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Qualitative Phytochemical Screening of Medicinal Plants Using Different Solvent Extracts

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ABSTRACT

Medicinal plants are rich in bioactive components that are utilized to treat various human ailments. They are crucial to healing as well. Phytochemical constituents are responsible for the medicinal activity of plant species. Phytochemical screening is an important step in identifying bioactive compounds present in particular medicinal plants. Hence, in this present work, phytochemical screening of leaf extract of some traditional medicinal plants, namely Cannabis sativa, Ricinus communis, and Bryophyllum pinnatum was carried out. The solvent extracts of the leaves of respective plants were prepared using the Soxhlet apparatus with acetone, chloroform, petroleum ether, and aqueous solvents. Qualitative phytochemical analysis of plants included tests for reducing sugars, flavonoids, steroids, glycosides, polyphenols, tannins, terpenoids, and coumarins. All eight tested phytoconstituents were found present in all three plants in any solvent extracts. Aqueous extract confirmed the presence of a maximum number of phytoconstituents in C. sativa in comparison to other solvents. Acetone confirmed the maximum and chloroform confirmed the minimum number of phytoconstituents in *R. communis*, while chloroform confirmed the maximum and aqueous extract confirmed the minimum number of phytoconstituents in *B. pinnatum*. These phytochemicals may be a source of innovative plant-based medications because their existence is connected with the therapeutic potential of these plants.

Keywords: Phytoconstituents, Cannabis sativa, Ricinus communis, Bryophyllum pinnatum.

INTRODUCTION

Medicinal plants are an excellent source as they provide a wide variety of possible therapeutic compounds that are both diversified and reasonably safe, compared to manufactured pharmaceuticals^{1,2}. According to the World Health Organization (WHO), traditional plant-based medicines constitute the major source of healthcare for more than 80% of the world's population in developing and underprivileged nations^{3,4}. The WHO has made an effort to identify all internationally used medicinal plants and recognized over 20,000 species⁵. The demand for plants originated raw materials is increasing at a rate of 15% to 25% annually and is expected to increase by over US\$5 trillion by the year 2050. The estimation of total trade by medicinal plants is approximately US \$ 1 billion annually

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in India⁶. India is incredibly rich in plant species that have therapeutic significance. Most people in society utilize these plants as herbal remedies or as pharmaceutical ingredients in contemporary medicine⁷. Researchers have been concentrating more on herbal remedies recently, and various plants are being investigated for potential therapeutic benefits⁸. Collaborative work on ethnobotanicals, ethnomedical, ethnopharmacological, and phytochemicals is crucial to attaining research progress in the field of medicinal plants9. Most of the studies have focused on the phytochemical screening of medicinal plants with an extraction efficiency of one or two solvents included in this study^{10,11}. The focus point of some of the studies was on the single solvents on multiple plants^{12,13,14}. So the present study was designed to include the preliminary phytochemical analysis of C. sativa, R. communis, and B. pinnatum and also shows the comparative metabolite extraction efficiency of acetone, chloroform, petroleum ether, and aqueous solvent extracts with the particular plant.

Medicinal uses of plant parts

C. sativa is an important herbaceous plant that originated from Central Asia that has been used in traditional medicine since the dawn of time. The plant has been used medicinally for centuries in a variety of civilizations in the treatment of various ailments. For example, for treating asthma, loss of appetite, depression, and sleeplessness¹⁵. In modern medicine, it has medical usage in the treatment of anorexia related to HIV/AIDS, nausea and vomiting in cancer chemotherapy^{16,17}, spasticity in multiple sclerosis¹⁸, gastrointestinal disorders, postpartum hemorrhage, difficulties during child labor, and in the management of sexually transmitted diseases¹⁹. There is significant evidence that cannabinoids are also effective in the treatment of several other disorders like neuropathic and chronic pain, movement disorders and spasms^{20,21}.

B. pinnatum is a succulent perennial plant native to Madagascar that has been used in medicines for a long time. This plant is used to cure prostate cancer as well as the common cold. *B. pinnatum* and other herb extracts in herbal compositions are said to operate as tonics, boosting health and respiration. The plant shows neurosedative activities, muscle relaxant activities²², anticonvulsant activities²³, nephroprotective, urolithic, antibacterial, antiallergic, antileishmanial, anticonvulsant, anti-inflammatory, antiulcer, and analgesic activities^{24,25}. Leaves of *B. pinnatum* also have Neuro-restorative potential²⁶. *R. communis* is a small woody tree found in India, South Africa, Russia and Brazil. The root, leaf, and seed oils of this plant have been used in Indian medicine to treat hypoglycemia, liver diseases, and inflammation^{27,28}. The plant parts have anticancer, antidiabetic, antitumor, antiasthmatic, antifertility, bone regeneration, cytotoxicity, antioxidant, insecticidal, antimicrobial, antiprotozoal²⁹ and antiulcer properties³⁰. The seed oil also has a laxative effect and induces labor in pregnant females³¹.

