

# Tracking aquatic animals with sediment DNA records

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## Introduction

Animals play key roles in aquatic ecosystems, both as connectors between primary producers and consumers, as well as mobile vectors, moving nutrients and other elements between habitats. Furthermore, many aquatic animals are associated with highly-valued ecosystem services including commercial and recreational fisheries, bird watching and other naturalist activities. Over the past few decades, we have seen numerous reports in the media of different wildlife populations declining (e.g., freshwater migratory fish in Canada<sup>i</sup>) or recovering from an earlier decline (e.g., cormorants in Great Lakes-St. Lawrence region<sup>ii</sup>). These assessments are possible in some cases where long-term records of population dynamics exist, but often such data are restricted to only a small proportion of aquatic animals and many observations are discontinuous over time. Long-term data can, however, be developed from lake sediment archives, providing key insights into unknown or unmeasured population and community level changes in recent decades. Furthermore, as sediment archives can date back over centuries, it is

possible to examine baseline conditions in aquatic ecosystems that predate major anthropogenic change.

Traditional paleolimnological methods have largely focused on organisms or parts of organisms that are microscopic in size (e.g., pollen, diatoms or algal pigments; Gregory-Eaves and Smol 2023). There has also been a long-standing effort to look at zooplankton subfossils represented both as pieces of the carapace or as resting stages in sediment cores. Classical paleolimnological methods also include the study of chironomid subfossil head capsules, which may be quite abundant in profundal sediments and have been examined over a large geographic area (Engels et al. 2020). In contrast, subfossils of fish and other animals (e.g., molluscs and waterbirds) are generally very sparse in sediment cores and thus have not been the subject of extensive study (but see Davidson et al. 2003; Walton et al. 2021).

Analyses of sediment DNA (sedDNA) have the potential to define how many animals have fluctuated over time, which was not possible with traditional paleolimnological approaches when no morphological remains were found. Earlier genetic analyses in paleolimnology began with the identification and genotyping of propagules (e.g., gametes, spores, resting eggs) left by organisms in their environment as a way to reconstruct past population dynamics and infer relationships between aquatic organisms (Orsini et al. 2013). Now, sedDNA approaches have largely been developed through modifications of environmental DNA (eDNA) and ancient DNA studies, which are more developed fields (Taberlet et al. 2018; Orlando et al. 2021). Furthermore, metabarcoding and metagenomic approaches can produce data streams of numerous taxa in a

single analysis, thereby providing a broader perspective of organisms that may have interacted in a food web.

In this chapter, we have approached the topic of aquatic animals by developing two main themes that are based on analyses of DNA preserved in lake sediments: a) zooplankton population and community dynamics and b) aquatic vertebrate dynamics. We end the chapter with a summary of the key findings and provide a perspective on topics that might be addressed in the near future.

#### **A. Zooplankton population and community dynamics**

Zooplankton occupy a central position in the aquatic food web as many are herbivores of phytoplankton (primary producers) and act as prey to planktivorous fish (note: some zooplankton also take advantage of the microbial loop or are predators of other zooplankton). Consequently, zooplankton communities are impacted by both top-down and bottom-up processes, and have been found to change across gradients in trophic status, temperature, and other lake variables (Paquette et al. 2022). Zooplankton offer a unique opportunity for understanding past ecological and evolutionary dynamics because several species can produce dormant forms (resting eggs; Cuenca-Cambronero et al. 2022), which can survive harsh conditions and remain preserved for long periods of time in aquatic sediments (Radzikowski 2013). A substantial diversity in zooplankton resting stages can be found in lake sediments; from cladocerans that generally preserve well (Vandekerhove et al. 2004), to species of rotifers and copepods that are less-well

preserved. Specifically, rotifer resting eggs tend to preserve poorly in sedimentary archives (Wallace and Snell 2010; Epp et al. 2010), offering limited possibilities for identification via morphological characters (Briski et al. 2011) and restricted success in reconstructing past rotifer population dynamics. In contrast, *Daphnia* produce resting eggs encapsulated in hard protective cases called ephippia. These heavy cases often settle at the bottom of lakes where they accumulate in sediments, acting as a reservoir for zooplankton populations once the environment becomes favorable again. Genetic analyses of resting eggs are typically done on individuals hatched in the lab from eggs retrieved from sediment cores. However, this approach may introduce biases in population estimates due to variable hatching success among genotypes and because conditions for successful hatching are unknown for most resting eggs (Schwartz & Hebert 1987). To overcome such issues, investigators may extract DNA from the eggs directly, without hatching them first (Montero-Pau, Gómez & Muñoz 2008; Lack et al. 2018; Cordellier et al. 2021). Methods have also been developed for extracting DNA directly from the ephippial cases (Ishida et al. 2012), meaning that *Daphnia* species can be determined even in the absence of eggs or from a cracked or incomplete ephippia, but success rates tend to be low. Finally, like other groups of organisms, zooplankton also leave pieces of DNA behind in the environment which can be analyzed by tapping directly into bulk sediments (sedDNA) even in the absence of visible fossilized remains or propagules. In a recent study, Tsugeki et al. (2022) demonstrated a strong congruence between quantitative (q)PCR analysis of sedDNA and morphological time-series data of resting eggs from *D. galeata* and *D. pulicaria* populations over the last 100 yrs. in Lake Biwa (Japan).

