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Influence of different *Fusarium* species on seed germination and seedlings growth of finger millet (*Eleusine coracana* L.)

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ABSTRACT

Effect of agriculturally important *Fusarium* species on the seed germination and seedling growth of finger millet (*Eleusine coracana* L.) was investigated. Species of *Fusarium* caused significant seed germination inhibition and seedling growth which varied with the species and age of the culture. Culture filtrates of *F. moniliforme*, *F. proliferatum*, *F. chlamydosporum*, *F. aethiopicum*, *F. heterosporum* and *F. sporotrichoides* were comparatively more toxic. The correlation coefficients between polished and unpolished variety of finger millet seed germination inhibition (0.574, $P=0.005$), shoot elongation inhibition (0.893, $P=0.0000$) and root elongation inhibition (0.175, $P=0.1770$) with culture filtrates of different species of *Fusarium* was recorded. Pathogenicity studies revealed that *F. roseum*, *F. sporotrichoides*, *F. proliferatum* and *F. oxysporum* caused seed-rot and seedling death. The maximum root elongation inhibition (52.20%), mean (36.39%) and minimum (14.73%) were recorded toward the toxicity of different species of *Fusarium*. Significant and positive correlation (0.802, $P=0.0026$) between the root and shoot, (0.393, $P=0.130$) between shoot and leaf, (0.121, $P=0.369$) between root and leaf elongation inhibition of finger millet could be observed. The culture filtrates of major mycotoxigenic strains of *Fusarium* revealed production of Zearalenone (ZEA), T2 toxin, nivalenol (NIV), Deoxynivalenol (DON) and Deoxyscripenol (DAS). Toxicity of *Fusarium* species to its seed germination and seedling growth may be attributed to their secondary metabolites including mycotoxins.

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KEYWORDS

Finger millet;
Fusarium species;
 Culture filtrates;
 Seed germination;
 Root elongation;
 Shoot elongation;
 Leaf elongation;
 Mycotoxins

INTRODUCTION

Finger millet (*Eleusine coracana* L.), commonly called as Ragi (India), is the staple food for tribal peoples, especially in dry areas of India, Nepal, Srilanka and china^[1]. It is rich in nutrients such as proteins, phosphorous, calcium, iron, thiamine, amino acid and fibre.

It is a gluten-free food and has strong therapeutic agent against liver diseases, high-blood pressure, heart weakness and asthma^[2], nourishing food for infants and invalids and is wholesome food for diabetics^[3].

In recent times considerable attention is being given to seed-borne fungi of this crop^[4-6]. Seriousness of the problem of mycotoxins was realized only in recent times

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probably their association with several mysterious diseases associated with food^[7]. The substrates for the mycotoxigenic fungi include plants grown and stored for human or animal consumption as well as processed food. There is also a growing awareness of the mycotoxins presence in the living and working environment^[8,9]. In the prevailing sub-tropical climatic conditions of this region, incidence of *Fusarium* species is reported to be significant in the cereal food chain, capable of reducing crop yields and elaborate mycotoxins such as trichothecenes, fumonisins (FB1, FB2 and FB3) and Zearalenone^[10].

Fusarium species, besides causing different diseases retard the seed germination and seedling growth^[11]. Seedling blight and head blight of *Fusarium* in small grain cereals is common in many parts of the world^[12]. *F. graminearum* and *F. culmorum* are the most important and frequently associated with different crops^[13] and posing serious health risks^[14]. Though incidence of fungi on finger millets has been reported from different parts of the world^[15-19], not much information is available on the incidence of mycotoxigenic fungi of finger millet. The present study aimed to understand the nature of association and role of the species of *Fusarium* on hulled (unpolished) and dehulled (polished) seeds of finger millet.

MATERIALS AND METHODS

Fungal cultures

Ten species of *Fusarium* (*F.aethiopicum*, *F.chlamydosporum*, *F.culmorum*, *F.heterosporum*, *F.moniliforme*, *F.oxysporum*, *F.poa*, *F.proliferatum*, *F.roseum* and *F.sporotrichoides*) associated with finger millet^[20] were maintained on Spezieller-Nahrstoffarmer Agar (SNA) and preserved at -4°C were employed for assessing their pathogenicity to finger millet.

Culture filtrates of *Fusarium* species on seed germination and seedling growth

Two varieties of seeds (polished and unpolished) were surface sterilized with 0.1% mercuric chloride and rinsed three times in sterile distilled water. The culture filtrates of different species of *Fusarium* as listed in table were collected from cultures grown in Spezieller-

Nahrstoffarmer broth (SNB) for 20 days at 27±2°C. Cultures filtrates were centrifuged at 12,000g to get cell-free filtrates. Hundred healthy surface sterilized kernels were suspended in 50ml of culture filtrates for 24 hours at 27±2°C and transferred to sterile-petri plates (9cm diameter) containing three layered wet blotter paper at the rate of 10 seed per plate and incubated for 5 days under illumination. Seeds soaked in un-inoculated broth served as control. At the end of incubation period, seed germination, shoot length and root length were measured over the control and their inhibition percentage was calculated with the formulae.

