Developing Proteomic Genotyping for the Common Good

UC Davis Proteomics

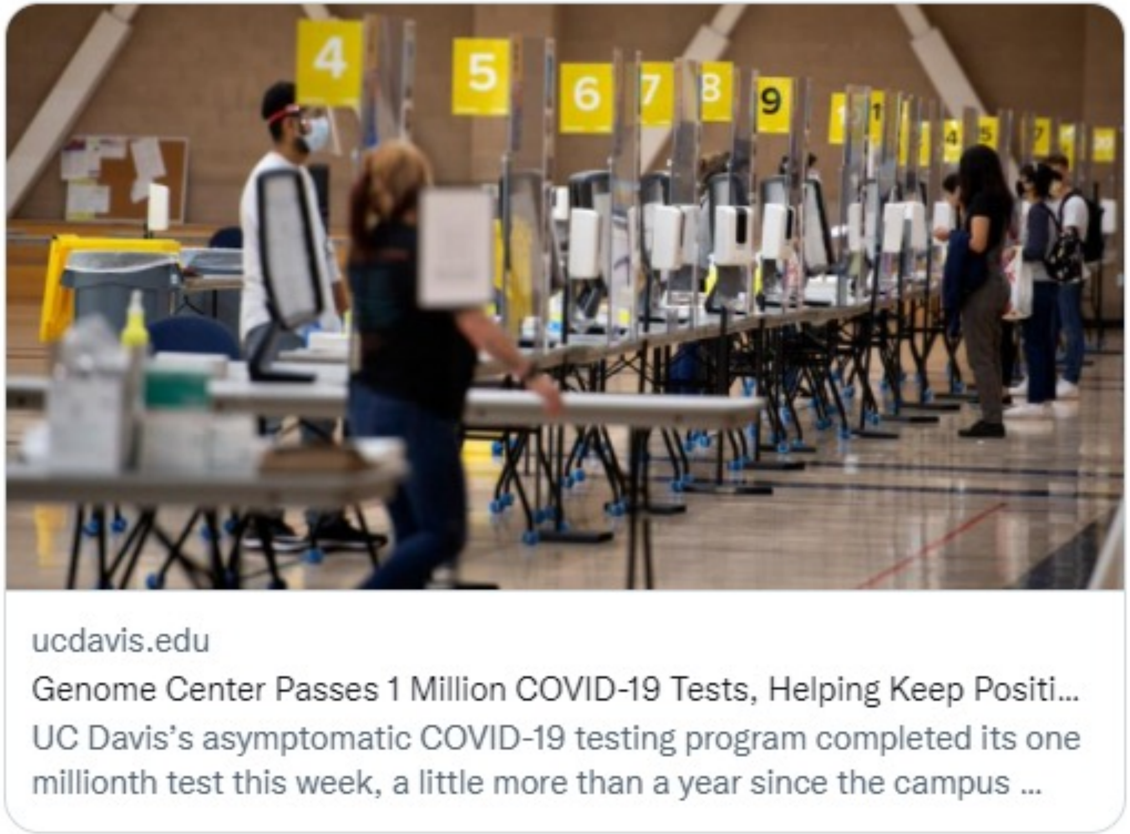
Proteomics.ucdavis.edu
blusky=@ucdproteomics.bsky.social

UC Davis Proteomics Core

- Established 2005
- Averages between 3-5 Staff Scientists and 3-5 undergraduate students
- Over 150 Authored publications by core scientists
  - Many more non authored (= acknowledged)
- 3 successful NIH S10 grants
- Generates typically between 600-800K a year in revenue
- Offers Online and in person (hands on classes) almost every year
  - https://video.ucdavis.edu/playlist/details/0_4jkc4swu - Online Videos
- Usually involved and spearheads several international proteins studies a year though the ABRF Proteomics Research group
  - 2020-2021 Pandemic Proteomic Beer Study
  - 2021-2022 Prote”omlet” egg glycopeptide study
- Only academic lab in the world that still does Edman sequencing and Amino Acid analysis

