Quantification of environmental DNA Transport over a River Network

L.M. Stancanelli
Department of Hydraulic Engineering, Delft University of Technology, Delft, The Netherlands

E. Ragno
Department of Hydraulic Engineering, Delft University of Technology, Delft, The Netherlands

ABSTRACT: An innovative technique based on the use of environmental DNA was proposed by Ficetola et al. (2008) for performing biodiversity assessment. Applications of this technique in rivers are still scarce, due to the limited knowledge on the dynamics of eDNA traces. Here, using the open source dataset by Carraro et al. 2020, we investigate the transport of eDNA. We first investigate the ability of eDNA to capture species abundance observed by classical methods via correlation measures. Finally, we setup a numerical simulation for evaluating the transport induced by advection and diffusion and the eDNA decay coefficient.

1 INTRODUCTION

Freshwater biodiversity is declining at large and unprecedented rate. For bending the curve of freshwater biodiversity loss, it is crucial to enhance the biodiversity monitoring strategy. The majority of the current biomonitoring practices are invasive and taxa-specific and, as such, fail to describe the entire ecosystem. Examples are the kicknet for insects and electrofishing or gill netting for fishes. The recent advent of environmental DNA opened a new way for performing fast and non-invasive biodiversity assessments.

Environmental DNA (eDNA) is a pool of genomic material originating from living organisms and their remains present in different types of environmental samples (Pawlowski et al. 2020). These genetic traces of animals and plants include reproductive stages such as gametes, tissue fragments, epithelial cells, or excretions produced or expelled by the organisms during their life cycle (Barnes and Turner 2016). We can estimate the genus relative of the genetic material by taking a sample (water), opportunely filtering it and extracting the eDNA with molecular lab-analysis. In particular, it is possible to perform: single-species detection and biodiversity survey. Single-spiecies detection is used for the monitoring of rare/endangered species and of invasive species (Pawlowski et al. 2020). The amount of DNA can be quantified using quantitative Polymerase Chain Reactions (qPCR). The biodiversity survey is performed adopting the eDNA metabarcoding, a method that provides data about community composition (Ruppert et al., 2019). Nonetheless, the quantitative interpretation of the eDNA concentration to derive species' abundance is still challenging (Carraro et al., 2020); owing to the large number of variables involved, e.g., the amount of eDNA release depending on the type of species, the transport of eDNA depending on water flow conditions, the eDNA deterioration depending on temperature. To develop accurate abundance distribution maps is important to understand the mechanisms that govern eDNA spreading in river networks.

Here, we aim at evaluating our ability to quantify the abundance of species from eDNA concentration data, studying the displacement of eDNA traces in the river network. We use open-source biomonitoring datasets (Mächler et al., 2019; Carraro et al., 2020) providing info on genera detections at different sections (60 in total), with either classical strategies (kicknet) and eDNA sampling. We focus on understanding of eDNA concentration distribution dynamics in running water ecosystems via statistical measures and physical based modeling.

The article is organized as follows: after this introduction, there is a brief overview to previous studies on eDNA (section 2). Then, it is presented the dataset that we adopt (section 3) for studying: the correlation between abundance species estimates by classical biomonitoring approach and eDNA approach (section 4); and the eDNA decay coefficient in running water ecosystems when adopting a numerical modeling approach (section 5). Finally, we present remarks and future prospective of this study (section 6).

2 ENVIRONMENTAL DNA

In 2008 Ficetola and co-authors (Ficetola et al. 2008) were the first to detect the presence of a species in fresh water, based on the persistence of DNA in the environment. They examined whether DNA fragments are preserved in the aquatic environment and whether they can be used for assessing the presence of species. Their research was successful in demonstrating the potential of this new and innovative approach, opening new perspectives for the assessment of biodiversity in aquatic ecosystems.

Since then, the number of eDNA applications for ecology and conservation research has increased at an exponential rate (Fediajevaite et al.,2021). Research efforts were concentrated on optimizing the molecular lab analysis focusing for example on primers, DNA amplification and sequencing, and taxa database. The rapid advancement in the molecular biology research, i.e., with the development of the metabarcoding approach capable to provide a rapid biodiversity assessment, facilitates a number of biomonitoring applications. Consequently, the interest on eDNA spreads easily into other research fields, such as ecology, hydraulics, hydrology, and water management. The research aim was oriented to reveal the origin and the fate of eDNA in the water ecosystem. Harrison et al., (2019) presented a review discussing the major processes that eDNA undergoes between organism and collection: eDNA shedding, decay, and transport.

