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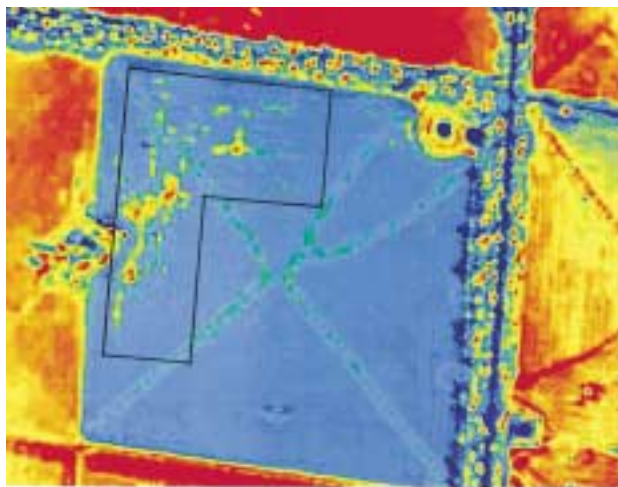
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Management practices to minimise pre-harvest aflatoxin contamination in Australian peanuts

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Abstract. Aflatoxin contamination in peanut kernels is a serious food safety issue throughout the world. Stringent implementation of the international regulatory limits for aflatoxin contamination has become a major factor affecting the economic viability of dryland peanut growers in regional Queensland. In this study, the effect of time of harvesting (digging) and threshing on kernel yield, seed grades, aflatoxin contamination and gross returns were examined with peanut (cv. Streeton), grown in large-scale on-farm trials in the Burnett District of Queensland, during the 1997–98 and 1999–2000 seasons. Aflatoxin contamination was widespread during the 1997–98 season because of a severe and prolonged end-of-season drought and associated elevated soil temperatures. During the 1999–2000 season, aflatoxin risk was low at 2 sites because of well-distributed rainfall and lower soil temperatures, in contrast to the other 2 sites where the risk was higher. In both seasons, early harvest and threshing under high aflatoxin risk conditions resulted in consistently lower aflatoxin concentrations and higher gross returns (up to 27%) than in delayed harvesting treatments. However, under low aflatoxin risk conditions crops could be left longer to realise higher potential yield and better seed grades. Indeed, early harvest under low aflatoxin risk resulted in lower gross returns because of lower yields and poorer seed grades. The current study highlighted the importance of assessing aflatoxin risk on a site-by-site basis in order to make appropriate decisions on timing of harvest so as to minimise aflatoxin contamination and maximise gross returns from dryland peanuts.

Introduction

Aflatoxins are a group of potent carcinogenic compounds produced by *Aspergillus flavus* (Link) and *Aspergillus parasiticus* (Speare) in peanut kernels when environmental conditions during crop maturation are characterised by elevated temperature (up to 35°C) and prolonged moisture deficit (Cole *et al.* 1989). Because of the toxicity and carcinogenicity to humans and livestock, aflatoxins are considered one of the world's major food safety issues and are accordingly closely monitored or regulated (van Egmond 1995). With increased worldwide consumer awareness and international mandatory regulations on aflatoxin contamination in processed peanut foods, there has been growing pressure on the Australian Peanut Industry to ensure delivery of aflatoxin-free products for human consumption.

In Australia, >60% of peanuts are grown under seasonally rainfed conditions in south-eastern Queensland (Fig. 1), with high probability (>50%) of end-of-season drought and aflatoxin risk (Wright and Hansen 1997). Although considerable research was conducted during the 1970s on strategies to manage aflatoxin contamination, uptake of this technology was limited because there were only minor price reductions imposed on aflatoxin-contaminated peanuts (Graham 1982*a*). However, aflatoxin contamination has more recently become a major issue for peanut growers following

the decision by peanut shellers to pass onto growers the real costs associated with removing aflatoxin-positive kernels from contaminated loads, using processes such as blanching and colour sorting. The pricing penalties imposed by the shellers on contaminated loads depend on the aflatoxin concentration in each load above a minimum allowable limit of 8 ppb. Penalties of up to $450/t are now being applied on loads depending on aflatoxin concentrations above 8 ppb. This change in pricing policy has created an urgent need to reassess on-farm aflatoxin risk and develop cost-effective management strategies to minimise aflatoxin contamination in dryland peanuts.

A significant amount of information on aflatoxin incidence and its management in peanut has been generated over the last 3 decades (Harkness *et al.* 1966; Dickens and Khalsa 1967; Mixon 1980; Graham 1982*b*; Coole *et al.* 1989). As a general rule, the greater the period of a crop's exposure to aflatoxin risk factors (i.e. end-of-season drought, elevated soil temperatures, high incidence of soil insects), the greater will be the likelihood of aflatoxin contamination. Control practices such as gypsum application to soil (Davidson *et al.* 1983; Cole *et al.* 1985*a*) or using chemicals on foliage and windrows (Petit *et al.* 1971; Bell and Doupnik 1972; Beuchat *et al.* 1974) have not been successful in reducing aflatoxin contamination under field conditions.

McDonald (1969) highlighted a range of cultural practices, including careful inter-row cultivation practices to avoid damage to pegs and pods, quarantining of dried patches, harvesting the crop at optimum maturity and pre-cleaning to remove immature pods, to minimise aflatoxin contamination. Agronomic practices such as irrigation, timely harvest, pre-cleaning and windrow management have also been shown to be effective in reducing aflatoxin contamination (Cole 1989; Wright and Cruickshank 1999).

Prolonged end-of-season drought not only increases the probability of aflatoxin contamination but can also delay crop maturity. Young *et al.* (1982) observed significant yield losses (up to 40%) following delayed harvest, depending on the variety and crop-growing conditions. Thus, under conditions of prolonged drought and elevated soil temperatures, aflatoxin contamination and harvest losses associated with delayed harvest can be major factors affecting gross returns from rainfed peanut crops. In contrast, under conditions of favourable rainfall and cooler temperature, early harvest can result in poor seed grades, which in turn has a negative impact on gross returns. It is therefore extremely important that aflatoxin risk is assessed for each season in order to adopt appropriate management practices to minimise aflatoxin contamination and maximise gross returns.

Although considerable information is available on the factors contributing to aflatoxin contamination, there has been limited on-farm research to validate the efficacy and economic value of implementing such management practices to minimise aflatoxin and maximise gross returns from dryland peanut crops grown under varied agro-climatic conditions. This study reports the impact of simple management practices such as harvest (digging) and threshing time on kernel yield, seed grades, aflatoxin contamination and gross returns in peanuts grown on commercial-scale farms.

