

Effect of Combination of Kasturi Fruit Juice (*Mangifera casturi*), Vitamin E and Dha on Interleukin-6 (IL-6) Gene mRNA Expression and IL-6 Levels in Lung Inflammation of Rats (*Rattus norvegicus*) Due to Coal Dust Exposure

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ABSTRACT

The respiratory tract can experience inflammation if exposed to coal pollutants. The purpose of this study was to analyze IL-6 gene mRNA expression and IL-6 levels in serum formed in experimental animals given a combination of ripe Kasturi fruit pulp juice (dose 7.8 mg/kg body weight/day dissolved in 2 ml H₂O), vitamin E (dose 22 mg/kg body weight/day) and DHA (dose 60 g/kg body weight/day) exposed to coal dust at 10 pm (N10S CellMicroSieves™ BioDesign Inc. of New York, USA) given 25 mg/day/m³ in 1 hour per day for 14 days. The experimental animals were grouped into 5 groups, group 1 was the control group (no coal dust exposure and no supplements), group 2 was the coal dust exposure group, group 3 was the combination of ripe kasturi fruit juice and vitamin E with coal dust exposure, group 4 was the DHA group with coal dust exposure and group 5 was the combination of ripe kasturi fruit juice, vitamin E and DHA with coal dust exposure. Coal dust exposure was carried out for 14 days. Using the post only control design method, IL-6 gene mRNA expression and serum IL-6 levels as pro-inflammatory parameters were examined. Statistical tests of homogeneity, normality and one-way anova SPSS 25.0 were performed with $\alpha = 0.05$. The results showed that IL-6 gene mRNA expression and IL-6 levels in serum were established in all groups. The average IL-6 gene mRNA expression obtained by group 1 was 6.486 ± 0.272 , group 2 was 10.868 ± 0.258 , group 3 was 8.420 ± 0.199 , group 4 was 9.096 ± 0.103 and group 5 was 7.655 ± 0.121 . The average serum IL-6 level obtained by group 1 was 52.21 ± 3.02 pg/ml, group 2 was 104.93 ± 2.98 pg/ml, group 3 was 80.70 ± 2.81 pg/ml, group 4 was 90.85 ± 2.36 pg/ml and group 5 was 65.81 ± 2.54 pg/ml. Statistical tests on all groups showed homogeneity and normality ($p > 0.05$) and one-way anova and Bonferroni post hoc found $p = 0.000$. The conclusion of this study is that IL6 gene mRNA expression and IL-6 levels have been established. In the group given a combination of ripe Kasturi fruit juice, vitamin E, and DHA with coal dust exposure for 14 years, IL-6 gene mRNA expression and IL-6 levels in serum were found to be the lowest compared to the group exposed to coal dust and the difference was statistically significant at $p = 0.000$.

Keywords: Kasturi fruit juice, vitamin E, DHA, mRNA expression, IL-6 levels, coal dust.

1. INTRODUCTION

The development of coal utilization as energy continues to increase in the world (Erdogan et al., 2024), along with increasing coal dust pollution (Asif et al., 2022). These pollutants can cause several health problems, especially in the respiratory tract. The respiratory tract can experience inflammation if exposed to coal pollutants (Vanka et al., 2022). Further prevention efforts against lung inflammation can be done by providing optimal inflammation relief. Optimal inflammation relief is currently a combination of medicinal ingredients, including natural ingredients. Natural ingredients are an important alternative because they have relatively mild side effects (Patil et al., 2019). *Mangifera casturi* is one of 31 mango species

found in Kalimantan, Indonesia. Other characteristics of *Mangifera casturi* are dark brown stems with rough and sticky surfaces, green leaves, lanceolate shape, purplish brown fruit skin, bright yellow to orange pulp, distinctive smell, sweet slightly sour taste, and lots of fiber. This endemic plant also contains bioactive compounds that are used as medicines (Sukmana et al, 2020). The taxonomy of *Mangifera casturi* is as follows: kingdom: Plantae, division: Spermatophyta, subdivision: Angiosperm, class: Dicotyledoneae, order: Sapindales, family: Anacardiaceae, genus: *Mangifera*, species: *Mangifera casturi* (Sutomo et al., 2014). A combination of anti-inflammatory and antioxidant is required to produce optimal treatment (Panebianco et al., 2019).

The flesh of ripe Kasturi fruit is a natural ingredient that contains antioxidants in the form of vitamin C and other chemical compounds that are beneficial for reducing inflammation. These natural ingredients may be combined with vitamin E (antioxidant) and DHA (anti-inflammatory). A study will be conducted on the effect of the combination of ripe Kasturi fruit juice, vitamin E and DHA on reducing inflammation in coal pollution cases by analyzing the expression of IL-6 gene mRNA and IL-6 levels in serum. The purpose of this study is to analyze the mRNA expression of the IL6 gene and IL-6 levels in serum formed in experimental animals given a combination of ripe Kasturi fruit juice, vitamin E and DHA and exposed to coal dust for 14 days.

