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Evaluation of *In vitro* Anti-Oxidant properties of Selected Medicinal Plants

[1]Rabia Usman, [2]Arshad Khan, [1]Saira Gul, [1]Abdur Rauf and [3]Naveed Muhammad

[1]Institute of Chemical Sciences, University of Peshawar, Peshawar, KPK, Pakistan
[2]School of Chemistry AND Chemical Engineering Southeast University Nanjing, 211189 China
[3]Departments of Pharmacy, University of Peshawar, Peshawar, KPK, Pakistan

Abstract: In order to explore the antioxidant potential of various plants fraction of crude extract viz *n*-hexane, ethyl acetate, Chloroform and methanol and ethanol fractions/extract of three selected medicinal plants of Pakistan were been carried out. Antioxidant profile was performed before bulk extractions to evaluate antiradical capacity of selected medical plants of K.P.K, Pakistan. In present study we carried out a systematic record of the relative anti-oxidant studies of some selected medicinal plants of Pakistan. Crude ethanolic extracts and various fractions of five plants namely*, Rumex dentatus, Solonum nigrum, Berginia ciliate, Lithospermum arvense* and *Soncus asper* were evaluated for their antioxidant potential using DPPH radical scavenging assay. The result of DPPH radical scavenging assay of all the crude extracts and various fractions revealed that ethanolic crude extract of *Rumex dentatus* 100µg/ml exhibits the most powerful antioxidant activity of (50.66 %) among the entire plants. This study opens the doors for new research direction for further investigation to isolate the leads compounds responsible for such bioactivities.

Key words: Antioxidant · Radical scavenger l-1-diphenyl-2-picryl hydrazyl · Pakistan Medicinal Plants

INTRODUCTION

Antioxidants are important in the prevention of human diseases. In human bodies free radicals are responsible for many diseases. Antioxidant is a molecule that is capable of inhibiting the oxidation of other molecules. Oxidation is a chemical reaction that transfers electrons from a substance to an oxidizing agent. Oxidation reaction can produce free radicals. In turn these radicals can start chain reactions that damage cells. Oxygen, though not dangerous by itself, is involved in the generation of various kinds of "Reactive oxygen species" (ROS). Reactive oxygen species (ROS) are chemically reactive molecules containing oxygen. Primarily ROS play an important role in the host defense mechanism against micro-organisms, but the increased production of ROS is associated with the onset of a variety of diseases including cancers, inflammation, neurogeneration, Parkinson's disease, atherosclerosis [1]. The current studies showed the antioxidant potential of medical plants of Pakistan.

Bergenia ciliata, belongs to family Saxifragaceae, the genus *Bergenia* consists of about 6 species, found in the Himalayas and Central and East Asia. Two species of *Bergenia* are found in Pakistan i.e *Bergenia ciliata* and *Bergeni astracheyi*. *B. ciliata is* used as a folk medicine for the treatment of various disease is discuss previously [2]. *Rumex dentatus* belongs to family Plygonaceae. It has is commonly known as dentate dock. Traditionally it is used as bactericidal, anti-inflammatory, antitumor, astringent and anti dermatitis diuretic, cholagogue, tonic and laxative agents. Leaves, stems and roots of *Rumex abyssinicus* L. *Rumex bequaertii* L. and *Rumex usambarensis* L. are used to treat pneumonia, cough, abscesses, stomach-ache and smallpox [3]. *Solanum nigrum* Linn. (Solanacea) is a thorny shrub widely distributed in Sikkim, Uttar Pradesh, Southern India and Sri Lanka in moist places. This plant is well known in English and Tamil system as 'Black night shade' and 'Kakamachi', respectively. The berries of *S. nigrum* (Solanaceae) have been reported in the ancient Indian medicinal literature with beneficial effects in inflammation,

Corresponding Author: Rabia Usman, Institute of Chemical Sciences, University of Peshawar, Peshawar, KPK 25120, Pakistan.

