Received: 26 November 2020 | Revised: 14 September 2021 | Accepted: 11 October 2021

DOI: 10.1002/edn3.259

Environmental DNA
Open Access
Dedicated to the study and use of environmental DNA for basic and applied sciences

WILEY

ORIGINAL ARTICLE

# Preservation of sedimentary plant DNA is related to lake water chemistry

**Weihan Jia**[1,2] | **Xingqi Liu**[1] | **Kathleen R. Stoof-Leichsenring**[2] | **Sisi Liu**[2,3] | **Kai Li**[2,4] | **Ulrike Herzschuh**[2,3,5]

[1]College of Resource Environment and Tourism, Capital Normal University, Beijing, China

[2]Polar Terrestrial Environmental Systems Research Group, Alfred Wegener Institute Helmholtz Centre for Polar and Marine Research, Potsdam, Germany

[3]Institute of Environmental Science and Geography, University of Potsdam, Potsdam, Germany

[4]College of Chemistry and Life Sciences, Zhejiang Normal University, Jinhua, China

[5]Institute of Biochemistry and Biology, University of Potsdam, Potsdam, Germany

**Correspondence**
Ulrike Herzschuh, Polar Terrestrial Environmental Systems Research Group, Alfred Wegener Institute Helmholtz Centre for Polar and Marine Research, Potsdam, Germany.
Email: Ulrike.Herzschuh@awi.de

Xingqi Liu, College of Resource Environment and Tourism, Capital Normal University, Beijing, China.
Email: xqliu@cnu.edu.cn

**Funding information**
European Research Council, Grant/Award Number: 772852; National Natural Science Foundation of China and German Research Foundation, Grant/Award Number: 41861134030; The Second Tibetan Plateau Scientific Expedition and Research Program, Chinese Academy of Sciences, Grant/Award Number: 2019QZKK0202; Capital Normal University, Grant/Award Number: 028185511700

## Abstract

Little is currently known about preservation of plant DNA in lake sediments. Most prior information originates from laboratory experiments while systematic field-based studies are still lacking. Here, we used the "*g*" and "*h*" universal primers for the P6 loop region of the chloroplast *trn*L (UAA) intron to amplify plant DNA from 219 lake surface sediments from China and Siberia. We introduce (i) the percentage of sequence counts with the best identity ≥95%, (ii) weighted average identity, (iii) weighted average DNA fragment length, and iv) rarefied richness of terrestrial seed plants of plant DNA metabarcoding as proxies for sedimentary DNA preservation and relate them to five environmental variables (lake water conductivity, lake water pH, mean July air temperature, and sampling depth, lake size) using boosted regression tree (BRT) analyses. Our results suggest that lake water chemical characteristics, that is, electrical conductivity and pH, are the most important variables for the preservation of plant DNA in lake sediments. Intermediate water conductivities (100–500 μS $cm^{-1}$) and neutral to slightly alkaline water pH (7–9) may facilitate plant DNA preservation. Furthermore, deep lakes seem to support plant DNA preservation as indicated by relatively high rarefied richness. We also find high rarefied richness in small lakes compared with large lakes, but this result needs to be assessed by more studies in the future. None of our BRT models shows that mean July air temperature is a key variable to limit plant DNA preservation. To conclude, our results suggest that sedimentary DNA studies can preferentially select deep lakes characterized by intermediate water conductivities and neutral to slightly alkaline pH conditions.

**KEYWORDS**
boosted regression tree, China and Siberia, environmental DNA, lake sediments, metabarcoding, plant DNA preservation

## 1 | INTRODUCTION

Sedimentary ancient DNA is increasingly used to reconstruct past ecosystem changes (Brown & Barnes, 2015; Capo et al., 2021; Pedersen et al., 2016; Ruppert et al., 2019; Willerslev et al., 2014). Since the development of next-generation sequencing (Glenn, 2011; Metzker, 2010; Shendure & Ji, 2008), the P6 loop region of the chloroplast *trn*L (UAA) intron (Taberlet et al., 2007) has become one of the most commonly used markers for identification of vascular plants in lake sedimentary (ancient) DNA studies (e.g., Alsos et al., 2018; Boessenkool et al., 2014; Clarke et al., 2019; Crump et al., 2021; Ficetola et al., 2018; Giguet-Covex et al., 2014; Liu et al., 2021; Niemeyer et al., 2017; Pansu et al., 2015; Parducci et al., 2012; Rijal et al., 2021; Sjögren et al., 2017). Accordingly, there is an urgent need to fully understand the taphonomy of this proxy including origin, transfer, and preservation of sedimentary plant DNA (Birks & Birks, 2016; Edwards, 2020; Giguet-Covex et al., 2019; Gugerli et al., 2005; Parducci et al., 2017). It has been shown that the plant DNA pool in lake sediments mainly originates from plant tissues such as leaves, seeds, roots, and wood remains (Parducci, Nota, et al., 2019; Willerslev et al., 2003). Conditions that support or hinder the preservation of plant DNA in lake sediments are by far less studied (Birks & Birks, 2016; Giguet-Covex et al., 2019; Parducci et al., 2017). Understanding how the environmental setting of a lake influences plant DNA preservation is essential to improve sampling strategies and to allow lake sedimentary DNA to become a reliable and interpretable proxy for past vegetation changes.

Damage to the DNA molecules can be efficiently repaired via DNA repair pathways in living cells (Lindahl, 1993). In contrast, various chemical reactions including hydrolysis (e.g., depurination and deamination), oxidation, alkylation, and Maillard reaction can degrade DNA after cell death (Eglinton & Logan, 1991; Hofreiter et al., 2001; Lindahl, 1993; Willerslev & Cooper, 2005). These reactions affect both intra- and extracellular DNA and lead to destabilization and breakage of DNA molecules, limiting the retrieval of complete DNA sequences from environmental samples (Hofreiter et al., 2001; Lindahl, 1993; Willerslev & Cooper, 2005). For example, nucleotide substitutions (C to T or G to A) caused by hydrolytic deamination mostly occur at the ends of ancient DNA fragments, which has become a common way to distinguish endogenous DNA from contamination (Briggs et al., 2007; Jónsson et al., 2013; Willerslev & Cooper, 2005). As a result, environmental DNA molecules are typically highly fragmented and damaged, reducing their chances of being correctly assigned to the taxa of their origin, which can impede sequence identification and species richness estimation.

The rate of chemically induced degradation is impacted by biotic (e.g., microbial activity) and abiotic factors (e.g., pH, temperature, ultraviolet (UV) radiation, and ionic strength). For example, low temperature, high pH, and high ionic strength can limit the rate of DNA depurination, which has been experimentally demonstrated *in vitro* by Lindahl and Nyberg (1972). DNA molecules can also be damaged from high-level UV and oxygen exposure (Lindahl, 1993; Ravanat et al., 2001; Strickler et al., 2015), which ought to be considered, especially for large lakes with long-distance surface transport due to large catchment areas. Furthermore, the adsorption of DNA to soil and sediment particles is regarded as the primary mechanism responsible for the long-term preservation of extracellular DNA in the environment because such adsorption inhibits the activation of DNA-degrading DNases produced by bacteria and fungi (Blum et al., 1997; Khanna & Stotzky, 1992; Ogram et al., 1988; Pietramellara et al., 2009), and this process is influenced by several factors such as ionic strength, pH, and sediment composition (Cai et al., 2006; Freeman et al., 2020; Greaves & Wilson, 1969; Kanbar et al., 2020; Khanna & Stotzky, 1992; Levy-Booth et al., 2007; Lorenz & Wackernagel, 1987; Romanowski et al., 1991; Yu et al., 2013). Most of the studies targeting DNA preservation performed experiments in specific laboratories using pure DNA molecules and preprocessed samples. However, inferences from laboratory analyses partly contradict the scarce field-based evidence. For example, high temperature is considered to accelerate DNA degradation (Hofreiter et al., 2001; Lindahl, 1993; Lindahl & Nyberg, 1972; Willerslev & Cooper, 2005), but many studies have successfully extracted PCR-amplifiable DNA from tropical lake sediments (e.g., Boessenkool et al., 2014; Bremond et al., 2017; Stoof-Leichsenring et al., 2012; Tabares et al., 2020). Thus, there is an urgent need to assess these experimental results against field-based studies.

In principal, lake sediment is an optimal place for DNA preservation. Sediments are frequently saturated with water, which seems to prevent downward leaching of DNA (Anderson-Carpenter et al., 2011; Giguet-Covex et al., 2014; Sjögren et al., 2017). In addition, when sediment particles are deposited at the lake bottom, adsorbed DNA becomes stored in a low-temperature, low-oxygen, and low-level UV radiation environment which limits microbial activity and is favorable for DNA preservation (Giguet-Covex et al., 2019; Parducci et al., 2017). Before the final burial, however, the quality of DNA at the water–sediment interface can be affected by environmental conditions, which are of crucial importance for the long-term preservation of sedimentary DNA molecules (Capo et al., 2021). Accordingly, investigation into the DNA characteristics of lake surface sediments can provide general insights into degradation pathways of sedimentary (ancient) DNA. However, reliable measures or proxies for preservation/degradation of lake sedimentary DNA have not yet been established.

DNA amplification, an essential step in the environmental DNA metabarcoding approach, is dependent on polymerase chain reaction (PCR; Taberlet et al., 2012). There are some potential biases in the current PCR-based analyses. For example, humic substances (e.g., humic acids and fulvic acids) in soils and sediments are assumed to be a major inhibitor of amplification and are difficult to be removed by standard DNA purification protocols (Matheson et al., 2010; Watson & Blackwell, 2000). Furthermore, primer mismatch (Piñol et al., 2015), polymerase selection (Matheson et al., 2010; Nichols et al., 2018), thermal cycling parameters (Wu et al., 2010), and other set-ups can also affect the outcomes of a metabarcoding study (Alberdi et al., 2018; Dopheide et al., 2018; Mathieu et al., 2020), especially for species richness and relative abundance

estimation. Therefore, the standard and same PCR protocol should be suggested to apply when we need to compare metabarcoding-based results from one dataset.

In this study, we used the "*g*" and "*h*" universal primers for the P6 loop region of the chloroplast *trn*L (UAA) intron to amplify plant DNA in 219 lake surface sediments from China and Siberia. Lake sediments were collected along a broad and extensive gradient of lake water conductivity, lake water pH, mean July air temperature (JulyT), sampling depth, and lake size. These five environmental variables were correlated to four proxies we have introduced for sedimentary DNA preservation using boosted regression tree (BRT) models (Elith et al., 2008). This paper aims (i) to establish and evaluate proxies for sedimentary DNA preservation; (ii) to quantify the relationships between the preservation of sedimentary plant DNA and various environmental variables; and (iii) to guide sampling designs for lake sedimentary DNA studies.

## 2 | MATERIALS AND METHODS

### 2.1 | Study lakes

Details about the sampled localities, including geographic coordinates, physical–chemical and environmental parameters, dominant surrounding vegetation, and sampling methods, are given in Stoof-Leichsenring et al. (2020). In this study, we investigated a subset of 219 lakes with available physical–chemical parameters located in the Qinghai-Tibetan Plateau (QTP), arid northwestern China, and Siberia (Figure 1; Appendix S1). The lakes cover a large geographical, climatic, and ecological gradient with elevation ranging from sea level to about 5000 m above sea level, lake water conductivity ranging from 5 to 57,194 μS $cm^{-1}$ (mean 1272 μS $cm^{-1}$), lake water pH ranging from 4.7 to 10.24 (mean 7.94), sampling depth ranging from 0.15 to 78.9 m (mean 8.76 m), and lake size ranging from 0.01 to 4400 $km^2$ (mean 63.88 $km^2$). The area of 45 extremely small lakes (or ponds) was replaced by 0.01 $km^2$. Data on JulyT downloaded from the WorldClim2 dataset (Fick & Hijmans, 2017; http://www.worldclim.org), containing the average climate data for the years 1970–2000 at a spatial resolution of 30 s (ca. 1 $km^2$), range from 4°C to 25°C (mean 12.17°C; Appendices S1 and S2).

