

MPI-SHH SUMMER SCHOOL 2021

Doorway to Human History

Microbial Archaeology

Dr. Irina Velsko and Zandra Fagernäs

Who are we?

Irina

- Has a Ph. D. in microbiology
- Works with dental calculus
- Also knows about living microbes
- Genetics, proteomics, metabolomics

Zandra

- Is trying to get a Ph. D. in microbiology
- Works with dental calculus
- Does a lot of lab work
- Genetics, proteomics

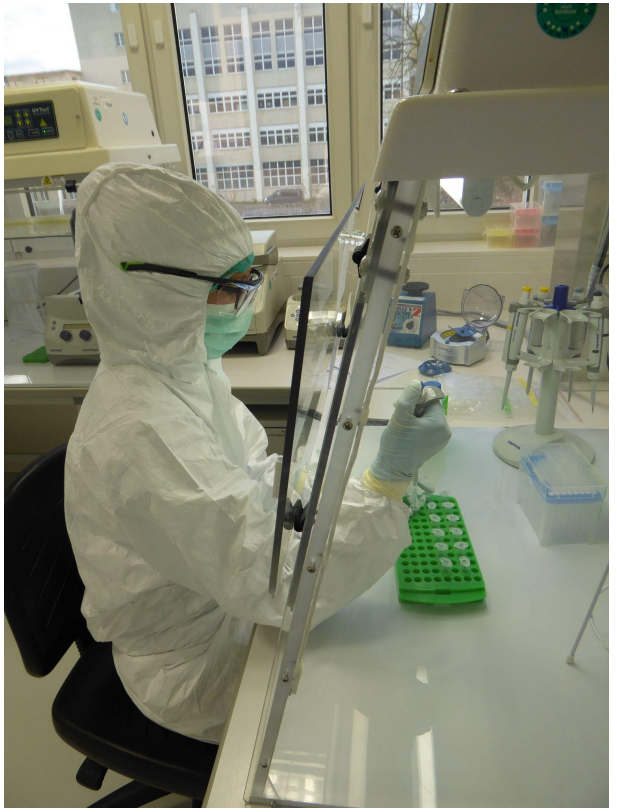
Is anyone working with ancient microbes?
Planning a project with ancient microbes?
Think ancient microbes are cool?

Write in the chat!

Why study microbes?

Where can you find ancient microbes?

Microbes are everywhere!!!



Disease

Past pathogens

- Changes in human lifestyle exposed us to new pathogens
  - Domesticated animals
  - Higher population densities
  - Mobility
- Epidemics and disease outbreaks have been recorded
  - Cause is often unknown
- Traditionally researched through palaeopathology of skeletal assemblages
- In the 1990s, aDNA came into the picture
- First genome published in 2011 (*Yersinia pestis*)

What pathogens?

Bacteria

- *Brucella melitensis* (brucellosis)
- *Gardnerella vaginalis* (bacterial vaginosis)
- *Helicobacter pylori* (GI ulcers)
- *Mycobacterium leprae* (leprosy)
- *Mycobacterium tuberculosis* (tuberculosis)
- *Tannerella forsythia* (periodontal disease)
- *Treponema pallidum* (syphilis/jaws/bejel)
- *Vibrio cholerae* (cholera)

What pathogens?

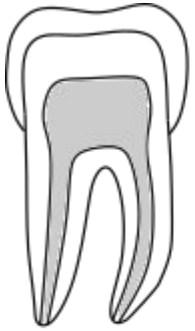
Viruses

- HBV (viral hepatitis)
- HIV (AIDS)
- Influenza virus (influenza)
- VARV (smallpox)

Eukaryotes

- *Phytophthora infestans* (potato blight)
- *Plasmodium falciparum* and *Plasmodium vivax* (malaria)

Where can we find pathogens?

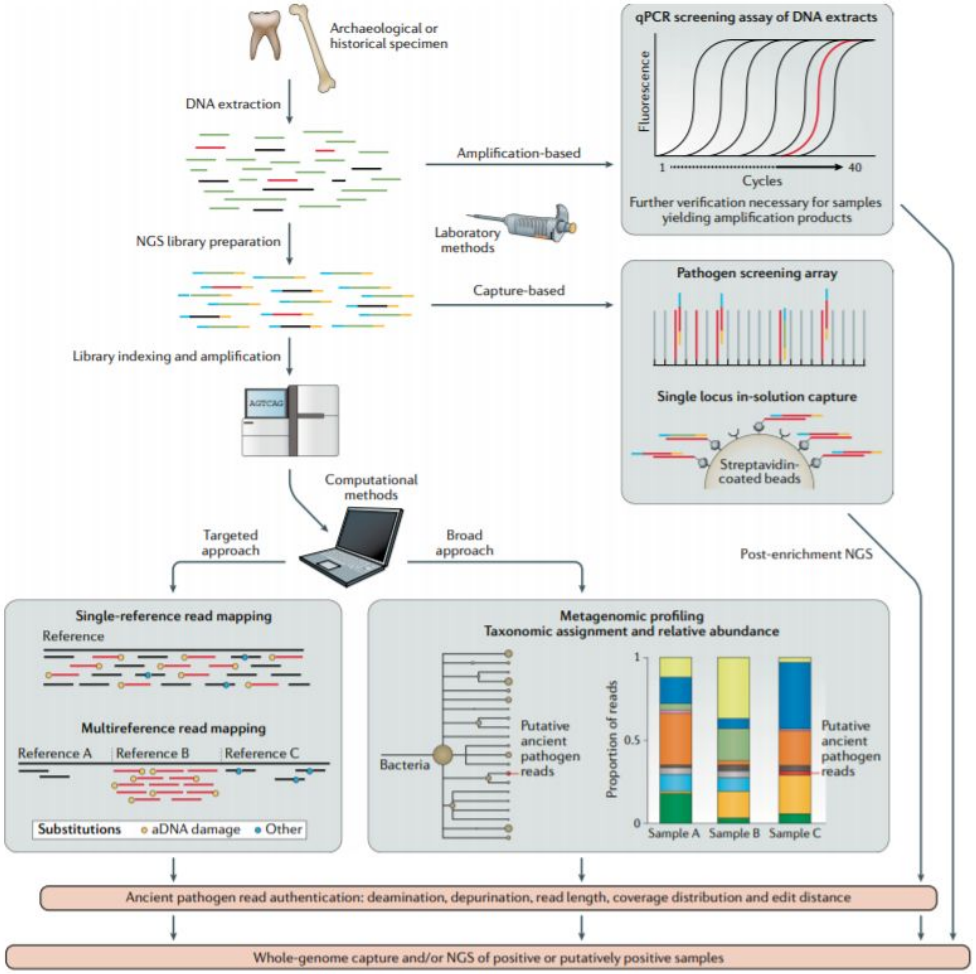


Pulp cavity

Bones

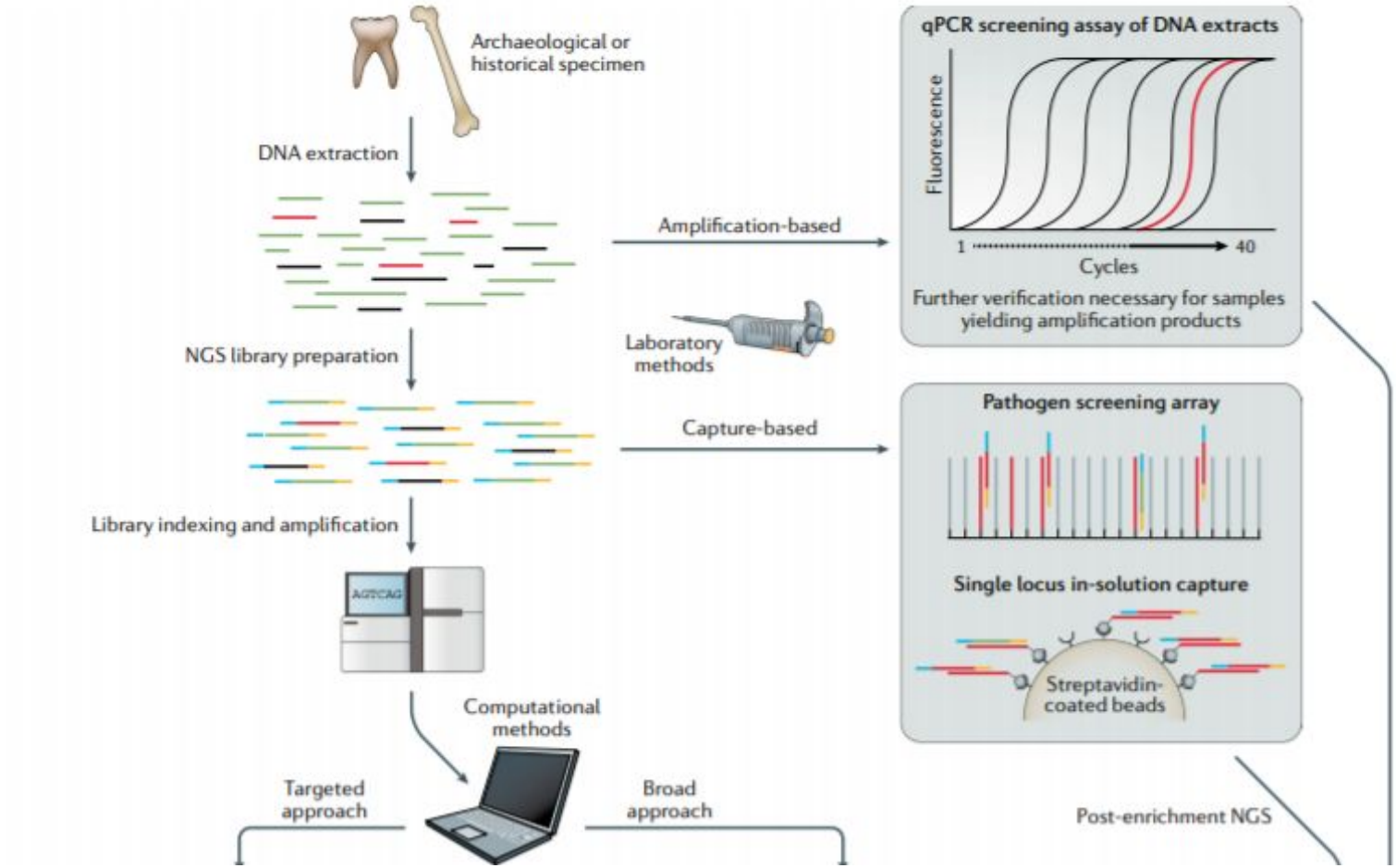
Skeletal lesions

Ancient pathogens



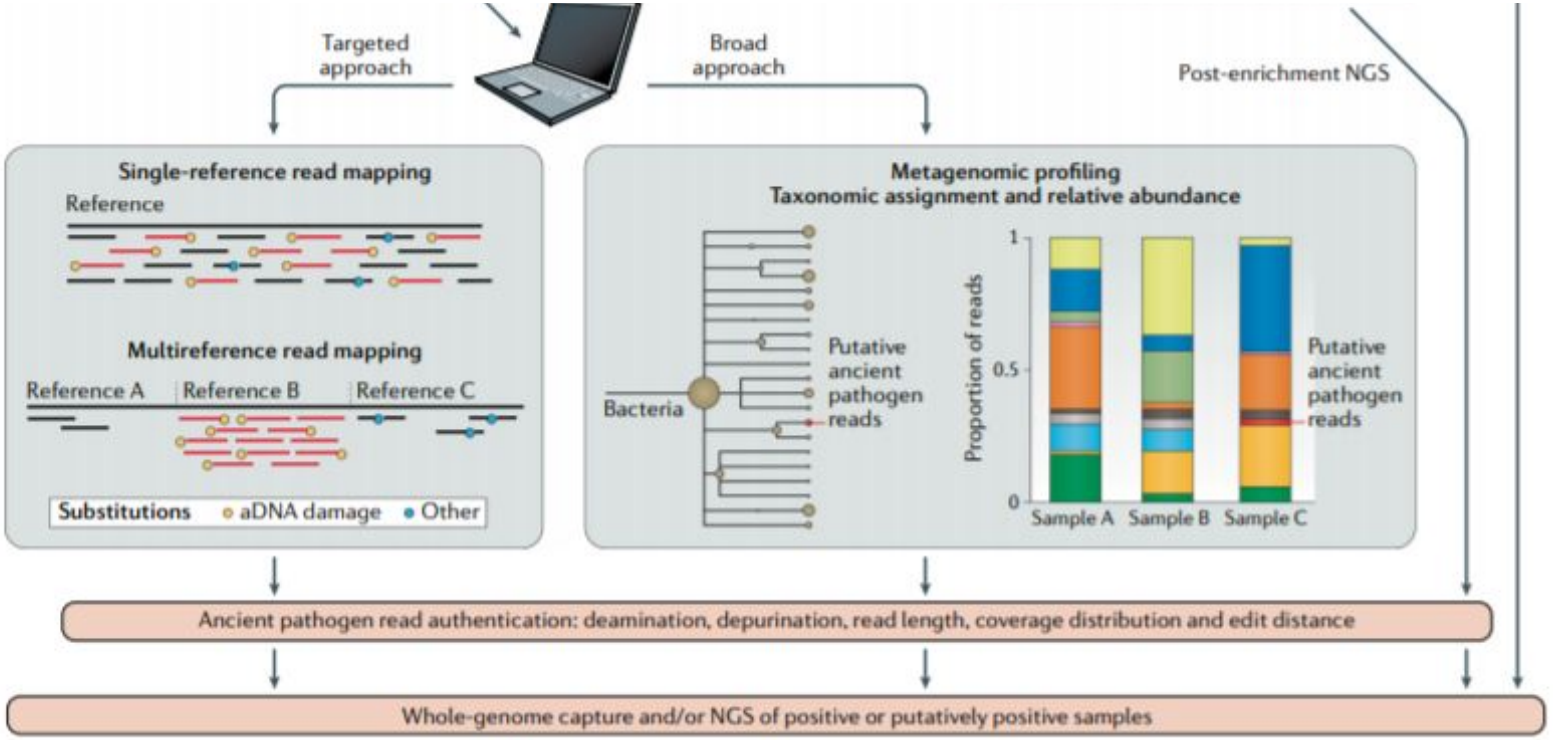
Spyrou *et al*. 2019 Nat Rev Genet.

