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Chapter

The Toxification and Detoxification Mechanisms of Aflatoxin B1 in Human: An Update

Qun-Ying Su

Abstract

Aflatoxin B1 (AFB1) is the most common carcinogen of aflatoxin, which contaminates many agricultural products in the daily diet of humans. More than 50% of patients with developing hepatocellular carcinoma (HCC) feature AFB1 exposure due to their shared consumption of contaminated food. One of the main mechanisms of AFB1-induced liver carcinogenesis is its biological activation and its interaction with DNA to produce AFB1-E-N7-dG adduct. This product may result in the formation of DNA damage and the mutations of tumor-associated genes such as TP53 and ras. In human, several pathways involving in AFB1 detoxification, including I- and II-type detoxification, DNA repair, have been reported. This study reviewed the detoxification mechanisms of AFB1 in human as well as AFB1 occurrence and toxification. Additionally, we also discussed prevention methods for AFB1 exposure.

Keywords: aflatoxin B1, toxification, detoxification, mechanism

1. Introduction

Aflatoxin B1 (AFB1), an important mycotoxin, is first identified in animal feed in 1961 due to the death of 100,000 turkeys in the UK where these turkeys were feed using the peanut powder with the high concentration of AFB1. Until now, this mycotoxin has proved to come from the secondary metabolites of *Aspergillus* under adverse conditions such as prolonged drought and insect-mediated damage to crops. Growing studies have also shown that individuals having crops contaminated by AFB1 often causes different poisoning. Its high-level exposure will cause acute poisoning and severe cellular damage, even death; whereas its long-term low-level exposure will induce chronic poisoning, genic mutations, even malignant alterations of cells. Evidence from epidemiological and clinicopathological studies displays that AFB1 has strong carcinogenicity, mutagenicity and teratogenicity. For human and animals, AFB1 often induces hepatocellular carcinoma, thus, is classified as one of I-class chemical carcinogen of hepatocellular carcinoma by the International Cancer Research Center [1–5]. Here, we will review the occurrence of AFB1 and its toxification effects on human. Additionally, we will also discuss all known detoxification of AFB1.

2. AFB1 occurrence

2.1 Toxic fungi and their classifications

Toxic fungi often live in the human crops and produce mycotoxins such as aflatoxins. Toxic fungi in crops can be divided into two categories according to whether their mycotoxins are produced before or after crop harvest. The first category is termed as field fungi, which often invade crops and produce mycotoxins before harvest. The another, also called storage fungi, mostly occurs in the storage of crops after harvest. The sources of both types of toxigenic fungi are affected by environmental factors. Crops before harvest, fungi can invade crops to produce toxins by interacting with other organisms, such as insects. The harvested crops are regulated by factors such as nutrients, temperature and humidity in the air, and biological agents (insects, competitive interference). Furthermore, toxigenic fungi can be divided into four types according to their effects on crops: *A. fungi* acting as plant pathogens, such as grass fungi; *b.* fungi producing fungal toxin and stressing plants, such as *Candida* and *A. flavus*; *c.* fungi acting as colonizers (such as *Aspergillus flavus*), which first colonize in harvest plants and subsequently, produce mycotoxin and contaminate crops; and *d.* fungi decomposing plants (such as *Penicillium chrysogenum* and *Aspergillus oryzae*), which often live in the soil [6, 7].

The crop fungi inoculate the growing crop kernels in the field and proliferate in storage under suitable conditions. Among known crop fungi, *Aspergillus*, *Fusarium*, and *Penicillium* have identified as toxin-producing fungi. Although many compounds produced by these toxin-producing fungi are known as their toxins, there have been only five important agricultural mycotoxins until now: deoxycinol, praurone, ochratoxin A, fumarin and aflatoxin. Mycotoxins produced by *Fusarium* include fumonisins, deoxycinol and zerneone. Although *Penicillium* and *Aspergillus* are storage fungi, they can also invade field stress plants and produce toxins. Increasing evidence has shown that *Penicillium* can produce ochratomycin, citrinin and patron, and that *Aspergillus* can produce aflatoxin, citrinin and baturin [8, 9].

