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*Full Length Research Paper*

# Phytochemical screening and *In-vitro* antioxidant activities of aqueous extract of *Enicostemma littorale* Blume

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**The present study was carried out to evaluate the potential of aqueous extract of *Enicostemma littorale* Blume, through phytochemical analysis and antioxidant activities. The phytochemical analysis indicated the presence of alkaloids, flavonoids, saponins, phenols, glycosides, tannins,carbohydrates, proteins, carotenoids and lycopenes. In the antioxidant studies, the DPPH assay of the plant extract showed potent antioxidant capacity with an $IC_{50}$ value of 31.09 µg/mg to that of standard (ascorbic acid) as 20.89 µg/mg. The extract showed potent scavenging of ABTS with $IC_{50}$ value of 27.81 µg/mg on comparison with the standard (ascorbic acid) as 18.08 µg/mg. The extract also exhibited efficient Ferric Reducing Antioxidant Potential (FRAP) activity with an $IC_{50}$ value of 18 µg of ascorbic acid/mg extract. These results indicated the antioxidant potential of aqueous extract of *Enicostemma littorale*.**

**Keywords:** *Enicostemma littorale*, phytochemical studies, antioxidant.

## INTRODUCTION

The knowledge of medicinal plants has been accumulated in the course of many centuries based on different medicinal systems such as Ayurvedha, Unani and Siddha (Muthu *et al.,* 2006). Medicinal plant lore or herbal medicines are major components of traditional medicine. Analysing the phytochemicals in the medicinal plants provides scientists with insight into how effective plants are medicinal and understanding how and why they are effective, can lead to the development of new medicines (Bussmann and Sharon, 2006). Numerous studies have been carried out on some plants, vegetables and fruits because they are rich sources of antioxidants such as vitamin A, vitamin C, vitamin E, carotenoids, polyphenolic compounds and flavonoids, which prevent free radical damage and reduce the risk of chronic diseases (Quinn and Tang, 1996; Zhou and Zhang, 1991).

*Enicostemma littorale* Blume (Synonym - *Enicostemma axillare*) belongs to the family Genitanaceae, is a glabrous, perennial herb attaining a height of 5 – 20 inches, producing yellow or white coloured flowers, which are arranged in clusters. The plant has been used in the treatment of diabetes mellitus (Stanley and Srinivasan, 2005), skin diseases, malaria, abdominal ulcers, arthritis, as anti–inflammatory (Jaishree *et al.,*2008), antimalarial (Soni and Gupta, 2009), antimicrobial (Sharadha *et al.,*2008), antipyretic (Garg, 2000), antirheumatic (Kavimani and Mani Senthilkumar, 2000), antipsychotic, antihelmintic (Vidhyadhar *et al.,*2010), diuretic and hepatoprotective (Vishwakarma and Goyal, 2004). It has the property to increase the HDL levels and decreases the serum cholesterol, triglycerides, LDL, VLDL and LDL/HDL ratio (Gopal *et al.,*2004). The present study was carried out to find out the potential antioxidant activities of the aqueous extract of *Enicostemma littorale*.

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**Table 1.** Screening of phytochemicals in the aqueous extract of *E. littorale*

| Aikaloids | Flavonoids | Saponins | Carbohydrates | Proteins | Phenols | Glycosides | Tannins |
|---|---|---|---|---|---|---|---|
| ++ | ++ | + | + | + | + | ++ | ++ |

++ present; + trace

**Table 2.** Quantification of phytochemicals in the aqueous extract of *E. littorale*

| Phytochemicals | Quantity (mg/g) |
|---|---|
| **Total proteins** | 20.4 ± 0.14 |
| **Free amino acids** | 6.18 ± 0.16 |
| **Carbohydrates** | 29.71 ± 0.15 |
| **Tannins** | 2.08 ± 0.16 |
| **Phenols** | 11.43 ± 0.16 |
| **Carotenoids** | 12.15 ± 0.13 |
| **Lycopenes** | 2.23 ± 0.09 |

