ROLE OF eHSP 72 ON THE INCREASE OF IL-6 AND NK CELLS IN PARTICIPANTS OF MAJELIS DZIKIR

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ABSTRACT

The background of this research was based on encouraging phenomena in today’s modern society. They do not only make dzikir group as a facility for praying, but also to have a sense of peacefulness, to be healthy, to have physical and psychological fitness, and even to heal diseases. This research goal was to disclose the mechanism of increasing immunity as a result of dzikir activities in dzikir group. The category of this research was observational. The design used in this research was time series by using 3 variables eHSP 72, IL-6 and NK cell. The result of this research showed that HSP 72 increased continuously from observation 1, observation 2 and observation 3, that was 0.006 (sig. = 0.006) or p<0.05. Moreover, the number of NK cells in 3 times observation was 0.076 (sig. = 0.076) or p > 0.05. Furthermore, the examination result of IL-6 in 3 times observation was 0.123 (sig. = 0.123) or p> 0.05. The conclusion of this research is that activities in dzikir can increase the value of eHSP 72 in the circulation. However, the increase of HSP 72 cannot significantly increase the IL-6 and NK cell, even though descriptive data show that IL-6 and NK cell increase during 3 times observation.

Keywords: dzikir group, immunity, eHSP 72, IL-6, NK cell

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INTRODUCTION

One phenomenon that was quite encouraging in today's modern society is their tendency to things related to spirituality. Sufism dhikr group or groups appear everywhere and are always flooded with participants. For example, under the leadership of dhikr council Ustadz Haryono, Ustadz Arifin Ilham and many more assemblies of dhikr that appears in these areas and also in demand by the Muslims as a part of modern society. The fact is happening today is that people not only make dhikr assemblies as a means of worship, but they also make use of assemblies of dhikr to get peace, fitness, physical and psychological endurance, and even treatment of a disease.

As one example is the assembly which is under the care of KH Sholeh Hudi Muhyiddin al-Amin in the boarding school Baiturrahmah Blimbing Malang. Assembly remembering is much visited by students from several areas with different purposes. There are among those who deliberately follow the activities in the assemblies of dhikr as a routine to draw closer to God, there are also among those who follow the activities of dhikr gain inner peace, and there are also among those who follow the activity with the purpose to cure a disease. Reality is quite encouraging is that many of those who follow the activities of remembrance for the purpose of healing of an illness, and in fact obtain satisfactory results. From the above phenomenon, it can be assumed that public participation in the assemblies of dhikr, which included a variety of activities dhikr, impact on the improvement of immunity. However, the mechanism of increased immunity after attending a remembrance activities can not be explained until now.
In this regard, the religious approach in the therapy process should need to get attention, in addition to other therapies. As the WHO agreement (1984) who has perfected the healthy limits by adding a spiritual element in it. Thus is healthy is not only healthy in the sense of physical, psychological and social, but also healthy in a spiritual sense. Realize the consequences of a healthy society, is not enough just to involve health professionals. Moreover the role of religious scholars cannot be ignored, especially in creating a healthy society in a spiritual sense.

Several studies have been conducted and the results stated that they are religious, better able to cope with the suffering he faced and it can accelerate the healing process. In a study on women with HIV, it is stated that there was a significant relationship between religiosity with the emotional status and immune status in African-American women are infected with HIV (T Wood 1999). In another study of cancer patients revealed that there is a relationship between religiosity with survival in patients with cancer (Ringdal 1996). Another study conducted on patients with metastatic breast cancer, and the results stated that there is a connection between spiritual expression with immune status in patients with metastatic breast cancer (Sephton et al. 2001). Similarly, a study of a new senior high school students who follow the activities tahajud pray regularly, and the result is that the prayer tahajud performed routinely and khusyu' can enhance the immune system (Sholeh 2003). There are very many similar studies published in medical journals, so that from the scientific facts show that there is a relationship between spirituality or religious activity by the immune system.

Viewed from the perspective of mental health, prayer and dhikr has a profound psychotherapeutic elements. Psychoreligious therapy with prayer and dhikr contain spiritual powers or spirituality that can generate self-confidence (Self-Confident) and a sense of optimism. These two things are very essential for the psychological condition of healing of an illness in addition to medicines and other medical measures (Hawari 2005). Additionally, the prayers and dhikr also contain benefits for the prevention of mental shock and psychiatric disorders, so it can be useful for coaching and increased enthusiasm for life. Thus, prayer and dhikr has the function of curative, preventive and constructive for health (Purwanto 2007).

