Reproductive biology and ecology of *Evolvulus alsinoides* and *Evolvulus nummularius* (Convolvulaceae)

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Abstract. The authors have found that *Evolvulus alsinoides* and *E. nummularius* grow and flower throughout the year, if the soil is wet. The pollen in both species is totally fertile and viable for 9–10 hours. This study has found that both species are facultatively xenogamous and refutes an earlier report that *E. nummularius* is facultatively autogamous. Furthermore, the flowers of these species resort to spontaneous autogamy during their closing time, which benefits maximization of the fruit set, but increase in the seed set rate is constrained by free styles and stigmas, which do not facilitate distinct contact with the anthers. According to earlier reports, *E. alsinoides* seemingly does not produce nectar, while *E. nummularius* definitely does not produce it. However, in this study it has been found that both species produce nectar and are pollinated by bees and butterflies, contrary to an earlier report that *E. alsinoides* is pollinated only by honey bees and snails. An additional finding is that both species display ballistochory, anemochory and hydrochory for seed dispersal; seeds have dormancy, as they germinate only during the rainy season. The woody rootstock also sprouts back to life to produce new foliage and reproduce sexually. These species have been found to be excellent sand binders as they usually form large patchy populations on sandy soils.

Key words: *Evolvulus alsinoides, Evolvulus nummularius*, facultative xenogamy, bees, butterflies, ballistochory, anemochory, hydrochory.

Introduction

Junquiera & Simao-Bianchini (2006) reported *Evolvulus* as a genus comprising about 100 species. Most of these species are South American, and many of them have extended into Central and North America. The genus is characterized by the presence of malpighiaceous trichomes (two-branched hairs), two free or basally-united styles, each with two filiform stigmas, and glabrous seeds. Austin (2010) reported that *E. alsinoides*, a perennial herb, is native to the Americas but it is now widespread across the tropical and subtropical regions of the world. Schmelzer & Bunyapraphatsara (2001) noted that *E. num*- *mularius* has spread across the tropics and subtropics, including much of tropical Africa, South Africa and Madagascar. These authors also mentioned that in Africa and Asia, *E. alsinoides* is widely used in traditional medicine as laxative, to treat stiffness of the limbs, sores and bleeding wounds, enlarged glands in the neck, leprosy, asthma, chronic bronchitis, skin infections, menstrual problems, as stomachic, against diarrhoea, cataract, depression, and for smoking. Austin (2010) noted that it is widely used in Ayurvedic medicine for memory enhancement, as antidepressant, antiepileptic and aphrodisiac, to cure syphilis and for hair growth promotion in India. Ketjarun & al. (2016) reviewed the reports on the medicinal value of E. alsinoides and E. nummularius. These authors noted that E. alsinoides produces several chemical components which have medicinal value. The plant is noted to be useful as brain and memory tonic, as anti-asthmatic cure, for treating uterine bleeding, insanity, epilepsy and nervous debility, for its antibacterial, antifungal, and antiulcer properties. E. nummularius is useful as a mild sedative and has anthelmintic properties. It was noted in the review that because of all these attributed properties, the two Evolvulus species are sold commonly via Internet as a powder used for brewing medicinal tea. Singh & al. (2010) provided some information on certain aspects of sexual reproduction in E. alsinoides. Sarma & al. (2007) reported that E. nummularius is facultatively autogamous and pollinated by snails and honey bees. Detailed information on the reproductive biology of both E. alsinoides and E. nummularius is not available, despite their medicinal importance and use as soil binders. Reproductive biology information on all other Evolvulus species is not available too. Therefore, the present study is aimed at providing details of reproductive ecology of these two species, discussed in the light of relevant literature.

Material and methods

Evolvulus alsinoides and E. nummularius growing in Visakhapatnam and its surroundings in India (17°42'N Latitude and 82°18'E Longitude) were studied from June 2014 to June 2016. Field trips were conducted to record the phenological aspects of the species. Ten inflorescences on five plants prior to flowering were tagged and followed daily to record the duration of flowering, anthesis schedule and timing of anther dehiscence. Twenty-five fresh flowers, five each from five plants, were used to record the floral morphological details. Nectar could not be measured and analyzed due to its secretion in minute quantity, further depleted by thrips in the mature bud stage and after anthesis. The methodology described by Cruden (1977) was used to estimate pollen output and pollen-ovule ratio; twenty mature, but indehisced anthers from ten plants were used for this study. In vitro pollen germination rate was observed using Modified Brewbaker and Kwack

