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Studies on Storage Seed Mycoflora of Groundnut (*Arachis hypogaea* L.)

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ABSTRACT

Present investigation was undertaken to study effect of different storage periods and different storage containers on incidence of seed mycoflora of two different varieties of Groundnut viz. TAG-24 and SB-XI. In both the varieties seeds stored for the period of 180 days showed the presence of more number of fungi (10) with maximum percent incidence while seeds stored for the period of 120 days and 300 days showed the presence of less number of fungi with poor percent incidence as compared with seeds stored for the other test storage periods. Seed borne fungi like Aspergillus candidus, Aspergillus flavus, Aspergillus fumigates, Aspergillus niger, Aspergillus terreus, and Rhizopus nigricans were found to be more common and dominant on the seeds stored for all the test storage periods but their percent incidence was different for different storage periods. It is evident that in case of both the varieties of Groundnut, the seeds stored in gunny bags as well as in cloth bags showed highest counts of mycoflora while the seeds in polythene bags showed no effect in number of fungi but their percent incidence was very low. It is also evident that the variety SB-XI showed more resistance against mycoflora than TAG-24.

Key Words: Seed Mycoflora, Groundnut, TAG-24, SB-XI.

INTRODUCTION

Seed is a living biological unit. The moment it develops and reaches maturity on its mother plant, the aging or senescence starts and gains momentum over time and it is highly dependent on the environmental conditions. Seed storage is an imperative and inescapable for sensitivity of seed to environment, seasonal demand, dormancy, specificity of planting time, necessity of carry over and need of buffer seed stock (Salunkhe and Desai, 1984). In storage the viability and vigour of seeds not only vary from genera to genera and variety to variety, but also regulated by many physico-chemical factors like moisture content, atmospheric relative humidity, temperature, initial seed quality, physical and chemical composition of seeds, gaseous exchange, storage structure, packaging materials etc. (Doijode, 1988). As the seed is hygroscopic in

nature, seed quality is affected by variation in moisture content, relative humidity, temperature *etc*.

Groundnut is one of the poor storer. Storing seeds after harvest till the next cropping season without impairing the quality is of prime importance for successful seed production. The problem of loss of seed viability is more severe in Groundnut harvested in the *Summer* season and about 50 per cent viability could be lost within 4-5 months of storage in such produce (Nautiyal *et al.*, 1990; Nautiyal and Ravindra, 1996). Seeds with high oil content appear to lose their germination and vigour in a short time despite the precaution taken during harvesting and drying (Nautiyal *et al.*, 1990). High temperature and high relative humidity causes rapid deterioration of viability and vigour of Groundnut seeds..

It is clear from the literature that seeds during storage show increase in their mycoflora and its composition was found to be variable with storage conditions. During storage, oilseeds increase in their mycoflora and its components were found to be variable with condition of storage. Taking this into consideration the present investigation was undertaken to study effect of different storage periods and different storage containers on incidence of seed mycoflora of two different varieties of Groundnut viz. TAG-24 and SB-XI.

MATERIAL AND METHODS

i) Collection of seed samples

The method described by Paul Neergaard (1973) has been adopted for the collection of seed samples. Accordingly, three random samples of seeds (one Kg each) were collected from oil mills, market place and Oil Seed Research Station (ORS), Latur. Groundnut cultivars used in the present study are TAG-24 and SB-XI.

ii) Effect of storage period on seed mycoflora of Groundnut varieties TAG-24 and SB-XI

To study effect of storage period on seed mycoflora, the seeds of Groundnut variety TAG-24 and SB-XI were stored in gunny bags at room temperature for different storage periods (120, 180, 240 and 300 days). Seed mycoflora of the stored seeds was detected at the end of every test period by moist blotter plate method.

iii) Effect of storage containers on seed mycoflora of Groundnut varieties TAG-24 and SB-XI

To study effect of storage containers on seed mycoflora, the seeds of Groundnut variety TAG-24 and SB-XI were stored separately in air tight containers like Tin boxes and Polythene bags and non-air tight storage containers like Gunny bags and Cloth bags for period of 180 days at room temperature. Seed mycoflora of the stored seeds from the test containers was detected by moist blotter plate method.

iv) Identification of storage seed mycoflora of Groundnut

The identification was confirmed with the help of latest manuals, Subramanian, (1971), Neergaard and Mathur, (1980), Jha, (1993) and Mukadam, (1997) Pure culture of the identified fungi were prepared and maintained on PDA (Potato Dextrose Agar) slants.

