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Review Article

Factors associated with chilling injury of mango and potential mitigation strategies

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ABSTRACT

Mango is a highly valuable fruit crop grown extensively in tropical and subtropical areas, offering considerable nutritional and economic benefits. However, mangoes are susceptible to chilling injury when exposed to temperatures below 13°C during storage, resulting in substantial postharvest losses. This paper explores the factors contributing to chilling injury in mangoes, including pre-harvest genetic variability, climatic conditions, nutritional status, and post-harvest handling and storage practices. The cellular and molecular mechanisms underlying chilling injury, such as membrane integrity, enzymatic activities, ROS accumulation, and hormonal changes, are discussed. Various mitigation strategies are reviewed, including breeding for chilling-resistant varieties, optimal agronomic practices, controlled atmosphere storage, heat treatments, chemical treatments, and emerging technologies like nanotechnology and genetic engineering. The review provided aims to inform better management practices and technological innovations to reduce chilling injury in mangoes, enhancing their shelf life, postharvest qualities and marketability.

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INTRODUCTION

Mango (*Mangifera indica* L.), a member of the Anacardiaceae family, is a commercially significant fruit crop extensively grown across tropical and subtropical regions in Asia, Australia, Africa, and Latin America (Litz, 2009); (Kiloes *et al.*, 2022). The global production of mango fruits has reached over 55 million metric tons in recent years. India is the largest producer, accounting for approximately 40% of the world's mango supply, followed by China, Thailand, and Indonesia. Mango is well-known for its sweetness, unique fragrance, and rich carotenoid content (Sivakumar *et al.*, 2011); (Sivankalyani *et al.*, 2016). The fruit is not only a significant source of nutrition, providing essential vitamins (mainly A and C), minerals, and antioxidants, but also holds substantial economic value (Manthey & Perkins-Veazie, 2020). Mango production supports the livelihoods of millions of farmers and contributes significantly to the economies of tropical and subtropical regions (FAO, 2020).

Mangoes are climacteric fruits that undergo rapid ripening after harvest. Similar to other fruits, their ripening process can be slowed, and shelf life extended by lowering the storage temperature. However, like many tropical and subtropical fruits, mangoes are vulnerable to chilling injuries when stored below a critical minimum temperature of 13°C (Chaplin *et al.*, 1991); (Lizada, 1991) As a result, prolonged storage triggers metabolic changes that cause chilling injury in fruits, leading to notable alterations in cell walls and membranes (Shewfelt & Del Rosario, 2000). His also reduces ethylene gas production and lowers the respiratory rate (Nair & Singh, 2009). During extended storage, chilling injury manifests as small patches around the lenticels on the fruit's surface, caused by cell death in those areas. Over time, these injuries expand and merge, resulting in large black spots on the fruit's skin (Mittler, 2002). Black lesions with surface pitting, water-soaked appearance, irregular ripening, loss of ripening ability, exocarp browning, and off-flavors are general symptoms of chilling injury in mangoes, which significantly reduces the aesthetic appeal and flavor of mango fruit (Shivashankar *et al*., 2022). These symptoms lower marketability and consumer acceptance, which ultimately suffer the producers and distributors. Likewise, the vulnerability of mango to chilling injury makes it difficulty for storage and transportation.

Factors associated with chilling injury

A. Pre-harvest factor

i. Genetic makeup

The vulnerability of mango to chilling injury varies greatly among the different cultivars. Due to genetic makeup, certain mango varieties are more vulnerable to chilling injury and some are not. For example: some cultivars, such as Okrong, are more resistant to chilling injury than others, such as Nam Dok Ma, based on skin discoloration (Phakawatmogkol *et al*., 2004)

ii. Climate

The vulnerability of mangoes to chilling injury can also be influenced by the climate where they are cultivated. Mangoes grown in regions with high and irregular temperature fluctuations are more prone to chilling injury compared to those cultivated in areas with consistent and moderate temper (Yahia *et al*., 2007).

iii. Nutrient

The vulnerability of chilling injury can be influenced by nutrients i.e. mainly mineral content content present in them. The risk of chilling injury can be minimized by enhancing their cell wall stability and membrane integrity during fruit growth with adequate levels of calcium and other essential minerals otherwise make them more susceptible (Kumar *et al*., 2011); (Singh *et al*., 2020).

B. Harvest and postharvest factors

i. Maturity stage at harvest

The degree of maturity during which mango are harvested plays a crucial role in their susceptibility to chilling injury. Generally, mangoes harvested at an immature stage are more susceptible to chilling injury compared to those picked at a mature but unripe stage (Nair *et al*., 2018); (Jha *et al*., 2021).

ii. Handling practices

Improper handling techniques/practices during and after harvest can make exacerbate chilling injury. For example: mechanical damage like bruising or cuts can serve as entry points for pathogens and make them vulnerable to chilling injury (Yahia *et al*., 2019); (Ghasemnezhad *et al*., 2020).

iii. Temperature

The storage and transportation temperature is a critical factor influencing the occurrence of chilling injury in mango. Temperature below 13°C (55°F) during storage and transportation makes them more susceptible to chilling injury so effective to maintain a temperature around 13°C (Fallik & Aharoni, 2018); (Li *et al*., 2022).

iv. Relative humidity

It can also affect the chilling injury. High humidity can promote mould growth and surface pitting, while low humidity can lead to dehydration and skin shriveling. To minimize these cases, an optimal relative humidity of around 85-90% is recommended (Zhou *et al*., 2017); (Su *et al*., 2021); (Han *et al*., 2022).

