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IMMUNE RESPONSE (CORTISOL, TNF α , HMGB1) IN TRAINED AND UNTRAINED ADOLESCENT AFTER 12 MINUTES RUN EXERCISE

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ABSTRACT

This study aims to explain the immune response (cortisol levels, TNF Alpha, HMGB1) in adolescents after moderate-intensity aerobic exercise (70-79%⁴⁶ NM) running for 12 minutes. The research was conducted by post test only on 15 basketball-trained students and 15 basketball-untrained students at SMAN 1 Banjarbaru with an average age of 17 years. The sampling technique was purposive sampling method. The value of VO₂ max was measured on the first day with MFT (Multistage Fitness Test). Measurement of cortisol, TNF Alfa, HMGB1 was carried out on the third day by taking blood after the students did aerobic exercise while running for 12 minutes. Data analysis used the Mann-Whitney test for VO₂ max, cortisol, TNF Alpha, and t-test was not fitted for HMGB1. The results showed that the average VO₂ max, cortisol, and HMGB1 scores of students with basket achievement were higher than those who were not found. On the other hand, the TNF alpha level that did not increase was higher. The result of data analysis is the difference in mean⁴⁵ between students finding baskets and not (p <0.05) on VO₂ max, Cortisol and TNF Alpha. There was no significant difference (p > 0.05) in HMGB1. There is a positive correlation in adolescents between Cortisol and TNF Alpha, Cortisol and HMGB1, HMGB1 and TNF Alpha.

The conclusion of this study is that adolescent Cortisol levels increased higher than not increased. Adolescent TNF alpha levels were lower than not. It has not been¹² proven that trained HMGB1 levels are different from untrained ones. In adolescents there is an increase in cortisol levels followed by an increase in TNF Alpha levels, an increase in⁵³ cortisol levels followed by an increase in HMGB1 levels and an increase in HMGB1 levels followed by an increase in TNF Alpha levels. Moderate-intensity aerobic exercise 12 minutes of running can be recommended as one of the simple exercises to maintain fitness and boost the immune system or exercise health during a pandemic.

Keywords: Immune response, 12 minutes of running, trained and untrained adolescents

INTRODUCTION

WHO states that in 2019 the majority of people around the world are less physically active, thus endangering their health in the future, so WHO recommends that physical exercise, especially

moderate intensity, is carried out for 150 minutes a week for someone aged 18-64 years. years (Bennie et. al., 2019). Regular exercise has effect of decreasing the risk of developing various diseases and increase life expectancy (Sand et. al., 2013; Singh et. al., 2019; Kokkinos et. al., 2012; McKinney et. al., 2016; Mallo et.al., 2019; and Nieman., 2012). One form of physical exercise is a game of basketball. Although not as popular as football, basketball is popular among Indonesians (Piepoli et.al., 2016; Singh Chahar, 2014). Basketball uses alternating aerobic-anaerobic metabolism, although anaerobic metabolism is more dominant (Gomes et. al., 2014).

Aerobic exercise is an activity that utilizes the continuous and normal contractions of large muscle groups a period of time and utilizes aerobic metabolism as a source of energy. One of the components of aerobic exercise is intensity (Ploughman and Smith, 2014). Moderate intensity aerobic exercise, an activity that consumes energy comparable to brisk walking, is the aerobic exercise most frequently studied (Plowman et.al., 2014; WHO, 2010; Patel et.al., 2017). Physical exercise is related to a person's ability to perform activities through its effect on VO₂ max (Habibi et.al., 2014). 12-minute running was used as a way to measure a person's VO₂ max value (Bandyopadhyay 2015; Penry et.al., 2011; Das, 2013). In addition, exercise is a stressor that stimulates the neuroendocrine system and immune response (Ploughman and Smith, 2014). The correlation between the immune system and exercise was first studied in David Nieman's study which showed a lack of reports of ARI complaints in individuals who routinely do moderate intensity physical exercise (Nieman, 2012; Simpson et.al., 2012). Physical exercise drastically affects the number of leukocytes circulating in the peripheral blood. Leukocytosis that accompanies acute exercise is a transient phenomenon because the number and composition of the leukocytes usually return to normal values within 6-24 hours after an exercise session. Leukocytosis that occurs in physical exercise is mostly caused by activation of neutrophils and lymphocytes, as well as a small proportion of monocytes (Gleeson et.al., 2013). Physical exercise activates the HPA response, which causes the hypothalamus to release corticotropin-releasing hormone (CRH) to cells in the anterior pituitary, which stimulates the release of adrenocorticotrophic hormone (ACTH) and stimulates the adrenal cortex to release glucocorticoids (cortisol) into the bloodstream (Habibi et. al., 2014).

