Cholinesterase Level, Hemoglobin and Blood Pressure among Vegetable Farm Workers Garuda Sakti — A Cross-Sectional Study

Hartini H*, Gela Saputri, Midun Gabriel

Medical Laboratory Technology of Akademi Kesehatan John Paul II Pekanbaru

Corresponding author: hartini.h@akjp2.ac.id

Abstract . Pesticides are chemical compounds used by farmers to eradicate pests in plants. Inappropriate use of pesticides can cause poisoning which can be detected by cholinesterase level. The purpose of this study was to determine the relationship between cholinesterase levels and blood pressure in vegetable farmers on Garuda Sakti, Kampar, Riau. The research methods were spectrophotometry and sphygmomanometer. A total of 7 samples obtained cholinesterase levels in the range > 1400 - 3500 U/L indicating mild poisoning. Cholinesterase levels in 2 samples ranged from 4,185.402 to 5,380.075 U/L, which mean normal. Sample 1 obtained cholinesterase levels in the range of 701 - 1,400 U/L indicating moderate poisoning. The results of the hemoglobin levels for those who were poisoned were mostly normal. The results of blood pressure measurement of farmers who were poisoned were predominantly normal. Future study is expected to link cholinesterase levels with farmer's leukocytes.

Keywords: Blood Pressure, Cholinesterase, Farmers, Hemoglobin, Pesticides

1 BACKGROUND

Pesticides or better known as pest repellents are chemical compounds commonly used by farmers to control or eradicate pest organisms (pests) on plants so that agricultural yields will increase (Sari et al., 2018). Farmers usually use pesticides without following the recommended usage rules. Farmers tend to use the cover blanket method, that is, the plants are still sprayed with pesticides even though the pests are not present. Farmers think that the more frequently they are sprayed with pesticides, the higher their yields will be because pests can be controlled (Nejatifar et al., 2022). According to

WHO (2006), 12,000 farmers die from pesticide poisoning per year in Indonesia (Ipmawati et al., 2016).

Factors related to pesticide poisoning are pesticide dose, work period as a sprayer, duration of spraying, frequency of spraying, and use of Personal Protective Equipment (PPE) (Hardi et al., 2020). Improper use of pesticides causes negative impacts on humans in the form of both acute and chronic poisoning (Alewu, B and Nosiri, C, 2011) . Acute poisoning caused by dizziness, nausea, vomiting, convulsions, fainting and even death (Goel, A and Aggarwal, P, 2007). Chronic poisoning is difficult to identify because the impact is not felt immediately, but in the long term it can cause cancer, mutagenic (genetic damage that causes chromosome changes), teratogenic (birth of disabled children) (Marisa & Pratuna, 2018). Apart from causing poisoning, it can also cause nervous disorders, liver disorders, hormonal disorders and increased blood pressure (S. Nurkhayati, N. Nurjazuli, 2018).

Exposure to pesticides can occur through breathing, mouth and skin (Nuryati et al., 2022). Pesticide poisoning can be identified through cholinesterase examination. Cholinesterase levels are influenced by organophosphate and carbamate pesticides (Hardi et al., 2020). The active substance of the pesticide is anti-cholinesterase (inhibits the work of the cholinesterase enzyme) which causes acetylcholine to not be broken down, resulting in a buildup of acetylcholine (Wafa et al., 2019).

Exposure to pesticides can also affect blood pressure which begins with pesticides entering the body through the skin, being swallowed by the mouth, or inhaled by the nose (Fajriani, Aeni, & Sriwiguna, 2019). When the active pesticide content enters the body, it can interfere with the process of breaking down acetylcholine (Agustina et al., 2018). Acetylcholine will bind to its receptors, namely muscarinic and nicotinic in the peripheral and central nervous system, causing disruption of the activity of the cholinesterase enzyme in nerve tissues and cells. The buildup of acetylcholine causes irregular blood flow in the blood vessels (Septiana et al., 2021). Blood flow in the blood vessels can be slower or faster, causing high blood pressure (hypertension) or low blood pressure (hypotension) (Agustina et al., 2018). Based on this background, researchers are interested in conducting research on cholinesterase level, hemoglobin and blood pressure in vagetable farm worker.