Resistance mechanisms of this body will be revealed more clearly and detailed through psychoneuroimmunology approach, which describes the interaction between psychological factors to physical factors (Ward 2007). Psychoneuroimmunology explains the relationship between psychosocial factors, the nervous and immune systems. Psychosocial stress would be captured by the five senses, and by the sensory nervous system will proceed to the nerves of the brain called the limbic system via neurotransmitters. Furthermore, psychosocial stimulus was through autonomic nervous system will proceed to the hormonal glands (endocrine) related to the immune system (Hawari 2005).

The concept of psychoneuroimmunology is not a union of three things (psychological, neuro and immune), but it is a discipline that third complementation establish the nervous system as the central point of capture. On that basis, then any stimulus to the central nervous system that causes the secretion of neurohormone responses will give change in activity in the cell body. Several neurohormonal is known is ACTH, cortisol, catecholamines, endorphins, encephaline and somatostatin (Setyawan 1995).

Some recent research says that there are specific proteins that can serve as indicators of both physical and psychological stressors, namely heat shock protein (HSP 72). HSP 72 is a family of HSP 70 that has a 70-kDa molecular weight. Included in the family of HSP 73 HSP 70 is a constitutive nature, where he was always there in the basal state or without stress, and HSP 72 that are inducible, where concentration is increased during stress conditions. In humans, HSP 72 can be found in the cells (iHSP 72) and in the peripheral circulation (eHSP 72). Intra-cellular HSP 72 (iHSP 72) play a role in eliminating protein aggregation, facilitate the refolding process, and act as protein chaperones. Intra-cellular HSP 72 (iHSP 72). This function provides protection against cell from the influence of stressors. While HSP 72 in the circulation/endogenous HSP72 (eHSP72) can be found in patients with various diseases, such as hypertension, atherosclerosis, coronary heart disease. eHSP 72 can also be found in high concentrations in normal individuals newly exposed to stressors, both physical and psychological. eHSP 72 plays a critical role in regulating mechanisms of the immune response (Johnson & Fleshner 2006).

eHSP 72 plays a role in modulating the immune response by stimulating the production of pro-inflammatory cytokines and innate immunity. In several studies have reported that eHSP 72 can stimulate the synthesis of nitric oxide (NO), TNF-α, Interleukin 1-β (IL-1β) and IL-6 produced by macrophages and neutrophils. In experiments conducted in mice, increased levels of IL-6 in the circulation was in response to acute stress tailshock. Similarly, IL-1β potentially has increased after exposure to tailshock stress (Ader 2007). In addition, eHSP 72 can also enhance NK cell proliferation and sensitivity (Johnson & Fleshner 2006). This study aims to uncover the mechanism of increased immunity to participants assemblies of dhikr through the examination of 72 eHSP that can trigger an increase in IL-6 and NK cells. If this can be disclosed and explained scientifically, then the
religion through *dhikr* assemblies can be removed as part of health services, either as a step promotive, preventive, and curative.

**MATERIALS AND METHODS**

This type of study chosen in this study is observational, while the design of the study is a time series. The population used in this study was participants *majlis dhikr* Miftahul Jannah Islamic boarding Darussalam Gadung Drisyorejo Gresik. Large sample in this study were at least 12 people taken with Simple Random Sampling technique, where samples that have met the inclusion criteria as many as 20 people were taken randomly to meet sample size has been determined, which is 12 people. To check eHSP 72 and IL-6 we used ELISA method (enzyme Imunosorbent Linked Assay). As for the examination of NK cells using the method immunocytochemistry. Data analysis was performed by descriptive and inferential statistics using SPSS for windows to reveal the standard deviation and average (mean) of all variables, statistical analysis is then performed using a repeated measurement on the grounds that the study sample is a single sample is examined in three time of observation.

**RESULTS**

The mean results of the research variables, namely eHSP72, IL-6 and NK cell that were carried out 3 times, i.e. once in the 3 days prior to the implementation of *dhikr*, and twice at 24 hours after the execution of *dhikr* (Table 1). It was found that 72 eHSP always known that increased from observation 1, observation 2 and observation 3 (Table 1). Similarly, IL-6 is always increased from observation 1 to observation to-3 as occurred in eHSP 72. While the number of NK cells, at the beginning of observation reaches an average value of 41.0000, subsequent observations have increased in the 2nd (68.4167) and slightly decreased in the observation of the 3rd (67.5000). The test results in 3 times 72 eHSP observations 0.006 (Sig. = 0.006) or p <0.05 which means an increase in HSP 72 significantly between observations 1, 2 and 3 (Table 2). Whereas, the results of the IL-6 in 3 times the observation 0.123 (Sig. = 0.123) or p> 0.05, which means there was no increase in IL-6 significantly between observations 1, 2 and 3 on the panel participants *dhikr* after attending activities *dhikr* (Table 3). The results of NK cells in three observation times .076 (Sig. = 0.076) or p> 0.05, which means there was no increase of NK cells significantly between observations 1, 2 and 3 on the panel participants *dhikr* after participating in activities (Table 4).

