# Influence of selenium and chitosan on physico-chemical properties of guava (Psidium guajava) under storage

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#### **ABSTRACT**

Guava (*Psidium guajava* L.) is an important fruit cultivated in tropical and sub-tropical countries. Being a climacteric fruit it has very high respiration and deterioration rates (shelf-life 3-4 days), making a challenge for storage. Therefore, the present experiment was conducted during 2016–17 to reduce the post-harvest losses and extension of shelf-life of guava fruits var. Hisar Surkha using the combination of selenium and chitosan treatments. The mature green freshly harvested guava fruits were treated with selenium solutions (0.01 ppm, 0.02 ppm, and 0.05 ppm) in combination with 1.5% chitosan containing 0.5% acetic acid for 5 min and stored at room temperature (20±2°C, 82±5% RH). The fruits were analyzed for physico-chemical attributes (physiological weight loss, fruit firmness, total soluble solids, and titratable acidity) and biochemical characteristics (total phenolic, flavonoid contents, and total antioxidant activity) at three days interval. The combined treatments of selenium and chitosan helped in delaying weight loss (9.66%), lower disease incidence, better retention of fruit firmness (4.70 kg/cm²), soluble solids (13.03 °Brix), titratable acidity (0.38%) compared to control on 12<sup>th</sup> day of storage. Treatment of guava fruits with selenium and chitosan also maintained higher total phenols (24.99 mg GAE/g dry wt.), flavonoids (3.51 mg CE/g dry wt.) and exhibited higher antioxidant activity (54.32%) throughout storage. The present study showed that combination of Se (0.02 ppm) and chitosan (1.5%) was most effective in maintaining post-harvest quality and enhance the shelf life of guava fruits up to 12 days during storage at room temperature.

**Key words:** Chitosan, Physico-chemical characteristics, Selenium, Storage and quality

Guava (Psidium guajava L.) is a good source of Vitamin C, dietary fiber, carotenoids, pectin, sugars, phenolic substances, and minerals such as phosphorus, iron, and calcium (Correa et al. 2011). Guava fruit matures and becomes soft very quickly after harvest and within 3-4 days, loses its texture and appearance at room temperature. Guava fruits contain a high amount of water which is associated with a high metabolic activity that lasts post-harvest and makes it very vulnerable to injury (Bassetto et al. 2005). The fresh produce also susceptible to post-harvest diseases that further reduces the quality during storage (FAO 2011). In developing countries like India, the harvesting techniques, including post-harvest infrastructure and handling are poor thus heavy losses occur during fruit distribution and marketing. There are some possible mechanisms to enhance the shelf-life of fruits by checking the rates of transpiration, respiration and microbial infestation. Though post-harvest changes in fresh fruit cannot be stopped, these can be slowed down by using preservation and management techniques which maintain

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the quality and enhance the shelf-life of fruits within certain limits. A number of chemicals and coatings were used to retard the process of fruits ripening to extend their shelf-life. These coatings have ability to modify the atmospheric conditions by forming a semi-permeable membrane which helps in reducing the weight loss, respiration rate and ethylene production (Ali *et al.* 2011).

Chitosan acts as a selective barrier to transpiration thereby limiting the rate of respiration, ethylene evolution, ascorbic acid loss, enzymatic browning, softening and delaying the ripening process (Bautista-Banos *et al.* 2006). On the other hand, selenium is also an important element which effectively retards ethylene production hence improves the shelf life of fruits and vegetables and maintained their quality (Liu *et al.* 2012). Literature survey revealed that no study has been published yet on the combined applications of selenium and chitosan on the post-harvest quality and shelf-life of guava fruits. The present study was aimed to investigate the effect of selenium and chitosan treatments on physico-chemical characteristics, disease incidence and quality attributes of guava fruits during storage at ambient condition.

## MATERIALS AND METHODS

Mature green, fresh, uniform size and shape fruits

of guava var. Hisar Surkha were harvested from the Horticultural farm, CCS Haryana Agricultural University, Hisar in the morning hours during 2016–17. The fruits were transported to the Department of Biochemistry in plastic bags, where they were sorted on the basis of uniform maturity, size and free of any visible defects. The selected fruits were washed to remove dust and dirt with water and dried using tissue paper. The fruits were treated with  $T_1$ : selenium (0.01 ppm) + chitosan (1.5%),  $T_2$ : selenium (0.02 ppm) + chitosan (1.5%),  $T_3$ : selenium (0.05 ppm) + chitosan (1.5%) and  $T_4$ : control (water) for 5 min.

