Research Article

Evaluation of Pesticide Residual Toxicity in Vegetables and Fruits Grown in Bangalore Rural District

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ABSTRACT

Two vegetables (Potato and Tomato) and one fruit (Grapes) were selected randomly from a farmer's field in Bangalore rural district. Pesticides were quantified by Gas Chromatograph and Electron Capture Detector (GC-ECD) and the analysis was carried out in Pesticide analysis laboratory, Department of Agriculture, Government of Karnataka, Bangalore. Insecticides identified and quantified were endosulfan, methyl parathion, malathion, chloropyrifos, cyhalothrin, quinolphos and fenvelerate. Eight potato samples were analysed in duplicate and six samples found to contaminated with endosulfan (75%), four were contaminated with parathion (50%). Greater MRL values were observed in two samples containing endosulfan ranging from 0.057-2.818mg kg^{-1} and one sample with quinolphos (0.007-0.09mg kg^{-1}), cyhalothrin and fenvalerate were completely absent in the samples. Tomato samples analysed indicated endosulfan and fenvalerate found to contain higher MRL values where percentage accumulation of endosulfan has residue range 0.05-2.24mg kg^{-1}. A few samples of grapes were contaminated with acephate, cypermethrin, Metaloxyl and monoprotophos. One sample of grape contaminated with Imidocloprid and the values ranged from 0.090mg kg^{-1} and 0.196mg kg^{-1}.

Keywords: Pesticides, Potato, Tomato, Grapes, Toxicity.

INTRODUCTION

India has fluctuating climatic condition with wide variety of soils on which vegetables and fruits can be grown. Thickly populated country in the world and consumption of fruits and vegetables are steadily increasing year after year. In India, vegetables are major constituents of diet as majority of Indians are vegetarian, with a per capita consumption of 135gm/day as against the recommended 300gm/day. However, several factors limit their productivity, mainly insect pests, and diseases. As several insect pests attack the vegetables, they are produced under very high input pressure[1]. Considerable emphasis is laid on production of these crops during the last two decades. Indian agriculture is predominantly traditional oriented and pest management is a built in process but not a separate activity. Pest damage severely constraints vegetable and fruit production in India. Different patterns of pesticide production and its use has been analyzed and insecticide use is around 75% in the country compared to 32% in the world. Herbicide is used only 12% compared to worldwide consumption of 47%. Carbomate and synthetic pyrethroids are most globally used (45% together). Organophospate constitute 50% of the consumption and bio-pesticides are used only up to 1% compared to 12% worldwide[2]. In India, DDT and BHC are the two chemicals used in agriculture and public health programmes. Though, these chemicals are banned commercially, they are very much in use due to ready availability at low cost and wide spectrum of activity and these molecules are very much stable in the environment. Plant protection products (more commonly known as pesticides) are widely used in agriculture to increase yield, improve quality, and extend the storage life of food crops[3,4]. Vegetables and fruits constitute the vital constituents used in day-to-day diet and residual toxicity occurs due to over usage of pesticides. Pesticides can cause respiratory and memory disorders, birth defects, depression, neurological deficiencies, miscarriages etc. The applied chemicals and/or their degradation products may remain as residues in agricultural products, which become a concern for human exposure. Selected sampling programmes can be used to investigate residual levels of pesticide in the environment, their movement and relative rates of degradation[5].

The main objectives of present investigation are:

- Qualitative and quantitative analysis of residues in selected fruits and vegetables
- To determine the range of values due to spraying of pesticides.
- To calculate the percentage of contamination due to pesticides.

MATERIALS AND METHODS

Sampling

Eight samples of two vegetables viz., tomato and potato and one fruit viz., grapes (one kg each) were collected from the growing areas of Bangalore rural district during January to march 2012. Samples were kept in a refrigerator (5°C) till analysis. All the samples were extracted fresh and information regarding pesticide applied to vegetable crops was collected from farmers at the time of sampling. Composite sample consisted of 1 kg was cut into small pieces and macerated in a grinder[6,7].

