

The Potential of Nitric Oxide in Minimizing Postharvest Physiological Changes and Ethylene Production in Stored Fruit

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Abstract: Fruit postharvest loss is the biggest challenge in addressing sustainable food security and profit maximization. Postharvest losses occur at any stage of the value chain from harvesting time to the final consumer. Fruit continue living processes after harvesting while using the stored nutrients and resulting in ageing, decay and quality loss. The Losses that occur during storage are mainly associated with the extreme fruit respiration rate and ethylene production. In order to maintain fruit quality and extend its shelf life during the postharvest period, there is a need to control the rate of respiration and ethylene production. Different treatments like the use of a controlled atmosphere and some other various compounds like ozone gaseous, Potassium permanganate donors, 1-Methylcyclopropene (1-MCP), Hydrogen sulfide (H₂S), hydrogen gas (H₂), carbon dioxide (CO₂), and chlorine dioxide (ClO₂) were approved to manage the respiration rate and ethylene production during ripening and extend fruit shelf life during storage. Nitric oxide (NO) is another gas that intervenes in many plant life processes from plant growth and development, fruit setting, ripening and senescence which also has the potential to inhibit ethylene synthesis by directly altering the activities of 1-aminocyclopropane-1-carboxylate (ACC) synthase and ACC oxydase or by formation of NO- S-adenosyl methionine (SAM) compound while binding with SAM synthetase and suppressing its activities in the ethylene synthesis pathway. The behavior of NO gas to intervene in different plant physiological processes and have a significant effects on managing postharvest losses in different fruit, nowadays makes NO a beneficial gas in food preservation industry.

Keywords: Nitric oxide, Ethylene, Fruit storage, Physiological changes, Postharvest loss

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1. INTRODUCTION

Fruits are some of the main components of a healthy diet. They contain water, carbohydrates, fats, proteins, fiber, minerals, organic acids, vitamins (Wargovich, 2000; Kader, 2001; Padhy and Behera,

2015) and different phytochemical contents like phenolic compounds, flavonoid, bioactive peptides (Yahia et al., 2019), that individually, or in combination, benefit human health (Septembre-Malaterre et al., 2018). Fruit not only

plays an important role in human health but also contributes more to the global economy in both developed and developing countries (FAO, 2011). Fruit nutritional and economical point of view remains the key driving forces to take advantage in increasing fruit production but it is also known that most of the horticultural products are highly perishable and susceptible to postharvest losses (Kitinoja and Kader, 2015). Therefore to maximize producer profit while also meeting consumers' needs and taking advantage of fruit potential in human life, there is a need for postharvest quality loss management. As the quality of fruit after harvesting cannot be improved all postharvest treatments are aimed at maintaining fruit nutritive values as well as preserving fruit for a long period (FAO, 2008; Mahajan et al., 2017).

All fruit continue their living processes after harvest which means that the post-harvest life of fruit depends on the rate at which it uses up its stored food reserves and its rate of water loss. The produce dies and decays when its food and water reserves run out. Anything that increases the rate of this process may make the produce inedible before it can be used and must be avoided to maintain the fruit's internal and external quality (FAO, 1989). Generally, the shelf life of fruit during storage is directly linked to postharvest biological processes within the stored fruit whereby the increased respiration and extreme ethylene production take the first place. Whenever these processes go on within fruits they result in physiological deterioration and losses as the fruits can no longer replace food materials or water. The produce undergoes an ageing process that is then followed by breakdown and decay even if the produce is not damaged or attacked by decay organisms, it will eventually become unacceptable as food because of this natural rot (FAO, 2008).

Considering the current world population expected to increase and adding to global food security, the increase in yield and productivity, without reducing postharvest losses, will not be sufficient to securing the availability of food in the world as well as maximizing producers' profit (FAO, 2008). Thus, to meet the global food demand all concerns regarding the reduction of fruits postharvest losses are extremely important. Therefore, the development of different approaches to maintain the postharvest fruit quality and extend its shelf life by managing fruit respiration and ethylene production during storage has been on agenda of many researchers for long. In addition to the use of controlled atmosphere (Bodobodak and Moshfeghifar, 2016; Kitinoja and Kader, 2015) to put ethylene and respiration rate under control, some other various compounds like Ozone gaseous (O₃) (Dickson et al., 1992), Potassium permanganate donors (Álvarez-Hernández et al., 2018; Álvarez-Hernández et al., 2019), 1-Methylcyclopropene (1-MCP) marketed under the commercial name 'SmartFresh™' (Kader, 2013), Hydrogen sulfide (H₂S) (Ziogas et al., 2018), hydrogen gas (H₂), carbon dioxide (CO₂), and chlorine dioxide (ClO₂) (Gonga et al., 2018) has been also examined to manager the respiration rate and ethylene production during ripening as way to extend fruits shelf life

