Review Article

ROS and *MAPK* Cascades in the Post-harvest Senescence of Horticultural Products

Mohamed Hawali Bata Gouda[1], Chunjie Zhang[1], Junhao Wang[1], Sijia Peng[1], Yuru Chen[1], Haibo Luo[1]* and Lijuan Yu[2]

[1]*School of Food Science and Pharmaceutical Engineering, Nanjing Normal University, Nanjing, P.R. China;* [2]*Institute of Agro-Products Processing, Yunnan Academy of Agricultural Sciences, Kunming, P.R. China*

ABSTRACT

Plants are naturally sessile and cannot move away from adverse environmental conditions. Environmental stress may induce loss of membrane integrity, which is a seminal feature of premature senescence. Therefore, plants must respond in other ways to protect themselves from abiotic and biotic stresses that involve protein kinases, which are crucial to signal transduction pathways. Protein kinases are involved in the phosphorylation of serine/threonine and tyrosine side chains of proteins. Among these protein kinases, mitogen-activated protein kinase (*MAPK*) cascade genes are key components of signal transduction pathways that help transduce extracellular signals to intracellular responses in animals, plants, and fungi. Interestingly, reactive oxygen species (ROS) are important and common messengers that are produced in various biotic and abiotic stresses; ROS are known to activate many of the *MAPKs*. In this review, we highlight the mechanisms of crosstalk between ROS and *MAPK* cascades in the post-harvest senescence of horticultural products and summarize recent findings about *MAPK* regulation and functioning in various cellular processes.

Keywords: ROS; *MAPK*; Signal transduction; Post-harvest; Senescence

INTRODUCTION

As perishable commodities, fresh horticultural crops are prone to post-harvest economic losses of quality and quantity due to biotic and abiotic stresses [1]. These stresses lead to a series of morphological, physiological, biochemical, and molecular changes in horticultural products [2]. To address these changes, plants have evolved both passive and active mechanisms to prevent infection. Active defense mechanisms are based upon the early detection of and quick response to invading organisms. Plants utilize resistance proteins (e.g., protein kinases) and other receptors to detect the presence or activity of pathogens [3]. Plants respond to abiotic and biotic stresses that involve protein kinases and are crucial to signal transduction pathways. Among these protein kinases, mitogen-activated protein kinase (*MAPK*) cascade genes are key components of signal transduction pathways that help transduce extracellular signals to intracellular responses in animals, plants, and fungi [4]. Interestingly, reactive oxygen species (ROS) are important and common messengers that are produced in various biotic and abiotic stresses, and are known to activate many of the *MAPKs* (Figure 1). ROS were initially thought to be toxic byproducts of
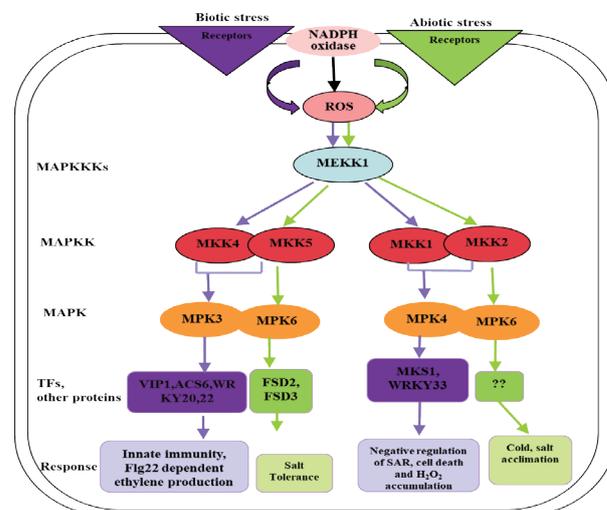

Figure 1: Response of *MAPK* cascades activated by ROS in biotic and abiotic stresses. *MEKK1* is a common *MAPKKK* activated by ROS that are produced in response to both biotic and abiotic stresses. *MEKK1* activates distinct downstream components of the *MAPK* cascade in *Arabidopsis*. Purple and green colors represent biotic and abiotic stresses, respectively.

aerobic metabolism, but they are now regarded as central players in the complex signaling network of cells [5].

