

Evaluation of the Effect of Ethephon in Postharvest Tomato (*Lycopersicon esculentum*)

ABSTRACT

Aims: The aim of this study is to determine the effect of applying ethephon on postharvest tomato.

Study design: Completely Randomized Design (CRD).

Place and Duration of Study: The study was conducted in the laboratory of the Department of Applied Food Science and Nutrition, Department of Food Processing and Engineering, Department of Applied Chemistry and Chemical Technology of Chittagong Veterinary and Animal Sciences University, Chittagong, Bangladesh. It was conducted for a period of six months from 1st January, 2018 to 30th June, 2018.

Methodology: TSS (Total Soluble Solid) was determined by using hand refractometer, proximate composition by standard AOAC method, vitamin C and β -carotene by UV-spectrophotometric method, sodium and potassium by flame photometric method, titratable acidity, calcium and magnesium by titrimetric method. To compare differences in control and treated group, significant difference was considered at the level of $P < 0.05$.

Results: Ethephon treated tomatoes ripen quickly with attractive surface color and had shorter shelf life than non-treated tomatoes (control). Physico-chemical properties including total soluble solid and titratable acidity increased significantly ($P < 0.05$) in treated group ($4.66 \pm 0.57^\circ\text{B}$; $0.32 \pm 0.01\%$ respectively) than control ($3.33 \pm 0.57^\circ\text{B}$; $0.28 \pm 0.01\%$ respectively). Moisture content also significantly ($P < 0.05$) increased in ethephon treated tomato ($94.15 \pm 0.27\text{g}/100\text{g}$) than non-treated ($85.54 \pm 0.23\text{g}/100\text{g}$). But significantly lower amount of carbohydrate, ash, crude fiber were observed in artificially ripened tomato by ethephon ($2.5 \pm 0.45\text{g}/100\text{g}$; $0.51 \pm 0.02\text{g}/100\text{g}$; $0.82 \pm 0.03\text{g}/100\text{g}$ respectively) than naturally ripened ($3.7 \pm 0.26\text{g}/100\text{g}$; $0.59 \pm 0.01\text{g}/100\text{g}$; $0.90 \pm 0.01\text{g}/100\text{g}$ respectively). Apparently protein and fat content were found insignificantly less amount in artificially ripened tomato by ethephon. The significant lowest concentration of vitamin C and β -carotene were observed in ethephon ripened tomato ($16.65 \pm 0.01\text{mg}/100\text{g}$; $413.33 \pm 1.15\mu\text{g}/100\text{g}$ respectively). Mineral contents of ethephon treated tomato (Na $3.65 \pm 0.47\text{mg}/100\text{g}$; K $199 \pm 1.00\text{mg}/100\text{g}$; Ca $4.75 \pm 0.07\text{mg}/100\text{g}$; Mg $8.57 \pm 0.19\text{mg}/100\text{g}$) found significantly ($P < 0.05$) fewer amounts than control tomato (Na $3.96 \pm 0.01\text{mg}/100\text{g}$; K $213.67 \pm 0.57\text{mg}/100\text{g}$; Ca $5.57 \pm 0.15\text{mg}/100\text{g}$; Mg $9.96 \pm 0.04\text{mg}/100\text{g}$).

Conclusion: Application of ethephon on postharvest tomato hastens ripening time and physico-chemical properties but reduces shelf life and nutritive value of tomato.

Keywords: Ethephon; tomato; nutritional quality; ripening time; shelf life.

1. INTRODUCTION

Fruits and vegetables are generally rich sources of vitamins, minerals, phytochemicals, dietary fibers, carotenoids and polyphenols [1-3]. Tomato (*Lycopersicon esculentum*) is the most important fruit of the world including tropical, sub-tropical and temperate regions [4]. It is widely used by food industries as a raw material for the production of different products such as puree or ketchup. It is the most common fruit in the Mediterranean diet, a diet known for its health, especially as regards the development of chronic degenerative diseases [5]. Worldwide tomato ranks third in area and production after potato and sweet potato but ranks first among processed vegetables [4]. It is one of the most important and popular vegetables in Bangladesh with a considerable total production of 3,88,725 metric tons produced in an area of 68,366 acres [6].

