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Changes in sugars, pectin and antioxidants of guava *(Psidium guajava)* fruits during fruit growth and maturity

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ABSTRACT

Biochemical changes during fruit growth, development and maturity of eleven genotypes of guava (*Psidium guajava* L.) fruit were studied at 30, 60, 90, 105, 120 and 127 days after fruit set (DAFS). Total sugars content showed a continuous increase from initial stages of fruit development till ripening in all the genotypes. Total sugar was increased rapidly at later stage of fruit development. The ascorbic acid content was linearly increased until fruit ripening (127 DAFS) in all the genotypes except RCG 1, RCG 2 and RCG 3, which showed the increasing trend up to 120 days and thereafter slightly decreased on 127 DAFS. Ascorbic acid content was low at initial development stages and increased with advancement of fruit maturity and ripening. The pectin content of fruits increased up to 105 days after fruit set among most of the genotypes except RCG 1, RCG 2 and RCG 3 which showed up to 90 days only thereafter it declined rapidly. However, phenol content in guava fruits was gradually increased up to 90 days in RCG 1, RCG 2 and RCG 3 and up to 105 days in remaining genotypes after which a rapid decline was observed up to 127 days after fruit set. Based on the present findings, genotypes RCG 1, RCG 2 and RCG 3 may be harvested after 105 days of fruit set while rest of the genotypes after 120 days of the fruit set to get the quality fruits.

Key words: Antioxidants, Fruit development, Guava, Maturity, Pectin, Sugars

Guava (*Psidium guajava* L.) is one of the most well known edible tree fruits grown widely in more than sixty countries throughout the tropical and subtropical regions of the world. The fruits are delicious, rich in vitamin 'C', pectin and minerals like calcium, phosphorus and iron. Guava fruits are used as fresh as well as for making jam, jelly, nectar, paste etc. (Patra *et al.* 2004). The agro-climatic condition of the north eastern region of India is quite suitable for commercial cultivation of guava and the farmers are looking for diversification of fruit crops to enhance their income.

A comparative study on various physico-chemical parameters associated with the fruit quality of different guava cultivars has much value especially for selecting a guava cultivar for the region, which could provide superior quality fruit. Since, guava plants bears flowering and fruiting once in a year during April-May and October-November, respectively in this region as compared to twice or thrice crops per year in other parts of the India. It is known that the quality and storage life of fruits depend on various physiological and biological changes, which occur during fruit growth, development and maturity. Harvesting at appropriate maturity is an important factor affecting fruit quality. Changes in sugar, pectin and antioxidants of guava fruit at various stages of maturity and ripening were reported by several workers at different locations (Singh and Jain 2007). These are the important criteria for determining the optimum stage of fruit harvesting to get quality fruits with extended shelf-life. However, no information is available under mid hill situation of Meghalaya on these aspects of guava.

MATERIALS AND METHODS

The present investigation was carried out at Horticultural Research Farm of ICAR Research Complex for North Eastern Hills Region, Umiam, Meghalaya, India during 2008 and 2009. The experimental site was situated at 25°41'-21" North latitude and 91°55'-25" East longitude and at an elevation of 1010 m above mean sea level. The climate of the site can be characterized as sub temperate to sub tropical with minimum and maximum temperatures ranging from 6.8 to 29.7°C. The detail weather parameters are given in Table 1.

The experiment was laid out in randomized block design

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Table 1 Monthly weather parameters from February 2008 to December 2009

