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Cold plasma technology for aflatoxin B1 detoxification in foods: focused review of mechanisms, efficiency and implementation challenges: a review

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ABSTRACT

Aflatoxin B1 (AFB1), as a secondary metabolite by *Aspergillus, Penicillium, Fusarium,* and *Alternaria,* a potent carcinogen, is a major global food safety concern. Traditional decontamination methods often compromise food quality or leave residues. Non-thermal cold plasma presents a promising alternative, effectively degrading AFB1 without heat. This technology utilizes reactive oxygen and nitrogen species (RONS) to break down the toxin. Different systems—including dielectric barrier discharge (DBD), plasma jets, and corona discharge—have demonstrated high efficacy while preserving food quality. Their success depends on key parameters like gas composition, treatment time, and the food matrix itself. However, challenges remain for industrial adoption. Scaling up the technology, ensuring regulatory approval, and maintaining the sensory and nutritional properties of treated food are critical hurdles. Future research should explore hybrid methods combining plasma with other treatments and conduct long-term in-vivo safety studies. This review assesses the current state and future potential of cold plasma as a sustainable solution for mitigating AFB1 contamination and enhancing food security

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1. Introduction

Food contaminated with foodborne pathogens can become a widespread health crisis. This contamination can occur not only during the production process, but also during food transportation, storage, and even preparation (1).

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Food contamination may concur because of physical, chemical, and microbial potential (2). Certain species of fungi, under field or storage conditions, secrete dangerous toxic compounds called mycotoxins. These toxins are actually byproducts of fungal metabolism that can easily contaminate food and threaten human health (3). While some fungi are beneficial, others produce harmful mycotoxins (4). Among all mycotoxins, aflatoxin is one of the most significant, with impacts ranging from acute to chronic disease (5). Grains, nuts, spices, and dried fruits are ideal breeding grounds for toxic fungi like *Aspergillus flavus* and *A. parasiticus*. These fungi produce a dangerous toxin called aflatoxin, which contaminates food and poses a serious health risk to consumers (3, 6, 7).

The most important aflatoxins from a food safety perspective include types B (B1 and B2), G (G1 and G2), and M (M1 and M2) (3). Aflatoxin B1 (AFB1), the most prevalent arising carcinogen, causes necrosis in vital organs and is a key concern in global trade due to strict regulatory limits (8, 9). Developing countries in Africa, Asia, and Latin America are particularly vulnerable due to inadequate storage conditions and lack of stringent regulatory measures (10). Even in developed nations, aflatoxin contamination remains a concern, especially in imported food products, highlighting the need for global surveillance and mitigation strategies (11).

Aflatoxin reduction can be achieved via chemical, biological, or physical methods (12), though chemical approaches are often unsuitable due to residual toxicity. Thus, non-thermal approaches like plasma technology are increasingly adopted. One of the emerging non-thermal methods which has increased progressively in the food manufacturing is the use of plasma technology. Plasma - the fourth state of matter - is an ionized medium comprising charged particles (electrons and ions), free radicals, metastable excited species, and vacuum ultraviolet radiation that behaves differently from the usual states of matter (13). Its properties vary by gas composition, pressure, and temperature (14).

Given the growing usage of plasma treatment in the food manufacturing, the current study investigates the efficacy of cold atmospheric plasma generated using different working gases in degrading AFB1in contaminated food models. The specific objectives are to evaluate the degradation efficiency of AFB1 under varying plasma treatment conditions such as exposure time, gas type, and power, to identify the primary reactive species responsible for toxin degradation. This research aims to provide a safe, non-thermal, and environmentally friendly approach for mitigating aflatoxin contamination in food products.

