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## Review

## Application of ozone in grain processing

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## ABSTRACT

Ozone is an effective fumigant for insect killing, mycotoxin destruction and microbial inactivation which has a minimal or no effect on grain quality. Studies have demonstrated that ozone which is a natural agent, may offer unique advantages for grain processing along with addressing growing concerns over the use of harmful pesticides. This paper focuses on the efficacy of ozone for preservation of food grains and discusses the possible effects on product quality. Inactivation mechanisms for micro-organisms and mycotoxins are detailed. Critical intrinsic and extrinsic factors governing inactivation are discussed along with potential grain applications.

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## 1. Introduction

Ozone finds wide application as a powerful disinfectant in water treatment, food processing and preservation and various other environmental applications. Ozone as an oxidant has numerous potential applications in the food industry because of its advantages over traditional food preservation techniques. Application of ozone either in gaseous or liquid form in fruit and vegetable processing is often employed for inactivation of pathogen and spoilage micro-organisms (Cullen et al., 2009). Apart from the wide spectrum of microbial inactivation, ozone also has the potential to kill storage pests and degrade mycotoxins. One of the potential advantages of ozone is that excess ozone auto-decomposes rapidly to produce oxygen and thus leaves no residues in food. Its efficacy against a wide range of micro-organisms including bacteria, fungi, viruses, protozoa, and bacterial fungal spores has been reported (Cullen et al., 2009; Khadre et al., 2001; Restaino et al., 1995). Such advantages make ozone attractive to the food industry and consequently it has been affirmed as Generally Recognized as Safe (GRAS) for use in food processing (Graham, 1997).

The possible application of ozone in food grain preservation would address the growing concern over the use of harmful

pesticides to kill storage pests. The Montreal Protocol on substances that deplete the ozone layer (Fields and White, 2002), increased insect resistance, and increased consumer demand for chemical free grains has led grain processors to seek alternatives to control storage pests. Currently, commonly used pesticides (fumigants) for grain storage include aluminium phosphide, methyl bromide and phosphine. Among these, the use of methyl bromide is nearly phased out as agreed in the *Montreal Protocol*. The persistent use of these pesticides has been reported to disrupt biological control systems by natural agents, leading to outbreaks of insect pests, widespread development of resistance, undesirable effects on non-target organisms, and environmental and human health concerns (Collins et al., 2005; Islam et al., 2009; Kells et al., 2001; Pimentel et al., 2009, 2007). The increasing concern about their adverse effects has highlighted the need for the development of selective insect-control alternatives (Fields and White, 2002). Use of ozone as food grain fumigant is a viable alternative from both environmental and economical perspectives. For example, studies by a number of authors (Islam et al., 2009; Pereira et al., 2008a,b; Pimentel et al., 2009) show the economical viability of using ozone to fumigate stored maize based on an economic analysis of maize stored up to 6 months at 20 °C. This paper reviews the efficacy of ozone for the storage and preservation of food grains, the effect of ozonation on product quality and the current status of ozone application in grain processing.

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## 2. Industrial production of ozone

The precursor for industrial production of ozone is that  $O_2$  is abundant and inexhaustible. Ozone ( $O_3$ ) results from the rearrangement of atoms when oxygen molecules are subjected to high-voltage electric discharge. The product is a bluish gas with pungent odour and strong oxidizing properties. Ozone is generated by the reaction of free oxygen radicals with diatomic oxygen to form triatomic oxygen molecules. Generation of the free oxygen radical occurs by breakage of strong O–O bonds, requiring a significant energy input. UV radiation and corona discharge methods can be used to initiate free radical oxygen formation and thereby generate ozone. In addition to photochemical (UV radiation) and electric discharge methods, ozone can be produced by chemical, thermal, chemonuclear and electrolytic methods (Kim et al., 1999). Generally the corona discharge method is used for generation of ozone at a commercial level. Two electrodes, high tension and low tension (ground electrode), separated by a ceramic dielectric medium provide a narrow discharge gap (Mahapatra et al., 2005). Electrons with sufficient kinetic energy (around 6–7 eV) to dissociate the oxygen molecule collide and a molecule of ozone can be formed from each oxygen atom (Güzel-Seydim et al., 2004). If atmospheric air is passed through the generator as the feed gas, 1–3% ozone can be produced; however, using high purity oxygen, up to 16% ozone production can be achieved. Consequently, ozone concentration cannot be increased beyond the point where the rate of formation and destruction are equal. Ozone is produced on site of operation as ozone gas cannot be stored since ozone spontaneously degrades back to oxygen atoms.

## 3. Application of ozone in grain processing

The patented Oxygreen® process was one of the most significant advances for application of ozone in food grains (Dubois et al., 2006, 2008). This process involves a premoistening of grains in a closed batch reactor followed by ozonation. Similarly, Yvin et al. (2001) patented a process to obtain flour with enhanced microbial safety from ozonated grains. Within the food industry, ozone is employed for fresh fruit and vegetable decontamination. However, a limited number of studies have been reported on ozone treatment of cereals and cereal-based products as an alternative to chlorine treatment.

Ozone treatment of grain is generally applied in silos or vessels. Prior to ozone application, it is necessary to characterise the dynamics of ozone movement through the various grain types to optimise ozone generators for use on large commercial storage bins (Shunmugam et al., 2005). Ozone moves through grain slowly because the gas reacts with the chemical constituents present in the outer layer of grain (seed coat). Diffusion of ozone into the grain depends upon the grain characteristics. Movement of ozone within a silo or column filled with grain can be in either of 3 directions (Fig. 1); namely movement in the transverse direction ( $x,z$ ) or movement in the vertical direction ( $y$ ) under the influence of ozone gas velocity ( $v_f$ ) and adsorption of ozone by the grain surface and possible reactions leading to degradation of ozone. Adsorption of ozone and subsequent penetration into the grain depends upon several intrinsic and extrinsic factors ( $k$ ) such as surface characteristics of the grain, microbial contamination, presence of insects and moisture content etc. Penetration and movement of ozone

