

## Incidence of Mycoflora in Indian Mustard (*Brassica juncea*) Seeds in Rajasthan

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**ABSTRACT:** Indian mustard [*Brassica juncea* (L.) Czern & Coss] is globally used as oilseed, vegetable and condiments. Mustard seeds are known to carry several pathogenic and non-pathogenic fungi which alter physiochemical properties of the seeds, losses of the seed weight, germination potential, and discoloration. *Alternaria* spp. is the most destructive pathogen of oilseeds in major growing areas. The aim of this study is to draw a systematic and comprehensive picture of important seed borne fungi of mustard which affect the production and productivity of mustard in major growing areas of Rajasthan. During present investigation, 100 seed samples were collected from ten major mustard growing districts of Rajasthan. Ten samples of each district were thoroughly mixed and one composite seed sample was established from every district and associated seed mycoflora and their incidence were quantified through blotter and agar plate methods. Results of dry seed analysis showed shriveled, discolored and damaged seeds in each sample. The maximum incidence (35.77 %) of seed mycoflora was recorded in un-surface sterilized seed sample of Alwar district and lowest in Bikaner and Jaipur sample (5.00%). A total of sixteen saprophytic as well as parasitic mycoflora (*Alternaria brassicae*, *Alternaria alternata*, *A. brassicicola*, *Aspergillus niger*, *A. flavus*, *A. nidulans*, *A. fumigatus*, *A. ochraceus*, *Curvularia lunata*, *Drechslera tetramera*, *Rhizopus nigricans*, *Mucor* sp., *Penicillium* sp., *Chaetomium* sp., *Metarrhizium* sp. and *Fusarium oxysporum*) belonging to nine genera were detected through Blotter and Agar Plate Methods. In conclusion, huge loads of mycoflora adversely affect seed germination and plant health which ultimately results in poor stand of crop and yield.

**Keywords:** Indian mustard, *Brassica juncea*, seed mycoflora.

### INTRODUCTION

The contribution of oilseeds to the agriculture economy of India ranks second only to food grains (Rathore *et al.*, 2018). *Brassica juncea* (L.) Czern & Coss belongs to family *Brassicaceae* is commonly known as Indian mustard and globally used as oilseed, vegetable and condiments (Saleem *et al.*, 2017). India is one of the leading oilseeds producing country in the world accounting for 11.2 per cent of the world's rapeseed-mustard production and ranks third in the world next to China and Canada. It is the second most important oilseed crop of India after groundnut in terms of area and production. In India, the crop was grown over 6.412 million hectares of land producing 6.33 million tonnes (Anonymous, 2017-18). Major Indian mustard growing states are Rajasthan, UP, Haryana, Punjab, Madhya Pradesh and Gujarat (Shekhawat *et al.*, 2012). Meena and Yadav (2018) stated that Rajasthan is the most giant mustard growing state and alone contributes 43 per cent of the total mustard seed production in India. In Rajasthan, rapeseed-mustard occupies a prime place amongst all the oilseed crops grown. Rajasthan ranks first both in area and production of rapeseed-mustard in the country. It is grown in the districts

namely Ajmer, Alwar, Bharatpur, Dausa, Jaipur, Sawai-Madhopur, Bikaner, Kota, Baran, Tonk. In Rajasthan, rapeseed-mustard comprising an area under cultivation was about 2.379 million hectares in the year 2018-19 with production of 3.588 million tonnes (Anonymous, 2018-19).

Indian mustard is the premier oilseed Brassica which covers about 85-90 per cent of the total area under cultivation of all these oilseed crops (Rao *et al.*, 2017). In India, mustard crop is grown in *Rabi* season from September-October to February-March. Sandy loam to clay loam soils but thrive best on light loam soils. Soils having neutral pH are ideal for their proper growth and development. Rapeseed-mustard oil is considered the best quality oil for human consumption as compared to other edible oils because of the lowest amount of harmful saturated fatty acids and adequate amount of two essential fatty acids i.e. linoleic acid and linolenic acid (Porter and Crompton, 2008). Mustard seed contains about 38 to 43 per cent oil which is yellowish in colour (Patel *et al.*, 2012).

