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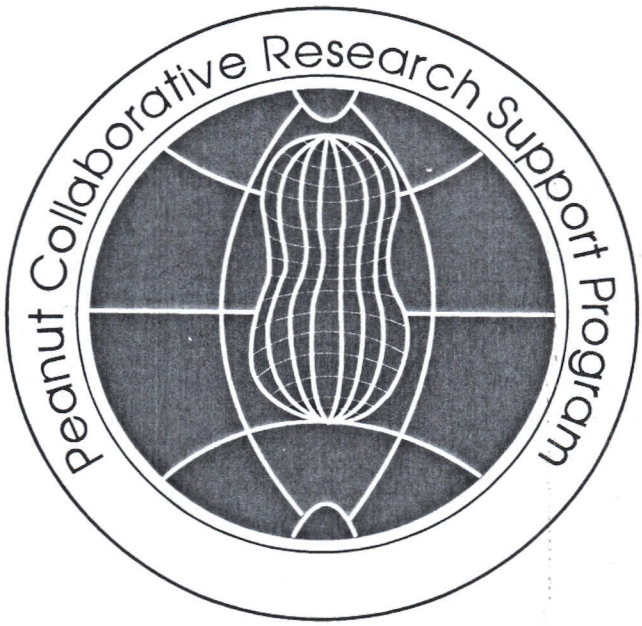

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The Incidence of Aflatoxin in Stored Groundnut in Ghana and The Effects of Some Plant Extracts on Growth of *Aspergillus parasiticus* and on Aflatoxin Synthesis

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INTRODUCTION

In Ghana, Groundnut (*Arachis hypogaea* L.) is an important food and oil crop. Much of the total national production of 113,000 metric tons (Anon., 1991) is in the Upper West, Upper East and Northern Regions of the country. These areas are within the Guinea and Sudan Savannas with an annual rainfall of about 1000 mm. Some amount of groundnut production takes place in the transitional belt around Takyiman, Kintampo and Atebubu (all in the Brong Ahafo Region) and Ejura in the Ashanti Region. The crop is also grown in the Volta, Western, Eastern, and Central Regions of the country, although production in these areas is relatively minor.

A major problem facing groundnut production in Ghana is decay and subsequent aflatoxin contamination of the seeds. Aflatoxins are carcinogenic compounds produced by *Aspergillus flavus* Link ex Fries and A. *parasiticus Speare*. Groundnut contaminated by aflatoxins is unsuitable for human and animal consumption because the aflatoxins can cause serious health problems (Enomoto and Saito, 1972; Cheng, 1992).

Even though considerable concern has been expressed in Ghana about the groundnut aflatoxin problem, studies highlighting the national importance of the problem are lacking, and the few studies attempting to address the problem have been limited in scope

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(Beardwood, 1964; Mintah and Hunter, 1989). Nor are we aware of any study in Ghana focussing on the inhibition of the aflatoxigenic fungi and control of aflatoxin synthesis.

The present research is an attempt to establish the level of contamination of stored groundnut in Ghana by fungi and aflatoxins. Results of our work on the effects of certain plant extracts on growth of *A. parasiticus* and on aflatoxin synthesis are also briefly presented.

Materials and Methods

Groundnut samples stored for 6-8 months were collected from markets in each of the ten regions of Ghana. Samples from a market were bulked together, sorted out into undamaged and damaged seeds, and the percentage of damaged seeds estimated. Fungi associated with the seeds were assayed on half strength potato-dextrose agar and identified using appropriate texts. Aflatoxins were extracted from the seeds following the method of Pons (1979). They were identified and quantified by reversed phase liquid chromatography with post column iodine derivatization (Shepherd and Gilbert, 1984). Eighteen plant extracts were tested for anti-Aspergillus and anti-aflatoxigenic action. Extracts were prepared as steam distillates, hot water infusions, and pure oils. The assay was accomplished on two aflatoxin inducing media, viz. Yeast Extract Sucrose (YES) medium and ½ strength Potato Dextrose Broth (PDB). Four milliliters of media in screw-capped vials were amended with 0.2 ml of each plant extract, inoculated with a Norsolorinic Acid (NOR) mutant of *A. parasiticus* (Keller et al. 1994) and incubated for eight days. NOR is a visible orange colored intermediate in the aflatoxin synthesis pathway and its absence from a vial shows absence of aflatoxins.