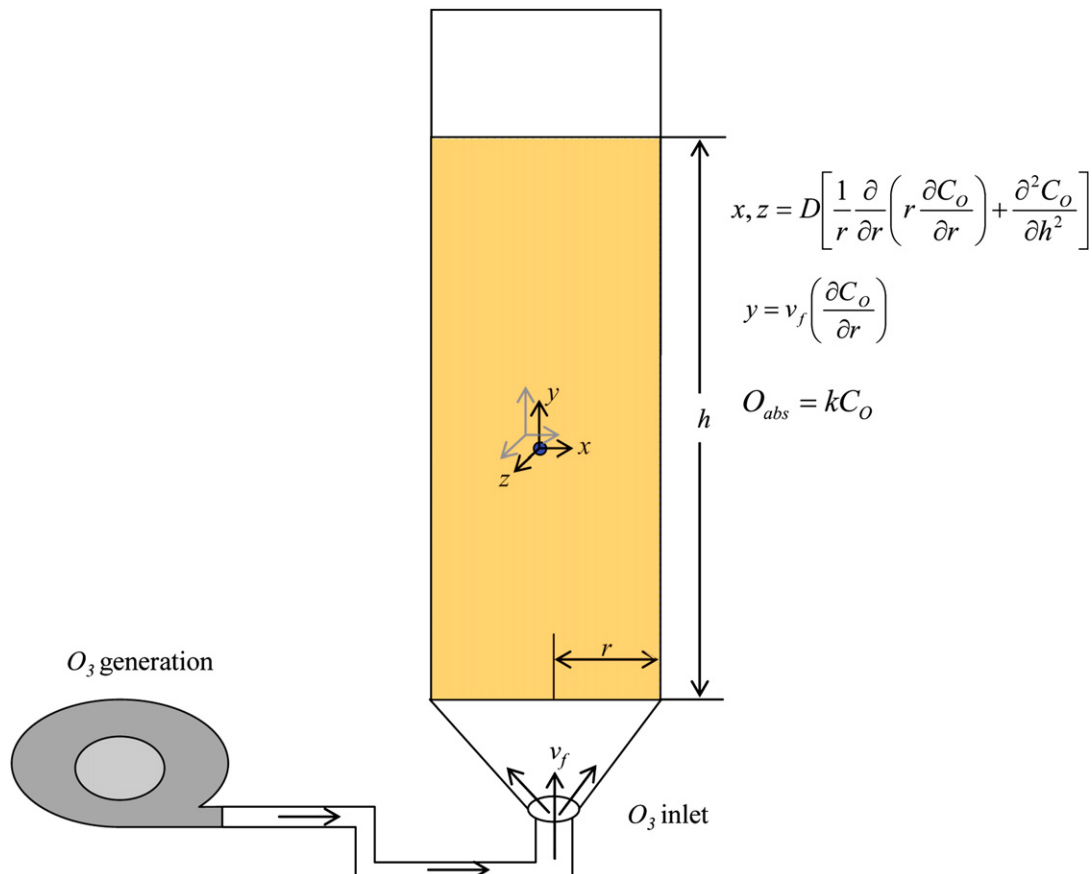


Fig. 1. Schematic diagram showing movement of ozone in a grain column.

within a grain column can be expressed by the differential kinetic – diffusion equation (Eq (1)) (Raila et al., 2006).

$$\frac{\partial C_0}{\partial t} = D \left( \frac{1}{r} \frac{\partial}{\partial r} \left[ r \frac{\partial C_0}{\partial r} \right] + \frac{\partial^2 C_0}{\partial h^2} \right) - v_f \left( \frac{\partial C_0}{\partial h} \right) - k C_0 \quad (1)$$

where:  $C_0$  – ozone concentration;  $D$  – diffusivity;  $r$  – radius of the bottom of the grain mound;  $h$  – grain mound height;  $k$  – factor of ozone absorption;  $v_f$  – air seepage velocity in the grain layer;  $t$  – duration of exposure to ozone.

Ozone movement through the grain layer is restricted by the highly reactive nature of ozone (Mendez et al., 2003). Kells et al. (2001) described the movement of ozone into two distinct phases for maize. The first phase is contact of ozone with grains during which the concentration of ozone reduces as it moves in the  $y$  direction through the grain due to interaction with organic materials present on or in the vicinity of the grain surface rapidly degrading ozone through oxidation reaction. Movement of ozone in phase 1 is restricted due to the ozone demand of the organic matter (Cullen et al., 2009; Kim et al., 1999). The second phase corresponds to free movement of ozone through grain layers once these reactive sites are eliminated (Kells et al., 2001; Mendez et al., 2003).

Ozone adsorption in the grain layer depends on ozone concentration in the feed gas, duration of exposure, gas flow rate, temperature, grain characteristics and the presence of other organic matter such as insects and surface microbial status of the grain. Presence of moisture also plays an important role in ozone reactivity with grain because water solubilizes ozone and increase contact between gas and grain. Raila et al. (2006) observed slower ozone penetration between grain layers with higher mycological contamination.

#### 4. Effects of ozone on the prevalence of storage pests

Grains are frequently stored for periods of up to 36 months at ambient temperatures in bulk silos and are often fumigated to prevent infestation and contamination (Kaushik et al., 2009). Within the grain processing industry, ozone is employed as a replacement for the existing fumigants such as methyl bromide and phosphine for the control of storage pests. Ozone as a fumigant is reported to kill stored-grain insects such as *Tribolium castaneum*, *Rhyzopertha dominica*, *Oryzaephilus surinamensis*, *Sitophilus oryzae*, and *Ephestia elutella* (Sousa et al., 2008). Table 1 lists reported applications of ozone for the control of insects. Laboratory and field studies report the efficacy of ozone in controlling both

phosphine-susceptible and phosphine-resistant strains of *Sitophilus zeamais*, *S. oryzae*, *R. dominica*, and *T. castaneum* (Qin et al., 2003). Ozone toxicity for insects varies depending on the stage within its life cycle. For example, larval and pupal stages of *T. castaneum* are ozone sensitive with sensitivity decreasing with age (Erdman, 1980). Isikber and Oztekin (2009) studied the mortality rate of 2 flour beetles (*Ephestia kuehniella* and *Tribolium confusum*) and observed insect mortality during ozonation was not only dependent upon the life stages specific for both the species but was also insect specific. They observed a higher susceptibility and high mortality for all 3 larvae, pupae and adult stages of *E. kuehniella* (90–100%) compared to *T. confusum* (1.3–22.7%) for similar experimental conditions. Similarly a higher susceptibility rate for insects is reported for Indian meal moth (*Plodia interpunctella*) compared to *T. confusum* (Leesch, 2003). However, Kells et al. (2001) reported high mortality rate for maize weevil, red flour beetle, and the Indian meal moth exposed to ozone (50 ppm for 3 days or 25 ppm for 5 days) (Table 1).

The respiratory system is considered to be the major entry route for toxic gases into an insect body, determining the lethality of a fumigant as the respiratory system of insects consists of highly branched cuticle lined tubes extending throughout the body (Lu et al., 2009). Insects breathe discontinuously to minimize oxidative damage due to oxygen toxicity (Hetz and Bradley, 2005). Ozone causes oxidative tissue damage even at low concentrations (Liu et al., 2007) resulting in DNA strand breaks, alteration of pulmonary function, bronchial responsiveness, membrane oxidation or mutations *in vivo* (Ballinger et al., 2005; Bornholdt et al., 2002). Increased respiration rate with increasing temperatures may result in more gas exchange due to the overall increase in metabolic and respiration rate (Pereira et al., 2008; Rozado et al., 2008). However, Sousa et al. (2008) did not observe any correlation between insect respiration rate and their susceptibility to ozone. Rozado et al. (2008) reported that exposure periods of 23.76 and 64.19 h are required to achieve 95% control ( $LT_{95}$ ) for adults of *S. zeamais* and *T. castaneum* in stored maize grains in modified atmosphere with an ozone injection at 50 mg kg<sup>-1</sup>. This dose did not affect the physiological quality of the grain. However, the lethal time ( $LT_{50}$  and  $LT_{95}$ ) for adult insects decreases with increase in grain mass temperature (Pereira et al., 2008) and with ozone concentration.

