# CHAPTER 2

# Review of Literature

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## 2.1 Orange fruit

The orange (*Citrus sinensis* L.) is a non-climatic citrus fruit belonging to the Rutaceae family. This fruit is widely consumed and is known for its sweet flavor and high nutritional value. Orange juices are comprised of moisture content (86–90%), ascorbic acid (15–65 mg/100 mL), flavanones (5.5–15 mg/100 mL), organic acids (0.5–1.0%), carotenoids (0.3–0.9 mg/100 mL), and dietary fibers (0.10–0.13%), respectively (Vavoura et al., 2022; Galaverna and Dall'Asta, 2014). The orange fruit has its origins in Southeast Asia, particularly in regions that include Southern China, India, and Myanmar (Wang et al., 2018). Oranges are generally round to oval, with a bright orange rind that can sometimes show green patches in warmer climates. The flesh is juicy and segmented, characterized by a sweet-tart flavor profile due to the balance of sugars and organic acids (Deng et al., 2020). Oranges are economically significant as they are one of the leading fruit crops globally. In 2022, approximately 76 million tonnes of oranges were produced worldwide, with Brazil being the largest producer (Deng et al., 2020). Orange processing generates many by-products, primarily peels, which contain bioactive compounds that could be utilized for nutraceuticals. This highlights an opportunity for sustainable practices by converting waste into valuable products (Nayana and Wani, 2024).

### 2.1.1 Varieties of oranges

Varieties of oranges also play a pivotal role in influencing juice quality and standardization. Oranges come in various cultivars, each with unique characteristics appealing to different tastes and uses. Notable varieties include the Navel orange, known for its sweetness and seedless nature, primarily consumed fresh (FAO, 1999). Valencia oranges, popular for their high juice content and balanced sweetness and acidity, are widely used for juice production (FAO, 1999). Blood oranges, with deep red flesh and berry-like characteristics, are used in salads, desserts, and juices. Clementines, a hybrid of mandarin and sweet orange, are sweet, easy to peel, and often eaten fresh. Bergamot oranges, valued for their aromatic peel, are primarily grown for extracting essential oils used in perfumes and Earl Grey tea (FAO, 1999).

The northeast region of India is renowned for its diverse citrus species, including unique orange varieties thriving in the region's distinctive climate and soil conditions.

Notable varieties include the Khasi Mandarin (*Citrus reticulata*), praised for its sweetness and juiciness, predominantly grown in Meghalaya's Khasi hills (Bhattacharya and Dutta, 1956). The Nagpur Mandarin, known for its rich flavor and easy peeling, is primarily cultivated in Maharashtra and grown in parts of Northeast India (FAO, 1999). The Coorg Santra, characterized by its sweet taste, is mainly cultivated in Karnataka and found in the Northeast regions (Venkataravanappa and Sonavane, 2022). The Assam lemon (*Citrus jambhiri*), although primarily a lemon, is often included in citrus discussions due to its prevalence in local cuisine (Hore and Barua, 2004). Arunachal Pradesh is also home to several indigenous orange varieties that contribute to the region's horticultural diversity, emphasizing the role of local varieties in enhancing the °Brix-acid balance and juice quality in the industry.

The Arunachal Pradesh orange, also known as Wakro orange, is a notable citrus variety cultivated in Arunachal Pradesh, India. This variety has gained recognition for its distinct characteristics and has been granted a geographical indication (GI) tag. Arunachal Pradesh oranges are known for their distinct sweet-sour taste, which is attributed to their moderate acidity and high total soluble solids. This balance makes them particularly enjoyable both as fresh fruit and in culinary applications. The fruit typically features an orange-colored peel of medium thickness, is round, and has a loose skin that facilitates easy peeling. The segments within are easily separable, contributing to their appeal for fresh consumption, known for its high juice content, they are ideal for juicing and are rich in ascorbic acid, adding to their nutritional value. Thriving in Arunachal Pradesh's unique climatic conditions, characterized by cool temperatures and ample rainfall, these oranges are a significant horticultural crop, accounting for approximately 90% of the state's total citrus production (Datta et al., 2021). The inclusion of Arunachal Pradesh oranges underscores their importance in enhancing juice quality and the °Brix-acid ratio, reflecting their potential to contribute to the global juice industry.

### 2.1.2 Standardization of fruit juice

Juice standardization is critical in the food and beverage industry to ensure consistent fruit juice quality and taste. The °Brix-acid ratio is one of the key parameters used for standardization, and it measures the balance between the sugar content (°Brix) and the juice acidity. This ratio significantly influences the sensory attributes of juice, such as sweetness, sourness, and overall flavor profile. Some studies reported that consumer acceptance of orange juice is closely linked to its °Brix-acid ratio, with an optimal range of 12:1 to 18:1, and for apple juice, it is 15:1 to 30:1 (Kimball, 1991; Jayasena and Cameron, 2008; Board and Woods, 2007).). Standardizing juice based on the °Brix-acid ratio ensures consistent flavor and complies with quality standards set by regulatory bodies like the Codex Alimentarius.

