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Review Article

Bioavailability and mechanisms of dietary polyphenols affected by non-thermal processing technology in fruits and vegetables

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ABSTRACT

Plant polyphenols play an essential role in human health. The bioactivity of polyphenols depends not only on their content but also on their bioavailability in food. The processing techniques, especially non-thermal processing, improve the retention and bioavailability of polyphenolic substances. However, there are limited studies summarizing the relationship between non-thermal processing, the bioavailability of polyphenols, and potential mechanisms. This review aims to summarize the effects of non-thermal processing techniques on the content and bioavailability of polyphenols in fruits and vegetables. Importantly, the disruption of cell walls and membranes, the inhibition of enzyme activities, free radical reactions, plant stress responses, and interactions of polyphenols with the food matrix caused by non-thermal processing are described. This study aims to enhance understanding of the significance of non-thermal processing technology in preserving the nutritional properties of dietary polyphenols in plant-based foods. It also offers theoretical support for the contribution of non-thermal processing technology in improving food nutrition.

1. Introduction

Fruits and vegetables are an integral part of a healthy diet because they are rich in polyphenols, vitamins, minerals, carotenoids, and dietary fiber, all of which benefit human health (Pu et al., 2023). Polyphenols, as one of the most significant active components of plants, are produced by the secondary metabolism and important for protective plant from degradation after harvest (Abdel-Aziz and El-Hadary, 2022). They are regarded as powerful natural antioxidants with a variety of biological and pharmacological properties, including antioxidant, cytotoxic, anti-inflammatory, antihypertensive, and anti-diabetic activities (Rana et al., 2022). However, when evaluating the health consequences of polyphenols in humans, in addition to their content, the accessibility, absorption, and bioavailability of polyphenols in the body are essential factors that cannot be ignored. The bioavailabilities of different phenolic compounds vary widely, and the studies have demonstrated that even the most abundant phenolic compounds do not necessarily have the optimum bioavailability, which can limit the beneficial effects exerted on the body. To establish conclusive evidence of the efficacy of dietary polyphenols on health benefits, it is necessary to accurately determine the bioavailability of polyphenols to assess their biological activity. Therefore, current studies have focused on the bioavailability of polyphenols and methods have been developed to increase their bioavailability (Teng and Chen, 2019).

Thermal treatment, mainly used for microbial safety and enzyme inactivation, leads to the loss of nutrients and antioxidant activity in fruits and vegetables (Oliveira et al., 2012). Due to the considerable thermal reactivity and sensitivity of polyphenols, the chemical properties of polyphenols are changed during heat treatment through differential isomerization, oxidative polymerization and degradation. These changes affect their biological activities (Zhang and Liu, 2022a,b). With the increasing awareness of nutritious and balanced diets, consumers are no longer satisfied with food safety alone, and their demand for fruits and vegetables with high nutritional value, good organoleptic properties, and bioactivity has increased. Non-thermal processing techniques, such as high-pressure processing (HPP), cold plasma (CP), pulsed electric field (PEF), and supercritical carbon dioxide (SC–CO_2), are well suited for solving the problem of tropical damage with optimal sterilization effects. They maximize the nutritional value and flavor of fruits and vegetables. Therefore, non-thermal processing technologies have

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been widely researched as alternatives to thermal processing technologies (Kumar et al., 2021). Recently, it has been shown that different non-thermal processing techniques that rely on various mechanisms of sterilization for fruits and vegetables affect the content and bioavailability of bioactive compounds (e.g., polyphenols).

This review aims to elucidate the changes in content and bioavailability of polyphenols treated with non-thermal processing, including HPP, CP, ultrasound, PEF, and SC-CO_2, and summarizes the potential underlying mechanisms of action, offering evidence-based justification for a better understanding of the nutritional properties of fruits and vegetables.

2. Polyphenols in plants

Polyphenols are secondary metabolites present in plant and plant-based foods, which are sources of natural antioxidants (Abdel-Aziz and El-Hadary, 2022). These compounds are distinguished by the presence of at least one aromatic ring with one or more hydroxyl groups attached. It is a heterogeneous group classified into flavonoids (anthocyanins, flavanols, flavanones, flavonols, flavones, isoflavones, and chalcones) and non-flavonoid molecules (phenolic acids, stilbenes, lignans, tannins, and curcuminoids) (Di Lorenzo et al., 2021) (Figure 1). They are typically conjugated with sugars and organic acids. Over 8000 phenolic structures have been found in fruits, vegetables, and cereals, as well as in derived beverages such as fruit juice, tea, coffee, and wine (Fan et al., 2022). These phenolic compounds are involved in many facets of their biological systems, including plant morphology, reproduction, growth, plant resistance against predators and pathogens, and anti-germination of pre-harvest seeds (Zhang et al., 2023).

Previous study has identified the top 100 richest dietary sources of polyphenols and determined the amounts of polyphenols present in food servings using standard serving sizes in a study based on the Phenol-Explorer database (Perez-Jimenez et al., 2010a). Berry fruits are generally high in polyphenols, which might be attributed to the high content of polyphenolic pigments, such as anthocyanins (Perez-Jimenez et al., 2010b). Polyphenols, such as anthocyanins, flavonols, hydroxycinnamic acids, and tyrosols, are mostly found in vegetables with the highest levels of polyphenols, such as olives, chicory, globe artichoke heads, and onions (Yang et al., 2021). The herbs and spices in the seasoning group also contained substantial concentrations of phenolic compounds. However, because these compounds are ingested in low amounts, their contribution to overall polyphenol consumption is therefore limited.

Polyphenols in plants are able to protect organisms from external stimuli and eliminate reactive oxygen species (ROS), which have the potential to affect human health (Sandoval-Acuna et al., 2014). Polyphenols play a crucial role in many diseases owing to their antioxidant, cytotoxic, anti-inflammatory, antihypertensive, and anti-diabetic properties. Previous study demonstrated that the using of plant dietary polyphenols as adjuvants reduce the burden of non-communicable diseases, including cancer, diabetes, obesity, cardiovascular diseases, and neurodegenerative disease (Quero et al., 2020; El-Hadary et al., 2023).

