Comparison of Biochemical, Hematological Parameters and Pesticide Expose-related Symptoms among Organic and Non-organic Farmers, Singburi, Thailand

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Abstract

Background: Main of agriculturists in Singburi was cultivated rice and major pesticides’ use is chlorpyrifos; however, farmers are applied bio-extracts to eliminate pest and trend to be organic farming. Hence, organic farmers may become lower risks from pesticides rather than non-organic farmers. Aim: To determine biochemical and hematological status between organic and non-organic farmers, Bang Rachan district, Singburi, Thailand. Materials and Methods: Farmers were separated into organic farmer group (N = 35) and non-organic farmer group (N = 45). Individual data for pesticide exposure were included, and pesticide-exposed relating symptoms were recorded. All demographic data of participants were documented before blood collection. Serum samples were analyzed for butyrylcholinesterase (BuChE) activity, liver function test, kidney function test, and lipid profiles. Each ethylenediaminetetraacetic acid-whole blood was determined for complete blood count (CBC). NaF plasma was analyzed for determine fasting blood glucose. All biochemical parameters were principled according to enzymatic assays and CBC was automatic analyzers. The data were represented in average ± standard deviation, and difference of biochemical and hematological status of two groups was statistically tested by unpaired t-test (P < 0.05). Results and Discussion: Blood glucose and lipid profiles of non-organic farmers were higher than reference ranges and significantly difference with other groups. Mean corpuscular hemoglobin (MCH) and MCH concentration values of non-organic farmers were lower than reference range and interpreted as anemia. Conclusions: Non-organic farmers were suchronic pesticide exposure with normal BuChE values and no related symptom; therefore, subclinical symptoms were disturbed biochemical and hematological status; and prediabetes, dyslipidemia, and anemia appeared.

Key words: Biochemical test, cholinesterase, complete blood count, pesticide exposure, organic farming

INTRODUCTION

Pesticides are broadly utilized for pest and pest-induced disease controls, especially in crop cultivation and vector-borne diseases control for public health work. World pesticides, including herbicides, insecticides, fungicides, and rodenticides, were annually applied approximately 2.4 million tons during 2006 and 2007.¹ Developing countries were used pesticide about 20% of the world, and Thailand was the third rank of pesticide usages in Asia-Pacific region.²⁻⁵ The health effects of intensive pesticide utilized and frequent pesticide exposures were caused acute and chronic intoxications, which were public health problems in Thailand. There was an increasing trend of pesticides imported from about 110,000 tons in 2007 to approximately 172,000 tons in 2013. Herbicides were the major pesticides with the highest proportion of import (62-79%), followed by insecticides (12-23%) and fungicides (5-11%). There were about 49,000 to 61,000 reported cases of pesticide intoxication each year with morbidity rate between

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76.4 and 96.6 per 100,000 populations. The reported cases of the toxic effects of substances during 2007-2013 were found predominantly in the Central region of Thailand (31-36%), followed by the North Eastern region (27-31%), whereas the annual proportion of the North (18-20%) were almost equal to those of the South (18-19%). The number of cases were usually increased during the growing season of many crops in rainy season (May - August) each year, and it was found mainly in farmers and farm workers.[4,6]

Pesticides exposures of agricultural workers were major occurred while were contact on mixing, loading, and spraying pesticides in farming and orchard area, and the rate of exposure may be several times of proportion in agriculturalist higher than in general people.12,5,7 Intensively, pesticide usage was lead high accumulation of toxic residues in environment (such as in soil, sediment, and water resources) and may spread to ecosystem (including aquatic animals, livestock, and agricultural products). The top of the food chain is humans; thus, pesticides are also contained in human body and hence made harmful health outcome. Acute and chronic clinical symptoms were characterized by pesticide exposures through water and food consumption.18-10 In literature reviews were mainly reported chronic effects of pesticides, which was focused on carcinogenesis in pesticide-exposed workers.11-13 However, other health effects were also related to pesticide exposure including effects in various human systems such as the immune, nervous, endocrine, and reproductive systems.14-20 According to epidemiologic data, pesticides were affected on enzymes, which were responsible for liver function, blood cell characteristics, and other biochemical pathways in persons who were pesticide exposed and had occupationally diseases.4,20-23 The current pesticide poisoning in Thailand was reported, and the chronic toxic effects of pesticide exposure were considered by worker, community, and government levels, which were pay attention to reduce the intensive pesticide usage. One way to reduce pesticide usage was the urgently promoted the organic farming practices, and search for the effective biopesticides or biological agents is used to control agricultural pests to substitute the chemical pesticides.4,6

