Expression of Catalase and Malondialdehyde Levels in Silicon Dioxyde-exposed

By Kusmiyati Kusmiyati

WORD COUNT 5148
TIME SUBMITTED 29-JUL-2020 08:11AM
PAPER ID 61650325

http://heanoti.com/index.php/hn

RESEARCH ARTICLE

URL of this article: http://heanoti.com/index.php/hn/article/view/hn20305

Expression of Catalase and Malondialdehyde Levels in Silicon Dioxyde-exposed Lung Tissue of Mice Treated with *Moringa oleifera* Leaves Extract

Kusmiyati[1(CA)], Soedjajadi Keman[2], Muhammad Amin[3], Suwarno[4]

[1(CA)]Doctorate Program of Health Science, Faculty of Public Health, Airlangga University, Indonesia; kus1979@yahoo.com (Corresponding Author)

[2]Department of Environmental Health, Faculty of Public Health, Airlangga University, Indonesia

[3]Department of Pulmonary and Respiratory Medicine, Faculty of Medicine, Airlangga University, Indonesia

[4]Department of Veterinary Microbiology, Faculty of Veterinary Medicine, Airlangga University, Indonesia

ABSTRACT

Silica particle such as silicon dioxide (SiO_2), is considered as a hazardous and cytotoxic particle. Silica particle exposure leads to oxidative stress in lung tissue. *Moringa oleifera* is a plant with potential antioxidant compounds. Therefore the aim of this study was to analyze the effect of *M. oleifera* leaves extract (MLE) on expression of catalase enzyme and malondialdehyde (MDA) levels in lung tissue of mice exposed to silica particles. This study was an experimental study with randomized posttest-only control group design using 30 male Balb/c strain mice, 8-10 weeks of age, 20-30 g body weight (BW), which were randomly divided into five groups. Group 1 was the negative control group, group 2 was exposed to SiO_2 particle and set as the positive control group, group 3 was treated with MLE 2 mg/20 g BW, group 4 was treated with MLE 5 mg/20 g BW, and group 5 was treated with MLE 8 mg/20 g BW. After 90 days, mice were sacrificed by cervical dislocation and the lung tissues were examined. Study results showed that expression of catalase in lung tissues of MLE-treated group was higher than that of positive control group, but not statistically significant. There was a significant difference of MDA level in lung tissue among groups. MDA level of groups treated with MLE 2 mg/20 g BW and 5 mg/20 g BW was lower than that of control group, while group treated with MLE 8 mg/20 g BW showed higher MDA level than control group (One Way ANOVA, $p<0.05$). It is concluded that administration of MLE indicates to prevent SiO_2 induced-oxidative stress in lung tissue of Balb/c mice.

Keywords: Antioxidant, Catalase, Malondialdehyde, *Moringa oleifera*, Silicon dioxyde

INTRODUCTION

Background

All Environmental pollution highly influences public health. One of the factors which may lead to health problems is air pollution due to dust particles. Many activities could contribute to air pollution, such as transportation, road construction, drilling, *etc*. One of the hazardous dust components is silica particle. Silica dust could be found in material of road construction, foundry, cement, coal, glass, and ceramic industry, as well as gold, sand, iron, and any minerals mining[(1)].

Silica particle such as silicon dioxide (SiO_2), is considered as a hazardous and cytotoxic particle. Human could be exposed to silica through various ways, one of them is inhalation. The lungs are the most impacted organ due to exposure of inhaled silica particle. Studies in some areas exposed to silica dust has been conducted. The foundry workers in Samsun, Turkey, developed symptoms such as phlegm (20.46%), cough (14.98%), breathlessness (8.06%), and wheezing (2.01%). A study of foundry workers in Taiwan has reported that lung function is correlated to the level of silica dust[(2)]. Prolonged silica exposure could lead to silicosis. Silicosis has been reported in dental supply factories workers, located in the States of Michigan and New York, i.e five cases[(3)], and over 69.1% among agate workers at Shakarpur[(4)]. A retrospective study conducted in a clinic located in Edinburgh, Scotland, has reported six cases of silicosis in stonemasons[(5)].

Prolonged or repeated exposure to silica may cause lung tissue damage. Silica dust that passes into the lung causes various responses in lung tissue, such as inflammation response and oxidative stress. Oxidative stress may

result from ROS generated at the silica particle surface, phagocytosis process, as well as inflammation process. Oxidative stress, arising as a result of an *imbalance* between antioxidant in the body and ROS production. Therefore, materials that could increase antioxidant status in the body are required.

One of natural plant materials that frequently consumed by people is *Moringa oleifera* leaves (*Moringa oleifera*, Indonesia common name is *kelor*). It is frequently consumed as vegetable, or as additive in food and beverages. Many studies has reported that *M. oleifera* leaves contain a variety of compounds with its health benefits. *M. oleifera* leaves contain various active compounds as potential antioxidant[8]. A study in Mexico performed by Valdez-Solana *et al*. (2015) has reported that *M. oleifera* leaves contain phenolic acids (gallic and chlorogenic acids) and flavonoid (rutin, luteolin, quercetin, apigenin and kaempferol)[9]. *M. oleifera* leaves also contain micro minerals such as Cu, Zn and Mn, as well as some vitamins[10].

Both *in vitro* and *in vivo* studies have demonstrated that *M. oleifera* leaves possess antioxidant[11], inflammatory and immunomodulatory[12], hypolipidemic, hepatoprotective[12], hypoglycemic[13], antibacterial, wound healing[14] and anticancer characteristic[15]. Nevertheless, the role of *M. oleifera* leaves to treat the impact of silica exposure has never been studied.

Purpose

This study aimed to analyze the effect of *M. oleifera* leaves extract on expression of catalase and MDA levels in lung tissue of mice exposed to silicon dioxyde (SiO_2) particles.

METHODS

Plant Material

Moringa oleifera plant was obtained from *Moringa oleifera* plantation in Sokon Village, Kupang, East Nusa Tenggara, Indonesia. The plant was identified in laboratory of Department of Biology, Faculty of Science and Technology, Airlangga University.

