



The Effect of Outdoor Sports and Green Outdoors on Lung Inflammation

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Abstract

This study aimed to analyze the effect of exercise in open spaces with exposure to pollution and activity in green open areas on lung inflammation. The true experimental method and The Randomized Posttest-Only Control Group Design were used in this study. Twenty-four adult male Wistar white rats weighing 200-250 grams aged 8-9 weeks were randomly divided using simple random sampling technique into four groups, NE (Non-Exercise) as control, NE+Pol (Non Exercise + Pollution Particulate Matter 2.5 >75 ppm, and Carbon Monoxide Meter <100 ppm), Ex (Exercise 5 times/week for four weeks), Ex+Pol (Exercise + pollution). The inflammatory marker Interleukin 6 (IL-6) was analyzed using Western Blotting. To determine the significant differences between groups, one-way ANOVA analysis and Post Hoc test were used. Based on the study results, it can be concluded that there is an effect of exercise in an open space with exposure to pollution on lung inflammation. And other results show that there is no effect of exercise in green open spaces on lung inflammation.

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INTRODUCTION

Inhaling exposure to air pollution such as particulate matter or excess diesel exhaust particles can cause airway and lung inflammation. It can exacerbate health problems such as asthma, chronic obstructive pulmonary disease, and heart disease (Shah & Balkhair, 2011). It is characterized by an increase in inflammatory cells, such as

neutrophils and macrophages, as well as inflammatory mediators including interleukin-6 (IL-6), interleukin-8 (IL-8), and prostaglandins (Gomes & Florida-James, 2014). Among these molecules, IL-6 deserves special attention because it is an essential mediator in developing the inflammatory process. Macrophages and IL-1 produce IL-6 and Tumor Necrosis Factor- α (TNF- α), stimulating local and systemic changes in the

inflammatory response (Febbraio & Pedersen, 2002; Steensberg et al., 2000). Exercising such as walking, jogging, and cycling done in open spaces has become the choice of the community to improve physical fitness. However, on the other hand, the effects of air pollution can also trigger physiological disturbances in the human body (Cutrufello, Rundell, Smoliga, & Stylianides, 2011).

Outdoor exercise with exposure to pollution has been shown to cause a decrease in lung function. Exposure to particulate matter 2.5 and ozone before or during exercise can impair lung function and increase lung inflammation (Giles & Koehle, 2014). At the level of submaximal training, breathing shifts from being dominated by the nose to being overwhelmed by the mouth, this transition causes the nasal filtering system to be bypassed, thereby potentially increasing the dose of air pollution that can worsen health (Niinimaa, Cole, Mintz, & Shephard, 1980). The results of a study conducted on trained runners who did an exercise with exposure to pollution revealed signs of lung inflammation and lung injury (Gomes, Stone, & Florida-James, 2011). Recent studies have also shown that physiological parameters (blood gases, cardiovascular and haematological parameters) are significantly affected by pollution after carrying out the Yo-Yo Intermittent Recovery Test (Aloui et al., 2017). This issue presents an interesting challenge to balance the benefits of exercise with the detrimental effects of air pollution on health.

In contrast to sports in open spaces with excessive exposure to pollution and many harmful effects on health, exercising in green open spaces is highly recommended. Green open spaces such as parks and urban forests that are not polluted can help reduce heat and ultraviolet radiation and improve air quality (Roy, Byrne, & Pickering, 2012). A systematic review found that parks' average daytime air temperature is 10 Celsius lower than in urban areas without greenery (Bowler, Buyung-Ali, Knight, & Pullin, 2010). Another study conducted in summer and warm temperatures found that the average garden air temperature reaches 20 to 30 Celsius with a surface temperature of 60 to 80 colder than non-green areas (Fintikakis et al., 2011). Green open spaces filled with trees and green plants, also known as the city's lungs, can produce oxygen and remove several airborne pollutants, including ozone, sulfur dioxide, and particulate matter (Grundström & Pleijel, 2014). Research in the United States revealed that urban forests are estimated to produce 67 tons of oxygen each year in the country (Nowak, Hoehn, & Crane, 2007). The results revealed that exercise in a pleasant environment such as a city park had a more significant effect on blood pressure, heart function, and mental health (Barton, Hine, & Pretty, 2009; Zubaida et al., 2021).