2 RESEARCH METHODS

The type of research is descriptive observational using a cross-sectional design. Sample in this study were 10 farmers of vegetable farmer in Garuda Sakti, Tapung, Kampar. The inclusion criteria in this study were male and female farmers, aged 20-60 years. This research was conducted at Clinical Chemistry Laboratory, Akademi Kesehatan John Paul II Pekanbaru and SMK Abdurrab. This study used centrifuge, vacutainer tubes without anticoagulants, syringes, tourniquets, 70% alcohol cotton, pasteur pipettes, test tubes, sterile gauze, Optiva Spectrophotometer, sfigmomanometer, stethoscope, cuvettes, 10-100 µL micropipette, 100-1000 µL micropipette, blue tips, yellow tips. The materials used in this study were reagent 1 (phosphate buffer), reagent

2 (butyrylthiocholine substrate), reagent 3 (dibucaine chlorhidrate), distilled water, and serum samples.

Blood pressure measurements. Systolic and diastolic blood pressure were measured with calibrated sphygmomanometer, in the right arm, following protocols recommended by the Blain (2011). Blood pressure measurements were taken after 10 min of rest. The subject was in a sitting position with antecubital fossa as high as the heart, with uncrossed legs. The arm should be supported at the level of the heart, resting on a cushion, pillow or arm rest. Ensure no tight clothing constricts the arm. Place the cuff on neatly 2cm above the brachial artery and aligning the 'artery mark'. The bladder should encircle at least 80% of the arm but not more than 100%. Use the cuff size recommended by the manufacturer of the monitor. Estimate the systolic beforehand: Palpate the brachial artery o Inflate cuff until pulsation disappears o Deflate cuff o Estimate systolic pressure. Then inflate to 30mmHg above the estimated systolic level to occlude the pulse. Place the stethoscope diaphragm over the brachial artery and deflate at a rate of 2-3mm/sec until you hear regular tapping sounds. Measure systolic (first sound) and diastolic (disappearance) to nearest 2 mmHg (Ibrahim et al., 2021).

Blood sampling followed the procedure of Warekois & Robinson (2016). The patient is asked to sit or lie down with the arms extended. The tourniquet is placed on the patient's arm ± 10 cm above the elbow crease. The patient is asked to make a fist and disinfect it with 70% alcohol cotton. Venous blood vessels with an angle of 15º using a syringe. Blood was taken as much as 3 mL. Blood is transferred into a vacutainer tube and homogenized.

Serum separation was carried out by Lestari et al., (2019). The vacutainer tube containing blood was centrifuged at 3000 rpm for 10 minutes. Serum and red blood cells were separated using a Pasteur pipette. Serum is put into a test tube.

Measurement of cholinesterase levels. Pipette 2700 µL of reagent 1 then put it in a test tube and add 300 µL of reagent 3 and homogenize as control reagent. Negative control reagen was prepare by pipette 1.5 mL of the control reagent and then put it into a test tube, add 50 µL of reagent 2 and homogenize. Pipette 10 µL of control serum then put it in a test tube and homogenize. Sample test by pipette 1.5 mL of Reagent 1 then put it in a test tube add 50 µL of reagent 2 and homogenize. Pipette 10 µL of farmer's serum then put it in a test tube and homogenize. Select Performance Test then press to select the type of inspection. Select Water then press to suck distilled water. Select Blank then press to suck the blank and wait for 3 seconds until the blank results come out. Select Sample then press to suck the sample and wait 2 minutes until the test results come out.

Hemoglobin test. Prepare the test tubes first and then label them. Then pipette 2500 µL of Drabkin solution. Pipette a 10 µL blood sample then homogenize it. Then incubate the sample in the tool for 5 minutes. Select "Performance Test" on the spectrophotometer then press to select the type of examination. Select Performance Test then press to select the type of inspection. Select Water then press to suck distilled water. Select Blank then press to suck the blank and wait for 3 seconds until the blank results come out. Select Sample then press to suck the sample and wait 2 minutes the results will come directly from the spectrophotometer.

Statistical analysis. Mean, standard deviation, frequency, and percentage were used in descriptive statistics, the Shapiro-Wilk test to check the data for normality.

3 RESULTS AND DISCUSSION

3.1 RESULTS

Characteristics of the participants. In this study, 10 vegetable farm workers participated. Of these, 3 were female and the 7 male vegetable farm workers. The mean of years working at the current vegetable farm was over 5 years, and there was no difference in the number of working months when comparing male and female workers or when comparing the two male and female study groups (Table 1). The mean working hours per day for the study groups was over 2 hours. The mean of the frequency of application was two times per week. Most of them used incomplete personal protective equipment.

