E-ISSN: 2706-8927
P-ISSN: 2706-8919
www.allstudyjournal.com
IJAAS 2025; 7(3): 01-04
Received: 25-12-2024
Accepted: 26-01-2025

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Effects of laser irradiation and wide: Spectrum light on the growth of colon cancer cells

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DOI: https://www.doi.org/10.33545/27068919.2025.v7.i3a.1383

Abstract
This study sought to ascertain if wide-ranging light, when combined with laser treatment, may accelerate and stimulate cell growth. Background Information: Broad-spectrum light can partially or fully counteract the effects of laser light. A few experiments have been conducted to investigate the benefits and drawbacks of combining broad-ranging light and laser irradiation light and laser irradiation. Methods: 635nm lasers were used to irradiate colon cells at different doses (J/cm^2) in both light and dark. MTT (3-(4,5-Dimethylthiazole-2-yl)-2,5-diphenyl-2H-tetrazolium bromide) tests have been used to measure variations in cell growth. Results: The findings showed that doses of 3.7 and 5 J/cm^2 were sufficient to cause noticeable alterations in colon cells by low-level laser therapy (LLLT). According to the MTT experiment, different doses employing 635 nanometers in the illuminated medium were more successful than 635 nm in the dark and more active promoting cell-reproduction. Conclusion: The results demonstrate that, as compared to the dark, red wavelengths are the most efficient in maintaining cell viability in the light-medium.

Keywords: Laser irradiation, broad-spectrum light, colon cancer cells

1. Introduction
Light is classified as belonging to the electromagnetic energy spectrum, which encompasses a broad variety of wavelengths, from extremely long waves at the infrared end of the spectrum to very short waves at the ultraviolet end (Figure 1). Numerous researches have looked at how different wavelengths affect cell proliferation; one study looked at how different wavelengths affected fibroblasts and proposed that cold laser treatment in the visible and near-infrared spectrum is what stimulates cell respiration [1]. One study found that biological effects were stimulated by monochromatic incoherent light *in vitro*. When the grown cells are first exposed to a laser beam, they exhibit biological effects; however, these effects diminish to nearly zero when they are subjected to a wide-spectrum light [2]. Thus, it implied that the experiment involves more than just the excitation of polarization-sensitive chromophores, and that ambient light may influence the results at Vivo and Vitro, which explains some of the discrepancies in the results/effects reported in the studies [2]. *In vitro*, fibroblast growth was suppressed by 810 nm, whereas visible lasers induced the highest proliferation, according to research on the effect of wavelength on fibroblast proliferation [3]. The findings show that the response of cell growth to laser treatment was influenced by both wavelength and cell type [3]. Although many aspects of their mechanisms are unclear, visible and infrared irradiation have a wound-healing effect as a result of local light impact [4]. To evaluate cell viability, researchers [5] using the trypan blue exclusion test and the MTT colorimetric assay. The advantages and disadvantages of combining a laser beam with wide-spectrum light have not been thoroughly examined in studies. This study examined the potential benefits of broad-spectrum light by exposing colon cells to varying laser doses (635 nm). This study suggests that the presence of broad-spectrum light in the laser therapy room might aid in accelerating cell proliferation.

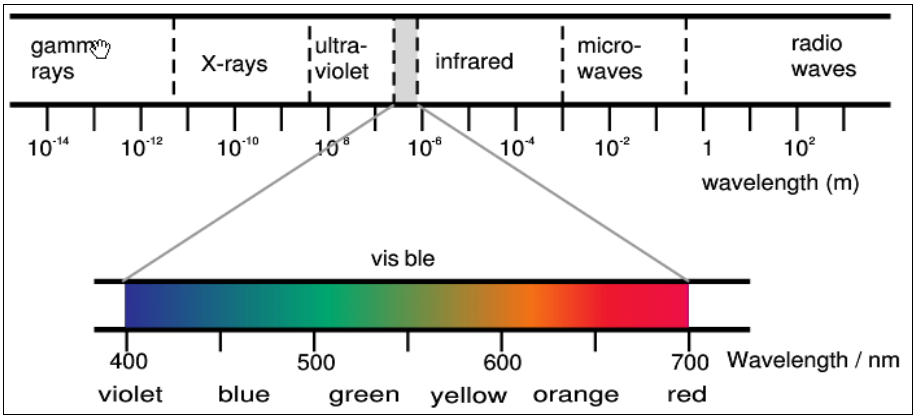

Fig 1: Visible light spectrum [6].