Based on the results of the studies above, sports in green open spaces such as city parks can be good sports places compared to sports in open spaces. This study aimed to analyze the effect of exercise in open areas

with exposure to pollution and exercise in green open spaces on lung inflammation.

METHODS

The method used in this study is a true experimental research method with The Randomized Posttest-Only Control Group Design (Ramadan & Juniarti, 2020). In this study, four groups were formed randomly. Group one as a control was not given exercise treatment, and without exposure to pollution (NE; Non-Exercise), group two was given no exercise treatment with pollution exposure (NE+Pol; Non Exercise + pollution), group three was given exercise treatment with pollution exposure (Ex+ Pol; Exercise + pollution), and group four was given exercise treatment without exposure to pollution (Ex; Exercise). After being treated, the lung inflammation results from the four groups will be measured, and the results will be compared.

Twenty-four adult male Wistar white rats weighing 200-250 grams aged 8-9 weeks from Biofarma Indonesia were randomly divided into four sample groups using a simple random sampling technique. The minimum sample size is determined based on Federer's formula, namely $(t-1)(n-1) \geq 15$, that t is the number of groups, while n is the number of samples per group so that $n \geq 6$. This study used six rats per group. Rats were put into clean cages with a length of 70 cm x width 50 cm x height 55 cm, and a maximum capacity of 6 pens could be used per cage. The cell maintenance every 3x a week was carried out

by replacing the husks of the rat cage pads, and the rats were given standard feed and given water ad libitum.

All samples adapted to the cage for one week, after which the samples in the Exercise and Exercise+pollution group adapted exercise on a treadmill for rats for one week with a running speed of 10 m/min. After adjusting, the Exercise and Exercise+pollution groups were treated with moderate-intensity running (20 m/minute) with a duration of 30 minutes. In the Non-Exercise+exposure to pollution and Exercise+pollution groups, they were exposed to pollution and poor air quality during the exercise. During exercise, pollution exposure was maintained using the Particulate Matter 2.5 >75 ppm and Carbon Monoxide Meter <100 ppm indicators. The Non-Exercise group as a control was not given any exercise treatment and was exposed to clean air. This protocol was performed five times/week for four weeks.

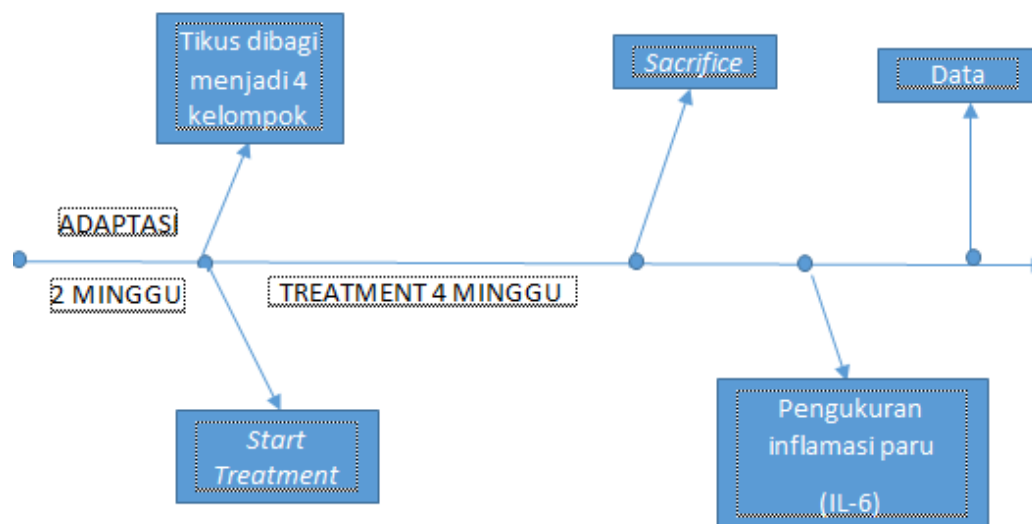
After being given treatment for 28 days, the sacrificed process was performed using anaesthetizing the rats with isoflurane, then cervical dislocation. After that, blood was taken from the heart and lungs, stored in a nitrogen solution, and then frozen in a freezer at -800 C. The blood and The lung was taken and terminated with the removal of the heart. After the sample organs are taken, IL-6 and TNF-alpha lung inflammation will be measured using Western Blotting. Western blotting is a technique for marking a protein on nitrocellulose, nylon, or another transfer membrane after the protein has been separated

