INTERNATIONAL JOURNAL
OF
PHARMACEUTICAL SCIENCES
AND
RESEARCH

ENUMERATION OF ORGANCHLORINE, ORGANOPHOSPHATE AND SYNTHETIC PYRETHROIDS IN BEANS GROWN IN FIVE DISTRICTS OF KARNATAKA

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ABSTRACT: Pesticide residues in food and crops are a direct result of application of pesticides to crops growing in the field. Large quantities of pesticides are being used to control various pests, which normally infest the economically important plants. Beans- grown across the country and provides an important source of income for the small and marginal farmers. Bean growing in five districts of Karnataka *viz.*, Bangalore urban, Bangalore rural, Ramanagara, Tumkur, Chikkaballapura and Kolar are the prime areas in the production of vegetables identified for the detection of pesticide residues. Bean samples spiked at 0.01, 0.5 and 1.0mg/kg and average recovery rate ranged from 77.0 to 95.4% & the average RSDs from 1.2% to 6.1%. Presence of Cyfluthrin- β, cyhalothrin-λ, cypermethrin, deltamethrin and fenvalerate Acephate, chlorphyriphos, dichlorvos, monocrotophos showed values Below Detection Level (BDL). 5% 0f the samples collected from the five districts showed mean phorate concentration (0.035 mg/kg) exceeding the MRL value of 0.05mg/kg in Bangalore urban district.

INTRODUCTION: Population explosion has resulted in tremendous pressure on the increased need of agricultural productivity. Ever increasing demand for food resulted in using chemical pesticide across the world to improve the quality and yield and to extend the storage life of food crops [1]. Reports indicated that 900 chemicals are used worldwide, legally and illegally, in various food products and for the treatment of crops and vegetables [2]. Applied chemicals and degradation product may remain as residues in the agricultural products, which is a concern for humans.

The residues are a matter of serious concern since increased use of chemical pesticides has resulted in the contamination of environment and has long term effects on human health and the problem is serious when these commodities are consumed [3, 4]. 85 % - 90 % of pesticides applied to agricultural crops never reaches the target organisms, but get dispersed through air, soil and water [5]. Pesticides are found to block the cellular processes of the target organisms, destroy or alter the pest metabolism or disrupt enzyme processes and affect nervous system.

Developing country like India consumes 600 g of pesticides per hectare, whereas that of developed countries is nearly 3000 g/hectare. Low consumption is attributed to fragmented land holdings, lower level of irrigation, dependence on monsoon and lack of using pesticides on grains [6].

Available reports indicated that Organochlorines, organophosphates and synthetic pyrethroids are commonly detected in the fruits and vegetables of both tropical and temperate countries. The objectives of the present study are to identify and assess the pesticide levels in the selected vegetables from the point view of safety to consumers, comparing the observed residue levels with PFA and codex standards, enumerate the most prevalent pesticides and making appropriate recommendations to facilitate extension workers to adopt safe pesticide usage in crop cultivation.

MATERIALS AND METHODS: The present study was conducted during the year 2011-2014 in five districts of Karnataka. Field study was conducted to collect the information about various aspects of pesticide use and their safety. This information was used as the baseline data to investigate the residue level of the analyzed pesticides.

Study area: Karnataka State is situated in the west central part of peninsular India, geographically located between 11^0 30' N to 18^0 30' N latitudes and 74^0 E to 78^0 30' E longitude. The state covers an area of 191,976 square kilometers (74,122 sq m) or 5.83% of the total geographical area of India. Five districts of Karnataka namely, Bangalore rural, Bangalore urban, Chikkaballapura, Kolar and Ramanagara (**Fig. 1**) are presenting the hub of agricultural activities were selected as study areas for the determination of pesticide residues in vegetable samples. A sporadic information and data was collected from farmers with respect to use of different pesticides and harvesting period of vegetables and fruits in the study area. The sampling period covered the pre- and post- rainy seasons, coinciding with maximum harvest period of vegetable samples coming from different sources of agriculture in the region were procured randomly between 2009 and 2013.

Sampling procedure transportation and storage: Randomly selected vegetable sample beans collected from the growing areas and district headquarters of five districts (Including major markets of Bangalore urban). Samples of marketable size (1 kg each) were collected and transported to the laboratory in fresh plastic bags

and kept in refrigerator (5 °C) until analysis in order to avoid any degradation of residues between sampling and analysis. Only the edible parts of vegetables were processed for residue analysis. A total of twenty vegetables samples were collected from five different districts of Karnataka.

