PHYTOCHEMICALS, TOTAL PHENOLIC CONTENTS AND ANTIOXIDANT ACTIVITIES OF WATER AND ETHANOL EXTRACTS FROM INCA PEANUT (*Plukenetia volubilis* L.) LEAVES

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Abstract

Inca peanut leaves (*Plukenetia volubilis* L.) is generally consumed in form of tea but there is less research which study about bioactivity of Inca peanut leaves cultivated in Thailand. The objectives of this study were to evaluate the phytochemical constituents, total phenolic contents and antioxidant activities of Inca peanut leaves. The fresh leaves and dried leaves were extracted by water (30°C), warm water (60°C), ethanol 95% (30°C) and warm ethanol 95% (60°C). It was found that dried leaves extracted with warm water (DW60) provided the highest percentage yield (19.01±0.43%w/w). The phytochemical screening of the extracts revealed the presence of phenols, flavonoids and alkaloids in all extracts. The main minerals in Inca peanut leaves were calcium, silicon and potassium. Dried leaves extracted with warm ethanol (60°C) (DE60) showed the highest ($p<0.05$) amount of total phenolic contents (22.05±1.20 mg GAE/g DW). The antioxidant activities were analyzed by ABTS radical cation scavenging activity and DPPH radical scavenging activity. It was found that DW 60 exhibited the highest ABTS radical cation scavenging activity (193.07±6.14 mg AEAC/g DW, IC50 3.07±0.91 µg/ml) while DE60 exhibited the highest DPPH radical scavenging activity (27.27±0.71 mg AEAC/g DW, IC50 119.81±4.35 µg/ml). Thus, the results of this work can be concluded that dried leaves extract obtained by warm water and warm ethanol 95% are suitable for further use as antioxidant for prevention and management of free radicals mediated oxidative stress.

Keywords: Antioxidant, Inca peanut, Phenolic, Phytochemical, *Plukenetia volubilis* L.

INTRODUCTION

Reactive oxygen species or ROS are broad term that include both oxygen free radicals, which have unpaired electrons and oxidizing agents that are not free radicals (Bayir, 2005). ROS production occurs continuously in all cells as part of normal cellular function. Excessive ROS are generated when expose to microbial infections, extensive exercise, pollutants, cigarette smoke, alcohol, and sunlight. Antioxidants are substances which prohibits oxidation and scavenging free radicals. Superoxide dismutase, catalase, glutathione peroxidase, glutathione and coenzyme Q are examples of endogenous antioxidant which prevent the damage of the cells caused by ROS (Halliwell, 2011). The imbalance between the production of ROS and the availability of antioxidants leads to oxidative stress (Poljsak, Suput & Milisav, 2013). Oxidative stress is linked to formation of many degenerative diseases, including cancer, cardiovascular disease, Alzheimer, cataracts, and aging (Kehrer & Klotz, 2015).

Natural antioxidants present in various parts of the plant such as stems, barks, leaves, flowers, pollen, roots, fruits, nuts and seeds. The most common natural antioxidants are phenolics, flavonoids, lignans, terpenes, cinnamic acid derivatives, coumarins, tocopherols and polyfunctional organic acids (Farias et al., 2013). Natural antioxidants have considerable interest in the field of food chemistry, pharmacy, cosmetic and medicinal due to a wide range of favorable biological effects. Natural antioxidants may function as reducing agents, free radical scavengers, hydrogen donors, singlet oxygen quenchers and metal chelators (Lakshmanashetty et al., 2010).

