

# Quantity of Salivary Immunoglobulin A, Lysozyme and Magnesium in Patients with Burning Mouth Syndrome and Xerostomia

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## Summary

*Recent studies suggest a connection between burning mouth syndrome (BMS) and the quantity and quality of saliva. The aim of our study was to determine quantities of salivary immunoglobulin A (sIgA), lysozyme and magnesium in unstimulated and stimulated saliva of patients with burning mouth syndrome and xerostomia. Salivary samples were obtained by sialometry. Salivary immunoglobulin A was determined by radial immunodiffusion according to Manzini, lysozyme levels were obtained according to Osserman and Lowlor, and magnesium was determined using atomic absorbance spectrophotometry. Levels of sIgA were decreased in stimulated whole saliva of patients with BMS and xerostomia when compared to those in unstimulated saliva ( $p < 0.001$ ). Lysozyme levels were also lower in stimulated whole saliva in such patients when compared to the levels in unstimulated saliva ( $p < 0.001$ ). Magnesium levels remain unchanged with regard to the salivary stimulation. The results of our study indicate that the quantity and quality of saliva could have an impact on symptoms of burning mouth syndrome.*

**Key words:** *burning mouth syndrome, quantity and quality of saliva.*

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## Introduction

Burning mouth syndrome is an intraoral pain disorder characterized by burning sensations of the oral mucosa with healthy appearance of the oral mucosa. Epidemiology varies in different countries, from 1.5% in England to 2.6% in the United States. However, prevalence is highest in menopausal women, being 10-40% (1,2,3). Symptoms are usually a burning sensation, sometimes even pain, taste distur-

bances and dry mouth (4). Most usually affected sites are the tongue, lips, and palate (1,3,5). Etiological factors can be divided into local, systemic and psychological. Factors contributing in the etiopathogenesis of BMS are candidiasis, galvanism, xerostomia, prosthetic appliances, contact allergy and parafunctional habits and mandibular dysfunction, hematological deficiencies of iron, calcium, zinc, vitamin B<sub>1</sub>, B<sub>2</sub>, B<sub>6</sub>, B<sub>12</sub> and folic acid (6-17). More recently numerous studies have reported a

connection between the quantity and quality of saliva in patients suffering from burning mouth syndrome. Data based upon correlation between BMS and xerostomia in literature are controversial, although most of the studies suggest a positive relationship between these two disorders.

Mott, Grushka, Sessle (3) report that although changes in saliva composition and their connection to BMS are only to be speculated upon, it could be of great significance to confirm that connection. The same authors report that the majority of studies have failed to demonstrate significant changes in the quantity and quality of saliva in patients with BMS, and also that some studies have reported changes in the protein quantity, immunoglobulins, phosphates and pH value in patients with BMS (19, 20).

Glass (7) suggests that xerostomia is a local contributing factor in the development of BMS, and other authors also found a higher or lower percentage prevalence of xerostomia in burning mouth syndrome patients (8, 9).

Controversial data exist in regard to protein composition in the saliva of patients with burning mouth syndrome (18,19,20,21).

Salivary IgA acts as a protective agent in the oral cavity, which has been investigated by numerous authors (22,23,24,25) in various countries, also in unstimulated and stimulated saliva (26-31).

Salivary lysozyme has been investigated both in healthy (24,32,33,34) and diseased individuals (23, 35,36,37,38) with regard to its antimicrobial activity.

To date investigations on salivary magnesium quantity (25,31,39) show completely different values in healthy persons.

## Materials and Methods

The aim of this study was to evaluate a correlation between burning mouth syndrome and xerostomia, and also levels of salivary IgA, lysozyme and magnesium in unstimulated and stimulated saliva of these patients.

Forty-three patients with burning mouth syndrome and xerostomia were examined from the Department of Oral Medicine, School of Dental Medicine in Zagreb, during 1998. There were 41

women and 2 men, average age 69.2 yrs. Quantification of saliva was determined by two measurements, using sialometry between 8-12 a.m. The unstimulated saliva was obtained by expectoration method in calibrated tubes (0.1 ml scale) during five minutes while participants were sitting. The stimulated saliva samples were obtained after chewing gum for one minute in dentate individuals or after drinking 1% C-vitamin solution followed by expectoration into calibrated tubes for five minutes. C-vitamin solution was made by dissolving 1g of ascorbic acid in 1 dcl of water. The results were expressed per one minute and levels below 0.2 ml per minute were considered xerostomic. Saliva samples were centrifuged at 800 rotations for ten minutes and stored at -20°C until analysis. The salivary analysis was performed in laboratories at the Clinical Hospital Centre Rebro.

