ASIAN JOURNAL OF HEALTH AND APPLIED SCIENCES
2023, VOL. 2, NO. 2, 20 – 26
https://doi.org/10.53402/ajhas.v2i2.354

Effect of Avocado *(Persea americana M.)* Juice on Superoxide-Dismutase (SoD) and Interleukin-6 (IL-6) Levels

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ABSTRACT

Cigarette smoke contains oxidants or free radicals, about 4700 harmful chemicals. The high level of free radicals in the body triggers the emergence of Reactive Oxygen Species (ROS), which results in oxidative stress. This can occur if there is an imbalance between the number of oxidants and antioxidants. In this process, there is an O_2 leak that will turn into a superoxide radical (O_2^-) that can form pro-inflammatory cytokines such as IL-6. The purpose is to determine the effect of avocado juice administration on Superoxide Dismutase (SOD) levels and IL-6 levels in male Wistar rats exposed to cigarette smoke. This study used Post Test Only Control Group Design. Samples were taken from 20 male rats that entered the inclusion criteria and divided into four random groups, namely K1, K2, K3, and K4. K1 is the control group with standard feed without exposure to cigarette smoke; K2 is the treatment group with standard feed given exposure to cigarette smoke, K3 was given standard feed plus avocado juice at a dose of 2.7 grams /day. Meanwhile, the K4 group was given standard feed plus avocado juice at a 5.4 grams/day dose. However, both were given 14 days of exposure to cigarette smoke. On the 15th day, male Wistar rats were drawn blood to examine the SOD using spectrophotometry and IL-6 using ELISA. Data were analysed using the One-Way Anova different test and continued with the Post hoc Tukey test. The average SOD levels were highest in the K1 with a value of 83,23, and IL- 6 levels were highest in the K2 with a value of 79,93. The One-Way ANOVA test obtained results that SOD and IL-6 levels in each group with a value of $P<0.05$, which means that there were significant differences in influence. Giving avocado juice has an effect on increasing levels of SOD and decreasing levels of IL-6.

KEYWORDS

Avocado Juice; Cigarette Smoke Exposure; SOD; IL-6

Received: June 11, 2023
Accepted: September 1, 2023
Published: September 20, 2023

Introduction

Smoking is a lifestyle habit that affects human health. Cigarette smoke contains around 1015-1017 oxidants or free radicals and around 4700 harmful chemicals, including aldehydes/carbonyls, NO_2 and SO_2 (Suryadinata, 2018). The high free radicals in the body trigger the emergence of Reactive Oxygen Species (ROS), which results in oxidative stress; this can occur when there is an imbalance between the number of oxidants and antioxidants. Antioxidants that act as the first defense system against free radical compounds are Superoxide Dismutase (SOD). SOD is the first antioxidant enzyme in the defense mechanism against superoxide anions (Parwata, 2015).

Free radicals in the human body result from oxidation and cell-burning processes that occur during breathing, cell metabolism, inflammation, and exposure to pollution (Suryadinata, 2018). Free radicals in the body will react with cell molecules to obtain electron pairs to become more stable. However, body cell molecules whose electrons are taken will turn into free radicals. This reaction will continue to occur in the body. Not stopping it will cause oxidative stress, which causes inflammation, DNA or cell damage, and causes various degenerative diseases (Parwata, 2015).

Regarding the effect of reducing SOD and interleukin-6 in avocado, avocado is a fruit rich in nutrients and antioxidants such as flavonoids, saponins, alkaloids and tannins. Apart from that, it also contains other compounds that are beneficial to the body, such as carotenoids, Monounsaturated Fatty Acid (MUFA), minerals, protein, vitamin A, B vitamins, vitamin C, vitamin E, which can protect cells from oxidative damage caused by ROS (Sadewo GB, 2019). The content of flavonoids in avocados can capture ROS and inhibit the action of enzymes that produce ROS so that DNA or cell damage can be prevented and oxidative stress does not occur again (Parwata, 2015). Previous research stated that avocado seed extract has potential when used as an anti-inflammatory agent (Tinesya D, 2019).

