ABSTRACT: *Eichhornia azurea* (Schwartz) Kunth is one of the most abundant species of aquatic macrophyte in the Upper Paraná River floodplain, and ephemeropteran nymphs frequently inhabit this plant. The objective of this study was to analyze the distribution of ephemeropteran nymphs associated with three sections (apical, intermediate, and basal) of *E. azurea* in two floodplain lakes of the Upper Paraná River as well as to establish the relation of this distribution to biotic and abiotic factors. The samplings of apical, intermediate, and basal sections of *E. azurea* were carried out monthly from October 1997 to October 1998. Higher concentration of dissolved oxygen may contribute to greater density of nymphs on the apical plant segments near the water surface. *Callibaetes willineri* was the most abundant taxon, occurring on the apical and intermediate plant sections whereas *Campsurus* spp. were recorded on the basal and intermediate sections of the plant, near the sediment and, mainly during high water. Lower dissolved oxygen concentrations near the lake bottom during the high water phase might suggest that during this phase the nymphs of *Campsurus violaceus* also migrate to *E. azurea* roots. It was found that changes in water level influenced the variation of the abundance of nymphs. Other genera, such as *Tricorythodes* and *Leptohyphes*, were recorded during the high water phase, when river water enters the lakes and their shores are flooded.

KEYWORDS: Ephemeroptera distribution, *Eichhornia azurea*, floodplain lake, Paraná River

*Eichhornia azurea* (Schwartz) Kunth is one of the most abundant aquatic macrophyte species in the Upper Paraná River floodplain. The particles adhered (organic matter or periphyton) to submerged segments of this plant serve as food source for a wide array of organisms. Of the invertebrates colonizing aquatic macrophytes, ephemeropteran nymphs are frequently found on *E. azurea*. The distribution of some species may be related to season (*Nolte et al.* 1996), fluctuations in water level (*Tabacchi et al.* 1993), type of substrate (*Giller and Campbell* 1989), and food supply (*Da Silva* 1998).

The objective of the present study was to analyze the spatial distribution of ephemeropteran nymphs associated with three sections (apical, intermediate, and basal) of *E. azurea* plants in Guaraná and Patos Lakes of the Upper Paraná River floodplain as well as to describe the relationship with abiotic factors.
The study was carried out in two lentic environments of the Upper Paraná River floodplain. Patos Lake (22º49'19''S and 53º31'33''W) is located in the floodplain on the left bank of the Ivinheima River, and it is distinguished mainly by its irregular shoreline. Depth during the study period varied from 1.2 to 4.0 m. Grasses and other macrophytes including *E. azurea* predominate along the shore. Guaraná Lake (22º43'26''S and 53º18'03''W) is located in the varzea on the right bank of the Baía River; it is connected with river by a permanent channel (Fig. 1A). Maximum depth varied from 1.2 to 3.0 m. Aquatic macrophytes, especially *E. azurea*, predominate in the channel and in the littoral zone of the lake, whereas grasses predominate on the shore.

Samples of *E. azurea* were obtained monthly from October 1997 to October 1998 in both lakes. One plant stolon was pulled out of the water by the floating leaf stem, until almost all root tufts were exposed. Next, the plant was partitioned into three sections: apical (stalk and submerged leaves, near the water surface), intermediate, and basal (submerged roots near the bottom) (Fig. 1B).

Each section (root and stalk) was placed in polyethylene flasks and preserved in 80 % alcohol. Later, plant parts were scraped and washed in 80% alcohol to remove adhering particles, and the portions of root and stalk were oven-dried at 60ºC. The density of organisms was calculated per 100 g plant dry weight. In the laboratory, nymphs were sorted, counted, and identified using a stereoscopic microscope.

At each sampling station, water transparency (by Secchi disc), depth, and temperature were measured. Water samples were collected from surface, middle, and near-bottom depths using a Van Dorn bottle, and pH and conductivity (µS cm⁻¹) were determined in the laboratory with a portable digital pH meter and conductivity meter, respectively. The Winkler method modified by Golterman *et al.* (1978) was used to determine dissolved oxygen concentration.

The Paraná and Ivinheima Rivers influence the water level in Guaraná and Patos

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**Fig. 1.** A) Study area and location of the sampling stations (● – Sampling stations ⚫ – Nupelia field laboratory ❂ – Direction of flow). B) Location of the sections of *Eichornia azurea*. Modified from Volkmer-Ribeiro *et al.* 1984.
lakes, respectively (Souza-Filho and Stevaux 1997). During the study period, the high water phase of the Paraná River occurred in the period from November 1997 to March 1998, and May 1998 and October 1998. In the Ivinheima River, high water occurred in November and December 1997, March through May 1998 and August to October 1998. The remaining months were considered low water phase (Fig. 2).

Five taxa of ephemeropteran nymphs were found: *Callibaetis willineri* Navás 1932, *Leptohyphes* sp. Eaton 1882, *Tricorythodes* sp. Ulmer 1920, *Caenis* sp. Stephens 1935, and *Campsurus* sp. Eaton 1868. *Caenis* sp. and *C. willineri* were frequent in both studied lakes (Figs 3A and B). *Tricorythodes* sp. was observed only in Patos lake on the apical section of *E. azurea* (Fig. 3A).

Abiotic variables were summarized using Principal Components Analysis (PCA). The first two axes were retained for interpretation (eigenvalues greater than 1). This axis explained 61.7% of the total variability of the data: the first component explained 38.1% whereas second 23.6%. Ordination separated the two lakes (Fig. 4A). Dissolved oxygen was positively associated with PCA 1, whereas conductivity and depth were negatively associated. For PCA 2, pH and transparency (Secchi) presented positive correlation (Fig. 4B).

