#### **Original Article**

## Medical and medicinal importance of *Rheum spp*. collected from different altitudes of the Kashmir Himalayan range



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#### 1. Introduction

The *Rheum* plant (commonly called Rhubarb), belongs to the family Polygonaceae of the group Monochlamydae of Dicots, is the world over-represented by 60 species, of which only 7 species have been reported from the Indian subcontinent [1]. According to Stewart [2], all the seven species (including *R. emodi, R. spiciforme & R. webbianum*) reported from the Indian subcontinent are also present in the Kashmir Himalaya. In taxonomical focus, inflorescence is pyramidal or spherical in which flowers are arranged in panicles and pedicel articulates at or below the middle. Rhubarb is derived from the Latin word "*rha*" (river) and "*barb*" (barbarian land) [1, 3].

<u>ABSTRACT</u>

Most of the plants are essential due to their medicinal properties and have contributed to the origin and evolution of many traditional herbal therapies. Rheum is also one of the famous and known medicinal plants of Kashmir Himalaya. It ranges from an altitude of 1700-5500 m. Its Roots are long and stout. The stem is erect, hollow, sulcate, glabrous, or strigose. It is native to Asia-Temperate to Asia-Tropical, from China to India, Nepal, and Pakistan. In India, its found in Himachal Pradesh, Jammu and Kashmir, and Uttar Pradesh. Rhubarb is the English name of the genus Rheum (family Polygonaceae). Emodi, Webbianum and Spiciforme are three species of Rheum found in Kashmir Himalaya. Rheum species are wellknown medicinal plant and is found on high altitudes. Its leaves and stalks are taken as food and cooked as vegetables. It's used as medicine and is generally proven more effective against certain ailments like antibacterial, anticancerous, anti-inflammatory, rheumatoid diseases, stomachache, and intestinal problems. It is also used for the treatment of indigestion, abdominal disorders, boils, wounds and flatulence. But Rheum has become endangered due to exploitation. Active compounds like aloe-emodin, rhein, emodin, chrysaphanol and physicion are found in this plant which is anti Cancerian agents. High molecular diversity is also observed in all the three species of Rheum.

> In ancient times, rhubarb roots were imported by the Romans from barbarian lands which were beyond the Volga or Rha River. According to Lindley's Treasury of Botany, the name is derived from the Greek rheo ('to flow'), in allusion to the purgative properties of the root. A total of 560 plant species of India are included in the Red List of Threatened species by IUCN, including *Rheum*species [1, 3]. *Rheum emodi* is included in critically endangered species. Thus it was identified as top priority species for conservation and cultivation. Due to its endangered nature, the export of *R. emodi* is banned or needs a special export permit [4, 5].

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*Rheum* is a strong vigorous plant and can live long, even in tough conditions. Rhubarb is a plant name for the many different species of *Rheum*, distributed in the temperate and subtropical regions of the Himalayas from Kashmir to Sikkim (Nautiyal *et al.* 2003) and growing wild in the mountains of the Western and North-western provinces of China and adjoining Tibetan territory. In Jammu and Kashmir, it grows on open slopes and shrubberies in Kashmir, Leh and Zanskar valley between 3,105-3,920 m [6].

Rheum emodi, commonly known as Himalayan rhubarb, is a medicinal herb Indian Ayurvedic system used in the of medicine. Rhubarbs are large perennial herbs. The flower stalk is solid or hollow and reaches a height of 1.5-2 m. The radical leaves, which have long, succulent petioles, form a rosette and are very large and usually entire. Rheum emodi can be found at elevations from 8,000 to 13,000 feet. This species has huge leaves which measure up to 3 feet in width. The plant can reach heights of 9 feet, especially in fertile soil. The inflorescence is panicled or spicate [7, 8].

The flowers are bisexual; male flowers sometimes occur. The fruit is brown, three-angled, winged achene. *Rheum* also prefers a full sun location and thrives in moist, fertile soil. Most *Rheum* can handle heavier clay soils better than many other herbs and makes a striking addition to the spring landscape [7, 8].

Rhubarb is propagated from seeds, seedlings, or pieces of rhizome. The first crop is obtained two years after planting. The stalks, which measure 25–70 cm long and 1.5–4 cm thick, are gathered (by breaking, cutting, or mowing) from early May through the first ten days of June. The yield in the sixth or seventh year is 250–300 quintals per hectare. An early harvest (by ten to 15 days), with twice the normal yield, may be obtained by covering the plants with a synthetic film. Rhubarb can also be raised in hothouses from three- or four-year-old rhizomes[7, 8].

The root of *Rheum* is commonly sold as a remedy for stimulating the bowels and has a mild purgative action when used in small quantities and should be used with some caution as its effect can be uncomfortable in

large doses. The leaf stalks, which are edible, contain as much as 2.5 percent sugars and approximately 3.5 percent organic acids (primarily malic, citric, oxalic, and succinic acids). They also contain potassium, calcium, phosphorus, and magnesium salts. Rhubarb is used to make compote, jelly, jam, and wine. The roots of *R. palmatum* var. *tanguticum* contain the laxatives emodin, chrysophanic acid, and anthracene glycosides; they are used medicinally in the form of powders, tablets, alcohol tinctures, and syrups [7-9].

