Dynamics of inorganic forms of nitrogen in soil
of the Nature Reserve Alúvium Žitavy

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Abstract


There was studied dynamics of inorganic soil nitrogen in the Alluvium Žitavy Nature Reserve in the years 2008 and 2009. We observed variability in the individual inorganic nitrogen forms in soil samples differing in the sampling depth, soil moisture content, sampling site and date of the sampling. After comparing the contents of N$_{in}$ over the research period, we found that higher values were in the first year (2008). Ammonium nitrogen values ranged from 3.80–16.87 mg kg$^{-1}$ and its average over the research period was 7.24 mg kg$^{-1}$. The contents of this N form in 2008 were higher than in 2009. Nitrate nitrogen displayed the opposite trend. The values of nitrate nitrogen were within 0.44 to 9.28 mg kg$^{-1}$ with an average of 2.03 mg kg$^{-1}$. The contents of this N form were higher in 2009. The content of ammonium nitrogen was found affected significantly by the year and date of sampling. The content of nitrate nitrogen was highly significantly influenced by the soil depth, year and sampling site. The coefficients of variation were high, consequently, there have not been identified correlations between the soil moisture content and amounts of the studied forms of inorganic nitrogen. The low values of nitrate nitrogen do not indicate a risk of pollution either of the groundwater or surface water.

Key words

ammonium nitrogen, nitrate nitrogen, soil, wetland

Introduction

Nitrogen is essential for growth and reproduction of all life forms, and except for legume crops and virgin soils with relatively high soil organic matter, soil N must usually be supplemented to sustain food, feed, and fiber production (DINNES et al., 2002).

Due to its importance in the functioning of different ecosystems, dynamics of microbial biomass and its role in plant nutrition under different ecosystem conditions is of considerable significance (TRIPATHI and SINGH, 2006).

Wetlands are subjected to changes in soil moisture as a result of both short-term seasonal climate variations and long-term changes in regional water resource management, both of which can modify the dynamics of ground and surface water inputs (YU and EHRENFELD, 2009).

Nitrogen cycling in wetland soils is thought to be highly sensitive to variations in soil moisture, which is a controlling variable affecting the redox potential of the soil. As wetland soils become increasingly anoxic, oxygen-dependent processes, in particular nitrification, are reduced or eliminated. At the same time, processes that require anoxic conditions, notably denitrification, may increase their rate (PINAY et al., 2002). Soil moisture in its turn varies with soil properties such as texture and organic matter content, which affect the water-holding capacity of the soils (BRULAND and RICHARDSON, 2004; WENDROTH et al., 2006; SLEUTEL et al., 2008), and the balance between inputs and outputs in the water budget of the soil profile. Changes to water budgets can be
critical in wetlands, where small alterations to inputs of precipitation, surface and groundwater can have large effects on the moisture status of the soil. Variations in climate, both seasonal and inter-annual, and long-term changes in regional water resource management can both cause change in the moisture content of wetland soils.

**Material and methods**

This experiment was pursued in the Nature Reserve (NR) Alúvium Žitavy. Located in the south-western part of Slovakia, in the cadastral territory of the town Hurbanovo and village Martovce. It is stretched along the lower stream Žitava, which is a part of the geomorphologic unit Podunajská nížina lowland. The NR with an acreage of 32.53 hectares was declared as a protected area in 1993, and its territory belongs to the 4th degree of protection. This protection has been declared for a woodland meadow with many diverse plant and animal species, including nesting birds. The area is characterized by a great diversity of habitats representing water vegetation, swamps, and floodplain forests. There are willow-poplar forests with rich scrub undergrowth providing shelters for many animal species. The protected plant species are *Nuphar lutea*, *Salix sp.*, *Iris pseudacorus*, *Leucojum aestivum*, and several others.

Much of the site area is flooded over the year, especially in spring. The fauna of the NR, is very rich in biodiversity. The ichthyofauna is represented by the species *Lepomis gibbosus*, *Perca fluviatilis*, *Carassius carassius*, and several others, the amphibians by the species *Bombina bombina*, *Hyla arborea*, *Rana esculenta*, and others, *Herpetofauna* by species of the genera *Lacerta* sp., *Natrix natrix*, and others, the birds by *Ardea purpurea*, *Falco cherrug*, *Numenius argus* and others and the mammals by *Ondatra zibethica*, *Martes martes*, *Talpa europaea*, *Valpes vulpes*, and many others. Because the NR is directly adjacent to the surrounding agro­ceneses, we can expect also them rich in animal species. The soils recorded in the Reserve are: Haplic Chernozems, Haplic Fluvisols and Haplic Histosols, moderate to severely polluted. The riparian vegetation and plant scrub are good refuges for fauna, especially for avifauna, and they are also an important element in landscape-forming (Palatická, 2009).

