THE SELF-REPORT OF HEALTH EFFECT ON BLOOD CHOLINESTERASE LEVEL OF PESTICIDE EXPOSURE; A CASE STUDY AMONG RICE FARMERS IN TARNLALORD, PHIMAI, NAKHON RATCHASIMA, THAILAND

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Abstract- Pesticide has been imported to Thailand for several years including insecticide, herbicide, and others coinciding with the expansion of the country's agricultural system. Among these chemicals are organophosphate and carbamate the groups of chemicals primarily used in agriculture. Exposure to these pesticides can cause harmful effect on human health. This study aims to find health effects of organophosphate and carbamate pesticides exposure and to assess to blood cholinesterase levels of acetyl cholinesterase (AChE) and plasma cholinesterase (PChE) in dry-season crops, among rice farmers (n=33) in Tarnlalord Sub-District, Phimai District, Nakhon Ratchasima Province, Thailand. The design of this study was a cross-sectional study with questionnaire by face to face interview and blood-tests by Test-mate ChE (Model 400) instruments. The average age (± SD) of the study was 46 ± 9.38 years old. The results showed the ChE levels in rice farmers including (1) the first blood collection, 24 hours after application, 72.70% of the farmers were abnormal (2). The farmers reported their adverse health effects related to gastrointestinal system, urinary system, eye, skin, and central nervous system. Additionally, the AChE level within 24 hours after first application was significantly associated to eye symptoms (Chi-square, $p<0.05$). This study showed that after the rice farmers applied pesticides at the beginning, both AChE and PChE levels were abnormal with self-recovery to normal levels by time. The appropriated self-practices and prevention from pesticides exposure should be recommended to rice farmer regarding proper use of personal protective equipment (PPE) and pesticides handling to reduce adverse health effects from pesticides exposure.

Index Terms - Health Effects / Cholinesterase / Organophosphate (OPs) And Carbamate / Male

I. INTRODUCTION

Thailand covers about 513,115 km2 and has a population of around 64 million [1], Thailand's major source of income and occupation is agriculture. In 2012, the morbidity rate from toxic substance was 2.35 per 100,000 populations or 1,509 people [2]. However, organophosphate (OPs) and Carbamate insecticides form are the groups of chemicals that are primarily used in agriculture. Pesticides have caused several harmful effects on the health of humans [3]. The quantitative determination of cholinesterase in whole blood can show the level of pesticide exposure. OPs and carbamate insecticides have the highest morbidity rate of poisoning among the farmer [4]. While, the measurements of biological monitoring that enters the body used are monitoring in blood, urine, saliva, or breast milk as biological media by the estimate of the amount of pesticide as its metabolite or its reaction product which is absorbed into the body [5]. Therefore, the aims of this research were to find blood cholinesterase levels of acetyl cholinesterase (AChE) and plasma cholinesterase (PChE) for 1 times in dry-season crops, and to assess the general health effect of organophosphate and carbamate pesticides exposure by self-report within 24 hours after pesticides application among rice farmers.

II. MATHERIALS ANDMETHOD

The design of this study was cross-sectional study. Participants of the study include male citizens [6], aged 18 to 59 years old, who have been rice farmers for more than 1 year, growing rice in this crop that live in Tarnlalord sub-district, Phimai district, Nakhon Ratchasima province, and commonly use OPs and carbamate pesticide in the paddy field. Those with a history of liver failure, myocardial infarction and malnutrition were excluded. The representatives of the farmers include those performing mixing, loading and spraying of pesticide by themselves (1 subject per household). The sampling by used the criteria to get target number of sample were 33 participants.

III. INSTRUMENT OF STUDY

1) *Questionnaire:* The questionnaires consisted of closed and open-ended questions. This questionnaire was modified from a questionnaire previously used in the pesticide safe use in rice farmers [5]. Three pesticide experts were brought on to inspect the

validity of questionnaire. The IOC score given by the experts was 0.90. After the reliability was improved, the questionnaire was administered to 30 farmers in Naimuang sub-district, Phimai district, Nakhon Ratchasima province. Then, the cronbach's alpha coefficient was calculated by using licensed SPSS Version 17. The found cronbach's alpha coefficient value was 0.731.

