



Research Article

ISSN : 0975-7384
CODEN(USA) : JCPRC5

Antioxidant capacities, phenolic, flavonoid and carotenoid content of various polarities extracts from three organs of *Sechium edule* (Jacq.) Swartz

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ABSTRACT

The objectives of this research were to study antioxidant activities from various fruit extracts of three organs of *S. edule* using two methods of antioxidant assays which were DPPH (2,2-diphenyl-1-picrylhydrazyl) and CUPRAC (Cupric ion Reducing Antioxidant Capacity) and correlation of their total flavonoid, phenolic and carotenoid content with IC_{50} of DPPH antioxidant activities and EC_{50} of CUPRAC capacities. Extraction was performed by reflux apparatus using different polarity solvents. The extracts were evaporated using rotary evaporator. Antioxidant capacities were tested using DPPH and CUPRAC assays. Determination of total phenolic, flavonoid and carotenoid content was performed by spectrophotometer UV-visible and their correlation with IC_{50} of DPPH scavenging capacities and EC_{50} of CUPRAC capacities were analyzed by Pearson's method. Ethyl acetate pedicel extract of *S. edule* (PD2) had the lowest IC_{50} of DPPH scavenging activity 1.3 $\mu\text{g/ml}$, while ethyl acetate fruit extract of *S. edule* (FR2) had the lowest EC_{50} of CUPRAC capacity 147 $\mu\text{g/ml}$. Ethyl acetate fruit extract of *S. edule* (FR2) had the highest total phenolic content (3.21 g GAE/100 g), ethyl acetate leaves extract of *S. edule* (LV2) had the highest total flavonoid content (11.64 g QE/100 g) and the highest total carotenoid content (12.73 g BE/100 g). There was negatively high correlation between total phenolic content in leaves extracts of *S. edule* with their IC_{50} of DPPH. The negative and high correlation between total phenolic, flavonoid and carotenoid content in fruit and pedicel extracts with their EC_{50} of CUPRAC capacities. The IC_{50} of DPPH scavenging activities of three organs of *S. edule* had no linear result with their EC_{50} of CUPRAC capacities.

Keywords: Antioxidant, DPPH, CUPRAC, three organs, *S. edule*, phenolic, flavonoid, carotenoid

INTRODUCTION

Oxidative stress was the important factor in many degenerative diseases. Antioxidant has potency to inhibit oxidative stress. Phenolic compounds are commonly found in plants, and they have revealed to have multiple biological effects, including antioxidant activity [1-3]. Many studies had reported that phenolic content in plants could be correlated to their antioxidant activities. Plants contained phenolic and polyphenol compounds can act as antioxidant [3-6].

Some of antioxidant methods such as DPPH (2,2-diphenyl-1-picrylhydrazyl), ABTS (2,2'-azinobis (3-ethylbenzothiazoline-6-sulfonic acid), FRAP (Ferric Reducing Antioxidant Power) and CUPRAC (Cupric ion Reducing Antioxidant Capacity) were widely used to predict antioxidant capacity of fresh fruits, beverages, and food [3,7-9]. Previous studies by Thaipong [3], Apak [8], Muller [10], Fidrianny [11] revealed that DPPH and CUPRAC methods could be used to measure antioxidant activity in many plants extracts. The previous research [12-14] exhibited antioxidant capacities of some plants including *S. edule*.

The objectives of this research were to study antioxidant activities of various extracts (n-hexane, ethyl acetate and ethanol) from three organs (leaves, fruit, pedicel) of chayote (*Sechium edule* (Jacq.) Swartz) using DPPH and CUPRAC assays, and correlations of antioxidant activities with their total phenolic, and carotenoid content.

EXPERIMENTAL SECTION

Materials: DPPH (2,2-diphenyl-1-picrylhydrazyl), neocuproine, gallic acid, quercetin, beta carotene purchased from Sigma-Aldrich (MO, USA), cupric chloride, organs of chayote. All other reagents were analytical grades.

Preparation of sample: Three organs from *Sechium edule* that were: leaves namely as LV, fruit as FR and pedicel as PD were collected from Boyolali, Center of Java, were thoroughly washed with tap water, sorted while wet, cut, dried, and grinded into powder.

