SAPIENZA
UNIVERSITÀ DI ROMA

DNA studies from lake sediments: what we know and what we want to know

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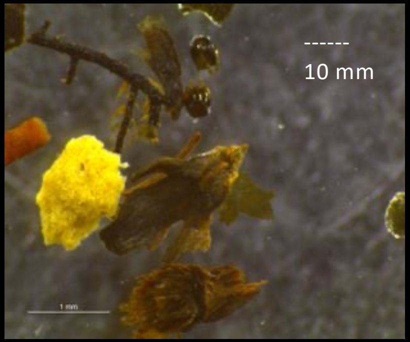
MPGD 2026: Statistical Methods for Post Genomic Data
29-30 January 2026 Grenoble (France)

Changing flora and palaeoenvironments

Investigating which plant (and animal) species were present in the past based on **fossil archives.**

Traditionally palaeoecologists look at:

- Microfossils (pollen, spores)
- Macro & megafossils.



Pollen & macrofossils (seeds, leaves, fruits, needles) retrieved from sediments.

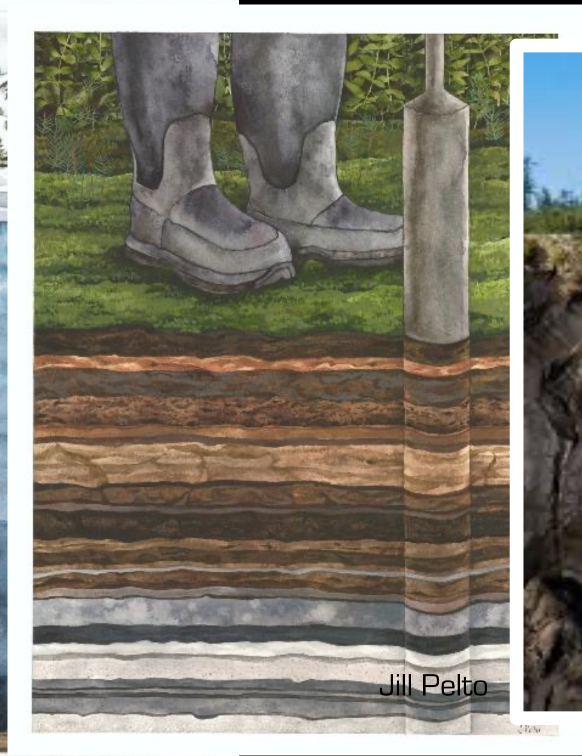
Megafossils retrieved from a small pool in Sweden: a **spruce cone** (9030 cal. yr BP), two **larch cones**, a **larch twig** (8160 cal. yr BP, composite date). Photo: Kullman, 12 July 2007.

Changing flora and palaeoenvironments

As fossil archives we use:

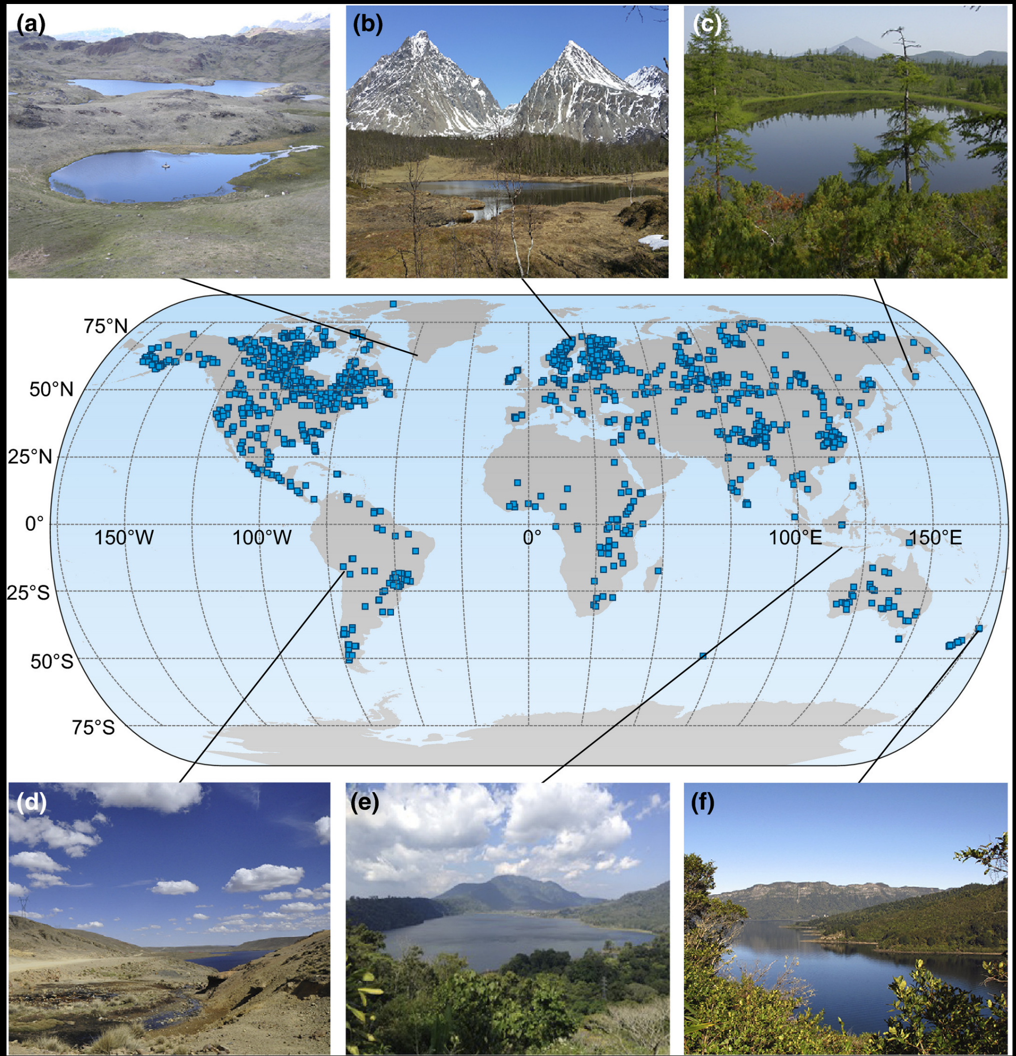
- lake sediments
- peat
- permafrost

Lake sediments



Peat sediments

Permafrost



Lake sediments

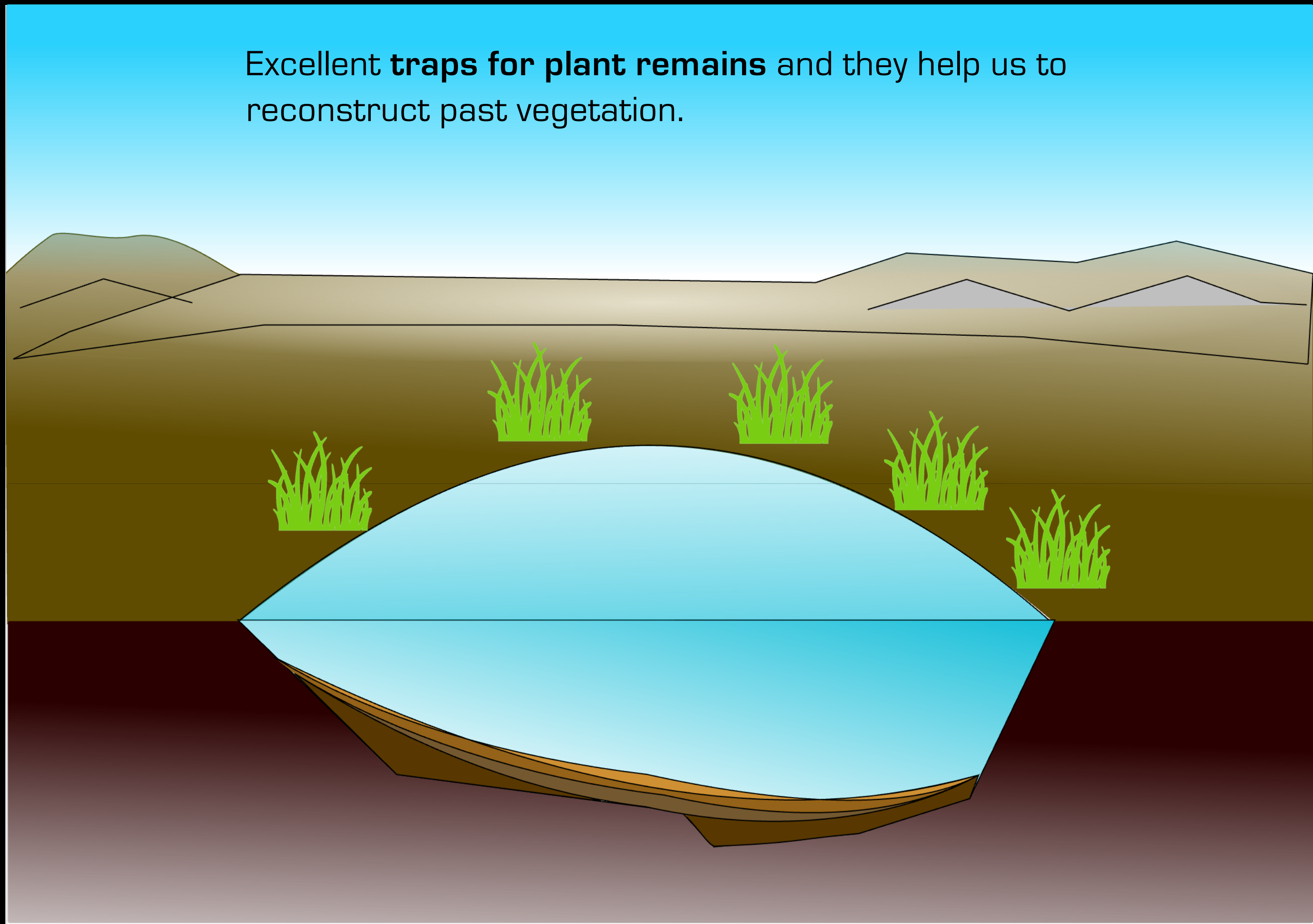
Abundant at high latitudes
Cold environments
Anoxic

