Indian J. Hort. 69(3), September 2012: 409-415

Comparison of cell wall degrading enzyme activities during ripening of guava fruit on-tree and in-storage

Arti Sharma*, R.K. Sharma, Saleem Siddiqui and Bindiya Sharma
Department of Horticulture, CCS Haryana Agricultural University, Hisar 125 004, Haryana

ABSTRACT

Changes in the activities of various cell wall degrading enzymes, *viz*., exo-polygalacturonase (PG), β-D-galactosidase, cellulase and pectin methyl esterase (PME) were assayed on alternate days. Activities of PG, β-D-galactosidase and cellulase increased continuously with the advancement of ripening on-tree as well as in-storage, whereas the activity of PME showed a fluctuating trend, it initially increased and thereafter showed a declining trend during ripening on-tree as well as in-storage. Further the activities of PG, β-D-galactosidase and cellulase showed a very high negative correlation with loss of fruit firmness during ripening, whereas the activity of PME did not showed a significant correlation with firmness. Changes in the activities of all the enzymes studied were faster during ripening in-storage as compared to ripening on-tree in both the cultivars; however cv. Hisar Surkha exhibited faster changes than cv. Hisar Safeda. Changes in enzymatic activities were rapid in rainy season than winter season crop.

Key words: Cellulase, exo-polygalacturonase, guava, pectin methyl esterase, β-D-galactosidase.

INTRODUCTION

Guava (*Psidium guajava* L.) is one of the most important tropical fruit. Fresh guava is rich source of vitamin-C and also contains appreciable amounts of niacin, calcium, phosphorus and pectin. Guava seed are potential source of iron and oil (Misra and Sheshadri, 16). Guava is a perishable fruit and highly prone to bruising and mechanical injuries. To reduce the percent losses in guava and to avoid glut, it is desirable to evolve technologies for prolonging its keeping quality through delaying softening process during ripening. Development of practical solution to the post harvest problems requires detailed understanding of biochemistry of fruit ripening. The various physiological and biochemical changes take place in guava during ripening have been studied by many workers (Selvaraj *et al.*, 22; Jain *et al.*, 11). It has been observed that ripening behavior of fruit while attached to tree may not be the same as in the detached fruit during storage (Nunes *et al.*, 18).

Softening during ripening involves structural as well as biochemical modifications of cell wall polysaccharides. While the mechanisms that regulate such modifications are not fully understood, cell wall hydrolytic enzymes are reported to contribute to softening in climacteric fruits such as apple, avocado, mango, papaya, pear and tomato (Huber, 9).Thus, the present investigation was undertaken with the objectives to compare the pattern of changes in activities of some hydrolyzing enzymes and find out the correlation of activities of enzymes (PG, PME, cellulase and β-D galactosidase) with loss of fruit firmness during ripening of guava fruits on-tree and in-storage.

*Corresponding author's present address: Division of Fruit Science, SKUAST-J, Main Campus Chatha, Jammu; E-mail: drartisharma02@gmail.com

MATERIALS AND METHODS

The present investigation was carried out during rainy and winter season crop of a year, at Post Harvest Laboratory of Department of Horticulture CCS HAU, Hisar. Fruits of guava cultivars Hisar Safeda and Hisar Surkha were harvested with secateur keeping a small intact pedicel with each fruit, from ten year old trees growing at Orchard of the department of Horticulture, CCS Haryana Agricultural University, Hisar. For studying ripening on-tree the fruits from tree were harvested on the basis of visual observation and firmness at three maturity stages, *viz.*, green mature stage (GMS): 100% green fruit; half ripe stage (HRS): 50% yellow and 50% green fruit; full ripe stage (FRS): 80% yellow and 20% green fruit (These stages were decided by visually observing the fruit skin colour and firmness was checked by putting pressure by hand. Fruits which were not compressed by applying pressure with hand were categorized into GMS, while those fruits which were slightly compressed were categorized as HRS and those which were punctured were categorized as FRS). For studying ripening, in-storage 10 kg uniform size fruits of both the cvs. Hisar Safeda and Hisar Surkha were harvested at green mature stage from the trees of uniform size and age. Fruits were divided into four replicates, each of 2.5 kg, packed separately in 2% perforated polythene bags (200 gauge) and stored at room temperature (Average room temperature during study was 30°C

