International Journal of Scientific Engineering and Applied Science (IJSEAS) - Volume-1, Issue-8,November 2015
ISSN: 2395-3470
www.ijseas.com

Bio-prospecting of some plants against seed-borne fungi of pulses.

Ashok S. Kandhare
K.M.C. College, Khopoli

Abstract:

Green gram, Black gram, Pigeon pea and chickpea are common pulses in diet rich in carbohydrates, proteins and minerals. Numerous fungi affect pulses adversely causing reduction in seed content and seed health. Total seventeen seed-borne fungi recorded from all four test pulses. Out of these seventeen seed-borne fungi, six were found to be common and dominant on all four test pulses. These seed-borne fungi cause adverse effect on yield and nutritive value of the pulses. In order to protect pulses from the pathological and economical damage lots of fungicides are used. Fungicides are harmful to the plant, environment and nutritive value of the pulses. Therefore; biological means like plant powders are tried during the study to curb the infestation of seed-borne fungi. Eighteen plants and three fungi that are more common and dominant on the four pulses studied. All plant powders were found to be effective on the selected fungi.

Key words: seed-borne fungi, pulses, plant powders, mycoflora

Introduction:

Pulses are the second most important group of food plants belonging to family Leguminosae. They form an important and indispensable part of daily diet. It is important source of dietary carbohydrates, proteins, essential amino acids and micronutrients such as calcium, phosphorus and iron. Therefore, pulses are important source of protein and essential amino acids for major vegetarians. Pulses like Green gram (*Vigna radiata* L.), Black gram (*Vigna mungo* L), Chickpea (*Cicer arietinum* L.) and Pigeon pea (*Cajanus cajan* L) etc are cultivated in Marathwada region of Maharashtra during Kharif and rabbi seasons, either as sole or intercrops, under rain fed or irrigated conditions.

Various seed-borne fungi affect pulses. Seventeen seed-borne fungi reported from all four test pulses i.e. Green gram, Black gram, Chickpea and Pigeon pea, of these six found to be common and dominant; these are *Aspergillus flavus, A. fumigatus, A. niger, Drechslera tetramera* and *Rhizopus stolonifer*. Out of six common and dominant seed-borne fungi three, *Aspergillus flavus, A. fumigatus* and *A. niger* are taken for present study.

Materials and methods:

i) Collection of plants and preparation of plant parts powder.

During present studies, eighteen plants available in the area were selected. The plants were identified from their morphological characters using 'Flora of Marathwada' (Naik, 1998). The collected plants were cut into different parts like stem, leaves and root. All parts were surface sterilized with 0.1% $HgCl_2$ and subsequently washed to remove disinfectant; with sterile distilled water. These plant parts were kept for drying in hot air oven at 60^{o}C for 48 hours.

The dried plant parts such as leaf, stem and root were crushed into powder with the help of grinder. The powders were passed through sieve to get fine powder. The powders of different plant parts were stored in polythene bags for the study.

ii) Evaluation of mycelial weight and sporulation.

Effect of different plant parts powder on dry mycelial weight and sporulation of common and dominant seed-borne fungi of pulses was studied. The seed-borne fungi were grown in liquid GN

medium (100 ml) supplemented separately with 5 g of leaf, stem and root powder of different plant parts in conical flasks for ten days at room temperature. After incubation, contents of the flasks were filtered through pre-weighted Whatman filter paper No.1. The filter papers with mycelial mat were oven dried for 24 hours at 60^{o}C and re-weighted. Growth of the seed-borne fungi in terms of dry mycelial weight was calculated by subtracting the initial weight of the filter paper from final weight of filter paper with mycelial mat. The common and dominant seed-borne fungi of pulses grown in GN medium without supplementation of plant powders served as control.

The sporulation of common and dominant seed-borne fungi of pulses was studied separately by collecting spore suspension of the respective fungi after passing the culture filtrate through muslin cloth. The spore suspension was placed on slide and sporulation was recorded by observing different microscopic fields under binocular microscope.

Results and discussion:

The results presented in the Table revealed that, all the test plant parts powder showed restrictive effect on growth of mycelium and sporulation of the fungus *Aspergillus flavus* in more or less degree. The most effective plants that caused maximum reduction in dry mycelial weight of the fungus were *Ocimum americanum* L. (leaf 20 mg), *Ocimum basilicum* L. (leaf 21 mg, stem 25 mg) and *Ocimum sanctum* L. (leaf 27 mg). The plants that caused minimum reduction of mycelial weight were *Adenanthera pavonia* L. (leaf 103 mg), and *Carum copticum* Benth & Hook. f. (103 mg), *Muntangia calabura* L. (root 102 mg) and *Samania saman* (root 102 mg).

