

Seed-borne fungi of chilli in Madhya Pradesh and their significance

USHA BHALE, M. S. BHALE, B.R. PANDEY AND R. P. PANDEY

Department of Plant Pathology, JNKVV, Jabalpur 482 004, M.P.

Under central Indian conditions, 16 fungi were found associated with 36 seed samples of chillies. *Colletotrichum dematium* and *Alternaria alternata* associated with chilli seeds were found responsible for severe seed rot and seedling decay. Standard blotter method was better than Agar plate method as it permitted 18 fungi to detect as compared to 6.0 per cent. Virulence of both the fungi were tested by seed and soil infestation method. Artificially injured and water soaked seeds had greater seed infection, 69 and 63 per cent as compared to 49 and 45 per cent in uninjured seeds with *C. dematium* and *A. alternata* respectively. Pre- and post-emergence mortality due to the fungi were recorded. Test tube water agar seedling symptom test was better in the detection of pathogen in seedlings.

Key words : *Colletotrichum dematium*, *Alternaria alternata*, seed-borne fungi, chilli

INTRODUCTION

Chilli (*Capsicum annuum* L.) is a major spice used as condiment as well as vegetable. The crop suffers due to a number of diseases from seed rot, seedling decay to meladies of adult plant (Chupp and Sherf, 1960; Singh, 1987). Many of these are seed-borne (Nobel and Richardson, 1968; Suryanarayana and Bhombe, 1961; Padagnaur and Naik, 1991; Agrawal and Sinclair, 1996; Rout and Rath, 1972; Grover, 1970). In recent studies seed rot, seedling blight, twig blight and fruit rot incited by *Colletotrichum dematium* and *Alternaria alternata* have conclusively been identified as major problem, however, very few attempts (Harne and Nema, 1967; Verma, 1973) have been made on this under central Indian conditions. Hence, studies are undertaken to know the extent of mycoflora associated with seeds and their significance.

MATERIALS AND METHODS

Detection of seed-borne mycoflora

In all 36 seed samples of chilli were collected and tested by standard blotter and agar plate method (ISTA, 1993). In blotter method untreated and pretreated (surface sterilized) seeds with 0.1 per cent mercuric chloride for 30 sec. followed by 3 washing of sterile water were used while in agar

plate method only pretreated seeds were used. Seeds were examined directly under stereoscopic binocular after incubation at 28°C on 7th day.

Virulence test

Seed samples of varying degree of natural infection of *Colletotrichum dematium* and *Alternaria alternata* were sorted and tested by seed and soil infestation methods.

Seed infestation

Seed samples free from natural infection were rolled on 10 day old actively grown pure culture of *C. dematium* and *A. alternata* and plated on moist blotters as in standard blotter method. Prior to rolling, seeds were surface sterilized and soaked in sterile water for 60 minutes. Seed samples having no infection of these fungi served as control. In other set, seed coat of pretreated seeds of the same sample were injured slightly with the help of sterile needle. Seeds injured but without infestation were kept simultaneously for incubation.

Soil infestation

Inoculum of the test fungi were grown in Earlenmeyer conical flask on potato sucrose broth (PSB) for 15 days and mycelial mat were mixed in sterile soil and filled in separate plastic pots (10 cm dia.). Prior to mixing the mycelial mats of each fungi were thoroughly washed in sterile water to remove

the toxic metabolites. After 10 days of establishment in sterile soil, surface sterilized seeds with no infection were sown and observed for pre- and post-emergence infections.

Seed to plant transmission

Test tube water agar seedling symptom test (Khare *et al.*, 1977) was employed. Two hundred rimless test tubes of 30 ml capacity were filled with 10 ml of one per cent water-agar, plugged and autoclaved. One seed was placed in each test tube without any pretreatment and incubated in growth chamber.

Surface sterilized 40 cm pots were used. Sterile sand was filled and 10 seeds were sown in each and observations on disease development were taken.

RESULTS AND DISCUSSION

Detection of seed-borne mycoflora

Sixteen fungi were found associated with 36 chilli seed samples collected. Maximum (16) fungi were recorded when standard blotter method was used as compared to agar plate method (8 fungi). Highest incidence of *Colletotrichum dematium* (29.0 per cent) and *Alternaria alternata* (22.0 per cent) were recorded which resulted in severe seed rot and seedling decay (Table 1). Species of *Fusarium* and *Curvularia* were also pathogenic while fungi like *Aspergillus* sp., *A. niger*, *A. flavus*, *Botrytis cinerea*, *Penicillium* sp., *Rhizopus* sp. and *Memmoniella* sp. were found in higher percentage which indicate improper condition during harvesting, threshing and storage. The Standard Blotter Method was superior as compared to agar plate method as it permitted more number of fungi to express (Table 1). Similar trend of fungi was observed in other parts of India when tested with the chilli seeds (Padaganur and Naik, 1991; Rout and Rath, 1972; Harne and Nema, 1967;

Manoharachari and Padmavati, 1976).

