

PESTICIDE EXPOSURE AND BLOOD CHOLINESTERASE LEVELS AMONG ADOLESCENTS FROM FARMING FAMILIES IN NORTHERN THAILAND

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ABSTRACT

Background. Adolescents living in agricultural communities may be at risk for the adverse effects of pesticide exposure because they are involved in agriculture either as a career or to support their families.

Objectives. The purpose of this study was to investigate the association of farm activities related to pesticide exposure on blood cholinesterase (ChE) levels among adolescents from farming families in the north of Thailand.

Material and Methods. This cross-sectional study included 336 adolescents aged 12-19 years from farming families in Chiang Dao District, Chiang Mai Province. Data on pesticide exposure was collected using a questionnaire, and blood ChE activity was assessed using a ChE reactive paper test kit via fingerstick blood sampling.

Results. Overall, 51.2% of participants had abnormal blood ChE levels. Univariable logistic regression analysis revealed that pesticide-related activities on farms associated with abnormal ChE levels were mixing/spraying (OR=10.54; 95%CI=4.63-23.99), assisting or working in areas with pesticide application (OR=5.54; 95%CI=3.45-8.89), and harvesting (OR=3.70; 95%CI=2.35-5.82). In a multivariable model (Nagelkerke R²=0.374), mixing/spraying (OR=4.90; 95%CI=2.03-11.83) and assisting or working in areas with pesticide application (OR=2.61; 95%CI=1.49-4.57) were significantly associated with abnormal ChE levels, but harvesting (OR=1.48; 95%CI=0.84-2.61) was not significant after adjusting for sex, age in years, and entering or walking through a farm.

Conclusions. The findings indicated that Thai adolescents living in farming families are at risk of pesticide exposure, particularly those involved in agricultural activities such as pesticide applicators. An intervention and measure to raise awareness and reduce the risk of pesticide exposure in adolescents is required.

Keywords: *pesticide, organophosphate, carbamate, agriculture, cholinesterase, teenager*

INTRODUCTION

According to the Food and Agriculture Organization of the United Nations (FAO) report, the total amount of pesticide use worldwide over the past 20 years (2011-2020) tends to continually increase [1]. In Thailand, pesticides are widely used to control insects and other agricultural pests, particularly organophosphates (OPs) such as chlorpyrifos and profenofos, and carbamates (CMs) such as methomyl and carbofuran. Farmers can be directly exposed to pesticides, while their farm families can be indirectly exposed through pesticide drift and take-home exposure [2-4]. This raises concerns about potential health risks, especially among children and adolescents living in agricultural communities who are highly susceptible to pesticide exposure and negative effects as a result

of developmental, dietary, and physiological factors [5-7]. A previous study found that exposure to OP pesticides among school-aged children living in Thai farming communities is impacted by farming activity, household environments, and child behaviors [8].

Blood cholinesterase (ChE) activity levels in the human body are used as a biomarker to determine internal exposure to OP and CM pesticides, which act as cholinesterase inhibitors. According to prior research, lower levels of ChE activity were observed among children and adolescents residing in rural and agricultural communities [2, 9-10]. In addition, adolescents can become occupationally exposed to pesticides by working on farms to support their families. Previous studies found that adolescent pesticide applicators had decreased ChE levels, which were associated with their health, including

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depression symptoms, neurological symptoms, and poorer neurobehavioral performance [11-18]. As a result, adolescents living in farm communities are at risk of pesticide exposure through a variety of routes, with serious health consequences. However, little is known about pesticide exposure patterns and health monitoring in Thai adolescents living in agricultural settings.

The main purposes of this study were to assess blood ChE levels and investigate the association between farm activities related to pesticide exposure and blood ChE levels among Thai adolescents from farming families in Northern Thailand. An additional goal of the study was to examine demographic factors and environmental pesticide exposure that are associated with ChE levels. The findings of this study can be implemented to the surveillance and prevention of pesticide-related health risks. It is also expected to make recommendations on health policy and planning to minimize pesticide exposure risks among Thai adolescents living in agricultural areas.

