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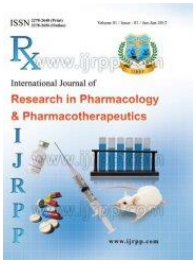

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Preliminary phytochemical screening and in vitro antioxidant activity of *commelina benghalensis L*

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ABSTRACT

Commelina benghalensis L. commonly known as **Benghal dayflower,** belongs to the family Commelinaceae. *Commelina benghalensis L.* is a perennial herb native to tropical Asia and Africa. Valaiyans of Piranmalai hills, Tamilnadu used the leaves for the treatment of rabies and wounds [1, 2] The kavirajes tribals of Bangladesh used the young leaves for external poisoning . The litertature survey of phytochemical screening of *Commelina benghalensis L* revealed the presence of tannins, phlobatannins, saponins, flavonoids, alkaloids etc.,The hydroalcoholic extract (70%) of *Commelina benghalensis L.* (Leaf) was subjected to preliminary phytochemical studies and antioxidant activity. The phytochemical screening of the hydroalcoholic extract (70%) of *Commelina benghalensis L.* (Leaf) revealed the presence of alkaloids, carbohydrates, sterols, saponins, tannins and phenolic compound, flavonoids, protein and free aminoacid, terpenoids, mucilage, betacyanin, quinone, phlobatannins, carotenoids. The inhibitory concentration (IC_{50}) of *Commelina benghalensis L.* (Leaf) against hydrogen peroxide scavenging effect was found to be 18 µg/mL in comparison with (ascorbic acid 7µg/mL). The inhibitory concentration (IC_{50}) of *Commelina benghalensis L.* (Leaf) in reducing power assay is found to be 12 µg/mL in comparison with (ascorbic acid 9µg/mL). The total antioxidant capacity is found to be 40 µg/mL in comparison with (ascorbic acid 10 µg/mL). It showed mild to moderate anti oxidant effect when compared with ascorbic acid.

Keywords: Antioxidant, *Commelina benghalensis L.*, Phytochemical

INTRODUCTION

Commelina benghalensis L. commonly known as **Benghal dayflower,** belongs to the family Commelinaceae. *Commelina benghalensis L.* is a perennial herb native to tropical Asia and Africa. Valaiyans of Piranmalai hills, Tamilnadu used the leaves for the treatment of rabies and wounds [1, 2]. The kavirajes tribals of Bangladesh used the young leaves for external poisoning [3]. The litertature survey of phytochemical screening of *Commelina benghalensis L* revealed the presence of tannins, phlobatannins, saponins, flavonoids, alkaloids,

steroids and flavonoids, carbohydrates, phytosterol, terpenoids, quinon, volatile oil, anthraquinone [4-12]. GC-MS analysis of *Commelina benghalensis L.* revealed the presence of bioactive compounds such as 3-dodecene, 1-hexadeconol, 9-eicosene and tetratriacontane, Phenol 2,4 bis(1,1 dimethyl ethyl), hexadecen1 ol trans9, 9,10 anthracenedione, tetracosane, 1,4 benzene-dicarboxylic acid, bis (2ethylhexyl) ester, 13 docosenamide, tetracosane 11 decyl [13]. The plant exhibited various pharmacological activites such as anti inflammatory activity [6], 15-lipooxygenase inhibition [10], anticoagulant activity [11], antibacterial activity [12, 16, 17], antimicrobial activity [14, 15], antiplasmodial activity, thrombolytic and cytotoxic activity and antidiarrhoeal & anthelmintic activity [18-20].

An attempt has been taken to study the preliminary phytochemical analysis for the leaves and to exhibit its anti-oxidant effect in comparison with ascorbic acid were determined.

MATERIALS AND METHODS

Collection & Authentication of plant material

Fresh leaves of *Commelina benghalensis L.* were collected from Madurai Medical College, Madurai (DT), during the month of August- 2017 and was authenticated by Dr. D. Stephen, M.Sc., Ph.D., Assistant professor, Department of Botany, American College, Madurai-20. The herbarium of this specimen was kept in the department for further reference.

Preparation of Hydroalcoholic Extract of *Commelina Benghalensis L.* (HAECB)

Procedure

The leaves were collected; shade dried was powdered coarsely and was defatted with petroleum ether (60-80˚c). The residue was dried and extracted with hydroalcohol (70%) by maceration until the complete extraction of the powder was filtered and concentrated under reduced pressure to obtain a solid residue (dark brown). (HAECB)

Phytochemical Studies

Hydroalcoholic extract of *Commelina benghalensis L.* (Leaf) was subjected to qualitative chemical analysis. The various chemical tests were performed on hydro alcoholic extract for the identification of secondary metabolites determined as per (Harborne; 1998) [21] and the results are displayed in **table 1.**

Invitro antioxidant Activity

Hydroalcoholic extract was subjected to in-vitro antioxidant studies. It includes hydrogen peroxide scavenging activity, reducing power assay and total antioxidant capacity.

