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Physico-chemical properties and sensory acceptance of mixed drinks of red cabbage (*Brassica oleracea* L.) and roselle (*Hibiscus sabdariffa* L.) extracts

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Abstract

The present work aimed to determine the best formulation of mixed drink made from red cabbage and roselle extracts. Six mixed drink formulations were prepared based on different ratios of red cabbage to roselle extracts: F1 (30:20); F2 (35:15), F3 (40:10); F4 (20:30); F5 (15:35) and F6 (25:25). Samples were prepared by pasteurising the mixed drinks with the pre-determined amount of ingredients at 90°C for 5 sec. The pH of roselle extract (2.06 ± 0.02) was found to be more acidic as compared to red cabbage extract (6.38 ± 0.03). This contributed to the low pH in mixed drink formulations (2.68-3.48). Total titratable acidity (TTA) (0.44-0.89% malic acid w/v) and total anthocyanin content (247.99-339.77 mg cyanidin-3-glucoside/L) were shown to increase significantly with increasing roselle extract concentration. Similarly, total soluble solids (TSS) (12.23-12.83°brix) was found to increase significantly with higher ratio of red cabbage extract due to high TSS content (7.67 ± 0.08°brix) in the extract as compared to roselle extract (4.63 ± 0.09°brix). There were significant differences among all the samples in L^*, a^* and b^* values. Mixed drink formulations that contained higher concentration of red cabbage extract were lighter and redder in colour. Significant differences were observed in sweetness, sourness, taste and overall acceptability of drinks, with F3 yielding the highest mean scores for all attributes. Mean score of sensory attributes of the mixed drinks were related to physico-chemical properties. Mean scores of all sensory attributes had negative correlation with TTA. Sweetness mean score was positively correlated to the L^* value, sourness mean score was positively correlated with pH, L^* and a^* values but negatively correlated with b^* value, and colour mean score had positive correlation with a^* value. Therefore, it can be concluded that panellists preferred less sour drinks, which were lighter and redder in colour.β-carotene onto the PKSAC in a model system but yielded the highest desorption efficiency. .

Introduction

The changes in lifestyles and the awareness on the importance of consuming a healthier diet has influenced the growth of the world juice market. Therefore, the demand for vegetable and fruit juices has risen all over the world including Malaysia. Currently, beverages like juices, blends, smoothies, fermented and fortified beverages are becoming a trendy way to consume fruits and vegetables; thereby contributing to a healthy lifestyle (Wootton-Beard and Ryan, 2011; Corbo *et al*., 2014; Marsh *et al*., 2014; Ramachandran and Nagarajan, 2014).

Red cabbage (Brassica oleracea L.) belongs to family Brassicaceae, and is a native vegetable of the Mediterranean region and south-western Europe (Arapitsas *et al*., 2008). In Malaysia, red cabbage is known as "kobis ungu". According to Mazza and Miniati (1993), anthocyanins from red cabbage could change from red at low pH (acidic) to blue/green at high pH (alkaline). Therefore, red cabbage's usage is not only restricted to acidic food products but can also be extended to neutral food products. There are many usages of red cabbage's anthocyanins such as to impart colour on various beverages, candies, dry-mix concentrates, chewing gums, yoghurts and sauces (Dorota and Janusz, 2007).

Roselle (*Hibiscus sabdariffa* L.) is widely grown

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in Malaysia and other countries such as Indonesia, Africa and America (Aishah *et al*., 2013). Roselle has unique brilliant red colour, and is locally known as "*asam susur*", "*asam paya*" or "*Ribena Malaysia*" due to its similarity with cranberries in flavour (Mohd-Esa *et al*., 2010). The roselle calyx can be processed into various food products such as syrup, juice drinks, jellies, wine, jams and natural food colourants (Tsai *et al*., 2002; Tsai and Huang, 2004; Duangmal *et al*., 2008; Hussein *et al*., 2010).

