



Monitoring of Aflatoxins in Peanuts

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Abstract

Peanuts (*Arachis hypogaea* L.) are one of the most important oilseed crops and snack foods in the world Agro-food trade market. The major producers/exporters of peanuts are the United States, China, Argentina, Sudan, Senegal, and Brazil. Peanuts are a perishable commodity, easily spoiled by fungi. Aflatoxins are a group of natural compounds mainly produced by *Aspergillus flavus* and *Aspergillus parasiticus*. They have been found to be carcinogenic, teratogenic, and mutagenic to humans and animals. Aflatoxin contamination of peanuts is one of the most important factors determining the quality of peanuts and has caused significant financial losses for producing and exporting countries. Therefore, monitoring of aflatoxins in peanuts and peanut-contained products is very important for protecting consumers. Various methods have been tried to decontaminate aflatoxin contaminated commodities (e.g. peanuts). These include physical methods (sorting, irradiation techniques, heating), chemical methods (acids, bases, oxidising agents), biological methods (microbiological). All EU member states have set tolerance limits for certain mycotoxin food combinations but at present no country has covered all important mycotoxins and all relevant commodities. The data varies greatly from country to country. The overall competent authority for carrying out a monitoring for aflatoxin levels in foodstuffs lies within the Ministry of Food, Agriculture and Livestock in order to estimate the actual dietary exposure of aflatoxin contaminants in the foodstuffs (eg. peanuts) concerned.

Keywords: Peanut, Mycotoxin, Aflatoxin, Decontamination

Introduction

Peanut (*Arachis hypogaea* L.) can produce energy due to their high oil, protein and fibre content. These characteristics led the nut to become sensitive to fungal contamination, both pre- and post-harvest (Canavar and Kaynak, 2013). Numerous moulds may be involved in peanut spoilage, such as species of *Aspergillus*, *Penicillium*, *Fusarium* and of *Alternaria* in low percentage (Passone et al., 2008).

Aflatoxins are secondary metabolites produced by species of *Aspergillus*, specifically *Aspergillus flavus* and *Aspergillus parasiticus* (Rustom, 1997). The name "aflatoxin" is derived from the first letter in *Aspergillus*, and the first three letters in *flavus* (Rawal et al., 2010). The most important types of aflatoxins are AFB₁, AFB₂, AFG₁, AFG₂, AFM₁, and AFM₂. They are highly toxic and carcinogenic compounds that cause disease in livestock and humans. Aflatoxin B₁ is most frequently found in plant substrates and shows the

greatest toxigenic potential. Aflatoxins are stable small molecules and cannot be destroyed by heat treatment or during processing (Chen et al., 2013). Though aflatoxins are very stable and do not degrade up to 270 °C (their melting temperature) in dry conditions, biologically they can be converted into further toxic derivatives, such as epoxide, M₁, or M₂, by metabolism in humans and animals or less toxic derivatives, such as B_{2a}, by microorganisms (Samuel et al., 2014). Thus, monitoring and prevention of aflatoxins in foods and feeds are important issues worldwide (Chen et al., 2013).

The worldwide accepted levels for AFB₁ and total AFT (the sum of AFB₁, B₂, G₁, and G₂) range from 1 to 20 mg/kg and from 0 to 35 mg/kg (FAO, 2004). The Codex Alimentarius Commission (CAC) Joint Food and Agricultural Organization of the United Nations and the World Health Organization food standards program adopted a level of 15 mg/kg for AFT for unprocessed peanuts and 10

mg/kg for ready-to-eat tree nuts (Ding et al., 2012). For peanuts, nuts, dried fruits and cereals, the maximum level of 2 ng/g for B₁ and 4 ng/g for total aflatoxins have been set by the European Commission (Afsah-Hejri et al., 2011).

Prevention of Aflatoxin in Peanut

Aflatoxin contamination may occur in the field before harvest, during harvesting, or during storage and processing, thus methods for the prevention of contamination can be divided into preharvest, harvesting and post-harvest strategies. Whereas certain treatments have been found to reduce aflatoxin formation in peanuts, the complete elimination of aflatoxin is currently not realistically achievable (Torres et al., 2014). The best way to control mycotoxin contamination of peanuts is to prevent it in the first place. This is not always possible, but technologies exist which, if available and affordable, can prevent much of the contamination that would otherwise occur (Dorner, 2008).

Pre-harvest control: Preharvest aflatoxin contamination of peanuts is associated with drought stress that occurs late in the growing season while the crop is maturing (Dorner and Cole, 1997). Pre-harvest contamination can be reduced by introduction of good crop husbandry and appropriate cultural practices that limit the growth of aflatoxigenic fungi (Rustom, 1997).