Laboratory Animal

Healthy male Balb/c strain mice *(Mus musculus)*, 8-10 weeks of age, weighing 20-30 grams, were obtained from biochemistry laboratory in Faculty of Medicine, Airlangga University. Mice were raised in Laboratory Animal Care Unit of biochemistry laboratory, Faculty of Medicine, Airlangga University. Ethics approval certification was obtained from Ethics Committee of Health Study, Faculty of Public Health, Airlangga University, No. 544-KEPK. Mice were fed pellets and water *ad libitum*. They were acclimatized for 1 week. A total of 30 mice were randomly divided into five groups of six each. The groups consisted of negative control group (without silica exposure and administration of MLE), positive control group (with silica exposure and administration of MLE), treatment group with silica exposure and treated with MLE 2 mg/20 g body weight (BW), treatment group with silica exposure and treated with MLE 5 mg/20 g BW, and treatment group with silica exposure and treated with MLE 8 mg/20 g BW. The extract was administered via oral gavage over 10 days before and 90 days after silica exposure. In the end of the treatment period, mice were sacrificed by cervical dislocation.

Extraction of *Moringa oleifera* leaves

M. oleifera leaves were extracted using maceration technique with ethanol 96% as a solvent. Extraction was performed in Phytochemistry Laboratory, Faculty of Pharmacy, Airlangga University. Identification of compounds contained in extract was done in Testing Service Unit, Faculty of Pharmacy, Airlangga University. Ethanol extract of *M. oleifera* leaves was diluted in CMC-Na 0.5% to obtain the desired concentration for administration via oral gavage to the mice.

Silicon dioxyde (SiO_2) Particle Exposure

In this study, 1-5µm silica particles SiO_2 (Sigma Aldrich) 80% were used. Silica was administered intratracheally, with SiO_2 concentration of 2.5 mg in 60µl NaCl 0.9%.

Measurement of catalase expression

Expression of catalase in lung tissue was measured using immunohistochemical methods with Anti-Catalase antibody [EPR1928Y](ab76110). Expression of catalase was observed under light microscope at a

magnification of 400x with 10 field of view regions. Measurement of catalase expression was performed in Electron Microscope and Integrated Medical Laboratory Unit, Faculty of Medicine, Airlangga University.

Measurement of MDA level

MDA level in lung tissue was measured using Enzyme-linked immunosorbent assay (ELISA) and the absorbance was read at 450 nm with a microplate reader. Mouse Malondialdehyde ELISA Kit (Cat. No E0625Mo) was used. Measurement of MDA level was performed in Immunology Laboratory, Faculty of Veterinary Medicine, Airlangga University.

Data and Statistical Analysis

The numerical data were expressed as mean and standard deviation[16]. One-Sample Kolmogorov Test was used to test the normality of data. Data from each group were compared using One-Way ANOVA with 95% confidence interval and Post-Hoc analysis with LSD method.

RESULTS

Body weight

At the initial of the study, Balb/c mice *(Mus musculus)*weight was recorded to ascertain that mice used in the study were from homogenous population. Mean and standard deviation of Balb/c mice *(Mus musculus)* weight are displayed in Table 1. The study involved Balb/c mice *(Mus musculus),* weighing 20-30 g. Statistical analysis showed that no significant difference in mice weight between groups (One Way ANOVA, $p>0.05$). Therefore, indicating that samples consist of mice from homogenous weight population.

Expression of catalase

Measurement of catalase expression was performed using immunohistochemical method with Anti-Catalase antibody. Means of catalase expression in negative control group and positive control group were 2.77±1.50 and 2.15±1.49, respectively. Means of catalase expression in lung tissue of mice in both treatment groups MLE 2 mg/20 g BW and MLE 5 mg/20 g BW were higher than positive control group (Table 1). Expressions of catalase in lung tissue of Balb/c mice *(Mus musculus)* treat with MLE tended to increase although statistical analysis showed that there was no significant difference between treatment groups (One Way ANOVA, $p>0.05$). Expressions of catalase on immunohistochemical examination are shown in Figure 1.

Malondialdehyde level

Measurement of MDA level is an indirectly assessment of free radical activity, thus what can be measured are products of the damage produced by free radicals, not directly the free radical compounds. In the study, MDA level in lung tissue was measured as the end-product of lipid peroxidation. The result showed the significant difference of MDA levels between groups ($p<0.05$). The result of Post-Hoc analysis with LSD method is presented in Table 1. It shows a significant difference of MDA levels between negative and positive control group ($p<0.05$), negative control group and MLE 8 mg/20 g BW group ($p<0.05$), positive control group and MLE 2 mg/20 g BW ($p<0.05$), positive control group and MLE 5 mg/20 g BW ($p<0.05$), positive control group and MLE 8 mg/20 g BW ($p<0.05$). MDA level of positive control group was higher than that of negative control group. The lowest MDA level was found in MLE 5 mg/20 g BW group, 0.3024±0.0328 nmol/ml, indicating that the dose level is able to decrease MDA. The results of linear regression showed that MDA contents was influenced by catalase expression ($p<0,05$).

Table 1. Body Weight, Expression of Catalase and MDA Levels in Lung Tissue of Balb/c mice *(Mus musculus)*

Group	Body Weight	Catalase	MDA
Negative control	25.17±1.72	2.77±1.50	0.3101 [a] ±0.0138
Positive control	25.33±1.21	2.15±1.49	0.3412 [b] ±0.0192
MLE 2 mg/20 g BW	25.67±1.03	3.08±1.96	0.3088 [a] ±0.0183
MLE 5 mg/20 g BW	25.33±0.82	4.77±2.19	0.3024 [a] ±0.0328
MLE 8 mg/20 g BW	25.00±1.41	2.40±0.79	0.3748 [c] ±0.0150

Data are presented as the means±SD, n= 6. Body weights are expressed in gram, catalase expressed in the number cells expressing catalase antibody and MDA levels are expressed in nmol/ml tissue. [abc] The same superscript indicates no significant difference between groups according One way ANOVA test followed by Pos Hoc LSD test.