3. Bioavailability of polyphenols

The composition of polyphenols is complex, the absorption and utilization degree is low, and the structure-activity relationship is unclear (Xie et al., 2016). The health benefits of polyphenols are related to their consumption and bioavailability. Although numerous studies have investigated the effects of polyphenols on human health, their limited bioavailability in human is hardly reported. From a nutritional perspective, bioavailability is defined as the fraction of intake that can be used for normal physiological functions. Bioavailability includes two more terms: bioaccessibility and bioactivity. Once a meal (or beverage)

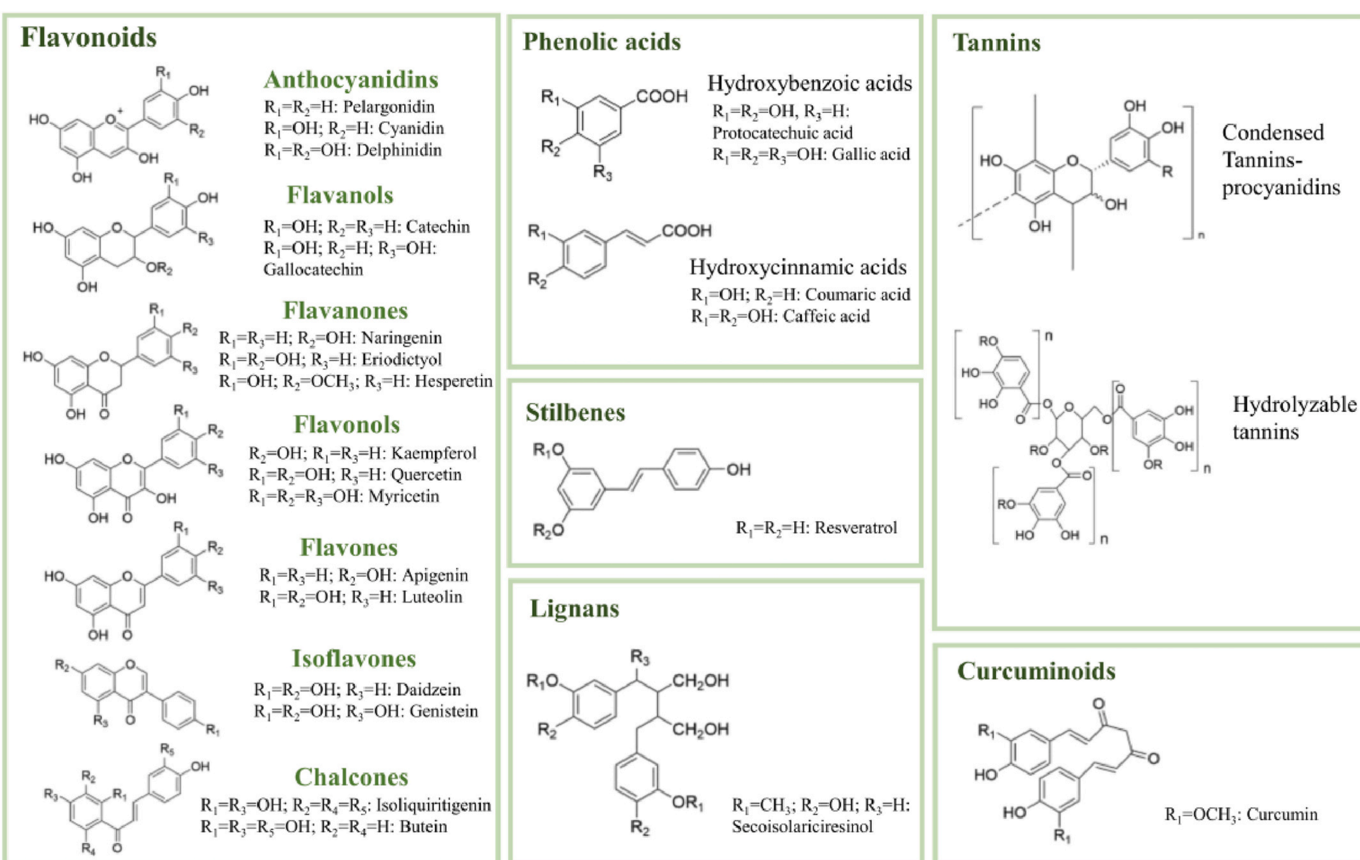

Fig. 1. Classes and chemical structures of dietary polyphenols.

is consumed, the first step in making a nutrient bioavailable is bioaccessibility (Galanakis, 2017). Bioaccessibility is described as the amount of a compound released from the food matrix in the gastrointestinal tract into the bloodstream and available for absorption. Before assessing the bioavailability of polyphenols and their metabolites in target tissues, they must be bioaccessible. Based on the definition, bioavailability and bioaccessibility can be assessed through in vitro and in vivo approaches, respectively. Scientists have commonly employed in vitro models such as simulated gastrointestinal digestion to assess the bioaccessibility of polyphenols, while the bioavailability of polyphenols is primarily assessed using in vivo models (Shahriyar Sahraeian and Mohammad-Taghi, 2023).

A range of physicochemical factors impact bioavailability, including compound structure, polarity, molecular weight, plant matrix, solid-state (crystalline or amorphous), digestibility of gastrointestinal enzymes, and absorption by enterocytes. Due to the complex structure and high molecular weight of polyphenols, some of them are absorbed by the stomach, and compounds such as catechins, flavanols, and flavonoids enter the circulatory system in the small intestine. Only 5–10% of ingested dietary polyphenols are directly absorbed by the small intestine. In contrast, most polyphenols (90–95%) reach the colon intact and undergo intestinal fermentation. The resulting polyphenol metabolites are absorbed and transported to the liver via the portal vein, where they are extensively degraded to form active metabolites (methylation, thioglycosylation, and sulfation). The metabolites then enter the body's circulation and reach the target tissues and cells, with the remaining or unused metabolites excreted in the urine (Yang et al., 2022). Proanthocyanidins belong to polyphenols and are one of the most studied polyphenols in absorption and metabolism (Zeng et al., 2020). The bioavailability of proanthocyanidins is largely affected by their degree of polymerization. The absorption rate of proanthocyanidin dimer is 5–10% of (-) -epicatechin. The absorptivity of trimers and tetramers is lower than that of dimers. Compared to (-) -epicatechin, absorbed intact dimers, trimers, and tetramers have limited phase II metabolism in rat gut and liver. The degree of polymerization exceeds 4 (DP > 4) Proanthocyanidins cannot be absorbed due to their macromolecular size and intestinal barrier (Ou and Gu, 2014). To comprehend the impact of polyphenols on human health, it is essential to first understand the nature of the primary polyphenols consumed, their dietary sources, their intake in different diets, their bioavailability, and the factors affecting their bioavailability (Grosso, 2018).

4. Effects of non-thermal processing technology on the content and bioavailability of polyphenols

Non-thermal processing technology has a great potential for application in the food industry and is mostly utilized for sterilization (Li et al., 2017). It retains high nutritional value while ensuring microbiological safety. Preservation (Chacha et al., 2021), drying of fruits and vegetables (Osae et al., 2020), juice processing, and the fresh-cut fruit and vegetable industry (Roobab et al., 2022) are the primary processing methods. Furthermore, current studies on the potential applications of non-thermal processing technologies focus on the extraction of bioactive compounds from processed fruits and vegetables (Abdel-Aziz and El-Hadary, 2022), as well as the improvements in the processing of fruit and vegetable by-products (Kumar et al., 2021).

Different thermal processing conditions lead to the diverse modifications in the structure and chemical properties of polyphenols, such as differential isomerization, oxidative polymerization, and degradation (Zhang and Liu, 2022a,b), which affect polyphenol content and the corresponding changes in biological activity. Emerging green non-thermal processing technologies use internal energy transfer to treat foods rather than high temperatures, which alters, among other things, cell structure and enzymes and, in turn, the polyphenol content and bioavailability of fruits and vegetables (Table 1 and Table 2).

