

**“Low power laser in the prevention of induced oral mucositis in bone marrow transplantation patients: a randomized trial”**

**HS Antunes**<sup>1,5</sup>, AM de Azevedo<sup>2</sup>, LFS Bouzas<sup>2</sup>, CAE Adão<sup>2</sup>, CT Pinheiro<sup>1</sup>, R Mayhe<sup>1</sup>, LH Pinheiro<sup>2</sup>, R Azevedo<sup>2</sup>, VD Matos<sup>1</sup>, PC Rodrigues<sup>3</sup>, IA Small<sup>4</sup>, RA Zângaro<sup>5</sup>, CG Ferreira<sup>4</sup>.

- 1- Section of Dentistry - Instituto Nacional de Câncer (INCA) - Brasil
- 2- Bone Marrow Transplant Center (CEMO) - INCA - Brasil
- 3- Therapy and Technology Development Section - INCA - Brasil
- 4- Clinical Research Service - INCA - Brasil
- 5- Institute for Research and Development - Universidade do Vale do Paraíba – Brasil

**Running Head: Laser and prevention of oral mucositis.**

**Héilton Spíndola Antunes:** Designed research, performed research, analyzed data, wrote the paper and final approval of manuscript.

**Alexandre Melo de Azevedo:** Designed research and final approval of manuscript.

**Luiz Fernando da Silva Bouzas:** Designed research, administrative support and final approval of manuscript.

**Carlos Alberto Esteves Adão, Cláudia Tereza Pinheiro, Renato Mayhe, Lucia Helena Pinheiro, Renato Azevedo, Valkiria D’Aiuto de Matos:** Performed research.

**Pedro Carvalho Rodrigues e Isabele Ávila Small:** Analyzed data.

**Renato Amaro Zângaro, Carlos Gil Ferreira:** Designed research, analyzed data, wrote the paper and final approval of manuscript.

Correspondence:

Héilton Spíndola Antunes

Seção de Odontologia - Serviço de Pesquisa Clínica - Instituto Nacional de Câncer (INCA)

Rua André Cavalcante 37, 2º andar.

Rio de Janeiro, Brasil.

CEP: 20231-050.

e-mail: [hspindola@inca.gov.br](mailto:hspindola@inca.gov.br)

## Abstract

We investigated the clinical effects of low power laser therapy (LPLT) on prevention and reduction of severity of conditioning-induced oral mucositis (OM) for hematopoietic stem cell transplant (HSCT). Were randomized 38 patients submitted to autologous (AT) or allogeneic (AL) HSCT. An diode InGaAlP was used emitting light at 660 nm, 50 mW and 4 J/cm<sup>2</sup> measured at the end of fiberoptic with 0.196cm<sup>2</sup> of section area. The evaluation of OM was done by OMAS and WHO scale. In the LPLT group 94.7% of patients had OM (WHO) Grade  $\leq$  2, including 63.2% with Grade 0 and 1, whereas in the controls group 31.5% of patients had OM Grade  $\leq$  2 (p<0.001). Remarkably the HR for grade 2, 3 and 4 OM was 0.41 (0.22-0.75, p = 0.002) and for grade 3 and 4 was 0.07 (0.11-0.53, p<0.000). Using OMAS by the calculation of ulcerous area, 5.3% of laser group presented ulcerous of 9.1 to 18 cm<sup>2</sup>, whereas 73.6% of the control group presented ulcers from 9.1 to 18 cm<sup>2</sup> (p=0.003). Our results indicate that the use of upfront LPLT in HSCT patients is a powerful instrument in reducing the incidence of OM and is now standard in our center.

**Keywords:** Low power laser therapy, Hematopoietic stem cell transplantation, Oral mucositis, Radiotherapy, Chemotherapy.

## Introduction

High-dose chemotherapy administered as part of the preparative regimens prior to HSCT has a direct cytotoxic effect on the oral epithelium, connective tissue and matrix extracellular leading to injury or disruption of the mucosal barrier<sup>1</sup>. Oral and gastrointestinal mucositis may occur in up to 100% of the patients undergoing high-dose chemotherapy (CT) with HSCT. Oral mucositis is associated with an increase in the incidence of systemic infections because of disruption of the natural mucosal barrier and impacts both the on length of hospital stay and on the complications associated with HSCT<sup>1,2,3</sup>.

Oral mucositis clinical presentation consists of mucosal blending, erythema, edema and progresses to ulceration with or without pseudomembrane formation, that develop most commonly on the non-keratinized mucosa of the floor of the mouth, tongue, buccal mucosa, and soft palate<sup>4</sup>. The initial stage of tissue injury occurs rapidly following the administration of radiation (RT) or chemotherapy who trigger both DNA and non-DNA damage. DNA strand breaks result in direct cellular injury that targets cells in the basal epithelium as well as cells within the submucosa. Although the mucosa seems to be absolutely normal at this stage, a cascade of events is ongoing in the submucosa that ultimately results in mucosal destruction<sup>5</sup>. The current management of such oral mucositis is directed at prevention, palliation, infection prevention and treatment<sup>6,7</sup>.

LPLT have been used in an attempt to reduce the incidence of oral mucositis and its associated pain in patients who are receiving high-dose CT or chemoradiotherapy prior to HSCT<sup>8,9,10,11,12</sup>. Over the last several years, appropriate laboratory and clinical evidence has been accumulating steadily to also support the use of LPLT to promote biomodulation<sup>13,14,15</sup>. It has been reported that LPLT promotes wound healing and reduces pain and inflammation. Different effects appear to be related to laser characteristics and the particular type of tissue being treated<sup>16</sup>. Helium-neon (He-Ne) laser (632.8 nm) treatment has been the most frequently studied form of LPLT for the prevention or reduction of oral mucositis and oral pain associated with cancer therapy (including HSCT). Research currently is underway on the use of diode lasers with wavelengths ranging from 650 to 905 nm. It appears that laser therapy produces no toxicity and is atraumatic to patients<sup>5</sup>.

