ORIGINAL ARTICLE

The Roll of Low Level Laser in diarrhea treatment

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ABSTRACT

Background: Escherichia coli (E. coli) bacteria normally live in the intestines of healthy people and animals. Most varieties of E. coli are harmless or cause relatively brief diarrhea.
Aim: To find the relationship between dose, and duration of laser beam with diarrhea patients.
Methods: *E. coli* species were taken from stool patients, and then it was cultured and incubated for 24 hours in Petri dishes, and coulter them in 24 tube of nutrition agar. 4 groups A, B, C, and D, from each Petri dish we take a culture and growth in nitric broth and then irradiated for different the same time (15 minutes) and for different number of times.
Results: Group A was considered as control, the intensity of laser beam was changed from 1.310 to 1.522 while the number of the bacterial species were decreased from 400, 300, 200, 100, 60, and 90 to groups A, B, C, D, E, and F respectively. **Conclusions**: the irradiation of these species with laser decrease the bacterial strain activity and this affected with respect to duration times, and with respects to time interval between each irradiation.
Keywords: *E. coli* species, diarrhea treatment, low level laser, irradiation

INTRODUCTION

Escherichia coli (abbreviated as *E. coli*) are bacteria that found in the environment, foods, and intestines of people and animals, they are a large and diverse group of bacteria. Although most strains of *E. coli* are harmless, others can make infected people sick and may cause diseases, and one them cause diarrhea, while others cause urinary tract infections, respiratory illness and pneumonia, as well as other illnesses. Escherichia coli typically live in the intestines of humans and animals and helps keep our guts healthy[1].

The types of *E. coli* that cause the majority of harmful infections in the U.S. produce a toxin called Shiga, and are appropriately called Shiga-toxin-producing *E. coli* (STEC). The CDC estimates that 265,000 Americans are infected with STEC per year, resulting in about 3,600 hospitalizations and 30 deaths[2].

Enter toxigenic *E. coli* (ETEC) is one of the leading causes of "traveler's diarrhea," which is often contracted when travelers from developed regions visit less-developed regions, according to Emory University[2]. The CDC estimates that anywhere from 30 to 70% of travelers may be affected, it depends on the time of year and destination, with areas such as Latin America, Africa and Asia having the highest risk of travelers developing ETEC[3].

Most common methods of administration of LLL radiation include lasers such as ruby (694nm), Ar (488 and 514nm), He-Ne (632.8nm), Krypton (521, 530, 568, and 647nm), Ga-Al-As (805 or 650nm), and Ga-As (904 nm)[4]. The reason why the technique is termed low level is that the optimum levels of energy density delivered are low and it is not comparable to other forms of laser therapy as practiced for ablation, cutting, and thermal tissue coagulation[5,6].

The first law of photobiology explains that for a low power visible light to have any effect on a living biological system, the photons must be absorbed by electronic absorption bands belonging to some molecular photo-acceptors, which are called chromophores[6,7]. The effective tissue penetration of light at 650 nm to 1200 nm is maximized. The absorption and scattering of light in tissue are both much higher in the blue region of the spectrum than the red, because the main tissue chromophores (hemoglobin and melanin) have high absorption bands at shorter wavelengths and tissue scattering of light is higher at shorter wavelengths. Water strongly absorbs infrared light at wavelengths greater than 1100 nm. Therefore, the use of LLLT in animals and patients almost exclusively utilizes red and near-infrared light (600-1100nm)[8-10].

But a few particularly nasty strains, such as E. coli O157:H7, can cause severe abdominal cramps, bloody diarrhea and vomiting. The use of low level laser to reduce pain, inflammation and edema, to promote wound, deeper tissues and nerves healing, and to prevent tissue damage has been known for almost forty years since the invention of lasers. Mitochondria are thought to be a likely site for the initial effects of light, leading to increased ATP production, modulation of reactive oxygen species, and induction of transcription factors. These effect in turn lead to increased cell proliferation & migration (particularly by fibroblasts)[11-12].

This study was designed and carried out to find the relationship between dose, and duration of laser beam and how it affect the bacteria that cause diarrhea.

