The Pharma Innovation Journal 2022; SP-11(7): 4466-4470

www.ThePharmaJournal.com

The Pharma Innovation

ISSN (E): 2277-7695
ISSN (P): 2349-8242
NAAS Rating: 5.23
TPI 2022; SP-11(7): 4466-4470

www.thepharmajournal.com
Received: 21-04-2022
Accepted: 24-05-2022

Sanjeet Kumar
Ph.D. Scholar, Department of Plant Pathology, TCA, Dr. Rajendra Prasad Central Agricultural University Pusa, Bihar, India

AK Mishra
Assistant Professor, Department of Plant Pathology, TCA, Dr. Rajendra Prasad Central Agricultural University Pusa, Bihar, India

CS Choudhary
Assistant Professor, Department of Plant Pathology, TCA, Dr. Rajendra Prasad Central Agricultural University Pusa, Bihar, India

Corresponding Author
Sanjeet Kumar
Ph.D. Scholar, Department of Plant Pathology, TCA, Dr. Rajendra Prasad Central Agricultural University Pusa, Bihar, India

Studies of variability in *A. brassicae* isolates causing blight disease of Mustard in Bihar

Sanjeet Kumar, AK Mishra and CS Choudhary

Abstract
The variability study of fifteen isolates of *A. brassicae* each collected from different district locations covering all 4 agroclimatic zones I, II, IIIA and IIIB of Bihar infecting mustard (*Brassica juncea*) revealed a distinct variation in terms of morphological characteristics viz. length, breadth, beak length and septations of conidia. The average conidial length, thickness, beak length, no. of transverse septa and no. of longitudinal septa varied from 150 to 243µm, 22.1 to 31.5 µm, 67 to 113.4 µm, 10.8 to 16 and 2.6 to 4.2 respectively. Severity of Alternaria blight disease of mustard in the term of percent disease intensity (PDI) also varied among source location of 15 isolates from 35.3 to 61.85%. Cultural characteristics like colony diameter, colour, growth pattern, margin and pigmentation among isolates also showed variability. The pathogenicity testing of isolates on mustard cultivar Varuna, revealed variable response among fifteen isolates. Four isolates namely MAB 01 Mzp, MAB 06 Prn, MAB 09 Bnk and MAB 14 Rts belonging to different districts under agroclimatic zones of Bihar as zone I, II, IIIA and IIIB respectively were found highly pathogenic and virulent in aggressiveness by causing spot size of more than 20 mm. However, seven isolates namely MAB 02 Gog, MAB 03 Swn and MAB 04 Cpr from zone I; MAB 07 Kth from zone II; MAB 08 Bgp and MAB 11 Skp from zone IIIA and MAB 15 Kmr from zone IIIB were found moderately virulent or pathogenic by causing mean spot size of 10-20 mm. Other 4 isolates were found as virulent or least pathogenic by causing spot size < 10 mm. This study may be useful in the development of integrated disease management strategy for Alternaria blight of mustard using breeding programs.

Keywords: Alternaria blight, mustard, *A. brassicae* isolate, cultural, pathogenicity, variability

Introduction
Rapeseed-Mustard are globally known as "Oilseed brassica", which holds the status of the third most important oilseed crop after soyabean and palm with the production of about 72 MT from about 35 m ha area. In terms of area and production, India stands third place after Canada and China, and fifth place in terms of productivity after Germany, France, Canada and China. (Jat *et al*., 2019) [8]. It is grown all over India in both tropical and subtropical regions covering 6.23 m ha of area producing 9.34 MT with 1499 kg/ha average productivity. In India, Rajasthan stands the first in its production covering the area of 2.37 m ha producing 4.08 MT with the average productivity of 1720 kg/ha. Bihar produces 0.11 MT from an acreage of 0.08 m ha with average productivity of 1305 kg/ha. (Agricultural Statistics at a Glance, 2019) [2]. In India, rapeseed mustard shares 23.5% area and 24.2% production of total oilseeds in the country. Despite being the third largest producer (11.3%) of oilseed brassica in the world, India meets 57% of the domestic edible oil requirements through imports and ranked 7th largest importer of edible oils in the world (Jat *et al*., 2019) [8].
Productivity of the crop, in the region is low due to a number of foliar diseases, viz., *Alternaria* blight, white rust, downy mildew and powdery mildew. Among these, Alternaria blight incited by *Alternaria brassicae* (Berk.) Sacc. is the most important and devastating disease. It has been reported to cause variable losses in yield, depending upon disease severity. Yield loss to the extent of 47 per cent has been reported (Chattopadhyay *et al.*, 2008 and Meena *et al.*, 2010) [4, 10]. The fungus, not only leads to yield reduction by causing foliar damage to the crop, but also damages siliqua in pod formation stage, severely deteriorating both seed and oil yield. (Choudhary et al. 2018) [5].
There are some reports on the existence of variability in mycelial growth, morphology and sporulation among different isolates of *A. brassicae* from different geographical regions of India (Ansari *et al.*, 1989; Kaur *et al.*, 2007; Goyal *et al*., 2011) [1, 9, 10]. Considering paramount importance of Alternaria blight disease of mustard crop, the present investigation focused on

