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### Factors Associated with Plasma Malondialdehyde Levels in People over 40 Years

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Research Article

## Factors Associated with Plasma Malondialdehyde Levels in People over 40 Years

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### Abstract



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Aging is a progressive process caused by a physiological decline in body functions. It is known that several factors can accelerate aging, namely age, lifestyle, oxidative stress, lipid peroxidation, and mitochondrial damage. Various pollutants such as cigarette smoke, motor vehicle fumes, industrial waste, and exposure to sunlight / ultraviolet light will form free radicals. Malondialdehyde (MDA) level is one of the indicators used to measure oxidative stress. Increased MDA levels are found in various degenerative diseases. This study aims to determine plasma MDA levels in the age group of 40 years and over and factors that can affect plasma MDA levels. This study was a cross-sectional study by including 42 subjects. Data collection was carried out using a questionnaire to obtain subject demographic data and physical activity, as well as physical examinations complemented by laboratory examinations and MDA levels. The mean age of the subjects was 52.7 years, and most of the 32 (76.2%) subjects were categorized as obese and had high activity. Pearson correlation test obtained body mass index statistically significantly correlated with plasma MDA levels with a value of  $p = 0.039$ , as well as physical activity statistically significantly correlated with a negative correlation, with a value of  $r = -0.26$  and  $p = 0.043$ . Conclusion Body mass index and activity are significantly correlated with plasma MDA levels.

**Keywords:** malondialdehyde, stress, oxidative, radicals

## INTRODUCTION

Aging is a progressive process caused by a physiological decline in body function.<sup>1</sup> A decrease in body homeostasis is accompanied by an increased risk of various chronic diseases including diabetes mellitus, cancer, neurodegenerative diseases, and heart disease. It is known that several factors can accelerate aging, namely age, lifestyle, oxidative stress, lipid peroxidation, and mitochondrial damage. Previous studies have shown that changes in sphingomyelin metabolism in the central nervous system and age are associated with neurodegenerative diseases.<sup>2</sup> In the US, aging is one of the most important factors in diabetes mellitus and heart disease. In 2015, an estimated 12% of the population had diabetes mellitus and an undetected 24% had diabetes mellitus. Heart disease is the highest contributor to mortality in the population aged 65-84 years.<sup>3</sup> Recent studies have shown that non-diabetic populations whose fasting glucose levels increase in the blood will increase the risk of coronary heart disease. But on the other hand, the data on fasting blood glucose increase is still debated in the population of cardiovascular disease patients who do not have diabetes mellitus. A recent study in North Sumatra showed a significant relationship between MDA levels in obese and non-obese populations.<sup>4</sup>

Free radicals are formed through two processes, namely

exogenous and endogenous processes. Various pollutants such as cigarette smoke, motor vehicle fumes, industrial waste, and exposure to sunlight/ultraviolet will form free radicals. Illness, stress, and excessive exercise/physical activity are endogenous factors produced by the human body that can form free radicals. The formation of free radicals occurs due to oxidation processes and metabolic processes that take place during exercise or activity, inflammatory processes, consumption of certain foods, and others. Free radicals will begin to activate with molecules in the surrounding cells to obtain electrons to stabilize the condition of one's body. The electrons that have been taken in the cell molecules will trigger the occurrence of free radicals. If this continues for a long time, it can cause oxidative stress which can cause inflammation, and DNA disease can also trigger the disease. Oxidative stress conditions characterized by increased production of Reactive Oxygen Species (ROS) have implications for various diseases such as hypertension, atherosclerosis, diabetes mellitus, stroke, chronic renal failure, heart failure, and other chronic diseases.<sup>5-7</sup>

Oxidative stress is an imbalance between oxidants (free radicals) and antioxidants. Free radicals that commonly cause oxidative damage are reactive oxygen species (ROS).<sup>8</sup> ROS compounds are formed through aerobic metabolism. These compounds can cause extensive damage through catalytic reactions. They are responsible for damage at the cellular level

by oxidizing proteins, lipids, DNA (Deoxyribonucleic acid), and other biomolecules. Various compounds can be used as markers of oxidative stress such as malondialdehyde (MDA), and protein carbonylation. 4-hydroxy-2-nonenal (4HNE), and 8-hydroxy-deoxyguanosine (8OHdG).<sup>9</sup>

