Chapter 10 *Exacum bicolor* Roxb. an Exquisite, Under Exploited Wild Ornamental

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Abstract *Exacum bicolor* an elegant flowering plant in the wild, is distributed in Peninsular India. Natural habitats of the plant are grasslands of both high and low altitudes, as well as scrub savannah. The unique feature of this plant is its restricted occurrence in small localized areas. Natural habitats are being exploited for tourism, the construction of highways, and as sites for agriculture and industry. Ex situ conservation is the best strategy for conservation in view of habitat destruction. The plant is an excellent ornamental suitable for regions of high humidity and heavy rainfall. Horticultural aspects, such as sturdy stems, a long flowering period, many flowers in a bunch, large flower size and extended life of the flowers under field conditions, result in excellent ornamental value of the plant for pot cultivation and bedding. Natural seed germination is very low and normally less than 5 %. Analysis of soil in regions where the plants occur shows the presence of mycorrhizae. Pollination is insect assisted; fruit is not set following selfing. Seedlings show wide variation in vegetative and floral characters. Consequently, there exists high potential for selection and improvement. There are many reports regarding the febrifuge, stomachic, and tonic properties of *E. bicolor*. Traditional healers in regions of India, such as Kerala and Karnataka, prescribe the plant for the treatment of fever, malaria, and blood purification. Identification tests have confirmed the presence of various compounds, including phenols, alkaloids, flavanoids, tannins and saponins in plant extracts. Thin layer chromatography for qualitative analysis confirmed the presence of these metabolites.

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10.1 Introduction

Exacum bicolor Roxb., commonly known as country kreat, is an exquisite ornamental flowering plant found wild in the humid tropics. The plant is an erect herb with a sturdy stem that does not lodge even during heavy rain. It bears numerous attractive flowers that retain their beauty for a considerable time. Attempts to domesticate this attractive flowering plant have not been successful. Reports in the ancient literature state the medicinal uses of the plant for various ailments. Severe destruction of the natural habitats has now endangered the plant in the Western Ghats and dry grasslands of Southern India.

10.2 Systematics

E. bicolor Roxb. is a member of the tribe Exacaceae in the family Gentianaceae. Six genera are included in this tribe. According to Struwe et al. (2002), the genus *Exacum* was introduced by Linnaeus in 1747, with two species, *E. pedunculatum* and *E. sessile*, being added later (Linnaeus 1753). Roxburg (1832) included another five species in this genus. Klackenberg (1985) revised the genus describing 65 species. Members of the genus have a wide paleotropical distribution occurring from Africa to Madagascar and Socotra Island, and also from India to New Guinea to the Northern part of Australia. Broadly, the genus *Exacum* is divided into two Sections; Section *Exacum* has 21 species distributed in India, Sri Lanka, and South East Asia, while section *Africana* with 44 species is distributed in Madagascar, Socotra, Oman, and tropical Africa (Struwe et al. 2002). John et al. (2001) reported 16 species of *Exacum* occurring in India. The majority of the *Exacum* species are characterized by restricted and local endemic distribution. Some of the species have been collected only a few times and hence not studied extensively. In contrast, there are species such as *E. tetragonum* which are widely distributed.

Many authors have reported *E. bicolor* as synonymous with *E. tetragonum*. Roxburg (1832) allotted separate species status to the above two species in his classification. Bentham (1876) in *Genera Plantarum* described *E. bicolor* as only a form of *E. tetragonum*. Hooker (1885) gave separate species status to *E. bicolor* and *E. tetragonum*, but Klackenberg (1985) described *E. bicolor* as a synonym of *E. tetragonum*. Sivarajan and Philip (1997) highlighted the need for further studies before describing the Himalayan *E. tetragonum* and South Indian *E. bicolor* as a single species. A difference of opinion still exists about separate species status for *E. bicolor* and *E. tetragonum*, and more studies may be required to explain the following, namely,

- 1. The wide variation in chromosome numbers of the two species viz. *E. tetra*gonum is 2n = 18 and *E. bicolor* 2n = 62 (Struwe et al. 2002).
- 2. Anthers of *E. bicolor* are curved (Gamble 1915; Hooker 1885) whereas *E. tetragonum* has straight anthers.

