Research Article

Pharmacognostic Standardization of roots, stems, leaves and fruits of *Fumaria parviflora* Lam. (Fumariaceae)

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https://doi.org/10.31024/ajpp.2018.4.2.23

Received: 19 February 2018 Revised: 22 March 2018 Accepted: 27 March 2018

Abstract

Objective: Present studies deal the quality control parameters of a locally occurring Indian medicinal plant, *Fumaria parviflora* Lam. which is used as folk medicine in Haryana (India). **Materials and methods:** In this study the organoleptic, morphological, microscopical characteristics, and physicochemical evaluation (ash value, extractive value, foreign matters and moisture content), fluorescence analysis of the root, stem, leaf and fruit of *Fumaria parviflora* were investigated. Ethanolic extracts were prepared by extracting the ground powders of root, stem, leaf and fruit. Preliminary phytochemicals analysis of ethanolic extracts of different parts of *Fumaria parviflora* was carried out for qualitative analysis. **Results and conclusion:** Preliminary phytochemicals analysis of ethanolic extracts of root, stem, leaf and fruit revealed the presence of carbohydrates, alkaloids, tannins and flavonoids. The pharmacognostic investigation of root, stem, leaf and fruit of *Fumaria parviflora* would be useful for maintaining the standards for the quality, purity and sample identification.

***Keywords*:** *Fumaria parviflora*, microscopy, pharmacognostic, flavonoids

Introduction

Fumaria parviflora Lam. (Fumariaceae) commonly known as "fine leaf fumitory, earth smoke", Indian fumitory, and wax doll in English (Rizvi et al., 2017). It is a loved medicinal herb in Indian system of medicine. It is well known annual weed growing throughout in India from Indus Ganga plain to down Nilgiris in South (Karuna modi et al., 2016). The major chemical constituents reported include alkaloids like Protopine, parfumine, d-bicuculline, hydrastine, N-methlhydrastine, N-methylhydrasteine, microcarpine, sanguinarine, adlumiceine, coptisine, fumaritine, sinactine, N-methylstylopine and sterols of plant are β-sitosterol, stigmasterol,and campesterol etc (Fafal et al., 2007; Paltinean et al., 2013). Entire herb has been widely used in Ayurvedic medicine system as bitter; cooling, expectorant, constipating, increases "vata" removes biliousness, fever, burning of the body, tired feeling, wandering of the mind, intoxication, urinary discharge, vomiting, thirst, enriches the blood, good in leprosy (API, 2004).

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The plant shows many biological properties such as hepatoprotective (Khan et al., 2017), antipruritic, antifeedant, Antiprotozoal, antiparasitic, anthelmintic, antidiabetic, antieczema, antioxidant, Antinociceptive, antimicrobial, prokinetic, laxative and spasmodic effect (Kumar et al., 2017).

As plant was not explored in depth but it have lot of ethanopharmacological importance. Therefore, we carried out the present studies which deal with extremely important information on micro morphological characteristics of this medicinal plant which would help in identification and authentification as well as provide basic pharmacognostic parameters. The pharmacognostic and phytochemicals analysis is key steps to develops the herbal pharmacopoeia standards and helpful in identification and quality control of the medicinal plants.

Materials and methods

Plant materials

The fruits bearing plants of *Fumaria parviflora* Lam. were collected from district Sirsa, Haryana (India) in month of February-March 2015. The collected plant was authenticated from Raw Material Herbarium and Museum,

Delhi (RHMD), CSIR-NISCAIR, New Delhi. A slide specimen/ sample have been deposited in the Raw Material Herbarium and Museum, Delhi (RHMD), CSIR-NISCAIR, Ref. No. NISCAIR/RHMD/Consult/2015/2811/04.

Chemicals

All chemicals were analytical grade used in this study purchased from SRL, CDH, SD fine and HIMEDIA. Formalin, absolute alcohol, safranin, haematoxyllin, fast green, glacial acetic acid, clove oil, canada balsam, chloral hydrate, phloroglucinol, H_2SO_4, NaOH, NH_3, lead acetate, $FeCl_3$, potassium hydroxide, ethyl acetate and chloroform etc.

Macroscopic and microscopic examination

Morphological characters were done by using simple microscope. The color, size, shape, odour and taste of roots, stems, leaves and fruits were determined. Microscopic characters were done by preparing thin section of root, stem, leaf and fruit of *Fumaria parviflora* Lam. (Tekin et al., 2017).