Materials and methods

All experiments were conducted on farmers' fields in the Burnett region of south-eastern Queensland (Fig. 1) during the 1997–98 and 1999–2000 growing seasons (October–May), under rainfed conditions, using the cv. Streeton released by Department of Primary Industries, Kingaroy, for dryland cultivation (Cruickshank *et al.* 2000). The soils are classified as Krasnozems (deep-red clay loam or Oxisol; Soil Survey Staff 1975), with a water-holding capacity of about 140 mm to a depth of 130 cm. After appropriate land preparation, fields were machine-sown with seed obtained from the PCA (Peanut Company of Australia). A spacing of 90 cm between rows and 15 cm between plants within a row was adopted at all sites. The crop was protected from pests and diseases throughout the season following appropriate plant protection measures.
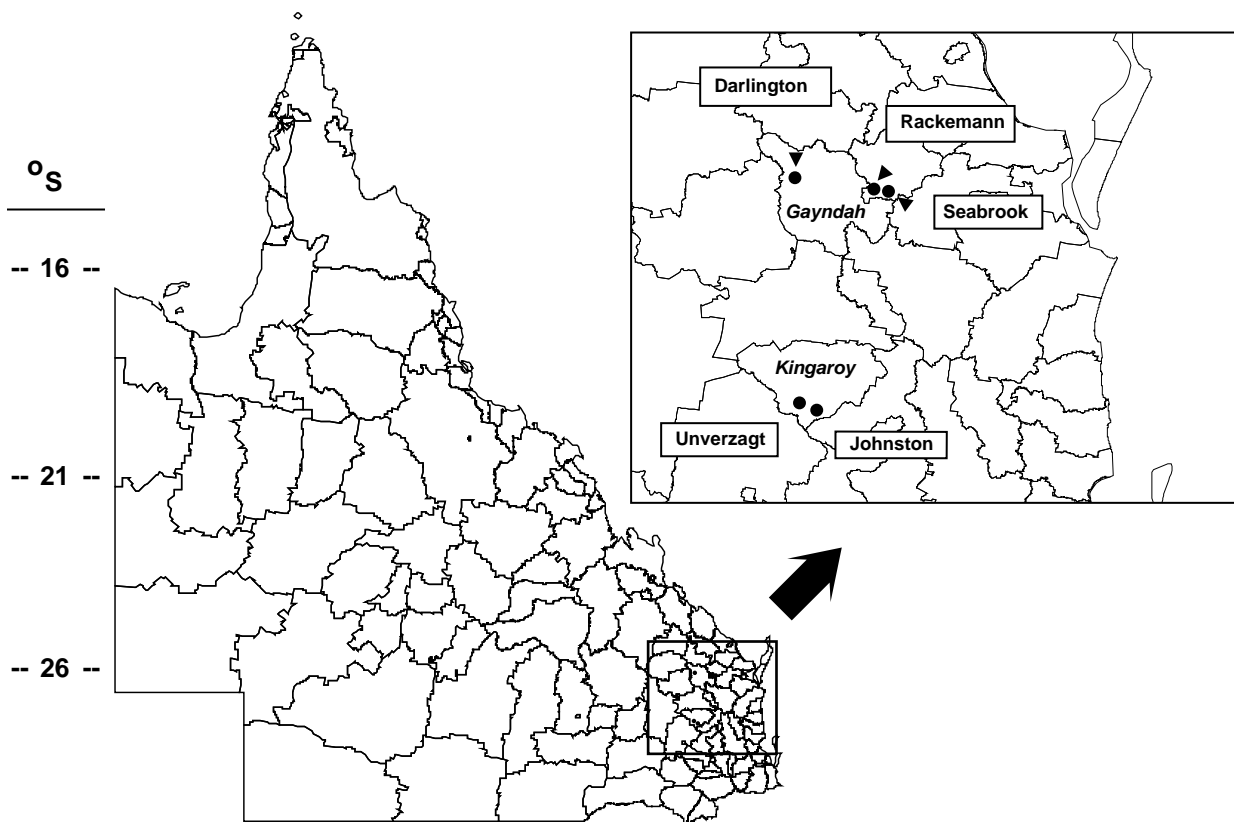

Figure 1. The map of Queensland showing the experimental sites in the South and North Burnett region (inset).

1997–98 season

The effect of harvesting and threshing time on kernel yield, seed grades, aflatoxin contamination and gross returns was examined at 2 locations in the North Burnett (Rackemann) and South Burnett (Johnston) regions of south-eastern Queensland (Fig. 1).

There were 3 harvest (or digging) times (H1, H2 and H3), where H1 is 2 weeks earlier than the farmers' practice; H2 is the farmers' practice and H3 is about 2 weeks later than the farmers' practice. Most farmers follow calendar time and harvest Streeton crops at 20–21 weeks after planting, depending on the growing conditions. However, farmers often leave crops in the ground until 24 weeks after planting, with an aim to maximise yields and seed grades. The harvest times H1 and H3 were designed around H2 in consultation with the growers. At each harvest, 2 threshing times (Thr1 and Thr2) were imposed; with the crop being threshed either 6 days (Thr1) or 10–14 days (Thr 2) after harvest. The experiments were arranged as a split-plot design with harvest treatments as main plots and threshing times as subplots, with 4 replications. The dates of planting, harvest and threshing for the 2 experimental sites are presented in Table 1. The plot size varied between sites from 4 rows (spaced at 0.9 m intervals) 12 m long each (42.2 m^2) to 4 rows 25 m long each (90 m^2). At each harvest time, the plot area was measured and the crop pulled using a mechanical digger and arranged into windrows. Plants were allowed to dry out in windrows until threshing using a small plot thrasher. Pods were collected in bags and dried in a bed drier at 35°C for at least 4 days before passing through a pre-cleaner to remove any extraneous matter and immature and shrivelled pods. The pre-cleaned pods were passed over a peanut belt screen and the pods with visual damage (growth cracks, soil insect damage) were handpicked and separated. Weights of the clean and damaged pod samples were recorded.

Kernel-grade analysis

Kernel grades were determined from a 1 kg pod sample randomly drawn from the pre-cleaned yield sample. Pods were hulled in a mechanical huller and passed through series of perforated (round holes) screens mounted on a modified Model 4 mechanical grader (Kingaroy Engineering Works), which separated kernels in to 3 different kernel grades. The 3 kernel grades used were Jumbos and 1s (riding over an 11 mm screen), 2s and 4s (passing through a <11 mm and riding over >9 mm screen), and oils (passing through an 8 mm screen). The kernel grades are presented as a percentage of kernel weight of each grade in the total sample weight.

