Coll. Antropol. 34 (2010) 3: 1039–1043
Original scientific paper

Effects of Low-Level Laser Treatment on Mouth Dryness

Danica Vidović Juras[1,2], Josip Lukač[3], Ana Cekić-Arambašin[1,2], Anđelko Vidović[4], Ivana Canjuga[2], Miroslav Sikora[5], Andreja Carek[2] and Mario Ledinsky[3]

[1] Dental Clinic, Zagreb University Hospital Center, Zagreb, Croatia
[2] School of Dental Medicine, Zagreb University, Zagreb, Croatia
[3] »Sestre milosrdnice« University Hospital, Zagreb, Croatia
[4] Dubrava University Hospital, Zagreb, Croatia
[5] Osijek University Hospital Center, Osijek, Croatia

A B S T R A C T

Mouth dryness (MD) is usually followed by inadequate mechanical cleaning of the mouth and decrease in the levels of salivary antimicrobial proteins (including secretory immunoglobulin A (sIgA)). It is accompanied by difficulties during speaking and food swallowing, with an unpleasant taste, burning sensations in the mouth and higher susceptibility to oral diseases. Low-level laser treatment (LLLT) can intensify cell metabolism and its application on salivary glands could improve salivation. The purpose of this study was to evaluate the effects of LLLT on salivation of patients suffering from MD. The study included 17 patients with MD. Their major salivary glands were treated with low intensity laser BTL2000 on 10 occasions. The whole unstimulated and stimulated saliva quantities were measured just before the 1st, after the 10th and thirty days following the last (10th) treatment. In the samples of unstimulated saliva concentrations of sIgA were estimated by using ELISA method and its quantity in the time unit was calculated. The visual analogue scale (VAS) score was used to assess burning and/or pain intensity at these three time points. Statistical tests revealed significant salivation improvement quantitatively and qualitatively, i.e. increase in the quantity of saliva and sIgA. VAS score was also significantly improved and no side effects were observed. Conclusions: According to the results of this study, application of LLLT to xerostomic patients' major salivary glands stimulates them to produce more saliva with better antimicrobial characteristics and improves the difficulties that are associated with MD. This simple non-invasive method could be used in everyday clinical practice for the treatment of MD.

Key words: *low-level laser treatment, mouth dryness, secretory immunoglobulin A*

Introduction

Mouth dryness (MD), dry mouth, hyposalivation and xerostomia are common names for chronic reduction in salivary glands secretion. It represents the state associated with numerous causes when the patient produces less than 1.5 ml of whole saliva over a 15 minute period without stimulation[1]. According to epidemiological studies, 10–47% of the population[2], depending upon diagnostic method, suffer from MD.

MD can have a significant adverse effect upon oral health and quality of life[1]. It is often accompanied by difficulties with speaking, chewing, food swallowing, by impaired or diminished taste sensation[3], and also by oral burning or soreness[4]. Furthermore, there is an insufficiency of other important salivary functions including mechanical cleansing action, control of pH, removal of food debris from the oral cavity, remineralization, maintaining the integrity of the oral mucosa and antimicrobial activity[5]. Impaired salivary functions and decrease in the levels of salivary antimicrobial agents, including secretory immunoglobulin A (sIgA), lead to higher susceptibility to oral infections, rampant caries[6] and periodontal diseases[7]. The treatment of MD remains very unsatisfactory[1]. It is primarily palliative, with emphasis on the use of saliva substitutes[4]. These are often not completely successful and systemic sialogogues, which are associated with risks of side effects as well, have not

Received for publication May 20, 2009

been shown to conclusively improve salivary function[8]. Therefore, new, efficient and safe solutions are required.

Numerous studies have shown beneficial effects of low-level laser treatment (LLLT) on biological tissues. Its mechanism of action is based on absorption of laser light by tissue and consequential enhanced synthesis of adenosine triphosphate[9], i.e. on increase of the energy available to cells. Therefore, at adequate wavelength, intensity and dose, LLLT is capable to achieve many advantageous effects such as increased cell proliferation and collagen production[10], accelerated collateral circulation and enhanced microcirculation[11], accelerated tissue repair[12], promoted nerve regeneration or pain relief[13]. Anti-inflammatory, antiedematous, analgesic[14] and immunomodulating[15] as well as many other desirable effects of LLLT are also well known. Moreover, there are no contraindications reported for its use in dentistry and no side effects associated with the use of LLLT have been reported[16].

