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# Non-thermal CO2 Laser Therapy (NTCLT): A Novel Photobiomodulative Approach for Immediate Pain Relief of Patchy Oral Mucositis Due to Chemotherapy of Solid Tumors

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## Abstract

**Introduction:** Chemotherapy-induced oral mucositis (COM) is a prominent complication of chemotherapy (CT). Non-thermal CO2 laser therapy (NTCLT) has been demonstrated as an innovative and safe photobiomodulative approach in some kinds of painful oral lesions. The purpose of this study was to evaluate the palliative effects of one session of NTCLT on COM lesions.

**Methods:** Patients with painful COM (WHO grade: ≥2) were included in this before-after clinical trial based on the eligibility criteria. The oral lesions were irradiated with a CO2 laser (power: 1 W, scanning the lesions with the rapid circular motion of the defocused handpiece) through a thick layer (3-4 mm) of a transparent gel containing a high-water content. The severity of pain in the lesions was self-assessed using a 0-to-10 visual analogue scale (VAS) for 7 consecutive days. The evaluating physician visited the patients on the 3rd and 7th days in search of any kind of complications.

**Results:** Seventeen adult patients with 35 patches of OM due to chemotherapy of solid tumors completed the trial. Immediately after NTCLT, the mean for non-contact VAS pain scores of the lesions significantly declined from 4.91 ± 2.356 to 0.29 ± 0.622 ($P<0.001$) and the mean for contact VAS pain scores from 7.77 ± 1.57 to 1.31 ± 1.18 ($P<0.001$). The mean VAS pain scores of the lesions showed statistically significant differences between the follow-up periods compared to the baseline ($P<0.001$). The process was completely pain-free and required no anesthesia. After NTCLT, no kind of thermal adverse effects such as irritation, destruction, aggravation and even erythema were observed.

**Conclusion:** Based on the results of this before-after clinical trial, NTCLT has the potential to be considered as a non-invasive and safe palliative option for the pain management of patchy OM due to chemotherapy of solid tumors.

**Keywords:** Chemotherapy-induced oral mucositis; Non-thermal CO2 laser therapy (NTCLT); Pain relief; Photobiomodulation; Solid tumors.

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## Introduction

Oral mucositis (OM) is a prominent and troublesome adverse effect of non-surgical cancer treatments, which can highly impair the quality of life in patients.[1-4] OM affects up to 100% of the patients undergoing radiation therapy (RT) for head and neck cancer (HNC) and up to 80% of the patients receiving high-dose chemotherapy (CT) before hematopoietic stem cell transplantation (HSCT).[1,2-5] It occurs in approximately 20%-40% of the patients treated by conventional-dose chemotherapy targeting solid tumors.[2,6,7]

The OM lesions typically initiate as erythematous mucosal lesions which may progress towards erosion and frank ulceration which may be extremely painful.[4,8] Pain is the most prominent symptom of OM which can negatively affect the quality of life in patients.[9-11] The pain may strongly impair basic physiological oral functions including eating, drinking, and speech. The severe pain may lead to opioid consumption, nutritional compromise, dehydration, enteral or parenteral nutrition, hospitalization, and even undesirable dose reductions or interruption of chemotherapy in spite of the increased risk of treatment failure.[7,9,10,12,13]

Despite the tremendous growth in OM research and the use of many drugs and non-pharmacological modalities for OM management, there have been no definite preventive or therapeutic options for OM management and its associated pain up to now.[2,13-15] Currently, the goals of OM management are to decrease the incidence of OM, minimize the duration and severity of the lesions, relieve the associated pain, support the appropriate diet, and prevent secondary infections.

