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ORIGINAL ARTICLE

Isolation of Aflatoxigenic Strains of *Aspergillus flavus* Associated with Stored Seeds of Lentil In Agra

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ABSTRACT

20 strains of *Aspergillus flavus* isolated from stored samples of lentils, out of which 16 were found to be aflatoxigenic. The average value of chlorosis index of these strains showed reduction in percentage germination of seeds. 0.7% YES+ salt medium gave the best results.

INTRODUCTION

The aflatoxigenic metabolites of certain strains of Aspergillus flavus Link ex Fries associated with agricultural commodities are known to be carcinogenic and present a serious problem of health hazards in human beings and animals. Many agricultural products have been tested as substrates for producing aflatoxins by A. flavus such as groundnut, maize, rice, wheat, rye, jowar, soyabean etc. (Hesseltine, *et. al*., 1966). There are some reports relating the effect of aflatoxins on plant tissues (Joffe, 1969; Kang, 1970; Mehan & Chowhan, 1973, 1974; Chowhan & Gupta, 1968; Maggon *et. al*., 1969). The present investigation deals with the screening of aflatoxigenic strains of A. flavus associated with stored seeds/dals of lentil in Agra dal market.

MATERIAL AND METHODS

Number of seeds and dal sample of lentil (*Lens esculenta* Moench) were collected from different dal mills, godowns, shops in the market in rainy, winter and summer seasons separately. 200 seeds were examined for the fungal occurrence following agar plate technique by using M.S.A. medium (Clarke, 1969). The plates were incubated at 25˚ C and 35˚ C for 7 days so as to reveal mesophilic and thermophilic strains. The pure cultures were maintained on C.D.A. medium in slants. Only 20 strains of A. flavus were isolated and grown over three liquid media (Czapek's Dox, SMKY and 0.7% YES+salt) by inculcating with 0.5 ml spore suspension of each strains. Five replicates were set up and incubated for seven days at 28± 1˚C. After the incubation period the contents of each Erlenmayer flask were filtered through Whatman filter paper No. 1. Further the culture filtrate of each strain was screened for the presence of aflatoxins following bioassay technique developed by Kang (1970). Observations were made by multiplication of numerical value and numb r of plants showing the symptoms of chlorosis and the average values were calculated. The results are tabulated in Table 1.

RESULT AND DISCUSSIONS

Of the twenty strains listed, sixteen strains were found to be aflatoxigenic as judged by the chlorosis index. The non-toxigenic strains were LS 4±, LS 5, and LS 10 and LS 18 which did not show the symptoms or chlorosis. Isolate no. LS 6 was found to be most toxigenic as apparent from the Table l. The percentage germination of seeds was also affected showing a decrease with an increase in the average value of chlorosis.

Some cultural characteristics like nature of growth and sclerotia production were studied. The strains having deeper yellow to orange colour, wrinkled growth and larger sclerotial production favoured higher chlorosis which might be related to aflatoxin production. Isolated number LS 6 showed these characteristics. Mattles and Adye (1965) also observed a similar correlation between production of aflatoxin and yellowing mycelium and substrate. Similar observations have also been reported by Kang (1970) and Mehan & Chohan (1973) in relation to A flavus isolated from Peanuts.

Interestingly, these cultural characteristics are mostly found to be positive in 0-7% YES+salt medium. Therefore, this study reveals that 0·7% YES+salt medium is most suitable for the screening of aflatoxin production.

Table 1: Germination percentage of Okra seeds and average value of chlorosis after soaking culture filtrate for 10 hours of the strains of *A. flavus* associated with lentil

S.No.	Isolate No.	0.7% Yes+salt		Observation based on 50 seeds			
				Czapek's		SMKY	
		G%	A.V.	G%	A.V.	G%	A.V.
1	LS 1	70.0	0.38	80.0	0.04	76.0	0.10
2	LS 2	74.0	0.08	80.0	0.00	84.0	0.00
3	LS 3	80.0	0.96	88.0	0.00	84.0	0.12
4	LS 4	88.0	0.00	92.0	0.00	90.0	0.00
5	LS 5	56.0	0.00	66.0	0.00	68.0	0.00
6	LS 6	84.0	1.40	76.0	0.00	88.0	0.00
7	LS 7	76.0	0.32	82.0	0.00	84.0	0.00
8	LS 8	76.0	0.28	92.0	0.00	96.0	0.00
9	LS 9	80.0	0.32	84.0	0.00	88.0	0.00
10	LS 10	84.0	0.00	88.0	0.00	88.0	0.00
11	LS 11	96.0	0.68	100.0	0.00	92.0	0.10
12	LS 12	84.0	0.54	88.0	0.00	86.0	0.10
13	LS 13	84.0	0.32	88.0	0.04	92.0	0.00
14	LS 14	80.0	1.20	92.0	0.00	84.0	0.00
15	LS 15	64.0	1.36	80.0	0.00	76.0	0.20
16	LS 16	76.0	1.04	86.0	0.16	80.0	0.08
17	LS 18	72.0	0.00	76.0	0.00	84.0	0.00
18	LS 20	82.0	0.32	86.0	0.00	84.0	0.00
19	LS 21	70.0	0.24	84.0	0.02	78.0	0.08
20	LS 22	86.0	0.26	90.0	0.00	88.0	0.00
Control		100.0	0.00	100.0	0.00		100.0
(Sterile water)							
Control		100.0	0.00	100.0	0.00		100.0
(Uninoculated medium)							

G. % - Percentage germination
A.V. – Average value

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