Three-way ANOVA was applied on axes 1 and 2 of PCA, with lake, plant section and hydrological period (high water and low water) as factors (normality and homoscedasticity assumptions were met). ANOVA identified significant differences of the scores between the lakes and hydrological period on PCA axis 1 ($F = 5.08; P = 0.028; F = 34.98; P < 0.01$; respectively) and among the lakes ($F = 47.42; P < 0.01$) on PCA 2. These results indicate differences in abiotic conditions summarized in PCA axis.

Nymphs distribution in the lakes and on different sections of the plant was summarized using Detrended Correspondence Analysis (DCA) (Gauch 1986). This analysis was performed in the case of abundance estimates of nymphs for sections of the plant and lakes. Axis 1 of DCA presented eigenvalue of 0.52 whereas for axis 2, the eigenvalue was 0.22. These two axes were retained for interpretation. To test for differences between lakes, plant sections (Fig. 5) and hydrological periods three-way ANOVA was performed using samples from DCA axes 1 and 2; no significant differences were observed in the interaction of the analyzed factors either for DCA (axis 1) and DCA (axis 2) ($F = 0.15; P = 0.86; F = 0.72; P = 0.48$, respectively).

To determine which environmental factors may be influencing nymphs distribution in the two lakes, correlations between PCA axes and DCA axes and hydrological periods were made. Seemingly, local
Factors have influenced the abundance of ephemeropteran nymphs, because DCA axis 1 was significantly correlated with PCA axis 1 ($r = 0.45; P < 0.01$) and axis 2 from PCA with hydrological periods ($r = –0.62; P < 0.01$). The physical and chemical characteristics of the lakes did not vary with depth; however, PCA demonstrated a clear separation of the lakes. Although the biotic variables (DCA axis 1), were only related to PCA axis 1, water level strongly influenced variation in dissolved oxygen. In these lakes, dissolved oxygen concentration varies with flood stage, and is lower during high water and higher during low water (Takeda et al. 1991). Dissolved oxygen concentration can be an important factor affecting the distribution of species of aquatic insects (Ward 1992). Kolar and Rahel (1993) established that nymphs of Callibaetis montanus cannot tolerate hypoxic conditions. One of the factors that may have contributed to the greater density of nymphs at apical plant sections is higher dissolved oxygen concentration at the surface.

Fig. 3. Mean (and ± SE) values (Oct. 1997–Oct. 1998) of Ephemeroptera nymph density (ind. 100 g d. w.−1) according to hydrological periods and sections of plants. A) Patos Lake B) Guaraná Lake (see Fig. 1).
Besides the effect of oxygen, the rise in water level may result in the occasional appearance of species not adapted to this particular environment, such as nymphs of *Tricorythodes* sp., which occurred only once in Patos Lake on apical plant section. According to Corkum (1989), these nymphs can be transported from the rivers to the lakes by water circulation. Nolte *et al.* (1997) also observed the occurrence of members of this genus in a lake of the Pantanal (Brazil) after several flood peaks.

First-stage nymphs of Baetidae (which are not identified to genus) were recorded only on apical plant segment. This is attributable to the egg-laying habit of the female, and to the development of the first stages that must take place on this part of the plant. Samways *et al.* (1996) reported that these nymphs occur on the plant near the water surface, to avoid the anoxic conditions and low temperatures of the bottom.

Nymphs of *Leptohyphes* were infrequent in both lakes, probably because they are...
more common along the banks of rivers, streams, and brooks (Edmunds et al. 1976, Froehlich and Oliveira 1997). Nolte et al. (1997) found a significant correlation between the abundance of *Leptophyes* and current velocity.

Nymphs of *Caenis* and *Callibaetis willineri*, the most abundant taxa, occurred on the apical and intermediate plant section. *Caenis* and *Callibaetis* feed on diatoms and other algae (Berner and Pescador, 1988). Da Silva (1997) established that in addition to algae, *Callibaetis guttatus* can eat fibers and decomposing remains of plants. The nymphs of *C. willineri* swim by means of contractions of the body and caudal appendages to reach other parts of the plant, and when disturbed they seek shelter in the roots. Their long, clawed front legs facilitate food capture and fixation to the substrate (Da Silva, pers. com.). Kaisin and Bosnia (1987) observed that vegetated areas together with a large quantity of organic matter provide favorable conditions for the development of *Caenis*, i.e., microhabitats, refuges from predators, and food resources.

The highest densities of species of *Campsurus* were recorded on the basal and intermediate section of the plant, mainly during high water in both lakes. Melo et al. (1993) suggested that nymphs of *Campsurus violaceus* in the Baia River have a life cycle adapted to the hydrological cycle of the Paraná River. They can migrate from the middle of the river to its banks during high water months, similarly to *Leptophleia cupida* from a northern population that seasonally migrates between a river and adjacent marshes (Hayden and Clifford after Ward 1989). Due to migratory patterns, nymphs of these two species avoid adverse conditions in rivers. The lower dissolved oxygen concentration near the lake bottom during the high water phase might suggest that during this phase nymphs of *Campsurus violaceus* also migrate into *Eichhornia* roots.

Changes in water levels influenced the temporal variation of the nymphs. However, other genera such as *Tricorythodes* and *Leptophyes* were recorded during the high water phase, when river water enters the lakes and their shores become flooded. The inundation and post-inundation periods may be important for dispersal of nymphs of these taxa, with the entry of new species into the environment in search of shelter on a certain section of the macrophytes.

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