Percentage of seed germination inhibition = 100 -

$$\frac{\text{Germination in treated seed}}{\text{Germination in control seed}} \times 100$$

Percentage of root elongation inhibition = 100 -

$$\frac{\text{Root elongation inhibition in treated}}{\text{Root elongation inhibition in control}} \times 100$$

Percentage of shoot elongation inhibition = 100 -

$$\frac{\text{Shoot elongation inhibition in treated}}{\text{Shoot elongation inhibition in control}} \times 100$$

Percentage of leaf elongation inhibition = 100 -

$$\frac{\text{Leaf elongation inhibition in treated}}{\text{Leaf elongation inhibition in control}} \times 100$$

Pathogenicity of *Fusarium* species by water agar method

Water-agar (WA) method as described by Girisham *et al.*^[21] was employed for testing the pathogenic potential of different species of *Fusarium*. Polished seeds were surface sterilized with 0.1% mercuric chloride and rinsed three times in sterile distilled water and placed on two percent (2%) sterilized water-agar (WA) in 25ml of culture tubes along with seven days old culture and incubated at 27±2°C for two weeks under illumination. Surface disinfected seeds without fungal inoculum were served as control. At the end of incubation period, root, shoot and leaf length were measured over the control and their inhibition percentage was calculated.

Profiling of toxin chemotypes of species of *Fusarium*

TLC analysis of toxin chemotypes

Single spore cultures of different species of *Fusarium* (listed) were grown in SNB broth for 10, 15 and 20 days at 27±2°C on rotary shaker (Yihder LM-450 D) at

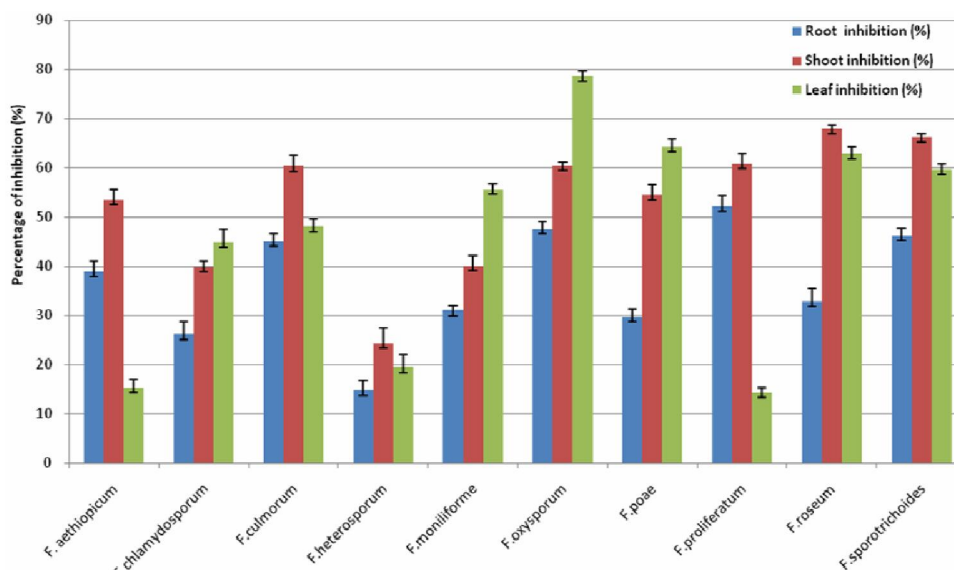


Figure 1 : Pathogenicity of species of *Fusarium* on seedling growth of finger millet

120 rpm. At the end of incubation period, the culture filtrates were harvested on Whatman No.1 filter paper and culture filtrates were centrifuged at 12,000g to get cell-free filtrates. The culture filtrates were acidified with 0.1M *o*-phosphoric acid and extracted twice with ethyl acetate (1:1, v/v) and concentrated by rotary evaporator and eluted in 1ml of methanol. The TLC plates were activated by immersing in oxalic acid solution (10% oxalic acid in methanol) for 5-10 min and heated at 110°C for 2min and allowed to cool at room temperature. Eluted 20µl extracts were spotted on activated TLC plates and then developed in suitable solvent system. The different mycotoxins produced by *Fusarium* species were identified by the colour of the fluorescence under short wave light (360 nm), and chemically confirmed by different spray reagent for different toxin-chemotypes described earlier^[20].

