Paleo-Metagenomics of Late Quaternary Packrat Middens

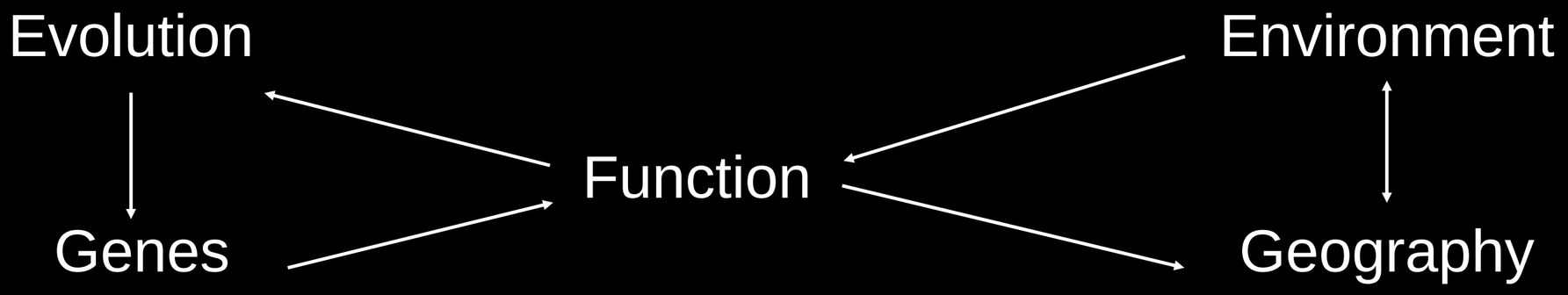
Robert Harbert,
Stonehill College

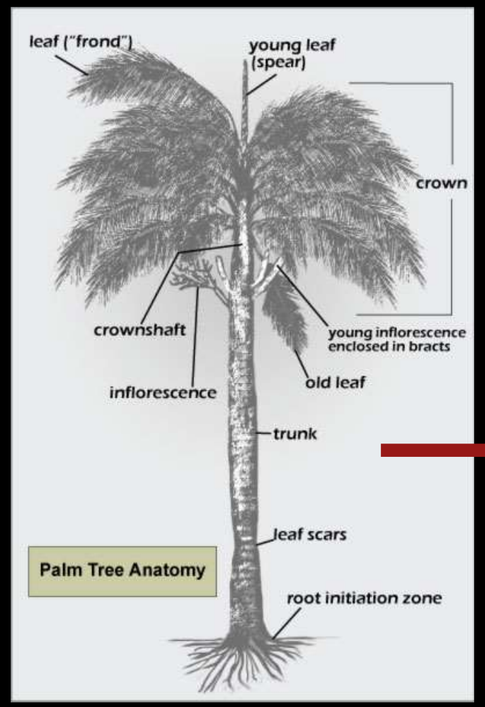
Topics

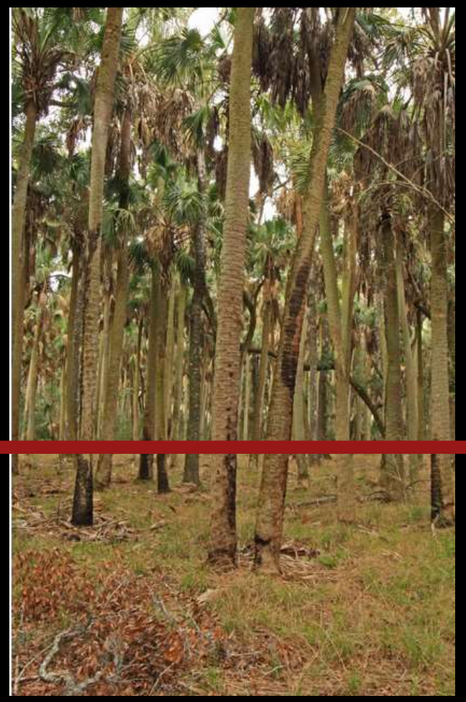
- 1) Packrat middens as paleoecological and paleoclimatological record
- 2) Ancient DNA profiling of fossil packrat middens
- 3) Current teaching/research topics

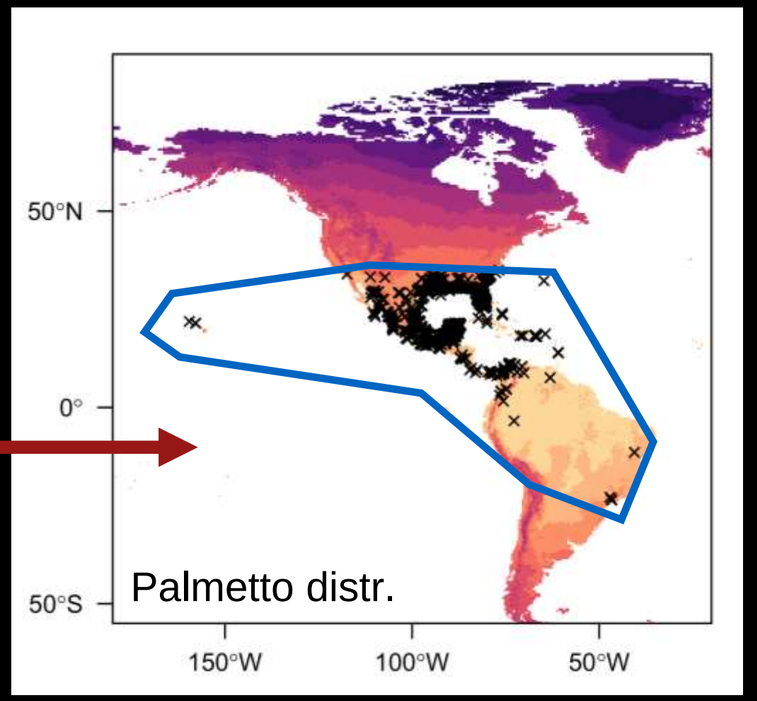
What can plant communities tell us about climate?

Ecological Feedback:

