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COLORIMETRIC DETERMINATION OF VITAMIN C IN FRESH AND DILUTE FRUIT JUICES AND EFFECT OF THERMAL EXPOSURE ON CONCENTRATION AT VARIOUS STAGES

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ABSTRACT

Among all vitamins, vitamin C plays a crucial role in maintaining good health. In the present study ten types of fruits are analyzed to compare ascorbic acid levels and the effect of boiling on its quantity is also determined by colorimetric method .It is concluded that its levels are gradually decreased with increase of boiling time. Cheaply and mostly available fruits have higher concentration of ascorbic acid than other fruits. In Psidium guajana, Citrus aurantium and Phyllanthus emblica the concentration of vitamin C is more. It is concluded that the concentration of ascorbic acid is more in semi-ripened stage of the fruit than ripened and unripened stages of Citrus limon, Citrus aurantifolia and Psidium guajana. But it is more in unripened fruits of Citrus medica and Citrus aurantium. In Psidium cattleianum sabine and Psidium littorale ascorbic acid concentration is more in fully ripened stage.

KEYWORDS: ascorbic acid, fruits, colorimetry, ripening, boiling.

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INTRODUCTION

Proper health is the basic thing for survival of human life. Fruits protect us from many diseases. Citrus fruits contain major quantity of ascorbic acid[1], which is an essential nutrient[2] and structurally relates to glucose.It is a weak sugar acid[3,4], found in many vegetables and fruits. Vitamin C helps to make collagen, absorbs iron[5]and neutralizes free radicals.[6] It is used as food additive[7], works as a co-factor in several enzymatic reactions[8], fight against infectious and chronic diseases [9-13] and strengthens immune system .[14]Its quantity decides the nutritional quality of the fruit.[15,16] During thermal exposure its stability is lost.[17,18] Our body is unable to synthesize vitamin C[19] but it has the role in preventing oxidative damage to DNA[20,21] and proteins in our body .Deficiency of this water soluble[22] and antioxidant [23,24] vitamin causes scurvy [25,26] which is identified by bleeding gums, anaemia[27], fatigue, depression, mal wound healing[28] and scaly skin. Ripening is the process by which fruits attain their desirable flavor, quality, color, palatable nature and other textural properties. It is associated with change in conversion of starch to sugar. During this process the fruits emit ethylene along with increased rate of respiration.

MATERIALS AND METHODS

Sample collection

Lime, lemon, citron, sour orange, Indian goose berry, star goose berry, sweet lime, red guava, green guava, white guava fruits of unripened, semi ripened and fully ripened are collected from local fruit gardens of Madhira mandal of Khammam district during winter season.

Sample preparation

Fruits are washed thoroughly with distilled water, manually sliced and pulped using a blender. Sample is divided into six portions, weighing five grams each and labelled as samples A-E. All these samples are diluted and exposed to heat treatment at 50^0C for 5, 10, 15, 20, 25 and 30 minutes respectively. Then absorbance is measured for both fresh samples and diluted samples using colorimeter.

Analysis

The processed samples are subjected to the following analysis immediately after sample preparation for vitamin C determination. 20ml oxalic acid solution, 0.2ml of 0.01 % methylene blue solution and 1ml of acetate buffer of pH 4.2 are taken in a 50ml beaker. To this fresh and heated ripened sample at 50^0C of 1ml solution is added separately and measured for absorbance periodically after 5, 10,15,20,25 and 30 minutes of boiling. Absorbance of each solution is read using Roy instruments digital photo colorimeter (RI_506 C) of infra instruments private limited at wavelength 540nm. Repeated the same procedure for semi ripened and unripened fruits of diluted and fresh juices.