Inca peanut leaves *(Plukenetia volubilis* L.*)* can be found in the Amazon region of South America. It has a star–shaped fruit capsule which contains edible dark brown oval seeds. The seeds have been utilized for oil production (Gonzalez–Aspajo et al., 2015). From recent years Inca peanut oil has caught attention as a novel source of oil rich in unsaturated fatty acids. The main composition of fatty acids in Inca peanut oil cultivated in Thailand was studied and found the presence of linoleic acid or ω6 (45.72%), linolenic acid or ω3 (42.27%), palmitic acid (6.42%) and stearic acid (4.53%) (Wuttisin, 2017). Some amount of oleic acid or ω9 (8.7–9.6%) was also detected (Chirinos et al., 2013; Follegatti–Romero et al., 2009; Guillén et al., 2003). It also contains essential amino acids such as cysteine, tyrosine, threonine, and tryptophan as well as vitamin E, polyphenols and minerals (Wang, Zhu & Kakuda, 2018). Inca peanut oil is available as edible oil and often used with pharmaceutical products and cosmetics. Fresh leaves, dried leaves, roasted leaves and commercial tea leaves were extracted

with hot water and revealed the presence of phenols, flavonoids, tannin, cardiac glycosides, steroids and terpenoids (Wuttisin, Nararatwanchai & Sarikaputi, 2021). The previous study also found that roasted leaves extract with hot water exhibited the highest phenolic content and exhibited the most potent antioxidant activity. Thus, the objectives of this research were to compare the phytochemicals, total phenolic contents and antioxidant activities of Inca peanut leaves extracts at different conditions. Fresh and dried leaves were extracted with water or 95% ethanol at 30°C and 60°C. The extracts were compared for their phytochemical constituents, total phenolic contents and antioxidant activities. The data might be provided more detail about the suitable extraction methods to achieve the active compounds in Inca peanut leaves which might be useful for supporting the utilization of Inca peanut leaves as antioxidant applications.

MATERIALS AND METHODS

1. Plant Material

Inca peanut was grown in Nanglae, Chiang Rai, Thailand. Inca peanut leaves (5 months old) were harvested during March 2020.

2. Chemicals

Gallic acid, Folin–Ciocalteu's reagent, 2,2–diphenyl–1–picrylhydrazyl (DPPH), 2,2–azinobis (3–ethylbenzothiazoline 6–sulfonic acid) (ABTS) and ascorbic acid were purchased from Sigma–Aldrich Chemical Co. (St. Louis, MO, U.S.A.). Other chemicals and reagents used in this study were analytical grade.

3. Preparation of leaves extracts

Dried leaves were prepared by air dried under shade for 2 days and in hot air oven (60°C) for 48 h. The leaves (fresh and dried) were cut into small pieces by blending in the blender (Sharp/ EM–11). Fresh and dried leaves were extracted with deionized water or ethanol 95% at a ratio of 1 : 10 (w/v) by incubated in incubating shaker (8 h) resulting in eight extracts: fresh leaves extracted with water (30°C) (FW30), fresh leaves extracted with warm water (60°C) (FW60), fresh leaves extracted with ethanol 95% (30°C) (FE30), fresh leaves extracted with warm ethanol 95% (60°C) (FE60), dried leaves extracted with water (30°C) (DW30), dried leaves extracted with warm water (60°C) (DW60), dried leaves extracted with ethanol 95% (30°C) (DE30) and dried leaves extracted with warm ethanol

95% (60°C) (DE60). The extracts were then filtered through Whatman® paper No.1 and dried using freeze dryer (Labconco). The percentages of yield were calculated by following equation (1):

$$\% \text{ Yield} = [\text{Mass of dried extract (g)} / \text{Mass of leaves (g)}] \times 100 \quad (1)$$

4. Phytochemical screening

The extracts were dissolved in water or ethanol (5 mg/ml) and determined for their phytochemical constituents according to several previously published standard protocols.

4.1 Screening for phenols

The 3–4 drops of 5% ferric chloride solution was added into test tube containing 1 ml of extract and observed for the purple color development (Harborne, 1973).

4.2 Screening for saponins

A 2 ml of distilled water was added into test tube containing 2 ml of extract. The mixture was shaken vigorously for 2 min and warmed. The formation of stable foam was observed (Banso & Adeyemo, 2006).

