Factors Influencing the Chemical Characteristics and Nutrient Retention / Release Potential of Wetlands in the Middle (Hungarian) and Lower (Bulgarian) Danube River

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Abstract: Several physical and chemical variables including nutrients (N- and P- forms) of wetlands in the Middle and Lower Danube River, each presented by five water bodies, were measured and analysed. The application of multivariate redundancy analysis (RDA) showed that variables reflecting the degree of wetland connectivity (isolation) to the main river and the macrophyte development are among the main and common factors explaining spatial variations of recorded variables, nutrient concentration ratios between wetlands and the river and ordination of wetland sampling sites. The morphological type of wetlands related to their connectivity to the river seems to influence the nutrient dynamics (*i.e.* release/ retention) stronger in the Lower Danube River, while factors such as flow availability and direction are of significant importance in the wetlands of the Middle Danube River.

Keywords: Wetlands, connectivity, ecosystem services, nutrients, trophic status, multivariate analysis

Introduction

Besides sustaining biodiversity, wetlands provide numerous additional ecosystem services, such as nutrient retention, drought and flood mitigation. These important ecological functions and the wetland ecological status as a whole seemed to be better defined by physical and chemical characteristics as shown by Peeters *et al.* (2009) for shallow lakes. Thus, depending on environmental conditions and connectivity, influenced by hydrology, the wetlands might act either as sources or sinks of nutrients (Hein *et al.* 2005, Bondar *et al*. 2007). According to Saunders, Kalff (2001), nitrogen retention in wetlands is higher than in lakes and rivers due to denitrification, uptake by aquatic plants and sedimentation. The alternation of wet and dry phases in wetlands may favor either denitrification or mineralisation of nitrogen (Venterink *et al*. 2002). In contrast, phosphorus, may be either bound in organic or inorganic form to sediments and assimilated by plants, or released by microbial mineralisation and redox-processes; such processes are controlled by the prevailing hydrological conditions (Hillbricht-Ilkovska 1999). In large rivers, in particular the Middle and Lower Danube, the wetlands play an important role in nutrient cycling. In this paper we evaluate the main factors that influence the chemical composition and nutrient retention potential as well as the trophic character of selected wetlands in the Middle and Lower Danube River.

Material and Methods

The Gemenc and Béda-Karapancsa floodplain area in the Middle Danube River extend between rkm 1489 and 1440. Five different wetland units of this large area were investigated, namely: the Grébec-Holt Danube (GDU), Rezéti-Holt-Danube (RDU), Vén-Danube (VDU) and Nyéki-Holt-Danube (NYHD) belonging to Gemenc, and the Mocskos Danube (MDU) belonging to Béda-Karapancsa (Fig. 1). Water sampling in July 2009 included *in situ* measurements of temperature, pH and conductivity by a WTW Multi 403i meter. In the laboratory Na, K, Cl, Ca, Mg, NO_2-N, and SO_4 ions were analysed by DX-120 ion chromatograph (Dionex). Furthermore, TOC (total organic carbon), DOC (dissolved organic carbon), TIC (total inorganic carbon), DIC (dissolved inorganic carbon), TC (total carbon), DTC (dissolved total carbon), TN (total nitrogen), and DTN (dissolved total nitrogen) were determined by a TOC analyzer (Elemetar-liqui-TOC). Standard analytical methods (Golterman *et al.* 1978) were used for determining HCO_3, SPM (suspended particular matter), m-alkalinity, NO_3-N, NH_4-N, PO_4-P, DTP (dissolved total phosphorus), TP (total phosphorus), and Chl-a (chlorophyll-a).

