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Organophosphate Pesticide on Fruits: Residue and Health Risk

Abata, E.O[1]

[1]*Department of Chemistry, School of Physical Sciences,*
Federal University of Technology, Akure, Ondo State, Nigeria.

Abstract

Fruits contain a lot of minerals and vitamins needed by human which is been advised even by nutritionist, but the production of this class of food is raising concerns because of the chemical products used during cultivation of these crops to ensure high yield and durability. The use of organophosphate pesticides has been favoured in some places like the US because of their fast decomposition after usage, but there are guidelines needed to be followed to ensure safe use and less pesticide residue in agriculture produce. This study determined the organophosphate residues in seven (7) fruits which are possible contamination route for human because they are often eaten raw, and also serve as data toward achieving a healthier community.

The result showed Bromophos is detected in watermelon, pineapple and cucumber with a concentration of 0.55mg/kg each, while chlorpyrifos is highest in banana with a concentration of 3.43mg/kg and orange showing the least concentration of fenamiphos with a value of 0.07mg/kg. Watermelon shows the highest pesticides residue of 12.72 mg/kg while orange had the least sum of 5.93 mg/kg. Some of this suggest a possible risk for the farmers themselves or those close to where the pesticides were used. The health hazard index of ethion suggest a possible health risk most especially with the children group. A drastic awareness, law and way to ensure farmers follow a good agricultural practice and pesticide usage is strongly advised.

Introduction

Agriculture to a large extent has contributed to the sustenance of human life, because it has ensured a continual production of crops and animals to meet a relative demand with comparison to the human population increase. The increase in production has in no small way being aided by the advancement in science and technology. This has created innovative ways to ensure better crops, high yield, long shelf life, resistance to pest and diseases and has ensure easy availability to the consumers. Some of these innovations include farm machineries, fertilizers, pesticides, hybrid seeds, microorganism for soil amendment, irrigation, trainings and others, while some are easy in application on farm, some are operated by experts or experienced operators and some are used by some farmers in ways they deem fit as long as yield is increasing or expected yield is met. Some of these include the use of fertilizers and pesticides. It has been reported that the indiscriminate application of pesticides on crops by farmers could be as a result of lack of training, money and possible illiteracy of farmers [1] for developing countries this can be high.

Pesticides are synthetic chemicals used to control plant disease and pests, these categories of chemicals if used moderately can greatly contribute to agricultural output. Recent industrial agricultural practices without these group of chemicals seems impossible [2] because produce loss can result in food scarcity threat [3]. There are many pesticides that has been banned in developed countries such as dichlorodiphenyltrichloroethane (DDT) [4], there are some that are persistent in nature after usage (does not break down easily) such as organochlorine pesticides while some are less persistent because they breakdown easily such as the organophosphates. Organophosphates are widely used in United States because of their acute toxicity level and these group of pesticides are commonly used on fruits [5]. Some of organophosphate pesticides (OPPs) include parathion, diazinon, malathion, dimethoate, chlorpyrifos and many more have been in existence since the 1940’s with high acute mammalian toxicity but easily breaks down [6]. Studies of effect of these pesticides has been made on occupational exposed people (farm workers and pesticides applicators) showing acute OPPs poisoning which can be severe. Its symptoms include nausea, dizziness, abdominal cramps, diarrhoea and convulsion [5]. A national representative study showed increasing level of OPP metabolites in urine of Attention deficit disorder /Attention deficit hyperactivity disorder ADD/ADHD for 8-15 years old [7].

Fruits contain essential minerals and vitamins needed by the body yet the use of pesticides makes it a major concern because of the possibility of pesticide residues [4], since most of fruits can be eaten raw and the major route of exposure of pesticides for non-occupational set of people is through the residual on the fruits through the mouth. In a research conducted covering a total of 141 countries, with additional data from the WHO Mortality Database. An estimation of about 385 million cases of pesticide poisoning, including around 11,000 fatalities occur annually world-wide. Based on a worldwide farming population a deduction was made that, about 44% of farmers are poisoned by pesticides every year [8] (Boedeker *et al*. 2020). The misuse of pesticides can lead to exposure through food and water and when people come in contact with large quantities, may lead to acute posing or long-term health effects, including cancer and adverse effects on production [4] (WHO, 2018). This made a major concern in developing countries like Nigeria especially in produce that can be eaten raw. This demand a continual assessment to ensure enough data is available in addressing the public health and adhering to international standards. This study aimed to determine the level of organophosphate residues in raw eaten fruits, compare it to acceptable daily intake and relate it to the health risk index.