Extraction: Three hundred grams of powdered samples were extracted by reflux apparatus using increasing polarity of solvents. The extraction using n-hexane was repeated three times. The remaining residue was then extracted three times using ethyl acetate. Finally the remaining residue was extracted three times using ethanol. So totally there were nine extracts: three n-hexane extracts (LV1, FR1 and PD1), three ethyl acetate extracts (LV2, FR2, and PD2) and three ethanolic extracts (LV3, FR3 and PD3).

IC₅₀ of DPPH scavenging activity: Preparation of DPPH solution was adopted from Blois [15] with minor modification. Various concentration of each extract were pipetted into DPPH solution 50 µg/ml (1:1) to initiate the reaction for obtaining a calibration curve. After 30 minutes incubation, the absorbance was read at wavelength 515 nm by using spectrophotometer UV-Vis Hewlett Packard 8435. Methanol was used as a blank. DPPH solution 50 µg/ml was used as control. Ascorbic acid was used as standard. Analysis was done in triplicate for standard and each extract. Antioxidant activity of each extract was determined based on the reduction of DPPH absorbance by calculating percentage of antioxidant activity [16]. IC₅₀ of DPPH scavenging activity of each extract can be calculated using its calibration curve.

EC₅₀ of CUPRAC capacity: Preparation of CUPRAC solution was adopted from Apak [8]. The CUPRAC solution was prepared in ammonium acetate buffer pH 7. Various concentration of each extract were pipetted into CUPRAC solution 50 µg/ml (1:1) to initiate the reaction for obtaining a calibration curve. After 30 minutes incubation, the absorbance was read at wavelength 450 nm by using spectrophotometer UV-Vis Hewlett Packard 8435. Ammonium acetate buffer was used as a blank. CUPRAC solution 50 µg/ml was used as control. Ascorbic acid was used as standard. Analysis was done in triplicate for standard and each extract. Antioxidant capacity of each extract was determined based on increasing in Cu (I)-neocuproine absorbance by calculating percentage of antioxidant capacity [8]. EC₅₀ of CUPRAC capacity of each extract can be calculated using its calibration curve.

Determination of total phenolic content (TPC): Total phenolic content were measured using the modified Folin-Ciocalteu method adapted from Pourmorad [2]. The absorbance was read at wavelength 765 nm. Analysis was done in triplicate for each extract. Standard solution of gallic acid 40-165 µg/ml were used to obtain a standard curve. The total phenolic content was reported as percentage of total gallic acid equivalent per 100 g extract (g GAE/100 g).

Determination of total flavonoid content (TFC): Total flavonoid content was measured using adapted method from Chang *et al.* [17]. The absorbance was read at wavelength 415 nm. Analysis was done in triplicate for each extract. Standard solution of quercetin 36-120 µg/ml were used to obtain a standard curve. The total flavonoid content was reported as percentage of total quercetin equivalent per 100 g extract (g QE/100 g).

Determination of total carotenoid content (TCC): Total carotenoid content was measured using the modified carotene method adapted from Thaipong *et al* [3]. Each extract were diluted in n-hexane. The absorbance was read at wavelength 470 nm. Analysis was done in triplicate for each extract. Standard solution of beta carotene 15-55 µg/ml were used to obtain a standard curve. The total carotenoid content was reported as percentage of total beta carotene equivalent per 100 g extract (g BE/100 g).

Statistical Analysis: Analysis of each sample was performed in triplicate. All results presented were the means (\pm SD) of at least three independent experiments. Statistical analysis (ANOVA with a statistical significance level set at $p < 0.05$ and post-hoc Tukey procedure) was conducted out with SPSS 16.0 for Windows. Correlations between the total phenolic, flavonoid, carotenoid content and antioxidant activities were made using the Pearson's method ($p < 0.01$).

RESULTS AND DISCUSSION

The previous research [5,12-14] exposed that *S. edule* had antioxidant capacity. There were no study regarding antioxidant capacity of various polarities extracts (which were n-hexane, ethyl acetate and ethanol) of three organs from *S. edule* using DPPH and CUPRAC methods.

