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Research Article

Development of a fruit-based carbohydrate-electrolyte drink and evaluation of its' physicochemical and sensory properties

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Abstract - Carbohydrate-electrolyte drinks (CEDs) are formulated to help maintain hydration during physical activity. This study aimed to develop a natural CED and evaluate its physicochemical, sensory, and microbiological properties. King coconut water, sweet orange juice, and water were used as the main ingredients. Three formulations (F1–F3) were developed by varying the proportions of king coconut water and added water, while other ingredient levels remained constant. Total sugar contents in F1–F3 were 86.84 ± 3.60, 91.47 ± 2.08, and 99.29 ± 1.86 g/L, respectively, and sodium contents were 446.33 ± 7.51, 446.33 ± 7.51, and 481.00 ± 13.00 mg/L, respectively—compliant with Standard 2.6.2 of the Australia New Zealand Food Standards Code for non-alcoholic beverages. Titratable acidity and ascorbic acid ranged from 0.55 ± 0.04 to 0.60 ± 0.04% and 147.07 ± 6.37 to 165.45 ± 19.10 mg/100g, respectively, with pH values near 3. Sensory evaluation indicated that formulation F2 had the most commercial potential. Proximate composition of F2 revealed 93.3% moisture, 6.5% carbohydrates, and 0.1% protein and ash; fat and crude fiber were not detected. The energy content was approximately 26.4 kcal/100 mL. Microbiological analysis confirmed compliance with safety standards, validating the effectiveness of pasteurization at 90 °C for 15 min. Heat treatment significantly affected ascorbic acid content, pH, total soluble solids, reducing sugars, and sodium content.

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These findings support the potential commercialization of this formulation as a natural CED with optimal functionality and acceptable taste.

Keywords: Carbohydrate-electrolyte drinks, king coconut, physicochemical properties, sensory attributes, sweet orange

1. Introduction

The normal water balance, electrolyte balance, and blood glucose level of the body at rest are disturbed when an individual is engaged in active physical activities. This affects the physical performance of the individual causing fatigue and resulting in the symptoms of dehydration. In such situations, a carbohydrate-electrolyte drink (CED) has a role to play as it is formulated as suitable for rapidly replacing fluid, carbohydrates, electrolytes, and minerals. When someone is engaging in an activity for less than one hour, it is sufficient to drink water before, during, and after to maintain adequate hydration. However, when the exercise is intense and long-lasting or is practiced in a particularly hot environment, it is advised to drink a carbohydrate-electrolyte beverage instead of plain water to maintain hydration, blood glucose levels, and energy production of the individual (Ruiz & Garcia, 2022).

Water, carbohydrates, electrolytes, and vitamins are the primary nutritional components of a CED. Among these, the total sugar and sodium content are regulated by standards. The amount of carbohydrates to be added to a carbohydrate-electrolyte drink should be within the range of 50-100 g/L while the amount of sodium should be within the range of 230 – 690 mg/L (Australia New Zealand Food Standards Code – Standard 2.6.2 – Non-alcoholic beverages and brewed soft drinks). The Scientific Committee on Food of the European Commission, in its report on the composition and specifications of food intended to meet the expenditure of intense muscular effort, especially for sportsmen, recommends that the sodium content of a CED should fall within the range of 460 to 1150 mg/L. In accordance with a position stand of the American College of Sports Medicine, the optimal composition of CEDs should range from 4 to 8 g per 100mL of carbohydrates (approximately from 4% to 8%) and from 10 to 30 mmol/L of sodium (approximately from 23 to 69 mg per 100 mL) (Silva et al., 2019). Therefore, a CED should be formulated relating to an appropriate specification depending on the requirement.

As modern consumers mostly go for natural options, most manufacturers try to follow the trend of making natural CEDs instead of making traditional synthetic CEDs. In that scenario, fruit components have been identified as a potential source of raw materials. Ruiz and Garcia (2022) suggest that fruit juice diluted at 50% is as effective as a commercial CED in rehydration and performance enhancement as they contain relatively similar amounts of carbohydrates that should be contained in CEDs. Moreover, the natural CEDs not only have economic advantages but also, they can provide additional beneficial nutrients and phytochemicals which are not found in commercial drinks (Rao et al., 2014). Therefore, fruit juices with added electrolytes are good sources for making natural CEDs. Although a wide range of flavours has been tried out by the product developers by incorporating fruit juices, the orange flavour has remained the principal flavour with the general public around the world. According to the literature, orange juice accounts for 29.1% of the global demand while the remaining share is divided between other juices and juice mixtures. Moreover, the orange flavour is within the top five when describing about the percentage introductions of fruit juices to new product developments

(Priyadarshani & Priyadarshani, 2018). These pieces of evidence suggest that the incorporation of orange flavour will maximize the attraction of consumers to the newly developing CED. Therefore, sweet orange (Citrus sinensis) which is considered as one of the most prominent fruit species of the Citrus group (Shravan et al., 2018), is incorporated into the product formulation to have the characteristic orange flavour into the product.

King coconut belongs to the semi-tall variety Aurantiaca within the species Cocos nucifera. King coconut water is a natural, nutritious, and healthy beverage which is attracted by both local and foreign consumers. The composition of king coconut water includes 94.46 ± 0.11% water, 4.95 ± 0.14% carbohydrates, 0.44±0.03% ash, 0.08 ± 0.01% protein, 0.03 ± 0.01% fat, and 0.02 ± 0.01% crude fibre (Jayasinghe et al., 2023). Moreover, it is rich in inverted sugars (mainly glucose and fructose), electrolytes (mainly Na+, K+, Ca^{+2}, and Mg^{+2}), and amino acids (mainly arginine, alanine, and cysteine) (Ediriweera, 1996; Jayasinghe & Hewajulige, 2021; Jayasinghe et al., 2022). Therefore, its chemical composition which includes mainly sugars and minerals, is ideal for using it in making CEDs.