#### 5. Microbial decontamination and mycotoxins

Ozone in gaseous or aqueous form is reported to reduce levels of the natural microflora, as well as bacterial, fungal and mould

**Table 1**  
Effects of ozone treatment on storage insects.

Food grain	Insects	Conditions	Mortality rate	Reference
Maize	RFB ( <i>Tribolium castaneum</i> ) MW ( <i>Sitophilus zeamais</i> ) IMM ( <i>Plodia interpunctella</i> )	50 ppm for 3 days	94.5% IMM 100% MW 92.2% RFB	Kells et al. (2001)
Maize	RFB ( <i>T. castaneum</i> ) MW ( <i>Sitophilus zeamais</i> ) IMM ( <i>P. interpunctella</i> )	25 ppm for 5 days	77.0% IMM 99.9% MW 91.4% RFB	Kells et al. (2001)
Wheat	<i>Ephestia kuehniella</i> and <i>Tribolium confusum</i> ,	Ozone concentration of 13.9 mg/L	<i>T. confusum</i> , 72.6% (Larvae) 1.3–22.7% (Adult) 90–100% (larve, adult)	Isikber and Oztekin (2009)
Stored products	<i>Tribolium castaneum</i> <i>Rhyzopertha dominica</i> <i>Oryzaephilus surinamensis</i>	50 ppm 30 °C and 70% RH	50% mortality 11.39–20.10 h (TC) 9.22–12.19 h (RD) 6.1–9.66 h (OS) 95% mortality 22.17–37.9 h (TC) 21.85–35.17 h (RD) 11.03–18.72 h (OS)	Sousa et al. (2008)
Maize	<i>T. castaneum</i>	50 ppm	50% mortality (71.4 h) at 20 °C 95% mortality (151.8 h) at 20 °C	Pereira et al. (2008)

IMM: Indian meal moth; Maize weevil; RFB: Red flour beetle; RD: *Rhyzopertha dominica*; OS: *Oryzaephilus surinamensis*.

contamination in cereals and cereal products, including spores of *Bacillus*, Coliform bacteria, *Micrococcus*, *Flavobacterium*, *Alcaligenes*, *Serratia*, *Aspergillus* and *Penicillium* (Naito and Takahara, 2006). Some studies show that up to 3 log reductions of micro-organisms in cereal grains, peas and beans can be achieved depending on ozone concentration, temperature and relative humidity conditions (Naito, 1989; Naito, 1990; Naito et al., 1987, 1988; Naito and Nanba, 1987; Naito and Shiga, 1989; Naito and Takahara, 2006). Transmission electron microscopic micrographs of *Bacillus* spores treated with ozone, suggest that ozone inactivates spores by degrading the outer spore component (spore coat layers comprise approximately 50% of the spore volume), thus exposing the cortex and core to the action of ozone (Foegeding, 1985; Khadre and Yousef, 2001).

Fungal or mould contamination of food grains is one of the most important issues determining grain quality, with both qualitative and quantitative losses reported due to microbes. Micro-organisms which are present in food grains either on their surface or internally, deteriorate the nutritional quality of the products and also produce metabolites (e.g. mycotoxins) which are dangerous to human and animal health (Raila et al., 2006). Ozone has been effectively used to control fungal growth and reduce mycotoxin contamination. Fungal and mould growth lead to the release of secondary metabolites known as mycotoxins. Fig. 2 lists some of the commonly occurring mycotoxins found in food grains. These are known to exhibit carcinogenic, teratogenic, immunosuppressive properties and cause several physiological disorders both in humans and animals (Fung and Clark, 2004; Leung et al., 2006; Mally and Dekant, 2009; Stockmann-Juvala and Savolainen, 2008; Wu et al., 2009).

The human health risks from mycotoxins in cereals and cereal-based products, (e.g. beer) and risk to animals from contaminated feed and cereal by-products is widely reported. Recently, Wolf-Hall (2007) reviewed the safety concerns arising from moulds and mycotoxins encountered during malting and brewing and highlighted the risks associated with mycotoxins in barley. Because of the potential grain losses to the farmer and toxicological hazards to the consumer, developing cost effective methods to detoxify mycotoxin-contaminated grains and foods is a priority (Young et al., 2006). Application of ozone in grain handling and storage could reduce or eliminate mycotoxins and undesirable microflora (fungi and moulds) from grain and grain products.

## 6. Degradation of mycotoxins

Ozone is reported to be effective in the detoxification and degradation of commonly occurring mycotoxins such as aflatoxin,

patulin, cyclopiazonic acid, secalononic acid D, ochratoxin A, and ZEN (Lemke et al., 1999; McKenzie et al., 1997). Table 2 lists reported effects of ozone treatment on fungal and mycotoxins in food grains. Similar to the killing of insects, fungal inactivation and subsequent decontamination of toxins depends upon several factors including ozone concentration, exposure time, pH and moisture content of the grain mass. For example Raila et al. (2006) observed 2.2 times higher fungal decontamination of wheat grains at a grain moisture content of 15.2% and 3 times higher at 22.0% grain moisture compared to dry conditions. This could be due to the greater efficacy of ozone in aqueous media compared to a gaseous phase, higher fungal growth in wet conditions than in dry conditions and slower movement of ozone within grain layers in moist conditions, thus allowing greater exposure time. Hence, moisture is an important factor influencing the efficacy of ozone. Application of dry ozone gas is reported to be less effective compared to moist conditions. For example, Young (1986) observed a 90% reduction in deoxynivalenol (also known as vomitoxin) during moist ozone (1.1 mol %) treatment and a 70% reduction after 1 h of ozone treatment under similar experimental conditions in corn. The efficacy of ozone is also reported to be influenced by the pH of the medium. Young et al. (2006) observed a rapid degradation of trichothecene mycotoxins at a low pH (pH 4–6) compared to a higher pH (pH 7–8). However, the effect of pH is unimportant as far as ozonation of food grain is concerned.

