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STUDY OF SEED BORNE FUNGI OF DIFFERENT CULTIVATED PLANTS

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ABSTRACT

Toxic metabolites of storage fungi of 6cultivated seeds were study. These fungi were *Aspergillusflavus A.carbonarius, A. niger, Fusariumoxysporum, Cladosporiumcladosporioides, Penicilliumcorylophilum, Rhizopusoryzae* and *Pythiumindigoferae*. The storage fungiare cultivated in Glucose Nitrate liquid medium and the culture filtrates were estimation for its phytotoxic effects. The culture filtrateseffect inhibition of seed germination of cultivated seeds and its pumule and radicle length. On inoculation, it appearsnoxiousspots on leaf lamina and caused wilting of seedlings *in vitro.*

KEYWORDS: *Storage fungi, Toxic metabolites,phytotoxic effects,cultivated seeds,*

Introduction:

A tremendous number of plant-pathogens are known to produce injurious metabolites in synthetic media. The phytotoxic effects of various fungi have been studied by different workers. Jahani*et al*.,state that *Bipolarissorokiniana* causing spot blotch of wheat create toxin, which play role in pathogenicity. It is known that different degree of pathogenicity of a fungus results with differences in producing of phytotoxic metabolites. Samota and Singh observed that the culture filtrates of fungi caused reduction in seed germination, radicle and plumule length. Ahamad[1] observed that seed germination and seedling growth of mustard reduced significantly by the aflatoxin produced by *A. flavus.*

The present study, cultivated seeds of 6 plants *viz,Sorghum, Phaseolus vulgaris*, *Pisumsativum, Triticumaestivum, Oryza sativa*and *Pennisetumglaucum* were studied for storage fungi. The seeds of these plants were used for cultivated purpose. The storage fungi are known for reduction indecline and vital principles of the seeds. Because of this the market value of the seeds and its germination are affected. In the present study 8 storage fungi *viz,Cladosporiumcladosporioides, Fusariumoxysporum, Aspergilluscarbonarius, A. flavus, A. niger, Penicilliumcorylophilum, Pythiumindigoferae*and *Rhizopusoryzae* isolated from seeds of the 6cultivated plants are selected for the investigation of toxic metabolites.

Materials and methods

Isolation of fungal metabolites:

Glucose nitrate liquid medium were prepared and the pH was adjusted to 5.5. 25 ml of the medium was poured in 250 ml conical flasks, sterilized and inject separately with 1ml standard spore suspension. The flasks were incubated at 25 ±2 ^{0}C for 7 days and harvested by filtering their contents through Whatman filter paper No. 1. The culture filtrates so collected are used for *further* investigation.

Toxicity of fungal metabolites:

The toxicity of the culture filtrates of fungi on seed germination and seedlings was assessed by the method adopted by Papdiwal and Deshpande. The toxicity was tested by using three different methods as follows:

I) Effect on seed germination:

The harmful effect of culture filtrates on seed germination was studied by keeping 8 seeds on filter paper in a Petridis.

The filter paper was soaked in 5 ml of the above solution. Filter paper soaked in 5 ml tapwater served as control. The filter paper was hold moist by adding the cultures filtrate or water in the particular petridises. The plates were incubated for 72 hours and percentage growth was recorded.

II) Effect on seedlings:

Twenty days old seedlings were put down in the test tubes containing 20 ml of the culture filtrate and also in test tubes containing tap water as control. Observations were recorded after 24 hours.

III) Effect on leaf lamina:

The poisonousof culture filtrates of fungi was assayed by leaf necrosis method. Healthy leaves from 2 months old plant were freshly collected and employed for the assay. The leaves areclean with sterile distilled water and are placed on moist filter papers in petri dishes. With the help of sterilized needle, minor injuries are made on leaf lamina, away from the main veins. 0.01 ml of the culture filtrate was placed on the injured area. Injured leaves applied with sterile fresh medium served as control. The leaves were incubated at 25±2 ^{0}C and the growth of necrotic lesions was recorded after 24 hours.

Results:

Effect on seed germination

Seeds of 6cultivated plants were surface clean with 0.1 %$HgCl_2$ solution for four minutes and washed repeatedly with sterile distilled water.

Effect of the metabolites of storage fungi on seed germination was studied as per the method described earlier. Suitable controls are maintained with sterilized distilled water. The seed germination of 6 plants under study was noted, and communicates as percent germination. The length of radicleof each germinated seed was noted, and its mean was calculated. The results wheredifferentiate with control and percent inhibition of seed germination, and average radicle length are calculated. The results are presented in table 1 to 6.