The study was focused on developing a commercially viable, Ready-To-Drink CED in accordance with Australia New Zealand Food Standards Code – Standard 2.6.2 – Non-alcoholic beverages and brewed soft drinks utilizing sweet orange juice and king coconut water as natural fruit components and evaluating its physicochemical and sensory properties. Further, the study was performed to analyse the proximate composition and microbiological quality of the best formulation identified and examine the effect of heat treatment on the physicochemical properties of the developed formulations.

2. Materials and methods

2.1 Raw materials

Ripe sweet oranges and king coconuts with the optimum maturity stage were purchased from a local market in Kurunegala and Kandy, Sri Lanka. Other ingredients used for the formulation of CED such as purified reverse osmosis (RO) water, sugar, ginger extract, and salt were supplied by Tropical Health Food (Pvt) Ltd, Heraliyawala Industrial Park, Malkaduwawa, Kurunegala, Sri Lanka.

2.2 Preparation of raw materials

Fresh juice from sweet oranges was extracted by the method previously used by Idangodage et al. (2023) with slight modifications. The sweet oranges were sorted out by their external appearance considering the colour of the peel, size of the fruit, weight of the fruit, and absence of physiological disorders to select matured and good quality oranges with maximum juice volume. Selected fruits were washed with running tap water to remove the dirt particles. They were dipped in 100 mg/L chlorine solution for 1-2 min to disinfect properly. After that, they were rinsed with running tap water several times until the chlorine smell was eliminated. Cleaned oranges were then peeled out and cut into two halves to bt ale extracted hygienically using a cleaned handheld squeezer. Obtained sweet orange juice was then filtered twice using a cleaned muslin cloth to remove the seeds and other particles. Finally, the fresh juice was preserved at 4 °C in airtight bottles until subsequent use. Additionally, ginger extract was prepared by grinding cleaned ginger tubers without adding water followed by squeezing the juice.

The extraction of king coconut water and preparation of the king coconut water mixture was performed using a modified method referring to Cappelletti et al. (2015) and Marapana et al. (2017). King coconuts were weighed using a balance (HANA 6405L, Turkey) to obtain

the whole nut weight which is one of the convenient physical parameters for selecting king coconuts with correct maturity (six months to eight months) for processing. Sorted-out king coconuts were washed thoroughly with water followed by 100 mg/L chlorinated water to remove sand, dirt, and microbes. King coconut water was extracted by cracking the coconut shell and making a hole using a sterilized steel knife. Collected king coconut water was filtered twice using a cleaned muslin cloth. Then king coconut water was measured into a pre-sterile container which was placed in an ice bath, and the required amount of sugar (average amount of 3 g/ 100 mL of king coconut water) was added and stirred until a mixture with a °Brix value between 9 to 10 was obtained (Gunathilake, 2012; Omeka et al., 2017). The °Brix values were obtained using a digital refractometer (ATAGO PR-1, Japan). Finally, the king coconut water mixture was poured into sterile glass bottles and they were sealed and stored at 4 °C temperature immediately to avoid any oxidation of king coconut water.

2.3 Formulation of carbohydrate-electrolyte drink 3.1 Data collection

The base of selecting the percentages of raw materials was the secondary data on the physicochemical parameters of sweet orange and king coconut water published in the literature (Muhammad et al., 2018; Shravan et al., 2018; Gangakhedkar et al., 2021; Jayasinghe et al., 2023) and the preliminary trials. As described by Shravan et al. (2018), the total sugar content of sweet orange juice was 8.36 g/100 g whereas Gangakhedkar et al. (2021) recorded a total sugar content of 7.59 ± 0.4 g/100 g. Moreover, a total sugar content of 9.35 g/100 g was reported by Muhammad et al. (2018). When comparing the values, most of them are within the range of 8–12 g/100 g. The slight changes in sugar content are due to the stage of ripening of fruits. According to Jayasinghe et al. (2023), the total sugars in king coconut water fluctuated, starting from a value of 4.18 ± 0.12 g/100 mL and increased gradually to 6.08 ± 0.05 g/100 mL, and 6.58 ± 0.11 g/100 mL at 7–8 months of maturity. Glucose and fructose are responsible for reducing sugar content in king coconut water and sucrose is responsible for non-reducing sugar content. As described by Jayasinghe et al. (2023), glucose content, fructose content, and sucrose content of king coconut water at 8 months of maturity were 2.98, 3.49, and 1.69 g/100 mL, respectively. Considering those information, three formulations (F1, F2, F3) were developed as shown in Table 1 assuring sugar and sodium contents were within the range of acceptability given in the standard 2.6.2 – non-alcoholic beverages and brewed soft drinks under Australia New Zealand Food Standards Code, by changing the percentages of purified RO water and king coconut water mixture while the percentages of sweet orange juice, salt, and ginger extract were kept constant.