The classical *MAPK* signaling cascade is minimally composed of three kinases, namely, *MAPK*, *MAPK* kinase (*MAPKK*), and *MAPKK* kinase (*MAPKKK*). These kinases operate as sequential signal transducers that channel, integrate, and amplify information from the cellular environment to transcriptional and metabolic response centers via phosphorylation. *MAPKs* are activated by *MAPKKs* through the phosphorylation of conserved threonine and tyrosine residues in the Thr-X-Tyr (T-X-Y) motif that is located in the activation loop (T-loop) between the catalytic subdomains VII and VIII. Furthermore, *MAPKKs* are activated by *MAPKKKs* through phosphorylation of serine and serine/threonine residues in the S/TXXXXS/T motif [6]. Previous efforts to identify and characterize *MAPK* cascade proteins in rice, *Arabidopsis* and other plants have provided a wealth of information. In this review, we highlight the mechanisms of crosstalk between ROS and *MAPK* cascades in the post-harvest senescence of horticultural products and summarize recent findings regarding *MAPK* regulation and functioning in various cellular processes.

LITERATURE REVIEW

New advances in the post-harvest senescence mechanism of horticultural products

Fresh horticultural products are living tissues that experience continuous changes after harvest. Their commodities are perishable products, with active metabolism during the post-harvest period [2], which induces post-harvest losses. These post-harvest losses, especially in terms of quality and nutritional composition, could be better controlled with an improved knowledge of their post-harvest physiology [7]. The primary objective of post-harvest research has been to maintain quality, while extending the shelf lives of harvested horticultural commodities. These objectives have been most effectively realized by regulation of the rate of respiratory metabolism [8], which changes throughout the life of the commodity. Luo et al. [9] found that the respiratory metabolic pathways of post-harvest fruits and vegetables vary during storage. Metabolic activity is especially high during the initial growth of the commodity, ripening of climacteric fruit, and periods of wound healing. After an initial surge to repair the wounds encountered during harvest, respiration typically declines in vegetative tissues and non-climacteric fruit (Figure 2) [8]. Plant senescence is affected by respiratory metabolism, which provides the energy for a range of biochemical processes [9]. In leafy crops such as salads, pre-harvest and post-harvest stresses (even of short duration) contribute to changes in metabolism that accelerate quality losses [10]. The major symptoms of post-harvest senescence of *Zizania latifolia* are shell etiolation, surface browning, transpiration, respiratory disorders, and tissue hollowness [9].

Respiration in plants involves several pathways, such as the Embden–Meyerhof–Parnas pathway, tricarboxylic acid cycle, mitochondrial electron transport, cytochrome pathway, pentose phosphate pathway, and glyoxylate pathway. Each pathway has unique biological functions. The Embden–Meyerhof–Parnas pathway, which oxidizes glucose to pyruvate, is a basic respiratory pathway followed by the tricarboxylic acid cycle and cytochrome pathway, which are essential for energy provision in post-harvest fruits and vegetables during storage. Unlike the many diverse
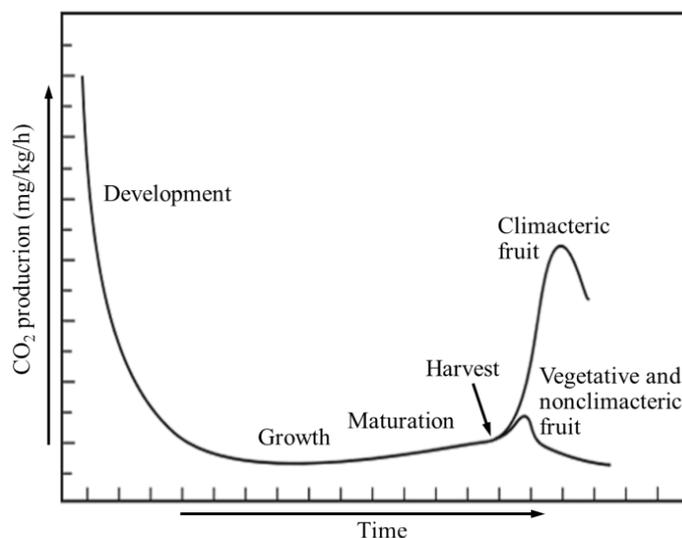

Figure 2: Changes in respiration (CO_2 production) during the development, growth, maturation, and harvest of vegetative commodities and of climacteric and non-climacteric fruit.

and often unique reactions that include secondary metabolism, these four pathways contain sets of reactions that are common to all vascular plants [8]. Recent studies have shown that the energy deficit caused by a decline in adenosine triphosphate synthesis is the main cause of membrane damage and browning of post-harvest horticultural products [11]. Moreover, Aghdam et al. [1] reported that sufficient intracellular adenosine triphosphate supply and supportive extracellular adenosine triphosphate signaling are crucial for attenuation of stresses, postponement of senescence, and maintenance of quality in post-harvest horticultural crops. In addition, reactive oxygen species accumulation can cause inhibition of the tricarboxylic acid cycle in mitochondria, as well as the upregulation of glycolysis and the oxidative pentose phosphate pathway [5].

