RESEARCH ARTICLE

SABRAO Journal of Breeding and Genetics
58 (1) 33-40, 2026
http://doi.org/10.54910/sabrao2026.58.1.4
http://sabraojournal.org/
pISSN 1029-7073; eISSN 2224-8978

MOLECULAR ANALYSIS OF PEPPER CULTIVARS FOR MATURITY STAGES USING EXPRESSION PROFILING OF *ACS* AND *ACO* GENES IN SAUDI ARABIA

RAHMA ALSHAMRANI

Department of Biological Sciences, King Abdulaziz University, Jeddah, Saudi Arabia
Author's email: ralshamrani@kau.edu.sa

SUMMARY

Pepper (*Capsicum annuum* L.) is a vital food commodity, and its shelf life plays a crucial role in determining economic viability. Ethylene, a key regulator in the ripening of climacteric fruits, such as peppers, considerably affects postharvest longevity. The ethylene biosynthesis pathway incurs management from gene families encoding 1-aminocyclopropane-1-carboxylate synthase (*ACS*) and 1-aminocyclopropane-1-carboxylate oxidase (*ACO*). The following study investigated the expression patterns of the *ACS* gene (*CaACS6*) and the *ACO* gene (*CaACO1*), along with a housekeeping gene (*CaGAPDH*), in three pepper cultivars (Cherry, Anaheim, and Bohemian), using quantitative polymerase chain reaction (qPCR). Gene expression assessment progressed across three maturity developmental stages (young green, large green, and ripening) to elucidate their influence on ethylene synthesis and pepper shelf life. The expression of the *CaACS6* gene showed a positive correlation with the ripening stage across all cultivars. In the Cherry cultivar, the expression of gene *CaACO1* significantly decreased at the ripening stage, indicating reduced ethylene production and potential for extended shelf life. Similarly, the Anaheim cultivar exhibited a decrease in the expression of both genes (*CaACS6* and *CaACO1*) during the ripening stage, revealing a reduced ethylene production, suggesting a potential for reduced ethylene production and extended shelf life. These insights provide valuable guidance for selecting pepper cultivars with prolonged shelf life, thereby enhancing postharvest quality and commercial sustainability.

Keywords: Pepper (*C. annuum* L.), cultivars, *CaACS6* and *CaACO1*, gene expression, ethylene production regulatory genes, maturity stages

Key findings: In pepper (*C. annuum* L.) cultivars, the gene *CaACS6* expression with an increased ethylene biosynthesis at the ripening stage, confirmed its role in maturation. Reduced gene *CaACO1* expression in pepper cultivars (Cherry and Anaheim) at the ripening stage suggested lower ethylene production, supporting their potential for extended shelf life.

Communicating Editor: Dr. Sajjad Hussain Qureshi

Manuscript received: July 27, 2025; Accepted: November 20, 2025.

Citation: Alshamrani R (2026). Molecular analysis of pepper cultivars for maturity stages using expression profiling of *ACS* and *ACO* genes in Saudi Arabia. *SABRAO J. Breed. Genet. 58 (1) 33-40. http://doi.org/10.54910/sabrao2026.58.1.4*.

INTRODUCTION

The fruit ripening process involves an intricate regulation and delineation into climacteric and non-climacteric fruits by ripening behaviors. Climacteric fruits, such as bananas, tomatoes, and peppers (*Capsicum annuum* L.), undergo pronounced surges in ethylene production and respiration, distinguishing them from non-climacteric fruits (Aizat *et al*., 2013). In the context of ethylene biosynthesis, understanding the rate-limiting steps and the regulatory genes involved in ethylene biosynthesis is paramount. Ethylene, a pivotal phytohormone, governs a myriad of physiological variations associated with fruit ripening phases, including alterations in color, texture, flavor, and aroma (Prasanna *et al*., 2007).

