PROTEINS, PROTEOMS, METAPROTEOMS

EVA CHOCHOLOVÁ
LABORATORY OF BIOLOGICAL AND MOLECULAR ANTHROPOLOGY
DEPARTMENT OF EXPERIMENTAL BIOLOGY

PROTEINS

- Collagens
- Keratins and corneous β-proteins
- Fibroin
- Amelogenin

} Previous presentation

- Mostly taxonomy, identification

Warinner et al., 2022 DOI: 10.1021/acs.chemrev.1c00703

SEX DETERMINATION

- Amelogenin cleaved and incorporated during enamel maturation
- Gene *AMEL*
- Non-mammals – autosomal (no difference)
- Mammals – *AMELX, AMELY*
  - Most species only *AMELX* funtional or even complete loss of *AMELY*
  - Some species *AMELX = AMELY* in sequence
  - Sequence difference enables sex determination in e.g., *Homo, Bovis, Capra, Ovis, Equus*…

- Ideal in cases of lower preservation (subadults, incomplete skeletal material, low DNA preservation, very old samples)
- Cost-effective

SEX DETERMINATION

RESEARCH ARTICLE | ANTHROPOLOGY |

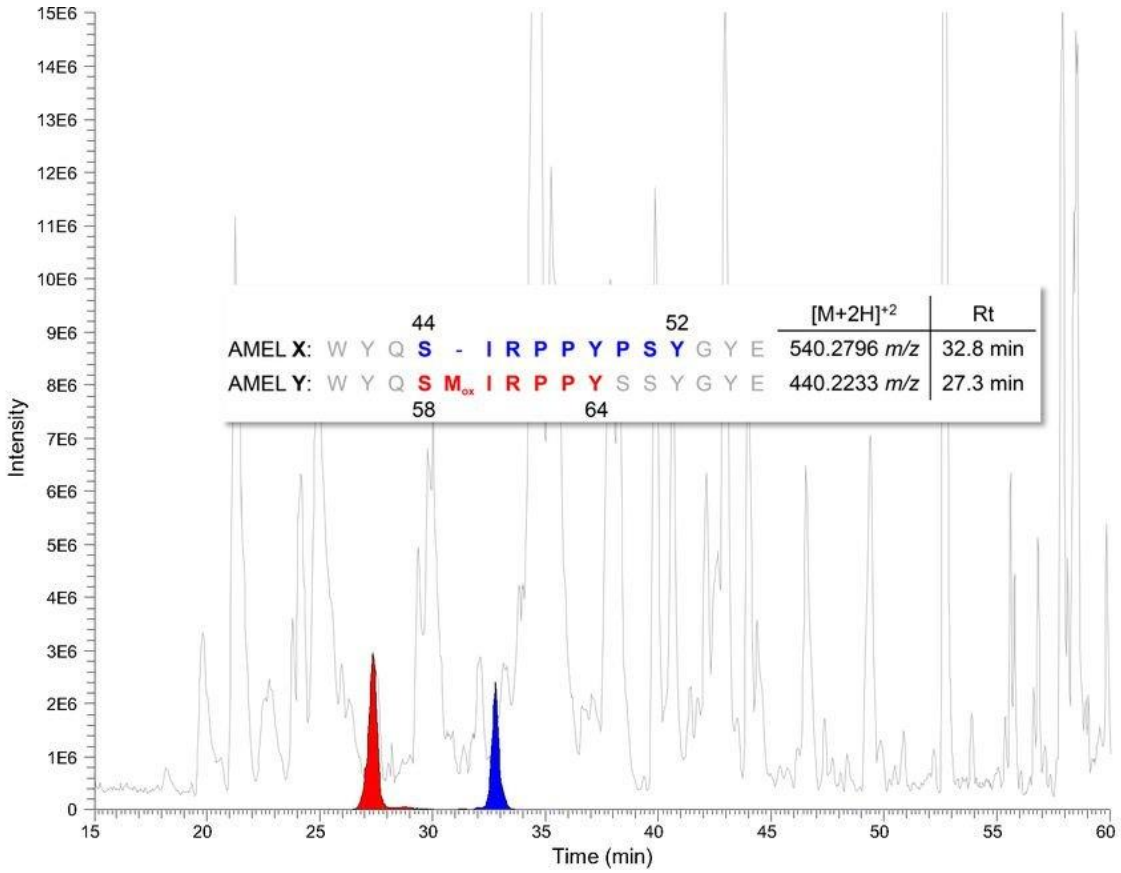
Sex determination of human remains from peptides in tooth enamel

Nicolas Andre Stewart, Raquel Fernanda Gerlach, Rebecca L. Gowland, Kurt J. Gron, and Janet Montgomery
Authors Info & Affiliations

Edited by Christopher Kuzawa, Northwestern University, Evanston, IL, and accepted by Editorial Board Member C. O. Lovejoy November 13, 2017 (received for review August 23, 2017)

December 11, 2017 | 114 (52) 13649-13654 | https://doi.org/10.1073/pnas.1714926115

- acid etching of tooth enamel – minimally destructive
- nanoflow liquid chromatography mass spectrometry (nanoLC-MS)



SEX DETERMINATION

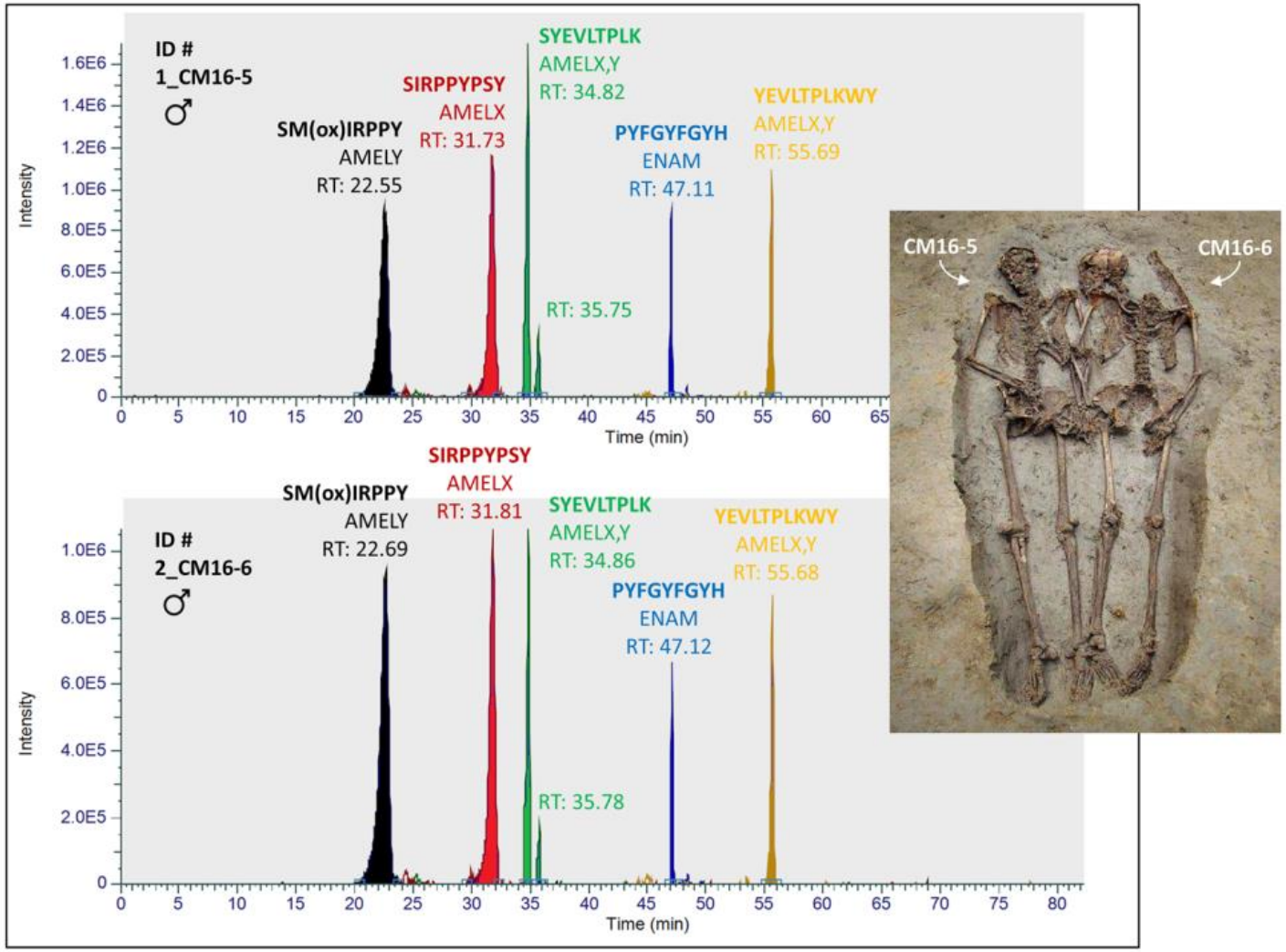
OPEN

Enamel peptides reveal the sex of the Late Antique 'Lovers of Modena'

