



## REVIEW

### Grain Processing Methods' Effectiveness to Eliminate Mycotoxins: An Overview

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Mycotoxins like aflatoxins, ochratoxins, patulin, citrinin, deoxynivalenol (DON), fumonisins, trichothecenes, *etc.*, produced by molds (*Aspergillus avus*, *A. parasiticus*, *Penicillin* spp, *etc.*) could occupy such grains as peanuts, millet, sunower, cassava, beans, sesame, sorghum, maize, *etc.* In order to either eliminate and or reduce the levels of mycotoxins, as well as detection limits, there are a number of grain processing techniques that have been reported in the literature. In this article, an overview of the effectiveness of grain processing methods to eliminate mycotoxins was performed. Specifically, the grain processing techniques considered include: extrusion, alkaline cooking, flaking, roasting, frying, baking, milling, cooking, cold plasma, cleaning, trimming, sorting, as well as chemical treatments like the use of sodium hydroxide, citric acid, sodium bisulfite and ammonia. Overall, while most methods seem effective in reducing some mycotoxins over others, more studies are required to identify how these methods particularly their combinations would elevate the removal/degradation of mycotoxins in grains, particularly to safer levels.

**Keywords:** Agrofood processing, Aflatoxins, Ochratoxins, Deoxynivalenol, Fumonisin.

## INTRODUCTION

Mycotoxins have been among the major problems of grains in some parts of the world, such as sub-Saharan Africa and Asia, usually resulting in the reduction in the economic and nutritional values of agricultural produce [1]. Some common mycotoxins include aflatoxins, ochratoxins, patulin, citrinin, deoxynivalenol (DON), fumonisins, trichothecenes, *etc.* These mycotoxins are produced by molds such as *Aspergillus flavus*, *A. parasiticus*, *Penicillin* spp, *etc.* and commonly found in peanuts, millet, sunower, cassava, beans, sesame, acha, sorghum, maize, *etc.* The causes of high proliferation of mycotoxins in foods and feeds are mostly because of poor pre-harvest, peri-harvest and post-harvest operations [1]. Inadequate government

legislation, as well as the unawareness (apparently, most grain handlers either unaware of mycotoxins or with limited knowledge) and insufficient education among agriculturists, farmers, consumers and entrepreneurs would be among the major reasons for the high proliferation of mycotoxins. Majority of people in various parts of the world consume grains and/or grain products on daily basis. As a result, they stand the risk of constant exposure to mycotoxins. Insufficient diversity in diets and poor food consumption patterns would increase the risks of mycotoxin exposures, with most staple foods being prone to mycotoxin proliferation [1]. The control and detection of these toxins remain marginal some extent, although some workers have developed portable detection device aimed at the on-site detection of mycotoxins, especially aflatoxins, in

grains and have also demonstrated that some starter cultures applied during production of few cereal beverages can bind to some mycotoxins.

Mycotoxins are well-known secondary metabolic compounds released by molds, which mostly colonize food crops and are food safety issues because of their chronic and acute toxicity on both humans and animals. Additionally, there are a number of well-known and yet equally precise methods and techniques for identifying and quantifying mycotoxins are continuously developed with the purpose of reducing/eliminating the levels of mycotoxins, reducing limit of detection, improving identification accuracy, among others. Furthermore, many industrial processes have been demonstrated to have the ability of reducing mycotoxin levels in foods and feeds. Additionally, the reduction in mycotoxin levels may result from normal natural processes used in the preparation of foods and feeds or through procedures purposely introduced in the course of processing, with aim of reducing mycotoxin levels [2]. In this article, an overview of the effectiveness of grain processing methods to eliminate mycotoxins was performed. Specifically, extrusion, alkaline cooking, flaking, roasting, frying, baking, milling, cooking, cold plasma, cleaning, trimming, sorting, as well as chemical treatments like the use of sodium hydroxide, citric acid, sodium bisulfite, ammonia, were considered. Table-1 shows the effectiveness of common food processing methods and techniques on mycotoxin reduction in foods and feeds.

**Common grain processing methods and their role in removing mycotoxins:** Some methods commonly used in processing grains and grain products include fermentation, sorting, trimming, cleaning, milling, heat treatment, cold plasma, *etc.* These common processing techniques offer alternative ways to mycotoxin concerns in foods and feeds. Although some of them are not originally intended to reduce mycotoxin levels, they have been shown to contribute significantly to their reduction. Food and agricultural processing cover all biological, chemical and physical processes raw grains undergo during the formulation and production of foods as well as nutraceuticals. In last two decades, many studies have been carried out regarding the levels of reduction of mycotoxins in processing worldwide [3]. The processes considered in this review are extrusion, alkaline cooking, flaking, roasting, frying, baking, milling, cooking, cold plasma, cleaning, trimming, sorting as well as chemical treatments such as use of sodium hydroxide, citric acid, sodium bisulfite, ammonia, *etc.* It is noteworthy to state that heat application in cooking and preserving products forms basis of thermal processing, such as extrusion cooking, normal cooking, canning, roasting, baking and frying.