In agriculture, depending upon the presence of fungi either on seed coat or in the seed, it is further called as external seed-borne fungi and internal seed-borne fungi, respectively. Ismail *et al.* (2012) stated that seed health

plays an important role in any crop production system. The goals of better yield cannot be achieved without the use of disease free seeds as they serve the important source of spread of various diseases. Singh *et al.* (2014) observed that rapeseed-mustard suffers with a number of diseases and insect-pests. Among the diseases, *Alternaria* blight, downy mildew, white rust and powdery mildew are of major significance and stated that *Alternaria brassicae* infects host species at all growth stages and affects seed germination and both quality and quantity of oil in mustard seeds. Ghosh *et al.* (2018) studied the, association of mycoflora adversely affects quality and health of seeds. They detected many fungal species in seed samples of oil seed crops and these were *Alternaria* sp., *Curvularia* sp., *Fusarium* sp., *Helminthosporium* sp., *Penicillium* sp., *Mommoniella* sp., *Aspergillus* sp., *Mucor* sp. and *Rhizopus* sp. and among these, *Alternaria* sp. as well as *Aspergillus* sp. were the most destructive pathogen of oilseeds. Dutta (2007) detected 31 and 20 fungal species from surface sterilized mustard seeds by Agar Plate and Blotter Methods, respectively. It was noted from the results that the percentage occurrence of fungal population increased with the increasing storage time in both Agar Plate and Blotter Methods. Mumtaz and Fatima (2017) found that the Agar Plate Method was to be suitable for isolation of mycoflora as there was higher per cent incidence of seed mycoflora. Shivpuri *et al.* (1990) collected eighty two seed samples of mustard from nine agro-climatic zones of Rajasthan and sixteen species of fungi were isolated from these samples. These are *Aspergillus caesiellus*, *A. nidulans*, *A. sejuntus*, *A. versicolor*, *Alternaria brassicae*, *pmoniliforme*, *F. acuminatum*, *F. oxysporum*, *Phoma lingam* and *P. rebulosa* etc. Ninety per cent of food crops are propagated by seed. Most widely grown oilseed crops in world agriculture (groundnut, sesame, mustard etc.) are all affected by seed borne diseases. Seeds of mustard (*Brassica campestris* L.) are known to carry several fungal pathogens which alter physio-chemical properties of the seeds during storage, losses of the seed weight, germination potential, medicinal properties and discoloration, causing the losses to the extent of 24 per cent (Ashraf and Choudhary, 2008). In India, various researchers have studied the incidence of seed borne fungi of several species of Brassica under storage environment from various geographical locations (Ghotekar and Hedawoo, 2010; Ghugal and Thakre, 2014). Siddiqui (2013) detected seed borne mycoflora of mustard (*Alternaria* sp., *Rhizoctonia* sp., *Aspergillus* sp., *Mucor* sp. and *Rhizopus* sp.) and concluded that the full potential of this crop is far from being exploited due to several abiotic and biotic stresses. The infected seeds may fail to germinate, transmit disease from seed to seedling and from seedling to growing plants.

## MATERIAL AND METHODS

### A. Collection of seed samples

Hundred seed samples of mustard were collected from 10 major mustard growing districts of Rajasthan (Table

1) during 2018 to know the incidence and adverse impact on seed health and quality. Two samples were collected from each village, five villages were selected in a tehsil and one tehsil was selected in a district. Therefore, a total of ten samples of mustard seeds were collected from one district and seed of each district mixed thoroughly and made one composite sample of every district.

### B. Examination of dry seeds

The method suggested by Agarwal and Sinclair (1987) with slight modification was followed. Twenty gram seeds from each sample were taken at random and divided into four fractions (5.0 g). Each fraction was spread on bottom of a Petri plate and examined with the help of a hand lens or if required under stereobionocular microscope. The inspected material was categorized as deformed seeds (shriveled), discolored (reddish brown), damaged seeds (by insects), impurities (plant debris: pieces of leaves and pods), inert material (stones and sand) and apparently healthy seeds. Seed and impurities of each category were pooled separately and weighed on modern electric balance and per cent content by weight was calculated.

