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### Phytochemical screening and ionomic analysis of selected RET medicinal plants of eastern Himalayan region

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#### Abstract

The study was conducted at Dept. of Horticulture, Sikkim University, Gangtok during 2017-2018 with an objective to study phytochemical and ionomic analysis of RET medicinal plants of Himalayan region. In the present study on phytochemical and ionomic analysis of RET medicinal plants, the stem and leaf samples of Swertia chirata, Clematix bouchiana, Gingko biloba and Valeriana jatamansi were collected from Kalimpong, W.B. The selected medicinal plants have been recommended as remedies for innumerable ailments in traditional medicinal system. The phytochemicals are thought to be largely responsible for the protective health benefits and are naturally occurring compounds in the plants. Among phytochemicals, screening was done for alkaloids, flavonoids, tannins, terpenoids and saponins. Study of trace elements in (RET) medicinal plants is of prime importance and ICP-MS was used for ionomic analysis. The presence of (Ba, Pb, Al, Ca, Cd, Co, Cu, S, Mg, Na, B, Zn, Hg. Si.) elements below the permissible limits for consumption were reported in this study. All the phytochemicals i.e. alkaloids, flavonoids, tannins and terpenoids were found in all the plants in this study except saponins. Diversity of groups of phytochemicals in the medicinal plants taken in present study helps us to strengthen potential of use of plants under this study as medicines and emphasis should be given on their intensive cultivation as well development of standard package of practices of these RET medicinal plants.

Keywords: RET medicinal plants, phytochemical screening, ionomic analysis

#### 1. Introduction

India has two out of the 18 biodiversity hot-spots in the world, which are in the Western Ghats and Eastern Himalayas. Sikkim, covering just 0.2% of the geographical area of the country harbours more than 26% flowering plants and has tremendous biodiversity and has been identified as one of the hot spots in the Eastern Himalayas. There is a growing concern throughout the world that the natural resources essential for human development and survival are being depleted and destroyed at an alarming and ever increasing pace. It is, therefore, necessary to plan development activities in such a manner that the precious environment is conserved on a sustainable basis. Conservation of rare, endangered and threatened (RET) plant species is an important issue. Hundreds of RET plants in India have already been recorded and their conservation was suggested. The Red Data Book has enlisted 622 vascular plant species (VPS) of Indian flora till 1990 this red figure rose to 1255 VPS3 till 2003, and it is on the increase day by day. In India, the RET species constitute 7.7% of known VPS3. Globally, 13.8% of VPS are RET (Rare, endangered and threatened). According to the International Union for Conservation of Nature and Natural Resources, the current species extinction rate is between 1000 and 10,000 times higher than it would naturally be. Once a species becomes extinct, the particular genetic resource is lost forever. Of the estimated 30,000 known medicinal plant species of the world, about 8,000 are found in India (Kumar and Katakam, 2002) [11].

In recent years, the demand for Indian medicinal plants has increased considerably both at local and global levels, which can be estimated by position of the country as the second largest exporter of raw herbal drugs to the global market. India is a major exporter of medicinal plants and is estimated that raw materials and drugs from medicinal plants of approximately 860 million are exported annually from India. In India, more than 41 million tribals and forest dwellers derive their earnings from these products.

India is one of the major exporters of crude drugs mainly to the six developed nations i.e. about 75% of the total exports as reported by Bhat *et al.* (2018) <sup>[5]</sup>.

