# The saliva immunology mechanisms and periodontal status in HIV infected subjects

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## Abstract

**Purpose:** The aim of this study was the evaluation of connection between parodontium determined by using GI and PBI indexes and specific immunity status and non-specific in HIV infected group and in control group.

Material and methods: The study was carried out in the group of 37 patients infected with HIV. Mixed non-stimulated saliva was used for the study. Peroxidase activity was determined using the method by Mansson-Rahemtull. Lysozyme and A, G, M antibodies concentrations were determined with the use of radial immunodiffusion method. The concentration of lactoferrin was determined by using ELISA method. The clinical state of parodontium estimated by means of GI and PBI evaluating quality changes in the gum.

**Results:** Deterioration of the immunological status of subjects was accompanied by the increase of the values of GI and PBI. The strong negative correlation between GI and PBI and the concentration of lactoferrin and positive activity of the peroxidase in the whole examined population was determined. In the infected group the correlation between the status of gingiva expressed by GI and concentration or activity of examined enzymes and immunoglobulins was not ascertained.

**Conclusions:** 

1. HIV infection is connected to worsening of paradontium status expressed by values of GI and PBI indexes.

2. Paradontium status correlated positively with immunological status of HIV positive subjects.

3. In HIV infected group, no connection between number of IgA, IgG, IgM, concentration of lysozyme, lacto-

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ferrin, activity of peroxidase and paradontium status was observed.

Key words: HIV infection, GI, PBI, lysozyme, lactoferrin, peroxidase, IgA, IgG, IgM.

## Introduction

HIV infection is a very important risk factor of periodontal disease manifestation [1]. It causes the disturbance of the immunological system, which is progressive deterioration of lymphocytes T CD4, leading to AIDS development. The research indicates, that one in ten infected persons is not aware of HIV infection. Changes within the oral cavity are often first symptoms of the HIV infection. The World Health Organization divided disease processes of the oral cavity in HIV infection into three groups [2-4]. The necrotizing ulcerative gingivitis and periodontitis are assigned to changes strongly bounded with HIV.

## Material and methods

The study was carried out in the group of 37 patients (11 women and 26 men, aged 19-65, mean age 32 years) infected with HIV, hospitalized in the Teaching Hospital of Infectious Diseases, the Medical University of Białystok. Patients were divided into 3 groups, according to immune disturbances, and a division criterion was laboratory tests concerning the number of CD4 helper lymphocytes T in peripheral blood. The control group comprised of non-infected individuals, counterpart of the examined group. The patients were informed about the aim of the study and they gave their consent. The study was carried out after the approval of the Bioethic Committee of the Medical University of Białystok.

Mixed non-stimulated saliva, collected using expectoration method in the amount of 3-5 ml 2 hours after meal, was used for

Examined groups	Number of examined subjects	Gingival Index	Standard deviation ±SD	PBI index	Standard deviation ±SD
Infected (1)	37	1.66	0.86	1.01	0.96
Non-infected (0)	37	0.89	0.73	0.68	0.65
Statistical Analysis ( $p < 0.05$ )	0v1 -				

### Table 1. GI and PBI indexes' values in examined groups

Table 2. Values of GI and PBI indexes and the time of infected subjects duration

Examined groups	Number of examined subjects	Gingival Indexs	Standard deviation ±SD	PBI index	Standard deviation ±SD	
Infected <4 years (1)	24	1.69	0.78	1.12	1.06	
Infected ≥4 years (2)	13	1.61	1.03	0.81	0.73	
Total (3)	37	1.66	0.86	1.01	0.96	
Control group (0)	37	0.89	0.73	0.68	0.65	
Statistical Analysis (p<0.05)	0v1 and 0v2 -					

Table 3. Values of GI and PBI indexes depending on immunological status of examined subjects

Examined groups	Number of examined subjects	Gingival Index	Standard deviation ±SD	PBI index	Standard deviation ±SD
CD4<200/µl (1)	8	2.02	0.77	1.36	0.92
CD4 200-499/µl (2)	19	1.63	0.87	1.2	1.06
CD4>500/µl (3)	10	1.45	0.9	0.39	0.45
Control group (0)	37	0.89	0.73	0.68	0.65
Statistical Analysis (p<0.05)		1v0 and 2v0		1v3	

the study. Saliva samples were centrifuged, divided into portions 200 µl each, and stored at -80°C.

Peroxidase activity was determined using the method by Mansson-Rahemtull et al. [5]. Lysozyme and A, G, M antibodies concentrations were determined with the use of radial immunodiffusion method, ready-made kits (Human NL Nanorid plate - The Binding Site Ltd., UK, BINARID - The Binding Site Ltd.). The concentration of lactoferrin was determined by using ELISA method (Bioxytech Lactof EIA - Oxis Health Products, Inc. USA). The clinical state of parodontium estimated by means of Gingival Index (GI) according to Löe and Silness and Papilla Bleeding Index (PBI) evaluating quality changes in the gum.