Best preservation conditions for DNA

Many **small glacial lakes** which offer **good catchment areas**

Parducci *et al.* 2017 New Phyt

Lakes are natural archives

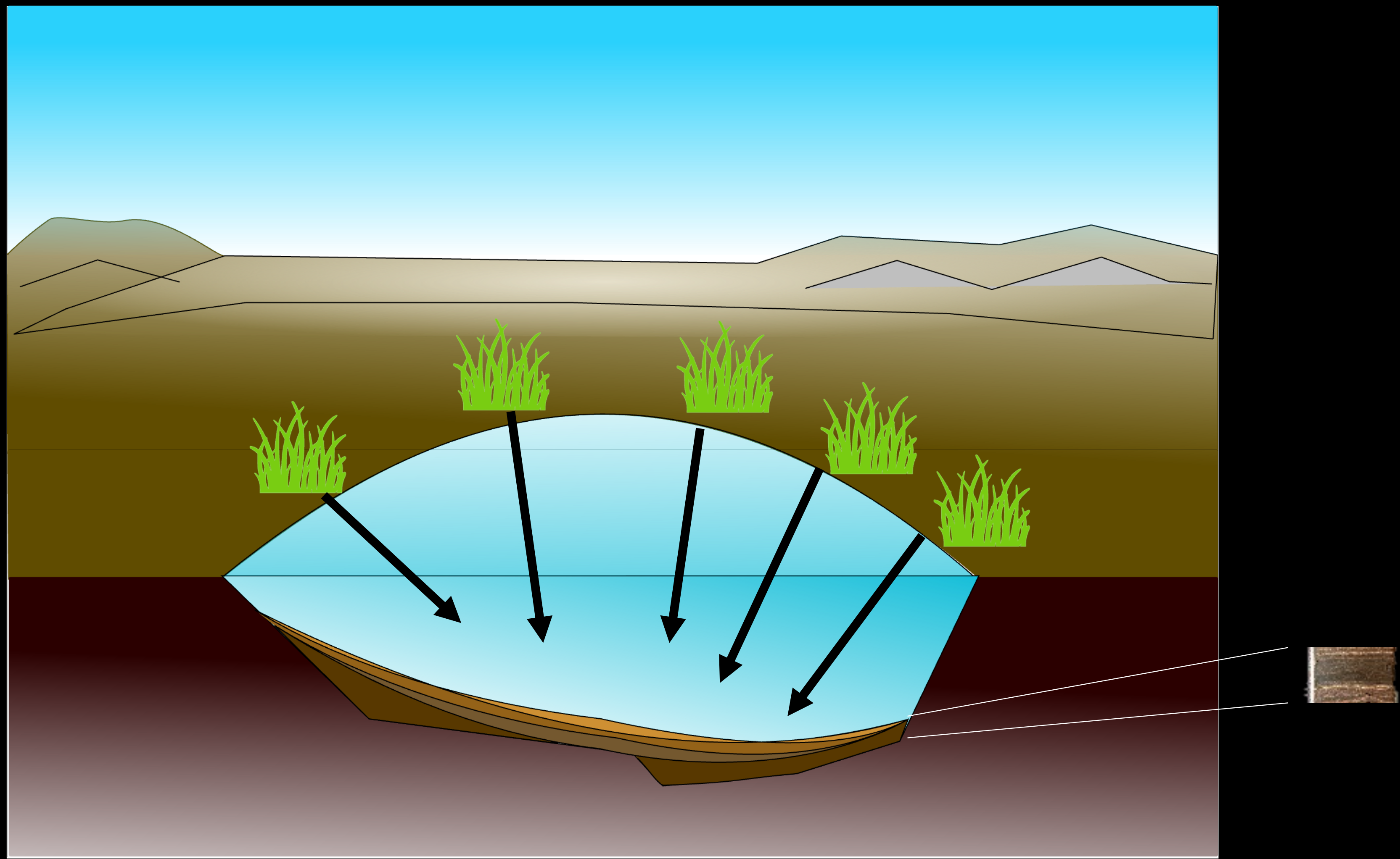


Post-glacial environment

15 -12 kr BP

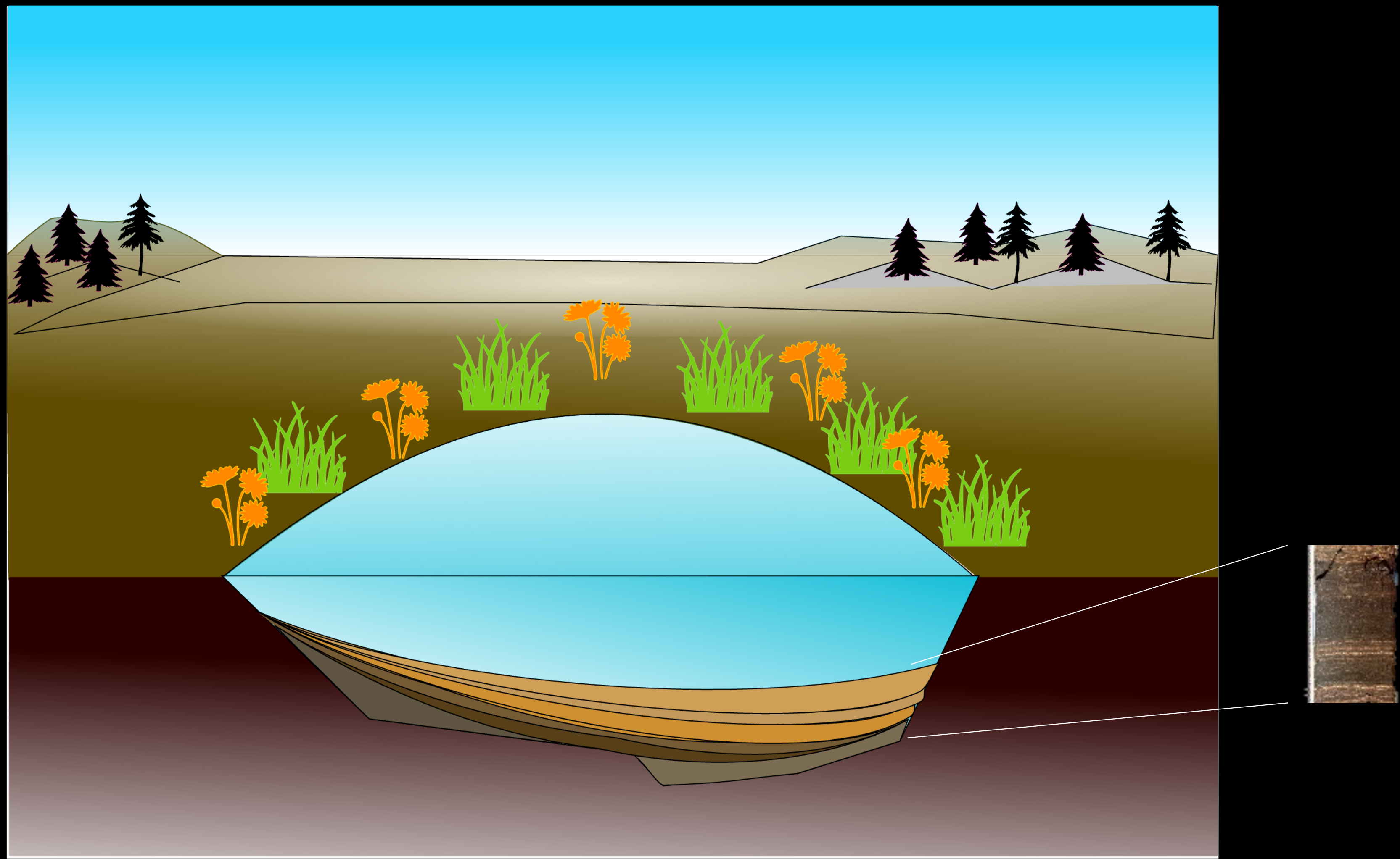
Post-glacial environment

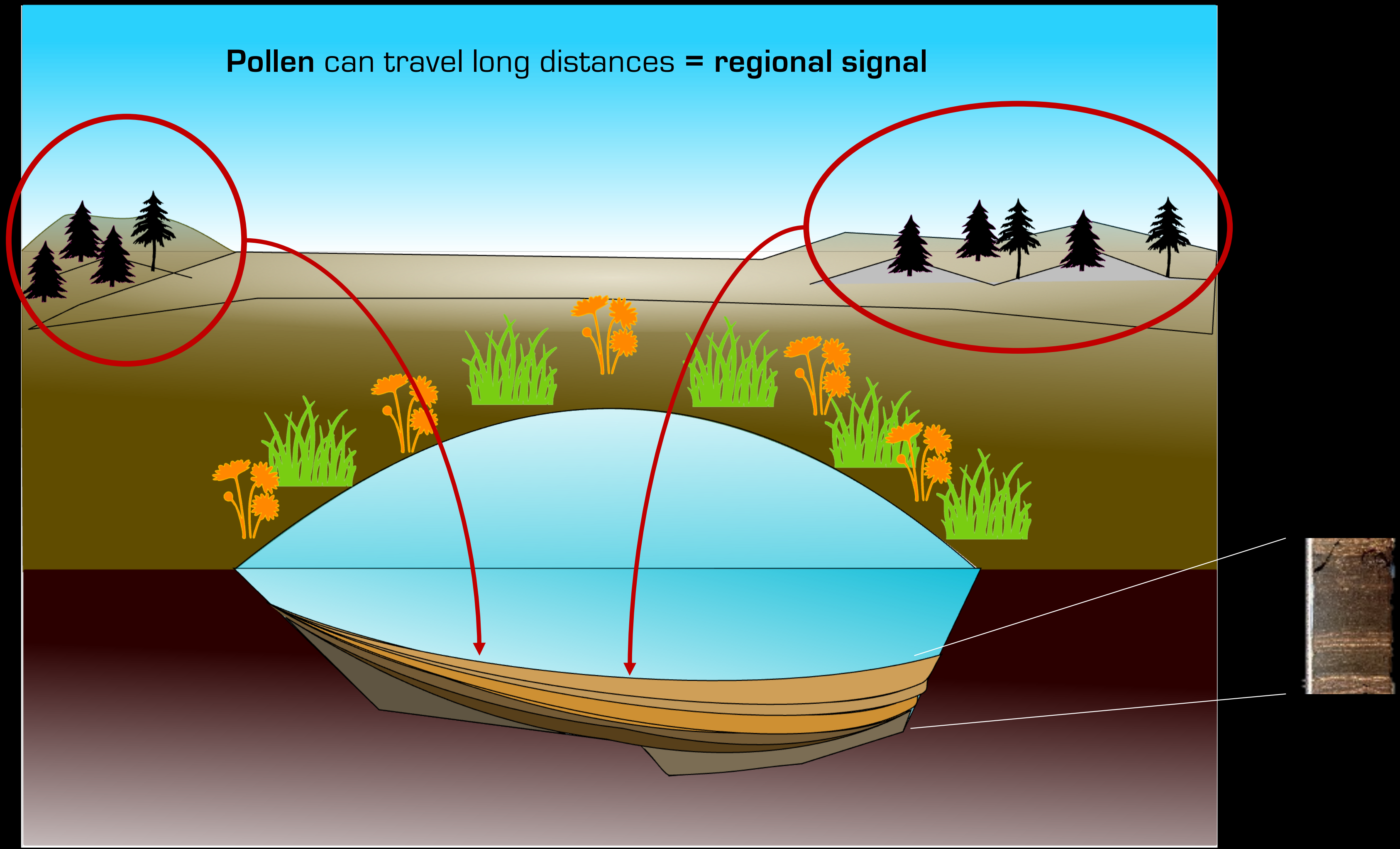
15 -12 kr BP



Tundra environment

12 -10 kr BP



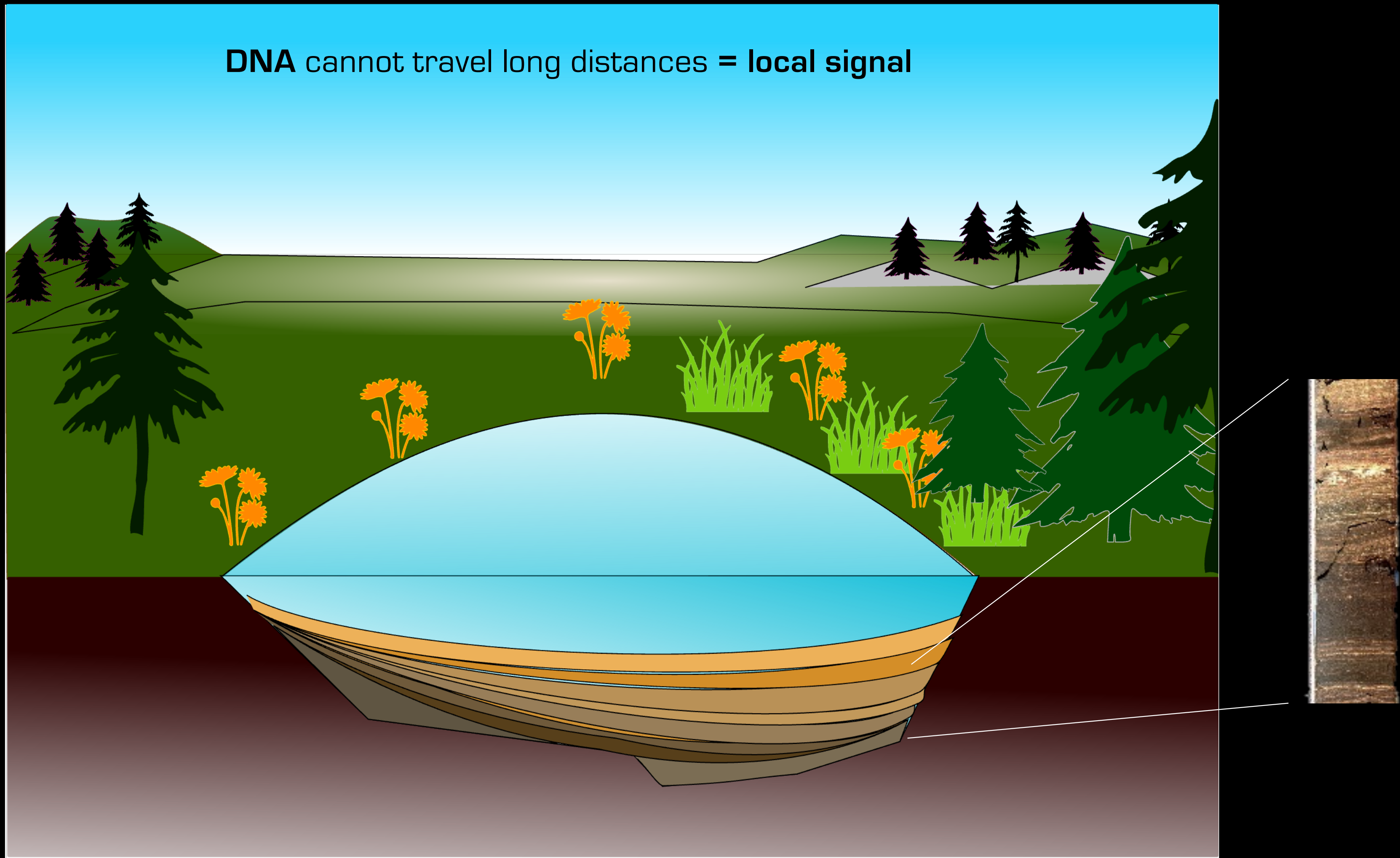


Tundra-boreal environment

10 kr BP

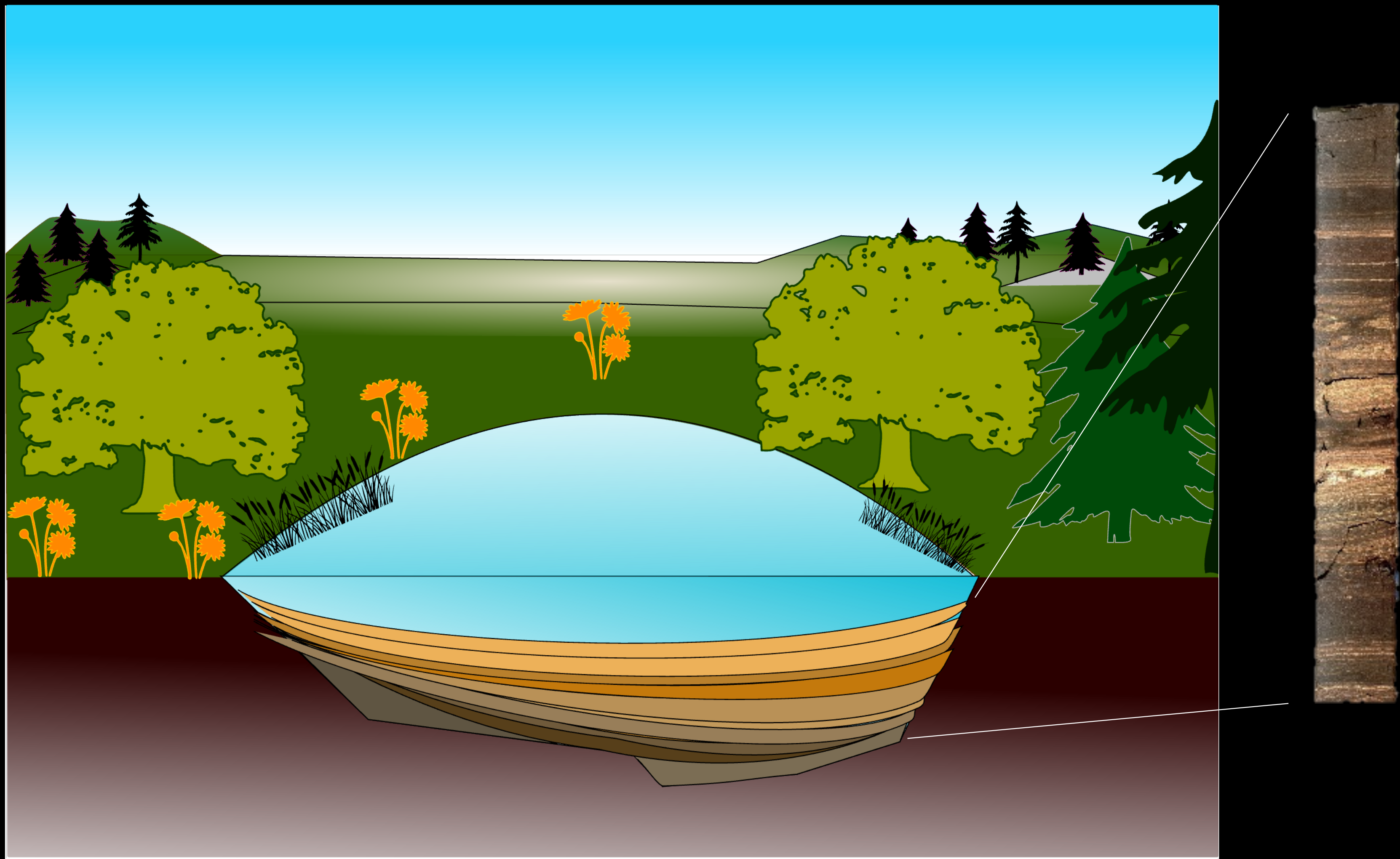