In climacteric fruits, such as tomatoes and peppers, ethylene production precedes color development and other ripening manifestations, showing the considerable regulation of ethylene biosynthesis (Klee and Giovannoni *et al*., 2011; Lü *et al*., 2018). In the papaya plant, these two genes have also undergone studies, cloning, and uses to develop papaya cultivars with delayed ripening characteristics (Magdalita *et al*., 200***2***, 2013). This increase in ethylene, as intertwined with climacteric respiration and biochemical transformations, leads to variations in pigment composition and cell wall structure (Afshar-Mohammadian *et al*., 2019; Abd-El-Moneim *et al*., 2020). Therefore, the role of ethylene in climacteric fruit ripening is well established; however, its role in non-climacteric fruit ripening remains less explored (Giovannoni, 2004; Pech *et al*., 2012).

Moreover, semi-climacteric fruits like peppers exhibited intermediate ripening patterns, where ethylene production was less distinct compared with climacteric fruits; however, it still influenced the ripening-associated variations (McGlasson *et al*., 1978). This complex regulation of ethylene synthesis in semi-climacteric fruits underscores the need for comprehensive studies to explore the underlying molecular mechanisms governing their ripening dynamics. Therefore, this study delved into the expression patterns of the crucial genes involved in ethylene biosynthesis, particularly focusing on peppers and shedding light on the complex interaction between gene regulation and fruit ripening processes.

In fruits, ethylene biosynthesis involves two key enzymes: ACS (1-aminocyclopropane-1-carboxylate synthase) and ACO (1-aminocyclopropane-1-carboxylate oxidase). The ACS catalyzes the conversion of S-adenosyl-L-methionine (SAM) to the ethylene precursor 1-aminocyclopropane-1-carboxylic acid (ACC) and often acts as the rate-limiting step (Lin *et al*., 2009). Subsequently, the ACO converts ACC to ethylene (Yang and Hoffman, 1984; Bleecker and Kende, 2000). However, both climacteric and non-climacteric fruits produce ethylene at the basal levels, referred to as System-1; climacteric fruits undergo a transition to System-2 upon maturation, increasing ethylene production that initiates ripening (Lelièvre *et al*., 1997; Pech *et al*., 2012).

Ethylene acts as a pivotal regulator of ripening, facilitating the transcription and translation of enzymes involved in these processes, such as cell wall degradation, pigment synthesis, and flavor development (Bapat *et al*., 2010). In peppers, a significant increase in expression of *ACS*, *ACC*, and *ACO* genes was evident during ripening, underscoring the importance of ethylene in maturity (Yokotani *et al*., 2009; Sharma *et al*., 2021). Moreover, synthetic compounds targeting ACS and ACO activities have reached exploration to mitigate postharvest losses (Ghosh *et al*., 2020). In ripening-related genes, including *PG*, *Psy*, and *ACO1*, the *ACS6* plays a remarkable role in ethylene biosynthesis and cell wall metabolism, influencing fruit texture and shelf life (Barry *et al*., 2000; Aizat *et al*., 2013).

Extending the fruit's shelf life is a key breeding objective, and modulating fruit ripening processes through transcriptional regulation of ripening-related genes holds promise in achieving this goal (Causier *et al*., 2002). Understanding the intricate interplay between ethylene biosynthesis and ripening-associated gene expression is essential for developing strategies to enhance fruit quality and prolong its shelf life, thereby contributing to the economic viability of fresh produce.

The genus *Capsicum* encompasses various domesticated pepper cultivars, alongside approximately 32 wild relatives (Magdy *et al*., 2019). These wild pepper phenotypes have attained adaptation to diverse environments, and the study focus primarily lies on local cultivars, which are more crucial from a breeding viewpoint, as they aim to enhance pepper quality and shelf life. In genus *Capsicum*, nine species identification was successful with varying ripening behaviors, and some displayed climacteric ethylene production as cultivated tomatoes (Grumet *et al*., 1981). Understanding the dynamics of ethylene biosynthesis during pepper fruit development is pivotal, as it considerably affects the ripening and shelf-life phases. Therefore, the presented study aimed to assess the ripening characteristics and shelf life of newly introduced greenhouse pepper cultivars in Saudi Arabia, leveraging the quantitative polymerase chain reaction (qPCR) analysis. Additionally, elucidating the roles of *ACO* and *ACS* genes in ethylene production provides valuable insights to support breeding efforts focused on enhancing pepper shelf life.