evaluation of morphological, cultural and pathogenic variability of fifteen isolates of *A. brassicae* of mustard from Bihar that is important to design disease management strategies by breeding resistant cultivars. Relationship of morphological variations among *A. brassicae* isolates with their pathogenic variability has also been investigated.

Materials and Methods

Survey of Alternaria blight of mustard and collection of isolates: To study the presence of variability of the pathogen in the attacked host, specimens of diseased plant parts characteristically showing typical symptoms of *Alternaria* blight on leaves having several brownish spots especially with concentric rings were collected during the survey of the disease from different locations of 15 districts of 4 different agroclimatic zones of Bihar. Survey for the PDI of the disease were done on the same date at same place in both years and mean data was calculated. Collected diseased specimens were wrapped in clean poly bags and were labelled according to the place and district they were collected from, respectively. The infected leaves were also preserved in refrigerator for further studies. These specimens were further used for microscopic examination, subsequent isolation, pathogenicity and variability studies. (Table 1).

Table 1: Severity status of Alternaria blight of Indian mustard in different parts of Bihar at Pod development growth stage during 2020-22

SL. No.	District (Place)	Latitude and longitude	Agro-Climatic Zones	Survey Dates	Per cent Disease Intensity			Name of Isolates
					2021	2022	Mean	
1	Muzaffarpur (TCA Dholi)	25°59'31.8"N 85°35'40.0"E	I	15.02	61.4	62.3	61.85	MAB 1 Mzp
2	Gopalganj (Jigna Gopal)	26°20'34.0"N 84°20'35.5"E	I	16.02	51.2	56.1	53.65	MAB 2 Gog
3	Siwan (Chhap)	26°15'22.8"N 84°21'02.7"E	I	16.02	45.8	57.1	51.45	MAB 3 Swn
4	Chhapra	25°47'38.9"N 84°45'25.6"E	I	17.02	44.7	54.3	49.5	MAB 4 Cpr
5	Araria (Sisauna)	26°06'47.8"N 87°28'18.1"E	II	20.02	35.7	47.7	41.7	MAB 5 Arr
6	Purnia (Baisa)	25°55'34.7"N 87°32'22.9"E	II	20.02	35.1	48.4	41.75	MAB 6 Prn
7	Katihar (Simaria)	25°34'39.3"N 87°30'37.5"E	II	21.02	43.7	51.8	47.75	MAB 7 Kth
8	Bhagalpur (Chaudharidih)	25°13'03.6"N 86°59'54.2"E	IIIA	22.02	32.8	37.8	35.3	MAB 8 Bgp
9	Banka (Khushalpur)	25°05'24.2"N 86°59'19.2"E	IIIA	22.02	40.5	34.5	37.5	MAB 9 Bnk
10	Munger (Hemjapur)	25°18'03.5"N 86°22'58.2"E	IIIA	23.02	40.8	47.1	43.95	MAB 10 Mgr
11	Sheikhpura (Ariyari)	25°21'59.2"N 85°32'10.7"E	IIIA	24.02	41.4	48.1	44.75	MAB 11 Skp
12	Patna (Bihta)	25°34'36.7"N 84°52'27.6"E	IIIB	28.02	36.5	55.1	45.8	MAB 12 Ptn
13	Bhojpur (Kulhariya)	25°34'40.5"N 84°46'59.5"E	IIIB	25.02	40.4	48.4	44.4	MAB 13 Bjp
14	Rohtas (Kumahu)	24°58'40.0"N 83°56'51.7"E	IIIB	27.02	41.2	56.6	48.9	MAB 14 Rts
15	Kaimur (Kudra)	25°03'08.7"N 83°47'19.7"E	IIIB	26.02	40.5	58.6	49.55	MAB 15Kmr
	C.D. at 0.05: 10.229; SE(m):3.513; C.V.:13.079 * Pooled Mean							