### 2.2 | Lake sedimentary DNA analyses

DNA extraction was carried out in the molecular genetic laboratories equipped for environmental DNA work at Alfred Wegener Institute Helmholtz Centre for Polar and Marine Research (AWI). Approximately, 5–10 g of wet sediment was taken from the samples and processed using the DNeasy PowerMax Soil Kit (Qiagen). One extraction blank was included for each batch of 10 samples and processed in the same way as the samples. PCR was performed with the "*g*" and "*h*" universal plant primers for the P6 loop region of the chloroplast *trn*L (UAA) intron (Taberlet et al., 2007). We produced two PCR replicates for most samples including extraction blanks and PCR negative template controls (NTCs). For the 13-TY samples from Taymyria, we also produced two PCR replicates but pooled them together because the same tag combinations

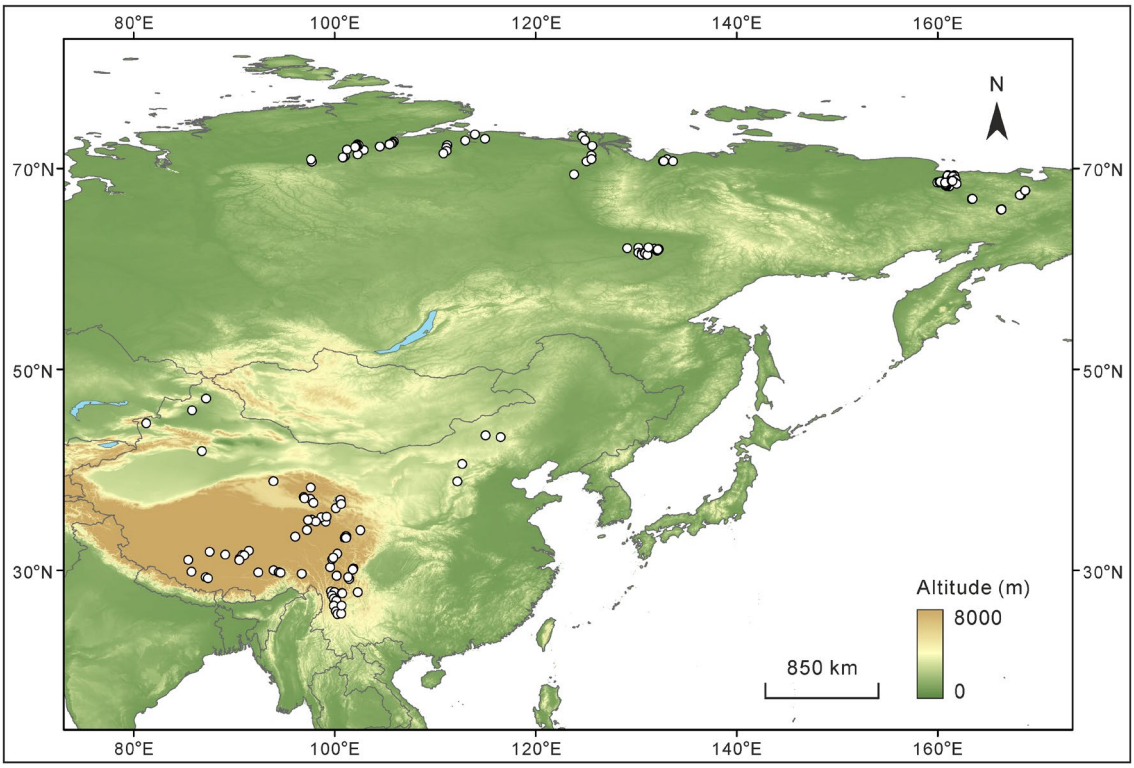


FIGURE 1 Location of the 219 sampled lakes in this study

(Binladen et al., 2007; Taberlet et al., 2018) were used and therefore not sequenced them individually. The only exception is that we produced up to six PCR replicates for most 16-KP samples from Chukotka only because they represent intra-lake samples. All PCR replicates were merged for the final interpretation. The same PCR protocol was applied for all samples to minimize potential biases in the results. After evaluation of PCR results, PCR products were subsequently purified using the MinElute PCR Purification Kit (Qiagen) and pooled in equimolar concentrations. Library preparation and next-generation sequencing on the Illumina platform were performed with four independent sequencing runs at Fasteris SA, Switzerland. Detailed descriptions of experimental steps are given in Stoof-Leichsenring et al. (2020).

Raw sequence data were directly reused from Stoof-Leichsenring et al. (2020) and processed by the OBITools package (Boyer et al., 2016), which includes *illuminapairedend* to align forward and reverse reads and then *ngsfilter* to assign the sequences to the samples. We subsequently used *obigrep* to exclude sequences shorter than 10 bp and with <10 sequence counts, and *obiuniq* to merge duplicated sequences. The *obiclean* program was run to remove putative PCR or sequencing errors. Taxonomic assignments with *ecotag* were finally performed without a defined identity threshold. Two reference libraries as described in Epp et al. (2015) were used to assign the sequences to taxa, which are the Arctic-Boreal vascular plant and bryophyte database (including 1664 vascular plants and 486 bryophyte species, published by Sønstebø et al. (2010), Willerslev et al. (2014), and Soininen et al. (2015)) and the European Molecular Biology Laboratory (EMBL) nucleotide database version 138 (Kanz et al., 2005; http://www.ebi.ac.uk/ena). We extracted sequences of the targeted region from the EMBL nucleotide database using ecoPCR (Ficetola et al., 2010). To maximize the number of sequences in the EMBL reference library, we allowed five mismatches between primers and target sequences in the ecoPCR analyses (Stoof-Leichsenring et al., 2020).

After running OBITools, four proxies were set up to quantify the extent of sedimentary DNA preservation/degradation. First, the percentage of sequence counts with the best identity ≥95% ($PCT_{id\geq 95\%}$), which is based on the assumption that the proportion of sequences that can be assigned to an entry in the databases (best identity ≥95%), is higher under ideal than under poor DNA preservation conditions. Second, weighted average identity (WAI) assumes that well-preserved DNA molecules have higher best identity values than poorly preserved DNA molecules. This proxy can be written as follows, for one sample:

$$\mathrm{WAI} = \sum_{i=1}^{n} \frac{x_i}{x}[ID_i]$$

where $n$ is the number of sequences, $x_i$ is the count of one sequence $i$, $x$ is the total count of all sequences, and $ID_i$ is the best identity of sequence $i$. Third, weighted average DNA fragment length (WAFL) is based on the assumption that DNA molecules are hydrolyzed into shorter fragments under poor than under ideal preservation conditions (Hofreiter et al., 2001; Lindahl, 1993; Willerslev & Cooper, 2005). This proxy can be written as follows, for one sample:

$$\mathrm{WAFL} = \sum_{i=1}^{n} \frac{x_i}{x}[FL_i]$$

where $n$ is the number of sequences, $x_i$ is the count of one sequence $i$, $x$ is the total count of all sequences, and $FL_i$ is the fragment length of sequence $i$. Fourth, taxonomic richness of terrestrial seed plants assumes that more terrestrial plant taxa can be detected in the samples under ideal than under poor DNA preservation conditions. To estimate the taxonomic richness, we excluded the following taxa from the dataset: (i) taxa with less than 100% identity; (ii) taxa that are not naturally found in China and Siberia; (iii) taxa that occur only once in the whole dataset; (iv) taxa that occur in the extraction blanks and NTCs with significantly high sequence counts. The filtered count data were subsequently subjected to rarefaction analysis (Birks & Line, 1992; the R code is available at https://doi.org/10.5281/zenodo.4562708). Detailed explanations of these four preservation proxies are given in section 4.1.

A series of BRT analyses (Elith et al., 2008), a flexible regression technique based on machine learning, were run to assess how each of the four proxies for sedimentary DNA preservation is related to five environmental variables (lake water conductivity, lake water pH, JulyT, sampling depth, and lake size). We also introduced number of years between field sampling and DNA extraction as a sixth environmental variable in the models and tested whether it was important for sedimentary DNA preservation. However, as this variable did not show a clear relationship with any preservation proxy, it was excluded from further analyses (Appendix S3). All BRT models were generated using the *gbm.step* function in the R-package *dismo* (Hijmans et al., 2017). The cross-validation program was first run to estimate the optimal number of trees and minimize the predictive deviance of the model. We used a Gaussian error distribution, a default bag fraction of 0.5, a slow learning rate of 0.0005, and a tree complexity of 2. Such a combination of parameters can achieve relatively low predictive error and high predictive accuracy of the model. Some tests with other (default) parameters indicated that our results were quite robust to the parametrization of the BRT model. Lake water conductivity, lake size, and sampling depth were log-transformed prior to analyses to ease visual inspection of the BRT output plots. Results from applying the *gbm.interactions* function were investigated to quantify interactions between the environmental variables in the BRT model. All statistical analyses were performed in RStudio (RStudio Team, 2015) with the R software (version 3.5.3; R Core Team, 2019). Pearson's correlation analysis was run using the *cor* and *corr.test* functions in the R-package *psych* (Revelle, 2021).

## 3 | RESULTS

Over 35 million sequence counts were assigned to more than 13,000 taxa, with 623 taxa having 100% sequence similarity with the reference libraries. Of these, more than 70% are assigned to terrestrial

plant taxa, and about half of these terrestrial plant taxa could be identified to species level. $PCT_{id\geq95\%}$, WAI, WAFL, and rarefied richness range from 0.25% to 99.93% (mean 77.5%), 59.91% to 99.96% (mean 93.09%), 16 to 86 bp (mean 53 bp), and 0 to 28 taxa (mean 10 taxa), respectively (Appendix S4). To keep as many samples as possible in the data analyses, we performed rarefaction analysis based on a very low sequence count ($n = 185$, SET-24), which is likely to be the main reason for the overall low rarefied richness in this study. For example, 80 unique terrestrial seed plant taxa are detected in Lake 13-TY-01 (Taymyria, Siberia), but its richness is reduced to 28.34 after rarefaction analysis. $PCT_{id\geq95\%}$ and WAI have a high positive correlation ($R = 0.96$, $p < 0.001$, $n = 219$). Though less strongly, WAFL is also positively correlated with $PCT_{id\geq95\%}$ ($R = 0.46$, $p < 0.001$, $n = 219$) and WAI ($R = 0.42$, $p < 0.001$, $n = 219$). Moderate or no correlations were found between rarefied richness and the three other proxies (Figure 2).

Figure 3 summarizes the relative contributions of the single environmental variables in the BRT model for each DNA preservation proxy. We found that all DNA preservation proxies are most strongly correlated to lake water conductivity, accounting for 28.4%–52.3% of the total relative influence. The fitted function values of all preservation proxies increase at low conductivities, reach a plateau at approximately 100–500 µS $cm^{-1}$, and decrease strongly at conductivities ≥1000 µS $cm^{-1}$ (Figure 4).

