

REPRODUCTIVE BIOLOGY OF THREE MEDICINAL PLANTS

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Abstract

This paper presents the flower morphology, pollination mechanisms and seed germination of three indigenous medicinal plants of Bangladesh namely, *Asclepias curassavica* L., *Clerodendrum viscosum* Vent. and *Scoparia dulcis* L. The minimum days taken for seed germination in *Asclepias curassavica*, *Clerodendrum viscosum* and *Scoparia dulcis* are 6, 14 and 9, respectively. Epigeal type of seed germination has been noted in *Asclepias curassavica* and *Scoparia dulcis*, whereas hypogeal type of seed germination has been found in *Clerodendrum viscosum*. Fruit production is highest in *Scoparia dulcis* among the species studied and a maximum of 304 seeds are produced per fruit in it. Maximum 108 seeds are produced per fruit in *Asclepias curassavica*. Among the species investigated *Asclepias curassavica* and *Clerodendrum viscosum* are cross-pollinated, and *Scoparia dulcis* is self-pollinated. Butterflies, bees and ants are noted to be the pollinators in *Asclepias curassavica* and in *Clerodendrum viscosum* black ants, butterflies and long tongue hawk moths are the pollinators.

Introduction

Medicinal plants are of enormous economic importance and they are used as raw materials for the extraction of active constitution in pure form, as precursors for synthetic vitamins and steroids, and as preparations for herbal and indigenous medicines (de Padua *et al.*, 1999). *Asclepias curassavica*, *Clerodendrum viscosum* and *Scoparia dulcis* are three medicinally important plants. *Asclepias curassavica*, a member of the family Asclepiadaceae is highly valued in both traditional and modern medicine. Leaf juice of *Asclepias curassavica* is administered in abdominal pain, arrests haemorrhages and having anthelmintic properties, whereas roots are applied for asthma, piles and gonorrhoea (Ghani, 2003). *Clerodendrum viscosum* belongs to the family Verbenaceae is used in fever, cough and bronchitis, also applied for herpetic eruptions and as vermifuge and bitter tonic. *Scoparia dulcis* falls under the family Scrophulariaceae, is used as analgesic, diuretic and antipyretic, to treat diarrhoea and dysentery, also in cough, bronchitis, hypertension, haemorrhoids and insect bite (van Valkenburg and Bunyapraphatsara, 2002; Ghani, 2003).

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Over the past half century, numerous studies have addressed various aspects of reproductive biology, pollination and seed germination. Several studies on reproductive biology and pollination mechanism have been carried out in different group of plants. Cox (1990) investigated pollination and the evolution of breeding system in the members of the Pandanaceae. Hassan and Khan (1996) carried out pollination studies in the genus *Polygonum* and showed that mechanism of cross pollination is operating as a rule in heterostylar plants, whereas self pollination occurs in non-heterostylar plants. Wyatt and Broyles (1990) highlighted the reproductive biology in milkweeds (Asclepiadaceae). The biological function of the neotropical orchid genera was elucidated by the study of the pollination biology (Singer and Sazima, 1999). Very recently, Pansarin and Amaral (2009) investigated the reproductive biology and pollination in the genus *Stanhopea*. Studies on seed germination in different plants are well known and factors affecting seed germination have been recognized in different species (Yang *et al.*, 1999; Hassan and Fardous, 2003; Chauhan and Johnson, 2008).

Even though studies on reproductive biology were carried out in different groups of plants, however, no study was conducted on this subject so far in the medicinally important species *Asclepias curassavica*, *Clerodendrum viscosum* and *Scoparia dulcis*. Because of habitat destruction and over-exploitation many medicinal plants are going to be endangered. Consequently, economically important plants should be brought under cultivation. Prior to bring them under cultivation their detailed reproductive biology should be studied. Therefore, the present study was undertaken to investigate different biological characteristics like mode of pollination and seed germination in these three indigenous important medicinal plants which might help us in bringing the plants under cultivation.

Materials and Methods

Three medicinal plants were selected for this study namely, *Asclepias curassavica*, *Clerodendrum viscosum* and *Scoparia dulcis*. Plants were collected from different parts of the country and planted in the Botanic garden of Dhaka University for closer observation and critical study. Flower morphology of each plant was studied critically.

