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TAXONOMY AND REPRODUCTIVE BIOLOGY OF THE GENUS *ZEPHYRANTHES* HERB. (LILIACEAE) IN BANGLADESH

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Keywords: Zephyranthes Herb.; Pollination; Seed germination; Pseudovivipary; Pollen viability.

Abstract

The genus *Zephyranthes* Herb. is revised along with its pollination mechanism, seed germination and vegetative propagation. Detailed taxonomy of four *Zephyranthes* species occurring in Bangladesh, namely, *Z. atamasco* (L.) Herb., *Z. candida* (Lindl.) Herb., *Z. carinata* Herb. and *Z. tubispatha* (L'Her.) Herb. *ex* Traub. was studied with their updated nomenclature, important synonyms, phenology, specimens examined, habitat, distribution, economic value and mode of propagation. A dichotomous bracketed key is provided for easy identification of the species. Pollination investigation reveals that all studied species of *Zephyranthes* are self-pollinated. Minimum five days were required for germination of seeds in *Z. atamasco*, and three days each in *Z. candida*, *Z. carinata* and *Z. tubispatha*. Pseudovivipary type of germination has been reported in *Z. candida* and *Z. carinata* for the first time. The maximum number of seeds (30) per fruit are produced in *Z. tubispatha*, whereas the minimum seeds (2) per fruit are found in *Z. atamasco*. Vegetative propagation through bulb was found more suitable than seeds in *Z. atamasco*, *Z. candida* and *Z. carinata*. Pollen viability has been found 100% in *Z. candida*, and *Z. tubispatha*, whereas, *Z. atamasco* and *Z. carinata* have shown 80% and 98% viability, respectively.

Introduction

The genus *Zephyranthes* Herb. (Liliaceae) comprises about 70 species and native to diverse areas of the New World including Argentina, the Caribbean, Mexico and North America (Chowdhury and Hubstenberger, 2006; Spurrier *et al*., 2015). In Bangladesh, this genus is represented by four species and found under cultivation. *Zephyranthes* are characterized by linear or lorate leaves, solitary flower, funnel shaped perianth, three carpels and sub-globose or depressed fruits. Pharmacological studies of *Zephyranthes* have revealed that the genus has anticancer, antifungal, and antibacterial activities (Katoch and Singh, 2015). Leaf decoction of *Z. candida* is used in South Africa as a remedy for diabetes mellitus (Pettit *et al*., 1984). Several studies on the genus *Zephyranthes* were carried out based on morphology (Spencer, 1973; Flagg and Flory, 1976; Flagg *et al*., 2002). Recently, Flagg and Smith (2008) studied three closely related species of *Zephyranthes*, i.e. *Z. atamasca* (the correct specific epithet is *atamasco*), *Z. treatiae*, and *Z. simpsonii* from southern United States, all of which have linear stigmatic lobes, green perianth tubes and white perianth segments. Based on cytological, herbarium, and field studies, and on Principal component analysis (PCA) and scatter diagram analysis, they concluded that all three taxa are distinct at the species level. Raina and Khoshoo (1972) studied cytogenetics of *Z. candida* and *Z. sulphurea*, while the breeding system of *Z. atamasco* was investigated by Broyles and Wyatt (1991).

Studies on reproductive biology disclose the nature of species, adaptation, speciation, hybridization, and systematics (Anderson *et al*., 2002; Neal and Anderson, 2005). Several studies

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on reproductive biology and pollination mechanism have been conceded in different group of plants (Cox, 1990; Wyatt and Broyles, 1990; Singer and Sazima, 1999; Liza *et al*., 2010). Studies on seed germination in different plants are also well documented, and factors affecting seed germination have been recognized in different species (Yang *et al.*, 1999; Hassan and Fardous, 2003; Chauhan and Johnson, 2008; Rahman *et al*., 2012; Ferdousi *et al*., 2014).

Despite the systematic studies of *Zephyranthes* were carried out in different countries (Flagg *et al*., 2002; Flagg and Smith, 2008; Arroyo-Leuenberger and Leuenberger, 2009) there has been no detailed study on taxonomy of this genus occurring in Bangladesh. Pollination, seed germination and propagation of *Zephyranthes* have never been investigated. Because of medicinal and ornamental value, the members of this genus need to be brought under cultivation, and prior to bring them under cultivation their reproductive biology need to be investigated in detail. The objectives of the present study are to revising the genus *Zephyranthes* and to investigate reproductive biological characteristics including mode of pollination and seed germination of these ornamental and economically important species, which might help in conveying the plants under rapid cultivation.

