

Full Length Research Paper

# Assessment of contamination by organochlorine pesticides in *Solanum lycopersicum* L. and *Capsicum annum* L.: A market survey in Nigeria

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This study investigated the presence and levels of organochlorine pesticides (OCPs) (hexachlorobenzene (HCB),  $\alpha$ -,  $\beta$ -,  $\delta$ -, and  $\gamma$ -hexachlorohexane (HCH), dieldrin, dichloro-diphenyl-trichloroethane (p,p'-DDT), dichloro-diphenyl-dichloroethylene (p,p'-DDE), dichloro-diphenyl-dichloroethane (p,p'-DDD) and trans-nonachlor) in *Solanum lycopersicum* and *Capsicum annum* samples commercially sold in Nigeria. OCPs were detected in all the samples analysed using Hewlett-Packard (HP 6890) gas chromatograph equipped with electron capture detector (GC-ECD). In *S. lycopersicum*, the total residue concentrations of  $\Sigma$ HCH (0.140 mg/kg),  $\Sigma$ HCB (0.144 mg/kg),  $\Sigma$ DDT (0.572 mg/kg),  $\Sigma$ dieldrin (0.073 mg/kg), and  $\Sigma$ trans-nonachlor (0.117 mg/kg) were found. In *C. annum*, the total residue levels of HCH, HCB, DDT, dieldrin and trans-nonachlor measured were 7.220, 0.096, 0.275, 0.037 and 0.117 mg/kg respectively. The  $\alpha$ -HCH isomer was not detected in *S. lycopersicum* and p,p'-DDD found only in *C. annum* samples. The OCPs levels were generally below the FAOs maximum residue limits except for HCH and trans-nonachlor. A pair-wise linear correlations among concentrations of OCPs in *S. lycopersicum* samples indicated statistically significant ( $P = 0.05$ ) relations between  $\delta$ -HCH and dieldrin ( $r = -0.997$ ,  $P = 0.047$ ), dieldrin and HCB ( $r = 0.99$ ,  $P = 0.043$ ), p,p'-DDD and p,p'-DDT ( $r = 1.00$ ,  $P = 0.006$ ).

**Key words:** Organochlorine pesticides, *Solanum lycopersicum*, *Capsicum annum*, vegetables, gas chromatography.

## INTRODUCTION

The contamination of the environment and food by chlorinated organic pesticides has become a topical issue of considerable concern in many parts of the world, and has led many researchers to investigate their occurrence, distribution and concentrations in commercially sold fruits, vegetables, milk and water (Yu et al., 2000; Soliman, 2001; How-Ran et al., 2006; Bai et al., 2006; Fontcuberta et al., 2008). Organochlorine pesticides have been used extensively worldwide since the early 1950s (Kin et al., 2006; Kuet and Seng, 2004) until restrictions were introduced in several developed and developing

countries due to their persistence in the environment and growing evidence of adverse associated health implications. In some developing countries like Nigeria, the use of pesticides in agriculture and public health as a quick and tailor-made panacea to numerous pest challenges continue despite well-known threats to human health and the environment. Literature on organochlorine pesticide residues in fruits, vegetables and foodstuffs from Nigerian markets are sparse (Atuma, 1985; Adeyeye and Osibanjo, 1999; Unyimandu and Duchy, 2002; Anyakora et al., 2008; Musa et al., 2010). Organochlorine pesticides are known to exhibit high lipophilicity and this is traceable to their environmental biopersistence and food chain contamination (Botella et al., 2004). The occurrence of organochlorine pesticides in food has been documented (Dong and Lee, 1999; Kuet and Seng, 2004; 438 Afr. J. Environ. Sci. Technol.

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Fontcuberta et al., 2008). The toxicity of OCPs pesticides can be acute and chronic. The chronic effects arising from exposure to contaminated food are mostly unknown but there is growing evidence of cancer, neurological damage, endocrine disruption and birth defects consequential from exposure (Miller and Sharpe, 1998; IARC, 2001; ATSDR, 2005a).