media. The fresh pollen grains were collected from virgin flowers after anther dehiscence and placed in Petri dish in lab conditions. They were put on cavity slides, added the nutrient media, covered with cover slip, and observed at hourly intervals to record the percentage of pollen germination. The methodology described by Dafni & al. (2005) was followed for testing stigma receptivity. Ten flowers, each from five plants, were used to test stigma receptivity. Furthermore, stigma receptivity was also observed visually to note the physical state and colour appearance. Twenty patches with two hundred and fifteen mature buds for E. alsinoides and four hundred thirty-five mature buds for E. nummularius were tagged and followed for three weeks to record fruit and seed set in open pollinations. A sample of 378 fruits of E. alsinoides and of 373 fruits of E. nummularius was collected randomly to record the seed set rate based on ovule number per flower. The fruit and seed morphological characteristics were recorded and measured to evaluate their adaptations for dispersal by different vectors. Fields visits were made during dry and rainy seasons to note the aspects of seed germination and production of new plants. Furthermore, field observations were carried out on the production of fresh growth and sexual reproduction from the prennial woody rootstock of the plant.

Insect foraging activity schedule and foraging visits of insects were recorded using a 2×2 m area of flowering patch for 10 min each hour, for the entire day, on four different days, and the data was tabulated to record the foraging pattern and the percentage of foraging visits made. During that period, the mode of approach, landing, forage collection behavior, and contact of insects with the floral sexual organs were carefully observed to note their role in pollen transfer and pollination. Bees were identified by the representative specimens available at the Department of Environmental Sciences, Andhra University, Visakhapatnam. Butterflies were identified by consulting the books of Kunte (2007). Ten specimens of each insect species were captured during peak foraging period and brought to the laboratory. Each specimen was washed in ethyl alcohol, stained with aniline-blue on a glass slide and observed under microscope to count the number of pollen grains present. Thus, the average number of pollen grains carried by each insect species was calculated for the purpose of pollen carryover efficiency.

Results

Evolvulus alsinoides L.

Phenology: The plant is an annual prostrate, diffuse, densely hispid, perennial herb with a woody branched rootstock and many spreading and ascending branches (Fig. 1a). It is a weed of sandy, open, dry and rock localities with variable soil moisture content. The stems are 20-70 cm long, slender, reclining, erect and deep-rooted. The leaves are sessile, simple, ovate, lanceolate-linear to elliptic, with blunt apex and densely clothed with appressed, white and silky hairs. The plant re-grows from the perennial ground rootstock and also from seed during rainy season. The plants, which grow from the already existing rootstock, flower in July and those growing from seed, in August. Duration of flowering and life of the plant depend on the soil moisture conditions. The flowering was profuse in July-October, when the soil was sufficiently wet due to heavy rainfalls (Fig. 1b). If the soil was sufficiently wet, even during the summer season, the plant survived and produced flowers depending on the soil nutrient status. In such plants, the flowering occurs across the year.

Flower morphology: The flowers are pedicellate, solitary, borne in leaf axils and quite distinct against the foliage. They are small (6.2 ± 0.4 mm long, 9.6 ± 0.6 mm wide), pale- to dark-blue with a white throat, funnel-shaped, odourless, actinomorphic, and bisexual. The calyx has five free sepals, partially fused at the base;

the sepals are green, densely hairy, lanceolate, acuminate, 2.6 ± 0.4 mm long and 2 mm wide. The corolla consists of five united blue petals forming a short tube at the base, bell-shaped to rotate, 8-10 mm wide and glabrous. The stamens are white, epipetalous, exerted and glabrous; they are dimorphic (3+2) due to insertion of their filaments into the corolla tube at different lengths (Fig. 1f). The anthers are white, glabrous, 1 mm long and dithecous. The ovary (6.4 ± 0.4 mm long) is green and bicarpellary, bilocular, with two ovules arranged on axile placentation in each locule. The styles and stigmas are pure white; the styles are two in number, short and each style extends into a long 2-lobed minutely capitate stigma (Fig. 1h-j).

Floral biology: Mature buds open at 06:00–07:00 h on clear sunny days and at 06:30–07:30 h on rainy days (Fig. 1c). The flowers open completely on sunny days, and remain partially open on rainy days. The anthers dehisce by longitudinal slits during anthesis and the pollen is presented latrosely (the split on the side of the anther positioned towards the other anthers rather than towards the inside or outside of the flower). The number of pollen grains per anther is 981.8±66.76 and per flower is 4909±333.84. The pollen-ovule ratio is 1227:1. The pollen grains are white, initially sticky, later powdery, spherical, 5-zonocolpate, 27.39 × 31.42 µm and with smooth exine (Fig. 1g). *In vitro* pollen grains are viable and have the ability to germinate as soon as the anthers dehisce,