by electrophoresis. The protein can then be detected by autoradiography, labelling with fluorescent compounds, labelling with protein-bound antibodies, lectins or other specific binding genes (Attwood, Campbell, Parish, Smith, & Stirling, 1998). The research timeline is shown in Figure 1.

Data analysis in this study used the Kolmogorov-Smirnov to analyze the normal

distribution of the data, then the Levene Test to analyze the homogeneity of the data. After that, one-way ANOVA analysis and Tukey's Post Hoc test were performed to investigate the significant differences between groups. P-value <0.05 was considered statistically significant.

Figure 1. Research Timeline



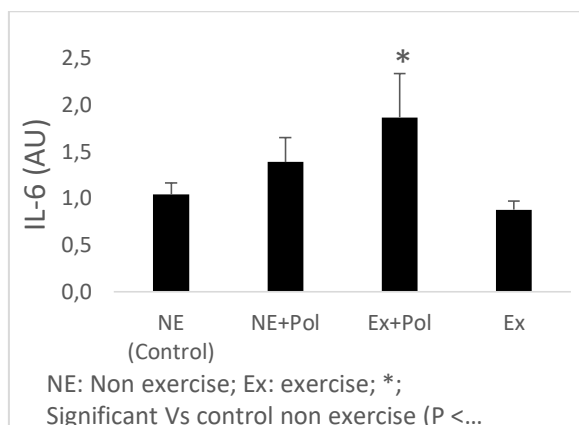
FINDINGS AND DISCUSSION

Finding

Figure 2 shows that the exercise experimental group with exposure to pollution had significantly higher levels of IL-6 inflammation than the control group (Ex+Pol: 1.87 AU; NE: 1.04 AU; $p=0.006$), with a mean difference of 0.83 AU. There was no significant difference between the experimental group exercise in green open spaces/without exposure to pollution and the

control group (Ex: 0.88 AU; NE: 1.04 AU; $p=0.839$), with a mean difference of 0.16 AU. Significant differences were seen between the experimental group of exercise with exposure to pollution and the experimental group of exercise in green open spaces/without exposure to pollution (Ex+Pol: 1.87 AU; Ex: 0.88 AU; $p=0.002$). Thus, it can be concluded that there is an effect of exercise in an open space with exposure to pollution on IL-6 inflammation in the lungs.

Figure 2. Effect of exercise with and without pollution exposure on IL-6 inflammation



Discussion

This study indicates that exercise in an open space with exposure to pollution can cause inflammation in the lungs, which is characterized by increased levels of inflammatory IL-6. The results of this study are consistent with the results of previous studies showing that exposure to diesel exhaust particles induces the systemic release of IL-6 levels, a cytokine that is considered a causative biomarker of the inflammatory process (Gimeno et al., 2011; Nemmar et al., 2010). Another study also revealed that exposure to diesel exhaust particles causes inflammation in the lungs, characterized by increased levels of IL-6, TNF-alpha and Keratinocyte Chemoattractant in rats who do moderate-intensity exercise for 60 minutes (De Paula Vieira et al., 2012; Putramadiansyah & Salahudin, 2021). Research by Devlin et al. (1999) also observed a significant increase in IL-6 inflammation after 4 hours of exposure to nitrogen dioxide followed by moderate-intensity treadmill exercise. In addition,

