PLANT SCIENCE TODAY
ISSN 2348-1900 (online)
Vol 11(sp3): 12-17
https://doi.org/10.14719/pst.5430

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RESEARCH ARTICLE

Biochemical profiling and byproduct utilization of *Garcinia* aril

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OPEN ACCESS

ARTICLE HISTORY
Received: 31 August 2024
Accepted: 18 November 2024
Available online
Version 1.0 : 25 December 2024
Version 2.0 : 14 February 2025

Check for updates

Additional information
Peer review: Publisher thanks Sectional Editor and the other anonymous reviewers for their contribution to the peer review of this work.

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CITE THIS ARTICLE
Lekshmi PRG, Aparna GS, Anith KN. Biochemical profiling and byproduct utilization of *Garcinia* aril . Plant Science Today. 2024; 11(sp3): 12-17. https://doi.org/10.14719/pst.5430

Abstract

Garcinia gummi-gutta, commonly known as Malabar Tamarind, is a fruit crop of significant economic value, extensively grown in the southern regions of the Western Ghats. The economically valuable part is the fruit peel or rind, which is used as a seasoning in its dried form, imparting a distinct flavour and taste to food preparations. During processing of the rind, the edible aril, a whitish pulp attached to the seed, is discarded. The aril of *G. gummi-gutta* fruits has a unique flavour with a sweet-acidic blend. Biochemical profiling of the aril is essential for developing food products with enhanced nutritional benefits. The present study reports the quantification of its biochemical components. Identification and quantification of various sugars and acids were performed using Liquid Chromatography Tandem Mass Spectrometry. Fructose (47.44 mg/g) emerged as the predominant sugar, while Hydroxy Citric Acid (522.99 mg/g) was identified as the principal organic acid. Biochemical profiling of the aril revealed its potential for functional food development. The aril contained significant amounts of Hydroxy Citric Acid (2.64%), known for anti-obesity properties, along with calcium (756.67 ppm), potassium 649.52 ppm, phenols 382.25 mg GAE/g, flavonoids 6.58 mg QE/g, and exhibited 87.32% antioxidant activity. Ready-to-serve blended fruit beverages were developed using the pulp of *G. gummi-gutta* aril combined with pomegranate, watermelon or dragon fruit juice. Sensory analysis indicates higher consumer acceptance and enhanced nutritional qualities. The utilisation of *G. gummi-gutta* byproducts can contribute to food product diversification, providing innovative options for novel products with enhanced taste and health benefits.

Keywords

bioactive compounds; byproduct utilisation; Garcinia gummi-gutta; organic acid profiling; ready-to-serve beverages; sugar profiling

Introduction

The genus *Garcinia*, commonly found in Asian and African tropical forests, is one of the oldest domesticated plant groups, valued for its importance in medicine and food (1). It belongs to the family Clusiaceae, which comprises 43 species and the Western Ghat mountain region of the Indian subcontinent serves as a biodiversity hotspot for *Garcinia*. *Garcinia gummi-gutta* (L.) Roxb. after formerly known as *G. cambogia* (Gaertn.), is an indigenous, semi-domesticated crop renowned for its nutraceutical and anti-obesity properties. Known Malabar tamarind or Brindle berry, it has been used in traditional medicine to treat respiratory diseases (2).

Phenolic acids, flavonoids and bioflavonoids are responsible for pharmacological properties in *Garcinia* (3). The leaves of *G. gummi-gutta* are

