THE AFLATOXIN CONTAMINATION PROBLEM IN GROUNDNUT - CONTROL WITH EMPHASIS ON HOST PLANT RESISTANCE*

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INTRODUCTION

Invasion of groundnuts by the aflatoxin-producing fungi *Aspergillus flavus* and *Aspergillus parasiticus*, and subsequent production of aflatoxins, is a serious problem in most groundnut growing countries. Aflatoxin contamination may occur pre- or post-harvest. Preharvest aflatoxin contamination is particularly important in the semi-arid tropics (SAT), especially under drought stress situations. Drought stress during late stages of pod development, a common occurrence in the SAT, predisposes seed to invasion by the aflatoxigenic fungi and consequently to aflatoxin contamination. Wet and humid conditions during postharvest drying can result in significant contamination of crop produce with aflatoxins. Aflatoxin contamination may also occur if dried, stored groundnuts absorb moisture from rainwater leakage, ground seepage, or from insect infestations.

This paper presents the current status of the aflatoxin problem worldwide with special reference to African

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groundnut producing countries, and underlines research needs. Possible practical control measures are discussed with special emphasis on use of host plant resistance.

Preharvest Aflatoxin Contamination

In this section the status of knowledge on the invasion of groundnuts by A. flavus and subsequent aflatoxin contamination before harvest is critically reviewed.

Several biotic and abiotic factors influence A. flavus invasion and aflatoxin contamination of groundnuts. Damage to pods by soil inhabiting pests and pathogens, mechanical damage to pods, and drought stress are all important factors predisposing the seeds to invasion by A. flavus (3,12,34,36). Delayed harvesting can also result in seed invasion by A. flavus and aflatoxin contamination (34,42). Soil types and soil temperatures also influence aflatoxin contamination of groundnuts.

Insect damage and aflatoxin contamination

A number of soil inhabiting pests such as pod borers, millipedes, mites, termites and nematodes attack groundnuts in the field, and have been implicated in A. flavus infection and subsequent aflatoxin contamination of groundnuts before harvest. The lesser cornstalk borer (Elasmopalpus lignosellus Zeller) is a common pest of groundnuts in the USA and has been found influential in predisposing groundnut fruit to *A. flavus* infection (12). Drought conditions favour infestation by lesser cornstalk borers which damage pods and feed on the kernels (19). *A. flavus* propagules may be carried by the insect to ideal infection sites where the kernel is damaged. Kernels from damaged pods generally contain very high levels of aflatoxins (12). Another serious pest of groundnut in the USA, the southern corn rootworm (*Diabrotica undecimpunctata howardi* Barber), has been reported to be associated with increased fungal invasion of groundnut fruit (53). The insect feeding sites provide portals of entry into the groundnut fruit for *A. flavus* and other soil fungi. An earwig (*Anisolabis annulepes* Dohrn) is a pest of economic importance in southern India and in Israel, particularly in black soils (2). Both adults and nymphs can bore into young tender pods and feed on developing kernels thus facilitating invasion by *A. flavus*.

Termites (*Microtermes* spp.) are important pests of groundnuts in India and several African countries (21,27). They cause pod scarification, and may also attack the tap root, causing wilting and premature death of plants. *Microtermes* spp. can also penetrate the pod, consuming the pod lining and occasionally the kernels. Pod scarification is restricted to the more mature pods. Such damage to pods can lead to invasion of seeds by *A. flavus* (26,34). *Odontotermes* spp. also feed on pods, scarifying and occasionally penetrating shells and rendering them susceptible to invasion by A. flavus and other soil fungi. Kernels from termite-damaged pods are likely to contain aflatoxins (35). It would be of interest to determine whether there is a quantifiable relationship between the degree of pod scarification and infection of the kernels by A. flavus.

