

Acetylcholinesterase levels in farmers exposed to pesticides: The prevalence and associated factor

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Acetylcholinesterase levels in farmers exposed to pesticides: The prevalence and associated factor

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Abstract

Background: The farmers' habit of pesticide management is an important factor in recovering the farmer's health from contamination. Most farmers have poor knowledge of pesticide safety management. **Aims:** This study aimed at analyzing factors related to the blood Acetylcholinesterase level as an indicator of pesticide contamination in the body. **Settings and Design:** This research was a cross-sectional study involved 57 people of rice and horticulture farmers in Ponorogo. The independent variables were age, working years as a farmers, educational background, activity, combination of pesticides, read the direction, storage and mixing, get information about pesticide safety management, use of PPE, smoking habit, mixing anorganic and organic, and personal hygiene. The dependent variable was blood acetylcholinesterase level. **Methods and Material:** The independent variables were observed by questionnaire and the dependent variable was measured with a photometer by the cobast method. The pesticide levels divided into four categories: (1) Very high risk of poisoning: 5-6 U/L; (2) High risk of poisoning: 7-8 U/L; (3) Medium risk of poisoning: 9-10 U/L, and (4) Low risk of poisoning : 11-12 U/L. **Statistical analysis used:** The data were analyzed by multiple linear regressions. The correlation within variables are analysed using Pearson Correlation test with 95 % of Confidence Interval (CI). The Statistical Package for Social Sciences (SPSS) 24 software were used in this research. **Results:** There were 6 farmers at very high risk, 17 farmers at high risk, 26 farmers at medium risk, and 8 farmers at low risk of pesticide contamination. The independent variable contributed 32.2% (R Square = 0.322) to the blood acetylcholinesterase level. The influencing factor was combination of pesticide (p-value = 0.02). **Conclusions:** Combination of

pesticide was the crucial factors in pesticide safety management. Farmers should do pesticide safety management properly start from storage, mixing, handling, spraying, and disposing of.

Keywords: Acetylcholinesterase, farmer, pesticide, associate factor

Key Messages:

The use of pesticides in combination was simultaneously affecting acetylcholinesterase levels in the blood. Farmers use pesticides based on hereditary experience can increasing pesticide contamination. Pesticide safety management were key factors in preventing pesticide exposure.

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Introduction

Indonesia as an agrarian country, most residents have a livelihood as a farmer. This is in accordance with data from the Indonesian Central Bureau of Statistics in 2019 which stated that the profession of farmers in Indonesia amounts to 38,109,196 or 29.46% of the total population (1). According to the Indonesian Central Bureau of Statistics in 2018, agricultural land in Indonesia including non-rice fields is 27,724,917 ha and rice fields were 7,105,145 Ha (2). Agricultural activities are very closely related to one of the chemicals called pesticides. Pesticides are substances used by farmers to eradicate pests and diseases of crops (3)(4).

Demangan Village, located in Siman Sub-District, Ponorogo District, has a majority of the population as farmers. Agricultural activities in the form of cultivation of rice, fruit, soybeans, and vegetables including chili and shallots. To maintain crop productivity, farmers use anorganic pesticides in hereditary and experience-based. Farmers realize that anorganic pesticides are more potent at eradicating pests than the organic one. Most of them have been farmers since their teens age. This condition needs attention, due to pesticides harm health especially in unsafe management (5)(6). Inappropriate use of anorganic pesticides in addition to being harmful to health can also be harmful to the environment especially springs and pesticide residues in agricultural products (6)(7)(8).

Most of farmers are chronically exposed to pesticides and it gets into the body through the skin. It can contaminates through breathing (4). The way pesticides work is to inhibit the enzyme acetylcholinesterase in the body so that it can disrupt the nervous system (5)(9). Inhibition of acetylcholinesterase becomes a biomarker in pesticide exposure (27). Symptoms of acute pesticide poisoning include irritation of the eyes, excessive fatigue and saliva, difficulty breathing, itching of the skin, cough, unclear vision, dizziness and burning of the tongue, throat and esophagus (10)(11).

In Demangan Village, farmers use pesticides according to experience; without looking at labels, considering the effects of toxicity, the use of personal protective equipment, as well as mixing pesticides. Farmers only use clothes that are tinged with the mouth and nose in place of a mask. It can increase pesticide contamination in the body of farmers (7)(11). Researchers also obtained data that 96% of farmers have poor knowledge of the dangers of pesticides in the body. Knowledge is one of the factors that supports a person's behavior (12).

