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CONTAMINATION, INFECTION AND TRANSMISSION OF *ALTERNARIA ALTERNATA* IN CUMIN.

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Out of 149 seed samples collected from 16 districts of Rajasthan, 85 samples had 0.25 to 26.5% black discoloured seeds which on incubation yielded 0.5 to 100% incidence of *Alternaria alternata*. Both symptomatic and asymptomatic seeds carried seed-borne infection. Conidia of pathogen were present externally on seed and its mycelium, inter as well as intracellularly in pericarp, carpophore and endosperm of seeds. Intraembryal mycelium was not seen.

From seeds infection reached to seedling/plant. Seed infection caused loss in their germination and later necrosis of leaves and stem.

Key Words: Cumin, seed-borne, infection, transmission *Alternaria alternata*.

Cumin (*Cuminium Cyminum* L.) seeds carry large number of seed-borne fungi, important being *Alternaria burnsii, Fusarium oxysporum* f. sp. *cumini*, F. spp.(Neergaard, 1977; Richardson, 1990). *A. alternata* (Fr) Keissler, a common seed- borne pathogen of many crops (Richardson, 1990) has been reported on cumin seeds from Kanpur (Swarup and Matur, 1972). The present paper deals with its incidence in cumin seeds collected from Rajasthan, alongwith seed infection and disease transmission.

MATERIALS AND METHODS

One hundred forty nine seed samples of cumin, collected from 16 districts of Rajasthan were tested by dry seed examination and incubation methods (Anonymous, 1976) and 78 samples were screened by seed washing test, for spore load of *A. alternata*.

Seeds were categorised as (i) bold symptomless (ii) bold black and (iii) shrivelled black discoloured ones and histopathological studies were made following techniques used by Singh *et al.* (1977).

The disease transmission studies were conducted using naturally infected seeds, by standard blotter method and growing on test in pots (Anonymous, 1985). Percent seed germination, seeds rot, seedling mortality and disease symptoms were recorded.

RESULTS

Black discoloured symptomatic seeds were observed in 57% seed samples, with 0.25 to 26.5% incidence. Seed washing test revealed spores of *A. alternata* in 70 samples of 14 districts with a spore load of 1000 to 24,000spores/g seed. The pathogen was isolated in 125 samples,with 0.5 to 100% incidence in incubation tests. Its occurrence was high in samples from Ajmer, Bhilwara, Jaipur, Jodhpur and Kota districts showing its wide occurrence on cumin seeds of Rajasthan.

A. alternata was isolated from pericarp (28-100%) of seeds of all the three categories in component plating. The endosperm of bold black and shrivelled black seeds (Fig. A) carried 2 to 8% and 46 to 54% infection respectively. The carpophore region carried high incidence (28-100%) of the pathogen. Cleared wholemount preparations of pericarp (Fig. B), endosperm (Fig. C) and carpophore revealed thick, dark brown, knotty, branched septate mycelium.

Microtome sections of bold symptomless seeds showed well differentiated pericarp consisting of 3 layers (exo-, meso - and endocarp) with fused seed coat, kidney shaped endosperm and tiny embryo. In such seeds only inter-and intracellular mycelium was seen in pericarp and carpophore, But in bold black category 30-40% seeds showed infection in all the 3 layers of pericarp (Fig.D) and cells of carpophore. Epithelial cells surrounding the vitae cavity revealed intercellular mycelium.

In shrivelled black seeds besides mycelial fragments, conidia were seen lodged in secondary ridge region of pericarp. Mycelial infection was observed in pericarp (100%). Cells of pericarp revealed necrosis,