*Rheum spiciforme* is a perennial growing to 0.3 m (1ft). Suitable for: medium (loamy) and heavy (clay) soils, prefers well-drained soil and can grow in heavy clay soil. It can grow in semi-shade (light woodland) or no shade prefers moist soil, flowers in June, and the seeds ripen in August. The leaves are large, rounded or heart-shaped, 15-30 cm across, thick and leathery, becoming red-brown on maturity. Leaves are carried on very stout stalks, 5-15 cm long, which also turn reddish. Flowers are greenish, turning reddish, and 3 mm across. Flowers are borne in 1-3 erect cylindrical spikes, 5-30 cm tall. The flowers are hermaphrodite (have both male and female organs) and are pollinated by Wind [10, <u>11</u>].

*Rheum* spiciforme is a medicinal plant of immense importance with large pharmacological applications. Besides having the above-mentioned pharmacological properties, it has been used as an ingredient of many herbal formulations, which are used for the cure of various ailments, in particular the regulation of blood pressure, fat, hepatitis, fever and cancer[10, 12].

*Rheum webbianum* is a tall perennial herb. It is found in all areas of Gilgit /Baltistan. Stem leaves ovate, smaller. Panicle large, 1- or 2branched. The flowers are small. Tepals are yellow-white, elliptic, jointed below the middle of the pedicel. Leaves orbicularcordate, 5-7 nerved, papillose or glabrous, tip rounded or sub acute, petiole long Inflorescence, panicle, axillary and terminal, flowers smaller pale yellowish. Fruit broadly ellipsoid or orbicular, 1-1.2 cm, ca. as long as wide, both ends slightly retuse; wings ca. 3.5 mm, broad, longitudinal veins near margin. Seeds are narrowly ovoid-ellipsoid, ca. 4 mm wide [10, 12].

The roots, stems, leaves and leaf stalks are purgative and are beneficial in treating indigestion, abdominal diseases, astringent, boils, purgative, wounds and flatulence. The roots are diuretic, laxative, purgative, febrifuge; used against indigestion, wounds and gastritis etc [6]. Rhubarb is extensively used in traditional Chinese medicine. Rhubarb has been studied for the management of gastrointestinal (GI) and renal function disorders and for the treatment of hyperlipidemia and cancer. Dried Rhubarb extract 20 to 50 mg/kg daily has been used in clinical trials. *Rheum*webbianum is used in the treatment of cancer. ulcer and liver ailments. The anthraguinone derivatives obtained from the rhizome/root extracts are found to possess antioxidant and anticancer activity. The root is purgative, and other uses are similar to *R.emodi* [10, 12].

Rhubarb contains a large number of derivatives anthraquinone such as chrysophanol, physcion, emodin, aloe emodin, emodin glycoside, rhein, etc. The methods commonly used to determine the anthraquinone compounds in Rhubarb are thin layer chromatography (TLC) and highperformance liquid chromatography (HPLC). Anthocyanins and flavonols also found in rhubarb may have biological effects in their own right [<u>13</u>, <u>14</u>].

roots of Rheumaustrale The (svn. Rheumemodi WALL, Polygonaceae) are widely used in Ayurvedic and Asian folk medicine. During the last years, several occurrences have caused a decline of Rheumspecies in the world. The increased rate of deforestation has put many of the specie in danger. It is extremely urgent to adopt management and conservation strategies. It is therefore essential to characterize, morphologically and genetically, the different populations of Rheum, which are considered the key elements in management programs.

#### 2. Medicinal properties of *Rheum*

It is used as a strong laxative and is known for its astringent effect on the mucous membranes of the mouth and nasal cavity[<u>15</u>]. Its astringent qualities help to improve bowel tone after it has purged the intestines, making it an excellent agent for improving the tone and health of the digestive tract. Its roots have been found useful in controlling gastrointestinal hemorrhage by promoting the formation of blood platelets. This increase in the number of platelets shortens blood clotting time and helps treat jaundice. It, therefore, has a truly cleansing action upon the gut, removing debris and then astringing with antiseptic properties [16, 17].

*R. webbianum* is used to treat indigestion, abdominal disorders, boils, wounds and flatulence. It improves the memory of senile patients, helpful in managing cancers [<u>18</u>, <u>19</u>].

R. emodi is purgative, stomachic. astringent, tonic and helps to cure skin diseases, used as an anti-oxidant<sup>[20]</sup>, antiviral nephroprotective activity [<u>21</u>], [22], antimicrobial, anti-tumor, anti-inflammatory, anticholesteroleamic. antifungal, antiparkinson, hepatoprotective [23], treatment of SARS [24], antidiabetic [25] and anti-cancer [26, 27], for textile dyeing [28] and used in cosmetics [29].

