Iraqi Laser Scientists Journal (ILSJ)/ www.ilsj-online.org
Vol .1, Issue 3; Pp; 1 - 6 , 2019

Original article

Impact of laser (Nd:YVO4 Crystals,532nm) radiation on white blood cells

Zahra, J.M.A. Al-Timimi
Laser Physics-College of Science for Women, Babylon University, Iraq

Abstract

Lasers have been flexible to utilization in multiple fields, one of these significant applicability in varied ranges in medicine for the advancement of health. Various models of lasers have been developed and efforts have been performed to incorporate it based on the precarious impressions they pose. The current study analyzed whether the low-level energy laser can be induced the leukocytes cell number count in human whole blood in vitro.

Hundred five ml samples of healthy and not-healthy blood obtained respectively, then the samples were additionally divided into four groups with 25 samples each, the four groups received different energy density. Each sample was then subdivided into 2 parts with 2.5 ml each ; was to labour as a control and the other as a test.

In examining the influence of the laser on the count numbers influenced by human leukocytes , the (Nd: YVO4 Crystals-diode-pumped, λ=532nm) laser was applied, in the first experimental group, the laser energy density was 0.5J/cm^2 at exposure duration of 1Sec , 1.5J/cm^2 at exposure duration of 3Sec, 3J/cm^2 at exposure duration of 6Sec and 5J/cm^2 at exposure duration of 10Sec for the second, third and fourth experimental groups. The examinations were assessing specifically the counts of leukocytes were conducted in an automated hematology analyzer. The leukocytes count in all the experimental groups did not vary and almost remained unchanged even after irradiation of the blood samples with different exposure duration.

The laser radiation on the blood samples appeared to have no significant influence on the leucocytes count number. The finding showed that the effect of the irradiation on these cells was less, nonetheless, the statistical analysis of the mean of the leukocytes cell count number of all groups noted the increase wasn't significant ($p>0.05$) , this intimates the advantageous role of a laser on numerous varieties of leukocytes,

Laser irradiation (Nd: YVO4 crystals-diode-pumped, λ=532nm) significantly improved the rheological characteristic by the protection and safeguard of leucocytes count essentially, these outcomes confirmed that there was no changing in the hemolysis markers and serum bilirubin concentration, the immune cell element wasn't considerably influenced by this laser . This proposes the importance of low-energy laser's in its biomedical application for various conditions.

Key words: Laser, White Blood Cells, Hematology

To cite this article: Zahra, J.M.A. Al-Timimi; Impact of laser (Nd:YVO4 Crystals,532nm) radiation on white blood cells ; Iraqi Laser Scientists Journal. Vol .1, Issue 3; Pp; 1 - 6 , 2019.

Introduction

Lasers have gained immense use due to the optimization of the wavelength for human tissue

interactions , (1–3) , other application of the laser irradiation include in aesthetic surgery for the repair of damaged or wrinkled skin through an effective and non-ablative therapy where intense pulsed laser is applied, wound healing and prevention of restenosis following coronary angioplasty, (4–6).

Biomedical & therapeutic administration of a laser is based on light - tissue interactions which, supervising to absorption by cellular elements such as a mitochondria cytochromes & endogenous chromospheres' which, give rise to the phenomena like a fluorescence, chemical reactions and thermal effects, (6–8). Among these interactions, thermal effects have proved to be the most versatile, (9). These laser therapies often involve red blood cells due to the high optical absorbance of hemoglobin, (10&11). Red blood cells (RBCs) are positioned in numerous organs in great concentrations. Consequently, the damaging influence of these radiations happens at the level of a cell pending the hemoglobin is placed in it. These physiological appearances arouse other rules in the cell-like enzyme inactivation , change of the metabolic rate & coagulation between other structural reforms in the cell, (6,12,13).