Pain control plays a crucial role in the management of OM. Relieving the pain of OM lesions greatly enhances the quality of life in the patients and reduces the need for concomitant analgesic agents including opioids. However, in spite of the importance of pain management of the OM lesions, there are no definite effective and safe therapeutic options for relieving the pain of these lesions.[9,10,15]

In order to standardize the protocols for the management of OM, the Multinational Association of Supportive Care in Cancer and International Society for Oral Oncology (MASCC/ISOO) developed evidence-based OM guidelines for the scientific management of OM.[13,14,16,17] These guidelines which are updated periodically, introduce the drugs and interventions which are most likely to be both effective and safe for appropriate management of OM.[13,14,16,17] The latest MASCC/ISOO clinical practice guidelines (2020) for OM management dedicated one section to photobiomodulation (low-level laser therapy, LLLT).[13]

Numerous clinical trials and several systematic reviews have been published about the application of photobiomodulation therapy (PBMT) for OM management.[13-26] In 2020, with the tremendous growth of high-quality studies with positive results, PBMT reached the level of evidence that allowed the Mucositis Study Group of MASCC/ISOO to recommend the application of PBM in specific groups of patients.[13,15] Currently, ample scientific evidence supports the application of PBMT (by using appropriate laser/light physical parameters) for the prevention of OM in HNC patients receiving RT (with or without CT) and high dose CT, especially for HSCT.[13,15,18,19,27] However, this guideline declared that due to insufficient scientific evidence, no guideline can be suggested or recommended for treating established OM and its associated pain with PBMT.[13,18,19]

During the last decade, the investigators pointed out the valuable effects of CO2 laser application in a non-destructive, non-thermal manner as a photobiomodulation device for pain relief of oral lesions, including recurrent aphthous stomatitis, oral lesions of pemphigus vulgaris, and oral aphthous ulcers of Behcet's disease, with no subsequent thermal adverse effects.[28-38] This non-ablative, non-thermal laser procedure which is completely painless was initially termed NACLT (non-ablative CO2 laser therapy).[30,31,38,39] However, after demonstrating the non-thermal nature of the procedure, it was changed to non-thermal CO2 laser therapy (NTCLT) to prevent misapprehension with surgical fractional non-ablative CO2 lasers applied for cosmetic purposes by dermatologists.[35-37] Due to the significant and instant analgesic impacts of NTCLT on oral lesions in our previous studies and its non-invasive, safe and non-thermal characteristics, this before-after clinical trial was carried out to evaluate the palliative impacts of one session of NTCLT on chemotherapy-induced oral mucositis (COM) lesions.

## Materials and Methods

### *Study Design*

This study was a before-after clinical trial (phase 1 and 2 clinical trials). The study protocol and informed consent were reviewed and approved by the Clinical Ethics Committee of the Royan Institute of Academic Center for Education, Culture, and Research (ACECR), Iran (Number EC/93/1103), and the study was registered in the Iranian Registry of Clinical Trials with IRCT identifier IRCT20220220054073N1 (https://www.irct.ir/trial/63766). Between April 2015 and March 2019, the patients with COM referred to the Department of Medical Laser, Medical Laser Research Center, Yara Institute, were evaluated for inclusion in the study. The study process used in the trial and its experimental character were completely explained to the participants before they signed the informed consent.

### *Patient Selection*

The patients with the diagnosis of COM were referred to

the laser clinic of Medical Laser Research Center, Yara Institute, by their oncologists. The participants were visited and examined by a dermatologist. The participants were included in this study based on eligibility criteria and obtaining written informed consent.

The inclusion criteria were as follows: the existence of painful lesions of OM due to chemotherapy with World Health Organization (WHO) score ≥ 2 OM (WHO OM scale: Grade 0: no findings, Grade I: erythema and soreness; no ulcer, Grade II: oral erythema, oral ulcers, solid diets can be tolerated, Grade III: oral ulcers, only liquid diets can be tolerated, Grade IV: intolerance to both solid and liquid diets), age ≥ 18, compliance with the study protocol, and signing the written informed consent form.

The exclusion criteria were current pregnancy or lactation, photosensitivity or consumption of photosensitizing drugs, past or current head and neck RT, and narcotic consumption.