METHOD

Cross sectional research was conducted on the population of all adolescents at Senior High School 1 (SMAN 1) Banjarbaru.. The sample in this study was divided into two, namely the sample was taken from 15 basketball-trained students and 15 untrained basketball students. The sample was cooperative and had filled out a consent form to be the subject of the study. The purposive sampling method was used to take samples according to the inclusion criteria, namely (a) male, (b) aged 15-18 years, (c) physically healthy, meaning that at the time of the study the probandus was not sick or infected. and has no history of heart or lung disease. and allergies, (d) Cooperatives, research subjects can be invited to work together to carry out research procedures, (e) Do not smoke, (f) Do not consume drugs that affect the number of leukocytes, monocytes, and neutrophils at least 2 days before blood collection, (g) Have a normal body mass index (BMI) (20-25). The sample of basketball-trained adolescents is students of SMAN 1 Banjarbaru who are members of a basketball sports club and routinely practice basketball at least three times a week for one hour per training

session for a year. The study will be stopped on subjects who experience signs of fatigue while doing the exercises so that they cannot complete the exercise.

The research was conducted for 3 days. On the first day, VO2 max measurements were carried out using the MFT (Multistage fitness test). Subject was rested on the second day. Blood samples were taken for analysis of levels of cortisol, Tnf alpha, and HMGB1 were checked on the third day. A 5 cc blood sample was taken from the brachial vein after running twelve minutes of moderate intensity aerobic exercise. Before training, the research subjects will have their maximum pulse rate (MHR) calculated using the tanaka formula. After knowing MHR, research subjects will use pulse oxymetry and warm-up in the form of running in groups consisting of 3 people per group until they reach the MHR target of 70-79%. After reaching the target of 70-79% MHR, the subject will continue to run for twelve minutes at the same rhythm. Blood sampling and subject analysis were carried out by trained personnel from the Prodia Banjarmasin laboratory. Trained health workers always accompany training sessions to prevent life-threatening things from occurring. A different test is used the Mann Whitney test for VO2 max, cortisol, TNF Alpha. whereas the unpaired student T test for HMGB1.

RESULTS

Sample characteristics were based on age, oxygen saturation, body mass index, pulse, height, weight, systolic and diastolic blood pressure. The average sample was 17 years and the mean value of pulse rate, systolic and diastolic blood pressure, body weight, body mass index of students trained in basketball was lower than students who were not trained in basketball. Pulse rate, systolic and diastolic blood pressure, body mass index, oxygen saturation were within normal limits. The table above shows that students who were trained in basketball were fitter than students who were not trained in basketball.