For selenium treatment, fruits were dipped for 5 min in solution of 0.01 ppm, 0.02 ppm and 0.05 ppm of sodium selenite. The chitosan treatment was given by dipping the fruits in a solution of 1.5% (w/v) chitosan consisting of 0.5% acetic acid (v/v). The fruits dipped in water for 5 min were used as a control. These treated fruits were allow to air dry and stored in cardboard boxes under ambient condition (20±2°C, 82±5% RH). The combined effect of both the treatments on physico-chemical and biochemical attributes of guava fruits were taken on same day of harvest and after 3, 6, 9 and 12 days of storage.

The physiological loss in weight (PLW) of guava fruits during storage was determined on initial weight basis by calculating the difference in initial weight and the subsequent weight at the time of observation and expressed in per cent. Fruit firmness was measured by hand held fruit pressure tester penetrometer tester (Model FT 327; TR Agricoli, Italy) and expressed in kg/cm<sup>2</sup>. Total soluble solids of fruit were measured with the help of Hand Refractometer (0-32 °brix range). Titratable acidity was estimated by method of Ranganna (2003) and results were expressed as the citric acid per cent. Total phenolics were determined by the method of Swain and Hillis (1959) and results were reported as mg gallic acid equivalents (GAE)/100 g dry weight. The flavonoid content was determined by the method of Delcour and De Varebeke (1985) and results were presented as mg catechin equivalents (CE)/100 g (mg CE/100 g) of dry sample. Total antioxidant activity was determined by the method of Shimada et al. (1992) using stable 2, 2-diphenyl-1picrylhydrazyl (DPPH) radical. The results of total antioxidant activity were expressed as per cent scavenging capacity of DPPH. Disease incidence was calculated by total plate counts of bacterial and fungal colonies on nutrient agar medium and dextrose agar medium, respectively and expressed as colony forming units (CFU). The findings of the present study were statistically analyzed using two factorial completely randomized designs (CRD). The sources of variation were storage duration, treatments, and their interactions. The means of different treatments were compared at 5% levels of significance. All experiments were replicated three times and analyzed using IBM SPSS Statistics® Version 25 software.

## RESULTS AND DISCUSSION

*Physiological loss in weight (PLW)*: The loss in weight during storage in fresh fruits and vegetables is principally due to the continuous transpiration and respiration processes.

The PLW increased progressively and significantly as the period of storage increased from 3<sup>rd</sup> day to 12<sup>th</sup> day in guava fruits (Table 1). The maximum weight loss of 17.49% was recorded in the control fruits after the 12<sup>th</sup> day of storage. Among the different treatments, the application of Se (0.02)ppm) + chitosan (1.5%) was most effective in delaying the PLW and showing 45.05% decline in PLW as compared to control fruits. The PLW exceeded the 10% threshold level in control (12.86%) at 9th day after storage, whereas the fruit treated with the combination of Se (0.02 ppm) + chitosan (1.5%) remained storable even up to the 12<sup>th</sup> day after storage (9.66%). The present investigation exhibited that selenium application at lower concentrations was found more effective in reducing weight loss by delaying the ripening processes. The findings of the present study are in agreement with the previous findings of chitosan treatment in guava (Anggarwulan et al. 2015) and papaya (Ali et al. 2011).

Fruit firmness: The firmness of guava fruit decreased significantly and progressively, irrespective of treatments with the increasing storage period (Table 1). The texture and appearance of the guava fruits at the time of harvest (0 day) and at 12th day of storage were showing visible differences (Fig 1). Among different treatments, the fruits treated with Se 0.02 ppm + chitosan 1.5% significantly maintained the texture and appearance up to 12th day during storage. This might be due to changes in cell wall components and the turgor pressure of the cells (Chen et al. 1983). The lower concentrations of Se (0.01 ppm and 0.02 ppm) in combination with chitosan (1.5%) were most effective in delaying the softening of fruits by maintaining their firmness up to 4.00 kg/cm<sup>2</sup> and 4.70 kg/cm<sup>2</sup> respectively, after the 12<sup>th</sup> day of storage. The chitosan formed a defensive layer over the surface of fruit thus reduced rate of respiration, transpiration, delayed ethylene production thus maintained more tissue rigidity. The lower selenium concentration was effective in delaying plant senescence due to decreasing the ethylene production rate thus slowed down the rate of fruit softening. The results of chitosan treatment obtained in the present study are in close conformity with the findings of Zhu et al. (2008) in mango, Ali et al. (2011) in papaya, Zahran et al. (2015) in pomegranate arils and Petriccione et al. (2015) in strawberry.

