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Research Article

PHYSICO PHYTO-CHEMICAL EVALUATION OF THE ROOT OF KANOTI (*Linaria ramosissima* Wall. Janch.)

Preeti Pandya[1], Rabinarayana Acharya[2], Shukla VJ[3], Renuka Kalyani[4], Komal Khanpara[5]

1. *Ph. D. Scholar, Dept. of Pharmacognosy, PGT-SFC, I.P.G.T. & R.A., Gujarat Ayurved University, Jamnagar, Gujarat, India.*
2. *Associate Professor, Dept. of Dravyaguna, I.P.G.T. & R.A., Gujarat Ayurved University, Jamnagar, Gujarat, India.*
3. *Head, Dept. of Pharmaceutical chemistry, I.P.G.T. & R.A., Gujarat Ayurved University, Jamnagar, Gujarat, India.*
4. *Ph. D. Scholar, Dept. of Pharmaceutical chemistry, I.P.G.T. & R.A., Gujarat Ayurved University, Jamnagar, Gujarat, India.*
5. *Ph. D. Scholar, Dept. of Pharmaceutical chemistry, I.P.G.T. & R.A., Gujarat Ayurved University, Jamnagar, Gujarat, India.*

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Abstract

Kanoti (*Linaria ramosissima* Wall. Janch) from the family Scrophulariaceae, also known as Bhintgilodi, is a reputed folklore drug. The plant is reported for its antidiabetic property specially root is used in the management of snake and scorpion bite. In the present study, root was evaluated, following the standard methods, for physicochemical parameters, flourescence analysis, qualitative tests, quantitative estimation of alkaloid & saponin and HPTLC study. Qualitative tests for root confirms presence of alkaloids, glycosides, tannins & flavonoids and total Saponins 1% w/w, and 1.2% w/w alkaloids quantitatively, while characteristic colour flourescence in different solvents with different wavelengths was observed. HPTLC profile shows 5 spots at 254nm, 4 spots at 366nm and 3 spots after spraying. The generated information of the present study will provide data which is helpful in the identification and authentification of the drug and also can be used for the further research.

Keywords: Kanoti, *Linaria ramosissima* Wall. Janch, Bhintgilodi, Scrophulariaceae, Phytochemistry

***Address for correspondence:**
Pandya Preeti,
Ph. D. Scholar, Dept. of Phamacognosy,
PGT-SFC, I.P.G.T & R.A.,
Gujarat Ayurved University, Jamnagar, Gujarat, India – 361008.
E-mail: preeti_ayu@yahoo.co.in

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INTRODUCTION

Linaria ramosissima wall. Janch. commonly known as kanoti is a folklore medicine plant highly effective in Diabetes.[1][2] The root of this plant is used in snake and scorpion bite.[3][4][5] Plant is a perennial, slender hairy herb, found throughout India in stony and rocky places and on ruined and old walls of forts and buildings.[1]-[5] The root[6] and stem[7] of this plant has been evaluated for their pharmacognostical characters. The phytochemical characters of the roots are not reported yet. Present study aims to carry out the phytochemical, physicochemical, qualitative and quantitative screening, fluorescence analysis and HPTLC fingerprinting of the root of the plant.

MATERIAL AND METHODS

Collection of plants

The plant was collected when it was in full bloom in rainy season, from the places located around Jamnagar and the authentication of the drug was confirmed by comparing the plant with different floras[1,2] and taxonomic experts of Gujarat Ayurved University and by comparing their Macroscopic characters mentioned in various floras. The plants were washed, dried, powdered and taken for further study. Herbarium was also prepared and submitted to Pharmacognosy museum of I.P.G.T. & R.A., Jamnagar, vide Herbarium no. 6015 for future reference.

