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Physical Treatments for Alleviating Chilling Injury in Fresh produce

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ABSTRACT

Fresh produce has a short shelf-life because the metabolic activity continues after harvest. Low temperature is one of the postharvest technology methods that suppress this activity along storage. Its effects prolong the shelf-life of fruits and vegetables. This method has long been recommended to reduce deterioration during storage so that it can maintain the quality of fruits and vegetables. However, it still has drawbacks such as chilling injury, especially in tropical and subtropical origins that are chilling sensitive. Therefore, another storage method is needed to alleviate chilling injury such as low-temperature conditioning, high-temperature conditioning, and intermittent warming which only use environmental conditioning during storage. The other one has modified atmosphere packaging and controlled atmosphere packaging. They utilize the ideal atmosphere for each fresh product during storage. The treatment proved that it could alleviate chilling injuries such as reduced pitting, flesh injury, failure of mature, scald, peel browning, weight losses, electrolyte leakage, malondialdehyde, respiration rates, production of superoxide radical anion (O_2-) & hydrogen peroxide (H_2O_2), lipoxygenase activity, phospholipase D, phenylalanine ammonia-lyase (PAL), and polyphenol oxidase (PPO).

1. INTRODUCTION

Fresh produce continues its biological activity after harvest. It causes the high metabolic activity while postharvest handling. The fruits and vegetables which are categorized as fresh produce have high moisture content than legumes and grains which can be a trigger for metabolic activity (Seymour *et al*., 1993). As scientist know, high metabolic causes rapid ripening and senescence leading to deterioration in chemical composition, texture, flavor, aroma, appearance, and color. This process is strongly influenced by temperature during storage.

Low temperature is one of the most effective methods since antiquity to reduce metabolic activity during storage. It is famously used in preserving quality fresh produce in the market or transportation (Concellón *et al*., 2007). Preserving the quality is important for fresh produce after harvest because the quality is closely related to the perception of consumers, which is influence the price of theirs. However, low-temperature storage is a problem for tropical and subtropical product that are sensitive to chilling injury.

Chilling injury is a physiological disorder that occurs in several commodities, especially those originating from tropical and subtropical regions. Examples of chilling-sensitive commodities are mango, cucumber, banana, papaya, persimmon, orange, pineapple, pear, apple, tomato, avocado, broccoli, zucchini, and eggplant. Physiological damage occurs in these chilling-sensitive commodities when they are stored at a low temperatures below 12°C (Lyons, 1973).

The phenomenon of chilling injury appears slowly day by day and the symptoms arise after the product is exposed to room temperature. The chilling injury symptoms in several commodities are pitting, water-soaked spots, uneven ripening, discoloration, failure to ripen, off flavors, browning in the peel (Paull & Jung Chen, 2000; Gross, 2016). The degree of severity in the product is depends on the temperature and time of exposure to low temperature, where it is reversible or irreversible. The reversible occurs when the product is returned to a non-chilling temperature so that the tissues reverse the damage and the irreversible occurs when the product has been exposed to low temperature for a sufficiently long time causing the membrane tissues to die (Lukatkin *et al*., 2012). This led to a deterioration in the quality of the product and reduced consumer acceptance.

Therefore, postharvest treatment to reduce chilling injury is required. Many researchers have carried out the experiment to alleviate chilling injury in low-temperature storage both physically and chemically post-harvest treatment. The example of physical treatment is intermittent warming, low-temperature conditioning, high-temperature conditioning, controlled atmosphere treatment, modified atmosphere, and chemical treatment is 1-Methylcyclopropene (1-MCP), Brassinosteroids, abscisic acid (ABA), nitric oxide, methyl jasmonate (Valenzuela *et al*., 2017). Over the years, new techniques have been discovered with advantages and disadvantages; chemical treatment needs special chemicals, and physical treatment needs special equipment. The natural treatments have been applied in different researches such as low-temperature conditioning, high-temperature conditioning, intermittent warming, modified atmosphere, and control atmosphere which did not require special equipment and some chemicals. Temperature condition gives an excellent effect if the settings temperature and kind of commodity are optimal. In this review, we focus to discuss physical treatment to reduce chilling injury during storage. From the existing research, this paper wants to dig into what kind of approaches to measure the symptoms of chilling injury and how far the physical treatment can reduce the chilling injury during storage.