<table>
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<th>Mean Square</th>
<th>F</th>
<th>Sig.</th>
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DISCUSSION

From the tables and graphs above 72 eHSP always known that increased from observation 1, observation 2 and observation 3. This fact can be explained that the activities of prayer and remembrance as well taushiyah followed by a new panel of dhikr Miftahul Jannah arguably the psychological stressors that can trigger increased levels of HSP 72 in the blood. This condition is consistent with research that explains that HSP 72 can also be found in the peripheral circulation in normal individuals, in cell death without due process of necrosis, as well as an increase in individuals who experience psychological stress (Asea 2007, Johnson & Fleshner 2006).

In a series of activities are provided taushiyah dhikr in the form of a review of the book of al-Hikam which describes the efforts to draw closer to God, to cleanse the liver through the avoidance of acts that are not good (al-madzmumah morality) and replace it with good deeds (al mahmudah morality). Through the senses of hearing, the entire congregation listen to what is delivered by the imam (murshid) to understand the study material.

Vilyanur Ramachandran explains that there are parts of the brain responsible for responses of human spiritual and mystical. They call it a god spot and located in the temporal lobes. In these lobes occur interpretation of what is heard. In this place, the message is read by the brain and sent to the other lobe, especially in the frontal lobe to be addressed (Pasiak 2005).

While prayer and dhikr is done is based on the book that serve as a guide by the assemblies of dhikr Miftahul Jannah under the leadership of the imam. All members of the congregational dhikr assemblies that mimic wird recited by the priest. For example bacaan paragraph chairs, brooms and prayers asmaul husna universe. All of this wird should be read with full concentration to achieve the perfect fervently. Khusyu' referred to is the condition of a relaxed, focused, and full concentration (Purwanto 2007).

However, new participants dhikr assemblies twice this dhikr is to follow the activities in the learning process (learning process) to be able to follow the activities of prayer and remembrance in khusyu' and full concentration in order to achieve a state of relaxation that is characterized by feelings of calmness and clarity of mind (Pasiak 2005). This learning process your mind becomes alert to digest all the information coming through the senses. This condition can be categorized as psychological stressors as evidenced by elevated levels of 72 eHSP significantly over the three observation times. That is, the condition of relaxation is still not achieved, due to new participants assemblies of dhikr is still in the learning process. In this learning period, it is possible there are some things that can interfere with concentration, among other things: the mind has not come into the self, there is a high emotional turmoil, mind distracted by the turmoil of passion, the mind is filled with many problems, have not been able to enjoy the worship, and worship only because of fear of sin (Hakim 2002).

Additionally, the prayers and dhikr performed within 90 minutes, for beginners is also a stressor that can trigger psychic tension. Psychic tension may occur as a result of the implementation of prayer and remembrance for a long time with the rules established by the assemblies of dhikr. In addition, the psychological tension may also occur as a result of grief about the sins that have been done, the desire to keep close to God when his name, and expectations are communicated through prayer and dhikr. For example when istighfar (astaghfirullahah adzim) which means the request forgiveness of all sins to God ever made, strands of monotheism sentence (laa ilaha illalllaah) which is an essential statement of the oneness of God, so that only the God-man is proper pleading for help.

This kind of psychic condition responded to by the hypothalamus. The hypothalamus has a crucial role in the regulation of emotions and responses to stress, so that the hypothalamus is also known as "stress center" especially given the role in mobilizing the body to react to stress. In this case, the hypothalamus controls the pituitary to produce several hormones. Control is very important to mobilize a physiological process in order to provide a response to fight or flight situation (Atkinson, et al, nd).

On the other hand, there is the hypothesis that increased eHSP 72 in the circulation can occur during stress, when

Table 4. Tests of Within-Subjects Contrasts NK cell

<table>
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<tr>
<th>Source</th>
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<th>Mean Square</th>
<th>F</th>
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</table>
Role of eHSP 72 on the Increase of IL-6 and NK Cells in Participants of Majelis Dzikir (Siti Nur Asiyah)

triggered by the sympathetic nerve activity and norepinephrine hormones. This hormone norepinephrine which then triggers an increase in the concentration of HSP 72 in the circulation through α1-ADR. Stimulation of α1-ADR This will cause an increase in intracellular Ca²⁺ and stimulates the release exosom containing HSP 72. Increased HSP 72 is able to modulate the immune response through the interaction of several surface receptors expressed on specific immune cells (Johnson & Fleshner 2006, Asea 2007).