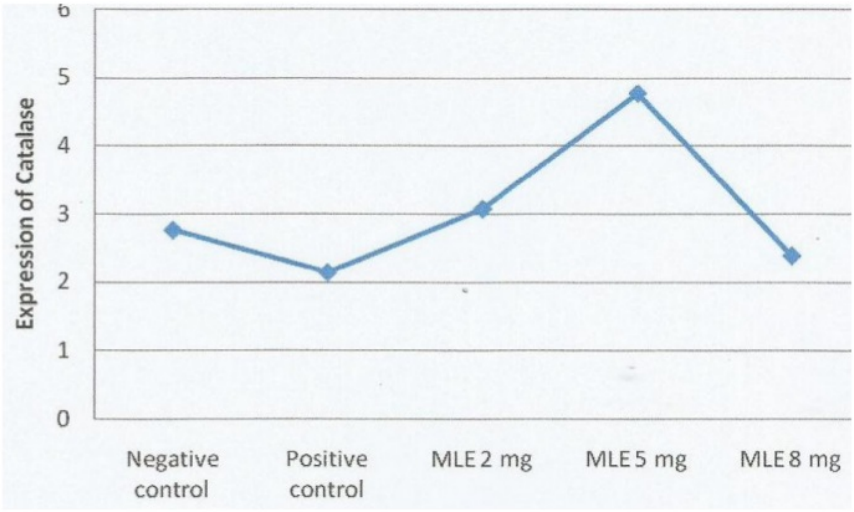

Figure 1. Expression of Catalase in Lung Tissue of Balb/c mice *(Mus musculus)*

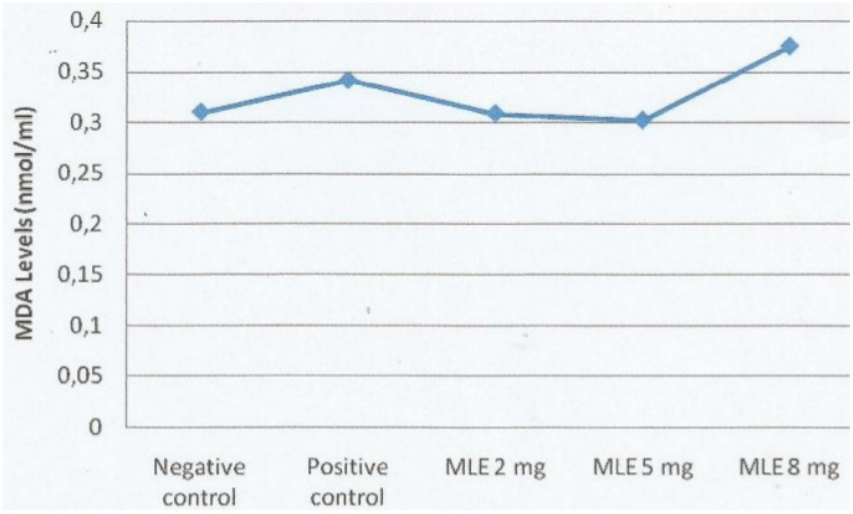

Figure 2. MDA Levels in Lung Tissue of Balb/c mice *(Mus musculus)*

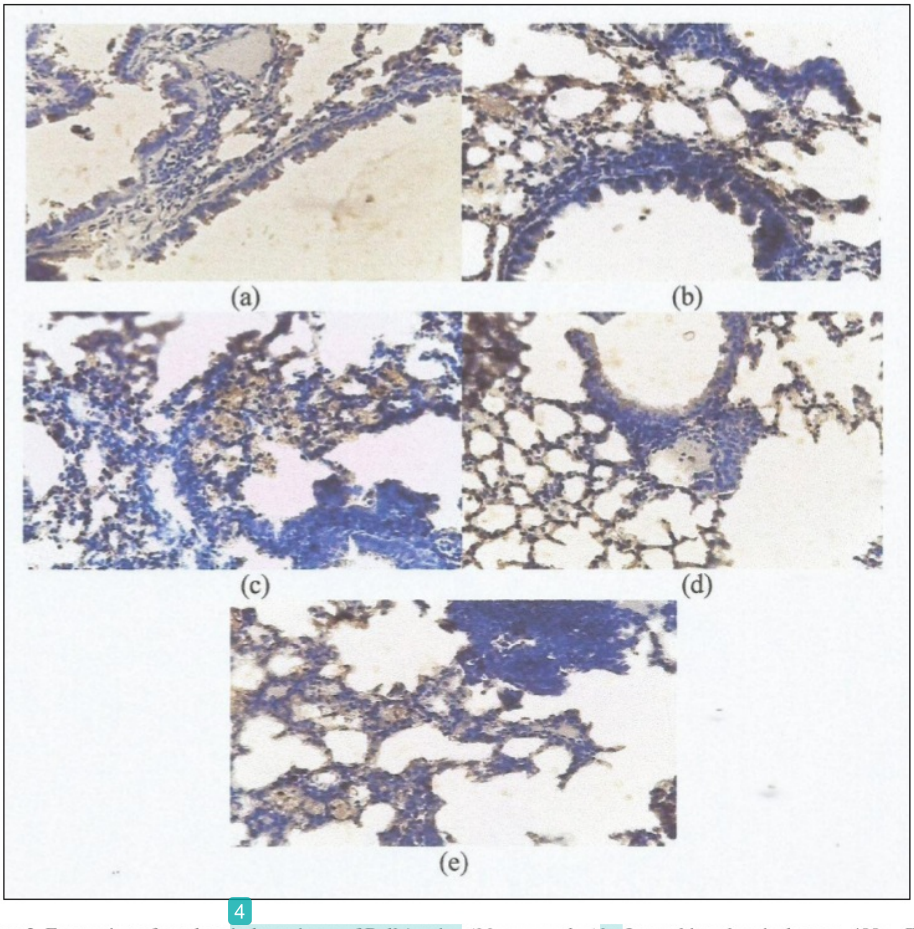

Figure 3. Expression of catalase in lung tissue of Balb/c mice *(Mus musculus)* by Imunohistochemical assay, 400x. Cells expressing catalase show a brown color. (a) Catalase expression in lung tissue of normal group. (b) Silica-treated group showing catalase expression decreasing. (c) MLE 2 mg/20 BW-treated group showing catalase expression increasing. (d) MLE 5 mg/20 BW-treated group showing catalase expression increasing. (e) MLE 8 mg/20 BW-treated group showing catalase expression decreasing.