*R. spiciforme* is also used in the treatment of boils, wounds, *rheum*atic pain. Roots are frequently used to treat bone fractures, backache and joint pain. It is also used as an adultrant. The usage of rhubarb has now been extended into functional food as health foods [11, 12].

Phytochemical surveys are now acting as the first step toward the discovery of useful drugs. Plants are the richest resource of drugs in traditional systems of medicine, modern medicines, food supplements, folk medicines, pharmaceutical intermediates and chemical entities for synthetic drugs[30-32]. About 50 to 60% of pharmaceutical drugs are either of natural origin or obtained through the use of natural products as starting points in their synthesis [33]. The *Rheum* species have been domesticated as medicinal plants for human consumption, as its roots and stem are rich in anthraquinones such as emodin, aloe-emodin, physicion, rhein, chrysophanol, physcion, alizarin, citreorosein and chrysaphanol [<u>34</u>].

#### 2. Materials and Methods

#### 2.1. Surveys

*R. emodi* was collected from various altitudes like Yousmarg, Dodhpathri, Thajwas Glacier (Sonmarg), Affarwat (Gulmarg), Dagvan (Tarsar Marsar), Aaroo (Pahalgaam), Laar and Dhara. *R. webbianum* was collected from ladakh region i.e. Tangsti, Khardungla, Panzila Top (Zanaskar), Tangole (Zanaskar) and Parakhachik (Zanaskar). *R. spiciforme* was collected from Gurez area i.e. Dawar Hills, Dahi Nala, Chakwali, Habbakhatoon Mountain and Tragbal.

All the study sites are depicted in Fig. 1 and Table 1. The specimens were identified by

consulting the Flora of China [<u>35</u>, <u>36</u>] and Alpine Flora of Kashmir Himalaya [<u>37</u>]. Further, the confirmation of the specimens was done by comparing the specimens in the Herbarium, Department of Botany, University of Kashmir, Kashmir (KU) and Punjabi University, Patiala (PUN) and the authenticated voucher numbers have been obtained from both the universities.

From the list of surveyed localities, it is clearly reflected that a vast area presenting a variety of vegetation types has been covered with altitudes ranging from 3352.8 to 4921.5 m.



Fig. 1. Red Circles denote surveyed areas

Species	Population	Populati	Altitu
Rheum emodi	Yousmarg	YM	4267.
	Sonamarg	SM	3462.
	Pahalgaam	PG	3962.
	Dodhpathri	DP	3862.
PUP (59513)/	Gulmarg	GM	4476.
KU (2010- KASH)	Tarsar Marsar	ТМ	4657.
Mishij	Dhara	DH	3751.
	Laar	LR	3657.
	Tissue cultured plants	тс	<i>h</i>
Rheum webbianum	Khardungla (Leh)	КН	4921. 5
	Tangsti (Leh)	TS	4725.
PUP (59515)/ KU (2011- KASH)	Panzila Top (Zanaskar)	РТ	4593. 8
PUP (59514)/	Tangole	TG	4266.
KU (2011- KASH)	Parkhachik (Zanaskar)	РК	3938. 4
	Tissue cultured plants	тс	
Rheum	Dawar Hills	DW	4419.
	Chakwali	CW	4684.
	Dahi Nala	DN	3810.
PUP (59516)/	Satni	SM	3962.
KU (2012- KASH)	Habba khatoon	HM	4756. 8
	Tragbal	ТВ	3352.
	Tissue	ТС	Q 

**Table 1.** Information regarding the differentlocalities of Kashmir visited during the presentstudies

#### 2.2. Studies of morphological features

Morphological traits cover the root length, root thickness, rhizome length, rhizome thickness, leaf length, leaf breadth, leaf number, number of branches, number of nodes, and plant height. These parameters were studied for scoring data regardingmorphological features of the plant species. The nodes are absent in the case of *Rheum webbianum* and *Rheum spiciforme*. So, this parameter was not studied in these two species.

#### 3. Morphology of *Rheum* species

In the present study, *R. emodi, R. webbianum* and *R. spiciforme* collected from different altitudes (natural habitats) were studied by using different morphometric parameters (using standard techniques).

#### 3.1. Rheum emodi

It grows from thick rhizomes. It has large, broadly ovate or orbicular, streaked green, radical and triangular leaves which are elevated on long fleshy and stout petioles. The flowers are small, numerous, bisexual, greenish-white and have large compound leafy and panicle inflorescences. It is a leafy perennial herb, cordate at the base, often long, 5 to 7 nerved papilose beneath. The maximum root length i.e., 67 cm was observed in plants from Sonamarg area and the maximum root thickness i.e., 7.3 cm, was observed in plants from Yousmarg (Fig. 2).

The *R. emodi* from Laar site in its natural habitat was registered 7 per m2.The maximum plant height in meters i.e., 5.6m was observed in plants from Dodhpathri. Maximum number of branches i.e., 19 and leaves i.e., 28 were seen in plants from Gulmarg area. The Maximum number of nodes i.e., 10 were recorded in plants from Laar and maximum leaf breadth i.e. 63 cm was observed Sonamarg plant(fig 3).