Previous research related to this study was a study of serum acetylcholinesterase levels in farmers in Thailand who took samples with finger blood tests and categorized them as unsafe and risky (13). Another similar study was analyzing of acetylcholinesterase levels in farmers exposed to pesticides in Malaysia using spectrophotometry with normal levels of pesticides of 3500 U/l (14). The difference with this study is the method used to analyze the level of acetylcholinesterase in the blood of farmers, namely by cobas method and categorization of pesticide levels in the blood. Furthermore, this study investigate the information on the characteristics of farmers with high levels of acetylcholinesterase by categorizing farmers on the use of anorganic, organic, and mixed pesticides.

The purpose of this study is to analyze acetylcholinesterase levels in farmers and the related factors. This research is expected to provide real data on the health condition of farmers related to pesticide use: thus, farmers realize about the importance of working safely and healthily.

Subjects and Methods

This research was a cross-sectional study involved 57 people of rice and horticulture farmers which was a combination of farmer groups in the Ponorogo District. The sampling was random-based, farmers willing to be the subject must have filled the informed content. The independent variables consisted of age, working years as a farmers, educational background, activity, combination of pesticides, read the direction, storage and mixing, get information about pesticide safety management, use of PPE, smoking habit, mixing anorganic and organic, and personal hygiene. The independent variables were observed by questionnaire and semi-structured interviews. The dependent variable was the blood acetylcholinesterase level. That dependent variable was measured with a photometer by the cobas method. Acetylcholinesterase levels in the blood were categorized as follows:

1. Very high risk of poisoning : 5-6 U/L
2. High risk of poisoning : 7-8 U/L
3. Medium risk of poisoning : 9-10 U/L
4. Low risk of poisoning : 11-12 U/L

The data were analyzed using multiple linear regressions to determine the relationship of independent variables with acetylcholinesterase levels in the farmer's blood with the formula $Y = a + b_1 X_1 + b_2 X_2 + \dots + b_n X_n$.

This research had been approved from the ethics commission of Dr. Harjono Hospital of Ponorogo with registered number: 3502021K121442020082100002/IX/KEPK/2020.

Results

The following are the demographic characteristics of farmers in Demangan Village:

Table 1: Demographic characteristics of farmers

Demographic	Frequency	Percentage (%)
Age		
< 40	7	12,28
≥ 40	50	87,72
Working years as farmer		
< 10	5	8,77
≥ 10	52	91,23
Educational Background		
Illiterate	4	7,02
Elementary School	23	40,35
Junior High School	12	21,05
Senior High School	16	28,07
Bachelor Degree	2	3,51
Activity		
Horticulture	5	8,77
Rice	19	33,33
Horticulture and Rice	33	57,89
Combination of Pesticide		
Use 1 kind of Pesticide	4	7,02
Use 2 kind of Pesticide	15	26,32
Use 3 kind of Pesticide	15	26,32
Use 4 kind of Pesticide	14	24,56
Use 5 kind of Pesticide	9	15,79
Read the Direction		
Yes	28	49,12
No	29	50,88
Storage and Mixing		
Home	25	43,86
Land	32	56,14