Received: 13 February 2019
Accepted: 7 August 2019
Published online: 11 September 2019

Federico Lugli[1,2], Giulia Di Rocco[3], Antonino Vazzana[1], Filippo Genovese[4], Diego Pinetti[4], Elisabetta Cilli[1], Maria Cristina Carile[1], Sara Silvestrini[1], Gaia Gabanini[1], Simona Arrighi[1], Laura Buti[1], Eugenio Bortolini[1], Anna Cipriani[2,5], Carla Figus[1], Giulia Marciani[1], Gregorio Oxilia[1], Matteo Romandini[1], Rita Sorrentino[1,6], Marco Sola[3] & Stefano Benazzi[1,7]

- Changing preconceptions
- Possible even in poor DNA preservation
- Suggest additional unique peptides beside 440.2233 (SM(ox)IRPPY), e.g. 432.2258 (SMIRPPY) and 396.7073 (M(ox)IRPPY)
- Most probably war comrades or relatives - unknown



Ion chromatograms representing selected peptides of the 'Lovers' enamel proteome[20]. Chromatograms search was performed using Xcalibur software (Thermo Scientific) with a mass tolerance of 5 ppm. Peptide sequences, protein names and retention times are reported in the graphs. The presence of peptide SM$_{(ox)}$IRPPY (AMELY; [M+ 2 H]$^{+2}$ 440.2233 *m/z*) in both the specimens suggests that the two individuals were males.

SEX DETERMINATION

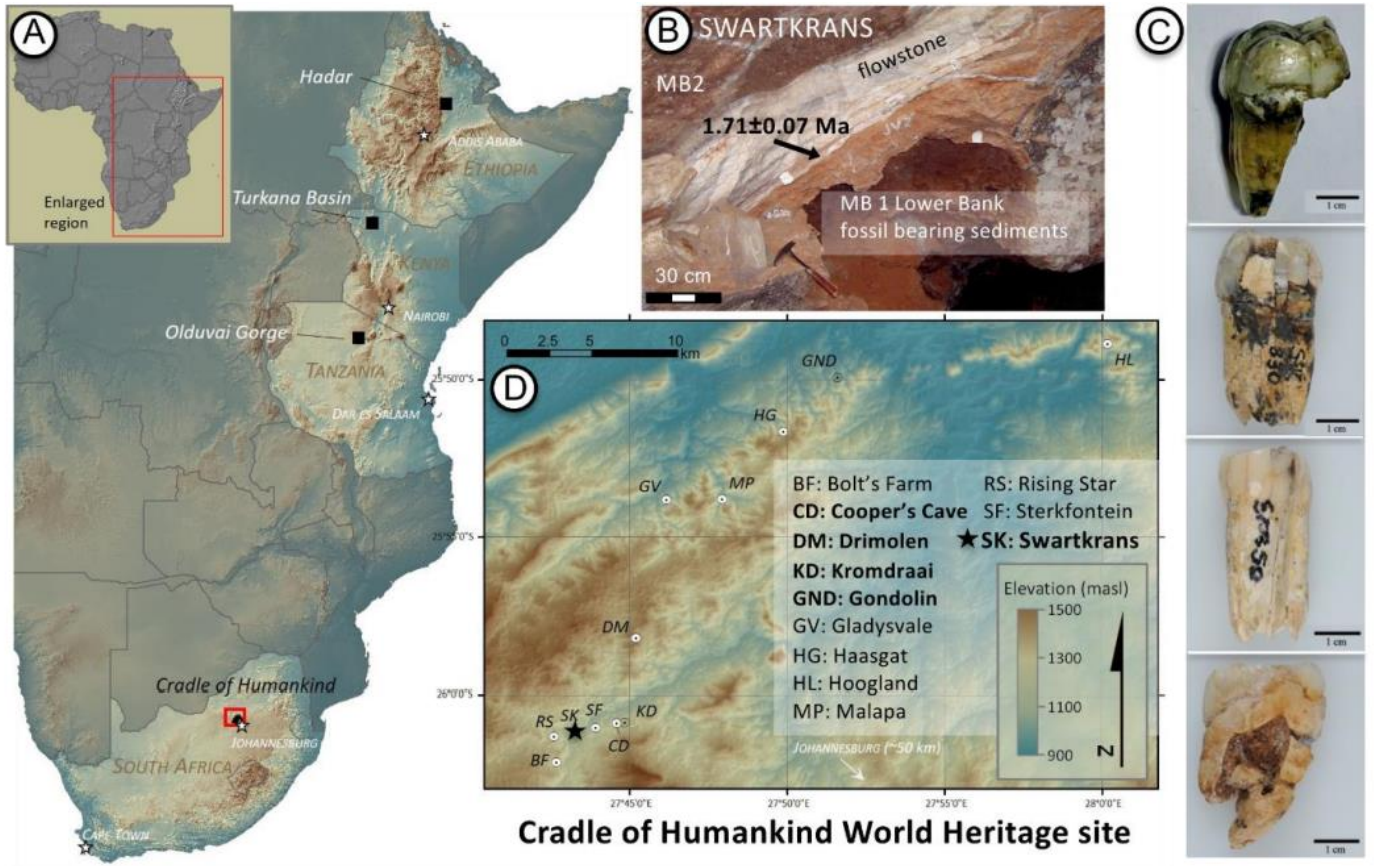
Enamel proteins reveal biological sex and genetic variability within southern African *Paranthropus*

Palesa P. Madupe, Claire Koenig, Ioannis Patramanis, Patrick L. Rüther, Nomawethu Hlazo, Meaghan Mackie, Mirriam Tawane, Johanna Krueger, Alberto J. Taurozzi, Gaudry Troché, Job Kibii, Robyn Pickering, Marc Dickinson, Yonatan Sahle, Dipuo Kgotleng, Charles Musiba, Fredrick Manthi, Liam Bell, Michelle DuPlessis, Catherine Gilbert, Bernhard Zipfel, Lukas F. K. Kuderna, Esther Lizano, Frido Welker, Pelagia Kyriakidou, Jürgen Cox, Catherine Mollereau, Caroline Tokarski, Jonathan Blackburn, Jazmín Ramos-Madrigal, Tomas Marques-Bonet, Kirsty Penkman, Clément Zanolli, Lauren Schroeder, Fernando Racimo, Jesper V. Olsen, Rebecca R. Ackermann, Enrico Cappellini

doi: https://doi.org/10.1101/2023.07.03.547326

preprint, 2023

- 2 million years old, *Paranthropus robustus*
- Oldest African hominin DNA about 0.018 Ma
- May help elucidate taxonomic placement