IC₅₀ of DPPH scavenging activity and EC₅₀ of CUPRAC capacity: DPPH is free radicals which dissolve in methanol or ethanol, and it has characteristic absorption at wavelength 515-520 nm. Colors of DPPH would be changed when the free radicals were scavenged by antioxidant [8,18]. Reagent of CUPRAC is cupric chloride which combined with neocuproine in ammonium acetate buffer pH 7. Sample will act as antioxidant, if it can reduces Cu (II) to Cu (I), at the same time it will be oxidized. Complex Cu (I) – neocuproine gives yellow color and show characteristic absorption at wavelength 450 nm. Yellow color intensity will be depended on amount of Cu (II) that is reduced to Cu (I). Redox potential of sample is important factor in CUPRAC assay. Sample will be oxidized if it had reduction potential lower than reduction potential of Cu (II)/Cu (I) 0.46 V.

The IC₅₀ of DPPH scavenging activities and EC₅₀ of CUPRAC capacities in various organs extracts from *S. edule* using DPPH and CUPRAC assays were shown in Fig 1 and Fig 2. The half minimum inhibitory concentration (IC₅₀) of DPPH scavenging activities and EC₅₀ of CUPRAC capacities compared to IC₅₀ ascorbic acid standard and EC₅₀ ascorbic acid standard.

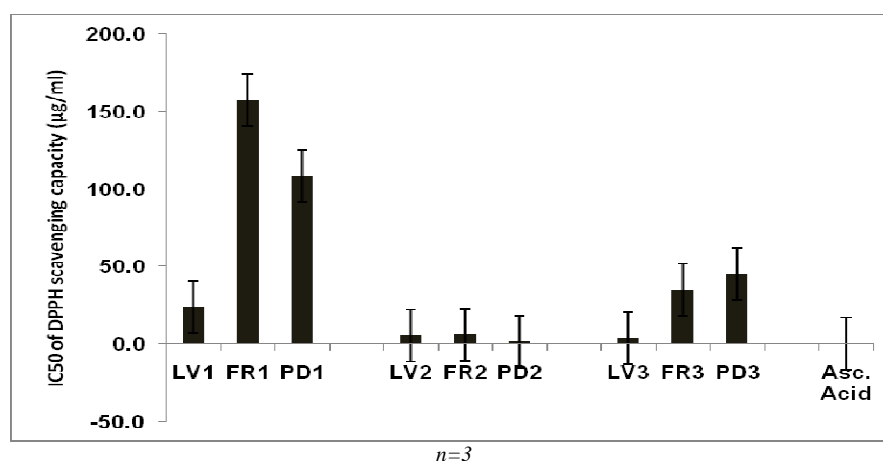


Fig 1: IC₅₀ of DPPH scavenging capacities in various organs extracts from *S.edule*

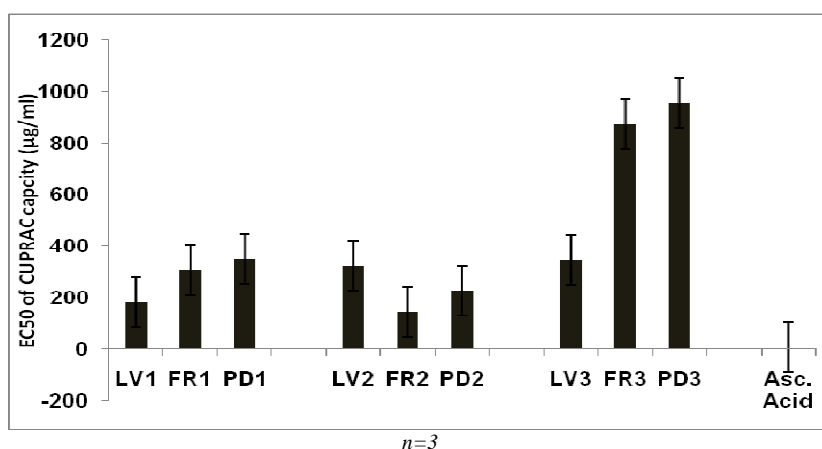


Fig 2: EC₅₀ of CUPRAC capacities in various organs extracts from *S.edule*

IC₅₀ of DPPH scavenging capacity is the concentration of sample or standard that can inhibit 50% of DPPH scavenging capacity, while EC₅₀ of CUPRAC capacity is the concentration of sample or standard that can exhibit 50% of CUPRAC capacity. The lowest IC₅₀ or EC₅₀ means had the highest antioxidant capacity. The IC₅₀ or EC₅₀ were used to classify antioxidant activity of a sample and compare to standard. Sample that has IC₅₀ or EC₅₀ less than 50 µg/ml is a very strong antioxidant, 50-100 µg/ml is a strong antioxidant, 101-150 µg/ml is a medium antioxidant, while IC₅₀ greater than 150 µg/ml is a weak antioxidant [15].

