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Cadmium (Cd) Levels With Kidney Function Examination As An Indication Of Kidney Damage In Petrol Station Operators In North Surabaya with Atomic Absorption Spectrofotometry

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ABSTRACT

Motor vehicle smoke contains several heavy metals that are harmful to humans, the one is cadmium. Petrol station operator workers in direct contact with motor vehicles are at risk of being exposed to the metal cadmium, which can accumulate in the kidneys, leading to decreased kidney function. Decreased kidney function can be seen from the value of creatinine and urea in bloods. This research aims to analyze the relationship between cadmium levels, creatinine and urea values of petrol station operator workers in North Surabaya. This research used correlational method with a cross-sectional design conducted at the Surabaya Health Laboratory and Clinical Laboratory of Health Analyst Department of Health Polytechnic of Ministry of Health Surabaya from November 2020 - June 2021. The sample of this study was 30 gas station operators using purposive sampling technique. Cadmium level examination used Atomic Absorption Spectrophotometry (AAS), then creatinine and urea values were examined using photometry. The results showed that the average cadmium levels 0.32 µg/L; the mean creatinine value 3.2 mg/L and the mean urea value 31 mg/L. From the research result, it can be concluded that there is a relationship between cadmium levels and urea value of gas station operator workers, but there was no relationship between cadmium levels and creatinine values of gas station operators. For further research, may include other variables such as a history of other diseases (diabetes, hypertension, gout) that can affect value of cadmium levels and kidney function examination.
Keywords: cadmium;creatinine;ureum;petrol stations operator;Atomic Absorption Spectrofotometry(AAS).

INTRODUCTION

Air pollution due to vehicle fumes has increased, especially in big cities in Indonesia, one of them is Surabaya. It can be seen from APSI data (air pollution standards index) in August 2019 indicates the index 67 with moderate status and value of PM 2.5 exceeds the quality standards that are harmful air pollutants(1). Exposure to the fumes may result in heavy metal poisoning one of which was cadmium. In North Surabaya region is an area that is quite impassable for trucks and buses and fueling at the gas station, so workers are at risk due to exposure to cadmium poisoning from the fumes.

Cadmium is a heavy metal that is highly toxic to humans. Cadmium into the human body can be through inhalation or from foods and beverages contaminated by cadmium. Cadmium into the body can cause itai-itai disease, which occurs softening of the bones until kidney failure(2). Cadmium into the body will enter the blood circulation and binds to specific metal-binding proteins, namely metallothionein, or circulate freely and combine with molecules, such as albumin, amino acids, sulfhydryl compounds, glutathione and cysteine(3). In all tissues, approximately 80-90% of cadmium will bind to metallothionein proteins to form CdMT, this bond has a strong affinity so it is difficult for most tissues to capture but can be captured by the kidneys(4). So, CdMT bonds will accumulate in the kidney. Cadmium levels in the blood represent recent or ongoing cadmium exposures, while urine cadmium levels indicate total levels of cadmium in the body(5).

Research conducted by O & Edna(6) said that there was an increase in blood cadmium levels in gas station workers when compared to controls, so there was exposure to cadmium at gas stations. Meanwhile, research by Iyanda(7) stated that there was an increase in blood cadmium levels in adolescents who worked less than 6 months is 0.29; less than 20 months is 0.44 and who has worked at a gas station is 0.29 compared to the control is 0.20.

The kidney is the main target organ due to cadmium exposure. Kidney damage may result from exposure by inhalation or ingestion. Chronic cadmium exposure is associated with progressive renal tubular dysfunction. High amounts of cadmium can cause various damage to the body. Low cadmium exposure can cause hepatotoxic effects, chronic renal failure, blood pressure changes, and changes in bone structure. High cadmium exposure can cause itai - itai disease, namely cadmium poisoning, which causes softening of the kidneys and damage to the kidneys, as in postmenopausal women who consume low vitamin D and calcium in Japan[8].

Laboratory tests conducted for the initial screening of the kidneys are the examination of creatinine and urea values in the blood called kidney faal examination. Creatinine and urea are the results of the renal excretion out with urine. Examination of creatinine in blood presents as Glomerulus Filtration Rate (GFR). Meanwhile, urea examination can be used as a diagnosis of acute kidney failure[9].

Research on welding workers conducted by Hernayanti[10] stated there was an increase in cadmium levels of 1.092 ppm compared to control of 0.12 ppm. And an increase in creatinine value of 1.58 mg/dL compared to the control of 0.76 mg/dL. So there is a relationship between increased levels of cadmium in serum with creatinine values in welding workshop workers.

Cadmium levels gas station workers can increase due to fumes around gas stations and can cause damage to the kidneys. Then the purpose of this study was to analyze cadmium levels with the value of kidney function examinations in petrol station workers in the North Surabaya region.