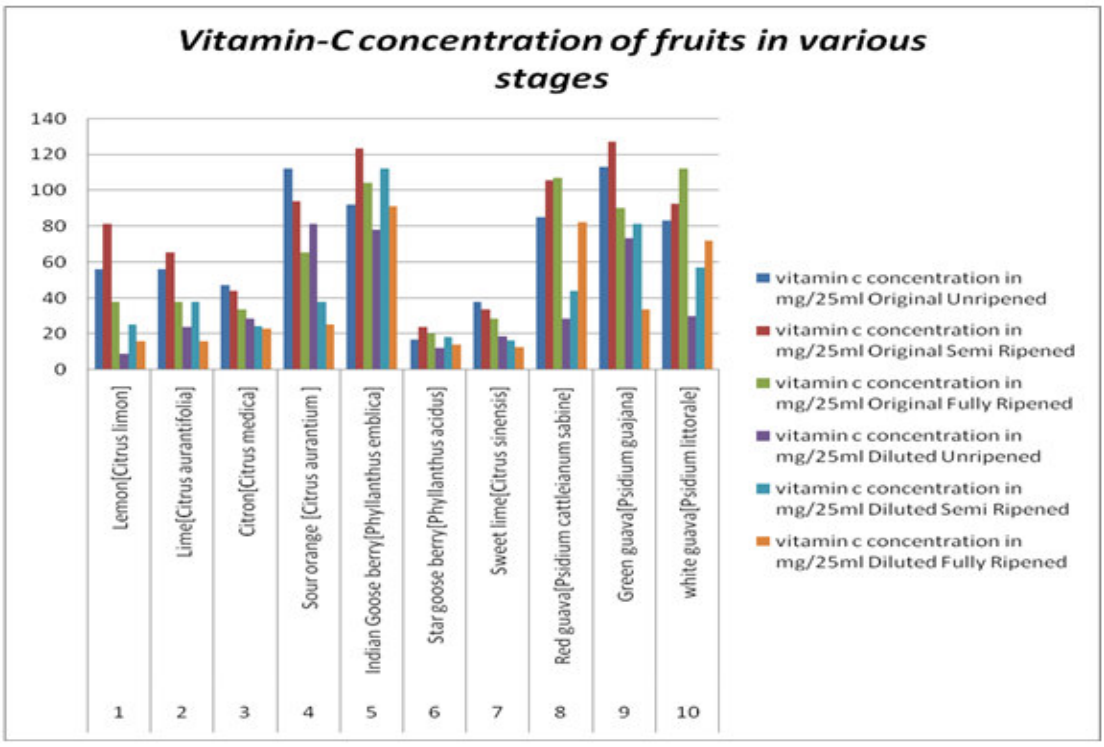
RESULTS AND DISCUSSION

In the calibration curve, the absorbances of standard vitamin C at various concentrations C are plotted. The concentration of vitamin C after boiling times of 5, 10, 15, 20, 25 and 30 minutes at 50^0C is plotted and it is found to be more in Psidium guajana, Citrus aurantium and Phyllanthus emblica. The concentration of original sample is prominent than that of boiled samples. The retention of vitamin C is often used as an indicator for the overall nutrient reduction of food products because it is highly sensitive to oxidation and leaching in water soluble media during heating. Higher temperatures does not retain vitamin C, causing it to leach out of the food into the water and also to be oxidized, first to dehydroascorbic acid and then to diketogulonic acid. This last compound has no vitamin C activity at all and is irreversible. It is better to maintain or store samples below the room temperature. Its concentration is decreased during fruit ripening in some fruits. Feeble vitamin C is easily deteriorated during boiling and finally depleted as the boiling time is increased. The present work revealed that the concentration of ascorbic acid is more in semi-ripening stage of the fruit than the ripened and unripened stages of lemon, lime, and green guava. It is noticed to be more in unripened fruits of sour orange (Fig 2) and citron (Fig 1). In fully ripened fruits of red guava (Fig 5) and white guava its quantity is more. So fruits should be consumed before processing to attain more concentration of ascorbic acid. It is diminished more in first 5 and 10 minutes of boiling time, and there is a regular decrease afterwards. In some fruits after periodical heating up to 30 minutes its concentration is completely reduced to zero. Dilution of heated samples causes further decrease of its quantity. The vitamin C concentration in following fruits is as follows: Green Guava > Indian Goose Berry > White Guava = Sour Orange > Red Guava > Lemon > Lime > Citron > Sweet Lime > Star Gooseberry.(Graph 4 to graph 24).

Table 1
Vitamin-C concentration of fruits in various stages

S no	Sample	vitamin c concentration in mg/25ml					
		Original			Diluted		
		Unripened	Semi Ripened	Fully Ripened	Unripened	Semi Ripened	Fully Ripened
1	Lemon[Citrus limon]	56	81.1	37.8	8.9	25	15.7
2	Lime[Citrus aurantifolia]	56	65.5	37.8	24	37.8	16
3	Citron[Citrus medica]	47	43.7	33.4	28.4	24.3	22.7
4	Sour orange [Citrus aurantium]	112	93.7	65.4	81.1	37.8	25.4
5	Indian Goose berry[Phyllanthus emblica]	92	123	104	78	112	91
6	Star goose berry[Phyllanthus acidus]	17	24	20	12	18	14
7	Sweet lime[Citrus sinensis]	37.8	33.4	28.4	18.9	16.2	12.6
8	Red guava[Psidium cattleianum sabine]	85	105.5	107	28.4	43.7	82
9	Green guava[Psidium guajana]	113	127	90	73.4	81.1	33.4
10	white guava[Psidium littorale]	83.2	92.3	112	29.7	57	72

Graph 1
Vitamin-C concentration of fruits in various stages



Bars in the graph represent the absorbances of fruits in original and dilute samples in the three ripening stages of fruits

Table 2
Vitamin-C concentration in original and diluted juices of fruits at various processing times

