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# PROMENE PLJUVAČKE KOD OSOBA SA BULOZNIM LIHEN PLANUSOM

# SALIVARY HUMORAL CHANGES IN ORAL BULLOUS LICHEN PLANUS

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**Sažetak**

***Cilj:*** *Cilj istraživanja bio je ispitivanje humoralnih mehanizama u pljuvački kod pacijenata sa buloznim lihen planusom u fazi egzacerbacije i remisije.*
***Materijal i metod:*** *Praćeno je devetnaest pacijenata sa oralnim buloznim lihen planusom. Pljuvačka je prikupljana u jutarnjim časovima, bez stimulacije, u količini 5-10 cm$^3$. Određivanje imunoglobulina, $C_3$ i $C_4$ u pljuvački vršilo se primenom tehnike „microelis". Cirkulišući imuni kompleksi u serumu i pljuvački određivani su PEG metodom. Dobijeni rezultati upoređivani su sa kontrolnom grupom i međusobno u fazi egzacerbacije i remisije. Rezultati su statistički obrađeni Studentovim „t" - testom.*
***Rezultati:*** *Ispitivanja su pokazala da je količina IgA u pljuvački značajno smanjena , dok je količina IgG i IgM značajno uvećana. Vrednosti imunoglobulina A , G i M u fazi remisije značajno su povećane kod bolesnika u odnosu na kontrolnu grupu. Upoređivanjem pljuvačke u fazi egzacerbacije i remisije dobile su iste količine IgG u oba stadijumima bolesti , ali evidentno povišene vrednosti IgA i malo značajno povećanje vrednosti IgM. U kontrolnoj grupi i grupi pacijenata u fazi egzacerbacije i remisije takođe su povećane CIC vrednosti . Podaci ukazuju na visoko značajnu depresiju komponente C3 u fazi egzacerbacije, dok je u remisiji vidljivo malo značajno smanjenje. Komplement komponente C4 u fazi remisije opada u poređenju sa kontrolnom grupom.*
***Zaključak:*** *Postoji evidentno učešće pojedinih komponenti pljuvačke u patogenezi oralnog lihen planusa, ali konačna pozicija za prevlast humoralnih mehanizama u patogenezi buloznog lihen planusa još uvek ne postoji.*

***Ključne riječi:*** bulozni oralni lihen planus, pljuvačka, imunoglobulini, komplement

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**Abstract**

***Background****: To investigate the salivary humoral mechanisms in patients with bullous lichen planus, in the phases of exacerbation and remission.*
***Material and Method****: Nineteen patients with bullous oral lichen planus were followed. Saliva was collected in the morning without stimulation in the amount from 5-10 cm$^2$. The determination of immunoglobulins, $C_3$ and $C_4$ in the saliva was performed using the micro-ELISA technique. The circulating immune complexes in the serum and saliva were determined by the PEG method. The results were compared with a control group and within the group in the phases of exacerbation and remission. The results were statistically processed by Student's t - test .*
***Results:*** *Salivary IgA showed a significant decrease, whereas IgG and IgM significantly increased. The values of immunoglobulin A, G and M in the phase of remission were significantly increased compared with the control group.*
*By comparing salivary immunoglobulins in the phases of exacerbation and remission, identical amounts of IgG in both stages of the disease were observed. However, considerably elevated values of IgA and slightly significant increase in the values of IgM were noted. CIC values in the control group and in patients' group in the phases of exacerbation and remission were increased. Data suggest a highly significant depression of $C_3$ component in the exacerbation phase, while in remission a slightly significant decrease was evident. Complement component $C_4$ in the remission phase compared with a control group was declined.*
***Conclusion:*** *There is an evident involvement of certain saliva components in the pathogenesis of oral lichen planus, but the final predominance of humoral mechanisms in the pathogenesis of bullous lichen planus does not exist yet.*

***Key words:*** bullous oral lichen planus, salivary immunoglobulins, complement.

## *Uvod*

Oralni lihen planus (OLP) je hronična inflamatorna bolest, koja se javlja uglavnom u srednjem dobu kod žena[1,2]. Njegova etiologija je nepoznata, sa mogućom multifaktorijalnom genezom[1], u okviru koje posebno treba istaći stres, genetsku predispoziciju, šećernu bolest i gastrointestinalne poremećaje [3]. Kliničke i histološke studije ističu ulogu nekih dentalnih restaurativnih materijala, kao što su amalgam[4], kompozit[5] i stomatološki akrilat[6], kao uzročnike ove bolesti, koja vrlo često može imati i malignu transformaciju. Ozbiljnost bolesti i njena nepoznata priroda zahtevaju konstantno ispitivanje etiopatogenetskih zbivanja u toku ove bolesti.