Four plants such as *Azadirachta indica* A. Juss (stem), *Cyperus rotundus* L. (rhizome), *Ocimum basilicum* L. (leaf, stem) and *Ocimum americanum* L. (leaf and stem) caused low sporulation in the test fungus. Rest of the plants showed moderate to high sporulation.

The most effective plants that caused maximum reduction of dry mycelial weight of the test fungus *Aspergillus fumigatus* were *Ocimum sanctum* L. (root 18 mg and leaf 20 mg), *Azadirachta indica* A. Juss (leaf 20 mg and root 30 mg) and *Eucalyptus globulus* Labill. (root 30).

The plants that showed stimulatory activity on mycelial growth of the test fungus were *Samania saman* (Jacq.) Merr. (leaf 106 mg and root 100 mg), *Ruelia tuberosa* L. (stem 100 mg), *Murraya koinigii* (L.) Spreng. (root 97 mg) and *Samania saman* (Jacq.) Merr. (leaf 106 mg and root 100 mg).

Sporulation was least with plants *Azadirachta indica* A. Juss (leaf and stem) and *Ocimum sanctum* L. (leaf, stem and root). All parts of *Samania saman* (Jacq.) Merr. caused highest sporulation and rest of the plants showed moderate to high sporulation.

In case of *Aspergillus niger* the plants with maximum inhibitory effect on mycelial dry weight were *Ocimum americanum* L. (leaf 18 mg), *Azadirachta indica* A. Juss (stem 20 mg) and *Ocimum basilicum* L. (leaf 21 mg). Some plants showed stimulatory activity to the mycelial dry weight of fungus. They were *Croton tiglium* L. (leaf 132 mg), *Tagetis erecta* L. (root 102 mg) and *Adenanthera pavonia* L. (leaf 101 mg), and where as *Croton tiglium* L. (root 99 mg) and *Muntangia calabura* L. (stem 99 mg) failed to arrest growth of mycelium of the test fungus.

Sporulation was minimum in six plants such as *Azadirachta indica* A. Juss (stem), *Cyperus rotundus* L. (rhizome), *Eucalyptus globulus* Labill. (leaf), *Ocimum basilicum* L. (stem and root), *Ocimum americanum* L. (leaf, stem and root) and *Ocimum sanctum* L. (stem and root). With treatment of rest of the plants sporulation was moderate to high.

Discussion:

Among all tested plants *Ocimum americanum* L., *Ocimum basilicum* L., *Ocimum sanctum* L., *Azadirachta indica* A. Juss, *Cyperus rotundus* L., *Eucalyptus lanceolatus*, *Murraya koinigii* (L.) Spreng. were found to be effective in reducing seed mycoflora of the test pulses. These plant powders found to reduce dry mycelial weight and sporulation of the common and dominant seed-borne fungi of pulses. *Croton tiglium* L. and *Tagetis erecta* L. in case of *Aspergillus niger* and *Aspergillus flavus*, *Murraya koinigii* (L.) Spreng. and *Samania saman* (Jacq.) Merr. in case of *Aspergillus fumigatus* and *Rhizopus stolonifer*, *Tagetis erecta* L. in case of *Drechslera tetramera*, *Samania saman* (Jacq.) Merr. were found to be stimulatory to mycelial growth of respective fungi. The phytochemicals like quircitin, azadiractin, nimbidin, nibonin etc from *Azadirachta indica* A. Juss. , chavicol, methyl chavicol, linalool, eugenol etc from *Ocimum* species is effective in controlling the seed-borne fungi. These phytochemicals could be harnessed instead of artificial chemicals to control menace of seed-borne fungal infestation.