Materials and Methods

Materials

The fruits (water melon, pineapple, orange, cucumber, cherry and banana) were purchased from Oja Oba market, Akure, Ondo State.

All chemicals used were of analytical grade.

All glass ware used (conical flask, beakers, measuring cylinder, volumetric flask) were washed with detergents and later soaked with the solvent, after which they were dried in an oven over a period of time prior to usage.

Methodology

➢ Extraction

Each fruit sample were washed thoroughly with distilled water and placed in a motar, the fruit were converted into paste separately by using mortar and pestle, and then spatula was used to move them to separate conical flask after which 20g of each sample was weighed on the weighing balance then 40ml of ethyl acetate was added and shaken thoroughly. A 5g of sodium hydrogen carbonate was added to the mixture to neutralize any acid present in the mixture followed by 20g of anhydrous sodium sulphate and the entire mixture was shaken vigorously for 1 hour. The procedure was repeated for other samples and the mixture was filtered into a labelled container before centrifuging at a speed of 1800rpm for 5 minutes. The organic layer was decanted into a conical flask and 5m of ethyl acetate was added [9]

➢ Clean Up of Fruits Extract

The fruits extract was cleaned up as follows: 10mm chromatographic column was filled with 3g activated silica gel and topped up with 2g of anhydrous sodium sulphate and 5ml of n- hexane was added thrice with the 2ml hexane.

The procedure was repeated for all the samples. The sample was collected in a 2ml vial sealed and placed in the refrigerator [10].

Qualitative Identification and Quantitative Estimation of the Organophosphate Pesticides

Qualitative identification and quantitative estimation of the pesticide residues were performed by reconstituting the dried sample eluents with 1 mL n-hexane. With the aid of a micro syringe, the injection of 1μL of the purified eluents was performed in a splitless injection mode on to the injection port of an Agilent 5977B Gas Chromatograph (GC) system equipped with Electron Capture Detector (ECD). Carrier gas was Helium at a flow rate of 1.2 mL/min and make up gas was Nitrogen. The run time was 25 minutes. The identification of OPPs was done by comparing the retention times of the peaks with those obtained from standard mixture of OPPs, while the quantification was based on external calibration curves prepared from the serially diluted standard solution of each of the OPPs. The separation was performed on a fused silica capillary column (DB-17, 30 m long x 0.250 mm internal diameter and film thickness of 0.25 µm). The temperatures of the injector and detector were 250^{o}C and 290^{o}C respectively. Oven temperatures programme started from 150^{o}C and increased to 280^{o}C at 6^{o}C per minute. The instrumental analysis was done at the Nigeria Institute of Oceanography and Marine Research (NIOMR) Laboratory, Victoria Island, Lagos, Nigeria.

Results and Discussions

Concentrations (mg/kg) of Organophosphate Pesticides in Fruits

Table 1 the concentrations of organophosphate in the fruits studied. The concentrations ranged from 0.124mg/kg of fenamiphos to 1.424mg/kg of azinfos-methyl. Dichlorvos showed a mean concentration of 0.167mg/kg. Mevinfos concentration is high in cherry with 0.49mg/kg followed by apple and banana with 0.29mg/kg and 0.28mg/kg respectively while the rest show 0.23mg/kg,0.24mg/kg and 0.26mg/kg. Diazinon ranges from 0.26 to 0.3mg/kg with water melon and pineapple having 0.3mg/kg each and banana having the least of 0.26 mg/kg. Etrimfos is not detected in banana while the rest showed between 0.23 to 0.24mk/kg. Phosphamidon shows a high concentration with 0.29 mg/kg in cucumber and the least in cherry with 0.18mg/kg. Parathion-methyl concentration ranges from 0.28 to 0.53mg/kg with apple showing the highest with 0.53mg/kg and pineapple showing the lowest with 0.28mg/kg. Fenitrothion is highest in cherry with 0.77mg/kg followed by cucumber with 0.59mg/kg then apple with 0.53mg/kg while pineapple showed the least with 0.31mg/kg. Pirimiphos-methyl showed a mean concentration of 0.269±0.007 across all the samples while malathion showed a mean concentration of 0.213±0.023. Chlorpyrifos showed highest concentration of 3.43mg/kg in banana followed by 3.12mg/kg in watermelon, next is 2.99mg/kg in cherry while the least is 0.71mg/kg in orange. The high occurrence of chlorpyrifos maybe a result of its accumulation and persistence in food matrices [11]. Bromophos-ethyl is not detected in orange, cherry, apple and banana while showing a mean concentration of 0.55mg/kg each in watermelon, pineapple and cucumber. Chlorfenvifos from the Table 1 showed a mean concentration of 0.247mg/kg while bromophos-methyl show a high concentration of 3.72mg/kg in cherry when compared to others followed by 1.4mg/kg in orange and the least is 0.42mg/kg in banana. Fenamiphos is lowest in orange with 0.07mg/kg followed by pineapple with 0.08mg/kg, next is 0.1mg/kg in cucumber and watermelon while a concentration of 0.2mg/kg in cherry as the highest. Ethion ranges from 0.08mg/kg in orange and cucumber to 1.42mg/kg in watermelon. Carbofenothion was not detected in pineapple while orang and cucumber showed 0.13mg/kg concentration each followed by 0.15 and 0.16mg/kg in banana and cherry respectively, then 0.25 and 0.37mg/kg in apple and watermelon respectively. Azinfos-methyl showed also a concentration range of 0.79mg/kg (in orange, cucumber and cherry each) to 2.89mg/kg in watermelon. Watermelon showed the highest sum of concentration to be 12.72mg/kg, followed by cherry with 11.58mg/kg, while orange having the lowest with 5.93mg/kg. The order is watermelon> cherry > banana > apple >