MATERIALS AND METHODS

Collection of plant materials

Fresh leaves of plants free from diseases were collected during January 2022. Taxonomic identification of plants was carried out by the department of Botany, Maharshi Dayanand University, Rohtak, Haryana.

Preparation of extracts

Collected plant leaves were washed thoroughly with running tap water. Leaf materials were cut down into small pieces and air-dried under shade for 22 days. An electric blender was used to grind the dried plant material into a fine powder and kept in small plastic bags with paper labeling. The crude plant extracts were prepared with different solvents like acetone, petroleum ether, chloroform and aqueous solvent using the Soxhlet extraction method for approximately 20 hours. The crude extracts were collected and kept in the refrigerator at 4°C in sealed bottles for further use.

Qualitative phytochemical analysis

The qualitative analysis of phytochemicals was done for different plant extracts with four different solvents, acetone, petroleum ether, chloroform, and aqueous solvent by using the following standard protocols. The experimental method is illustrated in Figure 1.

Test for steroid

2 mL of chloroform was added to the crude extract and concentrated H_2SO_4 was also added side by side. The evolution of red color in the lower chloroform layer directs the presence of steroids. Another test was also conducted, where 2 mL of chloroform was mixed with crude extract. After that 2 mL of acetic acid and 2 mL of concentrated H_2SO_4 were added to the mixture. Appearance of greenish color depicts the occurrence of steroids in the sample³².

Test for Terpenoids

The crude extract was mixed in 2 mL of chloroform and the solution was evaporated to dryness. 2 mL of concentrated H_2SO_4 was then added and get the solution heated for another 2 minutes. Appearance of grayish color indicates the availability of terpenoids in the sample^{33,34}.

Test for coumarins

3 mL of 10% sodium hydroxide (NaOH) was mixed with 2 mL of crude extract, appearance of yellow color depicts the presence of coumarins^{32,35}.

Test for reducing sugar (Fehling test)

1 mL of each of Fehling A and Fehling B reagents were mixed together and the mixture was then added to the crude extract and get the solution boiled. The formation of brick red color precipitates in the bottom of the vessel shows the availability of reducing sugars in the sample³³.

Test for polyphenols and Tannins

2 mL of 2% FeCl_3 solution was added to the crude extract. The appearance of a bluishgreen or bluish-black color shows the occurrence of polyphenols and tannins in the sample³⁴.

Test for flavonoids

The crude extract was mixed with a few small pieces of magnesium ribbon and then concentrated Hydrochloric acid was added to it drop by drop. After a few minutes, the appearance of pink or magenta-red color indicates the availability of flavonoids in the sample^{11,35}.

Test for glycosides (Salkowski's test)

2 mL of chloroform was added to the crude extract. Then 2 mL of concentrated H_2SO_4 was added and gently shaken. A reddish brown color shows the occurrence of steroidal ring, i.e., glycone portion of the glycoside^{10,32}.

Test for cardiac glycosides (Keller-Kiliani's test)

A few drops of 2% FeCl₃ solution were added to glacial acetic acid and 2 mL of this solution was mixed with the crude extract. The mixture was then transferred to another vessel having 2 mL of concentrated H_2SO_4 . The formation of a brown color ring at the interface indicates the availability of cardiac glycosides in the sample³⁵.

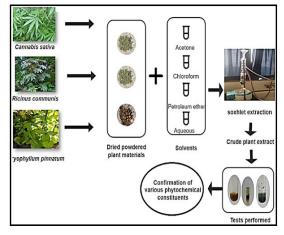


Fig. 1. A schematic diagram showing the experimental method RESULTS AND DISCUSSION

The phytochemical characteristics of leaf extract of *C. sativa* tested with different solvent extracts are summarized in Table 1, which shows the presence of medically active compounds in the plant. Reducing sugars, glycosides, cardiac glycosides, polyphenols, tannins, flavonoids, steroids, terpenoids and coumarins, all were found present in different solvent extracts. Aqueous extract yielded more metabolites in comparison to acetone, chloroform and petroleum ether extracts of *C. sativa*. Comparative studies for the same plant are shown in Table 2, which demonstrate the presence of most of the phytoconstituents confirmed by our study, but the solvents were not similar. Studies confirmed the maximum yield with aqueous extract.

Phyto-constituents	Acetone	Chloroform	Petroleum ether	Aqueous
Reducing sugars	-	-	+	-
Glycosides	-	-	+	+
Cardiac glycosides	-	-	-	+
Polyphenols and Tannins	+	+	+	+
Flavonoids	+	-	-	-
Steroids	+	-	+	+
Terpenoids	-	+	-	-
Coumarins	+	+	-	+

Table 1: Phytochemical analysis of leaf extracts of C. sativa

Present (+), Absent (-)

Plant part	Solvent	Phyto-constituents	References
Leaf, Stem,	Chloroform, Alcohol,	steroids, resins, fixed oil, alkaloids, flavonoids,	10
Root	Aqueous, n-hexane	terpenoids, tannin, amino acids, proteins,	
		glycosides, phenol, saponins	
Leaf	-	Alkaloids, flavonoids, cardiac glycosides, resins,	36
		terpenes, steroids	
Leaf	-	phenol, saponins, Alkaloids, flavonoids, glycosides,	11
		steroids	
Leaf	-	Anthocyanines, Steroids, Terpinoids	37

