## EFFECTS OF SOME LIMNOLOGICAL FACTORS ON THE GROWTH OF NELUMBO NUCIFERA GAERTNER

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#### Abstract

*Nelumbo nucifera* Gaertner was grown in a mesoscale culture by sowing seeds in cow dung compost and loamy soil (1 : 1). Rhizomes and young plantlets under *ex-situ* condition did not survive but germination was possible via seeds where the rate of germination was 50% and seedlings less than four months old also grew. Leaf area positively correlated with air temperature, water temperature, PAR, day length, rainfall, relative humidity, NO<sub>3</sub>-N, Chl *a* and phaeopigment. Petiole length showed positive correlation with rainfall, relative humidity and pH. Highest growth and flowering were found to be associated with a water depth of 100 cm under full sunshine and high nutrient conditions. Under shaded condition, plants did not produce flowers. Water flow rate of 50 ml/sec in the culture pit hampered flowering. Flowering was observed only in the stagnant condition.

## Introduction

Bangladesh has a wetland area of about 74,197 km<sup>2</sup> (Akonda 1989), where about 100 species of Angiosperms, 3 species of Bryophytes and 8 species of Pteridophytes commonly occur (Khan and Halim 1987). All these species play an important role in the socio-cultural and economic activities of the people living in the catchments of the wetlands. Nelumbo nucifera Gaertner (locally called Padda) is an important wetland macrophyte species which grows naturally in different Haor ecosystems of greater Sylhet and Kishoreganj districts. The species has also been reported from some Beel ecosystems of Rajshahi district (Ahmed et al. 2009). Their harvest is fully dependent on natural population which is frequently damaged by other ongoing activities in the Haor such as fishing, boating, agriculture, etc. Considering the significance of the plant, Goel et al. (2001) made an ex-situ conservation strategy where a national collection comprising of 60 species/races of *Nelumbo* have been maintained in the National Botanical Research Institute, Lucknow (India) under the biodiversity conservation programme. In Bangladesh, very few works on macrophytic vegetation have been carried out and those are mainly taxonomic and a few ecological (Islam 1993, Khondker et al. 1993, 1994, 2004, Alfasane et al. 2010). Recently biochemical composition of the seeds of N. nucifera and Euryale ferox Salisb. were studied by Alfasane et al. (2008, 2009). There exists almost no research work in Bangladesh on the growth studies of N. nucifera. The present research has therefore been undertaken to carry out an ex-situ growth studies of *N. nucifera* and its relationship with some limnological factors.

#### **Materials and Methods**

The present research was carried out in the arboretum of Department of Botany, University of Dhaka between 2005 and 2007. Cut pieces of rhizome of *Nelumbo nucifera* Gaertner were collected from a ditch near the Botanical Garden of Chittagong University in April, 2005 and transplanted in the earthen bin filled with tap water in the Botanical Garden of Dhaka University. But the rhizomes did not survive. Thereafter (in July, 2005) five plantlets of 6 months old (20-30 cm in length) of *N. nucifera* were collected from the National Botanical Gardens, Mirpur, Dhaka.

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These were transferred in the culture pit (cemented tank) of Botanical Garden of Dhaka University, but the plantlets failed to survive. Then again in August, 2005, five plantlets were grown in pots and acclimatized in the National Botanical gardens, Mirpur and the pots containing plants were transferred to the culture pit in the Botanical Garden of Dhaka University. These plantlets died within 15 days of transfer.

Later on (in September, 2005) twenty mature seeds of the plant were collected from National Botanical gardens, Mirpur and sown in the earthen bin (77 cm diam. and 40 cm deep; placed in the soil in such a way that the rim remains 6 cm above the gorund) in the Botanical Garden of Dhaka University and the germination study was carried out. Nearly one third (15 cm) of the volume of the bin was filled with 10 kg loam mixed with composted cow dung (1:1) and then it was charged with tap water. After twenty days seeds germinated and young seedlings were nurtured to carry out the experiments. Eight young seedlings (3 months old) of *N. nucifera* were transferred to the cemented tank (3 m long  $\times$  2 m wide) of which bottom was filled with cow dung compost and loamy soil in 1:1 ratio, to about 30 cm thick layer. Immediately after transplantation some tap water was added in the tank in such a way that only the leaves of the plants remain afloat. Finally it was filled with underground water upto 100 cm. The water depth was adjusted every day following the growth of the plants. The growth measurement was carried out on randomly selected petioles (n = 8) and leaves (n = 32) of the plants grown in the culture pit and limnological data were recorded fortnightly.