In the current trial we investigated the clinical effects of InGAIP (660nm) laser on prevention and reduction of severity of conditioning-induced oral mucositis for HSCT patients.

## **Patients and Methods**

### **Patients**

This was a randomized, placebo-controlled, quantity and prospective clinical trial. Between January 4<sup>th</sup>, 2004 and May 20<sup>th</sup>, 2005, 38 patients were evaluated and submitted to HSCT in Centro de Transplante de Medula Óssea (CEMO). The research was performed in compliance with the Resolution nº 196/96 of the National Health Counsel of Brazil, and was submitted to both the Ethics Committee of the Instituto Nacional de Câncer (INCA) and of the Universidade do Vale do Paraíba (UNIVAP). Patients were randomized on the day admission for the transplant between receiving lasertherapy (laser or experimental group), or not receiving the lasertherapy (placebo or control group).

#### **Inclusion Criteria**

Age  $\geq$  18 years old, hematologic disease nominate HSCT, oral mucous intact in the first day of the conditioning (D-7), absence of visible plaques on the teeth upon inspection, capacity to cooperate with the treatment, patients that after the information and instruction section have signed the consenting text giving free confirmation and being elucidated.

#### **Exclusion criteria**

Patients who did not fulfill the inclusion criteria (e.g. allogeneic or autologous not myeloablative transplant), patients who were receiving drugs for the treatment and/or prevention of mucositis, patients who were not previously evaluated and released by the author.

#### **Oral care**

Performed by the dentist before admission for the HSCT. Dental care included education about oral hygiene, panoramic radiograph, oral examination with attention to soft tissues, bones, tooth and periodontal exam, removal of sub and supragingival calculus. Eliminate sources of trauma caused by orthodontic bands and brackets, tooth or prosthesis, extraction of teeth with signs or symptoms indicative of potentially bad prognosis (active

periodontal disease, teeth requiring endodontic treatment or with extended caries and coronary destruction)<sup>17-18</sup>. All the patients had carried out oral hygiene with extra-soft toothbrushes, dental paste with peroxidase system after every meal and mouth rinses with an ethanol-free 0.12 % chlorhexidine solution<sup>19,20,21</sup> containing xylitol from D -7 until neutrophil recovery (granulocytes > 500/mm<sup>3</sup>), three times a day (morning, afternoon and night).

## **Methods**

### **Conditioning Regimens**

The characteristics of the conditioning regimens are summarized in table 1. Patients received Fluconazole 200 mg IV 12/12 h from D-2 and Acyclovir 500 mg/m<sup>2</sup> IV 8/8 h from D-2 until neutrophil recovery.

Samples for blood cultures were collected by central catheter and peripheral veins in case of febrile episodes followed by empiric therapy with broad-spectrum antibiotics.

### **Laser Application**

LPLT was started on the first day of the conditioning (D-7) and stopped on the day of the neutrophil recovery. The dentists were the only members of the team who knew to which group the patient was randomized to. The same equipment was used in all applications. The irradiation used was a 50 mW InGaAlP diode-laser, emitting continuous light at 660 nm, with a real power output of 46.7 mW and energy density (ED) of 4 J/cm<sup>2</sup> measured at the end of fiberoptic with 0.196cm<sup>2</sup> of section area during the experiment. It was applied in a punctual form, side by side, touching the material, during 16.7s per point, totalizing 15 points per region. The oral cavity regions previously treated were: upper lip, lower lip (redness and lip mucosa), buccal mucosa, dorsum, ventral and lateral tongue, floor of mouth, hard and soft palate. Before the application of the laser the tip was wrapped with a PVC film and after this procedure was disinfected with 70% alcoholic solution. For protection all patients received eyeglasses with total blocking of the light, to be used during the application of the laser.

A crossover was allowed for patients from the control group who presented a Grade 4 oral mucositis index of the World Health Organization (WHO)<sup>22</sup> and/or an ulcer area  $\geq 12$  cm according to the Oral Mucositis Assessment Scale (OMAS)<sup>23</sup>. From this moment, therapeutical laser with 8J/cm<sup>2</sup> per point was applied to these patients. The patients in the laser group who had presented erythema or ulcers continued to receive the preventive laser with 4J/cm<sup>2</sup>.

### **Laser therapy Evaluation**

With the purpose of minimizing interobserver variation and familiarizing the team with the measurements scales for mucositis a CD-ROM containing the research protocol, as well as photographic examples of normal and damage oral mucosa (mucositis) was given to all the professionals involved in the application of LPLT and on the evaluation of patients. One dentist and three nurses (blinded for the study), performed daily oral evaluation of the patients from day -7 until the neutrophil recovery. The results were catalogued and analyzed according with WHO's scale, OMAS and Visual Analogue Scale (VAS)<sup>23</sup>.

### **Statistics Analysis**

For the WHO's scale and OMAS the chi-square test ( $\chi^2$ ) was applied. Correlation between the scores in the WHO scale and OMAS was assessed by the F-test in analysis of variance (ANOVA), and Bonferroni's test. Mucositis-free survival was calculated from the first day of conditioning trough the neutrophil recovery by the Kaplan-Meier method. The concordance index (CI) between the evaluators was measured (CI=81.7% $\pm$ 1,96 $\sqrt$ 81.7x18.3:520). The first day and the duration (in days) of mucositis were compared between both groups, using the Mann-Whitney test. The VAS scores of pain were compared using Student's test. All p values <0.05 were considered as statistically significant.