Međutim, u poslednjih nekoliko godina, postaje jasno da imuni sistem ima primarnu ulogu u nastanku i evoluciji oralnog lihen planusa. U tom pogledu , smatra se da nivoi imunoglobulina u pljuvački mogu igrati ulogu u patogenezi oralnih mukoznih bolesti, uključujući lihen planus, ili kliničkih promena koje mogu uticati na bolest u okviru koje su prisutne[7]. U vezi sa ovim mogućnostima , pronađeno je povećanje nivoa seruma imunoglobulina IgA i IgG kod bolesnika sa OLP [8,9].

Ghalayani[10] je pokazao značajne razlike u distribuciji IgG + ćelija na različitim topografskim mestima oralnog lihen planusa ili lihenoidnih lezija, ali se razlike između raspodele IgG + ćelija između oralnog lihen planusa i lihenoidnih lezija nisu pokazale kao značajne. Iako je Sistig[7] pokazao povećanje nivoa salivarnih IgA i IgG kod ovih pacijenata, der Vaal[11] ne dokazuje ovu pretpostavku.

Imajući u vidu brojne kontroverzne i nedosledne stavove, krenuli smo u ispitivanje humoralnih mehanizama u pljuvački kod pacijenata sa buloznim lihen planusom u fazi egzacerbacije i remisije, nadajući se da će se razjasniti složeni etiopatogenetski mehanizmi ovog veoma čestog oboljenja.

## *Introduction*

Oral lichen planus (OLP) is a chronic inflammatory disease, which mostly occurs in the middle - aged women[1,2]. Its etiology is unknown with possible multifactorial genesis[1], such as stress, genetic predisposition, diabetes mellitus, or gastrointestinal disorders[3]. Clinical and histological studies highlight the role of some dental restorative materials such as amalgams[4], composites[5], and dentalacrylate[6] as the possible causes of this disease which very often has possible malignant transformation. The severity of the disease and the unknown nature require a constant study of etiopathogenetic activities during the course of the disease.

However, in recent years, it has become apparent that the immune system has a primary role in the emergence and evolution of oral lichen planus. In this regard, it is believed that salivary immunoglobulin levels may play a role in the pathogenesis of oral mucosal diseases, including lichen planus, or clinical changes which can affect the very disease[7]. In connection with this possibility, increased levels of serum immunoglobulins IgA and IgG were found in patients with OLP[8,9].

Ghalayani[10] showed significant differences in the distribution of IgG + cells in different locations of oral lichen planus or lichenoid lesions separately, but the differences in the distribution of IgG + cells between oral lichen planus and lichenoid lesions did not prove as significant. Although Sistig[7] proved the increased levels of salivary IgA and IgG in these patients, der Waal[11] did not prove this assumption.

Considering the numerous inconsistent and controversial views, we set out to examine the salivary humoral mechanisms in patients with bullous lichen planus in the phases of exacerbation and remission, hoping to participate in clarifying the complex etiopathogenetic mechanisms of this very common disease.

## *Materijal i metode*

Za realizaciju ovog cilja, na Klinici za oralnu patologiju i parodontologiju, na Stomatološkom fakultetu u Skoplju, izabrano je 19 bolesnika različitog pola i uzrasta. Dijagnostikovan je oralni bulozni lihen planus, bez obzira na topografsku lokalizaciju promena.

Studija nije uključivala pacijente sa kožnim manifestacijama i sa kožnim i oralnim promenama. Dijagnoza je postavljena na osnovu temeljne anamneze i objektivnih kliničkih nalaza.

Od svakog pacijenta uziman je uzorak pljuvačke. Pljuvačka je sakupljana ujutru jednostavnim dotokom bez stimulacije u količini od 5 do 10 $cm^3$. Zatim je materijal, najkasnije dva sata od sakupljanja, distribuiran na Institut za transfuziju, gde je zamrznut i kasnije dalje obrađivan.