Table: 1- Effect of culture filtrates on seed germination and length of radicle of *Phaseolus vulgaris*

Sr. No	Metabolites from the fungus	Seed germination		Average Radicle length	
		% germination	% inhibition	Length in cm	% inhibition
1	*Aspergilluscarbonarius*	16	65.00	1.7	55.00
2	*A. flavus*	11	70.00	1.8	48.60
3	*A. niger*	18	60.00	2.9	07.62
4	*Cladosporiumcladosporioides*	12	78.00	2.4	35.00
5	*Fusariumoxysporum*	20	60.00	1.8	56.38
6	*Penicilliumcorylophilum*	14	76.00	2.3	32.00

It is observed from table 1 that in case of *Phaseolus vulgaris*, the culture filtrates of 6 fungi decrease the percentage of seed germination. The highest inhibition of seed germination was observed in the culture filtrates of *A. flavus, C. cladosporioide* and *Penicilliumcorylophilum*(inhibition 75%).The adverse effect of culture filtrates was also observed on radicle length. The more inhibition of radicle length was observed in case of *Fusariumoxysporum*(inhibition 56.38%).

Table:-2 Effect of culture filtrates on seed germination and radicle length of *Sorghum.*

Sr. No.	Metabolites from the fungus	Seed germination		Average Radicle length	
		% germination	% inhibition	Length in cm	%inhibition
1	*Aspergilluscarbonarius*	41	52.00	1.9	57.00
2	*A. flavus*	32	64.60	2.6	42.00
3	*A. niger*	38	58.28	1.8	58.00
4	*Cladosporiumcladosporioides*	42	54.00	1.4	62.00
5	*Fusariumoxysporum*	54	38.40	2.4	48.00
6	*Penicilliumcorylophilum*	33	61.00	1.4	68.00

The data presented in table 2 reveal that all the 6 culture filtrates tested, showed inhibition of seed germination of *Sorghum.* The highest inhibition of seed germination (64.60 %) was observed in case of *A. flavus*and *P. corylophilum.* When the result of culture filtrate on the radicle length of the germinated seeds was observed, inhibition was observed in all cases. Maximum inhibition (68.00%) of radicle length was observed in case of *Penicilliumcorylophilum*culture filtrate.

Table:-3 Effect of culture filtrates on seed germination and radicle length of*Pisumsativum.*

Sr. No.	Metabolites from the fungus	Seed germination		Average Radicle length	
		% germination	% inhibition	Length in cm	% inhibition
1	*Aspergilluscarbonarius*	10	66.67	1.7	43.34
2	*A. flavus*	5	83.34	1.4	53.34
3	*A. niger*	10	66.67	1.6	46.67
4	*Cladosporiumcladosporioides*	5	83.34	1.4	53.34
5	*Fusariumoxysporum*	15	50.00	1.7	43.34
6	*Penicilliumcorylophilum*	5	83.34	1.8	40.00

The much moreinhibitions of seed germination were observed in case of *C. cladosporioides, A. flavus*and *P. corylophilum.* Inhibitory result of culture filtrates were also observed on radicle length. Large inhibitory effect on radicle length was with the culture filtrate of *C. cladosporioides*and *A. flavus*.

Table: - 4 Effect of culture filtrates on seed germination and radicle length of *Triticumaestivum.*

Sr. No.	Metabolites from the fungus	Seed germination		Average Radicle length	
		% germination	% inhibition	Length in cm	% inhibition
1	*Aspergilluscarbonarius*	50	37.50	1.5	57.15
2	*A. flavus*	40	50.00	2.5	28.58
3	*A. niger*	50	37.50	1.5	57.15
4	*Cladosporiumcladosporioides*	60	25.00	2.3	34.29
5	*Fusariumoxysporum*	60	25.00	2.0	48.86
6	*Penicilliumcorylophilum*	40	50.00	2.4	31.43

It is impact of Table 4 that the culture filtrates of the 6 fungi caused inhibition of seed germination and radicle length of *Triticumaestivum.* Largestinhibitions of seed germination

were observed in case of *P. corylophilum*and *A. flavus*. Maximum inhibition of radicle length was observed in case of *A. carbonarius*and *A. niger*.