Statistical analysis

The results are analysed statistically by applying one sample t test to compare the toxicity of fungi on seed germination, root, shoot, and leaf elongation inhibition variation at ($P < 0.005$) using GraphPad InStat version 5.03 (GraphPad Software, Inc.,)

RESULTS

Effect of culture filtrates of species of *Fusarium* on seed germination inhibition

The culture filtrates of different seed-borne species

of *Fusarium* of finger millet exhibited toxicity towards the seed germination and seedling growth. However, the degree of toxicity varied with both the variety of seed and the age of fungal culture filtrates depicted in TABLE 1. Twenty day aged culture filtrates of *F. moniliforme* and *F. proliferatum* were responsible for total (100%) germination inhibition of polished variety. Culture filtrates of *F. aethiopicum* and *F. chlamydosporum* were next in their toxicity towards seed germination of finger millet. Culture filtrates of *F. heterosporum* and *F. roseum* were least toxic, while culture filtrates of rest of species of *Fusarium* exhibited intermediate toxicity towards the seed germination of polished finger millet. Seed germination inhibition of unpolished variety was significantly high with 20 day aged culture filtrates of *F. aethiopicum*, *F. proliferatum*, *F. sporotrichoides* and *F. moniliforme* in a descending order and ranged between 94 and 97%. Least inhibition was recorded with *F. poae* and *F. oxysporum*. Culture filtrates of rest of the *Fusarium* species exhibited only mild toxicity and caused minimum seed germination inhibition of unpolished variety of finger millet. Toxicity of different species of *Fusarium* was statistically analysed and their mean, maximum, minimum and standard deviation for each experiment are precised in TABLE 2. The highest seed germination inhibition (100%) and lowest (16.20%) inhibition were recorded in the polished finger millet, while the lowest seed germination (24.50%), and the highest mean (97.60%) were found in unpolished finger millet. Mean

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TABLE 1: Effect of culture filtrates on seed germination, seedling growth of finger millet

<i>Fusarium</i> species	Age of culture filtrates (day)	Seed germination inhibition (%)		Root elongation inhibition (%)		Shoot elongation inhibition (%)	
		Polished	Unpolished	Polished	Unpolished	Polished	Unpolished
<i>F.aethiopicum</i>	10	39.8	68.6	19.8±5.6	36.7±4.3	26.9±5.8	32.9±2.7
	15	69.2	89.3	39.4±4.6	59.2±5.1	48.2±5.3	38.2±3.7
	20	89.7	97.6	48.9±2.9	71.2±5.9	69.6±4.4	59.6±2.3
<i>F.chlamydosporum</i>	10	32.8	65.2	29.6±5.1	32.7±5.7	28.4±3.7	31.7±4.3
	15	66.2	81.2	39.7±3.1	51.9±4.2	46.8±4.2	39.4±5.1
	20	89.6	91.3	49.3±5.2	71.2±4.5	61.6±2.9	46.2±5.9
<i>F.culmorum</i>	10	28.2	42.6	16.4±5.6	32.7±5.8	24.6±2.7	19.4±5.1
	15	46.6	54.9	19.3±4.2	51.9±2.6	28.2±3.7	23.2±3.2
	20	81.8	88.2	27.4±4.4	70.2±3.4	32.6±2.3	32.8±5.3
<i>F.heterosporum</i>	10	29.2	43.9	19.3±5.9	56.2±6.0	14.2±5.6	19.6±5.9
	15	42.6	69.2	29.1±2.7	71.6±3.4	29.2±4.3	29.4±4.4
	20	63.1	84.9	36.2±3.5	82.3±4.3	28.3±2.9	36.9±2.3
<i>F.moniliforme</i>	10	72.6	32.6	36.1±4.4	17.2±5.2	36.3±2.6	36.6±5.6
	15	80.2	61.2	42.9±5.2	48.1±2.6	65.3±3.4	66.8±4.3
	20	100.0	94.3	66.3±6.1	74.3±3.4	69.8±2.1	89.5±4.4
<i>F.oxysporum</i>	10	22.8	29.6	20.2±4.2	14.6±5.3	17.1±5.2	14.1±3.8
	15	43.6	38.2	26.1±5.3	20.9±3.9	27.2±4.3	27.6±4.1
	20	74.8	45.5	26.5±5.9	31.2±5.7	27.5±2.8	29.1±2.7
<i>F.poa</i>	10	21.3	29.9	20.6±5.3	15.7±5.3	24.6±2.5	16.4±5.5
	15	51.2	28.3	23.3±3.4	19.2±3.9	29.2±3.6	24.3±4.3
	20	72.3	39.7	28.7±5.8	22.6±5.8	31.2±2.1	33.9±2.7
<i>F.proliferatum</i>	10	39.8	48.2	53.6±5.3	17.3±5.2	42.6±2.7	43.9±3.6
	15	84.3	81.3	81.1±2.9	22.3±3.9	71.4±3.6	76.2±4.1
	20	100.0	96.2	85.1±4.3	26.2±5.1	74.2±2.1	79.3±2.7
<i>F.roseum</i>	10	21.5	24.5	19.8±5.2	18.4±5.2	23.8±3.6	28.4±5.5
	15	48.2	46.8	22.3±3.9	28.6±3.9	27.2±4.2	44.8±4.3
	20	71.6	72.6	29.7±5.9	30.2±5.9	32.3±2.8	56.2±2.9
<i>F.sporotrichoides</i>	10	16.2	79.3	20.8±2.5	19.8±5.9	24.8±5.8	26.8±5.2
	15	32.6	84.2	26.3±3.6	23.2±4.1	28.2±4.1	29.2±2.5
	20	64.8	96.1	30.1±2.1	30.7±5.1	33.2±5.3	39.4±5.8