2. Aflatoxin B1

Aflatoxin is one of the mutagenic, teratogenic, genotoxic, and carcinogenic mycotoxins created as a secondary metabolite by *Aspergillus, Penicillium, Fusarium,* and *Alternaria* but *Aspergillus flavus* , and *Aspergillus parasiticus*. Aflatoxin has various types (containing B1, B2, G1, G2, M1, and M2) with diverse levels of toxicity. Table 1 lists their toxicity, main side effects, and chemical structure. Among the many types of aflatoxins, types B (AFB1 and AFB2), G (AFG1 and AFG2), and M (AFM1 and AFM2) are the most abundant and prevalent in agricultural products (3). In addition, a wide range of aflatoxin-related pathologies, like malnutrition diseases, delayed physical and mental

maturity, alterations in reproduction, and nervous system diseases, have been verified in humans or animals (15). The most well-documented health effect of AFB1 is its role in hepatocellular carcinoma (HCC) (16). The International Agency for Research on Cancer (IARC) categorizes AFB1 as a Group 1 carcinogen, meaning there is adequate sign of its carcinogenicity in beings. Animal and human studies suggest that AFB1 impairs cell-mediated immunity by inhibiting T-cell function and reducing antibody production (17). In children, aflatoxin exposure has been linked to growth impairment (18). A longitudinal study in Benin and Togo found that children with higher AFB1 biomarker levels had significantly lower height-for-age scores, indicating stunted growth (19). This effect may be due to aflatoxin-induced intestinal damage and nutrient malabsorption (20).

The molecular architecture of AFB1 consists of three key components that contribute to its environmental persistence: (1) a bifuran ring system, (2) a coumarin moiety, and (3) a cyclopentanone ring. The bifuran ring, particularly the C8-C9 double bond, creates an electron-rich region that resists nucleophilic attacks and oxidative degradation (21). This structural feature is also responsible for AFB1's carcinogenic potential, as it enables the development of DNA adducts in liver cells (22). The conjugated π-electron system extending across the molecule provides exceptional stability against various environmental stresses. AFB1 demonstrates significant heat resistance, requiring temperatures above 160°C for substantial degradation (23). Conventional food processing methods such as boiling or baking (typically below 120°C) achieve less than 20% reduction in AFB1 content (24).

Chemical treatments show limited effectiveness against AFB1. While alkaline processing (nixtamalization) can cleave the lactone ring, it requires harsh conditions (>1% NaOH) that may affect food quality (25). Oxidizing agents such as hydrogen peroxide or ozone can degrade AFB1 but need prolonged exposure times (26). Physical methods like UV irradiation (254 nm) induce photodegradation but often produce toxic byproducts such as AFB2a (27). Gamma irradiation shows better efficacy, with doses exceeding 10 kGy required for 90% degradation (28). The exceptional stability of AFB1 under various environmental and processing conditions stems from its unique molecular architecture. This stability, combined with its potent toxicity, makes AFB1 one of the most challenging food contaminants to eliminate. Current research focuses on developing combination approaches (physical-chemical-biological) to effectively degrade AFB1 without generating harmful byproducts (24). So far, many methods have been used to reduce AFB1. Some of the chemical, physical and biological methods used have not had the desired effect on aflatoxin reduction or may cause adverse nutritional and organoleptic changes in food. Nowadays, the use of non-thermal treatments like UV and gamma irradiation, pulsed light treatment, and ozonation have been considered to remove aflatoxin (29). The chemical structure of AFB1 is shown in Fig. 1.

3.T ype of plasma treatment

Nowadays, non-thermal treatments are used to maintain the nutritious value and improve the microbial properties of food products. One of these treatments is plasma, the fourth state of material according to Fig. 2, plasma, often referred to as the

