The effect of infrasonic wave exposure (8-12 Hz) on the immune system (immunoglobulin M and G) in mice

**Abstract.** This research method uses infrasonic waves (8-12 Hz) in the form of infrasound waves, murottal al-Quran, and recitation of the Alquran verses. Immune system testing uses the hemagglutination method of antibody titers by observing the amount of Immunoglobulin M (IgM) and Immunoglobulin G (IgG) on the sixth day and eleventh day. Testing of mice blood serum is done by adding the same antigen namely goat red blood cells. Interactions between antigens and antibodies cause secondary reactions, namely in the form of agglutination or precipitation because antigens are insoluble small particles. The results showed that infrasonic wave exposure (8-12 Hz) affected the amount of Immunoglobulin G (IgG) but did not affect the amount of Immunoglobulin M (IgM). The results of statistical analysis of ANOVA test with a significance level of 5% showed no effect of infrasonic wave exposure (8-12 Hz) on immunoglobulin M (IgM) with a significance of 0.098. For Immunoglobulin G (IgG) there is an effect with a significance of 0,000 after that, then continued with the Duncan test which shows the highest value (3.1800) in the al-Quran reading treatment. Infrasonic waves (8-12 Hz) can increase immunoglobulin G (IgG).

1. Introduction

In approving life, talking about health is very important. Various diseases, most of which are infectious diseases carried by various types of microbes such as viruses, bacteria, parasites, and fungi. The body has the ability to overcome disease to a certain extent. For example, if an infection is released it will increase fever or increase body temperature, by increasing body temperature to form a protection from the body's defense system [1][2].

 The immune system (immune system) is a system in the body, both human and animal, which has the ability to recognize an object to the body and body then will respond in the form of neutralization, eliminate or include in the process, with the help of a favorable, or cause Damage to tissue damage [3][4].

 Various ways are done by humans to increase the immune system that is given regular exercise, getting enough sleep, avoiding stress or being approved for peace. In a calm situation, someone is in alpha brain waves (8-12 Hz). There are many ways to achieve defense by translating. In the Islamic language there is no known translation term, but Islam which advocates peace by means of dzikir. Dzikir here is realized by reading al- Quran [5][11].

ٱلَّذِينَ ءَامَنُواْ وَتَطۡمَئِنُّ قُلُوبُهُم بِذِكۡرِ ٱللَّهِۗ أَلَا بِذِكۡرِ ٱللَّهِ تَطۡمَئِنُّ ٱلۡقُلُوبُ.

 *“Those who have believed and whose hearts are assured by the remembrance of Allah. Unquestionably, by the remembrance of Allah hearts are assured." (QS. Ar Ro’d: 28).*

 Published research shows that it can discuss the brain in alpha (infrasonic) waves [5]. Transcendental (TM) meditation can significantly reduce the risk of death and stroke in coronary heart patients. Alpha waves are one of five categories of brain waves. Brain waves are electrical wave patterns found in everyone's brain. Alpha frequency is a condition in which the human brain is relaxed, calm but still in a conscious state. When the brain in alpha waves will produce the hormone serotonin and endorphin which causes a person to feel comfortable, calm, and happy. Serotonin and endorphin hormones can also increase body immunity, dilate blood vessels, stabilize heart rate and sharpen the senses [2][5]. In connection with the above, it is necessary to conduct research to determine the effect of infrasonic waves (8-12Hz) on immunity (Immunoglobulin M and Immunoglobulin G) in mice.

1. Method

This study used a randomized block design with 4 treatments 5 repetitions. The treatment of infrasound waves is in the form of reading verses of the Qur'an, murottal al-Quran, and the presentation of alpha (infrared) waves (CD alpha waves).

 The experimental animals used were male mice with a body weight of about 25-30 grams, 240 animals were divided into 4 groups, namely control, infrasonic wave transmission (CD alpha waves), reading verses of the Qur'an, and murottal al- Qur'an. Each group was given 10 mice (5 for M Immunoglobulin and 5 for Immunoglobulin G), each of which was treated with infrasonic wave exposure. In order to get valid data, it is repeated five times. In this study using male mice because they cannot be influenced by hormonal cycles that can affect the results of the study. The results of the study are also influenced by biological biology of experimental animals such as body weight, age, and environmental conditions.

