

# Effect of photobiomodulation therapy (808 nm) in the control of neuropathic pain in mice

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**Abstract** Neuropathic pain can be defined as the pain initiated or caused by a primary lesion or dysfunction of the central or peripheral nervous system. Photobiomodulation therapy (PBM) stands out among the physical therapy resources used for analgesia. However, application parameters, especially the energy density, remain controversial in the literature. Therefore, this study aimed to investigate the PBM effect, in different energy densities to control neuropathic pain in mice. Fifty (50) mice were induced to neuropathy by chronic constriction surgery of the sciatic nerve (CCI), treated with PBM (808 nm), and divided into five groups: GP (PBM simulation), GS (sham), GL10, GL20, GL40 (energy density of 10, 20, and 40 J/cm<sup>2</sup>, respectively). The evaluations were carried out using the hot plate test and Randall and Selitto test, before and after the CCI surgery, every 15 days during the 90 days experiment.  $\beta$ -Endorphin blood dosage was also tested. For both the hot plate and Randall and Selitto tests, the GL20 and GL40 groups presented reduction of the nociceptive threshold from the 30th day of treatment, the GL10 group only after day 75, and the GP group did not show any improvement throughout the experiment. The  $\beta$ -endorphin dosage was higher for all groups when compared to the GP group. However, only the GL20 group and GL40 presented a significant increase. This

study demonstrates that PBM in higher energy density (20, 40 J/cm<sup>2</sup>) is more effective in the control of neuropathic pain.

**Keywords** Laser therapy · Nerve injury · Pain control

## Introduction

Pain is described as a complex, subjective experience, which involves not only the transduction of harmful environmental stimuli but also the cognitive and emotional processing by the brain [1, 2]. Pain can happen in two ways during the inflammatory process, spontaneously and/or by the phenomena of sensitization, characterized by the exacerbated response to the harmful stimuli (hyperalgesia), as well as by pain in response to non-harmful stimuli (allodynia) [3–5].

Central or peripheral nervous system lesions may lead to a special kind of pain without nociception, named neuropathic pain [6]. As in other types of chronic pain, neuropathic pain promotes the activation of brain regions such as the parabrachial, amygdala central nucleus, gray matter and periaqueductal, and the cerebral cortex linked to emotion, sensory integration, and personality, which favors the emergence of comorbidities, such as anxiety and depression, generating a great social commitment of the individual [7].

Several experimental models have been developed for a better understanding of the neuropathic syndrome. Among these, we can highlight the partial or complete transection of the nerve and perineural inflammation, experimental diabetes, and chronic constriction injury of the sciatic nerve [8–11]. Chronic constriction injury of the sciatic nerve (CCI) has been extensively investigated due to its high reliability, wide reproducibility, and the development of thermal and mechanical hyperalgesias and allodynia, similar to the symptoms that occur in human patients with neuropathies [8, 12–14]. The signs

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of spontaneous pain along with thermal and mechanical hyperalgesia appear 24 h after surgery (CCI), remaining for about 4 months [8].

Currently, the pharmaceutical industry use painkillers as the main drugs to treat pain, although these have shown only 30% effectiveness in patients with neuropathic pain [15–17]. Thereby, many researchers have been seeking alternative treatments for this type of pain, such as physical therapy, acupuncture, psychotherapy, and anesthetics and neurosurgical procedures [2, 13, 18].

Among the resources used by physical therapists, photobiomodulation therapy (PBM) has been showing good results in neuropathic and chronic pain control [18–20]. The term PBM has recently been defined, at the 2014 joint North American Association for Laser Therapy (NAALT) & World Association for Laser Therapy (WALT) conference, as “The therapeutic use of light [e.g. visible, near infrared (NIR), infrared (IR)] absorbed by endogenous chromophores, triggering non-thermal, non-cytotoxic, biological reactions through photochemical or photophysical events, leading to physiological changes.” The analgesic effects induced by PBM can be explained by the modulation of the inflammatory chemical mediators, in addition to the stimulating synthesis of  $\beta$ -endorphin [21, 22]. The association of these factors tends to limit the excitability threshold reduction of pain receptors and eliminate allogeneic substances [23, 24]. However, several studies showing the use of PBM as a treatment for neuropathic pain report the use of different parameters, and these often differ with respect to energy density [19, 25, 26].

The facts above mentioned suggest that PBM is considered an effective practice in the treatment of painful processes, although the literature presents a great discrepancy in the use of the parameters, so it is believed that new research using experimental models to the establishment of appropriate protocols for the application this therapy in clinical practice are relevant.