4.3 Screening for flavonoids

4.3.1 Alkaline reagent test

A few drop of 10% sodium hydroxide solution was added into 1 ml of the extract. The intense yellow color indicates the presence of flavonoids (Tiwari et al., 2011).

4.3.2 Lead acetate test

A few drop of 10% lead acetate solution was added into 1 ml of the extract. The white or yellow precipitate indicates the presence of flavonoids (Bargah, 2015).

4.4 Screening for steroids

According to Salkowski's test, 1 ml of the extract was mixed with 1 ml of chloroform and 1 ml of concentrated sulfuric acid. The red color in the lower chloroform layer indicates the presence of steroids (Joshi, Bhobe & Saatarkar, 2013).

4.5 Screening for alkaloids

Potassium iodide (2 g) and iodine (1.27 g) were dissolved in distilled water (5 ml) and the solution was diluted to 100 ml with distilled water. Few drops of this solution were added into 1 ml of the extract. The brown precipitate indicates the presence of alkaloids (Abdullahi, Iyas & Ibrahim, 2013; Joshi, Bhobe & Saatarkar, 2013).

4.6 Screening for glycosides

Leave extract (0.5 g) was shaken with distilled water (5 ml). Glacial acetic acid (2 ml) containing a few drops of ferric chloride was added, followed by concentrated sulfuric acid (1 ml) along the side of test tube. The formation of brown ring at the interface give positive indication for cardiac glycoside and a violet ring may appear below the brown ring (Ayoola et al., 2008).

5. Micro X–Ray Fluorescence scanning

Inca peanut leaves were analyzed for elemental distribution by using Micro X–Ray Fluorescence (M4 Tornado, Bruker Nano GmbH, Berlin, Germany). This machine was equipped with two XFlash® silicon drift detectors (type SDD VH50P). It was operated at 50 kV and 599 µA. The X–ray beam coming from the excitation source is size defined by a focusing or collimating primary optic element. Once it excites the sample placed on a sample holder, the X–ray florescence energy is discriminated and counted by a detector (Rodrigues et al., 2018).

6. Determination of total phenolic contents

Total phenolic content was determined by spectrophotometry using Folin–Ciocalteu's assay (Waterman & Mole, 1994). A 20 µl of leaves extracts (2 mg/ml) was added with 100 µl of Folin–Ciocalteus reagent and 80 µl of 200 g/l sodium carbonate, consecutively. The mixture was incubated at ambient temperature for 1 h. The absorbance was measured with microplate reader at 765 nm (SPECTRO Star Nano Microplate Reader, BMG Labtech). Gallic acid was used as the reference standard. A calibration curve of gallic acid was prepared in the range of 1 to 10 µg/ml. The result was expressed as mg gallic acid equivalent per g dry weight (mg GAE/g DW).

7. Determination of antioxidant activities

7.1 The ABTS radical cation scavenging activity

The ABTS scavenging activity was performed according to the method of Re et al. (1999) with some modifications. The stock solution ABTS cation chromophore was prepared by the reaction between 7 mM ABTS solution (100 ml) and 2.45 mM potassium persulfate (final concentration) (100 ml) in the dark place at ambient temperature for 16 h. The ABTS solution was diluted with phosphate buffer, PBS (50 mM, pH 7.4) to an absorbance of 0.80 at 734 nm. The extracts were prepared at various concentrations (1.0, 0.5, 0.25 and

0.125 mg/ml). An aliquot of each extract (20 µl) was added to 180 µl ABTS solution. A mixture containing 20 µl of PBS solution and 180 µl ABTS solution was used as control. The resulting mixture was then incubated for 30 min at ambient temperature. The absorbance was measured at 734 nm with microplate reader (SPECTROStar Nano Microplate Reader, BMG Labtech). Ascorbic acid was used as the reference standard. The percent inhibition of ABTS radical cation was calculated by the following equation (2):

$$\% \text{ inhibition} = [(A \text{ control} - A \text{ sample or standard}) / A \text{ control}] \times 100 \quad (2)$$

Where A control is the absorbance at 734 nm without extract or standard. A sample or standard is the absorbance at 734 nm with extract or standard. The calibrations curve between percent inhibition and ascorbic acid concentration was established. The ABTS radical cation scavenging activity of the extract was expressed as mg ascorbic acid equivalent antioxidant capacity per g dry weight (mg AEAC/g DW) and IC50 value (µg/ml), indicating the concentration of extract scavenge 50% of ABTS radical cation.

