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Effect of Processing on Reduction of Profenofos and Chlorpyrifos Residues in Tomato Fruits

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ABSTRACT

Experiments were carried out to evaluate the effect of different decontamination processes on reduction of profenofos and chlorpyrifos residues in tomato fruits like washing, cooking, washing plus cooking and dipping in chemical solutions after application of Profex 50EC (profenofos 50%) and Lethal 20EC (chlorpyrifos 20%) individually on the crop. Profex 50EC was applied twice at the rate of 0.8ml/L and Lethal 20EC @ 2.50ml/L at 15 days interval on tomato crop. Tomato fruits were collected at 0 (2 hours) and 3 days interval after the last spray and subjected to decontamination processes. Washing of zero day contaminated fruit samples provided 28.08-69.07% relief from profenofos residues and 39.52-69.21% relief from chlorpyrifos residues. Cooking degraded profenofos residues up to 40.60-71.83% and chlorpyrifos residues by 44.88-58.19%. Washing plus cooking removed profenofos and chlorpyrifos residues up to 75% as compared to other processes and proved to be the best technique in removing the residues. Washing of fruits with 2% NaOH solution reduced the profenofos residues up to 69.07-71.42%, whereas washing with 0.05% solution of HCl reduced the profenofos residues up to 65.30-68.31%. Similarly chlorpyrifos residues were reduced to 69.21-67.33% after treatment with 2% NaOH solution and up to 61.84-65.30% after treatment with 0.05% HCl solution.

Key words: *Tomato, Processing, Profenofos, Chlorpyrifos, Residues*

INTRODUCTION

Vegetables are the inseparable component of Indian cuisine and are consumed throughout the country in different forms and preparations. They are the major source of vitamins and nutrients; hence they fulfill the requirements of our balanced diet[9]. Among the vegetables, tomato (*Solanum lycopersicum* L.) is an important cash crop of Himachal Pradesh which gives better return to the farmers and is infested by a large number of insect-pests and diseases[31, 30, 7]. The key pests of tomato are tomato fruit borer, mites, leaf miner, aphids, whiteflies etc. affecting both quantity and quality of the fruits. In a desperate bid to save the crop farmers sometime apply the pesticide repeatedly and at higher doses hence the repeated and intensive use of insecticides have lead to the development of resistance in insect pests[14].

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In Himachal Pradesh pesticides such as profenofos and chlorpyrifos have been used extensively by the farmers to control these major insect-pests of tomato crop. Since, the effect of insecticides is considered more toxic hence extra care should be taken to reduce the health hazards to the consumers[26]. The application of these pesticides near to harvest can leave residues on the fruits which may be harmful to the consumers[6].

Tomato fruits are picked frequently at short intervals and consumed as cooked or raw; hence chances of carrying pesticide residues to the consumers are more[25]. Hence Pesticide residues in tomato are of major concern to consumers due to their negative health effects. They have been found in both raw and processed fresh produce. There have been various reports suggesting use of different simple household processes in dislodging pesticide residues from food commodities thus making them safe for human consumption[28,2,10,11,13,19,27]. Operations such as Washing, peeling, blanching and cooking play a crucial role in the reduction of residues[13,18]. Each operation has a cumulative effect on the reduction of the pesticides[15].

So in the present scenario it is very important that some pragmatic solution should be developed to tackle this problem of food safety. Food safety is an area of growing concern worldwide on account of its direct bearing on human health. The presence of harmful pesticide residues in food such as tomato has caused a great concern among the consumers. Therefore, the present investigations were contemplated with the objective to study the effect of different decontamination processes in fruits for the reduction of profenofos and chlorpyrifos residues after its application on tomato crop in the field. Hence the techniques used in the present study focused on commercial and home processing of tomato and they included washing alone, washing with chemicals, cooking and washing followed by cooking.

MATERIAL AND METHODS

Chemicals and reagents

Profex 50EC containing 50% profenofos was obtained from M/S Nagarjuna Agrichem Ltd. and reagents like acetone, dichloromethane, hexane, toluene, sodium chloride, sodium sulfate anhydrous (AR grade), Celite 545 were all procured from M/S Merck Specialities, Mumbai. Activated charcoal decolorizing powder was obtained from M/S Darmstadt, Germany. All common solvents were redistilled in an all-glass apparatus before use.

Field trials

Tomato fruits (var. Him Sohna) were raised during 2009 at Entomological Farm, Dr YS Parmar University of Horticulture and Forestry, Nauni, Solan, Himachal Pradesh following recommended agronomic practices (Anonymous, 2009). The experiment was conducted in randomized block design (RBD) with three replications for each treatment.

