***Review Article***

**Fungal pathogen and mycotoxin management in agriculture and food processing**

**Abstract:**

Fungi may cause significant food deterioration and have a high growth tolerance over a wide range of food items as well as an agriculture. When fungus colonise food, they release poisonous secondary chemicals known as mycotoxins. These are strong toxins that are carcinogenic, teratogenic, and mutagenic, with grave health implications for humans. Mycotoxin contamination is increasingly common in agricultural goods. Mycotoxins are produced by a number of fungi on agricultural goods during or after harvest, and they are extremely harmful to both humans and animals. Aspergillus species create aflatoxins and ochratoxins, Penicillium produces ochratoxins and patulin, and Fusarium species produce fumonisins, deoxynivalenol, and zearalenone. These are the most common mycotoxins detected in food items. A number of acute and chronic disorders in humans and domestic animals are caused by fumonisins, patulin, aflatoxins, and ochratoxins, among other substances. Mycotoxins have been measured in dietary items. Modern detoxification techniques also need to enhance food product quality and be suitable from a financial and environmental standpoint. This Chapter examined innovative preventative methods for controlling fungi and mycotoxins that might jeopardise food safety and consumer health. As a result, among the green and emerging technologies examined were ionising and non-ionizing radiation, cold plasma (CP), pulsed light (PL), ultrasonic (US), pulsed electric field (PEF), and high-pressure processing (HPP). The main effects of using these novel technologies are destroy the structure of mycotoxins, significantly reduce the growth of mycotoxigenic fungus, and greatly preserve the nutritional value of food.

**Keywords:** Mycotoxin, agricultural goods, aflatoxins, cold plasma, pulsed light, ultrasound, pulsed electric field and high-pressure

**Introduction:**

Mycotoxins are harmful secondary metabolites that fungus make when they colonise food sources. These powerful poisons are carcinogenic, teratogenic, and mutagenic, with serious negative effects on human health. Global pre-harvest and post-harvest losses of agricultural commodities are estimated to be between 30% and 50%. In addition to endangering food security around the world, this practice also results in the loss of arable land ranging from 1.47 to 1.96 Gha, water from 0.75 to 1.25 trillion cubic meters, and energy from nearly 1% to 1.5% of the globally (Fox and Fimeche, 2013). Preharvest and postharvest losses in Agricultural crops and food commodities can be caused by a variety of biotic and abiotic causes. Fungal bio-deterioration of food commodities in storage systems is a persistent issue in tropical hot and humid condition area. Mycotoxins are produced by a number of fungi on agricultural goods during or after harvest, and they are extremely harmful to both humans and animals. Aspergillus species create aflatoxins and ochratoxins, Penicillium produces ochratoxins and patulin, and Fusarium species produce fumonisins, deoxynivalenol (DON), and zearalenone. These are the most common mycotoxins detected in food items. A number of acute and chronic disorders in humans and domestic animals are caused by fumonisins, patulin, aflatoxins, and ochratoxins, among other substances. Mycotoxins have been measured in food commodities and detected using various analytical methods.

Mycotoxins are produced by fungi that contaminate a variety of agricultural commodities either before or after harvest (FAO, 1991). Fungal proliferation and the production of mycotoxins are caused by tropical conditions like high temperatures and moisture, monsoons, unseasonal rains during harvest, and flash floods (Bhat and Vasanthi, 2003). Inadequate storage, improper harvesting techniques, and subpar conditions during transportation, marketing, and process. The majority of Africa shares these climatic traits as well as food production networks. As a result, there is a genuine risk that mycotoxins contamination of feeds and food can cause poisoning in humans as well as animals, which should be taken very seriously.

Over the past five years, there have been reports of the greatest mycotoxin-poisoning pandemic in a decade in Africa (Lewis *et al*., 2005; CDC, 2004). The US Food and Drug Administration (FDA) views some mycotoxins, such aflatoxins, as inevitable food contaminants. Therefore, minimising contamination has been the aim. However, due to the features of those nations' food systems and technical infrastructure, mycotoxin levels in developing nations are uncontrollably high, making it impractical to apply mycotoxin management techniques that are effective in affluent nations. The problem is exacerbated by the fact that cereal grains like maize, which are extremely vulnerable to mycotoxin contamination, constitute a mainstay diet in many African homes. In Africa and other tropical developing nations, aflatoxins and fumonisins are the food-borne mycotoxins that are most likely to be significant (WHO, 2006). Due to their substantial negative effects on human health, animal production, and trade, mycotoxins are gaining attention on a global scale (WHO, 2006; Wu, 2006).

Consumption of mycotoxin-contaminated food items has a negative impact on human and animal health, which become lowers the marketability of food commodities and raises questions about food safety (Soares *et al*., 2021). According to estimates, about five billion individuals eat contaminated food on a daily basis and are exposed to mycotoxins through unidentified mechanisms (Khodaei *et al*., 2021). Mycotoxicosis, or intoxication caused by consuming mycotoxins through food, can occur (Tanaka *et al*., 2007). When a mycotoxin causes acute or long-term toxicity involving hepatotoxicity, cytotoxicity, teratogenicity, neurotoxicity, mutation, and carcinogenicity, it is known as mycotoxicosis. Mycotoxins interact with nucleic acids to prevent DNA and RNA production at the cellular level (Smith *et al*., 2007).

In addition, the presence of mycotoxins in food products was eliminated using a variety of techniques, including chemical, biological, and physical ones. The results of thirty years of study on mycotoxins in important commercial food crops such as wheat, maize, sorghum, pearl millet, peanuts, oats, pulses, barley, oilseeds, rice, fruits, and fruit juices are compiled in this review. We also go over the various decontamination techniques, their drawbacks, and knowledge gaps, as well as the techniques for detecting the main mycotoxins. Data from thorough investigations on mycotoxins in food commodities are expected to be useful in determining research priorities and in the production of safer food.