Ancient pathogens



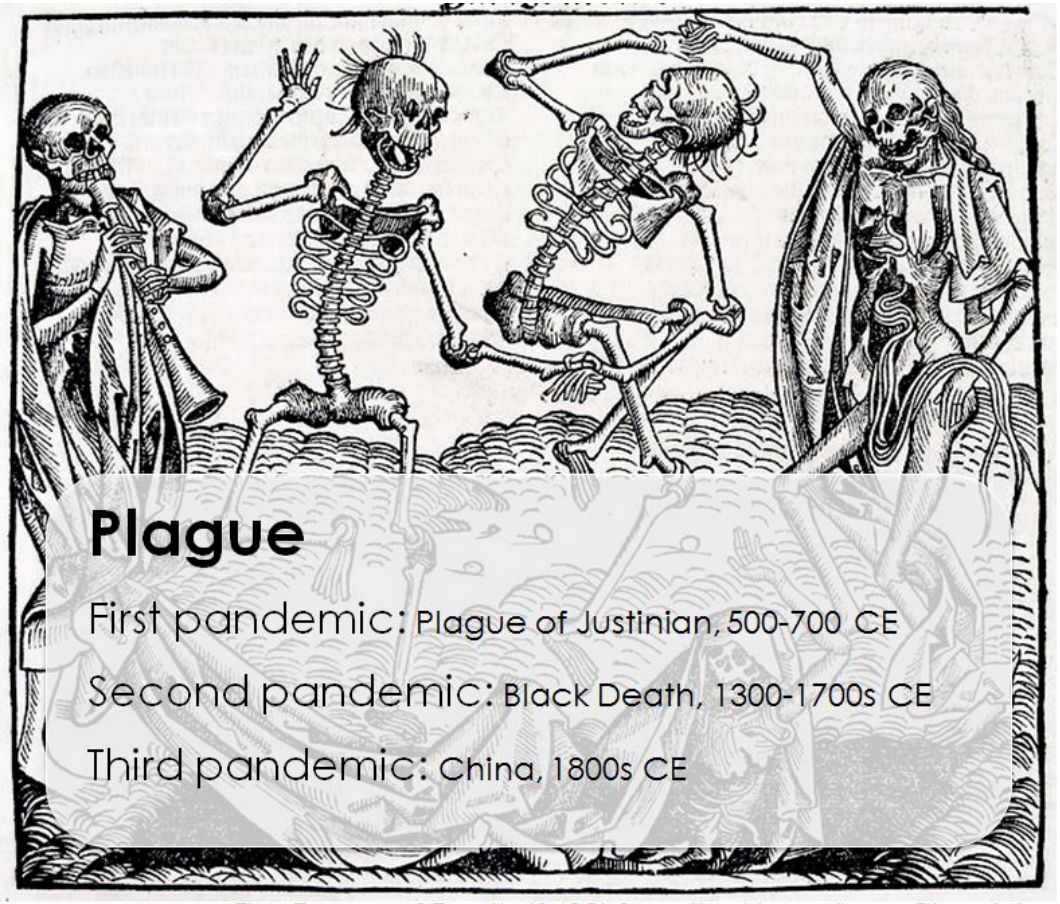
Spyrou *et al*. 2019 Nat Rev Genet.

Ancient pathogens



Spyrou *et al*. 2019 Nat Rev Genet.

1. Plague
2. Cocolitzli



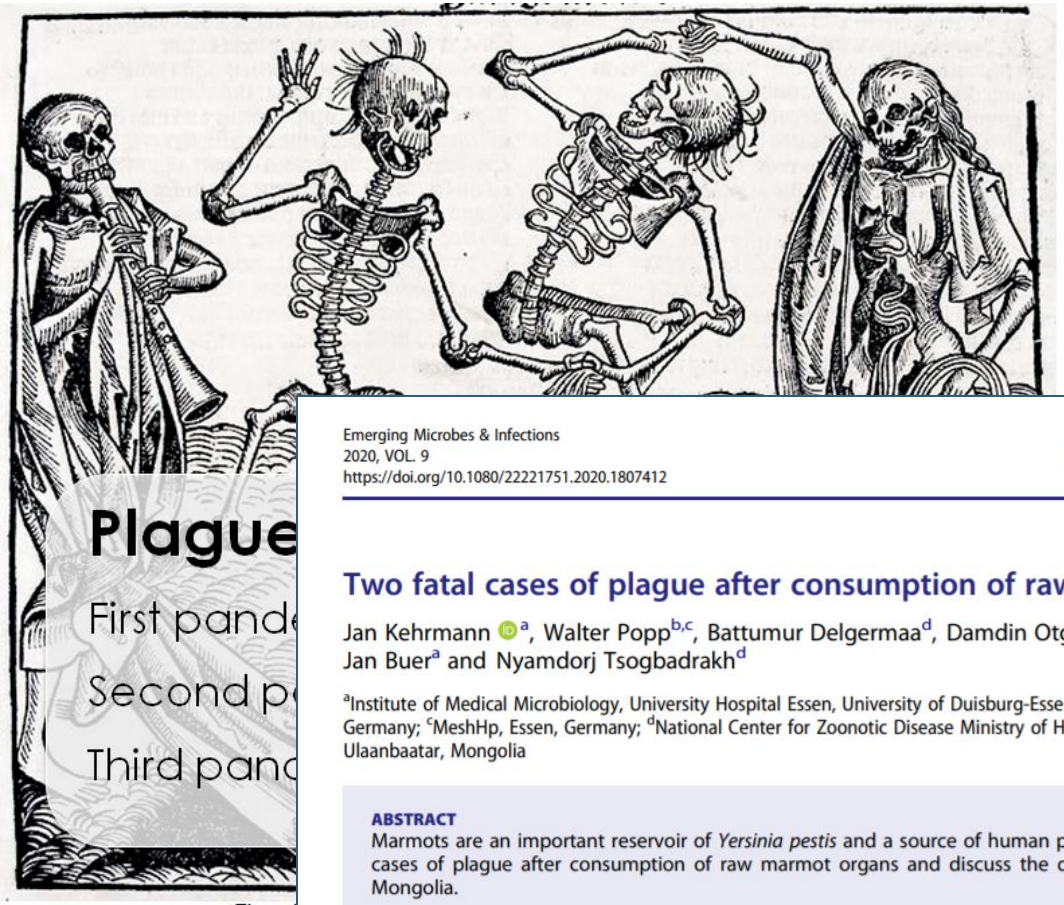
Plague

First pandemic: Plague of Justinian, 500-700 CE

Second pandemic: Black Death, 1300-1700s CE

Third pandemic: China, 1800s CE

The Dance of Death (1493) from the Nuremberg Chronicle



Plague

First pand

Second p

Third panc

The

Emerging Microbes & Infections
2020, VOL. 9
https://doi.org/10.1080/22221751.2020.1807412

EMi Taylor & Francis
Taylor & Francis Group

OPEN ACCESS Check for updates

Two fatal cases of plague after consumption of raw marmot organs

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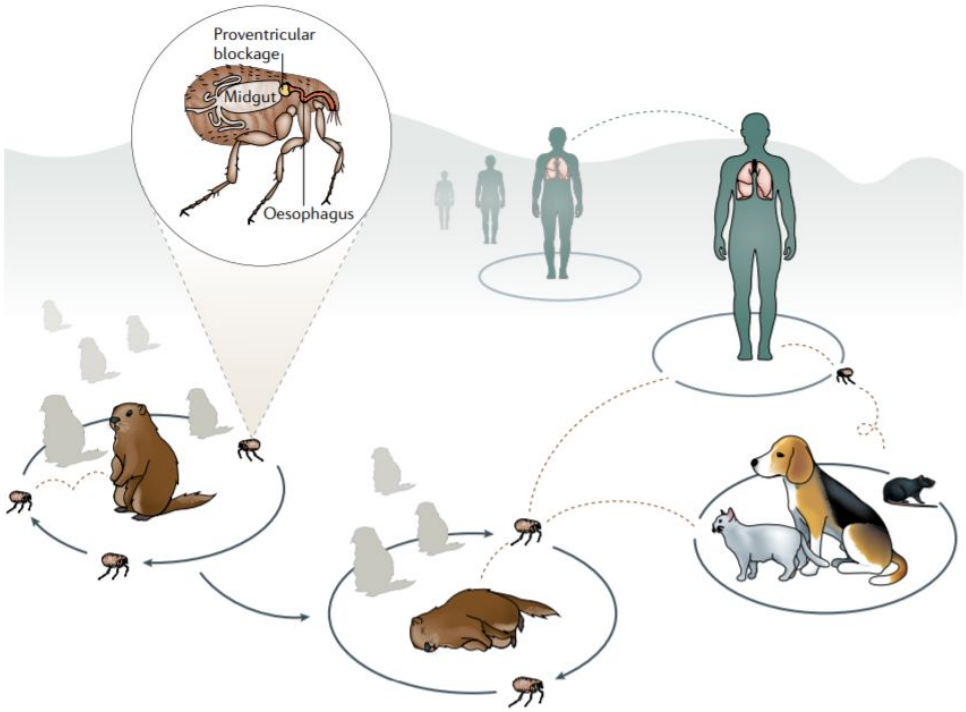
ABSTRACT
Marmots are an important reservoir of *Yersinia pestis* and a source of human plague in Mongolia. We present two fatal cases of plague after consumption of raw marmot organs and discuss the distribution of natural foci of *Y. pestis* in Mongolia.

ARTICLE HISTORY Received 12 May 2020; Revised 25 July 2020; Accepted 5 August 2020

KEYWORDS Plague; Mongolia; marmot; epidemiology; sepsis

Yersinia pestis

- Not human-adapted
- Maintained in wild rodents
  - Still has active foci
- Transmitted by fleas
  - Blocks gut
  - Flea feeds more



Spyrou *et al*. 2019 Nat Rev Genet.

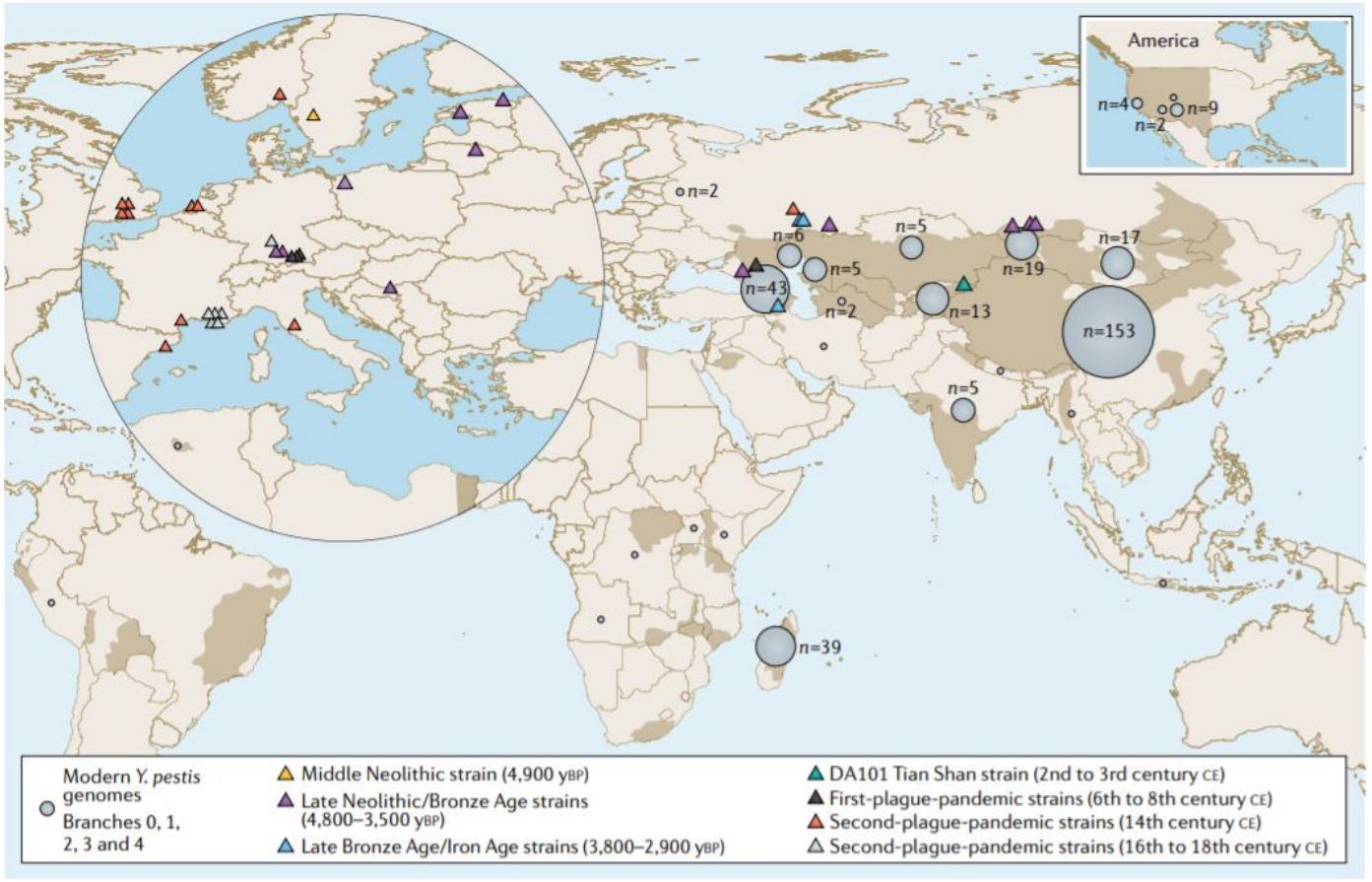
Plague in humans

1. Bubonic
    - Most common
    - Bacteria travel to lymph nodes, cause ‘buboes’
    - Up to 60% mortality if untreated

2. Pneumonic
    - Bacteria have travelled to the lungs
    - Human-to-human transmission
    - Nearly to 100% mortality if untreated

