

How to Cite:

Dabbous, O. A., Morsy, M., Abdelazeim, F. H., & Said, R. S. E. (2022). Effect of laser bio-stimulation with a wavelength of 632.8 on children with spastic cerebral palsy. *International Journal of Health Sciences*, 6(S4), 12684–12696.
<https://doi.org/10.53730/ijhs.v6nS4.12254>

Effect of laser bio-stimulation with a wavelength of 632.8 on children with spastic cerebral palsy

Ola A. Dabbous

Medical Department, National Institute of Laser Enhanced Sciences (NILES),
Cairo University, Giza, Egypt

Mona Morsy

Medical Department, National Institute of Laser Enhanced Sciences (NILES),
Cairo University, Giza, Egypt

Faten H. Abdelazeim

Professor, Department of Physical Therapy for Pediatrics, Faculty of Physical
Therapy, Cairo University and Dean, Faculty of Physical Therapy, October 6
University, Giza, Egypt

Rowida S. El Said

Benha Fever Hospital, Benha, Egypt

Corresponding author email: dr.rowida_pt@yahoo.com

Abstract---Cerebral Palsy (CP) defined as lasting clutters gather of development and posture advancement which is the cause of activity confinement that are credited to non-dynamic unsettling influences which happened in the developing fetal or newborn child brain. Concurring to European information, the average recurrence of CP is 2.08 per 1000 live births. Study proposed to confirm low laser acupuncture impact with wavelength 632.8 nm. on immunity improvement in children with spastic CP. This study was conducted on 40 children with spastic CP both sexes from 1 to 3 years old. They were chosen from the outpatient clinic of pediatric in the National Institute of Laser Enhanced Sciences (NILES) and physical therapy outpatient clinic at Benha Teaching Hospital. They were divided randomly into two groups equal in number: Group A: (Control group) received physiotherapy session only lasted for an hour for 3 sessions / week for 12 weeks. Group B: (study group): received laser acupuncture with wavelength 632.8 nm and power output: 50 mw for 3 sessions / week for 12 weeks as well previous program of physical therapy. Flow cytometry "BD Facscalibur" two-colure was used for gauge of natural killer cells for expression (CD₁₆ CD₅₆) for all children

of both groups before and after the treatment. The results were statistically analyzed, as it was concluded that there is a difference between post treatment measures for group A and B showed significant difference in natural killer cells count (CD₁₆, CD₅₆) in favor of group B. This study concluded to modify the immune response for the better in children with spastic CP after using of laser acupuncture therapy.

Keywords---spastic cerebral palsy, natural killer cells, low level laser therapy 632.8 nm, laser acupuncture, flow cytometry, CD16 CD56.

Introduction

Severe neuronal growth and normal myeline disorder that is caused by brain injury in newborns generates life-persisting sensory-motor and cognitive dereliction in the growing child with large variety of neurological symptoms, under the umbrella of cerebral palsy (Lambrecht et al., 2014). Obviously, the cause of the syndromes' symptoms of the CP is brain development disorder (Morgun et al., 2013). One theory discussed that cerebral palsy pathogenesis in children contains two major interconnected mechanisms. One of these mechanisms is: there is a damping defect in the cerebral nervous tissue related to ischemia and impairment of the regulation apoptotic cell death (Sun et al., 2012). Another concept, formation of auto reactivity against impaired nervous tissue in CP children due to fragility of immunity system regulation (Ortega et al., 2015). Natural killer cells (NK) defined as a sort of cytotoxic lymphocyte important for the innate immune system (Vivier et al., 2011). Low level laser therapy (LLLT) has altogether impact on blood pictures that shown the expanding in CD₄₅ lymphocytes and NK (CD₁₆ CD₅₆) (Mustafa et al., 2016). According to Mori et al., (2013) the expanded NK-cell activity in ordinary rats caused by permanent stimulation of acupuncture on ST 36 (*Zusanli*) one time per day for three days without affecting on spleen NK-cell populaces. Analysts proposed that increment of NK-cell activity was interceded by increments of interferon- γ (IFN- γ) and β -endorphin that were happened because of Electro Acupuncture. One of the preferred acupuncture points that have an effect on modifying the immune system response is called ST 36 that used as an analgesic effect. (Cabýoglu et al., 2006). Through the previous study, we confirmed the impact of laser acupuncture with wave length 632.8nm on modification of immunity response to the better in children suffered from spastic cerebral palsy.