Bagging experiment: In order to understand the pollination mechanism bagging experiments were carried out. Bags of fine cotton cloth were used for bagging which were done to note the mode of pollination of the flowers using the protocol of Hassan and Khan (1996). Unopened floral buds were caged by cotton bags and kept under observation. Emasculated floral buds were also brought under bagging. Observations on phenology were made throughout the year.

Seed germination experiment: Seeds were collected from mature fruits for germination experiments. Earthen pots of 10 inch in diameter filled up with a mixture of soil and compost (2:1) were used for seed sowing. Ten to thirty seeds at each time were

sown at different times of the year to record dormancy and viability, suitable time for germination, percentage and type of germination.

Results and Discussion

Flower morphology

Asclepias curassavica L., Sp. Pl.: 215 (1753).

(Fig. 1)

Flowers hermaphrodite, complete, c 1.7-1.8 cm across. Calyx 5, polysepalous, 3-4 mm long, 1 mm wide, narrowly lanceolate or linear-lanceolate, persistent. Petals 5, rotate, about 1.5-1.8 cm long, regular, elliptic, corolla tube short, crowned by a corona arisen from corolla, lobes reflexed, orange-red or bright crimson, corona adnate to the staminal

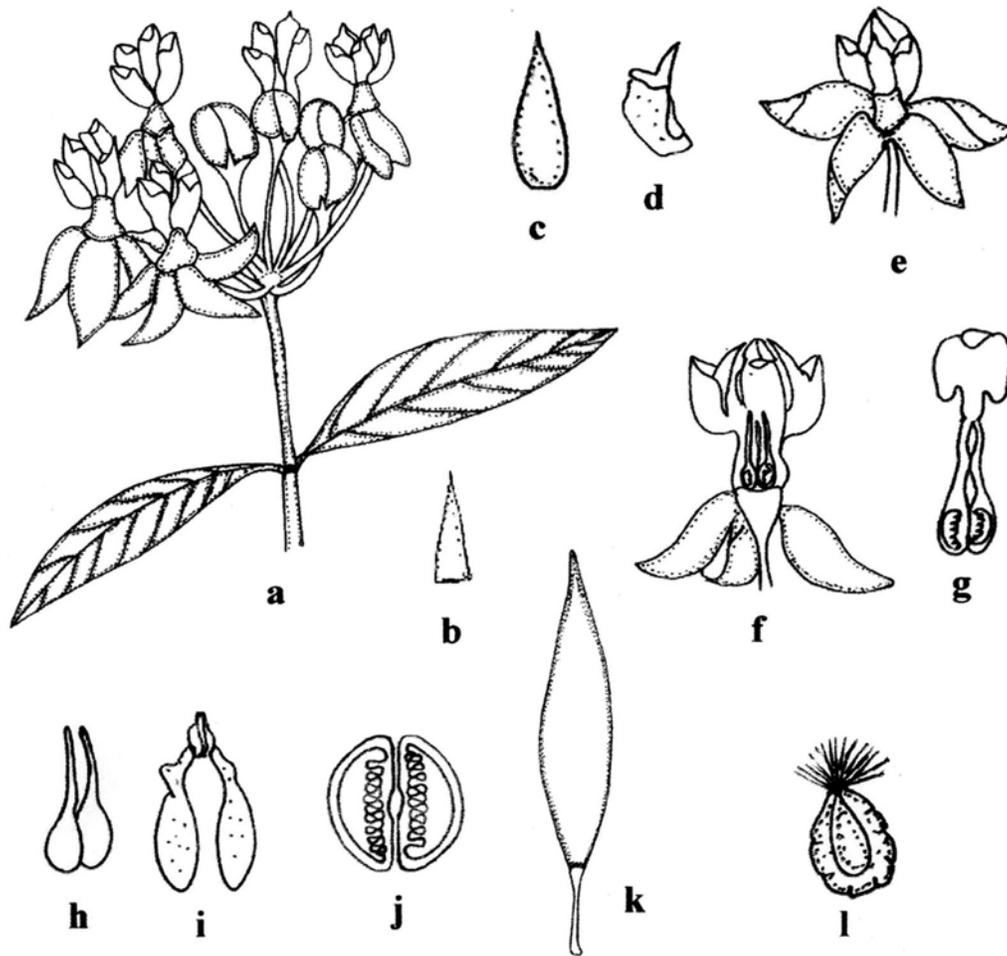


Fig. 1. *Asclepias curassavica* L.; a. Habit sketch with flowering branch ($\times 1$); b. Sepal ($\times 5$); c. Petal ($\times 1.5$); d. Corona ($\times 3.5$); e. Flower ($\times 1.5$); f. L. S. of flower ($\times 2.5$); g. Gynandrium ($\times 4$); h. Carpels ($\times 4.75$); i. Pollinia ($\times 20$); j. T.S. of ovary ($\times 18$); k. Fruit ($\times 0.6$); l. Seed ($\times 2.5$).

