Application of Photobiomodulation in Hearing Research: Animal Study

Jae-Hun Lee  
Jae Yun Jung  

1 Beckman Laser Institute Korea, College of Medicine, Dankook University, Cheonan, Korea  
2 Department of Otolaryngology Head and Neck Surgery, College of Medicine, Dankook University, Cheonan, Korea  
3 Department of Otolaryngology Head and Neck Surgery, Dankook University Hospital, Cheonan, Korea

Hearing organs have unique characteristics and have a role in processing external sensory signals. Sensory hair cells and nerve fibers in the organ of Corti can be damaged by various causes and they do not regenerate themselves. Medication used for clinical treatment for the inner ear is limited due to the anatomical structure of the inner ear. Photobiomodulation (PBM) is a therapeutic approach that uses various sources of light and the success of PBM therapy is highly reliant on the parameters of the light sources. The positive effects of PBM have been reported in various clinical fields. This paper summarizes the previously reported research on PBM for the treatment of hearing damage in animal models.

Keywords  
Photobiomodulation; Hearing research; Hair cell protection; Nerve regeneration; Diode laser
INTRODUCTION

Photobiomodulation (PBM) is a type of therapy using light source including laser and light emitting diodes (LED). More than 1,000 research papers have reported that its positive effect in pain relieving, neuronal regeneration, inflammation reducing, tissue repairing, and nerve promoting. Application and effect of PBM is highly rely on its parameters. These parameters including power density, wavelength, beam spot size, irradiation time, total energy, and number of repetitions are directly related with success of therapy. Combining of laser sources or delivery method can be additional factor to control the effect of PBM.

Hearing problems are the one of three most general social problem in aging society. Several cohort studies reported that over 45% of population in the world have hearing problems. Recently, the prevalence of hearing problems in young generation is increasing, and this could be resulted in communication dysfunction and social burden which also affect their family. Hearing aid and cochlear implant could be alternative way to compensate this problem but these approaches have clear limitation since these only aid remnant hearing function without treatment.

Diverse applications of PBM were reported after it approved by the United States Food and Drug Administration (FDA). Several studies performed PBM on patient with hearing dysfunction, especially for tinnitus. PBM has also been used in pre-clinical hearing research for treatment after damage in both in vitro and vivo (in review). In this paper, we shortly review the previous result using PBM for hearing dysfunction in animal models. We discuss what kind of PBM parameters was used and how is applied to animal.

MATERIALS AND METHODS

Strategy for article identification

Research was conducted using the following databases: PubMed, Google Scholar, and Springer. Keywords used: Photobiomodulation, hearing, LLLT, regeneration, hearing, recovery.

The titles, abstracts, and conclusions were screened and unrelated articles were excluded. Total 38 articles were included and were summarized. Evaluations of collected articles were performed by three reviews.

PARAMETERS OF PBM FOR HEARING RESEARCH

Wavelength

To apply PBM on the cochlea, penetration depth of wavelength is critical. Anatomically, cochlea is position inside of the temporal bone and light can be attached through tympanic membrane (Fig. 1). To compensate this huddle, red and near-infrared (NIR) light of wavelength 600-1000 nm which has greater penetration depth than other wavelengths is generally used.

Laser power and total intensity

Tympanic membrane which is first terminal of light for hearing treatment could be damaged depending on the total power of light. Since total power is composed with laser power, beam spot size, and irradiation time, appropriate parameters for each animal model is needed. Too strong power could lead pathological changes on the tympanic membrane and middle ear.

Irradiation and repetition time

Irradiation times of PBM in previous hearing researches are separated into two times (30 and 60 minutes). Since animal should be anesthetized during PBM irradiation, too long irradiation time is not appropriate for animal study. Furthermore, burning of tympanic membrane or pathological changes in the middle ear tissue caused by long duration of irradiation could occur in animal model.

PARAMETERS OF PBM FOR HEARING RESEARCH

Wavelength

To apply PBM on the cochlea, penetration depth of wavelength is critical. Anatomically, cochlea is position inside of the temporal bone and light can be attached through tympanic membrane (Fig. 1). To compensate this huddle, red and near-infrared (NIR) light of wavelength 600-1000 nm which has greater penetration depth than other wavelengths is generally used.

Laser power and total intensity

Tympanic membrane which is first terminal of light for hearing treatment could be damaged depending on the total power of light. Since total power is composed with laser power, beam spot size, and irradiation time, appropriate parameters for each animal model is needed. Too strong power could lead pathological changes on the tympanic membrane and middle ear.