2. MATERIALS AND METHODS

This study was conducted on female white rats (*Rattus novergicus*) weighing 125-190 g, 2-3 months old and physically healthy. The experimental animals were grouped into 5 groups, namely group 1 as control (without coal dust exposure and without supplements), group 2 as control (only exposed to coal dust), group 3 as a combination of ripe Kasturi juice and vitamin E with coal dust exposure, group 4 as DHA group and group 5 as a combination of ripe Kasturi juice, vitamin E and DHA with coal dust exposure. The dose for ripe Kasturi fruit juice was 7.8 mg/kg body weight/day dissolved in 2 ml H₂O, vitamin E in tablet form (dl- α -tocopheryl acetate 200 mg (vitamin E 200 IU under the brand name Santa-e® 200 from Sanbe) at a dose of 22 mg/kg body weight/day, and DHA capsules (OM3HEART 600, each capsule containing 120 mg DHA) at a dose of 60 g/kg body weight/day. All supplements were administered orally via sonde. Coal dust exposure at 10 pm (N10S CellMicroSieves™ BioDesign Inc. of New York, USA) was administered at 25 mg/day/m³ for 1 hour.

All studies were conducted for 14 days. Using the post only control design method, IL-6 gene mRNA expression and serum IL-6 levels as proinflammatory parameters were examined. The study was conducted at the Laboratory of Immunology and Microbiology, Faculty of Medicine, Hasanuddin University, Makassar. Statistical tests of homogeneity, normality and one way anova were performed with SPSS 25. $0\alpha = 0.05$.

Analysis of Content, Antioxidant Potential, and Phytochemicals in Kasturi Fruit Meat

The content of ripe Kasturi fruit pulp was obtained from the analysis report of IPB Bogor in 2008 with certification number 208-08/f1/4.2.4/lja/itp. Antioxidant potential was carried out by DPPH method at the Biochemistry Laboratory, Faculty of Medicine and Health, Lambung Mangkurat University Banjarmasin. Furthermore, phytochemical examination was carried out with certain methods (table 2) at the Pharmacology Laboratory, Faculty of Medicine and Health, Lambung Mangkurat University Banjarmasin.

Cytokine test

After treatment, real-time PCR was performed on the animals to determine the IL6 gene mRNA expression profile using a Real-time PCR machine (CFX Connect system, Biorad Laboratories, Real Time PCR 96 well 0.1 ml, USA) and following the manufacturer's protocol. IL-6 gene mRNA expression was performed by ELISA method (ELISA Reader 270 Biomerieux kit, France) and followed the kit manufacturer's protocol. The study was conducted at the Laboratory of Immunology and Microbiology, Faculty of Medicine, Hasanuddin University, Makassar.

3. RESULTS

Table 1. Nutrient Composition of Ripe Kasturi Fruit Flesh

No.	Test	Unit	Analysis Result	Method
1.	Water content	grams/100 grams	83,98	Gravimetry
2.	Ash Content	grams/100 grams	0,42	Gravimetry

3.	Fat	grams/100 grams	1,14	Soxhlet
4.	Protein	grams/100 grams	0,58	Kjeldahl-Micro
5.	Carbohydrates	grams/100 grams	13,88	Based on differences
6.	Energy	Kcal/100 grams	68,1	Calculations
7.	Ca	grams/1000 grams	202,94	AAS English
8.	Fe	mg/1000 grams	15,82	AAS English
9.	P	grams/100 grams	0,03	Spectrophotometry
10.	Vitamin A	Unit Units/100 grams	2542,18	HPLC
11.	Vitamin C	grams/100 grams	0,46	Spectrophotometry

Source: IPB Bogor Analysis Report, 2008, certification number 208-08/FL/4.2.4/LJA/ITP

The sample contains 83.98% water. The high water content indicates that most of the weight of the sample consists of water. Ash is the inorganic residue that remains after a complete combustion process. The ash content represents the total minerals in the sample. The sample contains 0.58% protein, calculated based on the nitrogen content converted to total protein. The sample had a carbohydrate content of 13.88%, calculated by the difference method (total = 100% - moisture content - ash - protein - fat).

The sample has 68.1 calories of energy per 100 grams, calculated based on fat, protein, and carbohydrate content (with their respective calorie factors). The sample contains 202.94 mg of calcium per 1000 grams, an important mineral for bone health. The iron content in the sample was 15.82 mg per 1000 grams, important for hemoglobin production. The sample contains 0.03% phosphorus, important for bone health and energy metabolism. The sample contains 2542.18 IU (International Units) of vitamin A per 100 grams, important for eye health and immunity. Vitamin C content in the sample was 0.46% (or 460 mg per 100 grams). Vitamin C functions as an antioxidant and boosts immunity.