The °Brix value represents the percentage of soluble solids, primarily sugars such as sucrose, glucose, and fructose, measured using a refractometer. Conversely, acidity is attributed mainly to organic acids like citric, malic, and tartaric acids and is measured as titratable acidity using a standardized base. The ratio between these two parameters, the °Brix-acid ratio, provides a composite index of the sensory balance in juices, effectively influencing perceived sweetness and sourness. For instance, citrus juices, particularly those derived from oranges and grapefruits, have a delicate interplay of sugar and acid essential for their characteristic taste. The °Brix-acid ratio becomes a critical tool in determining the right harvest time for fruits, with mature fruits exhibiting higher ratios due to increased sugar levels and reduced acidity (Singh et al., 2023).

In the context of consumer satisfaction, the °Brix-acid ratio has a crucial role in the acceptability of juices in different markets. Consumers in tropical and subtropical regions often prefer sweeter juices with higher °Brix-acid ratios, while those in temperate regions may favor a tangier taste profile. The role of sensory evaluation panels and consumer preference studies cannot be overstated, as they help manufacturers fine-tune the °Brix-acid ratio to cater to diverse palates. Research has indicated that preferences for °Brix-acid ratios vary geographically based on cultural and individual taste differences. This makes standardization a scientific and market-driven exercise (Kimball, 1991; Gayathri et al., 2014).

Achieving an optimal °Brix-acid ratio requires addressing several variables, including fruit variety, maturity, and environmental factors. Fruit varieties inherently differ in their sugar and acid profiles, necessitating careful selection for processing. For instance, Valencia oranges are ideal for juice production due to their balanced sweetness and acidity (Seminara et al., 2023). Similarly, climatic conditions such as temperature, sunlight, and rainfall impact the sugar-acid balance in fruits, influencing the final °Brix-acid ratio. Post-harvest handling practices, including storage and transportation, also affect the chemical composition of fruits. Prolonged storage may lead to changes in sugar content and acidity, making it essential for producers to monitor and adjust these parameters before processing (Rehman et al., 2019, Qureshi et al., 2023).

Processing techniques, such as blending, dilution, and adding sweeteners or acids, are commonly employed to standardize the °Brix-acid ratio. Blending juices from different batches or varieties helps achieve consistency by balancing high-sugar, low-acid juices with their low-sugar, high-acid counterparts (Sarbatly et al., 2023). In many cases, natural variation is insufficient to meet standard requirements, manufacturers may add food-grade sweeteners like sucrose or glucose syrup to increase the °Brix value or acids like citric acid to enhance acidity. However, such interventions must comply with regulatory guidelines to ensure product authenticity and safety. Concentration and evaporation techniques are also utilized to increase °Brix values, particularly in juices intended for reconstitution. Producers can achieve higher sugar concentrations by removing water through controlled evaporation, which is diluted to the desired ratio at the point of sale (Castro-Muñoz et al., 2022).

The importance of the °Brix-acid ratio extends beyond sensory attributes to include nutritional and functional considerations. The ratio indirectly reflects the balance of sugars and acids, which are critical for the nutritional profile of juices. Sugars serve as an immediate energy source, while organic acids contribute to metabolic functions and act as natural preservatives. Balancing these components enhances flavor and extends shelf life by inhibiting microbial growth. The acidity of juices, as measured through the °Brix-acid ratio, plays a key role in determining microbial stability and resistance to spoilage. Lower ratios, indicative of higher acidity, are particularly effective in creating an inhospitable environment for pathogens, thus enhancing food safety (Fellers et al., 1998).

In international trade, the °Brix-acid ratio is a benchmark for juice quality, with importing countries often specifying minimum or maximum values to ensure compliance with local standards. Regulatory bodies such as the FDA and the Codex Alimentarius provide detailed guidelines on acceptable ranges for °Brix and acidity, facilitating standardization across markets. Adherence to these standards ensures product safety and enhances consumer confidence. For manufacturers, achieving the specified °Brix-acid ratio can be a competitive advantage, as it signifies a commitment to quality and consistency (Dransfield, 1999).

Standardizing juice based on the °Brix-acid ratio is essential for ensuring consistent quality, flavor, and consumer satisfaction. By carefully balancing sweetness and acidity, manufacturers can produce juices that meet sensory and regulatory standards, enhancing market competitiveness. Advances in analytical techniques and processing methods continue to enhance the precision and efficiency of standardization, enabling the production of high-quality juices that appeal to diverse consumer preferences. As the juice industry evolves, the °Brix-acid ratio will remain a cornerstone of quality control and product development, underscoring its importance in the global beverage landscape (Kraus and Popek, 2013; Magwaza and Opara, 2015).