Sampling sites in the Lower Danube River encompassed 3 marshes located on Belene Island (Murtvo Blato, Peschin and Dyulova Bara), the middle of the side arm located opposite to Belene town, and 3 other wetlands (Kalimok Canal, Kalimok Marsh and Brushlen Canal) in the Kalimok-Brushlen protected area (Fig. 2). The water samples were taken in spring, summer and autumn from autumn 2009 to spring 2012. Temperature, pH, conductivity and oxygen concentration were measured *in situ* either by WTW - Multi 1970i or GMH 3510, or Greisinger electronic (pH) and Winkler titration (oxygen). The concentrations of NO_3-N, NO_2–N, NH_4-N, SiO_2, PO_4-P and Fe, as well as TN, TP, COD (chemical oxygen demand), and turbidity at 550 nm as absorbance, were determined colorimetrically by a Nova photometer 60 and kits of Merck. The samples of chlorophyll-a, after being filtered through 0.7 µm glass fiber filters and stored in liquid nitrogen, were

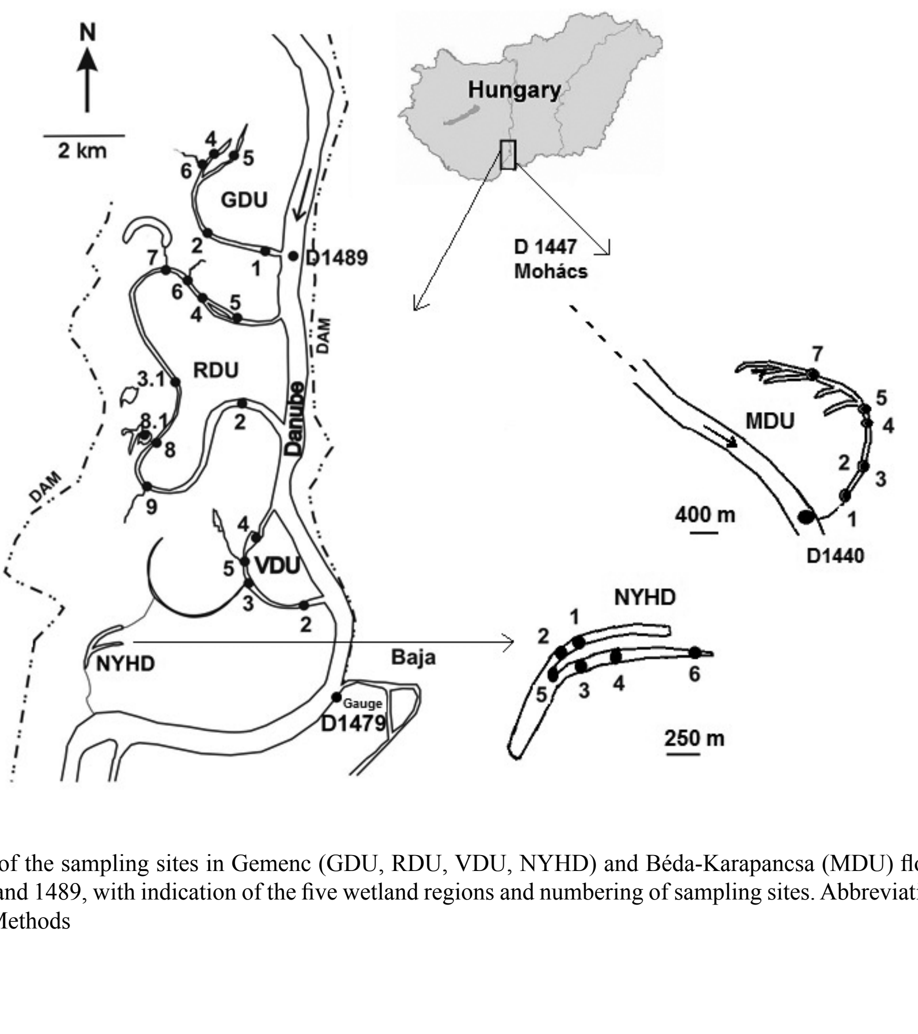

Fig. 1. Location of the sampling sites in Gemenc (GDU, RDU, VDU, NYHD) and Béda-Karapancsa (MDU) floodplain area, between rkm 1440 and 1489, with indication of the five wetland regions and numbering of sampling sites. Abbreviations are explained in Material and Methods

