© 2018 Journal of Pharmacy & Pharmacognosy Research, 6 (6), 448-457, 2018
ISSN 0719-4250
http://jppres.com/jppres

*Original Article | Artículo Original*

# Phytochemical characterization and antioxidant profile of *Sechium edule* (Jacq) Swartz (*Cucurbitaceae*) varieties grown in Costa Rica

[Caracterización fitoquímica y perfil antioxidante de las variedades de *Sechium edule* (Jacq) Swartz (*Cucurbitaceae*) cultivadas en Costa Rica]

**Jonathan Parra[1*], Priscilla Hernández[1], Fernando Ocampo-Maroto[2], Víctor Álvarez-Valverde[2], Yendry Carvajal-Miranda[2], Gerardo Rodríguez-Rodríguez[2], Cristina Herrera[1]**

[1]Instituto de Investigaciones Farmacéuticas (INIFAR), Facultad de Farmacia, Universidad de Costa Rica, Sede Rodrigo Facio, San Pedro, San José, 11501-2060. Costa Rica.
[2]Laboratorio de Fitoquímica, Escuela de Química, Universidad Nacional de Costa Rica, Campus Omar Dengo, Heredia, 86-3000. Costa Rica.
*E-mail: jonathan.parra@ucr.ac.cr

**Abstract**

*Context*: The *Sechium edule* fruit is grown and consumed because of their nutritional values in different countries of Latin America. Costa Rica is one of the most important producers and exporters. It is consumed as an antioxidant because its flavonoid content suggests such activity; however, the selection of varieties to crop does not involve the phytochemical profile of them.

*Aims*: To characterize the phytochemical and antioxidant profile of the *S. edule* varieties grown in Costa Rica.

*Methods*: The *in vitro* radical scavenging activity of different *S. edule* extracts were measured using DPPH and ORAC assays, as well as their inhibition of lipid peroxidation in rat liver and their redox potential by cyclic voltammetry. Flavonoids glycosides were isolated by HPLC and their structures were determined by NMR.

*Results*: The *S. edule* varieties grown in Costa Rica showed a good radical scavenging activity and inhibition of lipid peroxidation; there were significant differences between varieties. In addition, the redox potential determined by electrochemical methods suggested the presence of flavonoids, which was confirmed by the isolation of apigenin 7-*O*-rutinoside and luteolin 7-*O*-rutinoside.

*Conclusions*: The antioxidant potential of varieties of *S. edule* grown in Costa Rica was confirmed, identifying the 845 variety as the best antioxidant profile.

*Keywords*: antioxidant; chayote; flavonoid; *Sechium edule*.

Resumen

*Contexto*: El fruto de *Sechium edule* es cultivado y consumido por su valor nutricional en distintos países de América Latina, siendo Costa Rica uno de los principales productores y exportadores. Su consumo como antioxidante ha sido sugerido por su contenido de flavonoides, pero la selección de las variedades para el cultivo no involucra el perfil fitoquímico de las estas.

*Objetivos*: Caracterizar el perfil fitoquímico y antioxidante de las variedades cultivadas de *S. edule* en Costa Rica.

*Métodos*: Se midió la capacidad de eliminación de radicales *in vitro* de extractos de distintas variedades de *S. edule* a través de los métodos de DPPH y ORAC, la inhibición de la peroxidación lipídica en hígado de rata y el potencial redox por voltametría cíclica de dichos extractos. Se aislaron glicósidos de flavonoides por HPLC y su estructura se determinó por RMN.

*Resultados*: Las variedades de *S. edule* cultivadas en Costa Rica mostraron una buena capacidad de eliminación de radicales e inhibición de la peroxidación lipídica; mostrando diferencias significativas entre las variedades. Además, los potenciales redox determinados por métodos electroquímicos sugirieron la presencia de flavonoides, lo cual fue confirmado con el aislamiento de apigenina 7-*O*-rutinósido y luteolina 7-*O*-rutinósido.

*Conclusiones*: Se confirmó el potencial antioxidante de las variedades de *S. edule* cultivadas en Costa Rica, identificando la variedad 845 como la del mejor perfil antioxidante.

*Palabras Clave*: antioxidante; chayote; flavonoide; *Sechium edule*.

ARTICLE INFO
Received: February 5, 2018.
Received in revised form: July 22, 2018.
Accepted: August 5, 2018.
Available Online: August 14, 2018.
Declaration of interests: The authors declare no conflict of interest.
Funding: This study was supported by *Consejo Nacional de Rectores of Costa Rica* (CONARE) (Project number 817-B2-656).

## INTRODUCTION

*Sechium edule* (Jacq.) Swartz (*Cucurbitaceae*), also known as chayote, is an herbaceous plant originally native to Mesoamerica. This plant has been cultivated in Mexico and Central America since pre-Columbian times, where their fruits are very appreciated in rural zones because of their nutritional value. Nowadays, this crop is produced in many tropical countries such as China, India and Madagascar (Siciliano et al, 2004; Loizzo et al, 2016).

Costa Rica is one of the principals growing regions and has become the leading exporter worldwide; therefore, this country has a high degree of genetic diversity for the crop (Newstrom, 1991; Abdelnour and Rocha, 2008; Loizzo et al, 2016).