Major part of the exported raw material includes medicinal plants of the Himalayan Region (Rawat and Garg, 2005)<sup>[18]</sup>. It is estimated that approximately 90% of medicinal plants in use are collected from the wild in which 70% collection involves destructive harvesting (Ved et al., 1998)<sup>[21]</sup>. Due to lack of cultivation and prevailing ruthless in-situ harvesting, populations of these valuable plants are diminishing day by day coupled with loss of genetic diversity, habitat degradation and facing high risk of extinction. Resultantly, 17 species have already been placed in the Red Data Book of Indian plants (Nayar and Sastry, 1987, 1988, 1990)  $^{\left[15,\ \right]}$  and a number of plant species have become endangered and/or facing various levels of threats (Ved et al., 2003; Ved et al., 2005) <sup>[22-23]</sup>. Well known species threatened by excessive wild harvesting in the region include Taxus baccata, Swertia chirata, Piccorhiza kurroa, Valeriana jatamansi. These species are valued for yielding anti-cancerous drugs, Taxol and Podophyllotoxin respectively. If the threat continues and necessary steps are not taken, it is estimated that between 4000 and 10,000 medicinal plant species might now be endangered at global level. Study of trace element and toxic heavy metal is of prime importance as they play important role in the formation of active chemical constituents present in medicinal plants and they are, therefore, responsible for their medicinal as well as toxic properties (Abugassa et al., 2008) <sup>[1]</sup>. Some metals are essential nutrients (zinc, iron, copper, chromium, and cobalt) but are toxic at high concentration, while others (lead and cadmium) exclusively toxic without any beneficial effect.

Many medicinal herbs and their mixtures can present a health risk due to the presence of toxic elements. The quantitative estimation of various trace element concentrations is important for determining the effectiveness of the medicinal plants in treating various diseases and also to understand their pharmacological action. Moreover, trace element analysis of medicinal plants can be used to decide the dosage of the herbal drugs prepared from these plant materials. Therefore, determination of element compositions in foods and related products is essential for understanding their nutritive importance (Abugassa *et al.*, 2008; Nookabkaew *et al.*, 2006) <sup>[1, 16, 1]</sup>. The study was conducted with an objective to study phytochemical and ionomic analysis of selected RET medicinal plants of eastern Himalayan region.

#### 2. Materials and Methods

- i) The methodology employed during the course of investigation is being described in this section. The steps followed were (i) Collection of plant(s)
- ii) Authentication of plant material
- iii) Shade drying of plant material i.e. Leaves, Bark, Seeds, Root and Flowers
- iv) Oven drying of plant material i.e. Root/rhizome ring the course of investigation are described below
- v) Phytochemical Investigation(s)
- vi) Digestion of sample
- vii) Volume make up of digested sample
- viii) Multi elemental analysis using ICP-MS

The plants which were used for phytochemical and ionomic study were as follows:-

i) Swertia chirata

- ii) Clematis bouchiana
- iii) Gingko biloba
- iv) Valeriana jatamansi

#### 2.1. Phytochemical Investigation

The phytochemical test of the rare endangered and threatened species of medicinal plants was carried out using aqueous and ethanol extracts on the powdered specimens using standard procedures to identify the various constituents described by Bargah (2015)<sup>[2]</sup> and Edeoga *et al.* (2005)<sup>[6]</sup>.

#### 2.1.1. Collection and authentication of plant samples

The plant samples of leaves, stems, rhizomes of different medicinal plants were collected from the Uttar Banga Krishi Viswavidyalaya, Kalimpong during the month of December 2017.

#### 2.1.2. Drying of samples

The collected plant samples like (leaves, stems, rhizomes) were cleaned with tap water twice and were air dried. The rhizomes were thinly sliced and were oven dried at  $(40^{\circ}C)$  for 2 days and were ground into uniform powder using mortar and pestle.

#### 2.1.3. Preparation of plant extracts

After drying the powdered plants, samples were subjected to aqueous extraction and ethanol extraction for alkaloids, anthraquinones, tannins and terpenoid qualitative analysis,

Aqueous extract was prepared from dried powdered sample. Each sample was weighed 10 g on electronic weighing balance. Weighed samples were transferred into beaker and 20 ml of distilled water was added. The beakers containing samples were properly sealed with aluminium foil and were left at room temperature for 12 hours (Edeoga et al., 2005)<sup>[6]</sup>. For flavonoids, saponin ethanol extract was prepared from dried powdered sample. Each sample was weighed 2g on electronic weighing balance. Weighed samples were transferred into beaker and 6ml ethanol was added to it. The beakers containing samples were properly sealed with aluminium foil and were left at room temperature for 72 hours (Bargah *et al.*, 2015)<sup>[2]</sup>. Both the ethanol and aqueous extracts were filtered using funnel and filter paper of Whatman no. 42. The extracts obtained were used for phytochemical screening and left over were stored in a universal bottle and refrigerated prior to use (Bargah et al., 2015)<sup>[2]</sup>.