2.2 AFB1 occurrence

Several previous reviews have fully summarized the occurrence and biosynthesis of AFB1. Briefly, AFB1 are an important class of mycotoxins mainly produced by *Aspergillus flavus* and *Aspergillus parasiticus*. This term is so named and concerned because of the following several reasons: *a.* this mycotoxin has been identified in the *A. flavus* and regarded as pathologic agent of "turkey X" disease; *b.* this mycotoxin is the first B-type aflatoxin which can produce fluorescent characteristic under UV light; and *c.* AFB1 often display its severe toxic effects on human and animals. Usually, it is synthesized through 18 biological steps under the regulations of a huge neighbor gene cluster consisting of about 60–70 kb in original fungi. This biosynthesis at least involves in the three stages, consisting of the formation of primary product hydroxyversicolorone (the first to eighth step), middle product versicolorin B (the ninth to twelfth step) and ultimate product AFB1 (the thirteenth to eighteenth step). During the biosynthesis of AFB1, several key enzymes, including nicotinamide-adenine dinucleotide, nicotinamide-adenine dinucleotide phosphate reduced form, and 2S-adenosylmethionine, are required for the biosynthesis limitation [3, 10, 11].

3. AFB1 toxification and toxic mechanisms

3.1 The effects of AFB1 on the food chain

A. flavus is widely present in the soil, causing pollution to many crops such as corn, peanuts, rice, etc., and using these crops as a host produces aflatoxin, which in turn contaminates crop fruits. Aflatoxin contamination of food and animal feed has now become a major problem that threatens food safety. Crops can be contaminated with fungi in the field, harvested, and stored, making crop contamination control difficult. The Food and Agriculture Organization of the United Nations (FAO) estimates that 25% of the world's food crops are contaminated with mycotoxins [12]. Aflatoxins, the most harmful toxins, are the most difficult to deal with because they are commonly found in corn, peanuts and their products, cottonseed, peppers, peppers, pistachios and other foods. Studies have shown that the type of mold and its concentration of conidia, as well as the moisture content of corn, play a key role in the process of mold infection, spoilage and AFB1 production in corn. In the field, Aspergillus flavus enters the plant primarily by vaccination or secondary inoculation, which in turn infects the seed to produce aflatoxin. In spring, the source of inoculation of Aspergillus flavus spores is mainly from the propagules in the soil, the plant debris in the soil, and the wintering mycelium, insects or the Aspergillus flavus nucleus in the soil in the litter. In corn fields, spores are mainly derived from the spore-derived sclerotium conidia of Aspergillus flavus. Before the harvest, the insects damage the corn kernels to form the sclerotia. When harvesting, the sclerotia is dispersed in the soil, and the spring conidia are exposed on the surface of the sclerotium. A secondary inoculation source was found in the cotton field, and *A. flavus* was isolated from the leaves, flower buds and leaf discs of cotton, the content was 15, 94, or 56%, respectively. Most of the colonies were mainly distributed on the calyx tablets. Conidia are the main source of secondary inoculation. Fungal spoilage and mycotoxin contamination are major problems in crop contamination. Grains are affected by storage conditions and the environment after harvesting, such as storage containers, oxygen content in the air, water activity, temperature, and insects, all of which are factors of toxin contamination [7]. If stored poorly, it will increase the contamination of mycotoxins. As one of the main crops for human food and livestock feed, corn is planted annually at 120 million hectares and is one of the most polluted toxins. Aspergillus flavus is the main fungus that is infected after corn harvest. The drying and storage conditions of corn before storage are extremely important. Moisture can accumulate from the activity of pests, which provides ideal conditions for the proliferation of fungi and the accumulation of mycotoxins. In order to reduce the effects of mycotoxins on food and feed chains, it is necessary to control pest and fungal contamination [13, 14]. Humidity and temperature have important effects on mold growth and mycotoxin production. Therefore, humid and hot climates in tropical and subtropical regions provide favorable conditions for mold growth. The moisture content of the grain is generally expressed in terms of water content. Pathogenic fungi that invade crops prior to harvest typically require higher moisture levels (200–250 g/kg) to infect, while fungi that can proliferate during storage (130–180 g/kg) require higher moisture levels. Therefore, most feeds with a water content above 130 g/kg are prone to mold growth and formation of mycotoxins [15]. Therefore, the control of the moisture content in the grain becomes particularly important, especially in the control of moisture at the harvesting point, while the drying and storage of the grain before storage and the frequency of grain drying and plowing, as well as

