The Agricultural health surveillance of chilli farmers exposure to pesticides: A case study of agricultural area, Hua-Rua sub-district, Muang district, Ubonratchathani Province, Thailand

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การเฝ้าระวังสุขภาพของเกษตรกรผู้ปลูกพริกจากการรับสัมผัสสารกำจัดศัตรูพืช: กรณีศึกษาพื้นที่เกษตรกรรม ตำบลหัวเรือ อำเภอเมือง จังหวัดอุบลราชธานี

นางสาวณัฏฐา ฐานีพานิชสกุล

วิทยานิพนธ์นี้เป็นส่วนหนึ่งของการศึกษาตามหลักสูตรปริญญาสาธารณสุขศาสตรดุษฎีบัณฑิต
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Thesis Title	THE AGRICULTURAL HEALTH SURVEILLANCE OF CHILLI FARMERS EXPOSURE TO PESTICIDE: A CASE STUDY OF AGRICULTURAL AREA, HUA-RUA SUB-DISTRICT, MUANG DISTRICT, UBONRATCHATHANI PROVINCE, THAILAND
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ณัฏฐา ฐานีพานิชสกุล: การเฝ้าระวังสุขภาพของเกษตรกรผู้ปลูกพริกจากการรับสัมผัสสารกำจัดศัตรูพืช: กรณีศึกษาพื้นที่เกษตรกรรม ตำบลหัวเรือ อำเภอเมือง จังหวัดอุบลราชธานี. (THE AGRICULTURAL HEALTH SURVEILLANCE OF CHILLI FARMERS EXPOSURE TO PESTICIDE: A CASE STUDY OF AGRICULTURAL AREA, HUA-RUA SUB-DISTRICT, MUANG DISTRICT, UBONRATCHATHANI PROVINCE, THAILAND) อ.ที่ปรึกษาวิทยานิพนธ์หลัก: ผศ.ดร.วัฒน์สิทธิ์ ศิริวงศ์ อ.ที่ปรึกษาวิทยานิพนธ์ร่วม: ศ.ดร.มาร์ค เกรกอรี รอบสัน, 187 หน้า

การเฝ้าระวังทางสุขภาพของเกษตรกรผู้ปลูกพริกตำบลหัวเรือ อำเภอเมือง จังหวัดอุบลราชธานี จากการรับสัมผัสสารกำจัดศัตรูพืชได้ทำการศึกษาในช่วงระหว่างเดือนมีนาคม ถึงเดือนเมษายน 2555 โดยใช้เกษตรกร 40 คน ส่วนใหญ่เป็นเพศชาย มีอายุโดยเฉลี่ย 40.95 (±6.11) ปี และค่าดัชนีมวลกายเฉลี่ย 23.18 (±4.48) และผู้อยู่อาศัยในพื้นที่ 40 คน (กลุ่มเปรียบเทียบ) ซึ่งมีอัตราส่วนเพศชายเท่ากับเพศหญิง และมีอายุเฉลี่ย 38.15 (±11.28) ปี และค่าดัชนีมวลกายเฉลี่ย 23.01 (±4.21) จากการศึกษาพบว่าเกษตรกรได้รับสัมผัสสารกลุ่มออร์แกนโนฟอสเฟต (คลอร์ไพรีฟอส และโพรฟีโนฟอส) ผ่านเส้นทางการรับสัมผัสสารหลากหลายเส้นทางการรับสัมผัส โดยส่วนใหญ่ของเกษตรกรมีการใส่อุปกรณ์ป้องกันร่างกายระหว่างการพ่นสารกำจัดศัตรูพืช และกลุ่มโรคส่วนใหญ่ที่พบ คือโรคที่เกี่ยวข้องกับระบบประสาทส่วนกลาง จากการตรวจวัดสารกำจัดศัตรูพืชตกค้างบนร่างกายหลังจากการฉีดพ่น พบว่า สารกำจัดศัตรูพืชตกค้างบนร่ายกายมากที่สุด บนใบหน้าและมือ ตามลำดับ แต่ไม่พบสารกำจัดศัตรูพืชตกค้างบนเท้าของเกษตรกร จากการตรวจวัดสารกำจัดศัตรูพืชผ่านทางการหายใจ พบว่าเกษตรกรทั้งหมดได้รับสัมผัสผ่านเส้นทางการรับสัมผัสนี้ นอกจากนี้การรับผัสสารกำจัดศัตรูพืชทางผิวหนังและทางการหายใจนั้นไม่มีความสัมพันธ์กัน (r_s=0.155; $p>0.05$) ส่วนสารกำจัดศัตรูพืชตกค้างบนมือมีความสัมพันธ์อย่างมีนัยสำคัญทางสถิติกับการรับสัมผัสทางการหายใจ ($p<0.05$) จากการคำนวณค่าการรับสัมผัสสารต่อวันของเกษตรกรตามวิธีขององค์การพิทักษ์สิ่งแวดล้อม สหรัฐอเมริกา (US-EPA) พบว่า ค่าการรับสัมผัสสารผ่านทางผิวหนังมากที่สุด และเมื่อทำการระบุความเสี่ยงโดยใช้ค่าดัชนีบ่งชี้อันตราย (Hazard Index, HI) พบว่า กลุ่มเกษตรกรดังกล่าวอาจจะไม่ได้รับความเสี่ยงจากการรับสัมผัสสารคลอร์ไพรีฟอส และโพรฟีโนฟอสทางการรับสัมผัสทางผิวหนังและการหายใจ เนื่องจากค่าดัชนีบ่งชี้อันตรายของเกษตรกรทั้งหมดมีค่าน้อยกว่าค่าที่ยอมรับได้ (HI<1.0) แต่อย่างไรก็ตามจากการตรวจวัดหาระดับสารเมตาโบไลต์ของสารกำจัดศัตรูพืชกลุ่มออร์แกนโนฟอสเฟตในปัสสาวะ ผลการศึกษาพบว่า ระดับสารเมตาโบไลต์ของเกษตรกรหลังการฉีดพ่น 1 วันมีความแตกต่างอย่างมีนัยสำคัญกับก่อนการฉีดพ่นสารกำจัดศัตรูพืช และหลังการฉีดพ่นสารกำจัดศัตรูพืช 2 วัน($p<0.05$) รวมไปถึงระดับสารเมตาโบไลต์ของกลุ่มผู้อยู่อาศัยมีความแตกต่างอย่างมีนัยสำคัญกับระดับสารเมตาโบไลต์ของเกษตรกรหลังการฉีดพ่น 1 วัน ($p<0.05$) และพบว่าระดับสารเมตาโบไลต์ของเกษตรมีความสัมพันธ์กับการรับสัมผัสสารกำจัดศัตรูพืชผ่านทางผิวหนัง ดังนั้นเกษตรกรควรได้รับความรู้ด้านสาธารณสุขที่เกี่ยวข้องกับการใช้สารกำจัดศัตรูพืช และการใช้อุปกรณ์ป้องกันที่เหมาะสม เพื่อการเปลี่ยนแปลงพฤติกรรมการใช้สารกำจัดศัตรูพืชที่ถูกต้อง รวมไปถึงการปรับปรุงคุณภาพชีวิตของเกษตรกรให้ดียิ่งขึ้น

สาขาวิชา สาธารณสุขศาสตร์	ลายมือชื่อนิสิต
ปีการศึกษา 2555	ลายมือชื่ออ.ที่ปรึกษาวิทยานิพนธ์หลัก
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NUTTA TANEEPANICHSKUL: THE AGRICULTURAL HEALTH SURVEILLANCE OF CHILLI FARMERS EXPOSURE TO PESTICIDE: A CASE STUDY OF AGRICULTURAL AREA, HUA-RUA SUB-DISTRICT, MUANG DISTRICT, UBONRATCHATHANI PROVINCE, THAILAND. ADVISOR: ASST. PROF. WATTASIT SIRIWONG, Ph.D., CO-ADVISOR: PROF. MARK G. ROBSON, Ph.D., 187 pp.

Health surveillance of chilli farmers in Hua-rua sub-district, Muang district, Ubonratchathani province, Thailand was conducted during March to April, 2012. There were 40 chilli farmers and 40 non-chilli farmers getting involved in this study.Most participated chilli farmers were male. The average age was 40.95 (±6.11) years old and average body mass index (BMI) was 23.18 (±4.48). Male and female was equally in the non-chilli farmers group. The average age and BMI were 38.15 (±11.28) years old and 23.01 (±4.21) respectively. From interview using the face to face questionaire, most chilli farmers usually wore personal protective equipments and had health effects related to central nervous system, such as irritability and memory problem. Organophosphate pesticides (Chlorpyrifos and Profenofos) residue was mostly found on their body, face, and hand, respectively. On the other hand, the residue on feet was not detected. Pesticides were detected in all air samples using personal air sampling technique. Residue on dermal was not associated with inhalation (Spearman's rho =0.155; p>0.05). The Average Daily Dose (ADD) was calculated by the US-EPA recommendation, the highest ADD was obtaining from whole body (dermal contact). The Hazard Index (HI) for risk characterization indicated that the HI of farmers was lower than the acceptable level 1.0. The urinary metabolite level investigated from participants, there was the association between the first post application morning void and pre application morning void (Wilcoxon signed ranks; p<0.05), similar to the first post application morning void and the second post application morning void. The urinary metabolite of the first post morning voidfrom chilli farmers was significantly different from the urinary metabolite of non-chilli farmers' morning void. The main associated of pesticide exposure route and urinary metabolite was found from the dermal route (Spearman's rho= 0.405; p<0.05). This research suggested that public health education training programs including using of appropriate personal protective equipments (PPEs)should be conducted for the chili growing farmers according to improve their ability to handle pesticide and their quality of life.

Field of Study Public Health .... Student's Signature ....
Academic Year 2012 .... Advisor's Signature ....
Co-advisor's Signature ....

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LIST OF ABBREVIATIONS

ABS	= Absorption factor
ADD	= Average daily dose
BMI	= Body mass index
DAP	= Dialkylphosphate
DEP	= Diethylphosphate
DETP	= Diethylthiophosphate
DEDTP	= Diethyldithiophosphate
DMP	= Dimethylphosphate
DMTP	= Dimethylthiophosphate
DMDTP	= Dimehyldithiophosphate
μECD	= Micro electron capture detector
ED	= Exposure duration
EF	= Exposure frequency
FPD	= Flame photometric detector
GC	= Gas chromatography
HI	= Hazard index
HQ	= Hazard quotient
IRIS	= Integrated risk information system
LOD	= Limit of detection
MLA	= Mixing, loading and applying pesticide
NIOSH	= National institute for occupational safety and health
OP	= Organophosphate
OPPs	= Organophosphate pesticides
PPE	= Personal protective equipment
RfD	= Reference dose
RME	= Reasonable maximum exposure
US EPA	= United state environment protection agency
WHO	= World health organization

CHAPTER I

INTRODUCTION

1.1 Background of the study

Thailand is one of the world's largest exporters of agricultural products. Cultivation occupied approximately 40% of the country's area and more than a half of the total national workforce is currently engaged in agriculture (Panuwet et al., 2008; Kanatireklap et al., 2007). The geographic location and climate of Thailand are supported not only plenty of cultivation but insect and pest life cycle also. Thai farmer normally use chemical to speed up, increase, control their harvests and reduce pest and epidemic plants. In 2009, approximately 118,152 tons of pesticides, including insecticide, fungicide and herbicide, were imported to Thailand increasing rapidly 6 times from last decade (Office of agricultural economics, 2009).

Because of the heavy agricultural using pesticides, humans are continually exposed to a large amount of these chemicals (Barr, 2008). Pesticides may cause a health risk to agricultural workers during or after their use. Workers could be exposed to pesticide through multi-pathway; dermal, inhalation and accidental ingestion, during their involving in their job tasks. However, dermal might be the greatest exposure route, inhalation by significant gas vapor pressure and accidental ingestion are consider as following respectively (Chester, 2001).

Thai farmers are at high risk of pesticide poisoning because they has insufficient understanding and inappropriate pesticide use. They can be exposed to a variety of pesticides in their work. The common misuses include the use of larger volumes or concentrations of pesticide lead to the chance of high exposure. Normally, Thai farmer would like to create their own pesticide "cocktails" by mixing several pesticides together and add more than indicated on the label. The improper management and disposal of pesticides and a lack of awareness of pre-harvest intervals following application are also needed to concern (Panuwet et al., 2008; Quandt et al., 2010). Farmers generally use pesticides in the organophosphate group because of highly effective, less persistent and more biodegradable in the natural environment. Unfortunately, organophosphates lead many adverse health effects in human beings by inhibiting the function of the nervous system enzyme acetylcholinesterase (Jirachaiyabhas et al., 2004). The severity of pesticide exposure is related directly to a series of symptoms health effects (Lu, 2007).

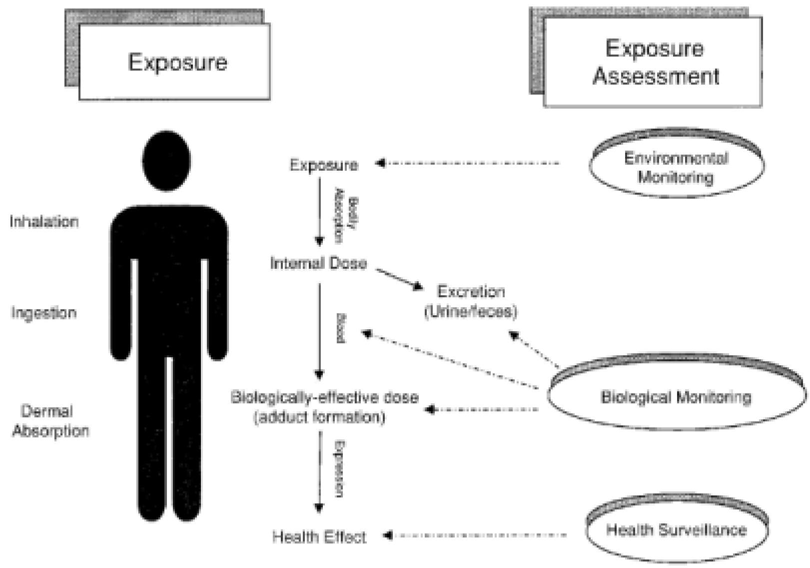


Figure 1.1 Schematic representation of the pathway of a toxicant from exposure to induction of health effects (Barr et al., 1999)

Human can be exposed to pesticides via multi-media and multi-route. Pesticide usage and environmental monitoring of exposure have to consider for all media and routes in order to calculate individual exposures. Measuring of exposures routes explaining as external dose, but, it may not estimate the absorbed dose. The internal dose, known as absorbed dose, of pesticide exposure is determined using biological monitoring. It includes the measurement metabolite, or a reaction product of a pesticide exposure biomarker such as human blood, urine, or tissues. Biological monitoring can determine whether an exposure has occurred and the body burden of the toxicant (Barr, 2008). The internal dose is an important for conducting health effects of exposure. The correlations between exposure and health effects are more precise if the dose assessment is conducted near the site of the induction of the health effect. Biological monitoring of pesticides is most often performed on the available body fluids to determine the internal dose resulting from pesticide exposure (Figure 1.1) (Barr et al., 1999). Thus, this study is going to figure out through all process from pesticide exposure till health outcomes from the exposure, starting from exposure assessment, biological monitoring, and health effects.

Chilli is a famous agricultural product of Thailand. It is normal ingredient in Thai food to give spicy taste. The large agricultural area growing chilli is settled in Northeastern of Thailand. Chilli is one of the crops using load of pesticides, especially Organophosphates pesticides. Hua rau sub-district, Muang district, Ubon Ratchahtani province, is a large area of agricultural. More than 70% of family in this area is farmers. Normally, farmers in this area do rice and chilli farms in each year. The chilli farm is started around December until March/ April in year round. After they harvest chilli products, the farmers continue rice crop in the same area. Unfortunately, chilli farm always apply pesticide excessively than another. There's a report showed that more than 80% of chilli-growing farmers in this area had low knowledge level to protect themselves from pesticide exposure and more than a half of the farmers didn't concerned about pesticide use and exposure (Norkaew et al., 2010). The risk assessment via dermal exposure (hands) of chilli-growing farmers in this area indicated that most farmers were not at risk from chlorpyrifos (organophosphate pesticide) exposure, nevertheless the farmers had mentioned on acute and repeated or prolonged effects of organophosphates after their application (Taneepanichskul et al., 2010). Another study of risk assessment from chilli consumption in this area demonstrated that the residue of profenofos (organophosphate pesticide) on chilli was higher than acceptable level explaining by hazard quotient (HQ > 1). Furthermore, the maximum level of chlorpyrifos and profenofos residues were higher than Reference dose (RfD), defined by US EPA, 45 and 110 times, respectively (Ooraikul et al., 2011).

1.2 Research Questions

1) Are chilli-growing farmers in Hua rau sub-district getting risk related to pesticides exposure via multi-exposure pathways?
2) Is there a relationship between biomonitoring and pesticide exposure routes in Chilli-growing farmers in Hua rau sub-district?
3) Is there an association between health effects and pesticides exposure in Chilli-growing farmers in Hua rau sub-district?

1.3 Research Objectives

The main objective of this study is to assess human exposure and health effect of common pesticides exposure through multi-exposure pathways: ingestion, inhalation and dermal contact of the farmers.

Specific Objectives

1) To identify health problems related to pesticide exposure
2) To study an effect of personal protective equipments on health.
3) To identify the main route of pesticide exposure of chilli-growing farmers by measuring concentration through multi-pathway.
4) To estimate pesticide exposure by biological monitoring (urinary metabolites).
5) To elaborate an association between biological monitoring and pesticide exposure routes.
6) To develop risk communication material of pesticide exposure protection for the chilli-growing farmers.

1.4 Research Hypothesis

1) Chilli-growing farmers in Hua rau sub-district are at risk of pesticides exposure from multi-exposure pathway.
2) There is a relationship between biomonitoring and pesticide exposure routes in Chilli-growing farmers in Hua rua sub-district.
3) There is an association between health effect and pesticide exposure in Chilli-growing farmers, Hua rua sub-district.

1.5 Conceptual Framework

INDEPENDENT VARIABLES

General Information
Individual information
-Gender
-Body weight (kg)
-Age (years)
-Height (Centimeters)
-Education
-Smoking
-Income
Agricultural works and farming descriptions
Handling and practicing of pesticides
Use of personal protective equipment (PPE)

Residue and Exposure Assessment:
Common pesticide use (Organophosphates)

Ingestion :
- Drinking water samples

Dermal contact :
- Wipe samples
- Patch samples
- Face Wipe

Inhalation:
- Personal air samples

DEPENDENT VARIABLES

Pesticide Contamination via Multi-pathways

Biological Monitoring
Urinary Metabolite Level

Health Effects from pesticides exposure

Risk Characterization

Risk Communication

Figure 1.2 Conceptual Frameworks

1.6 Operational Definition

Agricultural Health Surveillance

By the definition of "Agricultural health", it defines as the study of environmental, occupational, dietary, and genetic factors on the health of farmers, farm families, pesticide applicators, and others who work with and are exposed to agricultural chemicals. The World Health Organization defines "health surveillance" as the ongoing and systematic collection, analysis, interpretation, and dissemination of health information. Data surveillance will be generated initiatives provide a factual basis for the evaluation of intervention strategies, and for the development of rational public policies (Pickett et al., 2001)

In term of "Agricultural Health Surveillance" in this study defines as the process of systematic collection, analysis, interpretation, and dissemination of health information of farmers who expose directly to agricultural chemical and also provide a development of risk communication.

Farmer

In this study, farmer is a chilli-growing farmer whose age between 18 – 59 years old and living in Hua rau sub-district, Muang district, Ubon Ratchahtani province more than 5 years. He has to be pesticide applicator and engage all his farm activities, including mixing, loading and applying pesticide. Sickness farmers will be excluded from this study.

Common pesticides use

Common pesticide in this study concentrates on kinds of pesticide applying to chilli farm in this study area, especially Organophosphate. The pesticide has to apply to chilli farm normally, including use the same pesticides as previous.

Agricultural works and farming descriptions

This description in this part is related to the general information of agricultural and pesticide usage. The area, working years and tasks of farming are represented as the general information of agricultural. Pesticide usages include years of using pesticide, times of annual application and the equipment condition.

Handling and practicing of pesticides

The behavior of pesticide using in chilli farmer is a represent of handling and practicing of pesticide. The general behavior related daily pesticide applicator, such as the amount of pesticide use, storing place, safety consideration and pesticide exposure concern.

Use of personal protective equipment (PPE)

The use of personal protective equipment is defined as the cover which chilli-growing farmer use during the farm activities. It includes gloves, mask, goggles, boots, hat and coverall.

Residue of pesticides

The pesticide remaining on farmer’s body is defined as residue of pesticide in this study. The study tries to find out the concentration of pesticide on farmer’s hands, feet, face and body by using the specific method of analysis in laboratory.

Exposure assessment

Exposure assessment is an evaluation of the potential exposures to humans and the environment from the production, distribution, use, disposal and recycle of a chemical substance. The extent, duration, frequency and magnitude of exposures to a chemical (or multiple chemical) are estimated via various routes (ingestion, inhalation, dermal or transplacental/in utero exposure) for individuals or populations. Exposures can be estimated by measuring pollutant levels in various body tissues as biomarkers (WHO, 2008). In this study, exposure assessment is a process to estimate potential of pesticide exposure in chilli-growing farmer in study area via multi-route of exposure; inhalation, ingestion, dermal contact and biomarker.

Reasonable maximum exposure

The potential risk assessment in this study is based on the U.S. Environmental Protection Agency's (USEPA's) determination of what would result in an estimate of the reasonable maximum exposure (RME) expected to occur in this study area. The risk assessment procedure used in this study is likely to approximate the worst-case scenario defined as RME that is reasonably expected to occur at a site. RMEs are estimated for individual pathways. If a population is exposed via more than one pathway, the combination of exposures across pathways also must represent an RME (USEPA 1989 cited in Siriwong et al., 2010). Therefore, the upper confidence (95th percentile) on the arithmetic average concentrations was used to estimate the RME because the uncertainty associated with any estimate of exposure concentration might occur in this situation (Siriwong et al., 2010).

Drinking water

Drinking water in this study is a representative of pesticide contamination via chili-growing farmer ingestion route. The tank and/or cooler of drinking water which farmer takes to the farm and/or put in farm area are use as a sample. The drinking water at farmer's home is not including in this study.

Wipe samples

Wipe samples is a process of data collection. It defined as hands and feet wipe samples collecting for analyzing the contamination of pesticide residue on both hands and feet after a farmer finish his job task in farm.

Patch samples

In this study, patch sample is defined as a method to collect the residue of pesticide on farmer's body followed by WHO method. The standard patch will be put on 7 position of farmer's body during the farm activities.

Face Wipe samples

Face wipe sample is a procedure for collecting pesticide residues from workers' face during the agricultural activities. The sampling procedure in this study is modified from Dermal Face/ Neck wipe samples, agricultural handler exposure task force (Collier, 2009).

Personal air samples

In this study, personal air samples represent a inhale rate and pesticide concentration which the farmer inhale a contaminate air during farm activity. The measuring pesticide concentration via inhalation will be followed by NIOSH 5600 method (Organophosphorus pesticides, Issue 1, dated 15 August 1994).

Biological Monitoring

Biological Monitoring or biomonitoring is a common and useful tool for assessing human exposure to pesticides. Biological Monitoring involves the measurement of the parent pesticide, its metabolite or reaction product in biological media, typically blood or urine, to determine if an exposure has occurred and the extent of that exposure. Although not without its limitations, biological monitoring has great utility in integrating all routes of exposure allowing for one exposure measurement (Barr, 2008). In this study, biological monitoring is defined as urinary metabolite from organophosphate pesticide exposure during farm activities.

Urinary Metabolite Level

The urinary metabolite level in this study represents as the metabolite of organophosphate pesticides in urine which the specific metabolites are Dimethylphosphate[DMP], Dimethylthiophosphate [DMTP], Dimethyldithiophosphate [DMDTP], Diethylphosphate [DEP], Diethylthiophosphate [DETP] and Diethyldithiophosphate [DEDTP].

Health Effects from pesticides exposure

US EPA (Pennsylvania State University, 2009) defined the health effects of pesticides depend on the type of pesticide. Some, such as the organophosphates, affect the nervous system. Others may irritate the skin or eyes. Some pesticides may be carcinogens. Others may affect the hormone or endocrine system in the body. This study concentrates on acute health effect from pesticide exposure which refers to the chemical's ability to cause injury to a person or animal from a single exposure, generally of short duration. The harmful effects that occur from a single exposure by any route of entry are termed "acute effects" (Pennsylvania State University, 2009).

Risk Characterize from pesticide exposure

Risk characterization is the integration of the hazard identification, hazard characterization, especially dose-response, and exposure assessments to describe the nature and magnitude of the health risk in a given population. Once the risk characterization is completed, the results along with other information can then be used to develop priorities, strategies and program to protect those populations at risk (WHO, 2008). In this study, risk characterization use as a representative of defining the farmer's is at risk from pesticide exposure or not. The hazard quotient (HQ) and hazard index (HI) are explanation factors in this step.

Risk Communication

Risk communication was defined as ''an interactive process of exchange of information and opinion among individuals, groups and institutions. It involves multiple messages about the existence, nature, form, severity or acceptability of health risks'' (Tinker et al., 2000). Risk communication plan must be sound, with effective strategies, monitoring and evaluation to ensure the desired objectives are achieved. The planning requires expertise in various fields, such as program planning, evaluation, communications theory, and public health practice (Tinker et al., 2000). This study involves risk communication into the last step of research to give the information and provide the knowledge of pesticide exposure to the farmer for himself protection and reducing risk in this community. Risk communication is going to be a strategy for communicate between researcher and chilli-growing farmers for their understanding clearly.

CHAPTER II

LITERATURE REVIEWS

Pesticides, toxic chemicals, are introduced into the environment, especially agro-environment, for killing living organism, such as pest. Target species of pesticide are not only animals but also plants, bacteria and fungi. Due to human was considering as the larger species than target species, pesticides are applied on the qualities that not prove to be hazardous by our exposure. In fact, pesticides are definitely toxic to humans even in low doses. In our normal life, several sources of exposure could be posed to human accidental and intentional exposure. Pesticide can be present even in foods; drinking water affected our ordinal life. However, farmers are considered as the main pesticide exposure group by the application to their farm.

2.1 Pesticide use in Thailand

Intensive pesticide application has played an important role in Thai success in raising agricultural output to achieve food self-sufficiency and strong export growth since the 1970s. Heavily used on pesticide in Thailand seem to be a tool for increasing production level, quality of agro-product and its appearance, even though the usage together with misunderstanding of pesticide impact on human health and the environment, leads agricultural workers at their occupational hazard risks. The second common exposure a substance in Thailand was reported as “Organophosphate pesticide group”, nevertheless the trend of usage is continuously increased year by year (Issa et al., 2010; Ngowi et al., 2007; Panuwet et al., 2008, 2012; Posri et al., 2008; Wananukul et al., 2007).

The 11.5% of the country's Gross Domestic Product in the second quarter 2011, agriculture remains a highly significant despite the rise of industrialization. Pressures to sustain agricultural products have led to intense usage of pesticides. The quantities of imported agricultural pesticides have increased significantly from 1994 to 2005. Organophosphates still contributed the majority of imported pesticides since 1994, followed by Carbamates and Organochlorines (Taneepanichskul et al., 2012).

The Office of Agricultural Economics (OAE) and the Office of Agriculture Regulation (OAR) showed that pesticide use increased and imported into Thailand (Figure 2.1). Thus, it let Thailand ranked fourth out of 15 Asian countries in annual pesticide use and third in pesticide use per unit area (Panuwet et al., 2012)

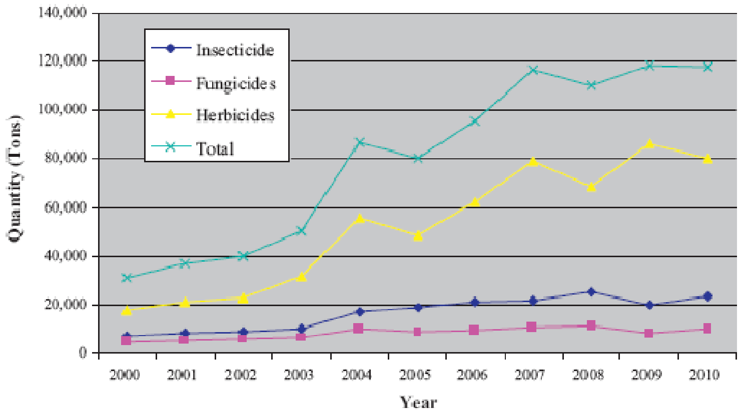


Figure 2.1 Summary of imported pesticide to Thailand 2000 - 2012 (Panuwet et al., 2012)

The liberal pesticide market in Thailand has resulted in the widespread availability and use of imported chemicals. Seventy-three percent of the agrichemical imports into Thailand are classified by the World Health Organization (WHO) as category IA (extremely hazardous), or IB (highly hazardous) (Kearns, 2008).

Thailand government has been encouraging farmers to switch to higher value horticultural crops. In 1992 all import duty on formulated pesticides was removed. As in many other countries, pesticides were supplied to them free or on credit. Widespread advertising also encouraged pesticide use, and public policies induced in farmers a belief that pesticides equated with 'modern' farming methods. The insecticides in most common usage are the extremely hazardous organophosphate pesticides. Even if farmers were aware of the hazards of some of these chemicals, it would be difficult for them to identify the active ingredients to avoid their use because pesticide is sold in Thailand under trade names (Dinham, 2003).

From human poisoning in Thailand during 2000 – 2004, reported by Ramathibodi Poison Center, pointed that pesticide poisoning was the major problem in Thailand. They were the most common poisons involved in human exposure, which counted for 41.5% of all cases of exposure. Among the pesticides, insecticides, herbicides poisoning were found to be 50.0% and 24.7% respectively. However, the unintentional occupational and accidental exposure to pesticides was only 8.6%. (Wananukul et al., 2007).

There are several reasons to be documented the problems of pesticides use in developing countries including Thailand as following (Dinham, 2003). Firstly, most farmers are never train in pesticide used and most information is depended on pesticide dealers. They believe that if they apply pesticide more than instructed. The result will be greater. Second they always mix different product because the effect is going to be better. In developing country especially Thailand some farmers are uneducated therefore they don't know about the chemicals name. They often use what is available rather than the exact pesticide for crop and pest. Moreover when they grow nontraditional crop, they can't separate between beneficial pests and pests. Third poor farmers, grower and agriculture doesn't have enough money to buy protective clothing and not separate work clothing from other. Furthermore they wear work clothing more than once without washing. While spraying pesticide and breaking, they commonly eat drink or smoke without washing their hands. Fourth pesticide containers are always kept in improper place such as left lying in fields, ditch and water courses. Fifth products label is always in unfamiliar language and instruction is always complex and difficult to understand so farmers may be use pesticide in the wrong ways. As a result of unclear instruction, application rate and timing, reentry periods after spaying, essential harvest intervals are unknown. Since inefficient using of pesticide on crop will lead to problem such as high application rates without any effect on pest levels, pest resistance and reduce productivity.

Due to the unsolved pesticide problems, a range of public as well as private schemes have emerged in response to heightened public concern. Thai environmental NGOs have been attempting to create alternative channels for sustainable agriculture, including organic farming, and have coordinated their efforts by establishing the Alternative Agriculture Network (Posri et al., 2008).

Pesticide exposure study in Thailand

The several studies related to pesticide exposure in Thailand had been reported. Summation of some study related to this study was reviewed and presented in Table 2.1.

Table 2.1 Summary of farmers' pesticides exposure studies in Thailand

References	Study population	Sample collection	Concentrated Pesticides	Results
Soogarun et al., 2003	35 male vegetable growers and 35 male references	70 blood samples	Organophosphates	A significantly lower level of serum cholinesterase among the vegetable growers, compared with the control group was found.
Jirachaiyabhas et al., 2004	33 farmers (traditional =18; integrate pest management= 15)	33 personal air samplers	Organophosphates	IPM farmers who use combinations of pesticides and non-chemical methods are at lower risk from being exposed to organophosphate pesticides during spraying compared with traditional farmers.
Thetkathuek et al., 2005	53 fruit farmers (age 16-60 yrs old)	106 blood samples (non-spray season =53; spray season =53)	Chlorpyrifos	Plasma cholinesterase (PChE) activity can be used as a biomarker for monitoring early toxicity induced by chlorpyrifos insecticide. Red blood cell cholinesterase (AChE) activities in nonspraying season and in the spraying season were not different.

References	Study population	Sample collection	Concentrated Pesticides	Results
Panuwet et al., 2008	136 male farmers (age 20-65 yrs old)	136 morning void urine samples	Organophosphates, pyrethroids and selected herbicides	DETP was the most detect frequency. Farmers, in difference topographically and crop, had different urinary metabolite level.
Siriwong et al., 2008	51 individuals consuming fish (age 10-75 yrs old)	15 of the most consumed fish species	Organochlorine pesticides (OCPs)	The residues of banned OCPs were detected at low concentrations, parts per billion (ppb) levels. The consumption of fish contributed to cancer risk by the calculated population cancer risk being greater than 1.0 (benchmark).
Jaipiam et al., 2009	33 vegetable growers and 17 references	100 personal air samplers (wet season = 50; dry season =50)	Organophosphates (chlorpyrifos, dicrotofos, and profenofos)	The concentration of pesticides in the vegetable growers was significantly higher than the references during both seasons. The vegetable growers may be at risk for acute adverse effects via the inhalation of chlorpyrifos and dicrotofos during pesticide application.

References	Study population	Sample collection	Concentrated Pesticides	Results
Jintana et al., 2009	90 pesticide applicators (age 18 – 65 yrs old) and 30 references subjects	210 blood samples (applicators = 180 [90 of high-exposed and 90 of low-exposed]; references = 30)	Organophosphates	Acetylcholinesterase (Ache) and Butyrylcholinesterase (BuChe) activities of low-exposure period were statistically significant decreased from high-exposure period. All enzyme activities in exposed group were statistically lower than in control group.
Kongtip et al., 2009	31 rice farmers	31 personal air samplers 31 blood samples	Chlorpyrifos	The chlorpyrifos exposure had high correlation with the levels of cholinesterase enzyme. The health risk of chlorpyrifos exposures through inhalation route was not acceptable by using Hazard Quotient evaluation (HQ>1).

