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RESEARCH ARTICLE

Occurrence of *Alternaria brassicae* in seed-producing cauliflower and its role of seed infection

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SUMMARY

The dark spot pathogen of cauliflower, *Alternaria brassicae*, is a common problem in east and north-east Bihar. Occurrence of this disease is repeatedly observed in crucifers of this region. The disease is more severe in those areas where seed production of crucifers is practiced. Therefore, we designed this experiment including the natural occurrence of dark spot disease and the survivability of the pathogen in the seed. In our location, the disease is started to appear in January. However, the peak period of this disease on foliage was middle February to middle March. This experiment also envisaged that *A. brassicae* can actively survive for a period of two months on the seed of cauliflower. The seeds obtained from late matured pods had significantly higher seed infection ($P < 0.05$) compared to seeds from early mature pods. This paper renders the evidence for development of new initiative to manage this disease such as development of physical or chemical seed treatment method that sustains effectively for two months and development of variety with early and synchronous podding, and other programmes may be encouraged. Management strategies of this pathosystem are briefly discussed.

Key Words : *Alternaria brassicae*, Cauliflower, Dark spot, Seed infection

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Different species of *Alternaria* infect plants like radish, cabbage, broccoli and cauliflower. *Alternaria brassicae* (Berk.) Sacc., causes dark spot disease of cauliflower (*Brassica oleracea* var. botrytis) that damages the production of cauliflower in both early and late season. In India, this pathogen also infects mustard, radish and cabbage in addition to cauliflower, and responsible for severe infection resulting substantial yield loss inciting premature ripening, siliqua dehiscence and seed shriveling (Seidle *et al.*, 1995). Yield losses of 20 to 30% due to this disease were recorded worldwide (McDonald, 1959; Conn *et al.*, 1990). *A. brassicae* causes dark spot of cultivated and wild crucifers (Smith *et al.*, 1988).

Cauliflower is widely grown as major vegetable in the north eastern region of Bihar and severely affected by *A. brassicae*. In this area, seed production of

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cauliflower is often practiced by the growers. The severity of this disease is sporadic to epidemic in this part of Bihar. Dark spot disease is not restricted to leaves but also damage stalk and pods, which turn black when colonised by *A. brassicae* leading to pre-mature ripening. Pre-mature ripening of the pods may lead to shedding of seeds (Maude and Humpherson-Jones, 1980). Infection of *A. brasicae* was recently reported on seedlings, leaves, seeds and pods (Kohl *et al.*, 2010). Mycelium of the fungus is observed over the seed superficially but it also progresses through the internal tissues of the seed. Seeds of infected pods tend to be shrunken and have low viability. The matured pods exhibit spots with circular markings. Saprophytically surviving mycelium on crop debris is foremost inoculum source for the disease.

This paper discusses about degree and severity of disease, and the viability of *A. brasicae* on seed of cauliflower. The current information would be useful of developing remedial strategy of seed production in cauliflower against dark spot disease.

MATERIAL AND METHODS

Disease assessment in the field :

The disease was observed once a week and assessments continued until maturity to find out intensity pattern in leaf and pod stage. Disease scoring of the leaves and pods was done in 10 randomly tagged plants using a 0-5 scale, which is recognized as: 0 = no infection, 1 = 1-5% infected area, 2 = 6-10% infected area, 3 = 11-20% infected area, 4 = 21-30% infected area, 5 = 31-100% infected area covered on the leaf. Further, infection of seeds and survival of the pathogen was examined under microscopic field. Seeds collected form infected plants were placed in a two-layer moist blotter paper adjusted in the Petri plates. The seed incubated for seven days allowed for germination and mycelial expansion of the fungal growth. Examination under a microscope at 40× magnification was done for presence and absence of the pathogen in the seed. Moreover, germinated seedlings (cotyledons and radicals) were also observed for the same objective.