-Resistance Varieties: The theoretically soundest approach of prevention is doubtless the breeding of cereals and other feed plants for resistance to mould infection and, consequently, mycotoxin production. Particularly in breeding wheat and corn, significant improvement of resistance has been achieved (Bata and Lasztity, 1999). However, resistance in peanuts to aflatoxin contamination under all conditions has still not been achieved and breeding efforts continue (Torres et al., 2014).

-Kernel moisture control: Pre-harvest aflatoxin contamination of peanuts essentially can be eliminated with proper and adequate irrigation. Developing and maturing peanuts are not susceptible to colonization by *A. flavus* and *A. parasiticus* until kernel moisture (water activity) begins to decrease in response to late season drought conditions with increased soil temperature (Dorner, 2008). For this reason, late season irrigation is recommended to help combat heat and drought stress, but this cultural practice seems to be impractical in some areas, especially in semi-arid and arid areas where water supplies are limited (Torres et al., 2014).

-Chemical and biological control: Several chemical control agents have been reported to inhibit aflatoxigenic mold growth and subsequent aflatoxin biosynthesis. While some studies suggested that pesticides and fungicides may be useful in controlling mycotoxin production under field conditions, other results have found that pesticides were ineffective in controlling mycotoxin production by *Aspergillus species* (Torres et al., 2014).

One strategy that has been developed for reducing preharvest aflatoxin contamination of crops is biological control, which is achieved by applying competitive non-toxigenic strains of *A. flavus* and/or *A. parasiticus* to the soil of developing crops (Dorner and Cole, 2002). This approach is based on the premise that when high number of spores of the nontoxigenic strains is added to soil, they will compete with naturally occurring toxigenic strains for infection sites for growth on peanut and for essential nutrients (Alaniz-Zanon et al., 2013).

Harvest control: It is very important to harvest the crop at optimum maturity, as excessive numbers of overmature or very immature pods at harvest can be reflected in high levels of aflatoxin in the final product. Also delays in harvesting will result in poor quality seed due to mold infections and subsequent aflatoxin contamination of the seeds/pods (Torres et al., 2014). During the harvesting process it is important that every effort is made to avoid physical damage to the agricultural commodities with crops which have been physically damaged being more susceptible to fungal growth (Kabak et al., 2006). The temperature, soil humidity and climate conditions of target peanut production areas are very useful in determining the best harvest time (Canavar and Kaynak, 2013).

Post-harvest control: Post-harvest contamination can be minimized by application of proper curing, drying, sorting and storage procedures (Rustom, 1997). According to the guide of Codex, to prevent an increase in aflatoxin contamination occurring during storage and transportation, it is important to control the moisture content, the temperature in the environment, and the hygienic conditions. The minimum moisture content for *A. flavus* growth on peanut is 8–10% at around 82% relative humidity, and aflatoxin production is generally correlated with kernel moisture contents of 10% or higher (Torres et al., 2014). Both the main aflatoxin producing *Aspergillus* strains *A. flavus* and *A. parasiticus* can grow in the temperature range from 10-12 °C to 42-43 °C, with an optimum in the 32 to 33 °C range. Aflatoxins are produced at

temperatures ranging from 12 to 40 °C (Sweeney and Dobson, 1998).

One strategy to reduce the entry of aflatoxin into the peanut chain is the use of chemical treatments such as acetosyringone, syringaldehyde and sinapinic acid and ammonia applications during post-harvest to reduce both fungal growth and toxin production (Canavar and Kaynak, 2013). From a human health perspective, the antioxidants such as butylated hydroxyanisole (BHA), propyl paraben (PP) and butylated hydroxytoluene (BHT) are allowed for use as antimicrobial agents by the US Food and Drug Administration (FDA) and are regarded as safe (GRAS) chemicals (Passone et al., 2008).

Detoxification Methods Of Aflatoxin in Peanut

Various methods have been tried to decontaminate aflatoxin contaminated commodities (e.g. peanut). These include physical methods (sorting, irradiation techniques, heating), chemical methods (acids, bases, oxidising agents), biological methods (microbiological) (Jewers, 1990).

According to the FAO any decontamination process to reduce the toxic and economic impact of mycotoxins needs the following requisites (Kabak et al., 2006; Rustom, 1997; Piva et al., 1995):

- It must destroy, inactivate, or remove aflatoxins;

- It must not produce or leave toxic and/or carcinogenic/mutagenic residues in the final products or in food products obtained from animals fed decontaminated feed;

- It should not adversely affect desirable physical and sensory properties of the product;

- It must be capable of destroying fungal spores and mycelium in order to avoiding mycotoxin formation under favorable conditions;

- It has to be economically feasible, and technically applicable.