Reactive oxygen species metabolism and senescence

Plants experience two principal stresses, biotic and abiotic, both of which can be deleterious. A common feature of the plant response toward these stressors is the production of ROS. Plant cells continuously generate ROS in various organelles from the electron transport chains of photosynthesis and respiration [12]. The reactive oxygen species signal itself may carry a decoded message, similar to the calcium signals that contain specific oscillation patterns within defined cellular locations [5]. NADPH oxidase, photorespiration, amine oxidase, and cell wall-bound peroxidases are sources of ROS production in plant cells [13]. Reactive oxygen species are known to play toxic roles in lipid peroxidation, membrane damage, and protein and DNA oxidization processes, resulting in cell death [14,15]. High levels of ROS can lead to phytotoxicity, whereas relatively low concentrations can be used for acclamatory signaling [2]. The roles of ROS as both toxic byproducts and as signaling molecules have been reported in fruit development and ripening [14]. To prevent accumulation of toxic levels of ROS, plants develop enzymes and non-enzyme antioxidant chemicals as defensive forces against excess ROS [12]. The enzymatic antioxidant system represents the primary mechanism for controlling production of ROS, thereby partly regulating the degree of lipid peroxidation. Superoxide dismutase, catalase, ascorbate peroxidase, and peroxidase are the key antioxidant enzymes of ROS scavenging

[15]. Glutathione peroxidase, superoxide dismutase, and catalase play important roles in balancing the induction and removal of ROS in plants; these proteins are encoded by families of closely homologous genes [14]. Indeed, it was found that superoxide dismutase can protect cells from oxidant stress by dismutation of $O_2^{\cdot}$ to H_2O_2; in contrast, catalase, peroxidase, and ascorbate peroxidase are enzymes that control the decomposition of H_2O_2. High levels of activity of these antioxidant enzymes and their coordinated action have been identified as components of the mechanism implicated in the alleviation of lipid peroxidation and the delay of senescence in many horticultural crops [15]. Loss of membrane integrity under senescence stress during storage of peach fruit is generally linked with the excessive accumulation of ROS, including $O_2^{\cdot}$ and H_2O_2, and the enhancement of lipoxygenase activity and malondialdehyde levels [15]. In the middle stage of fruit development, $O_2^{\cdot}$ and H_2O_2 act as potential signaling molecules. In the late stage of fruit ripening, H_2O_2 functions as a major toxic molecule to stimulate lipid peroxidation and oxidative stress [14]. Luo et al. [16] showed that browning in *Z. latifolia* was associated with a loss of membrane integrity, which resulted from lipid peroxidation. ROS metabolism, which is dependent on various functionally interrelated antioxidant enzymes, such as superoxide dismutase, catalase, and ascorbate peroxidase, is involved in lipid peroxidation; thus, Luo et al. concluded that chitosan/nano-chitosan coatings could effectively prevent the browning and lignification of cut *Z. latifolia* and maintain its fresh-cut color and texture for 12 days at 1°C. In general, lower doses of ROS act as signals that mediate at least a portion of the responses toward stress. At higher concentrations, they pose a significant threat that may eventually lead to programmed cell death [12]. Huang et al. [14] found that peaks of $O_2^{\cdot}$ and H_2O_2 occurred in the middle stage of peach fruit development, and that these peaks were closely followed by the rapid development of peach fruit. The production of ROS implies that they might serve as signaling molecules, stimulating the rapid development of fruit. However, in the late stage, H_2O_2 might be a major toxic molecule, which can induce oxidative stress. While ROS have the potential to cause oxidative damage to cells during environmental stress, they also play a key role in plants as signal transduction molecules involved in mediating responses to pathogen infection, environmental stresses, programmed cell death (accelerated by adenosine triphosphate depletion and altered redox states in cells [17]), and various developmental stimuli. These diverse stresses activate similar cell signaling pathways and cellular responses [2].

