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Application of ozone for degradation of mycotoxins in food: A review

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Abstract

Mycotoxins such as aflatoxins (AFs), ochratoxin A (OTA) fumonisins (FMN), deoxynivalenol (DON), zearalenone (ZEN), and patulin are stable at regular food process practices. Ozone (O₃) is a strong oxidizer and generally considered as a safe antimicrobial agent in food industries. Ozone disrupts fungal cells through oxidizing sulfhydryl and amino acid groups of enzymes or attacks the polyunsaturated fatty acids of the cell wall. *Fusarium* is the most sensitive mycotoxigenic fungi to ozonation followed by *Aspergillus* and *Penicillium*. Studies have shown complete inactivation of *Fusarium* and *Aspergillus* by O₃ gas. Spore germination and toxin production have also been reduced after ozone fumigation. Both naturally and artificially, mycotoxin-contaminated samples have shown significant mycotoxin reduction after ozonation. Although the mechanism of detoxification is not very clear for some mycotoxins, it is believed that ozone reacts with the functional groups in the mycotoxin molecules, changes their molecular structures, and forms products with lower molecular weight, less double bonds, and less toxicity. Although some minor physicochemical changes were observed in some ozone-treated foods, these changes may or may not affect the use of the ozonated product depending on the further application of it. The effectiveness of the ozonation process depends on the exposure time, ozone concentration, temperature, moisture content of the product, and relative humidity. Due to its strong oxidizing property and corrosiveness, there are strict limits for O₃ gas exposure. O₃ gas has limited penetration and decomposes quickly. However, ozone treatment can be used as a safe and green technology for food preservation and control of contaminants.

KEYWORDS

aflatoxins, *Aspergillus*, *Fusarium*, mycotoxins, ozone gas, *Penicillium*

1 | INTRODUCTION

Mycotoxins are a group of toxic fungal metabolites found in a wide range of food and feed products. Some mycotoxins such as aflatoxins (AFs), ochratoxin A (OTA) fumonisins (FMN), deoxynivalenol (DON), zearalenone (ZEN), and patulin have received public attention due to their severe

health effects. *Aspergillus*, *Fusarium*, and *Penicillium* are the main mycotoxin-producer fungal genera (Afsah-Hejri, Jinap, Hajeb, Radu, & Shakibazadeh, 2013). The genus *Aspergillus* is capable of producing both nephrotoxins (such as OTA) and carcinogens (such as AFs) (Tola & Kebede, 2016). Other mycotoxins produced by *Aspergillus* species are FMN, patulin, cyclopiazonic acid, and gliotoxin

(Moretti, & Susca, 2017; Varga, Baranyi, Chandrasekaran, Vágvölgyi, & Kocsubé, 2015). *Penicillium* species produce patulin in addition to AFs and OTA. Some minor mycotoxins produced by *Penicillium* are citrinin, cyclopiazonic acid, and penicillic acid (Perrone & Susca, 2017). *Fusarium* species produce ZEN, FMN, and trichothecenes such as DON (Bhat, Rai, & Karim, 2010; Munkvold, 2017; Torres et al., 2019) as well as T-2, nivalenol, and related derivatives (Moretti, Logrieco, & Susca, 2017). Mycotoxin contamination is a global problem but is more severe in warm and humid environments that favor the growth of fungi and mycotoxin production. Mycotoxin contamination imposes economic burdens on both the agriculture and food industry (Afsah-Hejri, Jinap, & Radu, 2013). Besides the economic losses associated with mycotoxin contamination of crops and food products, other issues such as human and animal health issues, reduced livestock production, and recall and disposal of mycotoxin-contaminated products are serious mycotoxin problems (Milicevic, Nesic, & Jaksic, 2015). Humans and animals can be exposed to mycotoxins through the ingestion of mycotoxin-contaminated food or feed, inhalation, or dermal contact (Gacem, Gacem, Telli, & Khelil, 2020).

Consumption of mycotoxin-contaminated food may cause acute or chronic health effects. Therefore, most of the countries have set strict regulations for the permitted level of some mycotoxins in food (Afsah-Hejri, Jinap, Arzandeh, & Mirhosseini, 2011). The most stringent regulations for mycotoxin in food had been set by the European Union (EU). The EU limits provide the maximum permitted levels for several mycotoxin-food combinations (Moretti & Susca, 2017). Aflatoxin B₁ (AFB₁) is the most harmful aflatoxin with hepatotoxic, mutagenic, and teratogenic effects in humans and animals (Afsah-Hejri, Jinap, Hajeb, et al., 2013). Due to the high toxicity and carcinogenicity, the EU set a tolerance level of 2 µg/kg for AFB₁ and 4 µg/kg for total AFs in cereals and cereal products (European Commission, 2006). OTA is a teratogenic, mutagenic, and nephrotoxic metabolite (Afsah-Hejri & Jinap, 2013; Afsah-Hejri, Jinap, & Mirhosseini, 2012; Pfohl-Leszkowicz & Manderville, 2007) that is classified under class 2B carcinogens (possibly carcinogenic to human) (IARC, 1993). EU limit for OTA in unprocessed cereals and coffee beans is 5 µg/kg (European Commission, 2006). FMN is also under class 2B (IARC, 1993); however, there have been some reports showing the high cancer-inducing property of fumonisin B₁ (FB₁), such as esophageal cancer in South Africa (Marasas, 1997). DON, the water-soluble trichothecene, is considered a potential organic pollutant for the water and environment (Zhou et al., 2020). Although DON inhibits protein synthesis and is known for its immunosuppressive effects, no carcinogenic or mutagenic effect has been reported (Afsah-Hejri,

Jinap, Hajeb et al., 2013; Ueno, 1983). The safe limit for FMN and DON level in grains used for human food in the United States is 2 to 4 and 1 mg/kg, respectively. However, the EU has lower safe limits for FMN (1 mg/kg) and DON (0.75 mg/kg) level in cereal (such as maize) used for human food (European Commission, 2007). Patulin is the most dangerous mycotoxin in fruits (specifically found in injured apples) and is both carcinogenic and genotoxic (Bhat et al., 2010; Diao, Ren, et al., 2018; Aafia, Rouf, Kanojia, & Ayaz, 2018). The United States and most European countries have an acceptable level of 50 µg/L for patulin in fruit juices (Anene, Hosni, Chevalier, Kalfat, & Hbaieb, 2016; Moake, Padilla-Zakour, & Worobo, 2005); however, EU set a low permitted level of patulin (10 µg/L) for fruit juices used in baby foods (European Commission, 2006). ZEN causes reproduction problems (Bhatnagar, Brown, Ehrlich, & Cleveland, 2002) and EU fixed a low level (75 µg/kg) for the maximum permitted level of ZEN in grains used for human consumption (European Commission, 2007).

Most mycotoxins are stable at food process practices. The polycyclic structure of AFs consists of a reactive bifuran group attached to a coumarin nucleus. Aflatoxin B series (B₁ and B₂) differ from G series (G₁ and G₂) by the presence of cyclopentenone ring instead of the β-lactone ring. AFB₁ and AFG₁ possess a double bond at their terminal furan ring (Kumar, Mahato, Kamle, Mohanta, & Kang, 2017; Proctor, Ahmedna, Kumar, & Goktepe, 2004). AFs are moderately soluble in polar solvents such as methanol and poorly soluble in water. AFs are heat stable but unstable at extreme pH values, in the presence of oxidizing agents, and oxygen + UV light (Afsah-Hejri et al., 2011). Depending on the type of food, moisture content (MC) of the food, and the processing method, AFs decompose at a temperature range between 237 and 306 °C (Pankaj, Shi, & Keener, 2018). Removing the double bond in the terminal furan ring of AFB₁ is the main target of most detoxification methods (Luo, Wang, Wang, Wang, & Chen, 2013).

OTA is a phenylalanine derivative, moderately soluble in polar solvents, and relatively unstable to air and light (Afsah-Hejri, Jinap, Hajeb et al., 2013). Pure OTA is stable up to 180 °C (Raters, & Matissek, 2008). Depending on the type of food and its moisture content, OTA degrades at a temperature range between 425 and 490 °C. A significant OTA reduction was observed in dark ground coffee roasted at temperatures higher than 425 °C (Van der Stegen, Essens, & Van der Lijn, 2001). ZEN, the macrocyclic β-resorcylic acid lactone, is slightly soluble in polar solvents and aqueous alkali. Both ZEN and DON are stable during regular food thermal processes. FMNs are thermally stable primary amines and are soluble in polar solvents (Afsah-Hejri, Jinap, Hajeb et al., 2013). FMNs are destroyed at 220 °C (Jard, Liboz, Mathieu, Guyonvarc'h,

FIGURE 1 Mycotoxin control and degradation methods.

& Lebrihi, 2011). Patulin is stable in acidic conditions but unstable at high temperature in alkali conditions (Scott & Somers, 1968).

2 | MYCOTOXIN DEGRADATION OR DETOXIFICATION METHODS

There is no available technique to completely eliminate mycotoxins from food or feed (Freitas-Silva & Venâncio, 2010). However, there are some techniques to prevent mycotoxin contaminations in food and agricultural crops. Hazard analysis critical control point (HACCP) system can be used to prevent and control risks associated with potential mycotoxin contamination. HACCP system is built on foundations of a good quality management system containing the following elements: (a) good agricultural practice, (b) good manufacturing practice, (c) good hygienic practice, and (d) good storage practice (Pineiro, 2001). The HACCP-based system can protect consumers against mycotoxin-contaminated food (Gil, Ruiz, Font, & Manyes, 2016).

The best management approach is to prevent fungal growth and subsequent mycotoxin production in food

(Aldred, Magan, & Olsen, 2004; Gacem et al., 2020). Figure 1 shows the most common mycotoxin control and degradation methods. Common mycotoxin-reducing strategies require the implementation of both pre- and postharvest tactics. The preharvest tactics mainly focus on prevention techniques at the farm level, whereas proper harvesting, processing, drying, and storage methods play a significant role at the harvest and postharvest prevention stages (Cinar, & Onbaşı, 2019; Neme, & Mohammed, 2017; Pankaj, Wan, & Keener, 2018). Biological factors (such as susceptible crops) and environmental factors (such as insect damage, fungal growth, mechanical injury, moisture, and temperature) affect mycotoxin occurrence in agricultural crops (Cinar, & Onbaşı, 2019; Neme, & Mohammed, 2017). Selection of fungal-resistant seeds, elimination of insect-damaged seeds, implementation of proper fungicides and insecticides, control of rodents, and soil management techniques are the most important preharvest mycotoxin prevention methods (Gacem et al., 2020). Proper drying and storage are the basic mycotoxin preventive measures at the postharvest stage. Mycotoxin detoxification can be achieved through physical, biological, or chemical processes (Peng, Marchal, & Van der Poel, 2018).

2.1 | Physical detoxification methods

Thermal process, irradiation, and adsorption techniques are the main physical methods used for mycotoxin detoxification. Extrusion cooking is a physical method that combines high pressure with a high temperature in a short time. Extrusion cooking can reduce the level of AFs and DON in corn flour (Cazzaniga, Basílico, González, Torres, & De Greef, 2001; Elias-Orozco, Castellanos-Nava, Gaytán-Martínez, Figueroa-Cárdenas, & Loarca-Piña, 2002) but thermal methods such as extrusion cooking can only be used for temperature-stable foods and cannot be applied to high-fat and high-protein content foods. Adsorbents such as active carbon (Khan & Zahoor, 2014) or mycotoxin-selective clay (Phillips, Sarr, & Grant, 1995) affect the quality of the food and can only be used for liquids such as milk or oils. Although irradiation can reduce the level of mycotoxins in food (Aygün, 2015; Calado, Fernández-Cruz, Verde, Venâncio, & Abrunhosa, 2018; Di Stefano, Pitonzo, Cicero, & D'Oca, 2014; Jajić, Jakšić, Krstović, & Abramović, 2016; Patras et al., 2017; Sen, Onal-Ulusoy, & Mutlu, 2019; Shanakhat et al., 2019; Vita, Rosa, Giuseppe, & Apparecchiature, 2014), in general it is not a recommended method for detoxification of mycotoxins in food products due to potential molecular reactions (Agriopoulou, Stamatelopoulou, & Varzakas, 2020; He, Zhou, Young, Boland, & Scott, 2010; Shi et al., 2018). Moreover, irradiation of food must be performed under specific standard operating procedures in laboratories approved by Food and Drug Administration (FDA) and International Atomic Energy Agency (IAEA) joint committee (Kalamatur, Kamasani, & Mudili, 2018).

2.2 | Biological detoxification methods

Fermentation and microbial metabolism fall under mycotoxin biological detoxification methods. Mycotoxins can be degraded into less toxic products by microorganisms (Gacem et al., 2020). Some studies focused on biological detoxification of mycotoxins and changing their chemical structures using *Flavobacterium aurantiacum* (Ciegler, Lillehoj, Peterson, & Hall, 1966), *Nocardia corynebacterioides* (Tejada-Castañeda et al., 2008), *Mycobacterium fluoranthenorivans* (Hormisch et al., 2004; Lapalikar et al., 2012), *Lactobacillus rhamnosus* (Abbès et al., 2013), *Saccharomyces cerevisiae* (Repečkienė, Levinskaite, Paškevičius, & Raudonienė, 2013), and *Enterococcus faecium* (Topcu, Bulat, Wishah, & Boyacı, 2010).

Biocontrol methods using saprophytic yeasts (Afsah-Hejri, 2013; Hua, 2013; Hua, Baker, & Flores-Espiritu, 1999) or atoxigenic strains of *Aspergillus* (Atehnkeng, Ojiambo, Cotty, & Bandyopadhyay, 2014; Doster, Cotty, &

Michailides, 2014) inhibit the growth of AF-producing fungi and prevent AF formation. Biological detoxification and biocontrol methods seem better options compared to the physical and chemical detoxification methods, but there are some concerns regarding the uptake of the nutrients in food by the microorganisms as well as the release of microbial metabolites in food (Peng, Chen, et al., 2018).

2.3 | Chemical detoxification methods

Chemical detoxification involves using chemical compounds or ozone for degradation of mycotoxins. Chemical treatments such as ammoniation (Nyandieka, Maina, & Nyamwange, 2009), neutral electrolyzed oxidizing water (Jardon-Xicotencatl, Díaz-Torres, Marroquín-Cardona, Villarreal-Barajas, & Méndez-Albores, 2015), citric acid (Méndez-Albores, Del Río-García, & Moreno-Martínez, 2007), sodium hydrosulfite (Jalili & Jinap, 2012), antioxidants (Gacem et al., 2020), alkaline solutions (Karaca & Nas, 2009; Tabata, Kamimura, Ibe, Hashimoto, & Tamura, 1994), and salts (Jalili, Jinap, & Son, 2011) had been tested for mycotoxin detoxification. Recently, nanomaterials and metallic nanoparticles have been used as antifungal agents to inhibit mycotoxin production (Abd-Elsalam, El-Naggar, Ghannouchi, & Bouquellah, 2020; Wang et al., 2019). Chemical treatments showed to be effective in the reduction of mycotoxin content but cause some irreversible changes and leave residue on the food or convert the structure of mycotoxin into another compound with an unknown structure.

It has been almost two decades since the FDA approved ozone as a safe antimicrobial agent for food industries (Asokapandian, Periasamy, & Swamy, 2018; FDA, 2001; Rice & Graham, 2001). Some advantages of ozone over other chemical oxidants are as follows: (a) ozone precursors are abundant, (b) ozone can be applied both in a gaseous or aqueous form, (c) it does not leave any residue after contact, (d) can be generated on-site, and (e) has no hazardous disposal (Pandiselvam et al., 2019; Torres et al., 2019). The efficiency of an ozone decontamination method depends on the type of treatment, type of food, temperature, pH, and contact time. Ozone treatment has been used on different types of food (Akbas & Ozdemir, 2008; Asokapandian et al., 2018; Concha-Meyer, Eifert, Williams, Marcy, & Welbaum, 2015; Doane & Johnson, 2018; Gonçalves, 2009; Mohammadi Kouchesfahani et al., 2015; Niemira, 2012; Trombete, Freitas-Silva, Saldanha, Venâncio, & Fraga, 2016, 2017; Xu, 1999). Studies showed that ozonization, exposing food to O₃ gas, can reduce the viability of bacterial and fungal contaminants and reduce microbial metabolite accumulation (Freitas-Silva & Venâncio, 2010). Previous studies reviewed some

applications of ozone technology in reduction of fungal contaminants in food (Diao, Hou, Chen, Shan, & Dong, 2013; Freitas-Silva & Venâncio, 2010; Ismail et al., 2018; Karaca, Velioglu, & Nas, 2010; Luo, Liu, & Li, 2018; Peng, Chen, et al., 2018; Peng, Marchal, et al., 2018; Udomkun et al., 2017; Womack, Brown, & Sparks, 2014). Nevertheless, there is no detailed publication available for the application of ozone technology on all six critical mycotoxins in food. This paper provides a comprehensive review on the application of ozone technology on the six critical mycotoxins in food and the mycotoxin-producing fungi as well as the pitfalls and future outlook for ozone technology.

3 | DEGRADATION OF MYCOTOXINS BY OZONE

“Ozein” is a Greek word that means “smell” (Asokapandian et al., 2018). Ozone, originated from the word ozein, is the triatomic form of oxygen and is a natural gas with a high oxidation/reduction potential (-2.07 V) that is higher than of the other common food industry oxidants such as chlorine (-1.36 V), hydrogen peroxide (-1.78 V), and hypochlorous acid (-1.49 V) (Brodowska, Nowak, & Śmigielski, 2018; Pandiselvam et al., 2019; Garud, Negi, & Rastogi, 2019; Khadre, & Yousef, 2001). Ozone has a pungent odor, and its molecular weight is 48 g/mol. Ozone gas has a density of 2.14 kg/m^3 at room temperature (Pandiselvam, Sunoj, Manikantan, Kothakota, & Hebbar, 2017; Rice, Graham, & Lowe, 2002). The stability of O_3 gas in an environment depends on the following factors: temperature, pH, pressure, and presence of organic matters and minerals. Depending on the pH, O_3 gas has a short shelf life between 20 and 30 min at room temperature and then decomposes to form oxygen. The stability of O_3 gas decreases at higher pH values (Freitas-Silva & Venâncio, 2010) and has a shorter shelf life in its aqueous form than its gaseous form (Asokapandian et al., 2018). However, the half-life of ozone in aqueous solution is very high (20 to 40 min) at low temperatures and $\text{pH} < 6.5$ (Pandiselvam et al., 2017). Ozone is considered a safe and environment-friendly sanitizer, which is recognized as an alternative to chlorine in the food industry. Depending on the type of food and its application, O_3 gas, ozonated-water, or ozone-mist can be used in food and agricultural crops (Rice et al., 2002).

O_3 gas is generated when the atmospheric air is exposed to a high-energy source such as corona discharge, ultraviolet (UV), or electrolysis. The concentration of O_3 gas can be measured with a UV meter. The feed rate of an ozone generating system is defined as its O_3 gas flow rate multiplied by O_3 gas concentration. The total applied ozone dosage is calculated by multiplying the O_3 gas concentration with

the exposure time and divided by the volume of the ozone-treated sample (Pandiselvam et al., 2017).

Mycotoxins have structural differences that account for their differences in response to ozone. Ozone treatment destroys the hypertoxic site of the furan ring in AFs (Luo et al., 2014b). Ozone attacks the double bond at C8–C9 of the furan ring of AFB_1 and AFG_1 , resulting in the creation of primary ozonides (Jalili, 2016). Later the degradation products will be rearranged into molozonides derivatives. These intermediate compounds have finite lifetimes (Tiwari et al., 2010). In a protic solvent, these molozonides derivatives will form organic acids or carbonyl compounds, such as ketones and aldehydes. Due to the lack of susceptible double bonds, the initial reaction of O_3 gas with AFB_2 and AFG_2 occurs at other sites of their molecule. The final products of AFB_1 ozone treatment are more polar than the AFB_1 itself (McKenzie et al., 1997). Fumonisin possess nitrogen heterocycles in their molecules (Tiwari et al., 2010). Interaction of O_3 gas with FB_1 initiates the loss of two hydrogen atoms from the parent FB_1 , resulting in the formation of an *N*-oxide at the primary amine of the molecule. McKenzie et al. (1997) used two bioassay systems (hydra bioassay and sphingoid base assay) to test the toxicity of FB_1 after ozonation. Based on the reactivity of the ozonation product with o-phthalic dicarboxaldehyde (OPA reagent) and the toxicity results from the two bioassay systems, they concluded that the final ketone product (3-keto FB_1 derivative) had an intact primary amine meaning that ozonation failed to prevent toxicity of FB_1 in hydra assay. They presented some reasons for their findings such as (a) the hydra assay had poor sensitivity (100 to 250 mg/kg) to FB_1 and (b) FB_1 needed to be more degraded to a compound that is less active than 3-keto FB_1 (e.g., by removing the primary amine in FB_1).