This method is used to measure the ability of compounds to capture or donate electrons to DPPH free radicals. DPPH radicals have a distinctive purple color, which will turn pale yellow when reacting with antioxidant compounds.

A wavelength of 517 nm was used to measure the absorbance of the DPPH solution using a spectrophotometer. IC50 is the concentration of antioxidant required to inhibit 50% of DPPH free radicals. The smaller the IC50 value, the stronger the antioxidant potential of a compound. Based on the classification of antioxidant potential:

- < 50 ppm** : Very strong activity
- 50–100 ppm** : Strong activity
- 101–150 ppm** : Medium activity
- 151–200 ppm** : Weak activity
- > 200 ppm** : Inactive or very weak

Table 2. Phytochemical Examination of Ripe Kasturi Fruit Flesh

No.	Compound Identification	Test	Results
1.	Flavonoids	Alkaline Reagent Test	+
2.	Phenol Alkaloids	Dragendroff Test	+
3.	Tannins	Gelatin test	+
4.	Phenol	Iron (III) chloride test	+
5.	Saponins	Foam test	+
6.	Antrakuinon	Interquinone test	-
7.	Steroids	Libermann Burchard Test	+
8.	Terpenoids	Salkowski Test	+

The sample contained flavonoids, which were identified using an alkaline reagent test. Flavonoids usually produce a yellow color, which can change in intensity when alkaline solutions are added. The sample also contained phenolic alkaloids, detected using Dragendroff reagent. Alkaloids are organic compounds that typically show an orange or red color with this reagent. The sample contains tannins, which can react with gelatin and cause a precipitate. Tannins are phenolic compounds with astringent properties. The sample contains phenols, which are detected with an iron(III) chloride test. Phenol gives a blue, green or purple color when reacting with this reagent. The sample contains saponins, which are characterized by the formation of a stable foam when shaken with water. Saponins are compounds that can reduce the surface tension of water. The sample does not contain anthraquinone compounds, which are usually tested using the interquinone test. If positive, this test shows a red color after the reaction. The sample contains steroids, which are identified by a green or blue color change when the Libermann-Burchard test is performed. The sample contains terpenoids, detected by the Salkowski test. The reaction usually results in a red ring or red coating on the solution.

Table 3. Results of IL-6 Gene mRNA Expression and Serum IL-6 Levels in Each Group

No.	Group	Not. Sample	Result RTPCR	ELISA (pg/ml) results
1.	K1 = Control Group (no exposure to coal dust and no supplement group)	Season 2	6,258 people	47,81
2.		Season 3	6,827 people	52,21
3.		Season 05	6,255 people	56,03
4.		Season 06	6,362 people	53,48
5.		Season 07	6,727 people	51,26
6.	K2 = Group Exposed to Coal Dust	Season 08	11,116 people	110,47
7.		Season 09	11,262 people	108,62
8.		S10	10.868	102,73
9.		S11	11,070 people	104,93
10.		S13	10.817	105,66
11.	K3 = Group that was given musk juice, vitamin E, and exposed to coal dust	S16	8,636 people	80,70
12.		S17	8,700 people	79,32
13.		S19	8,473 people	78,97

14.		S20	8,570 people	84,86
15.		Section 21	8.221	82,77
16.		Section 22	8.970	90,85
17.	K4 = Group given DHA and exposed to coal dust	S25	9,136 people	87,32
18.		S26	9.200	88,02
19.		S27	9,003 people	90,85
20.		S28	9,171 people	86,61
21.		S29	7.851	63,17
22.	K5 = Group given musk juice, Vitamin E, DHA and exposed to coal dust	S30	7,984 years	64,16
23.		S32	7,783 people	65,48
24.		S33	7,755 people	68,8
25.		English S34	7,656 people	62,19

Interleukin-6 (IL-6) is a proinflammatory cytokine that plays an important role in inflammatory and immune responses. IL-6 gene mRNA expression reflects the transcription level of the IL-6 gene, while serum IL-6 levels indicate the amount of IL-6 protein released in the body. The combined effect of diet on IL-6 gene mRNA expression suggests that certain diets may influence the transcription level of this gene. Diets with certain components, such as high fat or high antioxidants, may increase or decrease IL-6 expression, depending on their role in inducing or suppressing inflammation. Serum levels of IL-6 reflect the effect of diet on the release of IL-6 protein into the blood circulation. High levels of IL-6 in serum are often associated with chronic inflammation, oxidative stress or certain metabolic conditions.

In contrast, diets containing anti-inflammatory properties (such as polyphenol-rich or low-fat) can reduce serum IL-6 levels, suggesting a protective effect against inflammation. If the graph shows a decrease in IL-6 mRNA expression and serum IL-6 levels in a particular group, this indicates the diet has an anti-inflammatory effect. Conversely, if there is an increase in IL-6 mRNA expression and serum IL-6 levels, this indicates the diet may trigger inflammation.