Testing of mice blood serum was carried out by adding the same antigen namely 2% goat red blood cells (HRK). Preparation of 2% HRC suspension by means of 1 mL of goat's blood collected in a clean and dried tube containing 1 mg EDTA which functions as an anticoagulant. Then centrifuged at a speed of 1500 rpm to separate goat red blood cells (HRK) from the plasma. Goat red blood cells obtained were washed with PBS in a tube, then the tube was turned back and forth several times, then centrifuged again. Washing is done at least 3 times. After being centrifuged, PBS was separated so that the remaining HRK was 100%, then PBS was added with the same amount until a 50% HRK suspension was obtained, then as much as 0.4 mL was diluted with 9.6 mL PBS to obtain antigen suspension with 2% HRH concentration.

 After the antigen is obtained and then injected into each group of mice, the exposure is carried out 30 minutes every day. On the sixth day, observation of immunoglobulin, M activity was carried out and on the eleventh-day observation of immunoglobulin, G activity using the hemagglutination method. Hemagglutination test, Serum obtained is then diluted by "double dilution" with PBS with a ratio of 1/4, 1/8, 1/16, 1/32, 1/64, 1/128. From each of these comparisons piped 50 µL into 6 microtiters (well plate 96) plate wells after which 50 µL of goat red blood cell suspension was added 2% to each well and shaken for 5 minutes to be homogeneous. Then it was incubated at 37ºC for 60 minutes and left 24 hours at room temperature. After that, the highest dilution was observed from each blood serum of male mice which still could agglutinate goat red blood cells.

Data collection and analysis, data obtained from observations of the highest blood serum dilution of male mice that still showed agglutination from goat red blood cells were collected and converted using the [2log (titer) +1] formula. Furthermore, the data were analyzed statistically using factorial ANOVA analysis with a significance level of 5% to determine whether there was an effect of exposure to infrasonic waves (8-12 Hz) on immunity (Immunoglobulin M and Immunoglobulin G) in mice.

1. Result and Discussion

Immunoglobulin (antibody) is a molecule produced by B lymphocytes and macrophages that are stimulated by foreign antigens. Antigens used to be induced by antibody production are goat red blood cells (HRK) which are immunogens, namely antigens derived from the genes of other species. SDMK is a polyvalent antigen which is a protein with a greater potential determinant compared to monovalent antigens. The more foreign the antigen is used, the more effective it is to produce an immune response. This antigen is injected into the body of the mouse intraperitoneally before being exposed. This immunization is intended to give fish an immune response in experimental animals [7] .

 Interactions between antigens and antibodies cause secondary reactions, namely in the form of agglutination or precipitation because antigens are insoluble small particles. Clots formed between specific antigens and anti-serum will unite and eventually settle as large lumps and are easily visible with the liquid above remaining clear. This occurs because antibodies generally have more than one antigen binding receptor so that the antibodies react with other antigen molecules that may have bound to one of the antibody molecules and form a lump [1].

 On the 6th day, or 5 days after HRK induction, the blood of the mouse is taken intracardiac to observe IgM activity and 11th day for IgG. During this period, it is expected that B cell sensitization will proliferate, differentiate and develop into plasma cells that produce antibodies, IgM and IgG. IgM is an immunoglobulin that was first produced as an immune response to antigens followed by a switch to IgG production or other class of antibodies. This depends on the Th cell signal which requires binding with CD40 ligand (CD154) on the surface of T cells, and with CD40 in cell B. In addition, cytokines produced by T cells affect the constant regional genes that give rise to class diversion [1][10].