## **Results**

All the 38 patients completed the study and none were lost to follow-up or excluded for failure to complete the laser application protocol. The treatment was well tolerated and no toxicity from LPLT was recorded. Patients characteristics are summarized in table 2.

### **Mucositis evaluation by WHO criteria**

Using the WHO's scale it was observed that the laser group patients presented less intense oral mucositis (WHO Grade 0-1; Figure 1). The proportion of patients in the LPLT and placebo groups who developed Grade 0 or 1 mucositis (without ulcers) was 63.2% (12 of 19) including 3 patients submitted to total body irradiation (TBI) and 10.5% (2 of 19), respectively ( $p < 0.001$ ). Six patients in the LPLT group (31.5%) had small ulcers, (WHO Grade 2), totalizing 94.7% of the patients in this group with a WHO Grade between 0 and 2. The control group behaved in the opposite way ( $p < 0.001$ ). In order to better estimate the impact of LPLT, the mucositis-free survival was analyzed separately in the strata of patients with Grades 2, 3 and 4 and Grades 3 and 4 patients, respectively. The hazard ratio for Grades 2, 3 and 4 mucositis was 0.41 (0.22-0.757;  $p = 0.002$ ), whereas for Grade 3 and 4 only it was 0.07 (0.11-0.53; Figure 2 and 3).

### **Oral mucositis evaluation by OMAS criteria**

The evaluation by OMAS criteria was done both by using the calculation of the weighted average of the ulcerous area plus erythema's intensity and by ulcerous area only. Sixteen patients from the laser group (84.2%) stayed on the weighted average zone of 0-2.9, while only 5 patients from the control group (26.3%) stayed in the same zone ( $p = 0.007$ ; Figure 4). It was observed that the patients of the laser group presented small extension of the ulcerous area. with ( $p = 0.003$ ; Figure 5). In addition, it was observed that the control group patients showed mucositis earlier D +5 than the laser group D +6 ( $U^* = 0.42$  with  $p = 0.67$ ); stayed more time with mucositis, laser and control groups with an average of 6 and 9 days, respectively ( $U^* = 1.52$  with  $p = 0.13$ ) and, therefore, needed more time for it to heal, comparing to the laser group ( $U^* = 1.45$  with  $p = 0.15$ ), being observed that the average time for the laser application in the control group was 6 days . In total 24 patients presented ulcer in the oral cavity and the most affected areas were: buccal mucosa with 20.5 patients (85.4%), lateral of the tongue with 19 patients (79.1%) and ventral of the tongue with 17 patients (70.8%).

### **Correlation between WHO's scale and OMAS**

In this analysis arithmetic averages and standard deflection were used for comparison of the WHO's scale with the OMAS weighted average (WA). A significant difference was

detected between the Grade 1 (WA=1.25) and Grade 2 (WA=2.07); Grade 1 (WA=1.25) and Grade 3 (WA=3.72); Grade 1 (WA=1.25) and Grade 4 (WA=3.5); Grade 2 (WA=2.07) and Grade 3 (WA=3.72) and Grade 2 (WA=2.07) and Grade 4 (WA=3.5), with “F”=149.98 ( $p<0.001$ ). However a significant difference was not observed between Grades 3 (WA=3.72) and 4 (WA=3.5).

### **Level of agreement among evaluators and pain evaluation**

A significant agreement between the evaluators occurred, with an agreement index of 81.7%. Regarding the presence and intensity of pain, a significant difference was not noticed since 14 (73.7%) patients from the laser group (VAS 7) and 16 (84.2%) patients from the control group (VAS 8) reported pain ( $p=0.13$ ).

### **Impact of LPLT on clinical outcomes**

Concerning the results of the blood cultures, overall 28 (73.8%) patients presented negative blood cultures and 10 (26.3%) positive blood cultures. However no differences between LPLT and control group were observed. Further among the positive blood cultures no *Streptococcus* were identified and no tooth and gingival complication detected. Although not planned, an analysis of the impact of LPLT on survival and treatment related mortality was performed. Marginal differences in survival ( $p = 0.04$ ) but not in treatment related mortality (TRM) favoring the LPLT group were detected (data not shown).

## **Discussion**

The potential positive effects from LPLT as a preventive treatment method for patients with high probability to develop OM such as those submitted to HSCT has been postulated<sup>8,10,12</sup>. However confirmatory randomized trials with more appropriate design are lacking. The study presented here differs from two previous randomized trials in 3 ways: the population included, the LPLT and the criteria (scores) used for analysis.

The majority of our patients were submitted to allogeneic transplant (Table 2), whereas both Cowen<sup>10</sup> and Barasch<sup>8</sup> studies enrolled only HSCT autologous except for one patient. Since allogeneic transplant lead to a more severe OM, our population may be considered more vulnerable and the results more remarkable. However the fact that TBI was applied to 100% of Cowen's patients study, but only to 10% of our patients may have counterbalanced the previous difference making comparisons among the population described here and elsewhere even harder.