At present, most non-thermal processing technologies are still in the laboratory research and pilot stage, and there are limited studies on the energy cost of non-thermal processing technologies. Table 3 summarizes the current studies on the energy cost of non-thermal processing technologies. The traditional thermal processing (397.54 kJ/kg) cost slightly more in total energy cost compared to HPP and PEF (339.94 kJ/kg and 301.81 kJ/kg, respectively) (Vignali et al., 2022). Furthermore, CP incurred the lowest energy cost (87.78 kJ/kg) based on a 10g apple slice compared to other non-thermal processing techniques (Ranjbar Nedamani and Hashemi, 2022). Accordingly, non-thermal processing may have even lower energy costs compared to traditional thermal processing, as different substances were used for non-thermal and traditional thermal processing in two studies. Therefore, detailed comparisons between non-thermal processing technology and traditional thermal processing, along with their economic values, require further study in the future. The important thing is to focus on the energy cost and the additional economic value from non-thermal processing.

4.1. High-pressure processing (HPP)

HPP is commonly used for 3–5 min at a hydrostatic pressure of 300–600 MPa (Houska et al., 2022a). Pressure is produced by a hydraulic pump or piston and isostatically transmitted to the food product inside the pressure vessel almost instantly and uniformly (Aganovic et al., 2021). Sterilization is achieved by disrupting the structure of the microbial membrane. Simultaneously, the temperature is typically between 20 °C and 60 °C, which is significantly lower than the heat treatment temperature; thus, HPP may protect the flavor and nutrients of food (Wang et al., 2023).

The type of food, polyphenol family, HPP treatment time, and pressure affect the polyphenol content. High-pressure treatment increased the total phenolic content (TPC) of fruits and vegetables. For example, compared with the control group, HPP treatment of jujube pulp, increased the TPC by 7.9% at 600 MPa (Shen et al., 2016). Vega-Gálvez et al. (2014) observed that pressure treatment at 500 MPa for 5 min resulted in the highest TPC (increased by 26.25% relative to the control) in cape gooseberry pulp (Vega-Gálvez et al., 2014). HPP-treated (450, 550, or 650 MPa for 5 or 15 min) kiwi berries led to a significant increase in TPC from 37% to 80% (Blaszczak et al., 2021). The increase in TPC of fruits and vegetables may be due to the disruption of cell wall structures, inactivation of enzymes associated with phenolic loss, or polyphenols that are distributed and aggregated within the fruit pulp microstructure after high-pressure treatment. However, other studies have reported contradictory results. HPP treatment of astringent persimmons (200 MPa/25 °C/3 min and 200 MPa/25 °C/6 min) resulted in a nearly 20% decrease in TPC compared to that of the control samples (García-Cayuela et al., 2018), which may be due to the different fruits containing different types of polyphenols. Pressure levels and treatment times also affect the TPC. According to a theory, a dynamic balance occurs between homogenization and enzymatic reactions. At low temperature (4 °C) and low pressure (50 MPa), the TPC decreases because of the dominance of the enzymatic reaction, but with increasing pressure or temperature, the homogenization effect is enhanced and the TPC increases (Liu et al., 2022).

HPP treatment enhances the bioavailability of polyphenols by increasing their content. Bioaccessibility is a determinant of the release and solubility of polyphenols for subsequent absorption and assimilation during digestion and is another important factor in bioavailability. HPP applied to juices and beverages of plant foods promotes polyphenolic substance accessibility by modifying the food matrix and cell wall permeability, thereby improving bioaccessibility (Rodríguez-Roque et al., 2015). Di Nunzio et al. (2020) recently reported that, compared with those in pasteurized juices, the flavonoid content (hesperidin, narirutin, and didymin) decreased after processing but reached higher bioaccessibility in mandarin (cv Ortanique) juice pasteurized (63°C/15 s) and processed by low-pressure homogenization (20 MPa) (Di Nunzio et al., 2020). However, He et al. (2016) investigated the effect of

Table 1
Effects and mechanisms of different non-thermal processing techniques on polyphenols in fruits and vegetables.

Technology	Material	Treatment conditions	Change of TPC	Mechanisms	Ref
HPP	Jujube pulp	400, 500, and 600 MPa/20 min	TPC and total flavonoid content (TFC) increased by 7.9% and 18.4% at 600 MPa	Cell destruction and phenolic efflux	Shen et al. (2016)
	Cape gooseberry pulp	300, 400 and 500 MPa/1, 3 and 5 min	Some treatments increased while others reduced the TPC ($P < 0.05$). The maximum TPC was observed at 500 MPa/5 min	Disruption of cell walls and hydrophobic bonds	Vega-Gálvez et al. (2014)
	Kiwiberry	450 MPa, 550 MPa and 650 MPa/5 and 15 min	HHP processing (450, 550, or 650 MPa for 5 or 15 min) resulted in a significant increase in the TPC	Improved the release of polyphenols from plant tissues	Blaszczak et al. (2021)
	persimmon	200 MPa/25°C/3 min; and 200 MPa/25°C/6 min	HHP treatments(200 MPa/25 °C/3 min and 200 MPa/25 °C/6 min) affected the TPC, losing near to 20%	Increased the activity of oxidase enzymes (PPO and POD) after HHP	García-Cayuela et al. (2018)
	Mandarin juice	Homogenization at 20 MPa	Reduced the concentration of total phenolics and main flavonoids	Forces and temperature stresses generated in the homogenizing valve during processing	Di Nunzio et al. (2020)
CP	Cloudy apple juice	(a) spark discharge, (b) glow discharge Gas source: air Variable frequency: 20–65 kHz Time: 1–5 min	TPC increased significantly with increasing treatment time, reaching a maximum in the 4 and 5 min treated juices, where TPC increased by 69% and 64%, respectively	Increased cell membrane breakdown	Illera et al. (2019)
	Green tea leaves	DBD Gas source: air and N_2 Frequency: 6 kHz Time: 5 and 15 min	Gallic acid slightly increased and treatment at 15 W for 15 min increased catechins by 103.12% compared to the control	Ion bombardment in CP causes membrane rupture and phenolic efflux Degradation of phenolic compounds due to oxygen radicals	Keshavarzi et al. (2020)
	Fresh-Cut Apples	DBD Gas source: gas source: Frequency: 12.7 kHz Time: 30 and 120 min	TPC remainednchangede	Oxidation reaction and enzyme activity inhibition	Ramazzina et al. (2016)
Ultrasound	Apple juice	Frequency:25 kHz Temperature: 20 °C Time: 30, 60 and 90 min	TPC was significantly increased for 30, 60 and 90 min ($P < 0.05$)	Ultrasound-generated cavities cause enhanced cell wall disruption and promote the release of polyphenols	Abid et al. (2013)
	Raw tomato juice	Power: 500 W Frequency: 20–25 kHz Temperature: 10 °C Time: 0–30 min	TPC progressively increased from 17.32 mg GAE/100 g to 21.60 mg GAE/100 g as CUT was extended to 30 min	Cold ultrasound treatment at low temperatures protects polyphenols	Gao et al. (2019)
	Hand-pressed strawberry juice	Frequency: 25 kHz Temperature: 20 °C Time: 0, 15 and 30 min	The increase in TPC was from 81.76 mg/100 mg (GAE equivalent) up to 89.52 and 151.94 mg/100 mg in 15 and 30 min	The cell wall matrix in the juice pulp breaks down and the bound polyphenols in it may become unbound and become easily assayed	Bhat and Goh (2017)
	Watermelon juice	Power: 1500 W Temperature: 25–45 °C Time: 2–10 min	At higher treatment times of 10 min, TPC was significantly lower compared to the control ($p < 0.05$)	Temperature has a significant effect on TPC	Rawson et al. (2011)
	Blackberry juice	Power: 1500 W Frequency: 20 kHz Time: 15 and 25 min	US caused the increase of polyphenols in the dialyzed fraction of the samples, until 15% in the treatment at 80% amplitude 15 min (645.39 ± 26.74 mg GAE/100 g db)	The cavitation produced by ultrasound leads to the disruption of biological cell walls, which promotes the release of phenolic compounds	Ramírez Moreno et al. (2017)
PEF	Tomato Fruit	Frequency: 0.1 Hz Electric strength: 0.4–2 kV/cm	TPC increased ranges from 6.6% to 44.6%	MIPEF treatment induces plant stress response	Vallverdu-Queralt et al., 2012a,b
	Cranberrybush purée (Viburnum opulus)	Voltage: 30 kV Pulse duration: 40 ms Pulse width: 20 μs Electric strength: 3 kV/cm	TPC and TFC increased	Improved cell permeability, mass transfer and release of matrix-bound phenolic compounds	Ozkan et al. (2021)
	Wheat plantlet juice	Frequency: 1 kHz Pulse width: 80 μs Electric strength: 9 kV/cm	Significantly higher TPC and TFC	PEF intensity is higher than the cell membrane capacity, causing the cell membrane to rupture	Ahmed et al. (2021)
	frozen/thawed European blueberry (Vaccinium myrtillus L.)	20 μs monopolar square wave pulses Electric strength: E = 1, 3, 5 kV/cm Total specific energy input: W_T = 1, 5, 10 kJ/kg	The PEF treatment induced slightly higher release of polyphenols (up to +8.0%) and anthocyanins (up to +8.3%) compared to the control	Enhanced cell membrane permeabilization	Lamanauskas et al. (2015)
	Apple juice	Bipolar pulse (4 μs wide) Intensity: 35 kV/cm Frequency: 1200pps	PEF treatment resulted in 14.49% loss of phenolics	HTST treatment resulted in considerable phenolic loss (32.2%) compared to PEF treatment	Aguilar-Rosas et al. (2007)
	Apple juice	Intensity: 400 V/cm Time: 0–90 min	TPC increased first (15 min) and then decreased	PEF contributes to the activation and release of polyphenol oxidase	Grimi et al. (2011)