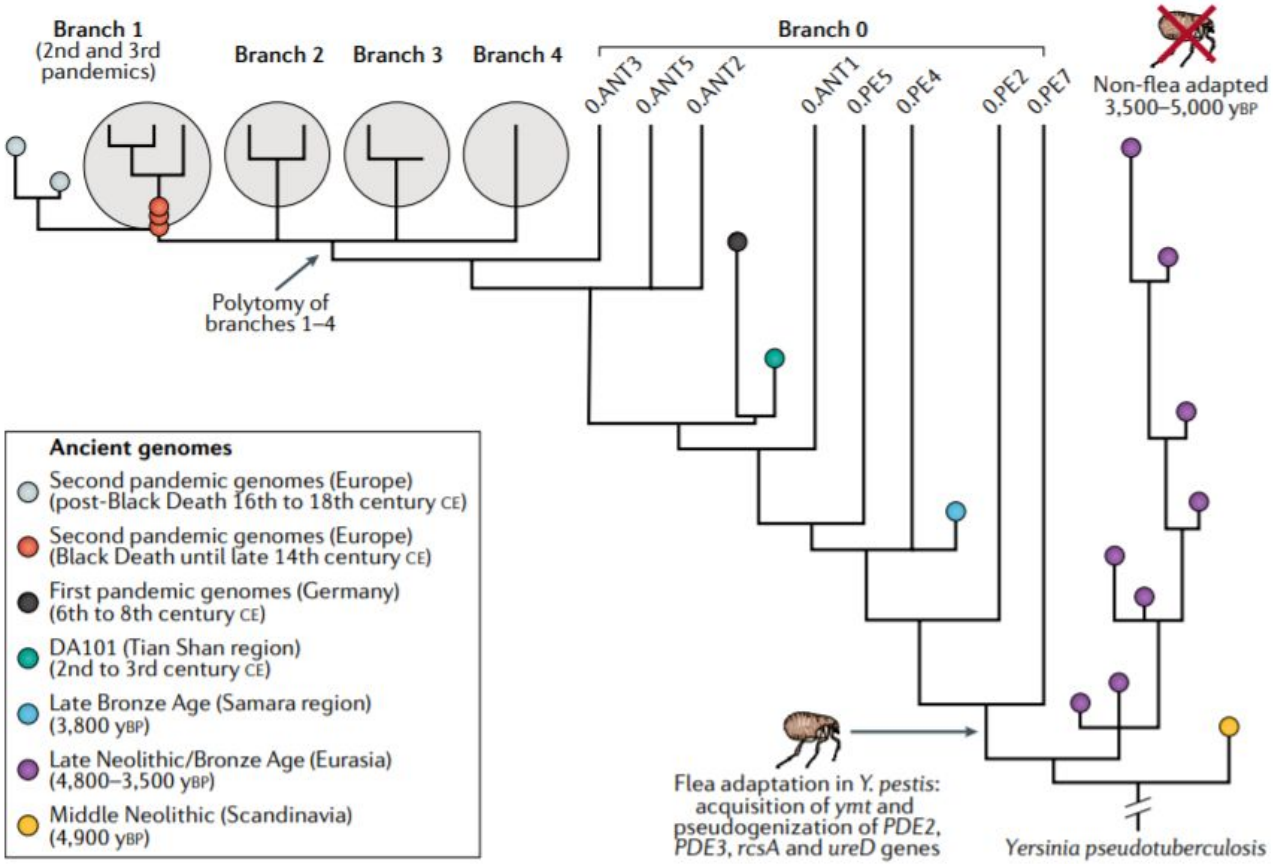
3. Septicaemic
    - Spreading in the bloodstream
    - Nearly to 100% mortality if untreated

Yersinia pestis genomes



Spyrou *et al*. 2019 Nat Rev Genet.

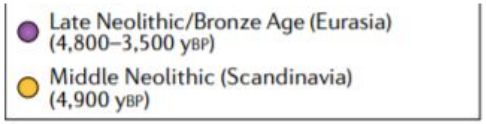
Yersinia pestis aDNA

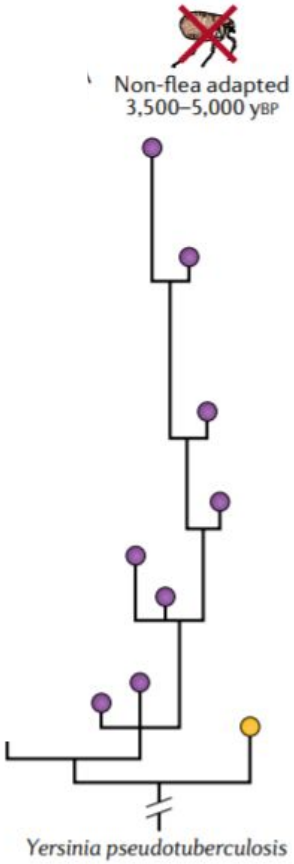


Spyrou *et al.* 2019 Nat Rev Genet.

Yersinia pestis aDNA

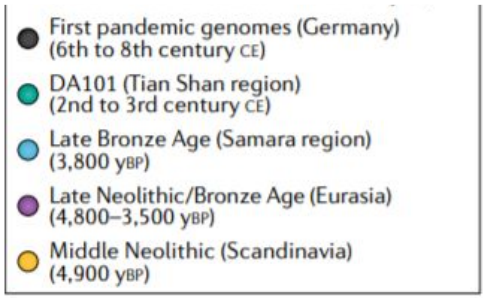
- Not flea-adapted
  - Not bubonic
- How was plague transmitted?
- What was the host population?
- Spread mirrors human migrations

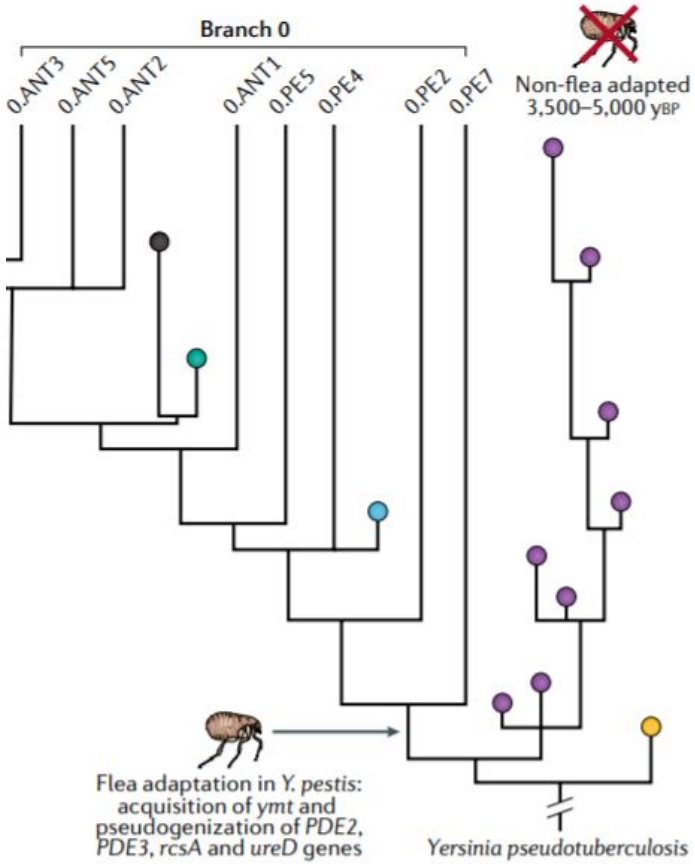




Spyrou *et al.* 2019 Nat Rev Genet.

Yersinia pestis aDNA





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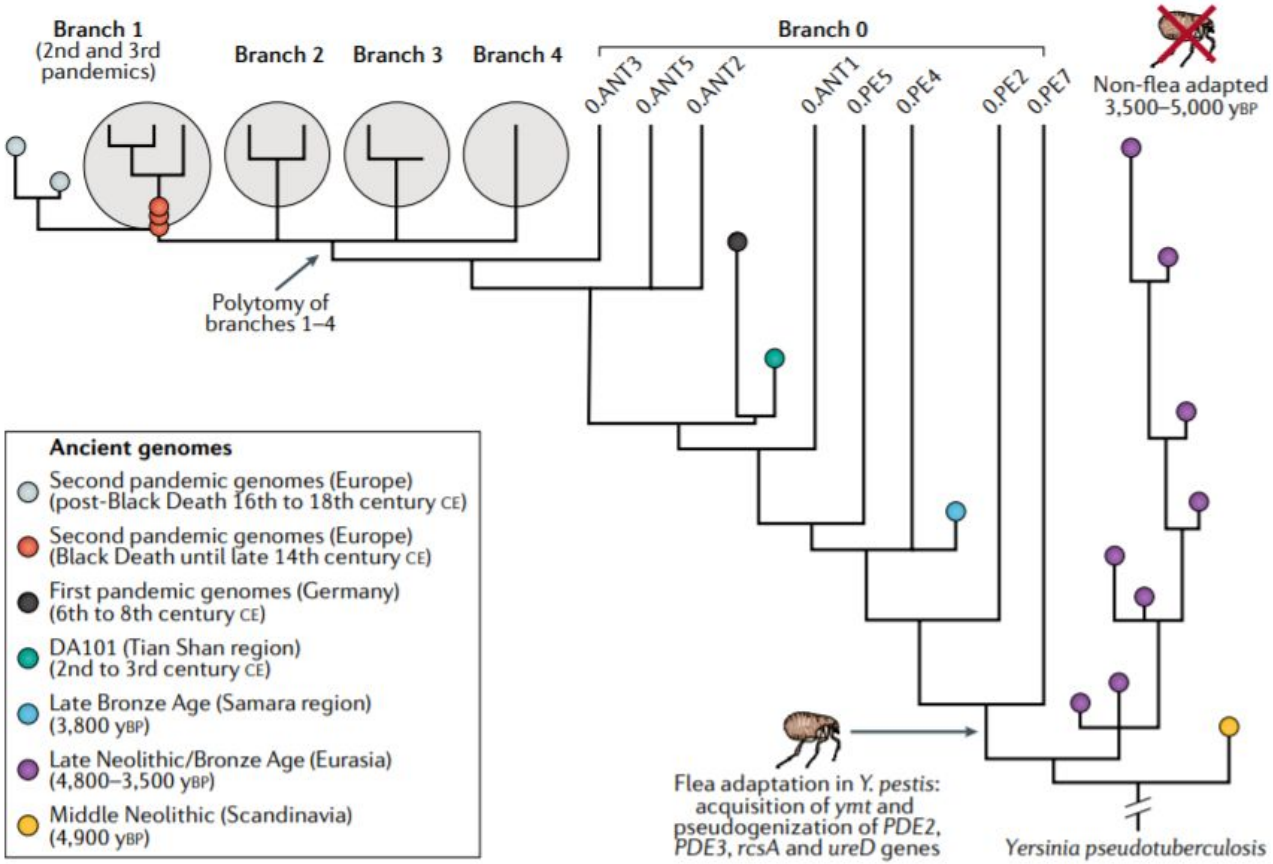
Yersinia pestis aDNA



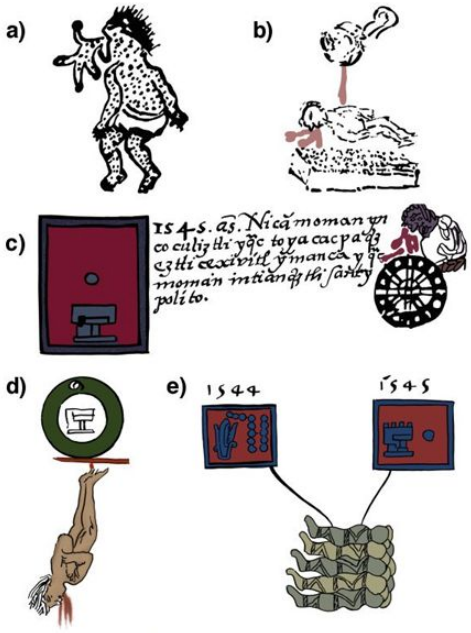
Spyrou *et al.* 2019 Nat Rev Genet.