Physicochemical parameters

Physicochemical study includes the parameters such as Loss on drying, Ash value, Acid insoluble ash, Extractive values in water and methanol, pH according to the various textual references.[8]

Qualitative Study

In qualitative analysis various chemical tests were performed to find out the presence or absence of chemical constituents like Saponins, Alkaloids, Glycosides etc.[9]

Quantitative Estimation

For quantitative evaluation of the Phytoconstituents the assay was carried out for total Alkaloids and total Saponins content by gravimetric method according to the references.[10]

Fluorescence Analysis

The fluorescence analysis of the plant drug powder was done in different solvent extracts. For this chloroform, methanol, water, hydrochloric acid, sulphuric acid and sodium hydroxide were taken as solvents. The extracts of the plant was prepared in selected solvents. All the extracts were subjected to long UV 366 nm and short UV 254 nm to observe the difference in colour.[11]

Chromatographic fingerprint

Preparation of sample solution

10 g of plant material was extracted with 5 ml of Methanol. The material was refluxed for half an hour. The extract was filtered and volume was made up to 10 ml to get solution concentration.[12]

Experimental Conditions

The methanolic extract of root was examined for HPTLC using the solvent system of Ethyl acetate: Dichloromethane: Glacial acetic acid in ratio of 7:2:1 v/v/v & stationary phase silica gel (Procoated Silica Gel TLC plates of merck) with the saturation time of 30 mins. The developed plate was observed under wavwlength of short UV 254nm and long UV

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366 nm. Spraying agent Vanillin H_2SO_4 was used for derivatization and visualization and then observed under day light with the conditions like, Camag Linomat V Application mode, Camag Twin trough Development Chamber, 30 min Chamber Saturation time, 30 min Development Time, Camag Scanner II,. Deuterium lamp & Tungstun lamp for Detection, and Win cats software Data System under the UV Spectrum scanning range of 200 to 700nm.[13]

RESULTS

The organoleptic characters of drug are shown in Table 1. Physicochemical analysis was carried out like, Loss on drying, Ash value, and the extractive values with different solvents and the results obtained are given in Table 2. In the flourescence analysis the plant root powder was treated with different solvents for observing the characteristic radiations. Sample when subjected to 254 nm and 366 nm and in visible radiation gives particular radiations which could be used as diagnostic characters and noted down in Table 3. Qualitative tests for various plant primary and secondary metabolites were detected and given in Table 4. While amongst detected metabolites total saponins and alkaloids were also detected in root. (Table 5) HPTLC profiles of methanol extract of the root showed distinct characteristic with common spot with similar Rf values as well as different values also noted down in Table 6. (Figure 3-7)

Table 1: Organoleptic characters of the root of *Linaria ramosissima* Wall.

Characters	Observation
Colour	Brown
Odour	Not distinct
Taste	Astringent, Bitter
Texture	Coarse, dull

Table 2: Physico-chemical parameters of the root of *Linaria ramosissima* Wall.

Sr.	Test	Sample D
1	Loss on Drying at 110 C % w/w	6.66
2	Ash Value % w/w	10.64
3	Acid insoluble Ash % w/w	0.94
4	Water Soluble extractive Value % w/w	14.76
5	Methanol Soluble extractive value % w/w	15.06
6	P^H	5.85

DISCUSSION

Organoleptic characters of sample shows brown colour, astringent & bitter taste while no any characteristic odour for the plant root powder. Physicochemical analysis was carried out to establish the standard quality parameters for the drug which can be useful for further research, identification and authentification and also for creating drug profile. For fluorescence analysis the plant root powder was treated with different solvents for observing the characteristic radiations if there is any at particular wavelengths of the light which is particular for every drug and helpful parameter for generating plant research profile. Fluorescence study of powder with different solvents also revealed distinguished colour characteristics treatment with sodium hydroxide, hydrochloric acid, methanol and chloroform showed diagnostic colour for powder of when subjected to 254 nm and 366 nm which could be used as diagnostic characters. Qualitative tests for various plant primary as well as secondary metabolites showed the presence of constituents like alkaloids, glycosides, saponins, tannins and flavanoids etc. While estimation of some detected phytochemical constituents confirmed amount of Total saponins 1% w/w, and 1.2% w/w alkaloids in root.

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Table 3: Flouroscence analysis of the root of *Linaria ramosissima* Wall.

Material	Visible Light	UV Light	
		Short (254 nm)	Long (366 nm)
Powder as Such	Brown	Flouroscent green	Flouroscent green
Aqueous extract.	Light brown	Brown	Brown
Methanol extract	Light yellow	Light Yellowish green	Light yellow
Chloroform extract	Olive green	Color less	Light reddish
Powder + Hcl	Dark reddish brown	Fluorescent green	Light grey
Powder + H_2SO_4	Dark reddish brown	Light greenish grey	Dark fluorescent blue
Powder + NaoH	Yellowish brown	Brown	Fluorescent green

Table 4: Preliminary Phytochemical analysis of the root of *Linaria ramosissima* Wall.