Fig. 1. Evolvulus alsinoides: **a** & **b** – habit with flowering phase; **c** – maturing bud; **d** & **e** – flower showing the relative positions of stamens and stailes; **f** – stamens; **g** – pollen grain; **h** – ovary with styles and stigmas; **i** – ovary; **j** – ovules; **k** & **m** – honey bee foragers; **k** & **l** – Apis cerana collecting nectar; **m** – Apis florea collecting pollen; **n** – Ceratina sp. collecting pollen.

and remain viable until 16:00 h, with a gradual decrease in germination rate (Table 1). The stigma becomes receptive thirty minutes after anthesis and remains so until 18:00 h of the same day. The stigma is shiny, dry and non-papillate during receptive phase. A nectary is situated at the base of the flower; it is limited to the area of the receptacle. The nectar is secreted in minute quantity during mature bud stage and is exposed on the anthesis. It is present in traces in open flowers due to feeding by thrips in buds. Thrips use the growing buds for breeding and emerge by the time the buds bloom. During mature bud phase, anthesis and post-anthesis, the thrips continually feed on nectar and pollen. The corolla, along with the stamens and receptive stigma, closes back by 12:00 h on the same day. The pollinated and fertilized flowers remain in place, while in unpollinated flowers the entire flower falls off on the third day.

Insect visitors and pollination: Thrips were found to be contributing to primary self-pollination by feed-

 Table 1. In vitro pollen germination in E. alsinoides and E. nummularius.

		E. alsinoides		E. nummularius			
Time (h)	Pollen sample	No. of pollen grains germinated	Germination (%)	No. of pollen grains germinated	Germination (%)		
0700	100	78	78	-	-		
0800	100	67	67	89	89		
0900	100	53	53	78	78		
1000	100	38	38	66	66		
1100	100	36	36	54	54		
1200	100	24	24	39	39		
1300	100	20	20	37	37		
1400	100	18	18	22	22		
1500	100	8	8	12	12		
1600	100	2	2	1	1		
1700	100	0	0	0	0		

Table 2. List of insect foragers on *E. alsinoides* and *E. nummularius*.

ing on both pollen and nectar since the beginning of anthesis. The flowers were foraged by honey bees, small carpenter bees and lycaenid butterflies during daytime, from 08:00 to 11:00 h, with concentrated foraging activity between 09:00-10:00 h (Fig. 3). The bees included Apis cerana (Fig. 1k,l), A. florea (Fig. 1m) (honey bees), Ceratina sp. (small carpenter bee) (Fig. 1n), while the lycaenid butterflies included Zizeeria karsandra, Zizina otis, Chilades laius, and Chilades pandava (Table 2). Of the total foraging visits recorded during the observation period, bees accounted for 59% and lycaenids for 41%. The bees approached the flowers in upright position, landed on the corolla and probed for pollen and/ or nectar. To collect nectar, they inserted their hairy tongue (proboscis) into the corolla throat to access the available nectar; in so doing, the ventral as well as dorsal side brushed against the dehisced anthers and the stigmas effecting both sternotribic and nototribic pollination. To collect pollen, the bees approached individual anthers and then only the ventral side and the stigmas brushed against the anthers effecting only sternotribic pollination. During most foraging visits, the worker honey bees and female small carpenter bees probed for nectar and pollen, while during other visits, both categories of bees (workers and females) collected either nectar or pollen. The butterflies approached the flowers



Fig. 3. Hourly foraging activity of insect foragers on *Evolvulus alsinoides*.

Order	Family	Genus	Species	Common name	E. alsinoides	E. nummularius	Forage sought
Hymenoptera	Apidae	Apis	<i>cerana</i> F.	Indian Honey Bee	+	+	P + N
		Apis	florea F.	Dwarf Honey Bee	+	+	P + N
		Trigona	Iridipennis	STINGLESS bee		+	P + N
		Ceratina	sp.	Small Carpenter Bee	+	_	_
Lepidoptera	Lycaenidae	Zizeeria	<i>karsandra</i> Moore	Dark Grass Blue	+	+	Ν
		Zizina	otis F.	Lesser Grass Blue	+	+	Ν
		Chilades	laius Stoll	Lime Blue	+	+	Ν
		Chilades	pandava Horsfield	Plains Cupid	+	+	Ν
		Euchrysops	cnejus F.	Gram Blue		+	Ν

P = Pollen, N = Nectar

with a head facing the corolla throat; then they inserted their proboscis to collect nectar and their head and ventral side brushed against the anthers and the stigmas effecting sternotribic pollination. The bees and butterflies visited 8-10 flowers consecutively and in some cases up to 18-23 flowers, depending on the flower density, before leaving the flowering patch. As the nectar was consistently collected by thrips, the insects made multiple visits to the same flowers in quest of nectar and/or pollen. The pollen carrying efficiency evaluated by body washings of captured insects indicated that the honey bees were more efficient in carrying pollen than the small carpenter bees and butterflies; the average number of recorded pollen grains varied from 71.3 to 58.8 for honey bees and 27.5 for small carpenter bees, and from 19.1 to 12.4 for butterflies (Table 3). The insects foraged the flowers in quick succession from one flower to another, in the same and/or different plant populations, in order to collect as much pollen and/or nectar as possible; this inter-plant foraging activity was considered to be promoting cross-pollination.