2) *Blood collection and measurement:* The blood cholinesterase levels were tested by using the basis of Ellman method with Test-mate ChE (Model 400), EQM. The blood test was collected at 20µL per person of farmers per time. Their blood was collected within 24 hours after 1st application. The analysis of blood ChE levels test in AChE and PChE was conducted by the researcher. Farmers have to wash their hands with soap first. Then, the nurse use surgical cotton moistened with alcohol to clean the finger in order to take the sample. After the alcohol dried off, a nurse uses a needle to puncture the finger. A nurse was to not squeeze the finger. If the blood is not enough, puncture the finger again. Therefore the reliability of lab measurement the researcher was blood tested on Test-mate ChE (Model 400).

ETHICAL CONSIDERATION

The experimental protocol was approved by the Ethics Review Committee for Research Involving Human Research Subjects, Health Sciences Group, Chulalongkorn University with the certified code COA No.055/2014.

IV. RESULTS

In this paddy field, the rice farmers consisted of 33 males. The average age (± SD) = 46 (±9.38). Most of participants graduated from primary school (78.80%). Twenty four percentages of participants had a total household income of less than 30,001 THB (1 USD ~ 33 THB) per year.

In the farming characteristics found the average (± SD) years of working duration on a farm was 19.64 (±1.43). The average history of pesticide use was 13.85 (±1.04) years. The average farm size was 18.58 (±1.41), (1 rais = 1,600 Sqm.). Most of participants had the duration of pesticide spraying between ½-1 hour/day (30.30%). 78.50% of them were used spraying by a backpack spray and 24.20% of them were used spraying by hand. The activities related in rice farm found all of the rice farmers were Spraying, Mixing, and Loading pesticides. 45% of participants were praying pesticides in the early morning (6:00 am.–9:00 pm.).

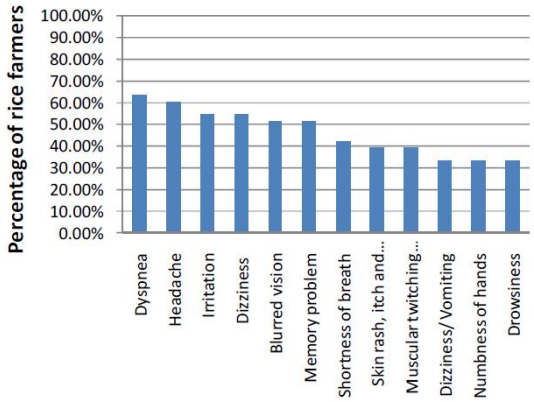

Figure I: Symptoms of pesticides that rice farmers exposure report (n = 33)

A. Self-report on health effects of organophosphate (OPs) and carbamate pesticides exposure

The general health effects of OPs and carbamate pesticides exposure to rice farmers by self-report, found that most the of rice farmers had respiratory disorders including dyspnea 63.60%, shortness of breath 42.40%, bronchorrhea 27.30%, and running nose 12.10%. The gastrointestinal systems were affected in a few of rice farmers; 33.30% of dizziness/vomiting, 18.20% of anorexia and 12.10% of stomach ache. Only few on them experienced loss of urinary control 12.10%. In the same direction, the glands also affected a few of rice farmers, 15.20% of sweating and 12.10% of hyper salivation. However, the main eye symptoms of participants were irritation 54.50%, follow by blurred vision 51.50% and the least were lacrimation 12.10%. For the skin systems; 39.40% of them had skin rash, itch, burn and muscular twitching and cramps and 33.30% of them had numbness of hands. Most symptoms occurred in the central nervous system. The majority of the participants experienced headache 60.60%, the inferior was dizziness 54.50% and the least is memory problem 51.50%. Conversely, only few symptoms of central nervous system are drowsiness 33.30%, ataxia 12.10% and slurred speech and trembling of hands 9.10% (n = 3.00), respectively.

B. Blood cholinesterase concentration in rice farmers

In this study, farmers registered in the study of blood cholinesterase had their blood collected to find levels of blood acetyl cholinesterase (AChE) and plasma cholinesterase (PChE). For the blood collection, blood was collected from the participants within 24 hours after 1st application. AChE and PChE levels means (U/ml) were showed the first blood collection demonstrated 27.30 % of participants' AChE levels were normal and 72.70% of the participant had an abnormal AChE levels.

