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FULL ARTICLE

Comparative study among three different phototherapy protocols to treat chemotherapy-induced oral mucositis in hamsters

Luana Campos, *Érika P. Cruz*, *Filipi S. Pereira*, *Victor E. Arana-Chavez*, *and Alyne Simões**

Laboratory of Oral Biology, Department of Biomaterials and Oral Biology, School of Dentistry, University of São Paulo, São Paulo 05508-000, Brazil

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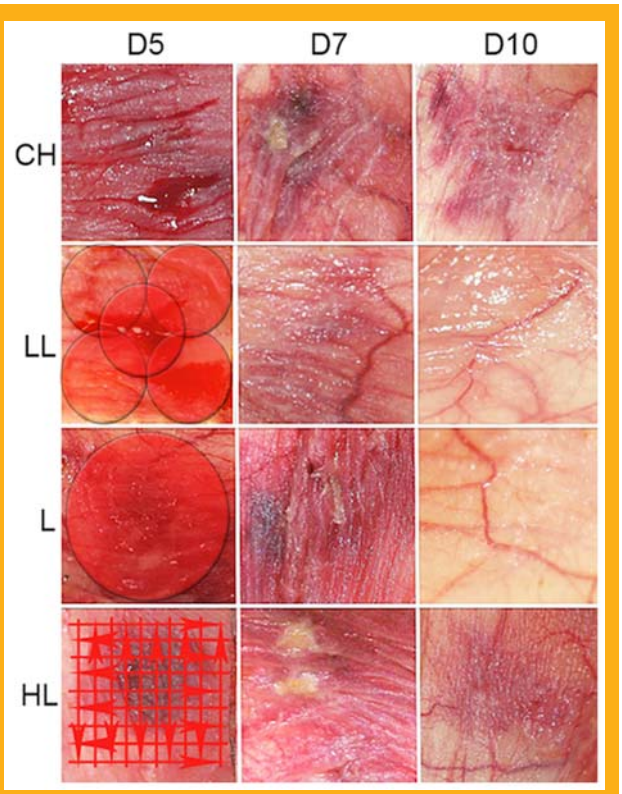
In this study, clinical, biochemical and histological analysis were used to compare different phototherapies, including LED, low and high-power laser (HPL) for the treatment of chemotherapy (CT)-induced oral mucositis (OM). One-hundred-fifty hamsters were divided into five groups: C: control; CH: CT/OM induction; L: CT/OM induction and treatment with LED (635 nm, 1.2 J), HL: CT/OM induction and treatment with HPL (808 nm, 10 J), LL: CT/OM induction and treatment with low-level laser therapy (LLLT) (660 nm, 1.2 J). OM was induced by scratches performed on check pouch mucosa after two injections of 5-Fluorouracil. The experiment lasted 10 days and OM was analyzed by specific clinical scales on days 5, 7 and 10. The animals were euthanized and the cheek pouch mucosa removed for biochemical (TNF-α concentration) and histological (light microscopy) analysis. After statistical analysis, the authors' results showed LED and LLLT therapies were efficient treatments for OM, decreasing TNF-α concentration on day 7 ($p < 0.05$) and completely healing the mucosa on day 10. HPL showed no interference in final healing of OM. According to the methodology used and the results obtained in the present study, LLLT and LED therapies were the best choices to decrease the severity of OM, accelerating tissue repair and decreasing the inflammatory process.



Clinical evaluation of OM in Groups CH, LL, L and HL and their respective arrangement of phototherapy treatments at different time intervals (5, 7 and 10 days).

* Corresponding author: e-mail: lysimoes@usp.br, Phone: +5511 2648.8091, Fax: +5511 2648.7840

1. Introduction

Oral mucositis (OM) is considered an acute side effect reported in patients undergoing mucotoxic chemotherapy (CT) [1]. When severe and/or widespread, it is associated with intense pain and bleeding, increasing the risk of systemic infection; need for fluids and nutritional support; and significant additional hospitalization costs [2, 3].

Although the pathogenesis of OM has not yet been completely elucidated, it has become accepted that proinflammatory cytokines are released in response to reactive oxygen species produced in the cells, resulting from the effect of CT [4, 5]. One of the cytokines most involved in the pathogeneses of OM is the tumor necrosis factor (TNF-α) related to damage to the epithelium, accelerating the formation of OM [5, 6].