Malondialdehyde (MDA) is a result of lipid peroxidation in the body. The concentration of MDA indicates an oxidation process that occurs in the body's cell membranes due to metabolic processes. One of the factors that cause aging is the production of excess reactive oxygen species (ROS). ROS are generated through the process of cell respiration in the mitochondria and the phagocytosis process of foreign body elimination. To stabilize the amount of ROS, the body can use endogenous and exogenous antioxidants. Disruption of the balance between ROS and antioxidants can cause irreversible damage to cell organelle components up to the DNA level.<sup>10,11</sup>

MDA levels are strongly influenced by many factors such as age, illness, stress, various pollutants, food consumption, antioxidant supplements and excessive physical activity or exercise, and others. The other study found that more than 30% of subjects had high MDA levels, and concluded that there was a significant relationship between eating habits, body mass index, glucose levels with MDA levels, as well as smoking habits significantly related to MDA levels.<sup>12</sup> Recent studies have shown that plasma MDA levels in populations aged 70 years and 60 years are not significantly different but there is an increase in the specific activity of the catalase enzyme that increases with age.<sup>10</sup> In another study, there was a significant difference in plasma MDA levels in the obesity group without metabolic syndrome compared to the obesity group with metabolic syndrome.<sup>4</sup>

There are still variations in MDA data in various groups with different ages and different disease groups, so this study aims to determine plasma MDA levels in the age group 40 years and over and factors that can affect plasma MDA levels.

## METHOD

This observational analytic study with a cross-sectional design approach was conducted from November 2022 to February 2023 in Angke village, West Jakarta, Indonesia. The inclusion criteria for this study were: men and women aged  $\geq 40$  years, willing to participate in the study and sign informed consent. Meanwhile, the exclusion criteria were having a history of malignancy, kidney failure, a history of autoimmune disease, taking drugs related to psychiatric conditions, and having limitations in mobility. This study used the calculation of sample size with the formula  $N / 1 + N \cdot \alpha^2$ , and obtained the minimum number of samples required was 40 subjects. Demographic data and physical activity were obtained from distributed questionnaires. Blood chemistry examination (complete blood, glucose, total cholesterol, liver enzymes (SGOT and SGPT) using an enzymatic method. Examination of malondialdehyde levels was carried out using the spectrophotometric method. This research has obtained approval from the ethics committee of the Faculty of Medicine, Trisakti University, Number: 178/KER/FK/1X/2022.

## RESULTS

A total of 42 subjects participated in this study, most of the subjects were female 27 (64.3%), and the mean age of the subjects was 52.7 years. Anthropometric data of the subjects are as follows body weight  $67.53 \pm 13.64$  and height  $156.02 \pm 8.73$  cm, while for abdominal circumference the average is 93.09 cm. Most of the subjects 59.5% fell into the normotensive category, with an average systolic blood pressure of 141.62 mmHg and an average diastolic blood pressure of 82.45 mmHg. A total of 32 (76.2%) subjects were

categorized as obese with BMI  $\geq 25.0$ , and most subjects had high activity levels. (Table 1)

Table 1: Distribution of Subject Characteristics (n=42)

Characteristics	n (%)	Mean $\pm$ SD
Gender		
Male	15 (35.7)	
Female	27 (64.3)	
Blood pressure (mmHg)		
Normal ( $< 140/90$ )	25 (59.5)	
Hypertension ( $\geq 140/90$ )	17 (40.5)	
Physical Activity		
Low ( $< 600$ Mets)	11 (26.2)	
High ( $\geq 600$ Mets)	31 (73.8)	
Body Mass Index (kg/m <sup>2</sup> )		27.69 $\pm$ 4.90
Not obese ( $< 25.0$ )	10 (23.8)	
Obese ( $\geq 25.0$ )	32 (76.2)	
Age (years)		52.7 $\pm$ 10.60
Body weight (kg)		67.53 $\pm$ 13.64
Height (cm)		156.02 $\pm$ 8.73
Abdominal circumference (cm)		93.09 $\pm$ 10.54
Blood pressure (mmHg)		
Systole		141.62 $\pm$ 20.77
Diastole		82.45 $\pm$ 12.70

The examination of the subject's plasma malondialdehyde levels obtained a mean of 0.77 with the highest value of 1.03 and the lowest of 0.51. Hemoglobin levels averaged 13.16 g/dL. Fasting blood glucose level was  $95.06 \pm 41.01$  mg/dL and total cholesterol was  $179.83 \pm 31.85$  mg/dL. Laboratory examinations to determine liver function were carried out by measuring SGPT and SGOT levels, obtaining mean values of 24.21 U/L and 26.31 U/L respectively. (Table 2)