Determination of Scavenging Activity of HAECB against Hydrogen Peroxide

The hydrogen peroxide scavenging was determined for the hydro-alcoholic extract of *Commelina benghalensis L.* as per **Rana MG et al., (1996)** [22]

Procedure

To 1 mL of different concentration of hydro-alcoholic extracts and ascorbic acid were treated with 3.8 mL of 0.1 M phosphate buffer solution (pH 7.4) and then 0.2 mL of hydrogen peroxide solution were added. The absorbance of the reaction mixture was measured at 230 nm after 10 min. Blank also prepared without reagents. Ascorbic acid was used as standard. The results are displayed in **figure 1&2.** And tabulated in **table 2.** The percentage inhibition of hydrogen peroxide was calculated using the formula,

% inhibition = [(Control–Test) / Control] × 100

Determination of Reducing Power Assay

The reducing power assay was determined for the hydro-alcoholic extract of *Commelina benghalensis L.* as per **Navnath et al, (2010)** [23]

Procedure

The reducing power ability of plant extracts was screened by assessing the ability of the test extract to reduce $FeCl_3$ solution as mentioned by Oyaizu *et al.*, (1986). 0.1 to 0.5 mL of plant extract solution (1 mg/mL) was mixed with 0.75 mL of phosphate buffer and 0.75 mL of 1 % potassium ferricyanide [$K_3Fe(CN_6)$] and incubated at 50^oC for 20min. About 0.75 mL of 10 % trichloro acetic acid was added to the mixture and allowed to stand for 10min. The whole mixture was then centrifuged at 3000 rpm for 10min. Finally 1.5 mL of the supernatant was removed and mixed with 1.5 mL of distilled water and 0.1mL of 0.1 % ferric chloride solution and the absorbance was measured at 700 nm in UV-Visible

Spectrophotometer. Ascorbic acid was used as standard and phosphate buffer was used as blank solution. The results are displayed in **figure 3&4.** and tabulated in **table 3.**

Determination of Total Antioxidant Activity

The total antioxidant activity was determined for the hydro-alcoholic extract of *Commelina benghalensis L.* as per **(Prieto *et al*., 1999)** [24]

Procedure

An aliquot of 0.3 mL of different concentrations of sample was treated with 2.7 mL of the reagent (H_2SO4, sodium phosphate and ammonium molybdate). In case of blank, 0.3 mL of methanol was used in place of sample. The tubes were incubated in a boiling water bath at 95°C for 90 min. The samples were cooled to room temperature, the absorbance of the aqueous solution of each concentration was measured at 695 nm against blank. The standard vitamin C was treated in a similar manner. The antioxidant activity was expressed as equivalents of ascorbic acid (µg/mL). The results are displayed in **figure 5&6.** and tabulated in **table 4.**

RESULTS AND DISCUSSION

Phytochemical screening

The phytochemical screening of the hydroalcoholic extract showed the following result and was displayed in table :1.

TABLE 1: PRELIMINARY PHYTOCHEMICAL SCREENING OF HYDRO – ALCOHOLIC EXTRACT OF *COMMELINA BENGHALENSIS L*. (LEAF) (HAECB)

S.NO	Test for phytoconstituents	Hydroalcoholic extract of *Commelina benghalensis L*. (Leaf)
1	Alkaloids	Positive
2	Carbohydrates	Positive
3	Anthraquinone Glycoside	Negative
4	Cardiac Glycosides	Negative
5	Sterols	Positive
6	Saponins	Positive
7	Tannins and Phenolic compounds	Positive
8	Flavonoids	Positive
9	Protein and Free Amino Acids	Positive
10	Mucilage	Positive
11	Quinone	Positive
12	Phlobatannins	Positive
13	Carotenoids	Positive
14	Terpenoids	Positive
15	Betacyanins	Positive

16	Emodin	Negative
17	Fixed oil & Gum	Negative
18	Anthocyanins & Lecoanthocyanins	Negative
19	Resins & Volatile oil	Negative

The phytochemical screening of the hydroalcoholic extract (70%) of *Commelina benghalensis L.* revealed the presence of alkaloids, carbohydrates, sterols, saponins, tannins and phenolic compound, flavonoids, protein and free aminoacid, terpenoids, mucilage, betacyanin, quinone, phlobatannins, carotenoids and absence of anthraquinone glycosides, cardiac glycoside, fixed oil, anthocyanin, lecoanthocyanin, emodin, gum, resins and volatile oil.

