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# **Cold plasma technology: does it have a place in food processing?**

<span id="page-1-7"></span><span id="page-1-6"></span>HollyBatt<sup>a</sup>  $\bullet$  and Jed W. Fahe[yb,](#page-1-1)[c,](#page-1-2)[d,](#page-1-3)[e,](#page-1-4)[f](#page-1-5)  $\bullet$ 

<span id="page-1-4"></span><span id="page-1-3"></span><span id="page-1-2"></span><span id="page-1-1"></span><span id="page-1-0"></span><sup>[a](#page-1-6)</sup>Department of International Health, Johns Hopkins Bloomberg School of Public Health, Baltimore, MD, USA; <sup>b</sup>Department of Medicine, The Johns Hopkins University S[c](#page-1-8)hool of Medicine, Baltimore, MD, USA; <sup>c</sup>Department of Psychiatry & Behavioral Sciences, The Johns Hopkins University School of Medicine, Baltimore, MD, USA; <sup>d</sup>Department of Pharmacology and Molecular Sciences, The Johns Hopkins University School of M[e](#page-1-10)dicine, Baltimore, MD, USA; <sup>e</sup>iMIND Institute, The Johns Hopkins University School o[f](#page-1-11) Medicine, Baltimore, MD, USA; <sup>f</sup>institute of Medicine, University of Maine, Orono, ME, USA

#### ABSTRACT

In recent years, there has been a growing demand for alternative food processing technologies that can improve food safety while preserving the nutritional quality of food products. Traditional thermal processing methods can lead to nutrient loss and degradation, prompting the exploration of novel approaches. Cold plasma (CP) technology, an emerging non-thermal food processing technique, has gained significant attention for its potential in the food industry. We provide herein, an introduction to CP and an overview of the technology, highlighting its potential advantages in safety, efficiency, and environmental friendliness.

#### <span id="page-1-11"></span><span id="page-1-10"></span><span id="page-1-9"></span><span id="page-1-8"></span><span id="page-1-5"></span>**KEYWORDS**

<span id="page-1-14"></span><span id="page-1-13"></span>Food safety; food processing; nutritional quality; antimicrobial; non-thermal

<span id="page-1-17"></span><span id="page-1-15"></span>Cold plasma (CP) technology offers a promising solution to enhance food safety without compromising the nutrient profile of food products (Ekezie et al. [2017](#page-10-0); Zhang et al. [2018;](#page-11-0) Rao et al. [2023\)](#page-11-1). The "cold plasma" is generated through the ionization of gas at either atmospheric or low pressure, resulting in a non-equilibrium state with high electron temperature and low ion temperature (Zhang et al. [2018](#page-11-0)). This unique characteristic of CP allows for the effective inactivation of pathogenic microorganisms on the surface of low-moisture foods, detoxification of mycotoxin-contaminated low-moisture foods, and extension of shelf life of ready-toeat foods, while minimizing the impact on food quality (Ekezie et al. [2017](#page-10-0); Zhang et al. [2018](#page-11-0)). Applications to other industries are manifold and have been reviewed elsewhere (Akishev et al. 2008;Matthes et al. [2013\)](#page-10-1), but we shall concentrate herein on food-based applications of CP.

<span id="page-1-20"></span><span id="page-1-16"></span>Compared to traditional thermal processing methods, CP technology has several advantages. CP is *environmentally safe* and does not produce long-lived toxic by-products (Yepez et al. [2022\)](#page-11-2). The minimal modifications observed in treated food products ensure that the *nutritional quality and sensory attributes remain largely intact* and of paramount interest to food processors. CP technology offers *efficiency* in food processing, with the ability to rapidly inactivate microorganisms on food surfaces (Rao et al. [2023;](#page-11-1) Thirumdas [2015](#page-11-3)).

Although CP technology holds great potential, there are challenges that need to be addressed if it is to be more widely implemented in the food industry. Factors such as cost-effectiveness, scalability, and regulatory considerations need to be carefully evaluated. Future trends in CP technology involve the development of portable devices, integration with other food processing technologies, and exploration of <span id="page-1-21"></span>novel applications (Thirumdas [2015;](#page-11-3) Ekezie et al. [2017;](#page-10-0) Zhang et al. [2018;](#page-11-0) Birania et al. [2022](#page-9-0); Rao et al. [2023](#page-11-1)).

# **Principles of CP generation**

<span id="page-1-18"></span>Plasma – the physico-chemical description as opposed to the biological definition – is characterized by ionized gas containing various active species, such as electrons, free radicals, and ions (Scholtz et al. [2015](#page-11-4); Dasan et al. [2017](#page-10-2); Zhang et al. [2018](#page-11-0)). This ionized gas can exist in either a ground or an excited state, with an overall neutral charge. There are two main categories of plasma: thermal and non-thermal plasma. Thermal plasma requires high-pressure levels (≥105Pa) and substantial power (up to 50MW) for its propagation (Scholtz et al. [2015](#page-11-4); Zhang et al. [2018\)](#page-11-0). On the other hand, non-thermal plasma is produced at lower pressures and power levels, without localized thermodynamic equilibrium, and is designated as non-equilibrium plasma (Scholtz et al. [2015](#page-11-4); Dasan et al. [2017;](#page-10-2) Zhang et al. [2018\)](#page-11-0).

<span id="page-1-19"></span><span id="page-1-12"></span>Various types of nonthermal plasma sources exist, each producing different quantities of reactive species. These sources include corona discharge, dielectric barrier discharge (DBD), microwave discharge, and a unique configuration known as a plasma jet (Scholtz et al. [2015](#page-11-4); Dasan et al. [2017](#page-10-3); Rao et al. [2023\)](#page-11-1) ([Figure 1\)](#page-2-0). Corona discharge involves the creation of a plasma region through the ionization of air molecules around sharp electrodes in a high electric field (Scholtz et al. [2015\)](#page-11-4). This results in the formation of ions and electrons in the vicinity of the electrode. Dielectric Barrier Discharge (DBD) operates between electrodes separated by a dielectric material, like glass or plastic.



<span id="page-2-0"></span>**[Figure 1.](#page-1-12)** Schematic overview of main methodologies used for atmospheric pressure cold plasma delivery that are relevant for food processing.

