



Role of Plant Growth Regulators and Eco-Friendly Postharvest Treatments on Alleviating Chilling Injury and Preserving Quality of Pomegranate Fruit and Arils: A Review

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Abstract

Punica granatum belongs to the Lythraceae family is one of the most important subtropical fruits native to Iran. Although the production of fruit has increased recently, there is still a gap between demand and supply. Improper handling, transportation, packaging and storage, mechanical damage, and susceptibility to chilling injury and its related physiological disorders during long-term storage are the most important causes of pomegranate postharvest losses. Fruit quality is lost with visible symptoms such as weight loss, shriveling, husk scald, fungal rot, aril color degradation, and off-flavor during long-term storage. Preserving the quality is the most important goal of the postharvest physiology industry. To minimize both qualitative and quantitative postharvest losses, it is crucial to apply appropriate knowledge and technologies during both the harvest and postharvest stages of pomegranate production. This helps to maintain the quality and shelf life of the fruit. This paper reviewed recent studies that used simple, eco-friendly, synthetic and organic plant growth regulator treatments in underdeveloped and developing countries, including proper packaging according to consumer demand and safe preservatives application, which significantly reduces postharvest losses and improves overall quality of pomegranate fruit and arils.

Keyword Edible coatings · Heat treatment · Micronutrients · Packaging · Plant growth regulators · Storability

Introduction

Daily consumption of fruits and vegetables is essential for human health due to their nutritional and bioactive compounds (Fraga et al. 2019). Pomegranate is an edible and medicinal fruit with high economic and nutritional value and rich in phenolic compounds, antioxidants, sugars, vitamins, organic acids, unsaturated fatty acids (unSFAs), minerals, and fiber (Mahesar et al. 2019; El-Mahdy et al. 2022). Pomegranate has an anti-inflammatory effect and reduces high blood pressure (Barati Boldaji et al. 2020; Pfohl et al. 2021), and also effective in preventing heart diseases, diabetes, and

cancer (Kushwaha et al. 2020; Stawarska et al. 2020; Chaves et al. 2020).

Fresh fruit losses are increasing in post-harvest processes (FAO 2019). Pomegranate fruits show chilling injury (CI) symptoms at temperatures below 5 °C, including rotting, browning, and cracking of the peel. In addition to CI, weight loss during storage causes the hardness of the peel and seeds, wrinkling, and senescence (Caleb et al. 2012, 2013). Weight loss and microbial decay due to transpiration and respiration are among the main problems of post-harvest storage (Kahramanoglu 2017). Long-term storage of pomegranate arils causes more weight loss due to lower resistance of the cell membrane against water loss (Belay et al. 2018). Fruit water loss during storage results in husk browning (Nerya et al. 2006). Additionally, polyphenol oxidase (PPO) and peroxidase (POD) activity enhances the brown superficial discoloration of pomegranate fruits (Baghel et al. 2021), which is the primary cause of quality decrease (Ioannou and Ghouli 2013). Weight loss increases CI symptoms by destroying the membrane integrity (Maghoumi et al. 2023). Damage to the membrane structures and a lack of resistance to cold are results of the decrease in unSFAs content and

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membrane fluidity, and it has been reported electrolyte leakage in the pomegranate peel has been linked to CI symptoms (Casares et al. 2019). Packaging and edible coatings reduces the vapor pressure difference between the surface and environment of the product by maintaining the relative humidity around the fruit, accordingly reducing the water loss of the product (Ngcobo et al. 2013). Also, nanoparticle technology helps to increase shelf life and reduce waste due to the controlled release of nutrients (Ding et al. 2022). Nano-elements can maintain antioxidant capacity with their antibacterial effects in food packaging (Sirelkhatim et al. 2015; Saba and Amini 2017). In addition, plant growth regulators improve tolerance to abiotic stresses by scavenging or reducing the accumulation of active oxygen species (AOS), electrolyte leakage, and expression of stress-specific genes (Rachapanaavar et al. 2022).

Pomegranate has a short ripening period with low storability (Ozdemir and Gokmen 2017; Melgarejo-Sanchez et al. 2021), and considering the nutritional value of pomegranate, maintaining its quality and nutrients is a research priority. The requirement to increase the shelf life of fresh fruits is to minimize the rate of biochemical reactions and enzymatic and microbial degradation (Kirandeep et al. 2018; Kumar et al. 2020). Therefore, this review aimed to investigate the mechanism of action of the main practical post-harvest treatments, which influence the quality and storage life of pomegranate fruit (Table 1).

Post-Harvest Management

Packaging Films

Food packaging preserves nutritional value by preventing contact with spoilage agents such as microorganisms, oxygen, and moisture (Khan et al. 2021). Films to improve the physicochemical properties and shelf life of fruits developed, and the usage of synthetic and semi-synthetic polymers is common (Ferreira et al. 2020; Shen et al. 2020). Polymer films are used for packaging due to their low production cost and excellent barrier properties against moisture and gases (Azeem et al. 2022; Dissanayake et al. 2022). The polymer film was successfully used on pomegranate fruit and improved overall quality and storage life of arils (Moradinezhad et al. 2018, 2020). Packaging fruit with micro- and macro-perforation high-density polyethylene (HDPE) reduced postharvest losses by minimizing moisture condensation, fruit rot, and shriveling (Lufu et al. 2021). Fruit packaged in the micro-perforated Xtend® had the most negligible weight loss and respiration rates compared to unpacked fruit (Kawhena et al. 2022). Arils packed with Xtend® maintained phenol, anthocyanin, ascorbic acid, and antioxidants compared to low-density polyethylene (LDPE)