Similar results were reported by Khirsagar and Meheta (1972) where substances from three ferns i.e. *Adiantum trapiziforma, Aleuritopteris farinose* and *Pterris vittal* were shown to possess antimicrobial property. Bhargava *et al* (1981) tested oils of *Ocimum americanum* L. leaf against *Aspergillus* spp. and found it to be antifungal. Singh and Prasad (1993) found leaf extracts of *Azadirachta indica* A. Juss, *Ocimum sanctum* L. and *Ricinus communis* effective against *Helminthosporium speiciferus, Fusarium oxysporum* and *Aspergillus flavus* causing fruit rot of banana. Khan *et al* (1996) reported plant extracts of *Azadirachta indica* A. Juss, *Calatropis procera*, *Cuscuta reflexa, Euphorbia pulcherrima* and *Solanum nigrum* were effective against fungi of pulses and further these plant extracts were compared with synthetic fungicides. Aridogan *et al* (2002) studied essential oils from eight plants for their antimicrobial activity against *Staphylococcus aureus* and *Escherichia coli* and found that, these oils were effective in more or less quantity as antibacterial. Ahmed and Aquilf (2003) tested antimicrobial activity of *Delonix regia* flower extract and found that these extracts showed antimicrobial activity in different degrees. Rukhsana and Saima (2005) studied antifungal activity of aqueous leaf extracts of two species of *Eucalyptus* against three pathogenic fungi namely, *Alternaria alternata*, *Drechslera hawaiiensis* and *Drechslera tetramera*. All the plant extracts were found to be antifungal. Dhekle (2007) studied plant powders of *Cocculus hirsutus*, *Holarrhina antidysentrica*, *Merremia aegyptiaca*, *Merremia quinquefolia*, *Mucuna puriens*, *Plumbago zeylanica* and *Wrightia tinctoria* against some seed-borne fungi, these plants showed antifungal properties in more or less degree. Tare (2001) studied oils of *Acorus calamus* L., *Azadirachta indica* A. Juss, *Cedrus deodara*, *Eucalyptus* spp. *Pongamia glabra*, *Salvadora oleoides*, *Sesamum indicum* and *Trachyspermum* spp. and found that, these oils were lethal to different insects. Kuldeep and Shah (2012) reported antifungal activity of marigold, lat jeera, lemon grass, mehandi, oninon and neem aginst *Dreschslera bicolor*. Emad et. al.(2012) reported different solvent extracts of azadirachta indica A. Juss found to be effective against different fungi.Trivedi et.al. (2013) found neem oil, mustered oil and azadirchtin effective to control viral and leaf spot infections.

Sr. No.	Plants used	GN medium + 5 gm powder of	*Aspergillus flavus*		*Aspergillus fumigatus*		*Aspergillus niger*	
			Dry weight of mycelium (mg)	Sporulation	Dry weight of mycelium (mg)	Sporulation	Dry weight of mycelium (mg)	Sporulation
01	*Acorus calamus* L.	Leaf	100	+++	37	++	68	+++
		Rhizome	60	++	33	+++	80	+++
02	*Adenanthera pavonea* L.	Leaf	103	+++	32	++	101	+++
		Stem	89	+++	44	++	93	++
		Root	60	++	38	++	88	++
03	*Azadirachta indica* A. Juss.	Leaf	40	++	20	+	40	++
		Stem	35	+	40	+	20	+
		Root	55	++	30	++	38	++
04	*Butea monosperma* (Lam.) Taub.	Leaf	93	++	52	++	61	++
		Stem	92	++	60	++	40	++
		Root	100	++	51	++	80	++
05	*Carum copticum* Benth & Hook. f.	Leaf	91	++	52	++	68	+++
		Stem	103	+++	60	++	50	++
		Root	99	++	62	+++	53	++
06	*Ciba pentandra*	Leaf	55	++	37	++	35	++
		Stem	70	++	52	++	52	++
		Root	75	++	44	++	62	++
07	*Croton tiglium* L.	Leaf	35	++	63	+++	132	+++
		Stem	58	++	80	++	97	++
		Root	97	+++	70	++	99	++
08	*Cyperus rotundus* L.	Leaf	43	++	40	++	32	++

		Rhizome	29	+	70	+++	47	+
09	*Eucalyptus globulus* Labill.	Leaf	42	++	44	++	40	+
		Stem	50	+++	38	++	48	++
		Root	52	+++	30	+++	88	++
10	*Melingtonia hortensis*	Leaf	77	+++	66	++	55	+++
		Stem	82	++	59	++	70	+++
		Root	68	+++	70	++	81	+++
11	*Muntangia calabura* L.	Leaf	85	++	37	++	61	++
		Stem	97	++	42	++	99	+++
		Root	102	+++	31	++	82	++
12	*Murraya koinigii* (L.) Spreng.	Leaf	43	++	70	++	60	++
		Stem	70	++	66	++	68	++
		Root	98	+++	97	+++	80	++
13	*Ocimum basilicum* L.	Leaf	21	+	32	++	21	++
		Stem	25	+	36	++	28	+
		Root	32	++	42	++	38	+
14	*Ocimum americanum* L.	Leaf	20	+	60	++	18	+
		Stem	31	+	50	++	35	+
		Root	38	++	40	++	40	+
15	*Ocimum sanctum* L.	Leaf	27	++	20	+	33	++
		Stem	33	++	29	+	40	+
		Root	40	++	18	+	52	+
16	*Ruelia tuberose* L.	Leaf	99	++	37	++	90	+++