Tomato is a rich source of vitamin A and C and is known as "poor man's orange". Among fruits and vegetables, this fruit occupy the 16th place as a source of vitamin A and the 13th as a source of vitamin C. Lycopene in tomatoes has anti-tumor properties. It also serves as an antioxidant because the β -carotene present in this fruit helps to prevent and neutralize free radical and ascorbic acid acts as an effective eliminator of superoxide, hydrogen peroxide, singlet oxygen and other free radicals [4].

Ripening is a natural process that brings a series of biochemical changes that are responsible for changes in color, respiration rate, ethylene production rate, tissue permeability, softening, nutrient composition and production of volatile compounds in the skin etc [7]. The ripening of the tomato is characterized by the loss of chlorophyll and the rapid accumulation of carotenoids, especially lycopene, since chloroplasts become chromoplasts [8].

For the prevention of postharvest losses, calcium carbide (CaC_2), ethylene (C_2H_2), ethephon ($\text{C}_2\text{H}_6\text{ClO}_3\text{P}$) and other non-recommended pesticides are used for the ripening of immature fruits quickly with a nice color [9]. Many well-known pesticides and chemical industries import ripening agents and market them under various names, such as Sundari, Harvest, Bevorder, Ripe, Ripen, Eden, Prolong, Ethrel, Remote, Garden, Action and Gold Plus. But, none of these is legal. In reality, the government has never approved a ripening agent. It has only approved two plant growth regulators (PGRs) and hormones which are used to make the fruit larger and more attractive. They leave because the government has no mechanism to verify which products are imported and sold. But even so, a section of traders is used the chemical spray to ripen tomatoes and is marketing them in different markets in the district. Traders generally use Ripen-15, a poisonous chemical spray to ripen tomatoes in a short time and for immediate profit, since the price of ripe tomatoes is comparatively higher than greens [10]. Ripen 15 contains 80% ethephon is used indiscriminately for ripening tomato [11].

Ethephon is widely used for tomato ripening [10]. It may have received registration as a plant growth regulator for early flowering. But this chemical is improperly used as ripening agent [11]. Ethephon ripened fruits have more acceptable colour and highest extent of moisture content as compared with the naturally ripened fruits [12]. Farmers easily get these chemicals from traders and use them excessively in tomatoes. But if this type of chemical is used in tomatoes, it increases the risk of heart attack, liver problem and kidney damage [10].

It is recommended to use a tomato ripening agent all over the world for uniform ripening, especially for commercial purposes. Tomatoes are usually harvested at mature-green or breaker or turning pink stage and ethylene gas is used to accelerate the ripening. In this process, the quality of the fruit is not affected at all. However, in Bangladesh, the practice of using the ripening agent is different. Here the fruits are collected at immature conditions and stored in the farmer's house and sprayed the ripening chemicals like ethephon. The fruits

ripen with an attractive red color, but the actual taste, flavor and quality are lacking due to the premature harvesting. The advantage from the point of view of intermediaries is that the cost of labor is reduced to a minimum due to the same collection time of all fruits and the ripe fruits remain rigid for a rather long period and the damage is reduced. Unfortunately, these benefits are at the cost of consumer satisfaction, fruit quality and health risks [11]. Though tomatoes are the source of micronutrient but ethephon which is used as ripening agent in tomatoes have effects on the nutritional quality of fruits [9]. Considering the forgoing problem, the present section describes the effects of ethephon on the changes of ripening time, shelf life, physico-chemical properties and nutritional composition (carbohydrate, protein, fat, fiber, ash, crude fiber, vitamin and mineral content) in postharvest tomato. It is necessary to put forward recommendations to the policy makers to develop strategic national plan to maintain the fruits quality in the supply chain.

2. MATERIAL AND METHODS

2.1 Collection of Sample

Fresh tomatoes were collected directly from production field of Khulshi and Oxygen area of Chittagong into clean polythene bags and transported to the laboratory for analysis.

2.2 Experimental Design

The experiment was arranged in completely randomized design (CRD). Selected tomatoes were divided into two experimental groups namely control (non-treated) and ripened by ethephon. In cases of non-treated, tomatoes were kept in bamboo basket and covered with straw. For ethephon treated group green fruits were dipped into ethephon solution. Each experiment was replicated for three times.