and polypropylene (PP). Also, the organoleptic quality increased due to the reduction of water loss and preservation of color (Dhineshkumar et al. 2017). Arils packed in the semi-permeable films had high polyphenols, anthocyanins contents, enzymatic activity (superoxide dismutase (SOD), catalase (CAT), and ascorbate peroxidase (APX)), and low PPO and POD activity (Adiletta et al. 2019). In a similar study, silver nano-bag maintained the taste, aroma, overall acceptability, anthocyanin, vitamin C, and antioxidant activity and reduced pectinase activity compared with Xtend bag, polyethylene bag, and polypropylene bag (EL-Eryan 2020). Passive modified atmosphere using XTend™ bags increased the anthocyanin concentration in the peel and arils and delayed the symptoms of CI (Valdenegro et al. 2022). In addition, vacuum (Moradinezhad et al. 2019) and modified atmosphere (Dorostkar and Moradinezhad 2022) packaging using LDPE bags significantly maintained fruit quality and reduced losses of pomegranate fruit cultivar Shishe-Kab. Reduction of microbial contamination and maintain the quality of arils was observed in polyethylene-polyester bilayer film compared to polypropylene biaxial orientation (Ranjbar and Ramezani 2022). Packaging can lead to structure preservation, less tissue damage and extending shelf life of aril due to increased vapor pressure and reduced cell wall polysaccharides degradation (Zhao et al. 2019) (Table 1 and Fig. 1A).

Edible Coatings

Edible coatings can maintain quality by creating a semi-permeable barrier against gas and moisture exchange. Also, they may carry active components such as nanoparticles that have antimicrobial or antioxidant activity against bacteria and ultraviolet (UV) rays, respectively, to improve the properties of coatings (Aristizabal-Gil et al. 2019; Sharma et al. 2020; Firdous et al. 2023). Nano-stimulants can be natural or chemical plant extracts, nanocomposites containing macronutrients, micronutrients, or chitosan. Polysaccharide-based coatings are colorless and have low calories with antioxidant and antibacterial characteristics (Harkin et al. 2019).

Post-harvest application of chitosan coating reduced respiration rate, weight loss, and shriveling symptoms of the pericarp surface of pomegranate fruit (Varasteh et al. 2018). Combination of modified atmosphere packaging and chitosan coating significantly reduced weight loss and husk scald symptoms (Candir et al. 2018). Similarly, CH-24-epibrassinolide coating reduced weight loss, respiration rate, electrolyte leakage, and microbial spoilage, followed by delayed texture, color, and total soluble solids (TSS) degradation (Mwelase and Fawole 2022). Combined chitosan and potassium sorbate (PS) decreased the CI symptoms, electrolyte leakage, and malondialdehyde (MDA) contents of fruit peel. Furthermore, it enhanced the activity of DPPH

Table 1 Physiological and molecular mechanisms involved in maintaining post-harvest quality of pomegranate

Treatment	Physiological and molecular mechanisms	References
Packaging films	Decreasing respiration rate and ethylene production, and delay ripening; Reducing weight loss and husk scald; Less chilling injury (CI), Increase of antioxidant activity; lower changes in acidity and soluble solids content; Inhibition of microbial activity by reducing pH and intracellular activities	Selçuk and Erkan (2015), Candir et al. (2018), Serry (2019)
Edible coatings	Inhibiting oxidative reactions, decreasing respiration rate, enzymatic browning, and release of volatile compounds, increasing the content of phytochemicals and delaying senescence, maintaining the balance of intracellular oxidation metabolism due to the ability to remove cytotoxic compounds through enzymatic antioxidants and non-enzymatic antioxidants; Effect on germination and hyphae morphology of fungal pathogens, preventing the growth of pathogenic and mycoparasitic fungi by increasing the permeability of the plasma membrane and nutrient limitation carbon and nitrogen and as a result cell wall structure with low branching and membranes rich in free polyunsaturated fatty acids such as linolenic acid; Regulating the expression of genes related to the glycolysis pathway and controlling the balance of aerobic-anaerobic metabolism and reducing genes related to ethylene production and ripening, expression of pathogenesis-related proteins	Palma-Guerrero et al. (2010), Lopez-Moya et al. (2015), Sayyari et al. (2016), Beatrice et al. (2017), Kumar et al. (2017), Liu et al. (2017b), Wang et al. (2017), Liu et al. (2018), Resende et al. (2018), Adil-etta et al. (2021), Hira et al. (2022)
Micronutrients	Accumulation of enzymatic and non-enzymatic antioxidants; Protection against pathogens by biosynthesis of proteins, carbohydrates and regulation of hormones, delay completion of cell division and growth cycle of microorganisms	Broadley et al. (2007), Saba and Amini (2017)
Melatonin	Induction of cold resistance through the oxidative pentose phosphate pathway, regulation of phenolic metabolism and increasing capacity inhibiting DPPH and reducing oxidative damage, stimulation of ROS, followed by enhancement of antioxidants, improves the activity of APX and GR, and reducing the ROS; Reduction of CI by decreasing the activity of PPO and membrane-degrading enzymes D (PLD) and LOX and increasing PAL, CAT, APX and SOD, supply of intracellular NADPH by promoting the activities of G6PDH and 6PGDH, increasing expression of antioxidant genes and positive regulation of gene expression of essential enzymes responsible for phenylpropanoid pathways such as PAL, CHS1, CHS2 and PSH and accumulation of phenols and flavonoids for cold resistance, expression of membrane fatty acid-inducing genes, such as FAD3 and FAD7 genes contributes to a higher unSFA/SFA ratio and increases membrane integrity, encoding calcium-dependent protein kinases (CDPK) and mitogen-activated protein kinases (MAPK), induction of Ca ²⁺ signaling pathways, activation of C-repeat binding factors (CBFs) as transcription factors for cold resistance, delay senescence by suppressing the expression of ethylene biosynthesis genes PcACS1 and PcACO1, increasing disease resistance by upregulating genes related to jasmonic acid synthesis (VaLOX, VaAOS, and VaAOC), genes related to pathogenesis proteins (VaGLU and VaCHT) and genes related to phenylpropane metabolism (VaPAL, VaC4H, Va4CL, VaCAD, VaPPO, and VaD)	Sun et al. (2016), Gao et al. (2018), Zhai et al. (2018), Jannatizadeh (2019), Aghdam et al. (2020a), Madebo et al. (2021), Qu et al. (2022)