2. APPROACHES TO MEASUREMENT OF CHILLING INJURY SYMTOMS

Chilling injury symptoms often occur after exposure to non-chilling temperatures such as 20 °C during marketing or wholesale. For many years, experiments about chilling injury topic in fruits and vegetables have been done, but the severity is difficult to define as quantitatively. Usually, the severity of the chilling injury is measured qualitatively by firmness and water losses, and visual observation such as chilling injury index and color change (Lyons, 1973). However, membrane damage due to the metabolic process which is triggered by environmental stress has a higher correlation with the degree of damage than the quality parameter. The scheme of membrane damage is shown in Figure 1.

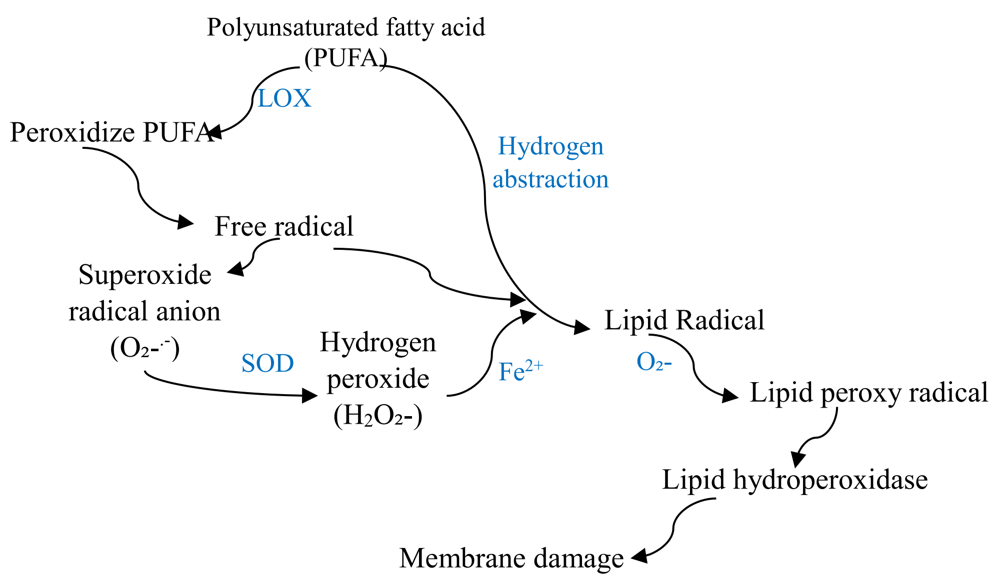

Figure 1. The Scheme of membrane deterioration in postharvest stress (Marangoni *et al*., 1996; Repetto *et al*., 2012).

Most of the previous research on environmental stress such as chilling injury has been based on the metabolic process, which correlates closely with the membrane deterioration (Marangoni *et al*., 1996; Parkin *et al*., 1989). The assessment of chilling injury that correlates with the metabolic processhas been measured by electrolyte leakage, lipoxygenase (Mao *et al*., 2007; Parkin & Kuo, 1989; Saltveit, 2005), fatty acid composition (Jin *et al*., 2015), malondialdehyde content (Ghani *et al*., 2017), enzyme activity of superoxide dismutase, catalase, ascorbate peroxidase (Valenzuela *et al*., 2017), reactive oxygen; superoxide radical anion (O_2-) & hydrogen peroxide (H_2O_2) (Shewfelt & Del Rosario, 2000), gene expression (Chen *et al*., 2017) and metabolomic analysis (Lurie, 2022).

2.1. Electrolyte Leakage (EL) and Malondialdehyde (MDA)

Lipid degradation in the membrane cell occurs by an enzymatic processor non-enzymatic process. In the non-enzymatically process, electrolyte leakage indicates that membrane damage has occurred due to an increase in permeability. The severity of these phenomena will continue when the low temperature exposed fruit or vegetable in a long time (Marangoni, Palma, and Stanley, 1996). EL increase at a chilling temperature (2.5°C) which indicate as chilling injury for some postharvest produce. In some experiment, the researchers have used the rate of electrolyte leakage to indicate the chilling injury symptoms. The permeability of the cell wall slowly increases day by day of storage and causes the cell membrane to break down. These phenomena are utilized to calculate the rate of EL during storage. Fahmy & Nakano, (2013) measured the rate of EL from total conductivity per hour to indicate the chilling injury symptoms in stored cucumbers. The researchers observed an upward trend in the electrolyte leakage of cucumber fruits when stored at 5°C with low and medium relative humidity. This is associated with membrane damage due to environmental stress. In addition, The MDA is an end product of lipid peroxidation. The amount of malondialdehyde can be an expression of the level of lipid peroxidation in the non-enzymatic process related with CI. Some researchers have attempted to use MDA levels to monitor CI status in stored cucumbers. Fahmy *et al*. (2015) mentioned that the MDA content increased at chilling temperatures. The increase in MDA was also associated with the increase in EL. Lado *et al*. (2016) also found that MDA concentration increased after two weeks of storage in grapefruits. Thus, we concluded that MDA is a mirror of CI symptoms for sensitive fruits. It is associated with chilling injury damage, which is exposure to long-time cold storage.