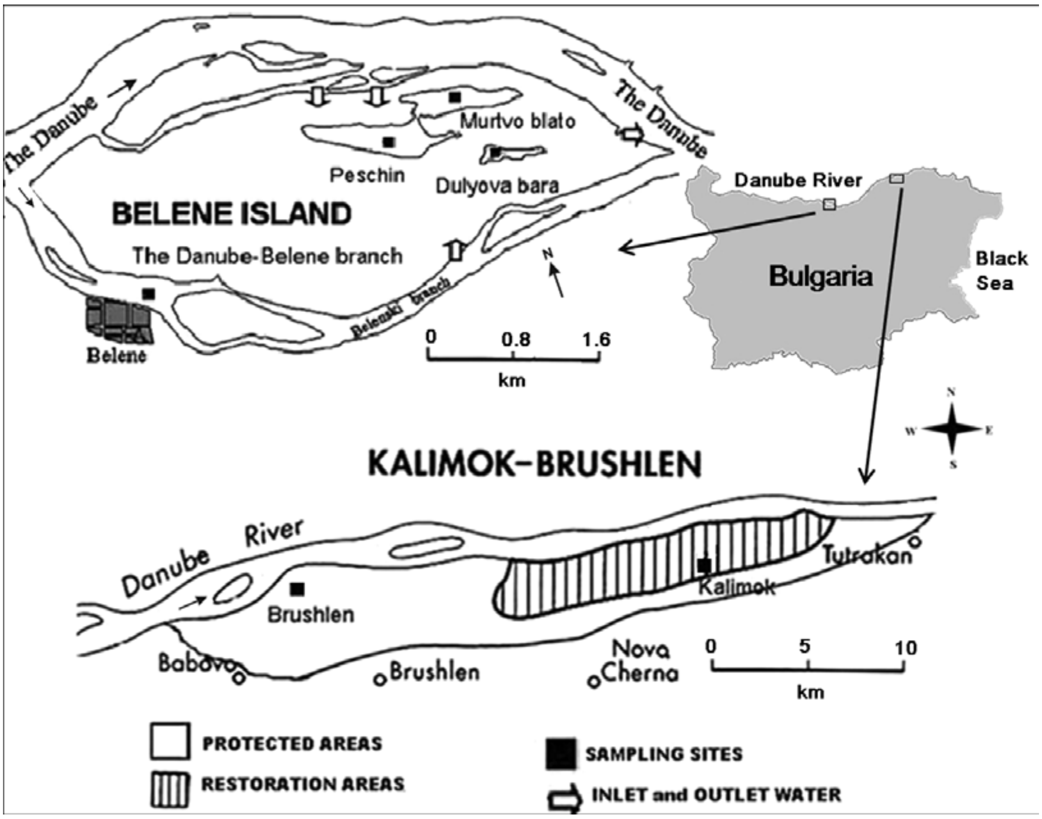

Fig. 2. Location of wetland sites on Belene Island (rkm 561-576): Murtvo Blato, Peschin and Dyulova Bara marshes; and in Kalimok-Brushlen area (rkm 440-465): Kalimok Canal, Kalimok Marsh, Brushlen Canal

analysed in the laboratory according to standard ISO 10260 (ISO 1992). Some additional characteristics of wetland sites, such as water depth, distance to the main river, presence/ absence or direction of flow, degree of connectivity defined by eu-, para-, plesio- and paleopotamal categories (Guti 2001), and percentage of water surface covered by macrophytes, etc., were used as environmental variables in the subsequent statistical analyses.

To find out whether the wetlands are potential sources or sinks of nutrients we calculated the ratios of inorganic N (sum of inorganic nitrogen compounds), PO_4-P, TN and TP concentrations in wetlands to those in the river. The statistical treatment included application of multivariate detrended correspondence analysis (DCA) and redundancy analysis (RDA) by Canoco 4.55 after ter Braak, Šmilauer (2002). The RDA revealed the effect of environmental variables on spatial variations of chemistry data, which in the case of Bulgarian data was done by partial RDA after eliminating the temporal variability by means of covariates. This ensures the comparability with Hungarian data, which do not include temporal variability.