in rainy season and 10°C in winter season). Three fruits were analyzed from each replicate and the activities of various cell wall degrading enzymes were assayed on respective stages for on-tree ripening and on alternate days for in-storage ripening. Activity of polygalacturonase (EC3.2.1.67) was assayed by the method of Bartley (2) , cellulase (EC3.2.1.4) activity by the method of Dick *et al.* (5) and the activity of pectin methyl esterase (EC 3.1.1.11) was assayed by the method of Hobson (8). Correlation between the activity of these enzymes and loss of fruit firmness during ripening was calculated using Karl Pearson's coefficient of correlation (r^2) as suggested by Panse and Sukhatme (19).

RESULTS AND DISCUSIION

Results related to changes in the firmness of fruits of fruits of guava hybrids, *viz.*, Hisar Safeda and Hisar Surkha followed a declining trend with the advancement of ripening on-tree as well as in storage (Tables 1 & 2). During on-tree ripening, in rainy season, fruit firmness was 13 kg/cm^2 at green mature stage, which reduced significantly to 5.7 and 3.8 kg/cm^2 at half ripe stage and full ripe stage, respectively while in winter season, it decreased from 13 kg/cm^2 at green mature stage to 7.00 and 4.30 kg/cm^2 at half ripe and full ripe stages, respectively. Varietal differences as well as interactions between varieties and stages of harvest were statistically non-significant. This loss of firmness may be attributed to either loss of turgor or degradation of cell wall (Brady, 3). During ripening in-storage, in rainy season, fruit firmness was 13 kg/cm^2 in both the hybrids at 0 day of storage which decreased to 8.2 kg/cm^2 in cv. Hisar Safeda and 6.8 kg/cm^2 in cv. Hisar Surkha on 2nd day of storage and showed a significant declining trend throughout the storage period, reaching to a minimum of 3.2 kg/cm^2 in cv. Hisar Safeda and 1.2 kg/cm^2 in cv. Hisar Surkha on 6th day of storage. In winter season, both the hybrids yielded the values of 13 kg/cm^2 upto

Table 1. Changes in the firmness (kg/cm^2) and activities (units*/g f.tw./h) of polygalcturonase (PG), β-D-galactosidase in guava fruits during ripening on-tree.

Harvest stage	Firmness			PG			ß-D-galactosidase		
	Hisar Safeda	Hisar Surkha	Mean	Hisar Safeda	Hisar Surkha	Mean	Hisar Safeda	Hisar Surkha	Mean
					Rainy season				
GMS	13.0	13.0	13.0	0.065	0.071	0.068	351	382	366
HRS	6.0	5.9	5.7	0.082	0.092	0.087	443	495	469
FRS	4.0	3.6	3.8	0.090	0.102	0.096	487	549	518
Mean	7.6	7.3		0.079	0.088		427	475	
r^2 with firmness					-0.99			-0.99	
CD at 5%									
Variety (A) :	NS				0.02			13.8	
Storage period (B) :	0.5				0.03			14.2	
A × B :	NS				NS			NS	
					Winter season				
GMS	13.0	13.0	13.0	0.042	0.053	0.048	158	252	205
HRS	7.1	6.9	7.0	0.062	0.078	0.070	233	371	301
FRS	4.5	4.1	4.3	0.068	0.089	0.079	254	423	338
Mean	8.2	8.0		0.057	0.073		215	348	
r^2 with firmness					-0.99			-0.99	
CD at 5%									
Variety (A) :	NS				0.02			12.6	
Storage period (B) :	0.9				0.03			13.5	
A × B :	NS				NS			NS	

1 unit* of PG = mg of galacturonic acid liberated

1 unit* of ß-D-galactosidase = micro mole of nitrophenol liberated

GMS = Green mature stage, HRS = Half ripe stage, FRS = Full ripe stage

Table 2. Changes in the firmness (kg/cm²) and activities (units*/g f.tw./h) of polygalcturonase (PG) and ß-D-galactosidase in guava fruits during ripening in storage.