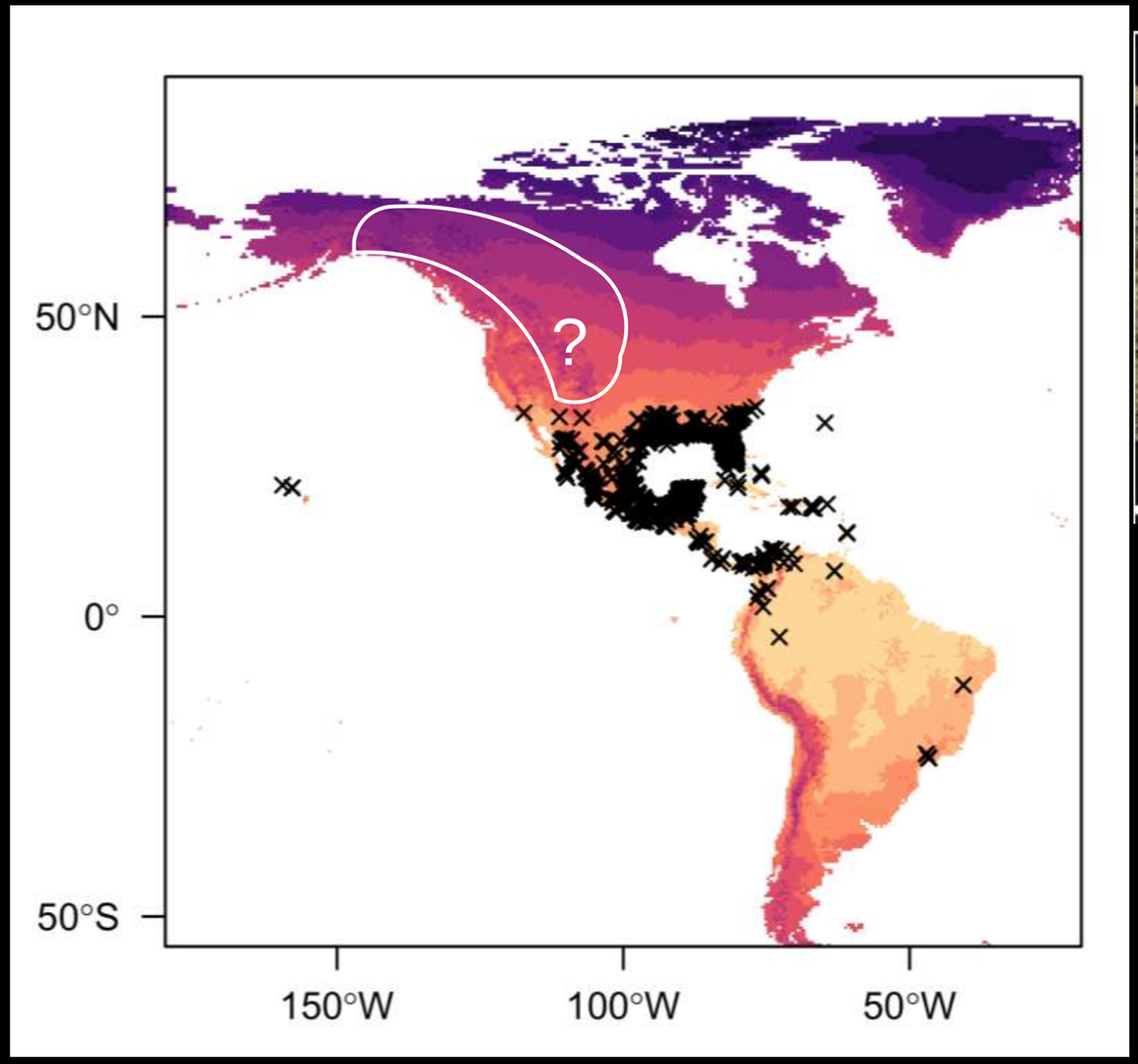








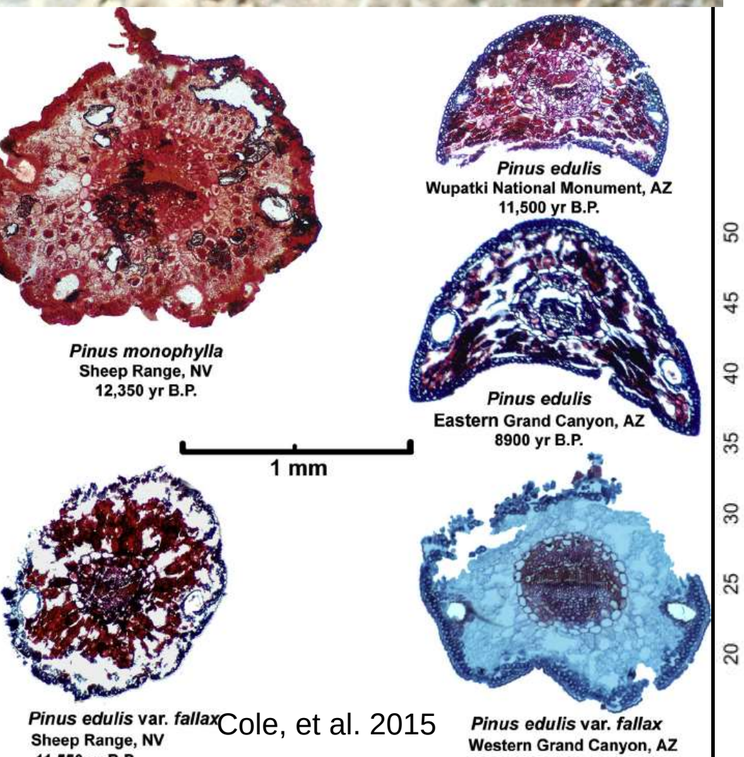
Extrapolation in the Fossil Record



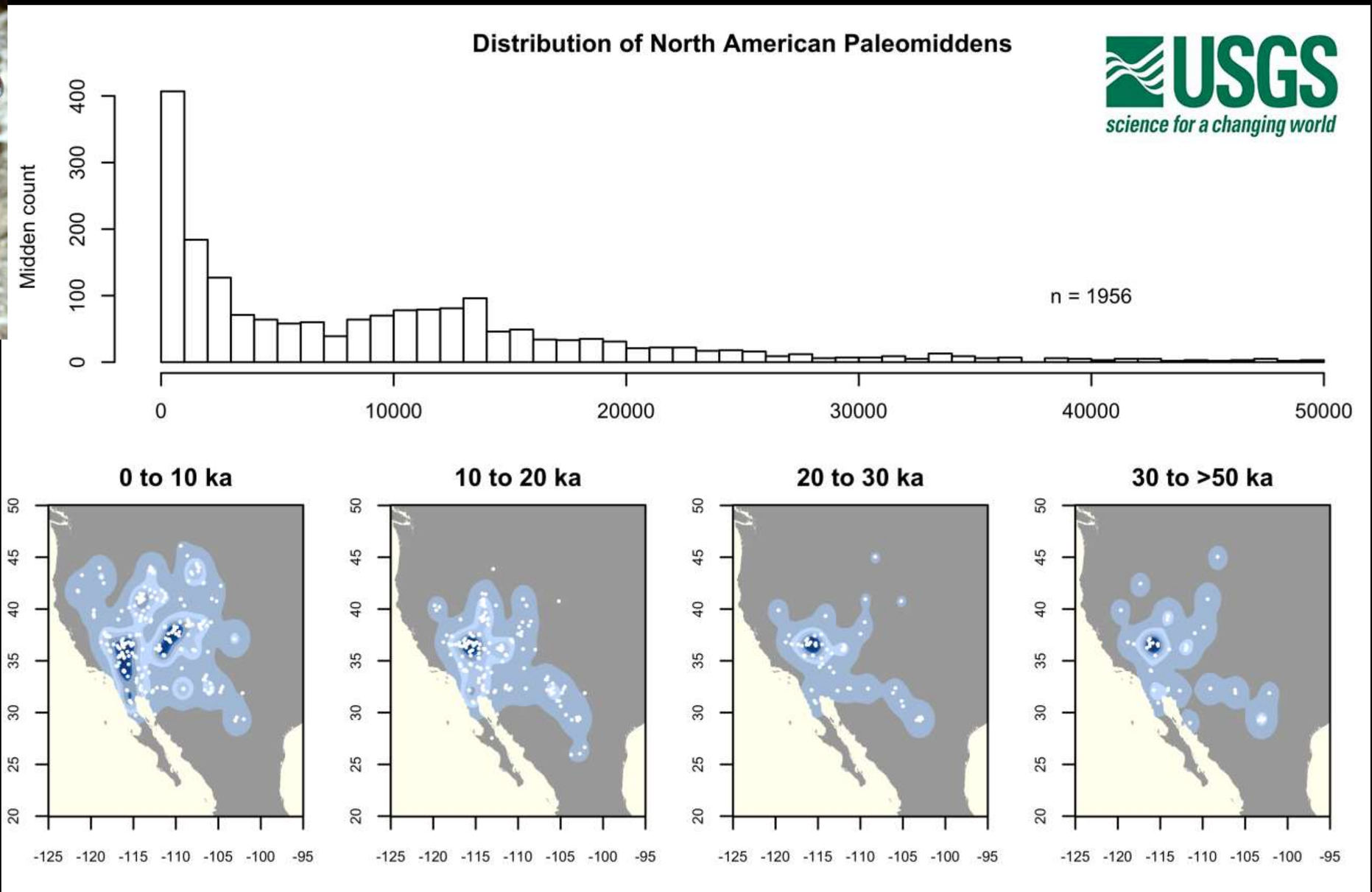
Packrats

Google image search: “packrat nest”

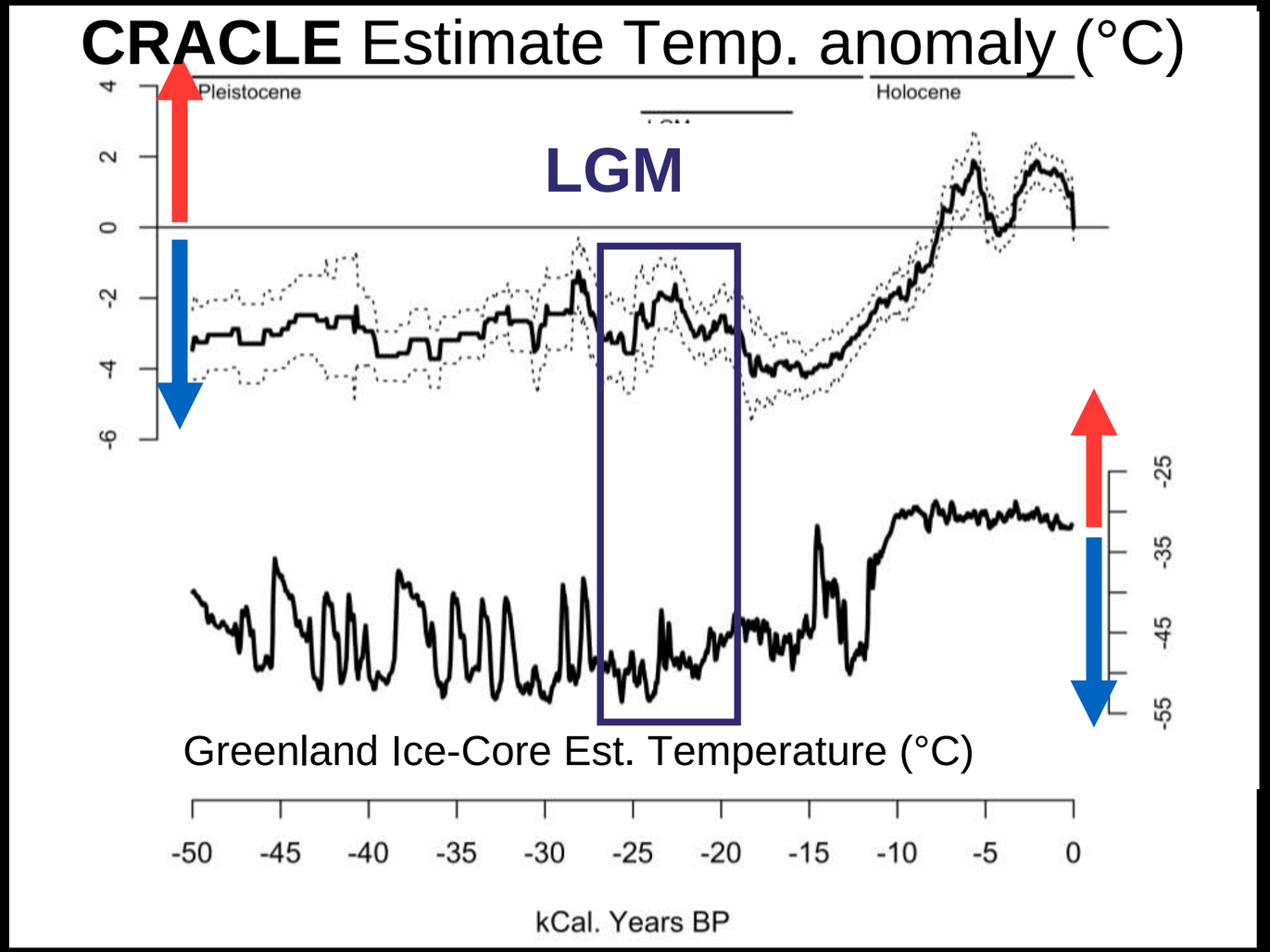
Late Quaternary Packrat (*Neotoma* spp.) midden macrofossils



Cole, et al. 2015



Harbert & Nixon. 2018. **Quantitative Late Quaternary Climate Reconstruction from Plant Macrofossil Communities in Western North America**



