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Evaluation of azoxystrobin (Amistar 25 SC) against early leaf blight and leaf spot diseases of tomato

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Spraying of azoxystrobin at various doses *viz*., 31.25, 62.50 and 125 g a.i. ha^{-1} revealed that 125 g a.i. ha^{-1} (500 ml ha^{-1}) recorded only 3.90 and 4.86 per cent disease index (PDI) of leaf blight and 0.00 and 2.42 PDI of leaf spot and the same treatment also recorded the higher yield of 27.60 and 26.30 tonnes ha^{-1} in the first and second season, respectively. No phytotoxic effect of azoxystrobin was observed in both the field trials of tomato even at four times the recommended doses of 125 g a.i. ha^{-1}. The persistence of azoxystrobin at 250 and 500 g a.i. ha^{-1} was observed upto seven days after last spraying. However, the persistence of azoxystrobin at 31.25, 62.50 and 125 g a.i. $ha^{-1}$was observed upto three to five days after last spraying. The safe waiting period for the harvest of tomato fruits was 0.61 and 0.17 days in the first and second field trial, respectively at azoxystrobin 125 g a.i. ha^{-1} (optimal dose).

Key words: *Alternaria solani*, azoxystrobin, *Septoria lycopersici*, residues, tomato

Introduction

Tomato (*Lycopersicon esculentum* Mill.), an important commercial crop is grown in an area of about 0.52 million ha with a production of 7.4 million tonnes in India (Anonymous, 2005) The major constraint to tomato production in India is early leaf blight and leaf spot caused by *Alternaria solani* [(Ell. and Mart.) Jones and Grout] and *Septoria lycopersici* Speg., respectively. Frequent sprays of copper containing (Bordeaux mixture and copper oxychloride) fungicides and certain other group of fungicides are required to check the diseases, which increase the cost of cultivation besides posing residue problem. Hence, newer fungicides are needed for leaf blight and leaf spot disease

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management in tomato. Azoxystrobin (Amistar 25 SC) possess a novel biochemical mode of action and its fungicidal activity results from the inhibition of mitochondrial respiration in fungi. This is achieved by the prevention of electron transfer between cytochrome b and cytochrome c. Because of its novel mode of action, azoxystrobin is effective against pathogens which have developed reduced sensitivity to other fungicides (Hewitt, 1998). Azoxystrobin exhibits no cross–resistance to the ergosterol biosynthesis inhibitors, phenylamides, dicarboximides and benzimidazole class of fungicides. Azoxystrobin shows a unique spectrum of disease control and is active against Oomycetes, Ascomycetes, Basidiomycetes and Deuteromycetes. No current commercial fungicide combines this breadth of spectrum with high levels of intrinsic activity at low rates. The present study was undertaken to study the bioefficacy, phytotoxicity and persistence of the newer fungicide azoxystrobin against tomato leaf blight and leaf spot diseases.

Materials and methods

Source of fungicides

The chemicals *viz*., azoxystrobin, mancozeb and carbendazim were obtained from M/S Syngenta Pvt. Ltd., India.

Bioefficacy of azoxystrobin

A field experiment was conducted during March–June, 2004 in the farmer's holding at Kaveripattinam, Krishnagiri Tamil Nadu, India with the variety PKM-1 tomato to study the bioefficacy of azoxystrobin against leaf blight and leaf spot diseases. The experiment was laid out in randomized block design with four replications with a plot size of 5 x 4 m (20 m^2). Regular agronomic practices were followed as per the Tamil Nadu Agricultural University crop production guide. The treatments of the experiment were T_1 - Azoxystrobin 25 SC @ 31.25 g a.i. ha^{-1}, T_2 -Azoxystrobin 25 SC @ 62.50 g a.i. ha^{-1}, T_3 -Azoxystrobin 25 SC @ 125 g a.i. ha^{-1}, T_4 - Mancozeb @ 1kg ha^{-1}, T_5 - Carbendazim @ 500 g ha^{-1} and T_6 – Control. Two rounds of sprays were given after 40 days after transplanting using a high volume ASPEE backpack sprayer with a spray fluid volume of 500 l ha^{-1} at 15 days interval. The disease incidence was recorded on 7 and 15 days after each spray. The intensity of early blight and leaf spot diseases was assessed with the score chart of 0 to 9 scale (0-No infection, 1-0 to10, 3-10.1 to15, 5-15.1 to 25, 7-25.1 to 50 and 9-More than 50 per cent leaf area affected) (Babu, 1994) The per cent disease index (PDI) was calculated with the following formula (Mckinney, 1923).

$$\frac{\text{Sum of numerical ratings}}{\text{Total number of leaves observed}} \times \frac{100}{\text{Maximum disease grade in the score chart}}$$

Another field trial was conducted during July–Oct, 2004 in a farmer's field at Vadivelampalayam, Coimbatore, Tamil Nadu, India using PKM-1 tomato variety in the same way to confirm the results obtained in the field experiment I.

Yield

The weight of fruits from each plot during harvest was recorded and the average yield per treatment was calculated.