Powdered microscopy

Air dried different parts of plant (leaves, stems, fruits and roots) were finely powdered (# 60) and observed under microscope. Small quantity of different plant parts powder were placed separately on slides and each slide was placed 2-3 drops of chloral hydrate solution and each slide was covered with cover slip then observed under microscope. Different cell contents i.e. epidermis, cork, collenchyma, schlerenchyma, parenchyma, reticulate vessels and stomatal cells were observed and snapshot was done by using digital camera (Sonibare et al., 2014).

Physiochemical examination

Physicochemical parameters of different plant parts of powdered as well as extract of crude drug such as foreign matter, total ash, water soluble ash, acid insoluble ash, alcohol and water soluble extractive values were determined. The moisture content of different plant parts of powdered and extracts were determined by using loss on drying method (Dhingra et al., 2014).

Fluorescence examination

Fluorescence examination of the different parts of plant powder was done by using standard method. The examination was done by treating the plant powder with different solvents including both acidic/basic and organic/inorganic. After treatment they were examined under visible light, short ultra-violet light and long ultra- violet light.

Fluorescence examination is an imperative mechanism for the screening of those constituents which have the assets of showing different colors under UV light. Some components are not fluorescent themselves but when they are reacted with solvents are converted into fluorescent derivatives. This phenomenon may be due to an exacting fluorescent substance or fluorescent derivative formed after treatment with reagents. Still many natural drugs are assessed qualitatively by using this parameter. Powdered roots, stems, leaves and fruits materials were observed under visible light, short ultra-violet light and long ultra-violet light simultaneously after treatment with different organic and inorganic reagents like KOH, NaOH, H_2SO_4, HCl, $FeCl_3$, iodine solution and HNO_3 (Akbar et al., 2014).

Microchemistry examinations of roots, stems, leaves and fruits powder

Microchemistry evaluations of different parts of plant with different chemical reagents were observed. Screening showed the presence of different phytoconstituents along with colour changes under ordinary day light by standard procedure.

Preparation of extract and preliminary phytochemicals examination

The different parts of the plant were air dried in shadow followed by grinding. Extraction of each part was performed with ethanol as solvent on soxhlet apparatus. The each extract was evaporated in rotary evaporator apparatus and air dried at room temperature for 2-3 days. These extracts were stored in refrigerator for further study. The preliminary phytochemicals examination of different plant parts extract of *Fumaria parviflora* was done by using the standard procedure.

Results

Macroscopic examination of roots, stems, leaves and fruits

The organoleptic and morphological study of *Fumaria parviflora* leaves as well as powder showed green colour. Leaves are compound, pinnatifid, 5 to 7 cm long, apex acute; petiole is very thin, 2.5 to 4.0 cm long and bitter in taste. The root was branched, cylindrical, about 8-10 cm long, 3 mm thick, cream coloured, and bitter in taste. The stem was pentagonal, pale green, smooth, hollow, about 2-4 mm thick, bitter and slightly acrid in taste. Fruits Capsule, are single seeded, 2 mm long and obovate, subtruncate, obscurely, apiculate, rugose and bitter in taste (Figure 1).

Microscopic examination

Roots microscopy

Transverse section of root shows a single layered epidermis. The cortex (ct) consisting of thin walled, rectangular, parenchymatous cells, outer 1 or 2 layers irregular and brown in colour. Endodermis is not distinct, secondary phloem very narrow and central part shows a