Subjects

Prospective clinical study was conducted from March 2021 to February 2022 in the pediatric outpatient clinic of the National Institute of Laser Enhanced Science (NILES) and physical therapy outpatient clinic at Benha Teaching Hospital on a sample of "forty children with spastic CP" of both sexes.

Inclusive criteria

All children were selected as the following:

1. Age range between 1 to 3 years old.
2. According to referral from the pediatrician, all children diagnosed with spastic CP.
3. According to Modified Ashworth Scale, their muscle tone ranged from 1 to up to 1+
4. All Children were rated at level 4 of gross motor function classification system (GMFCS)

Exclusive criteria

Children were excluded from the study for the following criteria:

1. Suffering from any deformity or any problem at back region.
2. Receiving irradiation therapy for Cancer treatment.
3. Knee joint deformity.
4. Cardiovascular or respiratory problems like pneumonia.
5. Receiving steroid or antibiotics medications.
6. Epilepsy.
7. Genetic syndrome.

Ethics

All parents of selected children before starting the study gave a written informed consent, according to the scientific, ethical committee rules of the National Institute of Laser Enhanced Sciences and of Benha Teaching Hospital.

Subjects were divided randomly into two groups equal in number and treatment duration for 12 weeks

Group A (control group) children received one hour physical therapy session per day only for 3 sessions per week.

Group B (study group) children received laser acupuncture with Wavelength: 632.8nm on immunity acupoint stomach 36 (ST₃₆ in Chinese Zusanli) and another acupuncture points: BL₁₈ (Gan Shu), BL₂₀ (Pi Shu) and BL₂₃ (Shen Shu) for three sessions per week as well as the previous program of physical therapy.

Procedures

Evaluation procedures

Flow cytometry "BD Facscalibur" two-colour was used for gauge of natural killer cells for expression of (CD₁₆ CD₅₆) for all children in both groups before and after treatment. Each child was evaluated for blood analysis for gauging natural killer cells by analyzing expression of CD₁₆⁺CD₅₆⁺ (arterial blood sample was by the drawn from radial artery by the chemist).

Treatment procedures

Physical therapy program

It was conducted to both control and study groups. The program was based on Neurodevelopmental treatment (NDT) approach with emphasis on improving both motor functions and coordination in form of stretches exercises, range of motion exercises, strengthening exercises, flexibility and endurance exercises. Also, the program was based on improvement of standing balance and walking.

Laser acupuncture

Low energy He-Ne Laser used for Laser Acupuncture to stimulate acupuncture points of immunity for all children of group B only. Laser parameters applied are shown in table 1.

Table (1): Laser parameters applied.

Irradiation parameter	Unite of measurement
Wavelength	632.8 nm
Power output	50 mw
Beam spot diameter	1 cm
Dose	3.0 J/cm ²
Treatment time /point	60 sec.
Number of points	8 points
Treatment interval	3 sessions / week

The probe was applied on immune acupoints as shown in figure (1).

- The acupoint ST36 was determined below patella and lateral to anterior crest of tibials anterior muscle bilateral.
- BL 18 (Gan Shu) was determined at back, lateral to lower border of spinous process of 9th thoracic vertebrae bilateral.
- BL 20 (Pi Shu) was determined at back, lateral to the lower border of spinous process of 11th thoracic vertebrae bilateral.
- BL 23 (Shen Shu) was determined at back, lateral to the lower border of spinous process of 2nd lumbar vertebrae bilateral.



Fig. (1): Case No. 10 (study group).

Results

He-Ne Laser Acupuncture on immunity indices in children with spastic CP was investigated. These results were intended for the collected data of CD16, CD56 and CD16 CD56 for each child in the two groups (A and B).

CD16

Compare between before and after treatment mean values of CD16 in groups (A and B)

The pre- treatment $\pm$ SD of group (A) value was 58.73 ± 14.71 , while that of post-treatment was 65.87 ± 10.75 and the significant was presented ($p= 0.001$) between before and after treatment mean values and % of change = 12.16 %. The pre- treatment $\pm$ SD of group (B) value was 56.63 ± 9.17 and post- treatment was 72.87 ± 8.05 and the significant was presented ($p= 0.0001$) between before and after treatment mean values and % of change = 28.68 % (table 2) and (figure 2).