Table 1 (continued)

Treatment	Physiological and molecular mechanisms	References
Salicylic acid	Induction of defense responses including upregulation of resistance genes to cause systemic acquired resistance (SAR), regulation of expression of genes related to pathogenesis; CI resistance by inducing the expression of different sHSPs (class I and II families) and high molecular weight HSPs with stabilizing function on the cell	Ding et al. (2001), Santisree et al. (2020)
Methyl jasmonate	Activation of the antioxidant system and defense compounds (such as phenolic compounds and heat shock proteins); Reducing membrane damage caused by decreasing the activity of LOX, the enzyme responsible for the production of superoxide free radicals, reduction of lipid peroxidation, MDA accumulation and electrolyte leakage; Inducing the expression of proteins related to pathogenesis and enzymes related to defense such as chitinase and β -1,3-glucanase and changes in phenolic biochemistry; Regulating the expression of genes encoding secondary metabolite biosynthesis enzymes including polyamine, glutathione and anthocyanins for CI resistance; Expression of jasmonate-related genes (JAZ, AOS1, AOC, LOX2, and COI1), interactions with other plant hormones (ABA, ET, SA, GA, IAA, and BR), and interaction with TFs (MYC2 and bHLH148), expression of MYC TFs and cold-responsive genes (MaCBF1, MaCBF2, MaKIN2, MaCOR1, MaRD2, MaRD5)	Jin et al. (2009), Zhao et al. (2013), Jiang et al. (2015), Hu et al. (2017), Yang et al. (2019)
Oxalic acid	Induction of cold resistance with physiological and biochemical changes in the metabolism of fatty acids, antioxidants and proline, increasing the expression of proline biosynthesis genes and inhibiting proline degradation genes	Awad et al. (2013)
Sodium nitroprusside	Reducing ethylene synthesis by inhibiting the enzymes 1-aminocyclopropane-1-carboxylic acid synthase (ACS), 1-aminocyclopropane-1-carboxylic acid oxidase (ACO) and S-adenosylmethionine synthetase (SAMS) through nitrosylation and reducing sensitivity to ethylene and delaying senescence; Reducing oxidative stress by inducing enzymes such as SOD, POD and CAT and suppressing LOX; Reduction of CI by S-nitrosylation of proteins and modulation of antioxidant response, regulating the AsA-GSH circulatory system to balance redox and reducing the accumulation of ROS and lipid peroxidation; Induction of stress-related gene expression by synergistic interaction with signaling molecules, such as Ca^{2+} , ET, SA and JA; Inducing the activity of enzymes involved in energy metabolism; Maintaining quality by reducing the gene expression of the xyloglucan endotransglucosylase/hydrolase (XTH) family and reducing the activity of cell wall hydrolyzes enzymes such as polygalacturonase, xyloglucan endoglycosyltransferase, cellulase and β -galactosidase; Regulation of lipid metabolism by increasing expression of genes encoding sn-Glycerol-3-phosphate acyltransferase, β -ketoacyl-ACP synthase, phosphatidylinositol bisphosphate and long-chain acyl-CoA dehydrogenase; Disease resistance by inducing PAL, 4-coumarate-CoA ligase, and cinnamic acid 4-hydroxylase enzyme activity, accumulation of antifungal compounds (such as phenylpropanoic acids, flavonoids, phenolics, and lignin) and induction of H_2O_2 accumulation	Ma et al. (2019), Yan et al. (2019), Zhao et al. (2021), Zuccarelli et al. (2021), Liu et al. (2023)

Table 1 (continued)