4. DISCUSSION

Based on the results of the study, it is known that the flesh of ripe kasturi fruit contains vitamin C of 0.46 g/100 g (table 1.), this indicates that the flesh of ripe kasturi fruit contains antioxidants due to its vitamin C content. To prove its antioxidant power, the antioxidant potential test was conducted and the IC₅₀ value = 498.46 ppm was obtained. This result shows that the material has antioxidant power and is classified as a weak antioxidant. In addition, the flesh of ripe kasturi fruit was subjected to phytochemical examination and the results showed that it contains: flavonoids, alkaloids, tannins, phenols, saponins, steroids and terpenoids (table 2.). These phytochemical compounds can also act as antioxidants, namely flavonoids. Flavonoids are organic compounds in plant materials consisting of phenolic compounds that have more than one hydroxyl group (Halliwell & Gutteridge, 2015) and have antioxidant activity (Huynh, 2024). Thus, the flesh of ripe kasturi fruit contains sufficient antioxidants, including: vitamin C and flavonoids.

Coal dust is one of the air pollutants that has the potential to increaseEnglish: which can have an impact on the surrounding community (Wang et al., 2023). Coal dust is one type of dust that is highly toxic and has been shown to cause several health problems, especially lung function disorders (Arumdani et al., 2024). The inflammatory process due to coal dust exposure in this study can be explained as follows. At the beginning of the process, the entry of particulates (PM 10) through environmental exposure into the lungs and the subsequent development of respiratory complications (Figure 1.), which includes an increased influx of macrophages, neutrophils, T lymphocytes, and B cells (Vanka et al., 2022). The inflammatory process due to coal dust exposure in this study can be explained as follows. At the beginning of the process, the entry of particulates (PM 10) through environmental exposure into the lungs and the subsequent development of respiratory complications (bronchitis and emphysema), which includes an increased influx of macrophages, neutrophils, T lymphocytes, and B cells in Figure 1. (Vanka et al., 2022).

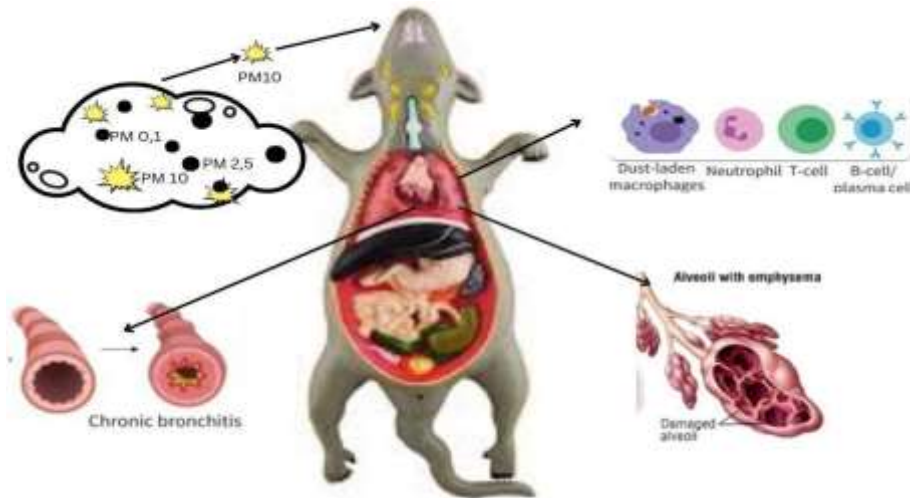


Figure 1. Inhalation Process of Coal Dust Exposure Pm 10

It further triggers a cascade of reactions, namely: cellular inflammation, Reactive Oxygen Species (ROS) and Reactive Nitrogen Species (RNS), cytokine production and DNA damage, which can lead to cell death and scar tissue formation (figure 2.) (Vanka et al., 2022). Inflammatory cytokine production includes: IL-6 (Riffo-Vasquez et al., 2000; Fachrurrodji et al., 2022). Research by examining the cytokine IL-6 in experimental animals can be carried out as an inflammatory variable (Khalil et al., 2018). One method that can be used to determine the concentration of IL-6 in serum from treated samples compared to (healthy) controls is ELISA (Ye et al., 2022).