fourth state of matter, is an ionized gas containing reactive species such as electrons, ions, free radicals, and photons (30). Plasma treatments are increasingly being explored for food safety claims, including the degradation of mycotoxins like AFB1 (AFB1) (31). Plasma treatment is one of the non-thermal treatments that has been used for various purposes today (13). the word plasma, which was first declared in 1928, states to a large number of different species like electrons, positive and negative ions, free radicals, gas atoms, molecules in the ground or excited state, and quanta of electromagnetic radiation (photons) (14). Plasma technologies can be broadly categorized into thermal plasma and cold plasma (non-thermal plasma), which differ significantly in their mechanisms, operating conditions, and effectiveness in toxin degradation (32). Cold plasma operates at near-ambient temperatures (30–60°C), making it appropriate for heat-sensitive food produces (33). The primary advantage of cold plasma is its ability to produce reactive oxygen and nitrogen species (RONS), such as ozone (O_3) (34), hydroxyl radicals (•OH) (35), and atomic oxygen (O) (36), which interact with AFB1's molecular structure, particularly targeting the C8=C9 double bond in the furan ring, a critical position for its carcinogenicity (37). Studies have demonstrated that cold plasma can degrade 70–100% of AFB1 in various food matrices, including corn, hazelnuts, and spices, without significantly altering nutritional or sensory properties (29, 38). Thermal plasma, in contrast, operates at high temperatures (several thousand degrees Celsius) and is typically generated by arc discharges or inductively coupled plasmas (39). While highly effective in decomposing organic compounds, its extreme heat can cause thermal degradation of food components, limiting its application in food processing. Few studies have explored thermal plasma for AFB1 degradation, as the high energy input may cause adverse changes in food quality, for instance protein denaturation and lipid oxidation (40). Cold plasma is generally preferred over thermal plasma for food applications owing to its non-thermal nature and selective reactivity (41).

Research indicates that cold plasma treatments, such as atmospheric pressure cold plasma (ACP) and high-voltage cold plasma (HVCP), achieve high AFB1 degradation rates (76–100%) within short treatment times (1–30 minutes) (29, 42). The reactive species in cold plasma break AFB1 into smaller, less toxic compounds, such as $C_{16}H_{16}O_6$ and $C_{17}H_{14}O_7$, without generating harmful residue (42). In contrast, thermal plasma, while capable of complete AFB1 destruction, is impractical for most food applications due to its high energy consumption and detrimental effects on food quality (43, 44).

Plasma is divided into different types based on gas composition, pressure, and temperature. The gas used for plasma may be air or separated gases. The meaning of separated gases is the use of Ar, He, O_2, Ne, and N_2. According to the pressure used in the plasma chamber, plasma treatment is alienated into three types of vacuum plasma, atmospheric pressure plasma and high pressure plasma. Changing the working power and the way they are activated produces cold and hot plasmas, at low and high temperatures, respectively (40). Based on the production method, plasma is divided into different types. Dielectric barrier discharge (DBD), jet plasma, corona discharge, and RF plasma are plasma production methods. Plasma

treatment is used for various cases such as improving microbial properties, reducing contamination, and changing the technological and physicochemical properties of food products on a laboratory scale. As illustrated in **Fig. 3,** plasma types can be classified by thermal behavior, gas input, and production method. Latest studies have elaborated more on the antimicrobial aspect of plasma treatment in the food manufacturing. So far, plasma treatment to reduce the microbial load caused by *Escherichia coli O157:H7*, *Listeria monocytogenes*, *Staphylococcus aureus* and Fungus species like *Aspergillus flavus*, *Aspergillus Spp*, and *Penicillum* have been used for heat- sensitive products (42, 45). One of the uses of plasma treatment is to decrease the risk of toxicity of mycotoxins in food, such as AFB1. Plasma treatment is known as a successful tool for the decontamination of mycotoxins such as *Alternaria* toxin, zearalenone, fumonisin, and aflatoxin. Plasma treatment, using active species of oxygen, nitrogen, hydroxyl radicals, superoxide, hydrogen peroxide, ozone, various nitrogen oxides, and electrons, affects complex compounds such as aflatoxin (46) and causes changes in its structure and breaking bonds or to form chemical bonds and reduce the amount of compound side (47). On the other hand, it prevents the adverse changes that may be caused by thermal treatments in food (29).

4. Effect of plasma treatments on AFB1

Plasma treatment has different types, not all of which are used in the food industry, and the generation of reactive species in it depends on parameters such as voltage, electrode gap distance, type of filled gas, gas relative humidity, and mode of reaction (direct or indirect) (42). Detoxification of aflatoxin may be done by removing the part contaminated with the poison or changing the chemical structure of the poison and reducing its toxicity (48). The toxicity of AFB1 has been attributed to the double bond at C8-C9 in its chemical structure. In the liver, cytochrome P450 enzymes break down aflatoxin B1 into AFB1-8, 9-epoxide compounds, that are hydroxylated and conjugated to glutathione or gluconic acid or are attached to DNA by nucleophilic bonds (49). One of the bonds broken by plasma treatment is the C8-C9 double bond, which reduces the toxicity of AFB1 and breaks its chemical structure into less dangerous compounds (50). Because some treatments such as thermal plasma may change the nature of the produced food (51, 52). In the following, we will examine some of the most widely used plasma treatments in the food manufacturing and their use in reducing the amount of AFB1.