Grain temperature also influences the efficacy of ozone in degradation of mycotoxins. Proctor et al. (2004) reported a greater degradation of aflatoxins in peanut kernels at higher temperature. They reported a reduced treatment time from 15 min to 10 min for a temperature rise from 25 °C to 75 °C to obtain a degradation of 77 % for AFB1 and 80% for AFG1 respectively. Further, they achieved a greater degradation in peanut kernel compared to flour, most likely due to the larger exposure area and also the fact that fungal or mould contamination is principally found on the grain surface. Lower doses of ozone (5 ppm in atmosphere) are reported to inhibit surface growth, sporulation and mycotoxin production by *Aspergillus flavus* and *Fusarium moniliforme* (Mason et al., 1997). Similarly Allen et al. (2003) reported that an ozone dose of 0.16 mg/g barley is sufficient to achieve 96% inactivation of fungal spores within 5 min.

Ozone gas was demonstrated to be effective for the degradation and detoxification of common mycotoxins (aflatoxins B1, B2, G1, and G2, cyclopiazonic acid, fumonisin B1, ochratoxin A, patulin, secalononic acid, and zearalenone) in aqueous solution (McKenzie et al., 1997). Ozone either completely degrades mycotoxins or causes chemical modifications, reducing their biological activity

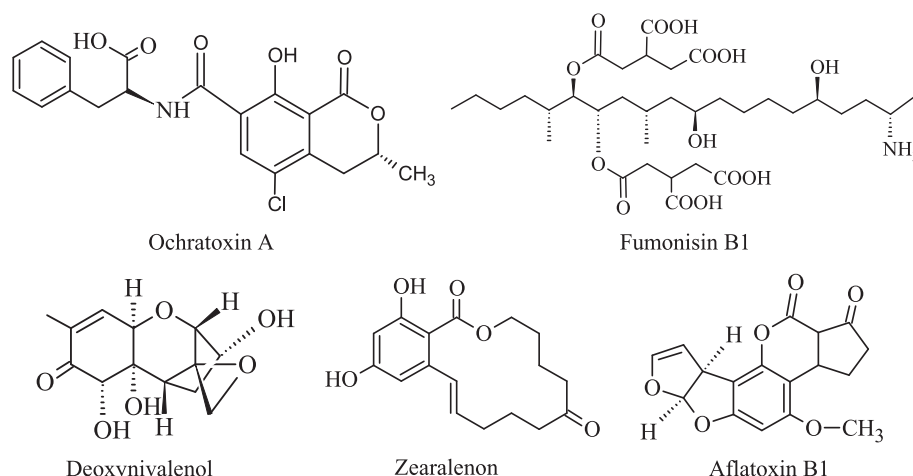


Fig. 2. Some of the commonly occurring food grain mycotoxins.

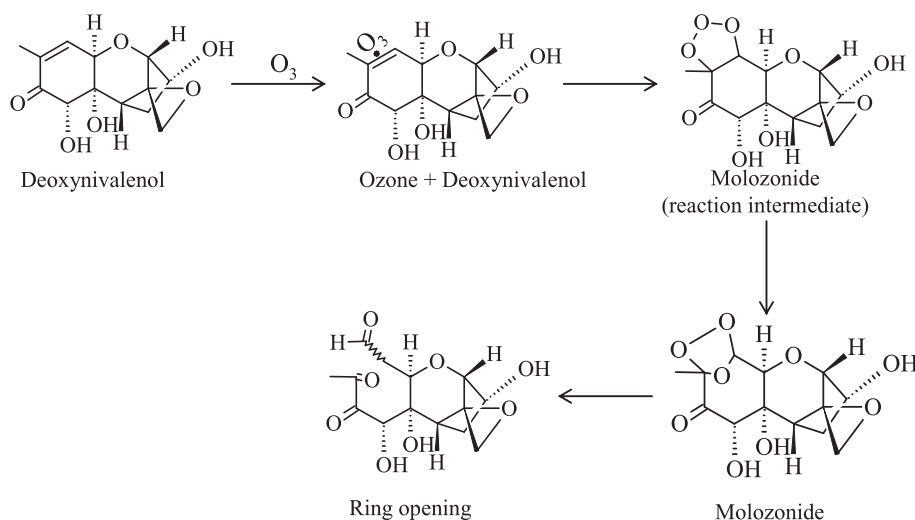
**Table 2**  
Effects of ozone treatment on mycotoxins.

Food grain or product	Mycotoxins	Treatment conditions	Degradation	Reference
Peanut meals	Aflatoxins	25 mg ozone/min	AFB1 and G1 (100% destruction) AFB2 (78%)	Dwarakanath et al. (1968)
Corn	Aflatoxins	Ozonation (10–12 wt%) reduced	aflatoxin levels by 92%	Prudente and King (2002)
Corn	Aflatoxin	92 h with O <sub>3</sub> at 200 mg/min	>95% reduction in AFB1	McKenzie et al. (1998)
Barley	Fungal spores and mycelia	0.16 mg of ozone (g wheat) <sup>-1</sup> min <sup>-1</sup>	96% fungal spore inactivation	Allen et al. (2003)
Maize	<i>Aspergillus parasiticus</i>	50 ppm for 3 days	63% reduction	Kells et al. (2001)
Wheat	Fungal spores and mycelia	0.33 mg of ozone (g wheat) <sup>-1</sup> min <sup>-1</sup>	96.9% fungal spore inactivation	Wu et al. (2006)
Wheat	Deoxynivalenol	Moist ozone (1.1 mol %) for 1 h	90% reduction	Young (1986)
Wheat	Deoxynivalenol	Dry ozone (1.1 mol %) for 1 h	70% reduction	Young (1986)
Barley	Fusarium	Gaseous ozone treatment 11 and 26 mg/g for 15 min	24–36% decrease in <i>Fusarium</i> survival	Kottapalli et al. (2005)
Peanut	Aflatoxin	Gaseous ozone (4.2 wt%) Temperature 25, 50, 75 °C	AFB1 (77%): 10 min at 75 °C AFB1 (80%): 5–10 min at 75 °C,	Proctor et al. (2004)

(Lemke et al., 1999; McKenzie et al., 1998). However degradation or chemical modification is specific to the structure of mycotoxins under investigation. McKenzie et al. (1997) observed greater resistance of aflatoxin B<sub>2</sub> and G<sub>2</sub> compared to B<sub>1</sub> and G<sub>1</sub> and the presence of double bonds at C<sub>8</sub>–C<sub>9</sub> position for B<sub>1</sub> and G<sub>1</sub> and the tendency of ozone to react at olefinic positions indicate the possibility for greater sensitivity towards ozone. According to the Criegee mechanism of degradation, the ozone molecule undergoes 1–3 dipolar cyclo addition with a double bond (Criegee, 1975). This leads to the formation of ozonides (1,2,4-trioxolanes) from alkenes and ozone with aldehyde or ketone oxides as decisive intermediates, all of which have finite lifetimes Criegee, 1975. Oxidative disintegration of ozonide and formation of carbonyl compounds result, while oxidative work-up leads to carboxylic acids or ketones (Cullen et al., 2009). Apart from the degradation of double bonded aliphatic or polycyclic aromatic hydrocarbons, the presence of chlorinated ring structures (e.g. Ochratoxin) and nitrogen heterocycles (e.g. Fumonisin) in mycotoxins (Fig. 3) are also subject to ozone attack, resulting in free chlorine or amino acids (Lemke et al., 1999).