Although the mechanism of detoxification is not very clear for some mycotoxins, it is believed that the oxidizing agents react with the functional groups in the mycotoxin molecules, change their molecular structures, and form products with less molecular weight, less double bonds, and less toxicity (Wang, Luo, et al., 2016). Ozone attacks the double bonds at the C9–C10 in DON molecule and also oxidizes the allylic carbon in the 8 position (He et al., 2010). Biological oxidation is similar to chemical oxidation but more specific. Biological detoxification studies showed the transformation of DON to less toxic compounds, namely, de-epoxy DON and 3-keto-DON (Jard et al., 2011; Li et al., 2011). It is believed that ozone attacks the chlorinated ring structure of OTA, resulting in amino acids or free chlorine (Tiwari et al., 2010). However, the oxidation mechanism of OTA and ZEN is not revealed yet, but it has been reported that phenylalanine and ochratoxin alpha are the main products of acid treatment of OTA (Jalili et al., 2011). Ochratoxin alpha was also detected during the

biotransformation of OTA. Biotransformation of ZEN resulted in less toxic compounds, namely, alpha zearalenol and beta zearalenol (Jard et al., 2011; Varga, Rigó, & Téren, 2000; Varga, Rigó, Téren, & Mesterházy, 2001). Patulin can be completely decomposed by O₃ gas. Diglycolic acid, oxalic acid, and CO₂ are the final products of patulin decomposition by ozone (Cataldo, 2008). Biological oxidation transforms patulin into a less-toxic compound, desoxypatulinic acid (Zhu et al., 2015).

An overview of the antifungal and mycotoxin degradation properties of ozone technology can be a useful tool to establish an appropriate mycotoxin detoxification method and improve the quality and safety of food products.

3.1 | Aflatoxins

AF contamination of crops results in huge losses on trades (Huertas-Pérez et al., 2018). Dwarakanath, Rayner, Mann, and Dollear (1968) were the first who reported the elimination of AFs by ozone. They used O₃ gas to treat high-moisture cottonseed meal (MC = 22%) and peanut meal (MC = 30%) and showed that ozone treatment completely destroyed AFB₁ and AFG₁ but was not effective on AFB₂. In a series of publications, they showed that total AFs level in peanut meal and cottonseed meal was reduced by 78% and 91%, respectively (Dollear et al., 1968; Dwarakanath et al., 1968; Rayner, Dwarakanath, Mann, & Dollear, 1971). Similar results were reported by Maeba, Takamoto, Kamimura, and Miura (1988), who showed immediate destruction of AFB₁ and AFG₁ by O₃ gas. They used thin-layer chromatography (TLC) for AF analysis. AFB₁ and AFG₁ transformed into new products after exposure to 1.1 mg/L O₃ gas and new fluorescent spots appeared on the TLC plates as a result of ozone treatment on AFB₁ and AFG₁. The appearance of these newly fluorescent products showed that the coumarin structure of AFB₁ and AFG₁ remained unchanged after exposure to O₃ gas, meaning that ozone attacked the double bond in their molecules. Prolonged exposure to O₃ gas disappeared the newly induced spots. High concentration of ozone (34.3 mg/L) destroyed AFB₂ and AFG₂ after 60 min exposure but no newly fluorescent product was observed. The difference between the sensitivity of AFB₁ and AFG₁ with AFB₂ and AFG₂ can be explained by their different molecular structure. AFB₁ and AFG₁ have high electron density due to the double bond at C8–C9, which is not present in AFB₂ and AFG₂. This suggested that ozone attacked the coumarin moiety at AFB₂ and AFG₂ and no fluorescent compound was produced. They also showed that ozone-treated AFs did not exhibit any mutagenic activity and suggested that ozone treatment of agricultural crops can be used as an effective method for detoxification of AFs

without producing any toxic compounds (Maeba et al., 1988).

Later, McKenzie et al. (1997) performed a detailed study on the detoxification of seven common mycotoxins using O₃ gas. AFB₁ and AFG₁ in solutions were immediately degraded after being exposed to 2 weight% O₃ gas. AFB₂ and AFG₂ were resistant to oxidation and required higher O₃ gas concentration (20%) and longer exposure time (1 min). They showed a linear relationship between the concentration of O₃ gas and its half-life. A constant source of O₃ gas was required due to the short half-life of ozone. Radiolabeled [¹⁴C] AFs were used to study the final products of ozonation. They suggested an ozone-dependent formation of by-products such as water-soluble compounds; however, no acidic product was detected by high-performance liquid chromatography (HPLC). More than 67% of AFB₁ was degraded in both corn and rice powders after exposure to 20 weight% of O₃ gas for 5 min, whereas AFG₁ was not detectable after the treatment. AFB₂ in both corn and rice powders reduced to 59.8%. They also explained that the difference in degradation rate between AFB₁ and AFG₁ with AFB₂ and AFG₂ was related to the lack of susceptible double bond in AFB₂ and AFG₂, resulting in the initial reaction with ozone at the less reactive sites of their molecules. They proposed that ozone treatment of AFB₁ formed AF molozonide, which was spontaneously transformed into AF ozonide. However, the proposed mechanism does not apply to AFB₂ due to the lack of a double bond at C8–C9. In another study, McKenzie et al. (1998) used ozonation as an environmentally friendly method to treat corn feed and showed the practical degradation of mycotoxins without any toxin byproducts or residues. The ozone-treated corn was used to feed turkey poults, and then their growth performance was monitored. Ozone treatment of AF-contaminated corn resulted in deactivation of AFB₁ and protected the turkey poults from significant weight changes related to AF-contamination of the feed.

Denvir et al. (2000, 2001) described the details of their ozone treatment methods and presented a treatment chamber that could be used for detoxification of a variety of food and agricultural products. O₃ gas was generated using a corona discharge process and uniformly distributed in the chamber. The apparatus had multiple chambers that were all equipped with temperature, pressure, and ozone sensors to continually monitor the parameters. A sampling port was placed at each chamber to extract samples for analysis and avoid overtreatment of samples. They used their system to detoxify AF-contaminated corn and showed that the rate of ozone detoxification was both concentrations and time dependent. A low dose of O₃ gas (67.6 g) for 10 min degraded 99.3% of AFs in whole corn grains. No AF was detected in corn samples after

96 hr exposure to a high dose of O₃ gas (1,025.28 g, flow rate = 178 mg/min at 20 psi).

To evaluate the safety and efficacy of the ozonation process in degrading AFB₁ in corn, Prudente Jr. and King (2002) performed a study on contaminated corn and reported a 92% reduction in AFB₁ after ozone exposure; however, the fatty acid composition of naturally contaminated corn was affected by ozonation (2.9% increase in saturated fatty acids and 2.9% decrease in total unsaturated fatty acids). Ozone-exposed AF-contaminated corn samples did not show any mutagenicity in the Ames assay. They showed that the mutagenicity potential of AFB₁ depended on its purity and the interfering materials in food. They suggested that the interfering materials in corn had antimutagenic properties, and later they used solvents to remove the interfering materials. The extracts from ozone-treated AF-contaminated corn samples showed a less inhibitory effect, and they suggested that it might be due to (a) ozonation process had destroyed the natural mutagen inhibitors in the contaminated samples or (b) ozonation process produced less mutagenic compounds. Further investigation confirmed that the presence of some materials (such as carotenoids, monoterpenes, or indoles) in corn decreased the mutagenic potential of AFB₁. They suggested that these compounds can minimize the formation of AFs or reduce the mutagenic or toxic effects of AFB₁. Prudente Jr and King (2002) also considered linoleic acid as a bioactive compound that contributed to the antimutagenicity property of corn extract. Although ozonation reduced the mutagenic potential of AFB₁, more studies are needed to investigate the ozone-AF reaction products.

The role of insects as vectors for mycotoxin-producing fungi has been discussed in some publications (Cardwell, Kling, Maziya-Dixon, & Bosque-Perez, 2000; El-Desouky, Elbadawy, Hussain, & Hassan, 2018; Hell, Cardwell, Setamou, & Schulthess, 2000; Niculina, Otilia, Veronica, Claudia, & Titus, 2019; Palumbo et al., 2014; Schatzki, & Ong, 2001; Widstrom, 1979; Wright, 1992). Studies showed that field infestation of grains is associated with *Aspergillus* infection and subsequent AF contamination; however, Wright (1992) showed that the main role of insects in *Aspergillus* infection and AF production in grains happens during storage. McDonough et al. (2011) investigated the effect of continuous-flow ozone treatment on AF degradation and fungal and pest reduction in corn. Corn kernels with *Aspergillus flavus* contamination and insect infestation (adult maize weevil [*Sitophilus zeamais* Motsch.] and adult red flour beetle [*Tribolium castaneum* Herbst]) were fed into a screw conveyor system with a continuous flow of ozone (47,800 mg/kg O₃ gas entering the screw conveyor) and a residence time of 1.8 min per pass. They observed 2-log *Aspergillus* count reduction and 100% insect mortality

in corn kernels after three passes through the screw conveyor. McDonough et al. (2011) did not recommend ozone treatment for commercial-scale continuous flow due to the short exposure time (1.8 min) and low AF reduction rate (20% to 30% reduction in AFB₁).

Luo et al. (2014b) showed that a high concentration of ozone (75 mg/L) and long exposure time (1 hr) significantly degraded both AFB₁ and total AFs in corn flour. Ozone treatment also decreased the moisture content of corn flour. They suggested the use of ozone as a practical tool for reducing AF levels as well as moisture content of grains during long-time storage. They performed toxicity tests using human hepatocellular carcinoma cell lines to evaluate the safety of ozone-treated grains. AF extract solutions of both contaminated corn and ozone-treated AFB₁-contaminated corn were used in cell culture solutions, and no noticeable difference was reported between the AFB₁-free culture solution (control) and ozone-treated culture solution (Luo et al., 2014a).

Recently, Porto et al. (2019) applied ozone gas on corn grifts and achieved a 57% reduction in AFB₁ after 480-min exposure to 60 mg/L O₃ gas. They used a factorial design and investigated the effect of the mass of grains, ozone concentration, and exposure time. The highest AF reduction (57%) was reported for the small size of the sample (1 kg), at a high concentration of ozone (60 mg/L), and long exposure time (480 min). Due to the larger surface area and lower MC of corn grifts, their exposure time was longer than that of corn grains (Jr & King, 2002) and corn flour (Luo, Wang, Wang, Li, Wang, et al., 2014b). El-Desouky, Sharoba, El-Desouky, El-Mansy and Naguib (2012) compared the effect of different exposure times (5 to 20 min) and ozone concentrations (20 and 40 mg/kg) at two different initial AFB₁ levels (10 and 20 µg/kg). They showed that despite the level of AFB₁ in wheat, the longer exposure time (20 min) was more effective than higher ozone concentration (40 mg/kg) in AFB₁ reduction. They explained it by the low diffusion rate of O₃ gas through grains and the slow reaction of O₃ gas with the seed coat. They also noticed a significant difference in the efficiency of the ozonation process for high-moisture grains, as ozone reactivity increased in the presence of moisture. Efficacy of ozonation in silos had been investigated by some researchers. Savi, Piacentini, and Scussel (2015) showed simultaneous degradation of AFB₁ and citrinin in wheat after 180-min ozonation and Trombete et al. (2016) reported simultaneous degradation of AFB₁ and DON in soft wheat after 300-min exposure to 60 mg/L O₃ gas. However, Piechowiak, Józefczyk, and Balawejder (2018) recommended ozonation in a fluidized state to control the enzymatic activity of grains. They showed that ozonation of wheat for 30 min (at 30 mg/L O₃ gas) in a fluidized bed decreased the activity of lipase, protease,

and amylase but significantly increased the activity of lipoxygenase.

The efficiency of ozonation is different on the flours than on the kernels or grains. Proctor et al. (2004) investigated the effect of O₃ gas and mild temperature on the degradation of AFs in artificially contaminated peanut kernels and flour. They explained that O₃ gas attacked the double bond at C8–C9 and formed the vinyl ether at the terminal furan ring of AFB₁ and AFG₁. The mechanism involved 1,3-cycloaddition of O₃ at the C8–C9 double bond, forming primary ozonides followed by rearrangement into molozonides. The highest level of AFB₁ degradation (72%) in peanut kernels reached after 10 min of exposure to 4.2% w/w O₃ gas at 75 °C. Regardless of the exposure time, the maximum degradation level of AFG₁ in peanut kernels (80%) was observed at 75 °C (lower AFB₁ and AFG₁ degradation in peanut flour [56% and 61%, respectively]). Similar to Maeba et al. (1988), Proctor et al. (2004) observed a low degradation level (51%) for both AFB₂ and AFG₂ at 75 °C. AFB₂ and AFG₂ showed more resistance to ozone due to the olefin double bond of the terminal ring. Ozone directly attacked AFB₂ and AFG₂ and opened the lactone ring.

Ozonation was more effective in kernels than flour, probably due to (a) the superficial contamination of kernels that can easily be exposed to ozone and (b) the protective effect of flour clumps formed during ozone treatment. Both the exposure time and the temperature had a significant impact on the degradation level of AFs; however, prolonged exposure time (longer than 15 min) had minimal impact. Proctor et al. (2004) suggested that ozonation at room temperature for 15 min is more efficient and cost-effective. They also recommended proper agitation to prevent clumping in flour.

Lower rate of AFB₁ degradation was reported by de Alencar et al. (2012), who investigated the effect of ozone gas on mold count, AF degradation, and physicochemical properties of naturally contaminated peanut kernels. Despite long exposure time (96 hr), only a 25% reduction in AFB₁ and a 30% reduction in total AFs were reported. They compared their results with Proctor et al. (2004) and concluded that their higher degradation rate was related to their artificially spiking method and accumulation of toxin on the surface of kernels that were accessible to O₃ gas. Nevertheless, de Alencar et al. (2012) showed that toxin diffusion was different from the outer part to the inner part of the naturally contaminated kernels. They also noted the influence of relative humidity (RH) on ozone efficiency; the higher the RH, the more efficient the ozonation process. In another study, three different naturally contaminated peanut varieties were exposed to O₃ gas to investigate the efficiency of ozonation on AFs degradation. Although the reduction level was low (18% for AFB₁), Abdel-Wahhab

et al. (2011) recommended the use of ozonation for lowering AFB₁ level in peanuts to meet the Egyptian acceptable level, which is 10 µg/kg for total AFs and 5 µg/kg for AFB₁.

Chen et al. (2014) observed significant degradation of both B and G AFs in ozone-treated peanuts. For the first time, they reported that G-AFs were more sensitive than B-AFs (78% degradation of AFG₁ and 65% degradation of AFB₁ after 30 min exposure to 6 mg/L O₃ gas). They suggested a minimum of 30 min of ozone treatment due to the slow saturation time and low residual concentration of O₃ gas. Moisture content was found to be a critical factor in the effectiveness of the ozonation process as it facilitated ozone adsorption by the kernel surface. Low-moisture peanut kernels (less than 5%) were less sensitive to ozonation. They did not observe any significant changes in the acid and peroxide values of ozone-treated peanuts. Recently, Li et al. (2019) reported that a combination of O₃ gas treatment with UV irradiation resulted in a higher degradation rate than O₃ gas treatment alone. They observed a 79% reduction in AFB₁ after 30 min exposure to 5 mg/L O₃ gas under UV irradiation. They achieved a higher degradation rate than Chen et al. (2014) and suggested to use combination UV/O₃ gas for long-term storage of peanuts. Hassan, Hussein, and Hawar (2018) compared the effectiveness of microwave treatment and ozonation technique for detoxification of AFB₁ in fish feed and reported a higher reduction rate (more than 4.3 times) with ozonation technique.

To evaluate the safety of ozone-treated AFB₁-contaminated peanuts (O-ACPs), Diao, Hou, Chen, et al. (2013) exposed naturally contaminated peanut kernels and peanut paste to 50 mg/L ozone gas for 60 hr and observed significant AFB₁ reduction (89.4%) for both samples. They achieved a higher AFB₁ degradation rate than Proctor et al. (2004) (77% degradation for AFB₁ in peanut kernels) and de Alencar et al. (2012) (25% reduction in AFB₁ in peanut). Rats were then fed with O-ACPs and they reported significant beneficial health effects (such as reduced kidney and liver damages, improved blood biochemical indexes, and decreased risk of cancer) in rats fed with O-ACPs, compared to the rats fed with AFB₁-contaminated peanuts.

In addition to peanuts, the effect of ozone treatment on pistachio kernels and Brazil nuts was also investigated. Akbas and Ozdemir (2006) studied the effect of ozone gas on AFs degradation and the physicochemical properties of pistachio nuts. They reported that regardless of the exposure time, ozone treatment was more effective in pistachio kernels than in ground pistachios, possibly due to the limited penetration of ozone in ground pistachios and weak contact. The highest level of AFB₁ degradation (23%) in pistachio kernels was observed after 420 min of ozone treatment (9 mg/L). Total AFs in pistachio kernels showed

a 24% reduction under the same conditions. Only a 5% reduction was reported for total AFB₁ in ground pistachios. No significant changes were observed in the moisture content, pH, color, and free fatty acid value of ozone-treated kernels and ground pistachios. The peroxide value of all ozone-treated samples was significantly different from the untreated samples except for the ground pistachios treated with 5 mg/L for 140 min, probably because of the limited penetration and short exposure time (less than 420 min). Regardless of the type of the sample, ozone concentration, and exposure time, the composition of five major fatty acids (linoleic, oleic, stearic, palmitic, and palmitoleic) did not show any significant difference after ozonation. Ozone treatment did not affect the appearance, flavor, sweetness, rancidity, and overall palatability of the whole kernels; however, a higher concentration of ozone (>5 mg/L) and longer exposure time (more than 140 min) significantly affected the sensory attributes of ground pistachio. Giordano, Nones, and Scussel (2012) suggested using ozone as an environmentally friendly control method for fungal reduction and AFs degradation in Brazil nuts. Depending on the exposure time and ozone concentration, 75% to 100% of AFs were degraded in naturally contaminated Brazil nuts. Fungal growth was drastically reduced, and no AF was detected during the storage.

Ozone gas was also effective in AF reduction in spices. Inan, Pala, and Doymaz (2007) used O₃ gas for degradation of AFB₁ in flaked and chopped red pepper. They reported an 80% decrease in AFB₁ in flaked red peppers after exposure to 33 mg/L O₃ gas for 1 hr, and a 93% decrease was reported for AFB₁ in chopped red peppers at higher ozone concentration (66 mg/L ozone for 1 hr).

Some researchers compared the efficacy of different forms of ozone (gas, aqueous, or ozonated water) on AF reduction. Zorlugenç, Kiroğlu Zorlugenç, Öztekin, and Evliya (2008) showed that O₃ gas was more effective than ozonated water in the reduction of AF level in contaminated dried Sarilop figs. Ozonated water (1.7 mg/L) had no significant effect on AFB₁ level at 30 min. However, ozonated water was effective at higher exposure times (more than 1 hr), and 83.25% and 88.62% reduction in AFB₁ level observed at 60 and 180 min, respectively. The highest reduction in AFB₁ level (95.21%) was reported for samples exposed to 13.8 mg/L ozone gas for 180 min. Their results were similar to the findings of McKenzie et al. (1998) for contaminated corn, but in contrast with the results reported by Wang, Liu, Lin, and Cao (2010), who for the first time used ozone mist for degradation of AFs in corn and compared it with ozone gas and ozonated water. Only a 52.4% reduction in AFB₁ level was observed for O₃-gas-treated corn, whereas ozonated water and ozone mist (called wet method) resulted in 78.1% and 85% reduction in AFB₁ level, respectively. Ozonated water was more effective

in the reduction of AFB₁ (92.2%) in wheat samples, followed by ozone mist (85.5%) and O₃ gas (56.8%). Ozone mist significantly reduced (94.4%) AFB₁ level in paddy rice after 12 hr. For paddy rice samples, AFB₁ degradation ratio for ozonated water and O₃ gas was 87.4% and 70.8%, respectively. Wang et al. (2010) concluded that the superiority of ozone mist and ozonated water to O₃ gas could be related to the reaction of ozone with water resulting in free OH radicals with stronger oxidation ability than ozone itself. Regardless of the type of grains, the final concentration of AFB₁ in 12-hr ozone-mist-treated samples was reduced to less than the maximum permitted level of AFB₁ in each grain. Germination capability of seeds was reduced by ozone treatments, but no significant changes reported for fatty acid content, odor, and color of the ozone-treated grains except for the color of ozone-mist-treated samples. Ozonation can be used for the degradation of AFB₁ in grains; however, further investigation is needed to study the effect of ozone mist and ozonated water on low moisture products.

