



JATROPHA SEED BORNE FUNGI IN THE HARYANA

^aPrabha Tiwari, ^aPooja Kannoja and ^bAmit Pandey

^aDepartment of Botany and Microbiology, H.N.B. Garhwal University, Srinagar, Uttarakhand, India. 246174.

^bForest Pathology Division, Forest Research Institute, Dehradun, Uttarakhand, India. 248006.

ABSTRACT

Seed mycoflorae adversely affect the quality of the seeds during storage resulting in low seedling vigour and oil yield. The present study aimed to identify fungi associated with seeds of *Jatropha curcas* in storage. The seed mycoflora of *Jatropha curcas* from different locations of Haryana were investigated using two different isolation techniques i.e. blotter paper and agar plate method. Total nine fungi were isolated and their occurrence percentage in the seeds was recorded. *Aspergillus* spp. were most frequently occurring fungi among all the isolated seed mycoflora. Occurrence of field fungi was found to decrease and the storage fungi percentage increased during storage.

KEY WORDS: *Jatropha curcas*, seed mycoflora and storage fungi.

INTRODUCTION

Due to diminishing petroleum reserves and the environmental consequences, biodiesel became an alternative energy fuel (Zurina, 2009). "Biodiesel" is well known chemically as the mono-alkyl esters of long-chain fatty acids. It is produced from less expensive feedstock containing fatty acids such as non-edible oils, animal fats, waste food oil and byproducts of the refining vegetables oils (Veljkovic *et al.*, 2006). In the situation of rapidly growing energy requirements, some non-edible oils have to play a significant role. The oil plant *Jatropha curcas* L. grows in tropical and subtropical climate across the developing world (Openshaw, 2000) & contains 20-40% oil.

Numerous examples exist in agriculture literature for the international spread of plant diseases as a result of the seeds infected or contaminated with pathogens (Agarwal and Sinclair, 1996). Seeds are regarded as important means for transporting plant pathogens over long distances. Seed fungal mycoflora are of considerable importance due to their influence on the overall health, germination, vigour, oil yield and final survival percentage of the plantations. A seed-borne pathogen may be present externally or internally or associated with the seeds as contaminant. Some of the seed-borne fungi were found to be very destructive, caused seed rot, and decreased seeds germination and also cause pre and post germination death (Bolkan *et al.*, 1976; Elarosi 1993) in different host species. Dharmaputra *et al.*, (2009) reported some irreversible degenerative changes in the quality of *Jatropha* seeds during storage, thus making the seed unfit for oil extraction, export purpose or sowing. Since farmers in Haryana are taking up the *Jatropha* plantation on large scale, the seed health becomes a prime concern and in the present study the seed mycoflora of *Jatropha curcas* from different locations of Haryana were investigated using two different isolation techniques i.e. blotter paper and agar plate method.