Table 2: Laboratory examination results of subjects (n=42)

Variable	Mean $\pm$ SD
Hemoglobin (g/dL)	13.16 $\pm$ 1.56
Fasting blood glucose (mg/dL)	95.06 $\pm$ 41.01
Total cholesterol (mg/dL)	179.83 $\pm$ 31.85
SGPT (U/L)	24.21 $\pm$ 13.50
SGOT (U/L)	26.31 $\pm$ 11.51
Plasma malondialdehyde	0.77 $\pm$ 0.26

Determining the type of correlation test to be used by first conducting a data normality test. The normality test used was Shapiro-Wilk because the number of subjects was less than 50 subjects. The correlation test used to assess the correlation between blood glucose, total cholesterol, SGPT, SGOT levels, age, body mass index and physical activity with plasma MDA levels was the Pearson correlation test. The test results are presented in Table 3. Blood glucose, total cholesterol, SGPT,



SGOT, and age did not have a significant correlation with MDA levels. Body mass index has a significant correlation with plasma MDA ( $p = 0.039$ ), with a positive correlation direction, and the strength of the correlation is statistically weak with a value of  $r = 0.396$  ( $<0.4$ ). The physical activity variable has a significant correlation with plasma MDA levels ( $p=0.043$ ) with a negative correlation direction but with a statistically weak correlation strength ( $r=-0.26$ ).

Table 3: Correlation Test of Age, Gender, Glucose, SGOT, SGPT, BMI, Physical Activity with Plasma MDA Levels (n=42)

	Plasma MDA Level	
	r	p
Age	0.098	0.756
Gender	0.124	0.108
Glucose	0.330	0.835
SGOT	0.104	0.513
SGPT	0.008	0.614
Body Mass Index	0.396	0.039*
Physical activity	-0.260	0.043*

\*  $p < 0.05$

## DISCUSSION

The level of oxidative stress can be measured using Malondialdehyde (MDA) concentration. MDA is the result of lipid peroxidation, which will cause cell damage. Oxidant compounds such as free radicals or non-radical species attack lipids that contain carbon-carbon double bonds, especially unsaturated fatty acids (PUFA). MDA is one of the biomarkers widely used to assess oxidative stress. The mean serum MDA in this study 0.77 was found to be lower than the previous study <sup>10</sup>, this is because the age difference of the subjects in this study was younger than in the previous study. In contrast, a study conducted in Iraq on hypertensive patients concluded that the increase in MDA was associated with an increase in age, gender, and duration of disease.<sup>13</sup>

Free radicals such as ROS are the etiology of most chronic diseases, as it is known that active free radicals are the result of metabolic reactions. Damage to body tissue cells occurs because active free electrons remain looking for other electrons to settle, if no other electrons are available then active free electrons will attack cells resulting in cell damage. Although free radicals naturally have benefits for the body, when the level of free radicals exceeds the normal value, it will cause harm to the body.<sup>11,12</sup>

Measurement of oxidative stress can be done by measuring plasma malondialdehyde (MDA) levels. MDA is a secondary result of the lipid peroxidation process, producing various active compounds that will damage cells. The process of MDA formation through enzymatic and non-enzymatic processes. Currently, MDA is one of the biomarkers used to assess oxidative stress.<sup>14</sup> Some previous researchers concluded that higher serum MDA was found in patients with hyperlipidemia. The condition of increased fat content in the body is in line with increased oxidative stress. Hyperlipidemia is associated with physical changes in cell membranes that facilitate the release of free radicals from the mitochondrial electron transport chain or activation of NADPH oxidase. In patients with diabetes mellitus (DM) there is also an increase in MDA levels, malondialdehyde interacts irreversibly and reversibly with proteins and phospholipids to have a profound effect, especially on the collagen of the cardiovascular system. MDA levels remain within normal limits due to the antioxidant

effect, but in DM, this protective effect is impaired resulting in increased MDA levels.<sup>1,15</sup>

Increased lipid peroxidation is considered a consequence of oxidative stress that occurs when the dynamic balance between prooxidants and antioxidants is disrupted. The statin class of drugs for hyperlipidemia therapy is proven to be anti-atherogenic, can improve endothelial function, inhibit vascular inflammation, and stabilize atherosclerotic plaques. In addition to statins, non-steroidal anti-inflammatory drugs (NSAIDs) can affect MDA concentration. Previous studies have found that NSAIDs have significant antioxidant properties to scavenge free radicals.<sup>16,17</sup>

