ISSN: 0975-833X

Available online at http://www.journalcra.com

International Journal of Current Research
Vol. 11, Issue, 01, pp.824-827, January, 2019

DOI: https://doi.org/10.24941/ijcr.34172.01.2019

INTERNATIONAL JOURNAL OF CURRENT RESEARCH

REVIEW ARTICLE

STUDIES ON THE PHYTOCHEMICAL, SPECTROSCOPIC CHARACTERIZATION AND ANTIBACTERIAL EFFICIENCY OF *SPATHOLOBUS PURPUREUS* BENTH. EX BAKER (FAMILY- FABACEAE)

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ARTICLE INFO

Article History:
Received 22nd October, 2018
Received in revised form
16th November, 2018
Accepted 08th December, 2018
Published online 31st January, 2019

Key Words:
Antimicrobial, Spectroscopical, Phytochemical, terpenoids, flavonoids, saponins, Alkaloids, flavonoids, steroids, *Spatholobus purpureus* Benth. ex Baker.

ABSTRACT

A large number of medicinal plants are used as alternate medicine for diseases of man and other animals since most of them are without side effects when compared with synthetic drugs. Each plant whether it may be shrub, herbs, algae have its own significance in pharmaceutical, medicinal, agricultural, industrial, biochemical and chemical sciences. The present investigation was focused on the preliminary phytochemical and Fourier Transform Infrared Spectral analysis and Antimicrobial Studies of solvents extracts of *Spatholobus purpureus* Benth. ex Baker results were clearly revealed that the plant contained different bioactive compounds such as of Alkaloids, Anthoquinones, Coumarins, Steriods and Flavonoids compounds were rich in the extracts of *Spatholobus purpureus* Benth. ex Baker are connected with defense mechanism against many microorganisms.

Citation: **Deshmukh, O.S. and Makode, K.U.2019.** "Studies on the phytochemical, spectroscopic characterization and antibacterial efficiency of spatholobus purpureus benth. ex baker (family- fabaceae)", *International Journal of Current Research*, 11, (01), 824-827.

INTRODUCTION

There has been an increasing interest worldwide on therapeutic values of natural products. The nature provides the mankind vast therapeutic flora with a wide variety of medicinal potential. The revival of interest in plant derived drugs is mainly due to the current widespread belief that "green medicine" is safe and more dependable than the costly synthetic drugs many of which have adverse side effects. In recent years, photochemical in vegetables have received a great deal of attention mainly on their role in preventing diseases caused as a result of oxidative stress which releases reactive oxygen species such as singlet oxygen and various radicals as a damaging side effect of aerobic metabolism. Characterization of extracts of medicinal plants is necessary, due to its numerous benefits to science and society. The information obtained, makes pharmacological studies possible. It also enabled structure-related activity studies to be carried out, leading to the possible synthesis of more potent drug with reduced toxicity. The mode of action of the plants producing the therapeutic effect can also be better investigated if the active ingredients are characterized. The use of phytochemicals as natural antimicrobial agents, commonly called 'biocides' is gaining popularity (Smid, 1999). The most essential of these bioactive constituents of plants are alkaloids, tannins, flavonoids and phenolic compounds. *Spatholobus purpureus* Benth. ex Baker are an attractive,succulent medicinal plant of the family Fabaceae. They are medicinal impotant plants distributed in Etawa forest of Betul District, Madhya Pradesh, India (Karuppusamy, 2013). Plants produce bioactive molecules in a diverse range making them a rich source of different types of medicines (Kala, 2011; Joselin, 2012; Florence, 2012 and Florence, 2014). Ramamoorthi and Kannan screened the bioactive group of chemicals in the dry leaf powder of *Calotropis gigantea* by FTIR analysis (Ramamurthy, 2007). Kareru *et.al*. detected saponins in crude dry powder of 11 plants using FTIR spectroscopy (Kareru, 2008). The most essential of these bioactive constituents of plants are alkaloids, tannins, flavonoids and phenolic compounds. Many of the indigenous medicinal plants are used as spices and food (Amin Mir, 2013). Traditionally herbal extracts were known to be effective against microorganisms as a result; plants form the basis of modern medicine. Plants produce phytochemicals to protect themselves; but recent studies indicate that many phytochemicals can also protect

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humans against infectious diseases (Jeeva, 2006; Domettila, 2013; Balakumar, 2011 and Rajan, 2011). Antimicrobial activity and phytochemical screening of *Spatholobus purpureus* root, in this study suggest that this plant can be used as an antimicrobial agent and expected that these may be used as therapeutic agents for various diseases (Sharma Yash, 2015).