Materials and Methods

Plant materials

Four species of *Zephyranthes* namely, *Z. atamasco*, *Z. candida*, *Z. carinata* and *Z. tubispatha* were collected from different places and planted in the Dhaka University Botanical Garden for further study. The collected specimens were critically examined, and the study was supplemented by the herbarium specimens preserved at the Dhaka University Salar Khan Herbarium (DUSH) and Bangladesh National Herbarium (DACB). Identification of the *Zephyranthes* species were confirmed in consultation with standard literature (Karthikeyan *et al*., 1989; Noltie, 1994; Hajra and Verma, 1996; Raven and Zhengyi, 2000; Utech, 2002; Siddiqui *et al.*, 2007) and by comparing with herbarium specimens deposited in DUSH and DACB. A dichotomous key to the species has been constructed for easy identification of the taxa. The voucher specimens have been deposited at DUSH.

Pollination

To study pollination the species were meticulously observed after flowering. Bagging experiment was conducted to understand the mechanism of pollination (Hassan and Khan, 1996). Bags of 15x12 cm made from fine cloth were used for bagging the flowers of individual plant at a stage, when all flowers were unopened. During bagging if open flowers exist, they were removed manually so that only unopened buds remained in the bags. Bagged inflorescence or flowers were kept under continuous observation for fruit formation which was compared with that of the plants kept under control.

Seed germination

Seeds were collected from mature fruits and sown in earthen pots of 10 inch in diameter filled up with a mixture of soil and compost (2:1). Seeds were sown at regular intervals in the earthen pot and the pots were kept in the semi-shaded position and watered everyday. Seeds were sown in different pots in different times of the year to record dormancy (if any), suitable period for germination, percentage and nature of germination.

Vegetative propagation

Vegetative propagation was performed through bulbs, separated from main plants.

Pollen viability

Pollen was taken from recently opened flower anthers. A drop of acetocarmine was taken onto the slide. After removing the anther from the flower pollen was touched into the acetocarmine placed on a slide and observed under light microscope. Viable pollen takes acetocarmine and the shapes are regular, whereas non-viable pollen remains non-coloured with acetocarmine and were very irregular in shape.

Results

Taxonomic treatment

Zephyranthes Herb., App. [Bot. Reg.]: 36 (1821).

Argyropsis M. Roem., Syn. Ensat. : 125 (1847); *Arviela* Salisb., Gen. Pl. Fragm.: 135 (1866); *Habranthus* Herb., Bot. Mag.: t. 2464 (1824); *Mesochloa*, *Plectronema* and *Pogonema* Rafin., Fl. Tell. 4: 10 (1836); *Pyrolirion* Herb., App. [Bot. Reg.]: 37 (1821).

Small herbs with tunicate bulbs. Leaves simple, linear or lorate, appearing with or after the flowers. Flowers solitary, usually at the top of the long scape. Perianth funnel-shaped, tube short or long, dilated upward; tepals 6, rarely up to 8, in 2 series of 3 each, united at the base. Stamens 6, rarely up to 8, adnate to the perianth base; filaments long; anthers linear, dorsifixed. Carpels 3, united, ovary 3-celled, ovules many; style filiform; stigma 3-lobed. Fruit a capsule, sub-globose or depressed, loculicidally 3-valved. Seeds oblong, black.

Key to species of *Zephyranthes*

1.	Leaves terete; spathe covering the ovary	*Z. candida*
-	Leaves flat; spathe not covering the ovary	2
2.	Spathe not 2-fid; flowers yellow	*Z. tubispatha*
-	Spathe 2-fid; flowers pink or white	3
3.	Outer 3 tepals obtuse, pink in colour	*Z. carinata*
-	Outer 3 tepals acute, white but turn pink at maturity	*Z. atamasco*

Zephyranthes atamasco (L.) Herb., App. Reg. 36 (1821); Utech, Fl. North. America 26: 298 (2002). *Amaryllis atamasco* L., Sp. Pl. 1: 292 (1753); *A. atamasco* Blanco, Fl. Filip. : 254 (1837). (**Figs 1 & 5A**).

English names: Atamasco Lily, Fairy Lily, Rain Lily, Easter Lily, Zephyr Lily.

Local name: *Sada Ghashphul*.

A perennial bulbous herb, bulb ovoid, c. 2.5 cm in diam., neck 2.5–5.0 cm long. Leaves linear, up to 15 cm long, bright green. Flowers solitary, terminal, bisexual; peduncle c. 21 cm long, hollow. Spathe simple, c. 3.0×0.6 cm, hyaline, tubular, 2-notched. Perianth segments 6, c. 5×2 cm, arranged in two rows, inner 3 smaller than the outer 3, white but lower 2 green in colour, changes from pure white to pink at maturity. Stamens 6, outer 3 large, c. 2.5 cm long, inner 3 small, c. 1.7 cm long; anthers linear, yellowish-orange. Carpels 3, united; ovary inferior, c. 0.4 cm long, 3-celled, placentation axile; style slender; stigma 3-notched, c. 4 cm long. Fruit a capsule, subglobose.

Flowering and fruiting: April to May.