Month	Temperature (°C)		Relative humidity (%)		Rainfall	Sunshine
	Max.	Min.	Morning	Afternoon	(mm)	hours
			2008			
February	20.3	6.8	62.5	58.6	4.7	6.8
March	26.1	11.8	58.3	49.5	69.7	5.7
April	29.7	15.7	60.4	53.1	58.4	7.7
May	28.5	17.2	78.2	72.0	296.2	6.0
June	27.9	19.6	86.4	77.0	345.3	3.7
July	28.0	20.4	90.7	72.9	285.4	2.9
August	27.8	20.2	90.1	73.7	426.1	3.0
September	28.1	18.9	86.4	73.8	401.5	4.4
October	26.5	15.9	85.1	70.7	228.5	NA
November	24.2	10.1	78.1	55.9	36.4	NA
December	22.1	9.1	82.5	60.3	19.3	NA
			2009			
January	21.5	7.9	79.7	51.9		6.2
February	24.0	9.3	76.7	50.4		7.7
March	26.9	12.3	70.1	56.2	40.2	6.7
April	28.4	16.7	67.0	59.1	56.3	7.1
May	28.1	17.4	77.5	73.7	381.5	6.2
June	28.7	19.5	84.9	75.2	352.3	4.1
July	29.0	20.7	86.5	73.7	419.0	5.7
August	27.9	20.1	90.9	78.6	504.4	2.8
September	29.0	19.4	84.8	72.8	367.5	4.9
October	27.4	16.2	86.5	70.7	141.4	6.1
November	24.3	11.6	82.6	60.5	70.0	6.6
December	21.5	8.0	82.9	60.3	7.0	6.1

with three replications and three plants per replication. Five years old bearing plant of eleven genotypes of guava, viz. RCG 1, RCG 2, RCG 3, RCG 11, RCGH 1, RCGH 4, RCGH 7, Allahabad Safeda, L 49, Lalit and Sangam were selected for this study. The selected trees were marked with metal tag for recording observations. Few branches from all side of the plant were selected and then flowers were tagged with numbered metallic labels on the day of anthesis. The fruits initially set were marked in each vine. Fifteen fruits were randomly harvested from each replication at 30, 60, 90, 105, 120 and 127 days after fruit set (DAFS) for analysis of sugar, pectin, ascorbic acid and phenol content in guava fruits from June to first week of November.

Twenty five ml of the solution used for the estimation of reducing sugars was taken and 2.5 ml of concentrated HCl was added and kept overnight. Next day, the solution was neutralized with 1N NaOH and the volume was made up to 75 ml. It was then titrated against Fehling's solution A and B (5 ml each) as done in case of reducing sugars. From the following formula, total sugar was calculated (Ranganna 1997).

$$\text{Total sugar as invert sugar (\%)} = \frac{\text{mg of invert sugar} \times \text{Dilution} \times 100}{\text{Titre value} \times \text{Weight or volume of sample}}$$

The total pectin content of guava fruit was estimated by modified method of Ranganna (1997) in which pectin precipitated as calcium pectate from an acidified solution. Fifty grams of blended sample was taken into 1000 ml beaker and added with 400 ml of 0.05 N HCl and boiled for about 2 hours till complete evaporation. After that sample was cooled and made up the volume of 500 ml with distilled water. The 100 ml aliquots was taken into a conical flask added with 250 ml of distilled water and neutralize with 1 N NaOH using phenolphthalein as an indicator then allowed to stand overnight. Thereafter, 50 ml of 1 N acetic acid followed by 25 ml of 1 N calcium chloride were added in solution and stand for 1 hour in room temperature, then boiled for 1-2 minutes and filtered through a previously weighed filter and then precipitate was dried in oven and weighed again. The amount of pectin was recorded as calcium pectate and expressed in percent.

Ascorbic acid content was determined by using 2, 6-Dichlorophenol-indophenol dye method (AOAC 1970). Five grams of the sample was grounded with about 25 ml of 4% oxalic acid and filter through Whatman No. 4 filter paper. The filtrate was collected in a 50 ml volumetric flask and the volume was made up with 4% oxalic acid and titrated against the standard dye to a pink point. The amount of ascorbic acid was recorded as mg ascorbic acid per 100 g of pulp. The amount of ascorbic acid was calculated using the following formula and expressed as mg/100g.

$$\text{Absorbic acid (mg/100 g)} = \frac{\text{Titre value} \times \text{Dry factor} \times \text{Volume make up} \times 100}{\text{Aliquot} \times \text{Weight of the sample}}$$