Isolation of the fungus: Infected plant parts showing symptoms of *Alternaria* blight were selected for collecting isolates. Small pieces of 3-5 mm size occurring next to healthy plant portion were cut and washed 3-4 times with tap water followed by their sterilization using 0.1% mercuric chloride (Hg Cl_2) solution for 30 seconds and washing 3-4 times in sterile distilled water to wash out all traces of Hg Cl_2 which were then aseptically transferred between the two-fold of sterilized blotting paper to remove excess amount of water to prevent bacterial contamination. Subsequently these pieces were aseptically transferred on to PDA poured petri plates and slants with the help of sterile inoculating needle which was plugged tightly with non-absorbent cotton and incubated in a BOD incubator at 25 ± 1ºC for 7 days to obtain a culture *A. brassicae* which produce circular white and smooth colonies with fluffy appearance. After seven days of incubation, the growing mycelium from the margin of apparently distinct colonies was sub-cultured on fresh PDA petri dishes. Culture of *A. brassicae* thus obtained was sealed with a paraffin wax strip and stored in a freezer to accomplish further investigations. In this way, the cultures of different isolates were obtained.

Identification and purification of the fungus: Temporary slides of the cultures of 15 *A. brassic*ae isolates prepared in lactophenol were examined under compound microscope to observe the characteristics of mycelia and spores. On the basis of their conidiophore and conidial morphology as described by Simmons (2007) [16], the pathogen was identified as *A. brassicae* (Berk.) Sacc. The pathogen *of A. brassicae* was purified by single spore technique. Single spore technique was used for maintaining the purity of each isolate (Gattani., 1954) [6]. Thus, fifteen isolates collected from different districts of Bihar were designated as Mustard Alternaria Blight Isolate (MAB) with numbers from 1 to 15 and 3 letters of the name of district from where they were collected viz. MAB-1 Mzp for Muzaffarpur, MAB-2 Gog for Gopalganj, MAB 3 Swn for Siwan, MAB 4 Cpr for Chapra, MAB 5 Arr for Araria, MAB 6 Prn for Purnia, MAB 7 Ktr for Katihar, MAB 8 Bgp for Bhagalpur, MAB 10 Mgr for Munger, MAB 11 Skp for Sheikhpura, MAB 12 ptn for Patna, MAB 13 Bjp for Bhojpur, MAB 14 Rts for Rohtas and MAN 15 Kmr for Kaimur districts.

Morphological variability of *A. brassicae* isolates: Among the fifteen *A. brassicae* isolates, morphological variability was evaluated using micrometry technique (Meena et al., 2005) [11]. In each slide 10 spores were examined at 40X magnification under light microscope. Ocular and stage micrometer were used to measure the size, shape and beak length of conidia and number of septation.

Cultural variability of *A. brassicae* isolates: The cultural characteristics of the fifteen isolates like colony, color, texture, growth, shape, margin and zonation were assessed by direct observation of 15 days old culture-grown on PDA incubated at 25±2 °C temperature.

Pathogenic variability of *A. brassicae* isolates: To confirm the identification of the disease and its causal agent, the pathogenicity test was carried out under polyhouse conditions under pot experiments using mustard (*B. juncea*) cultivar

Varuna. Pots were filled with sterilized soil and seedling were raised in. Pathogenicity test of the isolates was done, on one month aged healthy plants of mustard var. Varuna grown in pots under glasshouse condition. 15 days old cultures of *A. brassicae* on PDA was taken, blended with sterilized distilled water, and filtered through cheesecloth. The spore suspensions containing 15 to 20 spores/ml were taken in a 100 ml atomizer separately for each isolate. All the leaves of the *Brassica* species were inoculated in triplicate for each isolate of *A. brassicae* and one set was kept as check in which only sterilized water was sprayed. The inoculated plants were separately kept isolate wise in moist chambers. After 7 days of inoculation size of spots was measured. On the basis of mean size of spots developed the isolates were categorized as virulent or pathogenic for the diameter < 10 mm, moderately virulent for spot diameter of 10-19 mm and highly virulent for spot diameter of > 20 mm. Experiments were conducted in completely randomized design (CRD).