The utilization of *S. edule* as functional food has been suggested previously because of the content of flavonoids and their antioxidant activity (Ordoñez et al, 2006; Chao et al, 2014; Loizzo et al, 2016). Despite this potential, the choice of varieties for farming excludes the metabolome of the selection criteria, this situation reduces the potential of the crop as a functional food.

The chemical nature of the flavonoids into *S. edule* fruit and their concentrations have an important impact on the antioxidant potential of the fruit (Siciliano et al, 2004). Through multifactorial antioxidant screening methods, the antioxidant profile determination of the fruits helps to select the better samples for harvest (Tulipani et al, 2008, Riviello-Flores et al, 2018). Therefore, the phytochemical analysis and antioxidant profile determination of the germplasm of Costa Rican *S. edule* provide an important tool for improving the value of the crop as a functional food.

The aim of the present work was to characterize the phytochemical and antioxidant profile of the *S. edule* varieties grown in Costa Rica.

## MATERIAL AND METHODS

### Reagents

2,2-diphenyl-1-picrylhydrazyl radical (DPPH), 2,2'-Azobis-(2-amidino-propane) dihydrochloride (AAPH), (±)-6-hydroxy-2,5,7,8-tetramethylchromane-2-carboxylic acid (Trolox), *tert*-butyl hydroperoxide (TBHP), and deuterated methanol ($CD_3OD$) were obtained from Sigma-Aldrich® Chemicals (USA). Fluorescein sodium salt and thiobarbituric acid were obtained from Merck® (Germany).

### Animals

Inhibition of lipid peroxidation assay was performed with Sprague Dawley rats (both sexes, 180-220 g) obtained from the Biological Assays Laboratory (LEBI) Bioterium, University of Costa Rica. The animals were maintained under standard conditions of temperature and humidity, light/dark cycles of 12 h and food and water *ad libitum*. The use of animals in this study was approved by the Institutional Committee for the Care and Use of Animals (CICUA), University of Costa Rica (CICUA-14-10, July 10th, 2010).

### Plant material

Plant material was obtained from a germplasm collection at Santa Lucía Experimental Farm located in Barva, Heredia, Costa Rica (10°01'19" N 84°06'43" W). Three different types of *S. edule* (identified as *S. edule* 845, *S. edule* 846 and *S. edule* 853) (Fig. 1) were selected from 42 different varieties according to their high content of phenolic and steroid compounds previously determined by School of Chemistry, National University of Costa Rica (Rojas, 2014). The samples correspond to a 1:1 mixture of mature and unripe lyophilized fruit.

### *S. edule* extracts preparation

For DPPH, lipid peroxidation and cyclic voltammetry assays, 10 g of the samples of *S. edule*

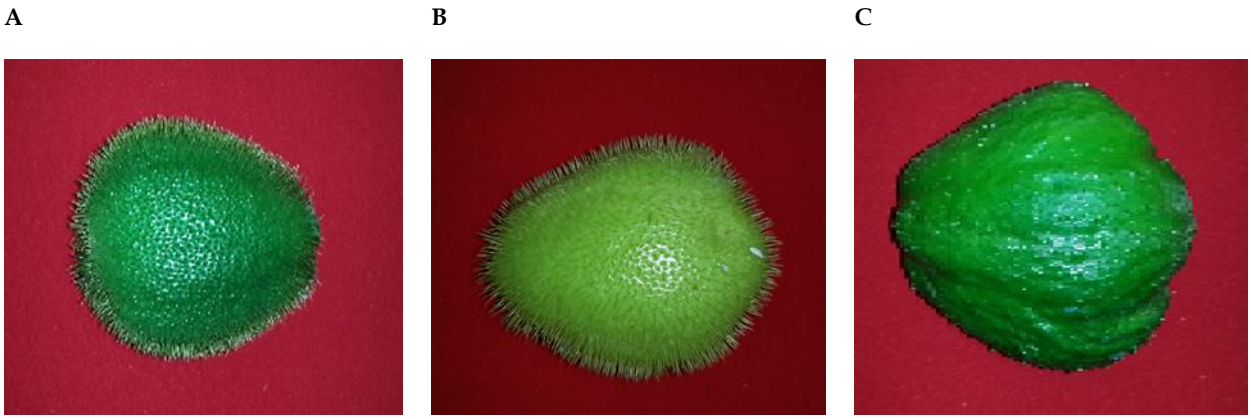


**Figure 1.** *Sechium edule* unripe fruit selected from 42 different Costa Rican varieties according to their high content of phenolic and steroid compounds. **A)** *S. edule* 845, **B)** *S. edule* 846 y **C)** *S. edule* 853.

were macerated for 24 h using as solvent 100 mL methanol: acetone: acidified water (M:A:A) at 7:7:6 proportion. Then, samples were filtered by cellulose acetate membrane disc filters 0.2 µm (Corning®, USA), concentrated using a rotatory evaporation (Büchi® Rotavapor®, Germany), and lyophilized (Labconco® 4.5, USA).

For the ORAC test, 10 g of the samples of *S. edule* were extracted in an ultrasonic bath (Branson 2510, USA) for 5 minutes using 100 mL of the same solvent before. Then, samples were centrifuged at 805 g for 5 min, and the supernatant was lyophilized (Labconco® 4.5, USA). The extraction process was carried out in triplicate.