Get Information About Pesticide Safety Management

Yes	18	31,58
No	39	68,42

Use of PPE

Yes	14	24,56
No	43	75,44

Smoking Habit

Yes	40	70,18
No	17	29,82

Mixing Anorganik and Organic Pesticide

Yes	30	52,63
No (Anorganic Only)	27	47,37

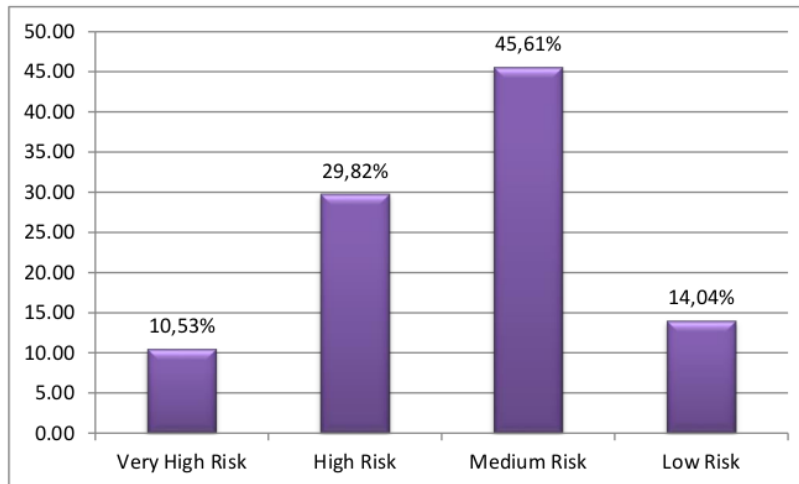
Personal Hygiene

Good	4	7,02
Poor	53	92,98

Based on the table 1 above, the obtained data of farmers in 87.72% were over 40 years old, 91.23% had been farmers over 10 years, 40.35% were elementary school, 57.89% were rice and horticultural farmers, only 7.02% of farmers did not mix pesticides, farmers who read the label before using pesticides as much as 49.12%. There were 43.86% of farmers still storing pesticides at home, 68.42% of farmers did not get information and socialization yet about safe pesticide management, 75.44% of farmers did not use personal protective equipment at all, 70.18% of farmers do smoking habits, 52.63% of farmers use anorganic mixing and organic pesticides, and 92.98% of farmers had poor personal hygiene.

The results of pesticide examination in the farmer's blood are as follows:

Figure 1: Pesticide Levels in Farmers' Blood



Based on figure 1, there were 10.53% of farmers at very high risk, 29.82% at high risk, 45.61% at medium risk and 14.04% at low risk. The following were the results of statistical analysis and cross-tabulation of the demographic characteristics of farmers with acetylcholinesterase levels:

Table 2: Analysis results of statistic and cross-tabulation of demographic characteristics with pesticide levels in farmers' blood

Demographic	Category				Total	p value	Sig.	R	R Square
	Very High Risk	High Risk	Medium Risk	Low Risk					
Age									
< 40	2	1	4	0	7	0,632			
≥ 40	4	16	22	8	50				
Working years as farmer									
< 10	0	1	2	1	4	0,906			
≥ 10	5	16	25	7	53				
Educational Background									
Illiterate	0	1	1	2	4	0,685			
Elementary School	3	8	8	3	23				
Junior High School	3	2	5	2	12				
Senior High School	0	6	11	0	16				
Bachelor Degree	0	0	1	1	2				
Activity									
Horticulture	0	1	4	0	5	0,252			
Rice	1	5	10	3	19				
Horticulture and Rice	5	11	12	5	33		0,086	0,580	0,336
Combination of pesticide									
Use 1 kind of Pesticide	0	1	2	1	4	0,02			
Use 2 kind of Pesticide	1	3	9	4	17				
Use 3 kind of Pesticide	2	4	6	1	13				
Use 4 kind of Pesticide	2	4	6	2	14				
Use 5 kind of Pesticide	1	6	2	0	9				
Read the Direction									
Yes	0	10	8	8	28	0,124			
No	4	12	7	8	29				
Storage and Mixing									
Home	2	8	10	7	27	0,52			
Land	4	9	16	1	30				
Get Information About Pesticide Safety Management									
Yes	0	8	8	3	19	0,457			
No	6	9	18	5	38				

Use of PPE						
Yes	1	2	7	4	14	0,09
No	5	15	19	4	43	
Smoking Habit						
Yes	5	13	16	6	40	0,995
No	1	4	10	2	17	
Mixing Anorganik and Organic Pesticide						
Yes	2	10	15	3	30	0,552
No	4	7	11	5	27	
Personal Hygiene						
Good	0	0	2	2	4	0,09
Poor	6	17	24	6	53	

Based on table 2, it was identified that there was no relationship between age (0.086), working years as a farmer (0.906), educational background (0.685), activity or type of agriculture (0.252), read the direction (0.124), storage and mixing (0.252),0.52), got information about pesticide safety management (0.457), used of PPE (0.09), smoking habit (0.995), mixing anorganic and organic pesticide (0.552) and personal hygiene (0.09) with pesticide levels in the blood. The combination of pesticides had a significant relationship with the levels of pesticides in the blood with a p value = 0.02 (< 0.05).

