



## INTEGRATED MANAGEMENT OF ZYZYPHUS FRUIT ROT CAUSED BY CARBENDAZIM RESISTANT MUTANT OF ASPERGILLUS NIGER USING MICRONUTRIENT COMBINATION WITH CARBENDAZIM

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### Abstract

Different agrochemicals and botanical pesticides were used for the integrated Management of Carbendazim resistance in *Aspergillusniger*. It was studied by mixing different Micronutrient and plant part extracts which have antifungal activity were mixed in carbendazim. In vivo studies carried out. Carbendazim and effective concentration of selected Micronutrient extracts of Medicinal plants were mixed well and ber fruits were treated with this solution. Plant extracts alone and in combination with carbendazim and other micronutrient were used individually and in combination with carbendazim for the management of *Zizyphus* fruit rot caused by resistant mutant of *Aspergillusniger*

**Keyword:** *Aspergillusniger*, Carbendazim Resistant Micronutrient



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### Introduction-

The fruit Ber (*Zizyphus mauritiana* Lamk.) are very nutritious fruit and are available at low cost hence it is really a poor man's fruit. It is xerophytes in nature. In Maharashtra the *Zizyphus* plant is found every field wherever irrigation facilities are less. The genus *Zizyphus* belongs to family Rhamnaceae and consist of 40 species in tropical and subtropical regions. There are more than 125 cultivars grown in India. However Ber suffers from various diseases like powdery mildew, sooty mold, Alternaria, Leaf spot and phoma leaf spot. The fruit are attacked by many pathogens at pre and post-harvest condition and spoil test and market quality. Among the post harvest pathogens *Aspergillusniger* was observed to be most common in Maharashtra state. There are reports that pre and post-harvest diseases of various fruits can be managed through systemic and conventional fungicides including carbendazim. However during last 30 years application of fungicides has plagued with several environmental problems and development of fungicide resistance in pathogen is major one. Many examples of fungicide resistance in fungal pathogens have been reported from U.S.A. Australia Europe, Israel, Japan, and from South East Asian countries. In India, it appears that management of pathogenic fungi with systemic and non-systemic fungicides has become more and more common over past 15 years. The aim of present study was therefore to examine the

possibility of development of resistance in *Aspergillusniger* against carbendazim and to find out the integrated methods to manage this pathogen causing fruit rot of *Zizyphus*. Integrated management of a disease have been emphasized now a day's hence agrochemicals were used individually and in combination with carbendazim for management of *Zizyphus* fruit rot caused by resistance mutant of *Aspergillusniger*.

### Material and Method

Diseased fruit samples of *Zizyphusmauritiana* were collected from various places. The observation for the pathogen was made by preparing slide and isolation on the medium. The identification of the pathogens was done through the referring earlier literature. It was noted that *Aspergillusniger* was present on maximum fruits. Hence it was selected for further studies as follows. Different micronutrient were used for the integrated management of carbendazim resistance in *Aspergillusniger*. It was studied by mixing different Micronutrient like Bo, Cu, Fe, Mg and MO These were agrochemical used individually and in combination with carbendazim for the management of *Zizyphus* fruit rot caused by resistance mutant of *Aspergillusniger*. In vivo studies were carried out. This was done by using mycelia suspension of *Aspergillusniger* strain. A.N. EMS.9 was inoculated on Ber fruits for pathogenicity test. Ber fruit were surface sterilised with 0.01% HgCl<sub>2</sub> solution and washed 10 times with sterile distilled water. They were inoculated with spore suspension of *Aspergillusniger* isolates or mutant resistant to carbendazim. Percentage Control Efficacy (PCE) was calculated (Cohen, 1989). In order to study the effect of carbendazim and other agrochemicals. Percentage control Efficacy (PCE) was calculated by using following formula.

$$PCE = 100(1 - X/Y)$$

Where X = Diameter of the colony on the plates containing carbendazim.

Y = Diameter of the colony on absolute control plates

Percentage control efficacy (PCA) was recorded after 12 days. In vitro wild sensitive isolate AN-9 was cultured on agar plates containing sublethal dose of carbendazim (2.5mg/ml). The and Mowere mixed in carbendazim by food poisoning technique (Nene and Thaphiyal, 1982). The Principle involved in this technique is to "poison" the nutrient medium with a fungi toxicant and allowing a test fungus to grow on such medium (Zapek-Dox) medium (2x) was prepared. It was sterilized and 10 ml of this medium was properly mixed with 10ml of carbendazim alone and Combination with other herbicide (2x a.i. concentration) selected for

study in sterile petriplates. These agrochemicals were used individually and in combination with carbendazim.