### *Study Procedure*

To perform NTCLT, prior to laser illumination, we covered the lesion and the tissue surrounding it with a thick (3-4 mm) layer of a non-anesthetic, completely transparent gel containing a very large percentage (87.5%) of water (Abzar Darman Co., Iran). Before initiating laser illumination, the patient and the medical staff put on safety goggles. The CO2 laser (wavelength: 10 600 nm; Lancet-2, Russia) beam (power: 1 W, in continuous mode) was irradiated through the gel layer. The lesion was scanned with the rapid circular motion of the handpiece over the lesion, while the distance of the tip of the defocused handpiece from the surface of the gel was about 5-6 mm. The laser beam was illuminated to a circle area of the lesion (with an approximate diameter of 1cm) for 5 seconds in every pass. If the pain persisted, the procedure was repeated. The gel layer was gently wiped after every pass, and a new gel layer was put on the lesion to preserve the water content of the gel and prevent subsequent tissue injury. Since the results of prior NTCLT studies demonstrated the pain-free character of the procedure and the patients in this trial reported no pain during NTCLT, no anesthesia was required.

### *Assessments*

The participants were instructed on how to measure the non-contact (non-stimulant) and contact pain scores of their lesions. Non-contact (non-stimulant) pain referred to the pain the patient felt in the lesion without any kind of mechanical or chemical stimulus. The contact pain referred to the pain the patient experienced in the lesion with the pressure contact of a medical cotton swab. The pain severity of the lesions was self-assessed using a 0-to-10 visual analogue scale (VAS). VAS = 0 showed "no pain" and VAS = 10 showed "maximum and the worst imaginable pain". The patients' pain scores were recorded before NTCLT, immediately after the procedure, 4, 8 and 24 hours later, and then every day for seven consecutive days. The evaluating physician visited the patients on the 3rd and 7th days in search of any potential complications.

### *Statistical Analysis*

The data regarding the patients' responses were analyzed by SPSS, version 18. The results were expressed as mean ± standard deviation (SD). Statistical significance was tested by using Student's *t* test for paired samples and repeated measures analysis. Statistical significance was set at $P < 0.05$.

## Results

Seventeen adult patients with 35 patches of OM due to chemotherapy were recruited in the trial from April 2015 to March 2019. All of these patients had been treated with conventional-dose chemotherapy for their solid tumors. The demographic and baseline attributes of the participants are presented in Table 1.

All the participants were female. Their mean age was 45.70 years, and the age range was 36-55 years. All the patients had solid tumors. The underlying malignancy in 14 (82.36%) patients was breast cancer, and in three (17.64%) patients it was metastatic cancer. All the patients had patchy OM, and none of them had diffuse OM. The most common sites of involvement were the lip (28.6%) and lateral tongue (28.6%). The other sites of involvement included the cheek (14.3%), the dorsal tongue (11.4%), the gum (8.6%), the ventral tongue (5.7%), and the tip of the tongue (2.9%). The WHO grade of OM in 11 (64.7%) patients was 2, in 5 (29.41%) patients was 3, and in one (5.89%) patient was 4.

The baseline VAS pain scores of the OM lesions are shown in Table 1.

### *The Results of the Analgesic Effects of NTCLT on Non-contact Pain Scores*

The mean ± SD of the baseline VAS pain scores for non-contact pain was 4.91 ± 2.356 (range: 2-10). Immediately after NTCLT, the mean ± SD of the VAS pain scores significantly declined to 0.29 ± 0.622 ($P < 0.001$), and in 77.1% of the patients decreased to zero. These means were 0.71 ± 1.1 and 1.09 ± 1.86 at 4 and 8 hours post-treatment, respectively. These means were 1.09 ± 1.91, 0.94 ± 1.90, 0.86 ± 1.83, 0.71 ± 1.82, 0.6 ± 1.81 and 0.6 ± 1.81 on the 2nd, 3rd, 4th, 5th, 6th and 7th days, respectively. The mean scores of non-contact pain showed statistically significant differences in the follow-up compared to the baseline scores ($P < 0.001$). The average non-contact pain scores (VAS) in the baseline and follow-up are shown in Figure 1.

