



In-vitro* Antioxidant Activity of Methanolic Extract of the Roots of *Bergenia ciliata

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Abstract

Bergenia ciliata is an essential medicinal plant used in regions where western medicines are inaccessible due to their unavailability and high cost. The methanolic extract of *Bergenia ciliata* roots was separated for phytochemical elements and *in-vitro* antioxidant activity. The plant extract showed the rich outgrowth of secondary metabolites that play the role for biological activities. The higher antioxidant functioning of the plant is due to the occurrence of reactive elements like phenols and flavonoids. The antioxidant functioning of the plant extract was measured by DPPH free radical scavenging assay. In DPPH free radical scavenging assay the IC₅₀ value of *Bergenia ciliata* was found to be 11.21µg/mL, while the IC₅₀ value of standard ascorbic acid was found to be 45.93µg/mL.

Keywords: antioxidant, DPPH, free radical scavenging, medicinal plants, methanolic extract.

Introduction

Bergenia ciliata is a well-known medicinal herb with thick rootstocks, 3.5 to 16.5 cm long. The plant is disseminated throughout Nepal at 1300-3000 m in moist, rocky places (Manandhar, 2002). Medicinal and aromatic plants play significant role in for livelihood health and socio-economic outlooks of the country. Most of the Nepal's population, especially tribal, ethnic groups and mountain people trusts on traditional medical applications (Ajayi *et al.* 2011). In many instances this application is transmitted orally from generation to generation and restricted to certain people (Edeogn *et al.* 2005). In present study plant sample was gathered from Manang district of Nepal to analyze its antioxidant activity and total phenol and flavonoid content. Antioxidant research is an essential topic in the medical arena as well as in the food industry. Antioxidants are

compounds that defend cells against the damaging consequences of reactive oxygen species, such as singlet oxygen, superoxide, peroxy radicals, hydroxyl radicals and peroxynitrite (Hafiza *et al.* 2002). A disproportion between antioxidants and reactive oxygen species resulted oxidative stress, followed by cellular damage. Oxidative stress has been connected to inflammation, ischemic injury, atherosclerosis, neurodegenerative diseases, aging, and cancer, (Arutselvi *et al.* 2012 & Igbinosa *et al.* 2009). Oxidants are proficient of stimulating cell division, which is a critical element in mutagenesis when a cell with a damaged DNA stand split. Thus, mutation can emerge which in turn is an essential factor in carcinogenesis. Both cigarette smoking and chronic inflammation are of the chief causes of cancer having free radical components in their

mechanism of action. Flavonoids may assist provide protection against these diseases by devoting, along with antioxidant vitamins and enzymes, to the whole antioxidant defense system of the human body. Epidemiological studies have displayed that flavonoid intake is inversely associated to mortality from coronary heart disease and to the occurrence of heart attacks (Tamilarasi *et al.* 2012). Flavonoids are most usually known for their antioxidant activity and the volume of flavonoids to act as antioxidants depends upon their molecular framework. The location of hydroxyl groups and other attributes in the chemical structure of flavonoids are essential for their antioxidant and free radical scavenging activities (Fernandez *et al.* 2004). Quercetin, the most usual dietary flavonol, is a potent antioxidant, because it has all the right structural attributes for free radical scavenging activity. It is usually assumed that constant consumption of plant extracted phytochemicals from vegetables, fruits, tea and herbs may bestow to shift the balance towards enough antioxidant status (Dehshahri *et al.* 2012). In the present study, antioxidant activity of the methanolic bark extract of *Bergenia ciliata* was assessed by using 1,1-diphenyl-2-picrylhydrazyl (DPPH) scavenging assay.

Materials and Methods

Plant materials

The plant sample was collected in month of June from the Manang district of Nepal grounded on the ethnobotanical uses. The plant sample was acknowledged at the National Herbarium and Plant Laboratories Government of Nepal, Godawari, Lalitpur.

Extraction

The plant sample was shade dried at room temperature and powdered material was then evaluated (50 g), soaked in methanol for 72h and filtered using WhatmanNo 1 filter paper. The filtrate attained was concentrated under reduced pressure in a rotatory evaporator to acquire the crude extract. The crude extract was used for further investigation of phytochemical constituents, total polyphenol content, flavonoid content and antioxidant properties.

Phytochemical screening

Phytochemical investigation of crude methanolic extracts of these medicinal plants was carried out based on the technique described on the standard protocol (Sucheta *et al.* 2011 & Saha *et al.* 2008).