There have been several reports on decontamination techniques for items containing fungi or to lower mycotoxins. But not every strategy is appropriate for the food sector. Certain features are necessary for an effective process to inactivate the fungi or remove the variety of mycotoxins from food, including high efficiency without producing toxic compounds or jeopardising technological or rheological properties, maintaining the nutritional values and minimising nutrient loss, and retaining the appetising qualities of food products. Thermal methods like pasteurisation and sterilisation reduce the amount of food deterioration brought on by fungus, but they also negatively impact food items' nutritional value and flavour, making them less appealing.

**Classification of foodborne mycotoxins**

 The mycotoxins generated by fungi are primarily categorised as follows: fumonisins (FBs), ochratoxin A (OTA), patulin (PT), sterigmatocystin (STC), zearalenone (ZEA), alternariol (AOH), tenuazonic acid, and alternariol monomethyl ether. **TABLE-1** lists several significant mycotoxins, the food products they contaminate, and the harmful consequences they cause. Mostly generated globally by *A. flavus* and *A. parasiticus*, **aflatoxin** is a very dangerous mycotoxin (Pandey *et al*., 2016). There are four main kinds in it: B1, B2, G1, and G2.

**Aflatoxin**

**Fig.1: Aflatoxin**

There are four main kinds in it: B1, B2, G1, and G2. It has been established that *A. flavus* generates B toxins, the most prevalent of which is the genotoxic and carcinogenic B1 (Payne and Brown, 1998; Abbas *et al*., 2008). While AFs from *A. flavus* are more prevalent in maize and cotton than in other crops (Hell *et al*., 2000), AFs from *A. parasiticus* are more common in groundnuts (peanuts) (Kaaya *et al*., 2006). ZEA, another mycotoxin, is sometimes referred to as F-2 mycotoxin. Numerous Fusarium species generate toxic compounds that are of great concern to producers of cattle and poultry, including DON, T2, and HT-2 toxins, ZEA, and diacetoxyscirpenol (DAS) (Kuiper-Goodman *et al*., 1987). As seen in Table 1, a variety of food products may be impacted by these pollutants. It can lead to infertility, abortion, and other breeding issues, particularly in pigs (Kuiper-Goodman *et al*., 1987).

 **Fig.2: Fumonisins**

Source: [Anne Koontz](https://www.alltech.com/about/senior-leadership/anne-koontz) (2021)

Fumonisins, a class of mycotoxins made up of FB1 and FB2, are generated by Fusarium. *Fusarium verticillioides* is a family of toxin-producing Fusarium moulds that mostly attacks grains, including wheat, maize and many others. Its most common member is called FB1. Additionally, it has been demonstrated that *F. moniliforme* and *F. verticillioides* generate FB2, which shares structural similarities with FB1. (Polisenska *et al*., 2020). In contrast to FB1, FB2 inhibits acyl sphingosine transferase and is more cytotoxic. FB2 also contaminates maize and other commodities. To date, fifteen distinct FBs have been reported; however, the majority of them have not been demonstrated to occur in the natural world (Polisenska *et al*., 2020).

Source: Purity coffee

**Fig.3: Ochratoxins**

Regarding ochratoxins, food commodities include three different forms of ochratoxins: OTA, OTB, and OTC. Produced by *Penicillium verrucosum* and species of Aspergillus, including *A. carbonarius* and *A. ochraceus*, OTA is the most common mycotoxin present in food (Al-Anati and Petzinger, 2006). Human contact may occur via consuming OTA-contaminated food items, such as coffee, cereals, pig products, grapes, and grape products (Richard *et al*., 1999). Furthermore, PT is produced by Aspergillus, Byssochlamys, and Penicillium and is generally associated with rotting apples. Research has demonstrated that PT exhibits antibacterial action against certain pathogens. Because of the health hazards, numerous nations have regulated the amount of PT in commodities as a consequence of health research.

**TABLE-1** List of significant mycotoxins, the foods that are most likely to get contaminated by them, and their main harmful consequences.