Results and Discussion

Damage in groundnut samples collected from all ten regions in Ghana ranged from 1.5 to 9.25% (Average 3.8%). Although we had expected a higher level of damage in seeds from the more humid parts of the country, we could not detect such a trend. Of the two aflatoxigenic fungi, *A. flavus* was more frequent, being observed in 31.7% of damaged seeds and from 12.8% of undamaged seeds. *Aspergillus parasiticus* was not as commonly encountered and was absent from 37 of the 42 groundnut samples assayed.

A. *niger van Thieghem* was recorded in high frequencies on both healthy (29%) and damaged (39%) seeds. Metabolites from this fungus have occasionally been shown to be toxic, so it may be useful to analyze the isolates of *A. niger* from groundnut for mycotoxins in the future.

Other fungi encountered on both damaged and unhealthy seeds, although in extremely low frequencies, were *A. candidus* Link, *A. tamarii* Kita, *A. ochraceous* Wilhelm, *Penicillium* spp, a *Trichoderma* sp, *Rhizopus stolonifer*, and a *Mucor* species. A few unidentifiable fungal species were also encountered. Some of these fungi have been reported as residents of stored peanuts (Joffe and Borut, 1966) and can produce toxins other than aflatoxins.

Aflatoxin was detected in all the damaged seed samples (Table 1). Most of these samples were characterized by aflatoxin levels greater than the WHO recommended limit of 30 ppb. Aflatoxins were not detected in some of the undamaged seed samples, and even when present in such seeds the levels were generally very low (Table 1). Thus, the groundnut aflatoxin problem in Ghana may be partly solved by sorting out damaged seeds from undamaged ones - which should not be difficult to do since the damage to groundnut in storage is low.

Steam distillates from *Xylopia aethiopica* (Dunal) Rich, *Monodera myrstica* (Gaerth), *Cinnamomum verum* Presl. and hot water infusion from *Piper nigrum* L. completely inhibited NOR formation in PDB. However, these extracts were ineffective in the YES medium. In preliminary experiments, however, we have noted that at concentrations of >0.5 ml /4 ml YES medium, distillates from *Ocimum gratissimum* L., *Cymbopogon citratus* (D.C.) Stapf. and *Xylopia aethiopica* completely prevented growth of *A. parasiticus*. This agrees with a previous report of anti-fungal activity by extracts from these three Ghanaian traditional medicinal plants (Awuah, 1989). Extracts from these plants and also those from *C. verum* and *Syzygium aromaticum*, which also completely inhibited fungal growth in the present study, should be considered for use in treating groundnut seeds to control the aflatoxin producing fungi.

Table 1. Aflatoxin levels in damaged and undamaged peanut seeds in Ghana.

	Total Aflatoxin Levels (ppb)1	
City/Town	Damaged seeds	Undamaged seeds
Accra (Greater Accra)	75.6	ND
Ashiaman (Greater Accra)	12.2	ND
Mankessim (Central)	17.7	0.2
Cape Coast (Central)	860.6	1.4
Koforidua (Eastern)	5.7	0.1
Takoradi (Western)	13144.3	ND
Kumasi (Ashanti)	105.0	0.4
Ho (Volta)	22168.0	ND
Sunyani (Brong-Ahafo)	54.2	ND
Takyiman (Brong-Ahafo)	5530.1	ND
Tamale (Northern)	71.4	0.5
Wa (Upper West)	14.5	ND
Navrongo (Upper East)	3505.2	154.2
Bolgatanga (Upper East)	301.8	12.2

1 ND = None detected.

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