The discharge is limited to the electrode vicinity due to dielectric properties, wherein electric induction and polarization cause ionization. The extent of DBD's corona discharge depends on the applied current and dielectric properties (Scholtz et al. [2015](#page-11-4); Rao et al. [2023\)](#page-11-1). In microwave discharges, electromagnetic waves with frequencies exceeding hundreds of MHz ionize gas to create plasma. This process occurs within a controlled chamber resonating with the microwave frequencies. The resulting non-thermal plasma finds applications in medical fields due to its unique characteristics (Scholtz et al. [2015\)](#page-11-4). Plasma jets are simple, maintainable atmospheric pressure plasma sources. They create a directed stream of charged particles and excited species by using an auxiliary gas flow. This reactive "plasma jet" is precise and powerful, making it useful for tasks like surface treatment and medical procedures. It's referred to as plasma pen, plasma torch, or plasma needle, highlighting its focused particle delivery capability (Scholtz et al. [2015;](#page-11-4) Rao et al. [2023\)](#page-11-1).

The parameters that influence plasma generation include gas composition, electric current voltage and frequency, and treatment time. Gas composition plays a crucial role in determining the type and concentration of reactive species generated in the plasma (Rao et al. [2023](#page-11-1)). A variety of gasses and mixes, including air, nitrogen, argon, and helium, can be used, each with its specific characteristics (Ekezie et al. [2017;](#page-10-0) Rao et al. [2023\)](#page-11-1). Voltage and frequency govern the energy delivered to the system and, thus, the characteristics of the plasma. Higher voltages and frequencies generally result in increased plasma density and greater generation of reactive species. Treatment time—duration of exposure to CP—can be adjusted based on the desired effects and on the characteristics of the food product being treated (Rao et al. [2023\)](#page-11-1).

In the context of the food industry, non-thermal plasma induced by electrical discharges is particularly relevant due to its potential for processing foods at low temperatures. The specific method employed for cold plasma generation determines the application trajectory, as well as the magnitude and composition of reactive species. Understanding the technological pathways for plasma generation, including those induced by atmospheric pressure and those operating under reduced pressure, is crucial in leveraging the potential of cold plasma technology in the food industry.

# **Mechanism of action**

CP's primary action with food involves generating reactive species, particularly reactive oxygen species (ROS) and reactive nitrogen species (RNS). These reactive species are pivotal for CP's antimicrobial and functional effects (Ekezie et al. [2017;](#page-10-0) Zhang et al. [2018;](#page-11-0) Birania et al. [2022](#page-9-0)).

CP proves effective in rapidly reducing harmful bacteria such as *Escherichia coli*, *Salmonella* spp., and *Listeria* spp. The outcome of CP treatment depends on factors such as product type, bacteria, food surface, and plasma behavior. However, fully comprehending CP's mechanism of action is intricate due to the interplay between plasma species and microorganisms (Zhang et al. [2018](#page-11-0); Rao et al. [2023](#page-11-1)).

During CP treatment, ionized gasses create charged particles, and reactive oxygen species (hydrogen peroxide  $(H_2O_2)$ , ozone  $(O_3)$ , and superoxide radicals  $(O_2$ <sup>-</sup>)) are produced, that disable microbes (Scholtz et al. [2015;](#page-11-4) Zhang et al. [2018](#page-11-0); Rao et al. [2023](#page-11-1)). Plasma-generated reactive agents target lipids and sugars on microbial cell surfaces, causing membrane rupture. Reactive oxygen and nitrogen species also interact with peptidoglycan in cell walls. As C–O and C–N bonds break, the entire bacterial cell structure is compromised (Rao et al. [2023\)](#page-11-1). Additionally, these agents impact proteins, DNA, and lipids within cells, leading to oxidation, lipid damage, and DNA fragmentation. Other studies propose that electrostatic forces damage cell membranes, culminating in cell death. Intense electric fields can even change protein structures, causing them to separate from cell membranes and potentially creating membrane perforations (Scholtz et al. [2015](#page-11-4); Rao et al. [2023\)](#page-11-1). These interactions between reactive species are not limited to the bacteria. These interactions affect the food components, influencing enzymatic reactions and potentially altering sensory and nutritional qualities of food. The alterations may be undesirable for food products. Thus, the implementation of cold plasma technology for food decontamination warrants a nuanced consideration of the target surface area. Notably, even if the cell surfaces of the food substrate undergo alterations during this process, it is imperative to emphasize that these cellular constituents are nonviable, thereby mitigating any safety concerns. Furthermore, the volumetric disparity between the food matrix and potential surface contaminants tends to render any surface-associated modifications negligible in comparison.

It is pertinent to discern that the ramifications of cold plasma treatment are contingent upon the nature of the substrate, differentiating between its application on solid substrates such as seeds or spices, where exposure remains localized to the superficial layers, and its use in the decontamination of liquid products like juices. In the latter case, comprehensive exposure across the entirety of the product is requisite to achieve the desired decontamination effect (Misra et al. [2016](#page-10-4)).

#### <span id="page-3-7"></span>**Cold plasma effects on food products**

# *Bacteria and virus inactivation*

Current research on cold plasma (CP) treatment for microbial inactivation focuses on identifying the most effective plasma regimes, determining minimal plasma power densities for appropriate microorganism reduction, and understanding the physical processes (Scholtz et al. [2015\)](#page-11-4). The efficiency of microbial inactivation depends on various factors, including the surface characteristics of the treated produce, the type of plasma device used, the gas composition, and the mode of exposure (Ziuzina et al. [2014;](#page-11-5) Ma et al. [2015;](#page-10-5) Scholtz et al. [2015;](#page-11-4) Rao et al. [2023](#page-11-1)). For instance, produce items with uneven surfaces may require more time for the complete destruction of microbes compared to smoother surfaces (Sani et al. [2023](#page-11-6)).