MATERIALS AND METHODS

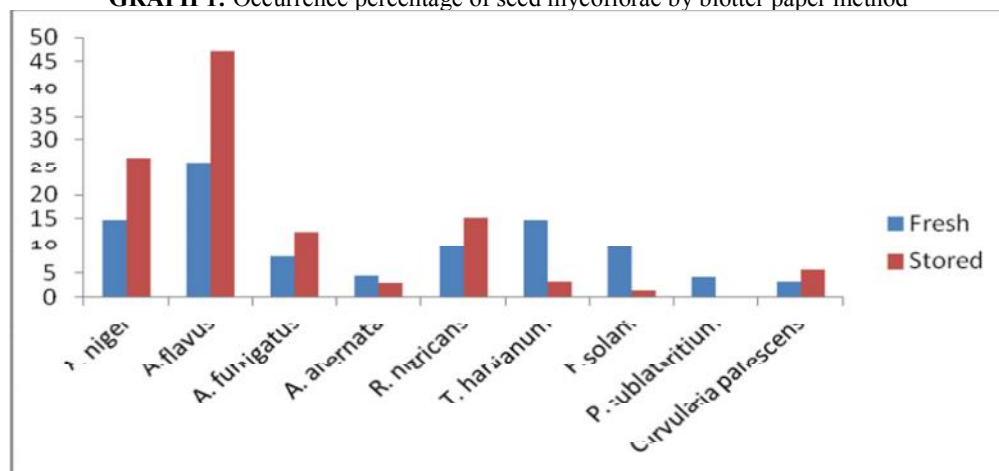
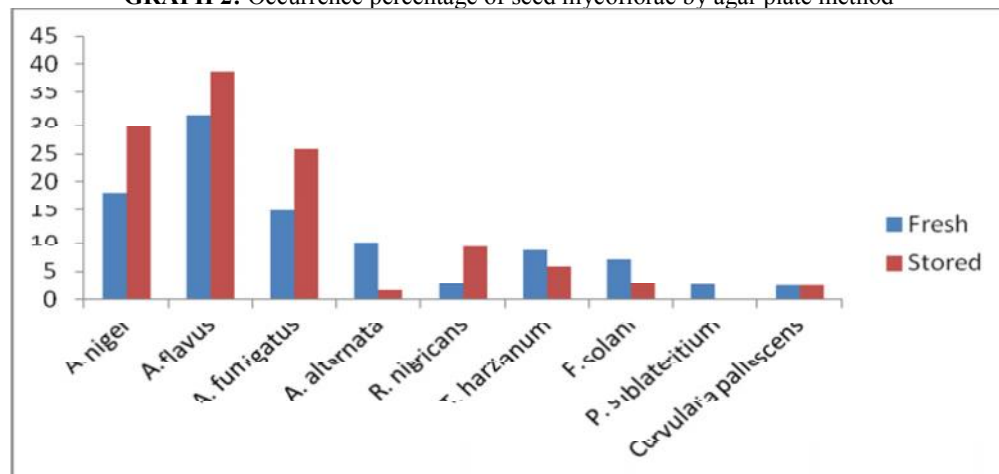
Seeds of *Jatropha curcas* were collected from field plantations and different local markets of Haryana. Two standard isolation procedures were employed for the isolation of pathogenic and saprophytic fungi *viz.* moist blotter and the potato dextrose agar (PDA) method. For determining seed mycoflora, seeds were surface sterilized with 0.1% mercuric chloride solution for one minute and then washed thoroughly with sterile distilled water and blotted dry between folds of sterilized blotter and placed on sterilized damp blotter paper. For potato dextrose agar method (PDA), seeds were surface sterilized as in the previous experiment and six seeds plated evenly on PDA plates. The culture plates were incubated at 25±2°C for 7 days. After the 7 days incubation period, conidia and hyphae of fungi growing on the seeds were picked off from the each infected seeds with fine forceps, mounted on a slide and examined with a compound microscope. Taxonomic identity of the isolated fungi was determined based on their morphological and microscopic characters using standard keys of Booth (1971), Ellis (1971) and Barnett and Hunter (1972).

RESULT AND DISCUSSION

The nine storage fungi recorded during the course of study were *Alternaria alternata*, *Aspergillus flavus*, *A. fumigatus*, *A. niger*, *Curvularia pallescens*, *Fusarium solani*, *Penicillium sublateritium*, *Rhizopus nigricans* and *Trichoderma harzianum*. The fungi detected from stored seeds were same as in fresh seeds except *Penicillium sublateritium* which was absent in stored seeds. There were variations in the occurrence of different fungal species in fresh and stored seeds (Table 1). In fresh seeds occurrence of *A. flavus* was highest in both, PDA (31.47) and blotter paper method (26.33). Lowest incidence of *Curvularia pallescens* i.e. 3.56 and 2.02 percent was recorded on blotter paper and agar plate method respectively.

TABLE 1: Percentage occurrence of mycoflorae in jatropha seeds in field and storage

S. No.	Fungi	Occurrence (%)			
		Fresh		Stored	
		Blotter Paper	Agar Plate	Blotter Paper	Agar Plate
1	<i>A. niger</i>	14.80	18.37	26.93	29.35
2	<i>A. flavus</i>	26.33	31.47	46.64	38.86
3	<i>A. fumigatus</i>	7.66	14.90	12.70	25.71
4	<i>A. alternata</i>	4.62	10.05	3.37	1.38
5	<i>R. nigricans</i>	10.42	2.37	15.1	8.7
6	<i>T. harzianum</i>	14.65	8.14	3.72	5.73
7	<i>F. solani</i>	9.45	6.83	1.16	2.49
8	<i>P. sublateralitium</i>	4.42	2.22	0.00	0.00
9	<i>Curvularia pallescens</i>	3.56	2.02	5.54	2.02

GRAPH 1: Occurrence percentage of seed mycoflorae by blotter paper method**GRAPH 2:** Occurrence percentage of seed mycoflorae by agar plate method