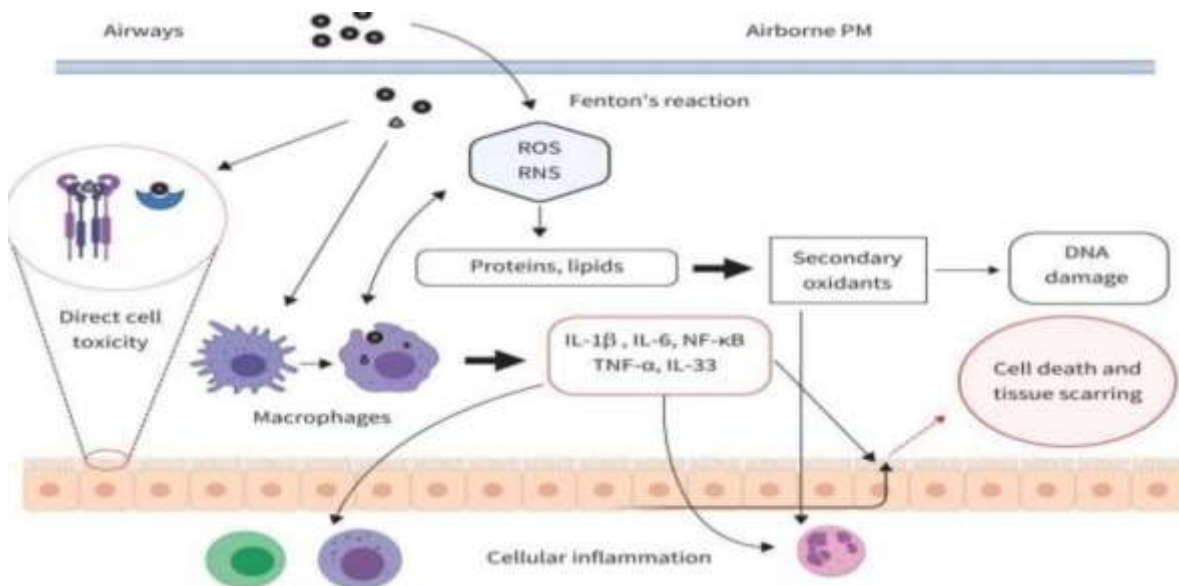


Figure 2. Formation of IL6 and ROS in Lung Inflammation Due to Exposure to Coal Dust PM 10

In the following study results, it was obtained that all treatment groups experienced the expression of the IL-6 gene mRNA. This is evidenced by the value of gene mRNA expression in each sample (table 3.). This expression shows the level of interleukin that is formed due to the inflammatory process. Furthermore, the average expression of IL-6 gene mRNA in each treatment group was presented compared to the control group. The average expression of IL-6 gene mRNA obtained from the results of the study can be seen in table 4. The results of statistical calculations from the results of the study on the expression of IL-6 gene mRNA and IL-6 levels in serum are as follows (One Way Anova & SPSS 25.0) in table 4.

Table 4. Statistical Calculation of IL-6 Gene mRNA Expression and IL-6 Levels in Serum

No.	Group	Mean expression of IL-6 gene	Average level of IL-6 (pg/ml)
1.	K1 = Control group (no exposure to coal dust and no supplement)	6.486 ± 0.272	52.21 ± 3.02
2.	K2 = Group exposed to coal dust	10.868 ± 0.258a	104.93 ± 2.98a
3.	K3 = Group that was given musk juice, vitamin E, and exposed to coal dust	8.420 ± 0.199ab	80.70 ± 2.81 inches
4.	K4 = Group given DHA and exposed to coal dust	9.096 ± 0.103abc	90.85 ± 2.36 ABC
5.	K5 = Group given musk juice, Vitamin E, DHA and exposed to coal dust	7,655 ± 0,121abcd	65.81 ± 2.54 abcd

Note: the value is presented as the mean ± SD; ap=0.00 compared to K1, bp=0.00 compared to K2, cp=0.00 compared to K3, and dp=0.00 compared to K4.

Judging from the average expression of IL-6 gene mRNA and IL-6 levels in serum, K1 obtained the lowest value compared to all treatment groups (K2, K3, K4 and K5), but K5 obtained the lowest value compared to all groups exposed to coal dust (K2, K3 and K4). Statistically, there were significant values between groups for the expression of IL6 gene mRNA ($p=0.000$) and serum IL-6 levels ($p=0.000$). The treatment group that was given ripe musk juice (K3 and K5) showed lower values than those who were not given (K2 and K4) ($p=0.000$), but higher than the control ($p=0.000$). The high value of these levels is in line with the subacute inflammatory process that occurs after exposure to coal pollutants for 14 days. A comparison between IL6 gene mRNA expression treatment groups can be seen in figure 3. and serum IL-6 levels in figure 4. below.

