# Feeding selectivity and growth of Nile tilapia (Oreochromis niloticus L.) fed on temperate-zone aquatic macrophytes

I. Šetlíková<sup>1</sup>, Z. Adámek<sup>2</sup>

<sup>1</sup>Faculty of Biological Sciences, <sup>2</sup>Research Institute of Fish Culture and Hydrobiology in Vodňany, University of South Bohemia, České Budějovice, Czech Republic

**ABSTRACT**: Feeding selectivity of Nile tilapia (*Oreochromis niloticus* L.) juveniles (9.3–20.9 g) to four aquatic macrophyte species and tilapia growth were studied in 4 consecutive experiments. Plant diet was provided to 8 tanks containing 20 fishes for 5 days. The fish were fed a carp diet between 4 experiments for 14 days. The consumption of 4 aquatic macrophyte species differed significantly  $[F(3,252) = 39.6; P < 10^{-6}]$ . *Elodea canadensis* was the most preferred plant (Chesson selectivity index =  $0.50 \pm 0.05$ , n = 4). *Potamogeton pectinatus* and *Spirodela polyrhiza* were consumed with about equal preference. *Myriophyllum spicatum* was the least preferred species. *Elodea canadensis* contained relatively more phosphorus, potassium and ash than the other three species. The daily plant dry weight intake ranged between 0.79 and 2.26% of body weight. The fish grew during the first two experiments (SGR = 2.54 and 3.18%/d, respectively), but lost weight during the 3rd and 4th experiments (SGR = -1.75 and -1.71%/d, respectively).

**Keywords**: fish nutrition; cichlids; plant diet; *Elodea canadensis*; *Myriophyllum spicatum*; *Potamogeton pectinatus*; *Spirodela polyrhiza* 

Despite the high abundance of macrophytes and microalgae in many freshwater bodies, there are only a few herbivorous fish species taking advantage of these resources. Among them are silver carp (Hypophthalmichthys molitrix Val.), grass carp (Ctenopharyngodon idella Val.), African tilapia species and, to a lesser extent, also European roach (Rutilus rutilus L.) and rudd (Scardinius erythrophthalmus L.) (Niederholzer and Hofer, 1979). Nile tilapia (Oreochromis niloticus L.) is an omnivorous species able to utilize food sources of various trophic levels, with a preference for blue-green algae, green algae and diatoms (Moriarty and Moriarty, 1973; Moriarty, 1973; Getachew, 1987; Khallaf and Aln-Na-Ei, 1987), but ingests also organic material (detritus) at different degrees of decomposition and filters bacteria, zooplankton and zoobenthos (Balarin and Hatton, 1979; Adámek and Sukop, 1995). Tilapia is also supposed to digest macrophytes effectively (Westoby, 1974; Khallaf and Aln-Na-Ei, 1987; Adámek and Mareš, 1990; Chapman and Fernando, 1994; Teferi, 1997). However a few studies conducted on the use of aquatic plants in tilapia (*Oreochromis* spp.) feeds had varying and sometimes conflicting results (for a short review see El-Sayed, 1999).

Nile tilapia are increasingly cultured in the temperate zone (e.g. heated effluents). Therefore the objective of the study was to examine which of the aquatic macrophytes commonly occurring in the temperate-zone would be preferentially consumed by Nile tilapia and which species would be useful for inclusion in prepared diets. The purpose of this study was to evaluate (1) the feeding behaviour of two-months-old Nile tilapia juveniles with respect to four temperate-zone macrophyte species on a selective and quantitative basis and (2) to assess the growth of Nile tilapia under experimental feeding regimes using macrophytes.

