Changes in total phenolic content and color of bottle gourd (*Lagenaria siceraria*) juice upon conventional and ohmic blanching

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Abstract Plant phenolics exist in a complex matrix and require a high separation capacity for analysis. Bottle gourd (BG) was blanched using conventional thermal and alternate thermal ohmic heating methods using different combinations of temperature (60–90°C) and time (1–5 min), and the effect of these methods on total phenolic content (TPC) and color was examined. Complete characterization on the basis of the phenolic profile of unblanched, ohmically blanched, and conventionally blanched samples of BG was studied. Gas chromatography-mass spectrometry and Liquid chromatography-mass spectrometry were used to detect volatile and nonvolatile phenolic compounds, respectively. The color profiles of the unblanched and conventionally and ohmically blanched samples were studied via HunterLab colorimeter using L*, a*, and b* coordinates. The highest increase in TPC was observed at 80°C, 4 min and 90°C, 5 min in the case of ohmic and conventional blanching, respectively. The conventionally and ohmically blanched samples both produced desirable green-color retention in comparison to the unblanched samples; however, highest retention was observed in the ohmically blanched samples. The ohmically blanched samples exhibited the maximum extraction of phenolic compounds and better color of BG juice in comparison to the other samples.

Keywords: bottle gourd juice, total phenolic content, color, blanching

Introduction

Bottle gourd (BG, *Lagenaria siceraria*), commonly known as lauki or dhoo di in India, is a good source of many essential nutrients. Fresh and tender fruits of BG are used as vegetables and for juice extraction. BG is well-known for its health benefits and prevention of many chronic diseases. The health benefits ascribed to BG are mainly related to the presence of bioactive components. BG contains natural antioxidants such as ascorbic acid, flavonoids, and phenolic compounds and can contribute significantly in scavenging free radicals if taken regularly in the diet (1).

Apart from containing beneficial bioactive components, BG is considered a good source of plant phenolics. BG phenolics are susceptible to degradative reactions as soon as juicing is done. Enzymatic conversion of anthocyanin occurs via the conversion of different-colored compounds into brown compounds during BG juice extraction. The brown pigment superposed on natural colors renders the BG juice (BGJ) unappealing. Oxidative reactions and enzymatic degradation of phenolics and conversion into compounds that are less stable are mainly responsible for lesser yield of phenolics (2).

Blanching is a unit operation performed prior to freezing, canning,

juicing, or drying. It involves heating of fruits or vegetables for the purpose of deactivating the enzymes; modifying the texture; preserving the color, flavor, and nutritional value; and removing trapped air (3). Apart from enzymatic deactivation, blanching has beneficial effects on phenolics. Certain colored phenolics, e.g., chlorophyll, are present in deeper layers of cells surrounded by gases that obscure the real color by reflecting light. Transmission of heat via direct or indirect methods removes these surrounding gases and facilitates phenolic extraction from deeper layers of cells. Temperature and time play a significant role in the extraction of phenolic components from plant material. Song *et al.* (4) determined that compared with other temperature–time combinations, blanching of vegetable soybeans at a high temperature and for a short period of time (100°C, 10 min) more effectively prevents loss of greenness and reduces the leaching water-soluble components. In contrast, longer blanching times cause more color and nutritive-value losses. Decreasing the temperature of processing, especially below the boiling temperature, preserves 80–100% of phenolic content in some vegetables (5). Moreover, the type of blanching also affects the retention and increase or decrease in TPC. Mainly, conventional blanching and ohmic blanching are used at the industrial scale.

Conventional blanching involves immersing or deluging food into food-grade water or food-grade steam. In both cases, food is directly exposed to a typical temperature range. This technique is limited to low heat penetration into the center of vegetables and may also result in excessive softening and uneven blanching (6). Size reduction may reduce the problem of uneven blanching due to largely exposed surfaces and increase the effectiveness of the process (7).