Table 1. Characteristics of research subjects

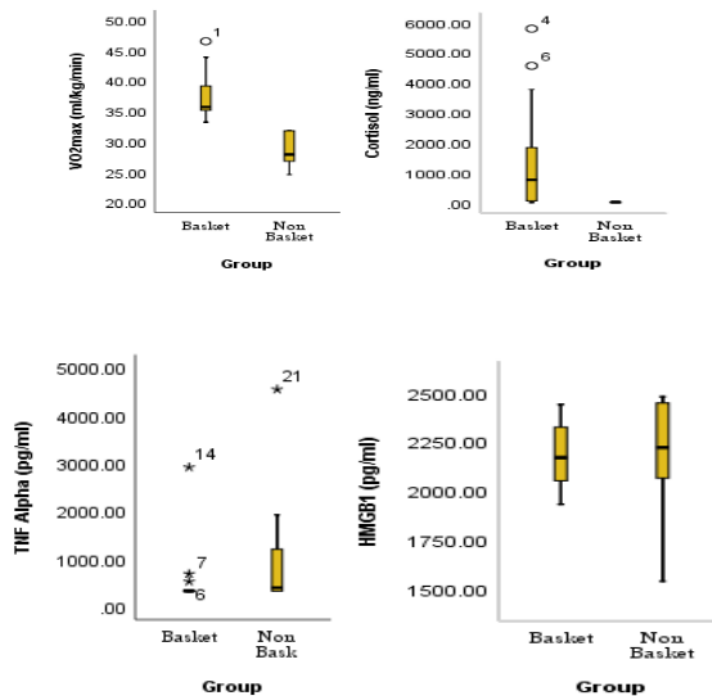
Characteristic (Mean ± SD)	Basket group (Trained Basketball Student n=15)	Non Basket Group (Untrained Basketball Student n=15)
Age (Years)	16.93±0.258	17.07±0.495
SO2 (%)	97.27±2.576	96.93± 3.731
Pulse per minute	84.20±11.384	92.93±12.981
BMI (kg / m2)	21.65±2.104	21.68±5.911
Height (cm)	167.37±8.067	172.00±5.305
Weight (kg)	61.13±10.034	64.33±18.289
Systolic blood pressure (mmHg)	127.53±14.282	132.20±13.078
Diastolic blood pressure (mmHg)	78.73±7.601	82.87±9.797

Table 2. Research Results

Marker	Trained Basketball Student (n = 15)		Untrained Basketball Student (n = 15)	
	Mean	SD	Mean	SD
VO ₂ max	37,49	26,1	28,85	18,8
Cortisol	1440,1	1863.1	26,3	16.1
TNF Alpha	532,0	667,2	963,7	1144,8
HMGB1	2180,6	166.4	2168,9	295.8

The mean VO₂ max, cortisol and HMGB1 concentrations of basketball-trained students were higher than untrained. In contrast, the untrained TNF alpha score was higher than the untrained.

Graph 1. Boxplot graph of VO₂ max Value, Cortisol, TNF Alpha, and HMGB1 concentration



VO₂ max

The mean VO₂ max of basketball trained students was 37.49 ± 26.1 ml / kg / minute higher than the untrained students, namely 28.85 ± 18.8 ml / kg / minute. With the Mann Whitney test, the VO₂ max value obtained was statistically significant difference in the VO₂ max value after 12 minutes of moderate intensity running training for students who were basketball trained and untrained p = 0.000 (p <0.05). Spearman's rho correlation test, there was a significant correlation

$p = 0.000$, with a very strong positive correlation (0.870^{**}) between VO_2 max and whether adolescents were trained in basketball. The more trained a teenager, the greater the VO_2 max value.

Cortisol Concentration

The average cortisol concentration of basketball-trained students was greater than that of untrained students. The mean blood cortisol concentration of basketball-trained students was $1,440.1$ ng / ml higher than the untrained, which was 26.3 ng / ml. In this study, statistically significant results were obtained using the Mann Whitney test and there was a significant difference in cortisol levels after twelve minutes of running with moderate intensity training in basketball trained and untrained students $p = 0.000$ ($p < 0.05$). Spearman's rho correlation test, there was a significant correlation $p = 0.000$, with a very strong positive correlation (0.720^{**}) between cortisol and whether adolescents were trained in basketball. The more trained a teenager, the greater cortisol levels will be.

TNF Alpha concentration

The mean TNF Alpha concentration of basketball-trained students was lower than that of untrained students. The mean TNF Alpha concentration for basketball-trained students was 532 pg / ml and untrained 963.7 pg / ml. Mann Whitney test showed a significant difference between the two after 12 minutes of moderate intensity training for basketball-trained and untrained students $p = 0.000$ ($p < 0.05$). There was no significant correlation between Spearman's rho test, $p = 0.077$, with a weak negative correlation (-0.327) between TNF Alpha and whether adolescents were trained or not in basketball. The more trained a teenager will have lower TNF Alpha levels.

Concentration of HMGB1

The mean HMGB1 concentration of basketball-trained students 2180.6 ± 166.4 pg / ml was higher than the untrained 2168.9 ± 295.7 pg / ml. There was no significant difference in HMGB1 levels after 12 minutes of moderate intensity exercise in basketball-trained and untrained students $p = 0.895$ ($p > 0.05$). There is no significant correlation between Pearson's correlation test $p = 0.895$, with a very weak positive correlation (0.025) between HMGB1 and whether or not teenagers are in basketball. The more trained a respondent is, the greater the HMGB1 level will be.