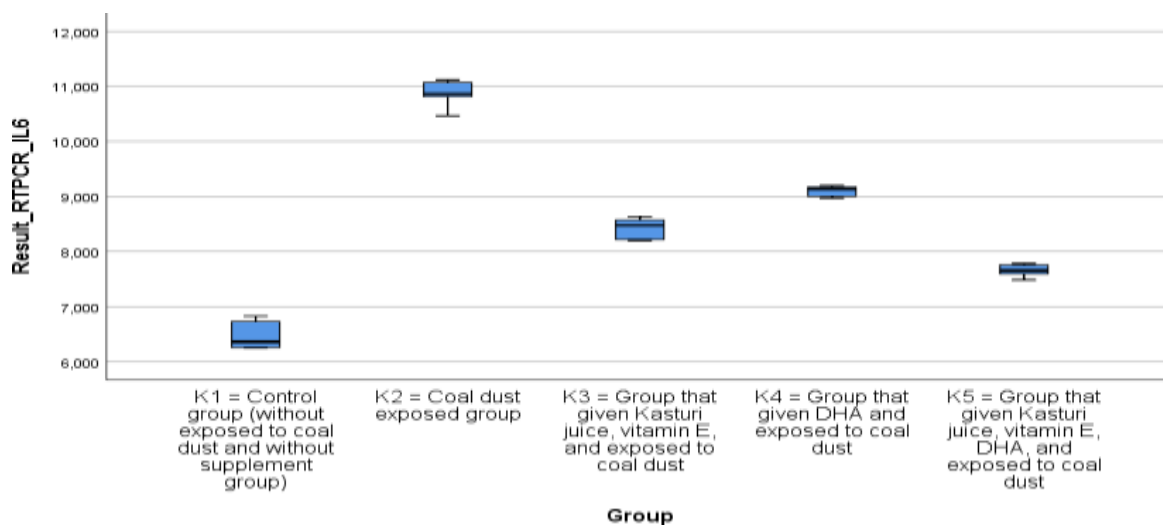


Figure 3. mRNA Expression of IL-6 Gene in Each Group

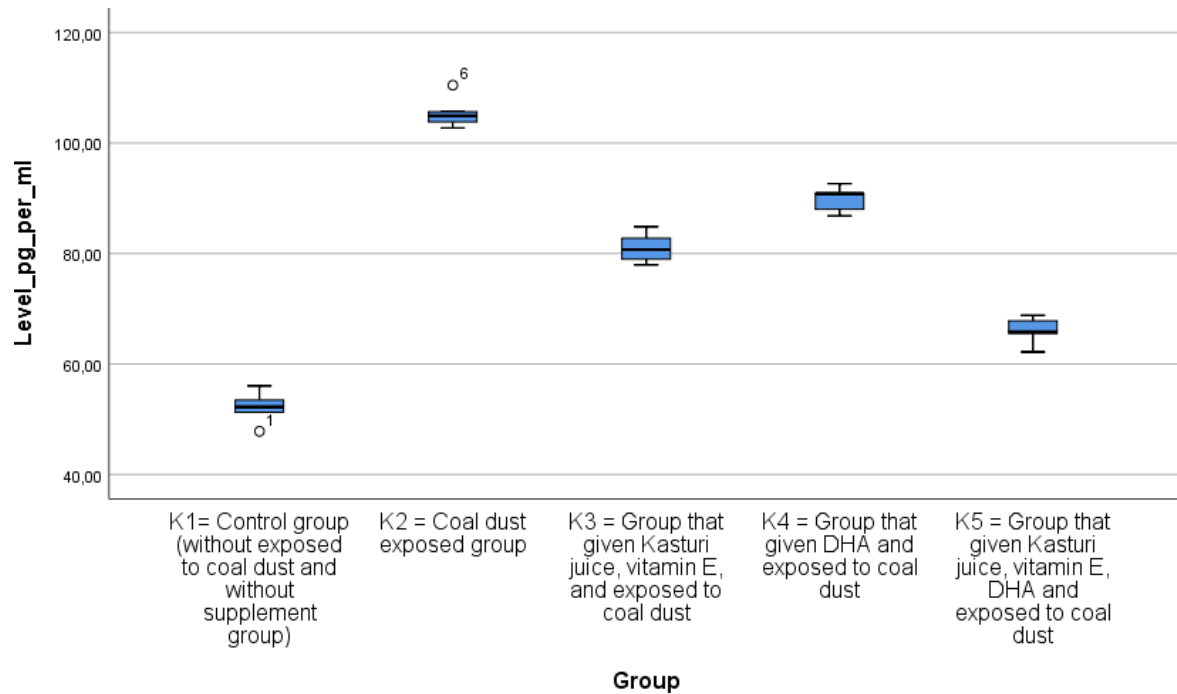


Figure 4. Levels of IL-6 in Serum in Each Group

Cytokinins play a role in the inflammatory process of the lungs as pro-inflammatory cytokines, including: IL-6 and TNF- α , both of which increase inflammation in response to coal dust. This has been proven by studies in rats exposed to coal dust for 1-4 weeks (Armutcu et al., 2007). IL-6 is a cytokine that has main activities in the form of: response in the acute phase, B cell proliferation, thrombopoiesis and cooperation with IL1 and TNF against T cells (Zhang & An, 2007). Increased expression of the IL6 gene can occur in inflammation (Santos et al., 2021).

