

Novel ready to serve beverage from green tomato

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ABSTRACT

Green tomato is a rich source of α -tomatine, which has anti-carcinogenic, antibiotic and cholesterol reducing effects in the human body. Therefore, present study was aimed to develop healthy ready to serve (RTS) green tomato beverage with α -tomatine content 10 to 11 mg/100 mL. The juice of green tomato was extracted and used at different concentrations (10-14%) with sugar (10-14°B) and citric acid (0.1-0.3% acidity) to standardize the best consumer acceptable formulation depending on the highest overall acceptability and vitamin C retention. Ginger, lemon juice and black salt each were also added at 1% concentration to increase acceptability of the product. Standardized formulation was further used to optimize the preservation process (thermal treatment with and without addition of preservative). The formulated RTS green tomato beverage processed with thermal treatment at 80°C for 2 min without preservative contained α -tomatine 10.40±0.07 mg/100 mL was judged to be highly acceptable for the overall acceptability and retained significant (p<0.01) amount of vitamin C, antioxidant activity and phenolic compounds with respect to the control. Further, loss of tomatine content was non-significant for 40 days of its refrigerated (7±2°C) storage.

Key words: Solanum lycopersicum L., α-tomatine, RTS beverage

INTRODUCTION

Tomato, belongs to the genus *Lycopersicon* (botanic family *Solanaceae*) is very versatile and popular agricultural crop grown globally. During the ripening process, tomato undergoes many physico-chemical changes, for instance, the green color of tomato changes to yellow, orange and then to red. The green (raw) tomato contains about 100 times higher tomatine (500 mg per kilogram of tomato weight) than that of fully ripe tomatoes (Friedman, 5). Therefore, composition of tomato depends upon its maturity.

Tomatoes contain both tomatine and dehydrotomatine from which tomatine exists in the form of α -tomatine (Kozukue *et al.*, 11). This α -tomatine has anti-carcinogenic and antibiotic effects on human pathogens like leukemic cells, virulent bacteria and protozoa. Furthermore, it can reduce cholesterol levels in the body (Friedman, 6), and has hemolytic activity against red blood cells (Nepal and Stine, 15). When green tomatoes are consumed, the α-tomatine present in green tomatoes is hydrolyzed to form tomatidine that can strengthen muscles. The steroidal compounds present in unripe green tomatoes prevent muscle atrophy, increase strength along with the endurance (Dyle et al., 4). However, when green tomato ripens, tomatine degrades, and the content of tomatine reduces significantly. In contrast, tomatine will not be affected or reduced

by food processing techniques such as frying or microwave (Meher and Gaur, 13).

The time has arrived to make daily foods nutritionally rich and easy to use to meet the increased demand for functional and nutritional foods in this contemporary era. Green tomatoes would be an aided ingredient in the context of functional and health foods (Dyle et al., 4). Additionally, consumption of green tomato products is not as widespread as that of red tomato. However, the synthetic juices which are rich in artificial sweeteners such as sugar can also be replaced with healthy green tomato juice. Furthermore, natural beverages have become popular today due to their pleasant taste, natural aroma and supportive effect on health. In general, tomato plants produce five different stages of tomato at the time of harvest viz. mature green, turning, pink, light red and red ones. This is one of the major impediment for mechanical harvesting of tomatoes. Manual picking of tomatoes based on maturity stages is labour intensive. However, in mechanical harvesting a significant quantity of small, immature, green tomatoes find very little market value. As these green tomatoes are very rich in heat stable bio-active components viz. α-tomatine, an attempt was made with the objective to utilize these for production of a high value beverage blending with ginger and lime.

MATERIALS AND METHODS

Mature green (20-25 days after flowering with white, star-shaped zone on its bottom end) tomato

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fruits of cv. Gujarat Anand Tomato 5 (GAT-5) without any external red colour were harvested for the experiment in the first week of the February 2021. Fruits were sorted for uniform colour and free of any external injuries. Fresh lemon, ginger, sugar, black salt and citric acid were obtained from the local market to prepare the ready-to-serve (RTS) green tomato beverage.