column. Gynostegium about 3 x 2 mm, cylindrical, anthers up to 2.5 cm long. Pollinaria 5, pollinia ovate-lanceolate, caudicular, pendulous, flattened. Ovary 2 or 3; styles 2 or 3, up to 3 mm long, glabrous. Flowering time: Almost throughout the year.

Clerodendrum viscosum Vent., Jard. Malm. 1: 25, t. 25 (1803).

(Fig. 2)

Flowers hermaphrodite, complete, white, pink at the centre, at evening sweet scented but odourless during the day. Sepals 5, gamosepalous, c 1.0-2.9 cm long, tubular, red, erect, lanceolate, silky pubescent. Corolla 5, tubular, lobes spreading, as long as the tube, white and purplish-pink at the mouth of the tube, rounded at the top, the tube 1.5-1.7 cm long, oblong, obtuse. Stamens 4, didynamous; filament c 1.5-4.2 cm long, white to purplish-red; anthers oblong, dorsifixed, 2-celled; pollen bursts transversely. Ovary glabrous; style slender, c 4.6 cm long; stigma short and 2-fid, c 0.2 cm long, white. Flowering time: January-June.

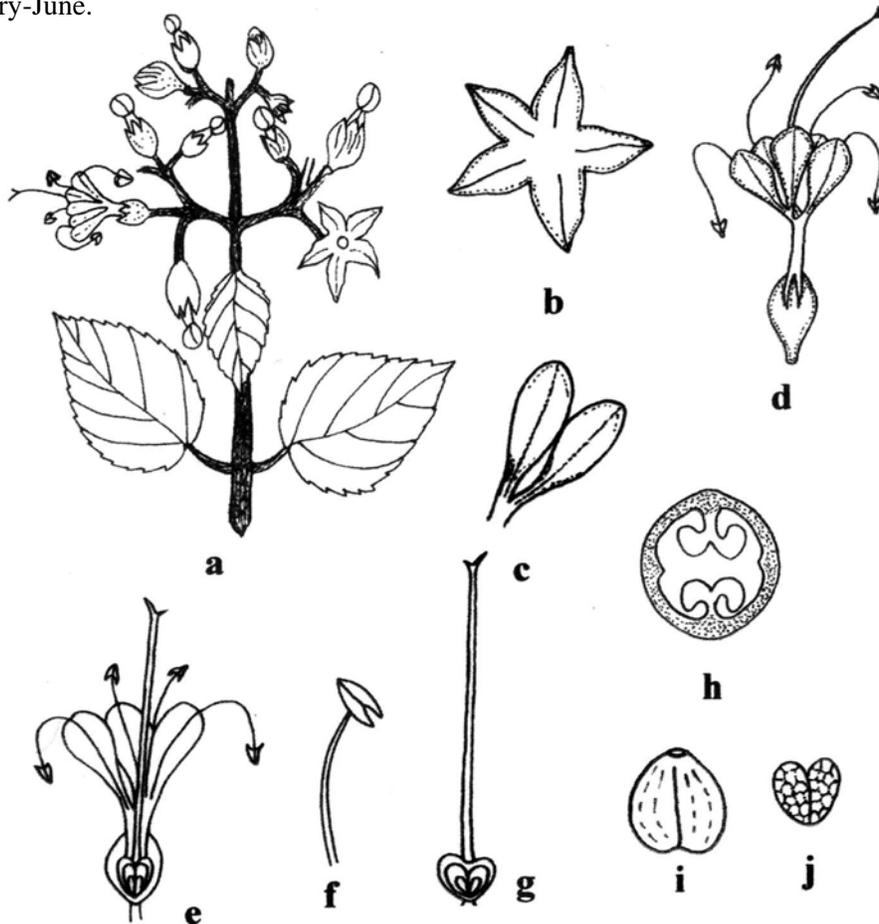


Fig. 2. *Clerodendrum viscosum* Vent.; a. Habit sketch ($\times 0.3$); b. Sepals ($\times 0.7$); c. Petals ($\times 1.6$); d. A flower ($\times 1.2$); e. L. S. of flower ($\times 1$); f. Stamen ($\times 0.6$); g. Carpel ($\times 1$); h. T.S. of ovary ($\times 20$); i. Fruit ($\times 1.2$); j. Seed ($\times 1$).

Scoparia dulcis L., Sp. Pl. : 116 (1753).

(Fig. 3)

Flowers hermaphrodite, complete, usually axillary, c 6-7 mm in diameter, 4-fid, rotate, regular. Sepals 4-5, gamosepalous, regular, calyx lobes oval-oblong, 2.5-3.0 x 0.8-1.0 mm, 3-nerved, glabrous, ciliate at the margin, persistent. Corolla pale yellow to white, corona present, tube densely hairy at the throat, lobes 2-4 mm long, apex obtuse, slightly curvy, upper lobes slightly larger than others. Stamens 4, exserted; filament inserted at the top of the corolla tube, glabrous; anthers dorsifixed. Style erect, c 2 mm long; stigma truncate to 2-partite, sometimes notched. Flowering time: Almost throughout the year.