The low-level energy lasers in up-to-date have been adopted in the non-invasive therapy of musculoskeletal & cutaneous complications so have gained developed treatment in wound healing, nerve repair, and pain management amongst other clinical applications, (14–16). In diabetic conditions, where wound healing is compromised, (5,17), low-energy intensity lasers have shown beneficial effects of healing impaired wounds.

The extraordinarily indispensable in resisting disease progress are Leucocytes considering they serve as the fundamental immune system protection for the body by utilizing multiple tools such as lysis & phagocytosis, (18–20).There have been extensive studies by Stadler *et. Al.*, (21) , on the effects of laser irradiation on the peripheral blood lymphocytes and RBCs in vitro . The study used red light (660nm) and demonstrated a marked increase in the populations of lymphocytes irradiated in the whole blood, (21).

Laser effect on these immune cells have been postulated to occur via the electron transport chain which causes an increase in adenosine triphosphate (ATP) production with subsequent increases in the cell metabolism which causes the photo- bio stimulation effect associate with laser irradiation, (22).

The **(**Nd: YVO4 Crystals-diode-pumped, λ=532nm laser being used in this study falls under the class IIIb laser with a power output of 100 mW. Class IIIb lasers are usually low power diode laser used for biostimulation. The low-energy laser has been advanced as an efficient instrument in the therapy of the host of neurological, vascular and soft tissue diseases, (9,23).

Lasers utilising photochemical influences, which don't destroy or break up tissue, (24). Photochemical -tissue interactions are substantial in therapy medications, alike photodynamic therapy, (21,25). Nevertheless, there has been an insufficient insight into the beneficial & damaging threshold levels of laser's radiations on leukocytes. Moreover, the safe limits of radiation and exposure duration have not been clearly ascertained. This study was to investigate whether the low laser energy density induces the WBCs number count in the whole human blood (in vitro).

Materials and Methods

In achieving our objectives, we subjected human blood cells to different lasers doses at different durations and determined the laser dose effects by comparing to set non-irradiated control blood samples. A study was organized following the guidelines of (National Medical Research) in Pulau Pinang General Hospital & sustained by (National Institute of Health) in Malaysia & Committee of Medical Research and Ethics. The collection of a hundred samples

of blood, 5 ml each, was done from healthy & non-healthy adults both female & male. The samples were handled with an anti-coagulant (EDTA), and then the samples were further subdivided into four tubes for laser irradiation. Each one of the samples was halved so that, one of the 2.5 ml samples was to labour as the control &the other as a test.

In guaranteeing the validity of the data, data was collected from 100 samples from patients of different races (Malaysians, Chinese, and Indians) sexes, age and of different health status. Most of the data collected from this study was from non-healthy samples since data was collected from a pathological laboratory. This was intended at realizing the objectives, which were to evaluate the influences of Nd: YVO4 crystals-diode-pumped, λ=532nm on the change of the rheological qualities of blood.Reliability of the data was assured by the size of each experimental group (n = 25) that was subjected to different laser doses.

Before the counting was produced, samples were left for 30 minutes at room temperature. Nd: YVO4 Crystals-diode-pumped, λ=532nm laser has been used as a radiation device with a power of 100mW. It comprises a powerful 808nm diode laser that pumps an Nd: YVO4 laser crystal, which in change outputs 1064nm. This quickly is doubled inside a non-linear KTP crystal, appearing in green light at the half-wavelength of 532nm.At the aperture, the diameter of the laser beam was (1.5mm) with a divergence (<1.5mRad), and the spot-size was ($0.2cm^2$).

The laser doses and their durations in seconds are as shown in Table1. In the 100 samples, they were clustered in four experimental groups that were to receive different lasers doses. In the first experimental group, the laser fluence was $0.5J/cm^2$ at exposure duration of 1 Sec followed by $1.5J/cm^2$ at an exposure duration of 3Sec for the second group, $3J/cm^2$ at an exposure duration of 6 Sec and $5J/cm^2$ at exposure duration of 10 Sec, for the third &fourth experimental groups.