Yersinia pestis summary

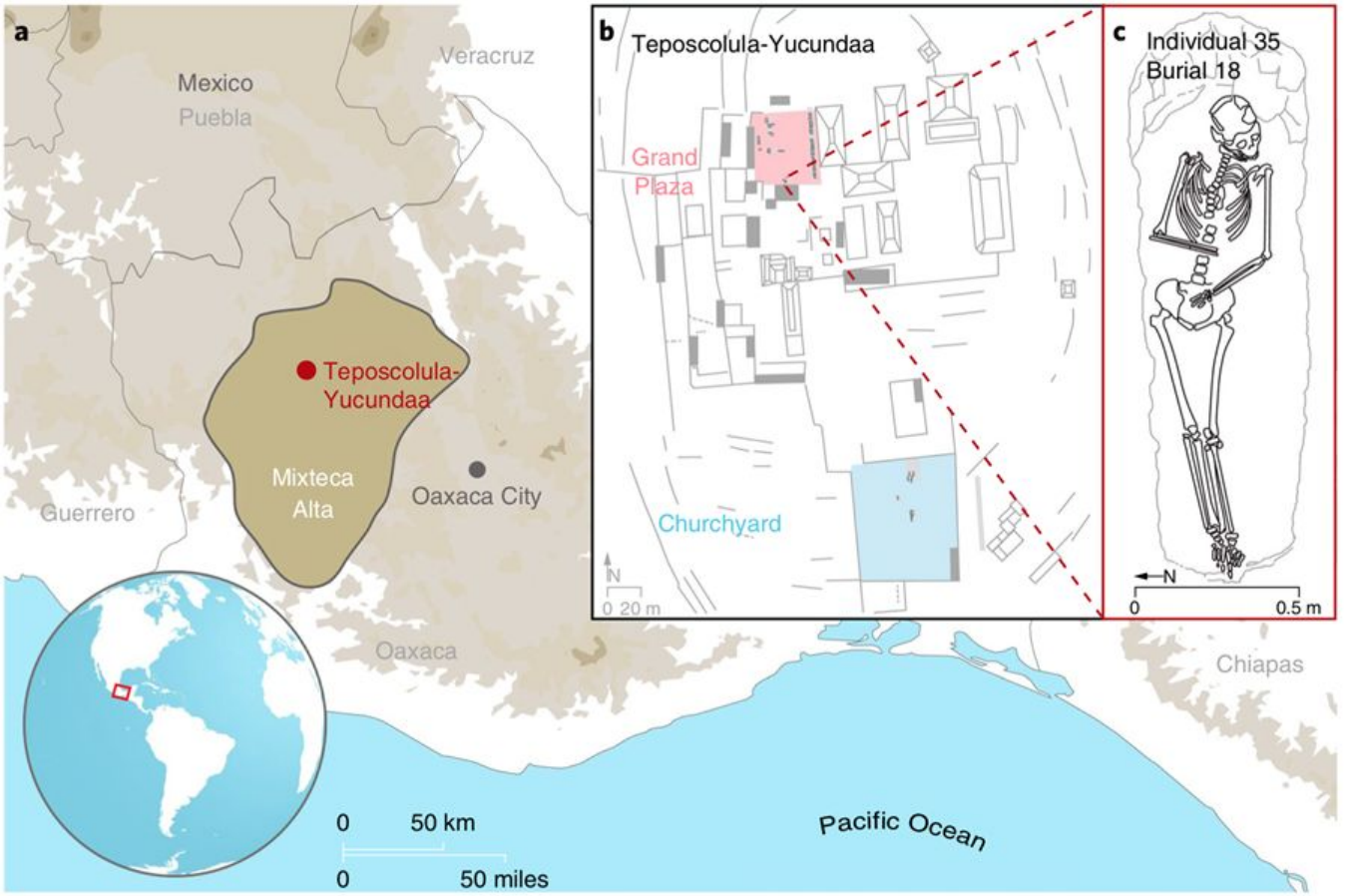
- By studying ancient genomes we have learned about
  - Transmission
  - Virulence
  - Evolution

Cocolitzli

- Poorly understood epidemic, mortality 60-90%
- During 16th century, started in Central Mexico
- Many indigenous sources depicting the epidemic
- No certainty on what the cause was
  - Typhoid fever
  - Epidemic typhus
  - Plague
  - Influenza
  - Hemorrhagic fever

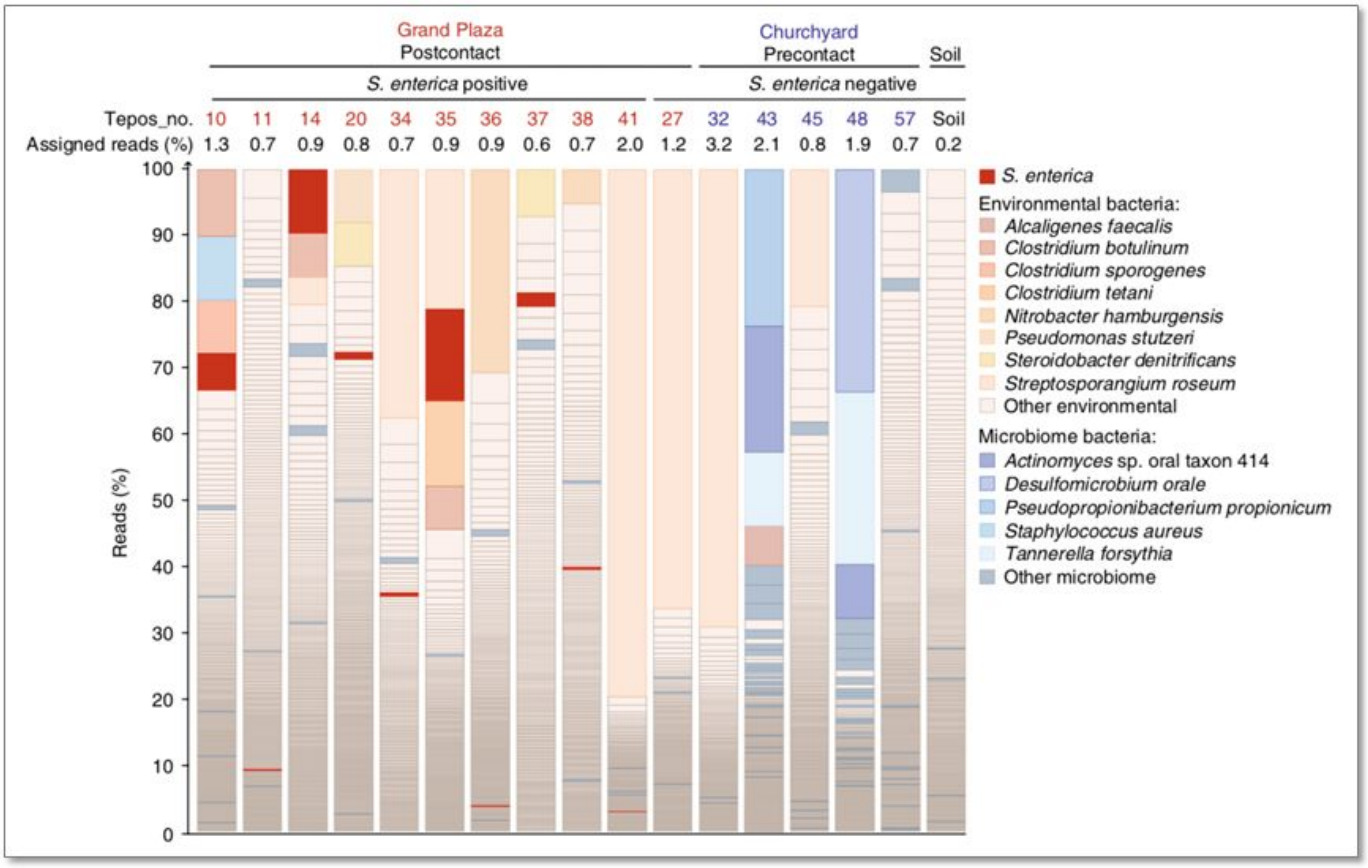


Cocolitzli-associated burial



Vågene *et al*. 2018 Nat Ecol Evol.

Metagenomic screening



Vågene *et al*. 2018 Nat Ecol Evol.

Cocolitzli-causing bacterium?

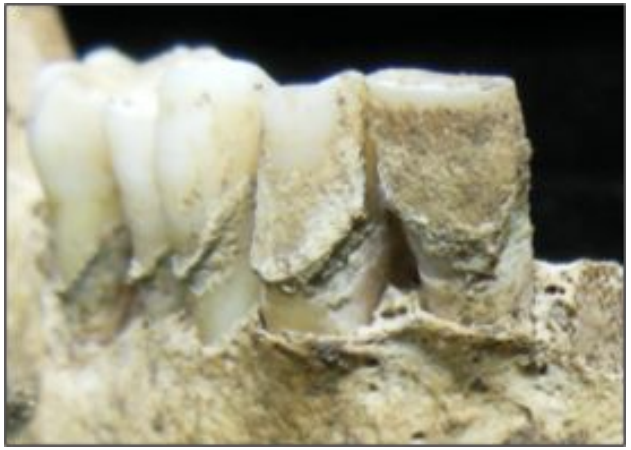
- *Salmonella enterica* subsp. *enterica* serovar Paratyphi C
  - Enteric fever
  - Infected individuals shed bacteria after symptoms have ended
  - Asymptomatic carriers
- Little is known about the past of this disease

Cocolitzli summary

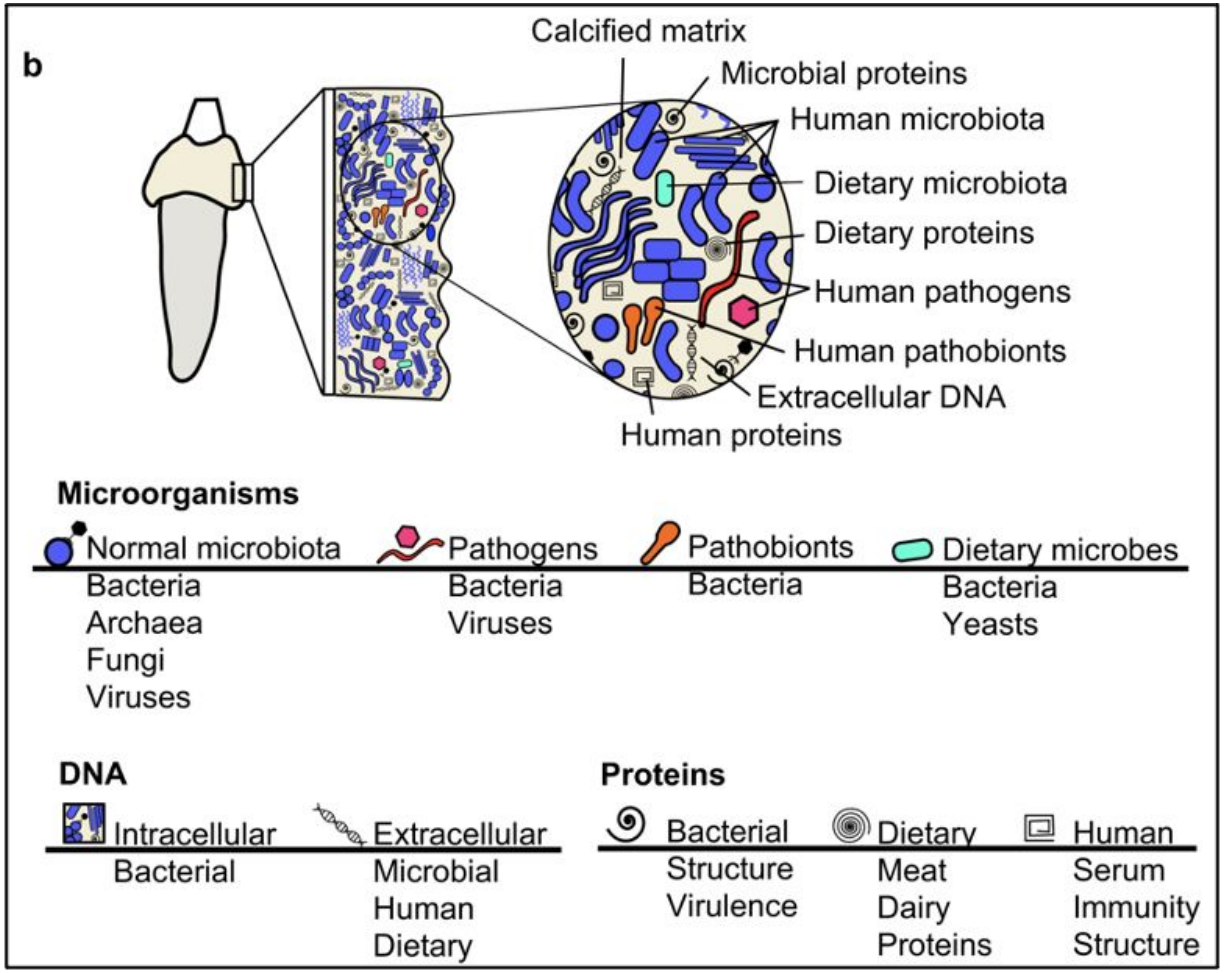
- First molecular evidence of what may have caused the epidemic
- More research needed
- We can start to solve ancient mysteries using aDNA
  - Connected with other lines of evidence

Microbiome

What is a microbiome?



Historic dental calculus
(~200 years old)



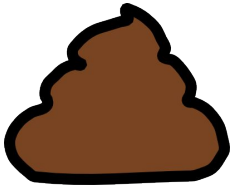
Modified from Velsko and Warinner 2017 *Bioarchaeology International*

Where are archaeological microbiomes?