Studies show that the biological toxicity of certain mycotoxins significantly decreases, following ozone treatment due to the formation of new products. McKenzie et al. (1997) reported the formation of Fumonisin B1 derivatives due to the ozonation of Fumonisin B1 in aqueous solution. They proposed that this might be due to the reaction of ozone with the primary amine (–NH<sub>2</sub>)

group leading to the formation of nitrogen oxide (–NO<sub>2</sub> or –N<sub>2</sub>O) coupled with the formation of ketone by the Criegee mechanism. However, the authors observed biological toxicity in a mycotoxin-sensitive bioassay possibly due to the presence of an intact primary amine in the Fumonisin B1 derivative. It has been shown that the primary amine group is necessary for the biological activity of fumonisin like compounds (Burns et al., 2008; Gelderblom et al., 1993; Stockmann-Juvala and Savolainen, 2008). Similarly, Young et al. (2006) studied the degradation of trichothecene mycotoxins (3-acetyldeoxynivalenol, 15-acetyldeoxynivalenol, diacetoxyscirpenol, 4-deoxynivalenol, fusarenon X, HT-2 toxin, 15-monoacetoxyscirpenol, neosolaniol, T-2 triol and verrucarol) by aqueous ozone. They observed a complete degradation of these mycotoxins to simple products at ozone concentrations of ≥25 ppm and intermediate products at ozone concentrations of <25 ppm due to partial degradation or chemical modification. The degradation mechanism of these mycotoxins can also be described by the Criegee mechanism as shown in Fig. 3. Intermediate compounds (keto trichothecenes or de-epoxy trichothecenes) formed after ozone treatment of trichothecenes are reported to be less toxic than their parent compounds (Beyer et al., 2009; Königs et al., 2009; Sundstøl Eriksen et al., 2004). Mammalian studies show that ozone effectively detoxifies mycotoxins. The potential to extend ozonation techniques for the remediation of contaminated feeds (Lemke et al., 1999; McKenzie et al., 1998) and food grains to ensure consumer safety has been demonstrated.



**Fig. 3.** Proposed mechanism for degradation of deoxynivalenol.

## 7. Effect on grain quality

Application of ozone at doses that are sufficient for the effective decontamination of grains may affect various quality attributes. The effects of ozone treatment on the quality and physiology of various food grains are reported in Table 3. Ozone is not universally beneficial and in some cases may promote oxidation degradation of chemical constituents present in the grains. Surface oxidation, discoloration or development of undesirable odours may occur from excessive use of ozone. Mendez et al. (2003) observed discoloration of ozonated rice (50 ppm for 30 days) husks which also had a vinegar odour compared to the control treatments. However, both the acidic odour and brown discoloration were removed during the milling process. Ozone is reported to alter the amino acid and fatty acid profile in aqueous solutions (Richard and Brener, 1984) by oxidizing the sulfhydryl group (–SH) of amino acids and oxidation of polyunsaturated fatty acids to peroxides (Guzel-Seydim et al., 2004) thus influencing the nutritional and metabolic value of grain. However, Mendez et al. (2003) did not observe any significant change in the amino acid and fatty acid profiles of ozonated wheat, soybean or maize. This indicates that penetration of ozone into the kernel is unlikely. Similarly, Prudente and King (2002) reported insignificant changes to the fatty acid profile of ozonated corn. Lower ozone concentrations (0.05–5 ppm) do not induce lipid oxidation in food grains. Similarly, Wang et al. (2008) showed the loss of protein content in ozonated corn samples compared to untreated corn.

However, higher ozone concentrations (>50 ppm) cause considerable oxidative damage to cereal grain flours (Naito, 1989). Mendez et al. (2003) investigated the effect of prolonged ozone treatment (6 times that needed to kill storage pests) on a range of food grains (Table 3) and did not observe any effect on the nutritional quality and processing characteristics of ozonated grains.

Storage of grains in ozone rich atmospheres does not influence the rheological properties of grains. For example Mendez et al. (2003) investigated the efficacy of ozone to control pests for stored wheat and rice. They reported that ozone treatment does not significantly change the bread-making properties of hard wheat, including tolerance of the dough to overmixing, absorption of water, dough weight, and proof height. Ozone treatments of rice grain during storage have no significant effect on the cooking quality (adhesiveness) of cooked rice.

Ozonation of wheat grain and subsequent flour milling and baking properties along with biochemical composition are

reported to be unaffected (Ibanoglu, 2001, 2002; Mendez et al., 2003). Ibanoglu (2002) studied the effect of ozonation of wheat grains and the subsequent effect on wheat flour properties. They reported that wheat washing with ozonated water (1.5 mg/L) for 30 min significantly reduces microbial load compared to normal water without affecting the rheological properties of wheat flour including extensibility and maximum resistance to extension (flour strength). However, soft wheat was more significantly affected by ozonation than hard wheat. This could be due to easier penetration of ozone into the endosperm in soft wheat compared to hard wheat. Farinogram profiles, an important rheological property characterising the baking properties of wheat flour were not affected for either soft or hard wheat samples. Studies conducted by Ibanoglu (2001) show that tempering of wheat grains with ozonated water does not have a significant effect on the rheological properties of wheat flour pertinent to baking. Ozonation of soft to medium wheat flour is reported to cause an increase in the resistance to extension of wheat flours and a decrease in extensibility (Naito, 1990).

A study conducted by Desvignes et al. (2008) examined the mechanical properties of the outer layers and the milling behaviour of wheat flour during ozone treatment (10 g/kg) by the patented ozone process “Oxygreen®”. Results indicated a reduction of the aleurone layer extensibility and the local endosperm resistance to rupture (i.e. an increase in friability) was affected. The ozonation of wheat grain causes a significant effect on the maximum viscosity and the setback value of ozonated wheat flour as indicated from viscoamylographs. Similarly, ozone treatment of rice starch is reported to enhance swelling with a reduced retrogradation tendency (An and King, 2009).

Ozone treatment of moistened wheat grain is reported to enhance dehulling of wheat grain (Coste et al., 2008). Desvignes et al. (2008) observed a significant reduction (10–20%) in the required total energy for milling without significant changes in biochemical characterisation of the milling fractions, demonstrating the potential of ozone application for food grains. This reduction in milling energy could be due to changes in the mechanical properties of wheat grain tissues. However, they observed a significant reduction in the coarse bran yield (~30%) with a comparable increase in the white shorts yield. This shows that ozonation has a tendency to enhance bran friability or easy separation of bran from the starchy endosperm. Further, a reduction in the level of starch damage in flours from ozonated grains due to the reduced energy requirement (Desvignes et al., 2008) may influence the potential starch enzymatic degradation and water absorption during dough making.