2.2. Reactive Oxygen (O_2- & H_2O_2) and Antioxidant Activity

Lipid peroxidation in chilling stress is the primary molecular mechanism in oxidative damage of the cell membrane. The process of lipid peroxidation has a role chain reaction-initiated hydrogen abstraction or radical oxygen which results in oxidative damage from polyunsaturated fatty acid (Repetto *et al*., 2012). Radical oxygen is known as reactive oxygen species (ROS) which is the most concern in a biological system. ROS mechanism is reduced by antioxidant activity such as superoxide dismutase (SOD), ascorbate peroxidase (APX), and catalase (CAT). SOD activity can convert superoxide radical anion (O_2-) to hydrogen peroxide (H_2O_2) (Mansoor *et al*., 2022). CAT and APX activity converts H_2O_2 to water (H_2O). APX is responsible for the modulation of ROS for signaling and CAT is responsible for the removal of excess ROS during stress (Mittler, 2002). Therefore, H_2O_2, SOD, CAT, and APX can be used as a parameters to measure the occurrence of chilling injury.

According to Yang *et al*. (2012), H_2O_2, SOD, CAT, and APX were used to determine the occurrence of chilling injury in cucumber stored at 2°C. Superoxide radical (O_2-) production and H_2O_2 content are also used as a parameters to determine the occurrence of chilling injury in stored mango at 5 °C because O_2- and H_2O_2 involved to lipid peroxidation in membrane damage (Zhang *et al*., 2017). High SOD, CAT, and APX activity could reduce the occurrence lipid peroxidation (Lado *et al*., 2016) and increased the H_2O_2 content indicates damage in the membrane cell (Zhang *et al*., 2017).

2.3. Fatty Acid Composition and LOX Activities

Post-harvest stress such as chilling injury affects membrane deterioration in the tissue, the process includes the changing temperature of bulk membrane lipids and decrease bulk membrane lipid fluidity. The process membrane fluidity and permeability are hydrolysis membrane phospholipids to free fatty acids by lipoxygenase (LOX) and will produce peroxidase fatty acid with the corresponding production of free radicals (Liu *et al*., 2020; Marangoni *et al*., 1996). Example mechanism membrane deterioration has been summarized in Figure 1. LOX is one of the two types of enzymes that are responsible for senescence and chilling injury in membrane damage, so an increase in LOX can indicate the occurrence chilling injury (Cao *et al*., 2009; Whitaker, 2012). Low LOX activity can maintain low superoxide radical activity which can contribute to the elimination of H_2O_2 (Cao *et al*., 2010). Mao *et al*., (2007) reported that LOX increased when cucumbers were stored at 2°C and decreased when cucumbers were heat treated prior to storage.

On the other hand, the change in fatty acid composition is also an indication of chilling injury when stress occurs (Rui *et al*., 2010a). Five fatty acids in membrane lipids are palmitic acid, stearic acid, oleic acid, linoleic acid, and linolenic acid, which are used as an indicator of chilling stress tolerance (Cao *et al*., 2009, 2011). The high percentage in fatty acid composition (unsaturated/saturated fatty acid) is responsible for acclimation to low-temperature condition (Mirdehghan *et al*., 2007). In cold storage kiwifruit, putrescine treatment reduced chilling injury symptoms due to higher unsaturated fatty acid (Yang *et al*., 2016).