Clinically, OM is characterized by pain and erythema, and when severe, it may be characterized by confluent ulcerative lesions that interfere with normal oral functions [7]. Although many palliative interventions have been used in OM management, recent studies have indicated the use of phototherapies as an effective and promising treatment [2, 3].

The MASCC/ISOO (Multinational Association of Supportive Care) clinical practice guideline recommends low-level laser therapy (LLLT) to prevent OM in patients receiving high doses of CT or chemoradiotherapy before hematopoietic stem cell transplantation [8, 9]. It is well accepted that the cells respond to a monochromatic radiation from laser and light emitting diode (LED), by their capacity to modulate metabolic process (mainly including ATP signaling pathway) [10, 11].

As regards OM healing, LLLT is known to increase fibroblast proliferation [12], favoring the collagen synthesis [13] and angiogenesis [14]; reducing COX-2, TNF-α and pro-inflammatory cytokines IL-6 and IL-1β [15, 16]; promoting differentiation of anti-inflammatory cytokines IL-2, IL-4, IL-8 and IL-10 [13]; and acting in the NFkB pathway [17]. In addition to analgesia, LLLT can enhance peripheral endogenous opioid production [18] and decrease serum prostaglandin E2 [19].

Furthermore, LED therapy has shown satisfactory, similar or even better results to those of LLLT with regard to biomodulatory effects [20–22]. Freitas et al., 2014, showed better healing of OM lesions in cancer patients submitted to LED therapy than in those receiving LLLT, although the authors concluded that both therapies were effective [20].

High Power Laser (HPL) irradiation on oral mucosa may be used in defocused mode, to avoid an increase in temperature and to act at low intensity, similar to LLLT [23–26]; therefore being used to promote wound healing as well as analgesia [26]. However, its healing effect is not as significant as its effects on pain relief [25]. In this sense, the mechanism of action of defocused HPL, described in the literature, is more related to the analgesic effect, showing neural inhibition, increased concentration of endogen opioids and inhibition of Na^+-K^+-ATPase [27–29], for example.

As regards the analgesic effect using LLLT, studies using low energy (0.05–1.5 J) for treatment and/or prevention of OM, have reported significant decrease in OM severity with consequent pain relief [24, 30–33]. In contrast, studies including high-energy application (2.0–3.5 J) have reported immediate analgesic effects, with decrease in the need for ingesting analgesics [24, 34].

Although there are several phototherapy protocols reported in the literature to treat or prevent OM in cancer patients, to the best of our knowledge, there is no study that compares these different protocols. Therefore, this study aims to evaluate and compare different protocols, by means of clinical, biochemical and histological analysis, with the use of 3 different types of equipment (LED, LLLT and HPL) for the treatment of CT-induced OM in hamsters.

2. Material and methods

The present study was conducted in compliance with the principles of laboratory animal care and national laws on animal use [35], and was authorized by the Ethical Committee for Animal Research of the University of São Paulo, Brazil. One-hundred-fifty male Golden Syrian hamsters, 8 weeks old, with body mass of approximately 150 g were used. All animals were maintained in the Laboratory of Oral Biology at the University of São Paulo, at temperature of 23 ± 3 °C, with 12-hour day/night cycles, fed with a standard laboratory diet and water *ad libitum*.

The animals were randomly and equally divided into 5 groups: control (C), CH, L, LL, and HL. C group received only the CT vehicle. All experimental groups received chemotherapy, as follow: CH group received only OM induction (described in *Oral Mucositis Induction Protocol* section below); L group received OM induction and LED therapy (1.2 J/cm^2, 1.2 J), LL group received OM induction and LLLT (6 J/cm^2, 1.2 J); and HL group received OM induction and HPL (10 J/cm^2, 10 J). One subgroup in each group was euthanized at days 5 (n = 50), 7 (n = 50) and 10 (n = 50).

2.1 Oral mucositis induction protocol

The protocol was based on a previously published protocol [36, 37]. The entire experiment lasted 10

consecutive days, and the OM was induced in groups CH, L, HL and LL, by two intraperitoneal injections of 5-Fluorouracil (5-FU) (Fluorouracil, Sigma Chemical CO, MO, USA), at days 1 and 3 of the experiment at doses of 100 mg/kg and 65 mg/kg body weight, respectively. The animals in Group C received only the CT vehicle (ammonia hydroxide, 1 M).

