Academic Sciences International Journal of Pharmacy and Pharmaceutical Sciences

ISSN- 0975-1491 Vol 4, Issue 1, 2012

Research Article

ANTIOXIDANT AND ANTIBACTERIAL ACTIVITIES OF *TRIANTHEMA DECANDRA* LINN

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Received: 6 Sep 2011, Revised and Accepted: 6 Dec 2011

ABSTRACT

Trianthema decandra (Vellai sharuni in Tamil and *Gadabani* in Hindi) is a prostrate weed widely distributed in India. Its root is traditionally used in the treatment of hepatitis and asthma and for wound-healing. The antioxidant and antibacterial potential of *Trianthema decandra* was assessed and the results revealed significant activity in the ethyl acetate and methanolic extracts of roots and leaves of the plant.

Keywords: *Trianthema decandra*, Antioxidant, Antibacterial, Phenolics, DPPH scavenging.

INTRODUCTION

Medicinal plants are gifts of nature to cure limitless number of diseases in human beings[1]. The abundance of plants on the earth's surfaces has led to an increasing interest in the investigation of different extracts obtained from traditional medicinal plants as potential sources of new therapeutic agents[2]. Antioxidants are a group of compounds that facilitate survival of plants and may promote the health of humans who consume a variety of plant foods[3]. In plants, the term 'antioxidant' refers to a wide range of phenolic compounds that vary from simple phenolic acids to highly polymerized compounds such as tannins. Phenolic compounds or polyphenols are categorized into 15 main classes with more than 8,000 identified compounds. The largest category is the flavonoid group, comprising 13 classes with over 5,000 compounds[4]. In plants, polyphenols are important for structural supports, as antiherbivorous substances, attracting pollinators, for protection from ultraviolet radiation and for wound repair[5]. The human body also synthesizes endogenous antioxidants such as superoxide dismutases, glutathione peroxidases, alpha-tocopherol and melatonin to counteract cellular damage by active oxygen and free radicals[6]. Many studies suggest that endogenous antioxidants or exogenous antioxidants from diet can function as free radical scavengers and improve human health. In this regard, consumption of a variety of plant foods may provide additional health benefits[6, 7].

The use of plant extracts and phytochemicals with known antimicrobial properties can be of great significance in therapeutic treatments. In the last few years, a number of studies have been conducted to prove such efficiency[8-10]. Many plants have been used because of their antimicrobial traits, which is due to compounds synthesized as secondary metabolites by the plant. These products are known by their active substances, for example, the phenolic compounds which are part of the essential oils[11], as well as tannins[12].

Trianthema decandra (Family: *Aizoaceae*) is a prostrate, glabrous, succulent and annual weed found almost throughout India, in cultivated and waste land. The genus *Trianthema* consists of 20 species, but only a few have been phytochemically investigated. It is commonly known as *gadabani* (Hindi) and *vellai sharuni* (Tamil)[13]. *Trianthema decandra* has been used in various parts of Asia, Africa, Australia and South America for curing various diseases. In some African countries, the plant is in popular use for treating skin diseases, asthma, hepatitis, wounds, fever and toothaches. In India, it is used for the treatment of ophthalmic[14]. The root applied to the eye cures corneal ulcers, itching, dimness of sight and night blindness. In orchitis, the root is ground in milk and administered[15]. The bitter roots are used for curing bacterial infections and it is also given in combination with ginger as a cathartic. The leaves contain huge amounts of vitamin C, and used to treat edema. The juice of leaves is used to treat the black quarter. The decoction of the herb is used as a vermifuge and is useful in rheumatitis. It is also used as an antidote to alcoholic poison.

The present work was carried out to evaluate the antioxidant and antimicrobial activities of the extracts of *Trianthema decandra* roots and leaves.