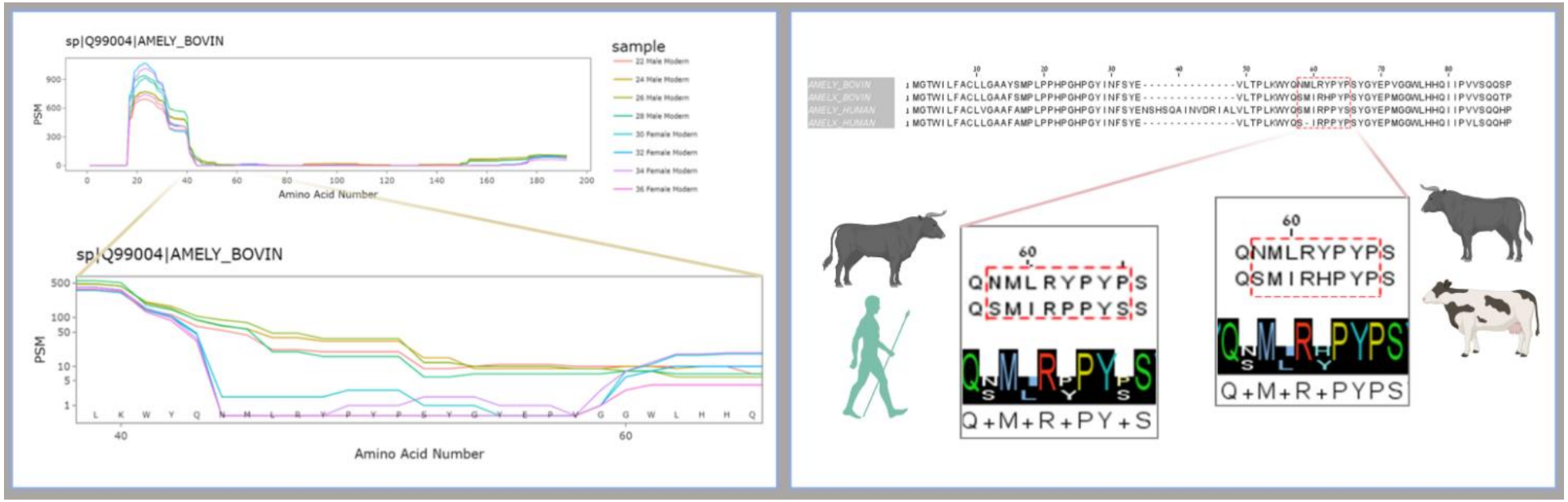
SEX DETERMINATION

A Peptidomics Method for Assessing Sex from Modern and Ancient Bovine Tooth Enamel.

Paula Kotli[1*], David Morgenstern[2], Liora Kolska Horwitz[3], Hamoudi Khalaily[4], Fanny Bocquentin[5], and Elisabetta Boaretto[1*]

preprint, 2024

- Sex determination in cattle



Left: Spectral counting-based coverage of AmelY protein in peptidomics data from eight modern samples - fourmales and four females, y-axis presents the number of ID spectra per amino acid position in the protein in log base 10 scale.Note, the difference between male and female samples in this region. Right: Human and cattle Amelogenin sequencealignments using Jalview (version 2.11.2.6). The close up are alignment and consensus sequences: Left, shows the uniqueAmelY peptide identified in human and its alignment with our unique cattle AmelY sequence. Right, cattle AmelY and AmelXalignments showing SAAVs obtained in this research that successfully determine sex in cattle.

PROTEOMS

- Bone and dentine
- Enamel
- Eggshells
- Mollusc shells
- Mummified remains
- Plants

Warinner et al., 2022 DOI: 10.1021/acs.chemrev.1c00703

SKIN

- Improved taxonomy, beyond taxonomic assignment
- Identification of proteins with expression varying in age groups – estimation of age of death from calf skin

Species Identification of Archaeological Skin Objects from Danish Bogs: Comparison between Mass Spectrometry-Based Peptide Sequencing and Microscopy-Based Methods

Luise Ørsted Brandt, Anne Lisbeth Schmidt, Ulla Mannering, Mathilde Sarret, Christian D. Kelstrup, Jesper V. Olsen, Enrico Cappellini

Published: September 26, 2014 • https://doi.org/10.1371/journal.pone.0106875

Sample no.	Find	MO+LM	LM+SEM	MS	MO+LM vs LM+SEM	MO+LM vs MS	LM+SEM vs MS
1	Baunsø, NM D11103a	Cattle	Horse	Goat	≠	≠	≠
2	Baunsø, NM D11103b	Goat	Cattle	Cattle	≠	≠	=
3	Baunsø, NM D11103c	Sheep	Sheep	Sheep	=	=	=
4	Borremose I, NM C26450	Sheep	Sheep	Sheep	=	=	=
5	Huldremose I dark, NM C3471	Sheep	Sheep	Sheep	=	=	=
6	Huldremose I light, NM C3471	Sheep	Sheep	Sheep	=	=	=
7	Karlby, NM D4854b	Sheep	Sheep	Sheep	=	=	=
8	Karlby, NM D4854c	Goat	Horse	Goat	≠	=	≠
9	Karlby, NM D4854e	Sheep	Cattle	Sheep	≠	=	≠
10	Møgelmose, NM 16316	Goat	Cattle	Cattle	≠	≠	=
11	Roum, NM C37412	Sheep	Sheep	Sheep/goat	=	=	=
12	Haraldskær, NM 3705	Cattle	*	Goat		≠	

"MO+LM": macroscopical observation and light microscopy, "LM+SEM": light microscopy and scanning electron microscopy, "MS": Mass Spectrometry-based peptide sequencing. *This item is thought to be deliberately de-haired and only few hairs are preserved on the surface. Therefore there was only sufficient hair for one microscopic analysis. =/≠ indicate same/different species identification achieved by the methods compared.
doi:10.1371/journal.pone.0106875.t002

SKIN

- Improved taxonomy, beyond taxonomic assignment
- Identification of proteins with expression varying in age groups – estimation of age of death from calf skin

RESEARCH ARTICLE | ANTHROPOLOGY |

Animal origin of 13th-century uterine vellum revealed using noninvasive peptide fingerprinting

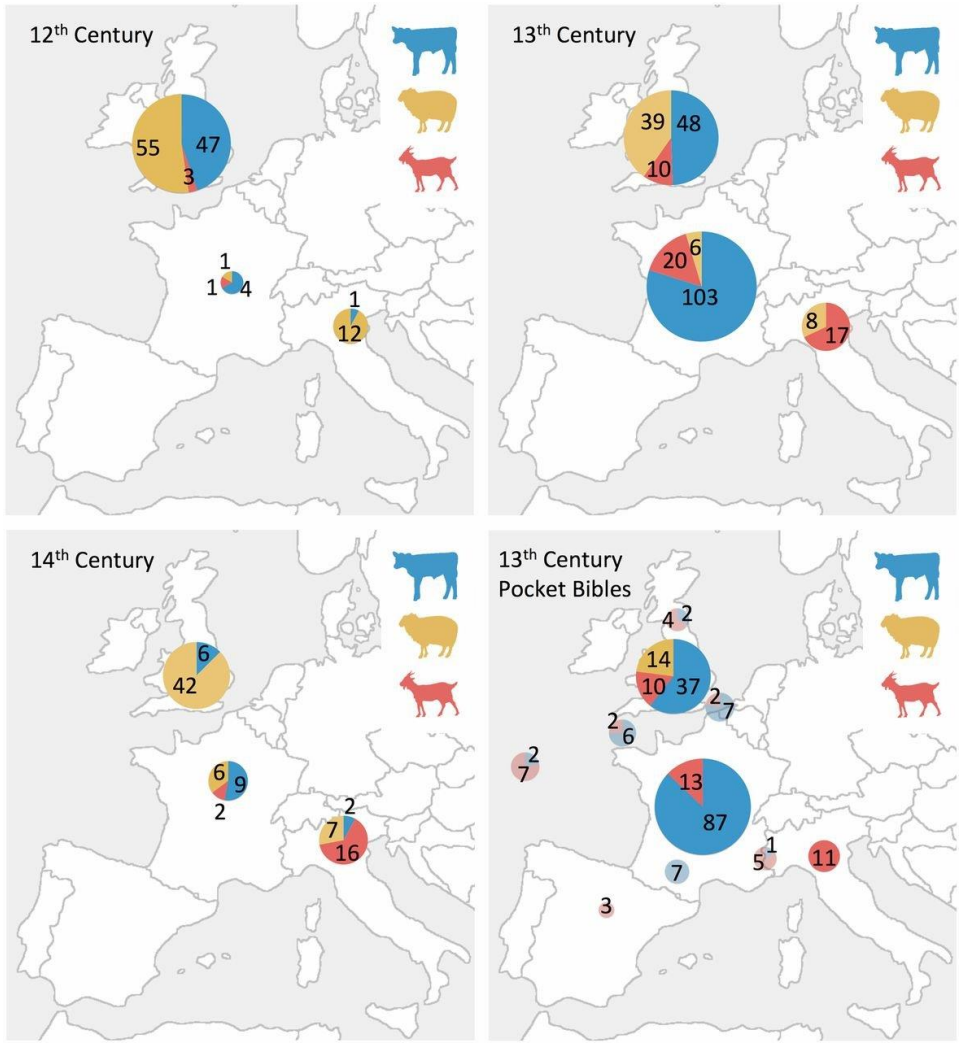
Sarah Fiddyment, Bruce Holsinger, Chiara Ruzzier, Alexander Devine, Annelise Binois, Umberto Albarella, Roman Fischer, Emma Nichols, Antoinette Curtis, Edward Cheese, Matthew D. Teasdale, Caroline Checkley-Scott, Stephen J. Milner, Kathryn M. Rudy, Eric J. Johnson, Jiří Vnouček, Mary Garrison, Simon McGrory, Daniel G. Bradley, and Matthew J. Collins