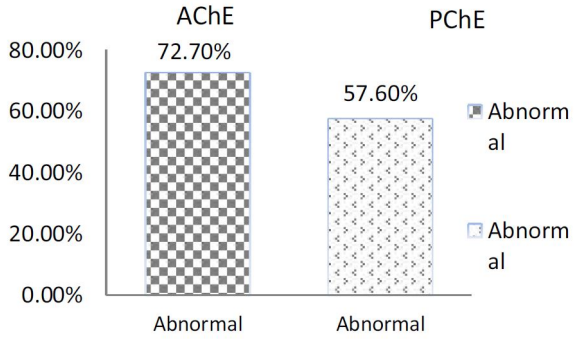

Figure 2: Blood cholinesterase level among rice farmers (n = 33)

C) The relationship between demographic characteristics and blood cholinesterase levels

The study showed that age was statistically significant and was related with AChE in the first application (p = 0.007). Most of participants aged between 46 – 59 years old had an abnormal AChE levels compared to other range groups. On the other hand, the relationship between demographic characteristics and AChE levels; education level and average total household income (Baht/Year) were not statistically significant. (Table II)

D) The relationship between farming characteristics and blood cholinesterase levels

The statistical analysis found that AChE in first application (within 24 hours after 1st pesticides application) were significantly associated with the duration of pesticide spraying (p = 0.018), type of sprayer (p = 0.047), most common type of pesticide (p = 0.012) and time of pesticide spraying period (p = 0.033). Other factors of farming characteristics did not have significant relationships with AChE levels. However farming characteristics were not significantly related to PChE. (Table III)

E) The relationship between blood ChE levels and health effect of Ops and carbamate pesticides exposure

The study of relationship between health effect of organophosphate and carbamate pesticides exposure and blood cholinesterase levels demonstrated the significant relationship between PChE in first application (within 24 hours) with blurred vision in the eye symptoms (p = 0.024, OR = 0.041, 0.836). The other symptoms of other health systems and PChE levels, on the other hand, were not significantly related. The results of the finding are shown in Table V.

DISCUSSION

In this study, female farmers were excluded because of a known influence of gender on ChE activity and there were only few women participants in the study [6]. The results showed that age was significantly related with AChE (within 24 hours after 1st application) as the researcher expected. Most of the farmers were 46–59 years old. As age increased, metabolism activity in body adverse also decreased. The pesticide exposed by rice farmers were eliminated out of the body at a slow rate and caused them to have lower AChE. The result was different to previous study of [7]. The presented study indicated both AChE and PChE levels were not significant with education level and average total household income (Baht/year).

The study found that AChE during the first application were significant with the duration of pesticide spraying, type of sprayer, most common type of pesticide and time of pesticide spraying period. These factors have made rice farmers who sprayed pesticide for more than 2 hours have lower AChE, The longer they sprayed pesticide, the more exposure they had [8]. The study of Magauzi reported similar results by using multivariable analysis to confirm the positive relationship between ChE levels within 24 hours after first application and type of pesticide used [9]. The rice farmers who used backpack sprayers have higher chance of getting high exposure from pesticides compared to hand sprayer because backpack spray create very small drop or aerosol of pesticide which is easily absorbed by the body (10). It is important to note that AChE activity in erythrocytes was also associated with the method of use, practice, chemical content, frequency and chemical type of farmers in Sam Chuk, Suphan Buri, Thailand [8] while other factors of farming characteristics and AChE levels were not significant. The study of personal protective equipment (PPEs) had the principle emphasis of aiming towards to avoiding exposure of skin, mouth, nose and eye when handling pesticide products. In this study, relationship between personal protective equipment (PPEs) and blood cholinesterase levels of the rice farmers was shown, although personal protective equipment (PPEs) including hat, paper mask or handkerchief, long sleeve shirt, long pants, gloves, boots, goggles, and apron and AChE within 24 hours after 1st application were not significant related. However, this study found the significantly relationship between PChE within 24 hours after 1st application and personal protective equipment (PPEs).

The rice farmers use PPEs when doing farming activities though the level of proper use was not taken into consideration. This is crucial for farmers because PPE is required for early entry to treated areas permitted under the worker protection standard. This involved contact with anything that

has been treated, such as plants, soil, or water. There must be coveralls such as chemical-resistant of long-sleeved shirt and long pants, chemical-resistant gloves, such as butyl rubber, nitrile rubber, or neoprene rubber, shoes and socks, protective eyewear, chemical-resistant headgear for overhead exposure [11]. The results of this study showed that proper use of personal protective equipment (PPEs) and blood cholinesterase levels within 24 hours after 1st application were not significant-when looking at AChE levels but were lower for PChE within 24 hours after 1st application. The reason why the rice farmers who wore proper gloves and boots had lower PChE but AChE was normal was because, personal protective equipment (PPEs) could minimize the risk [12] and rice farmers who used proper gloves and boots had less chance of pesticide exposure compared to those who use improper gloves and boots.