In cold plasma, energy is transferred from charged particles to neutral particles. The temperature of non-thermal plasma is between 30 and 60 degrees, which mostly refers to cold plasma and low-temperature plasma, and it is considered within the room temperature range. This type of plasma treatment is suitable for food products that are sensitive to high heat. It is similarly reasonable in terms of price and the amount of energy required. Cold plasma treatment is mostly produced by dielectric barrier discharges (DBD) and atmospheric pressure plasma jets (APPJ) methods (48). According to a study (29) the use of atmospheric pressure cold plasma resulted in a 100% reduction of AFB1 in corn kernels, while no negative effects were observed on the visual, sensory, nutritional, physicochemical, and technological properties of corn kernels. In another study, they motivated on the

detoxification of hazelnuts by cold plasma. Two types of atmospheric-pressure cold plasma and low-pressure cold plasma were accustomed reduce AFB1 in spiked samples. Both types of plasma treatment reduced 72-73% of AFB1 in hazelnut samples. Sensory evaluation in this study also showed that plasma treatment preserved the organoleptic properties of hazelnuts. The results showed that various types of plasma treatments can be used to reduce aflatoxin and detoxify in the food industry (53). In a study by (38), they investigated the reduction of aflatoxin by atmospheric pressure cold plasma treatment in peeled hazelnuts. The variables investigated in this study were device power, exposure time, and the type of gas used in plasma. More than 70% of AFB1 was destroyed in hazelnuts at 1000 watts of power and 12 minutes of treatment. Cold plasma was recognized as one of the approaches to reduce AFB1 in these studies. In addition, this treatment maintained the desired organoleptic characteristics of the product.

To use high voltage technology in the plasma system, non-thermal plasma is usually used to maintain the quality of the food product (45). The output voltage in this system is usually 0-120 kV (54). In (37) study high voltage cold plasma treatment was investigated on pure AFB1. Active gar species were found to be the main and important components in the inactivation and destruction of AFB1. Active oxygen and nitrogen species were the main components of plasma treatment in this study. Ozone was also one of the compounds produced by plasma treatment. Detoxification of aflatoxin by ozone gas is also common in the past. Ozone breaks down aflatoxin into two less dangerous chemical compounds (C16H16O6 and C17H14O7). The consequences of this study displayed that based on the relationship between bioactivity and chemical structure of aflatoxin, plasma treatment reduced the toxicity of this compound by breaking the C8-C9 double bond in the furfuran ring. Another method of reducing the toxicity of aflatoxin by plasma, which was mentioned in this study, is the addition of an aldehyde group, hydroxyl, or water molecule to the chemical composition of aflatoxin and reducing its toxicity. Water was added at the C8-C9 bond site. Another strategy of plasma treatment in reducing aflatoxin detoxification is oxidation and epoxidation reactions. In a different study, HepG2 cells were used to investigate the toxicity of aflatoxin after high-voltage cold plasma treatment in laboratory conditions. For this purpose, the cells infected with aflatoxin were subjected to high-voltage cold plasma treatment for 0, 2, 5, 10, and 20 minutes. The results revealed that the 20-minute plasma treatment reduced the oxidative damage of aflatoxin and its cytotoxicity. In this study, the reduction of AFB1 toxicity was determined by breaking the double bond in the C8-C9 region (50). In another study, high-voltage cold plasma treatment was used to reduce AFB1 in corn. The gas used was atmosphere and corn samples were exposed to plasma for 1, 2, 5, 10, 20, and 30 minutes. Up to 80% of AFB1 was reduced after 10 minutes of plasma treatment (42). As a form of atmospheric-pressure plasma, corona discharge occurs as a bright glowing region localized around a sharp electrode tip when placed in a highly non-uniform electric field (55, 56). In addition to its active species, corona discharge plasma jet treatment has received attention in the food industry as a result of its simplicity, reproducibility, and inexpensiveness (57). corona discharge plasma jet treatment is used at