Fruiting ecology: The pollinated and fertilized flowers grow continually and produce fruits in three weeks. The natural fruit set is 87.90% and the seed set is 68.25%. The fruit is partly enclosed by the calyx. It is a capsule, initially yellow-green and brown when mature; it is stalked, non-fleshy, non-hairy, 3–4 mm long and 2–4 mm wide. A single fruit produces one to four seeds and, accordingly, the fruit shape varies from globose to subglobose; 1-seeded fruit set is 3% and 2-seeded – 22%, 3-seeded – 36%, and 4-seeded – 39%. The seeds are brownish-black, smooth, reniform, glabrous and 2–3 mm in diameter. The fruits dehisce loculicidally to disperse seeds. The dispersed seeds fall to the ground quickly due to the low height of the mother

 Table 3. Pollen recorded in the body washings of insect foragers on *E. alsinoides* and *E. nummularius*.

		<i>E. alsinoides</i> Number of pollen grains			E. nummularius Number of pollen grains		
	Sample						
Insect species	(N)	Range	Mean	S.D	Range	Mean	S.D
Apis cerana	10	28-113	71.3	24.33	37-142	90.8	28.82
Apis florea	10	34-80	58.8	13.94	28-93	56.4	20.10
Trigona iridipennis	10	-	-	-	10-47	25.6	9.84
<i>Ceratina</i> sp.	10	16-43	27.5	9.15	-	-	-
Zizeeria karsandra	10	10-28	19.1	5.62	13-28	20.5	4.65
Zizina otis	10	11-25	18.5	4.05	11-25	15.6	5.08
Chilades laius	10	7-21	12.4	4.58	8-25	14.5	5.74
Chilades pandava	10	9-25	13.9	5.04	10-31	20.6	7.48
Euchrysops cnejus	10	-	-	_	6-23	13.4	4.36

plant. They are further dispersed by wind and rainwater. The seeds are dormant and germinate only during rainy season, which starts in June. Erratic rainfalls and long dry spells during rainy season were found to be terminating the growth and development of seedlings. The old plants with their robust underground root system grow continually and sequentially display phenological events. However, the aerial plants wither away under extreme dry soil conditions occurring during the dry season only.

Evolvulus nummularius (L.) L.

Phenology: Evolvulus nummularius is a weed of sandy, open dry localities with variable soil moisture content. It is an annual prostrate, creeping herb (Fig. 2a). It roots at the nodes and produces extensive branching. It is multi-stemmed and propagates through seeds, as well as vegetatively. The leaves are orbicular, with retuse apex, a rounded or slightly cordate base, and are alternately arranged on the stems. The plant re-grows from the perennial ground rootstock during rainy season. Individual plants occurring close to each other form expanded mats with overlapping stems due to their profuse growth. The plant is conspicuous during flowering period. The flowering occurs early in individuals emerging from rootstock, and late in those emerging from seed. The plants, which grow from the old rootstock, flower in July, and those growing from seed in August. Duration of flowering and life of the plant depends on the soil moisture. Thus, flowering is profuse in July-October, when the soil is sufficiently wet due to heavy rainfalls. If the soil is sufficiently wet, even in the summer season, the plant survives and produces flowers depending on the soil nutrient status. In such plants, the flowering occurs across the year. The flowers are pedicellate, solitary, densely villous and borne in leaf axils. They look quite distinct against the foliage. The plant with its extensively spreading branches and scattered solitary flowers is quite attractive during flowering season (Fig. 2b).