The second step, to induce the OM was performed on days 4 and 5, when both left and right cheek pouches of the animals were everted and the mucosa was irritated by superficial scratching with the tip of an 18-gauge needle. After this, a 1 cm^2 square area was demarcated in order to limit the irradiation field and the LED, HPL or LLLT treatments were conducted daily. Between days 4 and 10, all animals were anesthetized with Xylazine (Anazedan®, Vetbrands, Brazil) 13.8 mg/Kg and Ketamine (Dopalen®, Vetbrands, Brazil) 116 mg/Kg to receive the phototherapy protocol.

2.2 Laser parameters

For LLLT an InGaAlP diode laser (Twin Laser – MMOptics® Ltda, Brazil) with a wavelength of 660 nm, 40 mW, and energy density of 6 J/cm^2 was used, in punctual (5 points) and contact irradiation mode, delivering a total energy of 1.2 J. Irradiation time was 6 seconds per point based on the laser beam spot size of 0.04 cm^2.

The LED (Fisio LED – MMOptics® Ltda, Brazil) with a wavelength of 635 nm, 120 mW, irradiation time of 10 seconds per point, and energy density of 1.2 J/cm^2 was used, also in punctual and contact irradiation mode. Only one central point was treated, since the LED spot size is 1 cm^2, totaling 1.2 J of total energy delivered.

The HPL treatment was performed with a GaAlAs high power diode laser (Thera Lase Surgery, DMC Ltda, Brazil) with a wavelength of 808 nm, according to the protocol of Campos et al. [30] and Simões et al. [24]. Laser light was delivered through a 400 μm optical fiber; the power output at the display was set at 1.0 W, and the laser was applied in continuous-wave mode (power density of 1 W/cm^2). The irradiations were performed manually and perpendicular to the oral mucosa surface in defocused mode (non-contact) at an approximated distance of 1 cm from the mucosa lesion. The lesion area was irradiated for 10 seconds in scanning movements (5 seconds in vertical, and 5 seconds in horizontal movements) and energy density of approximately 10 J/cm^2 (Figure 1).

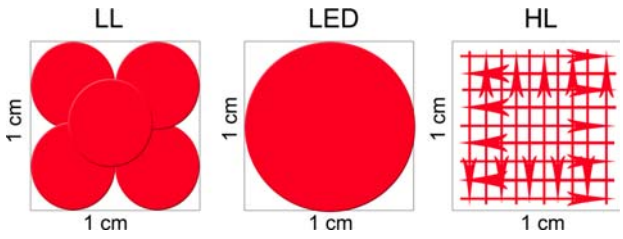


Figure 1 Arrangement of different irradiations (LLLT, LED and HL) in the demarcated areas (1 cm^2), corresponding to Groups LL, L and HL, respectively.

For all groups, the laser power was measured before irradiation using a Coherent® power meter, and the safety rules were followed, including the specific protective glasses, gloves and PVC (plastic film) to cover the laser pen tip.

2.3 Clinical evaluation

The clinical aspect of the cheek pouch mucosa was observed by one calibrated examiner on days 5, 7 and 10, and the degree of OM was evaluated by two specific assessment scales: criteria proposed by World Health Organization (WHO) [38] (Figure 2a), and Oral Mucositis Assessment Scale (OMAS) modified for hamsters according Wilder-Smith et al. (Figure 2b) [39]. The body mass, unconsumed food and drink of each animal were weighed daily.

2.4 Elisa immunoassay for TNF-α

For biochemical analysis, seventy-two samples were used. Immediately after euthanasia by cervical dislocation, the cheek pouch mucosa of all the animals was removed, weighed, clamped between aluminum tongs, precooled in dry ice and then stored at –80 °C until analysis. Before the analysis began, the tissue samples were thawed, homogenized at 10% and centrifuged at 1.540 × *g* for 10 minutes. The supernatant generated was used with the commercially available ELISA kit (RayBio®, Rat TNF-alpha, USA) to measure the TNF-*α*. The total protein concentrations of the tissue supernatant were determined with Folin-phenol reagent, using bovine serum albumin as a standard. The readings were taken at 660 nm [40]. Microplates were used and the data were standardized in terms of picograms of TNF-*α* per milliliter of total protein in the supernatant.

