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Research Article

IN VITRO ANTIOXIDANT ACTIVITY OF AQUEOUS FRUIT EXTRACT OF *PEDALIUM MUREX*.

Jalaram H. Thakkar [a]* Ajit N. Solanki[b], Mehul H. Thakkar[c], Himanshu K.Solanki[a], Natvarlal J. Patel[d]

[a]Department of pharmacy, SSR College of pharmacy, Sayli road, Silvassa, UT of D & NH, INDIA,
[b] Lambda Therapeutic Research Ltd, Near Gujarat High Court, S. G. Highway, Gota, Ahmedabad 380061, Gujarat, India
[c] Sun Pharmaceutical Industries Ltd, C1/2710 GIDC Phase-III, Vapi - 396 195, Gujarat.
[d] Department of Pharmacology, S. K. Patel College of pharma. Edu. & research, Ganpat University, Kherva, Gujarat, India.

ABSTRACT

Free radicals are involved in more than 80 diseases including Diabetes mellitus, arthritis, cancer, ageing. etc. in addition the free radicals are also play role in the pathogenesis of liver toxicity, peptic ulcer, diabetic nephropathy etc. In treatment of theses diseases, antioxidant therapy play key role. Current research is now directed towards finding naturally occurring antioxidant of plant origin. In Indian system of medicine *Pedalium murex (Bada Gokharu)* is an important medicinal plant and it has been used traditionally in various disorders and as a health tonic. To understand the mechanisms of pharmacological actions, the in vitro antioxidant activity of aqueous extract of *Pedalium murex* was investigated for DPPH scavenging activity and superoxide scavenging activity. Percentage inhibition of free radicals was measured. The antioxidant property may be related to the phenolic acids and micronutrients present in the extract. Results clearly indicate that *Pedalium murex* is effective free radical scavenger.

KEYWORDS - Antioxidant activity, *Pedalium murex*, free radical scavenging.lipschitz

INTRODUCTION

Majority of the diseases/disorders are mainly linked to oxidative stress due to free radicals[1]. The most common reactive oxygen species (ROS) include superoxide ($S0_2$.) anion, hydrogen peroxide (H_20_2), peroxyl (ROO-) radicals, and reactive hydroxyl (OH.) radicals. The nitrogen derived free radicals are nitric oxide (NO.) and peroxynitrite anion (ONOO.). ROS have been implicated in over a hundreds of diseases states which range from arthritis and connective tissue disorders to carcinogenesis, aging, physical injury, infection and acquired immunodeficiency syndrome [2,3]. In treatment of these diseases, antioxidant therapy has gained an immense importance. Current research is now directed towards finding naturally occurring antioxidants of plant origin. Antioxidants have been reported to prevent oxidative damage by free radical and ROS, and may prevent the occurrence of disease, cancer and aging. It can interfere with the oxidation process by reacting with free radicals, chelating, catalytic metals, and also by acting as oxygen scavengers [4,5]. Plant and plant products are being used as a source of medicine since long. The medicinal properties of plants have been investigated in the recent scientific developments throughout the world, due to their potent antioxidant activities, no side effects and economic viability [6]. Novel natural antioxidants from some plants have been extensively studied in the past few years for their antioxidant and radical scavenging properties [7,8,9].

Pedalium murex belongs to family Pedaliaceae is a herb mainly found near the sea coasts of South India, Gujarat, Konkan, Mexico and Tropical Africa. [10,11] The species found also in Delhi, Rajasthan and Punjab. In Hindi it is known as bada gokharu. Fruits of the plant are available in market for medicinal purpose. A variety of chemicals have been isolated and characterized from plant *P. murex.* They are classified under flavonoids, tri-terpenoids, lipids, steroids, phenolic acids, carbohydrates and amino acids. Specially fruits contain alkaloids, flavonoids (pedalitin and dinatin). Fruits are considered demulcent, diuretic, antispasmodic, antiseptic and aphrodisiac. Juice of fruit is believed to dissolve the kidney stone [12,13,14.15].

*Corresponding Author **Jalaram H. Thakkar** E mail id: jay_143143@yahoo.com

The objectives of the present study were to investigate the in vitro antioxidant activity of aqueous fruit extract of *pedalium murex* fruit through the DPPH free radical scavenging activity and superoxide anion radical scavenging activity [16,17].

MATERIALS AND METHODS [16]

Chemicals

1, 1-Diphenyl–2-picrylhydrazyl (DPPH), Nitro blue tetrazolium (NBT), Ethylene diamine tetra acetic acid (EDTA), Riboflavin. Other chemicals and solvents used were of analytical grade.

Preparation of extract and stock solution of pedalium murex

The fruits of the *pedalium murex* were collected and were identified and authenticated by a botanist of B. A college of agriculture, Anand Agriculture University. The fruits were finely powdered and used for extraction. The aqueous extract of the powder was collected and stock solution having concentration 1 mg/ml was prepared in distilled water.

