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Original Article

Effectiveness of Mar'ke Bilar Drink Formulation on Pro- Anti-Free Radicals in The Blood of Women of Childbearing Age Operators in Gas Stations

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Abstract

Background : Women of childbearing age who wortk at gas stations are vulnerable to pollution exposure due to fuel emissions. The workers are susceptible to heavy metal (Pb) poisoning in the fuel content. This can cause health problems such as reproductive disorders, impaired kidney function and can even cause anemia in women. The aims of this study was to determine the effectiveness of Mar'ke Bilar formulation on Pb, MDA, and SOD in blood.
Methods : The study was Quasi Experimental with a pre and post test group design that allows researchers to see the comparison of the control group with the experimental group. The sample in the study was Women of childbearing age who worked as operators at gas stations with a total of 60 people and were determined based on purposive sampling with inclusion criteria then divided into 4 treatment groups. Data analysis used the T-dependent test to see the differences in each group then continued with the One Way ANOVA and Post Hoc-Tukey tests.
Results : The results of the study showed that the three of Mar'ke Bilar formulations were effective in reducing Pb and MDA levels, as well as increasing SOD levels, which are endogenous antioxidant enzymes that are important in the body's defense against oxidative stress. While formula 1 proved to be the most effective in increasing SOD levels.
Conclusion : Mar'ke Bilar affects blood Pb, MDA, and SOD levels and formulation 1 is the most effective in increasing SOD levels. It is necessary to consume colored foods such as Marke'Bilar which are very good for avoiding diseases caused by free radicals by air and water pollution, especially those caused by heavy metals.

Keywords : Mar'ke Bilar, Pb, MDA, SOD, Free radicals

INTRODUCTION

Health problems in humans, which comes from air, water, soil pollution and motor vehicle smoke and also factory waste. Air pollution contributes to the health of workers and communities in the open air such as highways and their surroundings and in factory environments. Traffic police, street vendors, buskers at red lights, beggars and Public Fuel Station (SPBU) operators, are one of the groups of people who are most vulnerable to Pb exposure, where Pb is used as a mixture to improve fuel quality so that the combustion process becomes better.[1]

Operators at gas stations, especially women of childbearing age are vulnerable to exposure to Pb metals, especially when the vehicle is burning.[2] The location of the gas station which is also close to the highway, can increase the risk of exposure to heavy metals from vehicles traveling on the highway.[3]

The entry of Pb into the human body through the respiratory or digestive tract, then accumulates in the blood and through the reduction-oxidation cycle will produce free radicals in the form of *Reactive Oxygen Species* (ROS) including Hydrogen Peroxide ((H_2O_2), Nitrogen Oxide (NO), Oxygen Peroxide (O_2-).[4]

Several studies on women of childbearing age have found that Pb can cause reproductive disorders, increased cases of infertility, menstrual disorders, decreased sexual libido, toxicity to reproductive organs and can also cause disorders in Hb formation which causes decreased work productivity.[2,5]

Pb as a free radical trigger, can damage endogenous antioxidant enzymes such as GPx *(Glutathione Peroxidase)*, CAT *(Catalase)*, and SOD *(Superoxide dismutase)*. Other disorders resulting from the accumulation of Pb in the body in women of childbearing age can cause cervical cancer and breast cancer[6] due to disruption of the signal transduction process in DNA which can disrupt gene expression (DNA).

Moreover, the accumulation of Pb in the body can also cause gene mutations so that during the fertile period, if fertilization occurs, it can cause miscarriage or even initiation into the fetus, resulting in defects in the fetus and the baby being born and the worst prognosis is giving birth to a child with autism.[7]

An increase in *Malondialdehyde* (MDA) and a decrease in SOD *(Superoxide dismutase)* are markers of ongoing oxidative stress. SOD as an endogenous antioxidant and MDA as a marker of free radicals in the body are effects caused by exposure to Pb metal.[8] The application of the antioxidant system is a mechanism in self-defense against pollutant stress characterized by the loss or decrease in the content of antioxidant vitamins.[5]

Endogenous antioxidants obtained from the foods we consume daily are able to stimulate the antioxidant enzymatic system (SOD) and can reduce the formation of ROS and its effects on health. *Foodstuffs* containing vitamins A, and C and the mineral zinc (Zn) and bioactive substances can be found in colored foods and are found to have radicals scavenger ability.[9,10]

Persimmon *(Diospyros kki L.)* and purple passion fruit *(Ipomoea Batatas Var. antin 3)* are known to contain bioactive compounds and antioxidants such as carotenoids, anthocyanins, flavonoids, vitamin C, zinc, and Fe.[11] One hundred grams of persimmon contains vitamin A 81 IU, and vitamin C 7.5 mg, while purple passion fruit contains vitamin A 2410 IU and vitamin C 28mg. These components can help eliminate free radicals caused by heavy metal exposure by inhibiting free radicals' toxicity.[12] Persimmon and purple passion fruit are typical fruits of North Sumatra. Purple sweet potato is widely available in North Sumatra, especially in mountainous areas. Starch from purple sweet potato can be used as a sugar substitute, as well as a thickener and stabilizer for beverage products.[13] In 100 gr of purple sweet potato contains vitamin A 7700 IU and vitamin C 22 mg.

The three food ingredients derived from fruit and tubers have the potential to be made into a healthy drink called Mar'ke Bilar, through the process of sorting ingredients, steaming, heating, blending, filtering to become fruit juice that is ready to drink. The combination of these three ingredients has a sweet taste, a passion fruit aroma like citrus and a purplish pink color so that it increases the appetite for consumption. This drink has gone through an acceptance process with organoleptic tests and examination of nutritional and phytochemical content at the FMIPA and Agricultural Product Technology Laboratory, Brawijaya University.[13]

Mar'ke Bilar is a functional drink that combines fruits and tubers that are expected to complement each other, especially the bioactive and antioxidant components, so that it can be an option as a functional drink. The fruits and tubers used are local food products, easy to obtain so that the processing process becomes easier.

Based on the description above, it is necessary to conduct research that aims to determine the effectiveness of Mar'ke Bilar drink formulations on pro (Pb, MDA) and anti-free radicals (SOD) on Women of Childbearing age operators at gas stations.