particularly rich in ferulic acid, protocatechuic acid and caffeic acid (4). The fruit is an abundant source of Hydroxy Citric Acid, xanthones and flavonoids which have nutraceutical properties. The dried fruit rind, known for its bioactive compounds, is used as a seasoning that imparts a distinct flavour and taste to traditional cuisine. *G. gummi-gutta* rind was found rich in luteolin, caffeoylquinic acid, coumaroylquinic acid, kaempferol glycoside and apigenin as the phenolic acids (2). The most important phytochemicals present in *G. gummi-gutta* fruits were reported as organic acids, Hydroxy Citric Acid as the major one, xanthones (oxyguttiferone I, oxyguttiferone) and benzophenones (guttiferone I, N, J, K and guttiferone M) (5). During the processing of fruit rind, the edible aril, the whitish pulp attached over the seed, is discarded. The edible white aril of *G. gummi-gutta* fruits has a unique flavour with a sweet-acidic blend. Biochemical profiling of the aril is essential for developing food products with functional benefits; however, reports on its nutritional characterization are limited. This study focuses on quantifying the biochemical components of the aril, including total soluble solids, total and reducing sugars, acidity, protein, crude fiber, fat, ascorbic acid, carotenoids, minerals, Hydroxy Citric Acid, total phenols, total flavonoids and antioxidant activity, providing valuable insights into its nutritional profile. Liquid Chromatography Tandem Mass Spectrometry (LC-MS/MS) estimation was used to conduct profiling of sugars and acids providing essential information for the formulation of different processed foods. Value-added products from *G. gummi-gutta* aril have not been developed, despite its immense potential due to its bioactive content and unique flavour. Therefore, an attempt was made to develop Ready-to-serve beverages by blending *Garcinia* aril with various fruit juices, and their sensory and nutritional qualities were analyzed.

Materials and Methods

Fully ripe and uniform sized fruits of *G. gummi-gutta* var. *Gummi-gutta,* including varieties released from Kerala Agricultural University - Haritham, Nithya and Amrutham, along with a local cultivar (from selected homesteads), were collected for the study. The fruits were globose, deeply grooved with 6 to 10 grooves, seeds ovoid surrounded by white pulpy or reddish aril (6). The ripe fruits were analysed for various fruit characteristics, including fresh fruit weight, rind weight, rind Percentage, seed weight, seed %, pulp (aril) weight and pulp percentage using 30 replicates.

Biochemical parameters

The biochemical composition of the aril (pulp) of ripe fruits, as well as the biochemical parameters of the developed Ready-to-serve beverages, was determined using standard procedures. A digital refractometer (ATAGO; 0 - 85 ^{0}Brix) was used to determine Total Soluble Solids (TSS). Total acidity was measured using a titrimetric method with 0.1 N NaOH and expressed in terms of citric acid equivalent (7). The Vitamin C (ascorbic acid) was quantified with titrimetric method using 2, 6 dichloro phenol Indophenol dye and expressed as mg 100 g^{-1} of pulp weight. Protein content (g 100 g^{-1}) was measured according to the standard method using Bradford's colorimetric method (8). Hydroxy Citric Acid (HCA) was quantified by measuring the absorbance value at 485 nm of metavandate complex formed and standard curve was plotted with freshly extracted calcium salt of HCA. Acids from the pulp (aril) were extracted with 0.05 N H_2SO_4 to a final volume of 200 mL. To 1 mL of this extract, 1 N NaOH and 0.4 mL of 2.5% sodium metavandate were added and the solution was left at 28-32 °C for 30 min to form a coloured HCA-metavandate complex (9). Carotenoids were estimated using the colourimetric method (10) and crude fibre was determined by gravimetric method after chemical digestion (8). Fat content was determined by gravimetric method following the Twisselman method using petroleum ether as solvent (11). Iron content was estimated using di-acid digestion and measured with Atomic Absorption Spectrophotometry (AAS with Graphite Furnace of Analytik Jena India Model: HSN/SAC 90279000) and Calcium content was quantified using a titration method with Ethylenedi-aminetetraacetic Acid (EDTA) and potassium content was measured using a flame photometric method. Total phenol content was quantified as Gallic Acid Equivalent (8) and the total flavonoids were assessed to Quercetin Equivalent using the colourimetric assay (12). Total antioxidant activity of the aril was analysed with DPPH scavenging assay (2, 2- diphenyl-1-picrylhydrazyl) and expressed in percentage (13).

