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Fungi associated with oil loss in shelledcotton (*Gossypium hirsutum)* L. seeds stored at ambient temperature

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ABSTRACT: A survey of the fungal flora of shelled cotton (*Gossypium hirsutum*) seeds stored at ambient temperature and how it affects the oil content was carried out using cotton seeds obtained from some local markets in Benin City, Nigeria. Three fungi (*Rhizopus spp.; Aspergillus tarmarii* and *Candida spp.*) were isolated from the seeds. Oil loss increased with storage. Rhizopus-inoculated seeds and uninoculated ones were almost equally affected. Oil loss in seeds inoculated concurrently with the three fungi did not differ significantly from the control. Results showed that the inoculation of seeds with the three fungi had no synergistic effect on the ability of Rhizopus to degrade oil in cotton seeds whereas the reverse was the case in seeds inoculated with *Aspergillus* and *Candida*.

Key words: Fungi; Cotton seeds; Storage temperature; Oil loss.

Introduction

A member of the family malvaceae, cotton (*Gossypium hirsutum*) has about fifty genera and one thousand species (Espig, 1991). In the processing of cotton, the seeds are recovered by the removal of the lint (Langer and Hill, 1992; Gibbon and Pam, 1985). Cotton seed is a common soup ingredient in Nigeria, especially in Edo State where it serves as a substitute for melon (*Cucumeropsis edulis*) seeds, a more popular soup ingredient. Fungal flora of seeds is important in health as the former affects the deterioration of seeds which in turn detereioration to a large extent the suitability of seeds for human consumption.

Materials and Methods

Shelled cotton seeds displayed for sale in three markets (New Benin, Edaiken and Oba) in Benin City metropolis were stored in 11mm diameter plastic bowls and covered with lids. The bowls were then kept at room temperature (28 ± 2°C).

Determination of fungi present: A preliminary study was done to determine fungi originally present in apparently healthy seeds. This was done by plating out seeds on day 0 (day the seeds were collected from

markets). Ten whole seeds from each market type were surface – sterilized by shaking vigorously in Milton (5.75% Sodium hypochloite) solution diluted in the ratio 1:9 for three minutes and rinsed in several changes of sterile distilled water. Seeds were then transferred aseptically using a pair of flamed cooled forceps onto Potato Dextrose Agar, PDA (Oxoid, England) medium. The culture plates were incubated at ambient temperature (28 ± 2°C) for six weeks during which cultures were observed daily for fungal growth. Agar plates without seeds served as control.

Analysis of seeds for oil: The method of Agboola (1982) was used. The amount of oil in the samples was determined by extracting 25g ground seeds with petroleum spirit (Boiling range 60 – 8-°C) into a previously dried and weighed flask, using a soxhlet extractor. The residue in the extraction thimble was ground twice with sand, in a mortar and re-extracted. The solution was removed from the combined extracts using gentle suction to remove the last traces, and the flask containing the extracted oil heated for one hour in a ventilated oven at 100°C. The flask was then weighed after cooling in a dessicator.

Isolated fungi: Three fungi were isolated (Table 1). *Aspergillus tarmani* and *Candida sp.* were absent on day 0 whereas *Rhizopus* was present.

Oil loss: The amount of oil lost in storage increased steadily (Figure 1) with a maximum (64mg) in the sixth week. There was no significant difference in oil losses between seeds inoculated with *Rhizopus sp*. and uninoculated ones (Figure 2). For seeds inoculated with *Aspergillus tarmarii* the difference in oil losses in inoculated and control became significant from the fifth week upwards (Figure 3) whereas for seeds inoculated with *Candida spp*. it was significant in the sixth week (Figure 4). Oil loss in seeds inoculated concurrently with the three fungi did not differ significantly from the control (Figure 5). The oil loss in seeds inoculated with the three fungi was higher than that lost in seeds inoculated with *Rhizopus* or with *Candida*, but about the same as that lost in *Aspergillus* – inoculated seeds.

Table 1: Fungi isolated from cotton seeds stored for six weeks at room temperature.

Storage period (weeks)	**Isolated fungi**		
	Aspergillus tarmarii	***Candida sp.***	***Rhizopus sp.***
0	-	-	+
1	+	+	+
2	+	+	+
3	+	+	+
4	+	+	+
5	+	+	+
5	+	+	+

+ = Present
- = Absent.