Chemicals

The glassware were rinsed with acetone and dried in an oven at around 350°c prior to use. 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. Sodium chloride and anhydrous sodium sulfate, AR grade were procured from Himedia pvt .Ltd. India. Before use, anhydrous sodium sulfate was purified with acetone and heated for 4h at 600°C in muffle furnace to remove possible phthalate impurities. Florosil, 60-100 mesh, purchased from Merck India limited was activated at 350°C for 5h before use. The pesticide standards were procured from All India Network project on pesticide residues, Division of Agricultural Chemicals, Indian Agricultural Research Institute (IARI), Delhi, India[8,9].

Preparation of Standard Solution

An accurately weighed 10mg of an individual analytical grade pesticide was dissolved in 10ml volumetric flask using n-hexane to prepare the standard stock solution to 1000mg kg^{-1}. Standard stock solution of each pesticide was serially diluted to obtain immediate lower concentration of 100mg kg^{-1}. A mixture of standard stock pesticide solution was prepared by taking 0.1ml solution of compatible (acephate, aldrin, chlopyriphos, cyfluthrin-B, cyhalothrin, cypermethrin, deltamethrin, dichlorvos, dieldrin, a-endosulfan, B-endosulfan, endosulfan–sulfate, fenvalerate, α-HCH, β-HCH γ-HCH, heptachlor, monocrotophos, phorate and profenofos) pesticide in a 10ml volumetric flask and making the volume up to the mark with n-hexane. Standard mixture contained 10mg kg^{-1} of solution to determine the time of detection of the instrument. They were stored in a refrigerator at 5°C[10,11].

Extraction and Cleanup

Fresh fruits and vegetables samples were thoroughly shredded and homogenized .Approximately 20g of the sample was macerated with 40ml of ethyl acetate. Sodium hydrogen carbonate 5g and anhydrous sodium sulfate 20g was added to remove moisture and further macerated for 3minutes in ultra-turax macerator. Samples were then centrifuged for 5min at 3000rpm to obtain two phases. Extraction process was followed by clean-up step using solid-phase extraction with florisil. Florisil column (500mg/8ml) cartridge was conditioned with 10ml with ethyl acetate. Pesticides in sample extract (5ml) were eluted with 10ml of ethyl acetate. Concentrated to 1ml using rotary evaporator and dried by a gentle nitrogen stream. This was dissolved in 1ml of ethyl acetate. Pesticide were then quantified by gas chromatograph and electron capture detector (GC-ECD) and the analysis was carried out in Pesticide analysis laboratory, Department of Agriculture, Government of Karnataka, Banashankari, Bangalore[12-14].

RESULTS AND DISCUSSION

Potato

It is called *Solanum tuberosum* L belong to the family solanaceae. It is a starchy and tuberous crop. Word potato may refer to the plant as edible tuber. Commonly occurring disease is bacterial wilt, black leg, bacterial soft root, ring rot, black dot, early blight etc.

Table-1 Indicates the retention time percentage of contamination and residue range. Insecticides identified and quantified were endosulfan, methyl parathion, malathion, chlorpyrifos, cyhalothrin, quinalphos and fenvalerate. Eight samples of potato analyzed in duplicate for the presence of insecticide residues revealed that six samples were contaminated with endosulfan (75%), four were contaminated with methyl parathion (50%), three each were contaminated with chlorpyrifos (37.5%) and quinalphos (37.5%) and two samples with malathion (25%). Cyhalothrin and fenvalerate were not detected in any samples. It was observed from the table that, though potato is contaminated with endosulfan, >MRL values were noticed in two samples with residue ranging from 0.057-2.818mg kg^{-1}. Quinalphos which has retention time 12.27 and MRL value 0.01mg kg^{-1} was observed in one sample exceeding the MRL and was found to be in the range 0.007-0.09mg kg^{-1}.Rest of the insecticides methyl parathion, malathion and chlorphyrifos though contaminated the potato but still they were found well within the standard MRL values. A methyl parathion (0.013-0.372 mg kg-1) value does not exceed the MRL. Endosulfan contamination in a few samples of potato is high because pests are active during February to May when the vegetables are growing where a situation arises to illegally to use OCPs. Sometimes high temperatures in summer volatilized the OCPs from their reservoirs such as soil or vegetation and that the edible parts of the crop may have tapped some of the evaporating pesticides[15].

