

INSPIRATION bulletin

CL:AIRE's INSPIRATION bulletins describe practical aspects of research which have direct application to the management of contaminated soil or groundwater in an agricultural context. This bulletin describes an isotopic approach to determine the proportional contribution of nitrate sources in an Austrian river catchment.

Copyright © CL:AIRE.

The proportional contribution of nitrate sources in surface water in a mesoscale river catchment with a land-use gradient

1. Introduction

The European Union Water Framework Directive calls for water quality management on a river basins scale. Furthermore, it emphasises the need to prevent nitrate pollution of ground and surface waters as excess nitrogen from agricultural sources is one of the main causes of freshwater pollution in Europe. However, catchment-scale management approaches in large catchments are often hindered due to the poor understanding of the spatially and seasonally variable pathways of nitrate and its multiple possible sources. To overcome this problem the isotopic approach has been used to investigate the sources and fate of nitrate because different nitrate sources have distinctive isotopic nitrogen and oxygen signatures. Moreover, previous studies have proven that the probability distribution of the proportional contributions of nitrate sources can be estimated by the usage of Bayesian stable isotope mixing model. However, the number of surface water studies applying this model in large catchments characterised by a land use gradient is limited, and valuable information can be obtained by applying the model in areas characterised by different land uses to capture the differences in the proportional contributions of nitrate sources. Therefore, the Erlauf catchment was investigated as a mesoscale (631.5 km²) river system characterised by a land use gradient from pristine forests in headwaters to the agriculturally dominated area in the lower course of the river. The objectives were to: (1) determine the riverine nitrate (NO₃⁻) sources and their isotopic compositions in the study area, and (2) estimate and compare the proportional contribution of identified NO₃⁻ sources in sub-catchments that differ in their land use, by the usage of Bayesian stable isotope mixing model (MixSIAR).

2. Materials and Methods

2.1 Study site

The Erlauf catchment is situated in the Alpine foothills in Austria. The spring is located at 1200 m above sea level in the vicinity of the border between Lower Austria and Styria. The River Erlauf is a tributary of the Danube. The catchment is characterised by a land cover gradient. The southern part of the catchment is mainly forested in contrast to the northern part dominated by agriculture. Urbanisation is low, mainly rural with small villages and a few towns not exceeding 10000 habitants. The total catchment area is

631.5 km². At the gauging station located near the outlet, the mean discharge is 14.6 m³s⁻¹ (NO₃, 2019).

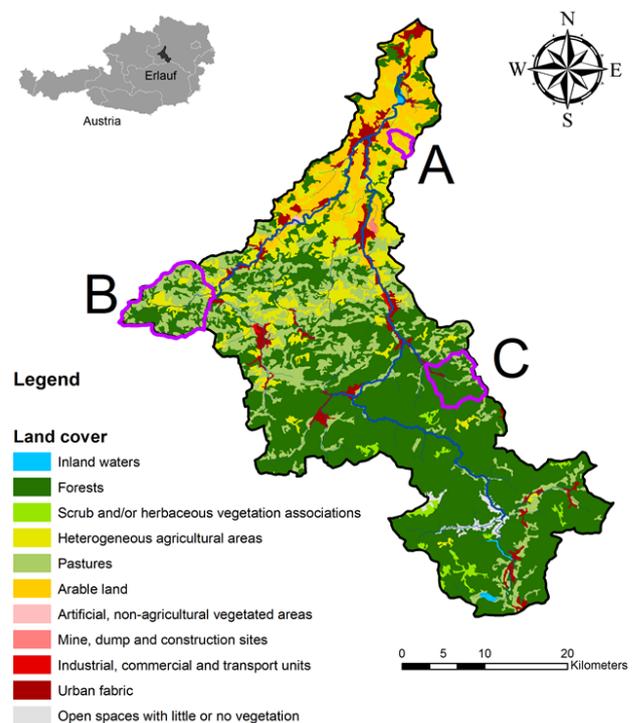


Figure 1: Location of the Erlauf catchment in Austria (left upper corner) and land cover map of the Erlauf catchment (EPA, 2012) with selected sub-catchments marked with purple (A, B, and C).

