ANTIOXIDANT PROFILE AND PHYTOCHEMICAL CONTENT OF DIFFERENT PARTS OF SUPER RED DRAGON FRUIT (HYLOCEREUS COSTARICENSIS) COLLECTED FROM WEST JAVA-INDONESIA

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ABSTRACT

Objectives: The goals of this research were to observe antioxidant properties from different parts of super red dragon fruit (Hylocereus costaricensis) using two antioxidant testing methods which were 2,2-diphenyl-1-picrylhydrazyl (DPPH) and 2,2′-azino-bis (3-ethylbenzthiazoline-6-sulfonic acid) (ABTS).

Methods: Antioxidant activities were determined using DPPH and ABTS assays, total phenolic content (TPC) using Folin-Ciocalteu reagent, flavonoid content by Chang’s method.

Results: Inhibitory concentration 50% (IC₅₀) of DPPH scavenging activity of all of the extracts in the range of 2.69 μg/ml was −94.17 μg/ml. The ethyl acetate peel extract of super red dragon fruit expressed the highest TPC (4.56 g GAE/100 g) and the highest total flavonoid content (12.63 g QE/100 g). TPC in flesh extract of super red dragon fruit had a negative and significant correlation with their IC₅₀ of ABTS. The IC₅₀ of DPPH and IC₅₀ of ABTS of flesh extract of super red dragon fruit showed positive and significant correlation.

Conclusion: All different parts extracts of super red dragon fruit (except n-hexane flesh extract) were categorized as a very strong antioxidant by DPPH method. Phenolic compounds in flesh extract of super red dragon fruit were the major contributor in antioxidant activities by ABTS method. DPPH and ABTS showed linear results in antioxidant activities of super red dragon fruit flesh extract.

Keywords: Antioxidant, Super red, Dragon fruit, Hylocereus costaricensis.

INTRODUCTION

Phenolic compounds are produced by the plant as a defense mechanism against different stress condition. Phenolic compounds are commonly used as a subject in many researchers. Phenolic compounds included flavonoid compounds have various effects such as antioxidant activity, antibacterial activity, antidiabetic activity, and hepatoprotector [1-4]. The excessive of free radical in oxidative stress condition can be inhibited by antioxidant, which related to many degenerative diseases. Fruits and vegetables are a natural antioxidant, because they contain phenolic and flavonoid compounds which have the antioxidant capacity [5].

Some methods have been used to observe antioxidant activity in many plants extracts such as 2,2-diphenyl-1-picrylhydrazyl (DPPH), ferric reducing antioxidant power (FRAP), and 2,2′-azino-bis (3-ethylbenzthiazoline-6-sulfonic acid) (ABTS) methods [6]. The previous researchers presented that DPPH, ABTS, and FRAP can be performed to observe the antioxidant activity of fruits, vegetables, and food [5-8].

Previous studies demonstrated that red dragon fruit contained major flavonoid compounds such as quercetin and catechin [9] and betalain against different stress condition. Phenolic compounds have various effects such as antioxidant activity, antibacterial activity, antidiabetic activity, and hepatoprotector [1-4]. The excessive of free radical in oxidative stress condition can be inhibited by antioxidant, which related to many degenerative diseases. Fruits and vegetables are a natural antioxidant, because they contain phenolic and flavonoid compounds which have the antioxidant capacity [5].

The goals of this research were to observe antioxidant potential in various polarity extracts (n-hexane, ethyl acetate, and ethanol) from different parts of super red dragon fruit grown in West Java-Indonesia using DPPH and ABTS assays, and correlations of total phenolic and flavonoid content with their antioxidant activities.

MATERIALS AND METHODS

Materials

DPPH, ABTS diammonium salt, gallic acid, and quercetin were purchased from Sigma-Aldrich (MO, USA), different parts of super red dragon fruit (H. costaricensis). All of other reagents were analytical grades.