The result of the present study exposed that IC₅₀ of DPPH scavenging activities of various organs extracts from *S. edule* ranged from 1.3 – 157.3 µg/ml. Ethyl acetate pedicel extract of *S. edule* (PD2) had the lowest IC₅₀ of DPPH radical scavenging activity 1.3 µg/ml, followed by ethanolic leaves extract of *S. edule* (LV3) 3.8 µg/ml, and ethyl acetate leaves extract of *S. edule* (LV2) 5.1 µg/ml, while ascorbic acid standard expressed IC₅₀ of DPPH scavenging activity 0.1 µg/ml. All of ethyl acetate and ethanolic leaves, fruit and pedicel extracts of *S. edule* had IC₅₀ of DPPH scavenging capacity less than 50 µg/ml, so they could be classified as very strong antioxidant. In the previous study [14] exposed that ethanolic leaves extract of *Luffa acutangula* (LA3) had the lowest IC₅₀ DPPH scavenging activity

73 µg/ml compared to ethanolic leaves extract of *Cucumis sativus*, *S. edule*, *Momordica charantia* and *Cucurbita moschata*. Ethanolic leaves extract of *S. edule* from Garut- West Java had IC₅₀ of DPPH scavenging capacity 94 µg/ml which was strong antioxidant, it was contrast with the present study which showed that the ethanolic leaves extract of *S. edule* from Boyolali –Center of Java gave IC₅₀ of DPPH 3.8 µg/ml which was categorized as very strong antioxidant. Research by Chao [13] demonstrated that IC₅₀ of DPPH of acidified methanol leaves extract of *S. edule* yellow 1503 µg/ml was lower than *S. edule* green 1801 µg/ml. Study by Souri [19] regarding the other plant of Cucurbitaceae family revealed that IC₅₀ of DPPH scavenging capacity of methanolic seed extract of *Cucumis sativus* was 1.25 µg/ml, it was different with the previous study [14] which showed that IC₅₀ of DPPH scavenging capacity of ethanolic leaves extract of *C. sativus* 416 µg/ml. Ethanolic leaves extract of *Momordica charantia* had IC₅₀ of DPPH scavenging capacity was 4.885 mg/ml [14], while study by Patel *et al.* [1] exposed that IC₅₀ of DPPH scavenging capacity of alcohol fruits extract and water fruits extract of *M. charantia* were 120 µg/ml and 182 µg/ml, respectively. Water fruits extract of cultivar N had the lowest IC₅₀ of DPPH scavenging capacity (181µg/ml) among in 16 cultivars of *M. charantia* (A-P) [20]. The previous research [14] exposed that ethyl acetate leaves extract of *S. edule* had the lowest EC₅₀ of FRAP capacity 759 µg/ml, while in the present study showed that its EC₅₀ of CUPRAC capacity was 347 µg/ml. Study by Ordonez [12] stated that ethanolic leaves and stem extract of *S. edule* by maceration method gave higher percentage of DPPH scavenging activities (85 % and 65 %, respectively) than using decoction method (80 % and 30 %, respectively).

TPC in various organs extracts from *S. edule*: TPC among the various organs extracts expressed in term of gallic acid equivalent using the standard curve equation $y = 0.004 x + 0.0025$, $R^2 = 0.998$. TPC in various organs extracts from *S. edule* exposed different result in the range of 0.88 – 3.21 g GAE/100 g. Ethyl acetate fruit extract of *S. edule* (FR2) had the highest TPC (3.21 g GAE/100 g) (Fig 3).