The *MAPK*-cascade signaling and senescence

MAPK cascades are evolutionarily conserved from unicellular to complex eukaryotic organisms, and constitute major signaling pathways involved in the regulation of a wide range of cellular activities from growth and development to cell death [18]. In the plant kingdom, *MAPK* cascades are a well-studied signaling mechanism, which includes a class of proteins that can sense stress signals and transduce them into suitable responses. These responses hold the key to the adaptation and survival of plants. A *MAPK* cascade is a divergent combination of at least three protein kinases, such as *MAPKKK* (*MKKK/MEKK, MAP3K*), *MAPKK* (*MEK/MKK*), and *MAPK* (*MPK*), which stimulate each other in a consecutive approach through phosphorylation [19]. The available data show that *MAPKs* are involved in plant signal transduction in response

to pathogens, drought, salinity, cold, wounding, ozone, ROS, and hormonal stimuli [19]. Multiple studies have demonstrated clear roles for this cascade in growth and development, as well as in defense/stress responses, in plants [18]. Some studies have been conducted to determine the response and regulation of rice *MAPKs*, particularly for their possible role/function in rice self-defense pathways [18]. In rice, 16 rice *MAPKs* (*OsMPKs*) have been identified; these are located on chromosomes 1, 2, 3, 5, 6, 10, and 11, with a particularly notable distribution on chromosomes 1, 5, and 6 [20]. Pedley and Martin [3] concluded that *MAPK* cascades participate in plant immunity. In addition, 28 *MPKs* and eight *MKKs* were identified in *Helianthus annuus* [4]. Pedley and Martin [21] identified two phylogenetically unrelated *MAPK* kinases (LeMKK2 and LeMKK4) in tomato that, when overexpressed in leaves, elicited cell death and activated Le*MPK2* and Le*MPK3*. In *Arabidopsis*, there are 20 *MAPKs*, 10 *MAPKs*, and approximately 60 putative *MAPKKKs*. Among these 60 putative *MAPKKKs*, 12 belong to the MEKK subgroup; the remaining putative *MAPKKKs* belong to the constitutive triple response 1 (CTR1) subgroup. Biochemical and genetic evidence has demonstrated that at least some of the 12 MEKK members are bona fide *MAPKKKs* [22]. In strawberry (*Fragaria vesca*), there are 12 *MAPK* genes in the genome. Protein domain analysis has shown that all FvMAPKs have typical protein kinase domains. Sequence alignments and phylogenetic analyses classified the Fv*MAPK* genes into four different groups [23]. In the apple (*Malus domestica*) genome, 28 putative apple *MAPK* genes (Md*MPKs*) and nine putative apple *MAPKK* genes (MdMKKs) have been identified [24,25]. Phylogenetic analysis revealed that Md*MAPKs* and MdMAPKKs can respectively be divided into four subfamilies (groups A, B, C, and D), respectively. The predicted MdMAPKs and MdMAPKKs were distributed across 13 of 17 chromosomes, with different densities. In addition, analyses of exon–intron junctions and of the intron phase inside the predicted coding region of each candidate gene revealed high levels of conservation within and between phylogenetic groups. These analyses demonstrated the existence of 14 *MAPKs*, five *MAPKKs*, 62 *MAPKKKs*, and seven *MAPKKKKs* in *Vitis vinifera* [26] and cucumber (*Cucumis sativus* L.). In *Cucurbitaceae* crops, which are highly susceptible to environmental stress and pathogen attack, analyses demonstrated the presence of at least 14 *MAPKs*, six *MAPKKs*, and 59 *MAPKKKs* [6]. The numbers of *MAPK* gene families in various plants are summarized in Table 1 [23-25].

Networking of ROS signaling with *mapk* signaling pathways

As mentioned above, reactive oxygen species (ROS) have a dual effect on plant metabolism: they can act as damaging byproducts of general metabolism; conversely, they can serve as signaling molecules [27]. These signaling networks include protein kinase networks, calcium signaling, cellular metabolic networks, and redox responses. In some instances, ROS accumulation has been found to precede the activation of signaling through these networks; in other instances, ROS accumulation was found to be a direct result of signaling through these networks [5]. In eukaryotes, the transmission of oxidative signals is controlled by protein phosphorylation involving *MAPKs* [13]. Therefore, a notable example of a ROS-activated signaling network is the *MAPK* cascade. An important *MAPKKK* in *Arabidopsis*, MEKK1, is activated upon exposure to abiotic factors (e.g., salt, cold, and wounds) and drought

Table 1: Numbers of *MAPK* gene families in assorted plants.

