PRELIMINARY PHYTOCHEMICAL EXPLORATION OF *HIBISCUS CANNABINUS* PLANT LEAF EXTRACTS USING DIFFERENT SOLVENTS

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ABSTRACT

Hibiscus cannabinus (Kenaf) is one of the most important medicinal plants. The aim of the present study is to evaluate the phytochemical constituents by using quantitative and qualitative analysis of *Hibiscus cannabinus* leaves using aqueous, chloroform, petroleum ether, ethyl acetate, and ethanol extracts with the help of standard techniques. Phytochemical screening reveals the presences of flavonoid, tannins, terpenoids, quinone, saponins, phenolic compound, steroids and coumarins. The Steroids and Terpenoids were present only in the ethanolic extract of Hibiscus cannabinus. The Alkaloids and Glycosides were completely absent in all the five extracts of *Hibiscus cannabinus* leaves. Quinone and Phenolic compounds were present in all the extract. Finally, the phytoconstituents of *Hibiscus cannabinus* leaves showed the high secondary metabolites with greater potentials.

Keywords: Phytochemical, *Hibiscus cannabinus*, Bioactive compounds, Aqueous

INTRODUCTION

Medicinal plants have higher bioactive compounds that has used in varied applications. In clinical studies used to develop drugs and in environmental treatments used to degrade dye stuffs and pigments (Edeoga *et al.*,2005 and Yadav *et al.*,2011). Plant products have been part of phyto medicines since time immemorial. This can be derived from barks, leaves, flowers, roots, fruits, seeds (Cragg *et al.*,2001). Plants containing beneficial phytochemicals may supplement the needs of the human body by acting as natural antioxidants (Suffredini *et al.*,2004). Phytochemicals are naturally occurring in the medicinal plants, leaves, vegetables and roots that have defense mechanism and protect from various diseases. Phytochemicals are primary and secondary compounds. Various studies have shown that many plants are rich source of antioxidants (Wadood *et al.*,2013).Phytochemicals are bioactive compounds present in plant foods which protects the body from the damaging effects of free radicals. Due to the beneficial effects of phytoconstituents in contributing to human health, scientists are exploring newer and cheaper sources of these bioactive compounds such as food industries by-products, agricultural bio wastes etc., (John *et al.*,2017). Knowledge of the phyto constituents of plants is desirable and the value for synthesis of complex chemical substances is quite easier (Mojab *et al.*,2003). Secondary metabolites are biologically active substances found in plants. The juice of the *Hibiscus cannabinus*, mixed with sugar and black pepper, is used in the treatment of biliousness with acidity. The seeds are aphrodisiac and stomachic. They are added to the diet in order to promote weight increase. *Hibiscus cannabinus*

belongs to the genus Hibiscus is probably native to Southern Asia. In Ayurveda the leaves are used in the treatment of dysentery and bilious, blood and throat disorders (Subi *et al.*,2015). Phytochemical analysis results showed as the presences of Alkaloids, Saponins, Tannins, Steroids, Glycosides, Flavonoids and Triterpenoids (Senguttuvan et al.,2014). In the present work, qualitative and quantitative phytochemical analysis was carried out in locally available *Hibiscus cannabinus*.

MATERIALS AND METHODS

Selection of Plant Species

The *Hibiscus cannabinus* plant leaves were collected from Manakkal, Lalgudi, Tiruchirapalli District, Tamilnadu. The collected plant materials were shown in **Fig.1**. The collected leaves were washed thoroughly 2-3 times with running tap water and once sterile with distilled water. Then cut into small pieces and d shade dried at room temperature for two weeks and made in to coarsely powdered separately and stored in well closed bottles for further analysis.

Fig.1 Collected locally available *Hibiscus cannabinus* plant leaves

Preparation of Plant Extracts

The fresh plant sample (leaves) were collected and washed under the running tap water to remove soil particles and other dust particles (Wadood *et al.*,2013). The leaves were air dried under the laboratory condition at room temperature for 15days.The dried leaves samples were ground well in to a fine powder with the help of mixer grinder (Yadav *et al.*,2011). A 10gm air dry plant was soaked into 50ml organic solvents. viz, aqueous, chloroform, petroleum ether, ethyl acetate, and ethanol extracts, separately for 24hrs in an orbital shaker at normal temperature. The extracts were filter through the Whatman filter paper No:1 (Alade *et al.*,1993). The extract was allowed to dry using rotary evaporator. The condensed extracts were stored in airtight container at 4°C till further investigation (Wong *et al.*,2006). Solvents used aqueous, chloroform, petroleum ether, ethyl acetate, and ethanol extracts were used as the solvents for the preparations of plant extracts. The process of extraction continues for 24 hours or till the solvent in siphon tube of an extractor become colorless. After that the

extract was taken in a beaker and kept on hot plate and heated at 30-40ºC till all the solvent got evaporated. Dried extract was kept in refrigerator at 4ºC for their future use in phytochemical analysis (Yadav *et al.*,2011).

Phytochemical Analysis

Phytochemical screening was performed to identify phytochemicals in the aqueous, chloroform, ethanol, Ethylacetate and petroleum ether; the preliminary phytochemical screening for the detection of various phytochemical constituents such as alkaloids, flavonoids, steroids, tannins.