MATERIALS AND METHODS

Plant materials and extraction

The plant *Trianthema decandra* was collected from Namakkal district, Tamil Nadu. The plant was taxonomically identified and authenticated by the Botanical survey of India, Coimbatore (Tamil Nadu) and a voucher specimen was deposited for future reference. Around 250 g of the roots and leaves (separately) were dried in shade, pulverized by a mechanical grinder and passed through a 40-mesh sieve to get a fine powder and stored in an airtight container. The dried powder (25 g) was extracted with petroleum ether (60-80 °C), wax removed and then extracted with ethyl acetate, 80% methanol and water sequentially in a soxhlet apparatus. The solvents from various extracts were then concentrated in rotary evaporator at reduced pressure below 40 °C.

Chemicals

1,1-diphenyl-2-picryl-hydrazyl (DPPH), butylated hydroxy toluene (BHT) and catechol were purchased from Sigma, St. Louis, MO, USA. All other chemicals and reagents were of analytical grade.

Bacterial strains

To study the antibacterial activity of various extracts of *Trianthema decanda* roots and leaves, the strains of bacteria were obtained form Microbiology Laboratory at PSG Hospitals, Coimbatore. The selected bacteria included *Staphylococcus epidermis, Escherichia coli, Klebsiella pneumonia, Staphylococcus aureus, Enterococcus faecalis, Pseudomonas aeruginosa, Pseudomonas putida* and *Proteus vulgaris.*

Antioxidant activity

Determination of total phenolic content

Total phenol content was determined by the method adapted from Singleton and Rossi[16] with some modifications using the Folin-Ciocalteu reagent. 1 ml of the extract was mixed with 1 ml of Folin-Ciocalteu's phenol reagent (1:10). After 3 min, 1 ml of saturated sodium carbonate (35%) was added to the mixture and it was made up to 10 ml by adding deionised distilled water. The mixture was kept for 90 min at room temperature in the dark. The absorbance was measured at 725 nm against the blank. The total phenolic content is expressed as mg of Catechol equivalents (CE) per g of dry extract.

Determination of DPPH free radical scavenging activity

The scavenging effect of the extracts on DPPH radicals was determined according to the method adapted from Shimada *et al.*[17]. Various concentrations of sample (1.5 ml) were mixed with 3 ml of

200 μM DPPH solution. The mixture was shaken vigorously and allowed to stand for 40 min, and the absorbance was measured at 517 nm, using butylated hydroxy toluene (BHT) as the standard. The percentage of inhibition was calculated according to the formula: $[(A_{control} - A_{sample})/A_{control}]x100$. IC_{50} values were determined by linear regression.

Determination of Antibacterial activity by disc diffusion method

The *in vitro* antibacterial activity of the sample solutions was studied by disc diffusion method. Plates were prepared by pouring 20 ml of sterile nutrient agar (Hi-media) into sterile Petri dishes and were inoculated with a loopful broth culture of each organism. Sterile discs (6 mm dia) impregnated with 20 μl (1.5 mg/disc) various extracts dissolved in dimethyl sulphoxide (DMSO) were air-dried and placed on the agar plates. The plates were incubated at 37 °C for 24 h. Control studies with polymixin-B and rifampicin 30 μg were used as antibacterial standard drugs. The control experiments with solvent DMSO were done concurrently[18]. The results were recorded by measuring the zones of growth inhibition surrounding the disc. Clear inhibition zones around the discs indicate the presence of antibacterial activity. All data regarding antibacterial activity are the average of triplicate analyses.

RESULTS AND DISCUSSION

Antioxidant activity

Total phenolic content

The total phenolic content of the various extracts of *Trianthema decanda* roots and leaves is given in Table 1. The ethyl acetate and methanolic extracts of both the roots and leaves showed higher levels of total phenolic contents than the petroleum ether and aqueous extracts. Phenols are very important plant constituents because of their scavenging ability due to their hydroxyl groups[19]. The imbalance between reactive oxygen species (ROS) and antioxidant defense mechanism leads to oxidative modifications in cellular membrane or intracellular molecules. Under pathological conditions, ROS are overproduced and results in lipid peroxidation and oxidative stress[20]. Antioxidant-based drugs or formulations are used for the treatment of complex diseases like atherosclerosis, stroke, diabetes, Alzheimer's disease and cancer[21].