In this study, there was no significant correlation between age and MDA levels. Similar results were also obtained by other researchers, cell damage due to oxidative stress is not related to increasing age but is associated with habits, behavior patterns, and physiological changes due to age. Various habitual patterns such as smoking, fatty food habits, lack of activity, and inflammatory processes can lead to increased oxidative stress. The habit of taking vitamin C (ascorbic acid) has a beneficial impact due to its antioxidant effect so cell damage and DNA damage are lower in this group.<sup>18</sup> MDA levels were significantly higher among all levels of smokers. MDA levels were lower in light smokers compared to heavy or moderate smokers. Higher vitamin C levels were found in the non-smoker group. All body tissues are susceptible to free radical damage, but based on location, the respiratory epithelium is particularly vulnerable to cigarette smoke. Oxygen radicals generated close to the cell membrane oxidize membrane phospholipids which will result in lipid peroxidation with the end products of this reaction being malondialdehyde, ethane, and pentane. this causes an increase in MDA levels in smokers.<sup>19</sup> Another study conducted in Klaten on foundry workers concluded that type of work, body mass index and smoking habits did not affect MDA levels, but length of work and marital status were associated with MDA levels. Long and continuous exposure to metallic materials will result in the accumulation of these materials in the body and will increase the oxidative process of polyunsaturated fats in cell membranes and worsen the level of lipid peroxidase, characterized by an increase in MDA levels as a reflection of the intensity and rate of lipid peroxidase reactions of damage to body cells.<sup>20</sup>

Hardiany's study also concluded that there was no significant difference in plasma MDA levels in the age population, but increasing age along with an increase in catalase-specific activity.<sup>10</sup> This increase in catalase-specific activity is a protective effect especially for elderly women to overcome high levels of oxidative damage. The role of endogenous antioxidant catalase is to suppress oxidative damage.<sup>18,21</sup> Oxidative stress plays an important role in the incidence of cardiovascular diseases including coronary artery disease. Oxidative stress is considered an important mechanism for the development of cardiovascular diseases especially atherosclerosis. Hypertension, cardiac hypertrophy, and ischemia-reperfusion injury are three other major cardiovascular conditions for which gender is a risk factor. Gender is also associated with differences in oxidative stress. Under physiological conditions, women appear to be less susceptible to oxidative stress. This may be due to the antioxidant properties of estrogen, sex differences in NADPH-oxidase activity or other mechanisms yet to be determined. Sex hormones including estrogen are globally present with their receptors in the heart so sex hormones can exert effects on the cardiovascular system. Estrogen can improve heart function and reduce injury to the heart. In addition, estrogen has antioxidant effects so that it can function as a cardioprotective.<sup>22,23</sup>

This study found a significant correlation between body mass index and MDA levels, the higher the BMI value, the higher the MDA levels. Similar results were also obtained in a study in India that concluded that in metabolic syndrome patients with one of the signs is an increase in BMI, there is an increase in MDA levels compared to the control group. In obese subjects, the level of MDA is higher than normal IMT. In a healthy body, there is redox hemostasis that occurs in cells, if an imbalance arises it will cause oxidative stress. Obese patients will have an increased metabolic load and mechanical load on the myocardium, increased body mass, and excessive food intake will cause the formation of lipid peroxidation, and free radicals.<sup>24</sup> The increase in some of these things will stimulate antioxidant enzymes in the long run, this will cause antioxidant enzyme reserves to be depleted and unable to overcome and balance the increase in oxidative stress.<sup>25,26</sup>

These types of training activities are exhausting, long exercise, overtraining syndrome, various types of exercise will induce a significant response to oxidative stress. Exercise activities such as light exercise, low-intensity exercise, and prolonged training will have an impact on improving endogenous antioxidant status. Reactive oxygen species play an important role in cell signaling and in regulating antioxidant gene expression. Exertion results in hyperregulation of nuclear factor kappa B and mitogen-activated protein kinases that activate gene expression of a number of enzymes and proteins with important roles in maintaining intracellular oxidative or antioxidant homeostasis.<sup>1,6,27</sup> The limitation of this study is that some factors that may affect MDA levels such as consumption of vitamin supplements, diet, and length of illness suffered were not examined in this study.

## CONCLUSIONS

Body mass index and activity were significantly correlated with MDA levels, while age, gender, blood glucose levels, and liver enzymes were not significantly correlated with MDA levels.

## Conflict of Interest:

The investigators declare there is no conflict of interest in this study.

