

## Original Research Article

### Screening of Seed Borne Mycoflora of *Cicer arietinum* L.

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#### A B S T R A C T

#### Keywords

*Cicer*;  
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*Aspergillus flavus*;  
*Botrytis cinerea*;  
*Fusarium*  
Seed Dressing.

*Cicer arietinum* is found prone to several fungal diseases. These diseases are soil borne due to prolonged saprophytic survival of the pathogen through sclerotia. Seed borne mycoflora of five cultivars of *Cicer* were studied under blotter paper method. All these varieties were found more susceptible to *Fusarium*, *Aspergillus niger*, *Aspergillus flavus*, *Botrytis cinerea*, *Sclerotium rolfsii*. Most susceptible variety was KAK2. Six fungicides were tested for their efficacy. Seeds treated with Indofin resulted in reduction of total seed mycoflora and enhanced the germination percentage by JG11 (64 to 69%), ICCV 95311(68 to 87%), KAK2 (58 to 77%), ICCV92944 (70 to 78%). The seedling vigor index was maximum (976.8) in the seeds treated with carbendazim of ICCV92944 variety followed by JG11 variety treated with captan. While least (72) vigor index was observed in KAK2 variety treated with *Trichoderma*.

#### Introduction

Chickpea (*Cicer arietinum* L.) is the third most important pulse crop, after dry bean and dry pea. As per the latest report of Food and Agriculture Organization (FAO) for 2011, India is the largest producer and consumer of chickpea in the world. In 2012-13 estimated production of chickpea is 8567.8 thousand tons.

Andhra Pradesh, shows six fold increase in production (621 kg/ha to 1397 kg/ha) in consequential increase area under chickpea cultivation (71,000 ha in 1991-93 to 5,17,000 ha in 2006-08). Chickpea is a

prime pulse crop of Rabi and is the cheapest source of protein and is the inseparable part of the daily diet of every Indian. It also has carbohydrates, Zinc, folic acid and a little fat.

The yield potential of present-day Chickpea cultivar exceeds 4 t/ha. However, average yield is less than 0.8t/ha, the gap between average yield is mainly due to disease and poor management practices. It is found attacked by about 67 fungi, 3 bacteria, 22 viruses and mycoplasmas and 80 nematodes

(Nene *et al.*, 1996). Major diseases in order of their of global magnitude are Ascochyta blight, Fusarium wilt, Botrytis gray mould, dry root rot, Verticillium wilt, Rust etc. Black root rot wilt caused by *Fusarium solani* (Mart.) Sacc. is one of the most serious disease which causes severe yield loss i.e., 60-70 per cent under favourable conditions (Tewari and Mukhopadhyay, 2003). The disease is both seed and soil- borne and affects seed germination and vigour to a great extent. To increase the production of Chickpea qualitatively and quantitatively farmers require healthy and quality seeds, with high percentage of germination and purity. Hence it is imperative that seeds must be tested before they are sown in the field. Another adverse effect of seed-borne pathogen is that it will contaminate the areas which were disease free previously. So it necessitates the eradication of seed-borne inoculum through various seed treatment and through enforcement of proper domestic and international quarantine act and procedures.

The seed vigor is debated by the infestation of fungal species like *Alternaria porri*, *A. alternata*, *Aspergillus amstelodami*, *A. flavus*, *A. fumigatus*, *A. nidulans*, *A. niger*, *A. sydowi*, *A. wentii*, *Botrytis cinerea*, *Cladosporium macrocarpum*, *Curvularia lunata*, *Fusarium equiseti*, *F. moniliforme*, *F. oxysporum*, *F. semitectum*, *Macrophomina phaseolina*, *Myrothecium roridum*, *Penicillium notatum*, *Rhizoctonia* sp., and *Rhizopus arrhizus* been reported from chickpea (Ahmad *et al.*, 1993). Of the different diseases *Ascochyta* blight caused by *Ascochyta rabiei* (Nene, 1980) is desolating .

The present investigation is conducted to study the effect of seed mycoflora on

germination and vigor index. The study includes seed health testing of different *Cicer* cultivars, and integrated disease management through use of seed dressing by fungicides.

## Materials and Methods

Five *Cicer* seed varieties Jaki 9218, JG11, ICCV95311, KAK2 and ICCV92944 were collected from Andhra Pradesh Seeds Development Corporation (APSDC). Standard blotter method was used to evaluate seed quality. Both untreated seeds and treated with fungicides (Bavastin, Trichoderma, Mancozeb, Indofil, Carbendazim, Captan) were placed in Petri plates with three layers of moistened blotters using sterile distilled water (10 seeds/ Petri). For each variety ISTA techniques (Anon, 1976) were used to study the mycoflora, where 40 untreated and 40 treated seeds per variety were used for the blotters test. The Petri plates were incubated at  $28^{\circ} \pm 2^{\circ}\text{C}$  under 12hr of alternating cycle of light and darkness for 7days. Seeds from each sample were examined for the percentage of mycoflora per 30 seeds under binocular microscope on the seventh day .