Table 1. Formulations of CED (per 100 mL)

Formulation	Purified RO water (%)	King coconut water mixture (%)	Sweet orange juice (%)	Salt (%)	Ginger extract (%)
F1	30	30	40	0.2	0.2
F2	25	35	40	0.2	0.2
F3	20	40	40	0.2	0.2

According to the formulations mentioned in Table 1, all the ingredients were mixed in sterile containers and heated up to 72 °C while continuously stirring (Jayasinghe et al., 2022). Then the hot filling was done separately into pre-sterile glass bottles and bottles were pasteurized at 90 °C for 15 min (Gunathilake, 2012; Ramos & Galvez, 2016; Omeka et al., 2017) in a steam sterilizer (NUVE OT 40L, Turkey). Finally, all the bottles (Figure 1) were cooled and stored for further analysis.

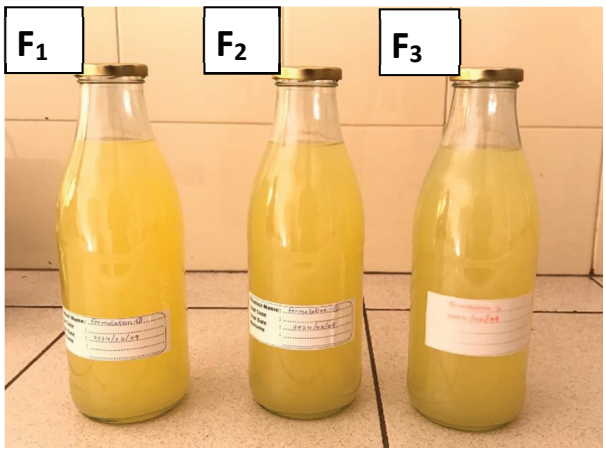

Figure 1. Formulations of CED after the heat treatment. Formulation 1 (F1), Formulation 2 (F2), Formulation 3 (F3)

2.4 Determination of physicochemical properties

The physicochemical properties were evaluated for the three formulations of CED before and after the heat treatment.

2.4.1 Total sugar content

The total amount of sugar was determined by the Lane and Eynon method referring to the method specified by Sewwandi et al. (2020) with appropriate modifications. Accurately, 10.0 g of each sample was weighed using an analytical balance (RADWAG AS 220 R2, Poland) into a 250 mL conical flask, and 100 mL of distilled water was added. After adding 3.0 mL of conc. hydrochloric acid (HCl), each sample was boiled for 3 min and allowed to cool. They were neutralized using a 10 % NaOH solution and transferred into 250 mL volumetric flasks separately and volumed up using distilled water. The burette was filled with the above-prepared solution. Then 5.0 mL of Fehling's A, 5.0 mL of Fehling's B solution, and 10.0 mL of distilled water were pipetted out into 100 mL conical flasks and mixed well.

A few pumice stones and 2-3 drops of methylene blue indicator were added into one conical flask. The preliminary titration was performed by titrating one aliquot which was heated for boiling, with the prepared sugar solution to obtain the approximate endpoint. The endpoint was determined at the point where the blue color of the indicator disappeared to colorless. After obtaining the approximate endpoint, another two aliquots were titrated directly near the endpoint without adding the indicator. When it was near the endpoint, the indicator was added, and titration continued until the endpoint was observed. From the obtained readings, the total sugar contents of each sample were calculated using Equation 1.

$$Total\ sugar\ content(gL^{-1}) = \frac{4.95 \times A \times D}{m \times V} \quad (1)$$

Where,

V is the volume of sugar solution required for the titration in ml

A is the volume of Fehling's A solution in ml

D is the dilution factor

m is the mass of the sample taken for the test in g

4.95 is the conversion factor (1 mL of Fehling's solution = 4.95 mg of glucose)

2.4.2 Reducing and non-reducing sugar content

For the determination of reducing sugars, 10.0 g of each sample was weighed using an analytical balance (RADWAG AS 220 R2, Poland) and transferred into 250 mL volumetric flasks using 100 mL distilled water. After mixing properly, the solutions were volume up. From this prepared solution the burette was filled and the titration was performed against Fehling's solution according to the same procedure as in the determination of total sugars in section 2.4.1. By subtracting the value of reducing sugar content from the value of total sugar content, the non-reducing sugar content was taken.

2.4.3 Sodium content

The sodium contents were determined according to the flame spectrophotometric method as described in AOAC method 966.16 (2005). First, sodium standard solution was prepared using reagent grade NaCl to prepare the dilution series consisting of 0, 10, 20, 30, 40, and 50 mg/L sodium solutions. The absorbance values for each dilution were taken using a flame photometer (JENWAY, D-07.2 CS.AL.02, Japan), and a standard curve was plotted. The samples were filtered using ashless filter papers (Whatman 42) and a known amount of the filtrates was diluted to reduce the sodium concentrations to the range covered by the flame photometer (JENWAY, D-07.2 CS.AL.02, Japan). Then they were aspirated directly into the flame one by one. Finally, the absorbance values for each sample were recorded and the standard curve was used to determine the sodium concentrations.

2.4.4 Total soluble solids (TSS)

TSS contents in terms of °Brix values of all the samples were measured in triplicates separately using a digital refractometer of 0-30 °Brix (ATAGO PR-1, Japan) according to the method specified in AOAC methods 932.12; 983.17 (2005). After making the refractometer into zero using distilled water, approximately 1.0 mL of the sample was placed on the lens of the refractometer and the reading was taken directly.

2.4.5 pH

The pH values of the samples were measured in triplicates using a calibrated digital pH meter (EXTECH Instruments PH100, China). The probe was placed in 10 mL of sample and pH was measured at

25 °C according to the method specified in Effiong and Udofia (2017).

