Enhancement of Surgical Vaginal Mucosal Wound Healing Using He-Ne Laser of 632.8nm with Topical Application of Tolidine Blue

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Abstract

Purpose of the study: This study aimed to prove the effect of low laser level therapy using He-Ne laser 1mw with topical application of tolidine blue dye as photosensitizer in promotion and enhancement of wound healing of vaginal mucosal layer.

Design: A twenty five women with vaginal mucosal wound of different causes were selected and divided into five equal groups, group one as control group, group two the wound exposed to laser and antibiotic after suturing .Group three the wound exposed to laser after painting with tolidine blue plus antibiotic, group four the wound exposed to tolidine blue and antibiotic only, group five expose d to tolidine blue paint plus laser therapy without antibiotics.

Results: Those patients who were exposed to laser and tolidine blue show significant reduction in the level of pain and rapid healing process that occur with in six days, this result was equal in both group III & group V, while group IV shows no difference in pain and tenderness in relation to control group. Group II show wound healing after 8 days but pain and tenderness show changes after the second exposure to laser.

Conclusion: LLLT can be used to enhance of mucosal wound healing of the vagina, to induce analgesic effects & prevent infection. The addition of photosensitizer (tolidine blue) leads to dramatic analgesic effects and more rapid healing.

Key word: low level laser therapy (LLLT), tolidine blue, vaginal wound

Introduction

In an extension of early experiments on mice in 1973, Mester and his group embarked on a series of animal studies, the results of which were published during the next 2 year [1]. All showed a photobiostimulation effect on the rate of tissue repair in various experimentally induced wounds as a result of low power He-Ne laser irradiation, and as a result of successful, clinical trails on small numbers of patients suffering from chronic unhealed wounds and sores of various etiologies which had previously been found to be unresponsive to other treatments, as a result of this pioneering work, laser therapy clinic was quickly established at Budapest. This type of therapy then extended mainly to Eastern Europe, China & the Soviet Union. In USA, the acceptance of LLLT, started in the early 1980s. [1] Laser therapy is a form of phototherapy which involves the application of monochromatic light over biological tissue to elicit a biomodulative effect within that tissue.

The main indications of photobiostimulation are:

- Wound healing.
- Soft tissue injuries, like muscle tears, hematoma and tendenopathies.
- Pain relief.
- Arthritic conditions of various aetiologies like small joint of the hands or feet. [2]

The interactions of the laser beam with biological tissue depend on the optical properties of the lased tissue.

The primary determinant of laser action is the absorption coefficient of the tissue, The scattering coefficient are additional factors that determine the absorption and penetration of laser energy. [3]

Low-intensity laser therapy can be used for pain relief.

A range of studies have indicated the apparent potential of low intensity laser radiation central in significantly altering the neurotransmitters of peripheral nervous system. [1, 3]

This therapy works on the principle of inducing a biological response through energy transfer, in that the photonic energy delivered into the tissue by the laser, modulates the biological process within that tissue, and those within the biological system of which that tissue is a part [2].

The Arndt-Schultz law of bio modulation infers that low dosage of photonic energy will stimulate that biological process, and higher dosage will inhibit them [4].

Haina in1981, Mashiko in 1983, Lievons in 1985, Abergel in 1987, Zarkovic in 1991, Fuad in 1993, Nazih in 2000 ,all of them said that low power laser therapy enhance granulation tissue formation [4,5,6,7,8,9,10]. Laakkso in 1994 said that 5 J/cm² is the
upper limit of therapeutic window & 8-12 J/cm² causing bio inhibition [11]. Inoue in 1989 suggested that LLLT has the potential to interfere with the operation of the human immune system in vitro [12]. Lutz Wilden in 2000 said that the importance of electromagnetic radiation of characteristics ranges of wave lengths on the mitochondrial energy transfer [13]. Walker in 1983, found elevation in the level of 5-hydroxyl indolacetic acid in those with pain relief after laser treatment [14]. Zakovic, Lombard, Lupyery, they suggested that laser can significantly alter serotonin metabolism, and increase the level of acetyl cholinesterase activity after exposure to He-Ne laser [15].

Patient, material & Methods:

A twenty-five multiparous with previous history of vaginal wounds, were selected so that those patients had a previous experience with such wounds; the duration of wound healing, pain and tenderness and its complication. They were between 25 to 35 years of age.

The selected patients having vaginal wound of different causes, it could be post delivery vaginal wound, vaginal wound due to anteroposterior repair. The selected patients were divided into 5 equaled groups.

- **Group I:** control group:- their wound treated by suturing with 000 chromic catgut plus antibiotic (amoxicillin).

- **Group II:** patients treated with suturing + laser (He-Ne) + antibiotic (Amoxicillin).

- **Group III:** patients treated with suturing + laser + painting with tolidine blue + antibiotic (Amoxicillin).

- **Group IV:** suturing + tolidine blue + antibiotic.

- **Group V:** suturing + laser + tolidine blue + without antibiotics.

Those who were allergic to penicillin were excluded from the study.

