**Proceedings of Pakistan Society for Horticultural Science**
2nd International Conference on Horticultural Sciences, February 18-20, 2016
Theme: Production Challenges and Food Security
Institute of Horticultural Sciences, University of Agriculture, Faisalabad, Punjab 38040, Pakistan

# Pre-Shipment Ethylene Treatment Influences Physiological, Biochemical and Phytochemical Attributes of Mango cv. 'Sindhri'

Saif ud din Taj*, Aman Ullah Malik, Muhammad Amin, Ahmad Sattar Khan, Saqib Shehzad
Postharvest Research and Training Centre, Institute of Horticultural Sciences, University of Agriculture, Faisalabad-38040, Pakistan
Email: maliksaif1990@gmail.com (S.T)

## Abstract

Physiologically mature 'Sindhri' mangoes were exposed to ethylene (100 ppm) at 24°C for 8 hours and subjected to stimulated sea shipment (12±1°C; 85-90% RH) and air freight (25±1°C and 33±2°C; 55-60% RH) conditions. Pre-storage ethylene treatment did not have any negative effect on fruit quality attributes. Peel colour, shrivelling, softness, acidity, ascorbic acid and anthracnose surged up in fruits of 25°C storage. However, ethylene, respiration rate and physiological weight loss increased in fruits of 33°C. Total antioxidants and total Phenolic contents were highest in fruits of 12°C. In this research concluded that sindhri mango fruit can be subjected to sea shipping conditions after ethylene treatment for 28 days with post storage shelf life of 4-5 days and in case of air freight, the pre-shipment ethylene treatment subsequently storage at 25±1°C (55-60% RH) for shelf life of 8 days and 33±2°C (55-60% RH) for 6 days can be an economical strategy.

## INTRODUCTION

Mango (*Mangifera indica* L.) is a preferred fruit worldwide due to its fantastic taste, unique flavour and premium nutritional value due to which it is known as "King of Fruits" (Asif *et al.*, 2002). Ripe mangoes contain bulk of carbohydrates, vitamins, amino acids, fatty acids, proteins, minerals, antioxidants and organic acids (Mukherjee and Litz, 2009). Eating mangoes in season may provide 250 KJ (60 Kcal)/100 g energy and ascorbic acid to our body (28.5 mg 100 $g^{-1}$) (Meadows, 1998).

Method and ripening agent play important role in fruit quality development and uniform ripening. Number of ripening strategies are used to trigger the ripening process. In some of the developing countries like Pakistan, Bangladesh and India calcium carbide ($CaC_2$) is used for ripening of fruits (Medlicott, 1986). The use of $CaC_2$ in commercial fruit ripening has been banned nationally and internationally from food safety and health prospective as it has been proven to be carcinogenic in nature and does not meet the current food safety standards (Medlicott *et al.*, 1987).

Different ripening (agents) alternate to $CaC_2$ are used including ethylene, ethephon and ethrel etc. The use of ethylene has now become a commercial feasible and globally

acceptable ripening strategy. Mature green mangoes are exposed to 20-100 ppm ethylene for 24-48 hours to trigger the ripening process in mangoes (Barmore, 1974). Mango being climacteric fruit, ripens after harvest (Prasanna *et al.*, 2007) and modification in texture and enzymatic activities linked with fruit ripening (Huber, 1983) in which breakdown of pectin, cellulose and hemicellulose cell wall polymers occurs (Payasi *et al.*, 2009). During ripening, physiological, biochemical and molecular changes occur that directly affect its quality traits (Osorio and Fernie, 2013). Some biochemical changes during postharvest ripening of mango include biosynthesis of carotenoids (Mercadante and Rodriguez-Amaya, 1998), increased activity of cell wall degrading enzymes (Ali *et al.*, 1995), changes in cell wall (Muda *et al.*, 1995), changes in phenolic content (Palafox-Carlos *et al.*, 2012), decline in ascorbic acid content (Hernández *et al.*, 2006), changes in color (Ornelas- Paz *et al.*, 2007), and increase in total soluble solids (Padda *et al.*, 2011). In mango, various enzymes documented to be involved in softening are polygalacturonase (Lazan *et al.*, 1986), and _-1,4-glucanases (Ali *et al.*, 2004).

Tissue softening is a physiological process particularly sensitive to ethylene (Gray *et al.*, 1994). Ethylene plays a very important role in regulation of fruit ripening (Lelievre *et al.*, 1997). After harvest, mango exhibits sharp rise in climacteric respiration rate and ethylene production which limits its postharvest shelf life with reduced eating quality (Tucker, 1993). Ripening reactions controlled by ethylene can be increased by exposure of the fruit to an atmosphere containing exogenous ethylene in order to produce homogeneous external color (Salveit, 1999).