(continued on next page)

Table 1 (*continued*)

Technology	Material	Treatment conditions	Change of TPC	Mechanisms	Ref
SC-CO_2	Pomegranate juice	Temperature: 45 °C Time: 40 min Pressure: 12.7 MPa	TPC increased 22%	Increased permeability of cell membranes	Bertolini et al. (2020)
	Sugar-preserved orange peel	Temperature: 31 °C Time: 20 min Pressure: 4, 7.4 and, 9 MPa	TPC increased by 1.09, 1.07, and 1.21 times than control	The huge pressure difference between the inside and outside the cell leads to slow oxidation of phenolics	Zhang et al. (2021)
	Beetroot juice	Temperature: 31, 39 or 55 °C Time: 10, 20, 30 min Pressure: 10, 30, 60 MPa	The highest degradation of polyphenols occurred at 60 MPa for 30 min at 55 °C	Effect of SC-CO_2 on enzyme inactivation	Marszałek et al. (2017)
	Apple juice	Temperature: 45 °C Time: 30 min Pressure: 10, 30, 60 MPa	Elevated degradation of phenolic compounds due to increased pressure	The solubility of polyphenols in CO_2 decreases as the pressure increases	Marszalek et al. (2018)

Abbreviations: CP: cold plasma; GAE: gallic acid equivalents; HPP: high-pressure processing; MIPEFs: moderate-intensity pulsed electric fields; PEF: pulsed electric field; POD: peroxidase; PPO: polyphenol oxidase; SC-CO_2: supercritical carbon dioxide; TFC: total flavonoid content; TPC: total phenolic content.

Table 2
Bioavailability of polyphenols in fruits and vegetables by different non-thermal processing techniques.

Technology	Material	Treatment conditions	Bioavailability of polyphenols	Mechanisms	Ref
HPP; HIPEFs	A blend of fruit juices (orange, pineapple, kiwi and mango)	400 MPa/5 min Temperature: 40 °C; Electric field strength: 35 kV/cm Width: 4-μs pulse Frequency: 200 Hz Time: 1800 μs Temperature: below 35 °C	38% increase in bioavailability of several phenolics (caffeic acid and p-coumaric acid from WB and MB; chlorogenic acid and ferulic acid from MB; hesperidin and rutin from all beverages)	Processing alters some of the physicochemical characteristics of phenolic compounds and, therefore, it can alter (increase or decrease) the bioavailability of these compounds	(Rodríguez-Roque et al., 2015)
HPHP	Mandarin juice	Homogenization at 20 MPa	HPHP increased bioaccessibility after in vitro digestion ($P < 0.001$)	The reduction in juice particle size facilitates the release of bioactive substances from the matrix; Molecular interactions between flavonoids and food substrates	Di Nunzio et al. (2020)
HPHP	Apple, grape and orange juice	High-pressure homogenized at 250 MPa for 10 min	Reduced the bioavailability of total phenols in apple juice by 29.3% and effectively improved the TPC of undigested grape and orange juice	HPHP could have ruptured the cellular structure and promoted the release of polyphenol, while inducing the oxidation, epimerization, and degradation of the fruit polyphenols	He et al. (2016)
Ultrasound	Blackberry juice	Power: 1500 W Frequency: 20 kHz Time: 15 and 25 min	The 80% amplitude treatment for 15 min increased the bioavailability of polyphenols	Promotes the release of polyphenolic compounds with higher absorption	Ramírez Moreno et al. (2017)
PEF	Fruit juice-Stevia rebaudiana mixture	Electric field strength: 25 kV/cm Temperature: below 35 °C	PEF treatment showed the highest stability of TPC (75.3%)	Acidic pH preserves better polyphenols in foodstuff	Buniowska et al. (2017)

Abbreviations: CP: cold plasma; HIPEFs: high-intensity pulsed electric fields; HPHP: high-pressure homogenization processing; HPP: high-pressure processing; PEF: pulsed electric field; SC-CO_2: supercritical carbon dioxide; TPC: total phenolic content.

Table 3
Specific working energy cost estimations for thermal and non-thermal technologies with no heat recovery.