Questionnaire survey: Field study was carried out through questionnaire so as to collect the information on awareness level of farmers regarding pesticide use, type and amount of pesticide they were using, their knowledge regarding the safety measures to be undertaken during and after pesticide. Twenty five farmers were interviewed and on the basis of these survey results, different pesticides that they were used in their farm are recognized and were selected for the laboratory analysis of the collected samples to study their residue level. Some banned pesticides were also considered for analysis.

Standard solutions: Pesticide standard stock solutions were procured from Indian Agricultural Research Institute (IARI), New Delhi. Working standard solutions containing a mixture of the analyte were prepared from the stock by appropriate solvent dilutions in n-hexane. For standards preparation, the required amount of pesticide standards were mixed with required volume of n-hexane (HPLC grade). The stock solutions, 100 ppm of each pesticide were prepared, labeled and stored in airtight clean bottles. From this, a single mix standard of 100 ppm was prepared, which was diluted firstly to 10 ppm and then to 1ppm. The 1 ppm mix standard was used to make the calibration standards of 0.01, 0.5 and 1.0 ppm. In this way, a series of calibration standards ranging from 1.0 to 0.01 ppm was prepared. The single and mixed stock solutions were stored at -5 ^{0}C while, the calibration standards were made on the day of analysis. The calibration standards so prepared (1 μl) were injected to GC and analyzed.

Reagents: All solvents like n-hexane, acetonitrile, petroleum ether and diethyl ether (HPLC grade) were procured from Sigma Aldrich Co., and were glass distilled before use. AR grade sodium chloride (NaCl) and anhydrous sodium sulphate (Na_2SO_4) was procured from HIMEDIA Pvt. Ltd.,

India. Before use, anhydrous sodium sulphate (Na$_2$SO$_4$) was purified with acetone and heated for 4 hr at 400 ^{0}C in a muffle furnace to remove possible phthalate impurities. Florosil (60-100 mesh) purchased from Merck India limited was activated at 450 ^{0}C and reheated at 130 ^{0}C for 5hr before use.

Pesticide analysis: During the investigation, residues of insecticides in beans were monitored in 20 vegetable samples from five districts of Karnataka using Gas Chromatograph with ECD and FTD (Shimadzu make, Model GC-2010). Preparation of the samples and determination of insecticide residues was based on the method described [7]. Pesticide residue analysis may be defined as the qualitative and quantitative analysis of the represented samples procured from agriculture field and market for pesticides and their toxic metabolites. Pesticide residue analysis involves three steps: Extraction, Cleanup and Analysis. The samples of vegetables were extracted by following acetonitrile extraction/ petroleum ether partitioning and cleaned up by column chromatography using florisil adsorbent. Efficiency of the method was validated with recovery.

Extraction: The most suitable approach in the determination of the pesticide residue contents in food samples is sample preparation method (Extraction and Cleanup) with various chromatographic methods. Sample preparation is often a neglected area, which over the years has received much less attention and research than the chromatographic separation or detection stages. The most efficient approach to pesticide analysis involves the use of multiclass, multi-residue methods (MRMs).

The first notable MRM was the Mills method developed in the 1960s for the determination of non-polar organochlorine pesticides in non-fatty food. The Mills method was based on acetonitrile extraction, the extract was then diluted with water, and the pesticides were partitioned into a non-polar solvent. The follow-up research was oriented towards extending the analytical polarity range to cover wider range of polarity of pesticides analyzed in a single procedure. New solvents for initial extraction and addition of NaCl for the partitioning

step were tested to reach higher recoveries of the more polar analytes. In 80s environmental and health concerns led to the avoidance of dangerous solvents, later Solid-Phase Extraction (SPE) was established to avoid Liquid-Liquid Partitioning (LLP) and as a cleanup step.

Increased urgency to further reduce solvent usage and manual labor led to the introduction of several alternative extraction approaches Accelerated Solvent Extraction/Pressurized Liquid Extraction (ASE/PLE),Gel Permeation Chromatography (GPC), Matrix Solid-Phase Dispersion (MSPD), Microwave-Assisted Extraction (MAE), Solid-Phase Extraction (SPE) / Dispersive Solid-Phase Extraction (DSPE), Solid-Phase Micro Extraction (SPME), Stir-Bar Sorptive Extraction (SBSE), Supercritical Fluid Extraction (SFE). Quick, easy, cheap, effective, rugged and safe method (QuEChERS) which aimed to overcome critical deficiencies and practical limitations of existing methods [8].