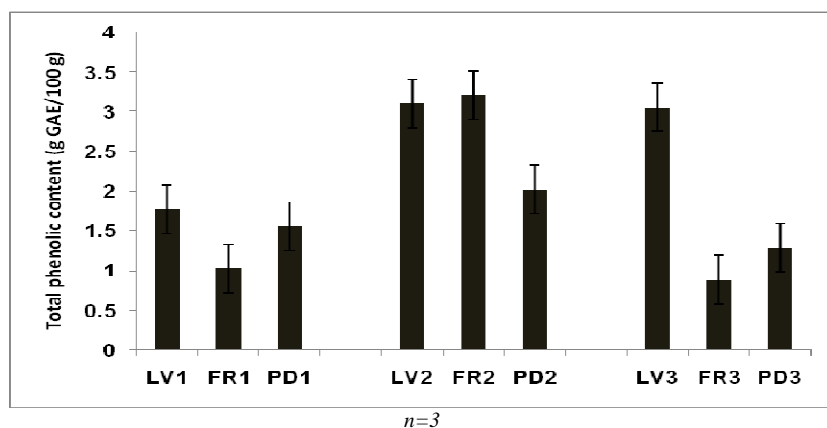


Fig 3: Total phenolic content in various organs extracts of *S. edule*

The total phenolic content can be contributed in antioxidant capacity [1-2,12-13]. Phenolic acid might contributed in antioxidant capacity. Chao [13] revealed that polyphenolic content in acidified methanol leaves extract of *S. edule* yellow was 0.063 g GAE/100 g and the green one 0.262 g GAE/100 g. Fidrianny [14] exposed that TPC in ethanolic leaves extract of *S. edule* from Garut 1.79 g GAE/100 g which was lower than ethanolic leaves extract of *L. acutangula* 2.88 g GAE/100 g, while in the present study TPC in ethanolic leaves extract of *S. edule* from Boyolali was 3.05 g GAE/100 g. Previous study [12] revealed that ethanolic leaves extract of *S. edule* by decoction and maceration method had TPC 0.91 and 1.16 mg GAE/ml, respectively, while in the current study expressed that ethanolic leaves extract of *S. edule* by reflux method had TPC 3.05 g GAE/100 g. TPC in ethanolic stem extract of *S. edule* by maceration method 0.25 mg/ml and similar with decoction method 0.23 mg/ml [12]. The present study showed that n-hexane, ethyl acetate and ethanolic pedicel extracts of *S. edule* were 1.56, 2.02 and 1.29 g GAE/100 g, respectively.

TFC in various organs extracts from *S. edule*: The TFC among the various organs extracts expressed in term of quercetin equivalent using the standard curve equation $y = 0.006 x - 0.019$, $R^2 = 0.998$. TFC in various organs extracts from *S. edule* showed different results in the range of 0.31 - 11.64 g QE/100 g (Fig 4). Ethyl acetate leaves extract of *S. edule* (LV2) had the highest total flavonoid content (11.64 g QE/100 g) and ethanolic pedicel extract of *S. edule* (PD3) had the lowest (0.31 g QE/100 g).