**Cleaning, trimming and sorting:** The preliminary processes such as cleaning, trimming and sorting may decrease the concentrations of mycotoxins in foods and feeds, but they do not remove all the mycotoxins. Often, the levels of these mycotoxins still remain within unsafe levels after undergoing these preliminary processes. Initial food (grain) conditions and contamination levels have significant effects on efficiency of cleaning. About 40% to 80% aflatoxins reduction could be achieved using physical cleaning, in which nuts, seeds or kernels damaged by molds are removed from whole grains [4]. In cleaned wheat,

deoxynivalenol (DON) was 4.6 mg per kg, whereas in dockage DON was 16.7 mg per kg; the initial level was reported to be 7.1 mg per kg [5]. About 74% decrease in levels of DON was reported after sorting samples with initial gross contamination. However, several kernels infected with *Fusarium*, which might have high trichothecenes levels, could not be physically distinguishable from wholesome grains and sorting did not remove it; thus, routine cleaning of grains can result in at most small (about 20%) decrease in levels of trichothecene [6]. Deoxynivalenol (DON) was reduced by 16% with screening, which showed 4.7-fold greater DON levels than cleaned soft US wheat done with air flow and screening combined [7]. Decrease in DON levels reported in cleaned wheat were from 6 to 19%, but had initial DON levels of 7.9 to 9.6 mg per kg [8]. Scouring has been shown to reduce levels of DON by 22% from initial 12.5 mg/kg level [9]. Cleaning reduced concentrations of Fumonisin in corn by 26 to 69% [10]. However, just 2 to 3% ochratoxin A reduction was achieved in barley using cleaning [11]. Overall, cleaning, trimming and sorting may decrease the concentrations of mycotoxins in foods and feeds, however, the levels of these mycotoxins remaining after these preliminary processes are usually still within unsafe levels.

**Roasting:** Roasting is a method used for cooking using dry heat, which often make use of oven, open flame or other sources of heat. The food may or may not come in direct contact with heating equipment. It can improve the flavours of foods through Maillard browning and caramelization, which mostly occur on the food surfaces. Roasting both naturally and artificially contaminated samples of cornmeal for 15 min at 218 °C caused nearly a total loss of the available fumonisins [12]. After roasting pistachio nuts for 120, 60 and 30 min at 150, 120 and 90 °C, respectively, aflatoxins in the nuts reduced by 17 to 63% and the reduction appeared as time and temperature dependent. Whole pistachio kernels, naturally contaminated with aflatoxins showed significant reduction of aflatoxins levels after roasting time of 30 min at 150 °C. In pistachio, roasting for 120 min at 150 °C degraded at least 95% of aflatoxin B<sub>1</sub> [13]. Aflatoxins reduction during the roasting of coffee bean was reported to depend on the temperature and type of roasting with decrease of 42-56% accomplished [14]. About 13 to 93% ochratoxin reduction in coffee can be achieved using roasting [15]. Roasting for 6-10 min using electric-powered oven resulted in reduction of about 40% mycotoxin levels, however, extending to 15 min duration resulted in nearly 75% reduction [16]. As high temperature treatment, roasting results in considerable destruction of some mycotoxins. However, after roasting, the remaining levels of these toxins could still pose danger, especially when within unsafe levels. Also, not all grain products can be roasted, including grain flours. Roasting at high temperature to reduce or remove mycotoxins can lead to massive loss of nutrients and other bioactive compounds present in foods.

**Baking:** Baking is a cooking process which exposes foods to dry heat usually in oven, powered by electricity, wood, coal, solar, or other sources of energy. Primarily, baking is used for preparing bread, quiches, tarts, pies, pastries and cakes. Baking can also be used for preparing baked beans, baked apples, baked

