



## Phytochemical Screening of Some Medicinal Plants of Chitrakoot Region

### KEYWORDS

phytochemicals, extraction methods, secondary metabolites.

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### ABSTRACT

The present study deals with the phytochemical screening of available parts (mainly leaves and stem) of twelve medicinal plants of four different families found in Chitrakoot region. Test plants were extracted with methanol, petroleum ether and water by cold and hot extraction methods and screened for the presence of carbohydrates, alkaloids, flavonoids, proteins, rasin, anthocynin, saponin, steroids, starch, tannins, starch, glycosides, phenol, phlobatannins and terpenoids. We found that the selected plants are good source of various phytochemicals. This study revealed the presence of various biologically active secondary metabolites which could be helpful in the prevention of chronic diseases.

### INTRODUCTION

Plants produce various bioactive phytochemicals which can be grouped under two categories; primary and secondary metabolites. Primary metabolites include proteins, carbohydrates, amino acids and chlorophyll while polyphenols, alkaloids, terpenoids are some examples of secondary metabolites<sup>1</sup>. Secondary metabolites are the chemicals that are not required for the immediate survival of the plant but synthesized to increase the survival of the plant by allowing it to interact with pathogens, herbivores insects and environment<sup>2</sup>. Secondary metabolites such as alkaloids, glycosides flavonoids, steroids, saponins and terpenoids play an important role in the protection of the plant from environmental stress, attacks of pathogens and insect pests<sup>3</sup>. Due to presence of these bioactive phytochemicals, plants provide a source of medicine since historic times and now these are an important part of all the world's pharmaceuticals and serve as starting material for drug development<sup>4</sup>. Phytochemicals are reported to have various health promoting effects. Studies also suggest that these natural phytochemicals modulate various molecular signal transduction pathways, involved in the phenomenon of inflammation, thereby preventing the onset of various chronic diseases like cancer, atherosclerosis, neurodegradation, obesity, articular rheumatism, skin aging and diabetes<sup>5</sup>. Most phytochemicals function as antioxidants in vitro and they can reduce oxidative stress and inflammation which are involved in the progression of type II diabetes mellitus<sup>6</sup>. These specific organic compounds of plant origin have shown antidiabetic activity and represent a source for the discovery and development of new type of antidiabetic molecules<sup>7</sup>. Polyphenolic compounds, immensely distributed throughout the plant kingdom, serve as antioxidants and neutralize deleterious free radicals, quenching singlet or triplet oxygen, or decomposing peroxides, and regulate carbohydrate metabolism<sup>8</sup>. Besides combating various afflictions, and possessing antidiabetic and antioxidative properties, phytochemicals could provide health benefits as, substrate for biological reactions, Co-factors for enzymetic reactions, inhibitors for enzymetic reactions, absorbants/sequestrants that bind to and eliminate undesirable constituents in the intestine, ligands that agonize or antagonize cell surface/ intracellular receptors, compounds that enhance the absorption or stability of essential nutrients, selective growth factors for beneficial gas-

trointestinal bacteria and selective inhibitors of deleterious intestinal bacteria<sup>9</sup>.

Chitrakoot situated in the northern region of Satna district of Madhya Pradesh, has a very rich wealth of medicinal plants which has also been described in our epics like Ramayana. The place is well known for its beautiful hill ranges, historical caves, perennial streams and diverse fauna and flora. Vast variety of herbs, shrubs, trees, climbers, having different flowers, fruits, roots are available<sup>10</sup>. Present work is aimed to screen different phytoconstituents found in some medicinal plants of Chitrakoot.

### MATERIAL AND METHOD:

Twelve plants of family Poaceae, Brassicaceae, Fabaceae and Apocynaceae were collected from different regions of Chitrakoot, and washed with 70% methanol. These were shade dried in room temperature and grinded using a grinder. Powdered sample was kept in air tight container.

Preparation of plant extracts- Dried plant materials were extracted with water, methanol (hot extraction) and petroleum ether (cold extraction). Methods of extract preparation were adapted from Pandey S<sup>11</sup>. (2014), with some modifications.

Preparation of water and methanol extract: 10 gm of dried sample was taken for maceration and dissolved in 50ml of water (to prepare water extract), and 50ml (90%) of methanol (for methanol extract) and kept in rotary shaker for 1 h. Then it was filtered through whatman No. 1 filter paper and the filtrate was used for the screening.