Several species of mites have been found to penetrate groundnut pods, feed on the kernels, and disseminate A. flavus spores (4). A number of studies have been conducted on the potential involvement of nematodes in the aflatoxin problem in groundnuts in Georgia, USA (7,24,43,44,45). These studies have failed to establish a definite link between nematode infestation and A. flavus infection or aflatoxin contamination.

The involvement of soil pests in the fungal infection process and the subsequent development of aflatoxins must be considered an important aspect of the overall aflatoxin problem in groundnut.

Pod damage by pathogens and aflatoxin contamination

Several soil inhabiting fungi such as Rhizoctonia solani, Sclerotium rolfsii, and Fusarium spp. commonly cause diseases of roots, stems, and pods. Premature death of plants, particularly during pod development and maturity, from root and stem infections by these pathogens increases the chance of seeds being contaminated with aflatoxins

(34,50). Lesions produced on shells by these pathogenic fungi also facilitate invasion of seeds by A. flavus (3,36). Some virus diseases such as bud necrosis disease, caused by tomato spotted wilt virus (TSWV), may also predispose groundnuts to invasion by the aflatoxigenic fungus.

Mechanical damage to pods during field cultivations and at harvesting also predisposes seeds to invasion by A. flavus and aflatoxin contamination (3,35,58). Mechanical injury, growth cracks, and pod damage by soil inhabiting pests have received considerable attention in relation to aflatoxin contamination, but very little is known of any association between diseases of groundnut and aflatoxin contamination of seeds. Schroeder and Ashworth (58), in the USA, found that kernels from pods with mechanical damage and growth cracks had higher levels of aflatoxin than those with rot and insect injury. It would be interesting to obtain information on the incidence of A. flavus and aflatoxins in seeds of pods from plants attacked by different diseases in different groundnut growing regions.

Drought stress and aflatoxin contamination

In the SAT, most A. flavus invasion and aflatoxin contamination of groundnuts occurs before harvest (36). About 67% of the total world production of groundnut is from rainfed areas of the SAT where groundnuts are usually grown under low, irregular rainfall. Drought stress during late stages of pod development, a common occurrence in the SAT, is the most important contributing factor in A. flavus invasion and aflatoxin contamination of groundnuts (11,15,42,52). Most reports of preharvest contamination of groundnuts with aflatoxins have been from areas where crops have been subjected to drought, and particularly from the semi-arid tropics (SAT).

An association between late season drought stress and increased A. flavus invasion and aflatoxin contamination in groundnuts was documented as early as 1965 in South Africa (59) and has been confirmed by researchers in Nigeria (35), in Senegal (69), in the USA (15,16,52,56,65), and in India (42). However, some of these studies revealed that drought stress alone was not responsible for aflatoxin production since drought-stressed groundnuts were not always contaminated with aflatoxins (22). Researchers in the USA (10,57), using novel experimental plots designed to monitor soil moisture and temperature, have defined the conditions for optimum A.flavus invasion and aflatoxin contamination as a mean pod-zone soil temperature of 28–30.5 C in drought conditions during the 40–50 days before harvest. They reported no aflatoxin contamination in kernels of undamaged pods from crops grown with adequate irrigation (irrespective of pod-zone soil temperature), or from drought-stressed crops when the mean pod-zone soil temperature during the last 40-50 days before harvest was <25 C or >32 C. This suggests that groundnuts grown under drought stress may not be contaminated with aflatoxins unless drought is accompanied by mean pod-zone soil temperatures of 25-31 C during late stages of pod development. Surprisingly, these researchers have reported high levels of A. flavus (25-70%) from different categories of undamaged pods even from irrigated plots. These levels of the fungus in seeds of undamaged pods at harvest appear extremely abnormal.

Reduced metabolic activity due to decrease in pod moisture content under drought conditions probably explains the increase in susceptibility of groundnuts to A. flavus infection and aflatoxin production. Another possible role of drought stress in preharvest aflatoxin contamintaion could be to suppress microbial competitors of the aflatoxigenic fungus by elevating the soil temperature in the pod zone.