References	Study population	Sample collection	Concentrated Pesticides	Results
Panuwet et al., 2009 (a)	136 farmers (age 18 – 65 yrs old) and 306 school children (age 10-15 yrs old)	449 urine samples	Methyl Parathion	More than 90% of all urine samples could be detected PNP (metabolite of Methyl Parathion and has been banned from Thailand). The positive correlation of PNP-DMP and PNP-DMTP was found.
Kachiyaphum et al., 2010	350 chilli-farm workers (age 18 – 60 yrs old)	350 reactive-paper finger blood test	Organophosphates and Carbamates	The high prevalence (32.6%) of abnormal SChE level was found among chilli-farms workers. The abnormal level was associated with gender, times and duration of spraying pesticide and pesticide s use behaviors.
Siriwong et al., 2010	30 fishermen	108 water samples	Organochlorine pesticides (OCPs)	The local fishermen might be particularly concerned about lifetime cancer risk associated with dermal contact based on reasonable maximum exposure.

References	Study population	Sample collection	Concentrated Pesticides	Results
Hanchenlash et al., 2012	8 vegetable farmers' families 8 fruit farmers' families (3 subjects were selected from each family composed of farmer, his/her spouse and child	144 urine samples (3 morning spot urine sample / subject)	N/A	Farmers' urinary metabolite was not correlated with their spouse or children collected at the same day. DAP level was moderately correlated with dermal exposure using a semi-quantitative observational method (DREAM)

2.2 Pesticide use in study area

Ubonratchathani Province, one of the largest agricultural areas, in the northeastern of Thailand, has produced fresh and dried chili products to support not only Thai residents but also export to some part of the world. Organophosphate pesticide, especially Chlorpyrifos and Profenofos, are widely used for controlling pest in chili crop. The most reported health effects related to pesticide poisoning is se verity of organophosphate pesticide. Inhibition of acetylcholinesterase (AChE) is generally accepted to be the most important acute toxic action of organophosphate compounds, leading to an accumulation of acetylcholine followed by dysfunction of cholinergic signaling (Ooraikul et al., 2011).

The evidence of pesticide exposure posed to health effects of adults including chili farmers themselves showed that more than 60 % of total population, getting involved in annual health check up survey by primary health care service, had abnormal acetylcholinesterase enzyme (AChE) level in blood in the last year. Unfortunately, the percentage of those is still increasing to around 90% in this year (Unpublished Data, Hua-rua Tambon Health Promoting Hospital, 2012) (Figure 2.2). The number projected to continuous using and exposing pesticide in the area. More than 80% of chili farmers had not much knowledge level to protect themselves and more than half of then didn't concern about pesticide exposure and usage (Norkaew et al., 2010). Moreover, the missing and strictly use of personal protective equipment (PPE) were detected once because of weather and humidity of Thailand. According to the Ethogram, that is not comfortable to the farmers to use PPE, therefore they are getting higher dose of exposure through multi-pathways during their job tasks, mixing, loading and applying pesticide.

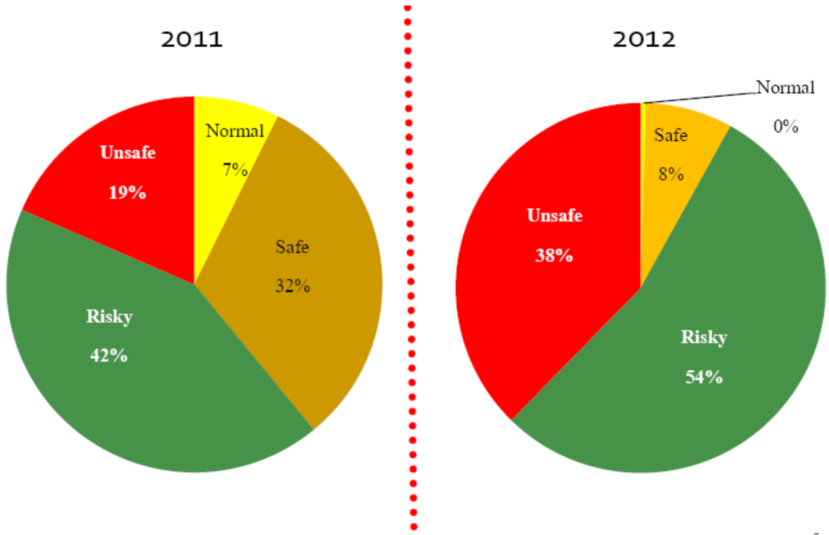


Figure 2.2 Cholinesterase Test using reactive paper during 2011 and 2012 (Unpublished, Hua-rua Tambon Health Promoting Hospital, 2012)

Pesticide exposure study in study area

Norkaew et al. (2010) assessed and evaluated the knowledge, attitudes, and practices on using personal protective equipment of chilli-growing farmers to protect themselves from pesticides and provided the recommendations and guidelines to reduce the farmers exposure to pesticides in Hua Rua sub-district, Muang district, Ubonratchathani Province, Thailand. A standardized questionnaire was used as measurement tools by face to face interviewing. There were 330 participated chilli-growing farmers. Of 71.2 % had educated in primary school. Most of them were applied pesticides by themselves. Nearly 90% of them recognized that they have to wear mask, boots, and cloth while spraying. Of 83.3% knew that pesticide can pass through their body. 45.5% of respondents knew that spraying should be done in the windward direction and they have to use PPE. Many of respondents commonly check equipment before using and wear clothing thoroughly while spraying. However, 77.2 % of chilli-growing farmers had low knowledge level, 54.5 % of the farmers' attitudes were not concerned about pesticide use and exposure and 85.0 % of farmers had fair practices level. The statistically significant association between knowledge and attitude, knowledge and practice, and attitude and practice were low positive correlation.

Ooraikul et al. (2011) conducted the study of human health risk assessment related to chili consumption during October 2010 to February 2011 in Hua Rua agricultural community at Hua Rua sub-district, Muang district, Ubon Ratchathani province. Socio-demographic and dietary survey were completed by face-to-face questionnaire among 110 local people. The study found that an average daily intake of chili was 0.018 kg/day which was higher than the average of general Thais (0.005 kg/day). Thirty-three chili samples were collected from farm after the day 7th of pesticides application and extracted using QuEChERS technique and quantified by gas chromatography equipped with flame photometric detector (FPD). Chlorpyrifos and profenofos were commonly detected in the range of < 0.010-1.303 mg/kg and 0.520-6.290 mg/kg, respectively. Moreover, a potential health risk characterization was indicated that risk characterization of chlorpyrifos did not exceed an acceptable risk ratio (hazard quotient; $HQ < 1.0$), but risk characterization of profenofos exceeded an acceptable risk ratio ($HQ > 1.0$).

Taneepanichskul et al. (2010) studied on risk assessment of Chlorpyrifos associated with dermal exposure in chilli-growing farmers during growing season from December 2009 to January 2010 at Hua-ruea sub-district, Muang district, Ubon Ratchathani province, Thailand. Chlorpyrifos residues on chilli-growing farmers' hands after spraying were collected using hand-wiping technique from 35 farmers. Hand surface areas of male and female were 0.088 m^2 and 0.075 m^2, respectively. The mean concentration (±SD) of chlorpyrifos analyzed by using gas chromatograph with a selective detector, flame photometric detector (FPD) was 6.95 ±18.24 mg/kg/two hands. To evaluate health risk of the chilli-growing farmers in this community, an Average Daily Dose (ADD) was calculated using reasonable maximum exposure (RME) at 95th percentile of chlorpyrifos concentration in order to concern health awareness and prevention. The ADD of farmers was 2.51×10^{-9} mg/kg/day and the ADD of male farmers (2.57×10^{-9} mg/kg/day) was higher than female farmers (2.41×10^{-9} mg/kg/day). Using hazard quotient (HQ) for risk characterization, it indicated that the HQ of farmers was lower than the acceptable level 1.0 ($HQ = 1.67 \times 10^{-6}$). Both of the HQ for male and female farmers were lower than the acceptable level, 1.71×10^{-6} and 1.61×10^{-6}, respectively.

Taneepanichskul et al. (2012) conducted a safety behaviour among the farmers, by gender, in Hua Rua sub-district Ubon Ratchathani Province. 35 randomly selected chilli-growing farmers had face-to-face interviews to investigate, general characteristics, frequency of spraying pesticide and pesticide safety behavior such as the pesticides used on their crops, use of personal protective equipment (PPE), cleaning gloves, pesticide container condition, and the place of changing and laundering work clothes and shoes. Most participants only rarely used protective equipment. 85.7% of them sprayed pesticide once per week. Nearly 80% did not wash or clean their gloves after application. The chilli-growing farmers in this area might be exposed to pesticides due to their pesticide using behavior which could contribute to increased health risk.

Taneepanichskul et al. (2012) investigated common pesticide used and health symptoms related to pesticide exposure among Chilli-growing farmers including an association between personal protective equipment (PPE) usage and health symptoms. The majority group of participants was men (65%) and had elementary level of education. Average income was around 1000 USD, approximately, with small growing area (2.05 ± 0.71 rai). Their handling and practicing of pesticide use were doubtful, with 20% storing pesticide at home without mixing them at their house and 95% using overdose of required amount of pesticide. PPE such as gloves, nose mask, boots, cover all (long sleeved shirts and long legged plants) were usually used during pesticide application. Some of the reported health symptoms due to pesticide exposure were irritation of the throat (40%), excessive salivation (65%), blurred vision (35%) and memory problem (70%). Some PPE usage related negatively to health problems were investigated in this study, such as skin problem versus wearing gloves (R= - 0.612**) and headache versus nose mask (R = - 0.745**). It was suggested that regular public health education training programs including how to use appropriated PPE should be organized for the Chilli growing farmers to improve their ability to handle pesticide.

2.3 Organophosphate pesticides (OPPs)

Organophosphates (OPs) are a large class of chemicals. An estimated several thousand OPs have been synthesized for various purposes since World War II. The majority of OPs are used as pesticides. The structures within class of OPs are similar although there are different compounds. Normally, they have a phosphorus atom and a characteristic phosphoryl bond (P=O) or thiophosphoryl bond (P=S) (Figure 2.3). OPs are esters of phosphoric acid with varying combinations of oxygen, carbon, sulfur, or nitrogen attached. Complexity in classification of OPs arises due to different side chains attached to the phosphorus atom and the position where the side chains are attached. Some OPs effect anticholinesterase activity, whereas others effect little or no anticholinesterase activity or need desulfuration to the analogous oxon before acquiring anticholinesterase activity (Gupta, 2006; Bleecker, 2008).

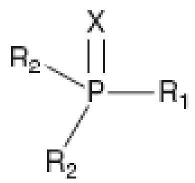


Figure 2.3 General structures of organophosphates which X is O or S

Chlorpyrifos

Chlorpyrifos (Figure 2.4) has been used as pesticides or ingredient of pesticide products to spray on the farm to control crop pests. It may also be applied to crops in a microencapsulated form. According to EPA, tolerances of Chlorpyrifos defined as raw agricultural commodities, foods, and animal feeds.

In the environment phase, Chlorpyrifos enters through volatilization, spills, and the disposal of chlorpyrifos waste. Volatilization is the major way when chlorpyrifos disperses into environment. Generally, Chlorpyrifos is broken down by sunlight, bacteria, or other chemical processes.

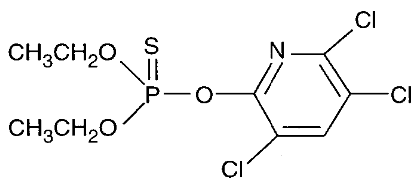


Figure 2.4 Chlorpyrifos Structure [O,O-diethyl- O-3,5,6-trichloro-2-pyridyl phosphorothioate]

The technical form (Table 2.2) is a white crystal-like solid with a strong odor. It does not mix well with water, so it is usually mixed with oily liquids before it is applied to crops (Cattani, 2004).

Table 2.2 Chlorpyrifos Properties

Chemical name	O,O-diethyl-O-3,5,6-trichloro-2-pyridyl phosphorothioate
Molecular weight	350.6
Empirical and structure formula	C9H11C13NO3PS
State	Crystalline solid
Color	White to tan
Odor	Mild mercaptan
Melting point	41.5 – 42.5 ^{o}C
Boiling point	>300 ^{o}C
Vapor pressure	3.35 mPa at 25 ^{o}C
Density	1.51 g/ml at 21 ^{o}C
Solubility, mean	Acetone >400g/l at 20 ^{o}C Dichloromethane > 400g/l at 20 ^{o}C Ethyl acetate >400g/l at 20 ^{o}C Methanol 250g/100ml at 20 ^{o}C Toluene >400g/l at 20 ^{o}C n-Hexane >400g/l at 20 ^{o}C Water 1.05ppm (W/V at 25^{o}C)

Profenofos

Profenofos (Figure 2.5) was first registered by the Agency in 1982 for use as an insecticide/miticide. This interim reregistration eligibility review is the Agency's first reevaluation of profenofos since its initial registration in 1982.

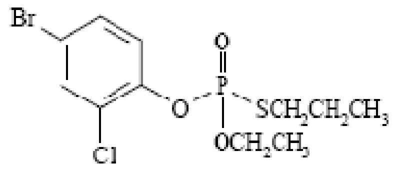


Figure 2.5 Profenofos Structure [O-(4-bromo-2-chlorophenyl)-O-ethyl-S-propyl phosphorothioate

Technical profenofos is a pale yellow liquid with a boiling point of 100°C (1.8 Pa) and a density of 1.46 g/cm^3 at 20 °C. Pure profenofos is an amber-colored oily liquid with a boiling point of 110 °C (0.13 Pa). Profenofos has limited solubility in water (20 ppm), but is completely soluble in organic solvents (ethanol, acetone, toluene, n-octanol, and n-hexane) at 25°C (US EPA, 2006).

Mechanism of OPs to health effect

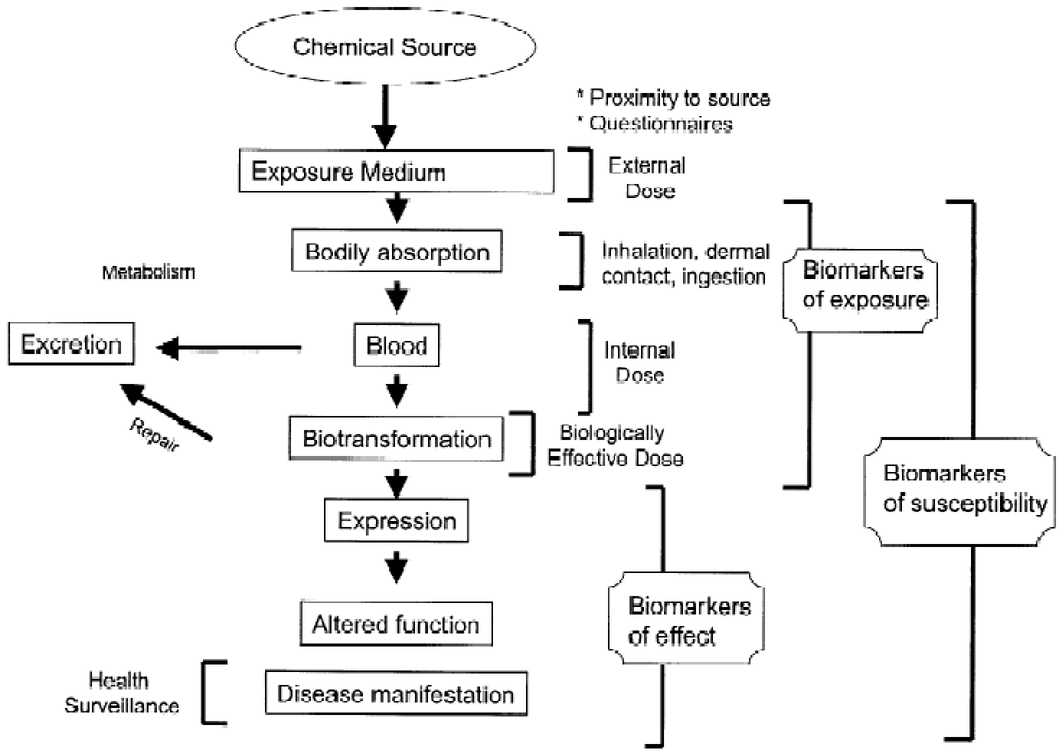
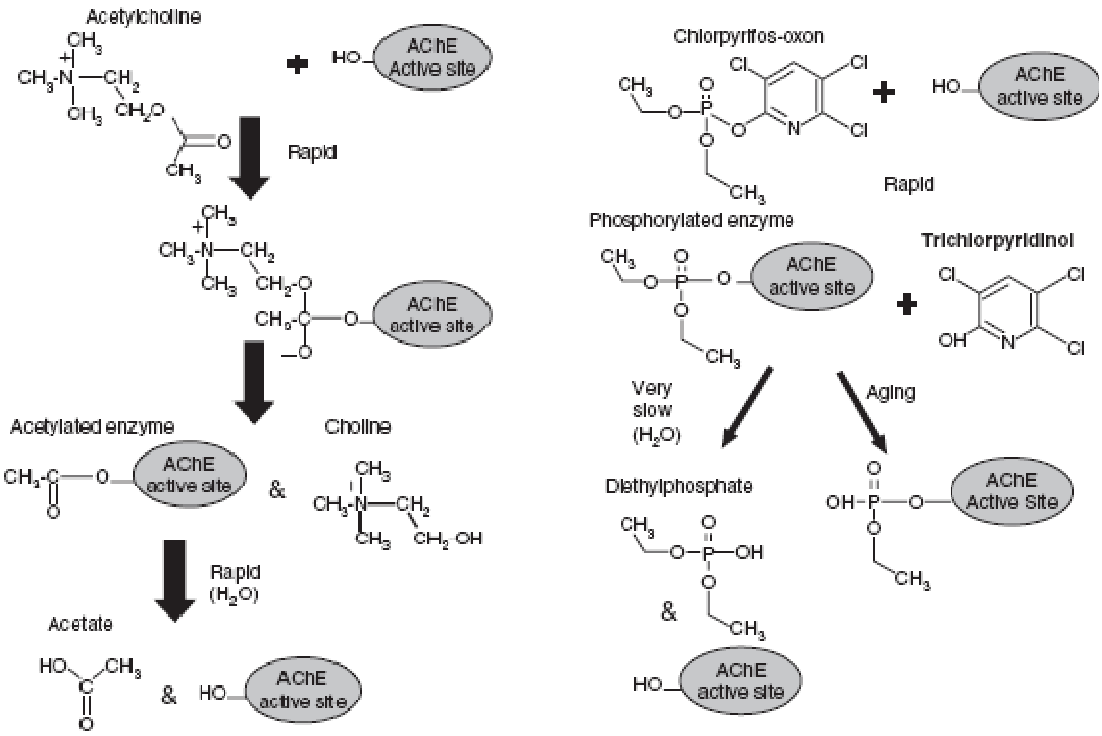


Figure 2.6 The pathway of a toxicant from exposure to induction of health effects (Adapted from Barr and Needham,2002)

Organophosphates is the most commonly cause systemic illness. Acute severe organophosphate poisoning is one of the most life-threatening human poisonings, but it is also treatable, often with a good outcome if treatment is begun promptly and early in the time course of poisoning (Snodgrass, 2010). Diagnosis of acute organophosphate poisoning is based on a history of exposure, clinical symptoms and signs, and, where available, a blood test of red cell cholinesterase and plasma pseudocholinesterase (Figure 2.6). List symptoms and signs of cholinesterase-inhibitor poisoning are shown in Table 2.3. Exposure to organophosphates may produce a broad spectrum of clinical adverse effects. These adverse effects may present clinically as headache, weakness, dizziness, blurred vision, psychosis, respiratory (pulmonary) difficulty, paralysis, convulsions, and coma. The onset of clinical manifestations of organophosphate poisoning usually occurs within 12 h of exposure.

The acute toxicity of OP is explained by irreversible inhibition of AChE activity at cholinergic synapses. The function of acetylcholinesterase (AChE) is to degrade the neurotransmitter acetylcholine (ACh). Inhibition of AChE (>70%) leads to accumulation of ACh at central and peripheral sites. OP share a common mode of insecticidal and toxicological action associated with their ability to inhibit the enzyme AChE within nerve tissue and at the neuromuscular junctions. In the brain, overstimulation of ACh receptors can lead to seizures. Inhibition of the breathing center in the brain results in asphyxiation. In the diaphragm muscle, overstimulation of nicotinic receptors leads to desensitization and paralysis of the breathing muscles. In the lung, overstimulation of receptors causes vast amounts of fluid to be secreted so that a person drowns in his or her own fluids. Heart rate is increased by sympathetic stimulation and decreased by parasympathetic stimulation. Depending on the relative effects on the two branches, OP compounds may produce tachycardia, bradycardia, fibrillation, or cardiac arrest. The comparison of AChE inhibition dynamics for the interaction of ACh between chlorpyrifos-oxon (OP) and Acetylcholine (AChE) is shown in Figure 2.7.



Interaction of acetylcholine   Interaction of OP chlorpyrifos-oxon

Figure 2.7 Interaction of acetylcholine and the OP chlorpyrifos-oxon (III) with the active site of AChE (Adapted from Gupta, 2007)

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Table 2.3 List symptoms and signs of cholinesterase-inhibitor poisoning

Exposure	Symptoms
Mild	Anorexia
	Headache
	Dizziness
	Weakness
	Anxiety
	Tremors of tongue and eyelids
	Miosis
Moderate	Nausea
	Vomiting
	Salivation
	Tearing
	Abdominal cramps
	Diaphoresis
	Bradycardia
	Muscular fasciculations
Severe	Diarrhea
	Pinpoint and nonreactive pupils
	Pulmonary/ventilatory difficulty
	Pulmonary edema
	Cyanosis
	Loss of sphincter control
	Heart block
	Convulsions
	Coma, possible death

2.4 Analysis of Organophosphate pesticides

Organophosphate (OP) pesticides are composed of a phosphate (or thio- or dithiophosphate) moiety and an organic moiety. The phosphate moiety is mostly O,O-dialkyl substituted. Cholinesterase inhibitors are effective by these pesticides. They can reversibly or irreversibly bind covalently with the serine residue in the active site of acetyl cholinesterase and prevent its natural function in catabolism of neurotransmitters. This action is not only effect to insects, but can also produce the same effects in wildlife and humans.

Once human exposure occurs, OP insecticides are usually metabolized to the more reactive oxon form which may bind to cholinesterase or be hydrolyzed to a dialkyl phosphate and a hydroxylated organic moiety specific to the pesticide. The organic portion of the molecule is released as a result of binding to cholinesterase. The cholinesterase-bound phosphate group may be “aged” by the loss of the O,O-dialkyl groups, or may be hydrolyzed to regenerate the active enzyme. These metabolites and hydrolysis products are then excreted in the urine, either in free form or bound to sugars or sulfates. The intact pesticide may undergo hydrolysis prior to any conversion to the oxon form and the polar metabolites are excreted.

Six dialkyl phosphate (DAP) metabolites of OP pesticides are the most commonly measured in urine. These methods use liquid–liquid extraction with polar solvents such as ethyl acetate or diethyl ether, cyclohexyl solid-phase extraction, azeotropic distillation, or lyopholization to isolate the DAPs from the urine matrix. The DAPs are derivatized using pentafluorobenzyl bromide (PFB). The derivatized extracts are analyzed using GC connected with flame photometric detection (FPD), flame ionization detection (FID), mass spectrometry and or tandem mass spectrometry. The data generated from these analyses do not provide unequivocal identification of a single pesticide, but rather a cumulative index of exposure to most of the members of the class of OPs. However, it is generally believed that OP exposure or exposure to OP hydrolysis products related to most urinary DAP. (Barr and Needham, 2002)

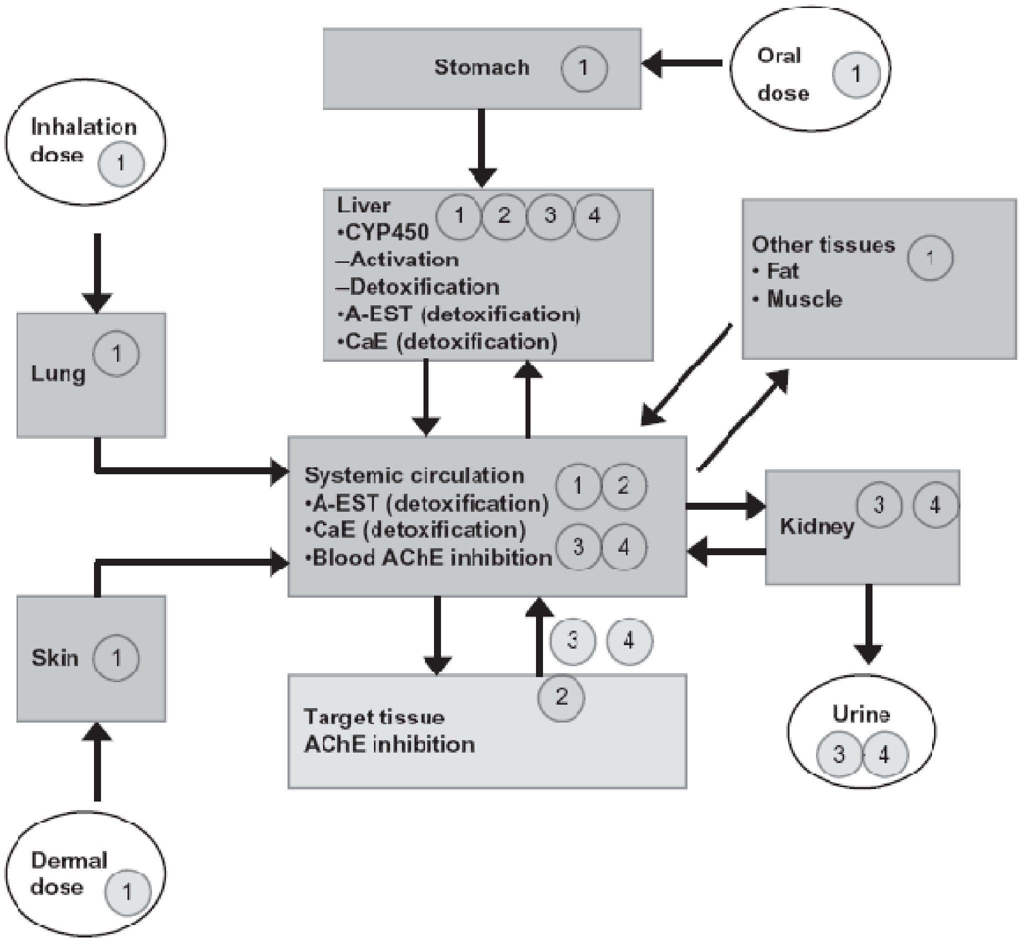


Figure 2.8 Diagram of the absorption, distribution, metabolism, and excretion of organophosphorus (OP) insecticides illustrating the critical tissue compartments (Timchalk, 2010).

Metabolism of Chlorpyrifos (Testai et al., 2010).

Chlorpyrifos is a weak acetylcholinesterase (AChE) inhibitor but it can be desulfurated by several isoenzymes to form the phosphate triester which is a powerful inhibitor of brain and serum AChE. The major site of CPF metabolism is the liver however, extrahepatic metabolism has been reported in other tissue as brain and intestine.

The major elimination pathway of Chlorpyrifos metabolized is urinary excretion, with TCP together with DEP, DETP, GSH conjugates, sulfates. The half-life for elimination of Chlorpyrifos from the various organs in rats is 10–16 hrs. , except for elimination from fat, which was estimated to be 62 hrs. The elimination half-life in humans has been estimated to be 27 h, with the maximum rate of TCP excretion occurring 24–48 h following dermal exposure.

The major pathways of Chlorpyrifos metabolism are shown in Figure 2.10. They include:

1. Oxidative desulfuration of the P=S moiety to P=O, catalyzed by cytochrome P450 (CYP), resulting in the toxic intermediate CPFO (bioactivation)
2. Dearylation catalyzed by CYP, resulting in 3,5,6-trichloro-2-pyridinol (TCP) and diethyl thiophosphat (DETP) (detoxification)
3. Hydrolysis by A-esterases (paraoxonases-PON1) of the phosphate ester bonds of CPFO to form TCP and diethylphosphate (DEP) (detoxification)
4. Hydrolysis by B-esterases as aliesterases (carboxylesterase-CE) and cholinesterase (BuChE), acting as molecular scavengers by binding stoichiometrically to CPFO (detoxification)
5. Conjugation of CPFO by glutathione-S-tranferases with reduced glutathione (GSH) (detoxification)
6. Conjugation of TCP by glucuronil-transferases and sulfotransferases to form the corresponding glucuronide and sulfate conjugates (detoxification)

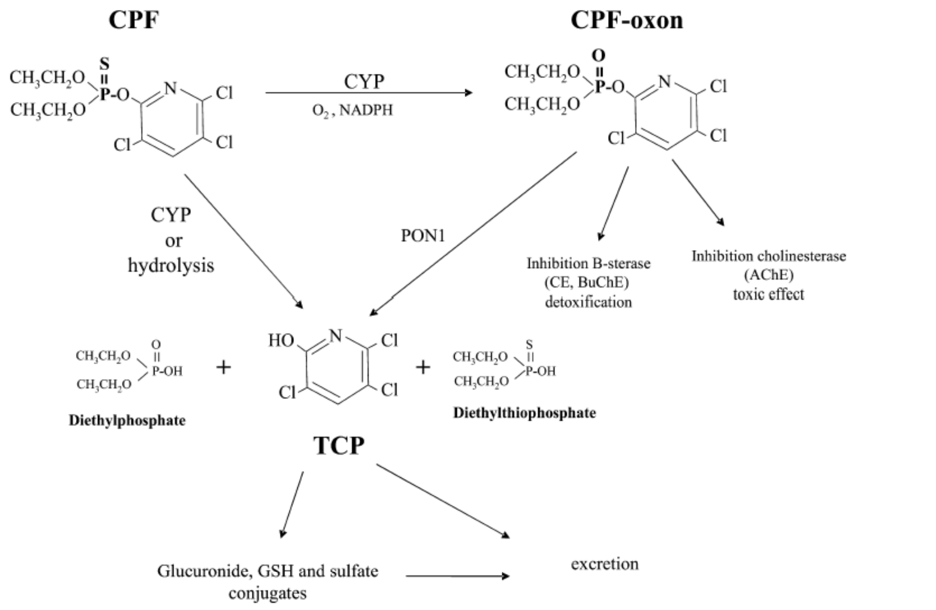


Figure 2.9 The major pathways of Chlorpyrifos metabolism (Adapted from Testai et al., 2010)

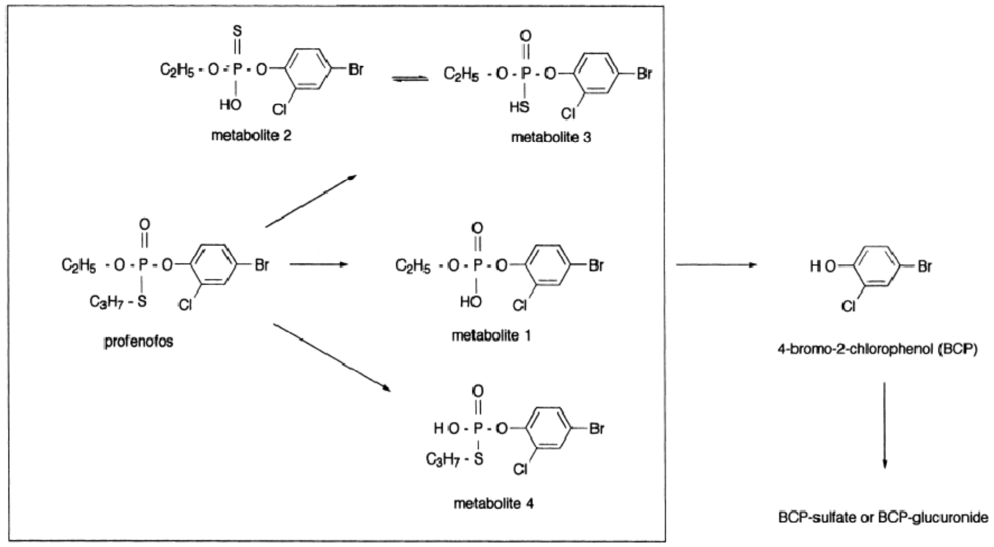


Figure 2.10 Metabolic pathways of profenofos in humans (Adapted from Gotoh et al., 2001)

2.5 Pesticide direct exposure

Occupational exposure to pesticides in agriculture and public health applications may cause acute and long-term health effects. Human exposures to pesticides may occur during worker contact relating the dermal and inhalation exposure routes mainly. Worker populations that are routinely exposed to pesticides include agricultural handlers involved in treatment of field crops and field workers. In many cases there are two distinct operations in the application of pesticide products. These operations are the mixing/loading that involves handling the concentrated product and diluting it in preparation for application, and the actual application of the diluted spray solution to the intended target. The risk assessment process enables regulatory agencies and the agrochemical industry to predict the extent of risk of adverse human health effects associated with the use of a given pesticide under specific use conditions. Because the evaluation of risk requires knowledge of both exposure and toxicity, exposures to the active ingredient associated with a given pesticide formulation must be assessed (Lunchick et al., 2010).

Pesticide poisoning among humans generally occurs either because of lack of compliance with existing pesticide regulations or because existing pesticide regulations are insufficient. The first cause involves cases that are preventable by following the precautionary measures specified on product labels and in governmental pesticide regulations. The appropriate interventions for these cases include enhanced education and enforcement. The second cause arises despite compliance with label instructions and regulatory measures and therefore requires interventions aimed at changing pesticide use practices and/or modifying regulatory measures (Calvert et al., 2001).

Occupational pesticide exposure studies

Exposure Assessment Studies

Curwin et al. (2005) studied on 24 farmers (cases) and 23 non-farmers (controls) pesticide exposure in the spring and summer of 2001. Hand wipe sample and an evening and morning urine sample were collected from each participant. They were analyzed for the parent compound or metabolites of six commonly used agricultural pesticides: alachlor, atrazine, acetochlor, metolachlor, 2,4-dichlorophenoxyacetic acid (2,4-D) and chlorpyrifos. Farmers, applying the pesticide, had urinary metabolite levels significantly higher than nonfarmers, farmers non-apply the pesticide, and farmers who had the pesticide commercially applied for atrazine, acetochlor, metolachlor and 2,4-D. For 2,4-D pesticide, association between time since application, amount of pesticide applied, and the number of acres was reported in this study. Farmers who reported using a closed cab to apply these pesticides had higher urinary pesticide metabolite levels, although the difference was not statistically significant. Most hand wipe samples were non-detectable, however, detection of atrazine in the hand wipes was significantly associated with urinary levels of atrazine above the median.

