

Future Exploration Using Photobiomodulation as a Treatment for Military Working Dogs

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ABSTRACT

Introduction: Photobiomodulation is a non-invasive therapy that addresses muscle fatigue, damage, and inflammation. This review explores the efficacy of PBMT using canine and murine models. By doing so, we show why PBMT warrants investigation as a medical technology to prevent and accelerate recovery from soft tissue, muscles, and minor wounds commonly sustained by Military Working Dogs (MWDs). Additionally, we aim to find the ideal PBMT fluence and wavelength that may accelerate the healing process and enhance physical performance.

Methods: We assessed 88 sources published between 1986 and 2023 from three databases, Google Scholar, Cochrane Library, and the Army Medical Department Virtual Library, using an appraisal checklist developed by the authors. Results: After excluding 45 articles that did not meet our inclusion criteria, we included 43.

Discussion: Our assessment showed that PBMT should be considered a supplementary treatment for animals affected by various muscular and health conditions. The therapy uses specific wavelengths and fluences of light, ranging from 632 nm to 1064 nm and 1 J/cm² to 50 J/cm².

Conclusion: Future empirical testing that is necessary to find the ideal dose parameters to treat specific types of injuries sustained in MWDs. Regrettably, many military canines suffer from musculoskeletal and soft tissue injuries. PBMT should be considered as a potential supplementary treatment technology for veterinarians and dog handlers to help dampen injuries and enhance physical performance and recovery, especially in a deployed setting. However, our working hypothesis should be tested through future quantitative field studies.

Keywords: Photobiomodulation; Light-Emitting Diode Therapy; Canines; Musculoskeletal Injury; Soft Tissue; Military Working Dogs

Abbreviations: MWDs: Military Working Dogs; PDMT: Photobiomodulation Therapy; ROS: Reactive Oxygen Species; NO: Nitric Oxide; SANRA: Scale for the Assessment of Narrative Review Articles; AMEDD: Army Medical Department; AVL: Virtual Library; LED: Light-Emitting Diodes; CCO: cytochrome C Oxidase; ETC: Electron Transport Chain; ATP: Adenosine Triphosphate

Introduction

The primary aims of this review are to examine Photobiomodulation Therapy (PBMT) as a treatment method, particularly in accelerating the recovery from muscular, soft-tissue, and minor wound-related injuries and preventing further damage. It is important to note that injuries in Military Working Dogs (MWDs) are often underreported. A 2019 U.S. Army public health report revealed that many medical encounters in MWDs are due to soft tissue and musculoskeletal injuries [1]. Currently there is a deficit in the existing scientific literature on PBMT which contains minimal collections of information on the scientific rationale behind the mechanisms of action of this therapy. This review discusses these deficits that need to be addressed for PBMT to be recognized as a practical treatment option that will be integrated into clinical practice. PBMT is a treatment that uses visible red and near-infrared light at specific wavelengths and intensities that stimulate enzymatic activity within cells. Currently a challenge faced through PBMT is that doses are often delivered at too high a fluence (W/cm^2), with too many treatment intervals, or for extended durations, which in turn increases levels of Reactive Oxygen Species (ROS) and Nitric Oxide (NO) leading to inhibitory effects and toxicity at high levels [2,3].

Well-defined PBMT dosing parameters for treating specific acute or chronic injuries are not well delineated in the literature, especially when it comes to the ideal PBMT treatment parameters based on desired therapeutic outcomes, which rely significantly on the fluence (W/cm^2) and doses (energy, time, treatment interval), energy density (J/cm^2), and frequency of application [4,6]. PBMT can be equated to medication that has active ingredients [6]. These are referred to as the irradiation parameters in PBMT, which include the wavelength (nm) used to treat, fluence (J/cm^2), pulse structure used (continuous vs. non-continuous), light length coherence, and polarization (linear/circular) [7]. The PBMT "dose" is the irradiation time and treatment periods (minutes, hours, days, weeks) that serve as the medicine for the treatment [6]. Therefore, the efficacy of light therapy in treating various injuries and health disorders depends on how well the intervention (measured in fluence and wavelength) is applied, through a set duration, with the proper application of the PBMT device to the affected part of the body.

