International Journal for Research in Applied Sciences and Biotechnology

ISSN: 2349-8889
Volume-7, Issue-3 (May 2020)

www.ijrasb.com

https://doi.org/10.31033/ijrasb.7.3.7

To Study the Ripening Process of Tomato Using Ethanol

Mayank Chauhan[1] and Shivangi Chauhan[2]
[1]Student, Department of Biotechnology, Amity University Noida, Banasthali University Jaipur, INDIA
[2]Student, Department of Biotechnology, Amity University Noida, Banasthali University Jaipur, INDIA

[1]Corresponding Author: mayankchauhan111@gmail.com

ABSTRACT

After the harvesting of tomato from the field the samples are stored in the room temperature (26.84°C±3.0°C) some sample kept as untreated samples around 4 to 5 and some samples are to be treated and are kept to the other side and treatment is done by ethanol around 2 to 5 ml per kg and after treatment in sample slightly less weight decrease in treated sample as compare to untreated samples and titrable acidity also decrease very slowly in treated sample as compare to the untreated samples. Some of the test is performed like chlorophyll test, carotenoids, titrable acidity, and weight of samples and shelf life of untreated is 6- 8 days and treated fruit shelf life is around 12 days after that decaying of the samples.

Keywords-- Ethylene, Untreated, Treated, Tomato, Ripening, Physiochemical Post-Harvestetc

I. INTRODUCTION

Tomato is the highly edible crop around the world it is one of the top most producing crop in the world and tomato cover largest area for its production and cultivation of its crop and India, United States and China. Around 130 million tonnes tomato produced annually 20 million tonnes are wasted or loosed every year due to post harvest losses. India it is Second largest after the production of onion, potato and sweet potato. [1] Tomato can be produced where moisture content is high and fertile soil should be present. It is the annual crop that is produced throughout the year and Tomato has a very high source of vitamin A, carbohydrate and some amount of protein etc.

Tomato is a climacteric fruit its shelf life is less around 8 -10 days and it contains 95% of water. Its demand is also high all around the world and due to its perishable nature it also has large post-harvest losses in developing countries. During ripening there are some physical and Bio-Chemical changes occur like change in colour, change in weight, change in texture cell wall, starch, organic acids, pigments, ascorbic acids, amino acid, protein, respiration, transpiration, etc. Ripening leads to loss chlorophyll and carotenoids etc. [2] To Stop ripening we can use technologies like freezing, lower temperature stop all the metabolic activities occur in tomato.

The samples are divided into two parts control or untreated and treated and fruit are divided into equal parts, one part that is untreated fruit is kept for observation and other is for treatment. Treatment is given by injection ethanol 95%v/v (around 2-5 ml) per tomato and is injected on the head of tomato and it is spread slowly in the whole part and which help in delaying ripening of fruit and after treatment all the metabolic activities will delay like respiration, transpiration, ethylene biosynthesis, synthesis of carotenoids and change in chlorophyll content etc and condition required are 21-22^0 C and relative humidity is around 75 % and they are kept in a container or box for the storage. [3] Ethylene is a natural plant hormone and ethylene initiates the ripening process for the unripe fruit green colour due to the chlorophyll by the enzyme hydrolase and after ripening of fruit red colour is due the chemical cause is anthocyanin and for unripe fruit hard surface of fruit and chemical cause is pectin and by enzyme pectinase. [5] and after ripening fruit become soft and due to the less presence of less pectin and sour taste of unripe fruit by enzyme kinase and there are various enzyme's which lead to ripening and softening is due to degradation of pectin by pectinase there are various pectinase like PMG, PG, PE, PL etc. and in apple, tomato hydrolysis of galactans to galactose by β-galactosidase.

After treatment by ethanol injection chlorophyll content will slow degradation of chlorophyll and slow synthesis of carotenoids but in untreated or control fruit fast degradation of chlorophyll. In untreated fruit, ethylene biosynthesis occurs in which S-adinosylmethyl iodine (SAM) lead to formation of 1-Amino-cyclopropane-1-carboxylic acid (ACC) in the presence of ACC synthase and ACC lead to formation of Ethylene in the presence of ACC oxidase. Loss of chlorophyll is due to synthesis of carotenoids and colour change from green to red in many climacteric fruits. [1]

Ripening is natural process after ripening fruit become fleshy and sweet in taste, texture will be soft and the nutritional quality of fruit will increase which is important in our diet and there are many climacteric fruits like tomato, banana, mango, avocado, plum, papaya, kiwi fruit and some non-climacteric fruits like grape, lemon, melon, grapefruit, orange, strawberry watermelon, etc. There are many ripening agents like calcium carbide, ethephon, ethylene glycol, ethylene etc. Fruits can be exposed with ripening agents to promote ripening when the fruits are plucked from trees before ripening. Ripening can be controlled by storing at lower

ISSN: 2349-8889
Volume-7, Issue-3 (May 2020)
https://doi.org/10.31033/ijrasb.7.3.7

temperature immediately after plucking of fruit to stop the metabolic activities and reduce the respiration rate and ethylene production and we can use some chemical like potassium permanganate, which is good ethylene absorber and coated with newspaper as the packing materials.