Storage (days)	Firmness			PG			ß-D-galactosidase		
	Hisar Safeda	Hisar Surkha	Mean	Hisar Safeda	Hisar Surkha	Mean	Hisar Safeda	Hisar Surkha	Mean
					Rainy season				
0	13.0	13.0	13.0	0.065	0.071	0.068	351	382	366
2	8.2	6.8	7.5	0.080	0.089	0.084	432	479	455
4	5.9	2.9	4.4	0.086	0.109	0.097	464	586	525
6	3.2	1.2	2.2	0.093	0.119	0.106	502	639	570
Mean	7.5	5.9		0.081	0.097		437	521	
r^2 with firmness					-0.99			-0.99	
CD at 5%									
Variety (A) :	0.2				0.03			14.2	
Storage period (B) :	0.3				0.04			14.7	
A × B :	0.4				NS			NS	
					Winter season				
0	13.0	13.0	13.0	0.043	0.053	0.048	158	252	205
2	13.0	13.0	13.0	0.048	0.058	0.053	176	276	226
4	8.1	6.8	7.4	0.056	0.066	0.061	205	314	259
6	6.2	3.9	5.0	0.063	0.074	0.068	231	352	291
8	3.5	3.0	3.2	0.066	0.084	0.075	242	399	320
Mean	8.7	7.9		0.055	0.067		202	318	
r^2 with firmness					-0.97			-0.97	
CD at 5%									
Variety (A) :	0.2				0.03			11.7	
Storage period (B) :	0.3				0.02			12.8	
A × B :	0.4				NS			NS	

1 unit* of PG = mg of galacturonic acid liberated

1 unit* of ß-D-galactosidase = micro mole of nitrophenol liberated

2nd day of storage thereafter the values of firmness declined significantly to 3.5 kg/cm² in Hisar Safeda and 3.0 kg/cm² in cv. Hisar Surkha on 8th day of storage. Cultivar Hisar Safeda retained higher firmness than cv. Hisar Surkha throughout the investigation in both the seasons. Tucker and Grierson (28) established the positive role of some degrading enzymes such as PG, PME and galactosidase with the fruit firmness. Loss of fruit firmness was more in rainy season than winter season which may be attributed to higher temperature in rainy season.

Activity of enzyme polygalacturonase (PG) increased progressively and significantly with the advancement of ripening on-tree as well as in-storage (Tables 1 & 2). During ripening on-tree in rainy season, at green mature stage, the PG activity was measured to be 0.068 units/gfw/h which further increased significantly to 0.087 and 0.096 units/gfw/h at half ripe and full ripe stages, respectively. In winter season PG activity was 0.048 units/gfw/h at green mature stage which showed a significant increase to 0.070 units/gfw/h and attained a maximum value of 0.079 units/gfw/h at full ripe stage. During ripening in-storage, in rainy season, PG activity increased from 0.068 units/gfw/h at 0 day to 0.106 units/gfw/h on 6th day of storage. In winter season, its value was recorded to be 0.048 units/gfw/h on 0 day of storage which eventually reached to the value of 0.075 units/gfw/h on 8th day of storage .The PG activity has been reported to be responsible for fruit softening (Tucker *et al.*, 29; Huber, 9). The increase in PG activity has been found responsible for a decrease in covalently bound pectin content due to its hydrolysis into free pectin (Siddiqui and Sharma, 27). The results reported herein are more or less in accordance with the earlier findings of Saroja *et al.* (20) and Goukh and Bashir

(7) in guava, Abu- sarra and Abu-Goukh (1) in mango, and Lazan *et al*. (15) in durian.

