Original Research Article

DOI: https://dx.doi.org/10.18203/2394-6040.ijcmph20205730

The level of residue of pesticides in vegetables by GC-MS, health risk and the negligence in implementation of policies, India

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Received: 31 October 2020 Revised: 16 December 2020 Accepted: 17 December 2020

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ABSTRACT

Background: Over the past years there has been an increase in the use of pesticides in vegetable farming developing countries. This study describes over spraying pesticide use among small-scale farmers in Jasra block of Allahabad district of Uttar Pradesh.

Methods: The purpose of this study was proposed to determine the pesticide residual level in 120 vegetable samples like brinjal, lady finger, tomato, chili, cabbage analysed by (GC-MS) gas chromatography mass spectrophotometry technique. Assessing the health risk due to the daily consumption of contaminated vegetables Jasra block of Allahabad district

Results: Samples were contaminated with organophosphorus group namely chlorpyriphos in bringal 1.215 mg/kg, and less carbamate (carbaryl) from 0.025 mg/kg, ladyfinger with the concentration of chlorpyriphos 0.418 mg/kg carbaryl 0.092 with very less residue of dichlorvos 0.005, green chilli with the concentration of chloropyriphos 1.507 mg/kg. Carbaryl 0.033 mg/kg, tomato with the concentration of chlorpyriphos 1.631mg/kg followed by less carbaryl 0.026 mg/kg, malathione, 1.890 mg/kg followed by cabbage with the concentration of chlorphyriphos 1.907 mg/kg. And less carbaryl concentration was recorded 0.064 mg/kg.

Conclusions: Five vegetable with pesticides residues, exceeding the maximum residue level (MRL) recommended by FSSAI, FAO/WHO. Farmers over spraying methods leads to acute diseases like respiratory tract infection, extreme weakness, and longer use pesticides exposure leads to chronic diseases like asthma, anxiety, depression, cancer, leukemia, allergies, nausea, vomiting, headache, blur vision, skin itching etc.to minimize the risk of diseases policy making and farmer protection standard and proper handling techniques, trained professionals to improve health safety.

Keywords: Acute, Carbamate, Chronic, FAO, FSSAI, GC-MS, Leukemia, Maximum residual limit, Organophosphorus, Pesticides residue, WHO

INTRODUCTION

Agriculture plays an important role to give food security in this highly populated country. The major crops grown in the country are rice, wheat, jute, potato, sugarcane, vegetables and tea. Chemical are always not very useful to humans and environment they may be useful for the productivity but they leads to many acute and chronic diseases with long term use when they remain as a residues in fruits and vegetable. That could lead to

possible health risk to consumers Investigation of pesticide residue levels in vegetables is a main concern of many researchers to evade possible risks of toxicity to human health.² Therefore there some national and international organizations set standard for maximum residual limit (MRL) for food items which usually monitor and regulate the amount of pesticides in foods. MRL for pesticide residues relates to maximum amount of that residue (expressed in mg/kg) that is lawfully permitted in particular food items (www.pmfai.org/

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stat.htm). The literary rate is somehow good in urban areas but the condition of rural areas are still in worse condition with no educational background that leads the farmers inappropriate practice of pesticides spray in vegetable farming. Contaminated vegetables are potential sources of compounds that can be hazardous to the health of consumers.3 Due to lack of education, cognizance and general evidence with regard to the usage of pesticides from government organizations/agencies, farmers are in distress from the bad effects of pesticides during improper handling, disposal and especially when they are not protected with personal protective equipments (PPE). Commonly the farmers depend on the use of synthetic pesticides. Though, Most of them know that these pesticides are hazardous chemicals and lethal to their well-being but still they are using repeatedly to get more yields from their cultivated lands. However, they are totally unaware to integrated pest management (IPM) approach. Use of high doses of pesticides lead to the adulteration of their products resulting to potential risk for the consumer's health.

METHODS

Study area

The study was targeted mainly the agriculture area of Jasra block of Prayagraj (Allahabad) district. Five vegetable samples brinjal, lady finger, tomato, chili, cabbage, were selected because these are vegetables mostly grown in this area. Commonly consumed by the local population. The vegetable samples were randomly collected from the agricultural fields of vegetable-growing regions

Study design

Pesticide residues were determined in 120 vegetable samples collected from the farm of Jasra area of Parayagraj (Allahabad),transported to Jamia Hamdard, laboratory, New Delhi, according to standard sampling procedure (Cook, 2002). The vegetable samples (24 of each crop) surveyed for the present study were brinjal, cabbage, green chili, ladyfinger tomato. Samples were stored at 4°C prior to extraction procedure.

