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STUDIES ON THE SURVIVAL OF *Alternaria brassicae* THE CAUSAL ORGANISM OF LEAF SPOT OF RAPESEED AND MUSTARD

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Alternaria brassicae lesions on siliqua was found directly associated with the seed infection. Intensity of seed infection varied with the number of lesions on a siliqua. Association of *A. brassicae* has been demonstrated with seeds of three crops of rape seed and mustard. However the population of the colonies of *A. brassicae* diminished with increased storage temperature and period. Temperature played a key role in survival of *A. brassicae* than storage material and period. *A. brassicae* survived with plant debris buried in field below the depth of 7.5 cm. *Chenopodium alba* has been recorded as a collateral host of *A. brassicae* in field.

Alternaria brassicae causes round light brown to dark brown spots with concentric rings on the leaves and stem, long to oblong and round sunken lesions on twigs and siliqua of all the *Brassica* species. This disease is responsible for quantitative and qualitative losses (Degenhardt *et al.* 1974, Ann' 1979). The lesions on the siliqua is directly responsible for shrivelling and dis-colouration of seeds and such seeds are responsible for carry-over of the inoculum (Richardson, 1970). Survival of *Alternaria brassicae* with seed, plant debris and in soil have been suspected (Vasudeva, 1958; Singh, 1978). Tsunoda and Skoropod (1977) have observed that *A. brassicae* survive in the form of microsclerotia and chlamydospores which appeared in later stage on infected leaves. In the present investigations, attempts have been made to find out survival source of *A. brassicae* in nature.

MATERIAL AND METHODS

Seed study: Infected siliqua of *Bras-*

sica juncea (Parkash) *Brassica campestris* (Yellow sarson, YSPB 24 and Brown sarson, BSH-1) were collected from field and graded in 0 to 5 scale, based upon 0 = no black spot, while 5 = heavily infected with *Alternaria brassicae*. Seeds collected from these siliqua were tested for the presence or absence of the organism by 'moist filter paper' method in 10 cm. petri-dishes. Organisms grown on the seed were examined and colonies of *A. brassicae* developed on those seeds were taken into account.

In another experiment, seeds samples of *B. juncea* and *B. campestris* were collected from threshing floor from crop infected with *A. brassicae* and kept in cloth bags on laboratory benches (Temperature ranged from 35-45°C). These seeds were examined for *A. brassicae* by the same method after an interval of one month.

In third experiment, seed samples were taken in cloth, polythene and

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paper bags and kept on laboratory benches (35-45° C) and in refrigerator (temp. ranged from 8 - 10° C). Seeds from these bags were examined periodically, for seven months (April to October).

Diseased plant debris:- Infected leaves, pods and husk were collected and kept on laboratory benches in paper bags for examination of conidial germination.

In another experiment, above materials were kept in sterilized soil in 25 cm earthen pots at the depth of 0.0, 7.5, 15 and 22.5 cm in 4 replicated sets. The pods were kept in green house under natural environment. In other set plant debris were kept in field soil (unsterilized) in half charcoal drums at the depth of 0.0, 7.5, 15.0, 22.5 and 30 cm in the replicated sets. These drums were buried in field to provide field conditions. In both the cases 10 and 50 seeds of *B. juncea* were sown in each pot and drum respectively. Seedlings received irrigation and interculture regularly. These plants were observed periodically for the appearance of leaf spot caused by *A. brassicae*.

Collateral host: Out of several plant species other than *Brassica* sp. which grow as weed, were kept in a plot of 4 x 2.5 m size and culture suspension of *A. brassicae* was sprayed on these weed plants thrice at the interval of 7 days. Out of several weeds *Chenopodium alba* responded. In another experiment leaves of *Chenopodium alba* were collected from field and Koch

postulate was proved. Cross inoculation was also made. Development of lesions under laboratory conditions by detached leaf technique was studied.

RESULTS AND DISCUSSION

Seed infection Study: It was noted that pods having up to 5 lesions with one or two bigger ones showed 2-5% (average being 5%) seed infection; Pods having 5 big and 3-4 small lesions were having 10-12% infected seeds. On those pods, where big lesions were more than 5 and less than 10 and with few small lesions were found to be infected by 20 - 25% (average being 22.5%). When more than 10 big lesions were found, seed showed 30-40% infection (average being 35%) (Table-1). In this grade, other seeds looking apparently diseased i. e. shrivelled and discoloured but all such seeds did not show infection. This may be due to the after effect of infection. Seeds from bulk infected population of *B. juncea*, *B. campestris* (Y S P B - 24) and *B. campestris* (BSH-1), were tasted by the same method immediately after harvest and was noted that out of 1360, 1220 and 1200 seeds 17.0, 32.7 and 41.3 per cent were found infected by *A. brassicae* respectively. A similar observation was made by Chahal and Labana (1980) while working on the association of *A. brassicae* with the seeds of Indian mustard under Punjab conditions

In another experiment seeds of *B. juncea* and *campestris* were collected from threshing floor and kept in cloth bags on laboratory benches. These seeds were examined after an interval

of one month. It was noted that *A. brassicae* infection was 11.8 and 15.5 percent in *B. juncea* and *B. campestris* respectively in the month of April but this population reduced to 1.7 and 1.9 percent respectively in May. Further examination of seeds revealed that in the month of June, population of *A. brassicae* disappeared. This indicates that high temperature during month proved lethal to this organism.

