Monitoring of pesticide residues in domestic vegetables in Thailand during 2015

Jitpaka Suntudrob¹, Wischada Jongmevasna¹, Thongsuk Payanan¹, RattiyaKorn Srikote¹, Weerawut Wittayanan¹*.

¹Bureau of Quality and Safety of Food, Department of Medical Sciences, Ministry of Public Health, Nonthaburi, Thailand
*Correspondent author: weerawut.w@dmsc.mail.go.th

Abstract

In Thailand, the Ministry of Public Health has the responsibility for regulatory monitoring safety of food. To assess the residue levels and to evaluate the exposure of the Thai population to pesticides, a nationwide surveillance program for pesticide residues in vegetables has been performed during 2015. A total of 934 samples of four types of vegetables including Chinese broccoli, ivy gourd, yardlong bean and water spinach were collected from wholesale markets by 77 Provincials Public Health Offices in collaboration with 14 Regional Medical Sciences Centers. All samples were sent to the accredited laboratory of the Bureau of Quality and Safety of Food and were examined for residues of 60 pesticides, including 20 organochlorines, 23 organophosphorus, 8 synthetic pyrethroids, 7 carbamates and 2 fungicides using multi-residue extraction and chromatography method. Predominantly, 22.3% of the samples had detectable pesticide residues at levels ranging from <0.01 to 5.9 mg/kg. Of the contaminated samples, the levels of 33.9, 31.9, 10.6 and 9.5% of the samples were pesticides detectable in yardlong bean, Chinese broccoli, ivy gourd and water spinach, respectively. The results showed a potential human dietary risk related to consumption of these vegetables. Governmental action plans should be developed with the cooperation of producers to reduce pesticide residues.

Keywords: Food safety, Monitoring program, Pesticide residues, Thailand, Vegetables

1. Introduction

To guarantee the sufficiency food supply, the use of pesticides in agriculture is still required [1]. However, the consumer’s health risk is concerned due to the presence of residues in treated food [2]. Thailand is one of the largest vegetable producers in the world and is an important exporter of tropical and subtropical fresh and processed vegetables, mainly to ASEAN countries, Japan, the United States of America and European countries [3 & 4]. Although agricultural products which were rejected from EU markets were mainly from China, Turkey and India, some of fresh vegetables from Thailand also had pesticide residues and started scarce worldwide [5]. Recently, the results from the monitoring and surveillance program called Total Diet Study (TDS) on pesticide residues conducted in Thailand during 1999-2003 [6] shown that 24 compounds from 4 different groups of pesticides were detected. From the program, main crops consumed by Thai population were analyzed every year and almost half of the tested samples presented at least one pesticide. Organophosphorus, synthetic pyrethroids and carbamates were among the most detected pesticides and seemed to be annually increased. Two of organochlorine pesticides, DDT and endosulfan, were detected even the total banned in Thailand because of their persistent property in environment [7]. These insecticides are acute toxic pesticides and were used worldwide. There are neurotoxic and can inhibit the enzyme acetylcholinesterase [8]. The detected metabolites produced by certain pesticides have been shown to be carcinogenic in human [9-11]. Fungicide residues have also been found on some fruits and vegetables, mostly from post-harvest treatments and some of them are dangerous to human health [12].
This study aimed to use a multi-residue method developed in our laboratory to analyze 60 compounds of pesticide and their metabolites in four types of vegetable including Chinese broccoli, ivy gourd, yardlong bean and water spinach in Thailand. To our knowledge, no survey has been carried out during a whole country in every province on the level of pesticides contaminated in these four vegetables. These group of crops constitute major part of the Thai diet contributing to population required nutrients and vitamins. In addition, the vegetables analyzed in this study have a high consumption ratio in Thailand, and consumers may ingest the residues in fresh, semi-cooked and cooked food. The incidence of detected pesticides reported in 2016 was exceptionally high Chinese cabbage and water spinach (more than 97% in both cases). A wholesale market based survey was conducted to investigate the possible contamination of vegetables sold in the major provincial and Bangkok metropolitan markets during 2015. The residues of pesticide were examined by multi-residue extraction and were quantified by using gas chromatography coupled with electron capture detector (GC-ECD), gas chromatography coupled with flame photometric detector (GC-FPD) and liquid chromatography with post column derivatization coupled with fluorescence detector. The primary goal of this program is to determine if the amounts and types of pesticides found on vegetables are in accordance with the tolerances set by Ministry of Public Health of Thailand. Violations of the law occur when pesticides are not used in accordance with label registration and are applied in excessive amounts, or when pesticides are applied to crops on which they are not allowed. The findings from this study would also be useful for the Thai government to ascertain the MRL of pesticides in these commonly consumed vegetables, and to incorporate other pest management strategies for the safe and appropriate use of pesticides. Additionally, the results obtained from the monitoring program could be evaluated to Thai population dietary risk related to consumption of these vegetables. Governmental authority should use this information to develop the food safety action plan and to minimize the health risk due to pesticide residues.