## Materials and methods

The present study was conducted according to the “Guide for Care and Use of Laboratory Animals” and approved by the Ethics Committee on Animal Experiments of the Federal University of São Carlos (UFSCar), protocol 026/2014. The animals were kept in the vivarium of the Physical Therapy Department at the UFSCar throughout the trial period and were housed individually in suitable standard polyethylene cages, under controlled environmental conditions (19–23 °C and light/dark cycle of 12/12 h). Food and drinking water were freely available, except during the brief test periods. A total of 50 mice, male Swiss-albino strain, weighing 25–30 g,

were used in this study. The animals were randomly divided into five groups ( $n = 10$ ) as follows:

- Sham group (SG): Simulation of the surgical procedure (CCI)
- Placebo group (PG): Subjected to surgical procedure, without undergoing irradiation
- Laser therapy group 10 J/cm<sup>2</sup> (LG10): Induction of neuropathy through CCI surgery with PBM application (808 nm), with energy density 10 J/cm<sup>2</sup>.
- Laser therapy group 20 J/cm<sup>2</sup> (LG20): Induction of neuropathy through CCI surgery with PBM application (808 nm), with energy density of 20 J/cm<sup>2</sup>.
- Laser therapy group 40 J/cm<sup>2</sup> (LG40): Induction of neuropathy through CCI surgery with PBM application (808 nm), with energy density of 40 J/cm<sup>2</sup>.

## Induction of neuropathy

The CCI experimental model was used for neuropathy induction. The method was performed initially by anesthesia under ketamine (1  $\mu$ l/kg, Agener, SP, Brazil) and xylazine (0.5  $\mu$ l/kg, Dopaser, SP, Brazil) (90 mg/kg, intraperitoneally, i.p.). The animal was placed in a ventral decubitus position with the right femur elevated to a 90° angle, fixed with adhesive tape. Next, an incision was made between the fascia of the gluteal and the femoral biceps thus exposing the sciatic nerve right next to its trifurcation. The tissue around the nerve was carefully cut at a distance of approximately 8 mm and nerve compression was achieved by placing four bandages using sterile non-inflammatory mononylon threads 5.0 [12–14].

## Photobiomodulation therapy

The animals received the first PBM application 3 days after surgery. The PBM was held with an infrared laser (Photon Laser III, DMC, São Carlos, Brazil), 808 nm wavelength, 30 mW output power, area of 0.028 cm<sup>2</sup>, and power density of 1 W/cm<sup>2</sup>. The parameters used for each group were as follows: LG10 with energy density of 10 J/cm<sup>2</sup>, total energy of 0.27 J, and irradiation time of 9 s; LG20 with energy density of 20 J/cm<sup>2</sup>, total energy of 0.54 J, and irradiation time of 18 s; LG40 with energy density of 40 J/cm<sup>2</sup>, 1.20 J, and application time of 37 s. PBM treatment was performed three times a week for a period of 90 days.

The in-contact punctual techniques were used in application, with the equipment pen positioned perpendicular to the tissue. The irradiation was carried, only in one point, out in the surgery site. At the time of treatment, the animals were immobilized by a cotton blanket for better application of the therapy.

## Functional evaluations

The evaluations were initiated in the pre-surgery period, so that data would be used as a baseline for the study. A new evaluation was conducted after 48 h of the CCI to verify the surgery effectiveness, and evaluations were conducted every 2 weeks during the 90 days of treatment. We performed eight evaluations throughout the experiment for each group, being that in each evaluation the animal was submitted to the tests only once. It is noteworthy that the animals were submitted to the acclimation tests 24 h before the start of the collections.

## Thermal hyperalgesia

The hot plate test is commonly used for the evaluation of thermal hyperalgesia in mice. The animals were placed on the hot plate (Hot Plate, Insight, Brazil) at approximately 52 °C and responses to thermal stimulation (withdrawal and licking of the superior or inferior members) were timed [27]. The longest they stayed on the plate was 25 s. This test was applied only once in each animal at each evaluation period.

## Mechanical hyperalgesia

The analgesymeter (Analgesymeter Randall Selitto, Insight, Brazil) equipment was used to carry out this evaluation; it is responsible for generating a linear increase of strength (in grams) on the animal paw dorsal surface, until it produces a response characterized by paw withdrawal. The paw withdrawal reflex is considered representative of the hypernociceptive threshold, i.e., the necessary force to be applied to the paw to induce an aversive response to a noxious stimulus. The force necessary for this animal to show such an answer is recorded in grams [28]. The animals submitted to this test were standardized so that the right lower leg (always submitted to CCI surgery) was always tested. Each animal was submitted once to the test, in each evaluation period, thus totaling eight evaluations in each animal throughout the experiment.