MATERIALS AND METHODS

Plant materials and growth conditions

The concerned experiment utilized three commercial pepper (*C. annuum* L.) cultivars (Cherry, Anaheim, and Bohemian). Planting the seeds proceeded in a non-heated, rain-sheltered greenhouse from April 2021 to January 2022, following a randomized scheme at the Department of Botany, Faculty of Agriculture, Ain Shams University, Cairo, Egypt. Plant fertilization continued weekly using NPK fertilizer (Polyhalite fertilizer, UK). Transplanting occurred when the seedlings were 50 days old. Plants entailed vertical training with a single stem and topping by cutting at the second leaf above the ninth truss. Each truss bore four or five pepper fruits. For this experiment, pepper fruit harvesting transpired at three different stages to monitor the expression patterns of *ACS* and *ACO* genes, irrespective of their commercial maturity: young green (10 days after pollination, or DAP), large green (30 DAP), and large red/ripe (60 DAP). The identification of ripening stages and their categorization followed the USDA color index (USDA, 2005; López-Camelo and Gómez, 2004), with the examples for the cultivars Cherry and Anaheim depicted in Figure 1. The sampling stages according to the expected ethylene pattern reached their designs based on previous studies (Yokotani *et al*., 2009).

RNA extraction and cDNA synthesis

The experiment followed the method outlined by Kim *et al*. (2006) with slight modifications. Each sample comprised three biological replications. Total RNA extraction occurred from fresh fruit samples using Triazole® reagent (Sigma-Aldrich, St. Louis, MO, USA), followed by purification via phenol-chloroform extraction. RNA quality assessment via 2% agarose gel electrophoresis used 2 µg RNA per lane (FUJIFILM), while the measurement of RNA concentrations utilized a Quantus™ Fluorometer (Promega, USA). The isolated total RNA underwent treatment with RNase-free DNase I (Promega, Madison, WI, USA) for 45 min at 37 °C. For each stage (10, 30, and 60 DAP), the usage of purified total RNA amounts of 1.5 and 2 µg succeeded. The conduct of first-strand cDNA synthesis applied M-MLV ^{RT}-reverse transcriptase (Invitrogen, Carlsbad, CA, USA) and random hexamers following the kit instructions. Upon completion, the cDNA storage at -20 °C ensued until further quantification.

Quantitative real-time PCR (RT-PCR)

Real-time quantitative PCR commenced using a 20 µl master mix, comprising 10 µl of SYBR green (Thermo Fisher Scientific's SYBR Green Master Mix), 0.5 µl of primer (P1), and 0.5 µl of primer (P2). They are newly designed primers using NCBI-Primer-BLAST (Table 1),

Table 1. Oligonucleotide primers used in the RT-PCR of ethylene-related genes in pepper cultivars.

Gene product	Gene name	Primer name	sequence (5′→3′)
ACS	*CaACS6*	*CaACS6F*	ATGCTGCCACGGTCTTTGAT
		CaACS6R	AGCCTTTCGGAACTCTCAGC
ACO	*CaACO1*	*CaACO1F*	GCATTTTCACGGAGCAAGGG
		CaACO1R	TCCCGTCCTTTTCAGCCATC
GAPDH	*CaGAPDH*	*CaGAPDHF*	GCTCGTCTGAATGGAGGAGG
		CaGAPDHR	GGTGGGTTAAAGTAGGGGCC

Figure 1. Illustration of fruit-ripening stages of pepper (*C. annuum*) cultivars: Cherry, Anaheim, and Bohemian at 60 DAP (mature stage).