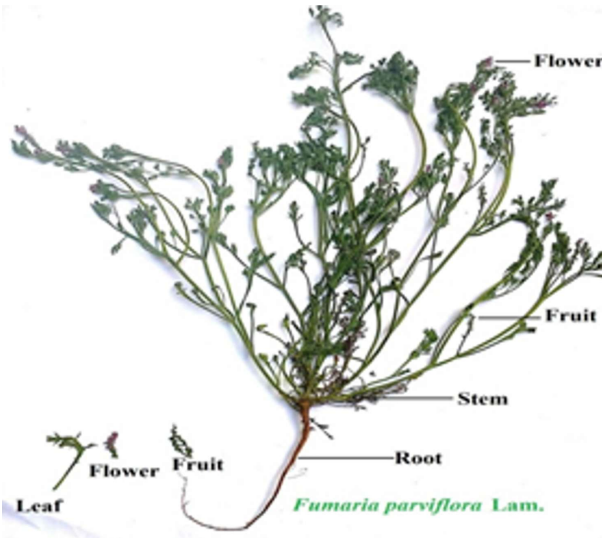

Figure 1. *Fumaria parviflora*

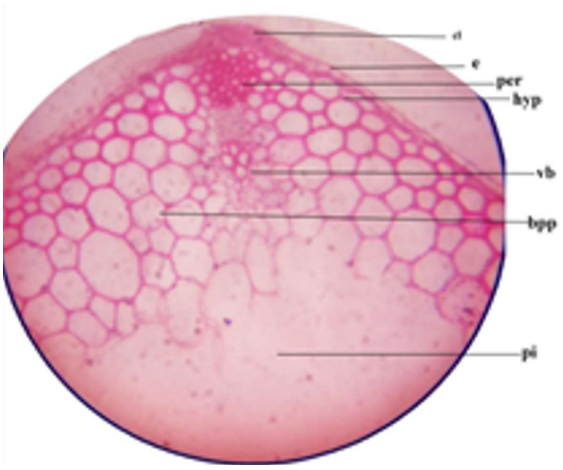

Figure 3. T.S. of *Fumaria parviflora* stems

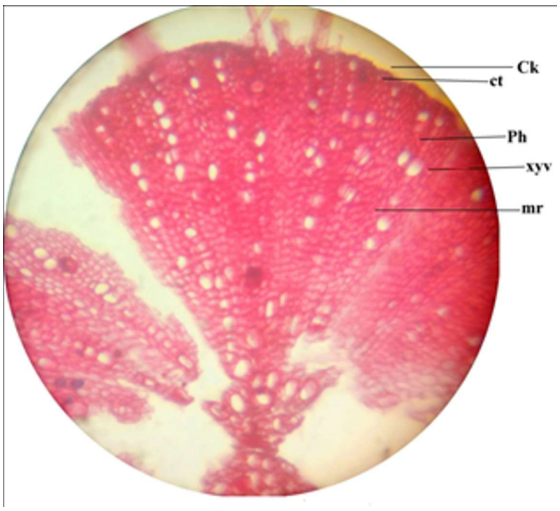

Figure 2. T.S. of *Fumaria parviflora* roots

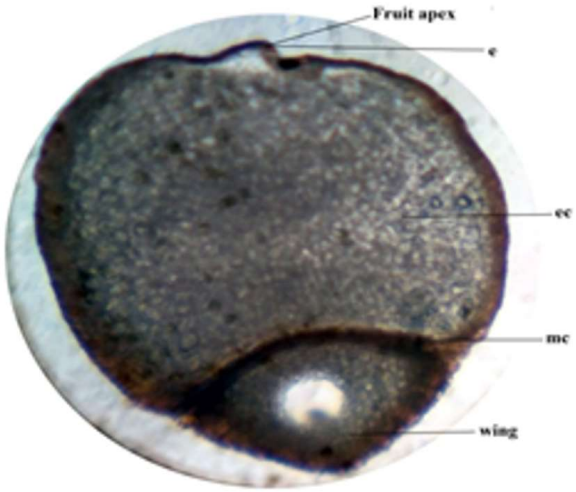

Figure 4. T.S. of *Fumaria parviflora* fruits

wide zone of xylem and consists of common elements. Xylem vessels (xyv) mostly single having reticulate and spiral thickening, medullary rays (mr) are less developed and mostly uniseriate (Figure 2).

Stems microscopy

Transverse section of Stem shows a single layered thin walled epidermis (e) of rectangular cells, covered with thin cuticle (cl), cortex (ct) narrow brown pitted parenchymatous (bpp). Pentagonal outline, of stem having well-known collenchymatous hypodermis (hyp); vascular bundles (vb) collateral, 5 or 6 arranged in a ring; each vascular bundle capped with pericyclic fibres (per). It has centrally hollow wide pith (pi) (Figure 3).

Fruits microscopy

Cross section of fruit wall we have observed 3 layers. Outer layer of fruit showed epidermis (e) with thin cuticle cells and some hypodermal cells. Mesocarp (mc) with schlerenchymatous cells. The endocarp (ec) one to many layers of endosperm cells developed under the apical region and folded on inner side of the fruit (Figure 4).