2.4.6 Titratable acidity

The titratable acidities in terms of citric acid percentage were determined according to the method specified in the SLS 729 (2010) with slight modifications. About 10.0 g of each sample was measured into a 100 mL volumetric flask using an analytical balance (RADWAG AS 220 R2, Poland) and volume up. A sample of 10.0 mL was pipetted into a conical flask and titrated against the standard 0.1 M sodium hydroxide (NaOH) solution using phenolphthalein as the indicator. Finally, the endpoint was taken by observing the color change from colorless to pink. The titratable acidity was calculated as anhydrous citric acid percentage by mass as shown in Equation 2.

$$Titratable\ acidity = \frac{192 \times V \times M \times D \times 100\%}{m \times 3} \quad (2)$$

Where,

V is the volume of standard NaOH solution required for the titration in ml

M is the molarity of standard NaOH solution

D is the dilution factor

m is the mass of the sample taken for the test in g

192 is the molecular weight of citric acid in g/mol

1/3 is the stoichiometry between citric acid and NaOH

2.4.7 Ascorbic acid content

The ascorbic acid contents were analyzed using the 2,6-dichloroindophenol titrimetric method as described in the AOAC method 967.21 (2005) with slights modifications. The standard ascorbic acid solution was prepared by weighing 50.0 mg of ascorbic acid reference standard using an analytical balance (RADWAG AS 220 R2, Poland) and transferring that amount into a 50 mL volumetric flask. The standardization of the 2,6-dichloroindophenol (DCP) solution was done using a freshly prepared standard ascorbic acid solution. A portion of 10.0 mL was pipetted from the prepared standard ascorbic acid solution and titrated against the DCP solution. The endpoint was observed as the point where the blue color of the DCP solution turned into a faint pink color and the dye equivalent value was calculated based on the reading. After the standardization process, test samples were analyzed. Accurately, 10.0 g of each sample was weighed using an analytical balance (RADWAG AS 220 R2, Poland) into 50 mL volumetric flasks and volumed up using a 2.5% metaphosphoric acid solution. Three aliquots of 10.0 mL were pipetted from it and titrated against the DCP solution until the endpoint was observed. The calculations were done as the milligrams of ascorbic acid in 100 grams of sample using the Equation 3.

$$Ascorbic\ acid\ content\left(\frac{mg}{100g}\right) = \frac{Z \times V \times D}{m} \quad (3)$$

Where,

Z is the dye equivalent value of DPC

V is the volume of DCP required for the titration of drink, in ml

D is the dilution factor

m is the mass of the sample taken for the test in g

2.5 Sensory evaluation

Sensory evaluation was carried out in individual booths at the sensory laboratory of the Department of Food Science

and Technology, Faculty of Agriculture, University of Peradeniya, Sri Lanka. The sensory panel consisted of 30 untrained sensory panelists who were chosen randomly from different ages (between 20-30 years) and gender groups (17-male and 13-female) among the university students from the Department of Food Science and Technology who were a target group of this product. Before the evaluation process, they were given clear instructions about the test. A descriptive test was performed using the category scaling method to quantify the sensory attributes. The color, aroma, taste, mouthfeel, and overall acceptability of the samples were assessed using seven points hedonic scale (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). The samples were distributed among panelists using a balanced complete block design. Each panelist received approximately 30.0 mL of refrigerated (4 °C) three samples, coded with random three-digit numbers. Water was provided to clean and rinse the palate between samples. Then they were asked to rate the intensity of the given attributes by assigning it a value referring to the given scale. Each of the responses was recorded on a separate ballot paper. (Marapana et al., 2017; Porfírio et al., 2019; Idangodage et al., 2023).

2.6 Proximate composition analysis and energy value

Proximate composition analysis was performed for the formulation selected from the sensory evaluation after the heat treatment. Moisture, protein, ash, fat, and crude fibre contents were determined according to the standard methods described by the Association of Official Analytical Chemists (AOAC, 2005). The total Carbohydrate content of the sample was determined as a total carbohydrate by difference, that is by subtracting the measured protein, fat, ash, fibre, and moisture from 100. The energy value was calculated using Equation 4 as mentioned in the AOAC (2005).

$$(Energy\ in\ calories = (9\times fat)+(4\times protein) + (4\times carbohydrates)\#) \quad (4)$$

2.7 Microbiological analysis

The Total Plate count test, Yeast & Mold count test, Coliforms, and *E. coli* tests were performed for the selected Formulation using ISO 4833-1: 2013, ISO 21527: 2008, ISO 4831: 2006, and ISO 7251: 2005 test methods, respectively.

2.8 Statistical analysis

All the assays were carried out in triplicate and the results were expressed as mean values and standard deviation (SD). The experimental design was a complete randomized design. The statistical significance among the formulations was tested by the one-way ANOVA followed by Tukey mean separation using the Minitab® 19 software. The statistical significance of the physicochemical parameters before and after the heat treatment was tested using the Paired T-test. Values with $P < 0.05$ were defined as statistically significant.

3. Results and discussion

3.1 Physicochemical properties of the formulations

The results on the TSS, pH, titratable acidity, ascorbic acid content, total sugar content, reducing sugar content, non-reducing sugar content, and sodium content of the formulations are presented in Table 2.