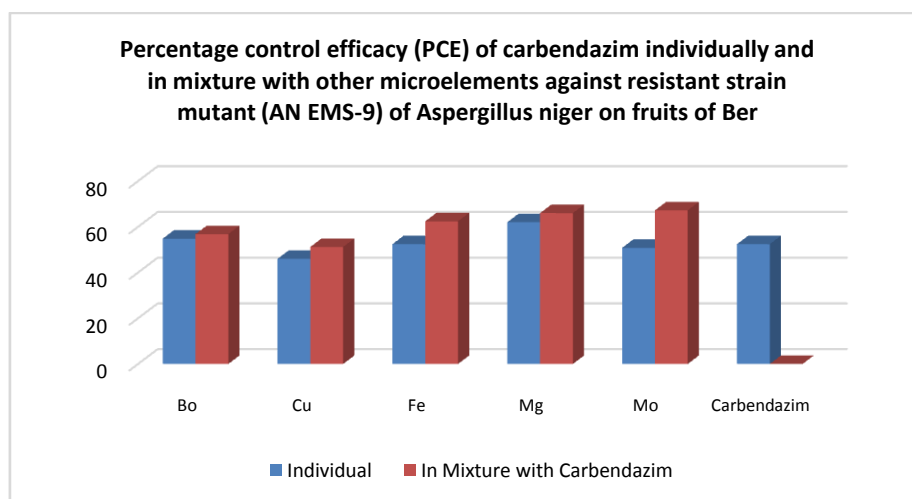
### Experimental results

#### Micro nutrients

Bo, Cu, Fe, Mg and Mo were used individually and in combination with carbendazim. Results in Table 34 and Fig. 25b indicate that all the micronutrients showed PCE on the *Zizyphus* fruits. Increase of the concentration again increased the PCE. Use of Mg alone gave higher PCE than other micronutrients, use of micronutrients in combination with carbendazim was found to more effective. Here Mo in combination with carbendazim was more effective than Cu, Mg, Fe and Bo.

**Table 01: Percentage control efficacy (PCE) of carbendazim individually and in mixture with other microelements against resistant strain mutant (AN EMS-9) of *Aspergillus niger* on fruits of Ber**

Sr. No.	Micronutrient (µg/ml)		PCE	
			Individual	In Mixture with Carbendazim
1	Bo	50	54.8	56.8
		100	62.4	70.0
2	Cu	50	46.0	51.2
		100	51.2	63.2
3	Fe	50	52.4	62.4
		100	63.2	74.0
4	Mg	50	62.0	66.0
		100	70.4	82.0
5	Mo	50	50.8	67.2
		100	66.0	71.2
6	Carbendazim only (2.5 µg/ml)		52.4	--



## Discussion

It was seen that altogether five diseases caused by *Aspergillusniger*, *Aspergillusflavus*, *Alternariasp.*, *Fusariumoxysporum* and *Rhizoctoniabataticola* were recorded. Mukherjee and Bhasin (1986) showed the fruit rot by 19 pathogens. However *Aspergillusflavus* recorded for the first time in this investigation. Further 10 isolates were tested against carbendazim. It was found that there was quite a large variation in the MIC of carbendazim against this pathogen. The MIC varied from 5 to 10 µg/ml *in vitro* and *in vivo*. Isolate AN-1 was sensitive while isolate AN-9 was tolerant. This indicated that the pathogen has a tendency to develop resistance against carbendazim. Variations in the sensitivity of isolates of different pathogens against various fungicides have been shown by many workers (Jones and Ehret, 1976; Gangawane and Saler, 1981; Hollomon, 1984; Khilareet. *al.*, 2003). There are also reports supporting these findings on the development of carbendazim resistance in various pathogens (Reddy, 1986; Gangawane, 1990; Chander and Thind, 1995). El-Goorani *et. al.*, (1984) and Spotts and Cervantes (1986) have reported benomyl resistance in different *Penicillium*spp. All of them agree that resistant mutants are emerged due to spontaneous mutation under selection pressure of fungicides. Gangawane and Reddy (1986) showed that certain micronutrients when used singly or in mixture with carbendazim reduce resistance in *Aspergillusflavus*. There are theoretical models developed in this basis, (Kable and Jeffery, 1980; Skylakakis, 1981; Levy *et. al.*, 1983) and practical examples (Delp, 1980; Dekker, 1981; Gangawane and Shaikh, 1988; Gangawane *et. al.*, 1995). Dekkar (1981) suggested that there is significant delay of resistance build up in the pathogens when mixture of different fungicides have been used. In the present study agrochemicals other than fungicides have also been proved useful in the management of carbendazim resistance in *Aspergillusniger* causing fruit rot of *Zyzyphus*

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