Technology	Treatment (heating/pressurizing + pumping)	Product cooling	Heat dissipation	Total	Ref
Conventional thermal treatment	211.39 kJ/kg	186.15 kJ/kg	0 kJ/kg	397.54 kJ/kg	Vignali et al. (2022)
HPP	339.94 kJ/kg	0 kJ/kg	0 kJ/kg	339.94 kJ/kg	Vignali et al. (2022)
CP	87.78 kJ/(kg·s)	0 kJ/(kg·s)	0 kJ/(kg·s)	87.78 kJ/(kg·s)	Ranjbar Nedamani and Hashemi (2022)
PEF	161.81 kJ/kg	140.0 kJ/kg	0 kJ/kg	301.81 kJ/kg	Vignali et al. (2022)

Abbreviations: CP: cold plasma; HPP: high-pressure processing; PEF: pulsed electric field.

high-pressure homogenization processing (HPHP) on the phenolic bioaccessibility of apple, grape, and orange juice and observed that HPHP reduced the total phenolic bioavailability of apple juice by 29.3% (He et al., 2016). This difference may be due to the HPHP-induced oxidation, exopolymerization, and degradation of polyphenols (Suarez-Jacobo et al., 2011).

4.2. Cold plasma (CP)

Plasma is the fourth state of matter and is an ionized or partially ionized gas composed of various chemically reactive species such as electrons, atoms, molecules, electrically charged ions, free radicals, photons, and visible light (Misra et al., 2016). Plasmas are classified as thermal or non-thermal based on their mechanism of generation and the relative temperature between electrons, ions, and particles with no electric charge. Non-equilibrium plasma, also called CP, is produced by electrical discharge in gases. The principal mechanism of plasma technology is the generation of reactive species, particularly reactive oxygen (ozone and O2-) and nitrogen (such as NO•) reactive species, which cause pore formation and cellular membrane disruption, irreversible DNA damage, induction of apoptosis, and alterations in vital endogenous protein pathways (Liao et al., 2017).

Illera et al. (2019) reported that the TPC of cloudy apple juice elevated by 69% and 64% at 10.5 kV for 4 and 5 min of treatment, respectively (Illera et al., 2019). The TPC and catechin content of green tea increased by 41.14% and 103.12%, respectively, after 15 min of nitrogen dielectric barrier discharge (DBD) cold plasma at 15 W of power generation (Keshavarzi et al., 2020). However, the TPC of apples assessed by HPLC-MS/MS was not significantly different from that of the control group. The effect of CP treatment on phenolic compounds was attributed to the synergistic effects of various reactive substances. The collective impact of numerous plasma-reactive substances, including N_xO_y, O_3, O_{2^-}, and OH, produce variations in the content of polyphenolic substances (Ramazzina et al., 2016). The increase in phenolic compounds could be due to CP-generated energy and plasma reaction substances, which promote cell membrane degradation and the release of phenolic substances (Rodríguez et al., 2017). However, oxygen radicals released in CP can also attack phenolic compounds, leading to their degradation. CP treatment also affects the activity of polyphenol oxidase (PPO) and peroxidase (POD), resulting in a decrease in PPO and POD activities, whereas prolonging the treatment time increases enzyme inactivation (Farias et al., 2020).

4.3. Ultrasound

Ultrasound is a sound wave with a frequency greater than 20 kHz. Ultrasonic cavitation is a unique physical phenomenon caused by the propagation of powerful ultrasounds in liquids. The energy generated by cavitation produces high-pressure, high-temperature, and high-gradient flow locally, which effectively disrupts the cell wall structure and promotes the diffusion of polyphenols (Maran and Priya, 2014). Ultrasound, as a non-thermal food processing technology, may be used to inactivate microbes and enzymes. It has a positive impact on food processing, including food preservation, mass transfer improvement, heat treatment assistance, texture modification, and food analysis (Soltani et al., 2019). Ultrasound applications in the food industry include cutting, freezing, drying, homogenization, foaming and defoaming, filtration, emulsification, and auxiliary extraction (Chavan et al., 2022). It can be combined with other treatments, such as pressure and temperature, to improve overall process efficiency.

A significant increase in TPC in ultrasound-treated apple juice at 20 °C for 30–90 min at a frequency of 25 kHz compared to that in the control sample (Abid et al., 2013). Cold ultrasound treatment of raw tomato juice (at a power of 500 W and a frequency of 20–25 kHz) gradually elevated the TPC from 32.100 mg GAE/21 g to 60.100 mg GAE/30 g when the treatment duration increased to 17 min (Gao et al., 2019). Bhat and Goh (2017) demonstrated that sonicated hand-pressed strawberry juice samples (0, 15, and 30 min at 20 °C, 25 kHz frequency), augmented the TPC from 81.76 mg/100 mg (GAE equivalent) to 89.52 and 151.94 mg/100 mg in 15 and 30 min, respectively (Bhat and Goh, 2017). When the treatment temperature, power, and time of ultrasound reach a particular threshold, the rupture of the cell wall tends to saturate, and excess ultrasound energy destroys additional phenolic compounds, while too high a temperature (60 °C) may also contribute to phenolic compound degradation (Rostagno et al., 2007). Thermal ultrasound treatment of watermelon juice with processing variables of temperature (25–45 °C), amplitude level (24.1–60 μm), and processing time (2–10 min) at a constant frequency of 20 kHz and pulse durations of 5 s on and 5 s off decreased the TPC with increasing temperature (Rawson et al., 2011). Ultrasound also has a significant impact on enzyme activity. High-intensity sonication has been shown to promote PPO enzyme inactivation by inducing aggregation and structural changes (Liu et al., 2017; Iqbal et al., 2020).

Ultrasound treatment of blackberry juice did not improve TPC but caused the increase by 15% in the dialyzed components of blackberry juice after digestion in vitro. The bioaccessibility and antioxidant activity of polyphenols in dialyzed components were both increased. This may be because the cavitation produced by ultrasound destroys the biological cell wall and promotes the release of polyphenols (Ramírez Moreno et al., 2017). However, ultrasound may have destroyed the polyphenols to some extent, changing their structure and breaking the polyphenols down into small molecules that are easily absorbed.

4.4. Pulsed electric field (PEF)

PEF is used to treat liquid and semi-solid foods with a higher electric field intensity (10–50 kV/cm), a shorter pulse width (0–100 μs), and a higher pulse frequency (0–2000 Hz). The PEF generates a magnetic field. When the pulsed electric and magnetic fields alternate, the cell membrane electroporates, and the material inside the membrane readily flows out. PEF has progressed from the laboratory and pilot plant stages to the industrial level as a potential non thermal food processing technology (Niu et al., 2020). PEF can be used alone or in conjunction with other types of processing. Its primary applications include microbe and enzyme inactivation, extraction of active ingredients (Xi et al., 2021), biomolecule modification (Zhang et al., 2021), chemical reaction augmentation, and accelerated aging of fermented foods (Feng et al., 2022).