Puzyr', Burov, Bondar', and Trusov (2010) studied the effect of ozonated water on the degradation of AFB₁ and combined it with an adsorption method using modified nanodiamonds (MND). They reported that 96.6% of AFB₁ was degraded after 5-min exposure to 19.5 mg/L ozonated water. A combination of ozonated water + MND resulted in the complete degradation of AFB₁ in 10 min. They showed that ozonated water significantly degraded AFB₁ and nanodiamonds absorbed the residual toxin.

One of the most important concerns in the application of ozone for mycotoxin decontamination is the final ozone degradation product, which should be identified and tested for its toxicity. Luo et al. (2014c) investigated and analyzed the structure of the degradation products of AFB₁ by aqueous ozone using a high-sensitivity, high-resolution, ultra-performance liquid chromatography. They identified six ozone degradation products (C₁₇H₁₀O₇, C₁₇H₂₂O₉, C₁₆H₁₂O₇, C₁₆H₁₆O₆, C₁₇H₁₄O₈, and C₁₆H₁₄O₇). In addition to measuring the mass of the products, double bond equivalents (DBEs) for AFB₁ degradation products were calculated. For AFB₁, they reported the DBE = 11.5 and identified six earlier mentioned degradation products with DBEs ranging from 6.5 to 10.5, all less than that of AFB₁. Lower DBEs (less than 11.5) for the degradation products confirmed that the double bonds in the terminal furan ring had been attacked by ozonated water. Ozone-degraded products showed no adverse effect on toxicity tests. Ozone treatment rapidly degraded more than 90% of AFB₁ and AFG₁ in methanol solution, whereas AFB₂ and AFG₂ were resistant to ozonation and showed 49.7% and 72% degradation after 16 hr ozone treatment (Ayranci & Karaca, 2018). Four ozonation products (C₁₇H₁₄O₇, C₁₆H₁₆O₆, C₁₅H₁₄O₅, and C₁₆H₁₄O₅)

were identified after the methanol solution of AFB₁ was exposed to O₃ gas (Luo et al., 2014c). AFB₁-ozone-degraded products did not show any adverse effect on the toxicity tests; however, more research is needed to study the AFB₁ ozonation products in highly contaminated samples and assure that the ozonated products are safe for animal and human consumption (Luo et al., 2018).

Diao et al. (2012) studied the ozonolysis pathways of AFB₁ in acetonitrile solution and detected 13 products, and six of them identified as the main ozonolysis products. Their DBE values were similar to those reported by Luo et al. (2013): DBE = 12 for AFB₁ and DBE = 11 to 13 for ozonolysis products. Nine ozonolysis products were produced through the first oxidative pathway of AFs, known as the Criegee mechanism. Criegee mechanism produces primary ozonides or Criegee intermediate products that will later decompose to carbonyl compounds. During the ozonolysis of AFB₁, a 1,3-dipolar cycloaddition of O₃ occurs at the C8–C9 double bond, producing an unstable molozone that spontaneously decomposes to a carbonyl compound of AFB₁. The second pathway of AFB₁ is based on the electrophilic and oxidative reactions and the benzene ring methoxy group is then oxidized. The main ozonolysis products of AFB₁ were C₁₇H₁₄O₁₀, C₁₈H₁₆O₁₀, C₁₆H₁₀O₆, C₁₉H₁₅NO₉, C₁₇H₁₂O₉, and C₁₇H₁₂O₉ (with different retention time and mass) (Diao et al., 2012). However, ozonolysis products did not show any toxic effects on animals (Diao, Hou, & Dong, 2013).

Recently, Agriopoulou, Koliadima, Karaiskakis, and Kapolos (2016) studied the kinetic and behavior of AF solution in water under different ozonation conditions. Similar to Chen et al. (2014), they found AFG₁ to be more sensitive to O₃ gas than AFB₁. They reported the sensitivity of AFs to ozonation as AFG₁ > AFB₁ > AFG₂ > AFB₂. AFB₂ was quite stable and showed only 17% to 29.6% degradation after 20-min exposure to different concentrations of O₃ gas. Regardless of the temperature and concentration of AFs, complete degradation of AFB₁ and AFG₁ observed after 3-min exposure to 13.5 mg/L O₃ gas. The higher degradation rate in AFB₁ and AFG₁ is related to their chemical structure and presence of the double bond at C8–C9 of their furan ring, which is not present in AFB₂ and AFG₂. The reaction of ozone with AFs was considered as a first-order reaction, and the true rate constants for decompositions of AFB₁ and AFG₁ by ozone at all temperatures were greater than those for AFB₂ and AFG₂. They also showed that the activation energies were concentration dependent and for AFB₁ and AFG₁ increased at higher concentrations of ozone. They concluded that AFB₁ was more sensitive at lower ozone concentrations (8.5 mg/L O₃ gas), whereas AFG₁ was more sensitive at higher concentrations of ozone (20 mg/L O₃ gas).

3.2 | Other mycotoxins

3.2.1 | Deoxynivalenol

Several studies proposed ozonation to be an effective, fast, and safe method for degradation of DON in agricultural commodities (Alexandre et al., 2018; Alexandre, Castanha, Calori-Domingues, & Augusto, 2017, 2018, 2019; Deng, Chen, Guo, & Zhang, 2011; Diao, Wang, Li, Wang, & Gao, 2018; 2019; Li, Guan, & Bian, 2015; Savi, Bittencourt et al., 2014; Sun et al., 2016; Torlak, 2019; Wang, Shao, et al., 2016; Wang, Luo, et al., 2016; Young, 1986; Young, Subryan, Potts, McLaren, & Gobran, 1986; 2006) (Table 1). In a series of studies, Young and colleagues showed that ozone degraded DON in corn samples, but not in wheat grains (Young, 1986; Young et al., 1986). Moist ozone (1.1 mol %) was more effective (90% reduction) than dry ozone (70% reduction) in DON-contaminated corn (Young, 1986). Dry ozone did not reduce the level of DON in naturally contaminated whole wheat kernels (Young et al., 1986). They exposed artificially contaminated oven-dried ground unhusked corn to both dry and moist ozone and observed different half-life of DON disappearance (2.5 hr for dry ozone and 15 min for moist ozone). The study showed that moisture plays an important role in the degradation of DON by ozone. Wheat grains were exposed to ozone for 3 hr and then left in an ozone-saturated chamber for 24 hr. Moist ozone was not effective on wheat samples, possibly due to the large sample size, or the matrix effect (Young, 1986). They assumed that ozone could not penetrate the whole wheat kernels as quickly as corn grains. In another study, they investigated the effect of pH on degradation of DON and the reaction between DON and ozone in an aqueous medium (Young, Zhu, & Zhou, 2006). They showed that ozone-saturated water (25 mg/kg) completely degraded DON to a nondetectable level. They observed some intermediate products at low ozone concentrations (0.25 mg/kg). Based on the UV and mass spectroscopy data, they assumed that ozone attacked the C9–10 double bond in DON molecule. They also showed that the reaction rate depended on the oxidation state at the allylic C8 position. The relative amount of ozone for 50% reduction of DON was higher in keto state than hydroxyl and methylene states. DON degradation by ozone was pH sensitive, and rapid degradation was observed at pH 4 to 6. DON reactivity with ozone depended on the oxidation state of C8 at pH 7 to 8, and very little or no reaction was reported at pH = 9.

Savi, Piacentini et al. (2014) showed that ozone inhibited fungal growth and degraded DON in wheat grains. DON was degraded in the pericarp and endosperm of wheat grains and no physicochemical changes observed after 120-min exposure to O₃ gas (60 µmol/mol). Li et al. (2015)

investigated the effect of ozone treatment on the quality parameters of wheat grains. Thirty seconds of exposure to 10 mg/L O₃ gas reduced 93.6% of DON (initial concentration = 1 µg/mL) in the solution. Higher degradation rates were reported for solutions with lower initial concentrations of DON (initial concentration <0.3 µg/mL). They showed that DON degradation was both concentrations and time dependent. They reported a 57.3% DON reduction in scabbed wheat (17% moisture content) after 12-hr exposure to 60 mg/L O₃ gas. Moisture content of wheat grains had a significant influence on DON degradation. No changes in the starch pasting properties of wheat observed after 4-hr ozone exposure. However, ozone treatment improved the quality of flour, and a slight rise in the stability and dough development time was observed. Wang, Shao, et al. (2016) reported that ozone treatment simultaneously degraded DON and improved flour quality. They reported 39.16% and 53.48% reduction in DON level after the 60- and 90-min exposure to 75 mg/L O₃ gas, respectively. Protein, starch, and amino acid content, fatty acid value, and carbonyl and carboxyl contents of ozone-treated samples remained unchanged. All ozone-treated samples had lower extensibility and yellowness as well as higher tenacity and whiteness. In another study, Wang, Luo, et al. (2016) evaluated the effectiveness of treatment parameters (such as exposure time, ozone concentration, moisture content, and type of raw material) on the degradation of DON. Similar to the observation of Li et al. (2015), Wang, Luo, et al. (2016) reported that DON degradation was time and concentration dependent and was significantly higher at longer exposure time and higher ozone concentrations. DON degradation was higher in whole wheat flour than wheat kernels. The higher degradation rate (78.66%) was observed in high-moisture samples (MC = 20.1%), and the maximum reduction of DON was reported for 20% MC samples. One-hour ozone treatment (100 mg/L) reduced DON in whole wheat flour from an initial concentration of 3.89 mg/kg to a final concentration of 0.83 mg/kg.

Sun et al. (2016) used saturated aqueous ozone for the degradation of DON in different contaminated grains. They reported 83% degradation of DON in solution after 7-min ozone exposure (80 mg/L). Lower detoxification rates were reported for contaminated wheat, corn, and bran (74.86%, 70.65%, and 76.21%, respectively) after 10-min exposure to 80 mg/L of ozone. Alexandre et al. (2017) evaluated the impact of ozonation on the rheological properties of flour. Although 80% reduction in DON was observed in high moisture samples, the process affected the rheological profile of whole wheat flour. They also studied the effect of ozonation on the nutritional quality of the wheat bran. DON reduction was only 32% after 240-min ozonation; however, the antioxidant capacity and total phenolic con-

tent of the bran were not affected (Alexandre et al., 2018). Recently, Piemontese et al. (2018) identified the optimum ozonation conditions that had no effect on the rheological characteristics of wheat and semolina. They showed that 6-hr ozone exposure (55 g/hr) degraded 29% of DON in wheat, significantly reduced fungal growth, and did not affect the rheological properties of semolina. Trombete et al. (2017) evaluated the effects of ozone concentration, exposure time, and grain mass on the levels of DON in wheat grains. A maximum of 48.0% DON reduction was observed when 2 kg of grain sample was treated with 60 mg/L of ozone for 300 min. They reported that ozone concentration and exposure time had positive effects on DON reduction, whereas the grain mass had a negative effect.

Researchers also studied the toxicity of degradation products of DON after ozone treatment (Li, Guan, & Bian, 2019; Ren et al., 2019; Wang et al., 2017; Xu, Ji, et al., 2019). Cytotoxicity tests performed by Xu, Ji, et al. (2019) revealed that ozone-treated and degraded DON products had cellular toxicity effects. Li et al. (2019) showed that treatment with gaseous ozone resulted in a 95.68% degradation in DON in ultrapure water within 15 s. The toxicity of 10 identified ozonized products of DON was significantly decreased because of de-epoxidation and the attack of ozone at the C9–10 double bond in DON. Cytotoxicity tests showed that the toxicity of DON in pure solution was significantly reduced after ozone treatment (Ren et al., 2019). Wang et al. (2017) investigated the safety of DON-contaminated wheat (DCWs) after ozone exposure and then test animals were fed with DCWs during the sub-chronic toxicity experiments. They showed that the toxic effects of DON were reduced by ozone and ozone itself had minimal harmless effects on mice in this process. The recent study by Ren et al. (2019) reported a complete degradation of DON in aqueous solution after 20-min exposure to 14.50 mg/L of O₃ gas with a flow rate of 80 mL/min.

3.2.2 | Ochratoxin A

McKenzie et al. (1997) studied the degradation of OTA at high concentrations of ozone. OTA totally degraded in solutions after 15-s exposure to O₃ gas (10% by weight) and the HPLC test detected no by-products. They measured the toxicity of OTA by a mycotoxin-sensitive bioassay method and showed that the toxicity of ozone-treated OTA solution was significantly reduced after 15 s of exposure to O₃ gas. According to Deng et al. (2011), OTA in corn (initial concentration = 80 µg/kg) completely degraded after 120-min ozone treatment at 30 g/m³ or 90 min at 60 g/m³. The study claimed that ozone treatment had a very little effect on the fatty acid composition in corn, which suggested

ozonation as a good treatment for degradation of OTA during corn storage. Qi et al. (2016) showed OTA degradation in solutions (65.4% after 120 s exposure to O₃ gas) and corn samples (70.7% after 180 min exposure to 100 mg/L O₃ gas). OTA degradation in corn was both concentrations and time dependent. High-moisture corn samples (19.6%) were more sensitive to ozone than low-moisture (14.1%) samples. They reported a decrease in the final MC of the ozone-treated corn samples. They also showed that 180-min ozone treatment increased both the whiteness and fatty acid value of treated corn samples.

Torlak (2019) investigated the effectiveness of O₃ gas on degradation of OTA in sultanas (raisins). The study showed that the initial level of OTA (16.7 µg/kg) in sultanas was reduced by 60.2% and 82.5% after 120- and 240-min ozone exposure, respectively. The study also claimed a 2.2-log reduction in the fungal population of sultanas after 120-min exposure to O₃ gas. No significant change in the concentration of phenolic substances of sultanas was reported.

3.2.3 | Patulin

An ideal degradation method should completely degrade the toxin up to water and carbon dioxide, which might not always be possible. Simple structure mycotoxins (such as patulin) tend to be more degraded than complex ones (Karaca et al., 2010). O₃ gas decomposes patulin to acids and CO₂ (Cataldo, 2008). McKenzie et al. (1997) showed significant degradation of patulin and ZEN in cornmeal and rice powder. They reported a complete degradation of patulin and ZEN in aqueous solutions exposed to 10% weight O₃ gas for 15 s. No degradation product was observed under UV light. Karaca and Sedat Velioglu (2009) studied the efficiency of ozone treatment on the degradation of patulin in the presence of some metal ions in model systems. Patulin showed very little resistance to ozone in model systems and 98% of initial patulin concentration was degraded in less than 1 min. Degradation rates of patulin in the presence of zinc, copper, calcium, and aluminum were almost the same as the one in the absence of these metals. Conversely, the presence of manganese and iron significantly reduced the detoxification rate of patulin by ozone. Ozone degradation of patulin did not produce any new byproducts. Diao, Wang, et al. (2018) used a self-developed ozone generator for the elimination of patulin in apple juice. They achieved 64.77% and 81.66% patulin reduction after 10 min exposure to 7 and 12 mg/L O₃ gas, respectively. The study showed that ozone concentration, treatment time, and pH had positive effects on the degradation efficacy of patulin, whereas initial patulin concentration and soluble solids content in apple juice had nega-

tive effects. Ozone detoxification reduced the color, ascorbic acid, malic acid, and total phenolic content of apple juice, but did not show any significant effects on its pH, soluble solids, and total acid. In another study, Diao, Ren, et al. (2018) exposed patulin to 10.6 mg/L O₃ gas for 90 s and tested the cytotoxicity of ozone-treated products on human hepatic carcinoma cells (HepG2) using MTT assay and apoptosis assay. They observed a 51.65% increase in cell viability due to ozone detoxification. They also reported an 11.06% reduction in total apoptotic cells after 180-s ozonation, suggesting that O₃ gas can significantly degrade patulin in drinks. According to this study, the half-maximal inhibitory concentration (IC₅₀) of patulin on HepG2 cells was 9.32 µmol/L after 24 hr of ozone treatment, which also showed a dose-dependent effect. They also investigated the efficiency of ozone processing on the degradation of patulin in apple juice (Diao et al., 2019). Using a self-developed ozonolysis reactor, they reported a 75.36% patulin degradation in apple juice. However, they reported some adverse effects of ozone processing on the quality of apple juice (such as changes in the major phenolic compounds and organic acids).

3.2.4 | Zearalenone

Qi et al. (2016) showed that 5 s of ozone exposure (10 mg/L O₃ gas) effectively degraded ZEN in corn samples to an undetectable level. Degradation of ZEN in corn samples increased at long exposure time and high ozone concentration. High moisture corn (19.6%) was more sensitive to ozonation than low-moisture corn (14.1%). They observed a 90.7% reduction in ZEN when high-moisture (19.6%) corn samples were exposed to 100 mg/L O₃ gas for 180 min. Ozone-treated corn samples had lower moisture content; however, their whiteness and fatty acid value increased. Alexandre et al. (2018) reported that 61% of initial ZEN concentration was destroyed after 240-min ozonation. They evaluated the nutritional quality of the wheat bran and showed that ozonation did not affect the total phenolic content and the antioxidant capacity of the bran. Xu, Wang et al. (2019) used aqueous ozone to reduce the level of ZEN in corn flour. They showed that 95.1% of the initial concentration of ZEN in contaminated corn flour was degraded after 90-min ozonation. They identified four degradation products, two of them were also detected in real samples of contaminated corn flour. They also evaluated the toxicity of the parent ZEN and its ozone-induced decomposition products. Ozonation products showed less toxic effects than ZEN, meaning that ozonation can be used to reduce the toxic effects of ZEN-contaminated products. According to a recent study by Alexandre et al. (2019), ozonation was effective in reducing ZEN contamination in whole maize

flour (WMF), with a maximum reduction of 62.3%. They also showed that ozone treatment modified the pasting properties, fatty acid profile, peroxide value, and affected functional and technological aspects of WMF.

3.2.5 | Fumonisin

Fumonisin are relatively stable molecules (Riley & Norred, 1999) and there is a very limited number of studies on fumonisin decontamination. It was very difficult to find research on fumonisin degradation by ozone; however, Mylona, Kogkaki, Sulyok, and Magan (2014) were the only research group who studied the effect of O₃ gas on *Fusarium* growth and fumonisin degradation. They showed that 30-min exposure to 200 mg/L O₃ gas inhibited growth of *Fusarium verticillioides* and subsequently reduced FMN production to a nondetectable level. Future research is needed to investigate the degradation of FMN in different food products and determine the safety of FMN ozonation products.

4 | EFFECT OF OZONE ON FUNGAL MICROBIOTA

Damage to the fungal membrane is known as the mechanism of the antifungal property of O₃ gas (Brodowska et al., 2018). Depending on the membrane structure, some fungal species show more resistance to ozone treatments. Gaseous O₃ is more effective in mycotoxin reduction, whereas aqueous ozone is known for its fungal growth control ability (Öztekin, Zorlugenç, & Zorlugenç, 2006; Palou, Smilanick, Crisosto, & Mansour, 2001). Therefore, it is suggested to use ozone as an alternative to fumigation for food and agricultural products (Whangchai, Saengnil, & Uthaibutra, 2006).

Hibben and Stotzky (1969) were the first who investigated the effect of O₃ gas on the germination of 14 fungal spores (Table 1). Actively metabolizing spores were very sensitive to O₃ gas. They showed that dry spores were less sensitive to O₃ gas, and the inhibitory effect of ozone increased at high RH conditions (95% to 99%). Large and pigmented spores (such as *Alternaria* spp.) were more resistant to O₃ gas, whereas the small and hyaline spores (such as *Fusarium* spp.) were very sensitive to O₃ gas. Medium sensitivity was reported for *Aspergillus* and *Rhizopus* species. The thickness of the cell wall and the presence of pigments in large spores could be responsible for their higher resistance. According to Hibben and Stotzky (1969), ozone could have affected the integrity and permeability of the membrane, deactivated enzymes, or oxidized the lipid fractions of the cell wall essential for

the synthesis of long-chain fatty acids. They also showed that RH was a key factor in the effectiveness of the ozonation process. Dry and nongerminating spores were more resistant to O₃ gas than the moist and germinating spores.

Ozone fumigation of grains is usually applied in silos. Ozone movement through the grains depends on the type of grain and must be optimized prior to ozonation. During ozone fumigation, O₃ gas primarily reacts with the seed coat and then diffuses into the grain (Tiwari et al., 2010). The penetration of gas into the grain depends on several factors such as moisture content, surface characterization of the grain, microbial contamination, and presence of insects.