DISCUSSION

Expression of Catalase

Catalase is endogenous antioxidant enzyme which catalyzes H_2O_2 into H_2O and O_2. The study result showed that silica-exposed groups had decreased catalase expression (Table 1). This is apparently due to silica could impair antioxidant system in the body[17]. Accordingly, it may impair the expression of antioxidant enzymes. Body has defense system to protect the body against ROS-induced oxidative stress via antioxidant system that consists of three line defense antioxidants. First, preventive antioxidants, i.e endogenous enzymes and minerals such as SOD, catalase, GPx, glutathione reductase, Se, Mn, Cu. Second, radical scavenger, such as glutathione, vitamin C, albumin, vitamin E, carotenoid, flavonoid. Beta-carotene scavenges singlet oxygen, vitamin C interacts with radicals such as O_2, OH. GSH acts as scavenger of free radicals such as O_2, OH, lipid hydroperoxide. Third, a group of enzymes which repairs damaged DNA, protein and lipids, and also biomolecular and cell membrane damage[18].

Enzyme activity in the body is highly influenced by enzyme cofactors, one of them is micronutrient *i.e*. selenium (Se). Se plays role in neutralizing process of ROS. Se acts as enzyme cofactor of GPx. A study involved patients with silicosis revealed reduced serum Se concentration[19]. Other study involved patients with silicosis also found different selenium level compared with control participants. Patients with silicosis were found to have a lower serum selenium concentration compared to healthy control[17].

The results showed that administration of *M. oleifera* leaves extract increased expression of catalase (Figure 1). This is due to *M. oleifera* leaves extract contains various compounds that could stimulate endogenous antioxidants. The results of identification of compounds in ethanol extract of *M. oleifera* leaves using thin layer chromatography revealed the presence of polyphenol, flavonoid, saponin, saturated sapogenin and unsaturated steroid compounds.

Table 1 showed that group treated with ethanol extract of MLE at dose 5 mg/20 g BW had the highest increase of catalase expression compared to those at dose 2 mg/20 g BW and 8 mg/20 g BW. *M. oleifera* leaves contain various bioactive compounds and nutrients which possess a lot of health benefits, e.g its function as antioxidant[20]. *M. oleifera* leaves are also the source of mineral and protein-rich food, and vitamin A, B, C and E[21]. A study of supplementation *M. oleifera* leaves powder could significantly improve serum GPx and SOD in postmenopausal women, and also decrease marker of oxidative stress i.e. MDA[22]. *M. oleifera* leaves extract also could restore antioxidant status in mice fed with high-fat diet[23].

The findings of identification compounds in *M. oleifera* leaves extract revealed the presence of flavonoid. Flavonoid found in ethanol extract of Moringa leaves possess antioxidant activity[24]. In addition, the presence of saponin was also shown in *M. oleifera* leaves extract. Saponin is bioactive compound with potential antioxidant capacity. A study conducted by Muhammad *et al*. (2017) showed that triterpenoidsaponins from the stems and bark of *Jaffrea xerocarpa*exhibited antioxidant activity in DPPH assay[25]. Other study also showed that administration of flavonoid, quercetin, could improve catalase activity in the bleomycin-treated lung[26].

Malondialdehyde Levels

Measurement of MDA level is an indirectly assessment of free radical activity, thus what can be measured are products of the damage produced by free radicals, not directly the free radical compounds. In the study, MDA level in lung tissue was measured as the end-product of lipid peroxidation. The result showed the significant difference of MDA levels between groups ($p<0.05$). The result of Post-Hoc analysis with LSD method is presented in Table 1. It shows a significant difference of MDA levels between negative and positive control group ($p<0.05$), negative control group and MLE 8 mg/20 g BW group ($p<0.05$), positive control group and MLE 2 mg/20 g BW ($p<0.05$), positive control group and MLE 5 mg/20 g BW ($p<0.05$), positive control group and MLE 8 mg/20 g BW ($p<0.05$). MDA level of positive control group was higher than that of negative control group. The lowest MDA level was found in MLE 5 mg/20 g BW group, 0.3024±0.0328 nmol/ml, indicating that the dose level is able to decrease MDA.

Exposure of silica particle could cause any changes in the body particularly the lungs. It results in some effects such as ROS formation and antioxidant imbalance in the body. Generation of oxidants by silica particles and by silica-activated cells results in the increased lipid peroxidation, cell and lung injury[19]. The present study did not perform directly measurement of ROS. Instead, MDA level as a product of lipid peroxidation by ROS was measured. The results showed that silica-exposed mice group had higher MDA level than control group. It indicated that exposure of silica particle leads to lipid peroxidation. It is similar to the study in humans. A study conducted in foundry plants which frequently exposed to metal dust and silica dust showed that among foundry workers, plasma MDA levels of exposure group were higher than that of in control group[27]. Administration of intratracheal silica to female Balb/c mice also elevated serum H_2O_2 level[28]. This indicated that silica could increase ROS in both animals and humans.