### ***1. External stressors as drivers of zooplankton population or community dynamics***

DNA-based paleolimnological evaluations of resting eggs have been broadly used to evaluate the temporal changes in the genetic structure of *Daphnia* populations in response to various external stressors such as eutrophication (e.g., Weider et al. 1997; Brede et al. 2009; Frisch et al. 2014; Möst et al. 2015). A frequent outcome of eutrophication and climate warming is the proliferation and accumulation of cyanobacteria in lakes. Because cyanobacteria are generally a poor food source for zooplankton compared to green algae, their proliferation can be a strong selection pressure on *Daphnia* populations. Experimental studies and the hatching of resting eggs in the laboratory has shown (Hairston et al. 1999) that small-bodied *Daphnia* and *Bosmina* are better adapted to the presence of cyanobacteria (even toxic taxa), and may initially outcompete larger-bodied *Daphnia* at the onset of toxic algal blooms (Jiang et al. 2017).

Apart from eutrophication and climate warming, other forms of anthropogenic stressors in aquatic systems have been shown to impact *Daphnia* population dynamics using sedDNA, such as the loading of aquatic contaminants. A study by Turko et al. (2016) based on genotyping of hatched resting eggs suggested rapid adaptation to lead pollution in the invasive *Daphnia galeata* of the peri-Alpine Lake Greifensee (Switzerland). *D. galeata* acquired resistance to lead pollution and became dominant, but populations showed a loss of resistance to lead after concentrations decreased. Likewise, sequencing of the 12S rRNA gene of DNA from resting eggs revealed that a past invasion of the Eurasian species *D. curvirostris* in Lake Onondaga (New York), a temperate dimictic lake, was at least partially facilitated by industrial contamination arising from the local chemical industry (Duffy et al., 2000). The invader largely disappeared once the lake water quality was restored.

## ***II. Interactions (e.g., host-parasite)***

Another topic of interest which can be explored using zooplankton is host-parasite interactions, which is an important structuring force in natural populations (Decaestecker et al. 2007). Using genotyping of hatched resting eggs from sediment cores collected in a natural pond (Heverlee, Belgium), Decaestecker et al. (2007) experimentally reconstructed the coevolutionary dynamics of *Daphnia* and its bacterial endoparasite *Pasteuria ramosa* providing evidence of *Daphnia* – *Pasteuria* coevolution. More recently, Rajarajan et al. (2022) investigated the role of natural genetic variability of the host *D. galeata* in determining the phylogenetic structure of the associated microbial community with a 5-yr long common garden experiment. The team then conducted a 16S rRNA gene metabarcoding analysis of microbial communities associated with *D. galeata* hatched from eggs recovered at two time periods in the sediments of Lake Greifensee (Switzerland). Their results showed that bacterial communities in the gut and body of *Daphnia* differed significantly between *Daphnia* genotypes, but that bacterial community beta-diversity was not correlated with the genetic distance between the hosts, suggesting that factors other than environmental selection and host genetics (including stochasticity) were at play in shaping *Daphnia* microbiome over time. Genetic analyses of sediment records are also unlocking information on the composition, prevalence and effect of viral infections on zooplankton.

As mentioned above, genetic reconstructions based on the genotyping or sequencing of resting eggs are limited for taxa other than *Daphnia*. However, a few studies have successfully conducted genetic analyses of on non-Daphnid resting eggs to investigate past population dynamics. For instance, Derry et al. (2010) looked at the survival of *Leptodiatomus minutus* copepods by hatching resting eggs from sediment intervals representing different levels of

acidification over a period of up to 100 years. Likewise, Epp et al. (2010) reconstructed the history of the rotifer genus *Brachionus* from its resting stages archived in a sediment record from an alkaline-saline crater lake (Lake Sonachi, Kenya) to elucidate the persistence and temporal dynamics of *Brachionus* haplotypes over a period of past environmental change. Analyses of the mitochondrial cytochrome oxidase I (COI) gene showed that the dominant haplotype in Lake Sonachi changed concomitantly to major environmental perturbations (volcanic eruption and major lake water level decrease).

## **B. Aquatic vertebrate population and community dynamics**

There is substantial interest in understanding the long-term dynamics of numerous aquatic vertebrates, in part because these taxa tend to be more iconic, represent important commercial species and, in some cases, may be taxa at risk of extinction. In addition to “desirable” taxa, there are also taxa that are not native to a particular site and thus are monitored as part of programs designed to limit their growth and spread. While studying the distribution and dynamics of both native and non-native taxa is necessary, it is also expensive and time consuming. As such, substantial investments have been made in developing efficiencies in the study of eDNA and sedDNA for issues of conservation and management (Taberlet et al. 2018; Ficetola et al. 2019; Pawlowski et al. 2022). Analyses of sedDNA targeting aquatic vertebrates are still in their early days, but this is an area that we anticipate will grow in the future (Capo et al. 2021). New methods specifically designed to detect sedDNA of aquatic vertebrates are also coming online (Thomson-Laing et al. 2022; Sakata et al. pre-print).

### ***I. Targeted approaches***

## **a) Fishes**

Typically, when one thinks of aquatic vertebrates, fish is the group of organisms that comes to mind and indeed they are a critical part of many aquatic food webs. Relative to other vertebrate groups, the most diverse group are the fish, with approximately 12,000 freshwater species (based on a global assessment in 2000). However, this is also the group that has seen more extinctions in the 20<sup>th</sup> century than any other vertebrates (Burkhead 2012). As a result, there is a growing need to understand the distribution and dynamics of a wide diversity of fishes.

To date, there has been a substantial number of papers that have applied eDNA approaches to study fish abundances, distributions and dynamics (Yates et al. 2021; Littlefair et al. 2021; Miya 2022). This foundational work from the water column has not only advanced the field of eDNA research but has also served the paleolimnological community well in terms of developing primers, improving lab workflows with information on inhibitors and streamlining bioinformatic pipelines. Experimental work has also been conducted in fish ponds or experimental vessels to quantify the persistence of fish DNA particles in water and sediment over time. For example, Sakata et al. (2020) collected samples of water and sediment from a fish pond and found that the concentration of DNA particles were generally greater and more persistent in sediments than in water, but that species composition based on metabarcoding was not different between sample matrices. These results are encouraging for sedDNA as they show that sediments have the potential to archive a longer term and representative portrait of fish assemblages.