Results and Discussion

Before applying the multivariate analyses we reduced the highly correlated chemical variables ($R \geq 0.80$) to one, since they neither add variance nor explain residual variance. The RDA of Hungarian data clearly separates the more isolated wetlands GDU, NYHD and MDU (plesiopotamal type) from the sites RDU and VDU, which have similar chemistry as the Danube River (Fig. 3). Obviously the almost simultaneous sampling of all sites (within one week) at times of high river water levels (about 5 m) was responsible for this similarity. The resemblance increases at high and decreases at low water levels as reported by Schöll *et al*. (2008) and Ágoston-Szabó *et al*. (2013).

Along with the plesiopotamal degree of isolation, the macrophytes and the outward flow of water as explanatory variables also contribute to the site separation (Fig. 3). The river sites (RDU and VDU) are characterised by high SPM, SO_4 and nitrogen (NO_3-N, TN, DTN) concentrations, while the more isolated sites are rich in TP (GDU), PO_4-P and NH_4-N (NYHD) concentrations, which may have al-

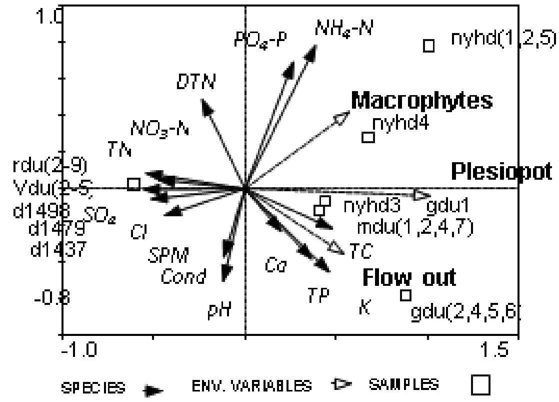

Fig. 3. RDA ordination of chemical variables from the Hungarian Danube River as response variables (species) and macrophyte coverage, Plesiopotamal and water 'flowing out' as explanatory variables (env. variables). The eigenvalues of the first canonical axis (0.451, significant for p=0.002) and of second axis (0.111) together explain 56.2% of total variation. All canonical axes are also significant for p=0.002. The other abbreviations are explained in Material and Methods

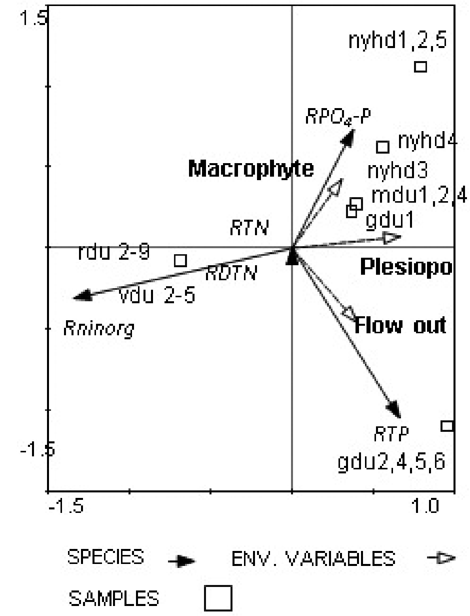

Fig. 4. RDA ordination triplot of wetland to river ratios of nutrient concentrations from the Hungarian Danube as response variables (species) and plesiopotamal type, macrophyte coverage and water 'flowing out' as explanatory variables (env. variables). The eigenvalues of the first canonical axis (0.228, significant for p=0.002) and of second axis (0.128) together explain 39.1% of total variation. All canonical axes are also significant for p=0.002. The other abbreviations are explained in Material and Methods

lochthonous (from the catchment) or autochthonous (from sediments rich in nutrients) origin.