Chemical standards and reagents

Pesticide standards were purchased from (Barafkhana, old Delhi, India) with purity between 97% and 99%. Cyclohexane and acetone and sodium sulfate anhydrous were purchased from individual stock solution of each pesticide standard was prepared in acetone at a concentration of 1 mg/kg and stored in a freezer at (-18°C). Working standard solutions were prepared by appropriate dilutions with cyclohexane and stored under refrigeration (4°C). The extraction process was followed by a clean-up step using column chromatography with a column packed to remove any residual components that

may interfere with the gas chromatography mass spectrophotometry (GC-MS) analysis proposed by Stan.⁴

Apparatus

The samples were analysed by GCMS instrument (Agilent 7890A), Agilent Technologies, USA) Equipped CTC-PAL auto sampler attached with a mass spectrophotometer detector (Agilent 5975C inert XL EI/CI MSD with triple axis detector, Agilent Technologies, USA). A HP-5MS capillary column 30 m × 0.25 mm I.D., 1 1.n film thickness.

The samples were injected with a 5:1 split ratio on to 30 m \times 0.25 mm \times 0.25 p,m Agilent, HP5MS column polymethylsiloxane (composed of 5% Agilent Technologies USA). Inlet temperature was set at 250°C. Carrier gas (He) flow was 1.5 ml/minute. After 3 minutes, solvent delay time at 50°C, the oven temperature was increased at a rate of 10°C/minute to 200°C for 5 minutes and 5°C/minute to 250°C for 10 minutes. High pure helium (99.999%) was the carrier gas set at a constant flow rate of 1.5 ml/minute the injection port, transfer line, and ion source temperatures was all set at 250°C. 70eV of EI was adopted, and the mass scanning range was set from 50 to 700 amu in full scan. Solvent delay time was set to 3 minutes for all samples generated by different methods. MSD chemstation software will be used to process data. The compound separated was identified by comparing the obtained mass spectra of the analytes with those of authentic standards from the PEST libraries and with the mass spectra published previously. Peak areas of all components were calculated by MSD chemstation, and relative amounts of volatile compounds was calculated on the basis of peak area ratios. Peak areas of all components were calculated by MSD chemstation, and relative amounts (RAs) of pesticide compounds was calculated on the basis of peak-area ratios

System suitability

System suitability test was carried out to ensure reproducibility of the equipment. 2.0 p,l of the standard solution of Chlorphyrifos and Carbaryl was injected six times. The RSD was found to be less than 2%.

Preparation of vegetable sample calibration of standard

Individual stock standard solutions (1 mg/ml) were prepared in ethyl acetate and stored in the dark at -20 °C. Prior to their use, they were kept for 1 hour at ambient temperature. A mixed stock standard solution of pesticides was prepared in ethyl acetate and hexane (1:1) at $10\mu/ml$ with respect to each pesticide. Spiked calibration curves at levels of 10, 50, 100, 200, 400, 500 ng/gm in triplicates were prepared by addition of $10~\mu l,~50\mu l,~100\mu l,~200\mu l,~400\mu l,~500\mu l,~of~mixed standard stock solution, respectively. A stock solution of chlorophyrifos and carbaryl in ethyl acetae hexane (1:1) at concentration of 1 mg/ml was used.$

RESULTS

Demographic data, clinical characteristics of the study

The reason for investigation to assess pesticide residues in Allahabad region is that it is one of major commercial center for vegetables not for only nearby areas but also for the whole region. Therefore, it is very vital to know the concentration status of pesticides residues of vegetables before coming to the market Also, to evaluate environmental burden of pesticides it is very essential to observe the amounts of pesticides in vegetable samples. Because spraying is prevalent from 20 years in this region