Only a handful of studies have been published thus far with the express intent of developing historical trajectories of fish dynamics. In all cases, these studies have taken a targeted approach where a genetic marker specific for a particular fish was evaluated (see Table 1). The earliest study conducted by Matisoo-Smith et al. (2008) confirmed the presence of a native fish in numerous samples from a lake sediment core in New Zealand. In later studies by Stager et al. (2015) and Nelson-Chorney et al. (2019), the authors were interested in resolving whether a focal fish was present in the lake ecosystem historically, or whether it was a relatively new addition to the food web. In two more recent studies, the authors have had access to complementary historical data on fish landings and in some cases, fish scale records, which they used to evaluate the coherency among records (Kuwae et al. 2020; Sakata et al. pre-print). For many of their focal fish time series, sedDNA concentrations were positively correlated with fish landings. In the more recent paper (Sakata et al. pre-print), the team corrected their sedDNA fish time series for diagenesis, using a model developed for chlorophyll *a* pigments and its derivatives (based on a first order degradation equation). In all cases, Sakata et al. (pre-print) found that the correlation between the sedDNA concentrations and the paired fish landing time series improved once the diagenesis correction was applied.

Overall, the rather limited number of published fish sedDNA studies may partly be due to challenges associated with studying fish DNA from sediment archives. Although comparative studies between water and sediments have reported higher concentrations of fish DNA in sediments (Sakata et al. 2020; Turner et al. 2015), there is mixed evidence for the successful detection of fish DNA in sediment archives. Some studies have pointed to limitations related to the diffusion of fish DNA from sediments to water (Dunker et al. 2016) and the decomposition of

sedDNA (Wei et al. 2018), as well as differential efficiencies in sedDNA extraction methods (Thomson-Laing et al. 2022) especially in sediments with high organic content. Monitoring fish is essential for many reasons (economical as well as ecological) and paleoreconstructions based on sedDNA have the potential to inform on fish dynamics that can contribute to better management. The sedDNA community is currently working towards optimizing protocols for fish DNA detection and extraction from sediment archives (e.g., Thomson-Laing et al. 2022 and other efforts underway)

#### **b) Other vertebrates and their parasites**

Targeted work on other aquatic vertebrates is likely to come in the near future, by focusing on birds and amphibians of interest. Already, there has been a decent amount of paleolimnological work tracking bird dynamics indirectly by looking at tracers of guano (reviewed Duda et al. 2021). In such cases, large colonies of birds produce a substantial amount of guano that is enriched in nutrients as well as numerous isotopes, heavy metals and pesticides, and these materials are transported to neighboring lakes. It is highly likely that the DNA from birds would also be preserved in sediment archives and could be evaluated through a targeted PCR approach, and possibly through more generalized sequencing methods (i.e., metabarcoding or metagenomics). We found a few cases where bird DNA was extracted from materials found in cave deposits (Willerslev et al. 2003; Haile et al. 2007), which is not too dissimilar from lake sediments. For example, Willerslev et al. (2003) successfully sequenced the DNA of a few birds from New Zealand in a deposit that was at least 10,000 years old.

Although we did not identify any published targeted studies on birds or amphibians extracted from lake sediments, we did find a few papers on parasites commonly associated with these organisms. For example, avian influenza virus (AIV) has been detected across numerous sediment samples. Based on a recent review by Coombe et al. (2021), AIV detection rates can vary between 0 – 56% in sediment- and soil-based substrates, and one study reported that probability of detection of AIV increased in sites with anthropogenic disturbance (Himsworth et al. 2020). Lang et al. (2008) reported on the potential to detect AIV in sediment archives, but to date we have not seen one such study published. Similarly, several papers have focused on amphibian pathogens, especially *Batrachochytrium* taxa which have been associated with global population declines of amphibians. Earlier work showed that *Batrachochytrium* DNA was persistent in soil for months (Stegen et al. 2017). In a comparative field study of DNA from frog skin swabs with environmental DNA from water and sediment samples (collected in the ponds the frogs inhabited), Brannelly et al. (2020) found that sediment samples generally had lower *Batrachochytrium* DNA concentrations than water samples, and limited seasonality relative to the other sample matrices. A logical next step would be to extract a core from a site where long-term data exist on *Batrachochytrium* infections to evaluate how congruent the sediment archive is with the epidemiological data.

## ***II. Application of Metabarcoding and Shotgun Sequencing***

Through both metabarcoding and shotgun sequencing (i.e., metagenomics) of environmental samples it is possible to detect a broader range of organisms, which has the advantage of considering multiple trophic levels. The primer sets used in metabarcoding are typically designed

to detect particular groups, with COI primers often used for invertebrate assemblages and 12S primers used for fish assemblages (although 16S and 18S primers can also pick up invertebrates and animals in general). To date, there have been only a few sedDNA metabarcoding studies that focused on aquatic animals, and none yet on fish (see Table 2).