Based on table 2, it was identified that the value of R was 0.580 which meant that between variables had a moderate relationship. R Square was 0.336 which means variable levels of pesticides in the blood could be explained by 12 independent variables simultaneously or simultaneously by 33.6% while others (66.4%) are described by other unexamined variables. The multiple linear regression equations are as follows:

$$Y = a + b_1 X_1 + b_2 X_2 + b_3 X_3 + b_4 X_4 + b_5 X_5 + b_6 X_6 + b_7 X_7 + b_8 X_8 + b_9 X_9 + b_{10} X_{10} + b_{11} X_{11} + b_{12} X_{12}$$

$$Y = 6,16 + 0,055 X_1 + 0,208 X_2 + 0,055 X_3 + -0,221 X_4 + -0,253 X_5 + 0,413 X_6 + -0,198 X_7 + -0,194 X_8 + -0,511 X_9 + -0,903 X_{10} + -0,157 X_{11} + 0,002 X_{12}$$

Discussion

In the present study, a related factor to acetylcholinesterase levels in the blood was the use of a combination of pesticides. This was observed in the data which was only 7.02% of farmers who use one type of pesticide, while 92.98% mix 2 to 4 types of pesticides. Farmers mixed pesticides to easily eradicate pests and increase pesticide kill power against pests and crop diseases. Pesticide mixing was done to increase the quantity of pesticide active ingredients, the frequency of spraying, the

effectiveness of spraying time, and save the cost of spraying pesticides, this is certainly harmful to the health of farmers and the environment (15)(16)(26)(31).

Plant pests were already resistant to pesticides so spraying one type of pesticide was less effective in pest control. Mixing 2 types of fungicides and 3 types of insecticides that had different active ingredients was able to increase the yield of shallots and decrease the attack of anthracnose and *spodopteraexigua* (30). Fungicides and insecticides could be mixed in spraying but with different active ingredients. Knowledge of pesticide toxin power classification of some active ingredients of agricultural pesticides should be conveyed to farmers to avoid inappropriate mixing of some types of pesticides. Mixing different types of pesticides with high toxin power greatly harmed the health of farmers, as it increased the risk of pesticide contamination in the blood. The combined properties of chemicals were very harmful to the body, especially additive and synergistic parts (15)(20).

According to previous research, chronic exposure to pesticides was found in farmers who mix pesticides with marked inhibitions of acetylcholinesterase enzyme activity (15)(16). It was also identified in the equation of multiple linear regressions with negative coefficients which means that every increase in the number of pesticides was mixed, then the level of acetylcholinesterase in the blood decreased, so there was inhibition of acetylcholinesterase. This was due to farmers who do not yet understand the additive and synergistic effects of chemical mixtures (15). Based on the cross-tabulation results in table 2, known that farmers who were at very high risk are farmers who mix as much as 2 even more types of pesticides. The effect of pesticide toxicity on the human body depended on the active ingredient contained (16). In table 2, some farmers mixed 2, 3, and 4 types of pesticides but they were at low risk. This was due to farmers in rice farming do less spraying than horticulture. While horticultural farmers who mixed 2, 3, 4 types of pesticides found out 2 people at low risk. This was due to the age of farmers categorized still under 40 years with a working period of 10 years.

The characteristics of the workforce that had 4 people in very high risk were illiterate, 66.67% aged ≥ 40 years and 33.33% < 40 years, had become farmers ≥ 10 years, had a smoking habit, was a farmer who has an activity in rice farming as well as horticulture, mixing two even more types of pesticides, not reading pesticide labels, 75% had never participated socialization related to pesticide management and 25% had participated socialization related to pesticide management, 75% store pesticides at home and 25% in rice fields, did not use personal protective equipment, farmers used anorganic pesticides, 75% had poor personal hygiene and 25% had good one. In addition, it was found that farmers in Demangan Village used insecticide pesticides that belonged to the organophosphate group and were proved to inhibit the acetylcholinesterase enzyme (22).

Low education had a profound effect on the behavior of farmers in pesticide management practices. This was identified in the double linear regression equation with a positive coefficient which meant that the higher education, the higher level of acetylcholinesterase in the blood would be. Low education in farmers also affected their awareness of pesticide hazards for health (25). In addition, factors influencing education included psychology, socio-cultural, the development of science and technology, socioeconomic, and ideology (17).

There were farmers aged 40 years old who were at very high risk, this was due to smoking habits, using 4 types of pesticides, never getting training, not using personal protective equipment, and having poor personal hygiene. Farmers with a working period of 10 years increased the risk of accumulating pesticides in the body. Farmers with high working life were in the category of very high risk and aggravated by spraying activities carried out more than 6 hours (18).