Latrines

↓

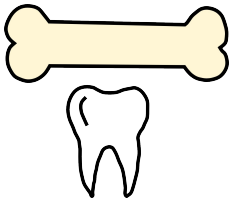
Parasites



Paleofeces

↓

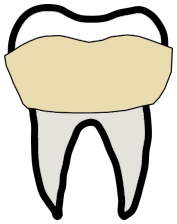
Gut



Bones & teeth

↓

Decomposition



Dental calculus

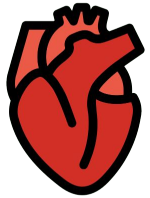
↓

Oral

Pottery

↓

Food/Drink



Preserved specimens

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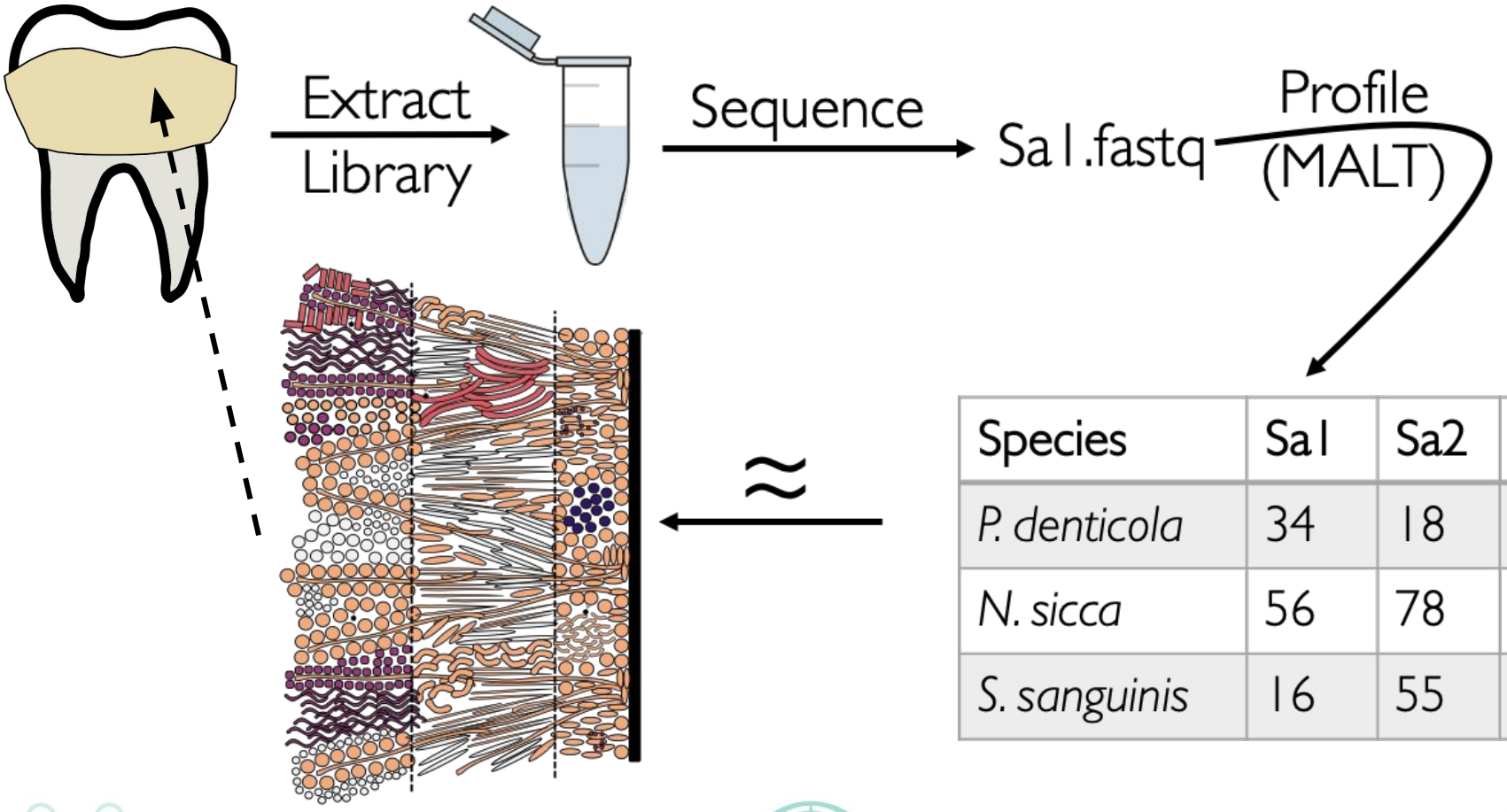
Other body sites

emojis from openmoji.org

How do we study microbiomes?



Species	Sa1	Sa2	Sa3
P. denticola	34	18	27
N. sicca	56	78	15
S. sanguinis	16	55	63

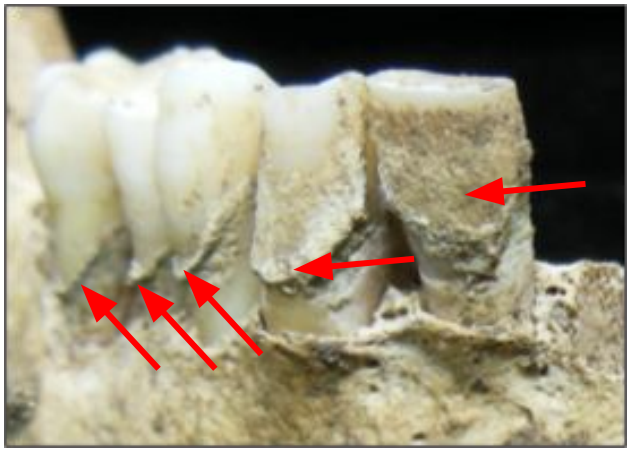
Dental calculus and paleofeces are common ancient human microbiomes

Hübner et al. in prep

Warinner et al. 2014

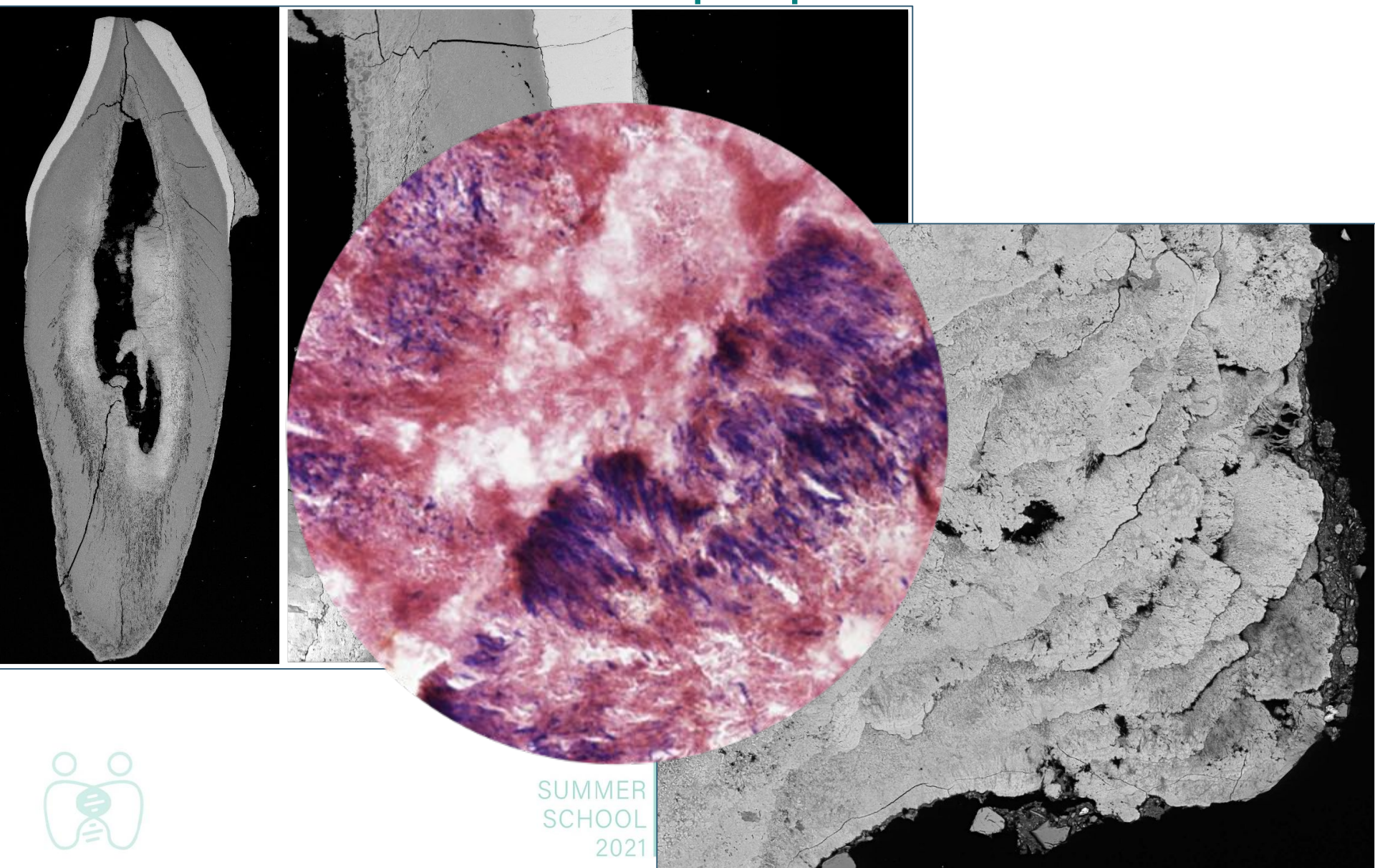
Ancient dental calculus...

- Mineralized dental plaque
  - associated diseases: dental cavities, periodontitis
- Ubiquitous in archaeological record
- Highly stable human microbiome
  - across time
  - across geography

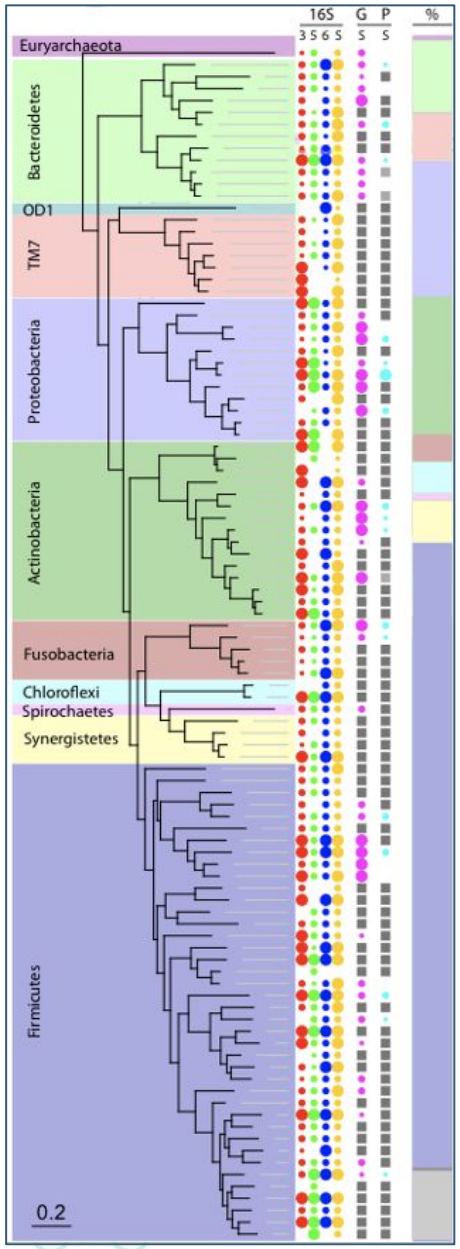


Historic dental calculus
(~200 years old)

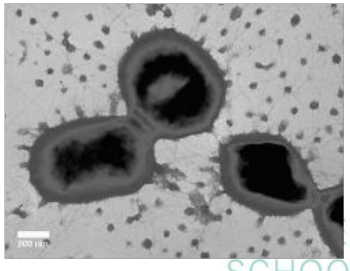
Ancient dental calculus looks like modern dental plaque

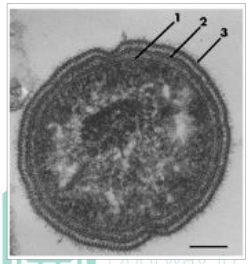


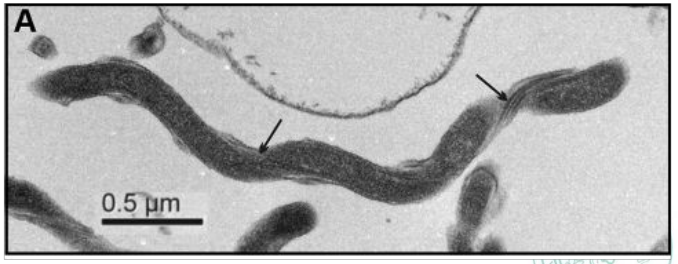
Ancient dental calculus resembles modern dental plaque



Pathogen[a,b]	Genes (contigs)	Proteins (peptides)	Virulence	Resistance[c]	Plasmid	CTn/Phage
Porphyromonas gingivalis	802 (2588)	7 (72)	++	+		+
Tannerella forsythia	1099 (11279)	10 (137)	++	+		+
Treponema denticola	917 (6106)	3 (15)	++	+	+	+

