ISSN:
Print - 2277 - 0593
Online - 2315 - 7461

Journal of Natural Sciences, Engineering and Technology

MYCOFLORA OF FRUITS AND SEEDS OF SOME *JATROPHA SPP.* AND *RICINUS COMMUNIS*

***M.I. IKHATUA AND M.G. DIKORU**

Department of Forestry and Wildlife, Faculty of Agriculture,
University of Benin, Benin City.
***Corresponding author:** iyayi.ikhatua@yahoo.com **Tel:** +234-8062946177

ABSTRACT

Fungi associated with the fruits and seeds of some *Jatropha* species and their related species *Ricinus communis* were investigated in Benin City. Samples were collected from the Nigerian Institute for Oil palm Research (NIFOR), and Egor villages in the outskirts of Benin City. Some seeds of *J. curcas* for sowing obtained from the Global Renewable Energy Development Company (GREDC) in Ilorin were also investigated. Isolation and subculture of isolates on Potato Dextrose Agar (PDA), identification of isolates and pathogenicity of fruit isolate were carried out. Isolates from the seeds were identified as *Fusarium* and *Fusoma spp*. from *J. curcas, Fusarium sp*, from *J. gossipifolia* and *Fusarium* and *Cornularia spp*. from *Ricinus communis*. The isolate from the fruit of *J. curcas* identified as *Colletotrichum sp*. gave positive pathogenicity results on fruits of J. curcas, J. gossipifolia and *R. communis* but proved negative on *J. multifida. Fusarium* and *Colletotrichum spp*. seem to be commonly associated with the seeds and fruits of the species respectively. Being a promising crop on bio-oil production, the common presence of *Fusarium* species may have implication on its storage and oil quality.

Key words: Mycoflora, fruits, seeds, *Jatropher, Ricinus*

INTRODUCTION

Agriculture contributes about 28.8% Gross Domestic Product (GDP) to the economy of Nigeria (USEA, 1997). However, over the years, emphasis has been placed on petroleum because it generates the highest percentage (80%) of national income (Adeoye and Alfred, 2010). Since petroleum is exhaustible, development of oil from renewable resources will help to balance or mitigate the adverse effect of petroleum fluctuation and change the emphasis of the nation on fossil fuel (James, 2010).

Plant oils in addition to being food are important as renewable resources for both fuel and chemical industries and *Jatropha* is one of such plants (Dyer and Mullen, 2008). *J .curcas* (Barbados nut) is often referred to as *Jatropha*. It produces seed with high oil content. The seeds are toxic and thus non-edible (Kurawa, 2010). The species is native of America (Holm *et.al.,* 1979) although it is almost pan tropical now as it easily establishes itself. It is a multipurpose plant with many applications. These include: seeds and fruits as contraceptives. (Horhammer 1979) hedge fences and dye (Richardson 1979; Mitchell and Rook, 1979); young leaves as vegetable (Ochise 1980); pesticidal (Tewari and Shukla, 1982); traditional medicine (Manandhar, 1989) and recently its oil from seeds as fuel to engines since its sulphur content is only 0.25% higher than that of normal

diesel (Forson *et al* 2004, Agarwal and Agarwal, 2007). Forson *et al. (*2004) reported on the performance of diesel *Jatropha* fuel blends in a diesel engine in Ghana. The bio-oil can also be the basis for soap making while the pressed residue of the seeds (pressed cake) is a good fertilizer and can also be used for biogas production (Kurawa, 2010).

The attack of pests and diseases is a limiting factor in achieving optimum production and sometimes make harvest fail. Some pests and diseases have been recorded for *Jatropha*. They include *Clitricybe tabescens* root rot, *Colletotrichum gloeosporioides* leaf spot, *Phalospora jatrophicola* rust (James, 1960); *Dothiorella, Colletotrichum, Oidium, Alternaria and Fusarium* foliage diseases (Grimm and Macs, 1999), and fruit spoilage by *Xanthomonas* and *Pseudomonas* (Jackson, 2009). In Nigeria Kurawa (2010) listed *Phytophthora*, *Pythium* and *Fusarium spp.* as damping- off and root rot organisms; and *Colletotrichum gloeosporioides* as leaf spot organism of *Jatropha*.