Discussion

Refuse dumps of food materials commonly found around markets in this locality might have been responsible to a large extent for the high *Rhizopus* spore load of the air. Halloin (1975) reported that cotton seeds not inoculated with *Rhizopus* and inoculated ones were infected similarly. The faster-growing *Rhizopus* might have restrained the other two fungi from making their appearances early. Osolu (1985) found that food debris (carbohydrate, protein and oil) trapped spores of *Aspergillus* cotton seed contains 25%.

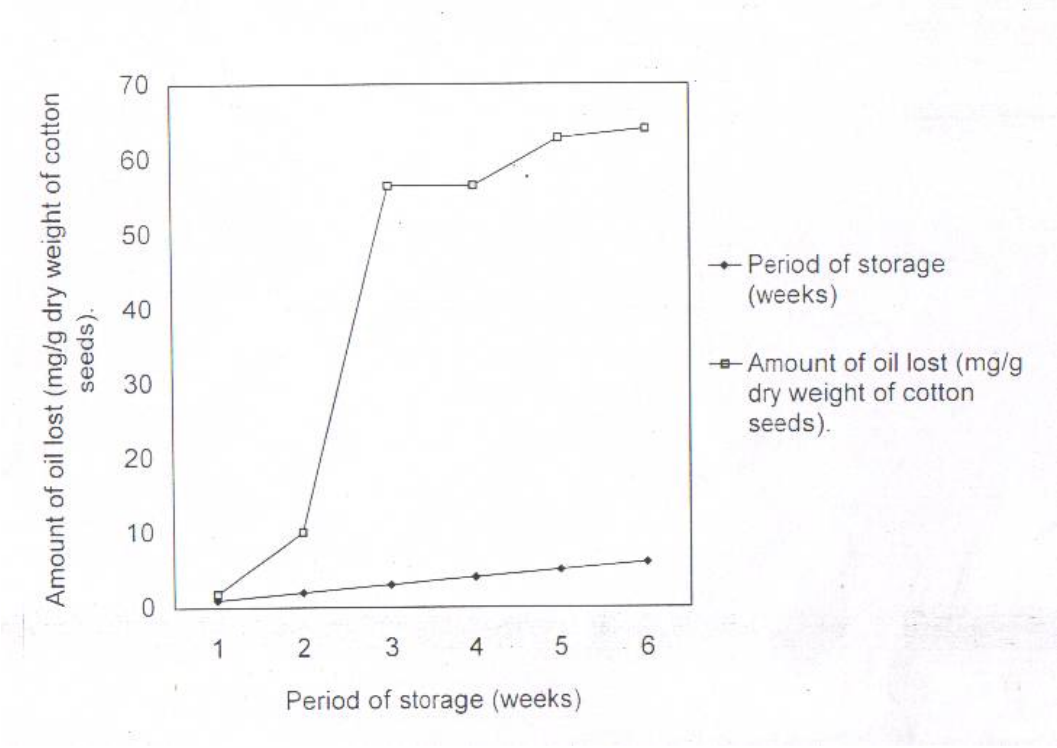

Fig. 1: Total mean oil loss in cotton seeds stored at room temperature (28 ± 2^{o}C) for six weeks.