# 2.2. Qualitative estimation of phytochemicals 2.2.1. Chemical test for Alkaloids

This test was carried out by taking 2ml of plant extract in 2ml of concentrated hydrochloric acid was added. Then few drops of Mayer's reagent were added. Presence of green colour or white precipitate indicates the presence of alkaloids.

#### 2.2.2. Chemical Test for Flavonoids

This test was performed by using 1 ml of plant extract to which 1ml of 10% lead acetate solution was added. The formation of yellow precipitate was taken as a positive test for presence of flavonoids.

## **2.2.3.** Chemical Test for Tannins Ferric chloride test

This test includes 0.5 g of the dried powdered samples which were boiled in 20 ml of water in a test tube and then filtered. Few drops of 0.1% ferric chloride was added to extract and

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brownish green or blue-black colour confirms presence of tannins.

# 2.2.4. Chemical Test for Terpenoids Salkowski test

This test was carried out by taking 5ml of each extract mixed in 2 ml of chloroform and concentrated  $H_2SO_4$  (3 ml) carefully added to form a layer. A reddish brown colour of the inter face was formed to show positive results for the presence of terpenoids.

#### 2.2.5.Chemical Test for Saponins

This test was carried out by taking 5 ml of extract and it was shaken vigorously with 5 ml of distilled water in a test tube and warmed in a water bath at  $60^{\circ}$ C. The formation of stable foam indicates presence of saponins.

#### 2.3. Elemental Analysis

Minerals or elements serve as structural components of tissues. They function in cellular and basal metabolism, water and acid-base balance, clotting of blood and formation of bones and teeth etc. Inductively Coupled Plasma-mass Spectrometry (ICP-MS) was used for the determination of elements in the dried powder of leaves, stems and rhizomes.

Elemental analysis by ICP-MS was conducted on the subset of 4 samples. The elements under consideration were toxic elements and trace elements. Collected plant samples were washed thoroughly and were sun dried and rhizomes were oven dried. Dried samples were ground in powder form in mortar and pestle and stored in sample container for further use. In case of total extractable elements digestion was carried out in an open air digestion system which is inexpensive and can be easily automated. All relevant parameters (time, temperature, introduction of digestion reagents). For digestion powdered samples were weighed 0.5g using weighing balance. Nitric acid (10 ml) and perchloric acid mixture (diacid) in ratio of 9:11 were added for digestion. The white fume and colourless solution confirmed the completion of digestion of powdered sample. The flasks containing digested sample were allowed to cool and then filtered using Whatman no. 1 filter paper. Filtrate samples volumes were made up to 50ml

using distilled water. The samples were digested with concentrated nitric acid and hydrogen peroxide in 9:4 ratio. The digested samples were stored in narrow mouth bottle at room temperature.

#### 2.3.1.Multi-Elemental Analysis Using ICP-MS

The multi-elemental analysis was performed using ICP-MS as prescribed by Perkin- Elmer Instrument Manual. Sample analysis was carried out by Perkin-Elmer Inductively coupled plasma mass spectrometry (ICP-MS) system with cross flow nebulizer. Digested samples were analyzed for the ionic constitution using Multi-elemental standard supplied by Perkin-Elmer containing analytes Ag, Al, B, Ba, Be, Bi, Ca, Cd, U, Co, Cr, Cs, Cu, Fe, Ga, Ge, In, K, Li, Mg, Mn, Mo, Nb, P, Rb, Se, Si, Ta, Ti, V, W and further the instrument was calibrated using standard reference material. After calibration, digested samples were analyzed for ionomic profiling.