Table 1: Quantitative estimation of pesticides in vego	etables.
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Samples	Pesticides residue detected	Range of pesticides residue detected (mg/kg)	MRL found (mg/kg)	Recommended MRL (FSSAI)
Bringal	Chlorpyriphos	0.000-1.101	1.215	0.2
	Carbaryl	0.000-0.010	0.025	0.5
Lady finger	Chlorpyriphos	0.000-0.101	0.418	0.2
	Carbaryl	0.000-0.020	0.092	0.5
	Dichlorvos	0.000-0.010	0.005	0.15
Green chilli	Chlorpyriphos	0.000-1.100	1.507	0.2
	Carbaryl	0.000-0.010	0.033	0.5
Tomato	Chlorpyriphos	0.000-1.000	1.631	0.2
	Carbaryl	0.000-0.010	0.026	0.5
	Malathion	0.000-0.210	1.890	3.0
Cabbage	Chlorpyriphos	0.000-1.001	1.907	0.2
	Carbaryl	0.000-0.010	0.064	0.5

The pesticides selected for this study were included organophosphate, organochlorine, carbamate pyrethrin classes, which could be useful to ensure the applicability of this study and to determine all different classes of pesticides in future. Chlorpyrifos is an organophosphate insecticide that inhibits acetylcholinesterase and is used to control insect pests. Chlorpyrifos is moderately toxic and leads to endocrine distruption. Its chronic exposure has been allied to neurological effects, developmental disorders, and autoimmune disorders. Carbaryl is one of most toxic carbamate pesticides and considered to responsible for highest acute toxicity to human as relate to any other insecticide used. It is used to control insects in a wide variety. Since its toxic effects are due to its activity as a cholinesterase inhibitor it is measured as neurotoxic pesticide. Malathion is an organophosphate para sympathomimetic pesticide that is widely used in agriculture, insecticide of relatively low human toxicity; however, absorption or ingestion into human body readily results in its metabolism to malaoxon, which is substantially more toxic. Several highly sensitive and selective analytical procedures are to determine the level of pesticidal residues residues in a variety of food matrices have been earlier described Self-perceived pesticides poisoning symptoms were also recorded. Like headache, vomiting, dizziness, abdominal pain, skin rashes and blurred vision by long term use of pesticides in field. There is very few information available on the levels of pesticide residues on fruits, vegetables and other crops produced in developing countries such as India. The present study was undertaken to determine the levels of pesticide residues on vegetables for use as a reference

point for future monitoring and to allow preventive measures to be taken to minimize the human health risks.

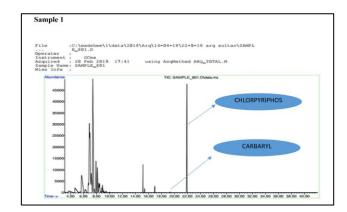


Figure 1: Chromatogram of brinjal.

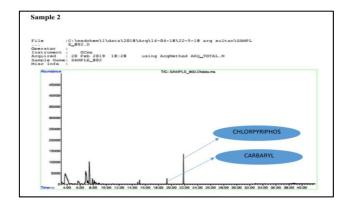


Figure 2: Chromatogram of ladyfinger.

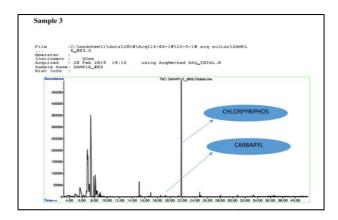


Figure 3: Chromatogram of chili.

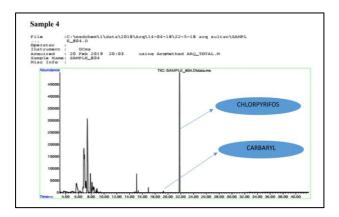


Figure 4: Chromatogram of cabbage.

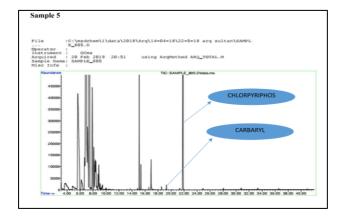


Figure 5: Chromatogram of tomato.

DISCUSSION

The above table shows the concentration of chlorpyriphos in is 1.215 and 24 carbaryl in brinjal is 0.025 which was collected from research area were of min. range was 0.000 and 1.101 whereas in case of carbaryl minimum was 0.000 and maximum was 0.011. The average value of chlorpyrifos residues obtained was 1.6 mg/kg against the standard MRL i.e., 0.5 mg/kg similar findings is also reported by Dogheim et al in Egypt showed that 45 samples were contaminated with the residues of chlorpyrifos and profenofos with the average chlorpyrifos

residues within the range of 2.8 mg/kg.⁵ Among carbamate pesticides, carbaryl residues were detected 0.82 mg/kg (MRL 0.05) in VS-2 (brinjal) and 1.6 mg/kg (MRL 0.5) in VS-7 (tomato) sample, whereas only one sample was contaminated with carbofuran, though some previous studies recorded carbamate residues in different vegetables in different regions of the world.