The eDNA shedding varies as a function of the type of species targeted. Indeed, the quantitative analysis of the genetic material dispersed in the ecosystem is the starting point for species abundance estimate. It is then important to assess the persistence of the genetic material in the ecosystem. The eDNA decay is the function that describes the time evolution of eDNA concentration. Studies on decay function demonstrate a logarithm decay in water and sand in relation to abiotic conditions (temperature, pH, salinity). Longer persistence of eDNA is observed in sand samples rather than in water samples (Sakata et al.2020). Finally, the transport of eDNA traces in the ecosystem allows to construct abundance distribution map. The understanding of eDNA transport dynamics is crucial for two aspects to improve: i) the estimate on the location of species communities; and ii) the probability to detect eDNA in field. The eDNA transport differs massively in relation to the type of ecosystems (Harrison et al., 2019). In lentic ecosystems, such as pond and lakes, we observe mainly a vertical transport and a stratification of eDNA; whereas in lotic ecosystems, such as streams and river, the horizontal transport has a major role.

3 DATASET OF ENVIROMENTAL DNA BY CARRARO ET AL. 2020

The dataset here used, it is the one by Carraro et al.,2020 (https://doi.org/10.5281/zenodo.3903330). This dataset is related to the biomonitoring assessment carried out in a subcatchment of the Thur basin, Switzeland. The basin has an extension of 740 km^2, along which they have selected 60 sampling staions, for investigating the biodiversity of insects, belonging to the following taxonomic orders: Ephemeroptera, Plecoptera, and Trichoptera. Sampling sites were chosen in order to represent all stream orders (Strahler number) in the river network (for more details, see Carraro et al. 2020, Supplementary Fig. 1). Streams belongings to order I are the one located upstream. They are characterized by slope of 0.2 and flow depth lower that 0.5 m and width of 1-3 m. Streams located in sections belongings to the lower part of the basin (order 3) have larger channel width, with values up to 20 m, and flow depth lower than 1.2 m. This dataset provides info on 50 genera detections at different sections (60 in total), with either classical strategies and eDNA strategy.

The classical strategy was consisting in the kicknet sampling in field and taxa recognition in the lab. They have immersed a kicknet in the water for 3 minutes, catching so insects that were then classified using a stereomicroscope.
The eDNA strategy was consisting in the collection of water samples that were then analyzed in the molecular biology lab for quantifying the DNA concentration.
Here, we used this dataset with the scope to investigate the dynamics of eDNA in the river network. We focus on the data relative to a single genus Leuctra, belongings to the taxonomic order Plecoptera.

4 ESTIMATE OF TAXA ABUNDACE IN A SECTION: COMPARISON BETWEEN CLASSICAL METHOD AND ENVIRONMENTAL DNA

In order to evaluate statistically the dynamic of the eDNA traces and how it evolves along the river network, we have compared data on taxa abundance acquired by means of kicknet and eDNA. In particular, for each river section we evaluate the rank correlation between the number of individuals detected with the kicknet and the number of eDNA reads detected in the water sample. Values of correlation may inform how the DNA traces distribute along the river network, and if their dynamics change at different stream orders. For example, low correlation between eDNA/kichnet data relative to sections located in the upper portion of the basin could indicate a rapid displacement of the traces downstream from its point of release. Whereas a poor correlation relative to sections located in the lower portion of the basin could be induced by the transfer and accumulation of traces along the river channel.

We use data of concentration estimate performed on the Leutra genus. From the kicknet sampling we have data measure in terms of number of individual detected while from the eDNA sampling we have the number of reads detected in the water sample. In order to facilitate the comparison between the two sampling methods which are characterized by different unit measure, we have separately ranked the two data vectors (Fig. 1a). After we calculate the Spearman's rank correlation between kicknet and eDNA abundance evaluated for the stream order (Fig. 1b). Value of correlation depends on the stream order, i.e. Strahler number (Fig 1b). Correlation increase when we consider exclusively the stations located in the upper part of the basin (Strahler order I). Such a results is underlining the fact that in case of the DNA methods the component of the transport plays a major role in the lower part of the basin.

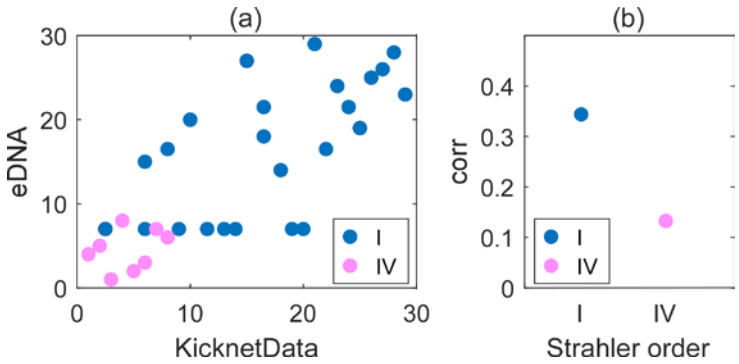

Figure 1. Concentration estimate of the Leuctra genus in the Thur river, dataset from Carraro et al. 2021. (a) relation between the ranked data vectors of the concentration of Leuctra, respectively estimated with traditional method, kicknet, and innovative method, eDNA. (b) the Spearman's rank correlation between kicknet and eDNA abundance for each stream order, Strahler number.