Treatment	Physiological and molecular mechanisms	References
Gamma-aminobutyric acid	Regulation of physiological responses by the interaction of signaling molecules including Ca ²⁺ , phytohormones, amino acids proline and polyamines; Reducing oxidative damage by regulating the transcription of antioxidant enzymes genes; AaGAD1 and AaGAD4 gene expression and endogenous GABA biosynthesis, decreasing the enzyme activity of ACC oxidase (ACO) and ACC synthase (ACS) by regulating the expression of AaACO1 and AaACO3, AaACS1 and AaACS2	Podlesakova et al. (2019), Li et al. (2021), Dong et al. (2022)
Heat treatments	Improving the integrity of the membrane due to the increase in the ratio of unsaturated fatty acids to saturated fatty acids; The expression and accumulation of heat shock proteins; Improving the performance of the antioxidant system and changing the activity of PAL and PPO enzymes; increasing sugar metabolism; Induction of cold resistance by regulating arginine biosynthesis pathways and production of signaling molecules such as polyamine, nitric oxide and proline	(Aghdam and Bodbodak 2014)

radical scavenging and antioxidant enzymes of arils and exhibited the lowest decay and weight loss (Molaei et al. 2021). It has been reported that the application of chitosan after organic acids treatment such as ascorbic, malic, and oxalic was practical for maintaining bioactive compounds, and antioxidant activity, reducing microbial spoilage and CI of pomegranate fruit during storage (Sayyari et al. 2016; Ozdemir and Gokmen 2017; Ehteshami et al. 2020). Also, emulsions and films of chitosan-oregano or cinnamon exhibited a complete inhibition against pathogens (Munhuweyi et al. 2017a). Coated fruits with chitosan and thymol had lower weight loss and higher anthocyanin, total phenol, flavonoid content, and sensory characteristics (Malekshahi and ValizadehKaji 2021). Chitosan nanoparticles containing clove essential oil also maintained fresh weight, TSS, and antioxidant activity and increased the shelf life and sensory quality of aril by reducing fungal contamination (Hasheminejad and Khodaiyan 2020). Similarly, savory essential oil encapsulated in chitosan nanoparticles was introduced to maintain the biochemical and sensory quality (Amiri et al. 2021). In a recent study, pomegranate peel extract and zinc nanoparticles loaded on chitosan coating reduced weight loss, microbial load, mold, and yeast and improved the sensory characteristics of the pomegranate fruit (Anean et al. 2023). The preservation of bioactive compounds is due to the role of coating in reducing oxidation (Saba and Amini 2017). In addition, there are reports on the role of chitosan in the transcription of genes that cause the synthesis of protective stimuli and the maintenance of phenolic content (González-Saucedo et al. 2019). The antimicrobial activity of chitosan nanoparticles has based on the electrostatic attraction between the protonated amine groups of chitosan and the negatively charged phospholipids of the cell wall

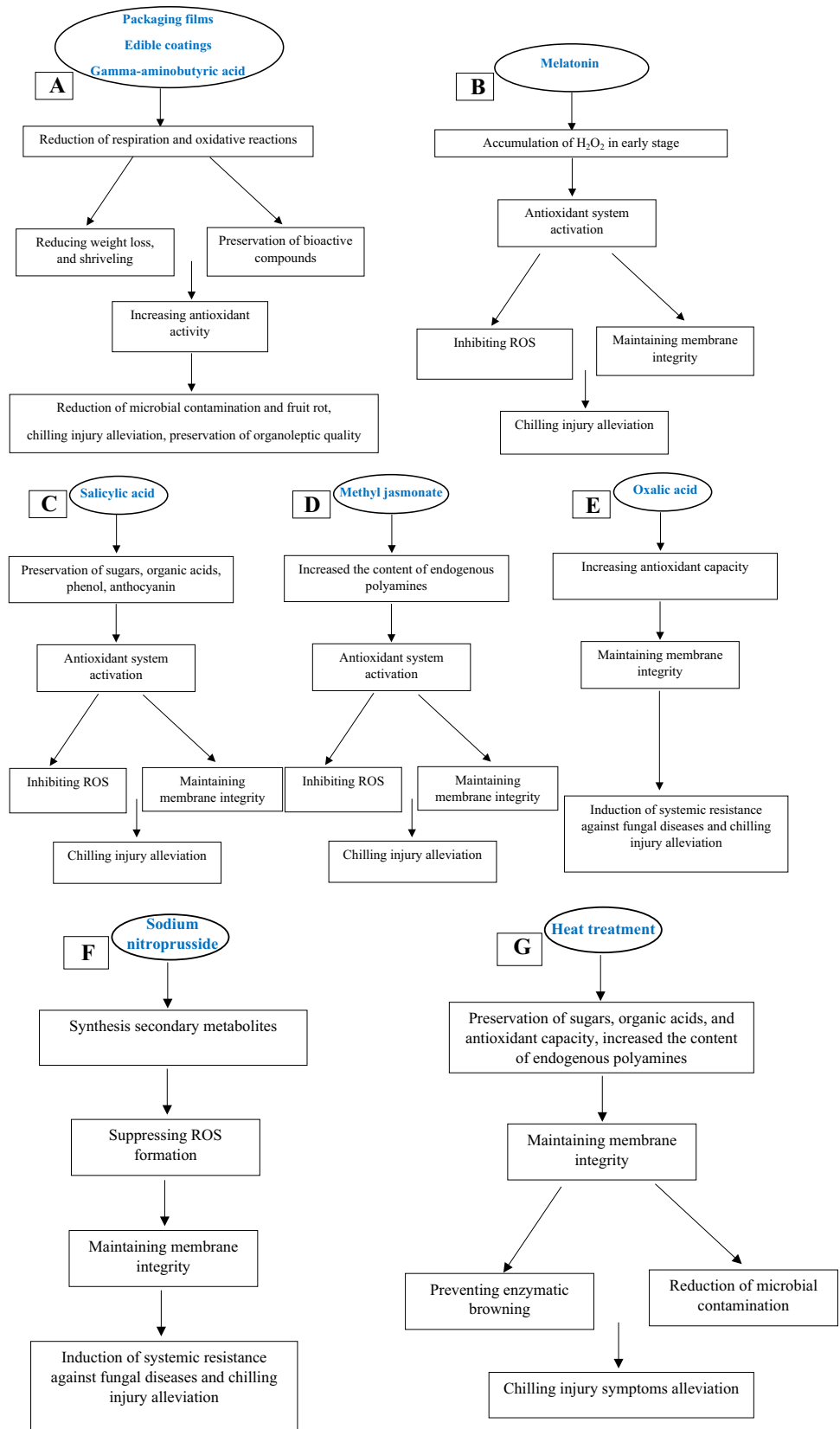
of microorganisms, which increases the permeability and degradation of the cell membrane (Li et al. 2015; Chandrasekaran et al. 2020; Yan et al. 2021). It has proved that chitosan affects protein biosynthesis and membrane fluidity and damages cell integrity by accumulating reactive oxygen species (ROS) in the microorganism cell and may be involved in energy metabolism (Ke et al. 2021).