PAR (photosynthetically active radiation) was determined with Li-Cor Quantum Meter, LI-185B, USA and air and water temperatures were measured with the help of a mercury centigrade thermometer. pH was measured with the help of a Griffin pH meter (PHJ-260-V-pH-meter, Model 50, UK). DO (dissolved oxygen) and soluble reactive silicate (SRS) were determined following Wetzel and Likens (1979). Alkalinity and nitrate-nitrogen (NO<sub>3</sub>-N) were measured after Mackereth *et al.* (1978) and Müller and Wiedemann (1955), respectively. SRP (soluble reactive phosphorus) was measured following Murphy and Riley (1962). Chlorophyll *a* and phaeopigment of the phytoplankton of tank-water was measured following Marker *et al.* (1980). Climatic data on relative humidity, total rainfall and day length of Dhaka Metropolis were collected from Bangladesh Meteorological Department, Dhaka (Table 1). Pearson Correlation study was made (SPSS program) to find the relationship between the measured limnological variables and the growth of petiole and leaf area.

## **Results and Discussion**

Petioles of *N. nucifera* plant was about 1.5 m in height and the leaves were as large as 50 cm in diameter. The leaves are green in winter, summer and monsoon but become yellow and starts decomposing during autumn.

The present investigation revealed that out of 20 mature seeds of *N. nucifera* sown in the culture pit, only 10 germinated after 20 days. Pagels (2001) reported that rate of seed germination of the plant ranged from 0-93% and after two weeks it was 18-93% when the seeds were gently rubbed on the sand paper. Goel *et al.* (2001) reported that *N. nucifera* multiplied by seeds and vegetatively by the division of rhizomes. The time period required for the germination of the seeds is in agreement with the observations made at the National Botanical Research Institute, Lucknow by Goel *et al.* (2001).

At the time of germination, some of the environmental factors were, PAR 776.83  $\pm$  107.21  $\mu$ E/m<sup>2</sup>/sec, day length 12.84  $\pm$  0.65 h, rainfall 114  $\pm$  56.48 mm, relative humidity (%) 91 $\pm$ 10.26, air temperature 34  $\pm$  2.33° C, water temperature 31  $\pm$  1.84° C, pH 7.10  $\pm$  0.09, alkalinity 4.0  $\pm$  0.87 meq/l, DO 8.45  $\pm$  1.83 mg/l, SRP 192.08  $\pm$  38.09  $\mu$ g/l, SRS 72.05  $\pm$  10.47 mg/l, NO<sub>3</sub>-N 53.5

 $\pm$  20.05 µg/l, planktonic chl *a* 34.5  $\pm$  10.2 µg/l and phaeopigment 11.2  $\pm$  5.8 µg/l. After germination of the seeds the young seedlings at less than four months old also grew well when transferred to another place. Masuda *et al.* (2005) reported that the seedlings of *N. nucifera* cultivated from May (i.e. summer) at 25-30° C for 2 months had more leaves.

The leaf area showed its average highest growth  $21.84 \pm 2.89 \text{ cm}^2/\text{day}$  in summer followed by the monsoon (18.31 ± 0.98 cm<sup>2</sup>/day) with lowest in winter (1.71 ± 0.55 cm<sup>2</sup>/day) and autumn (2.33 ± 0.29 cm<sup>2</sup>/day, Table 3). Seasonal highest growth rate of leaf area coincided with the seasonal average highest mean value of limnological parameters, PAR 776.83 ± 107.21  $\mu\text{E/m}^2/\text{sec}$ , water temperature 31.58 ± 1.95° C, air temperature 33.91 ± 2.30° C, alkalinity 4.10 ± 0.61 meq/1, NO<sub>3</sub>-N 108.72 ± 43.13 µg/l, chl *a* 102.34 ± 130.95 µg/l and phaeopigment 24.89 ± 36.98 µg/l (Tables 1 and 2). Another aquatic macrophyte *Euryale ferox* also showed increased growth rate of leaf area in summer (Alfasane *et al.* 2010).