In the present study, only the edible parts of vegetable samples (1.0 kg) were chopped and 50 g of samples were extracted in a warring blender with 100 ml acetonitrile for 2-3 min. The solvent was filtered through a Buchner funnel. The fruit residue was again subjected to extraction with 50 ml acetonitrile two more times. The extracts were evaporated under vacuum to about 5ml and then transferred to a separator funnel of capacity 1000 ml. 600 ml of 5 % sodium chloride was added and the extract was exchanged into petroleum ether layer by liquid-liquid partitioning thrice (100 ml, 2 × 50 ml). The extract was then passed through a layer of sodium sulfate (5 g) and evaporated to dryness in a rotary evaporator at a temperature below 40 °C.

Clean up: Glass column (60 cm length × 2.0c m I.D) was packed with a mixture of florisil (10 g), anhydrous sodium sulphate (10 g) and activated charcoal (0.2 g) supported on a cotton plug was used for cleanup and the sample was wetted with 50ml petroleum ether. Sample slurry prepared using petroleum ether was transferred to the column. The glass beaker containing extract was rinsed with acetone and was transferred to the column, which was allowed to stand for 45 min.

Subsequently, the petroleum ether present in the column was eluted drop-wise (5 ml/min). When about 5ml petroleum ether remained on the surface of the adsorbent, the extract was eluted with 200 ml each of freshly prepared 6 % solvent mixture (diethyl ether in petroleum ether), 15 % solvent mixture (diethyl ether in petroleum ether) and 50 % solvent mixture successively.

The eluents were concentrated to dryness in a rotary evaporator under vacuum and diluted to 10 ml with n-hexane for further analysis. From the dissolved residues, 1 µl was injected to gas chromatograph and peak areas were compared with those obtained from similar injections of standards.

Pesticide residue analysis: Pesticide residue analysis was performed on Gas Chromatograph GC-2010 (Shimadzu make) equipped with ECD (Electron Capture Detector) and FTD (Flame Thermionic Detector). A fused silica capillary column (BP5- 5 % Phenyl, 95 % Dimethyl polysiloxane) was used for the analysis. Insecticides like organochlorines (OCs) and pyrethroids (SPs) were analyzed using ECD (63Ni) and a capillary column BP-5 (60 m × 0.25 mm I.D. × 0.25 µm film thickness) with split ratio 1:10. Nitrogen flow rate of 30ml/min, injection port temperature of 250 ^{0}C and temperature of detector of 300 ^{0}C and an injection volume of 1µl were the Gas - Liquid Chromatography (GLC) working conditions maintained during the analyses. The column temperature was initially maintained at 80 ^{0}C for 5min and then slowly increased to 260 ^{0}C at the rate of 10 ^{0}C per min for 5min and finally increased to 290 ^{0}C for 5min.

In contrast, organophosphates (OPs) residues were analyzed with FTD and a split less capillary column DB-1 (10 m × 0.53 mm I.D. × 2.65 µm film thickness). The GLC working conditions maintained during the analyses were nitrogen flow rate of 60 ml/min, hydrogen flow rate of 3 ml/ min, air flow rate of 150 ml/min, injection port temperature of 280 ^{0}C, detector temperature of 300 ^{0}C and an injection volume of 1µl with split ratio of 1:10. The column temperature was initially maintained at 180 ^{0}C for 5 min and then gradually increased to 260 ^{0}C for 5 min.

Estimation / Quantification of residues: The carrier gas obtained from a steel gas cylinder passes through a flow regulator for the adjusted flow rate and enters into the sample injector. A little amount of the sample is introduced into the sample injector with the help of a hypodermic syringe. The sample injector is maintained at a temperature higher than the boiling point of the highest boiling component of sample in order to ensure rapid vaporization of the liquid samples. The carrier gas entering the sample injector sweeps off the vaporized sample and passes down the temperature programmed column. The components of the sample are distributed between the stationary and the mobile phases and pass down the column at different rates. This results in the separation of the components of the sample.