The consumers' demand for natural and healthy products has led to an increase in the innovation of mixed drink of vegetable- and fruit-origins such as cabbage and orange mixed juice (Frederick *et al*., 2016), beet and orange mixed juice (Porto *et al*., 2017) and mixed beverage from kale, ginger, coconut water and orange (Filla *et al*., 2018); which are not only rich in phytochemicals but also possess properties that could reduce the risk of diabetes, aging and chronic health conditions such as cardiovascular diseases and cancers (Pandey *et al*., 2009). There have been many studies on mixed drinks developed from the combination of roselle extract and vegetables/fruits such as roselle-apple, roselle-pineapple, roselle-mango and pineapple-carrot-orange-roselle drinks (Fasoyiro *et al*., 2005; Mgaya-Kilima *et al*., 2015; Ogundele *et al*., 2016). However, the combination of roselle and red cabbage extracts mixed drink has not been studied yet. Therefore, the aim of the present work was to determine the best formulation of mixed drink made from red cabbage and roselle extracts by characterising its physico-chemical properties and evaluating its sensory acceptance.

Materials and methods

Extraction of roselle and red cabbage

Dried roselle calyxes were obtained from MARDI Kuala Terengganu, Terengganu, Malaysia. The extraction of roselle was done based on the method described by Chumsri *et al*. (2008). Briefly, dried roselle calyxes were soaked in water at 1:10 ratio, and heated at 50°C for 30 min. Then, the roselle extract was filtered through a muslin cloth. Red cabbage was purchased from a local supplier at Kompleks Pasar Borong Seri Kembangan, Selangor, Malaysia, and extracted using a juice extractor (Santos, France). Then, the red cabbage extract was filtered through a muslin cloth. Both the roselle and red cabbage extracts were stored in high density polyethylene (HDPE) bottles at freezing temperature (-18°C) prior to mixed drink preparation.

Preparation of mixed drink of red cabbage and roselle extracts

Six formulations of mixed drinks were developed based on the proportion of red cabbage and roselle extracts respectively: F1 (30:20); F2 (35:15), F3 (40:10); F4 (20:30); F5 (15:35) and F6 (25:25), while C1 (0:50) and C2 (50:0) served as controls (Table 1). The mixed drinks were prepared by mixing the extracts with the ingredients, and pasteurised at 90°C for 5 sec. The mixed drinks were hot-filled into polypropylene (PP) bottles and cooled rapidly to room temperature by immersing the bottles in chilled water.

Determination of total soluble solids, pH and total titratable acidity

The total soluble solid (TSS) contents were measured using a pocket refractometer (Atago, Tokyo, Japan) with a scale of 0–53°Brix. The pH of the samples was measured using a pH meter (FE20, Mettler Toledo, Switzerland). The total titratable acidity was determined by titrating 20 mL samples with 0.1 M NaOH until pH 8.1, and the results were expressed as percentage of malic acid.

Determination of colour intensity

The colour intensity of the mixed drink samples were measured using Chroma Meter Minolta CR-400/410 (Minolta Co., Osaka, Japan) based on L^* a^*

Table 1. Formulation of controls and mixed drinks of red cabbage and roselle extracts

Sample	Red cabbage extract (%)	Roselle extract(%)	Sucrose(%)	Honey(%)	Arabic gum(%)	Citric acid(%)	Water (%)	Sodium benzoate (%)
C1	0.0	50.0	5.0	5.0	0.2	0.1	39.7	0.03
C2	50.0	0.0	5.0	5.0	0.2	0.1	39.7	0.03
F1	30.0	20.0	5.0	5.0	0.2	0.1	39.7	0.03
F2	35.0	15.0	5.0	5.0	0.2	0.1	39.7	0.03
F3	40.0	10.0	5.0	5.0	0.2	0.1	39.7	0.03
F4	20.0	30.0	5.0	5.0	0.2	0.1	39.7	0.03
F5	15.0	35.0	5.0	5.0	0.2	0.1	39.7	0.03
F6	25.0	25.0	5.0	5.0	0.2	0.1	39.7	0.03

b^* colour system. L^* denotes the lightness on a 0 – 100 scale from black to white, while a^* and b^* denote the redness (+) or greenness (–) and yellowness (+) or blueness (–) hues, respectively.