## MATERIALS AND METHODS

### Plant collection, preparation and analysis of the extract

The whole plant of *Enicostemma littorale* were collected during the month of December, from the local areas of Coimbatore district, Tamil nadu, India. The plant was authenticated (No. BSI/SRC/5/23/2010-11/Tech-2051) at the Botanical Survey of India, Southern regional centre, Coimbatore, India. The plant was shade dried at room temperature and 500g of *E. littorale* containing all vegetative and reproductive parts were coarsely powdered in a mixer grinder. 100g of the dried plant powder was mixed with 300 ml of water, cold macerated for 72 hours with intermittent shaking, filtered and the filtrate was then concentrated to dryness under reduced pressure at controlled temperature in a water bath. The extract yielded (14.6g %) was subjected to the phytochemical analysis to identify the constituents present in it.

A known quantity of the extract was also subjected to the quantitative determination of phytochemicals such as the total proteins (Lowry *et al,.* 1951), total free amino acids (Moore and Stein 1948), total carbohydrates (Hedge and Mofreiter 1962), tannins (Schanderl, 1970) and antioxidants such as the total carotenoids and lycopenes ( Zakaria *et al.,*1979) as well as total phenols (Singleton *et al*., 1999). The free radical scavenging assays of *E.littorale* extract were analysed by *in-vitro* antioxidant assays namely DPPH radical scavenging assay (Marxen *et al.,* 2007), ABTS assay (Roberta *et al.,* 1999) and FRAP assay (Thaipong. *et al.,* 2006). The experiments were carried out in triplicate inorder to compare the mean values of the investigated parameters.

## RESULTS AND DISCUSSION

### Phytochemical screening of the aqueous extract of *E. littorale*

The phytochemical analysis of the aqueous extract of *E. littorale* revealed the presence of alkaloids, flavonoids, saponins, carbohydrates, proteins, phenols, glycosides and tannins as shown in the table - 1.

### Quantitative analysis of the aqueous extract of *E. littorale*

In the quantitative analysis of phytochemicals and antioxidants in the aqueous extract of *E. littorale*, the total proteins, free amino acids, carbohydrates, tannins, phenols, carotenoids and lycopenes were found to be as shown in the table - 2.

### Free radical scavenging assays of the aqueous extract of *E. littorale*

### DPPH [2,2-Diphenyl-1-picrylhydrazyl] assay

DPPH test is based on the ability of DPPH, a stable free radical to decolorize in the presence of antioxidants, is a

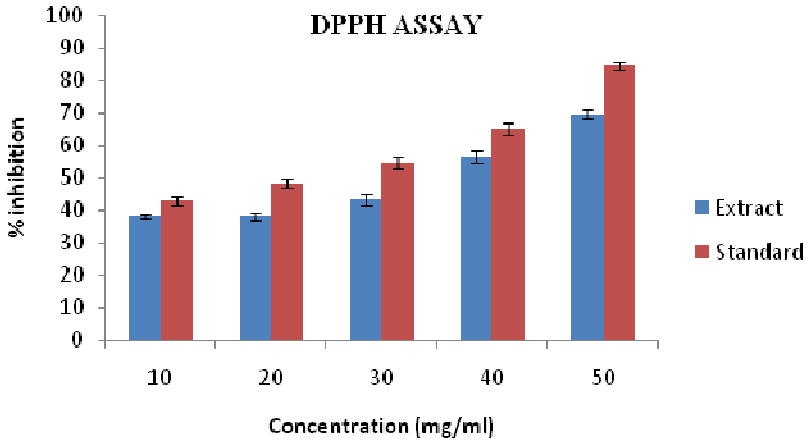


**Figure1.** Inhibitory potential of the aqueous extract of *E. littorale* and the standard ascorbic acid against DPPH free radical.