The study results showed that administration of MLE 2 mg/20 g BW and 5 mg/20 g BW could significantly decrease MDA level in lung tissue of mice. It proves that *M. oleifera* leaves possess antioxidant activity. These results were supported by *in vitro* study of *M. Oleifera*leaves extract through DPPH assay which showed its high antioxidant activity expressed as IC_{50}: 49.30µg/mL[29]. Other study involved Wistar rats treated with swimming test and aqueous extract of *M. oleifera* leaves showed increased activity of antioxidant enzymes and decreased blood concentration of MDA[30]. There was a negative correlation between serum MDA and total antioxidant status and GPx in red blood cell of Wistar rats treated with Moringa leaves. Serum MDA was observed to be significantly lower in rats fed with Moringa leaves than that of in control group. Total antioxidant status was also significantly higher in treatment group[31]. Various compounds in *M.oleifera* leaves extract are potential antioxidants in preventing oxidative stress or silica-induced lipid peroxidation. Vitamin content in *M. oleifera* leaves possibly plays role in reducing ROS. Saxena and Singh (2012) showed in their study that administration of vitamin C and vitamin E may recover histology of testis and epididymis of albino rats exposed to silica intraperitoneally[32].

Many natural antioxidants possess ability to inhibit oxidative stress through various mechanisms. Phenolic compounds in plants could be the source of natural antioxidants[33]. A study of ant-powder (*Myrmecodiapendans*) administration which contains antioxidants such as polyphenol, flavonoid, and tannin could increase serum SOD level and decrease serum MDA level of rats exposed to Pb-acetate[34]. Identification result of ethanol extract of *M. oleifera* leaves also found the presence of saponin. This compound also works as potential antioxidant. A study

of administration of saponin extracted from the root of *Garcinia kola* (Bitter kola) in diabetic animals model showed that saponin could decrease MDA level and increase SOD and catalase activity[35].

A study of saponin fraction extracted from stem bark of *Erythropheleum suaveolens* showed that it may inhibit lipid peroxidation and prevent free radical-induced damage[36]. Singh *et al*. (2017) reported that saponins in pulses were found to be beneficial in promoting healing which worked as antioxidant[37]. Other study showed that aqueous extract of *M. oleifera* leaves could inhibit lipid peroxidation in testis tissue by forming a complex with Fe^{2+}, thus preventing the initiation of lipid peroxidation[38]. Thus, saponins present in *M. oleifera* leaves may possibly decrease MDA level.

Flavonoid possess antioxidant activity through some mechanisms, such as scavenging free radicals, chelating metal, suppressing enzyme related to free radical formation, and stimulating internal antioxidant enzymes[39]. However, high dose of flavonoid molecules could possibly act as pro-oxidant[40]. The study results showed that administration of MLE at dose 8 mg/20 g BW to mice increased the MDA levels (Figure 2). This finding is in accordance with an *in vitro* study that showed administration of *M. oleifera* leaves extracted in ethanol 80% decreased oxidative stress, but administration of the extract at high dose (2000-3000 mg/l) could induce cytotoxicity[41]. Increase of MDA level could be caused by many factors. Environmental factor and prolonged treatment may influence the increase of MDA level. In addition, compounds in *M. oleifera* could possibly act as pro-oxidant.

In certain condition, flavonoid could act as pro-oxidant, which promotes oxidation in other components[42]. Pro-oxidant and antioxidant effect of flavonoid depends on its concentration[42],[43]. This is in accordance with a study by Shobot and Hadacek (2011) that found flavonoid antioxidant (myricetin) may also act as pro-oxidant. Pro-oxidant could occur due to oxidation process by phenolic flavonoid radicals, inhibition of mitochondrial respiration, absorption inhibition of low-molecular-weight antioxidants[43]. Pro-oxidant effect could also be beneficial[42]. The use of appropriate dose is crucial to obtain optimal benefit.

CONCLUSION

Ethanol extract of *M. oleifera* leaves 2 mg/20 g BW and 5 mg/20 g BW could inhibit oxidative stress in lung tissue of mice (*Mus musculus*) exposed to silicon dioxyde particle, which demonstrated by increased expression of catalase, and decreased MDA levels in lung tissue. These indicate that ethanol extract of *M. oleifera* leaves contains various compounds that work as potential antioxidant, such as phenolic, flavonoid, and saponin compounds. MDA levels affected by catalase expression.

Further study related to the effect of *M. oleifera* leaves extract on other antioxidant enzymes such as, SOD and GPx is needed to analyze the mechanism of ethanol extract of *M. oleifera* leaves in preventing oxidative stress-induced lung damage.

REFERENCES

1. IAPA. Silica In The Workplace. Industrial Accident Prevention Association; 2008.
2. Lin MH, Liou SH, Chang, CW, Huang IH, Strickland PT, Lai CH. An Engineering Intervention Resulting in Improvement in Lung Function and Change in Urinary 8-hydroxydeoxyguanosine Among Foundry Worker in Taiwan. Int Arc Occup Environ Health. 2011;84:175-183.
3. Hoz RED, Rosenman K, Borczuk A. Silicosis in Dental Supply Factory Workers. Respiratory Medicine. 2004;98:791-794.
4. Chaudhury N, Phatak A, Paliwal R, Raichaudhari. Silicosis Among Agate Workers at Shakarpur: An Analysis of Clinic-based Data. Lung India. 2010;27(4):221-224.
5. Nicol LM, McFarlane PA, Hirani N, Reid PT. Six Cases of Silicosis: Implications for Health Surveillance of Stonemasons. Occupational Medicine. 2015;65:220-225.
6. Kasolo JN, Bimenya GS, Ojok L, Ochieng J, Ogwal-Okeng JW. 2010. Phytochemicals and Uses of *Moringa oleifera* Leaves in Ugandan Rural Communities. Journal of Medicinal Plents Research. 2015;4(9):753-757.
7. Kumar P, Arora S, Yadav YC. Anti-inflammatory Activity of Coumarin and Steroidal Fractions From Leaves of *Moringa oleifera*. International Journal of Drug Discovery and Medical Research. 2012;1(1):20-25.
8. Jaiswal D, Rai PK, Mehta S, Chatterji S, Shukla S, Rai DK, Sharma G, Sharma B, Khair S, Watal G. Role of *Moringa oleifera* in Regulation of Diabetes-Induced Oxidative Stress. Asian Journal of Tropical Medicine. 2013;6(6):426-432.
9. Valdez-Solana MA, Mejia-Garcia VY, Tellez-Valencia A, Garcia-Arenas G, Salas-Pacheco J, Alba-Romero JJ and Sierra-Campos E. Nutritional Content and Element and Phytochemical Analyses of *Moringa oleifera* Grown in Mexico. Journal of Chemistry. 2015. 860381, 9 pages.
10. Moyo B, Masika PJ, Hugo A, Muchenje V. Nutritional Characterization of Moringa (*Moringa oleifera Lam*.) Leaves. African Journal of Biotechnology. 2011;10(60):12925-12933.