## Antioxidant activity

### *DPPH free radical scavenging activity*

The lyophilized fruit extract was dissolved in methanol to prepare stock solutions of 85 ± 3 mg/mL. The radical-scavenging activity was performed according to the method described previously (Brand-Williams et al, 1995; Dudonné et al, 2009). Briefly, 25 µL of fruit extract at different concentrations were incubated with 100 µL of DPPH 500 µM dissolved in methanol, at room temperature in the dark for 30 min. Then, the absorbance was measured at 520 nm against sample blanks for each concentration using a spectrofluorometer (BioTek® Synergy HT®, USA). A positive control prepared with DPPH was used to calculate the percentage of radical scavenging activity (RSA) as:

$$\%RSA = \frac{A_{control} - A_{Sample}}{A_{control}} \times 100$$

The percentage of RSA was plotted against the sample concentration to calculate the $IC_{50}$, which is the concentration of extract required to reach the 50% of inhibition of DPPH free radical formation (Londoño, 2012). This assay was performed in triplicates.

### *Oxygen Radicals Absorbance Capacity (ORAC) assay*

The lyophilized fruit extract was dissolved in phosphate buffer (PBS, potassium phosphate dibasic 15 mM, monobasic potassium phosphate 23 mM, pH 7.4) to prepare a stock solution of 1 mg/mL. The ORAC assay was performed according to the method described formerly (Wayner, 1985; Dudonné et al, 2009). Briefly, different concentrations of fruit extract were incubated with fluorescein 81.6 nM dissolved in PBS, at 37°C in dark 96-well plates for 10 min. Then, AAHP 153 mM dissolved in PBS was added to the mixture to induce oxidation. Fluorescence was measured every 1 min for 35 min, using 491 nm as wavelength excitation and 515 nm as wavelength emission, measures were carried using a spectrofluorometer (BioTek® Synergy HT®, USA). Results were expressed as µM of Trolox equivalents, according to Trolox calibration curve (6.25, 12.5, 25, and 50 µM)

with the interpolation of the area under the fluorescence decay curve (AUC) *versus* Trolox concentration. The AUC was calculated as:

$$AUC = 1 + \sum_{i=1}^{i=35} f_i/f_0$$

Where $f_0$ is the initial fluorescence reading at 0 min and fi is the fluorescence reading at time i. The experiment was performed in triplicate for each of the *S. edule* fruit extracts (Wayner et al., 1985; Huang D et al., 2002).

*Inhibition of lipid peroxidation in liver homogenates*

Five rats were anaesthetized with a mixture of $CO_2$ and air in a chamber for 1-2 min until lose consciousness, then sacrificed by decapitation. The liver tissue of each rat was obtained and homogenized in cold solution of PBS using a tissue homogenizer IKA® X Ultraturrax® T25 (Germany) equipment. Homogenates were centrifuged at 13 132 *g* for 15 min. Then, *S. edule* fruit extract at different concentrations was incubated with 0.75 mL of each liver supernatant, for 30 min at 37°C. Lipid peroxidation was then induced with *tert*-butyl hydroperoxide (TBHP) at a final concentration of 1.7 mM, for 1 h at 37°C. The concentration of malonyl dialdehyde (MDA) was measured as the end product of lipid peroxidation by the method described by (Heath and Packer, 1968). Briefly, 0.25 mL of liver homogenate pre-incubated with the extract, 0.25 mL of trichloroacetic acid (TCA) 35%, 0.25 mL of Tris buffer (50 mM, pH 7.4), and 0.5 mL of thiobarbituric acid 0.75% were mixed and heated at 100°C for 45 min. After cooling, 0.5 mL of TCA 70% was added to each sample and then centrifuged at 1575 *g* for 15 min. The absorbance of the supernatant was measured at 532 nm against a sample blank using a spectrofluorometer (BioTek® Synergy HT®, USA). The concentration of MDA was calculated as described previously (Heath and Packer, 1968; Ferreira et al., 2009; Dinakaran et al., 2011; Tang et al, 2012) and results were expressed as nmol of MDA per gram of liver tissue (nmol MDA/g tissue). Commercial Trolox was used as positive control. Basal levels of lipid peroxidation were established and correspond to liver homogenates without TBHP treatment. The experiment was performed in triplicate for each of the *S. edule* fruit extracts.

*Cyclic voltammetry*

Different concentrations of *S. edule* extracts dissolved in methanol were mixed with 0.1 M phosphate buffer (pH 7.2) in a ratio of 6:4 (v/v) according to the method described previously (Zhang et al, 2011). A potentiostat BAS 100B (Bioanalytical Systems®, USA) with a conventional three-electrode system: Ag/AgCl reference electrode, platinum auxiliary electrode, and glassy carbon working electrode, was used for all electrochemical measurements. Prior to each cycle, working electrode was polished with alumina paste 0.05 µm, rinsed with Milli-Q water and sonicated for 15 min. Also, in order to eliminate the oxygen, sample solutions were sonicated for 15 min prior to each cycle. Then, sample solutions were transferred into an electrochemical cell. Buffer phosphates was used as supporting electrolyte media, which allowed the redox reaction. The cyclic voltammograms were recorded from -1400 mV to +1400 mV at the scanning rate of 25 mV/s. All measurements were carried out at room temperature against a blank consisting of methanol and PBS. Quercetin 150 µM dissolved in methanol was used as a standard solution (Cosio et al, 2006; Zielinska et al, 2007; Zhang et al, 2011).

