ISSN 1392-3196 / e-ISSN 2335-8947 Zemdirbyste-Agriculture, vol. 106, No. 3 (2019), p. 203–212 DOI 10.13080/z-a.2019.106.026

Nitrogen concentration of the aquatic plant species in relation to land cover type and other variables of the environment

Edvina KROKAIT˹, Dinara SHAKENEVA¹, Erika JUŠKAITYT˹, Tomas REKAŠIUS¹.² Jolanta NEMANIŪTĖ-GUŽIEN˹, Jurgita BUTKUVIEN˳, Jolanta PATAMSYT˳, Vida RANČELIENĖ⁴, Regina VYŠNIAUSKIENĖ⁴, Laisvūnė DUCHOVSKIENĖ⁵, Lina JOCIEN˹, Zofija SINKEVIČIENĖ⁴, Donatas NAUGŽEMYS³, Violeta KLEIZAIT˳, Damian CHMURA⁶, Neil O. ANDERSON⁵, Donatas ŽVINGILA³, Eugenija KUPČINSKIEN˹

¹Vytautas Magnus University Vileikos 8, Kaunas, Lithuania E-mail: edvina.krokaite@gmail.com

²Vilnius Gediminas Technical University Saulėtekio 11, Vilnius, Lithuania

³Vilnius University Saulėtekio 7, Vilnius, Lithuania

⁴Nature Research Centre Žaliųjų Ežerų 49, Vilnius, Lithuania

⁵Lithuanian Research Centre for Agriculture and Forestry, Institute of Horticulture Kauno 30, Babtai, Lithuania

⁶University of Bielsko-Biala, Institute of Environmental Protection and Engineering Willowa 2, 43-309 Bielsko-Biala, Poland

⁷University of Minnesota, Department of Horticultural Science 1970 Folwell Avenue, St. Paul, MN 55108, USA

Abstract

Nitrogen (N) deposition data, together with inland water parameters, provide evidence that N load may affect the vegetation of Baltic States. There is much concern about eutrophication of the rivers, although information about physiological parameters of riparian plant species is still poor. The present study is aimed at comparison of leaf N concentration among populations of aquatic plant species of Lithuania, relating N concentration data to the type of land cover (classification system of COoRdinate Information on the Environment, CORINE) in the neighbouring areas, river state and size, intensity of agriculture in 1991–1996, and natural vice versa regulated fragments of the riverbed. The leaf N concentrations of widely spread native and invasive species (5 riparian and 2 water plants) were estimated for 241 sites (collection time 1st ten-day period of August, 2015) of the main river catchments of Lithuania. Only leaf blades were used for analyses and N concentrations were determined by the Kjeldahl method. The biggest (1.7 times) variation (p < 0.05) in leaf N concentration among populations was documented for Lythrum salicaria. According to the mean values (N % of dry mass, DM) of the leaf N concentration, species could be arranged into following order: Lythrum salicaria (3.0) < Stuckenia pectinata (3.1) < Phalaris arundinacea (3.5) < Bidens frondosa (3.8) < Phragmites australis (4.0) < Nuphar lutea (4.1) < Echinocystis lobata (4.2). Significantly higher (p < 0.05) concentrations of leaf N were found for L. salicaria populations growing near the small rivers (3.4% N DM) compared to the large ones (2.8% N DM). Other selected river and its environment parameters in most cases did not have significant effect on leaf N concentrations of aquatic plants of Lithuania. Leaf N concentration of the aquatic species was not influenced by land cover type (2000 and 2006 year data of database) and did not depend on the river size or state, based on the Water Directive guidance (EU, 2000). Among selected species the most nitrophylic was invasive for Lithuania species E. lobata, which is currently spreading along larger size (>1000 km²) rivers. The present levels of N entering riparian ecosystems are causing spread of the macrophyte species consuming relatively high amounts of N, although the main sources of macrophyte N remains

Key words: land cover, riparian plants, river regulation, river state, water macrophytes.

Please use the following format when citing the article:

Krokaitė E., Shakeneva D., Juškaitytė E., Rekašius T., Nemaniūtė-Gužienė J., Butkuvienė J., Patamsytė J., Rančelienė V., Vyšniauskienė R., Duchovskienė L., Jocienė L., Sinkevičienė Z., Naugžemys D., Kleizaitė V., Chmura D., Anderson N. O., Žvingila D., Kupčinskienė E. 2019. Nitrogen concentration of the aquatic plant species in relation to land cover type and other variables of the environment. Zemdirbyste-Agriculture, 106 (3): 203–212. DOI 10.13080/z-a.2019.106.026

Introduction

Nitrogen (N) is the most deficient element for plants in their intact natural state, although within the last century anthropogenic activities have doubled the amount of this element circulating on Earth (Pinay et al., 2018). In both instances, directly or indirectly, intensive agriculture, settlements, transport and industry are causing N pollution (Yoshikawa et al., 2015). Data of European Monitoring and Evaluation Programme (EMEP, 2017) provided information about increased emissions (21%) of ammonia (NH₂) within 2000–2015. For the present century, further growth of atmospheric N deposition is forecasted (Bobbink et al., 2010; Vet et al., 2014). In 2015, critical loads (based on N and sulphur (S) data) for eutrophication were exceeded (200 eq ha⁻¹ yr⁻¹ mean increase) in many countries of Europe (CPST, 2015). Reports of freshwater eutrophication are increasing (O'Hare et al., 2018). Freshwaters are permanently deteriorated by excess of N, which creates gradient of nutrients across the terrestrialaquatic ecosystems (Moldan, Wright, 2011; Pinay et al., 2018; Erős et al., 2019).

Compared to the data on eutrophication of the lakes, there has been significantly less work carried out on rivers (Mäemets et al., 2010; Hering et al., 2015). Riparian zones act as buffers for N, which incompletely consumed quantities are moving from agricultural fields to aquatic ecosystems (Yoshikawa et al., 2015; Hille et al., 2018; Pinay et al., 2018). In most cases, the physical and chemical characteristics of the water are recorded when evaluating the state of inland water bodies (Fuller et al., 2015; Yoshikawa et al., 2015). Data on aquatic N pollution (EMEP, 2017) provide only indirect evidence about possible effects on the health of aquatic organisms (Hettelingh et al., 2009). When eutrophication of rivers is discussed, more attention is paid to algal blooms compared to aquatic angiosperms (Yoshikawa et al., 2015; Erős et al., 2019). Monitoring networks usually assess macrophytes, preparing lists of the common plant species combined with the coverage data (Hille et al., 2018). Plant communities are formed during the longterm process; information about present availability of nutrients in different parts of the river basin could be obtained by measuring the N concentration of the leaves as a key important organ (Thompson et al., 1997; Han et al., 2005).