In addition, not only research on avocados. However, research in Taiwan states smokers' SOD levels are relatively lower than non-smokers (Jenifer HD, 2015). The decrease in SOD concentration is associated with the ability of ROS to oxidize proteins, including enzymes, causing a loss of enzymatic activity and SOD function (Phaniendra A, 2015). Several studies have reported that smoking behaviour can increase interleukin-6 levels in serum (KIS, 2019) - inflammation such as IL-6 (Aldaham S, 2015). This study is not the same as previous studies, which stated that there was no significant difference in the mean IL-6 serum levels between smokers and non-smokers (KIS, 2019).

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Not much research has been done on the effect of avocado juice on enzymatic and inflammatory antioxidant markers in cigarette smoke exposure, so it is necessary to measure SOD and IL-6 levels as markers of enzymatic and inflammatory antioxidants. This study aims to prove the effect of avocado juice on SOD levels and IL-6 levels in male Wistar rats exposed to cigarette smoke.

Literature review

Superoxide Dismutase (SOD) is a metalloenzyme containing copper, zinc, or iron atoms formed in the cytosol and manganese-containing ones formed in the mitochondrial matrix (Ighodaro OM and Akinloye OA, 2018). SOD is an enzyme that functions as a catalyst for the superoxide anion free radical dismutase reaction (O2-) to hydrogen peroxide (H2O2) and is converted into water molecules by glutathione peroxidase and catalase enzymes (Nurhayati S, Kisnanto T, Syaifudin M, 2011). SOD is an antioxidant enzyme with a strong effect and is the body's first defense against free radicals (Kristiningrum E, 2018). The SOD enzyme can be found in red blood cells, kidneys, liver, brain, testes, heart muscle, pancreas, and lungs (Munawwaroh R, Bintari YR, Purnomo Y, 2019).

SOD is an endogenous antioxidant that can inhibit oxidative stress. SOD is present in the cytosol and mitochondria, which will convert reactive superoxide radicals (O2-) into hydrogen peroxide (H2O2) (Wahyuni, S, 2015). Peroxides are catalyzed by the enzymes catalase (CAT) and glutathione peroxidase (GPx) (Zulham, 2020). The CAT enzyme converts 2 H2O2 molecules into 2 H2O and O2.15 (Widayati, E, 2012). GPx enzymes in erythrocytes and other tissues catalyze the destruction of H2O2 and hydroperoxide lipids using reduced glutathione (GSH), protecting membrane lipids and hemoglobin from oxidation by H2O2, thereby preventing hemolysis caused by peroxide attacks. GSH will be oxidized to oxygenated glutathione (GS-SG). GS-SG must be reduced back to GSH by the glutathione reductase (Gred) enzyme to make it available for GPx enzyme action. H2O2, which is not converted to H2O, will form reactive hydroxyl radicals (OH+) (Werdhasari, A, 2014).

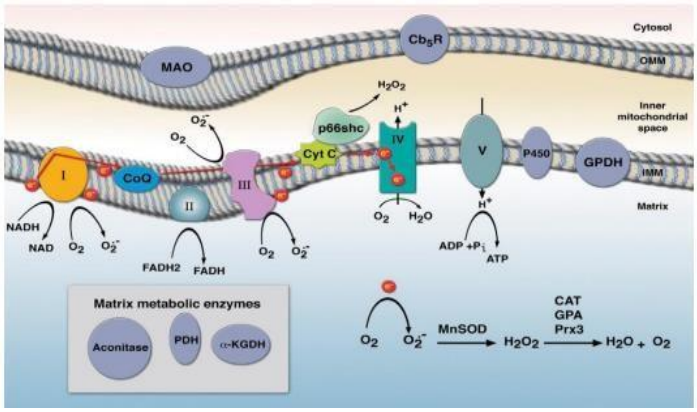

Figure 1. SOD Mechanism of Action (Wahyuni, N., 2015)