FRUIT	FRUIT STAGE	SAMPLE CODE	PROCESSING TIME(min)	mg/25ml	
				ORIGINAL	DILUTED
LEMON	UNRIPENED	A	5	17.5	9
		B	10	15.39	7
		C	15	15.31	6
		D	20	9.8	5
		E	25	6.9	4
		F	30	5	3
	SEMI RIPENED	A	5	56	14.2
		B	10	24	10.3
		C	15	23.5	7.8
		D	20	17.5	6.5
		E	25	16.45	5.8
		F	30	10.78	4
	RIPENED	A	5	14.2	4.9
		B	10	11.1	4.3
		C	15	7.5	4
		D	20	6.7	0
		E	25	5.7	0
		F	30	4.5	0
LIME	UNRIPENED	A	5	33.4	22.7
		B	10	24	16
		C	15	18.9	8.9
		D	20	16.3	6.2
		E	25	14	5.2
		F	30	11.2	4.4
	SEMI RIPENED	A	5	43.7	23.2
		B	10	33.4	17.5
		C	15	25.8	14.5
		D	20	24	12.4
		E	25	22.7	10.33
		F	30	18.9	8
	RIPENED	A	5	24	14.5
		B	10	22.7	11.4
		C	15	18.9	10
		D	20	16.4	6
		E	25	12.6	4.2
		F	30	11.1	3.9
SOUR ORANGE	UNRIPENED	A	5	63	29
		B	10	43.7	22
		C	15	22.7	17

		D	20	16.5	12
		E	25	16.2	9
		F	30	14	6.7
	SEMI RIPENED	A	5	43.7	24
		B	10	33.4	20
		C	15	25.8	15.6
		D	20	23	10.33
		E	25	18.9	7.8
		F	30	14.2	6
	RIPENED	A	5	28.4	20
		B	10	25.8	17.7
		C	15	22.7	16.3
		D	20	19	13
		E	25	16.2	8
		F	30	8.7	4.9
CITRON	UNRIPENED	A	5	43.7	24
		B	10	33.4	23.5
		C	15	25.8	14.2
		D	20	23	11.8
		E	25	22.7	10.33
		F	30	18.9	7.9
	SEMI RIPENED	A	5	33.4	22.8
		B	10	25.8	16.8
		C	15	18.2	13.8
		D	20	17.5	9
		E	25	16	7.5
		F	30	10	4.9
	RIPENED	A	5	25	18.2
		B	10	22.7	15.4
		C	15	17.9	13.8
		D	20	16	9
		E	25	12	6.9
		F	30	10.33	4.9
INDIAN GOOSE BERRY	UNRIPENED	A	5	72.5	52.4
		B	10	63.4	43.2
		C	15	52.3	32
		D	20	42	24
		E	25	35	14
		F	30	23	12
	SEMIRIPENED	A	5	97.4	62.3
		B	10	77.4	52.4
		C	15	65.8	37.8
		D	20	54	25.7
		E	25	45.3	20
		F	30	32	16
	RIPENED	A	5	82.3	60.4
		B	10	75	50.7
		C	15	62	33
		D	20	53	26
		E	25	42.5	16.5
		F	30	29.7	14
STARGOOSE BERRY	UNRIPENED	A	5	15.6	10.7
		B	10	14.2	8.4
		C	15	12.6	6.8
		D	20	8.7	4.5
		E	25	5.7	0
		F	30	4.5	0
	SEMIRIPENED	A	5	20.2	16.4
		B	10	17.2	14.7
		C	15	13	9
		D	20	9.5	6
		E	25	8.9	3
		F	30	6	0
	RIPENED	A	5	18	12.7
		B	10	16	10.9
		C	15	13	8
		D	20	9.5	5
		E	25	9	0
		F	30	5	0
SWEET LIME	UNRIPENED	A	5	33.4	17.4
		B	10	28	16.3
		C	15	24	14.7
		D	20	17	10.33
		E	25	15	8.7
		F	30	9	5.2
	SEMIRIPENED	A	5	24	8.7
		B	10	20	7.6
		C	15	17.2	6
		D	20	13.5	4.9
		E	25	8.9	0
		F	30	8	0
	RIPENED	A	5	30	13.2
		B	10	28	9.5