This separation of sites is strongly confirmed by ordination of the ratios of nutrient concentrations between wetlands and the Danube River (Fig. 4). The average values of PO_4-P and TP ratios (1.94 and 1.30, respectively) in wetlands covered by macrophytes (NYHD sites) and the N-inorganic (sum of inorganic N-compounds) ratio of the RDU and VDU sites (1.71, 1.76) are higher than the unity and therefore remote from the coordinate origin (Fig. 4). This means that these sites are potential sources of phosphorus and inorganic nitrogen. The nutrient concentrations at the MDU sites coincide with those at the Danube sites, despite their different chemical composition (Fig. 3). Therefore, the MDU sites seem to be neither a potential sink nor source of phosphorus (ratio average value for PO_4-P 0.97 and for TP 1.05 (Fig. 4). Obviously, the more isolated sites have lower inorganic and total nitrogen concentrations than the river (N-inorganic, TN and DTN ratios 0.36, 0.81 and 0.73 for GDU, 0.31, 0.76 and 0.90 for NYHD, and 0.18, 0.67 and 0.67 for MDU respectively); they might act as nitrogen sinks, especially for inorganic nitrogen, unlike the less isolated RDU, VDU which have either higher N-inorganic ratios (1.71, 1.76) or close to unity ratios for TN and DTN. Therefore, the RDU and VDU wetland sites might be sources of inorganic nitrogen.

The RDA spatial ordination for chemistry data on Bulgarian wetlands shows a clear separation of different water body types – river, drainage canals or marshes – due to low or absent connectivity between the river and wetlands (Fig. 5). The marshes of Belene Island are connected to the Danube River by ground and partly by surface water; the latter being human-regulated and flowing through the wetlands only for short intervals during periods of high water levels. The Kalimok Canal and Kalimok Marsh are also connected by groundwater and periodically inundated by surface water without flow-through effect, while the Brushlen drainage canal serves the collection of groundwater which is pumped back to the river. Therefore, in contrast to the Hungarian wetlands, the Bulgarian wetlands function predominantly as sinks of nutrients. The only explanatory variable with some significance for nutrient ratios is the coverage by emerged macrophytes (Fig. 6). The fact that eigenvalues of RDA for Bulgarian wetlands (Fig. 5) are much lower than those for Hungarian wetlands (Fig. 3) means that the explanatory variables provided and later selected by the analyses better correspond to the situation

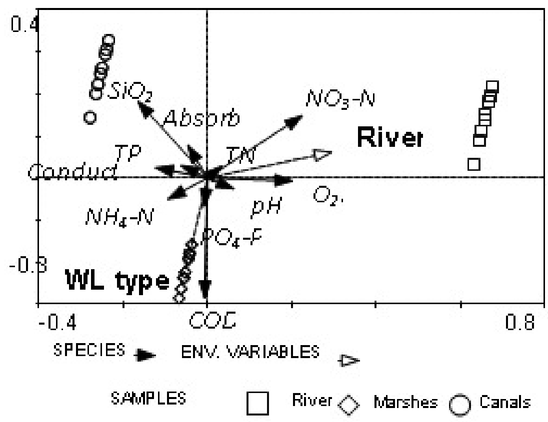

Fig. 5. Partial RDA ordination triplot of chemical characteristics in the Bulgarian Danube River as response variables (species), and type of sites presented as 'river' and type of wetlands (WL type) as 'canal or marshes' as explanatory variables (env. variables). The eigenvalues of the first canonical axis (0.095, significant for p=0.002) and of the second axis (0.049) together explain 18.1% of total variation. All canonical axes are also significant for p=0.002. For abbreviations see Material and Methods

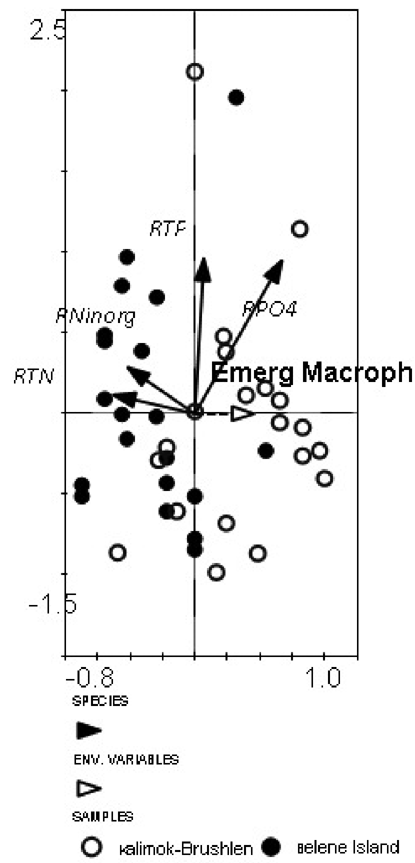

Fig. 6. Partial RDA ordination of sites and ratios between nutrient concentrations from wetlands and the river in the Bulgarian Danube as response variables (species) and % covered by emerged macrophytes, as explanatory variables (env. variables). The eigenvalues of the first canonical axis (0.032, significant for P=0.057) explain 5.7% of total variation. For abbreviations see Material and Methods