Fruits and seeds highly contaminated with fungi lead to the attack of the plants at different stages of development and subsequently during harvest and storage. Moulds, especially of fungi are a major cause of seed deterioration during storage. (Ashraf and Basu Chaudhary 1986, Mutters, 1998; Murray, 2000). All three *Fusarium spp*. isolated from seeds of *Brassica campestris* var *dichotoma* were found to reduce the oil content, progressively increase the fatty acid content and refractive index of oil while the oil also emitted a mouldy odour (Ashraf and Basu Chaudhary, 1986). Fungal contamination usually results in bio deterioration of fruits and seeds and reduced viability of seeds and vigour of seedlings (Valdehi, 2002).

There is paucity of information on the *mycoflora* of *Jatropha* fruits and seeds. Due to the deterioration by contaminants on fruits and seeds and the likely effect on quality of the diesel produced, this study was under taken to investigate the fungi associated with the fruits and seeds of *Jatropha*.

MATERIALS AND METHODS

Collection of Samples

Fruits of *Jatropha curcas, J. gossipifolia* and *J. multifida*, and seeds of *J. curcas*, and *J. gossipifolia* were obtained from NIFOR (Nigerian Institute for Oil Palm Research) and Egor villages in the outskirts of Benin City. Samples of a related species, *Ricinus communis* which is often mistaken for a *Jatropha* species, were also collected from NIFOR. Seeds of *J. curcas* obtained from the Global Renewable Energy Development Company (GREDC) Ilorin were also investigated. For ease of identification, the foliage and fruits of the species are shown in Plates 1-5.

Plates 1-5: Light micrographs of the fruits of *Jatrophar and Ricinus spp*. 1 and 2: *Jatropha curcas*, 3: *J.multifida*, 4: *J. gossipifolia*, and 5: *Ricinus communis*

Mycological/Pathological studies

At the time of this study only fruits of *J. curcas* showed signs of ripening/spoilage and there were no seeds from *J. multifida*. Isolation from the fruits of *J. curcas*, seeds of *J. curcas, J. gossipifolia*, and *Ricinus communis* was on Potato Dextrose Agar (PDA). Seeds and diseased chips from fruits were surface sterilised with bleach solution (1:4, v: v bleach to water) for 3 minutes, rinsed with sterile distilled water and plated on PDA in Petri-dishes. Plates were wrapped with cling film and incubated at a room temperature (30 $\pm$ 2) ^{o}C. Subcultures were maintained on PDA. Identification of isolates under the light microscope was based on morphological characteristics of vegetative hyphae and spores (Barnett and Hunter, 1998). Pathogenicity tests on fruits isolates were carried out on the fruits of *J .curcas, J. gossipifolia, J .multifida* and *R. communis*. Healthy fruits were surface sterilised with 70% ethanol and inoculated with 7-8 day old culture of the test isolate. These were incubated in moist humid chambers at room temperature. Control fruits were set up without the fungal inoculum.

RESULTS AND DISCUSSION

Fruits of *J. curcas* were commonly found to show symptoms of spoilage during the ripening process before drying up. The fruits turned brownish to black (Plate 6).

Plate 6: Light Micrograph of a healthy fruit (left) and disesed fruits of *J. curcas* in progressive symptom development

Fruits of other *Jatropha* species and *R. communis* did not exhibit this symptom. The isolate from this symptom was identified as *Colletotrichum sp*. Isolates commonly associated with the seeds of *Jatropha spp*. were *Fusarium* and *Fusoma spp*. while *Fusarium* and *Cornularia spp*. were associated with the seeds of *Ricinus communis*. Cultural and morphological characteristics of the isolates are shown in Table 1 and Plates 7-9.