Salivary immunoglobulin A was determined using radial immunodiffusion according to Manzini (40) on standardized kits (LC Partigen IgA, Behring Diagnostics, Marburg, Germany). LC partigen consists of plates for immunodiffusion which contain an agarose gel layer with nonspecific antisera on relevant human plasma protein. Antibodies are obtained from immunized rabbit.

Lysozyme was determined according to Osserman and Lowlor (41). The method is based on the ability of enzyme to lyse *Micrococcus lysodeicticus*. The sample was placed onto agarose plates containing microorganism and lyse zones determined by the concentration of enzyme in the sample. Human urinary lysozyme serves as a standard.

Magnesium levels were determined by atomic absorbance spectrophotometry (42). Statistical analysis was performed using Student t test and Wilcoxon test.

## Results

Average values of salivary immunoglobulin A in unstimulated whole saliva of our patients were approximately the same ( $X=0.37$  g/L;  $M=0.35$  g/L;  $Mod=0.20$  g/L) with a large dispersion in borders  $\pm 1SD$  ( $SD=0.20$  g/L) and span (0-1,10 g/L) and we concluded that distribution was normal. Average values of salivary immunoglobulin A stimulated whole saliva of our patients were approximately the

same ( $X=0.18$  g/L;  $M=0.147$  g/L;  $Mod=0.26$  g/L) with a large dispersion in borders  $\pm 1SD$  ( $SD=0.15$ g/L) and span (0-0.83 g/L) and we concluded that the appearance of distribution was normal (Figure 1).

Average values of lysozyme in unstimulated whole saliva of our patients were approximately the same ( $X=35.22$  mg/L;  $M=35$  mg/L;  $Mod=35$  mg/L) with a large dispersion in borders  $\pm 1SD$  ( $SD=20.74$  mg/L) and span (5-75.0 mg/L) and we concluded that distribution was normal. Average values of lysozyme in stimulated whole saliva of our patients were approximately the same ( $X=29.01$  mg/L;  $M=25$  mg/L;  $Mod=5$  mg/L) with a large dispersion in borders  $\pm 1SD$  ( $SD=20.74$  mg/L) and span (5-75.0 mg/L) and we concluded that distribution was normal. Average values of magnesium in stimulated whole saliva of our patients were approximately the same ( $X=29.01$  mg/L;  $M=25$  mg/L;  $Mod=5$  mg/L) with a large dispersion in borders  $\pm 1SD$  ( $SD=20.74$  mg/L) and span (4-72 mg/L) and we concluded that distribution was normal (Figure 2).

Average values of magnesium in unstimulated whole saliva of our patients were approximately the same ( $X=0.46$  mmol/L;  $M=0.43$  mmol/L;  $Mod=0.28$  mmol/L) with a large dispersion in borders  $\pm 1SD$  ( $SD=0.20$  mmol/L) and span (0.17-0.97 mmol/L) and we concluded that distribution was normal. Average values of magnesium in stimulated whole saliva of our patients were approximately the same ( $X=0.41$  mmol/L;  $M=0.39$  mmol/L;  $Mod=0.29$  mmol/L) with a large dispersion in borders ( $SD=0.18$  mmol/L) and span (0.15-0.79 mmol/L) and we concluded that distribution was normal (Figure 3).

Student t test between values of sIgA in unstimulated and stimulated saliva showed a statistically significant different values ( $t=5.88$ ;  $p<0.001$ ) and showed a statistically significant decrease of sIgA in stimulated saliva in relation to unstimulated saliva. Additional testing with nonparametric Wilcoxon test showed statistically significantly decreased values of sIgA in stimulated saliva when compared to the unstimulated saliva ( $Z=4.70$ ;  $p<0.001$ ).