Sugar profiling

Sugars were extracted from the oven dried *Garcinia* aril and 0.1 g dried pulp was diluted 20 times with mobile phase. The mobile phase comprised of Solvent A and Solvent B. Solvent A was acetonitrile and Water in an 80:20 ratio, while Solvent B was acetonitrile and water (30:70) with 0.1% ammonium hydroxide. A filtered 1 mL sample was collected and 2 µL of this sample was injected into LC-MS/MS system (Waters UPLC H class with TQD MS/MS) for profiling. The analysis began with an initial gradient of 100% solvent A for 1 min, transitioned to 88% solvent A and 12% solvent B at 8 min, shifted to 98% solvent A and 2% solvent B at 15 min and returned to initial conditions by 19 min for equilibration. The flow rate was 0.1 mL/min, using a 2.1 × 100 mm UPLC BEH-Amide column with 1.7 µm particles, maintained at 25 °C. Elution was monitored with a Photo Diode Array detector, directing effluent to the TQD-MS/MS system (14).

Organic acids profiling

The extraction procedure for organic acids was performed as previously (15). Dried *Garcinia* pulp (1 g) was diluted 20 times with mobile phase and 1 mL of the solution was filtered. From this, a 2 µL sample was injected into the LC-MS/MS system. Initially, the gradient was made up of solvent A: aqueous phase (100%) and solvent B: organic phase (0%) which maintained for 30 sec. The gradient was adjusted to 95% aqueous phase and 5% organic phase in the 5th min and it was maintained for 30 sec. At the 6th min, the system was brought back to the starting state and it was maintained for a min to equilibrate before the next injection. A constant flow rate of 0.1 mL/min was maintained. The UPLC BEH- Amide column of 1.7 µm particles (2.1 x 50 mm) protected by Vanguard column was used. The temperature was 25 °C and 4 µL sample was injected. The eluted acids were monitored by Photo Diode Array detector fitted with TQD-MS/MS (Waters)

system, which was optimized for organic acids analysis.

Formulation of Ready-To-Serve beverages

The pulp (aril) attached to the seeds of *G. gummi-gutta* was hand extracted and strained for the preparation of Ready-To-Serve (RTS) beverages, following the FSSAI (Food Safety and Standards Authority of India) specifications (Juice % ≤ 10, TSS ≤ 10 ^{0} Brix, Acidity ≥ 0.3%). The *Garcinia* pulp-only RTS beverage formulations were as follows: G1- 10% *Garcinia* pulp 10%, 10% sugar; G2 - 10% *Garcinia* pulp 10%, 15% sugar; G3 - 10% *Garcinia* pulp, 20% sugar. The 10% *Garcinia* pulp was blended with different fruit juices (pomegranate (GP), watermelon (GW) and dragon fruit (GD)) in varying proportions (10 to 20%) across 6 replications. These formulations were analyzed for sensory parameters and the best formulation from each fruit juice blend was subjected to biochemical analysis. The RTS formulations were evaluated for sensory attributes: colour, flavour, appearance, taste, texture and overall acceptability by a panel of 30 semi-trained members. Hedonic scores were given on a scale from 1 to 9, where: 9 - Like extremely, 8- Like very much, 7- Like moderately, 6- Like slightly, 5- Neither like nor dislike, 4- Dislike slightly, 3- Dislike moderately, 2- Dislike very much, 1- Dislike extremely (7). The mean score for each sensory attribute was statistically analysed. The best formulations from each RTS beverage blend (T_1 - *Garcinia* aril, T_2 - *Garcinia* aril + pomegranate juice, T_3- *Garcinia* aril + watermelon juice and T_4- *Garcinia* aril + dragon fruit pulp) were selected for biochemical parameters following the procedure as described above.

Statistical analysis

The fruit's physical parameters were analysed for 30 replicates and biochemical parameters were analysed for 15 replications. The data obtained were statistically analyzed with one-way analysis of variance and the significant difference was determined ($p < 0.05$). The Kruskall-Wallis chi-square value test was used to statistically analyse the sensory mean score.

