

ASSESSMENT OF GROUND NUT SEED MYCOFLORA**S. S. Ingle**

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sudhakaringle2012@gmail.com**Abstract**

The most common cause of spoiled ground nut seeds in storage is to due the growth of fungi. These fungi are responsible to cause seed borne diseases and produces mycotoxic substances. Seed samples of two varieties of ground nut were collected from local farmers to assess the mycoflora. Total thirteen species belonging to six genera of fungi were detected from sample in standard blotter test, Agar plate method and Seed washing test on both varieties. Aspergillus species of fungi were dominant.

Key words: Ground nut seeds, Mycotoxin, Aspergillus, Seed borne fungi.

Introduction

Ground nut (*Arachis hypogea* L.) is one of the most important and widely cultivated oil seed crop. The seeds of ground nut have been found to carry a number of fungi. The most common cause of spoiled ground nut seeds in storage is to due the growth of fungi. Storage fungi infect the seeds as the seeds is transferred into storage and can spread rapidly throughout the bulk under favorable conditions. These fungi grow on the ground nut seeds; they become visible and can kill the seed, and produce undesirable odour, taste and some time the seeds are not suitable for human consumption due to the production of mycotoxic substance by the seed fungi accompanied by change in the chemical nature of the seed. Seed deterioration due to various mycoflora is common feature leading to loss of viability and numerous fungi develop on stored seed (Lalithakumari *et al.* 1970) Singh (1988) reported that fungi have been a major cause of spoilage and many of them have been found to bring about biochemical deterioration. The germination of seed is also appreciably affected by certain fungi. (Mishra and Kanaujia (1973) Groundnut seeds infested with *Aspergillus flavus* showed great loss in protein resulting it into increase in free amino acids. Reduction in protein content of oil seeds was found to be highest due to *Fusarium oxysporum* followed by *Fusarium moniliforme*. (Bilgrami *et. al.* (1976). Present investigation was undertaking to find out the mycoflora associated with seeds of groundnut.

MATERIAL AND METHODS:

Seed samples of ground nut var.TLG -45 and LGN-1 were collected from farmers immediately after harvest to assess the seed mycoflora. Select four hundred seeds from each sample were taken to study and the seed-borne fungi of ground nut seeds were detected by agar plate and blotter test methods as recommended by International Seed Testing Association (1966), de Tempe (1953), Neergaard (1973) and Agarwal (1976). The fungi were identified by using manuals of Barnett (1955), and Subramanian (1983). The procedure of agar plate and blotter test methods is described as below. Three methods were used for isolation of externally and internally seed-borne fungi.

Seed washing test:

One hundred seeds were taken in flask which contained sufficient water for their soaking. These seeds were first shaken over a mechanical shaker for 5 to 10 minutes. The sediment thus obtained was examined under a compound microscope for the identification of fungi.

Ten seeds of each variety were washed in 20ml of sterile water and 1ml of seed washings obtained thus was plated on GNA. Fungi developed within 3 days were immediately transferred to PDA and /or GNA slants for further study.

Standard blotter test:

Seeds were equidistantly spaced on moist sterile blotters in petriplates moist chambers. 10 petriplates of 9" diameter each containing 10 seeds were incubated at 27±2°C for eight days. Under alternative cycles of 12/12hrs. of natural light and darkness. Observations were made for fungi appearing on seeds every 24 hours and growth was carefully transferred to PDA slants for further

studies. A minimum of 400 seeds were observed in each case. Untreated seeds were used for mapping external seed mycoflora whereas seeds were surface disinfected by treating with 0.01% mercuric chloride solution for 10 minutes, where used for internally seed mycoflora.

Agar plating:

Seeds were equidistantly plated on GNA plates aseptically. Colonies which developed during three days were picked up and maintained on PDA/GNA slants. Untreated seeds were used for mapping external seed mycoflora whereas seeds were surface disinfected by treating with 0.01% mercuric chlorite solution for 10 minutes were used for internally seed mycoflora.

Media: PDA and GNA media were used in this study.