 Observation of immunoglobulin activity was carried out by looking at the antibody titer namely the highest dilution by converting the antibody titer value with the formula [2 log (titer) + 1] shown in table 1. Then the calculation results were analyzed using Factorial ANOVA method with a significance level of 5% to find out whether or not the effect of exposure to infrasonic (alpha) waves on immunoglobulin activity, the results of the immunoglobulin M analysis can be seen in table 1 and analysis of immunoglobulin G in table 2:

**Table 1**. Results of statistical analysis of IgM using factorial ANOVA method.

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| Source | Type III Sum of Squares | Df | Mean Square | F | Sig. |
| Model | 753.404a | 7 | 107.629 | 1716.797 | .000 |
| Wave | .410 | 3 | .137 | 2.177 | .098 |
| Sunday | .230 | 3 | .077 | 1.220 | .309 |
| Error | 4.577 | 73 | .063 |  |  |
| Total | 757.980 | 80 |  |  |  |

aR Squared = .994 (Adjusted R Squared = .993)

 Knowing whether or not the influence of infrasonic wave exposure on M immunoglobulin mice can be done by looking at the significance value in the table, if the significance value is smaller than 0.05 then Ho is rejected which means there is an influence of infrasonic wave exposure (8-12 Hz) on Immunoglobulin M (IgM), if the significance value is greater than 0.05 then Ho is accepted which means there is no effect of infrasonic wave exposure (8-12 Hz). Based on the table above it can be seen that there is no effect of infrasonic wave exposure (8-12 Hz) on the amount of Immunoglobulin M (significance 0.098).

 The results showed that exposure to infrasonic waves did not affect the amount of IgM of mice because mice that were injected with antigen produced the same amount of IgM to be able to recognize antigens. And IgM cannot be organized properly because the antibodies and antigens have not been adopted due to too short a time.

**Table 2**. Results of statistical analysis of IgG with factorial ANOVA method.

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| Source | Type III Sum of Squares | Df | Mean Square | F | Sig. |
| Model | 750.672a | 7 | 107.239 | 2051.477 | .000 |
| Wave | 1.332 | 3 | .444 | 8.494 | .000 |
| Sunday | .252 | 3 | .084 | 1.607 | .195 |
| Error | 3.816 | 73 | .052 |  |  |
| Total | 754.488 | 80 |  |  |  |

aR Squared = .995 (Adjusted R Squared = .994)

 Knowing whether or not the influence of infrasonic wave exposure on mouse immunoglobulin G (IgG) can be done by looking at the significance values in the table, if the significance value is smaller than 0.05 then Ho is rejected which means that there is an effect of infrasonic wave exposure to Immunoglobulin G (IgG) if the value significantly greater than 0.05 then Ho is accepted which means there is no effect of exposure to infrasonic wave Immunoglobulin G (IgG). Based on the table above, it can be seen that there is an effect of exposure to infrasonic waves (alpha) on Immunoglobulin G (significance of .000).

 After knowing the effect of infrasonic wave exposure on Immunoglobulin G (IgG), it was followed by Duncan's test to find out whether there were differences in each treatment, namely the reading of verses of the Qur'an, murottal al-Quran and the appearance of alpha waves. Duncan test results can be seen in table 3:

**Table 3**. Results of analysis using the Duncan test.

|  |  |  |  |
| --- | --- | --- | --- |
| Wave | N | Subset 1 | Subset 2 |
| 1 | 20 | 2.8500 |  |
| 3 | 20 |  | 3.0600 |
| 4 | 20 |  | 3.1500 |
| 2 | 20 |  | 3.1800 |
| Sig. |  | 1.000 | .121 |

aWave 1: control mice group

bWave 2: groups of mice with the treatment of reading the Qur'an

cWave 3: groups of mice treated by Murottal al-Qur'an

dWave 4: groups of mice with the treatment of alpha waves

 From the table above shows that there is a difference between the control treatment and the treatment of infrasonic wave exposure. But there is no difference between the treatment of infra-electric waves, the treatment of murottal al-Quran, and the treatment of reciting ayat al-Quran. Of the three highest treatment values ​​is the treatment of reading the verse al-Quran (3.1800). Which means that reading verses of the Qur'an is very important because it can have a positive impact on health. Reading al-Quran also includes worshiping Allah, if someone wants to improve his health without having to be complicated by buying CD alpha waves just by reading al-Quran (dzikir).