METHODS

This type of research was a *quasi-experiment with pre- and Post Test Design with Control*.[14] The research was conducted for a month from February 10th to March 10th, 2023 at several gas stations that were willing to be used as research sites in Medan and surrounding areas. Only 8 gas stations were willing to be used as research locations, considering that due to their busy schedules, the time available was more limited. The research

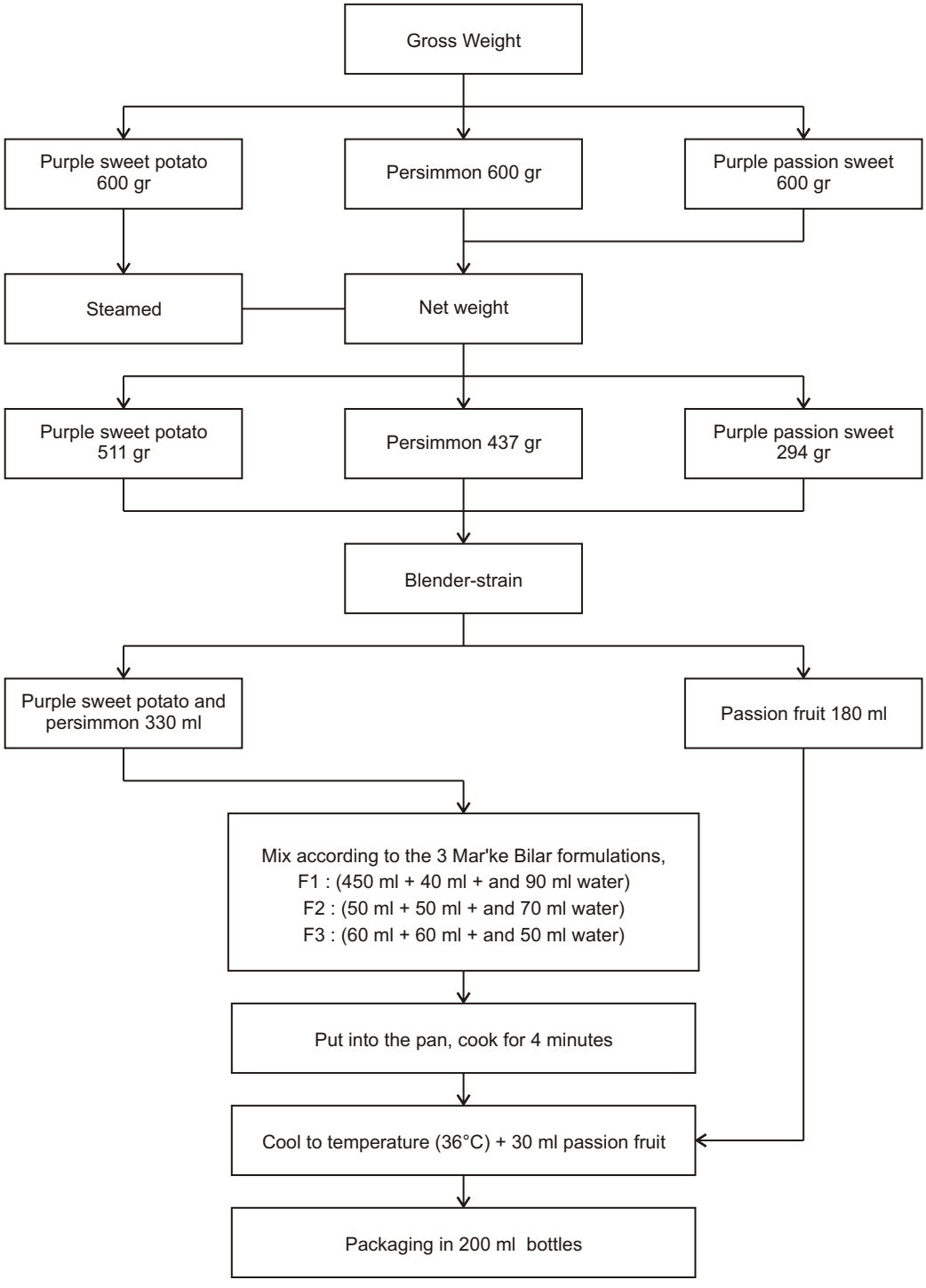

Figure 1. Procedure for Making Mar'ke Bilar Drink

population was Women of childbearing age as operators at gas stations in total 83 people The sample was determined by setting inclusion criteria: The sample was determined by purposive sampling, where researchers took samples based on the conditions and willingness to be sampled, so that researchers obtained samples combined with accidental sampling. The inclusion criteria were aged 20–35 years, still menstruating, not sick and actively working every day except holidays. Exclusion criteria: female gas station operators who are not pregnant because pregnant women are prone to initiation of heavy metal exposure to the fetus, Women of childbearing age in gas station operators who have worked ≤ 1 year, gas station operators who work part-

TABLE 1
Nutrient Content of Mar'ke Bilar with Various Formulations

	Marke Bilar Beverage Formulation			
Nutrients	**F1**	**F2**	**F3**	**Unit**
Vitamin C	51.73	56.81	65.87	mg/g
Beta carotene	2.79	4.49	5.1	µg/g
Zinc (Zn)	0.86	0.92	0.70	mg/kg
Iron (Fe)	3.93	4.68	4.42	mg/kg
Anthocyanins	170.99	233.5	268.51	ppm
Antioxidants	40.75	39.73	38.49	mg/ml
Phenol	0.38	0.57	0.91	%

Source: Brawijaya University Agricultural Products Laboratory in 2023

TABLE 2
Distribution based on sample characteristics

Characteristics		**Variable**	**Frequency Percent**
Sample		**N**	**%**
Age	20–30 years old	30	50.0
	>30–35 years old	30	50.0
	Total	60	100
Education	Primary School	1	1.7
	Junior High School	2	3.3
	Senior High School	57	95.0
	Total	60	100
Length of Service	<13 months	14	23.3
	13–24 months	18	30.0
	>24 months	28	46.7
	Total	50	100

time, male gas station operators, gas station operators aged > 35 years and < 20 years. Based on the criteria, a sample of 60 people was obtained, obtained from the availability and willingness of the gas station management and woman of childbearing age operators by signing an informed consent. then divided into 4 groups based on the close distance between gas stations, made into 1 group to achieve a balanced division between the control group, formulation group 1, formulation group 2 and formulation group 3. Mar'ke Bilar was given once a day at shift change times (≤ 20 minutes of time provided by the manager) for 14 days, at 14.00 to 14.20 WIB. To avoid bias against the accuracy of the provision of Mar'ke Bilar drinks, the sample was also controlled for nutrient intake that could be a confounding factor in this study, such as protein, vitamin A, vitamin C, and zinc intake. The process of providing Mar'ke Bilar drinks was directly supervised by researchers and enumerators based on the formulation. Here are 4 groups based on the formulation:

1. The control group (15 people) was given red syrup (a syrup drink with a color similar to Mar'ke Bilar made from a mixture of sugar and fruit-flavored essence) 200 ml/day.
2. Formulation group 1 (15 people) was given 200 ml/day of Mar'ke Bilar consisting of: purple sweet potato 40 g, persimmon 40 g, purple passion fruit 30 g, sugar 15 g and water 90 ml.

TABLE 3
Nutrient Intake Before and After Mar'ke Bilar Administration

Indicator	Control	*p*	F1	*p*	F2	*p*	F3	*p*
	Before	After	Before	After	Before	After	Before	After
	Average ±SD	Average ±SD	Average ±SD	Mean ±SD	Average ±SD	Mean±SD	Average ±SD	Mean ±SD
Protein (gr)	57.95 ± 8.82	61.64 ± 12.77	0.194 72.56 ± 20.5	75.56 ± 15.17 0.609	9 55.2 ± 23.25	56.9 ± 23.46 0.218	67.35 ± 21.46	69.67 ± 23.28 0.339
Vitamin A (mg)	68.00 ± 13.08	75.74 ± 8.87	0.237 65.74 ± 13.95	67.13 ± 23.69 0.563	74.35 ± 12.65	72.62 ± 18.45 0.183	71.84 ± 15.08	72.56 ± 24.93 0.217
Vitamin C (mg)	51.89 ± 6.39	57.13 ± 8.46	0.212 66.67 ± 18.13	71.22 ± 20.23 0.500	75.56 ± 15.33	78.37 ± 11.05 0.101	81.88 ± 21.67	83.42 ± 26.84 0.264
Zinc (mg)	7.11 ± 20.22	7.08 ± 18.70	0.117 7.318 ± 15.81	7.54 ± 22.62 0.427	7.67 ± 11.38	7.71 ± 25.10 0.617	7.61 ± 18.17	7.85 ± 09.17 0.352

TABLE 4
Distribution of Mean, SD of Pb levels before and after Mar'ke Bilar administration

Group	Pb	n	Min	Max	Mean ± SD	*p Value*
Control	Before	15	0.001	9.160	1.978 ± 2.687	0.09*
	After	13	1.750	30.500	9.434 ± 8.971	
	Difference	–	24.4050	0.800	7.471 ± 7.795	
Formulation 1	Before	15	2.260	17.380	8.680 ± 4.939	0.002*
	After	12	0.001	6.910	2.443 ± 2.271	
	Difference	–	4.250	13.770	5.713 ± 5.130	
Formulation 2	Before	15	2.530	19.550	9.227 ± 5.139	0.001*
	After	15	0.001	11.170	4.529 ± 4.630	
	Difference	–	2.210	19.549	4.697 ± 4.840	
Formulation 3	Before	15	1.570	23.070	12.438 ± 7.064	0.001*
	After	15	0.001	8.870	3.391 ± 3.418	
	Difference	–	0.100	18.279	9.046 ± 5.682	

Notes: *Paired T Test

3. Formulation group 2 (15 people) was given 200 ml/day of Mar'ke Bilar consisting of: purple sweet potato 50 g, persimmon 50 g, purple passion fruit 30 g, sugar 15 g and water 70 ml.
4. Formulation group 3 (15 people) was given 200 ml/day of Mar'ke Bilar consisting of: purple sweet potato 60 g, persimmon 60 g, purple passion fruit 30 g, sugar 15 g and water 50 ml.

The nutrient content and procedure for making Mar'ke Bilar can be seen in Figure 1 and Table 1.

Blood samples were taken as much as 2cc from the left upper arm using a 2.5 cc syringe by a medical laboratory technology analyst. Then, the blood was put into a tube containing ethylenediaminetetraacetic acid (EDTA) solution and then separated into serum to be examined in the Molecular Laboratory of the Faculty of Medicine and in the FMIPA Laboratory of Brawijaya University Malang. Malondialdehyde and superoxide dismutase examinations were carried out using the enzyme-linked immunosorbent assay (ELISA) kit method with a spectrophotometer (ELx808 Ultra Microplate Reader) and Pb examinations were carried out using the Atomic Absorbance Spectrophotometry method (AAS, Shimadzu AA-6200 & AA6300). Before data analysis, the data normality was first tested using Kolmogrov-smirnov and was found $p>0.05$. Then the

TABLE 5
MDA Level Data Before and After Mar'ke Bilar Administration

Group	MDA	n	Min	Max	Mean ± SD	*p Value*
Control	Before	15	4.24	10.59	9.85 ± 1.74799	0,436*
	After	13	8.21	15.85	9.88 ± 1.980	
	Difference	–	6.35	10.59	1.29 ± 4.654	
Formulation 1	Before	15	4.30	12.30	7.53 ± 2.12947	0.001*
	After	12	4.20	7.75	6.23 ± 0.921	
	Difference	–	1.32	8.74	2.56 ± -3.155	
Formulation 2	Before	15	6.28	15.85	11.48 ± 2.96045	0.001*
	After	15	7.23	10.20	8.38 ± 0.767	
	Difference	–	1.54	6.76	3.22 ± -2.677	
Formulation 3	Before	15	3.51	13.11	9.15 ± 2.31775	0.001*
	After	15	2.80	7.88	6.83 ± 1.256	
	Difference	–	1.31	6.35	2.32 ± -1.817	