The alteration of plant cell membrane permeability by PEF facilitates an increase in the content of bioactive compounds (e.g., polyphenols). Vallverdú et al. (2012) studied the effect of pulsed electric fields on the TPC of tomato juice (Vallverdu-Queralt et al., 2012a,b). They exposed a batch of tomatoes to moderate-intensity pulsed electric fields (MIPEFs) before immediately refrigerating them at 4 °C for 24 h. The treated and untreated juices were subjected to high-intensity pulsed electric fields (HIPEFs) or heat treatment (90 °C for 60 s). The results revealed that the highest TPC was found in tomato juice treated with the combination of MIPEF and HIPEF. The HIPEF-processed tomato juice contained higher levels of polyphenolic compounds (ferulic acid, caffeine-O-glucosinolates, p-coumaric acid, chlorogenic acid, rutin, and naringenin) than heat-treated tomato juice after processing and storage. Meanwhile, PEF can increase the content of polyphenol compounds (Lamanauskas et al., 2015; Ahmed et al., 2021; Ozkan et al., 2021). Recent research has shown that MIPEFs may be used to stress cells and induce the biosynthesis of secondary metabolites, such as polyphenols (Vallverdu-Queralt et al., 2012a,b), which is a plant response to stress. PEF has also shown examples of reduced polyphenol content (Grimi et al., 2011; Rybak et al., 2020). The explanation may be that the PEF treatment provides enough energy to perforate the cell membrane, and it promotes contact between the PPO in the plastid and the natural polyphenols in the vacuole. The energy of PEF may affect the structure of phenolic compounds and cause oxidation (Turk et al., 2010).

Current studies suggest that PEF treatment is an effective method for improving the bioaccessibility or bioavailability of phenolic compounds. Buniowska et al. (2017) revealed that PEF treatment increased TPC bioaccessibility in a fruit juice-Stevia rebaudiana mixture, most likely because it enhanced polyphenol release from the food matrix without the formation of new electrolytic products (Buniowska et al., 2017). In the HIPEF treatment of fruit juice-based beverages, there was an observed 38% increase in the bioaccessibility of several phenolic substances (caffeic and p-coumaric acids from both water-fruit juice beverage and milk-fruit juice beverage; chlorogenic and ferulic acids from milk-fruit juice beverage; hesperidin and rutin from all beverages). However, HIPEF reduced the bioaccessibility of some other phenolic compounds by 10–11% (chlorogenic and p-hydroxybenzoic acids from water-fruit juice beverage). (Rodríguez-Roque et al., 2015; Cilla et al., 2018). HIPEF, which has a higher intensity than PEF, affects the physicochemical features of various phenolic compounds differently, such as the phenol structure (hydroxylation, methylation, isoprenylation, dimerisation, and glycosylation, etc.) and/or the formation of phenol derivatives (by partial degradation of the combined forms, etc.). These changes ultimately impact the bioaccessibility of the phenolic compounds. It is possible that the general guidelines have certain conditions of application and do not apply to all specific case studies; therefore, further research in this field is needed.

4.5. Supercritical carbon dioxide (SC–CO_2)

SC-CO_2 is a carbon dioxide fluid maintained at a critical temperature and above a critical pressure. Supercritical fluids have a density comparable to that of liquids, a viscosity close to that of gases, and a diffusion coefficient higher than the liquids. These physical properties, combined with the absence of surface tension, result in high solubility and diffusion coefficients. The development of non-thermal processes is better suited to SC-CO_2 since it has a lower critical temperature and pressure than HPP.

Previous studies have indicated that SC-CO_2 treatment increases TPC levels in fruits and vegetables. Pomegranate juice treated with SC-CO_2 (45 °C for 40 min at 12.7 MPa pressure) raised TPC by 22% (Bertolini et al., 2020). The residual TPC of HPCD-treated orange peels was 1.09-, 1.07-, and 1.21-fold greater than that of untreated samples ($p < 0.05$) (Zhang et al., 2021). The mechanism could be that SC-CO_2 dissolves in water and gradually forms carbonic acid (Deotale et al., 2021), and a decrease in solution pH increases permeability of cell membranes (promoting the release of phenolics) and disrupts the structure of enzymes, thereby inactivating PPO and POD (preventing phenolic oxidation) (Xu et al., 2021). The high solubility and diffusion coefficient of SC-CO_2 allow it to rapidly infiltrate the cell structure. Studies have demonstrated that the dissolution of CO_2 in fruits and vegetables accelerates with increasing pressure and temperature. Under certain conditions, SC-CO_2 treatment of beet juice increased polyphenol content, but under others, polyphenols degraded (Marszałek et al., 2017). At shorter times (10 min), the TPC was lower than that of the control, and this loss could be compensated by either increasing the pressure or temperature. However, long time (30 min) and the high pressure (60 MPa) may not be possible to reverse this polyphenol loss. SC-CO_2 treatment of apple juice at increasing pressures (10, 30 and 60 MPa) resulted in increased degradation of phenolic compounds (Marszalek et al., 2018). Moreover, the solubility of polyphenols in CO_2 decreases as the pressure level increases (Zhang et al., 2021).

5. Influencing mechanism of non-thermal processing technology on polyphenols

5.1. Disruption of cell walls or cell membranes

Cell wall or cell membrane disruption is the most common and fundamental processing mechanism for non-thermal processing techniques. They cause damage to the cell wall or cell membrane using high energy rather than heat (e.g., high pressure), permitting the release of polyphenolic compounds from the cell, thus increasing the TPC. An increase in the TPC promotes bioaccessibility and bioavailability. Scanning electron microscopy revealed cell ablation and rupture on the surface of green tea leaves following nitrogen DBD cold plasma treatment in a study of CP-treated green tea leaves. Water penetrates the surface more easily and increases the TPC through the formation of cracks and pores (Keshavarzi et al., 2020). Frozen/thawed European blueberry (*Vaccinium myrtillus* L.) fruits were exposed to 20 μs monopolar square wave pulses with varying electric field strengths (1, 3, 5 kV/cm) and total specific energy inputs (1, 5, 10 kJ/kg), and their permeabilization was assessed using electrical impedance measurements and the cell disintegration index. With increasing PEF treatment intensity (E and W_T), the cell disintegration index increased significantly ($p < 0.05$) from 0.2 to 0.6 (Lamanauskas et al., 2015).

5.2. PPO and POD activities

Another major aspect is the influence of non-thermal processing techniques on enzymes, as certain enzymes (such as PPO and POD) in plant cells cause the oxidation of polyphenolic substances. Polyphenols are well protected when polyphenol-related enzymes are disrupted or even inactivated using non-thermal processing techniques. Enzymes are usually inactivated when their secondary and tertiary structures are disrupted. This disruption could be caused by various physical (e.g., heat, high pressure, and ultrasound) and chemical factors.