Germination percentage for all the varieties using paper towel method (ISTA 1996) was carried out. Three replications of 100 seeds by random pick were placed on the paper towel, which were kept in a beaker containing water for two weeks and also by Petri plate method where ten seeds per petriplate were kept. Shoot length and Root length were recorded for each seed in each sample. Seedling vigor index was calculated by multiplying the percentage of germination with the sum of root and shoot length that is expressed in centimeters (Abdul baki and Anderson 1973).

## Results and Discussion

The results of this study indicates the dominance of *Fusarium* sp. (50%) in all the varieties that were considered for study. It was found maximum in Jaki 9218 and ICCV92944 followed by *Aspergillus niger* (50%) in KAK2. Other fungi included species of *Botrytis* and *Sclerotium*. (Table 1) Present observation is in concurrence with Bretag and Mebalds (1987) and Sing *et al.*, (2005). This indicates that all these varieties are susceptible to seed borne fungi hence use of fungicides as an alternate practice to control the disease.

Singh *et al.*, (2002) have made similar observation in the *in vitro* study of some fungicides like Captan, Dithane M-45, Vitavax, and Bavistin for their efficacy in controlling *Fusarium* species on mungbean. Fungicidal treatment induced metabolic changes leading to development of toxic factors, resulting in the internal environment unfavorable for pathogens growth and activity, ultimately inducing the resistance and protection against infection.

Fungicides not only effectively increased the germination percentage 68 to 88% in ICCV 95311, 58 to 82 KAK2 variety, 64 to 74% in JG11 variety 70 to 72% in ICCV92944 variety when treated with captan (Table2) but also decreased the percent incidence of mycoflora in different varieties of *Cicer*. Captan showed best results in germination as well as in mycoflora control. These results are in concurrence with the observations made by Parimala *et al.*, (1998), Sandrou *et al.*, (1998) and Kumar and Dubey (2001) in cowpea, blackgram, brinjal and sunflower, respectively. Reports of De and Chaudhary (1999) are affirmative of the

present findings, who observed the minimization of seed mycoflora due to Bavistin. Fungicide treated seeds enhanced the rate of germination when compared to control. Untreated seeds recorded the highest seed mycoflora percentage and lowest seed germination percentage (58%).

In the present study, six fungicides were tested for their efficacy in overcoming seed borne infection, among them Indofil was observed to be effective followed by captan. Root length in JAKI 9218 is highest in seeds dressed with indofin followed by Carbendazim of the same variety (Figure 1). Shoot length is maximum in ICCV92944 variety treated with carbendazim followed by untreated variety of ICCV9511; minimum shoot length was observed in JG11 variety treated with captan in a fourteen day seedling. Most of the varieties both in treated and untreated showed an average shoot length of 0.5cm after 14 days (Figure 2).

Vigor Index was maximum in treated seeds of ICCV92944 (976) followed by JG 11(751). (Fig3). *Trichoderma viridae* treated seeds were more efficient than fungicides. Bunker and Mathur (2000) also reported the similar results with usage of *T. harzianum*. Present findings are in agreement with the reports of several workers (Dey *et al.*, 1989; Chakrabarty and Rao, 1992; Laxminaryan *et al.*, 1996). Thus, present study highlights the importance of seed treatment with most effective fungicides, to reduce the incidence mycoflora on *Cicer* seeds.

Seeds treated with bioagents showed beneficial effects on germination which resulted in increased root-shoot length and seedling vigor and also reduce the

mycofloral species that impede the seed germination. Incidence of diseases such as Ascochyta blight and root rot which affect production and quality of the produce. Use of resistant cultivars, crop rotation and other crop management practices need to be promoted. Significant loss during

storage, pest is reported in chickpea. Scientific pest management measures along with use of improved storage structures should be promoted. Creation of better grading, cleaning and packaging facilities quality seeds