Phytotoxic effect of azoxystrobin

A field experiment was conducted on PKM-1 tomato to study the phytotoxic effect of azoxystrobin during March–June, 2004 in the farmer's holding at Kaveripattinam, Krishnagiri, Tamil Nadu, India. The experiment was laid out *vide* bioefficacy trial. The treatments of the experiment were T_1 - Azoxystrobin 25 SC @ 31.25 g a.i. ha^{-1}, T_2 - Azoxystrobin 25 SC @ 62.50 g a.i. ha^{-1}, T_3 - Azoxystrobin 25 SC @ 125 g a.i. ha^{-1}, T_4 - Azoxystrobin 25 SC @ 250 g a.i. ha^{-1}, T_5 - Azoxystrobin 25 SC @ 500 g a.i. ha^{-1} and T_6 – Control.

Method of assessment

The crop was observed on 1, 3, 5, 7, 10 and 20 days after spraying for the phytotoxic symptoms such as injury to leaf tips, leaf surface, wilting, vein clearing, necrosis, epinasty and hyponasty. Leaf injury was graded based on visual rating on a 1-10 scale (1-1 to 10; 2-11 to 20; 3-21-30; 4-31 to 40; 5-41 to 50; 6-51 to 60; 7-61 to 70 ; 8-71 to 80; 9-81 to 90; 10-91 to 100 per cent leaf injury) (CIB, 1989). Another field trial was laid out at Vadivelampalayam, Coimbatore, Tamil Nadu, India during July–Oct, 2004 with PKM-1 tomato in the same way to confirm the results obatined in the field experiment I.

Persistence and harvest time residues of azoxystrobin

Two field experiments were conducted to determine the persistence of azoxystrobin in tomato fruits. The field experiment was conducted during March–June, 2004 and July–Oct, 2004 in the farmer's holding at Kaveripattinam and Vadivelampalayam, Tamil Nadu, India, respectively. The treatments of the experiments were as given above.

Analytical methodology

Sampling

Fruit samples were collected from all the concentrations of azoxystrobin treated plots and untreated control plots after last round of spraying to determine the harvest time residues. Samples were collected for dissipation studies at 0 (1h after spray), 1, 3, 5, 7, 10 and 14 days after application. Fruits (500 g each) were collected from each replication, pooled and after quartering, 25 g of laboratory analytical samples in duplicates were drawn in wide mouth containers having extraction solvent, acetonitrile : doubled distilled water (9:1v/v). The working samples were transported in an ice box and stored at -70 ºC in a deep freezer in the laboratory.

Extraction

The laboratory samples were homogenized with acetonitrile : water (9:1v/v). The extract was filtered under vacuum through a buchner funnel over laid with Whatman No. 1 filter paper into a round bottom flask. For further extraction, the residues were washed with the same solvent. All the aliquots were evaporated to near dryness on rotary evaporator <40 ºC and redissolved in dichloromethane : ethyl acetate mixture (95:5) for silica gel column clean up.

Clean up

For column chromatography, 1.5 cm (dia) x 50 cm (length) glass columns were used. The drip tip of the columns were plugged with cotton wool and packed to 6 cm height with activated silica gel sandwitched between 2 cm height layers of anhydrous sodium sulphate on either side. The packed column was prewetted with dichloromethane. To elute the compound, 25 ml of dichloromethane and ethyl acetate (7:3 v/v) was used after loading the condensed extract. Eluate was concentrated to near dryness and the residue was redissolved in 5-10 ml of HPLC grade acetonitrile for final determination using HPLC, Hitachi model L 6200 with the following operating parameters.

Mobile phase	**Acetonitrile (HPLC grade): water (HPLC grade) (80:20 v/v)**
Column	ODS 2
Flow rate	1 ml min^{-1}
Wave length	245 ήm
Quantity injected	20 µl (fixed loop)
Attenuation	3

The amount of residue present in the fruits was calculated by comparing the sample response with the response of standard by using the formula:

$$\text{Residue (ppm)} = \frac{\text{Sample peak height (cm)}}{\text{Standard peak height (cm)}} \times \frac{\text{Weight of standard (ηg)}}{\text{Weight of sample (g)}} \times \frac{\text{Volume of final extract (ml)}}{\text{Volume of sample injected (µl)}}$$