In Figure 1, it can be seen that the O_2* (superoxide radicals) produced in the change of NADH to NAD, $PADH_2$ to PADH is converted to H_2O_2 by MnSOD, and then the product H_2O_2 is converted to H_2O and O_2 by Catalase. SOD consists of 1) Copper Zinc Superoxide dismutase, a dimeric protein with two identical subunits linked non-covalently. Cu, Zn SOD plays an important role as the body's defense system against free radicals. Located in the cytoplasm and organelles with a size of 32,000 kDa. 2) MnSOD. Works as the main antioxidant in inhibiting the work of Superoxide Dismutase in mitochondria. Mn-SOD sized 40,000 kDA consisting of 4 subunits with manganese atoms. This type is synthesized mostly in the extracellular fluid by only a few cells, for example, endothelial cells and fibroblasts. 3) Fe-SOD. Enzymes are found in prokaryotes, plants, and bacteria. Three iron ions are bonded to three histidine, one aspartate, and one water molecule.

Various factors, including the content of cigarette smoke, influence SOD levels. The content of cigarette smoke is a source of free radicals that come from outside the body (exogenous). The body's natural mechanism in dealing with free radicals is by releasing endogenous antioxidants in the form of enzymes located in the body's tissues. The first line of antioxidant enzymes in efforts to protect against free radicals is (SOD) (Tazuyyun S, 2020).

Interleukin-6 (IL-6) is included in one of the pro-inflammatory cytokine groups so that this cytokine can be used as an indicator to assess the level of inflammation experienced by blood vessel endothelial cells (Yuniarti, E, 2014). In addition, interleukin-6 (IL-6) is also a pleiotropic cytokine that regulates cell growth and interactions between cells and triggers specific and non-specific immune reactivity. Interleukin-6 is secreted by T cells, macrophages, osteoblasts, blood vessels, endothelial cells, and smooth muscle cells to stimulate the immune system (Masfufatun M, Tania POA, Raharjo LH, Baktir A, 2018). In addition, IL-6 influences various cellular actions, including metabolic factors, effects on platelets, and coagulation factors. IL-6 plays an important role in atherosclerotic plaque, and its level increases in this event (6. Cahyani KIS, 2020). IL-6 circulates in multiple glycosylated forms with sizes varying from 22-27 kDa (Siagian D, 2018).

The normal value of serum interleukin-6 (IL-6) is < 4 > 4 pg/ml, which can be increased. Increased levels of IL-6 indicate an inflammatory process (25. Niu W, Liu Y, Qi Y, Wu Z, Zhu D, Jin W, 2012). An inflammatory process helps T lymphocytes divide into Th1, which produces proinflammatory cytokines interferon-γ (IFN-γ), tumour necrosis factor-α (TNF-α), tumour necrosis factor-β (TNF-β), interleukin-1, interleukin-6, interleukins -8, interleukin-12 which functions to activate the cellular immune system and the non-specific immune system. Th2 produces anti-inflammatory cytokines, namely interleukin-4 and interleukin-

10, which activate the humoral immune system (26. Wahyuniati, N and Maulana, R, 2015). IL-6 cytokines function as proinflammatory and anti-inflammatory secreted by T cells and macrophages. The cytokine interleukin-6 (IL-6) plays a role in stimulating the immune system's response to certain microbes, such as Mycobacterium leprae through pathogen en associated molecular patterns (PAMPs), which then bind to pattern recognition receptors (PRRs) and Toll-like receptors (TLRs) (Masfufatun M and Tania POA, 2018). The cytokine interleukin-6 stimulates hepatocytes to produce acute phase protein (APP) and cerebrospinal fluid (CPS) to stimulate progenitors in the bone marrow to produce neutrophils. In addition, interleukin-6 also stimulates the growth and differentiation of B cells into mast cells, producing antibodies for the specific immune system. Interleukin-6 is a growth factor (GF) in neoplastic plasma cells (myeloma). Interleukin-6 (IL-6) has a molecular weight between 21-28 kD, depending on ongoing processes such as glycosylation and phosphorylation. Through this process, the biological activity of interleukin-6 and its presence in specific tissues can occur. Interleukin-6 peptide consists of 212 amino acids with a gene located on chromosome 7p21 with five exons and four introns. Interleukin-6 is secreted by various heterogeneous proteins with a molecular weight of 19-70 kD, with the dominant isoform ranging from 23-30 kD. The interleukin-6 polypeptide binds to different carrier proteins such as albumin and soluble interleukin-6 receptors (Kang S, Tanaka T, Narazaki M, and Kishimoto T, 2019).

