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Original research article

Analysis of Laser Absorption Value Using LLLT-50 Device on Biological Samples with 650 nm Wavelengths

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ABSTRACT

The study aims to determine the absorbance coefficient value, with the light intensity observed before and after using Low-Level Laser Therapy-50 (LLLT-50) device on chicken and cow-meat tissues. The device has 20 mW, 25 mW and 50 mW output power and energy variations of 0.2J, 0.4J, 0.5J, 1.0J, 1.5J, and 2.0J. Samples of chicken meat with thicknesses of 0.65 mm, 0.85 mm, and 1.6 mm, as well as cow meat with thicknesses of 0.30mm, 0.60mm, and 0.85mm are measured using a spindle range. Each measurement is carried out 10 times to ensure the accuracy. The results show that chicken meat has a lower absorption coefficient than cow meat, due to lower myoglobin content and a simpler tissue structure. For chicken tissues with output power of 20 mW, 25 mW and 50 mW respectively, the LLLT-50 devices have absorption coefficients (μ) of -2.2, -2.17; and -2.9. However, the output capacity of the LLLT-50 devices with the same value on the cow meat tissue have μ of 1.2; 1.94; and 2.0. The findings support the development of non-invasive laser therapies, such as acupuncture lasers, by optimizing therapeutic parameters for better efficiency and safety. The research also provides insights into the interaction of laser light with biological tissues that are important for medical and scientific applications.

Keywords: Absorbance coefficient; Chicken-meat Tissue; Cow meat Tissue; LLLT-50; Laser 650 nm

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1. Introduction

Low-Level Laser Therapy (LLLT) is a method in the field of biomedics that can stimulate tissue healing, reduce inflammation, relieve pain without significant side effects, and be used as laser therapy acupuncture [1, 2]. LLLT was first observed by Endre Mester in 1967 when studies revealed that ruby laser exposure enhanced hair regrowth and wound repair in mice. Subsequent research over decades has optimized its medical applications through improved precision in wavelength selection and dosing protocols. In addition, LLLT is widely utilized for wound healing, pain management, and tissue regeneration due to its noninvasive nature and ability to stimulate cellular activity. Laser acupuncture is one of the therapies that combines the basic principles of traditional acupuncture with modern laser technology [3]. This acupuncture laser therapy uses low-level lasers to stimulate acupuncture points in the human body. In laser therapy acupuncture can replace the use of conventional needles. Such therapy aims to stimulate the flow of energy in the body and is able to improve various health conditions without causing pain due to use of needles [4, 5].

As a non-invasive kind of acupuncture therapy that can reduce pain, stiffness, and discomfort, acupuncture laser therapy is a recent breakthrough. Litscher (2018) found that the adenosine triphosphate (ATP) produced by the acupuncture laser stimulates cell metabolism and triggers the body's natural healing process by penetrating the cells with light photons [6]. In 2015, Hamblin et al [7], found that lasers can neutralize pain and enhance well-being; additionally, endorphins and other neurotransmitters can be released by acupuncture point laser activation. Supriyana and associates (2020), using these contemporary advancements, used "acupuncture points" in traditional therapy's preventative approach to treat certain diseases by connecting the skin's surface organs with therapeutic therapies [8, 9].

LLLT primarily targets mitochondrial cytochrome c oxidase, enhancing adenosine triphosphate (ATP) synthesis via the electron transport chain. This process reduces oxidative stress by lowering reactive oxygen species (ROS) and modulates intracellular signaling pathways, including NF-κB and Nrf2, which regulate inflammation and antioxidant responses. Secondary effects include increased angiogenesis, neurogenesis, and cytokine modulation (e.g., TGF-β, VEGF) [10, 11].

