



Antifungal activity of leaf extract of selected plants on seed mycoflora and its effect on seed germination and seedling health of Green gram

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Abstract

Green gram (*Vigna radiata* (L.) Wilczek) is one of the important pulse crops in India. The seeds of green gram carry a number of fungi. A total 11 fungal species belonging to 9 genera were isolated on seeds of green gram. These fungi were found to be responsible for seed deterioration making them unfit for consumption. Subsequently, these seed-borne fungi inhibit seed germination, seedling emergence and cause diseases. To control these plant pathogenic fungi harmful chemicals are used. Using of such chemicals unsafe for environment. To control these fungi, pesticide compound of plant origin are more effective, safe for environment. Eight plants were selected to evaluate their antifungal activity against seed borne fungi. Leaf extracts of all selected plants showed inhibitory activity against above mentioned seed borne fungi and increased seed germination percentage, root and shoot length as compared to control. Leaf extract of *Azadirachta indica*. A.Juss. and *Annona squamosa* L. shown significant antifungal activity.

INTRODUCTION

India is the major producer of green gram in the world and grown in almost all the States. Green gram is an excellent source of high quality protein (25%) having high digestibility. It is consumed as whole grains as well as "Dal" in a variety of ways in our food. Sprouted green gram is used in the preparation of curry or a savory dish (South India). It is supposed to be easily digestible and hence the patients prefer it. It is also a good source of Riboflavin, Thiamine and Vitamin C (Ascorbic acid). Green gram is also used as green manure crop. It being a leguminous crop has capacity to fix the atmospheric nitrogen (30-40 kg N/ha). It also helps in preventing soil erosion. Being a short duration crop, it fits well in many intensive crop rotations. Green gram can be used as feed for cattle. Many diseases have been reported as seed borne.

Seedling blight, rot, stem rot, leaf and pod rot caused by *Macrophomina*, *Curvularia*, *Alternaria* are some of the major fungal diseases of green gram. *Alternaria*, *Cladosporium*, *Fusarium* and *Rhizoctonia* are known to cause seed rot and pre and post emergence losses in green gram (Khare et al., 1990). Several workers reported *Alternaria* sp., *Aspergillus flavus*, *Aspergillus niger*, *Cladosporium* sp., *Curvularia lunata*, *Fusarium* sp., *Macrophomina phaseolina*, *Phoma medicaginis* and *Rhizopus* sp. are seed borne and seed transmissible (Raut and Ahire 1988, Patil et al, 1990). These fungi are potentially harmful for cultivation of green gram. Several factors are responsible for low production of Green gram. Among them, diseases play an important role (Nene 1980; Pal 1996). Many fungal pathogens are were found to be associated with seed at the stage of

harvest, transport and during storage, often reduce the germination ability or kill the infected plants or substantially reduce the productive capacity. Various scientist studied seed mycoflora of Green gram and other pulses. *Aspergillus flavus* showed maximum percent incidence followed by other. They also noted that, *Aspergillus flavus* was found to be more bio-deteriorating than the others. To control these plant pathogenic fungi harmful chemicals are used. Using of such chemicals unsafe for environment. The pesticide compounds of plant origin are more effective, safe for environment and no side effect on human hygiene (Jalandhar, 2011). Medicinal plants are good reservoirs of chemotherapeutants (Farooqui et.al.1998). Considering the importance of plant origin compounds to control the seed borne fungi present investigation was under taken to evaluate the leaf extract of selected medicinal plants for their antifungal activity against seed borne fungi of green gram.

Materials and Methods

Collection of plant material: *Azadirachta indica*. *A.Juss*, *Acacia nilotica* L., *Ocimum basilicum* L. *Annona squamosa* L., *Abrus precatorius* L. *Polyalthia longifolia* L., *Datura alba* L. and *Vernonia amygdalina* L. plants were selected. These are common and easily available plants in the vicinity. Their identification was confirmed using the 'Flora of Marathwada' (Naik, 1998) the fresh and healthy leaves of these selected plants was collected separately. The leaves of these plants were washed thoroughly with tap water, surface sterilized with 0.1% HgCl₂ and washed repeatedly with sterile distilled water for several times and kept in shade for drying. After drying, they were preserved separately in polythene bags at room temperature (27± 2°C) during the studies.

Preparation of plant leaf extracts: The dried leaves of selected plants were crushed separately in to fine powder with the help of blender. 10 gm powder of each of the plant leaves powder was dissolved separately in 100 ml sterilized hot distilled water in 250 ml pre sterilized conical flasks. Then extract were filtered with muslin cloth

and was heated to 55⁰ C in water bath for 15 minutes. Flasks were kept in oven for 24 hours at 4°C and the content was again filtered through Whatman's filter paper No.1. The filtrates were used as 5% plant leaf extracts.

Assessment of seed Mycoflora and seed treatment: Seed sample of green gram were obtained from local farmers and stored at room temperature 27⁰C. ISTA'S Standard blotter paper and agar plate method was used to isolates seed borne fungi (Neergaard,1979). Hundred seeds of from sample were soaked in aqueous leaf extracts (5%) of selected plants and keep overnight. The seeds soaked in sterile distilled water served as control. The treated and control seed were keep in shady place for drying. These dried seeds were plated on agar plates, incubated for seven days at room temperature and their seed mycoflora was detected, identified (Barnett, 1992) and percentage of seed germination, root length and shoot length are recorded.