Ozone can be used for insect management in stored grains (Hansen, Hansen, & Jensen, 2013; Isikber & Athanassiou, 2015; Isikber, & Öztekin, 2009; Pereira, Faroni, Sousa, Urruchi, & Paes, 2008; Tiwari et al., 2010) and as a potential alternative for phosphine against phosphine-resistant insects (Sousa, Faroni, Guedes, Tótola, & Urruchi, 2008). Ozonation can reduce both insect and fungal contamination in fruits (Al-Ahmadi, Ibrahim, & Ouf, 2016) and grains (Kells, Mason, Maier, & Woloshuk, 2001; Wu, Doan, & Cuenca, 2006). Jian, Jayas, and White (2013) reviewed the application of different concentrations of ozone against the most popular grain insects (*Sitophilus zeamais*, *Tribolium castaneum*, *Plodia interpunctella*, *Sitophilus oryzae*, *Tribolium confusum*, *Oryzaephilus surinamensis*, and *Rhyzopertha dominica*). They recommended 3 days of ozonation at 50 mg/L for the reduction of storage fungi and 8 days at 135 mg/L O₃ gas to eradicate insect infestation.

Kells et al. (2001) evaluated the efficacy of O₃ gas fumigation on fungi and insect reduction in stored maize. Ozone gas (25 and 50 mg/kg) was used to fumigate maize kernels contaminated with *A. parasiticus* and insects (adult maize weevil—*Sitophilus zeamais* [Motsch.], adult red flour beetle—*Tribolium castaneum* [Herbst], and larval Indian meal moth—*Plodia interpunctella* [Hübner]) for a period of 3 to 5 days. Three days of ozone fumigation (50 mg/kg O₃ gas) inhibited fungal growth and insect infestation by 63% and 92 to 100%, respectively. They optimized the fumigation parameters for the typical corn storage systems and their suggestion for fumigation of maize was to use 50 mg/kg O₃ gas at a velocity of 0.03 m/s for 1 day and 0.02 m/s for five subsequent days. According to them, more than 85% of O₃ gas penetrated 2.7 m into the grains at this optimum condition. They also suggested using ozone fumigation as an environmentally friendly alternative to phosphine fumigation. Despite the high incidence of mycotoxins in forage, ozonation is not recommended for ensiled feed due to their large volume (Ogunade et al., 2018).

TABLE 1 Application of ozone for degradation of six major mycotoxins (AFs, DON, OTA, patulin, FMN, and ZEN) and reduction of fungal microbiota

Product		Target of ozone	References
Corn	AFs		Denvir et al. (2000, 2001); Prudente Jr. and King (2002); Luo, Wang, Wang, Li, Bian, et al. (2014a); Luo, Wang, Wang, Li, Wang, et al. (2014b); McKenzie et al. (1998)
	AFs	<i>Aspergillus</i> <i>Fusarium</i>	Porto et al. (2019)
		<i>Aspergillus</i> <i>Fusarium</i> <i>Penicillium</i> <i>Rhizopus</i> <i>Mucor</i>	White (2007); White, Murphy, Bern, and van Leeuwen (2010, 2013)
		<i>Aspergillus flavus</i>	Hussein et al. (2015)
	AFs	<i>Aspergillus flavus</i>	McDonough et al. (2011)
		<i>Aspergillus parasiticus</i>	Kells et al. (2001)
	OTA and ZEN		Qi et al. (2016)
	FMN	<i>Fusarium verticillioides</i>	Mylona et al. (2014); Frisón et al. (2014)
	ZEN		Xu et al. (2019)
	OTA		Deng et al. (2011)
Corn flour	ZEN		Alexandre et al. (2019)
Corn powder and rice	AFs, OTA, ZEN, and Patulin		McKenzie et al. (1997)
Corn and wheat	DON		Young (1986); Young et al. (1986); Savi et al. (2014); Li et al. (2015)
Corn, wheat, and rice	AFs		Wang et al. (2010)
Corn, wheat, and bran	DON		Sun et al. (2016)
Corn, wheat, and oat		<i>Fusarium graminearum</i> <i>Fusarium verticillioides</i> <i>Fusarium langsethiae</i>	Mylona (2012)
Wheat	AFs		Savi et al. (2015)
		Fungal spores (not specified)	Wu et al. (2006)
	AFs	<i>Aspergillus</i> <i>Fusarium</i>	Trombete et al. (2016)
	AFs	<i>Aspergillus flavus</i>	El-Desouky et al. (2012)
		<i>Aspergillus clavatus</i> <i>Aspergillus niger</i> <i>Alternaria alternata</i> <i>Cladosporium cladosporioides</i> <i>Fusarium avenaceum</i> <i>Fusarium graminearum</i> <i>Fusarium poae</i> <i>Fusarium solani</i> <i>Fusarium tricinctum</i> <i>Fusarium sporotrichioides</i> <i>Penicillium aurantiogriseum</i> <i>Penicillium aurantiocandidum</i> <i>Penicillium funiculosum</i> <i>Penicillium verrucosum</i> , <i>Penicillium variable</i> <i>Penicillium expansum</i> <i>Rhizopus oryzae</i>	Raila et al. (2006)

(continues)

TABLE 1 Continued

Product		Target of ozone	References
		<i>Aspergillus flavus</i> <i>Penicillium citrinum</i>	Savi et al. (2015)
		<i>Aspergillus flavus</i> <i>Aspergillus parasiticus</i> <i>Fusarium graminearum</i> <i>Fusarium verticillioides</i> <i>Penicillium citrinum</i>	Savi and Scussel (2014)
	DON		Wang, Shao, et al. (2016); Wang, Luo, et al. (2016)
Wheat seeds		<i>Alternaria</i> <i>Aspergillus</i> <i>Fusarium</i> <i>Penicillium</i>	Granella et al. (2018)
Soft wheat	DON and AFs	<i>Aspergillus</i> <i>Fusarium</i>	Trombete et al. (2017)
Whole wheat flour	DON		Alexandre et al. (2017)
Wheat bran	DON and ZEN		Alexandre et al. (2018)
Wheat semolina	DON		Piemontese et al. (2018)
Wheat, pea, and barley		<i>Aspergillus</i> <i>Alternaria</i> <i>Fusarium</i> <i>Penicillium</i>	Ciccarese et al. (2007)
Barley		Fungal spores (not specified)	Allen et al. (2003)
Soybean		<i>Fusarium</i>	Gomes et al. (2020)
Peanut	AFs		Proctor et al. (2004); Chen et al. (2014); Li et al. (2019); Diao, Hou, Chen, et al. (2013)
	AFs	<i>Aspergillus flavus</i> <i>Aspergillus parasiticus</i>	de Alencar et al. (2012)
	AFs	<i>Aspergillus flavus</i> <i>Aspergillus niger</i>	Abdel-Wahhab et al. (2011)
		<i>Aspergillus flavus</i> <i>Aspergillus parasiticus</i> <i>Cladosporium</i> <i>Penicillium</i> <i>Rhizopus</i>	Laureth et al. (2019)
Cottonseed meal Peanut meal	AFs		Dwarakanath et al. (1968); Dollear et al. (1968); Rayner et al. (1971)
Pistachio	AFs		Akbas and Ozdemir (2006)
Brazil nut	AFs	<i>Aspergillus flavus</i> <i>Aspergillus parasiticus</i>	Giordano et al. (2012)
		<i>Aspergillus flavus</i>	Freitas-Silva et al. (2013); de Oliveira et al. (2020)
Dried figs	AFs	<i>Aspergillus flavus</i> <i>Aspergillus parasiticus</i> <i>Aspergillus niger</i> <i>Cladosporium cladosporioides</i> <i>Mucor hiemalis</i> <i>Mucor plumbeus</i> Bonord <i>Mucor racemosus</i> Fres	Zorlugenç et al. (2008)
		Fungal spores (not specified)	Öztekin et al. (2006)

(continues)

TABLE 1 Continued

Product	Target of ozone	References
Dried aromatic plants	<i>Alternaria</i> <i>Aspergillus</i> <i>Cladosporium</i> <i>Fusarium</i> <i>Penicillium</i> <i>Ulocladium</i>	Kazi et al. (2018)
Red pepper	AFs	Inan et al. (2007)
Muskmelon	<i>Fusarium sulphureum</i>	Hua-Li et al. (2018)
Dog food	<i>Aspergillus flavus</i>	Silva, Pereira, and Scussel (2018)
Feed	<i>Aspergillus</i> <i>Fusarium</i> <i>Penicillium</i>	Suian Jose, Raquel Bechlin, Werncke, and Christ (2018)
AFB ₁ solutions	AFs	Luo et al. (2013); Puzyr', Burov, Bondar', and Trusov (2010); Diao et al. (2012); Ayranci and Karaca (2018); Luo, Wang, Wang, Li, Zheng, et al. (2014c); Agriopoulou et al. (2016)
Raisins (sultanas)	OTA	Torlak (2019)
Apple juice	Patulin	Diao et al. (2019); Diao, Ren, et al. (2018)
Yeast extract-rose Bengal agar	<i>Alternaria</i> sp. <i>Aspergillus terreus</i> <i>Aspergillus niger</i> <i>Botrytis allii</i> <i>Chaetomium</i> sp. <i>Colletotrichum lagerlariurn</i> <i>Fusarium oxysporum</i> <i>Penicillium egyptiacum</i> <i>Rhizopus stolonifer</i> <i>Stetnphylium sorcinaeforme</i> <i>Stetnphylium loti</i> <i>Trichoderma viride</i> <i>Verticillium albo-atrum</i> <i>Verticillium dahliae</i>	Hibben and Stotzky (1969)
Spore suspensions	<i>Aspergillus ochraceus</i> <i>Aspergillus nidulans</i>	Antony-Babu and Singleton (2009)
	<i>Aspergillus niger</i>	Vijayanandraj et al. (2006)

Recently, Amoah and Mahroof (2019) used O₃ gas in wheat and rice silos to control insect (*Sitophilus oryzae*) infestation. They showed that eggs were more resistant to ozonation than immature and adult forms of *Sitophilus oryzae*. However, 24 hr exposure to 200 mg/L O₃ gas at 5-cm depth of samples inactivated both immature and adult forms of insect.

Wu et al. (2006) observed a high fungal inactivation rate in high-moisture (MC = 21.9%) wheat samples at high temperatures (40 °C). The reason is that high temperatures accelerate the decomposition rate of ozone to free radicals, and also ozone decomposes more readily in water and high-moisture products. Similar results were reported for barley grains by Allen, Wu, and Doan (2003).

High MC and high-temperature conditions are problematic for grain storage; therefore, Wu et al. (2006) studied the effect of ozone fumigation (0.33 mg/g O₃ gas) at 20 °C on wheat grains with MC 21%. Ten minutes exposure was enough to inactivate fungal spores without affecting the germination of seeds. An enhanced fungicidal effect of ozone was observed at high temperature (40 °C) and high water activity ($a_w = 0.9$ corresponding to the wheat MC = 21.9%), resulting in 96.9% and 90.3% fungal spore inactivation, respectively. The highest inactivation rate (96%) was reported for barley grains when high moisture grains (25%) were exposed to 0.16 mg/g O₃ gas followed by a 30 min hold in the sealed reactor (Allen et al., 2003). Mycotoxin in beer can be prevented by the application of ozone

on barley grains prior to the fermentation process (Pascari, Ramos, Marin, & Sanchis, 2018)

Fusarium contamination of wheat is a global problem causing both economical and health problems (Torres et al., 2019). A team of researchers used a mixture of air-ozone to dry wheat grains in the silo. They showed that *Fusarium* contamination was significantly reduced in ozone-air-ventilated grains, whereas *Aspergillus* and *Penicillium* survived the process (Raila, Lugauskas, Steponavičius, Railienė & Steponavičienė, 2006). They observed a higher inactivation rate in high moisture grains (22%), similar to those reported by Wu et al. (2006). Later, White (2007) recommended ozone fumigation of high-moisture corn to reduce both fungal and mycotoxin contamination of corn grains during storage. They found *Fusarium* and *Aspergillus* to be sensitive to O₃ gas, whereas *Penicillium* and *Rhizopus* were found to be very resistant to ozone fumigation in corn silos (White et al., 2013). The ozone susceptibility of *Fusarium* was later confirmed (Piacentini, Savi, & Scussel, 2017; Savi et al., 2014; Savi et al., 2015). In a series of studies, they showed complete growth inhibition of *Fusarium graminearum* after 120-min ozone fumigation followed by *Aspergillus flavus* (160 min) and *Penicillium citrinum* (180 min) (Savi et al., 2015; Savi et al., 2014). For small sample size, fungal growth inhibitions observed at a shorter exposure time (Savi & Scussel, 2014). Trombete et al. (2017) also showed that wheat volume in the silo was a key factor in the effectiveness of the ozonation process. The highest growth inhibition rates (3-log reduction) for both *Fusarium* and *Aspergillus* were reported for a small sample volume (2 kg) of wheat exposed to a high concentration (60 mg/L) of O₃ gas over a long period of time (300 min). Similar results were found by Granella, Christ, Werncke, Bechlin, and Coelho (2018), who reported a significant fungal count reduction (92.86%) after 45-min ozonated air-drying at 50 °C. Regardless of the type of ozone, both White et al. (2013) and Raila et al. (2006) found *Fusarium* to be the most susceptible strain to ozonation. The difference between the sensitivity of *Aspergillus* in their studies can be attributed to the type of ozone.

Fumigation of wheat with 40 mg/kg of O₃ gas for 20 min inhibited the growth of *A. flavus* by more than 95% (El-Desouky, Sharoba, El-Desouky, El-Mansy, & Naguib, 2012). Similar results were reported for maize seeds exposed to O₃ gas for a short period of time (83.3% *A. flavus* growth reduction after 10-min exposure to 2 g/min O₃ gas) (Hussein, Tuama, & Ali, 2015) as well as a 2-log reduction in *A. flavus* count after corn seeds passed three times through a screw conveyor with a continuous flow of ozone gas (47,800 mg/kg) (McDonough et al., 2011).

McDonough et al. (2011) reported a 95% reduction in *A. flavus* count after one single pass through the ozonated

conveyor system. Three-log reduction was reported in *Fusarium* and *Aspergillus* count in corn grits after 480-min exposure to 60 mg/L O₃ gas (Porto et al., 2019). Fungal count reduction was related to oxidative stress, metabolism disruption, and apoptosis that were induced by ozone.

In a similar study, White, Murphy, Bern, and van Leeuwen (2010) observed fungal growth on high-moisture (26%) corn samples fumigated with O₃ gas for 24 hr. They reported that only *Penicillium* survived at a high concentration of ozone (2 mg/kg O₃ gas per min for 24 hr). The order of fungal susceptibility to ozone was as follows: *Fusarium* > *Aspergillus* > *Mucor* > *Penicillium*. Their observations were in agreement with previous studies that reported *Penicillium* as the most ozone-resistant fungal strain. Similarly, *Penicillium* survived on wheat and barley seeds treated with 3% of their weight O₃ gas; however, pea seeds showed *Aspergillus* contamination after the same ozone fumigation conditions as wheat and barley (Cicarese, Sasanelli, Ciccarese, Ziadi, & Mancini, 2007).

Later, Mylona (2012) studied the effect of ozonation on *F. graminearum*, *F. verticillioides*, and *F. langsethiae* and observed 2-log reduction in the *Fusarium* population. The efficiency of high concentration of ozone (200 mg/kg) was not consistent for all three *Fusarium* spp.; however, complete inhibition was observed in low-moisture samples ($a_w = 0.94$) treated with 200 mg/kg O₃ gas for 30 min. Germination was also delayed at these conditions. Gomes et al. (2020) reported the total elimination of *Fusarium* in soybeans after 180-min exposure to O₃ gas in a bench-scale silo.

Antony-Babu and Singleton (2009) studied the effect of ozonation on *A. ochraceus* and *A. nidulans* and observed that, regardless of the concentration of ozone and exposure time, fungal structural development was affected in ozone-treated samples. *Aspergillus nidulans* failed to produce branched filaments and spores. Although *A. ochraceus* sporulated under ozone, its growth was very slow and limited. Some *A. niger* spores also survived the ozonation process; however, the survived spores formed sterile mycelia after germination. The colonies were not uniform, appeared as gray patches of mycelia, and failed to produce spores (Vijayanandraj, Nagendra Prasad, Mohan, & Gunasekaran, 2006).

Aspergillus contamination is a serious problem in nuts such as pistachios, almonds, and peanuts (Kluczkowski, 2019). Abdel-Wahhab et al. (2011) used O₃ gas to control fungal growth and reduce AF levels in peanut kernels. They reported that *Aspergillus* growth inhibition was dose dependent, and the ability of O₃ gas to reduce spore production was affected by the sugar content of the growth media. Laureth, Christ, Ganascini, and Coelho (2019) exposed peanut kernels to 50 mg/kg O₃ gas for 60 min and observed a significant fungal reduction (75.79% in peanut

grains and 82.66% in peanut pods) due to the rupture of the fungal cell envelope. The peroxide value of the kernels remained unchanged; however, the electrical conductivity of the kernels was affected by ozonation. de Alencar et al. (2012) reported an 80% reduction in the number of *A. flavus* and *A. parasiticus* in peanut kernels treated with 21 mg/L O₃ gas for 96 hr. They also reported the depigmentation of *Aspergillus* colonies due to the disorganization of fungal structure and oxidation of vital cell components. de Alencar et al. (2012) explained that ozone disrupts the cells through the following mechanisms: (a) oxidizing sulfhydryl and amino acid groups of protein and enzymes and (b) oxidizing polyunsaturated fatty acids. Aguilar et al. (2018) showed a first-order kinetic model for fungal pigment degradation during the ozonation process.

Giordano et al. (2012) observed slow *Aspergillus* growth after fumigation with low concentrations of O₃ gas (10 to 14 mg/L); however, 5-hr exposure to 31 mg/L O₃ gas completely inhibited the growth of *A. flavus* and *A. parasiticus* in Brazil nuts. They suggested fumigating Brazil nut containers with O₃ gas before shipping to control fungal growth and reduce AF contamination. A 3.1-log reduction in the *A. flavus* count was reported when contaminated Brazil nuts were exposed to 8.88 mg/L O₃ gas for 240 min (de Oliveira et al., 2020). Brazil nuts exposed to ozonated water (ozone concentration 20 mg/L) showed more than 91% reduction in the viable conidia count of *A. flavus* (Freitas-Silva, Morales-Valle, & Venâncio, 2013).

Öztekin et al. (2006) observed that fungal inactivation by O₃ gas was both time and concentration dependent in dried figs. They recommended the application of a low concentration of O₃ gas (5 mg/kg) over a long period of time (at least 3 hr) to reduce the fungal count in dried figs. They also compared the effectiveness of O₃ gas and ozonated water on fungal flora of dried figs and reported that 15-min exposure to 1.7 mg/L ozonated water completely inactivated *A. flavus*, *A. parasiticus*, and *A. niger*.

The contamination of aromatic plants used in ready-to-eat foods is a serious health concern. It was reported that 60-min ozonation (4 mg/L O₃ gas) reduced the fungal count by 2- to 4-log CFU/g in mountain tea, chamomile, thyme, oregano, and lemon verbena (Kazi, Parlapani, Boziaris, Vellios, & Lykas, 2018). Ozonation was suggested as a safe method to reduce the microbiological risks associated with aromatic plants. Ozonation also reduced fungal contamination in feed. Suian Jose, Raquel Bechlin, Werncke, and Christ (2018) showed 92.37% fungal spore reduction in a 10-cm layer of Sunn hemp seeds (*Crotalaria Spectabilis*) after 102.7-min ozonation. Silva, Pereira, and Scussel (2018) suggested using ozonation for fungal inactivation in dog feed. They showed a 98.3% reduction in *A. flavus* spore count after 120-min exposure to 40 µmol/mol O₃ gas. All the abovementioned studies showed that both

O₃ gas and ozonated water can inhibit the growth of mycotoxin-producer fungi, reduce mycotoxin formation, and degrade mycotoxins in agricultural crops. The sensitivity of different fungal strains to ozone is affected by the following factors: type of ozone, growth level, pH, humidity, temperature, and presence of other chemicals or compounds such as organic materials (Zorlugenç et al., 2008), and it is different in fruits than that in grains (Hua-Li et al., 2018). The optimum O₃ gas fumigation conditions depend on the type of food and its components. These optimum conditions must be evaluated for different crops and different varieties of a particular crop.

Table 1 presents the application of ozone for the reduction of fungal microbiota and the degradation of six major mycotoxins in different foods and agricultural products.

5 | ADVANTAGES, LIMITATIONS, AND FUTURE OUTLOOK

Ozone is a safe antimicrobial agent in food industries (FDA, 2001) and has remarkable benefits; however, ozone technology has some limitations. The advantages of ozone technology are as following.

5.1 | Environment-friendly decontaminating agent

Ozone has been widely used in food industries for sanitation and surface decontamination (Guzel-Seydim, Greene, & Seydim, 2004). O₃ gas has a short half-life and quickly decomposes to form oxygen and does not leave any residue on food (Pandiselvam et al., 2019). Ozone effectively kills a broad range of microorganisms such as mycotoxin-producing fungi and can be used as a residue-free fungal control agent. To the best of our knowledge, there is no other compound that can effectively control fungal contaminants without leaving residue on food.