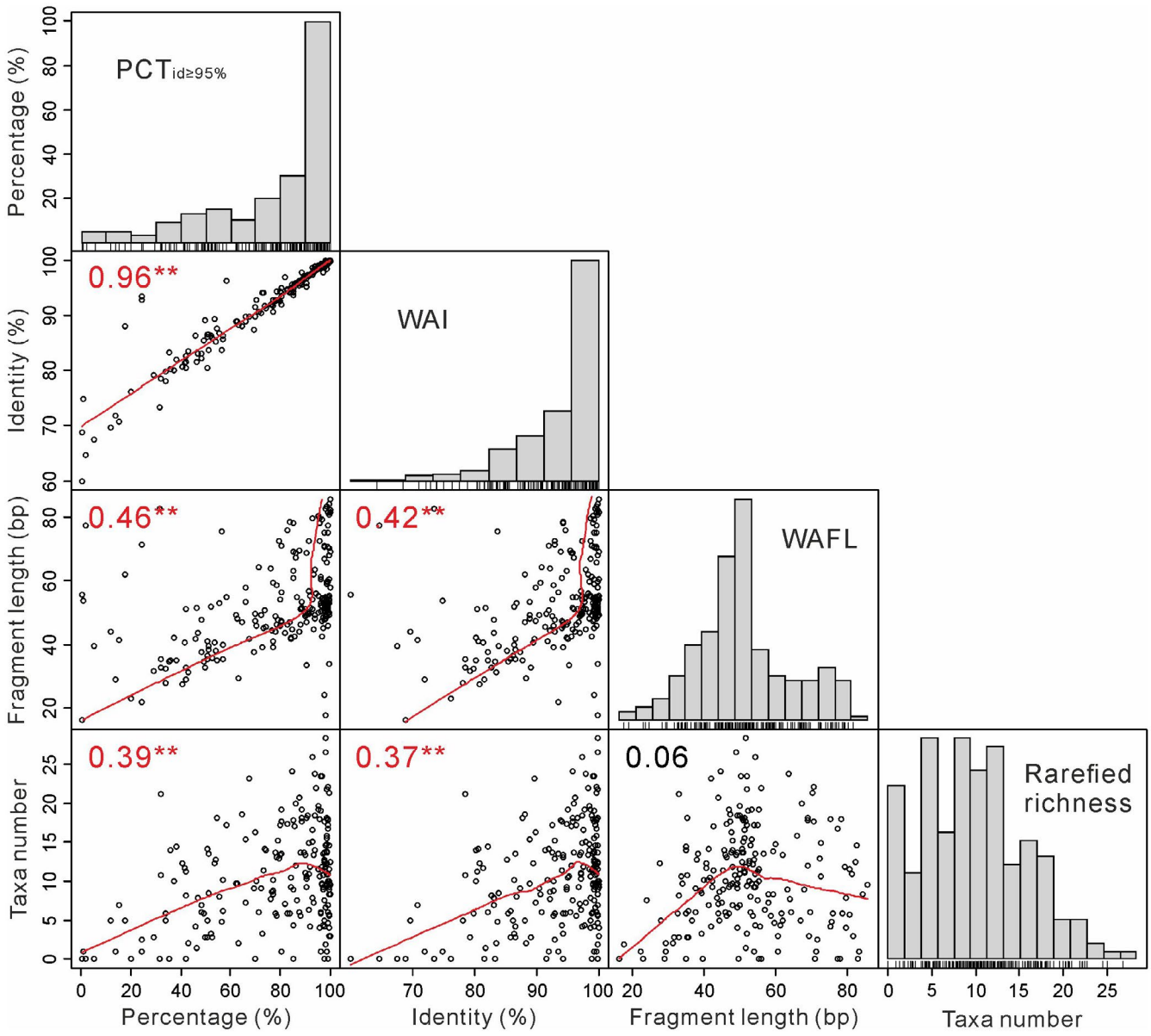


FIGURE 2 Pearson correlations between the four DNA preservation proxies proposed in this study, which are the percentage of sequence counts with the best identity ≥95% ($PCT_{id\geq95\%}$), weighted average identity (WAI), weighted average DNA fragment length (WAFL), and rarefied richness. The red curves in the scatter plots are LOESS curves fitted to the data. The double asterisk denotes a significant correlation at the 99% confidence level

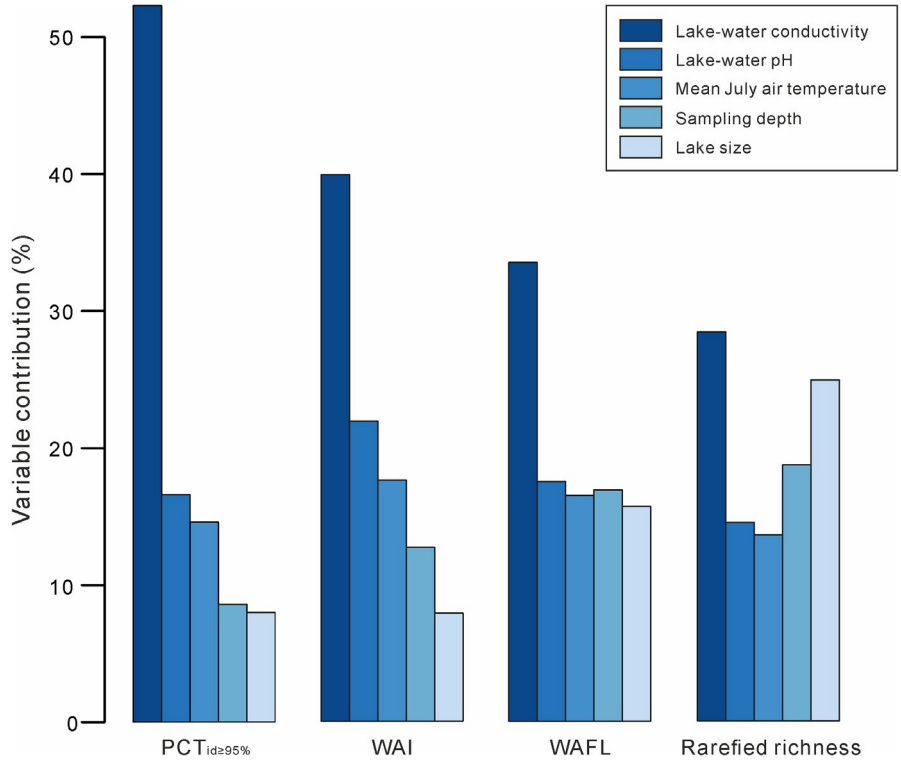


**FIGURE 3** Relative contributions of five environmental variables in the BRT models of four DNA preservation proxies, which are the percentage of sequence counts with the best identity ≥95% ($PCT_{id\geq95\%}$), weighted average identity (WAI), weighted average DNA fragment length (WAFL), and rarefied richness. Most variation in all DNA preservation proxies can be explained by lake water conductivity. Lake water pH is the second-most important variable in the models of $PCT_{id\geq95\%}$, WAI, and WAFL. Large variation in rarefied richness can be explained by lake size and sampling depth in addition to lake water conductivity

Lake water pH plays the second-most important role in the models of $PCT_{id\geq95\%}$, WAI, and WAFL, explaining 16.6%–21.9% of the total relative influence. The fitted function values of these three preservation proxies are relatively high for lake water pH between 7 and 9. Unlike these three proxies, lake size (24.9%) and sampling depth (18.7%) have a greater contribution than lake water pH (14.5%) in the model of rarefied richness, and high richness is found in small and deep lakes rather than big and shallow lakes (Figures 3 and 4).

Among the relative strength of pairwise interactions between the environmental variables, the one between lake water conductivity and lake water pH is by far the most important. We found that the fitted value of the model is relatively high when lake water conductivity is approximately 100–500 μS $cm^{-1}$ and lake water pH is approximately 7–9, as exemplified by the interaction plot of $PCT_{id\geq95\%}$ (Figure 5). Additionally, there is a statistically positive linear correlation between these two variables ($R = 0.65$, $p < 0.001$, $n = 219$).

The impact of the number of PCR replicates has been tested by merging only two PCR replicates for each sample (excluded 13-TY samples), and the output BRT results are almost the same (Appendix S5). In addition, rarefied richness of all taxa (i.e., terrestrial plants, aquatic plants, algae, ferns, and bryophytes) can be used in the same way as rarefied richness of terrestrial seed plants, and the similar BRT results can be outputted (Appendix S6).

## 4 | DISCUSSION

### 4.1 | $PCT_{id\geq95\%}$, WAI, WAFL, and rarefied richness as proxies for sedimentary DNA preservation

We introduce $PCT_{id\geq95\%}$, WAI, WAFL, and rarefied richness of terrestrial seed plants of plant DNA metabarcoding as proxies for sedimentary DNA preservation. Such proxies are needed because metabarcoding is currently the most widely used approach for sedimentary (ancient) DNA research, and assessing DNA preservation conditions directly from the output data would be highly valuable. Another more cost-intensive alternative is to apply metagenomic shotgun sequencing (Coissac et al., 2016), which can provide more information about DNA preservation. However, the majority of shotgun reads (>90%) cannot be fully analyzed because they cannot map any taxa in the reference libraries, which may be due to the incompleteness of genomic reference databases (Parducci, Alsos, et al., 2019; Pedersen et al., 2016; Slon et al., 2017). Currently, it is also very hard for shotgun sequencing to detect more plant taxa than metabarcoding at the same taxonomic levels (Parducci, Alsos, et al., 2019). Given that we only focus on modern plant DNA from lake surface sediments that can be well amplified by the *g-h* primers, metabarcoding seems to be a more useful and cost-efficient method for this study.

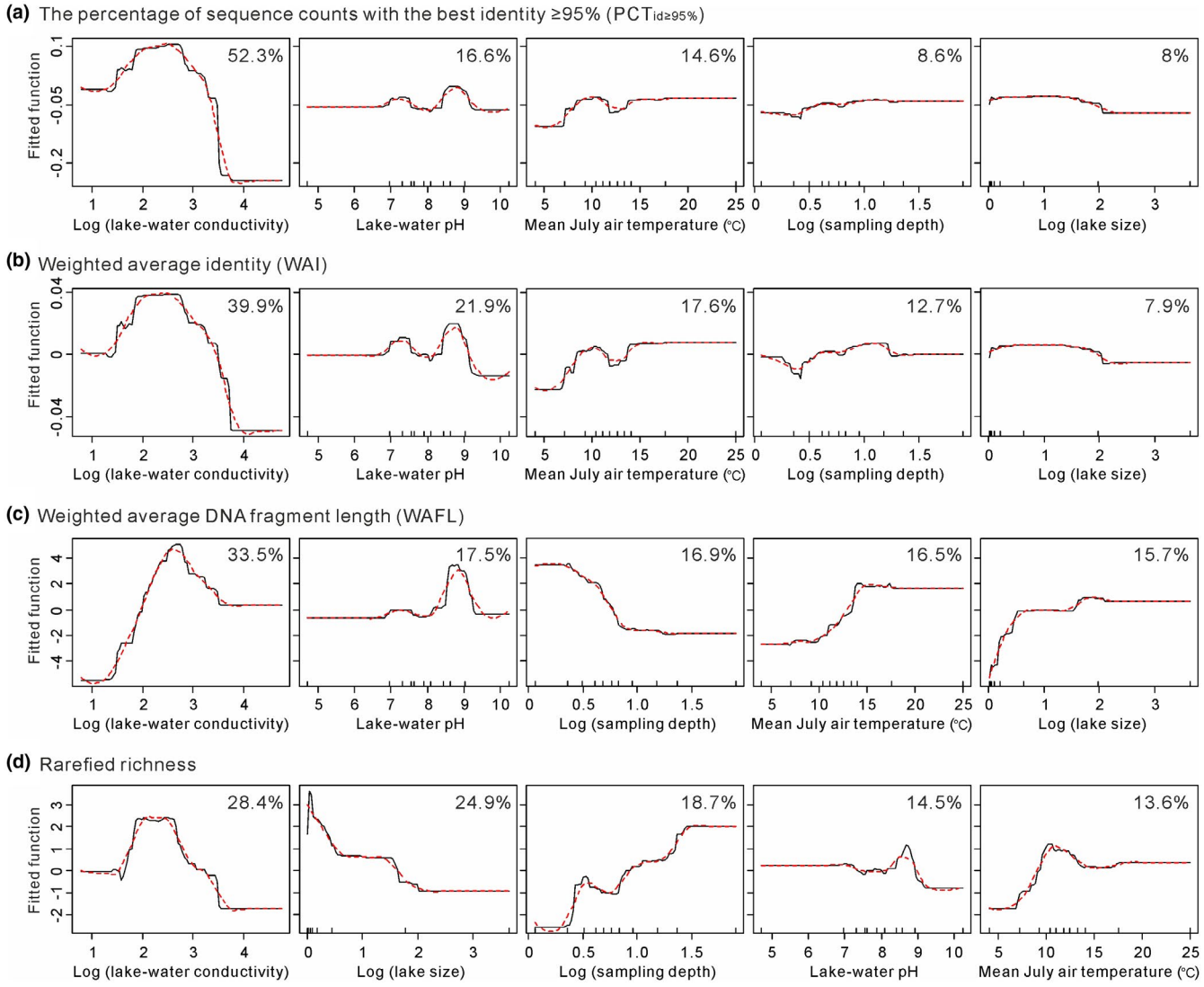


**FIGURE 4** Fitted functions between five environmental variables (ranked by relative contribution from left to right) and four DNA preservation proxies, which are (a) the percentage of sequence counts with the best identity ≥95% ($PCT_{id\geq 95\%}$), (b) weighted average identity (WAI), (c) weighted average DNA fragment length (WAFL), and (d) rarefied richness. The red dashed curves are a smoothed representation of the plots. Higher fitted function values indicate that the corresponding range of environmental variables is favorable for plant DNA preservation

Among the metabarcoding method, making use of plant DNA amplified by the "*g*" and "*h*" universal primers for the P6 loop region of the chloroplast *trn*L (UAA) intron to study sedimentary DNA preservation has several advantages. The primers are suitable for plant DNA studies because they amplify a short variable region with highly conserved sequences and there are a large number of reference sequences in public and customized databases available (Sønstebø et al., 2010; Taberlet et al., 2007; Willerslev et al., 2014). Chloroplast DNA occurs in high copy numbers in plant tissues and consequently has a better chance of being amplified (Morley & Nielsen, 2016; Sakamoto & Takami, 2018). Also, most amplified DNA is terrestrial plant DNA, which is assumed to be real allochthonous DNA transported by runoff from the catchment rather than in situ DNA from living aquatic macrophytes (Parducci, Nota, et al., 2019).

