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Floral Biology of *Strobilanthes Callosus* (Nees) Bremek.: An Underutilized Plant Species of The Western Ghats

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Abstract

Strobilanthes callosus, a member of the Acanthaceae family, is native to the open hill slopes and valleys of the Western Ghats, particularly in the Nasik region and other areas of Maharashtra and Peninsular India. While the genus Strobilanthes is found globally, S. callosus is primarily confined to specific hilly locations, notably at Anjaneri within the Trimbakeshwar tehsil of Nasik district. This species is known for its extraordinary mass flowering event, which occurs once every eight years. The earlier studies can collectively enhance understanding of Strobilanthes callosus; particularly in terms of its ethno-medicinal potential and phytochemical properties. However, specific research focusing solely on its floral biology remained limited, indicating a valuable opportunity for further investigation in this area. The current study has uncovered few fascinating aspects of the floral morphology, phenology, anther dehiscence, pollen viability including germination under controlled conditions, fruit, Seed characteristics, floral visitors of S. callosus etc. The Pollen viability was observed at 80%; however, a significant number of these pollen grains did not germinate under laboratory conditions due to the absence of the necessary microclimate and incubation period. Also, fieldwork highlighted few anthropogenic threats including grazing, tourism, and wildfire exposure which emphasized the need for further developmental biological studies to ensure its conservation in vivo.

Keywords: *Strobilanthes callosus, floral biology, Western Ghats.*

INTRODUCTION

Plant biologists have long been fascinated by the diversity in floral morphology among plant species, prompting the creation of a number of explanatory ideas. [46]. The genus name originates from the Greek 'Strobilos' (cone) and 'anthos' (flower), reflecting the floral architecture. *Strobilanthes* is the second largest genus in Acanthaceae, comprising over 350-454 accepted species worldwide [26], with around 150 species recorded in India [43], primarily in the Western Ghats and Nilgiri hills [4].

One of its most fascinating members is *Strobilanthes callosus* Nees. (syn. *S. callosa*), a shrub endemic to the open hill slopes and valleys of Maharashtra and parts of peninsular India [24]. Documented from districts including Ahmednagar, Satara, Ratnagiri, Pune, and Nasik [42], the species is locally known as 'Karvi'. It undergoes gregarious blooming every 7-10 years during the monsoon months (June-September), a phenomenon that blankets entire hillsides in hues of violet and white [3]. Previous studies have documented occurrence of *S. callosus* in several locations within the Western Ghat regions of Maharashtra such as Ahmednagar, Kolhapur, Pune, Raigad, Ratnagiri, Satara, Sindhudurg, and Thane. [42] [Fig.1A].

Ecological significance:

The mass flowering of *S. callosus* is a strategy that attracts a wide spectrum of pollinators- bees, butterflies, birds, enhancing the cross-pollination and reproductive success [15,16]. The blooms are playing crucial ecological roles such as enriching biodiversity through pollinator support, contributing to the forest floor nutrient cycle through post-bloom decomposition, aiding in soil stabilization and groundwater retention in semi-evergreen and deciduous ecosystems [39]. The life cycle and bloom rhythm of *S. callosus* not only mark a striking natural spectacle, but also underpin essential ecological processes in the Western Ghats. [20, 30].

Ethno medicinal role:

The genus *Strobilanthes* contains more than 209 bioactive compounds, including phenolics, flavonoids, and terpenoids, many with antibacterial and anti-inflammatory properties [7,47,48]. Traditionally, *S. callosus* has been used for treating inflammation and arthritis [19,37]. The stems are employed for hut construction and fencing by tribal communities, and the mass blooming supports the production of 'Karvi honey', highly valued for its unique taste and medicinal value [6,28]. In some parts of India; the roots are used as medicine and the locals employ this plant for their habitat, constructing walls for roofing, cow enclosures etc. [5]. GIS-based studies have focused on other species such as *S. kunthiana* and *S. blume*, highlighting the research gap for *S. callosus* [17].

Despite its ecological and cultural importance, detailed floristic and reproductive studies on *S. callosus*, particularly in Maharashtra, remain scarce. While prior studies have addressed broader ecological roles and pharmacology of *Strobilanthes* species, particularly in terms of a few species like *S. kunthiana, S. blume, S. ixiocephala;* there is a need to dearth

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systematic data on the floral biology of *S. callosus* including morphometrics of flowers, pollen and fruit characteristics, and reproductive efficiency of this underutilized plant species.

