demonstrates that the disharmony that occurs is suspected be a trigger of periodontal tissue inflammation. Occlusion disharmony is experienced due to the premature contacts, caries, attrition and loss of teeth leading to occlusal loads on periodontal tissue becomes excessive. The load on periodontal tissue will cause periodontal ligament damage. This damage will trigger inflammation and migration of inflammatory cytokines into the junctional epithelium [11]. This will be exacerbated by a low oral hygiene conditions. The results also show a lack of their knowledge on oral hygiene. Therefore, the occlusal disharmony causing excessive occlusal forces on the tissue and the possibility of bacterial plaque accumulation becomes a trigger factor of periodontal tissue inflammation.

Inflammation that occurs locally to the periodontal tissues will induce the immune system to increase the secretion activity of inflammatory cytokines i.e TNF- α [8]. These inflammatory cytokines found in gingival crevicular fluid (GCF), which are inflammatory exudatess collected in the gingival sulcus [12].

Inflammation causes increased vascular permeability resulting in extravasations of leukocyte cell. Serum proteins such as complement, acute phase proteins and plasmin system will further enhance inflammatory responses and activate endothelial cells to produce more cytokines. Cytokines activate macrophage cells to produce mediators such as TNF α , IL-8, 1L-6, IL-10, IL-12, PGE2, MMP, interferongamma (IFN-gamma), and chemokines such as MCP and MIP. After the initial phase of inflammation occurs, mononuclear cells such as macrophages and lymphocytes begin to infiltrate.

TNF- α is also a multipotential cytokines that have various biological effects and known to have a similar effect as IL-1. TNF- α is produced primarily by macrophages as the responses to agent such as lipopolysaccharides. TNF- α and IL-1 are both known to act on endothelial cells to increase the adhesion of polymorphonuclear neutrophils and monocytes, thus assisting to collect these cells into the inflammation site. Molecules TNF- α stimulates bone resorption by inducing the proliferation and differentiation of osteoclasts progenitors and indirectly activate osteoclast formation [12].

CONCLUSION

The results of this study concludes that the levels of TNF- α in GCF of *Osing* women suffering from occlusal disharmony is higher than that of women who does not suffer from occlusal disharmony. Occlusal disharmony causes the occlusal trauma that is a degenerative injury that occurs when the occlusal loads exceed the adaptive capacity of the periodontal tissues, thereby triggers TNF- α secresion. The presence of increased levels of TNF- α is one indicator of the occurrence of periodontal tissue inflammation.

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