

Preliminary phytochemical evaluation of leaf extracts of *Gymnema sylvestre*, *Phyllanthus amarus*, *Phyllanthus reticulatus* of Siddarabetta, Tumkur district, Karnataka

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ABSTRACT

The Ethno medicine involves the use of different plant extracts or the bioactive constituents of vital importance in the health application at an affordable cost and study of such ethanomedicine keenly represents one of the best avenues in searching new economic plants for medicine. In keeping this view in mind the present investigation is carried out in *Gymnema sylvestre*, *Phyllanthus amarus*, *Phyllanthus reticulatus* leaves collected from Siddarabetta, Tumkur district, Karnataka, India. Qualitative phytochemical analysis of three plants confirms the presence of various phytochemicals like alkaloids, flavonoids, tannins, saponins, terpenoids and Quinone. The bioactive compounds from different solvent extracts suspected of anti-diabetic properties.

INTRODUCTION

Plants have an almost limitless ability to synthesize aromatic substances mainly secondary metabolites, of which at least 12,000 have been isolated, a number estimated to be less than 10% of the total. In many cases, these substances serve as the molecules of plant defense against predation by microorganisms, insects, and herbivores. Further, some of which may involve in plant odour (terpenoids), pigmentation (tannins and quinines) etc., It is now clear that, the medicinal values of these plants lie in the bioactive phytochemical constituents that produce definite physiological effects on the human body. Though the traditional Indian system of medicine has a long history of use, they lack adequate scientific documentation, particularly in light modern scientific knowledge¹. These natural compounds formed the base of modern drugs as we are using today^{2,3,4}. "Phyto" is the Greek word for plant. There are many families of phytochemicals and they help the human body in a variety of ways. Phytochemicals may protect human from various diseases. Phytochemicals are non-nutritive plant chemicals that have protective or disease preventive properties. Phytochemicals are basically divided into two groups that are primary and secondary metabolites according to their functions in plant metabolism. Primary metabolites comprise common sugars, amino acids, proteins and chlorophyll while secondary metabolites consist of alkaloids, flavonoids, tannins and so on^{5,6}.

Gymnema sylvestre: Found in tropical forests of central and southern India. The woody *Gymnema* plant also grows in parts of Africa. The leaves of this long, slender plant have been used for more than 2,000 years in India to treat diabetes.

Phyllanthus amarus: It is a common Arabic weed of disturbed ground in Southern Florida, Bahmas, West Indies, Tropical America, and is naturalized in the old world tropics.

Phyllanthus reticulatus: *Phyllanthus reticulatus* is very common and widespread in India. It often grows in low altitudes. Ethnobotanical survey of these plants reveals anti-diabetic effects, although reliable research required and not yet been performed¹⁰. The present study is designed to explore the preliminary phytochemical analysis of *Gymnema sylvestre*, *Phyllanthus amarus*, and *Phyllanthus reticulatus* leaves for their pharmacological properties.

MATERIALS AND METHODS

Plant Materials: The fully mature leaves of *Gymnema sylvestre*, *Phyllanthus amarus*, and *Phyllanthus reticulatus* were collected from Siddarabetta, Tumkur district, Karnataka, India. During September 2011 and were washed thoroughly and shade dried.

Extraction of Plant Material: The dried leaves were grounded into a fine powder and the total mass was subjected for extraction by a hot percolation method with water, ethanol and Methanol in soxhlet apparatus for 72 hrs. Each solvent extraction step was carried out for 24 hrs. and after extraction the extracts were concentrated by evaporation and stored at 4°C for further study⁷.

Preliminary Phytochemical Screening: The phytochemical screening of the extracts was done using standard procedure as described⁷. The following qualitative tests were carried out as follows.

1) Steroids and Terpenoids: 10mg of the extract was dissolved in chloroform. Few drops of acetic anhydride were added followed by 1 ml of conc Sulphuric acid. Blue colour in chloroform layer which changes to green shows the presence of steroids, whereas the appearance of pink colour in chloroform layer shows the presence of terpenoids.