the insects and microorganisms in the stored grain are also Factors affecting water activity. It was found to be closely related to climate in the detection of aflatoxin levels after storage of Benin corn. Benin's aflatoxin contamination levels increase in dry and hot conditions in June each year; while the harvest season is affected by climate, the peaks of rainfall or the late planting of corn will increase aflatoxin levels during the rainy season [16]. Limiting the occurrence of AFB1 before crop harvesting can be achieved by reducing drought and temperature, controlling weeds, reducing insect damage, efficient harvesting techniques, and reducing soil Aspergillus spores through crop turnover. Using biological control, a competitive, non-toxigenic strain of Aspergillus flavus is applied to the developing soil to compete with naturally occurring toxigenic strains. Studies have shown that these biological control strategy can aflatoxin. The pollution is reduced by about 80–90%. Control of afb1 sensitive crops after harvest can be achieved by controlling factors that affect fungal growth, such as water activity, temperature, gaseous environment, and the use of pesticides or food preservatives. Harvesting only cereals with a moisture content of around 24% reduces the risk of grain damage and subsequent AFB1 production [17, 18].

3.2 The toxic effects of AFB1 on human and animals

In 1963, Asao et al. completed the structural clarification of AFB1, a member of the aflatoxin family containing a fused difuranyl group [19]. AFB1 is highly toxic to humans and several animals, and has three major characteristics: organophilic, genotoxic, and carcinogenic. Its pro-organism is mainly caused by damage to the liver, which can lead to hepatic hemorrhage and hepatocyte necrosis. The genotoxicity is mainly to induce the formation of AFB1-DNA adduct and the hot spot mutation of P53 gene. The carcinogenicity is mainly caused by hepatocellular carcinoma. The main toxicological effect of AFB1 is to induce DNA damage. AFB1 has been proven to be the main cause of liver cancer in patients with hepatitis B virus infection. It is a genotoxic liver cancer, which may cause cancer by inducing DNA adducts, leading to genetic changes in target cells, leading to DNA strand breaks and DNA base damage. And oxidative damage can eventually lead to cancer. AFB1 is mainly metabolized by the liver, and AFB1 taken from food is mainly metabolized by the cytochrome P450 enzyme to the final carcinogen AFB1-8-9-epoxide (AFBO). When AFBO reacts with DNA, it inhibits gene mutation in P53, a hotspot coding region of exon 249, by interacting with guanine bases, which may lead to HCC. AFB1 is metabolized by the P450 system into a number of hydroxylated products, including AFM1, AFQ1, AFP1, AFB2a [11, 20–25]. After aflatoxin is ingested into the human body, it mainly manifests as an acute or chronic disease. Acute attacks usually involve high concentrations of aflatoxins. For example, 317 cases of acute liver failure occurred in Kenya in 2004. The main reason is the consumption of aflatoxin-contaminated corn, and the case of patients with AFB1 lysine in serum. The adduct concentration was the highest in history. Growth retardation, immunosuppression, and carcinogenicity are chronic effects, and the incidence of chronic attacks in developing countries is higher because of exposure to low levels of aflatoxin intake [26].