2. Materials and methods

2.1 Samples

A total of 934 samples of four types of vegetables which are Chinese broccoli (n=254), ivy gourd (n=179), yardlong bean (n=248) and water spinach (n=253) were collected from wholesale markets by the Provincial Public Health Offices (PPHO) in collaboration with the Regional Medical Sciences Centers (RMSC) during 2015. It covered all Thailand’s territory including 76 provinces and Bangkok metropolitan area. The samples were collected with a minimum weighted of 1 kg and were packed in plastic bags with label. Then they were sent fresh to the accredited laboratory of the Bureau of Quality and Safety of Food (BQSF) for analysis of residues of 60 pesticides within 24 hours after collection. Four vegetables were selected because our unpublished primary results of monitoring program showed most contain chemical residues higher than other commodities and they are ingredients for Thai dishes the most admired [13]. The Codex guideline [14] (CAC/GL 50-2004) for sampling and sample preparation was used, and all samples were labeled and stocked at 4°C until the extraction was done. The samples were blended by homogenizer before weighing an analytical portion and other portion for internal quality control (IQC) sample. The rest of the blended sample portion was kept in a walk-in freezer at lower than -15°C.

2.2 Analytical method and instrumentation

Laboratory of pesticides and veterinary drugs residues of the BQSF was regularly inspected and authorized by the Bureau of Laboratory Quality Standard (BLQS) of the Department of Medical Sciences (DMSc) to ensure compliance with ISO/IEC 17025: 2005 requirements. The target pesticides included in this study were 20 organochlorines (OCs), 23 organophosphorus (OPs), 7 carbamates (CARs), 8 synthetic pyrethroids (SPs) and 2 fungicides. The AOAC 2007.01 official method [15] for extraction was modified and was employed for determination of OCs, OPs and SPs pesticides using GC-ECD and GC-FPD. Limit of quantitation (LOQ) was 0.05 mg/kg for gas chromatography technique. LC-amiable pesticides, CARs and Fungicides, were extracted by in-house method based on SPE extraction [16 & 17] and determined by HPLC-post column derivatization with the LOQ of 0.01 and 0.1 mg/kg, respectively. Both methods were validated and showed that its performance characteristics are adequate for use for this particular purpose.

2.3 Pesticide coverage

Compounds analyzed by the monitoring program included 5 groups, covered the list of 60 pesticides that can be detected by the methods. There were 20 organochlorine pesticides (OCs); aldrin, alpha-BHC, alpha-chlordane, gamma-chlordane, oxy-chlordane, pp’-DDE, pp’-TDE, pp’-DDT, dicofol, dieldrin, endrin, alpha-endosulfan, beta-endosulfan, endosulfan sulfate, heptachlor, trans-heptachlor epoxide, hexachlorobenzene, lindane, methoxychlor and tetrachloron. Other groups of pesticides included 23 organophosphorus (OPs); acephate, azinphos-methyl,
chlorpyrifos, diazinon, dichlorvos, dicrotophos, dimethoate, EPN, ethion, fenitrothion, malathion, methamidophos, methidathion, mevinphos, monocrotophos, ometheoate, parathion, parathion-methyl, phosalone, pirimiphos-methyl, profenofos, prothiofos and triazophos. Synthetic pyrethroid compounded 8 pesticides (SPs); bifenthrin, cyfluthrin, lambda-cyhalothrin, cypermethrin, deltamethrin, fenpropathrin, fenvalerate and permethrin. Carbamate pesticides (CARs) were aldicarb, carbaryl, carbofuran, 3-hydroxy carbofuran, methiocarb, methomyl and oxamyl. Finally, the two fungicides were carbendazim and thiabendazole. All pesticide standards were purchased from Dr. Ehrenstorfer (Augsburg, Germany). Purity of these pesticide standards was >96%.