## Analysis of $\beta$ -endorphin

For the determination of the  $\beta$ -endorphin levels, blood samples were collected through the decapitation of animals at the end of the experiment. The samples were centrifuged to separate serum and plasma, which was used for analysis. The  $\beta$ -END ELISA kit (MyBioSource, California, USA) was utilized, with polystyrene plates with 96 wells of conical base (Mybiosource, California, USA), covered with acetate tapes and automatic multichannel pipettes in several volumes, from 0.5 to 1000  $\mu$ L. The solutions and volume transfers were performed in polystyrene microtubes. We also used a plate washer (Wellwach BC, Thermo Scientific®) and a plate

reader (Multiskan FC, Thermo Scientific®). The absorbance for the referred  $\beta$ -endorphin was measured through the MicroWell 450 nm plate reader, as recommended in the kit.

## Statistical analysis

Results were expressed as mean and standard deviation. Intra-group and between groups variation was measured with one-way and two-way analysis of variance (ANOVA), respectively, using the software GraphPad Prisma 5.0. Tukey test was applied at the end of the analysis for multiple comparisons. The level of significance was set at  $p < 0.05$ .

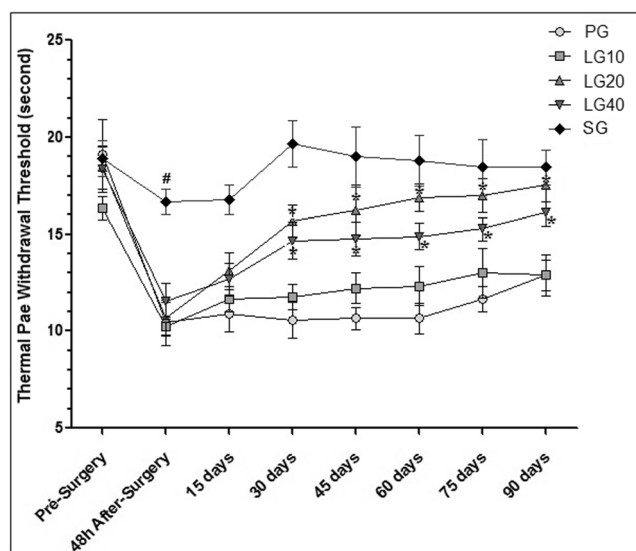
## Results

The animals were weighed and evaluated by the hot plate and Randall and Selitto tests before surgery. The animals were submitted to CCI surgery the following day, and after 48 h of surgery were again weighed, and evaluated showing the emergence of symptoms of hyperalgesia, allodynia, and reduction of body weight, which characterizes the existence of neuropathic pain. In the evaluations of thermal hyperalgesia, performed by the hot plate test, it was observed that the PG had not shown significant improvement of pain, being that the average obtained in the pre-surgery period was  $19 \pm 6$  s and the greatest average obtained throughout the experimental period was  $12.8 \pm 2, 2$  s, and there was absence of sensitivity recovery. The LG10, LG20, LG40 groups presented sensitivity recovery at the end of the treatment, but in the LG10 group this recovery was observed only from day 75 of treatment ( $p < 0.05$ ), whereas the LG20 and LG40 groups showed pain improvement and full sensitivity recovery from day 30 ( $p < 0.0001$ ) (Fig. 1).

The mechanical hyperalgesia evaluations, conducted by Randall and Selitto pressure test, showed significant improvement of pain and reduced symptoms of allodynia and hyperalgesia in LG10, LG20, and LG40 groups. However, for the LG20 group, treatment group demonstrated effectiveness from day 30, in the LG40 group from day 45 and the LG10 group this effectiveness was observed only in the last evaluation. The PG group did not show any improvement of symptoms, demonstrating the efficacy and duration of surgery during all the time of the experiment (Fig. 2).

The SG group showed no significant changes of data at any time of the experiment to the test for thermal hyperalgesia. However, for the mechanical hyperalgesia test, significant difference was observed on the 15th day, but this difference did not remain during the experiment, it can be justified as a post-surgical acute reaction.

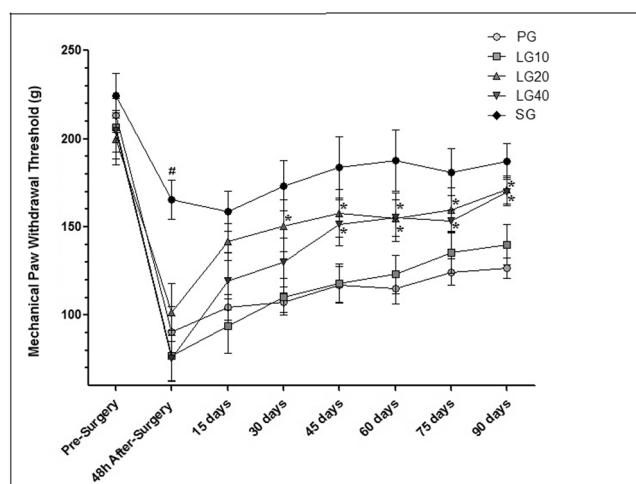
The intergroup results for functional evaluations showed significant improvement of symptoms and recovery of sensitivity with the use of PBM with energy density of 20 and 40 J/