5.2 | Recycling and reusing water

The waste water from food industries contains a broad range of microorganisms such as bacteria and fungi and particularly with the spores of these microorganisms. Ozone showed to be effective in waste water decontamination and lowering the biological oxygen demand and chemical oxygen demand of the water. Application of ozone on waste water can reduce its microbial load, increase the reusability of the processed water, and provide a chance for food industries to perform environment-friendly operations (Kim, Yousef, & Khadre, 2003). This is particularly important in processing plants that process

agricultural products with a high fungal load such as fig, pistachio, and peanut processing plants.

5.3 | Production of organic food

Ozone is a natural and residue-free compound that can be used in the production of “organic” food (Pandiselvam et al., 2019).

5.4 | Limitations of ozone technology

5.4.1 | Instability and quick decomposition

O₃ gas quickly decomposes to form Oxygen, and this decomposition is faster in water than in air. When ozone dissolves in water, it forms O₂ and highly reactive hydroxyl radicals. The rate of decomposition in water depends on the type and concentration of organic solutes. Primary alcohols and acids promote decomposition of ozone; however, bicarbonate and *tert*-butyl alcohol inhibit ozone decomposition (Oner & Demirci, 2016). Ozone decomposition is faster in alkaline pH values. In addition to the chemical compounds and pH of the solution, the half-life of ozone is affected by other factors such as temperature, type of treatment, and type of food. Ozone decomposition in the air is facilitated by ventilation. Due to the instability of ozone, it must be generated near its point of application and must be kept constant during the process (Abd-Elsalam et al., 2020; Pankaj et al., 2018; White, 2007).

5.4.2 | Exposure and health effects

Ozone is highly reactive and reacts with organic substances such as a human body. It is very important to continuously monitor both the work environment and people who may have contact with ozone. O₃ gas mainly affects the respiratory tract of human and specific care must be taken while working with it or being in an ozone-exposed environment (Zhu, 2018). Depending on the exposure time and concentration of ozone, acute or chronic toxicity may occur. Symptoms of ozone toxicity include burning sensation in the throat and eyes, cough, dizziness, and headache. Chronic toxicity symptoms are decreased memory, increased muscular excitability, and bronchitis (Guzel-Seydim et al., 2004). Figure 2 shows three health effect zones, which are defined based on the concentration of ozone in an environment: (1) *acceptable zone*, (2) *hazardous zone*, and (3) *critical zone* (White, 2007). Due to the health problems associated with ozone exposure,

human and animal contact must be limited. In the United States, a limit of 0.1 mg/L O₃ gas and an exposure time of 8 hr is set by Occupational Safety and Health Administration (OSHA) (Rakness, DeMers, & Blank, 1996). There is no shift limitation for ozone fumigation of grains in the silo; however, ozone fumigation of other agricultural crops in the storage room is usually performed during the night shift or when workers are not around. Potassium iodine solution is used by researchers to absorb the excess O₃ gas during the experiments and to prevent it from being released into the air.

It is very important to take safety precautions while working with ozonated water. Ozonated water (at concentrations more than 1 mg/kg) can release O₃ gas to the air, exceeding the safe level for workers. Warm water, high pressurized water, and small-size droplets increase the chance of toxicity. It is recommended to shroud over the application area or to use a fan to discharge the air outside (Smilanick, 2003).

5.4.3 | Corrosiveness

Ozone has a high oxidation/reduction potential (−2.07 V) (Garud et al., 2019; Pandiselvam et al., 2019), and due to the highly corrosive properties of ozone, equipment made of metal cannot be used in ozone fumigation systems. The use of ozone in food processing plants increases the chance of metal rusting and the release of metal pieces in food (Abd-Elsalam et al., 2020). Rubber or plastic containers can be used during ozonation but must be checked frequently for damage and cracks. Ozone-resistant heavy-duty chambers and equipment must be designed and developed for ozonation purposes (Gabler, Smilanick, Mansour, & Karaca, 2010). According to Jian et al. (2013), storage and processing equipment will be corroded in 2 months at high O₃ gas concentrations (47 to 106 mg/L).

5.4.4 | Penetration limitation

Ozone penetration is limited to the surface topography of the target (Boff, 1999). It has been shown that ozone fumigation in the silos has two distinct phases; in the first phase, O₃ gas had a very slow movement and rapid degradation, whereas in the second phase, O₃ gas had a free flow and little degradation. Ozone fumigation parameters had been optimized for the typical corn storage systems, and more than 85% of O₃ gas penetrated 2.7 m into the grains at the optimum conditions (Kells et al., 2001). To achieve higher efficiency in ozone fumigation of grains in small-size ozone treatment chambers, it was recommended to hold grains (30 min) in the sealed reactor after

FIGURE 2 Health effect zones of O_3 gas.

fumigation (Allen et al., 2003). Continuous stirring, both in ozone treatment chambers and washing tanks, can help to increase the surface contact. Constant stirring will expose more fungal filaments and spores to ozone and increase the efficiency of ozone treatment.

Toxin contamination may be either external or internal contamination. Ozone is a useful tool for the degradation of toxins on the external sites of crops but cannot penetrate the internal sites of colonization and toxin formation. Ozone efficiency is higher in artificially AF-contaminated samples due to the presence of toxin on the surface. Naturally contaminated kernels may have AFs inside the cotyledon, which results in less penetration and less efficiency (Diao, Hou, Chen, et al., 2013). It is suggested to increase the exposure time to solve the penetration limitation and increase ozonolysis efficiency. However, maintaining the quality attributes of the ozonated product should be taken into account (Udomkun et al., 2017).

5.4.5 | Presence of organic matter

The presence of ozone-reactive compounds in water can affect the efficiency of ozonated water. Water must be pre-

conditioned to reduce organic compounds and particulates (Smilanick, 2003). Organic matter (such as suspended matter in the rinse water or seed coatings) must be eliminated through a continuous filtration process (Boff, 1999)

5.4.6 | Formation of new products

Formation of new products is one of the main concerns associated with the ozone treatment of mycotoxins (Peng, Marchal, et al., 2018). There are three main degradation mechanisms for AFs: hydration, hydrogenation, and oxidation of the furan ring (Pankaj et al., 2018). Diao et al. (2012) described two ozonolysis pathways for degradation of AFB₁ and identified 13 ozone degradation products. All ozonolysis products showed significantly reduced toxicity. Luo et al. (2013); Luo, Wang, Wang, Li, Zheng, et al., 2014c) identified six ozone degradation products for AFB₁, none of them showed adverse effects on toxicity tests. Although AFB₁-ozone-degraded products did not show any adverse effect on the toxicity tests, more research is needed to study the AFB₁ ozonation products in highly contaminated samples and assure that the ozonated products are safe for animal and human consumption. Toxicity test on

ozonolysis products is a crucial obstacle in ozonation of mycotoxins, and more animal tests, as well as risk assessment on degradation products, must be done to assure the safety of ozonated products. Although no toxicity has been reported for the ozone-treated product of mycotoxins, there is a concern about the toxicity of these products after digestion. According to Freire and Sant'Ana (2018), such products may convert to their parent mycotoxin during digestion. These compounds can be toxic or have a higher bioavailability than their parent mycotoxin. Due to the potential health risks, more research on the chemical structure and toxicity of the digestion products is needed.

5.4.7 | Nutrients and sensory qualities

The nature of food, its constituents, pH, and MC affect the efficacy of ozone treatment (Pankaj et al., 2018). For example, hydrophobic surfaces such as fruits and vegetables increase the consumption rate of ozone (Boff, 1999). It is very important that the ozone-treated food should retain its appearance, nutrition value, and its overall quality. As several factors affect the detoxification ability of ozone treatment, conditions resulting in the highest fungal inactivation and maximum toxin degradation with minimum physicochemical effect on food products must be evaluated for every individual food product.

Studies showed that ozonation of pistachio and peanut kernels did not affect the fatty acid composition of nuts and no significant changes observed between the overall palatability, sweetness, flavor, and rancidity of ozonated and nonozonated pistachios and peanuts (Akbas & Ozdemir, 2006; de Alencar et al., 2012; Li et al., 2019). Ozone treatment of Brazil nuts did not affect the lipid profile of the raw Brazil nut oil (de Oliveira et al., 2020). However, hazelnut oil treated with ozone showed increased sensitivity to oxidation (Uzun & Ibanoglu, 2018). Unsaturated fatty acids of AF-contaminated corn were more susceptible to oxidation, and ozone treatment resulted in an increase in palmitic acid and saturated fatty acids (Jr & King, 2002).

Ozone treatment did not affect the color and overall appearance of ozonated red peppers, wheat, and rice (Inan et al., 2007; Wang et al., 2010). Ozone fumigation improved the storage property of wheat, corn, and rice grains (less fungal growth and mycotoxin) (Ferreira et al., 2018). According to Zhu (2018), moderate ozone treatments significantly increased the milling properties of wheat, swelling power of starch, and viscosity of dough. Excessive ozone treatment denatured wheat protein (such as gluten and glutenin) and affected dough rheology. Ozone treatment did not affect phytate, vitamin, and lipid content of wheat kernels; however, alpha amylase activity in ozone-

treated wheat flour was decreased. Ozonation of rice grains increased the viscosity of rice flour due to the enzyme inactivation but had no effect on the gelatinization properties of rice flour. Ozonation increased the lightness of wheat, sorghum, and corn flour.

During ozone fumigation in the storage rooms, O₃ gas reacts with atmospheric water in the storage room and decrease the RH of the air. RH of the storage room must be controlled and corrected during the ozone fumigation process to avoid drying of products (Freitas-Silva & Venâncio, 2010). Although some minor changes had been reported, these changes seem negligible compared to the benefits of ozone treatments and O₃ gas treatment can be used as a safe and green technology for food preservation and control of contaminants.

5.4.8 | Dose, time, and temperature dependency

The effectiveness of the ozonolysis process depends on ozone concentration, exposure time, and temperature. High temperature has a negative influence on the half-life of ozone, and ozone quickly decomposes at temperatures higher than 50 °C (Diao, Hou, & Dong, 2013). However, a high AFB₁ degradation rate was reported for cottonseed meal (Dwarakanath et al., 1968), peanut kernels, and peanut flour at temperatures above 50 °C (Proctor et al. (2004). Although high temperature accelerates the decomposition rate of ozone to free radicals and increases the effectiveness of the ozonation process, it cannot be used for most of the agricultural products due to its negative impact on the quality attributes of the products. Therefore, ozonation is usually performed at room temperature and the optimal conditions are set by adjusting the exposure time and concentration of ozone.

Ozonolysis efficiency increases with the increase in exposure time and ozone concentration. However, a high concentration of ozone and long exposure times have deteriorative effects on the quality attributes of ozone-treated food, and ozone application should not exceed a certain threshold (Diao, Hou, & Dong, 2013; Isikber & Athanasios, 2015). El-Desouky et al. (2012) showed that longer exposure time was more effective than higher O₃ concentration. Ozonation process must be optimized to find a combination of exposure times and concentration of ozone that results in the highest degradation of mycotoxins without affecting the nutritional value and quality of the product.

Ozone fumigation for fungal reduction is also performed at room temperature but uses higher ozone concentrations than the one used for mycotoxin reduction. The reason behind it is that a higher ozone dose is required to

penetrate the thick cell wall of fungal spores. Fumigation process must be optimized to find the optimum concentration of ozone and exposure time. It should be noted that fungal sensitivity to ozone depends on the fungal strain, growth level, physiological state of cells, RH, and components of the growth media (El-Desouky et al., 2012; Kim, Yousef, & Dave, 1999). Due to different reactions of different fungal species to ozonation, ozone has not been considered as a clear “fumigant” yet (Isikber & Athanassiou, 2015).

5.4.9 | Effect of MC and RH

Water content of the products is a key factor in the reduction of mycotoxins by ozone. Slower ozone penetration (Raila et al., 2006) and shorter ozone half-life (de Alencar et al., 2012) have been reported for grains with high moisture content. It is speculated that the activity of ozone is limited by the moisture of the food. However, higher fungal decontamination was reported in wheat grains with high moisture content. Raila et al. (2006) explained that ozone had a slow penetration rate (0.75 m) due to the high moisture of the grains (23.2%); therefore, it had a longer reaction with fungal strains on the surface of the grains.

RH is a key factor in the effectiveness of the ozonation process. The higher the RH, the more efficient the ozonation process (de Alencar et al., 2012). Dry and nongerminating spores are more resistant to ozone than the moist and germinating fungal spores (Hibben & Stotzky, 1969). Further research is needed to study the effect of the MC of the food on the effectiveness of the ozonation process.

5.4.10 | Ozonated water, ozone mist, or ozone gas

Effectiveness of ozonation methods depend on several factors such as type of food, size of particles, its constituents, and moisture content. Gaseous O_3 is more effective in mycotoxin reduction, whereas aqueous O_3 is known for its fungal growth control ability (Öztekin et al., 2006; Palou et al., 2001). Some studies compared the effectiveness of ozonated water, O_3 gas, and ozone mist in the degradation of AFBs. O_3 gas was more effective than ozonated water in the reduction of AFB₁ level in contaminated dried figs (95.21% and 88.62%, respectively) (Zorlugenç et al., 2008); however, different results were reported for grains. Ozonated water was more effective in the reduction of AFB₁ (92.2%) in wheat samples, followed by ozone mist (85.5%) and O_3 gas (56.8%). Ozone mist was the most effective method for the reduction of AFB₁ in paddy rice (94.4%)

followed by ozonated water (87.4%) and O_3 gas (70.8%) (Wang et al., 2010). It is believed that the superiority of ozone mist and ozonated water to ozone gas is related to the reaction of ozone with water resulting in free OH radicals with stronger oxidation ability than ozone itself.

5.5 | Future outlook

5.5.1 | Developing cost-effective and versatile ozone generation systems

Recent increase in the demand for ozone generators by different industries in combination with the advancement of manufacturing technology has resulted in the availability of more cost-effective ozone generators. Although modern ozone generators have low energy consumption, low maintenance, little metallic dust generation, and produce high concentrations of O_3 gas (Freitas-Silva & Venâncio, 2010), for some applications, they are still costly. Reducing the cost of ozone generators could increase their application in other industries. Another limiting factor for adopting ozone generators is their size and specifications. For example, most commercially available ozone generators are bulky and designed to work with an electrical supply of 120 or 240 V. For some applications in agriculture, there is a need for a compact and rugged system that can work with a 12- or 24-V DC supply. To expand the use of ozone to other applications, further research and development are needed to develop more compact and rugged systems that can work in harsh environments with a broad range of available voltage.

5.5.2 | Regulatory approval

FDA and many European countries had approved O_3 gas as a safe antimicrobial agent on food; however, the use of ozone as an antimicrobial agent is subjected to regulation as a pesticide under Environmental Protection Agency (EPA) rules (Loeb, 2018; FDA, 2001; Rakness et al., 1996). EPA Regulations of Pesticides categorized ozone generators and UV light systems as pest control devices designed to destroy or inactivate microbial pests (Keith & Walker, 1992; Tiwari & Rice, 2012). Ozone generators can be used for pest control (such as fungal control) and do not need FDA approval (Keener & Misra, 2016); however, such devices must be registered by EPA or have been made by EPA-registered establishments (Tiwari & Rice, 2012).

Although ozone technology proved to be effective in mycotoxin reduction, there are no regulations to allow detoxifying mycotoxin-contaminated crops for human use (Karaca & Velioglu, 2007). From the regulatory

FIGURE 3 Advantages, limitations, and future outlook of ozone technology.

perspective, approval of ozonation as a new detoxification and fungal control method requires a significant amount of data from both research and animal test results.

5.5.3 | Consumer confidence

Consumer attitudes toward an ozone-treated product is a key point. The majority of consumers perceive new technologies (such as food irradiation) as risky, unknown, and unacceptable (Keener & Misra, 2016). It is unclear how the public will react to ozone-treated crops. It is also very time-consuming and expensive to collect consumer opinion data.

Media and advertisements play an important role in providing information to the public and make them aware of the benefits of ozone technology for food industries as well as environmental and economic benefits. It is also the responsibility of researchers and scientists involved in the development of ozone technology to share their knowledge and findings with policymakers and the public to increase the awareness of the potential for ozone technology in food industries and the agricultural sector.

Although ozonation (in any form of O₃ gas, ozonated water, or ozone mist) has been proven as an effective

method for the inactivation of mycotoxins in contaminated agricultural crops, its acceptability and suitability had not been evaluated yet. Future research is needed to determine the durability, safety, and efficacy of the ozonation procedure to reduce mycotoxigenic fungi and mycotoxins in food as well as the safety of ozone-treated food products. The advantages, limitations, and future outlook of ozone technology is presented in Figure 3.

6 | CLOSING REMARKS

Ozone technology has a unique potential to be widely used in the food industry. Ozonation is a chemical-free and residue-free process capable of replacing current chemical decontamination methods. Although ozone has not been considered as a clear “fumigant” yet, ozone technology is potentially a green alternative to conventional chemical fumigation.

Numerous studies indicated that ozone could reduce mycotoxin level in food; however, there are notable variations on the exposure times and dose rates. Overall, higher levels of ozone concentration are needed for mycotoxin reduction than are required for suppressing mycotoxin-producer fungi growth. The physical state of food (e.g.,

liquid or solid, granular, or powder), level of fungal contamination, and nutritional factors in food (such as lipid content) should be considered when choosing ozone treatments. However, the grain mass and germination can be negatively affected by ozone treatment. These adverse effects can be lessened using a low concentration of ozone on high-moisture grains. Proper agitation can prevent clumping in flours and powders and increase the effectiveness of the ozonation process. Using ozone for patulin reduction in apple juice can negatively affect the total soluble solids content of the juice.

Although ozonation lowers the number of microbial contaminants, postozonation contamination can reduce the effect of ozone treatment. Ozonation can then be followed by coating fruits with a protective layer (such as edible wax) to prevent postozone treatment contamination and mycotoxin production. Other combinations such as ozonation + UV irradiation (for grains, peanut, and apple juice), ozonated water + pH adjustment using organic acids (for washing fruits), ozone gas + high pressure (for flours), and ozone gas + heat treatment (for dry fruits such as figs) can be used to reduce the ozone concentration or exposure time and moderate the negative impacts of ozone on the physicochemical properties of the ozone-treated product.

Despite the significant potential of ozone technology, this technology has not been widely utilized by the food industry. Low penetration, short half-life, corrosiveness, temperature dependency, and safety issues are examples of some limitations in the application of ozone at the commercial scale. More research is needed to address the earlier mentioned problems, standardize the application conditions, and deliver safe, cost-efficient, and effective ozonation technology for the industry. Most ozone applications require the application of ozone at a constant concentration within a limited range of temperature and humidity. Application of ozone in food processing plants, storage rooms, and packaging houses necessitates changes in design and equipment, process modification, and new training. The cost associated with these changes could also be a factor in the adaptations of this technology. In addition to issues that require more fundamental research, some issues need to be addressed by engineers. Issues such as how to resolve the low penetration of ozone in powdery materials or how to develop a better high throughput ozone application system for different types of food industries are examples of engineering research that are needed.

Despite all the challenges involved in its application, the future looks very promising for the use of ozone technology in the agricultural and food industries. Ozone technology has the potential to be used as a viable biofumigant against fungal contaminants and reduce mycotoxin contamination in the food industry.