11. Yassa HD, Tohamy AF. Extract of Moringa oleifera Leaves Ameliorates Streptozotocin-induced Diabetes Mellitus in Adult rats. Acta Histochemica. 2014;116:844–854.
12. Leone A, Spada A, Battezzati A, Schiraldi A, Aristil J, Bertoli S. Cultivation, Genetic, Ethnopharmacology, Phytochemistry and Pharmacology of *Moringa oleifera* Leaves: An Overview. International Journal of Molecular Sciences. 2015;16:12791-12835.
13. Olurishe C, Kwanashie H, Zezi A, Danjuma N, Mohammed B. Chronic Administration of Ethanol Leaf Extract of *Moringa oleifera Lam*. (*Moringaceae*) may Compromise Glycaemic Efficacy of Sitagliptin with no Significant Effect in Retinopathy in a Diabetic Rat Model. Journal of Ethnopharmacology. 2016;194:895-903.
14. Nayak D, Ashe S, Rauta PR, Nayak B. Assessment of Antioxidant, Antimicrobial and Anti-osteosarcoma Potential of Four Traditionally Used Indian Medicinal Plants. Journal of Applied Biomedicine. 2017;15:119-132.
15. Dany M, Madi N, Nemer N, Beyrouthy M, Abdoun S, Usta J. *Moringa oleifera*: Natural Leaf Extract with Potential Anti-cancerous Effect on A549 Lung Cancer Cells. Lung Cancer. 2012;77:S21–S27
16. Nugroho HSW. Descriptive Data Analysis for Numerical Data (Analisis Data Secara Deskriptif untuk Data Numerik. Ponorogo: Forum Ilmiah Kesehatan (Forikes); 2014.
17. Muzembo BA, Dumavibhat N, Ngatu NR, Eitoku M, Hirota R, Kondo S, Deguchi Y, Saito Y, Takahashi K, Suganuma N. Serum Selenium and Selenoprotein P in Patients with Silicosis. Journal of Trace Elements in Medicine and Biology. 2013;27:40-44.
18. Portap S, Pandey S. A Review On Herbal Antioxidants. Journal of Pharmacognosy and Phytochemistry. 2012;1(4):26-37.
19. Pandey JK, Agarwal D. Biomarker: A Potensial Prognostic Tool for Silicosis. Indian Journal and Environmental Medicine. 2012;16(3):101-107.
20. Saini RK, Sivanesan I, Keum Y. Phytochemicals of *Moringa oleifera*: A Review of Their Nutritional, Therapeutic and Industrial Significance. Biotech. 2016;6:1-14.
21. Jideani VA, Diedericks CF. Nutritional, Therapeutic, and Prophylactic Properties of Vigna subterranea and *Moringa oleifera*. 2014;187-207.
22. Kushwaha S, Chawla P, Kochhar A. Effect of Supplementation of Drumstick (*Moringa oleifera*) and Amaranth (*Amaranthus tricolor*) Leaves Powder on Antioxidant Profile and Oxidative Status Among Postmenopausal Women. J Food Sci Technol. 2014;51(11):3464–3469.
23. Das N, Sikder K, Ghosh S, Fromenty B, Dey S. *Moringa oleifera* Leaf Abstract Prevent Early Liver Injury and Restores Antioxidant Status in Mice Fed with Hight-fat Diet. Indian Journal of Experimental Biology. 2012;50:404-412.
24. Wang Y, Gao Y, Ding H, Liu S, Han X, Gui J, Liu D. Subcritical Ethanol Extraction of Flavonoids from *Moringa oleifera* Leaf and Evaluation of Antioxidant Activity. Food Chemistry. 2017;218:152-158.
25. Muhammad D, Lalun N, Bobichon H, Debar ELM, Gangloff SC, Nour M, Voutquenne-Nazabadioko L. Triterpenoid Saponins and Other Glycosides from the Stems and Bark of Jaffrea xerocarpa and their Biological Activity. Phytochemistry. 2017;141:121-130.
26. Verma VK, Singh N, Saxena P, Singh R. Anti-ulcer Antioxidant Activity of *Moringa oleifera (Lam)* Leaves Against Aspirin and Ethanol Induced Gastric Ulcer in Rats. International Research Journal of Pharmaceutical. 2012;02(02):46-57.
27. Liu H, Lin M, Liu P, Chan C, Chen C. Health Risk Assessment by Measuring Plasma Malondialdehyde (MDA), Urinary 8-hydroxydeoxyguanosine (8-OH-dG) and DNA Strand Breakage Following Metal Exposure in Foundry Workers. Journal of Hazardous Materials. 2009;170:699.704.
28. Sato T, Shimosato T, Alvord WG, Klinman DM. Suppressive Oligodeoxynucleotides Inhibit Silica-Induced Pulmonary Inflammation. The Journal of Immunology. 2015;7648-7654.
29. Fitriana WD, Ersam T, Shimizu K and Fatmawati S. Antioxidant Activity of Moringa oleifera Extract. Indones. J. Chem. 2016;16(3):297-301.
30. Lamou B, Taiwe GS, Hamadou A, Abene, Houlray J, Atour MM, Tan PV. Antioxidant and Antifatigue Properties of the Aqueous Extract of Moringa oleifera in Rats Subjected to Forced Swimming Endurance Test. Oxid Med Cell Longev. 2016;1-9.
31. Oparinde DP, Atiba AS. Moringa oleifera Leaf Prevents Oxidative Stress in Wistar Rat. European Journal of Medicinal Plants. 2014;4(9):1150-1157.
32. Saxena S, Singh SP. Efficacy of vitamin E and Vitamin C Against Silica Induced Toxicity on Male Reproductive Organs of Albino Rats. Journal of Applied and Natural Science. 2012;4(1):127-131.
33. Brewer MS. Natural Antioxidants: Sources, Compounds, Mechanisms of Action, and Potential Applications. Comprehensive Reviews in Food Scince and Safety. 2011;10:221-247.
34. Lamondo D, Soegianto A, Abadi A, Keman K. Antioxidant Effects of Sarang Semut (*Myrmecodia pendans*) on the Apoptosis of Spermatogenic Cells of Rats Exposed to Plumbum. Research Journal of Pharmaceutical. Biological and Chemical Sciences. 2014;5(4):282-294.