Preparation of petroleum ether extract: 10 gm of dried powdered was taken and soaked in 50 ml petroleum ether and kept in refrigerator for 1 hour. After 1h, it was filtered through Whatman No. 1 filter paper and the filtrate was used for analysis.

All the obtained extracts were then subjected to different qualitative tests to find out the presence of specific phytochemicals.

**(i) Test for Carbohydrates:** Molisch test: 1 ml of sample is placed in a test tube and two drops of Molisch reagent

was added. 2ml solution of concentrated  $H_2SO_4$  was added in test tube. Formation of red violet ring in the interface gave the positive Molisch test.

**Fehling test:** 2ml solution of Fehling A and Fehling B were taken in a test tube then dropwise sample were added. The mixture was shaken well and kept in a water bath for 10-15 minutes at 100 °C. A rusty brown or brick red colour precipitate confirms the presence of carbohydrates in the sample.

**Benedict test:** 2ml of Benedict reagent was added to the 1 ml of plant extract. Then the mixture was shaken well and placed in a water bath for 10-15 minutes. Formation of reddish precipitate indicates the presence of sugars in the sample.

**Anthron test:** 2 ml of anthron reagent was added to 500  $\mu$ l of extract. Formation of green blue colour gives a positive anthron test.

**(ii) Test for Alkaloids:** Mayer's test: 1ml of sample was added to a few drops of Mayer's reagent. Formation of white or pale yellow precipitate indicates the presence of alkaloids in the sample.

**Wagner's test:** 1.5% of HCl was added in 1 ml of extract and a few drops of Wagner's reagent were added to it. Appearance of yellow/ brown precipitate indicates the presence of alkaloids.

**Hager's test:** 1ml of extract was taken in a test tube, and few drops of Hagers reagent was added to it. Yellow precipitate confirms the presence of alkaloids in the sample.

**Dragondrof test:** 5ml of distilled water was added to the the 2 ml of sample, then 2M HCl and 1 ml of Dragondrof's reagent was added. Orange / orange red precipitate indicates the presence of alkaloids.

**(iii) Test for Flavonoids:**  $H_2SO_4$  test: A fraction of the extract was taken and treated with concentrated  $H_2SO_4$  and observed for the formation of orange colour.

**Shinoda test (HCl test):** Few fragments of Mg and dropwise HCl were added to 1 ml plant extract, which gives pink reddish/ brownish pink or green or blue colour in few minutes.

**(iv) Test for Proteins:** Biuret test: 1% of NaOH was added to 1 ml of extract and few drops of 1%  $CuSO_4$  were then added. Blue/ purple or violet/ pinkish colour indicates the presence of proteins.

**Millon's test:** 1 ml of test extract was mixed with  $H_2SO_4$  then Millon's reagent was added dropwise. White/yellow precipitate appears which turns into red colour precipitate, after heating the mixture. This indicates the presence of proteins.

**Ninhydrin's test:** 2 drops of freshly prepared Ninhydrin reagent (0.1% in n- butanol) is added to 1ml of extract and heat and observed for blue or red orange colour.

**(v) Test for Resins:** 1ml of ethanolic extract was dissolved in acetone and then 1 ml of distilled water is added. Turbidity indicates the presence of resin.

**(vi) Test for Tannins:** To 1 ml of the extract, 2ml of 5%  $FeCl_3$  is added which gives dark blue or greenish black colour and a positive tannin test.

**(vii) Test for Steroids:** Salvoski test: 1 ml of test sample was dissolved in 1 ml of chloroform and equal amount of concentrated  $H_2SO_4$ . Formation of Bluish red to cherry colour in chloroform layer shows the presence of steroids.

**(viii) Test of Saponin:** Foam test: A small amount of extract was shaken with water and observed for the presence of foam.

**Sodium Bicarbonate test:** Few drops of Sodium bicarbonate was added to 1 ml of plant extract. If honeycomb like structure forms, it confirms saponin.

**(ix) Test for Anthocyanin and Betacynin:** 1 ml of plant extract was treated with 1 ml of 2N NaOH then heated. Formation of bluish –green colour indicated the presence of Anthocynin while yellow colour indicated the presence of betacynin.

**(x) Test for Starch:** 1 ml of iodine solution is mixed in 1ml of extract, formation of blue colour indicated the presence of starch in the extract.

**(xi) Test for Glycosides:** To 1 ml of plant extract, 1 ml  $FeCl_3$  (5%), and equal amount of acetic acid is added, then few drops of  $H_2SO_4$  is added to the mixture. Greenish blue colour indicates the presence of glycosides.