Providing antioxidants vitamins C and E must go hand in hand, because the work of the two is synergistic (Murray et al., 2018). The administration of a combination of antioxidants from ripe musk juice (containing vitamins C) and E does not provide an optimal effect on inflammation (table 4.). Likewise, the administration of anti-inflammatory DHA alone does not have an optimal effect on inflammation. Musk juice and vitamin E with anti-inflammatory DHA produced an optimal effect on the decrease in mRNA expression of IL6 gene and IL-6 levels in serum in cases of inflammation due to exposure to coal dust, as it was the lowest expression of IL-6 gene mRNA and IL-6 levels in serum (table 4.) and the process can be seen in figure 5. The process has 4 steps, the first (1); Musk juice and vitamin E directly lower ROS and indirectly lower inflammation, the second (2); DHA lowers inflammation directly, the third (3); macrophages decrease, the fourth (4); The expression of the IL-6 gene mRNA decreases further the level of IL-6 in the serum decreases. Gene expression is associated with circulating levels of IL-6 (Lin et al, 2014). This is supported by Panebianco et al (2019), but the researchers fed the food directly with vitamin C and DHA, supplemented the diet with probiotics (*Bifidobacterium lactis*), and only measured IL-10.

5. CONCLUSION

The conclusion of this study is that the expression of the IL-6 gene mRNA and the level of IL-6 are known. In the group that was given a combination of ripe musk juice, vitamin E and DHA with exposure to coal dust for 14 days, the expression of IL-6 gene mRNA and IL-6 levels in serum was found to be the lowest compared to other groups exposed to coal dust and the difference was statistically significant at $p=0.000$. Based on research, the flesh of ripe musk fruit contains vitamin C of 0.46 g/100 g, indicating antioxidant properties. The antioxidant potential test produced an IC50 value of 498.46 ppm, which is classified as a weak antioxidant. In addition, the results of phytochemical examinations show that the flesh of ripe musk fruits contains compounds such as flavonoids, alkaloids, tannins, phenols, saponins, steroids, and terpenoids, where flavonoids play an important role as antioxidants.

Coal dust, as a harmful pollutant, triggers lung inflammation through the production of Reactive Oxygen Species (ROS), Reactive Nitrogen Species (RNS), cytokines (such as IL-6), and DNA damage. The results showed that the expression of IL-6 gene mRNA and IL-6 levels in serum increased due to exposure to coal dust. However, the group that was given ripe musk juice showed significant reductions in mRNA expression of the IL-6 gene and IL-6 levels compared to the group that was not treated, although the values were still higher than those in the healthy control group. The combination of antioxidants from ripe musk juice, vitamin E, and DHA provides an optimal effect in decreasing the expression of IL-6 gene mRNA and

IL-6 levels due to inflammation. These processes include ROS reduction, macrophage reduction, and inflammatory regulation. This proves the potential of the combination of musk juice and vitamin E with DHA as an anti-inflammatory therapy against the impact of coal dust exposure.