Nitrogen concentrations have been well examined for conifers and agricultural crops (Heinsoo et al., 2011; Butkutè et al., 2014; Pocienè, Kadžiulienė, 2016; Tan et al., 2018), defining N concentration ranges for deficiencies and toxicities (Bobbink et al., 2010). Under elevated N supply special morphological, physiological and biochemical strategies of the plants have been recorded: increases in shoot density, shoot length, leaf density, thickness of the leaves, concentration of free arginine, nitrate reductase activity and many other traits (Kupcinskiene, 2001; Chmura et al., 2016).

Worldwide information about plant N concentration is related to diverse parameters: mass of the aboveground part of the plants per plot, mass of aboveground part of the shoot, mass of the leaves and/or ratios of concentrations of nitrogen and phosphorous (N:P) (Han et al., 2005; Heinsoo et al., 2011; Butkutė et al., 2014; Brezinová, Vymazal, 2015; Pocienė, Kadžiulienė, 2016). In addition to the classical Kjeldahl

method, flow liquid spectroscopy, flash combustion method, hyperspectral technics are employed to estimate N for agriculture species (Tan et al., 2018). In some investigations only mean values of N concentration for very big set of the species are presented (Han et al., 2005), while in other surveys species N requirements are evaluated when employing the ready to-use Ellenberg indicatory values (EIV) (Ellenberg et al., 1992).

Despite worldwide large amounts of N data, in each situation there are different sets of plant species, growing under particular climatic or edaphic conditions with differential human pressure. Due to genetic and phenetic differences, distinct sampling approaches, methods and parameters of N determination, data obtained about quantities of N for certain species are heavily transformable from one region to another.

Excess of critical loads for eutrophication were documented in various parts of Lithuania (EMEP, 2017) and were higher when compared to the other Baltic States. For Lithuania, few records (Tumas, 1998) exist concerning past farming effects on the quality of the river waters.

In recent years, substantial data was collected on the frequency of distribution and abundance of aquatic plants, including riparian macrophytes, in Lithuania (Zviedre et al., 2015). In addition, pilot data has been collected on the adverse consequences on genetic diversity of populations of aquatic macrophytes under anthropogenic pressure (Anderson et al., 2018). Data concerning plant saturation by N is missing for riparian species in the Baltic States. Very little is known to what extent N concentration may vary between plant populations receiving unequal environment pressure.

The present study is aimed at comparison of N concentration among populations of riparian and aquatic plant species of Lithuania, relating N data to the type of the land cover in the neighbouring areas, river state and size, intensity of past agriculture activities and riverbed regulation.

Materials and methods

Sampling material and study sites. Phragmites australis (Cav.) Trin. ex Steud., Phalaris arundinacea L., Lythrum salicaria L., Echinocystis lobata (Michx.) Torr. & A. Gray and Bidens frondosa L. were selected as riparian species; Nuphar lutea (L.) Sm. and Stuckenia pectinata (L.) Börner were sampled as aquatic plant species (further all species, both riparian and water are titled under aquatic). Such list of the plants was based on frequent occurrence and abundance (P. arundinacea, P. australis, N. lutea, S. pectinata) or related to invasions (E. lobata and B. frondosa were categorized as invasive in Lithuania; L. salicaria, P. arundinacea, P. australis classified as native in Lithuania, but invasive in some habitats of North America). P. australis was sampled in 43 sites, P. arundinacea in 61 sites, L. salicaria in 39 sites, E. lobata in 18 sites, B. frondosa in 12 sites, N. lutea in 59 sites and S. pectinata in 9 sites. In total, plant material of seven species growing in 241 sites was analysed (Fig. 1).

To assess the anthropogenic effects of leaf N on aquatic species, plants were collected in the main river basins of Lithuania: Nemunas, Venta, Lielupė, and the seafront. Selection of the sites was based on availability of plant species. Sampling of plants was performed in

ISSN 1392-3196

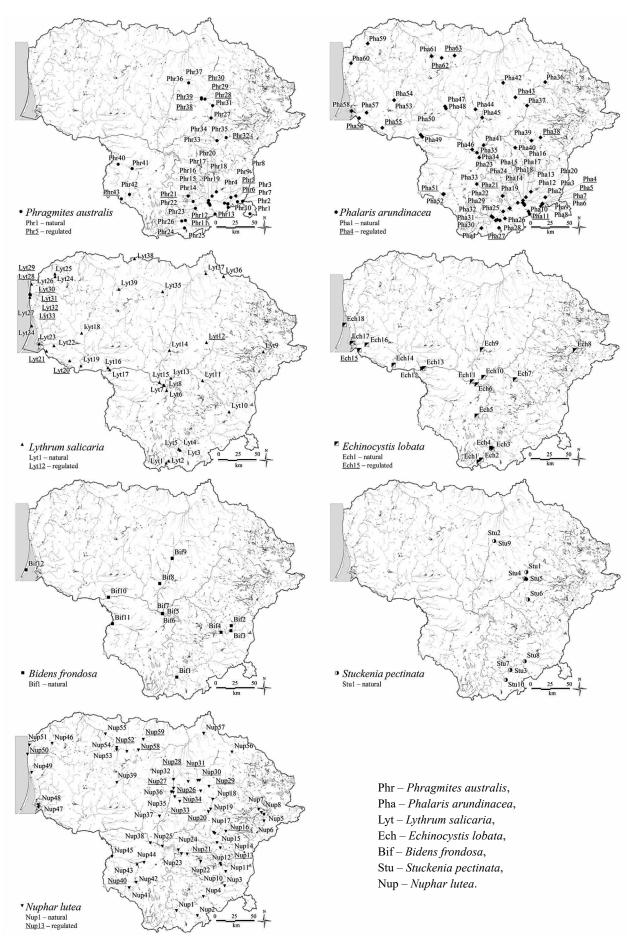


Figure 1. Geographical location of Lithuanian populations of riparian species, sampled for nitrogen concentration analyses

the 1st ten-day period of August, 2015 at the time plants were in flowering-ripening stage. For each species, three independent batches of the plant material were collected along 300 m length transects of separate site.