Preparation of sample

Different parts of super red dragon fruit (H. costaricensis) which were stem named as STE, peel as PEE, and flesh as FLE were collected from Bogor, West Java-Indonesia, were thoroughly washed with tap water, sorted while wet, cut, dried, and ground into powder.

Extraction

Each sample was extracted by reflux using different polarity solvents. 300 g of powdered samples was extracted using n-hexane was repeated 3 times. The remaining residue was then extracted 3 times using ethyl acetate. Finally, the remaining residue was extracted 3 times using ethanol. Hence, totally, there were nine extracts: Three n-hexane extracts (namely, STE1, PEE1, and FLE1), three ethyl acetate extracts (STE2, PEE2, and FLE2), and three ethanol extracts (STE3, PEE3, and FLE3).
Antioxidant activity by DPPH assay

Antioxidant activity by DPPH assay was conducted using a modification of Blois’s method [13]. 2 ml of various concentration of each extract were added into 2 ml DPPH solution 50 μg/ml. After 30 min incubation, absorbance was determined at wavelength 515 nm by UV-Vis spectrophotometer Beckman Coulter DU 720. Methanol was used as a blank, DPPH solution 50 μg/ml as control and ascorbic acid as standard. Analysis was conducted in triplicate for standard and each extract. Antioxidant activity was observed by calculating the percentage of reduction of DPPH absorbance. Inhibitory concentration 50% (IC₅₀) of DPPH scavenging activity of each extract can be determined using its calibration curve.

Antioxidant activity by ABTS assay

ABTS solution was prepared using a modification of Li et al. [14]. Each solution of ABTS diammonium salt 7.6 mm and potassium persulfate 2.5 mm were prepared in Agwadest and left in dark room for 12 h. The two solutions were mixed with 30 min incubation, left in the refrigerator for 24 h, and then diluted in ethano 2 ml of various concentration of each extract was added into 2 ml ABTS solution 50 μg/ml. The absorbance was read at wavelength 734 nm using UV-Vis spectrophotometer Beckman Coulter DU 720. Ethanol (95%) was used as a blank, ascobic acid as standard, and ABTS solution 50 μg/ml as a control. Analysis was performed in triplicate for standard and each extract. Antioxidant capacity of each extract by ABTS method was evaluated by observing percentage of antioxidant activity using reduction of ABTS absorbance. IC₅₀ of ABTS scavenging activity of each extract can be determined using its calibration curve.

Total phenolic content (TPC)

Determination of TPC used Folini–Ciocaltea reagent [15]. The absorbance was determined at wavelength 765 nm. Analysis was performed in triplicate for each extract. Standard solution of gallic acid (50-160 μg/ml) was used to evaluate a calibration curve. TPC was exposed as gallic acid equivalent per 100 g extract (g GAE/100 g).

Total flavonoid content (TFC)

Chang’s method [16] with minor modification was used to evaluate TFC. The absorbance was observed at wavelength 415 nm. Analysis was conducted in triplicate for each extract. Quercetin in solution 50-125 μg/ml was used to obtain a calibration curve. TPC was presented as quercetin equivalent per 100 g extract (g QE/100 g).

Statistical analysis

Each sample analysis was performed in triplicate. All of the presented results are means (± standard deviation) of at least three independent experiments. Statistical analysis using ANOVA with a statistical significance level set at p<0.05 and post hoc Tukey procedure was performed with SPSS 16 for Windows. Correlation between the total flavonoid and phenolic content and antioxidant activities and the correlation between two antioxidant testing methods were performed using the Pearson’s method.

RESULTS AND DISCUSSION

The activities and phytochemical content among the extracts can be compared if the density among the extracts were similar. One extract with high density may give the higher activity and the higher phytochemical content than low-density extract. Therefore, all extracts (nine extracts) which were used in the present study should prepare in similar density. The density of extract did not expose in 100% concentrated extract, due to its difficult to determine the density of concentrated extract using pycnometer, hence the density of the extracts was presented as density 1% extract (Table 1).