RESULT & DISCUSSION

i) Effect of storage period on seed mycoflora of Groundnut (By Moist Blotter Plate Method)

To study the effect of storage period on seed mycoflora, the seeds of groundnut variety TAG-24 and SB-XI were stored for different storage periods (120, 180, 240 and 300 days) in gunny bags at room temperature. The seed mycoflora was studied after each and every test storage period by moist blotter test method. The results are presented in table-1.

It is evident from the data summarized in table-1 and 2 that total 10 fungi were isolated from the seeds stored for all the test periods.

In both the varieties the seeds stored for the period of 180 days showed maximum percent incidence of seed borne fungi while the seeds stored for the period of 120 days and 300 days showed least percent incidence of seed borne fungi as compared with the seeds stored for the other test storage periods.

The seed borne fungi like *Aspergillus candidus*, *Aspergillus flavus*, *Aspergillus fumigates*, *Aspergillus niger*, *Aspergillus terreus*, and *Rhizopus nigricans* were found to be more common and dominant on the seeds stored for all the test storage periods but their percent incidence was different for different storage periods.

Table 1: Effect of storage period on per cent incidence of mycoflora on Groundnut (var. TAG-24 and SB-XI) seeds stored in gunny bags for different storage periods by moist blotter plate method. (After 7 days of incubation)

| Sr. | Seed Mycoflora | Per cent Incidence of seed mycoflora | | | | | | | | |
|-----|---|--|-------------|-------------|-------------|---|-------------|-------------|-------------|--|
| No. | | Storage period in days for variety TAG-24 | | | | Storage period in days for variety SB-XI | | | | |
| | | 120 Days | 180 Days | 240 Days | 300 Days | 120 Days | 180 Days | 240 Days | 300 Days | |
| 1 | Alternaria alternata (Fr.) Keissler | 10 | 10 | 00 | 00 | 10 | 05 | 00 | 00 | |
| 2 | Aspergillus candidus Link. | 10 | 15 | 20 | 10 | 10 | 10 | 05 | 05 | |
| 3 | Aspergillus flavus Link ex Fr. | 20 | 40 | 60 | 65 | 15 | 35 | 50 | 60 | |
| 4 | Aspergillus fumigatus Fresenius | 10 | 10 | 15 | 15 | 10 | 10 | 10 | 15 | |
| 5 | Aspergillus niger van Tieghem | 40 | 60 | 60 | 65 | 30 | 50 | 50 | 60 | |
| 6 | Aspergillus terreus Thom. | 10 | 10 | 20 | 25 | 10 | 05 | 10 | 10 | |
| 7 | Fusarium oxysporum Schlechtend emend Sny. & Hans. | 00 | 20 | 10 | 10 | 00 | 10 | 05 | 05 | |
| 8 | Macrophomina phaseolina (Tassi) Goldanich | 00 | 10 | 10 | 00 | 00 | 05 | 00 | 00 | |
| 9 | Rhizopus nigricans Ehrenb. | 00 | 20 | 20 | 20 | 00 | 10 | 05 | 05 | |
| 10 | Sclerotium rolfsii Sacc. | 20 | 40 | 60 | 00 | 10 | 05 | 05 | 05 | |
| | S.E. <u>+</u> | 3.43 | 3.88 | 4.04 | 2.36 | 2.87 | 2.88 | 3.02 | 2.50 | |
| | C.D. at 0.05% | 10.17 | 11.52 | 12.01 | 6.99 | 8.51 | 8.55 | 8.96 | 7.41 | |

Table 2: Effect of storage containers on per cent incidence of mycoflora on Groundnut (var. TAG-24 and SB-XI) seeds stored for the period of 180 days in different storage containers by moist blotter plate method. (After 7 days of incubation)

