Bangladesh J. Bot. **39**(2): 179-183, 2010 (December)

COMPARATIVE MORPHOLOGY AND FLORAL BIOLOGY OF THREE SPECIES OF THE GENUS *NYMPHAEA* FROM BANGLADESH

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Key words: Nymphaea, Comparative morphology, Floral biology

Abstract

Morphology and floral biology of three species of the genus *Nymphaea* (Nymphaeaceae) of Bangladesh revealed that *N. nouchali*, *N. pubescens* and *N. rubra*, were normal, protogynous or normal and protogynous, respectively. *N. pubescens* was night bloomer and each flower opened for three consecutive nights where as *N. rubra* opened for four consecutive nights and *N. nouchali* for three consecutive days. The behavior of anther dehiscence and stigma appendages had significant role in pollination. Excellent pollen germination and receptivity were observed in the exudate of stigma cup.

Introduction

As *Nymphaea pubescens* Willd. (Nymphaeaceae) is the national flower of Bangladesh, Its improvement with respect to economic and aesthetic aspect is of great significance. Prior to the genetic improvement of *Nymphaea*, study on morphology and floral biology in Bangladeshi materials were demanding. Other species should also be considered to study morphological variations. Three local species, namely *N. nouchali* Burm. f., *N. pubescens* Willd. and *N. rubra* Roxb. *ex* Andrews were studied in the present treatise. Floral biology studied by Schneider (1982), Prance and Anderson (1976) and Meeuse and Schneider (1980) on different species of *Nymphaea* mainly focused on protogynous nature and pollination mechanism. The present work is intended to study other behaviour of flowers that might influence the breeding behaviour of the species.

Materials and Methods

N. nouchali Burm. f., *N. pubescens* Willd., and *N. rubra* Roxb. *ex* Andrews collected from Bangladesh were studied. The results on floral biology were verified under tumbler condition both in Bangladesh and India. Morphological data of the species were taken from natural habitat of Bangladesh. Flowering and anthesis were recorded in September for three consecutive years. The opening and closing behaviour of flowers were measured with the help of a device developed by the author. The male reproductive nature was studied in respect of opening and closing of anthers, time of anther dehiscence and bending of stamens.

The female reproductive nature includes protogyny, stigma appendages, stigma exudates and stigma receptivity. The protogynous nature was tested by comparing the time of anthesis, stigma receptivity and anther dehiscence. Observations on the behaviour of stigma appendages were recorded from flowers on their first, second and third opening. The time and duration of stigma exudate secretion were examined. The presence of stigma exudates on stigmatic cup or its moist condition was considered as indication of stigma receptivity. It was confirmed by *in vivo* pollen germination percentage.

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Results and Discussion

Distinct morphological variations were observed in the *N. nouchali* Burm. f., *N. pubescens* Willd. and *N. rubra* Roxb. (Table 1, Fig. 1a-f).

Nymphaea nouchali: Stem, erect and tuberous, leaves glossy on both surfaces, light-green above, with or without purple patch and reddish-violet to bluish-violet beneath, peltate, margin waved and slightly dentate, lobes rounded. Flowers small, light-blue. Sepals not ribbed, glossy, entire, acute, reddish-green with blue inner surface. Petals acute, light-blue. Stamens with blue sterile appendages at the tip. Anthers triangular in cross section. Anther and stigma yellow, stigma saucer shaped with horn like appendages. Excellent fruit-set, seeds ellipsoid, enclosed by aril, floating.

Nymphaea pubescens: Stem, tuberous, erect, leaves ovate-elliptic, dentate, dark-green, glabrous above and purplish-green, pubescent and prominently veined beneath. Petioles and pedicels long. Sepals green and ribbed outside, white inside; Petals pure white, linear-oblanceolate and obtuse. Gradual transformation of petals towards stamen was prominent. Stamens arranged in three to four whorls, tip blunt and no sterile appendage. Anthers yellow, flat and possessed cream colored pollen grains. Anthers two-lobed, opened by longitudinal slit, introrse. Carpels many and syncarpous; stigmatic rays correspond to the number of carpels. Stigma cup shaped; stigma appendages oblong and yellow. Natural fruit formation observed which developed under water. Seeds ellipsoid, enclosed by transparent fleshy aril and floated on the water.

Nymphaea rubra: Stem, tuberous, erect, leaves glabrous, reddish-green above and reddish, pubescent and veined beneath. Flowers crimson-red, becoming lighter during winter season and bloomed round the year. Sepals 4, ovate-lanceolate, obtuse, reddish-green, ribbed outside and crimson-red inside. Petals crimson-red. Anthers and stigma appendages deep crimson-red. Stigma appendages very long, densely arranged, covering the stigma completely. No fruit formed naturally. *N. rubra* resembled *N. pubescens* in other respects.

The flower morphology of three *Nymphaea* species (Fig. 1a,b,c) in the present investigation is more or less similar to the observations of Mitra (1993) and Hossain *et al.* (2001). But it differed slightly with Conard (1905), Roxburgh (1932), Duthie (1960), Mosely (1961), Sen (1962), Prain (1963) and Khan and Halim (1979, 1987).

Table 1. Comparison of flower morphology of *Nymphaea* species.