3.2.1 Effect of aflatoxin on growth and development

An epidemiological survey was conducted in West Africa to measure exposure to aflatoxins in children between 9 months and 5 years of age, and their growth, development and height were examined against the reference population of World Health Organization (WHO) [27]. Studies have shown a strong association between

exposure to aflatoxin in children and dysplasia and underweight. In a field outbreak of aflatoxin, egg production fell by 5% [28]. The study data showed that for every 1 mg/kg of aflatoxin AFB1 in the feed, the growth rate of pigs would be reduced by 16% and broilers by 5% [29].

3.2.2 Immunosuppressive

In these animal studies, AFB1 has been shown to induce immunosuppression. For example, in studies of AFB1 exposed animals, it was found that the activity of B cells and T cells decreased, because T cells were more sensitive to AFB1 toxicity [30]. Research data from GY et al. showed that chicken phagocytic cells were severely damaged during aflatoxosis, and the ability to remove foreign substances from the circulation decreased, which may reduce the ability to process antigenic components. Aflatoxell chickens are more susceptible to infection [31]. In pigs, AFB1 exposure reduces lymphocyte response to mitogens, inhibits large phage migration and delayed skin allergic reactions [32]. Although many data on AFB1 immune effects have been obtained from animal studies, there is little data on the effects of long-term consumption of food contaminated with AFB1 on the human immune system. The effect on the immune system by aflatoxins in the diet of Gambian children found a decrease in sIgA levels in saliva, probably due to the high level of exposure to aflatoxins in the diet [33]. In a study of aflatoxin AFB1 exposure and cellular immune status in 64 Ghanaians, it was found that AFB1 exposure may result in a decrease in the major constituent cell T cells and B cells that cause lymphocyte subpopulations. High levels of AFB1 albumin adducts significantly reduced perforin- and granzyme a levels in CD8+ cytotoxic T cells compared to low levels of AFB1 albumin adduct. In participants with high levels of AFB1, changes in these immune parameters may result in impaired cellular immune function, thereby reducing host resistance to infection [34].

4. Detoxification of AFB1

Since the contamination of aflatoxins in food poses a risk to human health and leads to serious economic losses in crops, we have every reason to implement new methods to ensure the safety of food production. There are two main methods of implementation: (a) prevention of mold contamination and growth; (b) detoxification of contaminated products by opponents. Prevention of mycotoxin contamination can be achieved by storage before or after harvesting of the crop. However, the pollution of toxins is inevitable, and the detoxification pathway for contaminated food after harvest has been the subject of our in-depth research. Detoxification methods commonly used are physical methods and chemical methods. This article will focus on new research on detoxification of harvested contaminated crops.

4.1 Physical method

The most common way to remove AFB1 using physical methods is to heat and use gamma rays. Aflatoxins are highly thermostable. Studies have shown that AFB1 levels are significantly reduced by heating at 100 and 150°C for 90 minutes, respectively, at 41.9 and 81.2%. The AFB reduction rate of the soy milk after cooking was 97.9%, and the AFB1 reduction rate of the steamed soybeans after cooking was 33.6%. And studies have shown that high pressure cooking is better than ordinary cooking to remove AFB1. When the soybean is steamed or steamed in a pressure cooker, the reduction rate of the pressure cooker is about 10% higher than that

of the steam. Using autoclave cooking in rice can reduce AFB1 levels by 72–83%. The high-pressure cooking method is low in cost and easy to handle, and one of the challenges it faces is how to ensure the integrity of the food after heating. To ensure the integrity of the food, the use of maximum temperatures is often limited [35, 36]. The gamma ray has a strong penetrating electromagnetic wave that can penetrate the material without leaving any residue, which is its advantage. There have been many reports of the increase, decrease, or even unaffected mycotoxin produced by fungi under different conditions. Studies have shown that the fungal structure on paper with a minimum radiation dose of 16 kGy has been altered to avoid fungal growth. Library and file management staff use gamma radiation protection technology to provide a powerful means for the preservation of ancient books, archives and other paper materials [37]. A dose of gamma radiation exceeding 10 kGy can inhibit the germination of peanut seeds. Therefore, proper drying, packaging and environmental control measures with low relative humidity can reduce the growth of fungi and ensure safe, high quality peanuts [38]. The DI Stefano study showed that a radiation dose of 0.5–15 KGy resulted in a decrease in aflatoxin levels in the feed, while a 15 kGy gamma ray did not completely destroy ochratoxin A and aflatoxin in the test feed, FAO/International The IAEA/WHO Expert Committee on Food Irradiation has concluded in its report that foods with an average radiation dose of 10 kGy will not cause toxicological hazards and that toxicologically tested foods do not require retreatment. It is necessary to irradiate the food with radiation before the mold produces toxins [39].