2. Materials and Procedures

2.1 Culture of cells

Cell culture is a method for cultivating human or animal cells in a controlled setting. Given a medium that includes growth hormones and nourishment, cell culture is one of the most popular techniques in biomedical studies across a wide range of academic fields since it may be used to induce cells to mature *in vitro* or in their own tissue under the correct conditions. In a lab culture, cells are paired with the Vivo and allowed to grow similarly to how they do in the body. 37 degrees Celsius was used to brood the cell cultures. Inverse microscopy was used to examine the cells [7]. These cells are being stimulated to grow in handy plastic tubes using the complicated media. When a primary culture is a subculture, a cell line or strains with a restricted or continual existence in culture are produced [8].

2.2 Laser irradiation

Colon cancer cells were exposed to visible laser radiation at wavelengths of 635 nm, and doses of 3.7 and 5 J/cm^2. To ascertain the impact of these individual dosages and irradiation circumstances on cells, three irradiation tests were conducted in triplicate, and the average of the results was calculated. To enable the cells to attach, they are incubated overnight after planting. Before being exposed to radiation, the dishes had been incubated at 37°C [4]. Each dosage is measured for one minute for 3.7 J/cm^2 and 1 minute and 21 seconds for 5 J/cm^2 utilizing a laser intensity of 62 mW/cm^2 at 635 nm. Additionally, while the laser irradiation, the cells were maintained at Vitro temperature to observe whether any notable alterations occurred in the cells.

2.3 MTT test

The MTT test attempts to calculate cell viability in a high-resolution manner, avoiding the need for repeated cell counts. Because mitochondrial succinate dehydrogenase can only degrade soluble tetrazolium salts into a deeper purple formazan crystal in active cells, the level of activation is a strong predictor of probable cell division. The MTT test is based on the assumption that the majority of living cells have steady mitochondrial function and that variations in the number of viable cells are directly proportional to changes in mitochondrial activity. The formazan concentration, indicated as optical density, is assessed with a plate reader calibrated to 570 nm to ascertain variations in cell viability [7].

3. Results and Discussion

The impact of LLLT on colon cancer cells. The average value is used to summarize the results of the three repetitions of the tests. The viability ratios at 3.7 and 5 J/cm^2 were different from the control. The therapy sets' average value reveals show difference that is larger than would be expected by chance. Relative to the untreated group, this experiment demonstrated an elevation in cell counts for the 3.7 and 5 J/cm² dosages of 635 nm light (Figure 2), signifying enhanced mitochondrial function and survivability of the irradiated cells. Cells may have recovered from the first shock of light irradiation. The MTT examination, which uses 635 nm in the dark, showed to improve in viability with the various dosages (Figure 3). implying that following the first shock of laser irradiation, the cells are stimulating their own function, growth reorganization, and vitality. The results also demonstrate that dosages administered at 635 nm are more effective than controls in the presence of light, but less effective than those administered at 635 nm in the absence of light. Consequently, the results showed that 635 nm was the most efficient wavelength in the lighted environment. Different cell types respond differently to radiation, and the total cell response may show the average value rather than the true value. This is important for cell analysis but has little therapeutic importance. According to Karu [7, 8], the wavelength, dosage, and intensity of the radiation as well as the circumstances of the cell culture determine the laser's real effect. Due to its biological limitations, LLLT cannot cause the growth of rapidly proliferating cells or the activation of every cellular function. A multitude of *in vitro* experiments are being performed, and the behavior exhibited or not exhibited in a test sample illustrates the effect of laser therapy on a particular individual cell [9]. Laser light stimulates cells that are not responding appropriately throughout the irradiation process. Therefore, laser irradiation will provide no effect and no therapeutic advantage, regardless of whether cells are fully functioning at the moment of irradiation or proliferate under ideal conditions. Research has shown that broad-spectrum light may partially or completely affect laser light [10]. Using a red laser (635 nm), the results of the MTT test demonstrate that, when compared to controls, cell exposure in the dark exhibits the improvement in cell counts. The results indicate that, as compared to when they are in a dark room, cells subjected to irradiation at various dosages (J/cm^2) in a light room utilizing 635 nm have the best enhanced viability. This can be interpreted as the white light acting as a catalyst

either by increasing the light reaching the cells and thus increasing its absorption, which led to an increase in mitochondrial activity and thus increasing stimulation, which is what was observed in this research. The results demonstrated the influence of the therapeutic environment and, while most of the results were good, the doses at 635 nm and 70 mW were more successful than those with light but less effective than those at 635 nm and 70 mW in the dark. Consequently, research results indicate that 635 nm works best in the light [11].