Results and Discussion

Severity status of Alternaria blight disease of mustard in Bihar: Significant variability in PDI of Alternaria blight disease of mustard at pod development growth stage during 2020-22, was observed among 15 different mustard growing districts of Bihar (Table1). The data of mean PDI of 2 years in Table 1 revealed that the highest severity of the disease (61.85%) was found at Tirhut college of Agriculture, Dholi in Muzaffarpur district followed by (53.65%) in Gopalganj district. The data confirmed that the Dholi is the hot spot for Alternaria blight disease of mustard. The minimum severity of 35.3% was observed in Bhagalpur followed by that of 37.5% in Banka district. Severity of Alternaria blight of mustard in the form of PDI ranged from 35.3% to 61.85% among 15 districts of Bihar. The disease severity in four different agroclimatic zones viz. I, II, IIIA and IIIB was observed and recorded in the range of 49.5% to 61.85%, 41.7% to 47.75%, 35.3 to 44.75%, and 44.4 to 49.55% respectively. The highest disease severity was observed in agroclimatic zone I followed by zone IIIB and II with lowest in zone IIIA.

Morphological variability among isolates: In terms of conidial length, conidial width, conidial beak length, and number of transverse and longitudinal septa of *A. brassicae*, significant (P<0.05) morphological variation was observed among its 15 isolates of mustard. Maximum conidial length of *A. brassicae* was recorded for MAB 12 Ptn (242µm) which was significantly differed from MAB 04 Cpr (190µm), MAB 05 Arr (165µm), MAB 06 Prn (150µm), MAB 14 Rts (163µm) and MAB 15 Kmr (184µm). The minimum conidial length was recorded for the isolate MAB 06 Prn (150µm) followed by MAB 14 Rts (163µm. Maximum conidial thickness at the broadest part was recorded for MAB 01 Mzp (31.5µm) followed by MAB 12 Ptn (31.4µm) which were significantly higher than MAB 04 Cpr (23.4µm), MAB 05 Arr (22.1µm), MAB 06 Prn (23.5µm), MAB 07 Kth(22.6µm) and MAB 14 Rts (22.3µm). The minimum conidial thickness was recorded for MAB 05 Arr (22.1µm), followed by MAB 14 Rts (22.3µm) which were significantly at par with MAB 02 Gog (26.5µm), MAB 04 Cpr (23.4µm), MAB 05 Arr (22.1µm), MAB 06 Prn (23.5µm), MAB 07 Kth(22.6µm), MAB 14 Rts (22.3µm), MAB 09 Bnk (27.3µm) and MAB 15 Kmr (26.4µm) (Table 2). The maximum conidial beak length was recorded for MAB 10 Mgr (113.4µm) which was at par with 7 other isolates viz. MAB 01 Mzp(112.1µm), MAB 02 Gog(102.1µm), MAB 08 Bgp(102.7µm), MAB 09 Bnk(110.1µm), MAB 13 Bjp (95µm), MAB 15 Kmr(92.9µm). The minimum conidia beak length was observed for MAB 06 Prn(67µm) which is at par with MAB 04 Cpr(70.7µm), MAB 05 Arr (70.1µm), MAB 07 Kth(72.4µm), MAB 11 Skp(88.1µm), MAB 14 Rts(76.4µm).

Average number of transverse septa in conidia ranged from 10.8 to 16 among different isolates with the highest number shown by MAB 08 Bgp (16) followed by MAB 12 Ptn (15.3) and MAB 10 Mgr (15), whereas MAB 15 Kmr (10.8) showed minimum number of transverse septa. For the average number of longitudinal septa the maximum was found in MAB 09 Bnk (4.2) and minimum (2.6) in MAB 04 Cpr isolate (Table 2).