Total phenol content was determined using the Folin-Ciocalteu's reagent (Singleton and Rossi 1965). The 0.5 g fruit pulp was homogenized in 10 ml of 80% aqueous ethanol at room temperature and centrifuged at 10 000 rpm for 20 minutes and the supernatant was collected in petridish and residue was again re-extracted twice. The supernatant collected in evaporating dishes allow drying in room temperature. The residue was dissolved in 10 ml of distilled water. One ml of this extract was taken in 25 ml volumetric flask and added with 9 ml distilled water and 5 ml of Folin-Ciocalteu's reagent. After 3 minutes, 10 ml of 20% sodium carbonate was added and contents were mixed thoroughly and stored for 1 hour after that absorption reading was recorded with spectrophotometer at 650 nm. Total phenol content was expressed in gallic acid equivalents (GAE) in mg per 100 g fresh weight using a gallic acid standard curve.

RESULTS AND DISCUSSION

Ascorbic acid

The data presented in Table 1 revealed that a linear

increase in ascorbic acid was noticed till fruit ripening (127 DAFS) in all the genotypes except RCG 1, RCG 2 and RCG 3, which showed the increasing trend up to 120 days and thereafter slightly decreased at 127 DAFS. Ascorbic acid content was low at initial development stages and increased with advancement of fruit maturity and ripening.

The highest value of ascorbic acid was recorded in RCGH 1 (156.97 mg) followed by RCG 1 (150.28 mg) while, the lowest ascorbic acid was present in Allahabad Safeda (98.23 mg) on 105 DAFS. On 120 DAFS, the highest content of ascorbic acid was observed in RCGH 1 (198.73 mg) which was significantly superior over rest of the genotypes. The next best performers were RCG 1 (182.66 mg) closely followed by RCG 11 (182.48 mg) which were at par with each other but, significantly higher than others while, significantly lowest content was obtained in Allahabad Safeda (137.38 mg). Performance of genotypes differed significantly for the trait on 127 days also. The highest ascorbic acid was recorded in RCGH 1 (238.15 mg) followed by RCGH 7 (205.64 mg) and both were differed significantly with each other and higher than rest of the genotypes. The next best performers were Lucknow 49 (198.05 mg) followed by RCGH 4 (193.25 mg) which were at par with each other but, significantly higher than others while, lowest ascorbic acid was possessed in RCG 3 (129.13 mg) followed by RCG 2 (138.59 mg) and RCG 1 (162.46 mg) differed significantly among themselves. The result is in line with the findings of previous workers, Mitra and Bose (1996), who reported that ascorbic acid content increased very slowly at the initial stages of development, then increased rapidly and reached the maximum value at full ripe stage. It has been suggested that ascorbic acid production is linked with pectin degradation, galacturonic acids being postulated as a substrate for synthesis of ascorbic acid (Mapson and Isharwood 1956). Period of ascorbic acid accumulation corresponded with decrease of pectin content in the fruit. It also appeared that active synthesis of ascorbic acid during fruit development and early ripening might be attributed to inactivation of ascorbic acid oxidase due to high content of phenols. Increased in ascorbic acid content with advancement of fruit maturity and ripening in guava were also observed by several workers El-Bulk *et al.* 1997, Mercado-Silva *et al.* 1998, Hegde and Chharia 2004, Soares *et al.* 2007). However, decrease in ascorbic acid content in RCG 1, RCG 2 and RCG 3 at 120 days after fruit set, which might be due to climacteric rise in respiration resulting in oxidation of ascorbic acid by enzyme ascorbic acid oxidase. This result are in agreement with the observations of Selvaraj *et al.* (1999) and Bashir *et al.* (2003), they also observed a decreasing trend.