These types of treated fruit are easily supplied in the colder region where temperature is very less in zero degree or less. In these condition alcohol is essential to keep the body warm for the survival in these type of areas and supply of food is also very difficult and when the food is being supplied if the alcohol is present in these fruit then both the requirement will get completed and ripening also get delayed in the colder areas so these type of food will run for around 10- 15 days.[2] Commercial availability of These type of treated fruit can be easily available in a local market or big stores and they are similar in appearance like other untreated fruit. In the treated fruit there is no effect in the nutrition and presence of minerals will vary but very less as the shelf life of the fruit increase to some days because ethanol will increase the shelf life of the fruit but not affect its nutritional properties and treatment will increase its shelf life.

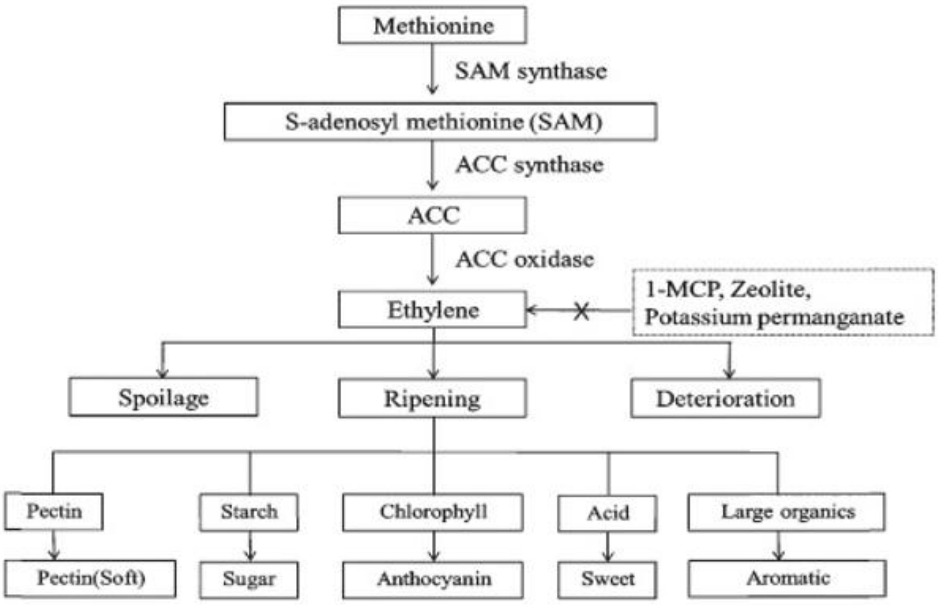


Figure 1: Ethylene Biosynthesis

II. MATERIAL AND METHOD

2. Plant Source and Treatment

The plant source (tomato) is collected from the local market and the ethanol is collected from the chemistry lab of amity university, Noida (UP). Tomato is divided into two parts treated and untreated and in treatment is done by ethanol 95%v/v injection and untreated tomato there is no such treatment done. [1] After the treatment we observe the colour change, change in weight, change in texture, and chemical characteristics -change in chlorophyll, test for starch, test for polysaccharide etc.

2.1: Quality Evaluation

2.1.1. Titrable Acidity

To measure the Titrable acidity of tomato take 50 ml of clear juice by cutting fruits, pressing with a hand press and filter it through cheese cloth and Make sure samples are at room temperature. [2] Weigh out 6g of juice into a 100 ml beaker Add 50ml distilled water then add 2 drops of phenolphthalein and titrate with 0.1N NaOH.

Use titration Equation,

$$\mathbf{N1V1 = N2V2}$$

$$\textbf{Titrable Acidity } (\%\textbf{acid}) = (-\boldsymbol{b}(\textbf{ml of NaOH used} * \mathbf{0.1} * \mathbf{100} * \mathbf{0.067}))/(\textbf{g of fruit juice})$$

2.1.2. Estimation of Chlorophyll in the Peel

To calculate the amount of chlorophyll in the tomato take 8g of fruit peel grounded in 15ml absolute ethanol and Centrifuge at 5000rpm for 1 min to remove debris and Divide supernatant into 3 equal volumes in different tube observe the absorbance at 480 nm, 645 nm, 663nm while using the spectrophotometer Use absolute ethanol as blank.