Flower morphology: The flowers are small (8.3 ± 0.6 mm long, 9.5 ± 0.4 mm wide), white, campanulate, odourless, actinomorphic and bisexual. The calyx has five free sepals partially fused at the base; the sepals are green, densely hairy, lanceolate, acuminate, 3.4 ± 0.4 mm long and 3 mm wide. The corolla consists of five united white petals forming a short tube at the base, 8-11 mm wide and glabrous. The stamens are white, epipetalous, exerted, glabrous, 3.4 ± 0.4 mm long;



Fig. 2. Evolvulus nummularius: **a** & **b** – habit with flowering phase; **c** – maturing bud; **d** & **e** – flowers; **f** – stamens; **g** – pollen grain; **h** – Gynoecium; **i** – ovary; **j** – ovules; **k** – 3-seeded fruit; **l** – 4-seeded fruit; **m** – Apis cerana collecting nectar; **n** – Apis florea collecting pollen; **p** – Zizeeria karsandra collecting nectar; **q** – Chilades pandava collecting nectar.

they are dimorphic (3+2) due to insertion of their filaments into the middle of the corolla tube at different lengths (Fig. 2f). The anthers are white, glabrous, 1 mm long and dithecous. The ovary $(6.4\pm0.4 \text{ mm long})$ is green and bicarpellary, bilocular, with two ovules arranged on axile placentation in each locule. The styles and stigmas are pure white; the styles are two in number, short and each style extends into a long 2-lobed minutely capitate stigma (Fig. 2h-j).

Floral biology: Mature buds open completely at 07:00-08:00 h on clear sunny days and partially at 07:30-08:30 h on rainy days (Fig. 2c-e). In mature buds, the corolla is slightly twisted and attains subrotate shape after complete opening. The anthers dehisce by longitudinal slits during anthesis and the pollen is presented latrosely (the split on the side of the anther is positioned towards the other anthers rather than towards the inside or outside of the flower). The number of pollen grains per anther is 885.4±46.96 and per flower is 4427±234.81. The pollen-ovule ratio is 1106.75:1. The pollen grains are white, initially sticky, later powdery, spherical, 6-zonocolpate, 35.69±5.31 µm, and with smooth exine (Fig. 2g). In vitro pollen germination tests indicated that pollen grains are viable and germinate after anther dehiscence and remain viable until 16:00 h, with a gradual decrease in germination rate (Table 1). The stigma becomes receptive thirty minutes after anthesis and remains so until 17:00 h of the same day. The stigma is shiny, dry and non-papillate during receptive phase. Nectary situated at the base of the flower secretes nectar in minute quantity during mature bud stage and

is exposed upon anthesis. Nectar is present in traces in open flowers, due to feeding by thrips since the beginning of anthesis. Thrips used the growing buds for breeding and emerge when the buds open; they continually feed on both nectar and pollen. The corolla, along with the stamens and receptive stigma, closes back by 12:00 h on the same day. The pollinated and fertilized flowers remain in place, while in unpollinated flowers the entire flower falls off on the second day.

Insect visitors and pollination: Thrips were found to be contributing to primary self-pollination by feeding on both pollen and nectar after anthesis. The flowers were foraged by honey bees, stingless bees and lycaenid butterflies at daytime, from 08:00 to 11:00 h, with concentrated foraging activity at 09:00 h (Fig. 4). The bees included *Apis cerana* (Fig. 2m), *A. florea* (Fig. 2n,o) (honey bees) and *Trigona iridipennis* (stingless bees), while the lycaenid butterflies included *Zizeeria karsandra* (Fig. 2p), *Zizina otis, Chilades laius, Chilades pandava* (Fig. 2q), and *Euchrysops cnejus* (Table 2). Of the total foraging visits recorded during the observation period,



Fig. 4. Hourly foraging activity of insect foragers on *Evolvulus nummularius*.

bees accounted for 55% and lycaenids for 45%. The bees approached the flowers in upright position, landed on the corolla and probed for pollen and/or nectar. To collect nectar, they inserted their hairy tongue (glossa) into the corolla throat to access the available nectar; in so doing, the ventral as well as dorsal side brushed against the dehisced anthers and stigmas effecting both sternotribic and nototribic pollination. To collect pollen, the bees approached individual anthers and then only the ventral side and stigmas brushed against the anthers effecting only sternotribic pollination. The butterflies approached the flowers with head facing the corolla throat; then they inserted their proboscis to collect nectar, during which their head and ventral side brushed against the anthers and stigmas effecting sternotribic pollination. The bees and butterflies visited 12-14 flowers consecutively and in some cases up to 20-23 flowers, depending on the flower density, before leaving the flowering patch. As thrips were using nectar, the insects made multiple visits to the same flowers in quest of nectar and/or pollen. The pollen carrying efficiency evaluated by body washings of captured insects indicated that honey bees were more efficient in carrying pollen than stingless bees and butterflies; the average number of recorded pollen grains varied from 90.8 to 56.4 in case of honey bees and 25.6 in case of stingless bees and from 20.6 to 13.4 in case of butterflies (Table 3). The insects foraged the flowers in quick succession, from one flower to the other, in the same and/or different plant populations, in order to collect as much pollen and/or nectar as possible; this inter-plant foraging activity was considered to be promoting cross-pollination.