### 2.1.3 Current status of orange juice processing

The processing and storage of citrus juices face several challenges that affect their quality, nutritional value, and marketability. One major issue is non-enzymatic browning, which compromises citrus juices' visual and sensory appeal during storage and processing. This browning often results from exposure to oxygen and heat, exacerbating chemical reactions that darken the juice and alter its flavor profile (Pandey and Negi, 2018). Maintaining low temperatures during the various processing stages is crucial to mitigate these effects. Additionally, the steps involved in processing citrus juices, such as evaporation and concentration, contribute to significant quality losses, including diminished color vibrancy, aroma volatiles, and nutritional content. These losses are directly influenced by the exposure time and temperature during processing, with longer and higher-temperature treatments leading to more pronounced deterioration (Akyıldız et al., 2023). As a result, striking a balance between efficient processing and quality preservation is one of the primary challenges for manufacturers.

Nutritional degradation of sensitive compounds, particularly vitamin C, is another concern. High-temperature processes like pasteurization are essential for ensuring microbial safety but often degrade essential nutrients such as vitamin C and other bioactive compounds. Vitamin C, being highly thermolabile, is particularly susceptible to breakdown under heat, which poses challenges in achieving effective sterilization without sacrificing nutritional quality (Gómez et al., 2011). Similarly, the formation of undesirable compounds during concentration processes further complicates juice production. At higher concentrations, gel-like substances can form, impeding further processing and packaging operations. Heating during concentration also promotes the generation of off-flavors, negatively impacting the final product's sensory attributes and reducing consumer acceptance (Afraz et al., 2024). These issues underline the importance of optimizing processing parameters and exploring advanced technologies to minimize quality and nutritional losses.

Technological limitations also challenge the juice industry. Conventional juice processing methods typically involve high temperatures, which can degrade the sensory and nutritional quality of juices. Emerging technologies such as low-temperature methods, oscillating magnetic field processes, ultrasound, high-pressure, and non-thermal plasma technologies is used to improve the nutrition and preserve the juice quality by reducing thermal exposure during concentration (Mukhtar et al. 2022, Sharma et al., 2024). However, adopting such technologies on a commercial scale remains limited due to high initial investment costs and operational complexities.

### 2.1.4 Changes in physicochemical properties

The processing of orange juice involves techniques that significantly impact its physicochemical, nutritional, and sensory properties. Pasteurization, concentration, and ingredient addition are critical to the product's quality. Pasteurization, while essential for microbial safety, often results in the degradation of thermolabile nutrients such as vitamin C, a vital component of orange juice’s nutritional profile (Akyıldız et al., 2023). This loss of nutrients during processing underscores the challenge of balancing safety and nutritional retention, a key focus in juice processing research. Similarly, adding functional ingredients, such as resistant maltodextrin, has been shown to alter the physicochemical properties of orange juice. Arilla et al. (2022) demonstrated that resistant maltodextrin not only enhances the viscosity of pasteurized orange juice but also improves its texture and mouthfeel, contributing to consumer satisfaction and product stability during storage. These modifications highlight the potential of ingredient innovation to address processing challenges while improving the overall quality of the juice.

Processing also affects the rheological properties of orange juice, which are crucial for both industrial operations and consumer acceptance. The viscosity of orange juice, a key rheological parameter, is highly sensitive to changes in soluble solids content, temperature, and concentration levels. Dahdouh et al. (2016) observed that higher levels of soluble solids significantly increase the viscosity of orange juice, which, while beneficial for texture, poses challenges to processing efficiency, particularly in pumping and bottling during operations. Temperature, a critical factor in most processing steps, further influences the rheological behavior of the juice, making it essential to optimize processing conditions to maintain desired flow characteristics. Additionally, concentration techniques such as evaporation exacerbate these changes by increasing the sugar content, further altering the juice's flow properties. These rheological changes affect the ease of processing and have implications for consumers' sensory experience, making them a vital consideration in juice production.

The color of orange juice, a primary quality indicator, is another attribute significantly affected by processing techniques. Thermal treatments such as pasteurization and concentration often lead to non-enzymatic browning, a reaction that diminishes the juice's visual appeal and perceived freshness. Vikram et al. (2005) noted that browning reactions are primarily driven by exposure to high temperatures during processing, which can also degrade carotenoids, the pigments responsible for the vibrant orange color of the juice. This loss of color, combined with the formation of off-flavors due to thermal degradation of aroma compounds, can negatively impact consumer perception and marketability. Maintaining optimal processing conditions, such as lower temperatures and shorter exposure times, is critical to mitigate these effects. Additionally, advanced processing technologies, including non-thermal methods like high-pressure processing, are being explored to preserve orange juice's sensory attributes and nutritional quality while ensuring microbial safety (Ambreen et al., 2023).

