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In vitro antioxidant activity of methanolic extracts of *Ageratum conyzoides* and *Ageratina adenophora* leaves

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Characterization of natural products based on the antioxidant activity gains tremendous interest in the past decade. In the current work, the antioxidant activity of the methanolic extract of *Ageratina adenophora* and *Ageratum conyzoides* leaves were determined. Both plants belong to the family Asteraceae and are invasive species in many tropical and subtropical countries, including northeastern India, China, Sri lanka, Nigeria. *A. conyzoides* is an annual herbaceous plant having a number of bioactive compounds. *A. conyzoides* is typically found in cultivated fields and other disturbed ecosystems; and is known for its medicinal use in the treatment of burns and wounds, arthritis, asthma, dermatitis, leprosy, malaria, and as an insecticide. Notable compounds include toxic pyrrolizidine alkaloids, phenolic acids, cumarin and polymethoxyflavones. In this study, antioxidant activity of the plant leaves was characterized for DPPH (2, 2-diphenyl-1-picrylhydrazy) and hydrogen peroxide radical scavenging activity. Antioxidant activity determines the potential of a compound to scavenge a free radical generated during oxidative stress in the body. The result suggests that *A. adenophora* and *A. conyzoides* leaves extracts showed potential antioxidant activity in terms of both DPPH and hydrogen peroxide scavenging activity.

Keywords: *Ageratina adenophora*, *Ageratum conyzoides*, antioxidant activity, DPPH, hydrogen peroxide.

INTRODUCTION

India is culturally rich in species of herbal medicine, and there are more than 2000 of such plants in record. But very few plants have been studied for pharmacological and active chemical composition that are used in treatment of various health diseases (Dursum *et al*., 2004). Recent interest has been increased in oxygen-containing free radicals in biological systems as they are causative agents of a variety of chronic disorder. Therefore, there has been a focus on the protective biochemical function of naturally-occurring antioxidants containing (Larson, 1988).

Antioxidant are important substances that have ability to protect the body from cellular damages by free radical induced oxidative stress (Tomero *et al*., 2005). Free radicals on human beings are closely related to toxicity, diseases like chronic renal failure, diabetes mellitus, cancer, immune dysfunction and aging (Halliwell and Gutteridge, 1986; Maxwell, 1995). Studies have shown that antioxidant properties of plants prevent oxidative stress defense and are thus important remediation for different human diseases including cancer, atherosclerosis, and aging process (Stajner *et al*., 1998). The presence of antioxidants in *Ageratum conyzoides* has been reported (Neelabh *et al*., 2017). The plants which exhibit antioxidant activity were mostly the phytochemical which shown presence of phenols, terpenoids, and flavonoids (Krishnaiah *et al*., 2011; Kumar *et al*., 2013).

MATERIALS AND METHOD

Plant materials

Fresh leaves of both *Ageratina adenophora* and *Ageratum conyzoides* were collected from in and around

Zemabawk campus of Regional Institute of Paramedical and Nursing Sciences, Aizawl, Mizoram, India. The identity of the plant was authenticated by a taxonomist at the Botanical Survey of India, Shillong. A voucher specimen of these plants are deposited in the Department of Pharmacy, RIPANS with Reference no: BSI/ERC/TECH/Plant Iden./2018/136

Preparation of plant extracts

The extraction of both plant metabolites were performed by procedure described in Dixit *et al*. (2005) with slight modification. The leaves were washed thoroughly with distilled water. Plant leaves were dried under shade until becomes completely dried and powdered coarsely by hand. The plant constituents are sequentially extracted using petroleum ether, chloroform, and methanol using the Soxhlet apparatus. The powdered leaves were extracted using each solvent to extract soluble matter. The extraction was continued until the extract solution became colorless. The extract was evaporated to dryness and store in refrigerator for the experiment.

Hydrogen peroxide scavenging capacity

The ability of *A. adenophora* and *A. conyzoides* methanolic extracts to scavenge hydrogen peroxide was determined with slightly modification to the method of Ruch *et al*. (1989). A solution of hydrogen peroxide (40 mM) was prepared in phosphate buffer (pH 7.5). 1 ml of the extract extract (100 µg/ml) in distilled water was added to hydrogen peroxide solution (0.6 ml, 40 mM). After 10 minute, absorbance was determined at 230 nm against a blank solution containing the phosphate buffer. The percentage of hydrogen peroxide scavenging were calculated by the formula (Keser *et al*., 2012):

$$\%\text{Scavenged } (H_2O_2) = (Ac\text{-}As)/Ac \times 100$$

Where Ac is the absorbance of the control and is the absorbance in the presence of the sample of the plant extract or standards.

DPPH radical scavenging

Sample stock solutions (1.0 mg/ml) were diluted to final concentrations of 20, 40, 60, 80, and 100 µg/ml, in ethanol. 1 ml of a 0.15nM DPPH ethanol solution was added to 2.5 ml of sample solutions of different concentrations, and allowed to react at room temperature. Ethanol (1.0 ml) plus plant extract solution (2.5 ml) was used as a blank. DPPH solution (1.0 ml; 0.15 mM) plus ethanol (2.5 ml) was used as a negative control. The positive controls were those using the standard solutions (Luciana *et al*., 2001). After 30 min, the absorbance values were measured at 518 nm and converted into the percentage antioxidant activity using the following formula:

$$\text{Radical scavenging } (\%) = (Ac\text{-}As)/Ac \times 100$$

Where Ac is the absorbance of the control, and As is the absorbance in the presence of the sample of the plant extracts or standards.