Result and Discussion

The result from agar plat method revealed the presence of 11 fungi belongs to 9 genera on seed sample of Green gram. These seed borne fungi were isolated and identified. Three species belongs to *Aspergillus* genera- *A. niger*, *A.flavus*, *A.fumigatus*., *Curvularia lunata*, *Fusarium oxysporum*, *Fusarium moniliforme*, *Rhizopus stolonifer*, *Alternaria alternate*, *Rhizoctonia soloni*, *Penicillum sp.* *Cladosporium sp.* Incidence *Aspergillus sp.* is more, this is also reported by Kandhare 2014. The seed of green gram associated with nine different fungi viz., *M.phaseolina*, *A. alternata*, *C. capsici*, *N. sphaerica*, *Chaetomium sp.*, *Aspergillus sp.*, *R. oryzae*, (Deshmukh et. al.,2016) Association of these seed borne fungi damage seeds in various ways. The quality of seed gets deteriorated and some time the grains are not suitable for animal consumption due to the production of mycotoxic substance by the seed fungi accompanied by change in the chemical nature of the seed. (Bhikane, 1988) The germination of seed is also appreciably affected by fungi. Moisture content of seeds has been

considered to be the next important factor for establishment of seed mycoflora. Safflower seeds having 13 % moisture favored sufficiently to develop species of *Aspergillus*, *Penicillium*, *Curvularia*, *Botrytis*, *Alternaria*, and *Cercospora* (Deokar, 1992). Patil *et al*, (1990) reported eighteen species of fungi belonging 14 genera on Green gram, of which 8 fungi *Alternaria alternata*, *Aspergillus fumigatus*, *A. niger*, *Curvularia geniculation*, *Fusarium moniliforme*, *F. oxysporum*, *Macrophomina phaseolina* and *Rhizoctonia bataticola* were pathogenic. Kandhare (2014) reported that sixteen fungi are associated seeds of green gram. Out of sixteen seed-borne fungi six were dominant; these are *Aspergillus flavus*, *A. fumigatus*, *A. niger*, *Drechslera tetramera*, *Fusarium moniliforme* and *Rhizopus stolonifer*. Attempts have been made on the bio-control of seed-borne fungi using plant extracts. The results obtained during the present investigations gave an idea that the plant leaf extracts can successfully control the spore germination, growth of seed-borne fungi like *Alternaria alternata*, *Aspergillus flavus*, *Curvularia lunata*, and *Fusarium oxysporum*. Plant leaf extracts also prevent percentage incidence of seed borne fungi and also can stimulate seed germination seedling emergence. All selected plants leaf extract showed good inhibitory activity against above mentioned seed borne fungi and increased seed germination percentage, root and shoot length as compared to control. *Azadirachta indica*, *A.Juss* and *Annona squamosa L.* extracts were showed good inhibitory activity against seed borne fungi 75 % of seed germination, 4.65 cm root length, 3.75 cm shoot length and 74% seed germination, 4.25 cm root length, 3.65 cm shoot length respectively. Followed by these plants *Polyalthia longifolia L.* showed good inhibitory effect against seed borne fungi they showed 57 % of seed germination, 3.79 cm root length, 2.96 cm shoot length and 63% seed germination, Similar results were observed in case of different crop plants by different workers. Rathore (1994) reported hot water, cold water and ethanolic leaf extracts of *Andrographis paniculata*, *Artabotrys hexapetalus*. He also found that the Garlic and *Artabotrys* extracts were very much effective in preventing the growth and sporulations of the test fungi. Jalandhar V. and M.Mamatha

<https://jbsd.in/edata>

(2017) studied the antifungal effect of weeds leaf extracts on seed borne fungi of Pigeon pea seed regarding seed germination percentage seedling length and vigor index. They have been reported that leaf extract of some weeds gave good result against seed borne fungi of Pigeonpea. The results obtained during the present investigations gave an idea that the leaf extract of *Azadirachta indica*. *A.Juss.* and *Annona squamosa L.* have shown significant antifungal activity which often leads to improved seed germination percentage and seedling health of test plant.

Table 1. Effect of leaf extract of selected plants on seed germination of Green gram

Sr. No.	Name of plants	% of germination	Root length in cm	Shoot length in cm
1	<i>Acacia nilotica L.</i>	49.10	3.25	2.70
2	<i>Abrus precatorius L.</i>	48.00	3.35	2.73
3	<i>Azadirachta indica A.Juss</i>	75.00	4.65	3.75
4	<i>Ocimum basilicum L.</i>	55.23	4.15	3.07
5	<i>Vernonia amygdalina L.</i>	51.09	3.09	2.87
6	<i>Datura alba L.</i>	44.00	3.01	2.90
7	<i>Annona squamosa L.</i>	74.00	4.25	3.65
8	<i>Polyalthia longifolia L.</i>	57.00	3.79	2.96
9	Control	45.00	2.77	2.01

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