Table 1. Characteristic of the participants

Variable		**f (person)**	**%**
Gender	Male	7	70
	Female	3	30
Work experience	> 5 years	10	100
	< 5 years	0	0
Spraying frequency	≤ 2 times a week	0	0
	> 2 times a week	10	100
Duration of work	≤ 2 hours	0	0
	> 2 hours	10	100
Direction	Match with the direction of a wind	10	100
	Not match with the label	0	0
Personal protective equipment	Wear a mask	0	0
	Not wear a mask	10	100
	Wear gloves	2	10
	Not wear gloves	8	80
	Wear a hat	8	80
	Not wear hat	2	20

The overall prevalence of abnormal Cholinesterase (less than 3.500 IU/L) was 8 farmers (80%) with mean 4.782,82 ± 844,8IU/L. Mild poisoning occurs to 7 farmers and moderate poisoning to 1 farmer with mean cholinesterase level 2.608,31 ± 491,3 IU/L and 739,321 ± 0 IU/L respectively (Table 2). Based on hemoglobin levels, it shows that 60% (6 farmer) had normal hemoglobin level, 20% (2 farmer) had polycythemia and 20% (2 farmer) had anemia. Based on blood pressure, it shows that 40% (4 farmer) had normal blood pressure, 30% (3 farmer) had pre-hypertension and 30% (3 farmer) had hypertension.

Table 2. Serum Cholinesterase, hemoglobin concentration and blood pressure

Variable		Mean ± SD	f (per-son)	%
Cholinesterase level	Normal (>3.500 IU/L)	4.782,82 ± 844,8	2	20
	Mild poisoning (>1.401-3.500 IU/L)	2.608,31 ± 491,3	7	70
	Moderate poisoning (701-1.400 IU/L)	739,321 ± 0	1	10
	Severe poisoning (< 700 IU/L)	0	0	0
Hemoglobin	Normal	14,4915 ± 1,2	6	60
Male (14-16 g/dL)	Polycythemia	19,275 ± 1,8	2	20
Female (12-14 g/dL)	Anemia	12,112 ± 0,8	2	20
Blood pressure	Normal		4	40
	Pre-hypertension		3	30
	Hypertension		3	30

4 DISCUSSION

Cholinesterase is an enzyme that hydrolyzes acetylcholine to produce acetic acid and choline (Rahayu & Sobayar, 2018). Based on the results of examining cholinesterase levels from 10 samples, 7 samples had mild poisoning and 1 sample had moderate poisoning. Based on questionnaire data, farmers who experienced poisoning did not use complete PPE. The reason given by farmers for not using complete PPE is because they feel uncomfortable when spraying. This is in line with research conducted by Fajriani et al., (2019) which states that incomplete use of PPE when spraying will result in a higher risk of farmers being exposed to pesticides. The aim of using PPE is to protect farmers from exposure to pesticides such as masks, gloves, head protectors, glasses and shoes (Darmiati, 2020). Farmers who experience poisoning should pay attention to the use of PPE when spraying pesticides to prevent poisoning from occurring to a more severe level (Damalas, CA and Koutroubas, SD, 2016)

Factors that can cause farmers to experience moderate poisoning apart from the use of PPE are work periods. Research conducted by Hardi et al., (2020) shows that there is a relationship between work period and frequency of pesticide spraying with cholinesterase levels in farmers. Farmers who experience moderate poisoning should not be exposed to pesticides for approximately 2 weeks so that cholinesterase activity returns to normal and does not progress to the level of severe poisoning.

Another factor that can cause a decrease in cholinesterase levels in the body is exposure to organophosphate pesticides (Jintana, S et al, 2019). The active pesticide substance that enters the body disrupts the breakdown of acetylcholine. Cholinesterase which should bind to acetylcholine will bind to the active substance of the pesticide (Agustina et al., 2018). The sulfate content in pesticides will form sulfhemoglobin bonds so that hemoglobin becomes abnormal and its function is disrupted in delivering oxygen (Purba, IG, 2011). The binding of sulfhemoglobin in red blood cells causes a decrease in hemoglobin levels, causing anemia (Marisa & Pratuna, 2018)

Based on table 2, 40% (4 farmer) had normal blood pressure, 30% (3 farmer) had pre-hypertension and 30% (3 farmer) had hypertension. Most of the blood pressure results obtained by farmers who experienced poisoning were normal. Factors that can influence blood pressure in farmers can be caused by genetics, age, gender, diet and physical activity (Naryati & Priyono, 2022).

5 CONCLUSION AND RECOMMENDATION

According to characteristic of farmers and pesticide exposure, 1 sample (10%) showed that cholinesterase levels were moderately poisoned, 7 samples (70%) showed cholinesterase levels that were mildly poisoned, and 2 samples (20%) had cholinesterase levels in the normal range, which means the farmers did not experience poisoning. Hemoglobin levels from 10 farmers were obtained, 6 farmers (60%) showed normal hemoglobin levels, 2 farmers showed polycythemia and 2 farmers (20%) showed anemia.

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