This hypothesis which has been demonstrated in this study, where there was an increase in blood levels of 72 eHSP after 24 hours of execution of dhikr. Similarly, when performed 72 repetitions of eHSP examination at 24 hours after the execution of the second week of remembrance. This was evidenced by statistical tests using a repeated measurement which generates a number of significance 0.006 (p <0.05) which means there is significantly increased levels of eHSP 72 on dhikr assembly participants after attending a remembrance activities.

In several studies have reported that eHSP 72 can stimulate the synthesis of nitric oxide (NO), TNF-α, Interleukin 1-β (IL-) and IL-6. In addition, eHSP 72 can also enhance NK cell proliferation and sensitivity (Johnson & Fleshner 2005). In this study the number of NK cells in the initial observation reaches an average value of 41.0000, subsequent observations have increased in the 2nd (68.4167) and slightly decreased in the observation of the 3rd (67.5000). Decrease in NK cell from observation 2 to observation 3 indicates that there has been a state eustress considered by the body does not require an increase in NK cell. That's because NK cell function is as a killer virus-infected target cells and other target cells coated with IgG that NK cells function as cytotoxic cells that depend on antibody (antibody dependent cell mediated ADCC cytotoxicity=) (Kresno 2001).

While IL-6 is always an increase from observation 1 to observation to-3 as occurred in eHSP 72, although the increase that occurred were not significant enough, as a result of psychological stressors during dhikr activities. The trend increase in IL-6 for 3 times the observation that IL-6 does not have a limiting factor as owned by IL-1.

IL-6 is a cytokine released by macrophages are activated by the acute phase response, and has contributed in host defense. IL-6 is able to increase body temperature, also known as endogenous pyrogen. One of the most important effects of IL-6 is initiated the formation of acute-phase response. In the acute-phase response will be formed by a protein synthesized and secreted by the liver, these proteins known as acute-phase proteins (APP). One of the acute phase proteins is C-reactive protein (CRP). CRP binds to the bacteria not only as an opsonin but also can activate the complement cascade by binding to C1q, which is the first component of complement activation in the classical pathway (Guntur H 2006). As a non-specific immunity, IL-6 stimulates hepatocytes to produce APP and with CSF (Colony Stimulating Factor) stimulate progenitor in the bone marrow to produce neutrophils (Baratawidjaja 2006).

From the results of statistical tests by using a repeated measurement found that IL-6 produced a sig. 0.123 (p>0.05) and NK cells produce a number sig. 0.076 (p>0.05), which means that there is no significant increase in levels of IL-6 and the number of NK cells in the assembly of dhikr participants dhikr following activities, although there has been increased 72 eHSP levels significantly.

This phenomenon can be explained that the activities undertaken by dhikr participants of Miftahul Jannah new assembly is the psychological stressors that can trigger increased levels of HSP 72 in the circulation. Stressors should result in stress perception. Stress perception is an early stage of defining the source of stress or stressors that received sensory organs and determine the accuracy of the stress response to stressors. Thus the conceptual nature of stress perception can be empiricized by measuring the change in the biological stress response of all systems associated with endurance. Furthermore, the individual will respond to these stressors can reach a new equilibrium (eustress) or lead to balance disturbances (distress) (Son 2005).

In this study, the stress response can be observed through the descriptive data showing the trend of increased levels of IL-6 and NK cells, although the increase has not occurred significantly. It may happen due to psychological stressors that participants gained during the dhikr assembly and activity was observed as many as three times the yield stress a different perception. As said by Ben Walgito that there are two factors that affect perception, the first is an internal factor that comes from the individual itself that can affect their emotional intelligence. These internal factors have two sources in terms of physical and psychological terms. In terms of physical health concerns of individuals, while covering the psychological aspects of experience, feeling, thinking ability and motivation. Second, in the form of external factors and environmental stimuli, stimulus to be perceived was strong enough and exceeds the threshold stimulus. Environment or special situations the background stimulus will also be influential in perception. Object and the background environment is unanimity or entity objects that are difficult to separate.
The same object with different social situations can produce different perceptions (Walgitto 2009).

In terms of physical health, all participants assemblies of dhikr which became the subject of this study was in good health, according to inclusion criteria previously set. Meanwhile, in terms of psychological experience, feeling, thinking skills and motivation is a condition that is very subjective and not observable, so it is very possible that considerable variation among individual study subjects. Psychological condition that varies is very likely determines the difference in stress perception. This perception of stress differences may affect the accuracy of the formation of stress response.