With shotgun sequencing, one aims to study all DNA in an environmental sample, and this can be considered advantageous as it removes the potential for PCR biases to be introduced. From a sedDNA extract, shotgun sequencing can provide an ecosystem-wide perspective (e.g., from bacteria and archaea to fish and mammals; Table 2). However, without any manipulation, the metazoan signal contained within the output of reads from a single environmental sample is rather small (i.e., less than 1% of assigned reads; Monchamp et al. 2022). Nonetheless, comparisons of classical zooplankton taxonomy analyses with metagenomic data based on eDNA extracted from water samples at the same sites, yielded results that were moderately congruent (Monchamp et al. 2022). Furthermore, shotgun sequencing can be applied to study *Daphnia* resting eggs where this analytical platform can provide insight into the differentiation of populations among time points, and identify presence of functional genes and viral pathogens (Hewson et al. 2013; Cordellier et al. 2021; O’Grady et al. 2022). For example, Hewson et al. (2013) used a shotgun metagenomic approach on resting eggs and bulk sedDNA from two temperate lakes in the USA to identify viruses suspected to be involved in the ecology of *Daphnia* spp. They were able to detect viruses in resting eggs retrieved from ~30 yr-old sediments of Oneida Lake and found a high prevalence of potentially fatal viruses (DMC1aHV+) prior to important population declines in *D. retrocurva*.

Several paleo-metagenomic studies have been conducted and recorded the presence of numerous aquatic animal taxa based on a handful of reads per sample (Table 2). For example, Pedersen et al. (2016) detected the presence of pike fish (*Esox* sp.) in a lake sediment record from western Canada (Charlie Lake) dating back 11,700 calendar years by adopting a metagenomic analysis. However, the greatest number of reads detected for pike was only 68, whereas the number of reads for some plant taxa were in the thousands. Other fish taxa detected in the record included stickleback and perch. To enhance the signal of taxa that typically have low read counts, one might adopt a hybridization approach, whereby one exposes the metagenomic library to RNA probes that hybridize to target sedDNA molecules and then these can be isolated from other DNA through the selection of probes by bacteria (streptavidin) that can be linked to a magnet (Ambrecht et al. 2021). Applying a comparative approach, Ambrecht et al. (2021) demonstrated an enhanced eukaryotic signal when hybridization was applied relative to extracts from marine sediments that did not receive this treatment. We anticipate that this approach will be applied more in the future.

## **Conclusions and future directions**

For most aquatic ecosystems, there are no long-term monitoring data on population or community dynamics, and thus lake sediment archives can fill key gaps in our understanding. The study of aquatic animals based on genetic material archived in lake sediments is largely rooted in population-based analyses of zooplankton resting stages. The field is now greatly expanding with the appearance of numerous exciting developments (e.g., shotgun sequencing + RNA probes), which is growing analyses to include many other taxa and the simultaneous study of

multiple trophic levels (see Chapter 10: Considering organisms in an interactive web with sediment DNA records). With such analyses, time series data that capture a wider trophic web can be developed and interrogated with sophisticated network analyses and joint-species distribution models.

Currently, there are several large initiatives underway that are developing more complete reference libraries for barcodes and full genomes (e.g., BIOSCAN: <https://ibol.org/programs/bioscan/> and the Earth Biogenome project: <https://www.earthbiogenome.org/>). These datasets will greatly improve taxonomic and functional gene assignments, and allow investigators to tease substantially more information from their sedDNA analyses. Collaborative efforts, facilitated through organizations like the SedaDNA Scientific Society (<https://sedadna.github.io/>) will also be instrumental in advancing the field and enhancing communication of results.

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## **References**

Armbrecht L, Hallegraeff G, Bolch CJS, et al. (2021) Hybridisation capture allows DNA damage analysis of ancient marine eukaryotes. *Scientific Reports* 11(1):3220. 10.1038/s41598-021-82578-6

Brannnelly LA, Wetzel DP, Ohmer MEB, et al. (2020) Evaluating environmental DNA as a tool for detecting an amphibian pathogen using an optimized extraction method. *Oecologia* 194(1):267-281. 10.1007/s00442-020-04743-4

Brede N, Sandrock C, Straile D, et al. (2009) The impact of human-made ecological changes on the genetic architecture of *Daphnia* species. *Proceedings of the National Academy of Sciences* 106(12):4758-4763. doi:10.1073/pnas.0807187106

Briski E, Cristescu ME, Bailey SA, et al. (2011) Use of DNA barcoding to detect invertebrate invasive species from diapausing eggs. *Biological Invasions* 13(6):1325-1340. 10.1007/s10530-010-9892-7

Burkhead NM (2012) Extinction Rates in North American Freshwater Fishes, 1900–2010. *BioScience* 62(9):798-808. 10.1525/bio.2012.62.9.5

Capo E, Gigu et-Covex C, Rouillard A, et al. (2021) Lake Sedimentary DNA Research on Past Terrestrial and Aquatic Biodiversity: Overview and Recommendations. *Quaternary* 4(1):6.

Coolen MJL, Orsi WD, Balkema C, et al. (2013) Evolution of the plankton paleome in the Black Sea from the Deglacial to Anthropocene. *Proceedings of the National Academy of Sciences* 110(21):8609-8614. doi:10.1073/pnas.1219283110

Coombe M, Iwasawa S, Byers KA, et al. (2021) A systemic review and narrative synthesis of the use of environmental samples for the surveillance of avian influenza viruses in wild waterbirds. *Journal of Wildlife Diseases* 57(1):1-18, 18. <https://doi.org/10.7589/JWD-D-20-00082>

Cordellier M, Wojewodzic MW, Wessels M, et al. (2021) Next-generation sequencing of DNA from resting eggs: signatures of eutrophication in a lake’s sediment. *Zoology* 145:125895. <https://doi.org/10.1016/j.zool.2021.125895>

Courtin J, Perfumo A, Andreev AA, et al. (2022) Pleistocene glacial and interglacial ecosystems inferred from ancient DNA analyses of permafrost sediments from Batagay megaslump, East Siberia. *Environmental DNA* 00:1-19. <https://doi.org/10.1002/edn3.336>