Irradiation and repetition time

Irradiation times of PBM in previous hearing researches are separated into two times (30 and 60 minutes). Since animal should be anesthetized during PBM irradiation, too long irradiation time is not appropriate for animal study. Furthermore, burning of tympanic membrane or pathological changes in the middle ear tissue caused by long duration of irradiation could occur in animal model.

![Fig. 1. Laser irradiation in the ear. Laser reaches to the cochlea through several obstacles. Curvy ear canal and tympanic membrane should be considered for photobiomodulation (PBM) in hearing research.](image-url)
EFFECT OF PBM IN HEARING RESEARCH

Several papers reported various regenerative and protective effect of PBM in hearing research. Information of researches with PBM and parameters of PBM is summarized in Table 1.

HAIR CELL PROTECTION

Rhee et al. (2012) firstly reported the protective effect of PBM on cochlear hair cell after traumatic noise exposure. PBM treated animal showed significant hearing threshold recovery with less hair cell loss in the middle turn of cochlear after 8 days of treatment. The wavelength and power of PBM in this study have been used as a standard PBM parameter for hearing research with animal model. Tamura et al. (2015) reported similar recovery of hearing threshold and similar hair cell protection in animal model with similar parameters of PBM in Rhee et al. (2012). Their further study found that these positive effect of PBM occurred though modulation of Reactive oxygen species (ROS) and activation of NF-kb. Furthermore, simultaneous PBM irradiations in both ears showed better recovery and protective effect then unilateral PBM in noise overexposed animal.

Protective effect of PBM on cochlear hair cell after ototoxic drug damage was also reported. Rhee et al. (2012) reported that survival of hair cell increased in PBM group after gentamycin treatment.

NEURONAL PROTECTION

Protective effect of PBM had reported in the neuronal structures in the cochlea. PBM using 808 nm protected damage of ribbon synapse between inner hair cell and spiral ganglion neuron against traumatic noise exposure. Number of post synaptic receptor were also significantly less damaged by neurotoxic drug application after PBM. Moreover, higher number of spiral ganglion neuron survived in neurotoxin with PBM group then neurotoxic damage only group.

POSSIBLE MECHANISM OF PBM IN HEARING RESEARCH

Molecular mechanisms of PBM can be categorized as chromophores, signaling molecules, and activation of transcription factors. Cytochrome c oxidase (Cox), which is the terminal enzyme of the electron transport chain, has known as typical chromophore related with PBM mechanism. Cox performs as a photo-receptor and transducer of photo-signals of light with red and near infrared regions. Reduction of oxygen in the Cox leading increment of mitochondrial membrane potential and the level of ROS and ATP as well. PBM can increases Cox activity to modulate cellular, resulting in reduction of nitrite.

There are several light sensitive ion channels in the cochlea. Transient receptor potential (TRP) channels have various isoforms with seven sub families. In the neurons, infrared light is able to generate laser-evoked neuronal voltage and TRPV4 channels were demonstrated to be the primary effectors of the chain reaction. Recent study supported this hypothesis by reporting the result that TRP channels involved in hair cell protection.

PBM also can activates signaling molecule. PBM can initiates mitochondrial ROS changes leading to activation of the transcription factor nuclear factor kappa B (NF-kB), which can sense the redox signaling. The fact that the addition of antioxidants inhibits the activation of NF-kB by 810 nm light reinforces this assumption. Protective of PBM in noise induced hair cell loss by NF-kB signaling was support this assumption.

Table 1. PBM treatment parameters of previous animal studies

| Laser type | Wavelength (nm) | Animal | Power (mW/cm²) | Irradiation time (minutes) | # of treatment (# per day) | Total laser energy (J/cm²) | Year | Reference number |
|------------|-----------------|--------|----------------|---------------------------|---------------------------|--------------------------|------|----------------|
| Diode laser | 830             | SD Rat | 200            | 60                        | 10 (1)                    | 7200                     | 2013 | [27]           |
| Diode laser | 808             | SD Rat | 110, 165       | 30                        | 5 (1)                     | 1980, 2970               | 2015 | [25]           |
| Diode laser | 830             | SD Rat | 200, 250, 300  | 30                        | 14 (1)                    | 10080, 12600, 15120      | 2016 | [26]           |
| Diode laser | 808             | SD Rat | 165            | 60                        | 7 (1)                     | 4158                     | 2019 | [28]           |
| Diode laser | 830             | SD Rat | 200            | 30                        | 7 (1)                     | 297                      | 2017 | [21]           |
| Diode laser | 830             | SD Rat | 100 to 165     | 60                        | 12 (1)                    | 4320 to 7128             | 2012 | [24]           |
| Diode laser | 808             | SD Rat | 165            | 30                        | 5 (1)                     | 1485                     | 2016 | [20]           |
| Diode laser | 808             | Gerbil | 200            | 60                        | 7 (1)                     | 4158                     | 2016 | [23]           |
| Diode laser | 808             | SD Rat | 165            | 60                        | 15 (1)                    | 8910                     | 2016 | [22]           |
CONCLUSIONS

PBM is a therapeutic approach with potency in hearing research. Non-invasive access without side effect of PBM therapy is the most efficient characteristic for clinical application. In this review, we summarized previous studies with PBM therapy in animal models. The success of PBM therapy in hearing research highly rely on the parameters of light source. For clinical application, more studies should be followed to address molecular mechanism of PBM therapy in the hearing area.