Nitrogen (N) analysis. Healthy (lacking in insect, fungi and/or bacteria damage), fully developed greenish leaves from the main shoot upper half were sampled for the analyses. Leaf blades (henceforth: leaves) were detached, dried at 70°C, ground to a fine powder using a mill Retsch MM400 (Germany) and sifted through a 1 mm screen. Over 750 samples were analysed by the Kjeldahl method (Allen, 1989) using Kjeldahl digestion unit DK-20S and automatic analyser UDK 159 (VELP Scientifica, Italy), following protocols of the manufacturer. Analyses were performed at the Department of Ecology and Environmental Sciences, Vytautas Magnus University. Nitrogen concentration was determined and expressed as % dry mass (DM) of the leaf tissue. Quality assurance was achieved using standard reference materials (SRM1515 and SRM1575) and certified reference material (CRM125045).

Environmental variables. Discerning the possible effects of features of the rivers and their environments on the N state of the plant, populations (evaluated according to the leaf N concentrations) were categorized in five ways: 1) three groups according to the land cover of area neighbouring riverbank: artificial areas (ART), agricultural areas (AGR), forest and semi-natural areas (further in the text named under forest areas, abbr. FOR), employing CORINE Land Cover database (classification level 1) available for 2000 and 2006 (CLC, 2006); 2) five groups according to the river status: high (H), good (G), moderate (MO), poor (P) and bad (B), using the classification of the Water Framework Directive (EU, 2000); 3) three location groupings: North-West (NW), Central (C) and South-East (SE), based on N concentrations in the rivers' (respectively 2.6–3.9, 2.9–4.8 and 0.6–2.1 mg L⁻¹) neighbouring agricultural areas during 1991–1996 (Tumas, 1998); 4) four groups based on the river size: small (<100 km²), medium-sized (100–1000 km²), large (1000–10000 km²) and extra-large (>10000 km²), following classification by the Water Framework Directive (EU, 2000); 5) two groups based on riverbed origin: natural and regulated (Gailiušis et al., 2001).

Statistical analysis. For comparisons (at 95% probability) of groups of populations of each species, median values were used and calculations of differences among groups were analysed employing statistical software *R*, version 3.4.4 (R Core Team, 2018) (Pohlert, 2014; Subirana et al., 2014).

Results and discussions

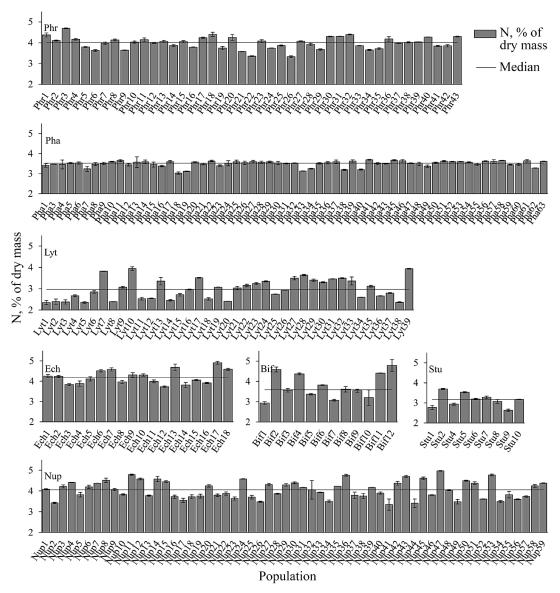
Leaf N concentrations of populations of the riparian and aquatic plant species. Mean of leaf N concentration for separate populations of *Phragmites australis* ranged from 3.33% to 4.41% of dry mass (DM) (Fig. 2); the most contrasting ones differed by 1.32 times (p < 0.05), being the lowest for the Phr26 population (Skroblus river) and the highest for Phr3 (Visinčia river). Mean leaf N concentration for all populations of *P. australis* was 3.99% DM. Mean of leaf N concentration for separate populations of *Phalaris arundinacea* ranged from 3.03% to 3.69% DM and was the most consistent among populations of all species examined. The most

contrasting values differed by 1.22 times (p < 0.05); the lowest N concentration was in the Pha18 population (Merkys river), while the highest was Pha41 (Neris river; Fig. 2). Mean leaf N concentration for all populations of P. arundinacea was 3.50% DM. Lythrum salicaria populations had mean leaf N concentrations ranging from 2.35% to 3.94% DM; the most contrasting ones differed by 1.68 times (p < 0.05). The lowest mean N concentration was in the Lyt1 population (Nemunas river), whereas the highest occurred in Lyt39 (Vokė river), while the mean leaf N concentration for all populations of L. salicaria was 2.98% DM. Separate populations of *Echinocystis* lobata ranged from 3.72% to 4.91% DM of leaf N; the most contrasting ones differed by 1.32 times (p < 0.05). The lowest concentration was in the Ech12 population (Nemunas river) to the highest in Ech17 (Atmata river); the grand mean = 4.18% DM for all *E. lobata* populations. Bidens frondosa populations ranged from 2.94% DM (Bif1; Nemunas river) to 4.79% DM (Bif12; Nevėžis river), with the most contrasting ones differing by 1.63 times (p < 0.05) and a grand mean of 3.77% DM. The leaf N concentration for Stuckenia pectinata populations ranged from 2.63% (Stu9) to 3.69% (Stu2) DM, both of which were located on the Nevėžis river (Figs 1 and 2). The most contrasting S. pectinata populations differed by 1.40 times (p < 0.05) with a grand mean of 3.14% DM for all populations. For *Nuphar lutea*, leaf N ranged from 3.34% DM (Nup41; Šešupė river) to 4.97% DM (Nup47; Pakalnė river), differing by 1.49 times (p < 0.05) with a grand mean of 4.06% DM (Fig. 2).

Land cover. Prevailing neighbouring land type according to use might be assumed as indirect evidence about the most favourable environment for one of the other aquatic species. Agricultural areas were the prevailing type of neighbouring land for 6/7 examined species, comprising 79% cases for *P. australis*, 72% for *P. arundinacea*, 53% in *L. salicaria*, 72% for *E. lobata*, 56% in *S. pectinata* and 66% for *N. lutea*. The only exception was a prevalence of artificial areas (67%) for *B. frondosa*. In all species, the range of N distribution among populations of different neighbouring land types overlapped (Fig. 3A). *S. pectinata* was not observed besides artificial areas, and *B. frondosa* was absent near forested areas.