Thousand years before present

https://www.openquaternary.com/articles/10.5334/oq.46/

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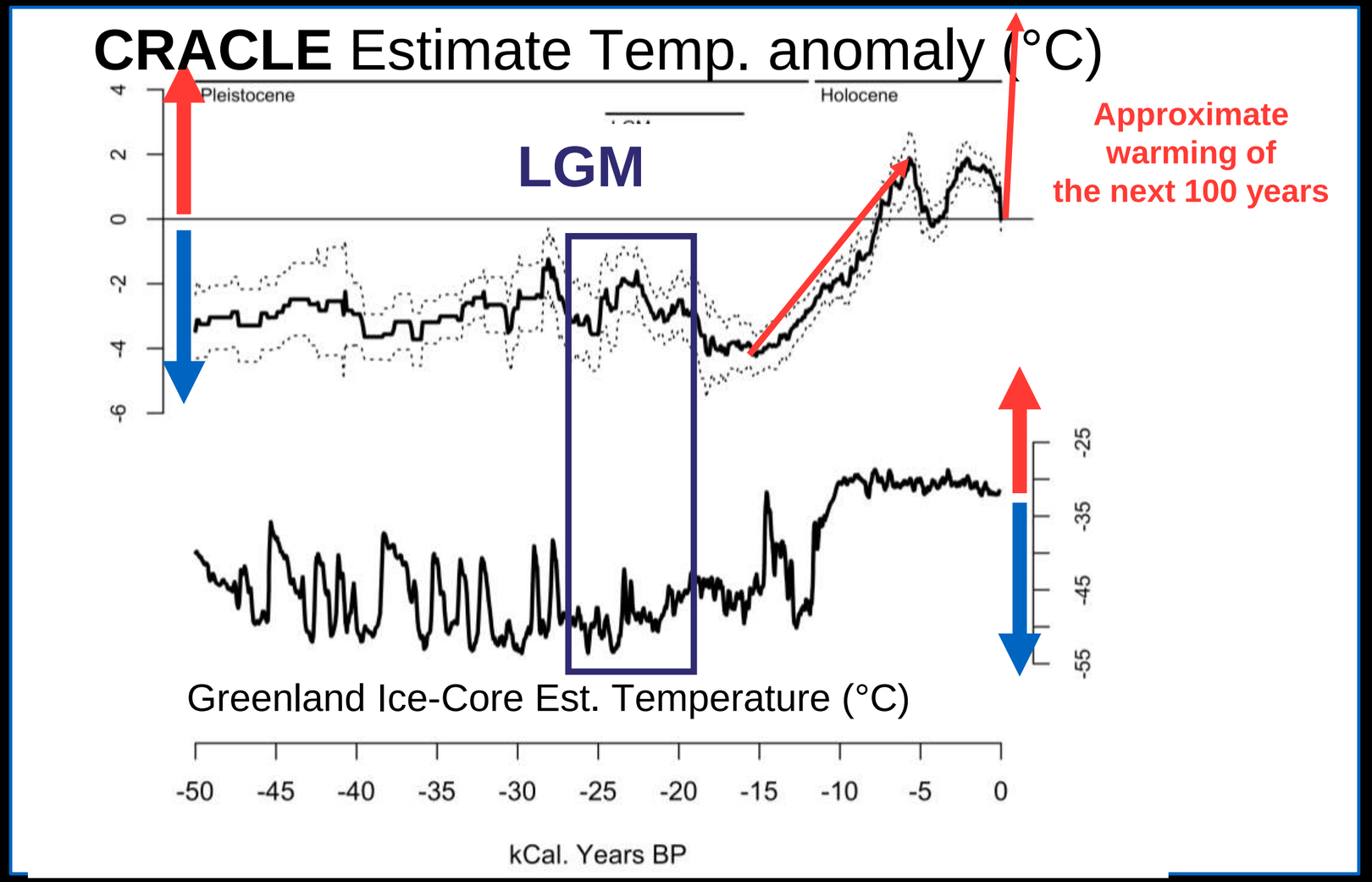


Thousand years before present

https://www.openquaternary.com/articles/10.5334/oq.46/

Coming Soon:

- CRACLE R package:
  - cRacle (https://github.com/rsh249/cRacle.git)
  - R implementation of the CRACLE paleoclimate estimation algorithm and associated functions for data access and visualization.

Ancient DNA

Ancient DNA from Ice-Cores

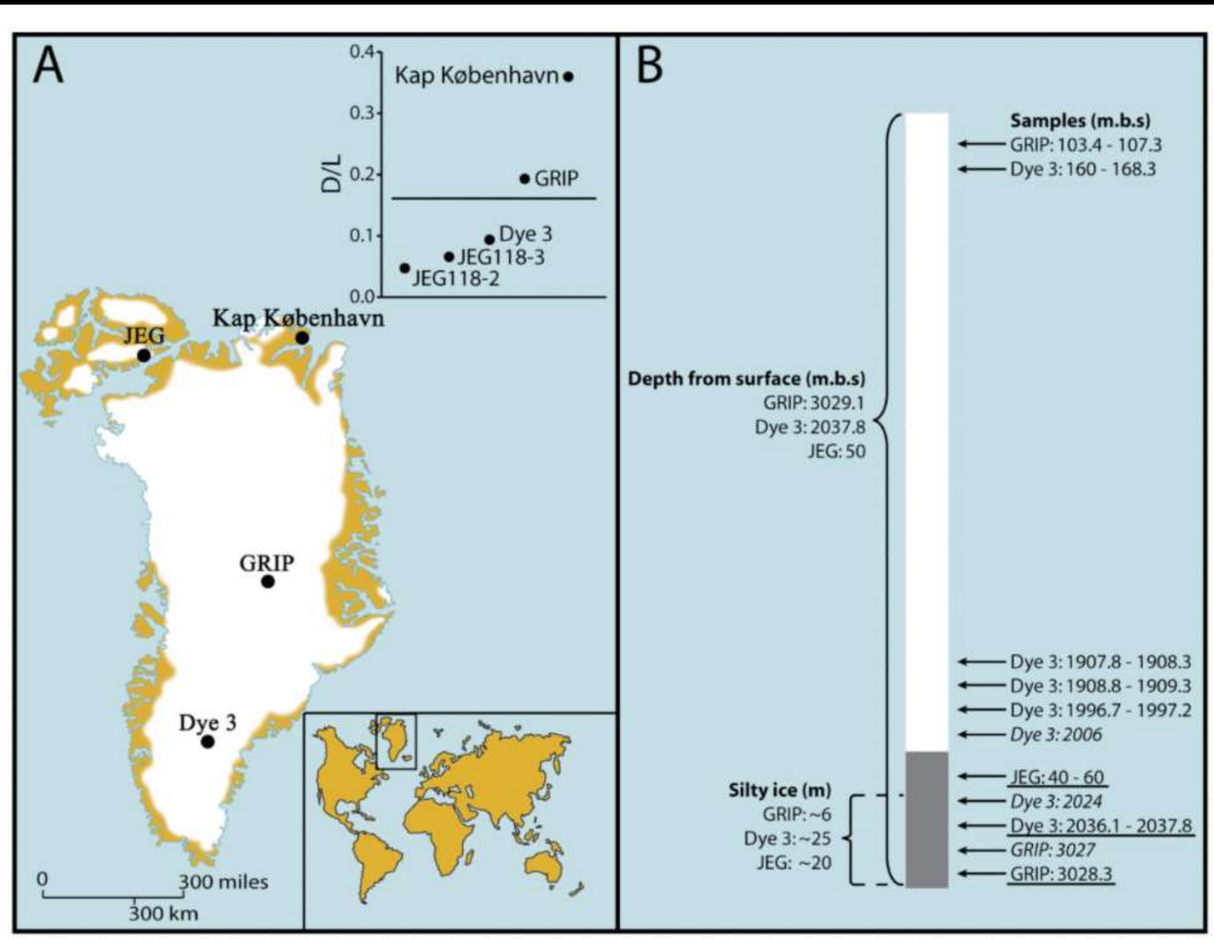
- DNA from plants and insects dating to >500,000 years ago!

Published in final edited form as:
Science. 2007 July 6; 317(5834): 111–114. doi:10.1126/science.1141758.

Ancient Biomolecules from Deep Ice Cores Reveal a Forested Southern Greenland

Eske Willerslev[1,*], Enrico Cappellini[2], Wouter Boomsma[3], Rasmus Nielsen[4], Martin B. Hebsgaard[1], Tina B. Brand[1], Michael Hofreiter[5], Michael Bunce[6,7], Hendrik N. Poinar[7], Dorthe Dahl-Jensen[8], Sigfus Johnsen[8], Jørgen Peder Steffensen[8], Ole Bennike[9], Jean-Luc Schwenninger[10], Roger Nathan[10], Simon Armitage[11], Cees-Jan de Hoog[12], Vasily Alfimov[13], Marcus Christl[13], Juerg Beer[14], Raimund Muscheler[15], Joel Barker[16], Martin Sharp[16], Kirsty E.H. Penkman[2], James Haile[17], Pierre Taberlet[18], M. Thomas P. Gilbert[1], Antonella Casoli[19], Elisa Campani[19], and Matthew J. Collins[2]

[1]Centre for Ancient Genetics, University of Copenhagen, Denmark [2]BioArch, Departments of