**Flavonoid isolation**

A mixture of the three varieties of *S. edule* was extracted several times using a mixture of methanol:dichloromethane (1:1) (extraction yield 46.5%). During the extraction, the sample was sonicated for 15 min at 30°C and filtered. After that, the solvent was concentrated using a rotatory evaporation (Büchi® Rotavapor®, Germany). In brief, 100 g of solid phase resin (Waters® Preparative C18 125Å, 55–105 µm, USA) was added to a glass column (Kontes, 2.5 cm internal diameter × 30 cm) and conditioned by passing 100 mL of methanol through the column bed followed by 500 mL water (1.0 mL/L trifluoroacetic acid, TFA). Sample was introduced in the column using methanol:water 1:9, and a mixture in different proportions of the same solvent was used as eluent. Fractions were

collected according to the result of a Folin-Ciocalteu test of the fractions. Once fractions rich in reducing compounds were put together, the solvent was removed using a rotatory evaporation (Büchi® Rotavapor®, Germany). Flavonoids from concentrated fractions were obtained using preparative HPLC with C18 column, a gradient of acetonitrile (30-100%, 25 min) as mobile phase, and DAD detector ($t_{R\,1}$: 11.4 min, $t_{R\,2}$: 9.8 min).

Pure compounds were analysed by 1D ($^{1}H$, $^{13}C$) and 2D (H-H COSY, HSQC, HMBC) NMR experiments at Varian® Mercury® 400 MHz (USA). Chemical shift was expressed in δ (ppm) referring to the solvent peaks $\delta_H$ 4.87 and $\delta_C$ 49.0 for $CD_3OD$. Spectra were analysed using MestReNova software (Mestrelab®, Spain).

The radical-scavenging activity of isolated flavonoids was determined by ORAC test as described above, with the following modification: AAPH reagent at 75 mM, time of incubation 20 min, 485 nm as wavelength excitation and 528 nm as wavelength emission.

### Statistical analysis

Results were expressed as the mean ± standard error of the mean (SEM). Statistical significance was determined by one-way analysis of variance (ANOVA) with Tukey as post hoc test using the IBM SPSS Statistics 22.0 (USA). A value of $p<0.05$ was considered statistically significant. All tests to determine the antioxidant activity were performed in triplicate.

## RESULTS AND DISCUSSION

### Antioxidant profile of *Sechium edule* varieties

The DPPH assay assesses the capacity of a substance to work through a reaction involving free radicals. Previously, *S. edule* reports an $IC_{50}$ value of 2-32 µg/mL for ethanolic extract of aerial parts (Ordoñez et al, 2006). Our results (Table 1) showed considerably less radical scavenging activity; nevertheless, it is difficult to compare results from different laboratories because of extract nature and reaction conditions variance (Sharma and Bhat, 2009). The ORAC assay is a widely used method for antioxidant potential in food. This assay uses peroxyl radicals that are closer to reactive oxygen species (ROS), which oxidized lipids in food. Previous data display values from 49 to 120 µmol TE/g DS (Chao et al, 2014), which are in the same order of our data (Table 1). While 845 variety shows the better activity in DPPH assay, the 846 variety presents higher ORAC values. In both radical scavenging *in vitro* tests, significant differences ($p<0.05$) were obtained between *S. edule* varieties.

**Table 1.** Antioxidant activity of three varieties of *Sechium edule* fruit extracts by 2,2-diphenyl-1-picrylhydrazyl (DPPH) free radical-scavenging and Oxygen Radical Absorbance Capacity (ORAC) assays.

| Variety | DPPH $IC_{50}$ (mg/mL) | ORAC (µmol TE/g DS) |
|---|---|---|
| 845 | 51.6 ± 0.1[a] | 31.1 ± 2.6[b] |
| 846 | 68.0 ± 2.0[a] | 39.6 ± 0.9 |
| 853 | 80.1 ± 0.8[a] | 25.5 ± 1.8[b] |
| Apigenin 7-*O*-rutinoside (**1**) | - | 6.9 ± 0.2 |
| Luteolin 7-*O*-rutinoside (**2**) | - | 6.9 ± 0.7 |

Results are present as the mean of the concentration necessary to reach the 50% radical-scavenging activity ($IC_{50}$) and micromoles of Trolox equivalents per gram of dried sample (µmol TE/g DS) ± standard error of three replicate experiments (n=3). [a]Significant difference between varieties ($p<0.05$). [b]Significant difference compared to 846 variety ($p<0.05$).