The statistical analysis concerning the differences of examined parameters was performed using the analysis of variances (for the variables of normal distribution) or Kruskal-Wallis test (for other variables). In case of significant difference between groups, post hoc analyses were conducted to compare all pairs of groups using t Student test with Bonferroni alteration/ amendment (for variables of normal distribution) or Dwass-Steele-Critchler-Flieger test (for other variables).

## Results

In HIV infected subjects, GI index was higher than in control group and corresponded to moderate gingivitis, with gingival erythema, tumour and haemmorrhage. In non infected group, the index value indicated subinflammation with mild gingival discoloration. The difference of GI values in those groups was statistically significant. PBI index reached higher value in

HIV infected persons than in control group (without statistically significant difference) (Tab. 1).

The value GI index did not change fundamentally along with the extension of infection duration. The visible mild tendency of PBI index value decrease with a duration of the HIV infection was not statistically significant (Tab. 2).

The value of the GI index indicated a tendency of increase along with the worsening of the immunity status of infected subjects. Its value indicated gingival subinflammation in examined subjects with the absolute number of lymphocytes CD4 >500/µl increased to moderate gingival inflammation in the group with the number of lymphocytes below 200/µl. Statistically significant differences appeared between the control group and examined infected subjects with the absolute number of lymphocytes CD4 below 499/µl. Data introduced in the table III indicate increase of PBI index value along with the worsening of the immunological status of examined subjects, however, statistically significant differences were ascertained only between groups with highest and lowest number of lymphocytes T CD4 (Tab. 3).

In persons, who were treated with HAART, the non-significantly higher value of the GI index in infected group, which was not treated with this method and significantly higher in control group was observed. The HAART therapy did not statistically influenced the change of the PBI index value. Only a little higher value in the infected subjects who underwent therapy was observed (Tab. 4).

GI values correlated statistically significantly with activity of peroxidase and concentration of lactoferrin in whole examined population. A strong negative correlation between GI and lactoferrin concentration and positive correlation with peroxidase

Examined groups	Number of examined subjects	Gingival Index	Standard deviation ±SD	PBI index	Standard deviation ±SD
Infected treated subjects (1)	28	1.82	0.75	1.06	0.99
Infected non-treated subjects (2)	9	1.17	1.04	0.86	0.90
Control group (0)	37	0.86	0.73	0.68	0.65
Statistical Analysis (p<0.05)		1v0		NS	

activity was observed. In the infected group the correlation between the gingival status expressed by Gingival Index and the concentration or activity of evaluated enzymes was not observed. In non-infected persons, the clinical manifestation of the paradontium correlated statistically positively significantly with the activity of the peroxidase and negatively with the level IgM.

Values of PBI index received in entire population correlated negatively statistically significantly only with lactoferrin concentration and positively with peroxidase activity.

## Discussion

The dental research essentially influence the early diagnosis of symptoms of the HIV infection or worsening of the immunological status of the infected person [7]. It has significant meaning especially in developing countries where an access to laboratory-research is limited [8]. Clinical symptoms of the immunological status worsening can have the significant prognostic meaning.

The specific immunodeficiency as result of HIV infection conjointly with changes in the non-specific immunity finds reflection in paradontium tissues and manifests itself with the high average value of GI and PBI in HIV infected persons in comparison to non-infected. As the infection's duration time proceeded values of both indexes decreased. These observations are confirmed by Yeung et al. research [9] according to infected subjects and Friedman et al. [10] research conducted within the group of drug addicts. Different results were obtained by Tukutuku et al. [11], because the gingival index in examined HIV infected population carried out average 0.42 and it was four times lower than in our research. Simultaneously, this result was better than obtained from our control group. There is almost no information about the connection between the immunological status of examined subjects and the paradontium status. In the accessible literature, this problem was examined by Barr et al. [12]. Their results prove the low significant influence of the immunity status on progress of changes in the paradontium. Our research confirmed these dependences and showed that together with worsening of the immunological status of examined subjects the values of GI and PBI increased. Nittayananta et al. research [13] indicate, that decrease of the absolute number of CD4 lymphocytes T below 200/µl is a significant risk factor of the pathological changes appearance within the entire oral cavity. In our research, we observed a correlation between the value of GI and PBI indexes and lactoferrin concentration and the peroxidase activity in the entire examined population. In the HIV infected group, the differences were not statistically significant. In non-infected persons, GI correlated statistically significantly positively with the activity

of the peroxidase and negatively with the IgM level. This relation was not confirmed in HIV infected subjects. Jentsch et al. [14] evaluated the influence of the lactoferrin concentration and activity of lysozyme and peroxidase on the healing process of the paradontium after the surgical treatment in the examined non-infected group. Their results indicate that only the lactoferrin concentration can be an appropriate marker to monitor the paradontium status, what is also confirmed by Fine et al. results [15].

## Conclusions

1. HIV infection is connected to worsening of paradontium status expressed by values of GI and PBI indexes.

2. Paradontium status correlated positively with immunological status of HIV positive subjects.

3. In HIV infected group, no connection between number of IgA, IgG, IgM, concentration of lysozyme, lactoferrin, activity of peroxidase and paradontium status was observed.

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