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Persistence of Metalaxyl and Mancozeb on Potato Leaves and Their Residues in Tubers

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Abstract: The persistence of fungicides on two commercial cultivars of potato was determined under field conditions at Punjab Agricultural University, Ludhiana, Punjab. Initial deposits of mancozeb on potato leaves were found to be 26.9 and 38.7 mg kg^{-1}, following application of ready mixture of fungicide metalaxyl 8% + mancozeb 64% (Ridomil MZ) at the rate of 1260 and 2520 g a.i. ha^{-1}, whereas metalaxyl residues were found to be 35.1 and 49.5 mg kg^{-1}, respectively. The residue level of mancozeb in potato leaves 15 days after application at single and double doze were 19.0 and 27.0 mg kg^{-1} showing a loss of 29.6 and 30.3%, whereas the values for metalaxyl at single and double doze were 0.40 and 0.80 mg kg^{-1} showing a loss of 98.9 and 98.4%, respectively. Residues of mancozeb and metalaxyl were not detected at 0.04 and 0.02 mg kg^{-1} level in potato tubers at harvest (PHI = 53 days) at both the dosages, respectively. The persistence and dissipation of mancozeb with the application of Ridomil MZ followed similar trend as in Indofil M-45. The rate of fungicide dissipation increased with time after application in both the potato cultivars 'Kufri Chandramukhi' and 'Chipsona'. No significant difference was observed on initial deposit, persistence and dissipation of the two molecules between the two potato cultivars.

Key words: Fungicide, mancozeb, metalaxyl, persistence, potato, residues

INTRODUCTION

Potato (*Solanum tuberosum* L.) is one of the most important vegetable crops widely grown in all agro-climatic zones of the world. It enjoys the status of staple food in most of the developed/developing countries. Nutritionally, potatoes are best known for carbohydrate content, besides vitamins. Its protein has very high biological value because all amino acids are present in comparatively large quantities and have a balanced ratio of protein to carbohydrate energy (Carputo *et al*., 2005). Potatoes are increasingly used as processed food. Because of high production potential and superior nutritional quality, potato is a useful tool to fight against hunger and malnutrition in developing and populous country like India. China and India are large potato producing countries and rank first and third with a production share of 22.7 and 7.8%, respectively. The potato production in India has increased by almost 850% between 1960 and 2000, partly in response to growing demand from higher-income urban populations. Since 1990, per capita consumption has risen from around 12 kg to 17 kg a year (Anonymous, 2008a). In India potato is grown over an area of 1401.4 thousand ha with annual production of 23.51 lakh metric tonnes (Anonymous, 2008b). The most important and commercial potato belt in India is the Indo-Gangetic plains including the states of Punjab, Haryana and Utter Pradesh. Potato crop is prone to a large number of fungal, bacterial and viral diseases. Of these late blight caused by *Phytophthora infestans* (Mont) de Bary is the dreadful disease responsible for causing huge economic losses to the potato crop (Bhattacharyya *et al*., 1990; Sokhi *et al*., 1993). Punjab experiences late blight epidemics after every 4 to 5 years resulting in significant crop losses (Thind and Mohan, 1998; Thind *et al*., 2004). Mancozeb sprays are effective in managing late blight of potato. The protective sprays of contact fungicides are usually applied before the disease

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appearance. Even under heavy disease situation, premixed preparations of non-systemic and systemic fungicide containing metalaxyl give good disease control (Anonymous, 2008c). It is an established fact that fungicides in general and mancozeb in particular are highly detrimental to human health, if consumed above Maximum Residual Limit (MRL). However, no information is available on the residue levels of mancozeb and metalaxyl in potato leaves and tubers under Indian conditions. So studies were undertaken to determine the residue level of both mancozeb and metalaxyl in potato tubers when different fungicidal spray schedule are adopted for the management of potato late blight.

MATERIALS AND METHODS

Technical grade mancozeb (98.0% purity) was supplied by Indofil Chemicals Company, Mumbai while metalaxyl (98.1% purity) was supplied by Syngenta India Ltd. Mumbai. A field experiment was conducted during 2007 and 2008 to study the persistence of fungicides applied against late blight of potato. The crop was raised in plots of 8x8 m as per standard agronomic practices. The samples of leaves sprayed with ready mixture of fungicide metalaxyl 8% + mancozeb 64% (Ridomil MZ) at the rate of 1260 and 2520 g a.i. ha^{-1}, were collected at 0, 3, 6, 9, 12 and 15 days after spray. The potato tubers were collected 20 days after last fungicide spray and at the time of harvest.