MATERIALS & METHODS

Study Area

Strobilanthes callosus is predominantly distributed across the Western Ghats of India, specifically in the states of Maharashtra, Karnataka, and Tamil Nadu. The present study was conducted in selected habitats of *S. callosus* within the Nashik district of Maharashtra, primarily at the Trimbakeshwar and Anjaneri hills, which are known for their rich floristic diversity and periodic blooming of *Strobilanthes callosus* [Fig. 1 A-B].

Field Sampling

Field investigations were carried out during the flowering season from August to December 2024. Plant specimens were collected and identified using standard regional floras. [8,11.] Weekly field visits were undertaken to monitor flowering phenology, which included the onset of leaf fall, bud initiation, anthesis, end of flowering, fruit development, and seed dispersal stages.

At each study site, 25 flowers from 10 randomly selected inflorescences were studied. Observations were first made in the field using a 60× handheld lens, and flowers were subsequently preserved in 70% ethanol for detailed laboratory analysis. Floral visitors were documented through direct observation during daylight hours, particularly between 07:00 and 11:00 hours, when pollinator activity was highest. Visitors were photographed and, when necessary, captured using insect nets for identification with the assistance of entomological keys and expert consultation.

Laboratory Analysis

Morphometric analyses of floral parts including bracts, petals, stamens, styles, stigmas, ovaries, fruits, and seeds were conducted using digital calipers and a stereo microscope. Phenological stages such as leaf senescence, bud initiation, anthesis, and fruiting were recorded systematically. Anther dehiscence was assessed by examining unopened flowers under a dissecting microscope to determine the proportion of dehisced anthers at anthesis.

Pollen grains were collected from freshly dehisced anthers and analyzed for size, shape, and aperture characteristics. Pollen viability was evaluated using the acetocarmine staining method at room temperature, following protocols [34]. *In vitro* pollen germination tests were performed employing both hanging drop and sitting drop methods, as described by [32,33]. Germination success and viability were documented microscopically and photographed for reference [Fig. 5 c-e].

Data Analysis

Observed data was analyzed using relevant statistical tools to ensure accuracy and reliability. The blooming and anther dehiscence data were analyzed using one-way ANOVA that revealed significant differences in the number of flowers opened and percentage of anther dehiscence across different time intervals. [Table 1]. Similarly, for morphological parameters and pollen viability data [Table 2 and Table 4], One-Way ANOVA was applied to compare plant height, internodes length, and leaf dimensions and assess pollen viability data across the sample plants.

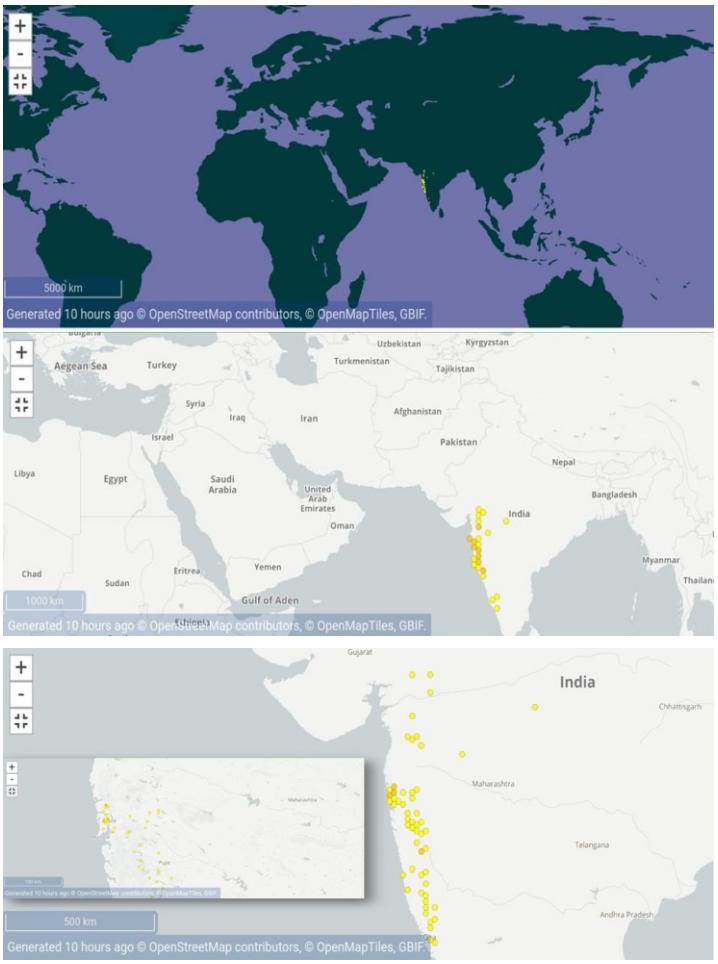

Fig.1A. Distribution of S. callosus Wall. ex Nees Bremek within India. (Source: GBIF-Global Biodiversity Information Facility)

