

# Proceedings of FIB

# Volume V



Anna Carolina R.T. Horliana Organizadora

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# **Proceedings of FIB**

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Universidade Nove de Julho

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#### Prefácio:

No V FÓRUM INTERNACIONAL DE BIOFOTÔNICA foram discutidos temas relacionados à Biofotônica e suas aplicações em Ciências da Saúde, com a finalidade de divulgar as amplas possibilidades de pesquisa e aplicação clínica resultantes de uma visão interdisciplinar aplicada à essa temática. O evento teve como público os pesquisadores, docentes e discentes de graduação e pós-graduação de todo o Brasil e da América Latina, bem como os profissionais de saúde dos serviços privados e do Sistema Único de Saúde (SUS). Foram promovidas discussões de caráter científico e tecnológico, proporcionando assim melhoria da formação dos pesquisadores e profissionais envolvidos nessa área de conhecimento. O evento foi totalmente gratuito, alcançando, portanto, um maior número de participantes. Foram realizadas 2598 inscrições on line, sendo que todas as inscrições foram realizadas *on-line*. Nesta edição, foi realizado um pré-evento (dia 2 de outubro de 2019) que contemplou o I Encontro Latino Americano de Biofotônica e o I Fórum de Inovação em Biofotônica.

O I **Encontro Latino Americano de Biofotônica** teve como objetivo de compartilhar experiências e discutir novos meios para difusão e colaboração na produção de novas tecnologias e inovações científicas entre os países da América Latina. Foram proferidas palestras pelos professores Prof. Dr. José Luis González Solís (Universidade de Guadalajara, México), Prof. Dr. Martín Eugenio Rapi (Universidade Nacional de Buenos Aires, Argentina), Prof. Dr. Jose Maria Aguilera (Universidade Nacional de Assunção, Paraguai) e pela profa Profa. Dra. Kristianne Porta Santos Fernandes (UNINOVE, Brasil). Ao final foi composta mesa de debate composta por representantes do PPG em Biofotônica aplicada às Ciências da Saúde e os professores convidados das instituições de referência na América Latina.

No período da tarde tivemos o **I Fórum de Inovação**, com intuito de discutir a Parceria Universidade / Indústria no âmbito da inovação e do empreendedorismo em Biofotônica. Participaram dessa discussão o representante da ABIMO, Dr. Joffre Moraes (Ass. Bras. da Indústria de Artigos e Equipamentos Médicos e Odontológicos), o representante da SISQUALY, Dr. Luiz Eduardo da Costa que falou sobre a regulação para equipamentos e insumos no Brasil. Também participaram representantes das nossas empresas-parceiras DMC. MMO, Cosmedical e Biolambda que expuseram seus produtos, planos de inovação, dificuldades de regulamentação e abrangência no mercado nacional e internacional.

O V Fórum Internacional de Biofotônica (3 e 4 de outubro de 2019) contou com apresentações plenárias, apresentações orais de trabalhos científicos de PPGs de todo o Brasil, e a sessão de apresentação de posters, com carga horaria total de 24h. A programação deste ano foi voltada para todas as áreas que podem se beneficiar das tecnologias de aplicação da luz em saúde, a saber: Medicina, Odontologia, Enfermagem, Estética, Fisioterapia, Farmácia, Biomedicina, Medicina veterinária e Biologia. Visto que a Biofotônica é uma área multidisciplinar, entendemos que a participação das diversas vertentes do conhecimento científico é esperada para evolução de diálogo construtivo. Portanto contamos com um grupo de altíssimo impacto científico, como o Profa. Dra. Celine Frochot (University of Lorraine, Nancy), a Profa. Dra. Cristiano Carlomagno (IRCCS Fondazione Don Carlo Gnocchi, Milano, Italy), Prof. Dr. Fábio Parra Sellera (FMVZ - USP), Profa. Dra. Ana Cecília Corrêa Aranha (FO-USP), Profa. Dra. Patrícia da Ana (UFABC), Profa. Dra. Ana Claudia Muniz Rennó (UNIFESP, Santos), Profa. Rita de Cassia Ferreira (Núcleo de Biofotonica - Conjunto Hospitalar do Mandaqui), Profa. Virgínia M.M. Rodrigues (UNINOVE, Brasil), Profa. Adriana Schapochnik (UNINOVE, Brasil) e Profa. Lidiane Rocha Mota (UNINOVE, Brasil). Como encerramento, propusemos uma nova atividade chama de Brainstorm: a pesquisa em Biofotônica e seu impacto na sociedade. O Prof. Dr. Nivaldo Antonio Parizotto (UNIARA, UFSCAR, Brasil) falou sobre o Histórico da Biofotônica e o prof Prof. Dr. Vanderlei Salvador Bagnato (IF USP-São Carlos, Brasil) sobre o desenvolvimento tecnológico em Biofotônica. Na sequência, os dois palestrantes foram convidados para participar de uma mesa de discussão com outros nomes relevantes na área da Biofotônica do Brasil e da América Latrina: Profa. Dra. Maria Cristina Chavantes (UNINOVE, Brasil), Profa. Dra. Martha Simões Ribeiro (IPEN, Brasil), Prof. Dr. Martín Eugenio Rapi (Universidade Nacional de Buenos Aires, Argentina), Prof. Dr. Renato Amaro Zângaro (Anhembi Morumbi, Brasil), Prof. Dr. Ricardo Scarparo Navarro (Brasil, Brasil), Profa. Dra. Sandra Kalil Bussadori (UNINOVE, Brasil), Profa. Dra. Silvia Cristina Nunez (Brasil, Brasil), Profa. Dra. Luciana Almeida Lopes (Nupen, Brasil), Prof. Dr. José Luis González Solís (México), A discussão foi mediada pela profa Profa. Dra. Kristianne Porta Santos Fernandes (Brasil) (UNINOVE).

Para este evento foram submetidos 104 trabalhos que foram avaliados pelo Comitê Científico do evento. Destes trabalhos 89 foram aprovados para publicação em Proceeding e 80 foram selecionados para apresentação em forma de banner. Os 8 melhores trabalhos submetidos foram selecionados para apresentação oral, sendo 4 deles selecionados para premiação gentilmente cedida pelas nossas empresas-parceiras. Nessa atividade foram divulgados os resultados mais atuais dos projetos de pesquisa realizados em 34 diferentes instituições, entre elas: FM ribeirão Preto, Hospital do Mandaqui, Instituto de Medicina Tropical, IPEN-CNEN/SP, São Leopoldo Mandic, UFABC, UFBA- Vitória da Conquista, UFMG-Ouro Preto, UFRJ, UFRS, UNIFESP-Santos, UNIFESP/SP, UNIMES-Santos, UNINOVE, Universidade Cruzeiro do Sul, Universidade da Virgínia/USA, Universidade Lusíadas, Santos, USP-São Carlos, USP-SP. Essas apresentações foram marcadas por muita discussão e interação entre os PPGs, que por consequência, melhorarão a qualidade da produção devido à discussão com importantes nomes da pesquisa nacional e internacional.

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### **Guest Speakers:**

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Lessa, Luis Gustavo Franco	LASERTHERAPY EQUIPMENT FOR APPLICABILITY IN ORTHODONTICS "and other areas, for oral use with protocols and patent"
Ligeiro-de-Oliveira, Ana	Clinical and histological quality of mammoplasty scars after photobiomodulation: a randomized, controlled, double-blind clinical trial
Paula	Anti-inflammatory effect of photobiomodulation in an experimental model of Chronic Obstructive Pulmonary Disease (COPD) caused by smoking: participation of Treg cells.
Lima, Barbara	Photodynamic therapy decreases cellular viability and sphere formation in oral squamous cell carcinoma cell lines
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	Evaluation of Mesenchymal Stem Cells Metabolism using Conditioning Treatment or Single Dose Irradiation
Lima, Rafael de	Evaluation of Mesenchymal Stem Cells Metabolism using Conditioning Treatment or Single Dose Irradiation
	The increase in radiant exposure do not increase methylene blue PDT antimicrobial action in Candida albicans planktonic culture when low irradiance is used
	Effect of photodynamic therapy on halitosis treatment in dentate patients.
Llanos Do Vale, Kátia	Efficacy of PDT and lingual scraper in the treatment of halitosis in elderly patients with complete denture: a randomized, controlled trial
Longo, Priscila Larcher	Evaluation of photodynamic therapy in pericoronitis: protocol of randomized, controlled, double-blind study.
	Analysis of muscular injury repair in rats submitted to systemic photobiomodulation
Lopez, Talita	Biochemical evaluation of the blood of rats with muscular injury submitted to systemic photobiomodulation
Lourenço, Matheus França	Evaluation of photobiomodulation in salivar production of patients with anti- hypertensive drug-induced xerostomia

Luz, Suzete Carvalho Landulfo	Photodynamic Therapy: Effectiveness and molecular mechanisms of curcumin action as a photosensitizer against Pseudomonas aeruginosa biofilm.
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Marcos, Rodrigo Labat	PHOTOBIOMODULATION USING LASER THERAPY IN THE CONTROL OF THE INFLAMMATORY PROCESS AND PROMOTING REPAIR IN THE TENDON IN THE EXPERIMENTAL TENDINITIS MODEL.
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Mayer, Marcia Pinto Alves	Evaluation of photodynamic therapy in pericoronitis: protocol of randomized, controlled, double-blind study.
Mello, Rafaela	Photobiomodulation in Oral Mucositis in Patients with Head and Neck Cancer: a cost- effectiveness study
Melo	Study of the larvicide activity of curcumine by Photodynamic Therapy in different concentrations and blue led (460nm) about wild Aedes aegypti's mosquitoes
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Mendes, Davi	UVA detection by melanopsin regulates proliferation, pigmentation, and molecular clock in normal and malignant melanocytes
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Motta, Pamella De Barros	Comparative study between photodynamic therapy with urucum and LED and probiotics in the halitosis reduction – protocol for a controlled randomized clinical trial
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Nascimento, Helenyce Reis Do	Hydroxyl radical generation by photosensitization of phenothiazinium dyes is affected by the media
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Pereira, Benedito Jorge	Effects of photobiomodulation in salivary glands of chronic kidney disease patients on hemodialysis
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	Level of pain, muscle strength and balance in women with knee osteoarthritis after a sensory-motor exercise program and photobiomodulation therapy via cluster.
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### EVALUATION OF SALIVARY AND SOROLOGICAL LEVELS OF PRO AND ANTI-INFLAMMATORY CYTOKINES PATIENTS WITH ORAL LICHEN PLANUS TREATED WITH PHOTOBIOMODULATION

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Oral lichen planus (OLP) is a chronic autoimmune mucocutaneous disease mediated by T lymphocytes with unknown etiology. Clinically has different reticular, atrophic and erosive aspect, associate with symptomatology. Actually treatment consist in topical and/or systemic use corticosteroids, which are associated with side effects. Many patients become refractory and irresponsive to this treatment and looking for a new one. Photobiomodulation (PBM) has shown promising results in OLP. Comparing results, pro and anti-inflammatory cytokines (IL6, IL1 $\beta$ , IL10, IL17 and TNF- $\alpha$ ) were evaluated at serological and salivary levels before and after treatments, through ELISA method.

Objectives: The objective of this study is to evaluate the effect of PBM in the control of inflammatory diseases, through a clinical, controlled, randomized and double blind clinical trial.

Methods: Thirty six patients with histopathological diagnosis of OLP were included, randomized into two groups. Corticoid group (n = 18), were treated with 0.05% clobetasol propionate gel three times a day for 30 consecutive days and PBM with laser turned off to disguise the treatment and another PBM group (n = 18), were treated with low intensity laser (660nm, 100mW, 177J/cm<sub>2</sub>, 5s, 0,5J per point) twice a week for 30 consecutive days and placebo gel 3 times daily during treatment to disguise the therapy. Blood and saliva were collected on the first and last days of the study to evaluate the expression of cytokines involved and their results after treatment. Shapiro-Wilk normality test, Wilcoxon and Test t were used. Majority of patients at study were female with average of 63.6 years old. Total of 121 OLP lesions were included.

Results: There was no statistically significant difference in the levels of IL6, IL1 $\beta$ , IL10, IL17 and TNF- $\alpha$  cytokines between control and PBM treated groups at the beginning and end of treatment.

Conclusion: In this study, both corticosteroid treatment and PBM treatment got equivalent results, however PBM has no side effects. There is few studies about it in literature and need to be more understood.



#### DIODE LASER EFFECTS ON BOVINE DENTIN REMINERALIZATION

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This study aims to validate a protocol for obtaining caries-affected dentin and the use of diode lasers for treatment purposes, through the in vitro process of demineralization of bovine dentin induced by Streptoccocus mutans biofilm (microbiological method). The validation of the protocol was performed evaluating the optical behavior of caries lesions in dentin irradiated with diode laser in the length 808 nm associated with fluortherapy speckle laser and optical coherence tomography (OCT) system. To perform this study, 48 blocks of bovine teeth were used. The experimental groups were composed from 2 variation factors: cariogenic challenge time (4 days) and dentin type (healthy and affected). After the cariogenic challenge, the specimens were evaluated in their healthy portions and affected by caries using different diagnostic methods and the results were compared. Subsequently, the specimens were initially evaluated with laser speckle and OCT. Specimens were randomly divided into 4 groups (n=12) and treated with acid phosphate fluoride and 808nm wavelength Diode laser. The experimental groups were divided: G1 caries lesion, treated with Diode and fluorine laser; G2 caries and fluoride lesion; G3 caries lesion, treated with fluoride and Diode laser; G4 caries lesion and treated with Diode laser. The analysis to verify the degree of demineralization and remineralization of dentin irradiated with 808nm Diode laser will be performed at the following times: initial, 7, 14, 21 and 28 days. Partial data, initial and 7 days were obtained. A calibrated examiner evaluated all the images obtained through visual inspection and the images obtained by OCT and Speckle laser. The data were tabulated and the statistical test was applied. No statistical differences between groups or times were observed. It was concluded that the proposed treatments for dentin remineralization were insufficient in the evaluation period.



### ANTIMICROBIAL PHOTODYNAMIC CHEMOTHERAPY (PACT) IN SURFACTANT VEHICLE FOR INACTIVATION OF PERIODONTOPATHOGENIC MICROORGANISMS

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Resume: Periodontitis is a chronic and multifactorial inflammatory disease caused by a complex bacterial biofilm infection. It produces host inflammatory response that causes degradation of periodontal tissue and special alveolar bone reabsorption that may compromise teeth viability and general health. Antimicrobial photodynamic chemotherapy (PACT) has been used as adjuvant to conventional treatment; however, literature presents unpredicted result on clinical situations. Aggregatibacter actinomycetemcomitans is a bacterial specie that play an important role in biofilm colonization and it is related to a number of periodontitis case. The aim of this study was to investigate PACT mediated by methylene blue (MB) in surfactant vehicle against A. actinomycetemcomitans. Bacterial suspension was prepared and PACT was performed using 100 µM MB in water solution and the same MB concentration in water with 25% SDS. It was irradiated by 0.1W laser (Photo LaseIII, DMC, São Carlos, Brazil) at 660nm, performing 15, 45, and 75 J/cm2 in a 96 well plate with an area of 0,4 cm2. Controls were performed for all test situations. Following treatments, bacterial suspension was quantified by colony form units (CFU) counts and it was converted to Log10(CFU/mL) for analysis. Bacterial reduction was achieved in both groups following 75 J/cm2 irradiation, with 4 logs of bacterial reduction for MB group and 1 log reduction for MB in SDS group. Under the parameters used in this experiment, the used of SDS in water solution did not improved PACT effect on A. actinomycetemcomitans.

Keyword: Antimicrobial photodynamic therapy, Oral periodontopathogenic, Surfactant vehicle, Periodontits



# EFFECTS OF LOW INTENSITY LASER ON BONE HEALING AFTER MOLAR EXTRACTION IN SPONTANEOUSLY HYPERTENSIVE RATS

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There is an association between hypertensive individuals and reduced bone mineral density. Low intensity laser has anti-inflammatory activity, accelerates the healing of bone defects in vivo and in vitro, stimulates blood flow, recruitment and activation of osteoblasts, osteosynthesis, and decreased osteoclastic activity. Objective: The hypothesis of this work is that photobiomodulation with LBI improves bone repair after exodontia in hypertensive rats. Methods: For the study, 24 female spontaneously hypertensive SHR rats (200-250 g) were used in which the right and left lower molars were extracted. The left alveolus was the control and the right one received the laser therapy according to the following parameters::  $\lambda$ : 808 (± 10) nm, P = 100 mW, time = 40 s, total energy = 4 J. Irradiation was performed over alveolar bone of a recent extracted rat molar with an estimated area of 0,03 cm2 of irradiated area. Irradiance of 3,33 W/cm2 and radiant exposure of 133 J/cm2 was achieved. The power density = 35.7 W/cm2, energy density 1429 J/cm2 was reached on the tip of laser fiber, which has a beam area of: 0.0028 cm2(tab. 1)."Two applications were performed, one in the immediate postoperative period and the other on day 7. On days 7, 14, 30 and 60 the animals were euthanized and the mandible fragments, staining with hematoxylin & eosin and Mallory's trichrome. The samples were evaluated to analyze the presence and type of inflammation, necrosis, bacteria, epithelialization, neoformed bone area and number of osteocytes. Results: The data were evaluated with t-test (SPSS v23, IBM) and there was no statistical difference between the groups in any of the parameters evaluated. Conclusion: It is concluded that under the conditions of the study, the application of LBI did not interfere in the bone repair of alveoli of spontaneously hypertensive rats.



## ANTIMICROBIAL PHOTODYNAMIC THERAPY MEDIATED BY PAPAMBLUE® ON PERIODONTAL DISEASE: RANDOMIZED BLIND CONTROLLED TRIAL

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The aim of this study is to investigate the clinical and antimicrobial effect of papainmediated photodynamic therapy in the clinical treatment of periodontal disease. Twenty patients with periodontitis were selected and randomly divided into two groups (n = 10), the control group received conventional periodontal treatment and group II - conventional treatment with antimicrobial photodynamic therapy (PACT). PACT was performed at the end of each periodontal treatment session, in locations with pockets greater than or equal to 4 mm. The PapaMblue® photosensitizer was inserted into periodontal tissues with a 1 minute syringe prior to irradiation. Then, a wavelength of 660 nm with a power of 100 mW was applied for 2 minutes with radiant exposure 30 J / cm2 and power density I = 250 mW / cm2. The patients underwent clinical evaluations before treatment (day 1), at 30, 60 and 90 days after the end of treatment; and microbiological evaluations before and 30 days after treatment. The data presented a normal distribution and statistical comparisons between and within groups were performed. There were no statistically significant differences in the comparison analyzes between groups. PapaMBlue® PDT is as effective as conventional therapy in subjects selected for this study. The findings of this ongoing study should be considered preliminary and interpreted with caution. In addition, further randomized clinical trials should be conducted to obtain more expressive conclusions.

Keywords: Photodynamic antimicrobial therapy, photosensitizer, laser, antimicrobial



### FLUORESCENCE SPECTROSCOPY OF CANDIDA ALBICANS BIOFILMS IN BONE CAVITIES TREATED WITH PHOTODYNAMIC THERAPY USING BLUE LED (450NM) AND CURCUMIN

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Introduction: Among the innumerable possibilities of technology use that employ fluorescence spectroscopy in the field of health, their applicability is focused on the diagnosis and control of infectious processes associated with bone lesions of the oral cavity. Objective: The aim of this study was to analyze, through fluorescence spectroscopy, Candida albicans biofilms formed in artificial bone cavities treated with photodynamic therapy (PDT) mediated with 450-nm blue light-emitting diode (LED) and curcumin. Another aim of this study was to analyze the existence of a correlation between the effectiveness of the photodynamic treatments and the fluorescence spectroscopy images. Material and methods: Artificial bone lesions (n = 40) were made in bovine bones and inoculated with standard suspensions of Candida albicans (ATCC 18804) for biofilm formation (14 days / 36 °C ± 1 °C). The 40 specimens were distributed among four experimental groups (n = 10): L-C- (control), L+C- (LED for 5 min 67mW/cm<sup>2</sup>, 20,1J/cm<sup>2</sup>), L-C+ (curcumin for 5 min), and L+C+ (PDT). Aliquots of 100 µL were collected from the bone cavities after treatments and seeded in duplicate on Sabouraud dextrose agar for 24 h at 36 °C ± 1 °C and the colony-forming units (CFU/ mL) were counted. Before and after each treatment, the specimens were subjected to spectral fluorescence and the images were compared using the Image J program. The log10 CFU/mL were compared with Kruskal-Wallis and Dunn's Multiple Comparison post-test (significance level at 0.05). The fluorescence histogram values before and after treatment were compared using Wilcoxon test (95%). The correlation between Candida albicans log10 CFU/mL and the number of the fluorescence red pixels spectroscopy was verified using Spearman correlation test. Results: The reduction of Candida albicans log10 CFU/mL in the L+C+ (PDT) group was the most relevant and the fluorescence spectroscopy was correlated to the microbiological result. Conclusion: It was concluded that there was a consistency between the log10 CFU/mL of Candida albicans and the red pixel data of the fluorescence images, demonstrating that the fluorescence diagnostic device reflects the true microbiological condition of Candida albicans biofilms in the bone cavities during the pre-treatment and post-treatment, thus providing the clinician the ability to dynamically, simply, and instantaneously verify the performance of the treatment used.

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## STUDY OF THE LARVICIDE ACTIVITY OF CURCUMINE BY PHOTODYNAMIC THERAPY IN DIFFERENT CONCENTRATIONS AND BLUE LED (460NM) ABOUT WILD AEDES AEGYPTI'S MOSQUITOES

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Introduction: In the first guarter of 2019, there were 273.193 confirmed cases of dengue in Brazil, which represented an increase of 74% over the same period in last year. Also in 2019, in the same period, 15.352 probable cases of chikungunva were register, with 2 deaths confirmed (1 in Bahia and 1 in Rio de Janeiro) and 14 deaths are under investigation. As for Zika, in this period, 2.344 probable cases were register in the country, with the North and Southeast being the regions with the highest number of probable cases. It is notorious that current policies for vector control of these arboviruses are not effective. Studies of the use of light-activated photosensitizers as an alternative to conventional insecticides for sustainable control of vector mosquitoes such as Aedes (dengue, yellow fever, chikungunya), Anopheles (malaria), Culex (yellow fever) can already be found, showing advantages over conventional insecticides (efficacy, safety, non-mutagenicity and fast degradation). Objective: The aim of this study was to evaluate the effectiveness of curcumin-mediated photodynamic therapy (PDT) and blue LED (460nm) on mortality of wild larvae of Aedes aegypti's mosquitoes. Material and methods: Traps were place in strategic regions identified by the LIRA issued by the Secretariat of Epidemiological Surveillance of the city of Vitória da Conquista-BA. The larvae collected in stages L2 and L3 were feed for 24h with curcumin at concentrations of 10, 20 and 50% mixed with fish food and then underwent blue LED irradiation for 2h (22mW/cm2 and 158.4 J/cm2). The larvae were place in a container with dechlorinated water and mortality was followed for 24 and 48h. Control groups in which only larvae were expose to blue LED for 2h and where larvae were only fed with curcumin mixed feed (10, 20 and 50%) were included in the study, in addition to the untreated group. All experiments were repeated after a 2-month interval for confirmation of the results. totaling 256 tests (tests 1 and 2, n = 16) between the PDT 10%, PDT 20%, PDT 50%, curcumin 10%, curcumin 20%, curcumin 50%, blue LED and untreated group. Results were compared by ANOVA test (5%). Results: All PDT groups showed larvicidal activity (p < 0.05). Conclusion: The PDT in all concentrations was effective in larval mortality, with the concentration of 20% curcumin showing the best activity with mortality of 100% in 24h. Funding sources: This study was financed in part by the Coordenação de Aperfeicoamento de Pessoal de Nível Superior - Brasil (CAPES) - Finance Code 001 and National Institute of Science and Technology of Basic and Applied Optics in Life Sciences (IFSC-USP).