MATERIAL AND METHODS

Design, setting, and subjects

This cross-sectional study was conducted on adolescents from farming families in Chiang Dao District, Chiang Mai Province, Northern Thailand. The area of Chiang Dao District was purposively selected because the favorable climate and geographical features provided conditions for cultivation throughout the year, with a large portion of the population engaged in various agricultural activities. These activities include crop cultivation by farmers who use pesticides, particularly OPs and CMs, to increase crop yields and protect against pests. Adolescents are commonly involved in agricultural activities to help their families, such as handling pesticides and assisting with requested tasks on farmland.

Participants for the study were chosen using convenience sampling. Local administrative organizations conducted public relations to invite adolescents who met the inclusion criteria through two schools that serve as gathering places for adolescents all over Chiang Dao District. Adolescents aged 12 to 19 who had spent at least one year living with a family member who worked as a farmer and used agricultural pesticides were eligible for inclusion. The sample size for estimating a population proportion [19] was determined by defining the estimated proportion at 50%, the confidence level at 95%, the level of precision at 0.05%, and the population size at 2,000. Adding 5% of the participants to prevent loss of data yielded a sample size of 340. The final number of adolescents for data analysis was 336, due to errors during blood sampling and testing.

This study received ethical approval from the Committee of Research Ethics, Faculty of Public Health, Chiang Mai University (No. ET014/2023). Before enrollment in the study, subjects aged 12 to 17 provided written informed assent, while those aged 18 to 19 provided written informed consent. For subjects under the age of 18, written informed consent was also obtained from their parents or legal guardians.

Data collection

Data was collected between July and August 2023. In-person interviews were conducted by assistance researchers who had received training prior to data collection. An interviewer-administered questionnaire contained the following sections: (a) demographic characteristics (sex, age, ethnicity, education level, underlying disease, smoking status, and alcohol drinking); (b) environmental pesticide exposure (entering or walking to agriculture area, distance between home and nearest farm, using household insecticides, and house cleaning with a wet cloth); (c) farm activities related to pesticide exposure (mixing and spraying pesticides, assisting or working in areas where pesticides were being applied, and harvesting), along with activity frequency, use of personal protective equipment (PPE), and handwashing and bathing after activities.

A registered nurse collected fingerstick blood samples to assess ChE activity in serum using ChE reactive paper (GPO, Thailand), a standard technique recommended by the Bureau of Occupational and Environmental Diseases, Ministry of Public Health. For laboratory testing, this method had sensitivity, specificity, and positive predictive value of 89.89%, 95.65%, and 94.59%, respectively, while for field screening, it had 77.04%, 90.01%, and 90.38% [20]. Blood ChE levels were classified into four levels based on color indication (Bigg's method): normal (yellow; ≥ 100 U/ml), safe (greenish yellow; 87.5-99.9 U/ml), risky (green; 75.0-87.4 U/ml), and unsafe (dark green; ≤ 75 U/ml) [20]. Unsafe and risky levels were defined as "abnormal", while safe and normal levels were defined as "normal".

Data analysis

Descriptive statistics, such as frequency and percentage, were used to analyze the sample variables. *Chi-square* tests were used to investigate the factors (demographic characteristics and environmental pesticide exposure) that are associated with blood ChE levels. Binary logistic regression was then used to determine the strength of the association between pesticide-related farm activities and blood ChE levels. This study examined both a single model, in which each farm activity was tested independently, and a multiple model, in which different farm activities

were tested together. A p-value of less than 0.05 indicated statistical significance, and an odds ratio (OR) with 95% confidence interval (95%CI) was reported. All statistical analyses were performed using SPSS Version 28 (IBM Corp., Armonk, NY, USA).