Results

Bioefficacy of azoxystrobin

Early blight

The first season results revealed that azoxystrobin was highly effective against leaf blight at 125 g a.i. ha^{-1} followed by 62.50 and 31.25 g a.i. ha^{-1} doses. The lowest concentration of azoxystrobin (31.25 g a.i.ha^{-1}) recorded 8.98 per cent incidence of leaf blight at 30^{th} day after spraying. The efficacy increased with the increase in concentration, but the rate of disease progress was found decreased in treated plots. The control plots recorded 13.55 PDI initially, which progressing upto 54.33 PDI as observed at the end of the experiment. The disease reduction over control was 92.82 per cent in azoxystrobin (125 g a.i.ha^{-1}) sprayed plot followed by its other doses *viz*., 62.50 g a.i. ha^{-1} (88.85%) and 31.25 g a.i. ha^{-1} (83.47%) as against mancozeb and carbendazim which recorded 83.61 and 78.69 per cent, respectively (Table 1). In the second season also, similar trend of results were obtained in which the higher dose (125 g a.i. ha^{-1}) of azoxystrobin recorded the lowest PDI of 4.86 followed by its lowest doses of 62.50 g a.i. ha^{-1} (7.53 PDI) and 31.25 g a.i ha^{-1} (11.10 PDI). Among the fungicides tested, carbendazim at 500 g ha^{-1} showed the least performance of 14.64 PDI. The control plots recorded the maximum disease incidence (42.66 PDI) at the end of the experiment (Table 2).

Table 1. Effect of azoxystrobin on leaf blight of tomato (Trial I).

Treatments	Before spray (PDI) *	1st Spray (PDI)*		2nd Spray (PDI)*		Per cent reduction over control
		7 DAS	15 DAS	7 DAS	15 DAS	
Azoxystrobin 31.25 g a.i ha^{-1}	13.90 (21.89)^{a}	14.68 (22.53)^{c}	13.26 (21.35)^{d}	10.41 (18.82)^{d}	8.98 (17.43)^{c}	83.47
Azoxystrobin 62.50 g a.i ha^{-1}	13.62 (21.65)^{a}	14.02 (21.99)^{cd}	12.50 (20.70)^{e}	9.08 (17.53)^{e}	6.06 (14.25)^{d}	88.85
Azoxystrobin 125 g a.i ha^{-1}	13.00 (21.13)^{a}	13.06 (21.18)^{d}	10.75 (19.14)^{f}	7.28 (15.65)^{f}	3.90 (11.90)^{e}	92.82
Mancozeb @ 1 kg ha^{-1}	12.80 (20.96)^{a}	16.46 (23.93)^{b}	15.20 (22.94)^{c}	12.10 (20.35)^{c}	8.90 (17.35)^{c}	83.61
Carbendazim @ 500 g ha^{-1}	12.89 (21.04)^{a}	16.90 (24.27)^{b}	15.76 (23.39)^{b}	12.66 (20.84)^{b}	11.58 (19.89)^{b}	78.69
Control	13.55 (21.59)^{a}	20.20 (26.67)^{a}	28.89 (32.13)^{a}	42.13 (40.47)^{a}	54.33 (47.48)^{a}	--

PDI - Percent disease index

*Mean of four replications. In a column, means followed by a common letters are not significantly different at the 5% level by DMRT

Values in parentheses are arcsine transformed values

Table 2. Effect of azoxystrobin on leaf blight of tomato (Trial II).

Treatments	Before spray (PDI) *	1st Spray (PDI)*		2nd Spray (PDI)*		Per cent reduction over control
		7 DAS	15 DAS	7 DAS	15 DAS	
Azoxystrobin 31.25 g a.i ha^{-1}	8.20 (16.64)^{a}	11.27 (19.62)^{b}	12.08 (20.34)^{c}	11.72 (20.02)^{c}	11.10 (19.46)^{c}	73.98
Azoxystrobin 62.50 g a.i ha^{-1}	8.05 (16.48)^{a}	9.15 (17.61)^{c}	9.39 (17.84)^{d}	8.67 (17.12)^{d}	7.53 (15.93)^{d}	82.35
Azoxystrobin 125 g a.i ha^{-1}	7.83 (16.25)^{a}	7.86 (16.28)^{c}	7.50 (15.89)^{d}	6.79 (15.10)^{e}	4.86 (12.74)^{e}	88.61
Mancozeb @ 1 kg ha^{-1}	7.83 (16.25)^{a}	11.30 (19.64)^{b}	12.20 (20.44)^{c}	11.90 (25.03)^{c}	10.68 (19.07)^{c}	74.96
Carbendazim @ 500 g ha^{-1}	7.92 (16.35)^{a}	11.43 (19.76)^{b}	13.52^{b} (22.00)	14.26 (22.18)^{b}	14.64 (22.49)^{b}	65.68
Control	8.03 (16.46)^{a}	13.58^{a} (21.64)	25.23 (30.15)^{a}	34.52 (35.98)^{a}	42.66 (40.78)^{a}	--

PDI - Percent disease index

*Mean of four replications

In a column, means followed by a common letters are not significantly different at the 5% level by DMRT. Values in parentheses are arcsine transformed values

Leaf spot

The higher doses of azoxystrobin (125 and 62.50 g a.i. ha^{-1}) exhibited 100 per cent reduction in leaf spot incidence (0.00 PDI) followed by its lower dose 31.25 g a.i. ha^{-1} which recorded the PDI of 2.00. Mancozeb and carbendazim sprayed plots recorded 4.10 and 7.32 PDI, respectively (Table 3). In the second season, all the doses of azoxystrobin were effective against the leaf spot and significantly superior over control. The disease incidence was 6.92, 5.14 and 2.42 PDI in the plots sprayed with azoxystrobin at 31.25, 62.50 and 125 g a.i. ha^{-1}, respectively on 15 days after second spray. In all the treatments decrease in disease trend was observed except in control, in which 30.29 per cent incidence was recorded (Table 4).