Although various studies have pointed to the importance of late season drought stress in aflatoxin contamination very little is known of the effects of early, mid season or multiple drought stress in the growing season.

The high level of pod and seed invasion by A. flavus in the soil has also been associated with over-maturity (35,42). In 1963, Nigerian groundnuts left in the ground for four weeks after maturity contained aflatoxin (35). Data from Alabama, USA also demonstrated that a much higher percentage of A. flavus invasion occurred in overmature seed and pods than in immature and mature seed and pods from the same plants at harvest (17). In India, Mehan *et al.* (42) showed that levels of *A. flavus* and aflatoxin B1 were much higher in seeds from overmature pods of several groundnut genotypes than in seeds from immature and mature pods, especially under drought stress conditions. Seeds become more susceptible to *A. flavus* invasion when the soil moisture in the pod zone approaches levels at which moisture content of the seed falls below 31% (14). Drought stress, lowered seed moisture content, over-maturity, and decreased vigour in groundnuts are interrelated and moisture related, and they all contribute to increased susceptibility to *A. flavus* invasion and aflatoxin contamination.

Soil types and aflatoxin contamination

Groundnuts are cultivated on a wide range of soils including light sandy soils, Alfisols, Oxisols, Inceptisols, and Vertisols, in different regions of the world, but little is known of the effects of these soils on preharvest aflatoxin contamination of groundnuts (20). Preliminary observations suggest that the incidence of *A flavus* invasion and aflatoxin contamination is likely to be much higher in groundnuts grown on sandy soils and Alfisols than in groundnuts grown on Vertisols (Mehan, unpublished data). This appears to be related mainly to the water-holding capacities of the soils; light sandy soils and Alfisols have low water-holding capacity and groundnuts grown on these soils are more prone to drought stress than those grown on

Vertisols that have high water-holding capacity. Light sandy soils and Alfisols appear to favour rapid proliferation of the aflatoxigenic fungus, particularly under dry conditions late in the growing season. Further investigations are needed to determine interactions between drought stress and A. flavus invasion and subsequent aflatoxin contamination of groundnuts in Vertisols.

Some research has been done on possible effects of calcium content of seeds on preharvest aflatoxin contamination, but no definite relationship has been established (9,66). If a relationship exists it could be a complex one as there is an interaction between drought and calcium deficiency.

Pod splitting is another factor contributing to aflatoxin contamination. Spanish and valencia groundnuts, maturing under fluctuating soil moisture conditions such as may occur in seasons of inadequate or irregular rainfall, are prone to pod splitting. Seeds in split pods are frequently invaded by A. flavus and subsequently contaminated with aflatoxins (20).

Infection of groundnut fruit by A. flavus

It is well established that A. flavus invasion can occur during pod development and maturation; the fungus entering by penetrating the pod wall or through a passage created by pod damage. However, the exact pathway of infection of groundnut fruit has not been fully delineated. Researchers in the USA (33,60,63) have suggested that *A. flavus* may invade through the flowers, travel down the pegs and become established in the developing seed. However, recent studies in Australia (Pitt 1984; Personal Communication) have failed to establish a definite link between flower and peg invasion, and between peg and fruit invasion. More research is needed to answer the important question "Can flower and peg invasion lead to invasion of groundnut fruit by *A. flavus* ?"; and Can this occur under both normal and drought stress situations" ?

