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PHYTOCHEMICAL AND PHYSICOCHEMICAL PROPERTIES OF SOME UNDERUTILIZED TROPICAL FRUITS

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ABSTRACT: *This study investigated the phytochemical (ascorbate, phenolic and flavonoids levels, and antioxidant activity) and physicochemical (pH, titratable acidity, brix and colour) properties of six underutilized fruits: Averrhoa carambola, Borassus flabellifer. Carica papaya, Garcinia xanthochymus, Terminalia catappa and Vitus doniana. The pH and titratable acidity of the juice expressed from these fruits range between 2.9-5.6 and 0.3-0.78 %, respectively. B. flabellifer had the highest brix of 18.2, while V. doniana had the lowest brix of 5.40. With respect to ascorbate, phenolic and flavonoid levels, C. papaya, V. doniana and G. xanthochymus had the highest levels of 91.80 mg/100 g, 94.67 mg GAE/100 g and 83.75 mg RE/100 g, respectively. The antioxidant activity of the fruits determined as IC_{50} was highest in V. doniana and the least in G. xanthochymus.*

KEYWORDS: Underutilised Fruits, Ascorbate Levels, Phenolic Content, Brix, Antioxidant Activity

INTRODUCTION

Fruits are essential nutrient suppliers which can help improve health and general wellbeing. This is due to the fact that fruits contain several beneficial phytochemical and bioactive constituents (Muthu et al., 2016). Despite their benefits, however, only hundreds of fruits are cultivated commercially for consumption leading to several fruits remaining underutilised (Padulosi et al., 2013). Among the factors that account for the underutilisation of some fruits include an under estimation of their potential use, non-availability of their complete botanical information and inadequate research on their commercial exploitation. Additionally, lack of knowledge on their food and nutritional potential, and stigma associated with their use has contribute to the underutilisation of several fruits (Dandin and Kumar, 2016).

Recently, a growing interest in the usage of underutilised fruit has emerged (Padulosi et al., 2013). This is due to the fact that some of these underutilised fruits have been found to harbour nutritionally rich bioactive compounds which could have nutraceutical and medicinal properties (Li and Siddique, 2018). Also, an increasing world population have led to the search for alternative food sources, leading to an increased interest in these underutilised fruits. Among the most common underutilised fruits include tropical fruits such as *Averrhoa carambola*, Borassus *flabellifer*. *Carica papaya*, *Garcina xanthochymus*, *Terminalia catappa* and *Vitus doniana*. These fruits can be consumed fresh when ripened, dried or processed to juices, jams and jellies to enhance their consumption (Padulosi et al., 2013). To ensure an improved usage of these fruits, information about their composition (nutritional and physiological) needs to be made available. This study was, therefore, aimed at evaluating the phytochemical and physicochemical composition of these underutilised fruits.

MATERIALS AND METHODS

Sample collection and preparation

Fruit samples were obtained from the University of Cape Coast Botanical Science Garden in Cape Coast, Ghana. All fruits were washed thoroughly with tap water to remove surface debris and peeled. Juices were pressed from the fruits and filtered through a cheese cloth. The expressed juice was used for the determination of pH, brix, colour, titratable acidity and phytochemical analysis. Ten (10) fruits were picked randomly from different trees for analysis.

Determination of pH, Brix, Titratable Acidity and Colour

The pH and brix of the juice was determined using a pH meter (B10P Benchtop pH Meter) and digital refractometer (MA871, Milwaukee Instruments USA) respectively (Ampofo-Asiama and Quaye, 2019a). The titratable acidity of the expressed was determined by titrating against 0.1 N NaOH using phenolphthalein as indicator (Ampofo-Asiama and Quaye, 2019b). The L*a*b* colour of the juice was determined using a colour meter (CS-10, CHN Spec, China). The colour of the outer surface of the juice was also determined prior to peeling the fruits.

Analysis of Flavonoid and Phenolic Content

Using mortar and pestle, 5 g of fruit mesocarp was homogenised in methanol to a total volume of 50 mL. The mixture was filtered and the filtrate used for the determination of flavonoid and carotenoid content.