during storage. Despite those compounds put ethylene under control, each of them has its limitation. More researches are going on to manage ethylene production as well as other physiological changes occur during fruits storage. Nowadays, new approach of using NO has also been a talks for different researchers to inhibit the ethylene production during fruits storage (Li et al., 2012; Zsuzsanna et al., 2019; Gonga et al., 2018).

The chemistry of Nitric oxide (NO) is determined by the fact that it is a radical diatomic molecule gas containing a double bond between the N and the O atoms that makes it a relatively unstable (i.e., an unpaired electron) as redox natural gas it can gain or lose one electron to form the ions (NO⁻ or NO⁺) (Lancaster, 2015). Ethylene (C₂H₄, CH₂=CH₂) is also unstable gas containing two C atoms with a double bond but with no radical (Bakshi et al., 2015) Therefore, differences concerns were laid from both gases having some similar behaviour. In addition both NO and ethylene can be synthesised in more similar pathway by plant from amino acids or applied from external source and influence plant physiological processes as as an agent for delaying or mitigating stress (Bakshi et al., 2015; Palma et al., 2019). NO was initially regarded as an environmental pollutant, but since the different trials showed its intervention into plant biochemical processes, it has gained more attention by world reserachers. Leshem (1996) reported that the endogenously-produced or exogenously-applied NO even at low concentration exert significant plant growth-promoting and ethylene inhibiting effects. From that time much more researches have been done to benefit what was considered as environmental pollutant. NO first started being used as agent of regulating defence during pathogen infection, afterwards it was described to involve in many other plant physiological processes from initial growing to senescence. NO has been reported to participate in many plant growth and development including fruit setting ripening and senescence of flesh fruit (Palma et al., 2019). The other example of effective use of NO in storage was shown in papaya fruits storage where fruits treated with NO showed lower rate of both CO₂ release (respiration) and ethylene production than the control fruits (Li et al., 2012). Therefore to have some insight into the involvement of NO in different stage of fruit value chain, this studies aimed at examining and bringing together the potentials of NO in regulating fruits postharvest physiological changes during storage and the interaction of NO and ethylene biosynthesis in postharvest shelf life of fruit crops.

1. Postharvest physiological changes and quality loss of fruit during storage

1.1. Postharvest physiological changes of fruit related to ethylene production

As fruits are living tissues they are subjected to continuous physiological change after harvest (Imahori, 2016). Ripening is the most developmental process

through which the fruit undergoes physiological and biochemical changes (Table1).

The main changes associated with ripening include color, firmness (softening by cell-wall degrading activities), carbohydrates, organic acids, lipids, phenolics, volatile compounds, structural polysaccharides and softening of texture and biosynthesis of pigments (carotenoids and anthocyanins) (Wills et al., 2000; Marti'nez-Romeo and Bailen, 2007). All these changes are well-known to be regulated by fruit respiration, ethylene production (Kader, 2013) and other metabolic activities.

While some changes are desirable, most are not. This means that postharvest changes in fresh fruits cannot be stopped, they can only be slowed to a certain extent to preserve the nutrient contents (Mahajan et al., 2017). The most important aspects of metabolism changes,

concerned with postharvest losses of fresh fruits are respiration, (aerobic and anaerobic), transpiration and water loss, physiological responses to stress or oxidative metabolism, extreme ripening and the final stage called senescence.

During the postharvest period most of all these changes vary with respect to the type of fruit whether is climacteric or non-climacteric even though changes occur in all fruit. Fruits that ripen after harvest are classified as climacteric and they typically ripen by softening significantly, by changing in color and other biochemical properties and becoming sweeter while non-climacteric fruits do not change significantly after harvest, they will soften a little, lose green color and develop rots as they get old but they do not change to improve their eating characteristics (Jobling, 2000).