Farmers who cultivated rice and horticulture were more at risk of pesticide contamination in the blood due to the use of 4-5 types of insecticides and 4 types of fungicides when they cultivated rice using 2 types of insecticides and 2 types of fungicides. Horticulture cultivation conducted intensive spraying to increase pesticide contamination in the blood (22)(26).

Reading the labels was the first step of pesticide safety management, it certainly required knowledge of it. Important information on pesticide packaging labels was essential in safe use for the health and environment (29). Yet, farmers had never gained knowledge on how to read pesticide labels so it was only experience-based. Previous research had stated that farmers with abnormal levels of Acetylcholinesterase did not follow the instructions for pesticide use on the label (25). In addition, the instructions on pesticide labels were very difficult for farmers to understand so it required some efforts to improve farmers' understanding of how to comprehend pesticide labels.

Proper and safe storage of pesticides was one of the keys to recover the health of farmers from pesticide exposure. Previous research had declared that decreased levels of acetylcholinesterase in the blood were influenced by storing unsafe pesticides (26). Some farmers stored pesticides in the rice fields and were at very high risk because the farmers were illiterate, aged 40 years, working period > 10 years, smoking, using pesticides of insecticide, and fungicides, and not using personal protective equipment.

There were farmers who had participated socialization related to pesticide management and were at very high risk. Based on the observations and interviews of the farmers, despite having obtained information regarding the safe management of pesticides, they still used pesticides according to their habits according to experience. If the prior pesticide measure had not reduced pests and plant diseases then the pesticide measure would be fulfilled. Therefore, the increasing dose of pesticides

increased pesticide contamination in the body. This was due to a lack of oversight and ongoing programs from the relevant stakeholders.

Farmers did not use personal protective equipment because of the following reasons: the unavailable facilities, spending money if they worked in safety. It was necessary to get attention from the relevant stakeholders to support safety agricultural activities. Previous research had informed that the completeness of PPE when using pesticides had an effect on acetylcholinesterase levels in the blood and most pesticide contamination through the skin followed by breathing (21)(24)(27). Especially in mixing and handling activities were riskier than other activities (27). Personal protective equipment that should be used by farmers was protective suits, aprons, face shields, goggles, full respirators, masks, gloves, and anti-chemical boots.

Smoking habits and without using the correct personal protective equipment increased the level of pesticide poisoning in the blood. Poor personal hygiene was not immediately washing hands after spraying and subsequently pesticides in the air sucked by farmers when smoking. Furthermore, tar and nicotine contained in cigarettes had an effect on acetylcholinesterase in the blood. It was simultaneously of the reactive oxygen production of species in exogenous and endogenous environments so that intracellular oxidative pressure improved multistage carcinogenic processes (19)(23).

At such time, farmers admitted to washing their hands in rice fields after doing spraying and bathing at home. The used clothes for spraying are still worn and washed upon arrival at home, thus it increased contamination to family members or take-home pesticide exposure pathways (6)(28). Absorption of pesticide levels in the blood through the skin occurred if the farmer did not shower immediately after spraying (7)(13). Personal hygiene had an important role to play in preventing pesticide contamination in farmers' bodies. Even previous research had stated that hand and face washing was ineffective at eliminating pesticide contamination (25). Thus, farmers should take a shower immediately after doing the spraying.

The strengths of this research were able to expose the characteristics of farmers who were at very high risk and investigated their reasons who did not do pesticide management safely. In addition, researchers also conducted observations on the behavior of farmers in rice fields. The limitation of this study was that blood sampling was carried out without considering the time-lapse of the activity of spraying farmers.

The conclusion of this study was the activity of using pesticides that were more than one type or mixing 2 types of pesticides even more significantly affects levels of acetylcholinesterase enzyme in the blood. Agricultural activities still became hereditary habits. Safe pesticide management needed to be applied since purchasing by reading pesticide labels, storage, mixing, handling, spraying, and also

cleaning activities after spraying. The use of personal protective equipment and personal hygiene was a supportive aspect of the protection of farmers from pesticide contamination. Farmers who were at very high risk should stop spraying activities 1-2 months to increase the levels of pesticides in the blood to the normal limit (24). The relevant stakeholders needed to create a structured program to monitor and to improve the level of health of farmers related to the dangers of pesticide use.

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