No.	Name of plant species	Abbreviation of *MAPK*	Total number of *MAPK* genes
1	*Aquilegia coerulea*	AcMPK	10
2	*Arabidopsis thaliana*	AtMPK	20
3	*Brachipodium distachyon*	BdMPK	16
4	*Brassica rapa*	BrMPK	30
5	*Capsella rubella*	CrMPK	18
6	*Carica papaya*	CpMPK	9
7	*Chlamydomonas reinhardtii*	CreinMPK Algae	6
8	*Citrus clememtina*	CcMPK	12
9	*Citrus sinensis*	CsMPK	12
10	*Coccomyxa subellipsoidea*	CsubMPK Algae	4
11	*Cucumis sativus*	CsMPK	14
12	*Eucalyptus grandis*	EgMPK	13
13	*Fragaria vesca*	FvMPK	11
14	*Glycine max*	GmMPK	31
15	*Gossipium Raimondi*	GrMPK	28
16	*Linum usitatissimum*	LuMPK	24
17	*Malus domestica*	MdMPK	28
18	*Manihot esculenta*	MeMPK	17
19	*Medicago truncatula*	MtMPK	17
20	*Micromonas pusila*	MpMPK	4
21	*Mimulus guttatus*	MgMPK	6
22	*Oryza sativa*	OsMPK	17
23	*Ostreococcus lucimarinus*	OlMPK	3
24	*Panicum virgatum*	PvMPK	27
25	*Phaseolus vulgaris*	PvMPK	14
26	*Physcomitrella patens*	PpMPK	8
27	*Picea abies*	PaMPK	14
28	*Populus trichocarpa*	PtMPK	21
29	*Prunus persica*	PperMPK	12
30	*Ricinus communis*	RcMPK	12
31.	*Selaginella moellendor ffii*	SmMPK	6
32	*Setaria italica*	SiMPK	16
33	*Solanum lycopersicum*	SlMPK	17
34	*Solanum tuberosum*	StMPK	12
35	*Sorghum bicolor*	SbMPK	16
36	*Thellungiella halophila*	ThMPK	17
37	*Theobroma cacao*	TcMPK	12
38	*Vitis venifera*	VvMPK	16
39	*Volvox car teri*	VcMPK	16
40	*Zea mays*	ZmMPK	12
41	*Zizania latifolia*	ZlMPK	18

and biotic factors (e.g., bacterial and fungal elicitors). In response to abiotic stimuli, MEKK1 activates the MKK2-*MPK*4/6 module; in response to biotic stress, it activates the MKK4/5-*MPK*3/6-VIP1/ACS6 module [28] (Figure 2). The *Arabidopsis MAPK* 8 (*MPK*8) reportedly connects protein phosphorylation, Ca^{2+}, and ROS in wound signaling pathways [13].

DISCUSSION

Under environmental stress conditions, *MPK*3 and *MPK*6 have been shown to play key roles in ROS signaling. Expression of

the genes encoding these proteins is induced by ozone treatment; plants that lack *MPK*3 or *MPK*6 are hypersensitive to ozone. Stress-induced expression of *MPK*3 and *MPK*6 leads to ROS accumulation and ethylene biosynthesis, thus inducing early leaf senescence [27,28]. Notably, *MAPK*s can be activated by the accumulation of H_2O_2; they can also trigger an H_2O_2-induced oxidative burst. In *Nicotiana benthamiana*, the *MAPK* cascades MEK2-SIPK/NTF4 and MEK1-NTF6 were reported to participate in the regulation of the radical burst induced by the oomycete pathogen *Phytophthora infestans* through nitric oxide and RbohB-dependent ROS generation. An experiment using *Arabidopsis*

protoplasts showed a correlation between the activation of plant *MAPK* cascades and H_2O_2, which was generated by various stress factors. H_2O_2 was found to activate the *MAPKKK*, ANP1, which then phosphorylated two downstream kinases, At*MPK3* and At*MPK6*. Protein phosphorylation through *MAPK* cascades has been suggested to trigger positive feedback regulation of Ca^{2+} and ROS through the activation of RbohD and RbohF in *Arabidopsis* [13]. Hydrogen peroxide is known to activate *MPK3* and *MPK6* in rice and can be activated by an upstream kinase, MKK6. This cascade is involved in providing resistance to fungal pathogens and tolerance against abiotic stresses (e.g., heavy metals, salt, cold, and UV rays) [28]. There is a feedback mechanism by which *MAPK* cascades, activated by ROS associated with particular stimuli, are known to regulate ROS production. Some studies have suggested that *MAPK* cascades exert positive feedback on ROS production. A study of maize revealed that abscisic acid activates 46 kDa *MAPK*, which acts downstream of H_2O_2 and positively regulates RBOH for H_2O_2 production. In addition to the positive regulation of ROS production, the NDPK2-*MPK3/6* cascade is known to negatively regulate ROS production, providing further tolerance against cold, salt, and oxidative stress [28]. These examples of ROS-mediated crosstalk among *MAPK*s suggest that ROS produced in response to various environmental stresses mediate the activation of similar *MAPK*s; however, the interactions within *MAPK* pathways and the final responses to these stresses become fundamentally distinct. The differences originate from the abilities of *MAPK*s to interact with separate downstream targets. Scaffolding proteins also play a major role in this interaction. It is also apparent that ROS have important roles as messengers that encode information for activation of different responses [28].