In stored seeds *A. flavus* was again found to be the most abundance fungus, with a contamination of 46.64 % and 38.86 % as detected by the moist blotter and agar plate method respectively. *A. niger* was the second most frequently detected species in both fresh and stored seeds. *Penicillium sublateralitium* was not found in stored seeds of *Jatropha curcas* in both the methods of isolation. This result was also supported by Srivastava *et al.*, (2011); Sahab *et al.*, (2011) and Singh *et al.*, (1996) that jatropha seeds are heavily deteriorated during storage, as they act as a source of stored nutrients for fungi such as *Aspergillus flavus*, *A. fumigatus*, *A. niger* and *Rhizopus*

nigricans. Jayaraman *et al.*, (2011) also isolated *Aspergillus flavus*, *A. fumigatus*, *A. niger*, *Penicillium* sp. and *Rhizopus* sp. from four samples of jatropha seeds from Chennai. Singh *et al.*, (1996) also reported fungal deterioration in jatropha seeds collected from central India which also decreased the oil yield and oil quality. It has also been found as mentioned in Table 1 that the occurrence percentage of some fungi such as *Alternaria alternata*, *Fusarium solani*, *Penicillium sublateralitium* and *Trichoderma harzianum* decreased in both the methods as compared to the fresh seeds during storage. These results are in consonance with the finding of Worang (2008);

Srivastava *et al.*, (2011) and Sahab *et al.*, (2011) who reported that the most of the field fungi found on the fresh seeds tend to decrease in their occurrence after three months of storage. The field fungi were generally replaced by storage fungi in *Jatropha curcas* seeds.

Hence, it could be suggested to arrest the fungal deterioration mainly by *Aspergillus flavus*, *A. fumigatus*, *A. niger* and *Rhizopus nigricans* to some extent by simple seed treatments with fungicides, botanicals and biological agents. This could solve the problem of fungal deterioration of seeds resulting in improved oil quality and productivity.

REFERENCES

Agarwal, V.K. & Sinclair, J.B. (1997) Principles of Seed Pathology (2nd ed.) CRC Press, U.S.A.

Barnett, H.L. and Hunter, B.B. (1972) Illustrated genera of Imperfect fungi. Burgess Publishing Co. Minnesota.

Bolkan, H. A., De Silva, A.R. and Cupertino, F.P. (1975) Fungi associated with soybean and bean seeds and their control in central Brazil. *Pl. Dis. Repr.* 60, 545-548.

Booth, C. (1971). The Genus *Fusarium* (1st ed.) Commonwealth Mycological Institute, Kew, England.

Dharamputra, O.S., Worang, R.L., Syarief, R. and Miftahudin. (2009) The quality of physic nut (*Jatropha curcas*) seeds affected by water activity and duration of storage. *Microbiology*, 3, 139-145.

Elarosi H. (1993) Diseases of vegetables. New Publishing House, Alexandria.

Ellis, M.B. (1971) Dematiaceous Hypomycetes. Commonwealth Mycological Institute, Kew, Surrey, England.

Openshaw, K. (2000) A review of *Jatropha curcas*: An oil plant of unfulfilled promise. *Biomass and Bioenergy*. 19, 1-15.

Sahab, A.F., Aly, S.E., Nawar, L.S. and El-Faham, S.Y. (2011) Fungal occurrence in physic nut (*Jatropha curcas*) seeds during storage and possibility aflatoxin production by *Aspergillus flavus* and *Aspergillus parasiticus* isolates. *Journal of American Science*. 7(5), 511-516.

Singh, N., Harsh, N.S.K. and Bhargava, A. (1996) Biodeterioration of *Jatropha curcas* seeds. *Ann. For.* 4(1), 52-54.

Srivastava, S., Sinha, A. and Srivastava, C.P. (2011) Screening of seed-borne mycoflora of *Jatropha curcas* L. *Research Journal of Seed Science*. 4(2),94-105.

Veljkovic, V.B., Lakicevic, S.H., Stamenkovic, O.S., Todorovic, Z.B. and Lazic, K.L. (2006) Biodiesel production from tobacco (*Nicotiana tabacum* L.) seed oil with a high content of free fatty acids. *Fuel*. 85, 2671-2675.

Worang, R.L., Dharamputra, R., Syarief, R. and Miftahudin (2008) The quality of physic nut (*Jatropha curcas*) seeds packed in plastic material during storage. *Biotropia*. 15, 25-36.

Zurina, Z.A. (2009) Extraction of oil from *Jatropha* seed-optimization and kinetics. *Am. J. Appl. Sci.* 6(7), 1390-1395.