Table 3. Effect of azoxystrobin on leaf spot of tomato (Trial I).

Treatments	Before spray (PDI) *	1st Spray (PDI)*		2nd Spray (PDI)*		Per cent reduction over control
		7 DAS	15 DAS	7 DAS	15 DAS	
Azoxystrobin 31.25 g a.i ha^{-1}	0.00 $(0.50)^a$	0.00 $(0.50)^b$	0.00 $(0.50)^c$	2.00 $(8.13)^b$	2.00 $(8.13)^d$	90.76
Azoxystrobin 62.50 g a.i ha^{-1}	0.00 $(0.50)^a$	0.00 $(0.50)^b$	0.00 $(0.50)^c$	0.00 $(0.50)^c$	0.00 $(0.50)^e$	100.00
Azoxystrobin 125 g a.i ha^{-1}	0.00 $(0.50)^a$	0.00 $(0.50)^b$	0.00 $(0.50)^c$	0.00 $(0.50)^c$	0.00 $(0.50)^e$	100.00
Mancozeb @ 1 kg ha^{-1}	0.00 $(0.50)^a$	0.00 $(0.50)^b$	1.00 $(5.74)^b$	2.00 $(8.13)^b$	4.10 $(12.08)^c$	94.07
Carbendazim @ 500 g ha^{-1}	0.00 $(0.50)^a$	0.00 $(0.50)^b$	1.50 $(7.04)^b$	2.98 $(9.94)^b$	7.32 $(13.31)^b$	81.06
Control	0.00 $(0.50)^a$	5.20 $(13.18)^a$	9.50^a (17.95)	18.34^a (25.36)	21.65^a (27.73)	--

PDI - Percent disease index

*Mean of four replications. In a column, means followed by a common letters are not significantly different at the 5% level by DMRT

Values in parentheses are arcsine transformed values

Yield

In the first season, azoxystrobin at 125 g a.i. ha^{-1} recorded the highest yield of 27.60 tonnes ha^{-1} compared to control (10.38 tonnes ha^{-1}). Significant yield increase was found among the different doses of azoxystrobin treated plots. Among the other two fungicides treated plots, Carbendazim (500 g ha^{-1}) recorded the lowest yield of 22.11 tonnes ha^{-1} (Fig. 1). Similarly in the second season also the

azoxystrobin at 125 g a.i. ha^{-1} recorded the maximum yield of 26.30 tonnes ha^{-1} and the control plots recorded the lowest yield of 9.26 tonnes ha^{-1}.

Table 4. Effect of azoxystrobin on leaf spot of tomato (Trial II).

Treatments	Before spray (PDI) *	1st Spray (PDI)*		2nd Spray (PDI)*		Per cent reduction over control
		7 DAS	15 DAS	7 DAS	15 DAS	
Azoxystrobin 31.25 g a.i ha^{-1}	5.48 (13.53)[a]	6.37 (14.61)[b]	7.66 (16.06)[c]	7.40 (15.78)[d]	6.92 (15.25)[d]	77.15
Azoxystrobin 62.50 g a.i ha^{-1}	5.95 (14.12)[a]	6.45 (14.71)[b]	6.72 (15.02)[cd]	6.02 (14.20)[d]	5.14 (13.10)[d]	83.03
Azoxystrobin 125 g a.i ha^{-1}	4.86 (12.74)[a]	5.11 (13.06)[bc]	5.15 (13.11)[d]	3.49 (10.76)[e]	2.42 (8.94)[e]	92.01
Mancozeb @ 1 kg ha^{-1}	5.82 (13.96)[a]	7.23 (16.59)[b]	8.92 (17.38)[bc]	10.23 (18.65)[c]	9.72 (18.16)[c]	67.91
Carbendazim @ 500 g ha^{-1}	4.79 (12.65)[a]	7.12 (15.47)[b]	10.22 (18.65)[b]	12.44 (20.65)[b]	11.57 (19.90)[b]	61.80
Control	5.08 (13.03)[a]	10.35 (18.90)[a]	15.24 (22.98)[a]	24.88 (29.92)[a]	30.29 (33.36)[a]	--

PDI - Percent disease index
*Mean of four replications
In a column, means followed by a common letters are not significantly different at the 5% level by DMRT
Values in parentheses are arcsine transformed values