**Fig. 1** Evaluation of thermal hyperalgesia. Paw withdrawal threshold, measured in seconds (s) of PG, LG10, LG20, and LG40 groups during the Hot Plate test in pre-surgery periods (*Pre-Surgery*), 48 h after CCI surgery (*48 h After-Surgery*), and during the 90 days of treatment on which the evaluations occur every 15 days. \* Statistically significant difference when compared with the PG ( $p < 0.05$ ). # Statistically significant difference when compared the SG with the others groups ( $p < 0.05$ )

cm<sup>2</sup>, for the LG20 and LG40 group, respectively. However, there were no significant differences between these two treatment energy density.

When PG group was compared with the LG10 group in the thermal hyperalgesia evaluation, there were no significant differences in comparison at any evaluated period. However, when compared with the LG20 and LG40 groups, significant



**Fig. 2** Evaluation of mechanical hyperalgesia. Paw withdrawal threshold, measured in grams (g), of PG, LG10, LG20, and LG40 groups, conducted by Randall and Selitto pressure test in the pre-surgery periods (*Pre-Surgery*), 48 h after surgery CCI (*48 h After-Surgery*), and during the 90 days of treatment in which the evaluations occur every 15 days. \* Statistically significant difference when compared with the PG ( $p < 0.05$ ). # Statistically significant difference when compared the SG with the others groups ( $p < 0.05$ )

differences were observed from day 30 of treatment to thermal hyperalgesia (Fig. 1) and day 45 of mechanical hyperalgesia (Fig. 2), thus demonstrating the effectiveness of the treatments of the groups LG20 and LG40.

It is worth mentioning that in the pre-surgical period, none of the five groups showed statistically significant differences in any evaluations between groups (Fig. 3a, b). In the 48-h period after surgery, it can be observed that in the comparison between groups, SG presented a statistical difference in relation to the other groups, without significant drop of SG values, both for the thermal hyperalgesia test (Fig. 3a) and the mechanical hyperalgesia (Fig. 3b), showing the CCI surgery efficacy in the other groups.

Figure 3 also highlights the mean values in the period 48 h after surgery and 90 days after surgery, characterizing the end of the experiment, it being possible to observe that the LG10, LG20, and LG40 showing sensitivity recovery due to treatment with LLLT, with final values similar to those found in the preoperative period. While SG did not present significant difference of the values during the whole experiment.

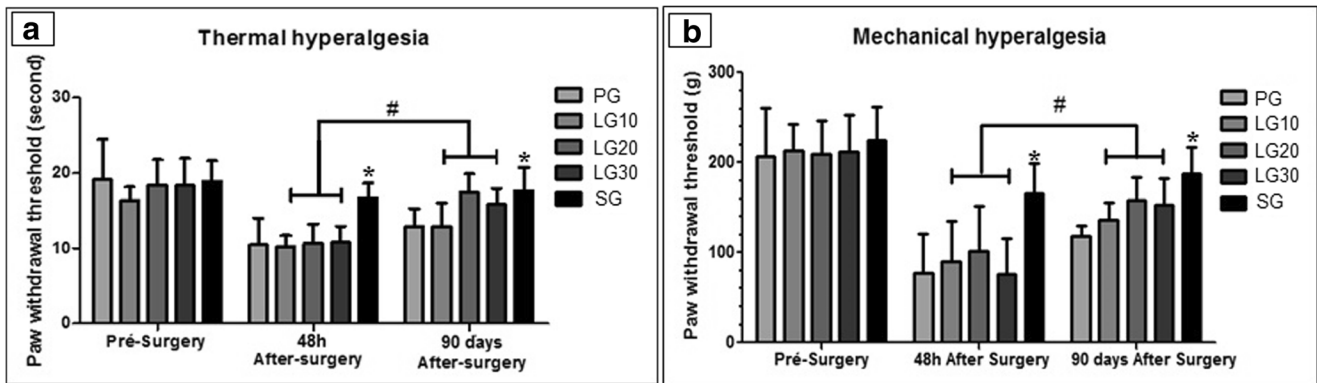
According to the ELISA assay protocol, it was possible to observe that when the PG group was compared with the other groups, all showed a greater amount of  $\beta$ -endorphin; however, only the LG20 and LG40 groups showed a statistically significant increase (Fig. 4).

