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Morphological Diversity and Nomenclature of *Swertia chirayita* (Gentianaceae)—Recovery of Endangered Medicinal Plant Population in North Eastern Himalaya

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Abstract

Swertia chirayita **is a highly endangered and very well known medicinal plant enlisted in the IUCN list of endangered plants in the world. The plant is currently having a very low population in eastern Himalayan region due to its very low germinability in its own natural habitat. The plant population was found to have a range of diversity in morphological observations and some of the morphovariants were found to be very low among the variant populations which created a concern due to its diminishing range of genetic base in this region. A basic group of 5 morphovariants and 24 sub variants were identified and separately maintained. It is a pluriannual plant and reproductive or flowering stage was observed after completion of two years of vegetative stage from the time of seed germination. A range of morphovariant diversity was observed in the populations of Neora Valley and Lava region of eastern Himalaya in our investigation. Diversity was critically observed in some of the parameters of different morphovariants in the population. Conservation practices were generally done by not making groups or sub groups of this endangered plant while we proposed the way of conservation by making groups for proper conservation of this vulnerable plant.**

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Keywords

A Conservation Model, Endangered Plant, Morphovariants, 5 Groups, 24 Sub-Groups

1. Introduction

Swertia chirayita is one of the most important medicinal shrubs, which is enlisted as one of the important vulnerable medicinal plants found in the world and enlisted by IUCN (International Union for Conservation of Natural resources) and CITES India (2014) http://bsienvis.nic.in/Database/bsi_3949.aspx. The plant is known to have various medicinal properties against various diseases.

1.1. Taxonomic Division

Order: Gentianales; **Family:** Gentianaceae; **Tribe:** Gentianeae; **Subtribe:** Swertiinae; **Genus:** *Swertia*; **Species:** *chirayita*.

Swertia chirayita (Roxb. Ex Fleming) H. Karst is also found to be named *Swertia chirayita*, Buch-Ham.; *Ophelia chirayita* Grisebach; *Agathotes chirayita* Don; *Gentiana chirayita* Roxburgh and *G. floribunda*. According to literature, the genus *Swertia* is taxonomically given name in the honour of Dutch gardener Emanuel Sweert (1552-1612) who published an important article in the early 1600s.

It has the chemicals like xanthones, polyphenolic compounds like amarogentin (secoiridoid glycoside) [1] [2] mangiferin, swertiamarin (seco-iridoid derivatives), chiratanin, flavonoids, terpenoids, iridoids, seco-iridoid glycosides [3]. Breeding behaviour of the plant is self pollinated [4] where it was first time reported to be self along with cytological observations but other reports were found to be cross pollinated in other regions of western Himalayan region [3] [5]. The authenticity of medicinal properties of *Swertia chirayita* are found in different published reports as antidibatic [6]-[11], anti inflammatory [12], hepato-protective [2], anti leishmaniak [13], anti-cancerous [14] [15] and activity against herpes simplex virus [16].

1.2. Plant Morphology Investigations

The plant is pluriannual in nature and reported to be grown in between the range of 1200 mt - 3000 mt above sea level [4] [17] [18], and some part of the population was found in North Eastern Darjeeling Himalyan region and some other part in Kashmir to Uttarakhand and Himachal Pradesh and rest of the part in Sikkim and Himalayan region [3]. The plants were found to have very low germination of this plant in natural conditions [3] [19], but reported 70% germination under good storage in warm polyhouse condition [20]. We also found good germination in Lava in polyhouse condition (**Figure 1(a)**). We found the range of plant population from the height less than 24 cm. (less than 1.5 ft) upto more than 145 cm (more than 6 ft) (**Figures 1(b)-(h)**). It was reported to have height in between the range of 2 - 3 ft long stem and middle part is round whereas upper portion is four angled with distinct decurrent line in each angle [3] [21] which differs from our investigation. The stems are cylindrical, sometimes quadrangular and orange brown with purplish in colour and have large continuous yellow pith [3].

1.3. Morphology of the Flower Investigations

The number of sepals are four, connate at the base (receptacle), gamosepalous with valvate (no overlapping) aestivation, lanceolate, equal sized and opposite [3] [4]. The number of petals are four, gamopetalous, regular, actinomorphic (showing radial symmetry), rotate, tetramerous with valvate aestivation [3] [4]. Petal is twice the size of the calyx. Petals are greenish yellow in colour and four divisions at the base [3] [4] [17].

The number of filaments is four. Filaments are alternated with petals, each uniform in size and almost equal in size with the stigma. Each filament bears dorsifixed, versatile and bi-lobed anthers, as the anthers are dorsifixed and versatile in nature, they are free moving [4]. Gynoceium has syncarpous ovary with hypogynous configuration. Stigma is bilobed [4].

The plant is found to have reproductive phenological plasticity where it was found flowering time started from July upto September [18] [21], where, it was found to have started from September and ends up to December in this region.