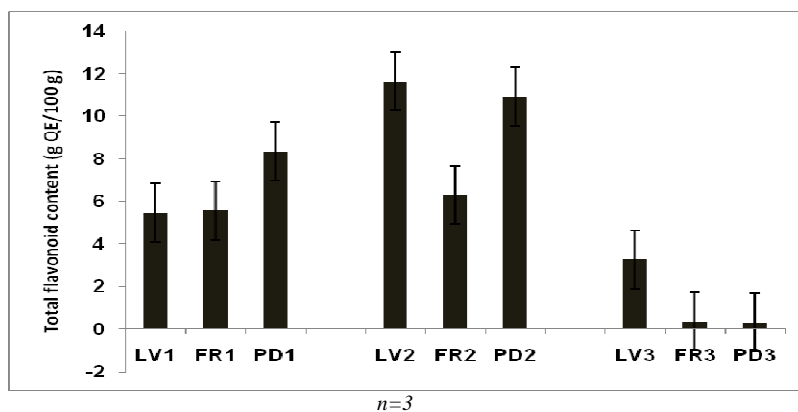


Fig 4: Total flavonoid content in various organs extracts of *S. edule*

In the present research exhibited that TFC in ethanolic leaves extract of *S. edule* was 3.26 g QE/100 g, while in previous study [13] stated that TFC in acidified methanol leaves extract of *S. edule* green 0.42 g QE/100 g and yellow 0.18 g QE/100 g. Study by Ordonez [12] exposed that TFC in ethanolic leaves extract of *S. edule* by decoction method (0.20 mg/ml) was lower than ethanolic leaves extract by maceration method (0.65 mg QE/ml). TFC in n-hexane, ethyl acetate and ethanolic pedicel extract of *S. edule* by reflux method were 8.34, 10.93, 0.31 g QE/100 g, while previous research [12] demonstrated that ethanolic stem extract of *S. edule* were 18 mg QE/ml by maceration method and 0.07 mg QE/ml using decoction method. The previous study [14] reported that TFC in ethanolic leaves extract of *S. edule* 5.47 g QE/100 g which was higher than ethanolic leaves extract of *C. sativus*, *L. acutangula*, *M. charantia* 1.71, 2.30 and 0.77 g QE/100 g, respectively.

TCC in various organs extracts from *S. edule*: The TCC among the various organs extracts expressed in term of beta carotene equivalent using the standard curve equation $y = 0.015x + 0.002$, $R^2 = 0.9999$. TCC in various organs extracts from *S. edule* showed different result ranged from 0.02 to 12.73 g BE/100 g (Fig 5). Ethyl acetate leaves extract of *S. edule* (LV2) had the highest TCC (12.73 g BE/100 g), while ethanolic fruit extract of *S. edule* (FR3) had the lowest carotenoid content (0.02 g BE/100 g). In the previous study [14] exposed that ethanolic leaves extract of *S. edule* had the highest TCC (0.6 g BE/100 g) compared to ethanolic leaves extract of *C. sativus*, *L. acutangula* and *M. charantia* (0.04, 0.09, 0.11 g BE/100 g), while the present study stated that TFC in ethanolic extract of leaves, fruit and pedicel of *S. edule* were 0.34, 0.02 and 0.03 g BE/100 g, respectively.

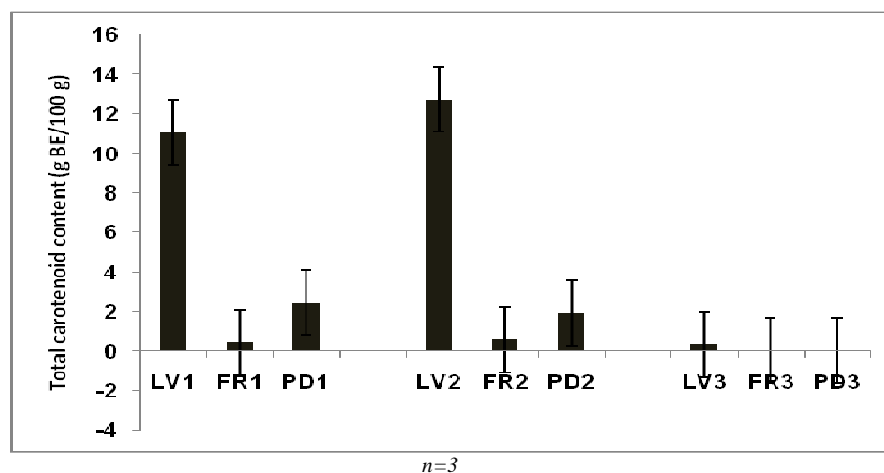


Fig 5: Total carotenoid content in various organs extracts of *S. edule*

Correlations between total phenolic, flavonoid, carotenoid content with DPPH scavenging activities, and CUPRAC capacities in various organs extracts of *S. edule*: Pearson's correlation coefficient was positively high if $0.61 \leq r \leq 0.97$ [3] and negatively high if $-0.61 \leq r \leq -0.97$. Sample which had the lowest IC_{50} of DPPH scavenging capacity or EC_{50} of CUPRAC capacity gave the highest antioxidant activity. So the highly and negative correlation between TPC, TFC and TCC with IC_{50} DPPH or EC_{50} CUPRAC expressed the good correlation.