Arcury et al. (2010) conducted a study on 196 farm workers due to the limited data of repeated pesticide absorption in an agricultural season. In 2007, 4 times at monthly intervals urine samples were collected from all participants in order to test for 12 pesticide urinary metabolites. Measurement of exposure risk was done by questionnaire data. The results showed that housing type had a significant association with metabolite detections. Most farm workers were exposed to an array of pesticides across an agricultural season and that many farm workers are repeatedly exposed to the same pesticides across an agricultural season. At least one urine sample collected from the great majority of farm workers contained the pesticide metabolites associated with the OP insecticides. All farm workers had detections for at least four different urinary metabolites. At least seven different urinary metabolites were found in most of farm workers in this study, especially OP pesticide urinary metabolites.

Blair et al. (2011) assessed the impact of exposure misclassification on relative risks using the range of correlation coefficients observed between measured post-application urinary levels of 2,4- dichlorophenoxyacetic acid (2,4-D) and a chlorpyrifos metabolite and exposure estimates from 83 pesticide applications. The results found that the correlations between urinary levels of 2,4-D and a chlorpyrifos metabolite and algorithm estimated intensity scores were about 0.4, for 2,4-D, 0.8, for liquid chlorpyrifos and 0.6 for granular chlorpyrifos. Correlations of urinary levels with kilograms of active ingredient used, duration of application, or number of acres treated were lower and ranged from 0.36 to 0.19.

Pesticide exposure health affects Studies

Lu (2007) investigated illnesses related to pesticide exposure among cutflower farmers in La Trinidad, Benguet. The study used personal physical health, laboratory examinations and questionnaire on work practices and illness as a measurement. Majority of exposed farmers, male gender, were symptomatic, with most common complaints being headache (48%), easy fatigability (46.1%) and cough (40.2%). An analysis showed that RBC cholinesterase levels were positively associated with age, selling pesticide containers, number of years of using pesticides, use of contaminated cloth, incorrect mixing of pesticides and illness due to pesticides Significant associations were also found for hemoglobin, hematocrit, RBC, white blood cell and platelet count.

Mitoko et al. (2000) study on assessing health hazards related to handling, storage, and use of pesticides, on agricultural estates and small farms. The 256 exposed subjects and 152 controls were completed questionnaire on symptoms experienced at the time of interview including sex, age, main occupation, and level of education. Symptoms on health effects of pesticides that inhibit cholinesterase (organophosphate and carbamate) reported during the high exposure period. Symptom prevalence in exposed subjects was higher during the high exposure period than the low exposure period, however no statistical significant was found. A clear and significant change in symptoms prevalence was found in the controls with a higher prevalence in the low exposure period. The relation between cholinesterase inhibition and symptoms showed that prevalence ratios were significantly >1 for respiratory, eye, and central nervous system symptoms for workers with >30% inhibition.

Zhang et al. (2011) investigated the prevalence and risk factors of acute work-related pesticide poisoning among 80 pesticide applicators from two villages in Southern China using face to face interview. Respondents who self-reported having two or more of a list of sixty-six symptoms within 24 hours after pesticide application were categorized as having suffered acute pesticide poisoning. A multivariate logistic model was use to assess the association between the composite behavioral risk score and pesticide poisoning. The results showed that. The most frequent symptoms among applicators were dermal and nervous system symptoms. After controlling for gender, age, education, geographic area and the behavioral risk score, farmers without safety training had an adjusted odds ratio of 3.22 (95% CI: 1.86-5.60). A significant “dose-response” relationship between composite behavioral risk scores calculated from 9 pesticides exposure risk behaviors and the log odds of pesticide poisoning prevalence was seen among these Chinese farmers.

Strong et al. (2004) studied on 211 farmworkers in Eastern Washington to assess the relationship between self-reported health symptoms and indicators of exposure to OP pesticides. The diagnosis of health symptoms most commonly reported included headaches (50%), burning eyes (39%), pain in muscles, joints, or bones (35%), a rash or itchy skin (25%), and blurred vision (23%). The proportion of detectable samples of various pesticide residues in house and vehicle dust was weakly associated with reporting certain health symptoms, particularly burning eyes and shortness of breath. However, no significant associations were found between reporting health symptoms and the proportion of detectable urinary pesticide metabolites.

2.6 Route of pesticide exposure

Exposure, described in terms of the magnitude, frequency, and duration of contact , including the chemistry and physical properties of the pesticide (Ballantyne and Salem, 2006), is the contact of an individual with a contaminant for a specific duration of time. For exposure to occur the contaminant and the individual must come together in both space and time. Exposure science characterizes and predicts this intersection (Sheldon, 2010).

Agricultural workers involved in the use of pesticides and post-application crop re-entry activities may be exposed to pesticides via the skin, by inhalation, or by accidental oral ingestion. Exposure is usually greatest by the dermal route, although inhalation can be an important route for pesticides that have significant vapor pressures, are applied in confined spaces, or have an application technique that generates a significant proportion of inhalable particles. The inhalation and dermal exposures received from mixing, loading, and applying the pesticide in the field, dietary intake and ambient air were included as potential sources for exposure to this pesticide (Chester, 2010) (Figure 2.11).

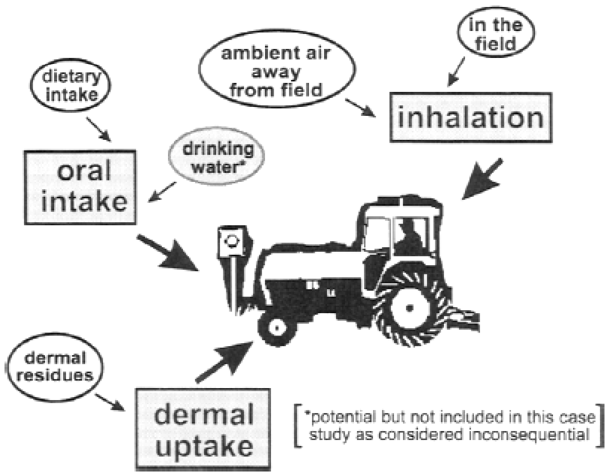


Figure 2.11 Multi exposure pathways via multimedia for the agricultural handler who has mixed, loaded, and applied a pesticide in a field (Adapted Dong et al., 2001)

Dermal Exposure

In practice, two measurements or estimations are usually made for all work activities associated with the use of pesticides:

1. Potential dermal exposure: the total amount of pesticide coming into contact with the protective clothing, work clothing, and skin.
2. Actual dermal exposure: the amount of pesticide coming into contact with the bare (uncovered) skin and the fraction transferring through protective and work clothing or via seams to the underlying skin, which is therefore available for absorption.

The biological availability or absorption of a pesticide via the dermal route of exposure is a property of the formulated product and the diluted material and is a separate subject in its own right. Given the significance of the dermal route, precise determinations of percutaneous absorption are key components of the overall assessment of the absorbed dose of the pesticide for risk assessment (Chester, 2010).

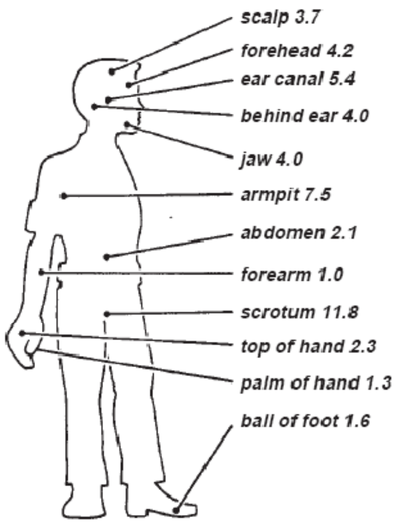


Figure 2.12 Rates of absorption through the skin are different for different parts of the body. (Adapted from: Johnson et al., 1999)

Inhalation Exposure

Setting aside volatile pesticides for the moment, the only spray droplets or particles that pose a potential risk comprise called inhalable or inspirable fraction, which is the mass fraction of airborne particulate capable of entering the respiratory tract via the nose and the mouth, so providing a source of absorption into the body, either from direct inhalation or from subsequent oral absorption. This is considered to be the most important indicator of potential inhalation exposure. The inhalable fraction depends on the speed and direction of the air movement, on the rate of breathing, and on other factors.

In risk assessment it is common to assume that volatile airborne pesticides are completely retained and absorbed via the respiratory tract, unless there are specific data to demonstrate otherwise.

Inhalation exposure is usually a small fraction of the total exposure and can, in some cases, be ignored, for example, the mixing and loading of liquid formulations, particularly if a closed loading system is involved. Conditions under which exposure by the inhalation route becomes important usually involve the use of volatile pesticides or of dusts, fumigants, and sprays, especially in enclosed spaces. It should, however, be borne in mind that a higher proportion of the inhaled dose may be retained systemically, compared with the proportion absorbed after dermal exposure, which could be as low as 1% or less of the available dermal dose (Chester, 2010).

Oral Exposure

Some of the larger airborne particulates may be trapped in the mouth or nasal passages and subjected to oral ingestion. Some of the exposure, which is measured as inhalation, might indeed be trapped and absorbed in this way.

No serious attempts have been made to measure separately the amount of exposure by this route because of the obvious difficulties involved. Biological monitoring takes into account all routes of absorption, but it is usually unable to distinguish between their relative contributions (Chester, 2010).

2.7 Risk Assessment

Risk assessment is a tool to predict the possibility of adverse effects to man and to identify the need of preventive actions. Risk assessment allows itself determining; the magnitude of the adverse effect posed by the pesticide product (Figure 2.13); the dose-response assessment to estimation of the relationship between dose and incidence and severity of the effect; the extent of exposure for measurement or prediction of doses to which humans are exposed; and the characterization of risk to express of adverse effects due to actual or predicted circumstances of exposure, and the nature and severity of such effects. (Maroni et al., 1999).

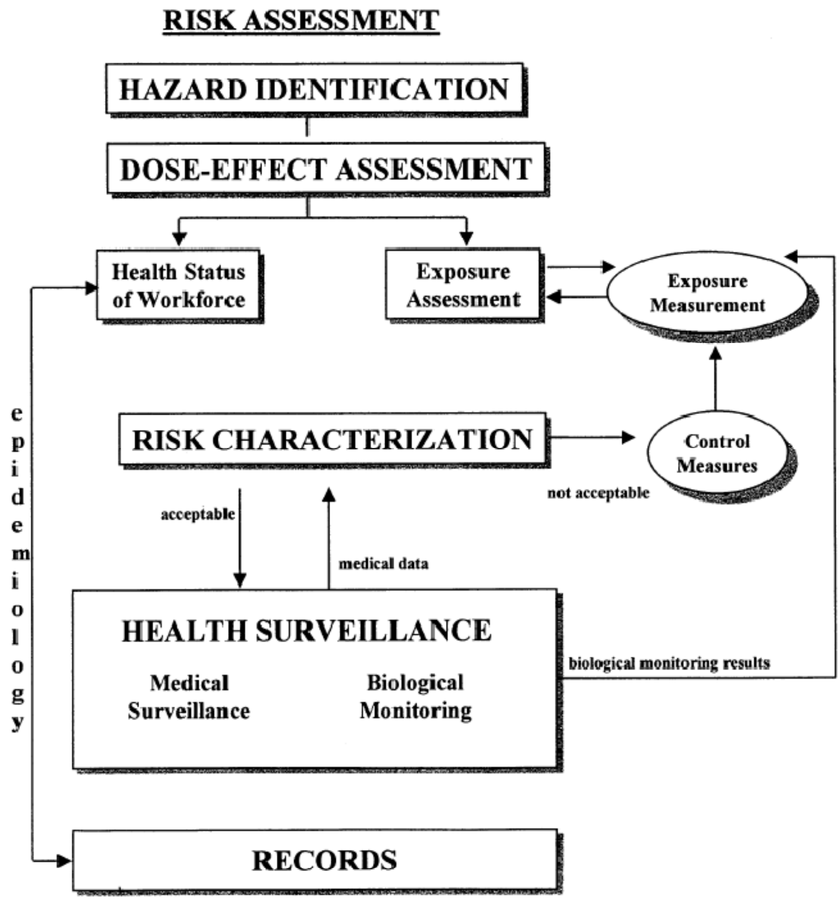


Figure 2.13 Health risk assessment **(**Adapted from Maroni et al., 1999**)**

Hazard Identification

Hazard identification refers to the potential health effects occurring from different types of pesticide exposure. It is strongly related to the extent and the type of a pesticide’s toxic properties. It is important to know whether the adverse effects observed in one species will occur in other species. Hazard identification may also involve characterizing the behavior of a pesticide or metabolite within the human body and chemical interactions within organs, cells, or even parts of cells. US EPA considers the following toxicity tests for hazard identification: (i) acute testing; (ii) sub-chronic testing; (iii) chronic toxicity testing; (iv) developmental and reproductive testing; (v) mutagenicity testing: assess the potential of a pesticide to affect the genetic components of the cell and (vi) hormone disruption (Frenich, et al., 2007).

Dose Respond Relationship

The dose–response assessment considers the degree or incidence of effects that occur, or are predicted to occur, at a given dose level including the amount of a substance a person is exposed to. Dose–response assessment considers the dose levels at which adverse effects are observed in test animals, and using these dose levels to calculate a corresponding dose for humans (Frenich, et al., 2007).

Exposure Assessment

Exposure assessment is the process for identifying potentially exposed populations and quantifying exposures. Exposure assessments search for characterizing "real-life" situations whereby potentially exposed populations are identified, potential pathways of exposure are identified, and the degree, frequency, and duration of chemical intakes or potential doses are quantified.

Exposure assessments may be conducted using direct and indirect approaches. Direct assessments measure the contact of the person with the chemical concentration in the exposure media over an identified period of time. There are very few cases where methods exist and are used to make direct exposure assessments. Personal monitoring techniques such as the collection of personal air or duplicate diet samples are used to measure exposure directly to an individual during a point in time.

Indirect assessments use available information on concentrations of chemicals in exposure media, along with information about when, where, and how individuals might contact the exposure media. The indirect approach then uses models and a series of exposure factors such as pollutant concentration, contact duration, contact frequency, to estimate exposure.

For a few pesticides, biomarkers can serve as a useful measure of direct exposure aggregated over all sources and pathways. It should be understood that biomarkers will measure integrated exposure from all routes. However, to use biomarkers for this purpose, several important criteria must be met. Biomarkers that can accurately quantitative the concentration of a pesticide or its metabolite in easily accessible biological media must be identified and available. The pharmacokinetics of absorption, metabolism, and excretion must be known. Time between pesticide exposure and biomarker sample collection must be known. Although there are a number of biomarkers that meet these criteria, very few studies using biomarkers have collected all of the information required to accurately estimate exposure (Sheldon, 2010).

Risk Characterization

Risk characterization integrates data from hazard identification, dose–response and exposure assessments to describe the overall risk from a pesticide. It develops a qualitative or quantitative estimate of the likelihood that any of the hazards associated with the pesticide will occur in exposed people. It also involves the assumptions used in assessing exposure as well as the uncertainties that are built into the dose–response assessment (Frenich, et al., 2007).

CHAPTER III

METHODOLOGY

3.1 Study Design

This study was figure out the health surveillance of chilli-growing farmers living in Muang district, Ubon Ratchathani Province, Thailand. The study design was "cross-sectional study design" because the information about health surveillance of pesticides exposure in this study was represented as it was going on only one point in time in the highest dose application (Olsen et al., 2004).

Sample was collected during March - April 2012 because farmers were use the highest dose of pesticides in this period of study. Normally, chilli crop was covered between October – May of each year. Most of pesticides were applied to the farms in the middle of the crop because the chilli-farmers were going to do the first harvest.

3.2 Study Area

The study area was located in the northeastern of Thailand, Hua-rua sub-district Muang district, Ubon Ratchathani Province. Hua-rua sub-district composed of 16 villages; however, there were only 3 villages (Figure 3.1) involving in this study because these villages were growing chilli during data collection and sampling.

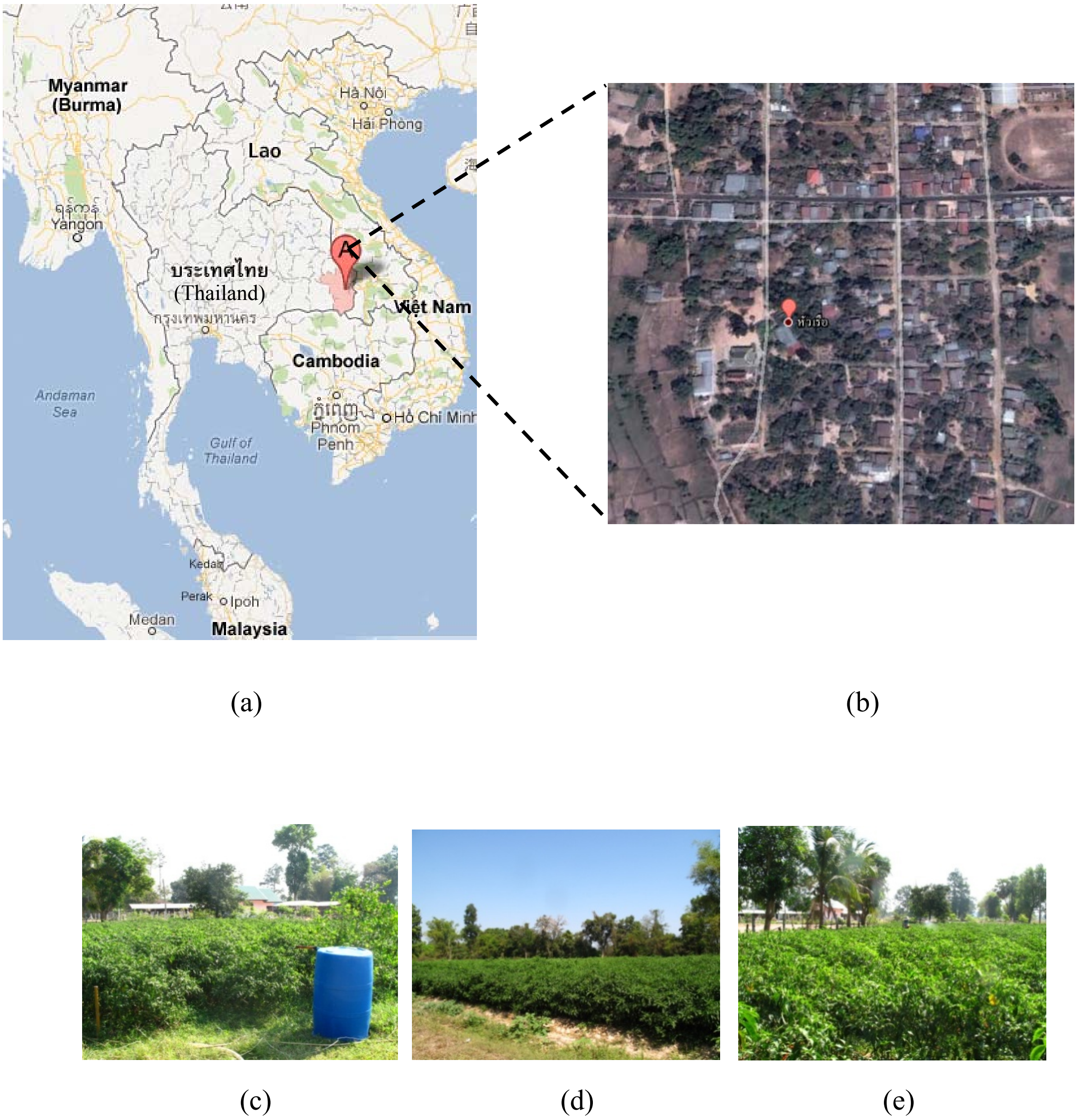


Figure 3.1 Topography map of the study area, Hua-rua sub-district, Muang district, Ubon Rachathani province, Thailand (a), (b) and chilli farm environment (c), (d), (e)

3.3 Study Population

Chili-growing farmer group (Cases)

Chili-growing farmers in Hua-rua sub-district Muang district, Ubon Ratchathani Province, who were normally apply pesticides to the field, were subjects in this study.

Inclusion criteria	Exclusion criteria
- living in the study area more than 5 years - their houses and farms are located at Hua-rua sub-district Muang district, Ubon Ratchathani Province - farmer age between 18 – 59 years old - daily work in the farm - must be applicators who mix, load and spray pesticides	- hired for apply pesticides - presented pesticide exposure symptoms and/or sickness during sample collection period

Reference groups (Controls)

Reference group represented control group in this study. All participants were living in the study area but didn't exposure to pesticide directly.

Inclusion criteria	Exclusion criteria
- living in the community of study area more than 5 years - their houses located at Hua-rua sub-district Muang district, Ubon Ratchathani Province - age between 18 – 59 years old - non- daily farm worker - did not use to apply pesticide on farm	- farmers (rice or chili or others) or had been a farmer before - presented pesticide exposure symptoms and/or sickness during sample collection period - pesticide vendor

3.4 Sample Size and Sampling Techniques

In this study, a total subject was 80 persons approximately in order to do a personal in-depth monitoring for pesticide exposure. The subjects were separated into 2 groups; Chilli-growing farmers (exposure; n=40) and reference group (non-exposure; n=40).

The PS program (power and sample size calculation; Version 3.0.43) (Dupont and Plummer, 1990; 1998) was used for sample size calculation. Independent t-test mode was chosen for cases and control sample size calculation because this study tried to find a difference between two independent groups (exposure vs. non-exposure) on the means of a continuous variable (figure 3.2).

From Curwin et al., (2005) study, urinary metabolite of chlorpyrifos, which was a widely pesticide use in Hua-rua agricultural area, was collected from 24 male farmers and 23 male nonfarmers. He found the non-farmers' urinary metabolite concentration was 3.3 (±3.2) µg/L and farmers' (sprayed by self) urinary metabolite concentration was 5.9 (±5.3) µg/L. In order to calculate the sample size, a difference in mean (δ) is equal to 2.6 µg/L. The variances of two groups (farmer and non-farmer) were pooled in order to achieve the best estimate of the (assumed equal) variances of the 2 populations (Ruxton, 2006) and the pooled variance was used to calculate the standard deviation (σ = 3.26) to fill in the program. The ratio (m) in this study was equal to 1. It meant that this study was planning of a continuous response variable from independent control and experimental subjects with 1 control(s) per experimental subject.

If the true difference in the experimental and control means was 2.6, we need to study 34 experimental subjects and 34 control subjects to be able to reject the null hypothesis was that the population mean of the experimental and control groups were equal with probability (power) 0.9. The Type I error probability associated with this test of this null hypothesis was 0.05 (α).

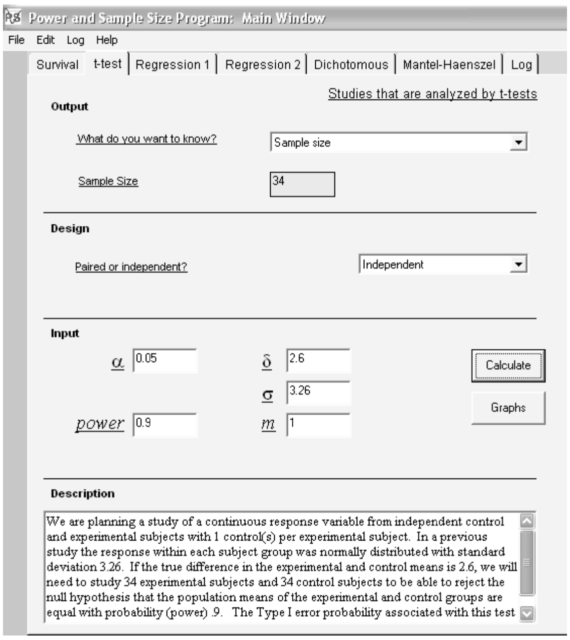
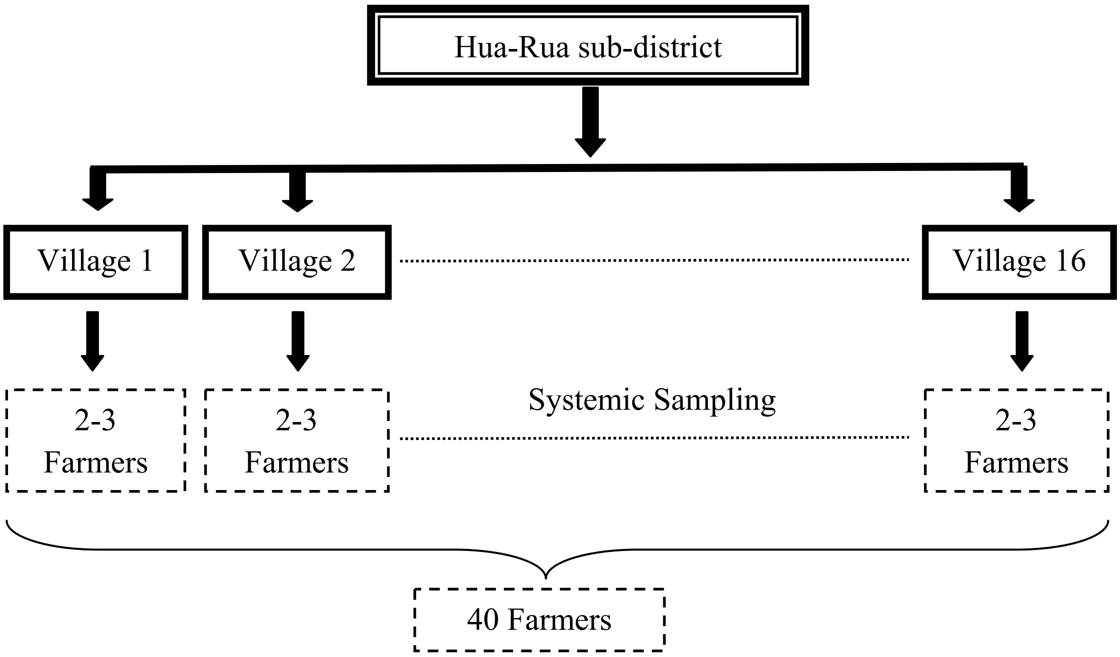


Figure 3.2 Sample size calculation using PS program

Sampling Techniques

Subjects in this study were selected by systemic sampling with every 10th individual from census records including convenience and willing to participate in the study. Exposure and non-exposure groups, 2-3 subjects per group, were included from 16 villages in Hua-Rua sub-district. They were selected by random number from census records, provided by primary health care workers in the study area. The sampling procedure was showed below.

Chilli-growing Farmers (40 subjects)



Non- Chilli-growing Farmers (40 subjects)

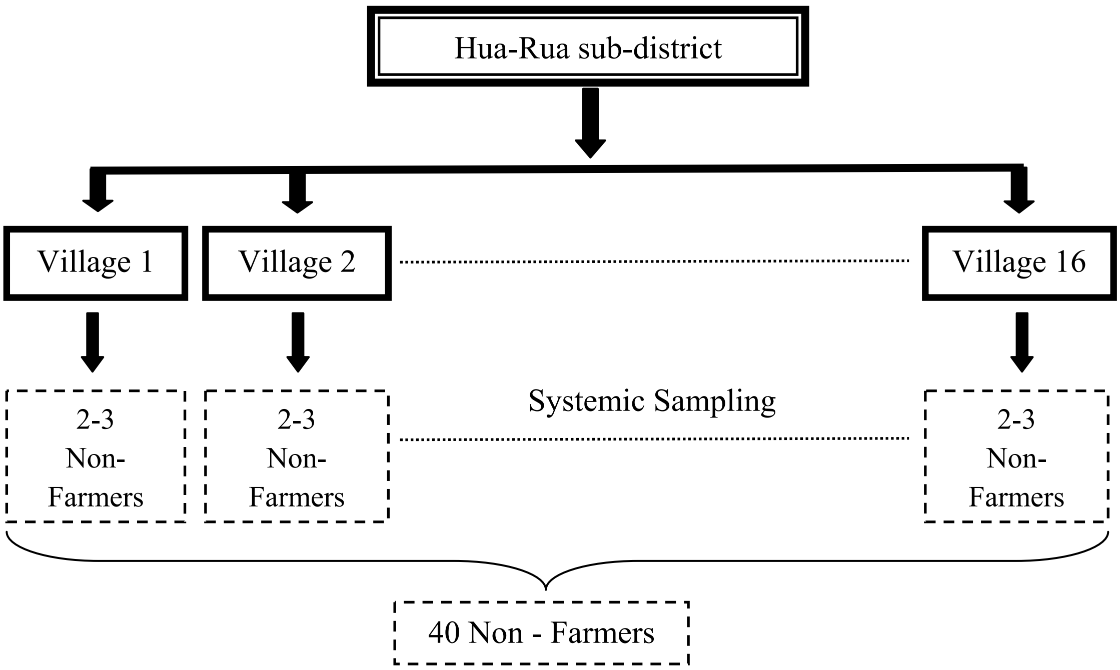


Table 3.1 The total collected samples in this study

Sample	Cases	Controls
Hand wipe	40	40
Feet wipe	40	40
Dermal patch	40	-
Face wipe	40	-
Drinking water	40	40
Air sample	40	40
Urinary Metabolite (Exposure : 3 samples/ subject Non-exposure : 1 sample/subject)	120	40
Total	560	

3.5 Face to face questionnaire study

Face to face interview method was applied to all subjects in this study. The questionnaire developed and adopted by researcher. It was composed of 3 parts covering general information, exposure information via multi-routes; inhalation, ingestion and dermal contact; and health effects related to pesticide exposure. Briefly, the first part questions asked about chilli-growing farmer's personal details, their field information including personal protective equipment (PPE) use. The exposure data, based on "How much? How long? How often?", was put in the second part using for average daily dose calculation and assess the risk. The questions were separated into 3 routes of exposure information which showed as previous. In the last part of this questionnaire, it was asked for health information related to organophosphate exposure. The information used to evaluate the health surveillance of this group of population. This questionnaire consisted of both open and close questions and the details of each part were showed in Appendix A (Jaipieam, 2008; Kavcar et al., 2009; Thomas et al., 2009; Issa et al., 2010). The summary of questionnaire is showed below;

Part 1: General Information

1. Gender
2. Body weight (kg)
3. Age (years)
4. Height (Centrimetes)
5. Education
6. Smoking

Agricultural works and farming descriptions

1. Area cultivated
2. Duration of application/ time
3. Years working in agriculture
4. Farming tasks
5. Years using pesticides
6. Number of annual pesticide applications
7. Application equipment and condition

Handling and practicing of pesticides

1. Using the required amounts of pesticides
2. Preparing (mixing) pesticides at home
3. Storing pesticides at home
4. Washing working clothes with the family clothes
5. Cleaning spraying equipment after work
6. Taking a meal at work place
7. Smoking while applying pesticides
8. Considering the safety period

Personal protective equipment (PPE)

1. Gloves
2. Mask
3. Boots
4. Hat

5. Short sleeved shirt
6. Long sleeved shirt
7. Short legged pants
8. Long legged pants

Part 2: Exposure Information (US EPA, 1997)

Inhalation

Exposure duration (hours)

Dermal contact

Event frequency (events/day)

Exposure frequency (days/year)

Ingestion

Source of drinking water

Number of glasses

Part 3: Health effects of pesticide exposure (Mitoko et al., 2000; Strong et al., 2004)

Skin Symptoms

Skin rash/ itching/ burning, tingling or numbness of hands, muscular twitching or cramps in the face, muscular twitching and cramps around the neck, muscular twitching and cramps in the arms, and muscular twitching and cramps in the legs

Respiratory Symptoms

Chest pain, cough, running nose, difficulties in breathing, shortness of breath, and irritation of the throat

Systemic Symptoms

Excessive sweating, nausea, vomiting, diarrhoea, excessive salivation, abdominal pain, headache

Eye Symptoms

Lacrimation and irritation, blurred vision

Neuro Muscular Symptoms

Difficulty in seeing, restlessness, difficulty in failing asleep, lacrimation, trembling of hands, and irritability, anxiety / anxiousness, memory Problem

The validity and reliability of questionnaire were tested. Three experts and professionals in related field of this study were asked for validity test in this questionnaire by using IOC (Index of item objective congruence) methods. The score equaled to 0.84. Pre-test questionnaire method from 17 persons were assessed for reliability (Cronbach's Alpha = 0.69).

3.6 Hazard Identification of Pesticide

"Hazard identification refers to the potential health effects that may occur from different types of pesticide exposure. It is strongly related to the extent and the type of a pesticide's toxic properties. This phase, usually involves the gathering of data on whether exposure to a pesticide causes an adverse effect. Hazard identification may also involve characterizing the behavior of a pesticide or metabolite within the human body and chemical interactions within organs, cells, or even parts of cells" (Frenich et al., 2007).

In this study, hazards were identified by the previous studies. In 2009, the face to face interview was conducted from 330 of 1,200 chilli farmers by convenient sampling method in Hua Rua sub-district, Muang district, Ubonratchathani Province by using standardized questionnaire. Twenty-three percent of farmers commonly applied Profenofos (Selecron) and 14.9% of them reported as Chlorpyrifos (Podium600) during their crop by self-spraying (Norkaew et al., 2010; Siriwong et al., 2011).

Residue of pesticides on chilli pre- harvesting products was also found by Ooraikul et al. (2011). Three samples from each of 11 chili farms were randomly collected 7 days after the last pesticide application in 2011 for a total of 33 chili samples. The detected pesticides were Chlorpyrifos (range <0.01 - 1.38 mg/kg) and Profenofos (range: 0.520 - 6.290 mg/kg). Moreover, of 15% profenofos contaminated samples were higher than the Maximum Residue Limits which is considered only the concentration of pesticide residue. The significant problem of chili-growing farmer in Hua Rua area was worm, aphid and plant louse. Furthermore, during sampling, the chili growing farmers reported that aphid was seriously presenting problem and most of them used the mixed of profenofos and chlorpyrifos due to they believed an increase in effectiveness resulted from the combination pesticides. Therefore, chlorpyrifos and profenofos were detected.