The soil samples were collected over the whole two-year research period at four sampling sites in the Nature Reserve Alúvium Žitavy:

**Sampling site 1 (SS1)** – a dense grassland, by a sparse willow (*Salix sp.*) stand – collector can be characterized as a typical wetland ecosystem.

**Sampling site 2 (SS2)** – a dense grassland, with dense vegetation consisting of phragmites (*Phragmites australis*) and willow (*Salix sp.*).

**Sampling site 3 (SS3)** – is covered with a dense grass vegetation, mostly flooded (during snow melting in spring intense rainfall episodes in summer when Žitava River flows over its bed).

**Sampling site 4 (SS4)** – a thick grassy vegetation, growing on the banks, consisting of cattail (*Typha latifolia*), phragmites (*Phragmites australis*), alder (*Alnus sp.*) and willow (*Salix sp.*).

The contents of inorganic forms of nitrogen (N-NO₃⁻ and N-NH₄⁺) were measured in soil samples taken from sampling sites at the boundary of the Nature Reserve. The soil was sampled monthly from two depths (0.0–0.3 m and 0.3–0.6 m). In these samples, we determined the contents of inorganic nitrogen forms in 1% solution of potassium sulphate (K₂SO₄), with using the following methods:

- N-NO₃⁻ – colour method by acid phenoldihydrosulphide
- N-NH₄⁺ – colour method by Nessler’s test solution.

The soil moisture content was determined by gravimetric method (in weight percent). For evaluation of pH value, a 1 mol dm⁻³ solution of potassium chloride was used.

The obtained values of inorganic nitrogen forms were presented in tables, figures and evaluated statistically. Because the data set did not show normal distribution, for statistical evaluation were used the Kruskal-Wallis test. Pearson’s correlation coefficient was used to express the correlation between the chosen evaluated factors (Program Statgraphics Plus 5.0.1).

**Results and discussion**

The average content of ammonium nitrogen (N-NH₄⁺ ) during the whole research period was 7.24 mg kg⁻¹ (Table 1), with a standard deviation of 2.91 mg kg⁻¹ (Table 2), which is higher than the value found in Žitavský luh by Kantor and Ondrišík (2005). The content of ammonium nitrogen was in the range 3.80–16.87 mg kg⁻¹ (Table 1). Over the whole period, the coefficient of variance for the ammonium nitrogen was 40.18%, which is twice higher than those detected by Kantor and Ondrišík (2005). The statistical data for the content
of ammonium nitrogen in the soil show a statistically high significant effect (significant level $\alpha = 0.01$) (Table 3) of the year and the sampling date. The sampling depth and sampling site showed statistically significant effects, too.

In summary, the highest contents of $\text{N-NH}_4^+$ were determined in the year 2008 (16.87 mg kg$^{-1}$) (Table 1). The Fig. 1 illustrates the dynamics of ammonium nitrogen in the two study years in the upper soil layer. We can see that the content of ammonium nitrogen had a decreasing character, but during the first year (2008), the decrease was more marked. The large decreases in soil N content after soil disturbances reported in this and other studies (EVANS and EHLERINGER, 1993) suggest that disturbance can modify the balance between N input and loss, causing a decrease in N storage within the

Table 1. Average contents of inorganic nitrogen forms over the whole research period (mg kg$^{-1}$)

<table>
<thead>
<tr>
<th>Monitored parameters</th>
<th>N-$\text{NH}_4^+$</th>
<th>N-$\text{NO}_3^-$</th>
<th>$\text{N}_\text{an}$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Year 2008</td>
<td>9.16</td>
<td>1.76</td>
<td>10.93</td>
</tr>
<tr>
<td>Year 2009</td>
<td>5.33</td>
<td>2.30</td>
<td>7.63</td>
</tr>
<tr>
<td>Depth 0.0–0.3</td>
<td>7.43</td>
<td>2.33</td>
<td>9.76</td>
</tr>
<tr>
<td>Depth 0.3–0.6</td>
<td>7.06</td>
<td>1.74</td>
<td>8.80</td>
</tr>
<tr>
<td>Sampling site 1</td>
<td>7.10</td>
<td>2.16</td>
<td>9.27</td>
</tr>
<tr>
<td>Sampling site 2</td>
<td>7.36</td>
<td>2.32</td>
<td>9.68</td>
</tr>
<tr>
<td>Sampling site 3</td>
<td>7.37</td>
<td>2.04</td>
<td>9.41</td>
</tr>
<tr>
<td>Sampling site 4</td>
<td>7.14</td>
<td>1.62</td>
<td>8.76</td>
</tr>
</tbody>
</table>