**Table 1.** Demographic Data and Baseline VAS Pain Scores of the OM Lesions

| No. | Age | Cancer type | WHO Grade | No of Lesions | Location of Lesions | VAS (Non-Contact Pain) | VAS (Contact Pain) |
|---|---|---|---|---|---|---|---|
| 1 | 36 | Breast | 3 | 1 | Lat. tongue | 10 | 10 |
| | | | | 2 | Cheek | 7 | 10 |
| 2 | 36 | Breast | 2 | 3 | Lip | 6 | 8 |
| | | | | 4 | Vent. tongue | 7 | 8 |
| | | | | 5 | Cheek | 7 | 9 |
| 3 | 50 | Breast | 2 | 6 | Lat. tongue | 5 | 10 |
| 4 | 37 | Breast | 2 | 7 | Tip of tongue | 2 | 7 |
| | | | | 8 | Lip | 3 | 7 |
| 5 | 55 | Breast | 2 | 9 | Dorsal Tongue | 5 | 8 |
| 6 | 54 | Metastatic cancer | 3 | 10 | lip | 5 | 7 |
| 7 | 42 | Breast | 3 | 11 | lip | 3 | 8 |
| | | | | 12 | lip | 2 | 8 |
| | | | | 13 | Ventral tongue | 3 | 7 |
| | | | | 14 | Dorsal tongue | 2 | 7 |
| | | | | 15 | Dorsal tongue | 3 | 7 |
| 8 | 36 | Breast | 2 | 16 | lip | 6 | 8 |
| | | | | 17 | Gum | 6 | 8 |
| | | | | 18 | Lat. tongue | 8 | 10 |
| | | | | 19 | Gum | 10 | 10 |
| | | | | 20 | Lat. tongue | 9 | 10 |
| 9 | 53 | Breast | 2 | 21 | Lat. tongue | 3 | 5 |
| | | | | 22 | Lateral tongue | 4 | 6 |
| 10 | 53 | Breast | 2 | 23 | Dorsal tongue | 6 | 7 |
| | | | | 24 | Cheek | 7 | 7 |
| 11 | 53 | Breast | 2 | 25 | Lip | 4 | 5 |
| | | | | 26 | Lip | 3 | 6 |
| 12 | 54 | Breast | 2 | 27 | Cheek | 4 | 7 |
| | | | | 28 | Lip | 4 | 8 |
| 13 | 38 | Metastatic cancer | 4 | 29 | Lip | 9 | 10 |
| 14 | 47 | Breast | 2 | 30 | Cheek | 4 | 10 |
| 15 | 42 | Breast | 3 | 31 | Lat. tongue | 3 | 7 |
| | | | | 32 | Lat. tongue | 3 | 7 |
| | | | | 33 | Lat. tongue | 3 | 8 |
| 16 | 48 | Breast | 2 | 34 | Lat. tongue | 3 | 4 |
| 17 | 43 | Metastatic cancer | 3 | 35 | Gum | 4 | 8 |

### *The Results of the Analgesic Effects of NTCLT on Contact Pain Scores*

The mean ± SD of the baseline VAS scores for contact pain was 7.77 ± 1.57 (range: 4-10). Immediately after NTCLT, these means significantly declined to 1.31 ± 1.18 ($P < 0.001$). These means were 1.91 ± 1.54 and 2.31 ± 2.33 at 4 and 8 hours post-treatment, respectively. These means were 2.37 ± 2.69, 2.14 ± 2.56, 1.74 ± 2.55, 1.43 ± 2.53, 1.11 ± 2.54 and 1.09 ± 2.58 on the 2nd, 3rd, 4th, 5th, 6th and 7th days, respectively. The mean contact pain scores showed statistically significant differences in the follow-up compared to the baseline ($P < 0.001$). The two sets of scores are shown in Figure 2.

The NTCLT process incurred no pain requiring no systemic or local anesthesia. Immediately following NTCLT, no thermal adverse effects including burn, ablation, destruction, aggravation of the lesions, or even erythema were observed. No kinds of visible complications were detected during the patients' follow-up periods.

