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Seed Mycoflora of *Lablab purpureus* L.

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A B S T R A C T

In the present study attempts have been made on the isolation and identification of fungi in order to study the mycoflora responsible to reduce the seed quality, longevity and vigor in the pulse of Hyacinth bean. *Lablab purpureus L*. is an annual plant that grows in poorly - drained soils belongs to the family Fabaceae. It contains high amount of protein, vitamins and minerals. Fungal cultures were isolated by using blotter and agar plate methods.22 fungal species and 14 genera were isolated. *Aspergillus spp, Alternaria spp, Curvularia spp, Fusarium spp, Rhizopus spp, Mucor spp* were dominant species. These species were found that they cause considerable amount of loss, spoilage of storage grains and also produce mycotoxins.

Introduction

Seeds of legumes or pulses form an important source of dietary proteins they provide essential amino acids to our predominantly vegetarian population (Mohd shaker *et al.*, 2010). Legumes also relatively rich in two essential micronutrients namely iron and calcium. Increasing population pressure, fast depletion of natural resources, poverty and low agricultural production are some of the problems faced by the developing countries. It is well documented that the developing countries do not produce enough food and of the right nutritional quality to meet daily needs (Kamatchi Kala *et al.*, 2010). To prevalent food shortage, attention is currently being focused on the exploitation of lesser known and non-traditional plant resources (Bresssani, 1975). Exploitation of under-utilized wild legumes is an important approach to combat the protein- malnutrition in developing countries. Food legumes source of protein, carbohydrates, minerals and vitamins. Being rich in protein, carbohydrates, calorific value, fiber, and vitamins, legumes constitute staple food in many countries.

*Lablab purpureus*is grown as a pulse crop in Africa, Asia and the Caribbean. It is also consumed as a green vegetable (Maas *et al.*, 2010). It is may suffer from low yields when grown as a main cash crop, and suggest that it

is more popular in home gardens and mixed-cropping schemes. Seeds contain 20to 28% crude protein (Cooke *et al.,* 2005) and contain large amounts of various vitamins and minerals.

Protein isolate from the bean can be used as a food additive. It has medicinal properties used in the Philippines and China as a stimulant, reduces flatulence, stimulates digestion and used as an anti-spamodic (Stuart, 2011). In Namibia, the roots have been used to treat heart conditions (Pennacchio *et al.,* 2010). It is used as a nitrogen- fixing green manure to improve soil quality.

The crops have been reported to suffer from various types of disease and majority of them are known to be caused by fungi which are seed- born in nature. Several seed- born fungi cause considerable loss in the seed content, among them degradation of starch, lipids, proteins in legumes due to seed-borne fungi which resulted in decreased quality of the seeds.

In the present investigation an attempt has been made to study the Isolation and identification of seed- borne mycoflora of *Lablab purpureus* seeds during storage condition.

Materials and Methods

Seed samples: Seed samples collected from the field and market places from the Indurthi village, Kharimnagar district, Telangan, India. Seed samples from different sources of the pulse crop was stored individually preserved in cloth bags at room temperature during the studies.

Detection of seed mycoflora

Isolation of seed mycoflora was done by blotter method De Tempe, (1953) and agar plate method by Muskett, (1948).

Standard blotter paper method

This is very simple, most convenient and efficient of all the incubation methods. Doyer (1938), De Tempe was first to adopt blotter paper method in seed health management. Three layers of sterile white blotter papers of 8.5cm diameter were soaked in sterile distilled water and were placed in pre-sterilized Petri plates of 9o mm diameter.

Ten seeds of test sample per Petri plates were then placed at equal distance on moist blotter. 100 seeds were used in each experiment. The plates were incubated at $28^0 \pm 2^0$C under diurnal conditions. On seventh day of incubation, seeds were first examined under electron microscope for determining the various fungal growths. The identification and further confirmation of seed borne fungi was made by preparing slides of fungi (Rathod *et al.,* 2012).

Agar plate method

In Northern Ireland, Musket (1948) first used this method for seed health management. In this method, pre-sterilized Petri plates were poured with15ml of autoclaved Potato Dextrose Agar (PDA). On cooling the medium, ten seeds per plate of the sample to be studied were equidistantly placed aseptically. Incubation and other details of the study were same as described for blotter paper method.