MATERIAL &METHOD

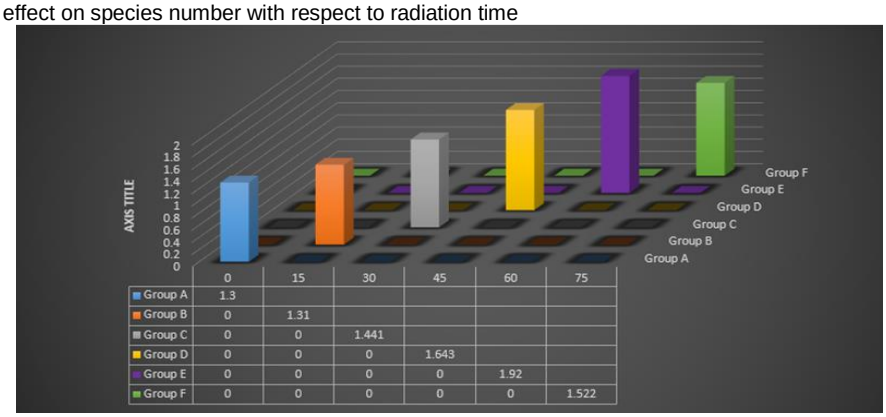
Ecolie species were taken from stool patients cultured and incubated for 24h in Petri dishes, and coulter them in 24 tube of nutrition agar, divided into 6 groups A (the control group), B, C, E, and F, and then irradiated as follow:
Group A: which contains 8 dishes each, cultured E-coli on Macungie agar.
Group B: irradiated directly with laser for 15 min continuously once daily.
Group C: irradiated with same laser for 15 min twice daily twice daily with time interval 20 min.
Group D: irradiated by the same laser for 15 min, three times a day with time interval 20 min.
Group E: irradiated by the same laser for 15 min four times a day, with time interval 20 min.

Group F: irradiated by the same laser for 15 min five times a day, with time interval 20 min.
All the incubated for 6hours and then taken for counting in spectrophotometer.

RESULTS AND DISCUSSIONS

The outcome results shows that there are a decreasing in the numbers of the species with respect to the duration of irradiation and also depends on the time interval between each irradiation with keeping the irradiated time fixed in order to get precise study for the effect of each group as shown in figures 1, 2 and 3.

Figure 1: Deference in effect on species number with respect to radiation time



	0	15	30	45	60	75
Group A	1.3					
Group B	0	1.31				
Group C	0	0	1.441			
Group D	0	0	0	1.643		
Group E	0	0	0	0	1.92	
Group F	0	0	0	0	0	1.522

Figure 2: Effect of laser on bacterial species numbers as an indicator of decreasing the intensity of the out com light when used spectrophotometer

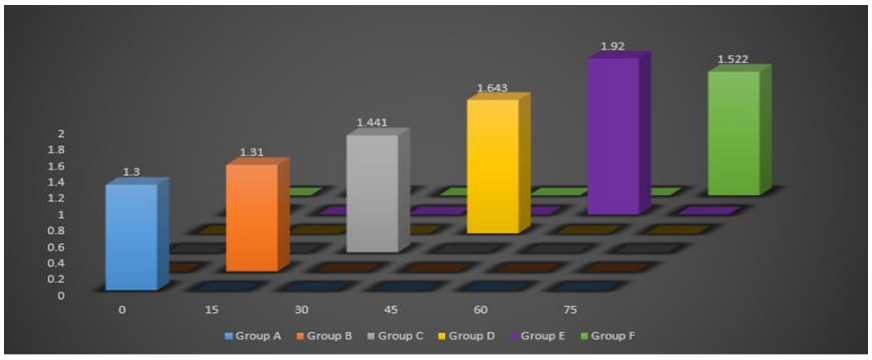
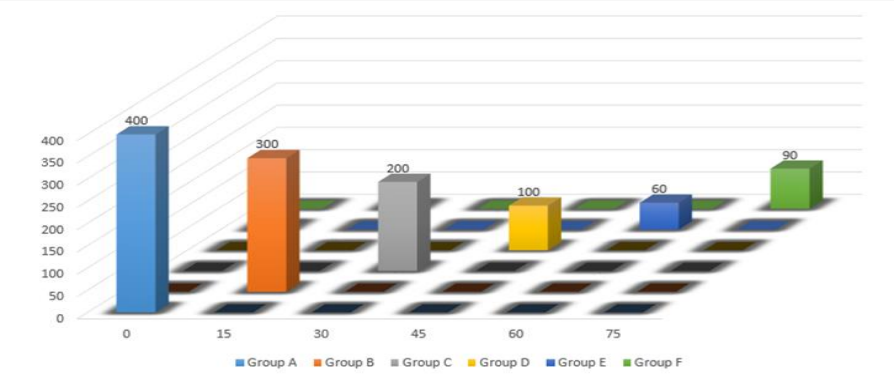