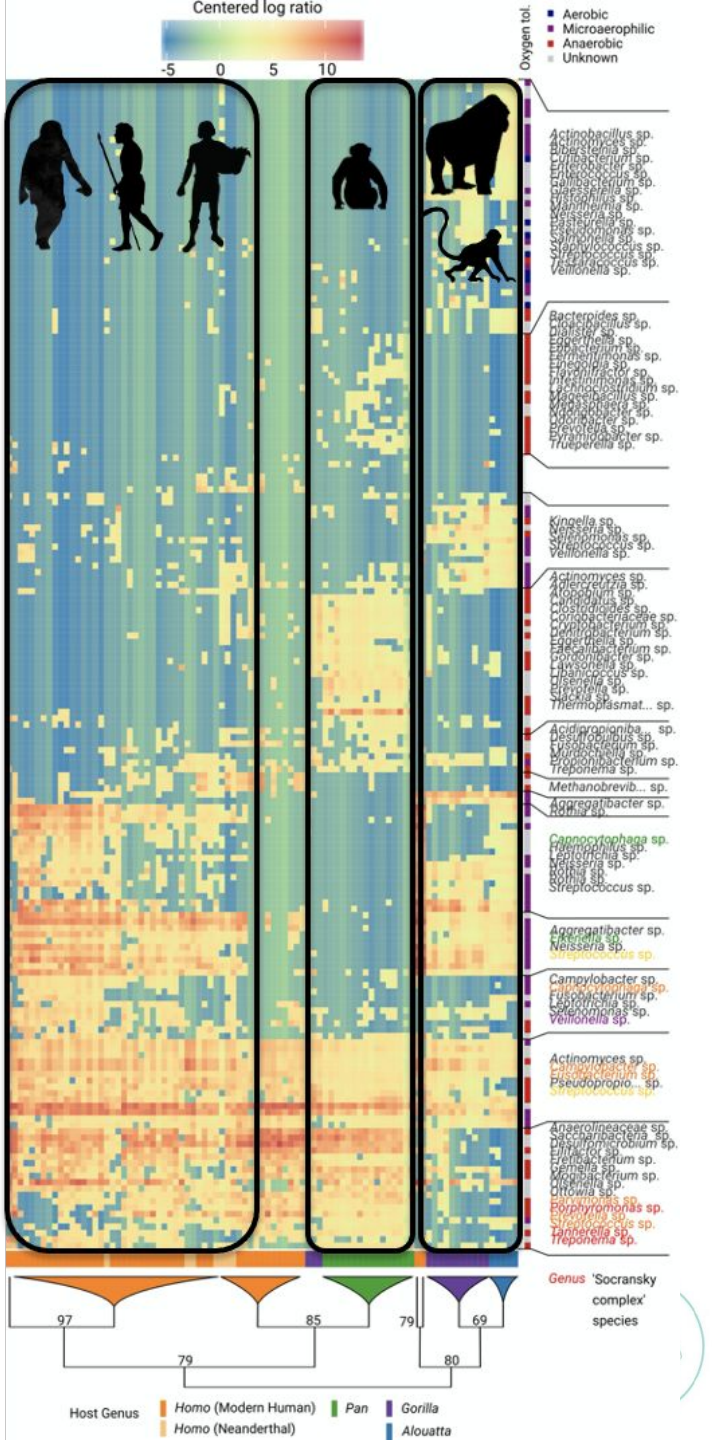






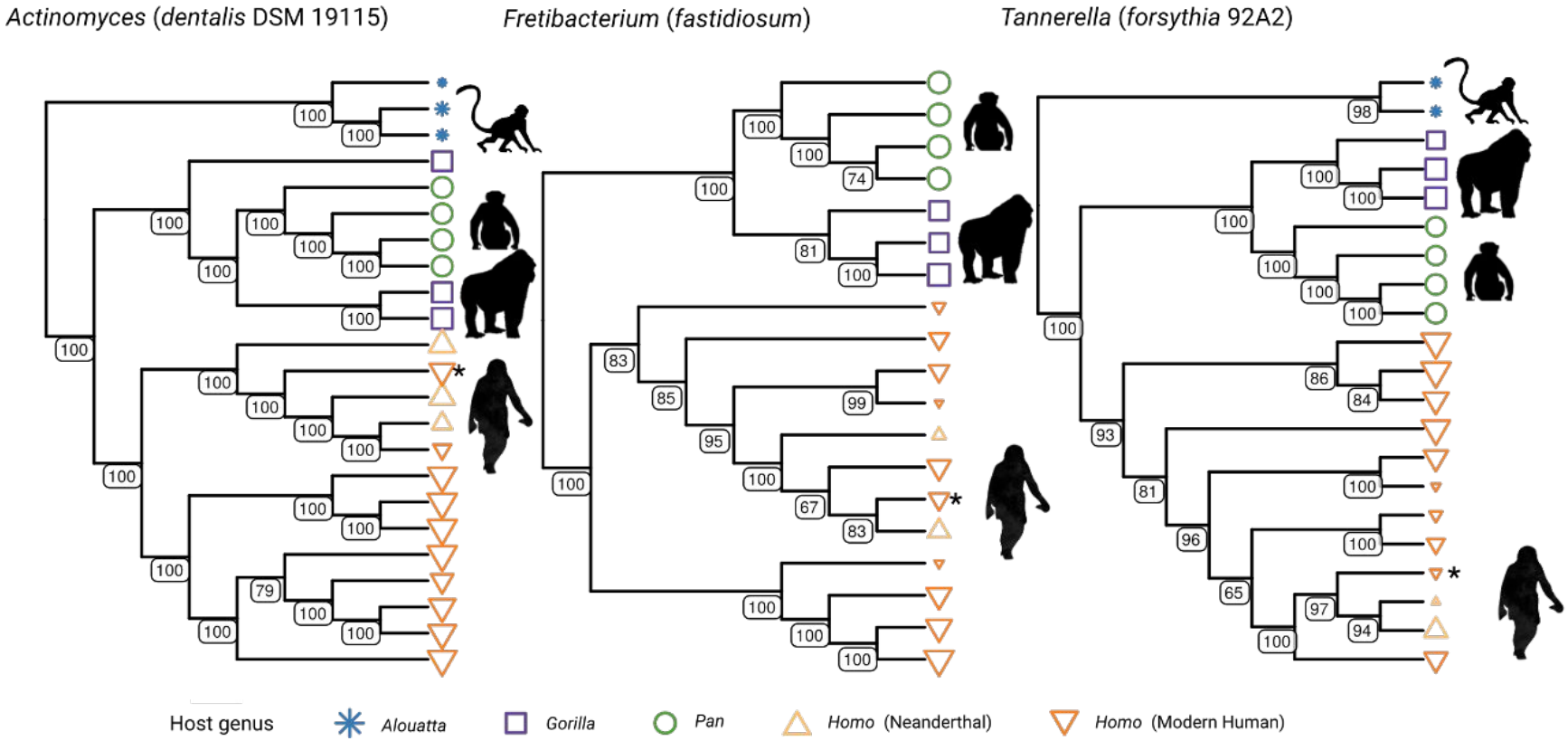
Calculus microbiomes are conserved through time

Modified from Fellows Yates, *et al.* 2021 *PNAS*



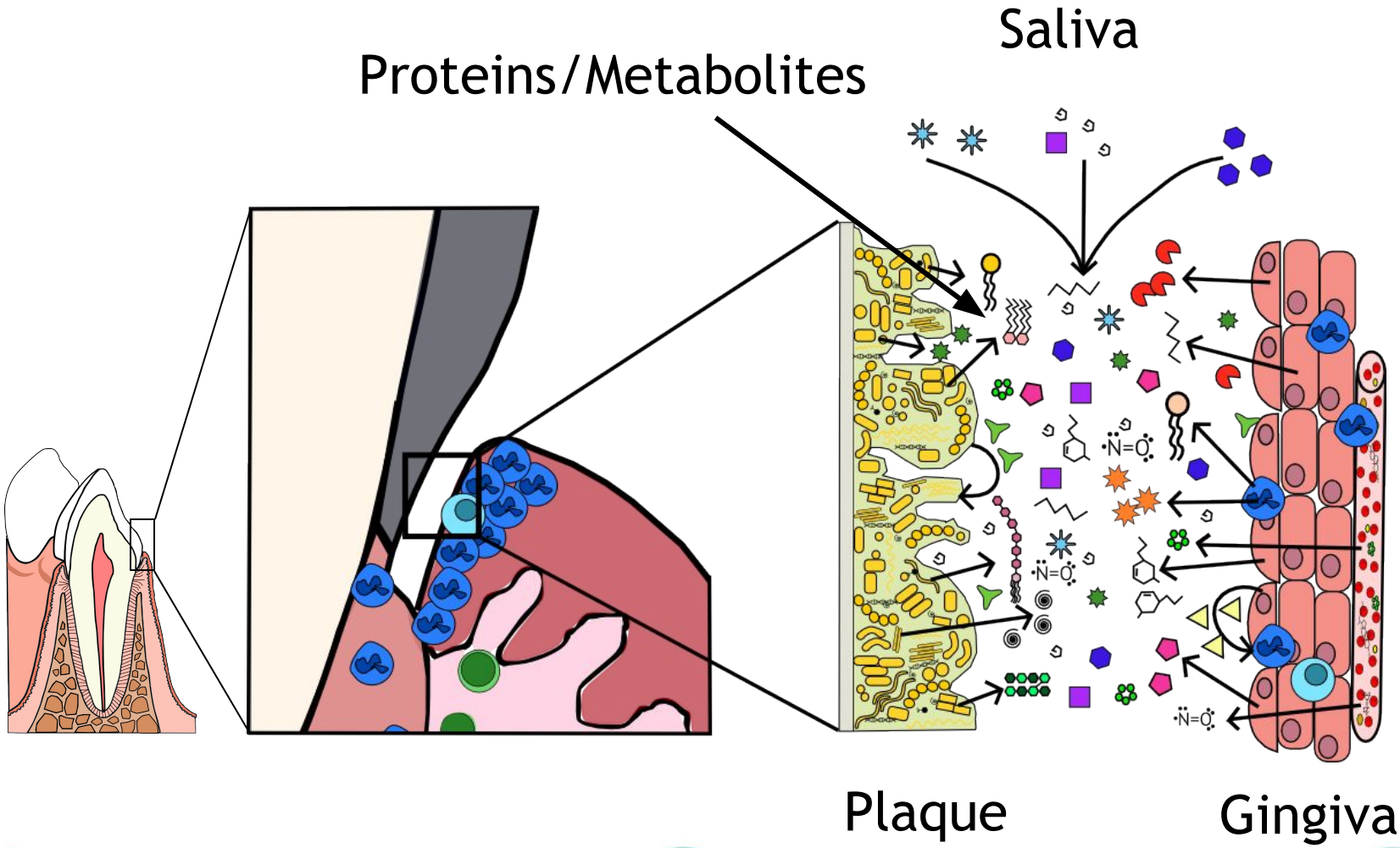
SUMMER SCHOOL 2021 Doorway to Human History