Cultivar Hisar Surkha exhibited higher PG activity than cv. Hisar Safeda which seems to be responsible for its fast ripening behavior. During the entire investigation activity of PG was found to have a significant negative correlation with fruit firmness. Siddiqui and Sharma (27) during ripening of *ber* recorded more or less parallel results. A significant and progressive increase in the activity of β-D-galactosidase (Tables 1 & 2) with the advancement of ripening was observed during the present investigation. During ripening on-tree, in rainy season, β-D-galactosidase activity was recorded to be 366 units/gfw./h at green mature stage, which increased to 469 and 518 units/gfw/h at half ripe and full ripe stages, respectively. Similar trend was repeated in winter season where its activity increased from 205 units/gfw/h at green mature stage to 301 and 338 units/gfw/h at half and full ripe stages, respectively.

During ripening in-storage, in rainy season, activity of enzyme β-D-galactosidase in guava fruits recorded on 0 day of storage was equivalent to 366 units/gfw/h which increased progressively throughout the storage period and reached to a maximum value of 570 units/gfw/h on 6th day of storage. During winter season, its value increased from 205 to 320 units/gfw/h from 0 to 8th day of storage. Activity of β-D-galactosidase was higher in cv. Hisar Surkha than Hisar Safeda, which might due to genetic variation among the hybrids. β-D- galactosidase activity was slower in winter season fruits as compared to rainy season fruits due to overall retardation in enzymatic activities at lower temperature. The present investigation revealed a very high and significant negative correlation between activity of β-D-galactosidase and loss of fruit firmness. Siddiqui and Bangerth (25) have also observed similar results during softening of *ber* and apple, respectively. The increased activity of β-D-galactosidase with ripening has also been reported in subtropical fruit crops (Lazan *et al*., 15), apple (Siddiqui and Bangerth, 24), and *ber* (Siddiqui and Sharma, 27).

Cellulase activity exhibited an increasing trend with the advancement of ripening (Tables 3 & 4) . During ripening on-tree, in rainy season, cellulase activity at green mature stage was measured to be 20.83 units/gfw/h which increased significantly to 24.31 units /gfw/h and 24.60 units/gfw/h at half ripe and full ripe stage, respectively. Similar trend was also recorded in winter season with an activity of 16.39 units /gfw/h at green mature stage which further increased significantly to 19.60 units/gfw/h at half ripe stage and then increased slightly to 20.32 units/gfw/h at full ripe stage. During ripening in-storage, in rainy season, cellulase activity on 0 day of storage was recorded to be 20.83 units/gfw/h it further increased significantly upto 6th day of storage and attained a maximum value of 29.22 units/gfw/h on 6th day. Cellulase activity of winter season fruits increased gradually from 16.68 to 21.60 units/gfw/h from 0 to 8th day of storage. Increased activity of cellulase enzyme during ripening is likely to cause loosening of cell wall thereby reducing fruit firmness (Lallan,14). These results are in agreement with the earlier results of chin *et al.* (4) and Goukh and Bashir (7) in guava, Ning *et al*. (17) in Chinese pear, and Ianneta *et al.* (10) in raspberry. However in contradiction to it Selvaraj *et al.* (21) have reported that cellulase activity increased from green to green mature stage and then declined until ripening stage in guava cvs. Allahabad Safeda and Sardar.

It was also noted that cv. Hisar Surkha showed higher cellulase activity than cv. Hisar Safeda. Furthermore, winter season fruits exhibited low level of cellulase activity. A strong negative correlation was observed in between cellulase activity and loss of fruit firmness during the entire investigation a similar correlation in between cellulase activity and softness of fruit has also been reported in mango (Abu-Sarra and Abu-Goukh, 1).