In vitro, the antioxidant activity can be categorized base on type of reaction, which are single electron transfer (SET) based assay and hydrogen atom transfer (HAT) based assay [17]. SET is based on the ability of an antioxidant to transfer one electron to reduce oxidant, meanwhile HAT based on the ability of an antioxidant to quench radical by hydrogen donation [18]. The degree of color change (either increase or decrease of absorbance of the probe at a given wavelength) is depended to the concentration of antioxidant in the sample [17]. SET and HAT mechanisms almost always occur together. Ionization potential (ΔIP), bond dissociation energy (BDE), redox potential, pH, and solvent will influence the predominant mechanism will be appeared [17,18]. Compound with ΔIP ->45 kcal/mol is predominantly by SET mechanism, while compound with ΔBDE of ~10 kcal/mol and ΔIP <-36 kcal/mol by HAT mechanism [18].

DPPH is free radical and show absorption at wavelength 516 nm. Antioxidant will transfer the hydrogen to DPPH to scavenge the free radical and DPPH will stable. DPPH in methanol give the purple color and color of DPPH solution will be changed to yellow when free radicals are scavenged by antioxidant [14]. The ability of an antioxidant to scavenge the free radical DPPH correlates with decreasing in absorbance of DPPH. IC₅₀ of DPPH scavenging activity is a concentration of sample or standard that can inhibit 50% of DPPH radical activity. The highest antioxidant activity will be shown by the lowest IC₅₀. IC₅₀ was used to observe the antioxidant activity of the sample and compared to standard.

2,2’-azino-bis-(3-ethylbenzthiazoline-6-sulfonic acid (ABTS) is not soluble in water and a polar solvent. Hence for determining antioxidant activity by ABTS assay should use ABTS diammonium salt. ABTS method is also called as Trolox equivalent antioxidant capacity (TEAC) method. TEAC is Trolox equivalent antioxidant capacity, which antioxidant activity exposed as Trolox equivalent. Sample which had higher antioxidant will give higher Trolox equivalent value.

Table 1: Density of different part extracts of super red dragon fruit

<table>
<thead>
<tr>
<th>Sample</th>
<th>N-hexane extract</th>
<th>Ethyl acetate extract</th>
<th>Ethanol extract</th>
</tr>
</thead>
<tbody>
<tr>
<td>Stem</td>
<td>0.661</td>
<td>0.891</td>
<td>0.800</td>
</tr>
<tr>
<td>Peels</td>
<td>0.664</td>
<td>0.891</td>
<td>0.799</td>
</tr>
<tr>
<td>Flesh</td>
<td>0.663</td>
<td>0.893</td>
<td>0.802</td>
</tr>
</tbody>
</table>

DPPH is free radical and show absorption at wavelength 516 nm. Antioxidant will transfer the hydrogen to DPPH to scavenge the free radical and DPPH will stable. DPPH in methanol give the purple color and color of DPPH solution will be changed to yellow when free radicals are scavenged by antioxidant [14]. The ability of an antioxidant to scavenge the free radical DPPH correlates with decreasing in absorbance of DPPH. IC₅₀ of DPPH scavenging activity is a concentration of sample or standard that can inhibit 50% of DPPH radical activity. The highest antioxidant activity will be shown by the lowest IC₅₀. IC₅₀ was used to observe the antioxidant activity of the sample and compared to standard.

The free radical of ABTS will be formed after ABTS reacting with potassium persulfate, and it will give blue color and the maximum wavelength at 734 nm. Antioxidant will scavenge the free radical, and the ability of antioxidant can be exhibited by decreasing in absorbance of the free radical ABTS.

IC₅₀ of DPPH and IC₅₀ of ABTS scavenging activities in different parts extracts of super red dragon fruit were demonstrated in Figs. 1 and 2. The lowest value of IC₅₀ means the highest antioxidant activity. Sample which had an IC₅₀<50 μg/ml was a very strong antioxidant, 50-100 μg/ml strong antioxidant, and 101-150 μg/ml medium antioxidant, while a weak antioxidant with IC₅₀>150 μg/ml [13].