Physical methods

Physical methods of aflatoxin contaminated peanut have been include: sorting, heating, irradiation.

- Sorting:** Physical removal or separation of aflatoxin-contaminated crops is an important strategy for reducing aflatoxin levels and can be achieved based on differing physical properties such as size, shape, color and visible fungal growth on the affected commodity (Womack et al., 2014). The most effective technique for managing aflatoxin contamination in

commercial shelling plants is electronic colour sorting (ECS). Peanuts that have been colonized by aflatoxigenic fungi are often discoloured, and ECS very efficiently removes a high percentage of the contaminated, discoloured kernels (Dorner, 2008).

- Irradiation:** Radiation is typically categorized as either ionizing (IR) or non-ionizing (NIR), with IR involving X-rays and gamma (γ) rays and NIR involving UV rays, microwaves, infrared rays and radio waves (Kabak et al., 2006). The use of gamma radiation to inactivate aflatoxins was investigated. The toxicity of a peanut meal contaminated with AFB₁ was reduced by 75% and 100% after irradiation with gamma rays at a dose of 1 and 10 kGy, respectively. However, doses higher than 10 kGy inhibited the seed germination, and increased the peroxide value of the oil in gamma-irradiated peanuts (Rustom, 1997).

- Heating:** Aflatoxins have high decomposition temperatures ranging from 237°C to 306°C. Solid AFB₁ is quite stable to dry heating at temperatures below its thermal decomposition temperature of 267°C (Rustom, 1997). Özkarslı and Var (2003) reported that microwave roasting of peanuts in a 2450 MHz for 90 seconds caused a 35.4% reduction in aflatoxin levels. In another study, Mobeen et al. (2011) achieved 50 to 60 % reduction in aflatoxin levels in peanut and peanut products by microwave roasting at 92°C for 5 min.

Chemical methods

A wide range of chemicals have been shown to reduce, destroy or inactivate mycotoxins (Piva et al., 1995). These chemicals include sodium hydroxide, hydrogen peroxide, ozone, sodium hypochlorite (Zhang et al., 2012). Although such treatment reduces nearly completely the mycotoxin concentration, these chemicals cause losses of some nutrients (Bata and Lasztity, 1999).

Ozone due to its safety, environment-friendly, low cost, high efficiency in decomposing aflatoxin B₁, has been widely studied and used in the food industry (Diao et al., 2013). Ozone eliminates the handling, storage, and disposal problems of conventionally used post-harvest pesticides (Zorlugenç et al., 2008). Proctor et al. (2004) achieved the highest level of degradation for aflatoxin B₁ (77±2%) after ozonation of peanut kernels for 10 min at 75°C. However, it is important to note that, as a chemical

detoxification method, ozonation with low ozone concentration and short treatment time is required to mitigate the damage to peanut nutrition (Chen et al., 2014).

Biological methods

Biological detoxification is the method of choice to deactivate mycotoxins. This comprises binding by adsorptive materials as well as microbial inactivation by specific microorganisms or enzymes (Schatzmayr et al., 2006).

Many microorganisms including bacteria, lactic acid bacteria and acid producing molds can metabolize and inactivate aflatoxins, with *Flavobacterium aurantiacum* as the most active organism (Rustom, 1997). Özkaya (2001) showed that *F.aurantiacum* strain NRRL B-184 reduced the amount of aflatoxin B₁ at 79-98.9 %, 92.6-99.8 % and 88.7-100.0 % in 48 hours in phosphate buffer, peanuts and dried red pepper, respectively. In another study, Zorlugenç (2009) reported that using *F.aurantiacum* strain NRRL B-184 the reduction of aflatoxin B₁ content in the rate of 84.28 % and 98.84 % in 72 hours at whole soy bean (1000 ng/g AFB₁) and milled hazelnut (500 ng/g AFB₁), respectively.

The importance of biological methods of aflatoxin degradation will likely increase if consumer resistance to chemical treatments continue to grow. However, the bright orange pigmentation associated with this bacterium would likely limit its applicability for food and feed fermentations (Bata and Lasztity, 1999).

Conclusion

Aflatoxin contamination of peanuts is one of the most important factors determining the quality of peanuts and has caused significant financial losses for producing and exporting countries. Thus, monitoring of aflatoxins in peanuts and peanut-contained products is very important for protecting consumers. Although the different methods used at present are to some extent successful, they have big disadvantages with, limited efficacy and possible losses of important nutrients and normally with high costs. Therefore, new methods of detoxification are necessary to prevent health risks and economic losses that result from aflatoxin contamination.

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