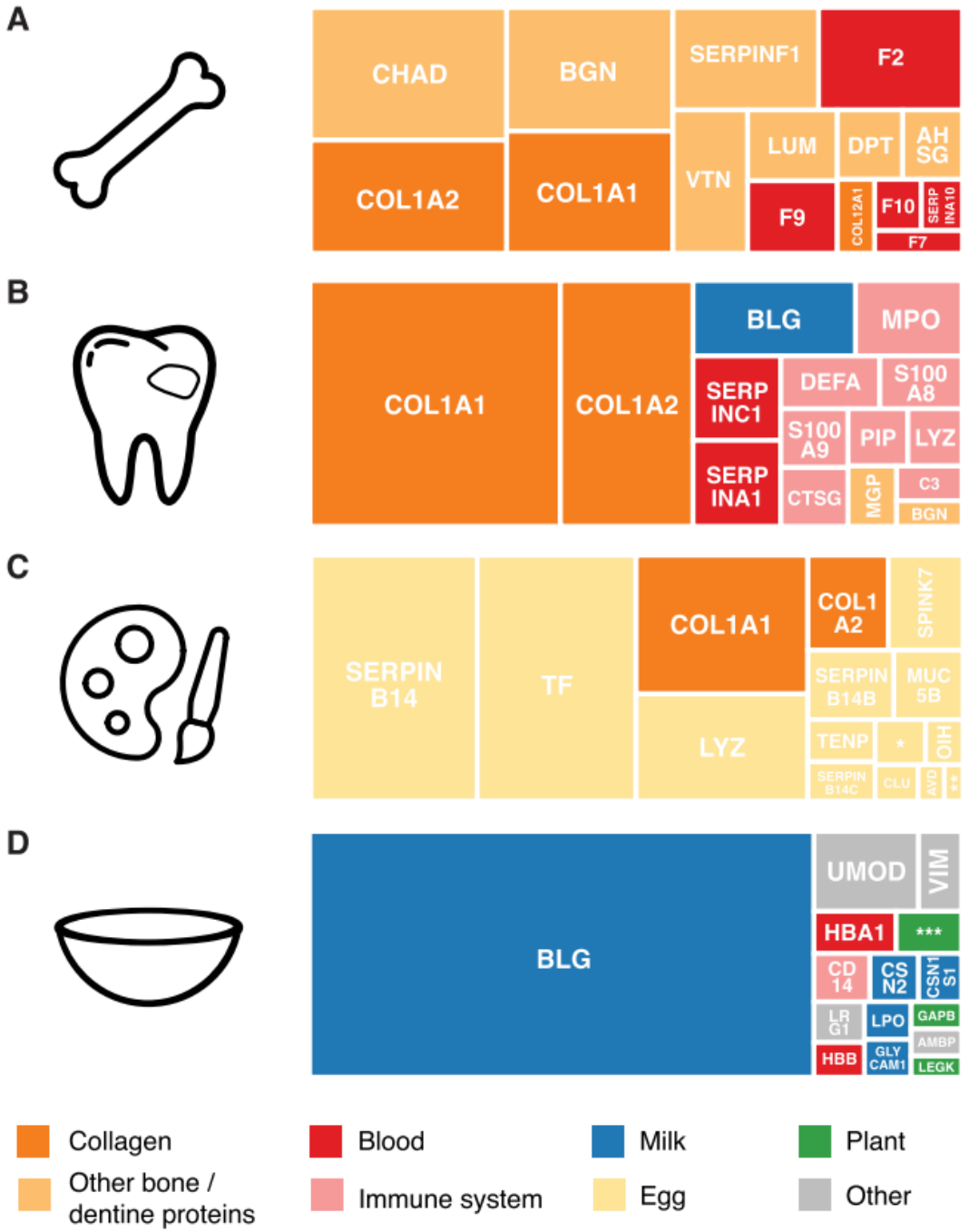
-16 Authors Info & Affiliations

Edited by Michael G. Campana, Smithsonian National Zoological Park, Washington, DC, and accepted by the Editorial Board October 22, 2015 (received for review June 23, 2015)

November 23, 2015 | 112 (49) 15066-15071 | https://doi.org/10.1073/pnas.1512264112

Tissue-thin parchment made it possible to produce the first pocket Bibles: Thousands were made in the 13th century. The source of this parchment, often called "uterine vellum," has been a long-standing controversy in codicology. Use of the Latin term *abortivum* in many sources has led some scholars to suggest that the skin of fetal calves or sheep was used. Others have argued that it would not be possible to sustain herds if so many pocket Bibles were produced from fetal skins, arguing instead for unexpected alternatives, such as rabbit. Here, we report a simple and objective technique using standard conservation treatments to identify the animal origin of parchment. The noninvasive method is a variant on zooarchaeology by mass spectrometry (ZooMS) peptide mass fingerprinting but extracts protein from the parchment surface by using an electrostatic charge generated by gentle rubbing of a PVC eraser on the membrane surface. Using this method, we analyzed 72 pocket Bibles originating in France, England, and Italy and 293 additional parchment samples that bracket this period. We found no evidence for the use of unexpected animals; however, we did identify the use of more than one mammal species in a single manuscript, consistent with the local availability of hides. These results suggest that ultrafine vellum does not necessarily derive from the use of abortive or newborn animals with ultrathin hides, but could equally well reflect a production process that allowed the skins of maturing animals of several species to be rendered into vellum of equal quality and fineness.





Representative examples of ancient proteomes. Well-preserved ancient proteomes contain distinctive groups of proteins that reflect the protein composition of the original tissue or material, such as human bone[364] (A), human dental calculus[445] (B), artist materials[137] (C), and pottery crusts[99] (D). As such, the composition of an ancient proteome can aid in its authentication. Data were searched against the SwissProt database using Mascot using the parameters described in ref 102. Protein identifications were established at <5.0% protein FDR and <1.0% peptide FDR in Scaffold v.5 (Proteome Software), and proteins with a minimum of 97% protein identification probability and at least two unique peptides were accepted. The top 15 proteins (by number of PSMs) per sample source were visualized as a treemap and labeled by their corresponding gene name; trypsin, keratins, serum albumin, and microbial proteins were excluded from the analysis. *Ovostatin; **riboflavin-binding protein; ***B3-hordein.