To assess the pesticide exposure of chilli farmers in this area, Chlorpyrifos and Profenofos were concentrated because of the significant finding from previous study. The main pesticides usage was already identified. Thus, the exposure assessment from multi-pathways were suggested to this study to figure out the main route of that exposure, the biological monitoring related to the exposure and health effects from this chilli farmers community.

3.7 Pesticide Exposure Assessment

3.7.1 Dermal Contact

Hands and feet wipe samples

Dermal (hands and feet) wipe samples was collected from chilli-growing farmers after complete their field activities (mixing, loading and applying pesticides) (Thomas et al., 2009). Two moistened patches with 40% isopropanal was used to wipe pesticide on each hand and foot of each farmer thoroughly. Both hands and both feet was collected and transferred to zip-lock bag separately. All wipe samples was closed, sealed, labeled and frozen for transport to laboratory and wait for analysis step (Jaipieam, 2008; Taneepanichskul, 2010).

Dermal skin samples

Standard patches (10 cm × 10 cm) used to collect exposure data via dermal route. There are 7 patches that fitted to the operator's clothing with safety pins. This method was based on the standard protocol for pesticides exposure developed by the WHO. Recommended sampling methods for assessing potential dermal exposure (PDE) had now been published by Health and safety executive (Johnson et al., 2004). The 7 positions set out in the positions as following, showed in Figure 3.3.

Position 1: on the hat, as close as practicable to the top of the head

Position 2: over the sternum, on the outside of normal clothing

Position 3: on the sternum, on the inside of normal clothing

Position 4: upper surface of the right forearm held with the elbow bent at right angles across the body, midway between elbow and wrist, on the outside of normal clothing.

Position 5: front of left leg, mid-thigh, on the outside of normal clothing

Position 6: front of left leg, above the ankle, on the outside of normal clothing

Position 7: on the back between shoulder blades, on the outside of normal clothing.

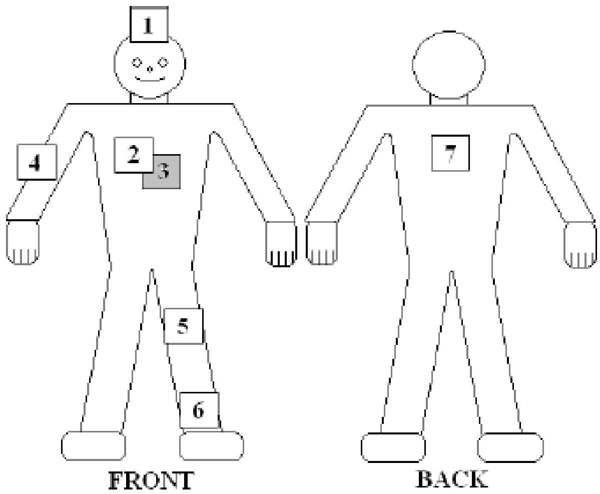


Figure 3.3 Position of the 7 sampling patches

Face wipes samples

Face wipe sample was a procedure for collecting pesticide residues from workers' face during the agricultural activities. The sampling procedure was modified from Dermal Face/ Neck wipe samples, agricultural handler exposure task force (Collier, 2009) showed as followed;

1. The field personnel collecting samples will wear clean, disposable gloves while collecting these dermal samples.

2. Dispense approximately 10 mL of 40% isopropanol alcohol/water solution on the gauze patch with the pipette

3. If the worker was wearing additional Personal Protective Equipment (PPE), such as goggles or a respirator, the worker removed all PPE before having the face/neck wipe collected.

4. Thoroughly wipe the worker's face/neck (front & back) with the moistened patch.

5. Wrap patch in aluminum foil (only if using a sealable bag) and place in the prelabelled bag, close the zip-lock, and place in frozen storage.

Face/neck wipe samples was collected before the workers eat anything and any time the workers would normally wash their face. Dermal face/neck wipe sample was collected from each worker after the hand wipe sample.

3.7.2 Ingestion

Drinking water samples

Drinking water procedure was modified from Sampling Guidance for Unknown Contaminants in Drinking Water (US EPA, 2008)

If the sample was being collected from a non-tap location, a clean 500 ml plastic container was used to dip the sample and fill the sample containers. Open the sample container, being careful not to contaminate the inside of the cap, the inside of the bottle, or the bottle threads. Fill the sample containers to ¼ inch from the top and cap the bottles. Wipe off the entire exterior of the container. Record the sample identification number, date and time of sample collection, sample location, and any other pertinent information on the sample label with a permanent marker and complete appropriate sample documentation form. Ensure that the appropriate sample label was permanently or securely affixed to each sample container. It was often easier to fill out the labels and attach them to the containers before mobilizing to the field.

3.7.3 Inhalation

Personal air samples (NIOSH, 1994; Jaipieam, 2008)

Air samples for measuring pesticide concentration via inhalation was followed by NIOSH 5600 method for sampling and analysis methods (Organophosphorus pesticides, Issue 1, dated 15 August 1994: Appendix D).

Air samples were collected at farmers' breathing zone by using personal sampling pump connected with sampler (Solid sorbent tube; OVS-2 tube: 13 mm quartz; XAD-2, 270 mg/140mg; appendix D). The pump was set and calibrated for each followed by NIOSH method;

Flow rate: 2 L/min

Volume Min: 12 L

Volume Max: 240 L

The sampler was set to connect with calibrated personal sampling pump with flexible tube and clipped to the applicator's collar where was in the farmer's breathing zone (Thomas et al., 2009). It was placed vertically with the large end down and it did not obstruct work performance. Flow-calibration checks for the pump were performed before and at the end of the sampling period. At the completion of sampling, the sampler capped both ends of it with plastic caps and pack for shipment. The sampler was stability at least 10 days at 25ºC and at least 30 days at 0ºC.

3.7.4 Biological Monitoring

Urinary Metabolite Samples

Urinary metabolite was concentrated on the primary exposure chemicals in this study. The major pesticides exposure was Organophosphate pesticides, thus all urine samples were analyzed for Dialkyl phosphate (DAP) metabolite. The biomarkers are shown on the table 3.2 below;

Table 3.2 Organophosphate pesticide and biomarker in this study

Pesticides	Biomarker
Organophosphate Insecticides	Dimethylphosphate (DMP)
	Dimethylthiophosphate (DMTP)
	Dimehyldithiophosphate (DMDTP)
	Diethylphosphate (DEP)
	Diethylthiophosphate (DETP)
	Diethyldithiophosphate (DEDTP)

3.7.5 Sample Analysis Procedures

Gauze patch analysis (Sample preparation)

Weight Sample 1 g

↓ Add DI water 10 ml
NaCl 5 g
Acetonitrile (HPLC) 10 ml
Magnesium Sulfate

Vortex Mixed 2 mines, Centrifuge 10 mines at 3000 rpm; 5ºC

↓

Transfer supernatant 5 ml to new tube

↓

Dry by N2 evaporator to less than 1 ml

↓

Adjusted volume to 1 ml

↓ Add Magnesium Sulfate ½ tea spoon
Add PSA Bowded Silica 1 tea spoon

Vortex Mixed 1 min, Centrifuge 3 mines at 3000 rpm; 5ºC

↓

Pass solvent to Syring filter Nylon 0.2 µm

↓

GC-µECD / GC-FPD

Drinking water analysis (Sample preparation)

Water sample 500 ml, Add NaCl 20 g in Separation funnel

↓

Extraction by CH_2Cl_2, 100: 50: 50 ml (3 times)

↓

Pass solvent through $NaSO_4$ anhydrous into 250 ml round bottom (3 times)

↓

Dry the residue by Rotary Evaporator (Dichloromethane Program; at 40 ºC)

↓

Adjusted volume by ethyl acetate (AR grade) 5 ml

↓

GC-FPD

Air sample analysis (Sample Preparation)

Added Toluene : Acetone (9:1, v/v) 20ml to air sampler

↓

Evaporated solvent by evaporator rotary (Acetone Program; 65ºC)

↓

Adjusted volume to 1 ml by Ethyl acetate

↓

GC-FPD

Urinary analysis (Sample preparation)

NaCl 2 g + Urine 5 ml

↓ Internal Standard (DBP; Conc 1.25 ppm) 50µl
HCL (6mol/L) 1 ml

Extraction by Ethyl acetate : Acetone (1:1, v/v) 2 x 5ml

↓ Shake 5 mines
Centrifuge 5 mines at 2000 rpm

Organic Solvent

↓ K_2CO_3 20 mg

Evaporated to 0.5 ml

↓ Acetonitrile 2 ml

Evaporated to dry

↓

Dry residue

↓ K_2CO_3 20 mg
Acetonitrile 3 ml
PFBBr 50 µl

Derivertization 15 hours at 50 ºC

↓ H_2O 4 ml
Hexane 2 x 5 ml

Liquid-Liquid Extraction

↓ Shake 5 mines
Centrifuge 5 mines at 2000 rpm

Organic Solvent

↓ Dry by N_2

Added Toluene 200 µl

↓

GC-FPD

Gas Chromatography (GC) condition for analysis

<u>GC-FPD</u>

Model:	Agilent Technology, 6890N, Made in USA		
Inlet:	Inject	2 µl	
	Temperature	200 ºC	
	Pressure	26 psi	
	Gas Type	Nitrogen	
Oven:	Initial temperature	80 ºC	
	Rate (ºC/min)	Final temp (ºC)	Time (min)
	12	195	0
	2	210	7
	15	225	10
	35	275	13
	Runtime	50.51 mines	
Column:	Capillary column		
	Model number: Agilent DB-1701 (30m x 0.248 mm x 0.25 µm particle size)		
	Flow: 2.6 ml/min		
Detector:	Flame Photometric Detector (FPD)		
	Temperature: 220 ºC		

GC-μECD

Model:	Agilent Technology, 6890N, Made in USA		
Inlet:	Inject	2 μl	
	Temperature	210 ºC	
	Pressure	14 psi	
	Gas Type	Nitrogen	
Oven:	Initial temperature	80 ºC	
	Rate (ºC/min)	Final temp (ºC)	Time (min)
	20	190	10
	3	215	3
	10	250	5
	20	280	10
	Runtime	47.83 mines	
Column:	Capillary column		
	Model number: Agilent 19091J-413 HP5		
	Flow: 3.1 ml/min		
Detector:	Micro-Electron Capture Detector (μECD)		
	Temperature: 320 ºC		

3.7.6 Quality Control

In term of inter and intra observer variation was controlled by using the standard laboratory (Center laboratory of Thailand in Khonkaen district) analyzing residue of pesticide. For biological monitoring (urinary metabolite), all samples were prepared by one researcher to reach the standard quality control at Laboratory. The laboratory will be assessed the analytical chemical technique to document method validation that AOAC Peer Verified Methods Program (1993) recommended.

Limit of Detection (LOD) and Limit of Quantitation (LOQ)

The limit detection (LOD) is the lowest concentration level that can be determined to be statistically different from a blank (99% confidence). The limit of quantitation (LOQ) is the level above which quantitative results may be obtained with a specified degree of confidence.

Method Detection Limit (MDL)

The method detection limit is the minimum concentration of a substance that can be measured and reported with 95% confidence that the analyze concentration is greater than zero.

Assessment of method precision

Relative Standard Deviation (RSD) or coefficient of variation (CV) used to estimate the precision for multiple samples. The precision acceptance criterion depends on the type of analysis. The precision in environmental analysis depends on the sample matrix, the concentration of analyte and the analysis technique. It is measured the variation between 2% and less than 20% (Siriwong, 2006).

Assessment of method accuracy

To access the method of accuracy is calculate by percent of recovery from analysis of reference materials, or laboratory control samples.

3.8 Data collection

Data collection was done by researcher and well-trained researcher assistants. The process of data collection was introduced and trained to all assistants before the data collection period by group and personal training. Some sample collections, such as dermal wipes and patch sample, was demonstrated by researcher.

Both chilli-growing farmers and non-chilli growing farmers was collected samples as following;

Questionnaire collection

Questionnaire collection, taking around 15-20 minutes, was started at the end of farm activities, starting from background information, exposure data, farm data, pesticide use, PPE and end up with sign and symptoms of pesticide exposure. Symptoms and sign of health effects were reported by chilli farmers at the time of spraying or within a few hours after it because this study concentrated on acute effects (Kishi et al., 1995). All question was answered by the farmers including observed by the researchers on farm in order to reduce bias from the reporter.

Dermal patch sample collection

Before they did their farm activities, loading, mixing and spraying pesticide, chili-growing farmers was asked to fit 7 patches in 7 positions (Johnson et al., 2004) to their clothing with safety pins on chest, back, arm and legs. There was a patch position put inside the cloth (position 3, figure 3.3). When the farmer had already done his job on farm, patches was removed from their body cautiously and collected them together in one square foil. After that, foil was labeled (farmer’s code) and kept in zip-lock and store in ice box with ice. Samples were in the ice until sent them to laboratory process.

Hands and feet wipe sample collection

Hands and feet wipe samples was collected from chilli-growing farmer after complete their field activities (mixing, loading and applying pesticides) (Thomas et al., 2009) before washing or cleaning their hands and feet. If he wore glove or boots, they were removed before sampling. Two moistened patches with 40% isopropanal was used to wipe pesticide residue on each hand and each foot of farmer thoroughly. Both hands and both feet was collected. Hands and feet wipe sample was kept separately and label farmer's code on square foil. Wipe samples was transfer to zip-lock bag separately. All wipe samples was closed, sealed, and frozen for transport to laboratory and waited for analysis step (Jaipieam, 2008; Taneepanichskul, 2010).

Face wipe sample

After hands and feet wipe sample collection, farmer was introduced to wipe his face by himself. Dispense approximately 10 mL of the surfactant solution (40% isopropyl alcohol/water) on the patch was given to the farmer. If he wore mask, it was removed before sample collection. The farmer's face was thoroughly wiped with the moistened patch and repeat again. The sample was kept in aluminum foil, place in the pre-labelled bag and frozen until analyze process (Collie, 2009).

Drinking water sample collection

In each sampling unit, the primary participant was asked about the main drinking water source, and samples was collected from tap or water container. If the source was tap water, samples were collected after 3-min flushing. The flow rate was reduced before sampling. Otherwise, it was collected from container on farm. The samples was filtered into 250-ml HDPE bottles (Kavcar et al., 2009). Sample preservation will be accomplished by storing the bottles at 4°C after sampling. Extraction will be carried out as soon as possible, within 7 days after collection (Quintana et al., 2001).

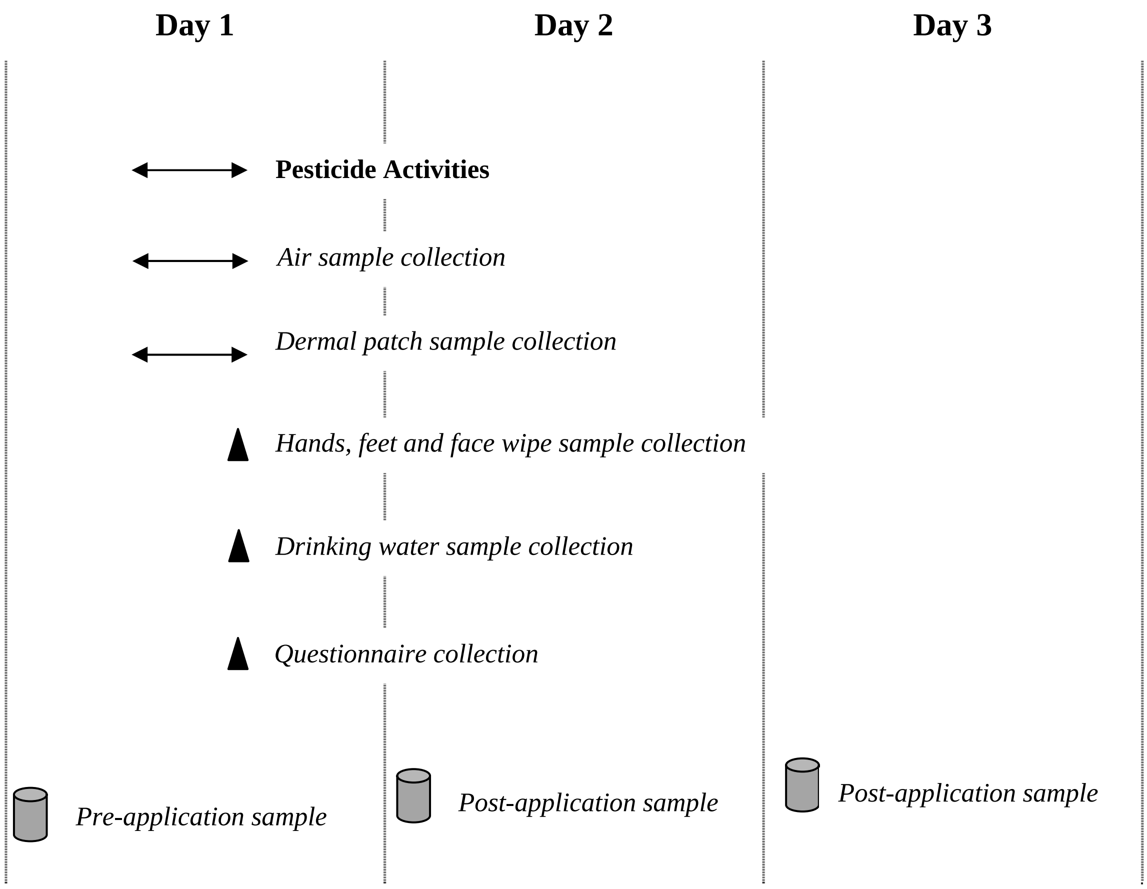
Personal air sample collection

Air samples were collected in the applicator's breathing zone during the pesticides activities (mixing, loading and applying). The sampler was clipped to applicator's collar and connected to the pump. It must not interfere subjects during his work. Flow calibrate was check before and after the completion of sampling period. After finish collection, the sampler was labeled and frozen in ice box and sent to laboratory (Thomas et al., 2009).

Urine sample collection

The subject was introduced how to collect the sample, including giving instruction, before the collecting period by the researcher. A morning void urine sample was collected from each chilli-growing farmer. Labeled polyethylene bottle, including instruction, was provided for all participants. Urine sample was collected from chili farmers 3 times/ farmer. Before the spraying day, farmer was asked for collecting first void urine on that day. Second urine sample was collected on the day after they do farm activity. On the third day, farmer was asked for collection urine sample again. All urine samples were first void morning urine. This research planed to collected urine 3 times because the urinary half-lives, following dermal dose, of 30 hours for dialkylphosphate metabolites (Thomas et al., 2009). Samples was transferred to zip-lock, labeled and kept in a cooler with frozen ice packs for transportation to the laboratory (Panuwet et al., 2008).

The collection timeline for one chilli-growing farmer in this study was showed on figure 3. , including all samples and pesticide activities.



Remark: First void morning urine sample collection

Figure 3.4 Time line for the sample collection

3.9 Data and Statistical Analysis

Statistical Analysis

In this study, analysis of data was done by using SPSS program version 17 for window. In term of describing the general information, descriptive statistic (mean, standard deviation (SD) and percentage) was use. The pesticide concentration via multi-route of exposure was concentrated at both mean and reasonable maximum exposure (RME) at 95^{th} percentile (upper bound) concerning of higher expose farmers, including average daily dose (ADD) exposure and Hazard Index (HI).

Chi-square test was used to find an association between wearing PPE and health symptoms reported form questionnaire. The significant of level was considered at 0.05 and 0.01, respectively.

Spearman's rank correlation was used to find the association between route of exposure and biological monitoring. Scatter plot graph to find the association and the relationship between each route of exposure also. Thus, the correlation between each pair was showed as an example followed;

Biological monitoring - Exposure routes

Urinary metabolite level - Dermal contact

Urinary metabolite level - Ingestion

Urinary metabolite level - Inhalation

Dermal – Ingestion

In term of statistical difference between cases and control pesticide exposure concentration, independent t-test was used to explain in each route of exposure.

Urinary metabolite (before MLA; exposure) Urinary metabolite (1st post MLA; exposure) Urinary metabolite (2nd post MLA; exposure)	} Urinary metabolite (non-exposure)

Paired t-test was used to explain the urinary metabolite statistical difference of each pair of urine samples.

Urinary metabolite (before MLA) - Urinary metabolite (1st post MLA)

Urinary metabolite (before MLA) - Urinary metabolite (2nd post MLA)

Urinary metabolite (1st post MLA) - Urinary metabolite (2nd post MLA)

Average Daily Dose (ADD) Calculation

To estimate the daily dose of chilli-growing farmers' exposure through muti-routes, the ADD equations of specific route was used. The factors in the equations wee asked from the farmers using questionnaires. The equations were showed as followed;

Dermal Contact (Siriwong et al., 2009; US EPA, 1997)

$$ADD_{dermal}(mg/kg - day) = \frac{CW \times SA \times PC \times ET \times EF \times ED \times CF}{BW \times AT} \quad \textit{eq.3-1}$$

Where:

CW	=	Chemical concentration in water (mg/l)
SA	=	Skin surface area (cm^2)
PC	=	Chemical-specific Permeability Constant (cm/hr)
ET	=	Exposure time (hours/day)
EF	=	Exposure frequency (days/year)
ED	=	Exposure duration (years)
CF	=	Volumetric conversion factor for water (1 l/1000 cm^3)
BW	=	Body weight (kg)
AT	=	Average time for carcinogenic effects, AT = ED

Ingestion (Lim et al., 2008)

$$ADD_{ingestion}(mg/kg - day) = \frac{C \times CF \times IR \times ED \times EF}{BW \times AT} \quad \textit{eq.3-2}$$

Where:

C	=	Contamination concentration (mg/kg)
CF	=	Conversion factor
IR	=	Ingestion rate per unit time (l/day)
ED	=	Exposure duration (years)
EF	=	Exposure frequency (days/years)

BW = Body weight (kg)

AT = Average time for carcinogenic effects, AT = ED

Inhalation (US EPA, 1997)

$$ADD_{dermal}(mg/kg - day) = \frac{C \times IR \times ED}{BW \times AT} \qquad \textit{eq.3-3}$$

Where;

ADD = Average daily dose (mg/kg/day)

C = Concentration of pesticide inhaled air (mg/m^3)

ED = Exposure duration (years)

EF = Exposure frequency (day/year)

BW = Body weight (kg)

AT = Averaging time (days) for non-carcinogenic effects (ED x 365 days)

Non-Carcinogen Risk Estimation

Hazard Quotient (HQ) expresses the risk estimation in this condition. The non-carcinogenic effects were calculated by the relationship below:

$$\text{Hazard Quotient (HQ)} = \text{Exposure / RfD} \qquad \textit{eq. 3-4}$$

Where:

Exposure = Chemical exposure level, or intake (mg/kg-day)

RfD = Reference dose (mg/kg-day)

If $HQ > 1$ adverse non-carcinogenic effect concern

$HQ \leq 1$ acceptable level (no concern)

In term of multiple substances and/or multi exposure pathway non carcinogenic effects calculation, it was expressed as hazard index (HI).

$$\text{Hazard Index (HI)} = \Sigma\,(\text{HQ}) \qquad \textit{eq. 3-5}$$

3.10 Ethic consideration

This study was approved by Institutional Review Boards (IRBs) of Ethical Committee of College of Public Health Sciences, Chulalongkorn University (ECCU group 1) (COA No. 038/2555; Date of approval: 9 March 2012). All participants had to sign a consent form prior to participation in this study. All subjects were asked to provide written, informed consent to participate in a study after they were provided with the requisite information on the pesticide (Chester, 2001).

CHAPTER IV

RESULTS

During the data collection period, there were only 3 villages form 16 villages where was growing chilli. All participants were selected by systemic sampling from that 3 villages by 10^{th} of census record provide by Hua- Rua tambon health promoting hospital workers. The participants were asked for their convenience and willing to participant before involving in this study. If they denied or were not willing, the next 10^{th} of census record was chose instead. Chilli farmers and non-chilli farmers had not to stay in the same house. All participants were explained thoroughly about this study by researcher before signed the consent form.

4.1 General Information and health effects related to pesticide exposure

4.1.1 Farmer and non-farmer characteristics’

Participants in this study composed of both 46 men and 34 women, separated into farmers and non-farmers group. Majority of age group were ranked between 30 to 39 years old (35.0%). Average weight (±SD) and height (±SD) were 61.11 (±12.46) kilograms and 162.54 (±6.79) centimeters, respectively. Most of them had finished elementary school (47.5%) and were non-smoker (80%).

Chilli growing farmers

Both male (65%) and female (35%) chilli-growing farmers participated in this study. Majority of chilli-growing farmers were 30 to 39 years of age while an average age (±SD) of all was 40.95 (±6.12) years old. Average BMI (±SD) was 23.18 (±4.48). Farmers completed elementary school (75%) was higher than those who had finished high school (25%). Chilli farmers’ average weight and height were 59.53 kilograms and 160.38 centimeters, respectively. They mostly were not smokers (Table 4.1).

Non-chilli growing farmers

From 40 non-chilli growing farmers, men and women were equal. Majority age group was younger than chilli farmer group (20-29 years old; 30%). An average BMI (±SD) was 23.01 (±4.21). Most of them had finished high school which was higher than another. The 80% of them reported as non-smoker. Average weight (±SD) and height (±SD) were 62.70 kilograms and 164.70 centimeters, respectively (Table 4.1).

In term of comparable of general characteristic between chilli farmers and non-chilli farmers, Chi-square test and fisher's exact test were applied. Sex, age, weight, height and BMI of both groups were comparable in term of statistical including smoking status. However, education level of non-chilli farmers was higher than chilli farmers and was not comparable (Pearson Chi-square; $p < 0.001$).

Table 4.1 General characteristics of study population

General Information	Chilli farmers	Non-Chilli farmers	Total
	(n = 40)	(n = 40)	(n=80)
Gender (n (%))			
Men	26 (65%)	20 (50%)	46 (57.5%)
Women	14 (35%)	20 (50%)	34 (42.5%)
Age groups (n (%))			
20 – 29	2 (5%)	12 (30%)	14 (17.5%)
30 – 39	18 (45%)	10 (25%)	28 (35.0%)
40 – 49	16 (40%)	10 (25%)	26 (32.5%)
50 or more	4 (10%)	8 (20%)	12 (15.0%)
Mean ±SD	40.95 (±6.11)	38.15 (±11.28)	39.55 (±9.12)
Range	26 - 52	19 - 57	19 - 57
Weight(kg) (mean ±SD)	59.53 (±10.99)	62.70 (±13.73)	61.11 (±12.46)
Height(cm) (mean ±SD)	160.38 (±5.10)	164.70 (±7.61)	162.54 (±6.79)

General Information	Chilli farmers	Non-Chilli farmers	Total
BMI (mean ±SD)	23.18 (±4.48)	23.01 (±4.21)	23.09 (±4.32)
	(n = 40)	(n = 40)	(n=80)
Education (n (%))			
Illiteracy	-	2 (5%)	2 (2.5%)
Elementary School [a]	30 (75%)	8 (20%)	38 (47.5%)
High School [b]	10 (25%)	18 (45%)	28 (35.0%)
College Graduate	-	8 (20%)	8 (10.0%)
Bachelor or higher	-	4 (10%)	4 (5.0%)
Smoking Status (n (%))			
Non-Smokers	30 (75%)	34 (85%)	64 (80.0%)
Smokers	10 (25%)	6 (15%)	16 (20.0%)

[a] *Elementary School: finished grade 6*
[b] *High School: finished grade 12*

4.1.2 Chilli farmer information related to pesticide exposure

Chilli farmers with over 10 years experience of pesticide application was the majority participants (85%), only 4 farmers had experience lower than 5 years. The average working hour of pesticide application was two hours for one time due to the application were performed in small cultivation area (average = 2.05 rai). An annual pesticide application was around 16 times. Average total annal income of chilli crop after cutting off capital was around 30,000 THB (995 USD) (Table 4.2).

Table 4.2 Agricultural works and farming characteristics (n=40)

Area cultivated(rai[a]) (mean ± SD)	2.05 (±0.71)
Duration of application/time[b] (Hrs) (mean ± SD)	2.0 (±0.3)
Years of using pesticides (n (%))(mean ± SD)	14.40 (±6.53)
0 – 9	6 (15 %)
10 – 19	18 (45%)
20 or more	16 (40%)
Frequency of annual pesticide applications (mean ± SD)	15.90 (±4.06)
Annual income (USD) (mean ±SD)	995 (±673)

[a] *Change Unit: 1 rai = 0.4 acre*
[b] *Duration of application including mixing and loading*

All chili farmers did not prepare and mix pesticide at their home; however some of them store their pesticide containers at home (20%) (Table 4.3). During pesticide application, all famers did not smoke and most of them did not take a meal to the farm (85%). Up to 95 % did not use the required amounts of pesticide shown on the label of pesticide container, and only 35 % cleaned or washed their equipment after finishing their work. About 85% of respondents indicated that they washed their working cloths and family clothes separately.

Table 4.3 Handling and practicing of pesticide use in Chili farmers (n=40)

	Chilli farmers: n(%)	
	YES	**NO**
Using the required amounts of pesticides	2 (5%)	38 (95%)
Preparing (mixing) pesticides at home	-	40 (100%)
Storing pesticides at home	8 (20%)	32 (80%)
Washing working clothes with the family clothes	6 (15%)	34 (85%)
Cleaning spraying equipment after work	14 (35%)	26 (65%)
Taking a meal at work place	6 (15%)	34 (85%)
Smoking while applying pesticides	-	40 (100%)

Table 4.4 was shown frequency of personal protective equipment (PPE) usage during pesticide application in chili farm. Most of them reported that they always wore PPE (gloves, nose mask, boots, hat, long sleeved shirts and long sleeved plants). The 100% PPE usage was long sleeved shirt. The 35% and 10% of all participants had never worn hat and nose mask respectively. Some chili farmers sometimes used some PPE as follows: gloves (40%), nose mask (10%), boots, hat and long legged plants (5%).

Table 4.4 Use of personal protective equipments (PPE) in chili farmers (n=40)

	Chilli farmers : n(%)		
PPE	**Never**	**Sometimes**	**Always**
Gloves	-	16 (40)	24 (60)
Nose mask	4 (10)	4 (10)	32 (80)
Boots	-	2 (5)	38 (95)
Hat	14 (35)	2 (5)	24 (60)
Long sleeved shirts	-	-	40 (100)
Long legged pants	-	2 (5)	38 (95)

4.1.3 Health effects related to pesticide exposure[1]

The reported acute health symptoms related to common pesticide exposure were recalled from chili farmers' experience after 24 hours pesticide application (Table 4.5). Most chilli farmers mentioned their health effects related to Central Nervous System (CNS). For skin symptoms, skin rash, the first reported symptom was itching and burning (20%). Irritation of the throat (40%), cough (35%) and shortness of breath (30%) had been diagnosed from the respiratory symptoms related to pesticide exposure. Few of them reported the running nose (20%). The main systemic symptoms were excessive salivation (65%) and excessive sweating (60%), respectively. Thirty five percent of them had headache problem. However, there were only 5% of chili farmers got an abnormal pain after their exposure. According to eye symptoms, irritation (30%) and blurred vision (35%) were reported nearly. Memory problem (70%) was pointed out as the first reported health symptom in this research. Nevertheless, restlessness and trembling of hands (10%) were mentioned from few participants.

The self-reported health symptoms from non-chilli farmers were used face to face interview with questionnaire. Most of them (65%) reported skin rash / itching / burning symptoms. For respiratory symptoms, they mentioned on chest pain (40%) and cough (50%) which were higher than chilli farmers' reports. Systemic and central nervous system (CNS) symptoms were found lower than the farmers, such as excessive sweating, excessive salivation and memory problem. Lacrimation and irritation were found 45% and 40%, respectively, which were higher than chilli farmer.

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[1] *Parts of this contents were published in Taneepanichskul, N., Norkaew, S., Siriwong, W., and Robson, G.M. 2012. Health effects related to pesticide using and practicing among chili-growing farmers, Northeastern, Thailand. Journal of Medicine and medical Sciences. 3(5) (2012), 319-325.*

Table 4.5 Subjective signs and symptoms related to common pesticide exposure

Symptoms	**Chilli farmers** (n=40) **n(%)**	**Non- Chilli farmers** (n=40) **n(%)**	**Total** (n=80) **n(%)**
Skin and extremeties			
Skin rash / itching / burning	8 (20)	26 (65)	34 (42.5)
Tingling / numbness of hands	2 (5)	8 (20)	10 (12.5)
Muscular twitching and cramps	4 (10)	4 (10)	8 (10.0)
Respiratory Symptoms			
Chest pain	10 (25)	16 (40)	26 (32.5)
Cough	14 (35)	20 (50)	34 (42.5)
Running nose	8 (20)	10 (25)	18 (22.5)
Difficulties in breathing	10 (25)	8 (20)	18 (22.5)
Shortness of breath	12 (30)	-	12 (15.0)
Irritation of the throat	16 (40)	16 (40)	32 (40.0)
Systemic Symptoms			
Excessive sweating	24 (60)	12 (30)	36 (45.5)
Headache	14 (35)	10 (25)	24 (30.0)
Vomiting/ diarrhea	4 (10)	4 (10)	8 (10.0)
Excessive salivation	26 (65)	6 (15)	32 (40.0)
Abdominal pain	2 (5)	14 (35)	16 (20.0)
Eye Symptoms			
Lacrimation	8 (20)	18 (45)	26 (32.5)
Irritation	14 (35)	16 (40)	30 (37.5)
Blurred vision	12 (30)	4 (10)	16 (20.0)
Central Nervous System (CNS) Symptoms			
Difficulty in seeing	10 (25)	4 (10)	14 (17.5)
Restlessness	4 (10)	8 (20)	12 (15.0)
Difficulty in failing asleep	12 (30)	8 (20)	20 (25.0)
Trembling of hands	4 (10)	4 (10)	8 (10.0)
Irritability	24 (60)	2 (5)	26 (32.5)
Anxiety / anxiousness	14 (35)	4 (10)	18 (60.0)
Memory problem	28 (70)	6 (15)	34 (42.5)

Association between health symptoms and wearing PPE was statistically analyzed (Table 4.6). Skin rash/ itching/ burning was associated with wearing glove ($p<0.01$) and nose mask ($p<0.05$). Muscular twitching and cramps was related with wearing glove ($p < 0.05$) and boots ($p<0.01$). Gloves and nose mask were suggested to wear for protecting farmers' themselves from chest pain. Moreover, gloves wearing also found the association for cough ($p<0.01$), running nose ($p<0.05$), shortness breath ($p < 0.05$), Vomiting/ diarrhea ($p<0.05$), trembling of hands ($p<0.05$) and Irritability ($p< 0.05$). Similarly, wearing nose mask was predicted as a protective factor with shortness breath ($p<0.01$), Vomiting/ diarrhea ($p<0.01$) and Abdominal pain ($p<0.05$). Difficulty in seeing found related with use of hat as same as wearing long legged plants. It could be suggested that wearing PPE was associated with incidence of health symptoms reduction.