35. Smith A, Adanlawo IG. In Vitro and In Vivo Antioxidant Activity of Saponin Extracted from The Root of *Garcinia kola* (Bitter Kola) on Alloxan-induced Diabetic Rats. World Journal of Pharmacy and Pharmaceutical Sciences. 2014;3(7):08-26.
36. Akinpelu BA, Igbeneghu OA, Awotunde AI, Iwalewa EO, Oyedapo. Antioxidant and Antibacterial Activities of Saponin Fractions of Erythropheleum Suaveolens (Guill. And Perri.) Stem Bark Extract. Academic Journals. 2014;9(18):826-833.
37. Singh B, Singh JP, Singh N, Kaur A. Saponins in Pulses and Their Health Promoting Activities: A review. Food Chemistry. 2017;233:540-549.
38. Akomolafe SF, Oboh G, Hunsi AAA, Akinyemi AJ, Adeyanju O. Inhibitory Effect of Aqueous Extract of Moringa oleifera and Newbuoldia laevis Leaves on Ferrous Sulphate and Sodium Nitroprusside Induced Oxidative Stress in Rat's Testes in Vitro. Open Journal of Medicinal Chemistry. 2012;2:119-128.
39. Kumar S, Pandey AK. Chemistry and Biological Activities of Flavonoids: An Overview. The Scientific World Journal. 2013;2013:1-16.
40. Rathee P, Chaundhary H, Rathee S, Rathee D, Kumar V, Kohli K. Machanism of Action Flavonoids as Anti-inflammatory Agents: A Review. Inflammation & Allergy-Drug Targets. 2009;8:229-235.
41. Sangkitikomol W, Rocejanasaroj A, Tencomnao T. Effect of Moringa oleifera on Advanced Glycation End-Product Fomation and Lipid Metabolism Gene Expression in HepG2 Cells. Genetics and Molecular Research. 2014;13(1):723-735.
42. Baldim JL, Alcantara GV, Domingos OS, Soares MG, Caldas IS, Novaes RD, Oliveira TB, Lago JHD, Chagas-Paula DA. The Correlation Between Chemical Structures and Antioxidant, Prooxidant and Antitypanosomatiol Properties of Flavonoids. Oxidative Medicine and Cellular Longevity.2017;1-12.
43. Chobot V, Hadacek. Extraploration of Pro-oxidant and Antioxidant Activities of the Flavonoid Myricetin. Redox Report. 2011;16(6):242-247.

Expression of Catalase and Malondialdehyde Levels in Silicon Dioxyde-exposed

ORIGINALITY REPORT

17%

SIMILARITY INDEX

PRIMARY SOURCES

1	Zhen Zhang. "Fatty acid extracts from Lucilia sericata larvae promote murine cutaneous wound healing by angiogenic activity", Lipids in Health and Disease, 2010 Crossref	52 words —	1%
2	worldwidescience.org Internet	47 words —	1%
3	"The nutraceutical effect of Scenedesmus dimorphus for obesity and nonalcoholic fatty liver disease–linked metabolic syndrome", Journal of Applied Pharmaceutical Science, 2020 Crossref	34 words —	1%
4	Wenhong Kan. "Lung, Spleen, and Kidney Are the Major Places for Inducible Nitric Oxide Synthase Expression in Endotoxic Shock: Role of P38 Mitogen-Activated Protein Kinase in Signal Transduction of Inducible Nitric Oxide Synthase Expression", Shock, 03/2004 Crossref	29 words —	1%
5	Mohammed Al-Abri, Mohammed Ashique, Aishwarya Ramkumar, Abderrahim Nemmar, Badreldin H. Ali. " Motor and Behavioral Effects of Leaf Extract ", Natural Product Communications, 2018 Crossref	28 words —	1%
6	journals.sagepub.com Internet	26 words —	1%
7	Sidney J. Stohs, Michael J. Hartman. " Review of the Safety and Efficacy of ", Phytotherapy Research, 2015	22 words —	1%

Crossref

8	Andrew B. Falowo, Felicitas E. Mukumbo, Emrobowansan M. Idamokoro, José M. Lorenzo, Anthony J. Afolayan, Voster Muchenje. "Multi-functional application of Moringa oleifera Lam. in nutrition and animal food products: A review", Food Research International, 2018 Crossref	20 words — 1%
9	www.scribd.com Internet	20 words — 1%
10	"E-Poster Presentations (Oral) | APDW 2019", Journal of Gastroenterology and Hepatology, 2019 Crossref	18 words — < 1%
11	file.scirp.org Internet	17 words — < 1%
12	"Abstracts of the Asian Congress of Nutrition 2019", Annals of Nutrition and Metabolism, 2019 Crossref	15 words — < 1%
13	onlinelibrary.wiley.com Internet	14 words — < 1%
14	journals.lww.com Internet	13 words — < 1%
15	www.wjpps.com Internet	12 words — < 1%
16	"Poster Abstracts", Neurogastroenterology and Motility, 7/2005 Crossref	12 words — < 1%
17	www.advancesagainstaspergillosis.org Internet	12 words — < 1%
18	"Nutritional Antioxidant Therapies: Treatments and Perspectives", Springer Science and Business Media LLC, 2017 Crossref	12 words — < 1%