| Sr. | Seed Mycoflora | Per cent Incidence of seed mycoflora of Groundnut | | | | | | | | |
|-----|---|---|-----------|---------------|-------|----------------|-------|---------------|-------|--|
| No. | | Air tight storage container Non-air tight storage container | | | | | | | | |
| | | Variety TAG-24 | | Variety SB-XI | | Variety TAG-24 | | Variety SB-XI | | |
| | | Tin box | Polythene | e Tin box | • | • | Cloth | Gunny | Cloth | |
| | | | bag | | e bag | Bag | Bag | Bag | Bag | |
| 1 | Alternaria alternata (Fr.) Keissler | 10 | 10 | 05 | 05 | 30 | 20 | 10 | 10 | |
| 2 | Aspergillus candidus Link | 10 | 00 | 05 | 00 | 20 | 10 | 10 | 05 | |
| 3 | Aspergillus flavus Link ex Fr. | 30 | 10 | 20 | 15 | 60 | 50 | 40 | 50 | |
| 4 | Aspergillus fumigatus Fresenius | 10 | 10 | 10 | 20 | 30 | 20 | 25 | 10 | |
| 5 | Aspergillus niger van Tieghem | 50 | 30 | 30 | 00 | 80 | 60 | 60 | 50 | |
| 6 | Aspergillus terreus Thom. | 10 | 10 | 05 | 00 | 30 | 20 | 20 | 15 | |
| 7 | Fusarium oxysporum Schlechtend emend Sny. & Hans. | 05 | 10 | 05 | 05 | 30 | 20 | 20 | 10 | |
| 8 | Macrophomina phaseolina (Tassi) Goldanich | 20 | 05 | 10 | 00 | 30 | 25 | 20 | 10 | |
| 9 | Penicillium sp. | 00 | 10 | 10 | 00 | 30 | 20 | 20 | 10 | |
| 10 | Rhizopus nigricans Ehrenb. | 10 | 30 | 30 | 05 | 80 | 70 | 60 | 50 | |
| | S.E. <u>+</u> | 3.86 | 4.00 | 2.87 | 2.13 | 3.84 | 4.80 | 4.50 | 4.48 | |
| | C.D. at 0.05% | 11.45 | 11.87 | 8.51 | 6.31 | 11.38 | 14.24 | 13.35 | 13.29 | |

Table-3: Microscopic characteristics used for the identification of Seed borne fungi of Groundnut.

| Sr. No. | Name of Fungi | Microscopic characteristics |
|---------|--|---|
| 1 | Alternaria alternata (Fr.) Keissler | The mycelium is profusely branched, brownish and septate. Conidiophores: Developed singly or in small groups, branched or unbranched. Conidia: In long chains (often branched), oval to ellipsoidal, with 2-7 transverse and 1-4 longitudinal or oblique septae, tapering end to form a short beak at the apex. The number of conidia in a chain varied from 2-8. |
| 2 | Aspergillus candidus Link. | Colony pale yellow, the fungus produces white as well as globose conidia-producing bodies which produce globose and sub-globose conidia. These conidia were smooth, thin-walled and revealed to be about $2.5-3.5\mu m$ in diameter. Vesicles spherical to sub-spherical which were entirely covered with metulae. |
| 3 | Aspergillus flavus Link ex Fr. | The mycelium is found to be submerged in the seed coat and forms a white to grey, tough mass. Conidiophores erect, simple, unbranched, hyaline, transparent and smooth. The apex of the conidiophores was inflated into a vesicle upon which radiating phialides are formed. Conidial heads were biseriate, globose to radiate often columnar, very light to deep yellow green, olive brown often brown. Conidia were found to be hyaline, single celled and produced in chains. They were globose to subglobose, often elliptical to pyriform and conspicuously echinulate. |
| 4 | Aspergillus fumigatus Fresenius | The mycelium produced blue-dull green colony on PDA medium, conidial heads being light green to dull blue green, vesicles uniseriate, pyriform, conidial head columnar, compact, densely crowded, Conidia globose to subglobose, green in mass 2-3 µm in diameter. |
| 5 | Aspergillus niger van Tieghem | Mycelium was found to be often scanty, hyaline to white or light yellow. Conidiophores were found to be developed directly from the seed coat They were hyaline to light brown, long, thin, unbranched, erect, brittle and terminating in to an inflated apex. Conidial heads appeared globose, but subsequently split into a few to several irregular or well-defined divergent columns of conidial chains. They were black, globose or radiate. Conidia found to be in chains on the sterigmata. They were single celled, pale to dark brown, more or less globose, with low to prominent ridges surfaces. |
| 6 | Aspergillus terreus Thom. | Mycelium found to be rapidly growing with variable colony appearance ranging from heavily sporulating colonies to fluffy, poorly sporulating colonies. The conidiophores were long, columnar, hyaline and smooth giving rise to subspherical biseriate vesicles. Conidia were found to be smooth walled, globose to slightly elliptical and striate. |
| 7 | Fusarium oxysporum Schlechtend emend Sny. & Hans. | The mycelium found to be white to light pink, aerial, unbranched or branched, very short monophialides bearing microconidia on false heads. The microconidia produced on micrconidiophores were abundant, hyaline, single celled, oval or elliptical. Macroconidia were found to be produced on the pale orange sporodochia. Macroconidia produced on macroconidiophores were hyaline, often 3 to 5 septate, falcate to almost straight, thin walled, with a curved apical cell and slightly foot-shaped basal cell. Chlamydospores were found to be terminal and intercalary, irregular in shape, thick walled with smooth surfaces. |
| 8 | Macrophomina phaseolina (Tassi) Goldanich | The mycelium found to be with thick hyphae. They were gray to brown or dark brown to black or dull white to light brown. Pycnidia were larger than the sclerotia, dark brown to black and scattered throughout the surface. They were found to be separate or confluent, rough, globose or irregular, beaked and ostiolate. Mature pycnidia were found to be dehisced and ooze conidia in a dull white, gelatinous mass. Conidia were aseptate, hyaline, ellipsoid to obovoid. Sclerotia were black, shiny, irregularly shaped. |