To prepare RTS green tomato beverage mature green tomato were washed with running water to remove adhered soil and contamination. Washed tomatoes were subjected to hot water blanching at 82°C for 15 seconds (Gould, 8). Dark green coloured juice was extracted from the blanched tomato pieces using centrifugal juicer and straining using double layered and quadruple layered muslin cloth. RTS green tomato beverage having TSS 4.40± 0.10 °B was prepared from this filtered juice. Additionally, lemon and ginger juice, and black salt were used to improve the taste of the beverage. Finally, the green tomato juice(GTJ) was mixed into the sugar syrup along with 1% ginger juice, 1% lemon juice and 1% black salt in order to increase the acceptability of the RTS green tomato beverage. Subsequently, hot filling was carried out at 85°C in 200 mL in pre-sterilized glass bottles to store the prepared beverage. Finally, filled bottles were crown corked and refrigerated (5-6°C).

To standardize formulation of RTS green tomato beverage was formulated using different concentrations of green tomato juice (10, 12 and 14%) with varying total soluble solids (10, 12 and 14 °B) and acidity (0.1, 0.2, and 0.3%) as shown in Table 1. Water was also added to maintain the desirable TSS in RTS beverage. The prepared beverage was evaluated after adequate cooling to standardize its formulation based on the responses recorded for vitamin C content, viscosity and sensory score in terms of overall acceptability (OA).

As shown in Table 1, preservation process variables were optimized for extended shelf life of the prepared RTS green tomato beverage. It was filled hot in glass bottle at 85°C temperature with and without the addition of preservative viz. sodium benzoate (SB). Furthermore, the filled bottles were thermal treated at different temperatures and time intervals as shown in Fig. 1. Both thermally and thermo-chemically processed RTS green tomato beverage samples were analyzed for its phenolics content, vitamin C content, antioxidant activity, tomatine, microbial load and sensory score. During processing of prepared beverage, control sample was prepared for comparative study.

Tomatine present in freshly prepared the juice and formulated RTS beverage was determined by direct reaction of tomatine with the anthrone reagent using the spectrophotometric method as described by Socic (19). Proximate composition (protein, crude fat, crude fibre, ash, moisture and total sugar), vitamin C content, β -carotene content and microbial load (viz. yeast and mould count, total plate count, and coliform count) was estimated using analytical method described by Ranganna (17).

Furthermore, acidity and TSS of the filtered GTJ and prepared beverage were measured as per methods described by Vahora *et al.* (20). Nine point hedonic scale was used for judging sensory attributes for overall acceptability by a panel consisting of 20 semi-trained judges. Viscosity of the beverage was measured using Brookfield viscometer (Model: DV-II+Pro) Brookfield Engineering Technology, Inc., USA with S-2 spindle at 100 rpm. The colour was measured

Parameters	Levels	Responses	
Formulation variables			
Green tomato juice (GTJ, %)	10, 12, 14	Sensory score	
Total soluble solids (TSS, °B)	10, 12, 14	Viscosity	
Acidity (%)	0.1, 0.2, 0.3	Vitamin C content	
Ginger juice (%)	1		
Lemon juice and (%)	1		
Black salt (%)	1		
Process variables			
Sodium benzoate (SB, ppm)	0, 120	Vitamin C retention	
Thermal processing (°C)	80, 85, 90	Phenolics content	
Time (min)	2, 4, 6	Antioxidant activity	
		Microbial load	

 Table 1. RTS green tomato beverage formulation and process variables.



Fig. 1. Preparation of RTS green tomato beverage.

using Lovibond colorimeter (Model: RT850i) (Vahora *et al.*, 20). Spectrophotometric Folin–Ciocalteau method described by Dhanani *et al.* (3) was used to estimate phenolics content. Antioxidant activity was determined using UV spectrophotometer as per methodology described by Dhanani *et al.* (3).

The data obtained for the various responses were analysed statistically and expressed as mean values ± standard deviations of three replications. Factorial completely randomized design (FCRD) was employed to evaluate the consequence of independent variables on its dependent variables. Statistical package software viz. Design-Expert version 7.0.0 (Stat-Ease, Minneapolis, USA) was employed to carry out the statistical analysis of experimental data. Total eighty-one experiments were conducted and their interactions were also studied to standardize the beverage formulation and fifty-four experiments to optimize the preservation process at 1% significance level.