Results are triplicate experiments were expressed in mean and standard deviation

seed germination elongation inhibition (56.55%) and (63.51%) were recorded in both polished and unpolished finger millet respectively. Significant difference ('t' test) on toxicity of *Fusarium* species on seed germination inhibition of polished (12.29%) and unpolished (14.31%) finger millets was recorded. A positive correlation (0.574, $P=0.0005$) and simple regression (0.3296) were recorded between the polished and unpolished seeds.

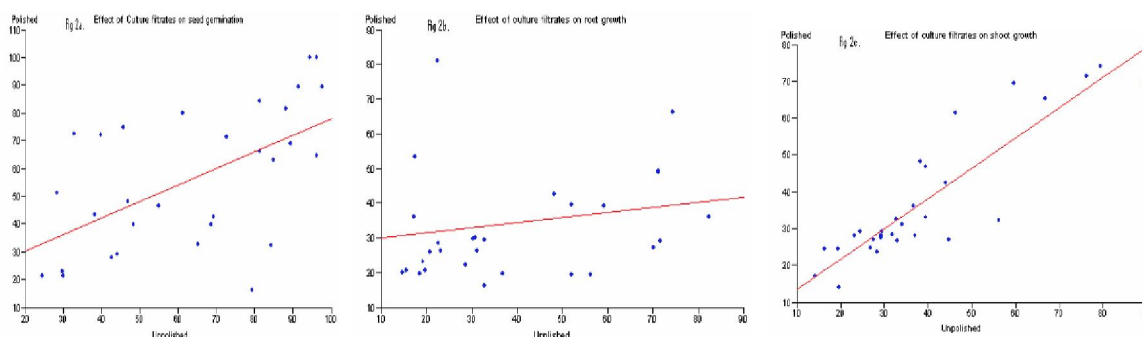
Effect of culture filtrates on root elongation inhibition

bition

Maximum root elongation inhibition was recorded with 20 day aged culture filtrates of *F. proliferatum* (85.1%) followed by *F. moniliforme* (66.3%), *F. chlamydosporum* (49.3%) and *F. aethiopicum* (48.9%), while least inhibition was recorded with *F. oxysporum*, *F. culmorum* and *F. poae* in a descending order. Culture filtrates of rest of the species of *Fusarium* were exhibited intermediate toxicity on root elongation of polished variety of finger millet. On the

TABLE 2 : Statistical analysis of culture filtrates of species of *Fusarium* on seed germination and seedling growth of finger millet

	Seed germination (%)		Root elongation inhibition (%)		Shoot elongation inhibition (%)	
	Polished	Unpolished	Polished	Unpolished	Polished	Unpolished
Minimum	16.20	24.50	16.40	14.60	14.20	14.10
25% Percentile	32.75	41.88	20.75	20.63	27.13	27.40
Median	57.15	66.90	28.90	30.95	29.20	33.40
75% Percentile	76.15	85.73	40.50	56.95	47.15	45.15
Maximum	100.0	97.60	85.10	82.30	74.20	89.50
Mean	56.55	63.51	34.46	38.94	37.48	39.06
Std. Deviation	25.20	24.31	17.74	21.42	17.42	18.90
Std. Error	4.600	4.438	3.238	3.911	3.181	3.451
One sample t test						
t, df	12.29	14.31	10.64	9.957	11.78	11.32
P value (two tailed)	< 0.0001	< 0.0001	< 0.0001	< 0.0001	< 0.0001	< 0.0001
CV*	44.55%	38.28%	51.47%	55.01%	46.49%	48.39%
Correlation	0.574	--	0.175	--	0.893	--
Regression	0.3296	--	0.0308	--	0.7966	--

CV*Coefficient of variation**Figure 2 : Correlation coefficient of polished and unpolished finger millets against culture filtrates of species of *Fusarium* on seedling growth; a: Correlation coefficient of seed germination, b: root growth and c: shoot growth of finger millet.**

other hand, highest root elongation inhibition of unpolished variety was recorded with culture filtrates of *F. heterosporum* followed by *F. chlamydosporum*, *F. aethiopicum* and *F. moniliforme*. *F. poae*, *F. proliferatum* and *F. oxysporum* exhibited least toxicity. Culture filtrates of rest of the species of *Fusarium* were mild in their toxicity on root elongation of unpolished variety of finger millet. The root elongation inhibition of polished finger millet ranged between (85.10%) and (16.40%), while inhibitory activity of different species ranged between (14.60%) and (82.30%) in unpolished finger millet. Mean root elongation inhibition of polished (34.46%) and unpolished (38.94%) finger millet was recorded. Significant difference of toxicity

among *Fusarium* species on root elongation inhibition of polished (10.64%), unpolished (9.95%) finger millet was recorded. Significant correlation coefficient (0.175, $P=0.1770$) and simple regression (0.0308) was observed between the polished and unpolished finger millet.