**Table 3**  
Effect of ozone treatment on food grain quality.

Food grain	Conditions	Changes in quality	Reference
Wheat	Ozone concentration 10 g kg <sup>-1</sup> Oxygreen® process	Flour yield (~), Milling energy required (↓), bran friability (↑), mechanical properties (√)	Desvignes et al. (2008)
Wheat	Ozonated water (1.5 mg L <sup>-1</sup> )	Extensibility (↓); water absorption (~); dough development time (~); dough stability (~); degree of softening (~); overall flour quality (~)	Ibanoglu (2001)
Barley	Gaseous ozone treatment 11 and 26 mg/g for 30 min	Germination energy (↓)	Kottapalli et al. (2005)
Soybean	50 ppm for 30 days	Fatty acid profile (~); Amino acid profile (~); rancidity (~)	Mendez et al. (2003)
Wheat	50 ppm for 30 days	Fatty acid profile (~); Amino acid profile (~); rancidity (~) bread-making properties (~); Milling performance (~)	Mendez et al. (2003)
Maize	50 ppm for 30 days	Fatty acid profile (~); Amino acid profile (~); popping volume (~)	Mendez et al. (2003)
Rice	50 ppm for 30 days	Adhesiveness (~); cooking quality (~)	Mendez et al. (2003)
Barley	Ozonation (10–12 wt%)	Fatty acid profile (~)	Prudente and King (2002)
Wheat	0.33 mg of ozone (g wheat) <sup>-1</sup> min <sup>-1</sup>	Germination rate (↓)	Wu et al. (2006)
Barley	0.16 and 0.10 mg of ozone (g Barley) <sup>-1</sup> min <sup>-1</sup>	Germination rate (↓)	Allen et al. (2003)

(×): Significant difference; (√) Insignificant; (↑): Increases; (↓): Decreases and (~): No change.

## 8. Conclusion

Within the grain processing industry, there is an increasing emphasis and trend toward the safe storage of food grains while minimizing qualitative and quantitative losses. Food laws and legislation to phase out chemical pesticides, increased pest resistance towards conventional fumigants and growing consumer demand for “greener” additives has forced processors, grain handlers, food scientists and entomologists to find alternatives. Ozone is an alternative method of grain disinfestation which is environment friendly resulting in no toxic residues. This review paper demonstrated that ozone is an effective greener alternative against a range of pests, micro-organisms and mycotoxins. However, the effectiveness of ozone depends on several factors including the amount of ozone applied, various environmental factors such as grain mass temperature, moisture and the surface characteristics. Ozone offers unique advantages for food grain processing with minimal or desired effects on the physicochemical properties. Hence ozone treatment is a potential greener alternative to conventional fumigants.