Table: - 5 Effect of culture filtrates on seed germination and radicle length of *Oryzasativ.*

Sr. No.	Metabolites from the fungus	Seed germination		Average Radicle length	
		% germination	% inhibition	Length in cm	% inhibition
1	*Aspergilluscarbonarius*	30	50.00	0.6	40.00
2	*A. flavus*	20	66.67	0.2	80.00
3	*A. niger*	30	50.00	0.5	50.00
4	*Cladosporiumcladosporioides*	35	41.67	0.5	50.00
5	*Fusariumoxysporum*	30	50.00	0.5	50.00
6	*Penicilliumcorylophilum*	20	66.67	0.2	80.00

The more inhibition of seed germination of *Oryza sativa*was observed with the culture filtrates of *P. corylophilum*and *Aspergillusflavus*(Table 5). Largest inhibition of length of radicle was observed when the seeds were kept in the culture filtrates of *P. corylophilum*and *A. flavus.*

Table:-6 Effect of culture filtrates on seed germination and radicle length of *Pennisetumglaucum.*

Sr. No.	Metabolites from the fungus	Seed germination		Average Radicle length	
		% germination	% inhibition	Length in cm	% inhibition
1	*Aspergilluscarbonarius*	40	50.00	2.5	50.00
2	*A. flavus*	30	62.50	2.0	60.00
3	*A. niger*	50	37.50	2.8	44.00
4	*Cladosporiumcladosporioides*	55	31.25	3.0	40.00
5	*Fusariumoxysporum*	45	43.75	3.0	40.00
6	*Penicilliumcorylophilum*	30	62.50	2.0	60.00

From the (table 6) it is noted that, the culture filtrates of 6 storage fungi were inhibitory for seed germination, and radicle length of *Pennisetumglaucum.* The more inhibition of seed germination was noted with the culture filtrate of *Rhizopusoryzae.* Moreover, the same culture filtrate caused maximum inhibition of radicle growth.

The data presented in Table 1 to 6 reveal that the culture filtrate of *P. corylophilum*and *Aspergillusflavus* cause large inhibition, of seed germination, among the 8 storage fungi studied. The culture filtrates of the fungi were found to cause maximum inhibition of seed germination of 6 plants, among the 6cultivated plants investigated. Largest inhibition of seed germination was observed in case of *Pisumsativum* seeds (Table 3), that was caused by the culture filtrate of *P. corylophilum*and *A. flavus.*The culture filtrate of *A. flavus*also caused radicle length inhibition of 4 plants among the 6 medicinal plants studied. 70% radicle length inhibition of *Pisumsativum*has been found to be caused by *A. flavus*culture filtrate.

It is also observed that *Fusariumoxysporum*showed highest inhibition percentage of seed germination and radicle length in case of *Oryza sativa*and*Triticumaestivum.* Similarly *A. flavus*was found to exhibit maximum inhibition percentage of seed germination and radicle length in case of *Sorghum,Pisumsativum*and*Oryza sativa.Rhizopusoryzae*was found to inhibit the seed germination and radicle length in case of *Pisumsativum.*

1) Effect on seedlings:

40 days old seedlings of *Oryza sativa, Pisumsativum.Triticumaestivum* was taken and put in test tubes containing 20 ml of culture filtrate of 6 storage fungi. The 4 seedlings were also kept in tap water (20 ml) as control. The seedlings were incubated at $25^{o} \pm 2^{o}$ C. Observations were recorded after 24 hrs. When observations were taken after 24 hrs, it was found that all the seedlings remain in the culture filtrate of 6 storage fungi wilted. However, the seedlings kept in water were found health-giving. Therefore, it can be culminated that the culture filtrates of 6 storage fungi, isolated from seeds of 6cultivated plants were containing certain chemicals, that were toxic to the seedlings; and therefore, wilting of seedlings were observed in all the cases.

Discussion:

The culture filtrates of different fungi are known to produce inhibitory effect on seed germination and seedling growth. Definitely non enzymatic phytotoxic substances play an important role in these processes. Data presented in Tables 1 -6 reveal that the culture filtrates of 6 different seed borne fungi of 6 plants reduce their seed germination percentage significantly. In addition, they also reduced radicle length of the germinated seeds. The percentage inhibition of seed germination and radical length varied with different fungal species. In all the seed samples, more inhibition of seed germination was observed in the culture filtrate of *Aspergillusflavus* and *Penicilliumcorylophilum.* When the toxicity of the culture filtrates of 6 common seed borne fungi of 6cultivated plant species under investigation was assayed by leaf necrosis method on the leaves of these plants; it showed toxic effect.

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