An Automated Hematology analyzer (Sysmex XE -2100TM) machine was applied to measure values of irradiated and non-irradiated (control) blood samples. This machine employs four methods in its hematological analysis. The RF/DC detection method is able to spot alteration in the sizes and densities of blood cells through detection of direct-current resistance and radio-frequency resistance respectively. The hydrodynamic focusing also referred as DC detection assists in ameliorating the accuracy of blood count and consequently its reproducibility. Physiological and chemical features of the cells are captured by the flow cytometry method, which employs the semiconductor laser.

Table1 : Laser doses used in the experiment

Fluence (J/Cm^2)	Exposure Time (Seconds)
0.5	1
1.5	3
3.0	6
5.0	10

Data Analysis

Means and standard deviations of the different experimental groups were done in the SPSS statistical software. The Significant differences accepted if the P value was less than (0.05) and rejected if the value was exceeded.

Results

In the irradiated groups, a small rapid systemic increase in WBCs counts during the Exposure Time of 3Sec & 10Sec. In the control group, the count number of the WBCs settled approximately constant, as pointed in figure 1, the levels of these WBCs didn't change and almost settled unchanged alike after

radiation with a different laser dose

The laser irradiation on blood samples seemed to have no significant effect on the leucocytes count. However statistical analysis of the mean number of WBCs of the groups showed the increase was not significant ($p>0.05$), as shown in the Table 2 below .This indicates to the beneficial role of 532nm solid state laser on various types of white blood cells in preserve and protect the cell from death or dissolution.

Table 2: Effects of laser (λ=532nm) irradiation on WBCs

Exposure Time	Energy Density (J/cm^2)	Mean Number of WBCs (10^3/µL)		p value n* = 25
		Control Groups	Irradiated Groups	
1	0.5	11.99± 5.63	11.62± 5.47	>0.05
3	1.5	10.69± 7.33	11.30± 7.14	> 0.05
6	3.0	10.44± 5.31	10.37± 5.23	> 0.05
10	5.0	9.83± 4.51	10.07± 4.24	> 0.05
*n = number of blood samples				

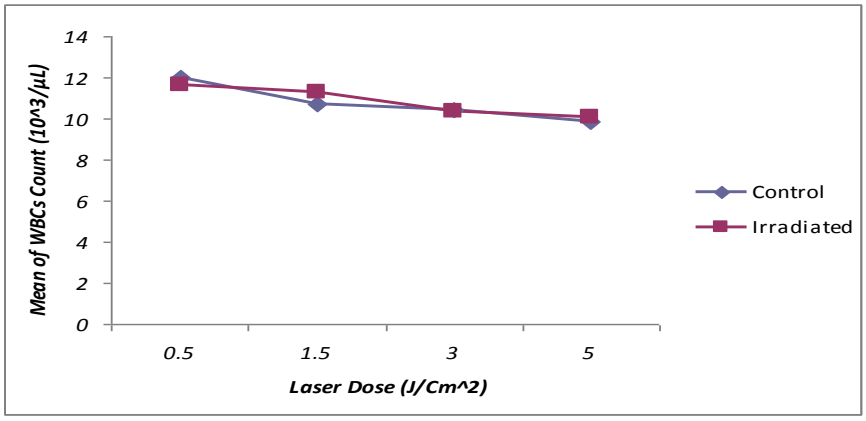

Figure 1: Graph showing the effects of laser (λ=532nm) irradiation on WBCs count.

Discussion

The laser effect on these immune cells has been postulated to occur via the electron transport chain which causes an increase in adenosine triphosphate (ATP) production with subsequent increases in the cell metabolism which causes the photo- bio stimulation influence connected with laser radiation, (26).

A shorter duration of the laser beam is a signed increase in optical absorbance of the Hgb, which decreases induced heating, (23). This is because of the aggregation of Hgb (Hgb coagulation) which increases the density of Hgb and consequently its optical absorbance, (27,28). This may suggest that the 532nm laser irradiation was not able to cause hemolysis and hence a subsequent reduction in the populations of WBCs.