Table 2. Physicochemical properties of the formulations

Physicochemical property	Formulation		
	F_1	F_2	F_3
Total sugar content (g/ L)	86.84 ± 3.60[a]	91.47 ± 2.08[a]	99.29 ± 1.86[b]
Reducing sugar content (g/ L)	44.35 ± 0.27[a]	45.29 ± 0.95[a]	46.95 ± 0.10[b]
Non-reducing sugar content (g/ L)	42.48 ± 3.74[a]	45.85 ± 1.74[a]	52.67 ± 1.58[b]
Sodium content (mg/ L)	446.33 ± 7.51[a]	446.33 ± 7.51[a]	481.00 ± 13.00[b]
TSS (°Brix)	6.37 ± 0.12[a]	6.50 ± 0.00[a]	7.17 ± 0.15[b]
Ascorbic acid content (mg/100 g)	147.07 ± 6.37[a]	161.77 ± 6.37[a]	165.45 ± 19.10[a]
pH	3.64 ± 0.02[a]	3.64 ± 0.01[a]	3.63 ± 0.01[a]
Titratable acidity (citric acid%)	0.55 ± 0.04[a]	0.60 ± 0.04[a]	0.60 ± 0.04[a]

Note: Values were presented as mean ± standard deviation (SD) of triplicate determination. Means deNoted by the same letters in a row represent non-significant differences ($P > 0.05$) between formulations. F_1- Formulation 1, F_2- Formulation 2, F_3- Formulation 3

Total sugar content is the main factor that determines the quality of a CED. Too-low concentration will not optimize the flavour and will not stimulate the urge to drink spontaneously. Enough carbohydrate content is needed for an immediate energy recharge (Kailaku et al., 2015) and glycogen replenishment to enhance exercise performance. On the contrary, the excess carbohydrates delay gastric emptying and intestinal absorption (Leiper et al., 2001; Murray & Stofan, 2001). To facilitate a rapid supply of nutrients and water to an individual from the ingested beverage, it is essential to have a rapid gastric emptying rate and complete intestinal absorption. Otherwise, negative physiological impacts such as gastrointestinal discomfort, diarrhea, feeling of satiety, and loss of palatability can be experienced. Therefore, the formulation of beverages should be carried out considering these factors in such a way as to optimize the gastric emptying rate and intestinal absorption rate. Normally in the small intestine, disaccharides and polysaccharides are digested to glucose by hydrolytic enzymes. Then glucose is directly absorbed by an active, Na+-dependent transport system across the mucosa. The rate of glucose absorption reaches its maximum at a glucose concentration of 200 mmol/L but tends to continue to increase until 555 mmol/L (Leiper et al., 2001). Therefore, total sugar content has been regulated by standards. According to standard 2.6.2 – non-alcoholic beverages and brewed soft drinks under the Australia New Zealand Food Standards Code, the amount of carbohydrates to be added to a carbohydrate-electrolyte beverage should be within the range of 50-100 g/L. The total sugar contents of F1, F2, and F3 formulations were 86.84 ± 3.60, 91.47 ± 2.08, and 99.29 ± 1.86 g/L, respectively. According to the statistical analysis, the F3 formulation significantly differed from the other two formulations as it had a higher total sugar content ($P < 0.05$). However, the total sugar contents of the three formulations were within the range of 50-100 g/L given in the standard 2.6.2 – non-alcoholic beverages and brewed soft drinks under the Australia New Zealand Food Standards Code. The reducing sugar contents of F1, F2, and F3 formulations were 44.35 ± 0.27, 45.29 ± 0.95, and 46.95 ± 0.10 g/L, respectively and the non-reducing sugar contents of F1, F2, and F3 formulations were 42.48 ± 3.74, 45.85 ± 1.74, and 52.67 ± 1.58 g/L, respectively. According to the statistical analysis, both reducing and non-reducing sugar

contents of the F3 formulation significantly differed from the other two formulations by having higher sugar contents ($P < 0.05$). In contrast, when considering the F1 and F2 formulations, their sugar contents were not significantly different ($P < 0.05$). When increasing the king coconut percentage, the sugar contents of the formulations should also be increased as the king coconut water mixture is the main contributor to the sugar contents of the formulations. As F3 had the highest percentage of king coconut water mixture, it showed higher sugar contents. Although F2 had a higher percentage of king coconut water mixture than F1, statistically they did not show a difference in sugar contents. This may have occurred due to the inhomogeneity of the king coconut water mixture when formulating.

The second most important factor that determines the quality of a CED is the electrolyte content. Among the different electrolytes, sodium is the most important electrolyte. Sodium helps to maintain a proper body fluid volume. As a slight deficiency of sodium may impair the performance of active individuals, it is added in the form of salts, such as sodium chloride or sodium citrate during the formulation of CEDs (Ashurst & Palmer, 2016). The effects of low amounts of sodium are less palatability, stimulation of urine production, and delay in the hydration process. Drinks with very high sodium concentrations may taste excessively salty (Murray & Stofan, 2001). Although only sodium plays an active role in sports science, most electrolyte drinks are also fortified with potassium, magnesium, calcium, and chloride. However, they do not have a discernible impact on the absorption of water and carbohydrates. They only contribute to the osmolality of the product (Leiper et al., 2001). Therefore, mainly the sodium content has been regulated by standards. The sodium contents of F1, F2, and F3 formulations were 446.33 ± 7.51, 446.33 ± 7.51, and 481.00 ± 13.00 mg/L, respectively. Statistical analysis revealed that the F3 formulation had a significantly higher ($P < 0.05$) sodium content than the other two formulations. This is due to the higher percentage of king coconut water mixture in F3 formulation which is the main source of electrolytes. However, the sodium contents in three formulations were within the 230-690 mg/L range given in the standard 2.6.2 – non-alcoholic beverages and brewed soft drinks under the Australia New Zealand Food Standards Code. In a similar study, the sodium content of an isotonic king coconut water drink was reported as 462 mg/L which is in concurrence with the above values (Marapana et al., 2017).