 The results of the analysis of this study indicate that exposure to infrasonic waves (8-12 Hz) affects the amount of Immunoglobulin G (IgG). Because the treatment of exposure to infrasonic waves (8-12) is performed well, right and continuously will improve positive emotional and coping activities. Emotional positivity can avoid stress so that you are in a calm state. Psycho-neuro-endocrine-immunology is a biopsychological approach that seeks two-way relationships namely the relationship of psychological conditions to the central nervous system (brain) and the relationship of psychological conditions to the immune system that can affect one's health status in the process of healing a disease [2][9].

 The mechanism of action of infrasound waves on Immunoglobulin G (IgG) is: First, infrasonic waves (8-12Hz) in the form of sound vibrations are then received and processed by the ear, then delivered to the brain part of the hypothalamus. To make it easier to see activity in this study using the ACTH-cortisol-immunity pathway. The hypothalamus is a small building in the lower part of the brain that is only about 0.3% of the total brain volume. The hypothalamus is also an integral part of the lymphatic system and is a signaling connector from various parts of the brain leading to the cerebral cortex. Axons from various sensory systems end up in the hypothalamus (except the olfaction system) before the information is passed on to the cerebral cortex. The hypothalamus functions as monitoring and controlling various activities of the body which are very much one of which is the immune system [8].

 Emotional positivity is transmitted to the limbic system and the cerebral cortex with a complex level of connectivity between the brainstem the left and right prefrontal hypothalamus the hippocampus-amygdala. So there is a balance between the synthesis and secretion of neurotransmitters, GABA and GABA antagonists by the hippocampus and amygdala, dopamine, serotonin and norepinephrine produced by prefrontal, acetylcholine, endorphins, and encephalic by the hypothalamus [2][11]. The balance of the synthesis and secretion of neurotransmitters which are inhibitory and ecstatic will also affect the secretion of CRF by PVN in the hypothalamus. Controlled CRF secretion, it will also control ACTH secretion by HIPAA. The control of ACTH secretion will affect the adrenal cortex balance in securing cortisol and some neurotransmitters, adrenaline and noradrenaline, catecholamines with alpha and beta receptors [2].

 Normality of cortisol levels will act as a stimulator to the immune response of the immunological body, both specific and non-specific, cellular and humoral. At specific cellular levels, normal cortisol stimulates cell synthesis, monocytes, neutrophils, eosinophils and basophils. While for specific, cellular and humoral immune response levels, normal cortisol can stimulate lymphocytes, both T lymphocytes and B lymphocytes that produce antibodies [2]. Its workflow starts from cortisol to stimulate macrophages or monocytes to secrete IL-1. The secretion of IL-1 by macrophages can stimulate B lymphocytes to differentiate into plasma cells which then produce antibodies or immunoglobulins, IgM, IgG, and IgA. In other pathways IL-1 secretion by macrophages can stimulate proliferating T cells and differentiate into Th-1, Th-2, Tc, mast/basophil cells, eosinophils, neutrophils, and directly affect NK cells [2].

 On one side Th-2 can stimulate B cells to differentiate into plasma cells, then produce immunoglobulins, IgG, IgM, IgE, and IgD. While cytotoxic T can also control B lymphocytes so they are not excessive in producing immunoglobulins. Besides that cytotoxic T is related to IL-1 to reactivate T-Helper [1]. Thus the three treatments in the form of alpha / infrasonic wave (8-12 Hz), murottal al-Quran, and recitation of the Qur'an if performed continuously can improve the immune system (Immunoglobulin G / IgG).

1. Conclusion

Finally, the conclusion can be derived from this research include: Infrasonic waves (8-12 Hz) can increase immunoglobulin G (IgG). The best infrasonic wave exposure (8-12 Hz) is in the treatment of Quran readings with a value of 3,800.Infrasonic waves (8-12 Hz) do not affect immunoglobulin M (IgM) because mice that are injected with antigen produce the same amount of IgM to be able to recognize antigens. And IgM cannot be organized properly because the antibodies and antigens have not been adopted due to the time being too short (± 5 days).

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