UC Davis Genome Center

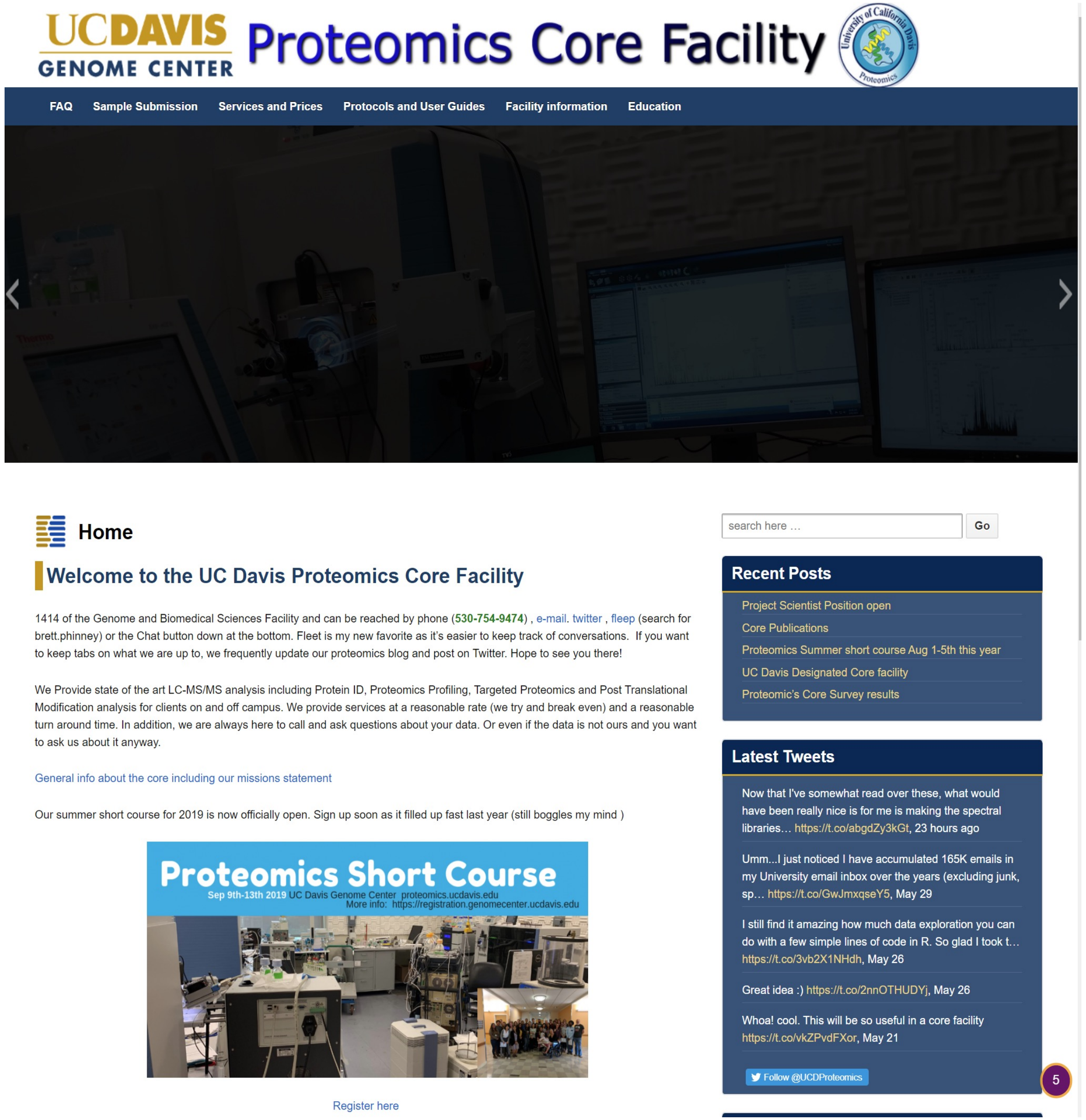
proteomics.ucdavis.edu



ucdavis.edu

Genome Center Passes 1 Million COVID-19 Tests, Helping Keep Positi...

UC Davis's asymptomatic COVID-19 testing program completed its one millionth test this week, a little more than a year since the campus ...



Home

Welcome to the UC Davis Proteomics Core Facility

1414 of the Genome and Biomedical Sciences Facility and can be reached by phone (530-754-9474) , e-mail. twitter , fleep (search for brett.phinney) or the Chat button down at the bottom. Fleet is my new favorite as it's easier to keep track of conversations. If you want to keep tabs on what we are up to, we frequently update our proteomics blog and post on Twitter. Hope to see you there!