Singburi province was a great city in Thai history for the heroic act of the villagers of Bang Rachan in battle, which located on the West bank of the Chao Phraya River, about 142 km from Bangkok, and it is an area of around 841 km². Geographically, it is a basin where three rivers: Chao Phraya, Noi, and Lopburi, flow through, are well known for abundant river fish, particularly the “Mae La” fish. More than 90% of agriculturists in Singburi were cultivated rice, and major pesticides’ use is chlorpyrifos, the member of organophosphates; however, Singburi farmers are applied to use bio-extracts for eliminate pest and trend to be organic farming by rapidly changing farming practices such as, relies on fertilizers of organic origin, i.e., compost, manure, green manure, and bone meal; crop rotation and companion planting; and biological pest control. Hence, organic farmers may become lower risks of health effects from pesticides rather than non-organic farmers. We were interested to the determined biochemical and hematological parameters and pesticide exposure-related symptoms between organic and non-organic farmers who habitat at Bang Rachan district, the most rice cultivate area of Singburi. This research results may be used as a guideline to health awareness of occupation workers or agriculturalists for changing from “risk” to more “safe” practices to prevent toxic effects from pesticide exposure. In case of non-organic farmers who were occurred subchronic effects, there may change the agricultural practices by public relations. These data were attractive for healthy consumers, which consumed safe rice products and may lead to increase organic rice demand in healthy food markets. Safety agriculture practices of farmers were benefited in organic farming due to health safety, cost reduction, and value added of agricultural products (as organic or high quality products).

MATERIALS AND METHODS

Subjects

Anthropometric data, characteristics of pesticide usage, and related symptoms for pesticide exposure were documented. Blood samples were collected from agriculturalists at Bang Rachan district, Singburi province that came for health service by Saun Sumandha Rajabhat University during November to December 2016. Farmers were separated into organic farmer group (N = 35), which were no past history of pesticide use in farming during the past 3 years ago, and non-organic farmer group (N = 45) was farmers who used pesticides frequently. Both of male and female farmers had same agricultural activities. Individual data for pesticide exposure were included the frequency of protective tool used and rate of pesticide exposure. The pesticide-exposed relating symptoms were recorded by interviewing. The participants with poor literacy had helped for filling the form of questionnaire. Informed consent of all participants was done, and study protocol was approved by the Ethical Review Committee. The demographic data, smoking habits, farming duration, and environmental habitat were significantly similar among two groups. After history documented, height and weight of each participant were measured using a fix stadia rod and an electronic scale, respectively, which were calculated the body mass index (BMI). All information of participants was documented before blood collection.

Blood collection, specimen preparation, and laboratory assay

Each blood sample was obtained by venipuncture from median cubital vein. 10 ml of fasting blood sample was collected and then divided into 6 ml of blood sample was drawn into clotting blood tube for prepare serum and the
remaining was drawn into ethylenediaminetetraacetic acid (EDTA) and NaF tubes (2 ml for each) for prepare EDTA-whole blood and NaF plasma. Each serum sample was analyzed for butyrylcholinesterase (BuChE) activity, liver function test (including total protein, albumin, aspartate aminotransferase [AST], alanine aminotransferase [ALT], alkaline phosphatase [ALP], total bilirubin, total bilirubin, and direct bilirubin), kidney function test (creatinine and blood urea nitrogen [BUN]), and lipid profiles (triglyceride, cholesterol, high density lipoprotein-cholesterol [HDL-c], and low density lipoprotein-cholesterol [LDL-c]). Each EDTA-whole blood was determined for complete blood count (CBC) including red blood cell indices, white blood cell count and differential of white blood cell, and platelet count; then hematological status was evaluated. Each NaF plasma was analyzed for determine fasting blood glucose. All biochemical parameters were principled according to enzymatic assays and operated by automatic analyzer, COBAS c501 (Roche-diagnostics, Rotkreuz, Switzerland) and CBC was analyzed by Celltac E MEK-7222 (Nihon Kohden, Tomioka, Japan). The laboratory assay was repeated in the triplicate analysis of three blood sample types. Quality controls were done by calculation of coefficient of variation, which ≤10% and performed in certified clinical laboratories. The values of biochemical and hematological data were interpreted by reference values according to Clinical and Laboratory Standards Institute.

Statistical analysis

The anthropometric, biochemical, and hematological data were represented in average ± standard deviation. Statistical analysis was performed using the SPSS computer program version 14.0 (SPSS, Chicago, IL, USA). The normal distribution and difference biochemical and hematological parameters between organic and non-organic farmers were statistically tested by Kolmogorov–Smirnov test and unpaired t-test, respectively. The statistical significance for each analysis was considered at $P < 0.05$.