LLLT demonstrates diverse clinical applications across multiple medical fields. In pain management, LLLT effectively alleviates chronic and acute pain, particularly in osteoarthritis and musculoskeletal disorders. A 2020 meta-analysis of 22 randomized controlled trials (RCTs) reported significant pain reduction in knee osteoarthritis, with a mean difference of -2.1 cm on the visual analog scale, attributed to anti-inflammatory effects and endogenous opioid release [12]. For tissue repair and wound healing, LLLT accelerates closure by stimulating fibroblast proliferation and collagen synthesis, with a 2021 review highlighting its efficacy in diabetic ulcers, where 70% of studies noted enhanced healing rates [12]. Post-surgical applications further reduce edema and expedite recovery. In dermatology, LLLT is utilized for acne vulgaris, psoriasis, and androgenetic alopecia; a 2019 RCT involving 60 participants demonstrated an 85% improvement in mild-to-moderate acne after 12 sessions, leveraging red and near-infrared wavelengths to target sebaceous glands and reduce inflammation [12]. Emerging neurological applications include traumatic brain injury (TBI) and neurodegenerative diseases, with a 2020 review noting improved cognitive function in TBI patients through neurogenesis and mitochondrial repair, while preclinical studies suggest potential in reducing amyloid-beta plaques in Alzheimer's disease. In dentistry, LLLT mitigates oral mucositis in chemotherapy patients and supports periodontitis treatment, with 2018 consensus guidelines endorsing 660–830 nm wavelengths for pain relief [12]. Additionally, sports medicine employs LLLT to enhance muscle recovery and reduce fatigue, exemplified by a 2019 study where athletes treated with 630 nm lasers (5 J/cm^2) exhibited faster recovery via decreased lactate levels and inflammation [12]. These findings underscore LLLT's versatility as a non-invasive therapeutic modality.

Depending on where the therapy is being administered, a laser used in acupuncture can go through different kinds of tissue and pass through a spot on the skin's surface at a certain depth and distance [1, 10]. Human skin is thought to be moderately resistant to radiation at wavelengths below 800 nm, with a thickness that varies from 1-3 mm depending on the area [11-13]. Therapists frequently wonder if the laser light files used in acupuncture can precisely reach the target spot at a certain depth because of the tissues' absorption throughout the process. It is anticipated that this study's attempt to ascertain each tissue's absorption coefficient would yield a picture and forecast of the laser light files' power resistance when therapy is implemented [14]. This relates to a conundrum that acupuncturist therapists frequently face: figuring out how strong of a laser to employ runs the risk of damaging the tissue that the laser is going through. Nevertheless, the laser cannot reach the intended spot if the power is too high. Thus, the existence of this research may be used to further develop and improve the LLLT-50 device as an acupuncture laser therapy tool. It is capable of optimizing treatment parameters such therapy duration, energy consumption, and laser output power or irradiation intensity [15, 16].

The tissue samples utilized in this investigation were samples of leg tissue from cows and chickens. The structure and organization of muscle, connective tissue, fatty tissue, and blood vessels in the meat tissues of sheep, pigs, and cows are comparable to those seen in humans [9, 17, 18]. In addition to studying patterns of laser light absorption and distribution with human biological conduits, the use of such biological tissue is done to understand how laser light interacts with human body tissue [19-21]. Tissues from chickens and cattle include chromophores similar to those seen in human tissues, including myoglobin and hemoglobin [22]. An earlier study found that chromophores have a significant role in the absorption of light at 650 nm wavelengths, which can enhance tissue oxygenation and blood circulation in laser treatment [23, 24]. The choice of cow meat and chicken tissue is also evaluated in terms of sample affordability; it is easily replicable, comes at a reasonable cost, is easily accessible, is more ethical than using human tissue, and has a low risk of infection. To optimize LLLT applications, it is essential to understand laser absorption characteristics in biological tissues. This study utilizes chicken and cow meat tissues as models for human tissues due to their comparable optical properties.

2. Materials and Methods

This research used an experimental method to examine the absorption value of laser 650 nm using LLLT-50 on chicken and cow meat tissue. Then, the researcher measured the value result quantitatively. The results are described on the chart in the result section. The instruments used in this research were a 010W handheld laser power meter and an HWLPM Mini 10W.

2.1 Correlation between the thickness of the different biological sample and the absorption value of laser 650 nm using LLLT-50

This research focused on the development of LLLT-50 device and the experimental impact analysis of thickness tissue on the power absorption using LLLT-50 device. We also analyzed the absorbance value of laser 650 nm using LLLT-50 output on the chicken-meat tissue and cow-meat tissue.

In this study, laser intensity interaction data were collected with 650 nm wavelengths on chicken and cow meat tissue. This study used the LLLT-50 laser device that can produce output power in 3 variations, namely 20 mW, 25 mW and 50 mW. LLLT-50 has a laser probe that uses 650 nm wavelengths. The LLLT-50 device has also had laser exposure energy variations with variations 0.2J, 0.4J, 0.5J, 1.0J, 1.5J, and 2.0J. This laser is one of the developments of the Red and Blue-Violet Laser device (Wardhani et al., 2022). Setup data retrieval was portrayed based on Fig. 1.