4.2 Biological treatment

Studies using biotechnology to reduce AFB1 levels in contaminated foods fall into two main categories: one that uses plant extracts to degrade AFB1 and the other that inoculates bacterial strains in food substrates. In recent years, natural plant products have attracted much attention as synthetic antibacterial agents because of their biodegradability, biosafety, effectiveness, and regenerability. At the same time, they are conveniently used as an eco-friendly technology for detoxification. Mycotoxins. Many studies have shown that plant essential oils can inhibit the growth of microorganisms and reduce the production of toxins. Bluma et al. showed that the addition of essential oils in corn kernels has a significant effect on the growth rate, hysteresis and accumulation of AFB1 of aflatoxin molds. Depends on water activity, AFB1 concentration and incubation time [40]. In addition to plant essential oils, water extracts of plants can also be used to dissolve AFB1. Another study by Vijayanandraj et al. also demonstrated the effect of different parameters on the detoxification of AFB1 aqueous extracts from different medicinal plants. They concluded that the leaf extract of Vasaka (*Adhatoda vasica* Nees) showed the greatest AFB1 degradation (≥98%) after incubation for 24 hours at 37°C; the A. vasica leaf extract was heated to 100 by high temperature. Celsius for 10 minutes or autoclaved at 121 degrees Celsius, the detoxification ability is significantly reduced; the A. vasica leaf extract is detected by mass spectrometry to the purified alkaloid, which is believed to be the principle of aflatoxin detoxification [41]. Iram et al. showed that *O. basilicum* leaves extract had significant degradation rates for aflatoxins B1 and B2, and the degradation rates were 90.4 and 88.6%, respectively. The structure of the degradation products was identified by mass spectrometry. Most of the products were passed. Formed by removing the double bond on the terminal furan ring and modifying the lactone group, the degradation property is significantly less toxic than AFB1. The plant extract is easy to obtain, cost-effective and bio-safe, and can be directly sprayed with aqueous plant extracts. Simple, does not involve technical knowledge, is a very good source of detoxification [42].

In another method, inoculation of the bacterial strain is to reduce AFB1 by physical binding or metabolism of the bacterial strain directly to AFB1. The biodegradation of aflatoxins has yielded some successful attempts, although most are carried out in sterile culture. The microbial degradation of aflatoxin is achieved by the activity of the enzyme, which is capable of decomposing the refractory polyheterocyclic molecules of aflatoxin. Brana et al. showed that *Pleurotus eryngii* can degrade AFB1 [43]. Farzaneh et al. showed that the repair rates of AFB1 by *Bacillus subtilis* UTBSP1 in nutrient broth and pistachio were 85.66 and 95%, respectively [44]. Liu et al. used cellulose bacteria to degrade the degradation ability of AFB1 in cottonseed meal. By improving the fermentation conditions, the degradation rate can reach 83.4% [45]. Carolyna and other studies have shown that AFB1 is combined with bacteria through weak non-covalent and acid. Of the bacteria treated, the binding may be intracellular rather than extracellular [46]. It is well known that lactic acid bacteria can degrade aflatoxins. Bueno et al. showed that lactobacillus and *Saccharomyces cerevisiae* can rapidly remove AFB1. The binding of AFB1 with microorganisms is a rapid process (no more than 1 minute), and this binding forms a reversible complex between the toxin and microbial surface without the need for chemical modification of the toxin [47]. Studies by Flora Oluwafemi et al. showed that AFB1 was significantly reduced (44.5%) in 50 ng/g contaminated corn, while AFB1 was the least reduced (29.9%) in 500 ng/g contaminated corn. Because lactic acid bacteria are non-toxic, they have many benefits for human health, and it is also possible to reduce the level of aflatoxin to a lower toxic dose. Lactic acid bacteria have broad application prospects as biopreservatives for food and feed [48].