2.5 Morphological studies

For morphological analysis, 72 samples were used, i.e.; for each group 5 animals were analyzed on day 5, 7

a

Score:	Description:
0	Pouch completely healthy. No erythema or vasodilation.
1	Light to severe erythema and vasodilation. No erosion of mucosa.
2	Severe erythema and vasodilation. Erosion of superficial aspects of mucosa leaving denuded areas. Decreased stippling of mucosa.
3	Formation of off-white ulcers in one or more places. Ulcers may have a yellow/gray due to pseudomembrane. Cumulative size of ulcers should equal about ¼ of the pouch. Severe erythema and vasodilation.
4	Cumulative seize of ulcers should equal about ½ of the pouch. Loss of pliability. Severe erythema and vasodilation.
5	Virtually all of pouch is ulcerated. Loss of pliability (pouch can only partially be extracted from mouth).

b

Ulcerated Area*		Severity of Erythema†		
		None	Not severe	Severe
		0	1	2
None	0	0	1	2
< 4mm²	1	1	2	3
4 – 9mm²	2	2	3	4
>9mm²	3	3	4	5

Figure 2 Mucositis assessment scales proposed by (**a**) WHO and (**b**) OMAS, to assess the severity of OM.

and 10. Immediately after euthanasia, the cheek pouch mucosa tissues were removed and fixed in 4% formaldehyde and 0.1% glutaraldehyde (Polysciences, PA, USA) buffered in 0.1 M sodium cacodylate at pH 7.2. After 6 hours at room temperature and storage overnight at 4 °C, the samples were rinsed in the same buffer until processing could be completed. The samples were dehydrated in a graded ethanol series to acetone, and embedded in historesin JB4 (Electron Microscopy Sciences, PA, USA). Three 3-µm-thick sections were obtained with a glass knife in a MICROM HM360 microtome (Germany) and stained with hematoxylin and acid fuchsin, examined and photographed in an Olympus BX 60 light microscope.

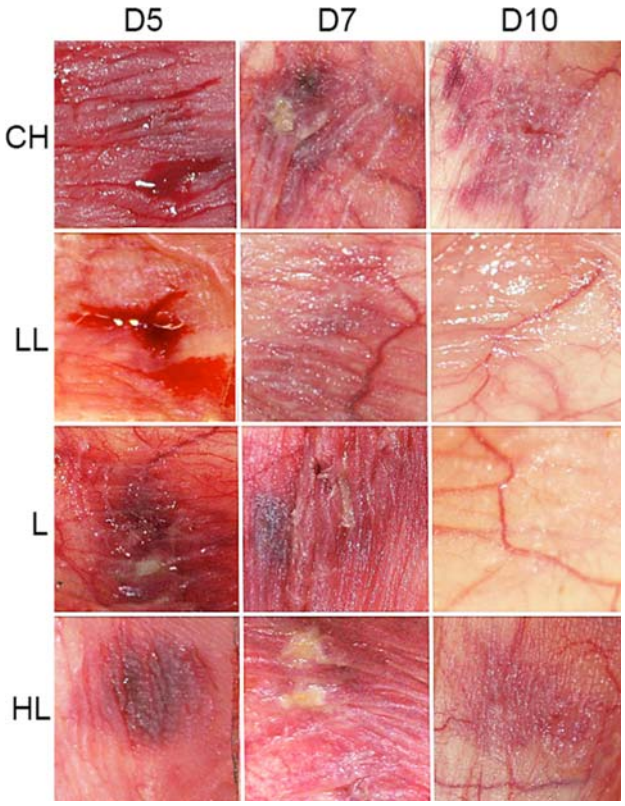


Figure 3 Clinical appearance of OM lesions in Groups CH, LL, L and HL at different time intervals (5, 7 and 10 days).

2.6 Statistical analysis

The results obtained were subjected to statistical testing using the analysis of variance (ANOVA) to compare quantitative variables and the Kruskal-Wallis, Fisher and Turkey contrast tests to compare qualitative variables. The significance level was set at 5%.

3. Results

3.1 General clinical evaluation

All experimental groups presented signs similar to those of oncologic patients undergoing chemother-

Table 1 Mean values of variance in loss of body mass (grams), food (grams) and water (mL) ingestion for all groups in 10 days of experiment ($n = 10$).