along with 1.2 µl of buffer, 6.6 µl of H_2O, and 1.2 µl of cDNA template, in addition to the TaKaRa rTaq polymerase (TaKaRa, Kyoto, Japan) kit. The qPCR program included an initial denaturation at 92 °C for 2 min, followed by 40 cycles of denaturation at 92 °C for 5 s, annealing at 56 °C for 15 s, and extension at 72 °C for 26 s. A dissociation test continued from 95 °C to 50 °C at 10-min intervals to determine the dimerization. Employing glyceraldehyde-3-phosphate dehydrogenase (*GAPDH*) served as a housekeeping gene (Coker and Davies, 2003).

Statistical analysis

In accordance with the method proposed by Livak and Schmittgen (2001), the cycle threshold (CT)'s computation used the default settings of sequence detection results obtained from the real-time software. Determining the number of cDNA molecules utilized the equation provided on the graph per microgram of mRNA-converted cDNA. The Ct values, as employed, estimated gene expression through relative quantification. Using the fold change method, the target genes attained a comparison with the control. The performance of a one-way analysis of variance (ANOVA) used SAS v8.2, based on a general linear model, which was followed by Duncan's multiple range test (DMRT) for means comparison and separation.

RESULTS AND DISCUSSION

The qPCR assay and validation

Real-time quantitative polymerase chain reaction (qPCR) was a widely used method for gene quantification due to its broad dynamic range, high sensitivity, and specificity (Mesfer *et al*., 2022). The qPCR has achieved effective application to both climacteric and non-climacteric fruits to detect ripening-related transcriptional changes with high accuracy, making it suitable for assessing ethylene-related genes in peppers (*C. annuum* L.).

Table 2. The PCR cycle threshold obtained by RT-PCR for genes *GAPDH*, *CaACS6*, and *CaACO1* at young green (10 DAP), large green (30 DAP), and large red/rip (60 DAP) stages of the three pepper cultivars.

Pepper line ID	*GAPDH*	*CaACS6*			*CaACO1*		
	HKG	10 DAP	30 DAP	60 DAP	10 DAP	30 DAP	60 DAP
Cherry	29	26	24	32	23	22	25
Anaheim	29	29	27	28	28	29	33
Bohemian	29	28	22	31	24	25	29

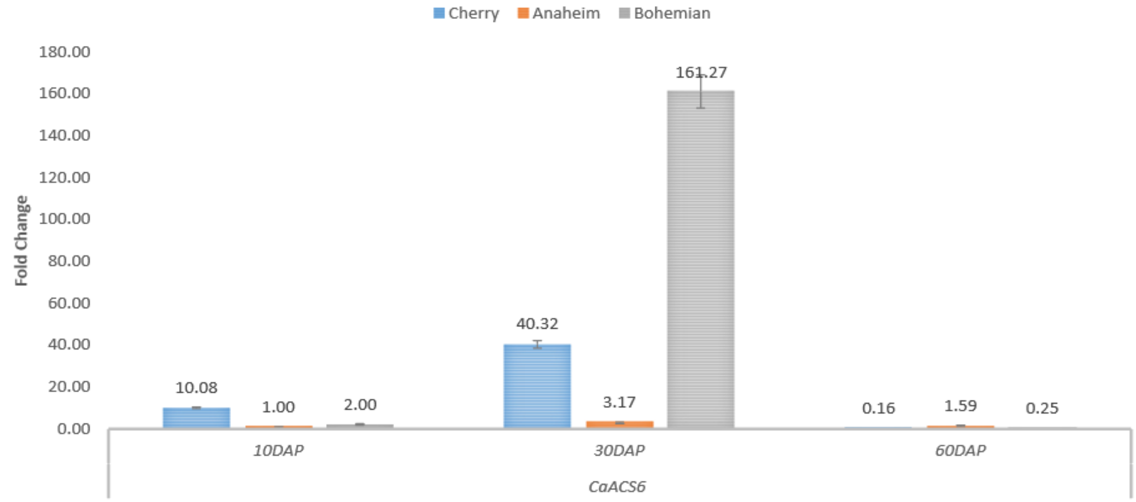

Figure 2. The fold-change ratios of relative expression for the ethylene synthase-associated gene *ACS* measured at three fruit-growth stages for the three imported pepper cultivars.