Table-3: Continued...

| Sr. No. | Name of Fungi | Microscopic characteristics |
|---------|----------------------------------|---|
| 9 | Rhizopus nigricans Ehrenb. | The colonies were found to be whitish, with aerial mycelium and black spots of sporangia and dark sporangiophores. Rhizoids were well developed. Sporangiophores (on stolons) were found to be brown, in groups of 1-3. Sporangia were blackish, powdery in appearance. Columellae were conical and mouse-grey. Sporangiospores angular-globose-ellipsoidal and distinctly striate. |
| 10 | Sclerotium rolfsii Sacc. | The fungus was found to be produced white, dense, radiating mycelial growth on potato dextrose agar medium. In the early stages, the fungus was found to be produced white mycelium and gradually lost its luster and became some what dull in appearance. Aerial hypae were not uniformly distributed. Initiation of sclerotial bodies were observed from fifth day onwards after inoculation. In the beginning, the sclerotial bodies were white which gradually turned to brwon colour and then to chocolate brown at maturity. The fully matured sclerotia were spherical, ellipsoidal. |

ii) Effect of air tight and non-air tight storage containers on seed mycoflora of Groundnut varieties TAG-24 and SB-XI (By Moist Blotter Plate Method)

Attempts were made to know the best storage container in order to get minimum incidence of mycoflora. For this, four types of containers namely Tin boxes, Polythene bags, Gunny bags and Cloth bags were used to store 1 Kg/container of seeds for a period of 180 days at room temperature. Then the seeds were plated on moist blotter and incubated for seven days at room temperature. The results obtained are summarized in table -2.

It was evident from the results that in case of both the Groundnut varieties TAG-24 & SB-XI, the seeds stored in gunny bags as well as in cloth bags showed highest counts of mycoflora while the seeds stored in polythene bags showed no effect in number of fungi but their percent incidence was very low.

It was also evident that the seeds of variety SB-XI showed more resistance against the mycoflora than the seeds of variety TAG-24.

iii) Identification of storage seed mycoflora of Groundnut

The fungi were identified by observing colony morphology and microscopic characteristic. The results are presented in table-3

CONCLUSION

In both the varieties the seeds stored for the period of 180 days showed the presence of more number of fungi (10) with maximum percent incidence while the seeds stored for the period of 120 days and 300 days showed the presence of less number of fungi with poor percent incidence as compared with the seeds stored for the other test storage periods.

The seed borne fungi like Aspergillus candidus, Aspergillus flavus, Aspergillus fumigates, Aspergillus niger, Aspergillus terreus, and Rhizopus nigricans were found to be more common and dominant on the seeds stored for all the test storage periods but their percent incidence was different for different storage periods.

It is clear from the results that in case of both the varieties of Groundnut (TAG-24 & SB-XI), the seeds stored in gunny bags as well as in cloth bags showed highest counts of mycoflora while the seeds in polythene bags showed no effect in number of fungi but their percent incidence was very low. It is also evident that the variety SB-XI showed more resistance against mycoflora than TAG-24.

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CONFLICT OF INTEREST

The author declares that there is no conflict of interest.

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