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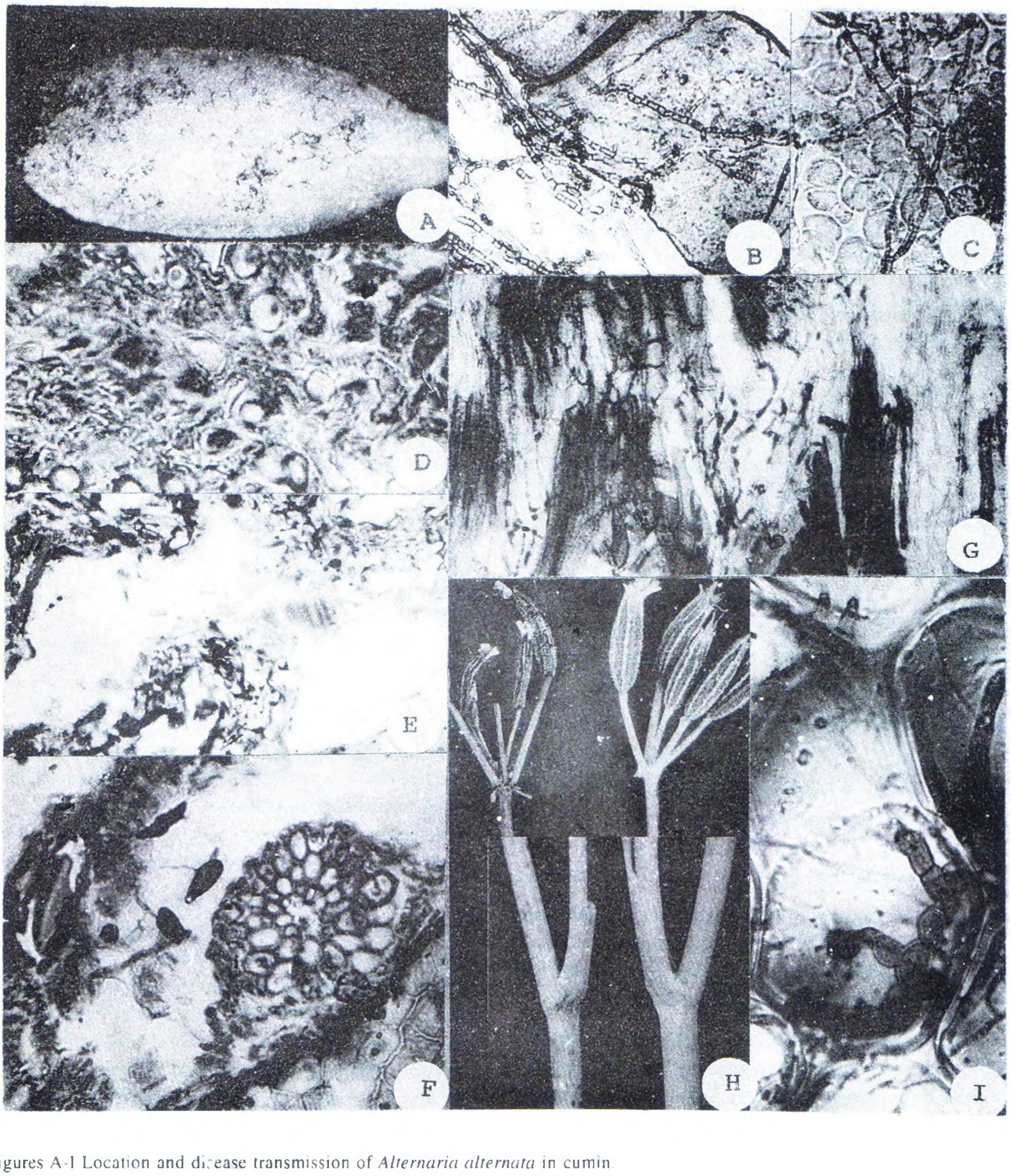

Figures A-I Location and disease transmission of *Alternaria alternata* in cumin.
Fig. A Incubated endosperm showing conidialchains of *A. alternata*. x 17.5.
Fig B,C Cleared pericarp and endosperm respectively showing branched, septate mycelium. x 125, x 250.
Fig. D Inter-and intracellular mycelium in exo-, mesoand endocarp region of pericarp. x 250.
Fig. E Necrosis and disintegration of pericarp tissue (due to heavy infection). x 125.
Fig. F,G Conidia (F) and mycelial aggregation (G) in carpophore region of shrivelled black seed. x 250.
Fig H Infected inflorescence (above) showing shrivelled black seeds and stem (below) with necrotic spots compared with healthy plant parts (right) x 12.8.
Fig. I T.S. of infected root showing inter-and intracellular mycelium in pith regionx 250.