Table 1. Timing of harvest and threshing treatments implemented at Johnston and Rackemann sites during the 1997–98 growing season

DAS, days after sowing; DAH, days after harvest

	Johnston	Rackemann
Date of sowing	1.xi.97	26.xi.97
Harvest 1 (DAS)	139	125
Thresh 1 (DAH)	4	6
Thresh 2 (DAH)	7	14
Harvest 2 (DAS)	152	139
Thresh 1 (DAH)	5	6
Thresh 2 (DAH)	10	10
Harvest 3 (DAS)	170	153
Thresh 1 (DAH)	4	5
Thresh 2 (DAH)	7	14

Table 2. Price and grading schedule at different aflatoxin concentrations for Streeton crop during the 1997–98 season

Seg1–4 indicate the range of aflatoxin concentrations

Kernel grading	Price ($/t kernel weight)			
	(<8 ppb) Seg1	(8–80 ppb) Seg2	(80–400 ppb) Seg3	(>400 ppb) Seg4
J +1	1225	1195	1125	650
2, 4	1100	1050	700	500
Oils + damaged	200	200	200	200

Aflatoxin analysis

Aflatoxin content was analysed in each kernel-grade sample as well as in the damaged kernel fraction. The weight of each kernel grade was recorded and aflatoxin in kernels extracted by blending the sample in 80% methanol in a ratio of 1:2 (sample size to methanol volume). The extract was passed through mini-column and concentration of the toxins was assessed by visually comparing the intensity of glow in the mini-column with a set of other columns containing standards with known aflatoxin concentration, under an UV lamp (Holaday and Passwater 1969). The aflatoxin content in total edible kernels was calculated as a weighted average using the actual weight of the kernels contributed by each grade.

Calculation of gross returns

Gross return from crop produce was calculated by applying the farm gate price and grading schedule adopted by the PCA in the 1997–98 season to the yield sample harvested from the experimental area (Table 2).

Weather data

Daily solar radiation, air temperature and rainfall data for each site was accessed from the 'data drill' (http://www.dnr.qld.gov.au/silo) website by providing the latitude and longitude of each farm, recorded using a ground positioning system (GPS) unit.

1999–2000 season

Trials were conducted at 4 sites on farmers' fields in the North (Darlington, Rackemann, Seabrook) and South (Unverzagt) Burnett regions of south-eastern Queensland. Crop management practices were followed as described for the 1997–98 season. However, there were only 2 harvest times, H1 and H2, where H1 is equal to about 2 weeks earlier than the farmers' practice and H2 is the farmers' practice. The treatments were replicated 4 times. Crops were threshed within 5 days of the harvest, using a small plot thrasher. There were no threshing time treatments. The sowing and harvest dates for the 1999–2000 season are presented in Table 3. Yield samples were processed as described for 1997–98 season. After pre-cleaning the yield sample, damaged pods

Table 3. Timing of harvests implemented at Darlington (DLT), Rackemann (RCK), Seabrook (SBK) and Unverzagt (UNV) sites during the 1999–2000 growing season

DAS, days after sowing

	DLT	RCK	SBK	UNV
Date of sowing	4.xi.99	16.xi.99	13.xi.99	18.x.99
Harvest 1 (DAS)	131	134	137	150
Harvest 2 (DAS)	145	147	150	162

Table 4.   Price and grading schedule at different aflatoxin concentrations for Streeton crop during the 1999–2000 season

Seg1–4 indicate the range of aflatoxin concentrations

Kernel grading	Price ($/t kernel weight)			
	(<8 ppb) Seg1	(8–80 ppb) Seg2	(80–400 ppb) Seg3	(>400 ppb) Seg4
Jumbos	1275	1000	850	600
Grade 1	1200	1000	800	450
Grade 2	1020	800	600	350
Manufacturing	400	200	100	100
Splits	700	500	300	200
Oils	150	150	100	100

were separated and weight of the damaged and clean pods determined before processing the clean pods for kernel-grade analysis.

Kernel-grade analysis

The procedure of kernel-grade analysis was modified to replicate the local industry standards, which involved grading the seeds into 6 categories, i.e. Jumbos (riding over an 11.51 mm screen), grade 1 (passing through 11.51 mm and riding over a 9 mm screen), grade 2 (passing through 9.92 mm and riding over a 8.73 mm screen), manufacturing (MFG) (passing through 8.73 mm and riding through a 6.75 mm screen), Splits (>6.75 mm) and oils (passing through a 6.74 mm screen).

Aflatoxin analysis

Aflatoxin content was analysed in the edible kernels (combination of Jumbos, 1s, 2s, MFGs, splits and oils) and damaged fraction of the yield sample, using the affinity column method (Truckess *et al.* 1991). Sample size and aflatoxin extraction procedures were similar to those described for the 1997–98 season.

Calculation of gross returns

Gross returns were calculated using the farm gate prices and grading schedule as given in Table 4.

Weather data

Automatic data loggers with 2 temperature sensors (Gemini data loggers, TinyTag) were installed at each site to monitor ambient air temperature at a height of 1 m above the crop canopy and soil temperature in the pod zone (5–7 cm depth), at hourly intervals throughout the season. Daily solar radiation and rainfall data for each site was accessed from the 'data drill' website.

Soil water balance analysis

Changes in plant-extractable soil water (PESW) at each site were computed for the 2 seasons, using the APSIM peanut model (Hammer *et al.* 1995).

Results

Weather during the 1997–98 season

A summary of mean air temperatures and rainfall during the pod-filling period as well as total rainfall during the 1997–98 season at the Johnston and Rackemann sites are presented in Table 5. The mean daily air temperatures during the pod-filling period at both sites were about 26°C up until H1. Although the mean daily temperature declined to about 23°C by H3, the diurnal fluctuation was ± 11°C around the mean. The Johnston site received a total of 377, 399 and 452 mm of rainfall by H1, H2 and H3, respectively, of which 60% fell during the seed-filling period. There was also some post-harvest rainfall, with 22 mm falling on windrows between H1 and H2, and a further 53 mm falling between H2 and H3. The Rackemann site received a total of 137, 150 and 252 mm of rainfall by H1, H2 and H3, respectively. However, in contrast to the Johnston site, the crop at the Rackemann site received <35% of the total rainfall during the seed-filling period, which resulted in a severe and prolonged end-of-season drought until H2 was harvested. There were, however, significant post-harvest rainfall events of 13 mm between H1 and H2, and 102 mm between H2 and H3, which interfered with harvest (for H3) and threshing operations for H2.

Seasonal changes in plant-extractable soil water

Changes in PESW, simulated using the APSIM peanut model, showed that at the Rackemann site PESW dropped continuously from 35 DAS until 140 DAS, resulting in severe and prolonged drought for a 115-day period (Fig. 2*a*). However, the Rackemann site received a total of 13 mm of rain between 125 (H1) and 139 (H2) DAS (Table 5), in 3 events of 3–5 mm. Because of high evaporative demand during the period, these rains were not sufficient enough to result in any change in the PESW. At the Johnston site, the crop experienced short mid-season drought episodes although there was a 55-day drought period between 115 and 160 DAS. At both sites, post-harvest rainfall occurred which interfered with the harvest and threshing operations.