3. Results and discussion

3.1 Monitoring general results for 2015

A total of 934 samples were analyzed for pesticide residues during 2015. Table 1. shows the five main pesticides classes analyzed in each commodity. Nearly eighty percent (77.7%) of the vegetable samples had no detectable pesticide residues. The number of samples which were positive for at least one pesticide (≥LOD) was 208 samples, representing 22.3% of total samples. Within all chemical properties compound groups, obviously no residual OCs was found in any sample. The levels of 33.9, 31.9, 10.6 and 9.5% of the samples were pesticides detectable in yardlong bean, Chinese broccoli, ivy gourd and water spinach, respectively. Moreover, residual OPs, SPs and CARs were found in the similar ratio in yardlong bean (12.9, 14.1 and 13.3%). Unsurprisingly, SPs pesticides, majority of cypermethrin, were the most frequently found in ivy gourd and water spinach and both had less pesticide residues than other vegetables, thanks to their resistance to pests and insects. In addition, slightly same proportion of OPs and SPs (15.7, 15.4%) were detectable in Chinese broccoli and this commodity was also the most contaminated by carbendazim (5.5%). Although the most popular classes of pesticides imported into Thailand are herbicides [18], no herbicide was investigated in this study. This was explainable by the limitation of the methods and instruments used.

Table 1. Samples analyzed by the Department of Medical Sciences monitoring program during 2015, as percent of positive samples and details with chemical group of pesticides.

<table>
<thead>
<tr>
<th>Vegetable</th>
<th>Sample analyzed</th>
<th>Positive samples, (%)¹</th>
<th>Groups of pesticides detected n, (%)²</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>OCs OPs SPs CARs FUNG</td>
<td></td>
</tr>
<tr>
<td>Chinese broccoli</td>
<td>254</td>
<td>81 (31.9)</td>
<td>0 (0) 40 (15.7) 39 (15.4) 8 (3.1) 14 (5.5)</td>
</tr>
<tr>
<td>Ivy gourd</td>
<td>179</td>
<td>19 (10.6)</td>
<td>0 (0) 3 (1.7) 15 (8.4) 5 (2.8) 1 (0.6)</td>
</tr>
<tr>
<td>Yardlong bean</td>
<td>248</td>
<td>84 (33.9)</td>
<td>0 (0) 32 (12.9) 35 (14.1) 33 (13.3) 8 (3.2)</td>
</tr>
<tr>
<td>Water spinach</td>
<td>253</td>
<td>24 (9.5)</td>
<td>0 (0) 2 (0.8) 21 (8.3) 1 (0.4) 3 (1.2)</td>
</tr>
<tr>
<td>Total</td>
<td>934</td>
<td>208 (22.3)</td>
<td></td>
</tr>
</tbody>
</table>

¹Presence of at least one pesticide residue at levels ≥ LOD.
²Number of target pesticides; 20 organochlorines (OCs), 23 organophosphorus (OPs), 7 carbamates (CARs), 8 synthetic pyrethroids (SPs) and 2 fungicides.

3.2 Irregularity of samples

A total of 189 samples (20.2%) were irregular (presence of a non-authorized, or residue levels higher than the Thai MRL) and these samples represented 90.9% of the positive samples (Figure 1). The Chinese broccoli and yardlong bean samples were the crops with the highest number of samples analyzed, having also the highest percentages of positive samples and percentages of irregular samples, representing 18.5 and 13.4% of the samples analyzed, respectively. The majority of the irregularities found in ivy gourd and water spinach for the period under study were related to the use of non-authorized chemicals (100% in both cases), mainly cypermethrin (37 samples), chlorpyrifos (4 samples) and carbendazim (4 samples).
The crops with the highest percentage of irregularity were Chinese broccoli and yardlong bean, mainly due to non-authorized organophosphorus compound, mostly chlorpyrifos, diazinon and profenofos. Figure 1 showed the percentage of residues that exceeded the national MRLs, considering all the positive samples. In about 20% of the cases, residues were up to 50% higher than the MRL, and in the majority of cases (65%), reached up to 2.5 times the MRL (15% exceeded of the MRL). The three highest residues above the MRL were cypermethrin in water spinach (5.82 mg/kg, MRL of 0.01 mg/kg), carbendazim and profenofos in Chinese broccoli (5.95 and 5.48 mg/kg, MRL of 0.01 mg/kg). Most of the residues (around 60%) found in the positive samples analyzed were in the range of 0.01 to <1 mg/kg, and only 4.3% had levels above 1 mg/kg.

