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PHARMACOLOGICAL STUDIES ON ROOTS OF RUELLIA TUBEROSA L

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Abstract

It is well known fact that the plants have ability to prepare different chemical compounds for different biological functions. In India the plants are used on large scale for different diseases considering these facts Ruellia tuberosa belonging to family Acanthaceae is selected for the present research work. Ruellia tuberosa is a small herb with fasciculate fleshy swollen roots. The plant is found to be with therapeutic effects as a natural remedy for constipation, antidote for poisons, leprosy, rheumatism, hemorrhoids and burning sensation. Many chemicals like Alkaloids, steroids, Phenolics, terpenoids and saponins are isolated from the roots of Ruellia tuberosa. The root extract is found to be contains different pharmacological properties. The present studies attempt the traditional uses, phytochemical analysis and pharmacological characters of roots of Ruellia tuberosa.

Introduction

The roots of *Ruellia tuberosa* are the tap roots. They are fasciculate tap roots. The roots are found to be contain many chemical compounds like alkaloids, tannins and coumarin. It is clear from the literature that these compounds have high pharmaceutical values. They serve as protective mechanism to the plants. They are found to be stored in the wood and bark of the roots. The compounds like tannins are found to be resistant to proteolytic

***Ruellia tuberosa* L.**

enzymes. They are found to be used in the Indian system of medicine as astringent to intestinal tract. The roots are proved to be good astringent and can be used in diarrhea, dysentery, ulcers, piles and fissures. The roots of *Ruellia tuberosa* are not so popular like the vegetables in food and the nutraceutical. They contain starch and claims for food values. Considering these facts studies were undertaken for the pharmacological characters and phytochemical analysis of roots of *Ruellia tuberosa* L.

Material And Methods

1. Study of Fluorescence Characters of roots of Ruellia tuberosa L. using different solvents

During the present studies the roots of *Ruellia tuberosa* were collected from the campus of Yeshwant Mahavidyalaya Nanded. They were cleaned and washed under tap water. They were dried in shade powdered and named as drug powder of roots of *Ruellia tuberosa*. The powder was packed in polythene bags and stored in refrigerator and it is used for the study of fluorescence Characteristics. For this 1 gm of drug powder was dissolved in 10 ml of different solvents. The test tubes containing the dissolved drug powder were observed in day light and Uv light at 254 nm for colours.

2. Study of ash values and exhaustive extractive values of Ruellia tuberosa L.

a) Determination of ash content: Ash of the crude drugs contain inorganic radicals like phosphate, carbonates, silicates of sodium, potassium, magnesium and calcium etc. The inorganic constituents like calcium oxalate, silica and carbonate contents of the crude drug affects the total ash value. These variables are then removed by treating with acid as they are soluble in HCl and then acid insoluble ash value was determined.

b) Determination of total ash value : The residue remaining after incineration is the ash content of the drug. 10 gm of powdered drug added and incinerated in silica crucible over the burner. The burned material was heated up to 500-600 ºC for six hours in muffle furnace. The residue remaining after incineration of sample is known as ash. The crucible is cooled in desiccators. The percentage of total ash content was calculated with reference to the air dried drug.

Where,

X- Weight of empty petridish

Y- Weight of used crude drug

Z- Weight of ash with petridish

Weight of total ash (A) = Weight of ash with petridish(Z) - Weight of empty petridish (X)

So , A= (Z-X) gm

Y gm of crude drug gives (Z-X) gm of the ash

100 gm of the crude drug gives $\frac{100}{Y}$ × (Z-X) gm of the ash.

Crude drug gives

Total ash value of sample = $\frac{100(Z-X)}{Y}$ %

c) Determination of acid insoluble ash : It is a part of total ash which is insoluble in dilute HCl. The ash was boiled for 5 min. in 25 ml of dilute HCl and after cooling the material was filtered through an ash less filter paper and wash with hot water. The material ignited and weighed. The percentage of acid insoluble ash was calculated with reference to the air dried drug sample.