### MATERIAL AND METHODS

Two-month-old Nile tilapia (mean initial individual weights see Table 1) were taken from a facility using heated effluents of the ironworks at Hrádek u Rokycan (Czech Republic), where fish were fed on a commercial carp diet (20% crude protein). The study was conducted in four consecutive experiments from June to August 1998 (1st experiment: 15 June–19 June, 2nd experiment: 4 July–8 July, 3rd experiment: 23 July-27 July and 4th experiment: 11 August–15 August). Each experiment had two parts: 4 days in which one of the four aquatic macrophytes species was offered and the 5th day on a mixture of these plants (Figure 1). Nile tilapia juveniles were stocked into 8 experimental tanks (0.3 m<sup>3</sup> each), with twenty individuals in each tank. The fish were fed ad libitum by introducing a weighed bulk (about 40 g fresh weight) of four aquatic plant species (Elodea canadensis L.C. Rich. Ex Michx., *Potamogeton pectinatus* L., *Myriophyllum spicatum* L. and *Spirodela polyrhiza* (L.) Schleiden). Aquatic macrophytes originated from small eutrophic ponds (mean depth of 1.5 m; [TP]: 0.09 mg/l and [TN]: 2.51 mg/l; mean from June–August 1998; n = 6). Prior to the weighing, fresh biomass of aquatic macrophytes was dried 5 times on its surface using filter paper. Each aquatic plant species was applied to two experimental tanks daily at 07.00 h for a period of 24 hours. The uneaten plant remains were removed from the tanks, dried again with filter paper and weighed (accuracy of fresh weight: 1 g and dry weight: 0.01 g). This part of all four experiments lasted 4 days.

On the 5th day of all four experiments, a mixture comprising one quarter by fresh weight of all four aquatic plant species was supplied to each tank. A randomised block design was applied in all four experiment replications. In order to protect the fish from a deficit of animal protein, they were fed *ad li-*

Table 1. Growth performance of the fish fed on plant diet (n = 8; mean  $\pm$  SD) and environmental conditions during the experiments (n = 120; mean  $\pm$  SD)

Experiment	1st	2nd	3rd	4th
Initial weight (g)	9.3 ± 1.2 <sup>a</sup>	11.3 ± 1.1 <sup>a</sup>	$16.9 \pm 1.6^{b}$	$20.9 \pm 2.0^{\circ}$
Final weight (g)	$10.5 \pm 1.0$	$13.2 \pm 1.5$	$15.5 \pm 1.2$	$19.1 \pm 1.6$
$FI^1$	$0.79 \pm 0.06^{a}$	$1.02 \pm 0.11^{b}$	$2.26 \pm 0.29^{\circ}$	$1.08 \pm 0.13^{b}$
SGR <sup>2</sup>	$2.54 \pm 0.88^{a}$	$3.18 \pm 1.49^{a}$	$-1.75 \pm 1.67^{\rm b}$	$-1.71 \pm 0.4^{\rm b}$
FCR <sup>3</sup>	$0.34 \pm 0.09^{a}$	$0.38 \pm 0.32^{a}$	$-3.78 \pm 5.64^{\mathrm{b}}$	$-0.35 \pm 0.15^{ab}$
T (°C)	$24.3 \pm 1.6^{a}$	$24.5 \pm 1.4^{a}$	$23.4 \pm 1.3^{a}$	$17.7 \pm 1.0^{\rm b}$
$O_2 (mg/l)$	$5.7 \pm 0.8^{ab}$	$6.0 \pm 1.3^{a}$	$7.4 \pm 0.8^{\rm ac}$	$6.1 \pm 0.6^{a}$
рН	$8.2 \pm 0.4^{a}$	$7.5 \pm 0.8^{b}$	$7.5 \pm 0.3^{b}$	$7.5 \pm 0.1^{b}$

<sup>&</sup>lt;sup>1</sup>food intake (g DW/100 g fish biomass/day)

Figures in the same row with different superscripts are significantly different (P < 0.05)

1 <sup>st</sup>	day	_	2 <sup>nd</sup>	day	_	3 <sup>rd</sup>	day	_	$4^{\mathrm{th}}$	day	_	5 <sup>th</sup>	day
1 P	2 S		1 S	2 S		1 S	2 E		1 M	2 E		1 SEMP	2 SEMP
3 <i>S</i>	4 M		3 M	4 P		3 P	4 S		3 <i>S</i>	4 S		3 SEMP	4 SEMP
5 E	6 M		5 P	6 E		5 M	6 M		5 P	6 P		5 SEMP	6 SEMP
7 P	8 E		7 E	8 M		7 P	8 E		7 E	8 M		7 SEMP	8 SEMP

1–8: tank number; E - M: 4 species of aquatic macrophytes (E - Elodea canadensis, M - Myriophyllum spicatum, P - Potamogeton pectinatus, S - Spirodela polyrhiza)

Figure 1. Example of randomised experimental design (1st experiment)

<sup>&</sup>lt;sup>2</sup>specific growth rate (%/day)