We Provide state of the art LC-MS/MS analysis including Protein ID, Proteomics Profiling, Targeted Proteomics and Post Translational Modification analysis for clients on and off campus. We provide services at a reasonable rate (we try and break even) and a reasonable turn around time. In addition, we are always here to call and ask questions about your data. Or even if the data is not ours and you want to ask us about it anyway.

General info about the core including our missions statement

Our summer short course for 2019 is now officially open. Sign up soon as it filled up fast last year (still boggles my mind)

Register here

Recent Posts

- Project Scientist Position open
- Core Publications
- Proteomics Summer short course Aug 1-5th this year
- UC Davis Designated Core facility
- Proteomic's Core Survey results

Latest Tweets

Now that I've somewhat read over these, what would have been really nice is for me is making the spectral libraries… https://t.co/abgdZy3kGt, 23 hours ago

Umm...I just noticed I have accumulated 165K emails in my University email inbox over the years (excluding junk, sp… https://t.co/GwJmxqseY5, May 29

I still find it amazing how much data exploration you can do with a few simple lines of code in R. So glad I took t… https://t.co/3vb2X1NHdh, May 26

Great idea :) https://t.co/2nnOTHUDYj, May 26

Whoa! cool. This will be so useful in a core facility https://t.co/vkZPvdFXor, May 21

Some of the samples in our Core Facility in the last year or so

- Human Hair & Skin & Fingermarks & 11K year old human teeth!
- Grape Sap
- Bull sperm cells
- Ferret Sperm
- Yeast/Beer
- Egg Whites
- All sorts of BioID experiments
- All sorts of PTMS (phospho, Sumo, ubiq)
- All sorts of livers ,Fish, mouse, human
- Walnut Bark and Pellicle (walnut skin)
- Tardigrades
- Horse Lung Lavage (horse snot)
- Dog tissue
- Sea Otter hearts (diseased cardiomyopathy, very fibrotic)
- Brain inclusion bodies
- Bovine uterine fluid
- Isolated HDL particles from serum
- Wheat
- Milk - human, cow, formula
- Plasma, human, mouse
- Mouse Knee tissue
- Covid Nasal Swabs
- Some weird sample that was green!

Horse Snot Collaboration with the Proteomics Core Here at OHSU

News & Research

Study identifies possible factor in newborn foals being prone to lung infections

September 7, 2023 Horsetalk.co.nz 0 Comments 2 min read

Share

Image by rihaij

Newborn foals appear to have a lower relative abundance of immune-related proteins in their lung lavage fluid than older foals and adult horses, researchers report.

You want me to put this in my mass spec?