Most studies using PBMT ineffectively depict an improper application of this treatment based on the injury or health disorder type [6,7]. Therefore, establishing a more comprehensive understanding of the application and delivery of PBMT based on the type of injury/pathology being treated must be further explored for successful treatment outcomes. PBMT has primarily been examined in murine models, but veterinarians have reported the efficacy of light therapy through case and observational studies [8,9]. Many of these studies have shown that photoreceptor receptor activation in the mitochondria regulates many biological processes and signal transduction pathways [8]. Based on the published data, PBMT is now a therapeutic

tool for certain wound types, injuries, muscle fatigue, strokes, and nerve injuries in canines and murine models [6]. Studies show that PBMT could hasten physiological recovery and improve performance [9,10]. MWDs often suffer acute and traumatic injuries while working in challenging environments, but there is limited research in this domain [1,11]. Therefore, the primary objective of this review is to fully understand the mechanisms underlying PBMT and clearly define parameters at which the treatment is maximally safe and effective to treat an array of muscular and soft-tissue injuries.

Methods

This NR was conducted according to Ferrari's framework for narrative reviews as described in "Writing Narrative Style Literature Reviews" [12]. The review was also written according to the Scale for the Assessment of Narrative Review Articles (SANRA), a tool developed by [13]. Primary, secondary and tertiary sources relating to the review's aims were found through a search of the Army Medical Department (AMEDD) Virtual Library (AVL) from 1986 to 2023, as well as the Cochrane Library and Google Scholar for the same period.

Inclusion Criteria

We established specific criteria for selecting primary and secondary sources for our review. These criteria align with the review's main objectives, as shown in Table 1. First, we only considered sources that addressed the population, focused on concepts related to the research aims, and reported the use of PBMT in a clinical or field setting. Second, we focused on the biochemical mechanisms, types of injuries in MWDs, wound healing, and the recommended PBMT dosing parameters to enhance physiological recovery. Collectively, we selected sources that examined these key concepts and contexts related to the population.

Search Strategy & Screening

We started our search in the Army Medical Department (AMEDD) Virtual Library (AVL) by selecting the Veterinary Medicine category from the menu. We then conducted an advanced search by selecting "Find all my search terms" in the search mode inquiry box. To ensure the most relevant results, we chose the "Apply related words" and "Search within the full text of the articles" options. We set our limiters to full-text articles, peer-reviewed, date published (1986-2023), Language English, and available in the Library Collection. Our search terms were (military working dogs) AND (canines) OR (low-intensity laser therapy) OR (photobiomodulation therapy) AND (musculoskeletal injury) OR (soft-tissue injuries) OR (muscle performance) OR (performance recovery). This search yielded 10 articles that were relevant to our research aims. For our second search in the AMEDD AVL, we used the same strategy as before but with different search terms. We entered working dogs (TX All Text) AND murine model (TX All Text) OR low-level laser therapy (TX All Text) OR photobiomodulation therapy (TX All Text) AND injury prevention (TX All Text). Our initial and secondary searches in the AMEDD AVL database produced

124 preliminary articles related to our working title, review aims, and objectives. We thoroughly searched the Cochrane Library for articles published between 1986 and 2023, using advanced title, abstract, and keyword search.