RESULTS

The demographics and environmental pesticide exposure of the 336 adolescents under study are presented in Table 1. Blood ChE levels were found to

Table 1. Demographic characteristics and environmental pesticide exposure among adolescents, classified by blood ChE levels (n=336)

Variables	n (%)	Blood ChE levels		p-value*
		Normal (n=164) n (%)	Abnormal (n=172) n (%)	
<i>Sex</i>				
Male	104 (31.0)	40 (38.5)	64 (61.5)	0.011
Female	232 (69.0)	124 (53.4)	108 (46.6)	
<i>Age</i>				
12-14 years	69 (20.5)	50 (72.5)	19 (27.5)	<0.001
15-19 years	267 (79.5)	114 (42.7)	153 (57.3)	
Mean \pm standard deviation (SD)	15.72 \pm 1.56	15.38 \pm 1.70	16.05 \pm 1.35	
<i>Ethnicity</i>				
Non-ethnic	189 (56.3)	99 (52.4)	90 (47.6)	0.138
Ethnic	147 (43.8)	65 (44.2)	82 (55.8)	
<i>Education level</i>				
Junior high school	103 (30.7)	60 (58.3)	43 (41.7)	0.021
Senior high school	233 (69.3)	104 (44.6)	129 (55.4)	
<i>Underlying disease</i>				
No	320 (95.2)	156 (48.8)	164 (51.2)	0.922
Yes	16 (4.8)	8 (50.0)	8 (50.0)	
<i>Smoking</i>				
No	325 (96.7)	157 (48.3)	168 (51.7)	0.317
Yes	11 (3.3)	7 (63.6)	4 (36.4)	
<i>Alcohol drinking</i>				
No	277 (82.4)	134 (48.4)	143 (51.6)	0.730
Yes	59 (17.6)	30 (50.8)	29 (49.2)	
<i>Entering or walking through a farm</i>				
No	53 (15.8)	41 (77.4)	12 (22.6)	<0.001
\leq 1 time per month	177 (52.7)	100 (56.5)	77 (43.5)	
>1 time per month	106 (31.5)	23 (21.7)	83 (78.3)	
<i>Home proximity to nearest farm[†]</i>				
<300 m	76 (23.2)	33 (43.4)	43 (56.6)	0.474
300 m – 1 km	75 (22.9)	36 (48.0)	39 (52.0)	
>1 km	176 (53.8)	91 (51.7)	85 (48.3)	
<i>Use of household insecticides[†]</i>				
No	74 (22.6)	39 (52.7)	35 (47.3)	0.460
Yes	253 (77.4)	121 (47.8)	132 (52.2)	
<i>House cleaning with a wet cloth[†]</i>				
<1 time per month	25 (7.6)	7 (28.0)	18 (72.0)	0.209
1-3 times per month	119 (36.4)	60 (50.4)	59 (49.6)	
At least 1 time per week	110 (33.6)	55 (50.0)	55 (50.0)	
Every day or almost every day	73 (22.3)	38 (52.1)	35 (47.9)	

[†] Unknown answer (n=9); * Chi-square test

Table 2. Farm activities related to pesticide exposure among adolescents, with frequency, PPE use, and hygiene behaviors classified by sex