The residues of Ethylene Bis-dithiocarbamate (EBDC) were analyzed by standard colorimetric method using CS_2 evolution method (Keppel, 1971). To a series of 25 mL volumetric flasks, varying amount of standard CS_2 solution (0.1-6.0 mL) was added by using graduated pipette. To each flask 15 mL colour reagent was added and diluted to the mark with ethanol. The flasks were allowed to stand for 15 min. and absorbance recorded at 435 nm against blank containing 15 mL colour reagent and 10 mL ethanol. The absorbance was plotted against standard concentration of CS_2 to obtain standard curve (Fig. 1).

The representative potato leaves/tubers were ground and 50 g homogenized sample was taken in 250 mL conical flask. To this 100 mL disodium salt of 1N Ethylene Dinitrilo-tetra-acetate (EDTA) solution was added. In first trap, 10 mL 10% NaOH solution was taken and 5 mL ethyl acetate was added to it. In second trap 15 mL chromogenic reagent was added. Procedure of standard protocol mentioned above was followed to get the CS_2 absorbed ethyl estate solution which was determined against a reference solution on spectrophotometer at 435 nm. CS_2 (µg) obtained was determined from standard

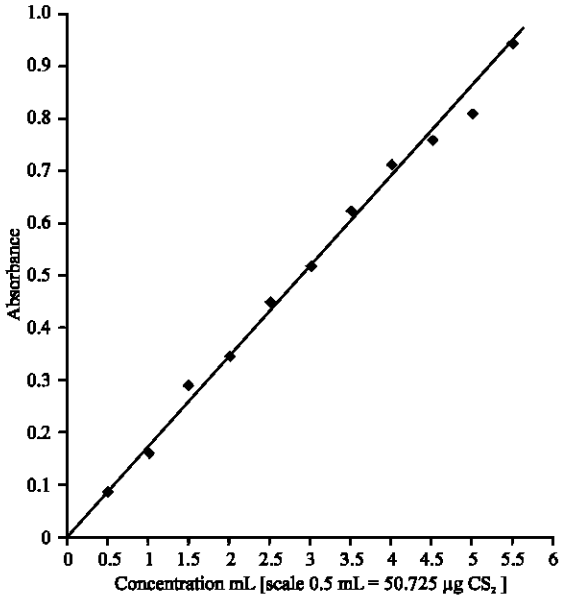

Fig. 1: Standard curve of Absorbance against µg CS_2 for determination of mancozeb residues

curve against absorbance recorded. The amount (µg) of mancozeb present in the sample was calculated using the formula:

$$\text{Amount of mancozeb (µg)} = \frac{\text{µg } CS_2 \text{x g mol. wt of mancozeb}}{2\text{x g mol. wt of } CS_2}$$

$$= \frac{\text{µg } CS_2 \text{x} 266.575}{2\text{x} 76.14}$$

$$= \text{µg } CS_2 \text{ x} 1.75$$

Mancozeb residues (mg kg^{-1}) were calculated by dividing µg mancozeb obtained by the total sample size.

Moisture free clear extract from ten grams of macerated leaf samples/(50 g for potato tubers) so obtained after separation by 75 mL dichloromethane and twice with 75 mL distilled hexane was filtered through Whatman filter paper No. 1. It was concentrated to near dryness and added about 20 mL hexane and again concentrated using rotary vacuum evaporator at 30°C. The process was repeated to completely evaporate dichloromethane and the final volume reconstituted to about 5 mL using distilled hexane.