Table 4.6 Statistical significant association between the use of PPE and reported health symptoms

PPE	Reported Symptoms	P-value
Glove	Skin rash / itching / burning	< 0.001**
	Muscular twitching and cramps	0.020*
	Chest pain	< 0.001**
	Cough	0.006**
	Running nose	0.042*
	Shortness of breath	0.037*
	Vomiting/ diarrhea	0.020*
	Trembling of hands	0.037*
	Irritability	0.020*
Nose Mask	Skin rash / itching / burning	0.037*
	Chest pain	< 0.001**
	Shortness of breath	0.005**
	Vomiting/ diarrhea	0.001**
	Abdominal pain	0.036*
Boots	Muscular twitching and cramps	0.008**

PPE	Reported Symptoms	P-value
	Running nose	0.036*
	Irritability	0.008**
Hat	Irritation	0.042*
	Restlessness	0.007**
	Difficulty in failing asleep	0.020*
Long legged pants	Difficulty in failing asleep	0.008**

*** Significant at 0.01 probability level*
** Significant at 0.05 probability level*

4.1.4 Body surface area calculation

Body surface area (SA) of participants in this study had to calculate because of an analysis of average daily dose (ADD) factor in the 3rd step (exposure assessment) of risk assessment process. The model of DuBois and DuBois (1916) (cited in US EPA, 2011) surface area calculation was used in this study (Equation 4-1). Specific average weight and height separated by group of participants (chilli farmers and non-chilli farmers) and sex were factor for this calculation (Table4.7).

$$SA = a_0 H^{a1} W^{a2} \qquad \textit{eq. 4-1}$$

Where:

SA = surface area (m^2)

H = height (cm)

W = weight (kg)

a_0, a_1, a_2 = constant values (US EPA, 1997)

The a_0, a_1 and a_2 in the equation were based on the US EPA's defaults values presented in the Appendix C.

Table 4.7 Average weight (kg) and height (cm) of participants (separated by sex and group)

Group / Sex	Male	Female	Male & Female
Farmers (n=40)			
Weight	60.9	56.9	59.5
Height	162	158	160
Non-Farmers (n=40)			
Weight	72.1	53.3	62.7
Height	170	159	165

Body surface area calculation was separated into specific parts of the body; head, face, hands, feet and total body surface area (Table 4.8). Head area was calculated due to the limit of default values on face surface area. Face surface area was equal to 1/3 of head area (US EPA, 2011). The data of weight and height in each calculation provided from questionnaire. Moreover, the specific defaults value of calculation was provide below table 4.8.

Table 4.8 Average surface area (m^2) of farmers in the study area

Average surface area (m^2)	Sex		
	Male	**Female**	**Male & Female**
Farmers			
Head	0.19^a	0.10^b	0.15
Face	0.06^c	0.03^c	0.05
Hands	0.19^d	0.12^e	0.16
Feet	0.08^f	0.08^f	0.08
Total	1.66^g	1.58^g	1.62
Non-Farmers			
Head	0.19^a	0.10^b	0.15
Face	0.06^c	0.03^c	0.05
Hands	0.19^d	0.12^e	0.16
Feet	0.09^f	0.07^f	0.08
Total	1.84^g	1.57^g	1.71

Defaults value of SA calculation
[a] a_0 = 0.0492, a_1 = 0.339, a_2 = -0.0950 (US EPA, 1985 cited in US EPA, 1997)
[b] a_0 = 0.0256, a_1 = 0.124, a_2 = 0.189 (US EPA, 1985 cited in US EPA, 1997)
[c] face area is equal to 1/3 of head area (US EPA, 2011)
[d] a_0 = 0.0257, a_1 = 0.573, a_2 = -0.218 (US EPA, 1985 cited in US EPA, 1997)
[e] a_0 = 0.0131, a_1 = 0.412, a_2 = 0.0274 (US EPA, 1985 cited in US EPA, 1997)
[f] a_0 = 0.000618, a_1 = 0.372, a_2 = 0.725 (US EPA, 1985 cited in US EPA, 1997)
[g] a_0 = 0.01545, a_1 = 0.54468, a_2 = 0.46336 (Gehan and George, 1970 cited in US EPA, 2011)

4.2 Pesticide exposure concentration via multi-exposure pathways: farmers and non-farmers

Personal pesticide exposure concentration samples were collected from both chilli farmers and non-chilli farmers for assessing and comparing the risk. Face wipe, hand wipe, foot wipe and body patch samples were estimated as dermal exposure. Inhalation exposure was investigated by personal air sampler connected with specific detector (Solid sorbent tube; XAD-2 OVS (Quartz Filter), 13>8 X 75 mm size, 2-section, 140/270 mg sorbent). In term of pesticide exposure through ingestion, drinking water was collected to measure that exposure from participants in this study.

The procedure of sample preparation and analysis were presented in Chapter 3. An analysis of sample in this study used gas chromatography with different specific detector. Personal air samples and drinking water were analyzed by gas chromatography (GC) connected with flame photometric detector (FPD). To detect multi-residue of OPPs in patch samples were specified by using GC with micro-electron capture detector (μECD).

4.2.1 Chilli farmers' pesticide exposure concentration

Pesticide exposure concentration was measured via multi-routes of its exposure from chilli farmers during and after their pesticide application (mixing, loading and applying: MLA). The procedure and timeline of sample collection was explained in the Chapter 3. Briefly, face wipe, hand wipe and foot wipe samples were collected after pesticide application process using moisten gauze patch with 40% iso-propanol; however, body patch and personal air samples were collected during their MLA. Drinking water was got from tank and/or cooler token to the farm by chilli farmers.

4.2.1.1 Detected samples frequencies

From 40 interviewed chilli farmer, 38 (95%) of them agreed to provide wipe and air samples. The results found that foot wipe and drinking water samples were not detected any OPPs pesticides. Hand wipe samples were detected both Chlorpyrifos and Profenofos at 10.53% and 26.32% respectively. Around 30% of face wipe sample could be found both chemical as previous as same as 80% of body patch samples were detected. All personal air samplers were detected Chlorpyrifos, but only 21.05% of them found Profenofos (Figure4.1)

4.2.1.2 Pesticide exposure concentration via multi pathways (dermal, oral and inhalation)

a) Dermal exposure concentration

Dermal exposure was concerned as the greatest route of pesticide exposure during MLA. Chilli farmers reported as usually wearing personal protective equipments (PPE), however inappropriate, misusing and homemade PPE were still found in this study area, as mention in Chapter 4. Thus, to figure out the residue of dermal contact during and after MLA was been a part. Hand wipe, foot wipe and face wipe after MLA including dermal patch collection during MLA were used as measurement tools.

In term of analysis, hands wipe sample (both hands) was analyzed together as same as 6 position of dermal patch sample. The procedure of body patch sample was recommended for 7 positions (Johnson et al., 2004) as follow; Position 1: on the hat, Position 2: over the sternum, on the outside of normal clothing, Position 3: on the sternum, on the inside of normal clothing, Position 4: upper surface of the right forearm held with the elbow bent at right angles across the body, Position 5: front of left leg, mid-thigh, on the outside of normal clothing, Position 6: front of left leg, above the ankle and Position 7: on the back between shoulder blades. However, the position 3 (on the sternum, on the inside of normal clothing) was cut off from this sample collection because of an inappropriate as Thai local tradition. Average concentrations and its distribution, including 95th percentile (RME), of hand wipe, face wipe and body patch samples were shown in table 4.9.

Figure 4.1 Detected personal exposure samples (%) of chilli-growing farmers after pesticide application including mixing, loading and applying pesticide (n=38)

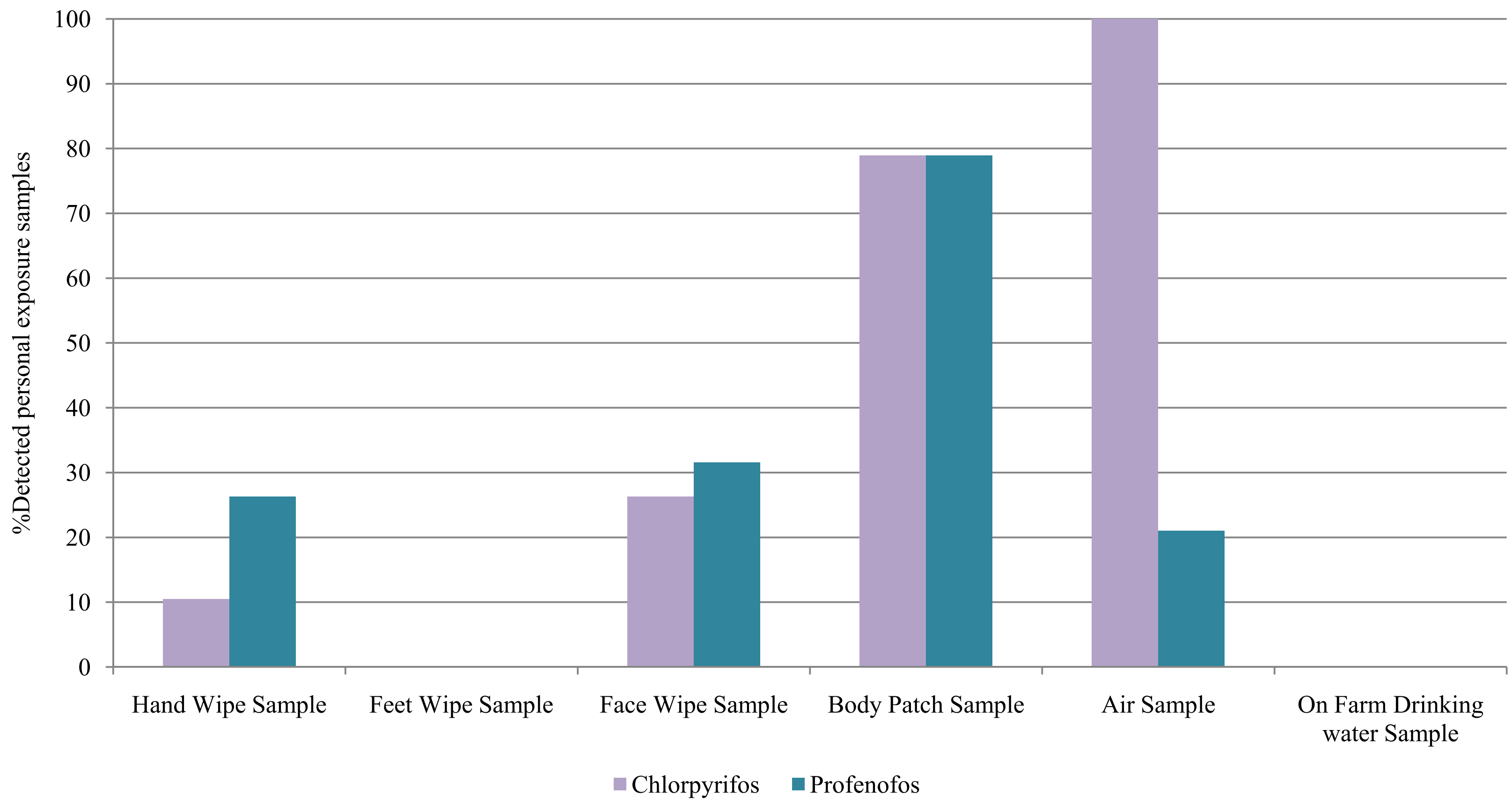


Table 4.9 Dose estimated of Organophosphate pesticides concentration in Chilli-farmers' dermal (mg/kg) (n=38)

	Mean	SD	Concentration at Percentile				Range
			25^{th}	50^{th}	75^{th}	95^{th}	
Hands							
Chlorpyrifos	0.043	0.068	<LOD	<LOD	<LOD	0.240	<LOD - 0.240
Profenofos	0.087	0.132	<LOD	<LOD	0.130	0.450	<LOD - 0.450
Face							
Chlorpyrifos	0.044	0.050	<LOD	<LOD	0.050	0.210	<LOD - 0.210
Profenofos	0.513	1.542	<LOD	<LOD	0.120	6.400	<LOD - 9.750
Body							
Chlorpyrifos	2.179	4.199	0.100	0.610	1.580	15.950	<LOD - 15.95
Profenofos	2.151	5.221	0.120	0.460	1.320	22.800	<LOD - 22.80

LOD (Limit of detection) <0.02 mg/kg

Hands wipe, face wipes and body patch samples could be detected both Chlorpyrifos and Profenofos, but foot wipe did not find any residue of pesticide in samples. Average concentration of Profenofos in hand and face wipe samples were higher than Chlorpyrifos, although both seemed to be equal in body patch sample.

b) Inhalation exposure concentration

Without any proper respirator protecting inhalation from pesticide during MLA, chilli famer could be accidentally exposed to pesticide and posed to acute and chronic health effects. Air samples were collected from farmers at their breathing zone representation by specific sampler. Totally, 38 air samples were collected from the farmers. Organophosphate pesticides, Chlorpyrifos and Profenofos, were detected in samples. Nevertheless, Profenofos was detected less than Chlorpyrifos (Table 4.10). Average, min, max including 95^{th} percentile of both pesticides concentration were also shown in Table 4.10.

Table 4.10 Dose estimated of Organophosphate pesticides concentration on Chilli-growing farmers' inhalation (mg/kg) (n=38)

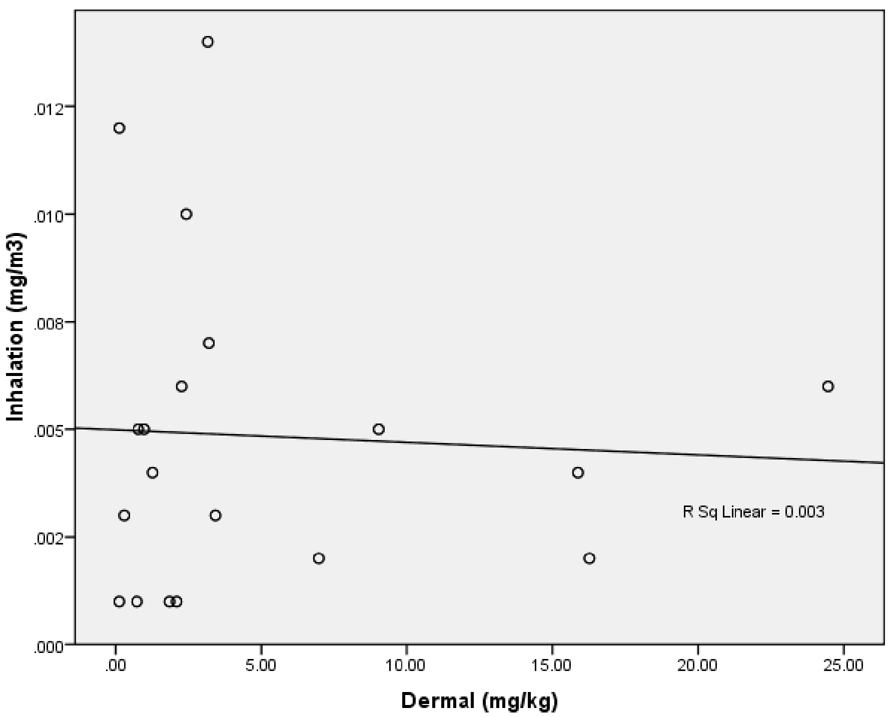
	Mean	SD	Concentration at Percentile				Range
			25th	50th	75th	95th	
Chlorpyrifos	0.004	0.004	0.001	0.002	0.005	0.014	<LOD - 0.010
Profenofos	0.001	0.002	<LOD	<LOD	<LOD	<LOD	<LOD - 0.006

LOD (Limit of detection) <0.001 mg/kg

c) Correlation between exposure route

In term of correlation of the concentration of pesticide exposure though multi exposure pathway, only association between dermal and inhalation exposure could be found, however the concentration of pesticide exposure through ingestion was not found. A scatter plot diagram showed the pattern of that association and there was not correlated in linear model (R^2=0.003) and there was no statistical association between dermal and inhalation exposure (Spearman rho's test; r_r = 0.155, p> 0.05) (Figure 4.2).

Figure 4.2 Scatter plot diagram of association between dermal and inhalation exposure



4.2.1.3 Exposure assessment and risk characterization

In the risk assessment, the exposure assessment and risk characterization were a process determining the dose of pesticide exposure and classify the risk of this study population. In term of exposure assessments, the direct assessments measuring the chemical concentration contact the person in the exposure media was done in this study. Personal monitoring techniques were used to measure exposure directly to an individual during a point in time (Sheldon, 2010).

The definition of Average daily dose (ADD), given by Integrated Risk Information System (IRIS) of US EPA, was “the mean amount of an agent to which a person is exposed on a daily basis, often averaged over a long period of time” (Exposure Factors Handbook, US EPA (2011)) and “dose rate averaged over a pathway-specific period of exposure expressed as a daily dose on a per-unit-body-weight basis and usually expressed in terms of mg/kg-day or other mass-time units”.

The hazard quotient (HQ) and hazard index (HI) were used in the step of risk characterization. The definition of hazard quotient (HQ) was “the ratio of estimated site-specific exposure to a single chemical from a site over a specified period to the estimated daily exposure level, at which no adverse health effects are likely to occur” (Terms of Environment, US EPA (2009)). Moreover, “The sum of more than one hazard quotient for multiple substances and/or multiple exposure pathways” was defined as Hazard Index (HI) by Waste and Cleanup Risk Assessment (US EPA, 2010).

a) Dermal Exposure Assessment

Exposure assessment through dermal route was calculated based on the equation, recommended by exposure factor handbook (US EPA, 1997). The dermal average daily dose (ADD) can be estimated by;

$$ADD_{dermal}(mg/kg - day) = \frac{DAevent \times EV \times ED \times EF \times SA}{BW \times AT} \quad eq.4\text{-}2$$

Where:

DAevent = absorbed dose per event (mg/cm -event)

EV = event frequency (events/day)

ED = exposure duration (years)

EF = exposure frequency (days/year)

SA = skin surface area available for contact (cm)

BW = body weight (kg)

AT = averaging time (days) for non carcinogenic effects, AT = ED

Hand Contact

The exposure estimation of hand contact was calculated by the following equation (*eq.4-3*), adopt from above equation (*eq.4-2*). The values in each factor of equation were presented in Table 4.11.

$$ADD_{dermal}(mg/kg - day) = \frac{Cs\ x\ SA\ x\ DAevent\ x\ EV\ x\ ED\ x\ EF}{BW\ x\ AT} \quad eq.4\text{-}3$$

Where;

ADD	=	Average daily dose (mg/kg/day)
Cs	=	Concentration of pesticide on both hands (mg/kg)
SA	=	Surface area (cm^2)
DAevent	=	absorbed dose per event (mg/cm^2-event)
EV	=	Event frequency (event/day)
ED	=	Exposure duration (years)
EF	=	Exposure frequency (day/year)
BW	=	Body weight (kg)
AT	=	Averaging time (days) for non-carcinogenic effects (ED x 365 days)

Face contact

For face contact exposure estimation, the ADD was calculated by the same equation as hand contact (*eq.4-3*), but the surface area was specific for only face of participants. The values of factor for this calculation was shown in Table 4.12.

Body skin contact

An exposure estimation of body skin contact was calculated by the following equation (*eq.4-4)*. The equation was similar to the previous (*eq.4-3*), but transfer efficiency from clothes to skin (TE) must be added because of the outside clothes concentration measurement. TE was equal to 0.1 without unit (Cal-EPA, 2004 cited in Jaipieam, 2008). The values of factor presented in Table 4.13.

$$ADD_{dermal}(mg/kg - day) = \frac{Cs \times SA \times DAevent \times EV \times ED \times EF \times TE}{BW \times AT} \qquad eq.4\text{-}4$$

Table 4.11 Value of factors in average daily dose (ADD) equation (hand contact) for chilli farmers in Hua rua sub district, Muang district, Ubon Ratchathani Province

	Concentration (Cs) (mean)	**Concentration (Cs) (95th percentile)**	**Surface area[a] (SA)**	**Absorbed dose per event[b] (DAevent)**	**Event frequency (EV)**	**Exposure duration (ED)**	**Exposure frequency (EF)**	**Body weight (BW)**	**Averaging time (AT)**
	(mg/kg)	(mg/kg)	(cm^2)	(mg/cm^2-h)	(hour/day)	(years)	(days/year)	(kg)	(days)
Chlorpyrifos	0.043	0.240	1.6×10^3	456×10^{-6}	2.00	14.40	15.90	59.53	5,256
Profenofos	0.087	0.450	1.6×10^3	456×10^{-6}	2.00	14.40	15.90	59.53	5,256

[a] SA values from direct calculation (Table 4.8)
[b] DAevent value from Griffin et. al.,1999

Table 4.12 Value of factors in average daily dose (ADD) equation (face contact) for chilli farmers in Hua rua sub district, Muang district, Ubon Ratchathani Province

	Concentration (Cs) (mean)	**Concentration (Cs) (95th percentile)**	**Surface area[a] (SA)**	**Absorbed dose per event[b] (DAevent)**	**Event frequency (EV)**	**Exposure duration (ED)**	**Exposure frequency (EF)**	**Body weight (BW)**	**Averaging time (AT)**
	(mg/kg)	(mg/kg)	(cm^2)	(mg/cm^2/h)	(hour/day)	(years)	(days/year)	(kg)	(days)
Chlorpyrifos	0.044	0.210	0.5×10^3	456×10^{-6}	2.00	14.40	15.90	59.53	5,256
Profenofos	0.513	6.400	0.5×10^3	456×10^{-6}	2.00	14.40	15.90	59.53	5,256

[a] SA values from direct calculation (Table 4.8)
[b] DAevent value from Griffin et. al.,1999

Table 4.13 Value of factors in average daily dose (ADD) equation (body skin contact) for chilli farmers in Hua rua sub district, Muang district, Ubon Ratchathani Province

	Concentration [a] **(Cs) (mean)** (mg/kg)	**Concentration** [a] **(Cs) (95th percentile)** (mg/kg)	**Surface area**[b] **(SA)** (cm^2)	**Absorbed dose per event**[c] ($mg/cm^2/h$)	**Transfer efficiency**[d] **(TE)**	**Event frequency (EV)** (hour/day)	**Exposure duration (ED)** (years)	**Exposure frequency (EF)** (days/year)	**Body weight (BW)** (kg)	**Averaging time (AT)** (days)
Chlorpyrifos	1.090	7.975	16.2×10^3	456×10^{-6}	0.1	2.00	14.40	15.90	59.53	5,256
Profenofos	1.076	11.40	16.2×10^3	456×10^{-6}	0.1	2.00	14.40	15.90	59.53	5,256

[a] Average concentration (Table 5.1) divided by 2 (Johnson et al., 2005)
[b] SA values from direct calculation (Table 4.8)
[c] DAevent value from Griffin et. al.,1999
[d] Transfer efficiency from clothes to skin (Cal-EPA, 2004)

The average daily dose (ADD) was calculated from the equation as previous. ADD was calculated at both mean and 95th percentile concentration for reasonable maximum exposure (RME) of participants concern. The upper confidence (95th percentile) on the arithmetic average concentrations was used to estimate the RME because the uncertainty associated with any estimate of exposure concentration might occur in this situation (Siriwong et al., 2010). The organophosphate pesticides (OPPs), Chlorpyrifos and Profenofos, were separated calculated for ADD including body part separation (Table 4.14). The calculation of ADD was shown in Appendix D.

The highest ADD was found in body skin contact to OPPs according to the larger surface area than others. The ADD of Profenofos on face contact was higher than Chlorpyrifos at both mean and RME. In the same way, hand contact of Profenofos was also more than Chlorpyrifos. It can be concluded by IRIS definition of ADD that chilli farmers in this study area exposed on Profenofos in daily basis life more than Chlorpyrifos.

Table 4.14 Average daily dose (ADD) of chilli farmers (on hand, face and body) in Hua rua sub district, Muang district, Ubon Ratchathani Province

ADD (mg/kg-day)	Hand	Face	Body surface
Chlorpyrifos			
ADD_{mean}	4.59×10^{-11}	1.47×10^{-11}	1.18×10^{-9}
ADD_{RME}	25.6×10^{-11}	7.01×10^{-11}	8.62×10^{-9}
Profenofos			
ADD_{mean}	9.29×10^{-11}	17.1×10^{-11}	1.16×10^{-9}
ADD_{RME}	48.1×10^{-11}	214×10^{-11}	12.3×10^{-9}

Dermal risk characterization

In term of non-carcinogenic risk characterization, Hazard quotient (HQ) and Hazard index (HI) were used. They was calculated by the following equation which recommended by US EPA (1997).

$$\text{Hazard Quotient (HQ)} = \frac{\text{Exposure}}{\text{RfD}} \qquad \textit{eq.4-5}$$

Where:

Exposure = chemical exposure level (mg/kg/day)

RfD = reference dose (mg/kg/day)

The explanation of HQ was shown below;

HQ > 1 adverse non-carcinogenic effect concern

HQ ≤ 1 acceptable level (no concern)

The exposure factor in *eq.4-5* was a representative of calculated ADD (Table 4.14). The reference dose with specific chemical, Chlorpyrifos and Profenofos, though dermal contact was presented in Table 4.15.

In term of multiple substances and/or multi exposure pathway non carcinogenic effects calculation, it was expressed as hazard index (HI).

$$\text{Hazard Index (HI)} = \Sigma\,(\text{HQ}) \qquad \textit{eq.4-6}$$

The interpretation of Hazard Index was as same as Hazard Quotient. HI was higher than 1, so adverse non-carcinogenic effect had to concern. In contrast, the lower or equal to 1 of HI was an acceptable level.

Table 4.15 Hazard Quotient (HQ) (on hand, face and body) of study population

HQ	Dermal RfD (mg/kg-day)	Hand	Face	Body skin
Chlorpyrifos				
HQ_{mean}	0.0015[a]	3.06×10^{-8}	0.98×10^{-8}	78.7×10^{-8}
HQ_{RME}	0.0015[a]	17.1×10^{-8}	46.7×10^{-8}	575×10^{-8}
Profenofos				
HQ_{mean}	0.00005[b]	1.86×10^{-6}	3.42×10^{-6}	2.32×10^{-5}
HQ_{RME}	0.00005[b]	9.62×10^{-6}	42.8×10^{-6}	24.6×10^{-5}

[a] Dermal RfD value from Jaipieam (2008)
[b] Dermal RfD value from US EPA (2009)

Hazard Quotients (HQ) for Chlorpyrifos and Profenofos at both mean and RME were not exceed the acceptable level for chilli growing farmers. HQ of Chlorpyrifos was highest via body skin contact as same as Profenofos. In term of comparison, HQ chlorpyrifos (hand contact) was lower than Profenofos at mean and RME. The face contact and body skin contact, the same interpretation as hand contact could be explained. The higher HQ of Profenofos exposure came from lower reference dose (RfD), divided factor in the *eq.4-5*, leading to the higher results from calculation.

b) Inhalation Exposure Assessment

Exposure assessment via inhalation was calculated based on the equation from "Exposure factor handbook" (US EPA, 1997). It was estimated by the following equation;

$$ADD_{dermal}(mg/kg - day) = \frac{C \times IR \times ED}{BW \times AT} \qquad eq.4\text{-}7$$

Where;

ADD	=	Average daily dose (mg/kg/day)
C	=	Concentration of pesticide inhaled air (mg/m^3)
ED	=	Exposure duration (years)
EF	=	Exposure frequency (day/year)

BW = Body weight (kg)

AT = Averaging time (days) for non-carcinogenic effects (ED x 365 days)

The values of each factor in the equation were shown in Table 4.16. The intake rate (IR) was came from the multiply of heavy breathing (3.9 m^3/hr) (US EPA, 2011) and duration of MLA (2 hr/day), so the IR was equal to 7.80 m^3/hr. The calculation of ADD via inhalation demonstrated in Appendix D.

Inhalation risk characterization

Risk characterization of inhalation was used the same procedure of dermal risk characterization in this study. Hazard quotient (HQ) and Hazard index (HI) were calculated based on the previous equation (*eq.4-5* and *eq.4-6*). The exposure factor in *eq.4-5* was a representative of calculated ADD (Table 4.17). The reference dose with specific chemical, Chlorpyrifos and Profenofos, though inhalation was presented in the same table as ADD.

Interpretation of Hazard Index and Hazard Quotient was as same as dermal contact. HI and/or HQ was higher than 1, so adverse non-carcinogenic effect had to concern. In contrast, the lower or equal to 1 of HI and/or HQ was an acceptable level.

Table 4.16 Value of factors in ADD equation (inhalation) for chilli farmers in Hua rua sub district, Muang district, Ubon Ratchathani Province

	Concentration (Cs) (mean) (mg/m^3)	**Concentration (Cs) (95th percentile)** (mg/m^3)	**Intake rate (IR)** (m^3/day)	**Exposure duration (ED)** (years)	**Exposure frequency (EF)** (days/year)	**Body weight (BW)** (kg)	**Body weight (AT)** (days)
Chlorpyrifos	0.001	0.004	7.80	14.40	15.90	59.53	5,256
Profenofos	0.0003	0.007	7.80	14.40	15.90	59.53	5,256

Table 4.17 Average daily dose (ADD) and Hazard Quotient (HQ) via inhalation of chilli farmers in Hua rua sub district, Muang district, Ubon Ratchathani Province

	ADD (mg/kg-day)	**RfD[a] (mg/kg-day)**	**HQ**
Chlorpyrifos		0.0003	
mean	5.71×10^{-6}		0.02
RME	22.8×10^{-6}		0.08
Profenofos		0.112	
mean	1.71×10^{-6}		1.53×10^{-5}
RME	40.0×10^{-6}		3.57×10^{-5}

[a] Inhalation RfD value from Jaipieam (2008)
RME - Reasonable maximum exposure at 95th percentile concentration (US EPA, 1989)

Table 4.17 showed ADD and HQ of Chlorpyrifos and Profenofos, including RfD. At mean level, the ADD of Chlorpyrifos (5.71×10^{-6} mg/kg-day) was higher than Profnofos (1.71×10^{-6} mg/kg-day), but the contrast of ADD was found at RME level because the highest detected Profenofos concentration was higher than the highest detected Chlorpyrifos was found in this study. Nevertheless, the RfD of Chlorpyrifos was lower than Profenofos for inhalation route. HQ of Chlorpyrifos seemed closed to 1 than Profenofos, it could be explained that chilli farmers got risk from Chlorpyrifos exposure more than profenofos. But, the HQ value was still not exceed than "1" so the adverse health effected was not need to concern for this population via inhalation during pesticide application.

The Hazard index (HI) of inhalation with Chlorpyrifos and Profenofos was calculated and got the same values as HQ of Chlorpyrifos at mean (HQ= 0.02) and RME (HQ= 0.08). The same figure of HQ and HI came from the lower HQ of Profenofos of inhalation (close to zero), thus it was not affected in the summation equation.

Table 4.18 showed the HI separated by chemical (Chlorpyrifos and Profenofos) and calculated HI. The summation of HI was calculated from *eq.4-8.*

$$HI = HQ_{hand} + HQ_{face} + HQ_{body} + HQ_{inhalation} \qquad eq.4\text{-}8$$

At mean, HI of Chlorpyrifos and Profenofos equaled to 0.02 and 4.38×10^{-5}, respectively. It could interpret that chilli farmers were not at risk from Organophosphate pesticides exposure even considerate at RME level (Table 4.18).

Table 4.18 Hazard Quotient (HQ) and Hazard index (HI) of chilli farmers in Hua rua sub district, Muang district, Ubon Ratchathani Province, identified by chemical

	HQ_{hand}	HQ_{face}	HQ_{body}	$HQ_{inhalation}$	HI
Chlorpyrifos					
Mean	3.06×10^{-8}	0.98×10^{-8}	78.7×10^{-8}	0.02	**0.02**
RME	17.1×10^{-8}	46.7×10^{-8}	575×10^{-8}	0.08	**0.08**
Profenofos					
Mean	1.86×10^{-6}	3.42×10^{-6}	2.32×10^{-5}	1.53×10^{-5}	$\mathbf{4.38 \times 10^{-5}}$
RME	9.62×10^{-6}	42.8×10^{-6}	24.6×10^{-5}	3.57×10^{-5}	$\mathbf{0.33 \times 10^{-5}}$

RME - Reasonable maximum exposure at 95th percentile concentration (US EPA, 1989)

Hazard index could be explained in the body part direction (Table 4.19). The summation of HI was came from HQ of two chemicals in each body part, the following equation;

$$HI = HQ_{chlorpyrifos} + HQ_{profenofos} \qquad eq.4\text{-}9$$

At mean and RME level, calculated HI were not exceed than 1. Body contact was found the highest HI comparing to other dermal parts but lower than inhalation. Chilli farmers had HI of face contact higher than HI of hand contact.

Table 4.19 Hazard Quotient (HQ) and Hazard index (HI) of Organophosphate pesticide exposure of chilli farmers in Hua rua sub district, Muang district, Ubon Ratchathani Province, identified by body part

	$HQ_{chlorpyrifos}$	$HQ_{profenofos}$	HI
Hand			
Mean	3.06×10^{-8}	1.86×10^{-6}	1.89×10^{-6}
RME	17.1×10^{-8}	9.62×10^{-6}	9.79×10^{-6}
Face			
Mean	0.98×10^{-8}	3.42×10^{-6}	3.43×10^{-6}
RME	46.7×10^{-8}	42.8×10^{-6}	43.3×10^{-6}
Body contact			
Mean	78.7×10^{-8}	2.32×10^{-5}	24.0×10^{-6}
RME	575×10^{-8}	24.6×10^{-5}	252×10^{-6}
Air			
Mean	0.02	1.53×10^{-5}	0.02
RME	0.08	3.57×10^{-5}	0.08

RME - Reasonable maximum exposure at 95th percentile concentration (US EPA, 1989)

The conclusion of HI was presented in the term of exposure routes, dermal exposure and inhalation exposure (Table 4.20). The HI of dermal exposure at mean was 2.93×10^{-5} and 30.5×10^{-5} at 95th percentile (RME). The exposure via inhalation got HI equal to 0.02 and 0.08 at mean and RME, respectively.