In vitro antioxidant studies

Determination of Hydrogen peroxide scavenging activity of *Commelina benghalensis L.* (leaf) (HAECB)

The inhibitory concentration (IC_{50}) of *Commelina benghalensis L.*(Leaf) against hydrogen peroxide scavenging effect was found to be 18 µg/mL in comparison with ascorbic acid 7µg/mL used as standard. The HAECB requires almost three times the concentration of ascorbic acid to scavenge the free radicals produced by hydrogen peroxide. The extract showed moderate anti-oxidant effect. The results are depicted in **figure 1 and 2** and are tabulated in

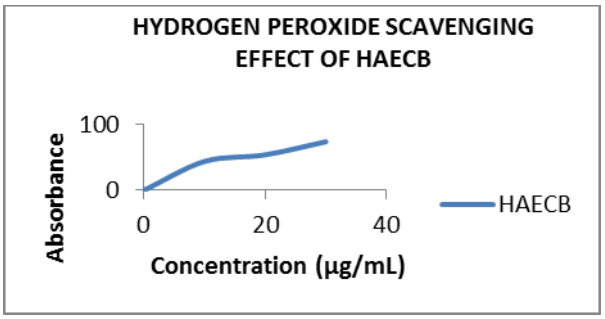


FIG. 1: Hydrogen peroxide scavenging effect of Ascorbic acid

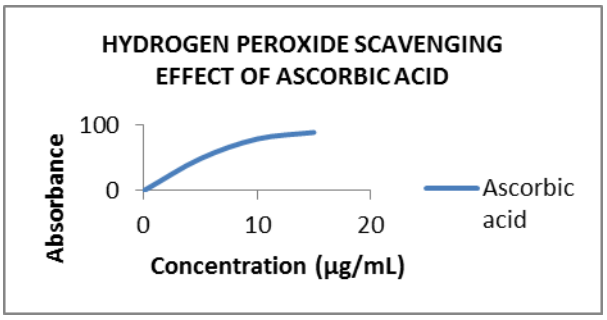


FIG. 2: Hydrogen peroxide scavenging effect of HAECB

TABLE 2: DETERMINATION OF HYDROGEN PEROXIDE SCAVENGING ACTIVITY OF *COMMELINA BENGHALENSIS L.* (LEAF) (HAECB)

S.NO	Concentration (µg/mL)		Percentage inhibition of ascorbic acid	Percentage of inhibition of HAECB
	Ascorbic acid	HAECB		
1	5	10	49 ± 0.333	44 ± 0.333
2	10	20	79 ± 0	54 ± 0
3	15	30	89 ± 0	74 ± 0
		IC_{50}	**7 µg/mL**	**18 µg/mL**

It is found that the extract requires three fold times as that of ascorbic acid to inhibit the free radicals. It showed mild anti oxidant effect when compared with ascorbic acid.

Determination of Reducing power assay of *Commelina benghalensis L.* (leaf) (HAECB)

The inhibitory concentration (IC_{50}) of *Commelina benghalensis L.* (Leaf) against **reducing power assay** was determined in comparison with ascorbic acid used as a standard. The inhibitory concentration (IC_{50}) of *Commelina benghalensis L.* (Leaf) in reducing power assay is found to be 12 µg/mL in comparison with ascorbic acid (9µg/mL).

