FREE RADICAL SCAVENGING ACTIVITY OF AQUEOUS AND ETHANOLIC EXTRACTS OF WILD AND L-ARGININE TREATED *CLEOME GYNANDRA L.*

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ABSTRACT

Cleome gynandra L. belongs to the family Cleomaceae, grows as a weed in most tropical countries. All over the world in different countries it is used to treat many diseases in their traditional systems for its remarkable nutritional and antioxidant properties. In India alone it is used by the traditional healers for many diseases e.g. .epilepsy, irritable bowel syndrome and in protozoal and worm infections etc. The present study is aimed at evaluating the DPPH (methanolic 0.1 mM) scavenging properties of aqueous and ethanolic extracts (100-500 µg/ml) of L-arginine treated and wild *Cleome gynandra* by using ascorbic acid as standard. This study revealed that ethanolic extract of L-arginine treated *Cleome gynandra* showed highest rate of DPPH scavenging activity than other extracts used.

Key Words: L-Arginine, *Cleome gynandra* L., DPPH, Anti-oxidant activity, Free radical scavenging.

INTRODUCTION

Cleome gynandra L. belongs to the family Cleomaceae is a potential medicinal plant, used as a vegetable in many African countries[1]. Phytochemical investigations of various extracts of this plant reveal the presence of metabolically active secondary metabolites such as Alkaloids, Carotenoids, Flavonoids, Phenols, Saponins, Tannins etc. most of these chemicals posses scientifically proven free radical scavenging activity[2,3,4,5]. *Cleome gynandra* was used by different scientists for anti-inflammatory, lysosomal stabilising potential and anti-oxidant potential of ethanolic leaf extract[7], antinociceptive activity from ethanolic and aqueous extract[8], antioxidant activity and radical scavenging activity[9], anti-oxidant potential of *Cleome gynandra* leaf extract on lymphoid organs in adjuvant induced arthritis in rats[10] , the roots of *Cleome gynandra* removes 'vata' stomachic, pain, ear ache, spleen enlargement and bilious fever[11] , immunomodulatory potential phagocytic activity, cell mediated and humoral immune system on albino rats[12] . All the above said uses of this plant are only because of the bioactive substances present in it. L-Arginine being the precursor of polyamines modifies the biochemical content and there by influence in antioxidant property. In the present study, we investigated the antioxidant activity of *Cleome gynandra*.

MATERIALS AND METHODS

L-Arginine Treatment

Seeds of *Cleome gynandra* were collected randomly from healthy plants and were soaked in 0.0mM to 0.5mM L-Arginine solution for 2 days then they were transferred to respective concentration labeled plastic posts filled with lateritic soil. The pots were irrigated with respective concentration of L-Arginine solution. On 30th day data were recorded.

Plant samples

Plant samples of 0.4mM treated (best yielded concentration) and wild were collected and used for further studies.

Extraction

The vegetative aerial parts of wild and L-Arginine treated samples were dried under shade and then powdered with a mechanical grinder. The dried powder of plant samples were extracted with Different solvents (Ethanol and Aqueous) in a Soxhlet apparatus. The extracts were evaporated to complete dryness by vacuum distillation.

Antioxidant Activity

The antioxidant activity of the plant extracts and the standard was assessed on the basis of the radical scavenging effect of the stable 1, 1-diphenyl-2-picrylhydrazyl (DPPH)-free radical activity by modified method (Braca A *et al.,* 2002)[3,13]. DPPH free radical scavenging activities were measured 0.2 ml extracts in various concentrations (100-500 µg/ml) of *sample*, 1ml DPPH solution (methanolic 0.1 mM DPPH) was added. Ascorbic acid was used as standard in 100-500 µg/ml solution. 0.002% of DPPH was prepared in all the solvent and 1 ml of this solution was mixed with 1 ml of sample solution and standard solution separately. These solution mixtures were kept in dark for 30 min and optical density was measured at 517 nm using Spectrophotometer. Solvent like methanol (1 ml) with DPPH solution (0.002%, 1 ml) was used as blank. The optical density was recorded and % inhibition was calculated using the formula given below (Bors W *et al.,* 1992)[14].