TABLE-1  
EFFECTIVENESS OF FOOD PROCESSING METHODS ON MYCOTOXIN REDUCTION

Processing method/ technique	Foods commonly applied to	Effectiveness	Ref.
Baking	Grain flour products, bread, pasta, breakfast cereals, <i>etc.</i>	In the pilot scale baking of Vienna and French bread, fermentation at 50 °C led to 56% and 41% reductions, respectively, in the levels of deoxynivalenol (150 mg/kg). Mycotoxins losses in baking are very low and vary based on the particular mycotoxin and treatment duration.	[22]
Cleaning	Grains, nuts, seeds, kernels, <i>etc.</i>	About 40 to 80% reduction in aflatoxins could occur using physical cleaning, removing nuts, seeds, or kernels damaged by molds from the whole grains	[4]
Cold plasma	Nuts, palm fruits, grains, <i>etc.</i>	A study used cold plasma and reported that the concentrations of deoxynivalenol and zearalenone reduced from 100 µg/mL to a handful µg/mL. A study was done on nuts with low-pressure cold plasma, where a?atoxins reduced by 50% from the food surfaces	[42-44]
Combined treatments	Grains, seeds, tubers, nuts, spices, vegetables, kernels, <i>etc.</i>	Combination of various processing treatments such as cleaning, washing, soaking, cooking, frying, <i>etc.</i> Can effectively reduce mycotoxin levels. A blend of washing and boiling of barley caused sequential losses in levels of mycotoxins. A study reported 0.347 ppm initial NIV levels in unwashed barley, which when washed first and second times reduced to 0.152 ppm and 0.066 ppm, respectively and then boiling the barley reduced the levels to 0.060 ppm	[17]
Cooking	Grains, seeds, nuts, spices, vegetables, kernels, <i>etc.</i>	Ochratoxin A reduction efficiency showed highest reductions in the mycotoxin were found after cooking rice samples in excess water (86.6%), while normal cooking and microwave cooking resulted in 83.0% and 82.4% respectively	[54]
Cornflake processing	Grits, bran, <i>etc.</i>	Ochratoxin levels were reduced after cornflake processing. While decrease in fumonisins during production of corn flake took place, presence of so-called masked or hidden fumonisins (protein bound) was reported in commercial samples of corn flake from retail shops	[32,33]
Deoxynivalenol	Grains, seeds, kernels, <i>etc.</i>	Scouring has been shown to reduce levels of DON by 22% from initial 12.5 mg/kg level	[9]
Extrusion cooking	Nuts, cookies, snacks, <i>etc.</i>	Extrusion cooking reduces levels of mycotoxins at rates which depend on various factors including the type of screw, type of extruder, use of additives, the raw material moisture content, screw speed, barrel temperature, initial concentration of mycotoxin and die configuration	[45,49]
Fermentation	Grains, seeds, vegetables, <i>etc.</i>	Patulin is inactivated by the fermentation process and so is not seen in apple beverages, such as cider. Wastes generated after alcohol fermentation had low mycotoxins concentration due to fermentation activities and ratio of decomposition of main mycotoxins in the waste had order of ZEA< NIV< DON	[17,61]
Frying	Grain products, seed products, <i>etc.</i>	High frying temperature result in decrease in fumonisins levels, however, DON being heat stable, show low reduction during the frying process. Frying wheat contaminated with DON (1.2 mg/kg) at 169, 205 and 243 °C for 15, 2.5 and 1.0 min, respectively, were found not to have any significant DON reduction effects	[22]
Milling	Grains, seeds, nuts, <i>etc.</i>	Milling effect on wheat flours was reported when the initial DON level was not determined but following modern milling level of DON was 0.36 mg per kg, while levels of DON in bran, shorts, reduction flour and break flour were 122.1, 84.4, 16.9 and 16.2 mg per kg, respectively. Milling results in the mycotoxin redistribution, with lesser levels in the flour richer fractions and higher levels in the bran richer fractions	[38-40]
Parboiling	Grains such as rice, seeds, <i>etc.</i>	The soaking, precooking and overall steeping processes rice undergo under commercial parboiling could either increase rice grains vulnerability to fungal exposure or convey these toxins deeper into the grains. Aflatoxins levels in grains increased significantly with increased soaking duration. Parboiling should not be considered as alternative processing for mycotoxins reduction	[64]
Roasting	Grains, seeds, coffee, kernels, <i>etc.</i>	In pistachio, roasting for 120 min at 150 °C degraded at least 95% of aflatoxin B1. Aflatoxins reduction during the roasting of coffee bean was reported to depend on the temperature and type of roasting with decrease of 42-56% accomplished. Roasting for 6-10 min using electric-powered oven resulted in reduction of about 40% mycotoxin levels, however, extending to 15 min duration resulted in nearly 75% reduction	[13,14,16]
Screening	Grains, seeds, nuts, spices, kernels, <i>etc.</i>	DON was reduced by 16% with screening, which showed 4.7-fold greater DON levels than cleaned soft US wheat done with air flow and screening combined	[7]
Sorting	Grains, seeds, nuts, spices, kernels, <i>etc.</i>	Sorting decreases the concentrations of mycotoxins in foods and feeds	[1]
Use of chemicals	Grains, seeds, spices, nuts, <i>etc.</i>	A number of chemicals used in treating cottonseed and peanut meals so as to inactivate aflatoxins, include ammonia, calcium hydroxide, formaldehyde, sodium hypochlorite, methylamine, hydrogen peroxide and ozone. Alkaline cooking (nixtamalization) reduced concentrations of FB1 (50-80%) via a combination of hydrolysis and extraction. Deoxynivalenol was reduced significantly because it is not stable in alkaline environment. Chemicals used promised substantial reduction in levels of mycotoxins	[69,70,72]