Within 2000–2006, agricultural net N load varied from 74.6% to 89.5% of the total N net load depending on the percentage of arable land and the load from point sources in five subcatchments of the river Nemunas (Šileika et al., 2013). The study (1995–2006) of the Merkys, Mūša, Nevėžis and Žeimena river basins, Lithuania has shown much higher N export coefficients from arable land compare to the export from forested land and pastures and meadows (Povilaitis, 2011). In our study, no significant differences in leaf N concentrations were found among groups of populations neighbouring artificial, agricultural and forested areas.

Within 2000–2006, the main input of the N from point sources entered Kaunas city (Šileika et al., 2013). Our study supports this fact: higher than median values of N concentration were documented for *B. frondosa* (site Bif6), *E. lobata* (site Ech6) and *L. salicaria* (Lyt7) growing in the centre of Kaunas (Fig. 2). Studies conducted in other countries have shown that pollution of the rivers and ground waters by N was mainly caused



Note. Bars above the columns are confidence intervals for each population; Phr – Phragmites australis, Pha – Phalaris arundinacea, Lyt – Lythrum salicaria, Ech – Echinocystis lobata, Bif – Bidens frondosa, Stu – Stuckenia pectinata, Nup – Nuphar lutea.

Figure 2. Mean (columns) and median (horizontal lines crossing the columns) values of the leaf nitrogen (N) concentration of groups of the aquatic populations of macrophytes

by runoff and leaching from agricultural fields with the contribution of forests and urban land cover being very small (Yoshikawa et al., 2015).

The absence of land cover-related differences in our study might be due to recent shifts of Lithuanian agriculture from intensive farming to ecological, improvement of wastewater cleaning systems in settlements and cities. An additional factor is that river neighbouring areas of different land cover are very fragmented in Lithuania; thereby their effects on plants might overlap.

River status. A moderate (MO) state for river fragments prevailed in populations of *L. salicaria* (40% of all populations), *E. lobata* (82%) and *B. frondosa* (82%), whereas a good (G) state occured for populations of *P. arundinacea* (47%), *P. australis* (41%), *S. pectinata* (56%) and *N. lutea* (34%). *P. arundinacea*, *L. salicaria* and *N. lutea* grew in the river fragments of "whatever state" (M, G, MO, P and B) (Fig. 3B). *P. australis* and

E. lobata did not grow in the fragments categorized as "bad state". S. pectinata and B. frondosa were not observed in the river fragments of the poor (P) and bad (B) states. According to the occurrence of populations in the river fragments of different state, as defined by Water Framework Directive (EU, 2000), the most resistant to pollution were P. arundinacea, L. salicaria and N. lutea, while the most sensitive included S. pectinata and B. frondosa. Wherever, L. salicaria, P. arundinacea and N. lutea were growing, including waters of poor (P) or bad (B) states, did not necessarily match higher leaf N concentrations. Among the examined species the lowest leaf N was for L. salicaria, whereas P. arundinacea was intermediate and the highest occurred with N. lutea. Bad state of the fragment of the river might also impair other factors than high concentrations of N.

River pollution by former agriculture. In the Baltic States, including Lithuania, the environmental consequences of intensive agriculture development within

period 1950-1990, remains poorly studied; evaluation of adverse farming effects on water pollution started later. In 1992-1996, a pilot study was performed subdividing Lithuanian territory into North-West, Central and South-East parts, according to the N and other element concentrations in the river fragments (2.6-3.9, 2.9-4.8, and 0.6–2.1 mg N L⁻¹, respectively) neighbouring agricultural areas (Tumas, 1998). For P. australis (72% of all populations), P. arundinacea (57%), and S. pectinata (67%) the biggest number of Lithuanian populations were collected in the South-East part representing the least polluted rivers (0.6–2.1 mg N L⁻¹ in 1992–1996; Tumas, 1998). E. lobata (39%), B. frondosa (58%), and N. lutea (49%) in the Central part represented the most polluted rivers (2.9-4.8 mg N L-1) and L. salicaria (44%) in the North-West part (2.6-3.9 mg N L⁻¹). No differences were found according to the leaf N concentrations, classifying aquatic macrophyte populations into North-West (NW), Central (C) and South-East groups (SE) (Fig. 3C), based on water pollution within 1992-1996 in the river fragments' neighbouring agricultural areas. Thus, former differences in river quality did not have longer-term consequences on plant nutrition. Complex nature of various factors affecting N losses from agriculture-dominated stream catchments in Lithuania was documented: 15-year data did not show any statistically significant trend either in the dynamics of the annual N concentration or in the annual N load in the streams (Stålnacke et al., 2014).

River size. The largest number of populations of *P. australis*, *P. arundinacea*, *L. salicaria* and *N. lutea* (53, 49, 33 and 56 % of all populations, respectively) were located in medium sized rivers, while for *B. frondosa* and *S. pectinata* (50% and 56%, respectively) found in large rivers, *E. lobata* (56%) – in extra-large rivers (Fig. 3D). Significantly higher (p < 0.05) concentrations of leaf N were found for *L. salicaria* populations growing near the small rivers (3.4% N DM) compared to the large ones (2.8% N DM). No other differences were found according to the leaf N concentration classifying aquatic macrophyte populations into groups based on river size. Small size rivers were missing among *P. arundinacea*, *S. pectinata* and invasive in Lithuania *E. lobata* and *B. frondosa*.

Our data showed that for plant invasions the most vulnerable are big rivers. It is obvious for E. lobata intensively spreading over the past decades. Large rivers of Lithuania are crossing the most populated cities, which are good donors for the seeds of alien species, initially grown as ornamentals. Large rivers may bring seeds from the countries located to the south, where temperature is more favourable for termophyllic E. lobata, whose requirement for temperature was scored as 8 out of 9 (1 = least termophillic, 9 = most termophillic species) (Ellenberg et al., 1992). As invader it was recorded in Central Europe, compared to Lithuania (Kopec et al., 2014). Large rivers are crossing wide agricultural areas and municipal wastewaters, bearing N pollutants, thereby creating the most favourable conditions for multiple introductions of invaders (Yu et al., 2018).