Possible signaling pathways of post-harvest senescence of horticultural products

Plants have evolved exceptional resistance mechanisms to manage many biotic and abiotic stresses. ROS are common messengers produced in response to abiotic and biotic stresses that activate *MAPK* pathways. On the basis of the available information, ROS can clearly activate a similar *MAPK* cascade in different stresses and can exert different responses accordingly. Understanding these pathways in horticultural products is very important for improving shelf life and quality retention during the harvest and post-harvest handling of horticultural products. The possible signaling pathways of the post-harvest senescence of horticultural products are shown in Figure 3. AC, adenylyl cyclase; AIF, apoptosis inducing factor; AKT, protein kinase B; AOC, allene oxide cyclase; AOS, allene oxide synthase; Apaf-1, apoptotic protease activating factor-1; Arts, apoptotic protease activating factor-1; ATF-2, activating
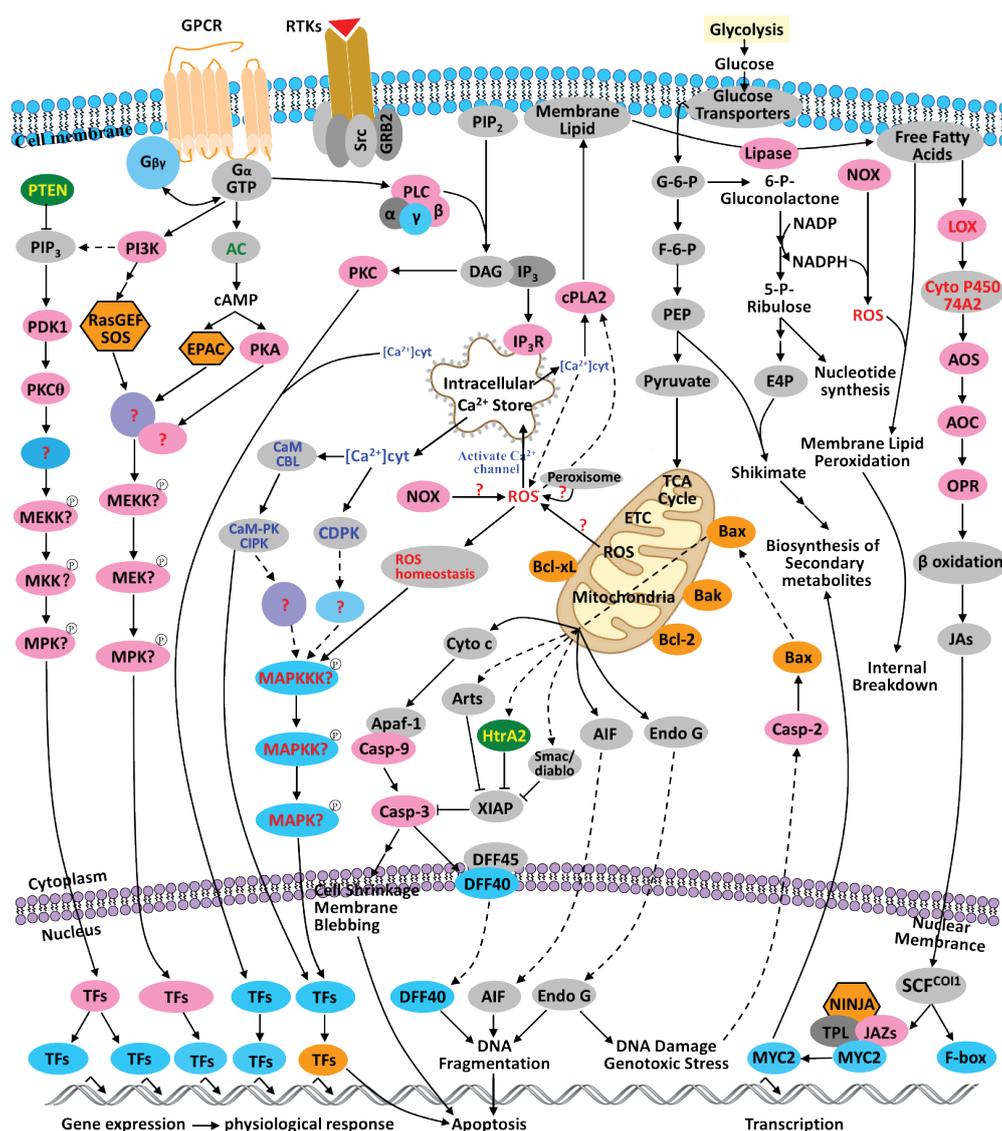