4.3 Chemical treatment

Mycotoxins can be removed or reduced chemically, and acids, bases, oxidizing agents, and reducing agents have been shown to destroy or extinguish mycotoxins. Acids are a natural part of foods that are added to the industry to add flavor to the food, and even some acids are used as preservatives or antioxidants. Organic acids in foods can degrade AFB1. AIKO et al. tested the degradation of AFB1 by various organic acids and considered that the effect of lactic acid was most effective in the organic acids tested. Since lactic acid is endogenous in the human body and is present in many foods, lactic acid is considered to be safe. Therefore, lactic acid can be recommended for food processing and as a preservative in fermented foods [49]. Rushing and other studies have shown that under acidic conditions, organic acids and arginine can be mixed to treat contaminated foods, and AFB1 can be rapidly converted to AFB2a-Arg within 20 minutes, reducing toxicity [50–53]. Aly et al. showed that HCL can effectively degrade AFB1 during acid hydrolysis [54]. Alkaline cooking is also used in the commercial to remove AFB1 from corn. Amination under high temperature and pressure conditions can also reduce AFB1 in corn. There are also many studies on the degradation of AFB1 in foods using ozone. Ozone has been reported as an antibacterial agent because it has antibacterial effects against spores and bacteria of fungi, bacteria, viruses, protozoa and fungi, and has a wide range of antibacterial agents. Ozone inhibits or microbial growth by oxidizing cell membranes and cell wall complex processes [55]. Diao et al. showed a significant decrease in AFB1 levels in peanut seeds at 13 and 21 mg/l ozone concentrations [56]. Proctor et al. showed that the use of ozone oxidation can degrade AFB1 in peanuts, and at an increased temperature of 75°C, AFB1 degradation rate reached 77% in just 10 minutes [57].

4.4 Sorbent additives

The above method of degrading AFB1 is to destroy or reduce the content of AFB1 in food, and the adsorbent is opposite thereto, which prevents AFB1 from

entering the intestinal tract after ingestion by binding to AFB1, so as to prevent hepatotoxicity of AFB1. Novasil clay minerals and aflatoxins are highly affinitive and high-capacity combinations in the gastrointestinal tract. The study of NS has been shown to absorb AFB1 in vitro in both animal models and human studies, reducing the bioavailability of blood toxins, and its use in humans has not affected the utilization of vitamins and trace elements in the body, and has been determined through clinical trials. A safe dose of NS, a NS content of up to 2.0% (w/w) in the diet does not cause significant toxicity [58–62]. Xue and other studies have shown that the enteral nutrient NovaSil can effectively regulate the toxicity and carcinogenicity of co-exposure to AFB1 and fumonisin B1. When the concentration in the diet is as high as 0.5%, liver changes, liver glutathione S The number and size of -transferase (GST-P+) foci were significantly reduced [63]. Another commonly used binder is chlorophyll. Studies in mammals and fish have shown that chlorophyll can inhibit the formation of carcinogens through the combination of AFB1, reduce the bioavailability of tissues, reduce DNA adduction, and reduce the incidence of tumors [64]. Smimonich observed in the study that after adding chlorophyll to the contaminated AFB1 feed, the AFB1-DNA adduct was reduced by 42%, and the AFB1 albumin adduct was reduced by 65%. AFB n7 - guanine Urinary adducts are reduced by 90%. In the same study, it was also shown that chlorophyll reduced the volume of GSTP lesions in the liver by 74% and the mean number of abnormal crypt lesions in the colon by 63%. Studies have shown that chlorophyll can be used as an early biochemical and advanced pathophysiological marker for AFB1 carcinogenesis in the liver and colon [65].