Despite the use of preventive LPLT a high incidence of ulcers was still observed in previous randomized studies<sup>8,10</sup>. In one study laser illumination continuous with 632.8 nm wavelength, power 25 mW, energy density of 1 J/cm<sup>2</sup>, from day -1 to +3 was applied<sup>8</sup>. It should be highlighted that in that study 20 patients served as their own control since the randomization was done between right and left of the buccal mucosa midline. Cowen<sup>10</sup> applied laser illumination continuous with 632.8 nm wavelength, power 25 mW, energy density of 1.5 J/cm<sup>2</sup>, from day -5 to -1 to 30 patients. In our trial both a higher energy density (4 J/cm<sup>2</sup>) and a longer administration (from day -7 until de neutrophil recovery) were applied. In marking contrast to previous data, this strategy showed to be highly effective since 63.2% of the patients did not present OM, whereas Barasch<sup>8</sup> and Cowen<sup>10</sup> report that 100% of the patients presented OM after HSCT. Further Migliorati<sup>12</sup> applied laser illumination continuous with 780 nm wavelength, power 60 mW, energy density of 2 J/cm<sup>2</sup>, from day -5 to + 5 to 11 patients and report that 63.7% of the patients presented OM Grades 3 and 4 (WHO) and 9% of the patients presented OM Grade 2 after HSCT and high-dose QT. Our data suggest that both higher laser energy density and length of the application may be pivotal for the outcome of LPLT preventive treatment. In line with that, recent data from a randomized clinical in which 60 children received LPLT (780 nm wavelength, power 60 mW and energy density of 4 J/cm<sup>2</sup>) for a short period of time (days 1 to 5) showed no difference between LPLT and control groups. It should be highlighted that the groups treated are heterogeneous including both patients treated with both conditioning and conventional chemotherapy regimens<sup>24</sup>.

Following the same rationale, a higher ED (8J/cm<sup>2</sup>) was applied with therapeutic intention for patients of the control group who only received LPLT when they achieved OM Grade 4 or 12 cm of ulcerous area. This unprecedented strategy was successful since

patients recovered in 6 days counting from the start of the laser application, whereas in previous report<sup>8</sup> this recovery took 16 days. Of note, the faster recovery from OM in our study cannot be attributed to neutrophil recover, since the medium score of neutrophils in this group was 94 mm<sup>3</sup>.

In contrast to the scores selected in previous randomized trials, here both WHO' scale and OMAS were used. The WHO's scale was selected due to its easy handling, straight forward applicability and previous validation, while the OMAS was picked because it measures and quantifies both ulcers and erythema. This is an important characteristic because some patients mention difficulty in swallowing due to pain in oropharyngeal even without presenting lesions on the oral cavity, what may mask the evaluation of WHO's scale or any other scale that mix signs and symptoms. The fact that both WHO's scale and OMAS showed a strong difference between LPLT and control groups in favor of the experimental arm coupled to the correlation between the two scales strengthen our data. Previous studies<sup>8,10</sup> did not evaluate the quantity and extension of the areas attacked by ulcers, but in the current study 7 patients from the laser group who presented ulcers had it in smaller proportion than the 17 patients from the control group. This fact confirms the methodological option for the use of OMAS. Theses results support OMAS as an instrument capable to portray the clinic condition of the patient and suggest that WHO and OMAS scale may be complementary in the evaluation of OM. It is worth to point out that none of the previous randomized studies used OMAS. Barach<sup>8</sup> used the modified Oral Mucositis Index Scale (OMI) and the Eastern Cooperative Oncology Group Oral Toxicity Scale (ECOG) and Cowen<sup>10</sup> used the scale published from Walsh<sup>25</sup>.

Although it was expected a delay in the beginning of OM in patients from laser group, it was not observed a significant statistical difference between laser and control group, what deserves further investigation. Further no significant statistical difference was seen between the laser and control group related to the total time with OM. It should be highlighted that control group patients who reached OM Grade 4 or a total area of OM of 12 cm, started to receive the therapeutic laser with 8J/cm<sup>2</sup> on the ulcerous areas. This crossover may have masked a true difference between the groups. In contrast to our trial Cowen<sup>10</sup> reported an increase in the time of mucositis Grade 0 and Grade 1 in the patients that received laser (17 days) compared to the control group (14 days). Here the comparison of the whole time of

OM between the groups did not show significant statistical difference, probably due to the above mentioned crossover.

Previous studies using VAS for pain report both a decrease in pain and in the use of morphine in patients receiving preventive laser<sup>8,10</sup>. In contrast to that, no difference between the two arms was noted in our series. It is important to emphasize that the initial idea was to use this scale to evaluate pain in the oral cavity. However it was observed that the pain started and was predominant in the oropharyngeal triggering the start of narcotics. Since these events preceded the onset of ulcers in the oral cavity, patients were already taking morphine when the oral lesions appeared and this may have been an important bias in the analysis of the VAS. Further it was observed that morphine provides a better pain control for oral cavity than for oropharyngeal lesions. These observations are unprecedented and should be taken into account and confirmed in future studies. Moreover no statistical difference was seen between Grade 3 and Grade 4, what probably reflects both the difficulty in the evaluation of the symptoms (subjective) and the administration of narcotics that masks the pain. This observation shocks the observation mentioned by Sonis<sup>26</sup>.

The presence of OM associated to the infection increases the hospital confinement time of the patient submitted to HSCT in 5 days increasing hospital costs<sup>26</sup>. The negative results of the blood cultures for *Streptococcus* in 38 patients indicate that the mouth environment adaptation method used was effective. The method adopted objectified supplying an eventual alteration in the quality and quantity of saliva, using a toothpaste with the lactoperoxidase enzyme, lysozyme and glucoseoxidase and the lactoferrin protein, bactericides, which in normal conditions are produced by the salivary glands, in association with the solution for mouthfuls containing chlorhexidine 0.12%, without ethanol. This approach is in harmony with Karthaus<sup>7</sup> who reported a positivity of *Streptococcus viridans* in blood cultures in 70% of the patients with severe OM; with Barker<sup>17</sup> and Meurman<sup>27</sup>, who referred to a possibility of a reduction of the quantity and quality of the saliva. Chlorhexidine (0.12%) has been regarded as a powerful antibacterial<sup>19,20,21</sup>.