Since the usefulness of LLLT has been shown in the management of numerous diseases and conditions, we assumed that it could be useful in MD as well. The aim of this study was to find out whether LLLT of the MD patients' salivary glands can result in salivation improvement. Particular attention was paid to the potential of LLLT to increase salivary flow rates, to induce synthesis of sIgA and to facilitate accompanied subjective difficulties.

Patients and Methods

The Ethics Committee of School of Dental Medicine, Zagreb University, Zagreb, Croatia, approved the study protocol. Written informed consent according to Helsinki II Agreement was obtained from each participant. Seventeen patients (two men and fifteen women, mean age 68.8±12.2 years) diagnosed with MD on the basis of salivary flow measurement voluntary participated in the study, conducted at the Department of Oral Diseases, Dental Clinic, Zagreb University Hosputal Center and School of Dental Medicine, Zagreb University.

At the time of the study all the participants were non-smokers and without clinical signs and symptoms of oral diseases and conditions other than MD. MD was diagnosed when subjects produced less than 1.5 ml of whole unstimulated saliva over a 15 minute period[1]. None of them were taking any medication influencing salivary secretion at the time of the study.

Patients´ major salivary glands were treated with low power semiconductor diode laser (BTL-2000, Prague, Czech Republic) by using infrared probe/diode (energy density 1.8 J/cm^2, frequency 5.2 Hz, output power 30 mW) on 10 occasions. LLLT was performed five days per week for two consecutive weeks. Each parotid gland was exposed for 5 minutes; submandibular glands were exposed for 2 and sublingual for 1 minute per treatment. The exposition of parotid and submandibular glands was performed extraorally and sublingual glands intraorally with the laser diode approximately 5 mm far from surface over glands anatomic sites. In order to cover the whole area of the glands treated, slow circulating movements were performed during the treatment. Energy dose delivered to each parotid gland was 7.2 J, to each submandibular gland 2.88 J and to sublingual gland 1.44 J per treatment. Hence, total energy dose delivered in a patient session was 23.04 J and the accumulated energy delivered on the whole in 10 sessions was 230.4 J.

The whole unstimulated and stimulated saliva were collected and quantified just before the 1st, after the 10th and thirty days following the last (10th) treatment. Each time this was performed between 9–11 a.m., at least 2 h after the last intake of food or drink. Under resting conditions, while participants were sitting with head bent slightly forward, just after saliva swallowing, participants spat whole unstimulated saliva every 60 seconds into calibrated containers (0.1 ml) during five minutes. Thereafter they were asked to rinse their mouths with 50 ml of 1% ascorbic acid solution for 60 seconds. After swallowing, participants spat whole stimulated saliva, in an equivalent way as unstimulated saliva, into other calibrated tubes for the next five minutes. Unstimulated and stimulated salivary flow rates were recorded for each participant and expressed in mililiters per five minutes. The samples of unstimulated whole saliva were frozen and stored at –20°C until used for sIgA determination. sIgA concentrations were measured by using commercially available indirect competitive enzyme immunoassay kit (Salivary SIgA EIA kit, Salimetrics, State College, PA, USA). According to the manufacturer's manual the intra-assay precision was determined from the mean of 10 replicates each (saliva samples) and coefficient of variation (CV) ranged from 4.49% to 6.99% for the samples with high, medium and low concentrations of sIgA. The inter-assay precision was determined from the mean of average duplicates for 8 separate runs with the CVs of 8.65% for the samples with high concentration and 8.93 for the samples with low concentration of sIgA.

The quantities of salivary IgA secreted in 5 minutes were calculated by taking in account sIgA concentrations and volumes of unstimulated saliva secreted in 5 minutes.

Just after saliva collection, at the same three time points, burning and/or pain intensity was assessed by using a 10 cm long visual analogue scale (VAS).

Statistical analysis

Normality of the distribution of variables was checked with Shapiro-Wilks' tests. Since all the variables followed Gaussian distribution, we performed repeated measures ANOVA for testing the effect of treatment on variables determined at three time points (3x1 design). If ANOVA proved to be significant, Tukey's post-hoc tests were performed to test the differences between single time points. $p<0.05$ was considered statistically significant. Data were analyzed using Statistica V6 (Statsoft, Tulsa, USA).

Results

Statistical analysis revealed that LLLT improved participants' salivation quantitatively and quantitatively, i.e. increased the quantities of saliva (Figures 1 a and b) and sIgA (Figure 1c).