Figure 3: Possible signaling pathways involved in postharvest senescence of horticultural products.

transcription factor-2; Bak, Bax, Bcl-2, apoptosis-promoting gene; bHLH, basic Helix-Loop-Helix; CaM, calmodulin; CaM-PK, calcium/calmodulin-dependent protein kinase; cAMP, cyclic adenosine monophosphate; Casp-2/-3/-9, caspase-2/-3/-9; CDPK, calcium-dependent protein kinases; COI1, coronatine insensitive 1; cPLA2, cytosolic phospholipase A2; C-Raf, downstream signal after RAS signal activation; Cyto c, cytochrome c; Cyto P450 74A2, cytochrome P450 74A2; DAG, diacyl glycerol; DFF, DNA fragment factor; Endo G, endonuclease G; EPAC, exchange protein directly activated by cAMP; ETC, electron transport chain; F-6-P, fructose 6-phosphate; F-box, F-box protein; FLIP, Fas-associated death-domain-like IL-1 β-converting enzyme inhibitory protein; G-6-P, glucose-6-phosphate G; GR, glutathione reductase; GRB2, growth factor receptor-bound protein 2; Gαβγ, small g-protein HtrA2, HtrA, HtrA serine peptidase 2; IP3, inositol 1,4,5-triphosphate; Jas, jasmonic acid; JAZs, jasmonate ZIM-domain; *MAPKKK*, MEKK, *MAPK* kinase kinase; *MAPKK*, MKK, MEK, *MAPK* kinase; *MAPK*, *MPK*, *MAPK*; MYC2, transcription factor; NINJA, Jaz coupling factor; NOS, nitric oxide synthase; OPR, 12-oxo-phytodienoic acid reductase; PDK1, phosphoinositide dependent kinase-1; PEP, phosphoenolpyruvate; PTEN, phosphatase and tensin homology deleted on chromosome 10; PI3K, phosphatidylinositol 3-kinases; PIP2, PtdIns(4,5)P2; PIP3, PtdIns(3,4,5)P3; PKA, protein kinase A; PKC, protein kinase C; PKC⍰, protein kinase C ⍰; PLC, phospholipase C; Rap1, Ras, small Rap1/Ras-related GTP-binding protein; RasGEF SOS, rat sarcoma guanine nucleotide exchange factor (GEF) son of sevenless; SCF, SKP1/CULLIN/F-box; Smac/Diablo, second mitochondria-derived activator of caspases; TCA, tricarboxylic acid cycle; TFs, transcription factors; XIAP, X-linked inhibitor of apoptosis protein.

CONCLUSION

In summary, many studies of the post-harvest senescence mechanisms of horticultural products have been conducted; however, the relevant senescence signals and their transduction mechanisms have not yet been fully clarified. Potential signal transduction pathways must be verified by further experiments. We presume that further in-depth studies will reveal the post-harvest senescence mechanisms of horticultural products, which could provide a theoretical basis for exploring better storage and transportation techniques for post-harvest horticultural products.

REFERENCES

1. Aghdam MS, Jannatizadeh A, Luo Z, Paliyath G. Ensuring sufficient intracellular ATP supplying and friendly extra cellular ATP signaling attenuates stresses, delays senescence and maintains quality in horticultural crops during postharvest life. Trends Food Sci Technol. 2018;76:67-81.

2. Madani B, Mirshekari A, Imahori Y. Physiological responses to stress. Annu Rev Nurs Res. 2019;7:51-69.

3. Pedley KF, Martin GB. Role of mitogen-activated protein kinases in plant immunity. Curr Opin Plant Biol. 2005;8(5):541-547.

4. Neupane S, Schweitzer SE, Neupane A, Andersen EJ, Fennell A, Zhou R, et al. Identification and characterization of mitogen-activated protein kinase (MAPK) genes in sunflower (*Helianthus annuus* L.). Plants. 2019;8(2):1-25.

5. Mittler R, Vanderauwera S, Suzuki N, Miller G, Tognetti VB, Vandepoele K, et al. ROS signaling: the new wave? Trends Plant Sci. 2011;16(6):300-309.

6. Wang J, Pan C, Wang Y, Ye L, Wu J, Chen L, et al. Genome-wide identification of MAPK, MAPKK, and MAPKKK gene families and transcriptional profiling analysis during development and stress response in cucumber. BMC Genomics; 2015;16(1):386.