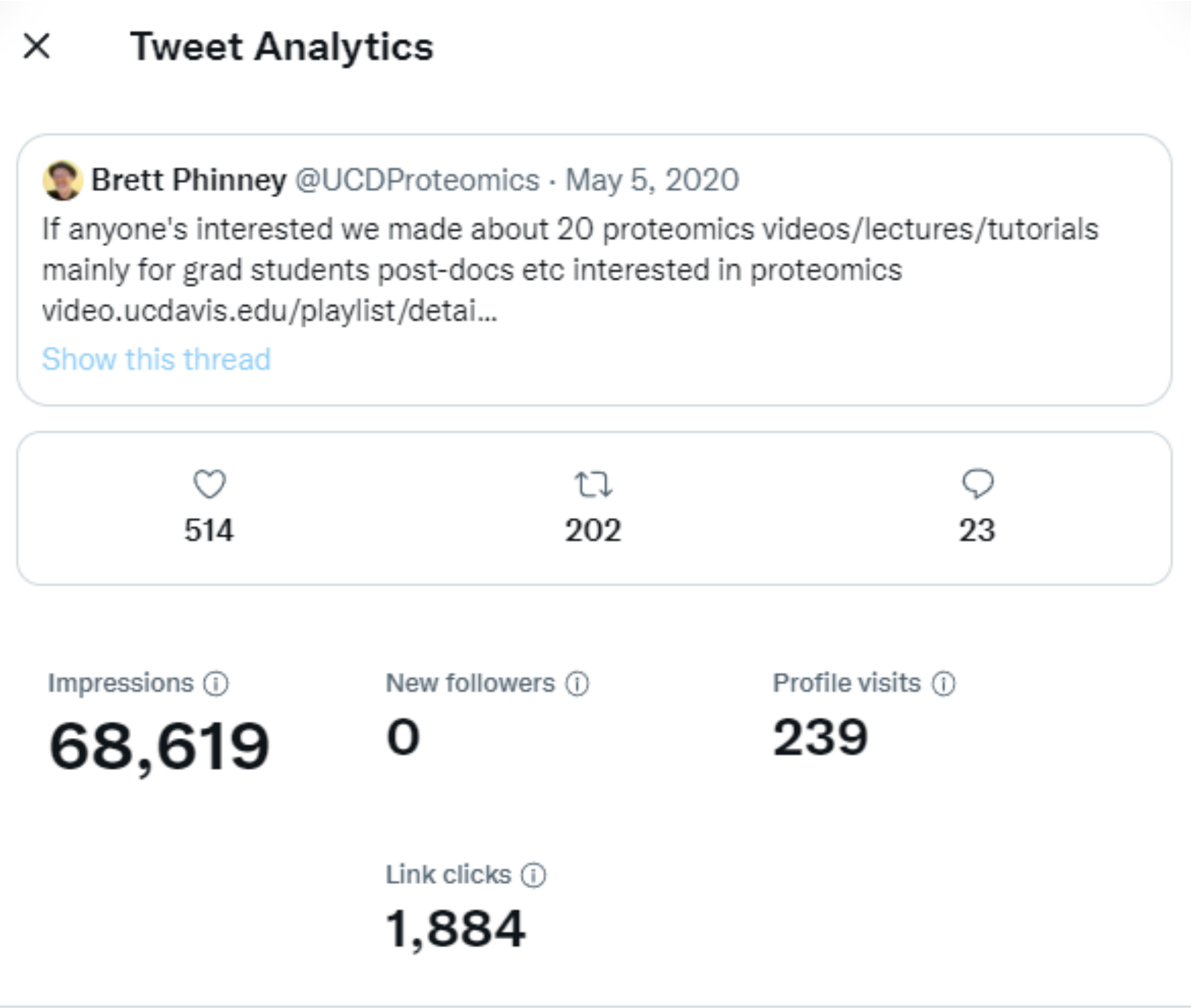
2023 course was last month

Sample prep/ LCMS / analysis

Virtual Class

- We did a virtual Pandemic Class available here

https://video.ucdavis.edu/playlist/details/0_4jkc4swu



Tweet Analytics

Brett Phinney @UCDProteomics · May 5, 2020

If anyone's interested we made about 20 proteomics videos/lectures/tutorials mainly for grad students post-docs etc interested in proteomics video.ucdavis.edu/playlist/detai...