RESULTS AND DISCUSSION

Data on proximate composition and other chemical attributes of the GTJ are presented in Table 2. The results obtained by Fuentes et al. (7) for the proximate composition of the GTJ showed some differences with the present data. This may be due to difference in variety of tomato and its agroclimatic condition. The data on chemical composition such as TSS and acidity has a reasonable agreement with the results obtained by Chomchalow et al. (2) for mature green tomato of "Sunny" variety whereas comparatively pH showed the lower value. Furthermore, vitamin C which is one of the major natural antioxidants present in the tomato fruit was found to be 10.35±0.02 mg/100 mL. Accordingly, the present study showed that GTJ contained 10.47 ± 0.09 mg/100 mL tomatine while Leonardi et al. (12) obtained 10.1 mg/kg of fresh weight for a cluster variety of tomato. The antioxidant activity and phenolics of GTJ were in reasonable agreement to the finding of Fuentes et al. (7).

Sensory profile of the product ascribes the acceptability and market potential of the product. In consideration to this, sensory evaluation of prepared RTS green tomato beverages was carried out. From the sensory evaluation of the control (GTJ) juice it was found that it had acceptable sour taste. Additional initial trials were conducted with different levels of ginger juice, lemon juice and black salt to improve its taste further and 1% level of each variable was found to be optimum. Mean data showing variations in OA score with respect to different levels of TSS, acidity and GTJ content are shown Fig. 1(a). Obtained result indicated that increased concentration of sugar had a positive effect on sensory attributes. However, above 12 °B TSS, it exerted a negative effect leading to lowest OA score of 6.83, observed for 14 °B TSS sample due to sweetest taste among all the samples. Similar trend was noted for variation in acidity of the beverage. In contrast, incorporation of the high amount of GTJ had increased OA. Maximum OA score (8.17) was recorded for 12 °B TSS, 0.2% acidity and 14% GTJ content sample. It might be due to combination of sugar syrup and high GTJ in the formulation, which led to best sugar-acid blend

Table 2. Proximate and chemical attributes of the green (unripe) tomato juice.

Characteristics	Mean value±SD (n=3)			
Proximate composition	1			
Moisture (%)	96.08±0.03			
Protein (%)	0.69±0.01			
Fat (%)	0.51±0.08			
Fiber (%)	1.06±0.01			
Ash (%)	0.15±0.01			
Carbohydrate (%)	1.51±0.04			
Chemical composition	'n			
TSS (°B)	4.40± 0.10			
рН	3.96± 0.11			
Acidity (% citric acid)	0.50± 0.01			
Vitamin C (mg/100 mL)	10.35± 0.02			
β - carotenoids	0.44± 0.03			
(mg/100 mL)				
Antioxidant activity (µmol Fe ⁺² /mL)	24.05± 0.23			
Phenolic content	25.81± 0.13			
(mgGAE/100 mL)				
Tomatine (mg/100 mL)	10.47± 0.09			
Colour value	L* a* b*			
	18.85±0.06 -3.30±0.07 6.60±0.07			

All values are mean value of three replications





Fig. 1. (a) OA, (b) Vitamin C content and (c) Viscosity of different formulated RTS green tomato beverages.