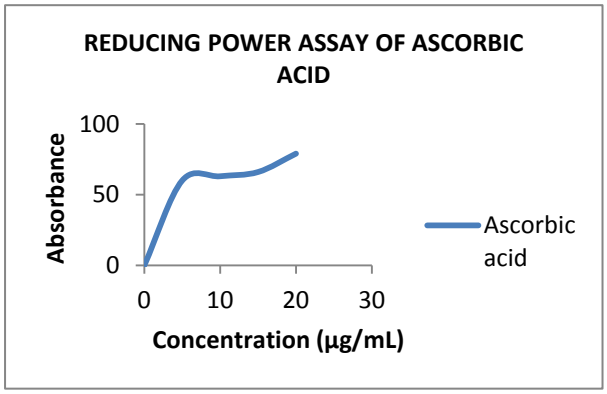


FIG. 3: Reducing power assay of Ascorbic acid

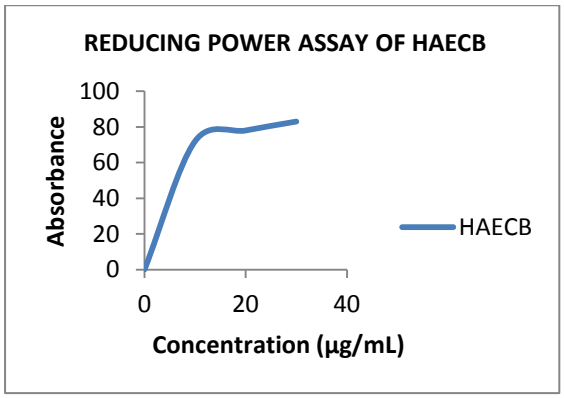


FIG. 4: Reducing power assay of HAECB

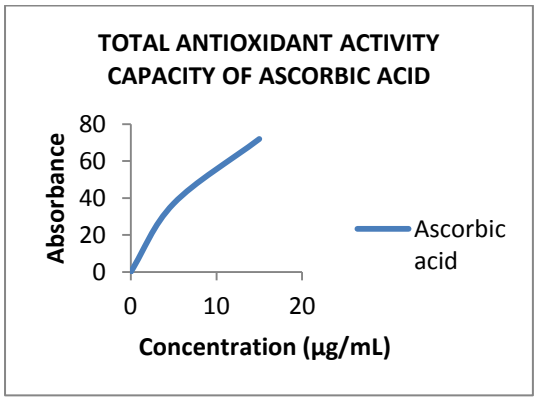
TABLE 3: DETERMINATION OF REDUCING POWER ASSAY OF *COMMELINA BENGHALENSIS L.* (LEAF) (HAECB)

S.NO	Concentration(µg/mL)		Percentage inhibition of Ascorbic acid	Percentage inhibition of HAECB
	Ascorbic acid	HAECB		
1	5	10	60 ± 0.333	72 ± 0.333
2	10	20	63 ± 0.333	78 ± 0.333
3	15	30	66 ± 0.333	83 ± 0.333
4	20		79 ± 0.333	
		IC_{50}	**9 µg/mL**	**12 µg/mL**

It is found that HAECB showed moderate antioxidant effect when compared with ascorbic acid.

Determination of Total antioxidant capacity of *Commelina benghalensis L.* (leaf) (HAECB)

The inhibitory concentration (IC_{50}) of *Commelina benghalensis L.*(Leaf) against **total antioxidant capacity** was determined in comparison with ascorbic acid used as a standard. The total antioxidant capacity is found to be 40 µg/mL in comparison with ascorbic acid 10 µg/mL.



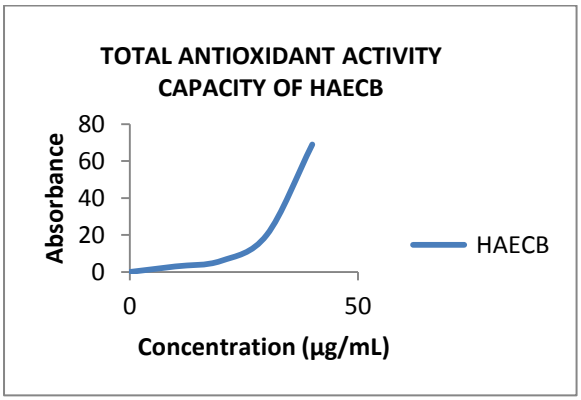


FIG. 5: Total antioxidant capacity of Ascorbic acid **FIG. 6: Total antioxidant capacity of HAECB**

TABLE 4: DETERMINATION OF TOTAL ANTIOXIDANT CAPACITY OF *COMMELINA BENGHALENSIS L.* (LEAF) (HAECB)

S.NO	Concentration (µg/mL)		Percentage inhibition of Ascorbic acid	Percentage inhibition of HAECB
	Ascorbic acid	HAECB		
1	5	10	37 ± 0.882	3 ± 0.577
2	15	20	72 ± 0.577	6 ± 0.577
3		30		20 ± 0.667
4		40		69 ± 0.577
		IC_{50}	**10 µg/mL**	**40 µg/mL**

Hydroalcoholic extract of *Commelina benghalensis L.* requires four fold times the amount of ascorbic acid to reduce the free radicals. It showed mild anti oxidant effect when compared with ascorbic acid.

CONCLUSION

The present research study reveals the phytochemical analysis of the *Commelina benghalensis L.* which adds additional scientific information to the existing research .It is concluded that this plant preparations may be formulated further, such preparations may be added as adjuvant therapy in the management of diseases.

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