Notes: *Paired T Test

TABLE 6
SOD Level Data Before and After Mar'ke Bilar Administration

Group	SOD (ppm)	n	Min	Max	Mean ± SD	*p Value*
Control	Before	15	14.59	39.31	25.31 ± 7.519	0.327*
	After	13	16.48	42.61	25.97 ± 16.48	
	Difference	–	22.83	16.02	0.0349 ± 9.775	
Formulation 1	Before	15	16.02	45.03	29.56 ± 6.926	0.001*
	After	12	19.18	66.28	49.78 ± 19.18	
	Difference	–	9.89	31.90	11.11 ± 2.127	
Formulation 2	Before	15	16.22	34.81	24.47 ± .281	0.001*
	After	15	18.98	60.91	39.62 ± 18.98	
	Difference	–	6.26	40.61	8.358 ± 13.190	
Formulation 3	Before	15	16.73	69.55	28.67 ± 13.530	0.001*
	After	15	25.24	80.28	45.22 ± 25.24	
	Difference	–	1.61	44.00	9.4801 ± 13.283	

Notes: *Paired T Test

data was analyzed using the T-dependent test to see the differences before and after the levels of MDA, SOD and Pb in each treatment of different formulations. Meanwhile, to see the most effective changes, One Way Anova test was carried out, then continued with a Post Hoc-Tukey test. This study has obtained ethical approval from the Ethics Commiitte of the Poltekkes Kemenkes Medan No: 01.1447/KEPK/Poltekkes Kemenkes Medan/ 2023.

RESULTS

The distribution of sample characteristics, nutrient intake before and after Mar'ke Bilar, minimum, maximum,

TABLE 7
The mean difference in Pb, MDA, and SOD levels between groups after Mar'ke Bilar Drink Administration

Indicator	Control	Formulation 1	Formulation 2	Formulation 3
Pb	+7.471 ± 7.795 [a]	5.713 ± 5.130[b]	4.697 ± 4.840[b]	9.046 ± 5.682[b]
MDA	1.29 ± 4.654[a]	2.56 ± 3.155[b]	3.22 ± 2.677[b]	2.32 ± 1.817[b]
SOD	0.03 ± 9.775[a]	11.11 ± 12.127[c]	8.35 ± 13.190[b]	9.48 ± 13.28[b]

ANOVA Test and Post Hoc-Tukey

mean, and SD values as well as the difference in Pb, MDA, and SOD levels before and after Mar'ke Bilar drink administration can be seen in the figures and tables below.

Table 2 showed that 60 samples of women of childbearing age aged 20–30 years were found in as many as 30 people (50%). Most of the respondents had a high school education as many as 57 people (95%), samples with elementary education were 1 person (1.7%) and junior high school was 2 people (3.3%). Based on the length of work, the largest sample was > 24 months, namely 28 people (46.7%), <13 months of work for as many as 18 people (30%). Based on the length of employment, the largest sample was in the gas station operator group >24 months, namely 28 people (46.7%). The length of employment will affect the condition of Women of childbearing age gas station operators who get tired easily and sometimes have difficulty concentrating. This is in accordance with Stamara's 2020 research. Pb as a heavy metal produces toxic effects and free radicals enter the body only for a few months if exposed to Pb with a working duration of 8–9 hours.[15]

Table 3 showed that the intake of nutrients in Women of childbearing age working as gas station operators has no difference before and after the Mar'ke Bilar healthy drink treatment, which is > 0.05. Nutrient intake before and after treatment was tested using a *paired t-test* to control the effectiveness of the Mar'ke Bilar healthy drink treatment. However, when viewed from the average number in each treatment group there is an increase in the intake of protein, vitamin A, vitamin C and zinc in each group tends to increase but some have decreased. But when viewed from the adequacy of Women of childbearing age aged 20–35 years in the control group and formulation group 2 looks still below. As for the adequacy of vitamin A of 600 mg/day, the daily intake of vitamin C is still below normal, for vitamin C intake the adequacy is 75 mg/day, and only formulation groups 2 and 3 have met the recommended adequacy rate. As for the adequacy of zinc at the age of women of childbearing age of 8 mg/day, while what is obtained based on the recall results is still below the AKG 2019.

Table 4 Shows a decrease in Pb levels after being given Mar'ke Bilar in formula groups 1, 2, and 3. The results of the paired t-test showed that the administration of Mar'ke Bilar showed a significant difference in formulation groups 1, 2, and 3, p<0.05. While in the control group without Mar'ke Bilar administration, there was an increase in the average Pb level from 1.978 µg/dl to 9.434 µg/dl with a difference of .7.471 µg/dl. When tested based on the paired t-test before and after p>0.05.

Table 5 shows a decrease in MDA levels after being given Mar'ke Bilar in formula groups 1, 2, and 3. The results of the paired t-test showed that the administration of Mar'ke Bilar showed a significant difference in formula groups 1, 2, and 3, p<0.05. While in the control group without Mar'ke Bilar administration, there was an increase in the average MDA level from 9.85 nmol/mL to 9.88 nmol/mL with a difference of 1.29 nmol/mL. When tested based on the paired t-test before and after p> 0.05.

Table 6 shows a decrease in SOD levels after being given Mar'ke Bilar in formula groups 1, 2, and 3. The results of the paired t-test showed that the administration of Mar'ke Bilar showed a significant difference in formula groups 1, 2, and 3, p<0.05. While in the control group without the administration of Mar'ke Bilar, there was an increase in the average MDA level from 25.31 ppm to 25.97 ppm with a difference of 0.0349 ppm. When tested based on the paired t-test before and after p> 0.05.

Table 7 shows that after a one-way ANOVA statistical test was conducted to see the differences between treatment groups, then a post hoc-tukey test was continued to see which group had the most influence for each Pb, MDA, and SOD levels. In the Pb examination indicator, the notation shows that for the control group there was a difference towards an increase indicated by the notation a (Table 4). While after the administration of Mar'ke Bilar in formula groups 1, 2 and 3 showed a decrease indicated by the notation changing to b.

In the MDA examination indicator, the notation shows that for the control group there was a difference towards an increase indicated by the notation a (Table 5). While after the administration of Mar'ke Bilar in formula groups 1,2 and 3 showed a decrease indicated by the notation changing to b.

In the SOD examination indicator, the notation shows that for the control group there was a difference towards an increase indicated by the notation a (Table 6).