Flavonoid content was determined by adding 0.1 mL of $NaNO_2$ to 0.5 mL of the methanol extract. The mixture was allowed to stand for 5 min and 0.2 mL of $AlCL_3$ was added. The resulting mixture was also allowed to stand for 6 min after which 0.5 mL of 0.5 M NaOH was added. This final mixture was incubated in the dark for 15 min and the absorbance measured at 512 nm using a spectrophotometer (Zhishen et al., 1999). Rutin (Sigma, UK) was used as the standard.

Phenolic content was quantified by adding 0.2 mL of Folin-Ciocalteau’s reagent to 1.5 mL of the fruit extract. The mixture was incubated for 4 min at room temperature, after which 0.5 mL of $NaCO_3$ (20 %) was added and kept in the dark for 30 min. The absorbance was measured at 725 nm and gallic acid was used as the standard (Ampofo-Asiama et al., 2019).

Analysis of Ascorbate Content and Antioxidant Capacity

For the determination of ascorbic acid, the fruit mesocarp was homogenised and mixed with metaphosphoric acid-acetic acid solution. To 1.5 mL of the extract 5 % trichloroacetic acid was added to a volume of 3 mL, 1 mL reagent mixture was added (Dinitrophenylhydrazine 2 g, Thiourea 5 %, Copper sulfate 0.6 %) and incubated for 1 h at 60 °C on a water bath. The mixture was cooled on ice and 2.5 mL H_2SO_4 added. After vigorous shaking, the mixture was incubated at room temperature for 20 min and the absorbance was measured at 540 nm (Ampofo-Asiama and Quaye, 2019a). Ascorbic acid was used as the standard.

The DPPH assay was used to determine antioxidant capacity after extracting with ethanol. DPPH in ethanol (0.1 mL) was added to 3 mL of ethanolic extract of the juice, shaken vigorously and allowed to stand at room temperature for 30 min. The absorbance of the mixture was measured at 517 nm using a spectrophotometer (Ampofo-Asiama and Quaye, 2019b). Ascorbic acid was used as the reference compound. Antioxidant capacity was estimated as the concentration of sample required to inhibit 50 % of DPPH free radical (IC_{50} value).

Statistical Analysis

Statistical analysis was performed using the student's t-test in SPSS (IBM, SPSS Statistics 20) at a significance level of 0.05.

RESULTS AND DISCUSSION

The measured pH, titratable acidity and brix of the juice expressed from the studied underutilised fruits is shown in Table 1. The lowest pH was observed in *A. carambola* (pH 2.9), while *B. flabellifer* had the highest pH of 5.6. *C. papaya* and *V. doniana* had the lowest and highest titratable acidity, respectively. With respect to brix, *V. doniana* had the lowest brix while *B. flabellifer* had the highest brix (Table 1).

In a study on *A. carambola* fruits, a pH of 2.20, a brix of 5.00 and a titratable acidity 0.66 % (Anim and Tano-Debrah, 2004), which is comparable to that obtained in this study was observed. Similar results on *A. carambola* have also observed by Narain et al. (2001) and Kesavanath et al. (2015). In *C. papaya* fruit, the measured pH and brix of 5.42 and 10.52, respectively, which is similar to that observed by Suwanti et al. (2018) was obtained.

Analysis on *V. doniana* gave a pH of 3.44, brix of 5.40 and titratable acidity of 0.67 % (Table 1). The measured pH is much lower compared to the 5.2 observed by Abu (2002), however, it is comparable to the pH of 3.88 observed by Ajenifujah-Solebo and Aina (2011). Although the measured acidity is comparable to that observed by Ajenifujah-Solebo and Aina (2011), a much lower brix was measured in this study. The analysis on *G. xanthochymus* [pH of 4.2, brix of 7.90, TTA of 0.66 % (Table 1)] gave results comparable to that observed by (Farinazzi-Machado et al. (2016) with the exception of a much higher brix of brix 12.3. In *B. flabellifer* fruit pulp, a pH of 5.6 and brix 18.3 was measured. The measured pH is in the range reported by Vengaiah et al. (2015), although a brix of 16.5 which is a lower than that observed in this study was observed.

