



BIOCHEMICAL ASPECTS LIKE TRACE ELEMENT OF CARBENDAZIM RESISTANT *ASPERGILLUS NIGER*

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ABSTRACT

The effect of nutritional sources such as, oxides and trace elements sources on growth and sporulation of resistant mutant was studied. The efforts were made to find out and biological characteristics of resistant strains of *Aspergillus niger*. There was quite a large variation in the biological Characteristics among the sensitive and resistant nutritionally the growth of resistant mutant was always higher on all nutritional sources when compared with the wild sensitive isolate. The growth of resistant mutant was highly supported by Cu, Fe, Mg, Mn, Na, Ca, when compared with the Carbendazim sensitive isolate. This was also related to the sporulation of resistant isolate.

KEYWORD: Carbendazim, Resistant, Sensitive, *Aspergillus niger*, trace element.

INTRODUCTION

The Ber (*Zyzyphus mauritiana* Lamk) is an important fruit crop of arid and semiarid tropics in our country. It is xerophyte in nature and survives well under water stress condition. The fruits are very nutritious and are available at low cost. The fruit are attacked by many pathogens at pre-and post-harvest condition and spoil the test and market quality. Under WHO the fruits are Supposed to be free from the diseases in order to export them aboard. Therefore, it was considered to study the post-harvest disease of *Zyzyphus* fruit in market and storage condition in Maharashtra and workout the management strategies. The samples of *Zyzyphus* fruits infected with different Pathogens were collected from market at different Places in Maharashtra. The pathogen was *Aspergillus niger* found to be dominated on maximum fruit. Hence isolate of *Aspergillus niger* were Isolated for the study. The sensitivity of isolates was tested. The samples of *Zyzyphus* fruits infected with different pathogen were collected from market at different Places in Maharashtra. The Pathogen was *Aspergillus niger* found to be dominated on maximum fruit. Hence isolate of *Aspergillus niger* was isolated for the study. The sensitivity of isolates was tested the ber suffers from various diseases like powdery mildew (Jayarajan and Chima, 1972). Sooty mold or black spot (Sharma *et al.*, 1983), *Alternaria* leaf spot, Phoma leaf spot and Fruit rot caused by *Phoma* spp., *Alternaria* spp., *Colitotrichum* spp. and *Pestalotiaversicolor*. It is also known that eighteen different fungal species cause post-harvest decay

of Ber fruits. Among this *Aspergillus flavus* infection is more common. The post-harvest pathogen not only resulted in maximum loss of ascorbic acid from the fruit tissues but also induced as aflatoxin production during pathogenesis (Singh and Sumbali, 2000).

Various types of the fruit rot of Ber have been reported by Pawar and Vyas (1974) and Gupta and Madaan (1978), Manoharachary *et al.*, (1989), Sharma *et al.*, (1981), Srivastava (1967), Sumbali and Mehrotra (1982) and Ullasa and Rawal (1986), Vayas and Pawar (1974) and Wadia and Manoharachary (1980)

MATERIAL AND METHODS

Nutritional studies: Various nutritional sources were added into CzapekDox agar medium in the plates and inoculated with both the carbendazim sensitive (AN-9) and resistant (AN EMS-9) strains were inoculated at the centre. Diameter of the colony and sporulation was recorded after 9 days incubation period.

Different insecticides were used for the integrated management of Carbendazim resistance in *Aspergillus niger*. It was studied by mixing different oxides trace element. These were used individually and in combination with carbendazim for the management of *Zyzyphus* fruit rot caused by resistance mutant of *Aspergillus niger*. In vivo studies were carried out. This was done by using mycelia suspension of *Aspergillus niger* strain.

A.N. EMS.9 was inoculated on Ber fruits for pathogenicity test. Ber fruit were surface sterilised with 0.01% HgCl₂ solution and washed 10 times with sterile distilled water. They were inoculated with spore suspension of *Aspergillus niger* 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 plate with Carbendazim only served as control. And different oxide, trace element was 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 oxides, trace element (2x a.i. concentration) selected for study in sterile Petri plates.

EXPERIMENTAL RESULT

Effect of trace elements

All the trace elements were used in sulphate form. Co, Cu, Fe, Mg, Mn, Na, Zn and Ca were used. The results are shown in Table 1. Fig. 1. It was seen that all the trace elements were inhibitory except Mg where slight growth was seen in case of sensitive isolate. However, the growth of resistant mutant was supported by Co, Fe, Mg, Mn, Na and Ca. But Cu and Zn were completely inhibitory for the resistant mutant. Only Mg showed the sporulation of sensitive isolate while all the trace elements except Cu and Zn supported the growth of resistant mutant.

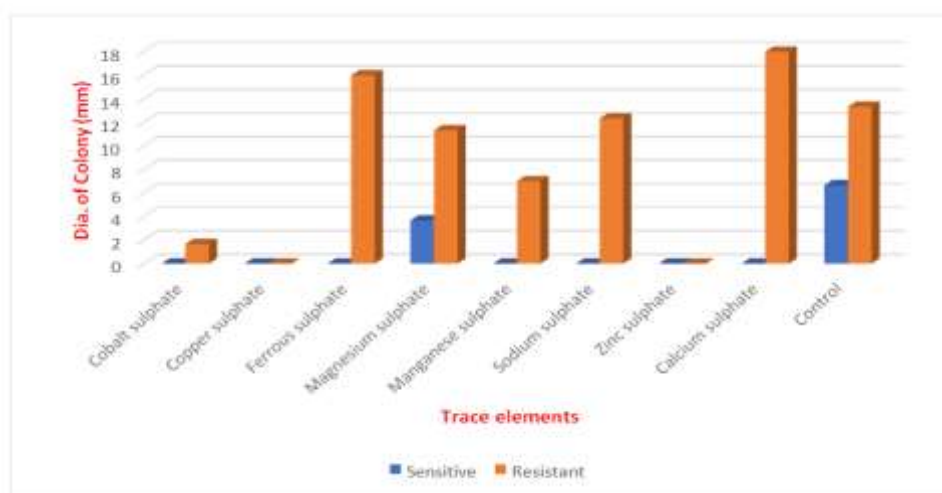


Fig. 1: Effect of different trace elements on the growth of wild sensitive isolate AN-9 and carbendazim resistant strain AN EMS-9 of *Aspergillus niger*.

Table 1: *In vitro* effect of trace elements on the growth and Sporulation of *Aspergillus niger* sensitive and resistant to Carbendazim causing fruit rot of Ber.

Trace element(0.03%)	Wild Sensitive		Resistant strain	
	Dia. of Colony(mm)	Sporulation	Dia. of Colony(mm)	Sporulation
Cobalt sulphate	-	-	1.66	+
Copper sulphate	-	-	-	-
Ferrous sulphate	-	-	16	+++
Magnesium sulphate	3.66	+	11.33	++
Manganese sulphate	-	-	7.00	++
Sodium sulphate	-	-	12.33	+++
Zinc sulphate	-	-	-	-
Calcium sulphate	-	-	18.00	+++
Control	6.66	+	13.33	+++
S.E.	1.50		2.09	
C.D. P .05	3.86		5.37	

+ = Few, ++ = Moderate, +++ = Good, ++++ = Abundant, - = Nil

DISCUSSION

Gangawane and Reddy (1986) showed that certain micronutrients when used singly or in mixture with carbendazim reduce resistance in *Aspergillus flavus*. 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 has been used.

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