## Discussion

This study provides evidence that NTCLT has the potential to be considered a novel, non-invasive, and safe PBM procedure for significant and instant pain relief of

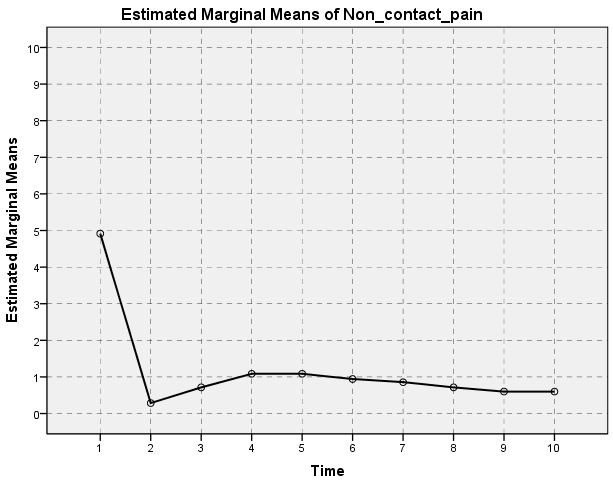


**Figure 1**. Mean Non-contact Pain Scores (VAS) in Baseline and Follow-up Sessions

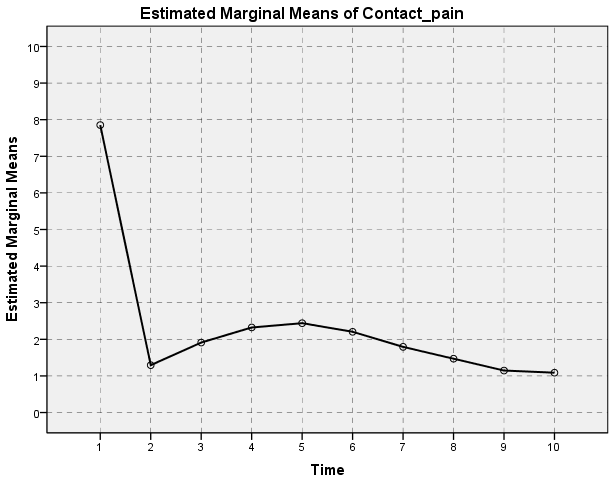


**Figure 2.** Mean Contact Pain Scores (VAS) in Baseline and Follow-up Sessions

patchy OM due to the chemotherapy of solid tumors. No thermal complications were detected after the procedure and throughout the patients' follow-up periods. NTCLT was completely painless and no anesthesia was required.

Recently, with the considerable growth of high-quality studies with positive results, PBMT reached the level of evidence that in 2020, the Mucositis Study Group of MASCC/ISOO recommended the application of PBMT/ LLLT for OM prevention and its associated pain in specific groups of patients, including adult patients who undergo HSCT conditioned with high-dose chemotherapy (with total body irradiation or without it) and adult HNC patients who undergo RT with or without CT.[13,18,19]

It should be noted that the vast majority of the studies have used PBMT for the prevention of OM rather than correction and alleviation of the lesions.[18,19,25] In these studies, with prophylactic purposes, PBMT was initiated before or concomitant with the administration of CT and/or RT and continued coincident with active cancer treatments.[19] Practically, the available clinical practice guidelines focus on the prevention of OM with PBMT.[18,19] The results of systematic reviews also confirm that the most effective approach for the application of PBMT in OM management is prevention rather than correction and palliation of the lesions.[18,19] In 2020, The Mucositis Study Group of MASCC/ISOO declared that due to insufficient scientific evidence, no guideline can be suggested or recommended for treating established OM and its associated pain with PBMT.[13,18,19] In fact, currently, there is no consensus on photobiomodulation protocols for the pain management of established COM, and practically the MASCC/ISOO clinical guidelines are used for the prevention of OM with PBMT.[13,18,19]

On the other hand, in patients receiving high-dose CT prior to HSCT and in HNC patients who receive RT, the OM incidence, severity and its lethal complications are much higher than that of conventional dose CT for treating solid tumors. Therefore, it is not surprising that the vast majority of the studies focus on the management of ROM and OM due to high-dose chemotherapy (especially HSCT patients) and not COM due to conventional dose CT.

