

Total Phenol and Flavanoid Content Analysis of Methanol Extract of *Lepidiummeyerii* (Maca)

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ABSTRACT: *Lepidiummeyerii* is yearly herbaceous plant native to Peruvian central Andes. In this study, we studied the phytochemical properties and total phenols, total flavonoid and total saponins contents of methanol extract of maca root. All bioactive compounds like saponins, triterpenoids, phenols and flavonoids were examined of methanol extract from maca root. Total flavonoids, saponins and phenols contents were high in the methanol extract from maca root. Antioxidant activities was analyzed by (DPPH) 1,1-diphenyl-2-picrylhydrazyl radical scavenging activity. Antioxidant activities were closely related with total phenol content in the methanol extracts of maca root. Thus, the methanol extract from maca root showed higher antioxidant activities and can be used as a nutritional source with highly potential health benefits.

KEYWORDS: DPPH, Maca, Triterpenoids, Antioxidant, *Lepidiummeyerii*.

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I. INTRODUCTION

Maca (*Lepidiummeyerii*) is yearly herbaceous plant of Brassicaceae family and native to central Andes. Maca is only reported species showing a joined hypocotyl and taproot founding an underground storing organ. The plant has been used for medicinal and food since pre-Columbian periods and also declared as a “lost crop of the Incas” (NRC, 1989). Maca increased attention outstanding reports on medicinal properties in the past two decades therefore it make a worthy for the nutraceutical market (Canales et al., 2000; Dini et al., 1994). These plants could be identified by their root colors. which are purple, yellow, white, black and gray (Zhang et al., 2017). Yellow maca is commercially ideal, yellow maca was abundant in macaene and phenolic compounds. (Clément et al. 2010) Maca was recommended as a safe for consumption by the FAD in 1992. Expert researcher finding secondary metabolites and nutritional compositions of maca, found it rich protein, fat, minerals ion and amino acid and also contains secondary metabolites such as macaene, alkaloid, glucosinolate, and many other components. (B. Cui et al., 2003; M. Ganzeret al., 2002) These bioactive compounds are considered correlated to the health effects of maca. Currently research shown the medicinal ability of maca like antiproliferative function, improving fertility, improving growth rate, improving sexual performance, antipostmenopausal osteoporosis, stress tolerance and ability in vitality. (Y. Wang et al., 2007) Maca has the capability to develop energy and modify the response against oxidative stress. Previously reported that maca includes various secondary metabolites such as, alkaloids, steroid hormones, saponins and polyphenol compounds (Tang et al., 2017). Özdemir et al. (2015) investigated on *Lepidium* species comprises triterpenoid, saponins as major secondary metabolites. Like other plants, also maca plant consist several antioxidant compounds. The amounts of these substances differ according to the maca ecotype, extraction method soil composition, the time of harvest and the drying process (Campos D et al., 2013) In vitro studies, antioxidant capacity measured by several methods such as 1,1-diphenyl-2-picrylhydrazyl (DPPH), hydroxyl radical scavenging ability (HRSA), ferric reducing antioxidant potential (FRAP), lipid peroxidation inhibition ability (LPIA), and 2,2'-azino-bis(3-ethylbenzothiazoline-6-sulphonic acid (ABTS) radical scavenging capacity of secondary compounds. Recent studies also investigated the inhibitory activity of plant extracts against pathogenic bacteria. The antimicrobial resistance against pathogens through traditional antibiotics has been increased. Treatment of infectious diseases initiated by antimicrobial resistant bacteria and examine for new antimicrobial resistant agent is important and necessary.

II. MATERIALS AND METHODOLOGY

Materials

Organic maca powder was purchased from Indigo Herbs co, UK. 1 g of plant powder material was weighed and added 50 mL of methanol. The extract was filtered by Whatman No. 54 filter paper under vacuum

and drying by rotary evaporation in an Eppendorf concentrator 5301. Dry extract was redissolved in 1% DMSO containing 10 mL of deionised water.

Qualitative phytochemical Analysis

Phytochemical screening of maca root methanol extract for the presence of alkaloids, phenolic compounds, flavonoids, saponins, triterpenoids, cardiac glycosides, anthraquinones and tannins was showed by earlier defined assays. (Vesoul J et al., 2012; Arkhipov A et al., 2014; Kalt FR, Cock IE, 2014)

Total Flavonoid Content

Total flavonoid in the crude methanolic extract from maca root was carried out through spectroscopic determination. (Mottaghipisheh et al. 2018). In reaction, 150 µL of sample was incubated at 37° C for 5 min than 1.7 mL of 30% methanol, 75 µL of 0.3 M AlCl₃, 750 µL of 0.5 M NaNO₂ solution were mixed and 500 µL of 1 M NaOH was added and absorbance was recorded at 415 nm. Total flavonoid contents was calculated as quercetin standard curve equivalents (mg QE/ 100 g dry weight).