**(xii) Test for phenols:** 1ml of plant extract, when treated with few drops of  $FeCl_3$  solution; it gives blue green colour and confirms the presence of phenols.

**(xiii) Test for Phlobatannins:** 1ml of plant extract was treated with 1 ml of 1% HCl and heat. Red colour precipitate indicates the presence of Phlobatannins in the sample.

**(xiv) Test for Terpenoids:** To 1ml of plant extract, 2ml of chloroform and 3ml of conc.  $H_2SO_4$  was added. Areddish brown precipitate at thr interface, confirmed the presence of terpenoids.

## RESULT AND DISCUSSION:

Table-1 to table- 4 present the results of phytochemical screening in all the plants of selected four families. Screening results indicated that all the plants are rich in diverse advantageous phytoconstituents, like phenols, flavonoids, alkaloids, anthocyanins rasins, saponin, steroids, tannins, starch, glycosides, terpenoids as well as proteins and carbohydrates. Except phlobatannins, all these bioactive compounds were found to be present in almost all the studied samples. Slightest amount of phlobatannins was shown by *C. thevatia* leaf extracts in our preliminary screening studies.

Polyphenols are the most prolific antioxidant in the diet. Their total dietary intake could be as high as 1g/d, higher than any other phytoconstituent and known antioxidants. Chief sources of polyphenols are plant based natural products; fruits, fruit juice, tea, coffee, red wine, vegetables, cereals, chocolates and dry legumes. Studies support that polyphenols prevent the cardiovascular diseases, cancer, osteoporosis, neurodegenerative diseases and diabetes. Various animal based experiments had supported the protective effects of polyphenols in neurodegenerative disorders and brain function deterioration. Antioxidant properties of polyphenols have been widely studied but their mechanism of action is far more than prevention of oxidative stress. As antioxidant they improve cell survival but their mechanism of action is still uncertain<sup>12</sup>.

Flavonoids consist of a large group of polyphenolic compounds having benzo- $\gamma$ -pyrone structure widely spread in plants. These are known to synthesize by plant in response to microbial infection and present in all plant parts. They are divided into a variety of classes such as flavons (flavon, epigenin and leutinol) Flavonoles (quercetin, kempferol, myricetin), flavonones (flavonone, hesperetin, naringenin) among which, flavonols are the most abundant flavonoids in food. These are the major colouring compounds of flowering plants. Flavonoids in food are responsible for colour, taste prevention of fat oxidation. Being phytochemicals, these cannot be synthesized by humans and animals.

Several studies suggested protective effects of flavonoids against bacterial, viral, cardiovascular diseases, cancer and other age related diseases. These act as secondary antioxidant defence system in plant tissues exposed to different biotic and abiotic stress. Their functional hydroxyl group is responsible for scavenging free radicals and chelating metal ions. Mechanism of antioxidant action can induce-

- 1- Suppression of ROS formation (either by enzyme inhibition or metal chelation)
- 2- Scavenging ROS
- 3- Upregulation or protection of ROS.

Some of the important enzymes involved in ROS generation are inhibited by flavonoids are monooxygenase, glutathione S transferase, mitochondrial succinoxidase, NADH oxidase etc. They protect lipid against oxidative damage. In addition there hepatoprotective, anti-inflammatory, anti cancer, anti bacterial, and anti viral activities have been also reported<sup>13</sup>. In the present investigation, we found that all the samples exhibited the presence of polyphenolic contents and flavonoids.

The alkaloids are one of the most diverse groups of secondary metabolites, found in living organisms and have an array of structure type, biosynthesis pathway and pharmacological activities. These are being used as drugs in potions, medicines, teas, poultices and poisons for 4000 years. These are pharmacologically active substances which possess various physiological activities in humans and animals.

Several alkaloids are still in use such as caffeine (Psychostimulant), codeine (Anti tussive agent that suppresses the coughing reflex), cocaine (local anaesthetic), morphine (analgesic) and quinine (antipyretic)<sup>14</sup>. All the samples were found to possess moderate or high amount of alkaloids, except methanolic extracts of *P. glaucum* stem, *B.oleracea*, leaves and *S. asoca* leaves, in our investigation.