#### 3.2. Rheum spiciforme

This species is also stout and leaves are basal, radical, dark green, broadly ovate or cordate, glabrous and the leaf blade is purplered abaxially, nearly leathery and has thick rhizome and thick roots. Flowers are small, numerous, greenish yellow and bisexual. In present studies, the maximum root length i.e. 64 cm and thickness, i.e. 8 cm was registered in plants of Chakwali and the maximum rhizome length, i.e.79 cm thickness with 59 cm breadth, was also observed in Chakwali plant (Fig 4).

The maximum plant height i.e. 2 m was observed in plants in the Chakwali area. The *R spiciforme* in its natural habitat in the Chakwali area was found to be higher in number (14 per m2). The maximum number of branches (14) and leaves (18) were recorded in plants of Habbakhatoon Mountain area (fig 5).

#### 3.3. Rheum webbianum

It can also be identified as having basal and cauline leaves. Its basal leaves are orbicular or broadly ovate, large and 30 - 60 cm long. It is abaxially pubescent, adaxially papilliferous or muricate, basal veins are 5, base is subcordate, deeply palmately 5-lobed, middle 3 lobes are pinnatisect, apex is narrowly acute. Stem leaves are ew and smaller than the basal ones. Ocrea and panicle are large. The flowers are small, bisexual, numerous and reddish pink in colour and grouped in large compound leafy inflorescences.

The root is stout, straight, and dark brown outside and yellow or red-yellow inside colour. The stem is stout, hollow, finely sulcate and glabrous. In present studies, the maximum root length i.e., 98 cm and thickness of root i.e., 14 cm of *R. webbianum* were registered in plants of Tangsti site (Fig 6).

The maximum rhizome thickness i.e., 68 cm of *R.webbianum* was observed in plants of Panzila Top and maximum rhizome length was registered in plants of Tangsti site. The maximum plant height i.e. 1.4m was observed in Panzila Top. The maximum number of branches i.e., 14 and leaves i.e., 18 were registered in plants of Parkhachik sites. The *R. webbianum* found in Parkhachik area was more in number i.e., 17/m2 (fig 7).



Fig. 2. Root of R. emodi (yousmarg)



Fig. 3. R. emodi in sonamarg



Fig. 4. Root of *R.spiciforme* in chakwali (Gurez)



Fig. 5. R. spicifome in Habba khatoon (Gurez)



Fig. 6. Root of *R.webbianum* in Tangsti (Leh)



Fig. 7. R.webbianum in Pakhachik (Zanaskar)

#### 3.4. Maintenance of germplasm

#### 3.4.1. (Ex-situ) in nursery

For germplasm establishment, plants, seeds, roots and rhizomes collected from different altitudes were grown in different beds in the Nursery at the University of Kashmir (Fig 8,9).



Fig. 8. (Ex-situ) in nursery



Fig. 9. Green house in University of Kashmir

#### 3.4.2. Growth in Nursery

The seeds, roots, rhizomes and whole plants collected from different locations were planted under natural, shady and suitable environmental conditions in the beds of net house at Naseem Bagh Campus of the University of Kashmir Srinagar. Each bed had its own name plate mentioning the place and date of collection. The plants were monitored on a daily basis for their growth responses (fig 10,11,12) *Rheum* is also called cliff hanger as it grows on rocks and at high altitudes from 2000 m to 6000 m. so it needs suitable and favorable conditions when grown ex-situ. So far, for establishing Rheum species in nursery, the soil was also taken from high altitudes along with the plant mixed with the soil of the net house. Different soil texture groups were also used with or without fertilizer urea.

### **3.4.2.1.** Response of *Rheum* species in the nursery

In the nursery, good seed germination ability was observed in natural soil, different soil texture groups and under the influence of urea. Sand was also mixed in the soil. However, maximum germination was recorded in sandy soil treated with urea in the ratio of 1:2.

#### 3.4.2.1.1. R. emodi

The germplasm of *R. emodi* collected showed a rapid growth response in nursery. The survival of the whole plants showed an increment from 70-90 % (fig.13)(table 2 & 3).

#### 3.4.2.1.2. *R. webbianum*

The germplasm of *R. webbianum* showed a slow growth response as compared to *R. emodi* in nursery. The survival rate during the year 2011-14 was 50-75 % (fig. 14) (table 4 & 5).

#### 3.4.2.1.3. R. spiciforme

The germplasm of *R. spiciforme* collected in 2011 also showed fast and good growth response and the survival rate observed in three years was 60-85%. (fig 15)(table 6& 7).