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# Factors Associated with Plasma Malondialdehyde Levels in People over 40 Years

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# Factors Associated with Plasma Malondialdehyde Levels in People over 40 Years

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## Acknowledgments 13

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**Abstract.** Aging is a progressive process caused by a physiological decline in body functions. It is known that several factors can accelerate aging, namely age, lifestyle, oxidative stress, lipid peroxidation, and mitochondrial damage. Various pollutants such as cigarette smoke, motor vehicle fumes, industrial waste, and exposure to sunlight / ultraviolet light will form free radicals. Malondialdehyde (MDA) level is one of the indicators used to measure oxidative stress. Increased MDA levels are found in various degenerative diseases. This study aims to determine plasma MDA levels in the age group of 40 years and over and factors that can affect plasma MDA levels. This study was a cross-sectional study by including 42 subjects. Data collection was carried out using a questionnaire to obtain subject demographic data and physical activity, as well as physical examinations complemented by laboratory examinations and MDA levels. The mean age of the subjects was 52.7 years, and most of the 32 (76.2%) subjects were categorized as obese and had high activity. Pearson correlation test obtained body mass index statistically significantly correlated with plasma MDA levels with a value of  $p = 0.039$ , as well as physical activity statistically significantly correlated with a negative correlation, with a value of  $r = -0.26$  and  $p = 0.043$ . Conclusion Body mass index and activity are significantly correlated with plasma MDA levels.

**Keywords:** malondialdehyde, stress, oxidative, radicals

## INTRODUCTION

Aging is a progressive process caused by a physiological decline in body function. <sup>(1)</sup> A decrease in body homeostasis is accompanied by an increased risk of various chronic diseases including diabetes mellitus, cancer, neurodegenerative diseases, and heart disease. It is known that several factors can accelerate aging, namely age, lifestyle, oxidative stress, lipid peroxidation, and mitochondrial damage. Previous studies have shown that changes in sphingomyelin metabolism in the central nervous system and age are associated with neurodegenerative diseases. <sup>(2)</sup> In the US, aging is one of the most important factors in diabetes mellitus and heart disease. In 2015, an estimated 12% of the population had diabetes mellitus and an undetected 24% had diabetes mellitus. Heart disease is the highest contributor to mortality in the population aged 65-84 years. <sup>(3)</sup> Recent studies have shown that non-diabetic populations whose fasting glucose levels increase in the blood will increase the risk of coronary heart disease. But on the other hand, the data on fasting blood glucose increase is still debated in the population of cardiovascular disease patients who do not have diabetes mellitus. A recent study in North Sumatra showed a significant relationship between MDA levels in obese and non-obese populations. <sup>(4)</sup>

Free radicals are formed through two processes, namely exogenous and endogenous processes. Various pollutants such as cigarette smoke, motor vehicle fumes, industrial waste, and exposure to sunlight/ultraviolet will form free radicals. Illness, stress, and excessive exercise/physical activity are endogenous factors produced by the human body that can form free radicals. The formation of free radicals occurs due to oxidation processes and metabolic processes that take place during exercise or activity, inflammatory processes, consumption of certain foods, and others. Free radicals will begin to activate with molecules in the surrounding cells to obtain electrons to stabilize the condition of one's body. The electrons that have been taken in the cell molecules will trigger the

occurrence of free radicals. If this continues for a long time, it can cause oxidative stress which can cause inflammation, and DNA disease can also trigger the disease. Oxidative stress conditions characterized by increased production of Reactive Oxygen Species (ROS) have implications for various diseases such as hypertension, atherosclerosis, diabetes mellitus, stroke, chronic renal failure, heart failure, and other chronic diseases.<sup>(5-7)</sup>

Oxidative stress is an imbalance between oxidants (free radicals) and antioxidants. Free radicals that commonly cause oxidative damage are reactive oxygen species (ROS).<sup>(8)</sup> ROS compounds are formed through aerobic metabolism. These compounds can cause extensive damage through catalytic reactions. They are responsible for damage at the cellular level by oxidizing proteins, lipids, DNA (Deoxyribonucleic acid), and other biomolecules. Various compounds can be used as markers of oxidative stress such as malondialdehyde (MDA), and protein carbonylation. 4-hydroxy-2-nonenal (4HNE), and 8-hydroxy-deoxyguanosine (8OHdG).<sup>(9)</sup>