Table 1: Some postharvest physiological and biochemical changes related to ethylene production

Fruit	Postharvest fruit physiological and biochemical changes	References
Apple	Ethylene production, loss of firmness, increase in total soluble solids (TSS) content and decrease in malic acid content. Softening of fruit as a result of the breakdown of bonds between adjacent cells (protopectins) in the cell walls form soluble pectins which in turn degrade into sugars, as well as changing in fruit color from green to yellow, resulting from the degeneration of chlorophyll in the skin. Conversion of starch to sugars and synthesis of other organic compounds	(Giné-Bordonaba et al., 2019) (Robert et al., 2018; (Ornelas-Paz et al., 2018; Kvikliene et al., 2006)
blueberry	Significant decrease in fruit firmness of blueberry fruit with respect to cultivar, change of fruit flavor an increase in TSS content and pH and high softening rate due to degradation of cell wall	(Liu et al., 2019)
Tomatoes	Softening of the fruit tissue from Polygalacturonase (PG) actions, conversion of starch to sugar, accumulation of secondary metabolites affecting appearance, taste and aroma	(Osei et al., 2017)
Pineapple	Change in metabolites such as amino acids and amines, organic acids, sugar, sugar acids and sugar alcohols and fatty acids	(Luengwilai et al., 2018)
Banana	Increase total soluble sugar and fruit appearance and taste. Enhance fruit quality and health related issues while assuring good flavor, texture, and uniform peel color and increasing vitamin C and titratable acids	(Hussen, 2014; Maduwanthi and Marapana, 2019)
Mango	Changes in the majority of the physico-chemical properties such as total soluble sugars, starch, and total soluble solids contents, titratable acidity, pH, firmness and the ratio of total soluble sugars to total organic acids.	(Brecht and Yahia, 2009; Joas et al., 2009; Ntsoane et al., 2019; Lei Yi et al., 2019)
Strawberry	Change in the color, texture, flavor, and aroma of fleshy fruit and an increases in water soluble polyuronides and significant mineral and vitamin content	(Kuchi and Sharavani, 2019)
Pomegranate	Increases in juice content and Total Soluble Sugar content TA decreased, pH increased, decrease in phenols and ascorbic acid level with parallel increase in anthocyanins and color change to dark red	(Pareek et al., 2015)

Climacteric fruits are characterized by an increase in the rate of respiration and ethylene production at the early

stages of ripening while non-climacteric fruits are those in which no increase or reduction in the respiration rate and

ethylene production takes place during the whole ripening period (Alós, et al., 2019) which means even though respiration and ethylene production exist in non-climacteric fruits, the rate is relatively constant. The main difference between those two types of crops is shown by the external ethylene treatment whereby climacteric fruit responds to ethylene application while non-climacteric fruit shows no response in terms of biochemical changes. The ethylene level around non-climacteric produce would seem to be in equilibrium with the surrounding ambient air rather than adding to the ambient air of the storage room (Wills et al., 2000).

During the ripening period there is also a change in fruits color, aroma, volatile gases and other organic acids and for all these changes the intervention of ethylene has been reported (Alós et al., 2019). For example, banana as climacteric fruit, it is harvested before ripening and can be artificially ripened to meet customer preferences. One of the most commonly used ripening agent is ethylene with proper temperature and humidity (Maduwanthi and Marapana, 2019). The applied ethylene accelerates the chemical changes associated with ripening and reduces time to banana climacteric rise, whereby starch decreases from about 20 to 1 % and sugars increase from about 1 to 18 % and the nitrogen content in the edible portion of the ripe banana amounts to about 0.2 % of the fresh weight (Hussen, 2014).

1.2. Ethylene involvement in fruit quality losses during storage

Food losses is a critical component of ensuring future global food security (FAO, 2011). Losses may occur at any point in the whole value chain, from the initial harvest through assembly, distribution and storage to the final consumer (Kitinoja and Kader, 2015a). The dramatic loss of fruits during harvesting and postharvest activities is due to their perishable nature and poor postharvest practices and facilities (Kitinoja and Kader, 2015; Teutsch, 2019; Porat et al., 2018)). Postharvest loss of fruit is difficult to predict as the major agents producing deterioration are those attributed to physiological changes and combinations of several organisms (FAO, 2011). Physiological changes of fruit during storage such as metabolic activities and biochemical reactions as natural processes lead to an increased respiration rate and ethylene production result in significant losses of nutritional value of fruit (Kitinoja and Kader, 2015; table 2). As reported by different studies, ethylene also known as the "ripening hormone or stress hormone" is a gas that plays a regulatory role in many processes of plant growth, development and eventually death (Dhall, 2013).