Postharvest Aflatoxin Contamination

Until the early 1970s aflatoxin contamination in groundnuts was attributed mainly to factors arising in postharvest field drying of the crop produce (16). In the mid-1970s it became clear that *A. flavus* invasion and aflatoxin contamination of groundnuts could occur before harvest (12,15,16). Preharvest invasion of groundnuts by aflatoxigenic strains of *A. flavus* can lead to serious aflatoxin contamination during drying of the crop produce in the field if environmental conditions favour development of the fungus during this stage. During postharvest drying there may be considerable invasion of seeds by *A. flavus* already established in the shell. This is encouraged if drying is slow and seeds are in the very susceptible range of 12-30% moisture content for extended periods. In the windrows, groundnuts generally dry fairly rapidly from an initial 40 to 50% moisture content to 30 to 15% moisture, a range conducive to growth of A. *flavus*. In warm, wet weather the drying time is extended and the risk of aflatoxin contamination is increased (23,64). If the pods dry quickly, chances of fresh invasion by A. *flavus* are unlikely, although the fungus may grow and produce aflatoxin in seeds which were already infected. If windrowed groundnuts are wetted by rain, drying is slower, and considerable seed infection and aflatoxin contamination may occur (23,64). In areas where rains continue after harvest, field drying of groudnuts can present problems and serious aflatoxin contamination is likely to occur.

Groundnuts are not invaded by A. *flavus* when their moisture content is below 8%. Under poor conditions of storage, seeds may be wetted by rain or may absorb moisture from the humid atmosphere to increase their moisture content to above this level resulting in rapid invasion by the aflatoxigenic fungus with consequent aflatoxin contamination. Groundnut seeds already infected with the fungus before storage can show increased levels of aflatoxin contamination if environmental conditions permit fungal growth and aflatoxin formation (12). High relative humidity and temperatures, rainwater leakage, condensation, non-uniform drying, pod damage, and insect infestation are all important factors contributing to aflatoxin contamination of groundnuts in storage (16).

CONTROL OF AFLATOXIN CONTAMINATION IN GROUNDNUT

Invasion by _A. flavus_ and subsequent aflatoxin contamination of groundnuts can be prevented or greatly reduced by adopting certain cultural, crop drying, and storage practices that will be discussed in this section.

Control of aflatoxin contamination before harvest

A. flavus invasion and aflatoxin contamination of groundnuts before harvest can be prevented or greatly reduced by avoiding drought stress, particularly during pod development and maturity. Providing adequate soil moisture for 4-6 weeks before harvest should prove effective in preventing _A. flavus_ invasion of groundnuts. Supplementary irrigation to the rainfed groundnut crop, especially during drought stress late in the growing season, prevents the risk of aflatoxin contamination prior to harvest and may also reduce damage to pods from soil inhabiting pests such as pod borers, and termites. The beneficial effects of irrigation in alleviating preharvest aflatoxin contamination may be negated if the entire groundnut field is not covered by the irrigation system. Since preharvest aflatoxin contamination occurs mainly in drought-stressed groundnuts, this factor can result in mixing of contaminated and non-contaminated groundnuts during harvest. In some areas under severe drought stress conditions, where soil becomes hard at the time of harvest, irrigation is given to facilitate lifting of the crop. In such cases lifting should be done immediately after irrigation, otherwise complete rotting of pods may occur. It is appreciated that very few groundnut farmers in SAT Africa have irrigation facilities.

Other cultural practices that alleviate drought stress and consequent aflatoxin contamination include weed control, optimum planting rates, adequate soil fertility, and proper choice of planting date. By proper choice of planting date and by using early-maturing (short-duration) cultivars, drought stress during the critical pod development period may be avoided. Groundnut genotypes have been reported to differ in their response to drought stress. Although it has not been definitely demonstrated that drought-tolerant cultivars have greater resistance than drought-susceptible cultivars to infection by _A. flavus_ and the level of aflatoxin contamination, it would appear reasonabe to grow drought-tolerant cultivars in areas where late season drought stress is of common occurrence.

Practices that lower the incidence of soil insects, mites, and nematodes will certainly contribute to improved yield and quality of groundnuts, but their value in reducing aflatoxin contamination has not been proven except in the case of termites in South Africa (59).

Preharvest aflatoxin contamination can also be substantially reduced by avoiding mechanical damage to pods during weeding and harvesting. Harvesting the crop at proper maturity and avoiding delayed harvest can also help reduce *A. flavus* infection and subsequent aflatoxin contamination.