In view of compelling evidence of health effects on humans based on studies especially in developed countries and weak implementation of government policy on regulation / ban or surveillance program for pesticides levels in foods in Nigeria, there is need for evaluation of concentrations of organochlorine pesticides in commercially sold food crops. *Solanum lycopersicum* L. (tomato) is a glossy red, or occasionally yellow, pulpy edible fruit that is typically eaten as a vegetable or in salad, or stew, while *Capsicum annuum* L. (chilli pepper) is a small hot-tasting pod of a variety of capsicum, used chopped (and often dried) in sauces, relishes, and spice powders. These fruits are commonly grown, commercially available and widely consumed in Nigeria especially in local delicacies. This research assesses the levels of organochlorine compounds in these fruits and attempts to evaluate whether the permissible residue levels in them are exceeded. The investigation identified two research issues: (1) assess the levels of organochlorine pesticides in *S. lycopersicum* L. and *C. annuum* L.; (2) establish the strength of relationship between individual pesticides analysed.

## MATERIALS AND METHODS

### Materials and reagents

Fifty (50) fresh samples each of *S. lycopersicum* L. and *C. annuum* L. were purchased arbitrarily from different retailers at a local market in Ota, Western Nigeria, and organochlorine pesticides were quantified by gas chromatography equipped with electron capture detector (GC-ECD). In order to have a fair representative of samples bought, *S. lycopersicum* L. and *C. annuum* L. were randomly selected and carefully grouped in units of ten (10) prior chemical extraction and analysis. These groupings were designated TOT 1 to 5, and TAT 1 to 5 for samples of *S. lycopersicum* L. and *C. annuum* L. respectively.

All solvents used were of analar grade and all the pesticide standards were > 95% pure. Stock solutions of each pesticide at different concentration level 1.0 to 400.0 mg/L were prepared in methanol and stored at 4°C. Preparation of different concentration levels of stock solution is due to their sensitivity to the ECD detector. Working standard solutions (0.5 to 2.5 mg/L) of a mixture of pesticides were freshly prepared before each analysis by volume dilution in distilled water, and an internal standard 1-chloro-4-fluorobenzene (2 mg/L) was added to the vial prior to GC analysis.

### Analyte residue analysed

The contaminants examined were hexachlorobenzene (HCB),  $\alpha$ -,  $\beta$ -  $\delta$ - and  $\gamma$ -hexachlorohexane (HCH), dieldrin, dichloro-diphenyl-trichloroethane (p,p'-DDT), dichloro-diphenyl-dichloroethylene (p,p'-DDE), dichloro-diphenyl-dichloroethane (DDD) and trans-nonachlor.

## Sample extraction and analysis

Ten grams of each sample was homogenized in a mortar with 10 g sodium sulfate ( $\text{Na}_2\text{SO}_4$ ). The homogenate was transferred to an extraction column and each sample was extracted twice with 40 ml of dichloromethane ( $\text{CH}_2\text{Cl}_2$ ). The extract was filtered through 0.45  $\mu\text{m}$  membrane filter paper to remove traces of water. The volume was reduced to 2 ml with a rotary evaporator. Adsorption chromatography was performed on the sample using activated silica gel to remove the crude hydrocarbons. The silica gel was deactivated with 3.0% water (w/w). A 3 g portion of the deactivated silica gel was packed into a glass column (1.0 cm id), covered with  $\text{Na}_2\text{SO}_4$  and eluted with 60 ml hexane. The eluent was concentrated by rotary evaporation to 0.5 ml. Aliquots were analysed by GC-ECD (Wang, 2002; Kin et al., 2006). The reference standards of organochlorine pesticides; hexachlorobenzene (HCB),  $\alpha$ -,  $\beta$ - and  $\delta$ -hexachlorohexane (HCH), dieldrin, dichloro-diphenyl-trichloroethane (p,p'-DDT), dichloro-diphenyl-dichloroethylene (p,p'-DDE), dichloro-diphenyl-dichloroethane (DDD) and trans-nonachlor of 1.0 mg/L were prepared and extracted with dichloromethane. The collecting organic layers were dried, evaporated and injected to GC-ECD (Hewlett-Packard, HP 6890) for the investigation of suitable extracting solvent. The reference standard mixture of eight OCPs was prepared in the range of 0.5 to 2.5 mg/L and mixed with 2.0 mg/L of pentachloronitrobenzene internal standard. The mixtures were then extracted and cleaned up at the suitable condition obtained prior injecting to GC-ECD for the calibration curves.