	Mean ± SD					
Sites/Altitude s (m)	Root leng th (cm)	Root thickn ess (cm)	Rhizo me length (cm)	Rhizo me breadt h (cm)	Leaf leng th (cm)	Leaf bread th (cm)
Gulmarg/447 6.68	47.6 ± 2.51 b	4 ±1ª	14 ± 1 <sup>b</sup>	$74 \pm 1^{\rm f}$	46 ± 1º	54 ± 1º
Dhara/ 3751.6	37.6 ± 2ª	2.3 ± 1.5 <sup>b</sup>	24 ± 1°	65 ± 1º	43.3 ± 1.5 <sup>d</sup>	32.6 ± 2.5 <sup>b</sup>
Pahalgaam /3962.4	38.3 ± 1.5ª	6.6 ± 1.5 <sup>d</sup>	12.6 ± 2ª	33.3 ± 1.5ª	26 ± 3.6 <sup>b</sup>	41.3 ± 3.2 <sup>e</sup>
Sonamarg/ 3462.2	67 ± 1 <sup>f</sup>	5.6 ± 1.5°	38.3 ± 2 <sup>f</sup>	$70 \pm 1^{\rm f}$	32 ± 2.6 <sup>e</sup>	63 ± 2 <sup>f</sup>
Yousmag/ 42.672	59 ± 1º	7.3 ± 1. <sup>5e</sup>	23 ± 2.6 <sup>d</sup>	42 ± 4.3 <sup>b</sup>	26.3 ± 1.5 <sup>b</sup>	27 ± 7.2ª
Dodhpathri/ 3862.4	49 ± 2°	3.3 ± 1.5 <sup>b</sup>	17 ± 2.6¢	38 ± 7ª	19.3 ± 1.5ª	37 ± 5 <sup>b</sup>
Tarsar Marsar/ 4657.2	$\begin{array}{c} 58 \pm \\ 2^{d} \end{array}$	$4\pm 2^{a}$	28.3 ± 2 <sup>e</sup>	52.6 ± 2 <sup>d</sup>	24.6 ± 3 <sup>b</sup>	31 ± 3 <sup>b</sup>
Laar/ 3657.6	47.3 ± 3 <sup>b</sup>	6.3 ± 1.5 <sup>d</sup>	36.6 ± 1.5 <sup>f</sup>	45.3 ± 3℃	50 ± 1 <sup>f</sup>	45 ± 3.6 <sup>d</sup>

**Table 2.** Morphological features (undergroundparts) of *R.emodi* growing under nurseryconditions

	± 5°	1.5	1.5	5	1	5.0
Values are	represe	ented	as me	an ±SD	(n=3),	Data
was analyz	ed by A	NOV	A using	g Dunca	ın's mu	ıltiple
range test	(SPSS1	7.0);	the va	lues w	ith diff	ferent
superscript	along	the	colum	ns are	statist	cically
significant a	t P<0.0	05				

**Table 4.** Morphological features (undergroundparts) of *R.webbianum* growing under nurseryconditions

			Mean	± SD		
Sites/Altitudes (m)	Root leng th (cm)	Root thickn ess (cm)	Rhizo me length (cm)	Rhizo me breadt h (cm)	Leaf leng th (cm)	Leaf bread th (cm)
Panaliza	38 ±	4.+1.5h	<b>2</b> ∔ 1h	24 ±	19 ±	39 ±
Top/4593.8	1 <sup>b</sup>	<b>∓</b> ±1.5°	2± 1°	1.32 <sup>b</sup>	1ª	1 <sup>b</sup>
Tangole/4266.	34 ±	2 + 12	1.3 ±	26 ±	24 ±	44 ±
1	1 <sup>b</sup>	$3 \pm 1^{\circ}$	0.1ª	1 <sup>b</sup>	$1^{b}$	1¢
Parkachik/393	18 ±	3 ±	7.9 ±	25 ±	34 ±	46 ±
8.2	1a	1.4ª	1.2c	1.45 <sup>b</sup>	1e	1e
Tangsti/4725.	98 ±	$14 \pm 14$	13.4 ±	29 ±	24 ±	24 ±
8	1 <sup>b</sup>	14 ± 1º	0.1d	1¢	1 <sup>b</sup>	1a
Khardungla/4	49 ±	5 + 1c	<b>2 + 1</b> h	14 ±	27 ±	27 ±
921.5	1c	$3 \pm 1^{\circ}$	$2 \pm 1^{\circ}$	1ª	1 <sup>b</sup>	1a

Values are represented as mean  $\pm$ SD (n=3), Data was analyzed by ANOVA using Duncan's multiple range test (SPSS17.0); the values with different superscript along the columns are statistically significant at P<0.005

Table	5.	Mor	phological	features	(above
groundp	arts)	of	R.webbianun	<i>i</i> growing	under
nursery	condit	tions	5		