**Table.1** Percentage of infection in thirty seeds set as three replications of ten seeds per variety

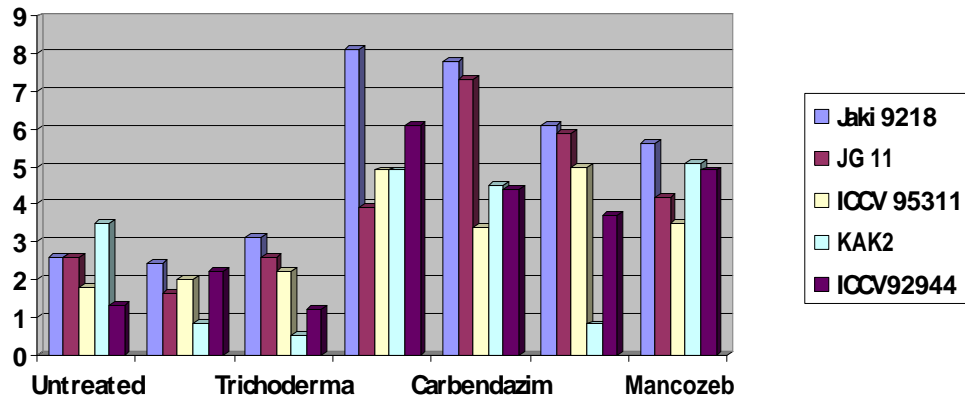
Percentage of infection					
<i>Cicer</i> Variety	<i>Fusarium sp.</i>	<i>Aspergillus niger</i>	<i>Aspergillus flavus</i>	<i>Botrys cinerea</i>	<i>Sclerotium rolfsii</i>
Jaki 9218	50	22	5	2	6
JG 11	40	27	5	3	5
ICCV 95311	48	30	15	2	5
KAK2	40	50	5	3	2
ICCV92944	50	28	10	5	2

**Table.2** Germination response of *Cicer* varieties in treated and untreated seeds.

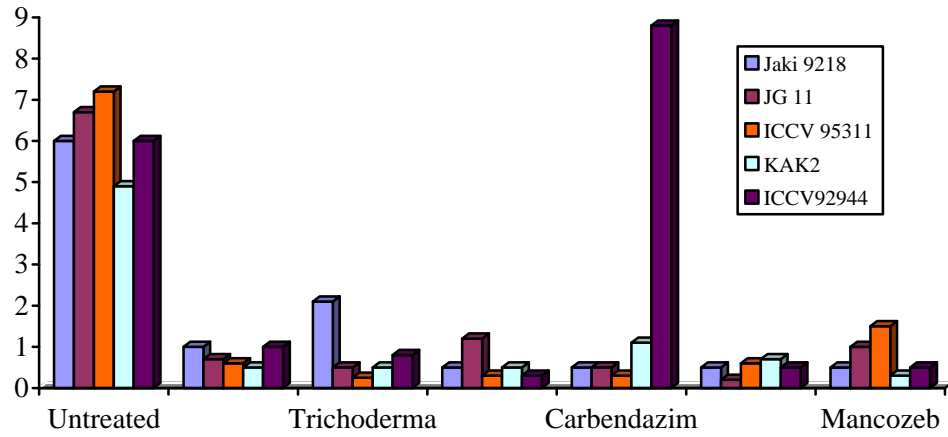
<i>Cicer</i> Variety	Percentage of Germination*						
	Untreated	Bavastin	Trichoderma	Indofil	Carbendazim	Captan	Mancozeb
Jaki 9218	72	71	68	67	66	58	71
JG 11	64	68	74	69	78	74	69
ICCV 95311	68	80	81	87	84	88	82
KAK2	58	68	72	77	81	82	80
ICCV92944	70	62	75	78	74	72	70

\*Average germination percentage of thirty seeds set as three replications of ten seeds.