The tertiary structure of the enzyme, which is maintained to a significant degree by electrostatic and hydrophobic interactions, is disrupted by HPP treatment at higher pressures. This process is accompanied by the penetration of water into the core of the protein molecule, resulting in a loss of contact between the non-polar structural domains of the molecule, culminating in conformational changes, partial unfolding, and loss of activity (Nagae et al., 2012). High pressures of 150–200 MPa cause quaternary structural rupture, resulting in the dissociation of the oligomeric enzyme into individual subunits. High pressure-induced denaturation can leave part of the enzyme structure intact; therefore, HPP alone is more likely to limit enzyme activity than inactivate it (Terefe et al., 2016). PPO is one of the most pressure-tolerant enzymes and typically requires pressure over 600 MPa as well as moderate temperatures and time to inactivate (Houska et al., 2022b). The relative sensitivity of POD and PPO varies depending on the substrate. However, under mild temperature conditions, high pressure may increase PPO and POD activity, possibly due to stress causing tissue fragmentation and the release of membrane-bound enzymes, as well as changes in protein structure leading to the activation of potential forms of the enzymes (Terefe et al., 2014).

The oxidation of reactive side chains of amino acids by plasma free radicals, particularly OH, O^{2-}, HOO, and NO, promotes changes in secondary protein structure and certain amino acid side chains of the enzyme, and could be the reason for the decrease in enzyme activity induced by CP treatment. However, there are reports of increased enzymatic activity. Hydroxyl radicals (OH-) generated by hydrolysis leads to increased POD activity. This phenomenon explains the damage to the cell membranes of fruits and vegetables caused by ROS generated by CP treatment. The natural response of these plants is to increase the production and activity of antioxidant enzymes, such as POD, PPO, and superoxide dismutase (SOD) (Yi et al., 2022). The reduced permeability of plasma also indicates an increase in POD activity (Misra et al., 2014).

At the molecular level, ultrasound can facilitate or damage enzymes, substrates, reactions between enzymes and substrates, and their surroundings. Ultrasound produces cavitation, magnetostrictive effects, and mechanical oscillations at low intensities and suitable frequencies. This process changes the conformation of the enzyme and accelerates the interaction between the enzyme and substrate, thus promoting its biological activity. In contrast, ultrasound can inactivate enzymes. Enzyme inactivation produced by ultrasound treatment is attributed to acoustic cavitation, which causes a rise in local pressure and temperature, as well as strong shear stress, resulting in altered secondary and tertiary changes in the protein (Feng et al., 2011). No significant reduction in activity was detected when the treatment temperature was lower than the denaturation temperature of PPO and POD (40–50 °C and 60 °C, respectively) (Bot et al., 2018). Enzyme inactivation was achieved only below the denaturation temperature for a sufficiently long time. This finding implies that the acoustic effects of ultrasound treatment may be insignificant and that heat directly induces enzyme inactivation.

Ultrasound treatment facilitates the release of enzymes from cells, which may boost enzyme activity while also increasing susceptibility to thermal inactivation (Bi et al., 2015).

PEF may alter the structure of enzymes, reducing their activity (Qian et al., 2016). It can change the secondary bonds that hold the enzyme together, and excessively high treatment conditions can cause helical structural disruption. A loss of α-helix and surge of β-page content in the secondary structure of the enzyme results in enzyme inactivation (Huang et al., 2012). PEF treatment may deactivate PPO and POD, thereby retaining phenolic compounds (Tian et al., 2018). HIPEF treatment, on the other hand, did not reduce β-glucosidase activity. Instead, it induced a slight increase activity when treated at 35 kV/cm for 1700 μs using square-wave pulses of 4 μs and a pulse frequency of 100 Hz (Aguiló-Aguayo et al., 2009). The activity increase may be due to the formation of more active sites or an increase in the size of existing sites (Ho et al., 1997).

The major causes of enzyme inactivation by SC-CO_2 are pH reduction and conformational changes in the secondary and tertiary enzyme structures. It has also been reported that the interaction of CO_2 with the (typically hydrophobic) protein core is crucial in understanding the inactivation process (Li et al., 2014). Proteins fold entirely to shield the hydrophobic core and its function from the surrounding aqueous medium; nevertheless, the addition of a non-polar molecule, such as CO_2, might alter the equilibrium.

5.3. Free radical reaction

Free radicals (O, OH, etc.), excited or reactive molecules (O2-, O3, NO, etc.), or charged particles (electrons and atomic or molecular ions) generated by ionized gases bombard the cells, causing pores or even cells to rupture, and the contents, such as polyphenols, to flow out. This is why CP treatment increases the TPC (Cao et al., 2021). However, when phenolic compounds are hydroxylated by reactive substances, hydroxyl ring carbonyl radicals are formed, which can set off a chain reaction that damages polyphenols (Abdel-Aziz and El-Hadary, 2022).

5.4. Plant stress response

Higher plants produce secondary metabolites that are not directly involved in the primary metabolic processes of plant growth and development but play a significant role in their adaptability to changing surroundings. They can defend predators, microbial attacks, plant competitors, and abiotic stresses including UV, ozone, and herbicides. Non-thermal processing techniques, such as ultrasound, CP, and PEF, can elicit abiotic stress responses in plants (Hasan et al., 2017). After 24 h of MIPEF treatment, potato tissue metabolism exhibited a plant stress response, as evidenced by alterations in polyphenols, amino acids, and hexose pools (Galindo et al., 2009). Under abiotic stress conditions, plants exhibit an increased synthesis of polyphenolic substances, such as phenolic acids and flavonoids, which help them cope with adverse environments. The antioxidant activity of phenolic compounds scavenges free radicals and thereby minimizes cell membrane peroxidation, protecting plant cells from the harmful effects of oxidative stress. The altered activity of various prime enzymes of the phenolic biosynthetic pathway, such as chalcone synthase, regulates phenolic production in stressful conditions.

5.5. Matrix interactions in fruits and vegetables

In addition to polyphenols, non-thermal processing techniques also affect other macromolecular components (e.g., polysaccharides, proteins, and lipids), that interact with polyphenols and either positively or negatively influence their release and bioavailability.

Fruits and vegetables are high in dietary fiber, which consists of different polysaccharides (e.g., cellulose, hemicellulose, and pectin polysaccharides) that form cell walls. Some phenolic compounds are bound to hemicelluloses in cell walls via hydrogen bonds and hydrophobic interactions (Zhu, 2018). Evidence suggests that polysaccharides in food interfere with phenolic compound assimilation. Phenolic compounds are more readily released from food matrices with low dietary fiber content, such as juices and beverages (Palafox-Carlos et al., 2011). Non-thermal processing techniques disrupt plant cell walls and act on polysaccharides (Liu et al., 2020). Polyphenols readily bind to plant cell wall polysaccharides, thereby disrupting the cell wall and facilitating the release of polyphenols (Phan et al., 2015). Such interactions can also occur through gastrointestinal mechanisms. Approximately 50% of phenolic intake in the Western diet is associated with dietary fiber, which can be absorbed only when the interaction is effectively hydrolyzed by the colonic microbiota.

Non-thermal processing techniques cause structural changes in proteins (including enzymes) in plant cells, altering their physiological activity (Barbhuiya et al., 2021). This may have an impact on their interactions with polyphenolic compounds(Xiao et al., 2023). Moreover, non-thermal treatments that modify the temperature and pH of the environment might influence their possible interactions (Ozdal et al., 2013). Most of these interactions are noncovalent, such as hydrogen bonding and hydrophobic interactions. Studies on protein-polyphenol interactions and their impact on bioaccessibility or bioavailability have shown inconsistent results, including negative, neutral, and positive effects (Jakobek, 2015). Based on the influence of non-thermal techniques on the interaction of polyphenols with proteins, it can be hypothesized that various non-thermal processing techniques under different treatment conditions result in the binding of polyphenols and proteins (especially enzymes) by covalent or non-covalent bonds, leading to diverse outcomes on contents and bioavailabilities of polyphenol.