- 3. *E. bicolor* is not seen naturally in sandy soils or marshy meadows or swamps of Kerala's South Western Ghats. The plant has restricted occurrence in Kerala. There are places with less than five plants in total. These findings do not conform to the studies of Klackenberg (1985), in which he reported a wide distribution for *E. tetragonum* in clay to sandy soils or waterlogged places, such as marshy meadows and swamps.
- 4. All the authors who have assigned separate species status to *bicolor* and *tet-ragonum* have mentioned the variation in flower color in the two species, *E. bicolor* having white flowers with blue corolla tips, while *E. tetragonum* has a blue/azure-colored corolla.
- 5. The present authors also feel that the choice of the epithet *E. bicolor* is more apt than *E. tetragonum*, as there are many species with quadrangular stems and tetramerous flowers in the genus *Exacum*.

10.3 Habitat Characterization

E. bicolor is a species endemic to Peninsular India. Natural habitats of the plant are grasslands of both high and low altitudes, as well as scrub savannah. The plant has been reported in the plateau region from the Konkan coast to the Southern tip of India, including Western Ghats. The species has wide adaptability to the climate prevailing in the plains of the tropics and high ranges in Western Ghats. A survey was conducted representing various habitats in the South Indian State of Kerala, the latter being a major portion of Southern Western Ghats, a hot spot of biodiversity. Floristic studies were undertaken initially to obtain a preliminary idea about the distribution of the plant. The majority of these studies represented forest areas such as Wild Life Sanctuaries, National Parks, and Bioreserves occurring at high altitudes. In the plains, where dry grasslands are present, survey work was carried out based on local enquiry. Dry grasslands usually occur in hillocks at 50-200 m elevation. Of a total of 125 sampling units surveyed, E. bicolor was cited only in 41 units with a frequency of 32.8 %. Nearly 70 % of these holdings are in private possession. Again, among the 41 units where the plant was recorded, 20 or less than 20 plants were recorded in 10 units during 2005. Among the 10 units with less than 20 plants, 6 had only 5 or less than 5 plants in the entire area (approximately half km²). The unique feature of the plant is its restricted occurrence in small areas even in a vast stretch of grassland. Sometimes, plants are seen only on one side of a hillock. During a revisit to 17 habitats in 2006, the plant disappeared from three of the sites due to complete destruction of the habitats. The status of various locations where E. bicolor was cited in Kerala is represented in Table 10.1. All the privately owned habitats are likely to be exploited at any time as decided by the owners for various reasons including laterite mining, soil excavation for various development activities, establishment of plantations of rubber and coconut, and also as industrial sites.

District	Number of habitats					
	Forest—shola grassland	Revenue— shola grassland	Revenue—dry grassland	Private—dry grassland	habitats	
Thiruvananthapuram	0	0	0	0	0	
Kollam	0	0	0	0	0	
Pathanamthitta	3	0 0	0	0	3	
Idukki	1	0	0	0	1	
Kottayam	0	1	0	0	1	
Thrissur	0	0	1	6	7	
Palakkad	1	0	0	2	3	
Malappuram	1	0	0	9	10	
Kozhikode	0	0	0	2	2	
Wayanad	5	0	0	0	5	
Kannur	0	0	0	7	7	
Kasargode	0	0	0	2	2	
Total	11	1	1	28	41	

 Table 10.1
 Status of habitats of occurrence of E. bicolor in Kerala

Table 10.2 Comparison of the altitude and occurrence of habitats of E. bicolor in Kerala

Sl.No.	Site particulars (Altitude from mean sea level)	No. of sites visited	No. of sites of plant occurrence	Frequency (%)	Density	Abundance
1	<500 m	80	29	36.3	2.2	3.9
2	>500 m	45	12	26.7	1.1	2.8

Altitudinal differences in plant distribution were also investigated (Sreelatha et al. 2009). For the study, sites were grouped mainly into two: those with an altitude less than 500 m, and those with an altitude exceeding 500 m. This grouping was done because above 500 m, the main associating flora, such as grasses, are very tall. Other associating plants were also similar from 500 m above mean sea level. Table 10.2 shows the frequency and numerical status of the plants above and below 500 m from mean sea level. It is evident that among the sites surveyed, plants were more frequent (36.3 %) at altitudes below 500 m. Greater density (2.2) and abundance (3.9) of *E. bicolor* were also recorded at these sites, indicating that the plant population is more concentrated in these areas than in high ranges (>500 m above mean sea level) of Kerala.