Total phenolic content

Total phenolic content of the methanolic crude extract from maca roots was examined using Folin–Ciocalteu method (Pontoni et al., 2017). In reaction, 0.5 mL of Folin–Ciocalteu reagent was added in 0.5 mL of extract and incubated at 37°C for 3 min. after incubation 1.5 mL sodium carbonate (10% w/w) solution was added. After 1 hour at room temperature, absorbance was recorded at 725 nm. Total phenolic content was calculated through gallic acid standard curve equivalents (mg GAE/100 g dry weight).

Total saponin content

The total saponin content of maca root methanol extract was determined by method of Nguyen et al. (2017). In reaction mixture contains 0.25 mL of the extract and 0.25 mL of 8% (w/v) vanillin solution were mixed and added 2.5 mL of 72% H₂SO₄ solution. The mixture was incubated at 60 °C for 15 min and using ice water bath to cooled the mixture. Absorbance was recorded at 560 nm. Escin was used as a standard. The total saponin content was calculated as escin equivalents (mg EE/ 100 g dry weight).

1,1-Diphenyl-2-picrylhydrazyl (DPPH) radical scavenging activity

The antioxidant capacity of methanol extract from maca roots was measured by DPPH (Maietta et al., 2018). Briefly, sample was dissolved in distilled water. 0.2 mL of maca extract with various concentration (1.0-5.0 mg/mL) was added to 0.8 mL of a DPPH solution. The mixture was incubated at room temperature for 15 min. and absorbance was observed at 517 nm The DPPH radical scavenging activity was calculated by the following Equation :

$$\text{Scavenging activity(\%)} = \{1 - (A2 - A1)/A0\} \times 100$$

A0: the absorbance of DPPH without sample

A1: the absorbance of sample without DPPH

A2: was the absorbance of sample and DPPH

III. RESULT AND DISCUSSION

Qualitative phytochemical screening

The dried extract was redissolved in 10 mL of deionised water. Qualitative phytochemical analysis report showed in Table 1.

Table 1: Qualitative phytochemical screenings of maca root methanol extract.

SN.	Phytochemicals Screen	Methanol Extract
1	Alkaloids	-ve
2	Phenols	+ve
3	Flavonoids	+ve
4	Cardiac glycosides	-ve
5	Saponins	+ve
6	Triterpenoids	+ve
7	Tannins	-ve
8	Anthraquinones	-ve

Total Phenolic Content

Phenolic compounds have various biological activities such as anti-atherosclerotic anti-inflammatory and anti-carcinogenic properties. These properties are closely correlated to their antioxidant activities (Talhaoui et al., 2015). Total phenol contents of methanol extract from maca roots was calculated by Folin–Ciocalteu method. The total phenol content of the methanol extract from maca root 0.20 mg GAE/ 100 g DW. Thus, the methanol extract from maca root shown rich phenolic content and can be used a potential antioxidant. Total phenol content in methanol extract of maca shown in Table 2.

Total Flavonoid Content

Flavonoids are very important portion of human nutrition because of radical scavenging capacity conferred through their hydroxyl groups. It is correlated to antioxidant activity (Sun et al., 2011). Total flavonoid contents of methanol extracts from maca root are shown in Table 2. The total flavonoids content in methanol extracts from maca root 0.02 mg QE/ 100g dry weight respectively.

Total Saponin Content

Saponins are secondary metabolites with diverse health benefits such as reducing the cardiovascular disease risk and some anti-cancer agents (Vuong et al., 2013). Saponins found in medicinal plants and plant-derived foods (Karimi et al., 2011). Total saponin contents of maca root methanol extract is shown in the Table 2. Total saponin content in maca roots 4.65 mg EE/ 100 g DW. Therefore, total saponins content the methanol extract from maca root could be a potential source of antioxidants.

Table 2. Total Phenol, Total Flavonoid and Total Saponin contents of methanol extract from Maca roots.

S.N.	Bioactive Compounds	Maca Root
1	Phenol (mg GAE/100 g dry weight)	0.20±0.19
2	Flavonoids (mg QE/100 g dry weight)	0.02±0.10
3	Saponin (mg EE/100 g dry weight)	4.65±0.38

DPPH Activity

DPPH radical scavenging activity is frequently used antioxidant activity of natural products. Antioxidant activity measured by calculating their hydrogen donating capacity (Lugasi et al., 1998). DPPH activity of maca root methanol extract shown in Table 3

Table 3. DPPH radical scavenging activity of the methanol extract from maca roots.

S.N.	Assay	Value (mg GAE/g maca)	Antioxidant activity
1.	DPPH	1.85	21.7%

IV. CONCLUSION

In this paper we conclude, the phytochemical properties analysis and total phenol content, total flavonoid content and total saponins contents in methanol extracts from maca roots was calculated and also expressed antioxidant activities of maca root methanol extracts. Total phenol (0.20 mg GAE/ 100 g dry weight), saponin (4.65 mg EE/ 100 g dry weight) and flavonoid contents (0.02mg QE/ 100 g dry weight) were evaluated in the methanol extract from maca root. The antioxidant activities of maca root was determined by DPPH radical scavenging activity assay. However, the total phenols content of maca root methanol extract was interconnected with the DPPH radical scavenging activity.

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