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## LEVEL OF PAIN, MUSCLE STRENGTH AND BALANCE IN WOMEN WITH KNEE OSTEOARTHRITIS AFTER A SENSORY-MOTOR EXERCISE PROGRAM AND PHOTOBIOMODULATION THERAPY VIA CLUSTER

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Background: Osteoarthritis (OA) is the most common chronic condition of the joints that leads to pain, stiffness and disability. The objective was to determinate the effectiveness of a sensory-motor exercises program (SMEP) and the PBM (associated or not) on knee flexor and extensor muscle strength, severity of injury, static balance, dynamic balance and pain of women with knee osteoarthritis (KOA). Methods: Thirty-two subjects with KOA were randomized in two groups: SMPG: Sensory-motor exercises associated with placebo PBM group (n=16) and SMAG: Sensory-motor exercises associated with active PBM group (n=16). The knee flexor and extensor peak torque, severity of injury, static balance, dynamic balance and pain were evaluated with the use of cell roll, Knee Injury and Osteoarthritis Outcome Score (KOOS), Y balance test, timed up and go (TUG), and numeric pain rating scale (NPRS), respectively. Interventions were realized with sensory-motor exercises (such as retrowalking, walking on their toes, leaning to the sides, balance board exercises) in addition to strengthening exercises, and PBM via cluster (808 nm, 100mW, 7 points each side, 56 J total) immediately after the end of each training session during 8 weeks, 2 times/week.

Results: No significant increase in the knee flexor and extensor peak torque was obtained. Although, after treatment, there was linear positive and significant correlation only in SMPG (p=0.002). The improvement in severity of injury (p=0.002), dynamic (p=<0.001) and static (p=<0.001) balances were significant statistically in both groups. And, the measurement of level of pain showed a significant improvement only in SMAG (p=0.043).

Conclusion: SMEP have beneficial effects on severity of injury, static and dynamic balance. The addition of PBM therapy is a resource that promotes extra effects in pain relief. Adding PBM to routine exercise regimes for patients with KOA should be investigated.



# BIOCHEMICAL EVALUATION OF THE BLOOD OF RATS WITH MUSCULAR INJURY SUBMITTED TO SYSTEMIC PHOTOBIOMODULATION

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Introduction: Photobiomodulation (PBM) is able to control the inflammatory process and improve the tissue repair when used on muscle injuries. This therapy, when applied to the bloodstream, is called systemic photobiomodulation (SPBM) and has been used to treat many diseases, but little is known about its effects on muscle repair. Objective: The aim of this study was to evaluate the biochemical markers (creatinine and urea) present in the blood plasma of rats submitted to injury and SPBM (preventive or therapeutic). Material and methods: Fifty male Wistar rats were used (CEUA - no 8446031018). The animals were randomly assigned to the following experimental groups: control: not subjected to cryoinjury or SPBM (n = 5); injury + simulated SPBM (injury): subjected to cryoinjury of tibialis anterior (TA) muscle and application of SPBM with the device turned off (n = 15); SPBM + injury: subjected to SPBM before TA muscle injury (n = 15); and injury + SPBM: subjected to TA muscle injury and subsequently treated with SPBM (n = 15). The SPBM was applied punctual in contact mode on the tail of the animals. The following parameters were used: 780 nm, 40 mW, 1 W/cm2, 10 J/cm2, 80s, 3.2 J. SPBM was performed once and started 24 h before the injury (SPBM + injury), or 2 h after the injury (injury, injury + SPBM), and was performed at 24 h intervals, totaling 1, 2, or 5 sessions. Five animals were sacrificed on day 1, 2 and 5 after the injury, and blood was collected for biochemical analysis. Data distribution was verified using the Kolmogorov-Smirnov test; the significant difference between groups was calculated using ANOVA and the Tukey test. Results: The results indicated that at 2 days, the treated groups (SPBM + injury and injury + SPBM) presented lower creatinine values than the injury group. At 5 days, there were significant differences between the treated groups, and the SPBM + injury group presented a lower amount of creatinine when compared to the other groups. Regarding the urea, the significant difference occurred at 2 days, being higher in the injury group than in the other groups. Conclusion: The results showed that the injury group presented data indicating protein catabolism at 2 days, while in the SPBM + injury group, a modulating effect on this process was observed.

Funding source: UNINOVE



## ASSOCIATION OF PHOTOBIOMODULATION WITH INTRADERMAL PLATELET-RICH PLASMA INJECTION FOR FACIAL REJUVENATION: DOUBLE-BLIND, RANDOMIZED CONTROLLED TRIAL PROTOCOL

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Introduction: Skin aging is an irreversible, slow and progressive process, mainly influenced by age, but also by external factors such as ultraviolet radiation, smoking, and alcohol, among others. It is increasingly common to look for procedures that slow down this process by limiting or hiding its effects on the skin and its appearance. Intradermal platelet-rich plasma (PRP) injections are a new modality of treatment with possibly beneficial effects on skin rejuvenation. In addition, studies have also shown that photobiomodulation (PBM) benefits the skin. Objective: To investigate the effects of combining the use of intradermal PRP injections on the face in conjunction with photobiomodulation for facial rejuvenation. Material and methods: A randomized, double-blind, controlled clinical trial will be conducted from October 2019 to January 2021, with voluntary participants who wish to improve facial aesthetics, treated at the Nove de Julho University (UNINOVE) Medical Specialties Outpatient Clinic in the city of Sao Paulo, Brazil. Participants will be divided into three groups: one group will undergo intradermal application of PRP in association with PBM; another group will undergo intradermal application of PRP and a placebo light application and a third group will receive an intradermal application of 0.9% saline (placebo) associated with PBM. Only one application of PRP or saline will be injected at defined points on the face. PBM sessions will be held immediately after the first application of PRP or saline is administered, 3 days after the first application and weekly for the following 3 weeks. The following variables will be studied for further analysis: face moldings analyzed by optical coherence tomography; skin viscoelasticity; histological study of the dermis; comparison, by plastic surgeons, of the evolution seen in photographic images (Wrinkle Assessment Scale) and participants' satisfaction level (FACE-Q). All data will be statistically evaluated according to their distribution. Rationale: PRP injection has gained notoriety in recent years as it is described as a non-immunogenic, low-cost procedure with minimal complications. On the other hand, the literature has described PBM as able to promote platelet activation, release of growth factors and modulation of the extracellular matrix also leading to positive results in facial rejuvenation. Thus it seems reasonable to suppose that combining these techniques together could have a synergistic effect. However, it was not possible to find any studies in the literature on the effects of the association of PRP with PBM on facial rejuvenation.

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## ANTI-INFLAMMATORY EFFECT OF PHOTOBIOMODULATION IN AN EXPERIMENTAL MODEL OF CHRONIC OBSTRUCTIVE PULMONARY DISEASE (COPD) CAUSED BY SMOKING: PARTICIPATION OF TREG CELLS.

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Introduction: Currently, COPD has a high impact on morbidity and mortality worldwide. The smoking habit, smoking history, airflow rate, obstruction and pulmonary emphysema are all related to the increase in CD8+ cells and/or CD8+/CD4+ ratio, the number of these CD4+, CD8+ cells express NF-kB, STAT4, IFN-y and perforin. A deficiency in T cells regulating CD4+CD25+Foxp3+ (Tregs) may impair the tolerance of the imune system and thus lead to immune disease. Thus, immunotherapeutic strategies, such as photobiomodulation (PBM), aim to regulate the levels of cytokines, chemokines and transcription factors. Objective: In this sense, the objective was to evaluate Foxp3 Treg cells as well as the production of IL10 in the lung after PBM in COPD model. Material and methods: We studied some parameters in C57BL/6 with COPD submitted to diode laser therapy (660nm,30mW, 180s) for 15 days. The protocol used for the induction of COPD consisted in the application orotracheal of extract of cigarette smoke (3x per week/7 weeks). Bronchoalveolar lavage (BAL) and lungs were collected for analysis. Data were submitted to the One-way ANOVA test followed by the Newman-Keuls test. Significance levels adjusted to 5% (p<0.05). Results: PBM reduced the number of inflammatory cells, the levels of IL1-β, TNF-α, IL-6, IFN-γ, MCP-1, GM-CSF (p<0,001), KC/CXCL1, LTB4 (p<0,01) and increased IL-10 (p<0,001). We also observed decrease of collagen, mucus, bronchoconstriction index, alveolar enlargement, CD4+, CD8+, CD4+STAT4+ and CD4+IFN-γ+. There was increased CD4+CD25+Foxp3+ and CD4+IL-10+. Discussion: Our results show that PBM reduces pulmonary inflammation, characterized by cellular quantification, CD4+CD25+Foxp3 and CD4+IL-10 in BAL, GM-CSF, MCP1, IL-1β, IL-6, IL-8 levels. , IL-10, TNF- $\alpha$ , IFN- $\gamma$  and LTB4, remodeling as well as alveolar enlargement demonstrating a promising role for the treatment of COPD. Conclusions: Thus, PBM can be used as immunotherapeutic strategy for the treatment of COPD through the possible mechanism of CD4+CD25+Foxp3+ e CD4+IL-10+.

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# EFFECT PHOTOMODULADOR OF RED AND INFRA RED LIGHT ON GROWTH OF RHODOTORULA SPP

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Introduction: Onychomycosis is a type of mycosis that affects nails and it is difficult to be diagnose and to be treated. Within this classification, have Rhodotorula spp being an emerging yeast responsible for causing opportunistic infections in humans. High frequency waves are widely used in aesthetic clinics and can help in the treatment of onychomycoses, because generated ozone and have fungicidal and bactericidal effect, can also stimulated the circulation of the place where it is applied and its vasodilatory function can be facilitates the penetration of systemic drugs. Another alternative is phototherapy (Low Level Laser Therapy), which is currently using in various areas of health. Objective: The aim of this study was to evaluate in vitro the effects photomodulator of red and infrared light on growth of Rhodotorula spp. Material and methods: The yeast from this work was isolated from humans by Dermatology Department from UNIFESP. For growth was used Sabouraud Agar in a petri dish and after 5 days, the light was applied (DMC-Therapy XT equipment) with a wave emission of 650 and 808nm with a power of 30mW and a dose of 3J/cm2 for 30 seconds per application, being done at 5 and 7 days. The application was made at 1cm for the surface of the plate. After periods of 2, 5, 7 and 12 days, photos and measurements of the area were performed by the Image J software. Statistical analysis t-test were performed in 3 repetitions. Results: Rhodotorula spp is a yeast, whose cells are spherical, ovoid producing pigment orange to red. In the period from 3 to 12 days, yeast in control plates grown 6cm2 of area. However, yeast plates that received red and infrared light the growth was paralyzed indicating that the laser acted as a modulator in period from 5 to 12 days. In addition, it was possible to verify the depigmentation of the colony in plates yeast+laser. Conclusion: The red and infrared laser under the methodological conditions showed effects on the cellular metabolism of Rhodotorula spp as the change in colony coloration and growth rate in vitro.



## PHOTODYNAMIC THERAPY: EFFECTIVENESS AND MOLECULAR MECHANISMS OF CURCUMIN ACTION AS A PHOTOSENSITIZER AGAINST PSEUDOMONAS AERUGINOSA BIOFILM

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Introduction: Pseudomonas aeruginosa is a gram-negative bacterium with high capacity to form dense biofilm, a complex structure which predisposes to development of antibiotic resistant phenotypes, contributing to healthcare-related infections becoming a serious worldwide public health problem. Objective: This is an in vitro experimental study, which has aimed to evaluate the antimicrobial effectiveness and molecular mechanisms of curcumin action at concentrations of 5 µg / ml, 20 µg / ml and 40 µg / ml irradiated by blue LED light 450 nm at energy doses of 3.24 J / cm<sup>2</sup>, 5.4 J / cm<sup>2</sup> and 10.8 J / cm<sup>2</sup>, against to Pseudomonas aeruginosa monospecies biofilms. Material and methods: Experimental groups treated with PDT (Photodynamic Therapy) had had their results compared among them with negative control group (without treatment) and positive control groups (LED and curcumin, alone), totaling 16 experimental groups. To evaluate the antimicrobial effect, the count of colony forming units per ml (CFU / ml) has been transformed to Log10. checking the assumptions of normality and homogeneity, with statistical analysis performed by Anova (p value <0.05) and Kruskal-Wallis (p value <0.05), with Bonferroni and Dunn post-test, respectively. The study of biofilm structure was based on images generated by LASER confocal scanning microscopy (CSLM) and, by Raman microspectroscopy, molecular mechanisms of curcumin action through peaks observed in spectrum have been evaluated. Results: The results have demonstrated an action centered on the disruptive effect of biofilm, as well as an antimicrobial effect in all PDT protocols, with more expressive results observed in protocols with higher photosensitizer concentration and higher energy dose. Conclusion: Curcumin-mediated PDT evaluated in that study has proved effective not only in reducing, in a single session, 1.3 log10 in bacterial cell viability, corresponding to a percentage reduction of more than 90% in CFU / ml count, but mainly due to its antibiofilm effect and may possibly be used as a potentiator of antimicrobial action.

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## EVALUATION OF DAILY PHOTOBIOMODULATION IN STEM CELL POPULATIONS OF ORAL SQUAMOUS CELL CARCINOMA CELL LINES

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Introduction: Studies have demonstrated that Oral Squamous Cell Carcinoma have a subpopulation of cells called as cancer stem cells (CSC), which are responsible for tumor growth, metastasis and therapeutic resistance. CSC show different phenotypes based on the CD44 and ESA, which are described as the mesenchymal CD44high/ESAlow phenotype with invasive capability and the epithelial CD44high/ESAhigh phenotype. The conventional treatment for OSCC is surgery associated with radio and chemotherapy, both associated with oral mucositis (OM). Some studies have demonstrated that Photobiomodulation (PBM) can act preventing the development of mucositis as well as reducing pain and promoting wound healing, mainly when applied daily in oral mucosa. However, some studies demonstrated that PBM promote tumor progression. Objective: The aim of this study was to evaluate the effects of daily PBM in the CSC phenotype of OSCC cell lines. Material and methods: OSCC cell lines (SCC9, Luc4 and CA1) were cultivated in DMEM/F12 with 10% FBS and supplements. Cells were irradiated using the LEDbox in direct contact mode, wavelength 660nm, power 80mW, power density 25.5mW/cm<sup>2</sup>, energy density of 3 and 6J/cm<sup>2</sup> with 120 and 240 sec and total energy of 9.6 and 19.2 J. respectively, for 3 consecutive days. After PBM, cells were collected and stained with CD44 and ESA and RNA were extracted to evaluate the expression of BMI1, POU5F1 (OCT4), IVL(Involucrin), S100A9 (CalgranulinB), SNAI2 (SLUG) and GAPDH by RT-gPCR. Data distribution was verified by Shapiro-Wilk normality test and significance difference between groups was calculated using T-Test and Mann-Whitney tests. Results: No difference between the CD44high/ESAhigh, CD44high/ESAlow and CD44low fractions was observed with 3 and 6J/cm<sup>2</sup> in relation to control. In addition, daily irradiation with 6J/cm<sup>2</sup> was associated with a significant decrease in the BMI1 mRNA expression. Conclusion: PBM with 3J/cm2 and 6J/cm2 (doses used to treat mucositis) showed no influence in the CSC phenotype in OSCC cell lines and was also associated with a decrease in the expression of BMI1 gene, responsible to maintain CSC self-renewal. Additional studies investigating the effects of PMB in CSC are necessary to understand its role in OSCC.

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### EFFECTS OF LOW-INTENSITY LASER ON VENOUS ULCER HEALING CUNHA LP1,\*, NOGUEIRA GA1, MARTINS CJ1

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Introduction: The literature indicates that among the ulcers found in the lower limbs, the ulcer of venous etiology is the one that has the highest prevalence, corresponds approximately from 80 to 90% of the ulcers found in this location, and the insufficiency Chronic venous disease is the main responsible for its emergence. The conventional treatment for patients with venous ulcer is the use of coatings and elastic compression. Studies point out that low intensity Laser (LBI) assists in the treatment of this lesion by stimulating tissue healing and pain control. Objective: The aim of this study was to evaluate the effect of low-intensity laser on venous ulcer patients. Material and methods: The patient COX, 80 years old, Caucasian, female. Medical diagnosis: Arterial hypertension. She presented as the main complaint pain in the area of the lesion and was in the lower right third. The same was irradiated using Laser Therapy XT, wavelength 660 nm and 808 nm, power of 100 mW, power density of 1.02 mW/cm<sup>2</sup>, Creep of 20 J/cm<sup>2</sup>, 20 sec and total energy of 2 J, in direct contact, for once a week. After 06 Irradiations in the wound presented complete healing of the area. Results: The lesion presented complete healing and decreased pain after LBI with 2 J. The patient had no adverse effects after the application of LBI. It was used in the perilesional NDerm skin and as hydrogel and alginate cover. Conclusion: The LBI proved to be effective in healing with 2 J, with analgesic effect and without the presence of adverse reactions, constituting a biostimulative and noninvasive therapy. In addition to the techniques employed in biophotonics are highly efficient and selective, with minimal reports of adverse events, it is necessary to develop research in this area.



### SURVIVAL IN ESCHERICHIA COLI CULTURES AND PLASMID DNA EXPOSED TO AMBER LED

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Introduction: Low power non-ionizing radiations, in infrared up to ultraviolet range of electromagnetic spectrum, are used for therapeutic purposes. For this, LEDs (light emitting diodes) and lasers are successful for pain relief and tissue regeneration. However, some studies have suggested that these radiations present sublethal effects on cells deficient in DNA repair mechanisms. Objective: The objective of this study was to evaluate effects of low power amber LED radiation on Escherichia coli cultures, proficient and deficient in DNA repair mechanisms, and on plasmid DNA. Material and methods: E. coli cultures AB1157 (proficient), JW1625 (deficient in endonuclease III), BH20 (deficient formamidopyrimidine DNA glycosylase - fpg) were exposed to amber LED radiation (617 61.7nm) at different fluences (160, 320 and 640 J/cm2, 30, 60 and 120s, respectively, spot size of 0.28 cm2, 1500mW), diluted, spread onto Petri dishes containing nutritive medium, incubated (18h, 37 °C), forming colony units were counted to calculate survival fractions and bacterial colonies were photographed to analyses by ImageJ software. Also, plasmids were exposed to amber LED radiation at same fluences, applied in agarose gels to electrophoresis procedure. After that, the gels were photographed and plasmid topological forms were analyzed by ImageJ. As controls, bacterial cultures and plasmids not exposed to amber LED radiation. Groups were compared by ANOVA and Bonferroni post-test considering p<0.05 as low level of significance. Results: Our data suggest no statistically significant alteration of survival fractions in proficient (AB1157) and deficient (JW1625 and BH20) strains in exponential and stationary growth phases. Plasmid topological forms were not modified, as well as, analysis of colony areas showed no statistically significant alteration in AB1157 in both growth phases and BH20 in exponential growth, but there was reduction of colony areas in BH20 stationary growth at all fluences, and in JW1625 at higher fluence in both growth phases. Conclusion: Although there is no cell viability and plasmid topological alterations, exposure to low power amber LED radiation could decrease cell division in E. coli cultures deficient in repair of oxidative lesions in DNA.

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### PHOTOBLEACHING OF METHYLENE BLUE IN BIOLOGICAL TISSUE MODEL (HYDROLISED COLLAGEN) USING 635NM IRRADIATION CONDITIONS

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Introduction: Methylene blue (MB) is used for Photodynamic Therapy (PDT), with a protocol which is efficient, inexpensive and also safely, used for decades in clinical applications. The quantification of efficacy of a photosensitizer in aqueous solution is well known in literature. Photobleaching, a photoinduced degradation or modification of photosensitizer, can procedures significant lack of efficacy in PDT treatment, since the generation of reactive oxygen species is directly associated with photons absorption of this photosensitizer. Objective: To study the photobleaching kinetics of MB in media that simulates the structured medium like biological tissue with hydrolyzed collagen. Material and methods: The samples were prepared with 0.1 mg/ml of hydrolyzed collagen in Milli-Q water, at different concentrations of methylene blue (50µM, 75µM, 100µM, and 125µM). The samples were placed in acrylic cuvettes (optical path length = 10mm). For the determination of photobleaching kinetics, the samples were irradiated with a collimated diode laser beam (Coherent-USA) with initial power (Po) of 7 mW and wavelength of 635nm, near MB visible absorption maximum (665nm). Temporal Transmitted power (Ptr) through the different concentrations of methylene blue in collagen-like tissue was measured by a powermeter (Fieldmax II Coherent) as a function of irradiation with laser. Data were analyzed using the Origin software using the function  $Ptr = P0^{*}(1$ e-Kt), where (in s-1) is the temporal photobleaching kinetic constant under different MB concentrations, Results: Photobleaching of MB in collagen like-tissue model due to irradiation can permits the increased light passage through samples, and these increased light were time- and MB-dependent. The rates constant in different MB concentrations obtained decreases with the concentration of methylene blue increases: 50µM = 605 s-1; 75µM =420 s-1; 100µM =395 s-1 ; and 125µM =350 s-1. Therefore, photobleaching's MB rates decreased as the concentration of methylene blue increased. Conclusion: These results indicate that the rates of the temporal photobleaching kinetics of methylene blue under 635nm laser irradiation (P0= 7mW) are proportional to the photosensitizer concentration. Further studies are underway, where we are increasing the complexity of the model to better understand photobleaching mechanisms. These results can helps in clinical conditions PDT treatments.

Funding sources: UFABC.

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Key words: Methylene Blue, Photobleaching, Photodynamic Therapy.



# EFFECT OF PHOTOBIOMODULATION IN BONE QUALITY OF BOVINE GRAFT IN ALVEOLUS OF RATS POST DENTAL EXTRACTION

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Introduction: The bone graft added to the dental alveolus immediately after tooth extraction prevents atrophy and deformity of the bone at the site of the loss of the element, enabling the rehabilitation with implants. Photobiomodulation accelerates bone healing process defects; stimulate blood flow; activate osteoblasts, promote osteosynthesis; decrease the osteoclastic activity and improve the integration of the scaffold material with the bone tissue. Improvement in the quality of the bone graft provides the primary stability and longevity of the implants in oral rehabilitation. Objective: The objective was to evaluate bone healing process in male Wistar rats submitted to molar extraction with or without bone grafting with lyophilized bovine bone and to verify the therapeutic effect of photobiomodulation with LED 850 nm in bone remodeling Materials and Methods. 48 animals were divided into 5 groups with 12 animals each, baseline G1 and G2 control (only molar extraction), G3 Exo / LED (carried out the extraction and treatment with LED  $\lambda$  = 850 nm, power = 100 mW, exposure time = 60s, energy = 6J, radiant exposure =  $30 J / cm^2$ , 1 spot irradiated 8 sessions 48/48 hours for 15 days), G4 Exo / Enx (performed the extraction and alveolus graft with lyophilized bovine bone Straumann® cerabone® 0, 5-1.0 mm, 1x 0.5 cc) and G5 Exo / Enx / LED (carried out the extraction, alveolus grafting with the biomaterial described above and treatment with LEDs in the same parameters described above). The animals were euthanized in 15 and 30 days. Histological analysis, optical coherence tomography, micro tomography and acid and alkaline phosphatase of the bone repair will be presented at the conference.

Results and conclusion: Study carried out and ongoing analyzes for respective data presentation and conclusion of the project at the International Biophotonics Forum (FIB).

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### UVA DETECTION BY MELANOPSIN REGULATES PROLIFERATION, PIGMENTATION, AND MOLECULAR CLOCK IN NORMAL AND MALIGNANT MELANOCYTES

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Introduction: UVA radiation can be sensed by several organic molecules and light sensitive proteins known as opsins. Melanocytes and melanoma cells express several opsins, of which melanopsin (OPN4) has been demonstrated to detect both temperature and UVA radiation. Although the effects of UVA radiation on skin are widely known, how opsins detect UVA and the physiological processes triggered by this detection are still largely unexplored. Objective: To evaluate whether daily stimulation of UVA radiation affects cell proliferation and death, melanin content, and molecular clock in normal and malignant melanocytes in the presence and absence of Opn4 (CRISPR knockout). Material and methods: Opn4+/+ and Opn4-/- normal and malignant melanocytes (Melana and B16-F10 cells, respectively) were seeded in 6-well plates at 37oC. At the beginning of the 4th day, cells were daily irradiated with UVA radiation (4.4 kJ/m2, 1.6 mW/m2 during 50 min) for 3 days, and were harvested immediately, 24, and 48 h after the end of the last stimulus. Cell death was determined by Trypan Blue exclusion and propidium iodine staining in flow cytometry. DNA damage was assessed by gama-H2AX staining, and melanin content by total absorbance at 475 nm. Molecular clock expression was assessed by real time bioluminescence in cells harboring Luciferase gene under the control of Per1 promoter, a clock gene. Results: In Opn4+/+ normal and malignant melanocytes, a reduction on cellular proliferation was observed, 24 h after the end of UVA radiation, in comparison with non-irradiated Opn4+/+ control cells, what was related to cellular death only in malignant cells. In both cell types, UVA radiation led to a persistent pigment darkening (PPD), and no DNA damage was observed. Upon knockout of Opn4 in normal and malignant melanocytes, all the above-effects were abolished in three independent Opn4-/- clones. As to clock genes, a reduction on bioluminescence was found in Opn4+/+ normal and malignant cells after each UVA stimulus. In the absence of Opn4, a bioluminescence increase was evident in both cell types. These data show that the molecular clock is repressed and activated in response to UVA radiation, in the presence and absence of Opn4, respectively. Conclusion: This is the first report that shows UVA radiation sensing by OPN4 in normal and malignant melanocytes, which mediates UVA-induced decrease of cellular proliferation, and activation of pigmentation and molecular clock. Therefore, OPN4 is an interesting pharmacological target in skin biology, and warrants further investigation.