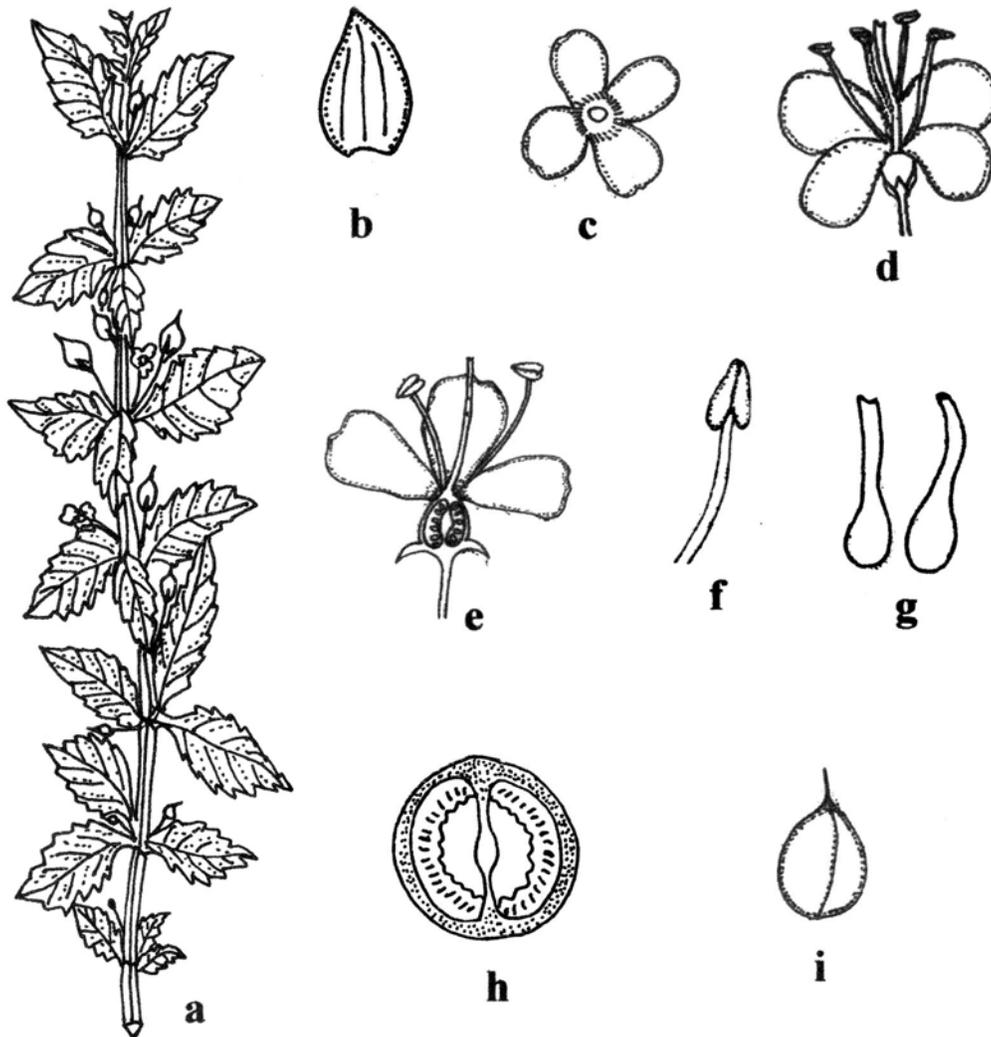


Fig. 3. *Scoparia dulcis* L.; a. A habit sketch ($\times 1$); b. Sepal ($\times 8$); c. Petal ($\times 4.5$); d. A flower ($\times 5$); e. L. S. of flower ($\times 5$); f. Stamen ($\times 6.5$); g. Carpels ($\times 8$); h. T.S. of ovary ($\times 50$); i. Fruit ($\times 6$).

Pollination

The results of pollination experiment are summarized in Table 1. In *Asclepias curassavica* five stamens are usually adnate to the stigma with the pollen agglutinated into pollinia united into pairs. So the 5-angled depressed stigma cannot accept pollens because of its structure. Besides, the larva of butterflies are seen that move from one flower to another. The bagging experiment did not reveal any fruit formation under the bag, however, fruits set under control indicating that cross-pollination is the usual method in *Asclepias curassavica*. Butterflies, bees and ants are the pollinators for this species (Table 1).

Table 1. Production of fruits in bagging experiment in three medicinal plants ('+' indicates positive fruit setting, '-' indicates negative fruit setting).

Sl. No.	Species	Bagging period (days)	Production of fruits		Type of pollination	Pollinators
			Bagged Plant	Control		
1.	<i>Asclepias curassavica</i>	c. 13	-	+	Cross-pollinated	Butterflies, ants, bees
2.	<i>Clerodendrum viscosum</i>	c. 10	-	+	Cross-pollinated	Ants, butterflies, hawk moths
3.	<i>Scoparia dulcis</i>	c. 12	+	+	Self-pollinated	-

In *Clerodendrum viscosum* the stigma cannot accept pollens for their arrangement. Many big black ants and butterflies are seen that moved on flowers to flowers but long tongue hawk-moth are also the pollinators for the tubular flower. Cross pollination occurs in this species as no fruit was formed