However, metabarcoding is highly dependent on PCR amplification, which also has several drawbacks. Template DNA (the P6 loop region) can be well amplified by the primers we used, but this method has limitations for the samples that lack targeted templates, which is probably caused by strong DNA fragmentation. Mismatches between the primers and multiple DNA templates can cause imbalances in amplification efficiency, which may overenrich or underenrich some taxa and lose the rare and hard-to-amplify taxa in our dataset (Jia, 2020; Piñol et al., 2015; Sønstebø et al., 2010; Taberlet et al., 2012); this is a major challenge in this field (Calderón-Sanou et al., 2019; Deiner et al., 2017). In addition, different bioinformatic strategies used for data filtering can also affect the final interpretation of metabarcoding results (e.g., Alberdi et al., 2018; Deiner et al., 2017; Kang et al., 2021). Therefore, there are still many uncertainties and limitations in the

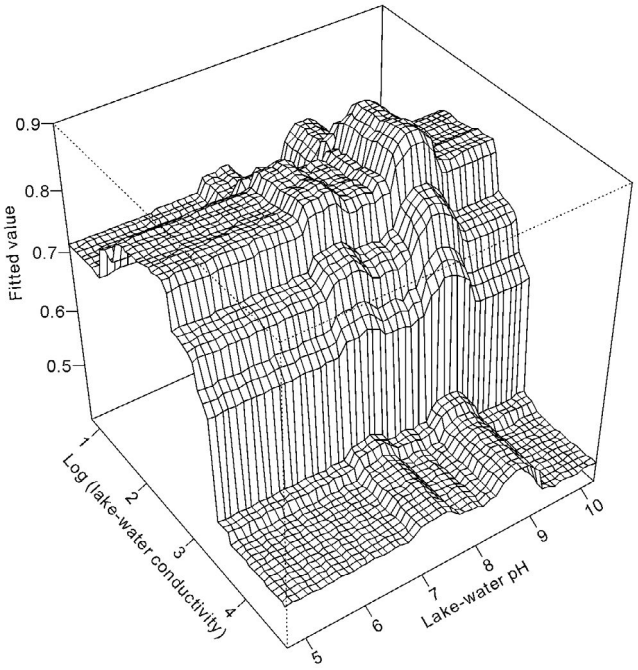


**FIGURE 5** Three-dimensional partial dependence plots for the strongest interaction in the BRT model of the percentage of sequence counts with the best identity ≥95% ($PCT_{id\geq95\%}$)

current workflow of DNA metabarcoding (Mathieu et al., 2020; Zinger et al., 2019), and the results of this study may need to be further assessed, for example, by non-targeting techniques such as shotgun sequencing.

$PCT_{id\geq95\%}$ and WAI are highly related to the best identity value given by *ecotag*. There are two main reasons for a low similarity between query and reference sequences under poor DNA preservation conditions: (i) the presence of extremely short (~10 bp) fragments, originating from strong DNA fragmentation, and long fragments (>220 bp), exceeding the theoretical maximum length of the marker (Calderón-Sanou et al., 2019; Taberlet et al., 2007). The latter can be produced by unspecific amplifications due to very low DNA template concentration. However, such fragments comprise only a small proportion of our dataset and are considered to have less impact on $PCT_{id\geq95\%}$ and WAI. (ii) The incorporation of mismatches caused by the degradation of DNA molecules such as depurination and deamination (Hofreiter et al., 2001; Lindahl, 1993; Willerslev & Cooper, 2005). Moreover, degradation can lead to low initial DNA template concentration, resulting in an increased number of PCR errors (Akbari et al., 2005; Taberlet et al., 1996), which can also create the differences between query and reference sequences. Aside from the effects of DNA preservation, the low best identity value can also be attributed to the incompleteness of the reference database. In this study, however, we used the same reference libraries for the whole dataset, and no strong regional patterns are found in $PCT_{id\geq95\%}$ and WAI (Appendix S7), indicating that both proxies are less biased by the incompleteness of the reference database in the different regions.

Fragment length has been widely used as a proxy to indicate DNA quality and preservation (e.g., Heintzman et al., 2014; Leino et al., 2009; Seutin et al., 1991). Under degradation conditions, strong DNA fragmentation allows only short fragments that still have primer binding sites to be amplified by PCR, whereas original long fragments that might lose primer binding sites are not amplifiable. Thus, average amplified fragment length can be used as an additional proxy for sedimentary DNA preservation. However, the marker we used is variable in fragment length (Taberlet et al., 2007), meaning that variations in fragment length may originate from variations in taxonomic composition rather than preservation characteristics (Alsos et al., 2018; Appendix S8). For example, the samples dominated by Potamogetonaceae (*Potamogeton*) might erroneously indicate good DNA preservation conditions because this taxon has a relatively long P6 loop fragment (~80 bp). Also, algae, ferns, and bryophytes usually have a relatively short P6 loop fragment (~10–20 bp), which may indicate poor DNA preservation conditions. However, only a small part of sequences can be assigned to these taxa in our dataset (<30% of all taxa with 100% identity), and most of the amplified DNA is terrestrial plant DNA with an average fragment length longer than 40 bp. Furthermore, WAFL has a positive correlation with $PCT_{id\geq95\%}$ ($R = 0.46$, $p < 0.001$, $n = 219$) and WAI ($R = 0.42$, $p < 0.001$, $n = 219$), which indicates that these three proxies might be intercorrelated and cross-validated. For example, we found that some low-WAFL samples (<40 bp) also have relatively low values in other preservation proxies (Figure 2).

Richness of terrestrial plant taxa has also been used as a proxy to study the taphonomic processes, including preservation, affecting sedimentary DNA records in recent years (e.g., Alsos et al., 2018; Giguet-Covex et al., 2019; Jia, 2020). However, it should be noted that this proxy is influenced not only by the preservation conditions of sedimentary plant DNA, but also by other taphonomic processes. For example, abundant aquatic macrophytes in the lake can decrease the ability to detect and amplify terrestrial plant DNA from sediments (Alsos et al., 2018, 2020; Rijal et al., 2021). Such dilution effect might explain the low rarefied richness found in some samples with a high proportion of *Potamogeton*. However, no significant correlation exists between the proportion of reads assigned to aquatic macrophytes and rarefied richness ($R = -0.13$, $p > 0.05$, $n = 197$), indicating that this effect is very weak. In addition, the rarefied richness of some lakes may be underestimated due to the incompleteness of the reference databases we used, especially for lakes located in remote areas with a unique plant community (e.g., the QTP), but no strong regional patterns are found in rarefied richness (Appendix S7). Furthermore, the influx of terrestrial materials into the lake also plays an important role in the detection of terrestrial plant DNA from sediments, which is related to many environmental factors, such as vegetation composition, DNA production, topographic relief, erosion dynamics, and the degree of connectivity across the catchment area (Giguet-Covex et al., 2019). Since we have very limited information on these factors for each lake, this issue requires further studies in the future. In our data, rarefied richness shows only a moderate or even no correlation with the three previous proxies, suggesting that these proxies might reflect different aspects of DNA preservation.

To conclude, although there are still some limitations in the four preservation proxies, the pattern of the BRT results of them is similar to some extent, which suggests that they are intercorrelated and cross-validated and could be useful proxies for sedimentary DNA preservation.

## 4.2 | Plant DNA preservation in relation to environmental variables

Our results indicate that plant DNA preservation is strongly related to lake water characteristics, particularly electrical conductivity and pH, while lake morphology and air temperature play minor roles. To our knowledge, this is the first systematic study on the impact of environmental conditions on the preservation of plant DNA in lake surface sediments that makes use of broad field-based information, while most previous studies on DNA preservation are based on laboratory experiments.

Plant DNA in lake sediments includes intracellular DNA, which belongs to plant fragments and living cells, and extracellular DNA, which is released into the surrounding environment by the degradation of plant tissues and the lysis of plant cells (Parducci et al., 2017; Taberlet et al., 2018). Because plant cells are lysed rapidly in the environment (Nielsen et al., 2007), extracellular DNA is considered the main component of total plant DNA in lake sediments (Parducci et al., 2017, 2019). From the moment of plant cell death, DNA repair mechanisms permanently cease and the plant DNA is released into soils and sediments. The rate of DNA degradation can be influenced by several factors (e.g., pH, temperature, UV radiation, microbial activity, and salinity; Giguet-Covex et al., 2019). Of these, microbial activity is considered to be the most direct factor to degrade extracellular DNA, because DNA molecules can be enzymatically hydrolyzed by DNases produced by bacteria, resulting in DNA strand damage and breakage (Blum et al., 1997; Pedersen et al., 2015; Strickler et al., 2015; Torti et al., 2015). Despite this, extracellular DNA can be well protected from the attack of nucleases by adsorbing to inorganic or organic surface reactive particles, such as clay minerals and humic substances (Cai et al., 2006; Crecchio & Stotzky, 1998; Greaves & Wilson, 1969; Nagler et al., 2018; Pedreira-Segade et al., 2018; Romanowski et al., 1991; Taberlet et al., 2018; Yu et al., 2013), which seems to be the primary mechanism of extracellular DNA persistence in soils or sediments (Blum et al., 1997; Kanbar et al., 2020; Ogram et al., 1988; Pietramellara et al., 2009). Adsorption inhibits the activation and transformation of DNases and separates the enzymes from their substrate (Demanèche et al., 2001; Khanna & Stotzky, 1992). The adsorption strength of extracellular DNA on soil and sediment particles is controlled by several environmental variables, such as the mineralogy of sorbent, ionic strength, pH, and DNA fragment length (Ogram et al., 1988).

Our results suggest that the electrical conductivity and pH of lake water might have a stronger effect on the preservation of sedimentary plant DNA than other environmental variables, and plant DNA tends to persist longer in lakes with intermediate water conductivities (100–500 μS $cm^{-1}$) and neutral to slightly alkaline water pH (7–9).

Previous studies have indicated that extracellular DNA adsorption is a charge-dependent process influenced by pH and cation concentration of the solution (Romanowski et al., 1991). The surface charge density of DNA is constant for a given pH (Romanowski et al., 1991). The isoelectric point of DNA is pH 5. At pH < 5, the phosphate moieties of DNA become protonated and exhibit a net positive charge, which is opposite to the surface charge of most clay minerals (e.g., montmorillonite and kaolinite) and humic substances. This means that extracellular DNA can be easily adsorbed to soil or sediment particles (Greaves & Wilson, 1969). However, acidic conditions catalyze hydrolytic processes and accelerate DNA degradation (Giguet-Covex et al., 2019; Lindahl, 1993; Lindahl & Nyberg, 1972; Seymour et al., 2018). At pH > 5, the phosphate moieties of DNA become deprotonated and exhibit a net negative charge, which is unfavorable for DNA adsorption (Greaves & Wilson, 1969). Furthermore, the electrostatic repulsion between DNA and soil or sediment particles increases with increasing pH values (Levy-Booth et al., 2007).

Electrical conductivity is the ability of water to conduct an electrical current (Davis & De Wiest, 1966), which is positively correlated with the ionic concentration of the solution (Zhuiykov, 2018). Ionic strength is a measure of the intensity of an electric field in the solution, which is positively related to the molar concentration and charge number of ions (Lewis & Randall, 1921). Thus, there can be a positive correlation between electrical conductivity and ionic strength, as has been evidenced by many previous studies (e.g., Alva et al., 1991; Gillman & Bell, 1978; Griffin & Jurinak, 1973; Ponnamperuma et al., 1966). In general, increases in ionic strength and electrical conductivity lead to increases in ionic concentration, which can synchronously increase cation concentration. Cations (e.g., $Ca^{2+}$, $Mg^{2+}$, $Na^{+}$, and $K^{+}$) play an important role in the process of DNA adsorption at pH > 5 because they can form bridges between the phosphate moieties of DNA and the particle surface and enhance DNA adsorption capacity (Greaves & Wilson, 1969; Khanna & Stotzky, 1992; Levy-Booth et al., 2007; Lorenz & Wackernagel, 1987; Paget et al., 1992; Pedreira-Segade et al., 2018; Romanowski et al., 1991). For example, Greaves and Wilson (1969) found that the adsorption of DNA by montmorillonite was greatly increased at all pH conditions in a buffered system with high concentrations of $Na^{+}$ ions. Romanowski et al. (1991) reported that more sand-adsorbed DNA could be detected when the water column had higher concentrations of $Mg^{2+}$ ions, even if the pH increased. In addition, the rate of depurination in DNA is relatively low at high ionic strength or electrical conductivity, which maintains the stability of DNA structure (Lindahl & Nyberg, 1972). Therefore, lake water conductivity could be a predominant variable influencing the preservation of sedimentary plant DNA under alkaline lake water pH conditions.