Farmers use pesticides in potato crop to overcome bacterial, viral and fungal diseases. With the exception of endosulfan and quinalphos, the accuracy and precision obtained for all pesticides were considered to be acceptable. Pesticides are commonly detected in potatoes. Chloropropham which is applied post harvest to control sprouting was most frequently used and was detected in 5% of potatoes. It was observed that, Azoxystrobin used as fungicide to control plant diseases in various crops including potatoes. All samples were below the Canadian MRL of 30mg kg^{-1}. Other pesticide detected in potato was imidocloprid which is a neonicotinoid insecticide used to control colorado potato beetle[16]. Potatoes (31.3%) were contaminated with OCPs metamidophos[17]. Maximum residue limits of metamidophos values found in the samples of potato did not exceed the levels permitted[18]. Fenitrothion and malathion insecticides were detected in 50% and 39.8% of the unprocessed potato samples with the mean concentration of 28.0ppb and 21.2ppb respectively[19]. No chlorinated insecticide or nitrogen containing halogenated fungicides were detected[20-22]. Farmers were personally interviewed at the place of sampling collection of potato mention that, they spray pesticides without consulting the experts or department officials. They blindly follow the instruction given and they are much ignorant about the dose, nature, methodology and chemistry of pesticide. They use pesticides indiscriminately without giving due weightage to the concentration of pesticides used. It was learnt that, farmers involved in cultivation of potato and other vegetables on the same land and may be due to regular spraying, pesticides might have adhere to the soils and long term persistence in the environment. Similarly farmers from the adjoining lands involved in other agricultural activities also spray pesticides to combat the diseases. This complex combination of various pesticides and their persistence nature may be the main reason for the occurrence of OCPs, SPs and OPs in potatoes selected for analysis. Farmers also spray pesticide for long term storage of potatoes and this may be the reason for contamination of potatoes with pesticides.

Tomato

Tomato lycopersicon lycopersicum (L.) Karsten is commonly grown throughout the globe and particularly for the red fruit it bears. Tomato was originated in South America and popularly consumed by large population in India. Fruit is rich in lycopene that may have beneficial health benefits. There are several diseases commonly infest tomato are leaf curling, occurrence of aphids, early blight, septoria, leaf spot etc.

Table 1: Pesticides, number of samples and residue range in Potato

S. No	Pesticide	Number of Samples		MRL Values (mg kg^{-1})	>MRL Values (mg kg^{-1})	Residue Range (mg kg^{-1})
		Analyzed	Contaminated			
1	Endosulfan	08	06	2	1	0.057-2.818
2	Methyl Parathion	08	04	1	NIL	0.013-0.372
3	Malathion	08	02	3	NIL	0.115-0.513
4	Chlorpyrifos	08	03	0.2	NIL	0.01-0.09
5	Cyhalothrin	08	00	0.2	NIL	ND
6	Quinalphos	08	03	0.01	1	0.007-0.09
7	Fenvalerate	08	00	2	NIL	ND

Table 2: Indicate the pesticide, number of samples and residue range in Tomato

S. No	Pesticide	Number of Samples		MRL Values (mg kg^{-1})	>MRL Values (mg kg^{-1})	Residue Range (mg kg^{-1})
		Analyzed	Contaminated			
1	Endosulfan	08	05	2	1	0.05-2.241
2	Methyl Parathion	08	06	1	NIL	0.04-0.078
3	Malathion	08	03	3	NIL	0.02-2.047
4	Chlorpyrifos	08	00	0.2	NIL	ND
5	Cyhalothrin	08	03	0.2	NIL	0.03-0.178
6	Quinalphos	08	00	0.01	NIL	ND
7	Fenvalerate	08	04	2	1	0.0024-2.317