Our search was limited to Cochrane Reviews and Cochrane protocols. We used the word variations “working dogs” OR “canines” OR “humans” AND “low-level laser therapy,” which resulted in 53 trials. However, only some of these trials were not relevant to our aims. Instead, they focused on using low-level laser therapy for orthodontic tooth movement and tooth-related pain. For our second search in the Cochrane Library, we used the word variations “working dogs” OR “canines” OR “human case clinical reports” AND “low-level laser therapy” OR “low-level light emitting diode” OR “photobiomodulation.” Unfortunately, we found no Cochrane reviews or protocols related to our primary and secondary objectives. However, we did locate 138

articles relevant to our population and concepts. For our final search in Google Scholar, we performed an advanced search for articles between 1986 to 2023. Search limits consisted of only finding articles “with all of the words” and “anywhere in the article” using the word variations “military working dogs” OR “canines” AND “low-level laser therapy” OR “photobiomodulation therapy” AND “murine model,” which yielded 1,800 articles. However, none of the articles were related to our review objectives. Hence, we performed a second basic search query in Google Scholar using the following word variations: military working dogs, injuries, low-level laser therapy, canines, and performance recovery. All three database searches resulted in retrieval of 88 articles that were assessed for eligibility, while 45 were excluded for not meeting populations, concepts, or context, leaving 45 articles for inclusion.