Effect of culture filtrates on shoot elongation inhibition

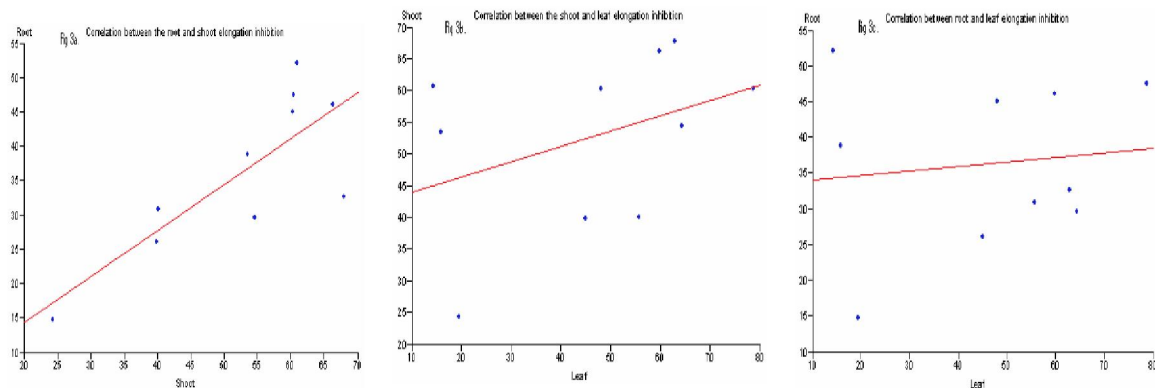
The toxicity of *F. proliferatum*, *F. moniliforme*, *F. aethiopicum* and *F. chlamydosporum* was maximum and caused shoot elongation inhibition (60-74%) of polished finger millet, while *F. oxysporum* and *F. heterosporum* were responsible for least inhibition. Culture filtrates of rest of the species of *Fusarium* were

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TABLE 3 : Statistical analysis of pathogenicity of species of *Fusarium* on seedling growth of finger millet

	Root elongation inhibition	Shoot elongation inhibition	Leaf elongation inhibition
Minimum	14.73	24.26	14.30
Median	35.82	57.39	51.80
Maximum	52.20	67.92	78.60
Mean	36.39	52.78	46.34
Std. Deviation	11.61	13.89	22.58
Std. Error	3.672	4.392	7.140
One sample t test			
t -value	9.910	12.02	6.490
P value (two tailed)	< 0.0001	< 0.0001	0.0001
CV*	31.91%	26.31%	48.72%
Correlation	0.802/0.0026	0.393/0.130	0.121
Regression	0.6434	0.1547	0.0147

CV*Coefficient of variation

Figure 3 : Correlation coefficient of pathogenicity of species of *Fusarium* on seedling growth of finger millet ; a: Correlation coefficient between root and shoot, b: Shoot and leaf growth and c: Root and leaf growth of finger millet

intermediate in their toxicity and caused moderate shoot elongation inhibition of polished variety of millet, while shoot elongation inhibition of unpolished variety of millet was between 45 and 90% under the influence of culture filtrates of *F. moniliforme*, *F. proliferatum*, *F. aethiopicum* and *F. roseum*. Culture filtrates of *F. oxysporum*, *F. culmorum* and *F. aethiopicum* were least in their toxicity in a descending order. Culture filtrates of rest of the species of *Fusarium* were intermediate in their toxicity on shoot elongation inhibition of finger millet. Culture filtrates of species of *Fusarium* were stimulatory to seed germination and seedling growth of both the varieties. The highest shoot elongation inhibition (74.20%) and least (14.20%) elongation inhibition were recorded in the polished finger millet, while the lowest root elongation inhibition was recorded (14.10%) and the highest mean (89.50%) were in un-

polished finger millet. Mean shoot elongation inhibition of polished (37.48%) and unpolished (39.06%) finger millet was recorded. Significant difference on toxicity among species of *Fusarium* on shoot elongation inhibition of polished (11.78%) and unpolished (11.32%) was recorded on both the varieties. Significant correlation coefficient (0.893, $P=0.0000$) and simple regression (0.7966) was observed between the polished and unpolished finger millet.