Table 1: Anthropometric characteristics and history for pesticide use in organic and non-organic farmers

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Organic farmers (n=35)</th>
<th>Non-organic farmers (n=45)</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age</td>
<td>61.5±10.8</td>
<td>62.1±12.3</td>
<td>&gt;0.05</td>
</tr>
<tr>
<td>BMI</td>
<td>23.6±1.5</td>
<td>24.5±2.6</td>
<td>&gt;0.05</td>
</tr>
<tr>
<td>Working duration for sprayers n (%)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1-5 years</td>
<td>11 (31.4)</td>
<td>13 (28.9)</td>
<td></td>
</tr>
<tr>
<td>5-10 years</td>
<td>2 (5.7)</td>
<td>22 (48.9)</td>
<td></td>
</tr>
<tr>
<td>&gt;10 years</td>
<td>10 (22.2)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Frequently of protective equipment use n (%)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Never</td>
<td></td>
<td>4 (8.9)</td>
<td></td>
</tr>
<tr>
<td>Rarely</td>
<td>3 (8.6)</td>
<td>6 (13.3)</td>
<td></td>
</tr>
<tr>
<td>Very often</td>
<td>9 (25.7)</td>
<td>25 (55.6)</td>
<td></td>
</tr>
<tr>
<td>Always</td>
<td>1 (2.8)</td>
<td>5 (11.1)</td>
<td></td>
</tr>
</tbody>
</table>

BMI: Body mass index

RESULTS

The 80 farmers were joined in this study, which divided into 35 and 45 for organic and non-organic farmers, respectively. Anthropometric data and working conditions of the participants were shown in Table 1. The average of BMI of both groups was risked to overweight (BMI = 23-24.9) and no statistical different between age range and BMI value in both groups ($P > 0.05$). Main of non-organic farmers (more than 50%) had worked with pesticide exposure more than 5 years. However, main of non-organic farmers (more than 70%) was often utilized protective equipment and some of organic farmers were agriculture with pesticide usage in the part. BuChE activity values of both groups were within the reference range (4.65-10.44 U/mL) and no significantly different [Table 2]. Other biochemical parameters of organic and non-organic farmers were evaluated and had interpreted with reference values [Table 2]. The average of each biochemical values including ALP, bilirubin, total protein, albumin, BUN, and creatinine in both groups were also no statistical different and values were within reference range. AST and ALT values in non-organic farmers were slightly higher than reference values; however, there was no statistical difference when compared to liver enzymes in organic farmers. The level of fasting blood glucose and lipid profiles including cholesterol, triglyceride, HDL-c, and LDL-c between both groups was significantly difference at $P = 0.047, 0.035, 0.026, 0.023$, and $0.039$, respectively. Non-organic farmer group was risked to pre-diabetes and dyslipidemia and trended to higher risk of cardiovascular diseases rather than organic farmers by higher in levels of triglyceride, cholesterol, and LDL-c and lower in level of HDL-c [Table 3]. Hematological parameters were determined in organic farmers and in non-organic farmers by CBC, which were shown in Table 4. The mean corpuscular hemoglobin (Hb) (MCH) and MCH concentration (MCHC) of non-organic farmers were lower (22.8 ± 4.2 pg and 29.1 ± 1.5 g/dL, respectively) than normal (MCH = 23-33 pg and MCHC = 31-37 g/dL); however,
no significantly different between two groups for all hematological parameters. Significant symptoms of pesticide exposure such as burning sensation in eyes and/or skin, chest lightness, dizziness, headache, and weakness rarely occurred in both groups.

**DISCUSSION**

BuChE activity is a reliable biomarker of pesticide exposure and generally used to estimate pesticide toxicity in occupational and clinical toxicology.\(^2\)\(^2\)\(^3\)\(^2\)\(^8\) and experts have recommended BuChE activity lower 60% of the reference value as critical value.\(^5\) In our results, BuChE activities in both groups were within the normal range and no statistical different. Normal BuChE levels in non-organic farmers may due to good practice for pesticide usage by often used protective equipment; however, normal BuChE levels in organic farmers were due to low pesticide exposed. Liver function test in both groups was no statistical different and almost within reference range;\(^2\)\(^5\) however, in non-organic farmers were slightly higher AST and ALT, which may unclear for the reasons between pesticide-exposed and/or may some medication of chronic diseases in elderly farmers, and this was the limitation of our study.\(^2\)\(^9\) Lack of individual characteristics rather than age and BMI may also a limitation of this study. Fasting blood glucose and lipid profiles in non-organic farmers were trended to a higher risk of cardiovascular diseases and significant different, which should be focus. The previous studies had reported the relation between organophosphate exposure and alteration of biochemical metabolism in animal model and epidemiological studies. Subchronic exposure of chlorpyrifos, the one member of OPs in farmer use, caused hyperlipidemia, glycogen storage reduction in liver, and increased oxidative stress, which was the risk of many chronic diseases such as cardiovascular diseases, hypertension, and diabetes mellitus (DM).\(^1\)\(^9\),\(^2\)\(^0\)-\(^3\)\(^2\)\(^3\)\(^2\)\(^3\)\(^2\) DM, particularly Type 2, is considered a multifactorial disease, in which genetics and lifestyle play a significant role, as well as environmental and occupational factors.\(^2\)\(^0\),\(^2\)\(^6\) Indeed, the previous study was demonstrated a strong correlation between the blood concentration of malathion, an organophosphate insecticide, and insulin resistance among farmers.\(^2\)\(^0\),\(^3\)\(^2\)\(^3\) Corresponded previous studies have demonstrated increased levels of pro-inflammatory cytokines in rats exposed to organophosphates.\(^2\)\(^0\),\(^2\)\(^2\)\(^3\)\(^4\),\(^3\)\(^5\) In addition, it has been demonstrated that the peripheral administration of interleukin-6 induces hyperglycemia and insulin resistance in humans and rodents.\(^3\)\(^6\) A recent study hypothesized that organophosphate pesticides can attenuate the incretin effect and produce insulin resistance through lipotoxic effects, inflammatory stimulation, and the induction of oxidative stress.\(^2\)\(^0\),\(^3\)\(^2\) The effects on hematological status by subchronic exposure were obtained not much attention. In our study, the mean of MCH and MCHC in non-organic farmers was lower than reference values, which