ISSN: 2349-8889
Volume-7, Issue-3 (May 2020)

https://doi.org/10.31033/ijrasb.7.3.7

2.1.3. To Measure Weight during Ripening of Fruit

Weigh each sample of treated and untreated one by one every day and observe the change in weight of the sample during ripening sample weight may increase or decrease. The weight of tomato is calculated by using the formula:

$$\textbf{Percent Weight Loss} = ((\textbf{Initial weight} - \textbf{Weight at the day of observation})/\textbf{Initial weight} \times \textbf{100})$$

2.1.4. Test for Carotenoids

To calculate the amount of **carotenoids** in the tomato take 8g of fruit peel grounded in 15ml absolute ethanol and Centrifuge at 5000rpm for 1 min to remove debris and Divide supernatant into 3 equal volumes in different tube observe the absorbance at 480 nm, 645 nm, 663nm while using the spectrophotometer Use absolute ethanol as blank and to calculate amount of carotenoids in the peel by using formula:

$$\mathbf{C(x + c) = (1000A470 - 2.13Ca- - 97.63Cb)/209}$$

III. RESULT AND DISCUSSION

3.1: Weight Loss

For Treated: After treatment by ethanol injection 95%v/v weight of fruit will decrease day to day and it will slow down all the metabolic activities(respiration) and transpiration of the fruit and plant hormone ethylene will effect ripening of fruit. For the treated tomato day 0 weigh is T1=41.52g, T2=27.87g, For day2 T3=39.6g, T4=26.53g, For day4 T5=38.6g, T6=25.41g.

For Untreated: For the untreated tomato as treatment is given to them they kept in the same environment as treated fruit are kept but the weight will decrease fast as compared to the treated tomato and rate of ripening(respiration) is also fast. For the untreated tomato day0 weigh is UT1=27.62g, UT2=42.45g, for day2 UT3=25.85g, UT4=40.62g, day6 UT5=24.47g, UT6=38.96g. Transpiration and respiration can be reason of high weight loss.

Table 1: Change of Weight of Tomato in Different Days

	DAY 0		DAY 2		DAY 4		DAY 6		DAY 8		DAY 10		DAY 12	
	CT	E.TR	CT	E.TR	CT	E.TR	CT	E.TR	CT	E.TR	CT	E.TR	CT	E.TR
S.NO	WEIGHT	WEIGHT	WEIGHT	WEIGHT	WEIGHT	WEIGHT	WEIGHT	WEIGHT	WEIGHT	WEIGHT	WEIGHT	WEIGHT	WEIGHT	WEIGHT
1	27.62 g	41.25 g	25.85 g	39.6 g	24.47 g	38.00 g	23.04 g	36.49 g	22.6 g	35.66 g	21.54 g	34.63 g	19.25g	32.56g
2	42.45 g	27.87 g	40.62 g	26.53 g	38.96 g	25.41 g	37.29 g	24.21 g	36.71 g	23.67 g	35.52g	22.84 g	33.25g	20.54g