Microbial stability is essential in orange juice processing, as it directly influences shelf life and safety. Thermal treatments, such as pasteurization, have long been employed to inactivate pathogenic microorganisms and spoilage agents, extending the product's shelf life. However, the effectiveness of these treatments often comes at the cost of quality degradation. Guerrouj et al. (2016) reported the interaction between microbial stability and preserving physicochemical properties in sonicated mild temperature treated orange juice. They found that sonicated temperature (43–45 °C) effectively reduces microbial load but can also exacerbate quality losses, including flavor, color, and texture changes. These findings have spurred interest in alternative methods, such as non-thermal pasteurization and low-temperature processing, which aim to achieve microbial stability without compromising juice quality.

## 2.2. Non-thermal approaches in juice processing

The food sector is progressively embracing non-thermal technologies as alternatives to conventional thermal processing methods for juice preservation, driven by consumer demand for minimally processed products that retain freshness, flavor, and nutritional value (Pankaj et al. 2018). Traditional thermal treatments, such as pasteurization and sterilization, are effective for microbial inactivation but often degrade heat-sensitive nutrients and sensory qualities, including flavor, aroma, and color, making way for innovative non-thermal methods such as cold plasma (CP), pulsed electric fields, ultrasonication, high hydrostatic pressure, and irradiation (Mukhtar et al. 2022; Bhatnagar et al., 2022; Kumar et al., 2023). These advanced techniques operate at ambient or low temperatures, effectively preserving the natural quality of juices while ensuring safety, extending shelf life, and reducing energy consumption. Moreover, their eco-friendly nature aligns with the increasing emphasis on sustainable food processing practices.

CP has emerged as a cutting-edge non-thermal technology that uses ionized gases containing reactive oxygen and nitrogen species (ROS and RNS) to inactivate microorganisms. The process operates at near-ambient temperatures, which helps preserve juices' physicochemical properties. Studies have shown its efficacy in microbial reduction in juices such as apples, oranges, and tomatoes, achieving significant decreases in microbial counts without altering sensory characteristics or compromising bioactive compounds like vitamins and antioxidants (Dasan and Boyaci, 2018).

Beyond microbial safety, CP technology has been reported to improve the functional properties of juice components, such as increasing protein solubility and enriching phenolic content. Such enhancements contribute to the final product's nutritional value and consumer appeal. Despite these benefits, the scalability and cost-effectiveness of CP systems remain significant challenges that require further research and technological advancements to enable their widespread adoption.

Pulsed electric fields represent another promising non-thermal technology that uses short bursts of high-voltage electric fields to disrupt microbial cell membranes and deactivate spoilage enzymes. This process effectively inactivates pathogens like *Escherichia coli* and *Listeria monocytogenes* in liquid foods while minimizing thermal exposure. As a result, PEF-treated juices retain higher levels of vitamins, antioxidants, and other bioactive compounds compared to their thermally processed counterparts (Timmermans et al., 2019; Mukhtar et al., 2022).

Ultrasonication is another non-thermal approach that has gained attraction in juice processing due to its versatility and effectiveness. This method uses high-frequency sound waves to create cavitation—the formation and collapse of microbubbles—which generates localized high-pressure and temperature effects. These conditions lead to microbial inactivation, improved mass transfer, and enhanced juice quality. Ultrasonication has been widely used to reduce microbial loads, improve emulsification properties, and increase the extraction of phenolic compounds and other bioactive ingredients in juices. Studies on sugarcane and orange juices have shown significant increases in antioxidant activity and phenolic content following ultrasonication treatment (Guerrouj et al. 2016; Mukhtar et al., 2022).

High hydrostatic pressure technology has proven to be one of the most effective non-thermal methods for ensuring juice safety while maintaining its freshness and nutritional value. The process involves subjecting juices to extremely high pressures (200 to 700 MPa), which disrupts microbial cell walls and deactivates spoilage enzymes without the need for elevated temperatures. This makes high hydrostatic pressure particularly suitable for preserving heat-sensitive nutrients like vitamin C, carotenoids, and polyphenols. Studies on apple and sugarcane fruit juices have highlighted the ability of high hydrostatic pressure to extend shelf life while maintaining sensory attributes such as taste, aroma, and color (Sreedevi et al., 2021; Szczepańska et al., 2021).