### Table 2: BuChE activity and liver and kidney function tests in organic and non-organic farmers

<table>
<thead>
<tr>
<th>Sample group</th>
<th>BuChE (U/mL)</th>
<th>Total protein (g/dL)</th>
<th>Albumin (g/dL)</th>
<th>Total bilirubin (mg/dL)</th>
<th>Direct bilirubin (mg/dL)</th>
<th>AST (U/L)</th>
<th>ALT (U/L)</th>
<th>ALP (U/L)</th>
<th>BUN (mg/dL)</th>
<th>Creatinine (mg/dL)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Organic farmers (n=35)</td>
<td>9.17±1.13</td>
<td>7.41±0.25</td>
<td>4.4±0.28</td>
<td>0.38±0.03</td>
<td>0.06±0.00</td>
<td>35.4±5.9</td>
<td>38.5±6.3</td>
<td>72.2±14.3</td>
<td>10.8±4.6</td>
<td>0.71±0.08</td>
</tr>
<tr>
<td>Non-organic farmers (n=45)</td>
<td>8.41±1.54</td>
<td>7.23±0.35</td>
<td>4.35±0.41</td>
<td>0.42±0.05</td>
<td>0.11±0.01</td>
<td>42.5±6.7</td>
<td>77.4±18.6</td>
<td>10.6±4.5</td>
<td>5.0±2.3</td>
<td>0.85±0.12</td>
</tr>
<tr>
<td>Reference range</td>
<td>4.65-10.44</td>
<td>6.60-8.70</td>
<td>3.50-5.50</td>
<td>0.30-1.20</td>
<td>0.00-0.50</td>
<td>37.0</td>
<td>53.0</td>
<td>128.0</td>
<td>5.0-23.0</td>
<td>0.5-1.5</td>
</tr>
<tr>
<td>P value</td>
<td>0.649</td>
<td>0.365</td>
<td>0.267</td>
<td>0.634</td>
<td>0.121</td>
<td>0.267</td>
<td>0.334</td>
<td>0.267</td>
<td>0.866</td>
<td>0.152</td>
</tr>
</tbody>
</table>

AST: Aspartate aminotransferase, ALT: Alanine aminotransferase, ALP: Alkaline phosphatase
corresponded to the previous report. The lowering of MCH and MCHC values and normocytic anemia caused by pesticide exposure, particularly organophosphates were still unclear; however, there may cause by general anemia in elderly caused by chronic blood loss, malnutrition, etc., and the subclinical symptoms caused by chronic pesticide exposure (particularly organophosphates) in long period may induce anemia in animal models. Furthermore, anemia due to pesticide exposure had been reported in Indian pesticide sprayers, which were significantly low in levels of Hb, Hct, MCV, MCH and MCHC, and impaired liver and kidney functions also occurred.

**CONCLUSION**

Age and BMI of organic and non-organic farmers in Bang Rachan district, Singburi, Thailand, were similar and no significant symptoms related to pesticide exposure which corresponded to normal levels of cholinesterase; however, AST and ALT values in non-organic farmers were slightly higher than reference values. Blood glucose and lipid profiles between non-organic farmers were higher than reference values, which were risked to pre-diabetes and dyslipidemia. MCH and MCHC values of non-organic farmers were lower than reference range and interpreted as normocytic anemia. In conclusion, non-organic farmers were subchronic pesticide exposure with normal BuChE values and no related symptom; therefore, subclinical symptoms were disturbed biochemical and hematological status, and pre-diabetes, dyslipidemia, and anemia appeared.

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**REFERENCES**

4. Sudjaroen Y. Biochemical and hematological status of...


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