potatoes, *etc.* [17]. For aflatoxins, evaluation of corn muffins produced from aflatoxins contaminated corn meal showed that 87% of the initial AFB1 levels in the cornmeal remained in the muffins after baking [18]. Baking biscuits, cookies and bread resulted in a deoxynivalenol level reduction of about 35% in cookies and biscuits and varied decrease from about 71-24% in bread, but gave no deoxynivalenol reduction in Egyptian flat bread [5,19,20]. Production of cookies by baking with standard recipe reduced level of deoxynivalenol (0.28 to 0.44 mg per kg) by 35% [21]. Baking using approved methods of the American Association of Cereal Chemists appeared not to noticeably reduce deoxynivalenol (0.52 to 0.31 mg per kg), however, there was variation in effects on its concentrations in the analyzed samples, the decrease ranged from 19-69% [8]. In the pilot scale baking of Vienna and French bread, fermentation at 50 °C led to 56% and 41% reductions, respectively, in the levels of deoxynivalenol (150 mg/kg) [22]. Although several studies reported that baking reduced the levels of deoxynivalenol, in contrast to other studies that (baking) alone did not result in considerable reduction levels. Baking at 205 °C for 30 min had no destructive effects on deoxynivalenol (about 4 mg/kg) [5]. Elsewhere baking at 170 °C for 30 min had no effect on the levels of deoxynivalenol (0.17 mg/kg) [23]. Additionally, baking at 210°C for 14 min had insignificant effects on deoxynivalenol levels (0.09 to 2.99 mg per kg) [24]. Baking Egyptian bread at 350 °C for 2 min did not reduce deoxynivalenol levels [19]. Study elsewhere showed that level of deoxynivalenol in bread did not reduce, however, the bio-toxicity was decreased significantly. This specific study shows that in bread during process of cooking such a new complex possibly form of deoxynivalenol binding carbohydrate or deoxynivalenol-binding protein has less cytotoxic potency than the deoxynivalenol itself [25]. Castelo *et al.* [26] studied fumonisins in some grains and reported that mixture of corn and muffin, which were artificially contaminated with FB1 (5 µg/g) and the ones that were naturally contaminated indicated insignificant fumonisins losses after baking. A different study reported that baking corn muffins for reasonable time at 200 and 175 °C led to 28% and 16% fumonisins reductions, respectively, with FB1 losses reported to be higher at surface compared to core of muffins [27]. Ochratoxin was reported to be stable during baking of bread, without any loss or decrease in its concentrations [11, 28]. Study also reported that baking of biscuits causes roughly 63.5% of toxin destruction or immobilizing [28]. All these research results clearly show that the mycotoxins losses in baking are very low and would vary, based on the particular mycotoxin type and treatment duration. However, baking for a longer time at very high temperature simply to destroy mycotoxins can result in excessive charring, poor quality products and/or excessive nutrient loss.

**Cornflake processing:** Cornflake processing has been reported to have some effects on mycotoxins. Cornflake processing effects on mycotoxins such as fumonisins and aflatoxins have been studied [29,30]. Cooking the aflatoxins contaminated grits, without or with addition of sugars caused 64 to 67% aflatoxins reductions. After flakes toasting without or with addition of sugars, aflatoxins reductions were 78-85% [31].

Ochratoxin levels were reduced after cornflake processing [32]. Castelo [12] reported that corn flake process with no added sugars caused 48.7% and 53.5% losses in fumonisin levels after toasting and cooking, respectively and 86% to 89% decrease when glucose was added. Fumonisins B<sub>1</sub> and B<sub>2</sub> stability during corn flake processing have been studied as well. Results showed that 60% to 70% decrease in fumonisin levels occurred, however 30% of the losses were associated with the extrusion stage, in which the materials were exposed for 2 to 5 min at 70-170 °C [30]. In a different study, a researcher studied the effects of cornflake process, gelatinization and extrusion cooking on fumonisins stability (B<sub>1</sub> and B<sub>2</sub>) and concluded that gelatinization and extrusion cooking reduced the levels of fumonisin to about 30 to 55%, whereas grits cooking for flaking resulted in the reduction of the fumonisins levels to 20 to 65% and fumonisins were reduced to 6-35% after roasting the flakes [29]. FB1 losses in the presence of high fructose corn syrup, maltose and sucrose mimicked those that occurred in corn flakes produced with no sugar. While the decrease in fumonisins during production of corn flake took place, presence of so-called masked or hidden fumonisins (protein bound) was reported in commercial samples of corn flake from retail shops [33]. Aflatoxins and fumonisins to significantly decrease as a result of cornflake processing are in literature. It is worthy to say that cornflake processing with sugars result in significant reduction in mycotoxin levels although the levels of these mycotoxins left after the processing may still remain within unsafe levels. Further studies are required to evaluate how the process can be improved to ascertain mycotoxin reduction to safe levels or complete elimination.