The number of studies in which PBMT was initiated after the establishment of COM is limited.[40-45] However, there is growing attention to the evaluation of the beneficial effects of PBMT/LLLT on established COM.[40,44]

In a before-after clinical trial, Nes evaluated the analgesic effects of LLLT (AsGaAl 830 nm, potency: 250 mW, 35 J $cm^{-2}$) on 13 adult patients with established COM. The patients were treated daily for five consecutive days. Every day, the VAS before-treatment and post-treatment pain scores were compared with each other. The average daily reduction of post-treatment VAS pain scores compared to the pretreatment scores of the same day was 67% ($P=0.007$). This reduction of VAS pain scores varied from 47% (the first day) to 94% (the fifth day).[45]

In another study, 40 adult patients with painful oral lesions of COM were treated with either the InGaAlP laser (660 nm, potency: 40 mW, 0.24 J per point) or the LED (630 nm, potency: 80 mW, 0.24 J per point) for 10 successive days, except for the weekends. The number of LLLT sessions required to achieve a VAS pain score 0 was 4.4-6 for patients in the InGaAlP laser group and 1.5-8 for patients in the LED group.[42]

In a pilot before-after clinical trial, Cauwels assessed the palliative impacts of LLLT (GaAlAs 830 nm, potency: 150 mW) on 16 children (mean age = 9.4 years) with established COM. LLLT sessions were performed every 48 hours until each lesion healed. Immediately after irradiation of the lesions, pain scores decreased considerably.[41]

In a multicenter RCT, 101 children with established COM with WHO grade > 2 COM were randomized to receive either PBM therapy (diode laser, 660 and 970

nm -combined wavelengths, irradiance: 320 mWcm$^{-2}$, fluence:36.8 J cm$^{-2}$) or placebo (sham treatment) for four successive days. A significant pain reduction was reported on the 7th day in the PBMT/LLLT group versus the sham group ($P < 0.006$). No significant complications were recorded.[43]

In these studies, semiconductor lasers or LED systems were used for PBMT. The significant advancements in the field of PBMT and the great expanding body of literature in PBMT have provided creative ways for the application of different laser/light sources including high-power, surgical lasers for PBM. Some studies demonstrated that in addition to the traditional PBMT devices (including He-Ne laser, semiconductor lasers, and LED systems) by some arrangements, it is possible to use surgical lasers for PBMT/LLLT. As Tuner declared, "When high-power lasers are used for biostimulation, one only needs to make the beam wide enough not to burn. An alternative is to scan rapidly over the lesion with a narrow beam. Therefore, the power density or average power is kept low enough to avoid burning, and their power density is set within the low-intensity laser therapy range" .[46] For example, surgical lasers such as CO2 laser 10 600 nm, Nd: YAG laser 1064 nm and ruby laser 694 nm, if used in the defocused mode at a setting in which their energy density is too low to avoid tissue burn, can be used for PBMT.[46] The famous experiment of Endre Mester, who irradiated the shaved areas of the skin of mice with very low-powered ruby laser and faster hair regrowing on the irradiated areas can be considered the first demonstration of using a surgical laser for PBMT/LLLT.[46]

The investigators have pointed out the valuable effects of the CO2 laser appliance in a non-destructive, non-thermal manner as a photobiomodulation device for relieving the pain of oral diseases such as recurrent aphthous stomatitis, oral aphthous ulcers of Behcet's disease, and oral lesions of pemphigus vulgaris.[28-38]

In addition to the helpful recommendations of Tuner for the application of surgical lasers such as the CO2 laser for PBMT (irradiation using a defocused handpiece or scanning of the lesion using a narrow beam), illumination of the laser beam through a gel with a very high percentage of water can be another contraption to significantly decrease the final beam power to the level of photobiomodulation lasers. For using the CO2 laser (10 600 nm) as a non-thermal and non-destructive laser for PBMT (NTCLT), these arrangements should be considered:

- Covering the lesion and the tissues surrounding it with a sufficiently thick layer of an appropriate gel. This gel should be essentially transparent, containing a very high percentage of water and with no anesthetic properties.
- Applying the CO2 laser using a defocused handpiece, with a 5–6 mm distance of the handpiece from the gel surface.
- Scanning the lesion by rapidly moving the CO2 laser handpiece in a circling motion.

