



## RELATIONSHIP OF DROUGHT TOLERANCE TRAITS WITH AFLATOXIN CONTAMINATION IN GROUNDNUT

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### SUMMARY

Fifty groundnut (*Arachis hypogaea* L.) genotypes were screened for low aflatoxin contamination under field conditions with two main treatments i.e., irrigated and simulated drought conditions. A severe strain of *A.flavus* was isolated by collecting native strains from different parts of Chittoor district in Andhra Pradesh and a sick plot was developed utilizing this strain. *A.flavus* inoculum was applied @ 2.5 g per meter row length and sufficient spore load was maintained till harvest. Mean aflatoxin content was higher under imposed moisture stress conditions compared to irrigated treatment. The genotypes differed in tolerance to aflatoxin in irrigated and simulated drought conditions. Among the fifty groundnut genotypes screened ICGV 86590, 89104, 94350, 99029, IC 48 and ICGS 76 had low aflatoxin levels (< 5 ppb) in both irrigated and simulated drought treatments. No consistent relationship was observed between seed colonization and aflatoxin production. The results indicate that aflatoxin production in groundnut is negatively related with relative water content (RWC), pod wall integrity and pod wall moisture content at harvest.

**Keywords:** Aflatoxin, pod wall integrity, pod wall moisture, relative water content

### INTRODUCTION

Aflatoxin contamination seriously affects the edible quality of groundnut and its products rendering them unfit for human and livestock consumption. Various surveys conducted in different parts of India have shown high aflatoxin levels in groundnut food products including raw peanut kernels (0.8 to 2200ppb), unrefined oil (0 to 786ppb) and groundnut cake (27 to 1122ppb) depending upon the agroclimatic location and storage conditions (Ghewande 1997). It is only in the last decade that a thorough understanding of the environmental and associated crop conditions that lead to high aflatoxin incidence have been explained (Deiner *et al.* 1987, Dorne *et al.* 1989). *A.flavus* infection could occur at pre-harvest harvest and post-harvest stages. Terminal droughts are often a pre-disposing factor for aflatoxin

contamination in groundnut. Damaged pods are likely to contain more of aflatoxins than the pods with undamaged shells. Soil temperature more than 28 °C and kernel moisture content less than 30 per cent favour aflatoxin production (Craufurd and Wheeler 2002). In the present study, the relationship between aflatoxin contamination with plant water status is examined.

### MATERIALS AND METHODS

Fifty groundnut genotypes from the groups of known tolerance to aflatoxin, foliar disease resistance, high water use efficiency, thick shell wall and different duration groups were screened for aflatoxin contamination during the *rabi* (2001-02) under mobile rain out shelters (ROS) covered with poly sheets at Regional Agricultural Research Station, Tirupati, Andhra

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**Table 1.** Aflatoxin content (ppb) and seed infection per cent of fifty groundnut genotypes under irrigated and simulated drought conditions