Results

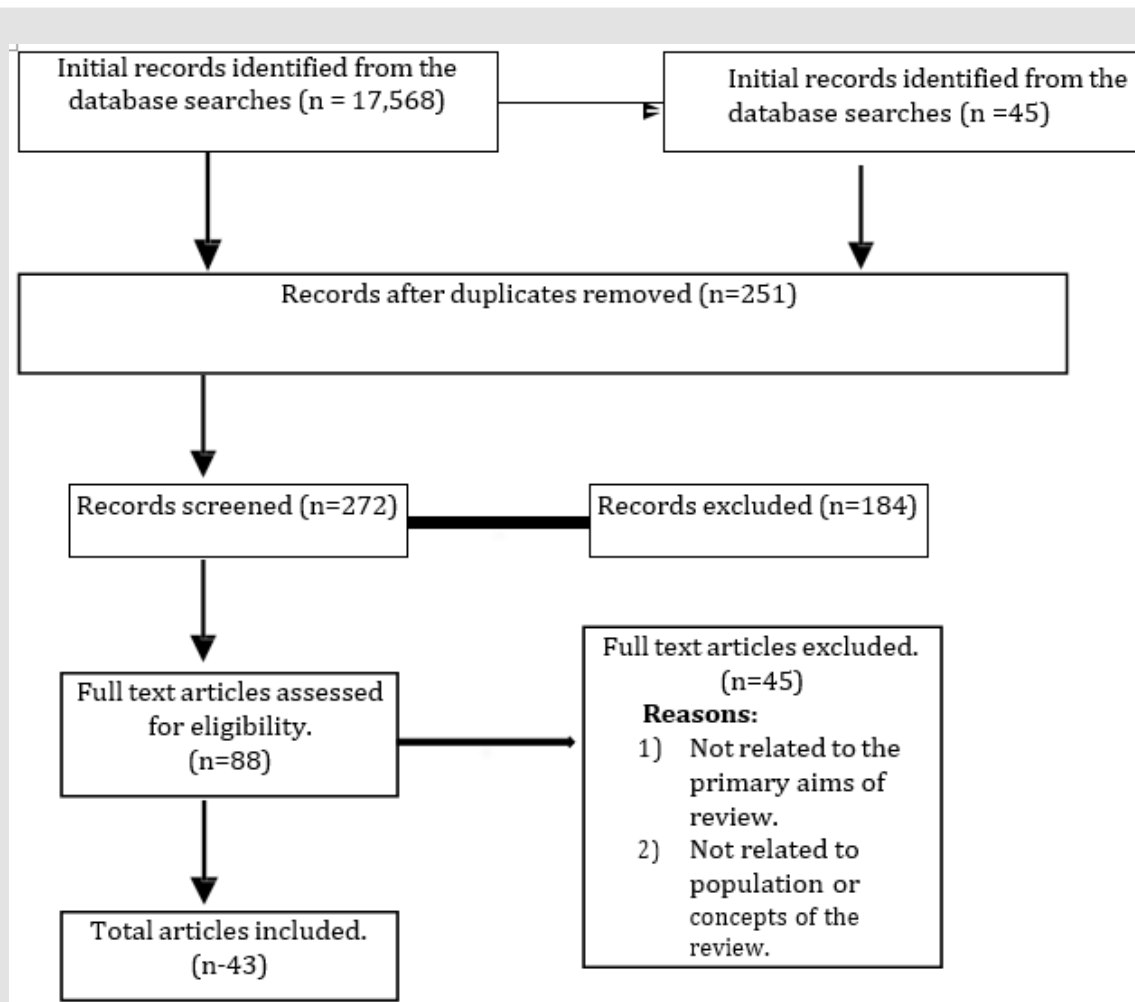


Figure 1: Flow chart of literature selection.

The flowchart for selecting sources is depicted in Figure 1. The articles were screened based on the inclusion criteria depicted in Table 1. Out of all the screened articles, 43 were selected for inclusion. The sources selected for inclusion included original research articles

(n=15), review articles (n=22), technical reports (n=2), case control study (n=1), retrospective studies (n=2), and conference/symposium (n=1).

Table 1.

	Inclusion Criteria
A.	Original research articles
B.	Case studies/retrospective case series
C.	Review articles of any type (invited, systematic)
D.	Observational, prospective studies
E.	Expert topical/commentary reviews
F.	Technical/special reports
G.	Free full-text article
H.	Population(s): canines, military working dogs, animal models
I.	Concepts related to PBMT use: biological mechanisms, use of PBMT in the military, injuries of canine and military working dogs, wound healing effects, acceleration of performance recovery, PBMT use on animal training models.
J.	Context: use of PBMT in clinical or field setting
K.	Types of PBMT devices and dosing treatment parameters.
L.	Published between 1986 to 2023
M.	Written in the English language

Note: Must meet at least one criterion (a-f), only eligible articles considered are (g-m).

Discussion

Recent studies suggest that PBMT therapy can be an effective tool in treating acute and chronic states of pain across multiple pathological and physiological causes [6,7,9,14]. This review breakdown the currently literary basis into four sections: the proposed biophysical mechanisms of PBMT, the most common injury presentations of canines, PBMT treatment ranges of effective treatment and utilizations of treatments across various types of muscular skeletal injuries in canines.

Biophysical Mechanisms Associated with PBMT

PBMT is a non-invasive medical treatment that is used to accelerate the healing of soft tissue injuries dyschromia's, and immunosuppressive-related disorders of the nervous system [9,14]. The therapy employs a small, charged particle known as a photon, which is a packet of electromagnetic energy known as a boson that interacts with electrons based on frequency (energy) and wavelength (momentum) [5,6]. Photons traveling at the frequency and wavelength within the red/NIR range of the visible and non-visible light spectrum, can be absorbed by affect cells at the molecular level, influencing mitochondrial activity of specific subcellular chromophores, such as cytochrome c oxidase (CCO), flavins, flavoproteins, and opsins [15,16]. PBMT light can penetrate the cellular membrane and activate biological processes, such as cell growth, propagation, and increased reactions of critical enzymes in the respiratory chain [17]. Red light has higher photon

energy, which can induce electrochemical changes at the tissue level, while NIR light has lower energy acting at the atomic level [5,18,19]. The depth of skin cell penetration relies on wavelength [19]. Further, the irradiation dose absorbed by the cells is a function of energy density and radiation treatment time [19,20].

The conflicting outcomes concerning the efficacy of PBMT can be attributed to the non-specific guidelines for a surplus of injuries and pathologies and often reflect the failure to adjust irradiation parameters (photon wavelength), irradiation time, and energy dispensed (dose) [5,6]. The applications of PBMT can be utilized for a variety of treatment remedies and with each, a parameter of effective wavelengths needs to be established. Recent studies have found that application on muscle tissues which contain higher concentrations of mitochondria responded well to PBMT wavelengths between 655 nm and 850 nm [17]. When applying this treatment in the realm of wound healing it has been found to accelerative healing of wounds at wavelengths ranging from 620 to 740 nm [17]. One must consider that the effectiveness of PBMT is dose-dependent which corresponds to the Arndt-Schulz Law (ASL), which says that small doses of stimuli enhance cellular activity, while more potent stimuli delivered inhibit activity, and results in harmful effects [6,17].