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### LOW-LEVEL LASER THERAPY MODULATES MUSCLE APOPTOTIC SIGNATURE IN RATS SUBMITTED TO HIGH-INTENSITY RESISTANCE EXERCISE

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INTRODUCTION: Acute high-intensity resistance exercise (HIRE) is characterized by inflammation, oxidative stress and muscle damage. From the lesion derives mainly necrosis, but there are studies that attribute importance to apoptosis. In this regard, photobiomodulation with low-level laser therapy (LLLT) has been used as a tool to attenuate cellular necrosis, but there are no studies that have applied LLLT to evaluate its action on HIRE-triggered muscle apoptosis. Therefore, this study aimed to analyze the effect of LLLT on muscle apoptosis in rats submitted to a session of HIRE. MATERIALS AND METHODS: Wistar female rats, 9-12 weeks old, weight of 180- 200 g, were distributed to one of the following experimental groups: Control (CTRL) - Placebo laser (device off) and not exercised; Exercise (EXE) - placebo laser and exercised; LLLT + Exercise - DMC Thera Laser equipment at 830 nm was applied on both legs at three points of the gastrocnemius muscle with 2J, 4J and 8J, respectively. LLLLT or placebo were carried out 60 sec prior to RE that consisted of four climbs bearing the maximum load with a 2 min time interval between each climb. Rats were euthanized immediately after the HIRE and the muscles was collected to analyze by Western blot of BCL-2, BAX, BAD, total AKT, phosphorylated AKT and cleaved Caspase 3. RESULTS AND DISCUSSION: No significant differences were found between the groups in the study of proteins: BAX, BAD, total AKT and phosphorylated Akt. BCL-2 protein expression was similar between the CRTL and EXE groups, but protein content was increased when comparing 2, 4 and 8J groups with the CTRL group. Cleaved Caspase 3 showed similar results between CTRL and 4J group. CONCLUSION: LLLT with 4J irradiation attenuated muscle apoptosis induced by HIRE in rats.

Key words: photobiomodulation, apoptosis, resistance exercise, low-level laser therapy



### THE INCREASE IN RADIANT EXPOSURE DO NOT INCREASE METHYLENE BLUE PDT ANTIMICROBIAL ACTION IN CANDIDA ALBICANS PLANKTONIC CULTURE WHEN LOW IRRADIANCE IS USED

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Introduction: The standard antifungal treatment is performed with the use of a medicine which may cause hepatotoxicity and microbial resistance. Given this scenario, photodynamic therapy (PDT) is a promising therapy which may be applied locally. It is based upon the use of a photosensitizer and a light source with the corresponding excitation wavelength, such as Laser and LED, generating reactive oxygen species responsible for cell death. However, there are many parameters that may influence the result of the therapy, such as the media for the photosensitizer agent administration, light exposure, irradiance, and others. Objective: The objective of this study was to evaluate the effect of PDT in different radiant exposures in C. albicans planktonic culture. Material and methods: C. albicans was cultured in sabouraud dextrose agar at 37°C for 48 hours. After growth, the cells were harvested, counted and 0.200mL of the inoculum (5x107 cells/mL) was plated in a 48-well plate with its respective treatment (0.200mL). The groups were divided into: control(CT); MB (MB 20mg/L aqueous solution); CTF - oral formula without MB; MBF - MB 20mg/L in oral formula. Subsequently, the samples were homogenized and incubated in the dark for 5 minutes. After incubation, the plate was submitted to light exposure (659 ± 9nm, 3.7mW/cm2) and 30, 40, 50 and 80 minutes (reaching 6.7; 8.8; 11.1 and 17.8 J/cm2). At the end of the irradiation, the samples were diluted in ultra pure water (10-1 to 10-5). The dilutions were spread in sabouraud dextrose agar and placed in an incubator at 37°C for 24 hours. After this period, colonies were counted (CFU/mL). Results: The MB group showed no reduction in relation to CT. CTF and MBF, exhibited a 1log10 reduction in relation to the CT. These results were observed in all the radiant exposures used. Therefore, PDT has no effect in Candida albicans culture when using 3.7mW/cm2 irradiance. Discussion: PDT with MB has been extensively studied as antimicrobial agent, however, frequently, it presents no desirable effects. The parameters of the therapy are essential for the achievement of antimicrobial effect. In here, we could show that increasing the time of exposure is not enough to reach some reduction in microbial growth, when using low irradiance. Conclusion: PDT did not reduce C. albicans colonies when low irradiance was used (3.7 mW/cm2) independently of the exposure time used. Further studies are needed to better evaluate the use of this therapy, and find a minimum irradiance to reach some microbial growth reduction.

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## ANALYSIS OF PHOTOBIOMODULATION ENFORCEMENT ON HEALING SKIN WOUNDS INDUCED IN MALNOURISHED RATS

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Photobiomodulation (PBMT) has been indicated for enforcement on healing skin wounds through the increase of mitotic activity, number of fibroblasts, synthesis of collagen and neovascularization, however the appliance of PBMT under unfavorable conditions, such as protein malnutrition, has not been fully studied. Therefore, this study aimed to verify the effects of PBMT applied for healing skin wounds, in proliferation phase, in malnourished rats – in hypoproteic diet. This study sample was composed for 40 male rats, age 12 weeks, of which 20 with 25% of protein considerated normal rate, and 20 rats on hypoproteic diet (8%). The rats composed four experimental groups (n = 10), they were submitted to skin wound through 8 mm punch, and then treated with PBMT (low level laser therapy - LLLT), parameters: 660nm, 50 mW, 1.07 W/cm2, 0.028 cm2, 72 J/ cm2, 2 J. The following analyses were performed at 7 and 14 days of tissue repair process: semi-quantitative histopathologic analysis, differentiation of the percentage of collagen type I and III by polarization of histological slides stained by picrosirus red and immunohistochemistry marking of MMP-3, MMP-9 fibronectin ,VEGF and mechanical resistance test in universal testing machine. The obtained results demonstrated significant differences between the normoproteic groups with injury and their respective treated groups (p<0.05), as well as in relation to treated and untreated hypoproteic groups p<0.001in histopathologic analysis semi-quantitative and immunohistochemistry for metalloproteinases 3 and 9 demonstrated that PBMT was able to decrease immunostaining with a statistical difference of p < 0.01. There was also decrease in collagen deposition process with statistical difference of p < 0.05 for both collagen types I and III. We can conclude that LLLT - PBMT application was proved effective in the treatment of cutaneous wounds in rats submitted to a hypoproteic diet, in different phases of tissue repair process, however, these alterations were shown to be more salient in the proliferation phase with increased blood vessel formation and granulation tissue, besides the reduction of metalloproteinases providing better mechanical resistance of the injured area in the remodeling phase with intensification of type I collagen.

Key-words: skin wounds ; photobiomodulation ; low level laser therapy.



### COMPARISON BETWEEN PHOTOBIOMODULATION THERAPY (PBMT) AND A COLD-WATER IMMERSION (CWI) IN RECOVERY SOCCER PLAYER: ANALYSIS OF BIOCHEMICAL MARKERS OF RECOVERY AND MUSCULAR OXIDATIVE STRESS.

#### DENIS SMITH, PAMELLA GABRIELA DOS SANTOS FERREIRA, SOLANGE ALMEIDA DOS SANTOS, JHENIPHE ROCHA CAIRES, PAULO DE TARSO CAMILLO DE CARVALHO.

This study aimed to investigate and analyze markers muscle damage and oxidative stress in soccer athletes after a match submitted to recovery strategies through cold water immersion CWI and Photobiomodulation therapy (PBMT). Sixteen male highlevel soccer players were recruited in this randomized, crossover, controlled clinical trial. After randomization of the volunteers, two groups of five (5) athletes were distributed. Two interventions were performed, one being CWI and the other PBMT irradiation was performed at 17 sites of each lower limb, employing a cluster with 12 diodes (4 lasers diodes of 905 nm, 4 light emitting diodes [LEDs] of 875 nm, and 4 LEDs of 640 nm, 30 J per site), 15 minutes after the end of the soccer match. Blood samples were then collected to determine the markers of muscle damage CK, LDH, and levels of oxidative damage to lipids and proteins, the activities of the TBARS, antioxidant enzymes SOD and CAT at the end of the interventions and after 48 hours of soccer match 1. After 7 days in soccer match 2 the inversion of the ducts between the groups was performed. Interventions were performed after 15 minutes at the end and 48 hours of soccer match 2. PBMT decreased CK activity (p < 0.05) from immediately and 48 h after soccer match compared to baseline and in PBMT with CWI comparison. CWI decreased LDH activity immediately after soccer match compared to baseline (p> 0.05) and was also better than PBMT (p> 0.05), in the 48h comparison both PBMT and CWI were able to reduce LDH activity compared to baseline. For the levels of oxidative damage to lipids and proteins both PBMT and CWI were not able to reduce TBARS near baseline (p < 0.01 and p < 0.001), respectively for analysis immediately after. For 48 hours PBMT reduced TBARS levels to values similar to basal values (p> 0.05). For SOD and CAT, there was an increase in PBMT antioxidant activity compared to baseline levels as well as CWI (p <0.05). We conclude that PBMT used the recovery of soccer players is the best modality for enhancement of prevention of muscle damage and oxidative stress when compared to CWI, leading to complete recovery baseline levels mainly 48h after soccer match.

Key-words: muscular oxidative stress ; photobiomodulation therapy ; muscle fatigue ; muscular recover soccer players.



### EFFECTS OF PHOTOBIOMODULATION ON NEURONAL CELL DEATH IN THE PREFRONTAL CORTEX OF RATS SUBMITTED TO NEONATAL ANOXIA

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Introduction: Neonatal anoxia is one of the major causes of neonatal deaths. The oxygen deprivation caused by this disease can lead to important sequelae to surviving newborns, including impairment of motor and cognitive functions. On the other hand, studies have demonstrated the positive effect of photobiomodulation on neuronal systems. The objective of this study was to evaluate the effects of photobiomodulation in the prevention of cell death in the prefrontal cortex of neonatal rats submitted to neonatal anoxia. Material and Methods: One to twoday old Wistar rats were divided in 4 groups: Control Sham (CS) and Anoxia Sham (AS), Control Laser (CL) and Anoxia Laser (AL). Anoxia groups were exposed to 100% nitrogen gas for 25 min, at 37 oC; control groups were exposed to ambient air. Five minutes later, the animals were irradiated with a diode laser ( $\lambda$ =808 nm) with 100 mW for 60 seconds. For analysis of cell death, the brain were collected 24 hours after treatment and prepared for immunohistochemical analysis using Fluoro-Jade C stain. The materials were analyzed under a fluorescence microscope and the marked cells were counted. Results: AS group presented the highest number of dead cells (p<0.05), showing that the anoxia induced cellular damage in the prefrontal cortex. Laser irradiation did not have effect on control group (CL compared to CS, p>0.05), whereas it reduced cell death after anoxia insult. AL group showed less dead cells compared to AS (p<0.05), but no difference was observed between AL and CS groups (p>0.05). Discussion: The results revealed that photobiomodulation inhibited cell death induced by anoxia. Although the mechanism of cell death in the present model is not totally elucidated, there is evidence that it occurs due to excitotoxicity and neuroinflammation. Furthermore, mitochondrial dysfunction is commonly presented after oxygen deprivation models. Thus we suggest that photobiomodultion could prevent cell death improving mitochondrial function and modulation neuroinflammation. Conclusion: Photobiomodulation decreased neuronal cells death throughout the prefrontal cortex of neonatal rats submitted to neonatal anoxia. Further studies investigating the effects of photobiomodulation on the central nervous system of rats submitted to neonatal anoxia is necessary to strengthen its evidence as a possible therapeutic method.

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### SYSTEMIC PHOTOBIOMODULATION BLUNTED THE INFLAMMATORY PARAMETERS IN ACUTE LUNG INJURY INDUCED BY SEPSIS

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Introduction: Sepsis is a severe disease with high mortality index. The acute lung injury (ALI), characterized by alveolar damage, lung inflammation, and impaired gas exchange, is a serious manifestation of sepsis, which is not efficient treated by current drugs. Therefore, novelty on ALI treatments is substantial. Considering that the photobiomodulation (PBM) treatment displays anti-inflammatory effects, here we studied the effects of systemic irradiation on the ALI.

Methods: Male rats were injected with lipopolysaccharide (LPS) or saline (i.p.) and, irradiated or not with light emitting diode in the caudal vein, for 150 s, 2 and 6 h after the LPS injections. After 24 h the LPS or saline injections, the inflammatory parameters were investigated. Device specifications: Bio Lambda LEDstar, Black Box Mini Model, São Paulo, Brazil; Probe Design, Single Probe; Wavelength: 660 nm; Radiant Power: 160 mW; Power Density: 38,5 mW/cm2; spot area: 4,15 cm2; Density of energy: 5,8 J/cm2; Issuance: Continuous (cw); Total Radiant Emission: 24 J.

Results: We showed that the systemic PBM treatment reduced LPS-induced cell influx into bronchoalveolar lavage and bone marrow, without alter the number of blood cells. We also observed decreased level of IL-1 $\beta$  and IL-6, and enhanced level of IL-10 in the bronchoalveolar fluid (BALF).

Conclusions: Our data show beneficial effects of systemic PBM treatment on ALI caused by sepsis, and suggest that PBM application as an inexpensive and non-invasive additional treatment to sepsis.

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### PHOTODYNAMIC THERAPY DECREASES CELLULAR VIABILITY AND SPHERE FORMATION IN ORAL SQUAMOUS CELL CARCINOMA CELL LINES

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Introduction: Squamous cell carcinoma is the most prevalent malignant neoplasm in the oral cavity. The main therapeutic modalities of OSCC are surgery and radiotherapy alone or in combination with cisplatin. Photodynamic Therapy (PDT) is based on the use of photosensitizers, whose cytotoxicity is activated by light, leading to the death of target cells. Some studies have demonstrated that PDT can be an alternative treatment to OSCC in initial stages. Cancer stem cells (CSC) are a rare subpopulation of cells capable to form colonies, growth in suspension as tumor spheres and resist to conventional treatment. Objective: The aim of this study was to evaluate the effects of PDT in the cellular viability of OSCC cell lines as well as to evaluate the sphere formation capability. Material and Methods: CA1 and Luc4 cell lines were cultivated in DMEMF12+ 10%FBS and supplements. 5x104 cells were plated in 96-well plate and divided in the following groups: control, LED, 5aminolevulinic acid (5-ALA, 0.25-2 mM) and 5-ALA-PDT (5-ALA+LED, 0.25-2 mM). Cells in the 5-ALA and PDT groups were incubated with different concentrations of 5-ALA at 37°C for 4h. Next, cells were irradiated with diode emission light (LED) using the BioLambda LedBOX, 660nm, 6J or 8J/cm<sup>2</sup> and 49.5 mW/cm<sup>2</sup>. Cell viability was evaluated after 24 and 48h of PDT using the MTT assay and the sphere formation was performed after 48h of PDT. Results: No difference in cellular viability was observed between the groups after 24h. CA1 and Luc4 cell lines showed a significant decrease in cellular viability after 5-ALA-PDT with 6J/cm2 and 8J/cm2, respectively, with both 1.5mM and 2.0 mM of 5-ALA. In addition, CA1 cell line showed lower number of spheres after 5-ALA-PDT with 1.5mM and 2.0 mM of 5-ALA than control group. Conclusion: PDT was able to decrease the cellular viability and the number of spheres in OSCC. Additional studies are necessary to understand the effects of PDT in the CSC derived from OSCC.

Keywords: Oral squamous cell carcinoma, photodynamic therapy, 5-ALA, cancer stem cell.



# PHOTOBIOMODULATION REDUCES SCORES IN PUSH AND BWAT SCALES FOR PRESSURE ULCERS

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Introduction: The high incidence of pressure ulcers is considered a serious public health problem and a negative indication of the quality of nursing care Objective: This study aimed to evaluate ulcer healing after photobiomodulation with red LED on the stage 3 and stage 4 pressure ulcers of patients admitted to a public hospital in the state of São Paulo, Brazil. Material and methods: The study was clinical, randomized and controlled, and divided into two groups, GC (conventional group, n = 8) and GLED (LED group, n = 7). The GC were treated according to standard hospital procedures, 1x per day. In the GLED the dressings were applied 2x per day with only a saline solution and red LED ( $\lambda$  = 664 ± 20 [nm], P = 15.0mW, I = 4.8mW / cm2, H = 2.9J / cm2, E = 9.0 J, t = 10 min). Healing was evaluated using the PUSH and BWAT scales as well as measurement of the area. OriginPro 2017 software was used to perform the statistical analysis, with  $\alpha$  = 0.05 in all tests. Results: PUSH scores decreased significantly after LED treatment, from 14.00 ± 2.50 down to 13.00  $\pm$  2.75 (p = 0.03125). There was also a decrease in BWAT scores after photobiomodulation, from 48.00 ± 12.00 down to 44.00 ± 15.25 (p = 0.02344), 6 sessions was determined to be the minimum number of sessions necessary to observe a significant reduction in PUSH scores, which was the primary variable. Conclusion; Photobiomodulation was as effective as the proposed conventional treatment in treating pressure ulcers, with the advantage of practicality and quality ofthe healing process, although there was no significant reduction in area measurement. The statistical power of the significant differences observed was 81%.

Keywords: ulcer; LED, phototherapy, wounds; dressing



## OCT EVALUATION OF DIFFERENT PROTOCOLS IN THE PARALYSIS OF RADIATION CARIES PROGRESSION ON ROOT DENTIN

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Introduction:Radiation-related caries is a late side-effect of radiotherapy of the neck and head. It is painless, affects teeth surfaces that are normally resistant to caries and it may lead to complete loss of dentition within several months of the onset of treatment. Due to the increased risk of opportunistic infections and osteoradionecrosis, oncologic patients should not be treated with conventional dentistry procedures. Thus, the development of minimally invasive protocols which may avoid the progression of carious lesions at early stages is essential. In this context, optical coherent tomography (OCT) may be a useful tool to monitor the development of radiation-related caries without exposing the oncologic patients to additional doses of ionizing radiation.Objective:To evaluate the effects of five different in vitro protocols on the progression of root dentin radiation-related caries by means of OCT. Material and methods:90 bovine root slabs, kept in artificial saliva (1.5 mM Ca; 0.9 P; 0.64 g/L thymol and 0.1 TRIS buffer; pH = 7.0), underwent radiotherapy (2 Gy/day, 5 days/week, 6 weeks). Afterwards, they were artificially demineralized for 32h (1.4 mM Ca, 0.91 mM P, 0.06 ppm F-, 0.64 g/L thymol and 0.05 acetate buffer; pH = 5.0) and then were randomly distributed into 6 groups (n = 15) for treatments: G1 - no treatment; G2 - acidulated phosphate fluoride gel (APG-gel); G3 – Nd: YAG irradiation (wavelength 1064 nm; pulse width 5 ns, total energy 280 mJ and laser beam diameter 5 mm); G4 – APF-gel + Nd:YAG; G5 -Biosilicate® (BS) and G6 – BS + Nd:YAG. After the treatments, all slabs were immersed in artificial saliva for 24 h. Then, all the slabs underwent a cariogenic challenge with S. Mutans(ATCC 25175 and 106 CFU/mL)during 48 hours. OCT was performed at sound dentin slabs and after radiotherapy, artificial demineralization, treatments and cariogenic challenge. Data distribution was verified by D'Agostino& Pearson and Shapiro-Wilk tests and significance difference among groups was calculated using one-way ANOVA ( $\Box$  = 5%). Results: The monitoring of total attenuation coefficient ( $\mu \overline{t}$ ) demonstrated that all experimental groupsexhibited equal or lower values of ut after cariogenic challenge in comparison to the post-treatments values. Only exception was G6 which presented an increase in this parameter. Conclusion: All the treatments, at some level, were capable of reducing the progression of early radiation-related caries lesions. Nonetheless, longitudinal microhardness and transversal microradiography should be performed in order to validate OCT results and understand G6 behavior.

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# EFFECT OF PHOTODYNAMIC THERAPY ON PERIODONTOPATHOGEN - IN VITRO STUDY

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Introduction: Periodontal disease consists of an inflammatory response of the tissues surrounding the tooth, caused by the accumulation of biofilm derived from deficiency in the oral hygiene and virulence of microorganisms. Conventional treatment consists of scraping and root planning and in some cases association with antimicrobials is required. Although working effectively, conventional treatment may not yield satisfactory results. An effective treatment that has been studied is antimicrobial photodynamic therapy (aPDT), which may act as an adjunct to conventional treatment. Such therapy consists of the use of a photosensitive dye (PS) associated with a resonant wavelength light source with the photosensitizer absorption spectrum, which in the presence of oxygen generate reactive oxygen species capable of causing damage. Objective: The aim of the present study was to evaluate the use of aPDT in Aggregatibacter actinomicentecomitans, as well as parameter optimization. Material and methods: For this, standard ATCC 29523 strain of A. actinomicentecomitans cultured in microaerophilia for 48h was used. Then experiments were performed in triplicate with 6 groups: 1) Control, no intervention 2) Light, which was only irradiated 3) 0, where only FS was used 4) 1, which was performed aPDT with 1min irradiation 5) 3, PDT with 3min irradiation and 6) 5, PDT with 5min irradiation. Methylene blue dye at 100 µM concentration was used as PS and Laser light (Photon Lase III, DMC, São Carlos, Brazil) of  $\lambda$  = 660nm with 100mW power and radiant exposure of 214.18 J/cm2. After microbial growth the cfu/mL count was performed and the mean and standard deviation were made. Results: Fortyeight hours after treatment, the control, light and only dye groups showed high microbial growth. In contrast, the groups in which the aPDT was performed presented logarithmic decay proportional to the irradiation time, and in 5min there was a growth of less than 1x103. Conclusion: Therefore, we conclude that aPDT was effective in microbial inactivation of A. actinomicentecomitans in vitro.

Keywords: antimicrobials, photosensitizer, photodynamic therapy

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### TRANSCUTANEOUS PHTOBIOMODULATION AS A CO-ASSISTANCE IN THE CONTROL OF BLOOD PRESSURE – CASE REPORT

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INTRODUCTION: This is a case report whose objective was to verify the effects of transcutaneous photobiomodulation in a patient with systemic arterial hypertension.

MATERIAL AND METHODS: The methodology used is a case report of a female pacient who is a university ambulatory patient who is part of a study that evaluates the effects of photobiomodulation on hemodynamic variables of hypertensive patients. The ethical precepts were respected.

RESULTS: Case report - Participant BMI, female, 60 years, divorced, hypertensive for 4 years, using beta-blocker, adherent to treatment. Ethical precepts were respected and the patient underwent photobiomodulation sessions at home for 3 months where she underwent 15 sessions, performed in 3 cycles of 5 consecutive days and pause of 20 days between each cycle. A red laser with 360 J per session with transcutaneous application over the radial artery was used. Systolic and diastolic blood pressures, SBP and DBP, respectively, as well as heart rate (HR) were monitored during the sessions. At the end of the cycles, SBP decreased from 129 mmHg to 96 mmHg, DBP decreased from 77 mmHg to 62 mmHg, and HR decreased from 102 to 65 beats per minute, in addition to beta-blocker suspension (under prescription doctor).