Meanwhile, after the provision of Mar'ke Bilar, it turns out that formula group 1 is significantly different from formula groups 2 and 3 which are shown with the notation c, while formulations 2 and 3 are in the notation b.

DISCUSSION

Sample Characteristics

The sample in this study amounted to 60 women of childbearing age. Based on the division of age groups, the percentage is divided into 2, namely 50% aged 20–30 years and the other 50% for the group >30–35 years. The selection of characteristics based on the age of Women of Childbearing Age is based on the fact that at the age of 20–29 years the process of maturation of a woman's reproductive cells is at its peak of maturity. Women in the age range of 20–29 years have a 95% chance of experiencing pregnancy, and will decrease to 90% if Women of Childbearing Age has reached the age of 30 years.[16]

If poisoning occurs due to Pb metal from exposure to motor vehicle fumes in women of childbearing age, it can cause reproductive system disorders in Women of Childbearing Age gas station operators by preventing infertility, decreased sex libido, and miscarriage, where according to Azami, *et al* (2022) states that the activity of biotransformation enzymes decreases and the resistance of certain organs to the effects of Pb decreases. This causes the older a person is, the higher the accumulation of Pb in body tissues will be.[17]

Based on the length of employment, 28 people (46.7%) were found to have worked for more than 24 months. Gas station employees who have worked for ≥4 years as operators have an average blood lead (Pb) level of 24.97 µg/dL. This is in line with the results of research by Stamara Ghina (2020) and Yeni (2021) which reported that women who work as gas station operators have blood Pb levels of 20.00 µg/dL.[15] According to the Centers for Disease Control and Prevention (CDC) in the United States and the Indonesian Ministry of Health, the normal threshold value is <10 µg/dL(1820), while the World Health Organization (WHO) sets an acceptable limit of ≤5 µg/dL.[19,21] Lead enters the body through the respiratory tract and then into the bloodstream, causing enzyme inactivation and acting as a highly reactive radical.[2]

Effect of Mar'ke Bilar on Pb levels

The results showed that there were differences before and after the administration of Mar'ke Bilar with various formulations (1, 2, and 3) on the blood Pb levels of Women of childbearing age working as gas station operators which tended to decrease. While in the control group, there was an increase in Pb levels without being given Mar'ke Bilar. The results of the ANOVA test were continued with the Post Hoc-Tukey Test, it turned out that the control group had different notations (a) with formulation groups 1, 2, and 3 (b). After being given the treatment, there was a decrease that could be seen from the difference in numbers that showed differences in notation. Mar'ke Bilar (Passion Fruit, Persimmon and Purple Sweet Potato) drink is one of the processed healthy drinks that contain antioxidants that can help prevent oxidative damage and prevent damage caused by Pb invasion into the blood including increasing free radicals, lysing erythrocytes and inactivating several enzymes in the body. Mar'ke Bilar contains vitamins that act as antioxidants such as Beta-carotene(pro-vitamin A)[22] and Vitamin C and the mineral Zinc. In addition, Mar'ke Bilar also contains bioactive substances anthocyanins and phenols that function to bind free radicals due to the accumulation of Pb in the blood.[23]

Vitamin A that functions as an antioxidant is beta-carotene (pro-vitamin A) obtained from colored foods that tend to be yellow and orange, such as food ingredients for making Mar'Ke bilar drinks. Beta-carotene antioxidants act as scavengers of free radical reactions and have a key role in preventing oxidative stress that occurs due to Lead in the body as antioxidants that act as scavengers that can collect free radical molecules in the body. Vitamin A is a natural antioxidant compound that fights free radical reactions and has a key role in preventing oxidative stress that occurs due to Lead in the body.[24]

Vitamin C can stimulate gelatin compounds that can reduce the availability of Pb metal in the blood. Vitamin C also has the function of translocating Pb metal to the disposal estuary either through the respiratory or urinary system. Vitamin C performs immobilization of heavy metals to reduce the absorption of Pb in the small intestine and vitamin C can inhibit the uptake of Pb at the cellular level so that the cytotoxicity process of Pb becomes decreased or even completely undetectable.[24–26]

Other antioxidants in the form of zinc minerals and bioactive substances such as anthocyanins and phenols in the Mar'ke Bilar content are able to suppress the movement of lead so that it does not interact with sulfhydryl groups or Pb metal cofactors in enzymes and molecules so that the metal prosthetic group cannot be replaced by Pb.[10,27]

This is in accordance with Sundari's 2017 study, where in this study, vitamin C given orally as much as 200 mg was found to be able to reduce Pb levels in female street sweepers in Denpasar, Bali.[25] Giving vitamin C and iron was able to neutralize Pb in the body so as to prevent the hemolytic process during the formation of Hb in adolescent girls.[28] While beta-carotene can reduce homocysteine in workers exposed to Pb.[19] Other studies of giving vitamin C to experimental animals, mice and

humans, the effectiveness of vitamin C in treating poisoning due to Pb metal, especially on neurotoxic effects.[29]

Effect of Mar'ke Bilar Administration on MDA Levels

The results showed that there were differences before and after the administration of Mar'ke Bilar with various formulations (1, 2, and 3) on the blood MDA levels of Women of childbearing age working as gas station operators which tended to decrease. While in the control group, there was an increase in MDA levels without being given Mar'ke Bilar.

Mar'ke Bilar has the special feature of being made from three food ingredients that are rich in antioxidants and bioactive substances that complement each other, this can be seen from the total antioxidant content in the three formulations (Formulation 1 is 40.75 mg/ml, formulation 2 is 39.73, formulation 3 is 38.49) which shows very strong activity.[11] The results of the ANOVA test were continued with the Tukey Post Hoc Test, it turned out that the control group had different notations (a) with formulation groups 1, 2, and 3 (b). After being given the treatment there was a decrease that could be seen from the difference in numbers that showed differences in notation.