Table (2): Compare between before and after treatment mean values of CD16 in groups (A and B)

Items	CD16			
	Group (A)		Group (B)	
	Pre-treatment	Post-treatment	Pre-treatment	Post-treatment
Mean $\pm$ SD	58.73 ± 14.71	65.87 ± 10.75	56.63 ± 9.17	72.87 ± 8.05
MD	7.14		16.24	
% of change	12.16 %		28.68 %	
t-value	3.87		14.26	
p-value	0.001		0.0001	
Level of Significant	S		S	

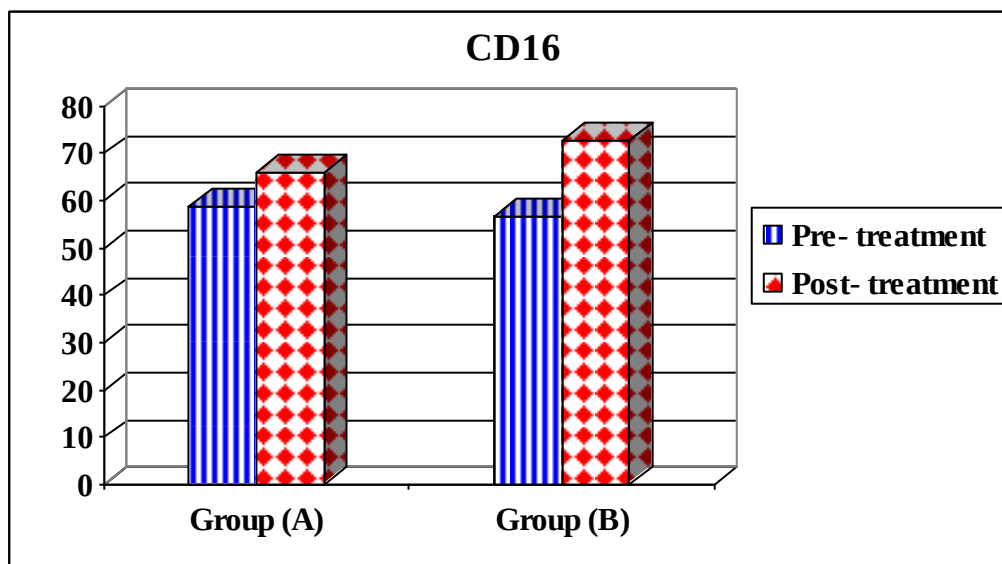


Figure (2): Compare between before and after treatment mean values of CD16 in groups (A and B)

After treatment mean values of CD16 between groups (A and B)

When comparing the post- treatment $\pm$ SD values between groups (A and B) were 65.87 ± 10.75 and 72.87 ± 8.05 respectively, the significant difference was revealed ($p= 0.0001$) in favor of group (B) and % of change was 10.36 % (table 3) and (figure2).

Table (3): After treatment mean values of CD16 between groups (A and B)

Items	CD16 (Post- treatment)	
	Group (A)	Group (B)
Mean $\pm$ SD	65.87 ± 10.75	72.87 ± 8.05
MD	7	
% of change	10.36 %	
t-value	2.33	
p-value	0.026	
Level of Significant	S	

CD56

Compare between before and after treatment mean values of CD56 in groups (A and B)

The pre- treatment $\pm$ SD of group (A) value was 13.66 ± 4.8 , while that of post-treatment was 17.68 ± 5.39 and the significant was presented ($p= 0.0001$) between pre and post- treatment mean values and % of change = 29.42 %. The

pre- treatment $\pm$ SD of group (B) value was 14.76 ± 3.59 , while that of post-treatment was 22.09 ± 3.68 and the significant was presented ($p= 0.0001$) between pre and post- treatment mean values and % of change = 49.66 % (table 4) and (figure 4).

Table (4): Compare between before and after treatment mean values of CD56 in groups (A and B)

Items	CD56			
	Group (A)		Group (B)	
	Pre- treatment	Post- treatment	Pre- treatment	Post-treatment
Mean $\pm$ SD	13.66 ± 4.8	17.68 ± 5.39	14.76 ± 3.59	22.09 ± 3.68
MD	4.02		7.33	
% of change	29.42 %		49.66 %	
t-value	6.07		16.59	
p-value	0.0001		0.0001	
Level of Significant	S		S	

