

Ain Shams University
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The Effect of Low Level Laser on Enzymes Involved in DNA Repair

Thesis submitted for the partial fulfillment of Doctoral Degree in Physics (Biophysics)

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\mathcal{T}_0 My grandmother \mathcal{T}_0 My parents \mathcal{T}_0 My brothers

You are the real owner of this Success

Abstract



Abstract

This Study aims to investigate the capability of activation of DNA repair enzymes involved in the biological cell that hosted a DNA damage via the exposure to Low Level Laser (LLL) at different fluences. The need for DNA repair enzymes activation or activation of DNA repair process becomes a serious demand after increasing genotoxic agents' sources and diseases caused by DNA damages. Low Level Laser (LLL) has been reported as an effective therapeutic tool in pain relief, wound healing, deeper tissues and nerves healing and bone regeneration. In addition, it could produce protection effect against DNA damages subsequent to ionizing radiation exposure and the proliferation in the biological cell. In this study, low level He-Ne laser was supposed as activator to the DNA repair process or DNA repair enzymes in human peripheral blood lymphocytes. These lymphocytes had DNA damages due to the previous exposure to different low doses of γ -radiation (2, 4, 6, 8, 10) Gy in Cs-137 unit. The effect of LLL was observed at different fluences (1, 2.5, 5) J/cm². Comet assay was used here to monitor the capability of LLL in removing the DNA damages produced by gamma radiation in the lymphocytes.

The measurement of DNA repair enzymes content or gene expression did not grow to be the effective routine in the judgment of enzymes activity. Comet assay measurements were used as indicator of DNA repair enzymes activity as function of their values. In this work, thecomet assay measurements of the samples pre-irradiated with ionizing radiation are compared to Cytochalasin-B Blocked Micronucleus (CBMN) at the same doses of y-radiation. The comparative study aims to validate

the comet assay procedures. Measured Comet assay parameters (tail DNA, tail length and tail moment) exhibit a significantly high correlation (p < 0.01) with the exposure dose. However, micronuclei (MNi) (CBMN parameter) show a p-value greater than 0.05. Thus, Comet assay is evidently more reliable in the estimation of the magnitude of DNA damage in peripheral blood lymphocytes due to exposure to gamma radiation.

On the second part, the comet assay measured parameters (tail DNA%, tail length and tail moment) are used to evaluate the contribution of LLL in activation of DNA repair enzymes by reducing DNA damages produced by different doses of gamma radiation at fluences of 1, 2.5 and 5 J/cm² individually. The observed results after immediate exposure to LLL reveal an irregular variation in the comet assay measured parameter at 1 J/cm² and 2.5 J/cm² at the applied Ionizing Radiation (IoR) doses. The issue seems to be different when observing the same cells after 5 hours from LLL exposure. A gradual reduction in comet assay parameters is observed as a function of LLL fluence for each applied IoR dose. Fluence of 5 J/cm² has the largest reduction in comet assay parameter. Thus, one may consider the LLL as activator to the DNA repair process where its capability is expressed as a function of LLL fluence.

This study put the LLL in a new vision as a therapeutic tool for DNA damages produced by IoR in biological cell. In addition It may be considered as an activator to Base Excision Repair (BER) enzymes which are the predominate pathway for IoR DNA damages. This study raises new questions that may be answered in future work about:

- (a) The ability of applying low level He-Ne laser at fluence 5 J/cm² as epidemiological procedure for workers in radiation filed or patient subject to radiotherapy.
- (b) The ability of applying low level He-Ne laser on cells that are deficient in DNA Repair enzymes.

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