Using the ASL rule, the principal PBMT dose parameters are wavelength (nm), treatment time, treatment intervals, and fluence (W/cm^2) [3,4]. Therefore there is a need to establish safe and effective

dosing parameters that are based on the type of injury or pathology being treated [17]. Mitochondria are tiny organelles found in the cells of living organisms that are vital for energy production, cellular respiration, and cell death [21,22]. These organelles are known as the “common chemical intermediate” as they aid the metabolic pathways that generate adenosine triphosphate (ATP) [21-24]. Mitochondria also possess mechanisms regulating cell death, which is activated at naturally occurring timepoints in the cell cycle or when cellular processes become dysfunctional. The conditions that predispose apoptosis can lead to metabolic, neurodegenerative, and cardiovascular diseases [21-24]. Therefore, preserving healthy mitochondria is critical for supporting energy transfer to organs and skeletal muscle tissue and preventing cell death. Red/NIR light therapy is suggested to be an effective treatment modality for *in vitro* studies with high numbers of mitochondria when Light-Emitting Diodes (LED) are delivered within the 660 nm to 810 nm wavelength range, while *in vivo* studies using LED were found to be effective within the range of 635 nm to 1064 nm [20]. This wavelength range is suggested to promote cellular activity, prevent cell death, and enhance ATP synthesis in the mitochondria [20].

Cytochrome c oxidase (CCO) is a protein found in the mitochondria of cells that plays a role in cellular respiration by hastening the transfer of electrons from cytochrome c to oxygen molecules [15,16,20,21,25,26]. This transfer produces the energy needed to yield ATP. The CCO can be excited by visible light penetrating the cellular membrane and triggering biochemical changes in the mitochondria [15,20]. The CCO subunit four in the mitochondrial Electron Transport Chain (ETC) is implied to be the most sensitive to the red/NIR light spectrum [26-28]. When exposed to the photons of light, the CCO subunit four promotes a series of photochemical events in the cell mitochondria's ETC, which foster enzymatic activity and amplify mitochondrial membrane potential [16]. Red/NIR light matches the absorption spectrum of CCO. It thus stimulates the CuA and CuB copper centers of CCO, allowing nitrogen oxide (NO) to dissociate from heme- $\alpha 3$ and CuB, increasing respiration and ATP synthesis and reducing oxidative cellular stress [16,24,25,27,28].

The latest studies on PBMT have revealed that it can have variable effects on cells with high mitochondrial activity [20]. Nevertheless, PBMT has also been revealed to hasten the healing of wounds, foster cell adhesion, augment collagen production, and decrease inflammatory response [21,25,28]. PBMT has been shown to reduce inflammation in rat models, downregulate pain-related symptoms in canines, and improve post-surgical recovery in humans and canines [29].

Synopsis of Combat & Noncombat Related Injuries in MWDs

In 2021, Schuh-Renner and colleagues conducted a retrospective study on 774 active military working dogs (MWDs) [30]. This study aimed to assess medical encounters, injury risk factors, and other related medical issues. The study included both univariate and

multivariate analyses of factors such as sex, breed, certification type, duty location, and service branch associated with a host of injuries and pathologies such as dermatologic, alimentary, dental, soft-tissue, and muscular issues [31]. Most of the MWDs were male (74%) with a dark coat (83%) certified in Explosive Detection (60%), and most of the breeds consisted of German Shepherds (39%) or Belgian Malinois (31%) [30]. It was found that 14% of non-surgical medical issues were related to musculoskeletal (MSK) conditions and soft-tissue injuries. The study also revealed that 83% of MWDs had suffered from a non-surgical medical issue [31]. For example, 28% were still affected by injuries, and 13% suffered from MSK-related conditions [31]. In 2019, the U.S. Army Public Health released a report that revealed that a significant number of MWDs suffer from MSK issues [1].