Carboxymethyl cellulose (CMC) is one of the most common modified celluloses with good solubility and reactivity (Pettignano et al. 2019). A decreasing weight loss and vitamin C of pomegranate arils in CMC coatings enriched with zinc oxide (ZnO) (Saba and Amini 2017) was observed due to a decrease in aerobic oxidation and followed by increasing antioxidant activity. CMC and chitosan combined with organic acids reduced hydrogen peroxide, electrolyte leakage, and MDA while maintaining total phenol content, catalase activity, and antioxidant activity (Ehteshami et al. 2019). Propolis hydrophobic composites with the formation of a biodegradable barrier prevent the diffusion of water vapor on the surface of the fruit and thus prevent weight loss during storage (Kahramanoğlu et al. 2018) and combined treatment of propolis with modified atmosphere packaging is more effective. Also, propolis extract prevents the losses of total soluble solids, titratable acidity, and ascorbic acid and improves sensory acceptance (Kahramanoglu and Usanmaz 2017; Kahramanoğlu et al. 2018) (Table 1 and Fig. 1A).

Micronutrients

ZnO nanoparticles have been Generally Recognized as Safe (GRAS) products by the U.S. Food and Drug Administration (FDA) for use in food packaging (Espitia et al. 2012). Recently, ZnO nanoparticles have been used in food

Fig. 1 A proposed model for postharvest treatments-mediated chilling injury resistance



packaging due to their low cost, nutritional and antibacterial characteristics (Dai et al. 2022; Sosa et al. 2023). Improving quality characteristics and shelf life of arils enriched with zinc sulfate ($ZnSO_4$) and nano-zinc oxide (nZnO) has been observed (Aminzadeh et al. 2022), which maintains intracellular acids by reducing microbial load and preventing aril weight loss. Similarly, $ZnSO_4$ and essential oil combination have been recommended for improving the quality characteristics, especially the increase of the Zn nutrient of arils to meet the body's nutritional needs (Aminzadeh et al. 2023). Coating carboxymethyl cellulose containing nZnO of pomegranate arils increases the shelf life by preserving phenol, anthocyanin, vitamin C, and antioxidant capacity and reducing mesophilic bacteria, mold, and yeast (Saba and Amini 2017). Zn and manganese are an activator of antioxidant enzymes. Activation of antioxidant enzymes such as SOD and CAT, also the accumulation of non-enzymatic antioxidants such as ascorbic acid and phenolics, could delay senescence and extend the shelf life (Gill and Tuteja 2010). After dissolving in water, zinc ions (Zn^{+2}) bind to the membrane of the microorganism and delay the completion of cell division and growth cycle (Atmaca et al. 1998), as after penetrating the bacterial cell wall, Zn^{2+} affects its cytoplasmic content and finally leads to the programmed cell death of bacteria (Table 1).

Plant Growth Regulators (PGRs)

Melatonin (MT)

Melatonin (N-acetyl-5-methoxytryptamine) is involved in physiological processes and regulating gene expression of biosynthetic/catabolic pathways of phytohormones (Arnao and Hernández-Ruiz 2021).

Post-harvest melatonin application by the accumulation of phenolic compounds increases the antioxidant capacity of pomegranate and scavenges ROS (Jannatizadeh 2019). The content of ascorbic acid is affected after melatonin treatment, and stimulating the accumulation of glutathione leads to an increase in anthocyanin and phenolic compounds (Aghdam et al. 2020a). MT treatment reduced ion leakage by increasing unSFAs and induced CI resistance by maintaining membrane integrity, reducing MDA, electrolyte leakage, and peel browning (Jannatizadeh 2019; Molla et al. 2022). Other researchers attributed the membrane integrity to less hydrogen peroxide (H_2O_2) accumulation following the activity of ROS scavenging enzymes such as CAT, SOD, APX, and glutathione reductase (GR) (Xu et al. 2019; Aghdam et al. 2020a). The maintenance of membrane integrity likely is due to the acceleration of electron flow in the mitochondrial electron transport chain by promoting NADH dehydrogenase, cytochrome b c1 oxidoreductases, and FoF1 -ATP synthase, which increases the capacity of ATP

synthase (Tan et al. 2013). In addition, exogenous application of melatonin induces CI resistance through the oxidative pathway of pentose phosphate and regulation of phenolic compounds metabolism (Aghdam et al. 2020a). Therefore, phenol accumulation and DPPH inhibition due to the high activity of the enzyme phenylalanine ammonia-lyase (PAL) and the low activity of the enzyme PPO are necessary for CI resistance. In general, an increase in membrane integrity, antioxidant enzyme activity, and a decrease in CI have been observed after MT treatment (Aghdam et al. 2020a) (Table 1 and Fig. 1B).