Determination of total anthocyanin contents

The total anthocyanin (TA) contents were determined by the pH-differential method (Guisti and Wrolstad, 2001). Absorbance was recorded using UV-Vis spectrophotometer (Perkin Elmer, United Kingdom) at 520 nm and 700 nm against water as a blank. The measured samples should be clear and contained no haze or sediments before readings were taken. Anthocyanin pigment concentration was calculated and expressed as cyanidin-3-glucoside equivalents, as follows:

$$\text{Anthocyanin pigment (mg/L)} = \frac{A \times MV \times DF \times 10^3}{\varepsilon \times 1}$$

where A = (A_{520}– A_{700}) pH 1.0 – (A_{520}– A_{700}) pH 4.5; MW (molecular weight) = 449.2 g/mol for cyanidin-3-glucoside (cyd-3-glu); DF = dilution factor; 1 = pathlength in cm; ε (molar absorptivity) = 26,900 molar extinction coefficients, in L × mol^{-1} × cm^{-1}, for cyd-3-glu; and 103 = factor conversion from g to mg.

Sensory acceptability test

The sensory evaluation was carried out at the Faculty of Science and Technology, USIM, Nilai, Negeri Sembilan, Malaysia by 40 untrained panellists. Sensory attributes were evaluated according to the degree of liking in the aspects of sweetness, sourness, taste, colour and overall acceptability. All samples were served and coded with random three digits number. Sensory attributes of the samples were evaluated using a 7-point category hedonic scale (1 = dislike very much; 4 = neither like nor dislike; 7 = like very much) as described by Meilgaard *et al*. (1999).

Statistical analysis

All analyses were done in triplicate. Experimental data were subjected to the analysis of variance (ANOVA), and the significant differences among means were determined by the Least Significant Difference (LSD) at p ≤ 0.05 using SAS software (Ver. 9.4., SAS Institute, Cary, NC, USA). Correlation analysis using the Pearson's correlation coefficient, with instrumental and sensory results, was carried out with SPSS 22.0 for Windows (IBM Corp., Armonk, New York).

Results and discussion

Total soluble solids, pH and total titratable acidity

There were significant differences in total soluble solid (TSS) contents of the formulated mixed drink samples as shown in Table 2. C2 showed significantly higher TSS (7.67 ± 0.09) than C1 (4.63 ± 0.09). Among the six formulations, F2 significantly yielded the highest TSS (12.83 ± 0.05°brix) while F4 yielded the lowest (12.23 ± 0.05°brix). The result also showed that TSS significantly increased in formulations with higher ratio of red cabbage extract. Furthermore, roselle extract naturally possesses low sugar content (3–5% dry weight) which consequently contributes to the low TSS (Wong *et al*., 2002; Da-costa-rocha *et al*., 2014; Mgaya-Kilima *et al*., 2015).

According to Tola and Ramaswamy (2014), pH can be considered as the most crucial factor in determining the heat resistance of bacterial spores. The pH of the formulated mixed drink samples in the present work

Table 2. Physico-chemical properties of controls and mixed drink formulations

Sample	Total Soluble Solids (°brix)	pH	Total Titratable Acidity(% malic acid)	Total anthocyanins content (mg cyd-3-glu/L)	Colour		
					L^*	a^*	b^*
C1	11.59 ± 0.03^g	2.50 ± 0.01^h	0.84 ± 0.01^b	227.84 ± 5.57^g	22.05 ± 0.21^b	2.13 ± 0.10^f	0.73 ± 0.04^e
C2	12.10 ± 0.00^f	4.30 ± 0.00^a	0.23 ± 0.00^g	200.79 ±1.23^h	23.40 ± 0.01^a	3.64 ± 0.07^e	-0.50 ± 0.02^g
F1	12.77 ± 0.05^b	3.13 ± 0.01^d	0.68 ± 0.03^c	271.15 ± 2.66^d	20.47 ± 0.10^e	6.08 ± 0.42^{cd}	1.04 ±0.07^d
F2	12.83 ± 0.05^a	3.29 ± 0.01^c	0.55 ± 0.01^e	254.56 ± 2.30^e	21.35 ± 0.07^d	7.95 ± 0.39^a	0.46 ± 0.03^f
F3	12.73 ± 0.05^b	3.48 ±0.01^b	0.44 ± 0.00^f	247.99 ± 4.98^f	21.53 ± 0.03^c	7.53 ± 0.08^b	-0.60 ± 0.04^h
F4	12.23 ± 0.05^e	2.79 ±0.01^f	0.61 ± 0.01^d	331.33 ± 2.71^b	20.47 ± 0.06^e	6.20 ± 0.03^c	1.38 ± 0.02^b
F5	12.37 ± 0.05^d	2.68 ±0.01^g	0.89 ± 0.00^a	339.77 ± 2.31^a	19.29 ± 0.02^g	3.58 ± 0.05^e	1.31 ± 0.02^c
F6	12.50 ± 0.00^c	2.98 ± 0.01^e	0.68 ± 0.00^c	297.84 ± 1.07^c	19.99± 0.06^f	5.82 ± 0.12^d	1.67 ± 0.03^a