The activity of enzyme pectin methyl esterase showed a fluctuating trend during ripening of guava fruits (Tables 3 & 4) . During ripening on-tree, in rainy season, activity of PME increased significantly from 63.85 units/gfw/h at green mature stage to 72.57 units/gfw/h at half ripe stage and then declined significantly to 68.37 units/gfw/h at full ripe stage. During winter season, activity of PME increased significantly from 57.65 units/gfw/h at green mature stage to 63.17 units/gfw/h at half ripe stage and thereafter reduced significantly to 60.39 units/gfw/h full ripe stage. During ripening in-storage, in rainy season fruits, activity of PME increased significantly from 63.85 to 68.05 units/gfw/h from 0 to 2nd day of storage and thereafter its activity did not change significantly upto 4th day of storage and then decreased significantly to 65.59 units/gfw/h on 6th day of storage. In winter season fruits, PME activity increased significantly upto 4th day of storage. On 0 day of storage its value was recorded to be 58.05 units/gfw/h, which was increased to a maximum value of 66.33 units/gfw/h on 4th day, thereafter it declined significantly to 62.45 and 58.00 units/gfw/h on 6th and 8th day of storage, respectively.

It has been postulated that PME assists PG enzyme for methylation or esterification of galacturonide chains of pectin (Huber, 9). Thus, decreased PME activity during later stages indicates that the role of PME was over after making the availability of pectic substances for PG enzyme to act upon. Similar results have also

Table 3. Changes in the firmness (kg/cm²) and activities (units*/gfw/h) of cellulase and pectin methyl esterase (PME) in guava fruits during ripening on-tree.

Harvest stage	Cellulase			PME		
	Hisar Safeda	Hisar Surkha	Mean	Hisar Safeda	Hisar Surkha	Mean
			Rainy season			
GMS	19.88	21.78	20.83	62.31	65.40	63.85
HRS	23.02	25.61	24.31	69.52	75.62	72.57
FRS	23.41	25.80	24.60	65.47	71.27	68.37
Mean	22.10	24.40		65.77	70.76	
r^2 with firmness		-0.99			-0.76	
CD at 5%						
Variety (A) :	1.36				2.56	
Storage period (B) :	1.10				3.02	
A × B :	NS				NS	
			Winter season			
GMS	15.48	17.31	16.39	55.20	60.11	57.65
HRS	18.51	20.69	19.60	60.02	66.33	63.17
FRS	19.62	21.03	20.32	57.82	62.97	60.39
Mean	17.87	19.67		57.68	63.14	
r^2 with firmness		-0.99			-0.67	
CD at 5%						
Variety (A) :	1.25				2.81	
Storage period (B) :	1.50				2.96	
A × B :	NS				NS	

GMS = Green mature stage, HRS = Half ripe stage, FRS = Full ripe stage

1 unit* of cellulase = µg of glucose liberated

1 unit* of PME = meq of ester hydrolysed

been reported earlier in guava (El-Zoghbi, 6; Jain *et al.*, 11), in *ber* (Siddiqui and Sharma, 27), and mango (Ketsa *et al.*, 13).

During the course of present study, cv. Hisar Surkha exhibited faster changes in PME activity and maintained overall higher activity than cv. Hisar Safeda. Fruits of winter season were slower in PME activity than rainy season fruits. Results relating to correlation of PME with loss of firmness were inconsistent with low values of correlation coefficient. Smith *et al.* (26) also observed a little correlation of PME activity with softening of banana fruits.

Guava fruits exhibited more or less similar pattern in the change of the activities of cell wall degrading enzymes except that the changes were rapid during ripening in-storage in comparison to ripening on-tree. A high negative correlation was observed between PG, β-D-galactosidase and cellulase activity and loss of fruit firmness. Thus, these enzymes are involved in softening of guava fruit during ripening, whereas PME plays a lesser role. Cultivar Hisar Surkha exhibited faster changes in the activities of cell wall degrading enzymes as compared to Hisar Safeda owing to its fast ripening character.

REFERENCES

1. Abu-sarra, A.F. and Abu-Goukh, A.A 1992. Changes in pectinesterase, polygalacturonase and cellulase activity during mango fruit ripening. *J. Hort. Sci.* **67**: 561-68.