Pathogenicity of different species of *Fusarium* on seedling growth

Effect of species of *Fusarium* on root elongation

Testing the pathogenicity of different species of *Fusarium* against finger millet revealed that *F. proliferatum*, *F. oxysporum* and *F. sporotrichoides* were significantly pathogenic and their pathogenicity

ranged between 46 and 52% while *F. heterosporum*, *F. chlamydosporum* and *F. poae* were mild pathogenic. Rest of the species of *Fusarium* exhibited intermediate in their pathogenicity against root elongation of finger millet. Pathogenicity of different species of *Fusarium* was statistically analysed and their mean, maximum, minimum and standard deviations for each experiment was calculated and precised in TABLE 2. The maximum root elongation inhibition (52.20%), mean (36.39%) and minimum (14.73%) were recorded with different *Fusarium* species. Significant difference of pathotoxicity ('t' test) of *Fusarium* species towards the root elongation inhibition (9.910 %), correlation coefficient (0.802, $P=0.0026$) and simple regression (0.6434) were observed between the root and shoot elongation inhibition of finger millet.

Effect of species of *Fusarium* on shoot elongation

F. roseum was highly pathogenic (67%) and caused seed-rot and seedling death, while *F. heterosporum* and *F. chlamydosporum* were least pathogenic and their pathogenicity ranged between 24 and 40%. *F. sporotrichoides*, *F. proliferatum*, *F. culmorum* and *F. oxysporum* were either nonpathogenic or caused marginal inhibition of shoot elongation of finger millet in a descending order. The pathogenicity ranged between 67.92% and 24.26% shoot elongation by different *Fusarium* species. Significant difference ('t' test) of pathotoxicity of *Fusarium* species towards the root elongation inhibition ($t=12.02\%$), correlation coefficient (0.393, $P=0.130$) and simple regression (0.1547) were observed between the shoot and leaf elongation inhibition of finger millet.

Effect of species of *Fusarium* on leaf elongation

Leaf elongation inhibition was significantly high in seeds inoculated with *F. oxysporum*, *F. poae* and *F. roseum*, while *F. proliferatum*, *F. aethiopicum* and *F. heterosporum* were responsible for least inhibition of leaf elongation of finger millet. Rest of the species of *Fusarium* were responsible for intermediate leaf elongation inhibition. The highest shoot elongation inhibition (78.60%), while least (14.30%) were caused by *Fusarium* species on leaf elongation. Significant difference in the pathotoxicity of different *Fusarium* species towards the leaf elongation ($t=6.490\%$), correlation

coefficient (0.121, $P=0.369$) and simple regression (0.0147) were observed between the root and leaf elongation inhibition of finger millet.

Profiling of mycotoxin-chemotypes

The chemical analysis of culture filtrates of different species of *Fusarium* revealed the production of different mycotoxins. Some species produced more than one mycotoxin chemotypes. In contrast different species of *Fusarium* produced same mycotoxin chemotype. Based on mycotoxins production pattern, species of *Fusarium* are classified in to 6 chemotypes. Chemotype I represented by *F. chlamydosporum*, *F. culmorum*, *F. oxysporum*, *F. moniliforme*, *F. solani*, *F. sporotrichoides*, *F. poae* and *F. proliferatum* produced zearalenone (ZEA), while chemotype II represented by *F. chlamydosporum*, *F. oxysporum*, *F. solani*, *F. sporotrichoides* synthesized T2 toxin. *F. chlamydosporum*, *F. poae*, *F. heterosporum* and *F. subglutinans* produced diacetoxyscirpenol (DAS) represents chemotype III. Chemotype IV represented by deoxynivalenol (DON) which is produced by *F. chlamydosporum*, *F. culmorum*, *F. graminearum*, *F. oxysporum*, *F. moniliforme* and *F. solani*. Chemotype V represented by *F. culmorum*, *F. graminearum*, *F. solani*, *F. latertium*, *F. sporotrichoides* and *F. heterosporum* produced nivalenol (NIV). Chemotype VI represented by *F. oxysporum* and *F. moniliforme* and *F. heterosporum* produced FB. Toxicity of these species of *Fusarium* on germination inhibition and retardation of the seedling growth (coleoptiles, radicle and leaf length) may be attributed to the production of any of above toxins or other minor or unidentified chemotypes produced by these fungi.

DISCUSSION

The results of the present investigations on pathogenicity revealed seed germination capacity and seedling growth of finger millets were adversely affected by one or other species of *Fusarium* to a varying degree. The adverse affect of *Fusarium* species growing on surface or inside the seed of finger millet^[22]. The water activity and early infection of *Fusarium* species are likely to reduce the germination of finger millets. Late infec-

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tion is likely to reduce the germination capacity only to certain extent^[12]. Culture filtrates of *F. proliferatum*, *F. aethiopicum* and *F. moniliforme* were comparatively more toxic. *Fusarium* species were pathotoxic to finger millets and retard shoot and root growth and comparatively more in unpolished variety than in polished seeds. A positive and significant correlation on between seed germination and shoot elongation, while non-significant and negative (weak) correlation was observed root elongation inhibition was recorded between the polished and unpolished seeds. This condition may be attributed caused to physiological inhibition mechanism of embryo^[23].