Laser parameters

Using the aiming beam of CO2 surgical laser, model BLITZ 50 SV, as a medical laser

Laser:- He-Ne laser, wavelength (632.8 nm)

Power: 1 mW

Divergence = 2 mrad

Output beam diameter = 0.61 mm

Time of exposure: 120 second for each spot

Energy density = 0.9 J/cm²

Spot size = 4mm

Procedure:

The patient was put in lithotomic position, exposing the interoitus, a piece of cotton introduced into the vagina to prevent uterine discharge during the procedure.

A graduated woody spatula was used as a guide to have 4mm spot size diameter and also to separate between the anterior & posterior vaginal walls. Tolidin blue was painted on the wound by using swab stick.

The exposure to LLLT was done 24 hours. After suturing the wound post-operatively every other day for four times, assessment during therapy as follows:-

1. The pain & tenderness, in comparison to the previous patient’s experience. Using short form of MaGill pain questionnaire (MPQ-SF)

2. Oedema around the wound.

3. Healing of the wound.

Laser Safety:

This type of laser used in this study is belonging to class two, so no specific safety measures were needed.

Results:

The study was depending on comparison between four groups of patients with control group (group one).

The assessment depends on the degree of pain according to short form McGill pain questionnaire (MPQ-SF) and the degree of tenderness, the severity of oedema of the presence of exudation.

The control group shows that the patient persisted to have pain and tenderness lasting 10 days and more with persistence of oedema for six days.

In group II, pain and tenderness changed to a moderate after the first laser exposure and slight decrease in oedema with persistence of exudates. This picture changed dramatically after the second exposure to laser that pain become mild, oedema become slight, little exudates, except for tenderness which persists moderate, the tenderness became mild after 3rd exposure and disappeared after the fourth exposure.

Group IV showed the same changes as control group (group I).

In group 3 and 5, we used laser plus photo sensitizer (tolidine blue paint) but in group 5, we didn’t use antibiotics.

Both groups show marked changes from the first exposure that the pain and tenderness became mild, the same for oedema with slight exudates, then after the second exposure the picture changed dramatically that the patients felt no pain on sitting, walking with mild tenderness, without oedema and dryness of the wound which showed complete healing after third exposure i.e. on day 6.
Table 1 shows that from first exposure, group 3 and group 5 that exposed to LLLT with. Tolidine blue painting got dramatic changes in pain, tenderness and inflammation, in relation to other groups. This table 2 shows that pain in group 3 and group 5 disappear but the tenderness became mild and the wound became dry with disappearance of oedema.

Table 2: The results of LLLT with or without photo sensitizer two days after the second exposure to laser.

<table>
<thead>
<tr>
<th>Number of group</th>
<th>Pain</th>
<th>Tenderness</th>
<th>Oedema</th>
<th>Exudates</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group one</td>
<td>Moderate</td>
<td>Moderate</td>
<td>Moderate</td>
<td>Obvious</td>
</tr>
<tr>
<td>Group two</td>
<td>Mild</td>
<td>Moderate</td>
<td>Mild</td>
<td>Slight</td>
</tr>
<tr>
<td>Group three</td>
<td>No pain</td>
<td>Mild</td>
<td>Negative</td>
<td>Dry</td>
</tr>
<tr>
<td>Group four</td>
<td>Moderate</td>
<td>Moderate</td>
<td>Moderate</td>
<td>Obvious</td>
</tr>
<tr>
<td>Group five</td>
<td>No pain</td>
<td>Mild</td>
<td>Negative</td>
<td>Dry</td>
</tr>
</tbody>
</table>

This table 3 shows that pain and tenderness after 3rd exposure in group 3 and 5 disappear with complete healing of the wound This table 4 showed that group 2 and group 4 still have tenderness and pain in spite of 4th exposure.

Table 3: The results of each group two days after the 3rd dose

<table>
<thead>
<tr>
<th>Number of group</th>
<th>Pain</th>
<th>Tenderness</th>
<th>Oedema</th>
<th>Exudates</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group one</td>
<td>Moderate</td>
<td>Moderate</td>
<td>Moderate</td>
<td>Slight</td>
</tr>
<tr>
<td>Group two</td>
<td>Mild</td>
<td>Mild</td>
<td>Slight</td>
<td>Slight</td>
</tr>
<tr>
<td>Group three</td>
<td>Negative</td>
<td>Negative</td>
<td>Negative</td>
<td>Dry</td>
</tr>
<tr>
<td>Group four</td>
<td>Moderate</td>
<td>Moderate</td>
<td>Moderate</td>
<td>Slight</td>
</tr>
<tr>
<td>Group five</td>
<td>Negative</td>
<td>Negative</td>
<td>Negative</td>
<td>Dry</td>
</tr>
</tbody>
</table>

Table 4: The results of LLLT with or without photo sensitizer 2 days after the fourth exposure.