The quantity of unstimulated saliva gradually increased during the study, from 0.6±0.3 mL/5min ($\bar{X}$± SD) just before the first LPLP, through 0.9±0.6 mL/5min after the 10th LLLT, to 1.1±0.8 mL/5min 30 day following the last LLLT. It was significantly higher 30 days after treatment when compared to baseline (Figure 1a).

The quantity of stimulated saliva was 1.4±0.6 mL/5min ($\bar{X}$±SD) at the beginning. It increased after the 10th LLLT (2.3±1.0 mL/5min) and remained elevated 30 days following the LLLT (2.4±1.3 mL/5min). Compared to baseline, at both time points the increase was statistically significant (Figure 1b).

The quantity of sIgA in unstimulated saliva secreted in 5 minutes just before the LLLT was 236.1±125.8 μg/5min ($\bar{X}$±SD). Immediately after the 10th LLLT it was higher (287.7±171.6 μg/5min), but this increase was not significant. Thirty days after the 10th LLLT it measured 384.9±214.6 μg/5min, which was significantly higher when compared to baseline (Figure 1c).

Intensity of burning and/or pain determined by VAS was 4.8±3.1 cm ($\bar{X}$±SD) at baseline, 2.2±2.1 cm immediately after the 10th treatment, and 2.0±2.1 cm 30 days after the 10th treatment. According to the participants' reports, the burning and/or pain intensity was reduced significantly after the 10th LLLT and it remained significantly lower 30 days after LLLT (Figure 1d). There were no side effects reported during this study.

Discussion

Numerous studies have been performed with the aim of investigating the possibilities of using low-level laser in dentistry. Despite this fact, the efficacy of LLLT in patients suffering from MD has been scantily investigated up to this point.

The results of our study prove the positive therapeutic effect of the LLLT on xerostomic patients' major salivary glands and on concomitant burning and/or pain sensations. By applying of the LLLT, it seems possible to stimulate major salivary glands to produce more saliva with better antimicrobial characteristics and to relieve the difficulties related to MD and eventually improve their quality of life. The fact that the advantages of LLLT were observed just after the last (10th) treatment but also in the follow up period of 30 days when they were even