7.2 The DPPH radical scavenging activity

The DPPH radical scavenging activity was determined according to the method of Gülçin et al. (2010). The extracts were prepared at various concentrations (1.0, 0.5, 0.25 and 0.125 mg/ml). An aliquot of each extract (20 µl) was added to 180 µl of DPPH radical solution. A mixture containing 180 µl of DPPH radical solution and 20 µl of 95% ethanol was used as control. After incubation in the dark place for 30 min, each mixture was measured absorbance at 517 nm with microplate reader (SPECTRO Star Nano Microplate Reader, BMG Labtech). Ascorbic acid was used as the reference standard. The percent inhibition of DPPH radical was calculated by following equation (3):

$$\% \text{ inhibition} = [(A \text{ control} - A \text{ sample or standard}) / A \text{ control}] \times 100 \quad (3)$$

Where A control is the absorbance at 517 nm without extract and standard. A sample or standard is the absorbance at 517 nm with extract or standard. The calibrations curve between percent inhibition and ascorbic acid concentration was established. The DPPH radical scavenging activity of the extract was expressed as mg ascorbic acid equivalent antioxidant capacity per g dry weight (mg AEAC/g DW) and IC50 value (µg/ml), indicating the concentration of extract scavenge 50% of DPPH radical.

8. Statistical analysis

SPSS version 23 were employed for all data analysis. One-way analysis of variance (ANOVA) Post Hoc multiple comparisons by Duncan's multiple-range test was performed to determine the significant difference ($p<0.05$) between sample groups.

RESULT

1. Inca peanut leaves extracts

The percentages yield of Inca peanut leaves extracts were shown in Table 1. DW60 provided the highest amount of the extraction yield (19.01±0.43%w/w) while FE30 provided the lowest amount of extraction yield (2.71±0.25%w/w).

Table 1 The percentage yield of Inca peanut leaves extracts and total phenolic contents

Extracts	Yield (%w/w)	Total phenolic contents (mg GAE/g DW)
FW30	5.56±0.09^{d}	10.72±1.06^{e}
FW60	5.86±0.72^{d}	12.14±0.99^{d}
FE30	2.71±0.25^{e}	19.22±1.02^{b}
FE60	11.92±0.60^{b}	15.45±0.04^{c}
DW30	10.34±0.26^{b}	10.28±0.38^{e}
DW60	19.01±0.43^{a}	12.20±0.90^{d}
DE30	10.25±1.20^{b}	18.83±1.27^{b}
DE60	7.52±0.68^{c}	22.05±1.20^{a}

Values are given as mean±S.D. from triplicate measurements.

Different letters in the same column indicate significant differences ($p<0.05$).

2. Phytochemical screening of Inca peanut leaves extracts

The phytochemical screening of Inca peanut leaves revealed the presence of phenols, flavonoids and alkaloids in all extracts as shown in Table 2.

Table 2 The phytochemical constituents of Inca peanut leaves extracts

Phytochemical	FW30	FW60	FE30	FE60	DW30	DW60	DE30	DE60
Phenols	+	+	+	+	+	+	+	+
Saponins	+	+	–	–	+	+	–	–
Flavonoids	+	+	+	+	+	+	+	+
Steroids	–	–	–	+	–	–	–	–
Alkaloids	+	+	+	+	+	+	+	+
Glycosides	–	–	–	–	–	–	–	–

+ = presence, – = absence

3. Micro X–Ray Fluorescence scanning

Inca peanut leaves were analyzed for elemental distribution by using Micro X–Ray Fluorescence. It was found that Inca peanut leaves composed of calcium (45.94%), silicon (21.76%), potassium (19.00%), magnesium (4.84%), sulfur (3.38%), phosphorus (2.74%), chlorine (1.03%), manganese (0.43%), aluminum (0.39%), titanium (0.23%), iron (0.13%), zinc (0.08%) and strontium (0.03%) as shown in Figure 1.