Sugars

Data presented in Table 2 showed an increasing trend for reducing sugar content in fruit pulp till last harvesting for all the genotypes.Performance of genotypes revealed that RCG 1 (2.94%) exhibited the highest value of reducing sugar followed by RCG 3 (2.82%) showing at par with each other while; the least content of reducing sugar was present in Sangam (1.94%) on 105 DAFS. Whereas on 120 DAFS, the highest sugar content was expressed by RCGH 1 (3.92%) followed by RCG 11 (3.88%) and RCGH 7 (3.77%) showing at par values. The lowest reducing sugar was present in Sangam (3.19%) followed by Lalit (3.22%) and RCG 3 (3.24%). The reducing sugar content on 127 DAFS was found to be significantly highest in RCG 11 (4.48%) followed

Table 2 Performance of guava genotypes for ascorbic acid at different days after fruit set

Genotypes	Ascorbic acid (mg/100 g)					
	Days after fruit set					
	30	60	90	105	120	127
RCG 1	17.23	66.38	113.78	150.28	182.66	162.46
RCG 2	18.89	58.56	103.52	144.32	173.97	138.59
RCG 3	14.90	54.34	96.77	130.42	154.82	129.13
RCG 11	26.78	54.12	98.91	135.45	182.48	212.83
RCGH 1	27.89	69.29	118.75	156.97	198.73	238.15
RCGH 4	22.53	43.25	84.35	118.05	162.26	193.25
RCGH 7	19.60	45.97	84.13	118.92	161.61	205.64
Allahabad Safeda	22.39	38.76	69.57	98.23	137.38	181.15
Lucknow 49	22.04	47.68	84.39	119.57	163.03	198.05
Lalit	21.62	43.05	79.00	113.61	156.04	171.92
Sangam	16.89	44.00	83.97	117.99	157.34	182.87
SEm±	1.29	1.63	2.05	2.56	2.25	2.35
CD (P=0.05)	3.87	4.67	5.85	7.31	6.43	6.72

Table 3 Performance of guava genotypes for reducing sugar at different days after fruit set

Genotypes	Reducing sugar (%)					
	Days after fruit set					
	30	60	90	105	120	127
RCG 1	0.80	0.86	1.81	2.94	3.70	3.78
RCG 2	0.79	0.87	1.78	2.67	3.50	3.59
RCG 3	0.81	0.93	1.92	2.82	3.24	3.31
RCG 11	0.86	0.96	1.46	2.28	3.88	4.48
RCGH 1	0.83	0.94	1.62	2.39	3.92	4.15
RCGH 4	0.76	0.89	1.35	2.50	3.36	3.45
RCGH 7	0.80	0.94	1.44	2.26	3.77	4.25
Allahabad Safeda	0.79	0.82	1.55	2.54	3.36	3.51
Lucknow 49	0.80	0.91	1.55	2.06	3.45	3.62
Lalit	0.71	0.81	1.38	2.11	3.22	3.51
Sangam	0.71	0.77	1.33	1.94	3.19	3.53
SEm±	0.03	0.03	0.05	0.06	0.06	0.08
CD (P= 0.05)	0.09	0.09	0.14	0.17	0.17	0.23

by RCGH 7 (4.25%) while, significantly low value of reducing sugar was exhibited by RCG 3 (3.31%) followed by RCGH 4 (3.45%) (Table 3).

Total sugar content showed a continuous increase from initial stages of fruit development till ripening in all the genotypes (Fig 1). Total sugar increased rapidly at later stage of fruit development. The highest value of total sugar was observed in RCG 1 (5.74%) followed by at par values of RCG 3 (5.48%) while, the lowest content was observed in Lucknow 49 (3.98%) on 105 DAFS. Whereas on 120 DAFS, the highest total sugar content was exhibited by RCG 11 (7.65%), which was at par with RCGH 1 (7.61%) but, significantly higher than RCGH 7 (7.38%) and other genotypes. Significantly lowest sugar was present in RCG 3 (5.96%) followed by Lalit (6.11%) showing at par values. The highest total sugar was exhibited by RCG 11 (8.61%) although at par with RCGH 1 (8.52%) but, significantly superior than RCGH 7 (8.32%) and others. The fruits with least sugar were obtained in RCG 3 (6.18%) on 127 days of fruit set. Increase in sugar during ripening might be due to depolymerisation of polysaccharides and conversion of fruit starch to sugars. The highest value of reducing and total sugar on 105 DAFS in RCG 1 and RCG 3 might be due to genetic characteristics of these genotypes to mature early after fruit set. Results are in accordance to findings obtained by Mitra and Bose (1996), El-Bulk *et al.* (1997), Mercado-Silva *et al.*(1998), Selvaraj *et al*. (1999), Bashir *et al*.(2003), Hegde and Chharia (2004) and Singh and Jain (2007).