<span id="page-3-17"></span><span id="page-3-14"></span><span id="page-3-13"></span>One study examined the sensitivity of nine microbial species, encompassing Gram-positive and Gram-negative bacteria, *Deinococcus radiodurans* and *Geobacillus stearothermophilus* spores, and the yeast *Candida albicans*. The sensitivity of bacterial vegetative forms was relatively similar across all bacterial species, however, inactivation of *Candida* and *Geobacillus* spores necessitated longer exposure times (Scholtz et al. [2010](#page-11-7)). A separate study found nitrogen-based cold plasma from a plasma jet from the product at a distance of 6cm to effectively reduce *Salmonella* and *Escherichia coli* on almonds (Niemiri et al. [2012](#page-10-6)). Others found a similar reduction in *Salmonella* when applying a microwave-powered CP system using nitrogen-cold plasma at 900W and 667Pa for 0, 2, 5, 10, and 20min to radishes (Oh et al. [2017\)](#page-11-8). Overall, the sensitivity of vegetative forms of planktonic (non-biofilm-contained) bacteria varied somewhat across species but was notably greater than that of spores or biofilm forms, which in general, required much longer exposure times for inactivation (Scholtz et al. [2015](#page-11-4)).

<span id="page-3-16"></span><span id="page-3-10"></span><span id="page-3-8"></span>Studies have found CP to be a lethal tool in inactivating viruses. Atmospheric pressure CP has been found to be a sufficient treatment for the disinfection of non-enveloped virus-contaminated surfaces (Yasuda et al. [2010\)](#page-11-9). One review examining the effects of CP on milk found CP to effectively reduce the pathogens of concern but did so without the risk of the microorganisms developing resistance (Rathod et al. [2021\)](#page-11-10).

# <span id="page-3-12"></span>*Fungal inactivation*

<span id="page-3-0"></span>Studies have explored the effectiveness of CP on different fungal species (Akishev et al. [2008\)](#page-9-1). The inactivation process varies among fungal species, and exposure times range from 30 s to 30min, depending on the microorganism and plasma conditions. However, the fungicidal effect of CP is generally weaker than its bactericidal effect, and further research is needed to fully understand the mechanisms of fungal inactivation and to optimize the plasma conditions for different fungal species (Scholtz et al. [2015](#page-11-11)).

# *Biofilms*

<span id="page-3-9"></span><span id="page-3-2"></span>CP treatment has proven to be highly effective at mitigating the proliferation of biofilms (Niemira et al. [2018,](#page-10-7) 2023; Hage et al. [2022](#page-10-8)). The pathogenic bacteria that create biofilms are highly resilient and can pose challenges for conventional sanitization methods. Numerous studies have underscored the efficacy of cold plasma treatment in reducing the biofilms formed by pathogens such as *Pseudomonas aeruginosa*, *Staphylococcus epidermidis*, *Escherichia coli* O157:H7, and *Streptococcus mutans* (Niermira et al. 2023; Matthes et al. [2013](#page-10-9)). Other research has demonstrated the ability of cold plasma treatment to completely eradicate *Staphylococcus aureus* and *Pseudomonas aeruginosa* biofilms, further highlighting its potential in the battle against biofilm-associated bacterial threats (Vandervoort and Brelles-Mariño [2014\)](#page-11-12).

# <span id="page-3-15"></span><span id="page-3-5"></span>*Sensory attributes*

<span id="page-3-3"></span><span id="page-3-1"></span>The sensory attributes of food, including color, aroma, taste, and texture, play a crucial role in consumers' food choices. Several studies have investigated the effects of cold plasma (CP) treatment on sensory quality, and they suggest that CP has minimal impact on these attributes. Basaran et al. [\(2008\)](#page-9-2) observed that treating nuts with air and sulfur hexafluoride plasma for 20min did not significantly alter the surface morphology or sensory properties compared to the control group (Basaran et al. [2008](#page-9-3)). Similarly, peanuts treated with an atmospheric pressure plasma jet had an overall appearance, likeability, taste, and texture that was not significantly altered by the use of CP treatment (Iqdiam et al. [2020](#page-10-10)). While some studies reported no differences in color, Pasquali et al. [\(2016\)](#page-11-13) and Lacombe et al. [\(2017](#page-10-11)) reported improved appearance of the foods they treated with CP (Pasquali et al. [2016](#page-11-14); Lacombe et al. [2017\)](#page-10-11). Lacombe et al. [\(2017](#page-10-11)) did report a slight change in the hardness (as a proxy for texture in their study) of blueberries, but Misra et al. [\(2014\)](#page-10-12) found the texture of their CP-treated cherry tomatoes to be unchanged after the application of the CP (Misra et al. [2014;](#page-10-12) Lacombe et al. [2017\)](#page-10-13).

#### <span id="page-3-11"></span><span id="page-3-6"></span><span id="page-3-4"></span>**Nutritional content**

Studies have assessed changes in vitamins, minerals, antioxidants, and other bioactive compounds of CP treated food products. Overall, CP treatment has demonstrated the ability to retain the nutritional integrity of food products. Researchers reported minimal or negligible changes in vitamins, minerals, antioxidants, and other bioactive compounds following CP treatment (Zhang et al. [2018](#page-11-0)). This suggests that CP technology can effectively extend shelf life without

compromising the nutritional value of perishable foods (Zhang et al. [2018;](#page-11-0) Rao et al. [2023;](#page-11-1) Sani et al. [2023](#page-11-15)). In some cases, there have been reports of increased food product antioxidant activity and enhanced retention of vitamins (Zhang et al. [2018](#page-11-0); Rao et al. [2023](#page-11-1)).

## *Impact on lipids*

<span id="page-4-1"></span>Lipid oxidation is a parameter that is crucial to food shelf life, as it can lead to the formation of detrimental primary and secondary oxidation products which negatively affect food quality (Rao et al. [2023\)](#page-11-1). Studies of the impact of cold plasma (CP) treatment on wheat flour showed no significant differences in the non-polar and glycolipid fractions of total extractable lipids. However, the levels of free fatty acids and phospholipids in wheat flour were significantly affected by CP treatment (Bahrami et al. [2016](#page-9-4)). This lipid oxidation can adversely affect the sensory properties of treated foods. However, it is important to note that the effect of CP on food lipids is not uniformly negative (Rao et al. [2023](#page-11-1)). Some studies found no significant differences in either hydroperoxides (oxidation by-products) or thiobarbituric acid reactive substances (TBARS) levels (which are used as an index of lipid oxidation in foodstuffs and elsewhere) between plasma-treated and control groups. This lack of differences was taken to indicate that by optimizing various plasma process parameters, such as modified gas mixtures with reduced oxygen levels, low input power, decreased treatment temperatures, and shorter processing times, lipid oxidation can be mitigated or down-regulated (Gavahian et al. [2018](#page-10-14)).