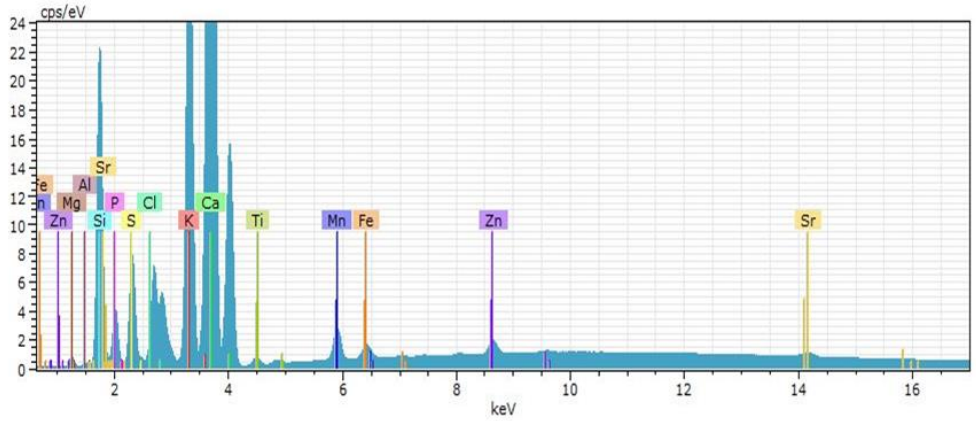

Figure 1 Elements distribution in Inca peanut leaves

4. Determination of total phenolic contents

The total phenolic contents of Inca peanut leaves extracts was determined with the Folin–Ciocalteu's reagent and expressed in term of gallic acid equivalent. The concentration of total phenolic contents was shown in Table 1. DE60 exhibited the highest contents of total

phenolic (22.05±1.20 mg GAE/g DW) while DW30 exhibited the lowest contents of total phenolic.

5. Determination of antioxidant activity

5.1 The ABTS radical cation scavenging activity

The free radical scavenging activities of the extracts were determined using ABTS radical cation scavenging assay and the results were displayed in Table 3. DW60 exhibited the highest antioxidant activity (193.07±6.14 mg AEAC/g DW, IC50 3.07±0.91 µg/ml).

Table 3 ABTS radical cation scavenging activity of Inca peanut leaves extracts

Extracts	Mean±S.D. (mg AEAC/g DW)	IC50 (µg/ml)
FW30	105.22±10.86^{C}	25.57±2.64^{c}
FW60	72.40±7.60^{d}	35.58±2.42^{e}
FE30	75.40±1.79^{d}	31.61±0.60^{d}
FE60	80.74±2.13^{d}	31.64±0.43^{d}
DW30	170.84±10.19^{b}	9.63±1.90^{b}
DW60	193.07±6.14^{a}	3.07±0.91^{a}
DE30	81.17±4.00^{d}	30.37±0.70^{d}
DE60	117.72±14.60^{c}	22.67±1.52^{c}

Values are given as mean±S.D. from triplicate measurements.

Different letters in the same column indicate significant differences ($p<0.05$).

5.2 DPPH radical scavenging activity

DPPH radical scavenging activity of the extracts were shown in Table 4. DE60, FE60, DE30 and FE30 exhibited the highest antioxidant activity (IC50 range from 119.81±4.35 to 166.58±8.17 µg/mL) while DW30 exhibited the lowest antioxidant activity (IC50 232.73±8.57 µg/ml).