### C. Detection of seed mycoflora

For detecting seed borne mycoflora, the seed health testing procedures as prescribed by International Seed Testing Association (ISTA, 1976) were followed for estimating incidence of mycotic genera. Four hundred and two hundred seeds per sample of mustard were tested by Blotter and Agar Plate Method, respectively.

## RESULTS AND DISCUSSION

### A. Dry seed examination

The dry seed inspection of ten composite seed samples of mustard belonging to major growing districts of Rajasthan was examined (Table 2). Each sample was categorized in five groups. Maximum deformity, in the form of shriveling (19.25%) and reddish brown discoloration (21.00%) was observed in Alwar sample and lowest in Bikaner sample (1.08% and 2.70%, respectively). Mechanically damaged seeds and impurities in the form of plant debris and inert materials (stone and sand/ash particles) were found in all the samples. Maximum apparently healthy seeds were observed in Bikaner sample (91.94%) while it was minimum (52.85%) in Alwar sample. Inspection of dry seed samples revealed the presence of deformed (shriveled), discolored (reddish brown) and damaged (insects and mechanically) seeds, in addition to inert material (impurities) and apparently healthy seeds. Presence of such deformation and discoloration along with impurities have also been reported during examination of dry seeds of gram, cowpea, moth bean and sesame by Ahmed and Reddy (1993); Khatik (1988); Goyal (1996); Cheema (1997); Mali (2001); Singh and Singh (1983), respectively.

**Table 1: Places of seed sample collection of mustard from major growing areas of Rajasthan.**

Sr. No.	Places of sample collection			Code No. (Composite Sample)
	District	Tehsil	Village	
1.	Ajmer	Kishangarh	Chitakheda, Chundari, Ganeshpura, Gudali, Khatoli	Maj
2.	Alwar	Ramgarh	Goha, Khedi, Mastabad, Niwali, Piproli	Mal
3.	Baran	Anta	Badi Pachel, Badusui, Bamla, Choti Pachel, Kachari	Mb
4.	Bharatpur	Nadbai	Bhadira, Kabai, Nadbai, Pachora, Ronija	Mbp
5.	Bikaner	Lunkarsar	Kakadwala, Kalavas, Lunkarsar, Malkisar, Ronjha	Mbk
6.	Dausa	Lalsot	Achalpura, Deedwana, Devali, Gumanpura, Khanpur	Md
7.	Jaipur	Jamwa Ramgarh	Andhi, Saipura, Gopalgarh, Herrawala, Lali	Mj
8.	Kota	Digod	Digod, Kacholiya, Kauruwa, Parasliya, Toran	Mk
9.	Sawai Madhopur	Gangapur City	Heerapura, Lalpura, Mahunkala, Motipur, Saloda	Msm
10.	Tonk	Uniara	Ahmadnagar, Kachrawta, Khedli, Uniara, Uniari	Mt

**Table 2: Seed abnormalities and impurities in different mustard seeds samples.**

Sr. No.	Categories of seed/impurities	Per cent content (by weight basis) per sample*									
		Mal	Maj	Md	Mbp	Mb	Mk	Msm	Mj	Mt	Mbk
1.	<b>Deformed seeds:</b>										
	(i) Shriveled	19.25	9.50	8.50	7.50	6.90	6.50	5.25	5.20	4.00	1.08
2.	<b>Discolored seeds:</b>										
	(i) Reddish brown	21.00	19.50	18.00	17.50	20.00	14.00	10.25	9.25	8.25	2.70
3.	<b>Damaged seeds:</b>										
	(i) Mechanically	3.00	1.50	2.00	1.50	1.00	0.50	1.50	0.45	1.50	1.23
	(ii) Insects	1.25	0.50	0.20	0.35	1.50	0.10	0.35	0.06	0.10	0.02
4.	<b>Impurities:</b>										
	(i) Plant debris	1.15	2.00	1.00	2.00	2.00	1.80	2.00	1.80	2.15	3.00
	(ii) Inert material	1.50	1.65	1.39	1.50	1.65	1.15	1.06	0.43	1.10	0.03
5.	<b>Apparently healthy seeds</b>	52.85	65.35	68.91	69.65	66.95	75.95	79.59	82.81	82.90	91.94

