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Compatibility of azoxystrobin and chaetoglobosin biomolecules with bacterial antagonists

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Abstract
The compatibility of azoxystrobin and chaetoglobosin biomolecules with bacterial antagonists was tested by using poison food technique. The results of this experiment denoted that, even at the highest concentration of azoxystrobin (2000 ppm) the bacterial growths were not inhibited, confirmed by recording the OD value of *Bacillus subtilis* and *Pseudomonas fluorescens* which was 1.86 and 1.84 as compared to 1.94 and 1.99 in control respectively. Chaetoglobosin also not controlled the multiplication of bacteria even at the highest concentration (2000 ppm) showed the OD value of 1.78 and 1.86 after 48 h of inoculation against *Bacillus subtilis* and *Pseudomonas fluorescens*. The OD values in control were 1.82 and 1.86 respectively against the same bio inoculants. Since not much difference observed between treatments and control, both the molecules could be compatible with bacterial bioinoculants.

Keywords: Azoxystrobin, chaetoglobosin, compatibility, bacterial antagonist

Introduction
Since fungicides may have lethal effects on the pathogen as well as antagonist, an understanding of the effect of fungicides on the pathogen and the antagonist, would provide information on the selection of suitable fungicides and fungicide resistant antagonist in combination to combat plant pathogens. The idea of combining biocontrol agents with fungicides is for the development or establishment of desired microbes in the rhizosphere (Papvizas and Lewis, 1981; Basamma and Kulkarni, 2017) [6, 1]. Vidhya *et al*., (2012) [9] tested *Pseudomonas fluorescens* as seed treatment bio-inoculant against *R. solani* in bean with the combination of azoxystrobin, fluidioxanil, pencyuron and tebuconazole at various concentrations and established a compatible reaction. Utkhede and Koach (2002) [8] suggested that azoxystrobin and *Bacillus subtilis* could be applied together as post inoculation spray against gummy stem blight of cucumber caused by *Didymella bryoniae* (Auersw.). Basha *et al*., (2012) [2] reported that an isolate (TPT15) of *Pseudomonas* sp exhibited very good compatibility with mancozeb (when used at a concentration of 0.2 per cent) and carbendazim (when used at a concentration of 0.1 per cent) to the tune of 99.50 per cent 92.88 per cent, respectively. These combination treatments were found to be very much effective against *Sclerotium rolfsii* causing stem rot of groundnut. Hence in the present study the compatibility of azoxystrobin and chaetoglobosin biomolecules with bacterial antagonists were tested under *in vitro.*

Materials and methods
The compatibility of the azoxystrobin and chaetoglobosin biomolecules with the bacterial antagonists was tested using Turbidometric method (ISI, 1964). One ml of the bacterial culture was transferred to a 250 ml sidearm flask containing 50ml of King's B and Nutrient broth for *Pseudomonas* and *Bacillus* respectively and amended separately with azoxystrobin and chaetoglobosin at four different concentrations *viz.,* 500, 1000, 1500 and 2000 ppm. The control was maintained without inoculation of bacterial culture and fungicides. The flasks were incubated at 28±2 ° C in a psycotherm shaker. The optical density values of the culture broth were determined in Spectro Photo colorimeter at 610 nm at regular intervals of 6 h.

Results
The results of this experiment denoted that, even at the highest concentration of azoxystrobin (2000 ppm) the bacterial growths were not inhibited, confirmed by recording the OD value of *Bacillus subtilis* and *Pseudomonas fluorescens* which was 1.86 and 1.84 as compared to 1.94

and 1.99 in control respectively. Chaetoglobosin also not controlled the multiplication of bacteria even at the highest concentration (2000 ppm) showed the OD value of 1.78 and 1.86 after 48 h of inoculation against *Bacillus subtilis* and *Pseudomonas fluorescens*. The OD values in control were 1.82 and 1.86 respectively against the same bio inoculants.

Discussion

Combinations of bacterial antagonist with fungicides would provide better control of plant diseases when compared to individual applications (Kataria *et al*., 2002). Application of azoxystrobin in combination with *P. fluorescens* strain W 36 showed better antagonist interaction against *Rhizoctonia solani* in bean and cucumber. Utkhede and Koch (2002) [8] reported that *B. subtilis* strain AGB1 along with azoxystrobin controlled the gummy stem blight (*Didymella bryoniae*) under green house condition. Sendhil Vel *et al*., (2004b) [7] found that *P. fluorescens* and *B. subtilis* growth were not affected by azoxystrobin at various concentrations *viz.*, 100, 150, 200, 250 and 300 ppm. Vidhya *et al.*, (2012) [9] denoted that the bacterial antagonists *B. subtilis* and *P. fluorescens* showed considerable tolerance against carbendazim and hexaconazole. Krishna Kumar *et al*., (2011) [4] reported that *B. subtilis* was compatible with the azole group of fungicides such as hexaconazole, propiconazole and tebuconazole up to 1000 ppm. (Basha *et al*., 2012 and Louis *et al*., 2016) [2, 5] found that *Pseudomonas* spp were highly compatible with mancozeb (99.5 per cent), and carbendazim (93 per cent) when used at 0.2 and 0.1 per cent concentrations respectively. Supported by earlier findings, the results of current investigation had revealed that the bacterial antagonist *B. subtilis* and *P. fluorescens* were fully compatible with azoxystrobin and chaetoglobosin. There was no growth reduction of bacterial bioinoculants observed even at the highest concentration of both molecules (2000 ppm). As there was no difference in the OD value between control and treatments, it is assumed that the tested bacterial antagonists might be compatible with azoxystrobin and chaetoglobosin biomolecules.