Morphological diversity of *Swertia chirayita* in populations at Lava

Figure 1. (a) *Swertia chirayita* population in nursery bed in Lava (6800 ft asl); (b) Morphotype: KSC-1-Plant height: 20 cm (<2 ft)—category—Extra dwarf; (c) Morphotype: KSC-2-Plant height 48.3 cm—category—Dwarf; (d) Morphotype: KSC-3- Plant height: 66 cm—Category—Medium; (e) Morphotype: KSC-4-Plant height: 91.9 cm—category—long; (f) Morphotype: KSC-5-Plant ht: More than 144 cm—Category—Extra long; (g) *Swertia chirayita* population in polyhouse; (h) Individual plants of 1 year age at lava nursery (6800 ft asl); (i) Seeds of the plant.

As per the conservation practices for any plant, this plant could not be conserved by simple collection and maintenance in the nursery bed or in the field in its natural habitat.

Objective our study:

1) New method of conservation for conservation of all diversified plant population by a model system.
2) Plant extinction rate should be minimised in the region by maintaining groups.
3) Characterization of plants quantitative as well as qualitative characters according to NBPGR

2. Materials and Methods

2.1. Study Regions

The study regions were lava (27.086˚N and 88.66˚E) and Neora Valley region (27.06˚N and 88.70˚E) of north eastern Himalayan region in Darjeeling district of West Bengal. We have screened the whole population after careful investigation for two and half years and after investigating in Lava (6800 ft asl) and Neroa Valley region (10,600 ft asl) population, we categorize the population into five groups according to the range of plant heights. Different reports of breeding behaviour was found where cytological study (Khoosho and Tandon, 1963 [5]; Joshi and Dhawan, 2005 [3]; Chakraborty *et al.*, 2009 [4]) was also conducted, where both self pollinated and cross pollinated behaviour of the plant was found.

2.2. General Method of Conservation

Generally conservation of any plant depends mainly on [22].

1) Identification of descriptors (both morphological and reproductive) of the plant,
2) Identification of reproductive behaviour of the plant,
3) Collection of various accessions from its natural habitat,
4) In-situ or ex-situ conservation of the population in natural or introduced habitat,
5) Maintenance of the population according to standard agronomic and breeding practices,
6) Collection and harvesting of the seed or reproductive material according to its normal life cycle and maintenance of next generations.

In any conservation programme, focus is laid out in collecting large number of accessions from different parts of the world, giving of accession number, maintaining of populations of accessions (as mass collection of seeds from accessions) and creating a large base population in a suitable place. But, this procedure, if is done in *Swertia chirayita*, morphological diversity of the plant will be lost very quickly.

2.3. Non-Applicability of General Rule of Conservation

The reason we found because it was reported to have a very low rate of germination [3] [19]. In many extra dwarf and dwarf plants, the population of this category is very low due to it lower number of capsules, lower number of seeds per capsules (seed-set per plant), very small seed size (**Figure 1(i)**) and natural lower germination propensity of this plant [19]. The seeds if are collected without separating them from mass collection of the whole accession, will be lost very easily and variation this type of population observed in this region will have a very little chance to survive.

As it is an endangered plant, breeder's objective could be to conserve the population according to the accessions [22]. We propose a model by grouping of the whole population and maintenance of each individual into groups and sub groups, and not by accessions. Accession names could be given after grouping of the population. In each accession, this group and sub-group division was found to be essential for effective conservation. Classification groups or sub-groups of any endangered plant specially, is very important, and after that according to groups or sub groups, the conservation programme should be progressed, which may be ignored at first attempt in normal plant species and are not practised in endangered or vulnerable according to IUCN by conversationalists [22].

We did a model system which can be applied in any population of *Swertia chirayita* by the basic five fundamental groups according to range of heights like 1) extra dwarf, 2) dwarf 3) medium 4) long and 5) extra long and made the population into further groups ranges from type of leaf size to differential time of flowering as per the diversity of the population found in this region.

This Model has three divisions. They are mainly:

1) Morphological diversity and variability

5 types of groups were identified in the character plant height [23] and their subsequent characteristics were done. Plant height range was grouped as:

a) Ksc-1—(<24 cm)—Extra Dwarf (**Figure 1(b)**); b) Ksc-2—(25 - 64 cm)—Dwarf (**Figure 1(c)**); c) Ksc-3—(65 - 104 cm)—Medium (**Figure 1(d)**); d) Ksc-4—(105 - 144 cm)—Long (**Figure 1(e)**); e) Ksc-5—(>144 cm)—Extra long (**Figure 1(f)**), Ksc (**Kalimpong *Swertia chirayita***) (as per rules of the Directorate of Medicinal and Aromatic Plants Research). The plant populations were maintained at Algirah (5500 ft asl) and Lava (6800 ft asl) field gene banks (**Figure 1(g)**). Accessions were collected from Naora Valley (10,600 ft asl) and Lava (6800 ft asl). Individual plants were found not having effective formation of stem after one year of age (**Figure 1(h)**). The plant height was found to be ceased only after flowering stage appeared in the population [3] [4].

It was therefore identification and measurement of the plant heights were taken after appearance of flowering stage.

A model of five groups and further sub categorizations were formed and 24 groups of populations were listed (**Table 1**).

2) Variability according to onset of flowering

a) Early flowering Type—1st week of September to 1st week of October,
b) Medium flowering Type—2nd week of October to 2nd week of November,
c) Late flowering Type—3rd week of November to last week of December.

