ISSN: 0974 - 0376

The Ecoscan : Special issue, Vol. IX: 431-435: 2016
AN INTERNATIONAL QUARTERLY JOURNAL OF ENVIRONMENTAL SCIENCES
www.theecoscan.com

EFFECT OF DIFFERENT SEED BORNE MYCO FLORA ON VIGOR OF WHEAT SEED SAMPLES COLLECTED FROM SOUTHERN RAJASTHAN

Hansraj Dhakar *et al.,*

KEYWORDS

Wheat
Myco Flora
Germination
Vigor

Proceedings of National Conference on
Harmony with Nature in Context of
Resource Conservation and Climate Change
(HARMONY - 2016)
October 22 - 24, 2016, Hazaribag,
organized by
Department of Zoology, Botany, Biotechnology & Geology
Vinoba Bhave University,
Hazaribag (Jharkhand) 825301
in association with
National Environmentalists Association, India
www.neaindia.org

The Ecoscan
AN INTERNATIONAL QUARTERLY JOURNAL OF ENVIRONMENTAL SCIENCES

HANSRAJ DHAKAR*, R. S. RATNOO, UMESH KUMAR DHAKAD AND ANITA JAT
Department of Plant Pathology, RCA, MPUAT, Udaipur - 313 001
e-mail: rajdhakar1@gmail.com

ABSTRACT

Seed borne myco flora of Wheat in two district of southern Rajasthan (Chittorgarh and Udaipur) were surveyed. A total of 80 seed samples, 40 of each district were collected during 2015-16. Data were recorded for per cent seed germination, per cent mortality and seedling vigor of wheat tested by seed inoculum, soil inoculum and seedling symptoms test. All the isolated myco flora significantly reduce the vigor of wheat seed samples. Maximum reduction in germination per cent *Curvularia lunata* (30.24%) radicle length (23.40mm) and plumule length (39.53mm) were due to *Fusarium moniliforme* tested by seed inoculum technique. In case of soil inoculum technique maximum reduction in plumule length (34.76mm) and radical length (17.65mm) by *Fusarium moniliforme*. In seedling symptoms test seedling mortality was ranged from 6.23% (*Trichoderma viride*) to 19.67% (*Fusarium moniliforme*).

***Corresponding author**

INTRODUCTION

Seeds are regarded as primary source of the infection for some of the diseases itself (Bhoyar *et al.*, 2014). Numerous examples exist in agriculture literature for the international spread of plant disease as a result of importation of seeds that were infected or contaminated with pathogens (Agarwal and Sinclair, 1996; Halt, 1994). Seed myco flora are chiefly responsible for the deterioration of seed in storage and reduce the viability of seeds and germination is also influenced (Singh *et al*, 2014). A seed borne pathogen may cause seed abortion, seed rot, seed necrosis, reduction or elimination of germination capacity as well as seedling damage resulting in development of disease at later stages of plant growth by systemic or local infection (Khanzada *et al*., 2002 ~ Bateman and Kwasna, 1999).

Many seed borne myco flora of wheat reported recently included *Alternaria alternata, Drechslera sorokiniana, Fusarium* spp. *Cladosporium herbarum*, *Stemphylium botryosum* (Glazek, 1997 and Mirza and Qureshi, 1978). These plant pathogenic fungi are also known to produce phytotoxic metabolites (Sharma *et al.*, 2001). So, keeping these things in view the present investigation aimed to isolate associated myco flora and their effects on vigor of wheat seed samples collected from two districts (Chittorgarh and Udaipur) of southern Rajasthan.

MATERIALS AND METHODS

Experimental location

The experiment was conducted at Department of Plant Pathology, Rajasthan College of Agriculture (RCA) MPUAT, Udaipur during year 2015-16.

Sources of experimental materials

Eighty seed samples of wheat were collected for detection of effect of seed borne myco flora on seedling vigor of wheat from Chittorgarh and Udaipur district of southern Rajasthan. 40 seed sample were collected from each district. All materials except seeds, which used in this experiment, were sterilized using 70% ethyl alcohol.

Seed inoculation technique

These method is followed by as described by Khokar (2012) in this apparently healthy surface sterilized seeds were taken. The seeds were rolled on 8 days old sporulating culture of individual fungi thriving on PDA in Petri plates. Sixteen (1 + 6 + 9) seeds per Petri plate were plated by using standard blotter paper test. The uninoculated surface sterilized apparently healthy seeds served as control. Observations on per cent germination and mortality were recorded on 10 days while seedling vigor (root and shoot length) were recorded on 15 days after germination.