**Figure.1** Root Length in Centimeters after seven days



**Figure.2** Shoot Length in Centimeters after fourteen days



**Figure.3** Seedling Vigor Index

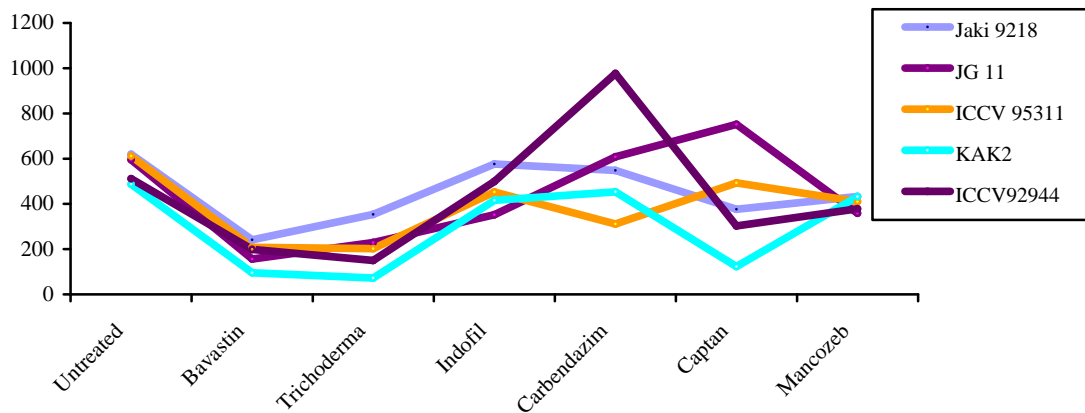


Plate 1: Seven day old seedlings



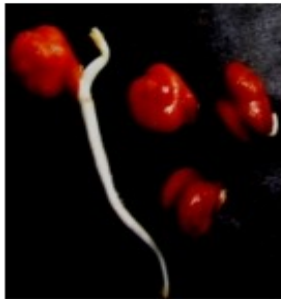
JG11 Trichoderma



JG11 Bavastin



JG11 Control



ICCV 92944 Trichoderma



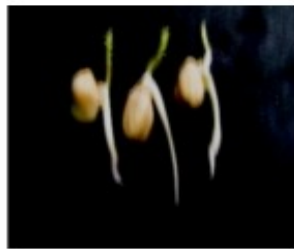
ICCV 92944 Bavastin



ICCV 92944 Control



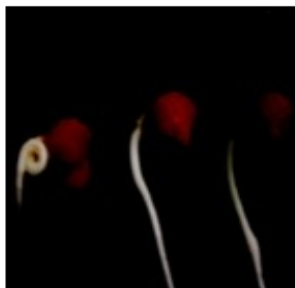
ICCV 95311 Trichoderma



ICCV 95311 Bavastin



ICCV 95311 Control



JAKI Trichoderma



JAKI Bavastin



JAKI Control

## References

- Abdul baki, A.A., and Anderson, J.D. 1973. Vigour determination in soybean seeds by multiple criteria. *Crop Sci.* 13:630-633.
- Ahmad, I., S. Iftikhar and Bhutta. A.R. 1993. Seed borne microorganism in Pakistan. A checklist 1991. Pakistan Agricultural Research Council, Islamabad, Pakistan. pp. 32.
- Bretag T.W., and Mebalds, M.I. 1987. Pathogenicity of fungi isolated from *Cicer arietinum* (Chickpea) grown in North- Western Victoria. *Australian J.Exp.Agric.* 27:141-148.
- Bunker, R.N., and Mathur, K. 2000. Integration of biocontrol agents and fungicides for suppression of dry rot of *Capsicum frutescens*. *J. Mycol. Plant Pathol.*, 31(3): 330-334.
- Chakrabarty, S.K., and Rao, R.D.J.P. 1992. Eradication of *Fusarium moniliforme* from maize seeds. *Indian J. Plant Protect.* 20: 105-107.
- De, K.R., and Chaudhary, R.G. 1999. Biological and chemical seed treatment against lentil wilt. *LENS Newslett.* 26(1&2): 28-31.
- ISTA., 1996. International rules for seed testing science and technology 13:299-513.
- Kandil, A.A., A.E. Sharief and Ahmed, S.R.H. 2012. Germination and Seedling Growth of Some Chickpea Cultivars (*Cicer arietinum* L.) under Salinity Stress. *J. Basic. App. Sci.* 8: 561-571.
- Kumar, D., and Dubey, S.C. 2001. Management of collar rot of pea by the integration biological and chemical methods. *Indian Phytopathol.* 54(1): 62-66.
- Laxminarayan, C., S.K. Sinha, S.K. Aujla, Consul, S.N. Sinha, M.M Payak, Y.L. Nene and Renfereo, B.C. 1996. Fungicidal seed treatment of maize in India. *Indian Phytopathol. Bull.* 3: 18-26.
- Nene, Y.L., 1980. A Review of *Ascochyta* blight of chickpea. *Trop. Pest. Manage.* 28: 61-70.
- Parimala, K., P. Thangavel, K. Saravanan, A. Anbuselvan and Ganesan, J. 1998. Effect of plant growth regulators and chemicals on seed germination in blackgram. In: proceedings of National symposium of future goal of physiological research for the improvement of plant research. 2: 42-47.
- Sandrou, S., K. Wahab and Kavitha, R. 1998. Effect of hardening on physiological growth and yield parameters of sunflower cv. Modern. In: proceedings of National symposium of future goal of physiological research for the improvement of plant research. 2:70-75.
- Sing Kiran., A.K. Sing and Singh R.P. 2005. Detction of seed mycoflora of chickpea (*Cicer arietinum*) *Ann. Pl. Protec.Sci.* 13(1):1-4.
- Singh, S.D., P. Rawal, N.S. Shekawat and Lodha, P.C. 2002. Management of mungbean (*Vigna radiata* (L.) Wikzek) seed mycoflora by seed dressing fungicides. *J. Mycol. Plant Pathol.* 23(1): 149.
- Tewari, A.K., and Mukhopadhyay, A.N. 2003, Management of chickpea root rot and collar rot by integration of biological and chemical seed treatment. *Indian Phytopath.* 56 (1):
- Woodstock, L.W., 1969. Seedling growth as a measure of seed vigour. *Proceedings of the ISTA.* 34(2): 273-80.