2.3 Pre-treatment of Samples

The skin (body) of tomatoes were washed gently with distilled water and then cleaned properly with cotton cloth to remove dust, adhered particles and agricultural chemicals. Then tomatoes were stored in a cool and dry place.

2.4 Treatment by Ethephon

Selected tomatoes were dipped into 1000 ppm ethephon solution for 10 minutes. Then tomatoes were air dried before packing in cardboard/cartoon. Selected tomatoes were stored at room temperature [13].

2.5 Determination of Ripening Time

The ripening time of the fruits were estimated by counting the days from the day of ethephon application to attain the red color surface of tomato between 60% to 90% with the help of tomato color chart [14].

2.6 Determination of Shelf Life

The shelf life was calculated by counting the days required to attain the last stage of ripening but up to the stage when tomato remained still acceptable for marketing [14].

2.7 Determination of Total Soluble Solid (TSS)

The total soluble solid content was recorded with the help of a portable refractometer. The crushed tomato pulp was placed in the prism of the refractometer and the readings were observed through the eyepiece. For a precise measurement, the readings taken were corrected for temperature variations at 20°C and the results were expressed as °Brix (°B) [15].

2.8 Determination of Titratable Acidity

A known weight of the crushed tomato sample was taken in a 100 ml volumetric flask and the volume was made up by the addition of distilled water. After filtration, 10 ml of the filtrate was taken in a separate conical flask and then titrated against 0.1 N sodium hydroxide using phenolphthalein as an indicator. The end point was determined by the appearance of a faint pink colour. Then titratable acidity was calculated [15]. The titratable acidity was determined by using the following formula

$$\text{Titratable acidity \%} = \frac{\text{Eq. wt of acid} \times \text{Normality of NaOH} \times \text{Volume made up} \times \text{Titre value}}{\text{wt of sample} \times \text{Volume of extract taken} \times 1000} \times 100$$

2.9 Determination of Proximate Composition Analysis

The moisture, protein, fat, ash, crude fiber content of the experimental groups were measured in triplicate using the standard AOAC methods [16]. The moisture was measured by oven drying at 105°C to constant weight, crude protein content by the Kjeldahl procedure (6.25 × N), total fat by extraction of ether, ash by incineration in a muffle furnace at 550°C to constant weight. Crude fiber was measured by digesting the sample. Carbohydrate content was calculated by difference method i.e

$$\% \text{Carbohydrate} = 100 - (\% \text{Moisture} + \% \text{Protein} + \% \text{Fat} + \% \text{Ash} + \% \text{Fiber})$$

2.10 Determination of Vitamin C

Vitamin C was determined by UV visible spectrophotometric method, as described by Rahman et al. [17].

2.11 Determination of β-Carotene

β-carotene content was analyzed by UV-spectrophotometric method, as described by Karnjanawipagul et al. [18].

2.12 Determination of Minerals

Sodium (Na) and potassium (K) content were determined by using flame photometer [19]. Sodium and potassium in solution was atomized into an oxy-hydrogen flame that emits atoms of sodium or potassium, causing them to emit radiation to specific wavelength. The amount of radiation emitted is measured on a flame photometer, under standard condition which is proportional to the concentration of sodium or potassium. Calcium (Ca) and magnesium (Mg) content were analyzed by using titrimetric method [15].

2.13 Statistical Analysis

Data collected in this study was analyzed using MS Excel, 2007 and SPSS (Statistical Package for the Social Sciences) version 22.0. *P* value was used to compare differences in the means of the ripening time, shelf life, total soluble solid (TSS), titratable acidity, moisture,

protein, fat, carbohydrate, ash, crude fiber, vitamin C, β -carotene and minerals. A significant difference was considered at the level of $P<0.05$.

3. RESULTS AND DISCUSSION

3.1 Effect of Ethephon on Ripening Time and Shelf Life of Tomato

Results of the effect of ethephon on ripening time and shelf life are shown in table 1. Significant differences ($P<0.05$) were observed in ripening time and shelf life of tomato those were ripened artificially with ethephon than naturally or control group.

