



Phytochemical Screening of Medicinal Plant *Dolichandrone falcata*

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ABSTRACT: The present study is conducted in order to discover the qualitative and quantitative analysis of the plant species *Dolichandrone falcata*. Qualitative phytochemical evaluation was carried out to test the presence of alkaloids, flavonoids, sugars, phenolic group, saponin, amino acid and essential oil in the extracts samples using modified methods. The present effort is targeted on the evaluation of the amount of total flavonoids and total phenol present in different plant parts such as bark, leaves and fruits. Estimated specific flavonoids (Rutin) was by using the method of Thin layer Chromatography. Plant species under estimation, *Dolichandrone falcata* belongs to the family Bignoniaceae.

Key words: *Dolichandrone falcata*, Phytochemical, Total flavonoid, Total phenol, Rutin.

INTRODUCTION

Phytochemistry is defined as the study of chemical composition of medicinal plants or phyto-drugs. Today we are witnessing a rapid growth of herbal drug industry and this growth story is accompanied by search of new herbal drugs. Standardized herbal extracts and phyto-chemicals are in huge demand for applied research as well as for commercial use. The identification of biologically active compounds is an essential requirement for quality control and dose determination of plant based drugs (Ganesan *et al.*, 2008).

There are many approaches for search of new biologically active principles in higher plants. One such approach is systematic screening which may result in the discovery of novel effective bio-compound. Screening techniques of biologically active medicinal compounds have been conducted on well known species of plants used in traditional medicines. Suffredini *et al.*, (2004) emphasized that approximately 20% of the plants found in the world have been submitted to biological tests for bioactive compound. Aparna *et al.*, (2009) reported that plant possess chemical constituents having antioxidant activity through 'Dolichandroside A. It a new -glucosidase inhibitor. Khalid *et al.*, (2010) mentioned that *Dolichandrone falcata* leaves extract have an anxiolytic effect. However, very little information is available on the screening of medicinal plant *Dolichandrone falcata*.

Morphology description (Habit)- *Dolichandrone falcata* commonly known as "Medhshing" in Hindi, "Mesasninghi" in Sanskrit (means look like a sheep horn, showing in Plate). It is medium size tree with dense foliage and profuse flowering, belonging to the family Bignoniaceae. Highly scented, creamish white flowers bloom in the evening and falls down in the morning (Plate). Ripe fruits of previous year remain on the tree when the flowers of next year are in bloom. Shetty and Singh, (1991) reported that it is the rare plant found in the rocky areas of forest. The bark leaves and fruits have medicinal properties. This tree should be introduced in city gardens, botanical gardens and conserved in forest area.

Ethnomedicinal uses of plant- The whole plant and its specific parts like leaves stem and roots are known to have medicinal properties. It has long history of use by indigenous and tribal people of world and India. The medicinal value of leaves and bark of this plant is also mentioned in Ayurveda. The decoction of bark is given in the treatment of nodules by the Bhil tribes of the kota district in Rajasthan. Leaves paste is mixed with Neem leaves and applied for curb swelling. Fruit paste and bark paste is applied with water three times to get relief from scorpion bite and snake bite. Leaves juices taken orally with water acts an antidote to snake bite by tribals of Kota region. In medicinal folklore Gnanavendhan, (1995) claimed that this plant have anti-snake venom activity.



Plate – Fruits and Flower of *Dolichandrone falcata*.

The information was collected by the course of ethnobotanical explorations of forests in the states of Madhya Pradesh and Bihar which revealed that it is also used in the treatment of liver disorder (Singh *et al.*, 2007). Yelne *et al.*, (2005) described that *Dolichandrone falcata* has been used in Ayurvedic system of medicine. The whole plant is used in traditional medicine; however the bark is mentioned to be the most powerful part.