Table 1. Model for retaining morphological diversity of plant population nomenclature of *Swertia chirayita*.

	Plant type nomenclature	Variability/ morphovariants	Sub-nomenclature	Plant height (range)	Leaf size	Onset of flowering
Gr-1	KSC-1	Extra dwarf	KSC-1-1	<24 cm.	Small (length < 5 cm, breadth < 3 cm)	early
	KSC-1	Extra dwarf	KSC-1-2	<24 cm	Small (length < 5 cm, breadth < 3 cm)	medium
	KSC-1	Extra dwarf	KSC-1-3	<24 cm	Small (length < 5 cm, breadth < 3 cm)	Late
Gr-2	KSC-2	Dwarf	KSC-2-1	25 - 64	Small (length < 5 cm, breadth < 3 cm)	early
	KSC-2	Dwarf	KSC-2-2	25 - 64	Small (length < 5 cm, breadth < 3 cm)	medium
	KSC-2	Dwarf	KSC-2-3	25 - 64	Small (length < 5 cm, breadth < 3 cm)	late
	KSC-2	Dwarf	KSC-2-4	25 - 64	Medium (5 - 8 cm, breadth 3 - 5 cm)	Early
	KSC-2	Dwarf	KSC-2-5	25 - 64	Medium (5 - 8 cm, breadth 3 - 5 cm)	medium
	KSC-2	Dwarf	KSC-2-6	25 - 64	Medium (5 - 8 cm, breadth 3 - 5 cm)	Late
Gr-3	KSC-3	Medium	KSC-3-1	65 - 104	Medium (5 - 8 cm, breadth 3 - 5 cm)	early
	KSC-3	Medium	KSC-3-2	65 - 104	Medium (5 - 8 cm, breadth 3 - 5 cm)	medium
	KSC-3	Medium	KSC-3-3	65 - 104	Medium (5 - 8 cm, breadth 3 - 5 cm)	late
Gr-4	KSC-4	Tall	KSC-4-1	105 - 144	Medium (5 - 8 cm, breadth 3 - 5 cm)	early
	KSC-4	Tall	KSC-4-2	105 - 144	Medium (5 - 8 cm, breadth 3 - 5 cm)	medium
	KSC-4	Tall	KSC-4-3	105 - 144	Medium (5 - 8 cm, breadth 3 - 5 cm)	late
	KSC-4	Tall	KSC-4-4	105 - 144	Large (>8 cm and breadth > 5 cm)	early
	KSC-4	Tall	KSC-4-5	105 - 144	Large (>8 cm and breadth > 5 cm)	medium
	KSC-4	Tall	KSC-4-6	105 - 144	Large (>8 cm, breadth > 5 cm)	late
Gr-5	KSC-5	Extra tall	KSC-5-1	>144 cm	Medium (5 - 8 cm, breadth 3 - 5 cm)	early
	KSC-5	Extra tall	KSC-5-2	>144 cm	Medium (5 - 8 cm, breadth 3 - 5 cm)	medium
	KSC-5	Extra tall	KSC-5-3	>144 cm	Medium (5 - 8 cm breadth 3 - 5 cm)	Late
	KSC-5	Extra tall	KSC-5-4	>144 cm	Large (>8 cm, breadth > 5 cm)	early
	KSC-5	Extra tall	KSC-5-5	>144 cm	Large (>8 cm, breadth > 5 cm)	medium
	KSC-5	Extra tall	KSC-5-6	>144 cm	Large (>8 cm, breadth > 5 cm)	Late

3) Variability according to leaf size at maturity stage

Measurement was done and categorization of leaf was done as broad leaf, medium size leaf and small leaf [23].

	Leaf length	Leaf breadth
Broad long leaf	>8 cm	>5 cm
Medium size leaf	5 - 8 cm	3 - 5 cm
Small leaf	<5 cm	<3 cm

In overall population, classification was done as

a) Small leaf was found in Morphotype-1 population: KSC-1; b) Small and medium leaf were found in Morphotype-2 population: KSC-2; c) Medium leaf was found in Morphotype-3 population: KSC-3; d) Medium and large leaf were found in Morphotype-4 population: KSC-4; e) Medium and large leaf were found in Morphotype-5 population: KSC-5.

Results obtained after harvesting was given in **Table 2** in different genotypes.

Statistical methods: Statistical method of Randomized Block Design (RBD) at P (<0.05) was adopted to measure the best results among the five groups [24]. C.D. (critical difference) and standard error mean (SEM±) were calculated in each group. In each group, 10 plants were randomly selected and average results were calculated in the year 2010-11 and 2013-14 respectively. The maturity of *Swertia chirayita* plants in this Himalayan region takes place after two and half years (2 and 1/2 years). We collected the seeds from the matured plants which were sown in the year 2010-11, and sow the seeds in the nursery beds in the Lava (6800 ft asl). The presented data (**Table 2**) were the pooled data of the harvested plants and seeds in the year (2010-11) and (2013-14) respectively. Characterization was also done according to minimum descriptors leveled at NBPGR at different growth stages [25].