2.4. Gene Expression

In postharvest physiology, gene expression is the most important because chilling stress, senescence, or metabolic processes are related to the expressed gene. It helps to understand the mechanisms that occur in plants or postharvest products during growth or storage, especially for membrane cell conditions. For example, (Lado *et al*., 2016) utilized gene expression as the tool to reveal the antioxidant system in chilling injury tolerance. Chilling injury in apple fruit such as soft scald, needs to be reduced by understanding the metabolism. One of the important things to understand is the correlation between specific metabolites and specific gene expression (Leisso *et al*., 2016). Maul *et al*. (2011) found LTP, MET, CAT, GTPP, LEA5, and SZP genes as a signal to express the chilling injury condition in stored grapefruit. Transcript levels were consistently higher in low-temperature treatment than in non-condition fruit. It was confirmed that genes expressions are affected by chilling and condition temperature. A heat treatment can alleviate CI with approaching gene expression. Hot water treatment (HWT) activates the expression of several genes related to stress CI, and conditioning treatment activates the expression of lipid membrane modification enzymes (Sapitnitskaya *et al*., 2006).

3. PHYSICALLY APPROACHES FOR CHILLING INJURY TREATMENT

3.1. Intermittent Warming

Intermittent warming is one of the natural treatments to reduce chilling injury for the post-harvest product by interrupting temperature. Warming before the chilling injury occurs is irreversible. Warm temperature is applied for one or more short periods when the fruit or vegetable stored at low temperature would increase the shelf-life of some chilling sensitive commodities (Wang *et al*., 2018).

Bell peppers are sensitive to chilling when stored at temperatures below 7°C. Apropriate intermittent warming (IW) such as warming at 20°C 24 h after 6 and 13 days of storage at 4°C, could preserve the integrity of the membrane because IW helps to delay the decline of unsaturated fatty acid content (Liu *et al*., 2015). Many researchers have tried intermittent warming (Table 1) because different kind of commodity require different temperature and time that need. Parkin & Kuo (1989) reported that during intermittent warming in storage of cucumber, the unsaturated of the glycolipid fraction was reduced. The intermittent warming was carried out with the warming at 14°C after storage for 3, 7, 10, 14, and 18 days at 4°C.

3.2. Temperature Conditioning

Fresh produce has a high temperature after being harvested in the field, a condition that causes high respiration which is associated with deterioration. The low-temperature conditioning provides the advantage for reducing the temperature. In practice, temperature conditioning such as low-temperature conditioning and high-temperature conditioning is applied to fresh produce before storing it at low temperatures. The aim of this treatment is to prevent membrane cells to avoid temperature shock and chilling injury during storage.

Low-temperature conditioning (LTC) is treatment above a critical chilling range. It is an alternative for increasing chilling injury tolerance. LTC at 5°C for six days before storage at 0°C for 33 days successfully alleviated chilling injury in 'Luoyangqi'loquat. LTC is useful to maintain quality of loquat both external and internal quality and could prolong

Table 1. Intermittent warming treatments to alleviating Chilling Injury

Commodity	Storage Temp. (°C)	Storage Frequency	Warm Temp. (°C)	Warm Period	Effects	Reference
Cucumber	2.5	Every three days	12.5	18 h	- Reduced pitting and decay - Reduced electrolyte leakage - Reduced ethylene production - 1-aminocyclopropane-1-carboxylic acid (ACC) level increase slightly	(Cabrera & Saltveit, 1990)
Green Tomato	8	Every six days	20	24 h	- Improve development color	(Biswas *et al.*, 2012)
Lime 'Tahiti'	5	Every 7 & 14 days	20	48 h	- Reduced CI symptoms based on visual assessments	(Kluge *et al.*, 2003)
Mandarin 'Balady'	2	Every 2 weeks	8	8 h	- Reduced weight loss & % decay - Increase TSS until last storage	(Mansour *et al.*, 2014)
Orange 'Olida'	3	Every three weeks	15	Two weeks	- Reduced CI symptoms based on visual assessments - Reduced decay incidence	(Schirra & Cohen, 1999)
Peach	2	Two cycles (14d, 28d)	20	One day	- Reduced chilling injury index & MDA content - Maintaining phenolic compounds	(Liu *et al.*, 2018)
Peach 'Paraguyo'	2	Every six days	20	One day	- Reduced scald and vitrescence	(Fernández-Trujillo & Artés, 1998)
Pears 'Nanguo'	0±0.5	Every 20 days	20±1	One day	- Slow down the occurrence of peel browning - Reduced electrolyte leakage and MDA - Increased in ATP concentration, energy charge (EC), gene expression of transcripts for ATP synthase (ATPase), NADH dehydrogenase (NDA) and vacuolar proton-inorganic pyrophosphatase (VPP)	(Wang *et al.*, 2018)
Potatoes	0	Every three days	15	One week	- Decreased respiration rates - Decreased sugar content	Hruschka *et al.*, 1969
Tomato 'Dario F-150'	9	Every six days	20	24 h	- Good firmness - Delay shriveling - Lowest total losses	(Artes & Escriche, 1994)
Tomato 'New Zealand'	6	Every seven days	20	24 h	- Reduced Chilling injury and decay development based on enhanced red color development until 27 days storage	(Biswas *et al.*, 2012)

