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Organochlorine Pesticide Residual Levels in Fruit Juice Produced in Accra, Ghana

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ABSTRACT

Organochlorine pesticides (OCPs) were determined in twenty (20) pineapple and cocktail fruit juice of eight (8) brands collected from various markets in the Greater Accra Region, Ghana. Each brand was replicated thrice or twice based on diffrrent batch number. A survey was conducted to ascertain farmers view on organochlorine pesticides and pesticide/insecticide used during pineapple and mango production. Fruit juice samples were also analysed to investigate pesticide residue levels in them. Fruit juice samples were analysed by gas chromatography prepared with electron capture detector for organochlorines. Thirty (30) mango and twenty-five (25) pineapple farmers interviewed used pyrethroid based pesticides in controlling insect pest on their farm.Aldrin (0.007 ± 0.004 mg/kg), Dieldrin (0.001 ± 0.003 mg/kg), Endrin (0.0004 ± 0.001 mg/kg) cis heptachlor (0.000001 ± 0 mg/kg), Heptachlor (0.09 ± 0.088 mg/kg), trans heptachlor (0.0003 ± 0.001 mg/kg), trans_nonachlor (0.003 ± 0.004 mg/kg), alpha_HCH (0.002 ± 0.001 mg/kg), beta_HCH (0.000001 ± 0 mg/kg), delta_HCH (0.007 ± 0.008 mg/kg), gamma_HCH (0.003 ± 0.006 mg/kg), cis chlordane (0.000001±0 mg/kg), Hexachlorobenzene (0.0008 ± 0.001 mg/kg), o_p_DDD (0.00024 ± 0.001 mg/kg), p_p_DDD (0.005 ± 0.007 mg/kg) o_p_DDE (0.000001 ± 0 mg/kg), p_p_DDE (0.0006 ± 0.001 mg/kg), o_p_DDT (0.0009 ± 0.001 mg/kg), and p_p_DDT (0.0003 ± 0.001 mg/kg) were present in all the samples collected. The observations made on this studies, suggest that, effective regulations guiding the use of pesticides on fruits should be enforced and regular checks of pesticide residues in freshly consumed products.

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Introduction

Consumption of locally produced natural fruit has increased tremendously in Ghana and increasing daily. This is due to the growing health conscious middle class and the campaign of consuming of made in Ghana products. The consumption of natural fruit juice range from production onsite such as at restaurants, juice bars to bottled fruit juice. This industry has resulted in over 37 companies producing fresh juice. However, the safety of these products is of paramount concern to consumers and international trade such as pesticide residue.

The current trends of pesticide usage in crop production in Ghana have raised concerns about the safety of crops produced and their related products. There has been exponential increase in the quantity and use of pesticides in agriculture since 2002 (Hodgson, 2003, Fianko *et al.,* 2011). These pesticides are used as deterrent against pests and diseases to avoid agricultural yield loss, or as disease trajectory control in the public health sector (Bempah *et al.,* 2011) but tend to pose health risk resulting from pesticide residues in food (Eskenazi *et al*., 2008). As a result, pesticide residue in food commodities has become a topical issue among researchers, government agencies or institutions. Pesticide applied during vegetable and fruit production range from pyrethroids, organophosphates, carbamates or organochlorines, or a combination (Fianko *et al.,* 2011 and Gonu *et al.*, 2012).

Among the groups of pesticide, organochlorine pesticides (OCPs) are noted for being potent in controlling insect pest and diseases hence farmers resort to the use of OCPs despite the obvious health hazards. The persistent nature of OCPs in the environment has raised concerns about the safety of the environment, vegetables and fruits being produced by farmers. Since dietary intake of pesticide residues through agricultural commodities and products are known to be toxic and carcinogenic (Zawiyat *et al*., 2007). These harms may occur as a result of the residues of the pesticide exceeding maximum residue level (MRL) standards that seeks to protect human health (IPC, 2012). Fruits and vegetables are mainly consumed raw or semi-processed compared to food groups such as bread and others and so are required to satisfy pesticide residue levels (World Health Organization, 2003 and Claeys *et al.*, 2011).These global standards impose the burden on locally produced foods and drinks of plant origin including fruit juice to be wholesome and safe for consumption.

In recent times, national and international trade of fruit juice has rapidly increased in Ghana (Bempah *et al.,* 2011). These juices include mainly mango, orange, pineapple or a cocktail. Fresh fruit juice which requires only the juice extraction from the fresh fruits are easy to make and mostly regarded as healthy, since they contain no additives or colours. The worry however is the pesticide residues in these fruit beverages in localities like Ghana where the use of pesticide in fruit production is common (Bempah *et al.,* 2011; Gonu *et al*., 2012). There was therefore the apparent need for evaluation of agro-pesticide residues in locally produced fresh fruit juices on the Ghana market,

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particularly in the Greater Accra Region where most of these juices are concentrated and also to investigate the use of agro-pesticide in fruit crop production in the Eastern Region which is a fruit hub.