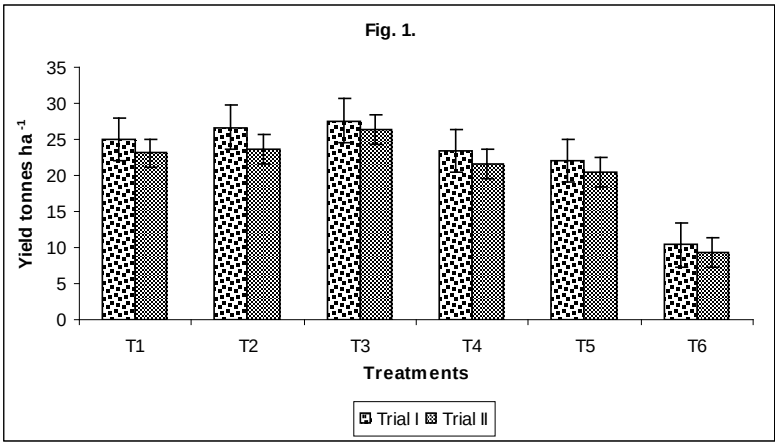

Fig 1. Effect of azoxystrobin on fruit yield of tomato.
T1 – azoxystrobin 31.25 g a.i. ha^{-1}; T2 – 62.50 g a.i. ha^{-1}; T3 – 125 g a.i. ha^{-1}; T4 – 250 g a.i. ha^{-1}; T5 – 500 g a.i. ha^{-1}; T6 - Control

Phytotoxicity

No Phytotoxicity symptoms were observed in all the tested concentrations of azoxystrobin (Table 5 and 6).

Table 5. Phytotoxic effect of azoxystrobin foliar application on tomato (Trial I).

Treatments	Phytotoxicity Particulars					
	Leaf injury	Wilting	Vein clearing	Necrosis	Epinasty	Hyponasty
Azoxystrobin 31.25g a.i. ha^{-1}	NP	NP	NP	NP	NP	NP
Azoxystrobin 62.50 g a.i. ha^{-1}	NP	NP	NP	NP	NP	NP
Azoxystrobin 125g a.i. ha^{-1}	NP	NP	NP	NP	NP	NP
Azoxystrobin 250 g a.i. ha^{-1}	NP	NP	NP	NP	NP	NP
Azoxystrobin 500 g a.i. ha^{-1}	NP	NP	NP	NP	NP	NP
Control	NP	NP	NP	NP	NP	NP

NP - No phytotoxicity

Table 6. Phytotoxic effect of azoxystrobin foliar application on tomato (Trial II).

Treatments	Phytotoxicity Particulars					
	Leaf injury	Wilting	Vein clearing	Necrosis	Epinasty	Hyponasty
Azoxystrobin 31.25g a.i. ha^{-1}	NP	NP	NP	NP	NP	NP
Azoxystrobin 62.50 g a.i. ha^{-1}	NP	NP	NP	NP	NP	NP
Azoxystrobin 125g a.i. ha^{-1}	NP	NP	NP	NP	NP	NP
Azoxystrobin 250 g a.i. ha^{-1}	NP	NP	NP	NP	NP	NP
Azoxystrobin 500 g a.i. ha^{-1}	NP	NP	NP	NP	NP	NP
Control	NP	NP	NP	NP	NP	NP

NP - No phytotoxicity

Persistence

Initial deposits of 1.3294, 1.9057, 2.1434, 3.5616 and 4.5270 µg g^{-1} were detected after application of azosytrobin at 31.25, 62.50, 125, 250 and 500 g a.i. ha^{-1}, respectively on tomato fruits in the field experiment I. The initial deposits dissipated from 19.45 to 38.05 per cent on first day after spraying (DAS) and reached below detectable residue level (BDL) after third DAS at 31.25 g a.i. ha^{-1}, fifth DAS at the dose of 62.50 and 125 g a.i. ha^{-1} and seventh DAS at 250 and 500 g a.i. ha^{-1} (Table 7). In the field trial II, spraying of azoxystrobin at 31.25, 62.50, 125, 250 and 500 g a.i. ha^{-1} left initial deposits of 0.9802, 1.5956, 2.0240, 3.1739 and 4.2481µg g^{-1}, respectively on tomato fruits. One day after spraying, the initial deposits dissipated by 19.14 to 65.19 per cent and reached below detectable level after three DAS at 31.25 g a.i. ha^{-1}, fifth day at 62.50 and 125 g a.i. ha^{-1} and after seven days at 250 and 500 g a.i. ha^{-1} (Table 8).

The best fit observed in tomato was first order kinetics for the most of the azoxystrobin treatments in both the trials and also followed the inverse power law (Table 9). The various statistical parameters like intercept (a), slope (b) of regression line and half life ($T_{0.5}$) with their confidence limits for the best fit function in tomato are presented in Table 33. The half life values were worked out for different doses *viz* ., 31.25, 62.50, 125, 250 and 500 g a.i. ha^{-1} were 0.8004, 1.0586, 1.5723, 1.9333 and 2.3852 days, respectively. Considering the maximum permissible residue limit of 2.0 ppm for tomato, the suggested waiting period after spraying of azoxystrobin at 62.50, 125, 250 and 500 g a.i. ha^{-1} was 0.2353, 0.6101, 1.9691 and 2.9746 days, respectively. In the field trial II, the half life values were 0.9022, 1.1358, 1.5847, 1.5025 and 2.1128 days and the suggested waiting period would be 0.1702, 1.5030 and 2.6848 days for azoxystrobin at 125, 250 and 500 g a.i. ha^{-1}, respectively.