Percent (%) DPPH activity = (A – B / A) X 100

(Where A = optical density of the blank and B = Optical Density of the sample.)

RESULTS AND DISCUSSION

Anti oxidant activity of ethanolic and aqueous extracts of *Cleome gynandra* was determined against 1ml of methanolic 0.1mM DPPH at 517 nm by UV spectrophotometer. This study revealed that the ethanolic extract of L-Arginine Treated *Cleome gynandra* at concentration of 500 µg/ml showed high rate of free radical (DPPH) scavenging activity (30.67%) among the tested samples and followed by ethanolic extract of wild *Cleome gynandra* (27.33%) the least radical scavenging activity among the tested sample was aqueous extract of wild *Cleome gynandra* (23.48%)(Fig1,2 and Table 1,2).

All the results were compared with same concentration of Ascorbic acid as a standard which showed the highest free radical scavenging activity of (18.96%) for 500µg/ml. All the samples at same concentration showed significantly different in radical scavenging activity due to the difference in secondary metabolite content(Fig1,2 and Table 1,2). At the same time increased concentration of samples showed the increased rate of radical scavenging activity.

Free radicals were most dangerous reactive oxygen groups they cause most dangerous diseases of the world viz. cancer, diabetes, cardio vascular problems etc[7]. The plant based drugs were need of the hour for nullifying the ill effects of these free radicals. The free radical scavenging activity of Flavonoid fractions from *Cleome gynandra* against DPPH, NO, peroxinitrile, superoxide, hydroxyl radical etc was proven[3]. Antioxidant activity of phenolic and flavanoid fractions of various extracts of *Cleome gynandra* was revealed against FRAP, ABTS and DPPH[6].

CONCLUSION

The present study revealed that L-Arginine treated plant samples showed more free radical scavenging activity than their wild forms. Among the tested samples ethanolic extract of L-Arginine treated plant samples showed more free radical scavenging activity than any other tested plant extracts.

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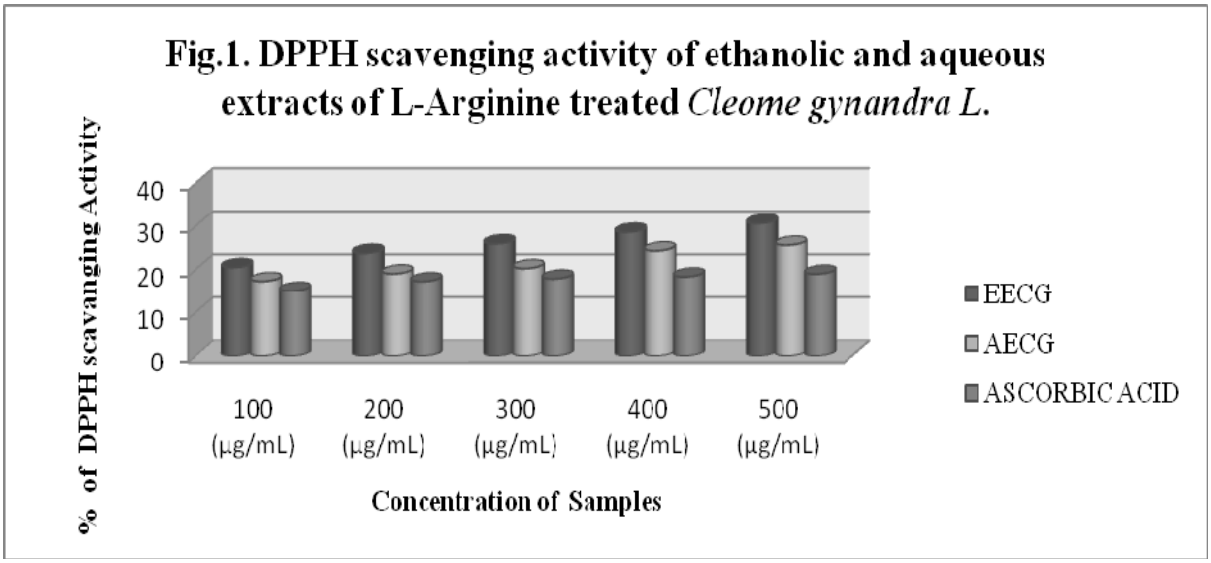
Table.1. DPPH scavenging activity of ethanolic and aqueous extracts of L-Arginine treated *Cleome gynandra L.*