Results and Discussion

The fresh fruit weight of ripe whole *G. gummi-gutta* fruits ranged from 99.76 g to 125.90 g and the varieties Haritham and Amrutham recorded the lowest fruit weight (Table 1). The variety Nithya had the highest fruit weight (125.90 g) and the local variety also recorded an average fruit weight of 117.90 g. The fruit rind, which is the economical part of *G. gummi-gutta* ranged from 68.40 g (Haritham) to 92.72 g in Nithya and the rind % was also the highest in Nithya (73.75%), followed by Amrutham, Haritham and the local cultivar. The seed weight was highest in Nithya (20.03 g) and the lowest was in the local cultivar (16.84 g). The seed% was highest in Amrutham (18.58%), followed by Haritham (17.31%), with the lowest value (14.27%) observed in the locally collected fruits. The highest average pulp/aril weight (20.38 g) was recorded for the local collection with 17.41% of total fruit weight. The lowest pulp % was observed for the varieties Nithya, Amrutham and Haritham, which indicated a higher rind % in improved varieties. This data revealed that *G. gummi-gutta* fruits collected locally, which are found majorly in homesteads of Kerala, are good source of aril/pulp that constitutes about 17.41% of the whole fruit weight. This edible pulp, even though peculiar flavour and sweet-acidic taste, is wasted during the processing of rind. Significant differences were observed in rind percentage, rind thickness, pulp percentage, seeds per fruit and seed percentage among the 2 morphotypes of Andamans (16). A wide variation in fruit yield, fruit rind thickness and dry rind recovery was reported in a *G. gummigutta* germplasm evaluation study conducted in Tamil Nadu (17).

The biochemical composition of *G. gummi-gutta* fruit aril was analysed and presented in Table 2. The pulp recorded a Total Soluble Solid (TSS) content of 14.30 ^{0}Brix, acidic in taste indicated by the high total acidity, good amount of vitamin C, low in protein, fat, iron and carotenoids. It had a good amount of Hydroxy Citric Acid (HCA, 2.64%), which is important for ant-obesity properties, with 1.44% fibre, high in calcium (756.67 ppm), potassium (649.52 ppm), total phenols (382.25 mg GAE/g), total flavonoids (6.58 mg QE/g) and 87.32% antioxidant activity as radical scavenging assay. The *G. gummi-gutta* fruit aril was found to be a good source of minerals and bioactive compounds. The fruit's physical and biochemical parameters were examined between the aril and rind of two *G. gummi-gutta* morphotypes of the Andaman and

Table 2. Biochemical composition of *G. gummi gutta* aril

TSS (^{0}Brix)	14.30 ± 0.093
Total acidity (%)	1.91 ± 0.046
Vitamin C (mg 100 g^{-1})	11.18 ± 0.341
Protein (%)	0.36 ± 0.032
HCA (%)	2.64 ± 0.109
Carotenoid (mg g^{-1})	0.003 ± 0.010
Fibre (%)	1.44 ± 0.026
Fat (g 100 g^{-1})	0.14 ± 0.006
Fe (ppm)	4.19 ± 0.048
Ca (ppm)	756.67 ± 0.263
K (ppm)	649.52 ± 1.26
Total phenols (mg GAE g^{-1})	382.25 ± 2.43
Total flavonoids (mg QE g^{-1})	6.58 ± 1.53
Antioxidant activity (%)	87.32 ± 1.47

Values are mean ± SD of 15 replicates

Table 1. Fresh fruit characters of *G. gummi-gutta* varieties

Varieties	Fresh fruit weight (g)	Rind weight (g)	Rind percentage (%)	Seed weight (g)	Seed percentage (%)	Pulp weight (g)	Pulp percentage (%)
Haritham	99.80 ± 13.97^b	68.40 ± 10.26^c	68.47 ± 1.92^b	17.20 ± 1.92	17.31 ± 1.00^a	14.20 ± 2.68^b	14.22 ± 1.55^b
Nithya	125.92 ± 7.49^a	92.78 ± 4.26^a	73.75 ± 2.26^a	20.03 ± 3.71	15.90 ± 2.75^{ab}	13.11 ± 2.90^{bc}	10.35 ± 1.76^c
Amrutham	99.76 ± 7.89^b	70.75 ± 8.99^{bc}	70.75 ± 4.39^a	18.44 ± 1.33	18.58 ± 1.93^a	10.57 ± 2.44^c	10.67 ± 2.68^c
Local	117.90 ± 8.84^a	80.69 ± 8.36^b	68.33 ± 2.43^b	16.84 ± 2.43	14.27 ± 2.29^b	20.38 ± 1.79^a	17.41 ± 2.43^a

Values are mean ± SD of 30 replicates

Nicobar Islands and the results were similar to the current study (16). The HCA and acidity as tartaric acid in dried *G.gummigutta* rind varied with genotypes (17).