The report examined a cohort of German Shepherds and Belgian Malinois and found that 13% of German Shepherds and 15% of Belgian Malinois had MSK problems [7]. Females were found to suffer from the highest percentage of MSK problems (18%) compared to males (13%) [1]. The study also discovered that 40% of 440 active MWDs suffered from soft-tissue-related injuries, which accounted for 10% of medical encounters [1]. Cumulative MSK injuries in MWDs are usually the result of repetitive use of the musculature and are underreported [1]. It is essential to understand the most common risk factors correlated with medical difficulties that can hamper the performance of MWDs [11,31,32]. The cost to acquire one MWD ranges from \$3,000 to \$4,000. However, after the necessary training requirements are fulfilled, the value of the canine ranges from an estimated \$11,000 to \$40,000, and canines trained in explosive detection can be worth up to roughly \$80,000 [32].

A retrospective descriptive study conducted in 2014 assessed 1,350 medical encounters from January 2009 through August 2010 and found that MWDs experienced dermatologic issues (25%), soft tissue trauma (21%), digestive diseases (17%), or musculoskeletal (MSK) issues (14%) [33]. PBMT may serve as a valuable complementary non-invasive treatment technology to accelerate the recovery of MWDs from muscle-related injuries, especially when veterinary services are not readily available downrange. However, to ensure PBMT's best use as an adjunctive therapeutic device for MWDs, it is necessary to assess safe and effective PBMT fluence (W/cm^2) and wavelength (nm) parameters and whether a continuous or pulsed wave is most effective in decreasing inflammation, lessening pain associated with chronic muscular disorders, and accelerating healing of superficial wounds [34]. Future investigators must report important information about the correct dosing parameters for PBMT, particularly regarding the type of injury, the tissue type being treated, and the frequency of its application to the injury site [5,7,9,33].

However, PBMT has analgesic and anti-inflammatory properties without side effects [29]. Therefore, its efficacy as an ancillary non-pharmacological therapy in an athletic canine cohort should be further examined. Earlier studies on living dog tissue have shown that PBMT can significantly hasten wound healing, improve post-opera-

tive rehabilitation, and lessen soft tissue and musculoskeletal edema pain [29]. This evidence highlights the possible use of PBMT as a supplementary treatment that could work in concert with conventional veterinary treatments to accelerate recovery from post-operative injuries and reduce pain associated with injury.

Impact of PBMT on Wound Healing

While light energy is shown to enhance the body's natural healing processes, more information is needed to describe the parameters of PBMT dosing schedules with guidelines on the upper and lower range at which the treatment is safe and efficacious, while also defining the side effects that may be seen when PBMT treatment is delivered at a dose outside safe ranges [6,35]. Specifically, there is no consensus among medical experts on the efficacy of PBMT for treating burn wounds, and paw pad lacerations, which are the most often reported non-combat injury in MWDs in a combat setting [1,6]. A recent review described PBMT is an effective non-invasive treatment modality in accelerating wound healing by targeting mitochondrial proteins and activating the release of nuclear factor kappa B (NF- κ B), which is essential for cytokine production, transcription of DNA, innate immunity, production of ATP, and syncretization of fibroblasts, which aid in the tissue and wound healing process [36]. An additional study suggested that PBMT using wavelengths of 680, 730, and 880 nm simultaneously, alone, or when combined with hyperbaric oxygen therapy, can accelerate the healing of oxygen-deprived wounds [37]. In their research, Mosca and colleagues explored using PBMT for treating a range of wounds [7]. This research emphasized the need for more objective reporting of the photochemical approach to ensure clinical safety, efficacy, accuracy, and validity in future studies, as they found a significant irregularity in the treatment doses employed by various investigators [7]. They also recommended the exploration of necessary molecular biomarkers of wound care that may be targeted by PBMT treatment, that the effectiveness of PBMT may vary depending on the wound type, and that the unpredictability of PBMT dosing parameters administered should be considered and accurately defined so that future wound care and healing research can deliver more successful treatment protocols [7].