7. Gundewadi G, Rakesh V, Bhimappa B. Physiological and biochemical basis of fruit development and ripening: a review. J Hill Agr. 2018;9(1):7-21.

8. Saltveit ME. Postharvest physiology and biochemistry of fruits and vegetables, Chapter 4-Respiratory Metabolism. Woodhead Publishing. 2019;73-91.

9. Luo H, Zhou T, Kong X, Tao M, Zhang J, Wang W, et al. iTRAQ-based mitochondrial proteome analysis of the molecular mechanisms underlying postharvest senescence of *Zizania latifolia*. J Food Biochem. 2019;43(12):e13053.

10. Cavaiuolo M, Cocetta G, Spadafora ND, Mu CT, Rogers HJ, et al. Gene expression analysis of rocket salad under pre-harvest and postharvest stresses: a transcriptomic resource for *Diplotaxis tenuifolia*. Plos One. 2017;12(5):1-27.

11. Lin Y, Lin Y, Lin H, Ritenour MA, Shi J, Zhang S, et al. Hydrogen peroxide-induced pericarp browning of harvested longan fruit in association with energy metabolism. Food Chem. 2017;225:31-36.

12. Petrov V, Hille J, Mueller-Roeber B, Gechev TS. ROS-mediated abiotic stress-induced programmed cell death in plants. Front Plant Sci. 2015;6:1-16.

13. Sewelam N, Kazan K, Schenk PM. Global plant stress signaling: Reactive oxygen species at the cross-add. Front Plant Sci. 2016;7:187.

14. Huan C, Jiang L, An X, Yu M, Xu Y, Ma R, et al. Potential role of reactive oxygen species and antioxidant genes in the regulation of peach fruit development and ripening. Plant Phys Biochem. 2016;104:294-303.

15. Gao H, Ke Z, Kang H, Cheng N, Yang Y, Wang DN, et al. Melatonin treatment delays postharvest senescence and regulates reactive oxygen species metabolism in peach fruit. Postharvest Bio Tech. 2016;118:103-110.

16. Luo H, Jiang LI, Bao Y, Wang L, Yu Z. Effect of chitosan/nano-chitosan composite coating on browning and lignification of fresh-cut *Zizania latifolia*. J Food Quality. 2013;36(6):426-431.

17. Takahashi H, Matsumura H, Kawai-Yamada M, Uchimiya H. The cell death factor, cell wall elicitor of rice blast fungus (*Magnaporthe grisea*) causes metabolic alterations including GABA shunt in rice cultured cells. Plant Sign Beha. 2008;3(11):945-953.

18. Agrawal GK, Iwahashi H, Rakwal R. Rice MAPKs. Biochem Bioph Res Co. 2003;302(2):171-180.

19. Colcombet J, Hirt H. *Arabidopsis* MAPKs: A complex signaling network involved in multiple biological processes. Biochem J. 2008;413(2):217-226.

20. Liu Q, Xue Q. Computational identification and phylogenetic analysis of the MAPK gene family in *Oryza sativa*. Plant Phys Biochem. 2007;45(1):6-14.

21. Pedley KF, Martin GB. Identification of MAPKs and their possible MAPK kinase activators involved in the Pto-mediated defense response of tomato. J Biol Chem. 2004;279(47):49229-49235.

22. Xu J, Zhang S. Mitogen-activated protein kinase cascades in signaling plant growth and development. Trends Plant Sci. 2014;20(1):56-64.

23. Wei W, Chai Z, Xie Y, Gao K, Cui M, Jiang Y, et al. Bioinformatics identification and transcript profile analysis of the mitogen-activated protein kinase gene family in the diploid woodland strawberry *Fragaria vesca*. Plos One. 2017;12(5): e0178596.

24. Zhang S, Xu R, Luo X, Jiang Z, Shu H. Genome-wide identification and expression analysis of MAPK and MAPKK gene family in *Malus domestica*. Gene. 2013;531(2):377-387.

25. Mohanta TK, Arora PK, Mohanta N, Parida P, Bae H. Identification of new members of the MAPK gene family in plants shows diverse conserved domains and novel activation loop variants. BioMed Genomics. 2015;16(1):1–20.

26. Çakır B, Kılıçkaya O. Mitogen-activated protein kinase cascades in *Vitis vinifera*. Front Plant Sci. 2015;6(556).

27. Kanojia A, Dijkwel PP. Abiotic stress responses are governed by reactive oxygen species and age. Annu Plant Rev. 2018;1:1-32.

28. Jalmi SK, Sinha AK. ROS mediated MAPK signaling in abiotic and biotic stress—striking similarities and differences. Front Plant Sci. 2015;6:769.