Fruiting ecology: The pollinated and fertilized flowers grow continually and produce fruits in three weeks. The natural fruit set is 85.74% and the seed set is 58.85%. The fruit is partly enclosed by the calyx. It is a capsule, initially yellow-green and brown when mature; it is stalked, non-fleshy, non-hairy, 3.4±0.4 mm long and 4.5±0.4 mm wide. Each fruit produces one to four seeds and, accordingly, the fruit shape varies from globose to subglobose; 1-seeded fruit set is 9%, 2-seeded is 34%, 3-seeded is 31% (Fig. 2k), and 4-seeded is 26% (Fig. 21). The seeds are brownish-black, smooth, ovoidtrigonous, glabrous, minutely tuberculate, and 1-2 mm in diameter. The fruits dehisce loculicidally to disperse seeds. The dispersed seeds fall to the ground quickly, due to the low height of the mother plant. They disperse further by wind and rainwater. The seeds are dormant and germinate only during rainy season, which starts in

June. Erratic rainfalls and long dry spells during rainy season were found to be terminating the growth and development of seedlings. The old plants with their robust underground root system grow continually and display sequentially phenological events. However, the aerial plants wither away during extremely dry soil conditions, which occur in the dry season only.

Discussion

This study indicates that *Evolvulus alsinoides* and *E. nummularius* display similar phenology in vegetative, flowering and fruiting events. In these species, the flowers stand out above the foliage and look attractive, especially in July–October, when flowering is very intense. Allard (1947) stated that *E. alsinoides* is easy to spot in the field with its twining habit and large brightly coloured and funnel-shaped flowers, with twisted corolla and clockwise in bud, and strongly plicate. In this study, it has been found that *E. nummularius* is also easy to spot in the field, since it is also similar in habit and floral characters.

Sarma & al. (2007) noted that a small percentage of pollen is sterile and fertile pollen grains are viable for about five hours only in E. nummularius. The present study showed that all pollen is fertile and viable for ten hours in E. alsinoides and nine hours in E. nummularius. Sarma & al. (2007) reported that E. nummularius is facultatively autogamous. But, in the present study, the pollen-ovule ratios and the spatial separation of stamens and styles delaying the occurrence of autonomous autogamy until the close-back of the flowers in both E. nummularius and E. alsinoides suggest that these species are facultatively xenogamous. Insects pollinate the flowers when the latter are in open state. The flowers resort to spontaneous autogamy during their closing time (at noon) due to contact between receptive stigmas and anthers. The extended period of stigma receptivity after the closure of the flower increases the chances for occurrence of spontaneous autogamy. In this breeding system, autogamy is delayed to promote cross-pollination, but in reality, both geitonogamy and xenogamy occur due to insect activity during the open state of flowers. Delayed autogamy brings an advantage for the plant, because it does not reduce the amount of pollen that could have been exported for outcross siring, nor does it cause seed discounting by reducing the number of ovules available for outcross

fertilization (Lloyd 1992). It also is a fitness benefit in habitats, where there is much spatial and/or temporal variation in the availability of pollen vectors (Morgan 2006). This means that when the pollinator availability is constant and adequate, plants can maximize their out-crossed seed set and then use delayed autogamy to fertilize any remaining ovules, while if pollinators are absent, the ovules may still be fertilized by autogamy and still retain a high relative fitness, if inbreeding depression is low (Pannell 2006). In a review on understanding plant diversity, Barrett (2010) explained why selfing evolves and what the advantages are. Selfing individuals have the advantage over outcrossers, when pollinators or mates are scarce. They also have a genetic transmission advtange through pollen that selfing variants experience, because selfers are both maternal and paternal parents of the seed they produce. These advantages confer reproductive assurance to the plants that have the ability to reproduce through selfing and outcrossing in the habitats, or in times when pollinators are scarce. In this study, the facultative xenogamous breeding system functional in E. alsinoides and E. nummularius is substantiated by the highest fruit and seed set in open pollinations. However, the seed set rate is significantly low, as compared to the fruit set rate in both species. This state of the seed set rate could be attributable to the free styles and stigmas, due to which their contact with the pollinators during flower life and closing time of the flower is not absolute; such situation would reflect on the fertilization rate of ovules of individual flowers, which characteristically produce four ovules. Therefore, it can be said that delayed autogamy in these species facilitates the production of 3-4 seeds per fruit, mostly from open pollinations and 1–2 seeds per fruit from selfing due to flower closure.