Packrat midden aDNA

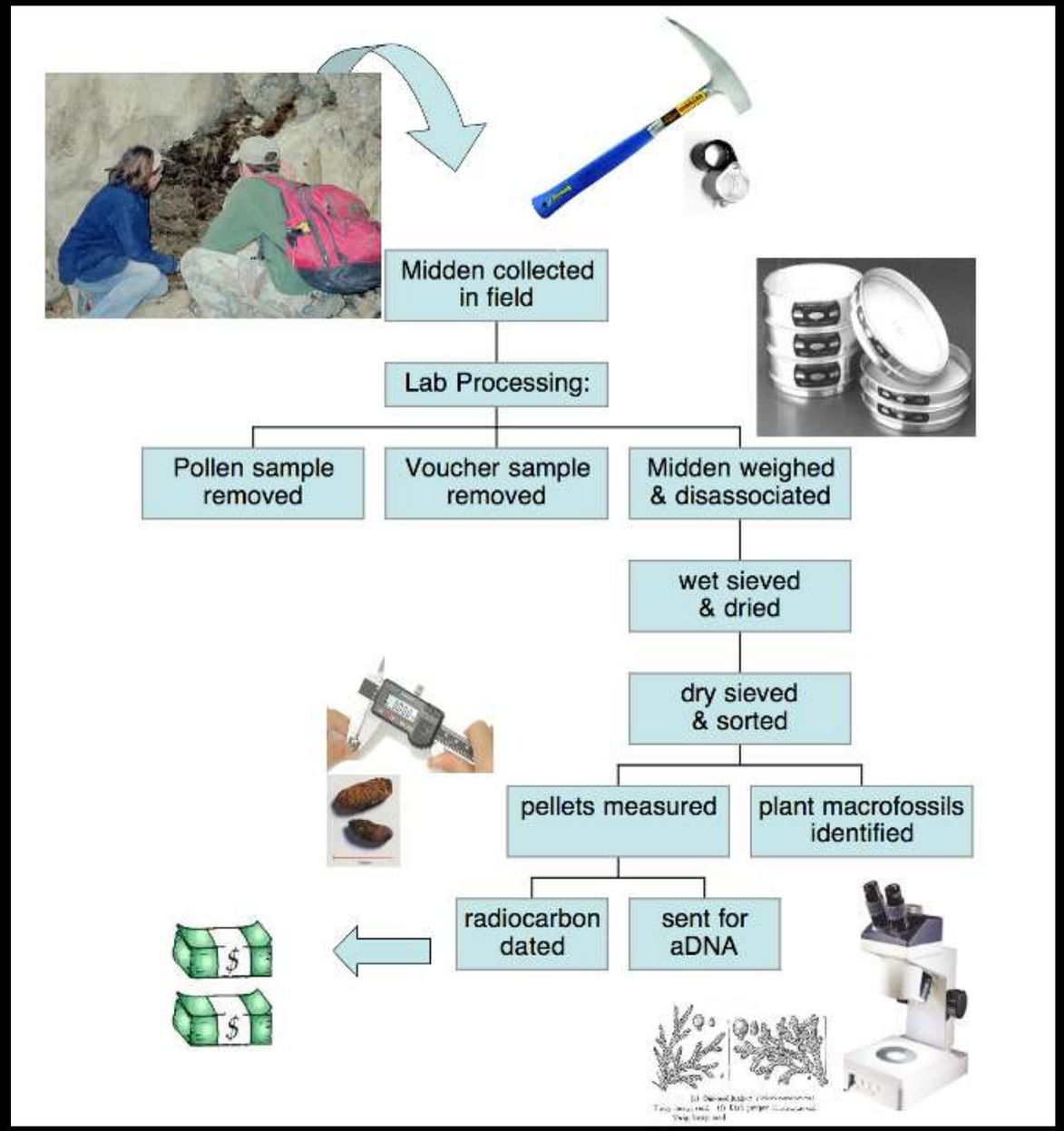


Photo Credit: J. Betancourt, K. Rylander (USGS)

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Packrat midden aDNA

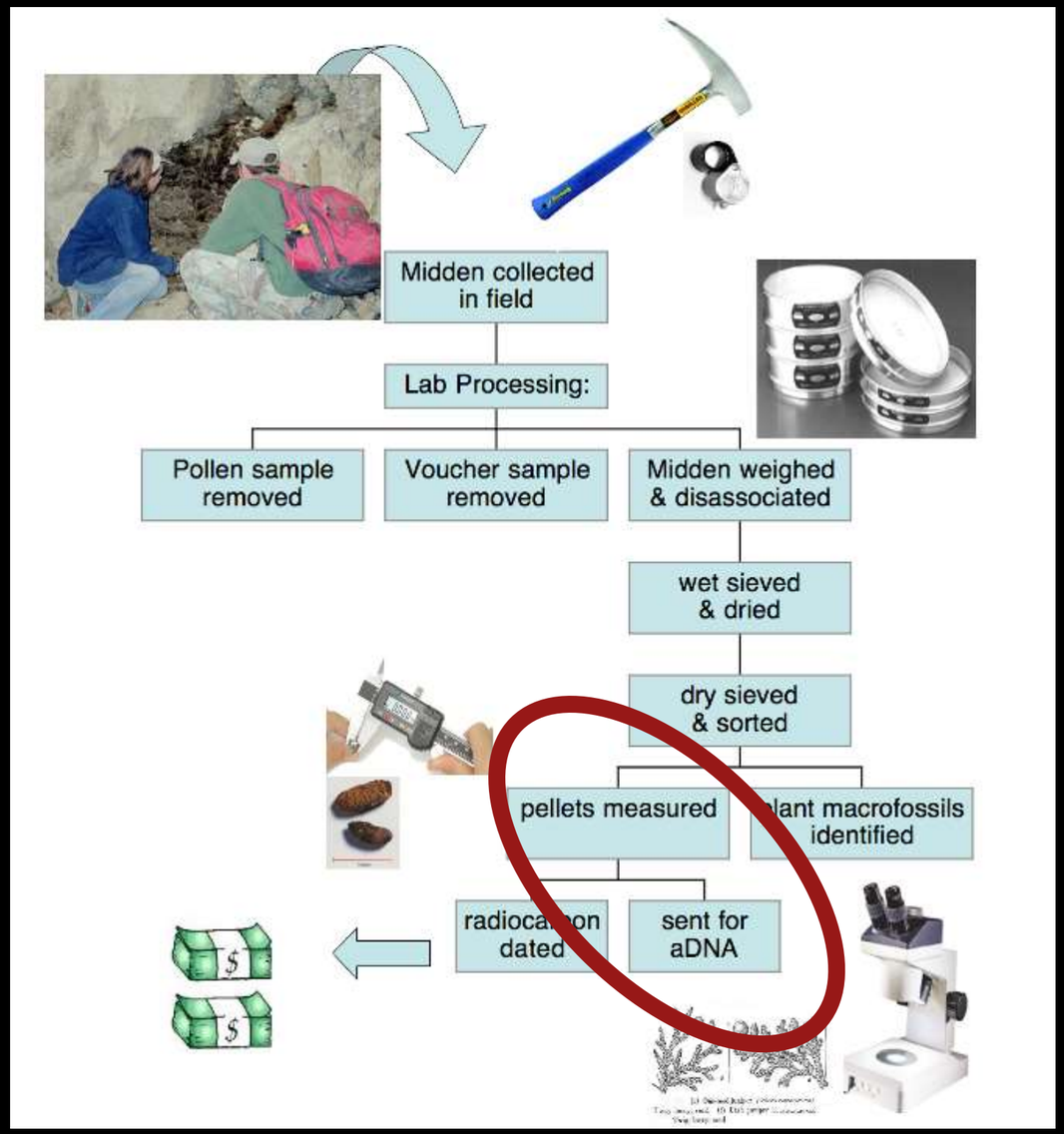
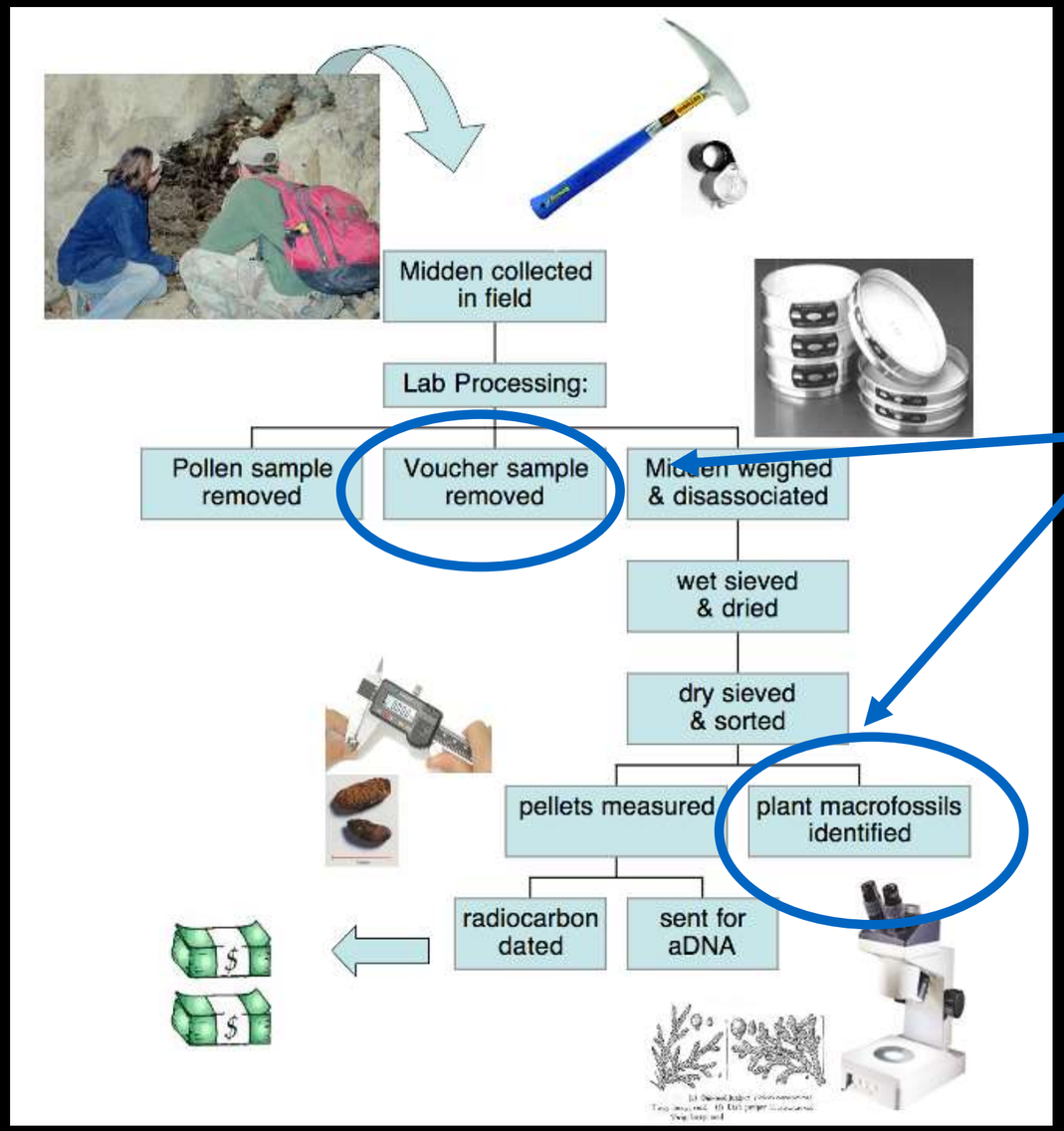


Photo Credit: J. Betancourt, K. Rylander (USGS)