Exposure characteristics	n (%)	Sex	
		Male n (%)	Female n (%)
<i>1. Mixing and spraying</i>			
No	274 (81.5)	73 (70.2)	201 (86.6)
Mixing	39 (11.6)	15 (14.4)	24 (10.3)
Spraying	7 (2.1)	4 (3.8)	3 (1.3)
Mixing and spraying	16 (4.8)	12 (11.5)	4 (1.7)
– Frequency of mixing and applying (n=62)			
Less than once per year	41 (66.1)	18 (58.1)	23 (74.2)
Yearly	15 (24.2)	7 (22.6)	8 (25.8)
Monthly or weekly	6 (9.7)	6 (19.4)	0 (0.0)
– Use of PPE (n=62)			
Goggles	9 (14.5)	3 (9.7)	6 (19.4)
Face mask	37 (59.7)	21 (67.7)	16 (51.6)
Gloves	23 (37.1)	11 (35.5)	12 (38.7)
Long sleeve shirt and long pants	40 (64.5)	20 (64.5)	20 (64.5)
Boots	14 (22.6)	7 (22.6)	7 (22.6)
– Handwashing and bathing after pesticide use (n=62)			
No	7 (11.3)	4 (12.9)	3 (9.7)
Yes	55 (88.7)	27 (87.1)	28 (90.3)
<i>2. Assisting or working in areas with pesticide application</i>			
No	145 (43.2)	38 (36.5)	107 (46.1)
Yes	191 (56.8)	66 (63.5)	125 (53.9)
– Frequency of any work (n=191)			
Less than once per year	121 (63.4)	38 (57.6)	83 (66.4)
Yearly	48 (25.1)	17 (25.8)	31 (24.8)
Monthly or weekly	22 (11.5)	11 (16.7)	11 (8.8)
– Use of PPE (n=191)			
Goggles	6 (3.1)	2 (3.0)	4 (3.2)
Face mask	56 (29.3)	26 (39.4)	30 (24.0)
Gloves	20 (10.5)	5 (7.6)	15 (12.0)
Long sleeve shirt and long pants	98 (51.3)	39 (59.1)	59 (47.2)
Boots	22 (11.5)	13 (19.7)	9 (7.2)
– Handwashing and bathing after work (n=191)			
No	37 (19.4)	10 (15.2)	27 (21.6)
Yes	154 (80.6)	56 (84.8)	98 (78.4)
<i>3. Harvesting</i>			
No	153 (45.5)	36 (34.6)	117 (50.4)
Yes	183 (54.5)	68 (65.4)	115 (49.6)
– Frequency of harvesting (n=183)			
Yearly	157 (85.8)	51 (75.0)	106 (92.2)
Monthly	20 (10.9)	14 (20.6)	6 (5.2)
Weekly	6 (3.3)	3 (4.4)	3 (2.6)
– Use of PPE (n=183)			
Face mask	20 (10.9)	6 (8.8)	14 (12.2)
Gloves	38 (20.8)	12 (17.6)	26 (22.6)
Long sleeve shirt and long pants	82 (44.8)	37 (54.4)	45 (39.1)
– Handwashing and bathing after work (n=183)			
No	32 (17.5)	12 (17.6)	20 (17.4)
Yes	151 (82.5)	56 (82.4)	95 (82.6)

be 0.9% for normal, 47.9% for safe, 42.3% for risky, and 8.9% for unsafe. In total, the prevalence of abnormal ChE levels was 51.2% (95% CI: 45.8-56.6). *Chi-square* tests revealed a significant association between blood ChE levels and demographic variables including gender ($p=0.011$), age ($p<0.001$), and education level ($p=0.021$). We also observed a significant association between entering or walking through agricultural areas and blood ChE levels ($p<0.001$).

Regarding pesticide-related activities on farms, approximately 18.5% of adolescents had a history of mixing or spraying pesticides, and more than half reported assisting or working in areas where pesticides were being applied (56.8%) and harvesting produce contaminated with pesticide residues (54.5%) (Table 2). No one reported wearing a chemical mask or respirator when handling pesticides or performing activities in areas where pesticides were being applied.

Association between pesticide-related activities on farms and blood ChE levels in adolescents is shown in Table 3. The modeled odds of abnormal ChE activity increased significantly for mixing or spraying pesticides (OR=10.54, 95% CI: 4.63-23.99), assisting or working in areas with pesticide application (OR=5.54, 95% CI: 3.45-8.89), and harvesting (OR=3.70, 95%CI: 2.35-5.82). After controlling for sex, age in years, and entering or walking through a farm, the modeled odds of abnormal ChE activity decreased but remained significant for each of the farm activities.

In the multiple exposure model, abnormal ChE activity was significantly associated all farm activities, including mixing or spraying pesticides (OR=5.75, 95% CI: 2.45-13.49), assisting or working in areas with pesticide application (OR=3.16, 95% CI: 1.86-5.35), and harvesting (OR=1.93, 95%CI: 1.15-3.25), as shown in Table 4. However, harvesting became insignificant after adjusting for sex, age in years, and entering or walking through a farm.