In climacteric fruits, application of ethylene stimulates and accelerates the ripening process, and has been employed as a treatment to both hasten and homogenize the ripening of several fruits (Salveit, 1999). The quality of fruit exposed to exogenous ethylene for ripening purposes is dependent upon the proper use of optimal levels of ethylene, $CO_2$ and $O_2$ concentrations, temperature, relative humidity and exposure time (Medlicott *et al.*, 1990; Salveit, 1999). Several works have been published on ripening acceleration and homogenization of mango by ethylene exposure for 12-24 h (Zamora *et al.*, 2004; Lagunes *et al.*, 2007).

Sindhri mangoes to export distant markets (28-35 days) must be transported at cold temperature (12°C) in order to reach till consumers in good physiological conditions (Amin *et al.*, 2012). Unlikely, at this temperature for offshore transport, mangoes show uneven colour development sometime, resulting less acceptance in market as well as fetch low price (Montalvo *et al.*, 2007). To mitigate the issue of poor cosmetic look due to uneven colour development, preshipment ethylene was applied in this experiment.

## MATERIALS AND METHODS

Physiologically mature and healthy mango fruit (cv. Sindhri) were harvested from a commercial orchard at district Multan. After desaping, fruit was packed in corrugated cardboard boxes and transported at 17°C in reefer van to laboratory Postharvest Research and Training Centre, Institute of Horticultural Sciences, University of Agriculture, Faisalabad, Pakistan. Total number of fruits were 144. In lab. their manual sorting and grading was done by experts . After that 12 boxes were made with 12 fruits per box. The fruits were treated with ethylene (100 ppm) at 24°C for 24 hours using shot method (8 hours interval) and 4 boxes of each were subjected to Condition-I: Storage at 12±1°C; 85-90% RH for 28 days followed by holding at ambient condition (33±2°C; 55-60% RH) till ripening (Simulated sea shipment condition); Condition-II: Holding at

25±1°C; 55-60% RH till ripening (Air freight condition); Condition-III: Holding at 33±2°C; 55-60% RH till ripening. The experiment was laid out under Completely Randomized Design along with two factor arrangements and replicated thrice with 144 fruits (approx. 12 fruits/box) in each replicate. The fruit were removed from low temperature storage after four weeks in accordance with stimulated sea shipment studies followed by five days of shelf studies at 33°C. Observations were made at final day of ripening to check the effect of treatments on peel colour, fruit textural softness and disease incidence. At ripe stage fruit were subjected to bio-chemical analysis such as TSS, total titratable acidity and vitamin C in order to evaluate the internal fruit quality (Amin *et al* ., 2007). Organoleptic evaluation of ripe fruit was done regarding taste, texture, flavor, pulp colour and aroma (Peryam & Pilgrim, 1957). Fruit peel colour, fruit textural softness were estimated by visual observations (Malik and Singh, 2005). Fruit colour was scored from 1 to 5 (1: 100% green – 0% yellow; 2: 75% green- 25% yellow; 3: 50% green- 50% yellow; 4: 25% green-75% yellow; 5: 0% green- 100% yellow). Similarly, fruit textural softness was rated from 1 to 5 scores (1: hard; 2: sprung; 3: slightly soft; 4: eating soft; & 5: over ripe).

**Statistical Analysis**

The data were subjected to analysis of variance (ANOVA) using Statistics 8.1 software and treatment means were compared using Least Significance Difference (LSD) Test at 5% level of significance ($P \leq 0.05$) (Steel *et al* ., 1997).

## RESULTS AND DISCUSSION

***Effect of Stimulated pre-shipment ethylene treatment on physic-chemical quality of cv. 'Sindhri'***

Data regarding fruit peel colour exhibited significant results at $P \leq 0.05$. Higher peel colour score of 4.93 and 4.87 was noted in fruit stored at 25±2 (8 days) and 33±1˚C (6 days), respectively. Whereas, fruit stored at 12±1˚C exhibited lower peel colour score of 3.80 when analysed after 28 days of storage (Figure 1a). Fruit softness score remained poor at lower temperature than higher ones. Fruit stored at 12˚C for 28 days exhibited minimum softness score (2.92) while maximum score was recorded in fruit stored at 25±1˚C analysed after 8 days of storage (Figure 1b). As the skin colour development and fruit softness was lower in fruit stored at 12 and 25°C than fruit kept at 33; it is obvious that post shipment ripening stages could be due to the exogenous ethylene application (before storage) and also that low temperature storage slows down the process of fruit ripening in mangoes (Govender *et al.*, 2005).