Where,

X- Weight of empty petridish

Y- Weight of used crude drug

Z- Weight of ash with petridish

Weight of total ash (A) =Weight of ash with petridish(Z) - Weight of empty petridish (X)

So , A= (Z-X) gm

Y gm of crude drug gives (Z-X) gm of the ash

100 gm of the crude drug gives $\frac{100}{Y}$ × (Z-X) gm of the ash.

'a' gm = Weight of residue (acid insoluble ash)

Y gm of the air dried drug gives 'a' gm of acid insoluble ash.

Acid insoluble ash value of the sample = $\frac{100\times a}{Y}$ %

d) Determination of water soluble ash : The ash was boiled for 5 min. with 25 ml of distilled water and after cooling the material was filtered through an ash less filter paper and washed with hot water and then the ash paper was ignited and the ash weighed.The weight of the insoluble matter was substracted from the weight of the ash.The difference in weight indicates the water soluble percentage. It was calculated with reference to the air dried drug sample.It is determined in a similar way to acid insoluble ash.

e) Determination of extractive values : The extracts obtained from the plant parts are indicated the approximate percentage of certain chemical constituents present in them. So the

extractive values are also useful for the evaluation of the crude drug. It is also useful for the estimation of specific constituents which are soluble in different solvents.

f) Water Soluble of extractives: 5 gm of powdered drug was macerated with 100 ml of distilled water in conical flask for 24 hrs with shaking at regular intervals. The solution was filtered and 25 ml of filtrate was evaporated in thin porcelain dish. Evaporate to dryness on a water bath and complete the drying in an oven at 100 ºC,cooled in desiccators and weighed. The percentage of extractives was calculated with reference to the air dried drug sample.

25 ml of Water extractives gives = X gm of residue

100 ml of Water extractives gives = 4X gm of residue

So, 5 gm of air dried drug gives = 4X gm of Water soluble residue

100 gm of air dried drug gives = 80 X gm of water soluble residue

water soluble extractive value of the sample= 80X %

g) Determination of alcohol soluble extractive: 5 gm of powdered drug was macerated with 100 ml of distilled water in conical flask for 24 hrs with shaking at regular intervals. The solution was filtered rapidly, taking precaution about loss of alcohol and 25 ml of filtrate was evaporated and weighed in thin porcelain dish and dried at 100 ºC and last weight is taken. The percentage of alcohol soluble extractives was calculated with reference to the air dried drug sample.

25 ml of alcohol extract gives = X gm of residue

100 ml of alcohol extract gives = 4X gm of residue

So, 5 gm of air dried drug gives = 4X gm of alcohol soluble residue

100 gm of air dried drug gives = 80 X gm of water soluble residue

water soluble extractive value of the sample= 80x %

3. Qualitative Phytochemical Analysis of root of *Ruellia tuberosa* L.:

a. Preparation of powder: The plant roots of *Ruellia tuberosa* L. were collected from the campus of Yeshwant Mahavidyalaya Nanded were washed with water and shed dried. The dried roots were powdered in grinder and were stored in air tight containers for further use. The Preliminary phytochemical screening was performed according to Johansen (1940) and Harborne (1984).

b. Preparation of Plant extracts: 10 gm of dried powdered samples were successively extracted with 100 ml of different solvents of various polarity was carried out as Petroleum ether (60 to 80ºC) Ethanol (60 to 80ºC), Methanol (65.5 to 70.5ºC), Acetone (56 to 60ºC)

and distilled water (80 to 100ºC) using Soxhlet apparatus. The extract were filtered and evaporated in hot water bath and weighed.

c. Qualitative Phytochemical Analysis:

Prepared extract of root were subjected for phytochemical screening by dissolving 1gm of extract in 10 ml of the different reagents and the phytochemical were detected.

1. Test for Alkaloids: For this finding 1gm of root extract was dissolved in 10 ml of dilute HCl and filtered. The filtrates were used for the following test :

Dragendroff's Test: In this test 1 ml of Dragendroff's reagent added to the above filtrate, the orange brown precipitate was observed which indicates the presence of Alkaloids. (The results shown in Table-2)

2. Test for Steroids: Salkowski reaction: To 2ml of extract, add 2 ml chloroform and 2 ml of conc.H_2SO_4.Shake well. Chloroform layer appears red and acid layer shows greenish yellow flurescence.