Attempts to show that *A. flavus* populations and aflatoxin levels are influenced by crop rotations have given conflicting results (25,51). Crop rotation is unlikely to be an important determinant of *A. flavus* populations in areas where droughts are frequent.

It would be interesting to assess the incidence of *A. flavus* invasion and aflatoxin contamination in early maturing groundnut cultivars that may fit into low rainfall environments and relay cropping systems, particularly those involving rice.

Control of Postharvest Aflatoxin Contamination

Postharvest aflatoxin contamination can be prevented or minimised by the following practices: (i) avoiding mechanical damage to pods during harvesting and subsequent processing, (ii) drying the produce in the field as rapidly as possible (at the small-scale farmer level harvesting in dry weather and drying the plants in inverted windrows is the most feasible system), (iii) preventing rewetting of the crop produce during or after the drying process, (iv) removal of any damaged or moulded pods from the produce, (v) drying the produce to a safe moisture level (8-9%) before storing, and (vi) storage of the produce at low temperature and low humidity. It is then important that the dried pods are protected from accidental wetting and are stored properly with protection from insect infestation.

Much research has been conducted on the conditions necessary to prevent mould growth during storage of agricultural commodities. Research is needed, however, on ways to apply the principles that have been developed to appropriate storage practices in various countries. Such revalidation is important if optimal conditions for storage are to be determined in respect of the varying environmental conditions in different countries. Where groundnuts are intended for human consumption, further reduction in contamination may be achieved by selective processes such as hand-picking and electronic sorting to remove visibly mould damaged seeds. Although there is no absolute correlation between visible mould damage of pods or seeds and their aflatoxin contents, mould-damaged seeds are more likely to have been invaded by _A. flavus_ than are clean, apparently healthy seeds. Electronic sorters for colour sorting of seeds have been tried out with variable results, and with some cultivars it has been possible to sort out discoloured seeds from healthy seeds and so reduce overall levels of aflatoxin in seed samples. However, there were problems associated with intrinsic seed colour differences between cultivars and with discolouration being sometimes caused by factors other than fungal invasion (12). In the USA, segregation of aflatoxin contaminated groundnuts has been very successful. Segregation may be carried out at various stages - by the farmer, by storage concerns, and by manufacturers of groundnut products (13). Contaminated lots are diverted for extraction and non-food uses. This type of aflatoxin control is better suited to advanced farming and industrialized processing conditions than to the small farmer and village level processing situations so common in developing countries.

Use of cultivars with resistance to *A. flavus* and/or aflatoxin production

An effective and practical way of controlling aflatoxin contamination would be to grow groundnut cultivars resistant to seed invasion by the aflatoxin-producing fungi.

A considerable amount of research has been aimed at finding cultivars with high levels of resistance to seed invasion and colonization by *A. flavus* based on protection from seed invasion by the testa (6,30,31,39, 40,46,47,48,69,70) and large numbers of genotypes and breeding lines have been screened. The test is carried out on undamaged, mature seeds that have been dried and stored for at least one month. These seeds are then rehydrated to around 20% moisture content, and surface inoculated with a spore suspension from an aflatoxigenic strain of *A. flavus*, and then incubated at 25 C for 8 days under high relative humidity. The percentage of seeds on which the fungus develops sporulating colonies is taken as a measure of the resistance (37,48 . Varietal ratings of from less than 10 to 100% seed colonisation have been shown. Researchers in the USA (6,47,48), West African countries (69,70), and at ICRISAT Center (39,40) have found several groundnut genotypes with marked resistance to in vitro colonisation by A. flavus of rehydrated, mature, stored seed (IVSCAF).

Most research points to testa resistance being physical (31,32,61,67,68) and it has been correlated with thickness, density of 'pallisade cell' layers, and absence of fissures and cavities. Permeability could also be an important factor as presence of wax layers on the testa have been noted. Seed coat tannins and specific amino acids have also been reported to be associated with resistance to seed invasion and colonization by A. flavus (1,55).