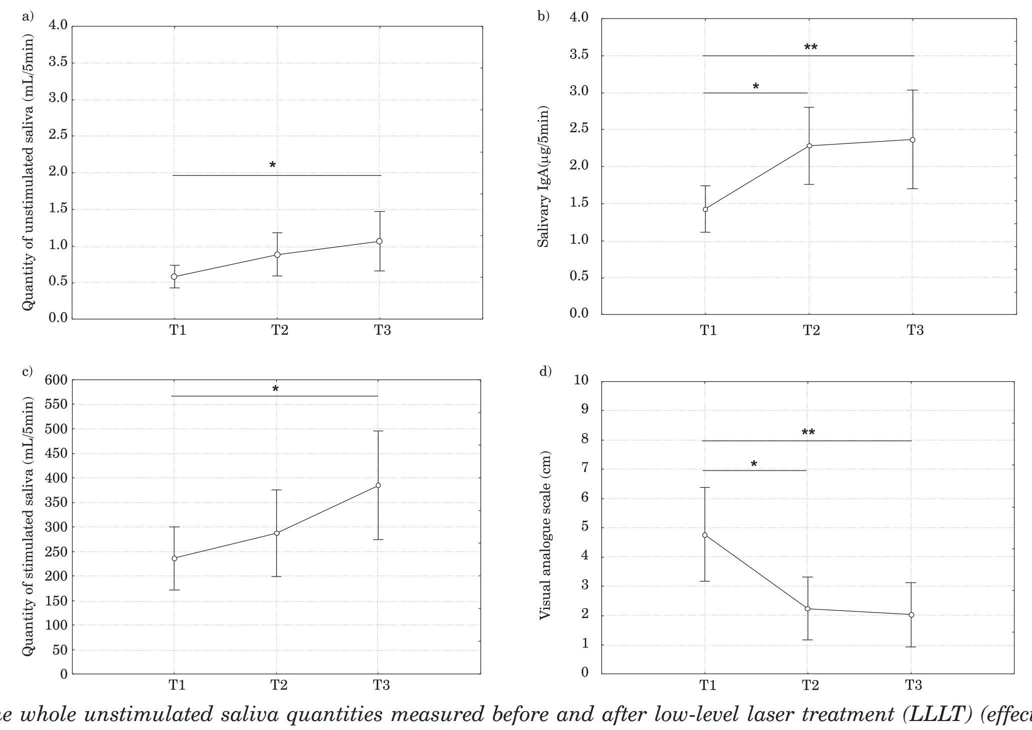

*Fig. 1. a) The whole unstimulated saliva quantities measured before and after low-level laser treatment (LLLT) (effect of treatment F(2.32)=5.614; p=0.008; *T1* vs. *T3, p=0.006). b) The whole stimulated saliva quantities measured before and after LLLT (effect of treatment F(2.32)=10.265, p<0.001; *T1* vs. *T2, p=0.002, **T1* vs. *T3, p<0.001). c) The quantities of salivary IgA in unstimulated saliva measured before and after LLLT (effect of treatment F(2.32)=3.940, p=0.029; *T1 vs.T3, p=0.025). d) The burning and/or pain intensity using visual analogue scale measured before and after LLLT (effect of treatment F(2.32)=15.827, p<0.001; *T1* vs. *T2, p<0.001, **T1* vs. *T3, p<0.001). T1 – before the 1st LLLT, T2 – after the 10th LLLT, T3 – 30 days following the last (10th) LLLT. Data were analyzed with repeated measures ANOVA (analysis of variance) and Tukey's post-hoc tests. Vertical lines denote 95% confidence intervals of the mean.*

more pronounced proves that LLLT effects are prolonged and indicates that, besides biostimulative, they also have regenerative effect.

Although none of the few available study reports on LLLT effect on salivary glands revealed similarly designed study, some findings of those studies are important for our results discussion. In the first experimental study of the effect of Ga–As semiconductor laser on salivary glands, which was carried out on rat submandibular glands, Takeda[17] found an increase in the mitoses of duct epithelial cells. Takahashi[18] proved regeneration of the intralobular duct and acinus in rat submandibular glands after YAG laser irradiation. Simões[19] observed positive effects of LLLT on salivary flow rate and some salivary parameters in rats. Lopes[20] stated that InGaAIP laser was effective in patients with head and neck cancer submitted to radiotherapy in prevention of radiotherapy-induced xerostomia, oral mucositis and related pain. In a double blind randomized trial Cowden[21] showed the efficiency of He-Ne laser in the prevention of oral mucositis and xerostomia induced by high dose chemoradiotherapy before autologous bone marrow transplantation. Tuner[22] and Simões[19] published results showing a fast response of the Sjögren's syndrome patients' glands to laser irradiation and efficiency regarding MD and inflammatory process. Kats[23] reported high therapeutic efficiency of LLLT in treatment of chronic nonspecific sialoadenitis resulting in rapid resolution of inflammatory symptoms and pain, high salivation, longer remission and a recovery of the gland structure. Ross[13] reported that LLLT results in nerve regeneration and this could mean also in stimulation of parasympathetic secretomotor innervation of the salivary glands. The above-mentioned reports are in accordance with our observations that LLLT is capable to increase saliva production. The exact mechanism responsible for such effect of LLLT on salivary glands and its secretion remains poorly understood[24]. Almost certainly, future explanation of the mechanism underlying these effects will include the ability of LLLT to increase the local-circulation through vasodilatation, to induce glandular cell proliferation, to improve cell respiration/ATP synthesis and release of growth factors and cytokines[25], to stimulate protein exocytosis[26], and to promote nerve regeneration[13].

Petrek[27] found stimulatory effect of He-Ne laser on the immune system of the patients suffering from chronic tonsillitis. Among other effects, shortly after the cessation of the treatment, he observed clinical improvement and significant increase of sIgA which was followed by increase of IgA serum levels after 4 weeks of follow-up. Kucerová[28] reported an increased sIgA and the pain diminishing following the He-Ne laser therapy after molar extraction. Likewise, our results showed that LLLT significantly increased sIgA and relieved patients' symptoms. A possible explanation for the increase of sIgA is that local laser irradiation intensifies the activation of B lymphocytes, whose differentiation into plasma cells contributes to an increase in immunoglobulin levels[27,28]. The potential of LLLT to increase sIgA, the predominant immunoglobulin found in saliva[29], could be of significant importance. This is due to the fact that sIgA, characterized by its antiadhesive action against microorganisms, is considered to be the main specific defence mechanism in the oral cavity[30].