<sup>&</sup>lt;sup>3</sup>food conversion ratio (dry food intake/fish weight gain)

Experiment	1 <sup>st</sup>		2 <sup>nd</sup>		3 <sup>rd</sup>		4 <sup>th</sup>
E or $M$ or $P$ or $S$ in each tank	1 <sup>st</sup> –4 <sup>th</sup> day	14 days carp diet	1 <sup>st</sup> –4 <sup>th</sup> day	14 days carp diet	1 <sup>st</sup> –4 <sup>th</sup> day	14 days carp diet	1 <sup>st</sup> –4 <sup>th</sup> day
E+M+P+S	5 <sup>th</sup> day		5 <sup>th</sup> day		5 <sup>th</sup> day		5 <sup>th</sup> day

(E – Elodea canadensis, M – Myriophyllum spicatum, P – Potamogeton pectinatus, S – Spirodela polyrhiza)

Figure 2. Design of all four experimental periods

bitum a commercial carp diet (crude protein: 20.0%, fat: 7.6%, fibre: 3.6%, ash: 5.5% and dry weight: 87.3%) between the four experimental periods for two weeks (Figure 2).

The experimental tanks were provided with constant aeration without water exchange during each of the four experimental periods and exposed to the natural light/dark cycle. Water temperature, dissolved oxygen concentration, and water pH were recorded three times a day using WTW Multiline P4.

Chemical analyses of aquatic plants. The aquatic plants were dried (105°C), weighed and then analysed for mineral nutrient content prior to being ashed at 500°C to determine ash content. Dried aquatic plants were mineralised using either mixture of H<sub>2</sub>SO<sub>4</sub> and H<sub>2</sub>O<sub>2</sub> or Kjeldahl method (in the case of TN) (Javorský, 1987). The content of mineral macronutrients (TN, TP, K, Ca and Mg) was determined in a commercial laboratory (Agro-la, Jindřichův Hradec, Czech Republic) according to methods described by Javorský (1987). Content of total nitrogen (TN) was determined using coulometric titration with electrogenerated hypobromite and biamperometric detection. Spectroscopic phos-

phovanadomolybdate complex method was used for determination of total phosphorus content (TP). Content of potassium (K), calcium (Ca) and magnesium (Mg) was determined using flame atomic absorption spectrometry. Mean carbon content in plant tissue was assumed to be 38% of dry matter (Moeller, 1978): this value was used to estimate the C: N ratio of the plants (Table 2).

**Feeding selectivity and fish growth**. Feeding selectivity was calculated as the alpha selectivity index of Chesson (1978), using the formula:

$$\alpha_i = r_i / p_i \left( \sum_{i=1}^n r_i / p_i \right)^{-1}; i = 1, ..., n$$

$$\sum_{i=1}^n \alpha_i = 1$$

where:  $r_i$  = the percentage of food item i in the fish ration  $p_i$  = the percentage of the same item in the total food offered

n = the total number of food items

When  $\alpha = 1/n$ , selective feeding does not occur. When  $\alpha_i < 1/n$ , less food item i occurs in the diet than expected from random feeding (negative selection). When  $\alpha_i > 1/n$ , more food item i occurs in the diet than expected (positive selection). Because n = 4 here, then 1/n = 0.25.

Table 2. Percentage concentrations of mineral nutrients and ash in the dry mass of the four macrophyte species (mean  $\pm$  SD, n = 4)

Species	Elodea canadensis	Myriophyllum spicatum	Potamogeton pectinatus	Spirodela polyrhiza
N	$2.04 \pm 0.52^{a}$	$1.51 \pm 0.34^{a}$	$2.06 \pm 0.47^{a}$	$2.19 \pm 0.37^{a}$
P	$0.59 \pm 0.23^{a}$	$0.18 \pm 0.07^{b}$	$0.26 \pm 0.08^{b}$	$0.18 \pm 0.04^{\rm b}$
K	$5.18 \pm 0.98^{a}$	$1.58 \pm 0.47^{\rm b}$	$2.33 \pm 0.77^{b}$	$2.53 \pm 0.31^{b}$
Ca	$2.32 \pm 0.43^{a}$	$2.14 \pm 0.51^{a}$	$1.76 \pm 0.47^{a}$	$1.47 \pm 0.47^{a}$
Mg	$0.18 \pm 0.02^{a}$	$0.43 \pm 0.58^{a}$	$0.23 \pm 0.11^{a}$	$0.25 \pm 0.11^{a}$
ash	$19.7 \pm 4.63^{a}$	$11.2 \pm 1.89^{b}$	$12.4 \pm 2.44^{b}$	$11.0 \pm 1.81^{\rm b}$
C:N	20:1ª	26:1ª	19 : 1 <sup>a</sup>	18:1ª