Warinner et al., 2022 DOI: 10.1021/acs.chemrev.1c00703

DINOSAUR FEATHERS

SEPTEMBER 22, 2023

Dinosaur feathers contain traces of ancient proteins, study finds

- „Modern bird feathers more stiff compared to dinosaur“

- Keratins (formerly α-keratins) – softer
- Corneous β-proteins (formerly β-keratins) – stiff
- Modern bird feather rich in β-keratins – stronger for flight

- Further research shows the differences were probably due to taphonomy – change observed in heated feathers (β » α)

- „you can't read the fossil record literally“

RESEARCH ARTICLE | BIOLOGICAL SCIENCES |

The molecular evolution of feathers with direct evidence from fossils

Yanhong Pan, Wenxia Zheng, Roger H. Sawyer, Michael W. Pennington, Xiaoting Zheng, Xiaoli Wang, Min Wang, Liang Hu, Jingmai O'Connor, Tao Zhao, Zhiheng Li, Elena R. Schroeter, Feixiang Wu, Xing Xu, Zhonghe Zhou, and Mary H. Schweitzer -12 Authors Info & Affiliations

Contributed by Zhonghe Zhou, December 15, 2018 (sent for review September 12, 2018; reviewed by Dominique G. Homberger and Chenxi Jia)

January 28, 2019 | 116 (8) 3018-3023 | https://doi.org/10.1073/pnas.1815703116

Article | Published: 21 September 2023

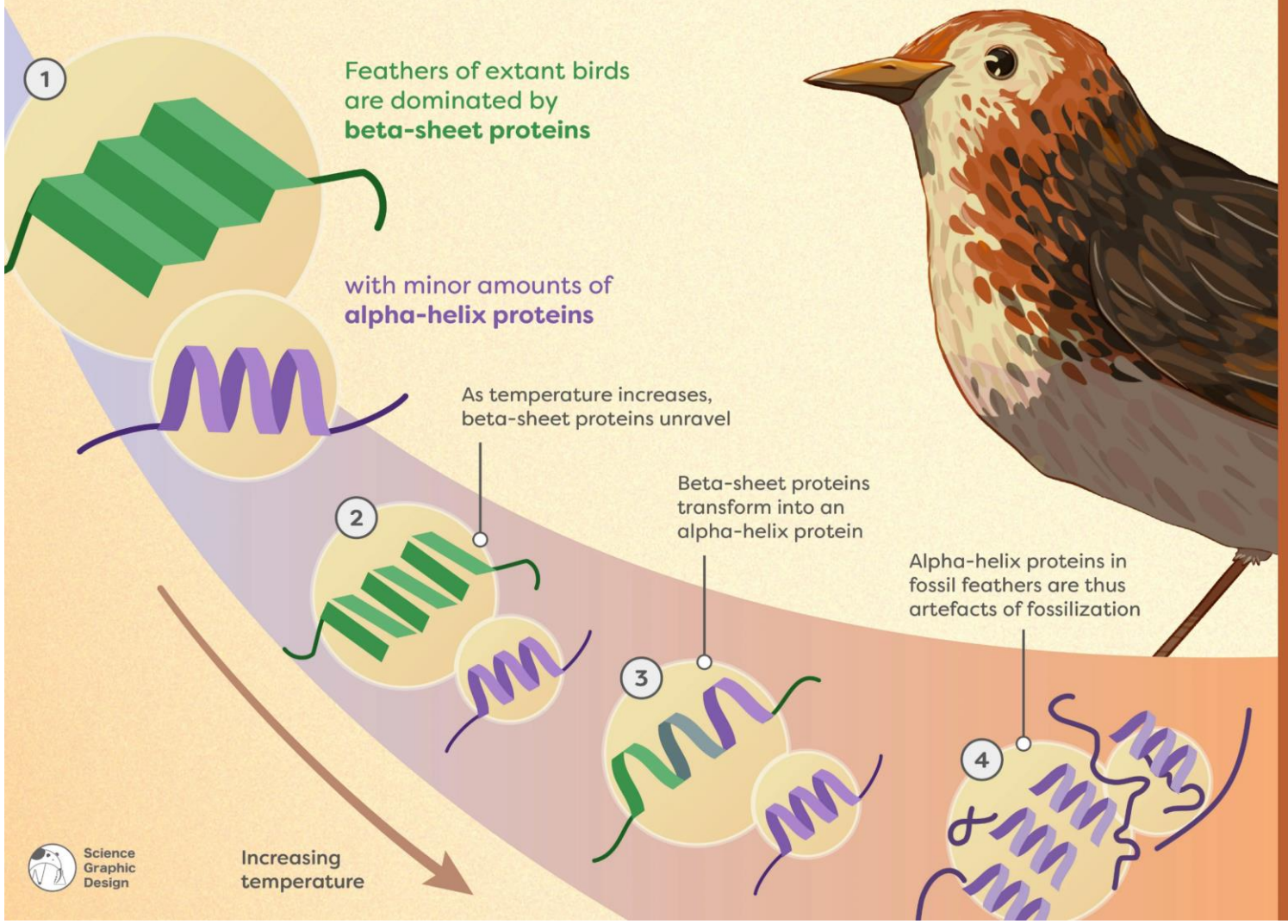
Preservation of corneous β-proteins in Mesozoic feathers

Tiffany S. Slater, Nicholas P. Edwards, Samuel M. Webb, Fucheng Zhang & Maria E. McNamara

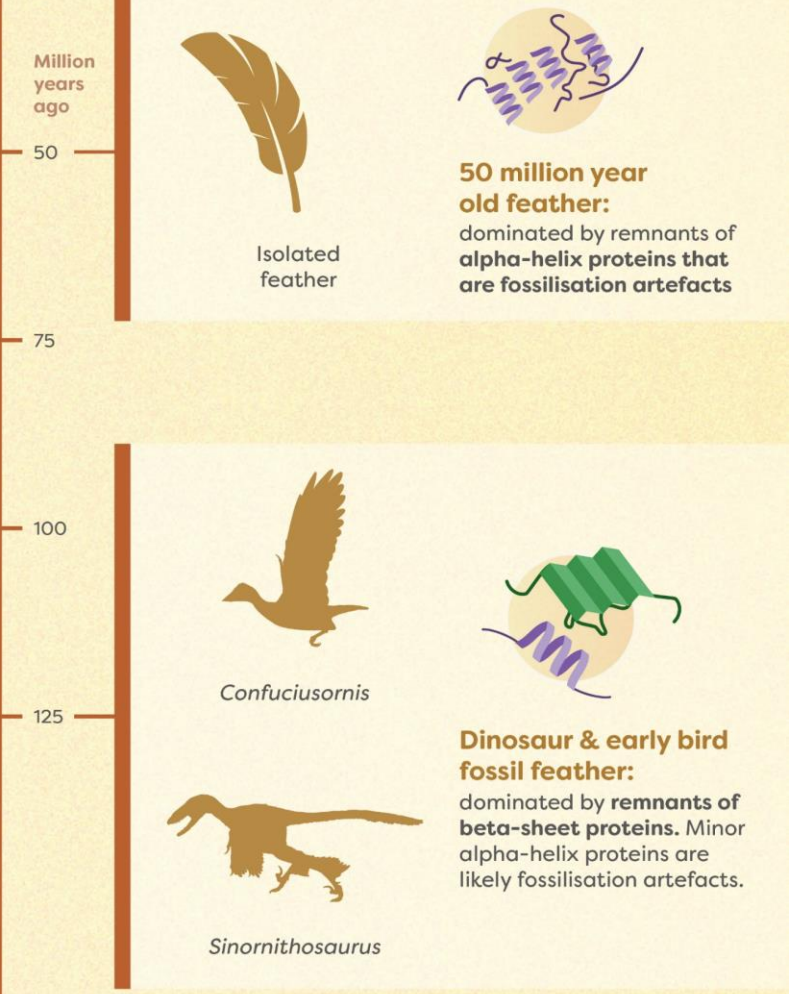
Nature Ecology & Evolution **7**, 1706–1713 (2023) | Cite this article

Ancient protein traces in fossil feathers

Our experiments found that feather protein remnants can survive during fossilisation
but beta-sheets alter to alpha-helices with increasing temperature



These experiments explain fossil feather chemistry



Slater et al., 2023 DOI: 10.1038/s41559-023-02177-8

METAPROTEOMS

- Microbiomes
- Food remains
- Pathogens and disease
- Cultural heritage

- Missing taxa in databases – often palaeoproteomic studies add both modern and ancient proteomes to databases

DIET AND SUBSISTENCE PRACTICES

- ZooMS
- Food remains, crusts …
- Fermentation
- Calculus
- Plant microremains

DIET AND SUBSISTENCE PRACTICES

- ZooMS
- Food remains, crusts …
  - Mostly ceramics
  - Calcified and charred food
  - Ingredients as well as processing
- Fermentation
- Calculus
- Plant microremains

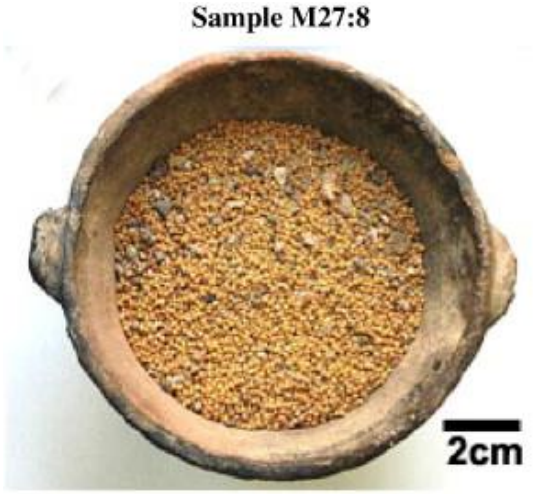
ELSEVIER

Journal of Proteomics
Volume 105, 13 June 2014, Pages 363-371

Proteomics identifies the composition and manufacturing recipe of the 2500-year old sourdough bread from Subeixi cemetery in China ☆