Taxa	Flower diameter (cm)	Number of petals	Number of stamens	Length of sterile appendages	Ovary diameter (cm)	Number of ovary chambers	Length of stigma appen-dages (cm)
N. nouchali	8.60 ± 0.11	16.80 ± 0.6	75.50 ± 2.38	0.34 ± 0.12	2.10 ± 0.12	13.55 ± 0.45	0.19 ± 0.02
N. pubescens	11.35 ± 0.42	19.50 ±1.80	61.50 ± 0.80	Nil	1.47 ± 0.06	17.00 ± 0.32	0.51 ± 0.03
N. rubra	19.95 ±1.12	22.18 ± 0.67	82.90 ± 2.22	Nil	1.77 ± 0.11	22.40 ± 0.59	1.11 ± 0.07

The taxonomic rank of *Nymphaea* species varied between species and varieties in many of the cases. The work of Hossain *et al.* (2001) and Hassan (2009) confirmed the identify of the Bangladeshi species worked on by the authors.

Flowering in *N. rubra* occurs round the year, while in the other two species flowering takes place from May to November. Optimum temperature for growth and flowering ranged from 31 to 36º C.

Anthesis showed great variation among different species of *Nymphaea. N. pubescens* showed anthesis for three consecutive nights, *N. rubra* for four consecutive nights and in *N. nouchali* it

was for three consecutive days. In all the species anthesis for the second time was found to take place about half an hour earlier. Opening and closing time of flowers were very much influenced by the intensity of sunlight and hence temperature also. According to Prance and Anderson (1976) temperature was more effective than sunlight for the opening and closing of *Nymphaea* flowers.

The anthers were introrse and split longitudinally in all the species. In *N. nouchali* the anther lobes opened for ever but in *N. rubra* and *N. pubescens* opening and closing correspond to the spreading of petals.

Fig. 1a-f. Top view of flowers showing stigma appendages: *N. pubescens* (a), *N. rubra* (b) and *N. nouchali* (c). Details of floral parts in longitudinal sections: *N. pubescens* (d), *N. rubra* (e) and *N. nouchali* (f).

The time of anther dehiscence differed in the three *Nymphaea* species. In *N. nouchali* anthers burst 24 hours prior to the anthesis. *N. pubescens* showed dehiscence 24 hours before or after anthesis and in *N. rubra* it was during the second anthesis. In *N. nouchali* the anther of different whorls burst gradually from outermost to innermost whorl during successive anthesis. But in *N. pubescens* and *N. rubra* all the anthers burst simultaneously. At the second anthesis, the stamen tip gradually bent inwards in *N. pubescens* and *N. rubra.* The bending formed a cone and completely covered the stigma in *N. rubra* but in *N. pubescens* the loose cone leaves a terminal small opening. The case was reverse in *N. nouchali* where the non-dehiscent inner whorl bent inside and the dehisced anthers spread outside.

Stigma showing receptivity before the dehiscence of anther indicated the protogynous nature of flowersd. *N. rubra* showed protogyny but *N. pubescens* was either protogynous or normal. *N. nouchali* was always normal. The results showed similarity with those of Schneider (1982), and Prance and Anderson (1976). Stigma appendages remain erect in first anthesis but bent from the

second anthesis. In *N. rubra* long stigma appendages completely covered the stigma inhibiting pollination. In *N. pubescens* the covering was partial and loose. Horn like small stigma appendages could not cover the stigma in *N. nouchali* (Fig. 1a,b,c). This behavior of the stigma appendages played an important role in breeding behaviour of the species.

The flowers of all species showed their stigma cup full of stigma exudates in the first anthesis. In the second anthesis the stigma of *N. rubra* dried up but in *N. nouchali* and *N. pubescens* stigma cup remained sufficiently moist. Prance and Anderson (1976) observed fluid on the stigma cup of *N. ampla* in unopened bud condition but supposed it as lake water. Meeuse and Schneider (1980) found the sugar level of stigmatic exudate to be 1 - 1.5%, boron concentration 5 ppm and ionic composition in terms of Ca, K and Mg very close to optimal for pollen germination. The presence and role of exudate were observed by others (Heslop-Harrison and Shivanna 1977, Mosely 1961, Schneider 1982).

The receptivity of stigma was confirmed by *in vivo* pollen germination. Pollen grain showed the highest germination percentage on the stigma of flowers opened for the first time. On second anthesis *N. rubra* lost its receptivity but *N. nouchali* and *N. pubescens* showed excellent pollen germination. Third anthesis showed negligible stigma receptivity. Shivanna and Sastri (1981) observed correlation between presence of stigma exudate and pollen germination in *Amaryllis vittata* and *Crinum defixum.* According to them the protein (esterase) required for pollen germination was present in the stigma exudates.

Acknowledgements

A part of Ph. D. thesis by the first author who obtained a scholarship from I.C.C.R. Delhi, India which is duly acknowledged. The author is also indebted to the Director General, Secondary and Higher Education under the Ministry of Education, Government of the People's Republic of Bangladesh, Dhaka for granting study leave for carrying out the research.

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(Manuscript received on 15 June, 2010; revised on 12 November, 2010)