Another important, analgesic effect of LLLT, also demonstrated in this study, is in line with the findings reported by many other authors[16,20,21,23,25,31,32] as well. Current explanation of analgesic effect of LLLT, although it hasn't been completely clarified, is based on its possibility to enhance synthesis of endorphins and bradykinins, to decrease c-fiber activity and to alter pain threshold[16,25]. Some authors hypothesized that this analgesic effect was mediated by serotonin and acetylcholine released centrally, and by histamine and prostaglandins released peripherally with the use of LLLT[25,33].

None of the available reports, publised results of international experimental and clinical research conducted in the period longer than last thirty years, indicated any true side effects associated with the use of low-level laser light[16]. Our findings confirm these observations.

On the basis of our study results, we can conclude that LLLT has been shown to be effective in treatment of MD and free of any side effects. Since currently used treatments of MD, including palliative short lasting saliva substitutes and systemic siagogogues associated with the risk of numerous side effects, are very unsatisfactory[1], LLLT seems to be superior and promising therapeutic solution. Although the results of our study indicate positive effects of LLLT in patients suffering from MD, further investigations with longer follow up period and/or addition of placebo group are needed to determine specificity and duration of beneficial effects and to ascertain if this simple, non-invasive and free of side-effects method could be introduced into everyday clinical practice as a routine procedure for the treatment of MD.

Acknowledgements

This study was supported by the Ministry of Science, Education and Sport of the Republic of Croatia (Project No. 065-0650445-0485).

REFERENCES

1. PORTER SR, VUČIĆEVIĆ BORAS V, Acta Stomatol Croat, 41 (2007) 95. — 2. THOMSON WM, LAWRENCE HP, BROADBENT JM, POULTON R, Health Qual Life Outcomes, 4 (2006) 86. — 3. CASSOLATO SF, TURNBULL RS, Gerodontology, 20 (2003) 64. — 4. GUGGENHEIMER J, MOORE PA, J Am Dent Assoc, 134 (2003) 61. — 5. BARTELS CL, Helping patients with dry mouth, BMC Pediatrics, accessed 29.01.2009. Available from: URL: http://www.oralcancerfoundation.org/dental/xerostomia.htm. — 6. SILVESTRE FJ, MINGUEZ MP, SUÑE-NEGRE JM, Med Oral Patol Oral Cir Bucal, 14 (2009) 8. — 7. MATEAR DW, BARBARO J, J R Soc Health, 125 (2005) 35. — 8.

GRISIUS MM, Oral Surg Oral Med Oral Pathol Oral Radiol Endod, 92 (2001) 156. — 9. CASTANO AP, DAI T, YAROSLAVSKY I, COHEN R, APRUZZESE WA, SMOTRICH MH, HAMBLIN MR, Lasers Surg Med, 39 (2007) 543. — 10. POSTEN W, WRONE DA, DOVER JS, ARNDT KA, SILAPUNT S, ALAM M, Dermatol Surg, 31 (2005) 334. — 11. IHSAN FR, Photomed Laser Surg, 23 (2005) 289. — 12. RIBEIRO MS, DA SILVA DDE F, DE ARAÚJO CE, DE OLIVEIRA SF, PELEGRINI CM, ZORN TM, ZEZELL DM, J Clin Laser Med Surg, 22 (2004) 59. — 13. ROSS G, ROSS A, Gen Dent, 56 (2008) 629. — 14. EKIM A, ARMAGAN O, TASCIOGLU F, ONER C, COLAK M, Swiss Med Wkly, 137 (2007) 347. — 15. GLUSHKOV OV, NOVOSELOVA EG, CHERENKOV DA, NOVOSELOVA TV, KHRENOV MO, LUNIN SM, CHUDNOVSKII VM, IUSUPOV VI, FESENKO EE, Biofizika, 51 (2006) 123. — 16. SUN G, TUNÉR J, Dent Clin North Am, 48 (2004) 1061. — 17. TAKEDA Y, J Oral Pathol, 17 (1988) 91. — 18. TAKAHASHI S, WAKITA M, Arch Histol Cytol, 56 (1993) 199. — 19. SIMÕES A, NICOLAU J, DE SOUZA DN, FERREIRA LS, DE PAULA EDUARDO C, APEL C, GUTKNECHT N, Clin Oral Invest, 12 (2008) 25. — 20. LOPES CO, MAS JRI, ZÂNGARO RA, Radiol Bras, 39 (2006) 131. — 21. COWEN D, TARDIEU C, SCHUBERT M, PETERSON D, RESBEUT M, FAUCHER C, FRANQUIN JC, Int J Radiat Oncol Biol Phys, 38 (1997) 697. — 22. TUNÉR J, HODE L, Low level laser therapy-clinical practice and scientific background (Prima Book, Grängesberg, Sweeden, 1999). — 23. KATS AG, BELOSTOTSKAIA IM, MALOMUD ZP, MAKAROVA LG, Vestn Khir Im I I Grek, 135 (1985) 39. — 24. SIMÕES A, SIQUEIRA WL, LAMERS ML, SANTOS MF, EDUARDO CDE P, NICOLAU J, Lasers Med Sci, 24 (2009) 202. — 25. PARKER S, Br Dent J, 202 (2007) 131. — 26. MARQUES MM, PEREIRA AN, FUJIHARA NA, NOGUEIRA FN, EDUARDO CP, Lasers Surg Med, 34 (2004) 260. — 27. PETREK M, HUBÁCEK J, ORDELTOVÁ M, Acta Univ Palacki Olomuc Fac Med, 129 (1991) 119. — 28. KUCEROVÁ H, DOSTÁLOVÁ T, HIMMLOVA L, BÁRTOVÁ J, MAZÁNEK J, J Clin Laser Med Surg, 18 (2000) 309. — 29. SEEMANN R, HÄGEWALD SJ, SZTANKAY V, DREWS J, BIZHANG M, KAGE A, Clin Oral Investig, 8 (2004) 233. — 30. MARCOTTE H, LAVOIE MC, Microbiol Mol Biol Rev, 62 (1998) 71. — 31. CEKIĆ-ARAMBAŠIN A, ĐURĐEVIĆ-MATIĆ A, MRAVAK-STIPETIĆ M, BILIĆ A, Acta Stomatol Croat, 24 (1990) 281. — 32. NAKASE M, OKUMURA K, TAMURA T, KAMEI T, KADA K, NAKAMURA S, INUI M, TAGAWA T, Oral Dis, 10 (2004) 217. — 33. CEYLAN Y, HIZMETLI S, SILI ^ Y, Rheumatol Int, 24 (2004) 260.