C. Physiological and biochemical factors

i. Membrane integrity and lipid composition

Chilling injury in mango is frequently associated with cellular membrane damage. Exposure to low temperatures can cause phase transitions in the membrane, leading to increased permeability and loss of integrity, which may result in ion leakage, tissue degradation, and other symptoms of chilling injury (Zhang *et al*., 2018); (Shi *et al*., 2023).

ii. Enzymatic activities

Chilling injury is primarily driven by the activity of specific enzymes, particularly lipoxygenase (LOX) and polyphenol oxidase (PPO), which are involved in oxidative reactions that cause browning and off-flavors in mango. When exposed to chilling stress, the activity of these enzymes increases, contributing to the development of chilling injury symptoms (Zhao *et al*., 2022).

iii. Accumulation of Reactive Oxygen Species (ROS)

Exposure to cold temperatures can lead to the accumulation of reactive oxygen species (ROS) in mango tissues, which contribute to cellular damage and serve as a key factor in the development of chilling injury (Li *et al*., 2023) .

iv. Ethylene production

Mangos undergo ripening because of a hormone present in plants called ethylene. Chilling stress can disrupt regular ethylene production and signaling pathways, leading to uneven ripening and other symptoms of chilling injury. The interactions between chilling stress and ethylene production can vary among the mango cultivars (Ballester-Costa *et al*., 2022); (Gao *et al*., 2023).

Mechanisms of chilling injury

A. Cellular and molecular mechanisms

Chilling injury in mango involves cellular and molecular pathways that interfere with regular metabolic functions. At the cellular level, chilling temperatures have the potential to induce a phase change in membrane lipids, which would enhance ion leakage and membrane permeability. This results onset of tissue deterioration and loss of cellular compartmentalization (Shi *et al*., 2023). In addition, chilling stress prompts the release of reactive oxygen species (ROS), which induce oxidative damage to proteins, lipids, and nucleic acids, leading to further cellular damage (Li *et al*., 2023).

B. Role of calcium, magnesium, and other minerals

Under stress conditions, minerals such as calcium and magnesium are essential for maintaining cellular structure and function. Calcium stabilizes the cell walls and membrane by binding to phospholipids and proteins, thus preventing leakage and degradation (Lester & Grusak, 2004). On the other hand, Magnesium is an essential cofactor for numerous enzymatic processes, including those associated with antioxidant defense. Adequate levels of these minerals can enhance the fruit's resilience to chilling stress and otherwise increase the susceptibility to chilling injury (Cakmak & Kirkby, 2008).

C. Hormonal changes associated with chilling stress

Chilling stress in mango causes major hormonal changes, especially in gibberellins (GA_3), ethylene, and abscisic acid (ABA). Ethylene is the key hormone in fruit ripening and senescence. Under chilling stress, disruptions in ethylene lead to an increase in susceptibility to chilling injury and uneven ripening of mango (Zhao *et al*., 2022). Similarly, other hormones such as abscisic acid, involved in stress responses, often accumulate under chilling stress and trigger various protective mechanisms, but also contributing to the symptoms of chilling injury (Wang *et al*., 2021). Gibberellins may also be affected further impacting the ability of the fruit to cope with chilling temperatures (Zhang *et al*., 2020); (Shivashankar *et al*., 2022).

D. Genetic and transcriptomic insights

Recent developments in genomics and transcriptomics have provided further insights into the genetic causes of chilling injury in mango. Studies have identified certain genes and gene networks that exhibit differential expression in response to chilling stress. These genes function in multiple activities, such as ethylene biosynthesis, antioxidant defense, membrane stability, and stress signaling (Dautt-Castro *et al*., 2019); (Zhang *et al*., 2022). Studying the expression patterns of these genes can aid in the development of molecular markers for breeding mango varieties that are resistant to chilling injury. For example: transcriptomic analyses have shown that, in chilling-tolerant cultivars relative to susceptible ones, there is overexpression of genes linked to ROS scavenging and downregulation of those implicated in cell wall disintegration (Luria *et al*., 2018); (Li *et al*., 2021).

Mitigation strategies

A. Pre-harvest strategies

i. Use of chilling-resistant varieties

Mango varieties resistant to chilling stress are being developed through breeding programs that select and cross-breed cultivars with genetic characteristics conferring tolerance to low temperatures (Dautt-Castro *et al*., 2019). Genomic methods and marker-assisted selection are being utilized more and more to identify and reproduce these traits. For instance, improving freezing resistance can be achieved by choosing features that are related to increased cell membrane integrity and effective antioxidant systems (Kuhn *et al*., 2017).

ii. Use proper agronomic practices

Mango fruits can be made considerably more resistant to freezing injury by utilizing ideal agronomic practices. For instance, preserving the integrity of the cell wall and overall fruit health depends on ensuring an appropriate supply of vital minerals, especially calcium and magnesium (Cakmak & Kirkby, 2008); (Hocking *et al*., 2016). According to (Zhang L. *et al*., 2019), proper irrigation techniques also help to prevent water stress, which can impair natural defense mechanisms against chilling.