The measured brix/acid ratio of the fruits is also shown in Table 1. A minimum and maximum brix/acid ratio of 8.06 (*V. doniana*) and 24.66 (*B. flabellifer*), respectively, was observed. The brix/acid ratio is an important factor that influences the sugar acid balance and the overall sensory perception of fruits (Bates et al., 2001). Generally, a brix/acid ratio of at least 8.0 is required for harvested fruits intended to be consumed fresh while fruits for juice preparation are expected to have a ration of at least 10.0 (Kimball, 1991). The results obtained shows that all fruits had a brix/acid ratio above 8.0, which means they are suitable for fresh consumption, while *B. flabellifer*, *C. papaya G. xanthochymus* and *T. catappa* can also be used in the preparation of fruit juices.

Table 2 shows the colour of the outer surface of the fruits as well as the pulp expressed from the fruit. Generally, the outer surface of the fruits recorded higher L* vales compared to the expressed fruit. This could be due to the waxy nature most fruits leading to an increased shininess/lightness of the fruit. Comparing the a*- and b*-values, it was observed that both the outer surface of the fruits and the expressed juice had the same colour, although differences in intensity could exist. For instances, although the b* values of *C. papaya* are both positive, the outer surface of the fruit, had a more intense yellow colour compared to the expressed juice. The measured hue angle of *C papaya* is similar to 50.88 observed by dos Santos et al. (2016), although they observed a lower Chroma of 13.91.

The phytochemical composition of the studied underutilised fruits is shown in Table 3. *A. carambola* had an ascorbate content of 12.63 mg/100 g, although, in other studies higher ascorbate contents of 23.4 mg/100g (Narain et al., 2001) and 35 mg/100 g were observed. (Kesavanath et al., 2015), however, observed ascorbate levels fruits which are comparable to that observed in this study. In *B. flabellifer* and *T. catappa*, lower ascorbate levels were measured compared to the observations of Vengaiah et al. (2015) and Dikshit and Samudrasok (2011). However, high ascorbate levels (more than twice) were measured in *G. xanthochymus* compared to the observation of Farinazzi-Machado et al. (2016). Also, the ascorbate level of *C papaya* was higher than the 51.2 mg/100 g and the range of 75-83 mg/100 g reported by Wall (2006) and De Souza et al. (2008), respectively. For *V. doniana*, the ascorbate levels measured in this study was lower than that reported by Abu (2002), Ajenifujah-Solebo and Aina (2011) and Egbekun et al. (1996), even though the value is similar to the levels observed by Pio-Leon et al. (2014).

The phenolic content of *A. carambola* (85.63 mg GAE/100 g) is similar to the 74.93 mg GAE/100 g observed by Khanam et al. (2015). For *C. papaya*, variable phenolic levels have been reported by different researchers. Uribe et al. (2015) observed a phenolic content rage of 23-130 in several varieties while (Maisarah et al., 2013) reported a much higher level of 272 mg/100 g.

In *G. xanthochymus*, flavonoid and phenolic contents of 83.75 mg RE/100 g and 58.75 mg GAE/100 g, respectively were measured. In a similar study, a much higher phenolic content of 219.23 mg GAE/100 g was observed, although the flavonoid content of 63.50 mg RE/100 g is similar to that observed in this study (Nabajyoti et al., 2015).

The phenolic content of *B. flabellifer* and *T. catappa* were 15.71 and 20.08 mg GAE/100 g. In *B. flabellifer* fruits collected from different locations in Sri-Lanka, an average phenolic content of 6.1 mg GAE/100 g (Kurian e al., 2017) was observed, while studies on *T. catappa* gave a phenolic content of 117.10 mg GAE/100 g.

The antioxidant capacity of the different fruits was determined by analyzing the IC_{50}. The lowest and highest IC_{50} was observed for *G. xanthochymus* (IC_{50} of 2.43) and *B. flabellifer* (IC_{50} of 23.73 mg/mL), respectively. Considering that the IC_{50} represents the concentration of expressed juice needed to inhibit 50 % of DPPH activity, the lower the IC_{50} the higher the antioxidant potential of the fruit. This means that *B. flabellifer* had the least antioxidant capacity of the studied fruits. An IC_{50} of between 0.04-7.5 mg/mL, has been reported for *B.*

flabellifer (Kurian e al., 2017), while in *C. papaya* (Maisarah et al., 2013) and *T. catappa* (Abdulkadir, 2013) an IC_{50} of 7.8 and 95.99 mg/mL has been observed, respectively.

