



Assessment of nutritional value and investigation of biological activities of *Hylocereus costaricensis*

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ABSTRACT

Hylocereus costaricensis (Dragon fruit), an edible cactus species of the Cactaceae family, is rich in essential nutrients like antioxidants, dietary fibers, minerals, and vitamins. The aim of the study was to analyze the proximate composition and investigate the biological activities of the edible parts (flesh and seed) of *H. costaricensis* cultivated in Bangladesh. Air-dried fruits were successively extracted by n-hexane, dichloromethane, and methanol. The moisture and ash contents of the fruits were found to be $85.95 \pm 0.53\%$ and $0.99 \pm 0.02\%$, respectively. UV and FT-IR spectral analysis showed the presence of different functional groups, which might be due to the presence of fatty acids, alkaloids, terpenoids, and phenolic compounds. The total carbohydrate content was 10.52g/100g as determined by the modified Molisch method. Fatty acid analysis revealed the presence of octanoic acid (5%), palmitic acid (16%), octadecanoic acid (41%), cis-9-oleic acid (29%), and linoleic acid (6%). The total phenolic content of different extracts was determined by the Folin–Ciocalteu method, and the value was found to be higher in the methanol extract (151.05 ± 0.34 mg GAE/g) than in the other two extracts. Total flavonoid content and total antioxidant content were determined by the aluminum chloride method and the phosphomolybdate assay, respectively. The methanol extract exhibited the highest activities (91.54 ± 0.22 mg QE/g and 40.08 ± 0.21 mg AAE/g, respectively) among all extracts in both cases.

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1. Introduction

Hylocereus costaricensis (red fleshed Dragon fruit), a tropical fruit, is very popular in South East Asia (Morton, 1987). The fruit is native to tropical areas of North and Latin America but it is now cultivated worldwide due to its high temperature and drought tolerance, well tolerated to soil salinities, nutritional and economic value. At present, dragon fruit is commercially cultivated in almost 22 countries including Australia, Cambodia, China, Malaysia, Thailand, Sri-Lanka (Nerd et al., 2002). Both edible and non-edible part contain different bioactive compounds e.g., flavonoids, polyphenols, terpenoids, alkaloids, tannins, carotenoids, betalainins that having antioxidant, antimicrobial, antiviral, anticancer, anti-inflammatory activities. *H. costaricensis*, a leafless cactus plant belonging to the Cactaceae family reaches up to 1.5 to 2.5 meters which having thin vinelike branches. The stem is triangular and having milky white flower petals. The fruit is moderately elongated which is 8.9-15.2 cm long. The pulp is pink coloured in which kidney shaped seeds are present (Abirami et al., 2021). The texture of the fruit is quite similar to kiwifruit due to the presence of black crunchy seeds. The flesh can be eaten as raw having mild sweet taste and low calories. The seed can be also eaten with flesh having nutty taste and a good source of essential fatty acids (Ariffin et al., 2009). Recently, this fruit has become very popular in Bangladesh but many of us do not know about the nutritional and medicinal

values as there is not enough study. So, the present study was designed for assessment of nutritional value and investigation of biological activities of *H. costaricensis* cultivated in Bangladesh.

2. Materials and methods

2.1. Collection of fruit materials

Fresh *H. costaricensis* fruits were gathered from the local market at Chow Rasta, Gazipur, Bangladesh. Subsequently, the fruits underwent a thorough water wash to eliminate undesired impurities and were then peeled. The fruits were subsequently sliced into small pieces of nearly the same diameter. These sliced fruits were air-dried and eventually ground into a powdered form.

2.2. Extraction

Dried fruit samples (300 g) were subjected to successive extraction using n-hexane, dichloromethane (DCM), and methanol via the maceration method, with each solvent being utilized for three days. The resulting mixture was filtered, and the collected filtrate was evaporated under reduced pressure. Ultimately, dried extracts of n-hexane (30.41 g), DCM (1.94 g), and methanol (36.92 g) were obtained.

2.3. Moisture and ash content

Approximately 2 g of finely chopped fresh fruit samples were placed into three previously weighed dried porcelain crucibles.