Irradiation is a non-thermal technology that uses ionizing radiation, such as gamma rays, X-rays, or electron beams, to eliminate pathogens and extend the shelf life of juices. This technique effectively targets a wide range of microorganisms, including *Salmonella* and *E. coli*, while preserving the sensory and nutritional quality of the juice (Shahi et al., 2021; Bhatnagar et al., 2022). Irradiation has been successfully applied to enhance the safety of orange juice without causing significant changes in their organoleptic properties (Foley et al., 2002). However, the perception of irradiation among consumers poses a significant challenge, as misconceptions about its safety and potential effects on food quality can limit market acceptance.

## 2.3 Cold plasma technology

Plasma, commonly called the fourth state of matter, emerges from continuously adding energy to a substance, transitioning sequentially from solid to liquid to gas (Harikrishna et al., 2023). Among the various advancements in plasma research, CP technology has gained significant attention for its versatile applications, especially in the agricultural and food industries. This innovative technology provides unique mechanisms of action and can function either as a stand-alone process or in combination with other preservation methods. Atmospheric cold plasma (ACP), a non-thermal technology, has shown remarkable potential in microbial inactivation. It is a promising tool for enhancing food safety, particularly in minimally processed or fresh produce (Misra et al., 2011). As consumer demand for fresh, high-quality fruits and vegetables continues to rise, the food industry faces the challenge of developing effective preservation techniques that ensure safety while maintaining product quality. Microbial contamination, especially in raw fresh produce, is a significant concern, necessitating advanced interventions to prevent spoilage and ensure safety without compromising food's nutritional and sensory attributes.

ACP stands out as an emerging method for microbial decontamination, offering a non-thermal alternative that aligns with modern food processing goals (Ozen et al., 2022). This is particularly relevant in minimizing the risks associated with microbial contamination in fresh produce. Unlike traditional methods such as thermal processing, which may result in losing essential nutrients and sensory characteristics, CP technology operates at low temperatures, making it an ideal solution for delicate food products (Zhang et al., 2022). In addition to its microbial decontamination capabilities, ACP has demonstrated the potential to prolong the shelf life of fresh produce, offering a sustainable solution to enhance food safety and quality. The principles of CP technology, which involve generating plasma at atmospheric pressure, contribute to its effectiveness in food preservation (Lacombe et al., 2015).

To enhance food safety and quality, CP technology can also be combined with other preservation methods, such as refrigeration or modified atmosphere packaging. The growing interest in CP technology stems not only from its potential for microbial inactivation but also from its ability to improve food safety sustainably. As the global demand for fresh and minimally processed foods rises, effective, non-thermal food preservation techniques are becoming increasingly important (Allai et al., 2023).

Technology's versatility and potential for integration into existing food processing systems make it a promising candidate for widespread adoption in the food industry. CP has demonstrated its ability to inactivate a broad spectrum of microorganisms, including bacteria, fungi, and viruses, without significantly affecting the sensory and nutritional qualities of food (Lacombe et al., 2015). This capability makes it particularly valuable for treating fresh produce, which is often consumed raw and highly susceptible to microbial contamination. ACP has been shown to effectively reduce microbial populations on the surfaces of fruits and vegetables, extending their shelf life and improving their safety for consumption. Furthermore, CP has the added benefit of targeting microorganisms resistant to conventional disinfection methods, offering a more effective solution for food safety. In addition to its microbial decontamination capabilities, ACP has positively affected food quality. Studies have demonstrated that CP treatment can enhance the appearance, texture, and nutritional content of food products, making it a promising method for enhancing both food safety and quality preservation. CP treatment has been shown to increase the antioxidant capacity of certain fruits and vegetables, potentially offering additional health benefits to consumers (Misra et al., 2011).

### 2.3.1 Plasma generation sources

CP technology employs a plasma generator to create an ionized gas state, typically at low pressures or atmospheric conditions, using gases like argon, oxygen, nitrogen, or mixture. Plasma generators are central to this technology and can operate in either batch or continuous systems, depending on the processing requirements. Various plasma generation methods have been explored for batch and continuous systems, such as dielectric barrier discharge (DBD), atmospheric pressure plasma jets (APPJs), radio frequency (RF), plasma jet, and gliding arc discharge (GAD) (Farooq et al., 2023). Gupta et al. (2024) noted that DBD systems are preferred for their scalability and ability to generate non-equilibrium plasma, which ensures minimal thermal effects on food matrices. In batch systems, the DBD electrodes parallel the product chamber, ensuring uniform plasma distribution over the treated surface or liquid. APPJs, on the other hand, are highly adaptable for continuous processes as they generate plasma jets that can treat liquid or solid food products during their flow or movement.