exposure to Particulate Matter 10 and black carbon was shown to increase TNF-alpha and Nuclear Factor-Kb in rats after 60 minutes of light-intensity exercise (Fashi, Alinejad, & Mahabadi, 2015). However, the mechanism by which air pollution can cause inflammation is not fully elicited. A recent study conducted by Pasqua et al. (2020) investigated whether air pollution from the city center would affect inflammatory and cardiorespiratory responses during long (90 minutes) moderate-intensity exercise. The results showed that pro-inflammatory did not increase at 30 minutes to 60 minutes of exercise duration. Pro-inflammation, characterized by increased IL-1, IL-6, TNF-alpha and Vascular Endothelial Growth Factor, was seen to increase after exercise duration of more than 60 minutes under conditions of exposure to vehicle air pollution. These results are certainly different from this study and are debatable. In this study, the sample only did one exercise and only saw the acute effect of pollution exposure on lung inflammation. The example did

exercise 5 times per week for four weeks in this study. Further research is needed to analyze exercise's acute and chronic effects with pollution exposure on inflammation. In addition, the duration and intensity of exercise are essential factors in the inflammatory response.

Research conducted by Flouris et al. (2012) also did not find a significant increase in IL-6 levels after exposure to secondhand smoke for 1 hour, followed by maximal exercise lasting about 10 minutes. Another study conducted by Jacobs et al. (2010) also found no significant difference in serum IL-6 after 20 minutes of moderate-intensity cycling under conditions of exposure to pollution. This can happen because the diversity of components and concentrations of air pollutants can cause different biological effects (World Health Organization, 2005). In this study, inflammation of the lungs can be caused by inhalation of carbon monoxide (CO) from the respiratory tract during exercise in an open space with exposure to pollution. Carbon monoxide causes a decrease in lung function and oxygen distribution, which prevents haemoglobin from carrying oxygen, and causes the inability of muscles to meet energy requirements during exercise (Giles & Koehle, 2014).

It has been shown that doing endurance exercise regularly provides many health benefits, such as increasing the rate of mitochondrial respiration, improving heart function, lowering obesity and blood pressure, and increasing longevity (Kokkinos & Myers,

2010). When done regularly and regularly, endurance training can also have a natural anti-inflammatory effect on lung diseases, such as asthma (Vieira et al., 2011), acute respiratory distress syndrome (Ramos et al., 2010; Syamsudin et al., 2021), and chronic obstructive pulmonary disease (Toledo et al., 2012). However, these various benefits can be obtained in an environment with good air quality and not polluted. The results of previous studies showed that samples who did moderate-intensity walking in a green background and lots of trees showed an increase in health markers at the molecular, physiological, and psychological levels, resulting in a systemic health improvement (Jane Elizabeth S. Thompson, 2012; Kuswari et al., 2021). These results are in line with this study which proved that the inflammatory markers IL-6 and TNF-alpha did not show an increase in samples who exercised in green open spaces/without exposure to pollution compared to samples who exercised with exposure to pollution.

The results of another study also showed no increase in TNF-alpha in samples that did exercise without being exposed to PM10 and black carbon compared to samples that did exercise with exposure to PM10 and black carbon (Fashi et al., 2015). Research conducted by De Paula Vieira et al. (2012) also showed that the inflammatory markers IL-6 and TNF-alpha were seen to be higher in samples who performed endurance sports with exposure to diesel exhaust particles compared to samples who exercised without exposure to

diesel exhaust particles. . Another study conducted by Gomes, Stone, & Florida-James (2011) also showed an increase in oxidative stress and lung inflammation characterized by increased IL-6 and CC16 in runners who ran 8 km with exposure to pollution. This study indicates that the inflammatory markers IL-6 and TNF-alpha in samples who exercise in green open spaces/without exposure to pollution are better than samples that exercise in open areas with exposure to pollution.

CONCLUSION

From the results of this study, it can be concluded that exercise in an open space with exposure to pollution on IL-6 inflammation in the lungs. There is no significant effect of exercise in green open spaces/without exposure to pollution on IL-6 inflammation in the lungs. Further research is suggested to analyze the acute and chronic effects of exercise with pollution exposure on inflammation and analyze the effect of duration and intensity of exercise when doing exercise with pollution exposure on inflammation.

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