		C	15	23.5	8.7
		D	20	16.4	7.6
		E	25	14	6.3
		F	30	8.9	4.9
RED GUAVA	UNRIPENED	A	5	55	38
		B	10	34	23
		C	15	28.9	18.7
		D	20	16.2	7.2
		E	25	13	6
		F	30	10	5
	SEMIRIPENED	A	5	68.4	48.3
		B	10	34	25
		C	15	20	19
		D	20	16.2	8.7
		E	25	13	7.1
		F	30	10.33	4.9
	RIPENED	A	5	81	54
		B	10	62.7	44.4
		C	15	48.9	24.8
		D	20	26.6	17
		E	25	16.2	11
		F	30	12	6
GREEN GUAVA	UNRIPENED	A	5	56	28.4
		B	10	32.7	15.2
		C	15	18	12.6
		D	20	15.6	8.7
		E	25	15	7.9
		F	30	12.6	4.9
	SEMI RIPENED	A	5	66	33.4
		B	10	43.7	25
		C	15	25.8	20
		D	20	25	17
		E	25	24	15
		F	30	22.7	11
	RIPENED	A	5	43.7	19
		B	10	24	15.2
		C	15	17.5	11.4
		D	20	15	9.2
		E	25	13.8	6.2
		F	30	11.2	4.9
WHITE GUAVA	UNRIPENED	A	5	35	28
		B	10	22.7	16.3
		C	15	18.9	14
		D	20	17.51	11.5
		E	25	14	7.1
		F	30	9.5	6
	SEMI RIPENED	A	5	48.4	25
		B	10	25.8	12
		C	15	18.2	10
		D	20	18	8.7
		E	25	14.2	8
		F	30	11.3	7.1
	RIPENED	A	5	65.8	48.2
		B	10	42.4	37.51
		C	15	32.7	24.9
		D	20	18.9	15
		E	25	17.51	8.5
		F	30	14.2	9

Figure 1
Citron [Citrus medica]

Figure 2
Sour orange [Citrus aurantium]

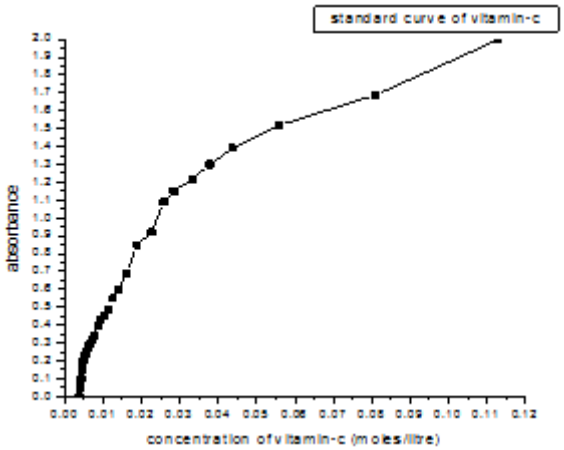
Figure 3
Citron [Citrus medica]

Figure 4
Sweet lime [Citrus sinensis]

Figure 5
Red guava [Psidium cattleianum sabine] grinded

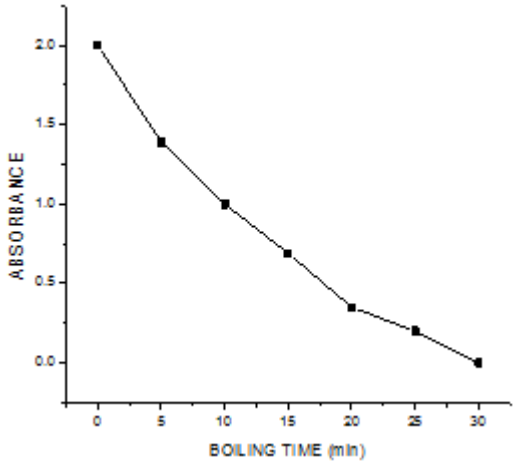
Figure 6
Red guava [Psidium cattleianum sabine] fresh cut pieces

Graph 2
Standard curve of vitamin – C at various concentrations



Absorbances are plotted at various concentrations of ascorbic acid. As the concentration is decreasing absorbance also decreases.

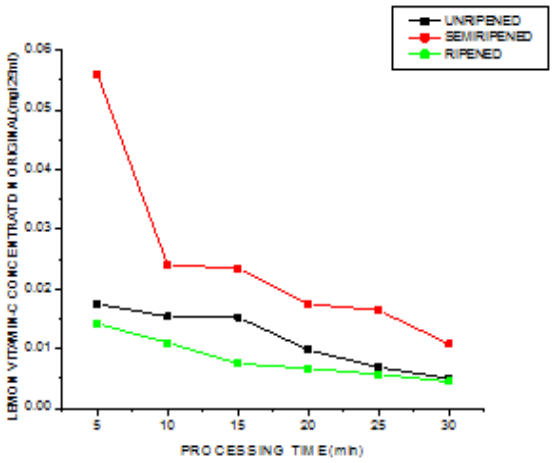
Graph3
Standard curve of vitamin – C at various boiling times



Absorbances are plotted at different boiling times. Absorbance of vitamin –c is decreased as the processing time is increased.