Terpenoids are also a class of secondary metabolites, widely distributed in plants. More than 40000 individual terpenoids are known to exist in nature with new compounds being discovered every year. Various classes of terpenoids have shown cytotoxicity against variety of tumor cells and anticancer properties in clinical animal models, These compounds have been reported in reduction of oxidative stress, suppression of inflammation, induction of apoptosis, regulation of cell cycle, inhibition of cell proliferation and modulation of multiple signal transduction pathways<sup>15</sup>. Our findings indicated that terpenoids were present in all the extracts except petroleum ether extract of *P. glaucum* stem, *A. nilotica* leaf and stem, *D. Sissoo* stem, *C. thevetia* stem, *S. asoca* stem and *C. roseus* stem methanolic extracts of *B.campestris* stem, *A. nilotica* leaves, *D. sissoo* leaves, *S. asoca* leaves & stem, *C. thevetia* stem. *C. thevetia* stem also did not show the presence of terpenoids.

Plant steroids are a distinctive class of phytoconstituents found throughout the animal and plant kingdom, a specific class of steroids, glucocorticoids are widely used for the suppression of inflammation in chronic inflammatory diseases which are associated with increased expression of inflammation genes by binding to glucocorticoid receptors on multiple signalling pathways. However, some adverse effects are also associated with their prolong use such as immunosuppression, hypertension, osteoporosis and metabolic disturbance<sup>16</sup>. We found that steroids were absent in petroleum ether extracts of *A. nilotica* leaves and stem, *S. asoca* stem, *C. thevetia* stem, *C. roseus* stem, water extract of *A. nilotica* leaves, *P. glaucum* grains and methanol extracts of *A. nilotica* leaves and *D. sissoo* stem.

Saponins are high molecular weight compounds in which a sugar molecule is combined with triterpene or steroid aglycon, so there are two major groups of saponins; triterpene saponins and steroid saponins. These are therapeutically important as they show hypolipidemic and anticancer activity of cardiac glycosides<sup>17</sup>. Result of our preliminary screening revealed that petroleum ether extract of many plants did not show saponins. Petroleum ether extracts of *C. dactylon*, *S. vulgare* leaves, grain and stem, *P. glaucum* leaves, stem and grain, *B. campestris* leaves and stem, *R. sativas* roots and leaves, *B. oleracea*, *A. nilotica* leaves and stem, *D. sissoo* leaves and stem, *S. asoca* leaves & stem, *C. thevetia* leaves and stem, *T. diversicata* stem, methanol extract of *S. vulgare* grain, *P. glaucum* stem, *T. diversicata* stem and water extract of *S. vulgare* grain and stem, and *P. glaucum* stem were found to be devoid of saponins.

Tannins are phenolic compounds of high molecular weight soluble in water and alcohol and found in root, bark, stem and other layers of plant tissues, due to presence of phenolic groups these are used as antiseptic. In ayurvedic medicine system, tannin rich plant based formulations are used to treat leucorrhoea, rhinorrhoea and diarrhea<sup>18</sup>. Except methanolic extract of *A. nilotica* leaves water and petroleum ether extract of *S. asoca*, stem, tannins were shown to be present in all the samples.

Anthocyanins are the members of the flavonoid group and are the most recognized visible members of bioflavonoid phytochemicals. Their free radical scavenging activity, antioxidant activity are well known but studies suggest that these phytochemicals possess other mechanisms of action which are responsible for other effects, beneficial for human health. Anthocyanin rich bioflavonoid mixtures and antocyanin isolates may provide protection from DNA cleavage, estrogenic activity, enzyme inhibition, increased production of cytokines, anti-inflammatory activity lipid peroxidation, decreasing capillary permeability and fragility<sup>19</sup>. Our result suggest that anthocyanins were present in all the samples, except petroleum ether extracts of *S. vulgare* grain and stem, *P. glaucum* grain and stem, *B. campestris* stem, *B. oleracea* leaves, *A. nilotica* stem and methanol & water extracts of *A. nilotica* stem.

Cardiac glycosides are the compounds used to treat congestive heart failure and cardiac arrhythmia. These compounds work by inhibiting the Na<sup>+</sup>/K<sup>+</sup> pump<sup>20</sup>. In our preliminary screening we found that glycosides were present in all the samples except methanolic and water extracts of *A. nilotica* stem, methanolic and petroleum ether extract of *C. thevetia* leaves, water and methanolic extract of *C. roseus* leaves and water and methanol extracts of *T. diversicata* leaves, phenolic compounds and flavonoid were present in all the samples. Similarly all the samples exhibited the pres-

ence of carbohydrates and proteins.

Resins are good traditional medicinal sources reported for the treatment of anti- inflammatory, antimicrobial, arthritis, wound healing, antitumor and antihyperlipidemia<sup>21</sup>.