Pectin content

Pectin content showed significant variation among the genotypes during investigation. The pectin content of fruits increased up to 105 days after fruit set among most of the genotypes except RCG 1, RCG 2 and RCG 3 which showed up to 90 days only thereafter it declined rapidly (Table 4).

Table 4 Performance of guava genotypes for pectin content at different days after fruit set

Genotype	Pectin content (%)					
	Days after fruit set					
	30	60	90	105	120	127
RCG 1	0.64	0.91	1.19	0.98	0.86	0.75
RCG 2	0.62	0.88	1.16	0.95	0.81	0.74
RCG 3	0.95	1.35	1.62	1.41	1.32	1.25
RCG 11	0.90	1.21	1.43	1.61	1.38	1.30
RCGH 1	0.92	1.26	1.52	1.71	1.53	1.36
RCGH 4	0.56	0.79	1.01	1.16	0.85	0.81
RCGH 7	0.89	1.27	1.54	1.73	1.57	1.40
Allahabad Safeda	0.89	1.00	1.27	1.36	1.03	0.97
Lucknow 49	0.94	1.16	1.29	1.42	1.20	1.03
Lalit	0.95	1.14	1.39	1.55	1.21	1.14
Sangam	0.76	0.99	1.27	1.41	1.09	0.95
SEm±	0.05	0.04	0.04	0.04	0.03	0.03
CD (P= 0.05)	0.14	0.12	0.12	0.12	0.09	0.09

The highest pectin content was recorded in RCGH 7 (1.73%) followed by RCGH 1 (1.71%) and RCG 11 (1.61%) which were at par while, the lowest content was observed in