Antioxidant activity test

DPPH radical scavenging activity

The free radical scavenging activity was tested by using DPPH assay. Different concentration of test samples (5, 10, 20, 30, 40, 50, 60, 70, 80, 90, 100 µg/ml) were produced while the concentration of DPPH was 0.2mM in the reaction amalgamation. These reaction amalgamations were taken in Eppendorf tubes and incubation at 37°C for 30min. Discolorations were tested at 517 nm using a UV-Visible Spectrophotometer. Percent radical scavenging activity by sample treatment was discerned by comparison with methanol treated control category; ascorbic acid was utilized as positive control. Measurement was executed at least in triplicate. The percentage scavenging of the DPPH free radical was computed using the following equation:

$$\% \text{ Scavenging Activity} = \frac{\text{Absorbance of the control} - \text{Absorbance of the test sample}}{\text{Absorbance of the control}} \times 100$$

The inhibition curve was strategized for the triplicate experiments and signified as percentage of mean inhibition \pm standard deviation and the IC₅₀ values were attained.

Results

Phytochemical screening result exhibited that, plant extract was the potent source of phytochemical constituents like polyphenols, flavonoids, alkaloids, steroids, and tannin except steroids and carotenoids.

Table: 1. Phytochemical investigation of plant extracts

| | | | |
|-------------|---|-------------------|---|
| Polyphenols | + | Reducing sugar | + |
| Steroids | - | Tannin | + |
| Alkaloids | + | Cardiac glycoside | + |
| Flavonoids | + | Anthraquinone | + |
| Terpenoids | + | Carotenoids | - |
| Glycosides | + | Saponin | + |

Key: + = Present - = Absent

Free radicals are chemical objects that can exist discretely with one or more unpaired electrons. The proliferation of free radicals can carry about thousands of reactions and thus may cause extensive tissue damage. Antioxidants can perform by converting the paired electrons to unpaired ones. The dose reliant inhibition of DPPH radical specifies that plant extract causes devaluation of DPPH radical in a stoichiometric method. The contemporary study was carried out to investigate the antioxidant activity of the methanolic plant extract of *Bergenia ciliata* barks. The DPPH radical scavenging activity (IC_{50}) of the plant extract was found to be $11.21 \pm 1.8 \mu\text{g/mL}$.

Khalaf *et al.* (2008), has reported the antioxidant activity (IC_{50}) of some medicinal plants such as *Camellia sinensis* Linn. $6.7 \pm 0.1 \mu\text{g/ml}$, *Eugenia caryophyllus* (spreng) $9.9 \pm 0.2 \mu\text{g/ml}$, *Zingiber officinale* $65.1 \pm 1.7 \mu\text{g/ml}$, *piper nigrum* Linn. $144.1 \pm 2.2 \mu\text{g/ml}$ and *Piper cubeba* Linn. $11.3 \pm 0.3 \mu\text{g/ml}$ which are found similar to the present study. Regarding the antioxidant activity of some medicinal plants. Nikolova *et al.* 2011 reported that antioxidant values (IC_{50}) of some plant extracts such as *Cardus nutans* L., *Leucosymplectum* L., *Crithmum aritimum* L., *Hedera helix* L., *Asparagus officinalis* L., *Fumaria officinalis* L. and *Daucus carota* L. has IC_{50} greater than $200 \mu\text{g/ml}$ which indicates the less potent antioxidant than that of present results. Sharma *et al.* (2015), has reported antioxidant activities of some chosen medicinal plants of Nepal and it is found that

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plants are the rich sources of antioxidant compounds.

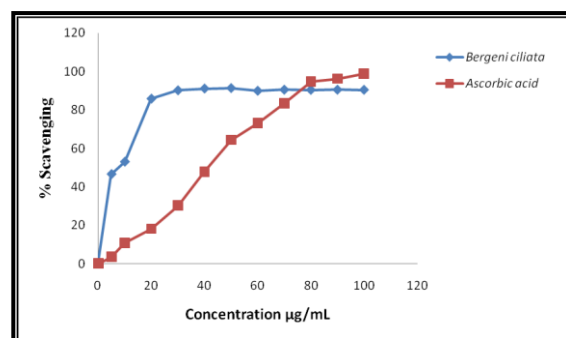


Fig 1: Percentage scavenging of DPPH free radical with plant extract and ascorbic acid

Conclusion

Phytochemical analysis showed that the root extract of *Bergenia ciliata* was a good source of secondary metabolites. The activity of free radical scavenging showed that plant extract was the potent antioxidant with IC_{50} of $11.21 \pm 1.8 \mu\text{g/mL}$. But, the IC_{50} value of standard ascorbic acid was $45.93 \mu\text{g/mL}$. It showed the sample is the potent antioxidant than the standard ascorbic acid.

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