3. Test for Coumarin: Take moistened dry powder in test tube. Cover test tube with filter paper soaked in dilute NaOH. Keep in water bath. After sometime expose filter paper to UV light. It shows yellowish-green fluorescence**.** But here no change is observed so the test is negative for coumarin.

4. Test for Tannin: To 2-3 ml of aqueous or alcoholic extract, add few drops of dilute HNO_3 you will get reddish to yellow ppt.Test is positive for Tannins.

5. **Test for Saponins: Foam test:** Shake the drug extract or dry powder vigorously with water. Persistent foam observed.

6. **Test for Flavonoids:** Heat test solution with zinc and HCl, pink to red colour is observed.

7. **Test for Quinones:** Borntrager's Test: To 3 ml extract, add dil. H_2SO_4.Boil and filter. To cold filtrate, add equal volume of chloroform. Shake well. Separate the organic solvent. Add ammonia. Ammoniacal layer turns pink or red.

8. **Test for Phenols:** To 2-3 ml of aqueous or alcoholic extract, add few drops of dilute acetic acid solution you will get red colour solution. Test is positive for Phenols.

9. Test for Gums: Hydrolyze test solution using HCl. Perform Fehling's test as mix 1 ml Fehling's A and Fehling's B solutions, boil for 1 min. add equal volume of test solution. Heat in boiling water bath for 5-10 minutes. then brick Red colour ppt. is observed. But here no such colour is observed.

10. **Test for Proteins:** Millon's test: Mix 3 ml of test solution with 5 ml Millon's reagent. White ppt. warm ppt. turns brick red coloured solution. But here no such colour is observed.

11. Test for Glycosides and sugars: Determine free sugar content of the extract. Hydrolyse the extract with mineral acid (dil.HCL/dil.H_2SO_4). Again determine the total sugar content of the hydrolised extract. Increase in sugar content indicates presence of glycoside in the extract.

12. Test for Fats and oils: Solubility test: Oils are soluble in ether, benzene and chloroform, but insoluble in 90% ethanol and water. Such solubility is not observed here so test is negative.

Experimental Results

1. Study of Fluorescence Characters of roots of Ruellia tuberosa L. using different solvents

During the present studies the roots of *Ruellia tuberosa* were collected from the campus of Yeshwant Mahavidyalaya Nanded. They were cleaned and washed under tap water. They were dried in shade powdered and named as drug powder of roots of *Ruellia tuberosa*. The powder was packed in polythene bags and stored in refrigerator and it is used for the study of fluorescence Characteristics. For this 1 gm of drug powder was dissolved in 10 ml of different solvents. The test tubes containing the dissolved drug powder were observed in day light and Uv light at 254 nm for colours. The results are presented in table-1

Table-1: Study of Fluorescence Characters of roots of *Ruellia tuberosa* using different solvents

Sr. No.	Solvent	Day light	UV light 254 nm
1.	Drug Powder	Pale green	Yellowish green
2.	Drug Powder+ 1N NaOH (Alcoholic)	Green	Yellowish green
3.	Drug Powder + 50% Sulphuric acid	Green	Yellowish green
4.	Drug Powder + Acetic acid	Pale green	Yellowish green
5.	Drug Powder + 1N HCl	Pale green	Green
6.	Drug Powder + 5% Iodine	Pale green	Green
7.	Drug Powder + Ferric chloride	Reddish brown	Yellowish green
8.	Drug Powder + 50 % Nitric acid	Reddish Orange	Yellowish green
9.	Drug Powder + HNO_3 + NH_3	Reddish Orange	Pale green
10.	Drug Powder+ 1N NaOH (Aqueous)	Yellow	Yellowish green
11.	Drug Powder + Picric acid	Yellow	Yellowish green

From the results presented in Table-1 it is clear that the drug powder is found to be pale green in day light and yellow green in UV light. It was interesting to know that the drug powder dissolved in aqueous 1N NaOH and in picric acid was found to be yellow in day light

and yellow green in UV light.The drug powder dissolved in alcoholic 1N NaOH and 50% Sulphuric acid was found to be green in day light and yellowish green in UV light.