To ensure the accurate experimental results, the tissue samples were handled using strict methods:

2.1.1 Initial Cleaning

The tissue was washed with a physiological saline solution (0.9% NaCl) to remove contaminants that could affect light absorption. Drying was performed using sterile gauze before cutting.

Fig. 1. Settings of laser intensity experimental measurement on chicken and cow meat samples tissue.

2.1.2 Cutting and Thickness Measurement

The samples were cut using a sterile surgical scalpel to obtain various tissue thicknesses. The thickness of the tissue was measured using a digital caliper with an accuracy of 0.05 mm to ensure precise measurements. The samples used were fresh tissue, not preserved, to maintain their natural optical characteristics.

The samples were prepared by cutting them into pieces of variable thickness, storing them at temperatures below 0°C to prevent tissue degradation before data collection. The sample variations performed in this study were 3 variations in the thickness of chicken meat and 3 variations in the thickness of cow meat. In both samples, the meat was derived from the leg parts for each animal type. The poultry tissue sample had variations of thickness 0.65 mm; 0.85 mm; and 1.6 mm. Meanwhile, the variation of the cow meat tissue samples was 0.30mm; 0.60 mm; and 0,85 mm. The measurement of the tissue specimen's thickness was performed using the length range with a rigidity tolerance of ±0.05 mm. The samples were not frozen (-20°C or lower) because the freezing process could alter the tissue structure, affecting light interaction with the tissue.

Before measurement, the samples were left at room temperature (±25°C) for 5–10 minutes to prevent significant changes in optical characteristics due to temperature differences.

2.2 Measurement of laser light absorption on the biological sample

On the measurement of laser light absorption, the sample was placed vertically with the position of the laser beam of the LLLT-50 device. The intensity of light was measured before and after passing through the sample using a power meter to determine the absorption value. Measurements were performed 10 times for each variation of the sample to ensure accuracy and minimize the occurrence of experimental errors. According to these repeated measurements, an average was calculated which was then used for further analysis. The measurement of laser absorption carried out at the distance between the sample and the laser light source was made fixed, and the samples were placed on the surface of the power meter. In addition, the measurement of laser light absorption in this tissue used the energy variation found in the LLLT-50 device. The energy variance used was 0.5 J and the laser output power was variated at 20 mW; 25mW; and 50mW. To reduce the presence of other factors, such as diffusion and reflection, laser light intensity measurements were conducted when no samples were available to determine the initial measuring limit. Analysis of light intensity data obtained by calculating absorption values using Eqs. (2.1)-(2.3), [25, 26].

$$P = P_0 e^{-\mu x} I = I_0 e^{-\mu x}, \qquad (2.1)$$

$$P = P_0 e^{-\mu x} P = P_0 e^{-\mu x}, \qquad (2.2)$$

where in Eq. (2.1) I is the light intensity (W/cm^2) on the tissue surface, I_0 is the intensity of light that is transmitted directly to a tissue sample, is the absorption coefficient, and x is the thickness of the sample tissue Eq. (2.2). In this study, the intensity or force that penetrates the tissue is recorded with a power meter on variations in the thickness of cow meat and chicken tissue. The value of the absorption coefficient (μ) on each variation of the sample tissue can be obtained from the μ value on the Eq. (2.3) [12, 25, 27].

$$\begin{aligned} \ln P &= \ln P_0 - \mu x, \\ \ln \ln P &= \ln P_0 - \mu x. \end{aligned} \qquad (2.3)$$

3. Results and Discussion

3.1 Comparison of 650 nm laser light intensity values with LLLT-50 devices measured on chicken and cow meat tissue

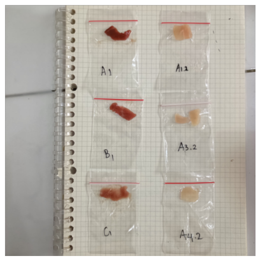

Fig. 2. Chicken and cow-meat tissue before laser applied.