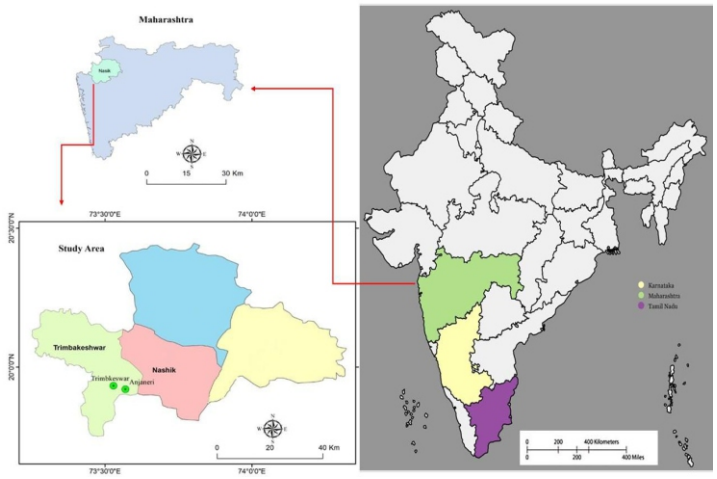

Fig.1B. Location of Study area showing general distribution of S. callosus within India

RESULTS

Strobilanthes callosus is a glabrate shrub with stiff, rough, and warted stems that grows 2-5 meters high. The leaves are 10-22 X 3-7.5, with one pair frequently smaller than the other. They are elliptic-lanceolate, acute or acuminate, with crenate and ciliate margins. [Table 2].

Phenological Observations

The flowering phenology of *Strobilanthes callosus* was monitored from August to December 2024. Leaf senescence began in mid-June, followed by new leaf initiation in early July. Bud emergence occurred during the last week of July, with full flowering observed between mid-August and late September. Anthesis occurred between 7:00 a.m. and 9:30 a.m., while

anther dehiscence was recorded within one hour of anthesis. Flowering ceased by mid-October, and fruiting was completed by the end of December. Fruiting starts in late December.

Floral Morphology

In *Strobilanthus callosus* displays zygomorphic, violet-blue tubular flowers arranged in terminal inflorescences [Fig.2]. The ovary is bicarpellary and superior with axile placentation. The stigma is bifid and exserted, adapted for pollinator contact. [Table 3 A-B; Fig. 4].

Anther dehiscence and Pollen viability

Among 60 anthers examined, 92% exhibited dehiscence at the time of Anthesis. Pollen grains were tricolpate, spherical, exine tectate, perforate with granular ornamentation, intine thin and smooth. [Fig.5-c, d], with a mean diameter of 42.3 ± 1.6 µm. Pollen viability tests using 1% Acetocarmine revealed 80.4% viable pollen, though *in vitro*. (Table 4; Fig. 5-e). Pollen germination success was found low (~18.5%) under lab conditions, possibly due to inadequate humidity or nutrient factors [32-35].

Fruit and Seed Characteristics

Each fruit is a Capsule, seeds are thin, obovate, acute, densely shaggy with white inelastic hair, developed by late October, containing 6-8 ellipsoid seeds. The seeds were blackish-brown, ~3.1 ± 0.2 mm long, [Fig. 5-a] with a rough surface texture aiding in water absorption and dispersal. [42].

Pollinator Observations

Bees were the most frequent visitors, contributing significantly to pollen transfer. Occasional visits by butterflies were also recorded. Floral visitors included *Apis dorsata, Apis cerana, Xylocopa spp.* were observed between 7:30 a.m. and 11:00 a.m. [Fig. 3].

DISCUSSION

The present work revealed few intriguing floral characteristics of *Strobilanthes callosus* which can be comparable to other species in the genus *Strobilanthes* such as *S. glandulata* from Sri Lanka [25,27], *S. brittoi* from Kerala [21], *S. warreensis* from Goa [23], *S. kunthiana* from south India [6].