The fruit stored at 25±1˚C and 33±1˚C revealed higher shrivelling scores of 4.72 and 4.12 after 6 and 8 days of storage, respectively and these were also statistically at par with each other. While fruit stored at 12±1˚C for 28 days exhibited lower shrivelling score of 3.45 (Figure 1c). The fruit stored at 25±1˚C for 8 days showed higher anthracnose score of 4.12; while, the fruit stored at 33±1˚C (6 days) and 12°C (28 days) attained the anthracnose scores of 3.27 and 2.92, respectively and these were statistically at par with each other (Figure 1d). Significantly higher skin shrivelling and disease development (stem end rot, anthracnose) at ripe stage in the fruit stored at higher temperature 33°C as compared to low temperature (12 and 25°C). These results are also confirmed by Nunes and Emond (2007) the relationship of temperature with the fruit quality pattern. Due to similar level of disease incidence and skin shrivelling at 12°C, storage of 'Sindhri' mango at 12°C (28 days) seems to be relatively more advantageous as compared to high temperature stored fruits.

Different storage temperatures showed variable response regarding the TSS. Fruit stored at 12±1°C for 28 days showed higher value of TSS (18.98°Brix). Meanwhile, the fruit stored at 33±1°C (6 days) and 25±1°C (8 days) exhibited TSS values of 18.10 and 14.52°Brix, respectively (Figure 2a). In the current study, unusually higher TSS was observed during ripening in the fruit stored at lower temperature (12 and 25°C) despite of lower rates of respiration and sugar conversion during storage as compared to fruit kept at higher temperature (33°C). It might be due to rapid sugar conversion rate during ripening under similar conditions, thus, resulting in higher TSS (Tefera *et al.,* 2008). Data regarding acidity showed that fruit stored at 12±1°C for 28 days exhibited lower total titratable acidity (0.319%) as compared to the fruit stored at 33±1°C for 6 days and 25±1°C for 8 days where acidity values were 0.344 and 0.405%, respectively (Figure 2b). Higher titratable acidity in the fruit held at 33°C seemed to be the impact of temperature as observed earlier by various researchers as compared to lower temperature (Seyoum, 2002). The ascorbic acid contents increased gradually with the increase of temperature but to a certain extent and then it decreased. Fruit stored at 25±2°C for 8 days exhibited higher ascorbic acid (31.04 mg/100 g FW). While, lower ascorbic acid (22.69 mg/100 g FW) was noted in fruit stored at 12±1°C after 28 days of storage (Figure 3c). Lowest ascorbic acid contents were recorded in fruit stored at 12 and 25°C as compared to 33°C, that might be due to the conversion of organic acids into sugars since this fruit has better TSS (Kader, 2008).

PWL increased progressively with the rise of storage temperature. Fruit stored at 12±1°C for 28 days showed less PWL (7.26%) as compared to fruit stored at 25±1°C (8 days) and 33±1°C (6 days) where values of PWL were 12.43 and 20.90%, respectively (Figure 3a). The fruit stored at 12±1°C for 28 days showed lower respiration rate (7.26 mmol $CO_2$ $Kg^{-1}h^{-1}$) as compared to the fruit stored at 25±1 (8 days) and 33±2°C (6 days) where respiration rate of 12.43 and 20.90 mmol $CO_2$ $Kg^{-1}h^{-1}$, respectively was measured (Figure 3b). The ethylene production rate showed positive variation among all the storage temperatures. Higher ethylene liberation rate (1.14 mmol $C_2H_2$ $Kg^{-1}h^{-1}$) was measured in the fruits stored at 33±2°C after 6 days. While lower ethylene liberation rate (0.57 mmol $C_2H_2$ $Kg^{-1}h^{-1}$) was recorded in fruit stored at 12°C analysed after 28 days. The ethylene liberation rate showed positive variation among all the storage temperatures. Higher ethylene liberation rate (1.14 mmol $C_2H_2$ $Kg^{-1}h^{-1}$) was measured in the fruits stored at 33±2°C after 6 days followed by 25±1°C (0.90 mmol $C_2H_2$ $Kg^{-1}h^{-1}$) after 8 days of storage. While, lower ethylene liberation rate (0.57 mmol $C_2H_2$ $Kg^{-1}h^{-1}$) was recorded in fruit stored at 12°C analysed after 28 days (Figure 3c). PWL was significantly remained poor in fruits stored at 12 and 25°C as compared to 33°C. Reduced respiration rate during ripening in the fruit stored at 12 and 25°C as compared to 33°C revealed the significant impact of storage conditions on physiological processes of cv. 'Sindhri' mangoes even after storage as described by Yamashita *et al.* (1997).