These crucibles were heated in an oven at 105°C for a duration of three hours. Following the heating process, the residue was cooled in a desiccator, and its weight was measured using an analytical balance. The percentage of moisture content was determined for each crucible using Eq. (1), and the mean value was subsequently determined. The remaining residue, subsequent to the moisture content assessment, was subjected to further heating within a muffle furnace (CARBOLITE-GSM 11/8) at 700°C for a period of 4 hours. After heating, the residue was cooled in a desiccator and weighed to determine the ash content using Eq. (2) (Park, 1996).

$$\% \text{ Moisture} = \frac{(\text{loss of weight (g)} \times 100)}{\text{weight of sample (g)}} \quad \text{Eq. (1)}$$

$$\% \text{ Ash} = \frac{(\text{weight of residue (g)} \times 100)}{\text{weight of sample (g)}} \quad \text{Eq. (2)}$$

2.4. FT-IR experiment of different crude extracts

Small amounts of each extract (n-hexane, DCM, methanol) were individually mixed with powdered potassium bromide. High pressure was applied to create KBr pellets. These pellets were sequentially inserted into an FT-IR spectrophotometer (Shimadzu IR Spirit), and the resulting spectra were recorded.

2.5. UV-Visible experiment of different crude extracts

A small quantity of each extract (n-hexane, DCM, methanol) was dissolved in the respective solvent. The λ_{max} value of each extract was measured using a double-beam spectrophotometer (Shimadzu UV 1800).

2.6. Total carbohydrate content

Total carbohydrate content was determined through a modified Molisch method (Fernandez et al., 2020). Roughly 10 g of the sample was placed in a 100 mL volumetric flask and water was added to the mark. After being kept at room temperature for 24 hours, the extract was filtered. Subsequently, 500 μL of this extract was mixed with 500 μL of 80% phenol, followed by 3 mL of H_2SO_4 . Absorbance was recorded at a wavelength of 488 nm using a double-beam spectrophotometer (Shimadzu UV 1800) against a blank. Standard glucose solutions of concentrations 250, 500, 800, 1000, and 1200 $\mu\text{g/mL}$ were prepared to draw a calibration curve ($y = 9.4224x + 0.1033$, $R^2 = 0.9972$) from the plot of absorbance (488 nm) versus concentration, allowing determination of carbohydrate amount.

2.7. Relative fatty acid composition

Fatty acids within the n-hexane extract's oil were assessed by comparing the retention time of standard methyl esters of fatty acids using gas-liquid chromatography. $\text{BF}_3\text{-NaOH}$ was employed to convert fatty acids into methyl esters, followed by analysis using a Gas Chromatograph with an FID detector. The HP-5 column was utilized to separate methyl esters, with the column dimensions being 30 m length, 0.25 mm diameter, and 0.25 μm thickness. The oven's temperature program ranged from 120°C for 1 minute (hold) to a gradual increase of 7°C per minute up to 270°C, followed by a 10-minute hold. Carrier gas N_2 (flow rate 2 mL/min) was employed in this analysis (Apurba et al., 2019; Jing et al., 2012).

2.8. Total phenolic content

Total phenolic content of different extracts was determined by Folin-Ciocalteu method (Alhakmani et al., 2013; Siddiqui et al., 2017). Approximately 0.5 mL (1 mg/mL) methanolic solution of each extract (n-hexane, DCM, and methanol) was taken into

separate test tubes. Following this, 5 mL of Folin-Ciocalteu reagent (1:10 v/v distilled water) and 4 mL of Na_2CO_3 solution were added. These solutions were vortexed and left to incubate for 30 minutes to develop color. Subsequently, absorbance was measured at 765 nm with a double-beam UV-Visible spectrophotometer against a blank. Calibration curve ($y = 0.0045x - 0.0494$, $R^2 = 0.9987$) was drawn using standard gallic acid solutions of concentrations 6.25, 12.5, 25, 50, 100, 200, and 400 $\mu\text{g/mL}$ in place of the extract. The determined total phenolic content was expressed as mg GAE/g of dry extract.

2.9. Total flavonoid content

Total flavonoid content was measured by aluminum chloride colourimetric method (Bhaigyabati et al., 2014). Each extract (n-hexane, DCM, methanol) solution (1 mg/mL in methanol) was mixed with AlCl_3 reagent (2% $\text{AlCl}_3 + 1\text{M CH}_3\text{COONa}$) in a volume of 5 mL. The mixtures were incubated for 30 minutes to allow color development, and absorbance was measured at 430 nm using a double-beam UV-Visible spectrophotometer against a blank. Calibration curve ($y = 0.0068x + 0.0002$, $R^2 = 0.9971$) was prepared using standard quercetin solution (2, 5, 10, 20, 40, 50, and 100 $\mu\text{g/mL}$) in the same manner as samples, excluding the extracts. The result was expressed in mg QE/g of dry extract.