Soil inoculation technique (Khokar, 2012)

All seed borne fungi separately and multiplied in sterilized rice medium (20 gm rice + 10 ml distilled water). Five hundred ml conical flasks were used for each organism containing one third of rice medium. These flasks were inoculated with 7 days old culture. The inoculated flasks were incubated at 25 + 2^0C for 10 days. Flasks were shaken every day to avoid clumping. The pots were filled with sterilized soil. Soil

was infested by each microorganism inoculum. For soil inoculation, upper 4 cm layer of the soil was thoroughly mixed with culture grown in rice medium @ 10g/pot. Pots were kept in pot house for 24 h for proper soil infestation before sowing of wheat seeds. Seeds were surface sterilized (1:1000 $HgCl_2$) before sowing followed by 3 washing with sterile distilled water and were sown in inoculated soil at the rate of 20 seeds per pot. Hundred seeds were sown for testing pathogenicity of each microorganism. A set of control was also kept with surface sterilized seeds sown in sterilized uninoculated soil. Pots were watered at regular interval. Observations on germination and mortality were recorded on 10 days while seedling vigor (root and shoot length) were observed on 15 days after germination.

Agar Test: Seedling symptoms in plain agar test tubes (Singh *et al.*, 2011)

One hundred seeds from each sample selected randomly were used. The seedling were raised in 160 x 16 mm test tubes each containing 10 ml of 1 per cent water agar (10 gm agar in 1000 ml distilled water) and plugged with a cotton plug and sterilized at 1.045 kg cm^{-2}. One seed was placed in each test tube and incubated at $25 + 2\,^0C$ under alternating 12 hr of light with 12 hr of dark period. After 5 days, plugs were removed and kept for 7 days for incubation and then seedlings were examined for symptoms appearance.

RESULTS AND DISCUSSION

All the seed samples (forty from each district) collected were mixed well and used for observing myco flora effect on germination and seedling vigor.

Seed inoculation technique

Healthy surface sterilized wheat seeds were taken. The seeds were rolled on 7 days old sporulating culture of individual myco flora and placed on PDA to observe per cent seed germination, per cent mortality and seedling vigor.

It is evident from fig 1 that seed germination and vigor of seedlings were influenced by the myco flora associated with seeds. The germination was markedly suppressed by the entire myco flora tested but the maximum inhibition of germination was observed in the seeds inoculated with *Curvularia lunata* (30.24%) followed by *Fusarium moniliforme* (19.08%), *Alternaria alternata* (13.68%) *Aspergillus flavus* (6.19%), *Trichoderma viride* (5.27%), *Mucor* spp.(3.31%), *Aspergillus niger* (2.66%), and *Rhizopus stolonifer* (2.36%) as compare to control. However, germination was obtained when the seeds were inoculated with *Curvularia lunata* (56.39%), *Fusarium moniliforme* (67.55%), *Alternaria alternata* (72.95), *Aspergillus flavus* (80.44%), *Trichoderma viride* (81.36%), *Mucor* spp. (83.32%), *Aspergillus niger* (83.97%), and *Rhizopus stolonifer* (84.27%).

Showing of data in fig 1also reveals that, maximum reduction in radicle length was due to *Fusarium moniliforme* (23.40mm) followed by *Curvularia lunata* (30.42mm), *Aspergillus flavus* (32.49mm), *Alternaria alternata* (35.60mm), *Aspergillus niger* (41.27mm), *Rhizopus stolonifer* (42.39mm), *Mucor* spp. (43.49mm) and *Trichoderma viride* (44.61mm) as compared to control (45.44mm).

Maximum reduction in plumule length was due to *Fusarium moniliforme* (39.53mm) followed by *Curvularia lunata* (46.58mm), *Aspergillus flavus* (52.44mm), *Alternaria alternata* (56.50mm), *Aspergillus niger* (63.79mm), *Rhizopus stolonifer* (63.98mm), *Mucor* spp. (64.26mm) and *Trichoderma viride* (64.69mm) as compared to control (66.33mm). Maximum (18.99%) mortality was caused by *Fusarium moniliforme* and minimum (5.42%) by *Mucor* spp.