Boreal environment

10 - 8 kr BP

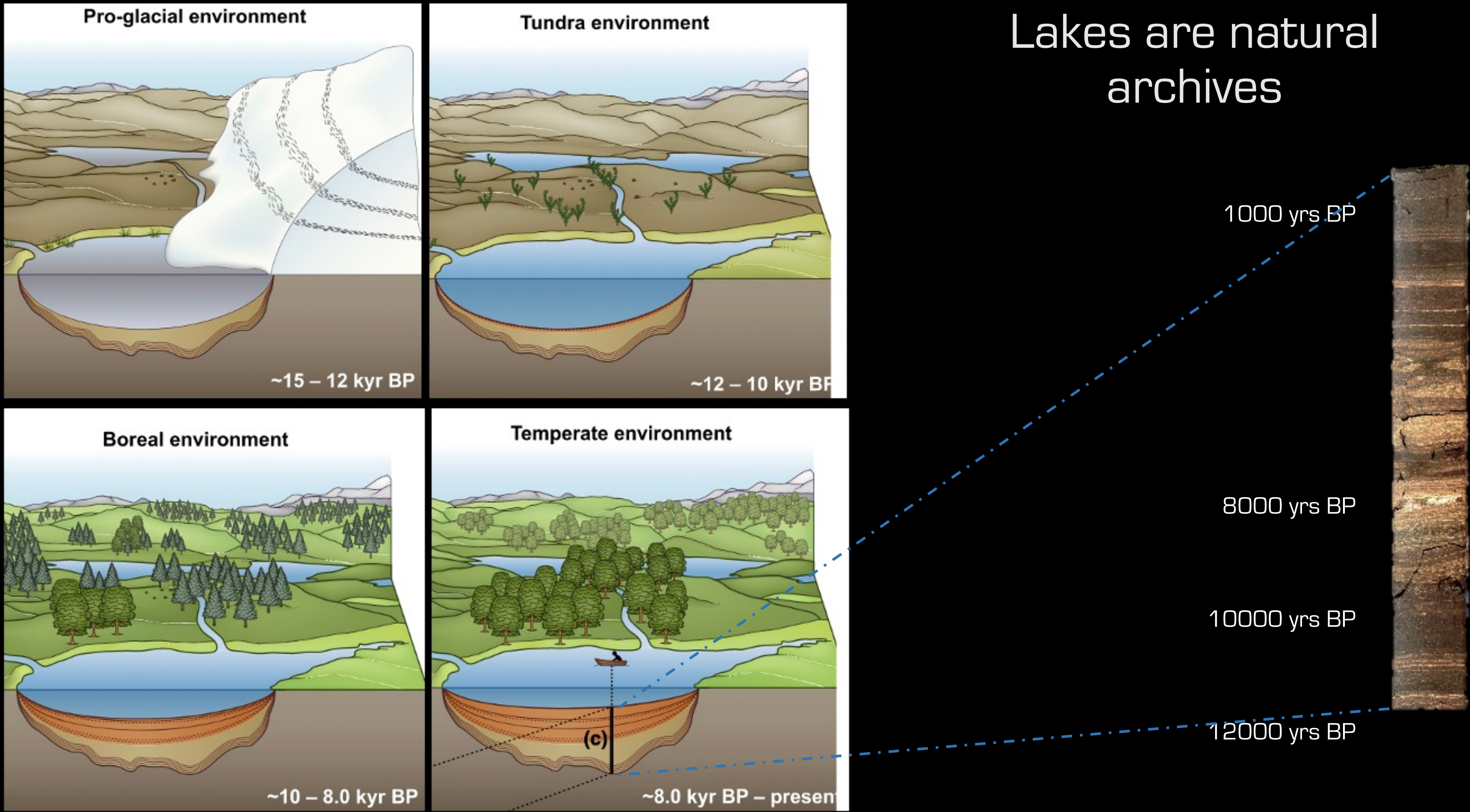


Temperate environment

8 - 4 kr BP

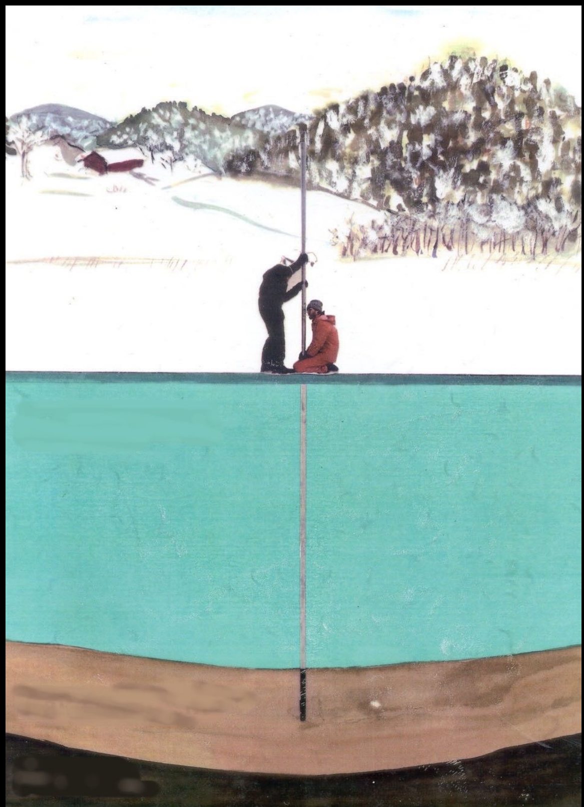


Lakes are natural archives



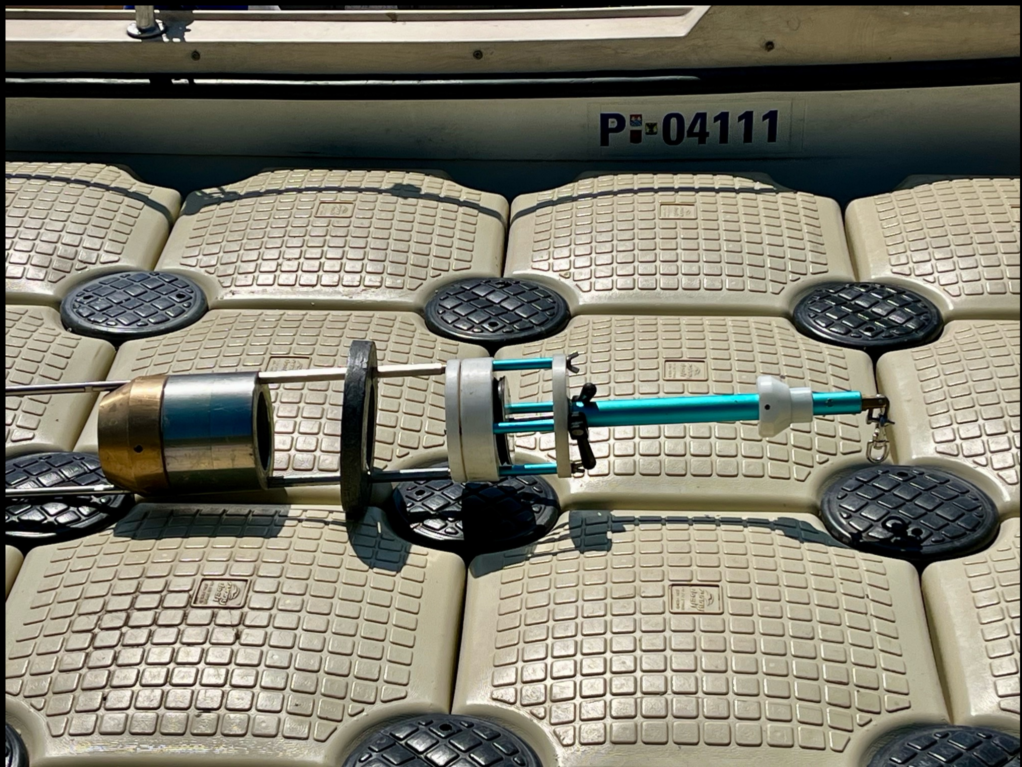
Parducci *et al.* 2017 *New Phyt*

Coring

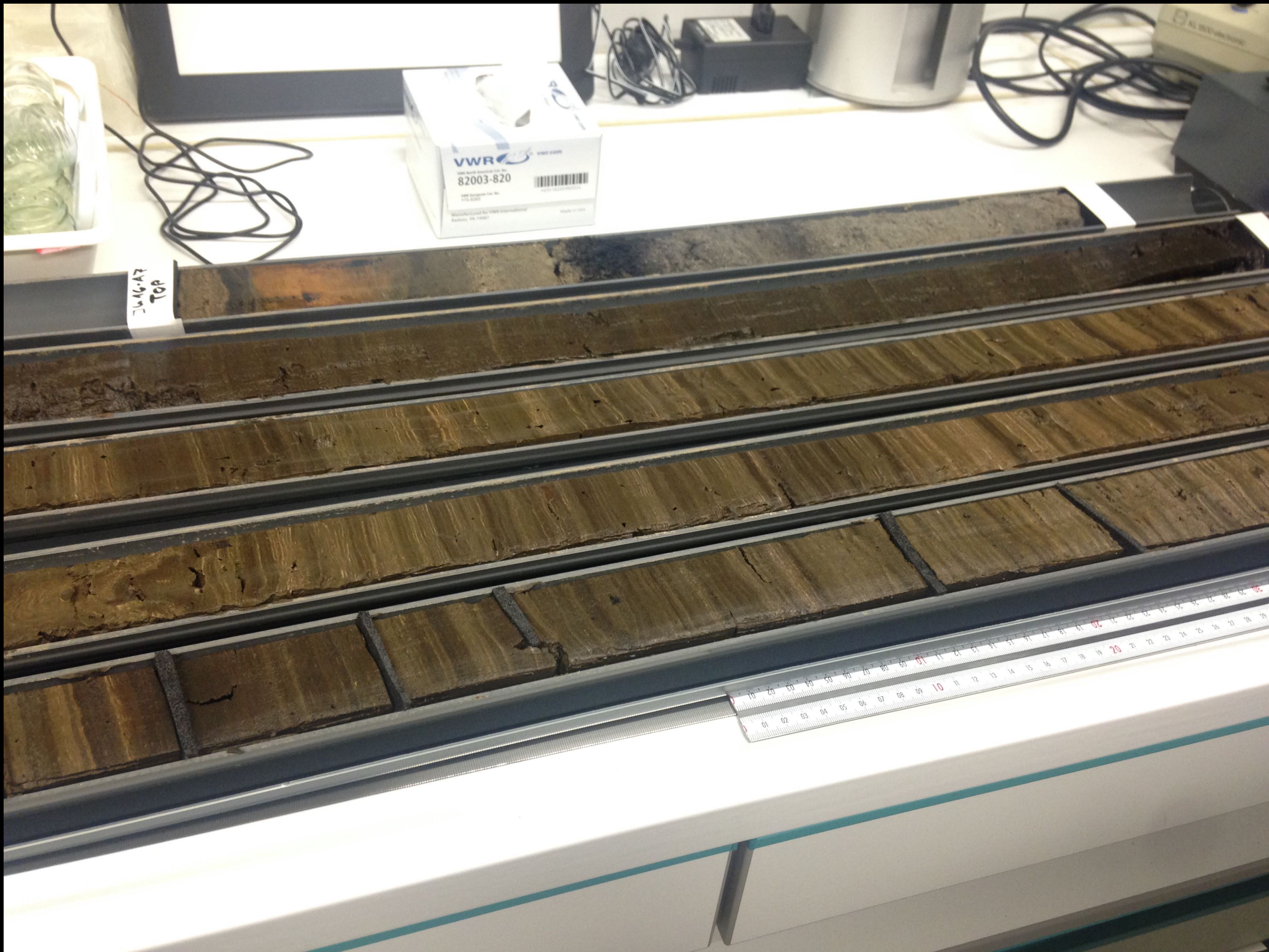


Lago di
Martignano
(central Italy)

Lago di Nemi (central Italy)



Lago di Nemi (central Italy)


VWR
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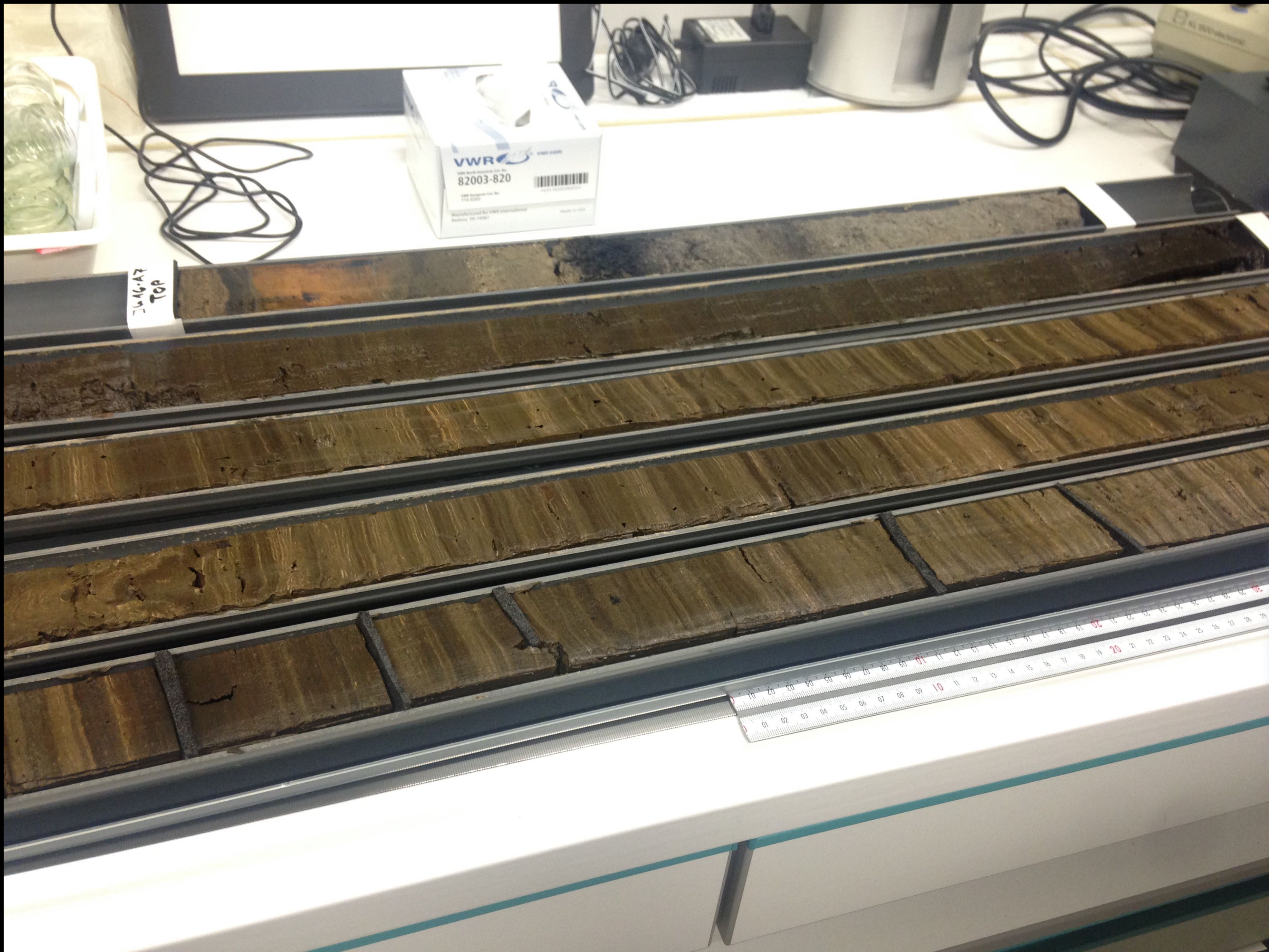