Table 3: Pesticide, number of samples and residue range in Grapes

S. No	Pesticide	Number of Samples		USFDA (MRL) (mg kg^{-1})	>MRL Values (mg kg^{-1})	Residue Range (mg kg^{-1})
		Analyzed	Contaminated			
1	Acephate	08	02	0.01	NIL	0.003-0.009
2	Cypermethrin	08	01	0.05	NIL	0.039-0.068
3	Delta-methrin	08	02	0.2	01	0.052-0.262
4	Endosulfan	08	02	0.05	NIL	0.012-0.027
5	Methyl Parathion	08	0	0.1	NIL	ND
6	Metaloxyl	08	0	2	NIL	ND
7	Imidacloprid	08	1	1	01	0.090-0.196
8	Monoprotophos	08	0	0.05	NIL	ND

Table-2 depicts the percentage contamination. Samples exceeded the MRL value and insecticides standard MRL value and range of residues. Eight samples were analyzed for insecticide residues. Results indicated that, endosulfan and fenvalerate found to contain higher MRL values. Six out of seven insecticides were detected and chlorpyrifos and quinalphos do not show any accumulation. Endosulfan contamination was found to be 67.5% and residue range 0.05-2.241mg kg^{-1}. Although methyl parathion, malathion and cyhalothrin found in the tomato sample and contamination vary from one insecticide to other. Methyl parathion has residue range 0.04-0.078mg kg^{-1} followed by 0.02-2.047mg kg^{-1} and 0.03-0.178mg kg^{-1} in malathion and cyhalothrin respectively. It was observed that, in tomato samples, percentage contamination was in the order methyl parathion (75%), fenvalerate (50%) and cyhalothrin (37.5%) and no detection was made in case of chlorpyrifos and quinalphos. Seven pesticide residues were detected, all being organochlorine pesticides. Endosulfan was extensively used in Ghanian agriculture to control aphids in tomato. Metabolites present in tomato samples as result of exposure to intense sun light or presence of an appropriate microbial species and suitable soil conditions and sometimes due to biodegradation of pesticide aided by fungi and bacteria[23]. Eight pesticides were observed in all samples analyzed, three organochlorine pesticides (Dicofol, chlorothalonil and endosulfan) and four pyrethroids (Bifenithrin, deltamethrin, lambda cyhalothrin and cypermethrin). Endosulfan is the pesticide that shows highest residue levels in tomato and lambda cyhalothrin with lowest residue. Out of 20 samples, eight samples found to have a residue value of 1-1.20mg kg^{-1} of endosulfan and two samples with 0.30-1mg kg^{-1} of delta methrin[24]. Insecticide residues were identified in seven tomato samples. Endosulfan was detected in all the samples, one sample each is contaminated with quinalphos and methyl parathion, malathion was recorded in two samples, chlorpyrifos in two samples and quinalphos and chlorpyrifos have been estimated qualitatively and quantitatively in tomato. In the samples of tomato, two pesticides namely endosulfan and cypermethrin were found and concentration of the detected pesticides determined from the area of package[25]. Most of the insecticides applied are potent toxins and their intensive use poses potential hazards to human's livestock and the environment[26,27].

From the study, it was observed that, only two samples exceeded the MRL. The MRL is not a toxicity level, but nothing to do with toxicology and due to non-observance of certain measures which includes respecting safety intervals between the last pesticide application and harvest of the crop or using the recommended rate of pesticide application[28]. In order to avoid such maladies, producer should follow the recommend, authorized and correct ways of using pesticides to control pests and diseases in their crops. These chemicals are toxic by nature, but when use in appropriate and safe manner as specified on the labels, they should not be harmful to the user, consumer or environment. It is their misuse which can be source of hazards. Vegetable if cleaned and washed, some percentage of insecticide could be removed. Some plant synthesizes insecticide and also could occur naturally. Variety of toxins are synthesized by plants for their protection against fungi, insect and animal predators and these natural pesticides occur in very much larger chemical structure types and at levels sometimes higher than synthetic pesticides[29]. According to farmer's perceptions, red spider mites and boll worms are the major pests of tomato. There is considerable increase in population of red spider mite incidence in Doddaballapura and Hoskote Taluks. The fact is that, farmers applied insecticides were not recommended for vegetable pest control is indicative of poor knowledge associated with pesticide use. Chemical pesticides disrupts the natural control systems, increase the risk of contamination of the farm environment and pesticide residues in fresh products.