Total soluble solids (TSS): The TSS content was increased progressively up to the 6<sup>th</sup> day of storage and declined thereafter irrespective of treatment (Table 1). The initial increase in TSS may be due to hydrolysis of complex polysaccharides into simple sugars by hydrolytic enzymes which further uses in the process of respiration and the level reduces under consecutive storage. The reduction in TSS was significantly less and slower in the treated fruits as compared with control. Among the treatments, selenium (0.02 ppm) + chitosan (1.5%) was most effective in maintaining the TSS content. The chitosan treatment delay in increase in TSS might be due to slowing down of metabolic and respiration and could delay the ripening process. These results are consistent with those of other studies concerning the effects

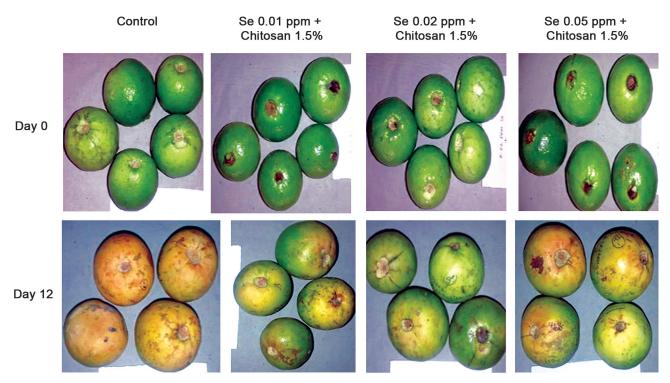


Fig 1 Guava fruits treated with chitosan and selenium at 0 and 12 days of storage.

of chitosan treatment on different fruits, such as mango and banana (Kittur *et al.* 2001), papaya (Ali *et al.* 2011) and pomegranate arils (Zahran *et al.* 2015).

Titratable acidity: The titratable acidity is used to measure the organic acid content of pulpy fruits. A progressive decline in titratable acidity observed in guava fruits during storage (Table 2). The acidity of the fruits decreased due to continuous consumption of organic acids in the respiration or due to metabolic changes during storage (Ghasemnezhad et al. 2011). Maximum titratable acidity (0.382%) was found in fruits treated with Se (0.02 ppm) + chitosan (1.5%), whereas control fruits had minimum titratable acidity (0.303%) at 12<sup>th</sup> day after storage. This effect of Se and chitosan might be due to the decrease in cellular metabolic activities such as respiration thereby, preventing loss of organic acids and hence, decreasing

the loss of acidity. Similar results with chitosan treatment have also reported in peach and litchi (Han *et al.* 2014) and pomegranate arils (Zahran *et al.* 2015).

Total phenolic, flavonoid contents and antioxidant activity: The application of selenium and chitosan treatment also influenced total phenolic, flavonoid and antioxidant capacity in guava during storage. The amount of total phenols and total flavonoids (Table 2) increased up to 3<sup>rd</sup> day and declined thereafter irrespective of treatment. All the treated fruits have higher phenols and flavonoids content as compared to control. Among the different treatments, Se (0.02 ppm) + chitosan (1.5%) were more effective in maintaining phenolic (24.99 mg GAE/g dry wt.) and flavonoid content (3.83 mg CE/g dry wt.) of guava fruits at 12<sup>th</sup> day of storage. These results are consistent with previous studies demonstrating that chitosan treatment improved

Table 1 Effect of chitosan and selenium on physiological loss in weight, firmness and total soluble solids in guava fruit during storage

Treatment	Physiol	ogical lo	ss in we	ight (%)		Firm	ness (kg/	/cm <sup>2</sup> )		Total soluble solids (°Brix)					
	3 Day	6 Day	9 Day	12 Day	0 Day	3 Day	6 Day	9 Day	12 Day	0 Day	3 Day	6 Day	9 Day	12 Day	
$T_1$	2.35	5.45	8.35	11.31	11.40	9.07	6.63	5.13	4.00	11.37	12.33	13.67	13.87	13.73	
$T_2$	1.63	4.15	7.76	9.66	11.40	9.37	6.87	5.43	4.70	11.37	12.10	13.37	13.43	13.03	
$T_3$	2.86	7.36	10.74	12.68	11.40	6.90	3.43	3.00	2.67	11.37	13.50	14.60	12.46	12.10	
$T_4$	3.30	8.79	12.86	17.49	11.40	6.70	3.33	2.23	1.40	11.37	13.27	14.37	12.63	11.60	
SEm±	$a \to 0.157, b \to 0.157,$ $a \times b \to 0.313$				$a \rightarrow 0$	.088, b -	→ 0.079,	a x b	0.176	$a \to 0.064, b \to 0.057, a \times b \to 0.127$					
CD (P≤0.05)	a –	$a \to 0.453, b \to 0.453,$ $a \times b \to 0.906$				.253, b -	→ 0.226,	a x b	→ 0.506	$a \to 0.182, b \to 0.163, a \times b \to 0.365$					