Table 4.20 Hazard index (HI) and Reasonable maximum exposure (RME) of chilli farmers in Hua rua sub district, Muang district, Ubon Ratchathani Province, identified by exposure route

	Mean	**RME**
Dermal exposure	2.93×10^{-5}	30.5×10^{-5}
Inhalation exposure	0.02	0.08

RME - Reasonable maximum exposure at 95th percentile concentration (US EPA, 1989)

Form the finding in this study, it found that chilli farmers were not at risk from Organophosphate pesticides (Chlorpyrifos and Profenofos) exposure by the procedure of risk assessment using risk characterization recommended by US EPA (2012). However, the comparison between routes found that chilli farmers got higher HI of inhalation route than dermal route. Thus, it can concluded that chilli farmers were exposed pesticides thought inhalation more than dermal contact so the health effects should be concern via inhalation more than dermal contact.

4.2.2 Non- Chilli farmers' pesticide exposure concentration

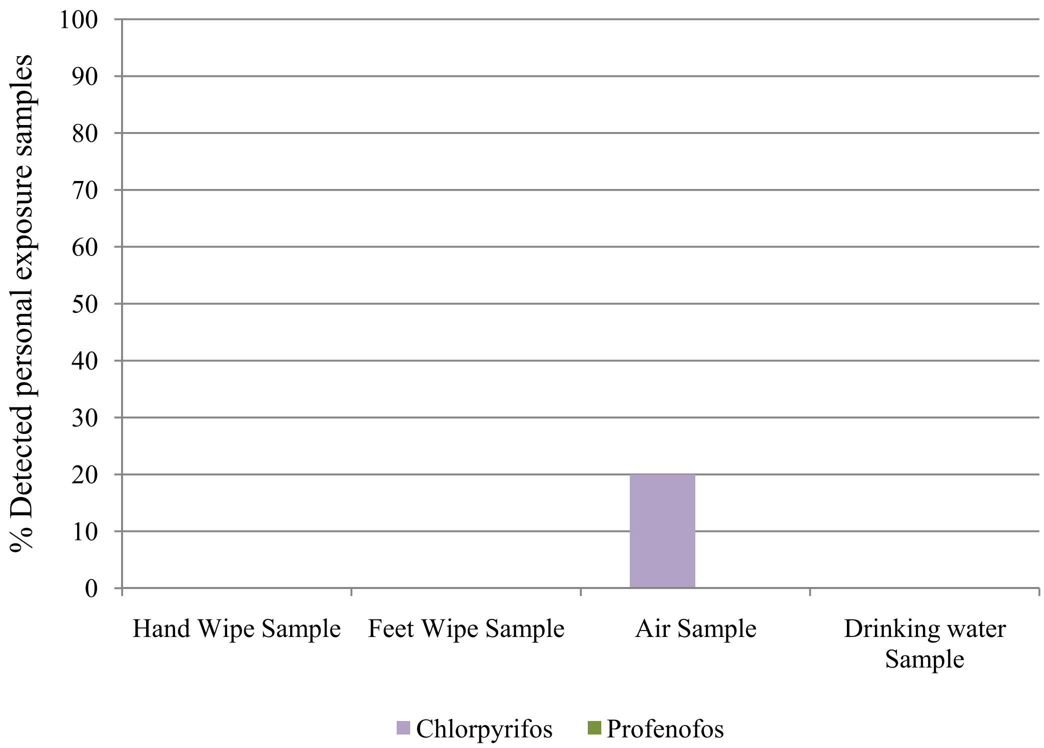
Indirect pesticide exposure was measure from people living in Hua rua sub district, Muang district, Ubon Ratchathani Province, but not participated in any agricultural section. Totally, 40 participants were included in this study part, as a control group. An investigation of multi-route of pesticide exposure was conducted in the same period of previous sample collection. The participants were randomly selected with specific criteria.

Briefly, hand wipe sample and foot wipe sample were collected by 40% iso-propanol moisten gauze as same as the procedure of chilli farmers. The participants did not be informed about the exactly sampling time; the samples were collected by accidentally because the sample was a represent of the real situation. After wipe samples, drinking water were collected and the air samples were collected for 24 hours in the common area of the house. So, the air samples were collected the day after sampling period.

4.2.2.1 Detected samples frequencies

All participants in this part of sample collection (control group) agreed to provide hand wipe, foot wipe, drinking water and air sample. So, there were 40 samples for each sending to central laboratory for analysis OPPs. Only 20% of air sample could be detected Chlorpyrifos (Figure 4.3). Nevertheless, there were no hand wipe, foot wipe and drinking water samples detected of OPPs (both Chlorpyrifos and Profenofos).

Figure 4.3 Detected personal exposure samples (%) of non-chilli farmers



4.2.2.2 Pesticide exposure concentration via multi pathways (dermal, oral and inhalation)

a) Dermal and oral exposure concentration

From laboratory analysis of personal samples, hand wipe and foot wipe used for estimating dermal exposure dose of non chilli farmers. Concentrations (mg/kg) were not detected in any samples (Figure 4.3). It could be reported that non-chilli farmers living this agricultural community, Hua rua sub district, Muang district, Ubon Ratchathani Province, were exposed to OPPs in the level less than limit of detection (LOD) in this study. Limit of detection in this study was equal to 0.02 mg/kg for patch analysis.

Similarly, drinking water analysis used for determining the level of pesticide exposure via oral route did not detected any OPPs. It could be reported that people living in this area exposed to OPPs though oral route (drinking water) less than LOD of analysis in this study.

b) Inhalation exposure concentration

Only 8 samplers form 40 collected sampler detected OPPs (Chlorpyifos). The detection level was rank from 0.001-0.002 mg/m^3. Thus, the estimation of mean, median and 95th percentile could calculate for 40 subjected because the concentrations were showed as less than LOD. The concentrations, presented in Table 4.21, were calculated by dividing only 8 subjects in this study in order to exposure assessment could be calculated for inhalation exposure. The mean concentration of air samplers was 0.0015 mg/m^3 and the RME (95th percentile) was 0.0020 mg/m^3.

Table 4.21 Dose estimated of Organophosphate pesticides concentration on non chilli-growing farmers' inhalation (mg/m^3) (n=8)

	Mean	Median	Min	Percentile				Max
				25th	50th	75th	95th	
Chlorpyrifos	0.0015	0.0015	0.0010	0.0010	0.0015	0.0020	0.0020	0.0020

4.3 Biological sample results: chilli farmers and non-chilli farmers

4.3.1 Creatinine Concentration in chilli farmers and non-farmers

Urinary creatinine concentration was measure for adjusting concentration of collected urine form chilli farmers and non-chilli farmers. The method of creatinine adjustment was done by divided DAP analyze concentration (µg/L) by creatinine concentration (mg/dL). The final unit of adjusted creatinine was micrograms concentration / grams creatinine (Barr et al., 2005).

For chilli farmers, the 1st post-MLA morning void urine was selected for cratinine analysis from each participant, representative of 3 collected urine samples. Before analyze DAP concentration, around 2 mL of the urine was separated from total urine sample and sent to laboratory for creatinine analysis. Other group of participants, non-chilli farmers, around 2 mL of each urine samples were separated as same as chilli farmers.

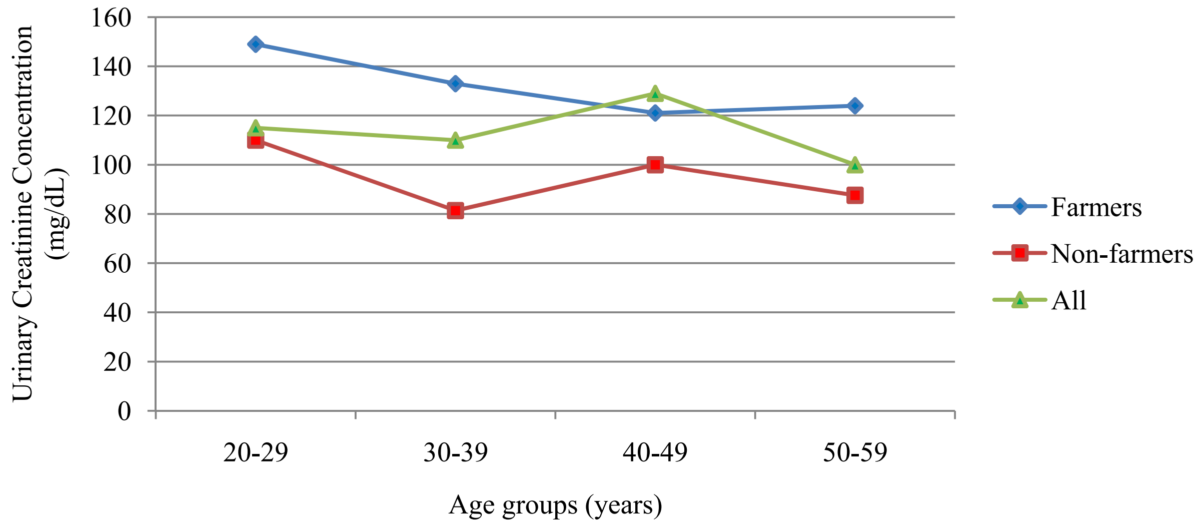
Total 79 urine samples, 39 of chilli farmers and 40 of non-chilli farmers, were sent to laboratory for creatinine analysis. The main reasons of missing samples came from not willing to provide samples from participants. The urinary creatinine concentration means, medians, ranks including 25th, 50th, 75th and 95th percentile were shown in Table 4.22. The results found that average creatinine concentration of chilli farmers (128 mg/dL) was higher than non-chilli farmers (95.8 mg/dL). Mean of urinary creatinine concentration for participants, separated by age groups, was presented in Figure 4.4.

Table 4.22 Urinary creatinine concentration (mg/dL) of chilli farmers and non-chilli farmers in Hua rua sub district, Muang district, Ubon Ratchathani Province

Group	**No.**	**Percentile**				**Rank**	**Mean**	**Median**
		25th	**50th**	**75th**	**95th**			
All	79	68.0	100	146	235	24.0 – 288	112	100
Chilli farmers	39	70.0	122	173	263	46.0 – 288	128	122
Non-chilli farmers	40	64.5	99.0	118	178	24.0 – 182	95.8	99.0

All – Chilli farmers and non-chilli farmers

Figure 4.4 Mean of urinary creatinine concentration (mg/dL) for participants, separated by age groups (years)



4.3.2 Chilli farmers' urinary DAP metabolite levels

The first morning void was collected from Chilli farmers before MLA and 2 following morning void after MLA. Procedure of urinary sample collection and analysis were shown in Chapter 3. All participants were signed the consent from before agreeing to provide urine samples.

The 39 (97.5%) of 40 participated chilli farmers had completely provided 3 morning void urine samples (Pre-MLA, 1st Post-MLA and 2nd Post-MLA). One farmer (2.5%) involved in interviewing section only and did not agree to provide any urine samples.

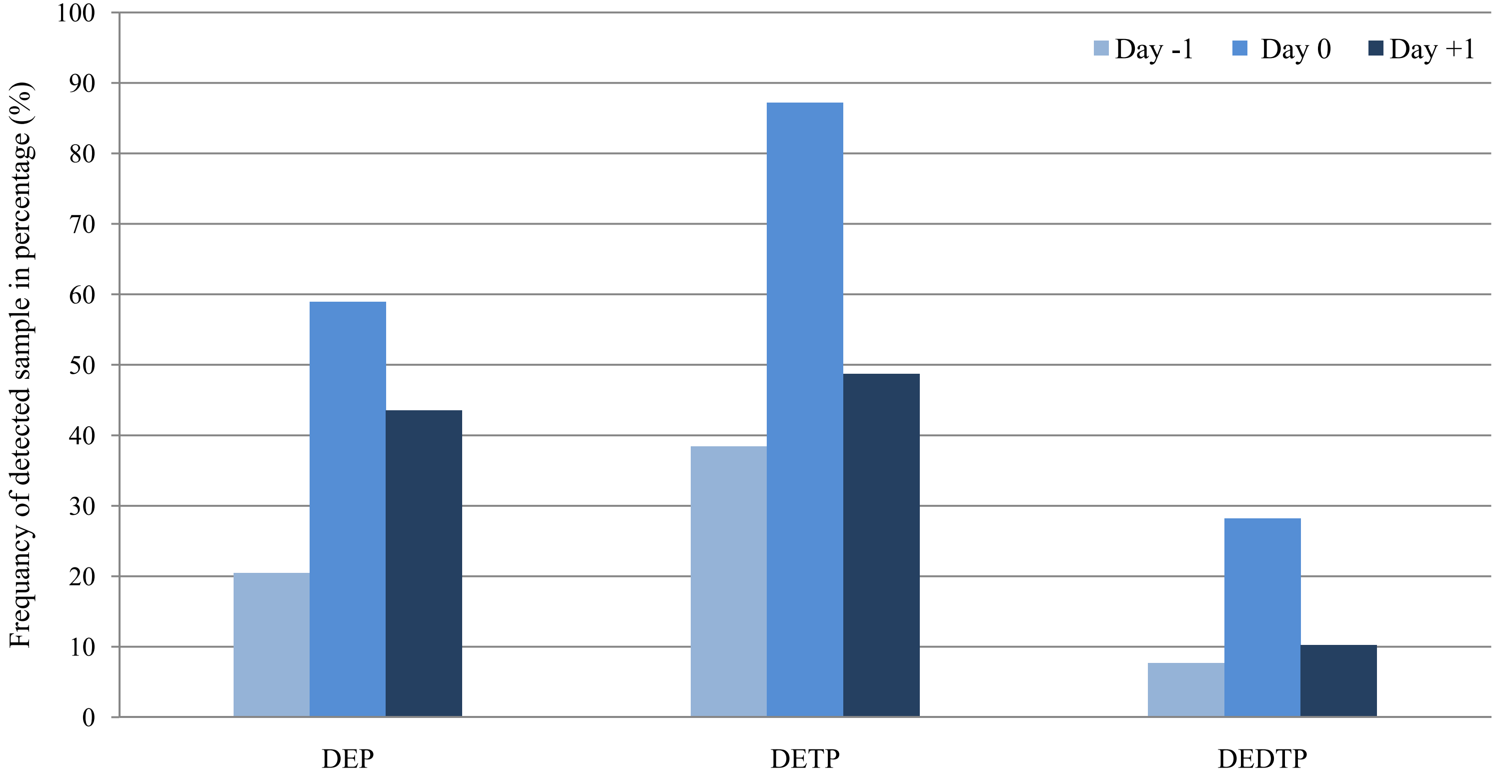
4.3.2.1 Detected samples frequencies of DAP metabolite

The percentage of detected frequency of DAP metabolite, Dimethylphosphate (DMP), Dimethylthiophosphate (DMTP), Dimehyldithiophosphate (DMDTP) ,Diethylphosphate (DEP), Diethylthiophosphate (DETP) and Diethyldithiophosphate (DEDTP), of 3 morning void urine samples was shown in figure 4.5. The dimethyl DAP metabolite (DMP, DMTP and DMDTP) was detected at low percentage of 3 urine sample, and DMDTP did not detected in any urine samples. For diethyl DAP metabolite (DEP, DETP and DEDTP), the 1st morning void following MLA had the highest detected frequencies, especially for DETP (85.7%). The changes of diethyl DAP level was increased from pre-MLA to 1st MLA and then decreased after 1st MLA to 2nd MLA.

From the parent compound in this study, only diethyl DAP metabolite should be detected because of Chlorpyrifos exposure. Profenofos did not produce any DAP metabolite (Bravo et al., 2004). On the first day before MLA, the most detected metabolite DETP (41.7%) followed by DEP (25.0%) and DEDTP (8.33%). The following day after MLA, DETP (85.7%), DEP (65.7%) and DEDTP (25.7%) was detected. On the last day, DAP metabolite was found DETP (48.5%), DEP (42.4%) and DEDTP (12.1%)

MLA – Mixing, Loading and applying pesticide

Figure 4.5 Detected urine sample frequency in percentage (%) of chilli growing farmers (n=39)



Day -1 : Morning void urine before MLA
Day 0 : 1st Morning void urine following MLA
Day +1 : 2nd Morning void urine following MLA

4.3.2.2 Urinary DAP metabolite levels of Chilli farmers

According to percentage of DAP metabolite analysis in this study and the parent exposure compound through muti-pathways (Chlorpyrifos), only diethyl DAPs were presented in this section. Diethy DAPs, composed of DEP, DETP and DEDTP, concentration showed in figure 4.6. The detected metabolites were DETP, DEP and DEDTP, respectively on the day after MLA (day 0).

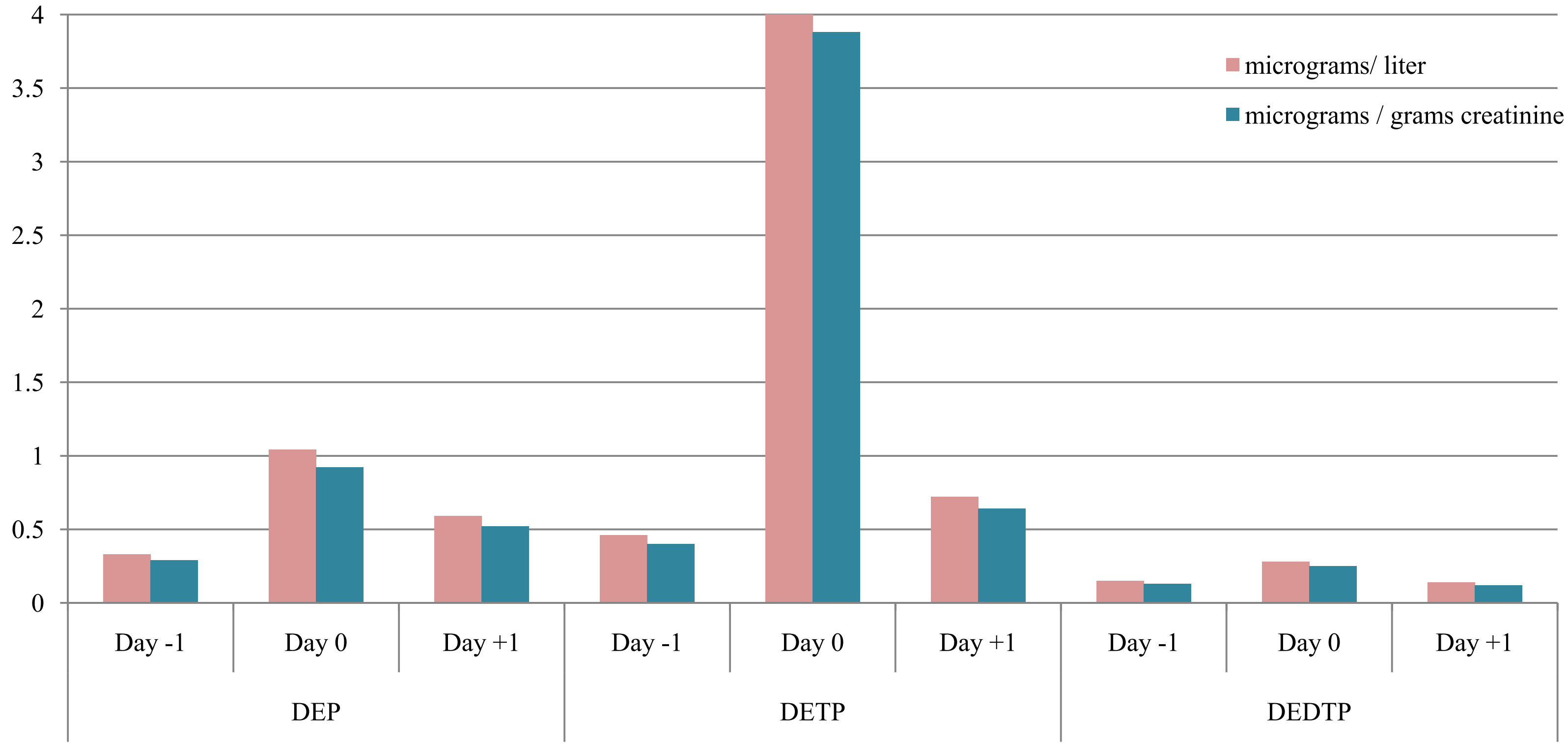
On the day before MLA (day -1), the range of each metabolite concentration was; DEP ranged from <0.20 – 7.06 µg/L (0.13 – 5.41 µg/grams creatinine), DETP ranged from <0.20 – 134 µg/L (0.14 – 143 µg/grams creatinine) and DEDTP ranged from <0.20 – 83.2 µg/L (0.06 – 83.2 µg/grams creatinine).

The 1st post-MLA (day 0), all diethyl DAPs metabolite concentrations were higher than day -1 because occurring from OPPs exposure. DEP could detected ranged from <0.20 – 9.97 µg/L (0.25 – 14.8 µg/grams creatinine), DETP ranged from <0.20 – 125 µg/L (0.82 – 186 µg/grams creatinine) and DEDTP ranged from <0.10 – 135 µg/L (0.06 – 63.7µg/grams creatinine).

Day +1 (the 2nd following MLA), all previous metabolite were detected and higher than day-1. The concentration of DEP ranged from <0.20 – 20.0 µg/L (0.15 – 30.5 µg/grams creatinine), DETP ranged from <0.20 – 15.7 µg/L (0.16 – 17.59 µg/grams creatinine) and DEDTP ranged from <0.20 – 6.02 µg/L (0.06 – 5.37µg/grams creatinine).

The geometric means of DEP, DETP and DEDTP were calculated and presented in figure 4.6. Both adjusted creatinine concentration and non-adjusted creatinine concentration were showed in the unit of micrograms/ liter (µg/L) and micrograms/ grams creatinine (µg/ grams creatinine). On the day-1, DETP was detected higher than DEP and DEDTP as same as day+1. The 1st following day after MLA (day 0), DETP and DEP had geometric mean higher than DEDTP. The more statistical analysis on diethyl DAP concentration including median and percentile (25th, 50th, 75th and 95th) were presented in Appendix E (Table E-1, Table E-2 and Table E-3).

Figure 4.6 Geometric mean concentration of urinary diethyl DAP metabolite concentration (n=39) (Cratinine adjusted and Non creatinine adjusted results)



Day -1 : Morning void urine before MLA
Day 0 : 1st Morning void urine following MLA
Day +1 : 2nd Morning void urine following MLA

Adjusted and unadjusted creatinine of diethyl DAP urinary metabolite concentrations showed in Table 4.23. Total diethyl DAP was calculated by the following equation;

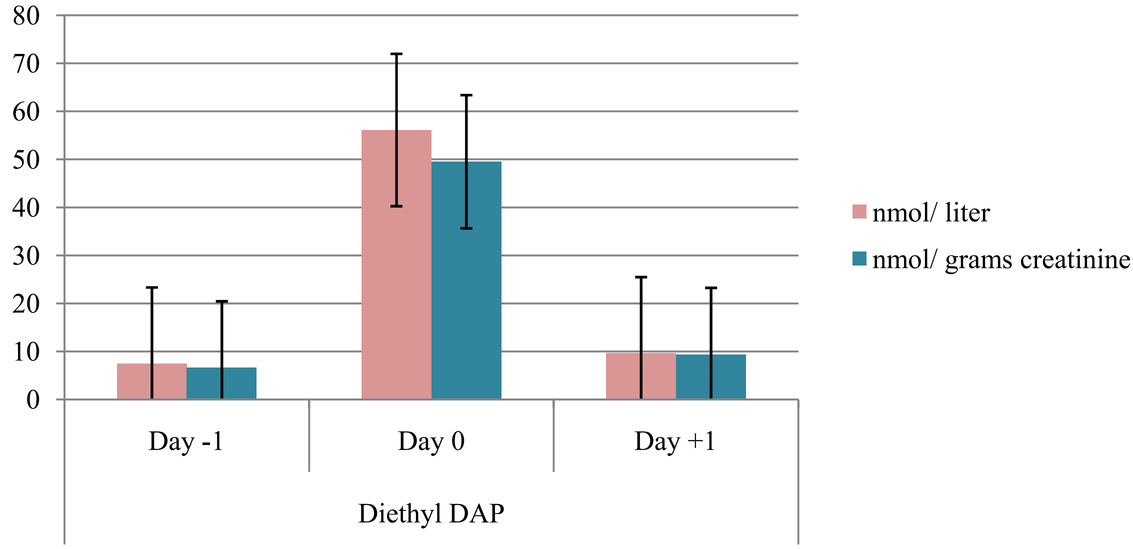
[Diethyl DAP] = [DEP] + [DETP] + [DEDTP] *eq.4-10*

Around 85% of 1st post-MLA samples detected diethyl DAP metabolite and more than a half of 2nd post-MLA samples also. Geometric concentration of pre-MLA, 1st post-MLA and 2nd post-MLA were 7.45 nmol/L (6.58 nmol/g cre.), 56.1 nmol/L (49.5 nmol/g cre.) and 9.60 nmol/L (9.37 nmol/g cre.). The comparison of geometric means presented in figure 4.7.

Table 4.23 Total diethyl DAP urinary metabolite concentration results (n=39)

	No. of detected samples			**Concentration**	
	n	%		Unadjusted Cratinine (nmol/L)	Adjusted Creatinine (nmol/g cre.)
Pre-MLA (Day -1)	17	43.6	Geometric Mean(± SE)	7.45 (±0.23)	6.58 (±24.0)
			Range	3.01 - 792	1.15 – 843
			Median	3.01	4.30
			95th Percentile	450	450
1st Post-MLA (Day 0)	35	89.7	Geometric Mean(± SE)	56.1(±0.36)	49.5(±42.4)
			Range	3.01 – 737	1.15 – 1098
			Median	73.4	60.3
			95th Percentile	736	893
2nd Post-MLA (Day +1)	21	53.8	Geometric Mean(± SE)	9.60(±7.90)	9.37(±9.25)
			Range	0.43 – 185	1.15 – 294
			Median	3.01	4.50
			95th Percentile	178	146

Figure 4.7 Comparison of total diethyl DAP of 3 urine samples of chilli farmers (n=39)



4.3.2.3 Comparison of urinary DAP metabolite levels for pre- and two post-application

The difference of 3 urinary metabolite samples (day-1, day0 and day+1) was found (Friedman test, $p<0.001$). The diethyl DAP urinary metabolite of chilli farmers in the 1st post application (day 0) was different from the day before application (day -1) and 2nd post application (day +1). However, the urinary metabolite level of the day before application was not different from the 2nd post application day (Table 4.24). So, the exposure to OPPs effected to the urinary metabolite level of chilli farmers in this study area. The metabolite was decreased from 1st post application day to 2nd post application statistical significantly.

Table 4.24 Statistical difference of urinary metabolite levels for before and after application

	P-value*
Day 0 & Day -1	< 0.001
Day 0 & Day +1	< 0.001
Day -1 & Day +1	0.131

*Wilcoxon signed ranks test

4.3.2.4 Association between exposure route and urinary metabolite level

Spearman rho's correlation was used to analyze correlation between route of pesticide exposure and urinary metabolite. Dermal and Inhalation routes were indentify as the route of pesticide exposure in this study and the urinary metabolite concentrations were separated to 1st post MLA and 2nd post MLA for analysis. The metabolite of pre-MLA was excluded from this part of analysis. The correlation between dermal exposure and 1st post MLA urinary metabolite was found at moderate level of correlation with statistical significant level (p<0.05) (Table 4.25). Nevertheless, other correlations were not statistical significant in this analysis.

Table 4.25 Association between concentration of exposure route and urinary metabolite

Route	Urinary metabolite	r_r
Dermal	1st post MLA (Day 0)	0.405*
	2nd post MLA (Day +1)	0.205
Inhalation	1st post MLA (Day 0)	0.108
	2nd post MLA (Day +1)	0.175

**Correlation is significant at the 0.05 level (2-tailed)*

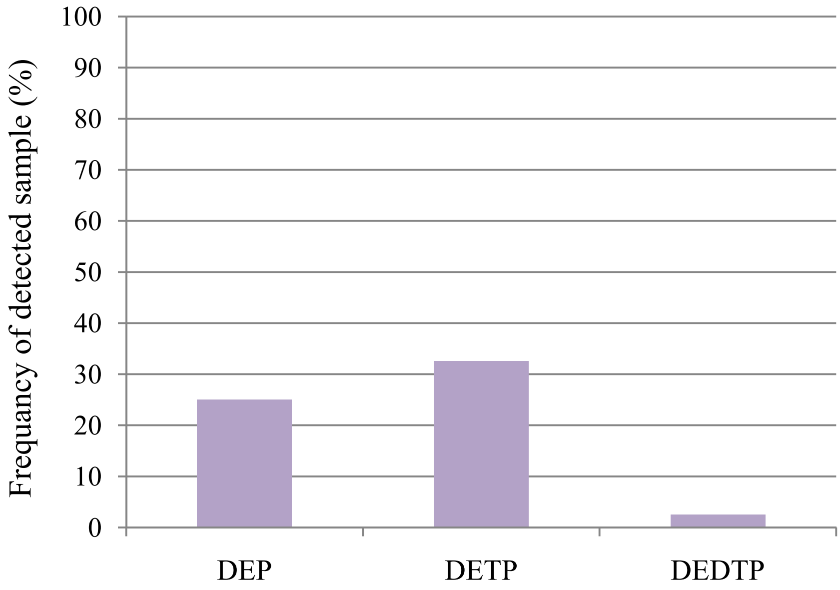
4.3.3 Non-chilli farmers' urinary DAP metabolite levels (Control group)

Urinary samples provided by non-chilli farmers were completely collected and all participants agreed to give the samples. Totally, 40 (100%) morning void urine samples were collected. The samples were collected the day after interviewing day. The urinary collected procedure, similar to chilli farmers, was suggested to participants.

4.3.3.1 Detected samples frequencies of DAPs metabolite

In this study, the main urinary DAPs metabolite, both chilli farmers and non-chilli farmers, was diethyl DAPs (DEP, DETP and DEDTP). The percentage of detected frequency of diethyl DAPs samples were shown in figure 4.8. From 40 samples, the main detected urinary DAPs metabolite was DETP (32.5%), followed by DEP (25%) and DEDTP (2.5%).

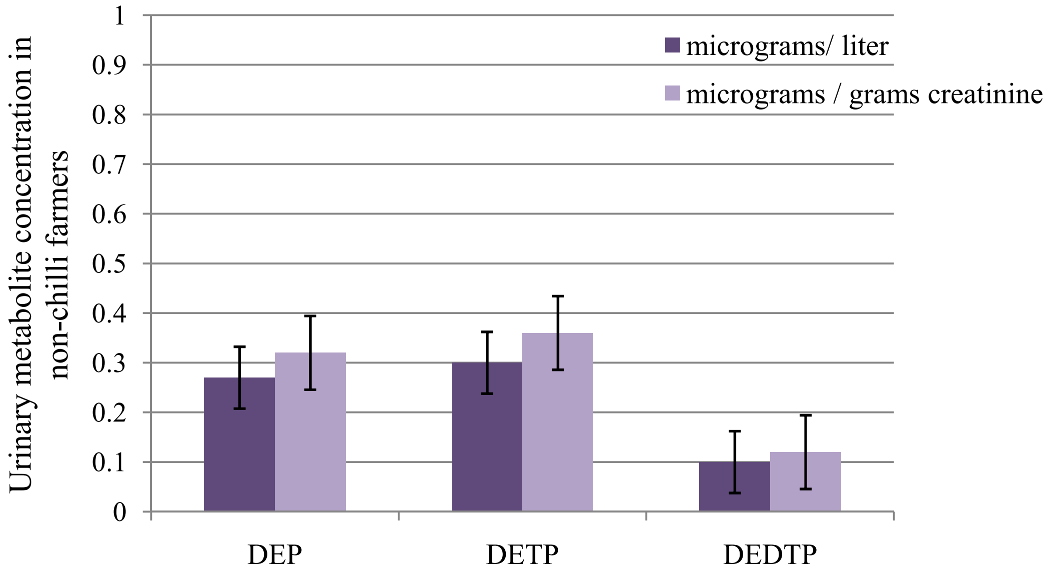
Figure 4.8 Detected urine sample frequency (%) of non-chilli farmers (n=40)



4.3.3.2 Urinary DAP metabolite levels of non-chilli farmers

Concentrations of urinary diethyl DAPs metabolite were presented in unadjusted and adjusted creatinine concentrations. The concentrations of diethyl DAPs metabolite were range from below 3.01 to 61.28 µg/L (1.66 -53.8 µg/g creatinine); separated to DEP ranged from <0.20 – 5.45 µg/L (0.17 – 4.78 µg/g creatinine), DETP ranged from <0.20 – 4.31 µg/L (0.17 – 6.06 µg/g creatinine) and ranged from <0.10 – 0.10 µg/L (0.08 – 0.42 µg/g creatinine) (Appendix F; Table F-1)

Figure 4.9 geometric mean urinary metabolite concentrations of non-chilli farmers (n = 40)



The geometric mean concentrations were presented in term of comparison between each metabolite. The adjusted creatinine geometric mean had higher concentration than unadjusted (figure 4.9). DEP, DETP and DEDTP concentration were 0.27 µg/L (0.32 µg/g creatinine), 0.30 µg/L (0.36 µg/g creatinine) and 0.10µg/L (0.12 µg/g creatinine), respectively.

Geometric mean concentration equaled to 4.31 µg/L (5.12 µg/g creatinine). The median and 95th percentile was also presented (Table 4.26).