## Discussion

It is known that painful processes, especially chronic pain, drastically reduce the individual quality of life and the persistence of symptoms can cause negative emotional reactions becoming debilitating and cause of suffering [29]. Treatment for these processes is becoming more challenging, and the search for new complementary therapies has been recurrent in literature [30].

The CCI method was used as an experimental model of neuropathic pain, being this method characterized by chronic constriction of the sciatic nerve, without nerve rupture. This work aimed to control of the painful process, like the other experimental models such as ligation of the spinal nerve (SNL), where one or more spinal nerves is constricted and cut, and the model of nerve injury (SNI) in which are cut the peroneal branch and tibial sciatic nerve [10–12]. The CCI stands out for being a model of easy execution and mimics the signs of neuropathic pain in the clinical treatment, such as mechanical allodynia, mechanical hyperalgesia, and thermal hyperalgesia [8, 12, 13].

Recently, several studies have investigated the mechanisms underlying the effects of different therapies such as joint manipulation [31], acupuncture [32], massage therapy [33], and phototherapy [34]. The data presented here reinforce that PBM has a significant analgesic effect. In addition, we



**Fig. 3** Evaluation of thermal (a) and mechanical hyperalgesia (b) in pre-surgery and 48 h after-surgical and 90 days after-surgery moments for all groups. # Statistically significant difference of the groups in relation to the

period 48 h after-surgical and 90 days after-surgery ( $p < 0.001$ ). \* Statistically significant difference the SG with other groups

expanded the possible targets by which the PBM would act to control pain, especially neuropathic.

The main finding of this study was that PBM with an 808-nm laser acts effectively in reducing neuropathic pain, noting that higher energy density are more effective in the promotion of analgesia. The fact that PBM promotes analgesia can be attributed to an increased production of  $\beta$ -endorphin caused by this therapy, as has been shown in other studies [21, 22, 35], especially in higher energy density, as presented in our study.

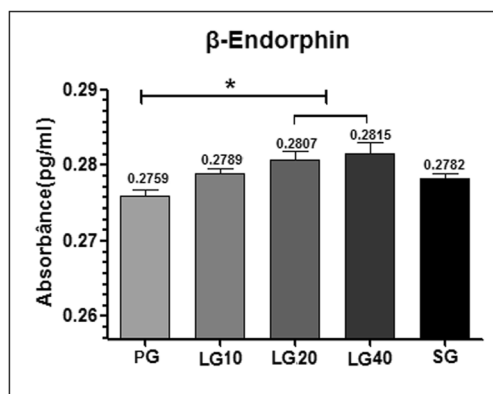
Literature currently suggests that PBM has effective action in reducing painful processes [18, 24, 36], highlighting that the possible PBM mechanisms of action in the production of analgesia can be attributed to the modulation of the inflammatory process, change of the excitation and conduction of the peripheral nerves, and by the stimulation in the increase of the endorphin synthesis [21, 22, 37].

Chow et al. [38] performed a literature review showing the possible mechanisms that PBM could affect during the treatment of peripheral nerve damage, which consequently trigger pain, reporting that the application in peripheral

nerves can slow the conduction velocity (CV) and decrease amplitudes of compound action potentials (CAP) or somatosensory-evoked potentials (SSEP). Importantly, findings establish the principle that photons delivered transcutaneously can inhibit or at least slow or partially block nerve conduction. This blocking or inhibition can be clarified by the action of PBM on the mitochondria that results in increased formation of reactive oxygen species (ROS) being responsible for the energy transduction of the laser in the cell. Chow et al. [39] still discuss in their work the action of PBM on nerve inhibition through the control of mitochondrial membrane potential, and it was observed that the use of PBM with high irradiance values results in a decrease in membrane potential, besides block fast axonal flow in small and medium diameter dorsal root ganglion neurons.

Hagiwara et al. [35] discussed in their study the possible PBM mechanisms of action on analgesia, noting that the pain mechanisms of action cannot be related only to the central nervous system but also to the peripheral nervous system and reported that PBM acts positively in both systems. The study demonstrated that PBM was capable of exerting anti-inflammatory action as well as the stimulation of peripheral opioids such as  $\beta$ -endorphin having a significant analgesic action. In addition, the study presented data corroborate with our study since the groups treated with PBM, especially at higher energy density (20 and 40 J/cm<sup>2</sup>), showed a higher expression of  $\beta$ -endorphin in the blood, being these the same groups that showed significant improvement in pain confirmed by the functional evaluations.

Yamamoto et al. [40] were among the first authors to discuss the PBM action in neuropathic pain in the literature, demonstrating that the action of this therapy may be related to the fact that laser stimulates greater production of endorphins, thus causing a significant analgesia, as found in our study.