Interleukin-6 has two transmembrane molecules, namely interleukin-6R and the signal transducing subunit. The pleiotropic role of interleukin-6 as a pro-inflammatory and anti-inflammatory agent related to interleukin-6R. interleukin-6 is upregulated and expressed in small amounts, except in conditions of infection and trauma. The pro-inflammatory role of interleukin-6 occurs in chronic processes, such as autoimmune diseases, for example, lupus, leprosy, and rheumatoid arthritis. Increased interleukin-6 levels occur in acute bacterial infection, chronic inflammation, and bacteremia conditions. (Kang S, Tanaka T, Narazaki M, and Kishimoto T, 2019).

Cigarette smoke contains free radicals and contains around 4700 harmful chemicals (Suryadinata RV, 2018). Exposure to cigarette smoke is associated with tissue damage, increased oxidative stress, increased production of lipid products, reduced levels of antioxidants, and an increased risk of several chronic diseases. This is due to the composition in cigarette smoke, such as Reactive Oxygen Species (ROS) and phenol-rich glycoproteins, which stimulate macrophages directly, triggering the production of cytokines both in acute and chronic exposure (Nusa G and Widyastiti N, 2016).

Tissue damage due to cigarette smoke can trigger the release of inflammatory mediators such as cytokines. Inflammatory mediators such as IL-6, IL-1, and TNF-α increase leukocyte activation. Leukocytes release cytokines as an initial response to tissue damage. Furthermore, cytokines mediate a series of inflammatory processes and the immune system. Cytokines are glycoproteins derived from helper T cells, natural killer (NK) cells, and macrophages that play an important role in the body's response. Proinflammatory cytokines such as IL-1, IL-6, and TNF-α can affect the function and synthesis of other cytokines through a complex cytokine network (Irawati L, Acang N, and Irawati N, 2008). Exposure to cigarette smoke can trigger the production of IL-6 cytokines by leukocytes. IL-6 plays a role in the liver's CRP and other acute-phase protein processing.

Free radicals are molecules that have one or more unpaired electrons. These electrons will cause free radicals to become a highly reactive compound in body cells by binding to electrons in cell molecules. This reaction is referred to as oxidation. Antioxidant compounds can neutralize the effect of these free radicals. Antioxidants can prevent damage caused by ROS. Recently there has been evidence that natural antioxidants can prevent the occurrence of ROS. Natural antioxidants are contained in medicinal plants, fruits, and vegetables (Winarni S, Nissa C, and Purnami CT, 2019).

Avocado (Persea americana M.) contains antioxidant compounds and high nutritional content, both eaten directly or juiced (Diyanti R, 2018). Avocados have the same per cent oil composition as olive oil, protein, Fe, and Ca minerals and contain many antioxidants (Ziraluo YPB and Duha M, 2020). Avocados also contain bioactive molecules that protect human body cells against free radicals. The phytochemical analysis also showed the presence of alkaloids, saponins, terpenoids, phenolics, flavonoids, and tannins in avocados (Asngad A and Subiakto DW, 2020).

Method

Participants

The type of research used is experimental research. The research design was the Post Test Only Control Group on experimental male Wistar Rats. Samples were taken from 20 male rats weighing 150-200 grams and eight weeks old. The variables used are the independent variable: avocado juice, the dependent variable: levels of SOD and interleukin-6; and the precondition variable: cigarette smoke.