3. Results and Discussion

Characterization is also done in various medicinal plants like *Coleus froskoli* [26], *Mentha* sps. [27] and field crops like diploid wheat [28], hexaploid wheat [29] *Caramlluma* sps [30] and *Artemisia* sps. [31].

Minimal descriptors were also listed in NBPGR (National Borough of Plant Genetic Resources) in different medicinal and aromatic plants. Characterization proforma according to **NBPGR** rule is done in *Swertia chirayita* [25]. The observations were taken at the mentioned necessary steps of this plant. They are recorded as below in North Eastern Himalayan region.

Characterization according to NBPGR rule [25]

1) **Plant habit**

Biennial

2) **Mode of reproduction**

Sexual

3) **Plant growth habit** (visual scoring)

Erect

4) **Root length** (cm) at harvest stage (average of 10 random roots)—**Quantitative character**—18.51 cm.

5) **Root diameter** (cm)—fully developed root at middle of the root at harvest stage (average of 10 random roots)—**Quantitative character**—4.64 cm.

6) **Root colour**—fully developed roots at harvest stage (visual scoring)—Pale yellow.

7) **Stem length** (cm)—recorded at late vegetative/flower initiation stage (average of 10 random plants) **Quantitative character**—86.75 cm.

8) **Stem branching**—recorded at late vegetative/flower initiation stage (visual scoring)—Trichotomously branched.

9) **Stem shape**—recorded at late vegetative/flower initiation stage (visual scoring)—Cylindrical, sometimes quadrangular.

10) **Stem colour**—recorded at late vegetative/flower initiation stage (visual scoring)—Dark brown.

11) **Number of branches per plant**—recorded on fully grown plants at initiation of the flowering (average of 10 random plants)—**Quantitative character**—4.53.

Table 2. Study of morphological characters of *Swertia chirayita*.

Genotypes	Av. No. of leaves per plant	Av. No. of primary branches per plant	Av. Root length/plant (cm)	Av. Length of the inflorescence (cm)	Av. Total fresh wt. per plant (g)
KSC-1-1	13.5	1.5	4	2.4	2.38
KSC-1-2	9.8	2	5.8	3.73	3.53
KSC-1-3 Group-1	12.4	2	4.5	3.14	5.62
SEM ±	0.98	NS	0.14	NS	0.58
C.D.	2.35	0.53	0.33	0.83	1.45
KSC-2-1	21.5	2.5	8.12	2.65	11.57
KSC-2-2	18.5	1.5	7.47	3.42	14.85
KSC-2-3	16.8	2.4	10.12	3.96	10.05
KSC-2-4	26.5	1.9	9.32	4.21	9.14
KSC-2-5	17.7	2.6	10.62	3.56	11.42
KSC-2-6 Group-2	20.4	3.4	7.86	3.25	10.26
SEM±	0.54	0.69	1.04	0.36	1.36
C.D	1.23	1.07	2.32	1.36	3.02
KSC-3-1	27.2	1.5	12.8	6.3	15.72
KSC-3-2	18.4	1.8	14.1	7.5	20.83
KSC-3-3 Group-3	22.4	1.6	12.5	8.5	23.84
SEM±	0.78	0.87	1.11	0.44	2.33
C.D.	2.36	NS	3.02	1.32	5.98
KSC-4-1	29.6	2.5	14.3	8.6	26.93
KSC-4-2	39.7	2.3	13.3	8.3	22.56
KSC-4-3	41.2	2.5	13.2	9.1	25.62
KSC-4-4	30.6	2.3	14.8	6.2	22.42
KSC-4-5	38.5	3.2	12.2	7.3	26.24
KSC-4-6 Group-4	27.6	2.8	13.8	6.8	22.72
SEM±	1.69	0.33	1.13	1.48	1.52
C.D.	5.98	0.98	2.69	2.78	3.79
KSC-5-1	49.8	2.8	15.2	8.9	25.73
KSC-5-2	59.7	3.3	12.3	6.1	28.53
KSC-5-3	61.2	2.9	10.2	8.1	31.31
KSC-5-4	75.4	3.2	9.8	7.6	26.32
KSC-5-5	63.8	4.3	13.9	7.1	28.91
KSC-5-6 Group-5	55.4	3.5	10.2	6.9	27.62
SEM±	1.05	0.22	0.78	0.88	1.98
C.D.	3.69	0.56	2.88	2.19	4.66