AUTHOR CONTRIBUTIONS

Dr. Afsah-Hejri: investigation, writing original draft (introduction, mycotoxin degradation methods, aflatoxins, fungal microbiota, challenges, future outlook, closing remarks, and tables), visualization, and revision of the completed document. Dr. Hajeb: writing original draft and revision of the other mycotoxins. Dr. Ehsani: review and editing the manuscript.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

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REFERENCES

- Aafia, S., Rouf, A., Kanojia, V., & Ayaz, Q. (2018). Ozone treatment in prolongation of shelf life of temperate and tropical fruits. *International Journal of Pure & Applied Biosciences*, 6(2), 298–303.
- Abbès, S., Salah-Abbès, J. B., Sharafi, H., Jebali, R., Noghabi, K. A., & Oueslati, R. (2013). Ability of *Lactobacillus rhamnosus* GAF01 to remove AFM1 in vitro and to counteract AFM1 immunotoxicity in vivo. *Journal of Immunotoxicology*, 10(3), 279–286. <https://doi.org/10.3109/1547691X.2012.718810>
- Abd-Elsalam, K. A., El-Naggar, M. A., Ghannouchi, A., & Bouqel-lah, N. A. (2020). Nanomaterials and ozonation: Safe strategies for mycotoxin management. In M. Rai and K. A. Abd-Elsalam (Eds.), *Nanomycotoxicology* (pp. 285–308). Cambridge, MA: Academic Press.
- Abdel-Wahhab, M. A., Sehab, A. F., Hassanien, F. R., El-Nemr, S. E., Amra, H. A., & Abdel-Alim, H. A. (2011). Efficacy of ozone to reduce fungal spoilage and aflatoxin contamination in peanuts. *Journal of Nuts*, 2(04), 1–14.
- Afsah-Hejri, L. (2013). Saprophytic yeasts: Effective biocontrol agents against *Aspergillus flavus*. *International Food Research Journal*, 20, 3403–3409.
- Afsah-Hejri, L., & Jinap, S. (2013). Influence of different mobile phase compositions on detection of Ochratoxin A. *Food Control*, 31(1), 244–250. <https://doi.org/10.1016/j.foodcont.2012.09.027>
- Afsah-Hejri, L., Jinap, S., Arzandeh, S., & Mirhosseini, H. (2011). Optimization of HPLC conditions for quantitative analysis of aflatoxins in contaminated peanut. *Food Control*, 22(3–4), 381–388. <https://doi.org/10.1016/j.foodcont.2010.09.007>
- Afsah-Hejri, L., Jinap, S., Hajeb, P., Radu, S., & Shakibazadeh, S. (2013). A review on mycotoxins in food and feed: Malaysia case study. *Comprehensive Reviews in Food Science and Food Safety*, 12(6), 629–651. <https://doi.org/10.1111/1541-4337.12029>
- Afsah-Hejri, L., Jinap, S., & Mirhosseini, H. (2012). Ochratoxin A quantification: Newly developed HPLC conditions. *Food Control*, 23(1), 113–119. <https://doi.org/10.1016/j.foodcont.2011.06.020>
- Afsah-Hejri, L., Jinap, S., & Radu, S. (2013). Occurrence of aflatoxins and aflatoxigenic *Aspergillus* in peanuts. *Journal of Food, Agriculture and Environment*, 11(3–4), 228–234.
- Agriopoulou, S., Koliadima, A., Karauskakis, G., & Kapos, J. (2016). Kinetic study of aflatoxins' degradation in the presence of ozone.

- Food Control*, 61, 221–226. <https://doi.org/10.1016/j.foodcont.2015.09.013>
- Agriopoulou, S., Stamatelopoulou, E., & Varzakas, T. (2020). Advances in occurrence, importance, and mycotoxin control strategies: Prevention and detoxification in foods. *Foods*, 9(2), 137.
- Aguilar, D., Morales-Oyervides, L., Contreras-Esquivel, J. C., Méndez-Zavala, A., Raso, J., & Montañez, J. (2018). Effect of ozone processing conditions on stability of fungal pigments. *Innovative Food Science & Emerging Technologies*, 45, 255–263.
- Akbas, M. Y., & Ozdemir, M. (2008). Effect of gaseous ozone on microbial inactivation and sensory of flaked red peppers. *International Journal of Food Science & Technology*, 43(9), 1657–1662. <https://doi.org/10.1111/j.1365-2621.2008.01722.x>
- Akbas, M. Y., & Ozdemir, M. (2006). Effect of different ozone treatments on aflatoxin degradation and physicochemical properties of pistachios. *Journal of the Science of Food and Agriculture*, 86(13), 2099–2104. <https://doi.org/10.1002/jsfa>
- Al-Ahmadi, S. S., Ibrahim, R. A., & Ouf, S. A. (2016). Possible control of fungal and insect infestation of date fruits using ozone. *Bio-sciences Biotechnology Research Asia*, 6(1), 17–28.
- Aldred, D., Magan, N., & Olsen, M. (2004). The use of HACCP in the control of mycotoxins: The case of cereals. In N. Magan & M. Olsen (Eds.), *Mycotoxins in food: Detection and control* (pp. 139–173). Boca Raton, FL: CRC Press.
- Alexandre, A. P., Castanha, N., Calori-Domingues, M. A., & Augusto, P. E. (2017). Ozonation of whole wheat flour and wet milling effluent: Degradation of deoxynivalenol (DON) and rheological properties. *Journal of Environmental Science and Health, Part B*, 52(7), 516–524. <https://doi.org/10.1080/03601234.2017.1303325>
- Alexandre, A. P. S., Castanha, N., Costa, N. S., Santos, A. S., Badiale-Furlong, E., Augusto, P. E. D., & Calori-Domingues, M. A. (2019). Ozone technology to reduce zearalenone contamination in whole maize flour: Degradation kinetics and impact on quality. *Journal of the Science of Food and Agriculture*, 99(15), 6814–6821.
- Alexandre, A. P. S., Vela-Paredes, R. S., Santos, A. S., Costa, N. S., Canniatti-Brazaca, S. G., Calori-Domingues, M. A., & Augusto, P. E. D. (2018). Ozone treatment to reduce deoxynivalenol (DON) and zearalenone (ZEN) contamination in wheat bran and its impact on nutritional quality. *Food Additives & Contaminants: Part A*, 35(6), 1189–1199. <https://doi.org/10.1080/19440049.2018.1432899>
- Allen, B., Wu, J., & Doan, H. (2003). Inactivation of fungi associated with barley grain by gaseous ozone. *Journal of Environmental Science and Health, Part B*, 38(5), 617–630. <https://doi.org/10.1081/PFC-120023519>
- Amoah, B. A., & Mahroof, R. M. (2019). Ozone as a potential fumigant alternative for the management of *Sitophilus oryzae* (Coleoptera: Curculionidae) in wheat. *Journal of Economic Entomology*, 112(4), 1953–1963.
- Anene, A., Hosni, K., Chevalier, Y., Kalfat, R., & Hbaieb, S. (2016). Molecularly imprinted polymer for extraction of patulin in apple juice samples. *Food Control*, 70, 90–95.
- Antony-Babu, S., & Singleton, I. (2009). Effect of ozone on spore germination, spore production and biomass production in two *Aspergillus* species. *Antonie van Leeuwenhoek*, 96(4), 413. <https://doi.org/10.1007/s10482-009-9355-2>
- Asokapandian, S., Periasamy, S., & Swamy, G. J. (2018). Ozone for fruit juice preservation. In G. Rajauria & B. K. Tiwari (Eds.), *Fruit juices* (pp. 511–527). Cambridge, MA: Academic Press.
- Atehnkeng, J., Ojiambo, P. S., Cotty, P. J., & Bandyopadhyay, R. (2014). Field efficacy of a mixture of atoxigenic *Aspergillus flavus* Link: Fr vegetative compatibility groups in preventing aflatoxin contamination in maize (*Zea mays* L.). *Biological Control*, 72, 62–70.
- Aygün, Ş. G. (2015). Detoxification of aflatoxin in peanut meal by heating and gamma irradiation (Doctoral dissertation). Middle East Technical University, Ankara, Turkey.
- Ayranci, U. G., & Karaca, H. (2018). *Degradation of aflatoxins by use of ozone gas*. Proceedings, 6th ASM International Congress of Agriculture and Environment (2018), 11–13 October 2018, Antalya, Turkey, pp. 108–114.
- Bhat, R., Rai, R. V., & Karim, A. A. (2010). Mycotoxins in food and feed_ present status and future concerns. *Comprehensive Reviews in Food Science and Food Safety*, 9, 57–81. Retrieved from <https://onlinelibrary.wiley.com/doi/pdf/10.1111/j.1541-4337.2009.00094.x>
- Bhatnagar, D., Brown, R., Ehrlich, K., & Cleveland, T. E. (2002). Mycotoxins contaminating cereal grain crops: Their occurrence and toxicity. In G. G. Khachatourians & D. K. Arora (Eds.), *Applied mycology and biotechnology* (Vol. 2, pp. 171–196). Amsterdam, the Netherlands: Elsevier.
- Boff, J. (1999). The use of ozone in controlling microbial growth on alfalfa sprouts during germination (Doctoral dissertation). The Ohio State University, Columbus, OH.
- Brodowska, A. J., Nowak, A., & Śmigielski, K. (2018). Ozone in the food industry: Principles of ozone treatment, mechanisms of action, and applications: An overview. *Critical Reviews in Food Science and Nutrition*, 58(13), 2176–2201. <https://doi.org/10.1080/10408398.2017.1308313>
- Calado, T., Fernández-Cruz, M. L., Verde, S. C., Venâncio, A., & Abrunhosa, L. (2018). Gamma irradiation effects on ochratoxin A: Degradation, cytotoxicity and application in food. *Food Chemistry*, 240, 463–471.
- Cardwell, K. F., Kling, J. G., Maziya-Dixon, B., & Bosque-Perez, N. A. (2000). Interactions between *Fusarium verticillioides*, *Aspergillus flavus*, and insect infestation in four maize genotypes in lowland Africa. *Phytopathology*, 90(3), 276–284.
- Cataldo, F. (2008). Ozone decomposition of patulin - A micotoxin and food contaminant. *Ozone: Science and Engineering*, 30(3), 197–201. <https://doi.org/10.1080/01919510801925930>
- Cazzaniga, D., Basílico, J. C., González, R. J., Torres, R. L., & De Greef, D. M. (2001). Mycotoxins inactivation by extrusion cooking of corn flour. *Letters in Applied Microbiology*, 33(2), 144–147. Retrieved from <http://www.ncbi.nlm.nih.gov/pubmed/11472523>
- Chen, R., Ma, F., Li, P. W., Zhang, W., Ding, X. X., Zhang, Q., ... Xu, B. C. (2014). Effect of ozone on aflatoxins detoxification and nutritional quality of peanuts. *Food Chemistry*, 146, 284–288. <https://doi.org/10.1016/j.foodchem.2013.09.059>
- Ciccarese, F., Sasanelli, N., Ciccarese, A., Ziadi, T., & Mancini, L. (2007, October). *Seed disinfection by ozone treatments*. Proceedings of the IOA Conference and Exhibition.
- Ciegler, A., Lillehoj, E. B., Peterson, R. E., & Hall, H. H. (1966). Microbial detoxification of aflatoxin. *Applied and Environmental Microbiology*, 14(6), 934–939.
- Cinar, A., & Onbaşı, E. (2019). Mycotoxins: The hidden danger in foods. In S. Sabuncuoglu (Ed.), *Mycotoxins and food safety* (pp. 1–21). Boston, MA: IntechOpen.
- Concha-Meyer, A., Eifert, J. D., Williams, R. C., Marcy, J. E., & Welbaum, G. E. (2015). Shelf life determination of fresh blueberries (*Vaccinium corymbosum*) stored under controlled atmosphere and

- ozone. *International Journal of Food Science*, 2015, 1–9. <https://doi.org/10.1155/2015/164143>
- de Alencar, E. R., Faroni, L. R. D. A., Soares, N., de, F. F., da Silva, W. A., & da Silva Carvalho, M. C. (2012). Efficacy of ozone as a fungicidal and detoxifying agent of aflatoxins in peanuts. *Journal of the Science of Food and Agriculture*, 92(4), 899–905. <https://doi.org/10.1002/jsfa.4668>
- de Oliveira, J. M., de Alencar, E. R., Bassay Blum, L. E., de Souza Ferreira, W. F., Campos Botelho, S. C., Racanicci, A. M., ... da Silva, C. R. (2020). Ozonation of Brazil nuts: Decomposition kinetics, control of *Aspergillus flavus* and the effect on color and on raw oil quality, *LWT - Food Science and Technology*, 123, 109106.
- Deng, J., Chen, W. J., Guo, B. X., & Zhang, Y. (2011). Effect of ozone treatment on the degradation of ochratoxin A and fatty acids in corn. *Food Science*, 21, 12–16.
- Denvir, A. J., McKenzie, K. S., Rogers, T. D., Miller, D. R., Hitchens, G. D., & Andrews, C. C. (2000). *US Patent No. 6,120,822*. Washington, DC: US Patent and Trademark Office.
- Denvir, A., McKenzie, K. S., Rogers, T., Miller, D., Hitchens, G. D., & Andrews, C. (2001). *US Patent No. 6,171,625*. Washington, DC: US Patent and Trademark Office.
- Di Stefano, V., Pitonzo, R., Cicero, N., & D'Oca, M. C. (2014). Mycotoxin contamination of animal feedingstuff: Detoxification by gamma-irradiation and reduction of aflatoxins and ochratoxin A concentrations. *Food Additives & Contaminants: Part A*, 31(12), 2034–2039.
- Diao, E., Hou, H., Chen, B., Shan, C., & Dong, H. (2013). Ozonolysis efficiency and safety evaluation of aflatoxin B1 in peanuts. *Food and Chemical Toxicology*, 55, 519–525. <https://doi.org/10.1016/j.fct.2013.01.038>
- Diao, E., Hou, H., & Dong, H. (2013). Ozonolysis mechanism and influencing factors of aflatoxin B1: A review. *Trends in Food Science and Technology*, 33(1), 21–26. <https://doi.org/10.1016/j.tifs.2013.06.002>
- Diao, E., Wang, J., Li, X., Wang, X., & Gao, D. (2018). Patulin degradation in apple juice using ozone detoxification equipment and its effects on quality. *Journal of Food Processing and Preservation*, 42(7), e13645.
- Diao, E., Ren, D., Liu, T., Zhang, J., Hu, W., & Hou, H. (2018). Ozone detoxification of patulin in aqueous solution and cytotoxic evaluation using human hepatic carcinoma cells. *Toxicon*, 155, 21–26.
- Diao, E., Wang, J., Li, X., Wang, X., Song, H., & Gao, D. (2019). Effects of ozone processing on patulin, phenolic compounds and organic acids in apple juice. *Journal of Food Science and Technology*, 56(2), 957–965.
- Diao, E., Shan, C., Hou, H., Wang, S., Li, M., & Dong, H. (2012). Structures of the ozonolysis products and ozonolysis pathway of aflatoxin B1 in acetonitrile solution. *Journal of agricultural and food chemistry*, 60(36), 9364–9370.
- Doane, P., & Johnson, A. (2018). *US Patent Application No. 15/757,522*. Washington, DC: US Patent and Trademark Office.
- Dollear, F. G., Mann, G. E., Codifer, L. P., Jr., Gardner, H. K., Jr., Koltun, S. P., & Vix, H. L. E. (1968). Elimination of aflatoxins from peanut meal. *Journal of the American Oil Chemists' Society*, 45(12), 862–865.
- Doster, M. A., Cotty, P. J., & Michailides, T. J. (2014). Evaluation of the atoxigenic *Aspergillus flavus* strain AF36 in pistachio orchards. *Plant Disease*, 98(7), 948–956.
- Dwarakanath, C. T., Rayner, E. T., Mann, G. E., & Dollear, F. G. (1968). Reduction of aflatoxin levels in cottonseed and peanut meals by ozonization. *Journal of the American Oil Chemists' Society*, 45(2), 93–95. <https://doi.org/10.1007/BF02890715>
- El-Desouky, T. A., Elbadawy, S. S., Hussain, H. B., & Hassan, N. A. (2018). Impact of insect densities *Tribolium Castaneum* on the benzoquinone secretions and aflatoxins levels in Wheat flour during storage periods. *The Open Biotechnology Journal*, 12(1), 104–111.
- El-Desouky, T. A., Sharoba, A. M. A., El-Desouky, A. I., El-Mansy, H. A., & Naguib, K. (2012). Effect of ozone gas on degradation of aflatoxin B1 and *Aspergillus flavus* fungal. *Journal of Environmental & Analytical Toxicology*, 2(128), 2161–0525. <https://doi.org/10.4172/2161-0525.1000128>
- Elias-Orozco, R., Castellanos-Nava, A., Gaytán-Martínez, M., Figueroa-Cárdenas, J. D., & Loarca-Piña, G. (2002). Comparison of nixtamalization and extrusion processes for a reduction in aflatoxin content. *Food Additives and Contaminants*, 19(9), 878–885. <https://doi.org/10.1080/02652030210145054>
- European Commission. (2006). Commission Regulation (EC) No 1881/2006 of 19 December 2006 setting maximum levels for certain contaminants in foodstuffs. *Official Journal of European Union*, 364, 365–324.
- European Commission. (2007). Commission Regulation (EC) No. 1126/2007 of 28 September 2007 (amending Regulation (EC) No 1881/2006) setting maximum levels for certain contaminants in foodstuffs as regards *Fusarium* toxins in maize and maize products. *Official Journal of the European Union*, 38, 157–160.
- Food and Drug Administration (FDA). (2001). Secondary direct food additives permitted in food for human consumption. *Federal Register*, 66(123), 33829–33830.
- Ferreira, B. C. F., Soares, C. D. S., Dutra, M. O., Rabelo, C. W., & Scuscel, V. M. (2018). Reduction of fungi and mycotoxin decontamination by ozone gas treatment in three stored rice (*Oryza sativa* L.) varieties. *Julius-Kühn-Archiv*. <https://doi.org/10.5073/jka.2018.463.241>
- Freire, L., & Sant'Ana, A. S. (2018). Modified mycotoxins: An updated review on their formation, detection, occurrence, and toxic effects. *Food and Chemical Toxicology*, 111, 189–205.
- Freitas-Silva, O., Morales-Valle, H., & Venâncio, A. (2013). Potential of aqueous ozone to control aflatoxigenic fungi in Brazil nuts. *ISRN Biotechnology*, 2013, 859830.
- Freitas-Silva, O., & Venâncio, A. (2010). Ozone applications to prevent and degrade mycotoxins: A review. *Drug Metabolism Reviews*, 42(4), 612–620. <https://doi.org/10.3109/03602532.2010.484461>
- Frisón, L., Bourilhon, P., Ocampo, H., Ponisio, D., & Basílico, J. (2014). Effects of ozone gas on *Fusarium verticillioides* and *F. proliferatum* strains producers of fumonisin. *Revista Venezolana de Ciencia y Tecnología de Alimentos*, 5(1), 31–42.
- Gabler, F. M., Smilanick, J. L., Mansour, M. F., & Karaca, H. (2010). Influence of fumigation with high concentrations of ozone gas on postharvest gray mold and fungicide residues on table grapes. *Postharvest Biology and Technology*, 55(2), 85–90. <https://doi.org/10.1016/j.postharvbio.2009.09.004>
- Gacem, M. A., Gacem, H., Telli, A., & Khelil, A. O. E. H. (2020). Mycotoxins: Decontamination and nanocontrol methods. In M. Rai and K. A. Abd-Elsalam (Eds.), *Nanomycotoxicology* (pp. 189–216). Cambridge, MA: Academic Press.