Identification and quantification of sugars of *G. gummi-gutta* fruit aril through profiling recorded 15 sugars, of which fructose was identified as the major sugar (47.44 mg/g) and other prominent sugars were glucose (21.759 mg/g) and mannose (2.61 mg/g). (Table 3 and Fig. 1). The organic acid profiling (Table 4 and Fig. 2) identified eleven distinct acids, with HCA being the most abundant acid (522.99 mg/g) followed by citric acid (110.00 mg/g). HCA is the main organic acid present in the *G. gummi-gutta* having ant-obesity properties and is valued most in the health and pharmaceutical industries. The HCA was reported as the major acid in *G. gummi-gutta* rind (17, 18) and it was found to be the major acid in *G. gummi-gutta* aril/pulp in the current study, narrating its importance in the food industry.

The RTS beverages from *G. gummi-gutta* fruit pulp, along with different fruit juice blends, were formulated and based on the sensory analysis conducted, the best formulations were selected (Fig. 3). The RTS beverage from *Garcinia* pulp alone, with 10% pulp and 15% sugar (G2) recorded the highest mean score for appearance (8.00), colour (7.80), flavour (8.20), taste (8.10) and overall acceptability mean score of 7.98. The *Garcinia*-pomegranate blended RTS beverage (GP3) recorded the highest sensory mean score of 8.40, 9.00, 8.40, 8.40, 8.60 and 8.56 for appearance, colour, flavour, texture, taste and overall acceptability respectively.

Table 3. Sugar profiling of *G. gummigutta* fruit aril

Sugars	(mg g^{-1})
Ribose	0.374 ± 0.010
Arabinose	1.824 ± 0.017
Xylose	0.013 ± 0.000
Rhamnose	0.017 ± 0.000
Fucose	0.018 ± 0.000
Fructose	47.445 ± 0.009
Glucose	21.759 ± 0.061
Mannose	2.631 ± 0.012
Galactose	0.436 ± 0.007
Inositol	0.150 ± 0.010
Sorbitol	0.519 ± 0.044
Sucrose	0.080 ± 0.001
Maltose	0.009 ± 0.002
Trehalose	0.001 ± 0.000
Lactose	0.002 ± 0.000

Values as mean ± SD of 3 replicates

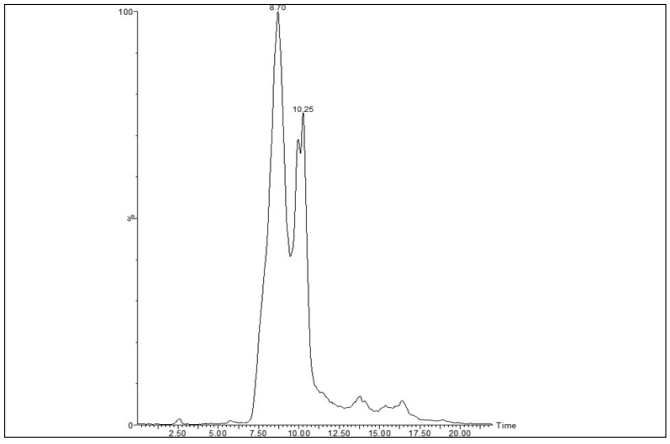

Fig. 1. Chromatogram of sugar profiling of *G. gummi-gutta* aril

The best formulation of *Garcinia*-watermelon blended RTS beverage (GW2) recorded the highest mean score of 8.20 for appearance, colour score of 8.10, flavour score as 8.00 and 8.04 for overall acceptability. The highest sensory score for flavour (8.00), appearance (8.30), colour (8.10) and overall acceptability (8.10) was recorded for *Garcinia*-dragon fruit blended RTS beverage (GD2). The blended squash from *G. gummi-gutta* rind with pineapple juice in the ratio 75:25 recorded a higher sensory mean score (19). The RTS beverage developed from *G. pendunculata* with sugar, jaggery and jeera powder recorded the highest sensory score for appearance, mouth feel, flavour and overall acceptability on a 9 point hedonic scale (20).