According to the fitted function curves (Figure 4), lake water conductivity tends to rise and reach a plateau at approximately 100–500 μS $cm^{-1}$, and decrease strongly at above 1000 μS $cm^{-1}$, indicating that high lake water conductivities might be unfavorable for the preservation of sedimentary plant DNA. The reason for this

phenomenon is highly due to high lake water pH. We notice that the threshold point of lake water conductivity (approximately 500 μS $cm^{-1}$) corresponds to a pH value of 9 due to their positive linear correlation ($R = 0.65$, $p < 0.001$, $n = 219$). Previous studies have shown that the negative charge of DNA and soil or sediment particles at pH ≥ 9 can highly reduce the efficiency of cations to mediate adsorption, and DNA can be desorbed almost completely from the negatively charged surface of clay minerals (Cai et al., 2006; Jia, 2020; Khanna & Stotzky, 1992; Levy-Booth et al., 2007; Ogram et al., 1988; Romanowski et al., 1991) even if their surface cations are saturated (Sheng et al., 2019). Another possible reason worthy of consideration is competitive adsorption. The amount of adsorbed DNA was found to be markedly reduced in the presence of additional phosphate, indicating that there was a strong competitive adsorption between phosphate groups ($PO_4^{3-}$) and DNA molecules on the clay mineral surface (Pietramellara et al., 2001; Yu et al., 2013). Before sediment particles are deposited and buried in the lake bottom, these desorbed DNA molecules will be free within the water column and available to DNase attack (Figure 6). In addition to these possibilities, the reasons for poor DNA preservation conditions at high lake water conductivities (≥1000 μS $cm^{-1}$) need to be further studied in the future and would require detailed ion measurements to rule out their putative function in DNA adsorption.

We notice that large variation in rarefied richness can be explained by lake size and sampling depth in addition to lake water conductivity (Figures 3 and 4). The surface sediments of shallow lakes or ponds might be exposed to high-level UV radiation, which can photochemically damage DNA (Ravanat et al., 2001; Strickler et al., 2015). On the other hand, if lakes are deep enough, the water column becomes thermally stratified, creating a cold (about 4°C) and frequently anoxic environment at the lake bottom, which will limit microbial activity and help preserve plant DNA (Parducci et al., 2017). In addition, underflows are common in many deep lakes with steep bottom slopes, which favors the concentration of terrestrial sediments at the lake bottom (Mirbach & Lang, 2018). We also find that the samples from large lakes (≥5 $km^2$) have lower rarefied richness than small lakes, which is not in accordance with our expectation. Large lakes, usually along with relatively wide catchment areas and well-developed hydrographical networks, are assumed to have more terrestrial inputs than small lakes or ponds that only receive inflow from limited streams. There are four hypotheses to explain this phenomenon. (i) Large lakes represent less than 20% of the total number of lakes in our dataset, which can strongly affect the degree of curve fitting in our BRT models. (ii) Nearly 80% of the large lakes in this study are located in the arid and semi-arid regions of China (e.g., the northern and central QTP, Inner Mongolia, and Xinjiang). The main vegetation types in this region are steppe and desert. In contrast, most small lakes are located in the humid and semi-humid regions (e.g., the southeastern QTP and Siberia; Appendix S9), where the vegetation types are dominated by forest, meadow, and shrub (Hou, 2001; Stone & Schlesinger, 2003). Hence, the overall plant richness of large lakes might be lower than that of small lakes because of the differences in vegetation composition and diversity in different regions. (iii) Many large lakes, especially Tibetan lakes in this study, have a very high water pH (Appendix S9), which is unfavorable for plant DNA preservation. (iv) The center of a small lake (or pond) is close to the lakeshore and local terrestrial plant materials and their DNA might be more easily transported into the lake by runoff from a small catchment (Alsos et al., 2018; Niemeyer et al., 2017). The relationships between rarefied richness and environmental variables, therefore, might be more complicated than other relationships described in this study and require further research.

Interestingly, our data do not provide clear evidence that mean July air temperature has a significant influence on the preservation

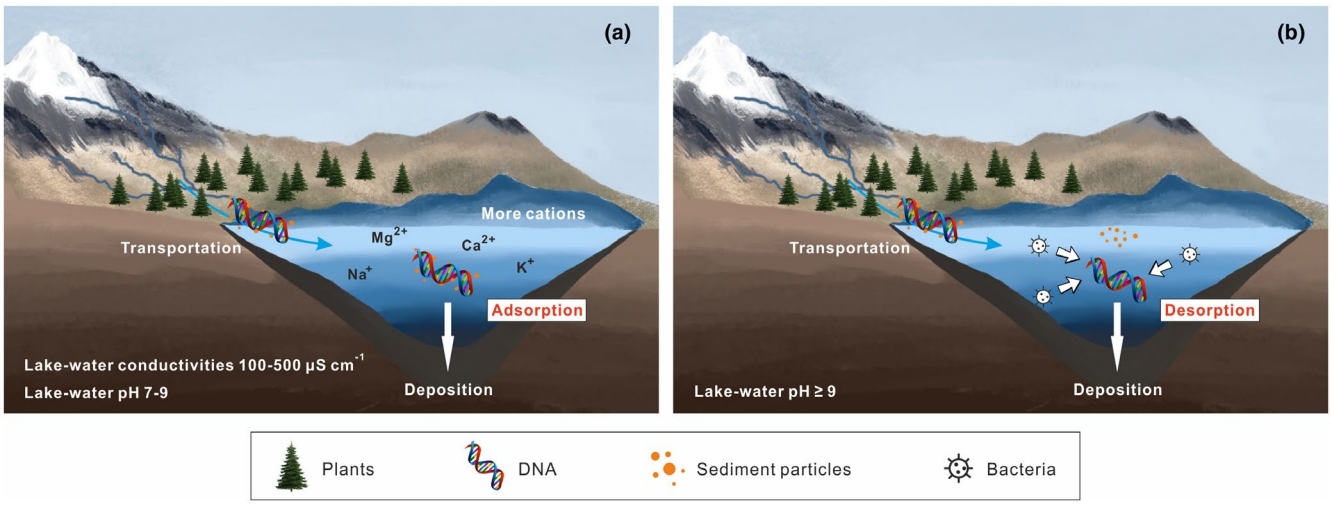


**FIGURE 6** Conceptual model of the preservation of plant DNA in lake sediments and its relationships with lake water chemistry. (a) Intermediate water conductivities (100–500 μS $cm^{-1}$) and neutral to slightly alkaline lake water pH (7–9) facilitate the adsorption of plant DNA to sediment particles, which is favorable for plant DNA preservation. (b) High lake water pH (≥9) leads to desorption of plant DNA from sediment particles, and DNA molecules are free within the water column and available to DNases produced by bacteria

of plant DNA in lake sediments. High temperature was considered a profound cause of DNA degradation (Hofreiter et al., 2001; Lindahl, 1993; Lindahl & Nyberg, 1972; Willerslev & Cooper, 2005). However, only approximately 15% of the total relative influence can be explained by JulyT in the BRT models, indicating that air temperature might not be a key variable to limit plant DNA preservation. For example, the QTP, with high elevation and low mean annual temperature, is theoretically an ideal place for DNA preservation (Hofreiter et al., 2014; Jia, 2020). However, plant DNA is not well preserved in the surface sediments of some lakes in the central QTP (e.g., Lake Pengco, Lake Bamuco, and Lake Selinco) due to their high water pH (Appendix S1). Furthermore, given that several previous studies have extracted PCR-amplifiable plant DNA from tropical lake sediments (Boessenkool et al., 2014; Bremond et al., 2017; Tabares et al., 2020), it is possible that the preservation of sedimentary plant DNA is more influenced by lake water chemistry than meteorological factors.

Although there are knowledge gaps in this field, the metabarcoding-based results from this paper could be used to design sampling strategies and to pre-select suitable lakes for studying sedimentary plant DNA, thus reducing additional time and costs during field work. Our results indicate that lake water chemistry should be given priority when considering study site selection. Lakes with intermediate water conductivities (100–500 μS $cm^{-1}$) and neutral to slightly alkaline water pH (7–9) may well preserve sedimentary plant DNA better than lakes with low or high water conductivities and acidic or strongly alkaline water pH. Furthermore, more terrestrial plant taxa might be detectable from deep lakes than from shallow lakes. Air temperature is not a key variable for study site selection. When considering these recommendations, we should keep in mind that not all potential environmental variables that affect sedimentary DNA preservation have been included in this study. For example, the importance of sediment type with respect to the mineralogy and grain-size composition of sediments for DNA preservation has widely been reported by previous studies (e.g., Freeman et al., 2020; Kanbar et al., 2020; Levy-Booth et al., 2007; Romanowski et al., 1991), and this may be one of the missing variables in our BRT models. In addition, the preservation conditions of upper modern DNA might differ from deep ancient DNA, which may also be impacted by other environmental factors (e.g., burial time and tectonic activity; Hofreiter et al., 2001; Pedersen et al., 2016).

## 5 | CONCLUSIONS

Using the amplified plant DNA records from 219 lake surface sediments from China and Siberia, we were able to quantify the effect of five environmental variables (lake water conductivity, lake water pH, JulyT, sampling depth, and lake size) on four proxies we introduced for sedimentary DNA preservation ($PCT_{id\geq 95\%}$, WAI, WAFL, rarefied richness) via BRT models. Our results suggest that electrical conductivity and pH of lake water are the two most important variables for the preservation of sedimentary plant DNA. Lakes with intermediate water conductivities (100–500 μS $cm^{-1}$) and neutral to slightly alkaline water pH (7–9) may be suitable for sedimentary DNA studies. Deep lakes seem to support plant DNA preservation as indicated by relatively high rarefied richness. The relationship between rarefied richness and lake size requires further assessment. Air temperature does not appear to be a key variable to limit the preservation of plant DNA in lake sediments.

It should also be noted that the conclusions are partially limited by the metabarcoding method, and non-targeting techniques such as metagenomics may provide more insightful information about DNA preservation in the future. In addition, not all potential environmental variables that affect sedimentary DNA preservation (e.g., catchment characteristics and sediment types) have been considered in this study. For sedimentary ancient DNA, we should carefully follow the above recommendations, because its preservation mechanism is even more complicated and can also be influenced by other environmental factors such as burial time and tectonic activity, which is different from that of DNA in lake surface sediments.

## ACKNOWLEDGEMENTS

This study was supported by a corporative project of the National Natural Science Foundation of China (NSFC) and the German Research Foundation (DFG; Grant No. 41861134030), and the Second Tibetan Plateau Scientific Expedition and Research Program (STEP), Chinese Academy of Sciences (CAS; Grant No. 2019QZKK0202). Weihan Jia received funding from Capital Normal University (Grant No. 028185511700) and the European Research Council (Glacial*Legacy*; Grant No. 772852) during his stay at AWI in Germany. We thank Junfeng Li, Yu Chen, Peng Han, Huashu Li, Xianyong Cao, Jian Ni, Steffen Mischke, and Luidmila A. Pestraykova for their support during field work, and Naimeng Zhang, Sichao Huang, Bastian Niemeyer, Sarah Olischläger, and Laura S. Epp for supporting us with the laboratory work. Qi Zhang is thanked for her help with Figure 6. We are thankful to Yiping Su for his helpful discussions. We also acknowledge Cathy Jenks for the English language editing.

## CONFLICT OF INTEREST

None declared.

## AUTHOR CONTRIBUTIONS

All authors conceived the study. W.J. and K.L. performed the laboratory work (guided by K.S.-L. and S.L.). W.J. applied the data analyses (guided by U.H., K.S.-L., and X.L.) and wrote the first draft of the manuscript. All co-authors reviewed and edited the final manuscript.

## DATA AVAILABILITY STATEMENT

DNA sequence data can be retrieved at Dryad (https://doi.org/10.5061/dryad.k6djh9w4r). Environmental data are described in Appendix S1 and available at Pangaea (https://doi.pangaea.de/10.1594/PANGAEA.920866).