The results of powermetry and some other physical investigations shows that by these provisions, the final beam power significantly drops by a factor of 200-500 after irradiation through the gel and to the level of photobiomodulation lasers.[30] With such provisions, the CO2 laser can be successfully applied as a non-thermal, non-destructive PBM device for the immediate and significant pain management of such mucosal lesions as oral aphthous ulcers, oral lesions of pemphigus vulgaris, and oral and genital aphthous ulcers of Behcet's disease with no subsequent thermal adverse effects.[30-38] It is a painless procedure and needs no anesthesia. This non-destructive laser technique was originally termed NACLT.[30,31,38,39,47] but after demonstrating its non-thermal features, it was called NTCLT to prevent misapprehension with the surgical fractional non-ablative CO2 laser applied for cosmetic purposes by dermatologists.[35-37] The results of an overview of systematic reviews in 2020 viewed NTCLT as one of the best two recommended photobiomodulative systems for both pain control and accelerating the healing of recurrent oral aphthous ulcers.[47] The scope of applications of NTCLT as an effective and safe PBM technique for pain relief of the lesions seems to be expanding to the non-mucosal lesions too.[48]

This study provides evidence that a single session of NTCLT has the potential to be applied for significant and instant pain relief of patchy OM due to chemotherapy of solid tumors without any visible side effects. The application of NTCLT as a "single session" procedure can be considered an important advantage of this PBM technique. Whether in OM patients with a relapse of pain, another session of NTCLT might be helpful for pain relief of the lesions or not is a concept that can be evaluated in future studies.

## Limitations and Suggestions for Future Studies

This study has some limitations too. In this before-after clinical trial, we did not assess the healing process of the OM lesions. Certainly, further randomized controlled clinical trials with control groups will be able to evaluate whether the healing processes of the OM lesions (at least to some extent) may impact the persistence of the analgesic effects of NTCLT or not.

In this clinical trial, all of the patients had patchy COM. Therefore, the positive results of this study cannot be extended to diffuse COM. We hope that further clinical trials will be able to study the palliative effects of NTCLT on diffuse OM. In addition, all the patients had solid tumors and they were treated with conventional-dose chemotherapy. Therefore, the positive results of this study cannot be extended to the more severe OM lesions

due to high-dose chemotherapy or radiotherapy. As another suggestion for future studies, the pain-relieving effects of NTCLT can be evaluated on established ROM and COM due to high dose CT including HSCT patients

## Conclusion

Based on the findings of this clinical trial, NTCLT can be considered a promising, non-invasive, and pain-free photobiomodulative technique for significant and instant pain relief of patchy OM due to chemotherapy of solid tumors. No thermal complications were detected after the procedure and in the patients' follow-up periods. NTCLT was completely painless and no anesthesia was required.

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### Authors' Contribution

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**Supervision:** Nasrin Zand.
**Writing–original draft:** Nasrin Zand, Jila Sadighi.
**Writing–review & editing:** Nasrin Zand, Jila Sadighi, Parvin Mansouri, Homa Mahdavi.

### Competing Interests

None.

### Ethical Approval

The study protocol was reviewed and approved by the ACECR Royan Bio-Medical Research Ethics Committee (Code: EC/93/1103). The study was registered in the Iranian Registry of Clinical Trials with the IRCT identifier IRCT20220220054073N1 (https://www.irct.ir/trial/63766). Respecting the confidentiality of identity information, the principal investigator protected the identifiable data of research participants.

The study process used in the trial and its experimental character were completely explained to the participants before they signed the informed consent form. The aim and protocol of the study were described clearly, and patients had sufficient time to make their decisions to participate in the study. The informed consent form to participate in the study included the following issues: the aim of the study, the possible benefits and risks of participating in the study, voluntary participation in the research process, and the right to withdraw from the research without affecting their current therapeutic care, emphasizing the participant's confidentiality and no cost for participating in the research.

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