Ohmic heating is an alternate heat-treatment technique that uses electrical energy for food processing. It involves passing alternate electrical current through food to generate internal heat as a result of electrical resistance. This technique has an advantage of fast, uniform heating owing to the generation of heat within the food material. Many studies have compared ohmic heating with conventional heating methods and have suggested that ohmic heating results in a better color and yield as well as imparts nutritional characteristics of many foods (8-10). Moreover, ohmic blanching requires lesser energy consumption, in terms of the heat required to heat the water to reach the desired temperature i.e., 0.5 kg of water/kg of the food product, as compared to conventional blanching that requires almost double the amount of water 2 kg of water/kg of food material hence, the energy required to heat that water also doubles in conventional blanching. Although blanching may prevent discoloration of the product and result in better extraction of phenolics, the process of blanching is very critical and needs to be performed with a proper temperature–time combination. Under blanching may accelerate the enzymatic activity of the product and result in faster degradation of color and phenolic compounds, whereas over blanching may cause excessive softening of product and thereby make juice extraction a difficult process as well as deteriorate its phenolic content. Therefore, this study was conducted to study the effect of conventional and ohmic blanching at varying temperature–time combinations on the TPC and color of BGJ.

Material and Methods

Reagents The Folin–Ciocalteu reagent (FCR), gallic acid, chloroform, hexane, ethyl acetate, ethanol, methanol, formic acid, and petroleum ether was purchased from Sigma-Aldrich (St. Louis, MI, USA). Silica gel and magnesium sulfate was purchased from Merck & Co. (Kenilworth, NJ, USA). Ultrapure deionised water (HP143BY; Cascade LS water, Thermo Fisher Scientific, Waltham, MA, USA) was used in all the experiments. N, O-bis (Trimethylsilyl) trifluoroacetamide with trimethylchlorosilane was purchased from Loba Chemicals (Mumbai, India). Absorbent tissues with 1-mm-thickness were purchased from Loba chemicals.

The entire glass assembly of the ohmic heater was fabricated at the Gupta Scientific Laboratory (Ambala, India). Other materials and chemicals used in the investigation were polyethylene terephthalate (PET) bottles (1 L and 500 mL), plastic container (500 mL), distilled water, hydrochloric acid (HCl), sodium hydroxide (NaOH); pipette tips

Fig. 1. Blanching vessel of the ohmic heater assembly
Labels

Diameter of beaker=10 cm

Height of glass assembly=9 cm

Distance between electrodes=7 cm

were available in the laboratory of the Department of Food Engineering and Technology, SLIET, Longowal.

Raw materials BG seeds (*Lagenaria siceraria* Var. kasha ganga), were purchased from Punjab Agriculture University (Ludhiana, India) and field-grown in SLIET, Longowal, (Punjab, India). Tender and good-quality vegetables free from any damage were used in this study.

Sample preparation The BGs were washed under running tap water and wiped. The outer skin of the BG was peeled using a handheld peeler before chopping. The BG were cut into cubes of uniform size ($2 \times 2 \times 2 \text{ cm}^3$) to ensure uniform blanching.

Blanching Blanching was performed according to the method followed by Bhat *et al.* (11); i.e., two-step blanching was performed: conventional and ohmic.

Conventional hot-water blanching was performed by simmering the BG cubes ($2 \times 2 \times 2 \text{ cm}^3$) into the water bath (NSW-133-BSS-2; Narang Scientific Works Pvt. Ltd., New Delhi, India) at different temperatures (60, 70, 80, and 90°C) for varying time periods (1, 2, 3, 4, and 5 min). The sample-to-water ratio was 1:5. The temperature of the water bath was controlled via an automatic temperature controller.

Ohmic blanching was performed using a laboratory-scale ohmic heater connected with a variable transformer to provide a power supply of 220 V and a fixed frequency of 50 Hz. The test cell used for blanching was made of borosil glass and had a capacity of 1,000 mL and total diameter of 10 cm. The height of the glass vessel was 9 cm, and the electrode-to-electrode distance was 7 cm. The electrode was fixed to the interior of the blanching vessel at a distance of 1.5 cm from the vessel surface. The electrodes were 6 cm long, 2 cm wide, and 0.1 thick and were made of stainless steel. Figure 1 shows the sketch diagram of the ohmic cell used for the blanching of the BG cubes.