Table 3. Association between pesticide-related activities on farms and blood ChE levels in adolescents: single exposure model

Exposure characteristics	Blood ChE levels		Crude OR (95% CI)	Adj OR ^a (95% CI)	Adj OR ^b (95% CI)
	Normal n (%)	Abnormal n (%)			
<i>1. Mixing/spraying</i>					
No	157 (57.3)	117 (42.7)	1	1	1
Yes	7 (11.3)	55 (88.7)	10.54 (4.63-23.99)*	8.63 (3.75-19.88)*	7.18 (3.03-16.99)*
<i>2. Assisting or working in areas with pesticide application</i>					
No	104 (71.7)	41 (28.3)	1	1	1
Yes	60 (31.4)	131 (68.6)	5.54 (3.45-8.89)*	5.04 (3.11-8.16)*	3.72 (2.21-6.25)*
<i>3. Harvesting</i>					
No	101 (66.0)	52 (34.0)	1	1	1
Yes	63 (34.4)	120 (65.6)	3.70 (2.35-5.82)*	3.34 (2.10-5.31)*	2.20 (1.31-3.71)*

^a Adjusted for sex and age in years; ^b Adjusted for sex, age in years, and entering or walking through a farm; * $p<0.05$

Table 4. Association between pesticide-related activities on farms and blood ChE levels in adolescents: multiple exposure model

Exposure characteristics	Model 1 ^a		Model 2 ^b		Model 3 ^c	
	OR (95% CI)	<i>p</i> -value	OR (95% CI)	<i>p</i> -value	OR (95% CI)	<i>p</i> -value
Mixing/spraying	5.75 (2.45-13.49)	<0.001	4.97 (2.09-11.80)	<0.001	4.90 (2.03-11.83)	<0.001
Assisting	3.16 (1.86-5.35)	<0.001	3.05 (1.79-5.20)	<0.001	2.61 (1.49-4.57)	0.001
Harvesting	1.93 (1.15-3.25)	0.013	1.88 (1.11-3.18)	0.018	1.48 (0.84-2.61)	0.180
Nagelkerke R ²	0.300		0.316		0.374	

^a Unadjusted; ^b Adjusted for sex and age in years; ^c Adjusted for sex, age in years, and entering or walking through a farm

DISCUSSION

Our findings revealed that the majority of adolescents from farming families in the north of Thailand had abnormal blood ChE levels, reflecting the degree of OP and CM exposure that entered the body via multiple routes, including inhalation, consumption, and dermal contact. Adolescents' high level of exposure may be due in part to living near pesticide-treated farmland, and families who work in agriculture unintentionally bring pesticide residues home on their clothing, shoes, or equipment [3-4]. It is consistent with previous research, which found lower levels of ChE activity among adolescents residing agricultural communities in Ecuador [2]. Similarly, 42.9% of Indonesian school-age children had low ChE levels [21]. Existing research suggests that environmental exposure to OP pesticides may cause changes in reproductive and thyroid hormone levels in adolescents [22-23]. In terms of occupational pesticide exposure, particularly OPs, decreased blood ChE activity has been linked to neurological symptoms and neurobehavioral effects in adolescent workers [11, 14-15, 24]. Furthermore, other pesticide classes are used in agricultural areas of Northern Thailand, in addition to OP and CM pesticides [25]. These findings indicate that adolescents from farming families are at high risk of health consequences from pesticide exposure, emphasizing the importance of health surveillance for them.

Regarding farm activities related to pesticide exposure, it was found that each one under investigation – mixing and spraying, assisting or working in areas where pesticides were being applied, and harvesting – was significantly associated with blood ChE levels in adolescents. Our findings indicated occupational pesticide exposure among Thai adolescents, which could contribute to their higher health risk. It is consistent with numerous studies that found that adolescent pesticide applicators in Egypt had significantly decreased plasma and erythrocyte ChE levels than non-applicators [11-12, 16]. About 18.5% of the adolescents reported having direct contact with pesticides via mixing or spraying, with most of applications occurring on a low frequency. This is comparable to the findings of adolescent Latino farmworkers, who reported applying or mixing agricultural chemicals at 21.6% [26].