The drug powder dissolved in 1N HCl, Acetic acid and Iodine was found to be pale green in day light and green, yellowish green and green in UV light respectively. The drug powder dissolved in 50% Nitric acid and HNO_3 +NH_3 was found to be reddish orange in day light and yellowish green and pale green in UV light respectively.

The drug powder of root of *Ruellia tuberosa* dissolved in Ferric Chloride was found to be reddish brown in day light, yellowish green in UV light.

Table-2: Ash values and exhaustive extractive values of Ruellia tuberosa L.

Sr. No.	Parameters	Ash values of roots of Ruellia tuberosa L. (%)
1.	Total Ash	6.87
2.	Water soluble ash	2.28
3.	Alkalinity of water soluble ash	0.34
4.	Acid soluble ash	0.03
5.	Alcohol soluble extractive	2.27
6.	Water soluble extractive	6.62

2. Study of ash values and exhaustive extractive values of roots Ruellia tuberosa L.

During the present studies the ash values were determined by different methods as described in RRL, Jammu, IDMA. Vol-I (1998). The results are presented in table-2

From the results presented in table-2 it is evident that the acid soluble ash of roots of Ruellia tuberosa L. showed very less percentage of ash value. The water soluble extractive ash showed very high ash value as compared to the other parameters.

Table-3: Qualitative Phytochemical Analysis of root of *Ruellia tuberosa* L .

Sr. No.	Phytochemicals	Phytochemical in roots of *Ruellia tuberosa* L.
1.	Alkaloids	+
2.	Coumarin	-
3.	Fats and oils	-
4.	Flavonoids	+
5.	Glycosides and sugars	+
6.	Gum	-
7.	Phenols	+
8.	Proteins	-
9.	Quinones	-
10.	Saponin	+
11.	Steroids	+
12.	Tannin	+

(+ Represents the presence of phytochemical, - Represents the absence of phytochemical)

3. Qualitative Phytochemical Analysis of root of *Ruellia tuberosa* L.

During the present studies the roots of *Ruellia tuberosa* were collected from the campus of Yeshwant Mahavidyalaya Nanded. They were cleaned and washed under tap water. They were dried in shade powdered and named as drug powder of roots of *Ruellia tuberosa* L.. The root extracts were prepared in different solvents and screened for different qualitative phytochemical analysis tests. The results are presented in table-3

From the results presented in table-3, it is clear that the roots of *Ruellia tuberosa* L. the presence of all the phytochemicals except Coumarin, Quinones, gum,proteins, fats and oils.

Discussion

Qualitative phytochemical screening and quantified total Phenolics and alkaloid contents of whole plant of *Ruellia tuberosa* L. revealed the presence of steroids, triterpenoids, Glycosides, saponins, tannins, alkaloids, phenols and carbohydrates. The methanolic extract shows higher amount of alkaloid and the ethanolic extracts has more phenolic compounds and antioxidant activity was found to be more in methanolic extract.

Phytochemical analyses of *Ruellia tuberosa* L.were performed. Result of phytochemical analysis of these extract revealed the presence of tannins, steroids, saponins, alkaloids, phenols, proteins, glycosides and sugars. The observations made in the present study have clearly showed the bioactive potential of the plant *Ruellia tuberosa* L in medicine. The leaves sample exhibited flurescence characteristics, ash values and extractive values. Leaves sample exhibited qualitative phytochemical properties. The work on similar lines was carried out by different workers with reference different medicinal plants. Amerjothy (2006) carried out phytochemical studies on Indian plant galls.It was found that the plant gall tissues are rich in IAA, Phenolics and proteins. Kalpesh et. al.(2012) studied antimicrobial activity and phytochemical analyses of Calotropis gigantea (L.) R. Br. Latex against selected cariogenic bacteria. They found that the test plant extract showed the presence of tannins, alkaloids sugars and glycosides. Mani ((1964) studied ecology of plant galls and suggested that the fundamental principles governing the gall morphogenesis and the normal morphogenesis are the same. Peach and Tracoy (1955) described modern methods of plant analysis. Sharmaet et. al. (2012) studied phytochemical evaluation and quantification of primary metabolites of Cassia pumila Lamk.

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