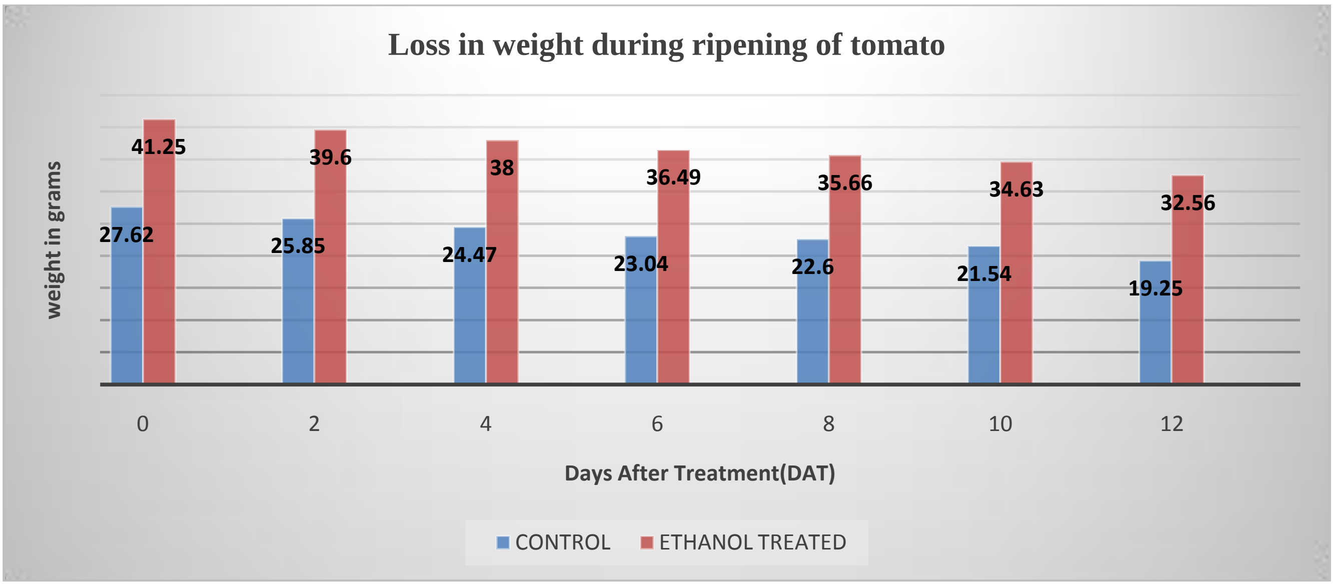


Figure 2: Graphical Representation of Loss in Weight

ISSN: 2349-8889
Volume-7, Issue-3 (May 2020)

https://doi.org/10.31033/ijrasb.7.3.7

3.2: Titarable Acidity

For Treated: Titarable acidity is used to determine acid present in the solution and it is expressed gram /litre (g/l) and it is obtained by multiplying by 10 to percent TA for the treated tomato when it is treated with ethanol injection and to measure the titrable acidity of the tomato it is titrated with the 0.01N NaOH using the phenolphthalein(2 drop) as an indicator the titrable acidity of tomato will decrease at slow rate this will improve the shelf life of the tomato and as the ripening follows in later stages sugar and acid ratio will vary as the ripening proceed in the later stages, while the ripening occurs high acid content present in tomato will get degraded by enzyme kinase and sugar content will increase its amount in the tomato and then it is measured by titration method by using the strong base NaOH.

For Untreated: As there is no treatment given in this method so the to measure the titrable acidity of the tomato it is again treated with 0.01N NaOH using phenolphthalein as an indicator to measure the titrable acidity will decrease at the faster rate because all the metabolic activities will occur from unripe stage to fully ripened stage.

Calculations: Use titration Equation,

$$N1V1 = N2V2$$

$$\textbf{Titratable Acidit y}(\%\textbf{acid}) = (\textbf{ml of NaOH used} * 0.1 * 100 * 0.067)/\textbf{g of fruit juice}$$

Table 2: Titrable Acidity Data of Tomato in Different Days

	DAY 0		DAY 3		DAY 6		DAY 9		DAY 12	
	UNT	TR	UNT	TR	UNT	TR	UNT	TR	UNT	TR
SR. NO	NAOH USED	NAOH USED	NAOH USED	NAOH USED	NAOH USED	NAOH USED	NAOH USED	NAOH USED	NAOH USED	NAOH USED
1.	5.3	4.75	3.4	4.25	3.15	4.05	3.05	3.95	2.94	3.84
TA%	0.59%	0.53%	0.38%	0.47%	0.35%	0.45%	0.30	0.42%	0.24	0.39%