The chemical analysis of culture filtrates of *Fusarium* species revealed that production of ZEA, DON, NIV, FB and HT2 toxin. The high levels of DON may responsible for decrease in the 30% of crop yield, attributed the decrease in crop yield in maize to DON production by *Fusarium* species^[24]. The environmental factors like temperature and relative humidity are reported to plays a major role in infestation of *Fusarium* species and elaborates mycotoxin chemotypes and reduce the seed germination and seedling growth^[25,26]. The present observations are in agreement with Narasimha Rao *et al.*^[27] who also recorded inhibition of seed germination of maize under influence of culture filtrates of species of *Fusarium*.

Fungal infestation in unpolished was comparatively more than in polished seeds which may be attributed to the moulds resting in the husk^[28], or the presence of nutrients^[29,30]. Reddy *et al.*^[31] who also reported that rice bran supported 14% more fungal infestation than its products. However, no difference was observed in seedling growth of hulled and dehulled finger millets. Only marginal difference was observed in shoot elongation inhibition with hulled and dehulled finger millet. Rashmi^[32] also recorded inhibition of seed germination of sorghum by its seed-borne fungi. The present investigations are in agreement with Kumar *et al.*^[33] who also recorded increased toxicity of culture filters with the age of fungus. The present findings can be positively correlated with Duverger *et al.*^[34] who also reported the increased production of DON and ZEA by *F. graminearum* with the progress of age^[35]. Alberts *et al.*^[36] recorded increasing FB1 and FB2 production with increasing age of *F. moniliforme*. In general pol-

ished variety was more resistant than the unpolished variety as observed by Schwarz *et al.*^[37] and Tekle *et al.*^[12].

Fusarium species reduced seed germination to varying degrees resulting seedling mortality and progressive disease development in the field and thereby reduced the yield and poor quality grain. The present findings are significant as they caused significant loss in agricultural products. Erpelding and L. Prom^[38], Fakhrunnisa *et al.*^[39] have also reported these species in different agricultural products. *F. proliferatum*, *F. oxysporum* and *F. sporotrichoides* were significantly toxic towards finger millet and positive correlation could be observed between the root and shoot, while, non-significant and weak correlation between the shoot and leaf, and root and leaf elongation inhibition respectively. Gachomo *et al.*^[40], Marley^[41], Mathur and Kongadal^[42] have also reported reduced seed germination of peanut seed infected with seed-borne fungi studied by them.

CONCLUSIONS

From the present investigations it can be concluded that many species of *Fusarium* associated with seeds of finger millet adversely affected the seed viability and seedling growth. However, in depth studies are needed to elucidate the real mechanisms of toxicity of species of *Fusarium* on seed germination and seedling growth involving large number of species of *Fusarium* and different seed varieties.

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REFERENCES

- [1] A.B.Obilana, E.Manyasa; Millets. In: Pseudo cereals and less common cereals. Grain properties and utilization potential P.S.Belton and J.R.N.Taylor eds.Springer-Verlag, Berlin, 176-217 (2002).
- [2] S.Sen, S.K.Dutta, S.Ghosh Dastidar; Int.J.Biomed.Pharmaceu.Sci., 5, 7-11 (2011).