Continued

Genotypes	Av total dry wt per plant (g)	Av. Number of capsules per plant	Av. Number of seeds per capsule	Av. Shoot wt per plant (g)	Av root wt per plant (g)	Av. Seed yield per plant (g)
KSC-1-1	0.98	24.35	90.64	3.01	0.48	0.52
KSC-1-2	1.45	31.42	68.32	2.92	0.84	0.49
KSC-1-3	1.30	27.46	56.14	3.05	1.02	0.95
SEM±	0.14	0.49	8.46	0.22	0.13	0.09
C.D.	0.33	1.37	23.23	NS	0.51	0.27
KSC-2-1	3.51	109.64	137.5	6.23	2.06	0.93
KSC-2-2	3.69	107.52	125.5	11.74	2.12	1.23
KSC-2-3	6.77	118.58	128.5	7.88	2.73	0.72
KSC-2-4	4.13	102.34	122.8	10.68	2.56	1.21
KSC-2-5	5.32	120.41	97.2	9.52	3.65	1.35
KSC-2-6	4.57	125.30	119.6	8.56	3.10	0.98
SEM±	0.61	7.01	4.01	1.09	0.22	0.23
C.D.	1.84	22.36	13.65	3.54	0.74	0.56
KSC-3-1	6.79	167.52	104.2	13.38	3.15	0.92
KSC-3-2	6.12	185.73	109.6	9.65	2.59	1.23
KSC-3-3	7.31	166.36	120.4	11.85	2.71	1.32
SEM±	0.85	9.01	7.12	1.21	0.18	0.24
C.D.	2.54	27.08	20.31	4.02	1.56	0.71
KSC-4-1	9.68	172.4	112.4	15.85	4.74	3.23
KSC-4-2	7.67	186.3	98.22	17.31	4.28	4.20
KSC-4-3	6.32	173.7	127.32	14.42	4.51	4.76
KSC-4-4	7.65	188.9	114.21	19.38	3.72	3.92
KSC-4-5	9.45	155.8	91.65	18.65	5.59	4.86
KSC-4-6	9.92	172.8	130.42	21.85	4.78	3.72
SEM±	0.98	7.36	6.32	1.32	0.39	0.33
C.D.	2.14	22.36	20.33	4.35	1.23	1.02
KSC-5-1	10.18	179.82	93.41	20.82	5.74	6.16
KSC-5-2	8.92	226.34	112.62	26.65	6.21	7.28
KSC-5-3	9.38	193.66	117.13	24.42	6.57	7.79
KSC-5-4	8.67	187.42	124.61	21.35	6.82	6.97
KSC-5-5	6.78	165.78	98.63	24.72	6.29	5.73
KSC-5-6	7.43	158.93	120.32	26.84	6.98	6.82
SEM±	0.44	5.36	4.98	1.69	0.22	0.29
C.D.	1.76	18.65	17.66	4.65	0.63	0.98

12) **Leaf length** (cm)—recorded on well developed leaves at full foliage stage (average of 10 random leaves)—**Quantitative character**—5.63 cm.

13) **Leaf width** (cm)—recorded on well developed leaves at full foliage stage (average of 10 random leaves)—**Quantitative character**—4.47 cm.

14) **Leaf shape**—recorded at full foliage stage (visual scoring)—Lanceolate.

15) **Leaf base**—recorded at full foliage stage (visual scoring)—Acute.

16) **Leaf attachment**—recorded at full foliage stage (visual scoring)—Sessile.

17) **Leaf colour**—recorded on 3rd leaf from the base at full foliage stage (visual scoring)—Green, purple green.

18) **Number of veins on lamina**—recorded at full foliage stage on well developed leaves (average of 10 random leaves)—5 numbers—**Quantitative character**.

19) **Number of leaves per plant**—recorded at full foliage stage (average of 10 random plants)—**Quantitative character**—59.58.

20) **Days to flower initiation**—recorded as number of days from seed sowing/transplanting to the flower initiation stage—**Quantitative character**—925 days (more than 2 and ½ years).

21) **Type of inflorescence**—recorded at full bloom stage (visual scoring)—Leafy panicles.

22) **Number of inflorescences per plant**—recorded at full bloom stage (average of 10 random plants)—**Quantitative character**—143.65.

23) **Number of flowers per inflorescence**—recorded at full bloom stage (average of 10 random inflorescences)—**Quantitative character**—14.32.

24) **Flower colour**—recorded at full bloom stage (visual scoring)—Greenish-yellow with purplish tinge.

25) **Plant height** (cm)—measured from ground level to the tip of inflorescence (average of 10 random plants)—**Quantitative character**—87.34 cm.

26) **Fresh herbage yield per plant**—recorded on dry weight basis when capsules are fully formed (average of 10 random plants)—**Quantitative character**—27.54 gm.

27) **Dry herbage yield per plant** (g)—recorded on dry weight basis when capsules are fully formed and plants are shade dried (average of 10 random plants)—**Quantitative character**—12.43 gm.

28) **Days to capsule formation**—recorded as number of days from planting to the day when at least 75% of the capsules have formed—**Quantitative character**—915.6 days.

29) **Number of capsules per plant**—counted when all capsules have formed on a plant (average of 10 random plants)—**Quantitative character**—179.82.

30) **Days to capsule maturity**—recorded as number of days from sowing/planting to the day when at least 75% of capsules have matured—**Quantitative character**—930 days.

31) **Capsule diameter** (cm)—recorded on matured capsules (average of 10 random capsules)—**Quantitative character**—0.45 cm.

32) **Capsule shape**—recorded on matured capsules (visual scoring)—Ovoid.

33) **Number of seeds per capsule**

To be recorded when seeds are matured and dried (average of 10 random capsules)—**Quantitative character**—103.48.

34) **Seed surface**—recorded on matured and dried seeds (visual scoring)—Smooth.

35) **1000 seed weight** (g)—recorded on completely dried seeds after harvest—**Quantitative character**—26.14 mg.