Cuenca-Cambronero M, Courtney-Mustaphi CJ, Greenway R, et al. (2022) An integrative paleolimnological approach for studying evolutionary processes. *Trends in Ecology & Evolution* 37(6):488-496. 10.1016/j.tree.2022.01.007

Davidson TA, Sayer CD, Perrow MR, et al. (2003) Representation of fish communities by scale sub-fossils in shallow lakes: implications for inferring percid–cyprinid shifts. *Journal of Paleolimnology* 30(4):441-449. 10.1023/B:JOPL.0000007247.97059.e2

- Decaestecker E, Gaba S, Raeymaekers JAM, et al. (2007) Host–parasite ‘Red Queen’ dynamics archived in pond sediment. *Nature* 450(7171):870-873. 10.1038/nature06291
- Derry AM, Arnott SE, Boag PT (2010) Evolutionary shifts in copepod acid tolerance in an acid-recovering lake indicated by resurrected resting eggs. *Evolutionary Ecology* 24 (1): 133–45. <https://doi.org/10.1007/s10682-009-9295-3>.
- Dommain R, Andama M, McDonough MM, et al. (2020) The challenges of reconstructing tropical biodiversity with sedimentary ancient DNA: A 2200-year-long metagenomic record from Bwindi Impenetrable Forest, Uganda. *Frontiers in Ecology and Evolution* 8 10.3389/fevo.2020.00218
- Duda MP, Hargan KE, Michelutti N, et al. (2021) Reconstructing Long-Term Changes in Avian Populations Using Lake Sediments: Opening a Window Onto the Past. *Frontiers in Ecology and Evolution* 9 10.3389/fevo.2021.698175
- Duffy MA, Perry LJ, Kearns CM, et al. (2000) Paleogenetic evidence for a past invasion of Onondaga Lake, New York, by exotic *Daphnia curvirostris* using mtDNA from dormant eggs. *Limnology and Oceanography* 45(6):1409-1414. <https://doi.org/10.4319/lo.2000.45.6.1409>
- Dunker KJ, Sepulveda AJ, Massengill RL, et al. (2016) Potential of Environmental DNA to Evaluate Northern Pike (*Esox lucius*) Eradication Efforts: An Experimental Test and Case Study. *PLOS ONE* 11(9):e0162277. 10.1371/journal.pone.0162277
- Echeverría-Galindo P, Anslan S, Frenzel P, et al. (2021) High-throughput identification of non-marine Ostracoda from the Tibetan Plateau: Evaluating the success of various primers on sedimentary DNA samples. *Environmental DNA* 3(5):982-996. <https://doi.org/10.1002/edn3.222>
- Engels S, Medeiros AS, Axford Y, et al. (2020) Temperature change as a driver of spatial patterns and long-term trends in chironomid (Insecta: Diptera) diversity. *Global Change Biology* 26(3):1155-1169. <https://doi.org/10.1111/gcb.14862>
- Epp LS, Stoof KR, Trauth MH, et al. (2010) Historical genetics on a sediment core from a Kenyan lake: intraspecific genotype turnover in a tropical rotifer is related to past environmental changes. *Journal of Paleolimnology* 43(4):939-954. 10.1007/s10933-009-9379-7
- Ficetola GF, Manenti R, Taberlet P. (2019) Environmental DNA and metabarcoding for the study of amphibians and reptiles: species distribution, the microbiome, and much more. *Amphibia-Reptilia* 40(2):129-148. <https://doi.org/10.1163/15685381-20191194>
- Frisch D, Morton PK, Chowdhury PR, et al. (2014) A millennial-scale chronicle of evolutionary responses to cultural eutrophication in *Daphnia*. *Ecology Letters* 17(3):360-368. <https://doi.org/10.1111/ele.12237>



Garcés-Pastor S, Wangenstein OS, Pérez-Haase A, et al (2019) DNA metabarcoding reveals modern and past eukaryotic communities in a high-mountain peat bog system. *Journal of Paleolimnology* 62(4):425-441. 10.1007/s10933-019-00097-x

Gauthier J, Walsh D, Selbie DT, et al (2021) Evaluating the congruence between DNA-based and morphological taxonomic approaches in water and sediment trap samples: Analyses of a 36-month time series from a temperate monomictic lake. *Limnology and Oceanography* 66(8):3020-3039. <https://doi.org/10.1002/lno.11856>

Gregory-Eaves I, Smol JP (2023) *Paleolimnology: Methods and Applications*. In: Jones I, Smol JP, editors. *Wetzel's Limnology: Lake and River Ecosystems* (4th Edition).

Haile J, Holdaway R, Oliver K, et al. (2007) Ancient DNA Chronology within Sediment Deposits: Are Paleobiological Reconstructions Possible and Is DNA Leaching a Factor? *Molecular Biology and Evolution* 24(4):982-989. 10.1093/molbev/msm016

Hairston NG, Lampert W, Cáceres CE, et al. (1999) Rapid evolution revealed by dormant eggs. *Nature* 401(6752):446-446. 10.1038/46731

Hewson I, Ng G, Li W, et al. (2013) Metagenomic identification, seasonal dynamics, and potential transmission mechanisms of a *Daphnia*-associated single-stranded DNA virus in two temperate lakes. *Limnology and Oceanography* 58(5):1605-1620. <https://doi.org/10.4319/lo.2013.58.5.1605>

Himsworth CG, Duan J, Prystajek N, et al. (2019) Targeted resequencing of wetland sediment as a tool for avian influenza virus surveillance. *Journal of Wildlife Diseases* 56(2):397-408. 10.7589/2019-05-135