pineapple > cucumber > orange. Among the eighteen organophosphate residues studied, bromophos-methyl with a concentration of 1.00mg/kg is lowest in watermelon and a concentration of 2.89mg/kg of azinfos-methyl is highest. Carbofenothion was not detected in pineapple but fenamiphos was, with a value of 0.08mg/kg (lowest) and azinfos-methyl with 2.18mg/kg (highest). In orange, bromophos-methyl is the only one above 1mg/kg while bromophos-ethyl was not detected and the rest has values below 1. In banana sample, etrimfos and bromophos-ethyl was not detected, dichlorvos, mevinfos, dimethoate, diazinon, phosphamidon, pirimiphos-methyl, malathion, chlorfenvifos, fenamiphos and carbofenothion fall below 0.3mg/kg while ethion, bromophos-methyl, fenitrothion and parathion-methyl fall between 0.3-0.65mg/kg then azinfos-methyl and chlorpyrifos are greater than 1mg/kg (1.31mg/kg and 3.43mg/kg respectively).

The concentration values in this study are higher than those reported by [12] which maybe a result from the need to use more of these pesticides yearly [13] or the level of how well informed the farmers are, in engaging in good agricultural practices and pesticides use [14] This scenarios of misuse of pesticides leaving its residue in crops could be one of the reasons why international market may reject exported crops when standards are not met [15]. Most of the residue in the samples in this study exceeded their maximum residue limit (MRL) given by [16]

Table 1 Concentrations (mg/kg) of Organophosphate Pesticides in Fruits [end of the chapter]

Hierarchical Cluster Analysis of Organophosphate Pesticides in the Studied Samples

The hierarchical cluster analysis was used to determine the relationship among the various pesticides using Euclidean distance as measure of similarity. This was performed using Statistical Package for Social Scientist (SPSS). Figure 1 shows the clustering analysis of the organophosphate pesticides in the studied fruit samples.

Cluster analysis (CA) grouped the organophosphate pesticides into clusters on the basis of similarities within a group and dissimilarities between different groups. Parameters belonging to the same cluster are likely to have originated from a common source and or similar chemical properties. The cluster analysis performed on the samples produced three major groups namely, A (Azinfos-methyl and chlorpyrifos), B (parathion-methyl and ethion), and C (diazinon, pirimiphos-methyl, phosphamidon, chlorfenvifos, malathion, mevinfos, dimethoate, etrimfos, dichlorvos, fenamiphos, carbofenothion, parathion-methyl, fenitrothion, and bromophos-ethyl) which showed closest clustering relationship. The close clustering relationship of the pesticides might be indicative of similar sources or suggestive that the pesticides are affected by similar environmental factors.