The inhibition of lipid peroxidation assay is an *in vitro* test that measures the degree of suppression of the lipid peroxidation in rat liver tissue homogenate through the detection of MDA generated during the oxidative destruction of lipid (Tang et al, 2012). In general, *S. edule* demonstrated the capacity of prevent the oxidative damage of the lipids in the liver tissue (Fig. 2), whilst the inhibition of lipid peroxidation was higher for varieties 845 and 853 at 400 mg/mL. These results are important because of the approximation to an oxidative damage in a biological context. In spite of showing high antioxidant activity at *in vitro* tests, 846 variety did not show good inhibition of lipid peroxidation. The non-correlation between chemi-

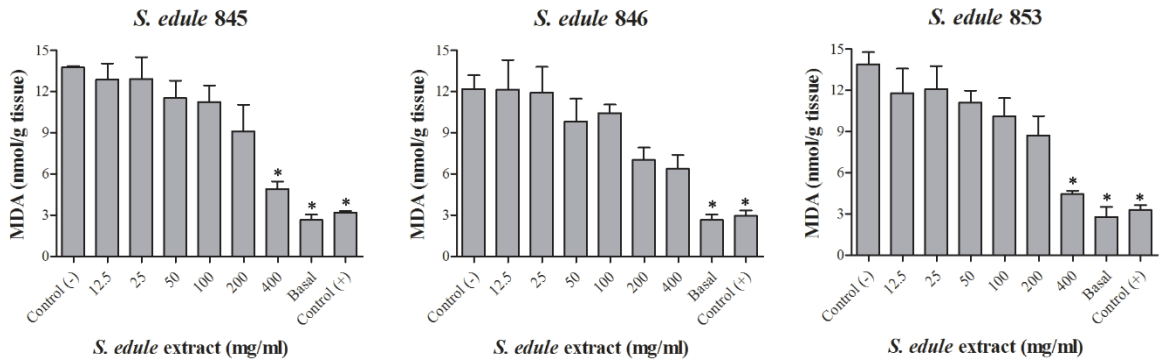


**Figure 2.** Inhibitory capacity of three genotypes of *Sechium edule* fruit extracts against lipid peroxidation induced by *tert*-butyl hydroperoxide in liver homogenates.

Negative controls correspond to liver homogenates expose to *tert*-butyl hydroperoxide without *S. edule* treatment. Positive controls correspond to liver homogenates exposed to tert-butyl hydroperoxide and treated with Trolox. Basal values correspond to liver homogenates without tert-butyl hydroperoxide treatment. Results were expressed as nmol of MDA per gram of liver tissue (nmol MDA/g tissue). Each value is the mean ± SE (n = 3). *p < 0.05 compared to negative control.

cal-based and cell-based assays has been discussed in many preceding works (Prior and Cao, 1999; López-Alarcón and Denicola, 2013), and it was related with different factors that make necessary to apply diverse analytical methods to evaluate antioxidant activity.

Cyclic voltammetry (CV) is a widely used electrochemical technique that has been suggested as an antioxidant assay. This technique allows measure the reducing potential of a sample as a function of two parameters: anodic peak current ($I_a$) and the peak oxidation potential ($E_{pa}$). These parameters analyse the antioxidant capacity of substances, where $E_{pa}$ is related to redox properties of the antioxidant and $I_a$ with the amount of charge transferred (Zhang et al, 2011; Prior and Cao, 1999). Previous reports related the antioxidant activity of *S. edule* with its flavonoid composition (Ordoñez et al, 2006; Chao et al, 2014; Loizzo et al, 2016); hence, the electrochemical behaviour of the extract gives an approximation to the kind of polyphenol present in the sample.

Fig. 3 presents the cyclic voltammogram of three genotypes of *S. edule* fruit extracts and quercetin as a reference. The quercetin shows three outstanding waves at 0.0, +0.5 and +1.0, which correspond to the three hydroxyl groups present at quercetin structure. On the other hand, *S. edule* extracts present only two outstanding waves at +0.5 and +1.0, suggesting activity by two hydroxyl groups, with a similar trend to apigenin. The $E_{pa}$ indicate electron-donating capacity, which is determined by the reactivity of the hydroxyl groups. It was considered that flavonoids with lower peak potentials would have higher radical-scavenging activity (Zhang et al, 2011). All the samples exhibited similar oxidation potential.

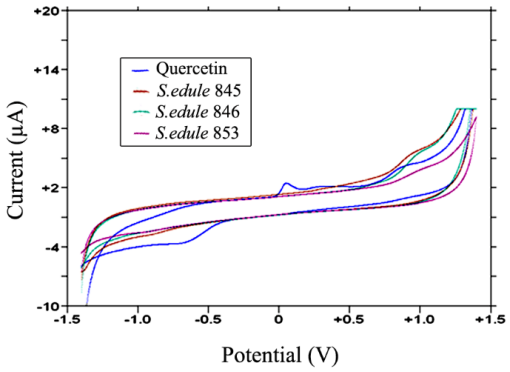


**Figure 3**. Cyclic voltammogram of three genotypes of *Sechium edule* fruit extracts dissolved in methanol at concentrations between 82-88 mg/mL.

The samples were measured with 0.1 M phosphate buffer (pH 7.2) in ratio 6: 4 v/v and registered through a potential range from -1400 mV to +1400 mV at a scan rate of 25 mV/s. Quercetin dissolved in methanol was used as a control. Blank consisted of methanol and 0.1 M phosphate buffer (pH 7.2).