Table 4 DPPH radical cation scavenging activity of Inca peanut leaves extracts

Extracts	Mean±S.D. (mg AEAC/g DW)	IC50 (µg/ml)
FW30	22.16±3.53^{bc}	181.92±41.99^{bc}
FW60	14.03±2.89^{d}	208.32±56.24^{cd}
FE30	18.84±0.68^{c}	166.58±8.17^{abc}
FE60	25.67±1.49^{ab}	126.76±8.55^{a}
DW30	10.00±0.42^{e}	232.73±8.57^{d}
DW60	22.35±2.75^{bc}	147.44±18.84^{ab}
DE30	25.52±2.69^{ab}	122.40±11.89^{a}
DE60	27.27±0.71^{a}	119.81±4.35^{a}

Values are given as mean±S.D. from triplicate.

Different letters in the same column indicate significant differences ($p<0.05$).

DISCUSSION

Fresh and dried Inca peanut leaves were extracted with water or 95% ethanol at 30°C and 60°C. The highest extraction yield was observed in dried leaves extracted with water at 60°C (DW60). This result found in accordance with the previous study that dried leaves extracted with water exhibited the highest yield when compared to fresh leaves (Wuttisin, Nararatwanchai & Sarikaphuti, 2021). The extraction yield was also increased when increasing polarity of the solvent used in extraction (Do et al., 2014). In addition, higher temperature increases solubility and simultaneously reduces surface tension of solvents which in turn contributes to higher extraction rate (Jusoh et al., 2019).

Inca peanut leaves extracts composed of phenols, flavonoids and alkaloids in all extracts. Phenol is considered the simplest class of phenolic compound. Flavonoids are the largest group of plant phenols which contain antioxidant capabilities (Saranya et al., 2017). Alkaloid can be extracted by water and ethanol (Ferrer–Serrano, Espada–Domínguez & Shasha, 2020). Alkaloids possess great health benefits against various chronic diseases. They showed anti–inflammatory, anticancer, analgesics, pain relief, antimicrobial, antifungal, and many other activities (Teng & Choi, 2012). Saponins were found in water extracts. The previous study indicated that water, as compared to ethanol, is a more suitable solvent to extract saponins (Mohaddes–Kamranshahi et al., 2019). Steroids can be found in only FE60. This

finding was the same as the previous study that found steroids in ethanolic extract of Inca peanut leaves (Nascimento et al., 2013; Shukla & Tyagi, 2017). However, steroids are non–polar so that the compounds can be extracted more perfectly in a non–polar solvent (Nuryanti & Puspitasari, 2017).

Our results were the same as the previous reports that described the presence of phenol, phenolic compounds, flavonoids, alkaloids, and steroids in Inca peanut leaves (Nascimento et al., 2013; Saavedra et al., 2010; Wuttisin, Nararatwanchai & Sarikaphuti, 2021). These phytochemicals are known to possess therapeutic activities including antimicrobial, cytotoxicity, anti–inflammatory, antitumor activity, anticarcinogenic and antioxidant with beneficial effects in the human diet. Calcium is the most abundant mineral found in Inca peanut leaves. Calcium functions as a constituent of bones and teeth. It plays a vital role in enzyme activation, hormonal release, regulation of nerve and also helps in muscle function (Soetan, Olaiya & Oyewole, 2010).

The total phenolic contents of the extracts were different depending on the type of extraction solvents and temperatures. The highest contents of total phenolic was 22.05±1.20 mg GAE/g DW from ethanolic extract at 60°C (DE60). The previous study found that roasted leaves extract with hot water exhibited the phenolic content at 21.36±1.90 mg GAE/g which lower than the present study (Wuttisin, Nararatwanchai & Sarikaputi, 2021). Ethanol has been known as a good solvent for phenolic compounds extraction. The previous study was also found that the total phenolic contents of the extracts decreased with increasing water content. The ethanolic extract may possess more phenol groups or have higher molecular weights than the phenolics in the water extract (Do et al., 2014). Then the best extracting solvent for total phenolic compounds in the present study was ethanol. Phenolic compounds are the largest category of phytochemical. They are mostly composed of flavonoids, phenolic acids, stilbenes, coumarins and tannins (Islam et al., 2015). Phenolic compounds have hydroxyl groups, which allow them to act as antioxidants by scavenging or stabilizing free radical through hydrogenation or complexion with oxidizing species (Uddin et al., 2018). Total phenolic contents could be used as a basis for rapid screening of antioxidant activity.