The carrier gas with the separated components enters the detector, which measure the change in composition of the carrier gas as it passes through it. This change is amplified before it is fed into a recorder which drives the recording pen on a moving strip of paper and a chromatogram is obtained. Currently rapid instrumental methods are available for data processing and obtaining chromatograms in computer compatible formats. The pesticide residue concentration was calculated using the equation,

$$\text{Residues value (µg/g)} = \frac{A_s \times V_{std} \times C_{std} \times D_f}{A_{std} \times V_s \times W_s}$$

Where
A_s = peak area of sample injected (mv),
A_{std} = peak area of standard injected (mv)
V_s = volume of sample injected (µl/ml),
V_{std} = volume of standard injected (µl/ml)
C_{std} = concentration of standard (µg/ml),
W_s = weight of sample taken (g)
D_f = dilution factor (ml)

Among the quantification of the targeted 20 residues, nine are organochlorines (*viz.,* aldrin, dieldrin, endosulfan-α, endosulfan-β, endosulfansulphate, HCH-α, HCH-β, HCH-γ, heptachlor), six are organophosphorus (i.e., acephate, chlorpyriphos, dichlorvos (DDVP), monocrotophos, phorate, profenophos) and five are synthetic pyrethroids (cyfluthrin-β, cyhalothrin-λ, cypermethrin, deltamethrin, fenvalerate).

By injecting 1µl of the standard solution or the cleaned up extract into the GC, retention times (RT) and peak areas of analytes were recorded. Residues were estimated by comparison of peak heights/peak areas of the standards with that of the unknown or spiked samples run under similar conditions. Efficiency of the method was validated with recovery.

Fortification / Recovery studies: The recovery studies for 3 replicates for each pesticide at three different fortification levels (1.0, 0.5 and 0.01 mg/kg) were carried out. For this purpose, vegetable samples were spiked with 1ml of desired concentration of pesticide. Resulting samples were mixed and allowed to stand for 30 min before extraction and then processed separately as per the methodology described above. The amount of pesticide residues in vegetable samples were calculated by measuring peak areas from extracted current profiles and comparing with those obtained from matrix-matched standards of a concentration similar to that of samples. Spiked samples were calculated in the same way as regular samples [9, 10].

Calculation of Percentage Recovery: The percentage recovery was calculated using the formula:

$$\% \text{ Recovery} = \frac{\text{Amount recovered}}{\text{Amount spiked}} \times 100$$

Maximum Residue Levels (MRLs): Maximum residue levels may be defined as the maximum levels of pesticide residue present in or on a produce when pesticide used under supervision following good agricultural practices (GAP). According to Environmental Protection Agency (EPA), it is the concentration of a pesticide residue that can remain in food and feed products, or commodities. It is also known as 'pesticide residue limits' or tolerances, which are set to protect human from harmful levels of pesticides in food. Food and Drug Administration (FDA) and the United States Department of Agriculture monitor the food in interstate commerce to ensure that these limits are not exceeded. MRL is primarily intended to be used in international trade. MRL is the maximum concentration of a pesticide residue (expressed as mg/kg), recommended by the Codex Alimentarious Commission (CAC) to be legally permitted in food commodities and animal feeds.

MRL are based on GAP data and foods derived from commodities that comply with the respective MRL are intended to be toxicologically acceptable.

Pesticide residues in beans: Variation in acephate, chlorpyriphos, dichlorvos, monocrotophos, phorate, cyfluthrin-β, cyhalothrin-λ, cypermethrin, deltamethrin and fenvalerate residues in beans samples are discussed below.

RESULTS AND DISCUSSION: Twenty different pesticide residues falling under organochlorines (OCs), organophosphorus (OPs) and synthetic pyrethroids (SPs) pesticide groups were analyzed in the beans. The vegetable samples were collected from five districts of Karnataka namely, Bangalore rural, Bangalore urban, Chikkaballapura, Kolar and Ramanagara. Among the pesticides residues analyzed, ten residues *viz*., acephate, chlorpyriphos, cyfluthrin-β, cyhalothrin-λ, cypermethrin, deltamethrin, dichlorvos, fenvalerate, monocrotophos, phorate falling under organo-phosphorus and synthetic pyrethroids pesticide groups were detected in the vegetable samples.