## ORCID

*Weihan Jia* https://orcid.org/0000-0003-3707-409X

## REFERENCES


Akbari, M., Hansen, M. D., Halgunset, J., Skorpen, F., & Krokan, H. E. (2005). Low copy number DNA template can render polymerase chain reaction error prone in a sequence-dependent manner. *The Journal of Molecular Diagnostics*, *7*(1), 36–39. https://doi.org/10.1016/S1525-1578(10)60006-2

Alberdi, A., Aizpurua, O., Gilbert, M. T. P., & Bohmann, K. (2018). Scrutinizing key steps for reliable metabarcoding of environmental samples. *Methods in Ecology and Evolution*, *9*(1), 134–147. https://doi.org/10.1111/2041-210X.12849

Alsos, I. G., Lammers, Y., Yoccoz, N. G., Jørgensen, T., Sjögren, P., Gielly, L., & Edwards, M. E. (2018). Plant DNA metabarcoding of lake sediments: How does it represent the contemporary vegetation. *PLoS ONE*, *13*(4), e0195403. https://doi.org/10.1371/journal.pone.0195403

Alsos, I. G., Sjögren, P., Brown, A. G., Gielly, L., Merkel, M. K. F., Paus, A., Lammers, Y., Edwards, M. E., Alm, T., Leng, M., Goslar, T., Langdon, C. T., Bakke, J., & van der Bilt, W. G. M. (2020). Last Glacial Maximum environmental conditions at Andøya, northern Norway; evidence for a northern ice-edge ecological "hotspot". *Quaternary Science Reviews*, *239*, 106364. https://doi.org/10.1016/j.quascirev.2020.106364

Alva, A. K., Sumner, M. E., & Miller, W. P. (1991). Relationship between ionic strength and electrical conductivity for soil solutions. *Soil Science*, *152*(4), 239–242. https://doi.org/10.1097/00010694-199110000-00001

Anderson-Carpenter, L. L., McLachlan, J. S., Jackson, S. T., Kuch, M., Lumibao, C. Y., & Poinar, H. N. (2011). Ancient DNA from lake sediments: Bridging the gap between paleoecology and genetics. *BMC Evolutionary Biology*, *11*, 30. https://doi.org/10.1186/1471-2148-11-30

Binladen, J., Gilbert, M. T. P., Bollback, J. P., Panitz, F., Bendixen, C., Nielsen, R., & Willerslev, E. (2007). The use of coded PCR primers enables high-throughput sequencing of multiple homolog amplification products by 454 parallel sequencing. *PLoS ONE*, *2*(2), e197. https://doi.org/10.1371/journal.pone.0000197

Birks, H. J. B., & Birks, H. H. (2016). How have studies of ancient DNA from sediments contributed to the reconstruction of Quaternary floras? *New Phytologist*, *209*(2), 499–506. https://doi.org/10.1111/nph.13657

Birks, H. J. B., & Line, J. M. (1992). The use of rarefaction analysis for estimating palynological richness from Quaternary pollen-analytical data. *Holocene*, *2*(1), 1–10. https://doi.org/10.1177/095968369200200101

Blum, S. A. E., Lorenz, M. G., & Wackernagel, W. (1997). Mechanism of retarded DNA degradation and prokaryotic origin of DNases in nonsterile soils. *Systematic and Applied Microbiology*, *20*(4), 513–521. https://doi.org/10.1016/S0723-2020(97)80021-5

Boessenkool, S., Mcglynn, G., Epp, L. S., Taylor, D., Pimentel, M., Gizaw, A., Nemomissa, S., Brochmann, C., & Popp, M. (2014). Use of ancient sedimentary DNA as a novel conservation tool for high-altitude tropical biodiversity. *Conservation Biology*, *28*(2), 446–455. https://doi.org/10.1111/cobi.12195

Boyer, F., Mercier, C., Bonin, A., Le Bras, Y., Taberlet, P., & Coissac, E. (2016). OBITOOLS: a UNIX-inspired software package for DNA metabarcoding. *Molecular Ecology Resources*, *16*(1), 176–182. https://doi.org/10.1111/1755-0998.12428

Bremond, L., Favier, C., Ficetola, G. F., Tossou, M. G., Akouégninou, A., Gielly, L., Giguet-Covex, C., Oslisly, R., & Salzmann, U. (2017). Five thousand years of tropical lake sediment DNA records from Benin. *Quaternary Science Reviews*, *170*, 203–211. https://doi.org/10.1016/j.quascirev.2017.06.025

Briggs, A. W., Stenzel, U., Johnson, P. L. F., Green, R. E., Kelso, J., Prufer, K., Meyer, M., Krause, J., Ronan, M. T., Lachmann, M., & Paabo, S. (2007). Patterns of damage in genomic DNA sequences from a Neandertal. *Proceedings of the National Academy of Sciences of the United States of America*, *104*(37), 14616–14621. https://doi.org/10.1073/pnas.0704665104

Brown, T. A., & Barnes, I. M. (2015). The current and future applications of ancient DNA in Quaternary science. *Journal of Quaternary Science*, *30*(2), 144–153. https://doi.org/10.1002/jqs.2770

Cai, P., Huang, Q., Zhang, X., & Chen, H. (2006). Adsorption of DNA on clay minerals and various colloidal particles from an Alfisol. *Soil Biology and Biochemistry*, *38*(3), 471–476. https://doi.org/10.1016/j.soilbio.2005.05.019

Calderón-Sanou, I., Munkemuller, T., Boyer, F., Zinger, L., & Thuiller, W. (2019). From environmental DNA sequences to ecological conclusions: How strong is the influence of methodological choices? *Journal of Biogeography*, *47*(1), 193–206. https://doi.org/10.1111/jbi.13681

Capo, E., Giguet-Covex, C., Rouillard, A., Nota, K., Heintzman, P. D., Vuillemin, A., Ariztegui, D., Arnaud, F., Belle, S., Bertilsson, S., Bigler, C., Bindler, R., Brown, A. G., Clarke, C. L., Crump, S. E., Debroas, D., Englund, G., Ficetola, G. F., Garner, R. E., … Parducci, L. (2021). Lake sedimentary DNA research on past terrestrial and aquatic biodiversity: Overview and recommendations. *Quaternary*, *4*(1), 6. https://doi.org/10.3390/quat4010006

Clarke, C. L., Edwards, M. E., Gielly, L., Ehrich, D., Hughes, P. D. M., Morozova, L. M., Haflidason, H., Mangerud, J., Svendsen, J. I., & Alsos, I. G. (2019). Persistence of arctic-alpine flora during 24,000 years of environmental change in the Polar Urals. *Scientific Reports*, *9*(1), 19613. https://doi.org/10.1038/s41598-019-55989-9

Coissac, E., Hollingsworth, P. M., Lavergne, S., & Taberlet, P. (2016). From barcodes to genomes: extending the concept of DNA barcoding. *Molecular Ecology*, *25*(7), 1423–1428. https://doi.org/10.1111/mec.13549

Crecchio, C., & Stotzky, G. (1998). Binding of DNA on humic acids: Effect on transformation of *Bacillus subtilis* and resistance to DNase. *Soil Biology and Biochemistry*, *30*(8–9), 1061–1067. https://doi.org/10.1016/S0038-0717(97)00248-4

Crump, S. E., Fréchette, B., Power, M., Cutler, S., de Wet, G., Raynolds, M. K., Raberg, J. H., Briner, J. P., Thomas, E. K., Sepúlveda, J., Shapiro, B., Bunce, M., & Miller, G. H. (2021). Ancient plant DNA reveals High Arctic greening during the Last Interglacial. *Proceedings of the National Academy of Sciences of the United States of America*, *118*(13), e2019069118. https://doi.org/10.1073/pnas.2019069118

Davis, S. N., & De Wiest, R. J. M. (1966). *Hydrogeology*. John Wiley and Sons.

Deiner, K., Bik, H. M., Machler, E., Seymour, M., Lacoursiere-Roussel, A., Altermatt, F., & Bernatchez, L. (2017). Environmental DNA metabarcoding: Transforming how we survey animal and plant communities. *Molecular Ecology*, *26*(21), 5872–5895. https://doi.org/10.1111/mec.14350

Demanèche, S., Jocteur-Monrozier, L., Quiquampoix, H., & Simonet, P. (2001). Evaluation of biological and physical protection against nuclease degradation of clay-bound plasmid DNA. *Applied and Environmental Microbiology*, *67*(1), 293–299. https://doi.org/10.1128/AEM.67.1.293-299.2001

Dopheide, A., Xie, D., Buckley, T. R., Drummond, A. J., & Newcomb, R. D. (2018). Impacts of DNA extraction and PCR on DNA metabarcoding estimates of soil biodiversity. *Methods in Ecology and Evolution*, *10*(1), 120–133. https://doi.org/10.1111/2041-210X.13086

Edwards, M. E. (2020). The maturing relationship between Quaternary paleoecology and ancient sedimentary DNA. *Quaternary Research*, *96*, 39–47. https://doi.org/10.1017/qua.2020.52

Eglinton, G., & Logan, G. A. (1991). Molecular preservation. *Philosophical Transactions of the Royal Society B-Biological Sciences*, *333*(1268), 315–328. https://doi.org/10.1098/rstb.1991.0081

Elith, J., Leathwick, J. R., & Hastie, T. (2008). A working guide to boosted regression trees. *Journal of Animal Ecology*, *77*(4), 802–813. https://doi.org/10.1111/j.1365-2656.2008.01390.x

Epp, L. S., Gussarova, G., Boessenkool, S., Olsen, J., Haile, J., Schrøder-Nielsen, A., Ludikova, A., Hassel, K., Stenøien, H. K., Funder, S., Willerslev, E., Kjær, K., & Brochmann, C. (2015). Lake sediment multi-taxon DNA from North Greenland records early post-glacial appearance of vascular plants and accurately tracks environmental changes. *Quaternary Science Reviews*, *117*, 152–163. https://doi.org/10.1016/j.quascirev.2015.03.027

Ficetola, G., Coissac, E., Zundel, S., Riaz, T., Shehzad, W., Bessière, J., Taberlet, P., & Pompanon, F. (2010). An *In silico* approach for the evaluation of DNA barcodes. *BMC Genomics*, *11*, 434. https://doi.org/10.1186/1471-2164-11-434

Ficetola, G. F., Poulenard, J., Sabatier, P., Messager, E., Gielly, L., Leloup, A., Etienne, D., Bakke, J., Malet, E., Fanget, B., Støren, E., Reyss, J.-L., Taberlet, P., & Arnaud, F. (2018). DNA from lake sediments reveals long-term ecosystem changes after a biological invasion. *Science. Advances*, *4*, eaar4292. https://doi.org/10.1126/sciadv.aar4292

Fick, S. E., & Hijmans, R. J. (2017). WorldClim 2: new 1-km spatial resolution climate surfaces for global land areas. *International Journal of Climatology*, *37*(12), 4302–4315. https://doi.org/10.1002/joc.5086

Freeman, C. L., Dieudonné, L., Collins, M. J., & Sand, K. K. (2020). Survival of environmental DNA in natural environments: Surface charge and topography of minerals as driver for DNA storage. *BioRxiv*, 922997. https://doi.org/10.1101/2020.01.28.922997

Giguet-Covex, C., Ficetola, G. F., Walsh, K., Poulenard, J., Bajard, M., Fouinat, L., Sabatier, P., Gielly, L., Messager, E., Develle, A. L., David, F., Taberlet, P., Brisset, E., Guiter, F., Sinet, R., & Arnaud, F. (2019). New insights on lake sediment DNA from the catchment: importance of taphonomic and analytical issues on the record quality. *Scientific Reports*, *9*, 14676. https://doi.org/10.1038/s41598-019-50339-1

Giguet-Covex, C., Pansu, J., Arnaud, F., Rey, P.-J., Griggo, C., Gielly, L., Domaizon, I., Coissac, E., David, F., Choler, P., Poulenard, J., & Taberlet, P. (2014). Long livestock farming history and human landscape shaping revealed by lake sediment DNA. *Nature Communications*, *5*, 3211. https://doi.org/10.1038/ncomms4211

Gillman, G. P., & Bell, L. C. (1978). Soil solution studies on weathered soils from tropical North Queensland. *Australian Journal of Soil Research*, *16*(1), 67–77. https://doi.org/10.1071/SR9780067