The selected best RTS beverage formulations were analysed for the biochemical composition and the results are

Table 4. Acid profiling of *G. gummigutta* fruit aril

Organic acids	(mg g^{-1})
Lactic acid	0.013 ± 0.001
Pyruvic acid	2.453 ± 0.88
Shikimic acid	0.560 ± 0.042
Malonic acid	1.014 ± 0.067
Maleic acid	0.066 ± 0.005
Succinic acid	2.811 ± 0.162
Malic acid	3.079 ± 0.146
Tartaric acid	0.274 ± 0.029
Fumaric acid	0.061 ± 0.002
Citric acid	110.005 ± 0.566
Hydroxycitric acid	522.990 ± 0.443

Values as mean ± SD of 3 replicates

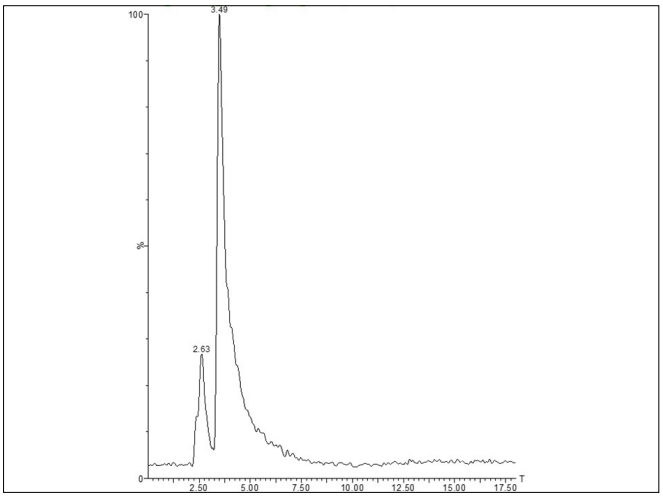

Fig. 2. Chromatogram of organic acid profiling of *G. gummi-gutta* aril

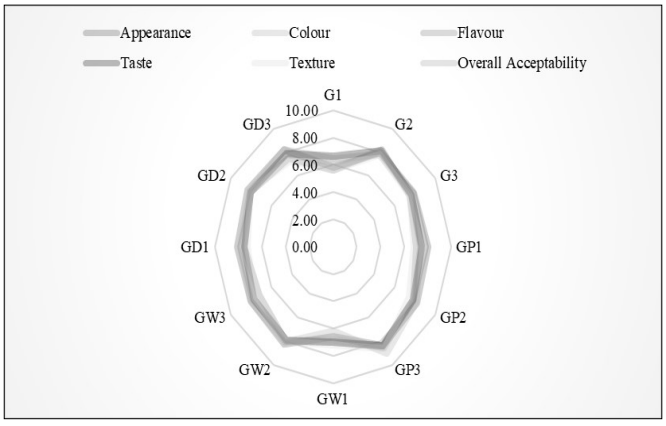

Fig. 3. Sensory mean score analysis of *G. gummi-gutta* RTS beverages

Table 5. Biochemical evaluation of developed *G. gummi-gutta* aril-based blended beverage formulations

RTS beverage formulations	TSS (⁰Brix)	Total sugar (%)	Reducing sugar (%)	Acidity (% citric acid)	Carotenoids (mg 100 g^{-1})	Vitamin C (mg 100^{-1})	Antioxidant activity (DPPH) (%)	Total phenols (mg GAE 100 g^{-1})	Total flavonoids (µg QUE g^{-1})
T_1- *G. gummi-gutta* aril	$18.10^a \pm 0.05$	$21.05^a \pm 0.03$	$9.80^a \pm 0.13$	$0.28^a \pm 0.02$	0.02 ± 0.010	$6.45^b \pm 0.101$	$63.20^b \pm 0.11$	$47.00^c \pm 0.08$	$49.44^c \pm 0.06$
T_2- *G. gummi-gutta* aril + Pomegranate juice	$17.20^b \pm 0.04$	$20.20^b \pm 0.07$	$9.26^b \pm 0.08$	$0.21^b \pm 0.04$	0.01 ± 0.005	$9.68^a \pm 0.068$	$93.12^a \pm 0.06$	$52.00^a \pm 0.13$	$51.84^a \pm 0.05$
T_3- *G. gummi-gutta* aril + Watermelon juice	$15.40^d \pm 0.15$	$14.18^d \pm 0.09$	$7.81^d \pm 0.11$	$0.20^b \pm 0.04$	0.01 ± 0.005	$3.23^c \pm 0.087$	$61.92^c \pm 0.05$	$50.00^b \pm 0.09$	$50.88^b \pm 0.07$
T_4- *G. gummi-gutta* aril + Dragon fruit pulp	$15.80^c \pm 0.14$	$18.35^c \pm 0.08$	$8.20^c \pm 0.09$	$0.15^b \pm 0.03$	0.02 ± 0.010	$3.23^c \pm 0.089$	$54.24^d \pm 0.04$	$44.50^d \pm 0.06$	$48.96^d \pm 0.08$
SEm±	0.054	0.036	0.052	0.016	0.004	0.044	0.036	0.046	0.033
CD (0.05)	0.167	0.110	0.162	0.050	NS	0.134	0.110	0.141	0.102