Harvest time residues

The residues of azoxystrobin at different concentrations were found at below detectable level in the harvested fruits of tomato (Table 10).

Table 10. Harvest time residue of azoxystrobin in tomato fruits.

Treatments	Azoxystrobin residues (µg g^{-1})	
	Trial I	Trial II
Azoxystrobin 31.25 g a.i. ha^{-1}	BDL	BDL
Azoxystrobin 62.5 g a.i. ha^{-1}	BDL	BDL
Azoxystrobin 125 g a.i. ha^{-1}	BDL	BDL
Azoxystrobin 250 g a.i. ha^{-1}	BDL	BDL
Azoxystrobin 500 g a.i. ha^{-1}	BDL	BDL
Control	BDL	BDL

BDL – Below Detectable Level, Determinability: 0.004 µg g^{-1}.

Discussion

Tomato early blight and leaf spot are the most destructive diseases which caused huge economic losses (Babu, 1994; Kumar and Sugha, 2003). Several fungicides have been reported to be effective in controlling these diseases including captafol, mancozeb, copper oxychloride, chlorothalonil for tomato leaf blight and leaf spot (Bhardwaj, 1991; Dillard *et al.*, 1997) Though these fungicides have been used for long time for the control of leaf blight and leaf spot of tomato, there are certain strains of fungi which are resistant to copper fungicides and some strains of cucumber powdery mildew fungi resistant to dinocap and benomyl (Carlile, 1986). Whereas, the compound such as azoxystrobin are known to break down this resistance because of their different mode of action as compared to other fungicides. In this context, a new fungicide with different mode of action against the pathogen becomes optionally important. In the present study, azoxystrobin was highly effective against early leaf blight and leaf spot of tomato at 125 and 62.50 g a.i. ha^{-1} followed by 31.25 g a.i. ha^{-1}. From this study it is evident that 125 g a.i. ha^{-1} (500 ml) of azoxystrobin was considered as the optimum dose to combat the early blight and leaf spot diseases.

The optimum dose of azoxystrobin (125 g a.i. ha^{-1}) sprayed plot recorded 92.82 and cent per cent reduction of early blight and leaf spot, respectively in first season trials. From the second season trials conducted against tomato early blight and leaf spot the optimum dose of azoxystrobin (125 g a.i. ha^{-1}) recorded 88.61 and 92.01 per cent disease reduction, respectively. Azoxystrobin @ 125 g a.i. ha^{-1} in the first season trials of tomato recorded 166.02 per cent increase in yield over control. In the second season tomato trials also, the same dose of azoxystrobin recorded a maximum yield of 26.30 tonnes $ha^{-1}$and 184.02 per cent increase over control. The results are in accordance with excellent control, curative, translaminar and systemic properties of azoxystrobin enables it to be used efficiently against downy mildew of grapevine and leaf blight of tomato at very low application rates (Hewitt, 1998; Ranganathan, 2001; Mejia Arreaza and

Hernandez, 2001). In other crops, the fungicide azoxystrobin provide an effective control of downy mildew and powdery mildew diseases of grapevine (Wong and Wilcox, 2001; Schwartz and Gent, 2005). The fungicide azoxystrobin was found effective against powdery mildew of sweet cherry at Oronda (Grover and Boal, 1998). The effectiveness of azoxystrobin against *Pythium aphanidermatum* (Eds.) Fitz. in cucumber root rot (Utkhede and Bogdanoff, 2003), *Claviceps africana* McRao in sorghum ergot (Prom and Isakeit, 2003) and *Alternaria alternata* (Fr.) Keissler in apple for moldy rot disease were also reported. Azoxystrobin proved its effectiveness in checking powdery mildew and downy mildew of summer squash and muskmelon, respectively and was found effective against metalaxyl resistant strains of *Phytophthora infestans* (Mont.) de Barry. The compound appeared to be effective against *Fusarium moniliforme* Sheld. (Sheath rot of rice), *Helminthosporium oryzae* Brade-de –Haan (brown leaf spot) and *Aspergillus niger* Van Tieghem (collar rot of groundnut) (Thind *et al.,* 2002) The results from early and the present study showed that azoxystrobin is an effective fungicide for controlling early leaf blight and leaf spot diseases of tomato. Azoxystrobin is of great advantage to the growers since they can use this systemic fungicide for all the dreaded diseases. Azoxystrobin was applied at different concentrations (31.25, 62.50, 125, 250 and 500 g a.i. ha^{-1}) and the phytotoxicity symptoms were not observed even at very high concentration (500 g a.i. ha^{-1}). This is an added advantage in azoxystrobin spray indicating its safety to tomato crop. Similarly, there was no phytotoxic symptom throughout the cropping season due to azoxystrobin application. Ranganathan (2001) and Sendhil Vel *et al.* (2004) also found that there was no leaf injury on grapevine at a higher concentration of azoxystrobin.