More than half (56.8%) of the adolescents reported assisting or working in pesticide application areas, which was linked to lower blood ChE levels. It is true that common farm activities such as helping parents mix pesticide, pulling pesticide spraying hoses, and cleaning equipment can increase the risk of pesticide exposure through inhalation and dermal contact. This is consistent with a previous study,

which found that being present during the application and having any direct contact with the application process was associated with changes in urinary biomarker concentrations in farm families such as children and spouses [27]. Poor PPE use among adolescents, particularly the failure to wear chemical masks or respirators while applying pesticides or engaging in other agricultural activities during the application, has been reported, potentially leading to increased exposure levels and adverse health effects [28-29]. The findings of this study suggest the need for pesticide training or an intervention to improve pesticide application practices, including the adoption of PPE, among adolescents engaged in farm activities, particularly those who are mixers or sprayers [26, 30].

Harvesting activities lost significance in the multiple exposure model after potential factors like entering or walking through a farm were accounted for. This could be due to indirect contact and low dose exposure caused by bodily contact with pesticide-treated crops during harvesting tasks such as picking, cutting, and sorting, which transfers pesticide residues contaminated on crop surfaces to their skin [31-32]. This is consistent with a study conducted among Indonesian schoolchildren, which found that cutting onion leaves (Crude OR=2.41, 95%CI: 1.35-4.29) and carrying onion harvest to other places (Crude OR=1.91, 95%CI: 1.21-3.03) are potential risk factors for OP pesticide exposure measured by metabolites [33]. Likewise, OP pesticides on hands were associated with lower plasma ChE levels among Thai vegetable vendors, implying primarily dermal exposure via hand contact [34]. According to a study by Chetty-Mhlanga et al. [35], picking fruit is the most frequently reported farm activities, and children aged 9-16 who engage in pesticide-related activities, particularly picking crops from the field, may be at a higher risk for health effects. However, 85.8% of the adolescents reported harvesting on a yearly basis, which may result in diminished pesticide exposure.

Demographic characteristics including sex, age, and education were found to be associated with ChE levels in Thai adolescents. This may also be because a higher proportion of male and older adolescents, as well as those with senior high school degree, reported participating in farm activities, leading to greater pesticide exposure and lower ChE activity. According to literature reviews, boys are more likely to participate in activities than girls [36], and men are typically responsible for direct contact with pesticides as farming is physically demanding [37]. Previous research revealed sex-specific effects, with males being more susceptible to OP exposure [38]. In addition, environmental pesticide exposure, like entering or walking through a farm, was associated with lower ChE levels. It is possible that adolescents

come into contact with pesticide-contaminated environments via various pathways while on the farm, such as playing in fields and plantations that have recently been treated with pesticides. A previous study in Thailand observed that toddlers' frequency of following caregivers to the farm was positively associated with CM residue on their feet [39]. This could imply that adolescents who enter or walk through a farm are exposed to pesticide residues via skin contact.

Regarding the study's limitations, this is a cross-sectional study in which causal relationships cannot be inferred. The study omitted many important details about pesticide exposure (for example, the last date of activities, hours and years of field work, length of time working with pesticides, pesticide active ingredient, and spraying method). Generalizability may be limited as a result of sampling by school unit in the investigated region. Therefore, larger-scale studies covering broader geographical areas should be conducted as well as an investigation into the health effects of pesticide exposure among Thai adolescents. Our study focused on blood ChE levels indicating OP and CM exposure; future research should consider using additional biomarkers to assess other pesticide classes, such as metabolites. However, this is a preliminary survey of teenagers from farming families, highlighting pesticide exposure both directly and indirectly as well as concern about a potential health risk, as a major public health issue.

CONCLUSIONS

This study provides evidence of environmental and occupational pesticide exposure among adolescents from farming families in the north of Thailand. Our findings revealed that pesticide-related activities in the fields, such as mixing and spraying, assisting or working in areas where pesticides were being applied, and harvesting, were associated with lower ChE levels. This raises significant concerns about potential health risks, particularly for pesticide applicators. Addressing pesticide exposure in adolescents necessitates raising awareness and public health education among individuals, parents or guardians, and communities.

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Conflict of interest

All authors declare they have no potential competing interest.

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