in the product. Similar observation was previously reported by Hamid et al. (9) for a RTS beverage from mulberry fruits. However, results revealed that all the formulations were found acceptable by the panelists and score varied from like slightly (6.83) to like very much (8.17) on the 9 point hedonic scale. Furthermore, addition of ginger juice, lemon juice and black salt in RTS green tomato beverage aided to improve acceptability. Sindhumati and Premalatha (18) reported that the papaya blended RTS flavored with cardamom and ginger was preferred over plain papaya RTS beverage. Statistically, it was found that the acidity and GTJ content had non-significant effect on the OA, whereas combined effect of TSS and GTJ content as well as acidity and GTJ content significantly (p<0.01) influenced the acceptability score. Data showing the vitamin C content of the prepared beverage samples due to variation in TSS, acidity and GTJ are depicted in Fig. 1(b). Vitamin C content was found in the range of 3.25 to 5.97 mg/100 mL. GTJ was the prime source of vitamin C in the formulation of the RTS green tomato beverage. Therefore, increase in concentration of GTJ in the formulation increased vitamin C in the final product. Accordingly, the beverage sample having 14% GTJ content had the highest amount of vitamin C. Similar trend was observed by Afreen et al. (1) during the preparation of RTS beverage form carrot with addition of sour-orange juices with different levels. ANOVA test indicated that, the GTJ concentration had the most significant effect (p<0.01) on vitamin C content in the final product followed by acidity and TSS. Moreover, among all interaction effects, only GTJ content and TSS influenced the availability of vitamin C in the prepared RTS beverage.

Viscosity of prepared RTS green tomato beverage was analyzed at 25±1°C temperature and the results are presented in Fig. 1(c). Experimental results revealed that with the increase in TSS, acidity and GTJ content, viscosity of the beverage was increased. It was found between 11.73 to 13.60 cP. This phenomenon can be attributed to the interaction of sugar, citric acid and GTJ to form a gel structure which had a crucial role in the alteration of rheological property of the RTS beverage. Hence, RTS green tomato beverage contained 14 °B TSS, 0.3% acidity and 14% GTJ had maximum viscosity, while minimum viscosity was noted for 10 °B TSS, 0.1% acidity and 10% GTJ containing beverage. Hemalatha et al. (10) observed a similar trend for RTS beverage from Cape gooseberry. Statistical analysis of the data indicated that two independent formulation variables i.e., TSS and GTJ content had highly significant (p<0.01) effect on the viscosity. Correspondingly, the interaction effects of TSS and GTJ content, and TSS and acidity significantly (p<0.01) influenced viscosity of the final beverage.

Data on quality characteristics (OA and vitamin C content) of different formulations were statistically evaluated to standardize RTS green tomato beverage formulation variables viz. TSS, acidity and GTJ content. With desirability of 0.898, a recipe of 12 °B TSS, 0.2% acidity and 14% GTJ content was obtained as a standardized formulation, having OA 8.17 and vitamin C content 5.89 mg/100 mL. Additionally, the final formulation contained 1% black salt, 1% ginger juice and 1% lemon juice. Further, it is well known fact that processing and preservation technique has substantial effect on the quality characteristics of the food products. Hence, this formulation was carried forward to prepare beverage for optimization of its preservation parameter.

Influence of thermal and thermo-chemical treatment, used to make RTS beverage safe for consumption and to avail extended storage life, on retention of vitamin C in processed RTS beverages is presented in Table 3 which shows significant (p<0.01) decrease in retention of vitamin C with increase in

temperature and time of thermal treatment. Maximum vitamin C retention, 2.06 mg/100 mL was observed at 80°C temperature and 2 min and minimum 0.88 mg/100 mL at 90°C for 6 min among the thermally treated beverage samples. Correspondingly, thermochemically processed RTS beverages exhibited analogues trend with respect to thermal time and temperature. However, thermally treated beverages retained higher amount of vitamin C as compared to thermo-chemically treated beverages. Vitamin C content of control sample was 2.58 mg/100 mL, 20.16% higher than the thermally treated beverage. ANOVA analysis indicated that all processing variables i.e., temperature, time and preservative significantly (p<0.01) influenced the retention of vitamin C independently. Vitamin C is heat sensitive and degrades when heat is applied to product. Hence, low temperature process for short time retained maximum vitamin C in the prepared beverage.