Recovery studies: In order to check the authenticity of the experimental procedure followed for extraction of different pesticides from vegetable samples, recovery studies were performed. All the targeted pesticides (aldrin, dieldrin, endosulfan-α endosulfan-β, endosulfansulphate, HCH-α, HCH-β, HCH-γ, heptachlor, acephate, chlorpyriphos, dichlorvos (DDVP), monocrotophos, phorate, profenofos, cyfluthrin-β, cyhalothrin-λ, cypermethrin, delta methrin and fenvalerate) were spiked at 0.01, 0.5 and 1.0 mg/kg on beans samples. Percent recoveries of insecticides for the bean samples indicated that recoveries are good and performance of extraction, clean-up and chromatographic parameters for the analysis of pesticide residues in vegetable samples are good.

The average recovery rates ranged from 77.0 to 95.4 % in beans and the average RSDs respectively ranged from 1.2 to 6.1 % in beans (**Table 1**). The variation in the recovery is not always possible to predict accurately but may be due to complete evaporation of the solvents on the rotary evaporator, oxidation due to solvent evaporation through direct air and degradation of pesticides [11].

Twenty pesticides residues in eight commodities (cucumber, tomato, pepper, green bean, eggplant, zucchini, melon and watermelon) of Spain at a recovery rate of 65–115 % and RSD value of 4-16 % were determined [12]. Scientists obtained mean recoveries of 78-104 % at fortification levels of 0.010 - 0.100 mg/kg and their relative standard deviations (RSDs) as ≤15 % [13]. It is reported that, pesticides recovered in vegetable samples ranged from 93% to 103% and RSD between 5 % and 10 % [14]. Scientists recorded an average recovery of 73.5 to 83.5 % for residues like α-cypermethrin and fenvalerate in grapes [15]. Pesticides recovered in spinach and eggplant in the range of 70-120 % and RSD less than 10 % [16]. It is reported that, pesticides recovered in fruits and vegetables ranged between 72 and 114 % with a RSD < 20 % [17]. It is observed that, mean recoveries of different insecticides in vegetables ranging from 94 to103 %, and the RSD below 10 % [18].

Analysis of fresh bean samples collected from five districts, Karnataka State indicated the presence of pesticide residues of different groups (OPs and SPs). The residue levels varied depending upon the type of vegetable and insecticide used. The pesticide residues in samples also differed from location to location which attributed to the different types and rates of application. It also reveals the preferred pesticide usage in the study area.

TABLE 1: AVERAGE RECOVERIES AND RSDS % OF DIFFERENT INSECTICIDES FROM SAMPLES OF BEANS AT FORTIFICATION LEVELS OF 1.0, 0.5, 0.01 MG/KG

Sl No	Pesticide groups	Name of the pesticide	Mean recovery (R.S.D) Beans (n=3) Level of fortification (mg/kg)		
			1	0.5	0.01
1	OCPs	Aldrin	90.6(3.8)	86.1(3.8)	87.4(3.7)
2		Dieldrin	82.7(5)	95.6(2.5)	82.5(5.1)
3		Endosulfan-α	73.2(4.5)	94(1.4)	77.6(2.1)
4		Endosulfan-β	78.8(2.1)	91.7(5.6)	80.6(3.6)
5		Endosulfan-Sulphate	85.2(3)	96.2(1.4)	88.5(5.6)
6		HCH-α	77.1(4.9)	88.7(5.6)	80.4(3.5)
7		HCH-β	83.8(3.2)	81.9(1.1)	85.6(3.5)
8		HCH-γ	79.1(6.2)	93(2.5)	76(1.5)
9		Heptachlor	89.4(5.5)	95.2(0.7)	90.7(2.1)
10	OPPs	Acephate	80.7(2.4)	89.2(6.2)	82.3(2.2)
11		Chlorpyriphos	86.4(1.4)	92.9(4.7)	84.5(5.4)
12		Dichlorvos	96.6(1.8)	92.2(2.3)	92.4(4.8)
13		Monocrotophos	88.4(1.5)	86.9(5)	88.4(1.5)
14		Phorate	84.3(5.9)	92.1(0.7)	85.6(3.5)
15		Profenofos	80.3(3.1)	88.5(2.7)	80.3(3.1)
16	SPs	Cyfluthrin-β	81.1(2.4)	86.9(5)	83(3.7)
17		Cyhalothrin	85.7(6.5)	82.6(2.2)	85.7(1.4)
18		Cypermethrin	86.2(2.6)	83.4(3.4)	93.9(2.6)
19		Deltamethrin	92.9(4.6)	91.5(1.6)	93.4(4.3)
20		Fenvalerate	76.1(1.6)	88.8(2.4)	77(2.6)