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| **SL No** | Mycotoxins | Related moulds | Symptoms/toxicology | Most contaminated food products | Ref. |
| 1 | Aflatoxins | *Aspergillus parasiticus, A. nomius,* and *A. flavus* | Depressed immune response, liver tumours, Liver necrosis, reduced growth, carcinogenic, hepatotoxic, mutagenic, teratogenic, vomiting, and pulmonary convulsions | Grain, cherries, strawberries, groundnut, raspberries, maize, peanuts, maize, cotton, pearl millet, sorghum, pistachios, chillies, cassava, oil seeds, spices, and dried fruits | Liu *et al*. (2006) |
| 2 | Cyclopiazonic acid | *A. flavus, A. oryzae,*  *A. versicolor, A. tamarii. P. patulum, P. verrucosum, P. camembertii, P. cyclopium, Penicillium griseofulvum,* and*P. puberulum* | Neurotoxin, cytotoxicity, weight loss, immunotoxicity, diarrhea, muscle, nausea, viscera necrosis, and convulsions | Groundnut, Maize, cheese etc. | Gonçalez *et al.* (2008) |
| 3 | Deoxynivalenol, Vomitoxin, Zearalenone | *Fusarium graminearum* and *F. subglutinans* | Diarrhoea, vomiting, decreased weight gain, feed refusal, infertility, hepatotoxic, genotoxic, immunetoxic, hemato-toxic, and oestrogenic effect | Wheat, maize, oats, rice, sorghum, and barley | Nakagawa *et al.* (2011) |
| 4 | Fumonisin B1 and Fumonisin B2 | *F. moniliforme* and *F. verticillioides* | Porcine pulmonary edema, equine leukoencephalomalacia, kidney disease, liver tumor, hepatotoxic, nephrotoxic, cytotoxic, and oesophagal cancer | Maize, rice, and wheat | Topi *et al.* (2021) |
| 5 | Trichothecenes | *F. culmorum, Trichoderma, F. graminearum, F. poae, Cephalosporium,* and *Trichothecium* | Food-related toxicities include aleukia, necrosis, mouth lesions in grill chickens, growth retardation, weight loss, vomiting, diarrhoea, haemorrhaging, fever, vertigo, and neurotoxicity. | Wheat, oats, and maize | Jimenez and Mateo (1997) |
| 6 | Ochratoxin | *A. ochraceus, P. verrucosum,* and *A. carbonarius* | Porcine nephropathy, genotoxicity, immunotoxicity, embryotoxicity, teratogenicity, neurotoxicity, and inhibitors of protein, RNA, and DNA synthesis are among the symptoms associated with poultry. | Wheat, spices, grapes, and coffee | Iqbal *et al.* (2018) |
| 7 | Patulin and Citrinin | *P. expansum* | Kidney damage, nephrotoxic,  immunotoxicity, teratogenic,  hepatotoxic, and foetotoxic | Apple, orange, grapes, and related products | Saxena *et al.* (2008); Oteiza *et al*. (2017) |
| 8 | Sterigmatocystin | *A. parasiticus, A. versicolor, A. flavus, A. nidulans, A. rugulosus, A. rubber, A. chevalieri, P. camembertii, A. amsyelodami, P. griseofulvum, P. communer* | Carcinogenic, mutagenic, immunotoxicity, cytotoxicity, diarrhea, nausea, and weight loss. | Maize, rice, wheat, and hay | Iqbal *et al*. (2018) |
| 9 | Alternaria toxins: alternariol, tenuazonic acid and others | *Alternaria* species | Cytotoxic, genotoxic, teratogenic, mutagenic, fetotoxic, and dermal toxicity | Grains, oil seeds, spices, and various fruits and vegetables | Dong *et al.* (2019) |

**Factors effect on fungal growth and mycotoxins production**

Primary factors that promote the growth of fungi and the generation of mycotoxin include pre-harvest handling techniques, unsanitary handling procedures during processing and shipping, and improper storage conditions, such as high moisture content, high temperatures, and heavy rains. Numerous elements have been identified by experts as favouring the formation of mycotoxins. These fall into three categories: chemical, biological, and physical. Environmental elements such as temperature, relative humidity, and insect infestation are examples of physical influences. While biological variables rely on the interactions between toxigenic fungus and substrate, chemical factors include the use of fungicides, insecticides, or fertilisers. Certain plant species are more vulnerable to fungal colonisation, whilst environmental factors may make certain plant species more resilient. Thus, factors can also be classified as extrinsic, intrinsic, implicit, processing, or water activity. These factors include moisture content, water activity, temperature, climate, oxygen level, type of substrate, type of plant, and nutrient composition; handling of grains, drying, blending, addition of preservatives, insect interactions, fungal strain, and microbiological ecosystem. The variables that influence the development of fungal growth and the formation of mycotoxins will be divided into many groups in this review based on pre- and post-harvest circumstances.

**Preharvest factors**

The most significant factors in determining the likelihood of preharvest contamination are those related to the type of soil, soil condition, drought, genotypes (plants bred to be resistant to fungal infection), plant density, amount of fertilisation, and insect activity. One natural factor that significantly affects the likelihood of fungal infection is soil. Crops cultivated in various soil types may significantly impact the amount of mycotoxin contamination and fungus development. For instance, peanuts cultivated in sandy soils encourage faster fungal development, particularly under dry conditions. Conversely, peanuts produced in heavier soils have less contamination because of their high water-holding capacity, which helps the plant withstand drought stress. The grade, quality, and market worth of agricultural products which are typically regarded as unfit for human and animal consumption are all negatively impacted when insects invade grains.

**Postharvest factors**

The post-harvest steps are the phases that come after harvest and precede primary processing, such as milling. Drying, storing, shipping, and processing procedures are among the post-harvest procedures that are seen to be crucial control points for tactics aimed at preventing mycotoxin contamination and fungal development. Food items' nutritional value can quickly decline as a result of poor post-harvest handling. In order to prevent fungal development and insect infestation and to extend the shelf life of agricultural products, it is imperative that crops be dried quickly to the lowest possible moisture content. If maize is kept at high moisture levels for three days, the contamination with aflatoxins might grow tenfold. It is generally advised that harvested crops be dried as quickly as possible to preserve the moisture level of 10–13%, which may be accomplished by simply sun-drying, in order to minimise fungal development and mycotoxin contamination. Fungal infection, insect infestation, and the buildup of mycotoxins can all happen during the crucial stage of storage. Inappropriate food handling practices or storage conditions can lead to mycotoxin contamination in feeds and food. Water activity (aW), substrate temperature, aeration, microbial interaction, insect activity, and rodent activity are some of the factors that affect the development of toxic fungus on grains during storage. In order to prevent the buildup of mycotoxins in grains that have been kept, moisture content control is crucial. Fungi often grow in environments with temperature ranges of 10–40 °C, pH values of 4–8, and aW levels greater than 0.70. Whereas *Aspergillus flavus* may germinate and grow at temperatures over 10ºC with aW ranging from 0.85 to 0.87, *A. ochraceus* can only develop at temperatures below 10ºC. On the other hand, *Penicillium expansum* may produce patulin at temperatures ranging from 0 to 24ºC. Different *Penicillium* species have different needs for moisture content; certain *Penicillium* spp. may thrive on soils devoid of water.