Total antioxidants activity remained poor at higher temperature as compared to lower temperature. Maximum total antioxidants activity (66.54%) was noted in fruit stored at 12±1°C after 28 days followed by 25±1 (8 days) and 33±1°C (6 days) where values of antioxidants activities were 52.37 and 44.77%, respectively and these were statistically at par with each other (Figure 5a). Total carotenoids remained higher as well as storage temperature increased but to a certain extent and then decreased gradually. Fruit stored at 12±1°C for 28 days exhibited lower total carotenoids (16.27 µg $g^{-1}$) as compared to the fruit stored at 25±1°C (8 days) and 33±2°C (6 days) where total

carotenoids of 24.09 and 35.29 µg $g^{-1}$, respectively were recorded (Figure 5b). The total phenolic contents declined positively as well as storage temperature increased. Higher total phenolic contents (40.49 mg GAE/100 g) were noted in fruits stored at 12±1°C after 28 days as compared to the fruits stored at 25±1°C (33.63 mg GAE/100 g) after 8 days and 33±2°C (21.65 mg GAE/100 g) after 6 days (Figure 5c). Fruit phytochemicals such as total carotenoids, total antioxidants and total phenolics showed significant variations regarding different storage temperatures in cv. 'Sindhri' mangoes. Fruit stored at 12 and 25°C showed lower total carotenoids contents as compared to 33°C. At higher storage temperature (33°C), the process of ripening seems to be at comparatively higher rate as compared to lower storage temperature (12 and 25°C), so that resulted in higher total carotenoids biosynthesis in the fruit. Previous reports also mention that low temperature retains green colouration for longer period and affects total carotenoid contents during storage (Medlicott *et al.,* 1986); while, Yahia and Pedro-Campos (2000) studied that total carotenoid contents were higher in fruit stored at 20°C than stored at 10°C. Fruit stored at low temperature (12 and 25°C) exhibited higher TPC and antioxidants activity which were possibly due to the activities of PAL enzyme that has been reported to be involved in TPC metabolism (Dixon and Paiva, 1995). In contrast, fruit stored at higher temperature (33°C) revealed reduced TPC and antioxidants activity and this reduction was may be due to the gradual enzymatic changes during storage. Moreover, our results are in strong agreement with the findings of Teiz and Zeiger (2010) who observed reduced activities of TPC during storage. Similarly, Pennycooke *et al.* (2004) also found that cold storage of fruit increased the total phenolics and antioxidant capacity in citrus.

Fruit stored at 12±1°C exhibited higher pulp colour score (7.20) as compared to the fruit kept at 25±1°C and 33±1°C where pulp colour scores were 6.30 and 5.78, respectively (Figure 4a). Fruit texture score showed a variation at different storage temperatures. Higher texture score (7.69) was recorded in fruits stored at 33±1°C for 6 days. Whereas, fruit stored at 12±1°C (28 days) and 25±1°C (8 days) exhibited texture scores of 6.35 and 5.65, respectively and these were statistically similar to each other (Figure 4b). Fruit stored at 12±1°C exhibited higher aroma score (7.15) as compared to the fruit held at 25±1°C (6.0) and 33±1°C (5.29) (Figure 4c). Fruit flavour score decreased gradually with the increase of temperature. Maximum taste score (7.81) was noted in fruits stored at 12±1°C analysed after 28 days (Figure 4d). Similarly, maximum taste score (7.81) was noted in fruits stored at 12±1°C when analysed after 28 days. While fruit stored at 33±1 (6 days) and 25±1°C (8 days) exhibited taste scores of 6.97 and 4.51, respectively (Figure 4e).

## CONCLUSION

The mango fruit cv. 'Sindhri' subjected to stimulated pre-storage ethylene (100 ppm) at 24°C for 24 hours did not have any negative effect on fruit quality attributes. The fruit could be subjected to sea shipping conditions (12±1°C; 85-90% RH) for 28 days with post storage shelf life of 4-5 days. While for air freight, the pre-shipment ethylene treated mangoes had 8 days shelf life at 25±1°C (55-60% RH) and 6 days at 33±2°C (55-60% RH). Therefore, 'Sindhri' mangoes have potential for pre-shipment ethylene treatment for sea and air export consignments. In future investigations, there is need to study the weekly and day to day changes under storage and ambient shelf respectively.