Pollen analysis

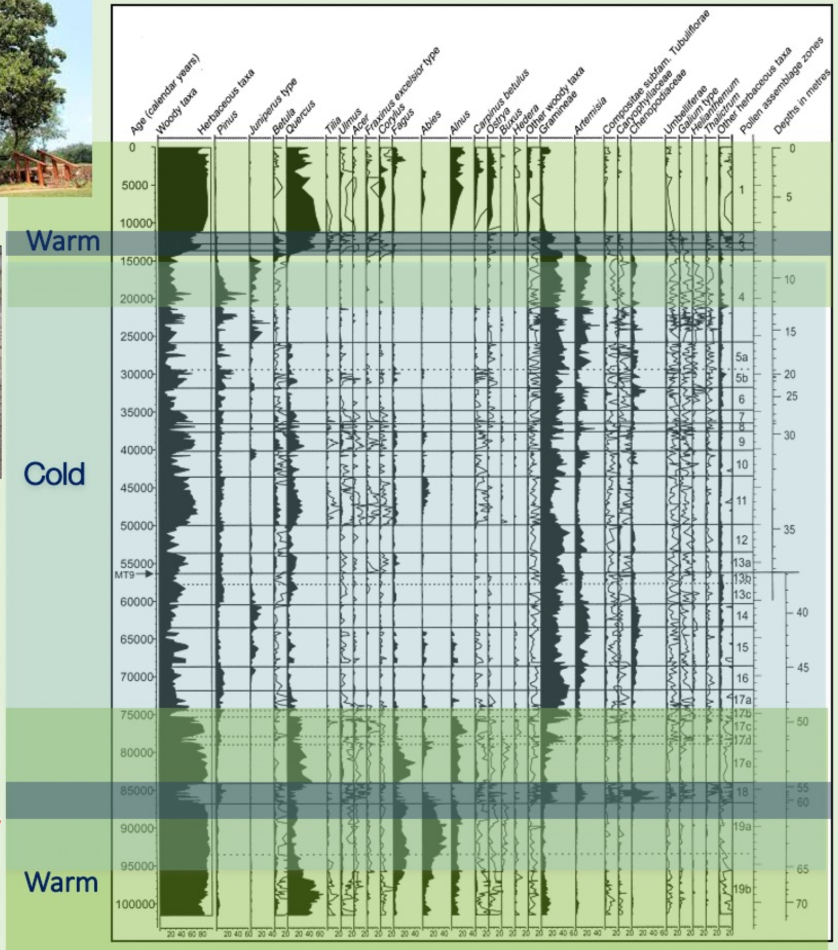
Palynology
The study of micro (pollen) and macrofossils and microscopic plankton organisms.

Pollen analysis

The last 110,000 years

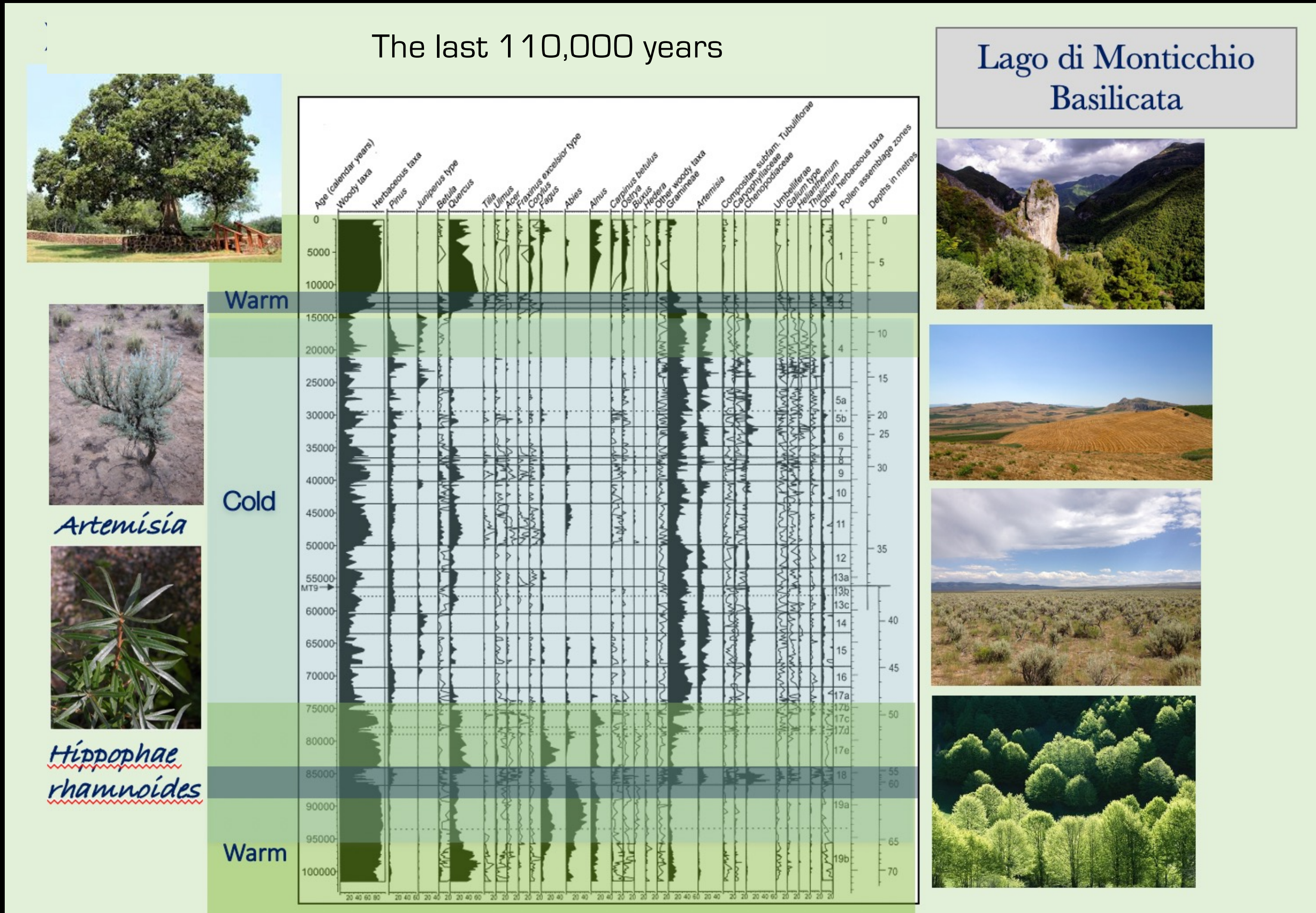
Artemisia

Hippophae rhamnoides



Lago di Monticchio Basilicata

Allen *et al.*, 1999 Nature



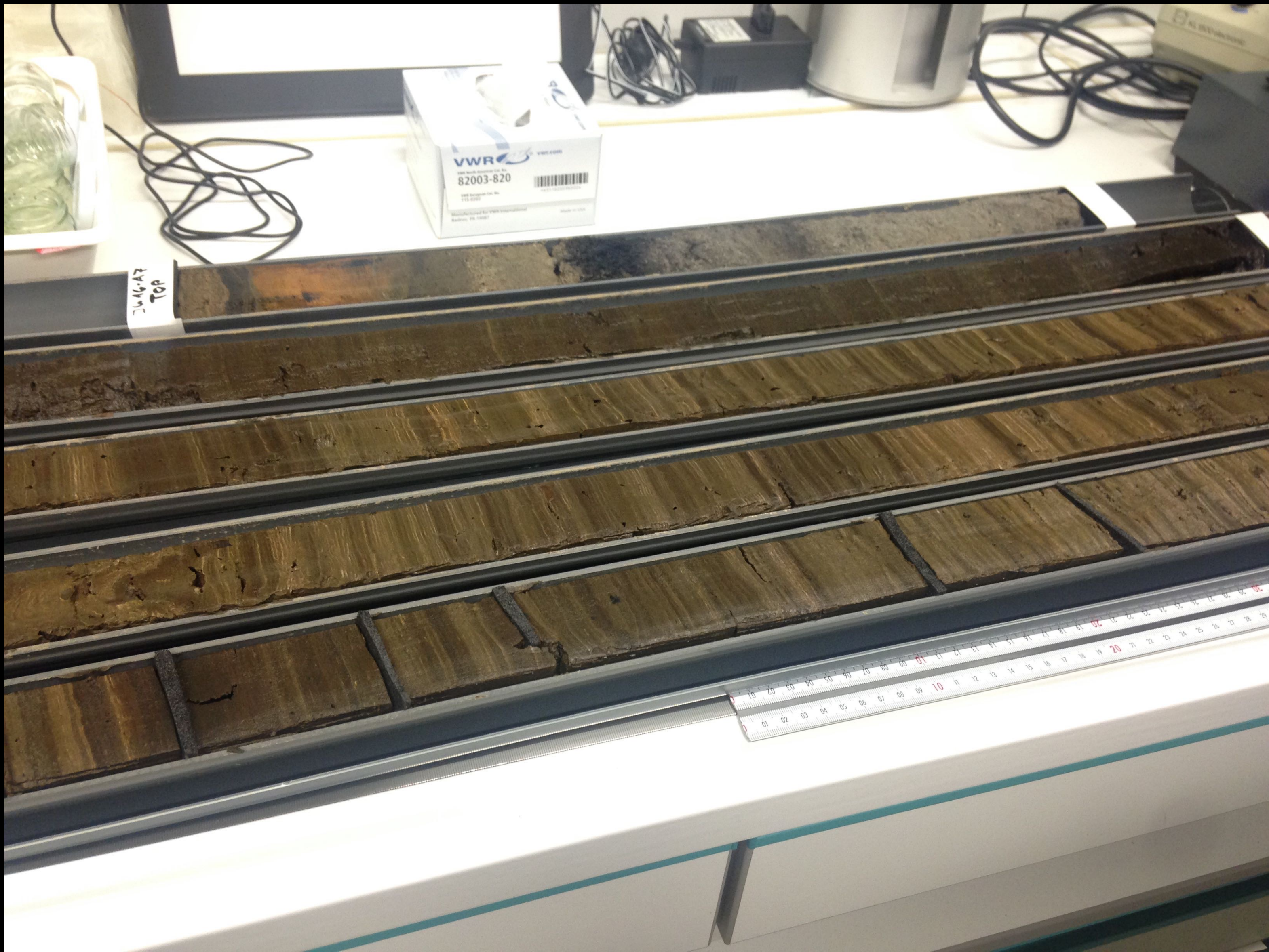
Allen *et al.*, 1999 Nature

Pollen analysis

Low taxonomic resolution – species often indistinguishable.

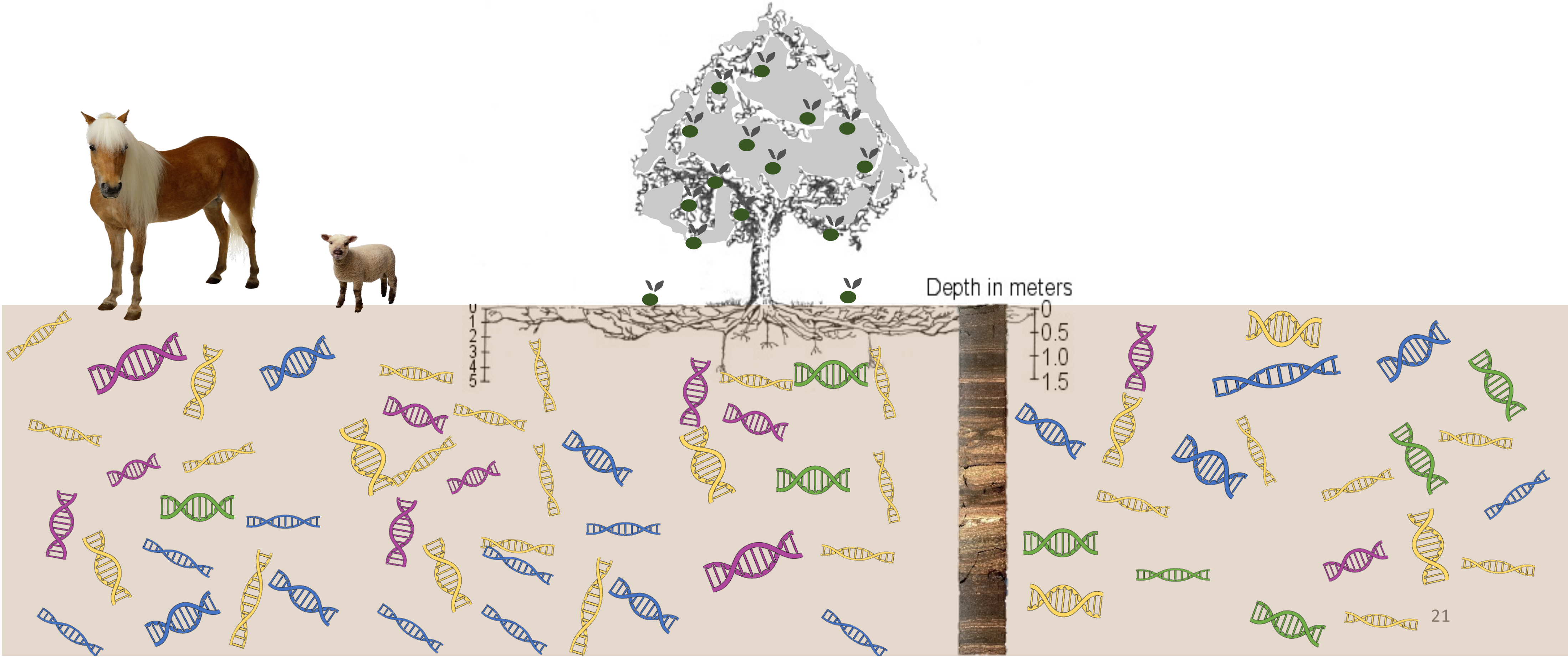
Long-distance transport – pollen can travel far.

Low pollen producers – some plants underrepresented.