Group	Body Mass	Food Intake	Water Consumption
C*	+5 (± 3.1) a	81 (± 17.5) a	119 (± 5.5) a
CH	–26 (± 16.2) bc	54 (± 23.6) b	89 (± 17.6) b
L	–32 (± 11.4) cd	27 (± 13.4) c	84 (± 15.0) b
HL	–40 (± 15.2) d	21 (± 7.1) c	82 (± 29.6) b
LL	–16 (± 13.3) b	42 (± 18.3) b	120 (± 41.3) a

* Same letters mean that there was no statistically significant difference between groups ($p < 0.05$).

apy treatment, such as diarrhea, decrease in water and food ingestion and prostration, with consequent loss of body mass.

The Group C consumed the most water and food, followed by the LL group, representing a positive relationship with the data on weight loss, since Group C showed weight gain of 4%, and Group LL animals were closest to this amount. Group CH showed a reduction of 18% in initial weight, and Groups L and HL showed a more pronounced weight loss of approximately 25% (Table 1).

Food intake was lower for Groups L and HL right from the beginning of the experiment. Group CH maintained a rate of consumption similar to that of Group C only on days 4 and 5, the rate being 26% and 33% lower than that of Group C on days 7 and 10, respectively. On day 10, Group C maintained a higher rate of consumption than the other groups; Groups CH and LL were similar and consumed approximately 40% less than Group C; and 50% more than Groups L and HL (Table 1).

Differently from the food intake, the water consumption was similar between Groups C and LL on all experimental days. When comparing all groups on day 5, there was no statistical difference. On day 10, the water consumption of Groups CH, L and HL was approximately 15% lower when compared with Groups C and LL (Table 1).

Table 2 Medium degree of OM, according to WHO and OMAS scales for each group (CH, LL, L and HL) on different days of analysis of the experiment (4, 5, 7 and 10 days).

Groups	Clinical Scales for OM			
	OMS			
	Days			
	4	5	7	10
CH*	3.7 Aa	4.2 Cb	3.2 Ea	2.4 Ga
LL	4.0 Ae	4.0 Ce	1.6 Dd	0.0 Fc
L	3.9 Ah	4.0 Ch	3.0 Eg	0.3 Ff
HL	4.0 Ak	3.3 Bj	3.7 Ek	1.6 Gi
W-Smith				
CH	3.2 Aa	4.1 Db	3.3 Fab	2.3 Ha
LL	2.8 Af	2.8 Bef	1.4 Ed	0.1 Gc
L	3.0 Aij	3.2 Bcj	2.6 Fhi	0.3 Gg
HL	3.0 Alm	3.5 Cn	3.2 Fmn	1.5 Hk

* Same letters mean that there was no statistically significant difference between groups. Capital letters indicate values between the groups; Lower case letters indicate values between the days.

3.2 Clinical evaluation of oral mucositis

Based on the scales, similar results were observed in the clinical assessment of OM lesions (Table 2). At the beginning of the experiment, Groups CH, L, LL and HL had a similar degree of OM, all presenting grades between 3–4. Group LL showed the lowest degree of OM on day 7 when compared with the other groups (1.5 and approximately 3, respectively). Group L was similar to Group LL on day 10, both with grade 0 ($p < 0.05$) (Figure 3) and Groups CH and HL maintained the highest degrees of OM on day 10, being 2.4 and 1.6 respectively, according to the WHO scale. When comparing the difference of OM grades on the experimental days for each group, the authors' observed that Group CH showed no significant difference between the initial and final degrees of OM (from 3.7 to 2.4), differing from Group HL that showed a decrease on day 10 (from 4.0 to 1.6) ($p < 0.05$) (Table 2).