3.1. Average Number of Leaves per Plant

Group 1: In case of Gr-1, KSC-1-2 was found to be statistically at par with the lowest value of KSC-1-3 at ($P < 0.5$), highest value was found in KSC-1-1 (**Table 2**).

Group 2: All the types are statistically at par with the highest and lowest value of the morphs (<C.D.1.23). That is, plants of height within the range of 25 - 64 cm. of all six types are statistically insignificant at ($P < 0.05$) (**Table 2**).

Group 3: KSC-3-3 was found to have highest number of leaves and KSC-3-2 was found to have lowest number of leaves. KSC-3-2 morph was statistically significant with highest and lowest value at ($P < 0.05$) (**Table 2**).

Group 4: KSC-4-2 was found showing highest number of leaves whereas lowest number of leaves was found in KSC-4-1. KSC 4-3 and KSC-4-6 are statistically at par with KSC-4-1 at ($P < 0.05$). All other morphs were found statistically at par with each other at ($P < 0.05$) (**Table 2**).

Group 5: KSC-5-2 was found to have highest number of leaver per plant. Lowest number of leaves was found KSC-5-5 at ($P < 0.05$) (**Table 2**). These variations were also found in morphological characters of *Mentha* sps. [27].

3.2. Average Number of Primary Branches per Plant

Group 1: In KSC-1-2 and KSC-1-3 were found to have more number of primary branches per plant. All the KSC types were found to have statistically at par at ($P < 0.05$) values regarding number of primary branches per plant (**Table 2**).

Group 2: KSC-2-6 was found to have highest number of primary branches and KSC-2-2 and KSC-2-4 was found to be statistically at par with KSC 2-6 at ($P < 0.05$). Lowest number of primary branches was found in KSC-2-4 (**Table 2**).

Group 3: All the three types were found to have non-significant values and they were all statistically at par with each other at ($P < 0.05$) (**Table 2**).

Group 4: All the six types were found to have non-significant values and they were all found to be statistically at par with each other at ($P < 0.05$) (**Table 2**).

Group 5: All the six types were found to be statistically significant and they were significantly different with the KSC-5-5 type that was found to have highest number of primary branches at ($P < 0.05$) (**Table 2**).

3.3. Average Root Length per Plant

Group 1: All the three types were found to have statistically significant values and they are significantly different from the highest value found in KSC-1-2 type at ($P < 0.05$) (**Table 2**).

Group 2: In all six types, KSC-2-2 and KSC-2-6 were found to be statistically significant with the KSC-2-5 type which was found to have root having highest number of root length at ($P < 0.05$) (**Table 2**).

Group 3: All the three types were found to be statistically significant with each other (**Table 2**).

Group 4: All the six types were found to be statistically at par with each other at ($P < 0.05$) (**Table 2**).

Group 5: The types KSC-5-2, KSC-5-3, KSC-5-4 and KSC-5-6 were found to be statistically significant with KSC-5-1 at ($P < 0.05$) which was found to have highest root length in all the six types (**Table 2**). Similar variations of root length were also found in *Coleus forskohlii* [26].

3.4. Average Length of the Inflorescence

Group 1: In case of average length of inflorescence, only KSC 1-1 was found to have significantly lesser than the value of KSC-1-2 which was found to have highest length of inflorescence. All other two types were found to have statistically at par values than KSC-1-2 at ($P < 0.05$) (**Table 2**).

Group 2: In this group, all the six types were found to be statistically at par with the highest length of inflorescence found in KSC-2-4 except the type KSC-2-1 which was found to have statistically significantly lower in length with KSC-2-4 at ($P < 0.05$) (**Table 2**).

Group 3: In this group of three types, KSC-3-3 type was found to have highest length of inflorescence. KSC-3-1 type was found to be statistically significantly lower in length with KSC-3-3 type and KSC-3-2 type was found to be statistically at par with KSC-3-3 type at ($P < 0.05$) (**Table 2**).

Group 4: In the six types, KSC 4-3 was found to have highest length of inflorescence and all the types except KSC-4-4 were found to be statistically at par with KSC-4-4 type. KSC-4-4 type was found to be statistically significantly lower in length with KSC-4-4 type at ($P < 0.05$) (**Table 2**).

Group 5: In the six types, KSC-5-1 was found to have highest length of inflorescence and all the types were found to be statistically at par with KSC-5-1 except KSC-5-2 which was statistically significantly lower in length than KSC-5-1 at ($P < 0.05$) (**Table 2**). [27] also did the groups and sub groups and found the variations in *Mentha* sps.

3.5. Average Total Fresh wt per Plant

Group 1: In this group, all the three types were found to be statistically significant with the highest total fresh wt found in KSC-1-3 at ($P < 0.05$) (**Table 2**).

Group 2: In this group, all the six types were found KSC-2-1 and KSC-2-5 were found to be statistically significantly lower with the highest fresh wt found in KSC 2-2 at ($P < 0.05$). All other types were found to be statistically at par with KSC-2-2 (**Table 2**).

Group 3: In this group of three types, KSC-3-1 was found to be statistically significantly **lower** than highest fresh wt found in KSC-3-3 at ($P < 0.05$). The other type KSC-3-2 was found to be statistically at par with KSC-3-3 (**Table 2**).