MATERIALS AND METHODS

Plant Collection: *Spatholobus purpureus (Fabaceae)* were collected from Etawa forest area of Betul district , Madhya Pradesh , India. Before picking the plant, the soil was moisted and then shade dried, were ground to get a course powder that was stored in airtight, high density poly ethylene container.

Spatholobus purpureus –Habit

Spatholobus purpureus - Root

Source and extraction: The organic solvent extract was prepared by adding 5 gm powder of ethno veterinary medicinal plants in 250 ml of organic solvent(Absolute Alcohol, Acetone, Petroleum Ether, Benzene, chloroform and Distil Water) for 6 hrs. by soxlhet method and filtrate was evaporated in controlled conditions of temperature of active constituents of preparations. Dried extracts were stored in labeled sterile wide mouthed screw capped bottle at 4^0c and used for further study.

Phytochemical screening: Phytochemical screening were performed to assess the qualitative chemical composition of different crude extracts using commonly employed precipitation and coloration reactions ,the methods of Harbone (Harborne, 1973), Trease and Evans (Trease, 1983) were used to identify the major secondary metabolites like Alkaloids, Flavonoids, Saponins, Carbohydrate, Protein, Phenols, Steroids, Tannins, Glycosides, Terpenoids, Phlobatannins, Coumarins, Emodins, Anthoquinones, Anthocyanins, Leucoanthocyanins in the extracts.

Antimicrobial screening: The standard pathogenic bacterial cultures were procured from IMTECH, Chandigarh, India. The bacteria rejuvenated in Nutrient broth (Hi-media – laboratories, Mumbai, India) at 37^0c for 18 hrs. and then stored at 4^0c on Nutrient agar subcultures were prepared from the stock for bioassay. Bacterial Pathogen used in study are, *E. coli, Pseudomonas fluroscene ,Salmonella typhi Bacillus subtilis, Klebsiella pneumoniae ,Staphylococcus aureus, Streptococus* Yeast candida, *Aspergillus niger.* The disc-diffusion test is based on the fact that for a given sample of plant extract. Antimicrobial susceptibility testing with discs is a simple and rapid method and provides a reproducible means of testing bacterial sensitivity to various antibiotics and chemotherapeutic agents.

Infrared (IR) spectroscopy analysis: This was done using infrared spectrophotometer of Shimadzu Corporation of model IR prestige 21. The extracts were scanned in accordance with ASTM 1252-98. A drop of each extract was applied on a sodium chloride cell to obtain a thin layer.

RESULTS AND DISCUSSION

Alkaloids: It was found that concentration of alkaloids have been extracted in Ethanol and Acetone extract. This is evident from the positive test with Hager's reagent. Benzene, Chloroform, Petroleum ether and Distil water extracts have shown negative test for Alkaloids with Hager's reagent.

Phenols: All extracts have shown positive test for Phenols.

Saponins: All extracts have shown positive test for Saponins.

Tannins: It was found that concentration of Tannins have been extracted in Ethanol and Acetone extracts. This is evident from the positive test with Braymer's reagent. Benzene, Chloroform, Petroleum ether and Distil water have shown negative test for Tannins.

Flavonoids: It was found that concentration of Flavonoids have been extracted in Ethanol and Acetone extracts. This is evident from the positive test. Benzene, Chloroform, etroleum ether and Distil water extracts have shown negative test for Flavonoids.

Terpenoids: It is found that concentration of Terpenoids have been extracted in Ethanol and Acetone extract. This is evident from the positive test. Benzene, Chloroform, Petroleum ether and Distil water extracts have shown negative test for Terpenoids.

Steroids-It is found that concentration of Steroids have been extracted in Ethanol, Acetone and Petroleum ether extract. This is evident from the positive test with Salkowski reagent. Benzene, Chloroform and Distil water extracts have shown negetive test for Steroids.