within the bag. On the other hand, fruit formation takes place under control. In *Clerodendrum viscosum* the types of cross-pollination are Myrmecophily, Psychophily or Phalaenophily. The emasculated flowers do not produce fruits under bagging condition in *Clerodendrum viscosum*. Therefore, it could be said that there is no possibility of apomixis.

In *Scoparia dulcis* the exerted stamens with filaments are inserted at the top of the corolla tube and truncate to bipartite stigma present. Fruit formation starts within 4-5 days within the bag and almost each carpel is turned to a fruit and fruits mature within 12 days. Therefore it is evident that self-pollination mechanism occurs in this species indicating *Scoparia dulcis* a self-pollinated plant.

Seed germination

Most plants reproduce through production of seeds. Seed germination experiments were carried out in *Asclepias curassavica*, *Clerodendrum viscosum* and *Scoparia dulcis*. Result of seed germination in *Asclepias curassavica* is depicted in Table 2. It is evident that seeds sown immediately after collection in January (14.1.2008) took more or less 6

days to germinate in *Asclepias curassavica*, whereas seeds sown in early March (8.3.2008) took about 13 days. Seeds sown after six month of storage did not germinate indicating that their viability was lost (Table 2). Seeds that fall on the ground after maturity of fruits usually germinated in January. Plants that germinated from seeds flower and fruit in the same season. A Plant of moderate size produces c 20-25 fruits per year. Maximum numbers of seeds produced in *Asclepias curassavica* are 108 per fruit. The type of seed germination was noted to be epigeal.

Table 2. Seed germination in *Asclepias curassavica*.