Instruments

Research tools: 1) Rat cages with feed containers measuring W: 40 cm, W: 30 cm, H: 30 cm. 2) Rat scales (Nigushi Scale). 3) Fumigation cage. 4) Beaker glass. 5) Gloves. 6) Eppendorf tube. 7) Digital camera. 8) Spectrophotometer. 9) 5 ml syringe. 10) Aerator hose. 11) Glass pipette. 12) Biovision SOD Spectophotometry/Colorimetric Assay Kit. 13) Serum IL-6 kits. 14) Biofin antibodies. 15) HRP conjugates. 16) Substrate reagent. 17) Stop the solution. 18) Electric blender.

The experimental animals used in this study were male Wistar rats aged eight weeks with a body weight of 150-200 grams. Male Wistar rats are the result of animal development obtained from the Laboratory of the Center for Food and Nutrition Studies (PSPG) Gajah Mada University, Yogyakarta; as many as 20 male Wistar rats were selected from the results of propagation for research purposes.

Data analysis

In this study, data on the average SOD and IL-6 level were presented in descriptive and tabular forms. Next, the data were tested for normality using the Shapiro-Wilk test, and the data homogeneity test was tested with the Levene test. The distribution of data on SOD and IL-6 levels was normal and homogeneous (uniform), so it was followed by a one-way ANOVA parametric test (One Way ANOVA) with a p-value <0.05, then continued with Tukey's posthoc test.

Results

This study used a sample of 20 male Wistar rats with a body weight of 150-200 grams divided into four groups, each consisting of 5 male Wistar rats, namely one control group and three treatment groups. Group 1 is the control group with standard feed without exposure to cigarette smoke; Group 2 is the treatment group with standard feed given exposure to cigarette smoke; Group 3 is the treatment group given standard feed and avocado juice with a dose of 2.7 gram/200-gram body weight of rats/day exposed to cigarette smoke, and group 4, namely the treatment group, was given standard feed and given avocado juice at a dose of 5.4 gram/200-gram body weight of rats/day exposed to cigarette smoke. On the 15th day, SOD and IL-6 levels were examined. SOD examination using spectrophotometry and IL-6 examination using ELISA. Research on giving avocado juice (Persea americana M) on SOD levels and IL-6 levels in male Wistar rats exposed to cigarette smoke for 14 days. The results of these studies are listed in Table 1.

Table 1. Measurement results of SOD levels (%) and IL-6 (pg/ml)

	Variable	**Group**				***P-Value***
		K1	K2	K3	K4	
SOD (%)	Mean ± SD	83,23 ± 3,97	28,23 ± 3,50	42,06 ± 3,04	73,53 ± 3,28	
	Shapiro Wilk	0,980*	0,899*	0,754*	1,000*	
	Levene Test					0,927[+]
	One Way Anova					0,000
IL-6 (pg/ml)	Mean ± SD	32,34 ± 0,59	79,93 ± 0,69	50,94 ± 0,88	38,68 ± 0,65	
	Shapiro Wilk	0,747*	0,926*	0,853*	0,784*	
	Levene Test					0,927[+]

SOD Level

Table 1 shows that the highest average SOD level is in the control group, namely 83.23 ± 3.97, while the lowest average value is in group 2, with a value of 28.23 ± 3.50. SOD levels between groups were normally distributed (p>0.05) and homogeneous (p>0.05). SOD levels between groups using the one-way ANOVA test differed significantly (p <0.05). The SOD levels between the two groups showed a significant difference between K1 and K2, K3, and K4. To find out which groups were significantly different, a post hoc test was carried out with the Tukey test.

Results of IL-6 (Interleukin-6) Levels

Table 1 shows that the highest average IL-6 level is in group 2 with a value of 79.93 ± 0.69, while the lowest average is found in group 1 with 32.34 ±. IL-6 levels between groups were normally distributed (p>0.05) and homogeneous (p>0.05). IL-6 levels between groups using the one-way ANOVA test.