5 NUMERICAL SIMULATION OF THE ENVIROMENTAL DNA SPREADING IN RIVER NETWORK

In order to evaluate the dynamics of eDNA traces in running water adopting a physical based approach, we have performed numerical simulations. We adopted Delft 3d for evaluate the hydrodynamics forces characteristics of the Thur basin and to evaluate the eDNA concentration spreading in the river network. We have imposed a rough schematization of the river channel geometry, assuming a rectangular channel. We have performed a 1D simulation of the hydrodynamic model and then we have evaluated the eDNA concentration spreading adopting the Delft 3d "water quality model". The goal of the numerical simulation is to evaluate of the eDNA transport by advection and diffusion. Successively we wish to calibrate the numerical model, using monitoring data and tuning the eDNA degradation coefficient. We are currently working on the evaluation of the characteristic eDNA degradation coefficient to be adopted at the different stream order. Results of the numerical simulations will be presented during the conference.

6 CONCLUSIONS

The advent of the eDNA analysis as a biodiversity assessment approach has opened a new paradigm for the biota conservation. Until now eDNA was mainly adopted for evaluated the presence/absence of species, and only few studies investigated the species abundance. Major concern is the perception of possible false positives detection caused by displacement of genetic material traces downstream from its point of release.

In this research, we provide a preliminary investigation from a statistical and a physical perspective on the rate of eDNA transport as a function of the different stream orders. We adopt an open-source dataset (Carraro et al. 2020), providing estimate of species abundance by kicknet sampling and eDNA sampling. Statistical analysis carried out on this dataset restitute higher correlation value between kicknet sampling and eDNA sampling in the upper portion of the basin. The lower portion is characterized by lower value, probably due to the transport/accumulation of genetic material in the network. For better understanding the transport we have also set-up a 1D numerical simulation of the eDNA concentration distribution in the river network, with the purpose to evaluate the eDNA degradation coefficient.

Future studies should focus attention on the development of a eDNA decay function in relation to hydrodynamics forces. This will lead to an accurate estimate of the fate of eDNA traces along the network.

REFERENCES

Barnes, M. A., & Turner, C. R. (2016). The ecology of environmental DNA and implications for conservation genetics. Conservation genetics, 17(1), 1-17.

Carraro, L., Mächler, E., Wüthrich, R., & Altermatt, F. (2020). Environmental DNA allows upscaling spatial patterns of biodiversity in freshwater ecosystems. Nature communications, 11(1), 1-12.

Carraro et al., (2020). DATASET: eDITH-thur (v1.0). Zenodo. https://doi.org/10.5281/zenodo.3903330

Fediajevaite, J., Priestley, V., Arnold, R., & Savolainen, V. (2021). Meta-analysis shows that environmental DNA outperforms traditional surveys, but warrants better reporting standards. Ecology and Evolution, 11(9), 4803-4815.

Ficetola, G. F., Miaud, C., Pompanon, F., & Taberlet, P. (2008). Species detection using environmental DNA from water samples. Biology letters, 4(4), 423-425.

Harrison, J. B., Sunday, J. M., & Rogers, S. M. (2019). Predicting the fate of eDNA in the environment and implications for studying biodiversity. Proceedings of the Royal Society B, 286(1915), 20191409.

Pawlowski J., Apothéloz-Perret-Gentil L., Mächler E. & Altermatt F. 2020: Environmental DNA applications in biomonitoring and bio-assessment of aquatic ecosystems. Guidelines. Federal Office for the Environment, Bern. Environmental Studies. no. 2010: 71 pp.

Ruppert, K. M., Kline, R. J., & Rahman, M. S. (2019). Past, present, and future perspectives of environmental DNA (eDNA) metabarcoding: A systematic review in methods, monitoring, and applications of global eDNA. *Global Ecology and Conservation*, *17*, e00547.

Sakata, M. K., Yamamoto, S., Gotoh, R. O., Miya, M., Yamanaka, H., & Minamoto, T. (2020). Sedimentary eDNA provides different information on timescale and fish species composition compared with aqueous eDNA. Environmental DNA, 2(4), 505-518.