#### 3. Results and Discussion

The results obtained in the present study are being illustrated as follows:-

#### 3.1 Phytochemical screening of RET medicinal plants

During the present study, ethanolic and aqueous extract of selected rare endangered and threatened species of medicinal plants of himalayan region were subjected for testing the presence of different metabolites mentioned above. With reference to Table no. 1, the results are being represented as follows:-

Sl. No.	Scientific name	Alkaloids	Flavonoids	Tannins	Terpenoids	Saponins
1.	Swertia chirata (leaves)	+	+	++	+	NA
2.	Swertia chirata (stem)	+	+	++	+	NA
3.	Ginkgo biloba (leaves)	+	+	+	+	NA
4.	Ginkgo biloba (stem)	+	+	-	+	NA
5.	Clematis bouchiana (leaves)	+	_	+	+	NA
6.	Clematis bouchiana (stem)	+	_	+	+	NA
7.	Valeriana jatamansi (leaves)	+	+	+	+	NA

Table 1: Phytochemical study of alkaloids, flavonoids, tannins, terpenoids and saponins

**Note:** +Positive, -Negative, NA= Not Accessed, ++ higher intensity of colour observed and indicates presence of respective phytochemical in higher quantity

**3.1.1. Swertia chirata**: Leaf and stem showed positive result in most of the phytochemical analysis, except saponins. The study conducted by Subedi, *et al.* (2018) <sup>[19]</sup> showed negative result in the saponin from leaf and stem of *Swertia chirata*.

**3.1.2. Clematis bouchiana:** Leaves and stem showed all the positive results in phytochemicals except the saponins. In the study conducted by Hawaze *et al.*,  $(2012)^{[7]}$  in their studies on phytochemical screening of Clematis species *C. longicauda and C. burgensis* leaves they found the positive result of saponin in methanol and water extract and negative in petroleum ether and here in *Clematis bouchiana* the absence may be due to the regional or climatic factors. In other studies carried out by Teshome *et al.*, (2013) in *Clematis simensis* the species of Clematis shows the negative

result of saponin content in different extracts *viz* petroleum ether acetone and methanol.

**3.1.3. Ginkgo biloba:** Leaves and stem showed all positive results in most of the phytochemical analysis tested in ethanol and aqueous solution but saponin was not found. The study conducted by Ibrahim, *et al.* (2016) <sup>[9]</sup> showed the positive result of saponin in their experiments on *Ginkgo biloba*. In present study the results were contrary and it might be due to climatic factors.

**3.1.4.** *Valeriana jatamansi*: Leaves showed positive results in most of the phytochemical analysis tested in ethanol and aqueous solution except saponins which were not defined. The findings of Babu *et al.*, (2017)<sup>[3]</sup> shows negative result in

saponin content of *Valeriana jatamansi* in different parts of leaves, rhizome etc.

#### 3.2 Ionomic analysis of RET medicinal plants under study

It is evident from Table no. 2 and as per analysis with ICP-MS the concentration of various elements found in RET medicinal plants under study is being represented as follows:-

#### 3.2.1. Barium (Ba)

In the present study barium was found highest in *Swertia* chirata stem i.e.  $125.13\mu g/g$  and lowest in leaves i.e.  $67.73\mu g/g$  and in *Clematis bouchiana* the highest was in leaf i.e.  $32.58\mu g/g$  as compared to stem that is  $31.9\mu g/g$  in *Ginkgo* biloba concentration is much higher i.e.  $1396.01\mu g/g$  as compared to stem i.e.  $61.11\mu g/g$  and in *Valeriana jatamansi*  $1302.31\mu g/g$ .

#### 3.2.2. Lead (Pb)

Lead was found in high concentration in leaves of *Swertia chirata* i.e. $0.51\mu g/g$  as compared to stem that is  $0.26 \mu g/g$  and it is much below the permissible limits i.e.  $1000 \mu g/g$  recommended for medicinal plants (WHO) 2007. In *Clematis bouchiana* concentration is higher in leaf i.e.  $0.18\mu g/g$  as compared to leaves i.e. $0.2\mu g/g$  and *Ginkgo biloba* the higher concentration is found in leaves  $0.56\mu g/g$  as compared to stem

i.e.0.55µg/g and in Valeriana jatamansi it was 0.33µg/g.