**Fig. 4** Absorbance (pg/mL) of  $\beta$ -endorphin found through the ELISA analysis. \* Statistically significant difference from the LG20 and LG40 groups to the PG group ( $p < 0.05$ )

Through photochemical theory, it is believed that the electromagnetic energy stimulates photoreceptor molecules or chromophores that respond to a specific band of light, performing the conversion of photochemical energy. Some authors support the hypothesis of a “therapeutic window” for effective photostimulation above a threshold value but below a value that triggers photo inhibition. The Arndt-Schultz’s Law shows this concept by the existence of a dose-dependent effect represented by a curve versus flow of biological response [41, 42].

Karu [24] suggests that the effectiveness of PBM is strongly related to the application of the appropriate energy density, reporting that high energy density can cause damage to the tissue and low energy density may not be effective. Thus, it is believed that the establishment of energy density used for treatments must have great scientific rigor. Thus, this study sought to identify the most effective flow in reducing neuropathic pain, reporting that energy density of 20 and 40 J/cm<sup>2</sup> promote meaningful analgesia.

Similarly, Bertolini et al. [43] presented a study with animals subjected to compression of the sciatic nerve and subsequently treated with PBM, with different energy density, 4 and 8 J/cm<sup>2</sup>, to reduce the pain. The results showed that PBM was effective in reducing pain, noting that higher energy density with consequently higher power showed a superior improvement over another dosage, as we demonstrated in our study, in which energy density of 20 and 40 J/cm<sup>2</sup> showed more positive responses compared to the fluency of 10 J/cm<sup>2</sup>.

Another study by James et al. [25] used the PBM in an attempt to reduce pain in animals submitted to CCI surgery, an energy 0.9 J per point being applied and for the evaluation of pain, the same tests presented in our study. The results corroborate our study since the PBM-treated animals showed significant improvement in pain at the end of the experiment. It is worth to note that despite the energy density used in study not close to our study, the energy applied by the energy density of 8 J/cm<sup>2</sup> was 0.88 J, and in our study, the energy applied by the energy density of 20 and 40 J/cm<sup>2</sup> was 0.5 and 1.2 J, these being the ones with the best results in the evaluations.

The literature shows that for the promotion of analgesia with PBM therapy, time of application knowledge and its consequences are of great importance [24], noting times greater than or equal to 30 s, not exceeding 60 s, have effective results in treating various types of pain [43]. These findings corroborate our study since the time used 18 and 37 s showed effective results in the promotion of analgesia.

Cotler et al. [44] reported in their study the importance of knowledge and detail of the parameters used in PBM, among the various parameters, the power density, in which the authors report that values up to 5 W/cm<sup>2</sup> are the most indicated

in the use of PBM for the treatment of pain. A review by Andrade et al. [45] reinforces the idea that the use of PBM in the treatment of neuropathic pain generates promising results for use in the clinical area; however, it is emphasized that the studies must present a greater detail of the parameters, so that the therapy is applied effectively.

Through the data presented, it is possible to report that energy density being a major factor at the time of establishing a treatment, beyond knowledge of the energy and time application to be applied is critical in determining the PBM therapy parameters.

PBM on different energy density can significantly reduce painful procedures such as those produced by neuropathy, acting in the stimulation to the release of beta-endorphin, this being a neurotransmitter responsible for the analgesia [46, 47].

However, comparison of studies about PBM in reducing pain, in particular neuropathic, is still difficult, since the dosimetric parameters are very different, in addition to absence of details and thereby impairing their reproduction for treatment [41, 48]. Thus, it is suggested that future studies, with further elaboration, are required to elucidate this question besides the research on higher energy density in an attempt to optimize the control of neuropathic pain.

## Conclusion

According to the data presented, it can be concluded that the PBM, 808 nm, acts positively on the reduction and control of neuropathic pain, noting that higher energy density, as 20 and 40 J/cm<sup>2</sup>, is more effective, besides stimulating greater production of  $\beta$ -endorphin.