Weather during the 1999–2000 season

The Darlington and Unverzagt sites received total rainfalls of 333 and 428 mm, respectively, compared with

Table 5.   Mean air temperatures and rainfall from the start of pod-filling until the three-harvest timings and total seasonal rainfall during the 1997–98 season at the Johnston and Rackmann sites

DAS, days after sowing

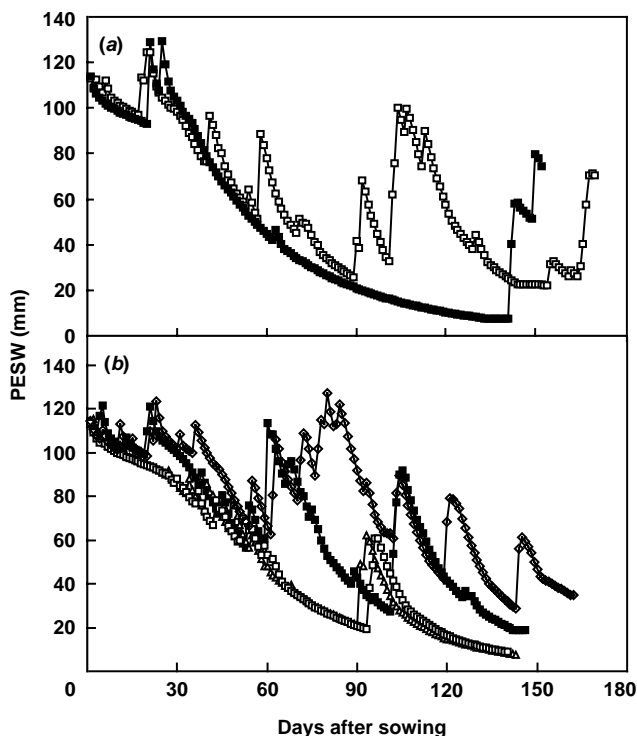| Parameter	Johnston			Rackemann		
	Harvest 1 (139 DAS)	Harvest 2 (153 DAS)	Harvest 3 (170 DAS)	Harvest 1 (125 DAS)	Harvest 2 (139 DAS)	Harvest 3 (153 DAS
Air temperature (°C)	25.5 ± 9.4	24.5 ± 10.6	22.5 ± 11.4	26 ± 10	24 ± 10	23 ± 11
Rainfall (mm)	214	236	289	43	56	158
Total rainfall (mm)	377	399	452	137	150	252

Figure 2. Seasonal changes in the plant extractable water (PESW) at (*a*) the Rackemann (RCK) and Johnston (JHN) sites during the 1997–98 growing season and (*b*) the Darlington (DLT), Unverzagt (UNV), Rackemann (RCK) and Seabrook (SBK) sites during the 1999–2000 growing season.

141 and 148 mm at the Rackemann and Seabrook sites, respectively (Table 6). Although about 50% of the total seasonal rainfall fell during the pod-filling period at all sites, lower rainfall amounts at the Rackemann and Seabrook sites resulted in a severe and prolonged end-of-season drought. Mean daily air temperatures during the pod-filling period ranged from 23 to 25°C by H1 across the 4 locations (Table 6). However, the Rackemann and Seabrook sites were characterised by higher mean soil temperatures during both harvest times (>26°C) than those recorded at the Darlington and Unverzagt sites (about 23°C).

Seasonal changes in plant-extractable soil water

Analysis of plant-extractable soil water simulated using the APSIM peanut model showed that all sites experienced end-of-season drought, although the intensity of drought varied substantially across sites (Fig. 2*b*). The Rackemann and Seabrook sites experienced prolonged drought from 90 DAP until final harvest, as evidenced by consistently lower PESW throughout the season. Well-distributed rainfall at the Unverzagt site resulted in relatively high levels of plant-extractable soil water throughout the season. The Darlington site experienced episodes of mid- and end-of-season drought; however, it was not as severe as that experienced at the Rackemann and Seabrook sites, owing to higher rainfall.

In summary, during the 1999–2000 season, the Rackemann and Seabrook sites were characterised by end-of-season drought and warm temperatures, while the Darlington and Unverzagt sites maintained relatively high levels of available soil moisture during the pod-filling period. In order to examine the environmental effects on aflatoxin contamination, the trial sites were grouped into high aflatoxin risk (HAR) (Rackemann and Seabrook) and low aflatoxin risk (LAR) (Unverzagt and Darlington) sites.

Effect of time of harvest and threshing on yield, seed grades, aflatoxin and gross returns during the 1997–98 season

At the Rackemann site, the timing of harvesting had a significant effect on kernel yield, with H2 and H3 resulting in yield losses of 12 and 56%, respectively, compared with the early harvest (H1) (Table 7). The late threshing (Thr2) also resulted in a significant loss of kernel yield for each of the harvest treatments (35% in H1 and H2 and 20% in H3). An analysis of kernel-grade composition from the Rackemann site revealed that the proportion of Jumbos increased from 15% in H1 to 25% in H2, although this declined back to 15% in H3 (Fig. 3). While the proportion of 2 + 4s grade kernels varied from 62 to 66% across harvests, the proportion of oil-grade kernels was high in H1 (17%), compared with only 8% in the H2 and H3. The proportion of damaged kernels increased with delay in harvest.

Table 6. Mean air and soil temperatures, and rainfall from the start of pod filling until the three-harvest timings and total seasonal rainfall during the 1999–2000 season at low and high aflatoxin risk sites

Parameter	Low aflatoxin risk				High aflatoxin risk			
	Darlington		Unverzagt		Rackemann		Seabrook	
	Harvest 1 (131 DAS)	Harvest 2 (145 DAS)	Harvest 1 (150 DAS)	Harvest 2 (162 DAS)	Harvest 1 (134 DAS)	Harvest 2 (147 DAS)	Harvest 1 (137 DAS)	Harvest 2 (147 DAS)
Air temperature (°C)	25 ± 10.4	24.9 ± 10.5	23.4 ± 9.5	23.1 ± 9.5	24.2 ± 11.7	22.2 ± 8.4	24.8 ± 12.1	22.8 ± 12.1
Soil temperature (°C)	22.8 ± 4.8	22.7 ± 4.6	23.2 ± 7.9	23 ± 7.9	26.5 ± 8.6	26 ± 8.5	26.1 ± 8.5	26.5 ± 8.5
Rainfall (mm)	147	147	212	212	71	71	74	74
Total rainfall (mm)	333	333	428	428	141	141	148	148

Table 7. Effect of timing of harvesting and threshing on kernel yield, aflatoxin contamination and gross returns at the Rackemann and Johnston sites during the 1997–98 growing season

	Kernel yield (kg/ha)		Aflatoxin conc. (ppb)		Gross return ($/ha)	
	Thr 1	Thr 2	Thr 1	Thr 2	Thr 1	Thr 2
Rackemann site						
Harvest 1	1578	1016	132	1002	1311	688
Harvest 2	1389	898	333	860	843	488
Harvest 3	692	560	796	1090	449	604
l.s.d. (*P* = 0.05)						
Harvest	108.0		434.3		359.7	
Thresh	171.7		573.9		178.4	
Johnston site						
Harvest 1	1830	1345	142	344	2913	2212
Harvest 2	1569	1353	536	686	1778	1521
Harvest 3	1515	793	235	655	1375	833
l.s.d. (*P* = 0.05)						
Harvest	180.5		170.4		857.1	
Thresh	305.8		176.1		462.0	

Timing of harvest at the Rackemann site had a significant effect on aflatoxin contamination, with aflatoxin concentration increasing linearly from 132 ppb in H1 up to 796 ppb in the H3. Also, the late threshing (Thr2) resulted in significantly higher aflatoxin concentration at each harvest (Table 7).