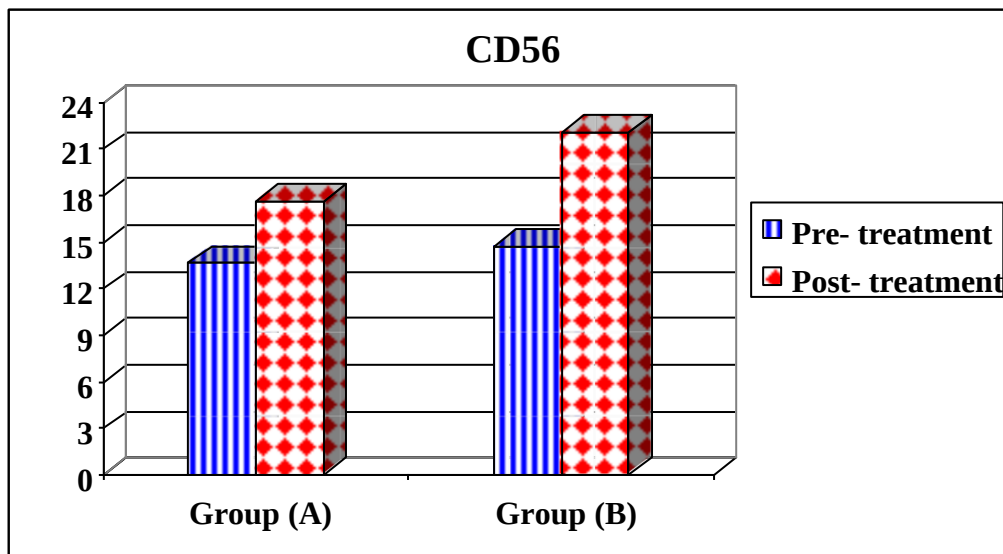


Figure (4): Compare between before and after treatment mean values of CD56 in groups (A and B)

After treatment mean values of CD56 between groups (A and B)

When comparing the pre- treatment $\pm$ SD values between groups (A and B) were 17.68 ± 5.39 and 22.09 ± 3.68 respectively, the significant difference was revealed ($p= 0.005$) in favor of group (B) and % of change was 24.94 % (table 5) and (figure 4).

Table (5): After treatment mean values of CD56 between groups (A and B)

Items	CD56 (Post- treatment)	
	Group (A)	Group (B)
Mean $\pm$ SD	17.68 $\pm$ 5.39	22.09 $\pm$ 3.68
MD	4.41	
% of change	24.94 %	
t-value	3.03	
p-value	0.005	
Level of Significant	S	

CD16 CD56**Compare between before and after treatment mean values of CD16 CD56 in groups (A, B and C)**

The pre- treatment $\pm$ SD of group (A) value was 3.77 ± 2.37 , while that of post-treatment was 5.32 ± 3.1 and the significant was presented ($p= 0.0001$) between pre and post- treatment mean values and % of change = 20.56 %. The pre-treatment $\pm$ SD of group (B) value was 3.59 ± 1.96 , while that of post- treatment was 7.69 ± 1.7 and the significant was presented ($p= 0.0001$) between pre and post- treatment mean values and % of change = 57.1 % (table 6) and (figure 6).

Table (6): Compare between before and after treatment mean values of CD16 CD56 in groups (A and B)

Items	CD16 CD56			
	Group (A)		Group (B)	
	Pre-treatment	Post-treatment	Pre-treatment	Post-Treatment
$\bar{X} \pm SD$	3.77 ± 2.37	5.32 ± 3.1	3.59 ± 1.96	7.69 ± 1.7
MD	1.55		4.1	
% of change	20.56 %		57.1 %	
t-value	4.76		4.91	
p-value	0.0001		0.0001	
Level of Significant	S		S	