\*Average of four replications (5 g seeds / replication)

**Detection of mycoflora.** The per cent incidence for each fungus was recorded from untreated as well as surface sterilized seeds on Blotter and surface sterilized seeds on Agar Plate Method.

**Through Standard Blotter Method (Un-surface sterilized).** Fifteen fungal species belonging to nine genera were isolated from untreated mustard seeds (Table 3) in Blotter Test. Fungi and their respective per cent incidence were *Alternaria brassicae* (0.25-4.75%), *Alternaria alternata* (0-2.50%), *A. brassicola* (0-

0.75%), *Aspergillus niger* (0.75-3.0%), *A. flavus* (0-3.75%), *A. nidulans* (0-2.25%), *A. fumigatus* (0-1.25%), *A. ochraceous* (0-0.75%), *Curvularia lunata* (0-2.50%), *Drechslera tetramera* (0-1.50%), *Rhizopus nigricans* (0-4.50%), *Mucor* sp. (0-2.50%), *Penicillium* sp. (0-4.25%), *Chaetomium* sp. (0-1.25%) and *Fusarium oxysporum* (0-4.00%). Total per cent mycoflora was recorded maximum in Alwar seed sample (35.75%) while it was minimum (5.00%) in Bikaner sample.

**Table 3: Per cent incidence of mycoflora of mustard seeds isolated by Standard Blotter Method (un-surface sterilized).**

Mycoflora	Per cent mycoflora / sample*										
	Mal	Md	Maj	Mbp	Mk	Msm	Mt	Mb	Mj	Mbk	Mean
<i>Alternaria alternata</i>	2.50	1.50	2.25	1.25	1.50	1.00	1.75	0.50	0.0	0.25	1.25
<i>Alternaria brassicae</i>	4.75	3.25	2.50	2.25	2.50	0.25	1.25	2.25	1.00	0.50	2.05
<i>Alternaria brassicola</i>	0.50	0.75	0.25	0.0	0.75	0.0	0.0	0.0	0.0	0.0	0.22
<i>Aspergillus flavus</i>	3.75	2.50	1.50	1.50	2.25	0.25	0.0	1.00	1.25	0.0	1.40
<i>Aspergillus fumigatus</i>	1.25	0.75	0.75	0.50	0.25	0.0	0.25	0.50	0.0	0.0	0.42
<i>Aspergillus nidulans</i>	2.25	1.25	2.25	0.50	0.25	0.50	1.25	0.0	0.0	0.75	0.90
<i>Aspergillus niger</i>	3.00	1.75	2.50	1.50	2.50	0.75	0.75	1.50	1.00	1.25	1.60
<i>Aspergillus ochraceous</i>	0.25	0.50	0.0	0.0	0.0	0.75	0.0	0.0	0.0	0.50	0.20
<i>Chaetomium</i> sp.	1.25	0.0	0.50	0.25	0.0	0.0	0.0	0.0	0.0	0.0	0.20
<i>Curvularia lunata</i>	2.00	1.25	0.0	2.50	1.25	2.00	1.00	0.0	0.0	0.0	1.00
<i>Drechslera tetramera</i>	1.50	0.0	1.25	0.0	0.50	0.0	0.0	0.0	0.0	0.0	0.32
<i>Fusarium oxysporum</i>	4.00	2.00	3.00	2.50	0.0	0.25	0.0	1.75	0.0	0.0	1.35
<i>Mucor</i> sp.	0.0	2.50	1.50	0.0	0.25	0.0	1.25	0.50	0.25	0.0	0.62
<i>Penicillium</i> sp.	4.25	2.25	2.75	0.50	0.0	2.50	0.0	0.0	1.50	1.75	1.55
<i>Rhizopus nigricans</i>	4.50	2.25	2.50	3.25	1.50	0.0	0.50	0.50	0.0	0.0	1.50
<b>Total % mycoflora</b>	35.75	22.50	23.50	16.50	13.50	8.25	8.00	8.50	5.00	5.00	