Malondialdehyde (MDA) is a result of lipid peroxidation in the body. The concentration of MDA indicates an oxidation process that occurs in the body's cell membranes due to metabolic processes. One of the factors that cause aging is the production of excess reactive oxygen species (ROS). ROS are generated through the process of cell respiration in the mitochondria and the phagocytosis process of foreign body elimination. To stabilize the amount of ROS, the body can use endogenous and exogenous antioxidants. Disruption of the balance between ROS and antioxidants can cause irreversible damage to cell organelle components up to the DNA level.<sup>(10,11)</sup>

MDA levels are strongly influenced by many factors such as age, illness, stress, various pollutants, food consumption, antioxidant supplements and excessive physical activity or exercise, and others. The other study found that more than 30% of subjects had high MDA levels, and concluded that there was a significant relationship between eating habits, body mass index, glucose levels with MDA levels, as well as smoking habits significantly related to MDA levels.<sup>(12)</sup> Recent studies have shown that plasma MDA levels in populations aged 70 years and 60 years are not significantly different but there is an increase in the specific activity of the catalase enzyme that increases with age.<sup>(10)</sup> In another study, there was a significant difference in plasma MDA levels in the obesity group without metabolic syndrome compared to the obesity group with metabolic syndrome.<sup>(4)</sup>

There are still variations in MDA data in various groups with different ages and different disease groups, so this study aims to determine plasma MDA levels in the age group 40 years and over and factors that can affect plasma MDA levels.

## **METHOD**

This observational analytic study with a cross-sectional design approach was conducted from November 2022 to February 2023 in Angke village, West Jakarta, Indonesia. The inclusion criteria for this study were: men and women aged  $\geq 40$  years, willing to participate in the study and sign informed consent. Meanwhile, the exclusion criteria were having a history of malignancy, kidney failure, a history of autoimmune disease, taking drugs related to psychiatric conditions, and having limitations in mobility. This study used the calculation of sample size with the formula  $N / 1 + N.\alpha^2$ , and obtained the minimum number of samples required was 40 subjects. Demographic data and physical activity were obtained from distributed questionnaires. Blood chemistry examination (complete blood, glucose, total cholesterol, liver enzymes (SGOT and SGPT) using an enzymatic method. Examination of malondialdehyde levels was carried out using the spectrophotometric method. This research has obtained approval from the ethics committee of the Faculty of Medicine, Trisakti University, Number: 178/KER/FK/1X/2022.

## **RESULTS**

A total of 42 subjects participated in this study, most of the subjects were female 27 (64.3%), and the mean age of the subjects was 52.7 years. Anthropometric data of the subjects are as follows body weight  $67.53 \pm 13.64$  and height  $156.02 \pm 8.73$  cm, while for abdominal circumference the average is 93.09 cm. Most of the subjects 59.5% fell into the normotensive category, with an average systolic blood pressure of 141.62 mmHg and an average diastolic blood pressure of 82.45 mmHg. A total of 32 (76.2%) subjects were categorized as obese with BMI  $\geq 25.0$ , and most subjects had high activity levels. (Table 1)

Table 1. Distribution of Subject Characteristics (n=42)

Characteristics	n (%)	Mean $\pm$ SD
Gender		
Male	15 (35.7)	
Female	27 (64.3)	
Blood pressure (mmHg)		
Normal (< 140/90)	25 (59.5)	
Hypertension ( $\geq$ 140/90)	17 (40.5)	
Physical Activity		
Low (< 600 Mets)	11 (26.2)	
High ( $\geq$ 600 Mets)	31 (73.8)	
Body Mass Index (kg/m <sup>2</sup> )		27.69 $\pm$ 4.90
Not obese (< 25.0)	10 (23.8)	
Obese ( $\geq$ 25.0)	32 (76.2)	
Age (years)		52.7 $\pm$ 10.60
Body weight (kg)		67.53 $\pm$ 13.64
Height (cm)		156.02 $\pm$ 8.73
Abdominal circumference (cm)		93.09 $\pm$ 10.54
Blood pressure (mmHg)		
Systole		141.62 $\pm$ 20.77
Diastole		82.45 $\pm$ 12.70

The examination of the subject's plasma malondialdehyde levels obtained a mean of 0.77 with the highest value of 1.03 and the lowest of 0.51. Hemoglobin levels averaged 13.16 g/dL. Fasting blood glucose level was 95.06  $\pm$  41.01 mg/dL and total cholesterol was 179.83  $\pm$  31.85 mg/dL. Laboratory examinations to determine liver function were carried out by measuring SGPT and SGOT levels, obtaining mean values of 24.21 U/L and 26.31 U/L respectively. (Table 2)