Chemical compounds are responsible for medicinal activity of the plants. The documentation of rare medicinal plants has been neglected for a long time and investigations on the phyto-chemical composition of useful plants are lagging behind. Many studies on the phytochemistry of medicinal plants concentrate only on few "fashionable" species that have been marketed heavily on a global scale. There are many medicinal plants for which limited phytochemical studies have been carried out and hence intensive efforts are required in this direction. Phyto-chemical quality of any herbal remedy is ensured by identification of biological compound present in it. Keeping this thought in mind, the present study was conducted in order to discover the qualitative and quantitative nature of leaves, bark, and fruit of *Dolichandrone falcata*.

MATERIAL AND METHODS

Plant Collection and extraction procedure - Plant parts of *Dolichandrone falcata* used in the present study was obtained from their natural habitats during early summer. The plant was identified from different floras and confirmed by Herbarium of Rajasthan University, Jaipur. All samples were pulverized with the help of stone pestle and mortar. Then the fine powdered samples were stored in airtight polythene covers. Now, 0.1g of each powder sample was inserted into a conical flask and 20ml of distilled water was added. The set up was made to stand for about 30 minutes (for appropriate

extraction) and then was filtered into different conical flasks.

Phytochemical analysis- The qualitative screenings of crude drugs for their active ingredients were carried out following standard procedures (Kumar *et al.*, 2007; Gupta and Sharma, 2005) for different phytochemicals. It is mandatory to note that total phenol and total flavonoid were estimated using Spectrophotometer System: UV5704M.

Estimation of Total Flavonoids: Flavonoid compounds are widely distributed in plant parts. These are poly-phenolic compounds. They contribute to the color and flavor of plants. Flavonoids fulfill many functions including producing yellow, red and blue pigmentation in flower and other parts of the plant.

Principle: Flavonoids react with phloroglucinol in vanillin reagent in acidic medium and produce reddish brown color. It has been reported by Swain and Hillis, (1959) and suitable modifications are suggested for getting results.

Estimation of Total Phenols: Phenols, the aromatic compounds with hydroxyl groups are abundantly present in plant kingdom. They occur in all parts of the plant. Phenols are known to offer resistance to diseases and pests in plants. Phenols include an array of compounds like tannins, flavonols etc. Total phenol estimation can be carried out with the Folin-Ciocalteu reagent. **Principle:** Phenols react with phosphomolybdic acid in Folin-Ciocalteu reagent in alkaline medium and produce blue coloured complex (molybdenum blue).

Thin Layer Chromatography (TLC) - The extracts of different plant parts were prepared by soaking in methanol. 250ml of methanol was used for soaking of each 100gm of dried plant part for 12 hours. Each extract was filtered three times by whatmen filter paper. The filtrate was used for further phytochemical screening through TLC.

Percolated silica gel nested on Aluminium plates (E. Merk) with 250 µm thickness was used as stationary phase for TLC profiling. Five different types of solvent system were used as mobile phase. Reference standard solvent of Rutin was purchased. For preparation of standard solution, 10 mg of Rutin was dissolved in 100 ml methanol. 10 µl plant extracts of the test solution taken using capillary tubes was applied in triplets on percolated silica gel TLC plates. The presence of rutin was confirmed by comparing the R_f values and colour band obtained with that of the reference standard.

RESULTS AND DISCUSSION

The biologically active chemical compounds can be identified by phyto-chemical analysis. A qualitative and quantitative analysis of phyto-chemical constituents present in medicinal plant *Dolichandrone falcata* belonging to family Bignoniaceae have been carried out. Qualitative phytochemical evaluation was carried out to test of the presence of alkaloids, flavones, sugar, phenolic group and saponin in the extracts samples using modified methods. Alkaloids, tanins, flavonoids and phenol were present in all the three parts of the plant under investigation. Saponin and terpanoids were not found in leaves (Table 1). The pharmacological action of crude drug was determined by the nature of its constituents such as alkaloids, terpenoids, flavonoids, glycosides, saponins, tannins, etc. Thirty plants species belonging to 19 families have been collected from Shahrabak area (in the west of kerman province) and Mahadavi *et al.*, (2006) screened for the determination by the nature of its constituents such as alkaloids, terpenoids and flavonoids.