Seed infection test :

To test whether the seeds were contaminated with the propagule and assisted for disease development in next cropping season, the investigation on viability of the pathogen was observed by above given procedure at regular intervals (from 15 to 90 days). For viability of seed, varying grade of infected pods (0-5 scale used for rating) was selected and tagged. Seeds were collected in two lots *i.e.* early matured pods (EMP) and late matured pods (LMP). In each pod, seeds were carefully threshed ensuring no injury made on seed. Thereafter, hundred seeds placed on two-layer moist blotter papers adjusted inside Petri plates and allowed for germination. The seed viability was then counted. Each seed viability experiment repeated thrice following Complete Randomized Design.

RESULTS AND DISCUSSION

The results obtained from the present investigation as well as relevant discussion have been summarized under following heads :

Disease development under field condition :

Our assessments showed the critical period for disease development on foliage was between second fortnight of February and second fortnight of March; however, disease appeared in the first week of January. During this period, dark spot epidemics appeared with high severity in cauliflower seed production plots. At first, the disease was observed as small grayish brown necrotic spots on leaves, which later coalesce and expressed as leaf blight. There was a rapid and abrupt progress of disease on the stalks and lastly black spot symptom resembled on the pods. Significant difference between leaf severity and pod severity of the disease was observed starting 3rd week after disease initiation (Fig. 1). During the study, comparative severity between leaf and pod was assessed; maximum severity on leaves (30.4%) was recorded at 5th week after disease onset (middle March) on foliage. Nevertheless, pod severity found significantly lower than the leaf severity for all the cases of observation.

This investigation advocates that application of management options like fungicide spray should be addressed during January or when the disease onset is taken place as described in Fig. 1. In those areas where the disease appears late, the strategy of fungicidal spray should be implemented as prophylactic management starting February. For this disease, a moderately effective fungicide should be used because the cruciferous plants are less susceptible at the younger stage (Humpherson-Jones, 1992). If infection on the foliage is neglected, the application of such fungicide should be addressed at early pod formation stage at regular interval. This strategy

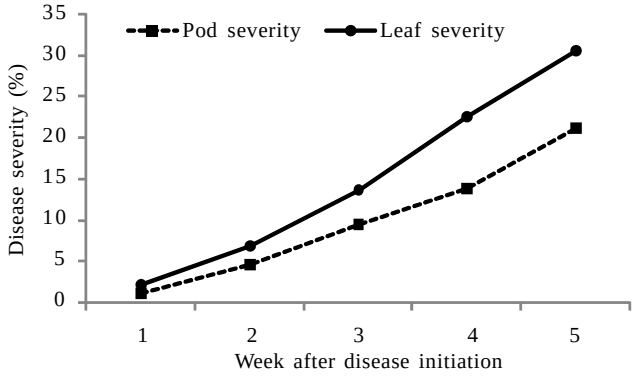

Fig. 1 : Severity of disease at weekly interval on leaves and pods

would reduce the pod infection; substantially it would also limit the pathogen survivability in the pod that leads to reduced seed germination in the successive season.

Disease dynamics in seed :

In the seed infection study, incidence of infection in germinating seed and non-germinating seed was counted over period of time immediately after threshing (Fig. 2). A geminated seed was considered infected if blackish dot found on the germinating cotyledon. Survival of the fungus in the germinating seed was found to be diminishing with progress of time. Nearly 50% reduction in the pathogen survival was observed at 15 days after threshing. No infected cotyledon (germinated seed) was observed on 60 days after threshing, which suggest that the seed-borne infection in the next season can be minimized by the application of a treatment effective for two-month period. This attempt will not only save the crop loss in successive season but also reduce inoculum pressure in the field by excluding pathogen from the seed. The non-germinated seed with the cottony fungal growth over the seed surface was considered as infected seed. Some seeds (15%) were severely infected with *A. brassicae* that did not allow the seed to germinate (Fig. 2). Three times reduction in complete inhibition of seed germination was recorded on 30 days after threshing. However, 1% non-germinated seed due to fungal infection was observed on 60 days after threshing indicated the prolong survival ability of the fungus. No effect on seed germination due to fungal infection was detected on 75 days after threshing.