shelf-life (Cai *et al.*, 2006). On the other hand, LTC could maintain taste and a specific bioactive compound of grapefruit for 16 weeks at 2°C (Chaudhary *et al.*, 2014). Table 2 list LTC treatments to alleviate CI for some commodities.

High-temperature conditioning has several methods, such as hot air temperature and hot water temperature. Initially, this treatment induced fungal pathogens, but now the treatments used to reduce disorder in chilling injury (Lurie, 2005). (Lurie *et al.*, 1991) applied high-temperature to reduce electrolyte leakage (EL) and malondialdehyde (MDA) in tomato storage at 2 °C. Temperature and time for high-temperature treatment is the primary key in the process because the treatment has heat damage for fresh produce such as peel browning (Klein & Lurie, 1992; Schirra & D'hallewin, 1997), pitting and scald (Donkin & Wolstenholme, 1995), and yellowing on zucchini (Jacobi *et al.*, 2001). Hot water treatment (HWT) is only used for the commodity which is resistant to water because this process sneeds hot water around 55 – 38 °C for 3 – 12 min. Mccollum *et al.*, (1995) reported that HWT at 38 or 42°C reduced the level of EL in cucumber storage at 2.5 °C. Table 3 list HWT treatments to alleviate CI for some commodities.

Table 2. Low-temperature conditioning treatments to alleviate Chilling Injury

Commodity	Storage Temp. (°C)	Low Temp. Period	LTC (°C)	Conditioning Period	Effects	Reference
Avocado	0	Three weeks	6-8	3-5 d	- Reduced the hard skin and tissue breakdown	**Woolf *et al*., 2003**
Eggplant	4	9 d	13	2 d	- Reduced MDA - Maintained the appearance - Increased peroxidase & catalase activity - Reduced loss of anth°Cyanin and total phenolic compound - Reduced polyphenol oxidase (PPO)	(Shi *et al*., 2018)
Grapefruit 'Marsh'	5	12 d	16 & 20	7 & 2 d	- Increased chilling tolerance based on the transcript of some genes	(Maul *et al*., 2011)
Mango	5	25 d	12	24 h	- Reduced electrolyte leakage, MDA - Inhibit the production of H_2O_2 & O^{-}_2	(Zhang *et al*., 2017)
Lime 'Mexican'	4	20 d	13	48 h	- Maintenance of antioxidant enzymes - Reduced of MDA content - The increase of SOD activity	(Fernando Rivera-Cabrera *et al*., 2007)
Loquat fruits	1	Five weeks	10	6 d	- Reduced electrolyte leakage - Maintained a high level of Glycine Betaine (GB) content & Betaine Aldehyde Hydrogenase (BADH) activity	(Jin *et al*., 2015)
Pears 'Huangguan'	0	60 d	10	3 d	- Reduced MDA content, Lipoxygenase (LOX) activity, PPO activity - Maintained a high level of total phenolic content	(Li *et al*., 2017)