Results and Discussion

Contaminated seeds can often result in poor germination and poor seedling vigor, resulting in an un-healthy crop. Healthy seed is the foundation of healthy plant a necessary condition for good yields. Field fungus associated with cause deterioration of seed quality, affects viability and reduces germination (Neeti saxena *et al.,* 2015; Tripati Agarwal *et al.,* 2011).

Twenty two fungal species, 14 genera were identified in two varieties of *Lablab purpureus* seeds, such as *Alternaria alternata, Aspergillus niger, Aspergillus flavus, Aspergillus fumigatus, Aspergillus nidulans, Aspergillus terreus, Aspergillus tenuis, Cladosporium spp, Curvularia lunata, Colletotrichum dematium, Cephalosporium acromonium, Fusarium oxysporum, Fusarium moniliformae, Macrophomin aphaseolina, Mucor racemosus, Mucorindicus, Nigrospora oryzae, Pencillium citrinum, Phomabetae, Rhizoctonia solani, Rhizopus nigricans, Rhizopus stolonifera* (Table 1). Most of the storage fungi isolated from the market variety than the Field variety. The difference is may be due to the age of the seed, seed type, and with the individual seed in the seed lot (Neetisaxena *et al.*, 2015). Storage fungi were present in low percentage in freshly harvest seed samples and became dominant as the storage period increased. Storage fungi require high osmatic pressure and no water (Manoharachary and Kunwar, 2006).

Table.1 Fungi identified from field and market seed varieties

S. No	Name of the Fungus	Field variety		Market variety	
		BP.M	AGP.M	BP.M	AGP.M
1.	*Alternariaalternate*	+	+	+	+
2.	*Aspergillus niger*	+	+	+	+
3.	*Aspergillus flavus*	+	+	+	+
4.	*Aspergillus fumigatus*	+	+	+	+
5.	*Aspergillus nidulans*	+	+	+	+
6.	*Aspergillus terreus*	+	+	+	+
7.	*Aspergillus tenuis*	+	+	+	+
8.	*Cladosporiumspp*	-	+	-	+
9.	*Curvularialunata*	-	+	-	+
10.	*Colletotrichumdematium*	-	+	-	+
11.	*Cephalosporiumacromonium*	-	+	+	+
12	*Fusarium oxysporum*	+	+	+	+
13.	*Fusarium moniliformae*	+	+	+	+
14.	*Macrophominaphaseolina*	+	+	+	+
15.	*Mucorracemosus*	+	+	+	+
16.	*Mucorindicus*	-	+	+	+
17.	*Nigrosporaoryzae*	-	+	+	+
18.	*Pencilliumcitrinum*	-	+	-	+
19.	*Phomabetae*	+	+	+	+
20.	*Rhizoctoniasolani*	-	+	+	+
21.	*Rhizopusnigricans*	+	+	+	+
22.	*Rhizopusstolonifera*	+	+	+	+

BP.M- Blotter paper method; AGP.M- Agarplate method

The present study has demonstrated that seeds of *Lablab purpureus* frequently carry a number of pathogenic fungi which can cause serious diseases in the field. Storage conditions in most parts of India are very conducive for mold invasion, proliferation and elaboration of mycotoxins (Girish and Goyal, 1986). *Alternaria, Aspergillus, Cheatomium, Cladosporium, Curvularia* associated with the seeds and these are

important seed borne fungi, Isolated from the both methods. The same results observed by Deo and Guptha, 2003; Mohdshekhar *et al.*, 2010). Seeds were adversely affected by the species of *Aspergillus, Alternaria, and Fusarium* species. Reddy, 1982 reported in many pulses. *Rhizopus, Cephalosporium spp, Pencillium* were observed in both methods (Jain and Patel *et al.*, 2004). *Fusarium* and *Macrophomina* were predominant in pulses. Chilkurind Giri, 2014; Kandare Ashok Sadhu (2014) observed and isolated them on the seeds of green gram.

More number of fungi were recovered in the agar plate than blotter method, this gives an idea that nutrients from the media might play an important role in initiation of growth of fungi on pulses also the media composition was more favorable than blotter method

In conclusion the fungi which were isolated from the blotter plate and agar plate method are seed- borne and cause deleterious effects on the seeds of stored grains. Both storage fungi and certain molds were observed. Which plays and important role in to reduce viability, longevity, quality of the seeds of both Field variety and Market variety of Indurthi village, Kharimnagar, Telangana, India.

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