Table 1. Pearson's correlation coefficient of total phenolic, flavonoid, carotenoid of organs extracts from *S. edule* and IC₅₀ of DPPH scavenging capacities, EC₅₀ of CUPRAC capacities

	TPC	TFC	TCC	EC ₅₀ CUPRAC LE	EC ₅₀ CUPRAC FR	EC ₅₀ CUPRAC PD
IC ₅₀ DPPH LE	-0.966**	-0.204 ^{ns}	0.439 ^{ns}	-0.993**		
IC ₅₀ DPPH FR	-0.591 ^{ns}	0.227 ^{ns}	0.094 ^{ns}		-0.132 ^{ns}	
IC ₅₀ DPPH PD	-0.519 ^{ns}	-0.133 ^{ns}	0.311 ^{ns}			0.056 ^{ns}
EC ₅₀ CUPRAC LE	0.938**	0.148 ^{ns}	-0.496 ^{ns}			
EC ₅₀ CUPRAC FR	-0.705*	-0.994**	-0.997**			
EC ₅₀ CUPRAC PD	-0.831**	-0.997**	-0.931**			

Note: DPPH = DPPH scavenging capacity, CUPRAC = CUPRAC capacity, TPC = total phenolic content, TFC = total flavonoid content, TCC = total carotenoid content, LE = sample LE, FR = sample FR, PD = sample PD, ns = not significant, * = significant at $p < 0.05$, ** = significant at $p < 0.01$

The highest and negative between TPC and IC₅₀ of DPPH scavenging capacity ($r = -0.966$, $p < 0.01$) was given by leaves extract of *S. edule*. The highest and negative correlation between TPC and EC₅₀ of CUPRAC capacity ($r = -0.831$, $p < 0.01$) was given by pedicel extract of *S. edule* (Table 1). Based on this data it could be supposed that antioxidant capacities in leaves extracts with DPPH method might be estimated indirectly by determining their TPC. EC₅₀ of CUPRAC capacities of fruit and pedicel extracts of *S. edule* had negatively and high correlation with TPC ($r = -0.705$, $p < 0.05$, $r = -0.831$, $p < 0.01$, respectively), TFC ($r = -0.994$, $r = -0.997$, $p < 0.01$, respectively) and TCC ($r = -0.997$, $r = -0.931$, $p < 0.01$, respectively). It means that increasing in TPC and or TFC and or TCC in fruit and pedicel extracts of *S. edule* would increase antioxidant activity by CUPRAC method. Based on this data it can be exposed that antioxidant capacity of leaves extract of *S. edule* by DPPH can be predicted indirectly by their TPC and antioxidant activity of fruit and pedicel extracts of *S. edule* by CUPRAC assay might be predicted indirectly by measuring their TFC and or TCC. In previous study [14] reported that TPC in leaves extract of *S. edule* and *L. acutangula* had positively high correlation with their percentage of DPPH scavenging capacities ($r = 0.875$, $r = 0.888$, $p < 0.01$, respectively).

Flavonoid, phenolic acid, tannins were included in phenolic compounds. Flavonoid which have OH in A ring and/or B ring are phenolic compounds. Ortho di-OH in phenolic compound would give higher antioxidant capacity than metha and para- di-OH position [21]. Phenolic compound which have -OCH₃ and -OH in ortho or para position have high antioxidant activity [8], but phenolic acid had lower antioxidant capacity than flavonoid [22].

Position of hydroxyl group in C-3'-C-4', OH in C-3, oxo function in C-4, double bond at C-2 and C-3 would give higher antioxidant capacity in flavonoid. Ortho position of hydroxyl group in C-3'-C-4' had the highest influence in antioxidant capacity of flavonoid. The flavonoid glycosides would give lower antioxidant capacity than flavonoid aglycone [22].

It could be seen in Figure 3, TPC in ethyl acetate leaves extract of *S. edule* (LV2) 3.11 g GAE/100 g was similar with TPC in ethanolic leaves extract (LV3) 3.05 g GAE/100 g, but IC₅₀ of DPPH scavenging capacity of LV2 (5.1 µg/ml) was higher than LV3 (3.8 µg/ml). It means LV3 had higher antioxidant capacity than LV2. Based on the data it can be supposed that many phenolic compounds in LV3 had high antioxidant capacity and many phenolic compound in LV2 had low antioxidant capacity.