REFERENCES

1. Clijsen R, Brunner A, Barbero M, Clarys P, Taeymans J. Effects of low-level laser therapy on pain in patients with musculoskeletal disorders: a systematic review and meta-analysis. Eur J Phys Rehabil Med 2017;53:603-10.
2. Pires de Sousa MV, Ferraresi C, Kawakubo M, Kaipper B, Yoshimura EM, Hamblin MR. Transcranial low-level laser therapy (810 nm) temporarily inhibits peripheral nociception: photoneuromodulation of glutamate receptors, prostatic acid phosphatase, and adenosine triphosphate. Neuropehonetics 2016;3:015003.
3. Turhani D, Scheriau M, Kapral D, Benesch T, Jonke E, Bantleon HP. Pain relief by single low-level laser irradiation in orthodontic patients undergoing fixed appliance therapy. Am J Orthod Dentofacial Orthop 2006;130:371-7.
4. Heo JC, Park JA, Kim DK, Lee JH. Photobiomodulation (660 nm) therapy reduces oxidative stress and induces BDNF expression in the hippocampus. Sci Rep 2019;9:10114.
5. Dos Santos TC, de Brito Sousa K, Andreo L, Martinelli A, Rodrigues MFSD, Bussadori SK, et al. Effect of photobiomodulation on C2C12 myoblasts cultivated in M1 macrophage-conditioned media. Photochem Photobiol. In press 2020.
6. Goo H, Kim H, Ahn J, Cho KJ. Effects of low-level light therapy at 740 nm on dry eye disease in vivo. Med Laser 2019;8:50-58.
7. Mesquita-Ferrari RA, Martins MD, Silva JA Jr, da Silva TD, Piovesan RF, Pavesi VC, et al. Effects of low-level laser therapy on expression of TNF-α and TGF-β in skeletal muscle during the repair process. Lasers Med Sci 2011;26:335-40.
8. Lee HJ, Kim YK. Burn wound successfully treated with 830-nm light emitting diode phototherapy combined with epidermal growth factor solution. Med Laser 2019;8:94-6.
9. da Silva Oliveira VR, Cury DP, Yamashita LB, Esteca MV, Watanabe IS, Bergmann YF, et al. Photobiomodulation induces antinociception, recovers structural aspects and regulates mitochondrial homeostasis in peripheral nerve of diabetic mice. J Biophotonics 2018;11:e201800110.
10. Adams PF, Hendershot GE, Marano MA. Current estimates from the National Health Interview Survey, 1996. Vital Health Stat 10 1999,(200):1-203.
11. Cruickshanks KJ, Wiley TL, Tweed TS, Klein BE, Klein R, Mares-Perlman JA, et al. Prevalence of hearing loss in older adults in Beaver Dam, Wisconsin. The Epidemiology of Hearing Loss Study. Am J Epidemiol 1998;148:879-86.
12. Cooper JC Jr, Gates GA. Hearing in the elderly—the Framingham cohort, 1983-1985: part II. Prevalence of central auditory processing disorders. Ear Hear 1991;12:304-11.
13. Mościcki EK, Elkins EF, Baum HM, McNamara PM. Hearing loss in the elderly: an epidemiologic study of the Framingham Heart Study Cohort. Ear Hear 1985;6:184-90.
14. Agrawal Y, Platz EA, Niparko JK. Prevalence of hearing loss and differences by demographic characteristics among US adults: data from the National Health and Nutrition Examination Survey, 1999-2004. Arch Intern Med 2008;168:1522-30.
15. Wallhagen MI, Strawbridge WJ, Cohen RD, Kaplan GA. An increasing prevalence of hearing impairment and associated risk factors over three decades of the Alameda County Study. Am J Public Health 1997;87:440-2.
16. Dalton DS, Cruickshanks KJ, Klein BE, Klein R, Wiley TL, Nondahl DM. The impact of hearing loss on quality of life in older adults. Gerontologist 2003;43:661-8.
17. Goodman SS, Bentler RA, Dittberner A, Mertes IB. The effect of low-level laser therapy on hearing. ISRN Otolaryngol 2013;2013:916370.
18. Zazzo M. Pain threshold improvement for chronic hyperacusis patients in a prospective clinical study. Photomed Laser Surg 2010;28:371-7.
19. Salahaldin AH, Abdulhadi K, Najar N, Bener A. Low-level laser therapy in patients with complaints of tinnitus: a clinical study. ISRN Otolaryngol 2012;2012:132060.
20. Teggi R, Bellini C, Piccioni LO, Palonta F, Bussi M. Transmeatal low-level laser therapy for chronic tinnitus with cochlear dys-function. Audiol Neurootol 2009;14:115-20.
21. Lee JH, Kim S, Jung JY, Lee MY. Applications of photobiomodulation in hearing research: from bench to clinic. Biomed Eng Lett 2019;9:351-8.
22. Tamura A, Matsunobu T, Tamura R, Kawauchi S, Sato S, Shiotani A. Photobiomodulation rescues the cochlea from noise-induced hearing loss via upregulating nuclear factor κB expression in rats. Brain Res 2016;1646:467-4.
23. Lee MY, Hyun JH, Suh MW, Ahn JC, Chung PS, Jung JY, et al. Treatment of peripheral vestibular dysfunction using photobiomodulation. J Biomed Opt 2017;22:1-7.
24. Lee JH, Chang SY, Mow WJ, Oh C, Kim SH, Rhee CK, et al. Simultaneous bilateral laser therapy accelerates recovery after noise-induced hearing loss in a rat model. PeerJ 2016;4:e2252.
25. Lee MY, Bae SH, Chang SY, Lee JH, Kim SH, Ahn JC, et al. Photobiomodulation by laser therapy rescued auditory neuropathy.
induced by ouabain. Neurosci Lett 2016;633:165-73.

