Aflatoxin Contamination Problems in Groundnut in Asia

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V K Mehan
C L L Gowda

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Contents

Aflatoxin Contamination of Groundnut: Prospects for a Genetic Solution through Conventional Breeding

H D Upadhyaya[1], S N Nigam[1], V K Mehan[2], and J M Lenne[2]

Aflatoxin contamination of groundnut is a serious problem in most groundnut-producing countries. The aflatoxin-producing fungi, *Aspergillus flavus and A. parasiticus,* can invade groundnut seed in the field before harvest, during postharvest drying and curing, and in storage. The semi-arid tropical environment is conducive to preharvest contamination when the crop experiences drought before harvest, whereas in wet and humid areas, postharvest contamination is more prevalent. Aflatoxin contamination can be minimized by adopting some cultural, produce-handling, and storage practices. However, these practices have not been widely adopted by small farmers in developing countries which contribute about 60% of the world's groundnut production. Cultivars resistant to seed invasion by aflatoxin-producing fungi or to aflatoxin production would be of great value to farmers in both developed and developing countries. Therefore, breeding for resistance to aflatoxin-producing fungi and/or aflatoxin production can play a significant role in preventing aflatoxin contamination in groundnut, consequent economic losses, and health hazards.

The alleviation of aflatoxin contamination through genetic manipulation has been attempted since the mid 1970s. In spite of the significant progress achieved to date, these efforts have not resulted in complete freedom from aflatoxin contamination. The current status and future prospects of genetic solutions to the aflatoxin contamination problem are briefly discussed in this paper.

Current Status of Genetic Resistance

In groundnut, depending on the site at which it operates, resistance to aflatoxin-producing fungi may be of three types—resistance to pod infection (pod wall), to

1. Genetic Enhancement Division, ICRISAT Asia Center, Patancheru 502 324, Andhra Pradesh, India.
2. Crop Protection Division, ICRISAT Asia Center, Patancheru 502 324, Andhra Pradesh, India,

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seed invasion and colonization (seed coat), and to aflatoxin production (cotyledons). The fungi have to penetrate the pod wall and the seed coat to reach the cotyledons from which they derive their sustenance. Resistance to pod infection is attributed to pod-shell structure, while resistance to seed invasion and colonization is physical, and has been correlated with thickness, density of palisade cell layers, absence of fissures and cavities, and presence of wax layers. There are conflicting reports regarding the role of fungistatic phenolic compounds in imparting resistance to seed colonization.

All the three types of resistance sources have been reported (Mehan 1989). These include Shulamit and Darou IV for resistance to pod infection, PI 337394 F, PI 337409, GFA 1, GFA 2, UF 71513, Ah 7223, J 11, U 4-47-7, Var 27, Faizpur, and Monir 240-30 for resistance to in vitro seed colonization by A *flavus* (IVSCAF), and U 4-7-5 and VRR 245 for resistance to aflatoxin production. The importance of preharvest aflatoxin contamination was realized only in the late 1980s, and some of the IVSCAF-resistant genotypes (PI 337394 F, PI 337409, GFA 1, GFA 2, J 11, UF 71513, Ah 7223) were reported to have considerably lower natural seed infection by *A. flavus* than various IVSCAF-susceptible genotypes (Mehan 1989).

The value of a resistance source depends upon the level and stability of its resistance. Resistance to pod infection has been reported to be highly variable and of a low level. Similarly, IVSCAF-resistance is not absolute and even the best sources show up to 15% seed colonization; only a few lines (J 11, PI 337394 F, and PI 337409) have shown stable resistance. For aflatoxin contamination, resistance levels are not very high (Anderson et al. 1995).

Relationships between Types of Resistance

There are conflicting reports on the relationship between IVSCAF-resistance and resistance to natural seed infection, and aflatoxin contamination in the field. In the breeding lines developed and evaluated at IAC, no correlation (-0.07) was observed between IVSCAF and seed infection in the field, indicating two independent genetic mechanisms. The high correlation observed in an earlier study (Mehan et al. 1987) might have been due to the inclusion of some selected germplasm lines; whereas the absence of correlation observed in breeding lines developed at IAC might have resulted from the recombination of genes controlling these mechanisms. The studies conducted, in the 1980s, in the USA and at IAC showed low levels of aflatoxin contamination in IVSCAF-resistant genotypes. However, the genotypes which were earlier reported to be resistant to IVSCAF or preharvest aflatoxin contamination contained high levels of aflatoxin, when subjected to an extended period of heat and drought stress, and none of them was more resistant than the susceptible cultivar Florunner in the USA (Anderson et al. 1995). Highly significant genotype (G) x environment (E) interaction effects for aflatoxin contamination were observed in this study. The exact information on the relationship between different resistance mechanisms, their interactions, and possible contributions in reducing aflatoxin contamination has not been clearly established.