Table 1: Effect of azoxystrobin on *Bacillus subtilis* and *Pseudomonas fluorescens.*

Days after inoculation	Absorbance at 610nm at different hours after inoculation													
	Bacillus subtilis							*Pseudomonas fluorescens*						
	12	18	24	30	36	42	48	12	18	24	30	36	42	48
500	0.82^{d}	1.09^{d}	1.62^{c}	1.98^{b}	2.08^{ab}	1.92^{bc}	1.63^{c}	1.46^{bc}	1.65^{b}	1.93^{a}	2.10^{b}	2.04^{a}	1.96^{a}	1.84^{ab}
1000	0.96^{c}	1.33^{c}	1.85^{b}	2.07^{b}	1.94^{b}	1.83^{c}	1.71^{bc}	1.39^{c}	1.76^{ab}	2.01^{a}	2.32^{a}	2.19^{a}	2.08^{a}	1.95^{a}
1500	1.30^{b}	1.40^{c}	1.89^{b}	2.32^{a}	2.10^{ab}	1.98^{abc}	1.65^{c}	1.40^{c}	1.61^{b}	1.97^{a}	2.13^{ab}	2.08^{a}	1.99^{a}	1.72^{b}
2000	1.34^{b}	1.65^{b}	1.98^{ab}	2.15^{ab}	2.26^{a}	2.12^{a}	1.86^{ab}	1.56^{ab}	1.74^{ab}	1.99^{a}	2.14^{ab}	2.01^{a}	1.93^{a}	1.84^{ab}
Control	1.52^{a}	1.86^{a}	2.14^{a}	2.34^{a}	2.27^{a}	2.04^{ab}	1.94^{a}	1.62^{a}	1.86^{a}	2.03^{a}	2.28^{ab}	2.19^{a}	2.07^{a}	1.99^{a}

Mean of four replications

In a column, means followed by same letter are not significantly different at the 5 per cent level by DMRT.

Table 2: Effect of chaetoglobosin on *Bacillus subtilis* and *Pseudomonas fluorescens.*

Days after inoculation	Absorbance at 610nm at different hours after inoculation													
	Bacillus subtilis							*Pseudomonas fluorescens*						
	12	18	24	30	36	42	48	12	18	24	30	36	42	48
500	0.93^{a}	1.17^{a}	1.98^{a}	2.11^{a}	2.03^{ab}	1.82^{ab}	1.76^{c}	1.38^{c}	1.69^{b}	1.93^{b}	2.04^{b}	1.98^{b}	1.87^{a}	1.65^{b}
1000	0.86^{a}	0.93^{c}	1.28^{d}	1.97^{ab}	1.84^{c}	1.71^{b}	1.79^{b}	1.61^{b}	1.82^{ab}	1.99^{ab}	2.28^{a}	2.23^{a}	2.01^{a}	1.78^{ab}
1500	0.89^{a}	1.04^{b}	1.43^{bc}	1.85^{bc}	1.82^{c}	1.77^{ab}	1.62^{d}	1.54^{b}	1.76^{ab}	1.98^{ab}	2.39^{a}	2.18^{ab}	1.87^{a}	1.72^{ab}
2000	0.75^{b}	0.98^{bc}	1.39^{cd}	1.77^{c}	1.96^{bc}	1.83^{ab}	1.78^{b}	1.69^{ab}	1.82^{ab}	2.03^{ab}	2.26^{a}	2.15^{ab}	1.94^{a}	1.86^{a}
Control	0.87^{a}	1.24^{a}	1.56^{b}	1.87^{bc}	2.17^{a}	1.94^{a}	1.82^{a}	1.78^{a}	1.93^{a}	2.16^{a}	2.37^{a}	2.06^{ab}	1.99^{a}	1.82^{ab}

Mean of four replications

In a column, means followed by same letter are not significantly different at the 5 per cent level by DMRT

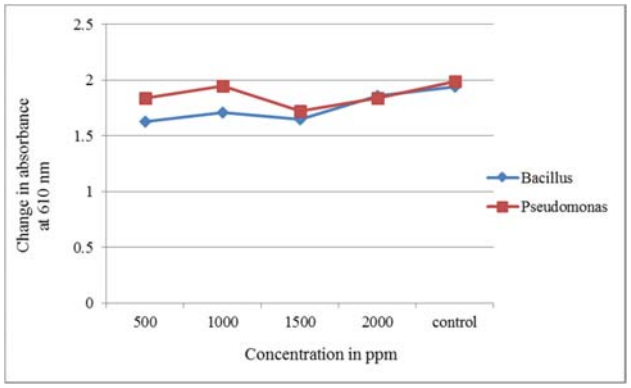

Fig 1: Compatibility of azoxystrobin with *Bacillus subtilis* and *Pseudomonas fluorescens.*

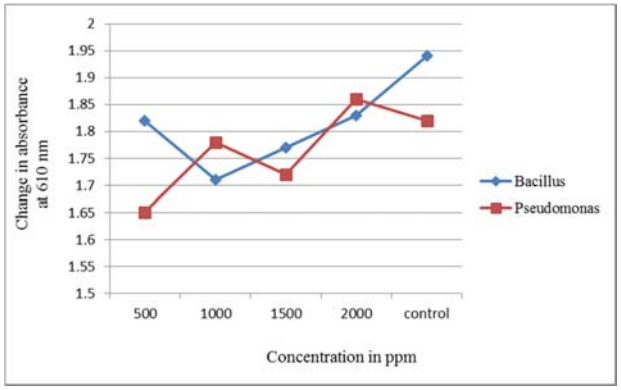

Fig 2: Compatibility of chaetoglobosin with *Bacillus subtilis* and *Pseudomonas fluorescens.*

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