CONCLUSIONS

This study showed that the studied underutilised fruits had high brix/acidity and appreciable levels of phytochemicals. The fruits also exhibited high antioxidant activities. This means that these fruits can be eaten fresh or processed into juice to enhance their usage and improve their economic value.

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APPENDIX

Table 1: pH, Titratable Acidity, Brix and Brix/Acid Ratio OF the Studied Underutilised Fruits.

Fruit	pH	Titratable Acidity (%)	Brix	Brix/Acid Ratio
A. carambola	2.90 ± 0.09	0.62 ± 0.15	5.50 ± 0.40	8.87
B. flabellifer	5.60 ± 0.11	0.74 ± 0.05	18.30 ± 1.99	24.66
C. papaya	5.42 ± 0.76	0.78 ± 0.67	10.52 ± 0.04	13.49
G. xanthochymus	4.20 ± 0.32	0.66 ± 0.07	7.90 ± 0.97	11.97
T. catappa	4.20 ± 0.30	0.30 ± 0.07	7.20 ± 1.54	24.00
V. doniana	3.44 ± 0.46	0.67 ± 0.14	5.40 ± 0.03	8.06

Table 2: Lab Colour, Hue Angle and Chroma of the Studied Underutilised Fruits

Fruit		L*	a*	b*	Hue	Chroma
A. carambola	Surface	43.79 ± 1.08	-2.52 ± 0.41	22.30 ± 2.86	96.45	22.44
	Juice	34.74 ± 0.87	-3.65 ± 0.45	2.34 ± 3.99	147.34	4.34
B. flabellifer	Surface	56.75 ± 2.69	-3.07 ± 0.69	24.64 ± 5.25	97.09	24.83
	Juice	43.34 ± 0.29	-1.59 ± 0.03	27.63 ± 1.99	93.29	27.68
C. papaya	Surface	65.57 ± 0.68	14.46 ± 0.99	18.96 ± 1.08	52.67	23.84
	Juice	34.59 ± 0.98	8.67 ± 1.99	10.41 ± 0.12	50.21	13.55
G. xanthochymus	Surface	62.73 ± 0.87	-15.15 ± 2.65	35.99 ± 0.97	112.83	39.05
	Juice	36.03 ± 1.67	-4.43 ± 0.21	3.15 ± 0.21	144.58	5.44
T. catappa	Surface	33.46 ± 0.76	-3.90 ± 0.51	1.88 ± 0.16	154.26	4.33
	Juice	25.87 ± 1.98	-2.59 ± 0.36	2.55 ± 0.29	135.45	3.63
V. doniana	Surface	28.08 ± 0.88	-0.97 ± 0.04	3.25 ± 0.62	286.62	3.39
	Juice	34.25 ± 0.87	-3.12 ± 0.65	3.42 ± 0.29	132.37	4.63

Table 3: Phytochemical and Antioxidant Composition of the Studied Underutilised Fruits

Fruit	Ascorbate	Phenols	Flavonoids	Antioxidant activity (IC$_{50}$)
	(mg/100 g)	(mg GAE/100 g)	(mg RE/100 g)	(mg/mL)
A. carambola	12.63 ± 3.68	85.63 ± 6.68	73.50 ± 4.76	14.20 ± 0.96
B. flabellifer	10.56 ± 1.51	15.71 ± 2.90	34.03 ± 5.77	23.73 ± 3.29
C. papaya	91.80 ± 12.77	19.73 ± 1.67	17.70 ± 1.47	11.59 ± 0.90
G. xanthochymus	25.13 ± 2.87	58.75 ± 6.86	83.75 ± 68.77	2.43 ± 0.06
T. catappa	41.96 ± 0.99	20.08 ± 1.96	22.70 ± 1.10	16.60 ± 4.65
V. doniana	13.28 ± 2.18	94.67 ± 13.65	18.03 ± 1.02	22.11 ± 2.68