		Mear	ı ± SD	
Collection area	No. of branches	No. of Leavs	Quadrant per (m²)	Plant height (m)
Panaliza Top/4593.8	9 ± 1.9 <sup>e</sup>	11 ±1.5 <sup>f</sup>	14 ± 1.5°	1.4 ± 0.1ª
Tangole/4266.1	$11 \pm 1^d$	14 ± 1 <sup>b</sup>	11± 1ªb	1.2 ± 0.1 <sup>e</sup>
Parkachik/3938.2	$14 \pm 1^{a}$	18 ± 1.4 <sup>a</sup>	$17 \pm 1^{b}$	1.1 ± 0.12 <sup>a</sup>
Tangsti/4725.8	9 ± 1.7°	$8 \pm 1^{e}$	$2 \pm 1^d$	1.2 ± 0.1 <sup>c</sup>
Khardungla/4921.5	6 ± 1 <sup>a</sup>	$5 \pm 1^{b}$	$3 \pm 1.4^{a}$	1.1 ± 1ª

**Table 3.** Morphological features (above ground<br/>parts) of *R.emodi* growing under nursery<br/>conditions

	mean ± SD						
Collection area	No. of branches	No. of Leavs	No. of Nodes	Quadrant per 1m	Plant height (m)		
Gulmarg	19 ± 1.9e	28.3 ±1.5 <sup>f</sup>	8.3 ± 1.5°	4± 1ª	2.1 ± 0.1 <sup>b</sup>		
Dhara	$12 \pm 1^{d}$	14 ± 1 <sup>b</sup>	$8 \pm 1^a b$	6 ± 1 <sup>e</sup>	1.4 ± 0.1ª		
Pahalgaam	$10 \pm 1^{a}$	7 ± 1.4ª	$7 \pm 1^{\rm b}$	$4 \pm 1.32^{a}$	1.6 ± 0.1ª		
Sonamarg	11 ± 1.7°	24 ± 1º	$9\pm1^{d}$	6 ± 1°	2.2 ± 0.1 <sup>b</sup>		
Yousmarg	7 ± 1ª	$9\pm1^{\mathrm{b}}$	4 ± 1.4 <sup>a</sup>	$4 \pm 1^a$	1.5 ± 0.1ª		
Dodhpathri	11 ±1¢	9 ± 2.6 <sup>b</sup>	$8 \pm 1^{ab}$	4 ± 1ª	5.6 ± 0.1e		
Tarsar Marsar	9 ±1 <sup>b</sup>	14 ± 2.5°	$12 \pm 1^{f}$	5 ± 1 <sup>b</sup>	1.6 ± 0.1ª		
Laar	11 ±1¢	15± 1 <sup>d</sup>	11 ± 1e	7 ± 1º	2 ± 1 <sup>b</sup>		

Values are represented as mean  $\pm$ SD (n=3), Data was analyzed by ANOVA using Duncan's multiple range test (SPSS17.0); the values with different superscript along the columns are statistically significant at P<0.005

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**Fig. 11.** Response of *Rheum spp*. In Net house



**Fig. 12.** Positive response of *Rheum spp.* In net house collected from different high altitudes of Kashmir Himalaya



Fig. 13. Good response of *Rheum emodi* in Net house

**Table 6.** Morphological features (underground parts) of *R.spiciforme* growing under nursery conditions

	Mean ± SD					
Sites/Altitude s (m)	Root leng th (cm)	Root thickn ess (cm)	Rhizo me length (cm)	Rhizo me breadt h (cm)	Leaf leng th (cm)	Leaf bread th (cm)
Satni mountain/39 62.4	49 ± 1 <sup>b</sup>	6 ±1 <sup>b</sup>	44± 1ª	29 ± 1ª	29 ± 1ª	31 ± 1ª
Dawar hills/4419.6	59 ± 1°	5 ± 1ª	49 ± 1ª	39 ± 1 <sup>b</sup>	44 ± 1°	54 ± 1d
Chakwali/46 84.8	64 ± 1 <sup>d</sup>	8±1°	79 ± 1 <sup>d</sup>	59 ± 1 <sup>d</sup>	54 ± 1 <sup>d</sup>	59 ± 1 <sup>bc</sup>
Habbakhatoo n mountain/47 56.8	59 ± 1°	8 ± 1°	64 ± 1°	54 ± 1 <sup>d</sup>	44 ± 1°	49 ± 1°
Dahi Nala/3810.0	39 ± 1ª	8±1°	54 ± 1 <sup>b</sup>	44 ± 1¢	44 ± 1°	50 ±
Traghal/335	49 ±	6±1b	61 ± 1¢	49 ± 1¢	34 ±	44 ±

Values are represented as mean  $\pm$ SD (n=3), Data was analyzed by ANOVA using Duncan's multiple range test (SPSS17.0); the values with different superscript along the columns are statistically significant at P<0.005

Table	7.	Morphological	features	(above
ground	part	s) of <i>R. spiciforr</i>	ne growin	g under
nursery	/ con	ditions		