Table 1: Total phenolic content (μg/ml) of the extracts of *Trianthema decandra* roots and leaves

Extract	Petroleum ether	Ethyl acetate	Methanol	Aqueous
Trianthema decanda roots	5.40 + 0.15	32.50 + 1.00	40.00 + 1.20	5.00 + 0.15
Trianthema decanda leaves	5.00 + 0.15	31.5 + 0.95	45.00 + 1.35	20.00 + 0.60

Values are expressed as mean + SD (n=3) as Catechol equivalents (CE).

DPPH free radical scavenging activity

The DPPH is considered to be a model of lipophilic radical. A chain reaction in lipophilic radicals was initiated by lipid autooxidation. The radical scavenging activity of *Trianthema decanda* roots and leaves extracts was determined from decrease in absorbance at 517 nm due to scavenging of stable DPPH free radical. The scavenging effects of *Trianthema decanda* roots and leaves extracts and BHT are given in Table 2. The extracts had significant scavenging effects on the DPPH radical. The positive DPPH test suggests that the samples are free radical scavengers. The IC_{50} values of the extracts in the DPPH free radical scavenging assay are illustrated in the Fig 1. These results indicate that the *Trianthema decanda* roots and leaves extracts, particularly the ethyl acetate and methanolic extracts, exhibit the ability to quench the DPPH radical, suggesting that the extracts are good antioxidants with radical scavenging activity.

Table 2: DPPH free radical scavenging assay of *Trianthema decandra* roots and leaves

Extract	% Inhibition			
	Petroleum ether	Ethyl acetate	Methanol	Aqueous
Trianthema decanda roots (1 mg/ml)	0.95 + 0.03	92.79 + 2.80	54.95 + 1.65	12.06 + 0.35
Trianthema decanda leaves (1 mg/ml)	46.85 + 1.40	61.26 + 1.85	81.08 + 2.45	77.48 + 2.30

Values are expressed as mean + SD (n=3)

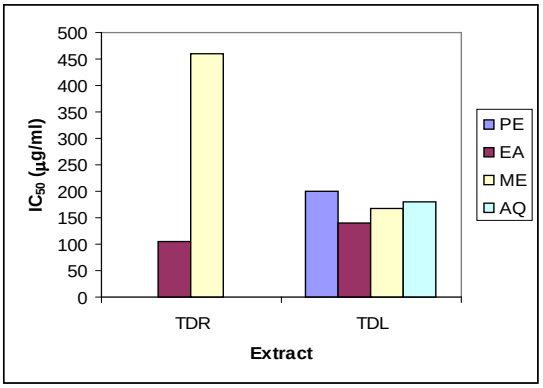

Fig. 1: IC_{50} values for DPPH free radical scavenging activity of *Trianthema decandra* roots and leaves

TDR - *Trianthema decanda* roots extracts

TDL - *Trianthema decanda* leaves extracts

PE – Petroleum ether extract

EA – Ethyl acetate extract

ME – Methanolic extract

AQ – Aqueous extract

Phenolic compounds may contribute directly to the antioxidative action[22]. The results indicate a strong relationship between total phenolic contents and radical scavenging activity, suggesting that phenolic compounds are responsible for the antioxidative properties of *Trianthema decanda* roots and leaves extracts. Phenolic compounds are reported to be effective hydrogen donors, which makes them good antioxidants[23, 24].

Antibacterial activity

The antibacterial activities of various plants have been reported by many researchers[25]. As the plants produce secondary metabolites in order to protect themselves from microorganisms, herbivores and insects, this antimicrobial effect is somehow expected from plants. Plants are important sources of potentially useful structures for the development of new chemotherapeutic agents. The first step towards this goal is the *in vitro* antibacterial activity assay[26]. Many reports are available on the antibacterial properties of plants[27, 28].