Packrat midden aDNA



Two types of samples that may be used for DNA analysis

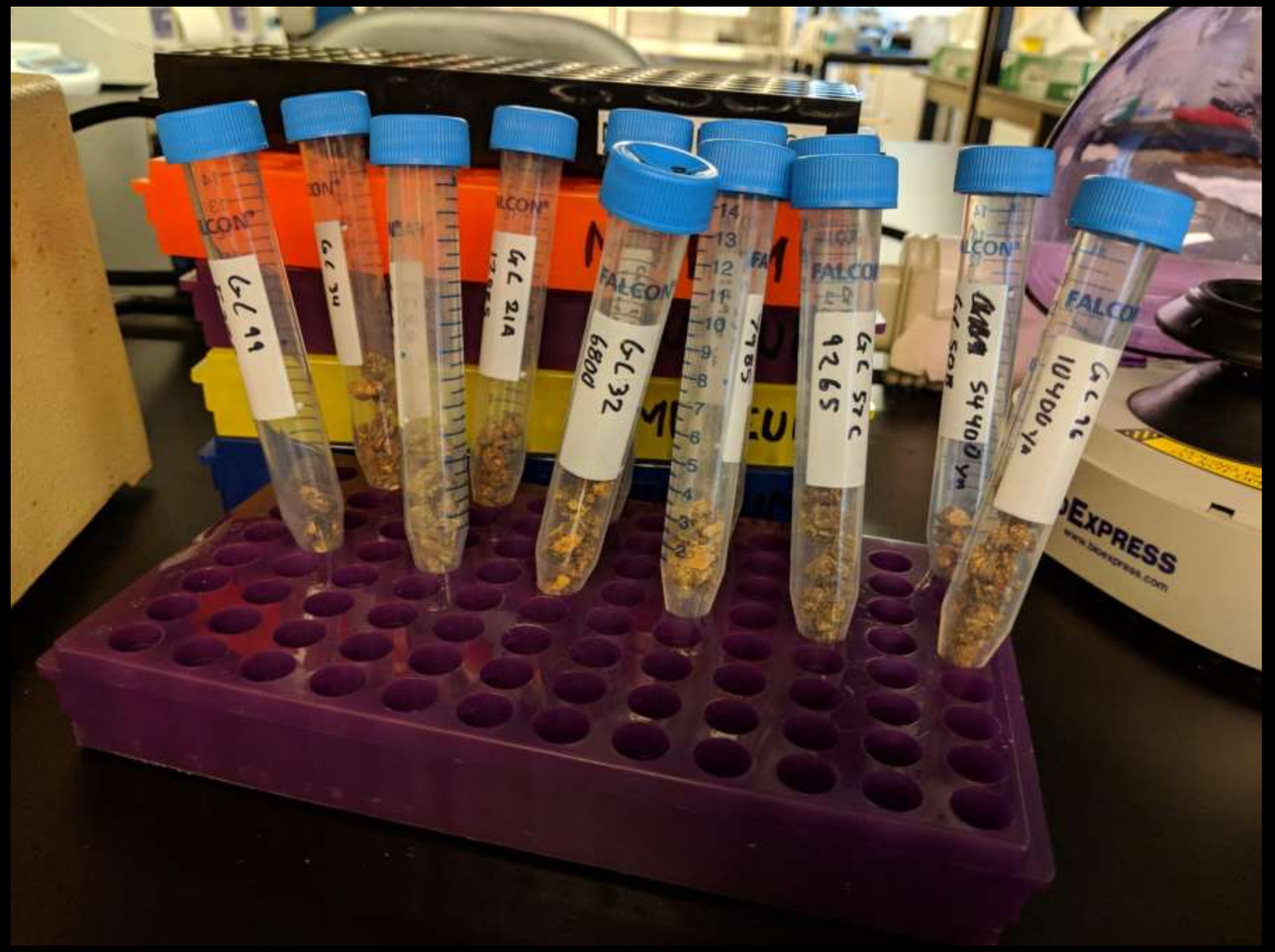
Photo Credit: J. Betancourt, K. Rylander (USGS)

City of Rocks
(COR)

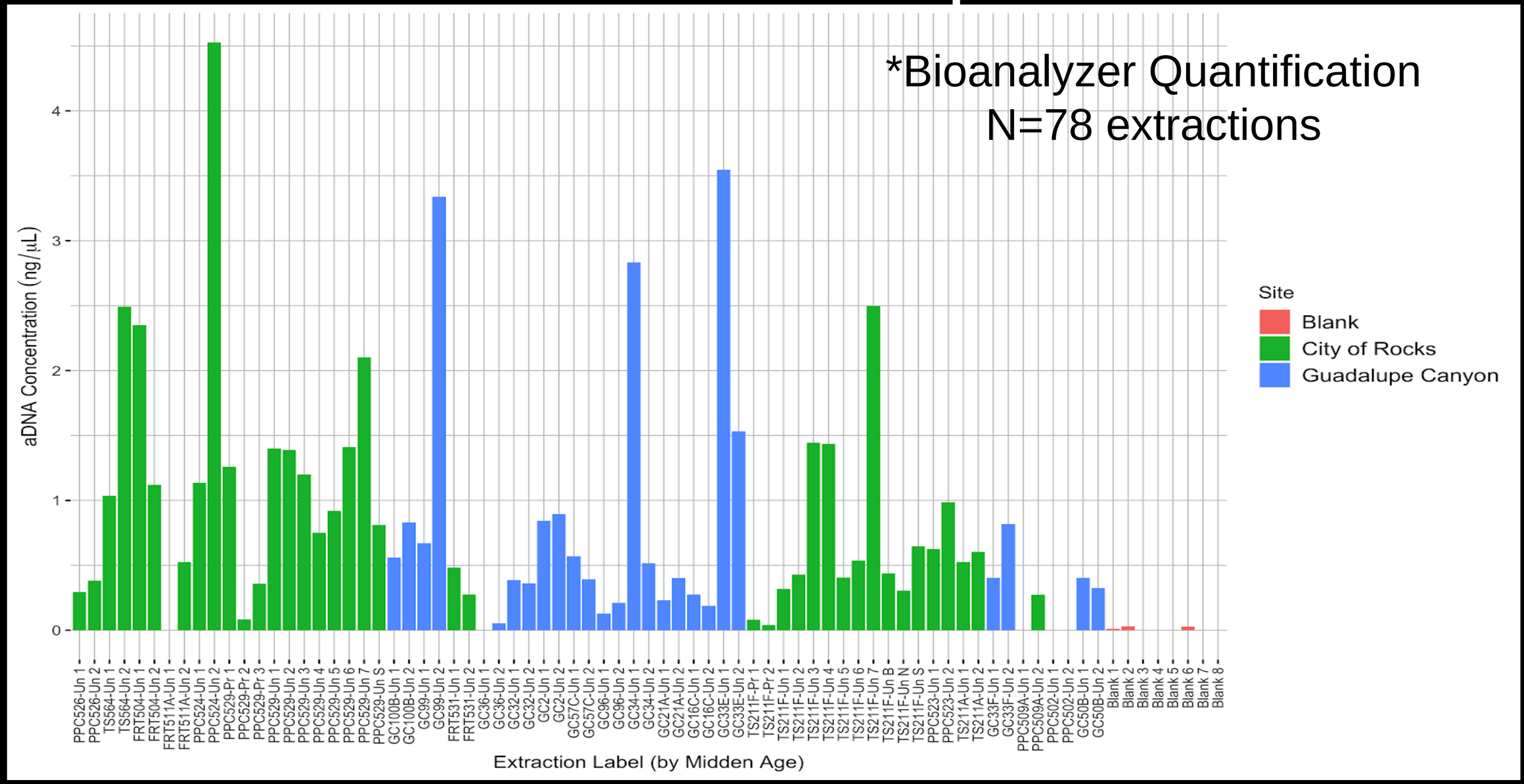
Guadalupe
Canyon
(GC)

aDNA Extraction Methods

- DNeasy PowerSoil® Kit protocol
- Measured resulting DNA concentration
  - Qubit® 2.0
  - Agilent 2100 Bioanalyzer



Ancient midden DNA quantification

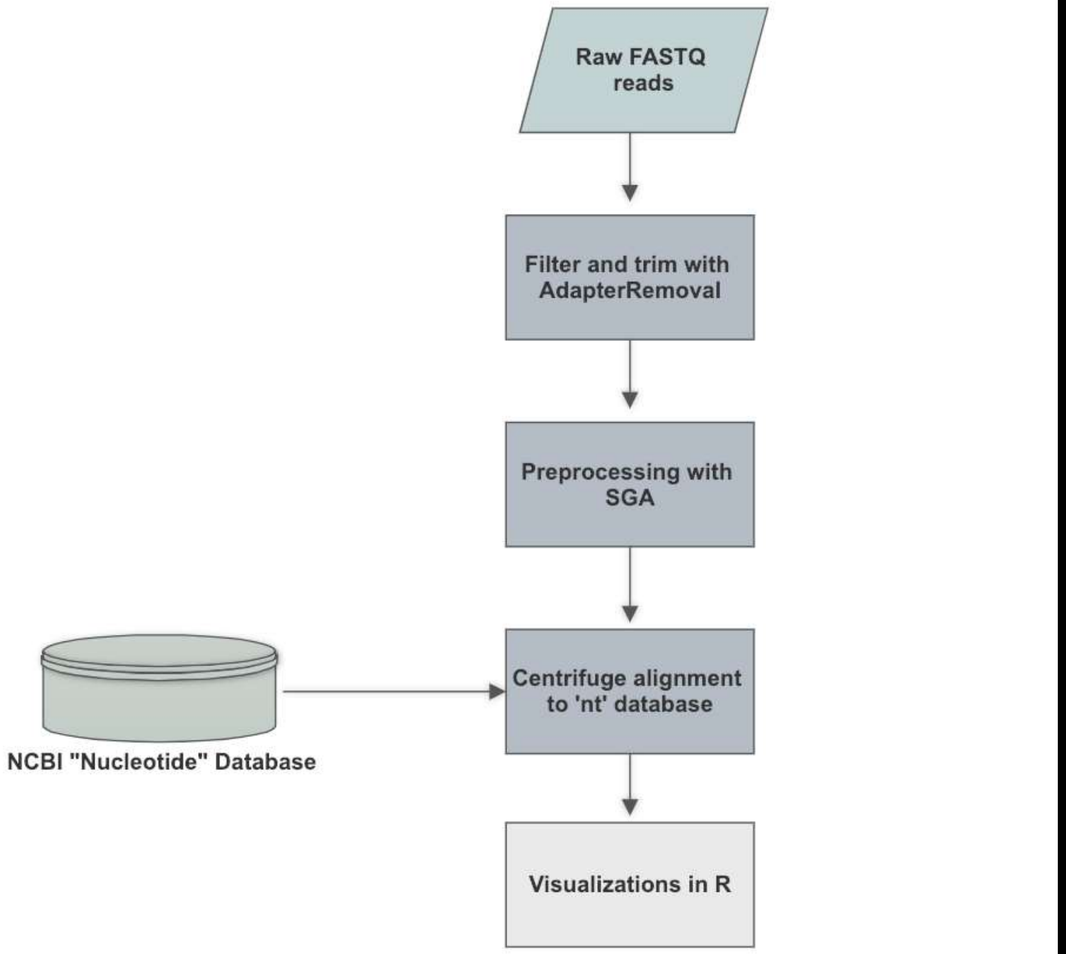


Sequencing

- Shotgun/Whole Genome
- Illumina HiSeq 2500, 2x125bp reads
- 22 samples submitted → 11 successful libraries
- ~30 – 60 million reads per sample