The promising study examined the *ACS* and *ACO* gene expressions at three ripening stages—young green (10 DAP), large green (30 DAP), and large red/ripe (60 DAP)—using *GAPDH* as the reference gene for normalization. The *GAPDH* stability in pepper and tomato has been confirmatory in developmental and stress studies, ensuring that observed variations in *ACS*/*ACO* gene expression reflect the biological variations rather than technical bias.

Melting curve analysis confirmed single, specific amplification peaks for genes *CaACS6* and *CaACO1*, indicating primer specificity. In the pepper cultivar Bohemian, the Ct values ranged from 22 (*CaACS6*) to 30 to 33 DAP (*CaACO1*) in FR-stage fruit (Table 2). Reports of similar Ct ranges for ethylene biosynthetic genes have also emerged in pepper during ripening phases (Lee *et al*., 2010), supporting the reliability and comparability of the present expression data.

Expression of the gene *CaACS6*

Assessing expression variations entailed calculating the delta Ct values by subtracting target gene Ct values from those of *GAPDH* (Livak and Schmittgen, 2001), allowing the estimation of relative expression and fold change. The highest *CaACS6* gene expression resulted in the pepper cultivar Bohemian at 30 DAP, with lower levels at the FR stage (Figure 2). Comparable qPCR-based fold variation patterns have been notable in tomato and melon fruits during the maturation process (Chen *et al*., 2022).

Across the pepper cultivars, *CaACS6* gene expression peaked at 30 DAP in the Bohemian and Cherry cultivars but remained low in the Anaheim cultivar. Similarly, *ACS* gene induction patterns have appeared in climacteric fruits, with the expression enhanced during active ripening (Tatsuki *et al*., 2007). This also authenticates the cultivar-specific regulation for ethylene biosynthesis.

The *CaACS6* gene encodes ACC synthase, which converts S-adenosylmethionine to ACC (Yang and Hoffman, 1984). Its downregulation at 60 DAP in the pepper cultivars Bohemian and Cherry coincides with ripening completion, whereas cultivar Anaheim's persistently obtained recordings with low expression, which may underlie its delayed ripening and extended shelf life (Bapat *et al*., 2010).

Expression of the gene *CaACO1*

The expression of the *CaACO1* gene reached primary detection at the young green (10 DAP) and large green (30 DAP) stages, with cultivar-specific differences (Figure 3). In the cultivar Cherry, *CaACO1* gene expression was high at 10 DAP (80.63) and peaked at 30 DAP (128.00), suggesting increased ethylene demand during early-to-mid ripening. In the cultivar Bohemian, the gene expression was relatively high at 10 DAP (32.00); however, it declined at 30 DAP (20.16), contrasting with the sharp *CaACS6* gene increase at this stage. This divergence signifies distinct regulatory control of the two genes in the pepper cultivar Bohemian.

In the cultivar Anaheim, the *CaACO1* gene expression remained low across all the fruit growth stages, with only minor fluctuations (2.00 at 10 DAP, 1.26 at 30 DAP, and 1.00 at 60 DAP, respectively). Such low levels indicate a reduced ethylene biosynthesis system, which may contribute to delaying the ripening and extending the shelf life. Similar tightly regulated *ACO1* gene patterns have succeeded their documentation in tomato, where expression may be negatively regulated in some cultivars (Nakatsuka *et al*., 1998).