4.5 DNA repair

In China, HCC is a common malignant tumor with a very poor prognosis, accounting for 55% of the world's HCC cases and more than 340,000 cases per year. This area of tumor-prone is mainly concentrated in eastern and southeastern China. Clinical epidemiological studies have shown that exposure to AFB1 and/or chronic infection with HBV and HCV is a major risk factor for liver cancer. Studies on the toxicity of AFB1 indicate that AFB1 damage to DNA plays a central role in the carcinogenic process of HCC associated with this toxin [48, 66]. AFB1 is metabolized by the cytochrome P450 enzyme into a reactive AFB1-8,9-epoxide (AFB1-epoxide), which is covalently bound to DNA to induce DNA damage. AFB1-induced DNA damage includes AFB1-DNA adducts, oxidative DNA damage, and gene mutations. The AFB1-DNA adduct in AFB1-induced DNA damage is 9-hydroxy afb1 (AFB1-N7-Gua), which is the most common type. The formation of the AFB1-N7-Gua adduct is first performed by pre-covalent insertion of the complex electrophilic between the double-stranded DNA and the high-stranded DNA, and then on the imidazole moiety of the formed AFB1-N7-Gua adduct. The charge generates another desired DNA adduct, a ring-opened carboxamide pyridine AFB1 (AFB1-FAPy) adduct. These adducts are capable of forming subsequent anti-repair adducts, dislocations, or lead to error-prone DNA repair, resulting in single-strand breaks (SSBs), double-strand breaks (DSBs), and base pair substitutions. The mutations caused by AFB1 exposure, the current major research and experiments believe that the P53 gene is closely related, there is a common mutation hotspot at 249 of TP53 (AGG to AGT) codon [11, 23, 25, 48]. However, epidemiological evidence suggests that although many people are exposed to the same level of AFB1, only a small percentage of the exposed persons have toxicological effects of AFB1, such as genetic mutations and HCC. The Nucleic Acid Excision Repair Pathway (NER), which has been shown to repair aflatoxin-induced DNA adducts, is the major DNA repair pathway. The repair steps of NER are mainly divided into: damage perception, opening denatured

bubbles, cutting damaged chains, transferring damaged oligonucleotides, filling gaps and ligation. There is increasing evidence that genetic polymorphisms in the NER gene are associated with DNA repair capacity and regulate the risk of cancer [66]. In China, molecular epidemiological studies of afb1-related HCC have investigated the association of several genes associated with the NER pathway, such as xeroderma pigmentosum C (XPC) and xeroderma pigmentosum D (XPD). In the oxidative damage of DNA caused by AFB1 exposure, the formation of 8-oxodG is important because it is abundant, highly mutagenic and hepatocarcinogenesis occurs. 8-oxodG lesions are mainly repaired by the BER pathway. The BER pathway promotes DNA repair through two common pathways: a. short patch BER pathway leading to a single nucleotide repair pathway; b. long patch BER pathway, resulting in at least two nucleotide repair pathways. Long et al. first reported DNA repair genes XRCC1, XRCC3, XRCC4, XRCC7, XPD, XPC (including rs25487, rs861539, rs7003908, rs28383151, rs3734091) by analyzing the AFB1-DNA adduct amount, TP53 gene mutation frequency and HCC risk. Genetic polymorphisms of (rs13181, rs2228001) and toxicological effects of AFB1 exposure. Studies have shown that the DNA repair gene XRCC1 gene mutation, XRCC3, XRCC4, XRCC7, XPC, and XPD may increase the AFB1-DNA adduct, the frequency of TP53M, and the risk of hepatocellular carcinoma, genetic mutations with lower DNA repair ability of these genes It should contribute to the toxicological effects of AFB1 and be of a preventive significance by identifying people with low DNA repair capacity [23, 67, 68].