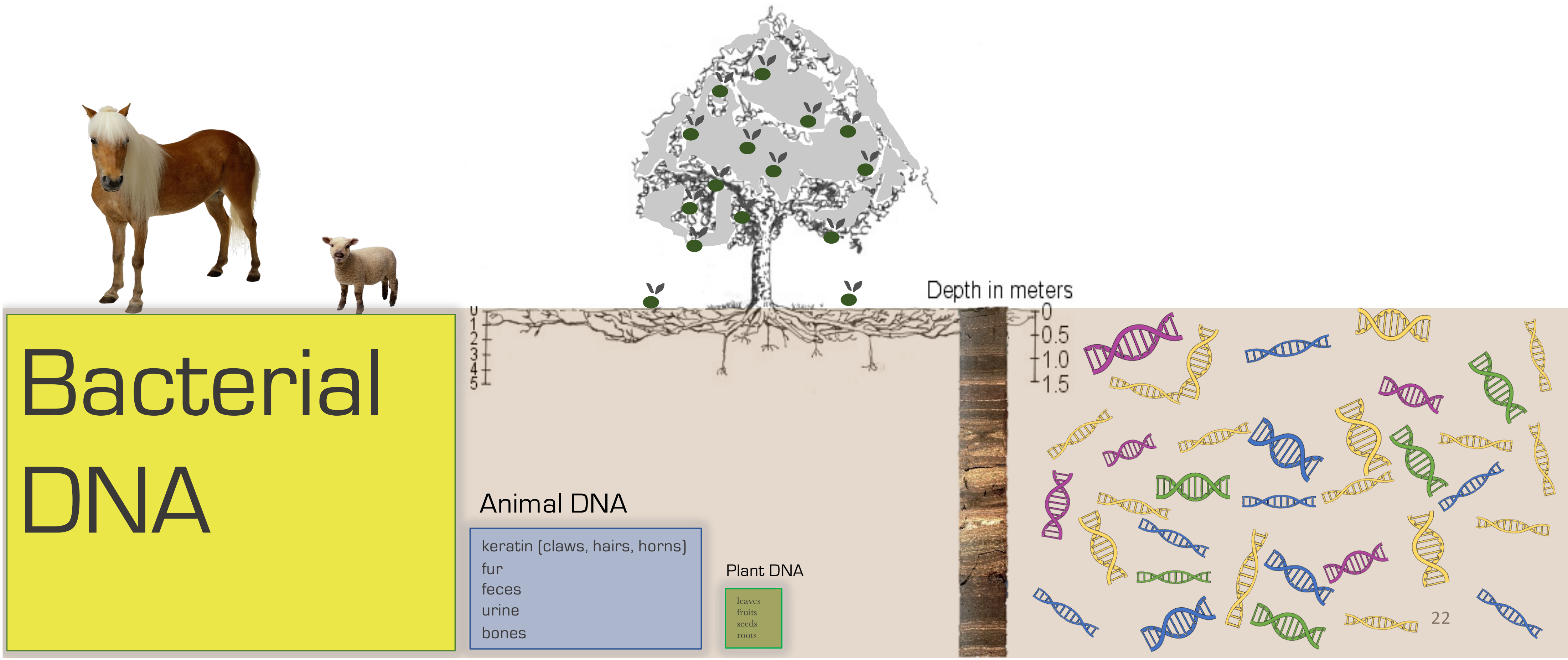


DNA analysis

Sedimentary DNA



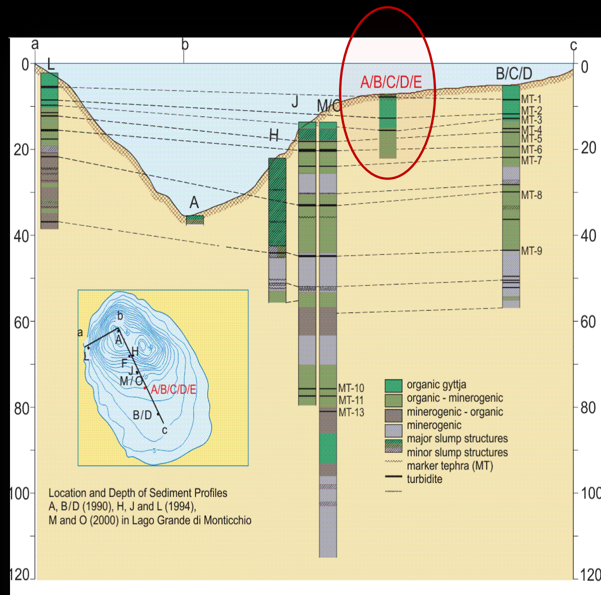
Sedimentary DNA



Sedimentary DNA analysis

LAGO GRANDE DI MONTICCHIO (BASILICATA)

- One of two **maar lakes** created after a major past volcanic explosion.
- Lies far enough south to have escaped direct **effect of the glaciations.**
- One of the **major glacial refugia** for European biota.
- Mediterranean climate with distinguished **seasonal changes**.
- Very **long fossil archive** covering **130.000 years**.



Coring

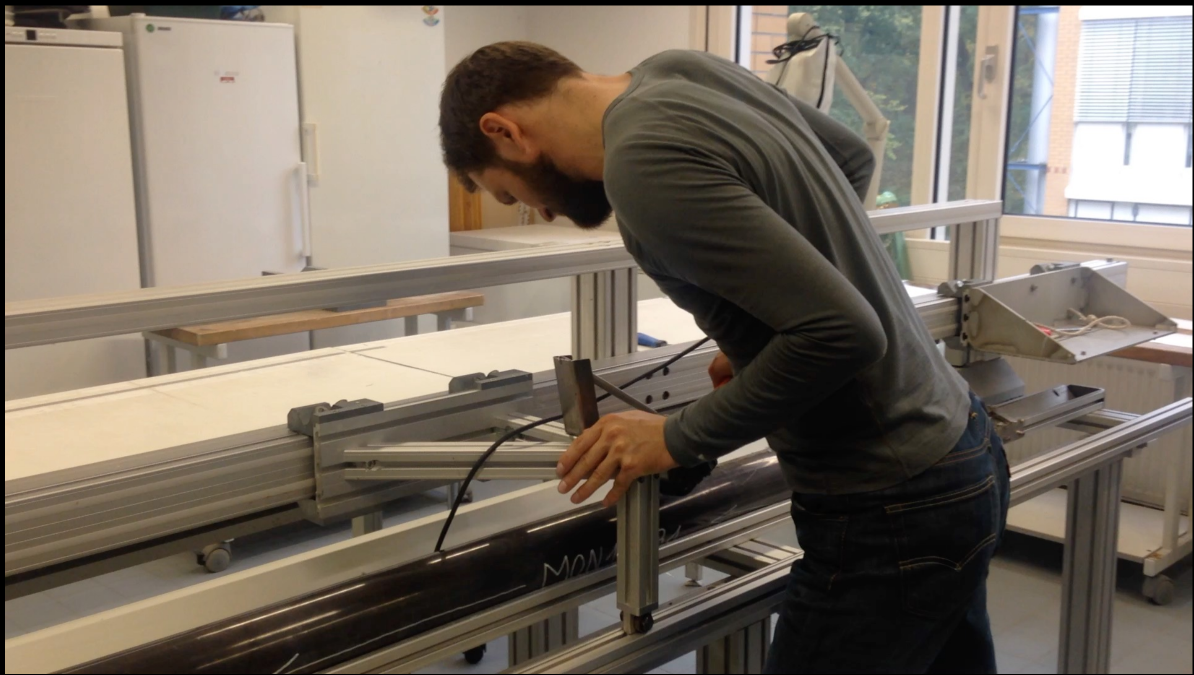
Short, hand-operated piston corer


UWITEC

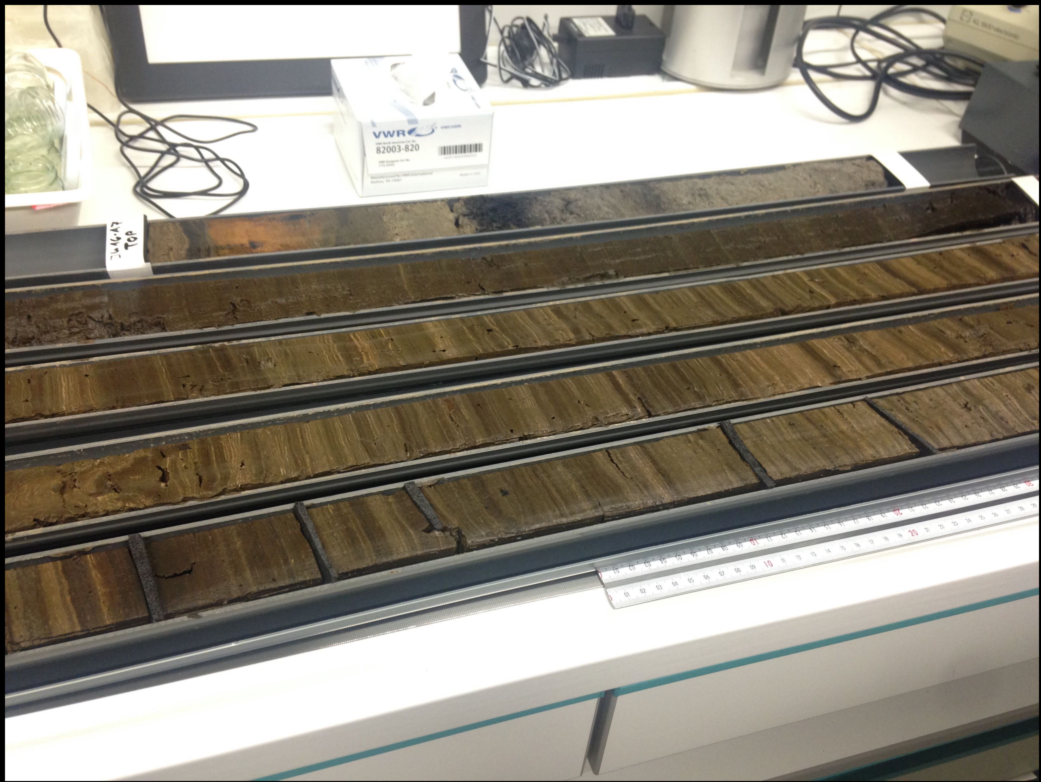
Sub-sampling for DNA analysis

Opening **MON16-B core** at the Helmholtz Centre for Geosciences, Potsdam, Germany

Sub-sampling for DNA analysis



Opening **MON16-B core**

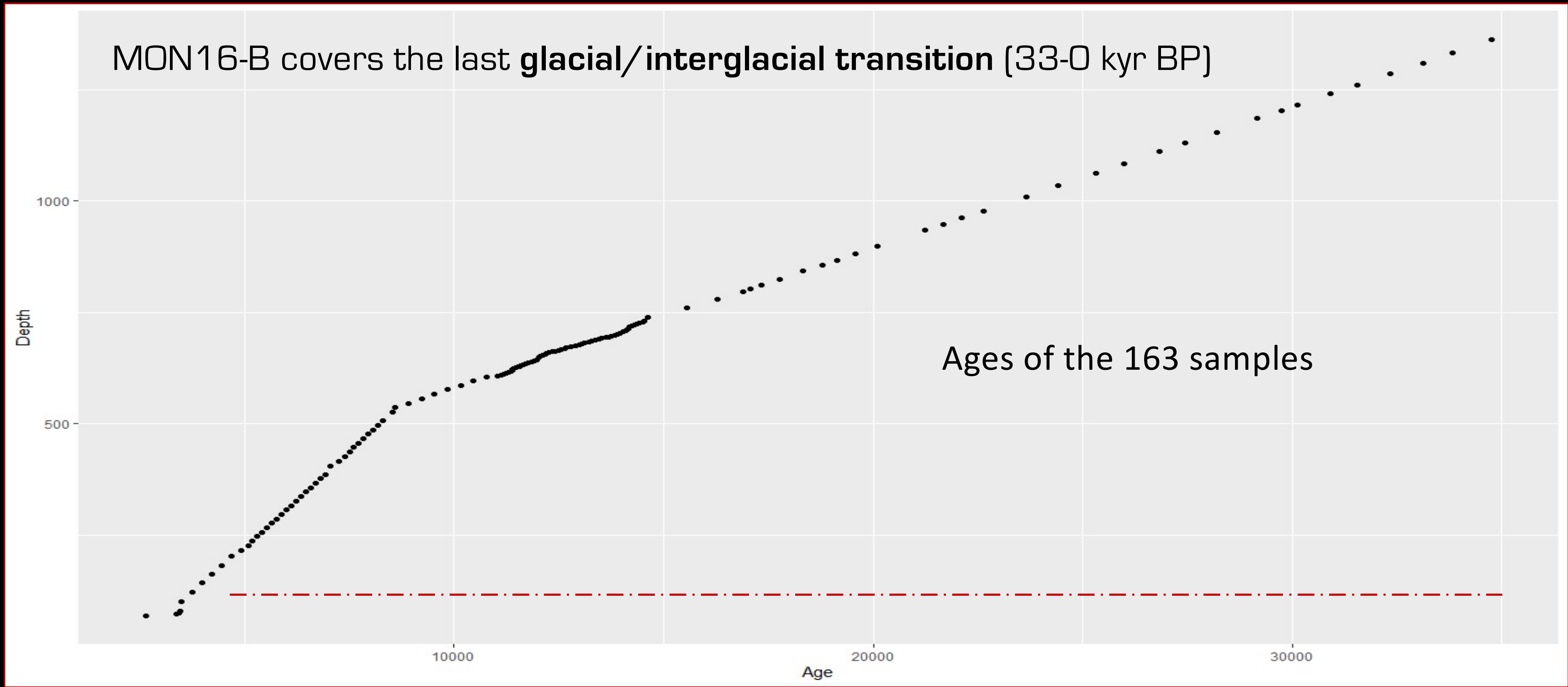


Geochemical analyses to determines the proportion of metallic (Cu, Pb Zn, Au, Ag) or non- metallic (Fl, S, P) elements which reflect natural or anthropogenic shifts in the watershed.