S.No.	Genotype	Aflatoxin content (ppb)			Seed infection per cent			Remarks
		Irrigated	Stress	Mean	Irrigated	Stress	Mean	
1	ICGS 11	7.75	1.00	4.37	26.0	2.00	14.0	Medium duration elite line
2	ICGV-86158	3.00	110.0	56.5	64.0	10.00	37.0	Dormancy & short duration elite line
3	ICGV-86590	1.00	1.00	1.00	6.00	2.00	4.00	Foliar disease resistant line
4	ICGV-86699	41.0	1.00	21.0	58.5	51.0	54.7	Foliar disease resistant line
5	ICGV-88145	3.25	2503.2	1253.2	36.5	4.00	20.2	Known aflatoxin resistant line
6	ICGV-89104	1.00	1.00	1.00	44.0	14.0	29.0	Known aflatoxin resistant line
7	ICGV-91114	1.00	4.00	2.50	14.0	80.0	47.0	Known aflatoxin resistant line
8	ICGV-91278	3.00	4.75	3.87	30.5	37.0	33.7	Known aflatoxin resistant line
9	ICGV91279	5.50	80.2	42.8	52.5	52.0	52.2	Known aflatoxin resistant line
10	ICGV-91283	6.00	5.50	5.75	76.5	30.0	53.2	Known aflatoxin resistant line
11	ICGV-91284	156.2	4.75	80.5	46.0	12.0	29.0	Known aflatoxin resistant line
12	ICGV-92206	6.00	1.00	3.50	74.5	50.0	62.2	Short duration elite line
13	ICGV-93280	2502	1.00	1251.5	8.00	4.00	6.00	Known aflatoxin resistant line
14	ICGV-93291	80.0	2510	1295	24.0	36.0	30.0	Known aflatoxin resistant line
15	ICGV-94341	3.00	18.5	10.7	22.0	16.0	19.0	Short duration elite line
16	ICGV-94350	1.00	1.00	1.00	40.0	30.0	35.0	Short duration elite line
17	ICGV-94358	9.00	1.00	5.00	30.5	14.0	22.2	Short duration elite line
18	ICGV-94433	18.0	120.0	69.0	34.5	48.0	41.2	Known aflatoxin resistant line
19	ICGV-95322	77.0	1.00	39.0	40.0	17.0	28.5	Short duration elite line
20	ICGV-95454	266.0	120.0	193.0	58.5	44.0	51.2	Known aflatoxin resistant line
21	ICGV-95460	1.00	4.50	2.75	22.0	24.0	23.0	Known aflatoxin resistant line
22	ICGV-95469	8.00	3.50	5.75	4.00	37.0	20.5	Known aflatoxin resistant line
23	ICGV-95477	70.0	1.00	35.5	22.0	60.0	41.0	Known aflatoxin resistant line
24	ICGV-95492	14.2	2513	1263.6	70.5	18.0	44.2	Known aflatoxin resistant line
25	ICGV-95494	5.00	1.00	3.00	46.0	55.0	50.5	Known aflatoxin resistant line
26	ICGV-98163	17.5	1.00	9.25	12.5	28.0	20.2	Medium duration elite line
27	ICGV-98170	10.7	12.5	11.62	26.0	47.0	36.5	Medium duration elite line
28	ICGV-98383	4.50	1.00	2.75	38.0	30.0	34.0	Foliar disease resistant line
29	ICGV-99029	1.00	1.00	1.00	16.0	4.00	10.0	Foliar disease resistant line
30	ICGV-99032	6.50	17.5	12.0	1.00	58.0	29.0	Foliar disease resistant line
31	ICGV-99054	6.75	3.00	4.87	2.00	22.0	12.0	Foliar disease resistant line
32	J-11	1.00	65.5	33.2	10.5	27.0	18.7	Known aflatoxin resistant line
33	NcAc-343	1.00	7.25	4.12	1.00	20.0	10.0	Water use efficiency
34	IC-48	1.00005.00	1.00	1.00	14.0	10.0	12.0	Water use efficiency

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S.No.	Genotype	Aflatoxin content (ppb)			Seed infection per cent			Remarks
		Irrigated	Stress	Mean	Irrigated	Stress	Mean	
35	JAL-17	5.00	1.00	3.00	18.0	22.0	20.0	Water use efficiency
36	TIR-31	5.50	177.5	91.5	10.5	38.0	24.2	Water use efficiency
37	IC-43	10.0	3.76	6.88	42.0	52.0	47.0	Water use efficiency
38	IC-10	9.27	10.0	9.63	14.0	46.0	30.0	Water use efficiency
39	TCGP-10	4.25	15.5	9.87	8.00	85.0	46.5	Thick shell line
40	TCGP-5	4.00	8.50	6.25	46.0	44.0	45.0	Thick shell line
41	TCGP-6	67.0	4.50	35.7	1.00	43.0	21.55	Thick shell line
42	TCGS-320	7.75	1.00	4.37	45.5	64.0	54.7	Nematode resistant line
43	TCGS-645	4.50	2500.5	1252.5	4.00	34.0	19.0	Soil insect tolerant line
44	TCGS-647	3.50	8.50	6.00	1.00	18.0	9.05	Soil insect tolerant line
45	TPT-3	3.50	3.00	3.25	1.00	33.0	16.5	Nematode resistant line
46	ICGV-86031	3.00	8.75	5.87	8.00	8.00	8.00	control
47	ICGS-44	5.00	120.0	62.5	28.0	13.0	20.5	control
48	ICGS-76	1.00	1.00	1.00	2.00	2.01	2.05	control
49	CSMG-84-1	9.25	17.5	13.4	2.50	8.00	5.25	control
50	JL-24	11.0	49.0	30	54.5	90.0	72.2	control
	<b>Mean</b>	69.9	271.0		27.1	31.9		
		<b>SED</b>	<b>CD(5%)</b>		<b>SED</b>	<b>CD(5%)</b>		
	<b>Treatments</b>	103.1	1309.5		0.10	1.28		
	<b>Genotypes</b>	615.6	1218.8		3.47	6.88		
	<b>Interaction</b>	870.6	1723.7		4.92	9.74		