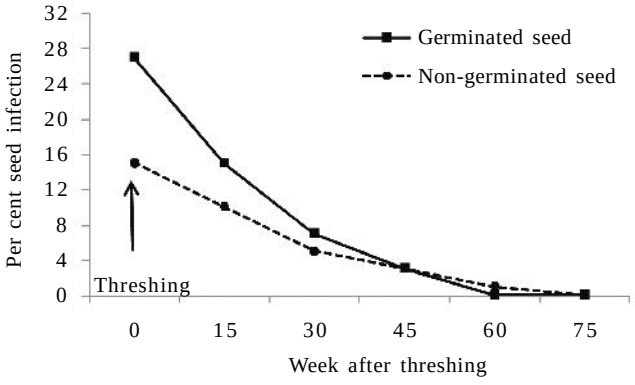

Fig. 2 : Survival of *Alternaria* on seed immediately after threshing

The result indicated that the pathogen was active upto two months after harvest in the pods (Fig. 2). In the bulk seed storage, the facility of physical or mechanical methods should be enriched in order to restrict the fungal infection in the pods. Such methods should be effective at a minimum period of two months. Moreover, the chemicals may also be identified that can inhibit the pathogenic colonization in the pods. However, the cheaper and practically feasible method should be popularising with a general understanding of environmental damage in the centre of the policy.

Evaluation of seed infection and viability of the pathogen :

Due to severe infection in pods germination ability of seed found reduced (Table 1). Seeds obtained from late matured pods (LMP) were more vulnerable to reduced germination than early matured pods (EMP). In LMP, severe infection of *Alternaria* was noticed due to presence of high inoculum, which was confirmed under a microscope (40×). Therefore, reduced seed viability was detected in the seeds obtained from LMP colonized with *A. brassicae*. Overall, the higher ratings associated with high per cent disease covered area (DCA) on the pods are significantly correlated reduced seed

Table 1 : Seed germination of cauliflower seed infected by *Alternaria brassicae* with varying degree of disease covered area

Rating	Per cent DCA[x]	Seed germination %	
		EMP[y]	LMP[z]
0	0	78.3 a	71.6 a
1	1-5	72.3 ab	67.0 a
2	6-10	65.3 b	53.0 b
3	11-20	50.3 c	42.0 c
4	21-30	41.6 c	28.3 d
5	30-100	23.6 d	10.6 e
	C.D. (P=0.05)	8.7	6.6

[x] Disease covered area
[y] Early matured pod
[z] Late matured pod

germination (Table 1). Maximum seed germination for both of the sources (pods) of seeds was noticed on the lowest rating. Although, 21.7 and 28.4% reduction in seed germination in EMP and LMP, respectively was observed at the 0 per cent DCA. At maximum rating, the seeds of LMP showed more than 50% reduction in seed germination compared to what recorded for the seeds of EMP. In case of the EMP seeds, no significant difference of seed germination was recognized among rating 3 and 4 ($P > 0.05$); however, the similar observation was made on rating 0 and 1 for the seeds of LMP.

Seeds obtained from the late maturity pods showed reduction in seed germination (Table 1). Humpherson-Jones (1992) reported that necrotrophic pathogens such as *A. brassicicola* and *A. brassicae* easily infect mature plant tissue of cruciferous crops than the younger one. This work vividly urges for development of cauliflower variety with early maturity character. Additionally, such varieties should have the synchronous maturity trait because higher seed infection was detected in the seeds obtained from pods that matured late. Because the resistance is decreased with plant age of crucifers (Humpherson-Jones, 1992), the young pods have high degree of resistance, and hence, a moderately effective fungicide would be a logical option that potentially inhibits the pathogen infection in the pod. Foundation of this strategy would prevent the early loss due to infection coupled with encouraging the safety measures of environmental hazard leading to development of an organic system. The use of moderately effective fungicide would accompany for prohibition of resistant strains of the pathogen. Resistant varieties are cheaper and reliable option for plant disease management (Hammond-Kosack and Jones, 1997). Therefore, the lines of cauliflower should be identified from the multi-location trials where high inoculum pressure of this pathogen is prevailed.

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