Soil inoculation technique

A pot experiment was conducted in case house where soil infested with individual myco flora and healthy seeds were grown to observe per cent seed germination, per cent mortality

(1) (2) (3) (4) (5) (6) (7) (8) (9)

(1) *Curvularia lunata*
(2) *Fusarium moniliforme*
(3) *Alternaria alternata*
(4) *Aspergillus flavus*
(5) *Mucor* spp
(6) *Aspergillus niger*
(7) *Rhizopus Stolonifer*
(8) *Trichoderma viride*
(9) Control (healthy)

Plate 1: Effect of different seed borne myco flora on germination and seedling vigour of wheat tested by seed inoculation technique

Table 1: Effect of different seed borne myco flora on germination and per cent mortality of wheat tested by seedling symptom test

S. No.	Seed myco flora	Seed germination (%)	Mortality (%)
1	*Alternaria alternata*	76.85 (61.24)	17.85 (24.98)
2	*Aspergillus flavus*	48.33 (44.02)	18.55 (25.50)
3	*Aspergillus niger*	58.81 (62.58)	14.78 (22.60)
4	*Curvularia lunata*	78.14 (62.11)	16.52 (23.97)
5	*Fusarium moniliforme*	55.27 (48.0)	19.67 (26.31)
6	*Rhizopus stolonifer*	81.36 (64.47)	7.61 (16.00)
7	*Mucor* spp.	81.68 (64.65)	8.44 (16.86)
8	*Trichoderma viride*	83.11 (65.73)	6.23 (14.44)
9	Control	84.35 (66.73)	4.89 (12.77)
SEm ±	0.875	0.198	
CD at 5%	2.604	0.59	
CV%	2.53	1.69	

* The value in parentheses is angular transformed

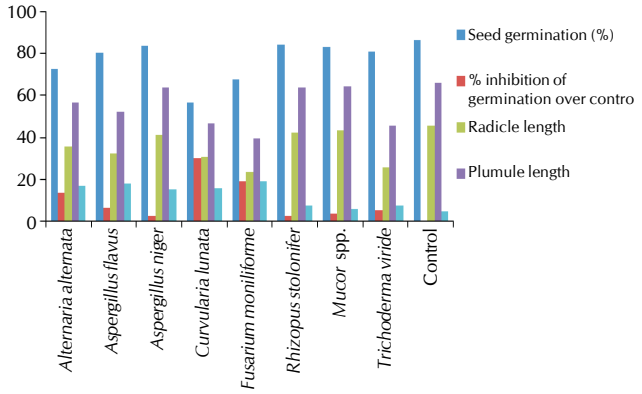

Figure 1: Effect of different seed borne myco flora on germination and seedling vigor of wheat seeds tested by seed inoculation technique

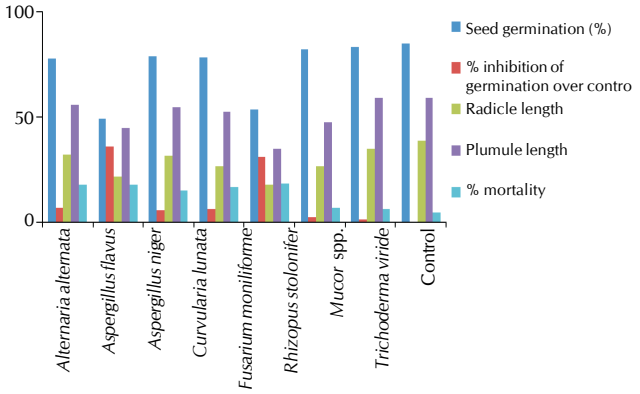

Figure 2: Effect of different seed borne myco flora on germination and seedling vigor of wheat seeds tested by soil inoculation technique

and seedling vigour. Data presented in Table 2 showed that, the fungi *Alternaria alternata, Aspergillus flavus, A. niger, Curvularia lunata, Fusarium moniliforme, Rhizopus stolonifer* and *Mucor* spp. inhibit the per cent germination, radicle and plumule length significantly as compared to control.

Results Presented in table 2 reveals that, maximum inhibition of germination was observed when soil was inoculated with *Aspergillus flavus,* (35.70mm) followed by

Fusarium moniliforme, (31.06mm), *Alternaria alternata* (6.82mm), *Curvularia lunata,* (6.03mm), *A. niger* (5.63mm), *Rhizopus stolonifer* (2.66mm) and *Mucor* spp. (1.14mm) as compared to control (uninoculated).

Maximum plumule length was also reduced when soil was inoculated with *Fusarium moniliforme,* (34.76mm) followed by *Aspergillus flavus,* (44.44mm), *Rhizopus stolonifer* (47.50mm), *Curvularia lunata,* (52.42mm), *A. niger,* (54.43mm), *Alternaria alternata,* (55.54mm) and *Mucor* spp. (58.63) as compared to control (58.63mm).