current levels (1.0-1.5 A) and electrode distances (15-35 mm) between the electrode tip and the sample, and the time of plasma treatment is different depending on the kind of food, its sensitivity, and the type of application (58). In (57) study, plasma treatment produced by corona discharge was used to reduce AFB1 in spiked samples of rice and wheat. After 30 minutes of treatment, 45-56% of aflatoxin in the samples was reduced. The main cause of aflatoxin reduction in this study was the oxidation of aflatoxin by reactive oxygen species produced by plasma treatment. In Table 2, more information on the plasma treatments used to reduce AFB1 is reported.

5. Limitations and challenges

Plasma technology has shown considerable promise as a non-thermal method for degrading AFB1 in various food matrices (59). However, several significant limitations and challenges must be addressed before this technology can be widely adopted by the food industry. One major challenge is the variability in treatment effectiveness across different food types. Research indicates that while plasma can achieve over 90% AFB1 degradation in powdered foods like spices, its efficacy decreases substantially in high-fat products such as nuts and oilseeds (29). This variation is primarily due to differences in food composition, surface characteristics, and the interference of lipid molecules with plasma-generated reactive species (37).

Another critical limitation involves the potential impact on food quality parameters. Although considered a mild processing technology, plasma treatment can induce undesirable changes in food products (60). These include oxidative degradation of sensitive nutrients like vitamins and polyunsaturated fatty acids (61), modifications to protein structures that may affect functionality (62), and the development of off-flavors caused by lipid peroxidation (53). Such alterations could compromise the nutritional value, sensory qualities, and overall consumer acceptability of treated products, particularly for delicate food items (48).

Scaling up from laboratory research to industrial implementation involves significant technological and financial challenges (63). Current challenges include the high energy requirements for continuous plasma generation (64), significant capital investment needed for large-scale plasma systems (65), and the lack of standardized protocols for different food commodities (66). These factors currently limit the economic viability and practical implementation of plasma technology in commercial food processing operations (67).

From a regulatory perspective, several safety concerns remain unresolved and require further investigation (68). These include the potential formation of toxic degradation byproducts (69), the long-term stability of plasma-treated products (70), and possible migration of reactive species into food matrices (71). Comprehensive toxicological assessments and stability studies are needed to ensure regulatory compliance and guarantee consumer safety when implementing this technology (72).Future research should prioritize several key areas to overcome these limitations. These include developing matrix-specific treatment protocols (57), optimizing plasma parameters to minimize quality deterioration (73), designing cost-effective and scalable plasma systems (74), and conducting thorough safety evaluations of plasma-treated foods.

Table 1. summary odf different types of aflatoxins, chemical structures, toxicity, and their heaith effects.

Aflatoxin Type		Chemical Structure	Toxicity (LD_{50} in Rats, mg/kg)(oral, rat)	Key Health Effects	References (ISI/Authoritative)
Aflatoxin (AFB_1)	B_1	Difuranocoumarin + Cyclopentanone	0.5	Potent hepatocarcinogen, mutagenic, immunosuppressive	(22, 75)
Aflatoxin (AFB_2)	B_2	Hydrogenated AFB_1 (no double bond)	1.7	Less toxic than AFB_1 but still carcinogenic	(76, 77)
Aflatoxin (AFG_1)	G_1	Difuranocoumarin + Additional lactone	0.5–1	Hepatotoxic, carcinogenic (less than AFB_1)	(78)
Aflatoxin (AFG_2)	G_2	Hydrogenated AFG_1	~2	Lower toxicity than AFG_1, but harmful to liver/kidneys	(79, 80)
Aflatoxin (AFM_1)	M_1	Hydroxylated metabolite of AFB_1 (found in milk)	0.3–0.5	Hepatocarcinogen, nephrotoxic, concerns in dairy products	(81, 82)
Aflatoxin (AFM_2)	M_2	Metabolite of AFB_2	~1.5	Less toxic than AFM_1 but still hazardous	(83, 84)

Table 2. Summary of some commercial plasma equipment for reduction of AFB1.