# <span id="page-4-4"></span>*Impact on proteins*

<span id="page-4-5"></span>In the quest for sustainable and environmentally friendly protein production, alternative protein sources, particularly plant and insect proteins, have become of interest to consumers. CP treatment provides a green approach to enhance the physical and chemical properties of plant proteins (Rao et al. [2023](#page-11-1)). When CP interacts with proteins, it generates reactive oxygen species that can lead to the derivatization of some of the component amino acids, creating active sites and enhancing the addition of hydrophilic groups to those proteins (Juan et al. [2021\)](#page-10-15). Furthermore, CP treatment holds the capacity to enhance or disrupt the functional and nutritional attributes of proteins by modifying their secondary, tertiary, and quaternary structures. This modification involves protein denaturation triggered by exposure to reactive species, leading to an augmented availability of amino acids. This denaturation process ultimately renders the protein structure more susceptible to hydrolytic enzymes, thereby boosting protein digestibility (Bu et al. [2023](#page-10-16))

#### <span id="page-4-2"></span>*Impact on carbohydrates*

CP treatment interacts with the structural components of plants in a process that involves the interaction of reactive species generated by plasma with sugar molecules. This leads to de-polymerization and cross-linking of starch, thus

<span id="page-4-9"></span><span id="page-4-7"></span>influencing the overall composition and properties of carbohydrates in food products (Thirumdas et al. [2015](#page-11-3)). Research on cashew apple juice demonstrated that CP treatment led to the degradation of both reducing and non-reducing sugars (i.e. fructose or glucose and non-reducing sucrose). However, extended exposure to CP resulted in an increase in sucrose due to the breakdown of oligosaccharides within fruit juices (Rodríguez et al. [2017](#page-11-16)). CP's influence on polysaccharides has been primarily observed in starch-containing legume and grain products. Through the generation of reactive plasma species, CP treatment causes surface etching and increases water binding sites by disrupting the starch. Consequently, this disruption can reduce the cooking time of brown rice (Thirumdas et al. [2015](#page-11-3); Liu et al. [2021](#page-10-17)). Research also highlighted the finding that CP could extend the storage time of fresh pasta by removing excess moisture in the product (Chen et al. [2020](#page-10-18)). CP effects on fiber (both soluble and insoluble) have not, to our knowledge, been rigorously evaluated in foods. Nor have the effects of CP on the lignin and cellulose structural backbones of plants been reported on, though we can presume that effects on crunchiness, mouthfeel, hardness and other related sensory parameters could possibly be linked to the effects of CP on these plant structural components.

# <span id="page-4-3"></span>**Bioactives**

#### *Phenolic compounds*

<span id="page-4-6"></span>Phenolic compounds have been extensively studied for their antioxidant capabilities. These compounds exert their effects through mechanisms including free radical scavenging, singlet-oxygen quenching, and metal chelation (Kumar and Goel [2019](#page-10-19)). The impact of cold plasma treatment on phenolic composition has yielded mixed results.

#### *Flavonoids*

<span id="page-4-10"></span><span id="page-4-8"></span><span id="page-4-0"></span>Cold plasma treatment has demonstrated varying effects on flavonoids across different foods. CP treatment increased detectable flavonoids in blueberries, this increase was attributed to the activation of specific metabolic pathways and the release of flavonoids bound to membranes (Sarangapani et al. [2017](#page-11-17)). Conversely, in foods like orange juice, marasca cherry, and grape juice, measurable flavonoid levels decreased after plasma treatment (Almeida et al. [2015;](#page-9-5) Garofulić et al. 2015; Pankaj et al. [2017\)](#page-11-18). Factors such as prolonged processing times and higher flow rates amplified the reduction of flavonoids. The transition of flavonoids from membrane-bound to free forms led to increased susceptibility to scavenging and damage by reactive species during processing, ultimately resulting in diminished flavonoid content. This complex scenario raises intriguing questions about the relative benefits of matrix-bound versus free flavonoids for human health, highlighting the challenges in assessing their impact through existing analytical and preclinical techniques (Almeida et al. [2015;](#page-9-5) Pankaj et al. [2017\)](#page-11-18).

# *Antioxidants*

<span id="page-5-4"></span><span id="page-5-3"></span><span id="page-5-1"></span>Antioxidants present in fruits and vegetables are pivotal in safeguarding lipids and preserving food quality by curbing-free radicals and thwarting oxidation. These bioactive components play a vital role in neutralizing-free radicals, thereby reducing the risk of diseases linked to oxidative stress (Rahaman et al. [2023](#page-11-19)). Thus, applying CP as a pretreatment on the extraction and upregulation of bioactives in plant foods has recently been framed as a positive effect (de Araújo Bezerra et al. [2023](#page-10-20)). A recent review assessed the effect of CP on phenolic compounds in liquids such as white grape juice, apple juice, tomato juice, and chocolate milk. Across these studies increased exposure time, power intensity, and flow rate decreased the number of phenolic compounds in the food products (Sruthi et al. [2022](#page-11-20)). The information to date contains anomalies that have yet to be explained based on mechanism. For example, CP generates high-energy electrons, which, under the right conditions, can lead to the production of ozone and other reactive species during treatment. Ozone can attack the aromatic rings of polyphenols, causing their breakdown and degradation (Almeida et al. [2015](#page-9-6)). Despite this, some fruit juices (blueberry, pomegranate, and cashew apple) showed an increase in the activation of enzymes involved in the synthesis of polyphenols (Gavahian et al. [2018\)](#page-10-21). Interestingly, some foods like radish sprouts showed minimal alterations in antioxidant activity even after undergoing mild plasma treatment (Pankaj et al. [2017\)](#page-11-21).