Table 2. Basic statistical characteristic of measured variables (N-$\text{NO}_3^-$, N-$\text{NH}_4^+$, $\text{N}_\text{an}$)

<table>
<thead>
<tr>
<th>Statistical characteristic</th>
<th>N-$\text{NH}_4^+$</th>
<th>N-$\text{NO}_3^-$</th>
<th>$\text{N}_\text{an}$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number of observation (n)</td>
<td>128</td>
<td>128</td>
<td>128</td>
</tr>
<tr>
<td>Average (x)</td>
<td>7.24</td>
<td>2.03</td>
<td>9.28</td>
</tr>
<tr>
<td>Standard deviation (s)</td>
<td>2.91067</td>
<td>1.26986</td>
<td>3.28249</td>
</tr>
<tr>
<td>Standard error ($S_x$)</td>
<td>0.25727</td>
<td>0.112241</td>
<td>0.290134</td>
</tr>
<tr>
<td>Minimum</td>
<td>3.80</td>
<td>0.44</td>
<td>4.71</td>
</tr>
<tr>
<td>Maximum</td>
<td>16.87</td>
<td>9.28</td>
<td>25.59</td>
</tr>
<tr>
<td>Coefficient of variation % (V)</td>
<td>40%</td>
<td>62%</td>
<td>35%</td>
</tr>
</tbody>
</table>

Table 3. Kruskal-Wallis analyses

<table>
<thead>
<tr>
<th>Nitrogen form</th>
<th>Source of variability</th>
<th>Test statistics (K–W)</th>
<th>Significant level</th>
</tr>
</thead>
<tbody>
<tr>
<td>N-$\text{NH}_4^+$</td>
<td>Depth</td>
<td>1.06</td>
<td>0.3022</td>
</tr>
<tr>
<td></td>
<td>Sampling site</td>
<td>2.15</td>
<td>0.5424</td>
</tr>
<tr>
<td></td>
<td>Sampling date</td>
<td>20.79</td>
<td>0.0041</td>
</tr>
<tr>
<td></td>
<td>Year</td>
<td>76.72</td>
<td>0.0000</td>
</tr>
<tr>
<td>N-$\text{NO}_3^-$</td>
<td>Depth</td>
<td>20.31</td>
<td>0.0000</td>
</tr>
<tr>
<td></td>
<td>Sampling site</td>
<td>10.52</td>
<td>0.0146</td>
</tr>
<tr>
<td></td>
<td>Sampling date</td>
<td>11.05</td>
<td>0.1363</td>
</tr>
<tr>
<td></td>
<td>Year</td>
<td>20.52</td>
<td>0.0000</td>
</tr>
<tr>
<td>$\text{N}_\text{an}$</td>
<td>Depth</td>
<td>6.85</td>
<td>0.0089</td>
</tr>
<tr>
<td></td>
<td>Sampling site</td>
<td>3.61</td>
<td>0.3072</td>
</tr>
<tr>
<td></td>
<td>Sampling date</td>
<td>3.16</td>
<td>0.0000</td>
</tr>
<tr>
<td></td>
<td>Year</td>
<td>39.99</td>
<td>0.0000</td>
</tr>
</tbody>
</table>
soil (Evans and Belnap, 1999). The average content at the first depth (surface layer) was 9.50 mg kg\(^{-1}\), whereas at the second depth it was 8.86 mg kg\(^{-1}\) (Fig. 2).

In terms of sampling sites, the content of ammonium nitrogen ranged from 7.10–7.37 mg kg\(^{-1}\). The highest content was observed in the sampling site 3, the lowest in SS 1. The same trend was also found in Žitavský luh (Kantor and Onoříšík, 2005). These authors detected the highest concentrations of ammonium nitrogen in sample sites representing a typical wetland ecosystem and the lowest in sample sites representing meadow ecosystems.

Significantly dropped contents of ammonium nitrogen were recorded on the first four sampling dates in 2008 at both soil depths.