DISCUSSION: Photobiomodulation is a technique increasingly used by various areas of health. Its use is generally employed for tissue repair, being beneficial in accelerating scar, inflammation and pain processes. Some studies reveal that photobiomodulation also promotes hypotensive effects on blood pressure, but gaps in the literature still exist. Considered as one of the most important non-pharmacological therapeutic advances today, the technique can enable efficient antihypertensive treatment.

CONCLUSION: The use of low intensity laser applied transcutaneously promoted modulating therapeutic effects, reducing blood pressure and heart rate in this patient.

DESCRIPTORS: Lasertherapy; Hypertension; Photobiomodulation.



### PHOTODYNAMIC THERAPY REVERSES THE NEGATIVE EFFECTS OF PERIODONTITIS ON THE UTERINE MICROENVIRONMENT DURING THE GESTATION

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Introduction: Periodontitis (P) is one of the most common infectious diseases that affects the periodontium and gradually destroys periodontal tissues. Among several systemic effects occasioned by P, alterations during the gestation have been studied. PDT is characterized by the association of a light source with a photosensitizing agent in order to cause cell necrosis and microbial death. Thus, our objective was to evaluate the effect of PDT on the negative repercussions of periodontitis during pregnancy, mainly in the uterine microenvironment.

Methods: Ten days before pregnancy, periodontitis was induced by ligature technique, and subsequently the rats were caged overnight with a male. Pregnancy was confirmed by vaginal smear. The treatment with PDT was performed 15 days after the induction the ligatures. Pregnant rats non-manipulated were used as control. Pregnant rats were euthanized at day 18 of gestation and the uterus was removed in order to investigate the parameters. The photosensitizer methylene blue (0.005%, CHIMIOLUX, DMC, São Paulo, Brazil) was administered at the two sites (vestibular and lingual). After three minutes, the periodontal pockets were irradiated with a red laser (MM OPTICS; Wavelength 660 ± 10 nm; Radiant power100 mW; Exposure duration 90s; spot size 0.02827 cm2; Radiant energy 9 J; Irradiance 3.5 W/cm2; Radiant exposure 318 J/cm2; Total radiant energy18 J).

Results: We showed that PDT had an important impact on the uterine microenvironment reducing the gene expression of IL-6, COX-1, COX-2 and NOS in the uterine tissue of pregnant rats with periodontitis. We also observed that PDT reversed the decreased level of IL-10 in the placenta.

Conclusions: Thus, our data showed the important role of oral health during gestation as well as PDT is an effective therapy. These studies might be useful in providing an important indicator of risk for future obstetric complications considering the impact in the offspring.

Ethical Approval and accordance: This study were performed according to the Brazilian Society of Laboratory Animals Science (Sociedade Brasileira de Ciência em Animais de Laboratório; SBCAL) and approved by the Animal Care Committee University Nove de Julho (CoEP-UNINOVE, AN005/2017).

Conflict of interest: The authors declare that there are no conflicts of interest regarding the publication of this paper.

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### TREATMENT OF HERPES LABIALIS BY PHOTODYNAMIC THERAPY: CONTROLLED, PROSPECTIVE, RANDOMIZED, DOUBLE-BLIND STUDY

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Background: Lesions of herpes labialis are caused by the herpes simplex virus type 1 (HSV-1) and cause pain and aesthetic compromise. It is characterized by the formation of small vesicles that coalesce and rupture forming extremely painful ulcers, that evolve to crusts, dry desquamations until their complete remission. Currently the treatment of these lesions is done with acyclovir. Although it diminishes the symptomatology, it causes viral resistance and does not prevent the recurrence of the lesions. It is known that photodynamic therapy (PDT) has numerous advantages, among them :the reduction of the time of remission, and does not cause resistance.

Materials and methods: A total of 30 patients with herpes labialis in the prodromal stage of vesicles, ulcers and crusts will be selected to participate in the study and randomized into two groups: G1 control and G2 experimental. After signing TCLE and TA, patients in group G1 will undergo the standard gold treatment for cold sores with acyclovir and simulated PDT treatment. Patients in the experimental G2 group will be treated simulating the gold standard treatment of herpes labialis (placebo) and PDT. In all patients will be collected saliva samples for analysis of cytokines, and will be performed exfoliative cytology in the lesions. The pain will be assessed through a pain scale and a questionnaire of quality of life related to oral health (Ohip- 14) will be given to them. Patients will continue to be followed up after 7 days, 1 month, 3 months, 6 months and 1 year and, if there is a recurrence of the lesion, they will contact the researchers.



### EFFECTIVENESS OF THE MODIFIED INTRAVASCULAR LASER IRRADIATION OF THE BLOOD (MODIFIED ILIB) METHOD AS AN ADJUNCT IN THE TREATMENT OF DYSMENORRHEA.

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Introduction: Menstruation is a periodic change that has important significance in the physical, emotional and reproductive aspects of women. Several women undergo painful discomfort during this period called dysmenorrhea, defined as pelvic pain associated with menstruation, constituting one of the most frequent gynecological complaints. Objective: The aim of this study was to evaluate the effectiveness of modified ILIB with 660 nm InGaAIP laser in the treatment of dysmenorrhea through pain scales. Material and methods: We selected twenty female subjects to apply modified ILIB for 30 minutes. The laser parameters were: 660 nm wavelength, 0.028 cm2 laser beam diameter, 100 mW power, 180 J energy dose, fluence of 918 J/cm2 and irradiance of 3,571.4 mW/cm2. For the application of the technique, the radial artery was located on the left wrist of the participants through digital pressure and a flexible and non-toxic carbon fiber bracelet was placed positioned with the perforation of the socket over the radial artery where the tip of the equipment was inserted, thus avoiding the beam displacement. The sessions began at the beginning of the menstrual period, daily, throughout the cycle. Painful symptoms were followed and evaluated using the Visual Analog Scale (VAS) and the McGill Pain Questionnaire (MPQ) immediately before patients started the proposed treatments and after 30 minutes of each session. Results: Measurement of pain using the VAS scale led to decreased pain, especially on the first day of treatment of each cycle, presenting a statistically significant difference when comparing initial pain with pain immediately after treatment at first and second cycle (p<0.05). Regarding the MPQ, pain sensation can be observed over the three days of treatment in each cycle, with a statistically significant difference between pain assessment before treatment and after treatment on the first day of the first menstrual cycle evaluated (p < 0.05). Conclusion: The modified ILIB with 660 nm laser was effective in reducing dysmenorrhea, especially on the first day of each menstrual cycle evaluated, proving to be a clinically useful tool to provide more comfort to patients suffering from dysmenorrhea.

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# 780 NM LASER THERAPY EFFECTIVENESS FOR BONE REGENERATION IN OPEN FRACTURES

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Introduction: Trauma is one of the most relevant public health problems as it is a major cause of death among young people, and causes major social and economic complications not only for the person who suffers it but also for the health system. In this context, new treatments with the ability to accelerate the process of bone regeneration are desirable. Objective: The aim of this study was to evaluate the effectiveness of 780nm LLLT as an adjunct to surgical treatment in bone fracture regeneration, to evaluate the analgesic effect, to analyze the influence on the use of prescribed analgesic and anti-inflammatory drugs and to verify changes in levels of inflammatory cytokines (TNF- $\alpha$ , interferon gamma, IL-1- $\beta$ , IL-10 and IL-17) before and after LLLT compared to the control group. Material and methods: Twenty patients with a clinical diagnosis of open limb fracture assisted by a public hospital in the interior of Bahia were selected and randomly divided into two groups: control (surgical treatment) and LTBI (surgical treatment and LLLT). LLLT parameters were: 780 nm, 0.04 cm2 beam diameter, power of 40 mW, 10 seconds per point, energy of 0.4 J, fluence of 10 J/cm2 and irradiance of 1 W/cm2. McGill pain assessment, analgesic and anti-inflammatory drug use, serum cytokine levels TNF-a, IFN-y, IL-1β, IL-10 and IL-17 were evaluated for all patients in the sample, and bone regeneration level was evaluated by digital standardized radiographs. Data were analyzed by Wilcoxon and Mann-Whitney tests (5%). Results: The LLLT group showed improvement in bone regeneration progress by comparing the images of days 30 and 60 by increasing bone density for this group (p = 0.005 and p = 0.002, respectively), as well as reducing pain after the 5th session compared to the first session (p = 0.019), reduction of medication use and increase in IL-1 $\beta$  dosage after 60 days of treatment. Conclusion: LLLT was effective as an adjuvant in the process of bone fracture regeneration, altered IL-1ß levels, reduced the use of analgesics and anti-inflammatories and decreased pain pattern throughout the sessions.

Funding sources: This study was financed in part by the Coordenação de Aperfeiçoamento de Pessoal de Nível Superior – Brasil (CAPES) – Finance Code 001.



### LASERTHERAPY EQUIPMENT FOR APPLICABILITY IN ORTHODONTICS "AND OTHER AREAS, FOR ORAL USE WITH PROTOCOLS AND PATENT"

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Introduction: Photobiomodulation using low-power lasers have been used for accelerating orthodontic treatment with success, the literature shows a speed of induced tooth movement around 40%. Many equipments and parameters have been used for this treatment, however most of them use lasers as a light source, are not affordable to the patients, require previous training to use the equipment and is not adapted for use in orthodontics. Objective: The aim of this study is to develop an intra-oral equipment, easy to use and affordable, to be used for the patients in home, during orthodontic treatment. Materials and Methhods: The photobiomodulation appliance is protected under the patent deposit BR 10 2019 009966 6. The equipment was developed using as a model a flexible sport mouth-guard. Nineteen LEDs emitting at 660nm and 850nm, with individual power output of 63 mW/per IR LED and 85 mW/per Red LED, resulting in a total of 1,4W per equipment, were adapted and installed covering all the buccal surface of the mouth-guard. The LEDs spotlight were directed to the tooth crown and gingiva over the buccal root surface. An electronical controller is used to allow an irradiation of 60 seconds, resulting in a total energy of 84Joules and an Energy density of 4,2J/cm2. The protocol for tooth accelerating was based in Suzuki et al., Jettar et al. and Fernandes et al., but a future protocol for analgesia after orthodontic activation is under development. Results: Two prototypes of the photobiomodulation appliance were developed and the parameters of irradiation and adaptation for intra-oral use was performed. The test for light delivery and LED distribution over the gingiva and tooth surface was calculated. Discussion: The use of photobiomodulation during orthodontic treatment is an effective treatment for accelerating tooth movement, reducing the total treatment time and consequently risk of caries and gum inflammation. However, the use of low-power lasers usually is not adapted for clinical use, since the small size of laser spot is time-consuming and uncomfortable for the patients and for clinicians. An intraoral appliance, that could irradiate the crown and gingiva of teeth involved in the orthodontic treatment is highly desirable. Conclusion: The photobiomodulation appliance presented and described at patent BR 10 2019 009966 6, is a promising approach for use in orthodontic treatment.



# SUGARS AND THEIR EFFECTS ON ANTIMICROBIAN BEHAVIOR OF PHOTODYNAMIC THERAPY IN STREPTOCOCCUS MUTANS

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Introduction: Streptococcus mutans are Gram-positive bacteria that can be found in the oral cavity, larynx and intestine. It has the ability to rapidly transport and metabolize a wide range of sugars, which may be directly related to its survival in oral biofilm and it has cariogenic potential in humans. In the presence of sugars, S. mutans activates membrane receptors by stimulating transport pumps, thereby increasing glucose uptake. Photodynamic therapy consists of the incorporation of a photosensitizer into the cell cytoplasm, followed by irradiation with visible light, which results in cell death. Objective: The objective of this work was to investigate the effect that sugars have on the antimicrobial behavior of photodynamic therapy in Streptococcus mutans (S. mutans). Material and methods: We used S. mutans (ATCC 25175) that it was cultured in onto agar brain and heart infusion (BHI) incubated at 37°C for 48 h under 5-10% CO2 atmosphere. Bacterial inocula were prepared by collecting pure colonies and suspend it in phosphate buffered saline (PBS) with and without 50 mM glucose and/or mannitol. Methylene blue (MB) photosensitizer (FS) was added to bacterial suspension to final concentration of 100 µM. Verapamil was used as a blocker for efflux pumps. Six groups were used: (control, irradiated with LED without FS, FS without irradiation, and PDT with time of 0, 60 and 120s). Results: Control, LED and FS groups did not present significant differences (P>0.5) with and without glucose or mannitol. After 2 min irradiation without glucose, we observed a reduction of 100% of the bacteria, while there was no significant microbial reduction in the presence of glucose or mannitol. In experiments with verapamil we observed a microbial reduction of approximately 2 logs in the presence of mannitol. We can conclude that PDT is a viable solution for inactivation S. mutans, and in the presence of sugars delayed microbial inactivation.

Funding sources: CAPES



### EVALUATION OF THE EFFECT OF PHOTOBIOMODULATION ON GRAFT SKIN DONOR AREA IN BURN PATIENTS IN RELATION TO PAIN: CASE STUDY

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Introduction: Introduction: The burns are skin wounds secondary to the direct or indirect action of chemical, thermal, electrical or radioactive agents that can cause partial or total destruction of the skin and reach even the deepest lavers, such as subcutaneous tissue, muscles, tendons and even even the bones. Depending on the assessment of the patient who has suffered burns, he may be hospitalized and subjected to systemic and topical therapy, including surgery for debridement and graft. The skin graft is obtained from a healthy donor area and this creates another wound. This wound is treated with coverage and the patient, in the first postoperative, reports severe pain and is treated with analgesics. As photobiomodulation (FBM) using low intensity laser and light-emitting diode (LED), has shown good results regarding anti-inflammatory action, pain, better tissue organization, acceleration of angiogenesis and stimulation of leukocyte chemotaxis, this study aimed to evaluate pain in patients with skin graft donor area submitted to PBM using LED. Material and Methods: Case study with methodology approved by CoEP of Nove de Julho University, including 2 patients with second degree burns and 1 patient with mixed second degree and 4 donor areas (one patient with two donor areas), divided into two groups: 2 donor areas in the control group (GC) treated with Membracel® (cellulose porous regenerating membrane) and two donor areas in the LED group, treated with Membracel® and LED, all without secondary coverage. The donor areas of the LED group were submitted to five PBM sessions (660 nm, 2.55 mW / cm2, 1.53J / cm2), lasting 10 minutes each, in the immediate postoperative, 1st postoperative period (PO), 3rd PO, 5th PO and 7th PO. To assess pain evolution, the analogic visual scale (AVS) was applied, with values from 0 to 10, in the postoperative period in which the LED sessions were performed. Results and discussion: In the patient with two donor areas, pain in the CG area remained high until the 7th PO and in the LED group, pain decreased in the 3rd PO. Conclusion: The results suggest an important action of PBM on pain reduction, mainly evidenced by the comparison between the two donor areas of the same patient.

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# IS IT POSSIBLE TO IMPROVE MESENCHYMAL STEM CELLS PARACRINE EFFECTS WITH LEDS?

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Introduction: Mesenchymal stem cells (MSCs) are being used in many clinical studies because of their paracrine action that can treat various diseases but the loss of MSCs at the transplanted site lowers the efficiency of cellular therapy. To address this problem, the use of light sources is a promising tool. Besides the beneficial roles that light sources may play in secreted cytokines and growth factors related to the modulation of inflammation, it is necessary to highlight the importance to seek the capability of the cells to remain stemless, viable, proliferating and self-renewing. Therefore, the present study aimed to irradiate MSCs using LEDs in order to improve cell secretome seeking a more efficient cellular therapy. In this milieu, we evaluated the secretion of key factors: proinflammatory (Tumor Necrosis Factor Alpha (TNFalpha)) and anti-inflammatory (Interleukin 10 (IL-10)) cytokines as well as Insulin Growth Factor 1 (IGF-1) that is responsible for promoting proliferation and selfrenewal; and Interleukin 6 (IL-6) that contributes to stemless characteristics, proliferation and protection of cellular apoptosis. Material and methods: Adipocyte derived MSCs were obtained from Fischer-344 male rats and irradiated with different radiant exposures (0.5, 2 and 4 J/cm<sup>2</sup>); the influence of the number of irradiations (single dose or once every other day in a total of three doses) was also assessed. Irradiations were performed using a 630 nm LEDbox (Biolambda, Brazil) at a constant irradiance of 15 mW/cm<sup>2</sup>. Control group was kept in the dark for the same time as 4 J/cm<sup>2</sup>-placebo. TNF-alpha, IL-10, IGF-1 and IL-6 analyses were performed 24 hours after the last irradiation. Results and Discussion: 0.5 J/cm<sup>2</sup> group showed a decrease in IL-10 secretion in the conditioning treatment compared to the control group, where, on the other hand, 2 J/cm<sup>2</sup> group showed an increase in TNF-alpha levels in a single dose irradiation compared to the control group. The 4 J/cm<sup>2</sup> group that received the conditioning treatment increased secretion of IGF-1 and IL-6 compared to the control group. Conclusion: The conditioning treatment of the group irradiated with 4 J/cm<sup>2</sup> improved cytokines responsible for stemless characteristics, viability, proliferation and self-renewal, unaltering the expression of pro and antiinflammatory factors.

Funding sources: São Paulo Research Foundation (FAPESP #2018/06865-7) and Higher Education Personnel Improvement Coordination (CAPES #1794278).

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## EVALUATION OF MESENCHYMAL STEM CELLS METABOLISM USING CONDITIONING TREATMENT OR SINGLE DOSE IRRADIATION

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Introduction: Mesenchymal stem cells (MSCs) are being studied due to the paracrine effects that help repairing various diseases, as an example, decreasing inflammation and scar formation and increasing local capillarity in infarcted myocardium. However, the loss of MSCs at the transplanted site is large. To address this problem, the use of light sources is a promising tool. It is well known that low level light sources play beneficial roles in mitochondria, an important organelle whose main function is essential for cell homeostasis through the generation of energy by the synthesis of adenosine triphosphate (ATP). In addition, the organelle also participates in NO metabolism and regulation of cell viability. Therefore, the aim of this study was to irradiate MSCs using LEDs in order to improve cell metabolism seeking a more efficient cellular therapy. Material and methods: Adipocyte derived MSCs were obtained from Fischer-344 male rats and irradiated with different radiant exposures (0.5, 2 and 4 J/cm<sup>2</sup>); the influence of the number of irradiations (single dose or once every other day in a total of three doses) was also evaluated. Irradiations were performed using a 630 nm LEDbox (Biolambda, Brazil) at a constant irradiance of 15 mW/cm<sup>2</sup>. Control group was kept in the dark for the same time as 4 J/cm<sup>2</sup>-placebo. MTT, AlamarBlue, NOx index (nitrate and nitrite ratio) and ATP analyses were performed 24 hours after the last irradiation. Results and Discussion: MTT increased in all single dose groups compared to the control group, however, this improvement in mitochondrial function was only confirmed in the group 4 J/cm<sup>2</sup> compared to the control group by AlamarBlue, NOx index and ATP quantification. Furthermore, when performed the conditioning treatment once every other day, only the group 4 J/cm<sup>2</sup> improved cell metabolism in all analyses compared to the control group. Conclusion: The use of red LEDs with 4 J/cm<sup>2</sup> improved all cell metabolism makers using single dose or conditioning treatment irradiation.

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Acknowledgment: We thank the Gynecology Department for the use of their Cell Culture Laboratory.



### PHOTOBIOMODULATION IN ORAL MUCOSITIS IN PATIENTS WITH HEAD AND NECK CANCER: A COST-EFFECTIVENESS STUDY

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Introduction: Oral Mucositis (OM) is an alteration due to toxicity caused by cancer treatment. Low intensity laser therapy has anti-inflammatory, analgesic and tissue repair properties. Objective: The objective of this work was to perform a systematic review and meta-analysis of the randomized clinical trials of the treatment of OM with laser in patients in treatment of cancer of the head and neck, followed by costeffectiveness analysis of the therapy. Material and Methods: The search terms "mucositis" AND "phototherapy", "laser therapy" AND "mucositis". "photobiomodulation" and "mucositis" were used to search the databases PubMed, Web of Science and MEDLINE. ", And" low level laser therapy "and" mucositis "in the last ten years. After the initial search we included only the randomized clinical trials and with two groups: one treated with laser and one placebo group. The studies were analyzed according to the criteria PRISMA and GRADE to evaluate the quality and degree of evidence. Results: After the first selection, 13 studies in the bibliometric analysis and the same were included in the systematic review, after the systematic review, only 6 studies were included in the meta-analysis. Regarding scientific production, we observed the research groups that published clinical trials on the subject are predominantly from India and Brazil. The results of the systematic review and meta-analysis showed that the laser presented good results in clinical improvement and pain reduction, decreasing the chance of the patients to be 64% (RR = 0.36 [95% CI = 0.29-0.44]). patients develop OM with degrees of debilitating lesions. The cost-effectiveness analysis resulted in an incremental cost of R \$ 3,687.53 for the laser group, with an incremental effectiveness of 132, 2 and the incremental cost-effectiveness ratio (ICER) was 27.89 for high OM cases avoided. Conclusion: It was concluded, therefore, that the photobiomodulation for OM in patients under treatment of head and neck cancer was clinically effective and costeffective.

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### ACTION OF ANTIMICROBIAN PHOTODYNAMIC THERAPY WITH RED LED IN REDUCING HALITOSIS: CONTROLLED CLINICAL TEST

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Introduction: Halitosis is the term used to describe any unpleasant odor relative to expired air. The prevalence of halitosis in the population is approximately 30%, of which 80-90% of the cases originate in the oral cavity resulting from proteolytic degradation by gram negative anaerobic bacteria. Antimicrobial photodynamic therapy (aPDT) has been widely used and with quite satisfactory results in the health sciences, it involves the use of a non-toxic dye, called photosensitizer (PS), and a light source of a specific wavelength in the presence of the oxygen in the medium. This interaction can create toxic species that generate cell death. Objective The objective of this controlled clinical study was to verify the effect of aPDT in the treatment of halitosis by the evaluation of the formation of volatile sulfur compounds with gas chromatography. Materials and Methods: Thirty-five young adults aged 18 to 25 years, with gas chromatographic halitosis were included in the study. The selected subjects were divided into 3 groups, G1 aPDT with 0.005% methylene blue with red LED 660nm, 4 irradiation points, 90s per point, 400mW of power, 36J per point, G2 Scraper and G3 Scraper and aPDT. All subjects underwent Oral ChromaTM evaluation before, after treatment and controls of 7, 14, 30. Wilcoxon test was used for the analysis of data from Oral ChromaTM intragroup and for the intergroup analysis the Kruskal Wallis test. A significance level of 95% (p < 0.05) was considered for all analyzes. Results: It was possible to observe statistically significant difference immediately after treatment for all groups, but the result was not maintained after 7 days. Conclusion: With the data obtained in this study it was possible to observe an immediate result in the reduction of halitosis after treatment with aPDT using red LED, but it was not maintained, requiring further studies addressing education and motivation with controls to define exactly the patient's return time in order to maintain treatment efficiency.

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### CLINICAL AND HISTOLOGICAL QUALITY OF MAMMOPLASTY SCARS AFTER PHOTOBIOMODULATION: A RANDOMIZED, CONTROLLED, DOUBLE-BLIND CLINICAL TRIAL

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Introduction: The surgical trauma, combined with injuries produced in accidents and secondary to other clinical conditions, represents a substantial expense for society, requiring a comprehensive multifactorial approach to solve problems related to wound healing. There are different options for treating scars and wounds, but no therapy promotes total skin regeneration and is adopted without dispute in the literature. Some procedures even present unsatisfactory or inconsistent healing results. Photobiomodulation (PBM) is the application of light to revive, repair, renew and protect damaged, degenerate or necrosis-threatening tissue. Therefore, PBM is a low-cost noninvasive procedure that will add to the postoperative healing therapeutic arsenal. Objective: To evaluate the clinical and histological quality of the operative scar of mammoplasty after photobiomodulation with the 660nm laser. Material and Methods: A randomized, double-blind, controlled clinical trial will be conducted from November 2019 to February 2021 with volunteer participants who need to perform mammoplasty on both breasts at the Mandagui Hospital Plastic Surgery Service in São Paulo. Participants with Fitzpatrick classification phototypes III, IV and V will be submitted to 660nm laser application during 06 weeks postoperatively in inframammary scar of one breast and will have therapy control (simulated laser) in the inframammary scar of the other breast. Thus, the intrinsic genetic characteristics of each individual will be maintained and the analysis will be reliable to the laser effect. Biomarkers of the local microenvironment and dermal histology will be studied at the beginning of the healing process - 06 weeks - and after 06 months, when the scar can be considered mature in the literature. At 06 months, 02 validated clinical perception questionnaires will also be applied - one for the patient and one for independent expert evaluators. All data will be statistically evaluated according to their distribution. Rationale: The histological analysis and objective evaluation of important biomarkers in the quality of the healing process will be of great value to study and prove the effectiveness of FBM. In addition, questionnaires for patients and independent specialists will add relevant information about satisfaction, symptoms and final aesthetic outcome of the scar following noninvasive treatment with no side effects, minimizing financial and psychological costs for patients, institution and professionals of health.