3.3 Samples with multiple residues

Figure 2 shows the vegetable presenting multiple residues from 1 up to 6 different pesticides. Yardlong bean was the crop with highest number of samples with multiple residues, with one sample has contamination of 6 different pesticides including methamidophos, acephate, chlorpyrifos, profenofos, ethion and cypermethrin. Chinese broccoli has the second highest number of pesticides contamination with 5 types of residues of diazinon, chlorpyrifos, profenofos, alpha-cyhalothrin and cypermethrin.
The main compounds found in each vegetable sample analyzed were shown in Figure 3. Cypermethrin was the pesticide most frequently detected in all commodities (36, 34, 15 and 21 samples in Chinese broccoli, yardlong bean, ivy gourd and water spinach, respectively. Cypermethrin is a moderately toxic to human but other animals, especially fish, are particularly susceptible to mortality effect [19]. Carbendazim, a benzimidazole systemic fungicide, was the single compound most frequently detected in all vegetables. Carbendazim (methyl 2-benzimidazolecarbamate) is widely used as a systemic fungicide in human food production because of its low toxicity. Nevertheless, it also inhibits proliferation of human cancer cells [20]. One yardlong bean sample had a residue of methamidophos which has been banned of use in Thailand since 2003 because it is highly acute toxic and high risk to user and it is classified as a WHO Toxicity Class “Class 1b” [21].

Other pesticides found in crops samples were registered and were authorized for use in Thailand [20]. However, some substances were misused and were prohibited for application to some vegetables due to their systemic and persistence properties. The four organophosphorus compounds most frequently found in the samples analyzed were chlorpyrifos, diazinon, profenofos and ometoate, representing the main problem for residual contaminants. Methomyl and carbofuran were two pesticides in carbamate group that were the most presented in all vegetables. Some yardlong bean samples had residues of organophosphorus compounds plus carbamates, compounds known to be inhibitors of the enzyme acetylcholinesterase (AChe).

**Figure 3** The pesticides most frequently detected in the samples by the DMSc monitoring program in 2015.

### 3.4 Evaluation by Health Areas

Under regulatory monitoring, samples were collected by Provincial Public Health Office (PPHO). In Thailand, each PPHO was affected to Health Area (HA) numbered 1 to 12 (no.1- no.12) except Bangkok where we gave unofficially number “0”. Samples were analyzed and were grouped by HA to be traceable which region has severe contamination problems. Figure 4. shows that these 934 samples were domestic foods, residuals were found with higher ratio in HA no.1 to HA no.6 and it shows more detailed data on domestic monitoring findings by commodity, including the percentage of detected samples and analyzed samples. The HAs which had the important percentage of pesticide detectable samples were located in north and central region. If Thai authority needs to improve the safety of vegetable, the most urgent cases of bad agricultural practice were found in theses suggested areas and would be solved.
3.5 Chronic risk assessment for dietary intake

The risk assessment from intake of pesticide residues in these vegetables was evaluated for adult populations (60 kg body weight, bw) based on the median or the highest experimental results of 6 top representative pesticides by employing the average daily consumption of 155.2 g vegetables/person from the national data documented by the National Health Examination Survey Office (NHESO) and the respective Acceptable Daily Intake (ADI) values from Codex pesticides databases. The results are expressed as percentages (Table 2) based on the Equation 1.

\[
ER(\%) = \left(\frac{(PR \times ADC)}{BW} \right) \times 100 \frac{ADI}{ADI}
\]  

where:  
- **ER** = Estimated chronic risk assessment of pesticide residues from vegetables intake  
- **PR** = the median or the highest pesticide residue results found (mg/kg)  
- **ADC** = Average Daily Consumption of vegetable (g/person/day)  
- **BW** = Body weight (kg)  
- **ADI** = Acceptable Daily Intake (mg/kg bw/day)

**Table 2** Estimative chronic risk assessment for Thai population due to vegetables intake.

<table>
<thead>
<tr>
<th>Pesticides</th>
<th>ADI1 (mg/kg bw)</th>
<th>Concentration levels found (mg/kg)</th>
<th>Estimated chronic intake (mg/person/day)</th>
<th>Estimated risk characterization (% ER)</th>
</tr>
</thead>
<tbody>
<tr>
<td>cypermethrin</td>
<td>0.02</td>
<td>median 0.27</td>
<td>0.0007</td>
<td>3.49</td>
</tr>
<tr>
<td></td>
<td></td>
<td>highest 5.82</td>
<td>0.0151</td>
<td>75.27</td>
</tr>
<tr>
<td>chlorpyrifos</td>
<td>0.01</td>
<td>median 0.06</td>
<td>0.0002</td>
<td>1.55</td>
</tr>
<tr>
<td></td>
<td></td>
<td>highest 1.90</td>
<td>0.0049</td>
<td>49.15</td>
</tr>
<tr>
<td>diazinon</td>
<td>0.005</td>
<td>median 0.05</td>
<td>0.0001</td>
<td>2.59</td>
</tr>
<tr>
<td></td>
<td></td>
<td>highest 0.30</td>
<td>0.0008</td>
<td>15.52</td>
</tr>
<tr>
<td>carbadaziom</td>
<td>0.03</td>
<td>median 0.40</td>
<td>0.0010</td>
<td>3.45</td>
</tr>
<tr>
<td></td>
<td></td>
<td>highest 5.95</td>
<td>0.0154</td>
<td>51.30</td>
</tr>
<tr>
<td>methomyl</td>
<td>0.02</td>
<td>median 0.09</td>
<td>0.0002</td>
<td>1.16</td>
</tr>
<tr>
<td></td>
<td></td>
<td>highest 0.89</td>
<td>0.0023</td>
<td>11.51</td>
</tr>
<tr>
<td>carbofuran</td>
<td>0.001</td>
<td>median 0.05</td>
<td>0.0001</td>
<td>12.93</td>
</tr>
<tr>
<td></td>
<td></td>
<td>highest 1.00</td>
<td>0.0026</td>
<td>258.67</td>
</tr>
</tbody>
</table>