Three sampling locations were selected to estimate and compare the proportional contribution of identified NO₃⁻ sources in sub-catchments with different land-use. The first sampling location is situated at the outlet of stream Mühlbach (Fig. 1, A) and was selected to represent an agricultural catchment, with most of the catchment area covered by arable land. The second sampling location is located at the outlet of stream Schliefaubach (Fig. 1, B), and was selected to represent a mixed land-use catchment. The main land-use type in this catchment is pasture, although forests and heterogeneous agricultural areas are also present. The third sampling location is situated at the outlet of stream Jeßnitz (Fig. 1, C) and it was selected

INSPIRATION bulletin

to represent a forested catchment. The catchment is mainly forested with pastures and urban areas. Table 1 presents details regarding the land use types in all sub-catchments.

Table 1: Land cover in selected sub-catchments expressed as a percentage of the total sub-catchment area.

Sub-catchment	A	B	C
Land cover [%]			
Arable land	60	0	0
Heterogeneous agricultural areas	40	13	0
Pastures	0	47	16
Forests	0	39	82
Urban fabric	0	1	2

2.2 Sample collection and laboratory analysis

In November 2013 surface water samples were collected at the outlets of the selected sub-catchments. Moreover, 61 precipitation samples were collected in the study area including rainfall and snow samples over a longer period of time (2012 – 2017). All samples were collected in HDPE bottles, filtered (0.22 μm cellulose acetate) and stored in a fridge prior to analysis within one month of collection. The isotopic composition of dissolved NO_3^- ($\delta^{15}\text{N}_{\text{NO}_3}$ and $\delta^{18}\text{O}_{\text{NO}_3}$) was measured by mass spectrometry. The NO_3^- stable isotopes were measured using the denitrifier method with bacteria strains of *Pseudomonas chlororaphis* (ATCC #13985) (Casciotti *et al.*, 2002; Sigman *et al.*, 2001). Moreover, all samples were analysed for stable isotopes of water ($\delta^2\text{H}_{\text{H}_2\text{O}}$ and $\delta^{18}\text{O}_{\text{H}_2\text{O}}$) by laser cavity ring-down spectroscopy (L1102-I, Picarro Inc.). The NO_3^- concentrations were analysed in the laboratory with a Segmented Flow Analysis according to DIN EN ISO 13395.

2.3 Estimation of the proportional contribution of nitrate sources in the study area with SIAR model

Based on a land-use four potential NO_3^- sources in the Erlauf catchment were determined, i.e. atmospheric deposition (AD), nitrate fertilisers (NF), manure and sewage (M&S), and nitrification of reduced N sources (RNS). The RNS source consists of NO_3^- derived from nitrification of nitrogen originating in soil organic matter, ammonium fertilisers, and ammonium in precipitation. In order to estimate the proportional contribution of the four potential NO_3^- sources in surface water the typical nitrate isotopic signatures of those sources have to be known, i.e., mean $\delta^{15}\text{N}_{\text{NO}_3}$ and $\delta^{18}\text{O}_{\text{NO}_3}$ values as well as their standard deviations.

The $\delta^{15}\text{N}_{\text{NO}_3}$ and $\delta^{18}\text{O}_{\text{NO}_3}$ means and standard deviations of atmospheric deposition were based on values measured from precipitation samples collected in the study area.

The $\delta^{18}\text{O}_{\text{NO}_3}$ values of RNS and M&S originate from nitrification and were not measured directly, but calculated. In theory, NO_3^- formed by microbial nitrification incorporates two oxygen atoms from soil water and one from atmospheric O_2 (23.5‰) (Andersson and Hooper, 1983; Snider *et al.*, 2010). Consequently, the expected $\delta^{18}\text{O}$ value of NO_3^- derived from nitrification may be estimated as follows:

$$\delta^{18}\text{O}_{\text{NO}_3} = \frac{2}{3}\delta^{18}\text{O}_{\text{H}_2\text{O}} + \frac{1}{3}\delta^{18}\text{O}_{\text{O}_2} \quad \text{Equation 1}$$

The soil water $\delta^{18}\text{O}_{\text{H}_2\text{O}}$ required in equation 1 was calculated by adding 5‰ to the mean $\delta^{18}\text{O}_{\text{H}_2\text{O}}$ measured from precipitation samples collected in the study area to account for the possibility of higher soil water $\delta^{18}\text{O}_{\text{H}_2\text{O}}$ due to evaporation (Mayer *et al.*, 2001; Spoelstra *et al.*, 2007), or a heavier reservoir of soil O_2 due to respiratory consumption in soil (Kroopnick and Craig, 1972).