Svi pacijenti su ispitivani na prisustvo imunoglobulina A, G i M, CIC i komponente komplemenata $C_3$ i $C_4$ u pljuvački. Testovi su sprovođeni dva puta, u fazi egzacerbacije i u toku remisije.Određivanje imunoglobulina iz pljuvačke je sprovedeno pomoću tehnike „microelis".

Određivanje komponenti komplemenata $C_3$ i $C_4$ u pljuvački vrši sa istim postupkom kao i određivanje imunoglobulina, osim što partigen ploče imaju posebna antitela za ove komponente. Normalne vrednosti za $C_3$ su od 0,80 do 1,40 g / l , a za $C_4$ od 0,2 do 0,5 g / l u serumu. Određivanje CIC u serumu i mešovitoj pljuvački izvedeno je po metodi PEG (polietilenski glikol). Dobijeni rezultati su upoređivani sa kontrolnom grupom i međusobno između pacijenata u fazi egzacerbacije i fazi remisije.

Kontrolna grupa se sastojala od 25 zdravih pojedinaca koji ne boluju od lihen planusa, niti bilo koje druge bolesti. Rezultati za vrednosti imunoglobulina A, G, M, CIC i $C_3$, $C_4$ u pljuvački upoređivani su sa kontrolnom grupom.

Svi rezultati dobijeni iz ovog istraživanja upoređeni su sa kontrolnom grupom i međusobno u fazi egzacerbacije i remisije bolesti.

Rezultati su statistički obrađeni primenom Studentovog t-testa radi određivanja značaja razlike u vrednostima.

## *Material and Methods*

To realize this goal, at the Clinic for Oral Pathology and Periodontology, the Faculty of Dentistry in Skopje, 19 patients of different sex and age were followed, diagnozed with oral bullous lichen planus, regardless of topographic distribution of changes.

The study did not include patients with dermal manifestations, and with skin and oral presentation. Diagnosis was made based on a detailed history taking and objective clinical findings. A sample of saliva was taken from each patient.

Saliva was collected in the morning by a simple overflow without stimulation in the amount from 5 to 10 $cm^3$. Then, no later than two hours of collecting, the material was transferred to the Institute of Transfusion where it was frozen and further processed.

All patients were assessed for immunoglobulin A, G and M, CIC, and complement components $C_3$ and $C_4$. The tests were conducted twice in the phases of exacerbation and remission. The determination of immunoglobulins in saliva was performed using micro-ELISA technique.

The determination of $C_3$ and $C_4$ complement components in saliva was done in the same way as determination of immunoglobulins, except that the partigen plates have specific antibodies to these components. Normal values for $C_3$ range from 0.80 to 1.40 g / l and for $C_4$ from 0.2 to 0.5 g / l in serum. CIC determination in serum and mixed saliva was performed by the method of PEG (polyetilen glycol).The results were compared with a control group and with each other in phases of exacerbation and remission.

The control group comprised 25 healthy individuals, who did do not suffer from lichen planus or any other disease. The results for the values of immunoglobulin A, G, M, CIC and $C_3$, $C_4$ in the saliva were compared with the control group.

All results obtained from comparative trials were compared with: the control group and each other in the phases of exacerbation and remission of the disease.

The results were statistically processed by applying the Student's t-test for the significance of differences in values.

## *Rezultati*

Dobijeni rezultati istraživanja u ovoj studiji prikazani su u tabelama 1-5.

Salivarne vrednosti za sve analizirane imunoglobuline ( IgA, IgG i IgM ) u ispitivanoj grupi i u kontrolnoj grupi ukazuju na statističku značajnost, gde je vrednost IgA pokazala značajno smanjenje, dok su vrednosti Ig G i IgM značajno povećane.

Vrednosti imunoglobulina A , G i M pljuvačke kod bolesnika sa buloznim lihen planusom u fazi remisije značajno su povećane u poređenju sa kontrolnom grupom ( $p<0,001$).

Komparativna analiza imunoglobulina pljuvačke u fazi egzacerbacije i remisije pokazuje istu količinu IgG u obe faze bolesti, ali evidentno povišene vrednosti IgA ($p<0,001$) i statistički značajno malo izražene uvećane vrednosti IgM ( $p<0,05$).

Salivarne vrednosti koncentracije CIC u kontrolnoj grupi pacijenata i pacijenata u fazi egzacerbacije i remisije idu u prilog povećanj u svih ispitivanih vrednosti ( $p<0,001$).