Table I: Cultural and morphological characteristics of the associated fungi with fruit and seeds of *Jatropha*

Part of Plant	Source	Isolate	Cultural characteristics	Morphological characteristics	Identification
J.curcas Fruit	NIFOR	1	A whitish moderately raised coarse growth which turned greyish to black, with many orange spore masses.	Hyphae hyaline and septate. Acervuli with dark spines or setae. Conidia hyaline, 1-celled and oblong.	Colletotrichum sp.
J. curcas seeds	NIFOR, Egor and GREDC	1	A whitish, cottony, raised fast spreading growth which turned greyish to black.	Hyphae hyaline and septae Conidia hyaline and born clusters. Macro conidia, 2-6 celled, oblong, canoe shaped with pointed ends. Microconidia 1-celled and ovoid.	Fusarium sp.
J. curcas seeds	GREDC	2	A whitish to greyish to blackish, cotton wool like fast spreading growth.	Hyphae hyaline and septate, Conidia hyaline, septate, 2-4 celled, cylindrical but not pointed at the ends	Fusoma sp.
J. gossipifo-lia seeds	NIFOR	1	A whitish, cottony, raised fast spreading growth which turned greyish to black.	Hyphae hyaline and septae Conidia hyaline and borne in clusters. Macro conidia, 2-6 celled, oblong, and canoe shaped with pointed ends. Microconidia 1-celled and ovoid.	Fusarium sp.
Ricinus com-munis seeds	NIFOR	1	A whitish, cottony, raised fast spreading growth which turned greyish to black.	Hyphae hyaline and septae Conidia hyaline and borne clusters. Macro conidia, 2-6 celled, oblong, canoe shaped with pointed ends. Microconidia 1-celled and ovoid.	Fusaruim sp.
		2	A white feather-like, fast spreading not raised growth which turned black. It pro-duced dark stalked pycnidia as culture aged.	Conidia hyaline needle-like or elongated and several celled.	Cornularia sp.

NIFOR- Nigerian institute For oil Palm Research
GREDC – Global Renewable Energy Development Company.

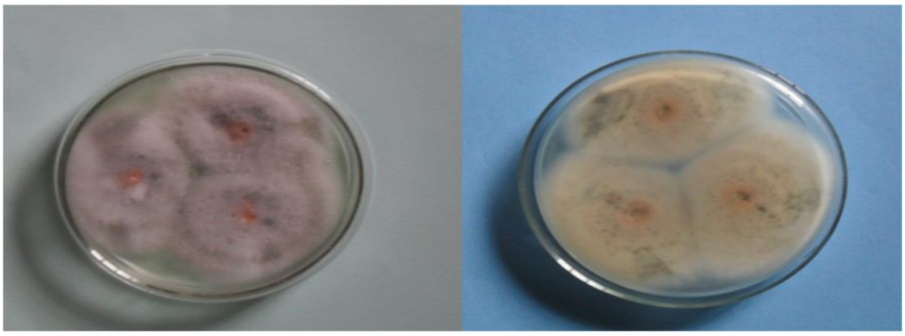

Plate 7: Light micrograph of *Colletotrichum sp*. in culture from *J. curcas* fruit. From Left to right are the upper and lower surfaces of growth

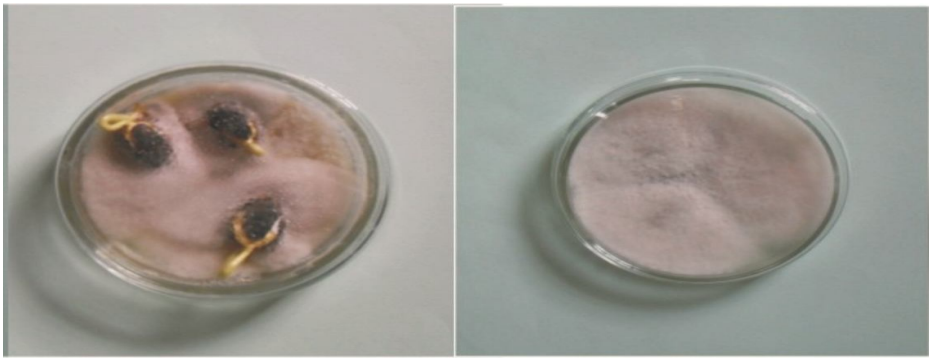

Plate 8: Light micrograph of *Fusarium* (left) and *Fusoma spp.* (right) from *J. curcas* seeds