tuberculosis, diuretics etc. This research was aimed to investigate the possible anticonvulsant and anti-inflammatory activities of *S. nigrum* in order to support or refute the claims by traditional herbalists in India [4]. *Lithospermum arvense,* a toxic plant belongs to the family Boraginaceae; contains pyrrolizidine alkaloids; capable of causing liver damage called also *Buglossoides arvensis*, corn gromwell. *Lithospermum arvense* L. (Field gromwell, corn gromwell) is a facultative winter annual weed of [5]. *Sonchus asper* (L.) Hill belongs to family Asteraceae, is used in various human disorder including wounds and burns, cough, bronchitis and asthma, gastrointestinal infection, inflammation, diabetes and cardiac dysfunction, kidney and liver disorders reproductive disorder like impotence (erectile dysfunction) in humans, jaundice and cancer [6].

MATERIALS AND METHODS

Plants Material: The plant materials were collected during the spring season from different parts of the Northern Province (Khyber Pakhthunkhwa) of Pakistan in March, 2010 and were confirmed by Prof Dr. Farrukh Hussain, Department of Botany, University of Peshawar.

Extract Preparation: The plant materials were dried in shade and then ground in a mortar. The powder plants materials were soaked in ethanol for three days and were then filtered using Whatt man filter paper (No 1). The filtrate was concentrated on rotary evaporator under reduced pressure to obtain a greenish gummy residue. The residue was suspended in water and fractioned with solvent of gradually increasing polarity profile starting from n-hexane, chloroform and ethyl acetate respectively as earlier discuss [7-12] and subsequently concentrating these fractions on rotary in vacuum yielding n-hexane, chloroform, ethyl acetate respectively. The dry residue and various fractions obtained was weighed and examined for their antioxidant potential.

DPPH Radical Scavenging Activity: The assay was performed according to standard protocol as earlier discuss [13, 17]. Dry crude extracts/fractions (25 mg) were taken and dissolved in distilled methanol, diluted it to prepare solution of various concentration by dilution method. 4 ml of each solution was taken and 1 ml of 0.001 M of DPPH was added. The Hydrogen atom or electron donation abilities of the consequent extracts/ fractions and standards were measured from the bleaching of the purple-colored methanol solution of 2,2-diphenyl-1-picrylhydrazyl (DPPH). All these solutions were kept in dark for 30 minutes. Also 5 ml methanol was taken and 1ml of DPPH solution was added, for control solution. At the end of the incubation period the mixtures were examined for the antioxidant activity at a wavelength of 517nm.Free radical scavenging abilities of the crudes/fractions were determined by measuring the change in absorbance of DPPH at 517nm, using the formula:

% DPPH Activity = Control –Crude Absorbance / Control x 100

RESULTS AND DISCUSSION

The crude ethanolic extract of *R. dentatus* proved to be a good source of antioxidant at tested concentration of 10, 20, 40, 60, 80 and 100 ppm with percent effect of 7.88, 10.22, 14.33, 18.55, 26.49 and 50.66 respectively. The methanolic and ethyl acetate fractions were comparatively showed good antioxidant effect, while the n-hexane and chloroform fractions were week antioxidant as shown in Figure 1. In case of *S. nigrum and B. ciliata* the methanolic fraction followed by ethyl acetate fraction were proved moderate antioxidant as presented in Figure 1. The crude ethanolic extract and its various solvent fractions of *L. arvense* and *S. asper* showed very week antioxidant effect as clear from Figure 1.

Table 1: List of medicinal plants tested for antioxidant activity

S. No	Plant name	Family	Folk uses
01	*B.ciliata*	Saxifragaceae	Fever, diarrhea and pulmonary affection.
02	*R.dentatus*	Plygonaceae	Bactericidal, anti-inflammatory, antitumor, astringent and anti dermatitis diuretic, cholagogue, tonic
03	*S. nigrum*	Solanacea	Inflammation, tuberculosis, diuretics
04	*L.arvense*	Boraginaceae;	Liver damage
05	*S. asper*	Asteraceae	Cough, bronchitis asthma, gastrointestinal infection, inflammation, diabetis cardiac