Podaci ukazuju na veoma izraženu značajnu depresiju $C_3$ komponente ($p<0,001$) u fazi egzacerbacije, dok je u fazi remisije evidentno malo značajno smanjenje ($p<0,05$).

Komplement komponente $C_3$ kod bulozne forme u fazi remisije u poređenju sa kontrolnom grupom opada ($p<0,001$). U fazi remisije, komplement komponente $C_4$ u pljuvački pokazuje neznatno smanjenje ($p<0,2$).

## *Results*

The results obtained in this study are shown in tables 1-5.

Salivary values for all analyzed immunoglobulins ( IgA, IgG and IgM ) in the examined and control group indicate statistical significance, whereas the value of IgA showed a significant decrease, and Ig G and Ig M significantly increased.

The values of salivary immunoglobulin A, G and M in patients with bullous lichen planus in the phase of remission are significantly increased compared with the control group ( $p<0.001$ ).

Comparative analysis of salivary immunoglobulins in the phase of exacerbation and remission points to identical amount of IgG in both stages of the disease, but an evidently increased value of IgA ( $p<0.001$) and statistically significant low elevated values of IgM ( $p<0.05$).

Salivary concentration values of the CIC in the control group of patients and patients in the phase of exacerbation and remission confirm the increase of all examined values ( $p<0.001$).

The data suggest a highly expressed significant depression of $C_3$ component ($p<0.001$) in the exacerbation phase, while in the stage of remission a low significant decrease is evident ( $p<0.05$ ).

Complement component $C_4$ in the bullous form in the phase of remission compared with a control group is declining ($p<0.001$). In the remission phase, complement component $C_4$ in saliva shows insignificant reduction ( $p<0.2$ ).

**Tabela 1**.Salivarne vrednosti imunoglobulina A, G i M u kontrolnoj i ispitivanoj grupi u fazi egzacerbacije
**Table 1.** Salivary values of immunoglobulin A, G and M in control and examined group in the phase of exacerbation

| Kontrolna grupa / Control group n = 25 | | | Ispitivana grupa n=19 (faza egzacerbacije) Examined group n=19 (phase of exacerbation) | | |
|---|---|---|---|---|---|
| gr/l | IgA | IgG | IgM | IgA | IgG | IgM |
| x | 1.10 | 0.02 | 0.11 | 0.71 | 0.08 | 0.29 |
| SD | 0.01 | 0.002 | 0.08 | 0.031 | 0.04 | 0.07 |
| t | | | | 6.14 | 7.31 | 7.62 |
| p | | | | < 0.001 | < 0.001 | < 0.001 |
| | | | | *** | *** | *** |

**Tabela 2.** Salivarne vrednosti imunoglobulina A, G i M u kontrolnoj i ispitivanoj grupi u fazi remisije
**Table 2.** Values of salivary immunoglobulin A, G and M in the control and examined group in the phase of remission

| Kontrolna grupa / Control group n = 25 | | | Ispitivana grupa n = 19 (faza egzacerbacije) Examined group n = 19 (phase of exacerbation) | | |
|---|---|---|---|---|---|
| gr/l | IgA | IgG | IgM | IgA | IgG | IgM |
| x | 1.10 | 0.02 | 0.11 | 0.71 | 0.08 | 0.29 |
| SD | 0.01 | 0.002 | 0.08 | 0.31 | 0.04 | 0.07 |
| t | | | | 6.14 | 7.31 | 7.62 |
| p | | | | < 0.001 | < 0.001 | < 0.001 |
| | | | | *** | *** | *** |

| Kontrolna grupa / Control group n = 25 | | | Ispitivana grupa n = 19 (faza egzacerbacije) Examined group n = 19 (phase of exacerbation) | | |
|---|---|---|---|---|---|
| gr/l | IgA | IgG | IgM | IgA | IgG | IgM |
| x | 1.10 | 0.02 | 0.11 | 0.99 | 0.06 | 0.29 |
| SD | 0.01 | 0.002 | 0.08 | 0.17 | 0.02 | 0.03 |
| t | | | | 6.14 | 7.31 | 7.62 |
| p | | | | **< 0.001** | **< 0.001** | **< 0.001** |
| | | | | *** | *** | *** |