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## Compliance with ethical standards

**Conflict of interest** The authors declare that they have no conflict of interest.

## References

1. Julius D, Basbaum AI (2001) Molecular mechanisms of nociception. *Nature* 413:203–210
2. Dworkin RH et al (2010) Recommendations for the pharmacological management of neuropathic pain: an overview and literature update. *Mayo Clinic proceedings*. *Mayo Clin* 85(3):3–14
3. Besson JM (1999) The neurobiology of pain. *Lancet* 353(9164):1610–1615
4. Urban MO, Gebhart GF (1999) Supraspinal contributions to hyperalgesia. *Proc Natl Acad Sci U S A* 96(14):7687–7692

5. Costigan M, Scholz J, Woolf JC (2009) Neuropathic pain: a maladaptive response of the nervous system to damage. *Rev Neurosci* 32:1–32
6. Dickenson A, Suzuki R (2005) Targets in pain and analgesia. In: Hunt SP, Koltzenburg M (eds) *The neurobiology of pain*. Oxford University Press, New York
7. Hunt SP, Bester H (2005) The ascending pain pathways. In: Hunt SP, Koltzenburg M (eds) *The neurobiology of pain*. Oxford University Press, New York, pp 115–137
8. Bennett GJ, Xie YK (1988) A peripheral mononeuropathy in rat that produces disorders of pain sensation like those seen in man. *Pain* 33(1):87–107
9. Seltzer Z, Dubner R, Shir Y (1990) A novel behavioral model of neuropathic pain disorders produced in rats by partial sciatic nerve injury. *Pain* 43(2):205–218
10. Gerard E, Spengler RN, Bonoiu AC, Mahajan SD, Davidson BA, Ding H et al (2015) Chronic constriction injury-induced nociception is relieved by nanomedicine-mediated decrease of rat hippocampal tumor necrosis factor. *Pain* 156(7):1320–1333
11. Mika J, Jurga AM, Starnowska J, Wasylewski M, Rojewska E, Makuch W et al (2015) Effects of chronic doxepin and amitriptyline administration in naive mice and in neuropathic pain mice model. *Neuroscience* 294:38–50
12. Kim KJ, Yoon YW, Chung JM (1997) Comparison of three rodent neuropathic pain models. *Exp Brain Res* 113(2):200–206
13. Reis FJ, Rocha NP (2006) Efeito analgésico de longa duração da dipirona sobre a hiperalgesia persistente induzida pela constricção do nervo ciático em ratos: participação do óxido nítrico. *Rev Bras Cienc Farm São Paulo* 42(2):513–522
14. Colleoni M, Sacerdote P (2010) Murine models of human neuropathic pain. *Biochim Biophys Acta* 1802(10):924–933
15. Serpell MG (2002) Gabapentin in neuropathic pain syndromes: a randomised, double-blind, placebo-controlled trial. *Pain* 99(3):557–566
16. Meier T, Wasner G, Faust M, Kuntzer T, Ochsner F, Hueppe M et al (2003) Efficacy of lidocaine patch 5% in the treatment of focal peripheral neuropathic pain syndromes: a randomized, double-blind, placebo-controlled study. *Pain* 106(1-2):151–158
17. Schestatsky P, Llado-Carbo E, Casanova-Molla J, Alvarez-Blanco S, Valls-Sole J (2008) Small fibre function in patients with meralgia paresthetica. *Pain* 139(2):342–348
18. Chow RT, Johnson MI, Lopes-Martins RA, Bjordal JM (2009) Efficacy of low-level laser therapy in the management of neck pain: a systematic review and meta-analysis of randomised placebo or active-treatment controlled trials. *Lancet* 374(9705):1897–1908
19. Lorenzini L, Giuliani A, Giardino L, Calza L (2010) Laser acupuncture for acute inflammatory, visceral and neuropathic pain relief: an experimental study in the laboratory rat. *Res Vet Sci* 88(1):159–165
20. Meireles A et al (2012) Avaliação do papel de opioides endógenos na analgesia do laser de baixa potência, 820 nm, em Joelho de ratos Wistar. *Rev Dor São Paulo* 13(2):152–155
21. Laakso EL, Cabot PJ (2005) Nociceptive scores and endorphin-containing cells reduced by low-level laser therapy (PBM) in inflamed paws of Wistar rat. *Photomed Laser Surg* 23(1):32–35
22. Bjordal JM, Baxter GD (2006) Ineffective dose and lack of laser output testing in laser shoulder and neck studies. *Photomed Laser Surg* 24(4):533–534
23. Campana EA et al (1999) The relative effects of He Ne laser and meloxicam on experimentally induced inflammation. *Laser Ther* 11(1):36–41
24. Karu T (1999) Primary and secondary mechanisms of action of visible to near-IR radiation on cells. *J Photochem Photobiol B* 49(1):1–17