Show this thread

Likes	Retweets	Replies
514	202	23

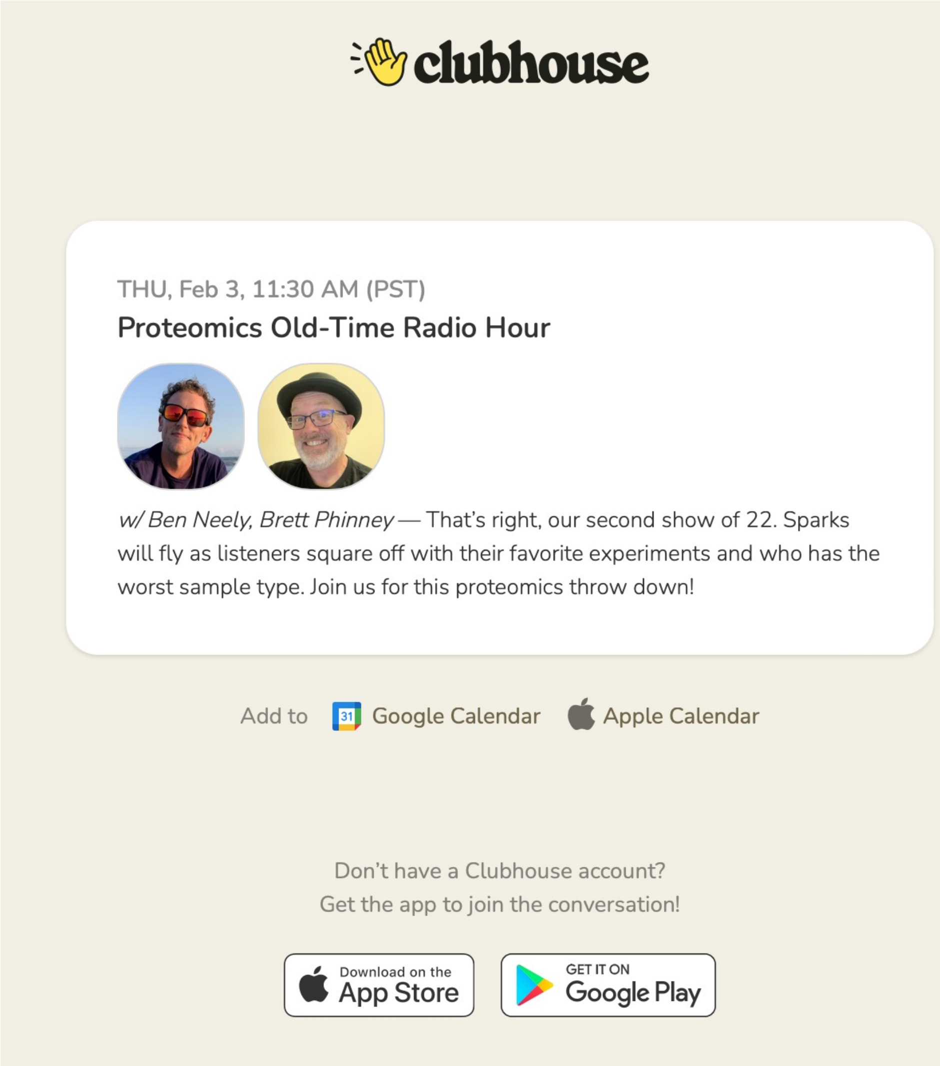
Impressions: 68,619

New followers: 0

Profile visits: 239

Link clicks: 1,884

Proteomics Community Building



THU, Feb 3, 11:30 AM (PST)

Proteomics Old-Time Radio Hour

w/ Ben Neely, Brett Phinney — That's right, our second show of 22. Sparks will fly as listeners square off with their favorite experiments and who has the worst sample type. Join us for this proteomics throw down!

Don't have a Clubhouse account?
Get the app to join the conversation!

Every other week clubhouse chat

Our Proteomics radio hour has been getting more popular

The 2021 ABRF Beer Study: Beer Proteomics at the Global Scale

- Preprint with 74 of co-authors is soon to be out!
- Largest beer proteomics resource ever (we think)

Brett Phinney, Andrew Marcus, Glen Fox, Hua Ding, Laura E Herring, Pratik D. Jagtap, Joanna Kirkpatrick, Vikas Kumar, Mukul K Midha, Leroy Martin, Magnus Palmblad, Baozhen Shan, Paul M Stemmer, Yan Wang, Dan Polasky, Austin Carr, Michael Shortreed, Benjamin A. Neely