Glenn, T. C. (2011). Field guide to next-generation DNA sequencers. *Molecular Ecology Resources*, *11*(5), 759–769. https://doi.org/10.1111/j.1755-0998.2011.03024.x

Greaves, M. P., & Wilson, M. J. (1969). The adsorption of nucleic acids by montmorillonite. *Soil Biology and Biochemistry*, *1*, 317–323. https://doi.org/10.1016/0038-0717(69)90014-5

Griffin, G. P., & Jurinak, J. (1973). Estimation of activity coefficient from the electrical conductivity of natural aquatic system and soil extracts. *Soil Science*, *116*(1), 26–30. https://doi.org/10.1097/00010694-197307000-00005

Gugerli, F., Parducci, L., & Petit, R. J. (2005). Ancient plant DNA: review and prospects. *New Phytologist*, *166*(2), 409–418. https://doi.org/10.1111/j.1469-8137.2005.01360.x

Heintzman, P. D., Elias, S. A., Moore, K., Paszkiewicz, K., & Barnes, I. (2014). Characterizing DNA preservation in degraded specimens of *Amara alpina* (Carabidae: Coleoptera). *Molecular Ecology Resources*, *14*(3), 606–615. https://doi.org/10.1111/1755-0998.12205

Hijmans, R. J., Phillips, S., Leathwick, J., & Elith, J. (2017). *dismo*: Species distribution modeling. R package version 1.1-4. Available at: http://cran.r-project.org/package=dismo

Hofreiter, M., Paijmans, J. L. A., Goodchild, H., Speller, C. F., Barlow, A., Fortes, G. G., Thomas, J. A., Ludwig, A., & Collins, M. J. (2014). The future of ancient DNA: Technical advances and conceptual shifts. *BioEssays*, *37*(3), 284–293. https://doi.org/10.1002/bies.201400160

Hofreiter, M., Serre, D., Poinar, H. N., Kuch, M., & Pääbo, S. (2001). Ancient DNA. *Nature Reviews Genetics*, *2*, 353–359. https://doi.org/10.1038/35072071

Hou, X. (2001). *Vegetation Altas of China*. Science Press.

Jia, W. (2020). Modern-process studies of plant DNA in lake sediments, Qinghai-Tibetan Plateau and arid northwestern China. Beijing: Capital Normal University. (in Chinese with English abstract).

Jónsson, H., Ginolhac, A., Schubert, M., Johnson, P. L. F., & Orlando, L. (2013). mapDamage2.0: fast approximate Bayesian estimates of ancient DNA damage parameters. *Bioinformatics*, *29*(13), 1682–1684. https://doi.org/10.1093/bioinformatics/btt193

Kanbar, H. J., Olajos, F., Englund, G., & Holmboe, M. (2020). Geochemical identification of potential DNA-hotspots and DNA-infrared fingerprints in lake sediments. *Applied Geochemistry*, *122*, 104728. https://doi.org/10.1016/j.apgeochem.2020.104728

Kang, W., Anslan, S., Börner, N., Schwarz, A., Schmidt, R., Künzel, S., Rioual, P., Echeverría-Galindo, P., Vences, M., Wang, J., & Schwalb, A. (2021). Diatom metabarcoding and microscopic analyses from sediment samples at Lake Nam Co, Tibet: The effect of sample-size and bioinformatics on the identified communities. *Ecological Indicators*, *121*, 107070. https://doi.org/10.1016/j.ecolind.2020.107070

Kanz, C., Aldebert, P., Althorpe, N., Baker, W., Baldwin, A., Bates, K., & Apweiler, R. (2005). The EMBL nucleotide sequence database. *Nucleic Acids Research*, *33*, D29–D33. https://doi.org/10.1093/nar/gki098

Khanna, M., & Stotzky, G. (1992). Transformation of *Bacillus subtilis* by DNA bound on montmorillonite and effect of DNase on the transforming ability of bound DNA. *Applied and Environmental Microbiology*, *58*(6), 1930–1939. https://doi.org/10.1128/AEM.58.6.1930-1939.1992

Leino, M. W., Hagenblad, J., Edqvist, J., & Strese, E. K. (2009). DNA preservation and utility of a historic seed collection. *Seed Science Research*, *19*(3), 125–135. https://doi.org/10.1017/S0960258509990055

Levy-Booth, D. J., Campbell, R. G., Gulden, R. H., Hart, M. M., Powell, J. R., Klironomos, J. N., Pauls, K. P., Swanton, C. J., Trevors, J. T., & Dunfield, K. E. (2007). Cycling of extracellular DNA in the soil environment. *Soil Biology and Biochemistry*, *39*(12), 2977–2991. https://doi.org/10.1016/j.soilbio.2007.06.020

Lewis, G. N., & Randall, M. (1921). The activity coefficient of strong electrolytes. *Journal of the American Chemical Society*, *43*(5), 1112–1154. https://doi.org/10.1021/ja01438a014

Lindahl, T. (1993). Instability and decay of the primary structure of DNA. *Nature*, *362*, 709–715. https://doi.org/10.1038/362709a0

Lindahl, T., & Nyberg, B. (1972). Rate of depurination of native deoxyribonucleic acid. *Biochemistry*, *11*(19), 3610–3618. https://doi.org/10.1021/bi00769a018

Liu, S., Kruse, S., Scherler, D., Ree, R. H., Zimmermann, H. H., Stoof-Leichsenring, K. R., Epp, L. S., Mischke, S., & Herzschuh, U. (2021). Sedimentary ancient DNA reveals a threat of warming-induced alpine habitat loss to Tibetan Plateau plant diversity. *Nature Communications*, *12*, 2995. https://doi.org/10.1038/s41467-021-22986-4

Lorenz, M. G., & Wackernagel, W. (1987). Adsorption of DNA to sand and variable degradation rates of adsorbed DNA. *Applied and Environmental Microbiology*, *53*(12), 2948–2952. https://doi.org/10.1128/AEM.53.12.2948-2952.1987

Matheson, C. D., Gurney, C., Esau, N., & Lehto, R. (2010). Assessing PCR inhibition from humic substances. *The Open Enzyme Inhibition Journal*, *3*, 38–45. https://doi.org/10.2174/1874940201003010038

Mathieu, C., Hermans, S. M., Lear, G., Buckley, T. R., Lee, K. C., & Buckley, H. L. (2020). A systematic review of sources of variability and uncertainty in eDNA data for environmental monitoring. *Frontiers in Ecology and Evolution*, *8*, 135. https://doi.org/10.3389/fevo.2020.00135

Metzker, M. L. (2010). Sequencing technologies – The next generation. *Nature Reviews Genetics*, *11*, 31–46. https://doi.org/10.1038/nrg2626

Mirbach, S., & Lang, U. (2018). Density-driven underflows with suspended solids in Lake Constance. *Journal of Soils and Sediments*, *18*(10), 3145–3152. https://doi.org/10.1007/s11368-017-1753-x

Morley, S. A., & Nielsen, B. L. (2016). Chloroplast DNA copy number changes during plant development in organelle DNA polymerase mutants. *Frontiers in Plant Science*, *7*, 57. https://doi.org/10.3389/fpls.2016.00057

Nagler, M., Insam, H., Pietramellara, G., & Ascher-Jenull, J. (2018). Extracellular DNA in natural environments: features, relevance and applications. *Applied Microbiology and Biotechnology*, *102*(15), 6343–6356. https://doi.org/10.1007/s00253-018-9120-4

Nichols, R. V., Vollmers, C., Newsom, L. A., Wang, Y., Heintzman, P. D., Leighton, M. K., Green, R. E., & Shapiro, B. (2018). Minimizing polymerase biases in metabarcoding. *Molecular Ecology Resources*, *18*(5), 927–939. https://doi.org/10.1111/1755-0998.12895

Nielsen, K. M., Johnsen, P. J., Bensasson, D., & Daffonchio, D. (2007). Release and persistence of extracellular DNA in the environment. *Environmental Biosafety Research*, *6*(1–2), 37–53. https://doi.org/10.1051/ebr:2007031

Niemeyer, B., Epp, L. S., Stoof-Leichsenring, K. R., Pestryakova, L. A., & Herzschuh, U. (2017). A comparison of sedimentary DNA and pollen from lake sediments in recording vegetation composition at the Siberian treeline. *Molecular Ecology Resources*, *17*(6), e46–e62. https://doi.org/10.1111/1755-0998.12689

Ogram, A., Sayler, G. S., Gustin, D., & Lewis, R. J. (1988). DNA adsorption to soils and sediments. *Environmental Science and Technology*, *22*(8), 982–984. https://doi.org/10.1021/es00173a020

Paget, E., Monrozier, L. J., & Simonet, P. (1992). Adsorption of DNA on clay minerals: protection against DNaseI and influence on gene transfer. *FEMS Microbiology Letters*, *97*(1–2), 31–39. https://doi.org/10.1016/0378-1097(92)90359-V

Pansu, J., Giguet-Covex, C., Ficetola, G. F., Gielly, L., Boyer, F., Zinger, L., Arnaud, F., Poulenard, J., Taberlet, P., & Choler, P. (2015). Reconstructing long-term human impacts on plant communities: an ecological approach based on lake sediment DNA. *Molecular Ecology*, *24*(7), 1485–1498. https://doi.org/10.1111/mec.13136

Parducci, L., Alsos, I. G., Unneberg, P., Pedersen, M. W., Han, L. U., Lammers, Y., Salonen, J. S., Väliranta, M. M., Slotte, T., & Wohlfarth, B. (2019). Shotgun environmental DNA, pollen, and macrofossil analysis of Lateglacial lake sediments from southern Sweden. *Frontiers in Ecology and Evolution*, *7*, 189. https://doi.org/10.3389/fevo.2019.00189

Parducci, L., Bennett, K. D., Ficetola, G. F., Alsos, I. G., Suyama, Y., Wood, J. R., & Pedersen, M. W. (2017). Ancient plant DNA in lake sediments. *New Phytologist*, *214*, 924–942. https://doi.org/10.1111/nph.14470

Parducci, L., Jørgensen, T., Tollefsrud, M. M., Elverland, E., Alm, T., Fontana, S. L., Bennett, K. D., Haile, J., Matetovici, I., Suyama, Y., Edwards, M. E., Andersen, K., Rasmussen, M., Boessenkool, S., Coissac, E., Brochmann, C., Taberlet, P., Houmark-Nielsen, M., Larsen, N. K., … Willerslev, E. (2012). Glacial survival of boreal trees in northern Scandinavia. *Science*, *335*(6072), 1083–1086. https://doi.org/10.1126/science.1216043

Parducci, L., Nota, K., & Wood, J. (2019). Reconstructing past vegetation communities using ancient DNA from lake sediments. In C. Lindqvist, & O. P. Rajora (Eds.), *Paleogenomics: Genome-scale Analysis of Ancient DNA* (pp. 163–187). Springer Nature Switzerland. https://doi.org/10.1007/13836_2018_38

Pedersen, M. W., Overballe-Petersen, S., Ermini, L., Sarkissian, C. D., Haile, J., Hellstrom, M., Spens, J., Thomsen, P. F., Bohmann, K., Cappellini, E., Schnell, I. B., Wales, N. A., Carøe, C., Campos, P. F., Schmidt, A. M. Z., Gilbert, M. T. P., Hansen, A. J., Orlando, L., & Willerslev, E. (2015). Ancient and modern environmental DNA. *Philosophical Transactions of the Royal Society B-Biological Sciences*, *370*, 20130383. https://doi.org/10.1098/rstb.2013.0383

Pedersen, M. W., Ruter, A., Schweger, C., Friebe, H., Staff, R. A., Kjeldsen, K. K., Mendoza, M. L. Z., Beaudoin, A. B., Zutter, C., Larsen, N. K., Potter, B. A., Nielsen, R., Rainville, R. A., Orlando, L., Meltzer, D. J., Kjær, K. H., & Willerslev, E. (2016). Postglacial viability and colonization in North America's ice-free corridor. *Nature*, *537*, 45–49. https://doi.org/10.1038/nature19085

Pedreira-Segade, U., Hao, J., Razafitianamaharavo, A., Pelletier, M., Marry, V., Le Crom, S., Michot, L., & Daniel, I. (2018). How do nucleotides adsorb onto clays? *Life*, *8*, 59. https://doi.org/10.3390/life8040059

Pietramellara, G., Ascher, J., Borgogni, F., Ceccherini, M. T., Guerri, G., & Nannipieri, P. (2009). Extracellular DNA in soil and sediment: fate and ecological relevance. *Biology and Fertility of Soils*, *45*, 219–235. https://doi.org/10.1007/s00374-008-0345-8