Mar'ke Bilar drink can reduce MDA levels which are free radical biomarkers due to Pb metal induction. Excessive Pb increases that invade the blood, resulting in increased reactions between Pb and unsaturated fatty acids commonly called lipid peroxidation. The occurrence of lipid peroxidation causes increased MDA levels in the body. The content of vitamin C, beta-carotene, and zinc minerals as well as their bioactive content can reduce MDA by protecting PUFA phospholipid membranes by donating Hydrogen ions (H+) to lipid peroxyl radicals (LOO*).[30] Free radicals damage cell membranes, where the main component is unsaturated fatty acids (PUFA) which contain double bonds so that they are more susceptible to free radical attack and will cause oxidative destruction of lipids from fatty acids called lipid peroxides. (Fauziah 2018, and Dewangga 2022).[31,32]

Mar'ke Bilar drink is also able to maintain the stability of the cell membrane where lipid peroxidation occurs so that the intensity of MDA formation can be inhibited. Another way antioxidants can break the chain of peroxy radical bonds with other molecules. This event will inhibit the formation of other peroxy radical chains so that MDA levels are not formed.[33]

Another function of Mar'ke Bilar which contains several antioxidants such as vitamin C helps form active free radicals into ascorbyl radicals which have a low reactive level. Vitamin C and beta-carotene are able to act as free radical scavengers by changing free radicals into reactive ones by providing hydrogen ions, including neutralizing singlet oxygen part of ROS.

The antioxidant content in Mar'ke Bilar such as Vitamin C, Betacarotene, and zinc minerals can stimulate the antioxidant enzymatic system to reduce the formation and effects of ROS so that MDA levels in the blood can decrease.[9] Other abilities of exogenous antioxidants include being able to repair body cells, neutralize reactive free radicals, especially oxygen types, then biochemically able to remain in the body.[9] Lee's 2018 research examining zinc in foodstuffs turned out to have a function that is an antioxidant and regulates oxidation-reduction reactions to be conditioned to decrease.[10] This research is also supported by other studies that provide antioxidant content in the form of vitamin C, and zinc which can prevent lipid peroxides in the liver and tissues so that the formation of oxidative stress can be prevented.[24,27]

Effect of Mar'ke Bilar Administration on SOD Levels

The results showed that there were differences before and after the administration of Mar'ke Bilar with various formulations (1, 2, and 3) on the blood SOD levels of Women of childbearing age working as gas station operators which tended to increase. In the control group, there was an increase but very small where the average increase was 0.0349. The results of the ANOVA test continued with the Post Hoc-Tukey Test, it turned out that the control group had different notations (a) with the formulation groups 2 and 3 (b) and the most effective in increasing SOD was formulation 1. After treatment there was an increase that could be seen from the difference in numbers that showed differences in notations. Formulation 1 with a relatively lower content of antioxidants and bioactive substances compared to formulations 2 and 3 turned out to be the most effective formulation. This is because the high antioxidant content in a food can also interfere with the production of enzymatic oxidants and even excessive doses can cause cell death.[10]

Other studies have stated that giving high doses of antioxidants can also be toxic and can also affect the rate of oxidation as the task of antioxidants.[20]

Mar'ke Bilar drink is a drink that is high in exogenous antioxidants and bioactive substances that can stimulate enzyme activity including SOD.[34] Another mechanism in increasing SOD by Mar'ke Bilar drink is that antioxidants and bioactive substances directly donate hydrogen ions and can neutralize the toxic effects of Pb and suppress the formation of free radicals so that they can directly increase endogenous antioxidant gene expression through several mechanisms.[35] The mechanism of increasing endogenous antioxidant gene expression is through the activation pathway of *Nuclear Factor Erythroid 2 Related Factor 2* (NRF2). This gene is

involved in the synthesis of endogenous antioxidant enzymes including SOD.[36,37]

SOD enzyme is an endogenous antioxidant that works by regulating ROS, thus the role of molecules that have activity as exogenous antioxidants such as colored fruits and vegetables and other foodstuffs that are high in pigment content of bioactive substances are needed to be consumed in counteracting oxidative stress.[38]

Dewangga's research in 2022, using purple sweet potato as an antioxidant, was able to increase blood SOD as an endogenous antioxidant.[32] Research conducted by Mochida in 2022 by giving persimmon was able to increase SOD due to the potential antioxidant content of persimmon.[32,39]

Exogenous antioxidants are compounds or systems that can safely interact with free radicals and stop the chain reaction before important molecules are damaged. Antioxidants (eg, flavonoids phenolic acids, tannins, vitamin C, and vitamin E) have diverse biological properties, such as anti-inflammatory, anticarcinogenic and anti-atherosclerosis effects. Several types of vitamins are important antioxidants for the body. Antioxidants that come from food include vitamin C, vitamin E, vitamin A, selenium, and zinc.[40]

Vitamin C is one of the water-soluble vitamins that has reducing properties so that this vitamin has important properties as an antioxidant that affects the body's redox potential. The role of vitamin C is good in helping to neutralize antioxidants, especially lead (Pb), because Pb also has oxidative stress properties, vitamin C is also able to be a free radical scavenger and can react directly. The results of a study conducted by Fauzi Susilo (2008) also found the same thing that giving vitamin C to albino mice at a dose of 0.2 mg/g/day for 36 days can protect nerve cells from free radical compounds caused by exposure to the heavy metal Pb. Another study by Silitonga (2018) stated that vitamin C can ward off free radicals, bind and clean heavy metals due to exposure from the environment.

Antioxidants increase the function of immune cells, maintain the integrity and function of lipids in cell membranes so that they can protect cellular proteins and nucleic acids that function to regulate signal transduction and DNA expression by means of DNA repair so that cancer prevention can occur. [26,31]

The process of preventing cancer in Women of Childbearing Age can also be done through preventing free radical invasion through inhibiting NF-κβ activation, preventing apoptosis which ultimately prevents the target of cancer p<38 in activating cancer cells.[41]

Antioxidants can also prevent disorders in the reproductive system of Women of Childbearing Age gas station operators by preventing infertility, decreased sex libido, miscarriage, and also increasing the regulation of luteinizing hormone which will affect the gonadotropin hormone produced by the pituitary gland in the brain which is needed by Woman of Childbearing age in the ovulation process.[19,38]

The urgency of antioxidants in Woman of Childbearing age gas station operators is the binding of Pb due to exposure to motor vehicle fumes. This process can increase the amount of lead in blood plasma so that Pb toxicity occurs, where antioxidants prevent an increase in Zinc Protoporphyrin (ZPP) levels so that the heme formation process can take place properly. Antioxidants also work to inhibit the activity of deta aminolevulinic acid dehydratase (ALAD), extend the life of red blood cells, maintain membrane stability, so that anemia in Women of Childbearing Age can be prevented.[19,21,26]

The impact on Women of Childbearing Age gas station operators, if consuming antioxidants in sufficient quantities will increase fitness and concentration while working can run optimally.[19]

The limitation of this study, it should be tested first on experimental animals as a pre-clinical test, so that the accuracy of the sample homogeneity process can be maintained. The number of samples obtained is still not maximal, this is because many gas station owners do not allow the blood drawing process to be carried out during working hours so that the blood drawing seems to be taken in a hurry. A calm atmosphere when taking blood can affect the results of the variable examination.