Type of equipment	Type of gas	Time of exposure	result	reference
High Voltage Atmospheric Cold Plasma (HVACP)	Atmosphere air (78% N2, 22% O2)	1, 2, and 5 min	AFB1 was degraded by 76% using a 5 min treatment	**(37)**
Low-temperature radio frequency plasma	Not mentioned	10,15, and 20 min	Exposure to plasma for 10 min resulted in a degradation rate of up to 88.3 %	**(85)**
Nitrogen gas plasma	Nitrogen	0-30 min	AFB1 was efficiently degraded to less than one tenth of the original level within 15 min	**(70)**
Corona discharge plasma jet	Atmosphere air	5, 10, 15, 20, 25 and 30 min	The average levels of AFB1 degradation ranged between 45-56% following corona discharge plasma jet treatment for 30 min	**(57)**
High voltage atmospheric cold plasma	Atmosphere air	0, 2, 5, 10, and 20 min	20 min of HVACP treatment for AFB1 significantly reduced AFB1	**(50)**
Cold atmospheric pressure plasma	Atmosphere air	30, 60, 120, 240 and 480 s	100% removal is achieved	**(29)**
cold plasma	Atmosphere air	1.7 and 30 min	Both plasmas reduced 72–73% of AFB1	**(53)**
Microwave-induced argon plasma	Argon	0, 1, 3, 5, and 10 S	AFB1 was completely removed after 5 s of plasma treatment	**(86)**
Cold Atmospheric Plasma	Atmosphere air	1, 2, 4, and 12 min	Plasma treatment reduced over 70% of AFB1	**(38)**
High Voltage Atmospheric Cold Plasma	Atmosphere air	1, 2, 5, 10, 20, and 30 min	Aflatoxin was degraded by 62% and 82% by 1 and 10 min plasma treatment	**(42)**

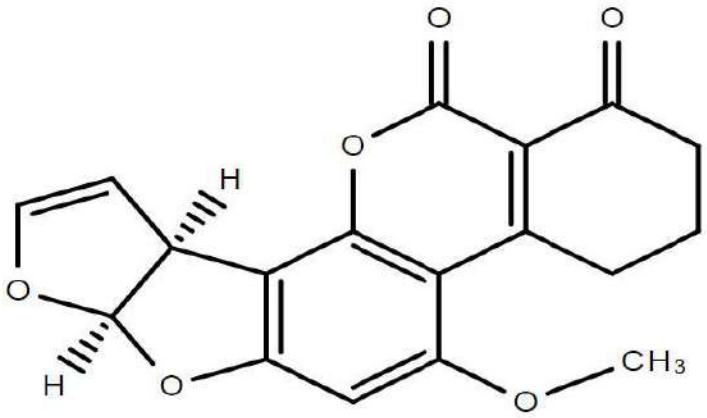

Figure 1. Chemical structure of AFB1.

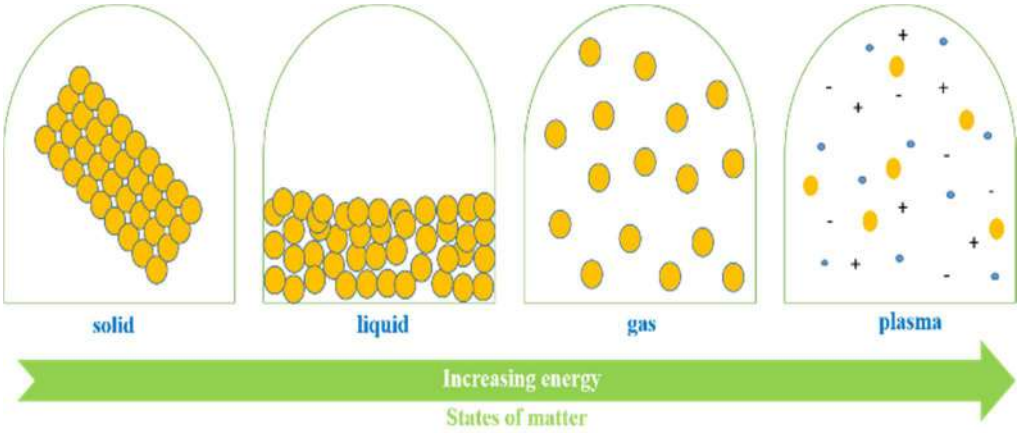

Figure 2. Schematic of state of matter from solid to plasma.