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REFERENCES

- [1] Abbas, AK, Lichtman, AH, Pillai, H. (2018). Cellular and molecular immunology, 9th edition. Philadelphia: Elsevier.
- [2] Armutcu, F., Gun, B. D., Altin, R., Gurel, A. (2007). Examination of lung toxicity, oxidant/antioxidant status and the effect of erdosteine in rats kept in an atmospheric cola mine. *Environmental Toxicology and Pharmacology*; 24: 106-113.
- [3] Arumdani, IS, Ilma, K. , Husni, SH., Rachmawati, I. (2024). PM 10 Toxicity of Coal Dust Particles and Factors Associated with the Incidence of Lung Function Disorders in Coal Mine Workers. *Tambusai Health Journal*, 5(2), 3074–3081 <https://doi.org/10.31004/jkt.v5i2.27734>
- [4] Asif, Z., Chen, Z., Wang, H., Zhu, Y. (2022). Update on air pollution control strategies for coal -fired power plants. *Clean Technology and Environmental Policy* (2022) 24:2329 –2347 <https://doi.org/10.1007/s10098-022-02328-8>
- [5] Erdogan, S., Pata, UK, Alola, AA. (2024). How do we position in reducing dependence on stone: Evidence from countries whose economies are heavily dependent on coal. *Energy Strategy Review* 54 (2024) 101444. <https://doi.org/10.1016/j.esr.2024.101444>
- [6] Fachrurroddji, Sidharta, BRA, Ariningrum, D., Suparyatmo, JB, Pramudianti, MID. (2022). Effect of Ginseng Extract on Serum Levels of Interleukin- in Diabetic Mellitus Patients Community Pneumonia Patients. *Indonesian Clinical Journal of Pathology and Medical Laboratories*; 3: 278 -284.
- [7] Halliwell, B., Gutteridge, J.M.C. (2015). Free radicals in biology & medicine, 5th edition. Oxford UK: Oxford University Press.
- [8] Huynh, TTH, Wongmaneepratip, W., Vangnai, K. (2024). The Relationship between the Chemical Structure of Flavonoids and Their Antioxidant Capacity in Preventing the Formation of Polycyclic Aromatic Hydrocarbons in a Heated Meat Model System. *Food* 13 (7), 1002; <https://doi.org/10.3390/foods13071002>
- [9] Khalil, A., Omran, H., Alsheikh, F. (2018). Pro-inflammatory and anti-inflammatory balance of cytokines in the livers of rats fed a high-fat diet exposed to gamma rays fractionated irradiation. *BMC Res Notes* (2018) 11:741 <https://doi.org/10.1186/s13104-018-3851-2>
- [10] Murray, RK, Granner, DK, Mayes, PA, Rodwell, VW. (2018). Harper's illustrated biochemistry 31st edition. New York: Mc Graw Hill Education.
- [11] Panebianco, C., Eddine, F. F. N., Forlani, G., Palmieri, G., Tatangelo, L., Villani, A., Xu, L., Accolla, R., Paziienza, V. (2019). The probiotic *Bifidobacterium lactis*, anti-oxidant vitamin E/C and anti-inflammatory dha attenuated lung inflammation due to exposure to pm 2.5 in rats. *kBenef Microbes*; 10(1): 69 -72.
- [12] Patil, KR, Mahajan, UB, Unger, BS, Goyal, SN, Belemkar, S., Surana, SJ, Ojha, S., Patil, CR. (2019). Inflammatory Animal Models for Anti-inflammatory Drug Screening: Implications for the Discovery and Development of Phytopharmaceuticals. *Int. J. Mol. Sci.* 2019, 20, 4367; doi:10.3390/ijms20184367
- [13] Riffo-Vasquez, Y., Pitchford, S., Spina, D. (2000). Cytokines in inflammation of the airways. *International Journal of Biochemistry and Cell Biology* Volume 32, Edition 8 Indonesian: 833-853. DOI: [https://doi.org/10.1016/S1357-2725\(00\)00029-7](https://doi.org/10.1016/S1357-2725(00)00029-7) *Journal of Management and Entrepreneurship*: 1907-5995
- [14] Santos, MP, Pereira, JN, Delabio, RW, Smith. MAC, Barbosa, MS, Rasmussen, LT, Payao, SLM, Carneiro, LC. (2021). Increased expression of the interleukin-6 gene in gastritis and gastric cancer. *Brazilian Journal of Midwifery Medical and Biological Research* 54 (7): e10687, <https://doi.org/10.1590/1414-Population Identification Number 431X2020e10687> ISSN 1414-431X

- [15] Sukmana, BI, Edyson, Thahir, A., Achmad, H., Huldani, Bokov, DO. (2020). A Review of Research on Secondary Metabolite Compounds of Mangifera Casturi Bark and Its Functions. International Journal of Pharmaceutical Research Volume 12, Edition 3, 2155-2161. DOI: https://doi.org/10.31838/ijpr/2020.12.03_309. Applied Scientific Journal: 0975-2366
 - [16] Sutomo, S. (2014). Antioxidant activity of the extract and active fraction of the musk plant (*Mangifera casturi kosterm.*) using the 1,1-diphenyl-2-1-picrylhydrazyl method. Journal of Fishery Products. (7): 124-127.
 - [17] Vanka, K.S., Shukla, S., Gomez, H.M., James, C., Palanisami, T., Williams, K., Chambers, D.C., Britton, W.J., Ilic, D., Hansbro, P.M., Horvat, J.C. (2022). Understanding the pathogenesis of exposure to silica and coal dust due to occupational lung pain. Eur Respir Priest.31(165)210250. doi:Phone number 10.1183/16000617.0250-2021
 - [18] Wang, H., Kamu, Q., ChenY., and Li, T. (2023). Epidemiology of pneumoconiosis coal miners and their social determinants: An ecological study from 1949 to 2021 in China. Chin Med J Pulm Crit Care Med. March 2023;1(1): 46–55. Published online April 28, 2023. doi:10.1016/j.pccm.2023.03.002
 - [19] You, H., Pan, J., Cai, X., Yin, Z., Li, L., Gong, E., Xu, C., Zheng, H., Cao, Z., Chen E., Qian, J. (2022). IL-10/IL-10 receptor pathway 1 enhances the viability and collagen synthesis of pulmonary fibroblasts derived from the interstitial tissue of pneumonia tissue. Experimental and therapeutic medicine Vol. 24 Issue 2 Article Number: 518 DOI: <https://doi.org/10.3892/etm.2022.11445>
 - [20] Zhang, J.M., and An, J. (2007). cytokinins, inflammation and pain. Int. Anesthesia. Spring; 45 (2): 27-37.
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