Table 3. High-temperature conditioning treatments to alleviating Chilling Injury

Commodity	Storage Temp. (°C)	Low Temp. Period	Warm Temp. (°C)	Warm Period	Media	Effects	Reference
Avocado	0	21 d	38	6 & 12 h	Air	- Reduced ethylene production - Reduced the symptom of flesh injury	(Florissen *et al*., 1996)
Avocado 'Hass'	0.5	28 d	38	60 min	Water	- Reduced external CI	(Woolf A., 1997)
Avocado	1.5	28 d	46	5 min	Water	- Reduced black cold damage	(Kremer-Köhne, 1999)
Kiwifruit 'Hongyang'	0	90 d	45	10 min	Water	- Reduced CI index - Lower MDA, lipoxygenase (LOX) activity, and ethylene production rate	(Ma *et al*., 2014)
Loquat Fruit	1	35 d	38	5 h	Air	- Reduced electrolyte leakage & MDA - Decreased phospholipase D and lipoxygenase activities	(Rui *et al*., 2010b)
Mandarin fruit	2	60 d	37	3 d	Air	- Reduced CI Index	(Holland *et al*., 2012)
Pepper fruit	1	28 d	53	4 min	Water	- Reduced CI & decay	(González-Aguilar *et al*., 2000)
Persimmon 'Fuyu'	0	6 weeks	47	0.5 h	Air	- Increased viscosity of the cell wall	(Woolf *et al*., 1997)
Persimmon 'Rojo Brillante'	1	32 d	50	3 min	Water	- Reduced firmness	(Besada *et al*., 2008)
Pomegranate	1.5	3 months	55	60 & 12 min	Air	- Reduce weight loss & electrolyte leakage	(Rahemi & Mirdehgan, 2004)
Tomato	2	3 weeks	38	48 h	Air	- Reduce CI index	(Lurie *et al*., 1997)
Valencia	4	Six months	53	3 min & 6 min	Water	- Reduced weight loss - Reduced decay of chilling injury	(Erkan *et al*., 2005)

3.3. Control and Modified Atmosphere

Fruit and vegetables after postharvest continue the biological activity caused the high respiration, especially climacteric product. The respiration triggers the quality degradation such as texture, color, phytochemical, vitamin and mineral. Environmental storage such as temperature, carbon dioxide and oxygen concentration related to the respiration rate. Control and modified atmosphere are one of the methods to reduce the respiration rate during storage. Control atmosphere (CA) could control the respiration that is suitable with the fresh produce characteristic while storage. It provides the ideal composition oxygen and carbon dioxide to suppress the respiration rate of fresh produce (Bodbodak & Moshfeghifar, 2016).

The effect of oxygen and carbon dioxide (CA) against the commodity has a variety depending on the kind or species of commodity, level of maturity, gas concentration, and temperature storage. Tolerance of a commodity against minimum O_2 and maximum CO_2 percentage we can look at Table 4. CA is not only useful in reducing chilling injury symptoms (Table 4) but also can maintain quality such as firmness, antioxidant, soluble solid, weight loss (Thompson, 2014). Lady pink apples stored at 1% O and 1% CO_2 showed a significant decreased in lipoxygenase during storage at 1°C (Villatoro *et al*., 2008). Valencia oranges stored at 0.02 % O_2 5°C can reduce respiration during storage (Ke & Kader, 1990) and CA preserve vitamin C during fruit storage (Lee & Kader, 2000).

Table 4. Minimum oxygen (O_2) and maximum carbon dioxide (CO_2) concentration tolerated in some commodities (Izumi *et al*., 1996; Kader *et al*., 1989).

Minimum O_2 concentration (%)	Maximum CO_2 concentration (%)	Commodities
1	10	Broccoli
1	15	Mushroom
2	2	Apricot, celery, lettuce, olive
2	5	Most cultivars of apples, kiwi fruit, nectarine, peach, plum, cantaloupe, cabbage, cauliflower, bananas
2	8	Papaya
2	10	Pineapple, Avocados
2	15	Cherry, strawberry
3	2	Tomato, artichoke
3	5	Onion
3	10	Persimmon, cucumber, sweetsop, mango
3	12	Rambutan
5	10	Asparagus, potato, cherimoya

Table 5. Control Atmosphere treatments to alleviating Chilling Injury

Commodity	Storage Temp. (°C)	Storage Period	CO_2 (%)	O_2- (%)	Effects	Reference
Avocado	5	Nine weeks	8	3	- Give the best quality after storage - Reduce the development of CI based on a physical parameter	(Meir *et al*., 1995)
Cucumber	6	6 d	5	5	- Reduce chilling injury based on CI scale	(Mercer & Smittle, 1992)
Cucumber	5	18 d	3	1	- Decreased respiration rate - Minimized losses of fructose, glucose, and malic acid content	(Yi Wang & Qi, 1997)
Empire Apples	3	Six weeks	1.5	3	- Reduce CI based on a percentage of scald incidence	(Burmeister & Dilley, 1995)
Nectarine	0	Six weeks	10	3	- Alleviated CI based on woolliness parameter	(Zhou *et al*., 2000)
Peaches	1	28 d	5 10	1.5 1.5	- Reduce the incidence of woolliness - Inhibition of pectin methylesterase activity	(Santana *et al*., 2011)