\*Sample size 400 seeds

**Through Standard Blotter Method (Surface sterilized).** Eleven fungal species belongs to seven genera were isolated from surface sterilized seeds (Table 4) in Blotter Test. Fungi and their per cent incidence recorded were *Alternaria brassicae* (0-5.75%), *A. alternata* (0.25-3.00%), *A. brassicicola* (0-2.25%), *Aspergillus niger* (0.5-2.50%), *A. flavus* (0-2.00%), *A. fumigatus* (0-0.75%), *Curvularia lunata*

(0-1.50%), *Drechslera tetramera* (0-0.75%), *Penicillium* sp. (0-2.5%), *Chaetomium* sp. (0-0.25%) and *Fusarium oxysporum* (0-4.50%). Total per cent mycoflora was recorded maximum in Alwar seed sample (25.00%) while it was minimum (2.50%) in Bikaner sample.

**Table 4: Per cent incidence of mycoflora of mustard seeds isolated by Standard Blotter Method (surface sterilized).**

Mycoflora	Per cent mycoflora / sample*										
	Mal	Maj	Md	Mbp	Mb	Mk	Msm	Mj	Mbk	Mt	Mean
<i>Alternaria alternata</i>	3.00	2.50	3.00	2.25	1.75	0.75	0.50	2.00	0.25	1.75	1.77
<i>Alternaria brassicae</i>	5.75	4.75	4.25	3.25	1.25	1.75	0.0	2.50	0.75	0.0	2.42
<i>Alternaria brassicicola</i>	2.25	1.75	0.75	0.0	0.0	0.0	0.0	0.0	0.25	0.0	0.50
<i>Aspergillus flavus</i>	2.00	1.25	1.50	0.0	1.75	0.50	1.00	0.0	0.0	1.25	0.92
<i>Aspergillus fumigatus</i>	0.0	0.50	0.50	0.25	0.75	0.25	0.0	0.0	0.0	0.0	0.22
<i>Aspergillus niger</i>	2.50	1.50	1.25	1.25	0.50	0.75	1.75	0.50	0.75	0.50	1.12
<i>Chaetomium</i> sp.	0.25	0.25	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.05
<i>Curvularia lunata</i>	1.50	0.0	1.00	0.25	0.0	0.0	0.50	0.25	0.0	0.25	0.37
<i>Drechslera tetramera</i>	0.75	0.25	0.0	0.50	0.0	0.0	0.0	0.0	0.0	0.0	0.15
<i>Fusarium oxysporum</i>	4.50	2.75	0.0	2.25	0.0	2.50	2.00	0.0	0.50	1.75	1.62
<i>Penicillium</i> sp.	2.50	1.25	0.25	2.25	0.50	1.25	0.75	0.0	0.0	0.0	0.87
<b>Total % mycoflora</b>	<b>25.00</b>	<b>16.75</b>	<b>12.50</b>	<b>12.25</b>	<b>6.5</b>	<b>7.75</b>	<b>6.50</b>	<b>5.25</b>	<b>2.50</b>	<b>5.50</b>	

\*Sample size 400 seeds

**Through Potato Dextrose Agar Plate Method (Surface sterilized).** Twelve fungi were detected from all the ten seed samples by Agar Plate Method (Table 5). Fungi and their per cent occurrence recorded were *Alternaria brassicae* (0- 10.0%), *A. alternata* (0-5.5%), *A. brassicicola* (0-2.70%), *Aspergillus niger* (0.25-2.0%), *A. flavus* (0-1.5%), *A. nidulans* (0-

0.5%), *A. ochraceous* (0-1.0%), *Curvularia lunata* (0-3.0%), *Drechslera tetramera* (0-1.0%), *Metarrhizium* sp. (0-2.5%), *Penicillium* sp. (0- 2.0%) and *Fusarium oxysporum* (0-6.5%). Total per cent mycoflora was maximum (36.70%) in Alwar sample while it was minimum (1.00%) in Bikaner sample.