DISCUSSION

The statistical tests in this study showed significant differences in VO_2 max, cortisol and TNF alpha levels between basketball-trained and untrained students. While another aspect, namely HMGB1, there was no statistically significant difference between students who were trained in basketball and those who were not trained.

Physical activity causes changes in the physiological functions of the body, both temporary changes (response), and changes that are permanent (Mulyono, 2016). This also occurs in people

who do sports, there is a physiological response so that the body's homeostasis is well maintained. An increase in workload due to exercise will cause an increase in the workload of the organ systems, for example in contracting skeletal muscles, an increase in glucose uptake from the blood, an increase in body metabolism, one of which is the release of heat as compensation characterized by sweating, an increase in the amount of energy needed, and the cardiovascular there is an increase in heart rate due to pumping blood to supply more to all organs (Giriwijoyo, 2007). The occurrence of a physiological response in the musculoskeletal system as an adaptation to physical exercise occurs by involving many molecular pathways that play a role in regulating muscle contraction and biosynthetic pathways for ATP formation, such as AMP activated protein kinase (AMPK), Sirtuins (SIRT's), mitogen activated protein kinases (MAPKs), and oxygen sensor prolyl hydroxylases (PHDs) (Camera et. Al, 2016; Fan and Evan, 2017; Kjøbsted et.al., 2018). All these changes require a good balance so that the work of the organs and body condition is not disturbed, that is what is called homeostasis. Thus, in athletes if the homeostatic response fails, it will reduce their physical performance and performance (Giriwijoyo and Sidik, 2012; Ashadi, 2014).

The VO₂ max score of basketball-trained adolescents was higher than the untrained. This is in line with the research of Buchan et al. In this study it was found that physical exercise three times a week for seven weeks at moderate intensity increased VO₂ Max values compared to those who did not exercise in adolescents (p = 0.000). The study explains that this is likely due to the effect of moderate intensity physical exercise on cardiac output, where physical exercise causes an increase in cardiac output due to an increase in stroke volume (Buchan et.al., 2011).

Macpherson et.al.'s research, 2011 describes two factors that cause an increase in VO₂ max, namely central factors and peripheral factors. Centrally, the increase in VO₂ max was due to an increase in stroke volume and a slight increase in maximal heart rate. Peripherally, the increase in VO₂ max occurs due to increased arterial-venous oxygen differences that are influenced by oxygen transport to active muscle fibers, local enzyme adaptation, and mitochondrial density. Peripheral VO₂ max increase occurs in sprint interval training (Lee et. Al, 2014).

In accordance with the study by Kohrt et al. In elderly aged 60-71 years, 53 men and 57 healthy women and a control group did not follow any exercise. The VO₂ max value is measured before and after aerobic exercise in the form of treadmill walking and running for 9-12 months. The results showed an improvement in the VO₂ value after a series of exercises in the male and female groups by 21 and 19%, respectively. It is provided that both elderly women and men can obtain VO₂ max improvement during routine exercise stimuli. Statistically, this study shows that the improvement in VO₂ max level does not depend on age, gender, VO₂ max value before undergoing exercise. or the intensity of the exercise undertaken (Wendi et.al., 2005).

Carazo-Vargas et al's research was conducted on 34 children aged 10-18 years who were physically healthy who participated in strength training. The results showed an increase in VO₂ max values after participating in training (Pedron et.al., 2015). In this study there was no significant difference in VO₂ max levels based on age, but another study by LeMura et al showed women experienced more VO₂ improvement than men after an exercise program (LeMura et.al., 1999).

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Baquet et al showed that there was no significant difference in VO₂ max values between athletes and non-athletes after exercising (Baquet et.al., 2003). Meanwhile, Huang in his journal states that greater VO₂ max improvement can be obtained with exercise for more than 20 weeks and an intensity of around 60% and 70% (Huang et.al., 2005).