METHODS

This research used a correlation method with a cross-sectional design implemented from November 2020 until June 2021. The cadmium level examination used Atomic Absorption Spectrophotometry (AAS) conducted at Surabaya Health Laboratory. Creatinine value and urea value were examined using photometry conducted at Clinical Laboratory of Health Analyst Department of Health Polytechnic of Ministry of Health Surabaya. The research respondents were 30 petrol station operators whose blood was drawn as research material using the purposive sampling technique. Respondents have criteria including male operators with a minimum term of two years, no kidney disease history, do not smoke, do not drink alcohol, do not take drugs, and willing to become respondents.

This research received a description of ethical exemption from Health Research Ethics Committee of Health Polytechnic of Ministry of Health Surabaya with ethical clearance number No.EA/448/KEPK-Poltekkes_Sby/V/2021 was approved on May 10, 2021.

Sampling

Blood specimens were taken was about 3 mL of venous blood. Placed in a tube containing 2 mL of EDTA anti-coagulant and 1 mL in a plain tube, and given the patient's identity on the tube. Then, carried out the destruction of the EDTA tube for cadmium examination and centrifugation on the plain tube for kidney function examination.

Cadmium Levels Examination

Blood specimens in EDTA tubes were subjected to wet digestion using concentrated HNO3 first to separate metal elements from organic compounds so that they could be read by Atomic Absorption Spectrophotometry (AAS). 10 mL of blood in the Nessler tube was put into the microwave, then added 10 mL of concentrated HNO3 and put back in the microwave for 30 minutes at a temperature of ±130oC and left overnight to disintegrate, which was marked by the solution entirely becomes clear. Then added aquadest free of heavy metals and put it into a volumetric flask and added aquadest up to the mark.

Absorbance measurements were conducted using Atomic Absorption Spectrophotometer at a wavelength of 228,8 nm. Before measuring the absorbance of the sample, be measured standard solution with various concentrations first and the absorbance value of the standard solution was used to create a calibration curve. Then the sample solution can be measured absorbance with a wavelength of 228,8 nm.

Creatinine Levels Examination

Creatinine examination used the Jaffe method without deproteination. The working principle of this test is that creatinine in an alkaline environment will form a red-orange complex with picric acid. The absorbance of this complex is equivalent to the level of creatinine in the sample. Blood in plain tube specimens that have been centrifuged serum taken. Serum was added working reagent and then homogenized. Then read the creatinine value using photometry with a wavelength of 492 nm.

Urea Levels Examination

Urea examination used the colorimetric enzymatic Berthelot method. The working principle of this test is that urease hydrolyzes urea into ammonia and carbon dioxide ions. Modifications Berthelot reaction with ammonium ions reacts with hypochlorite and salicylate form a green color. The intensity of the color formed is equivalent to the level of urea contained in the sample.

Serum in blood specimens that have been centrifuged homogenized with the first reagent and incubated for 5 minutes at 20-25°C. Then the second reagent was added and incubated for 10 minutes at 20-25°C. Then the sample was read using photometry with a wavelength of 578 nm.

Data Analysis

Data obtained by the normality test using Kolmogorov test then correlation test using Pearson correlation (r-Pearson) to determine the relationship of blood cadmium levels to the value of serum creatinine and urea values of petrol station operator workers in North Surabaya.

RESULTS

Based on the research results conducted on the analysis of cadmium levels by examining kidney function in gas station operator workers in the North Surabaya area, the data obtained can be seen in table 1.

In this study, respondents were 30 workers filling station operators in the North Surabaya region with an age range of 21-60 years and working period produce levels of cadmium and kidney examination value varies.

There is levels of cadmium are found in sample code 25 with a working period of 14 years producing cadmium levels of 0.52 g/L and there is levels of cadmium are found in sample code 2 with a working period of 14 years producing cadmium levels of 0.16 g/L. The average value of cadmium content is 0.32 g/L.

Then the creatinine value is found in sample code 16 with 14 years working periods and sample code 22 with 14 working periods produces a creatinine value of 3.7 mg/L, and there is creatinine value is in sample code 8 with 5 years working periods produces a creatinine value of 2.4 mg/L. The mean value of creatinine value is 3.2 mg/L

On the urea value, the value is found in sample code 6 with a working period of 6 years produces a urea value of 41 mg/L and there is the value of urea is found in sample code 8 with a working period of 5 years produces a urea value of 23 mg/L. The mean value of urea value is 31 mg/L.