5. AFB1-related legislation

In developing countries, AFB1 contamination of food is inevitable due to poor environmental and technical conditions, and it is not easy to be treated by high temperature, chemical, physical, etc. Humans can directly use contaminated foods (corn, peanuts, sorghum, rice, cashews, walnuts, pistachios, almonds) or animal products such as milk, eggs, etc. produced by using contaminated animals. The primary hazard of mycotoxin contamination in the food supply chain is human health, followed by animal health and productivity [69, 70]. Every country has strict controls on the mycotoxin contamination of food and feed to reduce human and animal exposure. Currently, Developed countries have access to federal regulatory bodies which set food safety standards and inspect domestic as well as imported/exported food products. Additionally, these countries have access to controlled storage conditions, which greatly reduces contamination post-harvest. These factors lead to lower overall contamination rates in developed countries. For example, the United States has reported acceptable AFB1 levels in corn (0–80 μg/kg during 1979–1983) and low daily intake of its citizens (0.34–197 ng/kg depending on the year and region of the country), which is much less than other undeveloped countries [71]. The European Union (EU) has some of the world's most stringent standards for mycotoxins in food and feed. Compared with the rest of the world, the European Union (EU) has the most extensive and detailed AFB1 presence in various foods and feeds provisions. It has been indicated that in many European countries the presence of AFM1 in milk and milk products was in lower range than the Asian and African countries [72, 73].

6. Summary and future direction

AFB1 is a kind of I-type chemical carcinogenic mycotoxin mainly produced by both *A. flavus* and *A. parasiticus* and is known to contaminate most of the world's food supply. AFB1 is the most potent of these compounds and has been well

characterized to lead to the development of hepatocellular carcinoma in humans and animals. The contamination of the food chain by AFB1 has a huge impact on human health and economic damage worldwide. The prevention and detoxification strategies of AFB1 have always been the goal of research. Although physical and chemical treatment is currently the main detoxification method, it is easy to lead to the loss of food nutrition. Biotherapeutics is a relatively new detoxification method. Natural plant extracts and plant essential oils are simple to produce, biosafe, and provide an excellent source of toxin detoxification. Studies have shown that individuals' susceptibility, such as genetic polymorphisms in DNA repair genes and/or metabolic genes, play a huge role in the different detoxification and the repair of DNA damage caused by AFB1. Thus, the biofunction supplementary methods which base on this kind of genetic difference may be not only regarded as potential new detoxification methods but served as potential biological markers for predicting the occurrence of such liver diseases as liver damage, liver cirrhosis, and hepatocellular carcinogenesis.

Conflicts of interest and source of funding

The authors declare no competing financial interests. This study was supported in part by the National Natural Science Foundation of China (Nos. 81,860,489, 81,760,502, 81,572,353, and 81,660,495), the Natural Science Foundation of Guangxi (Nos. 2018GXNSFAA281043, 2017GXNSFAA198002 and 2017GXNSFGA198002), Research Program of Guangxi "Zhouyue Scholar" (No. 2017-2038), Research Program of Guangxi Specially-invited Expert (No. 2017-6th), the '12th Five' Planning Program of Guangxi Education Science (No. 2015C397), the Innovative Program of Guangxi Graduate Education (No. JGY2015139), Research Program of Guangxi Clinic Research Center of Hepatobiliary Diseases (No. AD17129025), and Open Research Program from Molecular Immunity Study Room Involving in Acute & Severe Diseases in Guangxi Colleges and Universities (Nos. kfkt20160062 and kfkt20160063).

Abbreviations

Aflatoxin B1 (AFB1)
hepatocellular carcinoma (HCC)
Aspergillus flavus (*A. flavus*)
Aspergillus parasiticus (*A. parasiticus*)
The Food and Agriculture Organization of the United Nations (FAO)
World Health Organization (WHO)
AFB1-8-9-epoxide (AFBO)
Nucleic Acid Excision Repair Pathway (NER)
xeroderma pigmentosum C (XPC),xeroderma pigmentosum D (XPD)
European Union (EU)

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