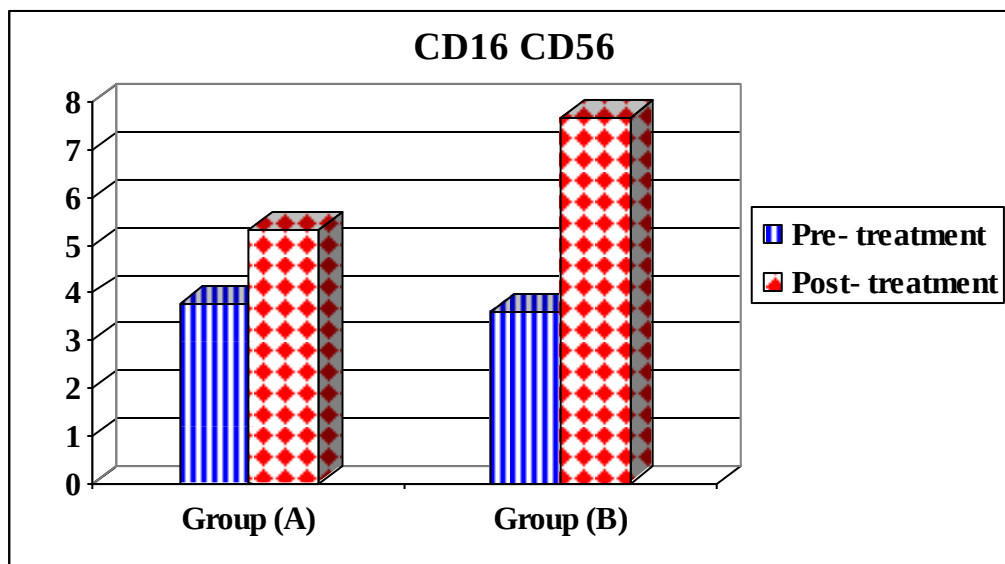


Figure (6): Compare between before and after treatment mean values of CD16 CD56 in groups (A and B)

After treatment mean values of CD16 CD56 between groups (A and B):

When comparing the pre- treatment $\pm$ SD values between groups (A and B) were 5.32 ± 3.1 and 7.69 ± 1.7 respectively, the significant difference was revealed ($p=0.006$) in favor of group (C) and % of change was 44.54 % (table 7) and (figure 6).

Table (7): After treatment mean values of CD16 CD56 between groups (A and B)

Items	CD16 CD56 (Post- treatment)	
	Group (A)	Group (B)
Mean $\pm$ SD	5.32 ± 3.1	7.69 ± 1.7
MD	2.37	
% of change	44.54 %	
t-value	3	
p-value	0.006	
Level of Significant	S	

Discussion

The spastic cerebral palsy children whom participated in the current study found their collected data of CD16 ranged from 58.73 to 56.63, CD56 ranged from 13.66 to 14.76 and CD16 CD56 ranged from 3.77 to 3.59 that means weak immunity. It may be explained by Thornton et al., (2012) the pathogenesis of cerebral parlay development in children happens through two major interrelated mechanisms. First mechanism: There's serious impairment of homeostasis within cerebral

nervous tissue auxiliary to interruption of blood flow and dis-regulation of apoptotic cell passing. Additionally, Ortega et al., (2015) found that second mechanism: As a result of the arrangement of auto reactivity against harmed nervous tissue, the immune dis-regulation occurs. From the previous studies, there are confirming thoughts from current study data about of cerebral paralysis pathogenesis and the improvement.

In other field, Fujimaki et al., (2003) conducted a think about looking at the impacts of LLLT on generation of receptive oxygen species (ROS) by human neutrophils. LLLT is a compelling helpful methodology for inflammatory conditions. An infrared diode laser (GaAlAs) with 830nm, 150mw/cm² was utilized for therapy. After illumination, ROS generation was measured by neutrophils and CD₁₆ on neutrophil surface by flow cytometer. The constricting impact of LLLT was bigger in neutrophils of smokers than non-smokers.

Improving of immune system in cerebral palsy children was confirmed by the physiological regulation of nervous tissue homeostasis; include better comprehending of the close interaction between nervous, endocrine, and immune systems. In the central nervous system, immune system is represented by 3 morphologically and functionally various cell factors are lymphoid cells of cerebrospinal fluid; non-lymphoid cells of the nervous tissue and endothelial cells of cerebral vessels. As well, crossing of the blood-brain barrier penetrating into the CNS and providing immunological surveillance are made by activated T-cells. The most modern concepts with cerebral palsy children in subject of physiological arrangement of limitations of blood flow of the nervous tissue, due to close relationship between the nervous and immune system demonstrated by Engelhardt & Liebner, (2014).

However Novoselova et al., (2006) demonstrated the impact of He-Ne laser with wavelength of 632.8 nm on splenic lymphocytes and macrophages for less than 60 seconds of introduction period, watched the incitement in interleukin-2 and nitric oxide generation, beside an increment within action of NK cells. While prolonged period of introduction more than 180 seconds, NO generation and natural killer cell activity diminished, but IL-2 generation was not diverse from the control level. No change occurred in interferon-gamma generation after laser light introduction of cells for sixty or one hundred and eighty second.