Table 2: Examples of postharvest fruit quality losses from extreme ethylene production

Fruit	Fruit quality losses related to ethylene	References
Apple	Causes a severe softening from reduced cell turgor during ripening process	(Jason et al., 2002; Robert et al., 2018)
Strawberry	Enhanced postharvest decay and chilling injury and reducing the quality of fresh fruit.	(Wills et al., 1999; Wills et al., 2000)
Zucchini	Loss of fruit weight, carotenoid content and high incidence to chilling injury	(Megías et al., 2016)
Papaya	Reduction of fruit folate content and taste	(García-Salinas et al., 2016)
Tomato	Firmness and weight loss of tomato fruit resulted from water loss and respiration rate whereby tomato fruit stored at 20 and 30°C showed significant increase in lycopene, β -carotene, chlorophyll degradation and weight and firmness loss	(Tadesse et al., 2015)
Pears	Enhanced senescence and losses of soluble sugars	(Zhai et al., 2018)
Mango	Substantial loss of organic acids, Loss of fruit weight associated to fruit softening and change in pectic substances cell wall as well as enhanced chilling injury at low temperatures.	(Brecht and Yahia, 2009; Lei Yi et al., 2019)
Pomegranate	The weight loss in mainly caused by water transpiration and CO ₂ loss during respiration	(Farid et al., 2018)

During storage of different climacteric fruit, ethylene has been scientifically proven to cause an increase in respiration, yellowing or spotting, enhanced production of ethylene as it is autocatalytic, accelerated ripening, ageing and decay, reduction in fruit nutrient contents (e.g. loss of vitamin C, carbohydrates and organic acids), taste and aroma changes (Wills et al.,

2000) and off-flavors such as bitterness resulting in decreasing fruits quality and shelf life and the degree of damage depends upon its concentration, length of exposure time, and storage temperature (Kader, 2013), while in non-climacteric fruit, ethylene induce fruit color change implies a parallel change in carotenoid content (Megías et al., 2016; Alos et al., 2019) as well as some

postharvest chilling injury in low temperature storage (Megías et al., 2016). An increased storage life in orange fruit with a decrease in ethylene concentration at 20°C was reported, however a similar effect of ethylene concentration on orange storage life at 2.5°C was limited by the appearance of chilling injury whereby there was a linear increase in the time to develop chilling injury with a logarithmic decrease in ethylene concentration (Wills et al., 1999). The study carried out on strawberries also showed that 30% of Strawberry loss occurred at 0.035-0.221 $\mu\text{L/L}$ ethylene level during storage (Wills et al., 2000).

2. General overview of the effects of Nitric oxide on biochemical changes and its quality control behavior during storage

Treatment with exogenous nitric oxide (NO) delays fruit ripening, prevents chilling damage, promotes disease resistance, and enhances the nutritional value (Palma et al., 2019). In papaya fruits, the level of vitamin C initially increased and then declined slightly during storage at a slower rate, low ethylene production, low weight loss, delayed changes in peel color maintained firmness and soluble solid content in the NO-treated fruits than in controls (Li et al., 2012). Several other studies examined the effects of NO treatments on different fruits and proved to be effective in maintaining postharvest fruits quality (Table 3).