The research on cyclical blooming patterns in *Strobilanthes* species has garnered a lot of attention of many workers. This phenomenon might represent a survival adaption to synchronize pollination and minimize herbivory, which gives it a tactical edge in increasing seed production by mass flowering. The fact that *S. callosus* blooms every eight years, could be due to changes in microclimate. However, like other species of *Strobilanthes;* the mechanism underlying this remarkable coordinated blooming occurrence is still unclear. Future scope lies in the research of this unique phenomenon. [6]. However, this study has confirmed a well-documented phenomena of *S. callosus*; i.e. synchronized blooming and mass flowering. Moreover, it can be concluded that the corolla morphology reflects an adaptation to various pollinators, most prominently bees. [22].

The life span of *S. callosus* is about seven to eight years. In the last year; all these plants die and at onset of monsoon, new plants are formed from seeds. Being monocarpic, *S. callosus* reproduces once after flowering and then dies. Blooming begins and lasts for around two to three months. After abundant blossoming, the capsules develop and dehisce in one or two months before withering and dying [10, 38].

During the summer, the pattern of drying up of whole plants while roots remaining alive in the ground, which is consistent with such earlier reports. [9,44]. Seed formation was observed in the same year in which the inflorescence, flowers, and vegetative development fade together. [2] [Fig.6].

Pollen grain studies conducted on *S. callosus* can aid in better understanding of the relationships within and between the taxa, as well as the phylogeny of the Acanthaceae family. [18,29]. Also, the Pollen morphology is known to have considerable taxonomic importance, and a basis of angiosperm phylogeny therefore the broad range of pollen morphological characters in terms of size, shape and texture as observed in *S. callosus* during the present study, which might be a relevant trait for delimiting species in this group. [25,45]. Moreover, pollen morphological characteristics of *S. callosus* found to be diverse, as in case of most Acanthaceae members and in accordance with the earlier such studies [12,36,40,41]. During the studies on pollen grains, lack of earlier reports on the pollen viability of *S. callosus* was observed. It was determined by using Acetocarmine stain (1%). It revealed that most of the viable pollen grains, despite of 80% pollen viability recorded, have failed to germinate after a 48-hour of incubation period. This might be due to several factors like specific laboratory *in vitro* conditions, suitability of nutrient media and pH levels in the medium. [14]. Additionally, temperature and humidity during incubation could have played a critical role. The intrinsic characteristics of the pollen grains could have contributed to such observations. Overall, these findings suggest that both environmental conditions and inherent pollen properties can influence pollen grain germination, highlighting the need for further research to optimize these factors. Also, the local differences in the abiotic environment can favour various floral morphologies in different places. According to few researchers, floral morphology can be described by the identity of floral visitors, with environment having only an indirect influence. However, in highly specialized interactions, biotic and abiotic factors may account for a significant amount of intraspecific variation in floral morphology [46].

The field observations, during early morning and throughout the daytime, indicated that the bees were dominant pollinators of *S. callosus* [31]. The preliminary findings regarding floral visitors of *S. callosus* have suggested the floral larceny by bees mostly in full bloom flowers and rarely in bud stage [1,13]. The occurrence of honey bees like *Apis* spp. is consistent with the other such studies which recorded it as major pollinator of this plant [31].

CONCLUSION

Floral biological studies on *S. callosus* demonstrates a remarkable combination of highly sophisticated strategies of reproduction, ecological niches, and medicinal value. Its Characteristic feature of synchronized mass-flowering behavior as studied in other species like *S. kunthiana* highlights evolutionary adaptations aimed at enhancing reproductive success through plant-pollinator interactions, further underscoring the ecological intricacy of this quite neglected and underutilized species. The presence of *Strobilanthes* species with proven antimicrobial, antioxidant, and anti-inflammatory activities highlights the need for more in-depth investigation into further therapeutic avenues. *S. callosus* represents a vital yet underexplored genus with profound ecological and ethnomedicinal significance.

A multidisciplinary approach integrating ecology, molecular biology, phytochemistry, and conservation science is essential to safeguard and harness the full potential of these remarkable plants for future generations.