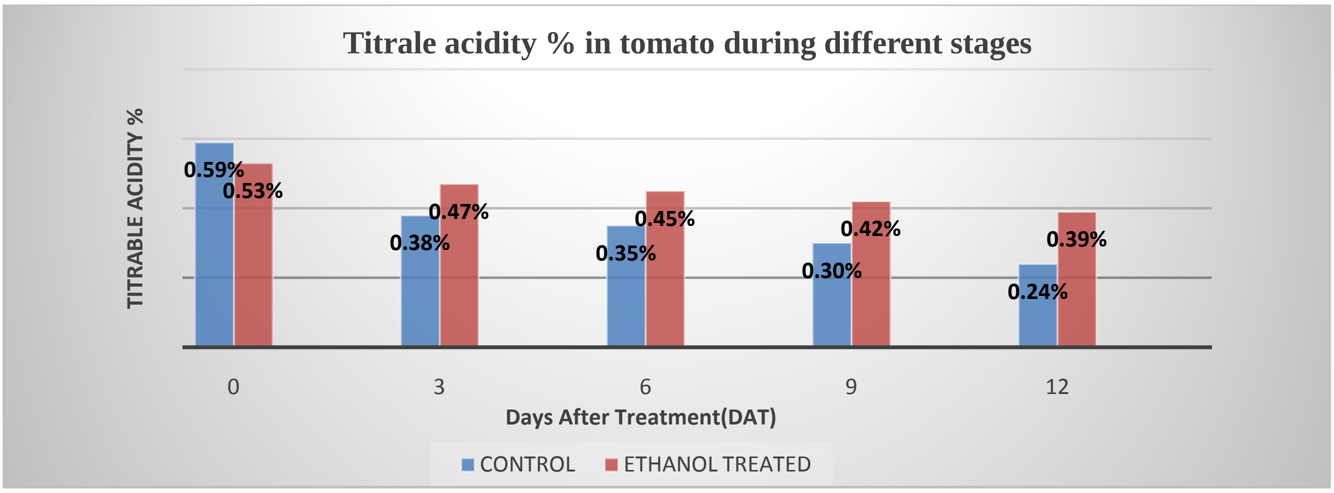


Figure 3: Graphical Representation of Titrable Acidity in Tomato

3.3: Chlorophyll

For Treated: After the treatment with ethanol in the tomato its chlorophyll content is measured by taking the absorbance at different wavelength 480nm,645nm,663nm optical density(OD) is recorded by an instrument called spectrophotometer. Chlorophyll a and chlorophyll b is calculated by the formula. At the 0 day at stage 1 the total chlorophyll content is (3.33) highest for the tomato and the chlorophyll at stage 2 of the tomato at half ripe stage is (2.55) and for the stage 3 of tomato is 2.18 which is ripened stage. [7] This type of treatment will increase the shelf life of fruit and it is easy for the long storage of tomato and this will reduce the post-harvest losses and this treatment does not have any side effect after it is being consumed. It was observed that treated tomato somehow showed slow degradation of chlorophyll and slow synthesis of carotenoids [12].

For Untreated: Pigment in unripe stage of tomato consists of chlorophyll a, chlorophyll b and carotenoids and we can estimate these pigments by spectrophoto metric techniques as observed for the treated tomato by taking the absorbance at different wavelength 480nm, 645nm, 663nm optical density (OD) is recorded by an instrument called spectrophotometer and the chlorophyll

ISSN: 2349-8889
Volume-7, Issue-3 (May 2020)

https://doi.org/10.31033/ijrasb.7.3.7

a and chlorophyll b is calculated by the given formula. [4] For the untreated tomato, ethylene breaks the chlorophyll and convert into anthocyanin and chlorophyll degradation can be observed peel of tomato from green to red in colour from unripen stage to fully mature ripened stage and in untreated tomato chlorophyll and carotenoids degradation was much rapid.

$$Chl - a = 13.36A664 - 5.19\ A649$$
$$Chl - b = 27.43A649 - 8.12\ A664$$

Spectrophotometric data of various stages of ripening is summed below and a respective graph is plotted.

Table 3: Chlorophyll Data of Tomato in Different Days

		Day 0		Day3		Day6		Day9		Day12	
		UNT	TR	UNT	TR	UNT	TR	UNT	TR	UNT	TR
S.NO	WAVE LENGTH	O.D.	O.D.	O.D.	O.D.	O.D.	O.D.	O.D.	O.D.	O.D.	O.D.
1	480nm	0.195	0.19	0.264	0.22	0.28	0.15	0.30	0.10	0.31	0.05
2	645nm	0.092	0.105	0.035	0.093	0.020	0.08	0.011	0.06	0.05	0.04
3	663nm	0.089	0.151	0.061	0.083	0.040	0.07	0.031	0.05	0.022	0.03
Chla :		0.88	1.64	0.68	0.80	0.48	0.67	0.36	0.47	0.14	0.27
Chlb :		1.69	1.70	0.52	1.74	0.18	1.50	0.10	1.13	1.04	0.77
Total Chl:		2.57	3.33	1.20	2.55	0.28	2.18	0.46	1.60	0.18	1.04
Carotenoid		0.13	0.10	1.02	0.24	1.25	0.25	1.39	0.17	0.99	0.14