#### 3.2.3. Aluminium (Al)

The highest concentration is found in the leaf of *Swertia* i.e.633.13 $\mu$ g/g as compared to stem i.e.37.13  $\mu$ g/g. in *Clematis bouchiana* higher concentration is found in leaves i.e. 29.53 $\mu$ g/g as compared to stem i.e. 5.09 $\mu$ g/g in *Ginkgo biloba* highest is found in leaves i.e. 104.77 $\mu$ g/g as compared to leaves i.e. 41.84 $\mu$ g/g and in *Valeriana jatamansi* 174.47 $\mu$ g/g. Orally administered aluminium compounds include iron, fluoride, phosphorus, strontium and to a lesser extent calcium (Parag *et al.*,2013).

#### 3.2.4. Calcium (Ca)

Calcium concentration was found highest in leaves of Swertia i.e.  $35.45\mu g/g$  as compared to stem i.e.  $25.47\mu g/g$  in *Clematis bouchiana* the highest concentration is in leaves i.e.  $75.55\mu g/g$  as compared to stem i.e. $21.24\mu g/g$  in *Ginkgo biloba* highest concentration is found in leaves i.e. $284.54\mu g/g$ as compared to leaves i.e.  $155.21\mu g/g$  and in *Valeriana jatamansi* 271.31\mu g/g Among all the health benefits of calcium is the most important ones are that it aids in maintaining bone health and dental health as well prevention of colon cancer and reduction of obesity.

Elements	Swertia chirata	Swertia chirata	Clematis bouchiana	Clematis bouchiana	Gingko biloba	Gingko biloba	Valeriana jatamansi
	(leaf)	(stem)	(leaf)	(stem)	(leaf)	(stem)	(leaf)
Ba	67.73	125.13	32.58	31.59	1396.01	61.11	1302.31
Pb	0.51	0.26	0.18	0.2	0.56	0.55	0.33
Al	633.13	37.13	29.53	5.09	104.77	41.84	174.47
Ca	35.45	25.47	75.55	21.24	284.54	155.21	271.31
Cd	0.3	0.23	0.27	0.42	0.06	0.12	0.11
Co	1.19	0.2	0.12	0.07	0.43	0.29	0.52
Cu	18.04	18.14	11.2	16.3	7.27	9.74	10.39
S	20.35	27.23	15.48	21.52	21.21	17	23.8
Mg	425.31	354.19	257.32	209.4	401.44	342.76	611.98
Na	5.93	3.87	1.78	2.43	9.52	6.46	23.1
В	0.87	1.25	0.68	0.91	4.13	1.02	4.36
Zn	1.31	1.52	0.38	0.58	0.34	0.38	0.63
Hg	0.34	0.23	0.1	0.26	0.74	0.56	0.44
Si	10.7	3.73	3.69	0.86	7.62	2.92	6.83

#### 3.2.5. Cadmium (Cd)

Cadmium was found highest in stem of *Swertia chirata* i.e.  $0.23\mu g/g$  as compared to leaves that was  $0.3\mu g/g$  in *Clematis bouchiana* concentration is higher in stem i.e. $0.42\mu g/g$  as compared to leaves i.e. $0.27\mu g$  in Ginkgo biloba highest concentration is found in stem i.e.  $0.12\mu g/g$  as compared to leaves i.e.  $0.06\mu g/g$  and in *Valeriana jatamansi*  $0.11\mu g/g$ . As it is below permissible limits. Samecka-cymerman. *et al.*, (2010) <sup>[20]</sup> reported mean concentration of cadmium to the tune of  $90\mu g/g$ . in *Taxus baccata* leaves from Poland.