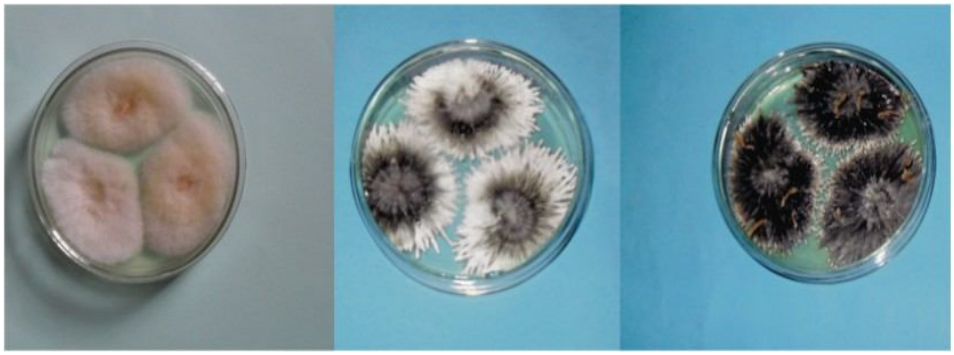

Plate 9: Light micrograph of *Fusarium sp.* (left) and *Cornularia sp* (middle and right) from *R. communis* seed

The results of pathogenicity tests with the *Colletotrichum* isolate proved positive on fruits of *J. curcas, J. gossipifolia* and *R. communis* (Plates 10 and 11) but negative on *J. multifida*. The incubation periods of *Colletotrichum sp.* on *J. curcas, J. gossipifolia* and *R. communis* were 1, 3 and 7 days respectively.

Plate 10: Light micrograph of the pathogenicity tests of *Colletotrichum sp*. on *J.curcas* fruits (control left, inoculated middle and right)

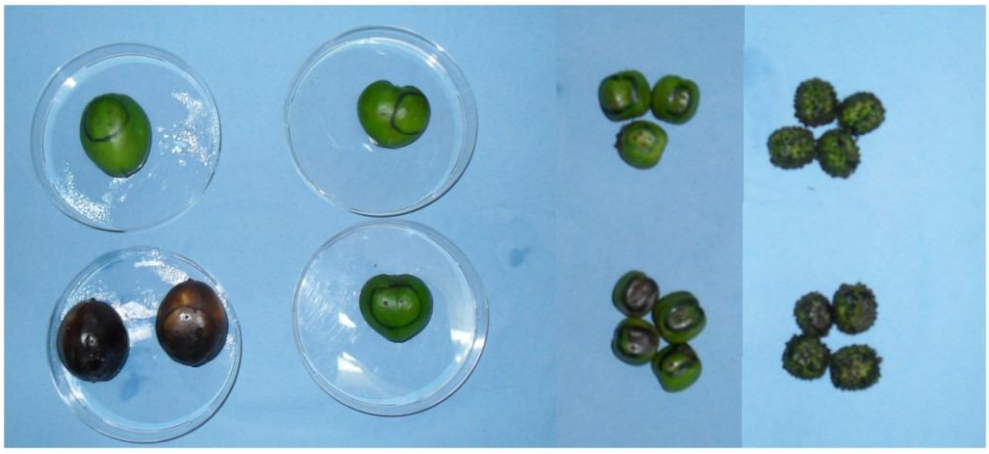

Plate 11: Light micrograph of the Pathogenecity tests of *Colletotrichum sp.* on *J.curcas*, *J. multifida*, *J. gossipifolia* and *R. communis* fruits respectively Top row are control fruits while bottom row are inoculated

The prevalence of *Fusarium spp*. on the seeds is in agreement with previous reports by Maurice (1999) and Grimm and Macs (1999) on the presence of the fungus as contaminant of seeds. Although this study has not dealt with the effect of the fungus on the seeds, they may as contaminants contribute to the post-harvest loss in quality of the seeds which may subsequently affect the quality of oils from the seed as reported by Ashraf and Basu Chaudhary (1986). According to Kurawa (2010), seeds of *Jatropha* being oily, should be properly stored and prepared for extraction to maintain a high quality of the final product. The presence of *Cornularia spp*. in association with fruits and seeds has been documented by Singh *et al*. (2008) as also shown in this study.. *Colletotrichum spp*. have been implicated as pathogen of foliar diseases of *Jatropha spp*. by James (1960), Grimm and Macs (1999) and Kurawa (2010) while this study has shown it as a pathogen of the fruits. Its cross infectivity potential on other *Jatropha* and related species has been shown with *J. multifida* resistant to its attack. In *J. carcus* the fungus may be aiding in the ripening and subsequent drying processes of the fruits which was not seen in the other *Jatropha spp.*