River regulations. There is big concern about harm caused by river regulations, since they are frequently accompanied by habitat fragmentation, eutrophication and diminished diversity of aquatic species (Banks et al., 2013; Brezinová, Vymazal, 2015). In some cases,

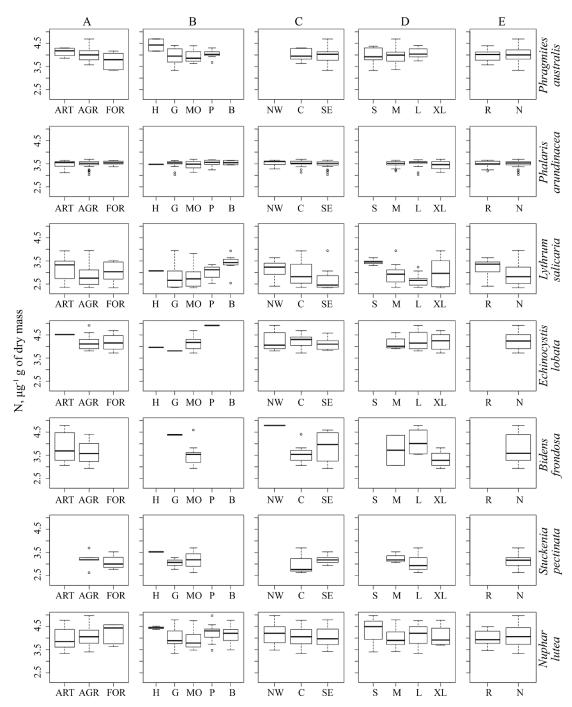
river regulations resulted in an increase of terophytes, including invasive species, such as *E. lobata* and *Bidens triparta* (Kopec et al., 2014). In Lithuania, 83% of the river fragments are regulated (Gailiušis et al., 2001).

Our results show that the highest numbers of populations for all species, were located in the natural river parts: 65% of all *P. australis* populations, 79% of *P. arundinacea*, 77% of *L. salicaria* and 71% of *N. lutea* (Fig. 3E). For some species, the natural river parts were the only locations, e.g., *E. lobata*, *B. frondosa* and *S. pectinata*. No differences were found according to the leaf N concentration, classifying macrophyte populations into groups of natural and regulated bed rivers. In parallel, our ongoing studies have demonstrated significant changes in genetic diversity due to river regulations (Anderson et al., 2018; Vyšniauskienė et al., 2018).

Based on previous investigations of numerous species, the leaf N concentration ranged between 1–2 and 5 µg mg⁻¹ DM (Thompson et al., 1997; Han et al., 2005). Our results are in agreement with these data. Compared to the leaf N data of the other *Poaceae* species (Thompson et al., 1997), concentrations of N determined for our studied aquatic species were not as high as for *Poa annua*, although much higher compared to *Arrhenaterum elatius*, *Elytrigia repens* and *Holcus mollis*.

Comparison of several aquatic species showed that the lowest mean of N concentrations is found for L. salicaria and the highest – for E. lobata populations. According to the mean values of leaf N concentration in all populations, species could be arranged into the following order: L. salicaria < S. pectinata < P. arundinacea < B. frondosa < P. australis < N. lutea< E. lobata. Relying on species indicator values for soil richness, defined by Ellenberg et al. (1992) (1 - least nitrophilic species, 9 - most nitrophilic species), six species in our survey could be classified as demonstrating higher mean requirements for the soil N: N. lutea (score = 6); P. arundinacea and P. australis (score = 7); E. lobata, B. frondosa and S. pectinata (score = 8). In the Ellenberg et al. (1992) survey, L. salicaria was defined as species with the widest range demand for N. Our data agree to it: L. salicaria population differences in leaf N concentration were the highest in comparison to the other species studied. B. frondosa was the second species with big differences in leaf N concentration among populations. It agrees with the findings concerning high plasticity of B. frondosa to N in the environment (Wei et al., 2016). Small numbers of Lithuanian populations of S. pectinata could be a reason for inconsistencies between our data and studies in Germany. S. pectinata and E. lobata were found only in less than 25 % sites, B. frondosa – in 20 % sites. Among the other reasons of differences (for *N. lutea*, in particular) might be distinct soil and climatic conditions, pollution and/or farming tradition changes after survey done by Ellenberg and coauthors (1992). Some discrepancies between indicatory values obtained by Ellenberg group and researchers in other than Germany countries are permanent subject for discussions (De Caceres et al., 2010).

Some of the studied species, i.e. *P. arundinacea*, *E. lobata* and *L. salicaria*, are ornamental crops (Anderson et al., 2018) and it would be worthwhile to elucidate details of their N requirements. Currently,



Note. ART – artificial area, AGR – agricultural area, FOR – forest area; H – high state of the water, G – good, MO – moderate, P – poor, B – bad; NW – North-West region of Lithuania, C – central, SE – South-East; S – small river size, M – medium, L – large, XL – extra-large; R – regulated vs N – natural fragment of the river; see text for specific details of these parameters.

Figure 3. Median values (boxes-whisker plots) of the leaf nitrogen (N) concentration of the groups of populations of the aquatic species

much attention is paid to harvesting either cultivated or natural plants for use as biofuels, such as *P. australis* and *P. arundinacea* (Mäemets et al., 2010; Heinsoo et al., 2011; Butkutė et al., 2014; Pocienė, Kadžiulienė, 2016). High leaf N concentrations in *P. australis* and *P. arundinacea* demonstrate their importance for removal of excess N entering the rivers of Lithuania.

While discussing eutrophication, frequently plant N is related to phytocenological parameters (Yu et al., 2018). According to a recent survey of 80 river sites for macrophyte species in Lithuania (Zviedre et al.,

2015), those with the highest frequency of occurrence and coverage included *N. lutea*. In our study, this species also had the highest N leaf concentration of all species. In the Zviedre et al. (2015) survey, phytocenological parameters were not provided for invasive *B. frondosa* and *E. lobata*, both of which accumulate high N concentrations. Since invasive plants can outcompete native vegetation in environments enriched with N (Fuller et al., 2015; Chmura et al., 2016; Yu et al., 2018), our leaf N data may indicate that the success of alien species (*B. frondosa* and *E. lobata*) in Lithuania may at least partially depend on

elevated N concentrations in the environment of aquatic macrophytes. It can be concluded that the present level of N entering riparian ecosystems is causing spread of the macrophyte species which consume high N levels, although the main sources of aquatic plant N remains to be explained.