Table 1. Seasonal mean values of different climatic parameters of Dhaka Metropolis.

| Parameters                    | Summer              | Monsoon             | Autumn            | Winter            |
|-------------------------------|---------------------|---------------------|-------------------|-------------------|
| PAR ( $\mu E/m^2/sec$ )       | $776.83 \pm 107.21$ | $653.78 \pm 202.67$ | $677.33\pm76.20$  | $620.28\pm51.30$  |
| Sunshine hour<br>(Day length) | $12.74\pm0.53$      | $12.84\pm0.65$      | $10.74\pm0.57$    | $10.98\pm0.32$    |
| Rainfall (mm)                 | $62.41\pm 66.12$    | $213.19\pm155.46$   | $38.83 \pm 52.09$ | $2.28\pm5.20$     |
| Relative humidity (%)         | $55.83 \pm 17.35$   | $79.59 \pm 9.61$    | $69.33 \pm 7.15$  | $55.86 \pm 11.07$ |

| Parameters                | Summer              | Monsoon           | Autumn            | Winter           |
|---------------------------|---------------------|-------------------|-------------------|------------------|
| Air Temp. (° C)           | $33.91 \pm 2.30$    | $32.00\pm2.24$    | $33.50\pm2.01$    | $25.49 \pm 4.02$ |
| Water Temp. (° C)         | $31.58 \pm 1.95$    | $29.93 \pm 1.99$  | $30.75 \pm 1.15$  | $24.28\pm3.89$   |
| pН                        | $6.67\pm0.07$       | $6.94\pm0.24$     | $7.09\pm0.06$     | $6.95\pm0.22$    |
| Alkalinity (meq/l)        | $4.10\pm0.61$       | $2.97\pm0.47$     | $3.45\pm0.34$     | $3.86 \pm 0.27$  |
| DO (mg/l)                 | $10.00 \pm 1.72$    | $5.65\pm0.97$     | $10.26\pm1.15$    | $11.08\pm0.94$   |
| NO <sub>3</sub> -N (µg/l) | $108.72\pm43.13$    | $60.97 \pm 41.93$ | $58.55\pm37.54$   | $95.28\pm65.17$  |
| SRP (µg/l)                | $113.46\pm65.34$    | $87.96 \pm 60.05$ | $138.81\pm29.04$  | $132.93\pm49.49$ |
| SRS (mg/l)                | $80.76\pm37.33$     | $74.52\pm25.29$   | $77.12 \pm 10.47$ | $85.22\pm25.08$  |
| Chl $a$ (µg/l)            | $102.34 \pm 130.95$ | $24.32\pm24.93$   | $24.99\pm8.85$    | $35.25\pm29.85$  |
| Phaeopigment (µg/l)       | $24.89\pm36.98$     | $5.67 \pm 5.64$   | $5.67 \pm 2.57$   | $6.85\pm5.65$    |

Table 2. Seasonal mean values of different limnological parameters in the culture pit.

Table 3. Growth rate of petiole (cm/day), leaf area (cm<sup>2</sup>/day) in *Nelumbo nucifera* in different seasons. n=8 for petioles, n=32 for leaves.

| Plant parts    | Summer           | Monsoon        | Autumn        | Winter        |
|----------------|------------------|----------------|---------------|---------------|
| Petiole length | $0.29\pm0.05$    | $0.34\pm0.10$  | $0.25\pm0.07$ | $0.22\pm0.05$ |
| Leaf area      | $21.84 \pm 2.89$ | $18.31\pm0.98$ | $2.33\pm0.29$ | $1.71\pm0.55$ |

The length of the petiole showed highest growth in monsoon followed by summer, autumn and winter (Table 3). Highest growth rate and flowering in some plants were found to be associated with a water depth of 100 cm. The present study reveals that the seasonal highest growth rate of petiole length associated with the seasonal average highest mean value of day length, total rainfall and humidity (Table 1). Similar type of observation was also recorded in *E. ferox* (Alfasane *et al.* 2010). From the study it is revealed that the highest growth of petioles of *N. nucifera* and *E. ferox* occurred in monsoon and this adaptation is because of their adjustment towards increasing water depth in monsoon.