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## References

- Allen, B., Wu, J.N., Doan, H., 2003. Inactivation of fungi associated with barley grain by gaseous ozone. *Journal of Environmental Science and Health Part B-Pesticides Food Contaminants and Agricultural Wastes* 38 (5), 617–630.
- An, H.J., King, J.M., 2009. Using ozonation and amino acids to change pasting properties of rice starch. *Journal of Food Science* 74 (3), C278–C283.
- Ballinger, C.A., Cueto, R., Squadrito, G., Coffin, J.F., Velsor, L.W., Pryor, W.A., Postlethwait, E.M., 2005. Antioxidant-mediated augmentation of ozone-induced membrane oxidation. *Free Radical Biology & Medicine* 38, 515–526.
- Beyer, M., Ferse, I., Mulac, D., Würthwein, E.U., Humpf, H.U., 2009. Structural elucidation of T-2 toxin thermal degradation products and investigations toward their occurrence in retail food. *Journal of Agricultural and Food Chemistry* 57 (5), 1867–1875.
- Bornholdt, J., Dybdahl, M., Vogel, U., Hansen, M., Loft, S., Wallin, H., 2002. Inhalation of ozone induces DNA strand breaks and inflammation in mice. *Mutation Research* 520, 63–72.
- Burns, T.D., Snook, M.E., Riley, R.T., Voss, K.A., 2008. Fumonisin concentrations and in vivo toxicity of nixtamalized *Fusarium verticillioides* culture material: evidence for fumonisin-matrix interactions. *Food and Chemical Toxicology* 46 (8), 2841–2848.
- Collins, P.J., Daglish, G.J., Pavic, H., Kopitke, R.A., 2005. Response of mixed-age cultures of phosphine-resistant and susceptible strains of lesser grain borer, *Rhyzopertha dominica*, to phosphine at a range of concentrations and exposure periods. *Journal of Stored Products Research* 41 (4), 373–385.
- Coste, C., Dubois, M. and Pernot, A. 2008. Dehulling wheat grains using ozone Patent Application Number: EP20070731758.
- Criegee, 1975. Mechanism of Ozonolysis. doi:10.1002/anie197507451.
- Cullen, P.J., Tiwari, B.K., O'Donnell, C.P., Muthukumarappan, K., 2009. Modelling approaches to ozone processing of liquid foods. *Trends in Food Science and Technology* 20 (3–4), 125–136.
- Desvignes, C., Chaurand, M., Dubois, M., Sadoudi, A., Abecassis, J., Lullien-Pellerin, V., 2008. Changes in common wheat grain milling behavior and tissue mechanical properties following ozone treatment. *Journal of Cereal Science* 47 (2), 245–251.
- Dubois, M., Coste, C., Despres, A.G., Efstathiou, T., Nio, C., Dumont, E., Parent-Massin, D., 2006. Safety of Oxygreen (R), an ozone treatment on wheat grains. Part 2. Is there a substantial equivalence between oxygreen-treated wheat grains and untreated wheat grains? *Food Additives and Contaminants* 23 (1), 1–15.
- Dubois, M., Canadas, D., Despres-Pernot, A.G., Coste, C., Pfohl-Leszkowicz, A., 2008. Oxygreen process applied on nongerminated and germinated wheat: role of hydroxamic acids. *Journal of Agricultural and Food Chemistry* 56 (3), 1116–1121.
- Dwarakanath, C., Rayner, E., Mann, G., Dollear, F., 1968. Reduction of aflatoxin levels in cottonseed and peanut meals by ozonation. *Journal of the American Oil Chemists' Society* 45 (2), 93–95.
- Erdman, H.E., 1980. Ozone toxicity during ontogeny of two species of flour beetles, *Tribolium confusum* and *T. castaneum*. *Environmental Entomology* 9 (1), 16–17.
- Fields, P.G., White, N.D.G., 2002. Alternatives to methyl bromide treatments for stored-product and quarantine insects. *Annual Review of Entomology* 47, 331–359.
- Foegeding, P.M., 1985. Ozone inactivation of *Bacillus* and *Clostridium* spore populations and the importance of the spore coat to resistance. *Food Microbiology* 2 (2), 123–134.
- Fung, F., Clark, R.F., 2004. Health effects of mycotoxins: a toxicological overview. *Journal of Toxicology–Clinical Toxicology* 42 (2), 217–234.
- Gelderblom, W.C.A., Cawood, M.E., Snyman, D., Vlegaar, R., Marasas, W.F.O., 1993. Structure-activity relationships of fumonisins in short-term carcinogenesis and cytotoxicity assays. *Food and Chemical Toxicology* 31 (6), 407–414.
- Graham, D.M., 1997. Use of ozone for food processing. *Food Technology* 51, 121–137.
- Güzel-Seydim, Z., Bever, J.P.I., Greene, A.K., 2004. Efficacy of ozone to reduce bacterial populations in the presence of food components. *Food Microbiology* 21, 475–479.
- Guzel-Seydim, Z.B., Greene, A.K., Seydim, A.C., 2004. Use of ozone in the food industry. *Lebensmittel-Wissenschaft und-Technologie* 37 (4), 453–460.
- Hetz, S.K., Bradley, T.J., 2005. Insects breathe discontinuously to avoid oxygen toxicity. *Nature* 433 (7025), 516–519.
- Ibanoglu, S., 2001. Influence of tempering with ozonated water on the selected properties of wheat flour. *Journal of Food Engineering* 48, 345–350.
- Ibanoglu, S., 2002. Wheat washing with ozonated water: effects on selected flour properties. *International Journal of Food Science and Technology* 37 (5), 579–584.
- Isikber, A.A., Oztekin, S., 2009. Comparison of susceptibility of two stored-product insects, *Ephesia kuehniella* Zeller and *Tribolium confusum* du Val to gaseous ozone. *Journal of Stored Products Research* 45 (3), 159–164.
- Islam, M.S., Hasan, M.M., Xiong, W., Zhang, S.C., Lei, C.L., 2009. Fumigant and repellent activities of essential oil from *Coriandrum sativum* (L.) (Apiaceae) against red flour beetle *Tribolium castaneum* (Herbst) (Coleoptera: Tenebrionidae). *Journal of Pest Science* 82 (2), 171–177.
- Kaushik, G., Satya, S., Naik, S.N., 2009. Food processing a tool to pesticide residue dissipation – A review. *Food Research International* 42 (1), 26–40.
- 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.
- Khadre, M.A., Yousef, A.E., 2001. Sporicidal action of ozone and hydrogen peroxide: a comparative study. *International Journal of Food Microbiology* 71 (2–3), 131–138.
- Khadre, M.A., Yuosef, A.E., Kim, J., 2001. Microbiological aspects of ozone applications in food: a review. *Journal of Food Science* 6, 1242–1252.
- 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, 1071–1087.
- Königs, M., Mulac, D., Schwerdt, G., Gekle, M., Humpf, H.U., 2009. Metabolism and cytotoxic effects of T-2 toxin and its metabolites on human cells in primary culture. *Toxicology* 258 (2–3), 106–115.
- Kottapalli, B., Wolf-Hall, C.E., Schwarz, P., 2005. Evaluation of gaseous ozone and hydrogen peroxide treatments for reducing *Fusarium* survival in malting barley. *Journal of Food Protection* 68 (6), 1236–1240.
- Leesch, J.G., 2003. The mortality of stored-product insects following exposure to gaseous ozone at high concentrations. In: Credland, P.F., Armitage, D.M., Bell, C.H., Cogan, P.M., Highley, E. (Eds.), *Advances in Stored Product Protection, Proceedings of the 8th International Working Conference on Stored-Product Protection, 22–26 July 2002, York, UK*. CAB International, Oxon, UK, pp. 827–831.
- Lemke, S.L., Mayura, K., Ottinger, S.E., McKenzie, K.S., Wang, N., Fickey, C., Kubena, L.F., Phillips, T.D., 1999. Assessment of the estrogenic effects of zearalenone after treatment with ozone utilizing the mouse uterine weight bioassay. *Journal of Toxicology and Environmental Health – Part A* 56 (4), 283–295.
- Leung, M.C.K., Diaz-Llano, G., Smith, T.K., 2006. Mycotoxins in pet food: a review on worldwide prevalence and preventative strategies. *Journal of Agricultural and Food Chemistry* 54 (26), 9623–9635.
- Liu, H., Wu, Y., Chen, H., 2007. Production of ozone and reactive oxygen species after welding. *Archives of Environmental Contamination and Toxicology* 53 (4), 513–518.
- Lu, B., Ren, Y., Du, Y.-z., Fu, Y., Gu, J., 2009. Effect of ozone on respiration of adult *Sitophilus oryzae* (L.), *Tribolium castaneum* (Herbst) and *Rhyzopertha dominica* (F.). *Journal of Insect Physiology* 55 (10), 885–889.
- Mahapatra, A.K., Muthukumarappan, K., Julson, J.L., 2005. Applications of ozone bacteriocins and irradiation in food processing: a review. *Critical Reviews in Food Science and Nutrition* 45, 447–461.
- Mally, A., Dekant, W., 2009. Mycotoxins and the kidney: modes of action for renal tumor formation by ochratoxin A in rodents. *Molecular Nutrition & Food Research* 53 (4), 467–478.
- Mason, L.J., Woloshuk, C.P., Maier, D.E., 1997. Efficacy of ozone to control insects, molds and mycotoxins. Nicosia. In: Donahaye, E.J., Navarro, S., Varnava, A. (Eds.), *Proceedings of the International Conference on Controlled Atmosphere and Fumigation in Stored Products*. Cyprus Printer Ltd., Nicosia, pp. 665–670.
- McKenzie, K.S., Sarr, A.B., Mayura, K., Bailey, R.H., Miller, D.R., Rogers, T.D., Norred, W.P., Voss, K.A., Plattner, R.D., Kubena, L.F., Phillips, T.D., 1997. Oxidative degradation and detoxification of mycotoxins using a novel source of ozone. *Food and Chemical Toxicology* 35 (8), 807–820.
- McKenzie, K.S., Kubena, L.F., Denvir, A.J., Rogers, T.D., Hitchems, G.D., Bailey, R.H., Harvey, R.B., Buckley, S.A., Phillips, T.D., 1998. Aflatoxicosis in turkey poults is