Group 4: In all the six types, KSC-4-2, KSC-4-4 and KSC-4-6 were found to be statistically significantly lower in fresh wt than KSC-4-1 at ($P < 0.05$) (**Table 2**).

Group 5: In all the six types, KSC-5-1 and KSC-5-4 were found to be significantly lower at ($P < 0.05$) than the highest total fresh wt found in KSC-5-3 v. KSC-5-5 and KSC-5-6 were found to be significantly at par with KSC-5-3. KSC-5-2 was found to be at par with KSC-5-3 (**Table 2**). Variation in fresh wt was also found in medicinal plants like *Mentha* sps [27], *Artemisia* [31] and wheat [32].

3.6. Average Total Dry wt per Plant

Group 1: In case of dry wt per plant in this group, KSC-1-1 was found to have significantly lower in dry wt than highest dry wt found in KSC-1-3 at ($P < 0.05$). KSC-1-2 was found to be significantly at par with KSC-1-3 at ($P < 0.05$) (**Table 2**).

Group 2: In this group, in the six types, all the morphotypes were found to be statistically significantly lower at ($P< 0.05$) in dry wt than KSC-2-3 which was found to have highest dry wt per plant (**Table 2**).

Group 3: In this group, all the three types were found to be statistically at par with each other and also with KSC-3-3 at ($P < 0.05$) which was found to have highest dry wt among here types (**Table 2**).

Group 4: In this group, KSC-4-1 and KSC-4-5 was found to have statistically at par with the highest value found in KSC-4-6, all other types were to significantly lower in dry wt than KSC-4-6 at ($P < 0.05$) (**Table 2**).

Group 5: In this group, Only KSC-5-3 was found to be statistically at par with KSC-5-1 which was found to have highest dry wt. All other types were found to have statistically significantly lower in fresh wt than KSC-5-1 at ($P < 0.05$) (**Table 2**). Dry wt. was also found variable while doing variability studies in wheat [27] and *Artemisia* [31].

3.7. Average Number of Capsules per Plant

Group 1: In this group, all the three morphotypes were found to be significantly different from each other and the KSC-1-1 and KSC-1-3 were found to significantly different from KSC-1-2 at ($P < 0.05$) which was found to have highest number of average number of capsules per plant (**Table 2**).

Group 2: In this group, except KSC-2-4 was found to have significantly lower at ($P < 0.05$) average number of capsules than the highest number of capsules found in KSC-2-6 (**Table 2**).

Group 3: In this group, all the three types were found to be significantly at par at ($P < 0.05$) with each other as well as the highest number of capsules found in KSC-3-2 (**Table 2**).

Group 4: In this group, KSC-4-1, KSC-4-2, KSC-3-3 and KSC 4-6 were found to have statistically at par at ($P < 0.05$) results of average number of capsules per plant when compared with KSC-4-4. Only KSC-4-5 was found to significantly lower than KSC-4-4 (**Table 2**).

Group 5: In this group, all the five types were found to be significant at ($P < 0.05$) with the highest value found in KSC-5-2 (**Table 2**). Similar observations were found in the plant population of wheat [29].

3.8. Average Number of Seeds per Capsule

Group 1: In this group KSC-1-3 was found to be significantly lower than the highest number of seeds found in KSC-1-1. KSC-1-2 was found to be statistically at par at ($P < 0.05$) with KSC-1-3 (**Table 2**).

Group 2: In this group, KSC-2-4, 2-5 and KSC-2-6 were found to be significantly lower in value at ($P < 0.05$) of average number of seeds when compared with KSC-2-1. KSC-2-2 and KSC-2-3 were found to be statistically at par with KSC-2-1 which was found to have highest number average number of seeds per capsule (**Table 2**).

Group 3: In this group, all the three types were found to be statistically at par with each other and KSC-3-1 and KSC-3-2 were found to be statistically at par with KSC-3-1 at ($P < 0.05$) which was found to have highest value of number seeds per capsule (**Table 2**).

Group 4: In this group, KSC-4-3 was found to be statistically at par with KSC-4-6 at ($P < 0.05$) which was sound to have highest number of seeds per capsule. Other types were found to be statistically significantly different from KSC-4-6 (**Table 2**).

Group 5: In this group, KSC-5-1 and KSC-5-5 were found to be statistically lower ($P < 0.05$) than the highest number of seeds per capsule found in KSC-5-4. All the other three types KSC-5-2, KSC-5-3 and KSC-5-6 were found to be statistically at par with KSC-5-4 at ($P < 0.05$) (**Table 2**). Similar observations were found in wheat by [32] and [28].

3.9. Average Shoot wt per Plant

Group 1: In this group, all the types were non-significant with each other, so they are not at par in between them (**Table 2**).

Group 2: In this group, KSC-2-4 was found to statistically significantly lower than KSC-2-2 at ($P < 0.05$) which was found to have highest shoot wt per plant. All other types were found to be statistically at par with KSC-2-2 (**Table 2**).

Group 3: In this group, all the three types were found to be statistically at par with KSC-3-1 which was found to have maximum shoot wt per plant at ($P < 0.05$) (**Table 2**).