In their report on the reproductive biology of *E. alsinoides*, Singh & al. (2010) have not mentioned whether this species produces nectar or not. Sarma & al. (2007) mentioned that *E. nummularius* does not produce nectar. The present study shows that both species produce nectar. Thrips feeding on nectar prior to the commencement of stigma receptivity prevents autogamy, but facilitates self-pollination after anthesis. However, self-pollination by thrips is limited by the divergent stamens situated far away from the styles and stigmas. Pollen feeding by thrips indirectly promotes flower visitation rate by pollen-collecting bees. Traces of nectar present in the flowers of both species promote flower visiting rate by nectar-feeding bees and

butterflies and, in effect, cross-pollination rate is enhanced. This finding is in agreement with a note by Hodges (1995) that overabundance of nectar may have a detrimental effect on the seed set by increasing the intra-plant pollinator movement.

On the basis of reports by various authors, Veronica et al. (2009) stated that the study of factors that influence the adjustments of floral traits among the species is a central issue in comparative plant ecology. In this context, these authors noted that many floral traits are related to the pollination and fertilization processes. Floral traits displayed by plants are products of selection to ensure reproductive success and, hence, collectively as components of a floral integrated design to maximize pollination and subsequently fertilization rate. Floral integrated designs vary and show adaptations for different classes of pollinators. Austin (1997) reported that pollination in Convolvulaceae is primarily effected by bees, but there are instances of pollination by moths, birds and bats. Singh & al. (2010) reported that E. alsinoides is pollinated primarily by honey bees and beetles, although other insects also pollinate this plant. Sarma & al. (2007) reported that E. nummularius is pollinated by the honey bee, Apis cerana, and the snail, Lamellaxis gracile. The present study indicates that snails have not been found in the habitats of E. alsinoides and E. nummularius; hence, snail-pollination is totally ruled out. The funnel-shaped blue flowers with white throats in E. alsinoides, and the campanulate, white flowers in E. nummularius, displaying the stamens and stigmas directly to the sunlight, project out against the foliage and their attraction is amplified by the patchy form of the growth habit. Despite such an abundance of flowers, especially during rainy season, the flowers are pollinated by a few bee and butterfly species only. Therefore, the pollinator guilds recorded in the study indicate that both species of Evolvulus are principally melittophilous and psychophilous.

Saharan & al. (2001) and Todd & al. (2002) stated that seeds of *E. alsinoides* are physically dormant. In this study, both *E. alsinoides* and *E. nummularius* display seed dormancy by germinating only during the rainy season. This dormancy enables these species to colonize the areas during rainy season, though erratic and long dry spells during the rainy season exercise an impact on the seedling establishment rate. However, re-growth from the well-established old rootstock withstands rain deficit and produces new plants, thus alleviating to some extent the loss of seedlings from seeds. Therefore, seed dormancy and production of new plants from the old rootstock enable the plant to occupy various habitats and to extend and expand its distribution range. In this context, it is appropriate to mention that the fruit capsule dehisces loculicidally facilitating seed dispersal to different distances on the ground (ballistochory) and the smooth and reniform seeds subsequently disperse by wind and rain water which is characteristic for anemochory and hydrochory. These three modes of seed dispersal benefit the plant in invading and colonizing new areas.

Knapp & Rice (1996) noted that there is a widespread interest in native herbs, partly due to recent availability of plant material, as well as to recognition of the role of native herbal species in restoration of biological diversity. The present study suggests that *Evolvulus* species are promising candidates for restoration of the ecological niches, where they successfully grow and colonize. With their clustered root system and spreading form of multi-stemmed branching pattern they are effective soil binders. Therefore, these species are important herbs in the natural and artificial restoration of habitats, which are either destroyed or degraded, or damaged.

References

- Allard, H.A. 1947. The direction of twist of the corolla in the bud and twining of the stems in *Convolvulaceae* and *Dioscoreaceae*. – Castanea, 12: 88-94.
- Austin, D.F. 1997. Dissolution of *Ipomoea* ser. Anisomerae (Convolvulaceae). J. Tor. Bot. Soc., **124**: 140-159.
- Austin, D.F. 2010. Baboquivari Mountain Plants: Identification, Ecology and Ethnobotany. Univ. Arizona Press, Tucson, p. 350.
- Barrett, S.C.H. 2010. Understanding plant reproductive diversity. Phil. Trans. R. Soc. B., 365: 99-109.
- Collevatti, R.G., Campos, L.A.O. & Da Silva, A.F. 1998. Pollination ecology of the tropical weed *Triumfetta semitriloba* Jacq. (*Tiliaceae*), in the Southeast Brazil. – Revista Brasil. Biol., **58**: 383-392.
- Cruden, R.W. 1977. Pollen-ovule ratios: a conservative indicator of breeding systems in flowering plants. – Evolution, 31: 32-46.
- Dafni, A., Kevan, P.G. & Husband, B.C. 2005. Practical Pollination Biology. Enviroquest, Ltd., Cambridge, p. 590.
- Dowarah, J., Boruah, H.P.D., Gogoi, J., Pathak, N., Saikia, N. & Handique, A.K. 2009. Eco-restoration of a high-sulphur coal mine overburden dumping site in northeast India: a case study. – J. Earth Syst. Sci., **118**: 597-608.
- Gebrekirstos, A., Teketay, D., Fetene, M. & Mitlohner, R. 2006. Adaptation of five co-occurring tree and shrub species to water