Cell lysis events in the dispersion of the hemoglobin, which is an influential laser absorbing element of RBCs & a succeeding decrease in the absorption efficacy of the blood ,(29).

Biological temperature for the denaturation of Oxy-Hgb complex is in most cases followed by haemoglobin denaturation, (30). These outcomes were faithful with the exception in hemolysis markers and serum bilirubin concentration, (25).

Conclusions

The short wavelength such as, high-energy ionizing irradiation causes the ionization of molecules in an indiscriminate manner with the far-infrared heating biological tissues, hence, only the near-infrared

& visible wavelengths which, in range of (400-10,600nm) are agreeable to the clinical utilization due to their discriminate characteristics.

When the laser dose time is increased there is no further change which is observed in WBCs. The explanation for this observation may be because at a longer time of laser dose exposure, there is the loss of the thermal loss in the irradiated blood samples. Also at laser dose for shorter duration, there is a marked increase in the optical absorbance of the Hgb, which minimizes laser-induced heating.

Bibliography

1. Siedhoff, MT, Wheeler SB, Rutstein, SE, et al. (2015); Laparoscopic hysterectomy with morcellation vs abdominal, hysterectomy for presumed fibroid tumors in premenopausal women: A decision analysis. Am. J. Obst. Gynecol. 212: 591.e1-591.e8.
2. Zahra, AT., (2017); Letter to the editor clinical evaluation of scalpel Er: YAG laser 2940 nm and conventional surgery incisions wound after oral soft tissue biopsy. Bangladesh Med. Res. Coun. Bull. 43: 149.
3. AL- Timimi Zahra , (2014); Investigating the Effects of Green Laser Irradiation on Red Blood Cells: Green Laser Blood Therapy. Int. J. Appl. Res. Stud. 3: 1–5.
4. Mustafa, FH., Jaafar, MS , Ismail, AH, et. al. (2011); Influence of hair color on photodynamic dose activation in PDT for scalp diseases, IFMBE Proceedings, 315–319.
5. Zahra, A-T., (2018); A Comparative Study of Determination the Spectral Characteristics of Serum Total Protein Among Laser System and Spectrophotometric: Advantage and Limitation of Suggested Methods. Curr. Anal. Chem. 14: 1–8.
6. Karu, T., (1989); Laser bio stimulation: A photo biological phenomenon. J. Photochem. Photobio.l B. Biol. 3: 638.
7. Al-Timimi, Z., Mustafa, FH., (2018); Recognizing the effectiveness of the diode laser 850nm on stimulate the proliferation and viability of mice mesenchymal stem cells derived from bone. Iraqi J. Vet. Sci. 32: 285–290.
8. Al - Timimi, Z, Saleem, I., Alhabeel, M., (2019); Laser Dental Treatment Techniques, In: Dr. Sivapatham Sundaresan (Ed.), Oral Cancer, In tech. Open, 1–16.
9. Kreisler, M, Christoffers, AB, Al-Haj, H, et, al. (2002); Low level 809-nm diode laser-induced in vitro stimulation of the proliferation of human gingival fibroblasts. Lasers Surg. Med. 30: 365–369.
10. Suhaimi, M, Zahra, J,, (2010); Therapeutic Laser for Chronic Low Back Pain. Bangladesh J, Med, Sci. 8: 118–128.
11. Hamad, F., Jaafar, M., Hamid A., Omar, A., Timimi Zahra, Houssein, H., (2009); Influences of different low level laser power at wavelength 635 nm for two types of skin ; dark and light. Proc. 7th. IMT-GT UNINET 3rd. Int. PSU-UNS Conf. Biosci. Influ. 7: 130–135.
12. Medrado, AR a P, Pugliese, LS, Reis, SR a, et. al. (2003); Influence of low level laser therapy on wound healing and its biological action upon my fibroblasts. Lasers Surg. Med. 32: 239–244.
13. Morgan, J, Oseroff, AR. (2001); Mitochondria-based photodynamic anti-cancer therapy. Adv. Drug. Deli. Rev. 49: 71–86.
14. Zhang, XF, Lin, Y, Guo, W, et, al. (2014); Spectroscopic insights on imidazole substituted phthalocyanine photosensitizers: Fluorescence properties, triplet state and singlet oxygen generation. Spectrochim Acta A Mol. Biomol. Spectrosc. 133: 752–758.