According to Zhu et al., (2006), the firmness of 5 and 10 $\mu\text{l L/L}$ NO-treated peaches decreased while the contents of soluble solids increased slowly during the first period of storage at both 5 and 25 °C. Increased fruit firmness, a lower respiration peak and reduced softening as well as delayed fruit pulp color development and ripening were also observed in mango fruits treated with nitric oxide (Tran et al., 2015). Low ethylene production in stored apple fruits with nitric oxide (Rudell and Mattheis, 2006) and inhibition of browning in apple slices treated with Nitric oxide donor compounds were also reported (Pristijono et al., 2008). Golden delicious apple variety sprayed with 5 sodium nitroprusside 14 days before harvest resulted in reduction of ethylene production during harvest and when stored at 18°C compared to non-treated fruit. In non-treated apple there was the rate of ethylene production of 155.46 $\text{nL g}^{-1} \text{FW h}^{-1}$, 2.28 fold higher than that of SNP-treated fruit. SNP-treated fruit contained higher NO concentrations and glucose content than control fruit and later after the glucose contents of control fruit increased at day 15 and 23, 51.11% and 33.64% higher, respectively, than in treated fruit but at 23 day the sucrose content in SNP treated fruit found to be 2.13 fold higher than initial and also to 89.76% higher than in control fruit 8 and 15 days after harvest which reflects the activities of NO in suppression of sucrose degradation to produce glucose

rather favor the sucrose accumulation during golden delicious apple fruit storage (Deng et al., 2013).

In strawberry storage, Zhu & Zhou, (2007) showed that 5 $\mu\text{l L/L}$ sodium nitroprusside (SNP) a nitric oxide donor compound could extend the post-harvest life of strawberry fruit where the ACCS activity of the fruit treated with SNP was lower than the control and exceeded storage days over the control. The similar result was found in another study conducted on selva strawberry where the fruits treated with SNP were less subjected to rot during storage compared to non-treated fruits (Abdollahi et al., 2013). Moreover, according to Zhang et al., (2014) the respiration and decay rate of strawberries dipped in solution of a combination of hydrogen sulfide and nitric oxide donor known as 'sodium nitroprusside' and stored at 20°C was lower than the control. In addition, the decay rate of the tomatoes treated with sodium nitroprusside was also lower than those treated with hydrogen sulfide alone as well as the control which shows the intervention of nitric oxide in reducing strawberry decay during storage.

According to Eum, et al., (2008), the nitric oxide (NO) treatment on tomato (*Solanum lycopersicum* 'Myrock variety') fruits delayed the burst of ethylene production and color development in both mature green and breaker stage fruits. Lai et al.,(2011) also reported that the peel color change and ethylene production of tomatoes stored at 25°C were suppressed by the application of sodium nitroprusside (NO donor). In first 12 day of storage the rate of ethylene was 5.7 $\mu\text{mol kg}^{-1} \text{h}^{-1}$ in tomato without SNP treatment and reduced gradually while the one of tomatoes treated with SNP was still lower at rate of 4.9 $\mu\text{mol kg}^{-1} \text{h}^{-1}$ even after 16 days. In addition to reducing ethylene production and suppressing of peel color change, the NO application maintain the quality of tomato by delaying the rate of total titrable acids (TA) and firmness decrease during storage.

In pomegranate fruit storage, the application of 1000 μM sodium nitroprusside (SNP) as a nitric oxide donor on pomegranate reduce the chilling injury at significant extent compared to untreated fruit and total soluble solid were high in untreated fruit than in SNP treated fruit reflecting a decrease in respiration and decrease the conversion of carbohydrates to sugars, as a consequence TSS decreases in SNP treated fruit (Ranjbari et al., 2016).

Table 3: The Effects of NO or its donor compounds (SPN) on quality preservation during fruit storage