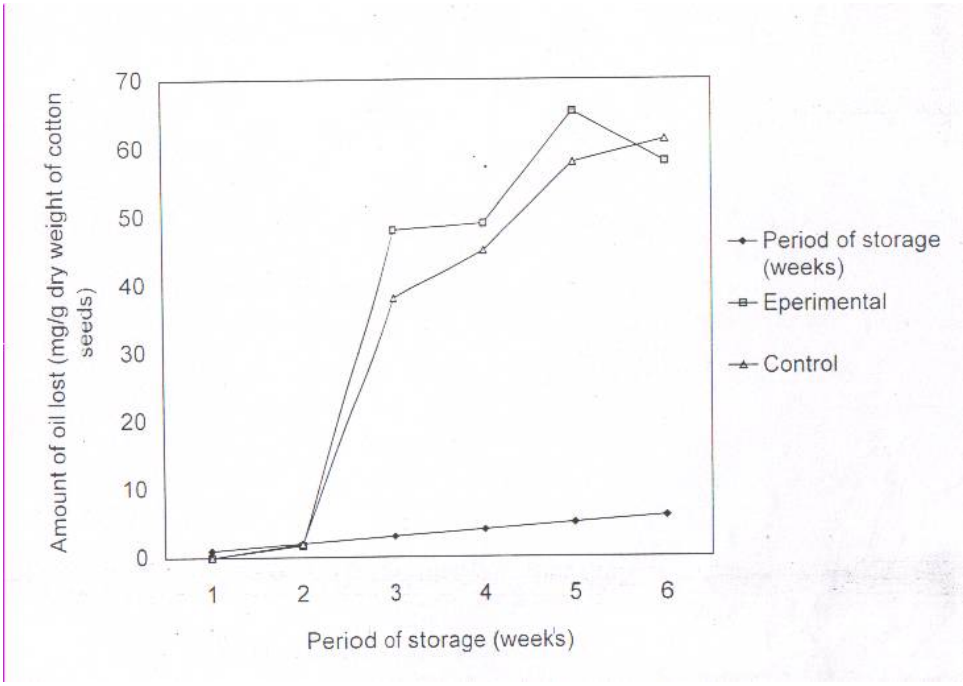

Fig. 2: Total mean oil loss in cotton seeds inoculated with *Rhizopus* spp. stored at room temperature (28 ± 2^{o}C) for six weeks.

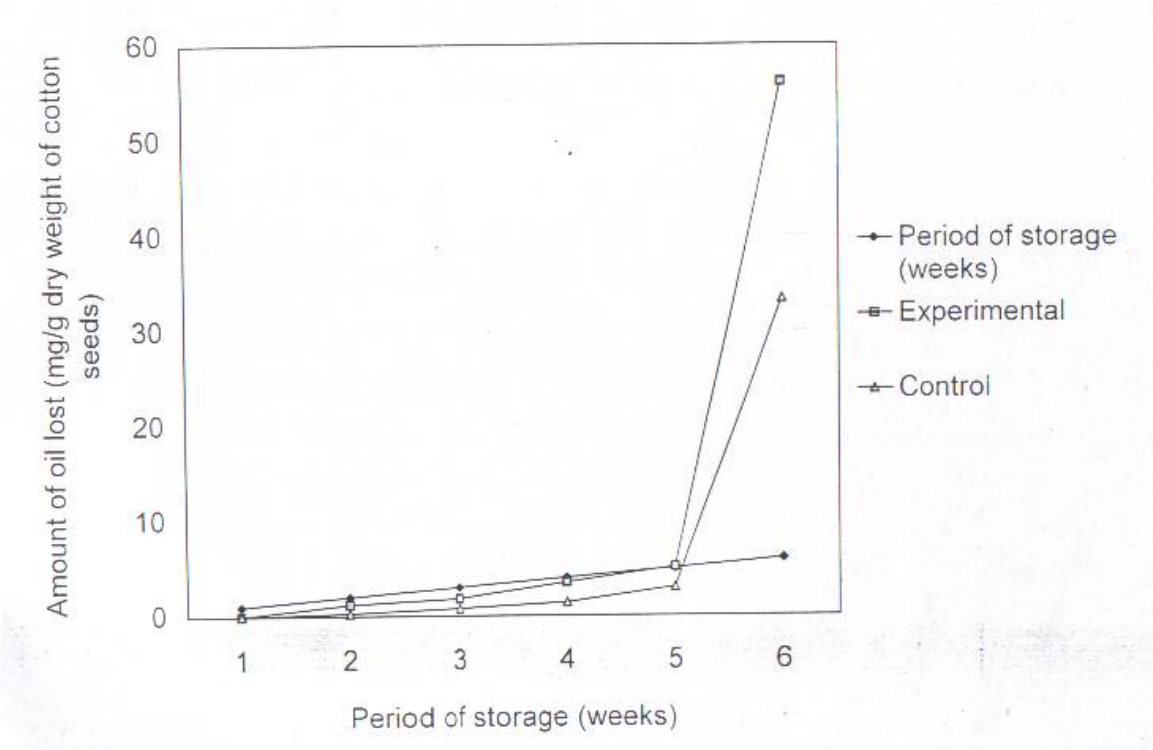

Fig. 3: Total mean oil loss in cotton seeds inoculated with *Aspergillus tarmarii* and stored at room temperature (28 ± 2°C) for six weeks.

