

Research Article

Synergistic Effect of Micronutrients on *Aureofungin* Resistance in *Alternaria Tenuis* causing Fruit Rot of Grape

K. S. Kadam

Department of Botany, K.K.M.College, Manwath-431505, Dist. Parbhani(MS), India

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Abstract: Fruit rot of grape caused by *Alternaria tenuis* was found to be resistant to Aureofungin. Micronutrients viz.Bo, Co, Cu, Fe, Mb, Mg, Mn and Zn individually and mixture with Aureofungin were tested both *in vitro* and *in vivo* against resistant mutant of *Alternaria tenuis*. Results showed that individually PCE was higher in Zn, Bo, Co, Cu, Fe and Mn on plates when compared with aureofungin at 100 µg/ml. Use of aureofungin in mixture with micronutrients, all micronutrients showed higher PCE.

Keywords: Fruit rot, *Alternaria tenuis*, Aureofungin.

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INTRODUCTION

Grape (*Vitisvinifera* L.) is one of the very important fruit crop in India and abroad. However, fruit rot of Grapes is caused by many fungal pathogens. Among these, Fruit rot of grapes caused by *Alternaria tenuis* is destructive disease in the field as well as during storage and transport (Chahal and Malhi, 1969; Krishnaiah et al., 1983; Rao, 1994). Aureofungin is most effective fungicide against *Alternaria spp.* (Ghosh and Gemawat, 1976; Krishna et al., 1998). Fungicide resistant cases in various plant pathogens have been reported in India as well as in other countries (Wild, 1980; Annamalai and Lalithakumari, 1990; Gangawane et al., 1995). The main objective of present study was to find out the synergistic effect of micronutrients on the management of Aureofungin resistant mutant of *Alternaria tenuis*.

MATERIAL AND METHODS

The sensitivity of *Alternaria tenuis* isolates to Aureofungin was determined by food poisoning technique (Nene and Thaplial, 1993). Czapek Dox agar plates containing different concentration (50 – 1000 µg/ml) of Aureofungin were prepared. Disc (4mm) of pathogen isolates taken from the margin of 7 days old colony were placed in the center of agar plates. These plates were then incubated at 26±3°C and linear growth

was measured at different intervals up to a week. MIC and ED₅₀ were calculated. Thus the sensitivity of twenty isolates was determined. There was a large variation in the sensitivity of isolates. During present investigation, disease resistance of the pathogen was developed by chemical mutation and it was used for further study as suggested by Dekker (1982). Thus the EMS-At-3 mutant was obtained with highest resistant factor 6 and used for present study. The agar plates containing sub lethal dose of Aureofungin and micronutrients (10 and 100 µg/ml) were prepared and inoculated with resistant mutant of *A. tenuis*. The plates were incubated at 26±1°C. The agar plate without treatment served as control. The percentage control efficacy (PCE) was calculated 8 days after incubation period as

$$PCE = 100 (1 - x/y)$$

Where, x = Diameter of colony in treated plates or Percentage disease index of treated fruits,

y = Diameter of colony in control or Percentage disease index of untreated fruits.

In vivo studies were carried out on fruit of grape. The fruits were surface sterilized by treating them with 1% HgCl₂ solution and were washed ten times with sterilized distilled water. The fruits were then treated with the mixture of Aureofungin and micronutrients (10 and 100 µg/ml). The resistant mutant *Alternaria tenuis* was inoculated by pin prick method on

the fruits and they were incubated for a week at $26 \pm 3^\circ\text{C}$ in the laboratory. Fruits without treatment served as control. Percentage disease index was calculated and

then on PDI, the percentage control efficacy was calculated as above equation.

Table 1 Percentage control efficacy of Micronutrients individually and in mixture with aureofungin against aureofungin resistant mutant (EMS-At-3) of *Alternaria tenuis* on agar plates.

Sr. No.	Micronutrients ($\mu\text{g/ml}$)	Individual (PCE)	Mixture (PCE)
1	Bo 10	32.62	60.26
	100	47.04	72.38
2	Co 10	38.73	64.33
	100	57.00	78.49
3	Cu 10	36.67	66.16
	100	54.19	75.56
4	Fe 10	50.19	71.25
	100	68.93	94.63
5	Mb 10	26.34	54.33
	100	46.24	66.49
6	Mg 10	35.67	65.13
	100	53.19	80.29
7	Mn 10	42.26	61.49
	100	61.93	84.63
8	Zn 10	47.36	74.31
	100	65.83	87.84
9	Aureofungin (800 $\mu\text{g/ml}$)	44.67	-
	S. E.	2.56	2.63
	C.D. at 0.05	52.08	76.67
	0.01	54.35	78.44

Table 1 Percentage control efficacy (PCE) of Micronutrients individually and in mixture With aureofungin against aureofungin resistant mutant (EMS-At-3) of *Alternaria tenuis* on grape fruits.

Sr. No.	Micronutrients ($\mu\text{g/ml}$)	Individual (PCE)	Mixture (PCE)
1	Bo10	21.73	52.73
	100	38.65	64.19
2	Co10	31.64	56.49
	100	47.66	69.73
3	Cu10	28.93	58.14
	100	40.70	68.62
4	Fe10	41.63	62.43
	100	52.31	83.94
5	Mb10	20.64	41.92
	100	58.34	58.66
6	Mg10	28.73	48.83
	100	42.62	70.15
7	Mn10	33.49	46.88
	100	50.46	63.56
8	Zn10	35.82	61.51
	100	56.44	73.69
9	Aureofungin (800 $\mu\text{g/ml}$)	38.94	-
	S. E.	2.64	2.60
	C.D. at 0.05	43.67	65.61
	0.01	45.46	67.37

RESULTS AND DISCUSSION

In vitro present study revealed that individually Bo, Co, Cu, Fe, Mg and Zn showed higher PCE when compared with aureofungin at 100 $\mu\text{g/ml}$. However, in mixture all the micronutrients showed higher PCE, Fe found higher PCE followed by Zn, Mg, Mn, Co, Cu and Bo in decreasing manner (Table -1). *In vivo* study revealed that individually PCE was maximum in Zn, Mn, Mg, Fe and Cu but when the fruits treated with aureofungin in mixture with micronutrients, all

micronutrients showed higher PCE when compared aureofungin, Fe showed higher PCE followed by Zn, Mg, Co, Cu, Bo, Mb and Mn in decreasing manner (Table 2).

The results are in agreement with the finding of earlier workers Gangawane and Reddy (1986) showed that certain micronutrients when used in combination with carbendazim reduced the resistance in *Aspergillus flavus*. Gangawane L.V. and Kamble S.S.

(2001) found that when carbendazim was used in combination with agrochemicals inhibited the growth of resistant isolate of *Macrophominaphseolina* causing charcoal rot of potato. Bhale et al (2009) showed that among micronutrients cobalt and molybdenum showed 100% PCE when used in mixture against *Fusariumoxysporum*. There are also theoretical models suggested by Kable and Jaffery (1980). Dekker (1981), suggested that there is significant delay of resistance build up in the pathogen when the mixture of different agrochemicals was used.

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