Means in the same column with the same letter are not significantly different at $p > 0.05$. C1 = red cabbage/roselle extracts (0:50); C2 = red cabbage/roselle extracts (50:0); F1 = red cabbage/roselle extracts (30:20); F2 = red cabbage/roselle extracts (35:15); F3 = red cabbage/roselle extracts (40:10); F4 = red cabbage/roselle extracts (20:30); F5 = red cabbage/roselle extracts (15:35); F6 = red cabbage/roselle extracts (25:25).

Table 3. Physico-chemical properties of red cabbage and roselle extracts

Sample	Total Soluble Solids (°brix)	pH	Total Titratable Acidity(% malic acid)	Total anthocyanins content (mg cyd-3-glu/L)	Colour		
					L^*	a^*	b^*
Red cabbage extract	7.67 ± 0.09[a]	6.38 ± 0.03[a]	0.15 ± 0.01[b]	442.62 ± 3.77[b]	19.39 ± 0.01[a]	2.10 ± 0.05[a]	-0.20 ± 0.06[b]
Roselle extract	4.63 ± 0.09[b]	2.06 ± 0.02[b]	1.62 ± 0.03[a]	667.72 ± 5.62[a]	19.34 ± 0.00[b]	0.99 ± 0.01[b]	0.60 ± 0.01[a]

Means in the same column with the same letter are not significantly different at $p > 0.05$.

ranged from 2.68 to 3.48, which could be classified as high-acid food (pH < 4.6; Babajide *et al*., 2013), thus rendering the samples resistant to microbial spoilage. Table 2 shows significant differences in pH of the mixed drink samples. F5 yielded the lowest pH (2.68 ±0.01) whereas F3 yielded the highest (3.48 ± 0.01). As expected, C1 yielded the lowest pH. The result revealed that pH values significantly decreased in formulations with higher ratio of roselle extract. This could be due to the low pH of roselle extract (2.06 ± 0.02) as compared to red cabbage extract (6.38 ± 0.03) (Table 3).

Similar trend was also observed on total titratable acidity (TTA). The TTA of the formulated mixed drink samples ranged from 0.44 - 0.89% of malic acid (Table 2). C1 significantly yielded higher TTA value (0.84 ± 0.01% of malic acid) as compared to C2 (0.23 ± 0.00). This result was clearly related to the lower pH of roselle extract as compared to red cabbage extract. According to Wong *et al*. (2000), roselle has been categorised as highly acidic fruit rich in organic acids such as oxalic, tartaric, malic and succinic acids. F5 yielded the highest TTA (0.89 ± 0.00) while F3 formulation yielded the lowest (0.44 ± 0.00% of malic acid). From the result, it can be seen that TTA significantly increased with increasing roselle extract concentration. This could be attributed to the naturally higher TTA of roselle extract as compared to red cabbage extract. The TTA result is in accordance to that of Mgaya-Kilima *et al*. (2015) on roselle-mango juice blends in which the TTA increased with increasing roselle extract concentration.