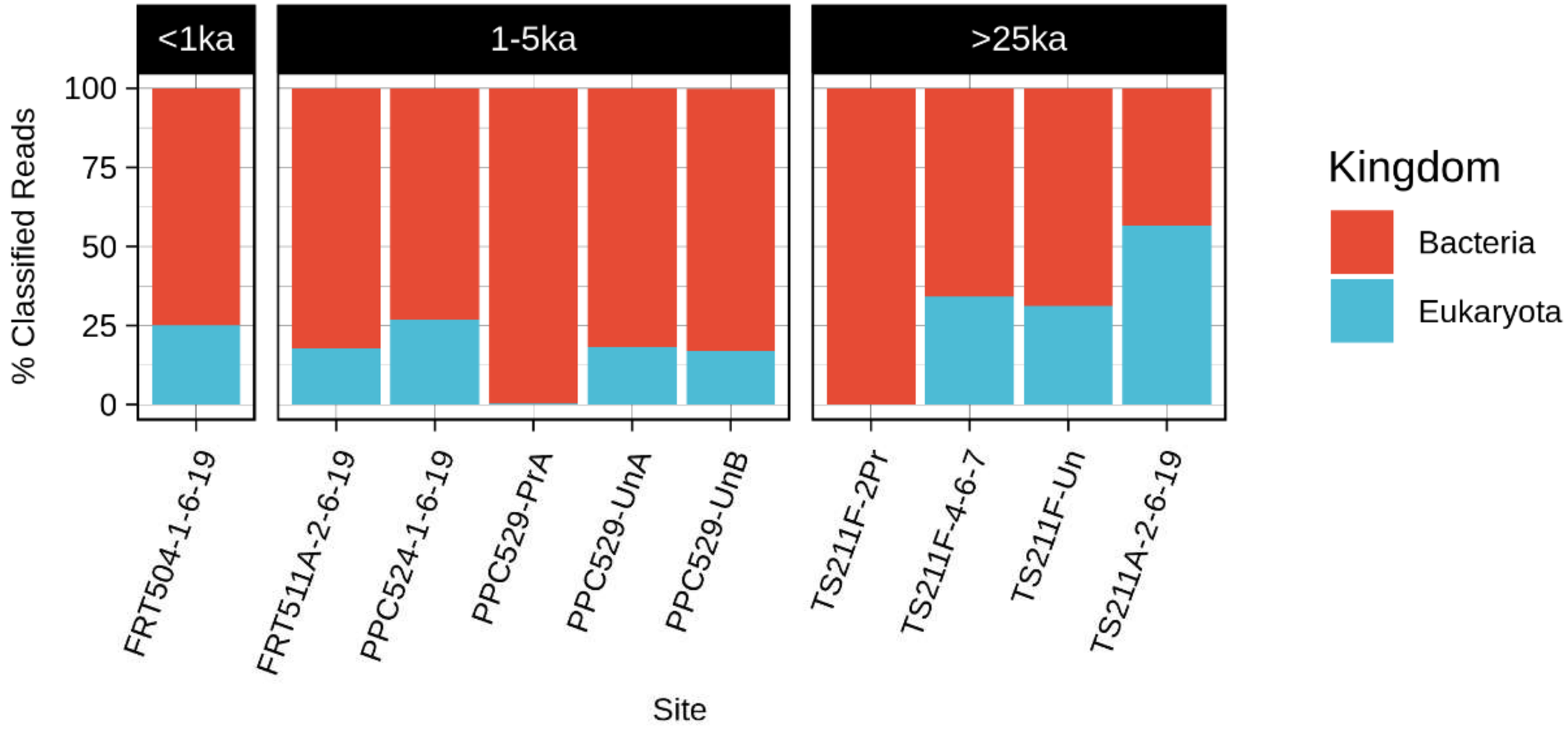
Metagenomics

Pipeline

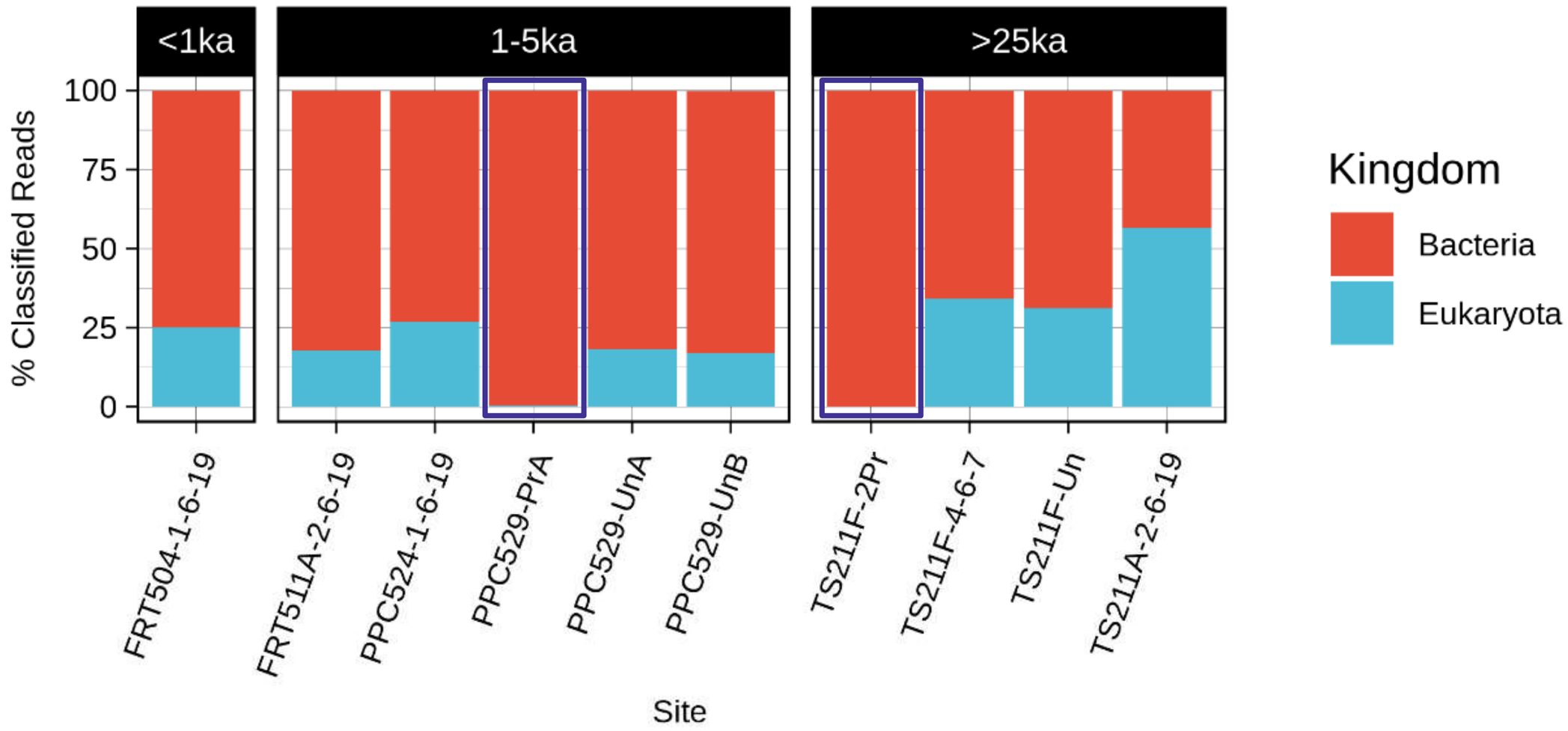


- Input: Raw Illumina ‘fastq’ files
- Data: HiSeq 2500, 2x125bp
- Output: Taxonomic classification of reads