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### COMPARATIVE STUDY BETWEEN PHOTODYNAMIC THERAPY WITH URUCUM AND LED AND PROBIOTICS IN THE HALITOSIS REDUCTION – PROTOCOL FOR A CONTROLLED RANDOMIZED CLINICAL TRIAL

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Introduction: Halitosis is the technical term for bad breath (foul odor emanating from the oral cavity), the origin of which may be local or systemic. The aim of the proposed project is to determine whether antimicrobial photodynamic therapy (PDT) and the use of probiotics are effective for the treatment of halitosis. Objective: The aim of this study was to evaluate in vitro the effects of daily PBM with dosimetric parameters used to treat OM in stem properties of OSCC cell lines. Material and methods : Fiftytwo students and employees of University Nove de Julho between 18 and 25 years of age with a diagnosis of halitosis (hydrogen sulfide concentration  $\geq$  112 ppb determined by gas chromatography) will be randomly divided into groups of 13 individuals submitted to different treatments: Group 1 -brushing, dental floss, and tongue scraper; Grupo 2 – brushing, dental floss, and antimicrobial PDT applied to the middle third of the dorsum of the tongue; Group 3 - brushing, dental floss, and probiotic; Grupo 4 – brushing and dental floss. Halimetric findings will be compared before, immediately after, seven days after and 30 days after treatment. Coated tongue samples will be submitted to microbiological analysis at these same evaluation times. The quantitative analysis will be performed with real-time PCR. The Shapiro-Wilk test will be used to determine the normality of the data. Parametric data will be compared using analysis of variance (ANOVA) and nonparametric data will be compared using the Kruskal-Wallis test. The results of the treatment in the different evaluation periods will be compared using the Wilcoxon test. Results: Expected results are that aPDT and the use of probiotics are effective in reducing halitosis. Discussion: Halitosis can affect interpersonal relations, exerting a negative impact on social wellbeing. Studies assessing the reduction of halitosis using PDT and probiotics are scarce. Conclusion: It is concluded that this project is important due to the scarcity of studies evaluating halitosis reduction through photodynamic therapy and the use of probiotics, presenting urucum as photosensitizer and LED as a light source more accessible to dentists.



### PHOTOBIOMODULATION USING LASER THERAPY IN THE CONTROL OF THE INFLAMMATORY PROCESS AND PROMOTING REPAIR IN THE TENDON IN THE EXPERIMENTAL TENDINITIS MODEL

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Introduction: Tendinopathy is a common disease that is difficult to treat. Traditional treatment uses anti-inflammatory drugs, with unsatisfactory results due to side effects. Photobiomodulation therapy is a promising auxiliary treatment for reducing the inflammatory process. However, there are few studies demonstrating the participation of lipoxin receptors in the resolution mechanism. Objective: The main of this study was to evaluate the effect of PBMT on inflammatory process resolution in an experimental model of tendinitis, evaluating the expression of COX2, TNFa, TGFb and Lipoxin receptors (LR), as well as myeloperoxidase (MPO) activity. Material and methods: Male Wistar rats were anesthetized and the tendinitis was induced by collagenase injection (100 micrograms / tendon) and distributed into 4 groups: Control (CTL), tendinitis untreated (NT) or treated with Sodium Diclofenac (DIC) or Laser (PBMT). Results: The NT group showed increased of the COX2 and TNFa expression and MPO activity. In contrast, there was a decrease in TGFb and LR expression. The DIC Group showed a decrease in COX2, but the LR remained low. The PBMT group presented reduction of COX2 and TNFa expression and MPO activity, with increase of TGFb and LR expression. Discussion and Conclusions: PBMT (810nm; 3J; 30mW) reduces the inflammatory process and directs the resolution process. The mechanism of action may be related to the increase of lipoxin receptors and resolution of tissue repair.



### PHOTOBIOMODULATION AS FINAL TREATMENT FOR THE CHONICAL PAIN IN THE SUBCUTANEOUS TRAJECT OF THE VENTRÍCULO-PERITHONEAL SHUNT: A CASE REPORT

### ALESSANDRA MUOIO, DANIELA TEIXEIRA, VALÉRIA MUOIO, EDGARD NETO AND FERNANDO PINTO

ABSTRACT: This is a case report in which patient S, EN 38 years old underwent a craniotomy about two years ago due to an arachnoid cyst and placement of a right ventricular-peritoneal valve and presented chronic pain throughout the entire course. subcutaneous valve passage. Initially the pain was treated with common analgesia, evolving to opioids and psychiatric follow-up. There was no pain improvement, on the contrary, it increased in intensity and its frequency became daily. A photobiomodulation protocol was then initiated focusing on analgesia. OBJECTIVE: To demonstrate the efficacy of analgesic treatment in a relevant and disabling clinical complaint despite oral analgesia. Photobiomodulation showed no alteration in shunt functioning based on clinical examination. MATERIAL AND METHOD: A 38-year-old patient underwent 10 photobiomodulation sessions with a 7-day interval using a lowintensity laser device, radiating 20 points along the cervical region to the right abdomen, red light,  $\lambda$  = 660nm, P = 25mW, I = 1W / cm<sup>2</sup>, E = 100mJ, H = 4J / cm<sup>2</sup>, t = 4s per point with 2cm equidistant points totaling 30 irradiated points. It was also used the visual analog scale for pain assessment in each subsequent photobiomodulation session. RESULTS: Immediately after the first session, the patient reported significant improvement, especially of the cervical region. During the treatment with photobiomodulation and, through the analogue visual pain scale analysis and total decrease of opioid use, the patient presented improvement of over 90%. DISCUSSION AND CONCLUSION: Photobiomodulation had a very effective analgesic effect on pathology unpublished in medical literature.



# EVALUATION OF LIGHT ABSORPTION OF CELL LINES CULTIVATED UNDER NUTRICIONAL STRESS

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Introduction: Photobiomodulation (PBM) is based in the use of light to modulate different cellular process, including proliferation, migration and cellular viability. The cellular response to PBM is dependent on the absorption of light photons by chromophores. In vitro studies have demonstrated that PBM can promote or inhibit the malignant behavior of oral cancer cells. In addition, the effects of PBM in a specific cell type is dependent of light absorption and culture conditions. Objective: The aim of this study was to evaluate the effects of nutritional stress in the absorption spectrum of cell lines. Material and methods: The oral squamous cell carcinoma cell lines SCC9, CA1 and LUC4, oral dysplastic cell line DOK, the immortalized skin keratinocyte cell line HaCAT and fibroblasts isolated from dental pulp were cultivated in the appropriate medium and divided in the following groups: 0% FBS, 5% FBS and 10% FBS. After 24h, cells were collected, counted and 1x105 cells were ressuspended in phosphate buffer solution (PBS). The absorbance was measured using the USB-2000 spectrophotometer. Results: It was observed that there was no significant difference in the cellular absorbance between the different FBS concentrations in CA1, LUC4, SCC9 and fibroblasts. There was a significant increase in the absorption of HaCAT cells cultivated with 5% and 10% of FBS when compared to cells cultivated with 0% FBS. DOK cells demonstrated a significant decrease in the absorbance with 10% FBS when compared to both, 0% and 5%. In addition, SCC9 cell line showed decrease in cellular absorbance in the 10% FBS group in relation to 0% and 5% FBS. Conclusion: Malignant and dysplastic cell lines were associated with reduced absorbance after 24h of nutritional stress and thus, the effects of PBM may be influenced by the in vitro culture condition.



# EFFECTS OF LOW-INTENSITY LASER THERAPY ON WOUND HEALING IN AMPUTATION

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Introduction: The World Health Organization points out that in 2019 diabetes is the main cause of non-traumatic amputation in Brazil. The conventional treatment for amputation wound is the use of toppings. Studies point out that low-intensity laser (LBI) assists in the treatment of this lesion by stimulating tissue healing and pain control. Objective: The aim of this study was to evaluate the effect of low-intensity laser in a patient with amputation wound. Material and methods: The patient E.L. C, 84 years old, Caucasian, female. Medical diagnosis: Diabetes mellitus, arterial hypertension and peripheral arterial obstructive disease. He presented as main complaint pain in the area of the lesion. The ulcer was located in amputation in the right lower limb without the presence of infection. The same was irradiated using Laser Therapy XT, wavelength 660 nm and 808 nm, power of 100 mW, power density of 1.02 mW/cm<sup>2</sup>, Creep of 20 J/cm<sup>2</sup>, 20 sec and total energy of 2 J, in direct contact, for once a week. After 09 Irradiations in the wound presented complete healing of the area. Results: The lesion presented complete healing and decreased pain after LBI with 2 J. The patient had no adverse effects after application. It was used in perilesional skin ozonized oil and as cover hydrogel and Alginate. Conclusion: The LBI proved to be effective in healing with 2 J, with analgesic effect and without the presence of adverse reactions, constituting a biostimulative and non-invasive therapy. In addition to the techniques employed in biophotonics are highly efficient and selective, with minimal reports of adverse events, it is necessary to develop research in this area.



# THE OCT EVALUATION OF WHITE SPOT LISIONS AFTER APPLICATION OF DIODE LASER 808NM AND FLUORIDES

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Introduction The association of laser use and fluoride seems to be able to accelerate the therapeutic process of initial caries lesions in dental enamel. The objective of this study was to evaluate the coefficient of retro-scattering in incipient caries lesion in dental enamel after irradiation with laser diode. The examples were forced for the formation of the initial lesion and irradiation with the diode laser and topical fluoride treatment. The C group causes only caries, CL cause of caries plus laser irradiation diode 808 nm and the other groups, CD caries lesion and with 5% fluoride varnish, CDL caries lesion and with diode laser 808nm and with fluoride varnish 5% Gallic acid is one of the main acidulated phosphate agents, the most potent gastric acid is the molecule of Phosphate C, CL, CD, CDL, CF and CFL has received treatments with fluorine. For a radiation characterization an optical coherence tomography (OCT), a measure of the value of the scattering coefficient of each of the units consisting of ANOVA and T-test, will be used. The OCT detected significant differences in mineral loss between samples in relation to time and type of treatment. The evaluation of the retro-scattering coefficient can surely be used in the in vitro evaluation of the diagnosis and / or progression of incipient lesions of dental enamel caries.

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### PHOTODYNAMIC INACTIVATION MODULATION OF CANDIDA ALBICANS WITH GLUCOSE

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Introduction: In a post-antibiotic era, antimicrobial photodynamic therapy (aPDT) emerges as a promising antimicrobial alternative. The combination of photosensitizer (FS) and light of adequate wavelength in the presence of oxygen can promote a phototoxic response in fungal cells. The expression of ABC family transports may diminish the effect of aPDT, however, association with glucose may be a solution to this problem. Material and Methods: To observe the effects of glucose on microbial inactivation with PDT, C. albicans ATCC10231, YEM 14 and YEM 15 strains were selected and cultured on Sabouraud Dextrose agar and incubated at 37 ° C for 24 hours. The proposed treatments were performed in phosphate buffered saline (PBS) suspensions and glucose at 0mM and 1mM concentrations. Methylene blue (AM) photosensitizer (FS) was added to the suspension at a final concentration of 100 µM. In the microbial inactivation assay 6 groups were used: (control; LED; PDT with time 0, 1, 3 and 6 min). Results: The Control, LED and aPDT 0 groups showed no significant differences with and without glucose. ATCC 10231 showed microbial reduction from 3 min in 0 and 1mM glucose concentrations, however, the reduction was more evident in the presence of glucose. At 6 min, the difference accentuated to 1 log, showing greater reduction in the presence of glucose. In YEM 14, inactivation occurred after 6 min with a reduction of 1 log, with and without glucose. On the other hand, YEM 15 showed inactivation after 3 min with and without glucose, but after 6 minutes, inactivation was higher in the presence of 1mM glucose. Discussion: High glucose concentrations may inhibit aerobic respiration in C. albicans. Thus, investigations should be directed to know the appropriate glucose concentration so that C. albicans can express its glucose transporters, increasing FS uptake and microbial inactivation through aPDT. Conclusions: PDT may be a viable solution for inactivation of resistant C. albicans in the presence of glucose. We can conclude that PDT may be a viable solution for inactivation by resistant C. albicans when glucose is present.

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## EFFECTS OF PHOTOBIOMODULATION IN SALIVARY GLANDS OF CHRONIC KIDNEY DISEASE PATIENTS ON HEMODIALYSIS

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Introduction: Chronic kidney disease (CKD) is a systemic condition characterized by the continuous and irreversible reduction of renal function due to a reduction in the glomerular filtrate rate. It is a worldwide public health question and its incidence has been increasing (comes up piling) worldwide, based on the aging population and the higher incidence of diabetes Melito and arterial hypertension. About 15 million Brazilians have some alteration of renal function which demonstrates a proportion of 50 cases/100,000 inhabitantsObjective: This randomized placebo-controlled trial evaluated the competence of photobiomodulation (PBMT) in salivary glands of patients with chronic kidney disease (CKD) who underwent hemodialysis. Material and methods: Forty-four patients on hemodialysis self-responded two questionnaires for oral health and salivary gland function perception. The subjects were evaluated for hypofunction of salivary glands and randomly allocated to two groups: PBMT group (three irradiations at 808nm, 100mW, 142J/cm2 and 4J per point); and placebo group. Patients were submitted to non-stimulated and stimulated submitted to sialometry and after the treatment at baseline, 7 and 14 days. Results: Salivary volume and biochemical of the saliva were analyzed. At baseline, most subjects had self-perception of poor oral health (52.6%) and salivary dysfunction (63.1%). Clinical exam revealed that 47.3% of subjects presented dry mucosa. PBMT promoted An increase in the volume of saliva collected-not stimulated (P = 0.027) and stimulated (P = 0.014) and the decrease of the urea concentration in both cases: non-stimulated (P = 0.0001) and stimulated saliva (p = 0. 0001). No alteration was detected in total proteins and calcium analysis. No alterations were detected in the total proteins and in the analysis of salivary calcium. Conclusions: These findings suggest that PBMT is effective in decreasing hyposalivation and Urea content in the saliva of patients with chronic renal disease. They may change the volume, concentration, and composition of saliva, which may affect the quality of oral health, but PBMT has the improvement of this oral condition.

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# TREATMENT OF MALE ANDROGENETIC ALOPECIA BY MICRONEEDLING ASSOCIATED TO PHOTOBIOMODULATION – CASE SERIES

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Background: Androgenetic alopecia is a pathology associated to hormonal imbalance. It affects men and women causing reduction and/or absence of hair in a particular region of the scalp. Some of the pharmacological treatments available in the market are inefficient and/or present risks and contraindications. In here, a combination of two minimally invasive techniques (photobiomodulation and microneedling) was proposed as a new treatment protocol. Objective: The objective of this study was to evaluate the efficacy of the association of microneedling to photobiomodulation for the treatment of male androgenetic alopecia. Material and one-month treatment consisted of Methods: The 12 applications of photobiomodulation (three times a week) using low intensity laser (4J per point, 40s, 660nm, 100mW) and two microneedling sessions (0.5mm roller) with a fifteen-day interval between them. The three participants were male, above 25 years old and presenting androgenetic alopecia. The procedures used within this study were approved by the Research Ethics Committee of the Nove de Julho University. The main outcome was the density of hair strands per square centimeter (n/cm2). The participants were asked regarding their dissatisfaction/discomfort with the dysfunction through a scale ranging from 0 to 10, where 0 meant no discomfort and 10 high discomfort. The scalp analysis was performed through a dermatoscope that registered scalp images and a software was used to count the number of strands. The images were recorded before the beginning of the treatment, after the sixth session (day 15), after the twelfth session (day 30) and thirty days after the end of the treatment (day 60). Results: At day 60 the hair count increased from 111.1 hair/cm2 to 140.6 hair/cm2 (pooled data from the three patients). The patients reported reduced dissatisfaction with hair loss after the end of the treatment. Discussion: The increase in hair count from 137.3 hairs/cm2 on baseline to 145.1 hairs/cm2 at the end of 14 weeks of PBM therapy (655 nm and 780 nm, 10 minutes daily, no radiant exposure was mentioned) was reported before, i.e. a 6% increase in hair density. In here, after 4 weeks of the combined treatment (PBM+microneedling) the hair count increased from 111.1 hair/cm2 to 140.6 hair/cm2, a 26% increase in hair density. Conclusions: The pooled data from the three patients showed 26% increase in hair density after the 4 weeks protocol of microneedling associated to photobiomodulation, showing that this is a promising and effective protocol.



# EFFICACY OF PHOTODYNAMIC THERAPY AND SCRAPING FACILITATED BY USE OF THE FIXED ORTHODONTIC APPLIANCE: RANDOMIZED AND DOUBLE-BLIND CONTROLLED CLINICAL STUDY

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Introduction: It is known that the presence of orthodontic brackets predisposes for a change in the quantity and quality of the biofilm, due to the challenge of brushing adequately. The sites are difficult to access with a toothbrush and periodontal curette, worsening inflammation, in addition, gingival hyperplasia is frequently associated. The hypothesis to be tested in this study is whether photodynamic therapy (PDT) could favor the decontamination, as the photosensitizer and light are capable of reaching areas that these instruments have difficulty accessing. Objective: To evaluate the impact of PDT as an adjuvant treatment to scaling, considering clinical immunoregulatory and microbiological parameters in patients with gingivitis with the predisposing factor of the use of a fixed orthodontic appliance. Material and Methods: A randomized, controlled, double-blind clinical study will include 34 patients, both genders, having used the fixed appliance for more than 12 months, with gingivitis present. Patients will be divided into two groups: Control group (n = 17) - Scaling and Root Planing (SRP) + PDT placebo and Experimental group (n = 17) - SRP + PDT. In G2 methylene blue 0.005%,  $\lambda$  = 660nm, 9J per inflamed site, irradiance = 3.5W / cm, radiant exposure = 318J/ cm2. In G1 the scaling will be performed with the aid of curettes and ultrasound. All participants will receive oral hygiene guidance (OHG) prior to SRP. The clinical periodontal data to be analyzed: plague index (PI), gingival index (GI) and probing depth (PD) by means of periodontal probe. Crevicular fluid (from 8 pre-determined sites) and non-stimulated saliva for analysis of the IL-6, IL-1 $\beta$ , IL-8, TNF- $\alpha$  and IL-10 cytokines will be collected using the ELISA method. The Universal bacteria count (16S rRNA gene) will be performed by gPCR. All analysis will be realized using the baseline (T0), 7 (T1) and 21 (T2) days after treatment. Oral health-related quality of life (OHRQoL) will be assessed using the OHIP-14 questionnaire at times T0 and T2. If sample distribution is normal, the Student T test will be used to compare the measurement of continuous and dependent variables. If it is not normal, the Mann-Whitney test will be used. The data will be presented in terms of  $\pm$  PD and the value of p will be defined as 0.05.

Key words: gingivitis, orthodontic appliance, photodynamic therapy.



# EVALUATION OF VO2 MAX, ANAEROBIC LIMIER AND PAIN LEVEL BEFORE AND AFTER AN AEROBIC EXERCISE PROGRAM ASSOCIATED WITH PHOTOBIOMODULATION EFFECTS IN WOMEN WITH KNEE OSTEOARTHRITIS.

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Introduction: The onset of chronic degenerative diseases is closely linked to the aging process. Among these diseases we can highlight osteoarthritis (OA), being predominantly in women. The most common manifested articulation is the knee. About 10% of people with age superior 55 years present pain and inactivation of the knee with OA. Given this, these women present i mpaired aerobics capabilities. One of the recommendations for treatment consist in treatment as aerobic exercise. In addition, features such as photobiomodulation (PBM) have shown promising results in enhancing the b enefits of aerobic exercise. Objectives: To evaluate the effects of an aerobic exercise protocol and the PBM in VO2máx, anaerobic limier and pain level, as well as sitffness and mobility of women with knee OA. Methods: Eleven women aged 55 to 70 years with grade II and or III knee OA were evaluated and randomized into 2 groups: Placebo P BM Exercise Group (PPEG, n=7) and Active P BM Exercise Group (APEG, n=4). The interventions were performed 2x/week for a period of 8 weeks. The protocol included aerobic exercise on ergometric bicycle, stretching of knee muscles groups and application of PBM on the affected knee. The PBM used is the cluster t ype with 14 diodes (8 08 nm, 100mW, 4J/point totaling 56J). Results/ Discussion: The results showed an improvement in the pain of the volunteers In addition, the results obtained through the KOOS questionnal re were also improved in most of the mentioned variables. However, for the cardiorespiratory variables there was not the same result . We will wait for more results until we determine a verdict on the effectiveness of aerobic exercise associated with PBM. Although some numerical data did not show any improvement, most volunteers reported t hat treatment with the aerobic exercise protocol on the ergometric bicycle was very effective in improving the functionality and cardiorespiratory fitness for performing daily tasks and re attending. environments due to symptoms arising from OA. Conclusion: Based on the partial descriptive results obtained in this study it is possible to conclude that the treatment with aerobic exercise protocol on the ergometric bicycle is been capable in improving symptom, stiffness, pain, activities, sports function and quality of life of the volunteers. However it did not occurred the same with the cardiorespiratory variables, despite the reports of the volunteers been the opposite from the obtained data. Given this, the continuity of the study is necessary to better compare the results between and inside the groups.



## THE ASSOCIATION OF PHOTOBIOMODULATION TO MICRONEEDLING IN FEMALE HAIR LOSS. PRELIMINARY RESULTS OF A RANDOMIZED DOUBLE BLIND CLINICAL STUDY

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Introduction: The female pattern hair loss is a clinical presentation commonly found in primary care. It is the progressive miniaturization of the hair follicles, and impact on patients quality of life, causing psychosocial limitations. The photobiomodulation and microneedling are listed as safe and effective tools to treat this condition. The goal of this study is to verify if there is an increase in capillary density of hairs, by combining photobiomodulation with 660nm laser and electronic microneedling. Material and Methods: Patients were divided into 3 groups, the first one including 3 microneedling sessions, the second 36 photobiomodulation sessions and the third one, combination of both techniques. The photobiomodulation was performed with Laser Elite Olympus - DMC, 660± 20 nm, 100mW, 40s per point, 4J, 5.6 W / cm2, 226 J / cm2, 1.5 cm between irradiated points around the affected area at the temporo-parietal region. The microneedling was carried out with an electronic pen (Derma Pen Erase Model DE77, VR Medical, needles 42, speed setting 3, and needle size 0.5 mm). The trichoscopy was performed with a AM7515MZTDino-Lite Edge Equipment. Results: The research is ongoing and the preliminary and partial results has shown that the group treated with combined techniques expressed the best results to date, however, the photobiomodulation group showed interesting results. Discussion: Today, photobiomodulation has proven to be an attractive alternative for treating alopecia, especially for those individuals who do not wish to continue with pharmaceuticals or surgical treatments. The ergonomic microneedling automated device uses and disposable needles, its use favors the action in the capillary region, being less painful and more economical. Its use on the scalp for alopecia is one of its recente advances. It has been compared with drugs in various alopecia, always showing a better one in combination. Conclusions: Preliminary results may suggest valuable clinical evidence leading us to anticipate that the association of resources favor hair growth, and that photobiomodulation, even in isolation, offers a good response to hair treatment. Final results will be published at the end of the study.