Table 1. Data analysis of cadmium levels with kidney function examination at gas station operators

No	Sample Code	Age (years)	Working period (years)	Cadmium Levels (µg/L)	Cretinine Levels (mg/L)	Urea Levels (mg/L)
1	1	40	14	0,23	3,2	25
2	2	40	14	**0,16**	3,1	32
3	3	39	13	0,27	3,3	35
4	4	33	14	0,42	3,6	30
5	5	29	12	0,25	3,4	28
6	6	50	6	0,19	3,3	**41**
7	7	38	14	0,24	3,1	36
8	8	32	5	0,33	**2,4**	**23**
9	9	51	14	0,28	3,1	34
10	10	41	14	0,48	3,4	31
11	11	35	14	0,22	3,1	37
12	12	23	4	0,37	3,2	26
13	13	40	12	0,26	3,4	27
14	14	41	14	0,21	3,5	32
15	15	39	14	0,32	3,2	34
16	16	32	14	0,48	**3,7**	29
17	17	30	13	0,28	3,2	24
18	18	49	5	0,22	3,6	38
19	19	37	12	0,29	3,1	29
20	20	38	14	0,37	3,1	32
21	21	50	13	0,24	3,4	37
22	22	41	14	0,43	**3,7**	34
23	23	35	14	0,28	3,2	34

No	Sample Code	Age (years)	Working period (years)	Cadmium Levels (µg/L)	Cretinine Levels (mg/L)	Urea Levels (mg/L)
24	24	21	5	0,34	3,0	27
25	25	53	15	**0,52**	2,5	29
26	26	33	8	0,41	2,9	31
27	27	26	5	0,38	2,6	28
28	28	28	3	0,29	2,9	31
29	29	24	7	0,46	2,6	27
30	30	29	12	0,51	2,6	24
Total				**9,73**	**94,4**	**925**
Average values				**0,32**	**3,15**	**30,8**

Table 2. Characteristics of gas station workers with cadmium levels above the cadmium level threshold

No	Sample Code	Age	Working Periods	Cadmium Levels
1	4	33 years old	14 years	0,42 µg/L
2	10	41 years old	14 years	0,48 µg/L
3	16	32 years old	14 years	0,48 µg/L
4	22	41 years old	14 years	0,43 µg/L
5	25	53 years old	15 years	0,52 µg/L
6	26	33 years old	8 years	0,41 µg/L
7	27	26 years old	5 years	0,38 µg/L
8	29	24 years old	7 years	0,46 µg/L
9	30	29 years old	12 years	0,51 µg/L

There are data on operator workers as many as 9 people with cadmium levels that exceed the threshold of 0.38 g/L(11). There is level of cadmium is found in sample code 25, which is 53 years old with a working period of 15 years, which is 0.52 g/L.

Based on age, sample code 29 is the youngest age at 24 years old with cadmium level of 0.46 g/L and sample code 25 is the oldest age at 53 years old with cadmium level of 0.52 g/L.

Then according to the working period, sample code 25 has the most extended working period of 15 years with a cadmium level of 0.52 g/L, while sample code 27 has the shortest working period of 5 years with cadmium level of 0.38 g/L.

DISCUSSION

The research was conducted at a gas station in the North Surabaya region with 30 respondents as operator workers at the gas station. Respondents obtained according to the sample criteria are male, have worked for more than two years, use PPE when working, in good health, do not smoke and do not take drugs, and have no history of kidney disease. History of diseases into the criteria only kidney disease, other diseases such as diabetes and hypertension that can affect kidney function are not included as sample criteria. Then there is no initial screening to checking the values of uric acid, blood sugar, and cholesterol using a test strip. These three values can also affect kidney function that may affect the values of creatinine and urea.

Age is one of the variables in analyzing cadmium levels in gas station operator workers. Generally, older people are more sensitive to cadmium activity in the body than young people because the activity of the biotransformase enzyme is reduced and the resistance of certain organs is reduced to the effects of cadmium. However, at a young age, high levels of cadmium can also be detected in the body because cadmium levels can come from foods consumed that contain cadmium, such as vegetables, rice, marine fish, and other foods contaminated with cadmium metal[12]. This study did not show a significant relationship between age and cadmium levels. This may be caused several supporting factors such as lifestyle, consumption of food and drinks, and different outdoor activities. This study explains that there is no significant relationship between cadmium levels and the working period. This study is similar to research conducted by Ayuda[8] which explains that there is no significant relationship between groups of the duration of exposure to cadmium less than ten years and more than ten years on the difference between the duration of work and cadmium levels in cadmium parking workers. A study conducted by Ghazali[13]

showed no significant relationship between years of service and levels of cadmium. However, there was a significant relationship between years of service and levels of lead and arsenic.

Cadmium levels in this study showed results that exceeded the threshold only 9 out of 30 respondents so was no significant increase in cadmium levels with increasing age and working period. This is not following the study conducted by Iyanda[7], which explained an increase in cadmium according to the length of exposure to cadmium in gas station workers. Cadmium in the blood is a biomarker of recent exposure and engages with acute symptoms. This study respondents, 70% of operator workers have worked for more than ten years resulting in cadmium in the body has accumulated high in the target organs, which can interfere with the function of these organs. In addition, according to a field survey conducted during sampling, operator workers have been wearing Personal Protective Equipment (PPE) in the form of masks regularly in the past year due to the Covid-19 pandemic. So may reduce the level of cadmium metal exposure from vehicle fumes that into through inhalation.