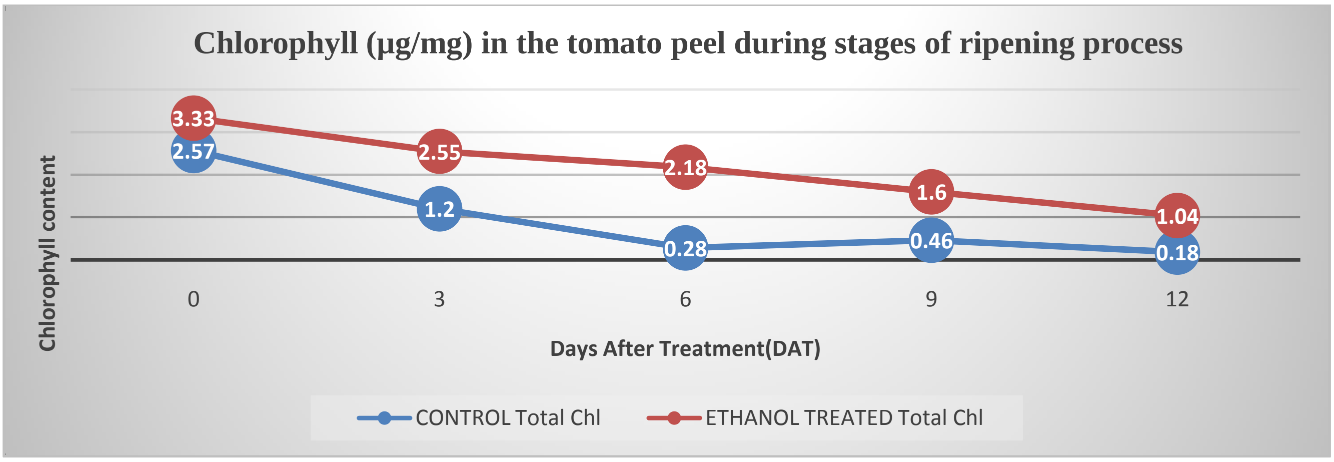


Figure 4: Graphical Representation of Chlorophyll Degradation in Chlorophyll

3.4: Change in Colour

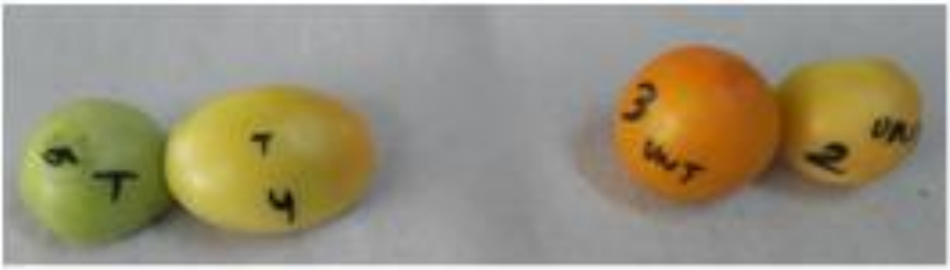
Change in colour is due to the to the degradation of chlorophyll in tomato peel and lead to synthesis of carotenes. the colour of mature unripe is green, colour of half ripe is yellowish to orange and mature ripe is red in colour and tomato become juicy. lightness will be going to decrease as the storage time increase. The observation of change in colour of tomato can be seen in figures. [17]

Day 1

All mature green tomatoes

ISSN: 2349-8889
Volume-7, Issue-3 (May 2020)
https://doi.org/10.31033/ijrasb.7.3.7

Day 3



Colour of untreated tomato starts changing

Day 5

Untreated tomato show fast coloration as compared to treated tomato while treated tomato are still green

Day 8

Even on the eight day starting from Day 5, a huge difference in colour of tomato is visible.

Day 12

Even after twelve days treated tomato are eatable but untreated tomato start decaying a false smell occur into the tomato

3.5: Carotenoids

Results of carotenoids in treated and control or untreated samples are listed in table. carotenoids are observed at each stage of tomato. In the Untreated tomato carotenoids will increase at faster rate as compare to the treated tomato due to the treatment of ethanol it effects transpiration and respiration and increase of carotenoids due to breakdown of chlorophyll and lead to the formation of carotenoids .as the level of chlorophyll will decrease in each stage will ultimately lead to increase of carotenoids. [8] The range of untreated tomato of carotenoids are 0.3 to 1.25 and for treated tomato is 0.10 to 0.25. Carotenoids can be calculated by this formula-

$$\boldsymbol{C(x+c) = (1000A470 - 2.13Ca - - 97.63Cb)/209}$$

$$\boldsymbol{Lycopene\ (mg/100\ mL) = -0.0458A663 + 0.204A645 + 0.372A505 - 0.0806A453}$$

ISSN: 2349-8889
Volume-7, Issue-3 (May 2020)
https://doi.org/10.31033/ijrasb.7.3.7