**Milling:** Milling is a process where grains are crushed or reduced in sizes to obtain flour. There are various stages involved in milling process. Milling can be done wet or dry. Corn subjected to wet milling resulted in 49% and 96% decrease in levels of ochratoxins in grits and germ, respectively [34]. Deoxynivalenol (DON) (2.0 mg/kg) contaminated soft wheats subjected to dry milling showed that the mycotoxin was distributed all over the parts of milled grains; shorts and bran [35]. Study on DON (0.17 mg/kg) contaminated wheat, simultaneously contaminated with ZON and NIV milled using Buhler experimental mill indicated that up to 60% of DON remained in the flour post-milling. Levels of DON in the bran was almost three times the levels in original wheat [23]. Similarly, wheat milling showed 24% to 48% decrease in DON in fractions of flour with 0.068 mg/kg initial DON level meant for consumption by human [36]. Compared to DON levels in cleaned wheat grains, there were 77% at those of uncleaned wheat grains, noting that initial levels of DON were 0.3-13.1 mg per kg [37]. Meanwhile, milling adverse effect on wheat flours was reported in another study. Although the initial values were not determined, the milling activity produced the level of DON at 0.36 mg per kg [38], whereas those in bran, shorts, reduction flour and break flour were 122.1, 84.4, 16.9 and 16.2 mg per kg, respectively [39]. Despite the notable differences in the above-mentioned studies, there appears to be one common fact, which is that the wheat bran has higher DON level. The bran deoxynivalenol level was above those in initial values of the wheat [37,39].



The contamination level of deoxynivalenol does not reduce during the milling process. Unfortunately, it results in the toxin redistribution, with lesser levels in the flour and higher levels in the bran richer fractions. As these bran fractions are used in foods meant for consumption by human, milling should not be completely considered as an efficient way of decontamination of mycotoxins in grains or grain products [40].

**Cold plasma:** A cold plasma, also called non-thermal plasma, which is not in thermodynamic equilibrium, due to the temperature of the electron is very hotter than the heavy species (neutrals and ions) temperature. In food industries, cold plasma appears associated with antimicrobial treatment, protein modification, enzyme inactivation, *etc.* and considered effective in eliminating pathogens and several toxic metabolites of microorganisms. Previous reports that have shown the potentials of strong antimicrobial effect of cold plasma, which points to its application to sterilize temperature-sensitive surface, including foods [41]. Cold plasma is primarily generated through atmospheric dielectric discharges, using working gas made up of synthetic air. A study used cold plasma and reported that the concentrations of deoxynivalenol and zearalenone reduced from 100 µg/mL [42]. Foods processed using plasma have to be evaluated for development toxic substances, which points to the need for more studies especially on the cold type. There are limited studies on use of cold plasma to reduce mycotoxins in foods. Basaran *et al.* [43] reported nuts subjected to low-pressure cold plasma, where aflatoxins reduced by 50% from the food surfaces. Elsewhere, palm fruits were subjected to argon cold plasma under atmospheric pressure, to tackle *A. niger* spores. After 9 min treatment, all the spores died and other mycotoxins levels appeared undetected, probably because they were below (detection) limits [44]. There are insufficient studies on use of cold plasma to reduce mycotoxins in foods, possibly because not all foods could be subjected to cold plasma treatment, cost and high technicality required, among others. Cold plasma alone would not be very reliable to reduce the mycotoxins to safe levels. The remaining percentage of mycotoxins after cold plasma treatment can still result to chronic mycotoxicosis.