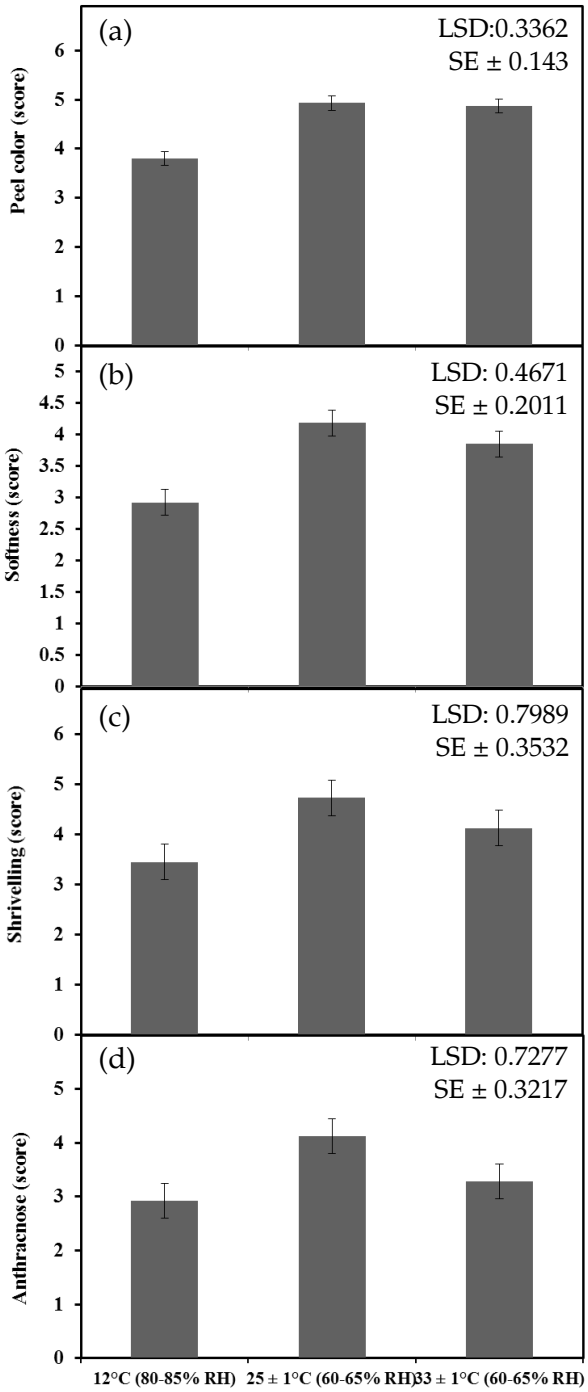


**Figure 1:** Effect of Stimulated pre-shipment ethylene treatment on (a) peel color, (b) fruit softness, (c) shrivelling and (d) anthracnose. n=44.

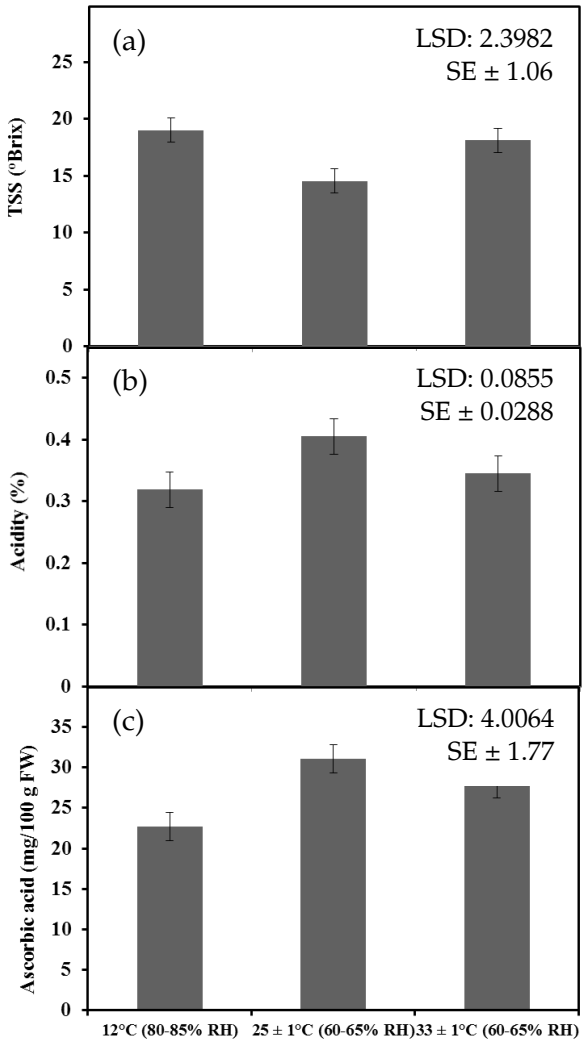


**Figure 2:** Effect of Stimulated pre-shipment ethylene treatment on (a) TSS, (b) TA and (c) ascorbic acid contents. n=44.