#### *Case study: spirulina*

<span id="page-5-0"></span>Beyrer et al. [\(2020](#page-9-7)) examined the effects of CP treatment on Spirulina powder (generally a mix of *Arthrospira platensis* and *A. maxima*) with a focus on two main aspects: (1) the inactivation of contaminating microorganisms, particularly *B. subtilis* spores, and (2) the impact on bioactive compounds such as polyphenols and antioxidants in Spirulina. They found that treatment with nitrogen plasma led to a significant increase in the total phenolic content (TPC) and Trolox equivalent antioxidant capacity (TEAC; a controversial metric for overall antioxidant potency) values. That increase was in linear correlation with the discharge power. On the other hand, treatment with air plasma caused a slight but non-significant change in TPC and a decrease in TEAC values, indicating a degradation of antioxidant compounds. Both chlorophyll-a and carotenoids in Spirulina were affected by the air plasma treatment, reducing the concentration of chlorophyll-a more significantly compared to carotenes/xanthophylls (Beyrer et al. [2020](#page-9-7)).

The efficacy of *Bacillus subtilis* spore inactivation was lower when those spores had been added to previously dried Spirulina powder compared to direct exposure of the spores used in inoculating that powder matrix. However, increasing the discharge power or treatment time with plasma led to greater spore inactivation. For instance, air plasma achieved about a 2-log reduction in spore count after just 1min at a low discharge power density of 15mW/cm<sup>2</sup>, while nitrogen plasma required 5min of exposure to achieve a similar level of inactivation (Beyrer et al. [2020\)](#page-9-7).

Concerning the behavior of bioactive compounds in plasma-treated materials, including Spirulina, the results varied depending on the experimental conditions. Short plasma treatments generally resulted in an increase in polyphenols and other antioxidants, attributed to the liberation of covalently bound antioxidant polyphenols, cell membrane disintegration, and depolymerization of polymeric polyphenols. However, longer or more powerful plasma treatments led to a decrease in these compounds.

Short, mild plasma treatments are likely to enhance product shelf-life by liberating covalently bound antioxidant polyphenols and increasing the availability of these compounds. This, in turn, can contribute to improved antioxidant activity, potentially delaying oxidative processes during storage. However, longer or more intense plasma treatments may have contrasting effects, as they could lead to a reduction in bioactive compounds. This decline could potentially compromise the material's antioxidant properties, making it more susceptible to oxidation over time.

In considering these speculative effects on shelf-life, it is reasonable to infer that the overall outcome may depend on a delicate balance. The ideal plasma treatment duration and intensity should be optimized to preserve or enhance the bioactive compounds while avoiding any excessive depletion, which might otherwise accelerate product deterioration. It would be beneficial for future research to delve deeper into the specific impacts of varying plasma conditions on Spirulina and similar materials to provide more concrete insights into product shelf-life and oxidation kinetics during storage (Beyrer et al. [2020\)](#page-9-8).

## *Case study: wheat and barley grains*

<span id="page-5-2"></span>Los et al. [\(2018](#page-10-22)) conducted a study to investigate how Atmospheric Cold Plasma (ACP), generated through high-voltage Dielectric Barrier Discharge (DBD), impacts microbial populations and grain quality in wheat and barley cereals. The researchers applied ACP directly to barley grains for 20min and allowed them to stand for 24h. This resulted in a significant reduction in bacterial counts (2.4  $log_{10}$  units) and fungal counts  $(2.1 \log_{10} \text{ units})$ . Under the same conditions, wheat grains exhibited notable reductions in bacterial counts (1.5  $log_{10}$  units) and fungal counts (2.5  $log_{10}$  units). The researchers subsequently inoculated the cereal grains with nonnative microorganisms. Of the nonnative microorganisms, *Bacillus atrophaeus* endospores displayed the highest resistance to ACP treatment. Conversely, the treatment significantly decreased *E. coli*, *B. atrophaeus* vegetative cells, and *P. verrucosum* spores. In general, the microorganisms introduced via inoculation proved more vulnerable to ACP compared to the naturally occurring microbiota.

The investigators also explored ACP's effects on seed germination rates. ACP application for 5min did not affect germination rates, whereas a 20-min direct treatment resulted in decreased germination rates for wheat grains. Using indirect plasma treatment for 5min followed by a 2-h retention period at 15°C resulted in the highest germination rate of 80%.

They assessed ACP treatment's effect on the physical characteristics of grain surfaces. Alterations in apparent

contact angles and free surface energy of these liquids pointed to potential modifications in grain surface characteristics due to the plasma treatment. The researchers assessed the contact angles for liquids like deionized water, ethylene glycol, and diiodomethane. The application of direct ACP treatment led to decreased contact angles with tested liquids. This indicates that treated grain surfaces became more receptive to liquid interactions (Los et al. [2018](#page-10-23)).

# **Applications in the food industry**

While some CP techniques for food decontamination have been scaled for batch manufacturing, there are limitations to the fundamental knowledge on the inactivation mechanisms with these technologies on a large scale (Thirmudas et al. 2015; Zhang et al. [2018](#page-11-0); Rao et al. [2023\)](#page-11-1). Applications of cold plasma (CP) in the food industry primarily focus on the inactivation of microorganisms present on the surface of solid foods (Birania et al. [2022](#page-9-0)). CP treatment provides targeted and precise food processing, allowing for selective action on specific food components without causing significant damage to other desirable attributes. This approach effectively decontaminates the surface and inactivates pathogens while minimizing modifications to the overall product (Rao et al. [2023;](#page-11-1) Thirmudas et al. [2015\)](#page-11-3). As a result, CP contributes to enhancing food safety, extending shelf life, and improving sensory attributes. For instance, studies have shown that CP treatment can significantly reduce *Salmonella* spp. on strawberries and effectively decontaminate cherry tomatoes containing harmful bacteria like *Escherichia coli*, *Salmonella typhimurium*, and *Listeria monocytogenes* (Ziuzina et al. [2014\)](#page-11-22)