It should not be neglected that our study was not blinded to one of the evaluators. This may have been balanced by the three additional evaluators who were not aware to which treatment arm the patients had been allocated and mainly by the fact that the method

adopted for evaluator's calibration proved to be efficient, since there was a concordance of 81.7% among them. Another caveat of the present study is the fact that it does not address if the high efficacy of the strategy adopted was due to the higher energy density, the length of application or the combination of both. In addition, this study did not assess a potential impact of LPLT on GvHD, days on antibiotics, use of total parenteral nutrition, and hospital confinement time. Additional studies with these endpoints as well as quality of life and cost-effectiveness analysis are warranted and should be pursued in future studies with preventive LPLT. Other endpoints that deserve further studies is survival. Although a marginal difference was observed in our study favoring the LPLT, these data should be analyzed with caution since our trial was not powered to detect these differences and confounding factors may be hampering the analyses.

In conclusion, our results indicate that the upfront use of LPLT in HSCT patients is a powerful instrument in reducing the incidence of OM and is now standard in our center.

Table 1

## Conditioning Regimens

Conditioning	Dose	Period	Laser Group	Control Group
Cyclophosphamide Carmustine Ethoposide	1800 mg/m <sup>2</sup> /day 450 mg/m <sup>2</sup> /day 2400 mg/m <sup>2</sup>	D -6 and D -3 D -2 D -7 ( 34 hours)	5	5
Cyclophosphamide TBI Anti-Thymocyte Globuline	60 mg/Kg/day 22 Gy 12/12 hours 15 mg/Kg/day	D -3 and D -2 D -7 to D -5 D -5 to D -4	3	5
Cyclophosphamide Bussulfan	60 mg/Kg/day 4 mg/Kg/Day	D -3 and D -2 D -7 to D-4	11	9

TBI- total body irradiation

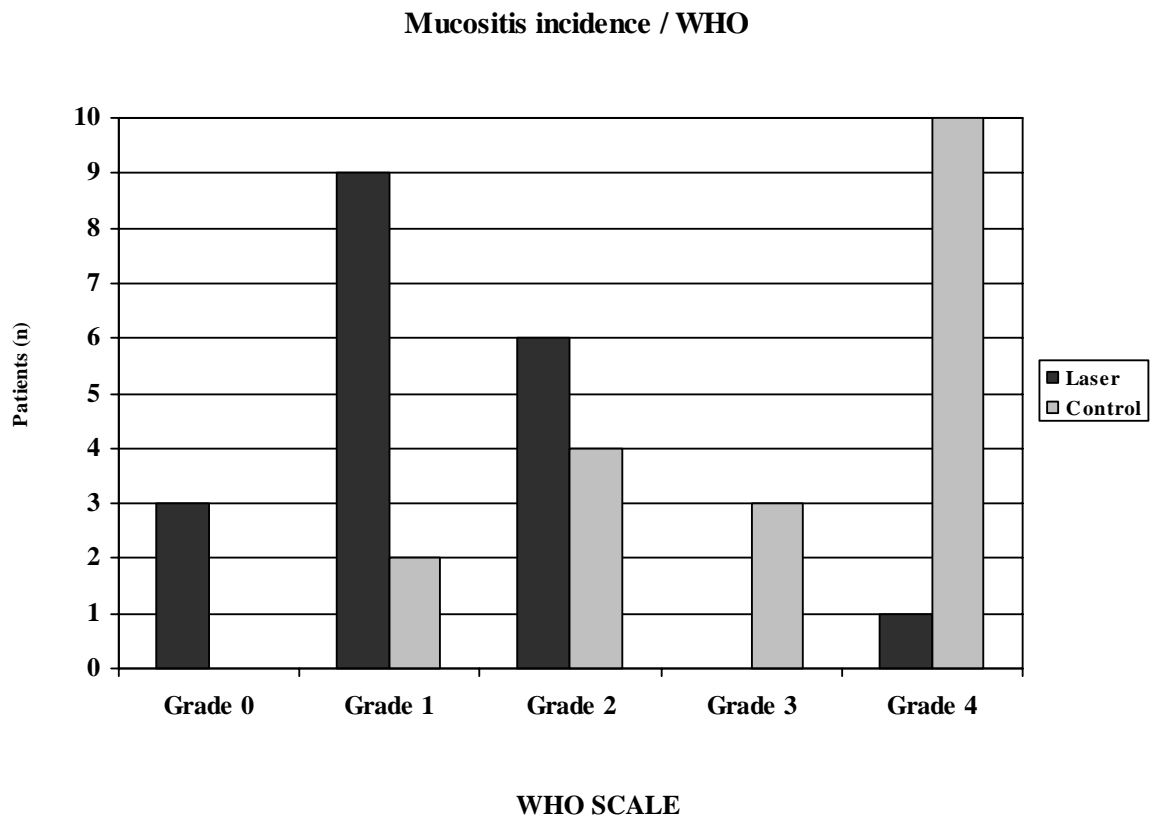
**Table 2**

Patients characteristics

Patients characteristics	Laser Group	Control Group
<b>Age</b>		
Average	36.5	36.8
Median	38.0	36.0
<b>Gender</b>		
Male	12	11
Female	07	08
<b>HSCT</b>		
Related Allogeneic	11	09
Related Allogeneic with TBI	00	02
Unrelated Allogeneic with TBI	01	03
Unrelated Allogeneic umbilical cord blood cells with TBI	02	00
Autologous	05	05
<b>Diagnostic</b>		
Chronic myeloblastic leukemia	08	08
Acute myeloblastic leukemia	03	03
Hodgkin's Lymphoma	06	02
Non- Hodgkin's Lymphoma	01	03
Acute lymphoblastic leukemia	01	00
Myelodisplasic Syndrome	00	03