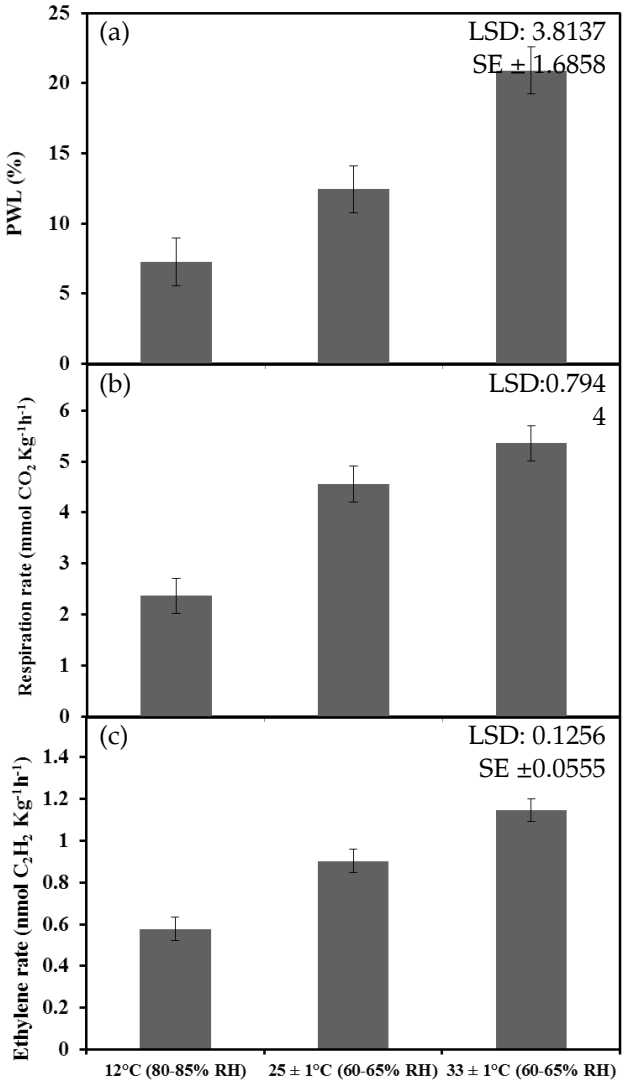


**Figure 3:** Effect of Stimulated pre-shipment ethylene treatment on (a) PWL, (b) respiration rate and (c) ethylene production. n=44.