# **Challenges and future trends**

<span id="page-6-0"></span>The implementation of CP technology in the food industry is accompanied by various challenges and limitations that require attention. One of the key challenges is the cost-effectiveness and scalability of CP technology. The equipment and infrastructure needed for CP generation can be relatively expensive, and scaling up CP systems for industrial production may present logistical and economic hurdles (Thirmudas et al. [2015](#page-11-3); Birania et al. [2022](#page-9-0); Rao et al. [2023](#page-11-1)). Even the number of academic or not-for-profit laboratories engaged in such research worldwide is quite limited ([Table](#page-7-0) [1\)](#page-7-0). Research and development efforts are necessary to optimize the technology for cost-effective implementation on a larger scale. Additionally, the regulatory landscape surrounding CP technology in the food industry is still evolving, necessitating the establishment of guidelines and regulations to govern its usage in different food applications to ensure compliance and widespread adoption (Birania et al. [2022](#page-9-0)). Furthermore, the design of CP equipment and its operational parameters play a critical role in achieving consistent and effective treatment outcomes (Thirmudas et al. [2015;](#page-11-23) Birania et al. [2022](#page-9-0); Rao et al. [2023\)](#page-11-1). Advancements in equipment design, such as developing portable and user-friendly devices, will aid in the practical integration of CP

technology into various food processing settings, benefiting the food industry stakeholders. Moreover, exploring the integration of CP technology with other food processing techniques, such as high-pressure processing, ultraviolet light, or modified atmosphere packaging, holds great potential for enhancing overall processing efficiency and product quality. Synergistic effects can be achieved by combining CP treatment with other technologies, which may further extend the shelf life and safety of food products.

#### **Consumer acceptance**

<span id="page-6-1"></span>Another hurdle may be the acceptance of CP treated foods by consumers. While CP offers advantages in maintaining food safety, nutritional quality, and sensory attributes, consumers often approach new technologies with skepticism, especially when unfamiliar terms or processes are involved (Birania et al. [2022;](#page-9-0) dos Santos Rocha et al. [2022](#page-10-24); Ekezie et al. [2017](#page-10-0); Sruthi et al. [2022\)](#page-11-24). Concerns about food safety, naturalness, and potential health impacts could hinder initial acceptance of CP-treated foods. However, emphasizing the benefits of CP, such as its non-thermal nature, minimal impact on sensory qualities, and "clean label" potential, along with effective communication and education strategies, could help improve consumer trust and acceptance. Clear labeling and transparent information about the safety and advantages of CP can play a crucial role in positively shaping consumer perceptions and increasing market adoption (dos Santos Rocha et al. [2022](#page-10-25)).

#### **Sustainability and economic advantages**

CP technology presents several sustainability benefits in food production, primarily by reducing resource use and environmental impact. CP systems operate at or near ambient temperatures  $(30^{\circ}C - 60^{\circ}C)$ , making them energy-efficient compared to thermal processing methods. The use of CP may reduce energy consumption up to 50% compared to thermal treatments for similar microbial inactivation levels (Farooq et al. [2023\)](#page-10-26). Lower energy demands may translate to a smaller carbon footprint for food processing facilities. CP's ability to utilize reactive species generated from air or other gases reduces the need for chemical sanitizers and preservatives, minimizing the environmental impact associated with chemical production, usage, and disposal (Rao et al. [2023\)](#page-11-1).

<span id="page-6-2"></span>From an economic standpoint, CP technology offers several cost-saving benefits. One of the most significant advantages is the extension of food product shelf life by effectively inactivating spoilage microorganisms. This longer shelf life reduces food waste throughout the supply chain, generating substantial cost savings for producers, retailers, and consumers (Birania et al. [2022](#page-9-0); Rao et al. [2023](#page-11-1)).

As consumer demand for minimally processed foods with fewer additives grows, cold plasma-treated products can be marketed as "clean label" or "naturally preserved," potentially commanding premium prices and enhancing market differentiation. The effective pathogen inactivation provided by cold plasma can also reduce the risk of foodborne illness

<span id="page-7-0"></span>**[Table 1.](#page-6-0)** Studies highlighting separate groups' primary research on CP technologies.