Figures in the same row with different superscripts are significantly different (P < 0.05)

Fish growth was characterised using specific growth rate (SGR), fish daily food intake (FI) and food conversion ratio (FCR):

SGR = 100 (In final fish weight – In initial fish weight)/n days of feeding (%/day)

FCR = dry food intake over each period/sum of mean wet fish increment

**Statistical analyses**. Statistical analyses were performed using the program Statistica 5.5. Normality was tested using the Kolmogorov-Smirnov test. Data on the consumption of plant dry weight as dependent on the plant species were processed by analysis of variance (ANOVA), and Tukey's multiple range test was used to compare the means that were considered significantly different at P < 0.05. Regression analysis was applied using the amount of plant dry matter consumed as dependent variable and water temperature as independent variable. Initial and final fish fresh weights were compared using t-test for dependent samples.

#### **RESULTS**

# **Environmental parameters**

The mean water temperature was 24.1°C (range 21.2 to 29.2°C; n = 360) during the first three experiments and decreased to 17.7°C (range 16.3 to 20.1°C; n = 120) during the fourth experiment (Table 1).

Mean oxygen concentrations 6.3 mg/l (range 5.1 to 8.5 mg/l) and pH 7.6 (range 7.0 to 9.1) were within acceptable limits during all experiments (Table 1).

# Feeding selectivity and behaviour

Nile tilapia readily consumed all aquatic plant species offered, i.e. *Elodea canadensis, Myriophyllum spicatum, Potamogeton pectinatus* and *Spirodela polyrhiza* in all four experiments. When the plants were first introduced into a tank, the fish immediately approached to them but then retreated. Assessment of palatability was different. A single fish was observed to leave the moving shoal and taste the plant on offer. This action lasted about 5 minutes and was repeated twice or three times before the whole shoal started to consume the plants.

The consumption of aquatic plants was not correlated with temperature (P=0.54). The analysis of variance (ANOVA) showed a highly significant difference [F(3,252) = 39.6;  $P<10^{-6}$ ] in the rates of dry matter consumption of the different aquatic plant species in all experiments (Figure 3). Mean intake of *Elodea canadensis* by tilapia was the highest (0.31–0.89 g DW/100 g fish biomass per day) during all four experimental periods when compared to other aquatic plant species. The intake did not differ significantly between *Potamogeton pectinatus* and *Spirodela polyrhiza* while *Myriophyllum spicatum* was the least consumed species (0.05–0.27 g DW

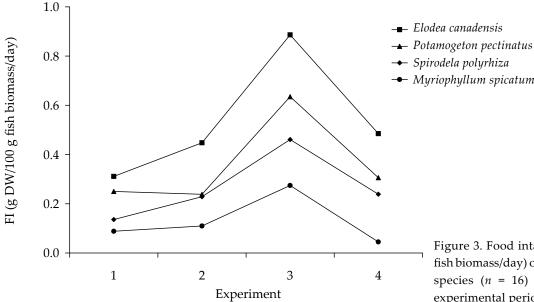


Figure 3. Food intake (g DW/100 g fish biomass/day) of 4 aquatic plants species (n = 16) during all four experimental periods

per 100 g fish biomass per day) (Tukey's test). The results were the same regardless of whether the four aquatic plant species were supplied separately or as a mixture.

Tilapia preferred the leaves of *Elodea canadensis* and almost all stems were left intact. In the case of *Spirodela polyrhiza*, the fish consumed the roots and young leaves. Whole plants of *Potamogeton pectinatus* were consumed. Fish grazed only on the young light green leaves of *Myriophyllum spicatum* while its stems with roots remained intact.