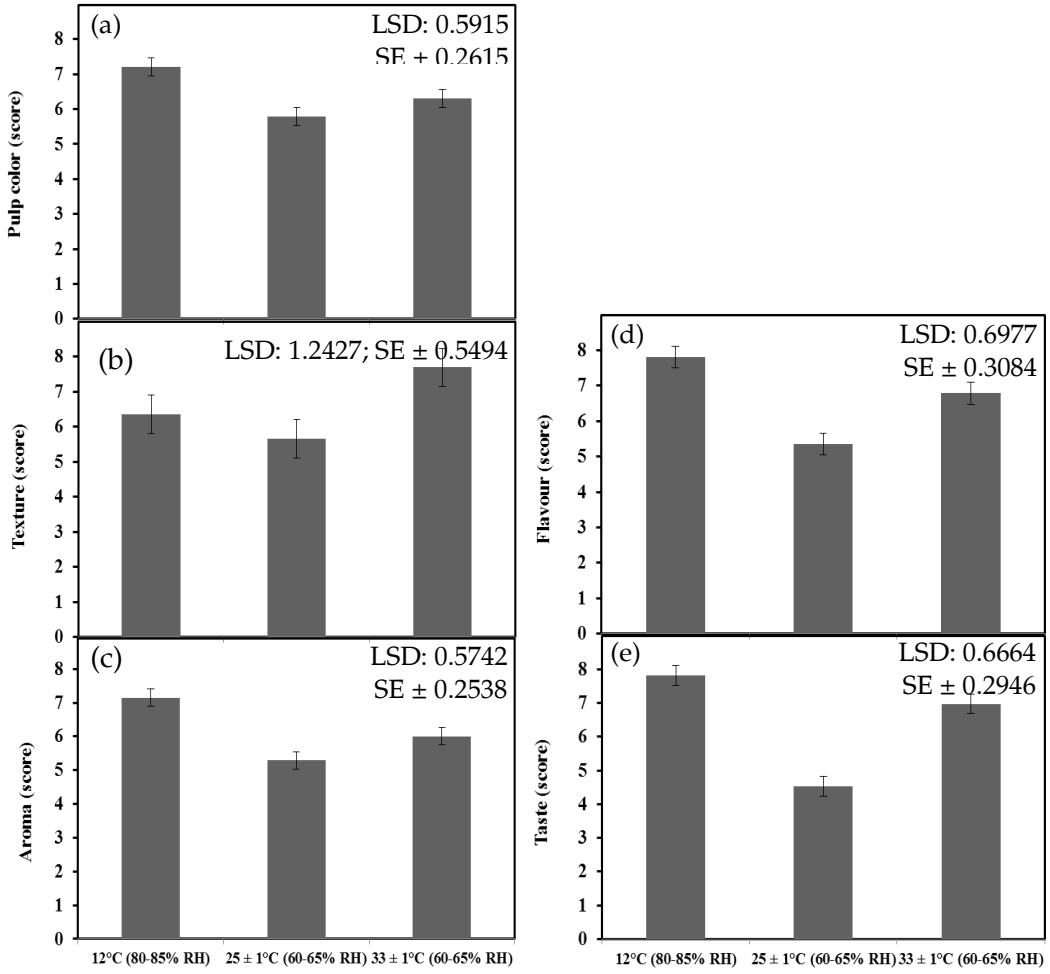


**Figure 4:** Effect of Stimulated pre-shipment ethylene treatment on (a) Pulp color, (b) Texture, (c) Aroma, (d) Flavor and (e) Taste. n=44.

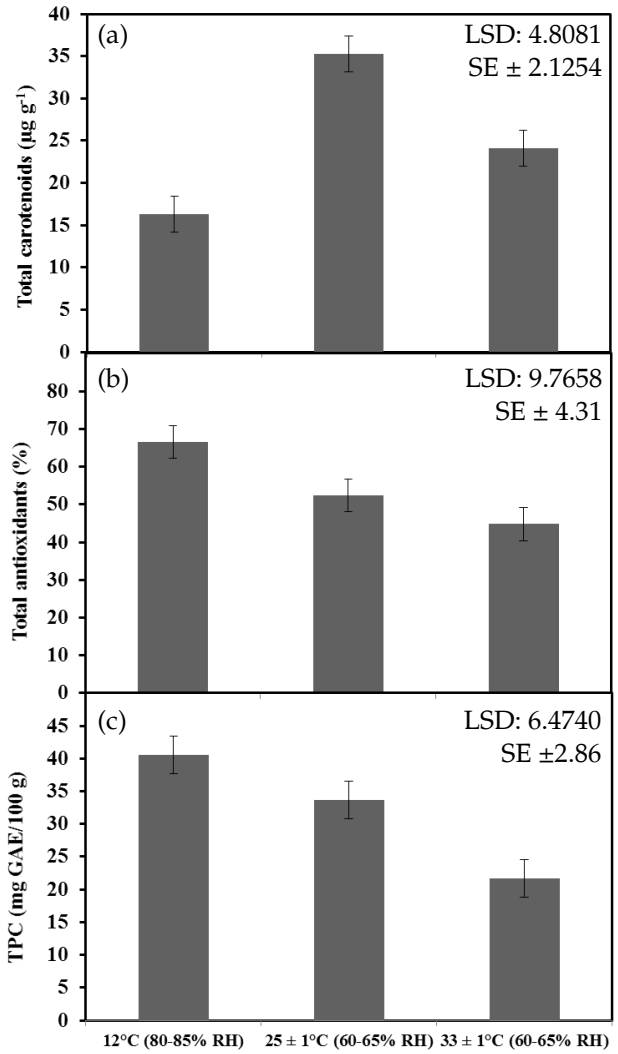


**Figure 5:** Effect of Stimulated pre-shipment ethylene treatment on (a) Total Carotenoids, (b) Total Antioxidants and (c) Total Phenolic Contents. n=44.

## REFERENCES


Amin, M. 2012. Integrated approaches for improving fruit quality and shelf life of two commercial mango cultivars of Pakistan. Ph.D. Thesis. Institute of Horticultural Sciences, University of Agriculture Faisalabad, Pakistan.

Asif, M., M. Usman, M.J. Jaskani and M.M. Khan. 2002. Comparative study of flower sex ratio in different cultivars of mango (*Mangifera indica* L). Int. J. Agric. Biol. 4:220-222.

Barmore, C.R. 1974. Ripening mangoes with ethylene and ethephon. Proc. Fla. State Hort. Soc. 87:331-334.

Cua, A.U. and M.C.C. Lizada. 1990. Ethylene production in the 'Carabao' mango (*Mangifera indica* L). fruit during maturation and ripening. Acta Hort., 269: 169-79

Curry, E.A. 1994. Pre-harvest applications of ethephon reduce superficial scald of Fuji and Granny Smith apples in Storage. J. Hort. Sci. 69: 1111-1116.

FAO. 2012. Food and Agriculture Organization of United Nations. (Available online with updates at: http://faostat.fao.org/site/339/default.aspx.)

Govender, L., L. Korsten and D. Sivakumar. 2005. Semi-commercial evaluation of *Bacillus licheniformis* to control mango postharvest disease in South Africa. Postharvest Biol. Technol., 38: 57-65.