Cleaning **MON16-B core**

Sub-sampling **MON16-B core**

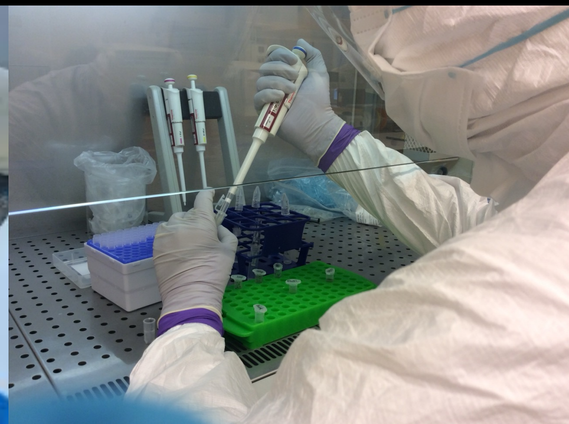
Radiocarbondating



Ancient DNA lab

- Bleaching and UV light
- Positive air pressure
- Isolated from modern DNA laboratories

Ancient DNA lab workflow



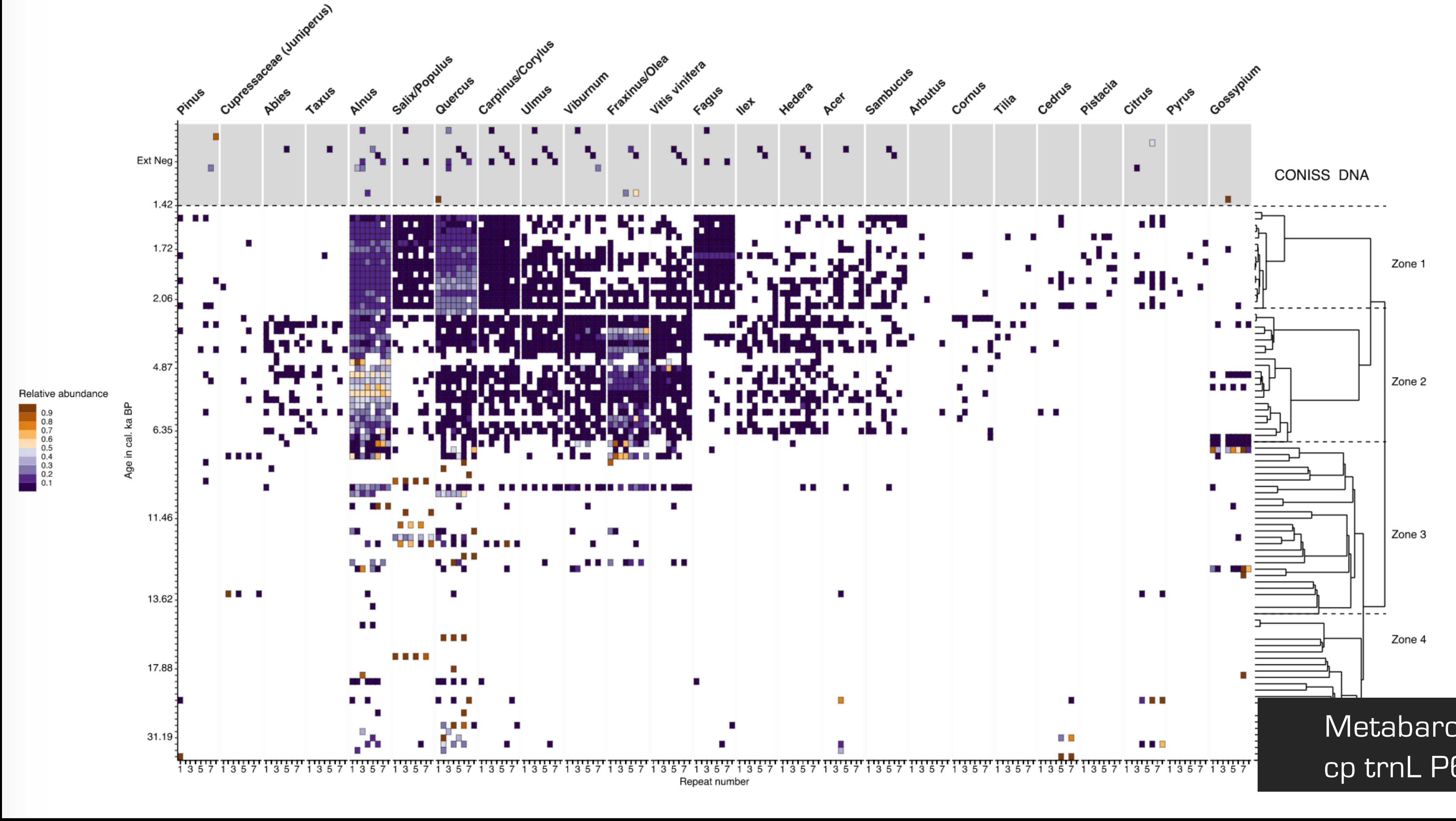
DNA extraction
0.25-10 g sediment
(Dneasy powersoil kit)

Metabarcoding PCR
cp **trnL P6 loop**
(8 replicates/sample)

Library preparation

Illumina sequencing

MiSeq metabarcoding
NovaSeq metagenomics



Metabarcoding
cp trnL P6 loop

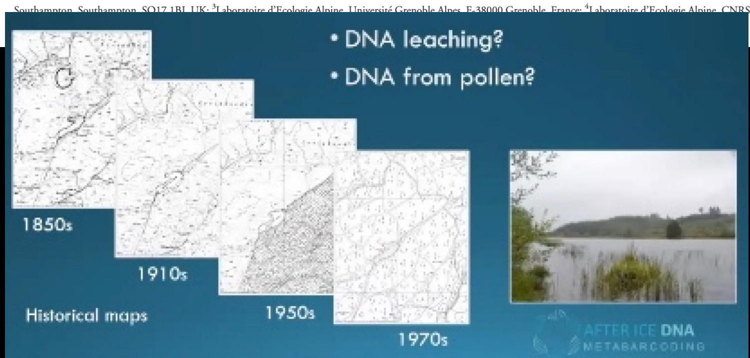
Is leaching a problem?

New Phytologist — Research

Lake sedimentary DNA accurately records 20th Century introductions of exotic conifers in Scotland

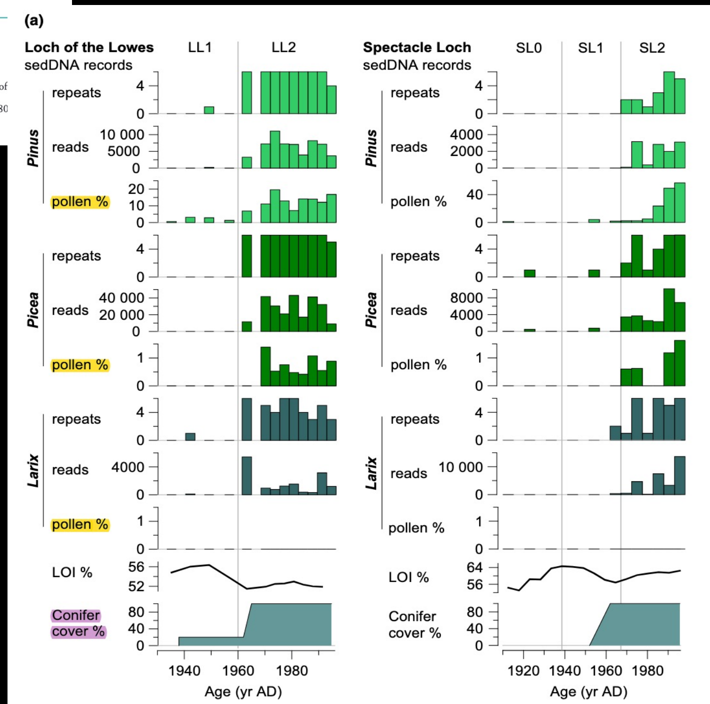
Per Sjögren[1], Mary E. Edwards[1,2], Ludovic Gielly[3,4], Catherine T. Langdon[2], Ian W. Croudace[5], Marie Kristine Føreid Merkel[1], Thierry Fonville[2] and Inger Greve Alsos[1]

[1]Tromsø University Museum, UiT – The Arctic University of Norway, Lars Thøringsvei 10, N-9037 Tromsø, Norway; [2]Department of Geography and Environment, University of



Sjögren *et al.* 2017 New Phyt

- No leaching
- Local signal (DNA signal not coming from pollen)



Metabarcoding
cp trnL P6 loop

How is vegetation around a lake represented?

Alsos *et al.* 2018 PLOS One

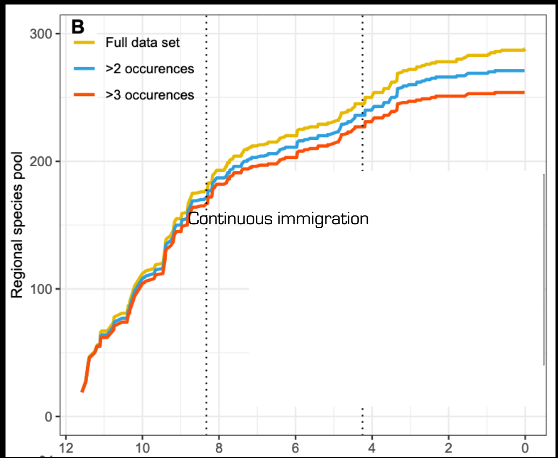
- 11 sites DNA and vegetation surveys
- DNA depends on **abundance**, **family**, **distance** and **site***
- DNA detects more taxa (species) than pollen
- 25-60% of plant taxa detected
- DNA detect a continuous increse of species during postglacial recolonization not seen in pollen
- Forbs better detected

* algae DNA presence reduces the amount of vascular plant DNA

Metabarcoding
cp trnL P6 loop

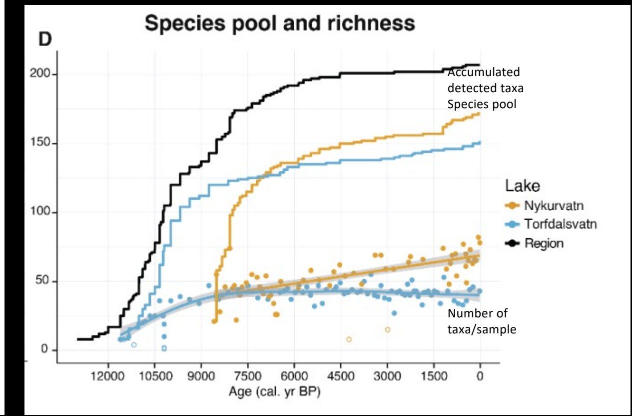
How do plant communities develop through time?

FENNOSCANDIA



Rijal *et al.* 2021 Sci Adv

ICELAND



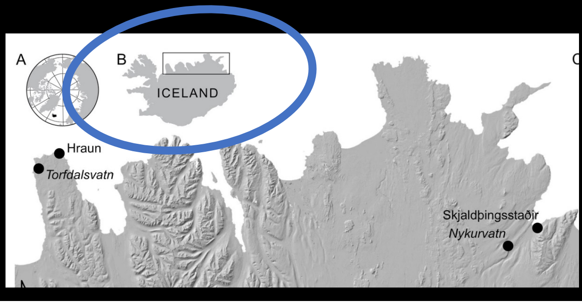
Alsos *et al.* 2021

- Species richness: clear levelling off in Iceland compared to Fennoscandia during mid Holocene

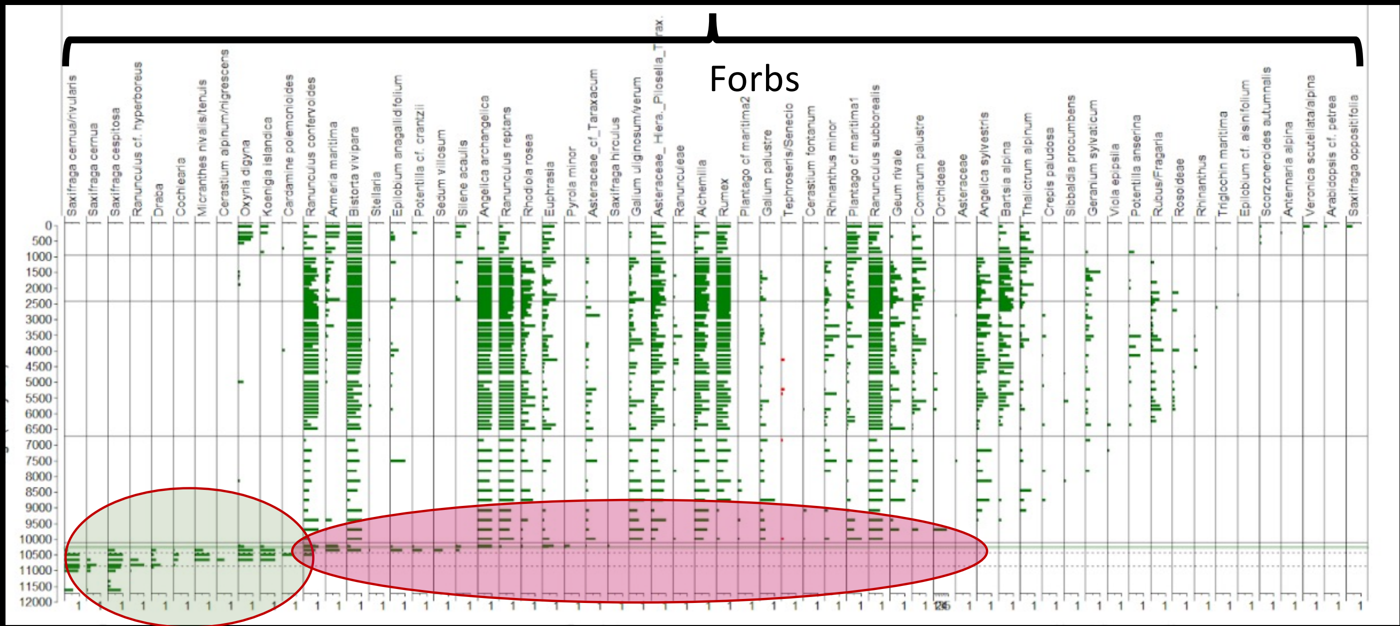
Reason: sea ice was the main dispersal vector for Iceland!

Metabarcoding
cp trnL P6 loop

Potential glacial survivors?



- Potential glacial survivors
- Warming > massive immigration at 10,100 BP
- Levelling off during mid Holocene



Alsos *et al.* 2021

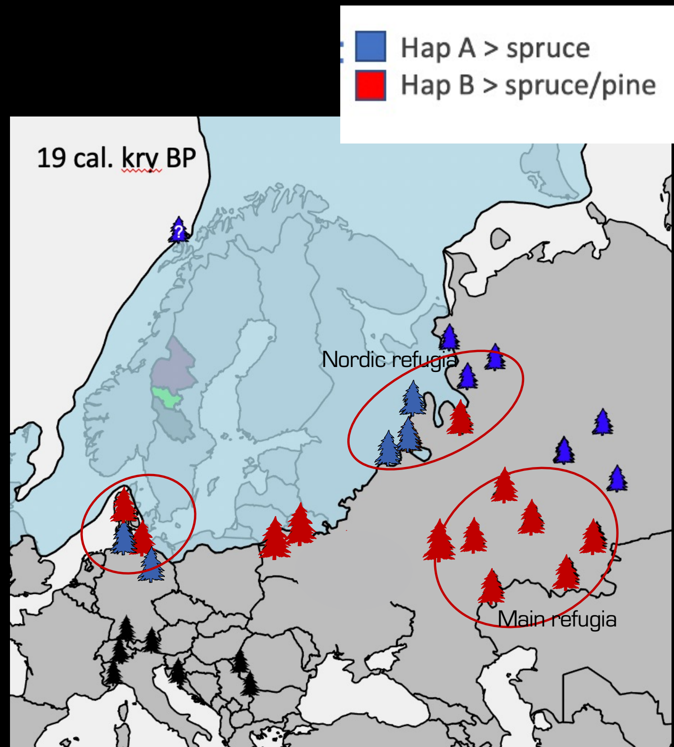
Metabarcoding
cp trnL P6 loop

Potential glacial survivors?