Non-thermal processing treatments accelerate lipid oxidation (Barbhuiya et al., 2021). However, the lipid content of fruits and vegetables is often minimal, and the interaction between lipids and polyphenols has a limited effect on their bioaccessibility and bioavailability (Jakobek, 2015). Fruits and vegetables include vitamins in general, particularly vitamin C, which has antioxidant properties. Vitamin C is susceptible to degradation due to temperature, pH, metal ions, and other factors. The variation in vitamin C content following non-thermal processing was similar to that of polyphenols (Ozkan et al., 2021). It can synergistically protect polyphenols from oxidation while also maintaining or increasing polyphenol content to some extent.

The components of the mechanism described above do not exist independently but are interconnected. These proteins form a complex network system that regulates the polyphenol content and bioavailability in non-thermally processed treatments (Fig. 2).

6. Conclusion

The emphasis on active compounds such as polyphenols has resulted in a preference for fruit and vegetable products with better nutritional value. Emerging non-thermal processing technologies are effective at conserving nutrients in fruits and vegetables while enhancing the content and bioavailability of polyphenols. Various non-thermal processing technologies have different principles and effects on fruits and vegetables. However, there are some limitations in this review. It lacks a comprehensive overview of the effects of non-thermal processing techniques on polyphenols in various fruits and vegetables, and the explanation of the mechanism is superficial. Many bioavailability studies primarily focus on in vitro bioaccessibility studies (simulations of gastrointestinal digestion), which are not as accurate and reliable as in vivo bioavailability studies. Based on the characteristics of different non-thermal processing technologies, appropriate choices may be made to meet the requirements of experimental or industrial production. However, to prevent unnecessary losses, it is essential to consider the drawbacks of various non-thermal processing techniques when polyphenolic compounds are present. Industrial manufacturing must also prioritize the development of systems suitable for industrial

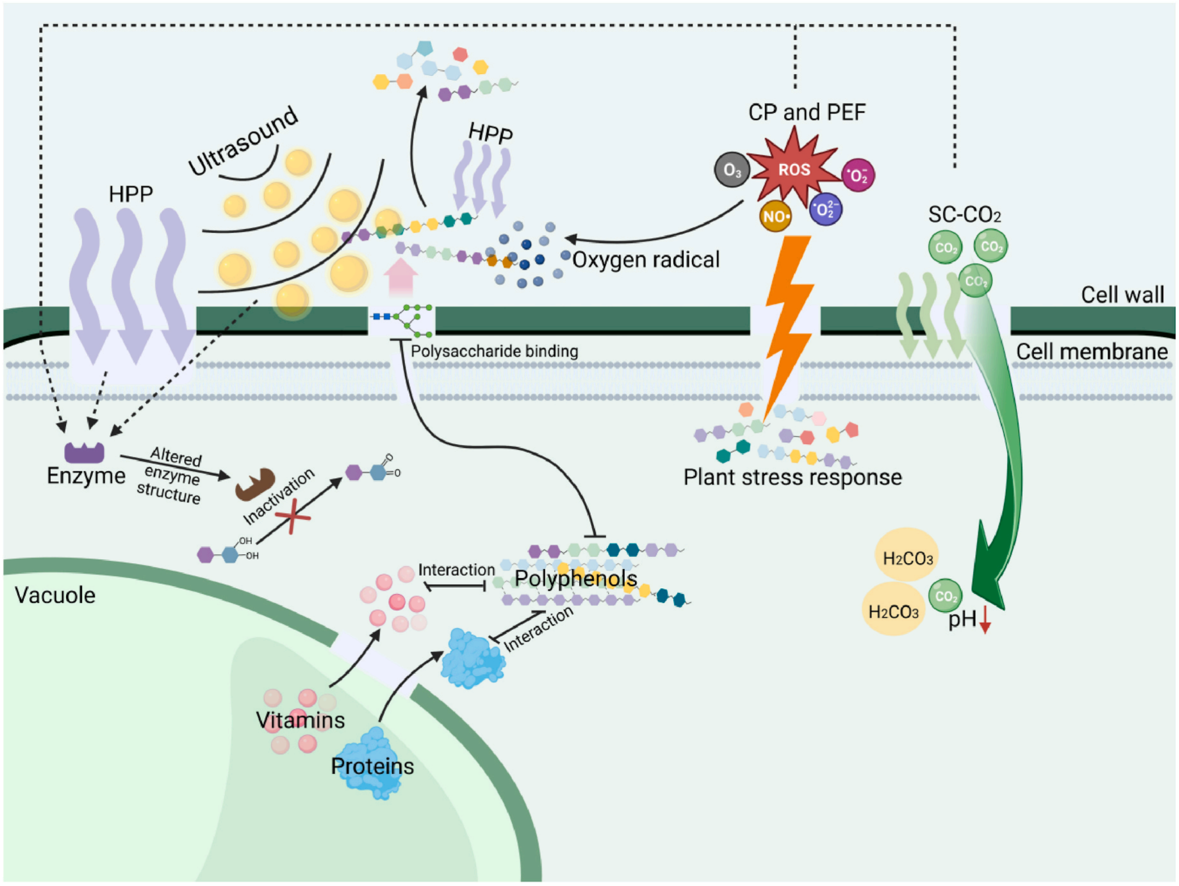

Fig. 2. Mechanism of the effect of non-thermal processing techniques on polyphenols in fruits and vegetables. CP: cold plasma; HPP: high-pressure processing; PEF: pulsed electric field; ROS: reactive oxygen species; SC-CO_2: supercritical carbon dioxide.

applications. In conclusion, emerging non-thermal processing technologies are more promising for the food industry than thermal treatment for the preservation, extraction, and bioavailability of polyphenols in fruits and vegetables.

CRediT authorship contribution statement

Yichen Liu: Conceptualization, writing and revision. **Jianjun Deng:** Supervision, Writing – review & editing. **Tong Zhao:** Writing – review & editing, revision. **Xiaojie Yang:** Writing – review & editing. **Juntao Zhang:** Writing and revision. **Haixia Yang:** Supervision, Writing – review & editing, Funding acquisition.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

No data was used for the research described in the article.

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Abbreviations

CP	cold plasma
DBD	dielectric barrier discharge
GAE	gallic acid equivalents
HIPEFs	high-intensity pulsed electric fields
HPHP	high-pressure homogenization processing
HPP	high-pressure processing
MIPEFs	moderate-intensity pulsed electric fields
PEF	pulsed electric field
POD	peroxidase
PPO	polyphenol oxidase
ROS	reactive oxygen species
SC-CO_2	supercritical carbon dioxide
SOD	superoxide dismutase
TFC	total flavonoid content
TPC	total phenolic content

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