## Gas chromatography analysis

The analysis of organochlorine pesticide residues was performed using Hewlett-Packard (HP 6890) gas chromatograph with an electron capture detector (ECD). A capillary column 30 m x 0.32 mm id with a 0.25  $\mu\text{m}$  film thickness was used with ultrapure nitrogen gas (99.99%) as the carrier gas, with a gas flow rate at 40 cm/s and the pressure at 94 kPa. The column was programmed from an initial oven temperature of 150°C and later raised at 4°C/m to 300°C, and then held for 10 m, with a total run time of 24 m. The splitless mode was used for the sample injection. The injector and detector temperature were 200 and 300°C respectively. Sample volumes of 2  $\mu\text{l}$  were injected.

## Statistical analysis and validation of method

Summary descriptive of data was performed using Statgraphics Centurion VI Software<sup>®</sup> with level of significance maintained at 95% for each test for nonparametric and parametric correlations. The percentage (%) recoveries together with limit of detection (LOD) and limit of quantitation (LOQ) were also calculated. The concentration of each OC pesticides was calculated by comparing the chromatographic peak area from the test sample to those of known amounts of the standards.

## RESULTS AND DISCUSSION

The linearity ( $r^2$ ), % recoveries, limit of detection and limit of quantitation of the obtained method are shown in Table 1. The mean concentrations of organochlorine pesticides (hexachlorobenzene (HCB),  $\alpha$ -,  $\beta$ - and  $\delta$ -hexachlorohexane (HCH), dieldrin, dichloro-diphenyl-trichloroethane (p,p'-DDT), dichlorodiphenyldichloroethyl ene (p,p'-DDE), dichlorodiphenyldichloroethane (DDD), and

**Table 1.** Linearity, percentage recoveries, limit of detection (LOD) and limit of quantitation (LOQ).

Organochlorine pesticides	Linearity ( $r^2$ )	Recoveries (%)	LOD (mg/Kg)	LOQ (mg/Kg)
HCB	0.9894	96.40	0.063	0.24
$\alpha$ -HCH	0.9956	98.16	0.000	0.00
$\beta$ -HCH	0.9987	96.03	0.063	0.24
$\delta$ -HCH	0.9995	97.07	0.060	0.20
Dieldrin	0.9989	98.23	0.126	0.42
p,p'-DDE	0.9890	99.12	0.303	1.01
p,p'-DDD	0.9893	98.86	0.258	0.86
p,p'-DDT	0.9969	99.32	0.030	0.10
trans-nonachlor	0.9949	99.08	0.102	0.34

trans-nonachlor) residues found in *S. lycopersicum* (TOT 1 to 5) and *C. annuum* (TAT 1–5) samples analysed are presented in Table 2. A Pearson product moment correlation between each pair of chlorinated organic pesticide residues is shown in Table 3. These correlation coefficients range between -1 and +1 and measure the strength of the linear relationship between the variables.

Also shown in parentheses are p-values, which test the statistical significance of the estimated correlations. P-values below 0.05 indicate statistically significant non-zero correlations at the 95.0% confidence level. A pairwise linear correlations among levels of organochlorine pesticides in *S. lycopersicum* samples indicated negative but statistically significant ( $p = 0.05$ ) relations between  $\delta$ -HCH and dieldrin ( $r = -0.997$ ,  $p = 0.047$ ). A correlation existed between dieldrin and HCB, p,p'-DDD and p,p'-DDT with  $r$  statistics recorded as 0.99 ( $p = 0.043$ ) and 1.00 ( $p = 0.006$ ) respectively (Table 3). This indicated that a positive and statistically perfect and significant relationship existed between these pesticides in *S. lycopersicum*. In general, positive but insignificant correlations were found between  $\beta$ -HCH and dieldrin ( $r = 0.699$ ),  $\beta$ -HCH and HCB ( $r = 0.746$ ),  $\beta$ -HCH and p,p'-DDE (0.269),  $\beta$ -HCH and trans-nonachlor ( $r = 0.895$ ),  $\delta$ -HCH and p,p'-DDE ( $r = 0.562$ ), dieldrin and HCB ( $r = 0.998$ ), dieldrin and p,p'-DDD ( $r = 0.005$ ), dieldrin and p,p'-DDT ( $r = 0.014$ ), dieldrin and trans-nonachlor ( $r = 0.306$ ), HCB and trans-nonachlor ( $r = 0.369$ ), p,p'-DDD and p,p'-DDT ( $r = 1.000$ ), p,p'-DDE and trans-nonachlor ( $r = 0.691$ ).