Fruits	Quality preservation during fruit storage	Treatments	References
Papaya	Delays the increase of TSS during storage by retarding the glycolysis process and maintains fruit firmness by reducing water loss and inhibiting the cell wall integrity Decreased polygalacturonase, pectin methyl esterase, pectate lyase and cellulase activities resulted in maintaining fruit firmness	60 μM /L NO	(Li et al., 2012)
		60 μM /L NO at 20 °C with 85% RH	(Guo et al., 2013)
Strawberry	Extends fruit postharvest life by delaying onset of rotting and fruit softening.	5 $\mu\text{mol L}^{-1}$ SNP 5-10 $\mu\text{mol L}^{-1}$ SNP at 20 and 5°C	(Abdollahi et al., 2013; Wills et al., 2000a)
Tamato	Delays peel color change and suppress the ethylene production by delaying 1-aminocyclopropane-1-carboxylate synthase (ACCS) and 1-aminocyclopropane-1-carboxylate oxydase (ACCO) activities. Preserves the fruit firmness during 20 days storage at 25 °C and enhances tomato fruit resistance against gray mold rot caused by <i>B. cinerea</i>	1mM GSNO+0.5 $\mu\text{L L}^{-1}$ 1-MCP.	(Steelheart et al., 2019);
		200 $\mu\text{L L}^{-1}$ NO	Eum et al., 2008).
		1mM SNP aqueous solution	(Lai et al., 2011)
Pomegranate	Increased the antioxidant activity, total anthocyanin content and reduced chilling injury	300 μM NO	(Ranjbari et al., 2018)
		1000 μM NO at 5 °C and 85% RH	Ranjbari et al., 2016)
Peach	delays senescence and altered the enzyme activities of sugar metabolism resulting in sustaining a higher sucrose content of fruit during storage Reduces the rate of fruit respiration and ethylene production and maintain the fruit fleshness	10 $\mu\text{L L}^{-1}$ of NO at 25 \pm 1 °C	(Han et al., 2018) (Liu et al., 2019a)
		100 μM SNP	
Apple	Stimulates sucrose synthesis by inhibiting decomposition of sucrose and Alters 1-aminocyclopropane-1-carboxylate synthase (ACCS) and 1-aminocyclopropane-1-carboxylate oxydase (ACCO) activities hence inhibit ethylene production	50 μM sodium nitroprusside (SNP)	(Deng et al., 2013)
Sweet cherry	Increases total phenolics content and vitamin C, Reduces decay index and weight loss.	10 μmolL^{-1} NO	(Asghari, Khalili, Rasmi, & Mohammadzadeh, 2013)
Mango	Reduces the rate of respiration and ethylene production, exhibits the maintenance of fruit firmness and water loss and inhibits mango peel and pulp color change as well as decreases the changes in TSS and TA in mango fruit.	1 or 2 μM of SNP solution for 30 min	(Linh, et al., 2014)

The similar results were reported in the study of the effectiveness of effectiveness of the individual application of nitric oxide or cellophane wrapping, and combination effects of these treatments on reducing chilling injury and quality improvement of pomegranate fruit during storage where the highest chilling injury index was observed in untreated fruit after 45 days and there a significant maintainance of antioxidant activity in treated fruit with 300 μM of nitric oxide compared to the control for 90 days of storage where total anthocyanin content of treated fruits was higher than the control during storage (Ranjbari et al., 2018).

3. Nitric oxide and ethylene biosynthesis and their relationship during fruits ripening

Ethylene (C₂H₄) plays a role in the postharvest life of many horticultural crops such as speeding up

senescence and reducing shelf life and as well as, improving the quality of the product by promoting faster, more uniform ripening before retail distribution (Dhall 2013). On the other hand there is a new concept of NO intervention ethylene production by acting as antagonism during fruits storage (Manjunatha et al., 2010). This brought a hope to increase fruits shelf life to cope with postharvest losses resulted from extreme ethylene production. Thus, the best way to understand the interaction of both gases is to have a look on their synthesis.

Ethylene synthesis starts by conversion of methionine (MET) amino acid to S-adenosyl methionine (SAM) with help of ATP. Then, ACC synthase, converts SAM to 1-aminocyclopropane-1-carboxylic acid (ACC), followed by the immediate production of ethylene by ACC oxydase (Adams and Yang, 1979; Fig.1). Actually, 1-Aminocyclopropane-1-carboxylic acid synthase (ACCS)

and 1-aminocyclopropane-1-carboxylic acid oxidase (ACCO) are the two enzymes specific for the ethylene biosynthetic pathway and any treatment that can alter or remove one of these enzymes will total inhibit the ethylene production (Rzewuski and Sauter, 2008) thus resulting to reduced senescence and increased shelf life (Dhall, 2013). Any postharvest intervention that reduces ethylene levels around produce will therefore have a positive effect on postharvest life. The application of 1-MCP had divergent effects on fruit respiration and ethylene production rates, and on decay development (Li et al., 2016). The 1-MCP efficiency of retarding ripening is linked with its ability blocking ethylene receptors and benefits post-harvest quality of fruits.

Nitric oxide (NO) is also one of gases that has been proven to reduce ethylene production during ripening. Many advances have been obtained regarding NO synthesis and its physiological effects in plants. However, the molecular mechanisms underlying its effects remain poorly understood (Ferreira and Cataneo, 2010).