Blanching was performed in a test cell using a sample-to-water ratio of 1:5 at varying temperatures (60, 70, 80, and 90°C) for 1, 2, 3, 4 and 5 min at a fixed frequency of 50 Hz and voltage of 220 V. The temperature was maintained using thermocouples.

Juice extraction The blanched BG cubes were wiped into an absorbent tissue before juice extraction. BGJ was extracted using a laboratory-scale juicer (Philips, Amsterdam, Netherlands). The extracted juice was filtered twice to remove any suspended solids, and clarified juice was used for the analysis.

Selection of solvent for the extraction of phenolics using the thin-layer chromatographic (TLC) technique Three samples of BGJ, i.e., control (unblanched), conventionally blanched (90°C, 5 min), and ohmically blanched (80°C, 4 min) were extracted using different solvents: petroleum ether, chloroform, and ethanol. The extracts were centrifuged (Remi PR -24; Remi Lab World, Mumbai, India), concentrated in a rotary evaporator (R-300; Remi lab world, New Delhi, India), and subjected to TLC profiling. Conventional TLC profiling via the one-dimensional ascending method using silica gel G (Merck Art 1/17631) was performed. The prepared extracts from petroleum ether, chloroform, and ethanol were spotted on a precoated TLC plate using a glass capillary. Approximately 1 µL was spotted at a distance of 1 cm and allowed to run for 20 min in the TLC chamber using different solvent systems (mobile phase). Different solvent systems, hexane, and ethyl acetate in a ratio of 9:1, 8:2, 7:3, 6:4, 5:5, 9:1, 8:2, 7:3, 6:4, and 5:5 as well as methanol, chloroform, and formic acid in a ratio of 7:2:1, 6:3:1, and 5:4:1 were used in the mobile phase.

The developed TLC was air-dried and shifted to an iodine chamber to detect the bands on the TLC plate. All detections were observed directly after drying. The movements of solute phase in sample containing active phenols was dragged by solvent phase in TLC plate and is expressed in terms of R_f .

$$R_f = \frac{\text{Distance travelled by solute}}{\text{Distance travelled by solvent front}} \quad (1)$$

Total phenolic content TPC was determined via FCR as per method followed by Bhat and Sharma (12) with certain modifications. Deionized water (DIH₂O) was used for preparing the gallic acid stock solution (1 mg/mL). Working standard concentrations of 0, 10, 25, 50, 100, and 250 µL/mL were prepared. Moreover, 50-µL samples of unblanched, ohmically blanched, and conventionally blanched juices were transferred into three test tubes (10 mL), followed by the addition of 450 µL of DIH₂O and 20 µL of FCR. The samples were mixed properly in a magnetic shaker, and 50 µL of 20% Na₂CO₃ and 450-µL DIH₂O were added. The samples were allowed to stand for 1 h at room temperature. Absorbance was measured at 725 nm using a spectrometer (Cole-Parmer, Mumbai, India). TPC was measured against the gallic acid calibration standard, and the results were expressed in terms of gallic acid equivalents.

Color measurement In the case of BGJ, color measurement was performed as per the method followed by Bhat *et al.* (11). The degree of browning was represented by hunter color parameters L*, a*, and b* using a Konika Minolta chromometer (model no. 410; Minolta, Osaka, Japan). The value of lightness L* varied between 0–100, with the highest one representing the highest lightness. The value of a* and b* ranged from –60 to +60 for redness and yellowness, respectively, and the negative values showed the respective greenness and blueness. The hunter color values L*, a*, and b* of BGJ in both the unblanched and blanched samples were measured using the illuminant D65/01 observer. The instrument was calibrated against a white standard. Total color difference (E) was calculated as

$$\Delta E = \sqrt{(\Delta L^*)^2 + (\Delta a^*)^2 + (\Delta b^*)^2} \quad (2)$$

In addition, the browning index (BI) of both the unblanched and blanched samples was estimated according to following equation:

$$BI = [100(x - 0.31)]/0.17, \quad (3)$$

where

$$x = (a^* + 1.75L^*)/(5.645L^* + a^* - 3.012b^*) \quad (4)$$

Gas chromatography-mass spectrometry (GC-MS) analysis Sample preparation for the Gas chromatography-mass spectrometry (GC-MS) analysis was performed as per the method of Zhang and Zuo (13) with some modifications. The unblanched, ohmically blanched (OB), and conventionally blanched (CB) juice samples were filtered through a Whatman filter paper (125 mm cat no. 1002 125). Moreover, 10 mL of each sample was extracted thrice by vortexing for 1 min in a 1.5 mL tube using a methanol: chloroform: formic acid ratio of 7:2:1. The mixture was then centrifuged for 10 min, and the top layer was removed and pooled in another tube. The extracts were passed through anhydrous MgSO₄ packed in a Pasteur pipet and evaporated to dryness under nitrogen. The extracts were derivatized by the addition of 50 µL of the derivatization reagent N, O-bis-trimethylsilyl trifluoroacetamide (BSTFA)+trimethylchlorosilane (TMCS) and heated at 70°C; 1 µL of each sample was injected for the GC-MS analysis.

The analysis was performed on a thermo GC (TRACE-1300; Thermo Fisher Scientific), which is an auto sampler interfaced to a mass spectrometer, thermo MS [Model TSQ 8000 (triple quadrupole)], operated in the electron impact ionization mode (EI). GC-MS was employed with a column: thermo TraceGold (TG) 5MS (30 mm×0.25 mm×0.25 µm, 5% phenyl methyl poly siloxane) operating in the electron impact mode at 70 eV; helium (99.9%) was used as the carrier gas at a constant flow of 1 mL/min. An injection volume of 1 µL of BGJ was employed in the split less mode at 250°C using an initial temperature of 60°C (2 min), which was then ramped to 240°C at 6°C/min (14).

Identification of compounds Identification of compounds from the fingerprints observed in the mass spectrum was performed using the database of the National Institute Standard and Technology

(NIST) library's mass spectral database. The spectrum of the unknown component was compared with that of the known components stored in the NIST library.

Liquid chromatography-Mass spectrometry (LC-MS) analysis

Sample preparation: The BGJ of the unblanched, ohmically blanched, and conventionally blanched samples was extracted (Philips); 100 mL of each sample was weighed and introduced into a 250-mL Erlenmeyer flask. Ultrasonically assisted extraction of phenolics was performed for 10 min at a 20-kHz frequency and at a temperature of 25°C. The samples were centrifuged at $1,957\times g$ for 10 min and ultrasonicated again for 10 min; 1 mL of sample was shifted to an Eppendorf tube along with 1 mL of the mobile phase. The sample was then centrifuged again, and 5 μ L was used for injection in LC-MS.

LC-MS analysis: LC-MS analysis was performed in SYNAPT G₂-Q-ToF-module (Medwow global, Manchester, UK). This Instrument was a hybrid quadrupole equipped with an electrospray ionization (ESI) probe with a mass range of 4,000 amu. A hexapole collision cell was used between the two mass analyzers to induce fragmentation for analyzing the structural investigations while using the above-mentioned instrument in the MS/MS mode. The analyte was separated on a symmetry C18 column (100 mm $\times$ 2.1 mm) and a guard column (20 mm $\times$ 2.1 mm) of the same stationary phase. In addition, 5 μ L was injected in a column under a flow rate of 0.2 μ L/min. The mobile phase comprised A: water containing 1% formic acid (v/v) and B: methanol with 1% formic acid (v/v). MS was performed with the positive-mode polarity of Electrospray ionization (ESI). Compound identification was done with effective usage of online libraries using molecular mass and structural formula of the compounds.