**Economic impacts of mycotoxins**

Mycotoxin-related economic losses can take many different forms, such as decreased availability of nutritious foods for humans and animals, decreased animal production due to feed rejection or illness, expensive medical expenses for treating toxicosis, high costs associated with finding substitute foods, advancements in detection and quantification techniques, and the creation of plans to reduce mycotoxin exposure. Mycotoxin contamination has significant negative economic effects when it comes to global trade. It lowers the price of crops and may result in significant food losses. In the United States, mycotoxin losses often amount to hundreds of millions of US dollars each year and are linked to both market expenses and adverse impacts on human health. The reduction in food availability, particularly in impoverished areas, regulatory rejection of exported goods, a decline in the market value of contaminated products in domestic markets, decreased marketability of crops that posed a clear threat to food security, as well as an increase in livestock, human diseases, and mortality, are all indicators of the economic impact of mycotoxin contamination in Africa. Additionally, this effect will raise the price of regulation and research initiatives meant to lower the possible dangers that mycotoxins pose to the health of people and animals.

**Mycotoxin regulations in food commodities**

Mycotoxins are controlled by the highest levels that are allowed in food to avoid their harmful effects on people (Claeys *et al*., 2020). Many nations have placed restrictions on the amounts of mycotoxins that can be found in food items (Puel *et al*., 2010). Many international and national organisations, including the FAOs, the World Health Organisation, the EU Commission, and the Codex Alimentarius Commission, additionally established regulations regarding the different mycotoxin types found in various foods in order to protect consumers (Adeyeye, 2016). While most nations do not have particular restrictions on any particular food or food product, basic rules apply to all food items. Nonetheless, a few nations in Europe and the USA have established dietary restrictions on particular items. Among mycotoxins, AFs exhibit strong genotoxic, carcinogenic, and immunosuppressive effects on people. Consequently, government organisations have set maximum amounts of aflatoxins, including AFB1, in the majority of food commodities (Bhat and Reddy, 2017). The highest concentrations (μg/kg) of significant fungal mycotoxins in key food items.

**Food safety and health hazard implications associated with mycotoxins**

The International Agency for Research on Cancer (IARC) designated AFB1 and aflatoxins combinations as Group 1 carcinogens in 1993 (IARC, 2002). The presence of mycotoxins on foods and feeds has the potential to be harmful to both human and animal health due to their diverse toxic effects and high thermal stability (Fellinger, 2006; Barug *et al*., 2003). There is no shortage of data to suggest that food-borne mycotoxins, especially aflatoxins and fumonisins, are heavily present in the diets of people living in sub-Saharan Africa. Miller (1996) asserts that illnesses made worse by aflatoxins account for 40% of the lost production in poor nations owing to illness. Unfortunately, a large portion of the populace in the area is ignorant of the dangers associated with ingesting mouldy goods. Notwithstanding efforts to ensure the safety of food items, customers may still be hesitant to pay more expenses and may even choose to purchase the less expensive goods due to poor literacy rates and other socioeconomic reasons.

**Emerging technologies in foods for reducing mycotoxins**

The food industry's future is in processed foods and novel products that use state-of-the-art technology to accomplish vital goals including higher yields, a longer shelf life, nutrient retention, sensory qualities and product freshness, improved quality, and food safety. These days, research is focused on innovative non-thermal techniques that don't harm consumers, such cold plasma (CP), ultrasonography (US), high-pressure processing (HPP), pulsed electric field (PEF), pulsed light (PL), and ionising and non-ionizing irradiation. Many foods may be employed with these technologies, including products containing biologically active chemicals and heat-sensitive foods, as well as dairy and meat products, canned foods, fresh vegetables and fruits, powdered products, and certain meals. But food may also be protected by these technologies from biological agents like mould, parasites, viruses, bacteria, and yeasts that can contaminate it. This is especially helpful when there are foodborne pathogens present, along with any potentially harmful byproducts like fungus and their mycotoxins.

Governments can protect the interests of food manufacturers and consumers by establishing food rules on the application of these technologies. Developing food processing technologies including radiation, UV, HPP, and PEF have all gone through regulatory clearance procedures run by governments and food safety organisations. These technological advancements safeguard food safety and quality while defending the rights of consumers.Under 21 CFR 179.39 of the Code of the Federal Regulations, for example, the FDA (Food and Drug Administration) has decided that certain food items with a high fat content that are packed in a vacuum or in an inert environment are not allowed to use ultraviolet (UV) rays (1 W of 2537 A. radiation per five to ten ft). A number of other limits apply to potable water as well, including the following: 0.19 per cent or less UV radiation; 100 gal/h per watt of 2537 A radiation flow rate; 1 cm or less water depth; and 36–46º C lamp operating temperature. In a 2000 petition to the FDA, the Food Irradiation Coalition (FIC) requested permission to employ ionising gamma radiation to treat a variety of human foods to a maximal irradiation dosage of 4.5 kGy (for non-frozen and non-dry items) and ten kGy (for frozen or dry products). For the commercial sterilisation of low-acid foods, the FDA approved pressure-assisted thermal sterilisation (PATS) in 2009. PATS is sometimes referred to as combining pressure and temperature in HPP processing.

**Irradiation**

Irradiation is a process that uses ionising energy to extract mycotoxins from food products. Radiation can oxidise fats or vitamins, give food an odd taste, or modify the colour of food, while being a non-thermal and efficient method (Mir *et al*., 2021). Gamma irradiation of a range of cereals (including rice, wheat, barley, and maize) has been used to lower AFB1 (Aziz *et al*., 2004; Aquino *et al*., 2005; Mohamed *et al*., 2015). This has been done. However, doses ranging from 0.5 to 10kGy have also been applied to reduce AFB2 (Aquino *et al*., 2005), ZEA (Aziz *et al*., 2004), OTA (Aziz *et al*., 2004), and FB1 (Aziz *et al*., 2007). The amount of moisture in the samples as well as the gamma irradiation dosage influence how quickly mycotoxins degrade.