These observed and analyzed results are in accordance with earlier workers (Awasthi and Kolte, 1989; Meena *et al.,* 2005; Sharma *et al*., 2013; Pramila *et al*., 2014) [3, 11, 15, 13], who also reported variability in morphology of *A. brassicae* isolates from difference places in India. Goyal *et al.* (2011) [7] reported variabilty in conidial morphology, mycelial growth and sporulation of thirteen isolates of *A. brassicae* collected from seven states of India. Similarly, Meena *et al.*, 2005[11]; Sharma *et al*., 2013[15] and Pramila *et al*., 2014[13] reported variability in the morphological characteristics in *A. brassicae* isolates collected from different places of India.

Table 2: Conidial morphology of different15 isolates of *A. brassicae* of Mustard in Bihar*

A. brassicae isolates	Length with beak (µm)	Thickness at the broadest part (µm)	Beak length (µm)	No. of Septa	
				Transverse	Longitudinal
MAB 01 Mzp	230	31.5	112.1	14.4	3.1
MAB 02 Gog	210	26.5	102.1	13.7	3.9
MAB 03 Swn	202	28.5	90.7	13.7	3.7
MAB 04 Cpr	190	23.4	70.7	13.3	2.6
MAB 05 Arr	165	22.1	70.1	13.4	2.8
MAB 06 Prn	150	23.5	67	13.6	3.1
MAB 07 Kth	181	22.6	72.4	14.5	3.4
MAB 08 Bgp	200	30.4	102.7	16	3.1
MAB 09 Bnk	220	27.3	110.1	14.3	4.2
MAB 10 Mgr	230	30.3	113.4	15	3.6
MAB 11 Skp	237	29.7	88.1	13.2	3.8
MAB 12 Ptn	242	31.4	91	15.3	3.6
MAB 13 Bjp	213	28.5	95	15	3.7
MAB 14 Rts	163	22.3	76.4	11	3.1
MAB 15 Kmr	184	26.4	92.9	10.8	3.2
C.D.	49.693	5.786	22.371		

SE(m)	17.734	2.065	7.984		
C.V.	27.882	24.221	27.955		
*Average of 10 conidia in each isolate.					

Cultural characteristic of *A. brassicae* isolates: Colony characters of pure culture of 15 isolates measured at 12 days of incubation at 25±1℃, showed marked variability in different characteristics. Radial growth of the isolate's colony, varied from 40 mm in MAB 10 Mgr to 56 mm in MAB 02 Gog and colour of the colony varied from olivaceous black to whitish black. Growth pattern of isolates were either adherent circular or fluffy circular with absent to multiple zonation with variation in margin as smooth thin white/gray and wavy thin gray. The pigmentation of the colony was observed as pinkish white, light brown, brown and dark brown (Table-3). The observed results are in accordance with Ansari *et al.*, 1989 [1]; Patni *et al.*, 2005 [12]; Kaur *et al*., 2007 [9]; Sharma *et al*., 2013 [15] who also reported the cultural variability in *Alternaria* species in respect of mycelial growth and sporulation.

Table 3: Colony characters of *A. brassicae* isolates of mustard in Bihar on PDA media after 12 days of incubation at 25± 1 ℃

A. brassicae isolates	Pure Culture (mm)*	Colony colour **	Growth pattern@	Zonation	Sectors	Margin#	Pigmentation
MAB 01 Mzp	55	OB	AC	One	No	STW	Dark brown
MAB 02 Gog	56	OB	FC	No	No	STW	Dark brown
MAB 03 Swn	54	OB	AC	One	No	STW	Brown
MAB 04 Cpr	53	BW	AC	No	No	STW	Brown
MAB 05 Arr	47	BW	AC	Multiple	No	WTG	Brown
MAB 06 Prn	48	BW	AC	Multiple	No	WTG	Brown
MAB 07 Kth	44	BW	AC	No	No	WTG	Brown
MAB 08 Bgp	45	OB	FC	No	No	STW	Brown
MAB 09 Bnk	42	WB	AC	Two	No	STG	Dark brown
MAB 10 Mgr	40	WB	AC	No	No	STW	Brown
MAB 11 Skp	43	WB	AC	No	Present	STW	Brown
MAB 12 Ptn	53	WB	AC	No	No	STW	Pinkish white
MAB 13 Bjp	51	WB	AC	Two	No	STG	Brown
MAB 14 Rts	52	WB	AC	No	No	STW	Brown
MAB 15 Kmr	53	WB	AC	No	No	SWO	Light brown
Mean	49.07	*Average of three replications.					
SE(m) ±	1.585	**Colony Colour: W: White, OB: Olivaceous black, B: Black, WB: Whitish Black, BW: Blackish White.					
C.D (P<0.05)	4.599	@Growth Pattern: AC= Adherent circular; FC= Fluffy circular.					
CV%	5.594	#Margin: STW=Smooth, thin and white, WTG= Wavy, thin and grey, STG= Smooth, thin and grey, SWO= Smooth, wide and olive.					