Anna Shevchenko [a 1], Yimin Yang [b c 1], Andrea Knaust [a], Henrik Thomas [a], Hongen Jiang [b], Enguo Lu [d], Changsui Wang [b] ✉, Andrej Shevchenko [a] ✉

- Sourdough bread made from millet and barley
- Fermenting with baker's yeast and lactic acid bacteria
- Broom corn (but not foxtail) millet



DIET AND SUBSISTENCE PRACTICES

- ZooMS
- Food remains, crusts …
- Fermentation
- Calculus
- Plant microremains
  - Usually waterlogged, mineralised, or charred seeds
  - Arid and/or cold areas
  - Low protein recovery compared to carbohydrates and lipids in seeds

Naturwissenschaften (2010) 97:205–217
DOI 10.1007/s00114-009-0629-3

ORIGINAL PAPER

A multidisciplinary study of archaeological grape seeds

Enrico Cappellini · M. Thomas P. Gilbert · Filippo Geuna · Girolamo Fiorentino · Allan Hall · Jane Thomas-Oates · Peter D. Ashton · David A. Ashford · Paul Arthur · Paula F. Campos · Johan Kool · Eske Willerslev · Matthew J. Collins

- DNA + carbohydrates + proteins from medieval *Vitis vinifera*

A guide to ancient protein studies

Jessica Hendy[1,9*], Frido Welker[2,3,9*], Beatrice Demarchi[4,5], Camilla Speller[5], Christina Warinner[6,7,8] and Matthew J. Collins[3,5]

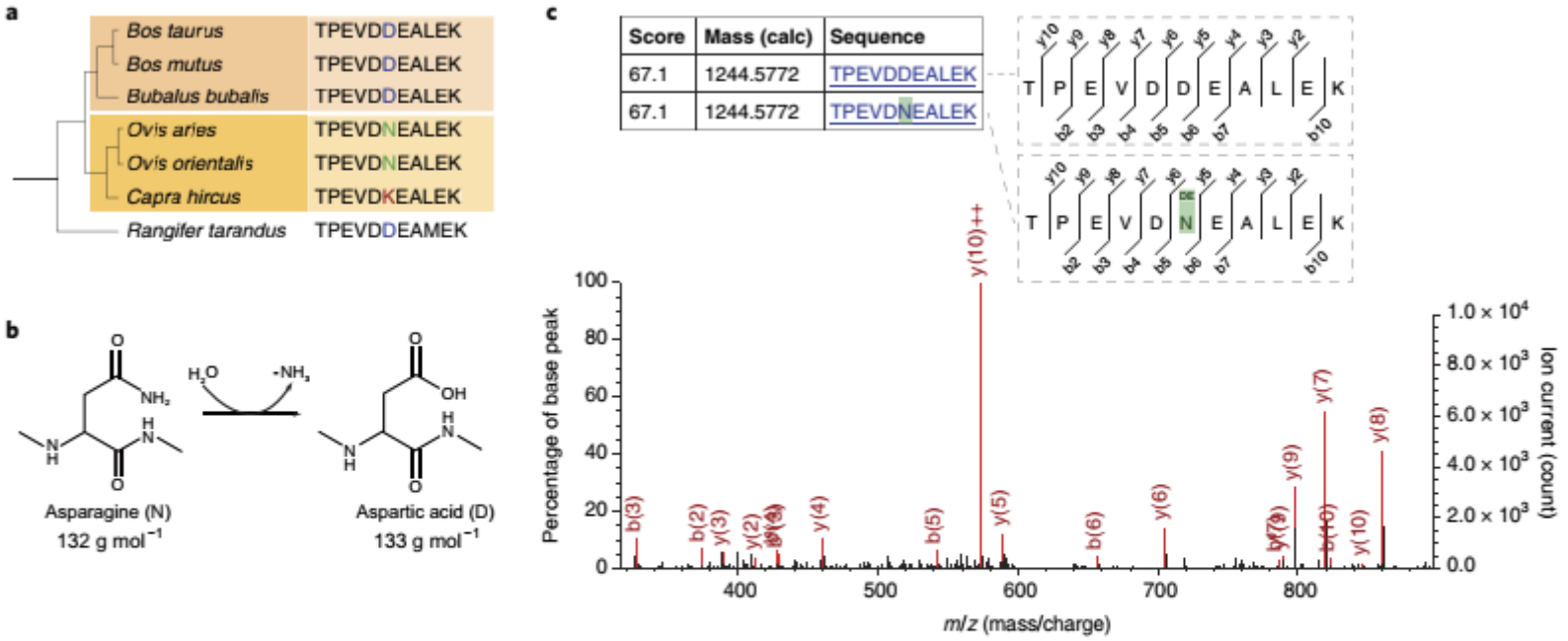


Fig. 4 | Damage-induced sequence ambiguity affects peptide taxonomic assignment for the whey protein beta-lactoglobulin. a, An important variant site that distinguishes Bovinae from Caprinae is an amino acid residue that is aspartic acid (D) in Bovinae, asparagine (N) in sheep and lysine (K) in goats. **b**, However, the deamidation of aspragine (N) results in its conversion to aspartic acid (D). **c**, Protein identification software is unable to distinguish an unmodified Bovinae residue (D) from a deamidated sheep residue (DE N) at this position. Data from ref. [12] demonstrate a Mascot output, including a b- and y-ion series, for this scenario. Incorrect precursor selection during MS analysis can result in similar ambiguity by selecting isotope-derived peaks for fragmentation, instead of the monoisotopic peak.

HEALTH AND DISEASE

- Pathogens
  - Better phylogenetic resolution with DNA
  - Enables research of pathophysiology
  - Often impossible to discern from other bacteria (e.g. *Mycobacterium*)
  - Best in combination with palaeogenomics (*Mycobacterium leprae* in calculus)

Research articles

Multi-omic detection of *Mycobacterium leprae* in archaeological human dental calculus

Anna K. Fotakis, Sean D. Denham, Meaghan Mackie, Miren Iraeta Orbegozo, Dorothea Mylopotamitaki, Shyam Gopalakrishnan, Thomas Sicheritz-Pontén, Jesper V. Olsen, Enrico Cappellini, Guojie Zhang, Axel Christophersen, M. Thomas P. Gilbert and Åshild J. Vågene See fewer authors

Published: 05 October 2020 https://doi.org/10.1098/rstb.2019.0584

- Palaeopathology
- Palaeogenomic analysis conclusive
- Only 4 proteins assigned to *Mycobacterium* genus

HEALTH AND DISEASE

- Pathogens
  - Better phylogenetic resolution with DNA
  - Enables research of pathophysiology
  - Often impossible to discern from other bacteria (e.g. *Mycobacterium*)
  - Best in combination with palaeogenomics (*Mycobacterium leprae* in calculus)

RESEARCH ARTICLE

Paleoproteomics of the Dental Pulp: The plague paradigm

Rémi Barbieri[1☯], Rania Mekni[1☯], Anthony Levasseur[1], Eric Chabrière[1], Michel Signoli[2], Stéfan Tzortzis[2], Gérard Aboudharam[1], Michel Drancourt[1]*

Table 2. List of four peptides retrieved from three individuals in a documented 18th century plague site, France; exhibiting 100% identity and 100% coverage (Blast on NCBI) with at least *Y. pestis*. * This peptide was found twice in the S22 individual.