Student t test between lysozyme values in unstimulated and stimulated saliva showed statistically significant different values ( $t=2.08$ ;  $p<0.05$ ) and showed a statistically significant decrease of lyso-

zyme in stimulated saliva in relation to unstimulated saliva. Additional testing with nonparametric Wilcoxon test showed statistically significantly decreased lysozyme values in stimulated saliva when compared to the unstimulated saliva ( $Z=1.86$ ;  $p<0.05$ ).

Student t test between magnesium levels in unstimulated and stimulated saliva did not show statistically significantly different values, as well as testing with Wilcoxon test.

## Discussion

The results of our study show that salivary immunoglobulin A (sIgA) levels decrease together with salivary stimulation, i.e. levels of sIgA are decreased in stimulated whole saliva of patients with burning mouth syndrome and xerostomia when compared to the sIgA values in unstimulated saliva. Our results are in concordance with the results of Rudney et al. (22) who also found negative correlation between sIgA levels and salivary stimulation in healthy individuals. Ryberg et al. (23) reported decreased levels of sIgA in the stimulated saliva of patients with asthma when compared to healthy individuals.

Mandel (24) reported that together with salivary stimulation levels of sIgA decrease as well in healthy individuals. The same results were reported by Gronblad (25), Brandtzaeg (26), Mandel and Khurana (27) and South et al. (28). Sreebny and Zhu (29) found elevated sIgA levels in whole unstimulated saliva of patients with Sjogren syndrome. Aguirre et al. (30) did not find any differences in sIgA quantity in parotid saliva with regard to the age of examinees. Syrjanen et al. (31) found decreased sIgA levels in unstimulated saliva of patients undergoing radiation therapy. Streckfus et al. (32) did not find any differences in sIgA quantity in diabetic patients when compared to a control group. Gahnberg and Krasse (33) found that levels of sIgA varied significantly in individuals over a four month period which could explain huge differences in sIgA reported in many studies. Narhi et al. (34) found negative correlation between sIgA levels and salivary stimulation.

Our results show that lysozyme levels are decreased in the stimulated whole saliva of patients with BMS

and xerostomia and are in concordance with the findings of Rudney et al. (22) and Narhi (34) in healthy individuals, although contradictory to the previous findings of Rudney (22). Jalil et al. (35) did not report any differences in lysozyme quantity in unstimulated and stimulated saliva of healthy individuals. Sreebny and Zhu (29) found elevated levels of lysozyme in the parotid saliva of patients with Sjogren syndrome. In HIV infected patients, lysozyme levels in unstimulated saliva tend to be elevated when compared to healthy individuals as reported by Tsang and Samaranayake (36).

Moutsopoulos et al. (37) found elevated levels of lysozyme in the unstimulated whole saliva of patients with Sjogren syndrome. Ryberg et al. (23) found decreased lysozyme levels in the unstimulated saliva of patients with asthma when compared to healthy individuals. Makkonen et al. (38) reported elevation of lysozyme quantity in patients undergoing radiation therapy.

Magnesium levels remain unchanged in unstimulated and stimulated saliva. Our results show that magnesium secretion is equal for a short period of time. Dawes (39) found also unchanged magnesium levels with regard to salivary stimulation in healthy individuals. On the other hand Mandel (24) reported decreased magnesium quantity in stimulated parotid and submandibular saliva after 2% citric acid stim-

ulation when compared to the levels in unstimulated saliva. Syrjanen (31) found decreased salivary magnesium levels in patients with galvanism compared to a control group

### Conclusion

On the basis of this study the following can be concluded:

1. Levels of sIgA in stimulated whole saliva are decreased when compared to the levels in unstimulated whole saliva of patients with burning mouth syndrome.
2. Levels of lysozyme are decreased in the stimulated whole saliva of patients with burning mouth syndrome when compared to the levels in unstimulated whole saliva. A possible explanation for these results is connected with decreased functional activity of the salivary glands in xerostomia and a possible role in the development of burning mouth syndrome.
3. Magnesium levels remain unchanged with regard to stimulation. Salivary glands do not have significance in magnesium production and salivary magnesium does not have an influence on burning mouth syndrome.