Conclusions

- 1. According to the mean values (N % of dry mass) of the leaf nitrogen (N) concentration, species could be arranged into the following order: *Lythrum salicaria* (3.0) < *Stuckenia pectinata* (3.1) < *Phalaris arundinacea* (3.5) < *Bidens frondosa* (3.8) < *Phragmites australis* (4.0) < *Nuphar lutea* (4.1) < *Echinocystis lobata* (4.2).
- 2. Differences in N concentration of the leaves of selected riparian species did not coincide in all cases with the Ellenberg indicatory values, as defined for Central Europe species.
- 3. Differences in leaf N concentrations among the populations were the smallest for *S. pectinata* and the largest for *L. salicaria*.
- 4. Significantly higher (p < 0.05) concentrations of leaf N were found for *L. salicaria* populations growing near the small rivers (3.4% N DM) compared to the large ones (2.8% N DM).
- 5. Leaf N concentration of the selected species was not influenced by riverbed regulations, water state and water former pollution (1991–1996) by agriculture either
- 6. Among water macrophytes studied, the highest N concentration was documented for the invasive Lithuania species *E. lobata*.
- 7. Based on present study leaf N data, the success of aliens *B. frondosa* and *E. lobata* in Lithuania may at least partially depend on elevated N concentrations in the environment of aquatic macrophytes.
- 8. It can be concluded that the present level of N amounts entering riparian ecosystems is big enough to cause the spread of nitrophilic species.

Acknowledgements

The research was funded by project (No. SIT-02/2015) granted by the Research Council of Lithuania. We would like to thank Dr. Ramūnas Vilčinskas for participation in the field work.

Received 07 01 2019 Accepted 31 05 2019

References

- Allen S. E. 1989. Analysis of vegetation and other organic materials. Allen S. E. (ed.). Chemical analysis of ecological materials (2nd ed.). Blackwell Scientific Publications, Oxford and London, p. 46–61.
- Anderson N. O., Jocienė L., Krokaitė E., Rekašius T., Paulauskas A., Kupčinskienė E. 2018. Genetic diversity of *Phalaris arundinacea* populations in relation to river regulation in the Merkys basin, Lithuania. River Research and Applications, 34 (4): 1–10. https://doi.org/10.1002/rra.3259

- Banks S. C., Cary G. J., Smith A. L., Davies I. D., Driscoll D. A., Gill A. M., Lindenmayer D. B., Peakall R. 2013. How does ecological disturbance influence genetic diversity? Trends in Ecological Evolution, 28 (11): 670–679. https://doi.org/10.1016/j.tree.2013.08.005
- Bobbink R., Hicks K., Galloway J., Spranger T., Alkemade R., Ashmore M., Bustamante M., Cinderby S., Davidson E., Dentener F., Emmett B., Erisman J. W., Fenn M., Gilliam F., Nordin A., Pardo L., De Vries W. 2010. Global assessment of nitrogen deposition effects on terrestrial plant diversity: a synthesis. Ecological Applications, 20 (1): 30–59. https://doi.org/10.1890/08-1140.1
- Brezinová T., Vymazal J. 2015. Nitrogen standing stock in *Phragmites australis* growing in constructed wetlands

 do we evaluate it correctly? Ecological Engineering, 74: 286–289.
 https://doi.org/10.1016/j.ecoleng.2014.10.017
- Butkutė B., Lemežienė N., Kanapeckas J., Navickas K., Dabkevičius Z., Venslauskas K. 2014. Cocksfoot, tall fescue and reed canary grass dry matter yield, chemical composition and biomass convertibility to methane.
 - Biomass and Bioenergy, 66: 1–11. https://doi.org/10.1016/j.biombioe.2014.03.014
- Chmura D., Krywult M., Kozak J. L. 2016. Nitrate reductase activity (NRA) in invasive alien *Fallopia japonica*: seasonal variation, differences among habitats types and comparison with native species. Acta Societatis Botanicorum Poloniae, 85 (3): 3514. https://doi.org/10.5586/asbp.3514
- CLC. 2006. CORINE Land Cover Nomenclature Conversion to Land Cover Classification System. http://www.igeo.pt/ gdr/pdf/CLC2006 nomenclature addendum.pdf
- CPST. 2015. Critical loads of oxidized sulphur, oxidized and nutrient nitrogen. 2015. http://oras.gamta.lt/files/ Critical%20Loads.pdf
- 10. De Caceres M., Legendre P., Moretti M. 2010. Improving indicator species analysis by combining groups of sites. Oikos, 119 (1): 1674–1684. https://doi.org/10.1111/j.1600-0706.2010.18334.x
- Ellenberg H., Weber H. E., Düll R., Wirth V., Werner W., Paulißen D. 1992. Zeigerwerte der Pflanzen in Mitteleuropa. Scripta Geobotanica, XVIII, 258 p. (in German).
- 12. EMEP. 2017. The European Monitoring and Evaluation Programme Status Report 1/2017. Transboundary particulate matter, photo-oxidants, acidifying and eutrophying components. http://emep.int/publ/reports/2017/EMEP_Status_Report_1_2017.pdf
- Erős T., Kuehne L., Dolezsaib A., Sommerwerk N., Wolter C. 2019. A systematic review of assessment and conservation management in large floodplain rivers – actions postponed. Ecological Indicators, 98: 453–461. https://doi.org/10.1016/j.ecolind.2018.11.026
- 14. EU. 2000. Directive 2000/60/EC of the European Parliament and of the Council of 23 October 2000 establishing a framework for Community action in the field of water policy. The Official Journal of the European Communities, L 327/1–327/72.
- 15. Fuller M. R., Doyle M. W., Strayer D. L. 2015. Causes and consequences of habitat fragmentation in river networks. Annals of the New York Academy of Sciences, 1355: 31–51. https://doi.org/10.1111/nyas.12853
- 16. Gailiušis B., Jablonskis J., Kovalenkovienė M. 2001. Lithuanian rivers. Hydrography and runoff. Lithuanian Energy Institute, 791 p. (in Lithuanian).
- 17. Han W., Fang J., Guo D., Zhang Y. 2005. Leaf nitrogen and phosphorus stoichiometry across 753 terrestrial plant species in China. New Phytologist, 168 (2): 377–385. https://doi.org/10.1111/j.1469-8137.2005.01530.x