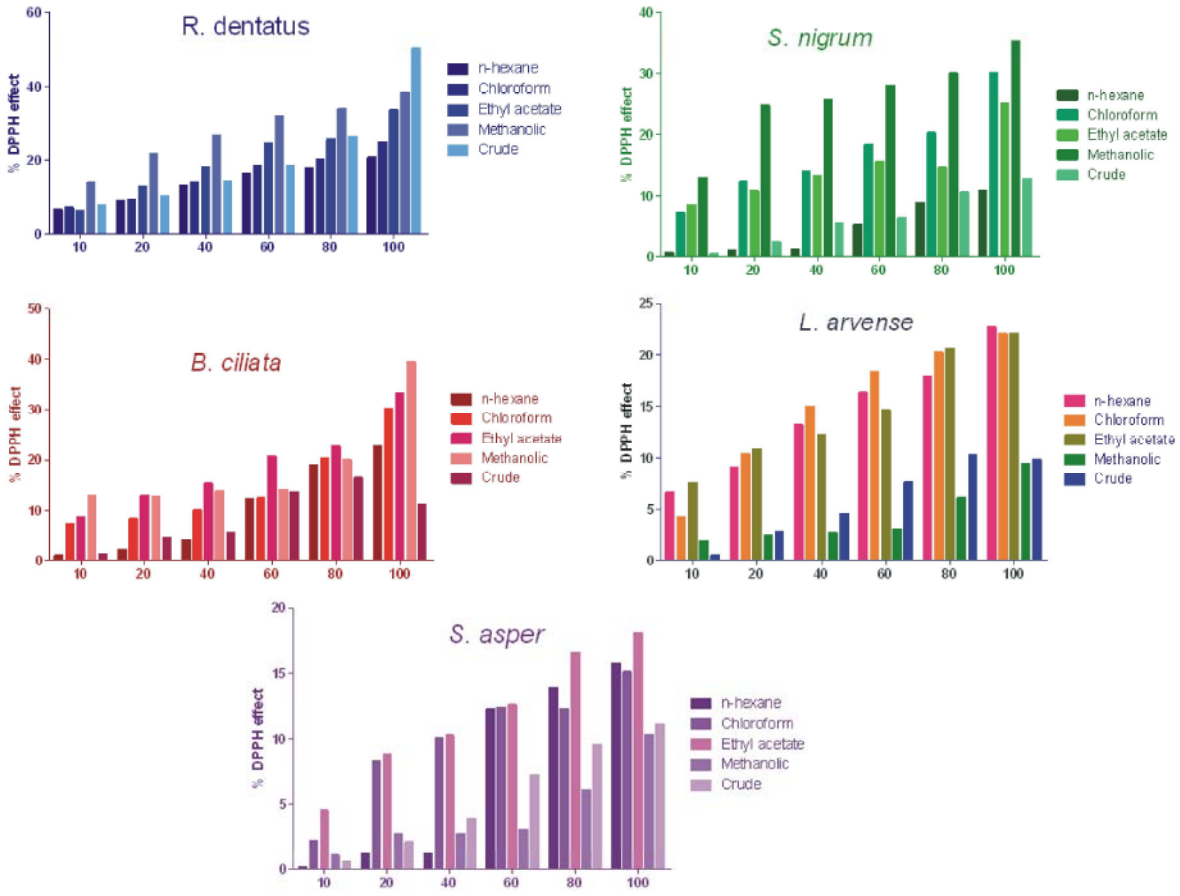

Fig. 1: Antioxidant activity of various plants at various concentrations

CONCLUSIONS

The present work showed that *R. dentatus* is rich source of antioxidant molecules while the rest of the plants exhibit moderate or no activity. It is strongly recommended through this research work that these tested plants should be used as antioxidant alone or in combination. The isolation of secondary metabolites from these natural medicines would be helpful in finding the mechanism underlying antioxidant.

Conflict of Interest Statement: We declare that we have no conflict of interest.

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