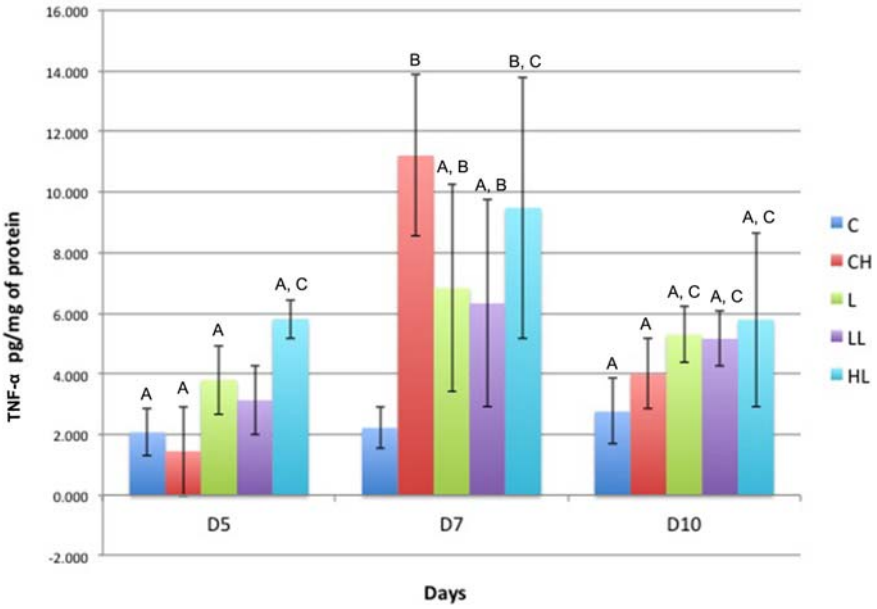
3.3 Elisa immunoassay

The biochemical analysis, performed by immunoassays for TNF-α, showed a similar concentration of the cytokine on days 5 and 10 for all groups; however, on the 7th day, Groups CH and LA showed an increase of approximately 500% and 400%, respectively, in TNF-α concentration when compared with Group C ($p < 0.05$); while Groups LL and L maintained similar values to those of Group C group ($p > 0.05$) (Graph 1).

3.4 Histological evaluation of oral mucositis

At histological levels, a number of morphologic changes were observed for all experimental groups.

On day 5, the animals from Group CH showed mucosal fragments of keratinized stratified squa-



Graph 1 Mean value of TNF-a concentration (pictograms per milligrams) per day (D5, D7 and D10) of experiment for each experimental group. Similar letters indicate similarity ($p < 0.05$).

mous epithelium, basal stratum with the largest number of cell layers and a central area with exposure of the connective tissue. In the lamina propria there was an increase in dense connective tissue with moderate inflammatory infiltrate (Figure 4a). On day 7, they showed an area of epithelial thickening, predominance of dense connective tissue with intense inflammatory infiltrate, associated with an area of necrosis and blood extravasation (Figure 4b). Whereas, on day 10, they presented hyperkeratinized stratified epithelium with a corrugated surface and basal layer with areas of hyperplasia. The lamina propria showed moderate inflammatory infiltrate, disorganized collagen fibers and blood extravasation (Figure 4c).

With regard to the animals from Group LL, on day 5 they showed the same characteristics as those of Group CH on the same experimental day (as described above) (Figure 4d). On day 7, mucosa samples revealed re-epithelialization, which was not observed in the other groups; the subjacent connective tissue exhibited inflammatory infiltrate, ranging from discrete to moderate (Figure 4e). On day 10, the mucosa fragments showed complete integrity of the epithelium, dense connective tissue with organized collagen fibers and neoangiogenesis (Figure 4f).

Whereas, the animals from Group L showed no difference in histological characteristics of the oral mucosa, when compared with Groups CH and LL on day 5 (Figure 4g). On day 7, however, they