According to Mehrez *et al*. (2016), when cereal samples were irradiated with 8 kGy and had a moisture content of 16%, the degradation of mycotoxins was much greater than when the samples had a moisture content of 11%. It might not, however, always eradicate every mycotoxin-producing fungus that is the target. Standing in an environment where things are being exposed to radiation might cause damage and cause cell mutations in individuals (Mir *et al*., 2021).

**Cold plasma**

Low-temperature plasma, sometimes referred to as non-thermal technology or cold plasma (C.P.), is typically created at atmospheric pressures and by electrical discharge in gases or lowered pressures (sub atmospheric pressures), requiring less power. The word refers to partially or fully ionised plasma gases, which are required to include free electrons, photon and ion particles, and atoms in either their excited or original condition. Since the amount of potential negative and positive charges in plasma is equal, it retains neutral as well as pure in charge. The fourth state of a chemical is commonly identified as plasma mode due to its distinct properties. Ozone (O3), electrons, free radical molecules, negative and positive ions, and ultraviolet photons are some of the components that make up cold gas plasma. Granular and fine-grained food, germinated seeds, and food surfaces may all be dried sterilised using it in the food processing industry.

It has been documented that cold plasma has beneficial effects on destroying the structure of mycotoxins and even inactivating or inhibiting the growth-promoting mycotoxins produced by fungus. Various forms of plasma chemicals have the ability to interact with biological molecules and cells, resulting in ongoing modifications at the molecular and morphological levels, ultimately rendering them inactive. Mycotoxin degradation has been attributed to a number of mechanisms, including ionisation, epoxidation, oxidation, direct interaction with free radicals, and deformation during the creation of the terminal furan rings and loss of double bonding in the ring. Based on the data shown in the table, it can be concluded that the cold plasma approach is highly effective in killing or inhibiting the growth of mycotoxigenic fungus, particularly Aspergillus and Penicillium species, in nuts and cereals. Moreover, it can lessen the possibility of harmful mycotoxins—most notably those of the A.F., OTA, DON, ZEN, AOH, and AME type in food.

**High-pressure processing**

High-pressure processing (HPP) or high hydrostatic pressure (HHP) technology is an additional non-thermal method of food preservation and sterilisation that guards against microorganisms and chemical food spoilage agents. In this method, either solid or liquid food is subjected to high pressure (often between 100 and >1000 MPa), which is applied uniformly throughout the food and whose intensity varies according to the volume and length of treatment. The high pressure inactivates microorganisms, spores, chemical, and microbial enzymes and can therefore extend the shelf life, quality, and safety of food products.

Mycotoxins' structures can also be altered by HPP, which will lower their toxicity and lower their concentrations in the environment. For instance, Kalagatur *et al*. discovered that using HPP (550 MPa of pressure) in combination with heating at 45º C and holding period of 20 min resulted in the greatest reduction in ZEA and DON levels (produced by *Fusarium graminearum*) in maize grains. Timmermans *et al*., on the other hand, applied HPP along with moderate heating to inactivate two heat-resistant fungi, *Aliivibrio fischeri* and *Talaromyces macrosporus*, in strawberry puree. They have demonstrated that no particular effects on the viability of the spores of these fungi were seen when heating alone (Tmax = 85º C for 60 min). However, many spores have been rendered inactive by the concurrent application of HPP (500–700 MPa) and heating (85–90º C) for holding durations longer than 13 minutes.

**Ionizing and non-ionizing radiation**

The process of exposing different food kinds to a prescribed quantity of energy which may be broadly classified into two categories: ionising and non-ionizing radiation is known as food irradiation. Ionising radiation, which functions by removing electrons from atoms and molecules of the substance and creating ions, such as gamma (g) and X-rays, as well as a, b, and neutron particles, is a non-thermal technique and kind of energy (high frequency, higher energy, with short wavelength).

Ionising radiation can cause direct or indirect harm to cells by altering DNA molecules and cellular division. Non-ionizing radiation, on the other hand, can destroy cells through heat damage, breaking the structure of DNA, and causing mutations in the microorganism cell. Examples of non-thermal methods include radio frequency (R.F.), ultraviolet (U.V.), visible light or spectrum, infrared (I.R.) and microwave (M.W.) as well as radio frequency (ELF) as low frequency radiation.

The food industry uses this technology, depending on the application, to pasteurise and sterilise food items, improve food quality, and extend food shelf life by disinfecting, stopping the growth and elimination of microorganisms, and breaking down the structure of any potential toxins formed in the food material. Through their interaction with mycotoxins or production of low-biological-activity compounds, highly reactive free radicals and water radicalisation are produced in radiation treatments.

The remarkable effectiveness of ionising and non-ionizing radiation in decontaminating mycotoxin kinds and controlling fungus, particularly *Aspergillus*, *Penicillium*, and *Fusarium* species. The majority of studies have shown that a more notable decrease in the breakdown of mycotoxins and fungi was seen with increased radiation exposure (in a regulated way and within the applicable criteria). According to these results, food contamination in the post-harvest food processing chain may be prevented by using radiation, particularly in agricultural products, dry goods, and powdered foods.

**Pulsed electric field**

There is no denying the role that low temperature, short duration, and low energy consumption play in pulsed electric field (PEF) sterilisation in agricultural and food engineering. The nuclear envelope and cell membrane's positional destruction is linked to the crucial impacts of PEF processing on biological cells. Transmembrane voltage is formed by the potential for differences to exist between the interior and exterior components of biofilms. Once the transmembrane potential or membrane voltage reaches a particular threshold, it induces apoptosis induction, persistent electroporation of the biofilm, and structural alterations in the cells. The transmembrane potential of filamentous fungal cells is determined by both electric and biological factors, including the quantity of hyphae, size, shape, and thickness of the cell membrane, as well as the number and duration of pulses.