Materials and Methods

The study consisted of survey and laboratory/experimental work

Materials

Reagents used in the study comprised the following: Fruits juice, HPLC grade 99.98%, hexane, ethyl acetate and distol-Pesticide grade acetonitrile, which were purchased from Fisher Scientific UK. Sodium sulphate, Silica, 60-100 mesh from the CDH Limited and the individual standards used for the quantification and identification of the organochlorine pesticides were obtained from Cambridge Isotope Laboratories Incorporated, USA.

A Shimadzu gas chromatograph, GC-2010 equipped with ^{63}Ni Electron Capture Detector (ECD), CTC AOC-20s autosampler, AOC-20i split-splitless auto injector, Programmed Pneumatic Control (PPC) and a computer running star workstation data processor. For separation, a 5 % diphenyl 95 % dimethyl siloxane capillary column (30 m × 0.25 mm, 0.25 µm film thicknesses) was used.

Sample selection

Fruits juice samples for the investigation obtained from Madina market in the Greater Accra Region. Ghanaian produced fruit juices were sampled purposively from eight (8) different brands. These brands are noted to be the major competitors in the industry. Twenty (20) fruit juices were purchased from retailers randomly at the market center. Each brand was replicated thrice or twice, based on batch number, production and expiry dates, thus to ensure variations among samples. Products were relabeled and transported to the laboratory in a cool box.

Analysis of Organochlorine

EPA Method 8081A was used to determine the concentrations of various organochlorine pesticides in extracts from liquid matrices, using fused-silica, open-tubular and capillary columns with electron capture detectors (ECD). Below were the various kinds of organochlorine pesticides this method was used for; Aldrin, cis heptachlor, Dieldrin, Endrin, Heptachlor, Mirex, trans_nonachlor, alpha_HCH, beta_HCH, cis chlordane, delta_HCH, gamma_HCH_lindane, hexachlorobenzene-HCB and o_p_DDD.

Twenty (20 ml) of the fruit juice sample was measured into an extraction jar after which 20g of Na_2SO_4 was added and mixed to homogenize. Ten (10g) of $NaHCO_3$ was added and content mixed by constant shaking for 2 min. Forty (40 ml) neutral ethylacetate was added for 2 min with constant shaking.

Mixture was placed in a sonicator for 20 min after which the organic layer was decanted. The ethylacetate extraction and decantation were repeated twice per sample. The extraction was followed by clean up where by Florisil column (500 mg/8 ml) cartridge was conditioned with 10 ml of ethylacetate. Five (5 ml) of extract was loaded into the column and eluted with 30 ml (2:1 v/v) of ethyl acetate-hexane. The extract was concentrated to dryness using the rotary evaporator with water bath aided with the water chiller.

Residues were dissolved in 2 ml of ethyl acetate and then transferred quantitatively into 2 ml vial for quantification on to the gas chromatograph Varian CP-3800 GC. Two (2 µL) of each dissolved residue was used for the GC analysis of organochlorine present in the juice.

The pesticide residue concentration was deduced

$$\text{Pesticide residue} = \frac{\text{concentration in final extract x final volume of extract}}{\text{weight of sample}}$$

Survey

A survey was carried out among farmers in Nsawam and its environs (25 pineapple farmers) as well as, in and around Somanya (30 mango farmers) to solicit their knowledge in pesticide usage and types of pesticides used. These areas are noted for pineapple, orange and mango, which are the main raw materials for fruit juice production.

Results and Discussion

Survey Pesticide used by farmers in controlling insect pest

Pesticide or insecticides used by farmers in controlling insect pests on their fruit farm are presented in Table 1. Cypadem, cydem super, Cymethoate, Akatesroro, Sardi super and Pyrinex quick were pesticide used by mango farmers in controlling insect pest (Table 1). These pesticides were described as very effective (80%-90%) at eliminating insect pests by 53.3% of mango farmers and 46.7% described theirs as effective (60%-70%). Pineapple farmers applied Cymethoate, Cyadem super, Cypadem, Diuron, Thermex, Desban and Pawa. Sixty (60%) these farmers described pesticides as very effective whiles 40% described them as effective. These pesticides used by farmers were pyrethroids. The list reveals that farmers did not apply organochlorine based pesticide. Generally, farmers (98.2%) preferred applying individual pesticides but some respondents (1.8%) preferred cocktail of two or more pesticides with the perception of effectively controlling insect pest.