Generally the NO synthesis involves nitric oxide synthase (NOS) which converts L-arginine to L-hydroxyarginine and subsequently to nitric oxide and citrulline with participation of O₂ and NADPH, despite NO biosynthesis in plant seems unclear, different studies report to be the same processes of NO biosynthesis in animal cells (Procházková et al., 2014; Baudouin and Hancock, 2014) and the same enzymes NO synthase responsible for NO synthesis was found in different plant species and the plant enzyme shares many biochemical and kinetic with animal NO synthase and also showed that NO biosynthesis inhibit methionine synthesis (Chandok et al., 2003) but recommended further research on mechanism involves. Therefore Methionine being the

precursor of S-adenosyl methionine (SAM), it is believed that inhibiting excess methionine will alter the formation of S-adenosyl methionine (SAM) and lead to reduce ethylene production.

Thus NO as bio-active molecule can regulate ethylene production via at least two mechanisms; through direct inhibition or suppressing the ethylene biosynthetic enzymes. The first mechanism, chemically NO inhibits the hydrogenation of ethane to ethylene and delay plant maturation and senescence (Leshem et al., 1998) while the second mechanism involve inhibition of ethylene biosynthesis by suppressing of 1-aminocyclopropane-1-carboxylic acid synthase (ACCS) and 1-aminocyclopropane-1-carboxylic acid oxidase (ACCO) activities whereby NO binds with 1-aminocyclopropane-1-carboxylic acid (ACC) and ACC oxidase to form a stable ternary complex known as ACC-ACO-NO complex (Manjunatha et al., 2012) resulting in reduction ethylene precursor 1-aminocyclopropane-1-carboxylic acid (ACC) content and decrease the ethylene production (Fig.2). From other different findings, It was observed that, even though there was no clear mechanism of how NO bind with ACCO, in peaches treated with 5 and 10 µl L/L NO, 1-aminocyclopropane-1-carboxylic acid (ACC) oxidase activity and ethylene production activity were reduced (Zhu et al., 2006). The reduction of ethylene production was also observed in apples treated with NO gas compound (Rudell and Mattheis, 2006). According to Zhu and Zhou, (2007) the application of nitric oxide (NO) donor sodium nitroprusside (SNP) of 5 µl L/L level to strawberry reduced the activities of ACCS and ACCO in particular that of ACCS which resulted in reduced ethylene production and ACC content and maintained the qualities of strawberry during storage.