At the Rackemann site, gross returns were the highest ($1311/ha) with the earliest harvest (H1) in combination with short harvest to threshing intervals (Thr1). A combination of yield losses and high aflatoxin contamination in H2 and H3 resulted in a linear reduction in gross returns from $1311/ha in H1 to $843/ha in H2 and $449/ha in H3. Delayed threshing in H1 and H2 resulted in even further reductions (up to 50%) in gross returns. However, in H3 gross returns were low ($449/ha at Thr1 and $604/ha at Thr2) and differences between Thr 1 and Thr 2 were marginal and not significant (Table 7).
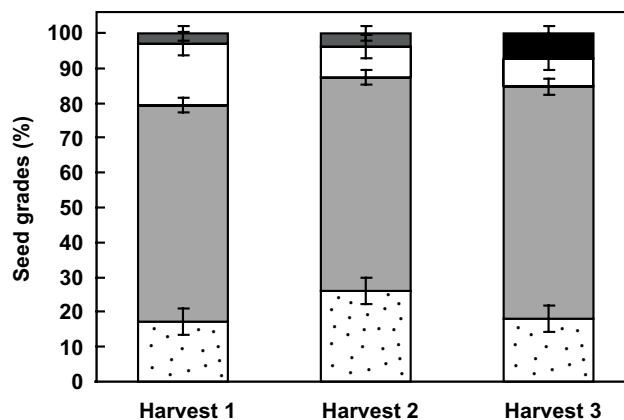
Although the Johnston site recorded greater yields than the Rackemann site because of a higher amount and better distribution of rainfall, the effect of timing of harvest and threshing on yield losses, aflatoxin contamination and gross returns were relatively similar (Table 7). For example, at the Johnston site kernel yield declined from 1830 kg/ha in H1 to 1569 kg/ha and 1515 kg/ha in H2 and H3, respectively. Late threshing (Thr2) in H1 further reduced yields from 1830 kg/ha to 1345 kg/ha. The combination of delayed harvest (H3) and late threshing (Thr2) resulted in the lowest harvestable kernel yield (793 kg/ha). Also results for kernel grades at different harvesting and threshing times at the Johnston site were similar to those at the Rackemann site (Fig. 3) (data not shown).

Severe end-of-season drought at the Johnston site resulted in very high levels of aflatoxin at all harvest and threshing time treatments (Table 7). Aflatoxin contamination was the lowest (142 ppb) in Thr1 of H1 and increased in later harvests (536 ppb and 235 ppb in H2 and H3, respectively). In a similar response as for the Rackemann site, late threshing (Thr2) at the Johnston site resulted in significantly increased aflatoxin concentrations at each harvest.

The combination of reduced yield and increased aflatoxin contamination resulted in a linear decline in gross returns from $2913 in H1 and Thr1 to $1778 and $1375/ha in H2 and H3, respectively. As observed at the Rackemann site, delayed threshing at each harvest resulted in further reductions in gross returns at the Johnston site.

Effect of time of harvest on yields, aflatoxin and gross returns during the 1999–2000 season

The effect of timing of harvest on yield and aflatoxin contamination varied across locations (Table 8). At the Darlington site, there was a significant yield benefit of 620 kg/ha in H2 over H1. At the Rackemann, Seabrook and

Figure 3. Seed grade profiles (stippled bars, J +1s; shaded bars, 2 + 4s; open bars, oils; solid bars, damaged) at three harvesting timings at the Rackemann site during the 1997–98 growing season.

Table 8. Effect of time of harvesting on kernel yield, aflatoxin contamination and gross return from peanuts grown at low (LAR) and high (HAR) aflatoxin risk sites during the 1999–2000 season

Values followed by different letters differ significantly between the treatments at a given site

Location (aflatoxin risk)	Kernel yield (kg/ha)		Aflatoxin conc. (ppb)		Gross return ($/ha)	
	Harvest 1	Harvest 2	Harvest 1	Harvest 2	Harvest 1	Harvest 2
Darlington (LAR)	2398a	3021b	1a	3a	2158a	2497b
Unverzagt (LAR)	1754a	1755a	0a	0a	1542a	1570a
Rackemann (HAR)	1197a	1243a	10a	64a	938a	831a
Seabrook (HAR)	1019a	1043a	0a	226b	835a	655b

Unverzagt sites, however, the yield differences between H1 and H2 were marginal and non-significant.

In general, aflatoxin contamination was lower during the 1999–2000 season than the 1997–98 season. There was negligible aflatoxin contamination (0 to <3 ppb) at the LAR Darlington and Unverzagt sites. However, substantial aflatoxin contamination was recorded at the other 2 HAR sites, with the concentration increasing from 10 ppb in H1 to 64 ppb in H2 at the Rackemann site, and from 0 in H1 to 226 ppb in H2 at the Seabrook site.

At the LAR sites, normal timing of harvest (H2) resulted in an increase in gross returns (15% at Darlington and 2% at Unverzagt), compared with the early harvest (H1). In contrast, there was a 12 and 27% reduction in gross returns in the H2, compared with H1 at the HAR Rackemann and Seabrook sites, respectively.

Aflatoxin contamination in different kernel grades during the 1997–98 and 1999–2000 seasons

During the 1997–98 season, aflatoxin contamination was determined in each of the kernel grades, including J +1s, 2 + 4s, oils and damaged kernel, at each of the harvest and threshing times (data not shown). However, in the 1999–2000 season, aflatoxin was determined only in the edible (pooled over J +1s, 2 + 4s, splits and oils) and damaged kernel fractions. To enable easier comparison across treatments and seasons, aflatoxin data are presented for the edible and damaged kernel fractions at different timings of harvest for the 1997–98 (Fig. 4) and 1999–2000 (Fig. 5) seasons. At the Rackemann site in the 1997–98 season, both edible and damaged kernels were contaminated but the aflatoxin concentrations were substantially greater in damaged kernels than in edible-grade kernels at each of the 3 harvests (Fig. 4a–c). In H1, edible-grade kernels contained considerable amounts of aflatoxin although the level increased significantly from 150 ppb in Thr1 to >400 ppb in Thr 2 (Fig. 4a). In H2, aflatoxin in edible-grade kernels increased to >800 ppb at both threshing times, while the damaged kernels maintained even higher levels of aflatoxin contamination (Fig. 4b). In H3, aflatoxin was present in considerable concentration in edible-grade kernels but the concentrations were lower (<400 ppb) than in H2. Damaged

kernels, however, maintained very high concentrations of between 1000 and 1500 ppb (Fig. 4c). Similar patterns of aflatoxin distribution among kernel grades were also observed at the Johnston site during the 1997–98 season (data not shown).