Organochlorine pesticide residue in fruit juice

Gas chromatography analysis of organochlorines in fruit juice (pineapple, fruit cocktail) revealed the presence of aldrin, cis heptachlor, dieldrin, endrin, heptachlor, mirex, trans_nonachlor, alpha_HCH, beta HCH, cis chlordane, delta_HCH, gamma_HCH, lindane, hexachlorobenzene_HCB, O_P_DDD, O_P_DDE, O_P_DDT, P_P_DDD, P_P_DDE, P_P_DDT, trans_chlordane and trans_heptachlor pesticide residues in the twenty one fruit juice sampled in the Greater Accra region, Ghana (Table 2). The residual levels of organochlorines in the fruit juice samples were insignificant. Heptachlor and mirex were the only residues above the EU's maximum residual levels (Table 2).

The survey revealed that farmers used only pyrethroid base insecticides, however organochlorine pesticide residues were disclosed in the fruit juice sampled. The identified metabolites, O_P_DDD, O_P_DDE, O_P_DDT, P_P_DDD, P_P_DDE, and P_P_DDT are indication of DDT degradation possibly from old usage before its ban (Canadian Council of Ministers of the Environment, 1999). This could be inferred from the trace amount of the metabolites recorded in the study (Table 2) and the lack of evidence of farmers' current usage (Table 1). However "total DDT" (sum of O_P_DDD, O_P_DDE, O_P_DDT, P_P_DDD, P_P_DDE, and P_P_DDT) were below European Union maximum residual level. Organochlorine pesticides are noted for having half-lives measured in years, or even decades leaving residues in water bodies, soil and in plant grown on residue retaining soils (Toan, 2013). These are banned agro-chemicals due to their carcinogenic property.

Table 1. Pesticides used by farmers in controlling insect pest

Pesticide trade name	Active ingredient
Cydem super	Cypermethrin and Dimethoate
Cypadem	Cypermethrin and Dimethoate
Cymethoate	Dimethoate
Pyrinex quick	Chlorpyrifos and Deltamethrine
Attack	Permethrin and Pirimiphos-methyl
Akatesroro	
Bendazene (fungicide)	
Karate	Lambda cyahalothrin
Diuron	Diuron
Sardi super	
Thermex	
Pawa	Lambda cyahalothrin
Dursban 20EC	Chlorpyrifos

Table 2. Pesticide residue in fruit juice sample

Pesticide residue	A	B	C	D	E	F	G	H	Average	EU MRLs
					mg/kg					
Aldrin	0.003998	0.003195	0.005885	0.003045	0.004364	0.012237	0.014571	0.005413	0.007	0.01
cis heptachlor	0.000001	0.000001	0.000001	0.000001	0.000001	0.000001	0.000001	0.000001	0.000001	0.01
Dieldrin	0.000001	0.001056	0.000001	0.000001	0.000001	0.008857	0.000001	0.000001	0.001	0.01
Endrin	0.000001	0.001548	0.000627	0.000001	0.000001	0.000787	0.000001	0.000613	0.0004	0.01
Heptachlor	0.0156	0.0272	0.027	0.0071	0.1524	0.1116	0.1498	0.2522	0.09	0.01
Mirex	0.18481	0.10714	0.18684	0.06744	0.09547	0.14507	0.17395	0.18223	0.14	0.01
trans_nonachlor	0.010639	0.002226	0.001846	0.000001	0.000001	0.005308	0.000001	0.000001	0.003	0.01
alpha_HCH	0.001472	0.001109	0.003923	0.000001	0.001133	0.002567	0.000958	0.001854	0.002	0.01
beta_HCH	0.000001	0.000001	0.000001	0.000001	0.000001	0.000001	0.000001	0.000001	0.000001	0.01
cis chlordane	0.000001	0.000001	0.000001	0.000001	0.000001	0.000001	0.000001	0.000001	0.000001	0.01
delta_HCH	0.00225	0.00206	0.00444	0.00198	0.00358	0.01131	0.02545	0.00254	0.007	0.01
gamma_HCH (lindane)	0.000229	0.002332	0.000856	0.000001	0.000637	0.001179	0.018436	0.000001	0.003	0.01
hexachlorobenzene-HCB	0.000001	0.000001	0.000797	0.000001	0.001278	0.004136	0.000001	0.000001	0.0008	0.01
o_p_DDD	0.000001	0.001724	0.000001	0.000001	0.00017	0.000001	0.000001	0.000001	0.00024	0.05
o_p_DDE	0.000001	0.000001	0.000001	0.000001	0.000001	0.000001	0.000001	0.000001	0.000001	0.05
o_p_DDT	0.001138	0.002061	0.000001	0.000001	0.00004	0.002427	0.001556	0.000001	0.0009	0.05
p_p_DDD	0.000001	0.000438	0.004795	0.00121	0.008761	0.020428	0.000001	0.003159	0.005	0.05
p_p_DDE	0.000001	0.002088	0.000575	0.000001	0.000001	0.001938	0.000001	0.000001	0.0006	0.05
p_p_DDT	0.009263	0.005094	0.008781	0.002033	0.00446	0.014049	0.006284	0.00691	0.007	0.05
trans_chordane	0.000202	0.000837	0.000001	0.00005	0.000001	0.002206	0.000001	0.000001	0.0004	0.01
trans heptachlor	0.000473	0.000384	0.000001	0.000001	1.1E-06	0.001469	0.000001	0.000001	0.0003	0.01