Table 2. Results of Analysis of SOD Levels with Post Hoc Tukey Test

	K2	K3	K4
K1	0,000	0,000	0,002
K2		0,000	0,000
K3			0,000

Information:
K1 : 5 male Wistar rats were given standard feed for 14 days
K2 : 5 male Wistar rats treated with standard feed and exposed to cigarette smoke for 14 days
K3 : 5 male Wistar rats were given standard feed treatment with the addition of avocado juice 2.7 grams/200 grams BW rats/day
K4 : 5 male Wistar rats were given standard feed treatment with the addition of avocado juice 5.4 grams/200 grams BW rats/day

Based on Table 2, it was found that there was a significant difference between each group in SOD levels (P <0.05). The difference is found in K1 and K2 with a value of 0.000, K1 and K3 with 0.000, and K1 and K4 with a value of 0.002. K2 with K3 with a value of 0.000, K2 with K4 with a value of 0.000, and K3 with K4 with a value of 0.000. Based on the data above, it can be concluded that each group has a significant difference in SOD levels.

Table 3. Results of Analysis of IL-6 Levels with Post Hoc Tukey Test

	K2	K3	K4
K1	0,000	0,000	0,000
K2		0,000	0,000
K3			0,000

Information:
K1 : 5 male Wistar rats were given standard feed for 14 days
K2 : 5 male Wistar rats treated with standard feed and exposed to cigarette smoke for 14 days
K3 : 5 male Wistar rats were given standard feed treatment with the addition of avocado juice 2.7 grams/200 grams BW rats/day
K4 : 5 male Wistar rats were given standard feed treatment with the addition of avocado juice 5.4 grams/200 grams BW rats/day

Based on Table 3, it was found that there was a significant difference between each group in IL-6 levels (P <0.05). The difference is in K1 and K2 with a value of 0.000, K1 and K3 with a value of 0.000, and K1 and K4 with a value of 0.000. K2 with K3 with a value of 0.000, K2 with K4 with a value of 0.000, and K3 with K4 with a value of 0.000. Based on the data above, it can be concluded that each group has a significant difference in IL-6 levels.

Discussion

Cigarette smoke contains free radicals and high free radicals in the body trigger the emergence of Reactive Oxygen Species (ROS), which results in oxidative stress; this can occur if there is an imbalance between the amount of oxidants and antioxidants. The treatment group exposed to cigarette smoke daily on as many as four sticks was K2, K3, and K4. The results of examining SOD levels in the cigarette smoke exposure group without avocado juice were lower compared to the control group, as shown in Table 1, which shows that exposure to cigarette smoke can reduce SOD levels. This is caused by exposure to cigarette smoke for 14 days can cause oxidative stress conditions. When high ROS concentrations occur during prolonged exposure to cigarette smoke, causing the SOD antioxidant defences are unable to neutralize ROS resulting in cell and tissue damage.

The SOD level in the group given avocado juice at a dose of 5.4 grams/200 gr/day and exposure to cigarette smoke was higher than the group given avocado juice at a dose of 2.7 grams/200 gr/day and exposed to cigarette smoke. Although the SOD level in the 5.4 g/200 gr/day dose group was the highest, it was lower than the control group, as shown in Table 5.1. This is because the control group was not exposed to cigarette smoke but was still given standard feed, so the SOD level had the highest value. Meanwhile, the group given avocado juice at a dose of 5.4 gr/200 gr/day and exposure to cigarette smoke was higher than the group given avocado juice at a dose of 2.7 gr/200 gr/day and exposed to cigarette smoke. The higher the dose of avocado juice affected, the higher the SOD level. The content of flavonoids, vitamins A, C, and E, can increase antioxidant enzymes and reduce peroxide content.