Pearson correlation matrix also showed positive correlation of leaf area of *N. nucifera* with air temperature, water temperature, PAR, day length, rainfall, humidity, NO<sub>3</sub>-N, chl *a* and phaeopigment (Table 4). Petiole length showed positive correlation with rainfall, humidity and pH. However, both the growth parameters of *N. nucifera* showed negative correlation with alkalinity, DO, SRS and SRP. Among these parameters positive correlation between leaf area and day length was found to be significant at 5% level (Table 4). Pearson correlation matrix of another experiment on *Euryale ferox* showed leaf area positively correlated with air temperature, water temperature, PAR, day length, rainfall, humidity, NO<sub>3</sub>-N and petiole length showed positive correlation with rainfall and humidity. Leaf area and petiole length of *Euryale ferox* both showed negative correlation with alkalinity, DO, SRS and planktonic chl *a* (Alfasane *et al.* 2010), a feature also found in *N. nucifera*.

| (uutu extructor         | i nom or oo prog |           |                    |                |           |
|-------------------------|------------------|-----------|--------------------|----------------|-----------|
| Limnological parameters | Growth rate      |           | Limnological       | Growth rate    |           |
|                         | Petiole length   | Leaf area | parameters         | Petiole length | Leaf area |
| PAR                     | - 0.818          | +0.682    | Alkalinity         | - 0.904        | - 0.019   |
| Day length              | - 0.195          | +0.976*   | DO                 | - 0.569        | - 0.567   |
| Rainfall                | +0.452           | +0.673    | NO <sub>3</sub> -N | - 0.853        | +0.269    |
| Humidity                | +0.811           | +0.238    | SRP                | - 0.501        | - 0.383   |
| Air Temp.               | - 0.229          | +0.565    | SRS                | - 0.233        | - 0.807   |
| Water Temp.             | - 0.248          | +0.619    | Chl a              | - 0.951*       | +0.624    |

- 0.776

Table 4. Pearson correlation between limnological parameters and growth rate of *Nelumbo nucifera* (data extracted from SPSS programme).

\* Correlation is significant at the 0.05 level (2- tailed)

+0.797

It has been found that the flower opens early in the morning and closes in the afternoon. The flower was usually pink, 10-25 cm in diameter with a single flower per peduncle. Petal length was  $10.38 \pm 1.33$  cm and breadth was  $5.36 \pm 1.44$  cm. The range of total number of petals were 17-19. Length of the stamen was  $2.86 \pm 0.26$  cm and the diameter of the stigma (seed pod) was  $3.2 \pm 0.5$  cm<sup>2</sup>. Goel *et al.* (2001) also mentioned phenotypic diversity of *N. nucifera* with a large number of racial variants with different shape, size and colour of the blossom ranging from white to dark pink and having 16-160 petals.

Phaeopigment

- 0.935

+0.663

In summer, flower was found to be on a thick peduncle rising about 22 cm above the water under full sunshine and high nutrient condition of the tank-water. The length of the peduncle was 1.2 m. Under the shaded condition due to the interruption of other large plants near the culture pit, some plants didn't produce flower. Flowering occurred during April to June. At the time of flowering different limnological parameters showed the following ranges: air temperature 28.5- $30.5^{\circ}$  C, water temperature 28-29° C, pH 7.23-7.28, alkalinity 1-3 meq/l, TDS 104-125 mg/l, conductivity 236-390 µS/cm, DO 4.80-5.89 mg/l, NO<sub>3</sub>-N 407.99 - 543.99 µg/l, SRP 21.98-53.01 µg/l, SRS 53.29-54.55 mg/l, planktonic chl *a* 5.30-95.75 µg/l and phaeopigment 1.71-22.00 µg/l.

pН

Goel *et al.* (2001) reported that flowering and fruiting in *N. nucifera* took place from March to October. They also observed decreased flowering and fruiting during extreme hot summer and rainy days. From the present study and others it appears that a moderate climate is suitable for the optimum growth and profuse flowering. Phenological studies have revealed that there is a thermoregulatory mechanism in lotus plants which maintain a steady environment inside the flowers to facilitate successful pollination by humming bees (Goel *et al.* 2001). In the present investigation visit by humming bees in the flower was also observed.

Water flow affected the flowering of *N. nucifera* and it was found that a flow rate of 50 ml/sec in the culture pit hampered flowering. Flowering was observed only in the stagnant condition. On another occasion, plants (seedlings less then 4 months old) when transferred into another culture pit with stagnant water on the south of the Department of Botany, survived very well and flowering also occurred within one year. To conserve threatened aquatic macrophytes of Bangladesh similar mesoscale culture experiments may be undertaken to ensure their *ex situ* conservation.

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