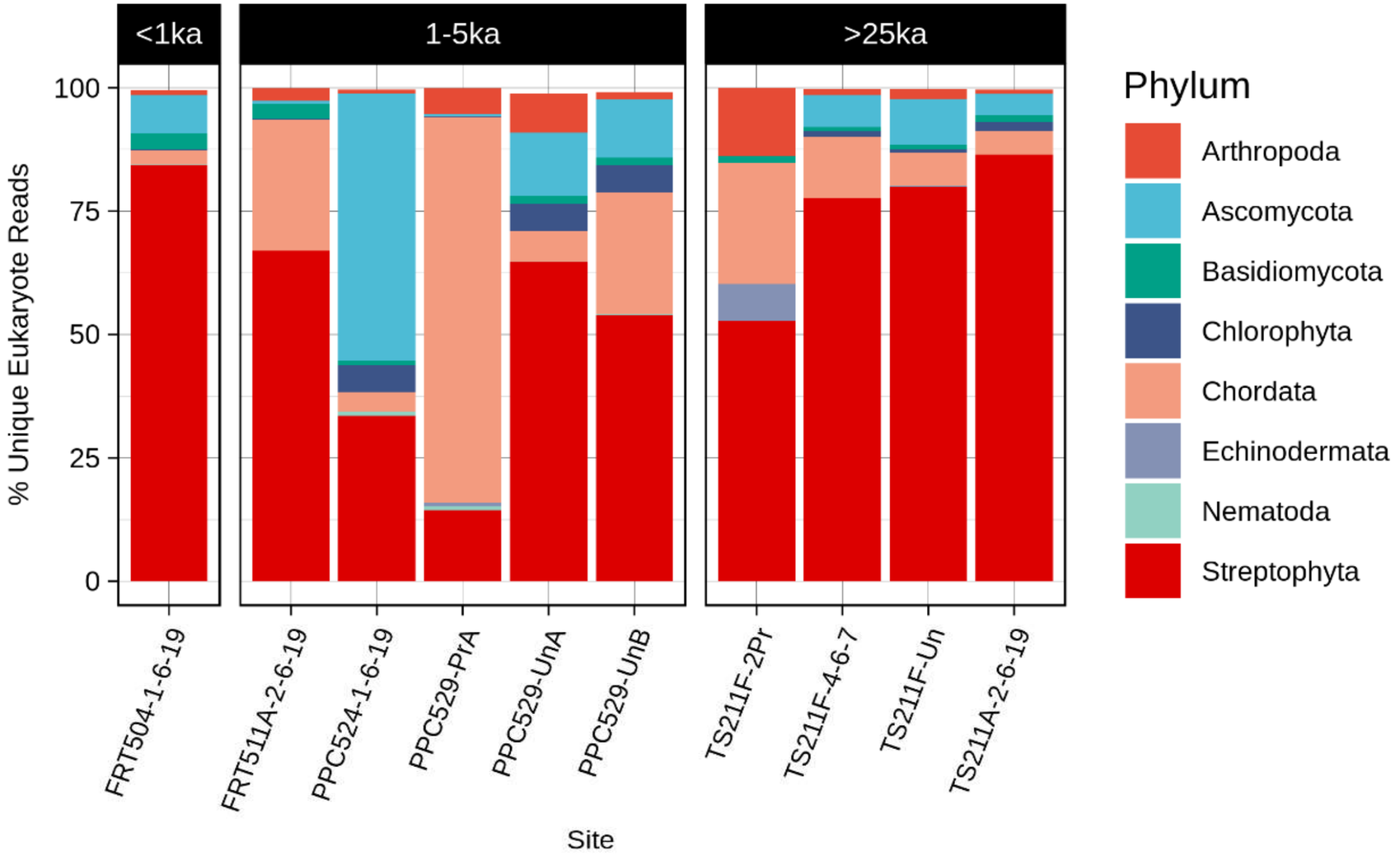
Taxonomic Classification



Taxonomic Classification



Eukaryotes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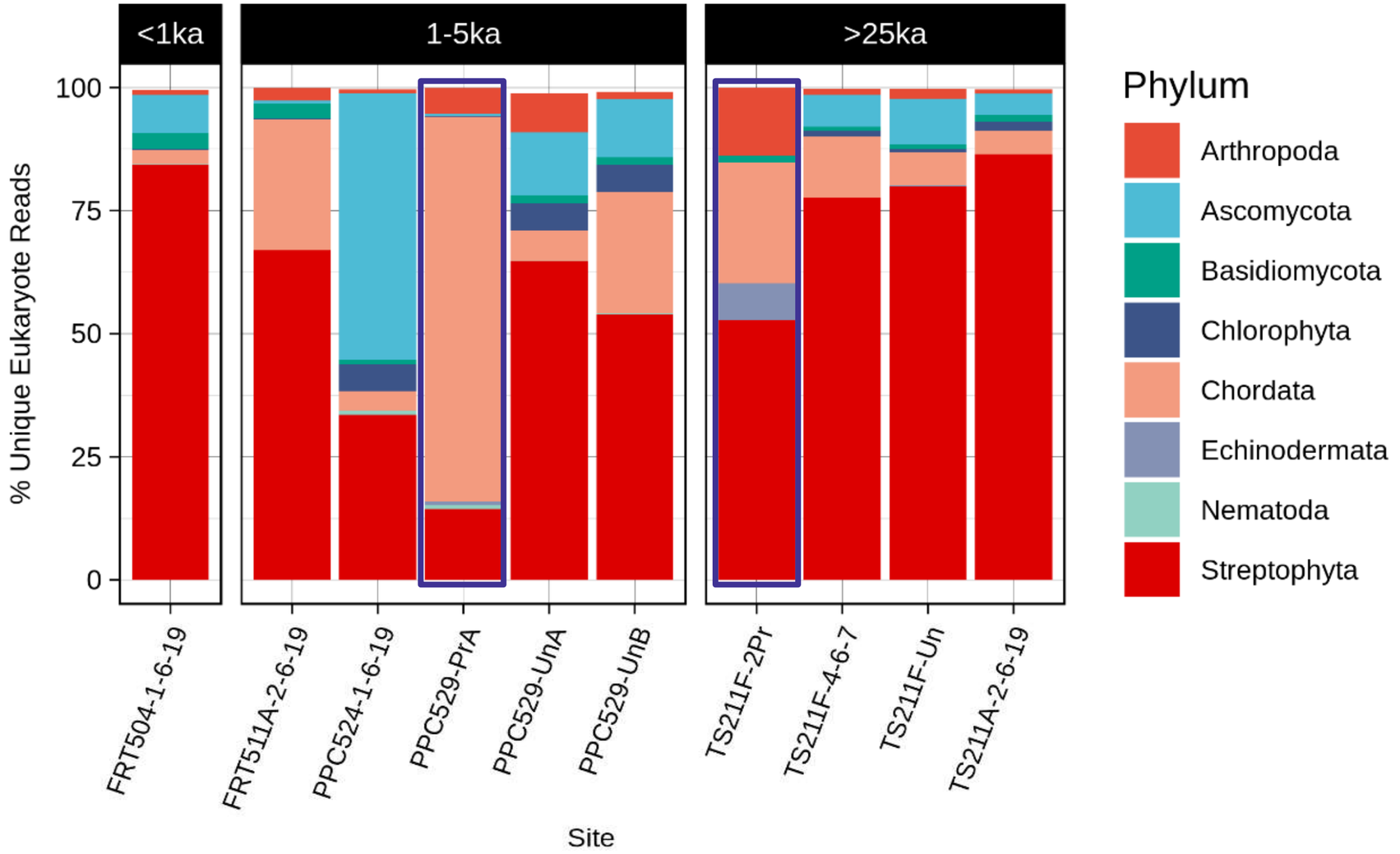
>25ka
% Unique Eukaryote Reads
100
75
50
25
0
FRT504-1-6-19
FRT511A-2-6-19
PPC524-1-6-19
PPC529-PrA
PPC529-UnA
PPC529-UnB
TS211F-2Pr
TS211F-4-6-7
TS211F-Un
TS211A-2-6-19
Site
Phylum
Arthropoda
Ascomycota
Basidiomycota
Chlorophyta
Chordata
Echinodermata
Nematoda
Streptophyta

Eukaryotes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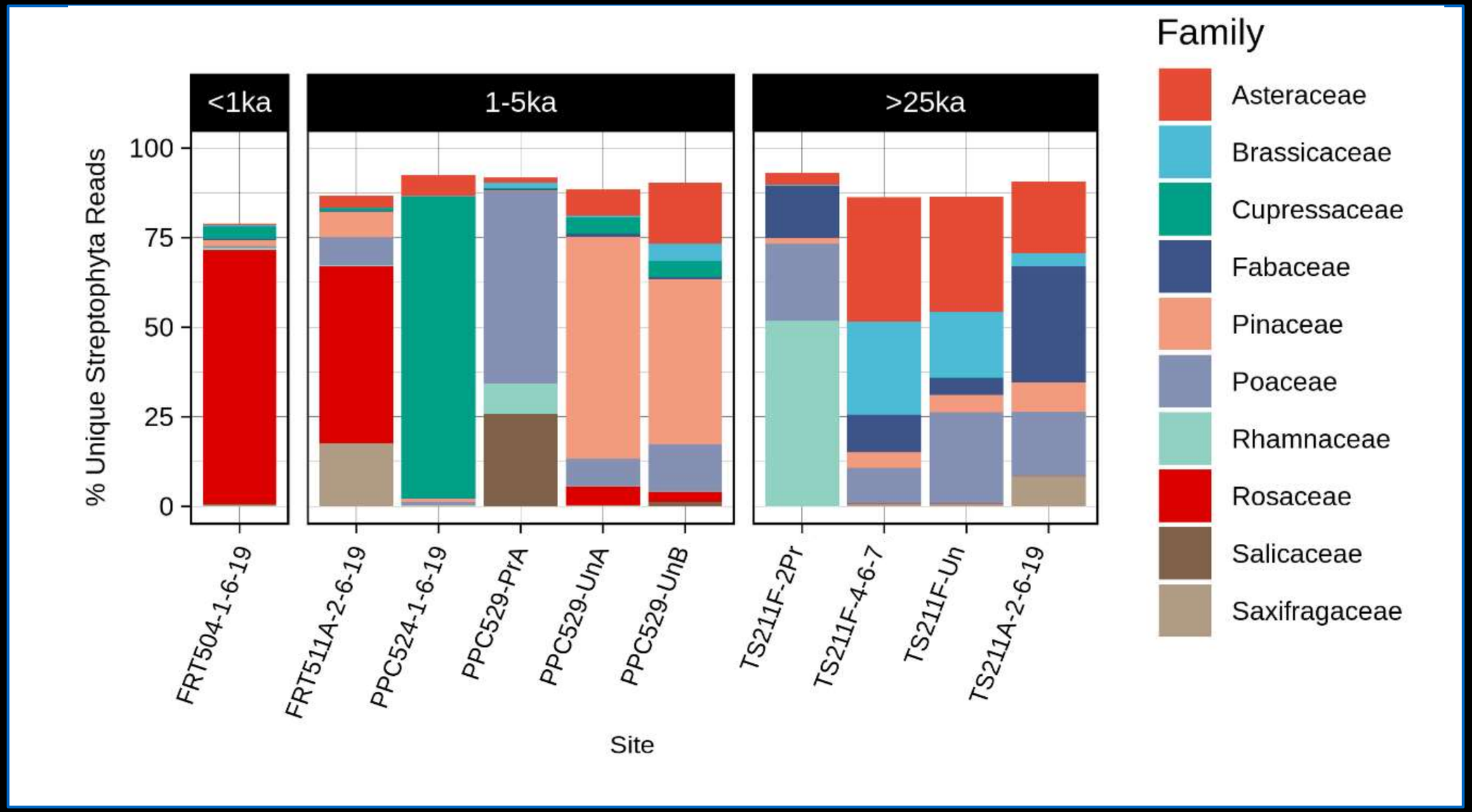
1-5ka
>25ka
% Unique Eukaryote Reads
100
75
50
25
0
FRT504-1-6-19
FRT511A-2-6-19
PPC524-1-6-19
PPC529-PrA
PPC529-UnA
PPC529-UnB
TS211F-2Pr
TS211F-4-6-7
TS211F-Un
TS211A-2-6-19
Site
Phylum
Arthropoda
Ascomycota
Basidiomycota
Chlorophyta
Chordata
Echinodermata
Nematoda
Streptophyta