10.4 Associated Flora

The major associating flora in low altitude grasslands are grasses such as *Heter*opogon contortus, Dimeria bialata, Ischaemum indicum, ground orchids like Habenaria diphylla, and H. heyneana together with other herbs such as Sopubia delphinifolia, Striga densiflora, Eusteralis quadrifolia, Sesamum laciniatum, Spermacoce pusilla and Oldenlandia corymbosa. In high altitude grasslands, E. bicolor is seen in association with the grasses Themeda cymbaria, Chrysopogon zeylanicus, Cymbopogon flexuous, and Arundinella purpurea, and with other herbs including Ageratum conyzoides, Osbeckia sp., Tephrosia purpurea, Anaphalis sp., Hypericum mysorensis, Swertia sp. and Phoenix lourieii.

10.5 Mycorrhizal Association

Soil samples collected from four different natural habitats of the plant showed the presence of mycorrhizae. Preliminary studies identified *Glomus* and *Acaulospora* as the two associating fungi. Spores of the fungi were grown on maize roots and mycorrhizae infected roots of maize were added to the potting medium while transplanting seedlings of *E. bicolor*. There was considerable difference in the number of flowers per plant. Mycorrhizae-treated plants produced almost twofold more flowers than the control plants. This particular aspect needs detailed investigation for further confirmation.

10.6 Characteristics of Exacum bicolor

Exacum bicolor is a herbaceous perennial (Fig. 10.1), and in dry grasslands grows to a height of 25–80 cm. In high altitude grasslands attached to shola forests, where grasses grow thick and tall, the height of the plants ranges from 40 to 120 cm. The stem is quadrangular in shape and hard at the base when mature. It is nonlodging even in severe rains. Leaves are sessile, and linear lanceolate with five prominent veins, and are arranged in an opposite fashion. Variations such as oblong and cordate type leaves are also observed. In the plain regions, plants flower during September to November, while in high altitude areas flowering lasts up to January. The inflorescence is a dichasial cyme; the number of flowers is 2–125 per plant. On average, there may be 40 flowers per plant, with 10–20 buds in bloom at any time. The flowering period lasts for 30–45 days, with a single flower remaining for 8–10 days in the field. Flowers open from 6.00 to 8.00 am, close in the evening, and reopen the next morning. This pattern continues for 2–3 days after which flowers remain open.

Flowers are bisexual and tetramerous, with four winged sepals and a four-lobed rotate corolla. Petal length varies from 1.0 to 3.8 cm and width from 0.5 to 1.9 cm. The androecium consists of four epipetalous stamens alternating with the petals; the filaments are short, the anthers oblong, curved, and dehiscing by apical pores. The ovary is two celled, with many ovules on an axile placenta. The style is long, the stigma capitate, and well exerted above the anthers. The fruit is a dehiscent capsule with numerous minute seeds. The size of individual flowers ranges from

Fig. 10.1 Habit of *Exacum* bicolor



2.2 to 7.8 cm, with an average of 5.0 cm. The petals are white with dark purple tips; these are the most attractive part of the flower. Petal tip color varies in intensity from dark purple to blue and sometimes even violet. Five to six days after flower opening, the purple pigmentation spreads gradually downward with the whole petal finally turning light purple. The bright yellow stamens add to the beauty of the flowers. A bunch of flowers is usually a colorful mix of white, dark purple, light purple, and yellow (Fig. 10.2). Pollination is assisted by insects such as stingless bees, bumble bees, and carpenter bees. Fruit set is not observed when flowers are bagged or plants are kept in cloth covered cages. Nearly 4 months are required from flowering to fruit maturity. The capsules dehisce to release the minute seeds.