Colour intensity

Colour plays an important role in product appearance as it can influence consumers' acceptance of a food product especially drink (Duangmal *et al*., 2008). There were significant differences among the formulated mixed drink samples for L^*, a^* and b^* values (Table 2). Both C1 (22.05 ± 0.21) and C2 (23.40 ± 0.01) had lighter colour (L^*) as compared to other six formulations. F3 significantly yielded the highest L^* value (21.53 ± 0.03). This result is in accordance to that of Aishah *et al*. (2013) in which different extracts namely *Hibiscus sabdariffa*, *Melastoma malabathricum* and Ipomoea batatas exhibited an increasing trend in L* values with increasing pH. On the other hand, F2 yielded redder colour intensity by having the highest a^* value (7.95 ± 0.39) while F5 yielded the least red colour intensity by having lowest a^* value (3.58 ± 0.05). According to Hee *et al*. (2011), acidic pH of less than 3.5 is essential to attain the desired red colour and stability of anthocyanins. F6 significantly yielded yellowish colour intensity by having the highest b^* value (1.67 ± 0.03) whereas F3 significantly yielded bluish colour intensity by having the lowest b^* value (-0.60 ± 0.04). This might be due to the higher proportion of red cabbage extract in the F3 formulation as the b^* values of C2 (-0.50 ± 0.02) and red cabbage extract (-0.20 ± 0.06) also had significantly lower b^* value as compared to C1 (0.73 ± 0.04) and roselle extract (0.60 ± 0.01). Result indicated that the addition of higher concentration of red cabbage extract in the formulations produced lighter, redder and bluish colour of the samples.

Total anthocyanin contents

There were significant differences in total anthocyanin (TA) contents among the formulated mixed drink samples as shown in Table 2, which ranged from 247.99 – 339.77 mg of cyd-3-glu/L. From the results obtained, F5 yielded the highest TA content (339.77 ± 2.31 mg of cyd-3-glu/L) while F3 yielded the lowest (247.99 ± 4.98 mg of cyd-3-glu/L). On the contrary, the TA contents of C1 (227.84 ± 5.57 mg of cyd-3-glu/L) and C2 (200.79 ±1.23 mg of cyd-3-glu/L) were lower as compared to the six mixed drink formulations. This might be due to the co-pigmentation of anthocyanins with other compounds (co-pigments) such as phenolic acids, flavonoids, amino acids, alkaloids and anthocyanins themselves (self-association) (Mazza and Brouillard, 1987; Mazza and Brouillard, 1990; Davies and Mazza, 1993). According to Bakowska *et al*. (2003), combination of different extracts containing anthocyanin was the key colour-stabilising mechanism in plants. Besides, it can also be seen from the results in the present work that the TA contents significantly increased

Table 4. Mean score for sweetness, sourness, taste, colour and overall acceptability of mixed drink formulations

Sample	Sweetness	Sourness	Taste	Colour	Overall acceptability
F1	4.10 ± 1.48[bc]	4.23 ± 1.58[b]	3.98 ± 1.53[bc]	5.38 ± 1.08[a]	4.10 ± 1.50[b]
F2	4.25 ± 1.32[b]	4.38 ± 1.39[ab]	3.98 ± 1.54[bc]	5.45 ± 0.99[a]	4.15 ± 1.55[b]
F3	5.20 ± 1.02[a]	5.10 ± 0.98[a]	5.15 ± 1.14[a]	5.53 ± 0.93[a]	5.08 ± 1.10[a]
F4	4.40 ± 1.53[b]	4.38 ± 1.60[ab]	4.23 ± 1.51[bc]	5.33 ± 1.14[a]	4.43 ± 1.52[ab]
F5	3.58 ± 1.57[c]	3.58 ± 1.88[bc]	3.63 ± 1.55[c]	4.85 ± 1.79[a]	3.75 ± 1.66[c]
F6	4.13 ± 1.70[bc]	3.98 ± 1.56[b]	4.30 ± 1.62[b]	5.50 ± 1.11[a]	4.35 ± 1.55[b]

Means in the same column with the same letter are not significantly different at $p > 0.05$. F1 = red cabbage/roselle extracts (30:20); F2 = red cabbage/roselle extracts (35:15); F3 = red cabbage/roselle extracts (40:10); F4 = red cabbage/roselle extracts (20:30); F5 = red cabbage/roselle extracts (15:35); F6 = red cabbage/roselle extracts (25:25).

with increasing roselle extract concentration as the roselle extract (667.72 ± 5.62 mg of cyd-3-glu/L) had higher TA content in comparison to red cabbage extract (442.62 ± 3.78 mg of cyd-3-glu/L) as shown in Table 2. Mgaya-Kilima *et al*. (2015) also found that TA content increased with increasing roselle extract concentrations in roselle-mango juice as the roselle extract is known as a reliable source of anthocyanins. However, the TA content of the roselle extract (667.72 ± 5.62 mg of cyd-3-glu/L) in the present work differed from that reported (502.33 ± 0.52 mg of cyd-3-glu/100g) by Chumsri *et al*. (2008), and this difference could possibly be due to roselle varieties and the methods of total anthocyanin determination.