Resins were absent in petroleum ether extracts of *C. dactylon*, *S. vulgare* leaves stem, *P. glaucum* grains and stem, *B. oleracea*, *S. asoca* leaves, *C. roseus* leaves and stem, and methanolic extract of *B. oleracea* and *A. nilotica* leaves.

Starch were absent an petroleum ether extract of *A. nilotica* leaves and stem, *D. sissoo* leaves and stem, *S. asoca* leaves and stem, *C. thevetia* leaves, *C. roseus* leavs, *T. diversicata* leaves & stem. methanolic extract of *S. vulgare* leaves and

water and methanolic extracts of *C. thevetia* stem and *C. roseus* stem.

Phlobatanins were absent in all the studied samples, except *C. thevetia* leaves.

**CONCLUSIONS**

Based on the results in this research, it can be concluded that all the tested plants contain high levels of different phytoconstituents, except phlobatannins which was present in only one sample, petroleum ether extract of *C. thevetia* leaves in low quality. Further work is needed to explore more information about these natural, non toxic and valuable compounds so that they can be used in drug formulation.

**Table -1: Phtochemical Screening of Poaceae family plants**

S. No.	Phytochemical	<i>C. dactylon</i>			<i>S. vulgare</i> leaves			<i>S. vulgare</i> grain			<i>S. vulgare</i> stem			<i>P. glaucum</i> leaves			<i>P. glaucum</i> grain			<i>P. glaucum</i> stem			
		W	M	PE	W	M	PE	W	M	PE	W	M	PE	W	M	PE	W	M	PE	W	M	PE	
1.	Carbohydrate	++	++	++	+	+	+	++	++	+	++	++	+	++	+	++	++	++	+	++	++	+	
2.	Alkaloid	++	++	+	+	+	+	++	++	+	++	+	+	++	+	+	+	+	+	+	-	+	
3.	Flavonoid	++	++	+	++	++	++	+	+	++	++	++	+	+	+	+	+	+	+	++	++	+	
4.	Protein	+	+	+	+	+	+	++	++	++	++	++	+	++	++	+	++	++	++	++	++	++	++
5.	Resin	++	+	-	++	+	-	++	++	+	+	+	-	++	++	+	+	+	-	+	+	-	
6.	Anthocyanin	+	+	+	++	+	+	+	+	-	+	+	-	+	+	+	+	+	-	+	+	-	
7.	Saponin	++	++	-	++	++	-	-	-	-	-	+	-	+	+	-	+	+	-	-	-	-	
8.	Steroid	+	+	+	+	+	+	-	+	+	+	+	+	++	++	++	+	+	+	+	+	+	
9.	Tannin	++	++	+	+	++	+	+	+	+	+	+	+	++	++	+	+	+	+	+	+	+	
10.	Starch	+	+	-	+	-	+	+	+	+	++	++	+	++	++	++	++	++	++	++	++	++	++
11.	Glycoside	++	+	+	++	++	++	+	+	+	+	+	+	++	++	+	+	++	++	+	+	+	
12.	Phenol	++	++	++	++	++	++	+	++	+	+	+	+	+	+	+	+	+	+	+	+	+	
13.	Phlobatanin	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
14.	Terpenoid	++	++	++	+	++	++	++	++	-	++	++	-	+	+	+	+	+	+	++	++	+	

W = Water extract, PE = Petroleum ether extract, M = Methanolic extract

**Table -2: Phtochemical Screening of Brassicaceae family plants**

S. No.	Phytochemical	<i>B.campestris</i> leaves			<i>B. campestris</i> stem			<i>R. sativus</i> root			<i>R. sativus</i> leaves			<i>B. oleracea</i>		
		W	M	PE	W	M	PE	W	M	PE	W	M	PE	W	M	PE
1.	Carbohydrate	+	++	+	+	+	+	++	++	++	+	+	+	+	+	+
2.	Alkaloid	+	+	++	+	+	+	+	+	++	+	+	++	+	-	+
3.	Flavonoid	+	++	+	++	+	++	++	++	++	++	+	++	+	++	++
4.	Protein	+	+	+	+	+	+	++	++	+	+	+	+	+	+	+
5.	Resin	+	+	+	+	+	+	+	+	+	+	+	+	+	-	-
6.	Anthocyanin	++	+	+	+	+	-	++	+	++	+	+	++	++	++	-
7.	Saponin	++	+	-	++	+	-	++	++	-	++	++	-	++	+	-
8.	Steroid	+	++	++	+	+	++	+	+	++	+	++	++	+	+	+
9.	Tannin	++	++	++	++	++	+	++	+	+	++	++	+	+	+	+
10.	Starch	+	+	+	+	++	++	+	+	++	+	+	++	+	+	+
11.	Glycoside	+	+	+	+	+	++	+	+	++	+	+	++	+	+	+
12.	Phenol	+	+	+	++	+	+	+	+	++	+	+	++	+	++	+
13.	Phlobatanin	--	--	--	-	-	-	-	-	-	-	-	-	-	-	-
14.	Terpenoid	++	++	+	-	-	+	-	+	++	-	+	++	+	+	++