**Tabela 3.** Salivarne vrednosti imunoglobulina A, G i M u ispitivanoj grupi u fazi egzacerbacije i remisije
**Table 3.** Values of salivary immunoglobulin A, G and M in the examined group in the phase of exacerbation and remission

| | Ispitivana grupa (faza egzacerbacije) Examined group (phase of exacerbation) n = 19 | | | Ispitivana grupa (faza remisije) Examined group (phase of remission) | | |
|---|---|---|---|---|---|---|
| gr/l | IgA | IgG | IgM | IgA | IgG | IgM |
| x | 0.71 | 0.08 | 0.29 | 2.42 | 0.08 | 0.33 |
| SD | 0.31 | 0.04 | 0.07 | 0.09 | 0.02 | 0.02 |
| Se | 0.07 | 0.009 | 0.01 | 0.02 | 0.004 | 0.00 |
| t | | | | 22.47 | 0.00 | 2.33 |
| p | | | | **< 0.001** | 0 | < 0.05 |
| | | | | *** | 0 | ** |

**Tabela 4.**Vrednost koncentracije CIC pljuvačke u kontrolnoj i ispitivanoj grupi u fazi egzacerbacije i remisije
**Table 4.** Values of salivary immunoglobulin A, G and M in the control and examined group in the phase of exacerbation and remission

| | Kontrolna grupa Control group n=25 | Ispitivana grupa Examined group n = 19 | |
|---|---|---|---|
| mgr / % | egzacerbacija exacerbation | remisija remission | |
| **x** | 0.02 | 0.09 | 0.04 |
| SD | 0.01 | 0.03 | 0.002 |
| t | | 10,64 | 4.23 |
| p | | < 0.001 | < 0.001 |
| | | *** | *** |

**Tabela 5.** Salivarne vrednosti koncentracija komplemenata $C_3$ i $C_4$ pljuvačke u kontrolnoj i ispitivanoj grupi u fazi egzacerbacije i remisije
**Table 5.** Values of salivary concentration of $C_3$ and $C_4$ complement components in the control and examined group in the phase of exacerbation and remission

| Kontrolna grupa / Control group N = 19 n = 25 | | | Ispitivan grupa / Examined group (remisija) / (egzacerbacija) (remission) / (exacerbation) | | | |
|---|---|---|---|---|---|---|
| gr/l | $C_3$ | $C_4$ | $C_3$ | $C_4$ | $C_3$ | $C_4$ |
| **x** | 0.76 | 0.13 | 0.28 | 0.09 | 0.57 | 0.12 |
| SD | 0.23 | 0.04 | 0.09 | 0.02 | 0.19 | 0.02 |
| t | | | 8.41 | 3.90 | 2.35 | 1.00 |
| p | | | < 0.001 | < 0.001 | < 0.05 | < 0.2 |
| | | | *** | *** | • | o |

## *Diskusija*

Iako su ranije studije ukazivale na ozbiljno učešće ćelijskog imuniteta, one ne daju definitivan i ubedljiv način njihovog delovanja u patogenezi i razvoju oralnog lihen planusa[4,8].

S druge strane , studije o učešću humoralnih imunoglobulina i serumskih komplemenata dovele su do nejednakosti i suprotnosti u istraživanjima[7,11]. Zbog ovih suprotnosti smatra se da nema adekvatnih informacija da li je pojava lihen planusa pod uticajem promena u humoralnom ili ćelijskom imunom odgovoru. Znajući poreklo i svojstva imunoglobulina pljuvačke, prirodno je očekivati promenu njihove koncentracije kod različitih bolesti usne duplje, uključujući i lihen planus.

Rezultati ovog istraživanja pokazuju niske nivoe IgA u kontrolnoj grupi. Imunoglobulini G i M se uvećavaju u fazi egzacerbacije i upoređujući ih sa kontrolnom grupom, pokazuju statistički značajnu razliku ($p<0,001$). Sistig[7] dolazi do identičnih zaključaka. Za više oralnih mukoznih oboljenja, uključujući oralni lihen planus , dobio je povećanje pojedinih potklasa imunoglobulina G, $IgG_1$, $IgG_2$, $IgG_3$ i $IgG_4$ u fazi egzacerbacije. U fazi remisije, vrednosti

## *Discussion*

Although previous in vitro studies indicated a serious participation of cellular immunity, they do not provide a definitive and convincing description of its activity in the course and pathogenesis of oral lichen planus [4,8].

On the other hand, studies about the participation of humoral immunoglobulins, and serum complement resulted in inequality and contradiction[7,11]. Due to these inconsistencies, it is considered that there is no adequate information whether lichen planus is influenced by changes in the humoral or cellular immune response. Knowing the origin and properties of salivary immunoglobulins, it is natural to expect a change in their concentration in various diseases that attack oral cavity, including lichen planus.