The first application of Profex 50EC (profenofos 50%) @ 0.8 ml/L and Lethal 20EC (chlorpyrifos 20%) @ 2.50ml/L of water were made at fruit formation stage followed by second application at an interval of 15 days. In control plots, only water was sprayed. Pesticide was sprayed as foliar application in three replications with the help of a knapsack sprayer, fitted with a hollow cone nozzle.

Sampling procedure

Fruit samples (1kg) from each replication were collected randomly at 0 (2 hours after spray) and 3 days intervals after last foliar application. The samples from each replication were collected randomly, packed in bags and brought to the laboratory for processing.

Decontamination Processes

Samples collected from the field were subjected to different decontamination processes viz. washing, cooking and washing followed by cooking[24].

A. Washing

1. Tomato fruits were washed under running tap water and hand rubbed for 2 minutes.
2. Samples were dipped in lukewarm water (50^0C) for 5 minutes and then, placed on filter papers for drying.
3. Samples were dipped in 2% NaCl (w/v) solution for 5 minutes followed by tap water washing.

4. Samples were dipped in 2% lukewarm salt solution (w/v) for 5 minutes followed by water washing.
5. Samples were dipped in 0.05% HCl (v/v) for 5 minutes, followed by water wash.
6. Samples were dipped in 2% (w/v) sodium hydroxide solution for 5 minutes, followed by washing with water.

B. Cooking

1. Open pan cooking: Unwashed samples from each replication were chopped and put in an open pan of 1 litre capacity containing 500 ml water and boiled till softness (10-15 minutes).
2. Steam cooking: Samples were chopped and steamed for 5 minutes in a pressure cooker.
3. Microwave cooking: Samples were kept in microwave for 5 minutes for cooking at 1400 W power output.

C. Washing followed by cooking

1. Washing+cooking: Samples were first washed by hand rubbing under a stream of running tap water for 2 minutes, followed by boiling in an open pan of 1 litre capacity containing 500 ml water till soft (10-15 minutes).
2. Washing+steam cooking: Samples were washed under running tap water and steamed for 5 minutes in a pressure cooker.
3. Washing+microwave cooking: Samples were first washed under the tap water and then, placed in microwave for 5 minutes for cooking at 1400 W power output.

After completing decontamination process, samples were extracted and cleaned up according to the method of Sharma[29].

Extraction and cleanup

The samples were processed and analyzed at the Pesticide Residue Analysis Laboratory, Department of Entomology, Dr YS Parmar University of Horticulture and Forestry, Nauni, Solan, Himachal Pradesh. Processed tomato fruits samples were homogenised in a domestic mixture.

A representative 100 g homogenised sample was taken up with 200 ml acetone in a 500 ml conical flask and kept for overnight. The extract was filtered through Buchner funnel by fitting a Whatman No. 1 filter paper. An aliquot of 60 ml (30 g equivalent) of sample was transferred to 1 litre separatory flask and extracted with 200 ml mixture of hexane and dichloromethane (1:1, v/v). The lower aqueous phase was transferred to another 1 litre separatory funnel containing ten millilitre saturated sodium chloride solution and partitioned twice with 100 ml dichloromethane. Lower aqueous phase was discarded and upper organic phase was pooled with first organic fraction. Pooled organic phase was passed through anhydrous sodium sulfate and evaporated to dryness at 45^0C by using vacuum rotary evaporator. Finally, the residues were taken up in 3 ml (1+2) acetone for cleanup. Profenofos and chlorpyrifos samples were cleaned up on charcoal column. Two millilitres of sample fraction of each was loaded in a charcoal column which was prepared by placing one inch layer of Celite 545, 6 g adsorbent mixture (1:4 w/w Charcoal:Celite 545) and then, overlaid with 2 g sodium sulfate. The column was eluted with 200 ml of 2:1 acetone: dichloromethane mixture. Eluant was evaporated to dryness, residues were dissolved in 2 ml n-hexane and 1µl was injected into a gas chromatograph.

Residue estimation

Residues of profenofos and chlorpyrifos were estimated by using Gas-Chromatograph (Agilent 6890N) having ECD detector and DB-5 Ultra Performance Capillary column (Cross-linked Methyl Silicon, length 30 m, 0.25 mm internal diameter with 0.25 µm film thickness). Oven temperature was programmed as: 100^0C for 1 minute, 30^0C/minute up to 150^0C, 3^0C/ minute up to 205^0C and finally 260^0C at rate of 10^0C/ minute. Injection port and electron capture detector (ECD) temperature were kept at 250^0C and 300^0C, respectively.