Hoisang et al. [38] studied chronic wounds in canines using PBMT. They used a single wavelength of 830 nm, an irradiation fluence of 4 J/cm², and simultaneous super-pulsed and multiple wavelengths (SPMW) of 660, 875, and 905 nm [38]. In this study, canines were randomly distributed into one of three experimental groups: 1) Control canines treated with irrigated saline and no PBMT, 2) PBMT-L1 canines treated with one wavelength of 830 nm and irrigated saline, or 3) PBMT-L2 canines treated with SPMW and irrigated saline. Wound healing rates significantly improved in the two PBMT treatment groups compared to the control [38]. The study's findings may not directly apply to MWDs due to differences in body mass hair length, breed type, and coat but should be considered through future empirical studies.

Influence of PBMT on Muscle, Performance & Recovery

Both humans and canines have four types of central skeletal muscle fiber types: Type I (oxidative) and Type IIa, which consists of a combination of oxidative and non-oxidative properties; and Type IIx and IIb (fast-glycolytic fibers), which have fewer mitochondria [39]. For example, well-trained athletic canines and humans will produce more lactate during physical activity but can uptake more and use it as a gluconeogenic precursor [40]. Thus, by increasing the mass of the mitochondrial network and expanding the number of lactate transporters, the athletic canine may enhance oxidative (aerobic) endurance performance, which could be vital to mission success. This lactate shuttling process occurs in the mitochondrial reticulum, transporting lactate and then oxidizing it as a functional energy substrate used by the skeletal muscles, kidneys, liver, and brain [40].

Transporter proteins move lactate from the outer membrane to the inner membrane of the mitochondria, whereby lactate is oxidized to pyruvate and moved into the Krebs cycle to be converted into adenosine triphosphate (ATP) [40-42,43]. Hence, one must consider the potential synergism of PBMT for accelerating metabolic and cellular transformations in muscle tissues. Priyadarshi et al. [42] found that using a continuous PBMT wavelength of 808 nm and a pulsed wavelength of 905 nm accelerated nerve repair and muscle recovery while also restoring nerve cells.

The formation of a mitochondria network depends on the type of physical training. Further, extended patrols beyond the muscular adaptive properties of a canine may result in excessive lactate production. PBMT should be studied as a synergistic device when combined with canine physical training using specific PBMT dosing parameters (fluence, wavelength, pulse structure), time (seconds), and treatment interval (hours, days, weeks) [6]. Limitations of dosimetry parameters, such as hair shaving due to hair length, breed type, coat texture, and skin pigmentation, may affect the penetration depths of the fluence and wavelength used to treat [48]. Studies focused on the effects of PBMT treatment in athletic canines are necessary to define the optimal wavelength (nm) and fluence (W/cm²) [9].

Determining the ideal therapeutic window for accelerating physiological recovery following exercise training bouts is also essential [9]. Specific outcomes assessed based on the specificity of the exercise training protocol need to be improved in the literature and require future investigations [9]. These protocols included muscular fatigue induced by neuromuscular electrical stimulation, swimming until exhaustion, anaerobic threshold running, and declined (downhill) treadmill running [44]. PBMT wavelengths ranging from 632.8 nm to 904 nm were shown to reduce muscle fatigue, tissue damage, and muscle inflammation [44].

Conclusion

PBMT has been shown to enhance the synthesis of ATP, reduce the release of reactive oxygen species, decrease cell death, promote

the growth of new capillaries, increase localized blood flow, stimulate signaling proteins that affect cellular processes, and downregulate inflammatory markers when used at effect doses. More reliable reporting of specific PBMT dosing parameters, devices used to treat, contact points treated, wavelength, and fluence used to treat before or after various types of activities performed by MWDs may serve as a valuable medical technology in the preservation of health and physical functionality of these canines.

Disclaimer

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Conflict of Interest

No economic interest or any conflict of interest exists.

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