Resistance to IVSCAF is of value when pods or seeds are wetted in storage. Such resistance should also prove useful when field conditions are unfavourable for rapid drying of the crop produce. Since the resistance depends upon the presence of an intact seed testa, any damage to the testa removes or greatly reduces resistance. This is unfortunate as most decortication processes cause damage to seeds. The resistance may therefore be of most value when groundnuts are stored in shell.

Resistance to A. flavus invasion of developing pods in the ground has received attention in recent years. It is obviously important to establish if IVSCAF-resistant genotypes also have seed resistance to invasion by A.

flavus before harvest. Researchers in Georgia, USA, failed to show any significant differences at harvest in *A. flavus* invasion or aflatoxin contamination of seed of genotypes with different levels of IVSCAF-resistance (8,11). But studies in Senegal (70) and in India (42) have shown that some IVSCAF-resistant genotypes also have resistance to the field infection of seeds by *A. flavus*. However, it should not be assumed that all IVSCAF-resistant genotypes will have resistance to pod and seed invasion by *A. flavus* in the field. In North Carolina, USA, Kisyombe *et al.* (28) demonstrated a linkage between IVSCAF-resistance and preharvest resistance in only one of 14 test genotypes. The resistance in the developing pod is likely to be complex, involving physical and biochemical factors. Environmental factors such as drought and soil types may influence competition and antagonism between *A. flavus* and other microbes in the geocarposphere.

It is important to develop an effective technique to field screen large numbers of germplasm and breeding lines for resistance to preharvest seed infection by *A. flavus*. This can best be done in sites in drought-prone areas with light sandy soils that provide a congenial environment for development of the fungus and also for seed infection. Levels of seed infection by *A. flavus* can be increased by subjecting the crop to drought stress during pod development and maturation. At ICRISAT, imposed drought stress has been used to field screen groundnut germplasm and breeding lines for resistance to preharvest invasion of seed by A. flavus. Several genotypes with resistance to preharvest A. flavus seed invasion have been identified. Some genotypes with preharvest resistance also have resistance to A. flavus invasion and colonization of rehydrated, mature, stored seed.

It is imperative to place emphasis on resistance of the groundnut fruit to A. flavus infection rather than solely on seed resistance. The resistance of the groundnut fruit to A. flavus invasion appears to be associated with certain structural and biochemical characters of the pod and seed, and there is a possibility that genotypes may have differential effects upon the populations of A. flavus in the geocarposphere. It would be interesting to determine if cultivars with different pod characters and in different botanical types show substantial differential susceptibility to A. flavus.

A different but none-the-less useful form of resistance would be one in which genotypes had seeds which could be invaded and colonized by aflatoxigenic strains of A. flavus but in which the fungus could not produce aflatoxins. Early research reported varietal resistance to aflatoxin production when autoclaved seeds of different genotypes were colonized by aflatoxigenic strains of A. flavus (29,54) but it was not borne out by confirmatory tests (5,18). However, there were indications that genotypes varied considerably in their efficiency as substrates for aflatoxin production (49,62). In 1979 research was started at ICRISAT to screen germplasm accessions to identify genotypes that did not support or poorly supported aflatoxin production. Significant varietal differences in rate and total accumulation of aflatoxin have been found between genotypes (38,41). At ICRISAT, we have identified two genotypes, U 4-7-5 and VRR 245, that support only very low levels of aflatoxin B1 following seed infection with an aflatoxigenic strain of A. flavus (41). Studies are currently in progress to determine whether these results can be repeated over seasons and locations. Comparisons of the chemical constitutents of seed of these genotypes and of susceptible ones from different seasons and different soil types may indicate possible mechanisms of resistance to aflatoxin production.