The Fundemic Beer Project

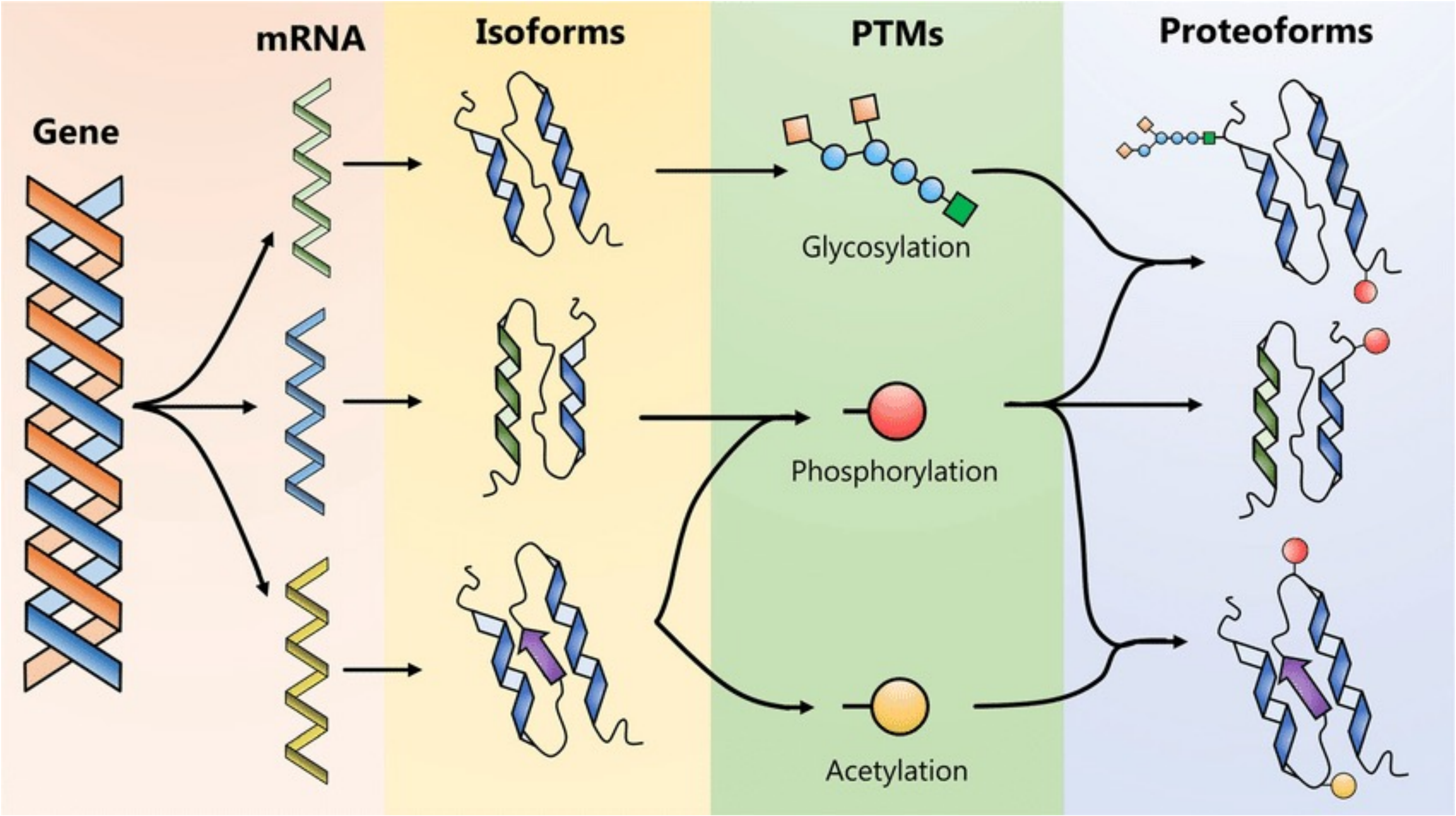
UC Davis Brewhouse

MassIVE MSV000088080 Partial Public