Acephate residues: The concentration of acephate residue in the beans from Bangalore rural, Bangalore urban, Chikkaballapura, Kolar and Ramanagara districts respectively varied from 0251 to 0.431 mg/kg (mean=0.132 mg/kg), 0.113 to 0.418 mg/kg (mean=0.12 mg/kg), 0.421 to 0.474 mg/kg (mean=0.169 mg/kg), 0.247 to 0.471 mg/Kg (mean=0.143 mg/kg), 0.271 to 0.421 mg/kg (mean=0.135 mg/kg). In all the five districts, 37.5 % of samples showed contamination with acephate residues. None of the samples showed acephate residue above MRL of 2.0 mg/kg. The trend of mean concentration of acephate residue in beans in district-wise is in the order of Ramanagara > Bangalore urban > Bangalore rural > Kolar > Chikkaballapura (**Table 2** and **3**). The results of our systematic study are in good agreement with the findings of an investigation conducted in India, Egypt and South America also. In the present case, the concentration of the results are in fair agreement with the other findings that, in the cabbage samples from different local markets of Egypt, acephate residue concentration varied from 0.182 to 0.457 mg/kg [19]. The results of the present

investigation are also supported the findings of a similar survey conducted by many workers, reported residue of acephate in the range of 0.05-0.37 mg/kg in the farmgate samples of cauliflower from Punjab [20] and the residue concentration of acephate in fruits and vegetables samples from South America as 0.06 - 0.028 mg/kg [21].

Chlorpyriphos residues: The concentration of chlorpyriphos residue in the beans from Bangalore rural varied from 0.152 to 0.182 mg/kg (mean = 0.064 mg/kg), with 37.5 % of samples having contamination. In Bangalore urban, it ranged from 0.133 to 0.172 mg/kg (mean = 0.044 mg/kg) with 25 % of samples showing contaminated with Chlorpyriphos. In Kolar, it varied from 0.165 to 0.181 mg/kg (mean = 0.043 mg/kg) and 25% of samples are found to be contaminated. Chlorpyriphos residue is below the detectable level

in the samples collected from Chikkaballapura and Ramanagara districts. The MRL for Chlorpyriphos residue is 0.2 mg/kg and none of the samples from Bangalore rural, Bangalore urban and Kolar districts showed chlorpyriphos residue above MRL of 0.2 mg/kg. The trend of mean concentration of chlorpyriphos residue in beans in different districts is in the order of Bangalore rural > Kolar > Bangalore urban. Chlorpyriphos pesticide residue in vegetable samples like potato, lettuce, tomato, onion, red pepper and green onion and recorded contamination with chlorpyriphos residue only in 12.5 % of the tomato samples, with residue concentration ranging from 0.14-0.16 mg/kg [22] and chlorpyriphos residue concentration of 0.003-0.115 mg/ kg in 82% of the vegetarian diet samples collected from homes, hostels and hotels periodically from Hisar, Haryana [23].

TABLE 2: SHOWING PESTICIDE RESIDUES (MG/KG) IN BEANS SAMPLES OF THREE DISTRICTS

Sl. No.	Pesticides	Bangalore rural (n=8)				Bangalore urban (n=8)				Ramanagara (n =8)			
		a(b)	min	a(b)	a(b)	a(b)	a(b)	a(b)	min	a(b)	min	max	mean
1	Acephate	3(37.5)	0.251	3(37.5)	3(37.5)	3(37.5)	3(37.5)	3(37.5)	0.251	3(37.5)	0.272	0.421	0.135
2	Chlorpyriphos	3(37.5)	0.152	BDL	BDL	BDL	BDL	3(37.5)	0.152	BDL	BDL	BDL	BDL
3	Dichlorvos (DDVP)	1(12.5)	BDL	BDL	BDL	BDL	BDL	1(12.5)	BDL	BDL	BDL	BDL	BDL
4	Monocrotophos	2(25)	0.011	BDL	BDL	BDL	BDL	2(25)	0.011	BDL	BDL	BDL	BDL
5	Phorate	2(25)	0.021	3(37.5)	3(37.5)	3(37.5)	3(37.5)	2(25)	0.021	3(37.5)	0.024	0.062	0.014
6	Cyfluthrin-β	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL
7	Cyhalothrin-λ	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL
8	Cypermethrin	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL
9	Deltamethrin	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL
10	Fenvalerate	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL

Note: BDL = Below detection limit, n = No. of samples analyzed, a = Contaminated, b = % of contamination

TABLE 3: PESTICIDE RESIDUES (MG/KG) IN BEANS SAMPLES OF TWO DISTRICTS

Sl. No.	Pesticides	Residue range (mg/kg)							
		Chikkaballapura (n=8)				Kolar (n=8)			
		a(b)	min	a(b)	min	a(b)	min	a(b)	min
1	Acephate	3(37.5)	0.421	3(37.5)	0.421	3(37.5)	0.421	3(37.5)	0.421
2	Chlorpyriphos	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL
3	Dichlorvos (DDVP)	2(25)	0.011	2(25)	0.011	2(25)	0.011	2(25)	0.011
4	Monocrotophos	2(25)	0.112	2(25)	0.112	2(25)	0.112	2(25)	0.112
5	Phorate	3(37.5)	0.021	3(37.5)	0.021	3(37.5)	0.021	3(37.5)	0.021
6	Cyfluthrin-β	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL
7	Cyhalothrin-λ	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL
8	Cypermethrin	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL
9	Deltamethrin	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL
10	Fenvalerate	BDL	BDL	BDL	BDL	BDL	BDL	BDL	BDL

Note: BDL = Below detection limit, n = No. of samples analyzed, a = Contaminated, b = % of contamination

Chlorpyrifos residues levels in 309 samples of fruits and vegetables (pineapple, lettuce, cabbage, cucumber and onion) samples sold in Ghanaian markets, with the residue level varying from 0.001-0.062 mg/kg. It is also found that chlorpyriphos in pineapple was higher than their respective

European Commission MRLs [24]. Mean concentration of chlorpyrifos was reported in eggplant (24.02µg/kg), cabbage (10.55µg/kg), cauliflower (2.85µg/kg), tomato (178.87 µg/kg) and ladyfinger (2.49 µg/kg) from Hyderabad, Andhra Pradesh, India [18] (**Table 2** and **3**).

Dichlorvos residues: For Bangalore rural district, the concentration of dichlorvos residue in beans was BDL to 0.021 mg/kg (mean = 0.003 mg/kg), with 12.5% of samples proved to be contaminated with Dichlorvos. In Chikkaballapura district, it ranged from 0.011 to 0.023 mg/kg (mean = 0.004 mg/kg), with 25 % sample contamination. It is BDL to 0.02 mg/kg (mean = 0.003 mg/kg) with 12.5 % sample contamination in Kolar. The dichlorvos residue concentration is not detected in beans samples collected from Bangalore urban and Ramanagara districts. None of the samples from Bangalore rural, Bangalore urban and Kolar districts showed dichlorvos residue above MRL of 0.15 mg/kg. The trend of mean concentration of dichlorvos residue in beans in different districts is Chikkaballapura>Bangalore rural=Kolar. These results of the present investigation are supported by findings of a similar investigation [25] reported dichlorvos residue concentration ranging from 0.004 – 0.022 mg/kg in cabbage, cauliflower, pea grains, brinjal, tomato, potato and green chilly samples collected from wholesale markets of Hisar, Haryana (**Table 2** and **3**).

Monocrotophos residues: In Bangalore rural district, the concentration of monocrotophos residue in beans varied from 0.011 to 0.05 mg/kg (mean = 0.008 mg/kg) and 25 % of samples were contaminated with monocrotophos residues. Contrast to this, in the samples from Chikkaballapura district, it is ranging from 0.112 to 0.153 mg/kg (mean = 0.033 mg/kg) with 25 % contaminated samples and in Kolar samples, it is range between 0.106 to 0.154 mg/kg (mean = 0.033 mg/kg) with samples having contamination accounting to 25 %. Monocrotophos residue concentration is below detectable limit in Bangalore urban and Ramanagara districts, while none of samples are having Monocrotophos residue values above the MRL of 0.2 mg/kg in Bangalore rural, Chikkaballapura and Kolar districts. The trend of mean concentration of monocrotophos residue in beans in different districts is

Chikkaballapura = Kolar>Ramanagara> Bangalore rural>Bangalore urban. 9.5 % per fruit samples contained 0.030 µg/g monocrotophos and 14.2 % of guava fruit samples had detectable amount of monocrotophos [26]. Similarly, a study revealed the presence of monocrotophos in the range of 0.001-0.044 mg/kg in vegetable samples (*viz.,* brinjal, cucumber, okra, ridge gourd and tomato) collected from Andhra Pradesh, India [27] (**Table 2** and **3**).