18. Heinsoo K., Hein K., Melts I., Holm B., Ivask M. 2011. Reed canary grass yield and fuel quality in Estonian farmers' fields. Biomass and Bioenergy, 35 (1): 617-625. https://doi.org/10.1016/j.biombioe.2010.10.022

Zemdirbyste-Agriculture

- 19. Hering D., Carvalho L., Argillier C., Beklioglu M., Borja A., Cardoso A. C., Duel H., Ferreira T., Globevnik L., Hanganu J., Hellsten S., Jeppesen E., Kodeš V., Solheim A. L., Nõges T., Ormerod D., Panagopoulos Y., Schmutz S., Venohr M., Birk S. 2015. Managing aquatic ecosystems and water resources under multiple stress - an introduction to the MARS project. Science of the Total Environment, 503-504: 10-21. https://doi.org/10.1016/j.scitotenv.2014.06.106
- 20. Hettelingh J. P., Posch M., Slootweg J. (eds.). 2009. Critical load, dynamic modelling and impact assessment in Europe: CCE Status Report 2008, Coordination Centre for Effects, Netherlands Environmental Assessment Agency, 234 p.
- 21. Hille S., Larsen S. E., Rubæk G. H., Kronvang B., Baattrup-Pedersen A. 2018. Does regular harvesting increase plant diversity in buffer strips separating agricultural land and surface waters? Frontiers in Environmental Science, 6: 58. https://doi.org/10.3389/fenvs.2018.00058
- 22. Kopec D., Ratajczyk N., Wolanska-Kaminska A., Walisch M., Kruk A. 2014. Floodplain forest vegetation response to hydroengineering and climatic pressure a five decade comparative analysis in the Bzura River valley (Central Poland). Forest Ecology and Management, 314: 120-130.
 - https://doi.org/10.1016/j.foreco.2013.11.033
- 23. Kupcinskiene E. 2001. Nitrogen fertilizer factory effects on the amino acid and nitrogen content in the needles of Scots pine. The Scientific World Journal, 1 (2): 449-456. https://doi.org/10.1100/tsw.2001.386
- 24. Mäemets H., Palmik K., Haldna M., Sudnitsyna D., Melnik M. 2010. Eutrophication and macrophyte species richness in the large shallow North-European Lake Peipsi. Aquatic Botany, 92 (4): 273-280. https://doi.org/10.1016/j.aquabot.2010.01.008
- 25. Moldan F., Wright R. F. 2011. Nitrogen leaching and acidification during 19 years of NH₄NO, additions to a coniferous-forested catchment at Gardsjon, Sweden (NITREX). Environment Pollution, 159 (2): 431-440. https://doi.org/10.1016/j.envpol.2010.10.025
- 26. O'Hare M. T., Baattrup-Pedersen A., Baumgarte I., Freeman A., Gunn I. D. M., Lázár A. N., Sinclair R., Wade A. J., Bowes M. J. 2018. Responses of aquatic plants to eutrophication in rivers: a revised conceptual model. Frontiers in Plant Science, 9: 451. https://doi.org/10.3389/fpls.2018.00451
- 27. Pinay G., Bernal S., Abbott B. W., Lupon A., Marti E., Sabater F., Krause S. 2018. Riparian corridors: a new conceptual framework for assessing nitrogen buffering across biomes. Frontiers in Environmental Science, 6: 47. https://doi.org/10.3389/fenvs.2018.00047
- 28. Pocienė L., Kadžiulienė Ž. 2016. Biomass yield and fibre components in reed canary grass and tall fescue grown as feedstock for combustion. Zemdirbyste-Agriculture, 103 (3): 297-304. https://doi.org/10.13080/z-a.2016.103.038
- 29. Pohlert T. 2014. The pairwise multiple comparison of mean ranks package (PMCMR). R package. https://CRAN.Rproject.org/package=PMCMR
- 30. Povilaitis A. 2011. Nutrient retention in surface waters of Lithuania. Polish Journal of Environmental Studies, 20: 1575–1584.
- 31. Stålnacke P., Aakerøy P. A., Blicher-Mathiesen G., Iital A., Jansons V., Koskiaho J., Kyllmar K., Lagzdins A., Pengerud A., Povilaitis A. 2014. Temporal trends in nitrogen

- concentrations and losses from agricultural catchments in the Nordic and Baltic countries. Agriculture, Ecosystems and Environment, 198: 97-103.
- https://doi.org/10.1016/j.agee.2014.03.028
- 32. Subirana I., Sanz H., Vila J. 2014. Building bivariate tables: the compareGroups package for R. Journal of Statistical Software, 57 (12): 1-16. https://doi.org/10.18637/jss.v057.i12
- 33. Šileika A. S., Wallin M., Gaigalis K. 2013. Assessment of nitrogen pollution reduction options in the river Nemunas (Lithuania) using FyrisNP model. Journal of Environmental Engineering and Landscape Management, 21 (2): 141-152. https://doi.org/10.3846/16486897.2012.663088
- 34. Tan C., Du Y., Zhou J., Wang D., Luo M., Zhang Y., Guo W. 2018. Analysis of different hyperspectral variables for diagnosing leaf nitrogen accumulation in wheat. Frontiers in Plant Science, 9: 674.
 - https://doi.org/10.3389/fpls.2018.00674
- 35. Thompson K., Parkinson J. A., Band S. R., Spencer R. E. 1997. A comparative study of leaf nutrient concentrations in a regional herbaceous flora. New Phytologist, 136 (4): 679-689. https://doi.org/10.1046/j.1469-8137.1997.00787.x
- 36. Tumas R. 1998. Regularities of river water quality under the interactions of physical geography factors and farming intensity. Proceedings of Nordic Hydrological Conference. Helsinki, Finland, p. 100–108.
- 37. Vet R., Artz R. S., Carou S., Shaw M., Ro C. U., Aas W., Baker A., Browersox V. C., Dentener F., Galy-Lacaux C., Hou A., Pienaar J. J., Gillett R., Forti C. M., Gromov S., Hara H., Khodzher T., Mahowald M. N., Nickovic S., Rao P. S. P., Reid N. W. 2014. A global assessment of precipitation chemistry and deposition of sulfur, nitrogen, sea salt, base cations, organic acids, acidity and pH, and phosphorus. Atmospheric Environment, 93: 3-100. https://doi.org/10.1016/j.atmosenv.2013.10.060
- 38. Vyšniauskienė R., Rančelienė V., Naugžemys D., Patamsytė J., Sinkevičienė Z., Butkuvienė J., Žvingila D. 2018. Genetic diversity of populations of Bidens genera invasive and native species in Lithuania. Zemdirbyste-Agriculture, 105 (2): 183-190. https://doi.org/10.13080/z-a.2018.105.024
- 39. Wei Ch., Tang S., Pan Y., Li X. 2016. Plastic responses of invasive Bidens frondosa to water and nitrogen resources. Nordic Journal of Botany, 35 (2): 001-008. https://doi.org/10.1111/njb.01331
- 40. Yoshikawa S., Takahashi H., Sasada Y., Mochizuki H. 2015. Impact of land use on nitrogen concentration in ground water and river water. Soil Science and Plant Nutrition, 61 (6): 898–909.
 - https://doi.org/10.1080/00380768.2015.1104521
- 41. Yu H., Wang L., Liu C., Fan S. 2018. Coverage of native plants is key factor influencing the invasibility of freshwater ecosystems by exotic plants in China. Frontiers in Plant Science, 9: 250. https://doi.org/10.3389/fpls.2018.00250
- 42. Zviedre E., Vītola I., Vizule-Kahovska L., Upena I. 2015. Evaluation of phytobenthos and macrophytes of the inland surface waters and ecological status defined by macrophyte reference index: report. Part II. Rivers. Latvian Environment, Geology and Meteorology Centre, 234 p. (in Lithuanian). http://vanduo.gamta.lt/cms/index?rubricId=a65cce5b-64c7-445f-883e-af3e11213469.