26. Rhee CK, Bahk CW, Kim SH, Ahn JC, Jung JY, Chung PS, et al. Effect of low-level laser treatment on cochlea hair-cell recovery after acute acoustic trauma. J Biomed Opt 2012;17:068002.

27. Tamura A, Matsunobu T, Mizutari K, Niwa K, Kurioka T, Kawachi S, et al. Low-level laser therapy for prevention of noise-induced hearing loss in rats. Neurosci Lett 2015;595:81-6.

28. Moon TH, Lee MY, Jung JY, Ahn JC, Chang SY, Chung PS, et al. Safety assessment of trans-tympanic photobiomodulation. Lasers Med Sci 2016;31:323-33.

29. Rhee CK, He P, Jung JY, Ahn JC, Chung PS, Lee MY, et al. Effect of low-level laser treatment on cochlea hair-cell recovery after ototoxic hearing loss. J Biomed Opt 2013;18:128003.

30. Lee JH, Lee MY, Chung PS, Jung JY. Photobiomodulation using low-level 808 nm diode laser rescues cochlear synaptopathy after acoustic overexposure in rat. J Biophotonics 2019;12:e201900145.

31. Karu TI. Multiple roles of cytochrome c oxidase in mammalian cells under action of red and IR-A radiation. IUBMB Life 2010;62:607-10.

32. Wu S, Zhou F, Wei Y, Chen WR, Chen Q, Xing D. Cancer phototherapy via selective photoinactivation of respiratory chain oxidase to trigger a fatal superoxide anion burst. Antioxid Redox Signal 2014;20:733-46.

33. Poyton RO, Ball KA. Therapeutic photobiomodulation: nitric oxide and a novel function of mitochondrial cytochrome c oxidase. Discov Med 2011;11:154-9.

34. Nilius B, Voets T. TRP channels: a TR(I)P through a world of multifunctional cation channels. Pflugers Arch 2005;451:1-10.

35. Albert ES, Bec JM, Desmadryl G, Chekroud K, Travo C, Gaboyard S, et al. TRPV4 channels mediate the infrared laser-evoked response in sensory neurons. J Neurophysiol 2012;107:3227-34.

36. Wang S, Geng Q, Hua L, Ma Y, Gao Y, Zhang W, et al. Transient receptor potential cation channel subfamily vanilloid 4 and 3 in the inner ear protect hearing in mice. Front Mol Neurosci 2019;12:296.

37. de Freitas LF, Hamblin MR. Proposed mechanisms of photobiomodulation or low-level light therapy. IEEE J Sel Top Quantum Electron 2016;22:7000417.

38. Chen AC, Arany PR, Huang YY, Tomkinson EM, Sharma SK, Kharkwal GB, et al. Low-level laser therapy activates NF-kB via generation of reactive oxygen species in mouse embryonic fibroblasts. PLoS One 2011;6:e22453.