Phenolics contribute to the protection against degenerative diseases. Previous study by Nisar *et al.* (16) reported that pasteurization and thermochemical treatment decreased phenolic compounds

	Time (min)	Vitamin C (mg/100 mL)	Phenolics content (mgGAE/ 100	Antioxidant activity (µmol	OA	Total Plate Count	Yeast and mold count	
			mL)	Fe⁺²/mL)				
Thermal treatment								
80°C	2	2.06 ± 0.06	12.47 ± 0.11	4.58 ± 0.01	7.83 ± 0.29	< 15	< 2	
	4	1.98 ± 0.00	11.54 ± 0.16	3.29 ± 0.31	7.67 ± 0.29	< 10	< 2	
	6	1.16 ± 0.29	9.52 ± 0.09	2.61 ± 0.12	7.33 ± 0.29	< 5	ND	
85°C	2	1.98 ± 0.00	9.34 ± 0.14	3.19 ± 0.25	7.5 ± 0.50	< 10	ND	
	4	1.65 ± 0.16	7.17 ± 0.55	2.25 ± 0.09	7.5 ± 0.58	< 5	ND	
	6	0.99 ± 0.00	4.55 ± 0.02	1.42 ± 0.07	7.33 ± 0.50	< 5	ND	
90°C	2	1.87 ± 0.19	7.52 ± 0.21	3.09 ± 0.00	6.83 ± 0.29	ND	ND	
	4	1.19 ± 0.21	5.17 ± 0.02	1.37 ± 0.25	6.83 ± 0.58	ND	ND	
	6	0.88 ± 0.00	2.99 ± 0.03	0.42 ± 0.00	6.50 ± 0.00	ND	ND	
Thermochemical treatment								
80°C + 12	0 2	2.01 ± 0.04	12.61 ± 0.32	4.59 ± 0.01	7.33 ± 0.29	< 5	ND	
ppm SB	4	1.65 ± 0.57	11.75 ± 0.13	3.27 ± 1.34	7.00 ± 0.00	< 5	ND	
	6	0.99 ± 0.00	9.56 ± 0.10	2.84 ± 0.16	7.00 ± 0.00	ND	ND	
85°C + 12	0 2	1.87 ± 0.19	9.47 ± 0.00	3.11 ± 0.29	7.17 ± 0.58	ND	ND	
ppm SB	4	1.35 ± 0.46	7.40 ± 0.40	2.29 ± 0.24	7.17 ± 0.29	ND	ND	
	6	0.91 ± 0.00	4.71 ± 0.10	1.42 ± 0.00	6.67 ± 0.29	ND	ND	
90°C + 12	0 2	1.68 ± 0.29	7.82 ± 0.33	3.15 ± 0.08	7.00 ± 0.50	ND	ND	
ppm SB	4	1.16 ± 0.26	6.06 ± 0.45	1.40 ± 0.13	6.67 ± 0.29	ND	ND	
	6	0.88 ± 0.00	3.02 ± 0.62	0.47 ± 0.06	6.17 ± 0.29	ND	ND	

Table 3. Quality attributes of processed RTS green tomato beverage.

All above values are mean value of three replications; ND=Not detected

in apple juice. Accordingly, the data pertaining to degradation of phenolic compounds in RTS beverage samples as shown in Table 3 signified similar effect of thermal and thermo-chemical treatment on the phenolics content of processed RTS green tomato beverages. Nevertheless, thermo-chemical treated beverages had more retention of phenolic compounds than the thermal treatment and maximum 12.61 mgGAE/100 mL phenolics content was found in the beverage contained 120 ppm sodium benzoate and processed at 80°C temperature for 2 min. However, the interaction effect of preservative, temperature and time was found to be non-significant at 1% (p<0.01) significance level whereas individual and interaction effect of temperature and time were found to be significant (p<0.01). In addition, individual effect of preservative was found to be significant (p<0.01). The degradation of phenolic compounds might be due to the complex physical and chemical reactions which formed byproducts from polyphenols that reacted with organic acid or carbonyl compounds, for instance, furfurals due to thermal processing.

From Table 3, RTS beverage, processed with and without preservative at 80°C for 2 min had maximum antioxidant activity, 4.59 and 4.58 µmol Fe⁺²/mL, respectively. Both thermal and thermo-chemical processing dropped the antioxidant activity of the beverages. The antioxidant activity of control sample was found to be 5.21±0.23 µmol Fe⁺²/mL. However, in consideration to control sample reduction of antioxidant activity was significantly low, 11.90 and 12.09% for process variable combinations 80°C temperature for 2 min as compared to 90°C for 6 min with and without addition of preservative, respectively. During thermal processing degradation of vitamin C and phenolic compounds dropped antioxidant activity. Antioxidant activity of RTS beverages was dependent on its processing temperature, time and their interaction (p<0.01).