### 2.3.2 Types of cold plasma system

CP system can be classified based on how the treatment process is conducted. These classifications refer to the mode in which the treated material is exposed to the CP. The CP systems are categorized as follows:

#### 2.3.2.1 Batch-type cold plasma technology

Batch-type systems are primarily used in laboratory-scale research or for treating small quantities of food, such as juice samples. Misra et al. (2011) DBD plasma systems are one of the most commonly used configurations for batch and continuous CP processing. In batch systems, DBD plasma is applied in a sealed chamber, exposing the product to plasma for a set duration. These systems offer precise control over treatment time and intensity, making them ideal for optimizing process parameters during experimental studies. The effectiveness of these systems depends on parameters such as gas composition, flow rate, power input, and treatment duration (Kumar et al., 2023). Xu et al. (2017) and (Wang et al. 2024) highlighted that batch and continuous plasma systems could effectively inactivate microbial contaminants in juices. Xu et al. (2017) reported that batch systems achieved higher microbial reductions due to longer residence times, whereas continuous systems maintained operational efficiency and prevented recontamination during large-scale treatments.

#### 2.3.2.2 Continuous type cold plasma technology

In contrast, continuous CP systems use conveyor belts or tubular reactors to ensure a seamless flow of the product through the plasma field. Misra et al. (2024) reported that the design and development of continuous system for fresh produce in food industry. This system is more efficient for high-volume processing, allowing uniform treatment and reducing operational downtime. Furthermore, continuous systems, often equipped with inline monitoring sensors, facilitate real-time quality assessment during processing. Wang et al. (2024) reported the role of CP in continuous systems for juice processing, highlighting its ability to maintain juice quality while effectively inactivating *Escherichia coli* and natural microorganisms in apple juice.

### 2.3.3 Cold plasma-assisted processing of juice

CP technology came as an innovative, non-thermal method with a significant ability to improve juices' safety, quality, and shelf life, as evidenced by extensive research over the last two decades. Misra et al. (2011) demonstrated that CP effectively reduced microbial loads in foods, achieving significant bacterial reduction without compromising juice quality. Research by Pankaj et al. (2017) highlighted the role of CP in preserving bioactive compounds such as phenolics and ascorbic acid in pomegranate juice, emphasizing the method's advantage in retaining nutritional content compared to traditional thermal pasteurization.

The enzyme inactivation capability of CP has also been explored extensively (Farias et al. 2022). The plasma application inhibited polyphenol oxidase (PPO) and peroxidase (POD) activity in apple juice, preventing enzymatic browning and preserving color. CP has shown promise in modifying juice composition and enhancing bioavailability. Hou et al. (2019) mentioned that plasma-treated juices exhibited higher antioxidant activity due to structural modifications of phenolic compounds during plasma exposure. Additionally, Paixão et al. (2019) investigated the impact of CP on volatile aroma compounds in siriguela juice, finding no significant loss of desirable flavor profiles, thus preserving consumer acceptability. Pipliya et al. (2024) demonstrated that plasma-treated juices exhibited prolonged microbial stability during storage. The mechanism involves the disruption of microbial cell walls, protein oxidation, and deoxyribonucleic acid (DNA) damage caused by reactive plasma species. Despite its benefits, CP technology is still under research, with challenges related to scalability, uniformity of treatment, and optimization of operational parameters. A study by Bermúdez-Aguirre et al. (2013) emphasize the need for further exploration of the plasma-juice interaction at the molecular level to ensure consistent outcomes and mitigate potential drawbacks, such as overproduction of reactive species that could impact juice quality. CP-assisted juice processing represents a cutting-edge approach that aligns with the demand for minimally processed, high-quality beverages with longer shelf life and enhanced nutritional profiles.

### 2.3.4 Effect of cold plasma on food quality

CP technology has gained considerable attention as a novel non-thermal approach for enhancing food safety while preserving or improving food quality attributes such as nutritional content, sensory characteristics, and functional properties. It employs RNS, ROS, and charged particles to inactivate microorganisms and enzymes without significant heat generation, thus preserving heat-sensitive nutrients. Misra et al. (2011) and Pankaj et al. (2017) have shown that CP effectively inactivates foodborne pathogens and spoilage organisms in various food products, including juices and fresh produce, without compromising their nutritional value or sensory quality. Hou et al. (2019) observed enhanced antioxidant activity in blueberry juice treated with plasma due to the structural modification of phenolic compounds, suggesting potential health benefits. Additionally, inactivating enzymes like POD and PPO during plasma treatment helps preserve the natural color and prevent enzymatic browning, as noted in studies on pineapple juice (Pipliya et al. 2022). However, some challenges remain, such as the potential formation of off-flavors or oxidative by-products during treatment, particularly at higher plasma intensities or prolonged exposure (Xu et al., 2017). CP is also known for its effect on food texture and functional properties, such as protein modification, which can enhance food ingredients' emulsifying and gelling properties (Zhang et al., 2020). CP has demonstrated significant potential to enhance food safety and quality, offering a sustainable and consumer-friendly substitute for traditional heat-based processing techniques.