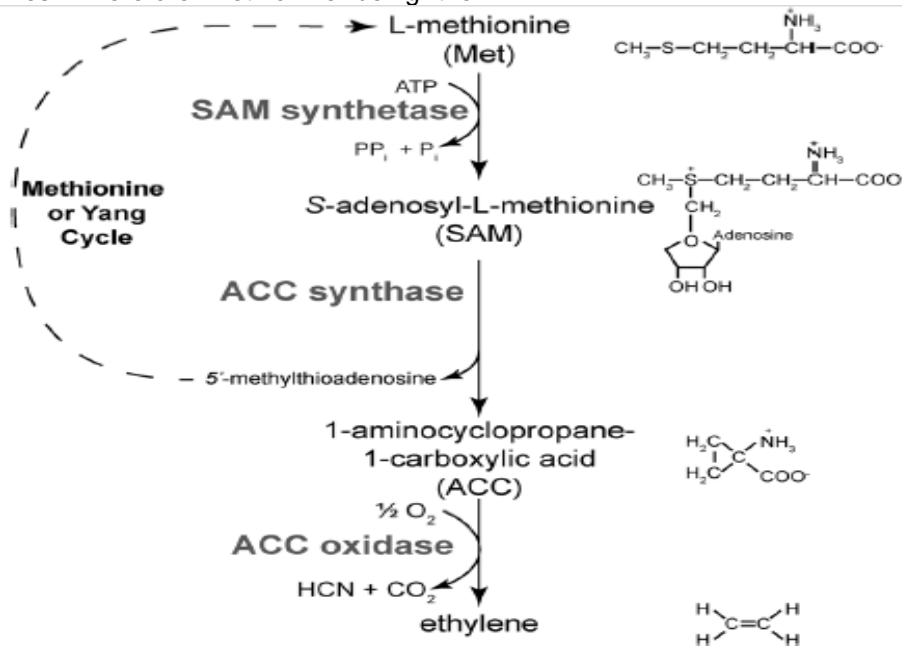


Figure 1: Biochemical pathway for ethylene biosynthesis in plants: In plants, ethylene is biosynthesized from the amino acid methionine. Three enzymes are involved: S-adenosyl methionine (SAM) synthetase converts methionine to SAM, ACC synthase converts SAM to ACC, and ACC oxidase converts ACC to ethylene. SAM synthetase and ACC synthase are part of the Yang or methionine cycle. The reaction catalyzed by ACC synthase is the rate-limiting reaction for ethylene biosynthesis (Bakshi et al., 2015).

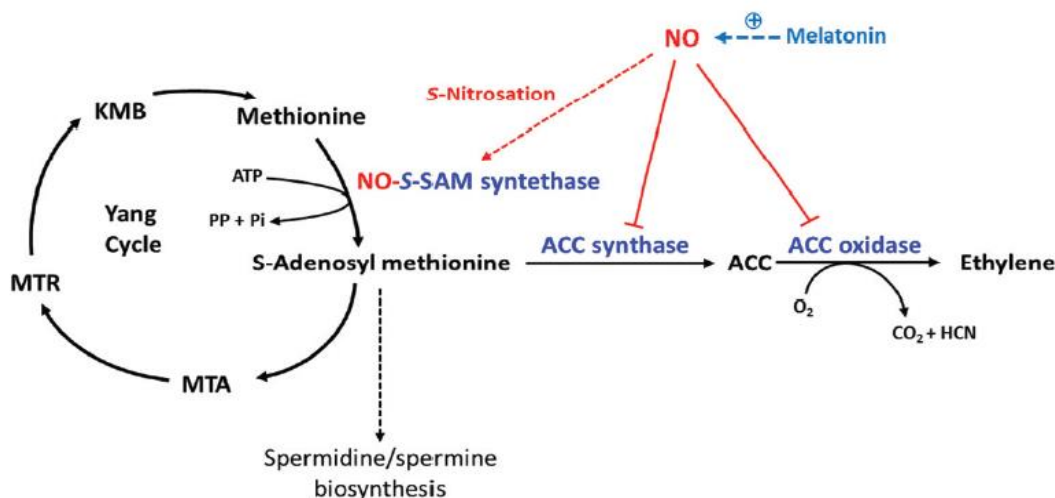


Figure 2: Ethylene biosynthesis pathway and methionine recycling in plants: Ethylene is synthesized in a two-step pathway that involves the enzymes ACC synthase and ACC oxidase. S-adenosyl methionine (SAM) is continuously replaced by a methionine cycle (the Yang cycle), although it can also be diverted to the biosynthesis of the polyamines spermidine and spermine. Nitric oxide (NO) inhibits the activities of ACC synthase and ACC oxidase, and it also affects SAM synthetase (also known as methionine adenosyltransferase, MAT) by a S-nitrosation event. Melatonin can stimulate the biosynthesis of NO. ACC, 1-aminocyclopropane-1-carboxylic acid; MTA, 5-methylthioadenosine; MTR, 5-methylthioribose; KMB, 2-keto-4-methylthiobutyrate (Palma et al., 2019).

4. CONCLUSIONS

Considering the current world population which is also expected to increase by more fold and add to global food demand, to meet this demand, food availability and accessibility cannot be reached by only the increasing of farm production but together with food losses management. Thus, the reduction of post-harvest food losses is a critical component of ensuring future global food security. Considering the facts that ethylene is one of most causes of fruit postharvest losses ethylene management plays a pivotal role in maintaining the postharvest life and quality of horticultural produce. Researchers have developed various approaches to manage ethylene and minimize postharvest losses during fruit storage. One of those approaches is the use of NO treatment during postharvest fruit storage. Despite NO gas that being initially considered as agent environment pollution, the use of NO gas in fruit preservation has become a new way to reduce postharvest losses and extend fruit shelf life. NO inhibits ethylene synthesis throughout the S-nitrosation pathway or directly altering the activities of ACC Synthase and ACC oxidase. Hence NO can be used to manage the postharvest losses mainly those associated with respiration and ethylene production.

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