Furthermore, by using less pesticides, PEF treatment can be environmentally favourable. For instance, the use of insecticides to eradicate *Rhizoctonia solani* might have detrimental effects on human health due to the residues left behind. The tubular hyphae (filaments) of *R. solani* are elongated, branching structures that hold many nuclei. Recent findings provide scientific evidence in favour of high voltage electric pulses' potential efficacy in eliminating this fungus. The PEF technique may be used on liquid diets; its application to solid food and its ability to inhibit the development of fungus and the creation of harmful metabolites have emerged as current study areas.

In their study, Vijayalakshmi *et al*. focused on examining the effectiveness of PEF technology in reducing the overall number of AFs that degrade, particularly the kind of AFB1 (%) by 20–65% output voltage, 10–26 ls pulse width, and pH conditions between 4 and 10. The results showed that AFB1 and total AFs were eliminated in 91–94% and 83–90% of cases, respectively.

The research states that heat-resistant species respond very well to PET therapy due to variations in pulse numbers, which change the temperature of the outflow. *P. bialowiezense*, a particularly heat-sensitive species, showed mitigation of conidia spores (4.3 logs) at 55º C, but *P. expansum*, a highly heat-resistant species, showed just 0.4 log elimination at 56º C. According to Zhong *et al.*, PEF has the greatest sterilisation efficiency of 99.84% and can kill *F. oxysporum* fungus in a nutritional solution in the least amount of time. The initial microbial density has little bearing on disinfection effectiveness, despite the PEF potency impact and exposure duration. Processing under ideal circumstances was found to have an exposure duration of 10 s and a PEF strength of around 5 kV/cm. Future food disinfection from fungi may be possible with the PEF technique, according to published research.

**Pulsed light**

Using a variety of light treatments, modern food processing techniques may be suitable for initiating chemical and biological events in food models that enable the controlled destruction of certain degradation by-products. The UV radiation is scattered by pulsed light (P.L.) in high-strength minor interval pulses with a flattened wavelength range of 100–1100 nm, which includes short wavelengths with strong bactericidal effects. These wavelengths have a significant potential for penetrating biological cells and affecting their key structural elements, including protein bands, cell membrane walls, and DNA structure, which can result in irreversible cell damage. P.L. has also shown promise in inactivating bacterial endospores, pathogenic parasites, microbial biofilms, and a variety of fungal species in a shorter amount of time.

**Ultrasound**

The foundation of ultrasound technology in the United States is sound waves, which are produced mechanically at extremely high frequencies (about 20 kHz) by molecules vibrating inside a diffusion matrix. Both low intensity (low energy and low power: frequencies 100 kHz and below 1 W/cm2 of intensities) and high intensity (high energy and high power: frequencies in the range of 20–500 kHz and intensities 1 W/cm2) waves, which are inaudible to human ears, are the two categories into which ultrasonication is divided. Through the inactivation of microbes, U.S. technology is utilised as a non-thermal technique to improve product quality, shelf life, and food safety. This includes limiting or inhibiting the growth of fungi and the formation of mycotoxins. The operational mechanism of the United States is linked to the formation of bubbles and acoustic cavitation.

The bubbles fluctuate in response to the propagation of ultrasonic vibrations, resulting in thermomechanical and chemical processes. The bubbles fluctuate and collapse as ultrasonic waves propagate, producing mechanical, chemical, and thermal consequences (collapse pressure, turbulence, and shear stress), as well as the formation of free radicals and extremely high temperatures and pressures in the cavitation zone. In order to cleanse wheat grain, Rudik *et al*. used U.S. (frequency range 2235 kHz and intensity from 0.3 to 1.5 W/cm2). With a frequency of 2426 kHz and an intensity of at least 1 W/cm2, they have been able to lower the fungal content and stop mycotoxins from forming in it. According to Villalobos *et al*., pre-treating fresh figs in the United States for 30 minutes at a frequency of 40 kHz and 60 W, in conjunction with other treatment techniques like osmotic dehydration and K2CO3 emulsion, can effectively regulate the growth of mycotoxigenic fungal and, in turn, the production of mycotoxin.

**Biological methods of detoxification of mycotoxins**

Although biological approaches don't leave harmful residues and are safe for the environment, choosing non-toxic, bio competitive bacteria can be challenging, and the detoxification process can take longer (Mir *et al*., 2021).

**Use of advantageous microorganisms**

Enzymes and a range of microorganisms, including bacteria, moulds, and yeasts, can deactivate or break down mycotoxins, reducing the number of dangerous compounds. A number of microorganisms have already been used to clean food, including *Saccharomyces cerevisiae*, *B. subtilis*, lactic acid bacteria, and *Bacillus licheni* forms. Mycotoxin detoxification procedures can employ microorganisms at any point (pre- or post-harvest), but they need to prove to be economical and successful. Kefir grains have been utilised by Ansari *et al*. (2015) to reduce AFB1 contamination of pistachios by 96.8%, whilst *Bacillus subtilis* UTBSP1 has been used by Farzaneh *et al*. (2012) to reduce AFB1 in the same matrix with an efficiency of 95%. To ascertain the safety and workings of helpful bacteria, more research is necessary.

**Use of botanicals**

The introduction of mycoflora into agriculturally significant food items has been controlled by the use of several synthetic fungicides. Physical and chemical techniques have not yet shown themselves to be effective or adequate in reducing or eliminating mycotoxins from food. Due to their detrimental impact on the food chain, these poisons can be removed by detoxification using botanicals (Gurney *et al*., 2014).

Apart from the debate around artificial preservatives, consumers are becoming more interested in natural food preservation methods to extend the shelf life and enhance food quality while guarding against mycotoxigenic microbial biodegradation (Pandey *et al*., 2016). Because they are safe and environmentally benign, researchers are looking into the antifungal properties of aromatic and medicinal herbs against mycotoxigenic fungus in both the academic and industrial sectors. Bioactive substances having anti-mycotoxigenic properties, such as phenolics, alkaloids, and terpenes, make up natural extracts.

