

## Preharvest Aflatoxin Problem in Linseed (*Linum usitatissimum*) in Bihar

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A survey of different regions of Bihar was conducted for the natural occurrence of aflatoxins and *Aspergillus flavus* in linseed. Out of 198 samples collected, 13 had aflatoxins. In all, 67 isolates of *A. flavus* were obtained from the samples, of which 36 isolates were toxigenic and elaborated aflatoxins in liquid culture in the range of 2.86 to 24.34 mg/l.

Key Words - Preharvest Aflatoxin Linseed Bihar Toxigenic isolates *A. flavus*.

Aflatoxins were first considered to be exclusively a post harvest problem (Christensen & Kaufmann, 1969), but subsequent studies have changed this concept (Subrahmanyam & Rao, 1974; Anderson *et al.*, 1975). So far, the preharvest aflatoxin problem has been studied mostly in case of maize and peanuts. Linseed, an important oil-seed crop of India, particularly in Bihar, is reported to be a susceptible substrate for aflatoxin contamination under storage (Sahay, 1984).

We have made an extensive survey for the natural incidence of aflatoxins and aflatoxin producing fungi in preharvest linseed crop.

**MATERIALS & METHODS** During February-March, 1987, an extensive survey of linseed growing areas of Bihar covering 16 districts was made to collect the samples. Altogether 198 samples were collected from fields during harvest. Each sample comprised 1 kg of representative fruits from crops of one field. From each sample, 100 surface sterilized seeds were plated on Czapek's Dox agar medium and incubated at  $28 \pm 2^\circ\text{C}$  for 6 days. Colonies of *Aspergillus flavus* group were isolated, purified and screened for their toxin producing potential in SMKY liquid medium (Diener & Davis, 1966).

Dried seeds 250g (at  $60^\circ\text{C}$  for 2 days) from each lot were ground and mixed. Sub-samples of 50g were used for extraction of aflatoxin (Thomas *et al.*, 1975). Qualitative analysis of aflatoxins was done on TLC plate (Reddy *et al.*, 1970) which was subsequently confirmed using thionyl chloride as well as by spraying 25%  $\text{H}_2\text{SO}_4$  (Przybylski, 1975). The spectrophotometric method of Nabney & Nesbitt (1965) was followed for the quantification of aflatoxin  $\text{B}_1$ .

**RESULTS & DISCUSSION** Out of 198 linseed samples, association of *A. flavus* was recorded in 112 samples. However, only 13 samples were contaminated with aflatoxins (Table-1). Of the contaminated samples, 9 had aflatoxin  $\text{B}_1$  alone, while 4 samples contained aflatoxin  $\text{B}_2$  in association with  $\text{B}_1$ . The concentration of aflatoxin  $\text{B}_1$  in the contaminated samples ranged from 18-460  $\mu\text{g}/\text{kg}$ . Out of 67 isolates of *A. flavus* isolated from contaminated linseed samples, 36 isolates were toxigenic (Table-1). Among the toxin producers the predominant aflatoxin  $\text{B}_1$  was elaborated by 19 isolates of *A. flavus*, while 13 isolates produced both aflatoxin  $\text{B}_1$  and  $\text{B}_2$ . Four isolates produced aflatoxin  $\text{G}_1$  also, but along with  $\text{B}_1$  and  $\text{B}_2$ . These toxigenic isolates elaborated aflatoxin  $\text{B}_1$  in the concentration ranged from 2.86-24.34 mg/l.

Table 1. Aflatoxin Incidence in Linseed Crop and Toxin Producing Potential of *A. flavus* Isolates

Aflatoxin : contamination / producing potential of	No. of : Seed samples / <i>A. flavus</i> isolates		Qualitative assay			Range of aflatoxin B <sub>1</sub>
	Analysed / Screened	+ve to aflatoxin	B <sub>1</sub>	B <sub>1</sub> B <sub>2</sub>	B <sub>1</sub> B <sub>2</sub> G <sub>1</sub>	
Seed	198	13	9	4	-	18-460 (μg/kg)
<i>A. flavus</i>	67	36	19	13	4	2.86-24.34 (mg/l)

Preharvest aflatoxin problem in linseed might be due to several factors including environmental, agronomic and biological. During fruit-development up to maturity of linseed crop, the climatic conditions remain congenial for *A. flavus* development. The principal source of inoculum might be conidia produced from the mycelium of the fungus that overwintered in plant debris in the soil (Jones, 1979). Another important source of primary inoculum might be the sporogenic germination of sclerotium with conidiophores and conidia. Wicklow & Donahue (1984) observed 5 of 7 *A. flavus* strains formed conidial heads on surface-sterilized sclerotia after 48 to 72 h of incubation on moist sand.

The difference in toxin producing ability of toxigenic strain may be considered as one of the important biological factors leading to variable degree of aflatoxin production in seed samples.

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