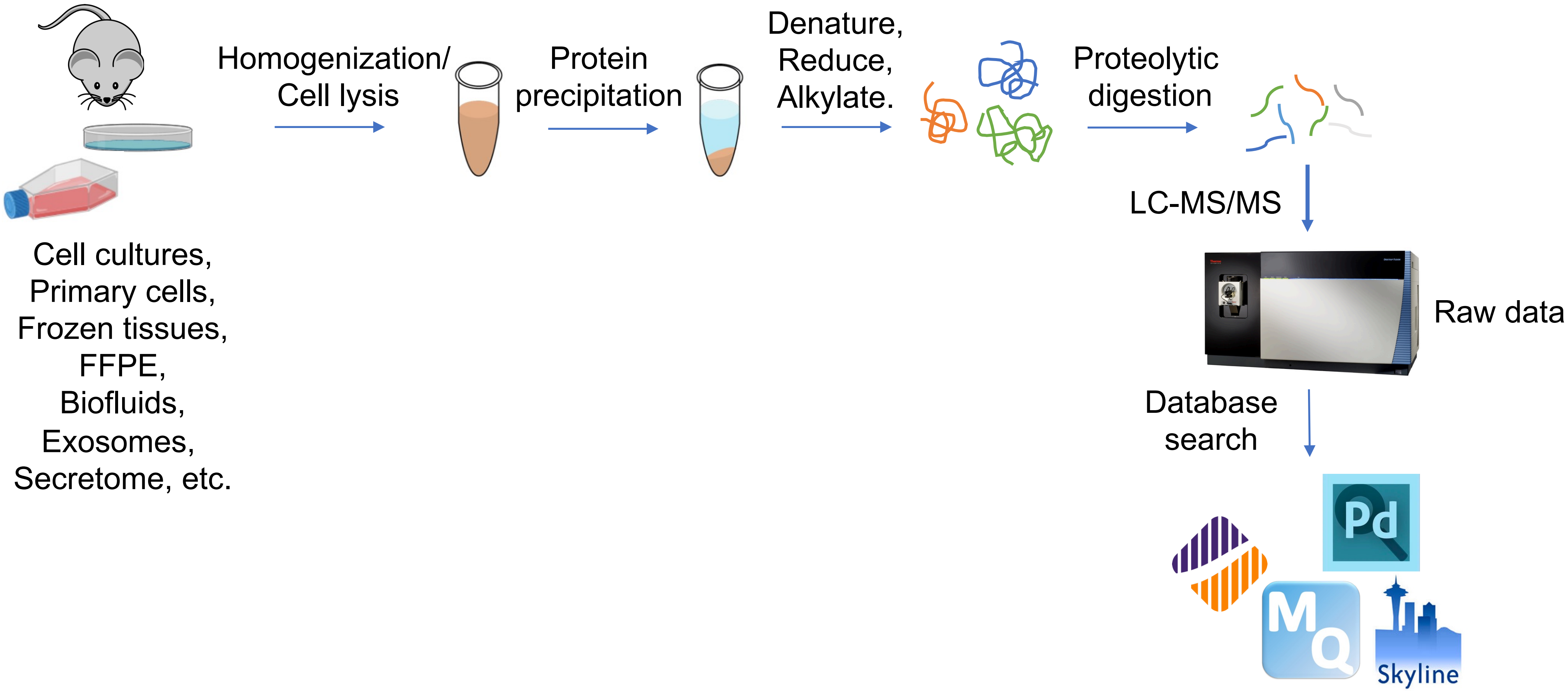
The 2020 ABRF Beer Study: beer proteomics at the global scale