Specimens examined: Dhaka: Dhaka University Botanical Garden, 11.4.2007, Sumona 21 (DUSH); 10.9.2013, Sumona 85 (DUSH).

Chromosome number: 2n = 12, 24 (Kumar and Subramaniam, 1986).

Habitat: Cultivated in gardens.

Distribution: Native to south-east America, naturalized in southern North America (Wade *et. al.*, 2014),widely cultivated in many countries including Bangladesh.

Uses: Cultivated as an ornamental plant in gardens. All parts are toxic especially bulb, may be fatal if eaten (Kates *et al.*, 1980).

Propagation: Through bulbs and seeds.

Zephyranthes candida (Lindl.) Herb., Bot. Mag. 53: t. 2607 (1826); Hajra & Verma, Fl. Sik. M. 1: 138 (1996); Raven & Zhengyi, Fl. China 24: 265 (2000); Utech, Fl. North America 26: 302 (2002). Hassan, Encycl. Flora & Fauna of Bangladesh 11: 351 (2007). *Amaryllis candida* Lindl., Bot. Reg. 9: t. 724 (1823). (**Figs 2 & 5B)**.

English name: Fairy Lily.

Local name: *Sada Ghashphul.*

A perennial clump-forming bulbous herb, bulb tunicated, ovoid, c. 2.5 cm in diam., neck 2.5–5.0 cm long. Leaves simple, terete, linear, up to 35 cm long and 0.5 cm in diam., hollow, obtuse, glabrous, dark green. Inflorescence solitary on terminal leafless scape. Flowers bisexual, incomplete, actinomorphic, epigynous, white; peduncle c. 26 cm long; spathe like bract present at the top of a long scape covered the ovary; bract c. 3×1 cm, brown in colour, lanceolate, glabrous. Tepals 6, c. 3.7×1.5 cm, free, ovate-lanceolate, white. Stamens 6, free, about half as long as the perianth; anthers c. 0.9 cm long, oblong, dorsifixed, yellow; filaments white, glabrous, more or less as long as anthers. Carpels 3, syncarpous; ovary inferior, c. 0.5×0.3 cm, 3-celled, ovules many; style slender, c. 1.7 cm long with stigma; stigma 3-notched; placentation axile. Fruit a capsule, subglobose, c. 0.8×1.2 cm, yellowish-green, 16–25 seeded. Seeds angular, flattened; testa black.

Flowering and fruiting: August to November.

Specimens examined: Dhaka: Science Library compound, University of Dhaka, 20.9.2007, Sumona 46 (DUSH); Nazrul Institute compound, University of Dhaka, 20.8.2011, Sumona 68 (DUSH).

Chromosome number: 2n = 19, 20, 36, 38, 40, 41, 48, 50 (Kumar and Subramaniam, 1986).

Habitat: Gardens, where it is widely cultivated.

Distribution: Originated from Argentina and Uruguay (Bateman *et al*., 2004). Native to South America, naturalized in South China, cultivated in many countries including Bangladesh (Siddiqui *et al*., 2007).

Uses: Used as an ornamental plant in gardens, containers or as a landscape plant. Bulb contains cytostatic constituents which can be used in the treatment of cancer (Pettit *et al.*, 1990).

Propagation: Through clumps of bulbs and seeds.

Zephyranthes carinata Herb., Bot. Mag. : t. 2594 (1825); Hajra & Verma, Fl. Sik. M. 1: 138 (1996); Raven & Zhengyi, Fl. China 24: 265 (2000); Utech, Fl. North America 26: 299 (2002). *Z. grandiflora* Lindl., Bot. Reg.: t. 902 (1825); Hassan, Encycl. Flora & Fauna of Bangladesh 11: 351 (2007). (**Figs 3 & 5C**).

English names: Pink Rain Lily, Fairy Lily, Zephyr Lily, Pink Storm Lily.

Local names: *Golapi Ghashphul*, *Peyazphul*.

A bulbous, clump forming perennial herb, bulb tunicated, up to 2 cm in diam. Leaves simple, exstipulate, linear, obtuse, entire, glabrous, green, up to 35.0×0.8 cm, appearing with flowers. Inflorescence solitary on terminal leafless scape. Scape c. 18 cm long, light green, produce a single maroon lipstick-like bud on a top. Flowers c. 7.5 cm long, c. 7.5 cm across, spreading, last a

few days, closing up at night. Spathe simple, c. 2.0×0.6 cm, hyaline, tubular, 2-notched. Perianth segments 6, rarely up to 8, funnel-shaped, rose or pink, 2–4 cm long, sub-elliptic to oblong–lanceolate. Stamens 6, sometimes 7–8, adnate to the throat of the perianth; anthers linear, yellow, narrow, dorsifixed; filament up to 2 cm long, white,. Carpels 3, syncarpous, ovary 3–celled, ovules many in each cell; style filiform, c. 2.5 cm long, placentation axile; stigma deeply 3–4 fid. Fruit a capsule, c. 0.5×0.5 cm, dark green, 6–10 seeded. Seeds black.