TBI- total body irradiation

Figure 1



p<0.001

---

Figure 2

Kaplan – Meier mucositis free- survival

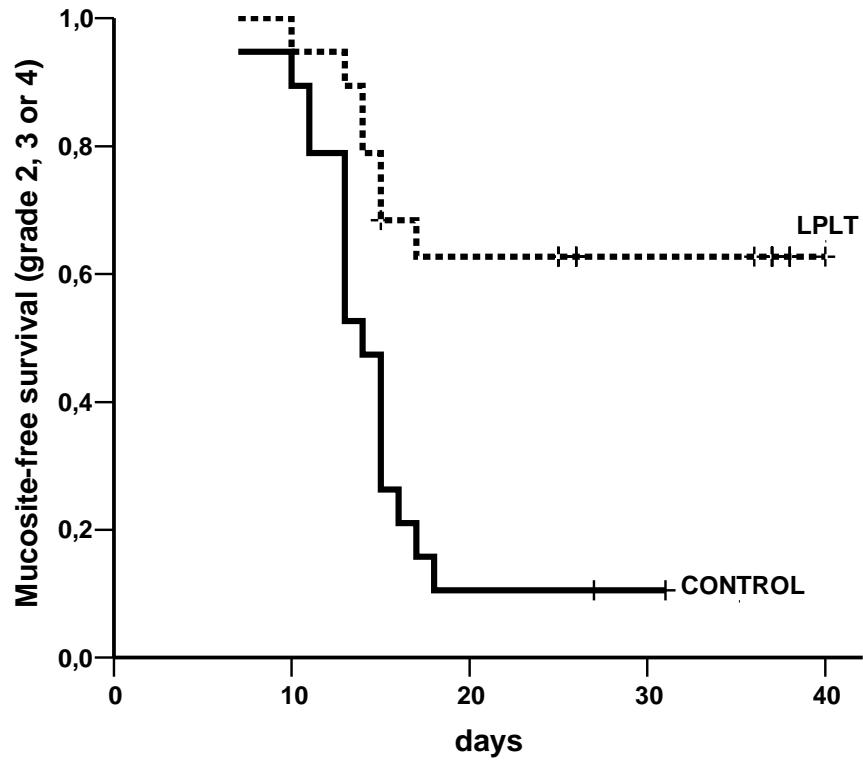


Figure 3

Kaplan-Meier mucositis free- survival(Grade 3- 4)

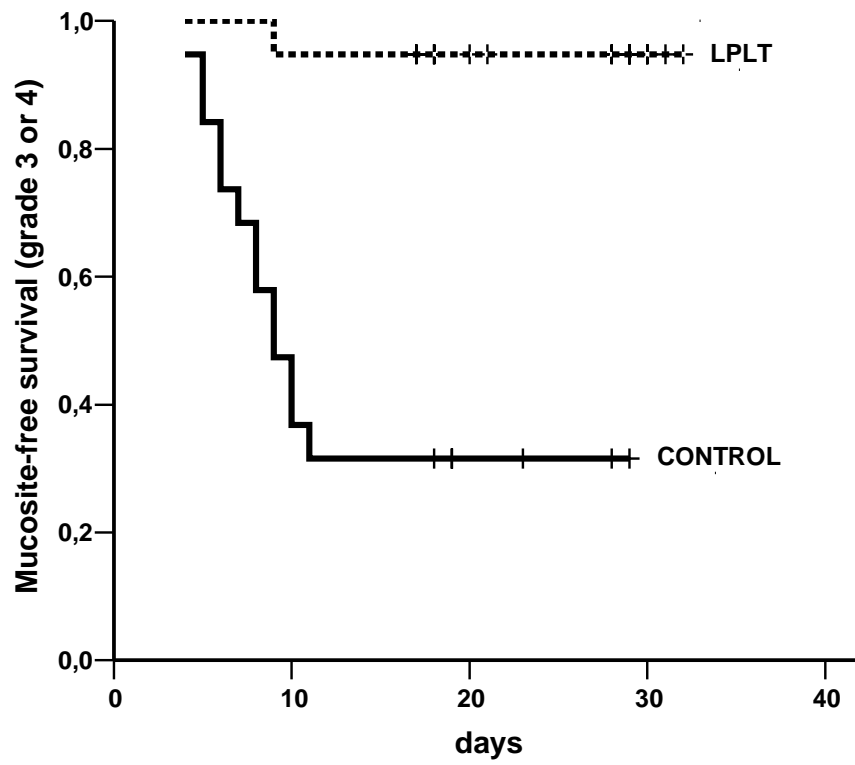
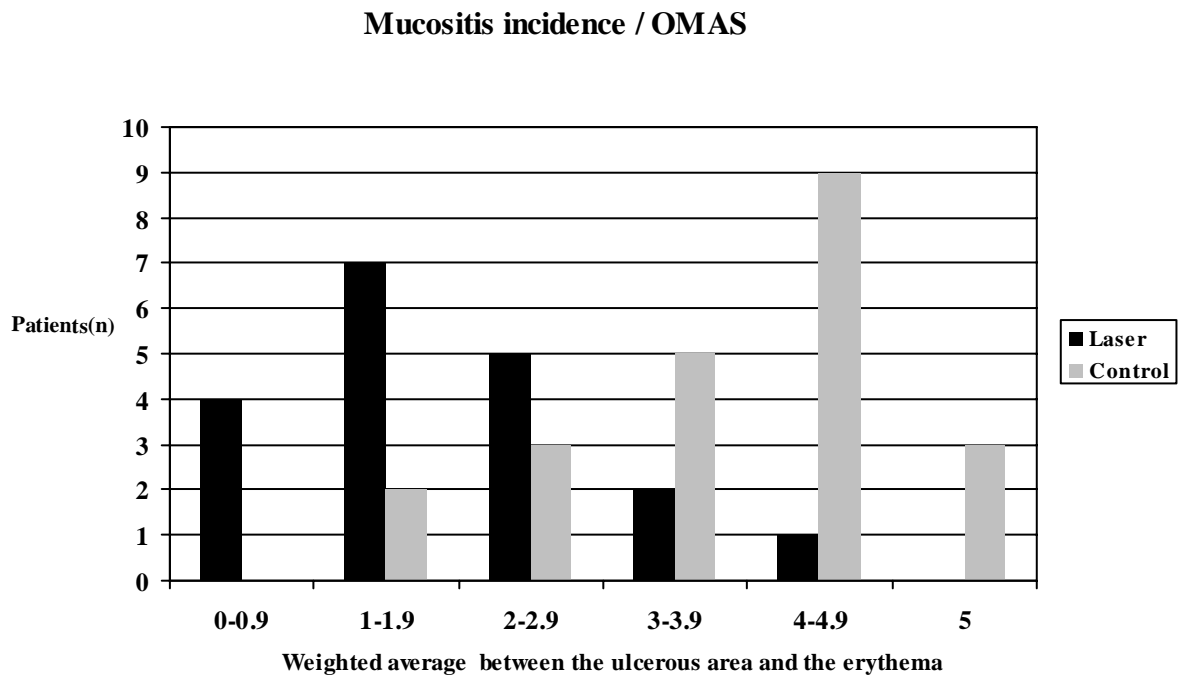