Furthermore, the $I_a$ produced is proportional to the combined concentration of antioxidant compounds (Zhang et al, 2011). Although 845 and 846

varieties displayed similar shape, 853 variety presented small peaks that suggest a lower concentration of flavonoids in the sample. This result is congruent with *in vitro* antioxidant activity, where 845 and 846 varieties showed more activity than 853 variety.

Summary, 845 and 846 varieties displayed higher antioxidant activity at radical-scavenging and electrochemical assays, while 845 variety showed higher activity at lipid peroxidation assay. Thus, there were differences between antioxidant activities of the *S. edule* varieties, where the 845 variety had the better antioxidant profile.

### Isolation and structure determination

In order to confirm the phytochemical profile of polyphenols into *S. edule*, flavonoids were isolated by HPLC. Two compounds (Fig. 4) were elucidated as apigenin 7-*O*-rutinoside (**1**) and luteolin 7-*O*-rutinoside (**2**) according to $^{1}H$ and $^{13}C$ NMR (400 MHz) data (Table 2). The spectral characterization of the compounds is congruent with reported data for flavonoid glycosides (Wang et al, 2003; Siciliano et al, 2004).

Both compounds were identified in *S. edule* formerly (Siciliano et al, 2004) and their isolation in the sample confirmed the results of the cyclic voltammetry, which suggest this kind of aglycones as discussed above. Future research should determine the concentration of these flavonoids in the samples in order to correlate them with their antioxidant profile.

### Radical-scavenging activity of the flavonoids

The isolated compound´s antioxidant characterization was carried out using ORAC method, showing values of 6.9 ± 0.2 and 6.9 ± 0.7 µmol TE/µmol of apigenin 7-*O*-rutinoside and luteolin 7-*O*-rutinoside, respectively (Table 1). It was expected that the value of the antioxidant capacity obtained from the pure compounds was higher than those found in the crude extract as a consequence of the eliminations of other compounds present in the extract with no antioxidant capacity or a low one (Dan et al, 2015). The results of the radical-scavenging activity of the flavonoids glycosides were congruent with the projected values according to previous reports for their aglycones (Zhang et al, 2013).

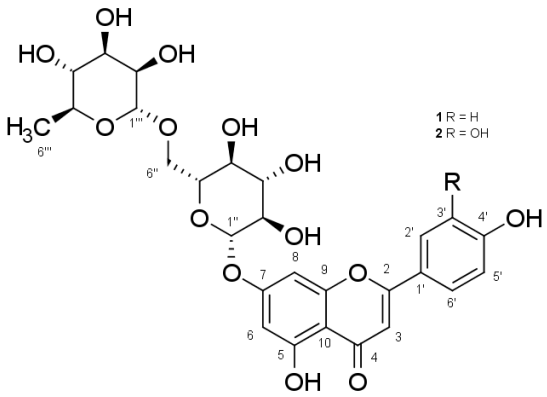


**Figure 4.** Structure of compounds (**1**) apigenin 7-*O*-rutinoside and (**2**) luteolin 7-*O*-rutinoside found in *S. edule* fruit varieties grown in Costa Rica.

## CONCLUSIONS

The fruit of *Sechium edule* could be considered as a functional food because of its antioxidant activity, which is related with the phenolic compounds into the vegetable; however, the selection of varieties for farming considers agronomic and morphologic characteristics, not the metabolome of the variety. The phytochemical selection approach helps to promote the health value of the food. In this study, in addition to confirming the potential of *Sechium edule* in health, differences in the antioxidant profile of the three varieties grown in Costa Rica were demonstrated. The 845 variety displayed the best antioxidant profile by *in vitro* tests. Moreover, apigenin 7-*O*-rutinoside and luteolin 7-*O*-rutinoside were identified in the samples and their antioxidant activity was confirmed.

This work could be the basis to promote the consumption of *Sechium edule* as healthy food and to make aware the selection of the right variety for this purpose.