Ishida S, Ohtsuki H, Awano T, et al. (2012) DNA extraction and amplification methods for ephippial cases of *Daphnia* resting eggs in lake sediments: a novel approach for reconstructing zooplankton population structure from the past. *Limnology* 13(3):261-267. 10.1007/s10201-012-0380-x

Jiang X, Xie J, Xu Y, et al. (2017) Increasing dominance of small zooplankton with toxic cyanobacteria. *Freshwater Biology* 62(2):429-443. <https://doi.org/10.1111/fwb.12877>

Kuwae M, Tamai H, Doi H, et al. (2020) Sedimentary DNA tracks decadal-centennial changes in fish abundance. *Communications Biology* 3(1):558. 10.1038/s42003-020-01282-9

Lack JB, Weider LJ, Jeyasingh PD. (2018) Whole genome amplification and sequencing of a *Daphnia* resting egg. *Molecular Ecology Resources* 18(1):118-127. <https://doi.org/10.1111/1755-0998.12720>

Lang AS, Kelly A, Runstadler JA (2008) Prevalence and diversity of avian influenza viruses in environmental reservoirs. *Journal of General Virology* 89: 509–519. DOI 10.1099/vir.0.83369-0

Littlefair JE, Hrenchuk LE, Blanchfield PJ, et al. (2021) Thermal stratification and fish thermal preference explain vertical eDNA distributions in lakes. *Molecular Ecology* 30(13):3083-3096. <https://doi.org/10.1111/mec.15623>

Matisoo-Smith E, Roberts K, Welikala N, et al. (2008) Recovery of DNA and pollen from New Zealand lake sediments. *Quaternary International* 184(1):139-149. <https://doi.org/10.1016/j.quaint.2007.09.013>

Miya M (2022) Environmental DNA Metabarcoding: A Novel Method for Biodiversity Monitoring of Marine Fish Communities. *Annual Review of Marine Science* 14(1):161-185. [10.1146/annurev-marine-041421-082251](https://doi.org/10.1146/annurev-marine-041421-082251)

Monchamp M-È, Walsh DA, Garner RE, et al. (2022) Comparative analysis of zooplankton diversity in freshwaters: What can we gain from metagenomic analysis? *Environmental DNA* <https://doi.org/10.1002/edn3.335>

Montero-Pau J, Gómez A, Muñoz J (2008) Application of an inexpensive and high-throughput genomic DNA extraction method for the molecular ecology of zooplanktonic diapausing eggs. *Limnology and Oceanography: Methods* 6(6):218-222. <https://doi.org/10.4319/lom.2008.6.218>

Möst M, Oexle S, Marková S, et al. (2015) Population genetic dynamics of an invasion reconstructed from the sediment egg bank. *Molecular Ecology* 24(16):4074-4093. <https://doi.org/10.1111/mec.13298>

Nelson-Chorney HT, Davis CS, Poesch MS, et al. (2019) Environmental DNA in lake sediment reveals biogeography of native genetic diversity. *Frontiers in Ecology and the Environment* 17(6):313-318. <https://doi.org/10.1002/fee.2073>

O'Grady CJ, Dhandapani V, Colbourne JK, et al. (2022) Refining the evolutionary time machine: An assessment of whole genome amplification using single historical *Daphnia* eggs. *Molecular Ecology Resources* 22(3):946-961. <https://doi.org/10.1111/1755-0998.13524>

Olajos F, Bokma F, Bartels P, et al (2018) Estimating species colonization dates using DNA in lake sediment. *Methods in Ecology and Evolution* 9(3):535-543. <https://doi.org/10.1111/2041-210X.12890>

Orlando L, Allaby R, Skoglund P, et al. (2021) Ancient DNA analysis. *Nature Reviews Methods Primers* 1(1):14. [10.1038/s43586-020-00011-0](https://doi.org/10.1038/s43586-020-00011-0)

Orsini L, Schwenk K, De Meester L, et al. (2013) The evolutionary time machine: using dormant propagules to forecast how populations can adapt to changing environments. *Trends in Ecology & Evolution* 28(5):274-282. <https://doi.org/10.1016/j.tree.2013.01.009>.

Paquette C, Gregory-Eaves I, Beisner BE (2022) Environmental drivers of taxonomic and functional variation in zooplankton diversity and composition in freshwater lakes across Canadian continental watersheds. *Limnology and Oceanography* 67(5):1081-1097. <https://doi.org/10.1002/lno.12058>

Pawlowski J, Bruce K, Panksep K, et al. (2022) Environmental DNA metabarcoding for benthic monitoring: A review of sediment sampling and DNA extraction methods. *Science of The Total Environment* 818:151783. <https://doi.org/10.1016/j.scitotenv.2021.151783>

Pedersen MW, Ruter A, Schweger C, et al. (2016) Postglacial viability and colonization in North America's ice-free corridor. *Nature* 537(7618):45-49. 10.1038/nature19085

Radzikowski J (2013) Resistance of dormant stages of planktonic invertebrates to adverse environmental conditions. *Journal of Plankton Research* 35(4):707-723. 10.1093/plankt/fbt032

Rajarajan A, Wolinska J, Walser J-C, Dennis SR, Spaak P (2022) Host-associated bacterial communities vary between *Daphnia galeata* genotypes but not by host genetic distance. *Microbial Ecology*, no. 0123456789. <https://doi.org/10.1007/s00248-022-02011-x>.