Coumarins-It is found that concentration of Coumarins have been extracted in Ehanol, Benzene, Chloroform, Acetone and Petroleum ether extract. This is evident from the positive test. Distil water extract have shown negative test for Coumarins.

Carbohydrates: It is found that concentration of Carbohydrates have been extracted in Ethanol and Acetone extracts. This is evident from the positive test with Molisch reagent. Benzene, Chloroform, Petroleum ether and Distil water extracts have shown negative test for Carbohydrates.

Table 1. Preliminary phytochemical screening of various extracts of *Spatholobus purpureus*

Present -- +ve Absent -- -ve

Plant parts	Test / Reagents Used	Ethanol extract E	Benzene extract B	Chloroform Extract C	Acetone extract A	Petroleum Ether P	Distil Water extract W
Root	Alkaloids (Hager's Test)	+	–	–	+	–	–
	Glycosides (Libermann's Test)	–	–	–	–	–	–
	Phenols	+	+	+	+	+	+
	Saponins (Foam Test)	+	+	+	+	+	+
	Tannis (Braymer's Test)	+	–	–	+	–	–
	Flavonoids	+	–	–	+	–	-
	Terpenoids	+	–	–	+	–	–
	Steroids (Salkowski Test)	+	–	–	+	+	–
	Phobatannins (Precipitate Test)	–	–	–	–	–	–
	Coumarins	+	+	+	+	+	–
	Proteins (Xanthoproteic Test)	–	–	–	–	–	–
	Emodins	–	–	–	–	–	–
	Carbohydrates (Molisch Test)	+	–	–	+	–	–

Table 2. Antimicrobial activity of root extracts of *Spatholobus purpureus* Benth. ex Baker

Sr. No.	Plant Extract	*E. coli*	*Pseudomonas fluroscene*	*Salmonella typhi*	*Bacillus subtilis*	*Klebsiella pneumoniae*	*Staphylococcus Aureus*	*Strepto cocus*	Yeast candida	*Aspergillus niger*
1	Ethanol	++	–	–	+ +	+	–	–	++	–
2	Petroleum Ether	+++	–	–	+++	–	–	–	–	–
3	Acetone	-	–	–	–	–	–	–	–	–
4	Benzene	-	++	++	++	–	–	–	–	–
5	Chloroform	+++	–	–	–	++	–	–	–	–
6	Distilled Water	+	–	–	+++	–	++	+	–	–

N.B. ─ Inactive (Resistance) , + Active (Zone of Inhibition)