Potato dextrose agar (PDA):

Potato slices, 200 g; Dextrose 20g; Agar 20g; Tap water 1000 ml. pH 5.5.

Glucose nitrate agar (GNA)

Glucose 10g; KNO₃ 2.5g; KH₂PO₄ 1g; MgSO₄ 0.5g; Agar 20g and Distilled water 1000 ml.

RESULT AND DISCUSSION

Seed borne fungi on ground nut var.TLG-45 and LGN-1 was assessed and presented in given Table .The fungi produced various colonies and pure cultures of individual fungi were obtained, each fungus was grown separately on PDA medium and its growth and sporulation were recorded. Comparing morphology of hyphae, conidiophores and conidia with standard manuals generic and specific names were determined.

Table: Percent of Incidence of fungi on two varieties of ground nut.

Name of the Fungus	TLG -45					LGN-1				
	SBT (% incidence)		APM (% incidence)		SWT (% incidence)	SBT (% incidence)		APM (% incidence)		SWT (% incidence)
	Ex.	In.	Ex.	In.		Ex.	In.	Ex.	In.	
<i>Aspergillus niger</i>	35	23	38	29	42	32	20	27	16	34
<i>Aspergillus flavus</i>	30	20	34	25	37	27	17	23	10	28
<i>Aspergillus terreus</i>	10	05	05	03	02	08	04	06	03	06
<i>Aspergillus fumigatus</i>	12	04	06	04	05	06	03	01	01	03
<i>Aspergillus nidulans</i>	10	04	05	03	03	--	-	01	-	04
<i>Aspergillus versicolor</i>	07	03	03	-	01	--	-	-	-	04
<i>Aspergillus wentii</i>	08	02	01	02	01	01	-	01	-	-
<i>Aspergillus humicola</i>	05	02	-	01	--	02	-	-	-	03
<i>Rhizopus stolonifer</i>	25	-	-	-	27	18	-	15	-	25
<i>Curvalaria lunata</i>	-	10	07	03	17	10	06	04	01	13
<i>Penicillium sp.</i>	03	02	04	02	03	02	-	03	-	06
<i>Fusarium moniliform</i>	05	01	03	01	02	01	-	01	-	02

<i>me</i>										
<i>Mucor sp.</i>	12	-	-	-	18	10	-	08	-	14
		-	-	-			-		-	

SBT- standard blotter test, APM- Agar plate method, SWT- Seed washing test Ex. External In.- Internal.

Total thirteen species belongs to six genera of fungi were detected from sample in standard blotter test (SBT), Agar plate method (APM) and Seed washing test (SWT) on both varieties. *Aspergillus* species of fungi were dominant (8) *Aspergillus niger*, *A. flavus* *A. terreus* *A. fumigatus* were detected from all test in both varieties. Percent of incidence is highest in seed washing test as compared to standard blotter test and agar plate method. *A. nidulans*, *A. versicolor* *A. wentii* *A. humicola* were detected in all test in var. TLG-45 where as rare % of incidence of these species in all test in var. LGN-1. *Rhizopus stolonifer* and *Mucor sp.* fungi were detected from sample in standard blotter test (SBT), Agar plate method (APM) and Seed washing test (SWT) on both varieties. % of incidence of *Penicillium*, *Fusarium* *Curvularia sp.* found in TLG-45 variety. From these results it is evident that seeds contaminated with these fungi responsible to cause diseases and mycotoxic substance produces by the seed fungi. Gupta and Chohan (1970) reported that *Aspergillus niger*, *A. flavus*, *Macrophomina phaseolina* and *Rhizopus arrhizus* were responsible for seed rot during germination and *Aspergillus niger*, *A. flavus* and *Macrophomina phaseolina* were also seedling invaders. Cherry *et al.* (1975) reported that, ground nut seeds infested with *Aspergillus flavus* showed great loss in protein resulting it into increase in free amino acids. Dominant saprophytic fungi were detected on JL-24 variety of ground nut these fungi play an important role in determining the seed health. (Deepali Sable *et al.*, 2008). Similar type of observation are made by Deshmukh and Kare (2010) on Niger, Safflower and soyabean seeds.

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