**Extrusion cooking:** Recently, extrusion cooking is gaining attention as among the food processing operations with promoting recognition worldwide given its many advantages over several conventional methods. In addition to its major objective to improve quality of final and intermediate processed foods, it could unintentionally improve food safety due to its potential to reduce levels of toxins there in, including mycotoxin [45]. Aflatoxins could be reduced by 50% to 80% using extrusion [46]. Similar results were reported when meal of peanut was processed using extrusion cooking and gave reduction of 23% to 66%. Aflatoxins reductions of up to 95% has been shown after cereals extrusion cooking [45]. For deoxynivalenol, extrusion experiments were shown to make safer the DON-contaminated corn flour [47]. Studies showed that spiked deoxynivalenol has stability in extruded pet foods and corn grits, with 12% DON reduction reported [48]. DON was reported to be stable at the pressures and temperatures

applied in processes [48]. In wheat, DON reduced significantly after soaking with the addition of sodium bisulphite. Extrusion was reported effective in reducing DON levels (over 95%) in all conditions of assessment [49], although low decrease of 55% was reported for DON, after cereals extrusion cooking [45]. Generally, extrusion cooking reduces levels of mycotoxins at rates which depend on various factors including the type of screw, type of extruder, use of additives, the raw material moisture content, screw speed, barrel temperature, initial concentration of mycotoxin and die configuration [45].

**Cooking:** Cooking is a process of preparing food for consumption using heat application, usually carried out conventionally in water. Cooking methods could vary, but most often depend on traditions and customs, affordability and availability of required resources [50]. Relevant literature is available about the effects of cooking on the levels of mycotoxins in foods (grains and grain products). Normal cooking of AFB1 contaminated rice was reported to show 34% average reduction. Pressure cooking caused better decrease of 78% to 88% [51,52]. In a different study, corn grits boiling resulted in aflatoxins reduction by 28%. AFB1 levels in dried wheat reduced to 90% and 50% after heating at 200 and 150 °C, respectively [53]. Comparing three different cooking forms for AFB1 reduction efficiency appeared highest after cooking rice samples in excess water (87.5%), whereas normal cooking and microwave cooking resulted in 84.0% and 72.5% reductions, respectively [54]. After boiling and draining for 10 and 5 min, respectively, DON (12.5 mg/kg) contaminated noodles showed 49% decrease, while the same process in another study with noodles reported 40% decrease after boiling [9]. Fumonisin B<sub>1</sub> is fairly stable in heating at temperature of boiling. No FB1 loss was reported after boiling *Fusarium verticillioides* culture for 30 min in water, following by drying for 24 h at 60 °C [55]. At very high temperature ranges, reduction could occur. Autoclaving oatmeal using 50% water resulted in 74% ochratoxin reduction, whereas autoclaving rice cereal or dry oatmeal resulted in greater losses, from 86 to 87.5% [56]. Cooking effect on ochratoxin A reduction in *Phaseolus vulgaris*, a bean variety "Carioca" has been studied after inoculation of suspensions of spores of *Aspergillus alutaceus*, an ochratoxigenic strain. Samples were taken after 10 days, analyzed for ochratoxin A levels and cooked using pressure, with and without prior soaking. The results showed that cooking noticeably decreased the ochratoxin A levels (about 84%). The effects appeared more effective when the bean was soaked for 12 h in the water prior to cooking for 45 min at 115 °C under pressure [57]. It can be said that significant losses in mycotoxins such as aflatoxins and ochratoxins can be achieved with cooking. While fumonisins appear to have stability at low temperatures of cooking, levels of DON showed lower reductions after cooking. While cooking may significantly reduce levels of some mycotoxins, other existing processing methods or even when combined can be explored to ensure mycotoxins are relatively reduced to safe levels or completely degraded/removed. Human exposure over a long period of time could still pose the risks of mycotoxicosis.

**Combined treatments:** Foods and feeds, including grains, generally undergo processing, which usually make use of use

of a combination of various processing treatments such as cleaning, washing, soaking, cooking, frying, *etc.* A blend of washing and boiling of barley caused sequential losses in levels of mycotoxins. A study reported 0.347 ppm initial NIV levels in unwashed barley, which when washed first and second times reduced to 0.152 ppm and 0.066 ppm, respectively and then boiling the barley reduced the levels to 0.060 ppm [17]. Dried beans washed using pure water for 120, 60, or 2 min, after which they were soaked for 10 h, 120 min or 60 min in water and then cooked for 120 or 60 min. The three treatments jointly removed approximately 50% of toxins in the beans [18]. The soaking, washing and cooking resulted in significant decrease in ochratoxin A levels in the dried beans. Therefore, the combination of different processing treatments would noticeably reduce the levels of mycotoxins. However, a combination of several processing treatments require further investigations to ascertain which combinations are more efficient in mycotoxin reduction, *viz.-a-viz.* maintaining the nutrient compositions and other bioactive constituents in the foods.