2. Bartley, I.M. 1978. Exo-polygalctouronase of apple. *Phytochem.* **17**: 213-16.

3. Brady, C.J. 1987. Fruit ripening. *Ann. Rev. Plant Physiol.* **33**: 999-1008.

4. Chin, L.H., Ali, Z.M. and Lazan, H. 1994. Comparative softening of guava fruits. Solubilazation and depolymerization of cell wall carbohydrates during ripening. *Proc. Malayasian Biochem. Soc. Conf.* **19**: 147-50.

Table 4. Changes in the activities (units*/gfw/h) of cellulase and pectin methyl esterase (PME) in guava fruits during ripening in-storage.

Storage (days)	Cellulase			PME		
	Hisar Safeda	Hisar Surkha	Mean	Hisar Safeda	Hisar Surkha	Mean
			Rainy season			
0	19.88	21.78	20.83	62.31	65.40	63.85
2	22.19	24.79	23.49	65.49	70.62	68.05
4	23.44	28.06	25.75	68.38	69.46	65.92
6	24.83	33.61	29.22	65.37	65.81	65.59
Mean	22.58	27.06		65.38	67.82	
r^2 with firmness		-0.96			-0.52	
CD at 5%						
Variety (A) :	1.50				3.10	
Storage period (B) :	1.67				2.42	
A × B :	NS				NS	
			Winter season			
0	15.80	17.56	16.68	55.73	60.38	58.05
2	17.58	19.36	18.47	60.67	65.46	63.06
4	18.52	20.30	19.41	63.56	69.10	66.33
6	19.48	21.31	20.39	61.25	63.66	62.45
8	20.49	22.87	21.60	56.07	59.93	58.00
Mean	18.37	20.28		59.46	63.70	
r^2 with firmness		-0.96			-0.02	
CD at 5%						
Variety (A) :	1.78				3.96	
Storage period (B) :	2.06				2.58	
A × B :	NS				NS	

1 unit* of cellulase = µg of glucose liberated
1 unit* of PME = meq of ester hydrolysed

5. Dick, A.J., Opuka-Gyamfua, A. and DE Marco, A.C. 1990. Glycosidases of apple fruit a multifunctional ß-galctosidase. *Physiol. Plant.* **80**: 250-56.

6. El-Zoghbi, M. 1994. Biochemical changes in some tropical fruits during ripening. *Fd. Chem.* **59**: 33-37.

7. Goukh, Abu and Bashir, H.A. 2003. Changes in pectic enzymes and cellulase activity during guava ripening. *Fd. Chem.* **83**: 213-18.

8. Hobson, G.E. 1963. Pectinesterase in normal and abnormal tomato fruit. *Biochem. J.* **86**: 358-65.

9. Huber, D.J. 1983. Polyuronide degradation and hemicellulose modification in ripening tomato fruit. *J. American Soc. Hort. Sci.* **108**: 405-9.

10. Lannetta, P.P.M., Berg, J. Van Den, Wheatley, R.E., McNicol, R.J. and Davies, H.V. 1999. The role of ethylene and cell wall modifying enzymes in raspberry (*Rubus indaeus*) fruit ripening. *Physiol. Plant.* **105**: 338-47.

11. Jain, Nisha, Dhawan, Kamal, Malhotra, Sarla and Singh, Randhir. 2003. Biochemistry of fruit ripening of guava (*Psidium guajava* L.): Compositional and enzymatic changes. *Plant Fd. Human Nutr.* **58**: 309-15.

12. Ketsa, S. and Daengkanit, T. 1998. Physiological changes during post-harvest ripening of durian (*Durio zibethinus* Murray). *J. Hort. Sci. Biotech.* **73**: 575-77.

13. Ketsa, S., Chidtragool, S., Klein, J.D. and Lurie, S. 1998. Effect of heat treatment on change in

softening in softening, pectic substances and activity of polygalacturonase, pectinesterase and β-galactosidase of ripening mango. *Plant. Physiol.* **153**: 457-61.