Date of seed collection	Date of seed sown	No. of seeds sown	Date of seed germinated	No. of seeds germinated	Days taken to germinate	% of seed germinated
	14.01.08	10	20.01.08	10	c 6	100
14.01.08	10.02.08	10	20.02.08	10	c 10	100
	08.03.08	10	21.03.08	6	c 13	60
	19.07.08	10	Not germinated	0	Not germinated	0

Seed germination in *Clerodendrum viscosum* is displayed in Table 3. The seed germination time for *Clerodendrum viscosum* is May to July and the minimum days taken to germinate when sown in May indicating that the usual germination period for this species is May. Seeds that fall on the ground after maturity of fruits usually germinate in May and early June. Plants that germinated from seeds do not flower and fruit in the same season. The type of seed germination in *Clerodendrum viscosum* was hypogeal.

Table 3. Seed germination in *Clerodendrum viscosum*.

Date of seed collection	Date of seed sown	No. of seeds sown	Date of seed germination	No. of seeds germinated	Days taken to germinate	% of seed germinated
	15.05.08	10	29.05.08	10	c 14	100
01.05.08	03.06.08	10	22.06.08	10	c 19	100
	01.07.08	10	24.07.08	4	c 23	40
	19.08.08	10	Not germinated	0	Not germinated	0

In *Scoparia dulcis* it takes 9-19 days to germinate seeds (Table 4). If seeds are sown after immediate collection, minimum days are required to germinate and germination rate becomes high. Seeds that fall on the ground after maturity of fruits usually germinate within 7-10 days. Plants that germinated from seeds usually flower and fruit in the same season. At every nodes of the plant usually 4 fruits are set. Plant of moderate size produces numerous fruits per year and each fruit contains a maximum of 304 seeds. Epigeal type of germination was found in *Scoparia dulcis*.

Table 4. Seed germination in *Scoparia dulcis*.

Date of seed collection	Date of seed sown	No. of seeds sown	Date of seed germination	No. of seeds germinated	Days taken to germinate	% of seed germinated
	19.07.07	30	28.07.07	30	c 9	100
19.07.07	03.08.07	30	16.08.07	26	c13	86.7
	19.01.08	30	02.02.08	24	c 14	80.0
	16.09.08	30	05.10.08	4	c 19	13.34

The present study shows that in *Asclepias curassavica* the usual germination period is January to February, in *Clerodendrum viscosum* the germination time is May to June, and in *Scoparia dulcis* it is almost throughout the year. It has been observed that among the species studied the minimum days taken to germinate seeds are in *Asclepias curassavica*, whereas the maximum days taken to germinate are in *Clerodendrum viscosum*. The development of seedlings from seeds up to maturity in the species studied has been shown in Plate 1.

In the present study we investigated floral morphology, seed germination and pollination mechanism of three medicinal plants. The present study reveals that hypogeal germination is found in *Clerodendrum viscosum*, whereas epigeal germination is observed in *Asclepias curassavica* and *Scoparia dulcis*. Self-pollination has been found to occur in *Scoparia dulcis* and cross-pollination is found in *Asclepias curassavica* and *Clerodendrum viscosum*. The main pollinators revealed from this study include black ants, butterflies, honey bees and hawk-moth. The role of butterflies, bees and ants have already been recognized as potential pollinators in different plants (Sazima *et al.*, 1993). *Asclepias* pollinators include bees, moths and butterflies (Kephart, 1983; Broyles and Wyatt, 1991). Purseglove (1968) postulated that flowers with white corolla, strong perfume, abundant nectar and sticky pollen are entomophilous and are visited by bees and thrips. However, our study showed that white flowers of *Clerodendrum viscosum* are visited by black ants, butterflies and hawk moths. In *Asclepias curassavica* cross-pollination has been done by butterflies, ants and bees. Among the species studied *Asclepias curassavica* and *Clerodendrum viscosum* are heterostylar plants, while *Scoparia dulcis* is a homostylar plant. The structures of stigma or the arrangement of stamens and carpels in *Asclepias curassavica* and *Clerodendrum viscosum* support the cross-pollination. In case of heterostylar species, no fruit is formed under bagged condition indicating cross-pollination as the breeding mechanism. Our results support the previous study on pollination mechanism involving bagging experiments indicates that mechanism of cross-pollination is operating as a rule in heterostylar plants due to the presence of the capitate stigmas, whereas self pollination occurs in homostylar plant (Hassan and Khan, 1996).