## DENTISTRY INTEREST MICROORGANISMS REDUCTION BY INTRAORAL PHOTODYNAMIC THERAPY MEDIATED BY LED ASSOCIATED WITH CURCUMINE AND PORPHYRIN IN AIDS PATIENTS

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Introduction: HIV / AIDS patients express different oral conditions due to immunocompromising, antiretroviral use, antibiotic and antifungal use, and associated infections that compromise oral health. Other factors such as smoking, drinking, use of narcotics, lack of adherence to ARVT, xerostomia and poor oral hygiene aggravate this condition whose treatment can contribute to the generation of resistant microbial strains and interferes with the local immune response of the individual. Thus the use of antimicrobial photodynamic therapy (PDT) for oral microbiota control in these patients emerges as a promising alternative. Objective: To test the effectiveness of total mouth PDT mediated by blue LED (450nm) associated with the curcumin photosensitizer and also the red LED (660nm) associated with porphyrin in AIDS patients. Material and methods: The study was conducted at the STD and AIDS treatment center in Vitória da Conquista-BA, where AIDS patients were screened and randomized between experimental groups. In the group using 0.75mg/mL curcumin (n = 30), it was spraved in all intraoral regions and after 5 minutes in dark environment, blue LED irradiation was performed for 10 minutes (67mW/cm2). and 20.1J/cm2), in the group with porphyrin 50µg/mL (n = 28), it was sprayed intraorally and after 10 minutes in dark environment, red LED irradiation was performed for 10 minutes (125mW/cm2 and 7.5J/cm2). Before and after treatments, saliva samples were collected and seeded in duplicate on BHI agar, Mac Conkey agar, Sabouraud dextrose agar with chloramphenicol, Mannitol agar and Mitis Salivarius agar. After incubation period (24h or 48h at 36°C), the colony forming units (CFU/mL) were counted and the results were submitted to the paired T-test (5%). Results: PDT showed antimicrobial effectiveness in the groups studied in individuals with LTCD4 and LTCD8 above 25% of normality (p <0.05). Conclusion: Curcumin-mediated PDT was effective in reducing oral Streptococcus spp. in AIDS patients with 25% or more than 25% of normal levels of LTCD4 and LTCD8, also reducing Staphylococcus spp., enterobacteria and the total count of microorganisms. Porphyrinmediated PDT was effective in reducing oral Enterobacteria in AIDS patients, either in those with levels greater than or below 25% of normal LTCD4 and in those with LTCD8 above 25% of normal.

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### DEVELOPMENT AND EFFECTIVENESS EVALUATION OF AN ORAL MOUTHWASH WITH NANOSTRUCTURED CURCUMIN FOR ANTIMICROBIAL PHOTODYNAMIC THERAPY

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Introduction: Antimicrobial Photodynamic Therapy is pointed in the national and international literature as a promising new approach as a high efficiency antimicrobial method regardless of the present antibiotic resistance status. However, studies that emphasize the possibility of using this technique to control oral biofilm are still rare. Objective: This research aimed to develop an oral mouthwash based on a nanostructured formulation as a vehicle for the curcumin photosensitizer (FS) and to evaluate the in vitro effectiveness of the product developed, associated with antimicrobial photodynamic therapy (PDT). Materiasl and Methods: A pseudoternary phase diagram (DFPT) was constructed to select the potential microemulsion (ME) formulations to be evaluated. The physicochemical characterization of the chosen ME was performed by means of droplet size, zeta potential, pH, relative density and rheological profile measurements. Analytical methodology for curcumin assay was standardized in the formulation developed by UV/Vis spectrophotometry. The product was subjected to accelerated stability study. The in vitro effectiveness of blue LED (450nm ± 10nm) mediated antimicrobial PDT was evaluated with emission at two radiation times (10 and 30 minutes) and intensity of 18 mW/cm2 associated with the developed formulation. The formulation was tested using FS curcumin at two different concentrations (30 and 60 µg.mL-1) on Candida albicans biofilm (ATCC 90029), methicillin resistant Staphylococcus aureus (ATCC 33591) and Escherichia coli (ATCC 25922). Results: The final product was defined as the composition of Polyethylene glycol 400, polysorbate 80, flaxseed oil, sorbitol, water and curcumin. pH 6.21  $\pm$  (0.036), relative density 1  $\pm$  0.0003 g / ml, droplet size 13.86 ± 0.20 nm, polydispersity index equal to 0.151 ± 0.0565, zeta potential of - 10.3 ± 2.6 and low viscosity Newtonian flow behavior 1.64 ± 0.01 cP. The final formulation was stable during storage under forced conditions. There was significant reduction of CFU/mL in all microorganisms tested (5% ANOVA and Bonferroni post-test). The biggest CFU/ mL reductions were 5.6175 log 10 CFU.mL-1 in the PDT (10 ') and PDT (30') group, both with 30 µg.mL-1for E. coli; 5.181 log 10 CFU.mL-1 in the PDT group (30 ') and 30 µg.mL-1 for MRSA; and 3,497 log 10 CFU.mL-1 in the PDT group (30 '). 60 µg.mL-1 for C. albicans. Conclusion: The developed mouthwash containing nanostructured curcumin is physically stable and effective in vitro for antimicrobial PDT application on biofilm of the tested microorganisms.

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# HYDROXYL RADICAL GENERATION BY PHOTOSENSITIZATION OF PHENOTHIAZINIUM DYES IS AFFECTED BY THE MEDIA

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In Photodynamic therapy (PDT), the therapeutic aid based in the use of a photosensitizer (PS) associated with light and the cell death is triggered by the production of reactive oxygen species (ROS), among them, singlet oxygen (102). Methylene blue (MB) is a phenothiazinium dye that have been studied in several PDT works, mainly due to its high absorption of light in the region between 550-700 nm and low cost, however, it is known that due to aggregation, the production of 1O2 may be compromised depending upon the medium used. Other phenothiazinium such as Azure A (AA), Azure B (AB) and dimethyl methylene blue (DMMB) may also present similar behavior. Thus, the objective of this study was to evaluate the aggregation of this series of compounds and measure the production of ROS in different media (water, phosphate buffer saline - PBS, physiological solution - 0,9% NaCl, urea 1mol/L and sodium dodecylsulfate 0,25% -SDS). The aggregation of the PSs was determined by spectrophotometry, by the calculation of the dimer-monomer ratios (D/M: employing the absorbance values for the dimer and monomer of each compound, respectively). The production of ROS was measured using the p-Nitrosodimethylaniline (RNO). The aggregation tendency observed was smaller for MB, higher for DMMB and intermediate for AB and AA, being slightly bigger for AA. Because they are more hydrophilic, the changes in D/M values of MB, AA and AB between the media is not very high. On the other hand, for the DMMB it is possible to observe the significant changes in the D/M in the different media, being lower in SDS, slightly higher in urea; in water the D/M value is intermediate, increasing in PBS and reaching the maximum in physiological solution. The ROS production increased depending upon the media used, being lower in urea, followed by PBS, NaCl, SDS and it was higher in water. The ROS production measured by RNO showed no relation to aggregation, suggesting that the oxidant species measured were produced by type I reaction. Some studies show that RNO measures production of hydroxyl radicals (•OH), since it do not react with 1O2 and superoxide. Thus, the production of •OH is similar between the compounds, and depends on the media used to deliver the PS. In SDS the •OH production differs between the compounds, being greater for DMMB.

Funding sources. São Paulo Research Foundation.



# PHOTOBIOMODULATION IN CUTANEOUS INJURY CAUSED BY USE OF CIRCULAR EXTERNAL FIXATOR: CASE SERIES

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Background: The Circular External Fixator (CEF) is an orthopedic device made of steel, aluminum or carbon, with the function of maintaining the rigidity or stability of the severely traumatized bone structure, which connects to the CEF through wires and/or pins. The injury to the skin caused by wires and/or pins does not receive as much attention as the bone injury, can leading to complications, such as pain and infection, increasing the recovery time or even leading to new surgery. Purpose: To avoid or alleviate the signs and symptoms of injuries to the skin insertions of wires and pins during the use of CEF. Material and Methods: Conventional treatment with 0.9% saline and essential fatty acids associated with laser photobiomodulation (PBM)  $\lambda$  = 660 nm, beam area of 0.126 cm2, I = 796 mW/cm2, H = 4J/cm2, t = 5s, E = 0.5 J, 4 points per insertion. The interventions occurred from the 3rd postoperative period, every 72h, for 16 weeks. Scale of Bates-Jensen, visual analogic scale of pain and lesion temperature of 6 participants were evaluated. Results are presented as mean ± standard deviation. Results: In the first session, the Bastes-Jensen scale score was  $16 \pm 3$ , pain  $2 \pm 0$  and  $36.1^{\circ}C \pm 6$ . In the 4th session, the Bastes-Jensen scale decreased to  $13 \pm 2$ , pain  $0 \pm 0$ ,  $36.0^{\circ}C \pm 6$ . Discussion and Conclusion: There was adequate healing, improved local pain, decreased inflammation, and restoration of physical function after FBM. New randomized controlled trial will be conducted.



## PHOTODYNAMIC THERAPY IN PREGNANT RATS WITH PERIODONTITIS PARTIALLY REVERSES THE LOW BIRTH WEIGHT WITHOUT ALTER THE PRETERM BIRTH

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Periodontitis (P) is an important buccal disease that affects the periodontium and gradually destroys periodontal tissues. It is controversial the effects of P during the gestation as well as its impact in the offspring. The association of a light source with a photosensitizing agent (PDT, photodynamic therapy) causes microbial death and this treatment emerges as important tool to treat P. Here, our objective was to evaluate the effect of P and its treatment with PDT on the repercussions in the birth weight and preterm birth. Ten days before the pregnancy, P was induced by ligature technique, and the treatment with PDT was performed 15 days after the induction of P. The photosensitizer methylene blue (0.005%, CHIMIOLUX) was administered and the periodontal pockets were irradiated with a red laser (Wavelength 660 ± 10 nm; Radiant power100 mW; Time exposure 90s; Radiant energy 9 J; Irradiance 3.5 W/cm2; Radiant exposure 318 J/cm2; Total radiant energy18 J). The parameters were evaluated immediately after the birth of puppies. Results: We showed that PDT treatment in pregnant rats with P partially reversed the low birth weight as well as the weight gain until weaning. On the other hand, PDT did not reverse the preterm birth. Conclusions: Thus, our data showed the important role of oral health during the gestation and PDT treatment seems to be an important tool in the P. Financial support: This study was sponsored by Fundação de Amparo à Pesquisa do Estado de São Paulo (2017/006444-9).



## EVALUATION OF CANDIDA ALBICANS INACTIVATION BY PHOTODYNAMIC THERAPY MEDIATED BY METHYLENE BLUE ASSOCIATED WITH FLUCONAZOLE

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Introduction: Antimicrobial Photodynamic Therapy (aPDT) offers a promising alternative in the treatment of infectious diseases traditionally treated with drugs. Candida albicans is a fungus that can be lethal when it develops drug resistance to fluconazole, commonly used in the treatment against this microorganism. However, several studies have shown satisfactory results in the treatment of infections caused by C. albicans using aPDT with methylene blue as a photosensitizer. The combination of therapies can provide a beneficial option for patients with diseases that are exclusively treated with drugs, reducing side effects and improving the quality of life. Objective: This study aimed to evaluate the alterations in the C. albicans killing when aPDT with methylene blue (MB) was associated with the antifungal fluconazole in vitro. Material and Methods: In this study, C. albicans were incubated with MB associated or not with fluconazole and were irradiated with 150 mW/cm<sup>2</sup> during 2 to 8 min with a laser emitting radiation of  $\lambda$  = 660 nm. The survival fraction of the cells submitted to aPDT was calculated using the CFU/mL count. In addition, two other analyses were performed, the analysis of MB uptake by quantifying MB outside the cells and the measurement of ROS formation during aPDT based on the degradation of the chemical compound RNO associated or not with fluconazole. Results: Experiments showed that after 2 min and 4 min of irradiation, there was approximately 1 log and 3 log increase in cell death of C. albicans previously incubated with fluconazole for 2 hours. After 8 min of irradiation, all fungal cells were killed. The amount and velocity of ROS generated MB-mediated aPDT was not altered by fluconazole. The analysis of MB diluted in the supernatant showed that fluconazole did not interfere on the MB uptake Discussion: The improvement of aPDT effect was not associated with an increase in the MB uptake or in ROS production. Since fluconazole inhibits ergosterol production, we suggest that the plasmatic membrane of fungal cells were less resistant to oxidative stress after incubation with fluconazole. Conclusion: The in vitro association of fluconazole and MB-mediated aPDT improved the antifungal effect on C. albicans, decreasing the amount of light energy necessary to kill all the fungal cells. These results suggest that the association of these therapeutic modalities may be beneficial for the treatment of infections caused by C. albicans. Acknowledgment: We thank the UFABC and IPEN/USP for the use of their equipment.



# EVALUATION OF THE SENSORY-MOTOR RESPONSE IN PATIENTS WITH MYELOMENINGOCELE FOLLOWING TREATMENT WITH PHOTOBIOMODULATION – STUDY PROTOCOL

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Myelomeningocele is a neural tube defect that occurs between the third and fourth week of gestation. Functional impairment occurs due to dysplasia of the nerve roots, which can lead to flaccid paralysis and sensory-motor dysfunction below the level of the lesion. Although advances in the multidisciplinary treatment of myelomeningocele have led to functional improvements in affected children, novel therapeutic modalities, such as photobiomodulation (PBM), could be a promising complement to treatment. Studies have demonstrated that red and infrared light have the potential to be effective, noninvasive forms of therapy, promoting axonal sprouting and sensory-motor recovery in experimental spinal cord injury models. Objective: Evaluate the effectiveness of PBM combined with physiotherapeutic exercises on the sensory-motor response in individuals with myelomeningocele at the lower lumbar and sacral level. Material and methods: Participants will be recruited from the Integrated Health Clinic of University Nove de Julho and randomized into two groups: Group 1 – treatment with PBM + physical therapy; Group 2 – sham PBM + physical therapy. For irradiation, the individuals will be positioned comfortably in lateral decubitus on the examining table. Four points will be irradiated with LED above the lesion level, wavelength 850 nm, 25 J and 50 seconds per point. The evaluations will involve electromyography of the gastrocnemius, anterior tibial and rectus femoris of both lower limbs. Touch sensation will be evaluated in L1 to S1 dermatomes through the Semmes-Weinstein (Smiles®) monofilament kit. The analyzes will be performed before starting the treatment (T0), after 12 sessions (T1) and after 30 days after the completion of the intervention (follow up) (T2). Results: The data will be analyzed with the aid of GraphPad PRISM version 7.0. The Kolmogorov-Smirnov test will be used to determine the normality of the data. Variables that fit the Gaussian curve will be expressed as mean and standard deviation values. The significance level will be set at  $\alpha = 0.05$ . Funding sources: São Paulo Research Foundation.



# ANTIMICROBIAL PHOTODYNAMIC THERAPY WITH AZURE B IN DIFFERENT MEDIUNS TO TREAT CANDIDA ALBICANS

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Introduction: The antimicrobial photodynamic therapy (aPDT) consists of the use of a photosensitizer (PS), irradiated with light at a specific wavelength in the presence of a microorganism. Methylene blue (MB) is a phenothiazinium PS widely studied in aPDT, but the results are not always effective. In addition to the lack of standardization of the aPDT application parameters, the effectiveness of the photodynamic action is directly related to the MB medium, due to its metachromacy. In view of this context, other phenothiazinium PSs whose photodynamic activity has not yet been studied, may exhibit a similar behavior. Objective: The aim of this study is to compare the efficacy of aPTD with Azure B (AB) conveyed in different media in the inactivation of Candida albicans. Material and methods: The C albicans strain was grown on Sabouraud Agar Dextrose 48 hours prior to the experiment. The cells were harvested, a water suspension was prepared and counted in the Neubauer Chamber. In microplates, were added 200µL of C albicans suspension (1x10 8 cells/mL) and 200µL of AB 0.002% conveyed in: water, PBS, physiological solution, 0.25% SDS, 1 mol/L urea. A control group without PS was also prepared. Cells were incubated in the dark with PS solutions for 5 minutes. It was then irradiated for 30 minutes (640 ± 12.5nm, 2.6mW/cm2, 4.7 J/cm2). At the end, an aliguot of each sample underwent successive dilutions (up to 10-5) that were spread in Sabouraud Dextrose Agar. Plates were held for 24h at 37°C and the number of CFU/mL counted. The experiments were performed in triplicate and three independent experiments of CFU/mL counted. Results: The results showed that AB when carried in media which induces to higher aggregation (PBS and physiological solution) did not reduce the number of CFU/mL in relation to the control group. When conveyed in water, the same occurs. In media where there is reduction of aggregation (SDS and urea) AB inactivate the microorganisms in SDS and no reduction was observed in urea. Conclusions: The medium of FS delivery influences the outcome of antimicrobial photodynamic therapy, but a direct relationship between aggregation and reduction has not been established. AB is more effective in inactivating C albicans when conveyed in SDS medium. Further studies are needed to establish the synergistic mechanism between SDS and AB in antimicrobial photodynamic therapy. The development of formulations for clinical use based on these principles may increase the efficacy of clinical treatments with aPDT.

Funding sources: Fundação de Amparo à Pesquisa do Estado de S. Paulo - FAPESP



# ANTIMICROBIAL EFFECT OF BLUE LIGHT IRRADIATION ON AGGREGATIBACTER ACTINOMYCETEMCOMITANS

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Introduction: Aggregatibacter actinomycetemcomitans is an oral bacteria related to bone loss on periodontal disease. The treatment of periodontitis is scaling and root planning and the resolution gingival inflammation is dependent of microbial biofilm control. Blue light irradiation has been implicated on bacterial inactivation by generating reactive oxygen species inside microbial cell. The aim of this study is to investigate the potential of bacterial kill by blue light. Material and Methods: We used A. actinomycetemcomitans ATCC 29523 and it was cultivated from frozen samples on brain heart infusion (BHI) medium on 5 - 10% CO2 at 37°C. Bacteria was irradiated by LED 403 nm (violet/blue) with output power of 1W, irradiance 854.7 mW/cm2, for 0, 1, 5, and 10 min performing radiant exposure of 0, 51, 256, and 512 J/cm2. Bacterial content was diluted from 10-1 to 10-6 times the original concentration and it was counted and transformed on colony form units (cfu) per mL. The data presented normal distribution and the average of cfu/mL was compared using ANOVA one-way and Tukey test groups were considered different when p<0.05. Results: The experiment was performed 3 times in triplicate. It was observed no bacterial reduction following blue light irradiation until 10 min. Discussion: Blue light active is based on the absorption of light by internal chromophores in bacterial compounds. However, bacterial that presents low level of proto-porphyrin IX, an endogen photosensitizer, could be less sensible to blue light. Conclusions: We concluded that Aggregatibacter actinomycetemcomitans is resistant to blue light inactivation under the parameters used in this study.

KEYWORDS: Antimicrobial photodynamic inactivation, Photodynamic antimicrobial chemotherapy, LED, Periodontal disease, periodontalpathogen Funding sources. The authors thanks CAPES.



## EVALUATION OF DENTAL WHITENING IN-OFFICE WITH LED VIOLET (405NM) WITH AND WITHOUT GEL: A CONTROLLED, RANDOMIZED, DOUBLE-BLIND CLINICAL TRIAL

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Introduction: Tooth whitening can be considered the most conservative cosmetic treatment in dentistry. The major advance in the last years was Violet LED of 405 nm wavelength, capable of producing tooth whitening without the use of chemical agent. This wavelength coincides with the absorption peak of the dentin pigment molecules, interacting selectively by breaking them down into smaller molecules, making the teeth lighter, with less sensibility.Objective:This randomized, controlled and double-blind clinical trial evaluates the effect in tooth color change with the violet LED(405 nm) system used in-office bleaching and tooth sensitivity after whitening. Material and methods: This study has been approved by Ethics Committee (#2.034.518) and registered (NCT03192852). Eighty patients will be randomized into 4 groups: G1 violet LED(n = 20), G2 violet LED + 35% cabarmide peroxide (CP)(n = 20), G3 35% CP(n = 20) and G4 35% hydrogen peroxide (HP)(control group)(n = 20). Colour was measured at baseline (TO), 15 (T6) and 180 (T7) day follow-up using the Vita Classical and the digital Easyshade V spectrophotometer. Tooth sensitivity was recorded by VAS 0-10cm scale at each session before and after whitening. The Psychosocial Impact of Dental Aesthetics Questionnaire (PIDAQ) was applied.Results:Using the visual scale both G1 and G3 perform consistently worse overtime when compared to G4 (p<0.05). G2 over time is not statistically different from G4 (p>0.05), implying that is at least as good G4. These results have been further corroborated using the CieLAB. For PIQAD, there was an overall decrease in score over time for all groups with no difference between them (p>0.05). In G4 there was the consistent reporting of pain, suggesting satisfactory results in terms of whitening at the expense of pain. Conclusions: Given that pain was not consistently reported in G2, one could suggest treatment with LED + 35%CP is as effective as 35% HP in terms of whitening but better in terms of pain. All treatments suggested an improvement in quality of life.

Keys word: dental Bleaching, Violet LED , color , carbamide peroxide, hydrogen peroxide, dental aesthetics

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### EFFECT OF LOW-INTENSITY LASER THERAPY COMPARED TO CURCUMINE PHOTODYNAMIC THERAPY IN PATIENTS WITH ORAL MUCOSITE

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Introduction: Oral Mucositis (OM) refers to erythematous and painful ulcerative lesions of the oral mucosa observed in cancer patients who are treated with chemotherapy and /or radiotherapy. Despite the great progress in our understanding of the biopathology of chemotherapy and /or radiotherapy-induced mucositis, there is still a need for new strategies that can at least mitigate the damage caused during the treatment process. Objective: The aim of this study was to evaluate the effectiveness of Low Intensity Laser Therapy (LLLI) and Curcumin-mediated Photodynamic Therapy (PDT) as an adjuvant to the treatment of cancer patients undergoing chemotherapy or radiotherapy with oral mucositis. Material and methods: Thirty patients were collected who met the inclusion and exclusion criteria of the research. Patients were divided into control group (n=10) treated with nystatin, group LLLI (n=10) treated with laser therapy with a fluency of 1.2 J/cm2 and PDT group (n=10) that were treated with the association of blue LED (450 nm), fluency of 20.1 J/cm2 and curcumin photosensitizer. Samples of saliva were collected after 7, 14, 21 and 30 days of starting treatment and seeded in selective culture for Candida sp. in addition to observing the pain level with Visual Analogic Scale (VAS) and the degree of OM was a evaluated with WHO scale for oral mucositis. Results: Intergroup comparisons indicated that the average percent reduction in Candida yeast after the application of therapies was significantly higher in the PDT group in all four evaluations (7, 14, 21 and 30 days), but with a statistical difference in the evaluations of 21. and 30 days (p < 0.05). As for pain sensitivity, a reduction was observed in the treatment groups compared to the control group after 30 days of treatment and an intragroup reduction after 14, 21 in the PDT, LLLI groups. Regarding the degree of mucositis, there was a statistical difference between the control groups, LLLI and PDT (p <0.05), and the control group showed an increase in the degree of mucositis from its 14th day of treatment, while the treatment groups presented reduction in the degree of mucositis from the 21st day of treatment. Conclusion: We can conclude that curcumin-mediated PDT and LLLI are noninvasive modalities for prevention and management of OM with advantages such as antimicrobial effect on C. albicans, analgesic effect and reduction of inflammation, improving the guality of life of patients with this lesion.

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## THE ASSOCIATION OF PHOTODYNAMIC THERAPY, LLLT AND CELLULOSE MEMBRANE DRESSING IMPROVE PRESSURE LESION HEALING: A CLINICAL STUDY

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Introduction: Pressure lesion (PL) is an area of tissue trauma caused by continuous and prolonged pressure, exceeding the normal capillary pressure applied to the skin and surrounding tissues, causing an ischemia that can lead to cell death. PL constitute a serious public health problem due to the large number of patients with changes in skin integrity. The high number of people with ulcers contributes to the burden on the public budget, as well as interfering with the population's quality of life. Objective: The aim of this study was to evaluate the synergistic action of photodynamic therapy (PDT), low level laser therapy (LLLT) and cellulose membrane dressing to control the contamination and contribute with the PL healing process. Materials and methods: Twenty individuals clinically diagnosed with PLs were divided into 2 groups: GI (control, conventional treatment) and GII (PDT, LLLT and cellulose membrane dressing). All individuals in the sample were followed for 20 weeks and the clinical characteristics, the isolation of the main contaminating microorganisms and the healing by measuring the areas in standardizes digital photographs were evaluated. In group GII, 2 sessions of PDT were performed using 1.5% curcumin emulsion and 450 nm blue LED for 12 minutes. irradiance of 30 mW/cm2 and fluence of 22 J/cm2. Microbiological collections before and after PDT were performed with sterile swabs. LLLT was performed with a 660 nm laser. spot diameter of 0.04 cm2, power of 40 mW, 10 seconds continue application per point, fluency of 10 J/cm2 and irradiance of 1 W/cm2. The LLLT treatment was made twice a week. Clinical characteristics were expressed as numbers and percentage of occurrence. Comparisons before and after PDTs were performed using paired-T test. Comparisons between the areas of the PLs were performed using the Mann Whitney 5% test. Results: There was a statistically significant difference between the collections made before and after PDT for all groups of microorganisms (p < 0.05). From the 10th week of treatment onwards, it was observed that the lesions of the GII group had a greater decrease in the mean area than the GI group, showing a statistically significant difference from the 19th and 20th weeks of treatment. Conclusion: The techniques employed in combination were effective in the treatment of the PLs promoting reduction of contamination and healing time.