This study resulted in data comparison of the 650 nm laser light power value with the LLLT-50 device transmitted to the tissue of chicken and animal samples as shown in Figs. 2-3. Based on the data obtained from the research results, the power value measured on the power meter using the variation of the laser power output on the LLLT-50 device experienced a significant decrease in the thicker tissue thickness (1.60 mm). Based upon Fig. 4, it can also be seen that the power measurement

Fig. 3. Chicken and cow-meat tissue after laser applied.

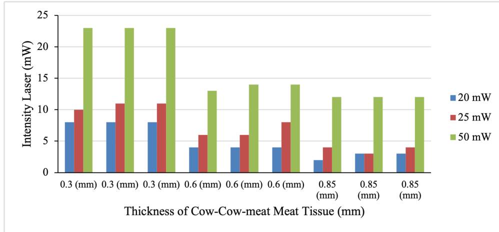

Fig. 4. Laser power intensity (mw) measured on the powermeter against the thickness of the chicken meat sample tissue (mm) using the LLLT-50 device.

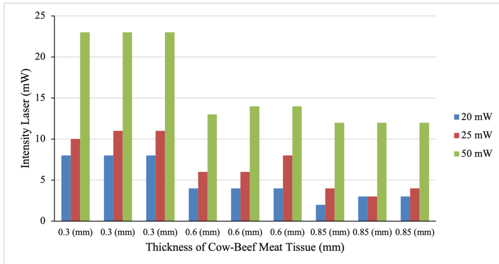

Fig. 5. Laser power intensity (mw) measured on the powermeter against the thickness of the cow-cow-meat meat sample tissue (mm) using the LLLT-50 device.

on the highest power meter was achieved at a variation 50 mW of the output power of the laser device LLLT-50 for the three variations of the thickness of the tissues of the chicken sample. In the variance of the lowest thickness (0.65 mm) of chicken tissue, the maximum measuring power value reached 60% of the maximum laser output of LLLT-50. Based on Fig. 5, the results of the observations of the laser power measured at the thickness variations of 0.30 mm, 0.60 mm, and 0.85 mm for the tissue variants of the cow meat sample are shown. On the result of the measurement, the characteristic trend of the result is the same as in Fig. 4. Whereas in Fig. 5, the value of the power measurable after passing through the highest cow-meat sampling tissue is the thinnest tissue thickness (0.30 mm) for the three laser output power variants on the device LLLT-50. Based upon Fig. 4, it can also be seen that the power of the most high-power meter is obtained at the 50-mW variation of the output of the device laser LLL T-50 for three variations in the tissue thickness of the meat sample.

Based on data from power measurements measured on power meters for variations in chicken and cow meat tissue samples, the overall power values in cow meat variants are lower than in chicken tissue variants. This is because the cow meat tissue has the surface color of red meat that can blur the red laser light used. Unlike colored chicken tissue that tends to be brighter, the intensity of red laser light can be optimally passed onto the power meter. The cow meat tissue is more red and darker due to its more complex structure, and it has a lot of connective tissue that can spread and absorb light. In addition, the color of cow meat tissue is also affected by the higher content of myoglobin than chicken. Myoglobin is a protein that is able to store oxygen in the muscles that can absorb red light. The cow meat sample also contains higher levels of collagen and fat compared to chicken tissue. The high levels of collagen and fats on the surface of the sample can inhibit the absorption of red light (650 nm) [25, 28, 29].

The biological tissue thickness used is closely related to the theoretical analysis of the physical power absorption of lasers at 650 nm wavelengths. Lambert-Beer's law says that tissue thickness increases light absorption because light must travel longer distances. [30, 31]. As a result, chromophores like hemoglobin and myoglobin absorb more energy. The thickness of the tissue also affects the penetration of light; thicker tissue tends to reduce the intensity of light passing through due to greater absorption and distortion. To ensure the efficiency and safety of laser therapy, understanding the relationship between 650 nm wavelengths and tissue thickness is crucial in laser therapies as it is important to optimize parameters such as the intensity and duration of irradiation to the desired penetration and therapeutic effect without damaging the tissue. The overall interaction suggests that tissue thickness should be considered specifically in clinical applications. Herewith attached the result of chicken and cow meat tissue before and after laser applied.