The results presented found that only washing hands before eating and drinking after used pesticides was significantly related with AChE within 24 hours after pesticide application. This report was supported by Sapbamrer [13] who also found the significant association between AChE and preventive practices during application contradicting with the studies of wilaiwan who found that washing hands before eating and after the use of pesticide were not significant association with AChE [14]. Washing hands before eating and drinking after using pesticides can protect them from pesticides exposure. Therefore, AChE level and other factors of handling pesticides including reading the label of pesticides products before use, using the recommended amounts of pesticides, self-mixing pesticides by themselves, taking shower by soap or shower cream immediately after applying pesticides, washing clothes by separated working clothes and normally clothes, washing equipment after use keeping of pesticide products at home, keeping of application equipment at home, digging a hole to bury a bottle of pesticide used, smoking while using pesticide (mixing, loading and spraying) and drinking while using pesticide (mixing, loading and spraying) did not demonstrate a significant relationship. The results from statistical PChE analysis found that wearing gloves when mixing, loading, spraying pesticides and digging a

Table II
Relationship between demographic characteristics and blood cholinesterase levels of participants (24 hours after 1st application) (n = 33)

Demographic characteristics	Blood cholinesterase levels			
	AChE		PChE	
	χ2	(P-value)	χ2	(P-value)
Age	10.06	**0.007***	1.18	0.554
Education level	2.588	0.274	0.983	0.612
Average total household income (Baht/Year) (1 USD ~ 30 THB)	1.219	0.748	0.838	0.840

Table III
Relationship between farming characteristics and blood cholinesterase levels of participants (within 24 hours after 1st application) (n = 33)

Farming characteristics	Blood cholinesterase levels			
	AChE		PChE	
	χ2	(P-value)	χ2	(P-value)
Working duration of farm	1.665	0.645	1.042	0.791
History of pesticide used	2.023	0.568	0.445	0.931
Farm size	0.085	0.994	4.225	0.238
Duration of pesticide spraying	7.648	**0.018***	1.306	0.728
Type of sprayer	3.960	**0.047***	1.313	0.252
Most common type of pesticide	8.800	**0.012***	0.794	0.672
Time of pesticide spraying period	6.615	**0.033***	0.294	0.961

Table IV

The relationship between health effect of organophosphate and carbamate pesticides exposure and blood cholinesterase levels (Within 24 hrs. after 1st application) (n = 33)

Health effect of organophosphate and carbamate pesticides exposure	Blood cholinesterase levels							
	AChE				PChE			
	χ2	(P-value)	OR	95%CI	χ2	(P-value)	OR	95%CI
Respiratory system								
Dyspnea	0.049	0.825	1.200	0.239-6.025	0.443	0.506	0.615	0.147-2.582
Bronchorrhea	0.229	0.632	1.500	0.284-7.934	0.021	0.886	1.120	0.239-5.251
Running nose	0.012	0.913	0.875	0.079-9.696	0.107	0.744	1.417	0.174-11.507
Shortness of breath	0.874	0.350	2.083	0.441-9.844	0.571	0.450	1.714	0.422-6.968
Gastrointestinal system								
Anorexia	0.416	0.519	0.475	0.048-4.740	0.248	0.618	0.625	0.097-4.012
Dizziness/ Vomiting	0.688	0.407	1.943	0.399-9.453	0.248	0.618	0.686	0.155-3.036
Stomach ache	1.707	0.191	NC	NC	1.977	0.160	4.909	0.452-53.267
Urinary system								
Loss of urinary control	1.707	0.191	NC	NC	1.977	0.160	4.909	0.452-53.267
Glands								
Hyper salivation	1.185	0.276	3.143	0.37126.621	0.107	0.744	1.417	0.174-11.507
Sweating	0.157	0.692	0.625	0.060-6.486	0.745	0.388	2.318	0.332-16.185
Eye symptoms								
Blurred vision	4.251	**0.039***	2.171	1.129-14.012	5.125	**0.024***	1.685	1.471-9.836
Lacrimation	1.707	0.191	NC	NC	0.107	0.744	1.417	0.174-11.508
Irritation	0.509	0.475	0.571	0.122-2.679	0.066	0.797	1.200	0.299-4.817
Skin symptoms								
Skin rash/ itching/ burning	1.528	0.216	0.338	0.058-1.972	0.138	0.710	0.764	0.184-3.169
Numbness of hands	0.688	0.407	0.476	0.081-2.811	0.248	0.618	0.686	0.155-3.036
Muscular twitching and cramps	0.132	0.716	1.333	0.282-6.300	0.138	0.710	0.764	0.184-3.169
Central nervous system								
Headache	1.354	0.245	0.400	0.084-1.913	1.146	0.284	0.462	0.111-1.921
Dizziness	0.733	0.392	2.000	0.404-9.909	1.340	0.247	0.438	0.107-1793
Drowsiness	0.000	1.000	1.000	0.197-5.000	0.248	0.618	0.686	0.155-3.036
Slurred speech	1.238	0.266	NC	NC	0.112	0.738	0.654	0.053-8.019
Ataxia	1.707	0.191	NC	NC	0.566	0.452	0.410	0.038-4.426
Trembling of hands	0.061	0.805	1.375	0.109-17.316	2.432	0.119	NC	NC
Irritability	0.383	0.536	0.571	0.096-3.409	0.907	0.341	0.468	0.096-2.271
Memory problem	1.638	0.201	0.357	0.072-1.780	1.588	0.208	2.475	0.597-10.269