Salicylic Acid (SA)

SA is involved in various physiological processes (Koo et al. 2020) and biotic and abiotic stresses (Sheteiwy et al. 2019). SA has been used in the postharvest storage of a wide range of products due to the absence of toxic residues (Asghari and Aghdam 2010).

Post-harvest application of SA, acetylsalicylic acid (ASA), and methyl salicylate (MeSA) preserved the quality and levels of total antioxidant, such as phenolics, anthocyanins, and ascorbic acid (Sayyari et al. 2011a, b; Dokhanieh et al. 2016). Furthermore, it reduces CI (Sayyari et al. 2011a, b; Boshadi et al. 2018) in pomegranate fruit. Exogenous SA led to the preservation of sugars, organic acids, phenol, anthocyanin, and antioxidant capacity by reducing respiration rate and PAL enzyme activity, and it was efficient in CI by reducing electrolyte leakage (Sayyari et al. 2009, 2011a). The combination of SA and putrescine increased bioactive compounds and fruit quality (Koyuncu et al. 2019). SA may stimulate anthocyanin synthesis through phenylpropanoid pathway activation (Sayyari et al. 2016), and in this regard, Koyuncu et al. (2019) observed the best fruit color in pomegranates treated with SA. SA increases ascorbic acid content by inducing APX activity and inhibiting ascorbic acid oxidase (AAO) activity (Rao et al. 2011). Post-harvest application of SA also maintained the chroma index of aril and peel, titratable acidity, and TSS (Koyuncu et al. 2019; Güneş et al. 2020). Arils treated with SA had good visual quality, no decay, and an unpleasant aroma (Shaarawi et al. 2016). Salicyloyl chitosan-treated pomegranate fruits had higher unsaturated/saturated fatty acid (unSFA/SFA) ratio (Sayyari et al. 2016), which delayed electrolyte leakage and internal and external browning. Also, the antioxidant capacity hydrophilic (H-TAA) and lipophilic (L-TAA) due to increasing phenol, anthocyanin, and ascorbic acid were high in the SA-treated fruits. The fruits showed lower weight loss, respiration rate and ethylene production, followed by higher firmness, total soluble solids, and more titratable acidity as a sensory quality (Sayyari et al. 2016). SA and methyl jasmonate in fresh-cut pomegranate significantly preserved antioxidant capacity under cold storage (El-Beltagi et al.

2023), followed by the reduction of oxidative stress suppressed the respiration rate (Aghdam et al. 2016a) (Table 1 and Fig. 1C).

Methyl jasmonate (MeJA)

MeJA is involved in physiological processes (Wang et al. 2019a; Pan et al. 2020) and improves the post-harvest quality of pomegranate fruit (Wang et al. 2021; Serna-Escolano et al. 2021).

Post-harvest application of MeJA mainly improves fruit quality by increasing phenolics and flavonoids, antioxidant capacity and volatile compounds production, and delayed senescence (Wang et al. 2021). MeJA reduces CI by delaying the ripening and preserving of antioxidant compounds (Sayyari et al. 2011a, 2017). In other studies, MeJA prevented CI by suppressing the activity of polyphenol oxidase and preventing the reduction of total phenol, reducing MDA, and maintaining the fluidity of the cell membrane (Chen et al. 2021). MeJA vapor treatment increased the content of endogenous polyamines, especially putrescine, and spermidine, which are efficient in resistance to CI (Valero et al. 2015). MeJA prevents membrane lipids degradation and permeability change and thus slows down the efflux of K^+ , Ca^{2+} , sugar, and other electrolytes and maintains intracellular stability. Examination of the structure of the pericarp in microscopic studies showed no damage to the lipophilic layer and cuticle, and the epidermal cells had a regular structure (Chen et al. 2021). In general, MeJA is a beneficial tool to prevent CI (Table 1 and Fig. 1D).

Oxalic Acid (OA)

OA has physiological functions, including induction of systemic resistance against fungal diseases by increasing the activity of antioxidants. Previous studies showed that its long-term use in non-climacteric fruits extended the shelf life (Valero et al. 2011; Ravi et al. 2017). In addition, endogenous OA causes intrinsic heat tolerance and increases antioxidant capacity (Osei-Kwarteng et al. 2023). The main effects of organic acids as anti-senescence agents are to delay ripening (Gimenez et al. 2017).