The ethephon treatment registered the lowest ripening time (2 days) while the highest was recorded in control (5 days). It clearly indicated that ethephon was more effective in early ripening of tomatoes as compared to untreated. Higher concentration of ethephon increases the ripening time of fruits [4, 9]. Improved ripening with post-harvest ethephon application is due to the binding of ethylene to the receptor, which forms an activated complex that leads to a wide variety of physiological responses, including ripening [20]. Enhanced ripening with the post-harvest application of ethephon has been reported in tomato [4, 9], pear [21] and mango [22, 23].

Mature green tomato had a higher storability than ethephon treated tomato. According to the present study, ethephon levels had also significant ($P<0.05$) effect on shelf life of tomatoes. The shelf life of control and ethephon treated tomato were recorded 15 days and 5 days respectively. Moreover, lower shelf life was recorded in the ethephon treated tomatoes which make the fruits unmarketable after 5 days of ripening period. The untreated (control) ripened fruits recorded maximum shelf life of 15 days. The lower shelf life in ethephon dip treatments may be due to direct contact of fruits with water. Because some unnoticeable injuries and bruises on fruit surface may absorb this water during dipping into ethephon solution which later became the entry point for the fungal infection [4]. Ethephon is responsible for faster respiration rate in fruits which leads to over softening and spoilage of fruit [24]. Similar types of results were corroborated in tomato [4, 9], pineapple and banana [9].

Table 1. Comparison of ripening time (days) and shelf life (days) between control and ethephon treated tomato

Sample	Variable (Days)	Control	Ripened by ethephon	P-value
Tomato	Ripening time	5.3 \pm 0.57	2.3 \pm 0.57	0.003*
	Shelf life	14.67 \pm 0.57	4.66 \pm 0.57	0.000*

**Statistically significant at 5% level*

Results are means \pm standard deviation (SD) of triplicate

3.2 Effect of Ethephon on Physico-chemical Parameter of Tomato

In the present study, the total soluble solid and titratable acidity content was influenced by ethephon treatment (table 2).

Total soluble solid (TSS) content of tomato pulp varied significantly ($P<0.05$) in control and ethephon treated. The ethephon treated fruits recorded maximum TSS content (4.66 \pm 0.57 $^{\circ}$ B). The control fruits recorded the lowest TSS content (3.33 \pm 0.57 $^{\circ}$ B). The increase in TSS during ripening may result from hydrolysis of starch and other polysaccharides to soluble form of sugar [4] and different types of anabolic and catabolic processes taking place in fruits, preparing it for senescence [25]. Higher TSS content

represents more ripeness of fruits among ripe fruits of the same variety [26]. The findings in this study is in consonance with the results were observed in tomato [4, 14, 27, 28] and pear [21].

Titrateable acidity of tomato was found to be maximum ($0.32\pm 0.01\%$) in ethephon treated fruits. The minimum acidity was noted in the control fruits ($0.28\pm 0.01\%$). A significant difference ($P<0.05$) in the acid content of the fruits was observed between ethephon treated and control fruits. In general, the acidity content of the tomato fruit decreases during the ripening period, which may be due to the utilization of organic acid in the pyruvate decarboxylation reaction that occurs during the fruit ripening process [29]. In the present observation, highest titrateable acidity was observed in ethephon treated fruit compared to control. The increased permeability of the membrane allows the acids stored in the cell vacuoles to breathe at a higher rate [4]. The similar observations were reported in tomato [4, 14], banana [9] and mango [23].

Table 2. Comparison on physico-chemical parameter between control and ethephon treated tomato

Sample	Variable	Control	Ripened by ethephon	P-value
Tomato	Total soluble solid (°B)	3.33 ± 0.57	4.66 ± 0.57	0.047*
	Titrateable acidity (%)	0.28 ± 0.01	0.32 ± 0.01	0.004*

*Statistically significant at 5% level

Results are means \pm standard deviation (SD) of triplicate

3.3 Effect of Ethephon on Proximate Composition of Tomato

The proximate analysis was conducted for comparing the tomato between naturally ripened (ethephon free) and artificially ripened after the treatment by ethephon. The results are represented in Table 3. The results were significant ($P<0.05$) for the moisture, carbohydrate, ash and crude fiber in tomato ripened by the ethephon.