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## PHOTOBIOMODULATION ON PROINFLAMMATORY CYTOKINES SYNTHESIS OF MUSCLE CELLS CULTIVATED IN M1 MACROPHAGE CONDITIONED MEDIA

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Introduction: Skeletal muscle is a tissue often involved in injury due its functional role and exhibits a great regeneration capacity related to the presence of satellite cells in the mature muscle fibers. Macrophages are indispensable effector cells since products secreted by them can influence and modulate the immune response during all phases of the inflammatory and tissue repair processes. Photobiomodulation (PBM) with low-level laser therapy has been used to benefit the muscle repair process. In vitro studies using this resource have demonstrated the modulation of muscle cells as well as cells involved in the inflammatory process, especially macrophages. Objective: The aim of this study was to evaluate the effects of PBM on the synthesis of cytokines IL-6 and TNF- $\alpha$  in satellite cells (C2C12) cultivated in the presence of a M1 phenotype macrophageconditioned medium (J774) previously submitted to the same irradiation parameters. Material and methods: J774 macrophages were activated with interferon-y and lipopolysaccharide for 2h to induce the M1 phenotype. Irradiation was performed once using an AlGaAs diode laser (780 nm, 70 mW, 17.5 J/cm2, 15 s, 1 J). C2C12 myoblasts were cultivated in a proliferation medium (DMEM + 10% FBS) and were irradiated with the same parameters used for the macrophages. After PBM, the myoblasts received 30% of M1 macrophage-conditioned medium (MCM1) with irradiated (+PBM) and nonirradiated macrophages. The IL-6 and TNF-α protein levels were detected 24 and 48h after C2C12 irradiation using ELISA kits. Results: Untreated and treated C2C12 myoblasts exhibited lower IL-6 levels in the presence of irradiated MCM1 at 24 and 48h. Treated myoblasts that received MCM1+PBM showed lower TNF-α levels after 24h in comparison to untreated C2C12 in non-irradiated MCM1. After 48h, untreated and treated C2C12 myoblasts exhibited lower TNF- $\alpha$  levels in the presence of MCM1+PBM. Conclusion: PBM performed concomitantly on myoblasts and proinflammatory macrophages was able to modulate the synthesis of IL-6 and TNF- $\alpha$  protein levels, which are important and directly involved in the differentiation of satellite muscle cells during the repair process.

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# LEVEL OF PAIN, BALANCE AND MOBILITY: THE EFFECTS OF PHOTOBIOMODULATION ASSOCIATED WITH AEROBIC EXERCISE IN WOMEN WITH KNEE OSTEOARTHRITIS

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Introduction: Osteoarthritis a chronic degenerative disease being well that lead to progressive loss of articular cartilage clinically reflected by pain, decreased range of motion, muscle weakness and loss of quality of life, especially in the elderly population over 65 years of age. In the context, an approach of non-pharmacological treatments such as physical exercise appears to be effective. In addition, resources like photobiomodulation have shown promising results. Objective: The aim of the study was to investigate the effects of photobiomodulation and aerobic exercise protocol, with a purpose decrease pain, increase mobility and balance. Material and methods: 10 subjects with II and III grade of knee AO were evaluated and randomized in 2 groups: A-Exercise and photobiomodulation Placebo Group (EPPG n= 6): exercise protocol associated with placebo irradiation; B- Exercise and photobiomodulation Active Group (EPAG n=4): treatment through exercise protocol associated with irradiation active. As interventions performed. The exercise program was performed 2x/week and included a warm-up phase, stimulus phase, and cooling phase. The cluster (808nm, 100mW, 4J / point totaling 56J) was applied to the most affected lower limb after exercise. For evaluation and reevaluation, the Knee Injury and Osteoarthritis Result Score (KOOS) and Analogic Visual Scale (AVS) was applied. In addition, 40m Fast Paced Walk Test, 30-second Chair Stand Test, Stair Climb Test and Y test was developed. For a data analysis, normality in terms of information distribution and analysis for a choice of appropriate statistical tests. Results: Regarding the partial results presented, the EVS had demonstrated pain reduction in both groups. The 40m fast paced walk test had presented time decrease of distance coverted in both groups. O sitting and getting up test had presented improvement in 30 seconds in both groups. Stair Climb Test had demonstrated time reduction to accomplishment in both groups. On Y test, there was no improvement to EPAG, on EPPG had improvemente on previous directions and left poster. On KOSS had had improvement related to symptoms of pain, rigidity, day by day activities and life quality in both groups. Although on sporting activies the average kept equal to EPAG. Conclusion: These results are partial and the continuious of this work becomes important in order to obtain the statistical results with final sample initially calculated. From this, we will can concluded about the effect of FBM on enhancing an aerobic exercise program for women with knee OA.

Funding sources: São Paulo Research Foundation.



# EFFECT OF PHOTODYNAMIC THERAPY ON HALITOSIS TREATMENT IN DENTATE PATIENTS

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Introduction: Halitosis is a symptom that manifests as an unpleasant odor emanating from the mouth, often demonstrating any change in the organism of the individual. A recent meta-analysis shows an increased prevalence of halitosis in the last years. Photodynamic therapy is very effective in eliminating halitosis. Although, studies shown halitosis returns in a week. Probably, it happens because of the bacteria in the dental sulcus could recolonize the dorsum of the tongue. Until nowadays, there is no study in adult population that associates halitosis and periodontal treatment with follow-up evaluation. Objective: The aim of this randomized, controlled, single-blinded clinical trial was to treat oral halitosis in healthy adults with photodynamic therapy (PDT), associated with periodontal treatment with 3 months follow up. Material and methods: Adult participants presenting at least 10 dental elements in the oral cavity and with halitosis were randomized into two groups. G1(experimental) PDT treatment and G2(control) Tongue scraper. In both groups, scraping and root planing were performed with ultrasound when needed. Halitosis was evaluated by quantifying volatile sulfur compounds -H2S- Hydrogen sulfide, CH3SH- metilmercaptanas e CH3SCH3dimetilsulfeto, which were measured by gas chromatography. Oral health-related guality of life was assessed using the OHIP-14 questionnaire. The outcomes were evaluated at baseline, 1 week and 3 months. The nonparametric Kruskal-Wallis test for statistical analysis was applied, comparisons were made with anova test, with the level of significance of 5%. Results: PDT group was more effective in decreasing H2S than scraper group (p<0,008). After 3 months the difference between groups decreased (p <0.019) however. PDT still had a better effect. The same was not observed when using CH3SH- and CH3SCH3- gases as outcome variables (p < 0.05) because there was no difference between groups and between measurements (p > 0.05). For total OHIP scores and its domains, no difference was found (p> 0.05) between groups in any time. Therefore, PDT showed good results when associated with periodontal treatment after 3 months. Conclusion: These results indicate that bacteria residing elsewhere could not recolonize the back of the tongue after the 3 months. Both treatments were not able to improve the quality of life of the participants in any of the evaluated moments, probably because there is no great aesthetic interference and often the patient himself is not aware, as it depends on the feedback of another individual to make patients aware. Funding sources: No.

Acknowledgment: No.



## EVALUATION OF PHOTODYNAMIC THERAPY IN PERICORONITIS: PROTOCOL OF RANDOMIZED, CONTROLLED, DOUBLE-BLIND STUDY

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Introduction: Pericoronitis is a common disease in the eruption phase of third molars, sometimes debilitating, with an impact on the quality of life. The most indicated treatment in the initial phase is the irrigation for cleanliness of the region. In order to reduce the chances of systemic dissemination of the infection and antibiotics use, it is mandatory to test effective treatments in the initial phase of pericoronitis avoiding the evolution of the infectious disease. Photodynamic therapy (PDT) is an interesting alternative because it is an effective antimicrobial treatment that is easy to perform and does not select bacterial resistance. The methylene blue (MB) used in PDT has been studied in an oral formulation, which optimizes the formation of monomers increasing its antimicrobial action. Objective: The aim of this study is to evaluate the effectiveness of PDT with MB in pericoronitis on the initial phase in healthy patients through microbiological, clinical, and immune response and to assess the effectiveness of PDT with MB in an astringent vehicle The impact of pericoronitis on oral health-related quality of life (OHRQoL) of these patients will also be evaluated. Material and methods: In this randomized, controlled, double-blind clinical bioequivalence protocol, 64 healthy patients with pericoronitis will be evaluated. Patients will be randomized into the positive control group (G1) (n = 32): irrigation with sterile saline and PDT (conventional MB at 0.005% concentration and irradiation with low intensity laser  $\lambda$  = 660 nm, 9J per point and radiant exposure of 318 J/cm), and the experimental group (G2) (n = 32): treatment identical to G1, however, MB will be delivered in a new formulation for oral use. Microbiological analysis will be performed by RT-PCR for the bacterium Tannerella forsythia. Gingival crevicular fluid and saliva will be collected to evaluate cytokines by Luminex assay (Luminex Corporation, Austin, TX). The pain (visual analogue scale), swelling and buccal opening (digital caliper), and OHRQoL will also be evaluated through the OHIP-14 questionnaire. The variables will be evaluated in T1 (baseline) and T2 (4th day after PDT). Registration: clinicaltrials.gov NCT03576105. Registered in July 2018.

Funding sources: The study will not receive funding. It will be carried out with funding from the researchers themselves. Acknowledgments: The authors have no conflicts of interest to disclose. The individual data of the patients will remain confidential. The results of this study may be presented at international conferences and published. All data will be saved on the laboratory of the Department of Biophotonics Applied to Health Sciences of Universidade Nove de Julho, São Paulo, Brazil.



# THE ROLE OF PHOTOBIOMODULATION ON THE PARAQUAT-INDUCED PULMONARY FIBROSIS

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Introduction: Pulmonary fibrosis (PF) is a chronic and progressive lung disease characterized by progressive lesion of the pulmonary parenchyma, inflammatory infiltrate and interstitial fibrosis. It is triggered by the excessive and disordered deposition of collagen and other extracellular matrix components, which results in severe changes in the architecture of the alveolus wall. Several factors can trigger PF among them exposure to chemical agents such as paraquat. Due to the absence of an effective treatment, the objective of the study was to investigate the effect of treatment with photobiomodulation on the course of PF.

Methods: Adult male C57BL6 mice were submitted to the induction of PF by the administration of Paraquat (10mg / kg, ip) and after 7 days of induction, the mice were treated during 7 days with Photobiomodulation (LED). Device specifications: BioLambda Apparatus LEDsabr, São Paulo, Brazil; Probe Design, Single Probe; Wavelength: 660 nm; Radiant Power: 160 mW; Power Density: 38,5 mW/cm2; spot area: 4,15 cm2; Density of energy: 5,8 J/cm2; Issuance: Continuous (cw); Total Radiant Emission: 24 J. Treatment specifications: Exposure time: 152 s; Irradiated points: 1 point; Irradiation Method: Direct skin contact; Anatomical location: trachea and lungs; Irradiation rhythm: punctual; Number of treatments: 1 day, seven applications; Optical properties of tissue: Healthy tissue; Animals not shaved. These parameters were measured.

Results: We showed that photobiomodulation reduced Paraquat-induced cell influx into the bronchoalveolar lavage and elevates the level of resolvin D1 without alter the levels of IL-6, TNF- $\alpha$ , IL-10, and IL-17A in the lung homogenates. In addition, did not alter the tracheal responsiveness.

Conclusions: The reduced cell migration induced by photobiomodulation might be attributed, at least in part, to elevated level of resolvin D1. Thus, photobiomodulation althought did not alter some parameters, showed beneficial effects on the inflammation and more studies are needed.

Ethical Approval and accordance: This study were performed according to the Brazilian Society of Laboratory Animals Science (Sociedade Brasileira de Ciência em Animais de Laboratório; SBCAL) and approved by the Animal Care Committee University Nove de Julho (CoEP-UNINOVE, AN0017/2016).

Conflict of interest: The authors declare that there are no conflicts of interest regarding the publication of this paper.

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# EFFECT PHOTOMODULADOR OF RED LIGHT ON GROWTH OF BIPOLARIS SOROKINIANA AND ANTIFUNGAL ACTIVITY WITH GUINE HIDROALCOOLIC EXTRACT

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Introduction: Barley is one of the major crops in the world, and spot blotch, caused by the fungus Bipolaris sorokiniana (Bs), is an important disease that affects it. One of the ways to control disease is the use of fungicides that can cause problems in the environment. In order to avoid risks, we sought to find a natural product that acts as an antifungal. In this case, the hydroalcoholic leaf extract of Petiveria alliacea L., popularly known as guines, was using for the ability to control disease. Objective: The aim of this study was to evaluate in vitro the effects photomodulator of red light on growth of Bs and antifungal activity with guine hidroalcohlic extract. Material and methods: Isolate from Bs came from Agraria Foundation (Paraná, SP) and growth in potato-agar-dextrose medium (PDA). 100g of dried guinea leaves was percolating into 100mL of 70% alcohol in one week. The extract showed 0.2mg protein and 0.093mg flavonoids. Twelve Petri dishes containing 10mL PDA medium were prepared and divided into 4 groups: 1) control plates + 100uL 70% alcohol, 2) control plates + 100uL alcohol + laser, 3) plates with 100uL extract, 4) plates with 100uL extract + Laser. The Laser (DMC-Therapy XT equipment) with a wave emission of 650 nm with a power of 30mW and a dose of 3J/cm2 for 30 seconds per application, being done at 5 and 7 days. The application was made at 1cm for the surface of the plate. After periods of 2, 5, 7 and 12 days, photos and measurements of the area were performed by the Image J software. Statistical analysis t test were performed in the 3 repetitions. At the end of 12 days, the conidia were removing from the plates and counted. Results: The guinea extract showed antifungal effect because there was a reduction of growth in 45% compared to control plate. Already observing the development of plate laser control, plate guinea laser with the plate control the reduction was only 6.5%. This indicated a photomodulating effect. When analyzing the guinea extract that have as antifungal effect. Control plate present 7x105 conidia and laser control plate 6x105 conidia without significant difference. The same occurred in plate with guinea+laser (1x105conidia) and plate with only guinea (3x105conidia). However, the guinea+laser extract plate (1x106 conidia) and laser control plate (6x105 conidia) presented significant difference. Conclusion: Guinea extract can inhibit new formation conidia and mycelium (antifungal) but Laser (650nm) treatment can be maintenance mycelium growth (photomodulador).



# EVALUATION OF THE CONCENTRATIONS OF INFLAMMATORY, PROTEIN AND OXIDATIVE BIOMARKERS OF PARKINSON'S DISEASE AFTER PHOTOBIOMODULATION FROM SUBLINGUAL LASER APPLICATION -CLINICAL, RANDOMIZED AND BLIND TEST – STUDY PROTOCOL

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Parkinson's disease (PD) is a multifactorial, complex, progressive and chronic syndrome, that leads to degeneration of dopaminergic neurons of the substantia nigra, has different subclinical types besides a great variability of changes during its course. It is a very common neurological condition and is considered the most prevalent motor alteration disease and the second most common neurodegenerative disease after Alzheimer's disease. The etiology of PD has not yet been fully elucidated, but evidences point to the involvement of mechanisms such as inflammatory process, mitochondrial dysfunction and oxidative stress both in the origin and progression of the disease. Positive findings related to the use of photobiomodulation in patients with neurological conditions cause a growing interest in the use of photobiomodulation as an aid in the treatment of diseases such as PD. In vitro and in vivo studies (animal models) that mimic PD and which used infrared light as treatment observed an increase in dopaminergic cell concentration, improved mitochondrial function, increased ATP production, and reduced clinical (motor) signs of the disease. Material and methods: Participants will be recruited from the Integrated Health Clinic of University Nove de Julho and randomized into two groups: Group 1 – treatment with PBM + physical therapy; Group 2 – sham PBM + physical therapy. For irradiation, the individuals will be positioned comfortably in dorsal decubitus on the examining table, a sub lingual irradiation will be performed at a single point, wavelength 808 nm, 36 J for 360 seconds. The effects on the concentration of inflammatory markers, oxidative stress, quality of life, clinical signs and functional capacity will be analyzed. The analyzes will be performed before starting the treatment (T0), after 18 sessions (T1). Results: The data will be analyzed with the aid of GraphPad PRISM version 7.0. The Kolmogorov-Smirnov test will be used to determine the normality of the data. Variables that fit the Gaussian curve will be expressed as mean and standard deviation values. The significance level will be set at  $\alpha = 0.05$ .

Funding sources: Universidade Nove de Julho.



# EVALUATION OF THE EFFECTS OF PHOTOBIOMODULATION ON ORTHODONTIC MOVEMENT OF MOLAR VERTICALIZATION WITH MINI-IMPLANT: A RANDOMIZED DOUBLE-BLIND PILOT STUDY

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Introduction: Loss of a dental element can generate several repercussions in the stomatognathic system. According to the latest survey by the Ministry of Health, in 2010, Brazilian adults had, on average, 7 missing teeth. This loss may lead to movement of the adjacent teeth and the antagonist, which would make prosthetic rehabilitation harder to do. Anchoring systems, such as mini-implants, have been increasingly used as a treatment option because they act with heavy but controlled forces and without side effects. Recent studies have shown that photobiomodulation (PBM) can accelerate orthodontic movement in molar intrusion. The objective of this study will be to evaluate the effect of photobiomodulation on the acceleration of the orthodontic movement of molar verticalization and its effect on pain and inflammation of the periodontal tissues. Material and Methods: Thirty-four healthy patients aged 30-60 years, who need to recover the prosthetic space for oral rehabilitation after loss of the posterior inferior dental elements and inclination of the adjacent element, will be randomly divided into 2 groups: G1 (control group) - verticalization by mini-implant + PBM simulation (placebo); G2 (experimental group) - verticalization by mini-implant + PBM. The movements will occur with the aid of mini-implants and elastomeric chains ligatures. The PBM will occur with diode laser application, 660 nm, 100mW, receiving 1J per point, 10s, 10 points (5 per buccal and 5 per lingual) and radiant exposure of 25 J / cm<sup>2</sup>. The orthodontic forces of verticalization (corresponding to any exchange of elastomeric ligation) will be applied every 30 days and the PBM will be applied immediately, 3 and 7 days of each month, for a period of 3 months. The crevicular gingival fluid (FGC) will be collected on the 1st, 3rd and 7th days after the first activation, and then on the 3rd day of the following two months. Interleukins IL1 $\beta$ , IL-6, IL-8, IL-10 and TNF- $\alpha$  will be analyzed by ELISA. Radiographic shots will be taken each month to ascertain the amount (in degrees) of verticalization. To evaluate the pain, the Visual Analogue Scale will be used in all the consultations, and to evaluate the quality of life, the OHIP-14 questionnaire will be applied. Analgesics will be given and the quantity of drugs will be counted. If the data are normal, they will be submitted to Student's t-test. The data will be presented as means  $\pm$  SD and the value of p will be defined as <0.05.

Key words: orthodontic treatment, photobiomodulation, mini-implant, verticalization of molars



## EFFECT OF PHOTOBIOMODULATION WITH LED (LIGHT EMITTING DIODE) IN ANALGESIA DURING LABOR

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Introduction: Pain during labor may be considered as an impediment for choosing a vaginal delivery. In an attempt to reduce pain during labor, several pharmacological and non-pharmacological methods are proposed. In the search for alternatives to pharmacological treatment for analgesia during labor which are effective and to be used in both public and private institutions Objective: The aim the of this research was to evaluate the effect of photobiomodulation with LED (Light Emitting Diode) in analgesia during labor Material and methods: , a clinical trial was performed with 29 women divided into two groups: G1 (experimental group-LED) and G2 (Control group - hot bath). In the experimental group, the LED plate (red and infrared) was placed in the dorsal region of the patient from T10 to S4 level for 10 minutes, and the plate switched off automatically. The hot temperature controlled bath was performed for 30 minutes. To verify the effect of LED on analgesia during labor, the following variables were evaluated: (1) perception of pain; (2) fetal conditions using cardiotocography or intermittent fetal auscultation; (3) Apgar score in the first and fifth minutes after birth and (4) time of delivery. Results: A statistically significant difference (p < 0.05) in pain reduction was observed by means visual scale before and after application in G1 - LED (7.92 + - 1.78). Regarding the other variables, there was no statistically significant difference between the groups when comparing fetal conditions, Apgar score and delivery time. Conclusion: LED can be considered an alternative, since it had as effect the reduction of the pain without changes in the other parameters during labor when comparing with the hot bath method used as protocol in the hospitals, demonstrating to be safe. It is suggested, however, the continuity in the clinical trials with larger samples and possible dosimetric adjustments to establish a photobiomodulation protocol for analgesia during childbirth.

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# ARTIFICIAL INTELLIGENCE IN THE DIAGNOSIS AND DETECTION OF INCIPIENT TOOTH DECAY AND EROSION INJURIES

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Introduction: The present study presents the evolution of the different methods applied for the segmentatio of laser speckle images - LSI applied to dental tissue lesions, specifically caries and erosion, which are in the early stages of development but of great relevance to several countries around the world. The application of computational vision methods associated to mathematical analysis proved to be a path rich in possibilities of automatic injury detection, currently applying AI methods such as Artificial Neural Networks and machine learning yields very satisfactory results. Objective: Through laser speckle imaging (LSI) and AI techniques develop a method of automatic detection and quantification of incipient tooth lesions, more specifically erosion lesions and caries lesions in parallel working also develop an intraoral equipment for Online capture of laser speckle images. Material and methods: We used the photo bank of erosion and carie lesions used in previous experiments. We divided these images into small windows and added to the vector statistical information calculated on them and classified the vectors into lesion vectors and non-lesion vectors. With this new vector bank we trained an Artificial Neural Network (RNA) getting a lesion classifier. We also used a commercial intraoral usb camera and adapted a 5mW laser for speckle laser frame generation to capture new images online. Results: We still have partial results. It creates a healthy and injured image bank and, consequently, a lesion-free or lesion vector bank for RNA training or machine learning. After training the RNA algorithm, is better and more efficient automatic detection of injury to teeth. Conclusion: Previous results indicate that the AI or RNA method becomes an efficient and fast way to detect dental injuries. We are now working on the algorithm to quantify this injury according to ICDAS.



# EVALUATION OF GINGIVAL INFLAMMATION AFTER IN-OFFICE DENTAL WHITENING WITH VIOLET LED (405NM) WITHOUT GEL: SPLIT-MOUTH MODEL

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Introduction: In-office tooth whitening is described in the literature using high concentrations of chemical agents that may be responsible for gingival irritation and dentin sensitivity problems. Although with well-executed gingival protection, the peroxide present in the gels may still pass to the tissues, causing damage to the gingival tissues, even if temporary. The Violet LED (405 nm) capable of producing tooth whitening without the use of chemical agent and can be used in-office. This wavelength coincides with the absorption peak of the dentin pigment molecules, interacting selectively by breaking them down into smaller molecules, making the teeth lighter. The effect of this light on gum tissue is not yet well understood. Objetive: To evaluate the degree of gingival tissue inflammation and cytokine IL-6 profile in gingival crevicular fluid 48 hours after Violet LED irradiation during in-office whitening. Material and Methods: This study has been approved by Ethics Committee (#2.034.518) and registered (NCT03192852). Thirteen participants with clinical indication for gingivoloplasty were treated. First session was of whitening with violet LED with partial gingival protection on the right side and total on the left side (split-mouth design). Forty-eight hours after being irradiated with violet LED, the gingival crevicular fluid was collected by means of an absorbent paper cone. It was introduced into the right and left gingival groove. The cones were placed in PBS (Phosphate-Buffered Saline) solution. The tissue was surgically removed by the gingivoplasty technique and submitted to histomorphometric evaluation. The left side was the control, where gingival protection was total. The 2nd, 3rd and 4th whitening sessions were performed only to contemplate the aesthetic treatment. Results: Measurement of the intensity of inflammation was assessed through the performance of scores: (0) - Absence of inflammation; (1) - Mild inflammation; (2) - Moderate inflammation and (3) - Severe inflammation. Crevicular gingival fluid analysis was performed using the t test and was used with a global level of 95% reliability (critical t = 2.6737) and p> 0.05. Is not statiscally different (p> 0.05) in inflammation between the irradiated side (right side) and the non-irradiated side (left side) of the gum. There was no statistically significant difference between right and left side (p> 0.05). Conclusions: No need to put gum protection barrier for Violet LED whitening treatment.