Antioxidant activity by DPPH assay

Antioxidant activity in different parts extracts of super red dragon fruit by DPPH and ABTS assays were carried out by determining IC₅₀ of DPPH and IC₅₀ of ABTS scavenging activities. IC₅₀ of DPPH and IC₅₀ of ABTS scavenging activities of each extract were compared to IC₅₀ DPPH and IC₅₀ ABTS of ascorbic acid standard.

Nurliyana [12] studied regarding antioxidant activities of peels and pulp of white dragon fruit (Hylocereus undatus) and red dragon fruit (Hylocereus polyrhizus). The result showed IC₅₀ of DPPH scavenging activity of 70% ethanol peels extract of red dragon fruit (300 μg/ml) was lower than...
peels of white dragon fruit and pulps of white and red dragon fruit. It was contrast to the present study which demonstrated that IC_{50} DPPH of ethanolic stem extract of super red dragon fruit (3.56 μg/ml) was lower than their ethanolic flesh extract (6.95 μg/ml) and ethanolic peel extract (16.45 μg/ml). All of the ethanolic parts extract of super red dragon fruit (stem, peel, and flesh) can be classified as a very strong antioxidant (IC_{50} <50 μg/ml). IC_{50} of DPPH of all parts extract of super red dragon fruit varied from 2.69 to 94.17 μg/ml, while the lowest was shown by ethyl acetate flesh extract of super red dragon fruit (2.69 μg/ml). Ascorbic acid as standard had IC_{50} of DPPH 0.49 μg/ml. It can be concluded that antioxidant activity of ethyl acetate ethyl flesh extract of super red dragon fruit was around one-sixth of antioxidant activity of ascorbic acid by DPPH assay. Based on Blois’s classification [13] it could be seen that all of parts (stem, peel, and flesh) extracts of super red dragon fruit (except n-hexane flesh extract 94.17 μg/ml) can be categorized as a very strong antioxidant.

The previous study presented that red dragon fruit (H. polyrhizus) pulps juice and then be precipitated with 96% ethanol, the frozen filtrate had EC_{50} of DPPH radical scavenging activity 2.9 mm Vitamin C [19]. The 80% acetone white dragon fruit (H. undatus) peel extract had antioxidant activity 177.14 μmol AEAC/100 g by DPPH method and 109.29 μmol AEAC/100 g by FRAP method [10]. Choo and Yong [11] exposed that EC_{50} of DPPH scavenging capacities of 50% ethanolic white dragon fruit (H. undatus) pulp extract (9.91 mg/ml) was lower than its fruit (peel and pulp) extract. The result was similar to IC_{50} of DPPH of 50% ethanolic red dragon fruit (H. polyrhizus) pulp extract (9.93 mg/ml). Luo et al. [20] revealed that IC_{50} of DPPH scavenging capacity of supercritical carbon dioxide peels extract of white dragon fruit (H. undatus) 0.91 mg/ml and red dragon fruit (H. polyrhizus) 0.83 mg/ml. Study by Halimoon and Hasan [21] expressed that percentage of DPPH scavenging activity of ethanolic white dragon fruit (H. undatus) extract (63.44%) was higher than its distilled water extract and methanolic extract.

Antioxidant activities can be stated by percentage of DPPH scavenging activity and compared to the percentage of DPPH scavenging activity of ascorbic acid as standard. The percentage of DPPH scavenging activity of ascorbic acid did not achieve 100% because there was still residual yellow color in solution after antioxidant giving hydrogen atom to DPPH [22,23]. The percentage of DPPH scavenging activity cannot present the real antioxidant activity because the higher concentration of sample does not always give the higher percentage of DPPH scavenging activity. The linear result will be seen in some concentration only. This condition can be occurred in extract or sample which contained more than one compound.

The extract consisted of many compounds, and not all of compounds have antioxidant activities. Some of them may act as an antagonist of antioxidant activities. The compounds will act as an antagonist of antioxidant activities if their minimum effective concentration has been reached. Therefore, this reason can explain why the extract with lower concentration can give higher activity than higher concentration.