Statistical analysis Statistical analysis was conducted using the SPSS software (version 16.0; SPSS Inc., Chicago, IL, USA). The experiments were performed in triplicates, and data were expressed as the mean $\pm$ standard deviation (SD) of the triplicate experiments. Two-way analysis of variance (ANOVA) was used to analyze the combined effect of different parameters on the samples. *P*-values less than 0.01 were considered to be statistically significant. Moreover, Duncan's post hoc multiple range test was used to examine the differences between the individual groups. *P*-values less than 0.05 were considered to be statistically significant.

Results and Discussion

Thin-layer chromatographic studies Various solvent systems with different polarities were studied using thin-layer chromatography to determine the most effective solvent system for phenolic separation. Petroleum ether, chloroform, and ethanolic extract of BGJ revealed the presence of many components with different retention factors. Among the different solvent systems used for detection, in terms of

Fig. 2. Effect of different temperature–time combinations on the total phenolic content in bottle gourd juice.

Labels

O=ohmic

C=conventional

their ability to separate components, a methanol: chloroform: formic acid ratio of 7:3:1 proved to be the most effective, producing multiple clearly visible spot separations. Other solvent systems and combined ratios showed no prominent clear-spot separation. The TLC studies revealed that petroleum ether extract showed maximum spot separation and was therefore selected for further analysis.

Effect of blanching on the phenolic compounds of BGJ

Plant phenolic compounds such as flavonoids, phenolic acids, and tannins play an important role in preventing various chronic disorders such as inflammation, cancer, and arteriosclerosis. These are one of the most important groups of natural antioxidants because of their diversity and extensive distribution. The retention, degradation, and extraction of phenolics depend on the type of phenolics, method of processing, and conditions of processing. It was observed that the extraction of phenolic compounds increased after blanching. The total phenolic content (TPC) of unblanched BGJ was 640 mg/100 mL, and it showed a significant ($p<0.01$) increase up to 644 and 697 mg/100 mL during conventional and ohmic blanching, respectively (Fig. 2). This may be due to the greater extraction yield of phenolics, which is linked to the increase of skin permeability caused by heat treatment. However, increasing the blanching temperature beyond a certain limit (Fig. 2) in both conventional and ohmic blanching led to a decrease in TPC. It is generally considered that applying a high temperature for a long holding time causes damage to functional components. The results showed that an increase in the temperature–time combinations up to 90°C, 5 min and 80°C, 4 min in the case of conventional and ohmic blanching, respectively, caused a significant increase in TPC. Temperature–time combinations beyond a certain level did not follow the similar trend that highlighted the role of heat in degradation in cellular materials first by increasing the phenolic

Table 1. Total color difference and browning index values during ohmic and conventional blanching

Time (min)	Temp. (°C)	Ohmic blanching		Conventional blanching	
		ΔE	BI	ΔE	BI
1	60	27.00	174.64	28.50	178.89
2	60	27.36	174.64	28.65	173.30
3	60	27.56	174.64	28.67	173.22
4	60	27.62	174.64	30.50	163.91
5	60	27.37	174.64	32.13	162.02
1	70	28.24	174.64	33.02	163.96
2	70	29.74	162.88	45.57	159.20
3	70	30.19	157.00	47.70	153.63
4	70	31.36	157.00	52.61	141.55
5	70	34.31	156.00	57.75	128.20
1	80	62.75	111.00	64.00	115.20
2	80	64.69	101.00	64.02	115.00
3	80	70.26	92.00	68.40	104.83
4	80	74.03	86.41	72.52	89.73
5	80	74.03	86.40	72.61	89.11
1	90	74.03	86.40	72.51	89.00
2	90	74.05	86.49	75.16	88.78
3	90	74.04	86.41	75.71	88.04
4	90	74.04	86.41	75.99	88.01

mass transfer and then degradation of these extracted phenolics at a high temperature for longer holding. Moreover, ohmic blanching showed the highest increase (8.9%) in TPC up to 80°C, 4 min followed by conventional blanching (Fig. 2). TPC losses due to the increased temperature–time may be attributed to the unstable nature of phenolics that are unstable at high temperatures. Moreover, extractability is highly influenced by the type of blanching performed to facilitate and transfer phenolics from the cell's interior to the external environment. In addition, free phenolics that are present at outer cell surfaces are leached out at faster rates than the bound phenolics are intact to cells. The high extractability and retention of TPC during ohmic heating may be attributed to the additional non thermal effects at frequencies greater than 40 Hz. Moreover, ohmic blanching requires lesser temperature and time than conventional thermal blanching, which maybe another reason for lesser degradation of phenolics. These results are supported by Jaiswal *et al.* (15) in the case of York cabbage.