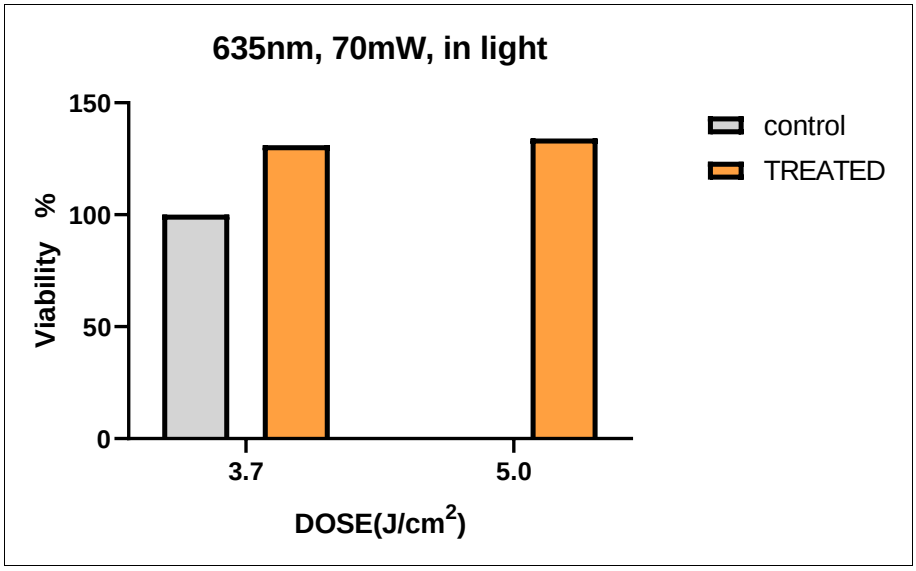

Fig 2: Illustrates how a 635 nm laser affects colon cancer cells when exposed to light.

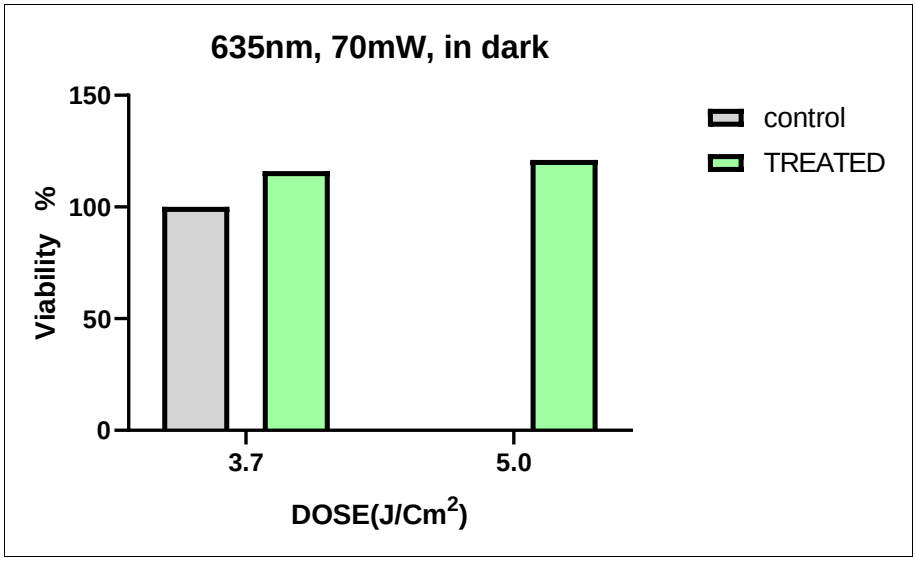

Fig 3: illustrates how a 635 nm laser affects colon cancer cells in the absence of light.

4. Conclusion

Irradiated cells using a wavelength of 635 nm in combination with wide-spectrum light combined an increase in survival, indicating that light may promote laser irradiation provided that the exposure duration is minimized to prevent undesirable thermal consequences. Using light at 635 nm enhanced the viability. In keeping with several studies that demonstrate that a medium dosage enhances cell development and functioning. These findings add credence to those of previous investigators who identified laser treatment as a very effective therapeutic method for tissue repair. Wide-spectrum light may have an impact on cell biological processes, as demonstrated by the results, which also indicated that the irradiation microenvironment influences the effects of laser irradiation. Despite claims that this light may lessen the effects of laser light completely or in part, our results show that using white light at a wavelength of 635 nm increases viability and has a stimulatory effect, suggesting that white light may have a potential therapeutic benefit. According to the results, it would be advantageous to use white light in combination with laser radiation to improve cell activation at a specific dose, wavelength, and exposure period.

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