TSS generally represents the sweetness of the drink and it plays a role in maintaining the appropriate palatability (Kailaku et al., 2015). TSS content or ^{o}Brix value is numerically equal to the percentage of dissolved solids in a solution, which includes reducing and non-reducing sugars, proteins, mineral salts, vitamins, organic acids, pigments, and other soluble dissolved substances (Beckles, 2012). The TSS contents of F1, F2, and F3 formulations were 6.37 ± 0.12, 6.50 ± 0.00, and 7.17 ± 0.15 ^{o}Brix, respectively. According to the statistical analysis, the F3 formulation significantly differed from the other two formulations as it had a higher TSS value ($P < 0.05$). The TSS content of this drink is determined mostly by sugars and electrolytes. According to Table 2, the F3 formulation consisted of higher amounts of total sugar and sodium content, resulting in a higher TSS content than the other two formulations. A similar study performed by Idangodage et al. (2023) has reported a higher TSS content of 9 °Brix, and it may be due to the addition of other recommended food-grade electrolytes during the formulation in addition to salt and sugar. Further, the variations in TSS contents could be due to the initial TSS contents of raw materials which were greatly affected by environmental characteristics (Beckles, 2012).

pH and titratable acidity are two important parameters that provide insight

into the product's quality. The pH values of F1, F2, and F3 formulations were 3.64 ± 0.02, 3.64 ± 0.01, and 3.63 ± 0.01, respectively indicating a non-significance among the formulations (P > 0.05). The titratable acidities in F1, F2, and F3 formulations in terms of citric acid percentage were 0.55 ± 0.04, 0.60 ± 0.04, and 0.60 ± 0.04%, respectively. They were not different significantly according to the statistical analysis (P > 0.05). The ascorbic acid contents in 100 g of F1, F2, and F3 formulations were 147.07 ± 6.37, 161.77 ± 6.37, and 165.45 ± 19.10 mg, respectively which were not significantly different (P > 0.05). Sweet orange juice mainly contributed to the pH values, titratable acidities, and ascorbic acid contents in the formulations. Natural citric acid from sweet orange juice contributes to the tartness and tangy taste that help to balance the sweetness of the drink. Further, citric acid acts as a natural preservative and an antioxidant. On the other hand, the low pH resulting from sweet orange juice is crucial for maintaining shelf stability and influencing the flavour profile. As the same amount of sweet orange was added, the pH values, titratable acidities, and ascorbic acid contents among the final formulations did not differ from each other. On the contrary, the pH value reported by Idangodage et al. (2023) who developed the isotonic drink using nas narang was lower than the present values. This might be due to the lower pH of nas narang juice than the sweet orange juice as reported by Herath et al. (2016). However, the results for titratable acidity were in concurrence with Idangodage et al. (2023). Moreover, Marapana et al. (2017) have developed an isotonic beverage that was made from king coconut water along with added ascorbic acid, with an ascorbic acid content of 400 mg/L. Its higher value than the present values may be due to the addition of ascorbic acid externally in addition to the naturally present ascorbic acid.

3.2 The effect of heat treatment on physicochemical properties of the formulations

Varying percentages of desirable constituents can be altered during the heat treatment, in addition to inactivation of microorganisms. Therefore, the effect of heat treatment on the physicochemical properties of three formulations was studied, and the experimental results are listed in Table 3.

Table 3. The effect of heat treatment on the physicochemical properties of the formulations

Physicochemical property		F1	F2	F3
Total sugar content (g/L)	Before	89.27 ± 1.95^{a}	93.28 ± 0.81^{a}	100.38 ± 2.51^{a}
	After	86.84 ± 3.60^{a}	91.47 ± 2.08^{a}	99.29 ± 1.86^{a}
Reducing sugar content (g/L)	Before	44.75 ± 1.23^{a}	48.23 ± 1.10^{a}	47.14 ± 1.61^{a}
	After	43.69 ± 0.88^{a}	45.29 ± 0.95^{a}	45.28 ± 0.48^{a}
Non-reducing sugar content (g/L)	Before	44.52 ± 0.73^{a}	45.05 ± 0.99^{a}	52.67 ± 1.58^{a}
	After	42.48 ± 3.74^{a}	45.85 ± 1.74^{a}	52.67 ± 1.58^{a}
Sodium content (mg/L)	Before	427.00 ± 3.46^{a}	424.67 ± 7.51^{a}	437.67 ± 7.51^{a}
	After	446.33 ± 7.51^{a}	446.33 ± 7.51^{a}	481.00 ± 13.00^{a}
TSS (^{o}Brix)	Before	6.63 ± 0.29^{a}	6.27 ± 0.31^{a}	6.47 ± 0.06^{a}
	After	6.37 ± 0.12^{a}	6.50 ± 0.00^{a}	7.17 ± 0.15^{b}
Ascorbic acid content (mg/100 g)	Before	180.16 ± 6.37^{a}	205.89 ± 12.74^{a}	213.20 ± 16.80^{a}
	After	147.07 ± 6.37^{b}	161.77 ± 6.37^{b}	165.40 ± 19.10^{b}

Table 3. The effect of heat treatment on the physicochemical properties of the formulations (cont.)