Pathogenic variability among isolates: Fifteen different isolates of *A. brassicae* of mustard showed pathogenic variability on host *B. juncea* cultivar Varuna. All the 15 isolates collected from different districts of Bihar were found to be pathogenic in nature (Table 4). On the basis of mean size of lesion produced after 7 days of inoculation, as of more than 20 mm, 4 isolates namely MAB 01 Mzp, MAB 06 Prn, MAB 09 Bnk and MAB 14 Rts were found and rated as highly virulent followed by 10-19 mm of the produced lesion size, 7 isolates namely MAB 02 Gog, MAB 03 Swn, MAB 04 Cpr, MAB 07 Kth, MAB 08 Bgp, MAB 11 Skp and MAB 15 Kmr were moderately virulent. Other 4 isolates namely MAB 05 Arr, MAB 10 Mgr, MAB 12 Ptn, MAB 13 Bjp produced lesions of size less than 10 mm and rated as virulent. Similar observations were reported by Saha *et al.*, 2014[14] who estimated pathogenic variability based on disease development by twenty-three *A. brassicae* isolates of cauliflower (*Brassica oleracea*). Pramila *et al*. (2014) [13] also reported pathogenic variability on the basis of size of spots among 10 isolates of *A. brassicae* collected from Indian mustard.

Table 4: Pathogenicity testing of *A. brassicae* isolates on mustard var. Varuna

A. Brassicae isolates	Pathogen Aggressiveness	Mean lesion size (mm)
MAB 01 Mzp	Highly virulent	25
MAB 02 Gog	Moderate virulent	17
MAB 03 Swn	Moderate virulent	18
MAB 04 Cpr	Moderate virulent	15
MAB 05 Arr	Virulent	8
MAB 06 Prn	Highly virulent	23
MAB 07 Kth	Moderate virulent	12
MAB 08 Bgp	Moderate virulent	14
MAB 09 Bnk	Highly virulent	23
MAB 10 Mgr	Virulent	8
MAB 11 Skp	Moderate virulent	13
MAB 12 Ptn	Virulent	7
MAB 13 Bjp	Virulent	8
MAB 14 Rts	Highly virulent	23
MAB 15 Kmr	Moderate virulent	14

C.D.	2.625
SE(m)	0.901
C.V.	10.273

Conclusion

The results of present investigations clearly depicted the existence of morphological, cultural and pathogenic variability in *A. brassicae* isolates collected from all different agroclimatic zones of Bihar. Significant variability in PDI of Alternaria blight disease of mustard was observed as the disease severity in four different agroclimatic zones of Bihar viz. I, II, IIIA and IIIB was found in the range of 49.5% to 61.85%, 41.7% to 47.75%, 35.3 to 44.75%, and 44.4 to 49.55% respectively with the highest severity of the disease (61.85%) at Dholi, Muzaffarpur and lowest of 35.3% in Bhagalpur.The results also show high level of de-arrangement with no clear grouping of *A. brassicae* isolates and non-existence of correlation among the morphological, cultural and pathogenic variability of different isolates. But in the present study 4 isolates namely MAB 01 Mzp, MAB 06 Prn, MAB 09 Bnk and MAB 14 Rts belonging to different districts under agroclimatic zones of Bihar as zone I, II, IIIA and IIIB respectively were found as highly pathogenic and virulent aggressiveness. However, 3 isolates namely MAB 02 Gog, MAB 03 Swn and MAB 04 Cpr from zone I, one isolate MAB 07 Kth from zone II, two isolates viz. MAB 08 Bgp, MAB 11 Skp from zone IIIA and only single isolate MAB 15 Kmr from zone IIIB, were moderately virulent or pathogenic. Other 4 isolates were found as virulent or least pathogenic. The evaluation of pathogenic variability among *A. brassicae* isolates is crucial for the development of most suitable management strategy for Alternaria blight disease according to different agroclimatic zones. This result of cultural, morphological and pathogenic variability may be useful in developing integrated disease management strategies and breeding programs for the mustard crop.