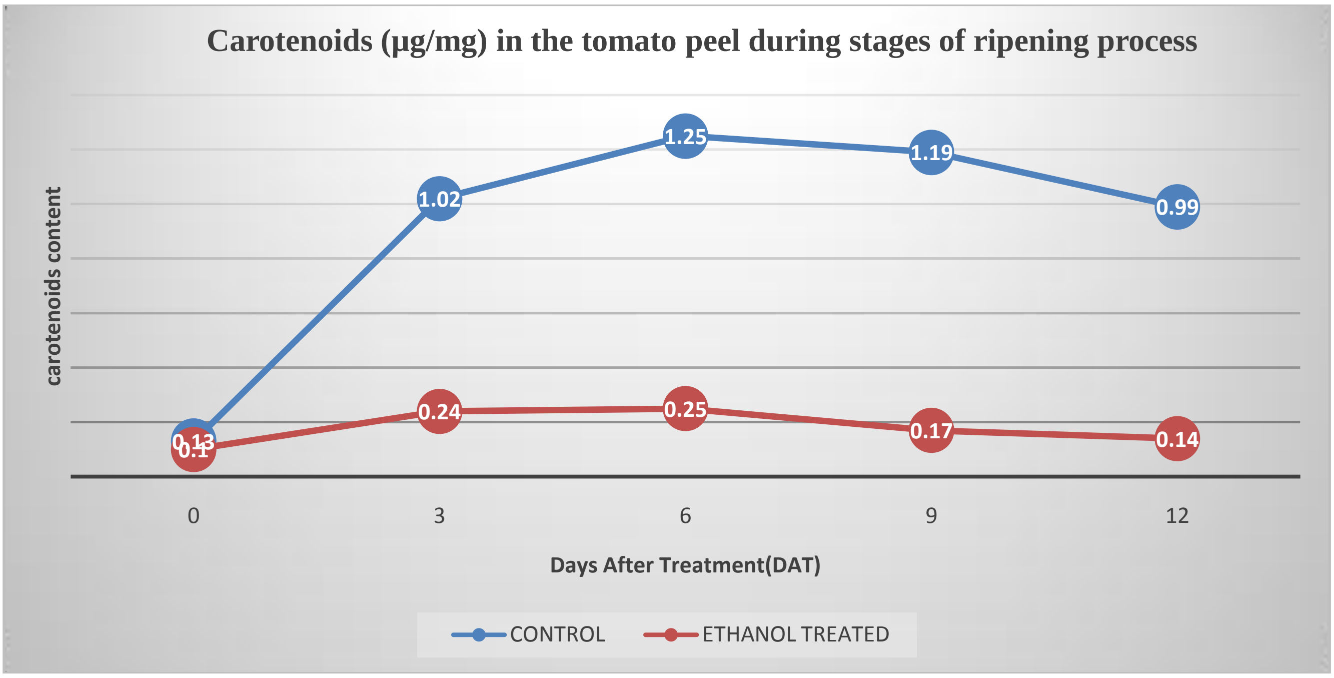


Figure 5: Graphical Representation of Carotenoids

IV. CONCLUSION

Ethanol clearly inhibited tomato fruit ripening in the presence of ethylene in this study. Clearly, the 3 and 4 mL treatments were not toxic since fruit eventually ripened without visible injury. The respiratory rate of tomato fruit exposed to 4 ml ethanol was significantly reduced. A possible explanation for the inhibition of tomato fruit ripening by ethanol is the inhibition of ethylene action. Ethanol may also regulate ethylene synthesis. Concentrations of ethanol accumulated in tomato fruit tissue either under anaerobic conditions or during exposure to ethanol vapour which were inhibitory to fruit ripening. Ethanol may, therefore, be an important regulator of fruit development, both in species in which it is synthesized under normal ripening and in fruits held in controlled atmosphere storage.

REFERENCES

[1] Khudairi AK (1972). The ripening of tomatoes. *J Amer Sci.*, 60, 696-707.
[2] Gonzalez J. (1999). Use of ethylene (ethephon) in uniform ripening of processing tomato (*Lycopersicon esculentum*) in California. *Acta Hort.*, 487, 179-182.
[3] Koskitalo DN, & Omrod DP. (1972). Effects of sub optimal ripening temperatures on the colour quality and pigment composition of tomato fruit. *J Food Sci.*, 37, 56-58.
[4] PFA. (2003). *Prevention of Food Adulteration Act 1954 Rules, In: International Law Book Co* (19th edn), Delhi, INDIA.
[5] Kasmire RF. (1981). Continuous flow ethylene gassing of tomatoes. California Tomatorama. *Fresh Market Tomato Advisory Board Information Bulletin* No.29.
[6] Cantwell M. (1994). Optimum procedures for ripening tomatoes. *Perishable Handling Newsletter. University of California*, Davis 80: 24-26.
[7] Sargent SA. (2005). Ripening and quality responses of tomato fruit to ethylene. *Postharvest Biol Technol.*, 36, 127-134.
[8] Bassetto E, Jacomino AP, Pinheiro AL, & Kluge RA. (2005). Delay of ripening of 'Pedro Sato' guava with 1-methylcyclopropene. *Postharvest Biol Technol.*, 35, 303-308. doi:10.1016/j.postharvbio.2004.08.003
[9] AOAC. (1990). *Official methods of analysis. (12thEdn) Association of Official Analytical Chemists*, Washington DC.
[10] Hunter S. (1975). *The measurement of appearance*. John Wiley and Sons. New York, USA.
[11] Singh S, Bansal ML, Singh TP, & Kumar R (1991). *Statistical Methods for Research Workers*. Kalyani Publishers, New Delhi, INDIA.
[12] Kretchman DW, Short TH. (1972). Studies on effect of ethephon on tomato cv. Napoli. *Scientia Hort.*, 65, 86-88.
[13] Mutton LL. (1978). Effect of moisture stress on the ethephon response in tomatoes. *Scientia Hort.*, 8, 299-305.
[14] Khedkar DM, More RM, Patil VK, Warke DC, & Ballal AL. (1981). *Studies on physico-chemical changes in banana during ripening. In: Symposium on recent advances in fruit development*, Ludhiana, INDIA.