Physicochemical property		F1	F2	F3
pH	Before	3.75 ± 0.04[a]	3.95 ± 0.02[a]	3.73 ± 0.02[a]
	After	3.64 ± 0.02[b]	3.64 ± 0.01[b]	3.63 ± 0.01[b]
Titratable acidity (citric acid%)	Before	0.60 ± 0.04[a]	0.60 ± 0.04[a]	0.60 ± 0.03[a]
	After	0.55 ± 0.04[a]	0.60 ± 0.04[a]	0.60 ± 0.04[a]

Note: Values were presented as mean ± standard deviation (SD) of triplicate determination. When considering one formulation and one parameter at a time, means deNoted by the same letters in a column represent non-significant differences (P > 0.05) between before and after the heat treatment.

The heat treatment did not affect significantly (P > 0.05) on total sugar content, reducing sugar content, non-reducing sugar content, and sodium content of all the formulations. It suggests that total sugar content, reduced sugar content, non-reducing sugar content, and sodium content had not been changed after the heat treatment. Moreover, the effect of heat treatment for these four parameters is independent of the product formulation.

The TSS contents of F1 and F2 were not affected significantly (P > 0.05) by the heat treatment. In contrast, the TSS content increased significantly from 6.47 ± 0.06 to 7.17 ± 0.15 °Brix in F3 formulation after the heat treatment (P < 0.05). When considering the product formulations, F3 had a significantly higher TSS content than the other two formulations. This may be the reason for having a significant effect from heat treatment only for the TSS content of F3. The increment in TSS content may be due to the evaporation of water from the product during the heat treatment (Beckles, 2012).

Irrespective of the type of formulation, the ascorbic acid contents before and after the heat treatment showed a significant reduction (P < 0.05) for all the formulations. Ascorbic acid is heat liable and its oxidation at pasteurization temperature is the reason for the reduction of ascorbic acid content after the heat treatment (Kumar et al., 2023). Ascorbic acid is reversibly oxidized into dehydroascorbic acid upon exposure to light, heat, transition metal ions, and pH (Yin et al., 2022).

The pH values before the heat treatment and the pH values after the heat treatment of all formulations showed a significant difference (P < 0.05) regardless of the type of formulation. It suggests that the pH values of all formulations are reduced after the heat treatment. The reduction in pH after the heat treatment may be due to the water evaporation and concentration of free hydrogen ions (Kumar et al., 2023). Moreover, Kong et al. (2020) have highlighted that the decrease in pH could be attributed to the dissociation of citric acid since it is the predominant acid. Although there is a possibility to degrade citric acids according to the observation of pH values, that degradation might be insufficient to indicate a significant reduction in titratable acidities. Therefore, the titratable acidities of the three formulations before and after the heat treatment did not change significantly (P > 0.05). These are related to the findings of Kong et al. (2020) who studied the quality of reconstituted pomegranate juice untreated, treated with high-temperature pasteurization and mild-temperature pasteurization. However, these results are not in agreement with Benattouche et al. (2020), who evaluated the physicochemical characteristics of normal orange juice and those heated at 90 °C for 1 min. According to their findings, heat treatment had no significant effects on pH, TSS, and titratable acidity.

Sensory evaluation

The results of the sensory evaluation are presented in Table 4. According to the results, color and aroma were not significantly different among the formulations ($P > 0.05$) suggesting that those attributes were similar among them. In contrast, taste, mouth feel, and overall preference were significantly varied ($P < 0.05$) among the formulations. There was no significant difference ($P > 0.05$) in the taste between F1 and F2. The taste of F1 and F2 were significantly different ($P < 0.05$) than F3 suggesting that it has a less preferable taste than F1 and F2. The mouthfeel of F2 was significantly different ($P < 0.05$) than F1 and F3, but there was no significant difference ($P > 0.05$) between F1 and F3, suggesting that F2 has a more favourable mouthfeel than the other two formulations. There was no significant difference ($P > 0.05$) in the overall acceptability between F1 and F2, but they were significantly different ($P < 0.05$) than F3, suggesting that it has less preferable overall acceptability than F1 and F2. Therefore, the formulation F2 was selected for further analysis of carbohydrate-electrolyte drink.

Table 4. Results of the sensory evaluation of the formulations

Sensory attribute (Variable)	Formulation		
	F1	F2	F3
Taste	5.70 ± 0.22^a	5.87 ± 0.21^a	4.73 ± 0.20^b
Mouth feel	5.00 ± 0.29^b	5.73 ± 0.21^a	4.97 ± 0.24^b
Aroma	5.33 ± 0.28^a	5.43 ± 0.24^a	4.93 ± 0.24^a
Colour	5.20 ± 0.32^a	5.63 ± 0.20^a	4.93 ± 0.27^a
Overall acceptability	5.60 ± 0.24^a	6.10 ± 0.20^a	4.63 ± 0.23^b

Note: Values were presented as mean ± standard deviation (SD). In the same variable, means with the same letter are not significantly different ($P > 0.05$) from each other.