### 2.3.5 Current status of plasma-assisted processing of fruit juices

Plasma-assisted processing of fruit juices has emerged as a promising non-thermal approach for enzyme inactivation, microbial decontamination, and preservation of nutrition and sensory attributes. In recent years, extensive research has demonstrated the potential of plasma technology to enhance the safety and shelf life of fruit juices without compromising their natural flavor, color, and bioactive compounds. Studies by Shi et al. (2011) and Ozen et al. (2022) have highlighted its efficacy in reducing microbial load in juices like orange and apple cider, achieving reductions of up to 5 log CFU/mL for common spoilage and pathogenic microorganisms. Moreover, plasma-assisted processing preserves heat-sensitive nutrients, including vitamins C and E, phenolic compounds, and antioxidants, as evidenced by Pankaj et al. (2017) in pomegranate juice. Enzyme inactivation is another crucial aspect, with research by Farias et al. (2022) reporting effective suppression of PPO and POD, which prevents enzymatic browning in juices such as apple. Plasma's ability to alter chemical bonds and improve the bioavailability of phytonutrients has also been observed, as described by Pipliya et al. (2023) in pineapple juice, where plasma offers an adequate level of POD inactivation while ensuring excellent extraction of phenolic components and high retention of antioxidants. On an industrial scale, plasma is gaining traction with advancements in continuous processing systems, which allow high-throughput treatment while maintaining quality and efficiency. Shi et al. (2011) highlighted the potential of CP for preserving orange juice with extended microbial stability during storage.

### 2.3.6 Influence of cold plasma processing parameters

#### 2.3.6.1 Voltage

The CP treatment's intensity increased with applied voltage and more prolonged treatment durations but diminished with greater juice depth (Kumar et al., 2023). The plasma generation voltage is directly correlated with the density of RNS and ROS (Pipliya et al., 2022). ACP produces reactive species such as hydroxyl radicals, nascent oxygen, nitrogen oxide, and ozone Baek et al., (2016). A study reported that the as the plasma voltage increased from 55 kV to 80 kV, the ozone concentration rose from 200 to 950 ppm (Wang et al., 2018). Baek et al. (2016) also reported an increased concentration of reactive oxygen-nitrogen species (RONS), including O, $O_3$, OHO, and NO, as the applied voltage was raised.

#### 2.3.6.2 Juice depth

Several processes, such as chemical transfer absorption, desorption, and diffusion, play a role in the movement of RONS from the gas phase into the liquid phase (Bruggeman and Leys, 2009; Zhou et al., 2018). Based on mass transfer kinetics, hydrogen peroxide and superoxide ions penetrate the liquid more deeply than other RONS (Chen et al., 2014). Perinban et al. (2019) observed that "both short-lived species (e.g., $O_2^-$ $ONOO^-$, and H) and long-lived species (e.g., $NO_3^-$, $H_2O_2$) reached the bottom of a 200 μm water layer". Similarly, Attri et al. (2015) reported a "decrease in hydroxyl radical concentration from 4.8 × $10^{-16}$ to 0.8 × $10^{-16}$ $cm^{-3}$ as the depth of the water layer increased from 2 to 6 mm".

#### 2.3.6.3 Treatment time

The duration of juice exposure to plasma significantly impacts the intensity of the treatment. Longer exposure times allow RONS to interact more extensively with various juice components. For instance, Wang et al. (2018) reported a 50 ppm increase in ozone production when the exposure time was extended to 9 min compared to 3 min at 80 kV. Prolonged treatment times have been shown to reduce enzyme activity in juices (Kumar et al., 2023; Pipliya et al., 2022; Xu et al., 2017; Illera et al., 2019). Similarly, extended treatment time led to a decrease in an enhancement in redness (*a**) in pork jerky (Yong et al., 2019) and pH levels in xanthan gum (Bulbul et al., 2019).

### 2.4 Response surface methodology

"Response Surface Methodology (RSM)" is a powerful statistical technique used for modeling and optimizing complex processes, including those in the food industry. RSM aims to find optimal operating conditions by exploring the relationships between multiple input variables and their effects on one or more output responses. It is beneficial when dealing with nonlinear, interactive, and multivariable systems, such as food processing, where numerous factors influence the final product quality. In thermal processing (e.g., pasteurization or sterilization), RSM has been employed to optimize parameters like temperature, time, and pressure for maintaining food safety while preserving sensory and nutritional qualities (Sreedevi et al., 2021). RSM provides a mathematical model to predict outcomes under different conditions, helping food processors identify optimal operating points for better product quality and process efficiency (Islam et al., 2019).