RESULTS

DPPH radical scavenging activity

Free radical scavenging activity in terms of DPPH for methanolic extract of *A. conyzoides* and *A. adenophora*

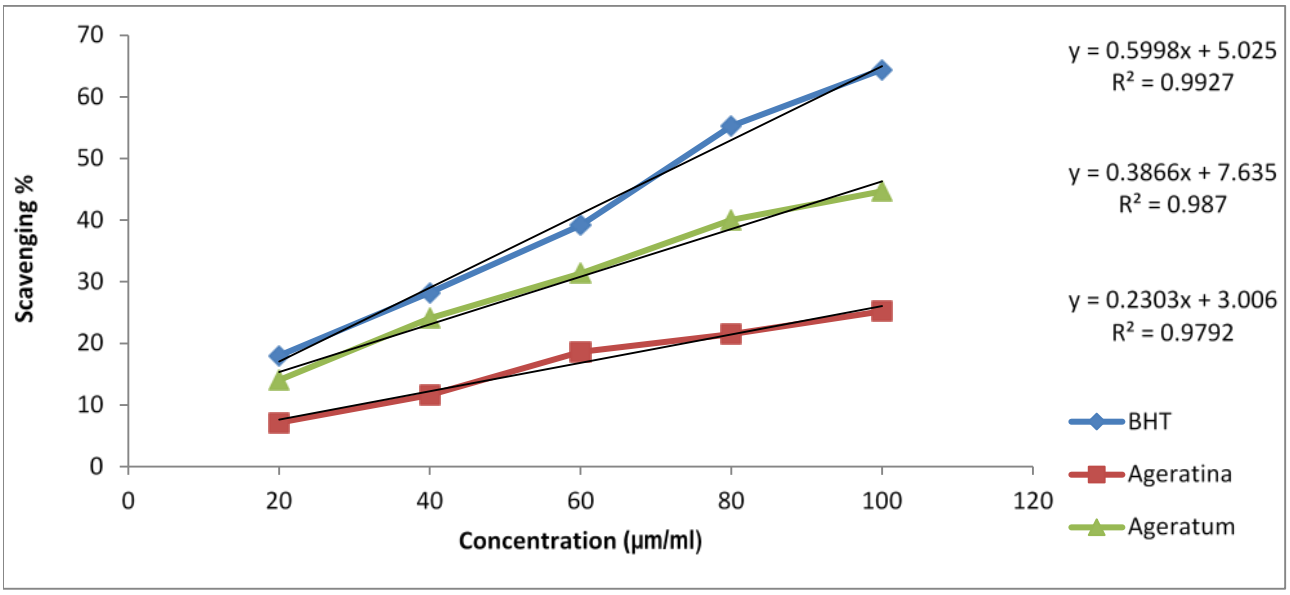

Figure 1: DPPH scavenging *of A. conyzoides* and *A. adenophora* with standard BHT.

leaves were performed and the resulta are shown in Figure 1. The IC_{50} values for plant extracts and standard butylated hydroxyl toluene (BHT) are shown in Table 1. BHT showed an IC_{50} of 68.043, *A. conyzoides* leaves extract showed IC_{50} of 70.489, and *A. adenophora* leaves extract showed IC_{50} of 92.791. Even though the leaves extract was not in pure formd it shows significant DPPH scavenging activity as compared to standard BHT. The antioxidant activity of the extract may be due to present phenol and other compound.

H_2O_2 Scavenging capacity

Table 1: IC_{50} value of methanolic plant extracts of *A. adenophora* and *A. conyzoides* with standard.

Sl.no	Sample	Part	IC_{50}
1	*A. conyzoides*	Leaves	70.489
2	*A. adenophora*	Leaves	92.791
3	BHT	Standard	68.043

The percentage scavenging capacity of methanol leaves extract *A. conyzoides* and *A. adenophora* with standard ascorbic acid are shown in the Table 2. The activity is lowest for *A. conyzoides* at 63.15%, followed by *A. adenophora* at 79.32%, and highest for ascorbic acid at 86.84% at 100 µg/ml. From this we can conclude that the scavenging capacity of both plants are comparable to that of the standard. The scavenging capacity of *A. adenophora* is very close to that of ascorbic acid), so we can say that this plant extract has high antioxidant property. Therefore, both the plant extracts can be further studied for therapeutic and other utilities.

Table 2: H_2O_2 scavenging activity of *A. adenophora* and *A. conyzoides* with standard ascorbic acid.

Sl.no.	Sample	Concentration (µg/ml)	% scavenging activity
1	Control	100	
2	*A. conyzoides*	100	63.15
3	*A. adenophora*	100	79.32
4	Ascorbic acid	100	86.84

DISCUSSION

From the study it was found that both *A. adenophora* and *A. conyzoides* possess antioxidant activity for the free radicals scavenging of DPPH and hydrogen peroxide. The antioxidant scavenges the free radicals generated in the body. The free radicals are mainly responsible for the oxidative stress in human body which lead to various diseases. The plant-based antioxidant can be utilized for various herbal formulations for mitigation and treatment because herbal formulations are cost effective as well as efficient. Thus, the antioxidant properties of these two plants can be utilized as an alternate source of treatment of various diseases (Choudhury *et al*., 2017).

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