The moisture contents of tomato were $85.54\pm 0.23\text{g}/100\text{g}$ for non-treated control group and $94.15\pm 0.27\text{g}/100\text{g}$ for ethephon-treated group. Analysis of data revealed that the ethephon treated samples had the highest extent of moisture content as compared with the untreated fruits. Due to the excessive formation of moisture, ethephon treated sample causes a significant increase in fruit weight [30]. Similar findings was reported in tomato and pineapple [9].

Table 3 indicates that carbohydrate, ash and crude fiber content in control or naturally ripened tomato was $3.7\pm 0.26\text{g}/100\text{g}$, $0.59\pm 0.01\text{g}/100\text{g}$, $0.90\pm 0.01\text{g}/100\text{g}$ respectively, which was significantly ($P<0.05$) reduced in ethephon treated ripe tomato. Similar trends were observed in the case of protein and fat content. Such findings revealed that ethephon treated fruits has poor nutritional quality as compared to naturally ripened fruits [9, 31]. The application of ethephon in postharvest tomato reduce protein, fat, carbohydrate, ash and crude fiber content in tomato [9] and banana [31].

Table 3. Comparison on proximate composition between control and ethephon treated tomato

Sample	Variable (g/100g)	Control	Ripened by ethephon	P-value
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Tomato	Moisture	85.54±0.23	94.15±0.27	0.000*
	Protein	1.24 ±0.05	1.17±0.02	0.361
	Fat	0.31±0.02	0.27± 0.02	0.063
	Carbohydrate	3.7±0.26	2.5±0.45	0.017*
	Ash	0.59±0.01	0.51±0.02	0.005*
	Crude Fiber	0.90±0.01	0.82±0.03	0.012*

*Statistically significant at 5% level

Results are means ± standard deviation (SD) of triplicate

3.4 Effect of Ethephon on Vitamin C and β-Carotene of Tomato

Vitamin C and β-carotene were decreased significantly ($P<0.05$) in tomato after ripened artificially by ethephon that are represented at table 4.

Tomato is a rich source of vitamin C (ascorbic acid). Results showed that non treated control group contained the highest quantity of vitamin C (20.28±0.01mg/100g) while the artificially ripened tomato by ethephon contained the lowest quantity of vitamin C (16.65±0.01mg/100g). The vitamin C content of fruit and vegetables also decreases in the correct preservation treatment due to the prolonged shelf life [11]. Hakim et al. [9] and Moneruzzaman et al. [27] also recorded the similar findings in the vitamin C content of tomato with application of ethephon.

Control or naturally ripend tomato contained 433.00±2.64µg/100gm β-carotene while the ethephon treated tomato contained 413.33±1.15µg/100gm. Similar results also reported in tomato, pineapple and banana [9].

Some metabolic intermediary substances are responsible for synthesis of the precursor of vitamin C and β-carotene [22]. Due to the application of ethephon, the production of these intermediary substances may be interrupted. So, vitamin C and β-carotene content was found lower in ethephon treated group than control group.

Table 4. Comparison on vitamin C and β-carotene between control and ethephon treated tomato

Sample	Variable	Control	Ripened by ethephon	P-value
Tomato	Vitamin C (mg/100g)	20.28±0.01	16.65±0.01	0.000*
	β-carotene (µg/100gm)	433.00±2.64	413.33±1.15	0.000*

*Statistically significant at 5% level

Results are means ± standard deviation (SD) of triplicate

3.5 Effect of Ethephon on Mineral Contents of Tomato

Minerals are important for the various metabolic activities of living tissue and even more for the fruit, which shows an enormous activity during the ripening process [9]. The results of the analysis of minerals (sodium, potassium, calcium and magnesium) of tomato samples are shown in Table 5. The analysis of data revealed that application of artificial ripening agent ethephon significantly ($P<0.05$) lower the mineral contents in tomato than naturally ripened. The maximum mineral content was observed in untreated (control) fruits. The ethephon treated fruits recorded minimum mineral content during the ripening period. Similar results

also observed in tomato [9]. Artificially ripened fruits contain less mineral contents than naturally ripened fruits [32].