The identification of biologically active compound is an essential requirement for the processing of plant based herbal drugs. Characterizing compounds or biomarker is identified from the plant part to assure the identity and quality of the preparation, this need to be responsible for the therapeutic activity.

Many studies have revealed that one or two glasses of red wine daily can protect against heart disease because of the presence of flavonoids. Flavonoids occur ubiquitously in colorful fruits and vegetables. Over 4,000 different flavonoids have been described, and they were categorized into flavonols, flavones, catechins, flavanones, anthocyanidins and isoflavonoids (Hollman and Katan, 1997). Svobodova, *et al.*, (2003) suggested that phenol containing compounds can be used to prevent ultraviolet light induced damage to hair and skin due to the UV-absorbing properties of the aromatic ring of the phenol. Wine as well as various fruits and vegetables have been applied to the photo damaged and wrinkled skin in various cultures over the centuries (Kellog, 2008). The present effort is targeted on the evaluation of the amounts of total flavonoids and total phenol present in different parts of the plant which is known to be medicinal but not yet evaluated for bio-prospecting interests.

Result shows that the amount (total flavonoids and total phenolic compound) vary from different parts of the same plant species. The results of the present study point out that bark has high concentrations of total flavonoid, recorded as 2.106 mg flavonoid in 1g plant material of *Dolichandrone falcata*. Particular order is not observed in the pattern of distribution of flavonoids (Table 2, Fig. 1). The leaves of studied plant have observed 1.456 mg/g and fruits have 1.256 mg/g as total flavanoids.

The phytochemical screening for the quantitative estimation of the total phenols of the plant *Dolichandrone falcata* showed that bark was rich in total phenolic compound. Presence of high amounts of total phenolic and total flavonoid content in the bark unveiled the medicinal importance of bark. Gordian *et al.*, (2007) reported the amount of total phenolic compounds found in eleven medicinal plants were studied in Nigeria. Estimation of total phenols was also observed in medicinal plants such as *Calotropis procera* and *Gymnema sylvestris* (Kumar *et al.*, 2007).

Table 1: Qualitative analyses for the presence of different Phytochemicals in *Dolichandrone falcata*

Plant Parts	Alkaloid (Mayer's reagent Wanger's reagent)	Tannin (Ferric chloride)	Saponin (Foam Test)	Terpanoids (Salkowski test)	Flavonoid (Ammonium solution)	Phenols (Ferric chloride sulphuric acid)
Bark	+	+	+	+	+	+
Leaves	+	+	-	-	+	+
Fruits	+	+	+	+	+	+

+ Present, -- Absent

Table 2: Quantitative analyses of Total Flavonoid and Total Phenol in different plant parts of *Dolichandrone falcate*.

Plant parts	Amount of phenolic product 1	Amount of flavonoid product 2
Bark	0.77+0.0721	2.106+ .436
Leaves	0.546 + .014	1.456+ .060
Fruits	0.316 + .0550	1.256 + .110