The estimation of metalaxyl residues was done by Gas Liquid Chromatograph (GLC) equipped with Nitrogen-phosphorus Detector (NPD) and a glass column (100x2 mM inner dia.) filled with 3% DC 200 on 100-120 mesh Gas Chrom Q. The operating conditions maintained were: detector temperature, 300°C; oven

Table 1: Recovery of mancozeb and metalaxyl in potato leaves and potato tubers

	Mancozeb		Metalaxyl	
Substrate	Level of fortification (mg kg^{-1})	Mean±S.D. (%)	Level of fortification (mg kg^{-1})	Mean±S.D. (%)
Potato tubers	0.04	83.67±4.16	0.02	79.60±0.65
	0.50	84.33±3.05	0.50	80.16±4.75
	1.00	85.00±2.00	1.00	82.88±3.00
Potato leaves	1.00	82.29±2.29	1.00	82.29±2.29
	5.00	84.00±4.09	2.50	80.55±0.95
	10.00	80.25±1.64	5.00	83.49±6.58

(column) temperature, 250°C; injector temperature, 270°C; carrier gas (N_2) flow rate, 30 mL min^{-1}; air flow, 145 mL min^{-1} and hydrogen flow, 3 mL min^{-1}. Before use, the column was primed with several injections of standard mixture of metalaxyl till a consistent response was obtained. Suitable aliquot of cleaned up samples were then injected and the compound in sample identified and quantified by comparison of retention time and peak heights of sample chromatograms with that of standard run under identical operating conditions.

Recovery experiments were carried out to know the efficiency of analytical method used. Samples of leaves and tubers from control plots were spiked with metalaxyl and mancozeb at level of 0.02 to 1.0 and 0.04 to 1.0 mg kg^{-1}, respectively (Table 1). These were extracted, cleaned-up and analyzed following the method already described. The control samples from untreated plots and reagent blanks were also processed in the same way so as to find out the interferences, if any, due to the substrate and reagents, respectively.

RESULTS AND DISCUSSION

The mean initial deposits of mancozeb on leaves of potato were 26.9 and 38.7 mg kg^{-1} after the application of ready mixture of fungicide metalaxyl 8% + mancozeb 64% (Ridomil MZ) at the rate of 1260 and 2520 g a.i. ha^{-1}, respectively. Three days after spray the residues dissipated to a mean level of 24.7 mg kg^{-1} thereby showing a loss of 8.2% at single dose and corresponding values at double dose was 36.1 mg kg^{-1} with 6.8% loss. The residue levels at the end of 6, 9, 12 and 15 days after spray were 22.4, 21.8, 20.8 and 19.0 mg kg^{-1}, respectively, whereas at double the dosage, the corresponding values were 34.7, 30.2, 28.1 and 27.0 mg kg^{-1} (Table 2). The dissipation of mancozeb was non-linear with respect to interval after spray at both the concentrations. Similar results were observed by Sharma *et al.* (1994) regarding deposition and dissipation of mancozeb in vegetable crops. In knol-khol the residues were low in heads as compared to leaves and persisted even after 30 days.

The dissipation of metalaxyl in potato leaves revealed that the average initial deposits of metalaxyl on potato

Table 2: Residues of mancozeb (mg kg^{-1}) following application of ready mixture of fungicide metalaxyl 8% + mancozeb 64% (Ridomil MZ) at the rate of 1260 and 2520 g a.i. ha^{-1} on potato leaves at different intervals under field conditions (average of 2 years data)

	Dosage			
	0.25%		0.505%	
Days after treatment	Mean*±SD (mg kg^{-1})	% dissipation	Mean*±SD (mg kg^{-1})	% dissipation
0	26.9±0.14		38.7±0.09	
3	24.7±0.14	8.17	36.1±0.07	6.76
6	22.4±0.22	16.7	34.7±0.06	10.3
9	21.8±0.09	19.0	30.2±0.05	22.0
12	20.8±0.11	22.9	28.1±0.06	27.4
15	19.0±0.07	29.6	27.0±0.05	30.3

*Mean of three replications

Table 3: Residues of metalaxyl (mg kg^{-1}) post-application of ready mixture of fungicide metalaxyl 8% + mancozeb 64% (Ridomil MZ) at the rate of 1260 and 2520 g a.i. ha^{-1} on potato leaves at different time intervals under field conditions (average of 2 years data)