According to Sultana *et al*. (2015), neem leaf extract helped lower the number of AFs that contaminated grains while they were being stored. Nevertheless, the powerful scent of this extract limited its application. *In vitro* detoxification effect of Brazilian medicinal plant extracts was reported by Ponzilacqua *et al*. (2019). These authors found that araca, sweet passion fruits, oregano, and rosemary all caused a time-dependent drop in AFB1, but none of the extracts under investigation had any effect on OTA or ZEA. After 48 hours, the extracts of oregano and araca had the greatest AFB1 drop (about 60%), with the rosemary extract coming in second.

Iram *et al*. (2016) examined the ability of aqueous extracts from Cassia fistula and *Ocimum basilicum* to disinfect pistachios using AFB1 and AFB2. According to these scientists, *O. basilicum* leaf extract was able to destroy 88.6% and 90.4% of AFB1 and AFB2, although *O.* *basilicum* twig and *C. fistula* leaf extracts were less effective. Natural extracts from various plant parts, such as leaves, fruits, and roots, show great promise in the decontamination of food from mycotoxins due to the growing demand for more "natural" additives and agents.

However, due to the effect of variables like the chosen cultivar, the portion of the plant utilised, and the edaphoclimatic circumstances, natural extracts exhibit significant heterogeneity in their composition (Mateus *et al*., 2021). To better manage the efficiency of these extracts, especially as anti-mycotoxigenic drugs, protocols for their standardisation must be established.

**EOs for detoxification of Afs**

Many researchers have suggested use EOs to stop the growth of fungi and stop *A. flavus* and *A. parasiticus* from creating AFs (Maraqa *et al*., 2007; El-Nagerabi *et al*., 2012). Many food-borne fungi are inhibited from producing aflatoxins (AFs) by EOs and flavonoids (Alpsoy, 2010).

At doses of 500 and 1,000 mg/kg, the extract from the seeds of *Azadirachta indica* totally prevented the formation of AFs in maize, whereas at 1000 mg/kg, the extract from the seeds of *Morinda lucida* hindered the development of AFs (Bankole, 1997). EOs produced from Iranian medicinal plants function in food systems as new antioxidants and AFs inhibitors. For instance, it was discovered that *Satureja hortensis* and its active components effectively inhibited the production of AFs by *A. parasiticus*. Carvacrol and thymol IC50 values for AFB1 and AFG1 were 0.50 and 0.06 mM, respectively. The two most powerful components of *S. hortensis* were discovered to be carvacrol and thymol, which might be utilised to lessen the number of AFs in food products (Razzaghi-Abyaneh *et al*., 2007).

Similar findings were made by El-Nagerabi *et al*. (2012), who discovered that by preventing *A. flavus* and *A. parasiticus* from growing, Nigella sativa EO may have reduced the amount of AFB1. Ageratum conyzoides EO was discovered to suppress the synthesis of AFs at 2.0μl/ml when generated by *A. parasiticus* (Ab2242) and 1.5μl/ml when produced by *A. flavus* (La3228; Adjou *et al*., 2012). According to Gomori *et al*. (2013), marjoram and clary sage essential oils considerably inhibit the development of *A. parasiticus*.

**EOs for detoxification of ZEA**

A review of the literature found that not much research has been done on the effectiveness of EOs in lowering ZEA levels in food items. It has been discovered that the essential oils of palmarosa, cinnamon, clove, lemongrass, and oregano are beneficial in lowering ZEA and DON levels. In naturally infected maize grains, *F. graminearum* was discovered to generate ZEA, whereas levels of both toxins were reported to be greater than 500 mg/kg in control crops (Marin *et al*., 2004). Similar to this, different research discovered that clove and palmarosa essential oils (EOs) were superior grain protectors for maize grains, as they both lowered the levels of ZEA and DON production as well as the growth rate of *F. graminearum* under a variety of environmental circumstances (Velluti *et al*., 2004).

**EOs for detoxification of FBs**

EOs have the ability to aid with FB detoxification as well. Researchers discovered that the FB1 production level in maize grains was suppressed by the essential oils (EOs) of cinnamon, lemongrass, palmarose, clove, and oregano (Velluti *et al*., 2003). In their evaluation of the EOs of *Aloysia triphylla*, *A. polystachya*, *Origanum vulgare*, and *Mentha piperita*, Lopez *et al*. (2004) discovered that while the EO of *A. triphyla* was found to increase the mycotoxin levels at lower doses, the EO of *O. vulgare* significantly reduced the FB1 level produced by *F. verticillioides*. In contrast, *O. vulgare* and *A. triphylla* EOs at 250 and 500μl/ml shown effective effectiveness against *F. verticillioides* that produced FB (Lopez *et al*., 2004).

In a further investigation, it was discovered that at 500μg/g dosages, cinnamon oil inhibited the development of *F. graminearum* and *F. culmorum* mycelia and the generation of FB (Hope *et al*., 2005). Furthermore, Yamamoto-Ribeiro *et al*. (2013) reported that the extract from Zingiber officinale inhibited the synthesis of FB1 and FB2 at dosages of 4,000 and 2,000μg/ml, respectively.