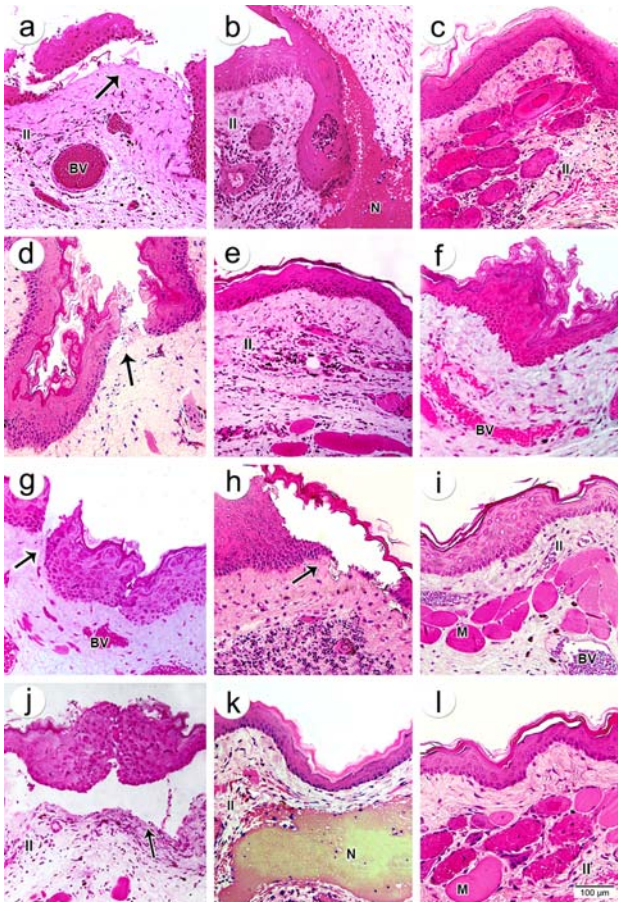


Figure 4 Photomicrographs of the oral mucosa in the different experimental groups that received induced OM. Analysis by Light Microscopy for Groups CH (**a**, **b**, **c**), LL (**d**, **e**, **f**), L (**g**, **h**, **i**) and HL (**j**, **k**, **l**), at different time intervals of the experiment (5, 7 and 10 days). The arrows indicate ulcers. *BV*, blood vessels, *N*, necrosis, *II*, inflammatory infiltrate, *M*, muscle fibers. 20,000× magnification.

showed a central ulceration covered by pseudomembrane, consisting of fibrin and inflammatory infiltrate, ranging from moderate to intense (Figure 4h). On day 10, they presented complete integrity of the epithelium with plane interface and underlying tissue. In the same way as Group LL, this group showed organized collagen fibers and neoangiogenesis (Figure 4i).

Finally, on day 5 the animals from Group HL showed the same characteristics as those of Group L, on the same experimental day (Figure 4j). On day 7, they revealed areas of epithelial thickening, predominance of dense connective tissue with intense inflammatory infiltrate, blood extravasation and an important area of necrosis (Figure 4k). Lastly, on day 10, they presented a palisade basal cell layer, chronic inflammatory infiltrate associated with disorganized collagen fibers (Figure 4l).

4. Discussion

Oral Mucositis is very painful mucosal damage as a consequence of cancer therapies, including high-doses of CT. Severe OM can negatively influence the patient's prognosis and have important economic impact, resulting from costs associated with management of the symptoms [41–43]. Studies have indicated that LLLT and LED can effectively reduce the severity of OM [2, 20, 22, 30, 36, 44–47], and HPL has been associated with a greater analgesic effect [24]. However, little is known about the comparison among LED, LLLT and HPL protocols to treat OM, at clinical, biochemical and histological levels. The present study showed that LLLT and LED promoted faster wound healing and reduced TNF-α expression, when compared with HPL.

The experimental OM model adopted in this study was performed according to the methodology proposed by Sonis et al. (1990) [37] modified by França et al. (2009) [36]. The clinical signs resulting from CT observed in all animals treated, such as diarrhea, gastrointestinal bleeding, decreased food intake and water consumption with consequent weight loss confirmed standardization of the methodology.

Although the animals in Group CH showed the worst condition of OM, the food and water consumption was similar to that of the other experimental groups. In fact, the 5-FU affects not only the oral cavity tissues, but the entire gastrointestinal tract, including major changes in the intestinal mucosa, resulting in a marked decrease in the absorption of water and nutrients, contributing to weight loss of the animals [48].

In the clinical analyses of OM severity, L and LL Groups showed complete repair by the end of the experiment, showing the effectiveness of these phototherapies. However, Group LL presented the best results; the animals showed less severe degrees of lesions, accelerated tissue repair and less inflammatory infiltrate as from day 7 of the experiment.

The effectiveness of laser and LED therapies to prevent and treat OM has been reported in the Refs. [2, 14, 20, 30, 44–46]. In a randomized prospective study to determine the effect of LLLT (660 nm, 15 mW and 3.8 J/cm^2) for prevention and treatment of OM in patients undergoing head and neck radiotherapy, the authors concluded that laser was effective in controlling the intensity of OM, and promoting an analgesic effect [45]. Freitas et al., 2014, on the other hand, showed better healing of OM lesions in cancer patients submitted to LED therapy than those receiving LLLT, although the authors concluded that both therapies were effectives [20].