Author	Year	Plasma type	Purpose	Findings
Akishev et al.	2008	Plasma jets	Reduction of fungi.	Effectively destroyed microorganisms and biofilms (Escherichia coli,
				Serratia marcescens, Bacillus subtilis, Mycobacterium flavescens,
Almeida et al.	2015	Atmospheric cold plasma	Test effects of CP on bioactive elements in juices.	Candida lypolitica and Aspergillus niger). ACP did not affect the quality of the phenolic compounds, antioxidants, or the color of treated prebiotic orange juice.
Basaran et al.	2008	Low pressure cold plasma	Reduction of pathogens on almonds.	LPCP was more effective on A. parasiticus early in the treatment, with SF6 plasma reducing the fungal population by 5-log in 10 min and air gasses plasma reducing total aflatoxins by 50% in 20 min, demonstrating the potential of plasma treatments for decontaminating aflatoxin-producing fungi in nuts.
Bu et al.	2023	Cold atmospheric plasma Atmospheric pressure plasma jet Two-dimension dielectric Barrier discharge, and nanosecond pulsed Discharge	Test CP's effect on the functional and nutritional attributes of protein structures.	Protein denaturation from exposure to reactive enhanced protein digestibility by modifying secondary, tertiary, and quaternary structures, increasing amino acid availability. 2D-DBD treatment minimally impacted amino acid composition
Garofulic et al.	2015	gas phase plasma	elements in juices.	Cold atmospheric pressure Test effects of CP on bioactive Short CP treatment of larger juice volume resulted in the highest concentrations of anthocyanins and phenolic acids. Plasma-treated sour cherry juice, under optimized conditions, contained more phenolic compounds compared to pasteurized and untreated juice.
Iqdiam et al.	2020	Atmospheric pressure plasma jet	Test CP's effect on the sensory effects and reduction of pathogens on peanuts.	APPJ did not change the overall appearance, likeability, taste, and texture of the peanuts, while effectively reducing Aspergillus flavus.
Lacombe et al.	2017	Atmospheric cold plasma	Reduction of pathogens on blueberries.	ACP significantly reduced both Tulane virus and murine norovirus.
Liu et al.	2020	Cold plasma treatment, helium plasma	Test CP's effect on the functional and nutritional attributes of rice.	CP decreased the cooking time, rice texture, and stability of the protein network while decreasing rice dough development, starch gelatinization, and starch breakdown.
Los et al.	2018	High voltage dielectric barrier discharge	Reduction of pathogens on grains.	HVACP treatment significantly reduced the microbial load of wheat and barley grains. Inoculated microorganisms were more susceptible to the CP than the naturally occurring microorganisms. HVACP did not negatively affect the quality of the grains.
Misra et al.	2014	Dielectric barrier discharge Test CP's effect on the	functional and sensory attributes of cherry tomatoes.	DBD resulted in insignificant changes in weight, pH, firmess, and color in treated cherry tomatoes.
Oh et al.	2017	Nitrogen cold plasma	Reduction of pathogens on radish sprouts.	NCP inhibited S. typhimurium on radish sprout surfaces and inhibited growth while in storage. The sensory qualities of radish sprouts were not affected by NCP.
Pasquali et al.	2016	Atmospheric cold plasma (dielectric barrier discharge)	Reduction of pathogens on vegetable leaves.	DBD reduced E. coli on radicchio and Listeria monocytogenes. There were not significant changes to the antioxidant activity, with insignificant changes to the quality attributes
Pankaj et al.	2017	High voltage dielectric barrier discharge	Assess the reduction of pathogens in grape juice.	HVACP reduced Saccharomyces cerevisiae with no significant alterations to pH, acidity and electrical conductivity of grape juice. There was a reduction in decrease in total phenolics, total flavonoids, DPPH free radical scavenging and antioxidant capacity, similar to that seen in thermal treatments.
Rodrigues et al.	2017	Nitrogen cold plasma	elements in juice.	Test effects of CP on bioactive Overall, vitamin C, flavonoid and polyphenol content and the antioxidant activity increased. NCP resulted in higher polyphenol content at the highest nitrogen flow. Vitamin C increased at the lowest flow.
Sarangapani et al.	2017	High voltage dielectric barrier discharge	Test effects of CP on bioactive pesticides, and sensory elements of blueberries.	HVACP effectively reduced pesticides on blueberries. elements in juices, effect on It increased flavonoids and total phenols but decreased ascorbic acid. HVACP did not significantly affect the color or firmness of the blueberries.
Scholtz et al.	2010		Negative corona discharge Assess the effect of CP on vegetative and spore forming bacteria.	NCD was found to be effective on vegetative bacteria but an order of magnitude less effective on spore forming bacteria.
Jin et al.	2022	Cold atmospheric plasma	Test CP's effect on bioactives in Winter Jujube	CP improved gene expression related to phenolic biosynthesis, increased total phenolic content, maintained antioxidant levels, and reduced oxidative damage.
Deng et al.	2007	Nonthermal plasma technology	Assess the reduction of pathogens on almonds	NTP reduced <i>Escherichia coli</i> on inoculated almonds.
Niemira et al.	2018	Nonthermal plasma technology	Assess the effect of CP on biofilms	NTP reduced <i>Escherichia coli</i> biofilms.
Bahrami et al.	2016	Atmospheric pressure cold plasma	Assess the effect of CP on microflora and functional qualities of wheat.	APCP did not have a significant impact on the wheat's microflora. It did alter change the molecular weight distribution of wheat protein polymer and oxidized free fatty acids and phospholipids.
Beyrer et al.	2020	Micro-discharge cold atmospheric pressure plasma	Assess the reduction of pathogens in spirulina.	SMD-CAPP reduced Bacillus spores and increased polyphenols and other antioxidants in spirulina in with short exposures.

outbreaks and costly product recalls, offering potential savings related to recall expenses and brand protection (Nwabor et al. [2022\)](#page-11-25).

<span id="page-8-6"></span>Despite these advantages, further research is needed to fully understand the potential and optimize the use of cold plasma technology in food processing. In addition to scale-up studies, comprehensive life cycle assessments should be conducted to quantify the overall environmental impact of cold plasma technology compared to conventional methods. Cost-benefit analyses will also help food manufacturers make informed decisions about adopting this technology, considering both the upfront investment and long-term savings.

#### **Commercialization of CP for food processing**

CP is emerging as a versatile, non-thermal processing method with significant potential for enhancing various food processing techniques such as hydrogenation, sterilization, and germination. CP has been argued to be an eco-friendly alternative to traditional oil hydrogenation, noting its ability to convert unsaturated bonds to saturated ones without producing harmful trans-fatty acids. However, optimization of plasma parameters and processing conditions is crucial to minimize the formation of secondary lipid oxidation compounds and trans-fatty acids. This study emphasizes the need for further research to harness CP's potential fully in the partial hydrogenation of oils (Thirumdas [2023\)](#page-11-26).

<span id="page-8-8"></span><span id="page-8-7"></span><span id="page-8-2"></span>When applying CP to beverage processing, Ozen et al. [\(2024\)](#page-11-27) and Chen et al. [\(2024\)](#page-10-27) examined the effects of atmospheric cold plasma (ACP) on the quality and stability of juices and coconut milk, respectively. Ozen et al. [\(2024\)](#page-11-27) showed that while ACP treatment generally maintains the physicochemical properties of apple and cantaloupe juices, it can significantly impact antioxidant activity and volatile compound profiles depending on the gas composition used (Ozen et al. [2024](#page-11-28)). Similarly, Chen et al. [\(2024\)](#page-10-27) reported that ACP treatment of coconut milk reduces microbial load and enhances stability without significantly altering sensory quality, making it a promising alternative to thermal pasteurization (Chen et al. [2024\)](#page-10-28).The agricultural applications of CP, particularly in seed germination and grain processing, have recently been explored. Bozhanova et al. ([2024\)](#page-9-9) reported that CP treatment improves the germination and drought resistance of durum wheat seeds, while Li et al. [\(2023\)](#page-10-29) demonstrates that CP pretreatment enhances the nutritional profile of germinated brown rice by reducing phytic acid and increasing γ-oryzanol and GABA content. Together, these studies suggest that CP could revolutionize food processing by providing safer, more efficient, and sustainable alternatives to conventional methods.

<span id="page-8-5"></span><span id="page-8-1"></span>While CP is gaining popularity in the food industry, commercial applications of CP are still relatively scarce. Three companies have taken steps toward harnessing the potential of CP in the food sector—Advanced Plasma Solutions (APS), Clean Crop Technologies, and Neoplas.