Cortisol

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The results of this study indicated that there was a significant difference in cortisol levels after 12 minutes of physical exercise running between trained basketball and not (P = 0.000). The basketball-trained cortisol value (1440.1 ng / μL) was higher than the untrained (26.3 ng / μL), this may be a homeostatic process not due to a stressor. And the average value of HMGB1 is also bigger. So that cortisol here functions as an anti-inflammatory and suppresses proinflammatory cell cytokines such as IL6, TNF Alpha, Leukocytes, Neutrophils and Monocytes. In contrast, untrained cortisol values were lower so that the anti-inflammatory function was not strong enough to suppress the inflammatory reaction as indicated by a greater increase in the TNF alpha concentration. Sports and physical activity can be a source of stress for the body and have an impact on other body systems and have the potential to cause homeostatic disorders (Mastorakos et.al., 2005).

Previous research has consistently shown that exercise with an intensity of more than 60% VO₂ max stimulates the release of higher cortisol concentrations in adults. Studies show that every teenager has the same body response to an increase in the HPA axis reaction and an increase in the cortisol response after exercise. In adolescents aged 15 to 16 years who exercise for 12 minutes with an intensity of 70-85% of the maximum pulse, there will be a striking increase in cortisol levels compared to the group that only does moderate intensity exercise (maximum 50-65% pulse rate) (Budde et.al., 2015).

Exercise with VO₂ max intensity of 60% (pre-intervention 12.3 ± 4.1 and post-intervention 20.1 ± 6.0) and 80% (pre-intervention 12.9 ± 6.3 and post-intervention) resulted in an increase in cortisol levels. significantly greater than the session with 40% exercise intensity (pre intervention 12.2 ± 4.3 and post intervention 10.8 ± 5.4). That is, cortisol levels in plasma increase in moderate and high intensity exercise. Conversely, exercise with light intensity did not show a significant increase in cortisol levels but decreased (Hill et.al., 2008).

Keyan et al's study of 62 healthy participants, 31 of whom did 10 minutes of intense exercise and 31 others did a leisurely walk. Examination of cortisol levels in saliva showed a significant increase in cortisol levels after intensive exercise compared to before exercise. (Zhang, et al, 2019).

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The cortisol concentration will continue to increase with increasing intensity and duration of exercise (Duclos and Tabarin, 2016). Different exercises have different effects on the hormonal system, a greater response is shown in strength training. Cortisol levels will increase according to the level of stimulation given. High-intensity exercise will cause an increase in the activity of stress hormones such as cortisol, ACTH, and catecholamines, which inhibit protein synthesis and trigger protein degradation resulting in the breakdown of skeletal muscle protein (Corazza et.al., 2014).

The level of cortisol secretion is influenced by the circadian cycle. Serum cortisol secretion begins to increase at midnight, peaking in the morning. In addition, it is possible that this increase is due to other factors that can increase cortisol secretion, namely psychological stress which also triggers the release of cortisol (Haslinda et.al., 2017).

The increase in cortisol levels after acute physical exercise has been proven in several studies (Sato et.al, 2016; Muscella et.al., 2019) The increase in cortisol levels is strongly influenced by the intensity of physical exercise and the subject's physical exercise habits. Hackney et..al's research shows that cortisol levels increase with increasing training load given to subjects aged 16-21 years (Hackney et.al., 2011). In addition, the research of Sato et.al., showed that there were differences in the increase in cortisol levels in athletes and non-athletes according to the intensity of the physical exercise given. Increased cortisol levels provide an anti-inflammatory effect by emphasizing the expression of pro-inflammatory cytokines, thereby inhibiting the increase in the number of leukocytes (Coutinho and Chapman, 2011; Ince et.al., 2019).

The release of cortisol is one form of body adaptation to stress due to exercise, this can be seen from the cortisol levels in blood plasma, as found in the results of this study. Cortisol causes an increase in fat content in the blood due to an increase in amino acids and fat mobilization which, if excessive, can disrupt tissue structure and function, and even cell death (Tkacova et.al., 2012).

Beserra et al conducted a review of 5 articles and 7 analyzes with the aim of looking at post-exercise cortisol levels in 463 respondents with major depressive disorder. Respondents did the exercises with different frequency, way of measuring cortisol, and control groups. The results found that physical exercise caused a decrease in cortisol levels. However, this decrease in cortisol depends on the type, frequency and measurement of cortisol taken. It is known that the decrease in cortisol levels can occur effectively after doing aerobic exercise with a frequency of 5 times per week (Beserra et.al., 2018).