Figure 4



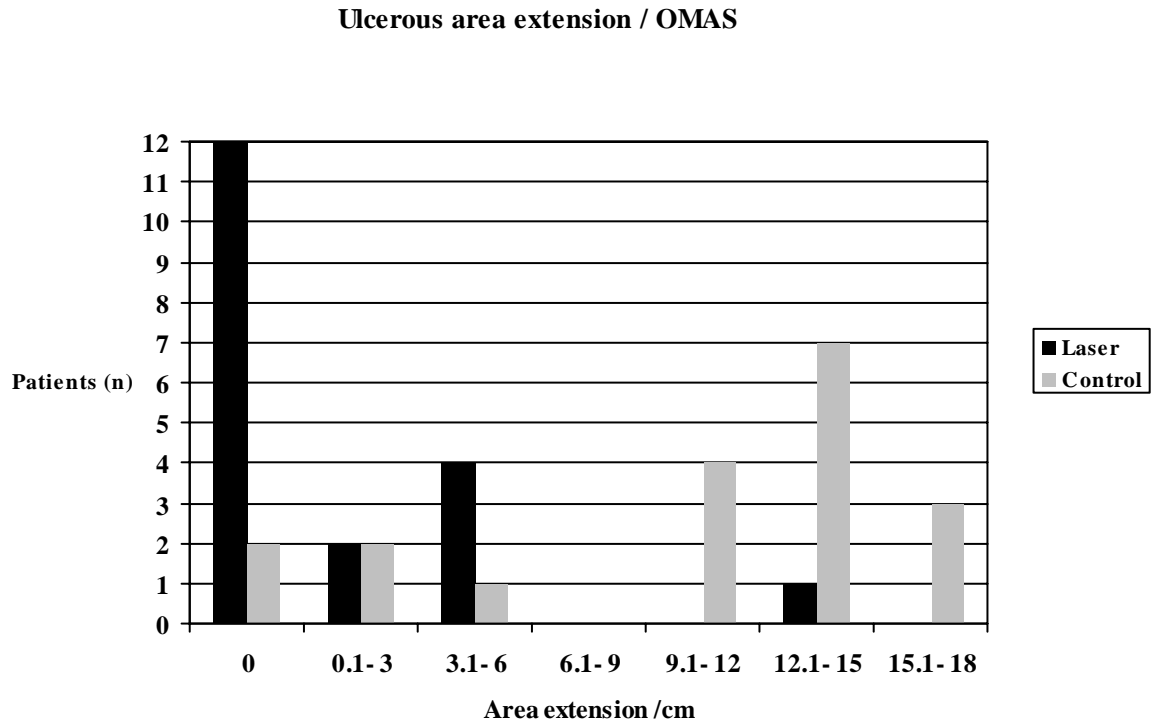
p= 0.007

---

(WA=2.5 x  $[(\sum u_i : 3 \times Nu) + (\sum e_i : 2 \times Ne)]$ , in which  $\sum u_i$ = sum of the ulcerous area, Nu= number of ulcerous areas,  $\sum e_i$ = sum of eritema's intensity and Ne= number of areas with erythema's.