B. Harvest and postharvest strategies

i. Optimal harvest time and techniques

Harvesting mangoe at the optimal maturity stage is essential for reducing chilling injury. Mangoes picked at a mature yet unripe stage are less vulnerable to chilling injury than those harvested prematurely (Subedi *et al*., 2020). Additionally, using proper harvesting techniques helps to minimize mechanical damage and consequently reduce susceptibility (Yahia *et al*., 2021).

ii. Temperature management

To prevent chilling injury in mango, it is important to maintain the correct temperature during storage and transportation. Mangoes should be kept at temperatures above the critical chilling threshold of 13°C (55°F) (Lalel *et al*., 2020); (Yahia *et al*., 2021).

iii. Controlled atmosphere storage

This technique involves the modification of oxygen and carbon dioxide concentration to slow down respiration and delay ripening, thereby reducing the risk of chilling injury. To minimize chilling symptoms, decrease the level of oxygen and increase the carbon dioxide concentration (Yahia *et al*., 2021).

iv. Use of edible coatings and waxes

The use of edible coatings and waxes can help preserve the internal environment of the fruit and minimize moisture loss. Edible coatings act as barriers to gas exchange and can also contain additives that provide additional protection against chilling injury. Similarly, it maintains the internal condition i.e. atmosphere and transpiration (Dhalsamant *et al*., 2020).

v. Application of heat treatments

Heat treatments, such as hot water dips and vapor heat treatments, can induce a form of "heat shock" response that enhances the fruit's tolerance to chilling temperatures thereby maintaining membrane integrity and reducing oxidative stress (Wang *et al*., 2019); (Lurie, 2020).

C. Chemical treatments

i. Application of calcium salts and other mineral dips

Dipping mangoes in calcium salts such as calcium chloride strengthens cell walls and membranes, thereby reducing susceptibility to chilling injury (Singh *et al*., 2020). Additionally, dipping mango in other solutions containing magnesium can also be beneficial (Shah *et al*., 2021).

ii. Use of antioxidants and anti-ethylene agents

The application of antioxidants helps alleviate oxidative stress induced by chilling temperatures, while anti-ethylene agents like 1-methylcyclopropene (1-MCP) can block ethylene activity, reducing uneven ripening and symptoms of chilling injury (Li *et al*., 2020).

iii. Postharvest fungicides

Postharvest fungicides such as thiabendazole help to reduce chilling injury by preventing fungal infections to the chilling-damaged tissues (Charles *et al.*, 2020).

iv. Use of phytohormones

The ability of Mango fruit to withstand chilling stress is strengthened by phytohormones such as methyl jasmonate, which function by preventing oxidative stress and enhancing energy metabolism (Huang *et al.*, 2024).

D. Emerging technologies

i. Use of nanotechnology-based solutions

Nanotechnology provides creative ways to lessen chilling injury. Nano-coatings and nano-encapsulated antioxidants offer targeted protection and controlled release of protective agents thereby enhancing the fruit's resilience to chilling temperatures (Zhang *et al.*, 2020); (Salinas-Roca *et al.*, 2021).

ii. Genetic engineering and CRISPR technologies

Advanced genetic engineering techniques, such as CRISPR/Cas9, can be used to develop mango fruits with enhanced resistance to chilling injury. Cultivars that are more resilient to low temperatures can be created by modifying particular genes related to stress response and membrane stability (Dhekney *et al.*, 2018).

iii. Advanced storage technologies

Precise control against chilling injury is possible because of advanced storage methods like dynamic controlled atmosphere storage, which continuously modify the gas composition in the storage environment based on real-time monitoring of the physiological status of the fruit (Kader, 2002).

CONCLUSION

Chilling injury presents a major challenge in mango postharvest management, impacting fruit quality, shelf life, and marketability. This complex issue is influenced by a range of pre-harvest, harvest, postharvest, and both physical and biochemical factors. The degree of susceptibility to chilling injury differs across mango cultivars, with genetic factors playing a key role. Additionally, climatic conditions during growth, nutritional status, and the maturity stage at harvest all contribute to the fruit's ability to withstand chilling stress. Effective temperature management, gentle handling practices, and maintaining optimal relative humidity are critical postharvest strategies to mitigate chilling injury. At the cellular level, chilling injury is associated with disruptions in membrane integrity, increased enzymatic activities, and oxidative stress due to ROS accumulation. Hormonal imbalances, particularly in ethylene production, are further prone to chilling symptoms. Advances in genomics and transcriptomics have identified key genes involved in chilling stress responses, providing potential markers for breeding chilling-resistant varieties. Ongoing research and technological progress are crucial for developing more efficient strategies to minimize chilling injury and enhance the postharvest quality and marketability of mango.

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