Effect of blanching on the color of BGJ An acceptable green color was retained after blanching of BG at 80°C, 4 min and 90°C, 5 min in the case of both ohmic and conventional blanching, respectively. The total color difference was estimated on the basis of L^* , a^* , and b^* values (Table 1). The blanched BG cubes showed higher E than the unblanched samples because of increase in L^* and decrease in a^* . The increased E indicates an increase in the lightness of samples with the temperature possibly due to the inactivation of enzymes that are mainly responsible for the formation of dark-colored compounds. The formation of brown compounds decreased because of an increase in the temperature–time combinations and was estimated

by a decrease in the BI. The ohmically blanched samples showed the highest E value at 80°C, 4 min and beyond, whereas the highest E value was observed at 90°C, 5 min for the conventionally blanched samples. Beyond 80°C, 4 min, the ohmically blanched samples showed negligible changes in E. The darkest green color was therefore retained at these time–temperature combinations. The control sample showed the lowest negative a^* value; therefore, the lightest green and darkest brown colors were observed in it. The increased L^* value and decreased a^* value during both conventional and ohmic blanching showed a decrease in browning. This can be verified from the decrease in BI, which was observed to decrease from 174.64 to 86.41 and from 178 to 88.00 with an increase in temperature–time combination in the case of ohmic and conventional blanched samples, respectively. The minimum BI of 86.41 was observed in the case of ohmically blanched samples at 80°C, 4 min. The retention of green color after blanching may be due to the release of chlorophyll-containing compounds. These compounds are trapped within the cellular network and surrounded by gases that partially obscure the hue by reflecting light. Mild heat treatment helps these surrounding gases escape into air or the surrounding water, thereby intensifying the green color. However, heat treatment beyond the boiling point degrades and starts releasing acids from plant cells that are responsible for the degradation of colored compounds. Similar results were obtained by Qudsieh *et al.* (16) in the case of sugarcane juice. Enzymatic and nonenzymatic browning is another main cause of brown-color formation in vegetable products. Compared with ohmic blanching, the BI was found to reduce at a faster rate (Table 1) and at lower temperature–time combinations in conventional blanching. However, it was observed that the conventionally blanched samples turned brown again after a short interval (30 min) when blanched at temperature–time combinations lower than 90°C, 5 min. Compared with the conventionally blanched samples, the ohmically blanched ones showed color retention for longer time periods even when blanched at 80°C, 4 min. This could be attributed to the enzymes and their heat-sensitive nature. With an increase in temperature, the enzyme activity decreased initially and required a particular temperature–time combination for complete inactivation. Inadequate blanching tended to slow the enzyme activity for a certain period rather than complete inactivation, which may have resulted in browning after a short interval of time. As the blanching proceeded, the enzymes deactivated, and complete inactivation was observed at 80°C, 4 min and 90°C, 5 min in ohmic and conventional blanching, respectively; hence, the maximum green-color retention was observed with this temperature–time combination (8). The reducing sugars present in BGJ are not in adequate amounts to initiate the Millard reaction at low temperatures. Therefore, non enzymatic browning contributed less to the brown-color formation in BGJ. The principle reason for the brown-color formation in BGJ could be the breakdown of natural phenolics by enzymes to produce melanin and benzoquinones. A high temperature leads to an increase in the internal energy of molecules and enzymes

Fig. 3. Total ion chromatograms of the phenolics in raw (A), ohmically blanched (B), and conventionally blanched (C) bottle gourd juice samples

and may consequently break the weak bonds that determine the three-dimensional structures of active proteins in enzymes. This breakdown may lead to thermal denaturation of protein and thereby deactivate the enzymes. Thus, the decrease in enzyme activity with increasing temperature allowed better retention of green color. The enzymes involved in the browning reaction were completely deactivated at 90°C, 5 min and 80°C, 4 min in the case of conventional and ohmic blanching, respectively (8).