Flowering and fruiting: June to October. Blooming soon after a heavy rainfall.

Specimens examined: Dhaka: Dhaka University Botanical Garden, 19.9.2007, Sumona 45 (DUSH); Dhaka University Botanical Garden, 24.8.2014, Sumona 92 (DUSH); Nazrul Institute compound, University of Dhaka, 20.8.2012, Sumona 76 (DUSH).

Chromosome number: 2n = 24, 36, 48 (Kumar and Subramaniam, 1986).

Habitat: Well-drained soils.

Distribution: Native of Central America and Mexico, distributed in warmer parts of America, widely cultivated in many countries with a warm climate (Siddiqui *et al*., 2007). In Bangladesh, it is widely grown in many gardens.

Uses: The species is valued as an ornamental plant, along walkway or at the front of a sunny border. In China, bulbs are used to break fever and a paste of the bulb is used for boils. Bulbs possess alkaloids which might be used in the treatment of cancer (Wiart, 2012).

Propagation: Propagated by bulbs or seeds.

Zephyranthes tubispatha Herb., App. Reg.: 36 (1821); Hook. f., Fl. Brit. Ind. 6: 277 (1892); Prain, Beng. Pl. 2: 797 (1903); (L'Her.) Herb. *ex* Traub, Taxon 7: 110 (1958); Hassan, Encycl. Flora & Fauna of Bangladesh 11: 352 (2007). *Amaryllis tubispatha* L'Her., Sert. Angl.: 9 (1789). *Z. nervosa* Herb., Amaryll. : 172 (1837). (**Figs 4 & 5D**).

English names: Zephyr Lily, Fairy Lily, Rain Lily.

Local name: *Holde Ghashphul*.

A small perennial herb with underground tunicated bulb, bulb c. 1.5×1.0 cm, grows singly. Leaves simple, linear, c. 30 cm long and 3 mm broad, green, entire, obtuse, appearing along with the flowers. Flowers solitary, pedunculate; bracts spathe-like, c. 2.3 cm long, situated at the top of a fistular scape, scape up to 28 cm long. Perianth segments 6, connate below, free above, funnel-shaped, c. 3.7 cm long, yellow. Stamens 6; anthers linear, dorsifixed, c. 0.7 cm long, orange, burst longitudinally; filament c. 1.4 cm long. Carpels 3, syncarpous, ovary 3-celled, c. 0.5 cm long, ovules many; placentation axile; style 1, c. 2 cm long, white; stigma 3–lobed, short. Fruit a subglobose capsule, loculicidally 3-valved, yellowish-green, c. 0.8×1.0 cm, 16–20 seeded. Seeds oblong, black, angled.

Flowering and fruiting: June to September.

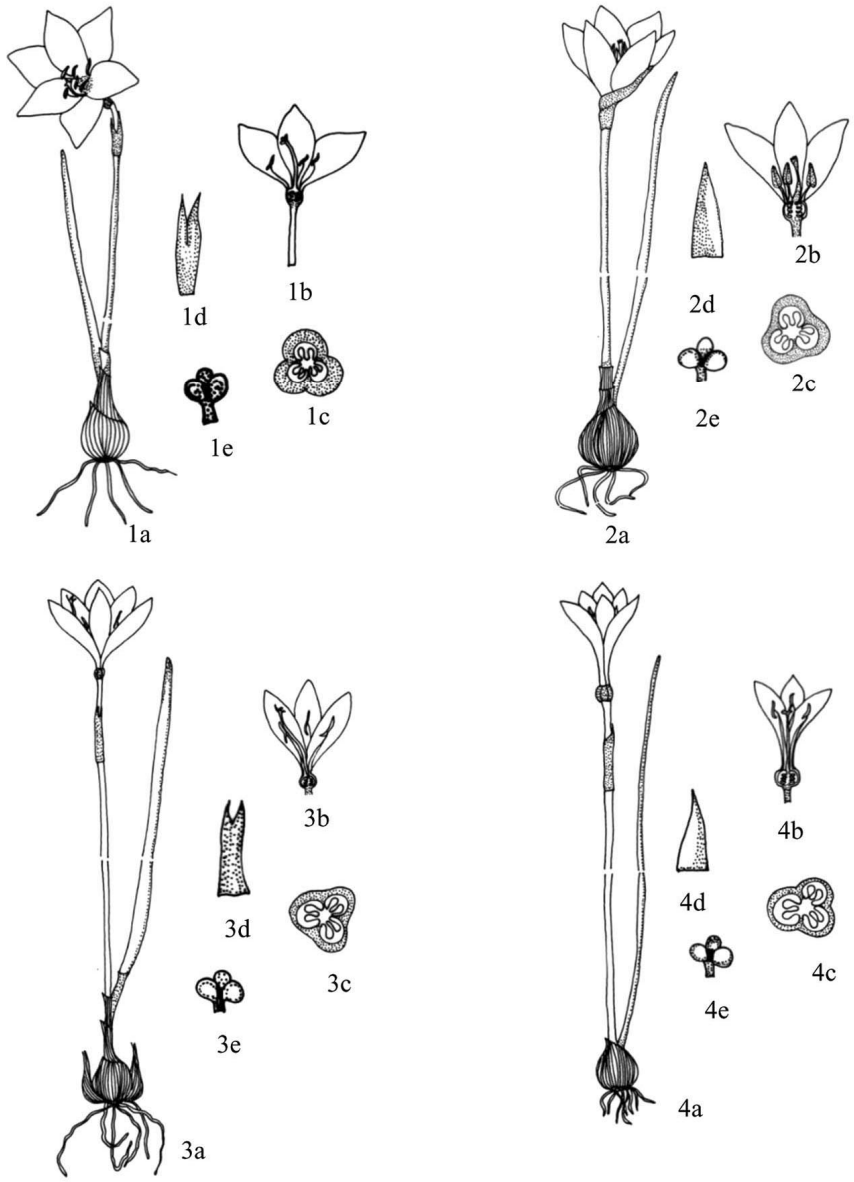
Specimens examined: Dhaka: Dhaka University Botanical Garden, 26.5.2007, Sumona 37 (DUSH); Dhaka University Botanical Garden, 10.4.1968, Mozahar 101 (DUSH); Uttara, Sector-3, 18.8.1998, M. Salar Khan K 10115 (DACB); Dhaka University Botanical Garden, 30.6.1970, A.M. Huq 78 (DACB).