In current study, there was increasing of NK cells counts in expression of CD16 CD56 in children with cerebral palsy come in agreement with Ezzo et al., 2006, who reported that acupuncture has a main role for increasing natural killer cells counts. For this cause, the Specialist may use it. Increasing of natural killer cells count is occurred by improving the pathway between immunity system and neurotransmitter network. Smyth et al., (2002) found that acupuncture improves NK Cell level and its functions, as NK Cell defines as an immune cell which plays a main role in regulating anti-cancer immune functions. Regarding to Lundgren et al., (2012) Laser radiation at 632 and 830 nm wavelengths is considered one of the preferred procedures for immunity system .because of creating an immune-invigorating impact; this is why the low laser radiation at wavelength 632.8nm in the current study was used to improve immunity.

The improvement of children immunity due to application of laser may be explained by the immune reaction is made up series of cells, tissues and atoms in which they act as a defense against pathogenic microorganisms. Occurrence of broken immune reactions from Immune system causes the individual defenseless to infection, bacterial and parasitic. In fact, Immune cells have ability to occur harm. The majority of famous infections are caused by unregulated immune cells, as confirmed by Nitish & Sudeep, 2011.

Laser Acupuncture (LA) Considers one of the preferred technique that applied to specific patients like pediatric because of noninvasive, painless, with less side effects techniques. In addition to the sessions duration of LA from ten to sixty seconds per acupuncture point that is shorter than with metal needle duration from ten to thirty minutes (Hu et al., 2014). Low level laser acupuncture at wavelength 632.8nm improved the immunity in children with cerebral palsy by increasing natural killer cells counts. As confirmed by El said et al. that laser acupuncture therapy at wavelength of 780nm increased the natural killer cells count which improved immunity indices in children who suffered from spastic cerebral palsy.

Mackanos & Contag, (2011) added the improvement of immune system may be due to photo energized advancement of interleukin-2 with nitric oxide and expanded movement of NK cells taking after exposure of He-Ne 632.8nm laser light. Low-power laser treatment has moreover appeared to operate on T-lymphocytes in an assortment of ways from both seen light and infrared, actuating lighted cells to more prominent action. From the previous we can say that infrared LLLT increments both human phagocytic and chemotactic advancement in human tumor cells. LLLT has an energizing effect within immune biological treatment of immune system maladies additionally invigorating and improving ordinary reaction of immune system cells to decide by outside living beings.

Conclusion

Modification of the immune response for the better in spastic CP children after using of LA therapy with wavelength 632.8 nm and power output: 50 mw on immune acupoint ST₃₆ and another acupuncture points: BL₁₈ (Gan Shu), BL₂₀ (Pi Shu) and BL₂₃ (Shen Shu) improved immunity indices by increasing of natural killer cells count for expression of "CD₁₆, CD₅₆ "in children with cerebral palsy without any other complain. The results were statistically analyzed, as it was concluded that there is a difference between post treatment measures for group A and B showed significant difference in natural killer cells count (CD₁₆, CD₅₆) in favor of group B.

References

- Ahmed, A. F., Olama, K. A., Zaky, N. A., & Abdelfadil, A. R. (2022). Oro-motor skills and gross motor abilities in children with diplegic cerebral palsy following electrical stimulation. *International Journal of Health Sciences*, 6(S1), 2171–2179. <https://doi.org/10.53730/ijhs.v6nS1.5144>