**Table 2.** $^{1}H$ and $^{13}C$ NMR (400 MHz) data of compounds **1** and **2** ($CD_3OD$, δ [ppm], J [Hz]).

| Position | Compound 1 | | Compound 2 | |
|---|---|---|---|---|
| | $\delta_C$ | $\delta_H$ | $\delta_C$ | $\delta_H$ |
| **Aglycon** | | | | |
| 2 | 166.9 | - | 166.9 | - |
| 3 | 104.2 | 6.68 s | 104.3 | 6.61 s |
| 4 | 184.1 | - | 184.0 | - |
| 5 | 163.0 | - | 163.0 | - |
| 6 | 101.1 | 6.55 d (1.8) | 101.1 | 6.54 d (2) |
| 7 | 158.9 | - | 158.9 | - |
| 8 | 96.3 | 6.80 d (1.8) | 96.1 | 6.75 d (2) |
| 9 | 164.7 | - | 164.7 | - |
| 10 | 107.1 | - | 107.1 | - |
| 1' | 123.1 | - | 123.5 | - |
| 2' | 117.1 | 6.98 d (8.6) | 116.9 | 6.94 d (9.2) |
| 3' | 129.6 | 7.91 d (8.6) | 147.0 | - |
| 4' | 162.9 | - | 151.2 | - |
| 5' | 129.6 | 7.91 d (8.6) | 114.3 | 7.42 ddd (2.4, 2.4, 6.4) |
| 6' | 117.1 | 6.98 d (8.6) | 120.6 | 7.42 ddd (2.4, 2.4, 6.4) |
| **Sugars** | | | | |
| *Glucose* | | | | |
| 1'' | 101.5 | 5.07 d (6.8) | 101.6 | 5.05 d (7.2) |
| 2'' | 77.8 | 3.52 d (4.8) | 77.8 | 3.52 dd (1.2, 8) |
| 3'' | 74.7 | 3.51 d (3.2) | 74.7 | 3.51 d (3.2) |
| 4'' | 77.1 | 3.68 m | 77.2 | 3.68 m |
| 5'' | 71.3 | 3.44 dd (8.8, 8.8) | 71.3 | 3.43 dd (10, 10) |
| 6'' | 67.4 | 4.07 d (9.8)<br>3.68 d (9.8) | 67.5 | 4.08 d (9.2)<br>3.68 d (9.2) |
| *Rhamnose* | | | | |
| 1''' | 102.1 | 4.74 d (1.8) | 102.1 | 4.74 d (1.2) |
| 2''' | 72.1 | 3.92 dd (1.8, 3.4) | 72.1 | 3.93 dd (1.6, 3.6) |
| 3''' | 72.4 | 3.73 dd (3.4, 9.6) | 72.4 | 3.75 d (2) |
| 4''' | 74.1 | 3.36 m | 74.1 | 3.37 m |
| 5''' | 69.8 | 3.67 m | 69.8 | 3.67 m |
| 6''' | 17.9 | 1.20 d (8) | 17.9 | 1.21 d (6.4) |

## CONFLICT OF INTEREST

The authors declare no conflict of interest.

## ACKNOWLEDGMENTS

The inhibition of lipid peroxidation assay was performed at Biochemistry Department, School of Medicine, University of Costa Rica; the authors thank to Silvia Quesada for her collaboration. The cyclic voltammetry assay was carried out at Research Center in Electrochemistry and Chemical Energy (CELEQ), University of Costa Rica; the authors thank to Ana Lorena Alvarado for her collaboration. Authors also acknowledge School of Chemistry, University of Costa Rica, for access to NMR instrument. This study was supported by a grant of *Consejo Nacional de Rectores of Costa Rica* (CONARE) (Project number 817-B2-656).

## SUPPLEMENTARY DATA

Supplementary data associated with this article can be found at http://jppres.com/jppres/pdf/vol6/jppres18.379_6.6.448.suppl.pdf

## REFERENCES

Abdelnour A, Rocha O (2008) Genetic characterization of a collection of chayote, *Sechium edule* (Jacq.) Swartz, in Costa Rica by using isozyme markers. Genet Resour Crop Evol 55(1): 163–170.

Brand-Williams W, Cuvelier M, Berset C (1995) Use of a free radical method to evaluate antioxidant activity. Food Sci Technol 28(1): 25–30.

Chao PY, Lin KH, Liu YF, Hsu JI, Yang CM, Lai, JY (2014) Antioxidant activity in extracts of 27 Indigenous Taiwanese vegetables. Nutrients 6(5): 2115–2130.

Cosio M, Buratti S, Mannino S, Benedetti S (2006) Use of an electrochemical method to evaluate the antioxidant activity of herb extracts from the Labiatae family. Food Chem 97: 725–731.

Dan L, Yuhian Z, Yan L, Ruifang S, Min X (2015) Purified anthocyanin supplementation reduces dyslipidemia, enhances antioxidant capacity and prevents insulin resistance in diabetic patients. J Nutr 14(4): 742–748.

Dinakaran S, Saraswathi N, Nalini V, Srisudharson, Bodanapu V, Avasarala H, Banji D (2011) Anti lipid peroxidation activity of *Piper trioicum* Roxb. and *Physalis minima* L. extracts. Pak J Pharm Sci 24(3): 411–413.

Dudonné S, Vitrac X, Coutière P, Woillez M, Mérillon JM (2009) Comparative study of antioxidant properties and total phenolic content of 30 plant extracts of industrial interest using DPPH, ABTS, FRAP, SOD, and ORAC assays. J Agric Food Chem 57(5): 1768–1774.

Ferreira PM, Monteiro LS, Coban T, Suzen, S (2009) Comparative effect of N-substituted dehydroamino acids and alpha-tocopherol on rat liver lipid peroxidation activities. J Enzyme Inhib Med Chem 24(4): 967–971.

Heath R, Packer L (1968) Photoperoxidation in isolated chloro-plants. I. Kinetics and stoichiometry of fatty acid peroxidation. Arch Biochem Biophys 125(1): 189–198.

Huang D, Ou B, Hampsch-Woodill M, Flanagan J, Prior RL (2002) High-throughput assay of oxygen radical absorbance capacity (ORAC) using a multichannel liquid handling system coupled with a microplate fluorescence reader in 96-well format. J Agric Food Chem 50(16): 4437–4444.