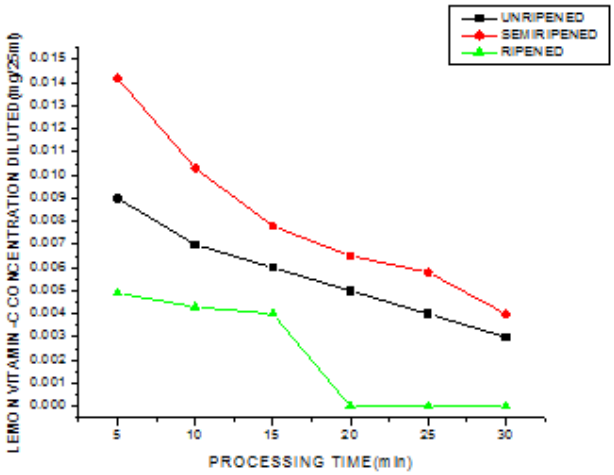
Graph4

Lemon vitamin – C concentration original



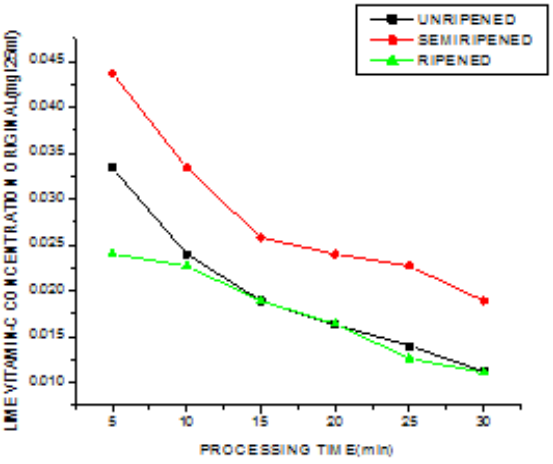
Graph5

Lemon vitamin – C concentration diluted



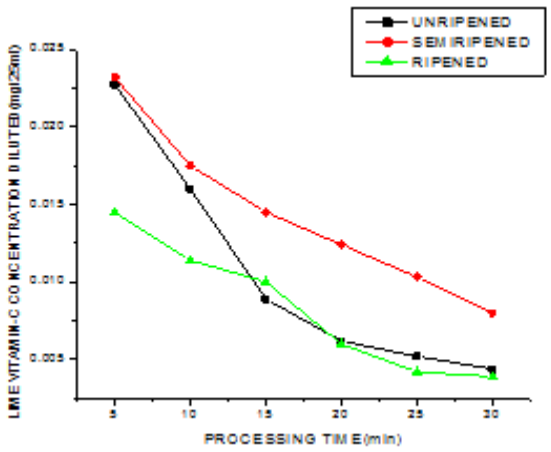
Graph6

Lime vitamin – C concentration original



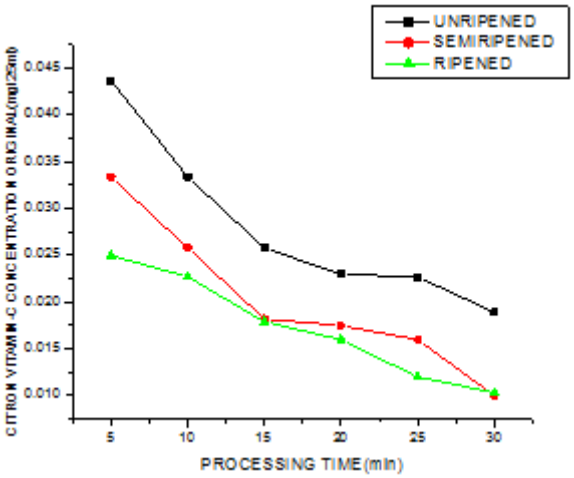
Graph7

Lime vitamin – C concentration diluted



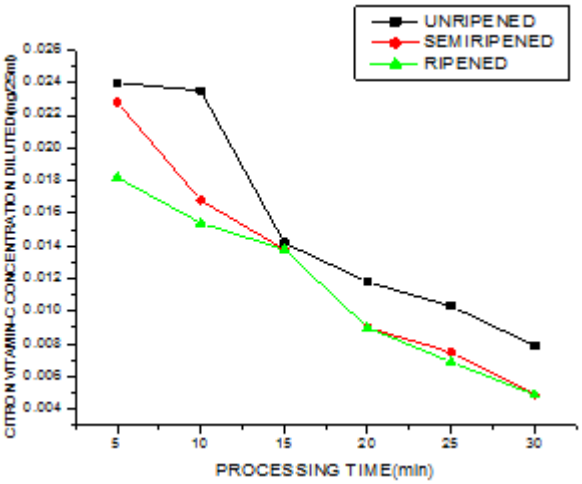
Graph8

Citron vitamin – C concentration original



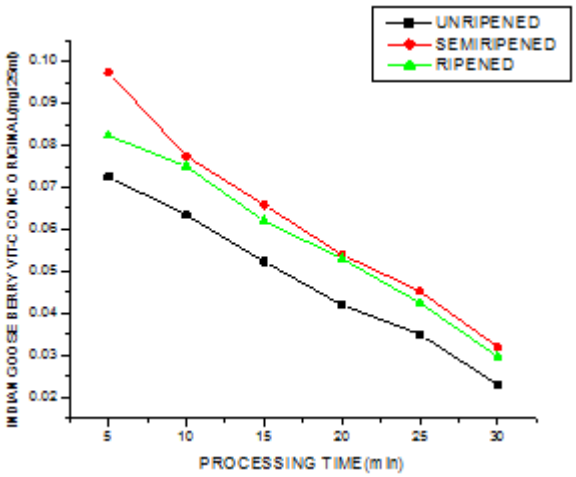
Graph9

Citron vitamin – C concentration diluted



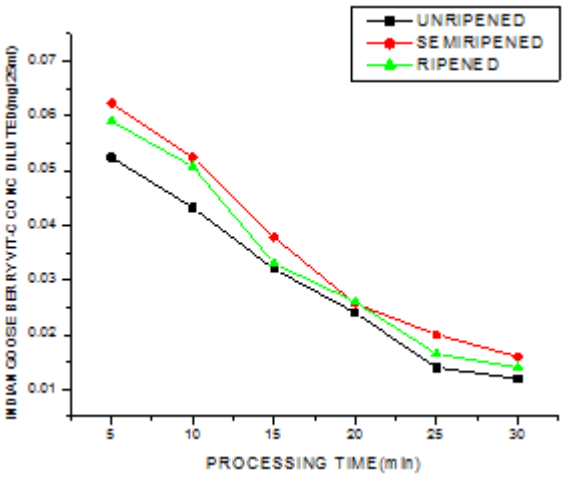
Graph10

Indian goose berry vitamin – C conc. Original



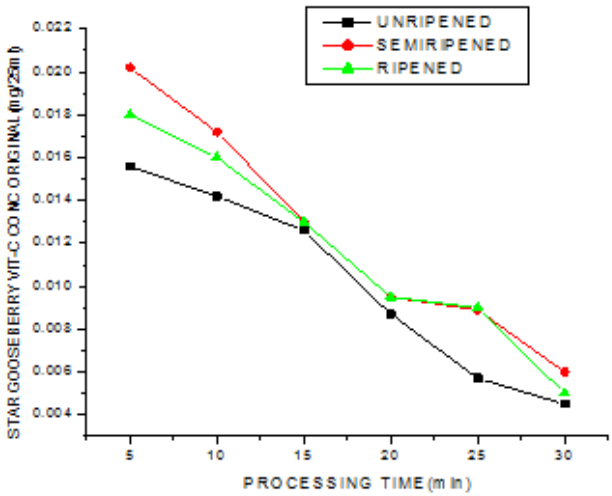
Graph11

Indian goose berry vitamin – C conc. diluted



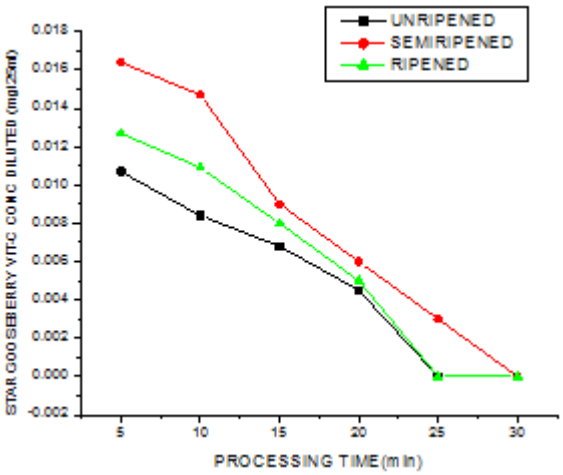
Graph12

Star goose berry vitamin – C conc. original



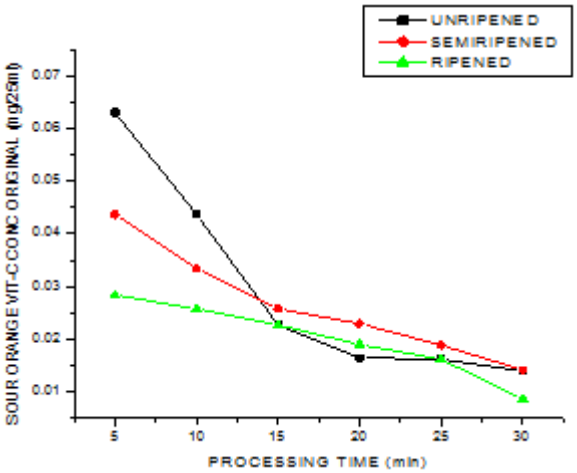
Graph13

Star goose berry vitamin – C conc. diluted