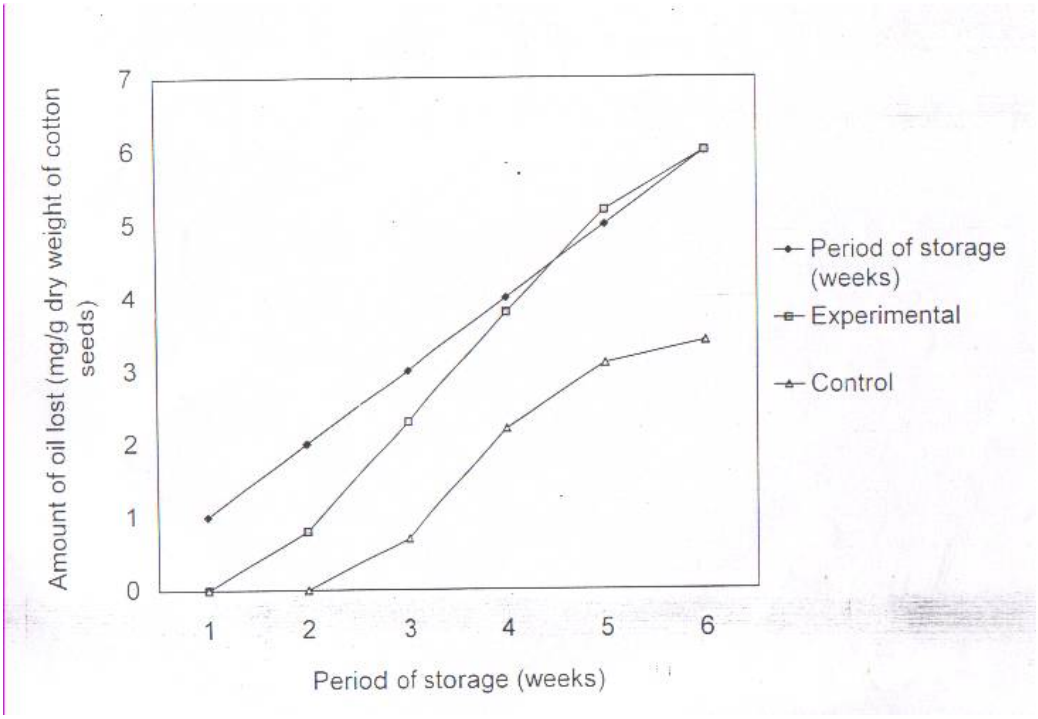

Fig. 4: Total mean oil loss in cotton seeds inoculated with *Candida* spp. and stored at room temperature (28 ± 2°C) for six weeks.

There was no much oil loss until the third week. It is possible that the fungi adapted to the substrate from this time (Peleg and Cole, 1998). The apparently equal losses in seeds inoculated with *Rhizopus* and uninoculated ones is supported by Halloin (1975) and Ekundayo (1987). The loss in oil increased with storage period for all three fungi *Rhizopus* and *Candida* seemed to degrade oil less efficiently than *Aspergillus* with either *Rhizopus* or *Candida*. Am ambient temperature of 28°C is high and could even be higher as occurs in tropical regions. High temperatures have been found to increase the rate of deterioration of stored seeds (Shama et al., 1998 and Abellana et al., 1999). The results from the study suggest that the three fungi have no synergistic effect on ability to degrade oil in cotton seeds. Therefore, in the removal or prevention of these fungi, single target approach may be more effective as the fungi may degrade more oil when they are present singly.