The study results are from previous studies, which stated that the ethanol extract of avocado seeds (Persea americana Mill) in the flavonoid group is categorized as a very strong antioxidant (Hasan T and Pharmacy PS, 2022). This is because the content of flavonoids can reduce the formation of free radicals in the body. The results of the study found that IL-6 levels in the smoke exposure treatment group without avocado juice (K2) were higher than the control group (K1), the group given avocado juice at a dose of 2.7 g/BB 200 gr/day and exposed to smoke. Cigarettes (K3) and group (K4) with a dose of avocado juice 5.4 g/BB 200 gr/day, as shown in Table 1. This is because exposure to cigarette smoke can hurt the body, namely an increase in free radicals. The body's large number of free radicals will cause tissue structural abnormalities related to the inflammatory response, namely decreased IL-6. IL-6 levels in the group given avocado juice at a dose of 5.4 grams decreased significantly compared to the group given avocado juice at 2.7 grams, as shown in Table 1. Avocado juice administration was shown to suppress IL-6 as a pro-inflammatory cytokine which can lead to inflammation due to oxidative stress conditions due to exposure to cigarette smoke.

The content of flavonoids in avocados can inhibit the action of enzymes that produce ROS so that cell and tissue damage can be prevented and oxidative stress does not occur again. The content of vitamins A, B, C, and E can protect cells from oxidative damage caused by ROS. The results of this study are in accordance with research which states that the ethanol extract of avocado seeds has potential when used as an inflammatory agent in carrageenan-induced hamsters. Types of compounds that can function as anti-inflammatory are flavonoids, this is because the content of flavonoids can counteract free radicals which cause the appearance of an inflammatory response (Tinesya D, Andhita N, and Vidmar R, 2019). The limitations of this study were not measuring ROS levels which are a factor causing oxidative stress, not carrying out a histopathological examination of tissues due to exposure to cigarette smoke, and not carrying out an examination to determine total antioxidant capacity.

Conclusion

Based on the research, it can be concluded that: Administration of avocado juice (Persea americana M.) affects SOD levels in male Wistar rats exposed to tobacco smoke for 14 days. 2) Administration of avocado juice (Persea americana M.) changed IL-6 levels in male Wistar rats exposed to tobacco smoke for 14 days.

Funding

This research received no specific grant from any funding agency in the public, commercial, or not-for-profit sectors.

References

Asngad A and Subiakto DW. (2020). Potensi Ekstrak Biji Alpukat Sebagai Hand Sanitizer Alami : Literatur Review. *J Fak Kegur dan Ilmu Pendidik*.;6(2):106-110. doi:10.23917/bioeksperimen.v5i1.2795

Diyanti R. (2018). *Daya Hambat Ekstrak Etanol Buah Alpukat (Persea Americana Mill) Terhadap Pertumbuhan Candida Albicans*.

Hasan T, Farmasi PS. (2022). Uji Aktivitas Antioksidan Ekstrak Biji Alpukat (*Persea Americana* Mill .) Asal Enrekang.166-175.

Irawati L, Acang N, Irawati N. (2008). Artikel Penelitian Ekspresi Tumor Necrosis Factor-Alfa (Tnf- A) Dan Inter Leukin-10 (Il-10) Pada Infeksi. *Maj Kedokt Andalas*.;10

Ighodaro OM and Akinloye OA. (2018). First line defence antioxidants-superoxide dismutase (SOD), catalase (CAT) and glutathione peroxidase (GPX): Their fundamental role in the entire antioxidant defence grid. *Alexandria J Med*. ;54(4):287-293. doi:10.1016/j.ajme.2017.09.001

Kang S, Tanaka T, Narazaki M, Kishimoto T. (2019). Targeting Interleukin-6 Signaling in Clinic. *Immunity*. ;50(4):1007-1023. doi:10.1016/j.immuni.2019.03.026

Kristiningrum E. (2018). Peranan SOD pada Tatalaksana Akne Vulgaris. *CDK–261*.;45(2):133-136.