Each capsule weighs 130–150 mg with a seed weight of 25–30 mg per capsule. On average, 1 mg of seed contains 140 seeds, the average seed size being 298 μ m. Considering an average of 40 flowers per plant, the number of seeds produced per plant ranges from 140,000 to 170,000. Even with this large quantity of seed production per plant, the spread of the plant to adjacent areas is not usually observed. This may be due to the fact that seeds dispersed by wind may not be deposited in suitable open places to germinate and also seeds may become buried deeply in low lands where there is water logging. Other supporting factors for the isolated occurrence of *E. bicolor* may be the discontinuous genus status reported by Good (1953) and the mycorrhizal association characteristic of the Gentianaceae (Meszaros et al. 2002). Natural seed germination is also less than 5 %, as in the case of other gentians.

Fig. 10.2 A bunch of flowers of *Exacum bicolor*



10.7 Propagation

In natural habitats, the plant is propagated from tuberous roots that perennate underground. This was confirmed from the sprouting of new plants from dry stumps of the previous year (Fig. 10.3) and the same pattern was observed throughout a study period by the present authors. In ex situ conditions, sprouting is not common and plants that sprout do not show vigorous growth. Hence, propagation through seed has been standardized. A compost of sand and coir pith in equal proportions is ideal for seed germination. Seeds germinate within 2 weeks in this medium provided high humidity is maintained. Propagation by cuttings has been unsuccessful.

The protocol for *in vitro* seed germination, shoot multiplication, and plantlet production has also been standardized. For *in vitro* seed germination, the optimum medium is that of Murashige and Skoog (1962) at one quarter strength, supplemented with 1.0 mg/l 6-benzylamino purine (BA). Darkness induced earlier increased seed germination. Multiple shoots from in vitro seedlings were best induced on MS medium at half strength supplemented with 0.5 mg/l BA with 0.5 mg/l indole-3-acetic acid (IAA). Optimum rooting was MS-based medium at one quarter strength supplemented with 1.0 mg/l indole-3-butyric acid (IBA), 20 g/l sucrose, and 0.125 % activated charcoal.

Fig. 10.3 Sprouting of *Exacum bicolor* from dry stumps



10.8 Variation in Seedling Progeny

Wide variation occurs in seedling populations of *E. bicolor*. A seedling population was raised from seeds obtained from wild plants collected from a natural habitat. A total of 350 seedlings were planted in beds and grown under the same management practices. Observations were recorded on various quantitative characters including plant height, internodal length, number of branches, number of leaves, leaf length, leaf width, number of flowers, flower size, petal length, petal width, and length of the dark purple patch at each petal tip. Qualitative characters were also observed such as leaf shape, flower shape, intensity of the purple color at the petal tip and flower color variants. All the quantitative characters showed wide variability (Table 10.3); maximum variability was expressed for the number of flowers per plant, from 15 to 215. The wide variation observed in seedling progeny offers immense scope for selection and further improvement of *E. bicolor*.

Qualitative characters also differed extensively especially in leaf shape, petal shape, and petal tip color. Leaf shape ranged from lanceolate, linear to cordate and oblong. Petal shapes, which ultimately decide the flower shape, also showed considerable variation. Petal tip color varied from low to high intensities of purple,

Sl.No.	Plant characters	Mean	SD	CV	Range
1	Height (cm)	48.4	12.6	26.1	23-100
2	Internodal length (cm)	7.3	2.1	28.7	3-14.5
3	Number of branches	8.0	3.1	39.1	3-18
4	Number of leaves	12.7	5.1	40.0	4–49
5	Leaf length (cm)	8.0	2.5	31.1	2.4-14.5
6	Leaf width (cm)	2.8	1.0	37.1	1-7.5
7	Number of flowers	66.4	36.1	54.4	15-215
8	Flower size (cm)	4.4	1.8	40.9	3-5.6
9	Petal length (cm)	2.1	0.4	17.1	1.2-4.0
10	Petal width (cm)	0.9	0.2	24.4	0.2-1.3
11	Length of purple patch at petal tip (cm)	0.7	0.2	30.7	0-1.2

 Table 10.3
 Variability in the seedling population of E. bicolor

with occasionally a blue color. A rare occurrence of flowers with fully white petals was also observed. Unlike the usual pattern of petals turning light purple on fading, some plants had flowers with petals that become dark purple during this period.