Selectivity index values differed significantly  $[F(3,124) = 234.7; P = 10^{-3}]$  between all aquatic plant species supplied except between *Potamogeton pectinatus* and *Spirodela polyrhiza* (Tukey's test, Table 3). *Elodea canadensis* had an  $\alpha$  value higher than 0.25 and thus can be considered as an aquatic plant species preferred by Nile tilapia. *Potamogeton pectinatus* and *Spirodela polyrhiza* were indifferent as regards the feeding preference of tilapia. *Myriophyllum spicatum* was eaten by the fish the least.

Table 3. Mean values of selectivity index according to Chesson (1978)

Species	Mean $\pm$ SD $(n = 32)$	Range
Elodea canadensis	$0.50 \pm 0.05^{a}$	0.40-0.75
Potamogeton pectinatus	$0.25 \pm 0.07^{\rm b}$	0.12-0.40
Spirodela polyrhiza	$0.22 \pm 0.05^{b}$	0.10-0.34
Myriophyllum spicatum	$0.06 \pm 0.03^{c}$	0-0.18

Figures in the same column with different superscripts are significantly different (P < 0.05)

# Chemical composition of plants

Significant differences in the phosphorus, potassium and ash concentrations were found between the four aquatic plant species. *Elodea canadensis* contained relatively more phosphorus, potassium and ash than the other species (Table 1). The plant material showed a high C: N ratio (Table 1). Nitrogen concentrations were significantly different in the four experiments [F(3,12) = 5.0; P = 0.02], i.e. they were highest during the first two experiments (average value 2.28% of dry mass) and decreased to 1.63% during the third and fourth experiments. The concentrations of other nutrients (P, K, Ca and Mg) in the plant tissues did not show much variability over the season.

# Fish growth

The growth response of the fish during the experiments is presented as the individual initial and final weight, mean of FI, SGR and FCR (Table 1). The individual initial fish fresh weight significantly differed from the final weight in all four experimental periods [ $t_{1\text{st exp.}} = -10.8$ ;  $P < 10^{-4}$ ,  $t_{2\text{nd exp.}} = -5.8$ ; P = 0.001,  $t_{3\text{rd exp.}} = 2.9$ ; P = 0.02 and  $t_{4\text{th exp.}} = 9.6$ ;  $P < 10^{-4}$ ]. The fish significantly grew during the first two experiments but lost weight during the third and fourth experiments. Initial individual fish fresh weights differed significantly from one another  $[\boldsymbol{F}_{\text{initial weight}}$ (3,28) = 98.3;  $P < 10^{-6}$ ] (Table 1). Mean food intake was 1.29 g DW/100 g fish biomass per day with the highest value during the third experiment (2.26 g DW/100 g fish biomass per day) (Figure 3). The efficiency of the plant utilization was very poor, large parts of the plant material appeared not digested in the fish excreta.

#### **DISCUSSION**

The mean water temperature during the first three experiments was optimal (22.7 to 26.6°C) but decreased to 17.7°C during the 4th experiment. This temperature is already too low for optimum feeding (22°C to 30°C; Dvořák *et al.*, 1989). However no correlation was found between the consumption of aquatic plants and water temperature. Similar results were described by Gur (1997) with the only increase of food intake during the spring temperature increase.

While Nile tilapia was not pre-adapted to ingestion of plants, it consumed all four aquatic macrophytes species readily from the beginning. Tilapia can easily and quickly adapt and thus there was no need to get it used to consume plants. The Chesson index ( $\alpha$ ) values (Table 3) correspond to the order of preference as expressed in the experiments with the ingestion of individual plant species (Figure 3). Different chemical composition (Bonar et al., 1990) and mechanical properties (Fischer, 1968; van Zon, 1977) could cause the different consumption rate of macrophyte species. In our experiments, Elodea canadensis was the most preferred plant species by the Nile tilapia, and had the highest content of phosphorus, potassium and ash as compared to the other three plant species tested (Myriophyllum spicatum, Potamogeton pectinatus and Spirodela polyrhiza). Leonard et al. (1998) reported that Oreochromis aureus (Steindachner) preferred leaves to roots of Azolla filiculoides Lam., which simultaneously had higher contents of P, K, ash and crude protein (calculated as N  $\times$  6.25). The availability of phosphorus in the feed is the limiting factor of plant protein utilisation by Nile tilapia (Gur, 1997). However, the addition of P to the 100% Spirulina diet only slightly improved the performance of tilapia (Oreochromis mossambicus (Peters)) fry in comparison with the same diet without phosphorus (Olvera-Novoa et al., 1998). The relation between plant consumption and its nutrient content was evident especially for Myriophyllum spicatum from which tilapia ate only young leaves. Young tissues contain the highest concentration of protein (nitrogen) and mineral nutrients (Begon et al., 1997).