- [3] S.Sen, S.K.Dutta; The European J.Plant Sci.Biotech., **6**, 103-108 (2012).
- [4] A.A.Ansari, A.K.Shrivastava; Indian.J.Agric.Sci., **61**, 228-229 (1991).
- [5] S.Navi, C.Reddy; APS Abstracts Phytopath., **95**, S74 (2005).
- [6] K.Saveetha, A.Sankaralingam, A.Ramanathan, R.Pant; Arch.Phytopath Pl.Prot., **39**, 409-419 (2006).
- [7] J.L.Richard; Inter. J. Food Microbiol., **119**, 3-10(2007).
- [8] B.Gutarowska, M.Piotrowska; Build.Environ., **42**, 1843-1850 (2007).
- [9] J.H.Wang, H.P.Li, B.Qu, J.B.Zhang, T.Huang, F.F.Chen, Y.C.Liao; Int.J.Mol.Sci., **9**, 2495-2504 (2008).
- [10] A.W.Schaafsma, R.W.Nicol, M.E.Savard, R.C.Sinha, L.W.Reid, G.Rottinghaus; Mycopathologia., **142**, 107-113 (1998).
- [11] K.R.N.Reddy, H.K.Abbas, C.A.Abel, W.T.Shier, C.A.F.Olivera, C.R.Raghavender; Toxin Reviews., **28**, 154-168 (2009).
- [12] S.Tekle, S.Helge, B.Åsmund; Eur.J.Plant Pathol., **135**, 147-158 (2013).
- [13] D.W.Parry, P.Jenkinson, L.McLeod; Plant Pathol., **44**, 207-238 (1995).
- [14] C.M.Placinta, J.P.F. 'Mello, A.M.C.Macdonald; Ani.Feed Sci.Technol., **78**, 21-37(1999).
- [15] S.I.Phillips, P.W.Wareing, A.Dutta, S.Panigrahi, V.Medlock; Mycopathologia., **133**, 15-21 (1996).
- [16] L.F.Kubena, T.S.Edrington, R.B.Harvey, S.A.Buckley, T.D.Phillips, G.E.Rottinghaus, H.H.Caspers; Poult.Sci., **76**, 1239-1247 (1997).
- [17] C.E.Magnoli, M.A.Saenz, S.M.Chiacchiera, A.M.Dalcero; J.Nat.Prid., **61**, 367-369 (1999).
- [18] J.W.Bennett, M.Klich; Clin.Microbiol.Rev., **16**, 497-516 (2003).
- [19] A.Dalcero, C.Magnoli, S.Chiacchiera, G.Palacios, M.Reynoso; Mycopathologia., **137**, 179-184 (2004).
- [20] P.Shilpa, V.Koteswara Rao, S.Girisham, S.M.Reddy; Asiatic. J.Biotechnol.Resor, **2**, 392-402 (2011).
- [21] S.Girisham, G.V.Rao, S.M.Reddy; Natl.Acad.Sci.Lett.(India), **8**, 333-335 (1985).
- [22] I.Morales-Rodriguez, M.J.de H.V.Yanz-Morales, G.Silva-Rojas, Garcia-de-los-Santos, D.A.Guzman-de-Pena; Mycopathologia., **163**, 31-39 (2007).
- [23] R.Tamura, N.O.Yasuyuki Hashidoko, H.L.Suwido, T.Satoshi; Ecol.Res., **23**, 573-579 (2008).
- [24] I.Kiecana, E.Mielniczuk, Z.Kaczmarek, M.Kostecki, P.Golinski; Eur.J.Pl.Pathol., **108**, 245-251 (2002).
- [25] D.C.Hooker, A.W.Schaafsma, L.Tamburic-Ilincic; Plt.Disease., **86**, 611-619 (2002).
- [26] V.Rossi, L.Languasco, E.Pattori, S.Giosue; J.Pl.Pathol., **84**, 53-64 (2002).
- [27] K.Narasimha Rao, G.Shyam Prasad, S.Girisham, S.M.Reddy; Bioinfolet., **3**, 255-261 (2006).
- [28] R.M.Clear, S.K.Patrick, T.Nowicki, D.Gaba, M.Edney, J.C.Babb; Can.J.Pl.Sci., **77**, 161-166 (1997).
- [29] A.C.Sales, T.Yoshizawa; J.Food Prot., **68**, 120-125 (2005a).
- [30] A.C.Sales, T.Yoshizawa; Food Addit.Contam., **22**, 429-436 (2005b).
- [31] K.R.N.Reddy, C.S.Reddy, U.N.Mangala, K.Muralidharan; J.Mycol.Pl.Pathol., **36**, 271-277 (2006).
- [32] P.Rashmi; Asian.J.exp.Biol.Sci., **2**, 127-130 (2011).
- [33] V.Kumar, M.S.Basu, T.P.Rajendran; Crop.Prot., **27**, 891-905 (2008).
- [34] F.Duverger, S.Bailly, A.Querin, L.Pinson-Gadais, P.Guerre, J.D.Bailly; Revue Méd.Vét., **162**, 93-97 (2011).
- [35] C.Ezekiel, N.Adegboyega, C.Odebode Stephen, O.Fapohunda; J.Biol.Environ.Sci., **2**, 77-82 (2008).
- [36] J.F.Alberts, W.C.A.Gelderblom, R.Vleggaar, W.F.O.Marasas, J.P.Rheeder; Appl.Env. Microbiol., **59**, 2673-2677 (1993).
- [37] P.B.Schwarz, J.G.Schwarz, A.Zhou, L.K.Prom, B.J.Steffenson; Monatsschrift Fur Brauwissenschaft., **54**, 55-63 (2001).
- [38] E.Erpelding, L.Prom; J.Plant Pathol., **5**, 106-112 (2006).
- [39] M.Fakhrumisa, H.Hashmi, A.Ghaffar; Pak.J.Bot., **38**, 185-192 (2006).
- [40] E.W.Gachomo, E.W.Muttu, O.S.Kotchont; Int.J.Agric.Biol., **6**, 955-959 (2004).
- [41] P.S.Marley; Trop.Agric., **6**, 11-19 (2004).
- [42] S.B.Mathur, O.Kongsdal; Common laboratory seed health testing methods for detecting fungi, published by the ISTA, P.D.Box 308, Switzerland, (2003).