Table 1: Blooming & Anther Dehiscence in Strobilanthes callosus (Nees) Bremek

Duration	No. of flowers opened	Duration	Anther dehiscence (%)
7.00- 9.00 am	42	**8.00- 10.00 am**	10.80
9.00- 11.00 am	55	**10.00- 12.00 am**	15.30
11.00- 01.00 pm	31	**12.00- 02.00 pm**	38.45

Table 2: Morphological parameters Strobilanthes callosus (Nees) Bremek

Plant Sample	Height of Plant (Meters)	Successive internode (Centimeter)	Length of Leaf (Centimeter)	Width of leaf (Centimeter)
Sample Plant 1	1-2 m.	2-3 cm.	6-15 cm	3-5 cm
Sample Plant 2	1.5- 2.5 m.	3-4 cm.	8-12 cm	4-7 cm
Sample Plant 3	2.5- 3 m.	2-3 cm.	12-18 cm	5-8 cm

Table 3A: Floral biological attributes of Strobilanthes callosus (Nees) Bremek

Parameters studied	Blooming stage	Full bloom stage
Color of corolla	Bluish-Purple	Violet
Width of Corolla	1-2 cm	3-4 cm
Type of Stamen	-	Didynamous
Length of Stamen	2.5- 3.5 cm	2.5- 4 cm
Anther Color	Yellow	Yellow
Attachment of Anther to filament	Basifixed	Basifixed
Position of Ovary	Superior	Superior
Fruit type	---	Capsule
Fruit length	---	1-3 cm
Fruit width	---	1- 1.5 cm
No. of Seed per fruit	---	2-4

Table 3B: Measurements of floral whorls of Strobilanthes callosus (Nees) Bremek

Floral whorls	Measurement (Mean ± SD)
Corolla	3.8 ± 0.2 cm
Style	2.1 ± 0.1 cm
Stigma	1.2 ± 0.05 cm
Ovary	0.8 ± 0.02 cm
Bract	2.3 ± 0.1 cm

Table 4: Percentage of Pollen Viability by Acetocarmine (1%) Test. [32,33]

Microscope Field No.	No. of Pollen grains in the field	No. of Stained Pollen grains			No. of Unstained Pollen grains	% Pollen Viability
		Dark-stained (Strong vitality)	**Light- stained (Weak vigor)**	**Total**		
1	19	09	05	14	05	73.68
2	17	11	03	14	03	82.35
3	21	12	05	17	04	80.95
4	18	11	04	15	03	83.33
5	20	10	06	16	04	80
Total	**95**	**53**	**23**	**76**	**19**	**80.4**

Fig. 2. Habit and Inflorescence of Strobilanthes callosus (Nees) Bremek

Fig. 3: Floral Visitors (Bees) of Strobilanthes callosus (Nees) Bremek

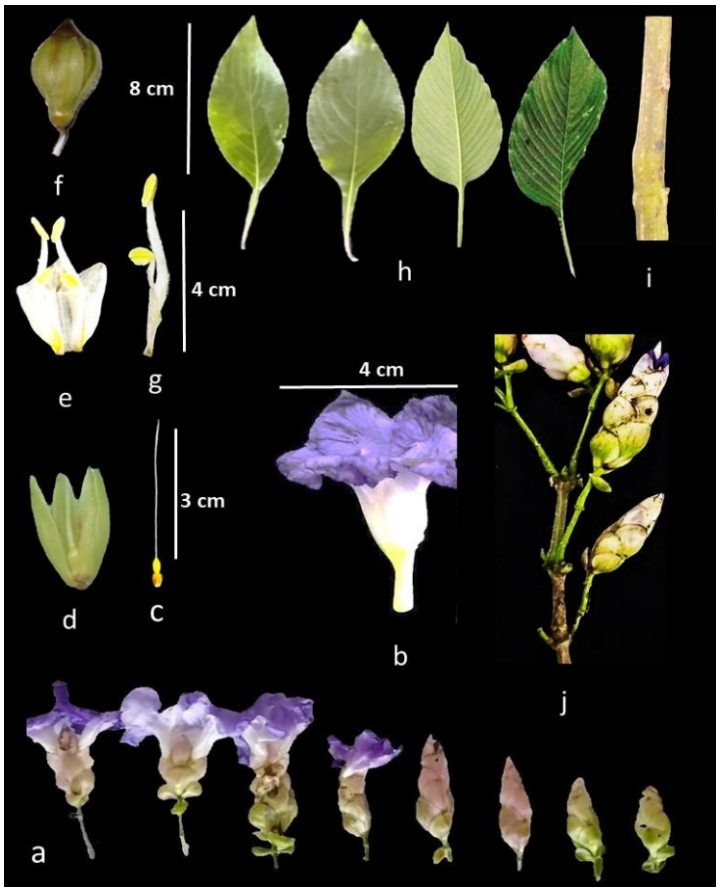

Fig.4: Floral attributes of Strobilanthes callosus (Nees) Bremek. a. Flowering stages b. Dissected flower c. Gynoecium d. Calyx. e & g. Didynamous Stamens f. fruit h. Leaves at different growth stages i. Stem showing node and internode j. Inflorescence.