Mean daily intake of aquatic plants (0.79–2.26 g DW/100 g fish biomass per day) was within the range (0.67–4.17 g DW/100 g fish biomass per day) reported for ingestion of aquatic plants by adult tilapia (Gaigher et al., 1984; El-Sayed, 1992; Hassan and Edwards, 1992; Leonard et al., 1998). Increased food intake during the third experiment (2.26 g DW per 100 g fish biomass per day) resulted in more rapid food passage through the digestive tract of the fish (large pieces of undigested plants were found in the excreta) and a shorter time available for digestion and absorption of the food. Fish match their food consumption to the digestible nutrient and energy content: if little digestible food is ingested, food consumption increases (Jobling, 1983; Bowen et al., 1995).

The fish grew only slightly when fed exclusively aquatic vegetation, and even lost their weight in the third and fourth experiments. This is in agreement with Okeyo (1987 - cited Okeyo, 1989), who observed that Oreochromis aureus (Steindachner) lost weight when fed three aquatic macrophytes (Elodea sp., Myriophyllum spicatum L. and Potamogeton gramineus L.) but his experiment lasted longer (35 days). The plant diet in our experiments had a higher C: N ratio than optimal. In the case of Myriophyllum spicatum, this ratio markedly exceeded 21:1, which could have resulted in conspicuous malnutrition of the fish (Russell-Hunter, 1970). Fish weight loss during the last two experiments could also have caused by decreasing contents of nitrogen (protein) in plants during the growing season (during the experiments). Although tilapia received standard food between the experiments, they probably were under nutritional stress.

The most preferred plant was *Elodea canadensis* when offered in combination with other three species. *Potamogeton pectinatus* and *Spirodela polyrhiza* were consumed and considered to be of about equal preference. *Myriophyllum spicatum* was the least preferred species. Low growth rates were recorded in Nile tilapia fed exclusively on aquatic plants.

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## **ABSTRAKT**

# Potravní výběrovost a růst tilapie nilské (Oreochromis niloticus L.) krmené našimi druhy vodních makrofyt

Ve čtyrech navazujících pokusech byla sledována potravní výběrovost našich druhů vodních makrofyt tilapií nilskou (*Oreochromis niloticus* L.) (9,3–20,9 g). Do každé z osmi plastových nádrží (0,3 m³) bylo nasazeno 20 ryb, kterým byla po dobu pěti dnů předkládána rostlinná potrava. Mezi pokusy byly ryby krmeny kapřími pelety po dobu dvou týdnů. Spotřeba jednotlivých druhů vodních makrofyt se statisticky průkazně lišila [F(3,252) = 39,6;  $P < 10^{-6}$ ]. Pořadí preferencí bylo: vodní mor kanadský (*Elodea canadensis*) (Chesson index selektivity = 0,50 ± 0,05, n = 4) > rdest hřebenitý (*Potamogeton pectinatus*) a závitka mnohokořenná (*Spirodela polyrhiza*) > stolístek klasnatý (*Myriophyllum* 

spicatum). Vodní mor obsahoval ve své sušině ve srovnání s ostatními třemi druhy vodních makrofyt více fosforu, draslíku a popela. Denní příjem rostlin (v suché hmotnosti) se pohyboval mezi 0.79-2.26% živé hmotnosti tilapie. Tilapie v průběhu prvních dvou opakování pokusu rostly (průměrná SGR = 2.54 a 3.18%/d), v průběhu třetího a čtvrtého pokusu ryby hubly (průměrná SGR = -1.75 a -1.71%/d).

**Klíčová slova**: výživa ryb; cichlidy; rostlinná potrava; *Elodea canadensis*; *Myriophyllum spicatum*; *Potamogeton pectinatus*; *Spirodela polyrhiza* 

Corresponding Author

RNDr. Irena Šetlíková, PhD., Jihočeská univerzita v Českých Budějovicích, Zemědělská fakulta, Katedra ekologie, Studentská 13, 370 05 České Budějovice, Česká Republika

Tel. +420 387 772 756, fax +420 385 310 122, e-mail: setlik@zf.jcu.cz