The relevant literature was used to obtain all other values, including $\delta^{15}\text{N}_{\text{NO}_3}$ and $\delta^{18}\text{O}_{\text{NO}_3}$ (Bateman and Kelly, 2007; Kendall, 1998 and references therein; Vitòria *et al.*, 2004), $\delta^{15}\text{N}_{\text{NO}_3}$ of RNS (Bateman and Kelly, 2007; Kendall, 1998 and references therein; Li *et al.*, 2007; Mayer *et al.*, 2001; Panno *et al.*, 2008; Rogers, 2008; Spoelstra *et al.*, 2007; Williard *et al.*, 2001; Zhang *et al.*, 2008), and $\delta^{15}\text{N}_{\text{NO}_3}$ of M&S (Bateman and Kelly, 2007; Curt *et al.*, 2004; Kendall, 1998 and references therein; Kreitler and Browning, 1983; Panno *et al.*, 2008; Rapisarda *et al.*, 2010; Rogers, 2008; Widory *et al.*, 2005).

The proportional contribution of four NO_3^- sources to surface water was calculated by applying a Bayesian stable isotope mixing model (MixSIAR). The model defines the set of N mixing measurements on NO_3^- isotopes with K sources and can be expressed as follows (Parnell *et al.*, 2010):

$$X_{ij} = \sum_{k=1}^k P_k(S_{jk} + C_{jk}) + \varepsilon_{jk} \quad \text{Equation 2}$$

$$S_{jk} \sim N(\mu_{jk}, \omega_{jk}^2)$$

$$C_{jk} \sim N(\lambda_{jk}, \tau_{jk}^2)$$

$$\varepsilon_{ij} \sim N(0, \sigma_j^2)$$

where X_{ij} is the observed isotope value j of the water sample i , P_k is the proportional contribution of source k , estimated by the model; S_{jk} is the source value k on isotope j and is normally distributed with mean μ_{jk} and standard deviation ω_{jk} ; C_{jk} is the isotope fractionation factor for isotope j on source k and it is normally distributed with mean λ_{jk} and standard deviation τ_{jk} ; ε_{ij} is the residual error describing additional inter-observation variance not described by the model and is normally distributed with mean 0 and standard deviation σ_j .

Based on water chemistry and isotope data denitrification was not observed in the respective sub-catchments (Bujak *et al.*, 2019). Moreover, calculations were performed for cold season (November) to reduce the possible impact of microbiological processes. Therefore, denitrification was assumed to have no effect on isotope signature in the respective sub-catchments and the fractionation factor was set to zero in equation 1 ($C_{kj}=0$).

3. Results and Discussion

Figure 2 shows the output of the model where source contributions differ between the different land-use areas.

The contribution of M&S calculated by the model in the agricultural sub-catchment was higher than in the mixed land-use and forested sub-catchments (74%, 40%, and 26%, respectively). Almost one-third of riverine NO_3^- was derived from manure and sewage in the forested catchment. This may seem a high result but considering that the measured NO_3^- concentration for the forested catchment was more than two times lower (5.0 mg L^{-1}) than in agricultural