Group 4: In this group, KSC-4-1, KSC-4-2 and KSC-4-3 were found to be statistically significantly lower in shoot wt than KSC-4-6 which was found to have highest shoot wt per plant. KSC-4-4, KSC-4-5 were found to have statistically at par with KSC-4-6 at ($P < 0.05$) (**Table 2**).

Group 5: KSC-5-1 and KSC-5-4 were found to be statistically significantly lower in shoot wt than KSC-5-6. All the types like KSC-5-2, KSC-5-3 and KSC-5-5 were found to be statistically at par with KSC-5-6 at ($P < 0.05$) (**Table 2**). This type of variability is also found in wheat [32] in case of characterization.

3.10. Average Root wt per Plant

Group 1: In this group, KSC-1-1 was found to be statistically significantly lower in root wt than KSC-1-3 which was found to be highest in root wt per plant. KSC-1-2 was found to be at par with KSC-1-3 (**Table 2**).

Group 2: In this group, KSC-2-1, KSC-2-2, KSC-2-3 and KSC-2-4 was found to be statistically significantly lower in root wt than KSC-2-5 which was found to have highest average root wt per plant. KSC-2-6 was found to be statistically at par with KSC-2-5 at ($P < 0.05$) (**Table 2**).

Group 3: In this group, all the three types were found to be statistically at par with the KSC 3-1 at ($P < 0.05$) which was found to have highest root wt per plant (**Table 2**).

Group 4: In this group, KSC-4-2 and KSC-4-4 were found to be statistically significantly lower in average root wt per plant than KSC-4-5 at ($P < 0.05$) which was found to have more root wt than any of five types. Rest of the types were found statistically at par with KSC-4-5 (**Table 2**).

Group 5: In this group, KSC-5-1, KSC-5-2 and KSC-5-5 were found to be statistically lower in root wt than KSC-5-6 at ($P < 0.05$) which was found to have highest root wt per plant. Rest of the types were found to be statistically at par with KSC-5-6 (**Table 2**). This type variability is also found in wheat [32] while characterization is done.

3.11. Average Seed Yield per Plant

Group 1: In this group, KSC-1-1 and KSC-1-2 were found to statistically significantly lower in seed yield per plant than KSC-1-3 at ($P < 0.05$) which was found to have highest seed yield per plant (**Table 2**).

Group 2: In this group, KS 2-1, KSC-2-2, KSC-2-4 and KSC-2-6 were found to be statistically at par with KSC-2-5 at ($P < 0.05$). Only KSC-2-3 was found to be statistically significant with KSC 2-5 (**Table 2**).

Group 3: In this group, all the types were found to be statistically at par with each other and also with KSC-3-3 at ($P < 0.05$) which was found to have highest seed yield per plant (**Table 2**).

Group 4: In this group, KSC-1-1 and KSC-1-6 were found to have statistically significantly lower than KSC 4-5 at ($P < 0.05$) which was found to have highest seed yield per plant. Rest of the types were found to be statistically at par with KSC-4-5 (**Table 2**).

Group 5: In this group, KSC-5-1 and KSC-5-5 were found to be significantly lower than KSC-5-3 which was found to have highest number of seeds per plant at ($P < 0.05$). Rest of the plants were found to have statistically at par values with KSC-5-3 (**Table 2**). Similar observations were found in wheat [32] and *Mentha* sps. [27].

From the overall characterization of the plant group [33] [34], different types were found to have statistically lower performance in character than the highest values recorded in each group (**Table 2**). Similar morphological characterization was also previously done in medicinal plants like *Mentha* sps. [27], *Coleus* sps [26] *Artimicia* sps [31] and also in field crops like wheat along with molecular markers to find out the extent of variability present in the population.

4. Conclusion

The morphological diversity in the population of *Swertia chirayita* was found in north eastern population where we did the investigation. A universal model of group and sub-groups is very much essential for effective conservation of this population. *Swertia chirayita* is a very vulnerable plant and cannot survive under the elevation of 5800 ft (asl). Even in the existing population, it is decreasing at an alarming rate due to its very low germination rate and increasing infestations by pathogens. This is the first attempt of making a model system of grouping of population for effective conservation of this plant in any natural or artificial conservation programme in its natural or introduced habitat. We also found minimum descriptors level according to the defined characters mentioned in NBPGR, we further propose categorization and sub-categorization in the above mentioned rule of grouping which is the only way for effective conservation of this plant. If it is not done, population will be completely lost after a few years and the plant will be extinct in near future due to loss of variability and genetic base especially in the extra dwarf and dwarf morphological variants.

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Author Contribution Statement: Contribution of Co-Authors

DM and SB worked with me as a group for the whole work of investigations where DM was assigned to look after the agronomical work for proper nutrient management of this endangered plant population both in seedling and transplanting stages and SB was assigned to look after the survival and disease management practices for this highly vulnerable plant. They also helped to collect the data at appropriate time of observation and collection and we approached the investigations in the locations of hills dividing our nature of work in that manner. DM also helped to do statistical works for valuable inference of the investigation.

Conflicts of Interest

The authors declare that they do not have any competing interests.

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