However, negative and statistically insignificant relations existed between  $\beta$ -HCH and  $\delta$ -HCH ( $r = -0.645$ ),  $\beta$ -HCH and p,p'-DDD ( $r = -0.711$ ),  $\beta$ -HCH and p,p'-DDT ( $r = -0.705$ ),  $\delta$ -HCH and dieldrin ( $r = -0.997$ ),  $\delta$ -HCH and HCB ( $r = -0.990$ ),  $\delta$ -HCH and p,p'-DDD ( $r = -0.078$ ),  $\delta$ -HCH and p,p'-DDT ( $r = -0.088$ ),  $\delta$ -HCH and trans-nonachlor ( $r = -0.236$ ), dieldrin and p,p'-DDE ( $r = -0.500$ ), HCB and p,p'-DDD ( $r = -0.062$ ), HCB and p,p'-DDE ( $r = -0.441$ ), HCB and p,p'-DDT ( $r = -0.053$ ), p,p'-DDD and p,p'-DDE ( $r = -0.868$ ), p,p'-DDD and trans-nonachlor ( $r = -0.951$ ), p,p'-DDE and p,p'-DDT ( $r = -0.873$ ), p,p'-DDT and trans-nonachlor ( $r = -0.947$ ).

The levels of Hexachlorobenzene (HCB) was generally low in *S. lycopersicum* (TOT 1-5) and *C. annuum* (TAT 1-5) with mean concentrations of  $3.74 \times 10^{-2}$ ,  $7.21 \times 10^{-2}$ ,  $3.46 \times 10^{-2}$ ,  $4.21 \times 10^{-2}$  and  $5.40 \times 10^{-2}$  mg/kg respectively (Table 2). HCB is a fungicide that has been banned globally under the Stockholm Convention on persistent organic pollutants (ACGIH, 1991; UNEP, 2004). Hexachlorobenzene is a known animal carcinogen causing increased incidences of liver, kidney (renal tubular tumours) and thyroid cancers (IARC, 2001). Chronic oral exposure in humans has been shown to give rise to a liver disease (porphyria cutanea tarda), skin lesions with discoloration, ulceration, photosensitivity, thyroid effects, bone effects and loss of hair. Neurological changes have been reported in rodents exposed to hexachlorobenzene. HCB may cause embryoletality and teratogenic effects. Human and animal studies have demonstrated that HCB is capable of crossing the placenta to accumulate in foetal tissues, which could be subsequently transferred to breast milk.

The mean concentration of hexa-chlorocyclohexane (HCH) as shown in Table 2 ranged from  $3.39 \times 10^{-2}$  to  $6.68 \times 10^{-2}$  mg/kg in *S. lycopersicum* samples while *C. annuum* samples recorded relatively higher mean concentrations of 3.14 and  $4.08 \times 10^{-2}$  mg/kg for TAT 1 and TAT 2 respectively. The detection of lindane and other HCH isomers in these vegetables is not a surprise since four of the HCH isomers ( $\alpha$ -,  $\beta$ -,  $\gamma$ -, and  $\delta$ ) are commonly in use in developing countries. Although lindane is the only isomer with pesticidal properties, they are known to be persistent, bioaccumulative, toxic, and mobile in the environment including the environmentally significant alpha- and beta-HCH isomers. Lindane, the gamma isomer, is produced and used commercially as an insecticide for decades. It is used in agriculture on fruit, vegetables, ornamental plants and as a soil and seed treatment to protect seeds and seedlings (ATSDR, 2005a). In this study,  $\alpha$ -HCH was not detected in any of the analysed samples. However, relatively low concentrations of  $\beta$ -HCH and  $\delta$ -HCH were found in *S. lycopersicum* samples. *C. annuum* samples recorded relatively higher  $\beta$ -HCH mean concentrations of 1.69 and

**Table 2.** Summary statistics for organochlorine pesticides concentrations (mg/kg) in *S. lycopersicum* and *C. annuum*.