Chromosome number: 2n = 24 (Kumar and Subramaniam, 1986).

Habitat: Well-drained soils and grassy ground of hilly areas.

Distribution: Native of Peru, tropical America and the West Indies (Siddiqui *et al*., 2007). This species is planted in gardens and has been naturalized in many countries including Bangladesh.

Uses: Used as an ornamental plant.
Propagation: By bulbs and seeds.



Figs 1-4. Habit sketch of four *Zephyrnthes* species. 1. *Z. atamasco*: 1a. Habit (×0.3); 1b. L.S. of flower (×0.2); 1c. T.S. of ovary (×2); 1d. Bract (×0.5); 1e. Fruit (×1). 2. *Z. candida*: 2a. Habit (×0.3); 2b. L.S. of flower (×0.2); 2c. T.S. of ovary (×3); 2d. Bract (×0.5); 2e. Fruit (×0.5). 3. *Z. carinata*: 3a. Habit (×0.3); 3b. L.S. of flower (×0.2); 3c. T.S. of ovary (×5); 3d. Bract (×0.5); 3e. Fruit (×0.5). 4. *Z. tubispatha*: 4a. Habit (×0.3); 4b. L.S. of flower (×0.2); 4c. T.S. of ovary (×2); 4d. Bract (×0.5); 4e. Fruit (×0.5).

Fig. 5. Habit of four *Zephyranthes* species: A. *Z. atamasco*; B. *Z. candida*; C. *Z. carinata*; D. *Z. tubispatha*.

Reproductive biology

Reproductive biology study on four *Zephyranthes* species revealed that *Z. atamasco*, *Z. candida*, *Z. carinata* and *Z tubispatha* all are self-pollinated. In *Z. atamasco* and *Z. carinata* single fruit sets each under both bagged and un-bagged condition after five days of bagging and no fruit formation occurs from emasculated flowers. In *Z. candida*, the emasculated flowers do not produce any fruit setting, while both bagged and un-bagged plants have single fruit setting after four days of bagging. Fruit setting starts after three days of bagging in *Z. tubispatha*, and a single fruit setting has been observed both in bagged and un-bagged plants.Seeds were germinated after five days of sowing in *Z. atamasco* and the rate of germination is very low (20%). There is no

dormancy period. In *Z. candida*, three to four days were taken for germination of seeds. The germination rate was 100% when they were sown in August through October indicating that these months are most suitable for seed sowing in this species. Seeds lost complete viability after three months in *Z. candida*. The minimum three days were taken for germination of seeds in *Z. carinata* and the germination rate was 100% when sown in July. After three months of seeds sowing they were not germinated and seeds lost their viability. The study also revealed that after maturation, viability of seeds decreased gradually. The results showed that three to four days were required for germination of seeds in *Z. tubispatha* and the rate of germination was very high when sown just after seed collection. There is no dormancy period in *Z. tubispatha*. Seeds lost their viability after three months. The optimum period of seed germination, minimum days taken for germination and percentage of germination in four *Zephyranthes* species are depicted in Table 1. Different stages of seed germination in *Z. atmasco, Z. candida, Z. carinata* and *Z. tubispatha* are shown in Figure 6.