Inca peanut leaves extracted with water 60°C (DW60) exhibited the highest antioxidant activity when determined by ABTS scavenging assay. ABTS scavenging assay depends on the antioxidant compound ability to scavenge ABTS radical cation. This assay can

measure antioxidant capacity of hydrophilic compounds (Awika et al., 2003). The slow reaction was observed when ABTS was reacted with samples in alcohol. The result indicated that water extract will provide high antioxidant activity when determined by ABTS radical scavenging activity than ethanolic extract (Shanab & Shalaby, 2013). DW60 exhibited higher ABTS radical cation scavenging activity (IC50 = 3.07±0.91 µg/ml) than roasted leaves extract with hot water (IC50 = 37.53±3.87 µg/ml) in the previous study (Wuttisin, Nararatwanchai & Sarikaputi, 2021). The extraction time of the previous study was 30 min. Then, the increasing of extraction time in the present study may contribute to the higher antioxidant activity of the extract (Wang, Cao & Prior, 1996).

Ethanolic extracts (DE60) gave higher antioxidant activity than aqueous extracts when determined by DPPH radical scavenging method which was in agreement with the highest content in phenolics found in the ethanolic extracts (Ekin et al., 2017). The DPPH assay is known to be sensitive towards some classes of reducing species such as phenolic and polyphenolic compounds (Sahu, Kar & Routray, 2013). Since DE60 exhibited the highest total phenolic content then it also possessed the highest antioxidant activity when determined by DPPH assay. It contained the highest total phenolic compounds which are able of donating hydrogen atom to a free radical to neutralize it (Truong et al., 2019). Moreover, our findings about DE60 performances agreed with previous studies reporting that DPPH assay method seems to work well on ethanol–extracted samples (Molyneux, 2004). DPPH radical scavenging method offers the approach for evaluating the antioxidant potential of plant extract. This assay measures the ability of the plant extract to donate an electron or H^+ ion. DPPH radical accepts electron or hydrogen from antioxidant molecules to become a stable molecule resulting in a decrease in absorbance at 517 nm (Ahmed, Zara & Baig, 2013).

Therefore, with the view of being cost–effective, the water extraction of Inca peanut leaves was recommended. DW60 possessed the highest yield and exhibited the highest antioxidant activity via ABTS scavenging assay. Furthermore, other studies revealed that ethanol and water mixture were more effective in extracting phenolic compounds than water (Tomsone et al., 2012). Then further studies might be performed to use ethanol and water mixtures for the extraction of phenolic compounds from Inca peanut leaves due to the wide range of phenols that the ethanol and water mixtures can dissolve (Alothman et al., 2009).

CONCLUSIONS

The results of the present study show the presence of some phytochemicals such as phenols, flavonoids, and alkaloids in Inca peanut leaves extracts. The extracts were determined for the total phenolic contents and antioxidant activities. Dried leaves extracts with warm water (60°C) provided the highest percentage yield and the highest ABTS radical cation scavenging activity. Dried leaves extracts with ethanol 95% (60°C) provided the highest total phenolic contents and the highest DPPH radical scavenging activity. The antioxidant properties of the extracts might be due to the presence of phenolic compounds, flavonoid and other phytochemicals present in Inca peanut leaves. Thus, it can be concluded that dried leaves extracted with warm water and warm ethanol 95% may be a potential source of antioxidant for prevention and management of free radicals mediated oxidative stress. The extracts should be applied into various applications related to antioxidant such as nutritional, cosmetic and pharmaceutical products. This is an ongoing study and further work is being carried to investigate its other important biological activities.

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