ISSN 1392-3196 / e-ISSN 2335-8947 Zemdirbyste-Agriculture, vol. 106, No. 3 (2019), p. 203–212 DOI 10.13080/z-a.2019.106.026

Vandens augalų rūšių azoto koncentracijų sąsajos su žemės dangos tipu ir kitais aplinkos veiksniais

- E. Krokaitė¹, D. Shakeneva¹, E. Juškaitytė¹, T. Rekašius^{1,2}, J. Nemaniūtė-Gužienė¹,
- J. Butkuvienė³, J. Patamsytė³, V. Rančelienė⁴, R. Vyšniauskienė⁴, L. Duchovskienė⁵,
- L. Jocienė¹, Z. Sinkevičienė⁴, D. Naugžemys³, V. Kleizaitė³, D. Chmura⁶, N. O. Anderson⁷,
- D. Žvingila³, E. Kupčinskienė¹
- ¹Vytauto Didžiojo universitetas, Lietuva
- ²Vilniaus Gedimino technikos universitetas, Lietuva
- ³Vilniaus universitetas, Lietuva
- ⁴Gamtos tyrimų centras, Lietuva
- ⁵Lietuvos agrarinių ir miškų mokslų centro Sodininkystės ir daržininkystės institutas
- ⁶Bielsko-Biala universiteto Aplinkos apsaugos ir inžinerijos institutas, Lenkija
- ⁷Minesotos universitetas, JAV

Santrauka

Baltijos šalių azoto (N) iškritų duomenys kartu su sausumos vandenų rodikliais rodo, kad azoto apkrova gali lemti augmenijos pokyčius. Nors upių eutrofikacija yra plačiai analizuojama, vis dar trūksta informacijos apie pakrantės augalų rūšių fiziologinius rodiklius. Tyrimo tikslas – palyginti Lietuvos vandens pakrančių augalų rūšių populiacijų lapų azoto koncentracijų duomenis, juos susiejant su tyrimo vietoms pagal gretimų žemių dangos tipu ir naudojimo paskirtimi, taikant aplinkos informacijos koordinuotos sistemą (CORINE), upių būkle bei dydžiu, žemės ūkio intensyvumu 1991–1996 metais ir upių vagų keitimu. Tyrimo medžiagą surinkus per 2015 m. rugpjūčio mėn. pirmąjį dešimtadienį iš pagrindinių Lietuvos upių 241 vietos, buvo įvertintos plačiai paplitusių penkių pakrančių ir dviejų vandens augalų (natūralių ir invazinių) azoto koncentracijos. Analizėms naudoti augalų lapai, kurių azoto koncentracijos buvo nustatytos Kjeldalio metodu.

Didžiausi (1,7 karto, p < 0,05) lapų azoto koncentracijos skirtumai tarp populiacijų nustatyti *Lythrum salicaria*. Pagal populiacijų lapų azoto koncentracijų (N % sausos masės, SM) vidutines vertes, tirtų rūšių augalai išdėstė taip: *Lythrum salicaria* (3,0) < Stuckenia pectinata <math>(3,1) < Phalaris arundinacea (3,5) < Bidens frondosa (3,8) < Phragmites australis (4,0) < Nuphar lutea (4,1) < Echinocystis lobata (4,2). Iš esmės didesnės <math>(p < 0,05) lapų azoto koncentracijos buvo nustatytos *Lythrum salicaria* populiacijų, augančių šalia mažų upių (3,4 % N SM) nei augančių šalia didelių upių (2,8 % N SM). Skirtingomis savybėmis pasižyminčiose upių vietose rinkti vandens augalai reikšmingai nesiskyrė pagal lapų azoto koncentracijas. Vandens augalų lapų N koncentracijos nesiskyrė tarp įvairios paskirties žemės tyrimo vietų (2000 ir 2006 m. duomenų bazės duomenys) ir upių fragmentų, besiskiriančių dydžiu arba būkle, įvertinta pagal Vandens pagrindų direktyvos gaires. Iš visų tirtų augalų nitrofiliškiausia buvo invazinė Lietuvoje rūšis – *Echinocystis lobata*, kuri pastaruoju metu plinta palei didesnio dydžio (>1000 km²) upes. Galima teigti, kad į pakrančių ekosistemas patenkantis azotas sukelia gausiai azotą naudojančių makrofitų rūšių plitimą, tačiau pagrindiniai makrofitų azoto šaltiniai dar nėra nustatyti.

Reikšminiai žodžiai: pakrančių augalai, upės būklė, upių reguliavimas, vandens makrofitai, žemės dangos tipas.