### 2.5 Enzyme and microbial inactivation

CP offers a versatile, non-thermal approach for microbial and enzyme inactivation. CP has several applications in microbial control, including surface sterilization in the food industry and medical disinfection. It is favored because it operates at low temperatures, preserving food and biological material quality (Singh and Thakur, 2024). CP impacts enzyme structure and function. The primary mechanisms involve the oxidation of amino acid residues, structural modifications, and inhibition of enzyme activity. ROS generated by CP can oxidize key amino acid residues in the active site of enzymes, leading to enzyme inactivation (Misra et al., 2016). CP can induce conformational changes in enzyme proteins, reducing their catalytic efficiency or rendering them completely inactive (Pipliya et al., 2022). Studies show that CP effectively inactivates various pathogens, including *Escherichia coli*, *Salmonella*, and *Listeria*, by disrupting cellular integrity and inhibiting vital metabolic processes (Ziuzina al., 2014). CP inactivates microorganisms by generating reactive ROS, RNS, and ultraviolet (UV) radiation. These reactive species can damage microbial cell walls, membranes, proteins, and nucleic acids. ROS, including hydroxyl radicals (•OH) and hydrogen peroxide ($H_2O_2$), disrupt microbial cell membranes and DNA, leading to leakage of cellular contents and cell death (Han et al. 2016). Nitric oxide (NO) and nitrogen dioxide ($NO_2$), key RNS generated by CP, interact with proteins and DNA, inducing structural alterations and enzyme inhibition (Roshanak et al. 2023). CP-generated UV radiation can cause DNA damage, including thymine dimer formation, preventing replication and leading to microbial death (Hosseini et al., 2020).

## 2.6 Kinetics modeling

Mathematical kinetics modeling plays a crucial role in understanding and optimizing various processes in food engineering, particularly those related to heat transfer, microbial inactivation, enzyme activity, and chemical reactions. In food processing, thermal and non-thermal treatments require accurate modeling to optimize conditions for maintaining food quality. Thermal processes, including pasteurization and sterilization, use models to predict the temperature distribution and the microbial and enzyme inactivation rate. Non-thermal processes like high pressure processing, pulsed electric fields, and CP treatments also benefit from kinetic models to optimize their effectiveness while minimizing detrimental effects on food quality (Kumar et al., 2024; Feroz et al., 2019). The kinetics of enzyme and microbial inactivation are typically described using first-order, Weibull, two-fraction, fractional conversion, $n^{th}$ order, biphasic, log-linear shoulder, Membre, Gompertz, logistic models, respectively, based on the temperature, voltages, and time of exposure to thermal or non-thermal treatments (e.g., high pressure, UV, or CP) (Kumar et al, 2024; Jaiswal et al., 2024).

## 2.7 Shelf-life and storage studies

Storage conditions, particularly temperature, significantly influence fruit juices' shelf life and quality. Temperature influences the microbial growth dynamics, enzymatic activity, and chemical stability of the juice, ultimately determining its flavor, color, nutritional value, and safety (Kumar et al., 2024). The shelf life of fruit juice is determined by a complex interplay of microbial, enzymatic, and chemical factors (Esua et al., 2023). Numerous studies have assessed how storage temperature influences these factors and the overall shelf life. Refrigeration is the most conventional way of extending juice shelf life without compromising the nutritional quality of food products. Szczepańska et al. (2021) reported that storing fruit juice at refrigeration temperatures (4–7 °C) significantly reduces the rate of enzymatic browning and microbial growth. At refrigeration temperatures, the enzymatic activities responsible for ascorbic acid degradation are slowed, maintaining juice quality for up to 7–14 days, depending on the packaging used and the type of juice (Feszterová et al. 2023). Alim et al. (2023) observed that refrigeration can maintain the juice's freshness, nutrient loss, particularly ascorbic acid degradation, continues slowly over time. In contrast, when juice is kept at room temperature (20–25 °C), oxidation takes place, and microbial spoilage occurs more quickly. Odriozola-Serrano et al. (2008) observed that juices stored at ambient temperature for a few days showed significant changes in flavor, color, and aroma, probably due to enzymatic activity and microbial contamination. Kaddumukasa et al. (2017) reported that increased microbial load at room temperature further accelerates spoilage, limiting shelf-life to around 2–3 days for unpasteurized, fresh juices.

The packaging material also influences the fruit and vegetable juices' shelf life and storage. Juice stored in glass containers had a longer shelf life than those stored in plastic or carton packaging (Pipliya et al. 2024). Glass bottles offer better protection from light and air, which reduces the oxidation rate and helps preserve the juice's flavor, color, and nutritional content. Sujeetha et al. (2020) reported that packaging materials that provide an airtight seal, such as vacuum-sealed bags or bottles, further extend shelf-life of food products by reducing oxidation and microbial contamination.

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