2.10. Total antioxidant capacity

Total antioxidant capacity of different extracts (n-hexane, DCM, and methanol) was determined through the phosphomolybdenum assay (Saroar et al., 2020; Prieto et al., 1999). For this, 0.3 mL of each extract solution (1 mg/mL in methanol) was allowed to mix with 3 mL of reagent (a mixture of 0.6 M H_2SO_4 , 28 mM Na_3PO_4 , and 4 mM ammonium molybdate in a 4:2:4 ratio). To develop color, the mixtures were incubated for 90 minutes at 95°C, then absorbance was measured in a double-beam UV-Visible spectrophotometer at 695 nm against a blank solution. Total antioxidant capacity was determined from the calibration curve ($y = 0.0093x + 0.1073$, $R^2 = 0.9771$), which was plotted using a standard ascorbic acid solution (5, 10, 20, 40, 50, and 100 $\mu\text{g/mL}$) prepared through the same process as the samples but without extracts. Ultimately, the result was expressed in mg AAE/g of the dry sample.

3. Results and discussion

3.1. Moisture and ash content

The amount of moisture content in a food sample regulates the stability, texture, and quality of the food sample (Park, 1996). The moisture content of fresh fruit of *H. costaricensis* was found to be $85.95 \pm 0.53\%$ which is in accordance with the value (86%) reported by Ruzainah (Ruzainah et al., 2009). Ash content refers to the inorganic materials remaining after the oxidation of organic material in a food sample, representing the mineral content (Sonkamble and Pandhure, 2015). The ash content was determined to be $0.99 \pm 0.02\%$, aligning with the value reported by Arivalagan (Arivalagan et al., 2021).

3.2. FT-IR analysis results

The FT-IR spectrum of the n-hexane and DCM extracts consists of -OH, C=O, C=C, sp^2 C-H, and sp^3 C-H stretching peaks, along with a long chain band at 722 cm^{-1} . This indicates that these extracts might contain different fatty acids. The methanol extract shows -OH, C=O, aromatic C=C, and sp^3 C-H stretching peaks, suggesting the presence of alkaloids, terpenoids, and polyphenolic compounds in the methanol extract. The observed vibrational frequencies and inferred groups are provided in Table 1.

3.3. UV experiments

The UV-visible spectral data of hexane, DCM, and methanol extracts were observed at 270, 279, and 283 nm, respectively, and indicated the presence of conjugated dienes and chromophores

with $n \rightarrow \pi^*$ and $\pi \rightarrow \pi^*$ transition. The spectral data revealed the presence of fatty acids, alkaloids, terpenoids, and phenolic compounds in these extracts (Pavia et al., 2013).

Table 1. FT-IR observed frequency and peak assignment (Pavia et al., 2013)

n-Hexane extract		DCM extract		MeOH extract	
Frequency (cm ⁻¹)	Inferred group	Frequency (cm ⁻¹)	Inferred group	Frequency (cm ⁻¹)	Inferred group
3423 (st)	-OH	3406 (st)	-OH	3397 (st)	-OH
3007 (st)	sp ² C-H	2926 (st)	sp ³ C-H	2929 (st)	sp ³ C-H
2926 (st)	sp ³ C-H	1744 (st)	C=O	1730 (st)	C=O
1745 (st)	C=O	1651 (st)	C=C	1641 and 1414 (st)	Aromatic C=C
1651 (st)	C=C	1462 (b)	-CH ₂ -	960-690(opp)	=C-H
1457 (b)	-CH ₂ -	1167 (st)	C-O		
1374 (b)	-CH ₃	723	Long chain		
1164 (st)	C-O				
722	Long chain				

Note: st = stretching, b = bending, opp = out of plane

3.4. Carbohydrate content

Total carbohydrate content of the dried powdered fruit sample was found to be 10.52g/100g. This value is comparable to the value (11g/100g) reported by Thokchom (Thokchom et al., 2019). The carbohydrate content in *H. costaricensis* is lower than that of different local fruits, e.g., Mango (15g/100g), Jackfruit (26g/100g) (Gupta et al., 2011; Ara et al., 2014).