On the other hand, the study by Wang et.al showed the opposite result with the research above. He conducted a study to assess the comparability of the effectiveness of aerobic exercise and resistance training in improving cognitive function in 42 young adults who were asked to do Stroop exercises after undergoing aerobic exercise, resistance training and sedentary conditions after a few days later. The saliva samples taken were at baseline conditions before exercise, immediately after exercise, and 30 minutes afterward. The results showed that cortisol levels increased after exercise, both compared with cortisol levels during the sedentary phase and baseline levels. Cortisol levels immediately after exercise were also higher than 30 minutes afterwards (Wang et.al., 2019). From this it is concluded that an increase in cortisol levels can occur after exercise, but is influenced by the type and intensity of exercise.

TNF Alpha

The results of this study indicated that there was a significant difference in the levels of TNF Alfa after 12 minutes of running physical exercise between trained basketball and not ($P = 0.000$).

Study by Dimitrov et al. Downregulation of monocytic TNF production during acute exercise mediated by high epinephrine levels (Dimitrov et.al., 2017). Terink et.al's study included 50 men

(mean age 58.9 ± 9.9 years) and 50 women (mean age 50.9 ± 11.2 years). The result was that all observed cytokine concentrations after exercise (IL6, IL8, IL10, IL1 beta, and TNF alpha) increased from baseline values ($P < 0.001$). Then the concentration decreased from day one to day two ($P < 0.01$). Exercise induces an increase in cytokines, but these levels will decrease in the following days even though the exercise is still carried out with the same training intensity and load (Terink et al., 2018). Nemet et. al. conducted a study on 11 healthy males of middle school age, ranging from 14 to 18.5 years, after 1.5 hours of single wrestling exercise. There was an increase in proinflammatory cytokines after exercise, namely IL6. TNF alpha and IL1 beta (Nemet et. Al., 2002).

In contrast to the results of the study by Steensberg et.al, six healthy male subjects were asked to do knee extensor exercises for 180 minutes. The result was that IL-6 and TNF-mRNA were both detected in the muscle sample at rest. IL-6 levels increased 100-fold during exercise, and there was no significant increase in arterial plasma TNF-alpha mRNA (Steensberg et.al, 2002).

Direct muscle contraction stimulates the release of IL-6, which is an anti-inflammatory cytokine, decreasing the production of TNF alpha and IL1 beta, in the acute phase and during cell proliferation. Moderate intensity exercise (MIT) is effective in reducing body fat, which can prevent fat cell damage and cell hypoxia, so that proinflammatory cytokines (IL6 and TNF) are reduced through increased adiponectin secretion and increased anti-inflammatory cytokines. (Huldani et al., 2020).

Souglis et.al's study compared inflammatory response rates and muscle damage indexes between four popular elite-level team sports. 72 elite male players from four sports: 18 each: soccer, basketball, volleyball and handball, were asked to finish the match, while 18 non-atlet as a control. Blood samples were drawn before, immediately after, and 13 and 37 hours post-match. The results found changes in TNF α levels in all types of sports, including basketball, in blood samples taken immediately after, 13 hours after, and 37 hours after a competitive match, compared to baseline values (measured in the morning about 8 hours before the match). Football resulted in a 3–4 fold increase immediately after the largest inflammatory cytokine match, namely Tnf alpha tumor. Volleyball showed the smallest increase in inflammation and markers of muscle damage compared to the other three sports (Souglis et.al, 2015).

HMGB1

The results of this study indicated that there was no significant difference in HMGB1 levels after doing physical exercise in the form of running 12 minutes between trained basketball and not.

The human body has endogenous danger signals to prevent inflammatory responses secondary to the release of intracellular inflammatory factors into the extracellular area, namely the molecular pattern of associated damage (DAMP). High Mobility Group Box 1 (HMGB1) is a protein from DAMP which is a sign of muscle cell damage and causes the mobilization of immune cells to the site of trauma. (Fan and Evans, 2017). HMGB1 levels will return to their original concentration after 30 minutes of rest after exercising (Huldani, 2016).