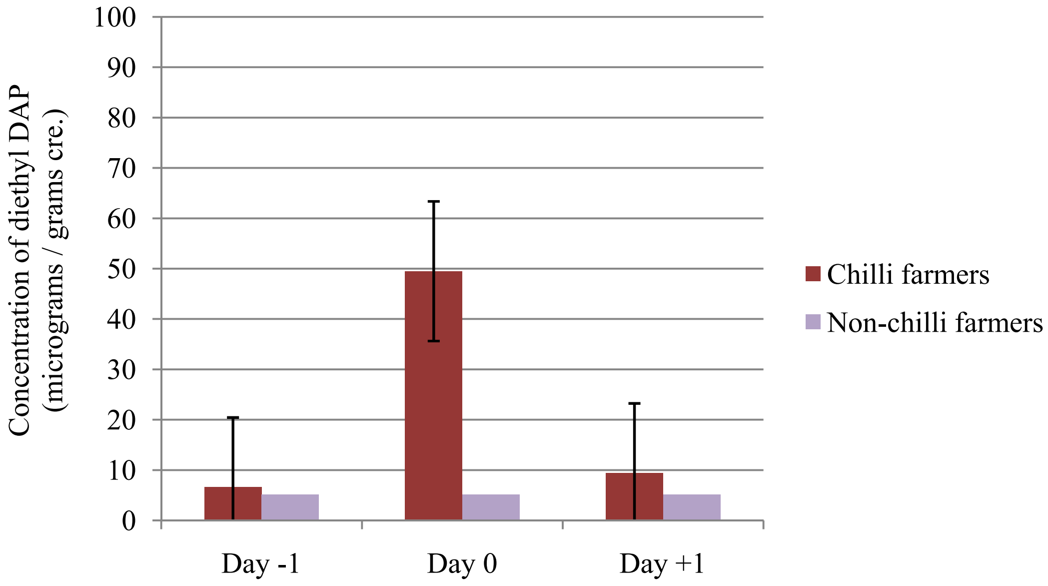
Table 4.26 Total diethyl DAP urinary metabolite concentration of non-chilli farmers results

	No. of detected samples			Concentration	
	n	**%**		Unadjusted Cratinine (µg/L)	Adjusted Creatinine (µg/g cre.)
Diethyl DAPs	11	27.5	Geometric Mean(±SE)	4.31(±1.63)	5.12(±1.86)
			Range	3.01 -61.3	1.66 – 53.75
			Median	3.01	3.76
			95th Percentile	30.9	41.8

4.3.4 Comparison of urinary DAP metabolite levels between chilli farmers and non-chilli farmer

Differentiate between chilli farmers and non chilli farmers urinary metabolite was tested. There was a different between urinary metabolite of non-chilli farmers and chilli farmers on the day after MLA (day 0) (Mann-Whitney U test; $p<0.001$). However, urinary metabolite of the day before MLA (day -1) and the 2nd day after MLA (day+1) of chilli farmers had no statistical different with non chilli farmers urinary metabolite. The geometric mean concentration of non chilli farmers and chilli farmers, separated by day, was show in the figure 4.10.

Figure 4.10 Geometric means concentration of both chilli farmers (n=39) and non chilli farmers (n=40)



CHAPTER V

DISSCUSSION

5.1 General Information and health effects related to pesticide exposure

5.1.1 General characteristics of participants

Most chilli farmers in this study were male in the middle age (30-39 years old) which was the same as previous studies (Ngowi at al., 2007; Perry et al., 2006), however there are some female involving in the study (Mancini et al., 2005). Normally, an agricultural activity in developing counties was done by men. The middle age was found as a subject in this research as same as previous. Body mass index (BMI) of both chilli farmers and non-chilli farmers were classified in the normal range (18.5 – 24.9) by WHO (1995).

More lack of education lead to more dose of pesticide exposure. Elementary education was the major group of population in this research. Safety instructions on containers are often written in unfamiliar languages, many farmers are illiterate, and the instructions themselves are difficult to follow (Eddleston et al., 2002). The comparable of education level between chilli farmers and non-chilli farmers was not found in this study. Most non-chilli farmers were reported occupation as government employees and local business owners so it leaded to the different of education levels. Thus, it could be affected to the knowledge and concern about pesticide exposure protection. Populations with little formal education might be at higher risk when using pesticides, possibly due to difficulties in understanding the use instructions and safety procedures included on the product labels. However, no significant correlation was found between reporting of symptoms related to pesticide exposure and education level (Recenaa et al., 2006).

Twenty five percent of chilli farmer and 15% of non-chilli farmers were reported as smoker. Comparing to rice farmers in the central of Thailand, the figure found that 28.6% of rice farmers, surveyed by Pan (2009), was smokers which higher than this study participants. Similar to a study of Jintana et al. (2009) demonstrated that applicator of OP pesticide (fruit farmers) in Rachaburi province (central of Thailand) was smoker around 26.7%.

5.1.2 Chilli farmer information related to pesticide exposure

From researcher observation, the missing and strictly use of personal protective equipment (PPE) were detected once because of weather and humidity of Thailand. The suitable and/or complete PPE were not available because of its pricing and not wildly using in the study area, rural area of the country. Thus, most farmers had to develop or made their own traditional PPE for themselves, not only to protect from pesticide exposure but also to reduce their expenditure. Simply and easy finding material in the area were used as PPE, such as using plastic bag as coverall, using helmet covering their head instead of mask and hat (Figure 5.1), wearing sock together with boots. However, rubber gloves and rubber booths were normally used in this study population because of reusable.

Figure 5.1 Inappropriate and misunderstanding of PPE in Chilli growing farmer (using helmet covering their head instead of mask and hat)

In generally, there were 2 types of sprayers, backpack sprayer (Figure 5.2b) and sprayer connected with motor and tank (Figure 5.2a), which chilli growing farmers used to spray pesticide. From the types of sprayers, the different pesticide contact should be found; the sprayer using man-power should have less pesticide contact than others. Spraying with a hand-pressurized backpack increased the visual score compared with the motor-pressurized backpack. The type of backpack sprayer determined the skin exposure of the farmers partly by influencing working practices, for example, using a hand-pressurized backpack was related to the practice of spraying with the nozzle directed straight ahead and with a shorter nozzle–applicator body distance (Blanco et al., 2005).

(a) (b)

Figure 5.2 Type of sprayers in the study area

(sprayer connected with motor and tank (a) and backpack sprayer (b))

Around 1000 USD was annually earned from chilli crop by the farmers. The income is associated with the inequity of environmental exposure on the job (Evans and Kantrowitz, 2002). Low-income marginal farmers were more often prone to severe poisoning (Mancini et al., 2005). Most chilli growing area was small; the average area was 1 acre approximately. Thus, the short time of pesticide activity was spent for each time of pesticide spraying. Mancini et al., 2005 reported that pesticide toxicity and exposure time were positively correlated with the extent to health symptoms related to pesticide exposure.

The use of PPE was mostly indicated from participants. They reports that they usually worn all PPE, especially long sleeved shirts. However, most of them mention as always worn long legged pants. Hat and Nose mask were reported as never from few chilli farmers. They reported that wearing hat was inconvenience during application. Moreover, misunderstanding of using hat was found; some farmers said that they always sprayed pesticide in the morning so they did not need to protect themselves from sunlight.

Most chilli farmer did not use the amount of pesticide following the instruction because they believed that the huge amount could protect pest with high efficiency. The previous study mentioned that Thai farmers are at great risk of pesticide poisoning largely because of inappropriate pesticide handling, improper use of personal protective equipment (PPE) as well as inadequate understanding of the toxicity of the chemicals with which they work (Panuwet et al., 2008).

From this study, family members could be exposed to pesticide by indirect exposure through storing pesticide at home, washing working cloths with their family. Other study of farmers' family exposure indicated that storage of pesticides in the home may lead to accidental exposure of household members and was more likely on smaller farms. Moreover, laundry methods for clothes worn when mixing or applying pesticides was another factor mentioned in this previous study (Gladen et al., 1998). Meal was sometimes taken with farmers to the farm, however other farmers reported that they finished their meal before work because their houses were close to the farm area.

5.1.3 Health effects related to pesticide exposure

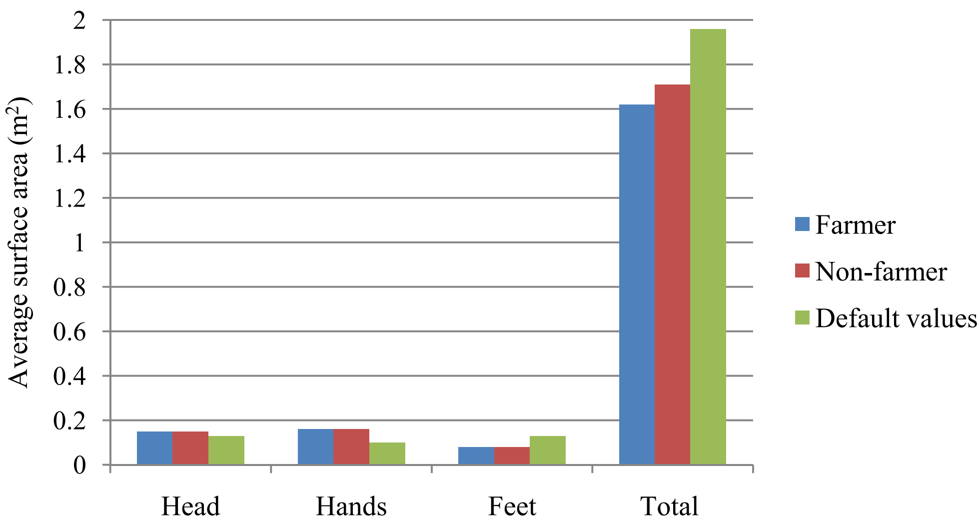
Most chilli farmers mentioned their health effects related to Central Nervous System (CNS). Several studied suggest that the main of pesticide related to illness are due to exposure that happens while working in the field (Strong et al., 2004). Irritation of the throat and cough related to respiratory symptoms was diagnosed from most participants. Excessive sweat and excessive salivation were the major heath symptoms. Ngowi et al. (2007) mentioned that excessive sweat, headache and dermal effects were more commonly reported. Memory problem was the first priority mentioned in this study which as same as the most frequency reported in Ribeiro et al. (2012) study.

The association between PPE and health symptoms was found. Form this study, the association was similar to Esechie et al. (2011). Therefore, these results indicate a relationship between nose mask usage and a reduced incidence of vomiting. Nevertheless, there is no doubt that the uses of PPE reduce pesticide exposure and the incidence of acute and chronic poisonings among these participants. Although these may not necessarily be direct "cause and effect" relationships, they do suggest that the use of PPE during pesticide application can greatly reduce the incidence of exposure and that the supplement is true (Esechie et al., 2011).

5.1.4 Body surface area calculation

Comparison of differentiation between defaults values (US EPA, 2011) and calculated body surface area for specific participants (male & female) presented in figure 5.3. Participants had head and hand surface area more than default values, however, feet and total surface area were less than. Due to the calculation of surface area, weight and height were directly considered by using the surface area calculated equation. However, the default surface area values, recommended by US EPA (2011), were not shown the default values of weight and height which use to calculated. Thus, the comparison between each factor in the equation could not be done. The difference of body surface area between male and female was affected directly to pesticide exposure through dermal route. The more surface area will get more pesticide contact. It showed that male will get higher dose of pesticide exposure more than female.

Figure 5.3 Comparison between calculated body surface area values and default body surface area values



5.2 Pesticide exposure concentration of farmers via multi-exposure pathways

5.2.1 Chilli and non-chilli farmers' pesticide exposure

From detected sample frequencies, no foot wipe and drinking water were detected OPPs residues. Face wipe and Body patch samples could detect both Chlorpyrifos and Profenofos with difference in percentage. The air sample detected Chlorpyrifos more than Profenofos around 80%.

Form researcher observation, foot wipe samples was not detected because all chilli farmers worn socks together with boots during MLA. They reported that they felt comfortable of wearing socks with boots more than putting on only their boots. The humidity and weather of Thailand can be caused of sweating during MLA of chilli farmers, so socks might be absorbed their sweat on their feet.

Drinking water samples in this study area were not detected of any OPPs because of several reasons. Firstly, OPPs are easily degradation (Freed et al., 1979), so it might be not detected or degrade from transportation process. The process of producing drinking water was another supportive reason. Normally, water was boiling for being drinking water for Thai. Moreover, participants in this study area use ground water for drinking and it could not detected residue of organophosphate pesticide in 2010. But, Younes and Gorchev (2000) reported that organophosphorus pesticides are readily hydrolysed in water and seldom found in drinking-water.

Around 30% of all face wipe samples were detected both Chlorpyrifos (Average = 0.044 mg/kg) and Profenofos (Average = 0.513 mg/kg). Similar to Schneider et al. (1992) studied the pesticide (Azinphosmethyl) residue on face/neck wipe of Peach Harvest Workers and found the concentration <0.002-0.05 mg for the face/neck wipes. Unlikely, the study of Aprea et al. (2001) found that the exposed face pads for had the concentration of pesticide residue below the detection limits.

Hand wipe samples were detected lower than face wipe samples. Ranking of Chlorpyrifos was found <0.02 - 0.240 mg/kg and Profenofos was ranked from <0.02 –

0.450 mg/kg. Similar to Curwin et al., (2005) study, the study conducted on farmers in Iowa, United State and found most of hand wipe samples were no detectable. The concentration which found in this study was less than the concentration of chlorpyrifos concentration exposure in the last study of Taneepanichskul et al. (2010) equal to 6.95 ±18.24 mg/kg/two hands. Moreover, the concentration of Chlorpyrifos and Profenofos in this study was lower than Pan et al. (2009) study. She found the rank of Chlorpyrifos was 0.29-105.62 mg/kg and rank of Profenofos was 0.51 – 22.86 mg/kg. However, the hands were always a source of contact with the pesticides (Aprea et al., 2001). The higher dose of pesticide exposure could also found from hand contact more than other part of body because of the concentrated pesticide during load to the sprayer equipment.

Profenofos and Chlorpyrifos were detected from chilli farmers in the ranked <0.001 – 0.010 mg/m^3 and <0.001 – 0.006 mg/m^3, respectively. Chilli farmers in this study were not exposed to organophosphate pesticide exceeded the time-weighted average (TWA) limit of the American Conference of Governmental Industrial Hygienist (ACGIH) recommendation value, equal to 0.02 mg/m^3. Similar to, Kongtip et al. (2009) found that the average occupational chlorpyifos exposure among rice farmers in Phatthalung Province was 0.062 ± 0.092 mg/m^3. Other study of ambient air breathed by farmers, conducted in Tambon Bang Rieng, Thailand, found the concentration of chlorpyrifos and methyl parathion ranging from 0.004 to 0.61 mg/m^3. Moreover, this study demonstrated that traditional farmers (0.19 mg/m^3) were exposed to the pesticides higher than integrated pest management farmers (0.037 mg/m^3) (Jirachaiyabhas et al., 2004). The air sample detected Chlorpyrifos more than Profenofos around 80% due to the solubility of the chemical property. Chlorpyrifos has the higher solubility than Profenofos, so this property could be effected to the process of laboratory analysis. Profenofos has limited solubility in water at 20 ppm but is completely soluble in organic solvents (ethanol, acetone, toluene, n-octanol, and n-hexane) (US EPA, 2006) and Chlorpyrifos has limited solubility in water at 1.05 ppm and soluble in toluene, analysis chemical, more than 400 g/L (Cattani, 2004).

Correlation between exposure routes

The test of association between dermal exposure and inhalation exposure of chilli farmers in this study found that there was not correlated in linear model (R^2=0.003) and test of association was not statistical significantly (Spearman rho's test; $p > 0.05$). Nevertheless, the correlation between hand contact to pesticide and inhalation was found (Spearman rho's test; $p < 0.05$). Likely, Aprea et al. (2001) suggested that the air concentration increases substantially the face pad concentration then increases proportionally and respiratory and dermal potential exposure may vary significantly across different organophospate pesticides.

Exposure assessment and risk characterization

Dermal Exposure Assessment

The highest ADD was found in body skin contact to OPPs according to the larger surface area than others. The ADD of Profenofos (17.1×10^{-11}) on face contact was higher than Chlorpyrifos (1.47×10^{-11}) at both mean. In the same way, hand contact of Profenofos was also more than Chlorpyrifos.

Hazard Quotients (HQ) for Chlorpyrifos and Profenofos at both mean and RME were not exceed the acceptable level for chilli growing farmers.

Inhalation Exposure Assessment

At mean level, the ADD of Chlorpyrifos (5.71×10^{-6} mg/kg-day) was higher than Profnofos (1.71×10^{-6} mg/kg-day). From Jirachaiyabhas et al. (2004) study, the calculated ADD of organophosphate pesticide inhalation was done and found that the traditional farmers absorbed 0.0006–0.0224 mg/kg-day of the pesticide, separated to 6.2–224% of chlorpyrifos or 3.1–112% of methyl parathion.The same figure of HQ and HI came from the lower HQ of Profenofos of inhalation (close to zero), thus it was not affected in the summation equation. Nevertheless, the RfD of Chlorpyrifos was lower than Profenofos for inhalation route. HQ of Chlorpyrifos seemed closed to 1 than Profenofos, it could be explained that chilli farmers got risk from Chlorpyrifos exposure more than profenofos. But, the HQ value was still not exceed than "1" so the adverse health effected was not need to concern for this population via inhalation during pesticide application.

The detectable of non-chilli farmers sample was found only air samples (20%). The small figure was found because of the indirect exposure to pesticide. The air was detected pesticide from the houses closed to farm area and the wind direction also affected. Agricultural pesticide detection was associated with housing adjacent to agricultural fields. The exposure has been considered high for farmworker families (Quandt et al., 2004).

5.3 Biological sample results: chilli farmers and non-chilli farmers

5.3.1 Creatinine Concentration in chilli farmers and non-farmers

Age and creatinine concentration was not associated from this study participants (Spearman rho correlation test: $p>0.05$) The highest concentration was found in 40-49 age group and decreased as the following age group (50-59 years). However, the association of creatinine concentration and age group in this study was not the same as previous study of Barr, et al. (2005) which found that in the U.S. population the creatinine concentration was reduced by age after the age rank of 20-29 years.

The guidelines of the World Health Organization (WHO) for valid urine samples for occupational monitoring, recommended that if a sample has creatinine concentration less than 30 mg/dL means too dilute for creatinine and the target chemical analysis. In the same way, the creatinine concentration more than 300 mg/dL means too concentrated for analysis (WHO 1996 cited in Barr et al., 2005). In this study, the too concentrated creatinine analysis was not found and 3 samples found cratinine concentration less than 30 mg/dL (too diluted). Nevertheless, the U.S. Department of Transportation accepted urine specimen which has a creatinine concentration more than 5 mg/dL for the screening of selected drugs of abuse (Barbanel et al. 2002). Thus, the too diluted creatinine samples were not cut off from this study because urine of "normal" persons would be unlikely to be excluded (Barbanel et al. 2002).

5.3.2 Chilli farmers' urinary DAP metabolite levels

Urinary DAP metabolite levels of Chilli farmers

Urinary metabolite level in this study found that geometric concentration of pre-MLA, 1st post-MLA and 2nd post-MLA were 6.58 nmol/g creatinine , 49.5 nmol/g creatinine. and 9.37 nmol/g creatinine, respectively. The highest detected frequency was the 1st post-MLA, 2nd post-MLA and pre-MLA. From the results, it could explain that the urinary half-lives for dialkylphosphate metabolites through dermal dose was 30 hours (Thomas et al., 2009).

Another Urinary DAP study with Thai farmers found that levels with average (geometric mean) levels are of 51.1 mg/g for vegetable farmers and 122.2 mg/g for fruit farmers (Hanchenlaksh et al.,2011). Moreover, Panuwet et al. (2008) assessed exposure pesticides of male farmers in Chiang Mai Province, Thailand and pointed out that no significant differences in metabolite concentrations of two farmer groups with differentiate in topographically different area. Moreover, Blair et al. (2011) demonstrated that correlations of urinary levels with kilograms of active ingredient used, duration of application, or number of acres treated were lower.

Sudakin and Stone (2011) pointed out that "the *in vivo* metabolism of organophosphates yields different DAPs, depending upon whether they undergo bioactivation or detoxification. The detection of urinary DAPs does not provide specificity with respect to the organophosphate from which they were derived, or their toxicological potency. Several recent studies documented the common presence of DAPs in residential environments and foods. Experimental studies support that DAPs have significant oral bioavailability, and undergo little to no metabolism prior to urinary excretion".

Association between exposure route and urinary metabolite level

In this study, the correlation between dermal exposure and 1st post MLA urinary metabolite was found at statistical significant level ($p<0.05$). Similar to Curwin et al. (2005) study, he found that "most hand wipe samples were non-detectable, however, detection of atrazine in the hand wipes was significantly associated with urinary levels of atrazine above the median". Unlikely, Bradman et al. (2007) demonstrated that total diethyl phosphate metabolites were weakly or negatively correlated with levels of chlorpyrifos in environmental media. Moreover, from Curl et al. (2002) found an association between dimethyl DAP levels in adult farmers urinary metabolite and exposure, however, the dimethyl DAP metabolite could posed to not only agricultural product exposure but also variety of OPPs. Furthermore, dermal exposure on day one correlated with total metabolites (DMP + DMTP) collected the following morning and total metabolites collected after 48 hours were less well correlated. Multiple regression analysis showed that urinary alkylphosphate was significantly correlated with the respiratory doses.

The highly detectable percentage of inhalation exposure was found from this study, nevertheless the correlation between this route and urinary metabolite was not found. The soluble of concentration of pesticide exposure through inhalation by the air could be explained this finding.

5.4 Risk Communication

Risk communication was defined as ''an interactive process of exchange of information and opinion among individuals, groups and institutions. It involves multiple messages about the existence, nature, form, severity or acceptability of health risks''. Risk communication plan must be sound, with effective strategies, monitoring and evaluation to ensure the desired objectives are achieved. The planning requires expertise in various fields, such as program planning, evaluation, communications theory, and public health practice (Tinker et al., 2000). In this study, the development of risk communication material was calendar connected with communicated using

PPE picture for encouraging chilli farmers to realize and concern their health (Appendix H).

In term of generalizability, this study could be generalized to other agricultural area where was growing the small plants as chilli. Due to the difference method of applying pesticides, the useful of this research was limited by the height of plants and the area of cultivation. However, the good manner of handling and practicing of pesticide and PPE usage, such as not storing pesticide at home and washing working clothes separately, could be introduced and suggested to other area by risk communication material; books and/or manuscript.

For policy implementation, this study could be suggested that the government should concentrate on dermal exposure, especially body contact. Because of percentage of detectable samples, the proportion between dermal contact and excretion of urinary metabolite was highly correlated (figure 5.4). To suggest the plan or policy to the government sector, the only body contact will be measured if the budget is limited. There are reasons to support the suggestion from this study. Firstly, the correlation between dermal contact and urinary metabolite level on the day after spraying was found from this study. Moreover, the percentage of detectable sample of body contact was the highest comparing to other dermal contact frequency. The frequency of inhalation was the highest detectable, however the correlation was not found. So, the policy implementation and plan was not recommended.

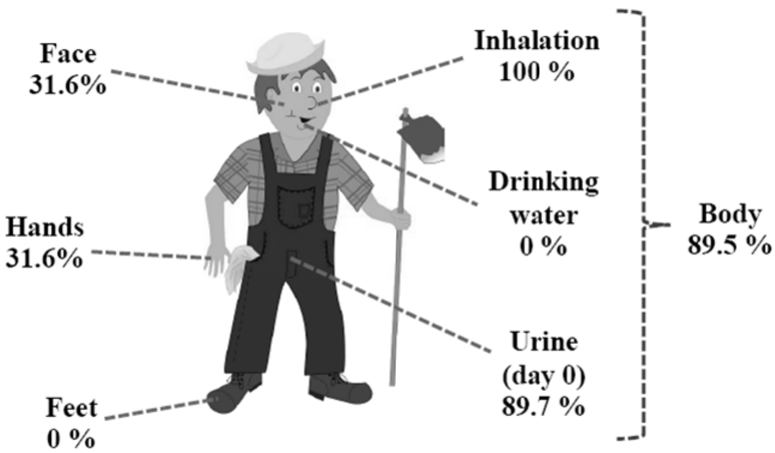


Figure 5.4 Detected frequencies in percentage of pesticide exposure multi-pathways

CHAPTER VI
CONCLUSIONS

6.1 Conclusion

To assess human exposure and health effects of common pesticides exposure through multi-exposure pathways, different measurement tools were used in this study. From the findings in this study, it would be concluded that chilli farmers had exposed to o+rganophosphate pesticides (Chlorpyrifos and Profenofos) through dermal and inhalation during their pesticide application. However, they were not classified in risk level of exposure by risk assessment procedure. Some common health effects form pesticide exposures were manifested in this farmers group.

Most chilli farmers in this study were male in the middle age (30-39 years old) and had only elementary education. Around 1000 USD was annually earned from chilli crop by the farmers Most chilli growing area were small; the average area was 1 acre approximately thus, the short time of pesticide activity was spent. The use of PPE was mostly indicated from chilli farmers. They usually wore all PPE, especially long sleeved shirts and long legged plants. Most chilli farmers mentioned their health effects related to Central Nervous System (CNS). Irritation of the throat and cough related to respiratory symptoms was diagnosed from most participants. The association between PPE and health symptoms was found, such as wearing mask and vomit and chest pain ($p<0.01$).

In the process of surface area calculation, participants had head and hand surface area more than default values recommended by US EPA, however, feet and total surface area were less than. From detected sample frequencies, no foot wipe and drinking water were detected OPPs residues. Face wipes and Body patch samples could detect both Chlorpyrifos and Profenofos with difference in percentage. The air sample detected Chlorpyrifos more than Profenofos around 80%. There was no association between dermal exposure and inhalation exposure of chilli farmers both in linear model (R^2=0.003) and Spearman rho's test ($p > 0.05$). However, the correlation between hand contact to pesticide and inhalation was found (Spearman rho's test; $p < 0.05$).

The highest ADD was found in body skin contact to OPPs according to the larger surface area than others. The ADD of Profenofos (17.1×10^{-11} mg/kg-day) on face contact was higher than Chlorpyrifos (1.47×10^{-11} mg/kg-day) at both mean. In the same way, hand contact of Profenofos was also more than Chlorpyrifos. Inhalation At mean level, the ADD of Chlorpyrifos (5.71×10^{-6} mg/kg-day) was higher than Profnofos (1.71×10^{-6} mg/kg-day). . Both hazard Quotients (HQ) and hazard index for Chlorpyrifos and Profenofos at both mean and RME did not exceed the acceptable level for chilli growing farmers in any route of exposure. It would be concluded that chilli farmers in this area were not at risk from OPPs exposure.

For creatinine analysis of urinary metabolite, age and creatinine concentration was not associated from this study participants (Spearman rho correlation test: $p>0.05$). Urinary metabolite level in this study found that geometric concentration of pre-MLA, 1st post-MLA and 2nd post-MLA were 6.58 nmol/g creatinine , 49.5 nmol/g creatinine. and 9.37 nmol/g creatinine, respectively. The highest detected frequency was the 1st post-MLA, 2nd post-MLA and pre-MLA. Correlation between dermal exposure and 1st post MLA urinary metabolite was found at statistical significant level ($p<0.05$).

6.2 Benefit from this study

1. To identified the specific pathway which chilli-growing farmer will be get higher risk than other routes
2. To identify a relationship between exposure routes and biological monitoring
3. To predict an association between health effects and pesticide exposure by using the model
4. Policy implementation and risk communication will be introduce to the community after finding the important pathways of pesticide exposure in order to keep sustainable behavior improvement of chilli-growing farmers in this community.
5. The association model will be apply to other farmers' community where use the similar pesticides and pattern of spraying pesticide.
6. Farmers, who have level of urinary metabolite over than usual, will be suggested to have a blood test at primary health care unit to confirm the results of analysis
7. The researcher will be introduced the way to protect themselves from pesticide exposure such as using personal protective equipment and reducing dose of pesticides usage. The monthly health check up at primary health care will be suggested to some farmers.

6.3 Limitation of the study

1. This study was concentrated on only chilli growing seasons in the year round, however there is another plants, such as rice, was grown in this area. Thus, the effected of difference pesticides could be found.
2. There were common pesticides that this study was focused. Other kinds of pesticides and herbicide which widely used in this area were not investigated. Thus, subjective signs and symptoms reports which were associated with common pesticides exposure might be caused by other pesticides.
3. Bias could recall from subjective signs and symptoms reports from chilli farmers and non-chilli farmers. The diagnosis was not done by the physician.

4. Weight and height was reported by participants. The equipment was not used to measure, thus the estimated weight and height could be found because the figure could usually change.
5. The comparable of education level was not found between two group of participants; chilli farmers and non-chilli farmers.
6. Type of sprayer equipments should be identified from chilli farmers because the different dose of pesticide exposure could be found.
7. For urinary metabolite part, only diethyl DAP was analysis in this study. The TCPY (3,5,6-trichloro-2-pyridinol) metabolite of Chlorpyrifos, dimethyl DAP and other urinary metabolite from pesticide exposure should be analyzed in order to find other kind of parent compound of pesticide in this study area.
8. The finding could be generalized to only the agricultural area where are growing low and/or small plants because of the different spraying method and equipments.

6.4 Recommendation for further study

1. The specific body skin samples, form 7 positions patches of dermal exposure procedure recommended by WHO (Johnson et al., 2004), should be analyzed to find the higher concentration of body skin contact. The finding could be suggested to chilli farmers to concern that part more than others.
2. Other period of chilli growing, harvested season, should be investigated from this study area because different kinds of pesticides and different dose might be used.
3. Pesticide exposure could also investigate by blood, but it could not specific the type of pesticide from exposure. However, the finger tip of blood checking should be done for confirmation of the finding in this study.
4. To complete the process of risk communication, the handbook and education program related to pesticide exposure protection should be done for this community. Suggestion of handling and practicing of pesticide use and

personal protective equipment should be added to the program to increase health awareness and health concern.

5. To know the benefits of risk communication material, evaluation in this area should be done in the future. If the pesticide contact and/or exposure of chilli farmers in this study area reduces, the effectiveness of this material will be suggested to other agricultural area.

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APPENDICES

Appendix A

Questionnaire (English)

Interviewer’s name_____________________ Code.______

Pesticide Exposure and Health Effects Questionnaire

Description

Questionnaire is separated into 3 parts; first and second parts consisted of opened and closed questions, the last past has only closed question. The details are showed as following:

Part 1: General Information

The questions ask about subjects’ background information including agricultural works and farming descriptions, handling and practicing of pesticides and personal protective equipment (PPE).

Part 2: Exposure Information

The information in this part is based on pesticide exposure frequency through dermal contact, inhalation and drinking water (oral route) in order to assess the risk for chilli farmers.

Part 3: Health effects of pesticide exposure

The general health effects related to pesticide exposure are put in this part. Farmers will report after their farm activities or within 24 hours after expose to pesticide in order to assess acute health effects.

****

<u>Part I: General Information</u>

Gender ☐ Male ☐ Female

Weight ________________Kilogram

Height ________________Centimeters

Education Background

- ☐ Uneducated
- ☐ Primary School
- ☐ Secondary School
- ☐ College Graduate
- ☐ Bachelor or higher

Smoking

- ☐ Never
- ☐ Ever
- ☐ Current smoke
    - o Type ____________________
    - o No. of cigarette / day ____________________
    - o How often? ____________________

Income (chilli crop/ year) ________________Bath

<u>Agricultural works and farming descriptions</u>

Area cultivated ________________Rai

Years working in agriculture ________________Years

Farming tasks of chilli growing

- ☐ Mixing
- ☐ Loading
- ☐ Applying

Years using pesticides _______________years ________________months

Number of annual pesticide applications ____________________times

Backpack sprayer condition

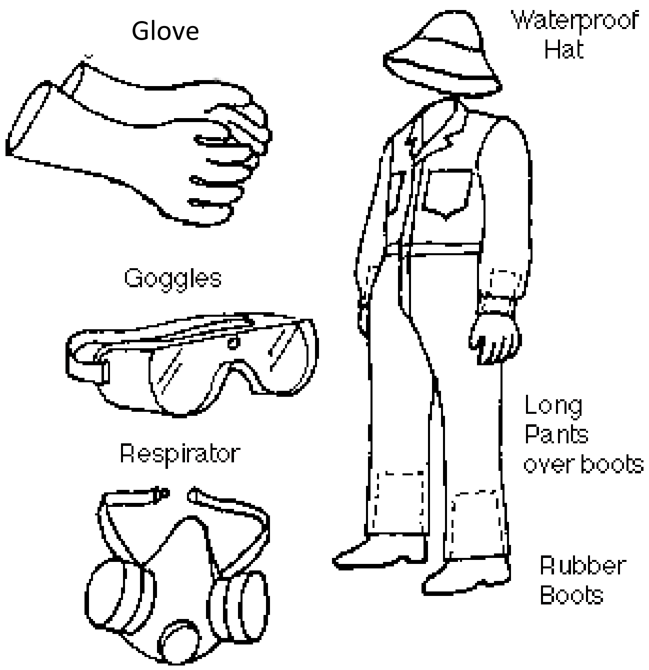
- ☐ Leaking
- ☐ Not Leaking

Handling and practicing of pesticides

	Every times	Sometimes	Never
Using the required amounts of pesticides			
Preparing (mixing) pesticides at home			
Storing pesticides at home			
Washing working clothes with the family clothes			
Cleaning spraying equipment after work			
Taking a meal at work place			
Smoking while applying pesticides			
Considering the safety period			

Personal protective equipment (PPE)

	Every times	Sometimes	Never
Use of Gloves ☐ Rubber ☐ Fabric ☐ Long ☐ Short			
Use of mask			
Use of boots ☐ Long ☐ Short			
Use of hat			
Use of short sleeved shirt			
Use of long sleeved shirt			
Use of short legged plants			
Use of long legged plants			



Part 2: Exposure Information

Duration of application/ time ________________Hours
(Mixing, Loading and Spraying)

Frequency of spraying pesticide

________________times / day

________________days / week

________________weeks / month

________________months / year

Drinking water during farm activities

Source of drinking water
- ☐ Tab water
- ☐ Underground water
- ☐ Mixed (Both Tab water and Underground water)
- ☐ Other ______________________

Cooler tank / Bottle condition
- ☐ Open
- ☐ Close

Number of glasses (Drinking Water) _______________________glasses

How much? (Approximately) _______________Liter

<u>Part 3: Health effects of pesticide exposure</u>

Skin Symptoms

	Yes	*No*
Skin rash / itching / burning		
Tingling / numbness of hands		
muscular twitching and cramps		

Respiratory Symptoms

	Yes	*No*
Chest pain		
Cough		
Running nose		
Difficulties in breathing		
Shortness of breath		
Irritation of the throat		

Systemic Symptoms

	Yes	*No*
Excessive sweating		
Nausea		
Vomiting / Dizziness		
Excessive salivation		
Abdominal pain / Stomachache		
Headache		

Eye Symptoms

	Yes	*No*
Lacrimation		
Irritation		
Blurred Vision		

Neuro Muscular Symptoms

	Yes	*No*
Difficulty in seeing		
Restlessness		
Difficulty in failing asleep		
Trembling of hands		
Irritability		
Anxiety / anxiousness		
Memory Problem		

Appendix B
Questionnaire (Thai)

ผู้สัมภาษณ์____________________ Code.______

แบบสอบถามการรับสัมผัสสารกำจัดศัตรูพืช และผลกระทบทางสุขภาพ

คำชี้แจง

แบบสอบถามชุดนี้แบ่งออกเป็นทั้งหมด 3 ส่วน ในส่วนที่ 1 และ 2 ประกอบด้วยคำถามในลักษณะเปิดและปิด ในส่วนที่ 3 จะเป็นคำถามปิดเพียงอย่างเดียว

ส่วนที่ 1 ข้อมูลทั่วไป

คำถามในส่วนนี้จะถามเกี่ยวกับข้อมูลพื้นฐานของผู้ตอบแบบสอบถาม รวมไปถึงข้อมูลเกี่ยวกับลักษณะการทำงานในพื้นที่เกษตรกรรม, ลักษณะการใช้สารกำจัดศัตรูพืช และการใช้อุปกรณ์ป้องกันร่างกาย

ส่วนที่ 2 ข้อมูลการรับสัมผัสสารกำจัดศัตรูพืช

คำถามในส่วนที่สองจะเกี่ยวข้องกับความถี่ของการรับสัมผัสสารกำจัดศัตรูพืชผ่านทางผิวหนัง ทางการหายใจ และทางการรับประทาน เพื่อนำข้อมูลที่ได้ไปวิเคราะห์ และประเมินความเสี่ยงของเกษตรกร

ส่วนที่ 3 ผลกระทบทางสุขภาพจากการรับสัมผัสสารกำจัดศัตรูพืช

คำถามส่วนนี้ เกี่ยวกับกลุ่มอาการที่เกี่ยวข้องกับการรับสัมผัสสารกำจัดศัตรูพืช โดยกลุ่มอาการนั้นจะถูกถามเกษตรกรผู้รับสัมผัสสารกำจัดศัตรูพืชภายใน 24 ชั่วโมงหลังการรับสัมผัส

****

ส่วนที่ 1 ข้อมูลทั่วไป

เพศ ☐ ชาย ☐ หญิง

น้ำหนัก _______________กิโลกรัม

ความสูง _______________เซนติเมตร

ระดับการศึกษาสูงสุด

☐ ไม่ได้ศึกษา

☐ ประถมศึกษา

☐ มัธยมศึกษา

☐ ปวส. / ปวช.