## Antioxidant activity by ABTS assay

The previous study [11] represented that 50% ethanolic pulp extract of white dragon fruit (H. undatus) had the highest antioxidant activity by ferrous ion chelating activity, followed by fruit (peel and pulp) extract of red dragon fruit (H. polyrhizus), fruit (peel and pulp) extract of white dragon fruit (H. undatus), and pulp extract of red dragon fruit (H. polyrhizus). It was different with the present study which exposed that ethanolic peel extract of super red dragon fruit (H. costaricensis) gave the highest antioxidant activity by ABTS method with IC_{50} of ABTS scavenging activity (0.55 μg/ml) compared to the other parts extracts, meanwhile IC_{50} of ascorbic acid was 0.52 μg/ml. Hence, it can be concluded that antioxidant activity of ethanolic peel extract of super red dragon fruit was around one-third of antioxidant activity of ascorbic acid, by ABTS method.

### TPC

The TPC among the various extracts were demonstrated in term of gallic acid equivalent using the linear regression equation y=0.004x+0.055, R^2=0.997. The TPC in various extracts from different parts of super red dragon fruit gave different result in the range of 0.82-4.56 g GAE/100 g, PE2 extract (ethyl acetate flesh extract of super red dragon fruit) showed the highest TPC (4.56 g GAE/100 g) (Fig. 3).

The TPC might contribute to antioxidant activity [15,24]. Cinnamic acid had higher antioxidant capacity than phenylacetic acid and benzoic acid [25]. Flavonoids, tannins, and phenolic acids are included in phenolic groups. Ortho and para hydroxyl substitution have the stronger antioxidant capacity [26].

Research by Choo and Yong [11] stated that 50% ethanolic pulp extract of red dragon fruit (H. polyrhizus) had the highest TPC 24.22 mg GAE/100 g compared to its distilled water extract and methanolic extract (4.01 g GAE/100 g) and flesh extract (1.05 g GAE/100 g). Previous study [15] presented that ethanolic peels extract of super red dragon fruit (H. undatus) showed the highest TPC (36.12 mg GAE/100 g) compared to peels extract of red dragon fruit (H. polyrhizus) extract (28.16 mg GAE/100 g), pulps extract of red dragon fruit (19.72 mg GAE/100 g), and pulps extract of white dragon fruit (3.75 mg GAE/100 g). It was similar to the present study which presented that ethanolic peels extract of super red dragon fruit had the highest TPC (4.56 g GAE/100 g) compared to its stem extract (4.01 g GAE/100 g) and flesh extract (1.05 g GAE/100 g). Previous study revealed that ethanolic fruit extract of white dragon fruit (H. undatus) gave the highest TPC (179.85 mg GAE/I) compared to its distilled water extract and methanolic extract [21]. Research by De Mello et al. [10] presented that 80% acetone peel extract of white dragon fruit (moisture of 90.23 g/100 g) had TPC 40.68 mg GAE/100 g. The frozen filtrate of red dragon fruit (H. polyrhizus) pulp juice after precipitating with 96% ethanol, gave TPC 17.22 mg GAE/g [19] and seed extract of red dragon fruit (H. polyrhizus) had TPC 13.56 mg/g dry weight sample [9].

The previous study stated that betalain content in 80% acetone extract of white dragon fruit (H. undatus) peaked extract was 101.04 mg betanin equivalent/100 g [10].
TPC in ethyl acetate peel extract of super red dragon fruit (PEE2) 4.56 g GAE/100 g was higher than ethanolic peel extract of super red dragon fruit (PEE3) 1.87 g GAE/100 g, however IC\textsubscript{50} of DPPH of PEE2 (17.12 μg/ml) was similar to IC\textsubscript{50} DPPH of PEE3 (16.45 μg/ml). The similar result in ABTS assay, PEE3 which had lower TPC than PEE2, gave the lower IC\textsubscript{50} of ABTS (1.55 μg/ml) than PEE2 (8.01 μg/ml). It can be supposed that many phenolic compounds in PEE2 had low antioxidant activities and many phenolic compounds in PEE3 with high antioxidant activities.