Profenofos and chlorpyrifos residues (mg/kg) were determined for each replication and then mean residues were calculated. Per cent relief from residues in each treatment was calculated from the mean residues, by the following equation:

$$\% \text{ relief} = 100 - \left(\frac{\text{Residue in processed sample (mg/kg)}}{\text{Residue in unprocessed sample (mg/kg)}} \times 100 \right)$$

Validation of Analytical Method

Unprocessed samples from untreated plot were spiked with profenofos insecticide at 0.05, 0.10, 0.20, 0.50 and 1.00 mg kg^{-1} and for chlorpyrifos it was 0.01, 0.05, 0.10, 0.50 and 1.00 mg kg^{-1} fortified levels. Data presented in Table 1 depicts reliability of analytical method, as the recovery of insecticides was above 86 per cent. Recovery of profenofos was between 88.00-93.00% with relative standard deviation (RSD) of 0.034-0.870% and for chlorpyrifos recovery was between 90.00-94.00% with RSD 0.021-1.007% (Table 1).

Table 1: Recovery of profenofos and chlorpyrifos from tomato fruits

Insecticides	Tomato fruits		
	Fortification level, (mg/kg)	**Mean recovery (%)**	**Relative standard deviation (%RSD)**
Profenofos	0.05	88.00	0.870
	0.10	86.00	0.349
	0.20	89.00	0.086
	0.50	91.00	0.034
	1.00	93.00	0.038
Chlorpyrifos	0.01	90.00	1.007
	0.05	90.00	0.934
	0.10	91.00	0.335
	0.50	92.00	0.107
	1.00	94.10	0.021

RESULTS AND DISCUSSION

Effect of Washing

Washing is the most common form of processing which is a preliminary step in both household and commercial preparation. Loosely held residues of several pesticides are removed with reasonable efficiency by varied types of washing processes[33]. Washing of 0 day sampled tomato fruits under running tap water provided 28.08% relief from profenofos residues whereas 39.18% relief was observed in 3 day old samples (Figure 1). Similar observations were recorded after washing of tomato fruits treated with chlorpyrifos (Figure 2). Aktar *et al*.,[3] reported that washing of cabbage head under running tap water removed 27.72-32.48% quinalphos residues which are in accordance with my findings. Similarly, Singh *et al*.,[32] also found that washing of okra fruits with tap water could remove the residues of cypermethrin to the extent of 36.25-42.76%. The initial diazinon residue level (0.822 ppm) on cucumbers was decreased by 22.3% by washing for 15 seconds rubbing under running water[8].