	Mean ± SD				
Site	No. of branches	No. of Leavs	Quadrant per (m²)	Plant height (m)	
Satni mountain/3962.4	$10 \pm 1.9^{e}$	5 ±1.5ª	7 ± 1.5 <sup>a</sup>	1.6 ± 0.1ª	
Dawar hills/4419.6	9 ± 1 <sup>b</sup>	11 ± 1 <sup>d</sup>	9± 1°	17 ± 0.1ª	
Chakwali/4684.8	11 ± 1¢	14 ± 1.4 <sup>e</sup>	$14 \pm 1^{d}$	2.1 ± 0.1 <sup>b</sup>	
Habbakhatoon mountain/4756.8	$14 \pm 1.7^{d}$	18 ± 1 <sup>f</sup>	8 ± 1 <sup>b</sup>	2 ± 0.1 <sup>b</sup>	
Dahi Nala/3810.0	9 ± 1 <sup>b</sup>	7 ± 1º	9 ± 1.4°	1.7 ± 1ª	
Traghal/3352.8	7 ±1ª	6 ±1 <sup>b</sup>	7 ±1ª	1.8 ±0.1ª	

Values are represented as mean ±SD (n=3), Data was analyzed by ANOVA using Duncan's multiple range test (SPSS17.0); the values with different superscript along the columns are statistically significant at P<0.005

#### 3.5. Ethnobotany

*R. emodi* was collected from different locations such as Dhara, Dodhpathri, Aaroo (Pahalgaam), (Affarwat) Gulmarg, Laar, (Thajwas) Sonamarg, Yousmarg. During the surveys, it was observed that tribal people used to preserve and conserve the *R. emodi* for their living. The root is dried for so many purposes by these local people. The leaves of R. emodiare taken as vegetable.

The *R.webbianum* was collected from Ladakh Tangsti, region i.e., Ganglas (Khardungla), Parkhachik (Zanaskar), Tangole (Zanaskar), Panzila top (Zanaskar) and *R.spiciforme* was collected from Gurez area i.e., Dawar hills (Tulail), DahiNala, Satni mountain, Habbakhatoon mountain, Chakwali and Tragbal. After interviews with local people of high altitude (Fig 16, 17), it was observed that *Rheum* species are used for so many diseases, as listed in Table 8.

The information of medicinal plants for different medicinal purposes was collected by conducting frequent field surveys. In the study, area and plants were collected from diverse habitats. The surveys and collection of *Rheum* species were mainly on high altitudes and information was taken from gujjars, bakkerwals and some local people. To gain

#### knowledge and information about medicinal plants these people were interviewed during the course of the present study.

Phoum species	Diseases	Partusod	Droparation		
Kileulii species	Diseases	I al t useu	The root is powdered in the form of paste and applied on		
	wounds and boils	Roots	wounds directly without massage.		
			Leaves are boiled in water with a pinch of salt and taken a		
	Jaundice	Leaves	cup orally twice a day.		
	Diaborrea Indigestion	Roots	50g Root is powdered and boiled in 500 ml water and taken		
			a cup thrice a day.		
	Cough and asthma	Roots	Leaves are crushed and boiled in water and then taken half		
			cup orally thrice a day.		
	Headache	Leaves	Leaves are crushed and bolled and then taken one cup twice		
			The root is ground into powdered form and mixed with		
	Muscular pain /Body ache	Leaves	some oil and then applied externally to the affected parts.		
R. emodi			The root is crushed, ground, and mixed with vinegar, and		
(Pumbhaak,	Freckle/Skin	Roots	then that paste is applied externally to the affected parts.		
Pumbchaalan)			Powder of root is applied with fresh milk also.		
			Fresh leaves and fresh roots are ground and the oil-like		
	Eruptions/ Skin diseases	Roots	substance came out from fresh roots and leaves and that oil		
	Swalling and		is applied on allected parts.		
	Inflammation / Rheumatic	Leaves/	Leaves and their stalks are cooked as a vegetable		
-	pain	Roots	heaves and then starks are cooked as a vegetable		
	A	Leaves/Stalks	50 g root powder is mixed with 10g ginger powder and then		
	Food Constipation		mixed with 200ml of water and then boiled, then taken half		
			cup four times a day.		
	Reducing obesity, weight/fat	Roots	10 g root and 3 g ginger is ground paste is made. Boil the		
	8 9, 8,		mixture in one 300ml of water and then taken two spoons		
			The root nowder is made into a fine naste and used on		
	Rheumatic pain/Wound	Roots	affected parts		
Duuchhimum	D:1	I	Leaves are crushed and boiled with water and taken orally		
<i>R. Webblanum</i> (Poyandchini	Piles	Leaves	thrice a day.		
Lachoo)	Chronic bronchitis	Leaves	Leaves are boiled and then the boiled water is taken a day		
Lucitooj		Beuves	thrice.		
	Constipation	Roots	Roots are ground and fine powder is made and then mixed		
	-		with water and then the tea spoons are taken a day thrice.		
	Wound/ swelling	Roots	make a fine paste and then applied to swellings and wounds		
			The powdered form of roots are mixed with water and one		
	Bone fracture/ Backache/	<b>D</b> .	spoon of turmeric, making a fine paste and then applied to		
	Joint pain	Roots	affected parts and two spoons are taken orally our times a		
R sniciforme			day.		
(Pumbhaak.	Food	Leaves	Leaves are cooked as a vegetable.		
Jarochotal)			The root is powdered into a fine paste and used against		
	Rheumatic pain	Roots	rheumatic pain and wounds. The 3-5 g powder once in a		
			The paste is applied to the affected portion externally. Tribal		
			people chew roots for any stomach problem. The root		
	Stomachache/Intestinal	Roots	powder is mixed with warm water and taken orally half a		
	infections Dysentery		cup twice a day for irritation in the stomach, dysentery and		
			intestinal infections.		