W = Water extract, PE = Petroleum ether extract, M = Methanolic extract

Table -3: Phtochemical Screening of Fabaceae family plants

S. No.	Phytochemical	A. nilotica leaves			A. nilotica stem			D. sissoo leaves			D. sissoo stem			S. asoca leaves			S. asoca stem		
		W	M	PE	W	M	PE	W	M	PE	W	M	PE	W	M	PE	W	M	PE
1.	Carbohydrate	+	+	+	+	+	++	+	++	+	+	+	+	+	++	+	+	++	
2.	Alkaloid	+	+	+	+	+	+	++	++	++	++	+	+	++	-	++	++	+	++
3.	Flavonoid	+	+	+	+	+	+	++	+	++	++	+	+	+	+	+	+	+	
4.	Protein	+	++	+	++	+	+	+	++	+	++	+	+	++	++	+	+	+	
5.	Resin	+	+	+	+	-	+	+	+	+	+	+	+	+	+	-	+	+	
6.	Anthocyanin	+	-	+	-	+	-	+	+	+	++	++	+	+	+	+	+	+	
7.	Saponin	++	+	-	+	+	-	+	+	-	+	+	-	++	++	-	+	-	
8.	Steroid	-	+	-	+	-	-	++	+	++	++	-	++	+	+	++	-	+	
9.	Tannin	+	++	+	++	-	+	+	+	+	+	+	+	++	+	++	-	+	
10.	Starch	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
11.	Glycoside	+	-	+	-	+	+	+	+	+	+	+	+	+	+	++	+	+	
12.	Phenol	+	++	+	++	+	+	+	+	+	++	+	++	++	++	+	+	+	
13.	Phlobatanin	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
14.	Terpenoid	-	+	-	+	-	-	++	-	++	++	++	-	+	-	+	-	-	

W = Water extract, PE = Petroleum ether extract, M = Methanolic extract

Table -4: Phtochemical Screening of Apocynaceae family plants

S. No.	Phytochemical	C. thevetia leaves			C. thevetia stem			C. roseus leaves			C. roseus stem			T. divericata leaves			T. divericata stem		
		W	M	PE	W	M	PE	W	M	PE	W	M	PE	W	M	PE	W	M	PE
1.	Carbohydrate	+	+	+	+	+	+	++	+	++	+	+	+	++	++	+	+	+	
2.	Alkaloid	+	+	+	+	+	++	++	++	++	++	+	++	++	++	+	+	++	
3.	Flavonoid	+	+	+	+	+	+	++	++	+	+	+	++	++	+	+	+	+	
4.	Protein	+	+	++	++	++	+	++	++	++	+	+	+	++	++	+	+	++	
5.	Resin	+	+	++	+	+	+	++	+	-	+	+	-	++	++	+	+	+	
6.	Anthocyanin	++	++	+	+	+	+	+	++	++	+	+	+	+	+	++	+	++	
7.	Saponin	+	+	-	+	+	-	+	+	++	+	+	+	+	+	+	+	-	
8.	Steroid	+	+	+	+	+	-	++	++	++	+	+	-	+	+	+	+	+	
9.	Tannin	+	+	++	+	+	+	++	++	+	+	+	++	+	+	+	+	+	
10.	Starch	-	-	-	-	-	+	-	-	-	-	-	+	-	-	-	-	-	
11.	Glycoside	+	-	-	+	+	+	-	-	+	+	+	+	-	-	+	+	+	
12.	Phenol	+	++	++	+	+	+	++	+	++	+	+	++	+	+	++	+	+	
13.	Phlobatanin	-	-	+	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
14.	Terpenoid	+	++	+	-	-	-	++	++	++	+	++	-	++	+	+	+	+	

W = Water extract, PE = Petroleum ether extract, M = Methanolic extract

## REFERENCE

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