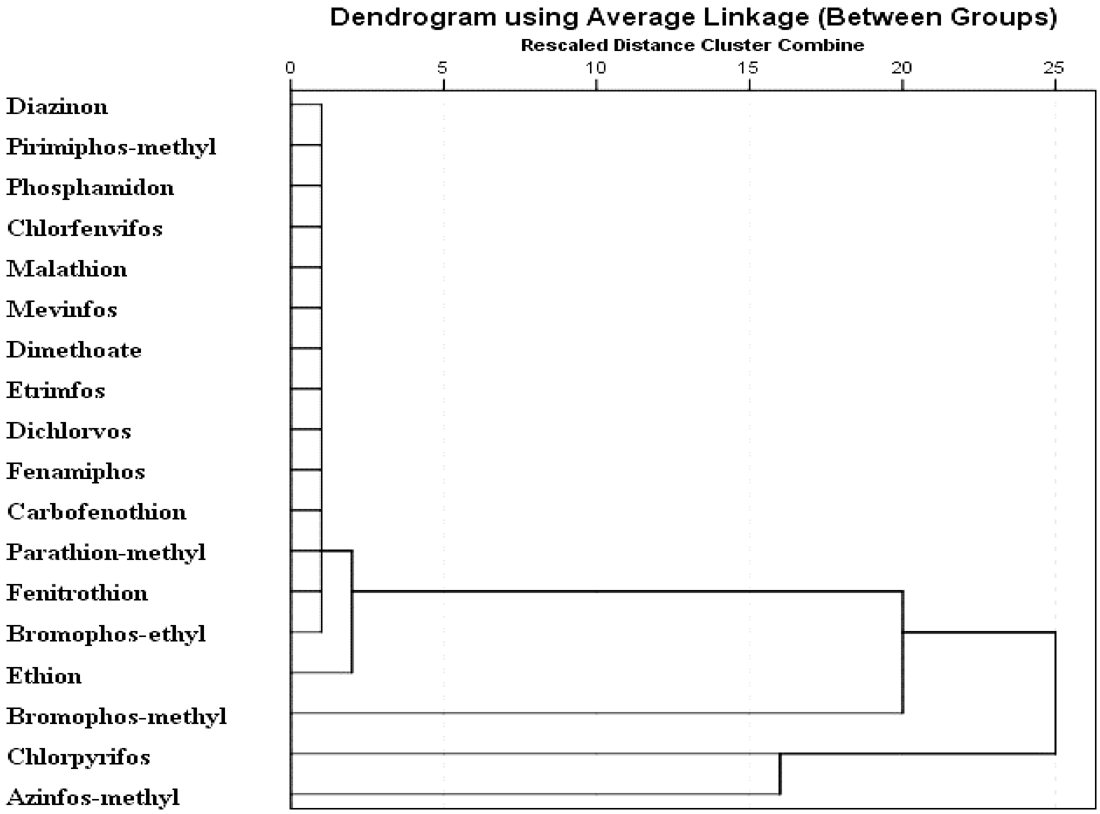

Figure 1.Hierarchical cluster analysis of organophosphate pesticides in fruits

Correlation Matrix of Organophosphate Pesticides in the Studied Samples

Correlation coefficient measures the srenght of the linear relationship between any two variables on a scale of -1 (perfect inverse relation) through 0 (no relation) to +1 (perfect sympathetic relation). In this study, the raw data was used in calculating the correlation coefficient using the Microsoft Excel computation software package.

Table 2 shows the correlation matrix of the organophosphate pesticides in the studied fruits. Positive and strong significant correlations exist between fenitrothion/mevinfos, pirimiphos-methyl/ mevinfos, chlorpyrifos/ mevinfos, bromophos-ethyl/ mevinfos, fenamiphos/ mevinfos, pirimiphos-methyl/ etrimfos, malathion/ etrimfos, fenitrothion/ parathion-methyl, fenamiphos/ parathion-methyl, carbofenothion/ parathion-methyl, pirimiphos-methyl/ fenitrothion, chlorpyrifos/ fenitrothion, bromophos-methyl/ pirimiphos-methyl, bromophos-ethyl/ malathion, fenamiphos/ chlorpyrifos, ethion/ chlorpyrifos, carbofenithion/ chlorpyrifos, carbofenithion/ ethion, and azinfos-methyl/ethion. Strong and significant positive correlations indicated chemical affinity, similar genetic origin and/or common background levels in the sample.

Table 2 Correlation matrix of organophosphate pesticides in fruits [end of the chapter]

Health Risk Assessment of Organophosphate Pesticides in Fruits

The health risk assessments were presented in Table 3 below, the table showed the estimated daily intake and health hazard index (HHI) based on exposure to organophosphate pesticides through ingestion of fruits.