Gas chromatography-Mass spectroscopy (GC-MS) analysis of phenolic compounds The total ion chromatograms (TICs) of the volatile components of the unblanched, ohmically blanched, and conventional blanched samples of BGJ were obtained using GC-MS. (Fig. 3A-3C). A large number of identified bioactive compounds were aromatic hydrocarbons, which contribute to color and flavor. The

main compounds identified were: rhodopin, nonanal, salicylic acid, 2,4 decadienal, decanal, 2,5-di-tet-butyl 1,4 benzoquinones, phytoalaxin, 2-pentanone, 9-octadecenal, myricetin, 1-monolinoleoyl glycerol trimethyl silyl, ethyl iso-allocholate, gallic acid, quercetin, quercetin-3-O-rutinoside, o-coumaric acid, and hydroxytyrosol. Among the 17 identified compounds, majority of the compounds were aromatic hydrocarbons with phenolic groups. Some phenolic acids such as salicylic acid and phenolic derivatives such as 2,5-di-tet-butyl 1,4 benzoquinones were also identified. Nonanal and gallic acid are colorless compounds with strong phenolic and antioxidant properties. Gallic acid is a widely studied compound and possesses anticancer and antimicrobial properties (17). Rhodopin contributes to the slight yellow color of BGJ; 2,4 decadienal is a strong flavor enhancer, whereas 1,4 benzoquinones possess selective and potent antioxidant properties and act as the precursor of hydroquinone,

Table 2. Major phenolic compounds identified in bottle gourd juice via GC-MS and LC-MS analysis

Compound	Retention time (min)	Peak area (%)	Identified ions (m/z)
Rhodopin	4.38	2.1	74,112
Nonanal	6.08	6.91	100,75,10
Salicylic acid	8.25	4.25	158
2,4 Decadienal	9.03	3.00	152,125,100
Decenal	9.62	5.96	121,100,70
2,5-Di-tet-butyl 1,4 benzoquinone	11.02	2.68	220,177,135,91
Phytoalaxin	12.26	0.38	71,94,121
2-Pentanone	13.42	2.15	95,135,181,20.1
9-Octadecenal	15.74	0.88	234.3, 208.1
Myricetin	17.69	0.32	98, 72, 120
1-Monolinoleoyl glycerol tri methyl siliyl	19.48	0.34	281,253, 207,135
Ethyl iso-allocholate	20.12	0.11	405, 327,281, 253,
Gallic acid	22.27	3.14	169
Quercetin	24.27	5.56	170
Quercetin-3-O-rutinoside	27.30	4.60	178,179
o-Coumaric acid	29.14	0.12	163
Hydroxytyrosol	31.10	0.11	153,192

Peak	m/z	RT	Ion	m/z fragments	Tentative identification	Juice type
1	30.83	32.2	[M+H] ⁺	37.51; 41.97; 30.20	Carboxy methyl glucan	C; OB; CB ¹⁾
2	44.31	47.1	[M+H] ⁺	44	N,N-Dimethyimethyl amine	C; OB; CB
3	52.11	48	[M+H] ⁺	52.37; 50.18	Ethanol	C; OB; CB
4	58.37	60	[M+H] ⁺	50.78	Glyoxal	C; OB; CB
5	61.02	62	[M+H] ⁺	64.43	N-Hydroxyl N-methyl	C; OB; CB
6	65.57	70	[M+H] ⁺	65.53	1,3-chloro pentadiene	C; OB; CB
7	79.95	72	[M+H] ⁺	76.90	Theaflavin-3-gallate	C; OB; CB
8	82.04	80	[M+H] ⁺	81.96; 82.66	Furan, 3-Methyl	C; OB; CB
9	87.63	90	[M+H] ⁺	87.35	Gallic acid	C; OB; CB