Powder microscopy

The powder characteristics of the root, stem, leaf and fruit were study under microscope and shown in the (Figure 5 a-d).

Physicochemical examination

Present study deals the various physicochemical parameters such as foreign matter, loss on drying, total ash, water soluble ash, acid insoluble ash values, and extractive values were determined in duplicate as depicted in table 1.

Fluorescence investigation

The fluorescence characteristics of the powder with different chemical reagents are depicted in table 2,3,4,5.

Microchemistry investigation of powdered roots, stems, leaves, and fruits

The different parts of the plant powder was treated with different chemical reagent showed the presence of

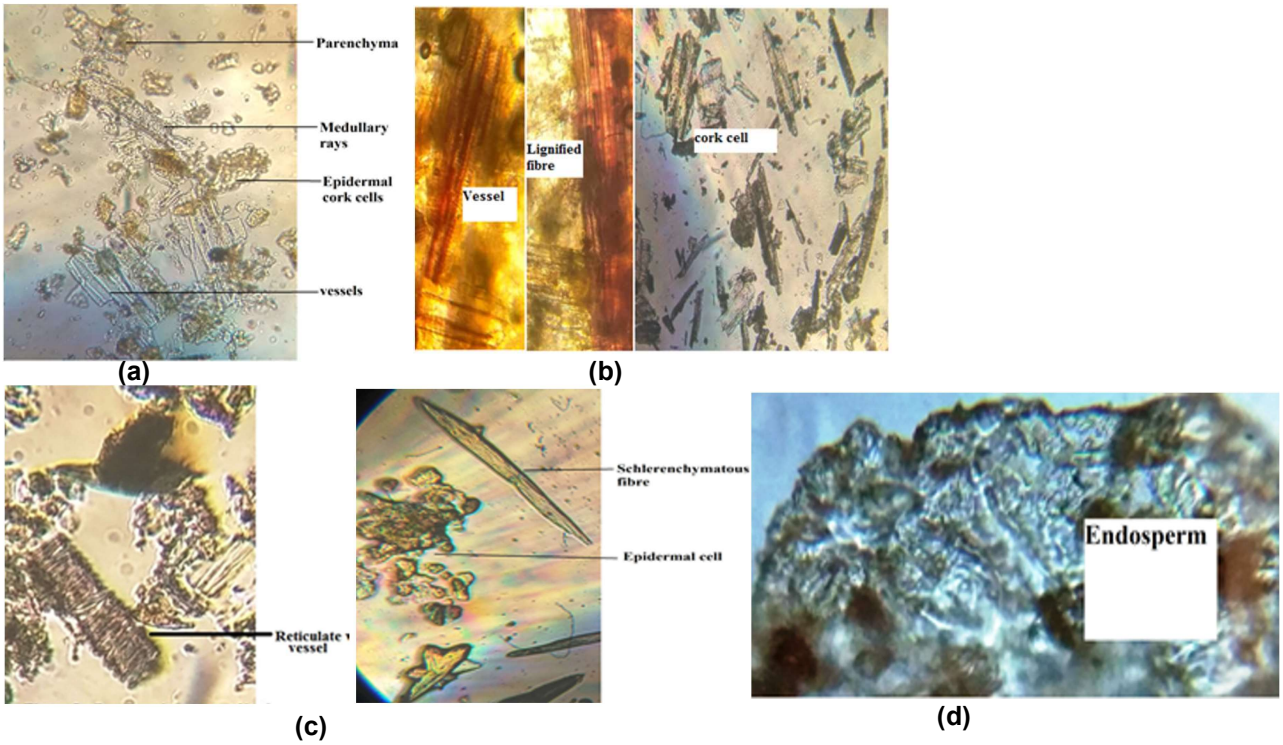

Figure 5 (a-d). Powder microscopy of *Fumaria parviflora*: (a) roots, (b) stems, (c) leaves, (d) fruits

Table 1. Physicochemical examination of roots, stems, leaves and fruits of *Fumaria parviflora* **Lam.**