Pesticide	Analyte	<i>S. lycopersicum</i>					Maximum limit (mg/kg)*	<i>C. annuum</i>				
		Min.	Max.	Range	mean±S.D.	C.V.%		Min.	Max.	Range	mean±S.D.	C.V.%
hexachlorobenzene	HCB	0.035	0.072	0.038	0.048±0.021	43.49	1.0	0.042	0.054	0.012	0.048±0.008	17.51
α-hexachlorobenzene	α-HCH	BDL	BDL	BDL	BDL			BDL	BDL	BDL	BDL	
β-hexachlorobenzene	β-HCH	0.0	0.040	0.040	0.023±0.021	89.41	ΣHCH-	1.690	2.340	0.650	2.015±0.459	22.81
δ-hexachlorohexane	δ-HCH	0.0	0.037	0.037	0.024±0.020	86.84		1.450	1.740	0.290	1.595±0.205	12.86
γ-hexachlorohexane	γ-HCH	BDL	BDL	BDL	BDL			BDL	BDL	BDL	BDL	
	dieldrin	0.0	0.073	0.073	0.024±0.023	95.83	0.1	0.0	0.037	0.037	0.019±0.003	16.04
dichloro-diphenyl-dichloroethylene	p,p'-DDE	0.0	0.175	0.175	0.058±0.011	18.97		0.007	0.105	0.098	0.056±0.007	12.32
dichloro-diphenyl-dichloroethane	p,p'-DDD	0.0	0.172	0.172	0.086±0.086	99.73	ΣDDT 2	BDL	BDL	BDL	BDL	
dichloro-diphenyl-trichloroethane	p,p'-DDT	0.036	0.056	0.020	0.046±0.010	21.83		0.064	0.099	0.036	0.081±0.025	31.17
	Trans-nonachlor	0.0	0.066	0.066	0.039±0.034	88.59	0.1	0.029	0.088	0.058	0.059±0.041	69.81

\*FAOs - Maximum Residue limits, S.D. – Standard deviation; C.V. – Coefficient of variation; Σ = Total; BDL – Below Detectable Limit.

2.34 mg/kg for TAT 1 and TAT 2 respectively, while δ-HCH high average concentrations of 1.45 and 1.74 mg/kg were obtained from TAT 1 and TAT 2 samples respectively. The International Agency for Research on Cancer (IARC) has classified HCH (all isomers) as possibly carcinogenic to humans. Long-term exposure to α-HCH, β-HCH, γ-HCH, or technical-grade HCH has been reported to result in liver cancer. It can also result in blood disorders, dizziness, headaches, and possible changes in the levels of sex hormones in the blood (IARC, 2001; ATSDR, 2005b). The concentrations of dieldrin in *S. lycopersicum* sample (TOT 2) analysed were generally low and was not detected in TOT 1 and

3 samples. It was equally not detected in TAT 1 samples of *C. annuum* but found in low level in TAT 2 with average concentration of  $3.73 \times 10^{-2}$  mg/kg respectively (Table 2). Dieldrin is an organochlorine insecticide highly persistent in the environment. It tends to biomagnify as it is passed along the food chain. Long-term exposure has proven toxic to a very wide range of animals including humans. Toxicity to humans, include carcinogenicity, reproductive and developmental toxicity, neurotoxicity, and acute toxicity. It has been linked to health problems such as Parkinson's, breast cancer, and immune, reproductive, and nervous system damage. It can also adversely affect testicular descent in the fetus if a

pregnant woman is exposed to dieldrin (ATSDR, 2002). Dichloro-Diphenyl-Trichloroethane (DDT) is one of the best known synthetic pesticides. The major metabolites and breakdown products of DDT in the environment are dichlorodiphenyldichloroethylene (DDE), which is produced by the dehydrohalogenation of DDT, and dichlorodiphenyldichloroethane (DDD). Total residues of DDT mean concentrations as shown in Table 2 ranged from  $1.329 \times 10^{-1}$  (TOT 2) to  $2.28 \times 10^{-1}$  mg/kg in *S. lycopersicum* samples while *C. annuum* samples recorded relatively low concentrations of  $1.685 \times 10^{-1}$  and  $1.069 \times 10^{-1}$  mg/kg for TAT 1 and TAT 5 respectively. The concentrations of trans-nonachlor were variable.