- Glacial survival and nordic glacial refugia for trees

Parducci *et al.* 2012 Science

mtDNA in 138 samples, 14 sites



Nota *et al.* 2022 Nature Comm

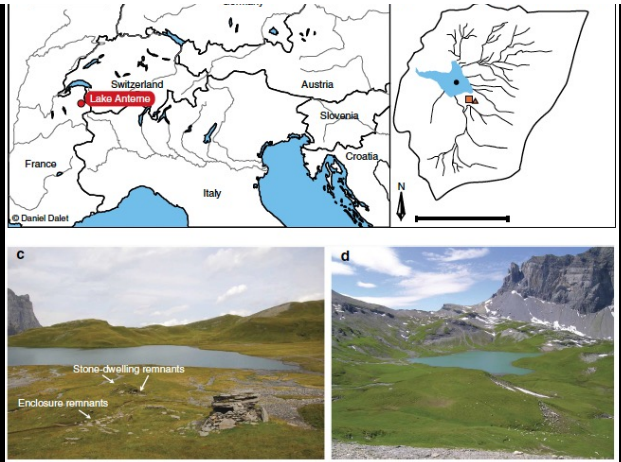
Anthropogenic impact, livestock farming

ARTICLE

Received 21 Mar 2013 | Accepted 7 Jan 2014 | Published 3 Feb 2014 DOI: 10.1038/ncomms4211

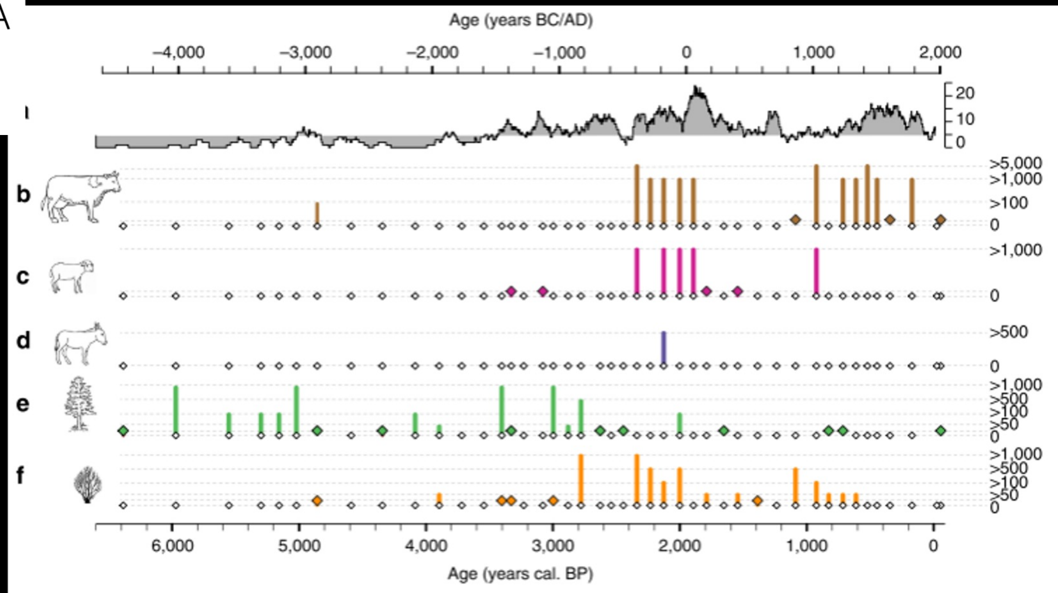
Long livestock farming history and human landscape shaping revealed by lake sediment DNA

Charline Giguet-Covex[1,2,*], Johan Pansu[1,*], Fabien Arnaud[2], Pierre-Jérôme Rey[2], Christophe Griggo[2], Ludovic Gielly[1], Isabelle Domaizon[3], Eric Coissac[1], Fernand David[4], Philippe Choler[1,5], Jérôme Poulenard[2] & Pierre Taberlet[1]



Giguet-Covex *et al.* 2014 Nature Comm

- DNA from domesticated animals and cultivated plants



Metabarcoding

How old is ancient DNA?

0,7-1 Ma

van der Valk *et al.* 2021 Nature

DNA from teeth in permafrost

How old is ancient DNA?

2 Ma!

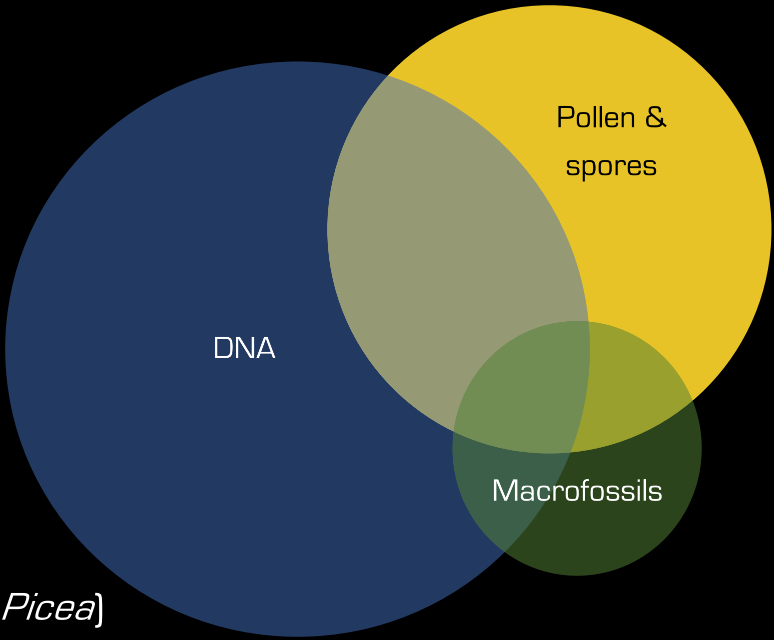
Sedimentary DNA
from permafrost,
Greenland

Kjær *et al.* 2022 Nature

Sedimentary DNA – What we know

Plants

- **Local signal**: long-distance pollen contributes minimally to the DNA signal
- **Complementary** to pollen and macrofossils
- **No leaching** from younger to older sediments
- **Plant richness higher** compared to pollen
- **Higher taxonomic resolution** compared to pollen
- **Forbs** better detected compared to pollen
- Certain taxa are preferentially detected (*Salix, Populus*)
- Low-frequency **contaminants** from plastics/reagents (*Pinus, Picea*)

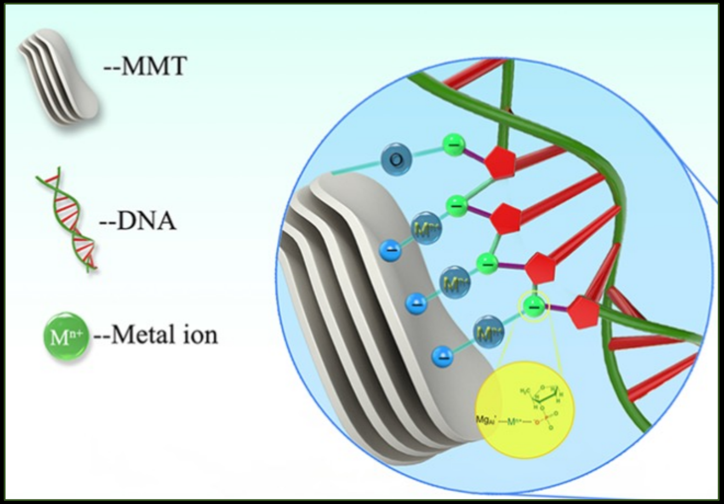


Parducci *et al.* 2017 New Phyt

Sedimentary DNA– What we know

- DNA can preserve for millions of year under cold conditions in terrestrial environments
- Mechanisms that preserve DNA in sediments are different from that of bones.
- **Adsorption at mineral surfaces modifies the DNA conformation**, probably impeding molecular recognition by enzymes, which effectively hinders enzymatic degradation

2 Ma

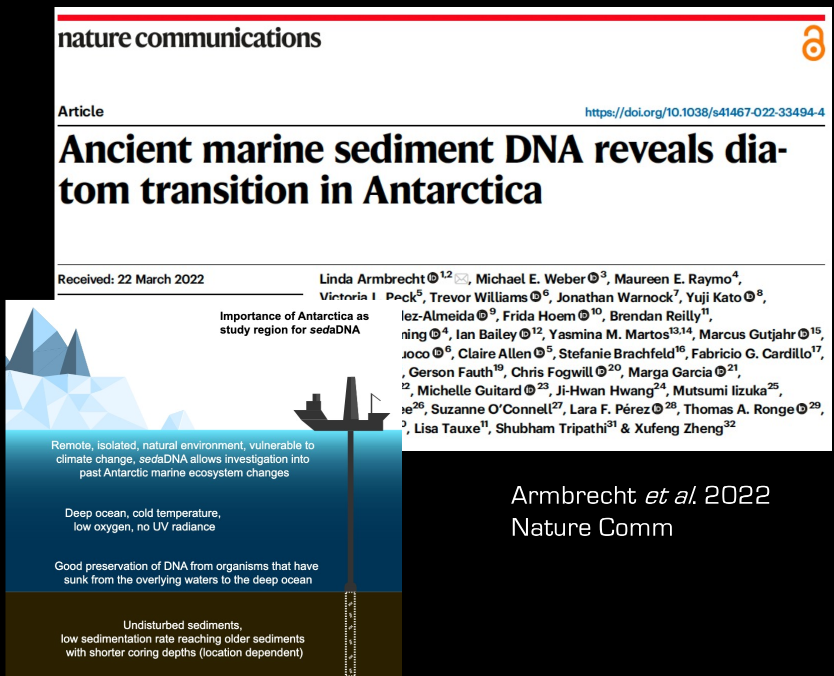


Metagenomics

Kjær *et al.* 2022 Nature

Sedimentary DNA– What we know

- DNA can preserve for millions of year under cold conditions in marine environments



Armbrecht *et al.* 2022
Nature Comm

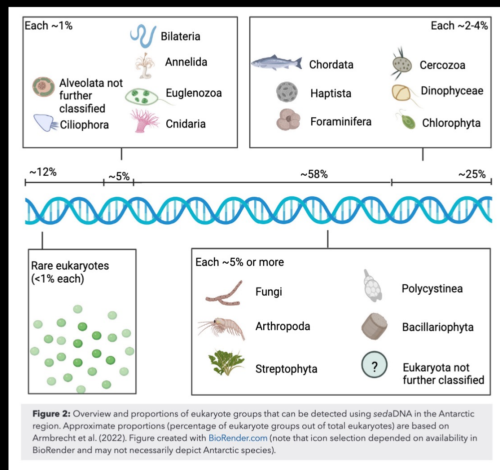


Figure 2: Overview and proportions of eukaryote groups that can be detected using sedaDNA in the Antarctic region. Approximate proportions (percentage of eukaryote groups out of total eukaryotes) are based on Armbrecht et al. (2022). Figure created with BioRender.com (note that icon selection depended on availability in BioRender and may not necessarily depict Antarctic species).

Authenticated marine eukaryote sedaDNA record of ~1 M years and diatom and chlorophyte sedaDNA dating back to ~540 ka.

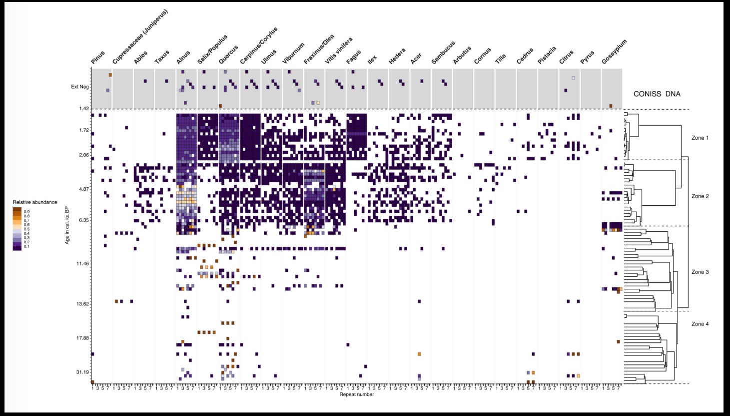
ca.1 Ma

Metabarcoding
SSU, LS, psbO

Sedimentary DNA – What we know

- DNA can preserve for several thousands of years in mid-latitude environments

ca. 30,000 yr



Nota *et al.* in prep

Metabarcoding PCR
cp trnL P6 loop

Sedimentary DNA – What we know

- DNA can preserve for thousands of year in tropical environments

ca. 5000 yr

Contents lists available at ScienceDirect

Quaternary Science Reviews

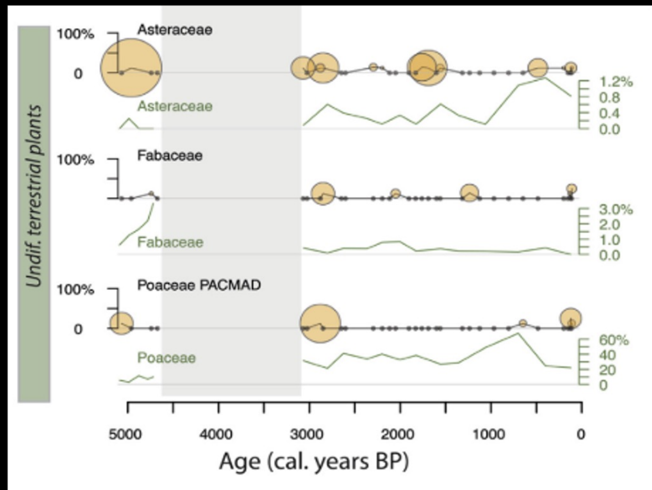
journal homepage: www.elsevier.com/locate/quascirev

ELSEVIER

Five thousand years of tropical lake sediment DNA records from Benin

L. Bremond [a, b, *], C. Favier [a], G.F. Ficetola [c, d, e], M.G. Tossou [f], A. Akouégninou [f], L. Gielly [c, d], C. Giguet-Covex [g], R. Oslisly [h], U. Salzmann [i]

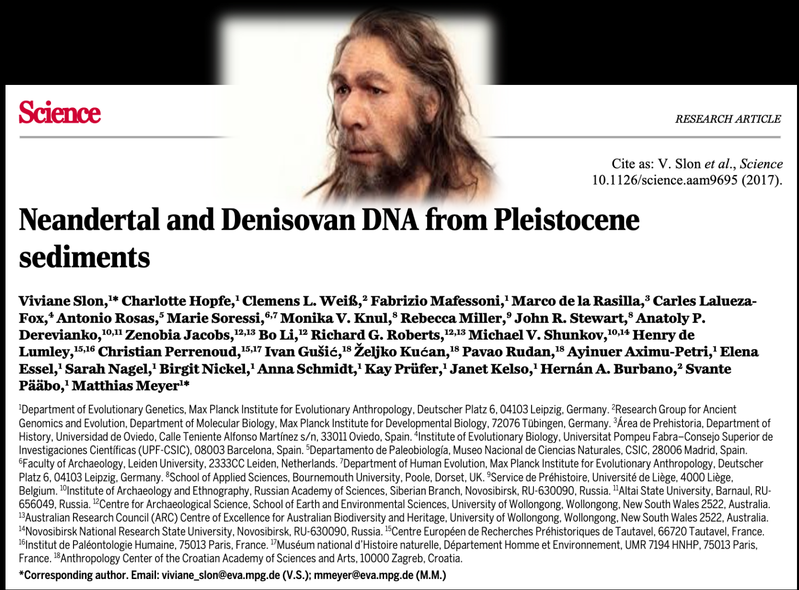
Bremond *et al.* 2022 QSR



Metabarcoding PCR
cp trnL P6 loop

Sedimentary DNA – What we know

- Human DNA can preserve in in absence of bones in cave sediments



Science

RESEARCH ARTICLE

Cite as: V. Slon *et al.*, *Science* 10.1126/science.aam9695 (2017).