The creatinine value in all respondents in this study showed a value above normal (0.7 – 1.3 mg/L). The creatinine values increased may occur due to cadmium in the body which accumulates in kidneys by 30% and in the liver by 30%. Cadmium in the body will bind to metallothionein proteins in the liver and kidneys, leading to increased free radicals in the body. This can cause a chain reaction of lipid peroxidation leading to decreased Glomerulus Filtration Rate (GFR), which increased blood creatinine values may mark. Creatinine is a product of creatine and phosphocreatine metabolism which is filtered and reabsorbed in the renal tubules. If there is damage to the kidneys and a decrease in GFR, the ability of the kidneys to filter creatinine will decrease that creatinine levels in urine will decrease may caused creatinine levels in serum to increase[14]. Research conducted by Yakubu[15] stated that a significant increase in creatinine and urea values in gas station operators compared to controls. There was an increase in creatinine value in all respondents in this study, but it did not significantly affect age and years of service. An increase in blood cadmium levels does not accompany increased creatinine value. This can occur because cadmium into the body accumulates in the kidney, leading to impaired renal function. In addition to the cadmium accumulates in the kidneys, increased creatinine values may occur due to an increase in muscle mass of the body, which in muscle mass associated with the level of protein intake.

The urea value in operator workers who became respondents showed 26 out of 30 respondents had urea levels exceeded the normal value (7-25 mg/L). The value of urea in serum reflects the balance of production and excretion. The value of urea is obtained by measuring the value of nitrogen in urea. Nitrogen in urea has a small but stable value. Urea is a waste product of protein metabolism excreted by the kidneys through urine. If the function of kidneys is damaged, the kidneys are not able to excrete urea, which can make urea circulate in the blood, resulting in the blood urea value becoming high. Heavy metals that enter the body will accumulate in the kidneys and cause damage to the nephrons, especially the tubular epithelial cells. This is marked by the Glomerular Filtration Rate (GFR) decreased, which can cause metabolic waste products such as urea that should be removed, resulting in a decrease in the value of urea in the urine and an increase in the blood. The effects of heavy metal exposure can cause cells to be unable to maintain their homeostasis so that certain types of cellular proteins are damaged and tissue apoptosis will increase. Cadmium can cause damage to cell membrane function by damaging the lipid composition of cell membranes. In an experiment conducted by Fadhilla[16] on male rats exposed to cadmium, there was a significant increase in blood urea and creatinine values. In this study, urea value increased, but have not a significant relationship with age and years of service. Increased levels of urea can be caused by several factors other than exposure to heavy metals, such as a history of diabetes, high blood pressure, lifestyle, body obesity level, and body mass index.

The results of statistical tests using Pearson's correlation on cadmium levels with creatinine values shows a p-value is higher than α value (0.05), this indicates that there is no significant correlation between cadmium levels and creatinine values. This is not comparable to the research conducted by Hernayanti(10) that there is a significant relationship between cadmium levels and creatinine values in welding workshop workers. However, it is comparable to Sugiharto[17] research that there is no significant relationship between cadmium levels and creatinine values in exhaust welding workers.

Statistical results of the Pearson test of cadmium levels with urea values showed p-value is lower than the value of α (0.05), which means that there is a significant relationship between cadmium levels and urea values in gas station operator workers. This is comparable to a study conducted by Yakubu[13] which stated that the increase in cadmium values in gas station workers was proportional to the increase in blood urea and creatinine values. From this research, it can be concluded that blood cadmium levels in gas station operator workers are not related to blood creatinine values but are related to blood urea values. The relationship of cadmium levels that do not exceed the maximum threshold in the body with serum urea values that exceed average values can occur because cadmium levels in the body have accumulated in the target organ is the kidneys. The kidneys have decreased function in excreting urea which should be excreted through urine but remains in the blood circulation, causing the blood urea value to increase.

CONCLUSION

Based on this research can be concluded that there is no relationship between blood cadmium levels and blood creatinine values. However, there is a relationship between blood cadmium levels and blood urea values of gas station workers in the North Surabaya region, with an average value of cadmium levels of 0.32 g/L; the mean value of creatinine value is 3.2 mg/L, and the mean value of urea is 31 mg/L.

There is a suggestion for gas station workers to be more alert to vehicle fumes around gas station by using PPE while working. And for further research, may include other variables such as a history of other diseases (diabetes, hypertension, gout) that can affect cadmium levels and kidney function examination.

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