S.N.	Parameters	Mean ± SD (%W/W)			
		Roots	Stems	Leaves	Fruits
Ash value					
1	Total ash	16.23±0.21	15.23±0.21	15.50±0.41	8.30±0.22
2	Water soluble ash	8.5±0.41	8.3±0.08	9.4±0.29	6.16±0.24
3	Acid insoluble ash	2.33±0.24	1.32±0.23	5.30±0.22	1.17±0.24
Extractive value					
1	Water soluble	12.28±0.21	26.19±0.05	29.73±0.25	18.03±0.21
2	Alcohol soluble	10.19±0.05	12.03±0.02	14.56±0.40	20.83±0.12
Moisture content					
1	Moisture content	0.59±0.01	0.45±0.02	0.82±0.02	0.16±0.01
Foreign matter					
1	Foreign matter	1.03±0.12	0.90±0.08	1.33±0.09	0.8±0.08

carbohydrate, lignin, tannin, alkaloid, saponin and flavonoids were shown in table 6.

Extractive values and preliminary phytochemicals examination

The extractive values of ethanolic extract of root, stem, leaf and fruit of *Fumaria* parviflora were calculated (table 7). Preliminary phytochemicals examination of ethanolic extracts of different parts of *Fumaria parviflora* revealed the presence of carbohydrates, proteins, steroids, alkaloids, glycosides, flavonoids, and tannins (table 8).

Discussion

As quality control of the allopathic medicine is essential, likewise the pharmacognostic standardization of herbal drug is also necessary for the quality control because substitute or adulterated plant drugs are mainly present in the market. This type of investigation will help to make sure the identity, quality, purity and safety of herbal drug for the medicinal use. The different parameters studied are organoleptic characters, macroscopic analysis, microscopic analysis and fluorescence analysis. Macroscopic and microscopic analysis is one of the easiest and cheapest methods to correctly identify the authentic herbal drug and the surety of raw material. Morphological and microscopical studies of root stem, leaf and fruit will be helpful in the identification of these parts of *Fumaria*

Table 2. Fluorescence examination of powdered roots

S.N.	Reagents	Colour observed in visible light	Colour observed under UV light	
			Short (254nm)	Long (365nm)
1	Powder	Light brown	Light brown	Light brown
2	NaOH (1N)	Brown	Yellowish green	Pale yellow
3	KOH (1N)	Brown	Green	Yellow
4	Conc. H_2SO_4	Pale yellow	Yellow	Yellow
5	Conc. HNO_3	Reddish brown	Green	Green
6	Conc. HCl	Brown	Yellow	Yellow
7	50% H_2SO_4	Reddish brown	Yellow	Yellow
8	Iodine solution	Dull Yellow	Black	Green
9	5% $FeCl_3$	Brownish black	Black	Black
10	Picric acid	Dull yellow	Green	Brown

Table 3. Fluorescence examination of powdered stems

S.N.	Reagents	Colour observed in visible light	Colour observed under UV light	
			Short (254nm)	Long (365nm)
1	Powder	Yellowish green	Green	Light green
2	NaOH (1N)	Sand	Lemon	Creamy
3	KOH (1N)	Sand	Lemon	Yellow
4	Conc. H_2SO_4	Reddish brown	Green	Yellow
5	Conc. HNO_3	Reddish	Yellow	Pale yellow
6	Conc. HCl	Pale yellow	Creamy	Creamy
7	50% H_2SO_4	Reddish	Lemon	Yellow
8	Iodine solution	Brown	Black	Black
9	5% $FeCl_3$	Black	Black	Black
10	Picric acid	Yellow	Walnut brown	Walnut brown

Table 4. Fluorescence examination of powdered leaves

S.N.	Reagents	Colour observed in visible light	Colour observed under UV light	
			Short (254nm)	Long (365nm)
1	Powder	Green	Green	Dull green
2	NaOH (1N)	Pale yellow	Light green	Light green
3	KOH (1N)	Pale yellow	Light green	Light green
4	Conc. H_2SO_4	Yellowish green	Green	Green
5	Conc. HNO_3	Reddish brown	Green	Green
6	Conc. HCl	Light brown	Purple	Purple
7	50% H_2SO_4	Yellow	Green	Green
8	Iodine solution	Blood red	Blood red	Blood red
9	5% $FeCl_3$	Bluish black	Black	Black
10	Picric acid	Yellowish green	Green	Green