The maximum reduction in radicle length was observed when soil was inoculated with *Fusarium moniliforme* (17.65mm) followed by *Aspergillus flavus* (21.36mm), *Curvularia lunata* (26.51mm), *Rhizopus stolonifer* (26.62mm), *A. niger* (31.22mm), *Alternaria alternata* (31.72mm) and *Mucor* spp. (34.36mm) as compared to control (38.64mm).

Seedling symptom test

The seeds were infested with individual isolated seed borne myco flora and placed one seed in each sterilized test tubes on plain agar media to observe per cent seed germination and seedling symptoms. Results showed that the maximum reduction in seed germination was observed in seed inoculated with *Aspergillus flavus* (48.33%) followed by *Fusarium moniliforme* (55.27%), *A.s niger* (58.81%), *Alternaria alternata* (76.85%), *Curvularia lunata* (78.14%), *Rhizopus stolinifer* (81.36%), *mucor* spp. (81.68%) and *Trichoderma viride* (83.11%) as compared to control (84.35%).

The Seedling mortality was ranged from 6.23 to 19.67% comprising minimum (6.23%) caused by *Trichoderma viride* and maximum (19.67%) by *Fusarium moniliforme*. However, *Alternaria alternata* produced foliar symptoms initially yellowing of leaves and blight symptoms on the seedling while, *Fusarium moniliforme* was also produce initially yellowing of leaves foliar symptoms and wilting on wheat seedlings.

The seed inoculation and soil inoculation methods proved quite efficient in testing seedling vigour (length of radicle and plumule) of *Alternaria alternata, Aspergillus flavus, A. niger, Curvularia lunata, Fusarium moniliforme, Rhizopus stolonifer, Mucor* spp. Inoculated myco flora inhibits the seedling germination and also reduced the plumule and radical length as reported by different workers. Anjorin and Mohammed (2014) were found that wheat seed harbor several species of fungi, which can reduce seed quality and cause plant disease. Fungi carried on or within seeds reduce seed germination, seedling emergence lead to less vigorous seedling.

REFERENCES

Agarwal, V. K. and Sinclair, J. B. 1996. Principles of Pathology. 2nd edi, CRC Press, Inc., *Boca Raton.* **I:** 539.

Anjorin, S. T. and Mohammed, M. 2014. Effects of seed borne fungi on germination and seedling growth of watermelon (*Citrullus lanatus*). *J. Agriculture and Social Science.* **5:** 1813-2235.

Bateman, G. L. and Kwasna, H. 1999. Effects of number of winter wheat crops grown successively on fungal communities on wheat roots. *Applied Soil Ecology*. **13:** 271-82.

Bhoyar, P. R., Chandankar, V. D., Bagde, V., Borkar, D. B. and Sonone, J. S. 2014. Studies on seed borne mycoflora and effect of bioagents and fungicides on wheat seed health. *The Bioscan.* **9(3):** 1285-1289.

Glazek, M. 1997. Mycoflora of winter wheat seeds harvested from flooded commercial fields in South-Western Poland in 1997. Plant Protection Institute Poznaniu, Sosnicowice Branch, Gliwicka St. Sosnicowice, Poland. **29:** 44-153.

Halt, M. 1994. *Aspergillus flavus* and aflatoxin B1 in flour production. *Eur J Epidermiol.* **10(5):** 555-558.

Khanzada, K. A., Rajput, M. A., Shah, G. S., Lodhi, A. M., and Mehboob, F. 2002. Effect of seed dressing fungicides for the control of seed borne mycoflora of wheat. *A. J. Pl. Sci.* **1:** 441-444.

Khokhar, M. K. 2012. Estimation of deteriorative effect of *Fusarium oxysporum* and *Aspergillus niger* on fenugreek seed germination, seedling vigour and *in vitro* efficacy of fungicides. *Internat. J. Plant Protec.* **5(2):** 286-289.

Mirza, J. H. and Qureshi, M. S. A.1978. Fungi of Pakistan. Thesis (Un

pub.) Submitted to University of agri., Faisalabad, Pakistan.

Sharma, P. K. 2001. Micro organism associated with seed of pea (*Pisum sativum* L.) their pathogenic potential and disease management. Ph.D. (Ag.) Thesis, MPUAT, Udaipur.

Singh, J., Srivastava, S., Shikha, Sinha, A. and Bose, B.2011. Studies on seed mycoflora of Wheat (Triticum aestivum L.) treated with potassium nitrate and its effect on germination during storage. *Res J. Seed Science*. **4:**148-156.

Singh, S., Sinha, A., Kumar, S. and Yadav, S. M. 2014. Determination of seed germination percent and effect of trichoderma harzianum rafai on fresh and stored seed by different methods. *The Ecoscan*. **5:**181-185.