¹⁾C, control; OB, ohmically blanched; CB, conventionally blanched

which is an antioxidant. Quercetin is a phenolic compound that mainly contributes to flavor. Phytoalaxin belongs to a group of phenolics that exhibit free-radical scavenging activity and are mainly utilized by the BG plant for defense against pathogens. The increase in TPC depends on the type of processing and conditions. Phenolics are not uniformly distributed within the tissue of plants. Certain phenolics are deposited in the outer cell wall in vacuoles and may be easily extracted with mild heat treatment. However, phenolics stored in the epidermal and sub-epidermal layers of tissues require appropriate processing treatment to open the food matrix; this allows the release of bound phenolics from deeper layers without much damage to the structural profile of phenols. Processing technologies such as conventional and ohmic blanching may assist in releasing bound phenolics by breaking down the cellular constituents of their cell wall. However, beyond certain temperature ranges, as observed in conventional and ohmic blanching (90°C, 5 min and 80°C, 4 min, respectively), TPC decreased. Moreover, browning in the control sample due to oxidation decreased the TPC content in BG. The highest browning and lowest phenolic content (640±0.5 mg/100 mL) was observed in the control sample of BG. The reason for the decrease in TPC with increased browning may be the degradation caused by the isomerization and oxidation of the basic structure of

phenolics and their consequent conversion into less useful compounds.

All the three samples showed similar ranges of compounds; the maximum peak intensity of each compound observed in the ohmically blanched samples was higher than that observed in the unblanched and conventionally blanched samples possibly because of the enzymatic conversion of phenolic compounds in the unblanched samples and heat degradation in the conventionally blanched BG samples. This was in good agreement with the results obtained from the estimation of TPC. The highest TPC (697±1 mg/100 mL) was observed during ohmic blanching at 80°C, 4 min, followed by conventional blanching (644±1.5 mg/100 mL); the lowest TPC was observed in the control sample (640±0.5 mg/100 mL).

Liquid chromatography-Mass spectroscopy (LC-MS) analysis of phenolic compounds Liquid chromatography-Mass spectroscopy (LC-MS) provides information on molecular mass without the need for isolation of trace components. Moreover, structural information enables identification of the detected compound. Nonselective LC-MS of BGJ for the detection of nonvolatile compounds was performed, and the results showed several fragmented and signature peaks.

Among the many peaks observed, nine compounds of our interest with phenolic properties were identified (Table 2). All the samples showed similar compounds with different relative occurrence, which represents the concentration of a particular fragmented ion. The ohmically blanched samples showed the highest occurrence of phenolic compounds, followed by the conventionally balanced samples; the lowest occurrence of phenolic compounds was observed in the control sample. The significant increase in the amount of phenolics during ohmic heating can be attributed to the fact that electrical energy can rupture the tissues and therefore release more phenolics. Ohmic heating has a potential to extract the maximum phenolic compounds with minimum damage to their structural integrity because it involves rapid heating and minimal heating of the product surroundings.

The highest increase was observed at 90°C, 5 min and 80°C, 4 min in conventional and ohmic blanching, respectively. The major phenolic compounds identified during the GC-MS analysis were decanal, salicylic acid, nonanal, and benzoquinones. An LC-MS revealed that the amount of gallic acid and theaflavin 3-gallate in the ohmically blanched samples of BGJ is higher than that in the conventionally blanched and unblanched samples. The BI values showed a continuous decrease with an increase in temperature–time combination upto 80°C, 4 min and 90°C, 5 min in the case of ohmic and conventional blanching samples, respectively. Beyond 80°C, 4 min, negligible change was observed in the BI of the ohmically blanched samples. The highest increase in TPC was also observed at 80°C, 4 min in the ohmically blanched samples. Moreover, compared with the unblanched and conventionally blanched samples, the color of BGJ in the ohmic blanched samples was more desirable.

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