in Table 5. The RTS from *Garcinia* pulp only (T_1) recorded a TSS of 18.10 ⁰Brix followed by *Garcinia*-pomegranate (T_2), *Garcinia*-watermelon (T_3) and *Garcinia*-dragon fruit (T_4) blended RTS beverage. The formulated *Garcinia* RTS beverages recorded a total sugar content in the range of 14.18 to 21.05%, 7.81 to 9.8% reducing sugars and 0.15 to 0.28% acidity. Carotenoid content in all the RTS beverage formulations ranged from 0.01 to 0.02 mg /100 gwithout any significant difference and the *Garcinia*-pomegranate (T_2) recorded the highest vitamin C (9.68 mg/100 g), antioxidant activity as radical scavenging assay (93.10%), total phenols (52.00 mg GAE/100 g) and total flavonoids (51.80 µg QUE/g). The higher amount of vitamin C, phenols and flavonoids in pomegranate juice, improved the nutritional and sensory qualities of blended beverages. *Garcinia*-watermelon (T_3) recorded a vitamin C content of 3.23 mg/100g, 61.90% antioxidant acidity, 50.00 mg GAE/100 g phenols, 50.80 µg QUE/g flavonoids. *Garcinia*-dragon fruit blended RTS beverage recorded the lowest vitamin C, antioxidant activity, total phenols and total flavonoids as compared to other blended beverages. The colour of the blended beverages was appealing as the *Garcinia* pulp alone beverage had no distinct colour. The developed *Garcinia* RTS formulations exhibited nutritional qualities with consumer acceptability. The RTS beverage developed from *G. pendunculata* was found to be nutritious with good antioxidant activity (20). The mangosteen (*G. mangostana*) fruit beverage also exhibited good sensory qualities with antioxidant activity potential and total phenol content (21). The *G. cambogia* extract was found to have higher antioxidant and antimicrobial activities and has the potential to be used as a natural preservative (22).

Conclusion

Analysis of the nutritional content of *Garcinia gummi-gutta* aril showed promising results for its potential for value addition through the development of processed products which can address the evolving demands of health-conscious consumers. Many of the phytochemicals including hydroxy citric acid, antioxidants, essential vitamins and other bioactive compounds in the aril could promote wellness. The nutritional profiling showed its suitability in the food processing industry for the development of value added products with functional properties. The present study suggests that the aril could be used for the development of blended fruit beverages with nutritional benefits and consumer acceptability.

Acknowledgements

The authors are grateful to Kerala Agricultural University for the funding and facilities for the conduct of research.

Authors' contributions

GPRL conceptualized and carried out the studies, manuscript preparation and finalisation, GSA participated in the research work, data collection and analysis, KNA carried out technical support and manuscript correction. All authors read and approved the final manuscript.

Compliance with ethical standards

Conflict of interest: Authors do not have any conflict of interest to declare.

Ethical issues: None

Declaration of generative AI and AI-assisted technologies in the writing process

During the preparation of this work, the authors used CHATGPT in order to improve the language. After using this tool/service, the authors reviewed and edited the content as needed and take full responsibility for the content of the publication.

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