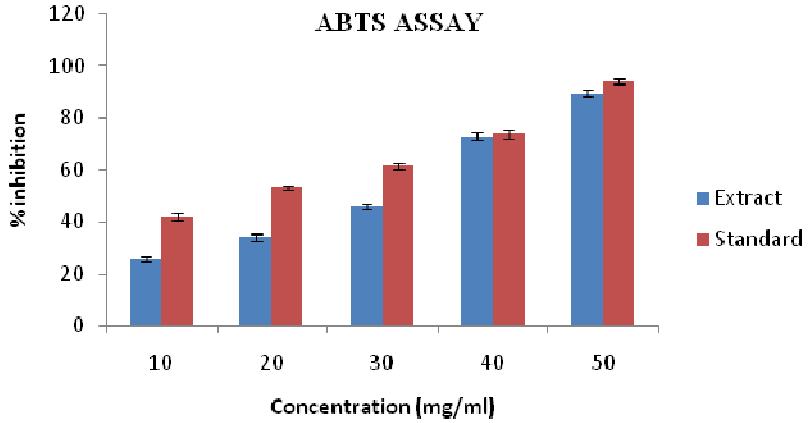


**Figure 2.** Inhibitory potential of the aqueous extract of *E. littorale* and the standard ascorbic acid against ABTS free radical.

direct and reliable method for determining the radical scavenging action (Hasan *et al.,* 2009). The demonstrated modified spectrophotometric method makes use of DPPH radical and its specific absorbance properties. The absorbance decreases when the radical is reduced by antioxidants. The DPPH radical scavenging activity of the aqueous extract of *E.littorale* is shown in the figure1.The $IC_{50}$ values of the plant extract was found to be 31.09µg/mg and that of the standard (Ascorbic acid) was found to be 20.89µg/mg.

## ABTS [2,2′-azinobis (3-ethylbenzothiazolinesulphonic acid)] Assay

ABTS assay is used for the screening of antioxidant capacity as a decolorization assay applicable to both lipophilic and hydrophilic antioxidants. The antioxidant activity is contributed by both the antioxidant and the duration of absorption of the free radical. The ABTS free radical scavenging activity of the aqueous extract of *E. littorale* was found to raise with increase in concentration. From the graph, as shown in the figure 2, the $IC_{50}$ values of the plant extract was found to be 27.81 µg/mg and that of the standard (Ascorbic acid) was found to be 18.08µg/mg.

## FRAP Assay

The FRAP assay is presented as a novel method for “antioxidant power”. Ferric to ferrous ion reduction at low pH causes colored ferrous-tripyridyltriazine complexes to form. The FRAP assay offers a putative index of

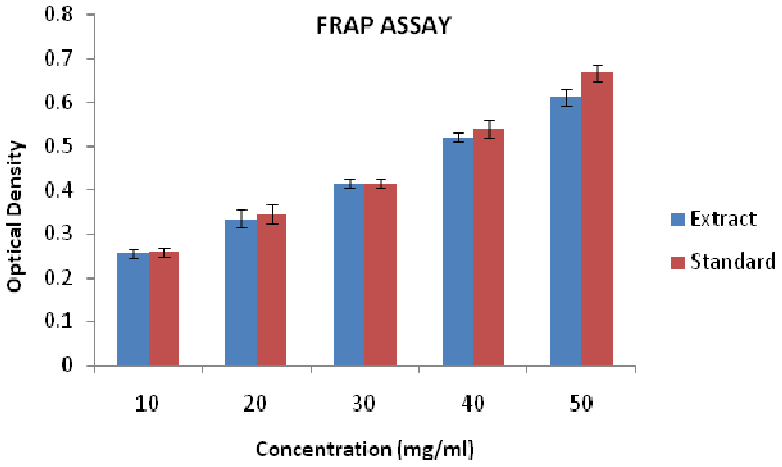


**Figure3.** Inhibitory potential of the aqueous extract of *E. littorale* and standard ascorbic acid

antioxidant, or reducing potential of biological fluids. The figure 3 shows the potentiality of the aqueous extract of *E. littorale* to scavenge the ferrous-tripyridyltriazine complexes in comparison with ascorbic acid. The $IC_{50}$ values of the plant extract was found to be 18 µg of ascorbic acid/mg extract.

## CONCLUSION

It is concluded that the aqueous extract of *Enicostemma littorale* was found to be rich in various phytochemicals and possess antioxidant properties.

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