During the 1999–2000 season, aflatoxin contamination was generally lower than during the 1997–98 season. At the HAR Rackeman site, there was negligible aflatoxin in edible kernels in H1; however, concentration increased significantly to >50 ppb in H2 (Table 8). Further analysis of aflatoxin in kernel grades showed that the damaged kernel fraction contained significantly higher concentrations of aflatoxin than the edible kernel fraction. Damaged kernels contained considerable concentration of aflatoxin (>150 ppb) at early harvest (H1), and the concentration further increased to >500 ppb in H2 (Fig. 5a). Similar patterns of aflatoxin distribution among kernel grades were also observed at the other HAR Seabrook site.

At the LAR Unverzagt site, there was no aflatoxin contamination in edible kernels in either H1 or H2 (Fig. 5b). However, damaged kernels contained traces of aflatoxin (<3 ppb) in H1 or H2. Similar results on aflatoxin contamination were observed at the other LAR Darlington site.

Discussion

A number of studies have shown that prolonged end-of-season drought coupled with elevated soil temperatures in the 25–30°C range are required for pre-harvest infection by the *Aspergillus flavus* and *A. parasiticus* fungi and subsequent aflatoxin production (Graham 1982a, 1982b; Hill *et al.* 1983; Cole *et al.* 1989). The differences in the intensity of aflatoxin contamination between the 1997–98 and 1999–2000 seasons could largely be attributed to the variability in the intensity and duration of drought as well as soil temperatures (Fig. 2a and b; Tables 5 and 6). During 1997–98, the severe aflatoxin contamination, including that in edible-seed grades (Fig. 4), could be attributed to prolonged drought periods (>45 days at Johnston and >90 days at Rackemann, Fig. 2a), along with elevated soil temperatures (Table 5). Sanders *et al.* (1985) showed that a drought of more than 20 days but less than

30 days, with optimum soil temperatures for aflatoxin production (25–30°C), was necessary to cause aflatoxin contamination in all kernel grades. Soil temperatures were not measured at the experimental sites during the 1997–98 season. However, extrapolation of the soil temperatures recorded from a nearby weather station suggested that mean daily soil temperatures at the Johnston site would have been in the range of 26–28°C during most of the seed-filling period during the 1997–98 season (data not shown). It is expected that the soil temperatures at the Rackemann site would have been at least comparable, if not more than those measured at the weather station near the Johnston site. It was also possible that the rapid decline in PESW throughout the

podding stage at the Rackemann site (Fig. 2*a*) would have resulted in a progressive decline in kernel moisture and consequent invasion of pegs and developing pods by *Aspergillus flavus*, as observed by Cole *et al.* (1985*b*). Although PESW at the Johnston site indicated better water availability than at the Rackemann site (Fig. 2*a*), high evaporative demand (>6 mm/day) during the pegging and pod-filling period would have resulted in severe water deficits in crops, as evidenced by the rapidly declining PESW before and after rainfall events. However, prolonged crop water deficits during the 1997–98 season could have resulted in reductions in kernel water activity (below an Aw of 0.97) and breakdown of the associated natural defence mechanisms (Wotton and Strange 1985), thus rendering the peanut plant vulnerable to infection by the *Aspergillus* fungi (Dorner *et al.* 1989).

Aflatoxin incidence was generally low in the 1999–2000 season, although the levels of contamination varied across sites (Table 8). The negligible aflatoxin contamination at LAR sites could be largely explained by maintenance of high kernel water activity (as a result of high PESW) (Fig. 2*b*), along with the occurrence of low soil temperatures (<2°C) (Table 6). The results from the current study therefore support the central hypothesis that elevated soil temperatures in addition to late-season drought are necessary for the production of aflatoxin in peanut kernels (Blankenship *et al.* 1984; Cole *et al.* 1985*b*).

In spite of widespread aflatoxin contamination during the 1997–98 season, aflatoxin concentrations were significantly lower with the early harvest (H1) than with the later harvest (Table 7). However, aflatoxin concentrations of >130 ppb at the earliest harvest (H1) could still be attributed to declining
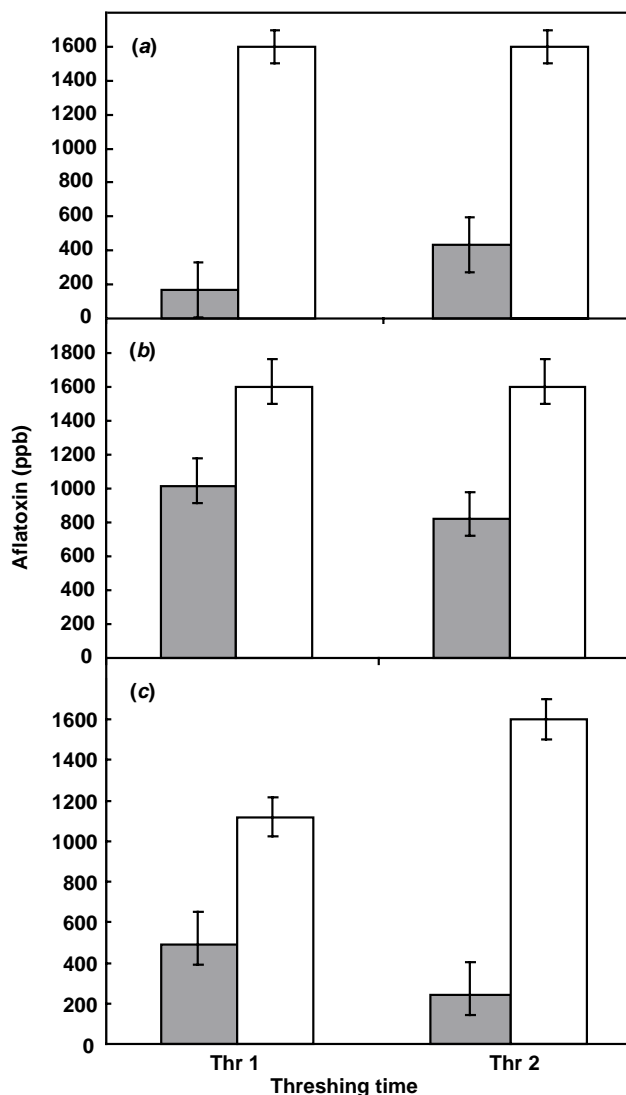

Figure 4. Aflatoxin concentrations in edible (shaded bars) and damaged kernels (open bars) measured in the two threshing time treatments, i.e. 7 days (Thr 1) and 14 days (Thr 2) after (*a*) harvest 1 (125 DAS), (*b*) harvest 2 (139 DAS) and (*c*) harvest 3 (153 DAS) at the Rackemann site during the 1997–98 growing season.
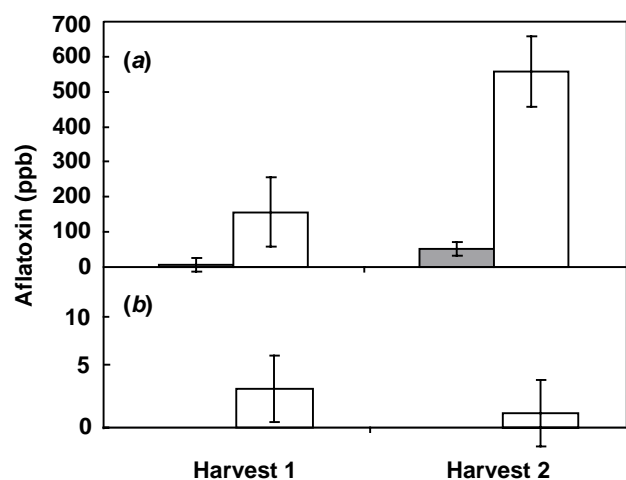