3.5. Relative fatty acid composition

Fatty acid analysis has shown that the n-hexane extract contains octanoic acid, palmitic acid, octadecanoic acid, *cis*-9-oleic acid, and linoleic acid (Table 2). Among these fatty acids, octadecanoic acid is predominant. Essential fatty acids (linoleic acid, *cis*-9-oleic acid) are important for human survival since they are important constituents of cell membranes, thus regulating membrane function. On the other hand, oleic acid can reduce the risk of coronary disease and decrease LDL in the body.

Table 2. Fatty acids composition in *H. costaricensis*

Fatty acids	Number of carbons	% Relative percentage
Octanoic acid	C8:0	5.06
Palmitic acid	C16:0	15.62
Octadecanoic acid	C18:0	40.98
<i>Cis</i> -9-oleic acid	C18:1 (ω -9)	28.96
Linoleic acid	C18:2	5.41

3.6. Total phenolic content

Phenolics are polyhydroxy aromatic compounds containing one or more benzene ring and hydroxyl groups (Dai and Mumper, 2010). They have antioxidant properties since they can neutralize free radicals (Zheng and Wang, 2001). Total phenolic contents for n-hexane, DCM, and methanol were 70.83 \pm 0.45, 91.27 \pm 0.34, 151.05 \pm 0.34 mg GAE/g, respectively. The methanol extract had the highest value, and the n-hexane extract had the lowest value of phenolic content. Thus, the methanol extract contains compounds with the highest antioxidant property.

3.7. Total flavonoid content

Flavonoids are compounds with variable phenolic structures that belong to secondary plant metabolites. Flavonoids are bioactive compounds since they exhibit different types of bioactivity, such as antioxidant, anti-inflammatory, and anticarcinogenic properties (Panche et al., 2016). Total flavonoid contents for different extracts were 30.09 \pm 0.09 (n-hexane), 50.12 \pm 0.15 (DCM), 91.54 \pm 0.22 (methanol) mg QE/g, respectively. It is observed that the highest value was obtained for the methanol

extract, which is consistent with the value (98.5 mg QE/g) reported by Irda (Irda et al., 2014) and the lowest value was found for the n-hexane extract.