ISSN: 2349-8889
Volume-7, Issue-3 (May 2020)

https://doi.org/10.31033/ijrasb.7.3.7

[15] Mahajan BVC, Kaur T, Gill MIS, Dhaliwal HS, Ghuman BS, et al. (2010). Studies on optimization of ripening techniques for banana. *J. Food Sci. Technol.*, *47*(3), 315-319.

[16] Brinston K, Dey PM, John MA, & Pridhan JB. (1988). Post harvest changes in *Mangiferaindica*L mesocarp walls and cytoplasmic polysaccharides. *Phytochem*, 27, 719-723.

[17] Solmos T, & Laties GG. (1973). Cellular organization and fruit ripening. *Nature*, 245, 390-391.

[18] Gonzalez J. (1998). Symposium on protected cultivation of solanacea in mild winter climates. *Acta Hort.*, 366, 105-107.

[19] Hicks JR, Mandez MJ, & Master JF. (1984). Influence of storage temperature and ethylene on firmness, acids and sugars of chilling sensitive and chilling tolerant tomato. *J. Amer. Soc. Hort. Sci.*, 109, 273

[20] C.S. Barry, & J.J. Giovannoni. (1997). Ethylene and Fruit Ripening. *J. Plant Growth Regul.*, *26*(2), 143–159.

[21] M.M.H. Senna, , K.M. Al-Shamrani, & A.S. Al-Arifi, (2014). Edible coating for shelf-life extension of fresh banana fruit based on gamma irradiated plasticized poly (vinyl alcohol)/carboxymethyl cellulose/tannin composites. *Mater. Sci. Appl.*, 5, 395–415.

[22] D.Mohapatra, S. Mishra, S. Giri, & A. Kar. (2013). Application of hurdles for extending the shelf life of fresh fruits. *Trends in Postharvest Technology.*, *1*(1), 37-54.

[23] J.H. Misir, F.M. Brishti, & M. Hoque. (2014). Aloe vera gel as a Novel Edible Coating for Fresh Fruits: A Review. *Am. J. Food Sci. Technol*., *2*(3), 93–97.

[24] A.Batu. (2004). Determination of acceptable firmness and colour values of tomatoes. *J. Food Eng. 61*(3), 471– 475.

[25] K.A.Athmaselvi, P. Sumitha, & B. Revathy. (2013). Development of aloe vera based edible coating for tomato. *Int. Agrophysics*, *27*(4), 369-375.

[26] R. Yulianingsih, D.M. Maharani, L.C. Hawa, L.Sholikhah, U.J. Ukpabi, U. Chijioke, E.N.A. Mbanaso, T. Sudargo, F.Z. Nisa, & S. Helmiyati. (2013). Physical quality observation of edible coating made from aloe vera on cantaloupe (Cucumismelo L.) minimally processed. *Pak. J. Nutr.*, *12*(9), 800–805.

[27] A.R. Vicente, M. Saladie, J.K.C. Rose & J.M. Labavitch. (2007). The linkage between cell wall metabolism27 and fruit softening: looking to the future. *J. Food Science and Agriculture, 87*(8), 1435-1448.

[28] Ankita Borah, Dr Komal Mathur, Dr GC Srivastava, & Mansi Agrawal. (2016). Effect of aloe vera gel coating and bagging of fruits in enhancing the shelf life of tomato. Int. j. innov. res. sci. eng. (Ghaziabad. Online), 2(6), 1-5.

[29] Joyanta Kumar Roy, Reza Akram, Md. Arshad Ferdowsh Shuvo, Habiba Khatun, Md. Shihabul Awal1, & Manobendro Sarker. (2017). Effect of ethanol vapor on ripening of tomato. *Agricultural Engineering International : The CIGR e-journal*, *19*(2), 168-175.