Table 1. Data on seed germination of four *Zephyranthes* species.

Species	Optimum period of seed germination	Minimum days taken for germination	Percentage of germination	Remark
Z. atamasco	May	5	20	Hypogeal
Z. candida	August-October	3	100	Pseudovivipary
Z. carinata	July	3	100	Pseudovivipary
Z. tubispatha	July-August	3	100	Hypogeal

In the present study, vegetative propagation in *Z. atamasco* and *Z. candida* through bulb has been found effective and more suitable than seeds. Plants propagated from seeds took about three years to bloom, whereas it took around two years from bulb separation. The study revealed that no bulblet was formed in *Z. tubispatha* indicating that seeds are the only means of regeneration in this species. The results showed that propagation through bulb separation took less time than that of seeds. Flower initiation took place through bulb is usually one year earlier than blooming through seeds. A comparative account of reproductive characters of *Zephyranthes* species i.e. time taken for seed germination, scape initiation to first flower, fruit formation after flowering, fruit maturation, number of flowers per scape, number of fruits per scape, number of seeds per fruit, time taken for flowering from seed germinated plants and time taken for flowering from bulb transferred plants are presented in Table 2.

Table 2. Reproductive characteristics of studied four species of *Zephyranthes*.

Species	Time taken for SG	Time taken for SIFF	Time taken for FFAF	Time taken for FMAF	No. of flowers/ scape	No. of fruits/ scape	No. of seeds/ fruit	Time taken for FFSG	Time taken for FFBT
Z. atamasco	5 days	8 days	4 days	7 days	1	1	2-8	3 years	2 years
Z. candida	3-4 days	7 days	3 days	6 days	1	1	6-20	3 years	2 years
Z. carinata	3-4 days	8 days	4 days	7 days	1	1	6-20	3 years	2 years
Z.tubispatha	3-4 days	7 days	3 days	6 days	1	1	8-30	3 years	Not possible

SG=Seed germination; SIFF= Scape initiation to first flowering; FFAF= Fruit formation after flowering; FMAF= Fruit maturation after formation; FFSG= Flowering from seed germination; FFBT= Flowering from bulb transfer.

Z. candida and *Z. carinata* showed pseudovivipary (Fig. 7). Seeds of these species are germinated inside the capsules after a heavy rainfall. In this process of germination, the hypocotyle elongated and came out of the seed forming a loop and developed narrow, straight

