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Research Article

PHYTOCHEMICAL SCREENING AND FREE RADICAL SCAVENGING ACTIVITY OF *LUFFACYLINDRICA* (LINN) FRUIT EXTRACTS IN NON POLAR TO POLAR SOLVENTS

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ABSTRACT

Luffacylindrica (Linn) commonly known as sponge gourd. It is a large monoecious climber. A tendril bearing herbbelonging to family Cucurbitaceae. The plant is widely distributed throughout India. Antioxidants are vital substances which possess the ability to protect the body from damages caused by free radical induced oxidative stress. The Present study was undertaken to analyze the presence of different phytochemical constituents and to evaluate antioxidant activity of *L.cylindricafruit* in petroleum ether,benzene, chloroform, acetone, ethanol and water extracts. All the extracts was testedfor1-diphenyl-2-picryl hydroxyl (DPPH) radical scavenging activity and compared with L-Ascorbic acid as standard. The antioxidant activity of these extracts was investigated based on their ability to scavenge (DPPH) stable free radical. Phytochemical screening of *L.cylindrica* revealed the presence of carbohydrate, protein, alkaloid, tannins, saponins, alkaloids, flavonoids and resin. A higher percentage free radical scavenging was found for aqueous extract as compared to all other extracts.

Keywords: *Luffacylindrica*, phytochemical screening, antioxidants,1-diphenyl-2-picryl hydroxyl.

INTRODUCTION

The skin antioxidant defense system (ADS) is essential in protecting the epidermis from damage by free radicals generated by environmental and endogenous factors. The antioxidants counteract free radicals by removing them from body.[1] Oxidation is a chemical reaction that transfers electron from a substance to an oxidizing agent. Oxidation reaction can produce free radicals which start chain reactions that can damage cells. Antioxidant may terminate the chain reactions either by removing radical intermediates or by inhibiting other oxidation reaction by being oxidized themselves.[2]In process of aging, oxidation plays an important role in human and other animals[3]

Oxidative stress (OS) is a general term used to describe the steady state level of oxidative damage in a cell, tissue or organ caused by the Reactive Oxygen Species (ROS)[4]Oxidative stress is a stress imposed on a biological system that requires oxygen to sustain a life. Oxidative damage is a result of oxidative stress. The extent of oxidative damage depends on many factors including rate of production of semi reduced oxygen species during aerobic metabolism as well as ability of biological system to withstand oxidative stress.[5] Free radical damage is what antioxidants are supposed to take care of either by stopping new damage or by reversing earlier damage caused by free radicals[6]

Luffacylindrica (Linn) commonly known as sponge gourd. It is a large monoecious climber. A tendril bearing herb belonging to family Cucurbitaceae.[7,8] The plant is widely distributed throughout India.[9]About 20-25 fruits are harvested at maturity as indicated by the yellowing of base and apex. The average yield is reported to be 24000 fruits per acre.

Fruit is used as a traditional medicine as an anthalmetic, carminative, laxative depurative, emollient, expectorant, diuretic and lactagogue and are useful in fever, syphilis, tumours, bronchitis, splenopathy, leprosy.[10] It is used as a vegetable either prepared like squash or eaten

raw like cucumbers[11,12]. Its seeds have been used in the treatment of asthma, sinusitis and fever. The seed oil is reported to be used for skin infections in the form of tincture. The fruit used in the treatment of jaundice and billiary and intestinal colitis and also in enlarged spleen and liver. The plant is reputed to have anti tubercular and antiseptic properties.[13]

MATERIALS AND METHODS

Plant materials and extraction

The fruits of *L.cylindrica* (Cucurbitaceae) were procured from the local market of Nagpur (Maharashtra) and authenticated in Department of Botany, RashtrasantTukdojiMaharaj Nagpur University, Nagpur.

Preparation of extracts

Fruits of *L.cylindrica* were washed and cut into very small pieces and then dried under the shade at room temperature for 8 days and later dried in an oven at 45^{0}C for complete removal of moisture to obtain constant weight then subjected to size reduction.200g of air dried powered fruit material was successively extracted in soxhlet assembly by using series of solvents in increasing order of polarity viz. petroleum ether,benzene,chloroform,acetone,ethanol,water.[14]Each extract was then concentrated by distilling off the solvent and then evaporating the solvent to dryness and weighed[15]. Their percentage extractive values were recorded.