Masfufatun M, Tania POA, Raharjo LH, Baktir A. (2018). Kadar IL-6 dan IL-10 Serum pada Tahapan Inflamasi di *Rattus norvegicus* yang terinfeksi *Candida albicans*. *J Kedokt Brawijaya*.;30(1):19. doi:10.21776/ub.jkb.2018.030.01.4

Munawwaroh R, Bintari YR, Purnomo Y, Munawwaroh R, Bintari YR, Purnomo Y. (2019). Efek Dekokta Daun Pulutan (*Urena lobata*) Terhadap Kadar Superoxide Dismutase (SOD) dan Malondialdehyde (MDA) Hepar Ikan Zebra (*Danio rerio*) Yang Dipapar Malathion Secara Kronik. *J Bio Komplementer Med*.;6(3):223-229.

Nusa G, Widyastiti N. (2016). Perbedaan Neutrophil-Lymphocyte Ratio Pada Subjek Bukan Perokok, Perokok Ringan Dan Perokok Sedang-Berat. *J Kedokt Diponegoro*.;5(4):903-910

Niu W, Liu Y, Qi Y, Wu Z, Zhu D, Jin W. (2012). Association of interleukin-6 circulating levels with coronary artery disease: A meta-analysis implementing mendelian randomization approach. *Int J Cardiol*; 157(2):243-252. doi:10.1016/j.ijcard.2011.12.098

Nurhayati S, Kisnanto T, Syaifudin M. (2011). Superoksida Dismutase (Sod) : Apa Dan Bagaimana Peranannya Dalam Radioterapi. *Bul Al*.;13(2):67-74. doi:10.23886/ejki.6.9670.

Simanjuntak E, Zulham. (2020). Superoksida Dismutase (SOD) dan Radikal Bebas. *J Keperawatan dan Fisioter*;2(2).

Siagian D. (2018). *Perbedaan Kadar Serum Interleukin–6 Pada Kanker Paru Perokok Dan Tidak Perokok.*. http://repositori.usu.ac.id/handle/123456789/8347

Sri Wahjuni MKDI. SUPEROKSIDA dismutase. Published online 2015.

Suryadinata RV. (2018). Pengaruh Radikal Bebas Terhadap Proses Inflamasi pada Penyakit Paru Obstruktif Kronis (PPOK). *Amerta Nutr*. ;2(4):317. doi:10.20473/amnt.v2i4.2018.317-324

Tazuyyun S. (2020). Pengaruh Pemberian Ekstrak Etanol Daun Kemangi (*Ocimum citriodorum*) Terhadap Kadar Superoksida Dismutase (SOD) Paru Tikus Wistar Setelah Paparan Asap Rokok. 000:137

Tinesya D, Andhita N, Vidmar R. (2019). Eksplorasi Potensi Ekstrak Biji Alpukat (*Persea americana*) Sebagai Agen Antiinflamasi. *LENSA (Lentera Sains) J Pendidik IPA*. ;9(2):52-56. doi:10.24929/lensa.v9i2.66

Wahyuniati N. (2015). Peran Interleukin-10 Pada Infeksi Malaria. *J Kedokt Syiah Kuala*. Published online :96-103

Werdhasari A. (2014). Peran Antioksidan Bagi Kesehatan. *Indones J Biotechnol Med*.;3(2):59-68. doi:10.22435/jbmi.v3i2.4203.59-68

Widayati E. (2017). Oxidasi Biologi, Radikal Bebas, dan Antioxidant. *J Maj Ilm Sultan Agung*. ;50(128).

Winarni S, Nissa C, Purnami CT. (2019). *Modul Makanan Kaya Antioksidan Untuk Peningkat Fertilitas*.https://docpak.undip.ac.id/1902/1/Modul.Makanan.Kaya.Antioksidan.Untuk.Peningka t.Fertilitas.pdf

Yuniarti E. (2014). Pengaruh Latihan Submaksimal Terhadap Kadar Interleukin-6 Pada Siswa Pusat Pendidikan Latihan Pelajar Sumatera Barat. *J Sainstek*. 2014;6(2):189-192.

Ziraluo YPB, Duha M. (2020). Diversity Study Of Fruit Producer Plant In Nias Islands. *J Inov Penelit*.;1(4):683-694.