stress and its implication in restoration of degraded lands. – Forest Ecol. Manag., **229**: 259-267.

- Hodges, S.A. 1995. The influence of nectar production on hawkmoth behavior, self-pollination, and seed production in *Mirabilis multiflora* (Nyctaginaceae). – Am. J. Bot., **82**: 197-204.
- Junqueira, M.E.R. & Smao-Bianchini, R. 2006. O genero Evolvulus L. (Convolvulaceae) no municipio de Morro do Chapeu, BA, Brasil. – Acta Bot. Brasil., 20: 152-172.
- Ketjarun, K., Staples, G.W., Swangpol, S.C. & Traiperm, P. 2016. Micro-morphological study of *Evolvulus* spp. (Convolvulaceae): the old world medicinal plants. – Bot. Stud., 57: 1-11.
- Knapp, E.E. & Rice, K.J. 1996. Genetic structure and gene flow in *Elymus glaucus* (Blue Wildrye): implications for native grassland restoration. – Restor. Ecol., 4: 1-10.
- Kumar, R., Ambasht, R.S., Srivastava, A., Srivastava, N.K. & Sinha, A. 1997. Reduction of nitrogen losses through erosion by *Leonotis nepetaefolia* and *Sida acuta* in simulated rain intensities. – Ecol. Engg., 8: 233-239.
- Kunte, K. 2007. A lifescape of Butterflies of Peninsular India. Univ. Press, Hyderabad, p. 254.
- Lloyd, D.G. 1992. Evolutionarily stable strategies of reproduction in plants: who benefits and how? – In: Wyatt, R. (ed.), Ecology and Evolution of Plant Reproduction: New Approaches, Chapman and Hall, New York, 137-168.
- Morgan, M.T. 2006. Selection on reproductive characters: conceptual foundations and their extension to pollinator interactions. – In: Harder, L.D. & Barrett, S.C.H. (eds), Ecology and Evolution of Flowers, Oxford Univ. Press, Oxford, 25-40.
- Okuyama, T. & Holland, J.N. 2008. Network structural properties mediate the stability of mutualistic communities. – Ecol. Lett., 11: 208-216.
- Pannell, J.R. 2006. Effects of colonization and metapopulation dynamics on the evolution of plant sexual systems. – In: Harder, L.D. & Barrett, S.C.H. (eds), Ecology and Evolution of Flowers, Oxford Univ. Press, Oxford, 223-238.
- Saharan, P., Kasera, P.K. & Chawan, D.D. 2001. Seed polymorphism and germination behavior of Shankhpushpi (*Evolvulus alsinoides*). Ann. Arid Zone, 40: 97-99.
- Sarma, K., Tandon, R., Shivanna, K.R. & Mohan Ram, H.Y. 2007. Snail-pollination in Volvulopsis nummularium. – Curr. Sci., 93: 826-831.
- Schmelzer, G.H. & Bunyapraphatsara, N. 2001. Evolvulus alsinoides (L.) L. – In: van Valkenburg, J.L.C.H. & Bunyaprabhatsara, N. (eds), Plant Resources of South-East Asia No. 12: Medicinal and Poisonous Plants 2. Backhuys Publ., Leiden, Netherlands, 258-260.
- Singh, K.P., Bhavana & Dhakre, G. 2010. Reproductive biology of Evolvulus alsinoides L. (Medicinal Herb). – Intl. J. Bot., 6: 304-309.
- Todd, B.L., Coons, J.M. & Owen, H.R. 2002. Scarification technique affects germination of *Stylisma pickeringii* (Patterson Bindweed), an Illinois endangered plant. – Am. Midland Nat., 148: 190-192.
- Veronica, A.F., Leonardo, G. & Julia, A. 2009. Influence of flower functionality and pollination system on the pollen size-pistil length relationship. – Organisms Diversity & Evolution, 9: 75-82.