TFC in ethyl acetate leaves extract of *S. edule* (LV2) 11.64 g QE/100 g was similar with TFC in ethyl acetate pedicel extract (PD2) 10.93 g QE/100 g, but IC₅₀ of DPPH scavenging activity and EC₅₀ of CUPRAC capacity of PD2 (1.3 µg/ml and 227 µg/ml, respectively) was lower than LV2 (5.1 µg/ml and 326 µg/ml, respectively). Based on the data above, it can be predicted that many flavonoid in PD2 had higher antioxidant capacity which had OH in C-3'-C-4', OH in C-3, oxo function in C-4, double bond at C-2 and C-3, while in LV2 many flavonoid had OH C5, C7, or C3' only, or C4' only, or C3 only without oxo function in C4, that had no and low antioxidant capacities. Regarding their CUPRAC capacity it can be supposed that many flavonoid in PD2 which had potential redox lower than potential redox of Cu (II)/Cu (I) 0.46 V, so it can be oxidized and at the same time it could reduce Cu (II) to Cu (I). Then Cu (I) formed complex with neocuproine and gave yellow color. CUPRAC method can detect lypophilic and hydrophilic antioxidant because of it can soluble in water and organic solvent [8], but ability of the sample to react with CUPRAC reagent depending on its potential redox.

Carotenoid had antioxidant capacity by scavenging free radical. More double bonds in carotenoid would give higher free radical scavenging capacity [23]. Carotenoid which contained more than 7 double bonds gave greater free radical scavenging activity than 7 double bonds [24]. Decreasing in lypophilicity of carotenoid would decrease free radical scavenging capacity [25]. Beta carotene was used as standard because it had conjugation double bonds due to its ability to scavenge free radicals [10,26]. TCC in n-hexane pedicel extract of *S. edule* (PD1) 2.45 g BE/100 g was greater than TCC in ethanolic leaves extract (LV3) 0.34 g BE/100 g, but IC₅₀ of DPPH scavenging activity of LV3 (3.8 µg/ml) which was categorized as very strong antioxidant compared to IC₅₀ of DPPH of PD1 (108 µg/ml)

medium antioxidant. Based on this data it can be predicted that many carotenoid in LV3 consisted of more than 7 double bonds and only a little carotenoid with more than 7 double bonds in PD1. In CUPRAC method it could be seen EC₅₀ of CUPRAC capacity of LV3 was similar with PD1. It might be many carotenoid in LV3 which had potential redox lower than 0.46 V and only a little amount in PD1.

The DPPH and CUPRAC methods had the different mechanism reaction. Mechanism of DPPH was electron transfer assay [27] and CUPRAC was redox assay [8], and the results of the two methods not always linear. The Pearson's correlation coefficient demonstrated that IC₅₀ of DPPH scavenging activities of leaves, fruit and pedicel extracts of *S. edule* had no correlation with their EC₅₀ of CUPRAC capacity. DPPH and CUPRAC assays gave no linear result for leaves, fruit and pedicel extracts of *S. edule*.

CONCLUSION

Different results could be given by different antioxidant methods. Variety of methods must be used in parallel to assess the antioxidant capacity of sample. All of ethanolic and ethyl acetate leaves, fruit and pedicel extracts of *S. edule* had IC₅₀ of DPPH scavenging activities less than 50 µg/ml that means were very strong antioxidant. There were negatively and high correlation between TPC in leaves extract with its IC₅₀ of DPPH scavenging activities. Phenolic compounds were the major contributor in antioxidant capacity in leaves extracts of *S. edule*. The negative and high correlation between TPC, TFC and TCC in fruit and pedicel extracts with their EC₅₀ of CUPRAC capacities. Phenolic and or flavonoid and or carotenoid compounds were the main contributor in antioxidant capacities of fruit and pedicel of *S. edule* using CUPRAC method. Antioxidant capacities of leaves, fruit and pedicel extracts of *S. edule* by DPPH method gave no linear result with CUPRAC method. Leaves, fruit and pedicel extracts of *S. edule* may be exploited its beneficial as sources of natural antioxidant .

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