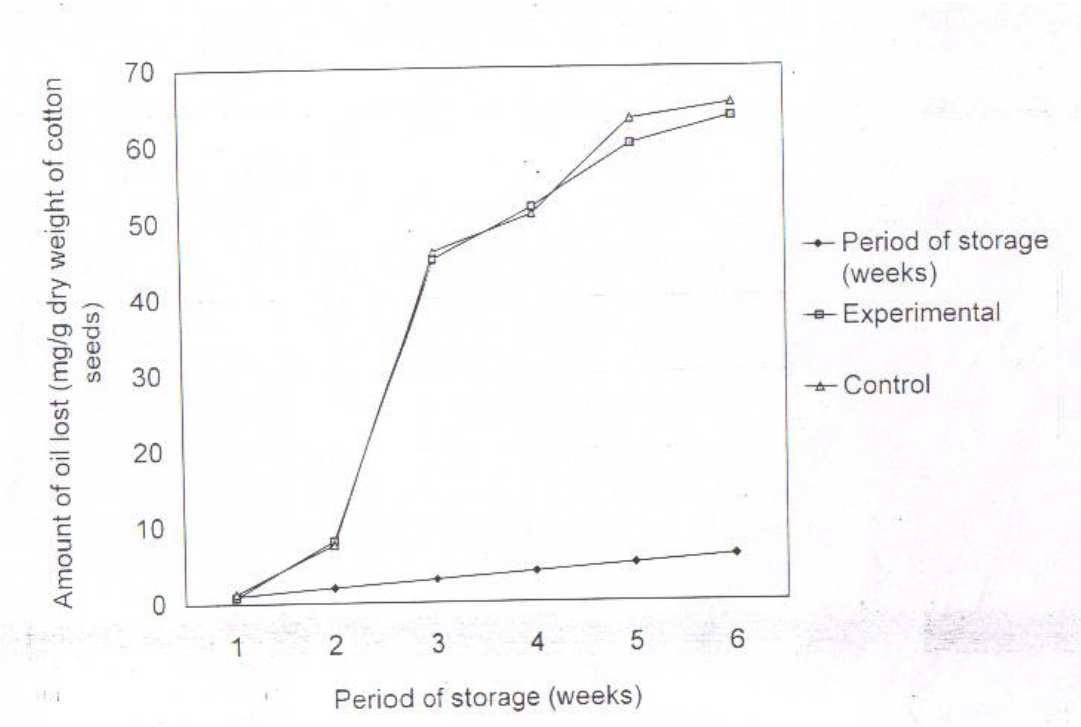

Fig. 5: Total mean oil loss in cotton seeds inoculated with *Rhizopus* spp., *Aspergillus tarmarii* and *Candida* spp. and stored at room temperature (28 ± 2^{o}C) for six weeks.

References

Abellana, M.; Benedi, J; Sanchis, V. and Ramos, A.J. (1999). Water activity and temperature effects on germination and growth of *Eurolium amstelodami*, *E. chevalieri* and *E. herbarium* isolates from bakery products. Journal of Applied Microbiology, 87 (3): 371 – 380.

Agboola, S.D. (1982). Research for effective food storage in Nigeria. Nigerian Stored products Research Institute, Occasional Paper; 4: 12 – 13.

Ekundayo, C.A. (1987). Mycoflora and vitamin content of sundried food condiments in Nigeria. Plant Foods and Human Nutrition, 37: 247 – 251.

Espig, G. (1991). The cultivated plants of the tropics. J. Margret (ed.). Eugen Ulmer G. MBH & Co. West Germany, pp. 97 – 101.

Gibbon, D. and Pam, A. (1985). Crops of the drier regions of the tropics. Longman, UK, pp. 54 – 57.

Halloin, J.M. (1975). Post harvest infection of cotton seed by *Rhizopus arrhizus,Aspergillus niger* and *Aspergillus flavus*. Phytopathology, 65: 1229 – 1232.

Langer, R.H.M. and Hill, G.D. (1982). Agricultural plants. Cambridge University Press, London, pp. 259 – 263.

Osolu, E.N. (1985). A survey of the Aspergilli in the air of Benin. Project Report, University of benin, Nigeria, 46p.

Oyeniran, J.O.(1980). The role of fungi in the deterioration of tropical stored products. Nigerian Stored products, Research Institute Occasional Paper, 2: 1 – 21.

Peleg, M. and Cole, M.B. (1998). Re-interpretation of microbial survival curves. Critical Review of Food Science and Nutrition, 38 (5): 353 – 380.

Shama, S.; Lai Cy, Antoniazzi, J.M.; Jiang, J.C.; Jazwinski Sm (1998). Heat stress – induced life span extension in yeast. Experimental Cell Research; 245(2): 379 – 388.