Figure 3: Defiance in bacterial species according to the deference in the time of radiation electric microscope



The decreasing of the intensity of the light that come out from the spectrophotometer was an indicator of decreasing the number of bacteria in the broth that under irradiation, also in order to let our work more precise we used another technique (using a mesh lens of electronic microscope) the counting of the species also decreased as figures 1, 2, and 3.

In group F the same result we get when we compared the other groups with respect to the control. This decreasing because of mitochondria, Cytochrome c oxidase (Cox) which represent as a multicomponent membrane protein which facilitate the transfer of electrons from water soluble cytochromec oxidase to oxygen. It is a terminal enzyme of the electron transport chain and plays a vital role in the bioenergetics of a cell[8]. Cox is the primary photo acceptor for the red-NIR range in cells because

absorption spectra obtained for Cox in different oxidation states was found to be very similar to the action spectra for biological responses to light[11].

The absorption of photons by Cox leads to electronically excited states, and consequently can lead to quickening of electron transfer reactions more electron transport necessarily causes increased production of ATP the light induced increase in ATP synthesis and increased proton gradient Cox, plays a vital role in the activation of the diverse biological cascade observed subsequently to laser irradiation[12].

The activity of cytochrome c oxidase is inhibited by nitric oxide (NO) .This inhibition can be explained by a direct competition between NO and O2 for the reduced binuclear center CuB/a3 of cytochrome c oxidase, and is reversible [(10, 11)]. It was proposed that laser irradiation could reverse this inhibition by photodissociating NO from its binding sites[12,13]. Because this coordinate binding is much weaker than a covalent bond, this dissociation is possible by LLL this effect was explained the decreasing of the bacteria after laser irradiation (Fig. 1, 2, 3). The dissociation of NO from Cox increases the respiration rate[13,14]. Light can indeed reverse the inhibition caused by NO binding to cytochrome oxidase, both in isolated mitochondria and in whole cells .LLL can also protect cells against NO-induced cell death[14].

Reactive Oxygen Species (ROS) and Gene Transcription LLLT was reported to produce a shift in overall cell redox potential in the direction of greater oxidation) and increased ROS generation and cell redox activity have been reported[10,14]..

In this show that the irradiation of these species may be decrease them activity with respect to duration times, also with respects to time interval between each irradiation. In the same time when we increased the number of irradiation, the activity of the cell increased also. It has been proposed that the redox state of a cell regulates cellular signaling pathways that control gene expression. Modulation of the cellular redox state can activate or inhibit signaling pathways. Several regulation pathways are mediated through the cellular redox state. Changes in redox state induce the activation of numerous intracellular signaling pathways, such as nucleic acid synthesis, protein synthesis, enzyme activation and cell cycle progression.

These cytosolic responses may induce transcriptional changes. Several transcription factors have been recognized to regulate by changes in cellular redox state. Based on the ability of LLLT to modulate cellular metabolism and alter the transcription factors responsible for gene expression, it has been found to alter gene expression.

Irradiation of LLL stimulates cell growth directly through regulation of the expression of genes related to cell proliferation and indirectly through regulation of the expression of genes related to cell migration and remodeling, DNA synthesis and repair, ion channel and membrane potential, and cell metabolism. All these factors explain the increasing again of the number of bacteria after increasing the number of irradiation by laser.

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