The HHI<1 indicate a safe level with malathion and fenitrothion, while ethion, parathion, diazinon and fenitrothion has HHI>1 suggesting a possible health risk, most especially for the children age group. Related health risks due to the consumption of all the fruit and vegetables samples by children indicates they more susceptible to the pesticide contaminants than adults [15, 17] reported a similar trend. The high HHI values of diazinon pesticide residue gives the highest possible risk (that is noncarcinogenic and carcinogenic health risks).

Table 3 Health risk assessment of organophosphate pesticides in fruits [end of the chapter]

Conclusion

The level of organophosphate residue in the studied fruits clearly showed a higher level which raise a major concern for the children from the health hazard analysis and these require prompt action since they are still developing. WHO recommendation is strongly advisable before consumption of these class of food, which include; washing before eating and peeling of fruit skins before eating raw. There should be a random visit of extension officers to ensure every farmer has a record book of every input especially the chemical-form input. A regular visit of expert to farmers for sample collection and encourage the farmers to always send some of their produce to the agricultural ministries where they will be tested for chemical residues and uptake by plants, and relate it to their chemical input. Ultimately it requires another agency linked to the food and drugs agency (National Agency for Food and Drug Administration and Control) that will handle locally produced raw food crops been circulated in the community.

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Table 1.Concentrations (mg/kg) of Organophosphate Pesticides in Fruits

Pesticides	**Water melon**	**Pineapple**	**Orange**	**Cucumber**	**Cherry**	**Apple**	**Banana**	**Mean±SD**
Dichlorvos	0.17	0.17	0.17	0.16	0.16	0.17	0.17	0.167 ±0.005
Mevinfos	0.24	0.23	0.24	0.26	0.49	0.29	0.28	0.29 ±0.091
Dimethoate	0.26	0.27	0.26	0.27	0.28	0.26	0.26	0.266 ±0.008
Diazinon	0.3	0.3	0.29	0.28	0.27	0.27	0.26	0.281 ±0.016
Etrimfos	0.24	0.23	0.23	0.23	0.23	0.23	bdl	0.199 ±0.088
Phosphamidon	0.23	0.2	0.2	0.29	0.18	0.22	0.28	0.229 ±0.042
Parathion-methyl	0.52	0.28	0.29	0.48	0.52	0.53	0.39	0.43 ±0.11
Fenitrothion	0.45	0.31	0.33	0.59	0.77	0.53	0.46	0.491 ±0.158
Pirimiphos-methyl	0.27	0.26	0.27	0.27	0.28	0.27	0.26	0.269 ±0.007
Malathion	0.23	0.24	0.2	0.22	0.21	0.22	0.17	0.213 ±0.023
Chlorpyrifos	3.21	0.75	0.71	0.89	2.99	2.94	3.43	2.131 ±1.272
Bromophos-ethyl	0.55	0.55	bdl	0.55	bdl	bdl	bdl	0.236 ±0.294
Chlorfenvifos	0.27	0.19	0.27	0.3	0.19	0.29	0.22	0.247 ±0.046
Bromophos-methyl	1	0.55	1.4	0.9	3.72	0.5	0.42	1.213 ±1.158
Fenamiphos	0.1	0.08	0.07	0.1	0.2	0.14	0.18	0.124 ±0.050
Ethion	1.42	0.22	0.08	0.08	0.14	0.27	0.61	0.403 ±0.484
Carbofenothion	0.37	bdl	0.13	0.13	0.16	0.25	0.15	0.17 ±0.115
Azinfos-methyl	2.89	2.18	0.79	0.79	0.79	1.22	1.31	1.424 ±0.815
$\sum OPPs$	12.72	7.01	5.93	6.79	11.58	8.6	8.85	

OPP = organophosphate pesticide, SD = standard deviation, bdl = below detection limit