Results are expressed in mg\g

1. Malik and Singh,1980; Sadashivam and Manickam, 1996.

2. Swain and Hillis, 1959 ; Mahadevan and Sridhar, 1986

Micro Controller Double Beam UV-VIS Spectrophotometer System: UV5704M

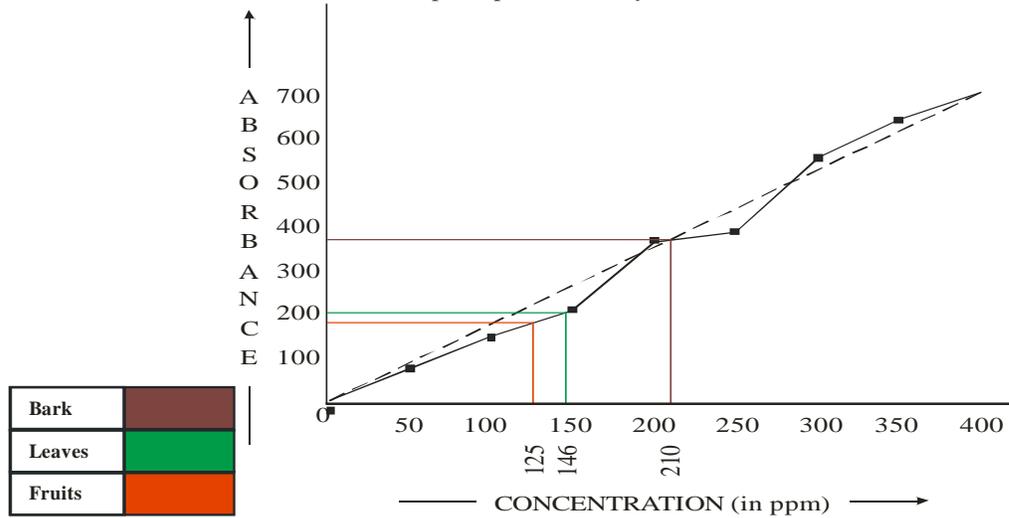


Fig. 1. Estimation of Total Flavonoid.

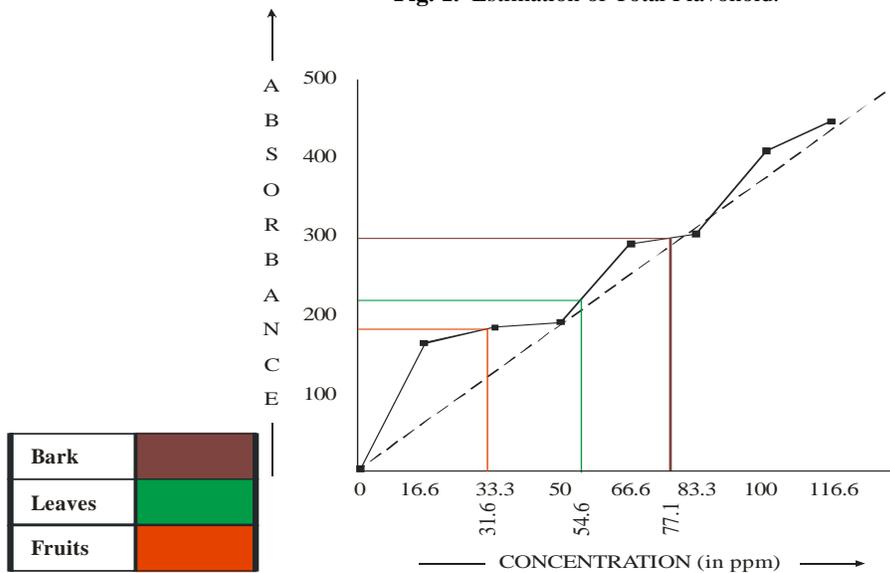
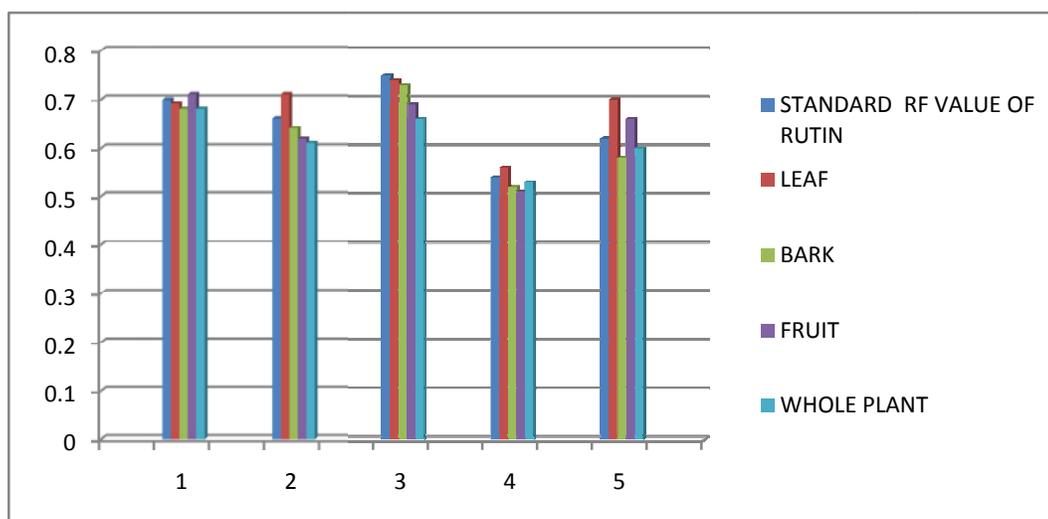