The results of our investigation registered low levels of IgA in the control group. Immunoglobulin G and IgM in the phase of exacerbation are increased, and compared with the control group, they show a highly statistically significant difference ($p<0.001$). Sistig [7] came to identical findings. For several oral mucosal diseases, including oral lichen planus, he recorded the growth of certain subclasses of immuno-globulin G, $IgG_1$, $IgG_2$, $IgG_3$ and

$IgG_1$ i $IgG_4$ su se vratile u normalu, a $IgG_2$ ostale povećane, što se delimično poklapa sa nalazima ovog istraživanja.

Eksperimentalno i teoretski je pokazano da je imunoglobulin A veliki protein čija je glavna funkcija zaštitna, direktno povezana sa odbrambenim mehanizmom oralne sluzokože. Smanjene vrednosti ovog imunoglobulina su objašnjene smanjenim otporom oralne sluzokože usled brojnih egzogenih ili endogenih faktora, dovodeći do pojave određenih patoloških stanja, manifestovanih kao klasične, potpuno benigne promene , sve do erozivno - ulceroznih lezija , sa brojnim dramatičnim kliničkim manifestacijama , čak i sa mogućnošću maligne alteracije . Ispitujući pljuvačne biomarkere, Lopez-Jornet[12] sugerise na moguću funkciju oksidativnog stresa koji izaziva varijacije u humoralnoj pljuvački koje utiču na oralnu sluzokožu.

Parichehr Ghaleiani[13] se oslanja na nalaze imunoglobulina pljuvačke u smislu razjašnjenja patogeneze oralnog buloznog lihen planusa, ali tvrdi da koristeći direktnu imunofluorescenciju, može detektovati imunoglobuline i komplemente komponenti u uzorcima tkiva iz lezija oralnog lihen planusa, kao korisni parametar u dijagnostici oboljenja. Do sličnih otkrića došao je i Sano[14], koji naglašava činjenicu da se upotrebom direktne imunofluorescence razjašnjava fiziologija buloznih bolesti u koje se uključuje i bulozni OLP.

Generalno, rečeno je da se ovi imunoglobulini u fazi egzacerbacije proizvode u većim količinama, bilo preko veće količine seruma ili kao rezultat lokalne sinteze pljuvačnih žlezda. Međutim, kapacitet ovog imunoglobulina da aktivira komplement i da se veže za stimulator antigena, da napravi povezanost sa CIC u pljuvački, omogućavajući smanjenje vrednosti pljuvačke[7,13] i niske vrednosti komplementa $C_3$ pljuvačke koje su inkorporirane u ovom kompleksu, naša je interpretacija dobijenih rezultata tipičnih za ovu fazu bolesti.

Sposobnost imunoglobulina A je utome da poveća fagocitne aktivnosti makrofaga i indirektno deluje na bakteriolizu. IgA ostvaruje svoju funkciju u prisustvu komplementa i lizozima, tako da je logično i očekivano, naći niske vrednosti komplementa komponente $C_3$ u ovom istraživanju.

$IgG_4$ in the exacerbation phase. At the stage of remission, $IgG_1$ and $IgG_4$ returned to normal, $IgG_2$ remained increased, which partly coincides with our findings.

Experimentally and theoretically is demonstrated that immunoglobulin A is a major protein whose main function is protective, directly related to the defense mechanism of the oral mucosa. Discounted values of this immunoglobulin are explained with the reduced resistance of the oral mucosa to numerous exogenous or endogenous factors, leading to certain pathological conditions, manifested up to possible classic, utterly benign, till the erosive-ulcerous lesions, with many dramatic clinical features, even with the possibility of malignant alteration. Examining the salivary biomarkers, Lopez-Jornet[12] suggests a possible function of oxidative stress that causes humoral salivary variations that affect the oral mucosa.

Parichehr Ghaleyani[13] relies on salivary immunoglobulin findings in clarifying the pathogenesis of bullous oral lichen planus, but argues that using a direct immunofluorescence can detect immuno-globulins and complement components in tissue samples from lesions of oral lichen planus, as useful parameters in the diagnosis of the disease. Sano[14] obtained similar findings, which emphasizes the fact that its use helps to clarify the physiology of bullous diseases which include the bullous OLP.