in the Middle than in the Lower Danube River. The averages of PO_4-P (2.03) and TP (2.14) ratios for all Bulgarian wetlands are higher, and those of TN (0.87) and N-inorganic (0.84) lower than unity, indicating that the Bulgarian wetlands predominantly retain nitrogen compounds but might act as sources of phosphorus.

The macrophytes seem to have decreasing effect on nitrogen and no effect on phosphorus ratios (Fig. 6) because the N/P ratio is low (geometric mean = 9) indicating nitrogen limitation, which is slightly stronger in Belene (lower N/P ratio) than in Kalimok-Brushlen.

The comparison in Fig. 7 shows that the Bulgarian wetlands have higher TP concentrations than Hungarian wetlands, which in turn show higher TN concentrations. According to TP values, most wetlands are classified as hypertrophic, but according to TN, this is true only for Hungarian wetlands. The lower TN concentrations in Bulgarian wetlands can be explained by macrophyte uptake (Kalimok Marsh) and by more frequent occurrence of anoxic conditions favoring denitrification (Saunders, Kalff 2001, Venterink *et al.* 2002, Hein *et al.* 2005), despite the temporary character and complete drying-out of some Bulgarian wetlands in years with low water as 2011.

Theoretically the permanent Hungarian wetlands should stimulate denitrification and have TN lower than the temporary wetlands in Bulgaria, whose dry periods should promote nitrogen mineralisation. However, the lower TP and high TN concentrations in Hungarian wetlands, especially during the studied summer period of high water levels, might be explained by the flushing effect of river waters washing out nutrient rich sediments and preventing oxygen depletion (Schöll *et al.* 2008). In contrast, such flushing effects in Bulgarian wetlands are minimized or completely absent. Therefore, they function predominantly as nutrient sinks, which leads to the accumulation of nutrients in sediments and frequent occurrence of anoxic conditions favoring denitrification and phosphorus release from sediments. The occasional drying events obviously did not compensate the strong macrophyte development observed during both the aquatic and terrestrial periods, which additionally increases nitrogen uptake, but also provides organic matter whose decomposition leads to more severe oxygen depletion

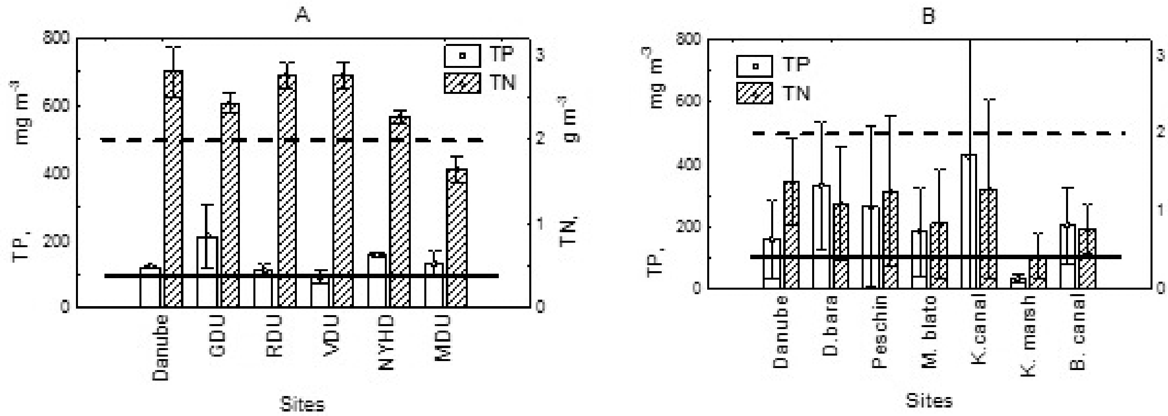

Fig. 7. The averages and standard deviations of total phosphorus (TP) and total nitrogen (TN) concentrations from wetlands and river sites of Hungarian (A) and Bulgarian (B) Danube wetlands. The solid and dotted lines indicate the lower hypertrophy threshold for TP and TN concentrations, respectively (after OECD 1982). The abbreviations of site names are explained in Material and Methods