The phytochemical characteristics of *R. communis* tested with different solvent extracts are summarized in Table 3. Medically active compounds, reducing sugars, glycosides, cardiac glycosides, polyphenols, tannins, flavonoids, steroids, terpenoids, and coumarins, all were found present in different solvent extracts. For *R. communis*, all the phyto-constituents were found present in acetone extract, glycosides (1 compound) was found absent in petroleum ether extract, reducing sugars and coumarins (2 compounds) were found absent in aqueous

extract, while cardiac glycosides, flavonoids and terpenoids (3 compounds) were found absent in chloroform extract. Results revealed that acetone extract yielded maximum and chloroform extract yielded minimum numbers of constituents. Various studies on the phytochemical analysis of *R. communis* are summarized in Table 4, which shows the presence of similar compounds in different parts of the plant with the same or different solvent extracts. Most of the studies doesn't revealed the extraction efficiency of solvents with the particular metabolites, that can be treated as the drawback of that study.

Phytochemical test	Acetone	Chloroform	Petroleum ether	Aqueous
Reducing sugars	+	+	+	-
Glycoside	-	+		
Cardiac glycosides	+	-	+	+
Polyphenols and Tannins	+	+	+	+
Flavonoids	+	-	+	+
Steroids	+	+	+	+
Terpenoids	+	-	+	+
Coumarins	+	+	+	-

Table 4. Comparative studies (11. Communis	Table 4:	Comparative studies	(R. communis)
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Plant part	Solvent	Phyto-constituents	References
Leaf	water, methanol, ethanol, acetone	Proteins Carbohydrates Phenols/Tannins Alkaloids, Flavonoids, Steriods,	38
Leaf	-	Flavonoids Saponins Glycosides Steroids Phenols, Tannins, Saponins, Starch	39
Seed, Root, Leaf	-	Phenol, Flavonoids, Glycoside, Steroid	40
Seed oil	-	Alkaloids, terpenoids, cardiac glycosides, tannins, steroids, saponins	41

The results of phytochemical analysis of different solvent extracts of *B. pinnatum* are shown in Table 5. Results revealed the presence of all tested compounds in plant, chloroform extract shows presence of all phyto-constituents, while reducing sugars were absent in acetone extract and flavonoids were found absent in petroleum ether extract. 4 compound were absent in the aqueous extract. It implies chloroform extract yielded maximum and aqueous extract yielded minimum metabolites in *B. pinnatum*. Table 6 is compiled on basis of some previous studies, which show similar results for the same and different solvent extracts for the leaf and other parts of the plant. In the case of different plants, the phytoconstituents extraction efficiency of different solvents varies greatly.

Phytochemical test	Acetone	Chloroform	Petroleum ether	Aqueous
Reducing sugars	-	+	+	+
Glycoside	+	+	+	-
Cardiac glycosides	+	+	+	-
Polyphenols and Tannins	+	+	+	+
Flavonoids	+	+	-	-
Steroids	+	+	+	+
Terpenoids	+	+	+	+
Coumarins	+	+	+	+

Table 5: Phytochemical ar	alysis of leaf extracts of <i>B. pinnatum</i>
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Table 6: Comparative studies	s (B. pinnatum)
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Plant part	Solvent	Phyto-constituents	References
Leaf	water, methanol, ethanol, acetone	Proteins, Carbohydrates, Phenols/Tannins, Flavonoids, Saponins, Glycosides, Steroids, Alkaloids	38
Wood, Stem bark	Hexane, ethyl acetate, methanol	Reducing sugars, saponins, steroids, tannins, alkaloids, flavonoids, phenols	42
Leaf, Root, Stem	-	Alkaloid, Tannin, Saponin, Flavonoid, Terpenoid, Glycoside, Phenols	43
Leaf	-	Flavonoid, Glycoside, Alkaloids, Triterpenoids, Tannins, Phenolic	44

CONCLUSION

The majority of the biologically active phytochemicals were found present in acetone, petroleum ether, chloroform, and aqueous extracts of leaves of *C. sativa*, *R. communis*, and *B. pinnatum*. *R. communis*, and *B. pinnatum* were more phytochemically rich in comparison to *C. sativa*. The medicinal plants were found rich in context of secondary metabolites, which are commonly employed in conventional medicine to treat and combat a wide range of illnesses. The antispasmodic, anti-inflammatory, analgesic, diuretic, and many other properties can be imputed to their high availability of polyphenols, flavonoids, tannins, terpenoids, steroids, glycosides, coumarins, and reducing sugars. The research carried out by us confirmed the therapeutic qualities of these plant species. It will be useful to do more research in the field of the quantitative analysis of these phytocompounds. Our study can be used as scientific support for the formulation of a variety of medications.

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Conflicts of interest

No such conflicts exist.

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