- Cabýoglu MT, Ergene N and Tan U (2006). " The Mechanism of Acupuncture and Clinical Applications". *Int.J. Neurosci.*:116(2):115–125.
- Engelhardt B & Liebner S (2014). Novel insights into the development and maintenance of the blood–brain barrier. *Cell Tissue Res* 355(3):687–699.
- Ezzo JM, Richardson MA and Vickers A(2006)."Acupuncture-Point Stimulation for Chemotherapy-Induced Nausea or Vomiting". *Cochrane Database of Systematic Reviews*. (2) Article ID CD002285.
- Fujimaki Y, Shimoyama T, Liu Q, Umeda T, Nakaji S, and Sugawara K (2003)." Department of Hygiene, Hirosaki University School of Medicine, Japan. Low-level laser irradiation attenuates production of reactive oxygen species by human neutrophils". *J Clin Laser Med Surg*. Jun. 21(3): 165-170.
- Hu WL, Chang CH, Hung YC, Tseng YJ, Hung IL and Hsu SF (2014). " Laser Acupuncture Therapy in Patients with Treatment-Resistant Temporomandibular Disorders. *PLoS One*". Crossref, Medline. 9(10): e110528.
- Lambrecht S, Urrea O, Grosu S and Nombela SP (2014). Emerging rehabilitation in cerebral palsy. *Emerging Therapies in Neurorehabilitation Biosystems & Biorobotics* 4:23–49)
- Lundgren K, Brown M, Pineda S, Cuzick J, Salter J, Zabaglo L, Howell A, Dowsett M and Landberg G(2012). Effects of cyclin D1 gene amplification and protein expression on time to recurrence in postmenopausal breast cancer patients treated with anastrozole or tamoxifen: a TransATAC study. *Breast Cancer Research* 14:R57.
- Mackonos MA & Contag CH (2011). Pluse duration determine level of Hsp 70 induction in tissuses following laser irradiation .*J.Biomed Opt.*, 7:078002.
- Morgun AV, Kuvacheva NV, Taranushenko TE, Khilazheva ED, Malinovskaia NA, Gorina IV, et al. (2013). Current concepts of perinatal ischemic injury in the brain neurovascular unit: molecular targets for neuroprotection. *Vestn Ross Akad Med Nauk* 12:26–35
- Mori H, Kuge H, Tanaka TH, Taniwaki E, Hanyu K and Morisawa T (2013)". Effects of Acupuncture Treatment on Natural Killer Cell Activity, Pulse Rate, and Pain Reduction for Older Adults: An Uncontrolled, Observational study". *J Integr Med.*:11(2):101–105
- Mustafa S, Al Musawil MS, Jaafar1 B, Al-Gailani M, Ahmed FMS, and Nursakinah S (2016). "Effects of Low-Level Laser Irradiation on Human Blood Lymphocytes in vitro". *Laser Med. sci.* 10:213.
- Nitish,CH and Sudeep,B.(2011)." Laser Basic Principle and Classification".*JRAP*,2:132-141.
- Novoselova EG, Cherenkov DA, Glushkova OV, Novoselova TV, Chudnovskii VM, Iusupov VI and Fesenko EE (2006). Effect of low-intensity laser radiation (632.8 nm) on immune cells isolated from mice. *Biofizika*. 51(3):509-18. Russian. PMID: 16808352.
- Ortega SB, Kong X, Venkataraman R, Savedra AM, Kernie SG, Stowe AM and Raman L (2015). "Perinatal Chronic Hypoxia Induces Cortical Inflammation, Hypomyelination, and Peripheral Myelin-Specific T Cell Autoreactivity". *J. Leukoc. Biol.* 99(1):21–29.
- Rowida S. El said, Ola A. Dabbous, Mona Morsy and Faten Abdelazeim." The Improvement of Immunity Indices in Children with Cerebral Palsy after Laser Acupuncture Biostimulation".previous work in process.

- Smyth MJ, Hayakawa Y, Takeda K and Yagita H (2002). "New Aspects of Natural-Killer-Cell Surveillance and Therapy of Cancer". *Nature Reviews Cancer*. 2(11):850–861.
- Sun Y, Zhang Y, Wang X, Blomgren K and Zhu C (2012). Apoptosis-inducing factor downregulation increased neuronal progenitor, but not stem cell, survival in the neonatal hippocampus after cerebral hypoxia-ischemia. *Mol Neurodegener* 7:17
- Suryasa, I. W., Rodríguez-Gámez, M., & Koldoris, T. (2021). Health and treatment of diabetes mellitus. *International Journal of Health Sciences*, 5(1), i-v. <https://doi.org/10.53730/ijhs.v5n1.2864>
- Thornton C, Rousset CI, Kichev A, Miyakuni Y, Vontell R and Baburamani AA (2012). "Molecular Mechanisms of Neonatal Brain Injury". *Neurol. Res. Int.* Article ID 506320, 16 p.
- Vivier E, Raulet DH, Moretta A, Caligiuri MA, Zitvogel L, Lanier LL, Yokoyama WM and Ugolini S (2011). "Innate or adaptive immunity? The example of natural killer cells". *Science*. 331 (6013): 44–9.
- Wibawa, I. M. S., & Putra, I. K. (2021). Design and manufacture of air quality measurements based on arduino ATmega 2560 using dust ZH03A laser sensor. *International Journal of Physical Sciences and Engineering*, 5(1), 8–15. <https://doi.org/10.29332/ijpse.v5n1.800>