Pradesh. The experiment was laid out in a factorial randomized block design with irrigation regimes as main treatments. Fifty groundnut genotypes were selected and each genotype was replicated thrice. Each genotype was planted in 3 rows of 4m length with 30cm row spacing and 15cm intra row spacing. Fertilizers were applied at the rate of 20:40:60 N, P<sub>2</sub>O<sub>5</sub>, K<sub>2</sub>O Kilograms per hectare. A severe strain of *Aspergillus flavus* was isolated by collecting native strains from different parts of Chittoor district in Andhra Pradesh and a sick plot was developed. *Aspergillus flavus* inoculum was applied @ 2.5 g per meter row length at 25 and 40 DAS and sufficient spore load maintained throughout the experiment. Moisture stress was imposed from 70 days after sowing till harvest. Soil temperatures and relative humidity were recorded daily using Gemini data loggers after the imposition of moisture stress. Soil moisture

contents were recorded periodically using soil moisture diviner. Physiological parameters, viz. Relative leaf water content, leaf temperatures, pod and kernel moisture content (Cole *et al.* 1993), pod wall integrity (Reddy *et al.* 2003) at harvest were measured. After harvesting, the natural seed infection per cent by *Aspergillus flavus* in the kernels was measured (Mehan *et al.* 1986) and aflatoxin content was estimated by indirect competitive ELISA method (Reddy *et al.* 1988).

## RESULTS AND DISCUSSION

The mean aflatoxin content was significantly low under irrigated (69.9 ppb) compared to simulated drought conditions (271.0 ppb) (Table 1). Among the fifty genotypes screened ICGV 86590, 89104, 94350, 99029, IC 48 and ICGS 76 had low aflatoxin levels (< 5ppb) in

both irrigated and simulated drought treatments. Interaction effects between irrigation treatments and genotypes were significant. Seed infection per cent values were high under simulated drought treatments (31.9%) compared to irrigated treatment (27.1%) (Table 1). Aflatoxin content and seed infections were high under moisture stress conditions indicating that pre-harvest drought conditions are highly congenial for invasion of *Aspergillus flavus* and production of AFB1 (Aflatoxin B1) in kernels. However the risk is limited under irrigated conditions. Though several workers have reported positive correlation between invitro seed colonization and field tolerance, the relationship is inconsistent (Mehan 1989). Cole *et al.* (1993) reported that under prolonged drought conditions, groundnut genotypes which maintained high kernel moisture showed enhanced resistance and produced low aflatoxin. The present study revealed no consistent relationship between seed infection per cent and aflatoxin production. Priyadarshini and Tulpule (1978) recorded no correlation between fungal infection and aflatoxin production and concluded that aflatoxin production depends on many other environmental factors besides *A. flavus* infection. Hence, the levels of *A. flavus* seed infection cannot be directly correlated to the aflatoxin production (Davidson *et al.* 1982).

Correlation studies between aflatoxin content with leaf relative water content, pod wall moisture and pod wall integrity indicate that aflatoxin production in groundnut is negatively related with leaf relative water content ( $r^2=0.25$ ,  $P<0.05$ ) (fig:1), pod wall moisture ( $r^2=0.28$ ,  $P<0.05$ ) (Fig:2) and pod wall integrity ( $r^2=0.33$ ,  $P<0.05$ ) (fig:3) at harvest. There is limited information on the pod wall characteristics and fungal invasion. These results further confirm that aflatoxin production in kernels is mitigated when plant maintain high relative water content and is the reason why drought is a predisposing factor for aflatoxin production in groundnut. Zambettakis *et al.* (1981) attributed pod infection to variation in shell structure. In conclusion the present study revealed that, genotypes that had low aflatoxin content both under irrigated and drought conditions maintained high leaf relative content, pod wall moisture content and pod wall integrity values. There was no consistent relationship between seed infection and aflatoxin production.