Sakata MK, Yamamoto S, Gotoh RO, et al. (2020) Sedimentary eDNA provides different information on timescale and fish species composition compared with aqueous eDNA. *Environmental DNA* 2(4):505-518. <https://doi.org/10.1002/edn3.75>

Sakata MK, Tsugeki N, Kuwae M, et al. (2022). Fish environmental DNA in lake sediment overcomes the gap of reconstructing past fauna in lake ecosystems. *Biorxiv*. <https://doi.org/10.1101/2022.06.16.496507>

Schwartz SS, Hebert PDN (1987) Methods for the activation of the resting eggs of *Daphnia*. *Freshwater Biology* 17(2):373-379. <https://doi.org/10.1111/j.1365-2427.1987.tb01057.x>

Stager JC, Sporn LA, Johnson M, et al. (2015) Of paleo-genes and perch: What if an “alien” is actually a native? *PLOS ONE* 10(3): e0119071. 10.1371/journal.pone.0119071

Stegen G, Pasmans F, Schmidt B et al. (2017) Drivers of salamander extirpation mediated by *Batrachochytrium salamandrivorans*. *Nature* 544: 353–356. <https://doi.org/10.1038/nature22059>

Taberlet P, Bonin AI, Zinger L, et al. (2018) *Environmental DNA: for biodiversity research and monitoring*. Oxford: Oxford University Press. Available from: <https://search.ebscohost.com/login.aspx?direct=true&scope=site&db=nlebk&db=nlabk&AN=1701932>

Thomson-Laing G, Howarth JD, Vandergoes MJ, et al. (2022) Optimised protocol for the extraction of fish DNA from freshwater sediments. *Freshwater Biology* 67(9):1584-1603. <https://doi.org/10.1111/fwb.13962>

Tsugeki N, Nakane K, Doi H, et al. (2022) Reconstruction of 100-year dynamics in *Daphnia* spawning activity revealed by sedimentary DNA. *Scientific Reports* 12(1):1741. 10.1038/s41598-021-03899-0

Turko P, Sigg L, Hollender J, et al. (2016) Rapid evolutionary loss of metal resistance revealed by hatching decades-old eggs. *Evolution* 70(2):398-407. <https://doi.org/10.1111/evo.12859>

Turner CR, Uy KL, Everhart RC (2015) Fish environmental DNA is more concentrated in aquatic sediments than surface water. *Biological Conservation* 183:93-102. <https://doi.org/10.1016/j.biocon.2014.11.017>

Vandekerckhove J, Declerck S, Vanhove M, et al. (2004) Use of ephippial morphology to assess richness of anomopods: potentials and pitfalls. *Journal of Limnology* 63(s1):75-84. 10.4081/jlimnol.2004.s1.75

Wallace RL, Snell TW. Chapter 8: Rotifera. In: Thorp JH, Covich AP, editors. *Ecology and Classification of North American Freshwater Invertebrates* (Third Edition). San Diego: Academic Press; 2010. p. 173-235.

Walton RE, Sayer CD, Bennion H et al. (2021). Once a pond in time: employing palaeoecology to inform farmland pond restoration. *Restoration Ecology* 29: e13301. <https://doi.org/10.1111/rec.13301>

Wei N, Nakajima F, Tobino T. (2018) A microcosm study of surface sediment environmental DNA: Decay observation, abundance estimation, and fragment length comparison. *Environmental Science & Technology* 2018 52 (21), 12428-12435. DOI: 10.1021/acs.est.8b04956

Weider LJ, Lampert W, Wessels M, Colbourne JK, Limburg P (1997) Long-term genetic shifts in a Microcrustacean Egg Bank Associated with Anthropogenic Changes in the Lake Constance Ecosystem. *Proceedings of the Royal Society B: Biological Sciences* 264 (1388): 1613–18. <https://doi.org/10.1098/rspb.1997.0225>.

Willerslev E, Hansen AJ, Binladen J et al. (2003) Diverse plant and animal genetic records from Holocene and Pleistocene sediments. *Science*. 300 (5620): 791-795. DOI: 10.1126/science.1084114

Yates MC, Glaser DM, Post JP et al. (2021) The relationship between eDNA particle concentration and organism abundance in nature is strengthened by allometric scaling. *Molecular Ecology*. <https://doi.org/10.1111/mec.15543>

## Footnotes

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<sup>i</sup> <https://www.theglobeandmail.com/canada/article-canada-is-failing-its-freshwater-fish-populations/>

<sup>ii</sup> <https://www.cbc.ca/radio/ideas/championing-the-cormorant-why-experts-argue-this-water-bird-is-unfairly-vilified-1.6200538>

Table 1. Compilation of studies using a sedDNA approach targeted to track past fish population or assemblage dynamics.

Authors	Year	Target taxa	Brief description of lab approach	Main finding
Matisoo-Smith et al.	2008	Common bully ( <i>Gobiomorphus cotidianus</i> )	150 bp sequence of 12S	Detected the presence of a native fish species
Stager et al.	2015	Yellow perch (YP; <i>Perca flavescens</i> )	species-specific barcode marker for YP; 124 bp cytochrome oxidase subunit 1 (CO1) gene	Despite concern that YP might not be native, all samples from 2200-year core tested positive for YP (negative controls were clean)
Olajos et al.	2018	Whitefish ( <i>Coregonus lavaretus</i> L.)	Whitefish specific primers designed from mitochondrial sequences	Based on analyses of ~10,000 yr. sediment records from Sweden, results show colonization of whitefish right after the region's deglaciation in one site, but colonization is much delayed in the second site
Nelson-Chorney et al.	2019	Yellowstone cutthroat trout (YCTT; <i>Oncorhynchus clarkii bouvieri</i> ) & westslope cutthroat trout (WCTT; <i>Oncorhynchus clarkii lewisi</i> )	Two diagnostic loci with SNPs unique to focal taxa used	DNA results challenged previous notion of recent introduction of WCTT as it was present in sediment record for ~100 years
Kuwae et al.	2020	Japanese anchovy, <i>Engraulis japonicus</i> , Japanese sardine, <i>S. melanostictus</i> , and jack mackerel, <i>Trachurus japonicus</i>	qPCR	Detected significant correlations between DNA concentrations and fish landings for 3 taxa groups, and modest correlation with Sardine fish scale records
Sakata et al.	preprint	<i>Plecoglossus altivelis</i> and <i>Gymnogobius isaza</i>	mitochondrial cytochrome b (cytb) gene (< 150 bp)	Positive relationships between preservation-adjusted DNA concentrations and historical CPUE estimates for <i>Plecoglossus altivelis</i>

