REVIEW ARTICLE

Effect of Low Level Laser Irradiation on White and Red Blood Cells after Different Storage Periods

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ABSTRACT

The aim of the present work was to investigate the effect of irritation with the He-Ne laser on the whole human blood cells. Samples were collected from healthy persons. Samples were divided into two groups: irradiation and control groups. irradiation groups received laser irradiation from He-Ne laser (30 mW) for 15 minutes. The results demonstrate that RBCs count start to decrease slightly after 6 hours of laser irradiation and significantly decreased after three weeks if irradiation followed by storage. Lymphocytes and T-cells counts showed sever dropping after 3 weeks of storage.

Keyword: Laser treatment, Blood storage, Low level Laser therapy, Blood cells.

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Conflict of interest: None

INTRODUCTION

Low intensity laser irradiation has widespread applications in different areas of medicine.[1] In hematology, a new method has been projected for the renewal of preserved Blood (i.e., increasing the ratio of young to old blood cells) using laser irradiation of stored blood. This method is supposed to be efficient, low-cost, non-invasive thus guarantee the absence of blood contamination. In this case, performing transfusions using the irradiated blood will better for the treatment of some hematological diseases.[2] A positive adjustment of some markers of the blood cells functional integrity has been recorded after laser irradiation, especially for the RBC parameters.[3,4] More research are required to understand the effects of low level laser (LLL) irradiation on these parameters.[5] Scientists continuously try to improve the transfusion process and the storage conditions of the blood, to minimize the transfusion risks. Important issues in blood transfusion that should be considered are the quality of preserved blood and the clearance of the transfused RBCs. During the storage period, Well-defined changes occur and are collectively called the storage lesion which would affect the quality of the blood, although, number of studies provide valuable evidence that the normal storage durations of blood do not have measurable adverse effects on the clinical outcomes in select transfused patient populations.[6] These changes involve the loss viability, changes in the mechanical and biochemical characteristics , and the oxidative damage.[7-9] Previous studies on intravenous irradiation reveled short-term effects of laser light on blood cells involving changes in erythrocyte membrane leading to the release of factors into the blood that appears to stimulate further changes, structural changes in plasma proteins, activation of complement and other components against foreign bodies, release of oxygen free radicals, decrease of platelet quantity and sometimes obstructing their functions, increased phagocytosis and many other effects.[5] Mostly, low level laser therapy utilizes He-Ne laser (wavelength of 632.8 nm). In this regard, He-Ne laser irradiation is thought to yield a protective effect on RBC membranes by stabilizing cell membrane thus reducing hypotonic hemolysis. Several erythrocytes (RBCs), hemoglobin quantity (HGB), and hematocrit (HCT) were strongly influenced by He-Ne laser action due to the strong absorption of hemoglobin in this wavelength.[2,4,5]

MATERIALS AND METHOD

Collection and Preparation of the Samples

Whole blood samples (3 mL) were collected from 50 healthy adults (mostly male), age 20–55 years. The blood aliquots placed in tubes containing an anticoagulant to avoid clotting. Before laser irradiation (zero time), a small volume of each sample (0.5 mL) was analyzed using hematology analyzer (Diagon D-cell5D) (Hungary). T-cells was measured after 6–8 hours of sampling. These measurements considered as a control or base line measurements.

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Irradiation Procedure Light

The remaining volumes were irradiated using a He-Ne laser (30mW) for 15 minutes. To measure T-cell percentage in blood samples before and after irradiation, *CyFlow® Cube 6* Flow Cytometry (Germany) with CD3 marker was used. After irradiation, blood samples were stored using a special refrigerator (Dairei refrigerator (Denmark) at -80°C in the laboratory of *Al-Amal National Hospital for Cancer Management* for 3 weeks. RBCs counts where measured after 6 hours, 1-week, 2 weeks, and 3 weeks of storage.

Statistical Analysis

SPSS program was used for statistical analysis of the results. Data were represented as mean ± SD. Statistical deference ≥ 0.001 considered significant.

RESULTS

The effects of low LLL irradiation on blood cells characteristics after storage are showed below. The RBC count was slightly decreased in samples tested after only 6 hours of exposure. RBC still slightly decreased in blood samples stored for two weeks, and strongly decreased after 3 weeks of storage as showed in Figure 1.