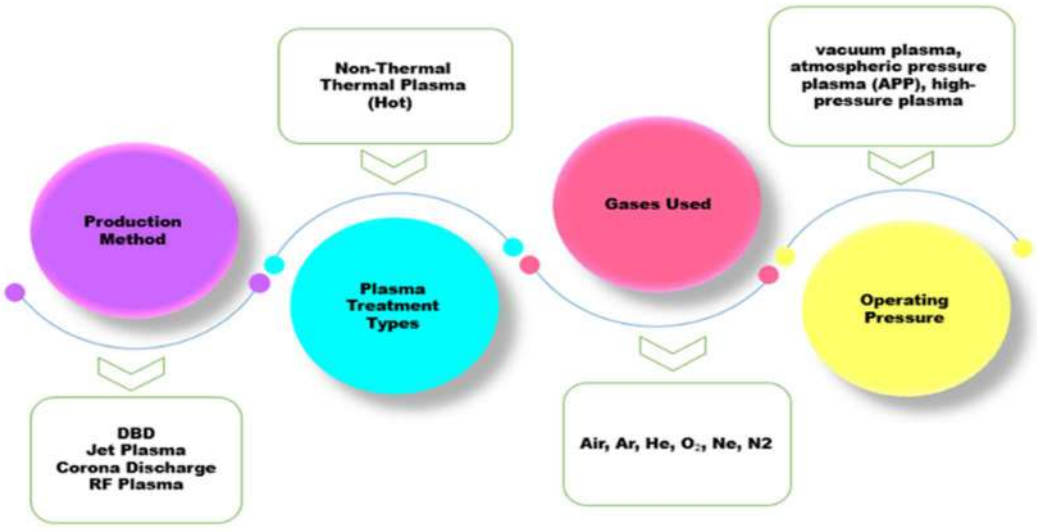

Figure 3. Overview of plasma treatment types categorized by method, gas used, and thermal nature.

Addressing these challenges through targeted research and technological innovation could facilitate the successful transition of plasma technology from experimental research to practical food safety applications in the industry (87). The ultimate goal is to develop a reliable, efficient, and economically feasible plasma-based solution for AFB1 reduction that maintains food quality while meeting all safety standards.

6. Conclusion

Cold plasma technology offers a highly effective method for degrading aflatoxin B1 (AFB1) in foods, surpassing traditional methods. Using non-thermal systems like dielectric barrier discharges, it achieves 70–100% toxin reduction while preserving food quality. The process relies on reactive oxygen/nitrogen species (RONS) that selectively break AFB1's toxic C8=C9 bond, detoxifying it without harmful residues. To realize its full potential, key challenges must be addressed. Treatment parameters need optimization for different food matrices to ensure consistent efficacy. Comprehensive safety assessments are required to evaluate any byproducts and the long-term stability of treated foods. Developing affordable, scalable systems is crucial for industrial adoption, especially in resource-limited settings. The public health impact could be transformative in high-risk regions like sub-Saharan Africa and Southeast Asia, where AFB1 contributes to liver cancer and childhood stunting. Widespread adoption could reduce these health burdens and improve economic outcomes for farmers by enabling access to international markets. As research overcomes current limitations, plasma technology may become indispensable for global food safety.

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Author contributions

Leila Zare contributed to project administration; resources; writing riginal draft. Javaneh Karimi contributed to project administration; resources; writing original draft. Ehsan Sadeghi contributed to conceptualization; data curation; resources validation; writing original draft; writing review and editing. Fatemeh Barzegar contributed to supervision; investigation; visualization; writing review and editing.

Declaration of competing interest

The authors have declared no conflicts of interest in this article.

Data availability statement

Data sharing is not applicable to this article as no new data were created or analyzed in this study.

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