INSPIRATION bulletin

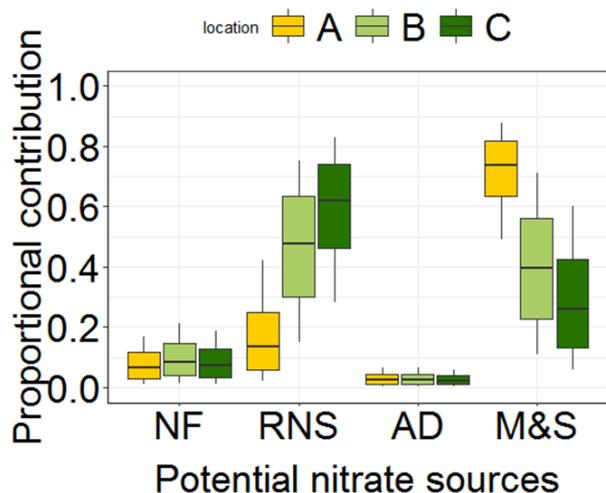


Figure 2: Proportional contributions of four potential NO_3^- sources in November 2013 estimated by the MixSIAR model for three sub-catchments (A, B, C) characterised by different land cover. Box plots illustrate the 10th, 25th, 50th, 75th, and 90th percentiles from bottom to top. Potential NO_3^- sources: nitrate fertilisers (NF), reduced N sources (RNS), atmospheric deposition (AD), manure and sewage (M&S).

catchment (12.1 mg L^{-1}) this cannot be neglected. The contribution of RNS calculated by the model in the forested catchment was higher than in the mixed land-use catchment and the agricultural catchment (62%, 48%, and 14%, respectively). Because the $\delta^{18}\text{O}_{\text{NO}_3}$ values of RNS and M&S were not measured directly but from the value expected from nitrification, the distinction of these two sources caused more constraints for the model and thus longer whiskers in the box plot (Fig. 2). According to the model, in all sub-catchments, AD contributed little to the riverine NO_3^- and less than 3%. The contribution of NF in all sub-catchments was low, from 6% to 8%.

4. Conclusion

Four potential NO_3^- sources in riverine water were determined in the Erlauf catchment and the proportional contribution of those sources was estimated using the MixSIAR model. Proportional nitrate source contributions in Erlauf catchment vary between catchment areas with different land uses. In the Erlauf catchment nitrification of reduced N sources was the main source of riverine NO_3^- in forested headwaters with a low percentage of the agricultural land cover. The proportional contribution of this nitrate source was substantially lower in the agricultural sub-catchment. Manure and sewage was the main source of nitrate in the agricultural sub-catchment. The land-use practices control the changes in nitrate sources contributions and nitrate concentrations in surface water in the study area. The results highlight the critical role of land management practices on regional N dynamics.

The MixSIAR model can be applied to estimate proportional contributions of four NO_3^- sources, namely manure and sewage, reduced N sources, nitrate fertiliser, and atmospheric deposition, in catchments characterised by different land-uses. The uncertainty of model estimations is higher for manure and sewage, and reduced N sources than for nitrate fertiliser and atmospheric deposition. An additional tracer (e.g. ^{11}Br) can be used to improve model estimates.

Acknowledgments

This research has received funding from the European Union's Horizon 2020 research and innovation programme under the Marie Skłodowska-Curie grant agreement No 675120 for the project entitled "Managing soil and groundwater impacts from agriculture for sustainable intensification – INSPIRATION".