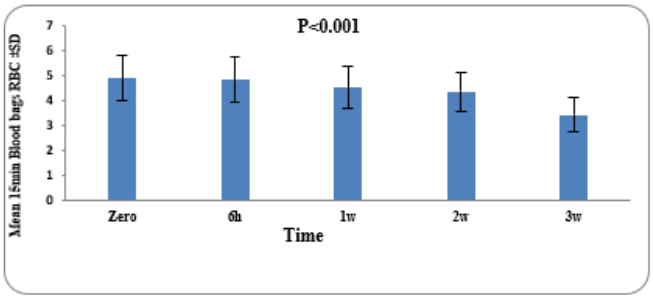

Figure 1: Effects of laser irradiation on RBC count after different blood storage periods

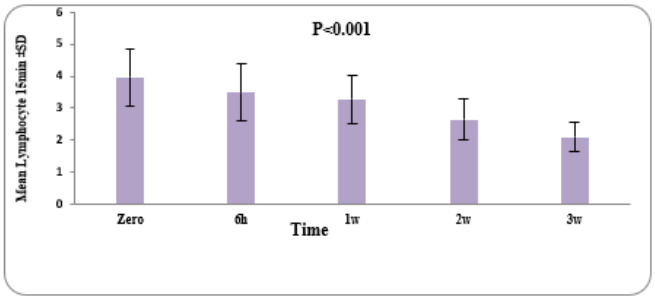

Figure 2: Effects of laser irradiation on lymphocytes count after different blood storage periods

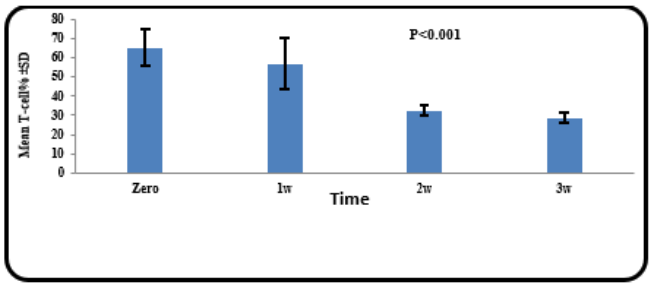

Figure 3: Effects of laser irradiation on T-cell count after different blood storage periods

Laser irradiation had an observed effect on lymphocytes when the results compared with the control samples. It was clear that after 3weekes of blood samples irradiation and storage, lymphocytes dropped to 50% of its original number before storage as shown in Figure 2.

The T-cell number was studied for blood samples which irradiated and stored for 1, 2, and 3 weeks by flow cytometry, as shown in Figure 3. T-cell numbers were significantly decreased up to 50% after 2 weeks of storage.

DISCUSSION

Low level laser (LLL) irradiation can cause many biological effects in living cells mostly via biochemical biomodulation rather than direct thermal effect. It is known that that red laser light affects blood at the molecular level. Laser light at 632.8 nm wavelength is selectively absorbed by hemoglobin. Under certain conditions, absorption of laser radiation by hemoglobin components leads to fractional photodissociation of the hemoglobin–ligand complexes, which in turn affect the oxygen affinity of hemoglobin.[3] Laser light also can affect RBC membrane protein structure leading to protein denaturation and cell lysis. The severity of these effects depends mainly on irradiation time and energy doses.[10] In the case if fresh blood irradiation, laser light increases lymphocyte count via improving cell proliferation and migration and prevention of cell apoptosis.[11] Laser light was found to cause the following effect when it interacts with blood cells: (1) increased activity of white blood cells; 2) stimulates redox activity in the cellular respiratory chain leading to cell activation; (3) enhanced cell proliferation and growth; (4) stimulation of protein, DNA, and RNA synthesis; and (5) activation of ATP production.[12,13] Previous study found that blood irradiation with 630 nm laser light caused significant decrease in WBC count, probably due to thermal injury caused by the irradiation or from initiation of primary, free radical reactions, while the decrease in RBC count may be due to changes in osmotic fragility and localized heating of the cells leading to cell rupture and hemoglobin release.[11,14]

CONCLUSIONS

The results of this study demonstrate that exposing blood samples to LLL (*in-vitro*) for 15 minutes has an observed effect on lymphocytes and T-cells more than its effect on RBC. These effects appear more clearly with increasing storage period namely after two weeks.

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