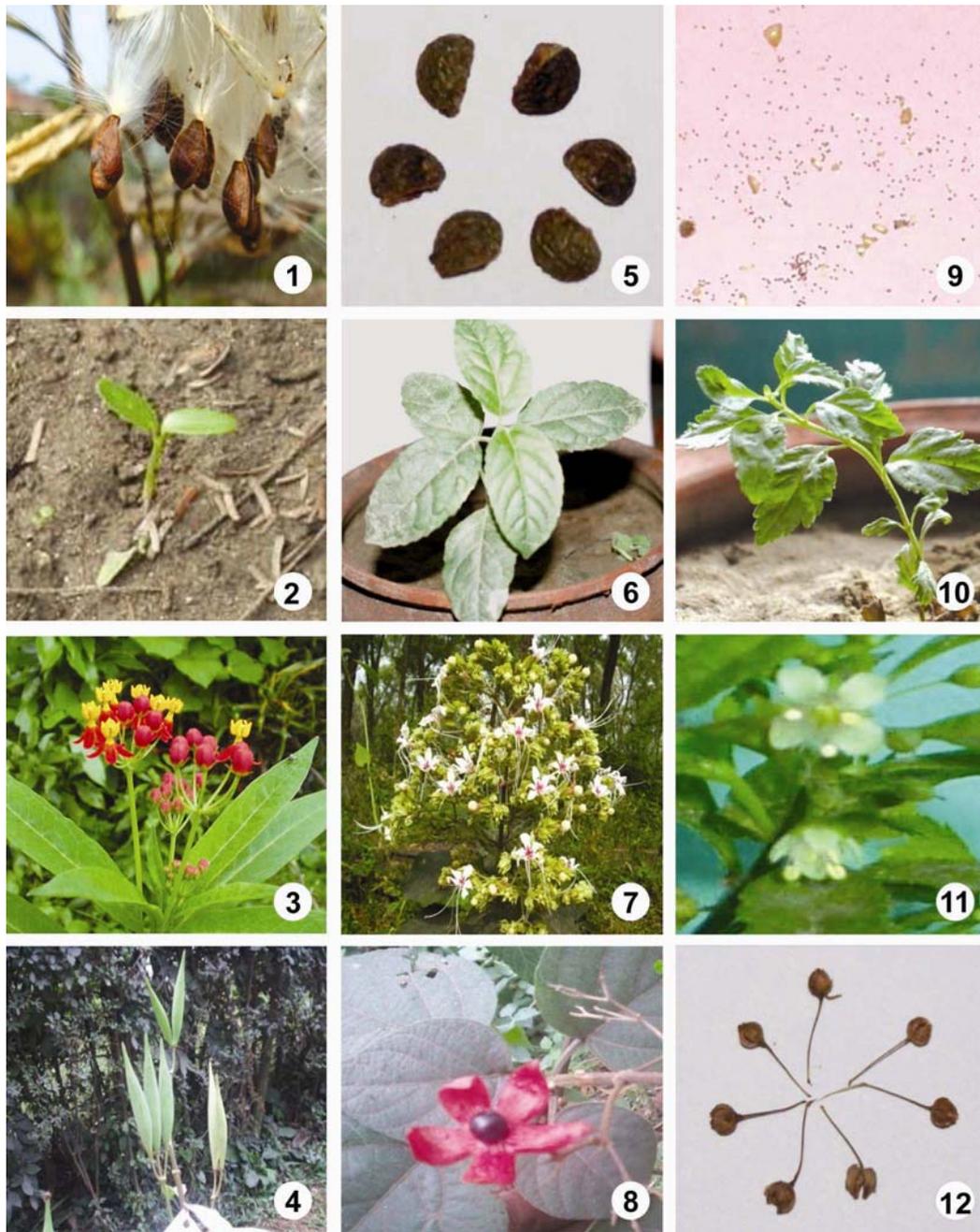


Plate 1. Development stages of three medicinal plants. 1-4. *Asclepias curassavica* (1. seeds; 2. seedling; 3. mature plant with flowering stage; 4. fruiting stage). 5-8. *Clerodendrum viscosum* (5. seeds; 6. seedling; 7. mature plant with flowering stage; 8. fruiting stage). 9-12. *Scoparia dulcis* (9. seeds; 10. seedling; 11. mature plant with flowering stage; 12. fruiting stage).

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