Plants


<1ka
1-5ka
>25ka
% Unique Streptophyta Reads
100
75
50
25
0
FRT504-1-6-19
FRT511A-2-6-19
PPC524-1-6-19
PPC529-PrA
PPC529-UnA
PPC529-UnB
TS211F-2Pr
TS211F-4-6-7
TS211F-Un
TS211A-2-6-19
Site
Family
Asteraceae
Brassicaceae
Cupressaceae
Fabaceae
Pinaceae
Poaceae
Rhamnaceae
Rosaceae
Salicaceae
Saxifragaceae

Top Plant Genera: Hints of A Changing Ecosystem

3,260 year-old midden

Pinus

Triticum

Diplostephium

Juniperus

Cercocarpus

28,460 year old midden

Triticum

Diplostephium

Lupinus

Poa

Artemisia

Images: nps.gov, en.wikipedia.org

Still working on:

- Amplicon Sequencing comparison
- Analysis of DNA damage patterns → Attempt to confirm ancient origins
- Evaluation of non-plant data

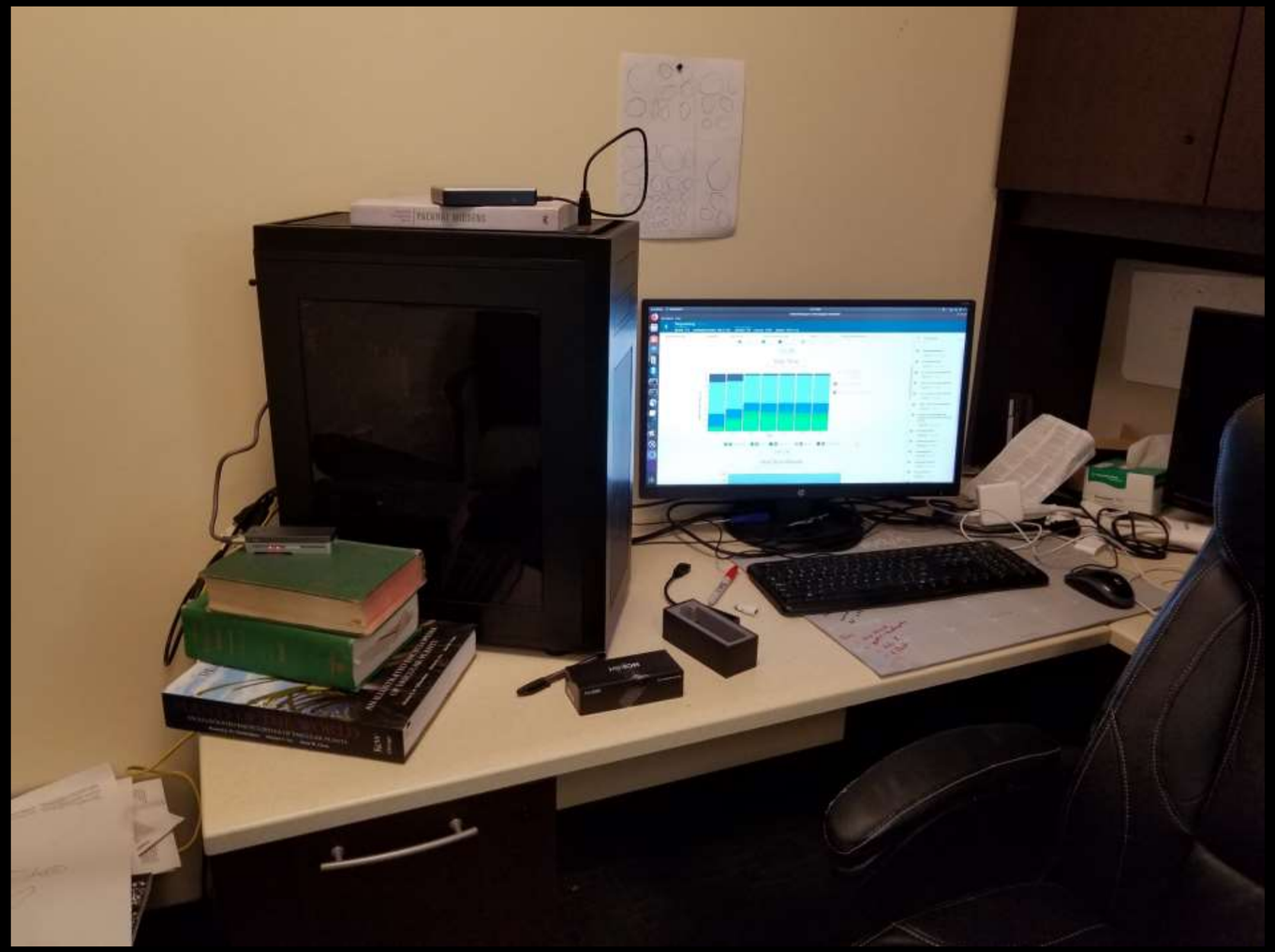
Current teaching/research topics

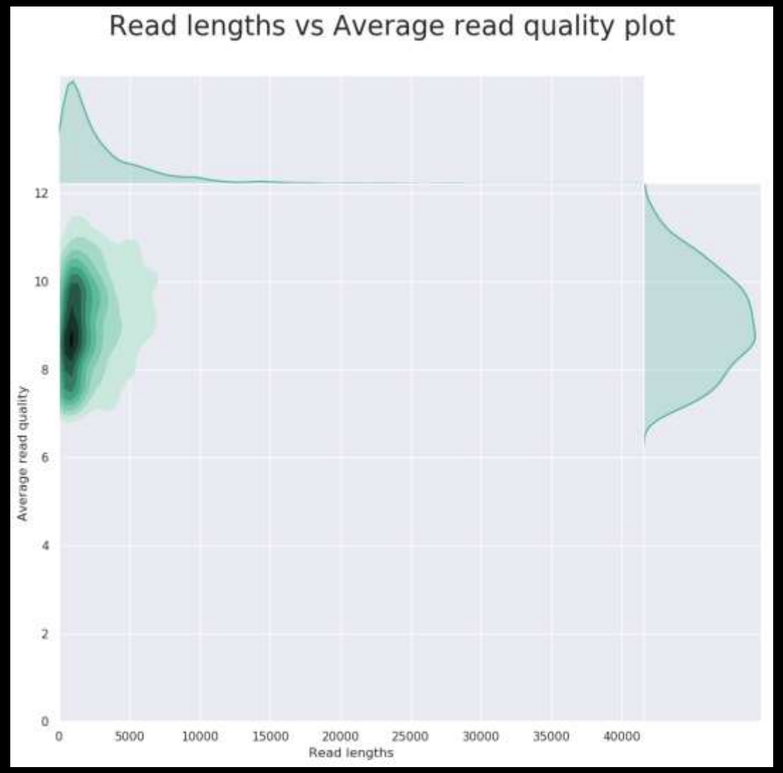
Teaching & Research

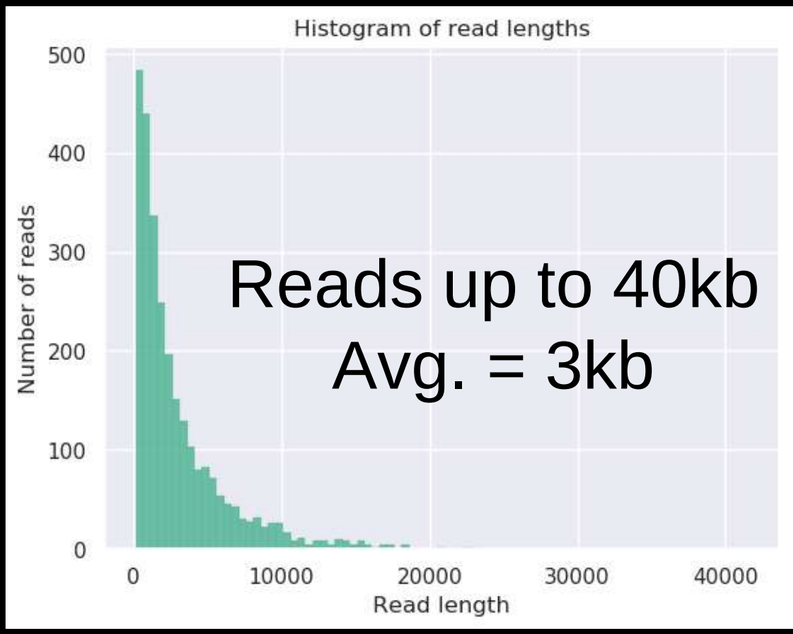
- Developing UG Bioinformatics courses
  - https://rsh249.github.io/bioinformatics
  - https://rsh249.github.io/applied_bioinformatics
- Oxford Nanopore MinION
- eDNA – Detecting plant communities from aquatic environmental DNA

https://pubs.usgs.gov/tm/02/a13/tm2a13.pdf

MinION: *E. coli*







~5000 reads in 6hrs = ~95% E. coli

Acknowledgements

Stonehill College
Alex Baryiames, Jordan Callahan, Caroline Pitten, Andrew Williams, Patrick O'Shea

Magda Pederson
Irvin Pan

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AMNH
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Cornell University
Kevin Nixon, Bill Crepet, Jeffrey Doyle, Thereis Choo, Daniella Allevato, Avery Hill

USGS — Packrat middens
Julio Betancourt
Packrat Midden Futures Working Group

Links

- These Slides: https://rsh249.github.io/files/harbert_seminar_UMassD_3_20_19.pdf

- Papers:
  - CRACLE https://bsapubs.onlinelibrary.wiley.com/doi/full/10.3732/ajb.1400500
  - Packrat Paleoclimate https://www.openquaternary.com/articles/10.5334/oq.46
  - Packrat aDNA **Coming soon to bioRxiv!**

- Courses:
  - Introduction to Bioinformatics – https://rsh249.github.io/bioinformatics
  - Applied Bioinformatics (Nanopore) – https://rsh249.github.io/applied_bioinformatics