S.No	Concentration of plant extract (µg / mL)	DPPH scavenging activity (%)		
		Ethanol	Aqueous	Ascorbic acid
1	100	20.49	17.26	15.00
2	200	23.77	19.02	17.23
3	300	25.96	20.24	17.86
4	400	28.63	24.31	18.32
5	500	30.67	25.62	18.96

Table.2. DPPH scavenging activity of ethanolic and aqueous extracts of wild *Cleome gynandra L.*

S.No	Concentration of plant extract (µg / mL)	DPPH scavenging activity (%)		
		Ethanol	Aqueous	Ascorbic acid
1	100	17.21	16.11	15.00
2	200	20.42	18.25	17.23
3	300	21.36	19.86	17.86
4	400	22.90	21.34	18.32
5	500	27.33	23.48	18.96

Fig.1. DPPH scavenging activity of ethanolic and aqueous extracts of L-Arginine treated *Cleome gynandra L.*



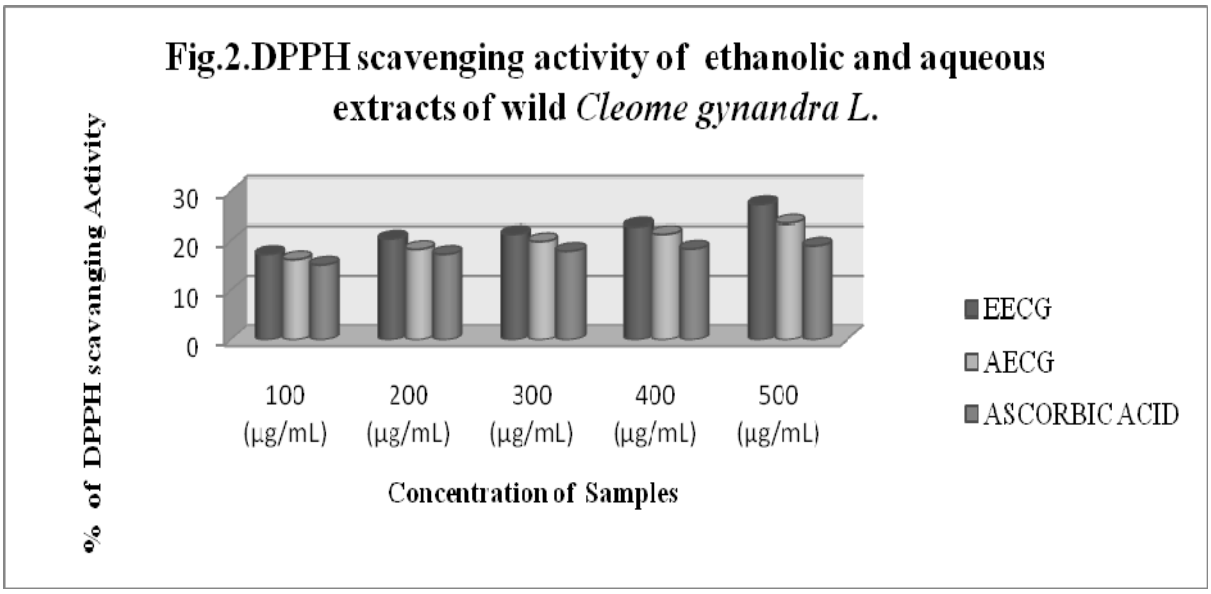


Fig.2.DPPH scavenging activity of ethanolic and aqueous extracts of wild Cleome gynandra L.
% of DPPH scavanging Activity
30
20
10
0
100 (µg/mL)
200 (µg/mL)
300 (µg/mL)
400 (µg/mL)
500 (µg/mL)
Concentration of Samples
EECG
AECG
ASCORBIC ACID