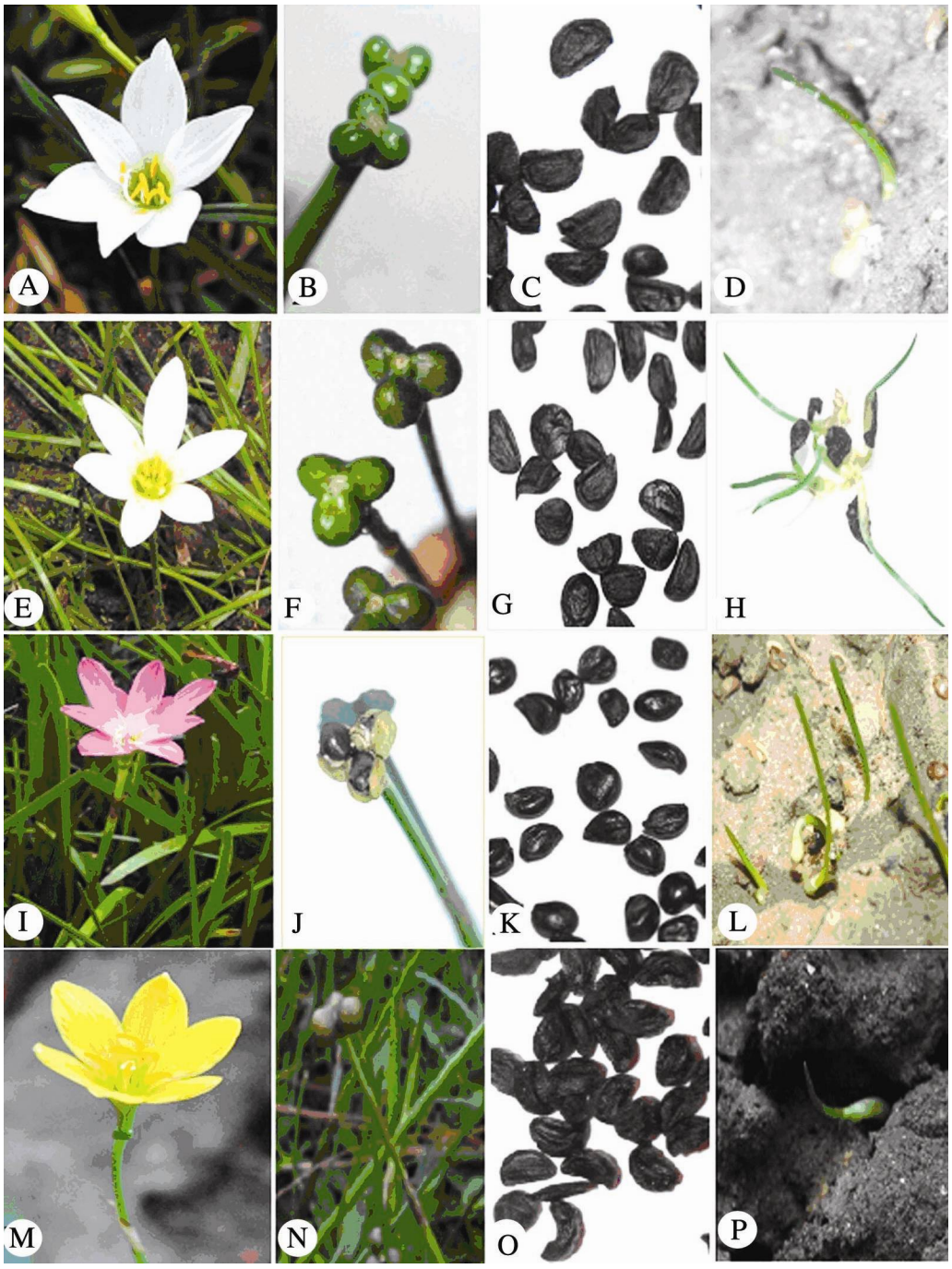


Fig. 6. Different stages of seed germination in four *Zephyraanthes* species: A-D. *Z. atamasco*; E-H. *Z. candida*; I-L. *Z. carinata*; M-P. *Z. tubispatha*; A,E,I&M: Flower; B,F,J&N: Fruits; C,G,K&O: Seeds; D,H,L&P: Seedling.

epicotyle. Hypogeal type of germination has been noticed in these species. Further investigation is needed to explore the mechanism of pseudovivipary in these species.

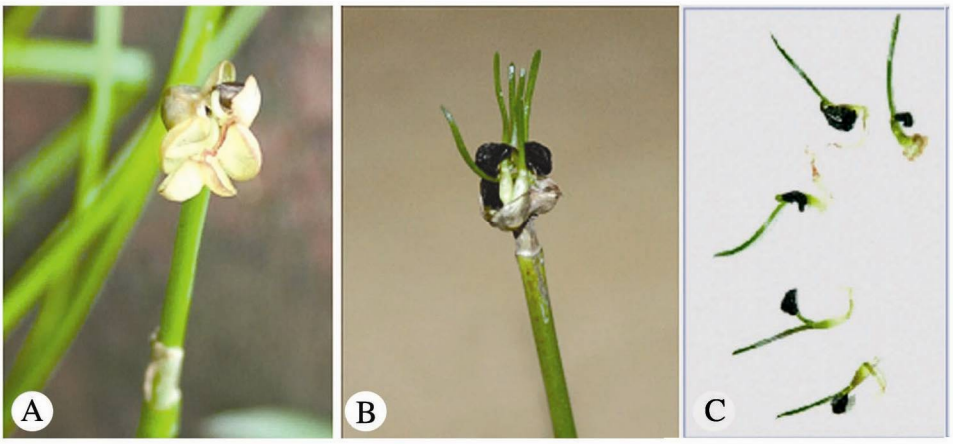


Fig. 7. Pseudovivipary in *Zephyranthes*: A. *Z. carinata*; B&C. *Z. candida*.

Pollen viability was tested in this study because of its importance in reproductive biology, and no seed formation takes place without viable pollen. The present investigation revealed that pollen viability ranged from 80 to 100% among the *Zephyranthes* species. The percentage of pollen viability was found 100% in *Z. candida* and *Z. tubispatha*, whereas, *Z. atamasco* and *Z. carinata* exhibited 80% and 98% viability, respectively. The viable pollens of these species are shown in Figure 8.