Funding sources: Own financial resources

Keysword: Gingivoplasty, Violet LED, gum, gingival crevicular fluid



## ANALYSIS OF MUSCULAR INJURY REPAIR IN RATS SUBMITTED TO SYSTEMIC PHOTOBIOMODULATION

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Introduction: Systemic photobiomodulation (SPBM) shows a biomodulatory, analgesic, immunomodulatory, vasodilatory and anti-inflammatory properties and is used to treat several diseases, including tissue repair, atherosclerosis, systemic arterial hypertension, acute myocardial infarction and stroke. In muscle acute injury, PBM is able to reduce edema, myonecrosis, leukocyte influx and leading to better collagen distribution and accelerating the repair process. However, little is known about the effects of SPBM on muscle repair. Objective: The aim of this study was to analyze the evolution of the injured muscle area's dimension in rats subjected to cryoinjury and SPBM (preventive or therapeutic). Material and methods: Twenty-eight male Wistar rats were used (CEUA no 8446031018). The animals were randomly assigned to the following groups: control: not subjected to cryoinjury or SPBM (n = 4); injury + simulated SPBM (injury): subjected to cryoinjury of tibialis anterior (TA) muscle and application of SPBM with the device turned off (n = 8); SPBM + injury: subjected to SPBM before TA muscle injury (n = 8); and injury + SPBM: subjected to TA muscle injury and subsequently treated with SPBM (n = 8). The SPBM was applied punctual in contact mode on the tail of the animals. The following parameters were used: 780 nm, 40 mW, 1 W/cm2, 10 J/cm2, 80s, 3.2 J. SPBM was performed once 24 h before the injury (SPBM + injury) or 2 h after the injury (injury, injury + SPBM). Next, animals received daily SPBM for 5 and 7 days, totaling 5 or 7 sessions. Four animals were euthanized on day 5 and 7 after the injury, and the TA muscle was removed for analysis. Injured area was evaluated by photos and measurements of the area were performed by the Image J software. Statistical analysis t test was performed in the 3 repetitions. Results: The results showed that the injury group had an average injured muscle area of 5,506 cm2 and 5,583 cm2, on days 5 and 7, respectively. In relation to the injury group at day 5, the animals from the SPBM + injury and injury + SPBM groups showed a reduction of 83.82% and 71.28%, respectively. At day 7, animals from the SPBM + injury and injury + SPBM groups demonstrated a significant decrease in the percentage of injured area when compared to the injury group. Conclusion: The results showed that SPBM was effective in reducing the injured area in acute muscle injuries in rats.

Funding source: UNINOVE



# PHOTOBIOMODULATION DECREASES PAIN AND IMPROVE CLINICAL ASPECTS IN PATIENTS WITH ORAL LICHEN PLANUS

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Introduction: Oral lichen planus (OLP) is an inflammatory mucocutaneous disease with different clinical presentations. Conventional treatment is the use of corticosteroids, associated with adverse effects and recurrence. Photobiomodulation (FBM) has been indicated as a therapeutic alternative, although the literature is controversial regarding its efficacy. Purpose: This study evaluated the effect of FBM on the treatment of OLP compared to corticosteroids. Material and Methods: Thirty-six patients with symptomatic OLP were included in the study and randomized into two groups: control (n = 18), treated with 0.05% clobetasol propionate 3 times a day for 30 consecutive days and with laser turned off to mask the treatment and PBM (n = 18), treatment with low level laser (660nm, 100mW, 177J / cm2, 5s, 0.5J per point) twice a week for 30 consecutive days and placebo gel 3 times daily during treatment to mask the therapy. The clinical aspect of OLP was assessed by scores and pain by VAS scale at the beginning of treatment (D0) and weekly during treatment (D7, D14, D21 and D30). Quality of life was assessed using the OHIP-14 questionnaire where items are distributed according to the following subscales: functional limitation, pain, psychological discomfort, physical disability, psychological disability, social disability and disability. Patients answered the questionnaire on the anamnesis day (baseline - D0) and on the 30th (D30). Results: Significant pain reduction was observed between D0 and D30 in the control group and between D0 and D14, D21 and D30 in the FBM group. Regarding the clinical aspect, significant improvement was observed in both groups at D14, D21 and D30 when compared to D0. There was no difference between the groups. Regarding quality of life, the improvement was more significant in the laser group. Discussion and conclusion: FBM has been shown to be as effective as corticosteroids in reducing pain and improving the clinical appearance of OLP, with the advantage of no side effects. The improvement in quality of life was higher in the laser group.



### EVALUATION OF INFRARED LED THERAPY ON THE IL-6 PROTEIN PRODUCTION BY HUMAN MONOCYTES POLARIZED TO PROINFLAMMATORY PHENOTYPE

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Introduction: Monocytes originate from myeloid stem cells and represent 10% of human leukocytes. After an injury, these cells can be recruited into the tissue, and can differentiate into macrophages or dendritic cells, which act to clean the inflammatory microenvironment by eliminating apoptotic cell debris. Monocytes are the major actors in the modulation of inflammation by the production of cytokines such as IL-1 $\beta$ , IL-6 and TNF- $\alpha$ . In this sense, several studies have demonstrated that photobiomodulation (Laser and LED) is an important therapeutic option in cases of tissue lesions. Objective: The aim of this study was to evaluate the effect of 850 nm Infrared LED therapy on the IL-6 protein production by human monocytes polarized to proinflammatory phenotype. Material and methods: Primary cultures from human peripheral blood samples were used. The cells were polarized with LPS for 2h, washed and irradiated with 850 nm infrared LED using 1J, 2J, 4J and 8J energy doses (single application after 2h of polarization). After the cells were incubated at 37°C for 24h, supernatants from the different experimental groups were collected for analysis of protein synthesis by enzyme immunoassay (ELISA). The experiments were performed in duplicate, using three blood samples from different individuals. Non-irradiated and non-polarized cells were used as control groups. All results were submitted to statistical analysis. Results: Monocytes polarized to proinflammatory phenotype (with LPS) showed a statistically significant increase in IL-6 protein production compared to non-activated and non-irradiated monocytes (control group). The treatment with infrared LED (1 Joule of energy) was able to reduce IL-6 protein syntheses in the irradiated monocytes polarized with LPS compared to monocytes which were only polarized. On the other hand, there was no statistically significant difference observed in the evaluations of infrared LED using 2J, 4J and 8J of energy. Conclusion: These results demonstrate the importance of dosimetry evaluation when choosing a light source for Biophotonics application and evidence the ability of the infrared wavelength to reduce the synthesis of inflammatory cytokine IL-6. Keywords: Monocytes; Inflammation; IL-6; Photobiomodulation; Ledtherapy.

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# APPLICATION OF PHOTOBIOMODULATION BEFORE OR AFTER HIGH-INTENSITY RESISTIVE EXERCISE HAS SIMILAR IMPACT ON MUSCULAR OXIDATIVE STRESS?

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Introduction: Acute exercise can significantly break homeostasis by inducing muscle damage, inflammation and oxidative stress. Recent data show that the application of low-level laser therapy (LLLT) before a high-intensity resistive exercise (RE) session attenuates muscle oxidative stress. However, when is the best moment to apply light (before- or after-RE) remain unclear. Therefore, the present study aimed to investigate the effect of LLLT before or after a RE session on muscle oxidative stress in rats. Material and Methods: Female Wistar rats were initially allowed to adapt to the act of climbing for three consecutive days and following (post-24 h rest period) the maximum load test. Rats were then kept at rest for 72 h and were assigned to one of the following experimental groups: Sham, non-exercised submitted to placebo-LLLT; RE, exercised submitted to placebo-LLLT; L+RE, pre-exercise LLLT; RE+L, post-exercise LLLT. RE session comprised four climbs bearing the maximum load with a 2 min time interval between each climb. An 830-nm DMC Lase Photon III was applied on three points in gastrocnemius muscles (two limbs) with the radiant energy of 4J. Animals were euthanized 24 hours after the exercise ended, and muscle tissue was removed to evaluate malondialdehyde (MDA), oxidized protein (OP) and nitric oxide (NO) levels. Activity of superoxide dismutase (SOD), catalase (CAT) and glutathione peroxidase (GPx) was also assessed. Results and Discussion: MDA and OP levels were significantly higher in the RE group, meanwhile CAT activity was downregulated by RE. All LLLT groups had prevention of increased OP, however, increased MDA was prevented only in the RE+L group. These positive results were associated with higher SOD activity. On the other hand, significant differences in GPx were observed only between L+RE and Sham groups. No significant treatment effect was found for CAT activity. There was no effect of RE and LLLT on NO levels. Conclusion: The moment to apply photobiomodulation therapy was decisive to obtain full positive results in prevents muscle lipoperoxidation and Keywords: protein oxidation. low-level laser therapy: oxidative stress; photobiomodulation; resistance exercise.



## METHYLENE BLUE ADMINISTRATION MEDIUM AND IRRADIANCE AFFECT PHOTODYNAMIC THERAPY EFFICACY

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Introduction Staphylococcus aureus is facultative anaerobic and gram positive. It can adapt quickly in unfavorable environments in pH, humidity, osmotic pressure and nutrient deficiency. Photodynamic Therapy (PDT) is a clinical treatment modality with potential application in the elimination of cells and microorganisms, using light at appropriate wavelength and a photosensitizing agent in the presence of oxygen. Phenothiazinium dyes, such as Methylene Blue (MB), can be used as a photosensitizing agent in PDT because it has high light absorption above 600nm. The PDT with MB may be effective against S. aureus, however the protocol parameters are essential for the success of the treatment. Objective: The aim of this study was to compare both the effect of MB media of administration and irradiance on the reduction of Staphylococcus aureus suspension in vitro. Material and Methods: The MB was used at 20µg/mL and incubated in the dark for 5 minutes with the bacterial inoculum (3,0 x107). The assays were performed in water and in BHI medium (Brain Heart Infusion), and these media without MB were used as control. For photodynamic treatment, the 660nm LED system (BioLambda, Brazil) was used. The samples were exposed to light during 30, at 18,6; 26.1 and 37.3 mW/cm2 irradiance. After treatment, the samples were cultured on 1% sucrose BHI agar and kept at 37 ° C for 24 hours in aerophilia. Results: In BHI medium there was no bacterial reduction in any of the parameters used, either in the control group (without FS) or in the treated group. On the other hand, in water there was no bacterial reduction in the groups without FS in all irradiance used while the treatments with MB caused a reduction in S. aureus growth (from 1 to 2 Log UCF / mL) which was higher the higher the irradiance used. Discussion: It is known that MB presents metachromacy, which means that it forms dimers or higher aggregates depending upon the medium. In here, the aggregation state of MB varies from water to BHI, being more effective to reduce microorganisms in water, due to bigger presence of monomers, which are photochemically more active than dimers. Conclusions: This study demonstrated that the medium used influences the efficacy of photodynamic therapy with MB, being more effective in water than in BHI. It was also possible to demonstrate a relationship between irradiance and efficacy, so that the in vitro reduction of S. aureus is greater the higher the irradiance used. Funding sources. São Paulo Research Foundation



## OPTICAL PROPERTIES OF HEALTHY AND BURNT MOUSE SKIN

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Introduction: The interaction between light and biological tissue occurs through reflection, refraction, absorption, scattering, and transmittance. Determining such properties is important for optical devices to be properly designed, diagnostic signals to be well interpreted, and for photonic therapy protocols to be safe once the optical properties of tissue affect the light's behavior. Diffuse optical spectroscopy has advanced as a promising tool for non-invasive determination of reflectance and transmittance. Once this data is collected, the Kubelka-Munk model for the indirect acquisition of the other parameters can be used to determine absorption coefficient, reduced scattering coefficient, reduced attenuation coefficient, and penetration depth. Since the skin is the major physical barrier during photonic therapies and that injured tissue behaves differently compared to healthy tissue, it is important that the optical properties of the skin be known in both physiological states. Objective: The aim of this study was to analyze the optical differences between healthy rat skin and rat skin after burn induction. Material and methods: Eighteen Wistar rats were divided into two groups: HG, which corresponds to the group of healthy animals; and BG, which corresponds to the group of animals that suffered induction of scalding burn. Through spectroscopic analysis, skin biopsies had their transmittance and reflectance values collected between 350 and 1400 nm wavelengths. Data was then processed by the Kubelka-Munk method to obtain the absorption coefficient, reduced scattering coefficient, and reduced attenuation coefficient. Results: Three days after burn induction the absorption coefficient, reduced scattering coefficient, and reduced attenuation coefficient was higher in group HG. Seven days after burn induction all the coefficients studied was higher in group BG. Fourteen days after burn induction, the absorption and reduced attenuation coefficients remained higher in group BG, while the reduced scattering coefficient was higher in group HG. Conclusion: It was observed that the optical properties of the skin change both when comparing the different physiological conditions and during the tissue repair process.

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# EFFICACY OF PDT AND LINGUAL SCRAPER IN THE TREATMENT OF HALITOSIS IN ELDERLY PATIENTS WITH COMPLETE DENTURE: A RANDOMIZED, CONTROLLED TRIAL

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Introduction: Halitosis is unpleasant odor emanating from the mouth or breath. 90% of halitosis cases are related to gases generated by metabolism of oral Gramnegative bacteria. The hydrogen sulfide gas emanate mainly from the bacteria housed in the dorsum of the tongue. There is a correlation between older age and halitosis, resulting in increased odor intensity. Conventional therapy involves a reduction in microorganisms with the use of antiseptic mouthwashes combined with tongue scrapers. Purpose: The aim of the present study was to compare the treatment of halitosis using PDT or a tongue scraper in older adults with full dentures. Material and Methods: A randomized, placebo-controlled clinical trial involving 40 individuals (60 years or older) with halitosis (hydrogen sulfide  $\geq$  112 ppb) with complete denture was performed. The patients were divided into 2 groups of 20: G1treated with tongue scraper; G2 -treated with PDT (Methylene blue+660 nm laser). The evaluation of halitosis was made with OralChromaTM, before and after treatments. If the halitosis is solved, the participants returned after one week for an additional evaluation. Results: Both groups showed a decrease in halitosis compared to the initial situation and this difference remained one week after treatments. However, PDT treatment was more effective than scraping both immediately (p = (0.0098) and after 1 week of treatment (p < 0.0001). Conclusion: PDT is an effective treatment for halitosis in elderly patients with full dentures and offers advantages such as low toxicity, not inducing bacterial resistance and preserving the oral microbiota.



# EVALUATION OF PAIN, FUNCTIONALITY AND QUALITY OF LIFE OF WOMEN WITH KNEE OSTEOARTHRITIS SUBMITTED TO AEROBIC EXERCISE AND PHOTOBIOMODULATION

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Introduction: Population aging is a worlwide phenomenon, mainly due to the decline in mortality and fertility rates. This growth of the elderly population generates chronic degenerative diseases. While population aging brought the benefits of greater longevity, it increased the morbidity and mortality profile, characterized by na increase in chronic degenerative diseases. Because of this situa-tion, nonpharmacological methods, such as physical exercise, are beneficial in several aspects. In addition, features such as photobiomodulation have shown favorable results. Objective: The aim of this study was to investigate the effects of aerobic exercise protocol and photobiomodulation on the pain, functionality, joint stiffness and quality of life in patients with knee OA. Material and methods: Ten individuals with II and III grade of knee OA were evaluated, randomized into 2 groups: Exercise and Photobiomodulation Placebo Group (EPPG, n = 6), treatment through an aerobic exercise protocol associated with photobiomodulation placebo or Exercise and Photobiomodulation Active Group (EPAG, n = 4), treatment through an aerobic exercise protocol associated with photobiomodulation active. The interventions was performed 2x / week for a period of 8 weeks. The exercise program included the warm-up, aerobic exercise, and cooling phase. The cluster (808nm, 100mW, 4J / point totaling 56J) was applied to the most affected lower limb after exercise. Visual Analogue Scale (VAS), Knee injury and Osteoarthritis Outcome Score (KOOS) and Short form 36 (SF-36) health survey guestionnaire was used for evalua-tion and reevaluation. In addition, Goniometry of knee flexion and the 30-second Chair Stand Test and Stair Climb Test was performed. Results: The descriptive results of this study showed a reduction in pain level, symptoms, joint stiffness, daily living activities and quality of life, as well as in the time to sit and get up and down stairs in both groups after treatment. For sports activities only the GEFP showed improvements, and the GEFA remained with the same average. Conclusion: These results are partial and the continuious of this work becomes important in order to obtain the statistical results with final sample initially calculated. From this, we will can concluded about the effect of FBM on enhancing na aerobic exercise program for women with knee OA.

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# EFFECTS OF LIGHT EMITTING DIODE (LED) ON MITOCHONDRIAL RESPIRATION, OXIDATIVE STRESS AND INFLAMMATORY PROFILE OF MESENCHYMAL STEM CELLS SUBJECTED TO DOXORUBICIN TOXICITY

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Introduction: A doxorubicin (DOXO) is a widely used chemotherapeutic agent that can cause major cellular damage, especially in cardiomyocytes. In some cases, progression to heart failure (HF) is not uncommon, and in this scenario different therapeutic options, such as the use of stem cells (CT), were tested. However, the therapeutic potential of CT after DOXO use may be mitigated by the hostile microenvironment following drug treatment, or may lead to drastically reduced cell survival after transplantation. Thus, this study proposes to test the use of Light Emitting Diode (LED) therapy as preconditioning in CT subjected to DOXO toxicity, a potential end of its resistive properties. Material and methods: Mesenchymal adipose tissue CT (CDTA) Fisher-344 isogenic rat fish coated (1x105 cells), cultured for 24h and irradiated (0.5J / cm<sup>2</sup> (0.5J group), 2J / cm<sup>2</sup> ( group 2J) and 4J / cm<sup>2</sup> (group 4J) 630nm) one minute before exposure to DOXO (25 µg / mL). One hour later, the following measurements were performed: mitochondrial metabolism, ATP, nitrite and nitrate dosage, and protein expression of VEGF, IL-1, IL-10, IL-6. Results: The group exposed to a non-irradiated DOXO (DOXO group) presented significant differences in relation to the control group in all analyzed parameters. Differences in the amount of ATP without mitochondrial metabolism and expression of VEGF and IL-10 were also found in all irradiated groups compared to the DOXO group. In addition, the 0.5J and 4J groups showed significant reduction in IL-6 expression and nitrate / nitrite ratio compared to the DOXO group. Conclusion: LED preconditioning was potentially effective as resistive properties of CT subjected to DOXO toxicity.



# PHOTODYNAMIC THERAPY REVERSES THE NEGATIVE EFFECTS OF PERIODONTITIS DURING THE GESTATION

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Introduction: Periodontitis (P) is one of the most common infectious diseases that affects the periodontium and gradually destroys periodontal tissues. Among several systemic effects occasioned by P, alterations during the gestation have been studied. PDT is characterized by the association of a light source with a photosensitizing agent in order to cause cell necrosis and microbial death. Thus, our objective was to evaluate the effect of PDT on the negative repercussions of P during pregnancy. Methods: Ten days before the pregnancy, P was induced by ligature technique, and the treatment with PDT was performed 15 days after the induction of P. Pregnant rats were euthanized at day 18 and the uterus and placenta were removed. The photosensitizer methylene blue (0.005%, CHIMIOLUX, DMC) was administered and the periodontal pockets were irradiated with a red laser (MM OPTICS; Wavelength 660 ± 10 nm; Radiant power100 mW; Time exposure 90s; Radiant energy 9 J; Irradiance 3.5 W/cm2; Radiant exposure 318 J/cm2; Total radiant energy18 J). Results: We showed that PDT reduced the gene expression of IL-6, COX-1, COX-2 and NOS in the uterine tissue of pregnant rats with P. We also observed that PDT reversed the decreased levels of IL-10 in the placenta. Conclusions: Thus, our data showed the important role of oral health during the gestation as well as PDT is an effective therapy.

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Key words: Periodontal disease; Gestation; Photodynamic therapy; Acute lung injury; Birth weight; Cytokines.



# EFFECT OF ER:YAG LASER AND ASSOCIATION PROTOCOLS ON THE DEMINERALIZED ENAMEL HARDNESS

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Introduction: Dental enamel is a highly mineralized tissue and defects that occur during amelogenesis may be manifested clinically in both the deciduous and permanent dentition such as Hipomineralization and Hypoplasia. Objective: The aim of this study was to analyse the hardness of demineralized enamel following different treatments. Materials and Methods Forty eight enamel blocs (4 x 4 x 7mm) were divided into six groups (n=8): (H) Sound; (DE) Demineralized; (DED) DE + Duraphat 5%; (DEL20) DE + Er:YAG laser (20mJ pulse mode; 0.20W; 10Hz; 60s; 1,18J/cm2; 11.83W/cm2); (DEL50) DE + Er:YAG laser (50mJ pulse mode; 0.50W; 10Hz; 60s; 2.95J/cm2; 29.58W/cm2); (DEL20D) DE + Er:YAG laser (20mJ) + Duraphat 5%. The irradiation was performed at 1mm distance from the surface using a tip (AS7066X, L-14mm, D-1.3mm in diameter) in water/air spray refrigeration (level 6). The enamel blocs were submited to DE-RE cycle (4h into DES solution + 20h into RE solution during 8 days and change the solutions following the 4th day). The Knoop hardeness was measured (50g/15s, six There were statistically diferences with the highest hardness observed for H and the lowest to DE (p<0,0001). Results: The DEL20D association was able to recover the enamel hardness similarly to H (p<0,05). Conclusion: Within the results obtained it may be concluded that the enamel microhardness was influenced by the studied treatments.



### EVALUATION OF PHOTOBIOMODULATION IN SALIVAR PRODUCTION OF PATIENTS WITH ANTI-HYPERTENSIVE DRUG-INDUCED XEROSTOMIA

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Introduction: Hypertension is a systemic condition that affects about 30% of the world population, according to data from the World Health Organization. Drugs used to control this disease have the potential to induce xerostomia, a buccal condition in which it is observed the decrease of the salivary flow and whose presence leads to the increase the index of caries, periodontal disease, until loss the teeth, dysgeusia, difficulty of mastication, dysphagia, bad breath, oral burning and impairment of the works installed in the buccal cavity, including retention of removable and total dentures. Photobiomodulation (PBM) has been shown to be effective in increasing saliva production in many conditions. Objective: The aim of this study was to evaluate a photobiomodulation in salivar production of patients with anti-hypertensive druginduced xerostomia. The patients (60) will be divided in 2 groups: G1: treatment with PBM (30); G2 simulate PBM (30). Material and methods: The method consists in the application of infrared laser in the three pairs of major salivary glands, one time per week, during 4 weeks. The parameters used will be: Laser Diode, DMC brand, emitting at  $\lambda$  = 808 nm, 4 J/site, CW, incidence (900) to the surface irradiated and in contact with it, distributed in 6 external points in each parotid, 2 external points in each sublingual and 2 points in each submandibular (internal), totaling 20 points. This protocol will determine the effectiveness of photodynamic therapy in xerostomia induced by antihypertensive drugs. Results: The study will verify if the PBM can increase the salivary flow of these patients, thus minimizing the side effects of antihypertensive drugs. Discussion: The literature is scarce when searching for PBM in patients with xerostomia induced by antihypertensive drugs. In the research carried out in the databases, no work was found that directed the PBM to patients with xerostomia induced by antihypertensive drugs. The great majority of the works deal with xerostomia related to chemotherapy and radiotherapy. Conclusion: This protocol will determine the effectiveness of photodynamic therapy regarding the reduction of xerostomia in older adults using antihypertensive drugs.

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