**Fermentation:** Fermentation is a process which make use of microorganisms during which starch is saccharified, yielding simple sugars that are primarily fermented to alcohol, lactic acid, carbon dioxide, *etc.* also, enzymes breakdown proteins to peptides of low molecular weight and amino acids [59]. Fermenting dough made with wheat flour reduced aflatoxins by about 50% [60]. Barley fermentation to produce alcohol reduced levels of mycotoxin by 100% (from initial level of 16.32 ppm to not detected). Wastes generated after alcohol fermentation had low mycotoxins concentration due to fermentation activities and ratio of decomposition of main mycotoxins in the waste had order of ZEA < NIV < DON [17]. Patulin is inactivated by the fermentation process and so is not seen in apple beverages, such as cider [61]. Cereals fermentation is very effective in reducing mycotoxin levels. Fermentation when properly optimized have been shown to be effective in reducing mycotoxin levels. However, only small number of foods and grain products require fermentation. Also, fermentation has not been shown to reduce all types of mycotoxins to safe levels. For example, fermenting dough made with wheat flour only reduced aflatoxins by about 50%.

**Frying (deep-frying and sautéing):** Frying involves cooking foods in oil or any other fat, which may take many forms, such as deep-frying, where foods are totally immersed in heated oil, and the process occurs in frying pan containing a thin oil coating. Frying remains among the fastest ways of cooking, as it efficiently transfers heat into the foods. In spite of using oil, frying has been regarded as dry cooking method given the absence of water in the process. Thus, in an ideal frying process, the food does not absorb the cooking oil [62]. Corn masa frying for 0-6 min at 140-170 °C did not result in fumonisins reduction, whereas frying tortilla chips for 15 min at 190 °C led to 67% fumonisins reduction [27]. Conversely, frying wheat contaminated with DON (1.2 mg/kg) at 169, 205 and 243 °C for 15, 2.5 and 1.0 min, respectively, were found not have any significant DON reduction effects [63]. Therefore, high frying temperatures result in decrease in fumonisins levels, however, DON being heat stable, show low reduction during

the frying process. Frying alone should not be seen as the only reliable way of reducing mycotoxins to safe levels. Frying at high temperature for long duration could have destructive effects on the contents of the foods, including nutritional composition.

**Parboiling:** Parboiling is a food processing technique that entails partially cooked water over a short time period complete cooking. Parboiling has some benefits including reduction in time of cooking when parboiled ingredient is added to foods. Rice is among the most commonly parboiled grains/ingredients. Aflatoxins levels in rice made through commercial parboiling (60-92 mg/kg AFB1) were found to be far higher compared to that in 'cottage' processed rice (12-29 µg/kg AFB1). The soaking, pre-cooking and overall steeping processes rice undergo under commercial parboiling seems to increase rice grains vulnerability to fungal exposure or helps to convey these toxins deeper into the grains. Aflatoxins levels in grains increased significantly with increased soaking duration [64]. Parboiling should not be considered as an alternative processing for mycotoxins reduction.

**Use of chemicals for treatment:** Aflatoxins inactivation using chemical treatment seems to provide the most feasible measure. There are two points in molecules of aflatoxins, which appear most vulnerable to attack by chemicals: the terminal furan double bond (when present) and the coumarin moiety internal ester. Several chemicals have been used to treat cottonseed and peanut meals in order to inactivate aflatoxins, including ammonia, calcium hydroxide, formaldehyde, sodium hypochlorite, methylamine, hydrogen peroxide and ozone [65]. Levels of deoxynivalenol in corn reduced by 95% through autoclaving for 1 h at 121 °C using 8.33% sodium bisulphite (aqueous) [66]. Adding calcium hypochlorite at 10 ppm to water used for soaking substantially reduced the effect of *A. flavus*, demonstrated by the reduction in aflatoxins levels in rice after parboiling [64]. Effects of additives used in bread on levels of deoxynivalenol post-bake showed that L-ascorbic acid and potassium bromate appeared unaffected. However, the application of ammonium phosphate, l-cysteine and sodium bisulphite showed a 40% decline in the levels of deoxynivalenol [67]. Interestingly in that study, isoDON, the metabolite of DON, was produced in the process [67]. Only extrusion cooking on the other hand, has been shown to have reduced the aflatoxins levels by 50% to 80%, however, ammonia was added, either as or bicarbonate (0.4%) or as hydroxide (0.7 and 1.0%), the reduction of aflatoxin levels accomplished was 95% [46]. Another study showed similar results after peanut meals were extruded in the presence of 2 to 2.5% (87% decrease) or absence (23 to 66% decrease) of ammonium hydroxide [68]. Alkaline cooking (nixtamalization) reduced the concentrations of FB1 (50-80%) *via* a combination of hydrolysis and extraction. Deoxynivalenol was reduced significantly given its instability in alkaline environment [69]. Corn has been processed into tortillas using nixtamalization by some researchers. The tortillas had FB1 of about 0.50 ppm, plus HFB1 of 0.36 ppm, which was 18.5% of initial concentration of FB1 (8.79 ppm). Another group of researchers [70] reported that the process of nixtamalization substantially reduced fumonisins levels in the