Sensory quality of the treated beverages was assessed for optimization of process parameter. Result reported in Table 3 revealed that thermally treated beverage samples were preferred over thermochemically treated samples for each respective treatment. Maximum OA score of 7.83 was obtained for sample treated at 80°C for 2 min, whereas minimum score 6.5 was at 90°C for 6 min with and without preservative. Addition of preservative in the thermochemical treatment altered flavour of the beverage leading to lower sensory score. Moreover, thermal treatment with preservative may formed benzene, which is unwanted compound in the product. In addition, loss of vitamin C also leads to the decrease in OA (Nagy et al., 14). In contrast, thermal processing produced undesirable flavours due to degradation of flavour components such as phenolic acids, essential oils, lipids, sugars, sulfur-containing compounds and ascorbic acid (Nagy *et al.*, 14). As a result, high processing temperature for longer time reduced sensory score. In comparison to control, OA score of RTS green tomato beverage processed at 80°C for 2 min and at 90°C for 6 min decreased 2.13 and 18.75%, respectively. There was significant effect (P<0.01) of temperature, time and preservative individually on the OA score whereas interaction effects were found to be non-significant.

Immediately after thermal and thermochemical treatment microbial load viz. total plate count, yeast and mold counts and coliform count in RTS beverages were determined and it was found to be decreased with increase in process temperature and time. As the data presented in Table 3, all treated samples were found to be safe for consumption in accordance with the guidelines of the Food Safety Standards Authority of India. Still, thermo-chemically treated beverage samples showed no detectable microbial load except for 80°C at 2 and 4 min. The coliforms were not detected in any sample including control.

Hence, preservation process has a vital role in maintaining original quality of the food products in addition to extending its shelf life. For the preservation of RTS green tomato beverage numerical optimized process parameters, 80°C temperature for 2 min without addition of preservative was obtained statistically with a desirability value of 0.946. It had optimum OA along with other nutritional factors. Therefore, standardized formulation and optimized preservation process for production of RTS green tomato beverage is 14% GTJ content, 12°B TSS and 0.2% acidity with the addition of 1% ginger juice, 1% lemon juice and 1% black salt, followed by hot filling at 85°C, crown corked and thermal treatment at 80°C for 2 min. Finally, the prepared RTS beverage was analyzed for its proximate and chemical composition and obtained results are presented in Table 4.

In conclusion, formulated RTS green tomato beverage is suitable for all age group as a healthy fruit based beverage having advantage of anticarcinogenic and antibiotic effects of green tomato along with its cholesterol reduction property and hemolytic activity against red blood cells. Further research needs to be carried out to develop different flavored beverages from the green tomato to replace the unhealthy sugar and water based carbonated beverages.

AUTHORS' CONTRIBUTION

Conceptualization of research (SHA); Designing of the experiments (SHA and AHP); Contribution

green tomato beverage.

Characteristics	Mean value±SD (n=3)
Proximate composition	
Moisture (%)	88.05±0.03
Protein (%)	0.43±0.01
Fat (%)	0.41±0.02
Fiber (%)	1.13±0.05
Ash (%)	0.95±0.03
Carbohydrate (%)	9.02±0.02
Chemical composition	
TSS (°B)	11.99±0.06
Acidity (% citric acid)	0.20±0.01
Vitamin C (mg/100 mL)	2.10±0.21
Antioxidant activity (µmol Fe+2/mL)	4.57±0.01
Phenolics content (mgGAE/100 mL)	12.48±0.12
Tomatine (mg/100 mL)	10.40±0.07

All above values are mean value of three replications

of experimental materials (ASH, BBP and HGB); Execution of lab experiments and data collection (AHP); Analysis of data and interpretation (BBP); Preparation of the manuscript (BBP).

DECLARATION

The authors declare that they have no conflict of interest to this work.

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