**Table 5: Per cent incidence of mycoflora of mustard seeds isolated by Agar Plate Method.**

Mycoflora	Per cent mycoflora / sample*										
	Mal	Maj	Md	Mbp	Mk	Mb	Mt	Msm	Mj	Mbk	Mean
<i>Alternaria alternata</i>	5.5	4.0	2.5	1.0	1.0	0.0	0.0	0.0	0.0	0.0	1.40
<i>Alternaria brassicae</i>	10.0	6.0	8.5	3.8	0.5	0.5	0.0	1.5	0.0	0.0	3.00
<i>Alternaria brassicicola</i>	2.7	1.2	1.2	0.5	0.0	0.5	0.2	0.0	0.5	0.0	0.68
<i>Aspergillus flavus</i>	1.5	1.0	0.5	0.5	0.0	0.5	0.0	0.0	0.0	0.0	0.40
<i>Aspergillus nidulans</i>	0.5	0.0	0.5	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.10
<i>Aspergillus niger</i>	2.0	0.25	1.5	1.5	1.0	1.5	0.5	0.5	0.5	0.5	0.92
<i>Aspergillus ochraceous</i>	1.0	0.5	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.15
<i>Curvularia lunata</i>	3.0	2.5	0.0	0.0	1.0	1.5	1.7	0.0	0.5	0.0	1.02
<i>Drechslera tetramera</i>	1.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.10
<i>Fusarium oxysporum</i>	6.5	4.5	2.0	3.5	2.0	1.0	0.0	1.0	0.0	0.0	2.05
<i>Metarrhizium</i> sp.	1.0	2.5	0.0	1.5	0.5	1.0	0.0	0.0	0.0	0.5	0.70
<i>Penicillium</i> sp.	2.0	1.0	0.0	0.0	1.5	0.0	0.5	0.0	0.5	0.0	0.55
<b>Total % mycoflora</b>	<b>36.70</b>	<b>23.45</b>	<b>16.70</b>	<b>12.30</b>	<b>7.50</b>	<b>6.50</b>	<b>2.90</b>	<b>3.00</b>	<b>2.00</b>	<b>1.00</b>	

\*Sample size 200 seeds

All the samples were analyzed through Blotter Method and Agar Plate Methods using both un-surface sterilized seeds and surface sterilized seeds. Throughout the study period, a maximum number of fungal species could be isolated by Blotter and the Agar Plate Methods, which may be due to the availability of high moisture content in the blotter's plate and the availability of fungal nutrients in the agar plate. These results are in agreement with the findings of Neergaard and Saad (1962); Singh *et al.* (1984) they concluded that the blotter and the agar plate methods are both valuable and supplementary to each other.

**Effect of surface sterilization (0.1% mercuric chloride) on seed borne fungi.** In Blotter Method, besides untreated seeds, surface sterilization of seeds

with 0.1 per cent mercuric chloride solution to seeds, was also used. In general, the chlorine surface sterilization reduced saprophytes to a considerable extent and their growth on seeds surface was also rendered sparse. However, in Blotter Test, the fungi recorded *Aspergillus nidulans*, *Aspergillus ochraceous*, *Rhizopus nigricans* and *Mucor* sp. were found only on untreated seeds and the surface sterilized samples showed decrease in per cent incidence value of *Aspergillus niger*, *Aspergillus flavus*, *Aspergillus fumigatus*, *Penicillium* sp., *Chaetomium* sp., *Curvularia lunata* and *Drechslera tetramera*. An increase in per cent incidence value of *Alternaria brassicae*, *Alternaria alternata*, *Alternaria brassicicola* and *Fusarium oxysporum* were observed.