☐ ปริญญาตรี หรือสูงกว่า

ท่านเคยสูบบุหรี่หรือไม่

☐ ไม่เคย

☐ เคย แต่เลิกสูบแล้ว

☐ สูบ

- o ชนิดของบุหรี่ ___________________
- o จำนวนมวน/ วัน ___________________
- o ความถี่ในการสูบ ___________________

รายได้จากการปลูกพริก / ปี _______________บาท

<u>*ลักษณะการทำงานในพื้นที่เกษตรกรรม*</u>

พื้นที่ในการเพาะปลูกพริก _______________ไร่

ระยะเวลาทั้งหมดที่ทำการเกษตร _______________ปี

กิจกรรมที่ทำในสวนขณะทำการปลูกพริก

☐ ผสมยา

☐ เทยาลงในเป้ฉีดพ่น

☐ ฉีดพ่นยา

ระยะเวลาที่ใช้สารกำจัดศัตรูพืชทั้งหมด _______________ปี _______________เดือน

จำนวนครั้งที่ฉีดพ่นสารกำจัดศัตรูพืชต่อปี ___________________ครั้ง

อุปกรณ์ที่ใช้ในการฉีดพ่นสารกำจัดศัตรูพืชมีรอยรั่วหรือไม่

☐ มี

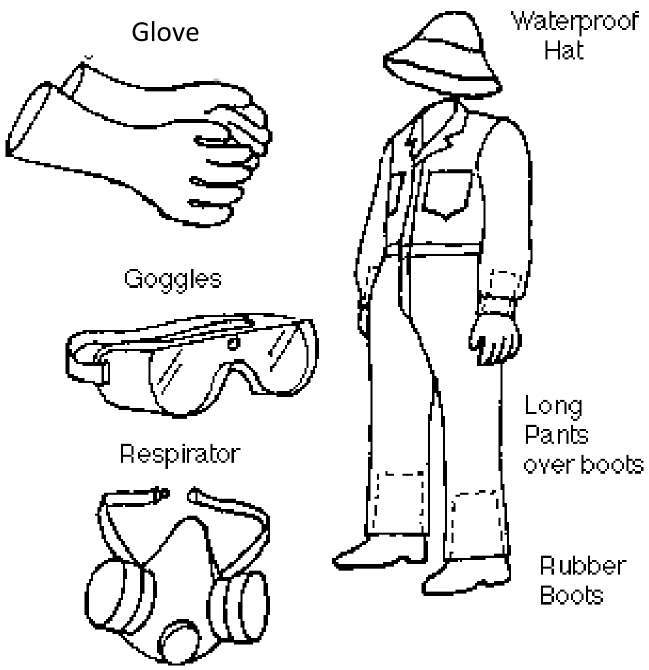
☐ ไม่มี

ลักษณะการใช้สารกำจัดศัตรูพืช

	ทุกครั้ง	บางครั้ง	ไม่เคย
ท่านใช้สารกำจัดศัตรูพืชตามที่ฉลากกำหนดหรือไม่			
ท่านเตรียมสารกำจัดศัตรูพืชที่บ้านหรือไม่			
ท่านเก็บสารกำจัดศัตรูพืชที่บ้านหรือไม่			
ท่านซักเสื้อผ้าที่ใช่ในการเกษตรรวมกับเสื้อผ้าทั่วไปหรือไม่			
ท่านล้างอุปกรณ์ฉีดพ่นยาหลังเสร็จการฉีดพ่นหรือไม่			
ท่านนำอาหารไปที่สวนพริกด้วยหรือไม่			
ท่านสูบบุหรี่ระหว่างฉีดพ่นหรือไม่			
ท่านได้คำนึงถึงความปลอดภัยของตัวท่านหรือไม่			

การใช้อุปกรณ์ป้องกันร่างกาย

	ทุกครั้ง	บางครั้ง	ไม่เคย
ท่านใช้ถุงมือทุกครั้งหรือไม่ ☐ ยาง ☐ ผ้า ☐ ยาว ☐ สั้น			
ท่านใช้ผ้าปิดจมูกทุกครั้งหรือไม่			
ท่านสวมรองเท้าบูททุกครั้งหรือไม่ ☐ ยาว ☐ สั้น			
ท่านสวมหมวกทุกครั้งหรือไม่			
ท่านสวมเสื้อแขนสั้นทุกครั้งหรือไม่			
ท่านสวมเสื้อแขนยาวทุกครั้งหรือไม่			
ท่านสวมกางเกงขาสั้นทุกครั้งหรือไม่			
ท่านสวมกางเกงขายาวทุกครั้งหรือไม่			



ส่วนที่ 2 ข้อมูลการรับสัมผัสสารกำจัดศัตรูพืช

ระยะเวลาทำกิจกรรมต่างๆในสวนพริกต่อครั้ง ________________ชั่วโมง

(ผสมยา, เทยาลงเป้ฉีดพ่น, ฉีดพ่น)

ความถี่ของการฉีดพ่นสารกำจัดศัตรูพืช

________________ครั้ง / วัน

________________วัน / สัปดาห์

________________สัปดาห์ / เดือน

________________เดือน / ปี

น้ำดื่มขณะทำกิจกรรมต่างๆในการปลูกพริก

แหล่งของน้ำดื่ม

- ☐ น้ำประปา
- ☐ น้ำใต้ดิน
- ☐ น้ำประปาและน้ำใต้ดิน
- ☐ อื่นๆ______________________

ลักษณะของภาชนะบรรจุน้ำดื่มในพื้นที่ปลูกพริก

- ☐ เปิด
- ☐ ปิด

ปริมาณน้ำที่ดื่มระหว่างทำงานในสวน ____________________แก้ว

รวมแล้วประมาณ______________ลิตร

ส่วนที่ 3 ผลกระทบทางสุขภาพจากการรับสัมผัสสารกำจัดศัตรูพืช

อาการทางผิวหนัง

	ใช่	ไม่ใช่
อาการคัน/ เป็นผื่น / มีรอยไหม้		
รู้สึกแปลบๆ หรือชาบริเวณมือ		
กล้ามเนื้อกระตุก หรือเป็นตะคริว		

อาการด้านระบบทางเดินหายใจ

	ใช่	ไม่ใช่
เจ็บหน้าอก		
ไอ		
น้ำมูกไหล		
หายใจไม่สะดวก		
หายใจเป็นจังหวะสั้นๆ		
ระคายคอ		

อาการด้านร่างกาย

	ใช่	ไม่ใช่
เหงื่อออกมากผิดปกติ		
วิงเวียนศีรษะ		
คลื่นไส้ / อาเจียน		
น้ำลายออกมาผิดปกติ		
ปวดท้อง		
ปวดหัว		

อาการด้านเกี่ยวกับตา

	ใช่	ไม่ใช่
น้ำตาไหล		
ระคายเคืองตา		
มองภาพไม่ชัดเจน		

อาการด้านระบบประสาท

	ใช่	ไม่ใช่
มองเห็น หรือสังเกตสิ่งต่างๆ ได้ยาก		
รู้สึกกระสับกระส่าย		
นอนหลับยาก		
มือสั่น		
ฉุนเฉียว หรือหงุดหงิดง่าย		
รู้สึกวิตกกังวล		
มีปัญหาด้านความจำ		

Appendix C

Body surface area

Table C-1 Equation Parameters for Calculating Adult Body Surface Area

Body Part	N	Equation for surface areas (m^2) a_0	W^{a1}	H^{a2}	P	R^2	S.E.
Head							
Female	57	0.0256	0.124	0.189	0.01	0.302	0.00678
Male	32	0.0492	0.339	-0.0950	0.01	0.222	0.0202
Trunk							
Female	57	0.188	0.647	-0.304	0.001	0.877	0.00567
Male	32	0.0240	0.808	-0.0131	0.001	0.894	0.0118
Upper Extremities							
Female	57	0.0288	0.341	0.175	0.001	0.526	0.00833
Male	48	0.00329	0.466	0.524	0.001	0.821	0.0101
Arms							
Female	13	0.00223	0.201	0.748	0.01	0.731	0.00996
Male	32	0.00111	0.616	0.561	0.001	0.892	0.0177
Upper Arms							
Male	6	8.70	0.741	-1.40	0.25	0.576	0.0387
Forearms							
Male	6	0.326	0.858	-0.895	0.05	0.897	0.0207
Hands							
Female	12[b]	0.0131	0.412	0.0274	0.1	0.447	0.0172
Male	32	0.0257	0.573	-0.218	0.001	0.575	0.0187
Lower Extremities[c]	105	0.00286	0.458	0.696	0.001	0.802	0.00633
Legs	45	0.00240	0.542	0.626	0.001	0.780	0.0130
Thighs	45	0.00352	0.629	0.379	0.001	0.739	0.0149
Lower legs	45	0.000276	0.416	0.973	0.001	0.727	0.0149
Feet	45	0.000618	0.372	0.725	0.001	0.651	0.0147

[a] $SA = a_0 W^{a1} H^{a2}$
W = Weight in kilograms; H = Height in centimeters; P = Level of significance; R^2 = Coefficient of determination;
SA = Surface Area; S.E. = Standard error; N = Number of observations
[b] One observation for a female whose body weight exceeded the 95 percentile was not used.
[c] Although two separate regressions were marginally indicated by the F test, pooling was done for consistency with individual components of lower extremities.
Source: U.S. EPA, 1985.

Adapted from: US EPA, 1997

Table C-2 Comparison of body surface area calculation and default values

Average surface area (m^2)	Sex					
	Male		Female		Male & Female	
	Calculation[a]	Default values[b]	Calculation[a]	Default values[b]	Calculation[c]	Default values[d]
Farmers						
Head	0.19	0.14	0.10	0.11	0.15	0.13
Face	0.06	-	0.03	-	0.05	-
Hands	0.19	0.11	0.12	0.09	0.16	0.10
Feet	0.08	0.14	0.08	0.12	0.08	0.13
Total	1.66	2.06	1.58	1.85	1.62	1.96
Non-farmers						
Head	0.19	0.14	0.10	0.11	0.15	0.13
Face	0.06	-	0.03	-	0.05	-
Hands	0.19	0.11	0.12	0.09	0.16	0.10
Feet	0.09	0.14	0.07	0.12	0.08	0.13
Total	1.84	2.06	1.57	1.85	1.71	1.96

[a] calculated body surface area values specific for participants in this study

[b] defaulted body surface area values (US EPA (1985) and NHANES 2005-2006 cited in US EPA, 2011: 7-43,7-44)

[c] average calculated body surface area values of male and female participants

[d] average defaults body surface area values

Appendix D

Average Daily Dose (ADD) Calculation of Chilli farmers

Hand contact

Chlorpyrifos

$$ADD_{mean} = \frac{0.043 \text{ mg/kg x } 10^{-6} \text{ kg/mg x } 456 \text{ x } 10^{-6} \text{ mg/cm}^2\text{-h x } 2 \text{ h/day x } 14.40 \text{ years x } 15.90 \text{ days/year x } 1.6 \text{ x } 10^3 \text{ cm}^2}{59.53 \text{ kg x } 14.40 \text{ year x } 365 \text{ days/year}}$$

$$= 4.59 \times 10^{-11} \text{ mg/kg-day}$$

$$ADD_{RME} = \frac{0.240 \text{ mg/kg x } 10^{-6} \text{ kg/mg x } 456 \text{ x } 10^{-6} \text{ mg/cm}^2\text{-h x } 2 \text{ h/day x } 14.40 \text{ years x } 15.90 \text{ days/year x } 1.6 \text{ x } 10^3 \text{ cm}^2}{59.53 \text{ kg x } 14.40 \text{ year x } 365 \text{ days/year}}$$

$$= 25.6 \times 10^{-11} \text{ mg/kg-day}$$

Profenofos

$$ADD_{mean} = \frac{0.087 \text{ mg/kg x } 10^{-6} \text{ kg/mg x } 456 \text{ x } 10^{-6} \text{ mg/cm}^2\text{-h x } 2 \text{ h/day x } 14.40 \text{ years x } 15.90 \text{ days/year x } 1.6 \text{ x } 10^3 \text{ cm}^2}{59.53 \text{ kg x } 14.40 \text{ year x } 365 \text{ days/year}}$$

$$= 9.29 \times 10^{-11} \text{ mg/kg-day}$$

$$ADD_{RME} = \frac{0.450 \text{ mg/kg x } 10^{-6} \text{ kg/mg x } 456 \text{ x } 10^{-6} \text{ mg/cm}^2\text{-h x } 2 \text{ h/day x } 14.40 \text{ years x } 15.90 \text{ days/year x } 1.6 \text{ x } 10^3 \text{ cm}^2}{59.53 \text{ kg x } 14.40 \text{ year x } 365 \text{ days/year}}$$

$$= 48.1 \times 10^{-11} \text{ mg/kg-day}$$

Face contact

Chlorpyrifos

$$ADD_{mean} = \frac{0.044 \text{ mg/kg x } 10^{-6} \text{ kg/mg x } 456 \text{ x } 10^{-6} \text{ mg/cm}^2\text{-h x 2 h/day x 14.40 years x 15.90 days/year x 0.50 x } 10^3 \text{ cm}^2}{59.53 \text{ kg x 14.40 year x 365 days/year}}$$

$$= 1.47 \times 10^{-11} \text{ mg/kg-day}$$

$$ADD_{RME} = \frac{0.210 \text{ mg/kg x } 10^{-6} \text{ kg/mg x } 456 \text{ x } 10^{-6} \text{ mg/cm}^2\text{-h x 2 h/day x 14.40 years x 15.90 days/year x 0.50 x } 10^3 \text{ cm}^2}{59.53 \text{ kg x 14.40 year x 365 days/year}}$$

$$= 7.01 \times 10^{-11} \text{ mg/kg-day}$$

Profenofos

$$ADD_{mean} = \frac{0.513 \text{ mg/kg x } 10^{-6} \text{ kg/mg x } 456 \text{ x } 10^{-6} \text{ mg/cm}^2\text{-h x 2 h/day x 14.40 years x 15.90 days/year x 0.50 x } 10^3 \text{ cm}^2}{59.53 \text{ kg x 14.40 year x 365 days/year}}$$

$$= 17.1 \times 10^{-11} \text{ mg/kg-day}$$

$$ADD_{RME} = \frac{6.400 \text{ mg/kg x } 10^{-6} \text{ kg/mg x } 456 \text{ x } 10^{-6} \text{ mg/cm}^2\text{-h x 2 h/day x 14.40 years x 15.90 days/year x 0.50 x } 10^3 \text{ cm}^2}{59.53 \text{ kg x 14.40 year x 365 days/year}}$$

$$= 214 \times 10^{-11} \text{ mg/kg-day}$$

Body contact

Chlorpyrifos

$$ADD_{mean} = \frac{1.090 \text{ mg/kg x } 10^{-6} \text{ kg/mg x } 0.1 \text{ x } 456 \text{ x } 10^{-6} \text{ mg/cm}^2\text{-h x } 2 \text{ h/day x } 14.40 \text{ years x } 15.90 \text{ days/year x } 16.2 \text{ x } 10^3 \text{ cm}^2}{59.53 \text{ kg x } 14.40 \text{ year x } 365 \text{ days/year}}$$

$$= 1.18 \times 10^{-9} \text{ mg/kg-day}$$

$$ADD_{RME} = \frac{7.975 \text{ mg/kg x } 10^{-6} \text{ kg/mg x } 0.1 \text{ x } 456 \text{ x } 10^{-6} \text{ mg/cm}^2\text{-h x } 2 \text{ h/day x } 14.40 \text{ years x } 15.90 \text{ days/year x } 16.2 \text{ x } 10^3 \text{ cm}^2}{59.53 \text{ kg x } 14.40 \text{ year x } 365 \text{ days/year}}$$

$$= 8.62 \times 10^{-9} \text{ mg/kg-day}$$

Profenofos

$$ADD_{mean} = \frac{1.076 \text{ mg/kg x } 10^{-6} \text{ kg/mg x } 0.1 \text{ x } 456 \text{ x } 10^{-6} \text{ mg/cm}^2\text{-h x } 2 \text{ h/day x } 14.40 \text{ years x } 15.90 \text{ days/year x } 16.2 \text{ x } 10^3 \text{ cm}^2}{59.53 \text{ kg x } 14.40 \text{ year x } 365 \text{ days/year}}$$

$$= 1.16 \times 10^{-9} \text{ mg/kg-day}$$

$$ADD_{RME} = \frac{11.40 \text{ mg/kg x } 10^{-6} \text{ kg/mg x } 0.1 \text{ x } 456 \text{ x } 10^{-6} \text{ mg/cm}^2\text{-h x } 2 \text{ h/day x } 14.40 \text{ years x } 15.90 \text{ days/year x } 16.2 \text{ x } 10^3 \text{ cm}^2}{59.53 \text{ kg x } 14.40 \text{ year x } 365 \text{ days/year}}$$

$$= 12.3 \times 10^{-9} \text{ mg/kg-day}$$

Inhalation

Chlorpyrifos

$$ADD_{mean} = \frac{0.001\ mg/m^3 \times 7.8\ m^3/day \times 14.40\ years \times 15.90\ days/year}{59.53\ kg \times 14.40\ year \times 365\ days/year}$$

$$= 5.71 \times 10^{-6}\ mg/kg\text{-}day$$

$$ADD_{RME} = \frac{0.004\ mg/m^3 \times 7.8\ m^3/day \times 14.40\ years \times 15.90\ days/year}{59.53\ kg \times 14.40\ year \times 365\ days/year}$$

$$= 22.8 \times 10^{-6}\ mg/kg\text{-}day$$

Profenofos

$$ADD_{mean} = \frac{0.0003\ mg/m^3 \times 7.8\ m^3/day \times 14.40\ years \times 15.90\ days/year}{59.53\ kg \times 14.40\ year \times 365\ days/year}$$

$$= 1.71 \times 10^{-6}\ mg/kg\text{-}day$$

$$ADD_{RME} = \frac{0.007\ mg/m^3 \times 7.8\ m^3/day \times 14.40\ years \times 15.90\ days/year}{59.53\ kg \times 14.40\ year \times 365\ days/year}$$

$$= 40.0 \times 10^{-6}\ mg/kg\text{-}day$$

Appendix E

Urinary metabolite results of chilli farmers

Table E-1 Urinary Diethylphosphate (**DEP**) metabolite results (n=39)

	No. of detected samples			**Concentration**	
	n	**%**		Unadjusted Cratinine (µg/L)	Adjusted Creatinine (µg/g cre.)
Pre-MLA	9	23.1	Geometric Mean	0.33	0.29
			Range	<LOD – 7.06	<LOD – 5.41
			Median	0.20	0.21
			Percentile		
			25^{th}	0.20	0.13
			50^{th}	0.20	0.24
			75^{th}	0.20	0.36
			95^{th}	2.99	3.75
1^{st} Post-MLA	23	59.0	Geometric Mean	1.04	0.92
			Range	<LOD – 9.97	<LOD – 14.8
			Median	1.38	0.60
			Percentile		
			25^{th}	0.20	0.25
			50^{th}	1.38	0.60
			75^{th}	6.02	4.50
			95^{th}	9.79	12.1
2^{nd} Post-MLA	14	35.9	Geometric Mean	0.59	0.52
			Range	<LOD – 20.0	<LOD – 30.5
			Median	0.20	0.29
			Percentile		
			25^{th}	0.20	0.15
			50^{th}	0.20	0.29
			75^{th}	2.73	2.06
			95^{th}	19.2	16.4

LOD < 0.2

Table E-2 Urinary Diethylthiophosphate (**DTEP**) metabolite results (n=39)

	No. of detected samples			**Concentration**	
	n	**%**		Unadjusted Cratinine (µg/L)	Adjusted Creatinine (µg/g cre.)
Pre-MLA	15	38.5	Geometric Mean	0.46	0.40
			Range	<LOD – 134	0.08 – 143
			Median	0.20	0.29
			Percentile		
			25th	0.20	0.14
			50th	0.20	0.29
			75th	0.98	1.03
			95th	6.65	3.54
1st Post-MLA	30	76.9	Geometric Mean	4.39	3.88
			Range	<LOD – 124.8	0.08 – 186
			Median	5.15	3.38
			Percentile		
			25th	1.68	0.82
			50th	5.15	3.38
			75th	14.2	10.3
			95th	125	151
2nd Post-MLA	16	41.0	Geometric Mean	0.72	0.64
			Range	<LOD – 15.7	0.08 -17.6
			Median	0.20	0.30
			Percentile		
			25th	0.20	0.16
			50th	0.20	0.29
			75th	3.35	2.89
			95th	13.3	11.9

LOD < 0.2

Table E-3 Urinary Diethyldithiophosphate (**DEDTP**) metabolite results (n=39)

	No. of detected samples			Concentration	
	n	**%**		Unadjusted Cratinine (µg/L)	Adjusted Creatinine (µg/g cre.)
Pre-MLA	3	7.69	Geometric Mean	0.15	0.13
			Range	<LOD - 83.2	0.03 – 83.2
			Median	0.10	0.11
			Percentile		
			25th	0.10	0.06
			50th	0.10	0.11
			75th	0.10	0.14
			95th	6.02	3.86
1st Post-MLA	9	23.1	Geometric Mean	0.28	0.25
			Range	<LOD – 135.1	0.03 – 63.7
			Median	0.10	0.12
			Percentile		
			25th	0.10	0.06
			50th	0.10	0.12
			75th	2.28	0.22
			95th	116	57.0
2nd Post-MLA	4	10.3	Geometric Mean	0.14	0.12
			Range	<LOD – 6.02	0.03 – 5.37
			Median	0.10	0.10
			Percentile		
			25th	0.10	0.06
			50th	0.10	0.10
			75th	0.10	0.15
			95th	3.39	3.86

LOD < 0.1

Appendix F

Urinary metabolite results of non-chilli farmers

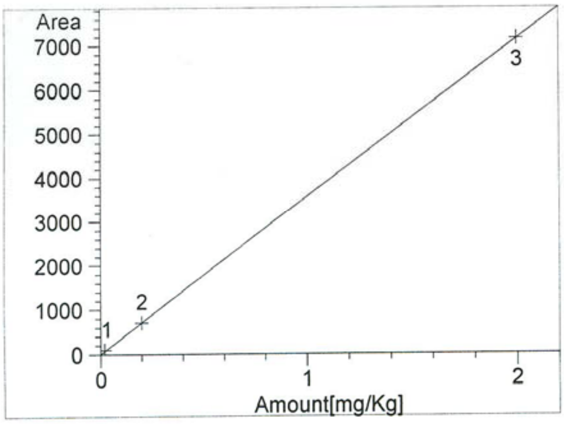
Table F-1 Urinary Diethyl DAPs metabolite results (n=40)

	No. of detected samples			Concentration	
	n	%		Unadjusted Cratinine (µg/L)	Adjusted Creatinine (µg/g cre.)
DEP	10	25.0	Geometric Mean	0.27	0.32
			Range	<LOD – 5.45	<LOD – 4.78
			Median	0.20	0.25
			Percentile		
			25th	0.20	0.17
			50th	0.20	0.25
			75th	0.20	0.53
			95th	1.67	2.86
DETP	13	32.5	Geometric Mean	0.30	0.36
			Range	<LOD – 4.31	<LOD – 6.06
			Median	0.20	0.25
			Percentile		
			25th	0.20	0.17
			50th	0.20	0.25
			75th	1.00	0.75
			95th	3.35	4.46
DEDTP	1	2.50	Geometric Mean	0.1	0.12
			Range	<LOD – 0.10	<LOD – 0.42
			Median	0.10	0.10
			Percentile		
			25th	0.10	0.08
			50th	0.10	0.10
			75th	0.10	0.15
			95th	0.10	0.38

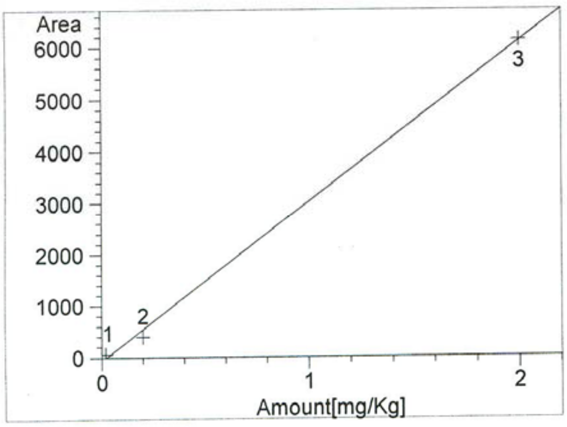
Appendix G

Calibration Curve and Laboratory Analysis

Chlorpyrifos and Profenofos calibration curve

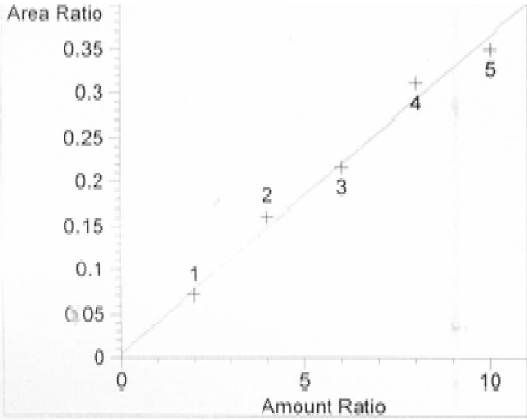



```
Chlopyrifos at exp. RT: 17.274
FPD1 A,
Correlation:              0.99999
Residual Std. Dev.:     14.32502
Formula: y = mx + b
       m:    3571.27049
       b:       6.36649
       x: Amount[mg/Kg]
       y: Area
```

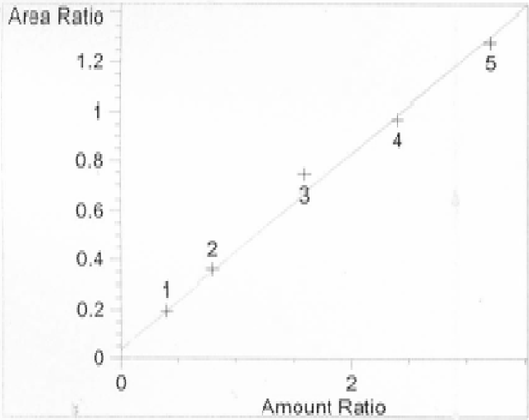



```
Profenofos at exp. RT: 23.466
FPD1 A,
Correlation:              0.99939
Residual Std. Dev.:    128.30549
Formula: y = mx + b
       m:    3091.16837
       b:     -68.90286
       x: Amount[mg/Kg]
       y: Area
```

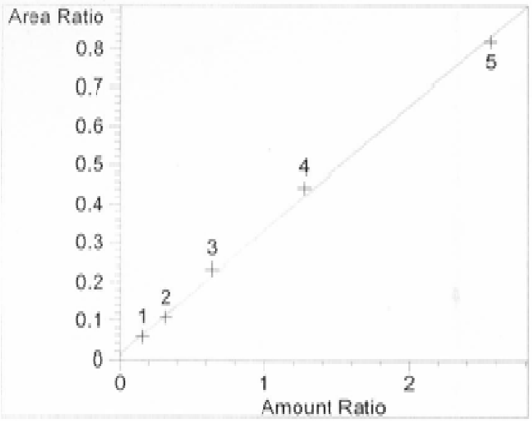
Urinary metabolite calibration curve



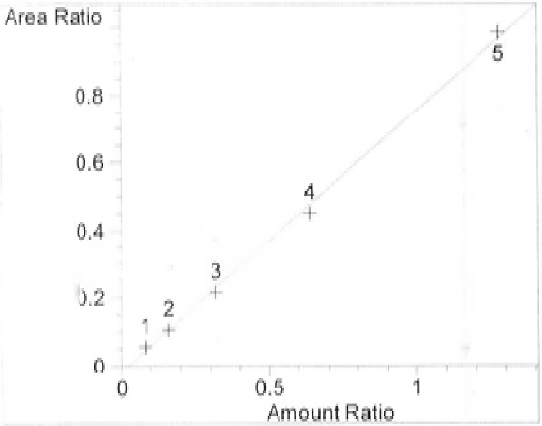
DMP at exp. RT: 3.840
FPD1 B,
Correlation: 0.99580
Residual Std. Dev.: 0.01392
Formula: y = mx + b
m: 3.61839e-2
b: 4.44552e-3
x: Amount Ratio
y: Area Ratio



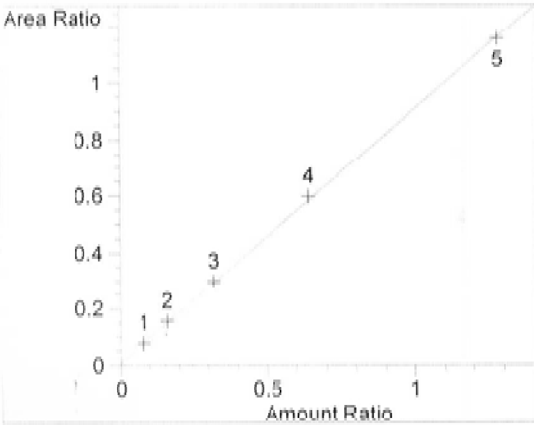
DEP at exp. RT: 4.600
FPD1 B,
Correlation: 0.99673
Residual Std. Dev.: 0.04426
Formula: y = mx + b
m: 3.95863e-1
b: 3.72111e-2
x: Amount Ratio
y: Area Ratio



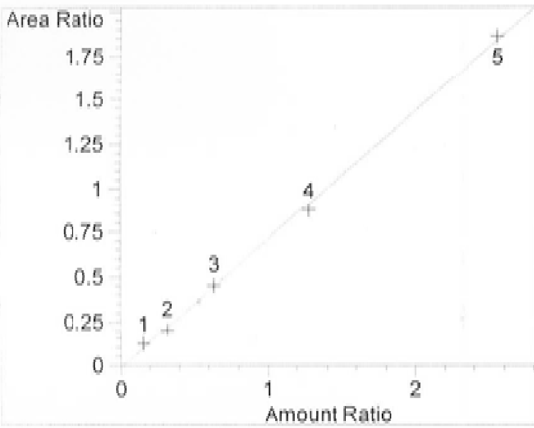
DMTP at exp. RT: 5.280
FPD1 B,
Correlation: 0.99881
Residual Std. Dev.: 0.01684
Formula: y = mx + b
m: 3.20705e-1
b: 1.19945e-2
x: Amount Ratio
y: Area Ratio



DMDTP at exp. RT: 6.380
FPD1 B,
Correlation: 0.99895
Residual Std. Dev.: 0.01893
Formula: y = mx + b
m: 7.68148e-1
b: -1.53890e-2
x: Amount Ratio
y: Area Ratio



DETP at exp. RT: 6.460
FPD1 B,
Correlation: 0.99986
Residual Std. Dev.: 0.00824
Formula: y = mx + b
m: 9.00562e-1
b: 7.48935e-3
x: Amount Ratio
y: Area Ratio



DEDTP at exp. RT: 7.260
FPD1 B,
Correlation: 0.99942
Residual Std. Dev.: 0.02664
Formula: y = mx + b
m: 7.24304e-1
b: -1.31402e-2
x: Amount Ratio
y: Area Ratio

Appendix H

Risk communication material

Monday	Tuesday	Wednesday	Thursday	Friday	Saturday	Sunday
			1	2	3	4
5	6	7	8	9	10	11
12	13	14	15	16	17	18
19	20	21	22	23	24	25
26	27	28	29	30	31	

VITAE

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Place of Birth : Bangkok, Thailand

Educational Achievement

- Bachelor of Science (Environmental Science), Faculty of Science, Chulalongkorn University, Bangkok, Thailand
- Master of Public Health, College of public health science, Chulalongkorn University, Bangkok, Thailand

Research Experience

- The association between Carbon Monoxide in underground car park and health effect
- Risk assessment of chlorpyrifos (Organophosphate pesticide) associated with dermal exposure in chilli-growing farmers at Ubonratchathani province, Thailand