D. Vidović Juras

Department of Oral Disease, Dental Clinic, Zagreb University Hospital Center, Gundulićeva 5, 10000 Zagreb, Croatia
e-mail: djuras@sfzg.hr

UČINKOVITOST LASERA NISKE IZLAZNE SNAGE U LIJEČENJU SUHOĆE USTA

S A Ž E T A K

Suhoću usta (MD) redovito prati neadekvatno mehaničko čišćenje usta i smanjenje količine salivarnih antimikrobnih proteina (uključujući sekretorni immunoglobulin A (sIgA)). Tu su i poteškoće tijekom govora i gutanja hrane, neugodan okus, osjećaj pečenja u ustima i veća sklonost prema oralnim bolestima. Tretman laserom niske izlazne snage (LLLT) može intenzivirati metabolizam stanica te bi njegova aplikacija na žlijezde slinovnice mogla poboljšati lučenje sline. Svrha je ovog istraživanja procijeniti učinke LLLT-a na salivaciju bolesnika koji pate od MD-e. Sudjelovalo je 17 ispitanika. Velike žlijezde slinovnice bolesnika 10 su puta bile tretirane laserom BTL 2000 niske izlazne snage. Neposredno prije prvog, te nakon desetog i tridesetog dana nakon zadnjega, desetog tretmana mjerili smo ukupnu nestimuliranu i stimuliranu slina. U uzorcima nestimulirane sline pomoću ELISA-e određivali smo koncentracije sIgA te izračunali njegovu količinu u jedinici vremena. Uz pomoć vizualno-analogne ljestvice procjenjivali smo intenzitet pečenja i/ili boli u sve tri vremenske točke. Statistička analiza rezultata otkrila je znatno poboljšanje salivacije nakon LLLT-a. Ono je bilo i kvantitativno i kvalitativno, tj. znatno je bila povećana količina sline i sIgA. Rezultati mjerenja prema ljestvici VAS također su bili mnogo bolji, a nepoželjni učinci nisu uočeni. Prema rezultatima ovog istraživanja, LLLT velikih žlijezda slinovnica bolesnika s MD-om potiče ih da proizvedu više sline s boljim antimikrobnim karakteristikama i olakšava poteškoće koje prate MD-u. Ta jednostavna neinvazivna metoda ima potencijala da se uvede u svakodnevnu kliničku praksu kao rutinski postupak za liječenje MD-e.