Pietramellara, G., Franchi, M., Gallori, E., & Nannipieri, P. (2001). Effect of molecular characteristics of DNA on its adsorption and binding on homoionic montmorillonite and kaolinite. *Biology and Fertility of Soils*, *33*, 402–409. https://doi.org/10.1007/s003740100341

Piñol, J., Mir, G., Gomez-Polo, P., & Agustí, N. (2015). Universal and blocking primer mismatches limit the use of high-throughput DNA sequencing for the quantitative metabarcoding of arthropods. *Molecular Ecology Resources*, *15*(4), 819–830. https://doi.org/10.1111/1755-0998.12355

Ponnamperuma, F. N., Tianco, E. M., & Loy, T. A. (1966). Ionic strengths of solutions of flooded soils and other natural aqueous solutions from specific conductance. *Soil Science*, *102*(6), 408–413. https://doi.org/10.1097/00010694-196612000-00009

R Core Team (2019). *R: A language and environment for statistical computing*. R Foundation for Statistical Computing. Available at: http://www.r-project.org

Ravanat, J., Douki, T., & Cadet, J. (2001). Direct and indirect effects of UV radiation on DNA and its components. *Journal of Photochemistry and Photobiology B: Biology*, *63*(1–3), 88–102. https://doi.org/10.1016/S1011-1344(01)00206-8

Revelle, W. (2021). *psych*: Procedures for psychological, psychometric, and personality research. R package version 2.1.6. Available at: http://cran.r-project.org/package=psych

Rijal, D. P., Heintzman, P. D., Lammers, Y., Yoccoz, N. G., Lorberau, K. E., Pitelkova, I., Goslar, T., Murguzur, F. J. A., Salonen, J. S., Helmens, K. F., Bakke, J., Edwards, M. E., Alm, T., Bråthen, K. A., Brown, A. G., & Alsos, I. G. (2021). Sedimentary ancient DNA shows terrestrial plant richness continuously increased over the Holocene in northern Fennoscandia. *Science. Advances*, *7*, eabf9557. https://doi.org/10.1126/sciadv.abf9557

Romanowski, G., Lorenz, M. G., & Wackernagel, W. (1991). Adsorption of plasmid DNA to mineral surfaces and protection against DNase I. *Applied and Environmental Microbiology*, *57*(4), 1057–1061. https://doi.org/10.1128/AEM.57.4.1057-1061.1991

RStudio Team (2015). *RStudio: Integrated development environment for R*. RStudio. Available at: http://www.rstudio.org

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. https://doi.org/10.1016/j.gecco.2019.e00547

Sakamoto, W., & Takami, T. (2018). Chloroplast DNA dynamics: Copy number, quality control and degradation. *Plant and Cell Physiology*, *59*(6), 1120–1127. https://doi.org/10.1093/pcp/pcy084

Seutin, G., White, B. N., & Boag, P. T. (1991). Preservation of avian blood and tissue samples for DNA analyses. *Canadian Journal of Zoology*, *69*(1), 82–90. https://doi.org/10.1139/z91-013

Seymour, M., Durance, I., Cosby, B. J., Ransom-Jones, E., Deiner, K., Ormerod, S. J., Colbourne, J. K., Wilgar, G., Carvalho, G. R., de Bruyn, M., Edwards, F., Emmett, B. A., Bik, H. M., & Creer, S. (2018). Acidity promotes degradation of multi-species environmental

DNA in lotic mesocosms. *Communications Biology*, *1*, 4. https://doi.org/10.1038/s42003-017-0005-3

Shendure, J., & Ji, H. L. (2008). Next-generation DNA sequencing. *Nature Biotechnology*, *26*, 1135–1145. https://doi.org/10.1038/nbt1486

Sheng, X., Qin, C., Yang, B., Hu, X., Liu, C., Waigi, M. G., Li, X., & Ling, W. (2019). Metal cation saturation on montmorillonites facilitates the adsorption of DNA via cation bridging. *Chemosphere*, *235*, 670–678. https://doi.org/10.1016/j.chemosphere.2019.06.159

Sjögren, P., Edwards, M. E., Gielly, L., Langdon, C. T., Croudace, I. W., Merkel, M. K. F., & Alsos, I. G. (2017). Lake sedimentary DNA accurately records 20th Century introductions of exotic conifers in Scotland. *New Phytologist*, *213*(2), 929–941. https://doi.org/10.1111/nph.14199

Slon, V., Hopfe, C., Weiß, C. L., Mafessoni, F., de la Rasilla, M., Lalueza-Fox, C., Rosas, A., Soressi, M., Knul, M. V., Miller, R., Stewart, J. R., Derevianko, A. P., Jacobs, Z., Li, B. O., Roberts, R. G., Shunkov, M. V., de Lumley, H., Perrenoud, C., Gušić, I., … Meyer, M. (2017). Neandertal and Denisovan DNA from Pleistocene sediments. *Science*, *356*(6338), 605–608. https://doi.org/10.1126/science.aam9695

Soininen, E. M., Gauthier, G., Bilodeau, F., Berteaux, D., Gielly, L., Taberlet, P., Gussarova, G., Bellemain, E., Hassel, K., Stenøien, H. K., Epp, L., Schrøder-Nielsen, A., Brochmann, C., & Yoccoz, N. G. (2015). Highly overlapping winter diet in two sympatric lemming species revealed by DNA metabarcoding. *PLoS ONE*, *10*(1), e0115335. https://doi.org/10.1371/journal.pone.0115335

Sønstebø, J. H., Gielly, L., Brysting, A. K., Elven, R., Edwards, M., Haile, J., Willerslev, E., Coissac, E., Rioux, D., Sannier, J., Taberlet, P., & Brochmann, C. (2010). Using next-generation sequencing for molecular reconstruction of past Arctic vegetation and climate. *Molecular Ecology Resources*, *10*(6), 1009–1018. https://doi.org/10.1111/j.1755-0998.2010.02855.x

Stone, T. A., & Schlesinger, P. (2003). *Vegetative cover of the Former Soviet Union, 1990*. Oak Ridge National Laboratory Distributed Active Archive Center. Available at: http://www.daac.ornl.gov

Stoof-Leichsenring, K. R., Epp, L. S., Trauth, M. H., & Tiedemann, R. (2012). Hidden diversity in diatoms of Kenyan Lake Naivasha: a genetic approach detects temporal variation. *Molecular Ecology*, *21*(8), 1918–1930. https://doi.org/10.1111/j.1365-294X.2011.05412.x

Stoof-Leichsenring, K., Liu, S., Jia, W., Li, K., Pestryakova, L., Mischke, S., Cao, X., Liu, X., Ni, J., Neuhaus, S., & Herzschuh, U. (2020). Plant diversity in sedimentary DNA obtained from high-latitude (Siberia) and high-elevation lakes (China). *Biodiversity Data Journal*, *8*, e57089. https://doi.org/10.3897/BDJ.8.e57089

Strickler, K. M., Fremier, A. K., & Goldberg, C. S. (2015). Quantifying effects of UV-B, temperature, and pH on eDNA degradation in aquatic microcosms. *Biological Conservation*, *183*, 85–92. https://doi.org/10.1016/j.biocon.2014.11.038

Tabares, X., Zimmermann, H., Dietze, E., Ratzmann, G., Belz, L., Vieth-Hillebrand, A., Dupont, L., Wilkes, H., Mapani, B., & Herzschuh, U. (2020). Vegetation state changes in the course of shrub encroachment in an African savanna since about 1850 CE and their potential drivers. *Ecology and Evolution*, *10*(2), 962–979. https://doi.org/10.1002/ece3.5955

Taberlet, P., Bonin, A., Zinger, L., & Coissac, E. (2018). *Environmental DNA: For Biodiversity Research and Monitoring*. Oxford University Press. https://doi.org/10.1093/oso/9780198767220.001.0001

Taberlet, P., Coissac, E., Pompanon, F., Brochmann, C., & Willerslev, E. (2012). Towards next-generation biodiversity assessment using DNA metabarcoding. *Molecular Ecology*, *21*(8), 2045–2050. https://doi.org/10.1111/j.1365-294X.2012.05470.x

Taberlet, P., Coissac, E., Pompanon, F., Gielly, L., Miquel, C., Valentini, A., Vermat, T., Corthier, G., Brochmann, C., & Willerslev, E. (2007). Power and limitations of the chloroplast *trn*L (UAA) intron for plant DNA barcoding. *Nucleic Acids Research*, *35*(3), e14. https://doi.org/10.1093/nar/gkl938

Taberlet, P., Griffin, S., Goossens, B., Questiau, S., Manceau, V., Escaravage, N., & Bouvet, J. (1996). Reliable genotyping of samples with very low DNA quantities using PCR. *Nucleic Acids Research*, *24*(16), 3189–3194. https://doi.org/10.1093/nar/24.16.3189

Torti, A., Lever, M. A., & Jørgensen, B. B. (2015). Origin, dynamics, and implications of extracellular DNA pools in marine sediments. *Marine Genomics*, *24*, 185–196. https://doi.org/10.1016/j.margen.2015.08.007

Watson, R. J., & Blackwell, B. (2000). Purification and characterization of a common soil component which inhibits the polymerase chain reaction. *Canadian Journal of Microbiology*, *46*(7), 633–642. https://doi.org/10.1139/w00-043

Willerslev, E., & Cooper, A. (2005). Ancient DNA. *Proceedings of the Royal Society B-Biological Sciences*, *272*(1558), 3–16. https://doi.org/10.1098/rspb.2004.2813

Willerslev, E., Davison, J., Moora, M., Zobel, M., Coissac, E., Edwards, M. E., Lorenzen, E. D., Vestergård, M., Gussarova, G., Haile, J., Craine, J., Gielly, L., Boessenkool, S., Epp, L. S., Pearman, P. B., Cheddadi, R., Murray, D., Bråthen, K. A., Yoccoz, N., … Taberlet, P. (2014). Fifty thousand years of Arctic vegetation and megafaunal diet. *Nature*, *506*, 47–51. https://doi.org/10.1038/nature12921

Willerslev, E., Hansen, A. J., Binladen, J., Brand, T. B., Gilbert, M. T. P., Shapiro, B., Bunce, M., Wiuf, C., Gilichinsky, D. A., & Cooper, A. (2003). Diverse plant and animal genetic records from Holocene and Pleistocene sediments. *Science*, *300*(5620), 791–795. https://doi.org/10.1126/science.1084114

Wu, J.-Y., Jiang, X.-T., Jiang, Y.-X., Lu, S.-Y., Zou, F., & Zhou, H.-W. (2010). Effects of polymerase, template dilution and cycle number on PCR based 16S rRNA diversity analysis using the deep sequencing method. *BMC Microbiology*, *10*, 255. https://doi.org/10.1186/1471-2180-10-255

Yu, W. H., Li, N., Tong, D. S., Zhou, C. H., Lin, C. X., & Xu, C. Y. (2013). Adsorption of proteins and nucleic acids on clay minerals and their interactions: A review. *Applied Clay Science*, *80–81*, 443–452. https://doi.org/10.1016/j.clay.2013.06.003

Zhuiykov, S. (2018). Semiconductor nano-crystals in environmental sensors. In S. Zhuiykov (Ed.), *Nanostructured Semiconductors* (pp. 475–538). Woodhead Publishing. https://doi.org/10.1016/B978-0-08-101919-1.00009-X

Zinger, L., Bonin, A., Alsos, I. G., Bálint, M., Bik, H., Boyer, F., Chariton, A. A., Creer, S., Coissac, E., Deagle, B. E., De Barba, M., Dickie, I. A., Dumbrell, A. J., Ficetola, G. F., Fierer, N., Fumagalli, L., Gilbert, M. T. P., Jarman, S., Jumpponen, A., … Taberlet, P. (2019). DNA metabarcoding - Need for robust experimental designs to draw sound ecological conclusions. *Molecular Ecology*, *28*(8), 1857–1862. https://doi.org/10.1111/mec.15060

## SUPPORTING INFORMATION

Additional supporting information may be found in the online version of the article at the publisher's website.

**How to cite this article:** Jia, W., Liu, X., Stoof-Leichsenring, K. R., Liu, S., Li, K., & Herzschuh, U. (2022). Preservation of sedimentary plant DNA is related to lake water chemistry. *Environmental DNA*, 4, 425–439. https://doi.org/10.1002/edn3.259