#### 3.2.6. Cobalt (Co)

Maximum concentration of cobalt is found in leaves of *Swertia chirata* i.e.  $1.19\mu$ g/g as compared to stem i.e.  $0.2\mu$ g/g in Clematis the higher concentration is in leaves i.e.  $0.12\mu$ g/g as compared to stem i.e. $0.07\mu$ g/g and in *Ginkgo biloba* the higher concentration is in leaves i.e.  $0.43\mu$ g/g as compared to stem i.e. $0.29\mu$ g/g and in *Valeriana jatamansi* leaves contain  $0.52\mu$ g/g. The study was carried out on eight herbs in Kenya as reported by Maobe *et al.*, 2012 <sup>[12]</sup> where high cobalt concentration was reported as compared to the present study

#### i.e. $967\mu g/g$ to $6067\mu g/g$ .

#### 3.2.7. Copper (Cu)

Copper concentration in tested plants was found to be the highest as in *Swertia chirata* stem i.e.  $18.14\mu$ g/g as compared to leaves i.e.  $18.04\mu$ g/g and in *Clematis bouchiana* highest is in stem i.e.  $16.3\mu$ g/g as compared to leaves i.e. $11.3\mu$ g/g. In *Ginkgo biloba* the highest concentration is in stem i.e.  $9.74\mu$ g/g as compared to leaves i.e. $7.27\mu$ g/g and in *Valeriana jatamansi* leaves the concentration is  $10.39\mu$ g/g respectively. Magia (2005) <sup>[13]</sup> reported that the copper concentration in seven medicinal plants was found in range of  $2.4-17.1\mu$ g/g.

#### **3.2.8. Sulphur (S)**

The concentration of sulphur was found highest in *Swertia* chirata stem i.e.  $27.23\mu g/g$  as compared to leaves i.e.  $20.35\mu g/g$ . In *Clematis bouchiana*, higher concentration was found in stem i.e.  $21.52\mu g/g$  as compared to leaves i.e.  $15.48\mu g/g$ . In *Ginkgo biloba* the higher concentration was found in leaves i.e.  $21.21\mu g/g$  as compared to stem i.e.  $17\mu g/g$  and in *Valeriana jatamansi* the concentration in leaves was

#### 23.8µg/g.

Following calcium and phosphorus, sulfur is the third most abundant mineral in the human body, representing ~0.3% of total body mass (Hewlings and Kalman, 2019)<sup>[8]</sup>. Sulphur is taken in as sulphur containing amino acids cysteine and methionine by human beings. The recommended dietary allowance for SAAs for human beings is 14 mg kg<sup>-1</sup> body weight. Lack of sulphur can lead to arthritis, muscle and joint stiffness, spondylitis etc. (Prasad and Shivaay, 2016)<sup>[17]</sup>.

#### **3.2.9. Magnesium (Mg)**

Magnesium was highest in *Swertia chirata* leaves i.e.  $425.31\mu g/g$  as compared to stem i.e.  $354.19\mu g/g$  and in *Clematis bouchiana* the highest concentration was found in leaves  $257.32\mu g/g$  as compared to stem i.e. $209.4\mu g/g$ . In *Gingko biloba* the highest concentration was found in leaves i.e.  $401.44\mu g/g$  as compared to stem i.e.  $342.76\mu g/g$  and in *Valeriana jatamansi* the concentration in leaves was  $611.98\mu g/g$ . Bardarov *et al.* (2015)<sup>[4]</sup> reported in his research on medicinal plants *Clinopodium vulgare* from Bulgaria that they found Mg concentration leaves as  $7004\mu g/g$  and  $3472\mu g/g$  in leaves and stem respectively.