**Table 3.** Correlation matrix for organochlorine pesticides concentrations in *S. lycopersicum*.

	$\beta$ - HCH	$\delta$ - HCH	dieldrin	HCB	p, p'-DDD	p, p'-DDE	p, p'-DDT	Trans-nonachlor
$\beta$ – HCH		-0.645 (0.553)	0.699 (0.507)	0.746 (0.464)	-0.711 (0.496)	0.269 (0.827)	-0.705 (0.502)	0.895 (0.295)
$\delta$ -HCH	-0.645 (0.553)		-0.997 (0.047)	-0.990 (0.089)	-0.078 (0.950)	0.562 (0.620)	-0.088 (0.944)	-0.236 (0.849)
Dieldrin	0.699 (0.507)	-0.997 (0.047)		0.998 (0.043)	0.005 (0.997)	-0.500 (0.667)	0.014 (0.991)	0.306 (0.802)
HCB	0.746 (0.464)	-0.990 (0.089)	0.998 (0.043)		-0.062 (0.960)	-0.441 (0.709)	-0.053 (0.966)	0.369 (0.759)
p,p'-DDD	-0.711 (0.496)	-0.078 (0.950)	0.005 (0.997)	-0.062 (0.960)		-0.868 (0.330)	1.000 (0.006)	-0.951 (0.201)
p,p'-DDE	0.269 (0.827)	0.562 (0.620)	-0.500 (0.667)	-0.441 (0.709)	-0.868 (0.330)		-0.873 (0.324)	0.671 (0.532)
p,p'-DDT	-0.705 (0.502)	-0.088 (0.944)	0.014 (0.991)	-0.053 (0.966)	1.000 (0.006)	-0.873 (0.324)		-0.947 (0.207)
Trans-nonachlor	0.895 (0.295)	-0.236 (0.849)	0.306 (0.802)	0.369 (0.759)	-0.951 (0.201)	0.671 (0.532)	-0.947 (0.207)	

Correlation ( ) P-Value.

*S. lycopersicum* samples recorded  $6.55 \times 10^{-2}$  and  $5.10 \times 10^{-2}$  mg/kg in TOT 1 and TOT 2 samples respectively.

However, this organochlorine hydrocarbon was not detected in TOT 3 samples. On the other hand, *C. annuum* samples recorded relatively higher concentrations of  $8.76 \times 10^{-2}$ ,  $6.48 \times 10^{-2}$  and  $2.97 \times 10^{-2}$  mg/kg for TAT 1, 2 and 4 respectively. Trans-nonachlor is the most bioaccumulative major constituents of the insecticide chlordane, a cyclodiene that was used extensively in home and agricultural applications. Like DDT, chlordane compounds are very persistent in the environment, resistant to metabolism, have a strong affinity for lipid, and biomagnify in aquatic food webs. It is obvious from the results that some organochlorine pesticide residues, specifically Dieldrin, DDT,  $\gamma$ -HCH,  $\alpha$ -HCH and  $\beta$ -HCH were found to be present in *S. lycopersicum* L. and *C. annuum* L. analysed. Generally, the concentrations of organochlorine pesticides in analysed *S. lycopersicum* and *C. annuum* were low.

However, these low levels of organochlorine pesticides are significant and cannot be neglected taking cognizance of the perceptible health risks associated with the consumption of tomato and chilli pepper. In Nigeria, farmers are known to use pesticides of organohalogenic families for pest control and foodstuff preservation imported into the country through unauthorized and

fraudulent routes. However, their use has been scaled down due to the ban on the use of most of the organochlorine pesticides in response to the Stockholm Convention on the use of Persistent Organic Pollutants (POPs).

## Conclusions

Despite considerable increased pesticide use over the past decades, little research has been done to determine their amount, fate and the toxicity or hazard of the pesticide and the likelihood of exposure via foodstuffs in Nigeria. This effort presents an attempt to address the concern pesticide residues might pose in Nigeria. Although the pesticide residue levels in *S. lycopersicum* L. and *C. annuum* L. are lower than FAO maximum residue limits (MRLs), the presence and eventual biomagnification of these toxic organohalogens in these vegetables raises health concerns considering that both foodstuffs are widely consumed in Nigeria.

More so, the presence of these toxicants in commercially available fruits underlines the need for routine analysis and assessment of pesticides in market foods in order to reduce their presence, or completely discourage their usage in agriculture. These determinations are

important in monitoring pesticides burden in consumable fruits and crops for the prevention, control and reduction of pollution as well as for occupational health and epidemiological studies.

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