Advanced Plasma Solutions (APS) is a pioneering force in the realm of Non-Thermal Plasma (NTP) technology. APS offers a comprehensive suite of services, from feasibility studies to full product development. Their company draws upon their expertise in plasma chemistry, electrical and mechanical engineering, design, and consulting. Their aim is to bridge the gap between research and market reality. Clean Crop Technologies is a company born out of an understanding of the challenges faced by agricultural supply chains. Their CP treatments focus on seeds, fruits, vegetables, cheese, meat, and seafood. This focus positions them strategically to enhance food safety and reduce wastage. Through CP technology, Clean Crop Technologies aims to elevate yields and protect food from molds, fungi, toxins, and pathogens, contributing to a more secure and sustainable global food supply. Neoplas is not singularly focused on the food sector, rather, they suggest their existing CP technology is a good fit for the food industry. Specifically, their website suggests their kinPen<sup>\*</sup>IND Portable Cold Plasma Device for Surface Treatment is a versatile solution for enhancing surface hygiene and quality across a wide range of applications—including the food and agricultural sector due to its ability to access intricate surface areas.

#### **The negative effects of CP**

CP treatments effectively reduce microbial load and improve the shelf life of some food products but they do come with some drawbacks. CP generates reactive oxygen species (ROS) and free radicals that trigger oxidative damage to lipids, proteins, and vitamins, altering sensory attributes and potentially affecting consumer preference. CP can also cause structural changes in the food matrix that impact the food's quality (Ekezie et al. [2017](#page-10-30); Zhang et al. [2018](#page-11-29); Birania et al. [2022](#page-9-10)). Other food sanitization methods come with benefits and disadvantages that food manufacturers and producers should consider when choosing which treatment is best to ensure both the safety and quality of their products. To wit, Ultraviolet irradiation's effectiveness in pathogen elimination is counterbalanced by its limited ability to penetrate foods with intricate structures or uneven surfaces, degradation of light-sensitive vitamins such as vitamin A and riboflavin, and can result in the generation of off-flavors, diminishing overall sensory quality. Unlike UV irradiation, which is limited to surface sterilization and can lead to material degradation over extended exposure, CP has deeper penetration capabilities and better material compatibility, particularly for heat-sensitive items (Cilliers et al. [2014;](#page-10-31) Guneser and Karagul Yuceer [2012](#page-10-32)).

<span id="page-8-4"></span><span id="page-8-3"></span><span id="page-8-0"></span>Gamma irradiation, another effective pathogen-reducing method, generates free radicals that induce oxidative damage, similar to CP, leading to textural changes and nutrient degradation. While CP may cause overexposure and alter the physical properties of treated materials, the concern is less pronounced with CP than gamma irradiation due to its more controlled generation of reactive species (Balakrishnan et al. [2021](#page-9-11)). Heat treatments (i.e. boiling, blanching, steaming, and microwaving) are a common method of pathogen control. These methods are known to cause nutrient loss and Maillard browning reactions that induce the formation of unhealthy advanced glycation endproducts (A.G.E.s) and

<span id="page-9-15"></span><span id="page-9-14"></span>affect flavor, color, and aroma. The structural changes induced by heat treatments can also disrupt texture (Lund & Ray, [2017](#page-10-33); Lee et al. [2018](#page-10-34)). Compared to heat treatments, which can cause significant thermal degradation of nutrients and sensory qualities in food products, CP operates at near-ambient temperatures, minimizing such damage (Farooq et al. [2023\)](#page-10-26). However, CP is not without its limitations, potential challenges include the optimization of treatment parameters to avoid the formation of undesirable byproducts, such as secondary lipid oxidation compounds or minor changes in volatile compounds. Therefore, while CP often presents a more balanced profile of effectiveness and material safety than UV, gamma irradiation, and heat treatments, careful parameter control is essential to mitigate its negative effects (Thirumdas [2023](#page-11-30)).

# **Conclusion**

<span id="page-9-12"></span>To effectively differentiate the focus and contributions of the current manuscript from similar studies, it is important to contextualize its unique approach and findings in relation to existing literature. Cherif et al. ([2023](#page-10-35)) provided a broad overview of cold plasma applications across various stages of food production, emphasizing its role in sustainable and safe food production by summarizing recent studies on microbial and pesticide treatments. Rao et al. ([2023\)](#page-11-31) focused on the application of cold plasma in low-moisture foods, highlighting its efficacy in inactivating pathogens, degrading mycotoxins, and promoting seed germination, while maintaining the quality of food.

Farooq et al. [\(2023](#page-10-36)) specifically delved into the advancements of cold plasma technology in food processing and its effects on physicochemical characteristics, presenting an in-depth discussion on optimizing product surface characteristics and processing parameters to enhance cold plasma efficacy. Kaavya et al. ([2023\)](#page-10-37) explore the influence of cold plasma on the rheological properties of foods, detailing its effects on viscosity, crystallization, and shear properties.

<span id="page-9-13"></span>In contrast, this study provides contextual evaluations that addresses the microbiological safety, nutritional retention, sensory qualities, and safety in food products processed with CP. It offers a distinct perspective by comparing the efficacy and potential of cold plasma with traditional and other novel thermal processing techniques, thereby filling a niche that emphasizes not just the general capabilities of CP, but its specific applications and outcomes. This targeted approach enhances the understanding of CP practical implementation in food processing and identifies the specific challenges and opportunities it presents for improving food manufacturing in real-world scenarios.

CP technology represents a promising advancement in the field of food processing. Its non-thermal nature and ability to effectively inactivate pathogens on the surface of low-moisture foods make it a valuable tool for enhancing food safety without the development of A.G.E.s that are present at high levels in many ultraprocessed food products and are extremely detrimental to health (Anti-AGEs Foundation 2024). Additionally, CP treatment offers the potential to extend the shelf life of perishable foods by

reducing the spoilage microorganisms and inactivating enzymes that contribute to food deterioration.

Furthermore, present research on CP technology suggests that it has an overall minimal impact on the nutritional content and sensory characteristics of food and food products, ensuring that overall quality and nutritional value may be maintained. Despite challenges related to cost, scalability, and regulatory considerations, ongoing research and development efforts are focused on optimizing CP technology and exploring novel applications. With the development of more portable devices that can be integrated with other food handling and food processing technologies, the future of CP technology holds promise for improving food safety, extending shelf life and enhancing the overall quality of our food products.

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