Table 3. Comparison of pesticide residues with that of EU MRLs

Pesticide residue	Minimum	Maximum	Grand mean±std	EU MRLs	F. Pr
		mg/kg			
Aldrin	0.00305	0.01457	0.007 ± 0.004	0.01	0.547
cis heptachlor	1E-06	1.00E-06	0.000001 ± 0	0.01	0
Dieldrin	1E-06	0.00886	0.001 ± 0.003	0.01	0.272
Endrin	1E-06	0.00155	0.0004 ± 0.001	0.01	0.824
Heptachlor	0.0071	0.2522	0.09 ± 0.088	0.01	0.388
Mirex	0.06744	0.18684	0.14 ± 0.047	0.01	0.382
trans_nonachlor	1E-06	0.01064	0.003 ± 0.004	0.01	0.716
alpha_HCH	1E-06	0.00392	0.002 ± 0.001	0.01	0.157
beta_HCH	1E-06	1.00E-06	0.000001 ± 0	0.01	0
cis chlordane	1E-06	1.00E-06	0.000001±0	0.01	0
delta_HCH	0.00198	0.02545	0.007 ± 0.008	0.01	0.397
gamma_HCH (lindane)	1E-06	0.01844	0.003 ± 0.006	0.01	0.298
Hexachlorobenzene (HCB)	1E-06	0.00414	0.0008 ± 0.001	0.01	0.254
o_p_DDD	1E-06	0.00172	0.00024 ± 0.001	0.05	0.662
o_p_DDE	1E-06	1.00E-06	0.000001 ± 0	0.05	0
o_p_DDT	1E-06	0.00243	0.0009 ± 0.001	0.05	0.716
p_p_DDD	1E-06	0.02043	0.005 ± 0.007	0.05	0.113
p_p_DDE	1E-06	0.00209	0.0006 ± 0.001	0.05	0.546
p_p_DDT	0.00203	0.01405	0.007 ± 0.004	0.05	0.358
trans_chordane	1E-06	0.00221	0.0004 ± 0.001	0.01	0.424
trans heptachlor	1E-06	0.00147	0.0003 ± 0.001	0.01	0.462

Findings of the study were similar to earlier work of Bempah *et al.,* (2011), who reported the presence of P_P_DDT, P_P_DDE, gamma_HCH, endrin, aldrin, and heptachlor in fruit juice sold in some supermarkets in Ghana. The degradation and low amount of DDT might have resulted from solar radiation (Bempah and Donkor, 2010) and probably the processing of the fruit juice.

Isomers of hexachlorocyclohexane (HCH) identified in the sampled fruit juice included alpha HCH, beta HCH, delta HCH and gamma HCH were below EU's maximum residual levels (0.01 mg/kg) which contradicts earlier reports by Bempah *et al*., (2011) who observed γ-HCH, pp-DDT, pp-DDE, endrin, endrin ketone, endrin aldehyde, heptachlor, aldrin, α-endosulfan and γ-chlordane residues in locally produced fruit juice above EU's maximum residual level.

The present study elucidates the adherence of farmers to the ban of organochlorine pesticide usage or rather the absence of it on the market. The observed organochlorine residues in the fruit juice nonetheless could be attributed to orgnochlorine long term ability to persist in the environment. Their identifications is a confirmation of their presence in the food chain (Bempah *et al*., 2011), though in insignificant trace amounts, but daily intake over a long period could pose health risks.

The higher heptachlor and mirex residue levels raise the red flags against the used fruit juices. In addition producers of raw materials for these fruit juices were thought to be engaged in the use of heptachlor and mirex pesticides or in recent past. Much campaign has been carried out on the banned of DDT or HCH however heptachlor and mirex might have less campaign on its usage.

Conclusion

Pesticides or insecticides used by farmers in cultivating mangoes and pineapples in Ghana were pyrethroid base. Farmers were aware of the consequences of organochlorines residues in the farm produce. Organochlorine pesticide residues were found in all the twenty (20) fruit juice sampled from the markets in the Greater Accra region, Ghana, though they are far below the EUs MRLs. The significant levels of heptachlor and mirex mall the wholesomeness of fruit juices.

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