**EOs for detoxification of OTAs**

Research on the use of plant EOs to reduce OTAs contamination and associated fungus has sometimes been conducted. According to Basílico & Basílico (1999) and Soliman and Badeaa (2002), OTA production levels by *A. ochraceus* might be inhibited by both oregano and mint essential oils. At 50 ppm dosages, the essential oils of clove, bay, and cinnamon reduced the amount of OTA in the wheat substrate; at 500 ppm doses, the toxin was completely inhibited (Cairns and Magan, 2003). The EO from *P. amboinicus* completely reduced the levels of OTA in meals containing hazardous strain of *A. ochraceus* around 500 mg/kg, as reported by Murthy *et al*. (2009). According to Murthy *et al*. (2009), the administration of 100 mg/kg of these EOs in food samples, such as maize, groundnuts, and chicken feed, also inhibited the growth of *A. ochraceus*. The EO from *A. framomumdanielli* has been shown by Aroyeun *et al*. (2009) to have ochratoxigenic effect in cocoa beans, as demonstrated by its capacity to lower OTA concentrations from 500 to 2,000 mg/kg.

Mohamed *et al.* (2012) discovered that the production of OTA was reduced from 135 to 98μg/ml by 0.10% of basil EOs. Moreover, EOs from *Cymbopogon citratus* (Sonker *et al*., 2014) and *Artemisia nilagirica* (Sonker *et al*., 2014) were observed to completely reduce the levels of OTA at doses of 1.6 and 0.8μl/ml, respectively.

Iradication

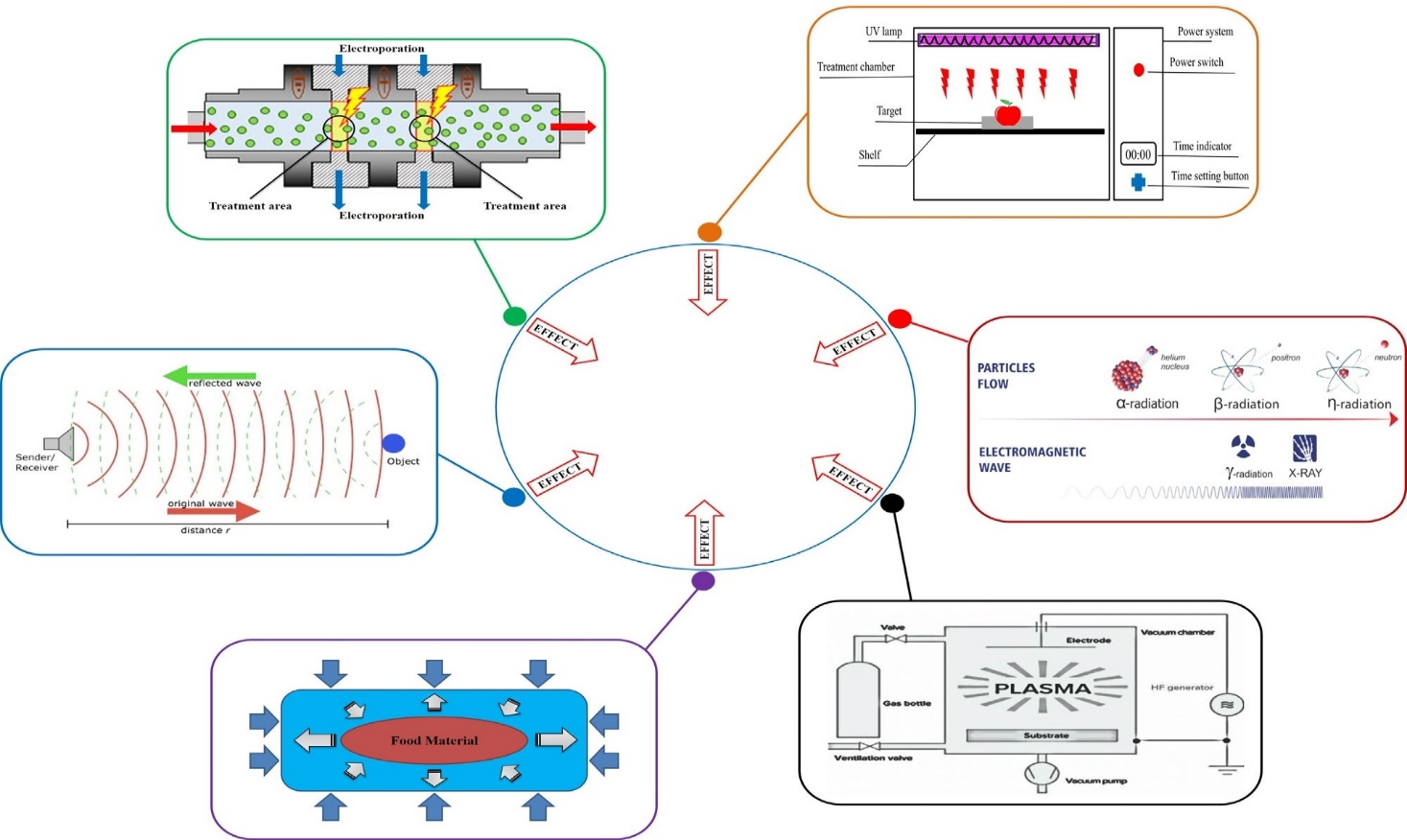
Pulsed Light

Pulsed Electric Field

**High Pressure Processing**

**Cold Plasma**

Fungi and mycotoxins in Food



**Irradiation**

Current Opinion in Food Science

**Fig.4: Controlling of fungi and mycotoxins in food Mirza Alizadeh *et al*.**

**Conclusion**

The development of fungus in food might be a significant sign that there is a considerable risk to consumers from the formation of mycotoxins. A variety of food items, including cereals, fruits, vegetables, and processed meals, can be contaminated with mycotoxins. Due to their extreme stability, mycotoxins are particularly difficult to remove from food after they have contaminated it. Control methods should thus be devised to keep an eye out for and stop mycotoxin contamination in the preharvest and postharvest phases. To determine the presence of mycotoxins in food and feed, several techniques were employed, including HPLC, TLC, HPLC-MS, HPLC-MS/MS, and GC-MS. Maximum levels of mycotoxins in food and feed are regulated by laws in a number of nations across the world.

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