In addition, a recent study with hamsters, using LED or LLLT, showed both treatments were effective in diminishing OM lesions [44]. The authors irradiated the 5-FU-induced OM with the same equipment and wavelength as those used in the present study. However, they used a different 5-FU dose and a higher energy per point, and consequently, a higher level of total energy [44].

Group HL maintained highest degrees of injury during all experimental periods, with edema and inflammatory infiltrate. However, it had milder degree of OM when compared with Group CH, indicating that the HPL may not be the best option, nevertheless it could be an alternative in the OM treatment. Similarly, Simoes et al., 2009 and Zand et al., 2012, compared the effect of HPL with LLLT or placebo, respectively, and suggested that HPL was unable to reduce the severity of oral lesions, but was more effective for promoting analgesia [24, 25].

The analgesic effect promoted by unfocused HPL has also been evaluated in rat ganglion cell culture. Chow, et al., using a laser with 830 nm, 1 W, defocused 4.5 cm from the surface of the coverslip and energy ranging from 1.5 J to 33 J suggested that laser irradiation can result in blocking the fast axonal flow, modulating nociception and reducing the pain [28]. In addition, some reports suggest an increase of endogen opioid concentration [49], an increase in synaptic activity of acetylcholine esterase [50], and at high energy densities, the inhibition of Na^+-K^+-ATPase [51].

Thus, based on evidences that the laser irradiation can inhibit nociceptive stimuli in humans and rats, in particular, specific inhibition of Aδ and C fibers, it is a plausible mechanism for the relief of acute and chronic pain with the use of lasers [52, 53]. These experimental findings in addition to the evidences that HPL has greater analgesic effect than LLLT [24, 25], support its use when the analgesia is the main propose of the treatment. In agreement

with this hypothesis, some studies [11, 24, 25, 53] have demonstrated that HPL was less effective for healing OM than LLLT, this being the reason for indicating HPL to be applied only in specific painful sites of OM.

Regarding the proinflammatory cytokine responsible for inducing inflammation and epithelial damage, Groups LL and L presented lower expression of TNF-α, on the 7th day of the experiment, when compared with the Groups CH and HL. These data contributed to the hypothesis that the treatment with LLLT and LED alter normal immunological response pattern by inhibiting the production of cytokines responsible for initiating and maintaining the inflammatory response and consequently, reducing the severity of the OM lesions [54, 55]. Furthermore, the LLLT can modulate the activity of macrophages, a connective tissue cell responsible for releasing TNF-α [56].

In a comparative study between LLLT and LED, both at fluence of 5 J/cm^2, the authors showed that the inflammatory phase for the phototherapy groups were better than for control group, with a reduced number of inflammatory cells, increased number of fibroblasts and increased collagen deposition [57]. More recently, the anti-inflammatory effect of LED was assessed in an experimental model of collagenase-induced tendinitis in rats; by histological analysis a significant decrease was shown in the inflammatory cytokines after LED treatment [58].

Considering that the animals from Groups HL showed a higher TNF-α concentration, necrotic area in the histological analysis and clinical presence of edema on day 7, the authors of the present study hypothesized that this could be associated with a thermal effect, since the cheek pouch mucosa of the hamsters is thinner than human oral mucosa. As HPL is known to act by increasing temperature, and even being used in non-contact mode in the present study, it may have promoted sufficient heating, thus justifying the intense inflammatory infiltrate observed [59–62].

In summary, LLLT and LED are atraumatic, safe and non-invasive techniques [13]; moreover LED therapy can be used as an alternative to LLLT, with some advantages being lower cost and ability to cover a wider area with a reduced treatment time. Whereas, although defocused HPL is atraumatic, non-invasive and promotes faster analgesia, it is more expensive and requires more extensive technical training of the operator to achieve successful treatment.

Finally, these findings provide some basic knowledge, essential for determining good phototherapy protocols for the treatment of OM, in order to provide the best care for patients submitted to cancer treatment and improve their quality of life.

5. Conclusion

According to the protocols used in the present study, the LLLT and LED therapies stimulated the wound healing process of oral mucositis lesions induced in hamsters by 5-FU injections and scratches. In addition, although the defocused high power laser could be an option for use in the treatment of painful oral lesions, it should be applied with caution. However, new recommendations and perspectives for clinical trials have to be considered.

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Conflict of interest No potential conflicts of interest were disclosed.

Author biographies Please see Supporting Information online.

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