Modified atmosphere (MA) depends on the type of packaging due to the permeability film to modify the ideal gas to extend the self-life fruits and vegetables (Cefola *et al*., 2016). Table 6 summarizes modified atmosphere treatments to alleviating Chilling Injury. The utilizing of perforated low-density polyethylene (LDPE) 107µm thickness and 0.075 m² size in cucumber storage at 4°C can reduce chilling Injury (CI) index, maintain firmness, color, and sensory characteristic (Manjunatha & Anurag, 2012). The utilizing of LDPE bag in stored cucumber solved the CI phenomenon. Low CO_2 in the packaging could suppress malondialdehyde (MDA) in 3 days of stored cucumber (Fahmy & Nakano, 2014). Polyethylene (PE) bag has delayed the browning and decay percentage of longan fruit at 2°C for 46 days, which could maintain the shelf life of this fruit (Khan *et al*., 2016). (D'Aquino *et al*., 2016) used oriented polypropylene (OPP) with laser-perforated to extent the self-life of cherry tomatoes. The 3% of O_2- and > 15% of CO_2 successfully reduced water losses, maintained vitamin C, total soluble solids, and firmness.

Table 6. Modified atmosphere treatments to alleviating Chilling Injury

Commodity	**Storage Temp. (°C)**	**Storage Period**	**Type of packaging**	**Effects**	**Reference**
Carambola	10	40 d	Low-density poly-ethylene (LDPE)	- Decrease water losses content and chilling injury index	(Ali *et al*., 2004)
Tomatoes	4	14 d	Polyethylene (PE)	- Reduce weight losses - Maintenance of firmness - Retarded of color change - Reduced CI index such as pitting score	(Park *et al*., 2018)
Pomegranates	5	12 weeks	Unperforated poly-propylene (UPP)	- Reduced weight losses, CI index, and decay	(Artés *et al*., 2000)
Bell pepper fruit	2	Six weeks	Micro perforated polypropylene	- Maintained putrescine content	(Serrano *et al*., 1997)
Green chilies	8	28 days	Anti-fog (RD45) film	- Maintained pigment stability, retention of phenolics, capsaicin, antioxidant, and ascorbic acid	(Chitravathi *et al*., 2015)
Mango	12	Three weeks	Micro perforated PE	- Reduced development of red or green spot	(Pesis *et al*., 2000)
Banana	10	30 d	Non-perforated polyethylene (PE) bags	- Reduced visible chilling injury (greyish peel browning) - Maintained total free phenolic - Reduced phenylalanine ammonia lyase (PAL) & polyphenol oxidase (PPO)	(Nguyen *et al*., 2004)
Cucumber	5	18 d	Seal LDPE	- Reduced CI index and weight losses - Maintained putrescine & spermidine content	(Yi Wang & Qi, 1997)

4. CONCLUSIONS

Many kinds of treatments have been applied by researchers to alleviate chilling injury, and the efficacy of treatment depends on sensitivity commodities, availability of tools, and the aim of the storage. It is divided into physical and chemical treatment to prevent commodity from CI phenomena. Each treatment has an advantage and a disadvantage. The advantage of physical treatment is less of chemical material but need some equipment to adjust the environmental condition such as ideal CO_2 & O_2 concentration and temperature. The popular physical treatments that successfully reduce the risk of chilling injury are intermittent warming, heat treatment, hot water treatment, low-temperature conditioning, controlled atmosphere, and modified atmosphere packaging. Intermittent warming, heat treatment, hot water treatment and low-temperature conditioning require tools to set and control the temperature during storage. Whereas modified atmosphere packaging requires plastic film to create the ideal CO_2 & O_2 concentration using the permeability of the plastic and controlled atmosphere requires special equipment to provide the ideal CO_2 & O_2 composition for fruit and vegetables during storage.

The physical treatments could successfully reduce pitting and decay, black cold damage, weight loss, the occurrence of peel browning, respiration rates, the hard skin and tissue breakdown scald, vitreousness, electrolyte leakage, MDA, lipoxygenase (LOX) activity, PPO activity, phenylalanine ammonia lyase (PAL), ethylene production, and inhibit production of H_2O_2 & O_2--. On the other hand, they may increase the 1-aminocyclopropane-1-carboxylic acid (ACC) levels, TSS, ATP concentration and energy charge (EC), gene expression, and maintenance of antioxidant enzymes, the high level of glycine betaine (GB) content & betaine aldehyde hydrogenase (BADH) activity, the high level of total phenolic content.

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