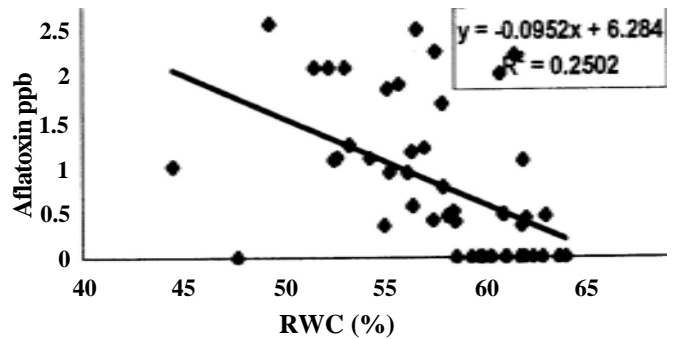


Fig. 1. Relationship between aflatoxin content and relative water content

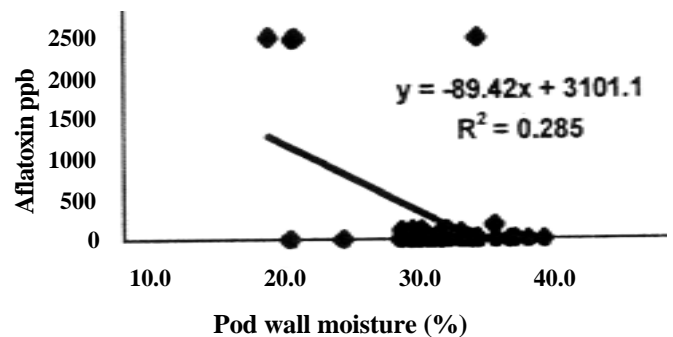


Fig. 2. Relationship between aflatoxin content and pod wall moisture

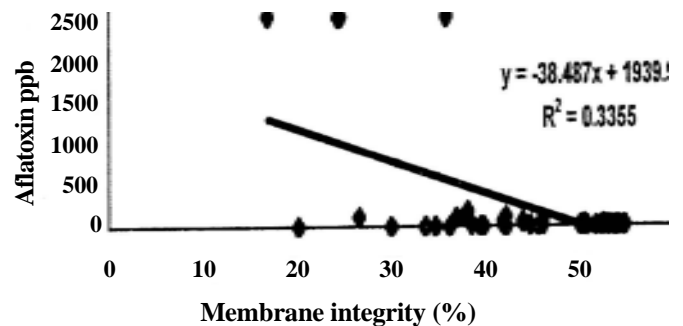


Fig. 3. Relationship between aflatoxin content and pod wall integrity

## REFERENCES

- Cole, R.J., Sobolev, V.S. and Dorner, J.W. (1993). Potentially important sources of resistance to prevention of preharvest aflatoxin contamination in peanuts. *Proc. American Res. Edu. Soc.* **78** (Abstract).
- Davidson, J.I., Hill, R.A., Cole, R.A., Mixon, A.C. and Henning, R.J. (1982). Field performance of two peanut cultivars relative to resistance to invasion by *A. flavus* and subsequent aflatoxin contamination. *Proc. American Peanut Res. Edu. Soc.* **14**: 74-78.

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- Diener, U.L., Cole, R.J., Sanders, T.H., Payne, G.A., Lee, L.S. and Klich, M.A. (1987). Epidemiology of aflatoxin formation by *Aspergillus flavus*. *Annu. Rev. Phytopathol.* **25**: 249-270.
- Dorner, J.W., Cole, R.J., Sanders, T.H. and Blanhenship, P.D. (1989). Interrelationship of kernel water activity, soil temperature, maturity and phytoalexin production in preharvest aflatoxin contamination of drought stressed peanuts. *Mycopathologia* **105**: 117-28
- Ghewande, M.P. (1997). Aflatoxin contamination of groundnut and its management in India. Pages 27-31 in Aflatoxin contamination problems in groundnut in Asia. *Proc. First Asia Working Group Meet.*, 27-29, May 1996.
- Mehan, V.K., Mc Donald, D., Ramakrishna, N. and Williams, J.H. (1986). Effects of genotypes and date of harvest on infection of peanut seed by *Aspergillus flavus* and subsequent contamination with aflatoxin. *Peanut Sci.* **13**: 46-50
- Mehan, V.K. (1989). Screening groundnut for resistance to seed invasion by *Aspergillus flavus* and aflatoxin production. In: Aflatoxin Contamination of Groundnut. pp. 323-334. *Proc. International Workshop*, 6-9 October, 1987. ICRISAT, India.
- Craufurd, P. and Wheeler, T. (2002). Factors contributing to pre-harvest and post-harvest infection and contamination. DFID Project, Workshop report-2002, pp. 1-7.
- Priyadarshini, E. and Tulpule, P.G. (1978). Relationship between fungal growth and aflatoxin production in varieties of maize and groundnut. *J. Agri. Food Chem.* **26**: 249-252.
- Reddy, D.V.R., Nambiar, P.T.C., Rajeswari, R., Mehan, V.K., Anjaiah, V. and Mc Donald, D. (1988). Potential of enzyme linked immunosorbent assay for detecting viruses, fungi, bacteria, mycoplasma like organisms, mycotoxins and hormones. In: *Biotechnology in Tropical Crop Improvement*. pp. 43-44. *Proc. International Biotech. Workshop*, 12-15 January 1987, ICRISAT, Patancheru, A.P., India.
- Reddy, P.V., Sudhakar, P., Sujatha, D. and Vijaya Kumar, B. (2003). Drought tolerance traits related to pre harvest aflatoxin contamination in groundnut. Abstracts: 2<sup>nd</sup> International Congress of Plant Physiology, Jan.8-12, 2003, New Delhi, India.
- Zambettakis, C., Waliar, F., Bockelee-Morvan, A. and de Pins, O. (1981). Results of four years research on resistance of groundnut varieties to *Aspergillus flavus*. *Oleagineux* **36**: 377-385.