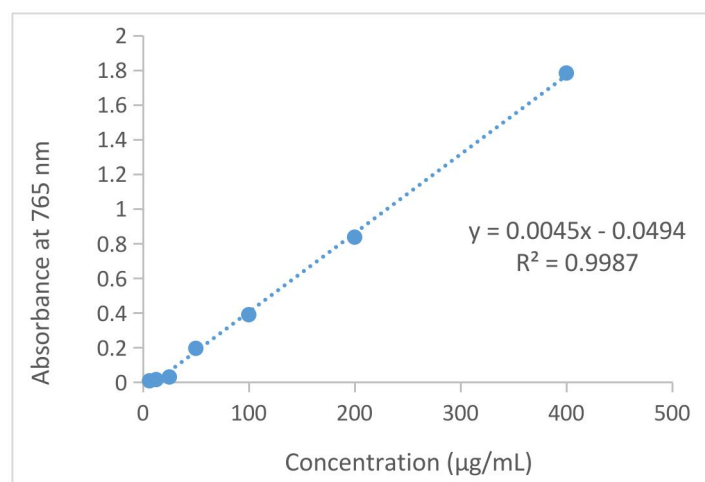


Fig. 1. Calibration curve for standard gallic acid

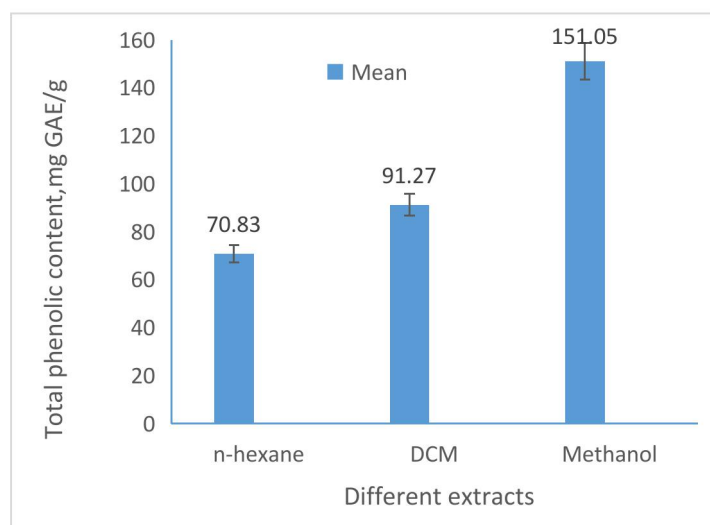


Fig. 2. TPC of different extracts of *H. costaricensis*

3.8. Total antioxidant capacity

Oxidative stress, resulting from the overproduction of reactive oxygen species, has negative effects on biomolecules such as lipids, nucleic acids, and proteins. This ultimately leads to various pathological and neurological disorders (Jennings et al., 2012; Muthusamy et al., 2008). The total antioxidant capacity of different extracts, namely n-hexane, DCM, and methanol, was found to be 15.95 ± 0.27 , 26.56 ± 0.22 , and 40.08 ± 0.21 mg AAE/g, respectively. The methanol fraction exhibited the highest antioxidant capacity, while the n-hexane fraction showed the lowest value. The presence of polyphenols and flavonoids in *H. costaricensis* contributes to its antioxidant capacity, allowing it to scavenge free radicals and have positive health effects, ultimately helping prevent various diseases.

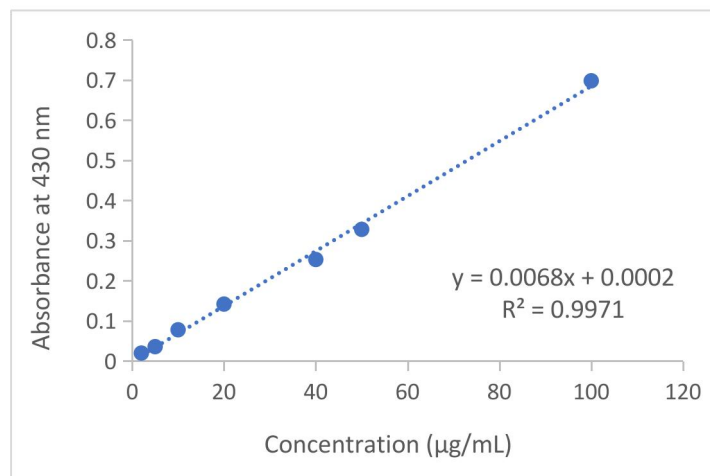


Fig. 3. Calibration curve for standard quercetin

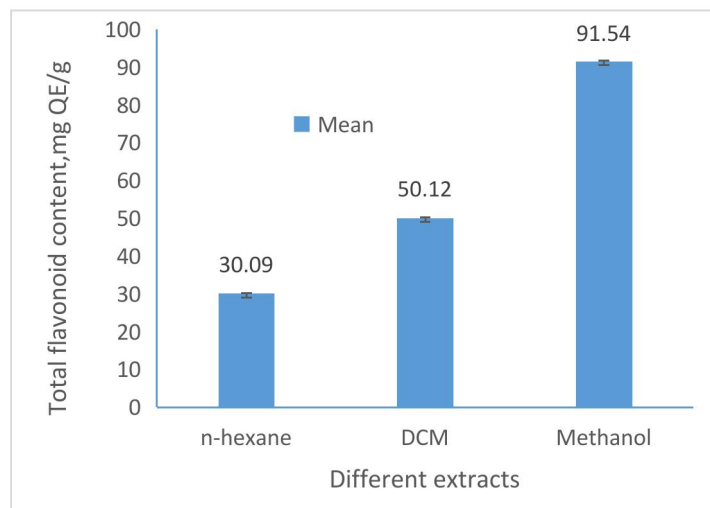


Fig. 4. TFC of different extracts of *H. costaricensis*

3.9. Total antioxidant capacity

Oxidative stress, resulting from the overproduction of reactive oxygen species, has negative effects on biomolecules such as lipids, nucleic acids, and proteins. This ultimately leads to various pathological and neurological disorders (Jennings et al., 2012; Muthusamy et al., 2008). The total antioxidant capacity of different extracts, namely n-hexane, DCM, and methanol, was found to be 15.95 ± 0.27 , 26.56 ± 0.22 , and 40.08 ± 0.21 mg AAE/g, respectively. The methanol fraction exhibited the highest antioxidant capacity, while the n-hexane fraction showed the lowest value. The presence of polyphenols and flavonoids in *H.*

costaricensis contributes to its antioxidant capacity, allowing it to scavenge free radicals and have positive health effects, ultimately helping prevent various diseases.