Figure 5. Aflatoxin concentrations in the edible (shaded bars) and damaged kernels (open bars) measured at two harvests, i.e. harvest 1 (131 DAS) and harvest 2 (145 DAS) at (*a*) high aflatoxin risk (Rackemann) site and (*b*) low aflatoxin risk (Unverzagt) site during the 1999–2000 growing season.

kernel moisture and consequent infection of developing pods by *Aspergillus* fungi, leading to aflatoxin production under continuing drought and high temperature. As observed by Cole *et al.* (1985*b*), longer exposure of developing pods to high aflatoxin risk conditions (low kernel water activity and high soil temperatures) led to increased aflatoxin concentrations at the later harvests (H2 and H3), at both Rackemann and Johnston sites (Table 7). The current study highlighted also the importance of the time interval between harvest (digging) and threshing operations in minimising aflatoxin risk (Table 7). At both the Rackemann and Johnston sites, aflatoxin concentrations were consistently higher at later threshing times with each of the harvest times (Table 7), suggesting the critical role of windrow configuration in post-harvest aflatoxin contamination. Under conditions where plants are poorly inverted in the windrow, pods covered by foliage are exposed to high relative humidity and warm ambient temperature conditions, which are conducive to growth of *Aspergillus flavus* and *A. parasiticus*, as well as for aflatoxin production (Diener and Davies 1967). Post-harvest rainfall at both Johnston and Rackemann sites (Table 5) would have also played a key role in delaying threshing operations, and thus prolonging the conducive conditions for fungal growth and aflatoxin production in windrows.

During the 1999–2000 season, aflatoxin contamination was generally low and confined to H2 at the HAR sites (Table 8). It was apparent that the period of high aflatoxin risk, characterised by a combination of drought and elevated soil temperatures, was insufficient for aflatoxin production at the early harvest (H1), but sufficient to result in aflatoxin production in H2 at the HAR sites (Cole *et al.* 1985*b*).

During the 1997–98 growing season, both the Rackemann and Johnston sites recorded a linear decline in kernel yield with delay in harvest and threshing operations (Table 7). The lower yields in H2 and H3 (than in H1) could be a result of harvest losses at digging as well as during threshing operations at each of the harvest times. Detachment of fully mature pods from the plant due to weakening of peg strength is a common occurrence when digging and threshing operations are delayed. Harvest losses of up to 40% (depending on the variety and growing conditions) have been reported with delay of harvest in peanut (Young *et al.* 1982).

During the 1999–2000 season, kernel yields were higher at LAR sites than at HAR sites (Table 8) because of the relatively higher levels of PESW throughout the season (Fig. 2*b*). At the LAR Unverzagt site, seasonal temperatures were 1–2°C lower, which prolonged the duration of the crop to >150 days (Table 6). At this site, the early harvest (H1) did not affect kernel yield (Table 8). However, at LAR Darlington site, where temperatures were higher, the early harvest (H1) resulted in lower yields and seed grades than the normal harvest (H2) (Table 8). The lower yields in the H1 treatment

at this site could be attributed to immaturity in the crop at the time of harvest. Several studies have shown significant losses in yield and seed grades resulting from too early or too late harvest of the crop, depending on the seasonal conditions (Young *et al.* 1982). A remarkable result from this study was that at HAR sites, early harvest had no significant effect on kernel yield but a major effect in reducing aflatoxin contamination (Table 8).

Gross returns from a peanut crop in Australia are determined by the price, kernel yield, seed grades and aflatoxin contamination in the produce. Results from the 1997–98 season showed that a combination of early harvest and early threshing (H1 and Thr1) resulted in the highest gross returns at both the sites (Table 7). Delayed threshing (Thr 2) at each harvest time resulted in significantly reduced gross returns at both sites because of either greater harvest losses or high aflatoxin contamination, or both. It was also of interest that gross returns at H1 were higher, even though seed grades were poorer than at the normal harvest time (H2) (Fig. 3). The higher gross returns at H1 than H2 implied that the combined effect of yield losses and aflatoxin contamination overrode any benefit of improved seed grades at H2.

It was also apparent that delayed harvest under high aflatoxin risk situations can result in a higher proportion of damaged pods (Fig. 3), which are known to contain high concentrations of aflatoxin. Indeed in both the seasons, damaged pods were potential sources of aflatoxin (Figs 4 and 5) as observed by earlier workers (McDonald 1969; Garaham 1982*b*; Cole *et al.* 1989).

The differences in aflatoxin contamination and gross returns among experimental sites during the 1999–2000 season also highlight the importance of monitoring aflatoxin risk during the season in order to maximise gross returns from dryland peanuts in Australia. Higher gross returns at LAR than HAR sites could be attributed to higher yields, better seed grades and low or negligible aflatoxin contamination. At HAR sites, gross returns were greater in H1 (by 13% at Rackemann and 27% at Seabrook) than in H2. These results reconfirmed findings from the 1997–98 experiments, which showed that under high aflatoxin risk situations, gross returns can be maximised by harvesting the crop earlier than the normal time. In contrast, at LAR sites, gross returns were greater from the normal harvest (H2) where yield and seed grades were maximised. For example, at the Darlington site, a significant increase in kernel yield (26%) and negligible aflatoxin contamination in H2 resulted in a gross return benefit of $339/ha over H1. However, at the Unverzagt site, yield and gross returns were similar in H1 and H2. At this site, lower night temperatures (<15°C) resulted in not only an extended growing period (Table 6) but also very low (almost zero) aflatoxin contamination (Table 8).

The conventional practice for dryland peanut cultivation in Australia has been to leave the crop in ground as long as possible with the aim to maximise yields and thus gross returns. Results from the current studies question this practice under all conditions. With the introduction of pricing penalties for aflatoxin-contaminated product, the decision of when to harvest and thresh the crop becomes critical in order to maximise gross returns.

Our results are in agreement with a multitude of previously published research and provide on-farm evidence of the value of simple agronomic practices such as timely harvest and threshing in minimising aflatoxin contamination in peanuts in high-risk years. The future challenge will be to better assess, and ultimately predict, aflatoxin risk during the season in order to make the most appropriate decision on timing of harvest to maximise gross returns.

Conclusions

The results from the current study showed that both late season drought and elevated soil temperatures are necessary for aflatoxin contamination to occur. Under conditions where aflatoxin risk is high, early harvesting and threshing practices should be followed to minimise aflatoxin contamination and maximise gross returns for peanuts produced under rain-fed conditions in Australia. However, under low aflatoxin risk conditions, crops can be harvested at the optimum time in order to realise potential yield, grades and hence gross returns. The study has also demonstrated the value of assessing the aflatoxin risk on a site-by-site basis in order to make appropriate harvesting management decisions.