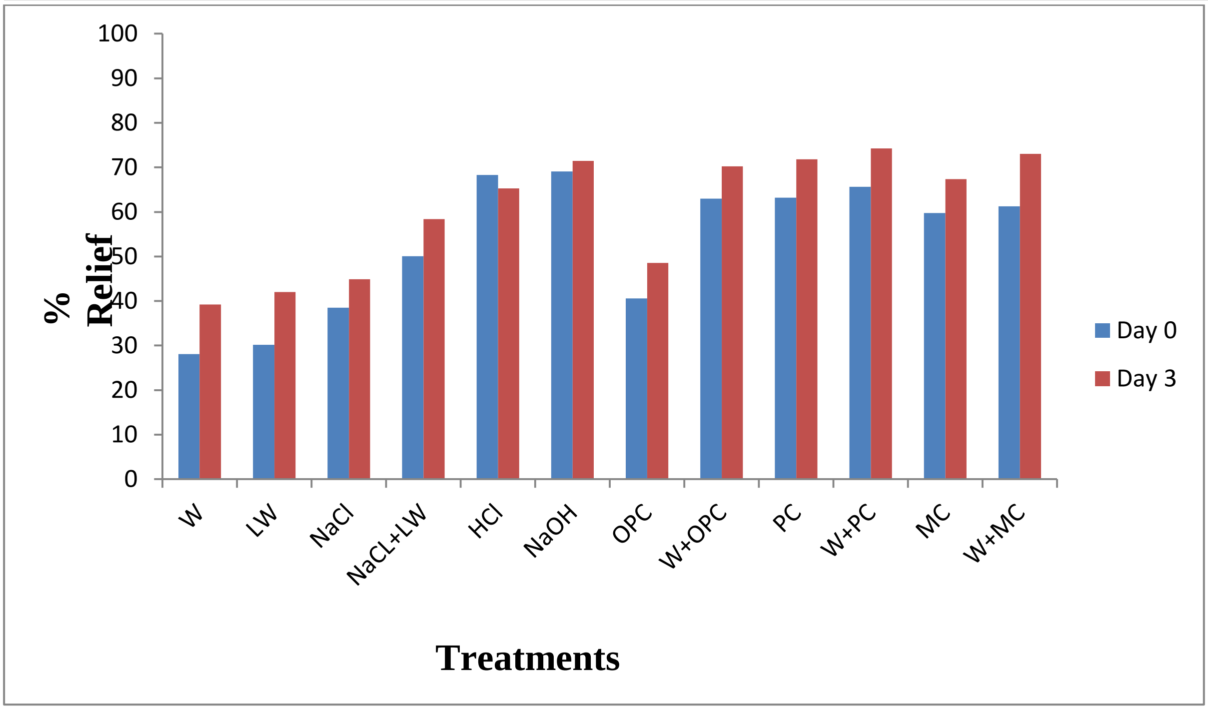

Fig. 1: Per cent relief from profenofos residues from different decontamination processes (W= Tap water washing, LW= Luke warm, OPC=Open pan cooking, PC=Pressure cooking, MC=Microwave cooking)

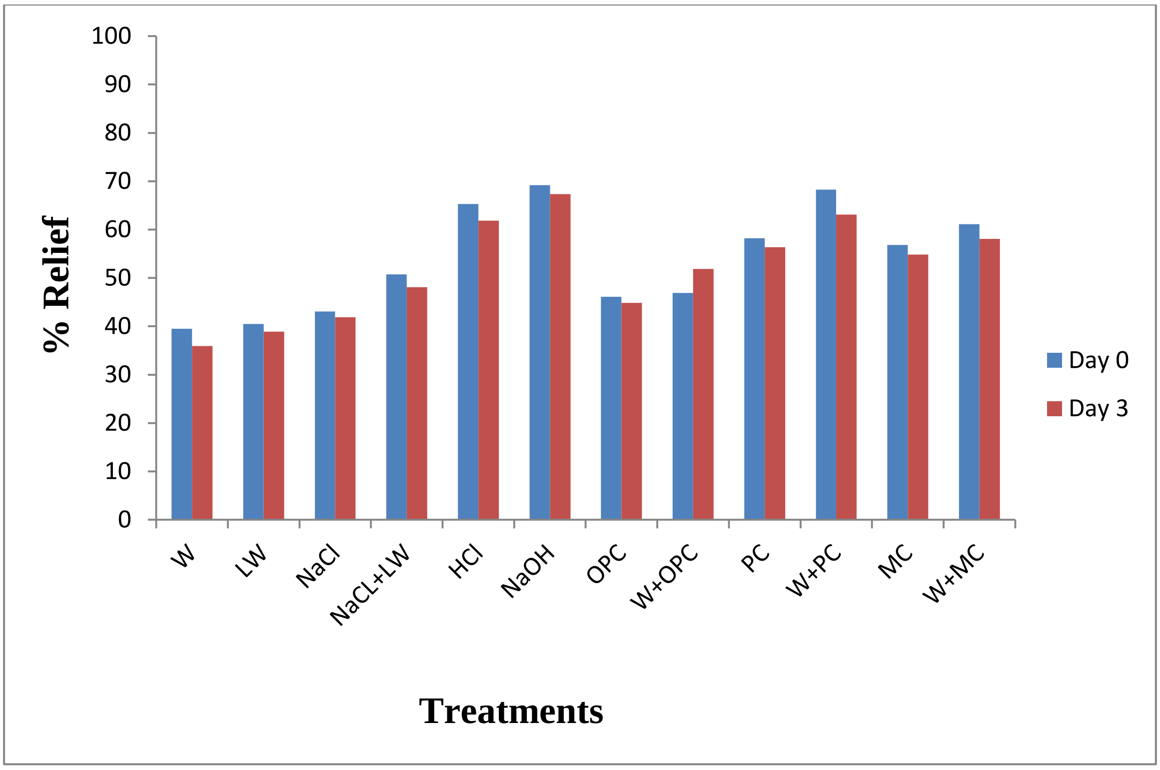

Fig. 2: Per cent relief from chlorpyrifos residues from different decontamination processes (W= Tap water washing, LW= Luke warm, OPC=Open pan cooking, PC=Pressure cooking, MC=Microwave cooking)

Lukewarm water washing of 0 day sampled tomato fruits provided 30.17% relief from profenofos residues whereas 42.04% relief was observed in 3 days old samples (Fig. 1). Similar observations were recorded after washing of tomato fruits treated with chlorpyrifos (Fig. 2) which are in accordance with Kanta *et al.*,[17] who reported 7-38 per cent reduction of *alpha*-cypermethrin residues by lukewarm water washing of cauliflower curds.