15. Houssein, HAA, Jaafar, MS, Ali, Z, et, al. (2011); Study of hematocrit in relation with age and gender using low power helium - Neon laser irradiation, IFMBE Proceedings, 463–466.
16. AL-TimimiZahra, (2014); Biological Effects of Yellow Laser-Induced of Cell Survival: Structural DNA Damage Comparison is Undergoing Ultraviolet Radiation Photocoagulation. Int. J. Eng. Res. Gen. Sci. 2: 544–548.
17. Zahra A-TT (2014); Exploration of Additional Mechanical Phenomena of Laser-Tissue Interaction Contributes to the Damage and Elimination of the Small Blood Vessels. Int. J. Sci. Eng. Res. 5: 672–675.
18. Lam, E, Kato, N, Lawton, M., (2001); Programmed cell death, mitochondria and the plant hypersensitive response. Nature, 411: 848–853.
19. Conlan, MJ, Rapley, JW, Cobb, CM., (1996); Bio stimulation of wound healing by low-energy laser irradiation A review. J. Cline. Periodontol. 23: 492–496.
20. Cannon, B. (2004); Brown Adipose Tissue: Function and Physiological Significance. Physio. Rev. 84: 277–359.
21. Nouri, K., (2018); Lasers in dermatology and medicine: Dental and medical applications, Springer International Publishing.
22. A. J. Welch, Jorge, H. Torres, W-FC., (1989); Laser Physics and laser tissue interaction. Laser Phys. 61: 961–964.
23. Peplow, P V., Chung, TY, Baxter, GD., (2010); Laser photobiomodulation of proliferation of cells in culture: A review of human and animal studies. Photomed. Laser Surg. 28: S-3-S-40.
24. Saygun, I, Karacay, S, Serdar, M, et. al. (2008); Effects of laser irradiation on the release of basic fibroblast growth factor (bFGF), insulin like growth factor-1 (IGF-1), and receptor of IGF-1 (IGFBP3) from gingival fibroblasts. Lasers Med. Sci. 23: 211–215.
25. Roukoz, D, Skandri N, Chammas, C. (2014); Pain Relief of Aphthous Ulcers by Lasers : A Literature Review. Dent. News (Lon.) 21: 12–22.
26. Peat, FJ, Colbath, AC, Bentsen, LM, et. al. (2018); In Vitro Effects of High-Intensity Laser Photo bio modulation on Equine Bone Marrow-Derived Mesenchymal Stem Cell Viability and Cytokine Expression. Photomed. Laser Surg. 36: 83–91.
27. Van Nimwegen, SA, Kirpensteijn, J. (2007); Comparison of Nd:YAG surgical laser and remorgida bipolar electrosurgery forceps for canine laparoscopic ovarioctomy. Vet. Surg. 36: 533–540.
28. Eduardo, F. de P, Bueno, DF, de Freitas, PM, et. al. (2008); Stem cell proliferation under low intensity laser irradiation: A preliminary study. Lasers Surg. Med. 40: 433–438.
29. Houssein, HAAA, Jaafar, MS, Ali Z, et, al. (2012); Correlations between lymphocytes, mid-cell fractions and granulocytes with human blood characteristics using low power carbon dioxide laser radiation. Mod. Appl. Sci. 6: 27–33.
30. Mahmood, S, Abolhab R, Mohamed, M. (2015); The Effectiveness of Diode Laser 810 nm in the Removal of Oral Pyogenic Granuloma in Repetitive Pulsed Mode. Iraqi J. Med. Sci. 213: 137–142.