Peptide	Specimen	Organism
(-)GIVYNPDNVADGFYYAEGGNFVQIYQYENPMFFEK(E) *	S22	*Yersinia pestis*
		Yersinia pseudotuberculosis complex
(K)LYDAANAALDVVDTEIAQGFPEPEWATQLREAIAEMNAPEPSEDEADWQR(F)	S16	*Yersinia pestis*
		Buttiauxella gaviniae
		Enterobacter cloacae
		Enterobacter hormaechei
		Enterobacteriaceae
		Klebsiella oxytoca
		Salmonella enterica
		Salmonella phage
(R)QSPEMDYFMAVFVPSFSLSLDEISLDSLD(-)	S23	*Yersinia pestis*
		Yersinia
		Yersinia frederiksenii
		Yersinia intermedia
		Yersinia pseudotuberculosis
		Yersinia wautersii
(R)KFNGNLNAER(I)	S23	*Yersinia pestis*
		bacteria symbiont BFo1 of *Frankliniella occidentalis*
		Brenneria goodwinii
		Chania multitudinisentens
		Enterobacter ludwigii
		Enterobacterales
		Erwinia
		Erwinia billingiae
		Erwinia persicina
		Erwinia typogaphi
		Ewingella americana
		Pantoea ananatis
		Pantoea sp.
		Pantoea stewartii
		Rahnella
		Serratia
		Serratia fonticola
		Yersinia pseudotuberculosis
		Yersinia ruckeri

HEALTH AND DISEASE

- Pathogens
  - Better phylogenetic resolution with DNA
  - Enables research of pathophysiology
  - Often impossible to discern from other bacteria (e.g. *Mycobacterium*)
  - Best in combination with palaeogenomics (*Mycobacterium leprae* in calculus)

- Immunity

Pathogens and host immunity in the ancient human oral cavity

Christina Warinner[1,2], João F Matias Rodrigues[3,4], Rounak Vyas[3,4], Christian Trachsel[5], Natallia Shved[1], Jonas Grossmann[5], Anita Radini[6,7], Y Hancock[8], Raul Y Tito[2], Sarah Fiddyment[6], Camilla Speller[6], Jessica Hendy[6], Sophy Charlton[6], Hans Ulrich Luder[9], Domingo C Salazar-García[10–12], Elisabeth Eppler[13,14], Roger Seiler[1], Lars H Hansen[15,16], José Alfredo Samaniego Castruita[17], Simon Barkow-Oesterreicher[5], Kai Yik Teoh[6], Christian D Kelstrup[18], Jesper V Olsen[18], Paolo Nanni[5], Toshihisa Kawai[19,20], Eske Willerslev[17], Christian von Mering[3,4], Cecil M Lewis Jr[2], Matthew J Collins[6], M Thomas P Gilbert[17,21], Frank Rühli[1,22] & Enrico Cappellini[17,22]

Calcified dental plaque (dental calculus) preserves for millennia and entraps biomolecules from all domains of life and viruses. We report the first, to our knowledge, high-resolution taxonomic and protein functional characterization of the ancient oral microbiome and demonstrate that the oral cavity has long served as a reservoir for bacteria implicated in both local and systemic disease. We characterize (i) the ancient oral microbiome in a diseased state, (ii) 40 opportunistic pathogens, (iii) ancient human–associated putative antibiotic resistance genes, (iv) a genome reconstruction of the periodontal pathogen *Tannerella forsythia*, (v) 239 bacterial and 43 human proteins, allowing confirmation of a long-term association between host immune factors, 'red complex' pathogens and periodontal disease, and (vi) DNA sequences matching dietary sources. Directly datable and nearly ubiquitous, dental calculus permits the simultaneous investigation of pathogen activity, host immunity and diet, thereby extending direct investigation of common diseases into the human evolutionary past.

HEALTH AND DISEASE

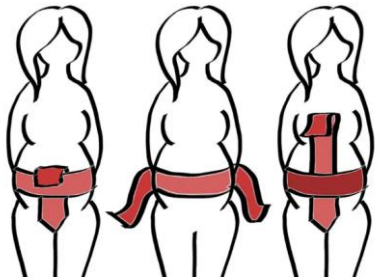
- Pathogens
  - Better phylogenetic resolution with DNA
  - Enables research of pathophysiology
  - Often impossible to discern from other bacteria (e.g. *Mycobacterium*)
  - Best in combination with palaeogenomics (*Mycobacterium leprae* in calculus)
- Immunity
- Medicinal practise

Girding the loins? Direct evidence of the use of a medieval English parchment birthing girdle from biomolecular analysis

Sarah Fiddyment, Natalie J. Goodison, Elma Brenner, Stefania Signorello, Kierri Price and Matthew J. Collins

Published: 10 March 2021 | https://doi.org/10.1098/rsos.202055

- Evidence for the use of honey, cereals, ovicaprine milk and legumes
- Cervico-vaginal fluid
- Opens up research of stains



A multidisciplinary approach for investigating dietary and medicinal habits of the Medieval population of Santa Severa (7th-15th centuries, Rome, Italy)

Angelo Gismondi[1], Marica Baldoni[2], Micaela Gnes[2], Gabriele Scorrano[2], Alessia D'Agostino[1], Gabriele Di Marco[1], Giulietta Calabria[2], Michela Petrucci[2], Gundula Müldner[3], Matthew Von Tersch[4], Alessandra Nardi[5], Flavio Enei[6], Antonella Canini[1], Olga Rickards[2], Michelle Alexander[4], Cristina Martínez-Labarga[2]*

- „stable isotope analysis from bone proteins and investigations on dental calculus using DNA analysis, light microscopy, and gas chromatography coupled with mass spectrometry“
- „knowledge of ethnopharmacological tradition and the application of medicinal plants (e.g. *Punica granatum* L., *Ephedra sp.* L.) were also identified“
- Artemisinin

CULTURAL HERITAGE

Proteins in Art, Archaeology, and Paleontology: From Detection to Identification

Sophie Dallongeville[†], Nicolas Garnier[‡], Christian Rolando[†], and Caroline Tokarski[*†]

View Author Information

Cite this: *Chem. Rev.* 2016, 116, 1, 2–79
Publication Date: December 28, 2015
https://doi.org/10.1021/acs.chemrev.5b00037

Extensive review – voluntary reading

- Paint
- Glue, binders

CULTURAL HERITAGE

Proteins in Art, Archaeology, and Paleontology: From Detection to Identification

Sophie Dallongeville[†], Nicolas Garnier[‡], Christian Rolando[†], and Caroline Tokarski[*†]

View Author Information

Cite this: *Chem. Rev.* 2016, 116, 1, 2–79
Publication Date: December 28, 2015
https://doi.org/10.1021/acs.chemrev.5b00037

- Paint
- Glue, binders
- Mortars

Case study

Application of peptide mass mapping on proteins in historical mortars

Stepanka Kuckova [a b c], Radovan Hynek [c], Milan Kodicek [c]

Show more

https://doi.org/10.1016/j.culher.2008.06.013

Table 1. Summary of protein additives in mortars and their classification by their effects on fresh and hardened mortars.

Effect	Protein additives
Accelerator of hardening	Egg white, blood, curd
Retarder of hardening	Egg white, blood
Plasticizer	Milk, egg white
Adhesive	Casein, animal glues, gelatin
Firmer	Milk, egg white, casein, cheese, blood

Some of the protein additives are matched into more, sometimes even contradicting, categories, due to their different effect on fresh and moderately hardened mortars.