** Significant at 0.05 probability level*
**Chi-square test*
NC – Not calculated

-hole to bury a bottle of pesticide used were significant with the level of PChE in first application, respectively. However the PChE levels and factors of handling pesticides were not statistically significantly and were similar to results of the previous study [14]

The results of this study showed that only blurred vision was significantly related with AChE within 24 hours after pesticides application in eye symptoms. These rice farmers spray pesticide with bare eyes and no protective equipment. The effects of pesticide are illnesses or injuries do not appear immediately (within 24 hours) after exposure to pesticide making farmers unaware, resulting in very few people wearing goggles to protect their eye. In this study, 32 out of 35 farmers did not used goggles. The results showed significant relationship between cholinesterase and eyes symptoms [14]. Adverse effects can be delayed for weeks, months or even years after the first exposure to a pesticide. Depending upon the toxicity of the compound, dosage and exposure time, the adverse effects of pesticides poisoning ranges from headaches, vomiting, skin irritation, respiratory problems to other neurological disorders [6]. The AChE measurements reflect this slow replacement rate. Thus, AChE is typically used as a marker of chronic exposure [15]. The reported OPs and carbamate poisoning showed the symptom would appear after a while. The symptoms could be acute (or sub-acute) and chronic. The acute symptoms usually emerge within 24 hours after exposure to OPs and carbamate pesticide and include dyspnea, dizziness, vomit, stomach ache, loss urinary control, blur vision. Sub-acute would be present 1-2 weeks after the exposure and most of the symptoms occur with respiratory system such as shortness and dyspnea. However, chronic cases will show the symptom around 2-4 weeks or more and some symptoms are distal weakness, muscle weakness and numbness of hand/leg [16]. The study reported PChE levels showed the significant relationship between PChE in first application and blurred vision in the eye symptom. Therefore, the study found that PChE levels were not significantly related with respiratory systems, gastrointestinal systems, urinary systems, glands, skin symptoms and central nervous systems. In contrast, PChE turnover is much quicker. PChE is a better short-term indicator due to its more rapid response to exposure; it is used as an indicator of recent, acute exposure (Brown et. al., 2006). However some pesticide-related symptom

cholinesterase studies demonstrate no significant relationship with AChE and PChE levels ([17], [6] and [18]).

CONCLUSION

This study showed value of blood cholinesterase (AChE and PChE) level for 3 times collected after rice farmers sprayed pesticides. Their AChE and PChE were found to be lower than the normal standard because of their level of pesticide exposure. As time passed, their AChE and PChE would increase back to the normal state. However, few of rice farmers still had lower AChE and PChE level which may be caused by more pesticide exposure from high frequency of us or from residing nearby farming areas. The rice farmers use PPEs but did not use it correctly. Many of them did not use due to the weather conditions and poor financial state. Those who used adopted incorrect practices such as not wearing masks and gloves when mixing, loading and spraying pesticides. This is the important as it increases pesticides exposure and risk of developing health effects. Method to reduce health effect from pesticides exposure is by wearing appropriate protection equipment and providing knowledge of the rice during pesticides application.

ACKNOWLEDGEMENT

This research was supported by College of Public Health Sciences Chulalongkorn University and the 90th Anniversary of Chulalongkorn University Fund (Ratchadaphiseksomphot Endowment Fund).

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