Preliminary Phytochemical Screening

All the extracts were subjected to preliminary phytochemical screening for evaluation of phytochemical constituents such as carbohydrate, protein, amino acid, alkaloids, tannins, fats and oil, flavonoids using standard procedure of analysis.[16,17,18]

Determination of antioxidant activity of *L.cylindrica* fruit extract by DPPH method[19]

The DPPH is a stable free radical and is widely used to assess the radical scavenging activity of antioxidant compound .This method is based on the reduction of DPPH in methanol solution in the presence of a hydrogen-donating antioxidant due to the formation of the non radical form DPPH-H*[20].The DPPH is reacted with methanol or absolute ethanol to yield purple color. The presence of antioxidants in the sample scavenge the formed DPPH radical and decrease in color is observed which is Spectrophotometrically measured at 517nm.[21,22] In one cuvette 3ml of methanol was taken and kept as a standard for all the extracts. In other cuvette 3ml of DPPH was taken. Absorbance for the blank samples at 517 nm was determined.[23] Cuvette of methanol was not disturbed. Now in another cuvette 3ml of DPPH was kept aside for 5min.To this cuvette ascorbic acid was added in microlitre in various concentrations. Absorbance at 517nm was read for each concentration. Scavenging activity was expressed as the % inhibition. Now ascorbic acid was replaced by extracts and followed same procedure.

The percentage of inhibition can be calculated using the formula

$$\text{Inhibition (\%)} = (A_0\text{-}A_1/A_0) \times 100$$

Where; A_0 is the absorbance of control and A_1 is the absorbance of test.

RESULTS

Extractive Value

The extractive value of petroleum ether, benzene, chloroform, acetone, ethanol and water were found to be1.33%w/w, 0.86%w/w, 0.96%w/w, 1.31%w/w, 2.13%w/w, 3.3%w/w respectively as recorded in Table no.1.Percentage yield of water extract (WECC) was found to be maximum i.e. 3.3%w/w as compared to other extracts.

Preliminary phytochemical screening

All the extracts were screened for presence of carbohydrate, protein, amino acid, alkaloid, tannin, fat and oil, flavonoid. Preliminary phytochemical screening showed the presence of carbohydrate,protein,alkaloid and flavonoid inwater extracts which is recorded in table no.2.

DPPH free radical scavenging activity

DPPH free radical scavenging activity of PELC, BELC, CELC, AELC, EELC, and WELC is depicted in fig.1.It was observed that water extract of *L. cylindrical* showed highest DPPH free radical scavenging activity then other extracts. Different concentrations of L-ascorbic acid were used as standard antioxidant.

IC_{50} Value for Antioxidant activity

IC_{50} value (Table no.3) states the amount of concentration of extract required to produce 50% free radical scavenging activity. Hence IC_{50}value is inversely related to the free radical scavenging activity. Here result clearly states that water extract of *L.cylindrica*fruit showed highest whereas Petroleum extract showed lowest DPPH free radical scavenging activity.

DISCUSSION

The phytochemical study of different extracts of *L.cylindrica* showed the presence of flavonoids

only in water extracts. Antioxidant activity of different extracts was found to be Water>Ethanol>Acetone>Chloroform>Benzene >Petroleum ether. Maximum antioxidant activity of water extracts could be contributed to presence of flavonoid.

CONCLUSION

Thus it can be concluded that *L.cylindrica* possesses antioxidant activity. Water extract possesses maximum activity while petroleum ether possesses minimum activity.