Loizzo M, Bonesi M, Menichini F, Tenuta M, Leporini M, Tundis R (2016) Antioxidant and carbohydrate-hydrolysing enzymes potential of *Sechium edule* (Jacq.) Swartz (Cucurbitaceae) peel, leaves and pulp fresh and processed. Plant Foods Hum Nutr 71(4): 381–387.

Londoño J (2012) Antioxidantes: importancia biológica y métodos para medir su actividad. Caldas, Colombia: Corporación Universitaria Lasallista.

López-Alarcón C, Denicola A (2013) Evaluating the antioxidant capacity of natural products: A review on chemical and cellular-based assays. Anal Chim Acta 763: 1–10.

Newstrom L (1991) Evidence for the origin of chayote, *Sechium edule* (Cucurbitaceae). Econ Bot 45(3): 410–428.

Ordoñez A, Gómez J, Vattuone M, Isla M (2006) Antioxidant activities of *Sechium edule* (Jacq.) Swartz extracts. Food Chem 97(3): 452–458.

Prior R, Cao G (1999) *In vivo* total antioxidant capacity: comparison of different analytical methods. Free Radic Biol Med 27(11): 1173–1181.

Rojas C (2014) Caracterización de la colección de germoplasma de chayote (*Sechium edule*) de la Universidad Nacional de Costa Rica, en cuanto al contenido de compuestos fenólicos y esteroideos totales. Grade Thesis, Chemistry School, National University of Costa Rica.

Riviello-Flores M, Arévalo-Galarza M, Cadena-Iñiguez J, Soto-Hernández R, Ruiz-Posadas L, Gómez-Merino F (2018) Nutraceutic characteristics of the extracts and juice of chayote (*Sechium edule* (Jacq.) Sw.) fruits. Beverages 4(2): 37.

Sharma O, Bhat T (2009) DPPH antioxidant assay revisited. Food Chem 113(4): 1202–1205.

Siciliano T, De Tommasi N, Morelli I, Braca A (2004) Study of flavonoids of *Sechium edule* (Jacq) Swartz (Cucurbitaceae) different edible organs by liquid chromatography photodiode array mass spectrometry. J Agric Food Chem 52(21): 6510–6515.

Tang X, Wu Q, Le G, Wang J, Yin K, Shi Y (2012) Structural and antioxidant modification of wheat peptides modified by the heat and lipid peroxidation product malondialdehyde. J Food Sci 77(1): H16–H22.

Tulipani S, Mezzetti B, Capocasa F, Bompadre S, Beekwilder J, Ric de Vos C, Capanoglu E, Bovy A, Battino M (2008) Antioxidants, phenolic compounds, and nutritional

quality of different strawberry genotypes. J Agric Food Chem 56(3): 696–704.

Wang M, Simon J, Aviles I, He K, Zheng Q, Tadmor Y (2003) Analysis of antioxidative phenolic compounds in artichoke (*Cynara scolymus* L.). J Agric Food Chem 51(3): 601–608.

Wayner D, Burton G, Ingold K, Locke S (1985) Quantitative Measurement of the total, peroxyl radical trapping antioxidant capability of human blood plasma by controlled peroxidation. FEBS Lett 187(1): 33–37.

Zhang D, Chu L, Liu Y, Wang A, Ji B, Wu W, Zhuo F, Wei Y, Cheng Q, Cai S, Xie L, Jia G (2011) Analysis of the antioxidant capacities of flavonoids under different spectrophotometric assays using cyclic voltammetry and density functional theory. J Agric Food Chem 59(18): 10277–10285.

Zhang D, Liu Y, Chu L, Wei Y, Wang D, Cai S, Zhou F, Ji B (2013) Relationship Between the Structures of Flavonoids and Oxygen Radical Absorbance Capacity Values: A Quantum Chemical Analysis. J Physic Chem A 117(8): 1784–1794.

Zielinska D, Szawara-Nowak D, Zielinski H (2007) Comparison of spectrophotometric and electrochemical methods for the evaluation of the antioxidant capacity of buckwheat products after hydrothermal treatment. J Agric Food Chem 55(15): 6124–6131.

**AUTHOR CONTRIBUTION:**

| Contribution | Parra J | Hernández P | Ocampo-Maroto F | Álvarez-Valverde V | Carvajal-Miranda Y | Rodríguez-Rodríguez G | Herrera C |
|---|---|---|---|---|---|---|---|
| Concepts or ideas | x | | | | | x | x |
| Design | x | | | | | x | x |
| Definition of intellectual content | x | | | | | x | x |
| Literature search | x | x | | | | | |
| Experimental studies | x | x | x | x | x | | x |
| Data acquisition | | x | x | | | | |
| Data analysis | x | | | | | | |
| Statistical analysis | | x | | | | | x |
| Manuscript preparation | x | | | | | | |
| Manuscript editing | x | | | | | x | x |
| Manuscript review | x | x | x | x | x | x | x |

**Citation Format:** Parra J, Hernández P, Ocampo-Maroto F, Álvarez-Valverde V, Carvajal-Miranda Y, Rodríguez-Rodríguez G, Herrera C (2018) Phytochemical characterization and antioxidant profile of *Sechium edule* (Jacq) Swartz (*Cucurbitaceae*) varieties grown in Costa Rica. J Pharm Pharmacogn Res 6(6): 448–457.