While external factors in the form of stimulus and the environment also influences the formation of perceptions. The stimulus in this case is the activity of prayer and remembrance as well as taushiyah by the assemblies of dhikr Miftahul Jannah Islamic boarding Darussalam Gadung Driyorejo Gresik. As already mentioned above that one object among the inclusion criteria of this study were new participants in the assemblies of dhikr Miftahul Jannah and have never followed a similar activity on other assemblies of dhikr. New participants who take part in assemblies of dhikr is observed three times, ie 3 days prior to the implementation of dhikr, and 24 hours after the implementation of the dhikr. Observations after dhikr only done 24 hours after the first and second dhikr. That is, participants assemblies of dhikr is only two times following the activities of dhikr. Meanwhile, one among the factors that influence the formation of social perception is based on the introduction of familiality i.e. repeated exposure of a stimulus so that the resulting stimulus is perceived to be more accurate, and the intensity associated with the degree of awareness about the stimulus person.

Thus can be explained that the stress response accuracy has not been formed in this study is due to the formation of stress perception has not been adequately. This is because the observation is only done twice after they follow the activities of the first and second dhikr. There is a possibility if the extension of observations made after they repeatedly follow the activities of dhikr, then the perception of stress will be formed adequately and will affect the formation of the stress response shown by an increase in immunity significantly.

Researchers are aware of the fact that there are differences in the formation of stress perception and stress response occurs in humans and animals. When the researchers took the initiative to conduct a second observation, at 24 hours after the execution of dhikr first and third observations at 24 hours after the execution of the second dhikr that has a time interval for one week, because the researchers refer to previous studies conducted in mice. Mice that were exposed to the stressor in the form of foot shock and then their blood samples taken for examination of the levels eHSP 72, then the result stating that there was an increase eHSP 72 followed by an increase in immunity. Elevated levels of eHSP 72 accompanied by an increase immunity in mice was re-occur after exposure to the same stressor (foot shock) during the first 7 days after exposure (Ader 2007). This condition is apparently not been seen clearly in the form of psychosocial stressors that occur in humans, but it should be recognized that there is an increasing immunity to the new participant Miftahul assembly dhikr dhikr Jannah after following activities were observed in 3 times and observations. The picture can be seen in the descriptive data from each of the parameters of immunity (For more details and pictures can be seen in the above table). Thus, this study has provided preliminary information that there has been an increasing trend of immunity to new participants assemblies of dhikr Miftahul Jannah after attending two events dhikr.

The fact is in line with Hawari opinion stating that the prayer and dhikr contains in-depth psychotherapeutic elements. Psychoreligious therapy is no less important with psychiatric psychotherapy, because therapy psychoreligious contain spiritual powers or spirituality that evokes a sense of confidence and a sense of optimism (Hawari 2005).

Zakiah Daradjat, experts and practitioners of counseling and psychotherapy Islam also believes that prayer can provide a sense of optimism, zest for life and relieve feelings of hopelessness when one deals with circumstances or issues that are less enjoyable for her. Support for the spirit of prayer into one's life. Praying can foster a sense of optimism in the self, and keep a sense of pessimism and despair, so that prayer contains benefits for the prevention of the occurrence of mental shock and psychiatric disorders. Moreover, the prayer has benefits for the development and improvement of the spirit of life (Purwanto 2007). Confidence and optimism was also instrumental in improving immunity to achieve a state of homeostasis. As McEwen opinion that the magnitude of the stress response not only depends on the stressors and the individual, the stress response also depends on the strategies adopted by individuals to cope with stress (Pinel 2009). One important factor is the ability of individuals to be able to control stress. Perceptions of control mediated the influence of stress on the human immune system. In one study of women with breast cancer found that patients who are pessimistic are more likely to experience a new tumor in a period of five years, even after they are taken into account the severity of physical illness (Gunawan & Sumadiono 2007).
CONCLUSION

With reference to research results and the above discussion, it can be concluded that the activities in the assemblies of dhikr that dhikr done regularly, directed and programmed, with a frequency of once per week with a duration of 120 minutes each time during the meeting, can raise levels of HSP 72 in circulation (eHSP). Increased eHSP 72 followed by the trend of increased levels of IL-6 and the number of NK cells, although there was no increase significantly. Thus the activities of dhikr in assembly, can be explained scientifically by using psychoneuroimmunology approach through the hypothalamic-sympathetic nerve-adrenal medulla can lead to elevated levels of eHSP 72 and potentially enhance immunity. In practical terms, the activities in the assemblies of dhikr that dhikr done with regular, targeted and programmed may act as an immunomodulator that is cheap, easy and valuable service.

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