Microbial species evolution traces host evolution

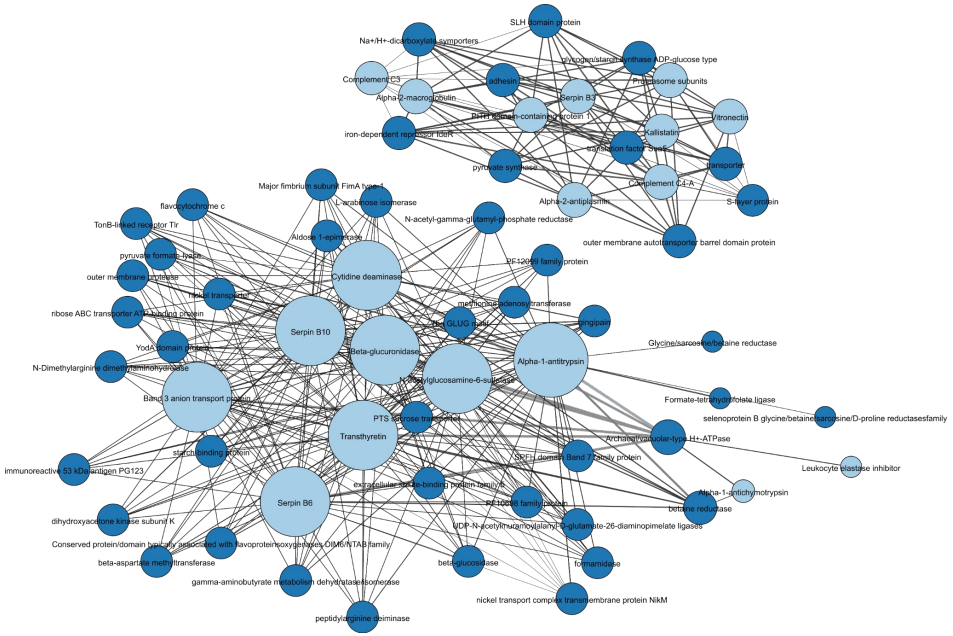


Modified from Fellows Yates, *et al*. 2021 *PNAS*

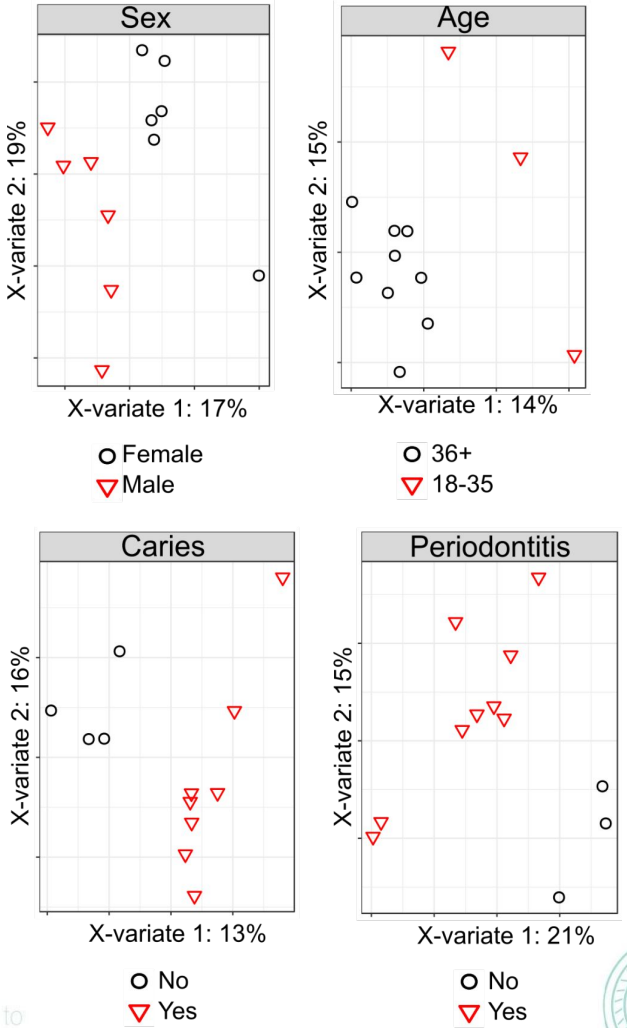
Biofilms are metabolically active



Proteins and metabolites preserve in ancient dental calculus



Modified from Velsko, *et al.* 2019 *Microbiome*

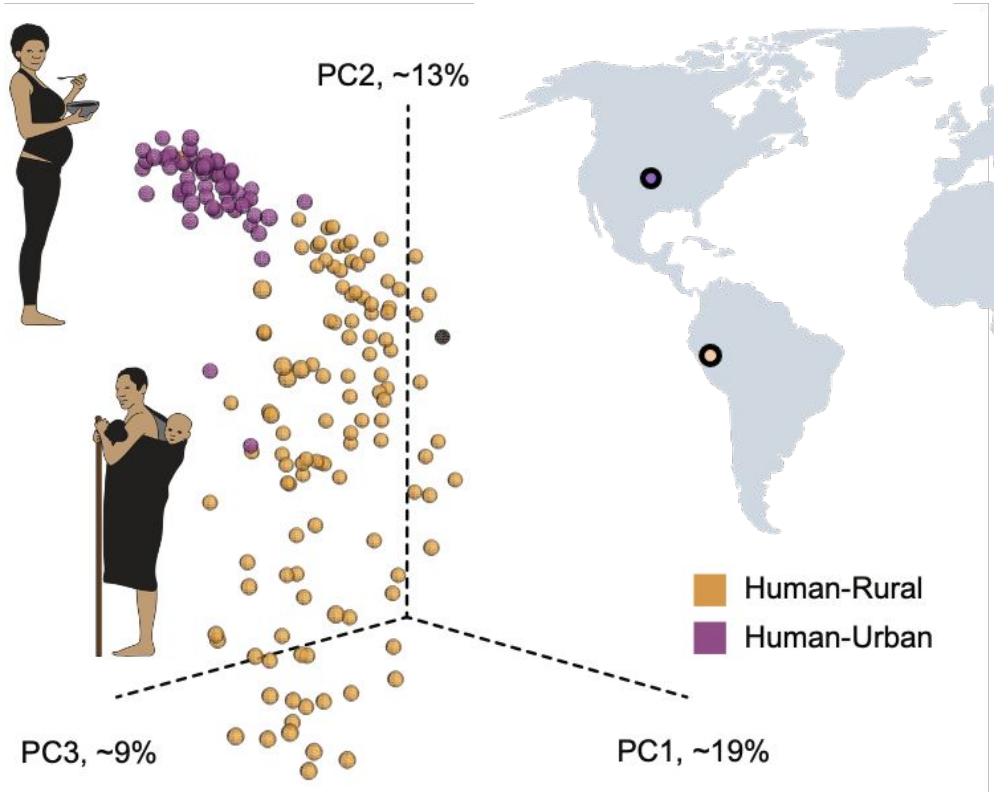


Modified from Velsko, *et al.* 2017 *Metabolomics*

Paleofeces

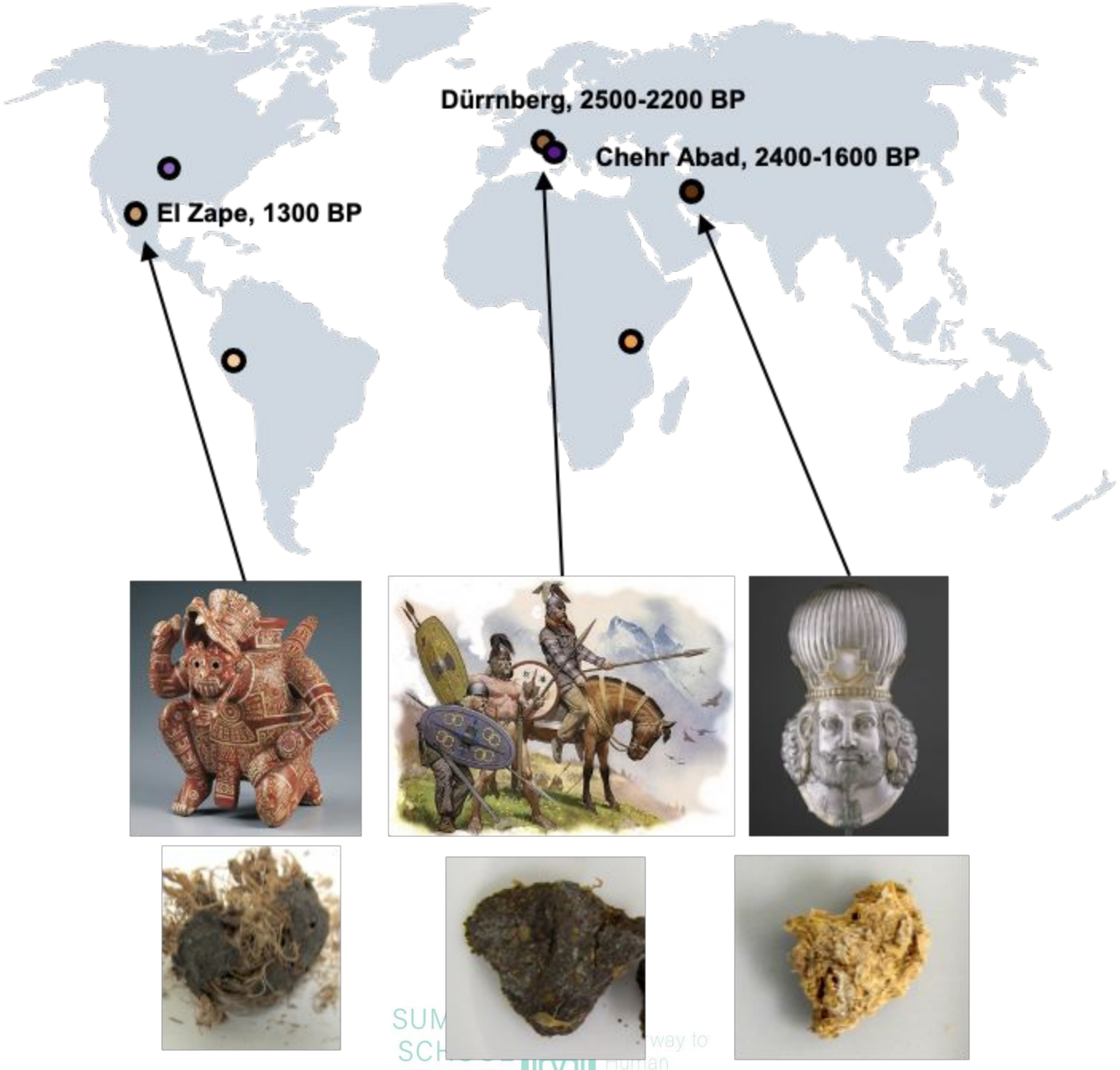
- Feces preserved under exceptional conditions
  - very dry (arid, salty)
- Associated with many inflammatory disorders
  - diseases of civilization/industrialization
- Uncommon in archaeological record
- Highly variable human microbiome

Rural and urban gut microbiomes differ

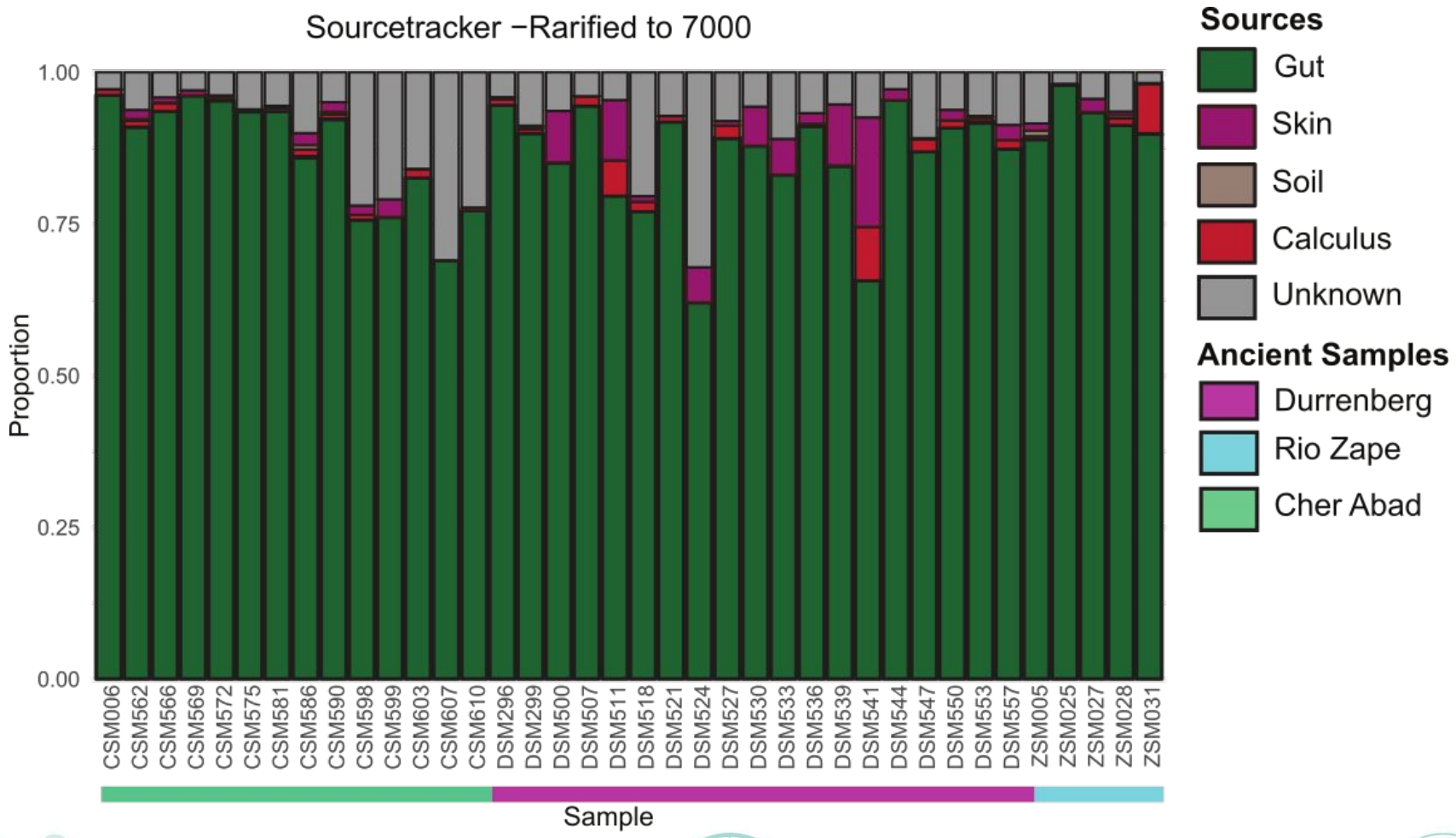


Paleofeces species composition

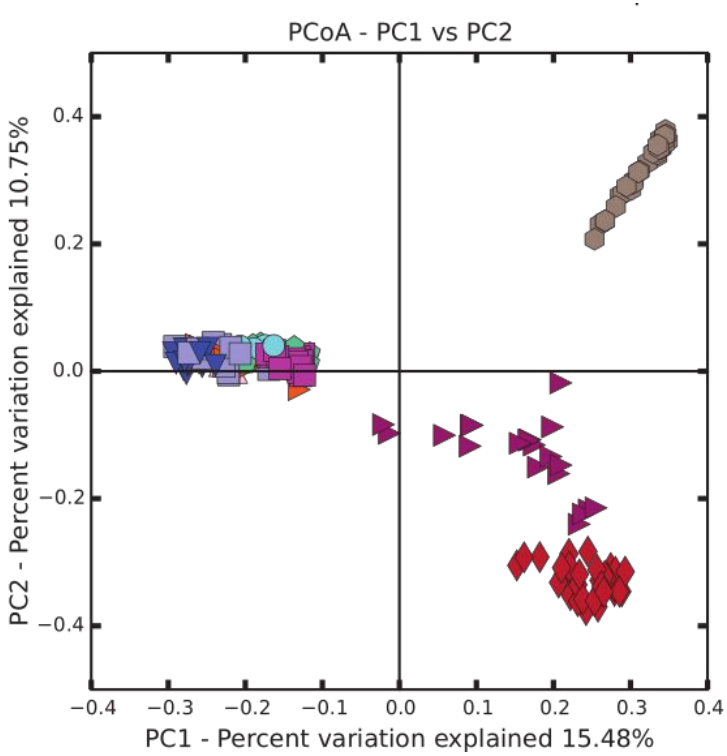
- When do changes happen?
- What drives changes?