Fig. 2. Estimation of Total Phenols.

Table 3: Quantitative analyses of Rutin Flavonoid through Rf values in different plant parts of *Dolichandrone falcata*.

S.No	Solvent Systems	Standard Value of Rutin	Rf Leaf	Bark	Fruit	Whole Plant
1.	Toluene : Acetic acid 40 : 20	0.70	0.69	0.68	0.71	0.68
2.	Toluene : Ethanol 46 : 4	0.66	0.71	0.64	0.62	0.61
3.	Toluene : Acetone : Formic acid 30 : 10 :10	0.75	0.74	0.73	0.69	0.66
4.	N-butanol : Acetic acid : Water 4 : 1 : 5	0.54	0.56	0.52	0.51	0.53
5.	Hexane : Ethyl acetate : Acetic acid 31:14: 5	0.62	0.70	0.58	0.66	0.60



Different types of flavonoids were found in members of family Bignoniaceae. Joshi and Singh (2004) analysed quinones and other constituents from root of *Tecomella undulata*. The identification of chemical constituents of some medicinal plants have progressively aided in the development of rapid and accurate methods for screening plant for a particular chemical. The flavonoids quercetin is known for its ability to relieve hay fever, eczema, and asthma. Chrysin-7-rutinoside was analysed from the leaves of *Dolichandrone falcata* (Subramanian *et al.*, 1972).

Recent studies have also confirmed that flavonoids Chrysin and Rutin can be used against cancer and as an antifertility agent. Such fundamental research will help to find the specific phyto-chemical which is helpful in treatment of non-curable diseases like cancer and AIDS. Identification of specific flavonoids (Rutin) will be done by using the method TLC.

The presence of active phyto-chemical like rutin (flavonoid) is represented by their Rf values in different parts of plant (Table 3). Rf values were estimated by Thin layer Chromatography. Determination and comparison of Rf values revealed that (Toluene:Acetone:Formic acid) is the best solvent system among the five solvents were used. Rajalakshmi and Kalaiselvi (2010) were also extracted Quercetin from plant species *Euphorbia hirta* by this solvent system (Toluene: Acetone: Formic acid). The result indicated that (N-butanol: Acetic acid: Water) solvent system estimated the lowest level of Rf value.

Kumar *et al.*, (2007) extracted common types of flavonoids such are Rutin, Quercetin, Myricitine from different medicinal plants. The Rf values of plant parts are very much similar to the standard values of Rutin in all the solvent system, thus the findings supported the presence of rutin flavonoid in plant *Dolichandrone falcata*.

Such fundamental research will direct a new pathway for specific phyto-chemical which might be helpful in treatment of non-curable diseases like cancer and AIDS. During experimentation it was observed that percolated TLC plates give the perfect result. Rutin is reported in medicinal plant species *Dolichandrone falcata*. TLC is the best method of qualitative evaluation. Identification of specific flavonoids in medicinal plants can be used by different techniques. New methods of HPTLC can be used for further estimations. Further studies are needed for this plant species to isolate, identify, characterize and elucidate the structure of their bio-active compound.

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