Comparative expression trends of *CaACS6* and *CaACO1*

By considering them together, the *CaACS6* and *CaACO1* genes exhibited distinct yet complementary expression patterns across pepper cultivars. In the Cherry cultivar, both genes' high expression emerged at 30 DAP, although the gene *CaACO1* also showed a pronounced peak at 10 DAP, suggesting earlier

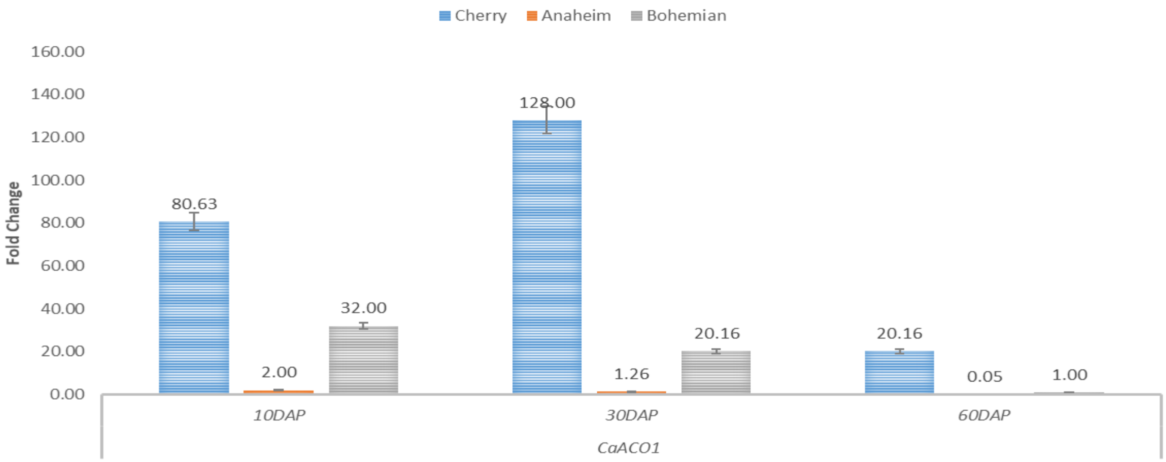

Figure 3. The fold-change ratios of relative expression for the ethylene synthase-associated gene *ACO1* measured at three fruit-growth stages for the three imported pepper cultivars.

activation of ethylene biosynthesis. In the cultivar Bohemian, the *CaACS6* gene displayed a dramatic rise at 30 DAP, whereas the gene *CaACO1* declined at the same stage. This indicates differential regulation of the two steps in ethylene biosynthesis, with the same findings also observed in tomato ripening studies (Nakatsuka *et al*., 1998; Wang *et al*., 2006). Meanwhile, in the cultivar Anaheim, both genes were consistently remarkable with low expression across all the fruit stages, supporting the cultivar's delayed ripening and extended shelf life. These results are consistent with findings that link reduced ACS and ACO activity to slower maturation and firmer fruit texture (Bapat *et al*., 2010). These combined patterns highlight that the *CaACS6* gene variation largely drives mid-ripening ethylene synthesis, while the *CaACO1* gene regulation may influence early- to mid-stage ethylene dynamics in a cultivar-dependent manner.

CONCLUSIONS

The presented study elucidates the genetic regulation of ethylene biosynthesis in peppers (*C. annuum* L.) and its role in fruit ripening, revealing significant cultivar-dependent differences. The low expression of *CaACS6* and *CaACO1* genes in the cultivar Anaheim correlates with the delayed ripening, reduced ethylene production, and extended shelf life. It identifies the said genotype as a promising breeding material and strong candidate for future studies on shelf life. These findings can help in marker-assisted selection and precision breeding to develop long-lasting cultivars adapted to Saudi Arabia's environmental conditions and market demands. Furthermore, biotechnological tools, such as clustered regularly interspaced palindromic repeats (CRISPR)-Cas9, RNA interference (RNAi), and postharvest ethylene inhibitors, offer complementary strategies for shelf-life extension. Integrating these genetic, molecular, and postharvest approaches holds substantial potential to enhance the sustainability and competitiveness of the regional pepper industry.