Overall, DAMP triggers the release of massive cytokines including TNF- α , IL-6, and IFN. These mediators enhance the activation, proliferation, maturation and recruitment of immune cells at the site of trauma, causing indirect activation of adaptive and innate immune cells such as DC or T cells (Mickael et.al., 2018).

HMGB1 exposure to human monocyte cultures will stimulate several pro-inflammatory cytokines such as IL6, IL-8, TNF, and inflammatory protein macrophages-1 (MIP-1). The kinetic responses to the LPS mediated release of TNF and HMGB1 were very different. HMGB1 stimulated biphasic TNF release with a the second wave is delayed, whereas LPS mediated TNF release occurred in the initial monophasic mode (Huan et.al., 2015).

Goh et.al conducted a study for 3 weeks with a high-intensity training program in 7 healthy young men. Aerobic and endurance training is done for 3 consecutive days each week at the same time. Saliva and blood samples were collected Pre and Post, and 30 minutes after exercise each week, and 24 hours after exercise at the end of the session at week 3. The results showed plasma HMGB1 increased from Pre to Post exercise ($P < 0.05$). (Goh et.al., 2020).

While Kaki et.al done a similar study on 40 male Wistar rats (weight 220 ± 10.2 g) which were randomly divided into four groups: diabetic neuropathy, diabetic neuropathy + exercise, healthy + exercise, and healthy controls. Diabetes was induced by injection of STZ (50 mg / kg). After confirming the development of diabetic neuropathy with a behavioral test, the exercise group received 6 weeks of continuous aerobic exercise at an average intensity of 15 m / min for 30 minutes on a treadmill. The serum HMGB1 level was measured by ELISA and the concentration of malondialdehyde (MDA) and the action of superoxide dismutase (SOD) and catalase (CAT) enzymes in the spinal cord were determined by biochemical methods. Two-way ANOVA and Tukey's post hoc test were used for statistical analysis. The results showed that aerobic exercise significantly reduced HMGB1 protein levels and MDA concentrations as well as increased SOD and CAT enzyme activity be compared to the diabetic neuropathy group ($p < 0.05$). Also, HMGB1 and MDA levels were increased and SOD and CAT enzyme activity decreased in the diabetic neuropathy group ($p < 0.05$). Aerobic exercise is known to alter levels of the protein HMGB1 and oxidative stress and increase the sensitivity of nociceptors to painful agents. It is recommended that aerobic exercise be used as a non-prescriptive therapeutic intervention for diabetes patients to reduce neuropathic pain (Kaki et.al., 2020).

In another sports study, 34 and 36 nonprofessional female and male runners and half marathon finish each race, immediately blood samples were collected 1-2 days before, and 2-7 days after the race. After the race HMGB1 serum concentration increased significantly by 1.5 times (half marathon; 3.13 ± 1.63 ng / mL to 4.78 ± 2.1 ng / mL) and 2.3 times (marathon; 2.58 ± 1.58 ng / mL to 6.02 ± 2.18 ng / mL), corresponding to an increase in circulation sRAGE for a half marathon but not a full marathon. During the recovery period, the concentration of HMGB1 and sRAGE increases back to the previous training level (Bekos et.al, 2016).

Not all HMGB1 respond positively to every exercise. In recent research, HMGB1 plasma remains below the limit of detection (78 pg / mL) of testing after 1,200 km of plyometric training or cycling (Behringer et.al, 2016).

There was a decrease in serum HMGB1 at two weeks of Nordic walking exercise combined with vitamin D in elderly women, this suggests regular exercise can weaken the alarmin response in healthy adults (Gmiat, et.al, 2017).

CONCLUSION

This study showed that basketball-trained students had better VO₂ max scores than untrained basketball students. Twelve minutes of moderate aerobic exercise resulted in higher increases in cortisol levels in basketball-trained students than in untrained students, while TNF alpha levels were opposite. A very strong positive correlation between VO₂ max and whether or not young people are trained in basketball. The more trained a teenager, the greater the VO₂ max value. There is a very strong positive correlation between cortisol and whether adolescents are trained in basketball. The more trained a teenager, the greater cortisol levels will be. Twelve minutes of aerobic exercise can be used as a method to boost the immune system and show the homeostasis process in adolescents is working well.

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