#### 3.2.10. Sodium (Na)

Sodium was found highest in *Swertia chirata* leaves i.e. $5.93\mu g/g$  as compared to stem i.e.  $3.87\mu g/g$  and in *Clematis bouchiana* the highest concentration was found in stem i.e.  $2.43\mu g/g$  as compared to leaves i.e. $1.78\mu g/g$ . In *Gingko biloba*, the highes. t concentration was found in leaves i.e. $9.52\mu g/g$  as compared to stem i.e. $6.46\mu g/g$ . In *Valeriana jatamansi* leaves Na concentration was found to be  $23.1\mu g/g$ . Sodium is a mineral and one of the elements found in salt. The human body requires sodium in relatively small amounts

#### 3.2.11. Boron (B)

The concentration of boron was found highest in *Swertia* chirata stem i.e.  $1.25\mu$ g/g as compared to leaves i.e.  $0.87\mu$ g/g. In *Clematis bouchiana* the higher concentration was found in leaves i.e.  $1.25\mu$ g/g as compared to stem i.e.  $0.68\mu$ g/g. In *Gingko biloba* the highest concentration was found in stem i.e.  $4.13\mu$ g/g as compared to leaves i.e. $0.91\mu$ g/g and in *Valeriana jatamansi* boron concrentration in leaves was found to be  $1.02\mu$ g/g.

#### 3.2.12. Zinc (Z)

The zinc concentration was highest in *Swertia chirata* stem i.e.  $1.52\mu$ g/g as compared to leaves i.e. $1.31\mu$ g/g. In *Clematis bouchiana* the higher concentration was found in stem i.e.  $0.58\mu$ g/g as compared to leaves i.e.  $0.38\mu$ g/g. In *Gingko biloba*, the highest concentration was found in stem i.e.  $0.38\mu$ g/g as compared to leaves i.e. $0.34\mu$ g/g and in *Valeriana jatamansi* zinc concentration in leaves was found to be  $0.63\mu$ g/g. Narayana *et al.* (2013) <sup>[14]</sup> reported in studies on trace elements of medicinal plants *Azadirachta indica* and *Ocimum sanctum* the concentration of zinc was found higher i.e. 200.8\mug/g to 254.5 $\mu$ g/g

#### 3.2.13. Mercury (Hg)

The concentration of mercury was found to be highest in Swertia chirata leaves i.e.  $0.34\mu g/g$  as compared to stem i.e.  $0.23\mu g/g$ . In Clematis bouchiana the highest concentration was observed in stem i.e. $0.26\mu g/g$  as compared to leaves i.e. $0.01\mu g/g$ . In Ginkgo biloba the higher concentration was found in stem  $0.74\mu g/g$  as compared to leaves i.e. $0.56\mu g/g$ 

and in *Valeriana jatamansi* leaves showed concentration of mercury as 0.44µg/g.

#### 3.2.14. Silicon (Si)

The silicon concentration was found to be highest in Swertia chirata leaves i.e10.7µg/g as compared to stem i.e.3.75µg/g and in Clematis bouchiana the concentration was found highest in the leaves i.e.3.69µg/g as compared to stem i.e. 0.86µg/g and in Gingko biloba the highest concentration was found in leaves i.e. 7.62µg/g as compared to stem i.e. 2.91µg/g. In Valeriana jatamansi the concentration of silicon was observed as 6.83 µg/g The beneficial elements are not deemed essential for all crops but may be vital for particular plant taxa. The distinction between beneficial and essential is often difficult in the case of some trace elements. Silicon (Si) is considered of beneficial for plants. This elements is not critical for all plants but may improve plant growth and yield. Pertinently, beneficial elements reportedly enhance resistance to abiotic stresses (drought, salinity, high temperature, cold, UV stress, and nutrient toxicity or deficiency) and biotic stresses (pathogens and herbivores) at their low levels (Kaur et al., 2015)<sup>[10]</sup>.

#### 4. Conclusion

All the detected elements in RET species of medicinal plants were detected below permissible limits. As phytochemicals are thought to be largely responsible for the protective health benefits and are naturally occurring compounds in the plants. All the phytochemical alkaloids, flavonoids, terpenoids, saponins and tannins were found in tested plants. Diversity of groups of chemicals in the medicinal plants taken in present study helped in strengthening the potential of plants under present study as medicines. These plants under present study can be cultivated intensively for development of next generation drugs.

#### 5. Acknowledgements

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