In the present investigation, a total of sixteen saprophytic as well as parasitic fungal species belonging to nine genera i.e. *Alternaria brassicae*, *Alternaria alternata*, *Alternaria brassicicola*, *Aspergillus niger*, *Aspergillus flavus*, *Aspergillus nidulans*, *Aspergillus fumigatus*, *Aspergillus ochraceous*, *Curvularia lunata*, *Drechslera tetramera*, *Rhizopus nigricans*, *Mucor* sp., *Penicillium* sp., *Chaetomium* sp., *Metarrhizium* sp. and *Fusarium oxysporum* were isolated from mustard seeds in Blotter method and Agar Plate Method. This finding is in line with the work of Qumberani (2001); Siddiqui (2013); Bhajbhuj (2014); Ghosh *et al.* (2018) while working on the seed-borne mycoflora of mustard.

**Comparison between the blotter and agar plate method.** Per cent incidence of fungi varied in Blotter Method and Potato Dextrose Agar Plate Method (PDA). PDA revealed lesser number of fungi as compared to Blotter Test. A total of 15 and 12 fungal species belonging to 9 and 7 genera were isolated in Blotter and PDA Tests, respectively. In Agar Plate Test, *Aspergillus flavus*, *Aspergillus nidulans*, *Aspergillus ochraceous*, *Aspergillus niger*, *Drechslera tetramera* and *Penicillium* sp., showed considerable low incidence whereas *Alternaria brassicae*, *Alternaria alternata*, *Alternaria brassicicola*, *Curvularia lunata* and *Fusarium oxysporum* were detected in high incidence as compared to Blotter Test. While some genera show no incidence in Agar Plate test, *Chaetomium* sp., *Mucor* sp. and *Rhizopus nigricans*. In general, variation was observed in Blotter Method and Agar Plate Method. Higher per cent counts of deep seated or pathogenic mycoflora were observed in Agar Plate Method as compared to Blotter Method. This variation might be due to the reasons that some of the weak and slow growing fungi could not grow in Blotter Method in comparisons to fast growing saprophytic fungi. Pre-surface sterilization of seeds and different substratum used in the method employed may be another reason (de-Tempe, 1961, Neergaard and Sadd, 1962; Jain, 1990).

## CONCLUSION

The maximum incidence (35.77 %) of seed mycoflora was recorded in un-surface sterilized seed sample of Alwar district and lowest in Bikaner and Jaipur sample (5.00%). A total of sixteen saprophytic as well as parasitic mycoflora (*Alternaria brassicae*, *Alternaria alternata*, *A. brassicicola*, *Aspergillus niger*, *A. flavus*, *A. nidulans*, *A. fumigatus*, *A. ochraceous*, *Curvularia lunata*, *Drechslera tetramera*, *Rhizopus nigricans*, *Mucor* sp., *Penicillium* sp., *Chaetomium* sp., *Metarrhizium* sp. and *Fusarium oxysporum*) belonging to nine genera were detected through Blotter and Agar Plate Methods. The fungal species recorded in PDA test were common to those observed in Blotter Test but per cent incidence of fungi varied in Blotter Test and Potato Dextrose Agar Plate Method (PDA). In conclusion, huge loads of mycoflora adversely affect seed germination and plant health which ultimately results in poor stand of crop and yield.

## FUTURE SCOPE

There is a significant economic impact of seed borne diseases particularly in underdeveloped countries where routine chemical treatment of seeds is prohibitively expensive and individual farmers can suffer huge yield reduction. Considering the importance of a rapeseed-mustard crop in the Indian economy, the urgent need for undertaking the basic and strategic research for stabilizing and increasing the production and productivity of mustard in our country. The present investigation has opened up new information and given rise to new ideas on seed borne fungi associated with seeds of Indian mustard. Hence the futures lines of work are needed with there is a need to undertake an intensive survey for associated seed borne mycoflora of mustard in major growing areas of Rajasthan and quantify the losses caused by them. To identify these fungal species helps to successful disease management. Research in this field will give direction and helps to reduce disease intensity in mustard crop. Integrated disease management strategies for seed borne mycoflora of mustard need to be developed.

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