Persistence of protective fungicide on the surface of the plant / plant parts plays an important role in determining their disease reduction potential and was highly useful in developing spray schedules. The results of persistence and dissipation of azoxystrobin in both the field experiments on tomato revealed that azoxystrobin at 31.25, 62.50, 125, 250 and 500 g a.i. ha^{-1} left an initial deposit ranged from 1.3294 to 4.5270 µg g^{-1} and 0.9802 to 4.2481 µg g^{-1} in the first and second field trial, respectively. Dissipation on initial deposits was from 19.45 to 38.05 and 19.14 to 65.19 per cent, respectively in the field trial I and II after one day of treatment. The residues reached BDL for 3 DAS at 31.25 g a.i. ha^{-1}, fifth day at 62.50 and 125 g a.i. ha^{-1} and seven DAS at 250 and 500 g a.i. ha^{-1} in both the field trials. The half life values of azoxystrobin at different doses were ranged from 0.8004 to 2.3852 days, and 0.9022 to 2.1128 days, respectively, in the first and second trial. Considering the maximum permissible residue limit of 2.0 ppm of tomato, the suggested waiting period after spraying of azoxystrobin at different concentrations was ranged from 0.2353, 0.6101 to 2.9746 days and 0.1702 to

2.6848 days in the first and second season, respectively. The results are in agreement with the findings of Gareur *et al.* (2002) who studied dissipation of azoxystrobin on tomatoes in green house, at the preharvest interval, the residues were below minimum residue level. The mechanism of disappearance showed that the decrease in residues was due to photo degradation. The effect of azoxystrobin residues on grapes from treatment to harvest and their fate in dried berries, wine and alcoholic beverages were reported. The disappearance rate (half life period $T_{0.5}$) was 3 - 4 days. The samples after drying did not show residue level *ie*, below detectable level. In the wines no detectable residues were found at the end of fermentation (Cabras and Angioni, 2000). It was also found azoxystrobin residues were recorded from grapevine fruits upto seven days and after that the residues were at below detectable level. He also stated that the half life ($T_{0.5}$) for fruit 2 to 3 days and 1.5 to 2 days for leaves after spraying (Sendhil Vel, 2003). Fungicides belonging to strobilurin groups are successful in controlling several plant diseases but their excessive, irrational and indiscriminate use can pose problems pertaining to the safety of the consumer. These chemicals may causes serious residue problems when they are applied at the maturing stage and a minimum waiting period is not followed. As many of the fruits and vegetables are consumed as raw products, fungicide residues on them may lead to health related problems. Work done on residues of fungicides in India is meagre although a number of fungicides are being used at present. The residue levels in the edible parts vary with the dose of the fungicides used and with the total number of sprays done (Tripathi *et al.*, 1976; Mithyantha *et al.*, 1977). If the dose used is high and it is applied at the improper time and a total number of sprays exceed than the recommended ones, there is every chance that the residues left in the crops at harvest time are higher than the tolerance limits prescribed. Standardization of fungicidal residue is an important activity as the quality parameters are interlinked with inherent toxicity, residual effects and phytotoxicity, etc. Analytical methods are given higher attention in order to ensure a higher degree of repeatability and reproducibility. The most widely used methods for faster, easier and sensitive analysis that permit screening of a large number of samples include GC and HPLC.

The present study illustrated with the reports of azoxystrobin residues on tomato fruits. The azoxystrobin residues were found at below detectable level in the harvested fruits of tomato. The minimum detectable level in tomato was 0.004 µg g^{-1} as the sample weight of 25 g of fruits. In tomato fruits, the MRL for azoxystrobin was 2.0 mg kg^{-1} (http: //www .hmso .gov.uk/legislation/scotland /ssi2002/20020271.htm, 2002). It was also reported that the residues of azoxystrobin were at below detectable level in harvested fruits of grapevine (Sendhil Vel *et al*., 2004). Majority of the fungicides were reported to be at

below detectable level in cucumber *viz.*, flusilazole and hexaconazole (Gupta and Gupta, 2001). The harvest time azoxystrobin residue in the present study was also recorded below detectable level in cucumber fruits. Hence, the fungicide can be safely used even upto 500 g a.i. ha^{-1} for the management of tomato early leaf blight and leaf spot diseases

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Table 7. Persistence and dissipation of azoxystrobin in tomato fruits (Trial I).