Acknowledgments

References

Bell DK, Doupnik B (1972) Chemicals in the windrow for controlling aflatoxins in peanuts. *Proceedings of the American Peanut Research and Education Association* **4**, 18–20.

Beuchat LR, Smith DH, Young CT (1974) Effect of foliar fungicides on aflatoxin, oil and protein content and maturing rate of peanut kernels. *Journal of the Science of Food and Agriculture* **25**, 477–482.

Blankenship PD, Cole RJ, Sanders TH, Hill RA (1984) Effect of geocarposphere temperature on pre-harvest colonization of drought-stressed peanuts by *Aspergillus flavus* and subsequent aflatoxin contamination. *Mycopathologia* **85**, 69–74.

Cole RJ (1989) Preharvest aflatoxin in peanuts. *International Biodeterioration* **25**, 253–257.

Cole RJ, Csinos AS, Blankenship PD, Sanders TH, Gaines TP, Davidson JI Jr (1985*a*) Evaluation of soil calcium as method of prevention of preharvest aflatoxin contamination in peanuts. *Proceedings of the American Peanut Research and Education Society* **17**, 71.

Cole RJ, Sanders TH, Hill RA, Blankenship PD (1985*b*) Mean geocarposphere temperatures that induce aflatoxin contamination of peanuts under drought stress *Mycopathologia* **91**, 41–46.

Cole RJ, Sanders TH, Dorner JW, Blankenship PD (1989) Environmental conditions required to induce preharvest aflatoxin contamination of groundnuts summary of six years' research. In 'Proceedings of the international workshop on aflatoxin contamination of groundnut'. ICRISAT 1987. (Ed. SD Hall) pp. 279–287. (ICRISAT Center: Patancheru, India)

Cruickshank AL, Wright GC, Nageswara Rao Rachaputi (2000) 'Streeton' — an aflatoxin tolerant peanut cultivar for the Australian peanut industry. *Proceedings of the American Peanut Research and Education Society* **32**, 27

Davidson JI Jr, Blankenshipm PD, Sanders TH, Cole RJ, Hill RA, Henning RJ, Guerke WR (1983) Effect of row spacing, row orientation and gypsum application on production and quality of non-irrigated Flourunner peanuts. *Proceedings of the American Peanut Research and Education Society* **17**, 71.

Dickens JW, Khalsa JS (1967) Windrow orientation and harvesting damage to peanuts. *Oleagineux* **22**, 741–746.

Diener UL, Davis ND (1967) Limiting temperature and relative humidity for growth and production of aflatoxin and free fatty acids by *Apsergillus flavus* in sterile peanuts. *Journal of American Oil Chemists' Society* **44**, 259–263.

Dorner JW, Cole RJ, Sanders TH, Blakenship PD (1989) Interrelationship of kernel water activity, soil temperature, maturity, and phytoalexin production in preharvest aflatoxin contamination of drought-stressed peanuts. *Mycopathologia* **105**, 117–128.

Graham J (1982*a*) Aflatoxins in peanuts: occurrence and control. *Queensland Agricultural Journal* **108**, 119–122.

Graham J (1982*b*) The occurrence of aflatoxin in peanuts in relation to soil type and pod splitting. *Food Technology in Australia* **34**, 208–212

Hammer GL, Sinclair TR, Wright GC, Meinke H, Bell MJ (1995) A peanut simulation model. I. Model development and testing. *Agronomy Journal* **87**, 1085–1093.

Harkness C, McDonald D, Stonebridge WC, A'Brook J, Darling HS (1966) The harvesting, handling and storage of crops in relation to development of toxic fungi, with special relation to groundnut production in Tropical Africa. *Food Technology* **20**, 72–78.

Hill RA, Blankenship PD, Cole RJ, Sanders TH (1983) Effects of soil moisture and temperature on preharvest invasion of peanuts by *Aspergillus flavus* group and subsequent aflatoxin development. *Applied Environmental Microbiology* **45**, 628–633.

Holaday CE, Passwater RA (1969) Minicolumn chromatography and fluorometry. *Fluorescence News* **4** (2).

McDonald D (1969) *Aspergillus flavus* on groundnut (*Arachis hypogaea* L.) and its control in Nigeria. *Journal of Stored Products Research* **5**, 275–280.

Mixon AC (1980) Potential for aflatoxin contamination in peanuts (*Arachis hypogaea* L.) before and soon after harvest — a review. *Journal of Environmental Quality* **9**, 344–349.

Petit RE, Taber RA, Schroeder HW, Harrison AL (1971) Influence of fungicides and irrigation practice on aflatoxin in peanuts before digging. *Applied Microbilogy* **22**, 629–634.

Sanders TH, Blankenship PD, Cole RJ (1988) Aflatoxin content of peanuts cleaned on a peanut belt screen before drying. *Proceedings of the American Peanut Research and Education Society* **20**, 25.

Sanders TH, Cole RJ, Blankenship PD, Hill RA (1985) Relation of environmental stress duration to *Aspergillus flavus* invasion and aflatoxin production in preharvest peanuts. *Peanut Science* **12**, 90–93.

Soil Survey Staff (1975) 'Soil taxonomy: a basic system of soil classification for making and interpreting soil surveys.' USDA agric. handbook no. 436. (US Government Printing Office: Washington, DC)

Truckess MW, Stack ME, Nesheim S, Page SW, Albert RH, Hansen TJ, Donahue KF (1991) Immunoaffinity column coupled with solution flurometry or liquid chromatography postcolumn derivatization for determination of aflatoxin in corn, peanuts, and peanut butter: collaborative study. *Journal of the Association of Official Analytical Chemists* **74**, 81–88.

Van Egmond HP (1995) Mycotoxins: regulations quality assurance and reference materials. *Food Additives and Contaminants* **12**, 321–330.

Wright GC, Hansen RB (1997) Climatic effects on aflatoxin incidence and management in peanut. In 'Proceedings of 2nd Australian peanut conference'. pp. 62–65.

Wright GC, Cruickshank AC (1999) Agronomic and crop modelling strategies to minimise aflatoxin contamination in peanuts. In 'Elimination of aflatoxin contamination in peanut'. ACIAR proceedings no. 89. (Ed. RG Dietzgen) pp. 12–18. (Australian Centre for International Agricultural Research: Canberra)

Wotton HR, Strange RN (1985) Circumstantial evidence for phytoalexin involvement in the resistance of peanuts to *Aspergillus flavus*. *Journal of General Microbiology* **131**, 487–494.

Young JH, Person NK, Donald JO, Mayfield WD (1982) Harvesting, curing and energy utilization. In 'Peanut science and technology'. (Eds HE Pattee, CT Young) pp. 458–485. (American Peanut Research and Education Society Inc.: Yoakum, Texas)