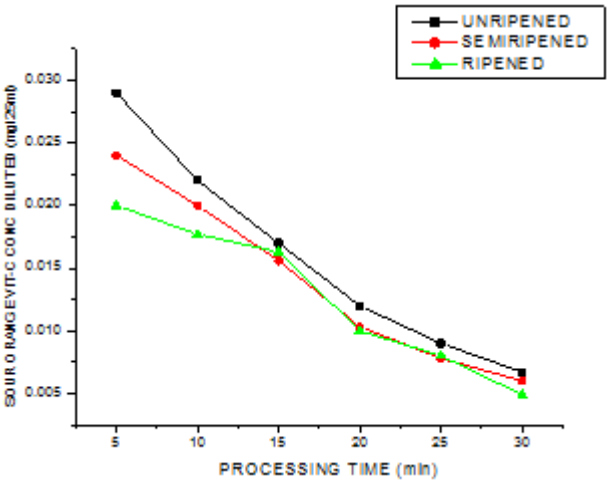
Graph14

Sour orange vitamin – C conc. Original



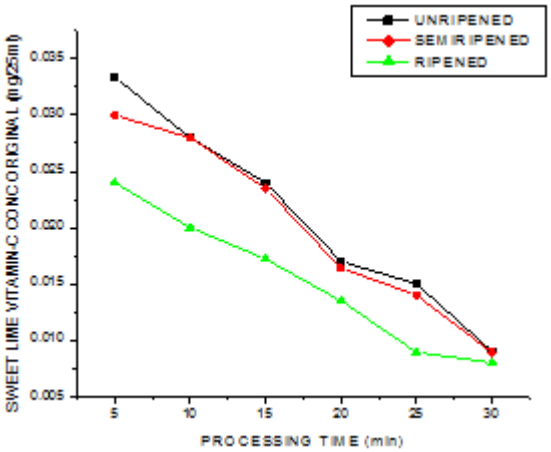
Graph15

Sour orange vitamin – C conc. Diluted



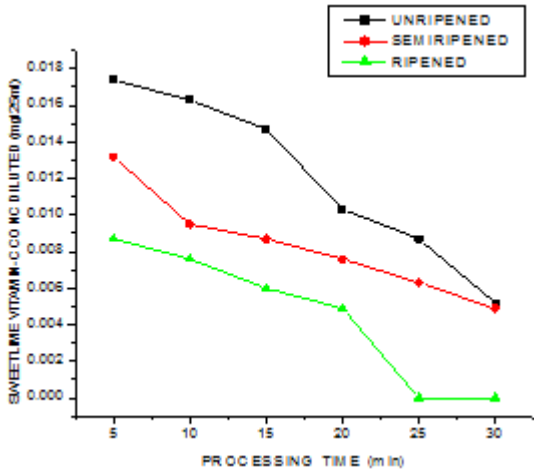
Graph16

Sweet lime vitamin – C conc. Original



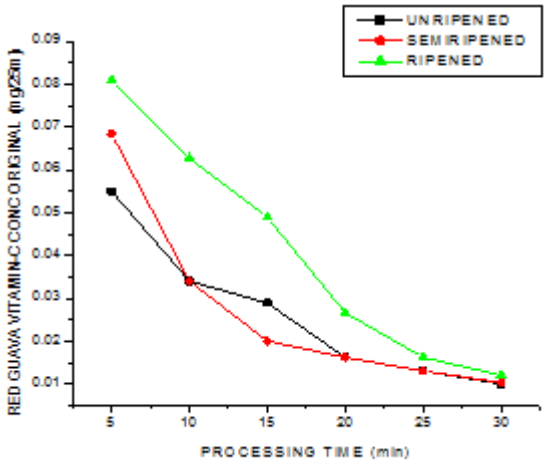
Graph17

Sweet lime vitamin – C conc. diluted



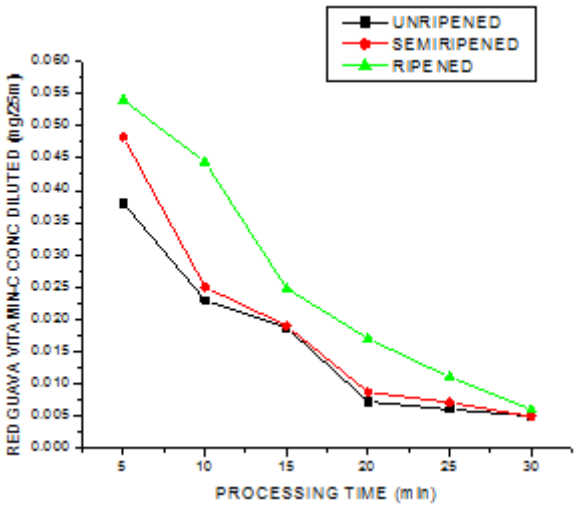
Graph18

Red guava vitamin – C conc. original



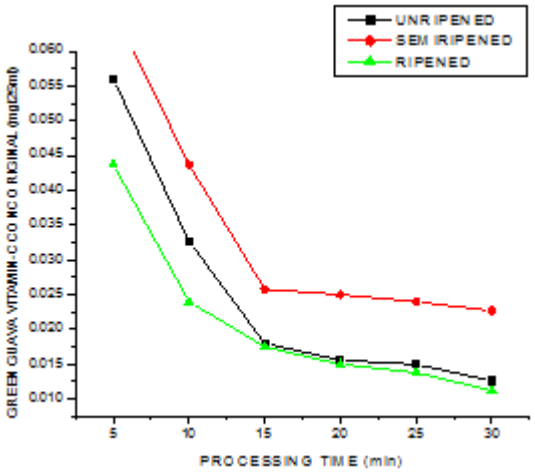
Graph19

Red guava vitamin – C conc. Diluted



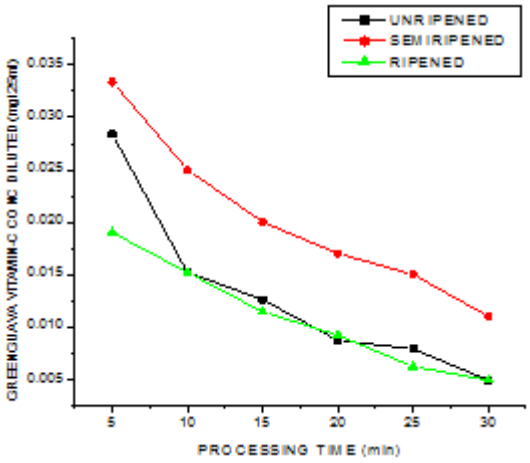
Graph20

Green guava vitamin – C conc. Original

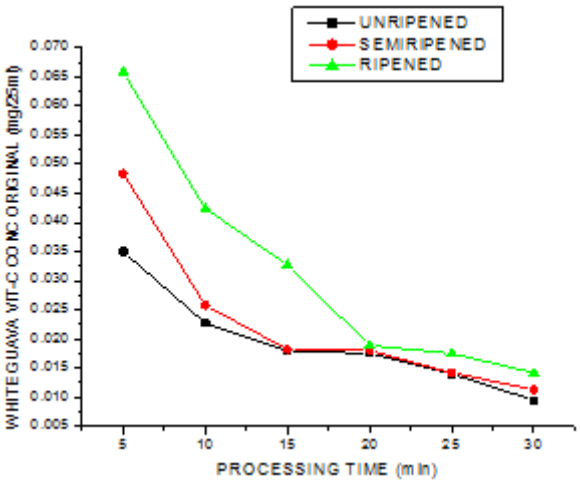


Graph21

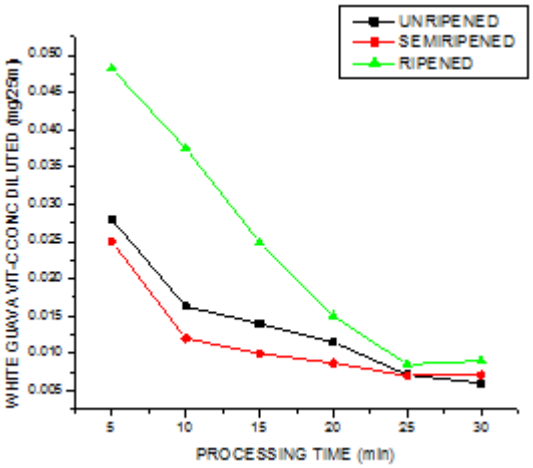
Green guava vitamin – C conc. diluted



Graph22
White guava vitamin – C conc. Original



Graph23
White guava vitamin – C conc. Diluted