Table 2. Laboratory examination results of subjects (n=42)

Variable	Mean $\pm$ SD
Hemoglobin (g/dL)	13.16 $\pm$ 1.56
Fasting blood glucose (mg/dL)	95.06 $\pm$ 41.01
Total cholesterol (mg/dL)	179.83 $\pm$ 31.85
SGPT (U/L)	24.21 $\pm$ 13.50
SGOT (U/L)	26.31 $\pm$ 11.51
Plasma malondialdehyde	0.77 $\pm$ 0.26

Determining the type of correlation test to be used by first conducting a data normality test. The normality test used was Shapiro-Wilk because the number of subjects was less than 50 subjects. The correlation test used to assess the correlation between blood glucose, total cholesterol, SGPT, SGOT levels, age, body mass index and physical activity with plasma MDA levels was the Pearson correlation test. The test results are presented in Table 3. Blood glucose, total cholesterol, SGPT, SGOT, and age did not have a significant correlation with MDA levels. Body mass index has a significant correlation with plasma MDA ( $p = 0.039$ ), with a positive correlation direction, and the strength of the correlation is statistically weak with a value of  $r = 0.396$  ( $< 0.4$ ). The physical activity variable has a significant correlation with plasma MDA levels ( $p = 0.043$ ) with a negative correlation direction but with a statistically weak correlation strength ( $r = -0.26$ ).



Table 3. Correlation Test of Age, Gender, Glucose, SGOT, SGPT, BMI, Physical Activity with Plasma MDA Levels (n=42)

	Plasma MDA Level	
	r	p
Age	0.098	0.756
Gender	0.124	0.108
Glucose	0.330	0.835
SGOT	0.104	0.513
SGPT	0.008	0.614
Body Mass Index	0.396	0.039*
Physical activity	0.260	0.043*

\* p < 0.05

## Discussion

The level of oxidative stress can be measured using Malondialdehyde (MDA) concentration. MDA is the result of lipid peroxidation, which will cause cell damage. Oxidant compounds such as free radicals or non-radical species attack lipids that contain carbon-carbon double bonds, especially unsaturated fatty acids (PUFA). MDA is one of the biomarkers widely used to assess oxidative stress. The mean serum MDA in this study 0.77 was found to be lower than the previous study <sup>(10)</sup>, this is because the age difference of the subjects in this study was younger than in the previous study. In contrast, a study conducted in Iraq on hypertensive patients concluded that the increase in MDA was associated with an increase in age, gender, and duration of disease.<sup>(13)</sup>

Free radicals such as ROS are the etiology of most chronic diseases, as it is known that active free radicals are the result of metabolic reactions. Damage to body tissue cells occurs because active free electrons remain looking for other electrons to settle, if no other electrons are available then active free electrons will attack cells resulting in cell damage. Although free radicals naturally have benefits for the body, when the level of free radicals exceeds the normal value, it will cause harm to the body.<sup>(11,12)</sup>

Measurement of oxidative stress can be done by measuring plasma malondialdehyde (MDA) levels. MDA is a secondary result of the lipid peroxidation process, producing various active compounds that will damage cells. The process of MDA formation through enzymatic and non-enzymatic processes. Currently, MDA is one of the biomarkers used to assess oxidative stress.<sup>(14)</sup> Some previous researchers concluded that higher serum MDA was found in patients with hyperlipidemia. The condition of increased fat content in the body is in line with increased oxidative stress. Hyperlipidemia is associated with physical changes in cell membranes that facilitate the release of free radicals from the mitochondrial electron transport chain or activation of NADPH oxidase. In patients with diabetes mellitus (DM) there is also an increase in MDA levels, malondialdehyde interacts irreversibly and reversibly with proteins and phospholipids to have a profound effect, especially on the collagen of the cardiovascular system. MDA levels remain within normal limits due to the antioxidant effect, but in DM, this protective effect is impaired resulting in increased MDA levels.<sup>(1,15)</sup>

Increased lipid peroxidation is considered a consequence of oxidative stress that occurs when the dynamic balance between prooxidants and antioxidants is disrupted. The statin class of drugs for hyperlipidemia therapy is proven to be anti-atherogenic, can improve endothelial function, inhibit vascular inflammation, and stabilize atherosclerotic plaques. In addition to statins, non-steroidal anti-inflammatory drugs (NSAIDs) can affect MDA concentration. Previous studies have found that NSAIDs have significant antioxidant properties to scavenge free radicals.<sup>(16,17)</sup>