- Garud, S. R., Negi, P. S., & Rastogi, N. K. (2019). Improving the efficacy of ozone treatment in food preservation. *Non-thermal Processing of Foods*, 5, 213–233.
- Gil, L., Ruiz, P., Font, G., & Manyes, L. (2016). An overview of the applications of hazards analysis and critical control point (HACCP) system to mycotoxins. *Revista de Toxicología*, 33(1), 50–55.
- Giordano, B. N. E., Nones, J., & Scussel, V. M. (2012). Susceptibility of the in-shell Brazil nut mycoflora and aflatoxin contamination to ozone gas treatment during storage. *Journal of Agricultural Science*, 4(8), 1–10. <https://doi.org/10.5539/jas.v4n8p1>
- Gomes, T., Canever, S. B., Savi, G. D., Piacentini, K. C., Cargnin, M., Furtado, B. G., ... Angioletto, E. (2020). Modeling and experimental of mould disinfestation of soybean silos with ozone. *Ozone: Science & Engineering*, 42(2), 183–193.
- Gonçalves, A. A. (2009). Ozone - An emerging technology for the seafood industry. *Brazilian Archives of Biology and Technology*, 52(6), 1527–1539. <https://doi.org/10.1590/S1516-89132009000600025>
- Granello, S. J., Christ, D., Werncke, I., Bechlin, T. R., & Coelho, S. R. M. (2018). Effect of drying and ozonation process on naturally contaminated wheat seeds. *Journal of Cereal Science*, 80, 205–211.
- Guzel-Seydim, Z. B., Greene, A. K., & Seydim, A. C. (2004). Use of ozone in the food industry. *LWT-Food Science and Technology*, 37(4), 453–460.
- Hansen, L. S., Hansen, P., & Jensen, K. M. V. (2013). Effect of gaseous ozone for control of stored product pests at low and high temperature. *Journal of Stored Products Research*, 54, 59–63.
- Hassan, F. F., Hussein, H. Z., & Hawar, S. N. (2018). Detection and detoxification of aflatoxin B1 from fish feedstuff using microwave and ozone gas. *Ibn Al-Haitham Journal for Pure and Applied Science*, 31(1), 28–36.
- He, J., Zhou, T., Young, J. C., Boland, G. J., & Scott, P. M. (2010). Chemical and biological transformations for detoxification of trichothecene mycotoxins in human and animal food chains: A review. *Trends in Food Science and Technology*, 21(2), 67–76. <https://doi.org/10.1016/j.tifs.2009.08.002>
- Hell, K., Cardwell, K. F., Setamou, M., & Schulthess, F. (2000). Influence of insect infestation on aflatoxin contamination of stored maize in four agroecological regions in Benin. *African Entomology*, 8(2), 169–177.
- Hibben, C. R., & Stotzky, G. (1969). Effects of ozone on the germination of fungus spores. *Canadian Journal of Microbiology*, 15(10), 1187–1196. <https://doi.org/10.1139/m69-215>
- Hormisch, D., Brost, I., Kohring, G. W., Giffhorn, F., Kroppenstedt, R. M., Stackebradt, E., ... Holzapfel, W. H. (2004). *Mycobacterium fluoranthenorans* sp. nov., a fluoranthene and aflatoxin B1 degrading bacterium from contaminated soil of a former coal gas plant. *Systematic and Applied Microbiology*, 27(6), 653–660. <https://doi.org/10.1078/0723202042369866>
- Hua, S. S. T. (2013). Biocontrol of *Aspergillus flavus* by *Pichia anomala*. In A. Mendez-Vilas (Ed.), *Microbial pathogens and strategies for combating them: Science, technology and education* (pp. 1067–1074). Badajoz, Spain: Formatex Research Center.
- Hua, S. S. T., Baker, J. L., & Flores-Espiritu, M. (1999). Interactions of saprophytic yeasts with a nor mutant of *Aspergillus flavus*. *Applied and Environmental Microbiology*, 65(6), 2738–2740.
- Hua-Li, X., Yang, B., Raza, H., Hu-jun, W., Lu-Mei, P., Mi-Na, N., ... Yong-Cai, L. (2018). Detection of NEO in muskmelon fruits inoculated with *Fusarium sulphureum* and its control by postharvest ozone treatment. *Food Chemistry*, 254, 193–200.
- Huertas-Pérez, J. F., Arroyo-Manzanares, N., Hitzler, D., Castro-Guerrero, F. G., Gámiz-Gracia, L., & García-Campaña, A. M. (2018). Simple determination of aflatoxins in rice by ultra-high performance liquid chromatography coupled to chemical post-column derivatization and fluorescence detection. *Food Chemistry*, 245, 189–195.
- Hussein, H. Z., Tuama, R. H., & Ali, A. M. (2015). Study the effect of ozone gas and ultraviolet radiation and microwave on the degradation of aflatoxin B1 produced by *Aspergillus flavus* on stored Maize grains. *Journal of Agriculture and Veterinary Science*, 8, 5–12. <https://doi.org/10.9790/2380-08510512>
- IARC Working Group on the Evaluation of Carcinogenic Risks to Humans, International Agency for Research on Cancer, & World Health Organization. (1993). *Some naturally occurring substances: Food items and constituents, heterocyclic aromatic amines and mycotoxins* (Vol. 56). Geneva, Switzerland: World Health Organization.
- Inan, F., Pala, M., & Doymaz, I. (2007). Use of ozone in detoxification of aflatoxin B1 in red pepper. *Journal of Stored Products Research*, 43(4), 425–429. <https://doi.org/10.1016/j.jspr.2006.11.004>
- Isikber, A. A., & Athanassiou, C. G. (2015). The use of ozone gas for the control of insects and microorganisms in stored products. *Journal of Stored Products Research*, 64, 139–145. <https://doi.org/10.1016/j.jspr.2014.06.006>
- Isikber, A. A., & Öztekin, S. (2009). Comparison of susceptibility of two stored-product insects, *Ephestia kuehniella* Zeller and *Tribolium confusum* du Val to gaseous ozone. *Journal of Stored Products Research*, 45(3), 159–164.
- Ismail, A., Gonçalves, B. L., de Neeff, D. V., Ponzilacqua, B., Coppa, C. F., Hintzsche, H., ... Oliveira, C. A. (2018). Aflatoxin in foodstuffs: Occurrence and recent advances in decontamination. *Food Research International*, 113, 74–85. <https://doi.org/10.1016/j.foodres.2018.06.067>
- Jajić, I., Jakšić, S., Krstović, S., & Abramović, B. (2016). Preliminary results on deoxynivalenol degradation in maize by UVA and UVC irradiation. *Contemporary Agriculture*, 65(3–4), 7–12.
- Jalili, M., & Jinap, S. (2012). Role of sodium hydrosulphite and pressure on the reduction of aflatoxins and ochratoxin A in black pepper. *Food Control*, 27(1), 11–15. <https://doi.org/10.1016/j.foodcont.2012.02.022>
- Jalili, M., Jinap, S., & Son, R. (2011). The effect of chemical treatment on reduction of aflatoxins and ochratoxin A in black and white pepper during washing. *Food Additives and Contaminants*, 28(4), 485–493. <https://doi.org/10.1080/19440049.2010.551300>
- Jalili, M. (2016). A review on aflatoxins reduction in food. *Iranian Journal of Health, Safety and Environment*, 3(1), 445–459.
- Jard, G., Liboz, T., Mathieu, F., Guyonvarc'h, A., & Lebrihi, A. (2011). Review of mycotoxin reduction in food and feed: From prevention in the field to detoxification by adsorption or transformation. *Food Additives & Contaminants: Part A*, 28(11), 1590–1609.
- Jardon-Xicotencatl, S., Díaz-Torres, R., Marroquín-Cardona, A., Villarreal-Barajas, T., & Méndez-Albores, A. (2015). Detoxification of aflatoxin-contaminated maize by neutral electrolyzed oxidizing water. *Toxins*, 7(10), 4294–4314. <https://doi.org/10.3390/toxins7104294>

- Jian, F., Jayas, D. S., & White, N. D. (2013). Can ozone be a new control strategy for pests of stored grain? *Agricultural Research*, 2(1), 1–8. <https://doi.org/10.1007/s40003-012-0046-2>
- Kalagatur, N. K., Kamasani, J. R., & Mudili, V. (2018). Assessment of detoxification efficacy of irradiation on zearalenone mycotoxin in various fruit juices by response surface methodology and elucidation of its in-vitro toxicity. *Frontiers in Microbiology*, 9, 2937.
- Karaca, H., & Sedat Velioglu, Y. (2009). Effects of some metals and chelating agents on patulin degradation by ozone. *Ozone: Science and Engineering*, 31(3), 224–231. <https://doi.org/10.1080/01919510902766662>
- Karaca, H., & Nas, S. (2009). Combined effect of pH and heat treatment on degradation of aflatoxins in dried figs. *Journal of Food Processing and Preservation*, 33, 329–339.
- Karaca, H., & Velioglu, Y. S. (2007). Ozone applications in fruit and vegetable processing. *Food Reviews International*, 23(1), 91–106. <https://doi.org/10.1080/87559120600998221>
- Karaca, H., Velioglu, Y. S., & Nas, S. (2010). Mycotoxins: Contamination of dried fruits and degradation by ozone. *Toxin Reviews*, 29(2), 51–59. <https://doi.org/10.3109/15569543.2010.485714>
- Kazi, M., Parlapani, F. F., Boziaris, I. S., Vellios, E. K., & Lykas, C. (2018). Effect of ozone on the microbiological status of five dried aromatic plants. *Journal of the Science of Food and Agriculture*, 98(4), 1369–1373.
- Keener, K. M., & Misra, N. N. (2016). Future of cold plasma in food processing. In N. N. Misra, O. Schlüter, & P. J. Cullen (Eds.), *Cold plasma in food and agriculture* (pp. 343–360). Cambridge, MA: Academic Press. <https://doi.org/10.1016/B978-0-12-801365-6.00014-7>
- Keith, L. H., & Walker, M. (1992). *EPA's pesticide fact sheet database*. Boca Raton, FL: CRC Press.
- Kells, S. A., Mason, L. J., Maier, D. E., & Woloshuk, C. P. (2001). Efficacy and fumigation characteristics of ozone in stored maize. *Journal of Stored Products Research*, 37(4), 371–382. [https://doi.org/10.1016/S0022-474X\(00\)00040-0](https://doi.org/10.1016/S0022-474X(00)00040-0)
- Khadre, M. A., & Yousef, A. E. (2001). Sporocidal action of ozone and hydrogen peroxide: A comparative study. *International Journal of Food Microbiology*, 71(2–3), 131–138. [https://doi.org/10.1016/S0168-1605\(01\)00561-X](https://doi.org/10.1016/S0168-1605(01)00561-X)
- Khan, F. A., & Zahoor, M. (2014). In vivo detoxification of aflatoxin B1 by magnetic carbon nanostructures prepared from bagasse. *BMC Veterinary Research*, 10(1), 1–14. <https://doi.org/10.1186/s12917-014-0255-y>
- Kim, J. G., Yousef, A. E., & Khadre, M. A. (2003). Ozone and its current and future application in the food industry. *Advances in Food and Nutrition Research*, 45, 167–218.
- Kim, J. G., Yousef, A. E., & Dave, S. (1999). Application of ozone for enhancing the microbiological safety and quality of foods: A review. *Journal of Food Protection*, 62(9), 1071–1087. <https://doi.org/10.4315/0362-028X-62.9.1071>
- Kluczkowski, A. M. (2019). Fungal and mycotoxin problems in the nut industry. *Current Opinion in Food Science*, 29, 56–63.
- Kumar, P., Mahato, D. K., Kamle, M., Mohanta, T. K., & Kang, S. G. (2017). Aflatoxins: A global concern for food safety, human health and their management. *Frontiers in Microbiology*, 7, 2170. <https://doi.org/10.3389/fmicb.2016.02170>
- Lapalika, G. V., Taylor, M. C., Warden, A. C., Scott, C., Russell, R. J., & Oakshott, J. G. (2012). F420H2-dependent degradation of aflatoxin and other furanocoumarins is widespread throughout the *Actinomycetales*. *PLoS One*, 7(2), e30114. <https://doi.org/10.1371/journal.pone.0030114>
- Laureth, J. C. U., Christ, D., Ganascini, D., & Coelho, S. R. M. (2019). Effect of ozone application on the fungal count and lipid quality of peanut grains. *Journal of Agricultural Science*, 11(5), 271–280.
- Li, H., Xiong, Z., Gui, D., Pan, Y., Xu, M., Guo, Y., ... Li, X. (2019). Effect of ozonation and UV irradiation on aflatoxin degradation of peanuts. *Journal of Food Processing and Preservation*, 43(4), e13914. <https://doi.org/10.1111/jfpp.13914>
- Li, M. M., Guan, E. Q., & Bian, K. (2015). Effect of ozone treatment on deoxynivalenol and quality evaluation of ozonised wheat. *Food Additives and Contaminants - Part A*, 32(4), 544–553. <https://doi.org/10.1080/19440049.2014.976596>
- Li, M., Guan, E., & Bian, K. (2019). Structure elucidation and toxicity analysis of the degradation products of deoxynivalenol by gaseous ozone. *Toxins*, 11(8), 474.
- Li, X. Z., Zhu, C., de Lange, C. F. M., Zhou, T., He, J., Yu, H., ... Young, J. C. (2011). Efficacy of detoxification of deoxynivalenol-contaminated corn by *Bacillus* sp. LS100 in reducing the adverse effects of the mycotoxin on swine growth performance. *Food Additives & Contaminants: Part A*, 28(7), 894–901. <https://doi.org/10.1080/19440049.2011.576402>
- Loeb, B. L. (2018). Forty years of advances in ozone technology. A review of Ozone: Science & Engineering. *Ozone: Science and Engineering*, 40(1), 3–20. <https://doi.org/10.1080/01919512.2017.1383129>
- Luo, X., Li, K., Xing, J., Qi, L., Yang, M., Wang, R., ... Chen, Z. (2018). In vivo toxicity assessment of aflatoxin B1-contaminated corn after ozone degradation. *Food Additives & Contaminants: Part A*, 35(2), 341–350. <https://doi.org/10.1080/19440049.2017.1395518>
- Luo, X., Wang, R., Wang, L., Li, Y., Bian, Y., & Chen, Z. (2014a). Effect of ozone treatment on aflatoxin B1 and safety evaluation of ozonized corn. *Food Control*, 37, 171–176. <https://doi.org/10.1016/j.foodcont.2013.09.043>
- Luo, X., Wang, R., Wang, L., Li, Y., Wang, Y., & Chen, Z. (2014b). Detoxification of aflatoxin in corn flour by ozone. *Journal of the Science of Food and Agriculture*, 94(11), 2253–2258. <https://doi.org/10.1002/jsfa.6550>
- Luo, X., Wang, R., Wang, L., Li, Y., Zheng, R., Sun, X., ... Tao, G. (2014c). Analyses by UPLC Q-TOF MS of products of aflatoxin B1 after ozone treatment. *Food Additives & Contaminants: Part A*, 31(1), 105–110. <https://doi.org/10.1080/19440049.2013.853323>
- Luo, X., Wang, R., Wang, L., Wang, Y., & Chen, Z. (2013). Structure elucidation and toxicity analyses of the degradation products of aflatoxin B1 by aqueous ozone. *Food Control*, 31(2), 331–336. <https://doi.org/10.1016/j.foodcont.2012.10.030>
- Luo, Y., Liu, X., & Li, J. (2018). Updating techniques on controlling mycotoxins - A review. *Food Control*, 89, 123–132. <https://doi.org/10.1016/j.foodcont.2018.01.016>
- MAEBA, H. I., TAKAMOTO, Y. U., Kamimura, M. I., & Miura, T. O. (1988). Destruction and detoxification of aflatoxins with ozone. *Journal of Food Science*, 53(2), 667–668.
- Marasas, W. F. O. (1997). Risk assessment of fumonisins produced by *Fusarium moniliforme* in corn. *Cereal Research Communications*, 25, 399–406.
- McDonough, M. X., Campabadal, C. A., Mason, L. J., Maier, D. E., Denvir, A., & Woloshuk, C. (2011). Ozone application in a modified screw conveyor to treat grain for insect pests, fungal contaminants, and mycotoxins. *Journal of Stored Products Research*, 47(3), 249–254. <https://doi.org/10.1016/j.jspr.2011.04.001>

- McKenzie, K. S., Kubena, L. F., Denvir, A. J., Rogers, T. D., Hitchens, G. D., Bailey, R. H., ... Phillips, T. D. (1998). Aflatoxicosis in turkey poult is prevented by treatment of naturally contaminated corn with ozone generated by electrolysis. *Poultry Science*, 77(8), 1094–1102. <https://doi.org/10.1093/ps/77.8.1094>
- McKenzie, K. S., Sarr, A. B., Mayura, K., Bailey, R. H., Miller, D. R., Rogers, T. D., ... Phillips, T. D. (1997). Oxidative degradation and detoxification of mycotoxins using a novel source of ozone. *Food and Chemical Toxicology*, 35(8), 807–820.
- Méndez-Albores, A., Del Río-García, J. C., & Moreno-Martínez, E. (2007). Decontamination of aflatoxin duckling feed with aqueous citric acid treatment. *Animal Feed Science and Technology*, 135(3), 249–262.
- Milicevic, D., Nesic, K., & Jaksic, S. (2015). Mycotoxin contamination of the food supply chain - Implications for one health programme. *Procedia Food Science*, 5, 187–190. <https://doi.org/10.1016/j.profoo.2015.09.053>
- Moake, M. M., Padilla-Zakour, O. I., & Worobo, R. W. (2005). Comprehensive review of patulin control methods in foods. *Comprehensive Reviews in Food Science and Food Safety*, 4(1), 8–21.
- Mohammadi Kouchesfahani, M., Alimohammadi, M., Jahed Khaniki, G., Nabizadeh Nodehi, R., Aghamohseni, Z., Moazeni, M., & Rezaie, S. (2015). Antifungal effects of ozonated water on aspergillus parasiticus: A new approach to prevent wheat contamination. *Journal of Food Safety*, 35(3), 295–302. <https://doi.org/10.1111/jfs.12159>
- Moretti, A., & Susca, A. (Eds.). (2017). *Mycotoxigenic fungi: Methods and protocols*. Totowa, NJ: Humana Press.
- Moretti, A., Logrieco, A. F., & Susca, A. (2017). Mycotoxins: An underhand food problem. In A. Moretti & A. Susca (Eds.), *Mycotoxigenic fungi* (pp. 3–12). New York, NY: Humana Press.
- Munkvold, G. P. (2017). Fusarium species and their associated mycotoxins. In A. Moretti & A. Susca (Eds.), *Mycotoxigenic fungi* (pp. 51–106). New York, NY: Humana Press.
- Mylona, K. (2012). *Fusarium species in grains: Dry matter losses, mycotoxin contamination and control strategies using ozone and chemical compounds* (PhD thesis). Cranfield University, Cranfield, UK.
- Mylona, K., Kogkaki, E., Sulyok, M., & Magan, N. (2014). Efficacy of gaseous ozone treatment on spore germination, growth and fumonisin production by *Fusarium verticillioides* in vitro and in situ in maize. *Journal of Stored Products Research*, 59, 178–184. <https://doi.org/10.1016/j.jspr.2014.08.001>
- Neme, K., & Mohammed, A. (2017). Mycotoxin occurrence in grains and the role of postharvest management as a mitigation strategies. A review. *Food Control*, 78, 412–425.
- Niculina, N. G., Otilia, C., Veronica, S., Claudia, D. C., & Titus, S. (2019). Research regarding the relationship among the pests *Ostrinia nubilalis*, *Helicoverpa armigera* and the fungi *Fusarium verticillioides*, *Aspergillus flavus* in corn in the climatic conditions from Lovrin (Timiş County). *Research Journal of Agricultural Science*, 51(4), 282–291.
- Niemira, B. A. (2012). Cold plasma reduction of Salmonella and Escherichia coli O157: H7 on almonds using ambient pressure gases. *Journal of Food Science*, 77(3), M171–M175. <https://doi.org/10.1111/j.1750-3841.2011.02594.x>
- Nyandieka, H. S., Maina, J. O., & Nyamwange, C. (2009). Destruction of aflatoxins in contaminated maize samples using ammoniation procedures. *East and Central African Journal of Pharmaceutical Sciences*, 12(3), 47–51.
- Ogunade, I. M., Martínez-Tupia, C., Queiroz, O. C. M., Jiang, Y., Drouin, P., Wu, F., ... Adesogan, A. T. (2018). Silage review: Mycotoxins in silage: Occurrence, effects, prevention, and mitigation. *Journal of Dairy Science*, 101(5), 4034–4059.
- Oner, M. E., & Demirci, A. (2016). Ozone for food decontamination: Theory and applications. In H. Lelieveld, J. Holah, & D. Gabrić (Eds.), *Handbook of hygiene control in the food industry* (2nd ed., pp. 491–501). Sawston, UK: Woodhead Publishing. <https://doi.org/10.1016/b978-0-08-100155-4.00033-9>
- Öztekin, S., Zorlugenç, B., & Zorlugenç, F. K. (2006). Effects of ozone treatment on microflora of dried figs. *Journal of Food Engineering*, 75(3), 396–399. <https://doi.org/10.1016/j.jfoodeng.2005.04.024>
- Palou, L., Smilanick, J. L., Crisosto, C. H., & Mansour, M. (2001). Effect of gaseous ozone exposure on the development of green and blue molds on cold stored citrus fruit. *Plant Disease*, 85(6), 632–638. <https://doi.org/10.1094/PDIS.2001.85.6.632>
- Palumbo, J. D., Mahoney, N. E., Light, D. M., Siegel, J., Puckett, R. D., & Michailides, T. J. (2014). Spread of *Aspergillus flavus* by novel orangeworm (*Amyelois transitella*) on almond. *Plant Disease*, 98(9), 1194–1199.
- Pandiselvam, R., Subhashini, S., Banuu Priya, E. P., Kothakota, A., Ramesh, S. V., & Shahir, S. (2019). Ozone based food preservation: A promising green technology for enhanced food safety. *Ozone: Science & Engineering*, 41(1), 17–34.
- Pandiselvam, R., Sunoj, S., Manikantan, M. R., Kothakota, A., & Hebbbar, K. B. (2017). Application and kinetics of ozone in food preservation. *Ozone: Science and Engineering*, 39(2), 115–126. <https://doi.org/10.1080/01919512.2016.1268947>
- Pankaj, S. K., Shi, H., & Keener, K. M. (2018). A review of novel physical and chemical decontamination technologies for aflatoxin in food. *Trends in Food Science & Technology*, 71, 73–83. <https://doi.org/10.1016/j.tifs.2017.11.007>
- Pankaj, S. K., Wan, Z., & Keener, K. M. (2018). Effects of cold plasma on food quality: A review. *Foods*, 7(1), 4. <https://doi.org/10.3390/foods7010004>
- Patras, A., Julakanti, S., Yannam, S., Bansode, R. R., Burns, M., & Vergne, M. J. (2017). Effect of UV irradiation on aflatoxin reduction: A cytotoxicity evaluation study using human hepatoma cell line. *Mycotoxin Research*, 33(4), 343–350.
- Pascari, X., Ramos, A. J., Marin, S., & Sanchis, V. (2018). Mycotoxins and beer. Impact of beer production process on mycotoxin contamination. A review. *Food Research International*, 103, 121–129.
- Peng, W. X., Marchal, J. L. M., & Van der Poel, A. F. B. (2018). Strategies to prevent and reduce mycotoxins for compound feed manufacturing. *Animal Feed Science and Technology*, 237, 129–153.
- Peng, Z., Chen, L., Zhu, Y., Huang, Y., Hu, X., Wu, Q., ... Yang, W. (2018). Current major degradation methods for aflatoxins: A review. *Trends in Food Science & Technology*, 80, 155–166. <https://doi.org/10.1016/j.tifs.2018.08.009>
- Pereira, A. D. M., Faroni, L. R. A., Sousa, A. D., Urruchi, W. I., & Paes, J. L. (2008). Influence of the grain temperature on the ozone toxicity to *Tribolium castaneum*. *Revista Brasileira de Engenharia Agrícola e Ambiental*, 12(5), 493–497.
- Perrone, G., & Susca, A. (2017). Penicillium species and their associated mycotoxins. In A. Moretti & A. Susca (Eds.), *Mycotoxigenic fungi* (pp. 107–119). New York, NY: Humana Press.
- Pfohl-Leschkowicz, A., & Manderville, R. A. (2007). Ochratoxin A: An overview on toxicity and carcinogenicity in animals and humans.