Kumari[20] also reported 32-100 per cent reduction of OP's insecticide residues by lukewarm water of cauliflower.

Chemical washing

Washing of treated tomato fruits with sodium hydroxide and hydrochloric acid provided a good relief from profenofos and chlorpyrifos residues in comparison to washing with sodium chloride and lukewarm sodium chloride solution. It may be due to hydrolytic property of profenofos and chlorpyrifos in strong acids and alkalies[34]. Sodium hydroxide provided 71.42% and 69.21% relief from profenofos and chlorpyrifos, respectively. Dip treatment of cauliflower curds with hydrochloric acid gave 68.31% relief from profenofos and 65.30% from chlorpyrifos residues. The present findings are in agreement with Patyal *et al*.,[24] who found that washing of treated apple fruits with 2% (w/v) NaOH and 0.05% (v/v) HCl gave 77.06 and 75.96%, relief respectively from endosulfan residues.

Dipping of tomato fruits samples in 2% sodium chloride solution (w/v) reduced profenofos and chlorpyrifos residues to 44.89% and 43.07% whereas lukewarm sodium chloride solution reduced residues to 58.36% and 50.72% respectively which is in agreement with the findings of Mukherjee *et al*.,[21] also observed that dipping of cauliflower curds in 1% brine solution followed by washing reduced the residues by 39.6% while in case of hot 1% brine solution, the reduction was 55.0%.

Cooking

Application of heat to the food commodities is commonly done through ordinary cooking, pressure cooking, microwave cooking, frying, sterilization and canning.

The effect of different cooking processing on removal of profenofos and chlorpyrifos residues in tomato was studied (Figure 1, 2). In all of the processes, cooking with pressure cooking was found to be more effective than in others. Pressure cooking reduced the residues up to 71.83%. These results are in accordance with the findings of Muthukumar *et al*.,[22] who also reported that pressure cooking was the most effective in reducing both α- and β-endosulfan by 64.59% and 61.60% as compared to boiling and microwave cooking.

Cooking of tomato fruits in open pan or under pressure or in the microwave resulted in 42-72% relief from profenofos and chlorpyrifos residues. The findings are in agreement with Dikshit[12] who observed that process of steaming dislodged the cypermethrin residues by 63-74% on stored pulses treated at 3 and 5 mg/kg levels. The disappearance of pesticide residues from boiling extract could be due to decomposition by the effect of heat, the stronger adsorption of pesticide onto plant tissues and or/the poor solubility of pesticides in water[1,4]. Walia *et al*.,[35] reported that microwave cooking reduced cypermethrin residues to the extent of 40.89 per cent in brinjal sprayed at 0.001 per cent concentration. Hence, processes involving heat can increase volatilization, hydrolysis or other chemical degradation and thus, reduce residue levels[16].

Washing followed by cooking

Washing is generally the first step in various types of treatments which are given to food commodities in combinations like washing followed by cooking, washing and drying, washing and peeling and washing, peeling and juicing to allow for effective decontamination from pesticides[18]. Washing of tomato fruits followed by cooking lead to more than 75% removal of profenofos and chlorpyrifos residues (Figure 1, 2). Similarly, Mukherjee *et al*.,[21] also reported that washing of cauliflower heads under running tap water removed 27.9% chlorpyrifos residues, cooking reduced residues to 41.4% and washing+cooking further reduced residues to 66.7%. Aktar *et al*.,[3] also reported that washing plus cooking of cabbage heads reduce more quinalphos residues (66.45-68.19%) in comparison to washing alone (41.30-45.20%).

CONCLUSION

A critical analysis of whole decontamination data revealed that the washing plus pressure cooking removed much higher residues from contaminated fruits as compared to the simple washings. Although, sodium hydroxide and hydrochloric acid treatments were superior over all other decontamination processes but such treatments can be used in the industries where large quantity of vegetables are processed for decontamination. Washing of vegetables with water followed by pressure cooking removed maximum residues up to 79% as compared to the other processes and proved good household practice.

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