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Phytochemical Study of *Hybanthus enneaspermus* (Linn.) F.Muell.

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ABSTRACT

To study the phytochemicals present in *Hybanthus enneaspermus* collected from the K.Puliyankulam, Virudhunagar District, Tamil Nadu, India. The present study reveals that the phytochemical analysis of nine different chemical compounds (Alkaloids Terpenoids, Saponins, Flavonoids, Tannin, Glycosides, Phenols, Steroids and Reducing sugars) were tested in three different extracts Ethanol, Acetone and Water respectively. Alkaloids, terpenoid, tannin, flavonoids, glycosides were found in Acetone extract; terpenoid, tannin, flavonoids, glycosides were found in Acetone extract; terpenoid, tannin, flavonoids, glycosides and phenolics were present in aqueous extract. All the three extracts have shown the negative result of saponin and reducing sugar. In acetone extract. Finally the solvency of *H. enneaspermus* in ethanol is high when compare with two other solvents.

Keywords: Alkaloid, flavonoid, Hybanthus enneaspermus, reducing sugars, tannin, terpenoid.

1. Introduction

Plants are a rich source of secondary metabolites with interesting biological activities. In general, these secondary metabolites are an important source with a variety of structural arrangements and properties ^[11]. Secondary metabolites are chemicals produced by plants; and their function in growth, photosynthesis, reproduction and other primary processes are not known yet. Secondary chemicals are important in plant use by widely used especially in Asia ^[21]. Plant phenolics include simple phenols, phenolic acids, coumarins, flavonoids, stilbenes, hydrolysable and condensed tannins, lignans and lignins. In plant, phenolics may act as phytoalexins, antifeedants, attractants for pollinators, contributors to the plant pigmentation, antioxidants and protective against Ultraviolet light, among others. Phenolics in general contribute to the bitterness, astringency, color, flavor, odor, oxidative stability of products in addition to health-protecting capacity of some. Flavonoids display a remarkable array of biochemical and pharmacological actions viz., anti-inflammatory, antioxidant, anti-allergic, hepatoprotective, anti-thrombic, anti-viral and anti-carcinogenic activities ^[3]. These compounds appear to play vital role in defence against pathogens and predators hence contribute to the physiological functions such as seed maturation and dormancy ^[4].

In the present study was undertaken to know the preliminary phytochemicals (qualitatively) present in various solvent extracts of *Hybanthus enneaspermus* leaves.

2. Materials and Methods

2.1 Preparation of Plant Material for Solvent Extraction:

H. enneaspermus leaves were collected from K. Puliyankulam, Virudhunagar District, Tamil Nadu, India. They were brought to the laboratory and washed well with tap water (2 to 3 times) and identified with help of Head of Department of Botany (Taxonomist), The Madura College. The collected leaves were kept under shade and allow it dried. After three weeks, they were powdered using a domestic grinder, It was successively extracted with ethanol, acetone and water using Soxhlet apparatus (500 mL). The last trace of solvent was removed under reduced pressure distillation and the crude extract was dried in a vacuum desiccator and used for the experiments. The concentrated extracts were stored in air tight screw cap vials and kept in refrigerator till further use.

2.2 Qualitative analysis of phytochemical substance

The extracts were subjected to phytochemical analysis to detect the presence of following biomolecules using the standard qualitative procedures ^[5].

a) Test for Alkaloids: 1ml of 1% HCl was added to 3ml of extract in a test tube and was treated with few drop of Meyer's reagent. A creamy white precipitate indicted the presence of alkaloids

b) Test for terpenoids: 5 ml of extract was mixed with 2 ml of $CHCl_3$ in a test tube. 3 ml of concentrated H_2SO_4 was carefully added to the mixture to form a layer. An interface with a reddish brown coloration was formed for the presence of terpenoids

c) Test for saponins: 5 ml of extract was shaken vigorously to obtain a stable persistent froth. The frothing was then mixed with 3 drops of olive oil and observed for the formation of emulsion, which indicated the presence of saponins.

d) Test for flavonoids: A few drops of 1% NH₃ solution was added to the extract in a test tube. A yellow coloration was observed for the presence of flavonoids.

e) Test for tannins: To 0.5 ml of extract solution, 1 ml of distilled water and 1-2 drops of ferric chloride solution were added and observed for brownish green or a blue black coloration.

f) Test for glycosides: 10 ml of 50% H_2SO_4 was added to 1ml of extract in a boiling tube. The mixture was heated in boiling water for 5min. 10ml of Fehling's solution (5 ml of each solution A and B) was added and boiled. A brick red precipitate indicated presence of glycosides

g) Test for phenols: Ferric Chloride Test: Extracts were treated with 3-4 drops of ferric chloride solution. Formation of bluish black colour indicates the presence of phenol.

h) Test for steroids: 2 ml of acetic anhydride was added to extract and then 2 ml of H_2SO_4 was added, the colour changes from violet to blue or green in samples extract indicates the presence of steroids.

i) Test for reducing sugars: A small fraction of extract was added vigorously with 5 ml of distilled water and filtered to the filtrates while equal volume of Fehling's solution was added and were shaken vigorously. A brick red precipitation indicates the presence of reducing sugars.

3. Results and Discussion

3.1 Qualitative Analysis of Phytochemicals from *H. enneaspermus*: The phytochemical analysis of nine different chemical compounds (Alkaloids Terpenoids, Saponins, Flavonoids, Tannin, Glycosides, Phenols, Steroids and Reducing sugars) were tested in three different extracts Ethanol, Acetone and Water respectively. However, all these chemicals were not extractable in one solvent. Alkaloids, terpenoid, tannin, flavonoids, glycosides, Steroids and phenolics were present in Ethanolic extract; Alkaloids, terpenoid, tannin and glycosides were found in Acetone extract; terpenoid, tannin, flavonoids, glycosides and phenolics were present in aqueous extract (Table I). All the three extracts have shown the negative result of saponin and reducing sugar. In acetone extract have shown additionally steroids and phenols were absent; alkaloids absent in aqueous extract. The solvency of H. enneaspermus in ethanol is high when compare with two other solvents.

Table 1: The qualitative test for	phytochemical analysis of various
extracts of H.	enneaspermus.

	Solvents		
Compounds	Ethanol	Acetone	Aqueous
Alkaloids	++	++	-
Teripioids	++	+	+
Saponins	-	-	-
Flavonoids	++	-	++
Tannin	+	++	++
Glycosides	-	+	+
Phenols	++	-	+
Steroids	++	-	-
Reducing sugars	+	-	-

The availability of specific phytochemicals in plant gives it specific medicinal properties. Therefore, presence of above phytochemicals in *H. enneaspermus* can be correlated with its medicinal potential. Similar reports on phytochemical composition of various medicinal plants were made earlier by many workers ^[6, 7-9]. However, it is very essential to isolate the bioactive fractions from these major groups so that it can be used further in designing specific drugs.

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