Sayyari et al. (2010) showed that OA preserves the total phenolics of pomegranate. Similarly, OA can increase the storage life of pomegranate by preserving bioactive compounds and antioxidant activity (Koyuncu et al. 2019). The mechanism of preservation of bioactive compounds by OA probably is due to its antioxidant properties that prevent lipid peroxidation. The combination of polysaccharide-based edible coatings and OA increased the phenolic content, CAT activity, and antioxidant activity while reducing H_2O_2 , MDA, electrolyte leakage, and CI in pomegranate fruit (Ehteshami et al. 2019, 2020). It seems OA reduces

CI symptoms by maintaining membrane fluidity (Ehteshami et al. 2019) and inducing antioxidant activity at low temperatures (Huang et al. 2016). The combination of controlled atmosphere and OA significantly reduced postharvest decay (Koyuncu et al. 2019). (Table 1 and Fig. 1E).

Sodium Nitroprusside (SNP)

SNP has been used as a nitric oxide (NO) donor for pre- and post-harvest treatment of apple fruit and might modulate shikimate and phenylpropanoid pathways (Ge et al. 2019). The shikimate pathway is a metabolic pathway that generates precursors to synthesize many secondary metabolites to enhance disease resistance (Karki and Ham 2014). SNP increases disease resistance by suppressing ROS formation and improves the fruit quality of *Cucumis melo* by suppressing ethylene production (Wang et al. 2019b; Sahu et al. 2020). Treatment with NO donors induces cold stress resistance in climacteric and non-climacteric fruits (Xu et al. 2012). The effects of NO are greater in non-climacteric fruits (Osei-Kwarteng et al. 2023). Reduces CI by exogenous application of NO is related to decrease membrane permeability, MDA content, ion leakage, and lipid peroxidation (Ranjbari et al. 2016, 2018; Wu et al. 2014). It seems to be associated with a decrease in the production or detoxification of ROS (González-Gordo et al. 2019) and an increase in the expression or activity of antioxidant enzymes (Wu et al. 2014; Babalar et al. 2018). SNP treatment increases the total quantity of adenosine triphosphate (ATP), the activity of enzymes involved in energy metabolisms such as H^+ -ATPase, Ca^{2+} -ATPase, succinic dehydrogenase (SDH), and cytochrome C oxidase (COX), which lead to an increase in cold stress resistance (Wang et al. 2015a). The application of exogenous NO in non-climacteric fruits reduces ethylene production and leads to a decreased respiration rate (Zhu and Zhou 2007). NO is an inhibitor of the mitochondrial respiratory chain, which binds to iron-sulfur proteins and inhibits their biological activities (Lin et al. 2012) (Table 1 and Fig. 1F).

Gamma-Aminobutyric Acid (GABA)

In the last decade, there has been an increasing trend to use natural stimulants to induce CI resistance and delay senescence in subtropical and tropical fruits (Shelp et al. 2017; Zhu et al. 2022). GABA is a four-carbon non-protein amino acid and the natural signal produced in plants under biotic and abiotic stress (Li et al. 2021; Liu et al. 2022). Activation of the enzyme and non-antioxidant defense system, maintenance of carbon–nitrogen ratio balance, involvement in the metabolism of carbohydrates and amino acids, regulation of plant growth, chlorophyll biosynthesis and membrane stabilization, osmotic regulation induced by exogenous GABA

in plants (Ji et al. 2018; Shomali et al. 2021). GABA is a safe edible coating in the food industry (Naila et al. 2010) that positively affected peel and aril firmness, flavor, texture and color, antioxidant activity, the content of phenolic compounds, and reduction of ion leakage and CI pomegranate fruit (Nazoori et al. 2020). Accordingly, edible coating based on carnauba wax with the addition of GABA delayed CI symptoms and preserved quality (Nazoori et al. 2022). In general, applying GABA post-harvest can increase the storage life of pomegranates by maintaining the quality characteristics.

Coatings act as a semi-permeable barrier against oxygen, carbon dioxide, and moisture, thus reducing water loss, respiration rate, and oxidative reactions (Maqbool et al. 2011). Edible coatings improve the firmness by maintaining the content of polysaccharides and subepidermal cells structure and integrity of the membrane and inhibiting the activity of pectin methyl esterase and polygalacturonase enzymes (Wang et al. 2015b). Exogenous application of GABA is a strategy to increase endogenous GABA levels and GABA shunt activity, which leads to an increase in carbon flux through the respiratory pathways, which will lead to an increase in NADH, NADPH, and ATP (Aghdam et al. 2018, 2020b; Shelp et al. 2021) and improves post-harvest quality (Aghdam et al. 2022). Storage at low-temperature delays senescence and improves resistance to CI by promoting the activity of the GABA pathway (Aghdam et al. 2022). GABA reduces the activity of phospholipase and lipoxygenase enzymes and increases antioxidant activity it maintains membrane fluidity by maintaining the ratio of SAFs to unSAFs (Aghdam et al. 2016b). GABA's role in increasing antioxidant activity is due to its ability to increase antioxidant compounds such as phenols and flavonoids (Wang et al. 2014). Phenols, especially anthocyanins, are related to antioxidant activity in pomegranates. GABA increases phenolic compounds, including anthocyanins, its effects are attributed to the activity of PAL, a key enzyme in the biosynthesis of phenols, and decreasing the activity of PPO (Ge et al. 2018; Habibi et al. 2020). Other mechanisms to reduce CI by exogenous GABA may be energy conservation by providing NAD, ATP, and inhibition of cytoplasmic acidification (Aghdam et al. 2016b, 2018).