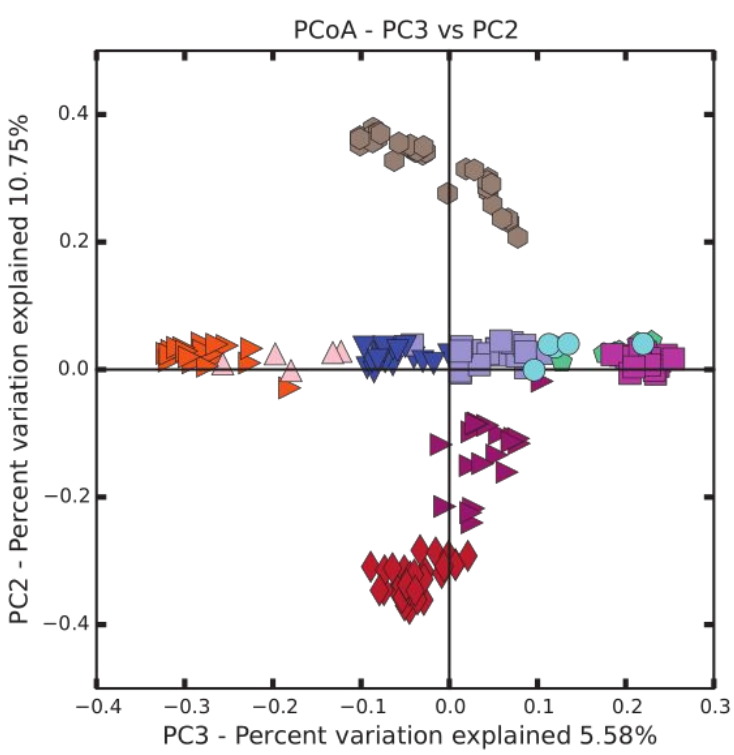


Well-preserved paleofeces resemble modern gut microbiomes



Well-preserved paleofeces resemble traditional food producer gut microbiomes





Sources

- Gut
- Skin
- Soil
- Calculus
- Unknown

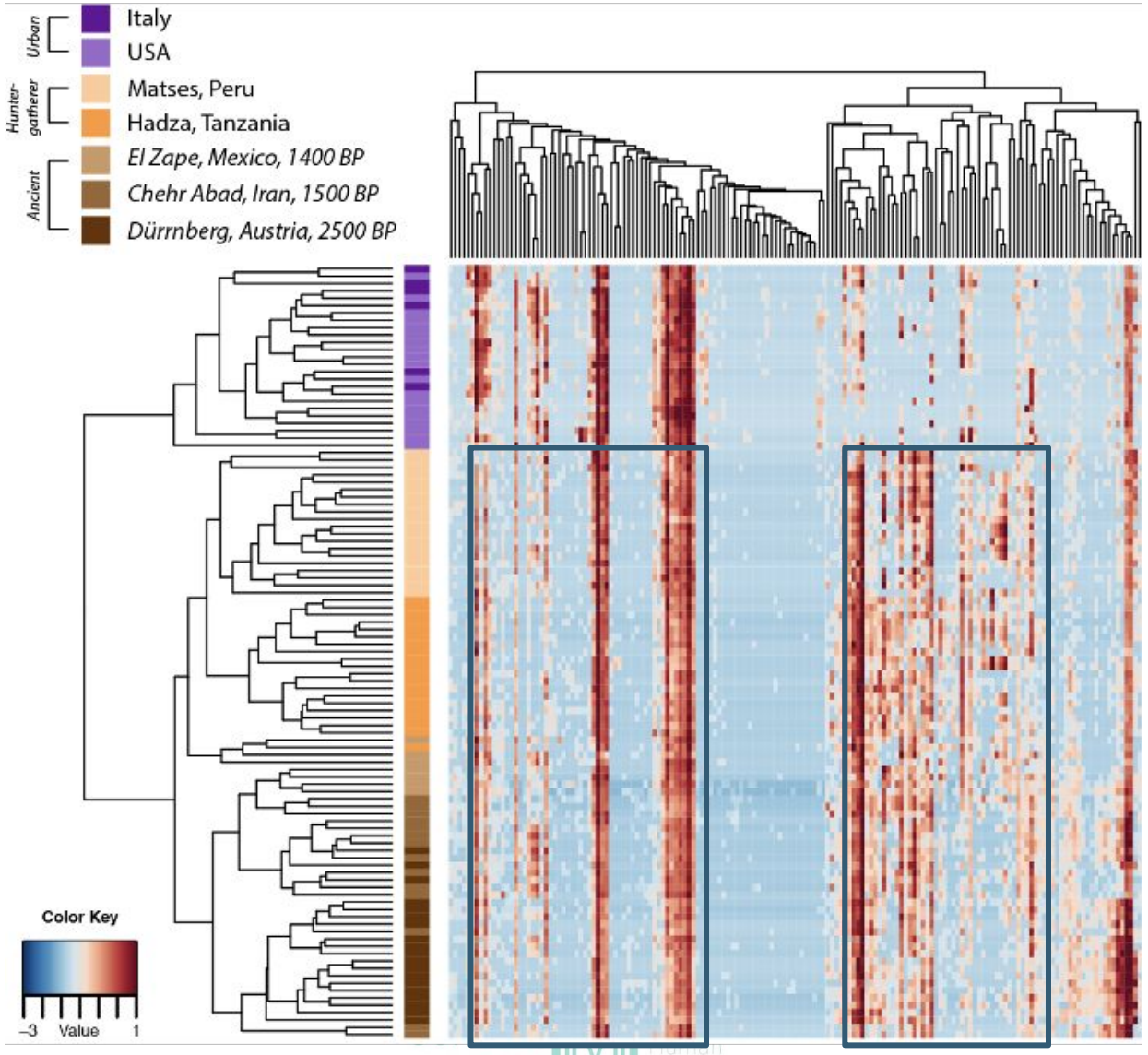
Ancient Samples

- Durrenberg
- Rio Zape
- Cher Abad

Modern Samples

- Matses
- Hadza
- USA
- Italy

Modern guts lack species found in rural gut and paleofeces microbiomes



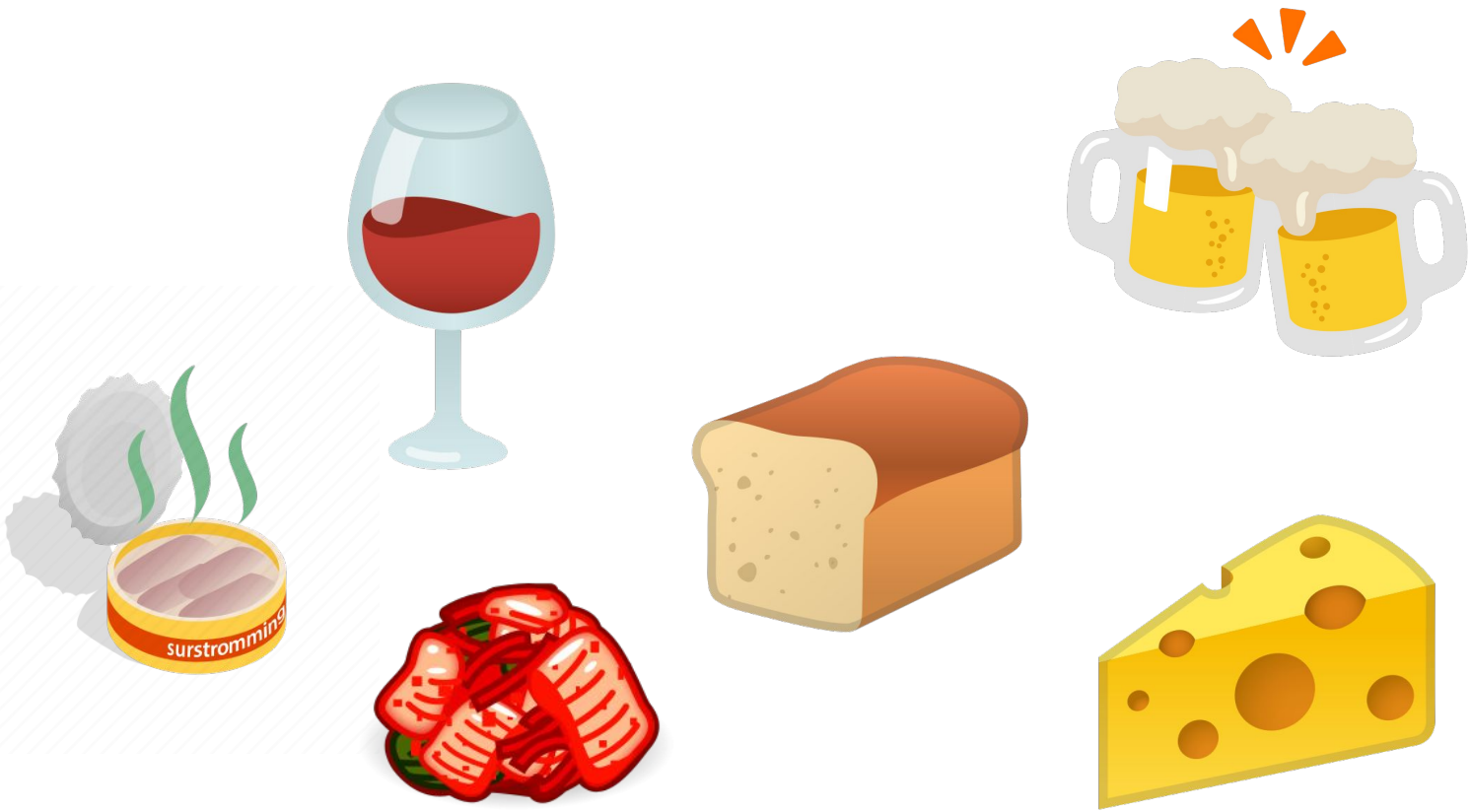
Microbiome take-home points

Microbiomes

- Preserve in the archaeological record
- Contain information about
  - Microbial taxonomy
  - Microbial activity (proteins/metabolites)
- Reflect host changes
- Help us understand historic health and disease
  - Long-term changes to human condition

Cuisine

Dietary microbes



The dairy mystery

The dairy mystery

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Proteomics evidence for kefir dairy in Early Bronze Age China

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ABSTRACT

Cheese making has been inferred at several sites in northern Europe as early as the 6th millenniu
and was common in Egypt and Mesopotamia in 3rd millennium BC. However, the remains of ar
cheeses have never been found and recipes of ancient dairy, its production scale, social and econ
impact remain poorly understood. Here we present direct proteomics evidence for the production
earliest known cheese that was found as an organic mass associated with the mummies of Early Br
Age cemetery of Xiaohe (1980–1450 BC) in Xinjiang, China. Kefir fermentation of ruminant milk
symbiotic culture of *Lactobacillus kefiranofaciens* and other lactic acid bacteria and yeasts was the ba
robust, scalable, probiotic, lactose-free dairy and a key technological advance that introduced econ
benefits of extensive herding into a semi-pastoral household of the Eastern Eurasia population alrea
the Early Bronze Age.

The dairy mystery

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Direct evidence of milk consumption from ancient human dental calculus

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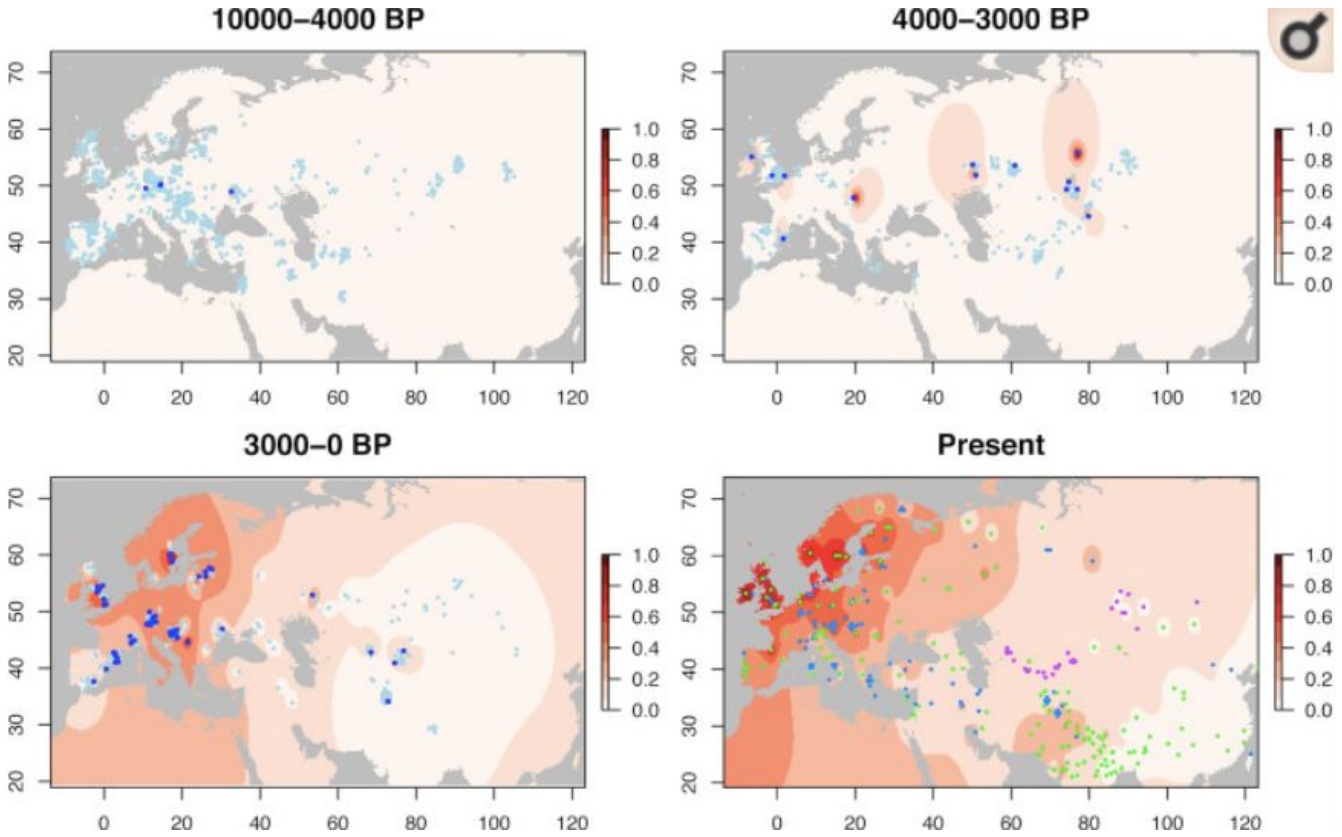
Lactase persistence



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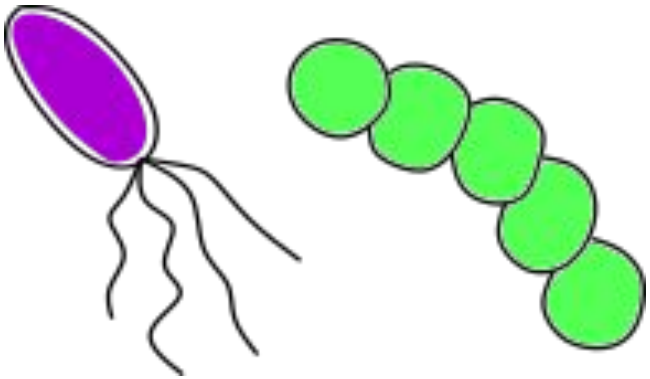
Microbes? Cooking?

The gut microbiome?



The dairy mystery

Archaeogenetics
Palaeoproteomics
Ethnography
Modern microbiology
Archaeology...

Stay tuned!

Overall conclusions slide

- Microbes are awesome.
- Microbes are abundant in the archaeological record
- They can be identified with various biomolecular techniques
- They can tell us about multiple aspects of the human past

Discussion questions

- Are there historically recorded diseases that you might want to investigate with aDNA? What are they?
- Can you think of places to find ancient microbes other than the ones we presented?
- How can studying ancient microbes help us with modern medicine and microbiology?
- How can ancient animal samples be used to study the origin of zoonotic (animal-derived) diseases?
- How can studying microbiomes help us understand human lifestyle changes?
- How have the words used to explain/describe infectious disease evolved over time?