References

- Andersson, K.K., Hooper, A.B. 1983. O_2 and H_2O are each the source of one O in NO_2^- produced from NH_3 by *Nitrosomonas*: ^{15}N -NMR evidence. *FEBS letters* 164 (2), 236-240.
- Bateman, A.S., Kelly, S.D. 2007. Fertilizer nitrogen isotope signatures. *Isotopes in environmental and health studies*. 43 (3), 237-247.
- Casciotti, K.L., Sigman, D.M., Hastings, M.G., Böhlke, J.K., Hilkert, A. 2002. Measurement of the oxygen isotopic composition of nitrate in seawater and freshwater using the denitrifier method. *Anal. Chem.* 74 (19), 4905-4912.
- Curt, M.D., Aguado, P., Sanchez, G., Bigeriego, M., Fernández, J. 2004. Nitrogen isotope ratios of synthetic and organic sources of nitrate water contamination in Spain. *Water, air, and soil pollution*. 151 (1-4), 135-142.
- Kendall, C. 1998. Tracing nitrogen sources and cycling in catchments, in: *Isotope tracers in catchment hydrology*. Elsevier, pp. 519-576.
- Kroopnick, P., Craig, H. 1972. Atmospheric oxygen: Isotopic composition and solubility fractionation. *Science*. 175 (4017), 54-55.
- Li, X., Masuda, H., Koba, K., Zeng, H. 2007. Nitrogen isotope study on nitrate-contaminated groundwater in the Sichuan Basin, China. *Water, air, and soil pollution*. 178 (1-4), 145-156.
- Mayer, B., Bollwerk, S.M., Mansfeldt, T., Hütter, B., Veizer, J. 2001. The oxygen isotope composition of nitrate generated by nitrification in acid forest floors. *Geochimica et Cosmochimica Acta*. 65 (16), 2743-2756.
- NÖ. 2019. Wasserstandsnachrichten und Hochwasserprognosen. NÖ Landesregierung (accessed 13.02.2019). <https://www.noel.gv.at/wasserstand/#/de/Messstellen>.
- Panno, S.V., Kelly, W.R., Hackley, K.C., Hwang, H.-H., Martinsek, A.T. 2008. Sources and fate of nitrate in the Illinois River Basin, Illinois. *Journal of Hydrology*. 359 (1-2), 174-188.
- Parnell, A.C., Inger, R., Bearhop, S., Jackson, A.L. 2010. Source partitioning using stable isotopes: Coping with too much variation. *PLOS ONE*. 5 (3), e9672.
- Rapisarda, P., Camin, F., Fabroni, S., Perini, M., Torrisi, B., Intrigliolo, F. 2010. Influence of different organic fertilizers on quality parameters and the $\delta^{15}\text{N}$, $\delta^{13}\text{C}$, $\delta^{2}\text{H}$, $\delta^{34}\text{S}$, and $\delta^{18}\text{O}$ values of orange fruit (*Citrus sinensis* L. Osbeck). *Journal of agricultural and food chemistry*. 58 (6), 3502-3506.
- Rogers, K.M. 2008. Nitrogen isotopes as a screening tool to determine the growing regimen of some organic and nonorganic supermarket produce from New Zealand. *Journal of agricultural and food chemistry*. 56 (11), 4078-4083.
- Sigman, D.M., Casciotti, K.L., Andreani, M., Barford, C., Galanter, M., Böhlke, J.K. 2001. A bacterial method for the nitrogen isotopic analysis of nitrate in seawater and freshwater. *Anal. Chem.* 73 (17), 4145-4153.
- Snider, D.M., Spoelstra, J., Schiff, S.L., Venkiteswaran, J.J. 2010. Stable oxygen isotope ratios of nitrate produced from nitrification: (^{18}O)-labeled water incubations of agricultural and temperate forest soils. *Environmental Science & Technology*. 44 (14), 5358-5364.
- Spoelstra, J., Schiff, S.L., Hazlett, P.W., Jeffries, D.S., Semkin, R.G. 2007. The isotopic composition of nitrate produced from nitrification in a hardwood forest floor. *Geochimica et Cosmochimica Acta*. 71 (15), 3757-3771.
- Widory, D., Petelet-Giraud, E., Négrel, P., Ladouche, B. 2005. Tracking the sources of nitrate in groundwater using coupled nitrogen and boron isotopes: a synthesis. *Environmental Science & Technology*. 39 (2), 539-548.
- Williard, K.W.J., DeWalle, D.R., Edwards, P.J., Sharpe, W.E. 2001. ^{18}O isotopic separation of stream nitrate sources in mid-Appalachian forested watersheds. *Journal of Hydrology*. 252 (1-4), 174-188.
- Zhang, Y., Liu, X.J., Fangmeier, A., Goulding, K.T.W., Zhang, F.S. 2008. Nitrogen inputs and isotopes in precipitation in the North China Plain. *Atmospheric Environment*. 42 (7), 1436-1448.

For more information on the INSPIRATION Project, please visit:
www.inspirationitn.co.uk

If you would like further information about other CL:AIRE publications
please contact us at the Help Desk at www.claire.co.uk