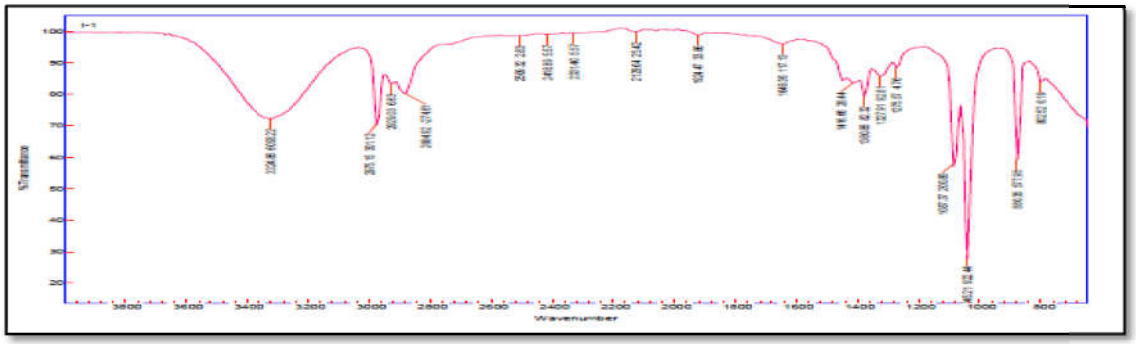

Fig. 1. FT-IR spectrum of *Spatholobus purpureus* in Ethanol extract

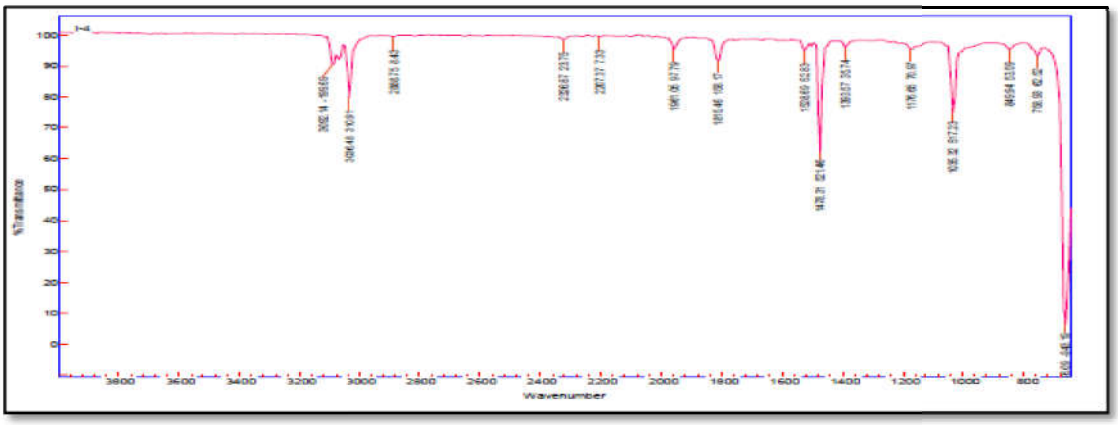

Fig. 2. FT-IR spectrum of *Spatholobus purpureus* in Benzene extract

IR interpretation

It exhibited characteristic band at 3324.86 cm^{-1} shows the presence of N-H stretch (Primary, secondary Amine, amide) and at 2975.15 cm^{-1} shows the C-H stretching (Alkanes), at 1416.66 cm^{-1} shows the C-C stretching(Aromatic amine), at 1087.37 cm^{-1} shows C-N stretching(Aliphatic amine), at 945.21 cm^{-1} shows O-H bend (Carboxylic acid), at 802.52 cm^{-1} shows C- Cl stretching(Alkyl halides), at 880.35 cm^{-1} shows N-Hl way (primary, secondary amine) in Ethanolic extract. It exhibited characteristic band at 3092.14 cm^{-1} shows the presense of C-H stretching (Aromatic) and at 3036 cm^{-1} shows the C-Hstretching (alkanes), at 1478.31 cm^{-1} shows the C-C stretching (Aromatic), and at 1035.32 cm^{-1} shows C-N stretching (aliphatic amine), at 758.93 cm^{-1} shows C-Cl stretching (alkyl halide) in Benzene extract.

From table 2 it is clear that root extracts of *Spatholobus purpureus,* **t**he ethanol extract showed activity against *E. coli, Bacillus subtilis, Klebsiella pneumoniae, Yeast candida* and inactive against *Pseudomonas fluroscene, Salmonella typhi, Staphylococcus aureus, Streptococus, Aspergillus niger*. The petroleum ether extract showed activity against *E. coli, Bacillus subtilis* and inactive against all remaining pathogens. The benzene extract showed activity against *Pseudomonas fluroscene, Salmonella typhi and Bacillus subtilis*. The Chloroform extract showed activity against *E. coli, Klebsiella pneumonia.* The distilled water extract showed activity against *E. coli, Bacillus subtilis, Staphylococcus aureus, Streptococus.*

Conclusion

Our results were clearly revealed that the plant contained different bioactive compounds such as of Alkaloids, Anthoquinones, Coumarins, Steriods and Flavonoids. These compounds were rich in the extracts of *Spatholobus purpureus* (Fabaceae) are connected with defense mechanism against many microorganisms. These plants have antimicrobial activity against some gram positive and gram negative bacteria such as, *E.coli, Pseudomonas fluroscene, Salmonella typhi Bacillus subtilis, Klebsiella pneumoniae, Staphylococcus aureus, Streptococus, Yeast candida*, *Aspergillus niger.* Ethanolic extract showed good antimicrobial activity against tested bacterial strains such as *Escherichia coli, Staphylococcus aureus, and Bacillus subtilis Klebsiella pneumoniae, Yeast candida, Salmonella typhi*. Thus these plants can be utilize as an alternative source of useful drug.

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