Being a promising crop on bio-diesel production, the common presence of *Fusarium spp*. may have implication on its storage and oil quality. Thus this present study serves as a preliminary one on which more research on the associated fungi will be based as well as their effects on the seeds and their products.

REFERENCES

Adeoye, Y., Alfred, D. 2010. Nigeria: Oil still accounts for 40% GDP, 80% Foreign exchange; Fed. Govt. Vanguard of May 3rd 2010.

Agarwal, D., Agarwal, A.K. 2007. Performance and emissions characteristics of *Jatropha* oil (preheated and blends) in a direct injection compression ignition engine. Applied Thermal Engineering, 27(2): 2314-2323.

Ashraf, S.S., Basu Chaudhary, K.C. 1986. Effect of seed-borne *Fusarium* species on the Physico-chemical properties of rape seed oil. *Journal of Phytopathology,* 117(2): 107-112.

Barnett, H.L., Hunter, B.B. 1998. Illustrated Genera of Imperfect Fungi. 4th edition. APS Press, St Paul, Mnnesota. 240pp.

Cochise, J.J. 1980. Vegetables of the Dutch East Indies reprinted. *Journal of Food Science,* 3: 77-97.

Dyer, J. M., Mullen, R.T. 2008. Engineering plant oils as high -value industrial feedstocks for biorefining: the need for underpinning cell biology research. Physiologia Plantarum 132: 11-22.

Forson, F.K., Oduro, E.K., Hammond-Donkoh, E. 2004. Performance of *Jatropha* oil blends in a diesel engine. Renewable energy 29: 1135-1145.

Grimm, D., Macs, I. 1999. Biological control can make use of beneficial arthropods. *Plant Journal* 1:17-33.

Holm, L.G, Pancho, J.V., Herberger, J.P., Plucknett D.L. 1979. A Geograp Atlas of World Weeds. John Wiley and sons. New York. 391 pp.

Horhammer, L. 1979. Hayer's hand buck. Der pharmazantischem praxis vol 2-6 springer-verlag. Berlin.

Jackson, R.W. 2009. Plant pathogenic bacteria genomics and molecular biology Caister Academic Press. 330pp.

James, A.D. 1960. Agricultural Handbookn 165. Index of Plant Disease in the United States. USGPO Washington. D.C 531pp.

James, L. 2010. *Jatropha* biofuel. Aus-Nig.com.

Kurawa, A.S. 2010. *Jatropha* cultivation, processing and biofuel production. A *Jatropha* Manual. 118pp.

Manandhar, R. 1989. Inherently sustainable technology. *African Journal of Biotechnology,* 2: 362-363.

Maurice, O.M. 1999 Problems caused by fungi:In Richard, K.R, Carl, A.B. and Pradip, D.P (ed) Encyclopedia of Food Microbiology 3: 2056-2509.

Mitchell, J.C., Rook, A. 1979. Botanical dermatology Green glass Ltd. Vancouver BC. Canada. 787pp.

Murray, M.A. 2000. Cereal production and processing. In: Nicholson Paul T. and Lan S. (eds). Ancient Egyptian Materials and Technology Cambridge. pp 609 – 655.

Mutters, S. 1998. Monitoring Rice Management Practices. Progress report of the work from fall 1993-fall 1994. University of California.

Richardson, M.J. 1979. An annotated list of seed-borne diseases. Ed. 3. Commonwealth mycol. Kew. England. 320pp.

Singh, P., Srivastava, B., Kumar, A., Dubey, N.K. 2008. Fungal contaminants of raw materials of some herbal drugs and recommendation of C*innamonum camphora* oil as herbal fungitoxicant. Microb. Ecol., 56: 555-560.

(Manuscript received: 6th June, 2012; accepted: 31st July, 2012).