Test for Alkaloids

To each extract 2ml was acidified with a few drops of dilute hydrochloric acid. Then 1ml of Dragendorff's reagent was added. The appearance of orange to red precipitate indicates the presence of alkaloids.

Test for Flavonoids

To 2 ml of extract, 1 ml of 2N sodium hydroxide(NaOH) was added. Presence of yellow color indicates the presence of flavonoids.

Test for Steroids

Chloroform 10ml was added to 2ml of all three plant extracts. To these extracts 1ml of acetic anhydride was added and then 2ml of concentrated sulphuric acid was added along the sides of the test tube. Color formation at the junction is noted. The appearance of blue green color indicates the presence of steroids.

Test for Tannins

To 2ml of each extract a few drops of 10% lead acetate were added. The appearance of white precipitate indicates the presence of tannins.

Test for Terpenoids

To 0.5 ml of the each extract was treated with 2 ml of chloroform and conc. sulphuric acid. Formation of red brown color at the interface indicates the presence of terpenoids.

Test for Quinones

To 1 ml of each extract, 1 ml of concentrated sulphuric acid was added. Formation of red color indicates presence of quinones.

Test for Coumarins

To 1 ml of each extract with10% sodium hydroxide was added to 1ml of the extract. Formation of yellow color indicates the presence of coumarins.

Test for Glycosides

To 2 ml of filtered hydrolysate, 3 ml of chloroform was added and shaken well, chloroform layer was separated and 10% ammonia solution was added to it. Formation of pink color indicates presence of glycosides.

Test for Saponins

To 1ml of each extract taken in a measuring jar, 9ml of distilled water was added and shaken vigorously for 15seconds and extract were allowed to stand for 10min.Formation of stable foam (1cm) indicates the presence of saponins.

Test for Phenols

To 2 ml of distilled water followed by few drops of 10% ferric chloride was added to 1ml of the extract. Formation of blue or green color indicates the presence of phenols.

RESULT AND DISCUSSION

In this study, the preliminary phytochemical screening of locally available collected *Hibiscus cannabinus* were analyzed the phytoconstituents with different solvents and the phytochemical analysis done in water, chloroform, ethanol, ethyl acetate and petroleum ether. The extracts of *Hibiscus cannabis* leaves revealed the presence of flavonoid, tannins, terpinoids, quinone, saponins, phenolic compound, steroids and coumarins. The Steroids and Terpenoids were present only in the ethanolic extract of Hibiscus cannabinus. The Alkaloids and Glycosides were completely absent in all the five extracts of *Hibiscus cannabinus* leaves. Quinone and Phenolic compounds were present in all the extract. Results of this preliminary phytochemical investigation in the extract of *Hibiscus cannabinus* leaves were shown in the **Table 1** and **Fig.2.**

Table 1: Phytochemical investigation in the extract of *Hibiscus cannabinus*

S.No	Phytochemical	Water	Ethanol	Chloroform	Ethyl acetate	Petroleum ether
1.	Alkaloids	-	-	-	-	-
2.	Flavonoids	+	+	-	-	-
3.	Steroids	-	+	-	-	-
4.	Tannins	+	+	+	-	+
5.	Terpenoids	-	+	-	-	-
6.	Quinone	+	+	+	+	+
7.	Coumarins	+	+	-	-	-
8.	Glycosides	-	-	-	-	-
9.	Saponins	-	+	-	-	+
10.	Phenols	+	+	+	+	+

+ = Presence, - = Absence

Fig.2 Laboratory Analysis in different extracts of *Hibiscus cannabinus*

The present findings are strongly supported by the following findings. Flavonoids belong to the group of polyphenolic compounds and are typically known for their health promoting potentials such as anti-oxidant, anti-allergic, anti-inflammatory, anti-microbial and anti-cancer properties (Boots *et al.*,2008). Phytochemical analysis conducted on the plant extracts revealed the presence of constituents which are known to exhibit medicinal as well as physiological activities (Njoku *et al.*,2009). Phenolic constituents were found to be higher in concentration than the alkaloids and flavonoids. (Hussain *et al.*,2011). Steroids have been reported to have antibacterial properties and they are very important compounds especially due to their relationship with compounds such as sex hormones (Raquel *et al*.,2007). Sometimes the investigation results shows the variation in the contents like alkaloids, flavonoids, phenol and carbohydrate when compared to above mentioned results. These variations are due to number of environmental factors such as climate, altitude, rainfall etc. as mentioned (Kokate et *al*., 2004).

CONCLUSION

The Preliminary phytochemical analysis of *Hibiscus cannabinus* leaves was analysis to maximum classes of phytoconstituents is present. The medicinal plants have highest therapeutic efficiency by both clinical and environmental fields. The qualitative phytochemical analysis were studied and represented in table, if the phytochemical compound is present denotes positive sign and absence mark as negative sign. The phytochemical compounds identified in this study have earlier been proved to be bioactive and present numerous secondary metabolites. In the present study, all the five extracts are analyzed and to indicate the more positive result in ethanolic plant leaf extract. Several studies confirmed the presence of phytochemical constituents and these contributes medical as well as environmental treatments to safeguard out community.

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