Table 5. Fluorescence examination of powdered fruits

S.N.	Reagents	Colour observed in visible light	Colour observed under UV light	
			Short (254nm)	Long (365nm)
1	Powder	Dark brown	Black	Black
2	NaOH (1N)	Pale yellow	Parrot colour	Green
3	KOH (1N)	Reddish brown	Light green	Light green
4	Conc. H_2SO_4	Reddish	Purple	Purple
5	Conc. HNO_3	Light brown	Green	Green
6	Conc. HCl	Dark brown	Green	Green
7	50% H_2SO_4	Red	Reddish brown	Reddish brown
8	Iodine solution	Red	Parrot colour	Green
9	5% $FeCl_3$	Black	Dark Green	Green
10	Picric acid	Pale yellow	Green	Green

Table 6. Microchemistry investigation of powdered roots, stems, leaves and fruits

S.N.	Reagents	Roots		Stems		Leaves		Fruits	
		Colour/ppt	Constituents	Colour/ppt	Constituents	Colour/ppt	Constituents	Colour/ppt	Constituents
1	Iodine solution	Pale yellow	Cellulose (+)	Pale yellow colour	Cellulose (+)	Pale yellow	Cellulose (+)	Pale yellow	Cellulose (-)
2	Iodine solution+66% H_2SO_4	Bright blue	Cellulose (+)	Bright blue colour	Cellulose (+)	Bright blue	Cellulose (+)	Bright blue	Cellulose (-)
3	Phloroglucinol + HCl	Red	Lignin (+)	Red colour	Lignin (+)	Red	Lignin (+)	Red	Lignin (+)
4	Water	No change	Saponin(-)	No change	Saponin(-)	No change	Saponin(-)	No change	Saponin(-)
5	Molisch'S reagent	Violet colour	Carbohydrate(+)	Violet colour	Carbohydrate(+)	Violet colour	Carbohydrate(+)	Violet colour	Carbohydrate(+)
6	Aqueous $FeCl_3$	Black	Tannin(+)	Black	Tannin(+)	Black	Tannin(+)	Black colour	Tannin(+)
7	Mg-HCl	No change	Flavonoid(-)	Orange colour	Flavonoid(+)	Red to purple colour	Flavonoid(+)	Red colour	Flavonoid(+)
8	Picric acid	Yellow ppt	Alkaloid(+)	Yellow ppt	Alkaloid(+)	Yellow ppt	Alkaloid(+)	Yellow ppt	Alkaloid(+)

parviflora Lam. Physicochemical analysis of root, stem, leaf and fruit are helpful to establish quality standards of the plant. Therefore, various parameters used for identification of different plant parts are important for drug evaluation. The results of all types of analysis are helpful in establishing quality control standards and purity assurance of drugs. Phytochemicals analysis is also the important part of herbal drug quality parameters. These simple, economical but consistent principles can be useful even for an inexpert person whenever using the drug as folk medicine. This investigation will also be helpful for producer for assessing the quality and purity of raw material. Concisely, the different parameters were described here can be considered as distinguishing to identify and authenticate this herbal drug.

Table 7. Extractive values of different parts of *Fumaria parviflora* extract

Extracts	Yield (%W/W)	Colour of extract	Consistency
Root ethanolic extract	6.25	Light brown	Solid
Stem ethanolic extract	9.75	Yellowish green	Viscous
Leaf ethanolic extract	17	Blackish brown	Gummy
Fruit ethanolic extract	14.65	Dark brown	Viscous

Table 8. Preliminary phytochemicals examination of ethanolic extracts of different parts of *Fumaria parviflora*

Phytochemical constituents	Chemical tests	Roots ethanolic extract	Stems ethanolic extract	Leaves ethanolic extract	Fruits ethanolic extract
Alkaloids	Dragendorff's reagent	+	+	+	+
	Mayer's reagent	+	+	+	+
	Wagner's reagent	+	+	+	+
	Hager's reagent	+	+	+	+
Cabohydrates	Molisch's reagent	+	+	+	+
Proteins	Biuret reagent	-	-	-	-
Lipids	Paper Staining test	-	-	-	+
Saponins	Foam test	-	-	-	-
Glycosides	Borntrager's test	-	-	-	-
	Keller- Killiani test	-	-	-	-
Tannins	Ferric chloride test	+	+	+	+
	Lead acetate test	+	+	+	+
Flavonoids	Shinoda test	+	+	+	+
	66% H_2SO_4	+	+	+	+
Steroids	Salkowski test	+	-	-	+
	Conc. H_2SO_4	+	-	-	+

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