Fig. 1. Dynamics of ammonium nitrogen at a depth of 0.0–0.3 m in the years 2008 and 2009

Fig. 2. Dynamics of ammonium nitrogen at a depth of 0.3–0.6 m in the years 2008 and 2009
In comparison with the ammonium nitrogen, the nitrate nitrogen showed slower dynamics over the whole research period. The average content of nitrate nitrogen over the whole research period (2008 and 2009) was 2.03 ± 1.27 mg kg⁻¹ (Table 1, Table 2). This low concentration does not mean a threat to the quality either of groundwater or of surface water. Its value is even lower than the one obtained by Kantor and Onáriš (2005) in the wetland Žitavský luh. Gilliam et al. (1999) report that the inorganic N forms of nitrogen were extractable almost equally to NO₃ and NH₄ in old field soils. The anaerobic conditions in the swamp soils, however, prevent nitrification. Pinelands soils, including upland and mineral wetland soils, in general show very low or zero rates of net nitrification, and no or in very low concentrations nitrates in soil pore water (Ehrenfeld et al., 1997a, b).

The interval in both years was 0.44–9.28 mg kg⁻¹ (Table 1). For nitrate nitrogen was found a higher coefficient of variance (62.43%) than for ammonium nitrogen.

This higher variation coefficient values were connected with marked nitrate nitrogen – see Fig. 6. The exceptions were observed in case of the first sampling date, the second sampling site, at the both sampling depths. The content of nitrate nitrogen on the first sampling date was 9.11 mg kg⁻¹ (the first depth 0.0–0.3 m) (Fig. 3) and 9.28 mg kg⁻¹ (the second depth 0.3–0.6 m) (Fig. 4). Many observations of drained wetlands confirm increases in both soluble inorganic N content and N process rates (Regina et al., 1996, 1999; Freeman et al., 1997; Olde et al., 2002; Tiemeyer et al., 2007). Similar responses were observed in riparian wetlands drying out during summer months (Bechtold and Naiman, 2006). However, Regina et al. (1996) found that there were differences in effects of drainage on nitrate production between minerotrophic fens and ombrotrophic bogs, suggesting that soil characteristics control the response of wetland soils to drying. In a series of studies, Bridgham (1995) and colleagues (Udegraft et al., 1995; Bridgham et al., 1998) show that many types of histosols had higher rates of N mineralization and nitrification when they were incubated under aerobic conditions than in case of anaerobic conditions. However, the authors found that the relative change in nitrification rate, particularly the share of nitrification in proportion to the total net mineralization, varied considerably among the soil types, reflecting differences in the substrate quality. In mineral wetland soils, N mineralization reaches maximum at intermediate levels of soil moisture (57–78% waterfilled pore space) (Sleutel et al., 2008); the precise values vary with soil texture and soil organic matter amount.

More variability was also observed during the second year (2009) in all sampling sites (Fig. 4). The highest content of nitrate nitrogen was found on the first sampling date in the first sampling site (5.8 mg kg⁻¹), and a more marked increase was identified on four sampling dates (IV.2009, V.2009, VI.2009, VII.2009) at the second sampling depth. In the case of nitrate nitrogen, statistically high significant effect was obtained for the year and sampling depth. Statistical significant effect (significant level α = 0.05) was found only in the sampling site (Table 3).

As for sampling sites, the content of ammonium nitrogen ranged between 1.62–2.32 mg kg⁻¹. The highest content was found in the sampling site 2, the lowest in SS 4.
Fig. 4. Dynamics of nitrate nitrogen in depth of 0.3–0.6 m in the years 2008 and 2009

Fig. 5. Dynamics of ammonium nitrogen at depths 0.0–0.3 m and 0.3–0.6 m in the years 2008 and 2009

Fig. 6. Dynamics of nitrate nitrogen at depths 0.0–0.3 m and 0.3–0.6 m in the years 2008 and 2009
The comparison between the two nitrogen forms (ammonium and nitrate) shows that the values ammonium nitrogen were almost the same at the both sampling depths (Fig. 5); on the other hand, there were some differences in the contents of nitrate nitrogen between the depths (Fig. 6). The average value of ammonium nitrogen at the first depth was 7.43 mg kg$^{-1}$ and at the second depth 7.06 mg kg$^{-1}$. In the case of nitrate nitrogen, the average nitrogen content value at the first depth was 2.33 mg kg$^{-1}$, at the second depth 1.74 mg kg$^{-1}$.

In the Table 2 we can see that the average value of N-NH$_4^+$ content (7.24 mg kg$^{-1}$) was higher than the content of the N-NO$_3^-$ (2.03 mg kg$^{-1}$). Gilliam et al. (1998) found out that anaerobic conditions in the swamp soils, prevent nitrification; consequently, in the swamp soils there was only a little amount of nitrogen in the form of NO$_3^-$ and more than 98% of extractable nitrogen was present in the form of NH$_4^+$.