There is no relationship between IVSCAF-resistance and the ability of the seed to support aflatoxin production. It is possible that a genotype with both these desirable traits may be found, or it may be possible to combine these traits by crossing selected genotypes. It would be interesting to know how these resistances to IVSCAF, to natural infection of seed by A. flavus in the field, and to aflatoxin production operate and how they are inherited.

The use of cultivars resistant to A. flavus and aflatoxin production in combination with crop management practices designed to minimize risk of aflatoxin contamination could provide a solution to this serious problem.

RESEARCH NEEDS

Most groundnut producing countries in Africa and Asia, with the exception of those with significant export trade with Europe and North America, have underestimated or even ignored the problem of aflatoxin contamination in groundnuts. But in the long run economic implications in terms of decline in their export trade and health hazards to humans and animals have stimulated their interest in the aflatoxin problem to the point of earnest consideration. In recent years several countries such as Senegal, Sudan, India, and Thailand have taken up research on several aspects of the aflatoxin problem in groundnut including monitoring of aflatoxin contamination in groundnuts and their products for export purposes. In the past 25 years following the recognition of the aflatoxin problem in the early 1960s, most of the research on the aflatoxin problem in groundnut has been done in the USA, India, and Nigeria. Considerable information on the extent of the problem and on how to approach control at different stages in production and storage has been generated in these countries. This information provides a good basis for similar research or for further investigations into the problem for those countries with no research plans or where only limited research has been initiated. However, much remains to be done to define the full ramifications of the aflatoxin problem in groundnut in all groundnut producing countries and to establish effective control. There is an obvious need to conduct systematic surveys in different seasons to determine the extent to which groundnuts are contaminated with aflatoxins at different stages - at lifting, during field drying, and on-farm storage in different agroclimatological zones. Such information could help in establishing a plan for effective control of aflatoxin contamination. Also, it would be possible to identify high-, and low-aflatoxin contamination risk areas.

Little is known of the effects of different soil types on preharvest seed invasion by *A. flavus* and aflatoxin contamination, and this should be examined in different agroclimatological zones. It would be interesting to determine the effects of soil temperature and moisture on *A. flavus* seed invasion and subsequent aflatoxin contamination before harvest.

Although much is known about the effects of severe late season drought stress on seed infection by *A. flavus* and aflatoxin contamination, it is imperative to investigate the effects of moderate drought stress, and of occurrence of drought at different growth stages in a growing season. The possibility of invasion of groundnut fruit in the soil being initiated through infection of flowers and pegs needs to be properly investigated. If this is established it would be helpful in predicting aflatoxin contamination before harvest.

Preliminary results have shown that it is possible to identify groundnut cultivars with resistance to seed invasion by aflatoxin-producing fungi. More emphasis should be given to research on preharvest seed invasion by _A. flavus_. Genotypes reported to be resistant to seed invasion by the aflatoxigenic fungus should be tested in different environments in different soil types, and particularly in drought-prone areas. The resistant genotypes must be compared with commercial cultivars in farmers' fields to demonstrate their usefulness in terms of prevention or substantial reduction in aflatoxin contamination. The aflatoxin contamination status of all components of the saleable yield should be determined; most studies have concentrated on undamaged, full-sized, mature seeds.

Limited research has been done on finding groundnut genotypes that do not support, or support only very low levels of aflatoxin production following seed infection by aflatoxigenic strains of _A. flavus_. This is obviously an important area for further intensified research. Mechanisms of resistance to aflatoxin production need to be further investigated.

Efforts to breed high-yielding groundnut cultivars with resistance to _in vitro_ seed invasion and colonization by _A. flavus_ (IVSCAF) have been successful. It is obviously important to emphasize breeding for resistance to seed infection by _A. flavus_ in the field. Mechanisms of resistance to seed and pod infection should be determined and their inheritance investigated. Pod characters are likely to be important in this respect.

Research needs to be done to determine if seed position in the pod has any relation to aflatoxin contamination. This would be important in terms of improving sampling procedures for monitoring aflatoxin contamination of the crop produce.

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