The balance of SOD activity and H₂O₂ scavenging enzymes can be critical for cell survival during cold storage. GABA scavenges ROS and protects plant tissue against active carbonyl damage through higher SOD activity (Malekzadeh et al. 2017; Habibi et al. 2019) (Table 1 and Fig. 1A).

Heat Treatments

Heat treatments, such as vapor, water immersion, hot water rinsing, and bruising are simple and eco-friendly

technologies to maintain functional and nutritional properties and extend the fruit shelf life.

Ascorbic acid, anthocyanins, and phenolics are responsible for total antioxidant activity in pomegranate fruits, and heat treatment helps preserve health-promoting compounds by increasing total antioxidant activity (Kulkarni et al. 2005). Heat-treated pomegranate fruit showed higher total antioxidant activity than the control (Mirdehghan et al. 2006). The higher antioxidant is related to high levels of total phenol, ascorbic acid, and anthocyanin content. In addition, the preservation of sugars (glucose and fructose) and organic acids (malic, citric, and oxalic acids) maintained the organoleptic quality of arils (Mirdehghan et al. 2006). The effect of heat on the increase of sugar concentration can be attributed to the increase of activities of glucosidase, galactosidase, and arabinase, which causes the release of sugar from cell wall polymers (Beirao-da-Costa et al. 2006). Preservation of organic acids may be due to respiration rate inhibition under heat treatment (Serrano et al. 2004). Heat treatment increases free putrescine and spermidine and delays fruit softening (Mirdehghan et al. 2007). Higher polyamine levels and maintaining the ratio of SAFs to unSAFs maintain the integrity and fluidity of the membrane and induce a mechanism of tolerance to low temperatures (Mirdehghan et al. 2007). Mild heat treatment, such as hot water, reduces microorganisms and inactivates destructive enzymes (Talaie et al. 2004; Mirdehghan et al. 2006). Heat shock likely induces ROS, followed by the activation of oxygen radical scavengers such as SOD, POD, and CAT, as a defense system against oxidative stress (Moller 2001). Hot water treatment of arils suppressed the PAL and PPO activity while increasing POD activity and subsequently preventing enzymatic browning and quality reduction (Maghoumi et al. 2013). Investigating the effect of intermittent warming and hot water treatment showed that intermittent heating treatment increased shelf life and reduced pomegranate fruit decay (Moradinezhad and Khayyat 2014); however, storage life was higher in hot water treated-fruit, which is probably due to improved defense-system is against post-harvest pathogens. Intermittent warming, especially a warm period at the beginning of storage, resulted in higher enzymatic antioxidant activity and phenolic content and lower PPO activity in the peel, resulting in less cold damage (Taghipour et al. 2021a). Preservation of unSFAs against peroxidation, lower MDA production, continuous increase of spermidine, and higher levels of putrescine were observed as the membrane immune system immediately after treatment (Taghipour et al. 2021b). Maintaining antioxidant enzyme levels is probably responsible for reducing lipid peroxidation. Warming before the appearance of irreversible CI symptoms is a practical method for cold storage (Table 1 and Fig. 1G).

Conclusion and Prospects

The quantitative and qualitative characteristics of the fruit are affected by pre- and post-harvest management. Therefore, appropriate processes at pre- and post-harvest stages lead to reducing stresses and increasing quality. Also, the food produced in the correct management system will be suitable from the nutritional value, sensory quality, and safety aspects. Packaging protects the product against mechanical injury and microbial spoilage and improves organoleptic quality and marketability. Considering that pomegranate fruit is non-climacteric, the use of polymer films may have a good potential for the maintenance of its quality. Polymer films create a modified atmosphere and have a significant effect on preventing chilling injury and maintaining fruit quality. However, there is a need for extensive studies to develop the packaging system for pomegranate arils and fruit in different commercial cultivars. On the other hand, the increase in polymer production for post-harvest application indicates low recycling rates and environmental issues, therefore the assurance of the existing technology for edible coatings production must be considered to meet consumer demand. Although edible coatings limit moisture loss and gas exchanges and increase shelf life, however, further research is essential to ensure the moisture barrier properties of hydrophilic edible coatings, improving the adhesion and durability of the coating during storage. In addition to extending shelf life, improving nutritional quality is one of the important research priorities. The biofortification of plants with nanoparticles help to improve nutritional quality. However, further research is needed to optimize the concentration to reduce postharvest physiological disorders. MT and SNP, as signaling molecule, affects metabolism by regulating endogenous levels of hormones and oxygen free radicals. Besides, approval of the FDA is essential regarding the possibility of commercialization despite the high cost and long-term treatment time. SA, JA, OA, and GABA are safe compounds to maintain quality and antioxidant activity during storage. However, detailed knowledge of the mechanisms that increase bioactive compounds and antioxidant capacity is essential. Post-harvest heat treatment is efficient in preserving total antioxidant activity and bioactive compounds. However, the optimal temperature and duration of heat treatment to prevent irreversible oxidative stress should be determined in commercial pomegranate cultivars.

All the processes used in the previous research significantly increase the nutritional quality. However, further research in various commercial cultivars is vital to select the best treatments at the optimal concentration for extending the storage life of pomegranate fruit and arils.

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Declarations

Conflict of interest The authors declare that they have no conflict of interest.

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Consent to Participate On behalf of all co-authors I believe the participants are giving informed consent to participate in this study.

Consent for Publication I, Farid Moradinezhad give my consent for submitted manuscript to be published in the JPGR.

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