Table 1 : Association of mycoflora with chilli seeds as detected by standard blotter and agar-plate method

Mycoflora	Standard Blotter Range	Agar Plate Range
<i>Alternaria</i> sp.	1.0 - 7.0	—
<i>Alternaria alternata</i>	3.0 - 22.0	1.0 - 12.0
<i>Aspergillus flavus</i>	1.0 - 12.0	1.0 - 3.0
<i>Aspergillus niger</i>	1.0 - 15.0	1.0 - 5.0
<i>Aspergillus</i> sp.	1.0 - 5.0	1.0 - 2.0
<i>Botrytis cinerea</i>	1.0 - 8.0	0 - 0
<i>Colletotrichum dematium</i>	3.0 - 29.0	1.0 - 18.0
<i>Chaetomium</i> sp.	1.0 - 6.0	—
<i>Curvularia lunata</i>	1.0 - 9.0	1.0 - 2.0
<i>Curvularia</i> sp.	1.0 - 3.0	—
<i>Drechslera</i> sp.	1.0 - 5.0	—
<i>Fusarium oxysporum</i>	2.0 - 17.0	1.0 - 3.0
<i>Fusarium solani</i>	1.0 - 9.0	—
<i>Memmoniella</i> sp.	1.0 - 4.0	—
<i>Penicillium</i> sp.	1.0 - 12.0	1.0 - 1.0
<i>Rhizopus</i> sp.	1.0 - 4.0	—

(—) No association was observed.

Virulence test

Pathogenicity of *C. dematium* and *A. alternata* was tested by seed and soil infestation method (Table 2). The fungi were responsible for 49.0 and 45.0 per cent seed rot while the injury resulted in 69.0 and 63.0 per cent infection due to *C. dematium* and *A. alternata* respectively. This also indicated that injury increased the chances of seed rot.

In case of soil infestation method, pre-emergence mortality, 41.0 and 29.0 per cent was recorded when the seeds were sown in soil infested with *C. dematium* and *A. alternata*. The seed-borne inoculum of these fungi also adversely affected the final stand (Table 2).

Table 2 : Influence of seed and soil-infestation with *C. dematium* and *A. alternata* on germination, pre and post-emergence losses in chillies

Fungus	Seed infestation				Soil infestation		Percent final stand
	Percent germination		Percent Infection		Percent mortality*		
	U	I	U	I	Per	Post	
<i>C. dematium</i>	65.0	62.0	49.0	69.0	41.0	7.0	52.0
Control	80.0	71.0	00.0	00.0	0.0	0.0	91.0
<i>A. alternata</i>	63.0	61.0	45.0	63.0	29.0	11.0	60.0
Control	83.0	73.0	0.0	0.0	7.0	0.0	93.0

U = Uninjured, I = Injured chilli seeds, * = After 15 days

Role of seed-borne *C. dematium* and *A. alternata* in causing seedling diseases was confirmed by test tube water agar seedling symptom test (Tables 3 and 4). Both the fungi resulted in severe (31.0 and 30.0 per cent) seed rot and seedling blight. Infected seedlings were pale

Table 3 : Role of seed-borne *C. dematium* in causing diseases on chilli seedlings in test tube water agar seedling symptom test and in pots.

Place	Percent Germination	Mortality of chilli seedlings (%)*			
		Pre-emergence		Post-emergence	
		Total	Due to <i>C. dematium</i>	Total	Due to <i>C. dematium</i>
Test tube with 1% water agar	47.00	53.0	31.0	15.0	8.0
Plastic pots with sterile sand	35.0	65.0	28.0	5.0	3.0

* After 20 days

Table 4 : Role of seed-borne *A. alternata* in causing diseases on chilli seedlings in test tube water agar seedling symptom test and in pots.

Place	Percent Germination	Mortality of chilli seedlings (%)*			
		Pre-emergence		Post-emergence	
		Total	Due to <i>A. alternata</i>	Total	Due to <i>A. alternata</i>
Test tube with 1% water agar	57.00	43.0	30.0	4.0	8.0
Plastic pots with sterile sand	42.0	58.0	28.0	9.0	9.0

* After 20 days

and died due to fungal attack. On isolation infected parts yielded the fungi indicating the movement from seed to seedling. Test tube method was better. The symptoms on roots could be seen through transparent water agar.

REFERENCES

- Agrawal, V.K. and Sinclair, J.B. (1996). *Principles of Seed Pathology*. CRC Press, Florida, USA.
- Chupp, C. and Sherf, A.F. (1960). *Vegetable diseases and their control*. Ronald Press Co. USA.
- Grover, R. K. (1974). Seed-borne diseases of chilli, their implications and control. 61st session of Indian Science Congress, Nagpur, 1974.
- Harne, S.M. and Nema, K.G. (1967). Seed borne fungi of some vegetables. *JNKVV Res. J.* **3** (2) : 130.
- ISTA (1993). International rules for seed testing. *Seed Sci. Technol.*, **21** (Supplement) p: 296.
- Khare, M.N., Mathur, S.B. and Neergaard, P. (1977). Test tube water agar seedling symptom test, a technique for detection of seed borne pathogen. *Seed Sci. Technol* **5** : 613-617.
- Manoharachary, C. and Padmavati, K. (1976). Fungi associated with chilli seeds. *Geobios* **3** (3) : 99-100.
- Nobel, M. and Richardson, M.J. (1968). *An annotated list of seed-borne diseases* (2nd ed.) Hand book on seed Health Testing **1** (1) : 191.
- Padagnaur, G.M. and Naik, K.S. (1991). Mycoflora of chilli seeds from fruit rot affected and healthy seeds. *Curr. Res* **20** (2) : 183-184.
- Rout, B.K. and Rath, G.C. (1972). Note on seed-borne diseases of chilli (*Capsicum annum* L.). *Indian Phytopath* **25** (4) : 597-598.
- Singh, R.S. (1987). *Diseases of Vegetable Crops*. Oxford and IBH Co., New Delhi P. 362.
- Suryanarayan, S. and Bohmbe, B.B. (1961). Studies on the fungal flora of some vegetable seeds. *Indian Phytopath* **14** : 30-41.
- Verma, M.L. (1973). Comparative studies on virulence of isolates of 4 species of *Colletorichum* parasitic on chillies. *Indian Phytopath* **26** (1) : 28-31.

(Accepted for publication April 6 2000)