25. Jameie SB, Masoumpoor M, Janzadeh A, Nasirinezhad F, Kerdari M, Soleimani M (2014) Combined therapeutic effects of low power laser (980nm) and CoQ10 on neuropathic pain in adult male rat. *Med J Islam Repub Iran* 28:58
26. Masoumpoor M, Jameie SB, Janzadeh A, Nasirinezhad F, Soleimani M, Kerdari M (2014) Effects of 660- and 980-nm low-level laser therapy on neuropathic pain relief following chronic constriction injury in rat sciatic nerve. *Lasers Med Sci* 29(5):1593–1598
27. Kuraishi Y et al (1983) Separate involvement of the spinal noradrenergic and serotonergic systems in morphine analgesia: the differences in mechanical and thermal analgesic tests. *Brain Res* 273:245–252
28. Randall LO, Selitto JJ (1957) A method for measurement of analgesic activity on inflamed tissue. *Arch Int Pharmacodyn Ther* 111(4):409–419
29. Griffis CA, Compton P, Doering L (2006) The effect of pain on leucocyte cellular adhesion molecules. *Biol Res Nurs* 7(4):297–312
30. Dray A (2008) Neuropathic pain: emerging treatments. *Br J Anaesth* 101(1):48–58
31. Martins DF et al (2013) Ankle joint mobilization affects postoperative pain through peripheral and central adenosine A1 receptors. *Phys Ther* 93(3):401–412
32. Ward U, Nilsson UG (2013) Acupuncture for postoperative pain in day surgery patients undergoing arthroscopic shoulder surgery. *Clin Nurs Res* 22(1):130–136
33. Mitchinson AR et al (2007) Acute postoperative pain management using massage as an adjuvant therapy: a randomized trial. *Arch Surg* 142(12):1158–1167
34. Cidral-Filho et al (2014) Light-emitting diode therapy induces analgesia in a mouse model of postoperative pain through activation of peripheral opioid receptors and the L-arginine/nitric oxide pathway. *Lasers Med Sci* 29:695–702
35. Hagiwara S, Iwasaka H, Okuda K, Noguchi T (2007) GaAlAs (830 nm) low-level laser enhances peripheral endogenous opioid analgesia in rats. *Lasers Surg Med* 39(10):797–802
36. Medalha CC, Di Gangi GC, Barbosa CB, Fernandes M, Aguiar O, Faloppa F et al (2012) Low-level laser therapy improves repair following complete resection of the sciatic nerve in rats. *Lasers Med Sci* 27(3):629–635
37. Yan W, Chow R, Armati PJ (2011) Inhibitory effects of visible 650-nm and infrared 808-nm laser irradiation on somatosensory and compound muscle action potentials in rat sciatic nerve: implications for laser-induced analgesia. *J Peripher Nerv Syst* 16(2):130–135
38. Chow RT, Armati P, Laakso E-L, Bjordal JM, Baxter GD (2011) Inhibitory effects of laser irradiation on peripheral mammalian nerves and relevance to analgesic effects: a systematic review. *Photomed Laser Surg* 29(6):365–381
39. Chow RT, David MA, Armati PJ (2007) 830 nm laser irradiation induces varicosity formation, reduces mitochondrial membrane potential and blocks fast axonal flow in small and medium diameter rat dorsal root ganglion neurons: Implications for the analgesic effects of 830 nm laser. *J Peripher Nerv Syst* 12(1):28–39
40. Yamamoto H, Ozaki A, Iguchi N, Kinoshita S (1988) Antinociceptive effects of laser irradiation of Hoku point in rats. *Pain Clin* 8:43–48
41. Tiphlova OA, Karu TI (1987) Action of monochromatic low-intensity visible light on growth of *E. coli*. *Microbiology* 60:626–630
42. Low L, Reed A (2001) *Eletroterapia Explicada: Principios e Prática*, 3th edn. Manole Ltda, Barueri-SP
43. Bertolini GR, Artifon EL, Silva TS, Cunha DM, Vigo PR (2011) Low-level laser therapy, at 830 nm, for pain reduction in experimental model of rats with sciatica. *Arq Neuropsiquiatr* 69(2b):356–359
44. Cotler HB, Chow RT, Hamblin MR, Carroll J (2015) The use of low level laser therapy (LLLT) for musculoskeletal pain. *MOJ Orthop Rheumatol* 2(5):00068
45. de Andrade AL, Bossini PS, Parizotto NA (2016) Use of low level laser therapy to control neuropathic pain: a systematic review. *J Photochem Photobiol B* 164:36–42

46. Matera JM, Tatarunas AC, Oliveira SM (2003) Uso do laser arseneto de gálio (904nm) após excisão artroplástica da cabeça do fêmur em cães. *Acta Cir Bras* 18(2):102–106
47. Chavantes MR (2009) *Laser em biomedicina princípios e prática: guia para iniciantes, pesquisadores e discentes na área de saúde e exatas*. Atheneu, São Paulo
48. ME K, Kazemikho N, Aghili R, Forough B, Lajevardi M, Dabaghian FH, Goushegir A, Malek M (2011) Diabetic distal symmetric polyneuropathy: effect of low-intensity laser therapy. *Lasers Med Sci* 26(6):831–835