**TFC**

The TFC among three parts extracts of super red dragon fruit were exposed in term of quercetin equivalent using the linear regression equation y=0.006x−0.098, R\textsuperscript{2}=0.996. TPC in parts super red dragon fruit extracts were varied from 0.70 to 12.63 g QE/100 g. The highest TFC (12.63 g QE/100 g) was presented by ethyl acetate peels extract of super red dragon fruit (PEE2) (Fig. 4).

The frozen filtrate of pulp juice of red dragon fruit and after precipitating with 96% ethanol showed TFC 0.23 g catechin/100 g [19]. It was similar to the present study which expressed that TFC in ethanolic flesh extract of super red dragon fruit (0.52 g GAE/100 g) and lower than its stem extract (2.66 g GAE/100 g). Research by Adnan et al. [9] exhibited that there were two major compounds of flavonoid group were catechin 3.60 mg/g and quercetin 1.31 mg/g dry weight seed extract of red dragon fruit (H. polyrhizus). Flavonoid may have antioxidant effect as hydrogen donating compound, metal chelating ion, single oxygen transfer, and singlet oxygen quencher [27]. Hydrogen donating and metal chelating is related to ortho di-OH structure in ring B, C-2-C-3 double bond and oxo group at C-4 [27]. Flavonoid had higher antioxidant capacity than phenolic acid [25]. Flavonoid would give higher antioxidant capacity if flavonoid had ortho di-OH in C3'-C4',-OH in C3, oxo function in C4, double bond at C2 and C3. The di-OH with ortho position in C3'-C4' had the highest influence to the antioxidant capacity of flavonoid. The flavonoid glycoside would give lower antioxidant capacity than flavonoid aglycone [25]. The previous research succeeded to identify betalain as pigment in red dragon fruit (H. polyrhizus) pulps [19]. Purple color in super red dragon fruit can be correlated with betalain or anthocyanin in super red dragon fruit. Anthocyanin is one of the phenolic compounds in the plant. Betalain is not belong to anthocyanin group because betalain contains nitrogen whereas anthocyanin does not. Betalain may be contributed in antioxidant activity. Based on betalain structure, it has high antioxidant activity, because it does not have ortho di-OH which gives influence on antioxidant activity. Anthocyanin will give influence on high antioxidant activity if the anthocyanin has ortho di-OH in ring B.

In Fig 4 it can be seen that the ethyl acetate peel extract of super red dragon fruit (PEE2) had TPC (12.63 g QE/100 g) higher than ethyl acetate flesh extract of super red dragon fruit (FLE2) 4.78 g QE/100 g, but IC\textsubscript{50} DPPH of FLE2 (2.69 μg/ml) lower than IC\textsubscript{50} DPPH of PEE2 (17.12 μg/ml). In TPC determination, any compound which has ortho di-OH at benzene ring can react with aluminum (III) chloride and form a complex. The same reaction can occur in any compound which has ortho di-OH-OCH form a complex. The same reaction can occur in any compound which has ortho di-OH in benzene ring. Ortho di-OH has the highest influence in antioxidant activity. Based on the result, in can predicted that many flavonoid compounds in FLE2 were flavonoid which had ortho di-OH in ring B which had highest antioxidant activity or double bond in C2-C3, -OH in C3 an oxo function in C4 which also contribute to the high antioxidant activity. The flavonoid compound might be quercetin which has ortho di-OH in ring B and stated as the major flavonoid in red dragon fruit by Adnan et al. [2011].

**Pearson's correlation coefficient**

Coefficient of Pearson correlation was significantly negative if -0.61 ≤ r ≤ -0.97 and significantly positive if 0.61 ≤ r ≤ 0.97 [6]. The lowest IC\textsubscript{50} of DPPH and IC\textsubscript{50} of ABTS scavenging activities will give the highest antioxidant activity. Increasing in TPC and TFC can influence increasing in antioxidant activities, which was exposed by lower IC\textsubscript{50} of DPPH and IC\textsubscript{50} of ABTS scavenging activities. Therefore, the good correlation between TPC and TFC with IC\textsubscript{50} of DPPH or IC\textsubscript{50} of ABTS was significantly negative correlation [28].