Phorate residues: In Bangalore rural, the concentration of phorate residue in beans varied from 0.021 to 0.026 mg/kg (mean = 0.006 mg/kg) showing 25 % of samples contaminated with phorate. In Bangalore urban, it ranged from 0.011 to 0.035 mg/kg (mean = 0.01 mg/kg) illustrating 37.5 % of samples contamination. In Chikkaballapura, it is 0.021 to 0.026 mg/kg, with a mean value of 0.009 mg/kg and contamination in 37.5 % of samples. It ranged from 0.022 to 0.061 mg/Kg (mean = 0.01 mg/kg) with 25 % of samples contamination in Kolar and 0.024 to 0.062 mg/kg (mean = 0.014 mg/kg) with 37.5 % contamination in Ramanagara district. None of samples from all the districts crossed the MRL for phorate residue being 0.05 mg/kg excepting 12.5 % of the samples each from Kolar and Ramanagara districts. Mean concentration trend of phorate residue in beans in different districts is Bangalore urban = Kolar>Chikkaballapura> Bangalore rural. These results are in fair conformity with the findings of earlier work [28] revealed the presence of phoratein Shanghai green (0.0257 µg/g) and Chinese cabbage (0.0398 µg/g) from Nanjing, China. The present investigation results are also endorsed by earlier findings [29] reported that, phorate residues in fruits and vegetables which vary from BDL to 0.405 mg/kg from Xiamen, China (**Table 2** and **3**).

Deltamethrin residues: The concentration of deltamethrin residue in beans ranged from 0.361 to 0.413 mg/kg (mean = 0.111 mg/kg) with 25 % of samples contaminated with it in Bangalore urban district. The measured concentration is above the MRL of 0.2 mg/kg. In contrast, it is not detected in the bean samples from Bangalore rural, Chikkaballapura, Kolar and Ramanagara districts. 68 % contamination in vegetarian diet samples with deltamethrin residue (0.008-0.102 mg/kg) represented by samples from homes, hostels and hotels from Hisar, Haryana [23].

The levels of deltamethrin varied between 0.007-0.010 mg/kg (mean: 0.008 mg/kg) in pear and 0.026-0.062 mg/kg (mean: 0.044 mg/kg) in pineapple samples from Kumasi markets [30] (**Table-2** and **3**).

Cyfluthrin-β, cyhalothrin-λ, cypermethrin and fenvalerate residues: The concentration of cyfluthrin-β, cyhalothrin-λ, cypermethrin and fenvalerateresidues is below detectable level in the beans from all the five districts. It was reported that, residue of cyfluthrin-β (0.11-0.18 mg/kg) in the farmgate samples of cauliflower from Punjab [20] and cyfluthrin-β residue contents to vary from 0.004-0.010 mg/kg in watermelon (mean: 0.008 mg/kg) to 0.018-0.021 mg/kg in pineapple (mean: 0.020 mg/kg) samples collected from markets in Kumasi [30]. It was observed that, cyhalothrin- λ (0.14 mg/kg) in farmgate samples of cauliflower from Punjab [20] and cyhalothrin-λ residue value of 0.057-0.125 mg/kg in fruits and vegetables samples from South America [21]. Cyhalothrin- λ residue of 0.073 mg/kg was reported in 132 samples of fruits, vegetables, herbs and spices collected from Egyptian local markets [31] (**Table 2** and **3**).

CONCLUSION: Crop producers also have to apply the principle of 'due diligence' to ensure the application of pesticides only where pest/disease pressures and /or potential crop losses make it cost effective. Prolonged use of pesticide will promote pest resistance, each time when higher doses are applied. Regular monitoring of the vicinity should be encouraged to avoid possible consumption of food stuff. Farmers should be educated and encouraged and to protect people from the impact of higher levels of pesticides establishing standards for pesticides in vegetables and fruits are necessary. The standard so established should be reviewed from time to time.

CONFLICT OF INTEREST: Authors have declared that no conflict of Interests exists.

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