3.4 Proximate composition analysis and energy value

The proximate composition and energy value of the F2 formulation are shown in Table 5. The percentages of moisture and carbohydrate were 93.3% and 6.5%, respectively while 0.1% per each accounted for protein and ash. However, fat and crude fibre were not detected. According to the study of Marapana et al. (2017), an isotonic king coconut beverage contained 5.4 % carbohydrate content with minor quantities of protein, fat, and ash and no crude fibre aligning with the present study. Moreover, the proximate composition analysis is almost consistent with the values reported earlier by Idangodage et al. (2023). The carbohydrate contents of commercially available Yeti® and SL Sports® sport drinks have been mentioned as 5.83 g/100 mL and 7.4 g/100 ml, respectively on the labels. According to the recommendations of the American College of Sports Medicine, carbohydrate-electrolyte drinks should contain 4-8% carbohydrate content which was fulfilled by the present study, to maximize gastric emptying, enhance fluid absorption from the intestine, as well as supply energy to the working muscles. The percentage of ash content indicates the total minerals that are important in replacing electrolytes lost through sweat after prolonged exercise. The low amount of protein may be either free amino acids or enzymes or any non-protein constituents that contain nitrogen. No detection of crude fibre may be due to the filtering process carried out during the

processing (Idangodage et al., 2023). Further, an energy value of 26.4 kcal/100 mL was obtained, and it was slightly comparable to the energy values of 22.05 kcal/ 100 g and 29.44 kcal/100 mL previously reported by Marapana et al. (2017) and Idangodage et al. (2023), respectively. Moreover, the energy values of commercially available Yeti® and SL Sports® sport drinks are within the same range as they are mentioned as 25 kcal/100 mL and 28 kcal/100 ml, respectively on the labels.

Table 5. Proximate composition analysis and energy value of F2 formulation

Tested parameters	Result
Moisture (%)	93.3
Fat (%)	Not detected
Protein (%)	0.1
Ash (%)	0.1
Carbohydrate (%)	6.5
Crude fibre (%)	Not detected
Energy (kcal/100 ml)	26.4

3.5 Microbiological analysis

As shown in Table 6, the microbial count obtained for the aerobic plate count test was 9 CFU/ml while the yeast and mold counts were 5 CFU/ml. Further, Coliform and E. coli were not detected. As in the present study, yeast mold count, and Coliform counts were not observed in the study done by Marapana et al., (2017) suggesting that the pasteurization process has increased the microbial stability of the final product. Further, Omeka et al. (2017) suggested that 5 mins at 100 ºC gives microbial stability at the lowest time according to the obtained results. The obtained results for the microbial analysis of aerobic plate count, Coliform, and E. coli were within the limits given for the Ready-To-Serve fruit drinks in SLS 729: 2010. In contrast, the yeast and mold count were slightly higher than the required limit specified in SLS 729: 2010. This can be described using the findings of Jayasinghe et al. (2022). They have done a study on the identification of potentially hazardous microorganisms and assessment of physicochemical deterioration of thermally processed king coconut water under different processing conditions in Sri Lanka and found a pattern of initial lower microbial counts with gradually increased microbial counts at intermediate processing steps and significantly lowered ($P < 0.05$) counts after thermal treatment. They have identified a few thermal-resistant yeasts and molds, which have survived in the thermally processed king coconut water. Therefore, based on the results of the above study, it can be suggested that the thermal process validation of king coconut water should be targeted according to the revealed knowledge of the identified hazardous microorganisms while adhering to Good Manufacturing and Hygienic Practices with minimized handling time to avoid rapid quality deterioration. However, a shelf-life study should be conducted in future research to evaluate the shelf-life stability of the product.

Table 6. Microbiological analysis of F2 formulation

Test	Microbial count
Aerobic plate count	9 CFU/mL
Yeast and mold count	5 CFU/mL
Coliform	Not detected
E. coli	Not detected

4. Conclusion

Three formulations of fruit-based CED were developed assuring sugar and sodium contents were within the range of acceptability as per the international standards, by changing the percentage of water and king coconut water mixture while the percentages of other ingredients were kept constant among the formulations. Among the three formulations, formulation consisting of RO water, king coconut water mixture, sweet orange juice, ginger, and salt with percentages of 25, 35, 40, 0.2, and 0.2%, respectively (F2) was found to have the best sensory attributes of color, aroma, mouthfeel, taste, and overall preference. The total sugar content of F2 was 91.47 ± 2.08 g/L and the sodium concentration was 446.33 ± 7.51 mg/L. Other physicochemical properties of pH, TSS, titratable acidity, ascorbic acid content, reducing sugar content, and non-reducing sugar content of F2 were 3.64 ± 0.01, 6.50 ± 0.00%, 0.60 ± 0.04%, 161.77 ± 6.37 mg/100 mL, 45.29 ± 0.95 g/L, and 46.18 ± 2.11 mg/L, respectively. According to the proximate composition analysis, the percentages of moisture and carbohydrate were 93.3% and 6.5%, respectively while 0.1% per each accounted for protein and ash. However, fat and crude fibre were not detected. The energy value of the drink was 26.4 kcal/100 ml. The obtained results for the microbial analysis of aerobic plate count, Coliform, and E. coli were within the limits given for the Ready-To-Serve fruit drinks in SLS 729: 2010. In contrast, the yeast and mold count were slightly higher than the required limit suggesting the requirement of maintaining Good Manufacturing and Hygienic Practices during processing. Thus, the study reflects that fruit components are good sources of developing natural CEDs while maintaining good palatability as well. Further, this research demonstrates the significant effects of heat treatment on physicochemical properties. The ascorbic acid content, pH, TSS, reducing sugar content, and sodium content were affected by the heat treatment. The ascorbic acid content, pH, and reducing sugar content were significantly decreased while TSS and sodium content were significantly increased ($P < 0.05$) due to the heat treatment. However, it is recommended to perform further research on the osmolality of the drink is essential to comprehensively understand its effectiveness in terms of hydration.

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Conflicts of interests

The authors declare no conflicts of interest.

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