Fig. 14. Good response of *Rheum webbianum* in Net house



**Fig. 15.** Good response of *Rheum spiciforme* in net house



**Fig. 16.** Collection of Ehtnobotasnical information bt interacting with local people (Interaction with locals in Leh(left); Interaction with locals in Dhara(right))



Fig. 18. Interaction with locals in Gurez

#### 4. Discussion

Morphological features are important in classifying the genotypes into different groups and records of these traits in different populations give an estimate of the range of these traits found in a species. Further, many of the morphological features may have a positive correlation with the synthesis and production of any phytoconstituent/phytochemical of the plant. Thus, the study of morphology becomes more important as it can indicate the best genotypes in relation to some active principles found in plants [4, 5, 10].

Therefore, in the present study, we recorded at least nine morphological features in all three species of *Rheum*. These three species of *Rheum*(*R. emodi, R. webbianum* and *R. spiciforme*) were collected i.e. from different altitudes of Kashmir Himalaya and studied by using different morphometric parameters (using standard techniques). *Rheum emodi* is a perennial plant that grows from thick and erected rhizomes and as well as from long roots[5, 7, 17].

The leaves are large, triangular and broadly ovate with fleshy petioles. The flowers are small, greenish-white on large compound leafy inflorescences. Nodes are also present. *R. webbianum* also has basal and cauline leaves. Basal leaves are orbicular or broadly ovate and are triangular in shape with long, fleshy petioles. Flowers are reddish pink in colour. Nodes are absent in this species [24-27].

*R. spiciforme* is also a stout herb. Leaves are also broadly ovate and triangular in shape. It also has thick rhizomes and long roots. The flowers are small and yellowish green. Nodes are absent in this species. These species were grown in the net house (Kashmir University) under suitable conditions as they have been listed under the endangered category and are under threat. *Rheum* species are one of the known medicinal plants. *Rheum* has vanished from lower localities and now are found only at higher altitudes [5, 7, 17].

In the net house, all the three *Rheum*species showed good response and survival rate was also good, but *R. emodi* and

*R. spiciforme* showed good survival rates as compared to *R. webbianum.* The ex-situ germplasm of *R. emodi* showed rapid and fast growth response. The germplasm of *R.webbianum* showed a slow growth response in net house. The germplasm of *R. spiciforme* also showed a fast and good growth response[7, <u>16</u>].

The *Rheum* species are used by tribal people, local people who live in hilly areas and at high altitudes. They consume the *Rheum* plants in their daily lives. They use *Rheum* leaves as vegetables and they dry the roots of *Rheum* plants for different purposes, e.g. wounds, boils, scars and digestion problems[1, 15, 36].

#### 5. Conclusion

The *Rheum* species i.e. *R. emodi, R. webbianum* and *R. spiciforme* are identified on the basis of main features i.e. *R. emodi* has nodes, whereas nodes are absent in R. *webbianum* and *R. spiciforme*. The flowers of *R. webbianum* are reddish pink, whereas the flowers of *R. emodi* are greenish white and the flowers of *R. spiciforme* are yellowish green. The *R. emodi* and R. spicifome are cliff hangers, they are mostly found in rocky areas, whereas *R. webbianum* is found mostly on plain slopes of hills and they are short herbs.

The collected germplasm was grown in the net house of Kashmir University and at least nine quantitative traits were recorded in all three species. Of the nine traits studied, *R. emodi* exhibited maximum diversity in six (rhizome breadth, leaf length, leaf breadth, no. of branches, no. of leaves, and plant height) morphological traits while *R. webbianum* showed maximum diversity in two (root length and root thickness) traits and *R. spiciforme* showed maximum diversity in one (rhizome length) morphological trait.

The diversity detected in studied traits in all these species may be attributed to the adaptive behavior of each species towards the climatic conditions prevailing in habitats of each species.

Information regarding local uses of *Rheum* species was collected by interacting with local and tribal people. Different parts of all the

*Rheum* species are being used by local people to control many diseases like piles, constipation, skin eruptions, intestinal infection,

wounds, joint pains and also used as a food

#### **Conflict of Interest**

The authors hereby declare that they have no conflict of interest.

#### Author's contributions

All authors equally participated in designing experiment analysis and interpretation of data. All authors read and approved the final manuscript.

#### **Consent for publications**

All authors have read and approved the final manuscript for publication.

#### Availability of data and material

The authors have embedded all data in the manuscript.

#### Ethics approval and consent to participate

The authors did not use human or animals in the research

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