Jha, S.N., K. Narsaiah, A.D. Sharma, M. Singh, S. Bansal and R. Kumar. 2010. Quality parameters of mango and potential of non-destructive techniques for their measurement – A review. J. Food. Sci. Technol. 47:1-14.

Kader, A. and B. Mitcham. 2008. Optimum procedures for ripening mangoes. (Available online with updates on: http://postharvest.ucdavis.edu/Pubs/Pub_Desc_9.pdf.)

Lalel, H.J.D., Z. Singh and S.C. Tan. 2003a. Aroma volatiles production during fruit ripening of 'Kensington Pride' mango. Postharvest Biol. Technol. 27:323-336.

Lalel, H.J.D., Z. Singh and S.C. Tan. 2003b. Distribution of aroma volatile compounds in different parts of mango fruit. J. Hortic. Sci. Biotechnol. 78:131-138.

Li, Z.F., N. Wang, G.S.V. Raghavan and C. Vigneault. 2009. Ripeness and rot evaluation of 'Tommy Atkins' mango fruit through volatiles detection. J. Food Eng. 91:319-32.

Meadows, J. 1998. Florida food fair. Cooperative Extension Service for Sarasota Country. University of Florida: Extension Institute of Food and Agriculture Sciences.

Medlicott, A.P. 1986. Report on a visit to Institute of Technology Assessment and Compliance Certification (ITAC) Brasil to investigate the effects of maturity, storage and gas treatment on mango fruit ripening. Trop. Devlp. Res. Inst. UK, Visit Report.

Medlicott, A.P., J.M. Sigrist, S.B. Reynolds and A.K. Thompson. 1987. Effects of ethylene and acetylene on mango fruit ripening. Ann. Appl. Biol. 111:439-444.

Montalvo, E., H.S. García, B. Tovar and M. Mata. 2007. Application of exogenous ethylene on postharvest ripening of refrigerated 'Ataulfo' mangoes. Food Sci. Technol.40: 1466-1472.

Mukherjee, S.K. and R.E. Litz. 2009. Introduction: Botany and importance. In: R.E. Litz (Ed.). The Mango: Botany, Production and Uses (2nd Ed.). CAB International, Wallingford, UK.p.1-18.

Nagel, M.C. 1989. The Fruits of Ethylene. ChemMatters. **7**:11-13.

Padda, M.S., V.T.A. Cassandro, M.G. Raphael, C.S. David and J.M. Elzabeth. 2011. Methods to analyze physico-chemical changes during mango ripening: A multivariate approach. Postharvest Biol. Technol. 62:267-274.

Per, H., S. Kurtoglu, F. Yagmur, H. Gumus, S. Kumandaş and M.H. Poyrazoglu. 2007. Calcium carbide poisoning via food in childhood. J Emergi Med. 32: 179-80.

Peryam, D.R. and F.J. Pilgrim. 1957. Hedonic scale method of measuring food preferences. Food Tech. 11: 9-14.

Sankat, C.K., K. Bissoon and R. Maharaj. 1994. Ripening quality of Julie mangoes stored at low temperatures. Acta Hort. 368:712-722.

Sergent, E., B. Schaffer, P. Lara and L. Willis. 1993. Effect of ethephon on mango (*Mangifera indica* L). fruit quality. Acta Hort. 341: 510-517.

Seyoum, T. 2002. The improvement of the shelf life of vegetables through pre- and postharvest treatment. PhD thesis. Univ. Free State, South Africa.

Siddiqui, M.W. and R.S. Dhua. 2010. Eating artificially ripened fruits is harmful. Curr. Sci. 99:1664-1668.

Singh, Z. and J. Janes. 2001. Effect of postharvest application of ethephon on fruit ripening, quality and shelf life of mango under modified atmosphere packaging. Acta Hort. 553: 599-601.

Steel, R.G.D., J.H. Torrie and D.A. Dicky. 1997. Principles and Procedures of Statistics. A Biometrical Approach, 3rd ed. McGraw Hill Book Co. New York., USA.

Tefera, A., T. Seyoum and K. Woldetsadik. 2008. Effects of disinfection, packaging and evaporatively cooled storage on sugar content of mango. African J. Biotechnol. 7: 65-72.

Usman, M., B. Fatima, M.M. Khan and M.I. Chaudhry. 2003. Mango in Pakistan: A chronological review. Pak. J. Agri. Sci. 40:151-154.

Yamashita, A., T. Sugiura and K. Waku. 1997. Acyltransferases and transacylases involved in fatty acid remodeling of phospholipids and metabolism of bioactive lipids in mammalian cells. J. Biochem. 122:1-6.