Generally, it is said that this immunoglobulin in the exacerbation phase is produced in larger quantities, through spillover of serum, or as a result of its local synthesis of the salivary glands. However, the capacity of this immunoglobulin to activate the complement, to bind to the antigen stimulator, and to make the association with CIC in the saliva, enabling reduced salivary values [7,13] and low salivary values of $C_3$ which are incorporated in this complex, is in fact our interpretation of the obtained results, typical of this stage of the disease.

The ability of this immunoglobulin is to increase the phagocytic activity of macrophages and indirectly to act on bacteriolysis. IgA realizes its function in the presence of complement and lysozymes, so it is logical and expected to find the low levels of complement component $C_3$ in our research.

Dominantna komponenta u kliničkom ispoljavanju bulozne forme oralnog lihen planusa je prisutvo enantema i udružene eksudacije, u čijoj su osnovi vaskularna neravnoteža, prvenstveno naglašena filtracija kapilara i naglašena transudacija ali i proteini koji su poreklom iz seruma. Na osnovu ovih nalaza se može reći da je povećana koncentracija imunoglobulina A u pljuvački kod posmatranih grupa znak hemodinamskih varijacija.

U fazi remisije, jedna od karakteristika IgA je da pomaže proces fagocitoze, da se aktivira komplement kroz produkciju $C_3$b komponente, kada počinje aktivacija patološkog razvoja. Konstantna invazija antigenog materijala stimuliše kontinuiranu proizvodnju novih frakcija imunoglobulina , uključujući i IgG . Jedan deo njih, aktiviran preko patoloških procesa, posredovan preko sistema komplemenata, pravi vezu sa antigenom izgradnjom CIC.

S obzirom na povećanu biološku efikasnost i podobnost alternativnog puta i brzine kaskadnih procesa, počevši od $C_4$ komponente u odnosu na klasičan put, prirodno je očekivati njegovu veću vrednost u odnosu na imunoglobulin A, čije se vrednosti smanjuju u fazi egzacerbacije. Ako se doda povećana potreba organizma za odbranom, onda postoji opravdanje za objašnjenje dobijenih rezultata u ovom istraživanju u pogledu vrednosti IgG i dobijenog povećanja vrednosti u pljuvački u svim kliničkim oblicima u obe faze bolesti [15-17].

Prekomerna produkcija imuno-globulina M je direktno povezana sa akutnom fazom bolesti. Pre svega, uključena je u odbranu organizma na nivou sluzokože u ranoj fazi bolesti, tzv. fazi egzacerbacije. Uporna zapaljenska reakcija i sekundarna infekcija stimulišu lokalnu sintezu ovog imunoglobulina, što zapravo tumači rezultate I koji su dobijeni u ovom istraživanju. Imunoglobulin A je glavno antitelo u svim sekretima i pljuvački koji je u potpunosti odgovoran za sprovođenje mehanizama odbrane površine sluzokože. Iako u tom kontekstu prioritet pripada upravo ovom imunoglobulinu, ne treba zanemariti i druge nosioce humoralnog imuniteta, imuno-globuline M i G [18,19].

The dominant component in the clinical manifestation of the bullous form of oral lichen planus is the presence of erythema and associated exudation, in which basis are the vascular imbalance, primarily emphasized capillary filtration and marked transudation, not neglecting even those proteins that come from serum. Based on these findings, we believe that the increased concentration of immunoglobulin A in saliva in the studied group are due to the underlying hemodynamic variations.

In the remission phase, one of the many features of IgA is to assist phagocytosis, to activate the complement through $C_3$b component production, when the activation of the pathological development begins. The constant invasion of antigen material stimulates a continuous production of new immunoglobulin fractions, including IgG. One part of them, activated through the pathological processes, mediated by the complement system, makes a link with antigen, building CIC.

However, considering the increased biological effectiveness and suitability of alternative path and speed of a cascading process, starting with the $C_4$ component versus classical pathway, it is natural to expect a higher value compared with immunoglobulin A, whose values are reduced in the phase of exacerbation. If you add the growing need of the body for defense, then there is justification for our obtained results in terms of the value of IgG and received increased salivary values in all clinical forms in both stages of the disease[15-17].