- Molecular Nutrition and Food Research*, 51(1), 61–99. <https://doi.org/10.1002/mnfr.200600137>
- Phillips, T. D., Sarr, A. B., & Grant, P. G. (1995). Selective chemisorption and detoxification of aflatoxins by phyllosilicate clay. *Natural Toxins*, 3(4), 204–213.
- Piacentini, K. C., Savi, G. D., & Scussel, V. M. (2017). The effect of ozone treatment on species of *Fusarium* growth in malting barley (*Hordeum vulgare* L.) grains. *Quality Assurance and Safety of Crops and Foods*, 9(4), 383–389.
- Piechowiak, T., Józefczyk, R., & Balawejder, M. (2018). Impact of ozonation process of wheat flour on the activity of selected enzymes. *Journal of Cereal Science*, 84, 30–37.
- Piemontese, L., Messina, M. C., Marconi, E., Falasca, L., Zivoli, R., Gambacorta, L., ... Solfrizzo, M. (2018). Effect of gaseous ozone treatments on DON, microbial contaminants and technological parameters of wheat and semolina. *Food Additives & Contaminants: Part A*, 35(4), 761–772.
- Pineiro, M. (2001). *Manual on the application of the HACCP system in mycotoxin prevention and control* (No. 73). Rome, Italy: Food & Agriculture Organization.
- Porto, Y. D., Trombete, F. M., Freitas-Silva, O., De Castro, I. M., Direito, G. M., & Ascheri, J. L. R. (2019). Gaseous ozonation to reduce aflatoxins levels and microbial contamination in corn grits. *Microorganisms*, 7(8), 220.
- Proctor, A. D., Ahmedna, M., Kumar, J. V., & Goktepe, I. (2004). Degradation of aflatoxins in peanut kernels/flour by gaseous ozonation and mild heat treatment. *Food Additives and Contaminants*, 21(8), 786–793. <https://doi.org/10.1080/02652030410001713898>
- Prudente, A. P., Jr., & King, J. M. (2002). Efficacy and safety evaluation of ozonation to degrade aflatoxin in corn. *Journal of Food Science*, 67(8), 2866–2872.
- Puzyr', A. P., Burov, A. E., Bondar', V. S., & Trusov, Y. N. (2010). Neutralization of aflatoxin B1 by ozone treatment and adsorption by nanodiamonds. *Nanotechnologies in Russia*, 5(1–2), 137–141. <https://doi.org/10.1134/s1995078010010143>
- Qi, L., Li, Y., Luo, X., Wang, R., Zheng, R., Wang, L., ... Chen, Z. (2016). Detoxification of zearalenone and ochratoxin A by ozone and quality evaluation of ozonised corn. *Food Additives and Contaminants - Part A*, 33(11), 1700–1710.
- Raila, A., Lugauskas, A., Steponavicius, D., Railiene, M., Steponaviciene, A., & Zvicevicius, E. (2006). Application of ozone for reduction of mycological infection in wheat grain. *Annals of Agricultural and Environmental Medicine*, 13(2), 287–294.
- Rakness, K. L., DeMers, L. D., & Blank, B. D. (1996). Ozone system fundamentals for drinking water treatment. *Opflow*, 22(7), 1–5.
- Raters, M., & Matissek, R. (2008). Thermal stability of aflatoxin B1 and ochratoxin A. *Mycotoxin Research*, 24(3), 130–134.
- Rayner, E. T., Dwarakanath, C. T., Mann, G. E., & Doller, F. G. (1971). *US Patent No. 3,592,641*. Washington, DC: U.S. Patent and Trademark Office. <https://doi.org/10.1111/j.1559-3584.1927.tb04229.x>
- Ren, D., Diao, E., Hou, H., Xie, P., Mao, R., Dong, H., & Qian, S. (2019). Cytotoxicity of deoxynivalenol after being exposed to gaseous ozone. *Toxins*, 11(11), 639.
- Repečienė, J., Levinskaitė, L., Paškevičius, A., & Raudonienė, V. (2013). Toxin-producing fungi on feed grains and application of yeasts for their detoxification. *Polish Journal of Veterinary Sciences*, 16(2), 391–393. <https://doi.org/10.2478/pjvs-2013-0054>
- Rice, R. G., & Graham, D. M. (2001). US FDA regulatory approval of ozone as an antimicrobial agent—what is allowed and what needs to be understood. *Ozone News*, 29(5), 22–31.
- Rice, R. G., Graham, D. M., & Lowe, M. T. (2002). Recent ozone applications in food processing and sanitation. *Food Safety Magazine*, 8(5), 10–17.
- Riley, R. T., & Norred, W. P. (1999). Mycotoxin prevention and decontamination: A case study on maize. *Food, Nutrition and Agriculture*, (23), 25–32.
- Savi, G. D., Bittencourt, K. O., Stein, S. M., Santos, K., Martins, C., & Scussel, V. M. (2014). *Ozone treatment efficiency on toxigenic fungi and mycotoxins decontamination from postharvest wheat (Triticum aestivum L.) grains*. 11th International Working Conference on Stored Product Protection.
- Savi, G. D., Piacentini, K. C., Bittencourt, K. O., & Scussel, V. M. (2014). Ozone treatment efficiency on *Fusarium* graminearum and deoxynivalenol degradation and its effects on whole wheat grains (*Triticum aestivum* L.) quality and germination. *Journal of Stored Products Research*, 59, 245–253.
- Savi, G. D., Piacentini, K. C., & Scussel, V. M. (2015). Ozone treatment efficiency in *Aspergillus* and *Penicillium* growth inhibition and mycotoxin degradation of stored wheat grains (*Triticum aestivum* L.). *Journal of Food Processing and Preservation*, 39(6), 940–948. <https://doi.org/10.1111/jfpp.12307>
- Savi, G. D., & Scussel, V. M. (2014). Effects of ozone gas exposure on toxigenic fungi species from *Fusarium*, *Aspergillus*, and *Penicillium* genera. *Ozone: Science and Engineering*, 36(2), 144–152. <https://doi.org/10.1080/01919512.2013.846824>
- Schatzki, T. F., & Ong, M. S. (2001). Dependence of aflatoxin in almonds on the type and amount of insect damage. *Journal of Agricultural and Food Chemistry*, 49(9), 4513–4519.
- Scott, M., & Somers, E. (1968). Stability of patulin and penicillic acid in fruit juices and flour. *Journal of Agricultural and Food Chemistry*, 16(3), 483–485.
- Sen, Y., Onal-Ulusoy, B., & Mutlu, M. (2019). Detoxification of hazelnuts by different cold plasmas and gamma irradiation treatments. *Innovative Food Science & Emerging Technologies*, 54, 252–259.
- Shanakhat, H., Sorrentino, A., Raiola, A., Reverberi, M., Salustri, M., Masi, P., & Cavella, S. (2019). Technological properties of durum wheat semolina treated by heating and UV irradiation for reduction of mycotoxin content. *Journal of Food Process Engineering*, 42(3), e13006.
- Shi, H., Li, S., Bai, Y., Prates, L. L., Lei, Y., & Yu, P. (2018). Mycotoxin contamination of food and feed in China: Occurrence, detection techniques, toxicological effects and advances in mitigation technologies. *Food Control*, 91, 202–215.
- Silva, J., Pereira, M. N., & Scussel, V. M. (2018). Ozone gas antifungal effect on extruded dog food contaminated with *Aspergillus flavus*. *Ozone: Science & Engineering*, 40(6), 487–493.
- Smilanick, J. L. (2003, December). *Use of ozone in storage and packing facilities*. Washington Tree Fruit Postharvest Conference Wenatche, Washington, pp. 1–10.
- Sousa, A. D., Faroni, L. D. A., Guedes, R. N. C., Tótola, M. R., & Urruchi, W. I. (2008). Ozone as a management alternative against phosphine-resistant insect pests of stored products. *Journal of Stored Products Research*, 44(4), 379–385.
- Sun, C., Ji, J., Wu, S., Sun, C., Pi, F., Zhang, Y., ... Sun, X. (2016). Saturated aqueous ozone degradation of deoxynivalenol and its application in contaminated grains. *Food Control*, 69, 185–190.

- Suian Jose, G., Raquel Bechlin, T., Werncke, I., & Christ, D. (2018). Ozone penetration in a column containing sunn hemp (*Crotalaria spectabilis* Roth) seeds and the effect on the quality. *Poljoprivredna Tehnika*, 43, 1–10. <https://doi.org/10.5937/PoljTeh1803001S>
- Tabata, S., Kamimura, H., Ibe, A., Hashimoto, H., & Tamura, Y. (1994). Degradation of aflatoxins by food additives. *Journal of Food Protection*, 57(1), 42–47.
- Tejada-Castañeda, Z. I., Ávila-Gonzalez, E., Casaubon-Huguenin, M. T., Cervantes-Olivares, R. A., Vásquez-Peláez, C., Hernández-Baumgarten, E. M., & Moreno-Martínez, E. (2008). Biodetoxification of aflatoxin-contaminated chick feed. *Poultry Science*, 87(8), 1569–1576. <https://doi.org/10.3382/ps.2007-00304>
- Tiwari, B. K., Brennan, C. S., Curran, T., Gallagher, E., Cullen, P. J., & O'Donnell, C. P. (2010). Application of ozone in grain processing. *Journal of Cereal Science*, 51(3), 248–255. <https://doi.org/10.1016/j.jcs.2010.01.007>
- Tiwari, B. K., & Rice, R. G. (2012). Regulatory and legislative issues. In C. O'Donnell, B. K. Tiwari, P. J. Cullen, & R. G. Rice (Eds.), *Ozone in food processing* (pp. 7–17). Hoboken, NJ: Blackwell Publishing.
- Tola, M., & Kebede, B. (2016). Occurrence, importance and control of mycotoxins: A review. *Cogent Food & Agriculture*, 2(1), 1191103. <https://doi.org/10.1080/23311932.2016.1191103>
- Topcu, A., Bulat, T., Wishah, R., & Boyacı, I. H. (2010). Detoxification of aflatoxin B 1 and patulin by *Enterococcus faecium* strains. *International Journal of Food Microbiology*, 139(3), 202–205.
- Torlak, E. (2019). Use of gaseous ozone for reduction of ochratoxin A and fungal populations on sultanas. *Australian Journal of Grape and Wine Research*, 25(1), 25–29.
- Torres, A. M., Palacios, S. A., Yerkovich, N., Palazzini, J. M., Battilani, P., Leslie, J. F., ... Chulze, S. N. (2019). Fusarium head blight and mycotoxins in wheat: Prevention and control strategies across the food chain. *World Mycotoxin Journal*, 12(4), 333–355.
- Trombete, F. M., Freitas-Silva, O., Saldanha, T., Venâncio, A. A., & Fraga, M. E. (2016). Ozone against mycotoxins and pesticide residues in food: Current applications and perspectives. *International Food Research Journal*, 23(6), 2545–2556.
- Trombete, F. M., Porto, Y. D., Freitas-Silva, O., Pereira, R. V., Direito, G. M., Saldanha, T., & Fraga, M. E. (2017). Efficacy of ozone treatment on mycotoxins and fungal reduction in artificially contaminated soft wheat grains. *Journal of Food Processing and Preservation*, 41(3), e12927.
- Udomkun, P., Wiredu, A. N., Nagle, M., Müller, J., Vanlauwe, B., & Bandyopadhyay, R. (2017). Innovative technologies to manage aflatoxins in foods and feeds and the profitability of application – A review. *Food Control*, 76, 127–138. <https://doi.org/10.1016/j.foodcont.2017.01.008>
- Ueno, Y. (1983). *Trichothecenes: Chemical, biological, and toxicological aspects* (Vol. 4). Amsterdam, the Netherlands: Elsevier Science.
- Uzun, H., & Ibanoglu, E. (2018). Oxidation kinetics of hazelnut oil treated with ozone. *Grasas y Aceites*, 68(4), e222.
- Van der Stegen, G. H., Essens, P. J., & Van der Lijn, J. (2001). Effect of roasting conditions on reduction of ochratoxin A in coffee. *Journal of Agricultural and Food Chemistry*, 49(10), 4713–4715.
- Varga, J., Baranyi, N., Chandrasekaran, M., Vágvolgyi, C., & Kocsu, S. (2015). Mycotoxin producers in the *Aspergillus* genus: An update. *Acta Biologica Szegediensis*, 59(2), 151–167.
- Varga, J., Rigó, K., Téren, J., & Mesterházy, Á. (2001). Recent advances in ochratoxin research: II. Biosynthesis, mode of action and control of ochratoxins. *Cereal Research Communications*, 29, 93–100.
- Varga, J., Rigó, K., & Téren, J. (2000). Degradation of ochratoxin A by *Aspergillus* species. *International Journal of Food Microbiology*, 59(1–2), 1–7. [https://doi.org/10.1016/S0168-1605\(00\)00230-0](https://doi.org/10.1016/S0168-1605(00)00230-0)
- Vijayanandraj, V. R., Nagendra Prasad, D., Mohan, N., & Gunasekaran, M. (2006). Effect of ozone on *Aspergillus niger* causing black rot disease in onion. *Ozone: Science and Engineering*, 28(5), 347–350. <https://doi.org/10.1080/01919510600900035>
- Vita, D. S., Rosa, P., Giuseppe, A., & Apparecchiature, C. G. (2014). Effect of Gamma irradiation on aflatoxins and ochratoxin A reduction in almond samples. *Journal of Food Research*, 3(4), 113. <https://doi.org/10.5539/jfr.v3n4p113>
- Wang, L., Wang, Y., Shao, H., Luo, X., Wang, R., Li, Y., ... Chen, Z. (2017). In vivo toxicity assessment of deoxynivalenol-contaminated wheat after ozone degradation. *Food Additives & Contaminants: Part A*, 34(1), 103–112.
- Wang, L., Shao, H., Luo, X., Wang, R., Li, Y., Li, Y., ... Chen, Z. (2016). Effect of ozone treatment on deoxynivalenol and wheat quality. *PLoS ONE*, 11(1), 1–13. <https://doi.org/10.1371/journal.pone.0147613>
- Wang, L., Luo, Y., Luo, X., Wang, R., Li, Y., Li, Y., ... Chen, Z. (2016). Effect of deoxynivalenol detoxification by ozone treatment in wheat grains. *Food Control*, 66, 137–144. <https://doi.org/10.1016/j.foodcont.2016.01.038>
- Wang, S., Liu, H., Lin, J., & Cao, Y. (2010). Can ozone fumigation effectively reduce aflatoxin B 1 and other mycotoxins contamination on stored grain? *Julius-Kühn-Archiv*. <https://doi.org/10.5073/jka.2010.425.167.172>
- Wang, X., Qin, X., Hao, Z., Luo, H., Yao, B., & Su, X. (2019). Degradation of four major mycotoxins by eight manganese peroxidases in presence of a dicarboxylic acid. *Toxins*, 11(10), 566.
- Whangchai, K., Saengnil, K., & Uthaibutra, J. (2006). Effect of ozone in combination with some organic acids on the control of postharvest decay and pericarp browning of longan fruit. *Crop Protection*, 25(8), 821–825. <https://doi.org/10.1016/j.cropro.2005.11.003>
- White, S. D. (2007). *Using ozone to control fungi in high moisture corn* (Master's thesis). Iowa State University, Ames, IA.
- White, S. D., Murphy, P. T., Bern, C. J., & van Leeuwen, J. H. (2010). Controlling deterioration of high-moisture maize with ozone treatment. *Journal of Stored Products Research*, 46(1), 7–12.
- White, S. D., Murphy, P. T., Leandro, L. F., Bern, C. J., Beattie, S. E., & van Leeuwen, J. H. (2013). Mycoflora of high-moisture maize treated with ozone. *Journal of Stored Products Research*, 55, 84–89. <https://doi.org/10.1016/j.jspr.2013.08.006>
- Widstrom, N. W. (1979). The role of insects and other plant pests in aflatoxin contamination of corn, cotton, and peanuts—A review. *Journal of Environmental Quality*, 8(1), 5–11.
- Womack, E. D., Brown, A. E., & Sparks, D. L. (2014). A recent review of non-biological remediation of aflatoxin-contaminated crops. *Journal of the Science of Food and Agriculture*, 94(9), 1706–1714. <https://doi.org/10.1002/jsfa.6520>
- Wright, V. F. (1992). *Assessment of insect infestation in stored maize and their relationship to Aspergillus flavus contamination*. Mycotoxin Prevention and Control in Food Grains, UNDP/FAO REG-NET and ASEAN Grain Postharvest Programme, Bangkok, pp. 110–116.
- Wu, J., Doan, H., & Cuenca, M. A. (2006). Investigation of gaseous ozone as an antifungal fumigant for stored wheat. *Journal of Chemical Technology & Biotechnology*, 81(7), 1288–1293. <https://doi.org/10.1002/jctb>

- Xu, L. (1999). Use of ozone to improve the safety of fresh fruits and vegetables. *Food Technology*, 53(10), 58–62.
- Xu, Y., Ji, J., Wu, H., Pi, F., Blaženović, I., Zhang, Y., & Sun, X. (2019). Untargeted GC-TOFMS-based cellular metabolism analysis to evaluate ozone degradation effect of deoxynivalenol. *Toxicon*, 168, 49–57.
- Xu, Y., Wang, Y., Ji, J., Wu, H., Pi, F., Zhang, Y., & Sun, X. (2019). Chemical and toxicological alterations of zearalenone under ozone treatment. *Food Additives and Contaminants - Part A*, 36(1), 163–174. <https://doi.org/10.1080/19440049.2018.1547425>
- Young, J. C. (1986). Reduction in levels of deoxynivalenol in contaminated corn by chemical and physical treatment. *Journal of Agricultural and Food Chemistry*, 34(3), 465–467.
- Young, J. C., Subryan, L. M., Potts, D., McLaren, M. E., & Gobran, F. H. (1986). Reduction in levels of deoxynivalenol in contaminated wheat by chemical and physical treatment. *Journal of Agricultural and Food Chemistry*, 34(3), 461–465.
- Young, J. C., Zhu, H., & Zhou, T. (2006). Degradation of trichothecene mycotoxins by aqueous ozone. *Food and Chemical Toxicology*, 44(3), 417–424. <https://doi.org/10.1016/j.fct.2005.08.015>
- Zhou, Y., Wu, S., Wang, F., Li, Q., He, C., Duan, N., & Wang, Z. (2020). Assessing the toxicity in vitro of degradation products from deoxynivalenol photocatalytic degradation by using upconversion nanoparticles@ TiO₂ composite. *Chemosphere*, 238, 124648.
- Zhu, F. (2018). Effect of ozone treatment on the quality of grain products. *Food Chemistry*, 264, 358–366. <https://doi.org/10.1016/j.foodchem.2018.05.047>
- Zhu, R., Feussner, K., Wu, T., Yan, F., Karlovsky, P., & Zheng, X. (2015). Detoxification of mycotoxin patulin by the yeast *Rhodosporidium paludigenum*. *Food Chemistry*, 179, 1–5. <https://doi.org/10.1016/j.foodchem.2015.01.066>
- Zorlugenç, B., Kiroğlu Zorlugenç, F., Öztekin, S., & Evliya, I. B. (2008). The influence of gaseous ozone and ozonated water on microbial flora and degradation of aflatoxin B1 in dried figs. *Food and Chemical Toxicology*, 46(12), 3593–3597. <https://doi.org/10.1016/j.fct.2008.09.003>

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