Table 5. Comparison on mineral contents between control and ethephon treated tomato

Sample	Variable (mg/100g)	Control	Ripened by ethephon	P-value
Tomato	Na	3.96±0.01	3.65±0.47	0.000*
	K	213.67±0.57	199±1.00	0.000*
	Ca	5.57±0.15	4.75±0.07	0.001*
	Mg	9.96±0.04	8.57±0.19	0.000*

*Statistically significant at 5% level

Results are means ± standard deviation (SD) of triplicate

4. CONCLUSION

From the results of this study it has been revealed that, application of ethephon in postharvest tomato fruits provide attractive uniform color, quick ripening time and higher physico-chemical parameter. But, it greatly affects the shelf life and nutrient composition of postharvest tomato. Moreover, ethephon is hazardous for human health. So application of ethephon in tomato should be monitored properly and governments should take proper action against this. Not only governments but also producers and consumers should be aware about considering the aspects of using ethephon in postharvest tomato.

REFERENCES

1. Khoo H, Prasad KN, Kong K, Jiang Y, Ismail A. Carotenoids and their isomers: color pigments in fruits and vegetables. *Molecules*. 2011;16(2):1710-1738. DOI:10.3390/molecules16021710
2. Masibo M, He Q. Mango bioactive compounds and related nutraceutical properties-a review. *Food Reviews International*. 2009;25(4):346-370. DOI:10.1080/87559120903153524
3. Slavin JL, Lloyd B. Health benefits of fruits and vegetables. *Advances in Nutrition*. 2012;3(4):506-516. DOI: 10.3945/an.112.002154
4. Dhall RK, Singh P. Effect of ethephon and ethylene gas on ripening and quality of tomato (*Solanum lycopersicum* L.) during cold storage. *J Nutr Food Sci*. 2013;3(6):244. DOI: 10.4172/2155-9600.1000244
5. Leonardi C, Ambrosino P, Esposito F, Fogliano V. Antioxidative activity and carotenoid and tomatine contents in different typologies of fresh consumption tomatoes. *J. Agric. Food Chem*. 2000;48(10):4723–4727. DOI: 10.1021/jf000225t
6. BBS. Bangladesh Bureau of Statistics. Yearbook of Agricultural Statistics of Bangladesh-2017. Ministry of Planning, Government of the People's Republic of Bangladesh. 2018.
7. Tripathi K, Pandey S, Malik M, Kaul T. Fruit ripening of climacteric and non climacteric fruit. *Journal of Environmental and Applied Bioresearch*. 2016;4(1):27-34.
8. Khudairi AK. The ripening of tomatoes: A molecular ecological approach to the physiology of fruit ripening. *J Amer Sci*. 1972;60(6):696-707.