On further investigation to find out the presence of *A. brassicae* colony in relation to storage conditions and storage temperature of the seeds of above two crops were stored in polythene, paper and cloth bags and kept at $8\pm 2^{\circ}\text{C}$ (Refrigerator temp) and $40\pm 5^{\circ}\text{C}$ (Laboratory conditions) in 1979--80, it was noted that at low temperature, the *A. brassicae* remained with seeds upto October. However, population was reduced drastically from 10 per cent in April to 1.5 per cent in October when stored in cloth bag. In case of polythene bags, it came down to 0.5 per cent but in paper bags stored seeds, not a single colony of *A. brassicae* was recovered in October (Table 2) in case of *B. juncea*. Similarly, in case of *B. campestris* (yellow sarson), the population of infected seeds remained only 1.2 per cent in October from 16 per cent of April in case of cloth bags, while 1.5 and 0.8 per cent when stored in polythene and paper bags respectively (Table 2). On the other hand, under laboratory conditions, the colonies count reduced drastically from 10 per cent to 5.4 per cent and from

16.0 per cent to 0.40 per cent where seeds of *B. juncea* and *B. campestris* (yellow sarson) were stored in cloth bag respectively after a month (May test). This population reduced further when stored in polythene and papers bags and kept under laboratory conditions irrespective of crop. Tests of June and further months did not yield any *A. brassicae* colony on seeds. From the above experiments, it was concluded that storage materials have no effect on carry over of *A. brassicae* from seed to new crop and it is only temperature which played major role in reducing the population.

Diseased plant debris: It was noted that no lesion developed up to 30 days in case where material were kept in sterilized soil under green house conditions and after 45 days lesions started appearing irrespective of burrial depth and even in control sets where plant debris were not kept. This showed that spores came from nearby fields. In another trial, where field in which no *Brassicae* crop was grown for the last 3 years, were kept in iron drums. *Alternaria* leaf spot started appearing after 20 days. Number of lesions on the leaves were more when plant debris were burried at the depth of 7.5 cm and less, when it was burried at the depth of 15 cm. No lesion was observed where material was burried at the depth of 22.5 cm in drums.

Above observation indicated that *A. brassicae* did not survive in earthen pots having sterilized soil kept in green house. It may be due to shift in the microclimate under sterilized soil in

green house. on the other hand, it survived when plant debris were kept in field soil at the depth of 7.5 or below. In case where plant debris kept on surface, it was liable to exposure to sun and due to high temperature of summer the organism was destroyed as in case of laboratory observations.

Collateral hosts: There are reports that *A. brassicae* survives and flourish well on several Brassicae species (singh, 1979). However, during these investigation, authors have come across that *A. brassicae* produced redish, circular with concentric rings on (*Chenopodium alba* L.) which comes up as a weed along with rape seed and mustard crop. After proving koch's postulates, cross inoculation i.e. from *B. juncea*, to *Chenopodium alba* and from *Chenopodium alba* to *B. juncea*, revealed that *Chenopodium alba* isolate of *A. brassicae* produced lightbrown circular lesion on *B. juncea* by detached leaf technique after 7 days and *B. juncea* isolate also caused light-brown lesions on detached *Chenopodium alba* leaves. These lesions did not develop into normal lesions because the detached leaves of both the plants became yellow.

This observation indicated that *Chenopodium alba* a common weed of rapeseed and mustard helps in

dissemination of *A. brassicae* under favourable conditions. Therefore, removal of this weed will be an additional tool to check the intensity in nature.

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Table 1 Presence of *Alternaria brassicae* in infected seeds of *Brassica juncea* in relation to siliqua infection.

Grade	No. of lesions on one siliqua	Conditions of seeds	Per cent range of infection	Per cent average
1.	No lesion/one or two small pin head size	Bold normal colour	0	0
2.	One or two big and 2-3 small lesions	Bold with few shrivelled and discoloured	2-5	5
3.	5 big and 3-4 small	Bold with few shrivelled discoloured	10-12	10
4.	5-10 big lesions	Majority shrivelled and discoloured	20-25	22.5
5.	More than 10 big lesion 10-15 small lesion.	Few normal seeds and most are shrivelled and diseased.	30-40	35

Table-2 Effect of storage temperature and storage material on the survival of *Alternaria brassicae*

Crops	Temperature (°C)	Percent <i>Alternaria</i> infected seed detected during the month											
		April			May			June			July		
		1	2	3	1	2	3	1	2	3	1	2	3
<i>B. juncea</i>	8+0°C	10.0	10.0	10.0	8.4	8.0	7.5	3.6	3.0	4.8	3.0	2.0	1.1
	40+5°C	10.0	10.0	10.0	3.3	2.5	5.4	0.8	0.0	1.9	0.0	0.1	0.0
<i>B. campestris</i> (YSPB-24)	8+2°C	16.0	16.0	16.0	8.2	8.2	10.0	7.0	7.5	4.4	6.0	2.1	3.4
	40+2°C	16.0	16.0	16.0	4.0	1.1	4.0	0.3	0.0	0.0	0.0	0.0	1.1

Crops	Temperature (°C)	Percent <i>Alternaria</i> infected seed detected during the month								
		August			September			October		
		1	2	3	1	2	3	1	2	3
<i>B. juncea</i>	8+0°C	1.0	1.0	1.6	2.4	0.0	1.8	0.3	0.0	1.5
	40+5°C	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
<i>B. campestris</i> (YSPB-24)	8+2°C	2.5	2.6	3.1	4.0	3.5	2.9	1.5	0.8	1.2
	40+2°C	0.0	0.0	0.0	0.1	0.0	1.0	0.0	0.0	0.4

Note 1: Seeds stored in polythene bag. 2 = stored in paper bag, 3 = stored in cloth bag