Genetics of Resistance

There are only three published reports on the inheritance of resistance, which give estimates of broad sense heritability and combining ability. The broad sense heritability estimates ranged from 55 to 79% for seed colonization, from 27 to 87% for seed infection, and from 20 to 47% for aflatoxin production. These studies were conducted in the USA (Mixon 1979, Utomo et al. 1990) and India (Upadhyaya and Nigam, unpublished). A report from the USA indicates that there is no significant correlation among the three types of resistance, indicating that they are controlled by different genes (Utomo et al. 1990). In a diallel study, significant reciprocal effects were noticed in some crosses indicating maternal influence on testa structure (Rao et al. 1989).

The genetics of resistance mechanisms has not been clearly established. The allelic relationship among various sources for each resistance trait needs to be elucidated to enable breeders to pyramid the non-allelic genes for each resistance mechanism.

Current Status of Resistance Breeding

Breeding efforts for resistance to pod infection have not received any attention. Further, it was assumed that if shell thickness was related to resistance, resistance breeding would result in low shelling percentages. In the past, seed colonization resistance received the maximum attention due to the ease of screening procedures. Of late, natural seed infection and aflatoxin production have received increasing attention, although screening for resistance to aflatoxin production is expensive. A much cheaper ELISA-based methodology was recently developed at ICRISAT.

Research on breeding for resistance to aflatoxin contamination is in progress in India, Senegal, Thailand, and the USA. The groups at Tifton, USA, and IAC, India, have successfully transferred IVSCAF-resistance to different genetic backgrounds. The group at Tifton produced six breeding lines GFA-1, -2, AR-1, -2, -3, and -4 (Mixon 1983a and 1983b). GFA-1 and -2 (both runner market types), whose yields were equal to or better than that of Florunner, had equal or less than average seed colonization than the resistant control genotype (PI 337409). The yield potentials of AR-1, -2, -3, and -4 are too low for their practical use as commercial cultivars.

In India, resistance breeding activities are mainly conducted at IAC and the National Research Center for Groundnut (NRCG). At IAC, research on breeding for resistance to aflatoxin contamination started in 1976. Several hundred breeding lines have since been tested for yield and IVSCAF-resistance, and several lines with IVSCAF-resistance and high yield have been identified. Four hundred and seventy-two lines were evaluated for preharvest seed infection and yield. Some of them have seed infection and colonization equal to or less than the best resistant control cultivar, J 11, and high-yield potential across seasons/years and locations. Of these, ICGV 88145 and ICGV 89104 have been released as improved germplasm lines (Rao et al. 1995). Recently, four such lines (ICGVs 91278, 91279, 91283, and 91284) were evaluated for yield and other agronomic traits in national programs in Thailand and

Vietnam, and they have performed very well. Three lines (ICGVs 87084, 87094, and 87110), bred at IAC for resistance to seed infection, were also found to be resistant in Niger, Senegal, and Burkina Faso in West Africa (Waliyar et al. 1994).

In Thailand and Senegal, PI 337394F, PI 337409, UF 71513, and J 11 are commonly used as resistant donors. The lines AR-1, -2, -3, and -4 are also being used in Thailand as sources of resistance; 55-437 has been used in Senegal.

In the breeding scheme at IAC, the selection for resistance traits is delayed until later generations. However, it would be desirable to screen segregating generations and select only resistant plants/progenies. This would require modification of screening techniques currently being used to make them more suitable at the single plant level.

Future Prospects of Breeding for Aflatoxin Resistance

Although researchers have not been able to locate germplasm lines which show complete resistance to fungi at the pod-wall, seed-coat, and cotyledon levels, it was expected that the levels of resistance could be improved further by pyramiding resistance genes, from different and diverse sources. It was also thought that by combining the three different kinds of resistance in one genetic background, the problem of aflatoxin contamination could be overcome to a large extent. Unfortunately, the progress made so far in conventional breeding has not been able to meet these expectations. The recourse to biotechnology, through modification of the aflatoxin biosynthesis pathway or the use of variants of hydrolytic enzymes (chitinases and glucanases), to provide transgenic protection to groundnut against infection by aflatoxin-producing fungi may help in obtaining groundnuts free from aflatoxin.

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