Days after spraying	Azoxystrobin residues (µg g^{-1})						Dissipation (%)					
	31.25 g a.i. ha^{-1}	62.50 g a.i. ha^{-1}	125 g a.i. ha^{-1}	250 g a.i. ha^{-1}	500 g a.i. ha^{-1}	Control	31.25 g a.i. ha^{-1}	62.50 g a.i. ha^{-1}	125 g a.i. ha^{-1}	250 g a.i. ha^{-1}	500 g a.i. ha^{-1}	Average dissipation (%)
0	1.3294	1.9057	2.1434	3.5616	4.5270	BDL	--	--	--	--	--	--
1	0.8235	1.2809	1.6487	2.7401	3.6462	BDL	38.05	32.79	23.08	23.07	19.45	27.29
3	0.1157	0.4860	1.1988	1.6358	1.9831	BDL	91.29	74.50	44.07	54.07	56.19	64.02
5	BDL	0.0689	0.2095	0.8339	1.2129	BDL	100.0	96.38	90.23	76.59	73.21	87.28
7	BDL	BDL	BDL	0.2646	0.5809	BDL	100.0	100.0	100.0	92.57	87.17	95.95
10	BDL	BDL	BDL	BDL	BDL	BDL	100.0	100.0	100.0	100.0	100.0	100.0
14	BDL	BDL	BDL	BDL	BDL	BDL	100.0	100.0	100.0	100.0	100.0	100.0

BDL – Below Detectable Level , Determinability: 0.004 µg g^{-1}

Table 8. Persistence and dissipation of azoxystrobin in tomato fruits (Trial II).

Days after spraying	Azoxystrobin residues (µg g^{-1})						Dissipation (%)					
	31.25 g a.i. ha^{-1}	62.50 g a.i. ha^{-1}	125 g a.i. ha^{-1}	250 g a.i. ha^{-1}	500 g a.i. ha^{-1}	Control	31.25 g a.i. ha^{-1}	62.50 g a.i. ha^{-1}	125 g a.i. ha^{-1}	250 g a.i. ha^{-1}	500 g a.i. ha^{-1}	Average dissipation (%)
0	0.9802	1.5956	2.0240	3.1739	4.2481	BDL	--	--	--	--	--	--
1	0.3412	0.9372	1.3539	2.2500	3.4351	BDL	65.19	41.26	33.11	29.10	19.14	37.56
3	0.0730	0.4558	0.7161	1.4189	2.2143	BDL	92.55	71.43	64.62	55.29	47.87	66.35
5	BDL	0.0668	0.2143	0.6330	1.0048	BDL	100.0	95.81	89.41	80.06	76.34	88.85
7	BDL	BDL	BDL	0.0814	0.4233	BDL	100.0	100.0	99.56	97.44	90.04	97.50
10	BDL	BDL	BDL	BDL	BDL	BDL	100.0	100.0	100.0	100.0	100.0	100.0
14	BDL	BDL	BDL	BDL	BDL	BDL	100.0	100.0	100.0	100.0	100.0	100.0

BDL – Below Detectable Level , Determinability: 0.004 µg g^{-}

Table 9. Intercepts, slope and half life of azoxystrobin residues in tomato fruits.

Treatments	A	LL	UL	B	LL	UL	$T_{0.5}$	LL	UL	Waiting period (Days)	Predicted equation
Trial – I											
T1	5.0591	4.4066	5.7117	- 0.8660	- 1.0866	- 0.6454	0.8004	0.5965	1.0043	--	Y = 5.0591 – 0.8660X
T2	5.4524	4.2524	6.6524	- 0.6548	- 1.0605	- 0.2491	1.0586	0.4027	1.7144	0.2353	Y = 5.4524 – 0.6548X
T3	5.5673	4.0184	7.1161	- 0.4409	- 0.9645	0.0828	1.5723	- 0.2951	3.4396	0.6101	Y = 5.5673 – 0.4409X
T4	6.0043	5.5142	6.4943	- 0.3585	- 0.4781	- 0.2390	1.9333	1.2886	2.5780	1.9691	Y = 6.0043 – 0.4781X
T5	6.1628	6.0040	6.3251	- 0.2906	- 0.3302	- 0.2510	2.3852	2.0601	2.7103	2.9746	Y = 6.1618 – 0.2906X
Trial – II											
T1	4.4276	3.8022	5.0531	- 0.7683	- 0.9797	- 0.5568	0.9022	0.6539	1.1505	--	Y = 4.4276 – 0.7683X
T2	5.2059	3.95798	6.4539	- 0.6103	- 1.0321	- 0.1884	1.1358	0.3506	1.9211	--	Y = 5.2059 – 0.6103X
T3	5.3728	4.7814	5.9641	- 0.4374	- 0.6373	- 0.2375	1.5847	0.8604	2.3090	0.1702	Y = 5.3728 – 0.4374X
T4	6.0269	4.8376	7.2162	- 0.4849	- 0.7751	- 0.1947	1.5025	- 1.6975	0.5984	1.5030	Y = 6.0269 – 0.4849X
T5	6.1791	5.8018	6.5565	- 0.3281	- 0.4201	- 0.2360	2.1128	1.5199	2.7057	2.6848	Y = 6.1791 – 0.3248X

T1 – azoxystrobin (25 SC) 31.25 g a.i. ha^{-1}; T2 – 62.50 g a.i. ha^{-1}; T3 – 125 g a.i. ha^{-1}; T4 – 250 g a.i. ha^{-1}; T5 – 500 g a.i. ha^{-1}

A – Intercepts; LL- Lower Limit; UL – Upper Limit; B - Slope ; T 0.5 – Half Life.