- prevented by treatment of naturally contaminated corn with ozone generated by electrolysis. *Poultry Science* 77, 1094–1102.
- Mendez, F., Maier, D.E., Mason, L.J., Woloshuk, C.P., 2003. Penetration of ozone into columns of stored grains and effects on chemical composition and processing performance. *Journal of Stored Products Research* 39 (1), 33–44.
- Naito, S., Nanba, Y., 1987. Studies on utilization of ozone in food preservation .4. The effect of ozone treatment on thiamin and riboflavin contents of cereal-grains, cereal grain powders, peas, beans and whole species. *Journal of the Japanese Society for Food Science and Technology-Nippon Shokuhin Kagaku Kogaku Kaishi* 34 (12), 794–800.
- Naito, S., Shiga, I., 1989. Studies on utilization of ozone in feed preservation .9. Effect of ozone treatment on elongation of hypocotyl and microbial counts of bean sprouts. *Journal of the Japanese Society for Food Science and Technology-Nippon Shokuhin Kagaku Kogaku Kaishi* 36 (3), 181–188.
- Naito, S., Takahara, H., 2006. Ozone contribution in food industry in Japan. *Ozone-Science & Engineering* 28 (6), 425–429.
- Naito, S., Okada, Y., Sakai, T., 1987. Studies on utilization of ozone in food preservation .3. Microbicidal properties of ozone on cereal-grains, cereal grain powders, peas, beans and whole spices. *Journal of the Japanese Society for Food Science and Technology-Nippon Shokuhin Kagaku Kogaku Kaishi* 34 (12), 788–793.
- Naito, S., Okada, Y., Sakai, T., 1988. Studies on utilization of ozone in food preservation .5. Changes in microflora of ozone-treated cereals, grains, peas, beans and spices during storage. *Journal of the Japanese Society for Food Science and Technology-Nippon Shokuhin Kagaku Kogaku Kaishi* 35 (2), 69–77.
- Naito, S., 1989. Studies on utilization of ozone for food preservation .6. The influence of ozone treatment on lipids contained in cereal-grains, cereal grain powders, peas, beans and pulse products. *Journal of the Japanese Society for Food Science and Technology-Nippon Shokuhin Kagaku Kogaku Kaishi* 36 (11), 878–883.
- Naito, S., 1990. Effect of ozone treatment on the rheological properties of wheat-flour. *Journal of the Japanese Society for Food Science and Technology-Nippon Shokuhin Kagaku Kogaku Kaishi* 37 (10), 810–813.
- Pereira, A.d.M., Faroni, L.R.D., Silva Jr., A.G.d.S., Sousa, A.H.d., Paes, J.L., 2008a. Economical viability of ozone use as fumigant of stored corn grains. *Engenharia Na Agricultura* 16 (2), 144–154.
- Pereira, A.d.M., Faroni, L.R.D.A., De Sousa, A.H., Urruchi, W.I., Paes, J.L., 2008b. Influence of the grain temperature on the ozone toxicity to *Tribolium castaneum*. *Revista Brasileira de Engenharia Agrícola e Ambiental* 12 (5), 493–497.
- Pimentel, M.A.G., Faroni, L.R.D., Tótolá, M.R., Guedes, R.N.C., 2007. Phosphine resistance, respiration rate and fitness consequences in stored-product insects. *Pest Management Science* 63 (9), 876–881.
- Pimentel, M.A.G., Faroni, L.R.D., Guedes, R.N.C., Sousa, A.H., Totola, M.R., 2009. Phosphine resistance in Brazilian populations of *Sitophilus zeamais* Motschulsky (Coleoptera: Curculionidae). *Journal of Stored Products Research* 45 (1), 71–74.
- 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.
- Prudente Jr., A.D., King, J.M., 2002. Efficacy and safety evaluation of ozonation to degrade aflatoxin in corn. *Journal of Food Science* 67 (8), 2866–2872.
- Qin, Z.G., Wu, X., Deng, G., Yan, X.P., He, X.C., Xi, D.K., Liao, X.W., 2003. Investigation of the use of ozone fumigation to control several species of stored grain insects. *Adv. Stored Prod. Prot.* 846–851.
- 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.
- Restaino, L., Frampton, E., Hemphill, J., Palnikar, P., 1995. Efficacy of ozonated water against various food related micro-organisms. *Applied Environmental Microbiology* 61, 3471–3475.
- Richard, Y., Brener, L., 1984. Removal of ammonia and other nitrogen derivatives from drinking water with ozone. In: Rice, R.G., Netzer, A. (Eds.), *Handbook of Ozone Technology and Applications*, vol. II. Butterworth, Boston, pp. 99–121.
- Rozado, A.F., Faroni, L.R.A., Urruchi, W.M.I., Guedes, R.N.C., Paes, J.L., 2008. Ozone application against *Sitophilus zeamais* and *Tribolium castaneum* on stored maize. *Revista Brasileira de Engenharia Agrícola e Ambiental* 12 (3), 282–285.
- Shunmugam, G., Jayas, D.S., White, N.D.G., Muir, W.E., 2005. Diffusion of carbon dioxide through grain bulks. *Journal of Stored Products Research* 41 (2), 131–144.
- Sousa, A.H., Faroni, L.R.D.A., Guedes, R.N.C., Tótolá, 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.
- Stockmann-Juvala, H., Savolainen, K., 2008. A review of the toxic effects and mechanisms of action of fumonisin B-1. *Human & Experimental Toxicology* 27 (11), 799–809.
- Sundstøl Eriksen, G., Pettersson, H., Lundh, T., 2004. Comparative cytotoxicity of deoxynivalenol, nivalenol, their acetylated derivatives and de-epoxy metabolites. *Food and Chemical Toxicology* 42 (4), 619–624.
- Wang, Y., King, J.M., Xu, Z., Losso, J., Prudente, A., 2008. Lutein from ozone-treated corn Retains Antimutagenic properties. *Journal of Agricultural and Food Chemistry* 56 (17), 7942–7949.
- Wolf-Hall, C.E., 2007. Mold and mycotoxin problems encountered during malting and brewing. *International Journal of Food Microbiology* 119 (1–2), 89–94.
- Wu, J.N., Doan, H., Cuenca, M.A., 2006. Investigation of gaseous ozone as an anti-fungal fumigant for stored wheat. *Journal of Chemical Technology and Biotechnology* 81 (7), 1288–1293.
- Wu, Q., Jezkova, A., Yuan, Z., Pavlikova, L., Dohnal, V., Kuca, K., 2009. Biological degradation of aflatoxins. *Drug Metabolism Reviews* 41 (1), 1–7.
- Young, J.C., Zhu, H., Zhou, T., 2006. Degradation of trichothecene mycotoxins by aqueous ozone. *Food and Chemical Toxicology* 44 (3), 417–424.
- 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.
- Yvin, J.-C., Bailli, A., Joubert, J.-M. and Bertaud, O. 2001. Method and installation for making flour from ozone-treated grains, World Patent, WO0143556.