In the present study, the methanolic extract of *Trianthema decanda* roots was found to be effective against *S. epidermis, K. pneumonia, S. aureus, E. faecalis,* and *P. vulgaris*, while the ethyl acetate extract of roots was active against *K. pneumonia, S. aureus, P. aeruginosa* and *P. vulgaris* (Table 3). But petroleum ether extract of the roots was found to be effective against *K. pneumonia, S. aureus* and *P. vulgaris,* while aqueous extract was active only against *S. aureus.* Of the extracts of *Trianthema decanda* leaves, the ethyl acetate extract of *Trianthema decanda* showed inhibitory activity against all the bacteria tested. The methanolic extract of leaves was found to be active against all other organisms except *S. epidermis* and *E. coli.* While the aqueous extract of leaves showed mild activity against *P. aeruginosa,* the petroleum ether extract was found to be ineffective against any of the bacteria tested.

Table 3: Antibacterial activity of *Trianthema decandra* roots and leaves

Extracts	Zone of inhibition (mm dia)									
	Petroleum ether		Ethyl acetate		Methanol		Aqueous		Standards	
Organisms	TDR	TDL	TDR	TDL	TDR	TDL	TDR	TDL	PM	RF
Staphylococcus epidermis	-	-	-	25	10	-	-	-	30	18
Escherichia coli	-	-	-	25	-	-	-	-	28	11
Klebsiella pneumonia	10	-	14	20	20	22	-	-	28	30
Staphylococcus aureus	16	-	20	26	20	20	22	-	38	25
Enterococcus faecalis	-	-	-	20	14	30	-	-	35	30
Pseudomonas aeruginosa	-	-	8	30	-	20	-	10	10	18
Pseudomonas putida	-	-	-	22	-	8	-	-	36	10
Proteus vulgaris	18	-	20	20	20	8	-	-	24	4

Values are expressed as mean (n=3)

TDR - *Trianthema decanda* roots extracts

TDL - *Trianthema decanda* leaves extracts

PM – Polymyxin – B

RF – Rifampicin

The ethyl acetate and methanolic extracts of both the roots and leaves of *Trianthema decandra* showed good antibacterial activity against most of the tested bacteria, suggesting the presence of antibacterial agents in the plant.

Taking into account the total phenolic contents, DPPH free radical scavenging and antibacterial activities, the ethyl acetate and methanolic extracts of both the roots and leaves of *Trianthema decandra* showed higher levels compared to the petroleum ether and aqueous extracts. Antioxidants that retard the oxidation process may additionally exhibit antimicrobial activity[29]. Thus there is correlation between the antibacterial activity and the antioxidant activity of the *Trianthema decandra* extracts. Our results suggested that both the roots and leaves of *Trianthema decandra* could be a promising source of natural antioxidants, as in ripe Pepino furit[30]. Some medicinal plants are potent antioxidants and may be efficient as preventive agents in many diseases. The World Heath Organization has also recognized the importance of traditional medicine and has created strategies, guidelines and standards for botanical medicines. Proven agro-industrial technologies need to be applied to the cultivation and processing of medicinal plants and the manufacture of herbal medicines[31].

CONCLUSION

The present results reveal that the ethyl acetate and methanolic extracts of roots and leaves of *Trianthema decandra* possess antioxidant and antibacterial potential, by virtue of their phenolic contents, DPPH free radical scavenging and antibacterial activities. The results also revealed that the leaves have more antioxidant and antibacterial potential than the roots. The broad spectrum antibacterial activity of the above extracts may not be due to a single phytomolecule, but may be due to the presence of a number of bioactive metabolites.

ACKNOWLEDGEMENT

The authors are thankful to the management, K.S. Rangasamy College of Technology, Tiruchengode, India, for providing the fund and facilities.

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