Table 2.Correlation matrix of organophosphate pesticides in fruits

	Dichlorvos	**Mevinfos**	**Dimethoate**	**Diazinon**	**Etrimfos**	**Phosphamidon**	**Parathion-methyl**	**Fenitrothion**	**Pirimiphos-methyl**	**Malathion**	**Chlorpyrifos**	**Bromophosethyl**	**Chlorfenvifos**	**Bromophos-ethyl**	**Fenamiphos**	**Ethion**	**Carbofenothion**	**Azinfos-methyl**
Dichlorvos	1																	
Mevinfos	-0.55	1																
Dimethoate	0.17	0.02	1															
Diazinon	0.1	-0.16	-0.41	1														
Etrimfos	-0.25	0.22	0.18	0.36	1													
Phosphamidon	-0.04	-0.08	-0.09	-0.05	-0.09	1												
Parathion-methyl	-0.49	0.54	-0.32	0.07	0.39	0.32	1											
Fenitrothion	-0.7	**0.85**	-0.04	-0.25	0.32	0.22	**0.75**	1										
Pirimiphos-methyl	-0.56	**0.58**	-0.31	0.07	**0.56**	-0.11	**0.69**	**0.71**	1									
Malathion	-0.13	-0.14	-0.13	0.51	**0.69**	-0.2	0.19	0.02	0.29	1								
Chlorpyrifos	-0.16	**0.55**	-0.29	-0.11	-0.04	0.25	0.71	**0.59**	0.34	-0.15	1							
Bromophos-ethyl	0.09	-0.36	-0.13	0.49	0.38	0.35	0.11	-0.11	0.07	**0.63**	-0.23	1						
Chlorfenvifos	-0.01	-8.26E-17	0.13	0.16	0.3	0.49	0.32	0.18	0.05	-0.01	0.12	-0.05	1					
Bromophos-ethyl	-0.28	**0.68**	0.24	-0.13	0.42	-0.21	0.23	**0.6**	**0.66**	0.04	0.11	0.03	-0.19	1				
Fenamiphos	-0.32	**0.81**	-0.22	-0.3	-0.15	0.25	**0.59**	**0.77**	0.38	-0.35	**0.8**	-0.31	-0.04	0.39	1			
Ethion	0.16	-0.03	-0.26	0.39	-0.02	0.25	0.41	0.04	0.01	0.1	**0.65**	0.21	0.2	-0.2	0.18	1		
Carbofenothion	0.1	0.1	-0.04	0.03	0.12	0.15	**0.55**	0.28	0.32	0.04	**0.61**	-0.02	0.47	0.04	0.2	**0.72**	1	
Azinfos-methyl	0.39	-0.09	0.22	0.48	0.16	0.08	0.03	-0.17	-0.34	0.29	0.26	0.29	0.22	-0.18	-0.08	**0.71**	0.38	1

n= 12, r ≥ 0.55 at 95% confidence interval

Table 3.Health risk assessment of organophosphate pesticides in fruits

		Estimated daily intake (EDI)						
Pesticides	**Population**	**Water melon**	**Pineapple**	**Orange**	**Cucumber**	**Cherry**	**Apple**	**Banana**
Chlorpyrifos	Adults	0.015	0.003	0.003	0.004	0.014	0.014	0.016
	Children	0.01	0.016	0.01	0.012	0.012	0.003	0.042
Malathion	Adults	0.001	0.001	0.0009	0.001	0.001	0.001	0.0008
	Children	0.3	0.003	0.003	0.002	0.003	0.002	0.003
Ethion	Adults	0.006	0.001	0.0003	0.0003	0.0006	0.001	0.003
	Children	0.002	0.001	0.003	0.001	0.001	0.001	0.003
Parathion	Adults	0.002	0.001	0.001	0.002	0.002	0.002	0.001
	Children	0.001	0.004	0.004	0.004	0.006	0.003	0.007
Diazinon	Adults	0.001	0.001	0.001	0.001	0.001	0.001	0.001
	Children	0.0002	0.003	0.004	0.003	0.004	0.003	0.003
Fenitrothion	Adults	0.002	0.001	0.001	0.002	0.003	0.002	0.002
	Children	0.005	0.006	0.004	0.006	0.008	0.003	0.007
Health Hazard Index (HHI)								
Chlorpyrifos	Adults	1.582	0.369	0.349	0.438	1.473	1.449	1.69
	Children	4.654	1.087	1.029	1.29	4.335	4.263	4.973
Malathion	Adults	0.003	0.003	0.003	0.003	0.003	0.003	0.002
	Children	0.011	0.011	0.009	0.01	0.01	0.01	0.008
Ethion	Adults	3.499	0.542	0.197	0.197	0.345	0.665	1.503
	Children	10.295	1.595	0.58	0.58	1.015	1.957	4.422
Parathion	Adults	2.562	1.38	1.429	2.365	2.562	2.612	1.922
	Children	7.54	4.06	4.205	6.96	7.54	7.685	5.655
Diazinon	Adults	7.392	7.392	7.1464	6.9	6.653	6.653	6.407
	Children	21.75	21.75	21.025	20.3	19.575	19.575	18.85
Fenitrothion	Adults	0.443	0.305	0.325	0.581	0.759	0.522	0.453
	Children	1.305	0.899	0.957	1.711	2.233	1.537	1.334