Test/ Reagent	Functional group	Observation	Result
Dragendorff's reagent	Alkaloids	Orange Brown ppt	+ve
Wagner's reagent	Alkaloids	Reddish brown ppt	+ve
5% fecl3	Tannin & Phenolic compd.	Deep blue black color	+ve
Borntrager's test	Antnraquinone glycosides	No color change	-ve
Biuret reagent	Protein	No color change	-ve
Fehling's test	Carbohydrate	First yellow, then brick red ppt observed	+ve
Liebermann-buchard	Steroids	First red, then blue and finally green color appears	+ve
Lead Acetate	Flavonoids	Yellow ppt	+ve
Shaking in test-tube	Saponins	Frothing with honeycomb appearance	+ve

(+ve : Present; -ve : Absent; ppt : precipitate)

Table 5: Quantitative Estimations of the root of *Linaria ramosissima* Wall.

Sample	Saponin content (%w/w)	Alkaloid content (%w/w)
Root	1.2	1

Table 6: HPTLC profile of the methanolic extract of the root of *Linaria ramosissima* Wall.

Sample ID	254 nm		366 nm		After spraying with Anisaldehyde H_2SO_4	
	No. of spots	Rf value	No. of spots	Rf value	No. of spots	Rf value
MeOH extract of Root	5	0.01,0.07,0.19,0.49,0.50	4	0.01,0.07,0.17,0.90	3	0.01,0.07,0.19

HPTLC profiles of methanol extract of the root showed distinct characteristic with common spot with similar Rf values at 0.01 and 0.07. When the plate was observed at 366 nm shows 2 spots while 3 spots were detected when plate was observed in short UV (254 nm). Other than this the same Rf valued spots were observed.

When derivatisation (Vanillin Sulphuric Acid spray reaction) was carried out for identification of chemical constituents in the sample, 3 spot with same Rf values were observed in the sample. Hence, HPTLC profile could be used as a diagnostic tool to identify the plant.

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Figure 1: Natural habitat of the plant *Linaria ramosissima*

Figure 2: Root of the plant *Linaria ramosissima*

Figure 3: UV Short 254nm

Figure 4: UV Short 366nm

Figure 5: After Spraying

Figure 6: Peak display (254nm)

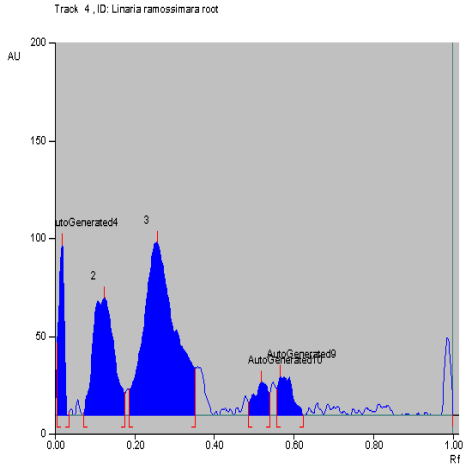
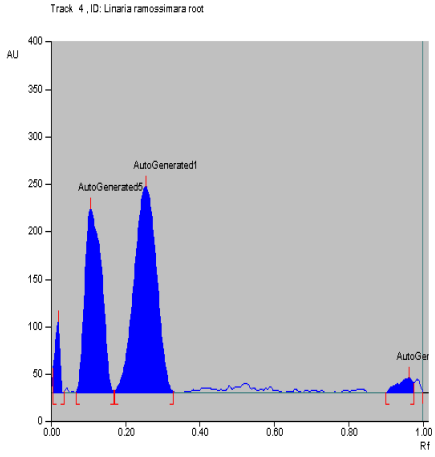


Figure 7: Peak display (366nm)



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CONCLUSION

Linaria ramosissima Wall. Janch. root reveals the presence of alkaloids, glycosides, tannins and flavonoids, in qualitative analysis. It contains a good amount of good amount of Saponin and alkaloid. Its root extracts shows characteristic colour flourescence with different solvents, at different wavelengths.

HPTLC profile shows 5, 4 and 3 spots at 254 nm, 366nm and after spraying, respectively. The observed physicochemical and phytochemical data may be useful as diagnostic tool for identification and authentification of the drug and for further research.

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