<sup>\*</sup>a =treatments, b =days of storage,  $a \times b =$ interaction

Table 2 Effect of chitosan and selenium on titratable acidity, total phenolics and flavonoids in guava fruit during storage

Treatment	Titratable acidity (%)					Total p	henolic	s (mg G	AE /g d	ry wt.)	Flavonoids (mg CE/g dry wt.)				
	0 Day	3 Day	6 Day	9 Day	12 Day	0 Day	3 Day	6 Day	9 Day	12 Day	0 Day	3 Day	6 Day	9 Day	12 Day
$T_1$	0.527	0.512	0.433	0.425	0.356	22.28	26.19	25.92	25.41	25.00	3.36	6.26	5.48	4.32	3.51
$T_2$	0.527	0.521	0.450	0.429	0.382	22.28	26.48	26.11	25.59	24.99	3.36	6.50	5.83	4.55	3.83
$T_3$	0.527	0.469	0.438	0.390	0.324	22.28	25.25	24.79	24.41	24.06	3.36	5.74	5.13	3.48	3.05
$T_4$	0.527	0.439	0.397	0.358	0.303	22.28	25.55	24.91	24.23	23.63	3.36	5.25	4.47	3.36	2.52
SEm±	$a \rightarrow 0$ .	004, b -	→ 0.004	, a x b -	→ 0.008	$a \rightarrow 0$ .	071, b -	→ 0.064,	a x b -	→ 0.142	$a \rightarrow 0.0$	053, b -	→ 0.048,	, a x b -	→ 0.107
CD (P≤0.05)	$a \rightarrow 0$ .	011, b –	→ 0.010,	a x b -	→ 0.023	$a \rightarrow 0.2$	204, b-	→ 0.182	, a x b -	→ 0.408	$a \rightarrow 0$ .	153, b -	→ 0.137,	, a x b -	→ 0.307

a, treatments; b, days of storage; a × b, interaction

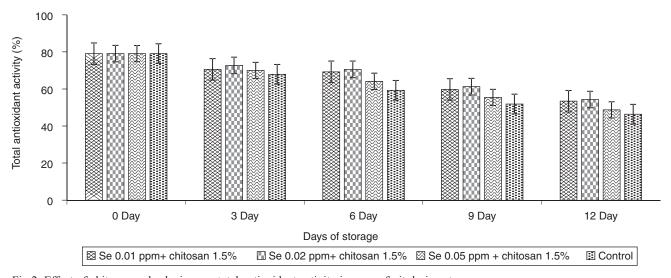


Fig 2 Effect of chitosan and selenium on total antioxidant activity in guava fruit during storage.

the nutraceutical properties of fruits by maintaining high levels of phenols, anthocyanins and flavonoids during post-harvest (Wang and Gao 2013). Selenium has also been defined to induce antioxidant capacity in plants, by increasing tocopherol and phenolic compounds may be due to its ability to inhibit the reaction of polyphenol oxidase enzyme thus retains more phenolic and flavonoid content (Xu et al. 2003). The total antioxidant activity decreased progressively in control as well as treated guava fruits throughout the storage (Fig 2). The fruits treated with Se (0.02 ppm) + chitosan (1.5%) had maximum total antioxidant activity. These results are in agreement with those reported earlier in chitosan treated strawberry fruit during storage (Petriccione et al. 2015).

Disease incidence: In this study, a combination of 0.02 ppm Se and 1.5% chitosan was found highly effective against bacterial and fungal growth in guava till 12th day of storage. The total plate counts of bacterial and fungal colonies (CFU) were maximum in the fruits under control conditions, which had uncountable bacterial (>1010) and fungal (>10<sup>12</sup>) colonies. Significant reduction of storage rots has been recorded in apples, kiwifruit, pears treated with chitosan (Bautista-Banos et al. 2004). Se treatment also significantly inhibited spore germination of the fungal pathogen and effectively controlled gray mold in harvested tomato fruit (Wu et al. 2016).

It can be concluded from the present investigation that the combined treatments of selenium (0.02 ppm) and chitosan (1.5%) was most efficient in maintaining the fruit quality and extending the fruit storability up to 12 days under ambient storage conditions. However, the higher concentrations of selenium in combination with chitosan had a negative effect on quality attributes during storage due to its pro-oxidant activity at higher concentration. Thus, lower selenium concentrations with chitosan can be used to extend the storability, post-harvest life, and quality of guava fruits under ambient storage conditions.

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