Overproduction of immunoglobulin M is directly associated with the acute phase of the disease. It is primarily involved in defending the body at the level of the mucous membrane in the early stage of the disease, the so-called stage of exacerbation. Persistent inflammatory reaction and secondary infection stimulate local synthesis of this immunoglobulin, which actually explains the the results obtained in this study. Immunoglobulin A is the main antibody in all secretions and in saliva, which is fully responsible for the implementation of the defense mechanisms of mucosal surfaces. Although the priority is here given to this immunoglobulin, other carriers of humoral immunity, immunoglobulin M and G should not be neglected [18,19].

S obzirom na identične vrednosti imunoglobulina G u fazi remisije i egzacerbacije kod bulozne forme, pretpostavlja se da ovaj imunoglobulin nema presudnu funkciju u pljuvački kao imunoglobulin A . Evidentno povećanje imunoglobulina G u fazi remisije , u odnosu na kontrolnu grupu , i povećanje salivarnog imunoglobulina M, u poređenju sa kontrolom, najverovatnije je dodatni faktor u delovanju salivarnih imunoglobulina, sa ciljem odbrane organizma na nivou oralne sluzokože. Verujemo da frakcije ova dva imunoglobulina imaju dodatnu ulogu u odbrambenim mehanizmima IgA u toku imunogenih događaja na oralnoj sluzokoži.

Analiza CIC u fazi egzacerbacije i remisije ukazala je povećanu vrednost u odnosu na kontrolnu grupu, dok su vrednosti komplementa komponente $C_3$ smanjene kod svih pacijenata u obe faze bolesti. Tumačenje ovih nalaza je analogno prethodnim. Što se tiče vrednosti $C_4$ komponente u pljuvački u fazi egzacerbacije, one su niske. Pored toga, razlika između bulozne forme i kontrolne grupe je vrlo značajna ( $p<0,001$). U fazi remisije, komponenta $C_4$ kod pacijenata, u poređenju sa kontrolnom grupom, se smanjuje, tako da je statistički značajna.

Prema ulozi medijatora u odgovoru na sistem $C_4$ komplementa, logičan je i očekivani pad vrednosti u fazi egzacerbacije i u fazi remisije bolesti. Ovaj proteolitički enzim je sposoban da nastavi odvijanje reakcije, delujući direktno na komponentu $C_3$.

Pretpostavlja se da je ova komponenta instalirana u CIC i zbog toga njena vrednost opada u cirkulaciji, analogno komponenti $C_3$. Hronicitet oralnog lihen planusa i činjenica da je kod pacijenata u najvećem procentu prisutan do kraja svog života ide u prilog njegovoj stalnoj aktivnosti i funkciji.

Given identical values of immunoglobulin G in the phase of remission and exacerbation in the bullous form, we assume that this immunoglobulin has not a crucial function in the saliva as immunoglobulin A. Evident increase in the immunoglobulin G in the stage of remission, versus the control group, and increased salivary immunoglobulin M, compared with control, probably is an additional factor in the effectiveness of salivary immunoglobulin system, aimed at defending the body at the level of the oral mucosa. We believe that these two immunoglobulin fractions have accessory role in the defense mechanisms of IgA immunogenic events in the oral mucosa.

CIC analysis in the phases of exacerbation and remission indicated an elevated value compared to the control group, whereas the values of the complement component $C_3$ are reduced in all patients in both phases of the disease. The interpretation of these findings is analogous to the former. As for the values of $C_4$ component in the saliva in phase of exacerbation, they are low. In addition, the difference between the bullous form and the control group is highly significant ($p<0.001$). In the remission phase, component $C_4$ in patients, compared to the control group, is easily reduced, so it is statistically insignificant.

According to the role of mediators in the response to the $C_4$ complement system, a decrease in the value is logical and expected in the phase of exacerbation and in the remission phase of the disease. This proteolytic enzyme is able to continue the progress of the reaction, acting directly on the component $C_3$.

We assume that this component is installed in the CIC, and therefore its value declines in the circulation, analog to component $C_3$. Chronicity of oral lichen planus and the fact that lichen planus in the largest percentage of patients is present till the end of their life is something that tells about its constant activity and function.

## *Zaključak*

Definisan stav o dominaciji humoralnih mehanizama još uvek ne postoji . Naravno, moglo bi se reći da je evidentno učešće pojedinih komponenti pljuvačke u patogenezi oralnog lihen planusa.

## *Conclusion*

There is still no concensus on the dominance of the humoral mechanisms. It could be postulated that the involvement of certain components from the saliva in the pathogenesis of oral lihen planus is evident.

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