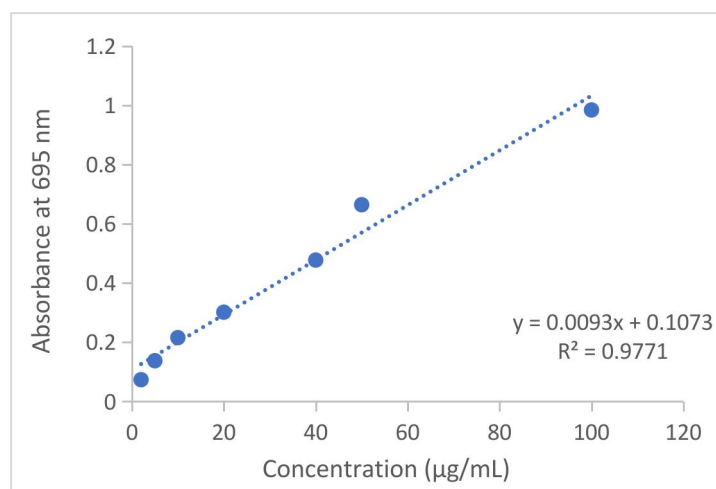


Fig. 5. Calibration curve for standard ascorbic acid

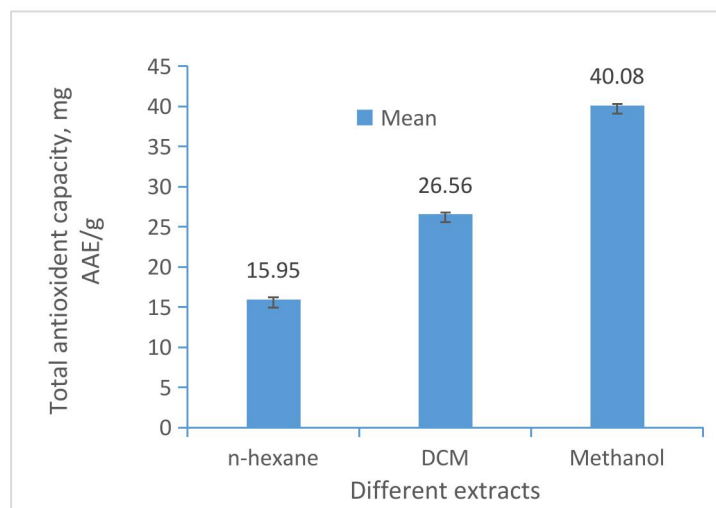


Fig. 6. TAC for different extracts of *H. costaricensis*

Dragon fruit is highly popular in Southeast Asia due to its adaptability to different climates, as well as its nutritive and medicinal properties. Additionally, it has a significant impact on the economy, particularly in developing countries within this region. The domestic demand for dragon fruit has surpassed that of other local fruits, attributed to its nutritional value and appealing taste. Commercial products like dragon fruit juice, jam, jelly, candy, and syrups are now being manufactured, contributing to the economy. Furthermore, the fruit peel can be utilized as a coloring agent and raw material in the food industry. Its low cultivation and maintenance costs have led to widespread cultivation in the region, creating employment opportunities, particularly for unskilled labor. Educated unemployed individuals are now engaging in dragon fruit cultivation, addressing unemployment issues in these developing countries while also boosting their economies. In conclusion, this fruit serves as a valuable source of daily nutrition and has a substantial economic impact.

4. Conclusion

The aforementioned study underscores that *H. costaricensis* is rich in essential nutrients, including different essential fatty acids, lower carbohydrate content, and higher mineral content. Various extracts are abundant in polyphenols and flavonoids, exhibiting extensive antioxidant activity. Thus, *H. costaricensis* proves to be a valuable source of antioxidants.

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Conflict of interest

The authors declare there is no conflict of interest in this study.

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