The assessment of the estimated chronic risk from vegetables intake is indicated in Table 2. The health risk characterization, based on the median and highest results found in this work for top 6 often found, ranged from 1.16 to 12.93% and from 11.51 to 258.67% of the ADI respectively for Thai population. Dietary exposure to pesticides is of health concern due to the potentially toxic effects. Vulnerable groups such as the elderly, diabetics, or pregnant women may be more susceptible to pesticides. Children are much more sensitive to the toxicity of contaminants than adults because of their lower body weight and active developmental processes.
3.6 Discussion and suggestion

The DMSc pesticides monitoring program data collected in this paper shows that 22.4% of the 934 samples analyzed during 2015 were positive for at least one pesticide residue. A recent report from the ThaiFDA in 2009 indicated that only 3.25 percent of 54,140 fresh food samples available in local markets were contaminated with pesticide residues. These results were obtained from a monitoring program using screening test kit with low sensitivity and selectivity. The results from our laboratory are more likely to be precise and converge to reflect the common situation because the capacity with sophisticated instruments and international standard methods were applied. On the other hand, Wanwimoetrak S. et al. found that over 97 percent of vegetable samples available in markets and supermarkets in Thailand during 2013 were pesticides detected. These rates are questionable and are much higher than those seen in any countries in the world. Pesticide residues determination is one of the most complicated fields of sciences. Experiences in extraction step and the ability for using instruments for detection are required to be able to quantify correctly such analytes. The lake of experimental training could translate into important error of results interpretation. Chinese broccoli and yardlong bean were the crops with the highest percentage of positive samples (around 30%). For area based studied, samples produced in central and northern region were more likely to contain residues than those from north-eastern and southern areas. It was explainable by the different climatic condition of each region and its intensive production destined for sell or for consume locally. However, the prevalence of non-authorized pesticides in this study was very low. The pesticide formulation management organized by the Department of Agriculture (DOA), Ministry of Agriculture and Cooperatives has successfully applied. Some problems still subsist in rural area due to the profile of the agricultural population of the country. On the other hand, high percentage of irregular samples was the use of pesticides with no MRL. It is possible that a farmer will use a pesticide registered for on crops in other also grown on the property, regardless of its registration status. Farmers with limited education and lack of knowhow on handling the pesticides preferred mixing high potency pesticides to make highly toxic cocktails, increasing pesticide dosages over recommended limits, for strong and fast acting pesticides. In order to minimize non-authorized pesticides used on minor crops, the Food and Drug Administration of Thailand (Thai FDA) who is responsible for implementation in accordance with the national legislation has to extend the MRL to all crops within a group based on supervised residues analyses conducted only on representative crops for that group. Furthermore, Thai government authorities should facilitate the implementation of regulations and laws on pesticide residues and food safety. These are extremely primordial to reduce the health risks of local consumers associated with pesticide residues in vegetables.

4. Conclusion

Residue data obtained in this study indicated that good agricultural practices (GAP) are not being followed by Thai farmer, since most of pesticides found in vegetables are higher than MRL or are not authorized for use. The results show a potential human dietary risk related to consumption of these vegetables. In order to ensure the safety of our food, governmental action plans should be developed with the cooperation of producers to reduce pesticide residues. National regulatory agencies included in food chain must cooperate synchronously and continually. Finally, scientific data on residues and other aspects of chemical use must be open to the public, to satisfy the right-to-know of citizens.

5. Acknowledgements

The authors acknowledge Miss Jaruwan Limsajasakul, the director of the Bureau of Quality and Safety of Food and are grateful to the Regional Medical Sciences Centers.

6. References


