Assessment the Frequency of HLA-DRB1, Allele and Salivary Level of Receptor Activator Nuclear Factor KB Ligand (RANKL) / Osteoprotegerin (OPG) in Chronic Periodontitis

A Thesis

Submitted to the council of College of Dentistry / University of Baghdad in partial fulfillment for the requirement for the degree of Master of Science in Oral Microbiology

By

Sara Mohammed Mohssen

B.Sc. Biology (2008-2009)

Supervised by:

Assist. Prof. Dr. Batool Hassan Al-Ghurabi

PhD. in Medical Microbiology / Clinical Immunology

Baghdad - Iraq

2013 A.D. **1434 A.H.**

Abstract

Background: Periodontal diseases are initiated by microbial plaque, which accumulates in the sulcular region and induces an inflammatory response. This response may progress in certain susceptible individuals to chronic destructive inflammatory condition termed periodontitis. There is increasing evidence that host susceptibility plays an important role in initiation and progression of periodontitis. The Receptor activator of nuclear factor-kappa B ligand/ osteoprotegerin (RANKL/OPG) axis is involved in the regulation of bone metabolism in periodontitis, in which an increase in receptor activator of nuclear factor-kappa B ligand or a decrease in osteoprotegerin can tip the balance in favor of osteoclastogenesis and the resorption of alveolar bone that is the hallmark of periodontitis.

Aims of Study: This study was performed to investigate the association of human leukocyte antigens class II genetypes (HLA-DR) and the susceptibility to chronic periodontitis in Iraqi patients, as well as to study the role of salivary levels of RANKL and OPG in pathogenesis of chronic periodontitis.

Subjects and Methods: Fifty five subjects with chronic periodontitis (25smokers and 30 non-smokers) with ages range from 24-64 years and 25 apparently healthy volunteers their ages and sexes were matched with the patients were participated in this study. Periodontal parameters used in this study were plaque index, gingival index, probing pocket depth, clinical attachment level and bleeding on probing. Saliva samples were collected from all patients and controls, whereas blood was collected from thirty patients only and twenty controls. DNA was extracted from blood samples, and then HLA-RD genotyping was performed by polymerase chain reaction-sequence specific oligonucleotide probes (PCR-SSO). While enzyme-linked immunosorbent assay

was carried out for estimation the salivary level of RANKL and OPG in studied groups.

Results: The present data revealed that the frequencies of HLA-DRB1*03 and HLA-DRB1*11 alleles were significantly higher in patients than in healthy controls (P= 0.004 and P=0.006 respectively), on the other hand low frequency of HLA-DRB1*04 allele was found in patients when compared with healthy control (P= 0.020). Furthermore; this study showed that median salivary levels of RANKL was elevated in patient as compared with control group (P<0.001), whereas the salivary levels of OPG doesn't show any significant differences between the study groups (p>0.05). In contrast the ratio of RANKL/OPG ratio was significantly higher in patients when compared with the ratio in control group. Interestingly negative significant correlation was noticed between RANKL and OPG.

Regarding correlation between salivary (RANKL and OPG) and clinical periodontal parameters, RANKL levels was showed significant positive correlation with each of probing pocket depth and clinical attachment level. Otherwise no association between OPG levels and clinical parameters of periodontitis was found. Moreover; the ratio RANKL/OPG was showed significant positive correlation with each of gingival index, probing pocket depth and clinical attachment level. Finally the current results indicated that smoking had no effect on the salivary levels of RANKL and OPG.

Conclusion: This study demonstrates that HLA-DRB1*03 and HLA-DRB1*11 alleles may contribute to the increased susceptibility to chronic periodontitis. In addition salivary levels of RANKL and OPG play a crucial role in pathogenesis of periodontitis, and the relative RANKL/ OPG ratio appears to be indicative of disease occurrence.