CULTURAL HERITAGE

Proteins in Art, Archaeology, and Paleontology: From Detection to Identification

Sophie Dallongeville[†], Nicolas Garnier[‡], Christian Rolando[†], and Caroline Tokarski[*†]

View Author Information

Cite this: *Chem. Rev.* 2016, 116, 1, 2–79
Publication Date: December 28, 2015
https://doi.org/10.1021/acs.chemrev.5b00037

- Paint
- Glue, binders
- Mortars

- Coffins, clothing, tools, toys, religious objects, kayaks, paintings …

- Tissue type

Cite this: *Analyst*, 2013, **138**, 4849

Identification of collagen-based materials in cultural heritage

Daniel P. Kirby,*[a] Michael Buckley,[b] Ellen Promise,[c] Sunia A. Trauger[d] and T. Rose Holdcraft[c]

Cite this: *Analyst*, 2013, **138**, 4849

Identification of collagen-based materials in cultural heritage

Daniel P. Kirby,*[a] Michael Buckley,[b] Ellen Promise,[c] Sunia A. Trauger[d] and T. Rose Holdcraft[c]

Table 2 A selection of the Alaskan Native objects analyzed and identified through PMF covering a wide variety of object types and materials. Numbers in parentheses are Peabody Museum of Archaeology and Ethnology accession numbers

Object	Sampling location	Museum ID	PMF ID
Gutskin bag (2103)	Gut	Seal	Eared seal
Gutskin bag (2103)	Painted stripe	Seal	Seal (Phocidae/phocini)
Gutskin bag (2103)	Sinew	Unknown	Caribou
Gutskin bag (48414)	Gut	Walrus or sea lion	Eared seal
Gutskin bag (48414)	Black border	Walrus or sea lion	Bear
Gutskin bag (48414)	Sinew	Unknown	Caribou
Gutskin bag (76018)	Red painted stripe	Bear	Eared seal
Gutskin bag (76018)	Gut	Bear	Eared seal
Gutskin bag (76018)	Border sinew	Unknown	Eared seal
Gutskin bag (76018)	Embroidery sinew	Unknown	Dog/wolf
Gutskin bag (48414.1)	Gut	Unknown	Bear
Gutskin bag (48414.1)	Red and green border	Unknown	Bear
Gutskin bag (48414.1)	Sinew	Unknown	Caribou
Gutskin cap (48415)	Gut	Seal	Eared seal
Gutskin cap (48415)	Painted border	Unknown	Seal (Phocidae/phocini)
Gutskin cap (48415)	Sinew	Unknown	Caribou
Spear with pouch and sinew (1620)	Pouch	Sea lion	Eared seal
Spear with pouch and sinew (1620)	Sinew	Whale	Caribou
Gutskin coat (56749)	Gut	Seal	Eared seal
Gutskin coat (56749)	Black overlay	Unknown	Eared seal
Gutskin coat (56749)	Sinew	Unknown	Eared seal
Kayak model (1203)	Skin	Unknown	Seal (Phocidae/phocini)
Kayak model (1203)	Sinew	Unknown	Right whale
Kayak model (1204)	Skin	Sea lion	Eared seal
Kayak model (1204)	Sinew	Unknown	Right whale
Kayak model (11255)	Skin	Unknown	Eared seal
Kayak model (11255)	Gut	Unknown	Eared seal
Kayak model (11255)	Sinew	Unknown	Fin whale
Kayak model (11256)	Skin	Unknown	Humpback whale
Kayak model (11256)	Gut	Unknown	Eared seal

CULTURAL HERITAGE

Proteins in Art, Archaeology, and Paleontology: From Detection to Identification

Sophie Dallongeville[†], Nicolas Garnier[‡], Christian Rolando[†], and Caroline Tokarski[*†]

View Author Information

Cite this: *Chem. Rev.* 2016, 116, 1, 2−79
Publication Date: December 28, 2015
https://doi.org/10.1021/acs.chemrev.5b00037

- Paint
- Glue, binders
- Mortars

- Coffins, clothing, tools, toys, religious objects, kayaks, paintings …

- Tissue type

- Biocodicology

Cite this: *Analyst*, 2013, **138**, 4849

Identification of collagen-based materials in cultural heritage

Daniel P. Kirby,*[a] Michael Buckley,[b] Ellen Promise,[c] Sunia A. Trauger[d] and T. Rose Holdcraft[c]

CULTURAL HERITAGE

Proteins in Art, Archaeology, and Paleontology: From Detection to Identification

Sophie Dallongeville[†], Nicolas Garnier[‡], Christian Rolando[†], and Caroline Tokarski[*†]

View Author Information

Cite this: *Chem. Rev.* 2016, 116, 1, 2–79
Publication Date: December 28, 2015
https://doi.org/10.1021/acs.chemrev.5b00037

- Paint
- Glue, binders
- Mortars

- Coffins, clothing, tools, toys, religious objects, kayaks, paintings …

- Tissue type

- Biocodicology

Species identification of ivory and bone museum objects using minimally invasive proteomics

CATHERINE GILBERT, VACLAV KRUPICKA, FRANCESCA GALLUZZI, ALEKSANDRA POPOWICH, KATELL BATHANY, STÉPHANE CLAVEROL, JULIE ARSLANOGLU, AND CAROLINE TOKARSKI Authors Info & Affiliations

SCIENCE ADVANCES · 26 Jan 2024 · Vol 10, Issue 4 · DOI: 10.1126/sciadv.adi9028

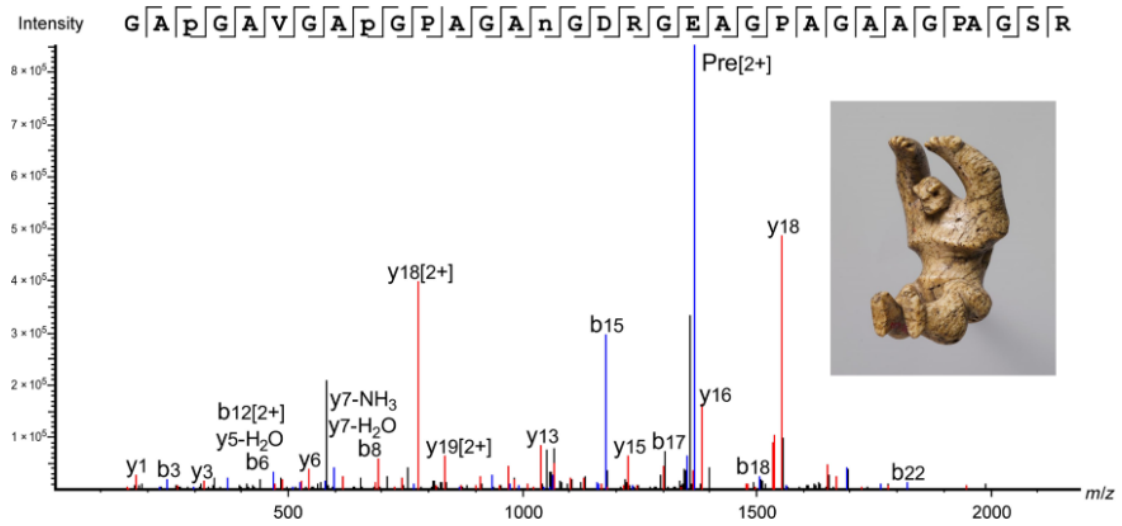


Fig. 2. Sperm whale peptide marker identified in Hawaii bone pendant sample—1979.206.1587, 18th to 19th century, The Metropolitan Museum of Art.
Peptide from collagen α2(i), unique to *P. macrocephalus*, detected in object 1979.206.1587, but also identified in object 2012.346. GAP(+15.99)GAVGAP(+15.99)GPAGAN(+0.98)GDRGEAGPAGAAGPAGSR, *m/z*: 1366.1423, *z*: 2, parts per million (ppm): 0.1.

A guide to ancient protein studies

Jessica Hendy[1,9*], Frido Welker[2,3,9*], Beatrice Demarchi[4,5], Camilla Speller[5], Christina Warinner[6,7,8] and Matthew J. Collins[3,5]

- Preservation of proteins is higher
- Different information than DNA
  - Lower taxonomical resolution, tissue specifity, active processes

- No latex gloves, wool, silk, leather or exposed skin
- Ancient biomolecules laboratories

- Feasibility of a few samples

- Blanks, injections of blanks between samples in LC-MS/MS
- Inject oldest and most precious samples first
- Diagenesis (mostly deamidation)

- Contaminating taxa in search (cRAP - common Repository of Adventitious Proteins)

- Share data in public repositories

- Combine with other methods!

- Dinosaurs, beta and aplha keratins, taphonomy
- Birthing girdles, analysis of stains
- Kayaks from multiple species
- Grape and all seeds
- Sex determination

CASE STUDIES

- Groups of 2 or 3
- Pick a case

- Individual proposal
- Brainstorming together only AFTER individual work

- Project proposal is only a suggestion, you can think of very different approach!
- Try what you personally would like to or could do

- Scaleable – try showing cost-effective, minimal scenario, as well as wild research fantasy without funding limits

- Project proposal (max 1 page, no minimum, bullet points allowed) BEFORE May 15th
- 10 min presentation of the project on May 15th
  - PowerPoint / no props / science slam style / shark tank / (almost) anything goes

EXAM

- Suggestion for final exam: everyone together with coffee and cookies, defense afterwards
- Wednesday, 29th May, 2pm ?