14. Lallan, Ram 2001. Physiological and biological changes during fruit maturation of Kinnow mandarin on tree and in-storage. M.Sc. thesis submitted to CCS Haryana Agricultural University, Hisar, India.

15. Lazan, H., Salamat, M.K. and Ali, Z.M. 1995. ß-galactosidase, polygalacturonase and pectinesterase in differential softening and cell modification during papaya fruit ripening. *Physiol. Plant.* **95**: 106-12.

16. Mishra, K. and Sheshdri, T.R. 1968. Chemical component of the fruits of guava (*Psidium guajava* L.). *Phytochem.* **7**: 641-45.

17. Ning, B., Kubo, Y., Inaba, A. and Nakamura, R. 1997. Softening characteristics of Chinese pear 'YaLi' fruit with special relation to change in cell wall polysaccharides and their degrading enzymes. *Scientific Report of the Faculty of Agriculture, Okayama University*, **86**: 71-78.

18. Nunes, M., Cecilia, N., Brecht, Jeffery, K., Morais, Alcina, M.M.B. and Sargent, S.A. 2006. Physicochemical changes during strawberry development in the field compared with those that occur in harvested fruit during storage. *J. Sci. Fd. Agric*. **86**: 180-90.

19. Panse, V.G. and Sukhatme, P.V. 1967. *Statistical Methods for Agricultural Workers*. ICAR, New Delhi, pp. 167-74.

20. Saroja, S., Pushpa, A. and Shanthi, V.P. 1988. A study on the activity of the selected enzymes in the ripening of banana and guava. *Ind. Nutr. Diete.* **25**: 207-11.

21. Selvaraj, Y., Pal, D.K., Edward Raja, M. and Rawal, R.D. 1998. Biochemistry of guava (*psidium guajava* L.) fruit ripening: Changes in respiration rate, ethylene production and enzyme activity. *Indian J. Hort*. **55**: 1-9.

22. Selvaraj, Y., Pal, D.K., Edward, Raja, M. and Rawal, R.D. 1999. Changes in chemical composition of guava fruits during growth and development. *Indian J. Hort*. **56**: 10-18.

23. Sharma, R.K and Siddiqui, S. 2001. Ripening of ber (*Zizyphus mauritiana* Lamk.) on tree and in storage the study of flesh firmness, cell wall composition and found enzymes. Paper presented in *International Conference of Nature Farming and Ecological Balance* (ICNFEB-2001) on March 7-10, 2001 held at CCS HAU, Hisar, India.

24. Siddiqui, S. and Bangerth, F. 1995. Differential effect of calcium and strontium on flesh firmness and properties of cell walls in apple. *J. Hort. Sci*. **70**: 949-54.

25. Siddiqui, S. and Bangerth, F. 1996. The effect of calcium infiltration on the structural changes in cell walls of stored apples. *J. Hort. Sci*. **70**: 295-98.

26. Smith, N.J., Seymor, G.B., Jeger, M.J. and Tucker, G.A. 1990. Cell wall changes in banana and plantains. *Acta Hort*. **269**: 283-89.

27. Siddiqui, S. and Sharma, R.K. 2003. Ripening of ber (*Zizyphus mauritiana* Lamk.) fruits on tree and in-storage: Change in the activities of wall bound enzymes. *Haryana J. Hort*. *Sci*. **32**: 44-47.

28. Tucker, G.A., Robertson, N.G. and Grierson, D. 1980. Changes in polygalacturonase isoenzymes during the ripening of normal and mutant tomato fruits. *European J. Biochem*. **112**: 119-24.

29. Tucker, G.A. and Grierson, D. 1987. **In**: *The Biochemistry of Plants*, Vol. 12 Daries, D. (Ed.) Academic Press, New York, pp. 265-19.

Received : February, 2011; Revised : January, 2012; Accepted : April, 2012