Figure 5

Ulcerous area extension



---

P= 0.003

## References

1. Stiff P. Mucositis associated with stem cell transplantation: current status and innovative approaches to management. *Bone Marrow Transplant*. 2001; 27 (Suppl. 2): S3–S11.
2. Spijkervet FKL, Sonis ST. New frontiers in the management of chemotherapy–induced mucositis. *Curr Opin Oncol*. 1998; 10 (Suppl I): S23–S27.
3. Sonis ST, Oster G, Fuchs H et al. Oral mucositis and the clinical and economic outcomes of hematopoietic stem-cell transplantation. *J Clin Oncol*. 2001; 19(8): 2201–2205.
4. Sonis ST. The pathobiology of mucositis. *Nature Reviews Cancer*. 2004; 4: 277-284.
5. Rubenstein EB, Peterson DE, Schubert MM, et al. Clinical practice guidelines for the prevention and treatment of cancer therapy-induced oral gastrointestinal mucositis. *Cancer*. 2004; 100(9): 2026-2046.
6. Köstler WJ, Hejna M, Wenzel C, Zielinski CC. Oral mucositis complicating chemotherapy and / or radiotherapy: options for prevention and treatment. *CA Cancer Journal of Clinicians*. 2001; 51(5): 290-315.
7. Karthaus M, Rosenthal C, Ganser A. Prophylaxis and treatment of chemo- and radiotherapy-induced oral mucositis- are there new strategies? *Bone Marrow Transplantation*. 1999; 24:1095-1108.
8. Barasch A, Peterson DE, Tanzer JM, et al. Helium-neon laser effects on conditioning-induced oral mucositis in bone marrow transplantation patients. *Cancer*. 1995; 76(12): 2550-2556.
9. Ciais G, Namer M, Schneider M, et al. La laserthérapie dans la prévention et le traitement des mucites liées à la chimiothérapie anticancéreuse. *Bull Cancer*. 1992; 79: 183-191.
10. Cowen D, Tardieu C; Schubert M, et al. Low energy helium-neon laser in the prevention of oral mucositis in patients undergoing bone marrow transplantat: results of a double blind randomized trial. *Int. J. Radiation Oncology Biolo. Phys*. 1997; 38(4): 697-703.
11. Bensadoun RJ, Franquin JC, Ciais G, et al. Low- energy He/Ne laser in the prevention of radiation-induced mucositis: a multicenter phase III randomized study in patients with head and neck cancer. *Support Care Cancer*. 1999; 7: 244-52.
12. Migliorati C, Massumoto C, Eduardo FP, et al. Low-energy laser therapy in oral mucositis. *The Journal of Oral Laser Applications*. 2001; 1: 97-101.

13. Karu T, Pyatibrat LV, Kalendo GS. Photobiological modulation of cell attachment via cytochrome c oxidase. *Photochemical and Photobiological Sci.* 2004; 3: 211-216.
14. Karu T. Cellular mechanism of low power laser therapy: new questions. In: Simunovic, Z. *Lasers in medicine and dentistry: basic science and up-to-date clinical application of low-energy level laser therapy- LLLT.* Rijeka-Croá: Vitagraf (ed); 2003: 79-100.
15. Zhang Y, Song S, Fong CC, Tsang CH, Yang Z, Yang M. cDNA microarray analysis of gene expression profiles in human fibroblast cells irradiated with red light. *The Journal of Investigative Dermatology.* 2003;120(5): 849-857.
16. Woodruff LD, Bounkeo JM, Brannon WM, et al. The efficacy of laser therapy in wound repair: a meta analysis of the literature. *Photomedicine and Laser Surgery.* 2004; 22(3): 241-247.
17. Barker GJ. Current practices in the oral management of the patient undergoing chemotherapy or bone marrow transplantation. *Support Care Cancer.* 1999; 7:17- 20.
18. National Cancer Institute. Oral complications of chemotherapy and head and neck radiations. [http://www.cancer.gov/cancertopics/pdq/supportivecare/oral\\_complications](http://www.cancer.gov/cancertopics/pdq/supportivecare/oral_complications). Accessed April 6, 2005.
19. Addy M. Anti-sépticos na terapia periodontal. In: Lindhe J. *Tratado de periodontia clínica e implantodontia oral.* Rio de Janeiro, RJ: Guanabara Koogan S.A (ed); 1999: 332-349.
20. Epstein JB, McBride BC, Stevenson-Moore P, Merilees H, Spinelli J. The efficacy of chlorhexidine gel in reduction of *Streptococcus mutans* and *Lactobacillus* species in patients treated with radiation therapy. *Oral Surgery, Oral Medicine, Oral Pathology.* 1991; 71(2): 172-178.
21. Ferretti GA, Hansen IA, Whittenburg K, Brown AT, Lillich TT, Ash RC. Therapeutic use of chlorhexidine in bone marrow transplant patients: Case studies. *Oral Surgery, Oral medicine, Oral Pathology.* 1987(6); 63: 683-687.
22. Parulekar W, Mackenzie R, Bjarnason G, Jordan RCK. Scoring oral mucositis. *Oral Oncology.* 1998; 34: 63-71.
23. Sonis ST, Eilers JP, Epstein JB, et al. Validation of a new scoring system for the assessment of clinical trial research of oral mucositis induced by radiation or chemotherapy. *Cancer.* 1999; 85(10): 2103-2113.
24. Cruz LB, Ribeiro AS, Rech A, Rosa LGN, Castro CG Jr, Brunetto AL. Influence of low-energy laser in the prevention of oral mucositis in children with cancer receiving chemotherapy. *Pediatric Blood Cancer.* Prepublished on July 21, 2006, as DOI 10.102/abc. 20943.

25. Walsh LJ, Hill G, Seymour G, Roberts A. A scoring system for the quantitative evaluation of oral mucositis during bone marrow transplantation. *Special Care in Dentistry*. 1990; 10(6): 190-195.
26. Sonis ST. Mucositis as a biological process: a new hypothesis for the development of chemotherapy-induced stomatotoxicity. *Oral Oncology*. 1998; 34: 39-43.
27. Meurman JH, Pyrhönen S, Teerenhovi L, Lindqvist C. Oral sources of septicaemia in patients with malignances. *Oral Pathology*. 1997; 33(6): 389-397.