Neandertal and Denisovan DNA from Pleistocene sediments

Viviane Slon,[1*] Charlotte Hopfe,[1] Clemens L. Weiß,[2] Fabrizio Mafessoni,[1] Marco de la Rasilla,[3] Carles Lalueza-Fox,[4] Antonio Rosas,[5] Marie Soressi,[6,7] Monika V. Knul,[8] Rebecca Miller,[9] John R. Stewart,[8] Anatoly P. Derevianko,[10,11] Zenobia Jacobs,[12,13] Bo Li,[12] Richard G. Roberts,[12,13] Michael V. Shunkov,[10,14] Henry de Lumley,[15,16] Christian Perrenoud,[15,17] Ivan Gušić,[18] Željko Kućan,[18] Pavao Rudan,[18] Ayinuer Aximu-Petri,[1] Elena Essel,[1] Sarah Nagel,[1] Birgit Nickel,[1] Anna Schmidt,[1] Kay Prüfer,[1] Janet Kelso,[1] Hernán A. Burbano,[2] Svante Pääbo,[1] Matthias Meyer[1*]

[1]Department of Evolutionary Genetics, Max Planck Institute for Evolutionary Anthropology, Deutscher Platz 6, 04103 Leipzig, Germany. [2]Research Group for Ancient Genomics and Evolution, Department of Molecular Biology, Max Planck Institute for Developmental Biology, 72076 Tübingen, Germany. [3]Área de Prehistoria, Department of History, Universidad de Oviedo, Calle Teniente Alfonso Martínez s/n, 33011 Oviedo, Spain. [4]Institute of Evolutionary Biology, Universitat Pompeu Fabra–Consejo Superior de Investigaciones Científicas (UPF-CSIC), 08003 Barcelona, Spain. [5]Departamento de Paleobiología, Museo Nacional de Ciencias Naturales, CSIC, 28006 Madrid, Spain. [6]Faculty of Archaeology, Leiden University, 2333CC Leiden, Netherlands. [7]Department of Human Evolution, Max Planck Institute for Evolutionary Anthropology, Deutscher Platz 6, 04103 Leipzig, Germany. [8]School of Applied Sciences, Bournemouth University, Poole, Dorset, UK. [9]Service de Préhistoire, Université de Liège, 4000 Liège, Belgium. [10]Institute of Archaeology and Ethnography, Russian Academy of Sciences, Siberian Branch, Novosibirsk, RU-630090, Russia. [11]Altai State University, Barnaul, RU-656049, Russia. [12]Centre for Archaeological Science, School of Earth and Environmental Sciences, University of Wollongong, Wollongong, New South Wales 2522, Australia. [13]Australian Research Council (ARC) Centre of Excellence for Australian Biodiversity and Heritage, University of Wollongong, Wollongong, New South Wales 2522, Australia. [14]Novosibirsk National Research State University, Novosibirsk, RU-630090, Russia. [15]Centre Européen de Recherches Préhistoriques de Tautavel, 66720 Tautavel, France. [16]Institut de Paléontologie Humaine, 75013 Paris, France. [17]Muséum national d'Histoire naturelle, Département Homme et Environnement, UMR 7194 HNHP, 75013 Paris, France. [18]Anthropology Center of the Croatian Academy of Sciences and Arts, 10000 Zagreb, Croatia.

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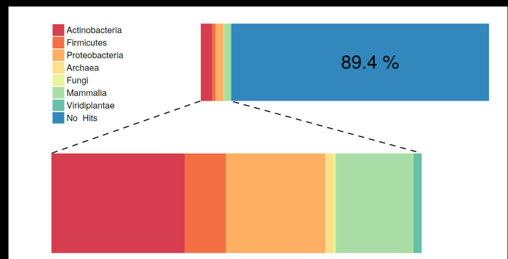
Slon *et al.* 2017 Science

- A paper published in 2017 found Neanderthal and human's DNA in cave sediments using shotgun sequencing + capture enrichment
- They sequenced millions of DNA sequences from multiple sediment samples
- A large proportion of the DNA recovered was of microbial origin.



Shotgun and capture enrichment

Sedimentary DNA – What we want to understand better

- **Taphonomy:** processes **driving the archiving of extracellular DNA** from plants and animals in the sediments

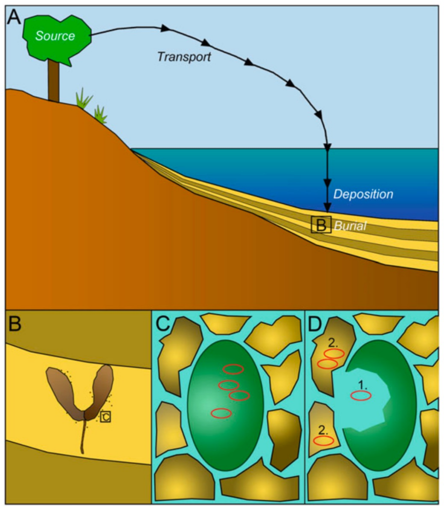
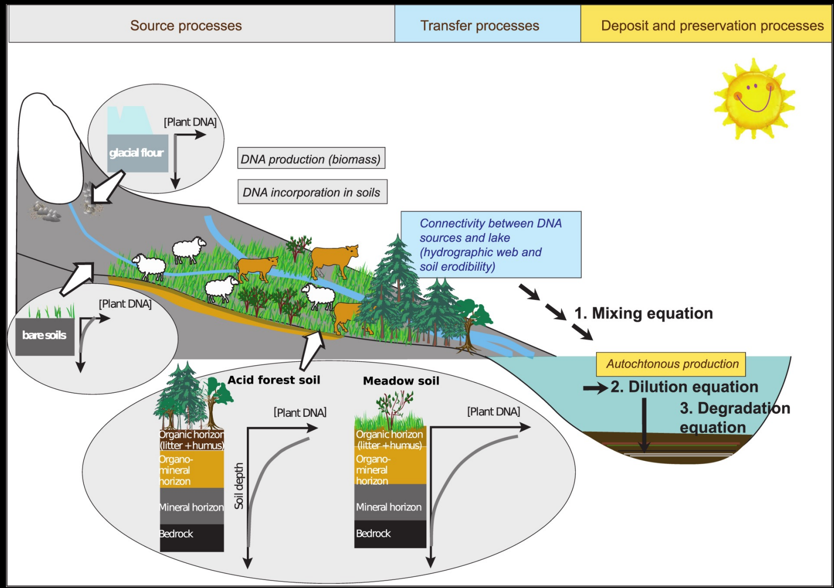


Fig. 3 Diagram illustrating different aspects of taphonomy and preservation of plant DNA in lake sediments: a seed from a tree (source of DNA) is transported via wind and deposited in the lake where it is buried in the sediments (**a**); DNA may be extracted from the seed itself, if preserved (**b**), but the tissues may begin to degrade in which case DNA will be present in cells or organelles (e.g. chloroplasts (**c**)) within the sediment. Over time these may also degrade, releasing DNA, which can be free within the sediment (1) or adsorbed to mineral grains within the sediment (2)

Parducci *et al.* 2018



Williams *et al.* 2023

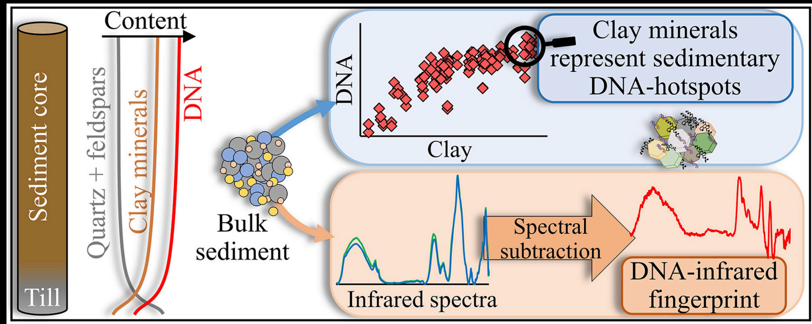
Taphonomic processes acting at the **source** and driving the **transfer**, **deposit** and **preservation** of the DNA in the lake sediments.

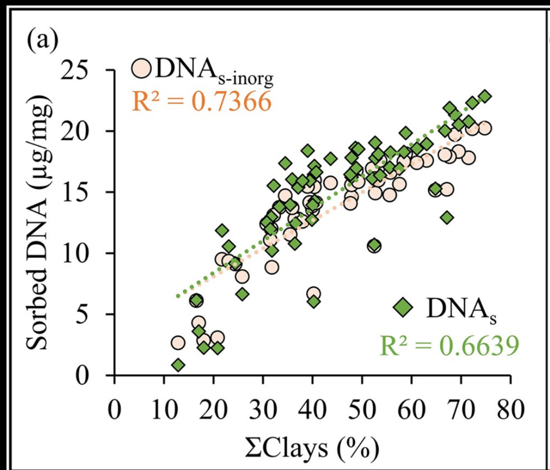
Sedimentary DNA – What we want to understand better

- Potential **DNA-hotspots in sediments**

Minerals, especially **clay minerals**, enhance the preservation of DNA.

However, the role of minerals and organic matter on DNA adsorption onto heterogeneous sediments is still not clear.



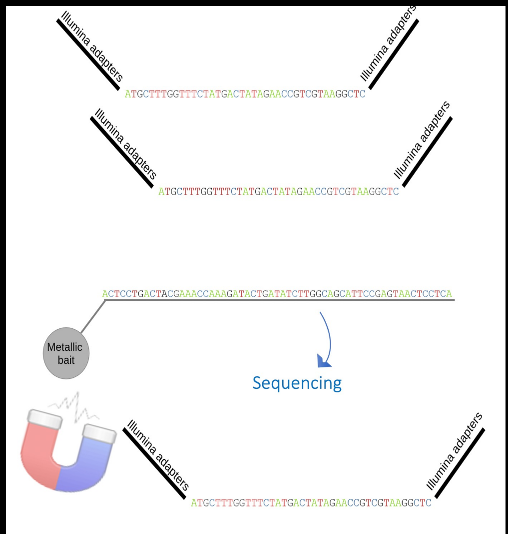


Adsorption of DNA onto sediments and **sediment without organic matter** in µg/mg, according to amount of clay.

Kanbar *et al.* 2020 Appl Geochem

Sedimentary DNA – What we want to understand better

- **Capture enrichment techniques**: do they increase sequencing efficiency, taxonomic resolution, and enable quantitative biodiversity inference from sediments?



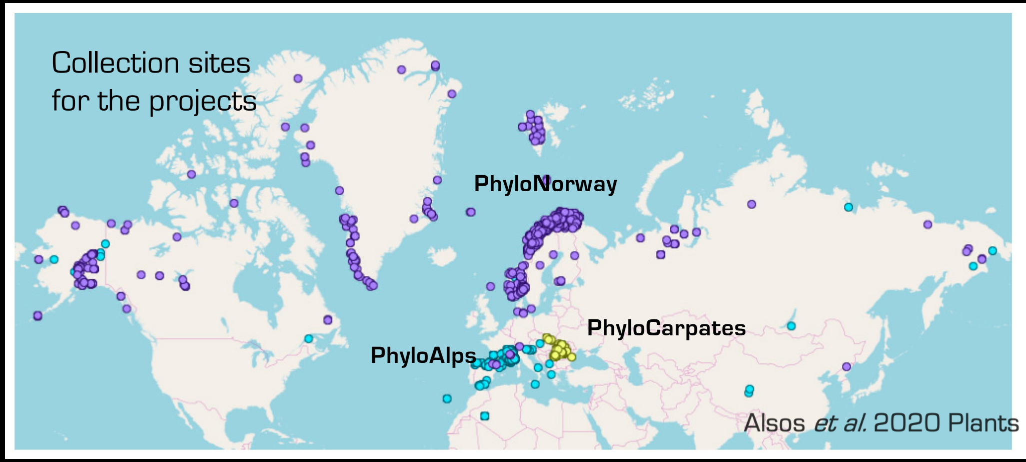
We designed and are testing **capture probes** targeting:

- plant DNA barcodes (rbcL, matK) from major plant orders (Poales, Asterales, Pinales, Fagales);
- plant and animal DNA barcodes (rbcL, matK, COI) from key indicator taxa of lake water quality.

Nota *et al.* 2022
Marconi *et al.* (in preparation)

Sedimentary DNA – What we want to understand better

- Importance of **reference libraries**



Large-scale genome skimming assembly project of cp and nuclear rDNA sequences from:

- 2051 herbarium specimens from Norway/Polar regions
- 4604 modern specimens from Alps and Carpathians

Custom and reduced reference libraries (PhyloNorway, PhyloAlps, PhyloCarpates) have significantly improved taxonomic identification in **metabarcoding studies**.

Sedimentary DNA – What we want to understand better

- Importance of **reference libraries**

BROAD DATABASES

Capture-based analyses appear to benefit from **broad databases**, as bacterial DNA remains abundant and reduced databases may artificially force read assignment to target taxa.

In **mid-latitude and tropical regions**, the high diversity of flora and fauna makes custom-database construction and sequencing impractical.

Some conclusions

What we know about sedDNA

- As with bones, DNA quality depend more on **preservation conditions** than **age**.
- Mechanisms that preserve DNA in sediments are different from that of bones.
- **Adsorption at mineral surfaces modifies the DNA conformation**, probably impeding molecular recognition by enzymes, which effectively hinders enzymatic degradation
- For plants, DNA results depend on **abundance**, **family**, **distance** and **site**
- DNA provides a **local signal** complementary to pollen and macrofossils

What we want to understand better

- **Taphonomy** - how organic remains pass from the biosphere to the lithosphere
- Potential **DNA-hotspots** in sediments
- Efficiency of **capture** vs. shotgun and metabarcoding approaches
- Importance of **reference databases** and **bioinformatic tools** for analyses

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