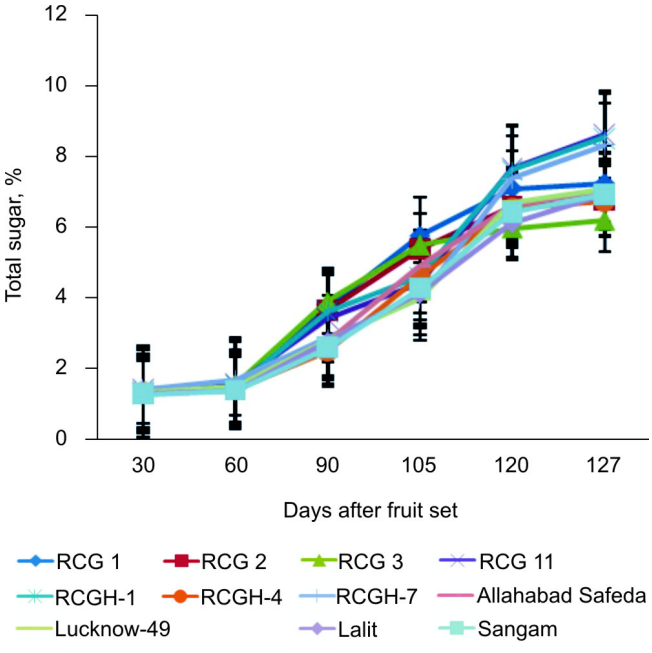

Fig 1 Changes in total sugar content of guava genotypes

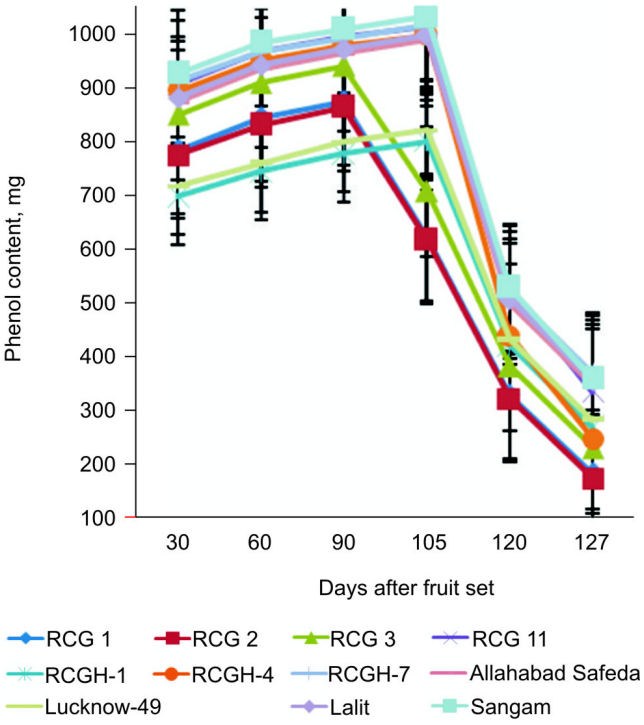

Fig 2 Changes in phenol content of guava genotypes

RCG 2 (0.95%) followed by RCG 1 (0.98%). At 120 DAFS, the highest total pectin content was exhibited again by RCGH 7 (1.57%), which was at par with RCGH 1 (1.53%) but, significantly higher than RCG 11 (1.38%) and RCG 3 (1.32%). Significantly lowest pectin was present in RCG 2 (0.81%) followed by RCGH 4 (0.85%) and RCG 1 (0.86%) showing at par values. On 127 days of fruit set also, the highest pectin content was exhibited by RCGH 7 (1.40%), which was at par with RCGH 1 (1.36%) and significantly higher than RCG 11 (1.30%). Whereas, significantly lowest pectin content was analyzed in RCG 2 (0.74%) followed by RCG 1 (0.75%) and RCGH 4 (0.81%) which were at par (Table 3). Similar finding in guava cultivars at various locations were also reported by El-Bulk *et al.* (1997), Selvaraj *et al.* (1999), Hegde and Chharia (2004), Suryakanth and Mukunda (2007). Pectin degradation is linked with ascorbic acid production and it is postulated that glacturonic acids are substrates needed in synthesis of ascorbic acid. Period of ascorbic acid accumulation corresponded with falling of pectin content in fruit (Yan *et al.* 2006). Increase in pectin content during fruit development might be due to conversion of other forms of pectin into water soluble form of pectin and in later stage the decrease in pectin could be due to enzymatic degradation of pectin with advanced ripening (Paul and Chen 1983).

Phenol content

The phenol content in guava fruits was gradually increased up to 90 days in RCG 1, RCG 2 and RCG 3 and up to 105 days in remaining genotypes after which a rapid decline was observed up to 127 days after fruit set (Fig 2). The highest phenol content was noticed in Sangam (1 034.93 mg) and lowest in RCG 2 (617.33 mg) on 105 DAFS. Whereas on 120 DAFS, the highest phenol content was possessed again in Sangam (529.94 mg) which was at par with RCG 11 (525.60 mg) followed by RCGH 7 (520.39 mg) and Lalit (508.60 mg) while, the lowest was registered again in RCG 2 (322.48 mg) followed by RCG 1 (328.45 mg) Which were at par. The highest phenol content on 127 DAFS was exhibited by RCGH 7 (368.73 mg) and was at par with Sangam (358.95 mg) and Lalit (356.88 mg) while, significantly least phenol content was recorded in RCG 2 (173.70 mg) and RCG 1 (183.45 mg) showing at par each other. This result was in agreement with the findings of Hegde and Chharia (2004), Yan *et al.* (2006) and Reddy *et al.* (2010) in guava. Increase in phenol content during early stage of fruit growth might be due to increase of its biosynthesis in fruits initially but, decrease in later stage might be due to increase in activity of polyphenol oxidase during ripening of guava (Mowlah and Itoo 1982).

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