REFERENCES

Abd-El-Moneim D, Alqahtani MM, Abdein MA, Germoush MO (2020). Drought and salinity stress response in wheat: Physiological and TaNAC gene expression analysis in contrasting Egyptian wheat genotypes. *J. Plant Biotechnol*. 47: 1–14.

Afshar-Mohammadian M, Fallah SF, Rezadoost MH (2019). Different expression of kiwifruit ethylene-related genes during low storage temperatures. *J. Consum. Prot. Food Saf.* 14: 113–120.

Aizat WM, Able JA, Stangoulis JC, Able AJ (2013). Characterisation of ethylene pathway components in non-climacteric capsicum. *BMC Plant Biol.* 13: 191.

Bapat VA, Trivedi PK, Ghosh A, Sane VA, Ganapathi TR, Nath P (2010). Ripening of fleshy fruit: Molecular insight and the role of ethylene. *Biotechnol. Adv.* 28: 94–107.

Barry CS, Llop-Tous MI, Grierson D (2000). The regulation of 1-aminocyclopropane-1-carboxylic acid synthase gene expression during the transition from system-1 to system-2 ethylene synthesis in tomato. *Plant Physiol.* 123: 979–986.

Bleecker AB, Kende H (2000). Ethylene: A gaseous signal molecule in plants. *Annu. Rev. Cell Dev. Biol.* 16: 1–18.

Causier B, Kieffer M, Davies B (2002). MADS-box genes reach maturity. *Science* 296: 275–276.

Chen H, Bai S, Kusano M, Ezura H, Wang N (2022). Increased ACS enzyme dosage causes initiation of climacteric ethylene production in tomato. *Int. J. Mol. Sci.* 23: 10788.

Coker JS, Davies E (2003). Selection of candidate housekeeping controls in tomato plants using EST data. *Biotechniques* 35: 740–748.

Ghosh A, Saha I, Dolui D, De AK, Sarkar B, Adak MK (2020). Silver can induce oxidative stress in parallel to other chemical elicitors to modulate the ripening of chili cultivars. *Plants* 9: 238.

Giovannoni J (2004). Genetic regulation of fruit development and ripening. *Plant Cell* 16: S170–S180.

Grumet R, Fobes JF, Herner RC (1981). Ripening behavior of wild tomato species. *Plant Physiol.* 68: 1428–1432.

Kim HB, Kwon M, Ryu H, Fujioka S, Takatsuto S, Yoshida S, An CS, Lee I, Hwang I, Choe S (2006). The regulation of DWARF4 expression is likely a critical mechanism in maintaining the homeostasis of bioactive brassinosteroids in *Arabidopsis*. *Plant Physiol.* 140: 548–557.

Klee HJ, Giovannoni JJ (2011). Genetics and control of tomato fruit ripening and quality attributes. *Annu. Rev. Genet.* 45: 41–59.

Lelièvre J, Latchè A, Jones B, Bouzayen M, Pech J (1997). Ethylene and fruit ripening. *Physiol. Plant.* 101: 727–739.

Lin Z, Zhong S, Grierson D (2009). Recent advances in ethylene research. *J. Exp. Bot.* 60: 3311–3336.

Livak KJ, Schmittgen TD (2001). Analysis of relative gene expression data using real-time quantitative PCR and the 2−ΔΔCT method. *Methods* 25: 402–408.

López-Camelo AF, Gómez PA (2004). Comparison of color indexes for tomato ripening. *Hortic. Bras.* 22(3): 534–537.

Lü P, Yu S, Zhu N, Chen YR, Zhou B, Pan Y, Tzeng D, Fabi JP, Argyris J, Garcia-Mas J, Ye N, Zhang J, Grierson D, Xiang J, Fei Z, Giovannoni J, Zhong S (2018). Genome encode analyses reveal the basis of convergent evolution of fleshy fruit ripening. *Nat. Plants* 4: 784–791.