The laser absorption coefficient value (μ) is a parameter that can describe how effectively a material or tissue receives or absorbs laser light with a certain wavelength [28, 29]. This absorptive coefficient has different values depending on each tissue affected by the chemical composition as well as the physical construction of the tissue [32]. Chicken tissues, with simpler tissue structures and lower levels of myoglobin, result in more light being transmitted on the power meter. Meat tissues, with more complex tissue and more myoglobin, and containing white fat, has higher absorption values so that it can reduce the transmission of light on the power meter. High white fat (intramuscular) is capable of absorbing and spreading light.

Therefore, based on the observations presented in Figs. 6-7, the 650 nm laser

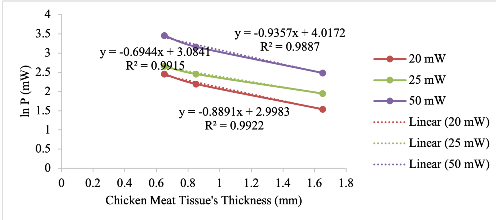

Fig. 6. Graphic correlation of measured power (ln *P*) with thickness of net of chicken meat sample.

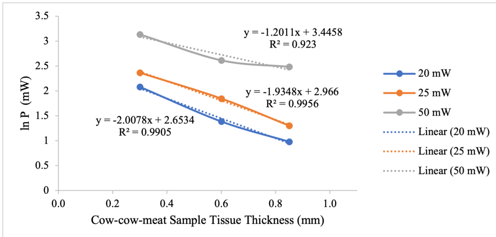

Fig. 7. Graphic correlation of measured power (ln *P*) (mW) to cow-meat tissue sample thickness (mm).

absorption coefficient value in the chicken sample tissue has a lower value compared to the value of the cow-meat samples tissue absorption coefficient. This can be seen in the slope value on the linear chart for each data variation of the sample. The slope values in Figs. 6-7 are the values of the absorbance coefficient derived from the correlation between the measured laser power and the thickness of the samples. At Eq. (2.3), when a graph gradient analysis is performed, the value is the graph gradient value of Figs. 6-7. In the results of Fig. 6, the absorption values in chicken tissue are obtained for each variation of the power output of the device LLLT-50, respectively, 0.89. (20 mW); 0.69 (25 mW) and 0.94 (50 mW). In the result of Fig. 5, the absorbance values on cow meat tissue for each variant of the output power of the unit LLLT-50 are obtained, respectively. 2.00 (20 mw); 1.94 (25 mw) and 1.20 (50 mW).

In previous studies it was seen that chicken has a lower absorption coefficient than cow meat. This is because chicken contains less myoglobin compared to cow meat, so chicken will absorb light at a wave of 650 nm. A simpler and less dense chicken structure compared with cow meat also causes more red light to be passed on to the power meter than the light absorbed in the meat.

4. Conclusion

Low-level laser therapy that is widely used in acupuncture lasers can stimulate healing and reduce pain without significant side effects. The study conducted a 650 nm laser light absorption test with the LLLT-50 device on various thicknesses of chicken and cow meat tissue to determine the absorbance coefficient value, with the light intensity observed before and after passing the sample with the power meter. Based on the results of the study, it was concluded that the 650 nm laser absorption coefficient value with the LLLT-50 device in chicken tissue has lower values than cow meat because of the content of myoglobin and the simpler tissue structure (no complex muscle tissue), does not contain much fat, and the color is brighter than the cow-meat. Besides variations in in myoglobin concentration, the research demonstrates that chicken tissue displays lower absorption coefficients (μ = 0.69–0.94 cm^{-1}) relative to cow-meat tissue (μ = 1.20–2.00 cm^{-1}) at 650 nm, due to collagen/fat composition, and structural complexity. The result is consistent with literature indicating that heme groups in myoglobin and hemoglobin strongly absorb red light (600–700 nm) [16, 22].

In the study, the use of chicken and cow meat tissue to determine the interaction of laser light with human body tissue could be done to optimize acupuncture laser therapy parameters with the LLLT-50 device. This study can address concerns about the proper laser power to reach the therapeutic point without tissue damage. These results help refine LLLT settings for therapeutic use, particularly in acupuncture laser therapy. Future research should explore in vivo studies and optimize treatment parameters for human applications.

The ex vivo model excludes dynamic factors like blood flow, which alter in vivo absorption. Additionally, focusing solely on 650 nm limits comparisons to studies using broader spectra. Future research could incorporate computational approaches, such as Monte Carlo simulations, to model scattering or validate results with human tissues under clinical conditions.

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