2) Alkaloids: 10mg of the extract was dissolved in conc HCL and filtered. A few drops of solution are poured into the center of watch glass. Mayer reagent is added along

Table: 1 Phytochemical screening

Phyto compounds	Phyllanthus amarus			Gymnema sylvestre			Phyllanthus reticulatus		
	W	E	M	W	E	M	W	E	M
Steroids	-	+	-	-	-	-	-	+	+
Terpenoids	+	-	-	+	-	+	-	-	-
Alkaloids	-	-	-	+	+	+	-	-	-
Flavonoids	+	-	-	+	-	-	+	-	-
Coumarins	-	-	-	-	-	-	-	-	-
Saponins	-	-	-	+	-	+	-	-	-
Tannins	-	+	+	+	+	-	-	+	+
Phenol	+	+	+	-	-	-	+	+	+
Anthraquinone	-	-	-	-	-	-	-	-	-
Quinone	+	+	+	+	+	+	+	+	+
Catechin	+	-	-	-	-	-	-	-	-

W=water extract E=Ethanol extract M=Methanol extract

the sides of the watch glass with the help of a glass rod. Formation of a gelatinous white precipitate at the junction of two liquid shows the presence of alkaloids.

3) Flavonoids: 10mg of the extract was dissolved in methanol. Magnesium turnings were added into this followed by conc HCL. A magenta colour shows the presence of Flavonoids.

4) Coumarins: 10mg of the extract is dissolved in methanol and alcoholic KOH was added. The appearance of yellow colour which decolorizes while adding conc HCL shows the presence of Coumarin.

5) Saponins: Extract was dissolved in water and shaken well. Froth which last for a long time shows the presence of saponins

6) Tannins: 10 mg of the extract was boiled with 1 ml water for 30 min. The extract is filtered clear and to this 0.5 ml 2% gelatin was added. A curdy white precipitate indicates the presence of tannin.

7) Phenolic compounds: Extract was dissolved in alcohol and 1 drop of neutral ferric chloride was added to this. The intense colour indicates the presence of phenolic compound.

8) Anthraquinone: To the extract Magnesium Acetate solution was added the pink colour developed indicates the presence of Anthraquinone.

9) Quinone: Few mg of the substrate in alcohol is treated with sulphuric acid. The colour developed indicates the presence of Quinone.

10) Catechin: Few mg of the substrate in alcohol is treated with a few drops of Ehrlich reagent and a few drops of concentrated Hcl. The pink colour developed indicates the presence of catechin.

RESULTS

The present study carried out on the plant samples revealed the presence of medicinally important bioactive compounds. The phytochemical characters of all the plants investigated are summarized in the Table: 1. The water extract of *Phyllanthus amarus* was found to contain terpenoids, flavonoids, phenol quinones and catechin. And ethanol extracts show steroids, tannins, phenol, quinones and methanol extracts exhibit only tannins,

phenol and quinones. The water extracts of *Gymnema sylvestre* exhibit terpenoids, alkaloids, flavonoids, saponins, tannins, and quinine's. Whereas methanol extracts exhibits terpenoids, alkaloids, saponins and quinones. The water extracts of *Phyllanthus reticulatus* exhibit flavonoids phenol and quinones. Ethanol extracts show steroids, tannins, phenol and quinones. And methanol extracts showed steroids, tannins, phenol and quinones.

DISCUSSION

The phytochemical screening and qualitative estimation reveals leaves were rich in phenol, quinones and little tannins. *Gymnema sylvestre* exhibits rich alkaloids in all solvent extract flavonoids found only in water extracts in all plants. Saponins were noticed in water and methanol extracts of *Gymnema sylvestre*. Steroids present only in ethanol extracts of *Phyllanthus amarus* and in methanol and ethanol extracts of *Phyllanthus reticulatus*. Catechin is identified only in water extracts in *Phyllanthus amarus* and no other leaves reveal the presence of Coumarins. The plants studied here can be such as a potential source of useful drugs. Further studies are required to detect the antibiotic activity of the plants were used as a traditional medicine to treat diabetes mellitus and studies required to characterize bioactive compounds useful in treating diabetes mellitus.

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