9. Hakim MA, Huq AKO, Alam MA, Khatib A, Saha BK, Haque FKM, et al. Role of health hazardous ethephone in nutritive values of selected pineapple, banana and tomato. *Journal of Food, Agriculture and Environment*. 2012;10(2):247-251.
10. Sarma PK. Pre-Scaling up of improved finger millet technologies: Postharvest losses of tomato: a value chain context of Bangladesh. *International Journal of Agricultural Education and Extension*. 2018;4(1):85-92.
11. Hassan MK, Chowdhury BLD, Akhtar N. Post harvest loss assessment: A study to formulate policy for loss reduction of fruits and vegetables and socioeconomic uplift of the stakeholders. Final report (PR 8/08). 2010;8:106-185.
12. Rahman AU, Chowdhury FR, Alam MB. Artificial ripening: What we are eating. *Journal of Medicine*. 2008;9(1):42-44. DOI: 10.3329/jom.v9i1.1425.
13. Kumari P, Douhan S, Bala S, Kumar J. Effect of ethylene and calcium carbide on ripening of mango (*Mangifera indica*) during storage at ambient temperature. *The Bioscan*. 2016;11(3):1441-1443.
14. Moniruzzaman M, Khatoun R, Hossain MFB, Rahman MT, Alam SN. Influence of ethephon on ripening and quality of winter tomato fruit harvested at different maturity stages. *Bangladesh Journal of Agricultural resources*. 2015;40(4):567-580.
15. Rangana S. *Handbook of Analysis and Quality Control for Fruits and Vegetable Products*. 2nd ed. New Delhi: McGraw Hill Publishing Company Limited; 1991.
16. AOAC. *Official Methods of Analysis of the Association of Official Analytical Chemist*. Washington D.C., USA. 20th ed; 2016.
17. Rahman MM, Khan MMR, Hosain MM. Analysis of vitamin C (ascorbic acid) contents in various fruits and vegetables by UV-spectrophotometry. *Bangladesh Journal of science and Industrial Research*. 2007;42(4):417-424.
18. Karnjanawipagul P, Nittayanuntawech W, Rojsanga PL, Suntornsuk L. Analysis of β -carotene in carrot by spectrophotometry. *Journal of Pharmaceutical Science*. 2010;37(1-2):8-12.
19. Chekri R, Noël L, Vastel C, Millour S, Kadar A, Guérin T. Determination of calcium, magnesium, sodium, and potassium in foodstuffs by using a microsampling flame atomic absorption spectrometric method after closed-vessel microwave digestion: method validation. *Journal of AOAC International*. 2010;93(6):1888-1896.
20. Yang SF. Regulation of ethylene biosynthesis. *Hort Sci*. 1980;15:238-243.
21. Mann SS, Singh B. Effect of ethrel on ripening of fruits of Patharnakh pear harvested on different dates. *Acta Hort*. 1990;279(59):529-532.
DOI:10.17660/ActaHortic.1990.279.59
22. Yah ARC, Novelo SAG, Cortes JAT, Argumedu JJ, Duch ES. The effect of ethephon on the colour, composition and quality of mango (*Mangifera indica*, cv Kent). *J Food Sci Technol*. 1998;4(3):199-205.
23. Kulkarni SG, Kudachi VB, Vasantha MS, Keshava MN, Aravinda B, Ramana KVR. Studies on effect of ethrel dip treatment on the ripening behavior of mango (*Mangifera indica* L.) variety 'Neelum'. *J Food Sci Technol*. 2004;41(2): 216-220.
24. Bondad ND, Pantastico EB. Ethrel induced ripening of immature and mature green tomato fruits. *Econ Bot*. 1972;26(3):238-244.
25. Smith RB, Loughheed EC, Franklin EW, McMillan I. The starch iodine test for determining stage of maturation in apples. *Can J Plant Sci*. 1979;59(3):725-735.
26. Anthon GE, LeStrange M, Barretta DM. Changes in pH, acids, sugars and other quality parameters during extended vine holding of ripe processing tomatoes. *Journal of the Science of Food and Agriculture*. 2011;91(7):1175-1181.

27. Moneruzzaman KM, Hossain ABMS, Sani W, Safiuddin M. Effect of stages of maturity and ripening conditions on the biochemical characteristics of tomato. *American Journal of Biochemistry and Biotechnology*. 2008;4(4):336-344.
DOI: 10.3844/ajbbbsp.2008.336.344
28. Helyes LJ, Dimeny Z, Lugasi A. Effect of maturity stage on content, colour and quality of tomato (*Lycopersicon lycopersicum* (L.) (Karsten) fruit. *International Journal of Horticultural Science*. 2006;12(1):41-44.
29. Rhodes MJC, Wooltorton LSC, Galliard T, Hulme AC. Metabolic changes in excised fruit tissue I. Factor affecting the development of a malate decarboxylation system during the ageing of disks of pre-climacteric apples. *Phytochem*. 1968;7(9): 1439-1451.
30. Khorshidi S, Davarynejad GH. Influence of preharvest ethephon spray on fruit quality and chemical attributes of 'Cigany' sour cherry cultivar. *Journal of Biological and Environmental Sciences*. 2010;4(12):133-141.
31. Rabaya T, Samad MA, Abedin MZ, Sikder MA, Hossain MI, Zubair AM. Study of artificial ripening agent and its effects on banana (*Musa* spp.) collected from Tangail area, Bangladesh. *IOSR Journal of Environmental Science, Toxicology and Food Technology*. 2017;11(9):14-19.
DOI: 10.9790/2402-1109011419
32. Mahmood T, Iftekhhar S, Humera A, Iffat M. Comparative study to evaluate the effect of calcium carbide (CaC₂) as an artificial ripening agent on shelf life, physio-chemical properties, iron containment and quality of *prunus persica* l. Batsch. *European academic research*. 2012;1(5):685-700.