		Stem	92	+++	100	++	40	+++
		Root	100	+++	90	++	97	+++
17	*Samania saman* (Jacq.) Merr.	Leaf	70	+++	106	+++	52	++
		Stem	78	++	95	+++	70	+++
		Root	102	+++	100	+++	80	++
18	*Tagetis erecta* L.	Leaf	93	+++	40	+++	66	++
		Stem	68	++	80	++	88	++
		Root	95	++	70	++	102	+++
19	Control	GN medium	120	+++	95	+++	98	+++

- No sporulation
+ Low sporulation
++ Moderate sporulation
+++ High sporulation

References:

Kuldeep singh jadon and Rakesh Shah (2012). Antifungal activity of different plant extracts against Drechslera bicolour causing leaf blight of bell pepper. *Archieves of phytopathology and plant protection*. Online 18 April 2012.

Amit trivedi, Sharma S.K., Hussain (2013). Application of biodynamic preparation, bio-control agent and botanicals for organic management of virus and leaf spots of blackgram (vigna mungo L. Hepper). *Journal of agricultural research*, vol. (4), 60-64, url: http://academiapublishing.org/ajar/pdf/2013/April/recordnumber20143001867.

Emad M. El- Kholie, Mohammed A. T. Abdelreheem and Seham A. Khader (2012). *Azadirachta indica* A. Juss extracts influenced some pathogenic fungi. *African journal of microbiology reseacrh*, Vol. 6(27), pp 564-5649. url: http://www.academicjournals.org/AJMR.

Kshirsagar, M.K. and A.R. Mehta (1972). Survey of ferns in Gujrath state (India) for presence of antibacterial substances. *Planta med*. 22:386.

Bhargava, K.S., Dixit, S.N., Dubey, N.K. and R.D. Tripathi (1981). Effect of leaf extracts of *Datura stramonium* and *Rauvolfia serpentine* on the growth of some soil borne *Fusaria*. *Proce. 71st Indian sci. cong*. Part III Abst. Bot.: 38.

Singh, H.M.P. and M.M. Prasad (1993). Evaluation of medicinal plant extracts against Banana. *Proceeding Indian sci. Cong*. Part III Abst. Bot. 74.

Khan, M.I., Ashaq, M., Bhat, M.Y. and Fazal, M. (1996). A comparative study of the efficacy of some plant extracts and fungicides on mycoflora of Chick pea seeds. *Annals of plant protection sciences*. 4 (1): 85-87.

Aridogan, B.C., Daydar, H., Kaya, S., Demirici, M., Ozbasar, D. and Mumcu, E. (2002). Antibacterial activity and chemical composition of some essential oils. *Archives of Pharmaceutical research*, v. 25(6): 860-864.

Ahmad, I., Aqil, F. (2003). Broad spectrum antibacterial and antifungal activities and potency of crude alcoholic extract and fractions of *Delonix regia* flowers. *2nd world congress on "Biotechnological developments of herbal medicine" NBRI, Lucknow, UP, India:* 74, February 20-22.

Rukhsana Bajwa and Saima Iftikhar (2005). Antifungal activity of allelopathic plant extracts VI: In vitro control of fungal pathogens by aqueous leaf extracts of *Eucalyptus*. *Mycopath*. 3(1, 2): 7-12.

Dhekle, N.M. (2007). Antifungal activity of some medicinal plants against aflatoxin producing fungi. Ph.D. thesis, Swami Ramanand Tirth Marathwada University, Nanded (M.S.).

Tare, V. (2001). Bioactivity of some medicinal plants against chosen insect pest/ vectors. *Journal of medicinal and aromatic plant sciences*. 22 (4A): 120-124.

Naik (1998). *Flora of Marathwad*. Volume I and II, Amrut prakashan, Aurgangabd.