Preparation of test sample solution

From the stock solution the test sample solutions having concentration 100, 300, 500, 750, 1000 µg/ml were prepared.

DPPH free radical scavenging activity

Accurately weighed 4.3 mg of DPPH was dissolved in 3.3 ml of methanol in a test tube. Solution was protected from light by covering with aluminum foil. 150 µl of above solution was taken and diluted up to 3ml with methanol, the absorbance of this solution was taken immediately at 516 nm on UV spectrophotometer using methanol as blank. This reading was served as control reading. For the test and standard, the aliquots of different concentration ranging were prepared. For the assay 150 µl of the test or std solution was added to 150 µl of DPPH solution and diluted up to 3ml with methanol, the absorbance of this solution was taken after 15 min at 516 nm on UV spectrophotometer using methanol as blank. The absorbance was taken in triplicate manner.

The % Scavenging activity was found by using following formula,

$$\% \text{ scavenging activity} = \frac{\text{O.D of control} - \text{O.D of test}}{\text{O.D of control}} \times 100$$

IC50 value was found out by plotting graph of Concentration Vs % inhibition, R^2 value was also found and represented.

Super Oxide free radical scavenging activity

100 µl Riboflavin solution [20 µg], 200 µl EDTA solution [12mM], 200 µl methanol and 100 µl NBT (Nitro-blue tetrazolium) solution [0.1mg] were mixed in test tube and reaction mixture was diluted up to 3ml with phosphate buffer [50mM] The absorbance of solution was measured at 590nm on U.V spectrometer using phosphate buffer as blank after illumination for 5min.This is taken as control.

For the test solution 150 µl of test solution was taken and 100 µl riboflavin, 200 µl of EDTA, 200 µl of methanol and 100 µl of Nitro-blue tetrazolium was added. Dilute this solution up to 3 ml with phosphate buffer and illuminate for 5 min. measure the absorbance at 590nm on U.V spectrometer. % scavenging activity was founded out by using following equation.

$$\% \text{ scavenging activity} = \frac{\text{O.D of control - O.D of test}}{\text{O.D of control}} \times 100 \%$$

RESULTS:

Table 1: DPPH free radical scavenging activity of aqueous extract of fruits of *P. murex*.

Sr. No.	Substance	Concentration (mcg/ml)	% Scavenging activity	IC_{50} Value (mcg/ml)
1	**Control**	-	-	-
2	**Ascorbic acid (std)**	5	21.77 ± 0.2	10.15
		10	49.22 ± 0.9	
		50	65.33 ± 0.8	
		100	80.56 ± 0.4	
		150	82.78 ± 0.3	
	R^2 Value	0.973		
3	***P. murex***	100	24.01 ± 0.3	524.47
		300	36.32 ± 0.4	
		500	51.01 ± 0.2	
		750	66.50 ± 0.8	
		1000	73.66 ± 0.2	
	R^2 Value	0.979		

Table 2: Super oxide free radical scavenging activity of aqueous extract of fruits of *P. murex*.

Sr. No.	Substance	Concentration (mcg/ml)	% Scavenging activity	IC_{50} Value (mcg/ml)
1	**Control**	-	-	-
2	**Ascorbic acid (std)**	5	20.22 ± 0.4	9.45
		10	52.89 ± 0.6	
		50	62.33 ± 0.8	
		100	77.52 ± 0.4	
		150	83.68 ± 0.3	
	R^2 Value	0.996		
3	***P. murex***	100	18.28 ± 0.9	598.54
		300	32.33 ± 0.8	
		500	42.56 ± 0.4	
		750	63.41 ± 0.4	
		1000	72.88 ± 0.3	
	R^2 Value	0.987		

DISCUSSIONS:

Traditional medicament play an important role in our day to day life in spite of overwhelming influence of modern medicine in treatment of various disorders like diabetes, viral infection, rheumatic disease, allergic condition, obesity, respiratory diseases, cardiovascular diseases, etc. [16]

The DPPH scavenging activity of the samples was compared with reference compound, ascorbic acid. The IC50 value of aqueous extract of *pedalium murex* and ascorbic acid shows that aqueous extract of *pedalium murex* having moderate antioxidant activity compare to the ascorbic acid that shows very potent antioxidant activity.

The super oxide scavenging activity of the test sample also compared with the reference standard ascorbic acid. IC50 value of both suggests that the aqueous extract of *pedalium murex* having moderate superoxide scavenging activity and anti oxidant activity is might be due to presence of flavonoids.[17]

CONCLUSION

The aqueous extract of *pedalium murex* showed moderate type of anti oxidant activity against DPPH and super oxide free radical. Its anti oxidant activity is might be due to presence of flavonoids.

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