Table 1: Per cent isolation of Alternaria alternata in blotter test from different parts of infected (IS) and healthy looking seedlings (HLS) and plants of cumin of different ages obtained from asymptomatic and symptomatic seeds in pot experiment.

Days from Emergence	30				70		161	
	Asymp.		Symp.		Asymp.	Symp.	Asymp.	Symp.
Seedling/Plant Parts	IS	HLS	IS	HLS				
Alternaria alternata								
Ac. No. 8079								
Root	6	-	78	14	8	42	12	48
Stem	8	-	84	22	24	82	26	72
Leaf	28	4	92	26	32	94	28	100
Flower	-	-	-	-	10	46	-	-
Fruit	-	-	-	-	-	-	12	54
Ac.No. 8080								
Root	4	-	72	10	14	38	8	54
Stem	12	2	86	24	22	74	24	76
Leaf	32	10	90	28	34	96	36	96
Flower	-	-	-	-	14	42	-	-
Fruit	-	-	-	-	-	-	8	62

Asymp-Asymptomatic. Symp-Symptomatic

dense mycelial aggregation, disintegration of tissues leading to the formation of large cavities (Fig. E). The fibrovascular bundles showed hyphae around the vessels (Fig. G) and conidia loged in carpophore region (Fig. F). However the cells of endosperm were normal. Mycelium was not seen in embryo.

DISEASE TRANSMISSION- In blotter test, seeds germination began after 24 hrs of sowing and was 86%, 97% and 49, 84% in asymptomatic and symptomatic seeds respectively. Ungerminated seeds were covered with fungal growth and rotted. Symptoms appeared on 4th day as yellowing, wrinkling of leaves, brown necrotic spots and streaks and rotting of hypocotyl.

Test tube seedling symptom test exhibited 88, 90%; 78, 82 and 52, 58% germination in asymptomatic, bold black and shrivelled black seeds of the two seeds samples. Symptoms appeared after 19 days of sowing as yellowing of lower leaves, followed by upper leaves, necrotic spots and streaks on stem (Fig. H) and leaves. Infected plants were stunted,possessed less number of inflorescence bearing bold black and shrivelled black seeds (Fig. H) resulting in high yield loss. Stand of healthy seedlings/plant was 78, 82%, 40, 44%and 0% and total loss was 10, 14% 30, 34% and 92, 94% in symptomless, bold black and shrivelled black categories respectively. Pathogen was isolated from 30 to 161 days old seedling/plants (Table 1.). Leaves followed by stem, inflorescence and root revealed high incidence of pathogen. Cleared tissues and hand cut sections showed inter-and intracellular mycelium in all plant parts (Fig. I) which confirms transmission of *A. alternaria* from seeds to seedling/plant.

A. alternata, wide spread in Rajasthan, has been reported as seed-borne pathogen of many crops, such as sunflower (Singh *et al.*, 1977), sorghum (Rastogi, 1984), wheat (Agarwal *et al.*, 1987), intra embryal infection. However, Thakkar (1988) recorded its extraembryal infection in barley seeds. In present study the pathogen occurred as contaminant as well as pathogen of seeds. Presence of inter-and intracellular mycelium suggests its internally seed-borne nature. Extent of seed-borne colonization varied with the degree of seeds discolouration. Dense mycelial aggregation in pericarp and carpophore indicates entry of pathogen through pedicle, from where it spreads in the tissues of pericarp and carpophore, penetrating further in the endosperm. The multilayered endospermic tissue acts as mechanical barrier to penetration into the embryo. Thus embryo of all categories are free from infection. Occurrence of symptoms (necrosis) and recovery of pathogen from all plant parts suggests seeds to seedling/plant transmission of *A. alternata* in cumin.

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