

## Short Communication

# Elaboration of mycotoxins by seed-borne fungi of finger millet (*Eleusine coracana* L.)

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**Mycotoxin producing potential of fungi associated with finger millet was investigated. Many species of *Aspergillus* elaborated aflatoxins, patulin, terreic acid and sterigmocystin, while species of *Fusarium* elaborated zearalenone, fusarinone-X, deoxynivalenol, nivalenol, diacetoxyscripenol, neosolanil and HT-2 toxins. *Penicillium griseofulvum* elaborated cyclopiazonic acid. The toxigenic potential of individual fungus varied.**

**Key words:** Mycotoxins, seed-borne fungi, finger millet.

## INTRODUCTION

Deoxynivalenol (DON) and nivalenol (NIV) are a group of closely related secondary fungal metabolites, that are produced predominantly, although not exclusively, by several species of the genus *Fusarium*, especially *F. graminearum*. Finger millet, known as 'ragi' in India is an important staple food for people belonging to the low socio-economic group, several reports have shown that millet (Pathak et al., 2000) are inexpensive and nutritionally comparable or even superior to major cereals. Regular consumption is known to reduce the risk of diabetes mellitus (Gopalan, 1981) and gastro intestinal tract disorders (Tovey, 1994). These seeds are vulnerable to the huge diversity of opportunistic microbes especially the *Fusarial* species, further, it is anticipated seeds that are more vulnerable to mycotoxin contamination.

Though, there are several recent reports of infestation of finger millet by mycotoxin, producing fungi from different parts of the World (Nikema et al., 2004; Ana et al., 2009). There are only limited studies in India (Rajan et al., 2006; Vinod kumar et al., 2008). Further, very limited information is available from this region. Moulds, besides depleting the nutrients, may also produce toxic substances that are potential health hazards to animals and, in turn to humans (Fazekas et al., 1996; Trucksess, 2001). These mycotoxins can be very stable to food processing (Molini'e et al., 2005) and be present in final products. Hence, an attempt has been made to study the

toxigenic potentials of different fungi associated with finger millet in different parts of the state. Majority of the *Fusarium* species produce trichothecene mycotoxins. Trichothecenes are esters of sesquiterpenoid alcohols containing the trichothecene tricyclic ring system (Pestka and Smolinski, 2005).

## MATERIALS AND METHODS

A total of 110 pre-packaged samples were selected from local retail commerce in Andhra Pradesh of India. No particular preference was used in selecting samples or locations. The sample size was 250 gm were analyzed. The mycoflora of seed as well as the mycotoxins were assayed.

Fusarial mycotoxins were analyzed using thin layer chromatography (TLC). For this purpose, fusarial culture filtrates were extracted twice with ethylacetate (2 × 50). The combined extracts were passed through an anhydrous Na<sub>2</sub>SO<sub>4</sub> bed to remove moisture and then evaporated to dryness before dissolving in 1 ml of methanol and spotting onto the TLC plates. The toxins were identified by spraying the plates with different spray reagents (Table 1) as suggested by Kamimura et al. (1981); Ramakrishna et al. (1985), and the compounds thus separated were identified based on the color of the fluorescence of the spot and by the R<sub>f</sub> values, as compared with standards. The R<sub>f</sub> was calculated by using the formula:

$$R_f = \frac{\text{Distance travelled by the compound}}{\text{Distance travelled by the solvent}}$$

## RESULTS AND DISCUSSION

From Table 1, it is evident that, most of the fungal strains

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**Table 1.** Incidence of mycotoxin producing fungi associated with finger millet.

Name of the fungus	Number of strains	Number of strains producing toxin	Solvent system	Fluorescence under UV		Rf value	Chemical conformation	Mycotoxin
				Before spray	After spray			
<i>Aspergillus flavus</i>	120	79	C:A (90:10)	Blue green	-	B1-0.42 B2-0.40 G1-0.32	-	Aflatoxins
<i>Aspergillus nidulans</i> i.e solvent	28	22	C:M:A (1:01:01)	Dull brick	Yellow	0.91	AlCl <sub>3</sub> (Ramakrishna et.al., 1985)	Sterigmocystin
<i>Aspergillus terreus</i>	36	21	T:Ea:F (50:40:10)	Dark brown	Yellow	0.45	2% Phenylhydrazene hydrochloride	Patulin or Terreic acid
<i>Fusarium</i> spp. <i>F. usarium oxysporum</i> , <i>F. moniliforme</i> , <i>F. graminearum</i> , <i>F. culmorum</i> , <i>F. sporotrichoides</i>	48	24	Ea:T:F (50:40:10)	Green	Blue	0.63 0.35 0.21 0.78 0.42	-	Zearalenone Fusarinone-X Deoxynivalenol Nivalenol Neosolaniol,HT-2
<i>Trichothecium roseum</i>	5	3	C:M (98:2)	Blue Blue-green	Pink	0.97 0.65	Phloroglucinol	Trichothecin
<i>Trichoderma viride</i>	2	2	C:M (98:2)	Blue	Pink	0.97	Phloroglucinol Resorcinol	Trichodermin Gliotoxin
<i>Penicillium griseofulvum</i>	8	4	C:Ea:F:T (50:40:10:2)	Dark brown	Purple	-	FeCl <sub>3</sub>	Cyclopiazonic acid

C = chloroform; A = acetone; M = methanol; T= toluene; Ea = ethylacetate; F = formic acid.

isolated from finger millet produced one or the other mycotoxin. However, aflatoxins producing fungi were dominant. The incidence of toxigenic strains of *A. nidulans* and *A. terreus* come next. Species of *Fusarium* also elaborated variety of

toxins of which Zearalenone was most common. On the other hand, Vesonder et al. (1978) reported that most of the food samples were contaminated with deoxynivalenol. Other trichothecene producing fungi not only formed

minor production of spermosphere but also their toxigenic potentials were limited. Doohan et al. (2003) commented that production of trichothecenes by *F. culmorum* and *F. graminearum* is favoured by warm and humid conditions. The

trichothecenes including DON, NIV and fusarenone-X, are common fungal contaminants of millets (Magan and Olsen, 2004; Jennings et al. (2000). Most of the species of *Penicillium* were non-toxigenic and hence less hazardous. Present observations are similar to those of Bilgrami et al. (1981) , Reddy and Reddy (1983), Girisham et al. (1985) who also observed the association of fungi with seeds of maize, sesamum and Pearl millet respectively with varied mycotoxigenic potentials and pose a threat to the health to man.

From the present investigations it can be concluded that variety of fungi harboring finger millet seeds are potentially toxigenic and not only hazardous directly to man but also may be responsible for diseases of poultry and livestock. Consumption of these toxins is a potential problem for humans and farm animals (Erisken et al., 1998; Rotter et al., 1996). Thus these fungi may be responsible for primary and secondary mycotoxicoses in man. Hence, more detailed investigations are desired in order to suggest measures to check the mould infestation of finger millet grains/product.

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