Note 1: CPUE = catch per unit effort and SNP = single nucleotide polymorphism

Table 2. Summary of metabarcoding and metagenomic studies addressing aquatic fauna distributions and dynamics in sediments

Authors	Year	Target taxa	Brief description of lab approach	Main findings
<b>a. Metabarcoding studies</b>				
Coolen et al.	2013	Copepods	18S rRNA (317 bp fragment) amplified with general eukaryote and copepod specific reverse primer (Cop317r)	Presence of particular copepod taxa suggest establishment of saline conditions in Black Sea
Garces-Pastor et al.	2019	Invertebrates and more broadly eukaryotes	313 bp fragment of the mitochondrial COI, using the Leray-XT primer set & V7 region of nuclear-encoded ribosomal 18S rRNA gene (< 117 bp)	Relatively more arthropoda detected with the COI vs 18S primer sets. Some of the most prevalent arthropods were mites, collembola, coleoptera, maxillopoda and ostracoda
Echeverria-Galindo et al.	2021	Ostracodes	16S, 18S and COI primer pairs tested <i>in silico</i> and on surface sediment samples	Greatest % of ostracod sequences obtained with 2 different 18S primers, but sediment volume extracted had little effect
Gauthier et al.	2021	Eukaryotes	v7 region of 18S rRNA (~260 bp) results compared with zooplankton morphological counts in sediment traps and water	Three maxillopoda ASVs dominated water and sediment samples, whereas morphological taxa were dominated by brachiopoda. Greatest congruence between zooplankton sedDNA and morphology was apparent by looking at extracellular sedDNA fraction.
Capo et al.	2021	Eukaryotes	Comparison of extraction approaches and sediment volumes using 18S rRNA v7 region	Lake-specific differences much greater than extraction differences. When zooming in on metazoan reads, no clear patterns were associated with sample volume. Slight tendency for chordate reads to be more abundant in extracellular DNA fraction.
<b>b. Metagenomic studies</b>				
Pedersen et al.	2016	All taxa	Metagenomics analyses of lake sediments from past Beringia sites of western Canada	Diversity of aquatic fauna detected as far back as 12,000 cal yr. BP
Dommain et al.	2020	All taxa	Metagenomic analyses of swamp core from Uganda dating back 2200 cal years BP	Substantial diversity in metazoa detected from including fishes, insects, nematodes as well as birds and mammals
Courtin et al.	2022	All taxa	Metagenomics analyses of permafrost sediments from Siberian dating back to mid Pleistocene	Culicidae of the mosquito family detected in all samples, among other organisms from viruses to mammals
Note 1: additional 18S metabarcoding studies have been published but we only highlight ones where animals were the focus.				
Note 2: additional 18S metabarcoding studies where the focus was on protists are featured in the chapter 6 by Barouillet et al.				
Note 3: bp = base pairs				

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This chapter is part of the not yet published book:

# **Tracking Environmental Change Using Lake Sediments**

## **Volume 6: Sedimentary DNA**

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**Chapter 1** “Using sedimentary DNA to unravel past changes in biological communities” [Eric Capo, Cecilia Barouillet, John P. Smol](#)

**Chapter 2** “The source and fate of lake sedimentary DNA” [Charline Giguet-Covex, Anthony Foucher, Ludovic Gielly, Femke Augustijns, Marina Morlock, Stanislav Jelavic, Isabelle Domaizon, Eric Capo](#)

**Chapter 3** “The workflow of the sedimentary DNA approach” [Peter D Heintzman, Kevin Nota, Benjamin Vernot, Linda Armbricht, Alexandra Rouillard, Tyler Murchie, Youri Lammers, Sandra Garces Pastor](#)

**Chapter 4** “Bacterial and archaeal DNA from lake sediments” [Aurèle Vuillemin, Marco Coolen, Susanne Liebner, Stefan Bertilsson, Pilar Junier, Jens Kallmeyer](#)

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**Chapter 6** “DNA archives of protists in lake sediments” [Cecilia Barouillet, Isabelle Domaizon, Eric Capo](#)

**Chapter 7** “Diatom DNA from lake sediments” [Katharina Dulias, Kathleen Stoof-Leichsenring, Laura Epp, Ulrike Herzsuh](#)

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**Chapter 10** “Considering organisms in an interactive web with sediment DNA records” [Zofia Taranu](#), [Irene Gregory Eaves](#), and [Marie-Eve Monchamp](#)

**Chapter 11** “Terrestrial vegetation DNA from lake sediments” [Sandra Garcés-Pastor](#), [Sarah Crump](#), [Laura Parducci](#), [Kevin Nota](#), [Dilli P. Rijal](#), [Sisi Liu](#), [Weiham Jia](#), [Maria Leunda](#), [Christoph Schwörer](#), [Inger G. Alsos](#)

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**Chapter 13** “Perspectives and future development in sedimentary DNA research” [Mikkel W Pedersen](#)