In case of ladyfinger the quantity of chlorphyrifos of carbaryl in ladyfinger which was collected from research area having the min range was 0.000 and max was 0.101 mg/kg (ppm) with the total concentration of 0.418. Whereas in ladyfinger the carbaryl concentration range was minimum 0.000 and maximum was 0.020, whereas the total concentration was 0.092 mg/kg, similar findings was also reported in Uttar Pradesh tested okra samples and found Chlorpyrifos and Cypermethrin residues above the recommended MRL value which were 5.75 mg/kg and 0.63 mg/kg respectively, against 0.2 mg/kg⁶. The concentration of dichlorvos was found to be 0.005 mg/kg with the mean concentration of 0.000-0.010 mg/kg. These results of the present investigation are supported by findings of a similar investigation 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, which was found to be in very less concentration. Whereas in Green chilli over spraying of pesticides increases the rate of pesticidal residues concentration in chili was found to be 1.507 mg/kg (ppm) with the mean value of (0.000-1.100), whereas the concentration of carbaryl as found to be 0.033 mg/kg with the mean value ranging from (0.000-0.010). It was observed in tomato that concentration of chlorpyriphos in tomato is 1.631 mg/kg with the mean value ranging from (0.000 to 1.000) whereas concentration of carbaryl in tomato was found to be 0.026 mg/kg which ranges from (0.000 to 0.010) mg/kg, whereas malathion residue concentration in tomato was recorded as (1.890) with the mean value of (0.000-0.210) mg/kg. In cabbage the mean value of chlorpyriphos of total 24 sample ranges from (0.000-1.001) mg/kg, while the pesticides concentration was recorded as 1.631 mg/kg, Whereas the mean value of carbayrl was recorded as 0.000-0.010 mg/kg and concentration was found to be 0.026 mg/kg similar finding was in Rajasthan measured a total of 28% samples to be contaminated. He found that residue of monocrotophos was 5.12% above the MRL value and value for chlorpyriphos was 2.56% above the standard MRL.⁷ Farmers should undergo proper training on the appropriate methods for spraying and should have PPE using and pesticides handling techniques for the safe levels of pesticide use prior to spraying of pesticides to the crops. Training and increased awareness programs can help to reduce the level of detected pesticide residues in vegetable farming.

The limitations of study is for certain area of research and also limited to 5 vegetables only, if more vegetables are

checked there are possibilities of increase of maximum residual limits.

CONCLUSION

A multiresidue method was successfully validated for the rapid detection of pesticide residues in different chemical classes in vegetable samples by GC-MS. The high pesticides level in some the samples suggest that these pesticides are being used extensively, which possibly can cause significant health problems, not only to the consumers but also to the farmers who are working in the field. Regular monitoring of the pesticide spraying techniques of farm workers on vegetables cultivation and proper education on the high risks and safe use of pesticides is important. Our study may serve as a basis for the national regulatory authorities to take proper measures to certify that pesticide residues on vegetables are within the acceptable limits for safe consumption and that the occupational exposure is reduced.

With basic research on public perception on risk assessment and analysis would be useful in promoting widespread acceptance and adaption. Role for the public sector in research, product development, pesticides product testing and registration, implementation of pesticide use strategies, and public education about pesticides.

ACKNOWLEDGEMENTS

We are highly thankful to the Department of public health (SHUATS). We are also thankful to Jamia Hamdard (Delhi) for laboratory work and helping out in different ways. We are also thankful to Ministry of Minority Affairs New Delhi for providing financial support without which it would never be possible. A. Khan is a recipient of MANF-SRF fellowship funded by Ministry of Minority Affairs New Delhi India.

Funding: No funding sources Conflict of interest: None declared

Ethical approval: The study was approved by the

Institutional Ethics Committee

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Cite this article as: Khan A, Srivastava JP, Bose DK. The level of residue of pesticides in vegetables by GC-MS, health risk and the negligence in implementation of policies, India. Int J Community Med Public Health 2021;8:407-11.