TPC in flesh extract of super red dragon fruit had a significant and negative correlation with their IC\textsubscript{50} of ABTS scavenging activities.

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**Table 2: Pearson's correlation coefficient of total phenolic, flavonoid content in parts extracts of super red dragon fruit with their IC\textsubscript{50} of DPPH and IC\textsubscript{50} of ABTS scavenging activities**

<table>
<thead>
<tr>
<th>Antioxidant parameter</th>
<th>Pearson's correlation coefficient (r)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>TPC</td>
</tr>
<tr>
<td>IC\textsubscript{50} DPPH STE</td>
<td>0.96**</td>
</tr>
<tr>
<td>IC\textsubscript{50} DPPH PEE</td>
<td>0.685*</td>
</tr>
<tr>
<td>IC\textsubscript{50} DPPH FLE</td>
<td>0.649**</td>
</tr>
<tr>
<td>IC\textsubscript{50} ABTS STE</td>
<td>-0.808*</td>
</tr>
<tr>
<td>IC\textsubscript{50} ABTS PEE</td>
<td>-0.606**</td>
</tr>
<tr>
<td>IC\textsubscript{50} ABTS FLE</td>
<td>0.613*</td>
</tr>
</tbody>
</table>

IC\textsubscript{50} DPPH, IC\textsubscript{50} DPPH scavenging activity, IC\textsubscript{50} ABTS, IC\textsubscript{50} ABTS scavenging activity. STE: Stem of super red dragon fruit, PEE: Peel of super red dragon fruit, FLE: Flesh of super red dragon fruit. TPC: Total phenolic content. TFC: Total flavonoid content. ns: Not significant, *significant at p<0.05, **significant at p<0.01. IC\textsubscript{50} Inhibitory concentration 50%, DPPH: 2,2-diphenyl-1-picrylhydrazyl, ABTS: 2,2'-azino-bis 3-ethylbenzthiazoline-6-sulfonic acid
l=0.614, p<0.05). In Figs. 1 and 3 can be seen that TPC in n-hexane flesh extract, ethanolic flesh extract and ethyl acetate flesh extract of super red dragon fruit (0.82, 1.05, and 3.70 g GAE/100 g, respectively) gave TPC of ABTS 234.13, 27.69, and 6.25 μg/mL, respectively), which higher TPC gave lower IC50 of ABTS. Based on the result it can be supposed that phenolic compounds were the contributors major in antioxidant activity of flesh extract of super red dragon fruit, by ABTS method.

The previous result by Nurliyana et al. [12] figured that TP had no correlation with their IC50 of DPPH. It was similar to the present study which revealed that TPC and TSC of stem, peels and flesh extract of super red dragon fruit were not significant correlation with their IC50 of DPPH (Table 2). IC50 of flesh extract of super red dragon fruit extract had a significantly positive correlation with their IC50 of ABTS (r=0.988, p=0.01). It can be concluded that DPPH and ABTS assays gave linear result in antioxidant activity of flesh extract of super red dragon fruit.

CONCLUSION
Different results can be shown by various methods, hence for evaluating antioxidant activities should be carried out by different methods in parallel. All different parts extracts of super red dragon fruit H. costaricencis (except n-hexane flesh extract) can be categorized as very strong antioxidant using DPPH assay. Phenolic compounds in flesh extracts of super red dragon fruit were the major contributor to their antioxidant activity by ABTS method. DPPH and ABTS methods gave linear results in antioxidant activity of flesh extract of super red dragon fruit. Stem, peel, and FLE (H. costaricencis) have many benefits to prevent oxidative stress. The waste products of super red dragon fruit (peel and stem) were potential as sources of natural antioxidants for further exploitation.

REFERENCES