and stimulation of denitrification during the aquatic phase. While the concentrations of N and P may sufficiently explain the wetland trophic status, a static ratio between concentrations in wetlands and the main river arm has a limited power to define the dynamic processes of wetland nutrient release and retention. Obviously the retention of nutrients is a result of complex of processes of import and export, accumulation and release, composition and decomposition of organic matter, etc. The full understanding of such complex and dynamic processes is a demanding task requiring long lasting simultaneous hydrological and chemical, measurements at in- and output sites. Such data have to be combined into a mass balance model accounting for autochthonous and allochthonuos point and diffuse nutrient sources as well as for the diverse and heterogeneous character of wetland ecosystems.

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References

Ágoston-Szabó E., K. Schöll and M. Dinka 2013. Limnological characteristics of a Danube oxbow-lake (Danube-Drava Natinal Park, Hungary). – *River Systems,* **20** (3-4): 277-287.

Bondar E., V. Kurcera-Hirzinger, S. Preiner, G. Weigelhofer, F. Schiemer and T. Hein 2007. The impact of an artificial water enhancement scheme on phosphorus dynamics in an urban floodplain system in Vienna (Austria). – *Internat. Rev. Hydrobiol.,* **92: (**4–5): 413-427.

Golterman H. L., R. S. Clymo and M. A. M. Ohnstad 1978. Method for physical and chemical analysis of freshwaters. IBP Handbook No 8, Blackwell Sci. Publ., 213 p.

Guti G. 2001. Water bodies in the Gemenc floodplain of the Danube, Hungary (A theoretical basis for their typology). – *Opusc. Zool., Budapest*, **33**: 49-60.

Hein T., W. Reckendorfer, J. H. Thorp and F. Schiemer 2005. The role of slackwater areas for biogeochemical processes in rehabilitaed river corridors: examples from the Danube. – *Large Rivers Vol. 15, No1-4, Arch. Hydrobiol. Suppl.,* **155** (1-4): 425-442.

Hillbricht-Ilkovska A. 1999. Shallow lakes in lowland river systems: Role in transport and transformations of nutrients and in biological diversity. – *Hydrobiologia,* **408/409:** 349–358.

ISO 10260, 1992. Water quality – Measurement of biochemical parameters – Spectrometric determination of the chlorophyll-a concentration, p. 1-4.

OECD 1982. Eutrophication of waters: Monitoring, assessment and control. Vollenweider R. A., J. Kerekes, OECD, Paris.

Peeters E. T. H. M., R. J. M. Franken, E. Jeppesen, B. Moss, E. Becares, L.-A. Hansson, S. Romo, T. Kairesalo, E. M. Gross, E. van Donk, T. Nõges, K. Irvine, R. Kornijów and M. Scheffer 2009. Assessing ecological quality of shallow lakes: Does knowledge of transparency suffice? – *Basic and Applied Ecology,* **10**: 89–96.

Saunders D. L., J. Kalff 2001 Nitrogen retention in wetlands, lakes and rivers. – *Hydrobiologia,* **443**: 205–212.

Schöll K., M. Dinka, A. Kiss and E. Ágoston-Szabó 2008. Hydrobiological surveys in specific hydrological situations at the Gemenc floodplain of the Danube (Hungary). 4th ECRR Conference on River Restoration, Italy, Venice S. Servolo Island 16-21 June 2008, 141-148.

ter Braak C. J. F., P. Šmilauer 2002. Canoco Reference Manual and CanoDraw for Windows, User's Guide, Biometris, Wageningen and České Budějovice 499 p.

Venterink H. O., T. E. Davidsson, K. Kiehl and L. Leonardson 2002. Impact of drying and re-wetting on N, P and K dynamics in a wetland soil. – *Plant and Soil,* **243**: 119-130.