Acknowledgements

The authors are thankful to the Head, Department of Plant Pathology, PGCOA, Dr Rajendra Prasad Central Agricultural University Pusa, Bihar for providing necessary facilities and encouraging support in carrying out the present investigation.

References

1. Ansari NA, Khan MW, Muheet A. Effect of some factors on growth and sporulation of *Alternaria brassicae* causing Alternaria blight of rapeseed and mustard. Acta Bot. Ind. 1989;17:49-53.
2. Agricultural Statistics at a Glance. Directorate of Economics & Statistics, DAC & FW, Ministry of A & FW, Govt. of India, 2019, 73.
3. Awasthi RP, Kolte SJ. Variability in *Alternaria brassicae* affecting rapeseed and mustard. Indian Phytopath. 1989;42:275.
4. Chattopadhyay C, Agrawal R, Kumar A, Bhar LM, Meena PD, Meena RL, *et al.* Epidemiology and forecasting of Alternaria blight of oilseed Brassica in India-a case study. J Plant Dis. Protect. 2008;112:351-365.
5. Choudhary CS, Mishra AK, Singh RS, Mukherjee U, Pandey A. Management of *Alternaria* Blight of Indian Mustard in Bihar. Int. J Cur. Microbiol. App. Sci. 2018;7:1053-1058.
6. Gattani ML. Agar plate spore germination method for testing fungicides. Phytopathology. 1954;44:113-115.
7. Goyal P, Chahar M, Mathur AP, Kumar A, Chattopadhyay C. Morphological and cultural variation in different oilseed Brassica isolates of *Alternaria brassicae* from different geographical regions of India. Indian J Agric. Sci. 2011;81(11):1052-1058.
8. Jat RS, Singh VV, Sharma P, Rai, PK. Oilseed brassica in India: Demand, supply. policy perspective and future potential. OCL, 2019, 26(8). Available online at: www.ocl-journal.org
9. Kaur S, Singh G, Banga SS. Documenting variation in *Alternaria brassicae* isolates based on conidial morphology, fungicidal sensitivity and molecular profile. In: Proceeding of the 12th International Rape-seed Congress, GCIRC, Wuhan, China. 2007 Mar;4:87-89.
10. Meena PD, Gupta R, Rani A, Sharma P, Rai PK, Chowdappa P. Morphological and cultural variability among *Alternaria brassicae* isolates from India. National Symposium on Molecular Approaches for Management of Fungal Diseases of Crop Plants. Bangalore, 2010, 184-185.
11. Meena PD, Chattopadhyay C, Kumar VR, Meena RL, Rana US. Spore behaviours in atmosphere and trends in variability of *Alternaria brassicae* population in India. J. Mycol. Plant Pathol. 2005;35:511.
12. Patni CS, Kolte SJ, Awasthi RP. Cultural variability of *Alternaria brassicae*, causing Alternaria blight of mustard. Annals of Plant Physiology. 2005;19(2):231-242.
13. Pramila, Giri P, Tasleem M, Taj G, Mal R, Kumar A. Morphological, cultural, pathogenic and molecular variability amongst India isolates of *Alternaria brassicae* in Uttarakhand. African Journal of Bio-technology. 2014;13(3):441-448.
14. Saha S, Garg R, Venkataravanappa V, Mishra PK. Molecular and Cultural Characterization of *Alternaria brassicae* Infecting Cauliflower in Uttar Pradesh, India. Proc. Natl. Acad. Sci., India, Sect. B Biol. Sci., 2014. DOI 10.1007/s40011-014-0472-y.
15. Manika S, Swati D, Dinesh SB, Chowdappa P, Selvamani R, Sharma P. Morphological, cultural, pathogenic and molecular studies of *Alternaria brassicae* infecting cauliflower and mustard in India. African Journal of Microbiology Research. 2013;7(26):3351-3363.
16. Simmons EG. Alternaria: an identification manual. CBS, Utrecht publisher, Netherland, 2007.