	Dosage of fungicide			
	0.25%		0.50%	
Days after treatment	Mean*±SD (mg kg^{-1})	% dissipation	Mean*±SD (mg kg^{-1})	% dissipation
0	35.1±0.18		49.5±0.41	
3	16.0±0.13	54.6	19.6±0.46	60.3
6	6.0±0.18	82.9	8.1±0.08	83.6
9	2.1±0.14	93.9	3.9±0.05	92.0
12	0.4±0.01	98.9	0.8±0.02	98.4
15	BDL		BDL	

*Mean of three replications; Below Detectable Level (BDL) <0.02 mg kg^{-1}

leaves were 35.1 and 49.5 mg kg^{-1}, respectively, following the application of ready mixture of fungicide metalaxyl 8% + mancozeb 64% (Ridomil MZ) at the rate of 1260 and 2520 g a.i. ha^{-1} (Table 3). More than 98% metalaxyl residues dissipated after 15 days of the spray at both the dosages.

The persistence of metalaxyl followed similar trend as that of mancozeb, except that its dissipation with time was much higher. Milgroom and Fry (1988) observed that metalaxyl residues decreased rapidly in the first two days after application and thereafter the decrease was quite slow (Table 4). They further observed that rainfall had no effect on the efficacy of metalaxyl in controlling the disease Singh and Pundhir (2004) observed only 15% metalaxyl residues after 15 days of application. Thind *et al.* (2004) observed the persistence of Ridomil MZ for more than 10 days on one month old potato plants as compared to Dithane-M 45 which lasted for 5 days only.

The fungicide residues in potato tubers cv. 'Kufri Chandramukhi' were determined 20 days after spray and at harvest (PHI = 53 days). The mancozeb residues were below the detectable level of 0.04 mg kg^{-1} in potato tubers harvested 20 days after spray and harvested at full

Table 4: Residues of metalaxyl and mancozeb (mg kg^{-1}) in potato tubers of cv. *Kufri Chandramukhi* (average of 2 years data)

		Residue			
		20 days after last spray		At harvest (PHI = 53)	
Treatment schedule	Disease* severity (%)	Mancozeb*	Metalaxyl*	Mancozeb*	Metalaxyl*
Ridomil MZ 1260 g a.i. ha^{-1}	0	BDL	0.15±0.01	BDL	BDL
Ridomil MZ 2520 g a.i. ha^{-1}	0	BDL	0.28±0.04	BDL	BDL
Control	65	BDL	BDL	BDL	BDL

Below Detectable Level (BDL) <0.02 mg kg^{-1} for metalaxyl and <0.04 mg kg^{-1} for mancozeb*Mean of three replications, PHI = Pre-harvest interval

maturity. However, in tubers harvested 20 days after spray the mean residues of metalaxyl in potato tubers were 0.15 and 0.28 mg kg^{-1} at single and double the dosages, respectively. The residues found at this stage were much higher than the MRL (0.05 mg kg^{-1}) prescribed by the Codex (Anonymous, 2001). However, residue of metalaxyl was below the detectable limit of 0.02 mg kg^{-1} in potato tubers harvested at maturity. Singh (1998) observed that the extent of metalaxyl residues in potato tubers was more at 5 days than at 20 days after harvest, which was drastically reduced at 40 days after harvest. They observed that higher dose of fungicide resulted in increased residue levels.

CONCLUSION

The dissipation of metalaxyl in potato leaves revealed that the average initial deposits of metalaxyl on potato leaves were 35.1 and 49.5 mg kg^{-1}, respectively, following the application of ready mixture of fungicide metalaxyl 8% +mancozeb 64% (Ridomil MZ) at the rate of 1260 and 2520 g a.i. ha^{-1}. More than 98% metalaxyl residues dissipated after 15 days of the spray at both the dosages. The persistence of metalaxyl followed similar trend as that of mancozeb, except that its dissipation with time was much higher. The dissipation of mancozeb was non-linear with respect to interval after spray at both the concentrations. However, no significant difference was observed on initial deposit, persistence and dissipation of the tow fungicides among the two potato cultivars. Metalaxyl residue of 0.13 to 0.28 mg kg^{-1} was detected in potato tuber 20 days after last foliar spray on potato crop, however, no metalaxyl residue was detected at harvest time. The mancozeb residue could not be detected in potato tubers before and at the time of harvest. Likewise, residues of mancozeb and metalaxyl was below detectable limits in potato tubers of different cultivars harvested from farmer's fields.

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