CONCLUSION

Giving Mar'ke Bilar formula 1, 2, and 3 reduces Pb and MDA levels and increases SOD levels, where formula 1 is the most effective treatment in increasing SOD levels. It is necessary to consume colored foods such as Mar'ke Bilar which are very good for avoiding diseases caused by free radicals caused by air and water pollution, especially those caused by heavy metals.

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REFERENCES

1. Klopfleisch B, Sutomo AH, Iravati S. Kadar timbal pada petugas stasiun pengisian bahan bakar umum. Ber Kedokt Masy. 2017;33(4):205.
2. Windusari Y, Aini IN, Setiawan A, Aetin EN. Deteksi Frekuensi Distribusi Timbal Dalam Darah Pekerja Pengisi Bahan Bakar: Studi Kasus SPBU di Plaju, Sumatera Selatan. J Kesehat Lingkung Indones. 2019;18(1):62.
3. Almunjiat E, Sabilu Y, Ainurrafiq A. ANALISIS RISIKO KESEHATAN AKIBAT PAJANAN TIMBAL (Pb) MELALUI JALUR INHALAS PADA OPERATOR DI STASIUN

PENGISIAN BAHAN BAKAR UMUM (SPBU) DI KOTA KENDARI TAHUN 2016 (STUDI DI SPBU TIPULU, WUA-WUA, ANDUONOHU DAN SPBU LEPO-LEPO). J Ilm Mhs Kesehat Masy Unsyiah. 2018;1(3):185158.
4. Sani A, Darma AI, Abdullahi IL, Musa BU, Imam FA. Heavy metals mixture affects the blood and antioxidant defense system of mice. J Hazard Mater Adv. 2023;11(July):100340.
5. Mayaserli DP, Renowati R, Biomed M. ANALISIS KADAR LOGAM TIMBAL (Pb) PADA RAMBUT KARYAWAN SPBU. Sainstek J Sains dan Teknol. 2018;9(1):19.
6. Ningtyas AW. Analisa Komparasi Kadar Logam Berat Mercuri pada Urine Pasien Kanker Payudara dengan Kadar Logam Berat Marcuri pada Urine Wanita Sehat. J Inohim. 2013;1(2):88–98.
7. Ding M, Shi S, Qie S, Li J, Xi X. Association between heavy metals exposure (cadmium, lead, arsenic, mercury) and child autistic disorder: a systematic review and meta-analysis. Front Pediatr [Internet]. 2023;11(July):1–15. Available from: https://doi.org/10.3389/fped.2023.1169733
8. Anggraeni S, Setyaningrum T, Listiawan MY. Perbedaan Kadar Malondialdehid (MDA) sebagai Petanda Stres Oksidatif pada Berbagai Derajat Akne Vulgaris. Berk Ilmu Kesehat Kulit dan Kelamin Period Dermatology Venereol. 2017;29(1):36–43.
9. Javed M, Ahmad MI, Usmani N, Ahmad M. Multiple biomarker responses (serum biochemistry, oxidative stress, genotoxicity and histopathology) in Channa punctatus exposed to heavy metal loaded waste water /704/172/4081 /631/601 article. Sci Rep. 2017;7(1):1–11.
10. Lee SR. Critical role of zinc as either an antioxidant or a prooxidant in cellular systems. Oxid Med Cell Longev. 2018;2018.
11. Kusumah SH, Pebrianti SA, Maryatilah L. Test the Antioxidant Activity of Fruit and Purple Passion Fruit Syrup Using Dpph Method. 2021;2(1):2746–1209.
12. Makmun A, Rusli FIP. Pengaruh Vitamin C Terhadap Sistem Imun Tubuh Untuk Mencegah Dan Terapi Covid-19. Molucca Medica. 2020;12:60–4.
13. Siahaan G, Tarigan N, Siregar IR. Daya Terima dan Uji Kandungan Fitokimia (Antioksidan , Serat , Indeks Glikemik , dan Vitamin C) p ada Minuman Sehat Mar ' ke Bilar sebagai Alternatif Pencegahan Obesitas Acceptability and Phytochemical Assessment (Antioxidant, Fiber, Glycemic Index ,. 2023;7(2):224–31.
14. Harun I, Susanto H, Rosidi A. Pemberian tempe menurunkan kadar malondialdehyde (MDA) dan meningkatkan aktivitas enzim superoxide dismutase (SOD) pada tikus dengan aktivitas fisik tinggi. J Gizi dan Pangan. 2017;12(3):211–6.
15. Rinawati D, Barlian B, Tsamara G. IDENTIFIKASI KADAR TIMBAL (Pb) DALAM DARAH PADA PETUGAS OPERATOR SPBU 34–42115 KOTA SERANG. J Med (Media Inf Kesehatan). 2020;7(1):1–8.
16. Siahaan G, Widodo E, Lestrina D, Oppusunggu R. Impact of Mar'ke Bilar Healthy Drink on Blood Levels of Malondialdehyde, Superoxide Dismutase, and Hemoglobin in Soccer Athletes at PPLP North Sumatra. Asian J Sport Med . 2024;15(1).
17. Azami M, Tardeh Z, Mansouri A, Soleymani A, Sayehmiri K. Mean blood lead level in iranian workers: A systematic and meta-analysis. Iran Red Crescent Med J. 2018;20(1).
18. Yenni M, Sugiarto, Husaini A. Analisis Kadar Logam Timbal Darah Petugas Stasiun Pengisian Bensin Umum (Spbu) Kota Jambi. Cetak) J Innov Res Knowl. 2021;1(5):773.
19. Putra A, Fitri WE, Febria fuji astuti. Toksisitas Logam Timbal Terhadap Kesehatan Dan Lingkungan. J Kesehat Med Saintika. 2023;14(1):158–74.
20. Simanjuntak K. PERAN ANTIOKSIDAN FLAVONOID DALAM MENINGKATKAN KESEHATAN. Adv Ceram Mater. 2012;3(4):328–31.