Table 1: Extractive Value % (W/W)

S.No	*L.cylindrica* Fruit Extracts	% (W/W)
1	PELC	1.33
2	BELC	0.86
3	CELC	0.96
4	AELC	1.31
5	EELC	2.13
6	WELC	3.3

Table 2: Preliminary phytochemical screening of *L. Cylindrical* fruit extracts

S.No.	Phytochemical	Test	PECC	BECC	CECC	AECC	EECC	WECC
1	Carbohydrate	Fehling test	_	-	-	-	+	+
2	Protein	Biuret test	_	-	-	-	-	+
		Xanthoprotein	_	-	-	-	-	+
3	Amino acid	Ninhydrin test	-	-	-	-	-	-
4	Alkaloid	Hager's Reagent	-	-	+	-	-	+
		Wagner's Reagent	-	-	-	-	+	+
5	Tannins	Ferric chloride reagent	-	-	-	-	-	-
		Lead acetate Test	-	-	-	-	-	-
		Potassium dichromate Test	-	-	-	-	-	-
6	Fat and Oil	Spot Test	+	-	-	-	-	-
7	Flavonoid	Shinoda Test	-	-	-	-	-	+

Table 3: DPPH free radical scavenging activity of *L.cylindrica* fruit in Non-Polar to Polar Solvents

Conc. of Extracts (µg/ml)	PELC % Inhibition	BELC % Inhibition	CELC % Inhibition	AELC % Inhibition	EELC % Inhibition	WELC % Inhibition
Control	-	-	-	-	-	-
20	2.61	2.40	4.70	6.19	10.26	11.60
40	5.74	6.88	8.67	7.99	18.30	17.51
60	8.80	8.66	11.18	18.88	24.44	26.68
80	12.43	12.31	14.52	25.07	26.34	39.02
100	17.24	21.81	19.43	29.17	30.79	46.94
120	25.39	26.09	23.92	33.89	35.02	49.05
140	27.06	28.60	29.04	39.24	42.01	55.37
160	36.46	36.11	35.21	43.54	49.31	55.80
180	37.40	44.78	40.85	48.58	55.44	58.64
200	43.46	48.47	49.73	54.03	59.57	60.62
220	46.49	51.07	52.24	58.86	63.17	68.14
240	47.46	53.86	57.05	67.26	68.04	69.30
260	49.94	58.76	59.03	68.41	68.78	70.04
280	51.72	66.17	61.42	69.35	72.06	72.46
IC_{50}	**260 µg/ml**	**210 µg/ml**	**200µg/ml**	**190 µg/ml**	**160 µg/ml**	**120 µg/ml**
IC_{50} (Std.)Ascorbic acid - 60 µg/ml						

Abbreviation: LC-*Luffacylindrica*; PE-Petroleum ether extract ; BE-Benzene extract ; CE-Chloroform extract ; AE-Acetone extract; AE-Alcohol extract; WE- Water extract

DPPH free radical scavenging activity of *L.cylindrica* fruit in Non-Polar to Polar solvents

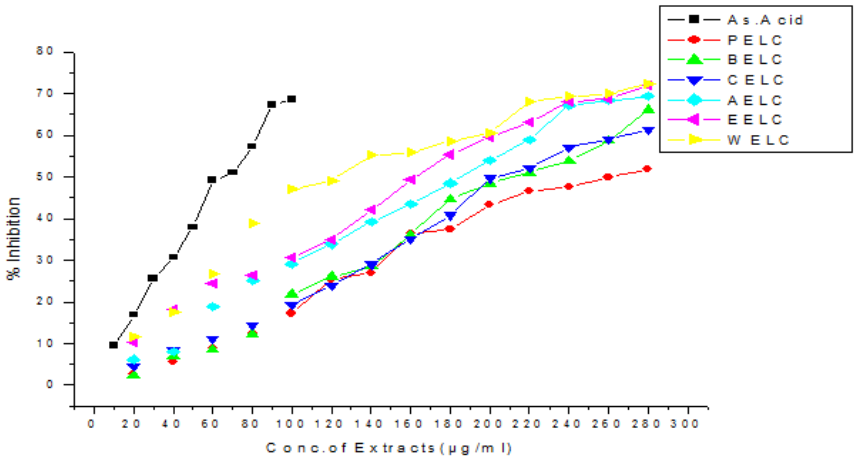

Fig. 1: DPPH free radical scavenging activity of *L.cylindrica* fruit in Non-Polar to Polar solvents

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