10.9 Ornamental Uses

The ornamental value of the plant was reported earlier by Woodrow (1910) in his book "*Gardening in the Tropics.*" Although beautiful, the plant was reported by the author as being most difficult to cultivate. The ornamental value of *E. bicolor* has been reported by several scientists (Rao 1986; John et al. 2001; Sreelatha et al. 2006). The most valued character of the plant is its tolerance to heavy rain and high humidity, but it needs these conditions for optimum growth. The plant could be utilized as a potted ornamental as well as for flower borders. Annuals that are nonlodging and suitable for cultivation are rare in tropical, high rainfall areas. A long flowering period of 30–45 days, pleasingly large individual flowers and long field life of the flowers, add to the aesthetic value of this plant. In Kerala, the plant was very popular about 5–8 decades ago and flowers of *E. bicolor* were given considerable religious importance in certain regions of the State. It was one of the choice flowers to adorn the earthen deity "Trikkakarayappan," worshipped during the important regional festival "Onam" in Kerala. The vernacular name, *Kannamthali*, very often finds a place in local linguistic poems and film songs.

10.10 Medicinal Uses

Like any other gentian, *E. bicolor* is also reported to have many medicinal uses. Dried stalks of the plant were sold in South Indian markets during the early twentieth century (Rao 1914) and were widely used for their febrifuge properties.

Sl. No.	Plant organ	Seedling stage			Bud initiation stage			Flowering stage		
		Starch (%)	Soluble sugar (%)	Phenol (%)	Starch (%)	Soluble sugar (%)	Phenol (%)	Starch (%)	Soluble sugar (%)	Phenol (%)
1	Shoot	6.7	7.5	2.3	7.3	5.5	1.9	6.0	7.1	1.8
2	Root	6.5	6.1	0.7	4.0	4.6	0.9	6.9	4.7	0.6
3	Flower	-	-	-	-	-	-	8.5	9.8	3.0

Table 10.4 Quantitative analysis of dry plant samples of E. bicolor

The tonic and stomachic properties of the plant were reported as a substitute for *Gentiana* and *Swertia* (Anon, 1952). Chopra et al. (1956) and Srivastava (1989) quoted similar medicinal properties for the plant. However, the plant is not in commercial trade anywhere in India. Local enquiry during studies by the present authors revealed that the plant was used for treatment of fever, eye, and skin diseases and urinary disorders by traditional medical practitioners. However, substitutes are being used today due to lack of availability of the plant.

Primary metabolites like starch and sugars are essential for plant growth; many primary metabolites act as precursors for pharmacologically active compounds. Preliminary phytochemical screening has been done to analyze the content of primary and secondary metabolites. Quantitative analysis of dry plant samples for starch, soluble sugar, and phenol are given in Table 10.4. During the flowering stage, the content of primary metabolites reached a maximum which could support the medicinal property of the whole plant. Identification tests for various compounds such as alkaloids, flavonoids, tannins and saponins in dry plant samples have given positive results. Jeeshna and Paulsamy (2011) isolated six phytochemical compounds of medicinal importance from powdered whole plant material of *E. bicolor*. Of these compounds, two were polyphenolic, two were alkaloidal, one was glycosidal, and one was steroidal in nature.

10.11 Conservation Strategies

In Kerala, nearly 70 % of the natural habitats are in private ownership. Severe habitat destruction continues in all these areas causing depletion of the plant at an alarming rate. Habitat destruction is also reported from the Western Ghats regions of other States. *Ex situ* conservation is the only solution in this context. For any *ex situ* conservation programme, plants have to be domesticated, and for this to be achieved, the habitat, plant habit, propagation, floral biology and phenology must be studied in depth. This needs urgent attention in the prevailing social situations in Kerala and other South Indian States.

10.12 Conclusions

Wide adaptability to the climate of plains and high ranges, nonlodging stems even in severe rain, improved flower morphology, a long flowering period, and better adaptations to field conditions give potential for *E. bicolor* to be domesticated as an ornamental plant. Future research should concentrate on standardization of production technologies for popularizing *E. bicolor* as a garden plant. Although there are several reports about the medicinal value of this plant, scientific studies are required for commercial exploitation of its medicinal properties.

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