21. Suhartono. Pajanan TImbal (Pb) dan Kesehatan Ibu. 2022. 59 p.
22. Fadlilah AR, Lestari K. Review: Peran Antioksidan Dalam Imunitas Tubuh. Farmaka. 2023;21(2):171–8.
23. Andrés Juan C, Manuel Pérez de la Lastra J, Plou FJ, Pérez-Lebeña E, Reinbothe S. Molecular Sciences The Chemistry of Reactive Oxygen Species (ROS) Revisited: Outlining Their Role in Biological Macromolecules (DNA, Lipids and Proteins) and Induced Pathologies. Int J Mol Sci. 2021;22:4642.
24. Kamilatussaniah, Yuniastuti A IR. Pengaruh Suplementasi Madu Kelengkeng terhadap Kadar TSA dan MDA Tikus Putih yang Diinduksi Timbal (Pb). J MIPA. 2015;38(2):108–14.
25. Sundari LPR, Dinata IMK, SHA LMI. Pemberian Vitamin C 250 Mg Per Oral Menurunkan Kadar Timbal Darah Wanita Penyapu Jalan di Kota Denpasar. J Med. 2017;48(3):185–8.
26. Ahmad Khan W. Preventive Agent of Vitamin C Against PB-Induced Toxicity: A Review. Int J Adv Biol. 2022;9(3):1–13.
27. Wiyasihati SI, Wigati KW. Potensi Bayam Merah (Amaranthus tricolor L) sebagai Antioksidan pada Toksisitas Timbal yang Diinduksi pada Mencit. Maj Kedokt Bandung. 2016;48(2):63–7.
28. Levanta DS, Hananingtyas I. Paparan timbal dalam urin remaja pada kejadian gangguan sistem saraf dan keseimbangan di kecamatan Curug. Public Heal Risk Assesment J. 2023;1(1):38–55.
29. Autifi MAH, Mohamed WY, Abdul Haye WM, Elbaz KR. The Possible Protective Role of Vitamin C against Toxicity Induced by Lead Acetate in Liver and Spleen of Adult Albino Rats (Light and Electron Microscopic Study). Egypt J Hosp Med. 2018;73(10):7650–8.
30. dos Reis LCR, Facco EMP, Salvador M, Flôres SH, de Oliveira Rios A. Antioxidant potential and physicochemical characterization of yellow, purple and orange passion fruit. J Food Sci Technol. 2018;55(7):2679–91.
31. Fadila AR, Kurniasari R. Pengaruh Edukasi Gizi Menggunakan Media Terhadap Peningkatan Pengetahuan dalam Upaya Pencegahan Obesitas pada Remaja di MTs Al-Khairiyah. J Untuk Masy Sehat. 2022;6(2):113–9.
32. Dewangga MW, Irianto DP. POTENSI UBI JALAR UNGU (Ipomoea batatas var Antin 3) SEBAGAI ASUPAN ANTIOKSIDAN UNTUK ATLET. J Tumbuh Obat Indones. 2022;15(2):136–45.
33. Raeeszadeh M, Karimfar B, Amiri AA, Akbari A. Protective Effect of Nano-Vitamin C on Infertility due to Oxidative Stress Induced by Lead and Arsenic in Male Rats. J Chem. 2021;2021.
34. Khairani S, Made N, Sandhiutami D, Permata I, Farmakologi S, Farmasi F, *et al*. Peningkatan Aktivitas Sod Dan Penurunan Kadar Mda Dari Ekstrak Etanol Daun Lobak (Raphanus Sativus L) Pada Mencit The Effect Of Ethanol Extract Of Radish (Raphanus Sativus L.) In Increasing Sod Antioxidant Activity And Decreasing Mda Levels In Mice. Farmasains. 2023;10(1):1–8.
35. Ighodaro OM, Akinloye OA. First line defence antioxidants-superoxide dismutase (SOD), catalase (CAT) and glutathione peroxidase (GPX): Their fundamental role in the entire antioxidant defence grid. Alexandria J Med. 2018;54(4):287–93.
36. Altobelli GG, Van Noorden S, Balato A, Cimini V. Copper/Zinc Superoxide Dismutase in Human Skin: Current Knowledge. Front Med. 2020;7(May):1–8.
37. Arslan AS, Seven I, Mutlu SI, Arkali G, Birben N, Seven PT. Potential ameliorative effect of dietary quercetin against lead-induced oxidative stress, biochemical changes, and apoptosis in laying Japanese quails. Ecotoxicol Environ Saf. 2022;231:113200.

38. Sheilaadji MU, Listiawan MY, Ervianti E. Hubungan Kadar Antioksidan Superoxide Dismutase (SOD) dengan Indeks Bakterial (IB) pada Pasien Kusta Baru Tipe Multibasiler (MB) tanpa Reaksi (Correlation of Superoxide Dismutase (SOD) Antioxidant Level with Bacterial Index (IB) in New Multibacillary (MB). Berk Ilmu Kesehat Kulit dan Kelamin – Period Dermatology Venereol. 2019;31(3):200–9.
39. Mochida N, Matsumura Y, Kitabatake M, Ito T, Kayano SI, Kikuzaki H. Antioxidant Potential of Non-Extractable Fractions of Dried Persimmon (Diospyros kaki Thunb.) in Streptozotocin–Induced Diabetic Rats. Antioxidants. 2022;11(8).
40. Dieny FF, Fitranti DY, Panunggal B, Safitri I. Pengaruh pemberian sari umbi bit (beta vulgaris) terhadap kadar hemoglobin dan performa atlet sepak bola. J Gizi Indones (The Indones J Nutr. 2017;5(2):119–26.
41. Sivaranjani N, Venkata Rao S, Rajeev G. Role of reactive oxygen species and antioxidants in atopic dermatitis. J Clin Diagnostic Res. 2013;7(12):2683–5.