Grapes

Grapes technically called *Vitis vinifera* L. belongs to the family Vitiaceae. It is cultivated in many parts of India. Fruits are edible and the crop is confronted with various diseases such as anthracnose, *Downey mildew*, *Powdery mildew*, bacterial leaf spot etc. To prevent the incidence of diseases different pesticides are sprayed.

Table-3 depicts residue range, percentage contamination MRL and samples exceeded MRL values. Residues in grapes were analyzed for eight compounds. Pesticide analyzed in the samples includes acephate, cypermethrin, deltamethrin, endosulfan, methyl parathion and imidcloprid showed greater MRL values. Further a few samples of grapes were contaminated with acephate, cypermethrin, metaloxyl and monoprotophos. Percentage contamination was 25% in acephate, deltamethrin and endosulfan and cypermethrin (25%) and imidacloprid (25%) were also noticed. Acephate has residue range 0.003-0.009mg kg^{-1} was found well within the MRL (0.01mg kg^{-1}). Cypermethrin though detected but has minimum residue values 0.039mg kg^{-1} and maximum of 0.068mg kg^{-1} compared to MRL (0.01). Endosulfan has residue range 0.012-0.027mg kg^{-1} where as one sample was contaminated with imidacloprid with minimum and maximum residue ranges showing 0.090mg kg^{-1} and 0.196mg kg^{-1} respectively. Cypermethrin, dimethoate, malathion and profenofos insecticides and dicarbomate fungicides were detected in grapes samples[30]. Synthetic pyrethroids analysis carried out for cypermethrin, fenvalerte, fluvalinate and delta methrin. Fluvalinated and fenvalerate was detected in six samples of grapes. Among fruits, grapes contained maximum residues[31]. Pesticides such as imazalil, thiabendazole, decamethrin were present in the analysed samples of grapes[32]. During interaction with the farmers at Devanahalli taluk, they informed regarding usage of several pesticides to control *Downy mildew*. They commonly use captan, malathion, dithane M-45 to combat fungal diseases. Unfortunately, spraying of pesticides was indiscriminate without giving due advantage to chemistry of pesticides and physiological behavior of grape plants. Results indicated that, deltamethrin was a dominant contaminant and most possible reason of more frequent residues of deltamethrin for controlling fruit fly attack close to harvest stage. Presence of OP and SP residues in vegetables and fruits is an indicative of change in usage pattern of insecticides in India. Monitoring studies are imperative to know actual status of contamination due to toxic pesticide residues for future polices as well as to strengthen the confidence of consumer in quality of food[33]. Consumer should aware of the concept of Acceptable Daily Intake (ADI) in the evaluation of safety of pesticide residues in food. ADI of pesticides is the amount of those pesticides that can be ingested daily by man during an entire life time without an appreciable risk to the consumer health on the basis of all known facts[34].

CONCLUSION

Farmers of Bangalore rural district supply considerable amount of vegetables to the large populace of Bangalore to suffice their needs. The district enjoys warmer climatic condition, vegetables tend to be very susceptible to pest infestation and diseases. Farmers have been spraying the pesticide in order to combat pest and diseases and to increase the crop yield since several years. Chemicals accumulate in soil, persistent in air and sometimes remain on stagnant and runoff water. Majority of the farmers are ignorant of chemicals and chemistry of pesticides. Some spray chemicals blindly following the instruction given. Prior consultation with department official or any resource person is much essential. Farmers should also observe the safety period before the harvest of fruits and vegetables. Prolonged use of pesticide will promote pest resistance, each time when higher doses are applied. In view of an increasing trend in pesticide uses in Karnataka, continuous monitoring of pesticide residue in food crops is desirable in order to protect the end user from health hazards and generate baseline data upon which, future plans could be envisaged.

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