Magdalita PM, Laurena AC, Perez MTM (2013). Cloning and characterization of partial 1-aminocyclopropane-1-carboxylate oxidase gene and antisense transformation into yellow 'Solo' papaya via *Agrobacterium tumefaciens*. *J. Int. Soc. Southeast Asian Agric. Sci.* 19: 63–76.

Magdalita PM, Laurena AC, Yabut-Perez BM, Zaporteza MM, Mendoza EMT, Villegas VN, Botella JR (2002). Towards transformation, regeneration, and screening of papaya containing antisense ACC synthase gene. In: I.K. Vasil (Ed.), Plant Biotechnology 2002 and Beyond. Springer, Dordrecht, pp. 323–327.

Magdy M, Ou L, Yu H, Chen R, Zhou Y, Hassan H, Feng B, Taitano N, van der Knaap E, Zou X, Li F, Ouyang B (2019). Pan-plastome approach empowers the assessment of genetic variation in cultivated *Capsicum* species. *Hortic. Res.* 6: 108.

McGlasson WB, Wade NL, Adato I (1978). Phytohormones and fruit ripening. In: D.S. Letham, P.B. Goodwin, and T.V.J. Higgins (Eds.), Phytohormones and Related Compounds — A Comprehensive Treatise, Vol. 2. Elsevier, New York, pp. 447–493.

Mesfer AS, Safhi FA, Alshaya DS, Ibrahim AA, Mansour H, Abd El-Moneim D (2022). Genetic diversity using biochemical, physiological, karyological and molecular markers of *Sesamum indicum* L. *Front. Genet.* 13: 1035977.

Nakatsuka A, Murachi S, Okunishi H, Shiomi S, Nakano R, Kubo Y, Inaba A (1998). Differential expression and internal feedback regulation of 1-aminocyclopropane-1-carboxylate synthase, 1-aminocyclopropane-1-carboxylate oxidase, and ethylene receptor genes in tomato fruit during development and ripening. *Plant Physiol.* 118: 1295–1305.

Pech JC, Purgatto E, Bouzayen M, Latché A (2012). Ethylene and fruit ripening. In: M.T. McManus (Ed.), Annual Plant Reviews, Vol. 44: The Plant Hormone Ethylene. Wiley-Blackwell, Oxford, UK, pp. 275–304.

Prasanna V, Prabha TN, Tharanathan RN (2007). Fruit ripening phenomena – an overview. *Crit. Rev. Food Sci. Nutr*. 47: 1–19.

Sharma K, Gupta S, Sarma S, Rai M, Sreelakshmi Y, Sharma R (2021). Mutations in tomato 1-aminocyclopropane carboxylic acid synthase2 uncover its role in development beside fruit ripening. *Plant J.* 106: 95–112.

Tatsuki M, Endo A, Ohkawa H (2007). Influence of time from harvest to 1-MCP treatment on apple fruit quality and expression of genes for ethylene biosynthesis enzymes and ethylene receptors. *Postharvest Biol. Technol.* 43: 28–35.

USDA (2005). United States Standards for Grades of Sweet Peppers. USDA, Washington, DC, USA. https://www.ams.usda.gov/grades-standards/sweet-peppers-grades-and-standards

Wang W, Zhu B, Lü J, Luo Y (2006). No difference in the regulation pattern of calcium on ethylene biosynthesis between wild-type and never-ripe tomato fruit at mature green stage. *Russ. J. Plant Physiol.* 53: 54–61.

Yang SF, Hoffman NE (1984). Ethylene biosynthesis and its regulation in higher plants. *Annu. Rev. Plant Physiol.* 35: 155–189.

Yokotani N, Nakano R, Imanishi S, Nagata M, Inaba A, Kubo Y (2009). Ripening-associated ethylene biosynthesis in tomato fruit is autocatalytically and developmentally regulated. *J. Exp. Bot*. 60: 3433–3442.