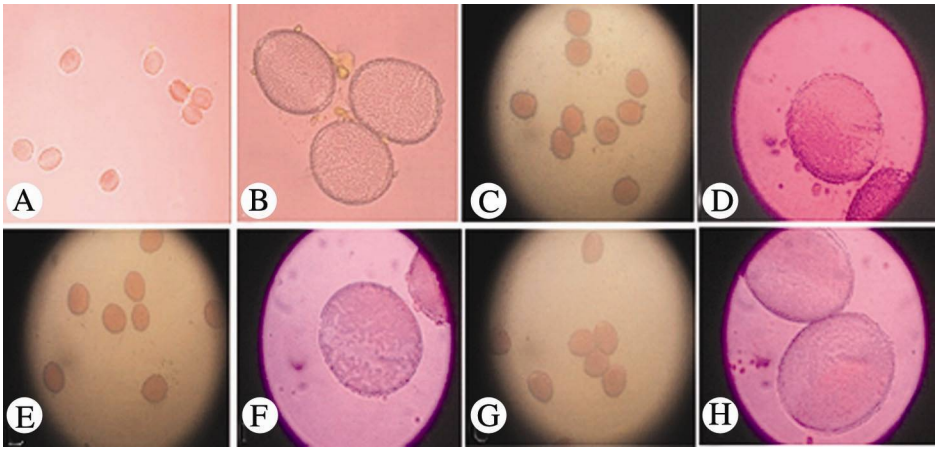


Fig. 8. Pollen viability of four *Zephyranthes* species: A&B. *Z. atamasco* (×10, ×40); C&D. *Z. candida* (×10, ×40); E&F. *Z. carinata* (×10, ×40); G&H. *Z. tubispatha* (×10, ×40).

Discussion

Zephyranthes are remarkable for the wide ecological niche they occupy, from xeric to temporarily flooded conditions, having many coveted ornamental characteristics. Flowers of

Zephyranthes appear in spring through fall after the first rains and they last one to two days, depending on sunlight and temperature, however, new flowers continuously develop for several days (Knox, 2009). In the present investigation we studied floral morphology, phenology pollination, seed germination and vegetative propagation of four *Zephyranthes* species occurring in Bangladesh, *viz*., *Z. atamasco*, *Z. candida*, *Z. carinata* and *Z. tubispatha*. Among them *Z. candida* can easily be distinguished from the remaining species by its terete leaves and spathe covering the ovary.

Previous investigations in *Zephyranthes* have indicated that species with styles that are long relative to the stamens are self-incompatible, whereas species with short styles are self-compatible. Species with styles as long as the stamens may be either self-compatible or self-incompatible (Broyles and Wyatt, 1991). Studies on breeding system of a long-styled *Z. atamasco* revealed that 5%, 78%, and 92% fruit-set occurred in flowers which were bagged, self-pollinated, or cross-pollinated, respectively (Broyles and Wyatt, 1991). In our study, we found that *Z. atamasco* produced fruit setting in bagged and un-bagged plants, while no fruit formation occurred in emasculated flowers. Although it took very short time for germination of seeds, the germination rate is only 20% as observed in *Z. atamasco*. In *Z. candida*, all seeds sown during August to October were germinated only after three to four days of sowing which indicates this period as the most suitable time for seed sowing. Seeds lost their viability after three months in *Z. candida*. All seeds of *Z. carinata* sown in July were germinated, while the rate of seed germination was 100% when they were sown in July and August. In case of *Z. tubispatha* the rate of seed germination was 100% when sown in July and August, while this rate decreased to 80% when seeds were sown in September. None of the seeds was germinated in any of the four species of *Zephyranthes* employed in this study when they were sown after October. This indicates that seeds are not germinated after rainy seasons and they lost their viability in this period.

Pseudovivipary describes plants that produce apomictic or asexual propagules such as bulbils or plantlets in the place of sexual reproductive structures. Species with true vivipary tend to inhabit shallow marine habitats, either in mangrove or in seagrass communities, while pseudovivipary is most prevalent among terrestrial plants occurring in strongly seasonal environments, either growing at high altitudes and latitudes, or in semi-arid to arid areas (Elmqvist and Cox, 1996). All of these habitats are characterized by extraordinarily coarse-grained environments for seedling establishment, even though with major differences in patch size.

Vegetative propagation through pseudovivipary is known from over 100 species of grasses (Poaceae) and this can be caused by genetic factors, injury or unfavourable environmental conditions (Milton *et al*., 2008). This phenomenon has also been reported in members of some dicotyledonous families including Crassulaceae (Mabberly, 1987), Oxalidaceae (van der Pijl, 1983) and Saxifragaceae (Lid and Lid, 1994). In Liliaceae, pseudovivipary was documented in *Allium* (Stebbins, 1950) and *Crinum viviparum* (Ansari and Nair, 1987). However, this phenomenon has never been described in *Zephyranthes*. Our study is the first of its nature reporting pseudovivipary in *Z. carinata* and *Z. candida*. Several authors have argued that pseudovivipary has evolved in response to a short growing season, enabling plants to complete the cycle of offspring production, germination and establishment during the few weeks of an arctic or alpine growing season (Lee and Harmer, 1980). Molau (1993) has pointed out that pseudovivipary among tundra plants is mostly prevalent among late-flowering species. The present investigation reveals diagnostic feature of pseudovivipary as noticed in *Z. candida* and *Z. carinata*. In order to understand the mechanism of pseudovivipary in *Zephyranthes* species further detailed study is needed. Based on the present investigation, it could be concluded that reproductive biological characters somehow can be used for delimiting *Zephyranthes* species; however, further studies

including more taxa are needed for better understanding the taxonomy and interspecific relationships of the genus *Zephyranthes*.

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