maize. Sorghum flour with aflatoxins contaminated of 140 ppb were extruded with aqueous citric acid or lactic acid at six concentration levels. Reduction in levels of aflatoxins was more effective when 92% citric acid (aqueous) was used, than when 67% lactic acid (aqueous) was used [71]. Effects of eighteen different chemicals, including oxidizing agents (sodium hypochlorite and hydrogen peroxide), salts (sodium sulfate, sodium chloride, sodium hydrosulfite, sodium bisulfite and acetate ammonium), alkaline compounds (calcium hydroxide, potassium hydroxide, sodium hydroxide, sodium bicarbonate and ammonia) and acidic compounds (acetic acid, citric acid, benzoic acid, phosphoric acid, chloridric acid and sulfuric acid), on reduction in levels of AFB<sub>1</sub>, AFB<sub>2</sub>, AFG<sub>1</sub>, AFG<sub>2</sub> and ochratoxin A were studied in white and black pepper. Nearly all of chemicals used indicated substantial reduction of levels of mycotoxins. Adding to the highest and lowest decrease in AFB<sub>1</sub>, the most toxic aflatoxins, was 54.5% with sodium hydroxide and 20.5% with benzoic acid. No significant difference occurred between white and black peppers [72]. Use of chemicals or their combination with other methods/techniques can be effective in reducing mycotoxins to safe levels.

**Analytical detection of mycotoxins in grains (merits and demerits):** A number of methods/techniques have been used to analyse mycotoxins, which include the spectral analysis technique (SAT), liquid chromatography-mass spectrometry (LC/MS), high-performance liquid chromatography (HPLC), thin-layer chromatography (TLC) and enzyme-linked immunosorbent assay (ELISA) [73-81]. The SAT has merits such that it reveals the quantitative and qualitative information about the mycotoxins structure and quick screening of large sample size can be performed. However, its demerits are that there could arise issues like spectra overlapping, cross-reactivities with a related mycotoxin, given that it is semi-quantitative, with a narrow range of operation. Also, sometimes, the reading of spectral data could be complicated, given by problems of matrix interference [82]. LC/MS on the other has merits as well. For example, it has a sensitive and selective detection and can generate information about analyte structure, with low detection limits. However, the disadvantage of LC/MS is that of being very expensive and requires specialist expertise to operate [79,83]. Moreover, LC/MS is not much different from HPLC, in addition to that, it may require derivatization. The merit of HPLC includes good selectivity, short analysis time given its high sensitivity, presence of an autosampler. Others include its precise identification and its automated platform [76,78,79,83]. TLC, although it also requires specialist expertise to operate, has merits like precise identification, automated and good selectivity, with ease in identification of mycotoxins [75,79,80,83]. On the other hand, whilst ELISA's sensitivity depends on the technique of ionization, the merits include ease of operation, minimum requirements for preparation of the sample, sensitive detection and convenience in its operational steps. It can also simultaneously analyze several mycotoxins given its rapid screening of samples requiring limited organic solvents [73,74,77,80,81]. Consequently, novel precise methods and techniques for identifying and quantifying mycotoxins continue to be developed so as to eliminate and or reduce the

levels of mycotoxins, reducing limit of detection, improving identification accuracy, among others.

## Conclusion

Aflatoxins, ochratoxins, patulin, citrinin, deoxynivalenol (DON), fumonisins, trichothecenes, *etc.* being mycotoxins produced by molds such as *Aspergillus avus*, *A. parasiticus*, *Penicillin* spp, *etc.* remain commonly found crops like peanuts, millet, sunower, cassava, beans, sesame, acha, sorghum, maize, *etc.* In this article, an overview of the effectiveness of grain processing methods to eliminate mycotoxins were successfully performed. For emphasis, the processes considered in this review included extrusion, alkaline cooking, flaking, roasting, frying, baking, milling, cooking, cold plasma, cleaning, trimming, sorting, as well as chemical treatments such as use of sodium hydroxide, citric acid, sodium bisulfite, ammonia, *etc.* Consequently, efforts have to be consistent to increase the precision of novel precise methods and techniques for identifying and quantifying mycotoxins. These efforts have to continue to increase the focus to eliminate and or reduce the levels of mycotoxins, as well as reduce the limit of detection, which would improve identification accuracy, among others. Moving forward, the efficiency and reliability of grain processing methods require further exploration, particularly to identify other methods or combinations of methods to ensure complete removal/degradation of all mycotoxins or at least reduce them to safe levels.

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## CONFLICT OF INTEREST

The authors declare that there is no conflict of interests regarding the publication of this article.

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