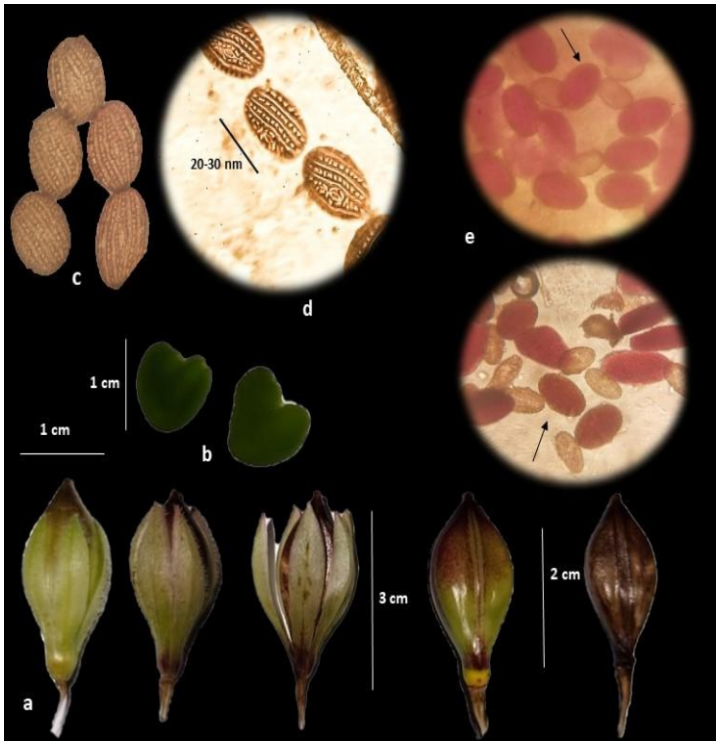

Fig. 5: a. Fruit and Pollen morphology of Strobilanthes callosus (Nees) Bremek. showing a. Obovoid two seeded capsule with white appressed hairs. b. Broadly ovate Seeds c.-d. Subspherical Pollen grains having tectate exine perforated with granular ornamentation. e. Viable pollen grains after using Acetocarmine (1%) stain.

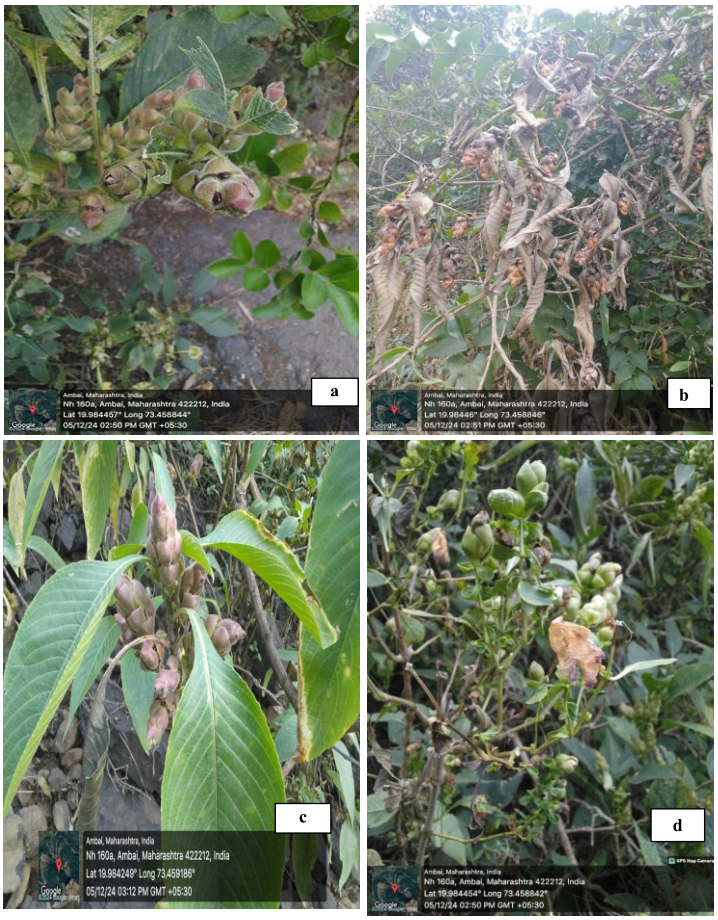

Fig. 6. Drying up of inflorescence (b, c), faded flower (d) and fading of vegetative growth after the formation of the seeds (a) in the same year.

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