Among the three stages of ripening, least concentration of ascorbic acid is observed in case of star goose berry whereas maximum concentrations are observed for green guava at unripened and semi ripened stages only. In case of fully ripened white guava maximum concentration is observed. In semi ripened and ripened stages of red guava (Fig 5 and 6) the concentrations are almost similar. In case of dilute samples, the least quantity is observed for lemon at unripened stage. For sweet lime (Fig 3) at semi ripened and fully ripened stages the least concentrations are noticed.Highest concentration at unripened stage is reported for sour orange (Fig 2).In case of Indian gooseberry major concentrations are observed at both semi and fully ripened stages. In citron (Fig 1) more concentration is observed in unripened stage. For lime at semi ripened stage higher concentration is noticed. With increase in processing time also similar decrease in concentration of ascorbic acid is observed. After dilution of lemon juice taken at ripened stage during processing time after 20, 25 and 30 minutes almost the concentration of ascorbic acid is completely reduced to zero. For dilute sample of star goose berry the concentration of ascorbic acid is completely reduced to zero at all the three ripening stages.

CONCLUSION

From the present investigation it is concluded that cheaply available guava has more concentration of vitamin C than other costly fruits. This method is more appropriate to determine the quantity of ascorbic acid. It is advisable to take fresh fruits than stored pulps and juices. Because processing of a fruit into juice and pulp involves thermal exposure which leads to deterioration of its quality.

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CONFLICT OF INTEREST

Conflict of interest declared none.

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