In this study, there was no significant correlation between age and MDA levels. Similar results were also obtained by other researchers, cell damage due to oxidative stress is not related to increasing age but is associated with habits, behavior patterns, and physiological changes due to age. Various habitual patterns such as smoking, fatty food habits, lack of activity, and inflammatory processes can lead to increased oxidative stress. The habit of taking vitamin C (ascorbic acid) has a beneficial impact due to its antioxidant effect so cell damage and DNA damage are lower in this group.<sup>(18)</sup> MDA levels

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were significantly higher among all levels of smokers. MDA levels were lower in light smokers compared to heavy or moderate smokers. Higher vitamin C levels were found in the non-smoker group. All body tissues are susceptible to free radical damage, but based on location, the respiratory epithelium is particularly vulnerable to cigarette smoke. Oxygen radicals generated close to the cell membrane oxidize membrane phospholipids which will result in lipid peroxidation with the end products of this reaction being malondialdehyde, ethane, and pentane. this causes an increase in MDA levels in smokers.<sup>(19)</sup> Another study conducted in Klaten on foundry workers concluded that type of work, body mass index and smoking habits did not affect MDA levels, but length of work and marital status were associated with MDA levels. Long and continuous exposure to metallic materials will result in the accumulation of these materials in the body and will increase the oxidative process of polyunsaturated fats in cell membranes and worsen the level of lipid peroxidase, characterized by an increase in MDA levels as a reflection of the intensity and rate of lipid peroxidase reactions of damage to body cells.<sup>(20)</sup>

Hardiany's study also concluded that there was no significant difference in plasma MDA levels in the age population, but increasing age along with an increase in catalase-specific activity.<sup>(10)</sup> This increase in catalase-specific activity is a protective effect especially for elderly women to overcome high levels of oxidative damage. The role of endogenous antioxidant catalase is to suppress oxidative damage.<sup>(18,21)</sup> Oxidative stress plays an important role in the incidence of cardiovascular diseases including coronary artery disease. Oxidative stress is considered an important mechanism for the development of cardiovascular diseases especially atherosclerosis. Hypertension, cardiac hypertrophy, and ischemia-reperfusion injury are three other major cardiovascular conditions for which gender is a risk factor. Gender is also associated with differences in oxidative stress. Under physiological conditions, women appear to be less susceptible to oxidative stress. This may be due to the antioxidant properties of estrogen, sex differences in NADPH-oxidase activity or other mechanisms yet to be determined. Sex hormones including estrogen are globally present with their receptors in the heart so sex hormones can exert effects on the cardiovascular system. Estrogen can improve heart function and reduce injury to the heart. In addition, estrogen has antioxidant effects so that it can function as a cardioprotective.<sup>(22,23)</sup>

This study found a significant correlation between body mass index and MDA levels, the higher the BMI value, the higher the MDA levels. Similar results were also obtained in a study in India that concluded that in metabolic syndrome patients with one of the signs is an increase in BMI, there is an increase in MDA levels compared to the control group. In obese subjects, the level of MDA is higher than normal IMT. In a healthy body, there is redox hemostasis that occurs in cells, if an imbalance arises it will cause oxidative stress. Obese patients will have an increased metabolic load and mechanical load on the myocardium, increased body mass, and excessive food intake will cause the formation of lipid peroxidation, and free radicals.<sup>(24)</sup> The increase in some of these things will stimulate antioxidant enzymes in the long run, this will cause antioxidant enzyme reserves to be depleted and unable to overcome and balance the increase in oxidative stress.<sup>(25,26)</sup>

These types of training activities are exhausting, long exercise, overtraining syndrome, various types of exercise will induce a significant response to oxidative stress. Exercise activities such as light exercise, low-intensity exercise, and prolonged training will have an impact on improving endogenous antioxidant status. Reactive oxygen species play an important role in cell signaling and in regulating antioxidant gene expression. Exertion results in hyperregulation of nuclear factor kappa B and mitogen-activated protein kinases that activate gene expression of a number of enzymes and proteins with important roles in maintaining intracellular oxidative or antioxidant homeostasis.<sup>(1,6,27)</sup> The limitation of this study is that some factors that may affect MDA levels such as consumption of vitamin supplements, diet, and length of illness suffered were not examined in this study.

## Conclusions

Body mass index and activity were significantly correlated with MDA levels, while age, gender, blood glucose levels, and liver enzymes were not significantly correlated with MDA levels.



**Conflict of Interest:**

The investigators declare there is no conflict of interest in this study.

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