<table>
<thead>
<tr>
<th>Number of group</th>
<th>Pain</th>
<th>Tenderness</th>
<th>Oedema</th>
<th>Exudates</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group one</td>
<td>Mild</td>
<td>Mild</td>
<td>Mild</td>
<td>Negative</td>
</tr>
<tr>
<td>Group two</td>
<td>Negative</td>
<td>Mild</td>
<td>Negative</td>
<td>Dry</td>
</tr>
<tr>
<td>Group three</td>
<td>Negative</td>
<td>Negative</td>
<td>Negative</td>
<td>Negative</td>
</tr>
<tr>
<td>Group four</td>
<td>Mild</td>
<td>Mild</td>
<td>Mild</td>
<td>Negative</td>
</tr>
<tr>
<td>Group five</td>
<td>Negative</td>
<td>Negative</td>
<td>Negative</td>
<td>Negative</td>
</tr>
</tbody>
</table>
Discussion:

The vagina is a site of repetitive surgical wounds (episiotomies) and for that reason this study was designed to see the effects of LLLT on vaginal mucosal wounds, the pain and tenderness decreased to a mild degree from first exposure that the patient insisted to have such treatment mainly because of analgesic effects.

These rapid changes allowed the patients to return to their usual home work rapidly, sitting freely in comparison with their previous experience with a wound in such area.

In this study duration of healing of the wound was shortened after exposure to LLLT (within 8 days) and this goes with previous studies which were mentioned previously, but the addition of photo sensitizer on the wound made the process of healing faster(within 6 days), no infection good analgesic effect, these changes were more dramatic than those exposed to laser therapy alone.

The most acceptable explanation of the beneficial effects of LLLT is what Passerella said in 1988, he suggested that laser irradiation extend to the mitochondria and induce changes in cellular homoeostasis, which entails a whole cascade of reactions and propose that a number of the components of the respiratory chain (eg. Cytochrome oxidase, flavine dehydroenase). Absorb laser light at appropriate wave lengths.

The activation of electron transport chain in this way in turn results in enhanced synthesis of ATP. Laser also affect hydrogen level in the cell, which is coupled with an increase in ATP, causing activation of other membrane ion carriers such as sodium an potassium and altering the flow of calcium between mitochondria and cytoplasm. [17, 18]

The variation of such parameters is a necessary component in the control of proliferative activity of the cells and the changes in ion concentration further affect cell metabolism and developments by influencing the cyclic nucleotide level which is involved in events leading to initiation of DNA synthesis. [16,17]

Regarding pain, changes in the level of serotonin and acetylcholine play a role in analgesic effects that explained by Walker in 1983. He said that 5-hydroxy indoleacetic acid (5-HIAA), a metabolite of 5-HT, excreted in high level in those patients experiencing pain relief as a result of laser treatment. [14]

Zarkovic 1989, Lombard’s 1990 also suggested that laser has the potential, at least under some circumstances, to significantly alter serotonin metabolism. [15]

Lupy 1986 confirmed the role of acetylcholine in this process, as he saw an increase in the level of acetyl cholinesterase activity after exposure to He-Ne laser therapy. [16]

The addition of photo sensitizer at the same time with LLLT made the result more obvious and clear. It seems that tolidine blue makes the tissue more sensitive to He-Ne laser so that produce the laser effects rapidly.

The group exposed to photo sensitizer plus LLLT without antibiotics showed no sign of infection and it seems that the addition of tolidine blue to the wound may cause suppression of bacterial growth which goes with previous observation by Wilson. He used a combination of low power laser light in the presence of a photosensitizer, using He-Ne laser of power out of 7.3 mw emitting light with a beam diameter of 1.3 mm at a wave length of 632.8 nm , the photosensitizer was tolidine blue 0 which has an absorption maximum at 632nm . Significant number of bacteria was killed in supragingival plagues samples. Wilson again in 1993 used a photosensitizer and LLLT to kill methicillin– resistance bacteria. [19,20]

The explanation of this bacterial growth inhibition may due to release of free radicals and then super oxide anion, hydrogen dioxide or a transfer of energy to oxygen molecules in which excited singlet oxygen which is very reactive, leading to cellular oxidation and necrosis and because the photo sensitizer is used as a paint on the surface of the wound it acts only at that area causing inhibition of bacterial growth. [19]

Conclusions

1-Low level laser therapy is a method that can be used in promotion of healing in gynecological and obstetrical wounds.

2-Low level laser therapy with a photo sensitizer like tolidine blue is a good method to enhance healing of vaginal mucosal wound, and to induce analgesia.

3- LLLT plus photo sensitizer prevent infection without the need to give systemic antibiotics.

4-The advantages of local photo sensitizer are; easy application, no toxic side effect from systemic applications and can be easily removed.

Suggestions

1-This method of therapy can be used in gynecology and obstetrics like in treatment of episiotomy for rapid healing.

2-We need to look for laser with different wave lengths and to use other photo sensitizers to have more flexibility.

3-One can do a study to see the effect of this method in treatment of infected wounds or ulcers.

4-To see the effect of LLLT on wound healing in different age groups, on different sites and with different wavelengths.
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