Sensory acceptance of red cabbage and roselle mixed drinks

The mean scores given by the panellists for sensory attributes of the formulated mixed drink samples are presented in Table 4. Results showed that there was no significant difference in colour among the samples. However, significant differences were observed in sweetness, sourness, taste and overall acceptability. Panellists gave F5 the lowest mean score for all attributes, including overall acceptance. This could probably be due to the sour taste of the formulation and greater TTA which were 2.68 and 0.89% of malic acid, respectively, that affected the attributes and overall acceptance of the sample. On the other hand, F3 received significantly highest score for sweetness (5.20 ± 1.02), sourness (5.10 ± 0.98), taste (5.15 ± 1.14), colour (5.53 ± 0.93) and overall acceptability (5.08 ± 1.10) as compared to the other samples. A minimum sensory score of 5.0 for all attributes would be considered essential and acceptable to establish the best formulation of the mixed drink. Therefore, F3 was chosen as the best formulation for the red cabbage and roselle mixed drink.

Correlations between sensory acceptance and physico-chemical properties of red cabbage and roselle extracts mixed drinks

The mean score of sensory attributes of the formulated mixed drinks were distinctly related to physico-chemical properties (Table 5). The mean scores of all sensory attributes were negatively correlated with total titratable acidity which means that an increase in red cabbage extract in the mixed drinks would receive higher mean scores from the panellists. The increase in red cabbage extract would mask the acidity since this extract had lower TTA as compared to roselle extract. Lawless *et al*. (2012) and Bechoff *et al*. (2014) also reported that sweetness

Table 5. Correlation between mean score of sensory attributes and physico-chemical properties of mixed drinks made from red cabbage and roselle extracts

Variables	Sweetness	Sourness	Taste	Colour	Overall Acceptability
Total soluble solids	0.332	0.448	0.236	0.516	0.200
pH	0.763	0.820*	0.683	0.747	0.666
Total titratable acidity	-0.929**	-0.954**	-0.814*	-0.852*	-0.860*
Total anthocyanins content	-0.630	-0.711	-0.539	-0.738	-0.521
L^* value	0.832*	0.912*	0.663	0.760	0.706
a^* value	0.755	0.828*	0.594	0.860*	0.648
b^* value	-0.789	-0.835*	-0.699	-0.393	-0.678

* and ** values indicate significance at $p < 0.05$ and $p < 0.01$ respectively. Correlations graphically represented were linear.

was inversely correlated to titratable acidity of the infusions and concentrates. The sweetness mean scores were positively correlated to the *L** values which means that lighter colour of mixed drink would receive higher mean score by the panellists. The sourness mean scores were positively correlated to pH, *L** and *a** values but negatively correlated to *b** values. This could be explained by the fact that F3 was the least acidic but had lighter, redder and less yellow in colour, thereby receiving the highest mean score by the panellists. Vázquez-Araújo *et al*. (2010) working on mixed fruit juices also reported juices with high acid content had a lower acceptability. The colour mean scores were also positively correlated with the *a** values which means that panellists preferred redder colour for the mixed drinks. Bechoff *et al*. (2014) also reported that there was a positive correlation between overall acceptability with *a** values of roselle syrup.

Conclusion

The physico-chemical properties and sensory acceptance of mixed drinks formulated from red cabbage and roselle extracts have been assessed. The results showed that the total soluble solids, pH, total titratable acidity, colour, total anthocyanin contents and sensory acceptance of the formulated samples were significantly affected by the different ratios of red cabbage and roselle extracts in the mixed drinks. The increase in roselle extract in the formulations had significantly resulted in lower pH value but higher total titratable acidity and total anthocyanin contents. On the contrary, the increase in red cabbage extract in the formulations significantly resulted in higher total soluble solids and better sensory acceptance of the samples. For sensory acceptance, panellists preferred F3 formulation (40:10; red cabbage extract to roselle extract) as evidenced by the highest mean score received for all attributes as compared to the other samples. Therefore, it can be concluded that F3 formulation was the best formulation of mixed drink made from red cabbage and roselle extracts. As anthocyanins also possess potent antioxidant, it is recommended to include antioxidant activity analysis in the future research.

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