19	de la Hoz, R.E.. "Silicosis in dental supply factory workers", Respiratory Medicine, 200408 Crossref	12 words —	< 1%
20	www.heanoti.com Internet	12 words —	< 1%
21	X He, M Yi, B Liu, L Xu, X Li, A Lu. "Anti-oxidant and Immunomodulatory Acitivities of Ganoderma lucidum and American Ginseng in Rats and Immunosuppressive Mice", Planta Medica, 2011 Crossref	11 words —	< 1%
22	www.bioone.org Internet	11 words —	< 1%
23	www.nutridom.ca Internet	10 words —	< 1%
24	bmccancer.biomedcentral.com Internet	10 words —	< 1%
25	Bonoy Lamou, Germain Sotoing Taiwe, André Hamadou, Abene, Justin Houlray, Mahamat Mey Atour, Paul Vernyuy Tan. " Antioxidant and Antifatigue Properties of the Aqueous Extract of in Rats Subjected to Forced Swimming Endurance Test ", Oxidative Medicine and Cellular Longevity, 2016 Crossref	9 words —	< 1%
26	Noumo Ngangmou Thierry, Tatsadjieu Ngouné Léopold, Montet Didier, F. Mbofung Carl Moses. "Effect of Pure Culture Fermentation on Biochemical Composition of <i>Moringa oleifera</i> Lam Leaves Powders", Food and Nutrition Sciences, 2013 Crossref	9 words —	< 1%
27	V. Kuete. "Moringa oleifera", Elsevier BV, 2017 Crossref	9 words —	< 1%
28	www.degruyter.com Internet		

9 words — < 1%

29 Sujogya Kumar. "Chapter 15 Assay Guided Comparison for Enzymatic and Non-Enzymatic Antioxidant Activities with Special Reference to Medicinal Plants", IntechOpen, 2012
Crossref
9 words — < 1%

30 www.jpsionline.com
Internet
9 words — < 1%

31 Souravh Bais, Guru Sewak Singh, Ramica Sharma. " Antiobesity and Hypolipidemic Activity of Leaves against High Fat Diet-Induced Obesity in Rats ", Advances in Biology, 2014
Crossref
9 words — < 1%

32 repo.unand.ac.id
Internet
9 words — < 1%

33 K I N Rahayu, I P S Suharto, A N Etika, S E Nurseskasatmata. "The Effect of Ginger Extract (zingiber officinale roscoe) on the Number of Neutrophil Cells, Fibroblast and Epithelialization on Incision Wound", Journal of Physics: Conference Series, 2020
Crossref
9 words — < 1%

34 millenniumzoology.com
Internet
8 words — < 1%

35 www.researchgate.net
Internet
8 words — < 1%

36 Basilua Andre Muzembo, Narongpon Dumavibhat, N'landu Roger Ngatu, Masamitsu Eitoku et al. "Serum selenium and selenoprotein P in patients with silicosis", Journal of Trace Elements in Medicine and Biology, 2013
Crossref
8 words — < 1%

37 Pedalino, C.. "Effect of Atropa belladonna and Echinacea angustifolia in homeopathic dilution on
8 words — < 1%

experimental peritonitis", Homeopathy, 200410
Crossref

38 Bani, S.. "Anti-arthritic activity of a biopolymeric fraction from Euphorbia tirucalli", Journal of Ethnopharmacology, 20070301 — 8 words — < 1%
Crossref

39 V.S. Neergheen-Bhujun, Z.B. Ruhomally, Y. Dunneram, R. Boojhawon, M. Chan Sun. "Consumption patterns, determinants and barriers of the underutilised Moringa oleifera Lam in Mauritius", South African Journal of Botany, 2020 — 8 words — < 1%
Crossref

40 Fatma Abd El-Fat, Heba Hassan Sal, Samah Mosbah El-, Hoda Samir El-S, Hamdy Abdel-Hady. "Utilization of Natural Antimicrobial and Antioxidant of Moringa oleifera Leaves Extract in Manufacture of Cream Cheese", Journal of Biological Sciences, 2018 — 7 words — < 1%
Crossref

41 Chatchada Sutalangka, Jintanaporn Wattanathorn, Supaporn Muchimapura, Wipawee Thukham-mee. " Mitigates Memory Impairment and Neurodegeneration in Animal Model of Age-Related Dementia ", Oxidative Medicine and Cellular Longevity, 2013 — 7 words — < 1%
Crossref

42 "Abstracts of the 35th Annual Meeting of the European Society of Human Reproduction and Embryology", Human Reproduction, 2019 — 7 words — < 1%
Crossref

43 Lei Wang, Qiong Zou, Jinxing Wang, Junjie Zhang, Zeping Liu, Xiaoyang Chen. "Proteomic Profiles Reveal the Function of Different Vegetative Tissues of Moringa oleifera", The Protein Journal, 2016 — 7 words — < 1%
Crossref

44 Sri Agus Sudjarwo. "Purification and characterization protein of anti-dengue specific immunoglobulin Y for diagnostic kit of dengue", Journal of Applied Pharmaceutical — 7 words — < 1%

Science, 2012
Crossref

45 Shruti Saxena, S. P. Singh. "Efficacy of vitamin E and vitamin C against silica induced toxicity on male reproductive organs of albino rats", Journal of Applied and Natural Science, 2012 — 6 words — < 1%
Crossref

46 Z.F. Ma, J. Ahmad, H. Zhang, I. Khan, S. Muhammad. "Evaluation of phytochemical and medicinal properties of Moringa (Moringa oleifera) as a potential functional food", South African Journal of Botany, 2019 — 6 words — < 1%
Crossref

47 R. K. Saini, N. P. Shetty, P. Giridhar. "GC-FID/MS Analysis of Fatty Acids in Indian Cultivars of Moringa oleifera: Potential Sources of PUFA", Journal of the American Oil Chemists' Society, 2014 — 6 words — < 1%
Crossref

EXCLUDE QUOTES ON
EXCLUDE BIBLIOGRAPHY ON
EXCLUDE MATCHES OFF