The next statistic indicator – the Pearson’s correlation coefficient indicated only very weak relations between N-NO$_3^-$ and N-NH$_4^+$.

Table 4. Pearson’s correlation coefficients with fitting P-values

<table>
<thead>
<tr>
<th>Pairs</th>
<th>Pearson’s correlation coefficient</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>N$_m$ – N-NO$_3^-$</td>
<td>0.4696</td>
<td>0.0000</td>
</tr>
<tr>
<td>N$_m$ – N-NH$_4^+$</td>
<td>0.9228</td>
<td>0.0000</td>
</tr>
<tr>
<td>N$_m$ – moisture</td>
<td>0.3322</td>
<td>0.0001</td>
</tr>
<tr>
<td>N-NH$_4^+$ – moisture</td>
<td>0.2610</td>
<td>0.0029</td>
</tr>
<tr>
<td>N-NO$_3^-$ – moisture</td>
<td>0.2604</td>
<td>0.0030</td>
</tr>
</tbody>
</table>

Fig. 7. Dependence of N-NH$_4^+$ on soil moisture

Fig. 8. Dependence of N-NO$_3^-$ on soil moisture
Table 4 shows Pearson’s correlation coefficient between that pairs of variables with P-value lower than 0.05. P-values below 0.05 indicate statistically significant non-zero correlations at the 95% confidence level. The values of Pearson’s coefficients for correlation between moisture and N-NH₄⁺ (0.2610) and moisture and N-NO₃⁻ (0.2604) show a week correlation even for the P-value lower than 0.01. The graphical interpretations are in Figs 7 and 8.

Conclusions

During the years 2008–2009 we monitored contents and dynamics of inorganic N forms in the Nature Reserve Alúvium Žitavy. From the acquired results, we can draw the following conclusions:

- The content of the ammonium nitrogen ranged between 3.80–16.87 mg kg⁻¹ with an average of 7.24 mg kg⁻¹. Higher concentrations were observed in the first year (2008).
- The most dominant inorganic form of nitrogen was ammonium, the proportion of the bulk inorganic nitrogen was 78.08%.
- The dynamics of the ammonium nitrogen during the year 2008 was decreasing; in the second year, it has been somewhat stabilised.
- The highest content of ammonium nitrogen was measured in sampling site 3 (7.36 mg kg⁻¹), the lowest in sampling site 1 (7.10 mg kg⁻¹).
- The dynamics of nitrate nitrogen during the two years was different from the dynamics of ammonium nitrogen. The higher concentration (2.30 mg kg⁻¹) was found in the second year.
- The highest content of nitrate nitrogen was measured in sampling site 2 (2.31 mg kg⁻¹), the lowest in sampling site 4 (1.62 mg kg⁻¹).

The important information is that the nitrate nitrogen concentrations maintain low (especially in depth from 0.3 to 0.6 m) and pose no risk of pollution to either surface water or groundwater.

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References


Regina, K., Nykanen, H., Silvola, J., Martikainen, P.J. 1996. Fluxes of nitrous oxide from boreal peatlands
as affected by peatland type, water table level and nitrification capacity. *Biogeochem.*, 35: 401–418.


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**Dynamika anorganických foriem dusíka v pôde Prírodnej rezervácie Aluvium Žitavy**

**Súhrn**


Po porovnaní obsahov N\(_{\text{am}}\) počas celého výskumného obdobia sme zistili, že vyššie hodnoty boli v prvom roku (2008). Amónny dusík sa pohyboval v hodnotách 3,80–16,87 mg kg\(^{-1}\) a jeho priemer v sledovanom období bol 7,24 mg kg\(^{-1}\), pričom v roku 2008 bol jeho obsah vyšší ako v roku 2009. Dusičnanový dusík mal opačnú tendenciu. Hodnoty dusičnanového dusíku boli v rozsahu 0,44–9,28 mg kg\(^{-1}\) s priemerom 2,03 mg kg\(^{-1}\) s vyššími hodnotami v roku 2009.

Štatisticky vysoko preukazný vplyv na obsah amónneho dusíka mali rok a dátum odberu. Na obsah dusičnanového dusíka štatisticky vysoko preukazne vplyvala hĺbka, rok a miesto odberu.

Vzhľadom na vysoké variačné koeeficienty neboli zistené žiadne korelácie medzi vlhkosťou pôdy a obsahmi sledovaných foriem anorganického dusíka.

Nízke hodnoty dusičnanového dusíka neznamenajú riziko znečistenia podzemných ani povrchových vôd.