357 injections

Complexity & Variation: Isoforms and Proteoforms



A typical workflow in bottom-up proteomics experiment



Cell cultures,
Primary cells,
Frozen tissues,
FFPE,
Biofluids,
Exosomes,
Secretome, etc.

Homogenization/
Cell lysis

Protein
precipitation

Denature,
Reduce,
Alkylate.

Proteolytic
digestion

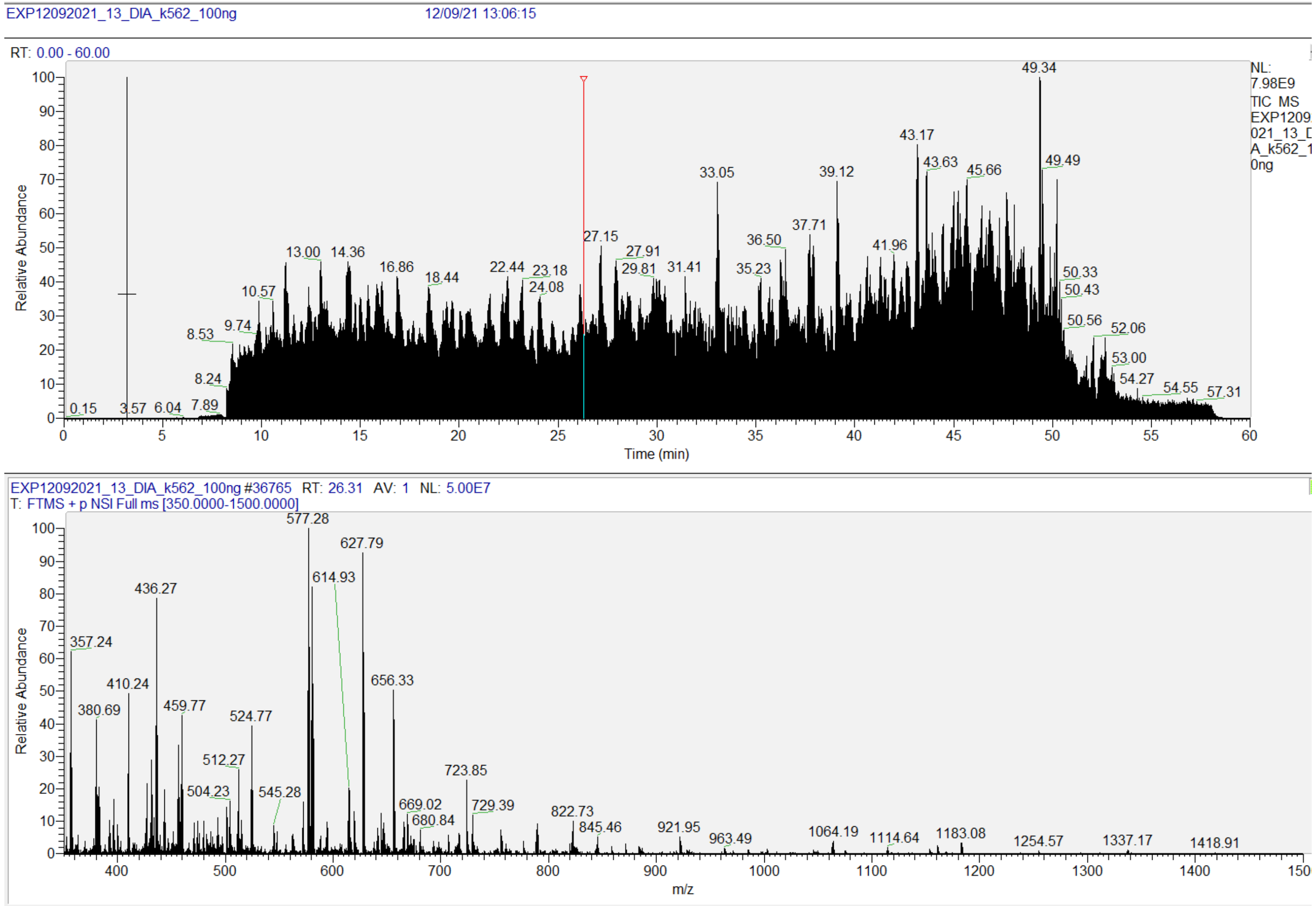
LC-MS/MS

Raw data

Database
search

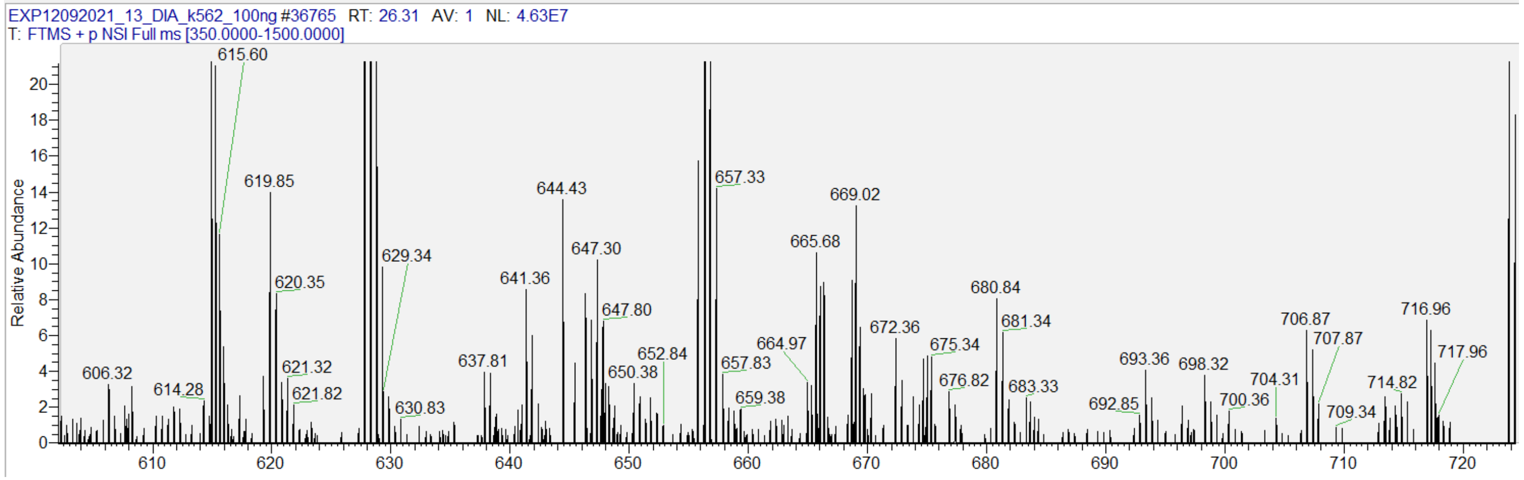
Shotgun Proteomics

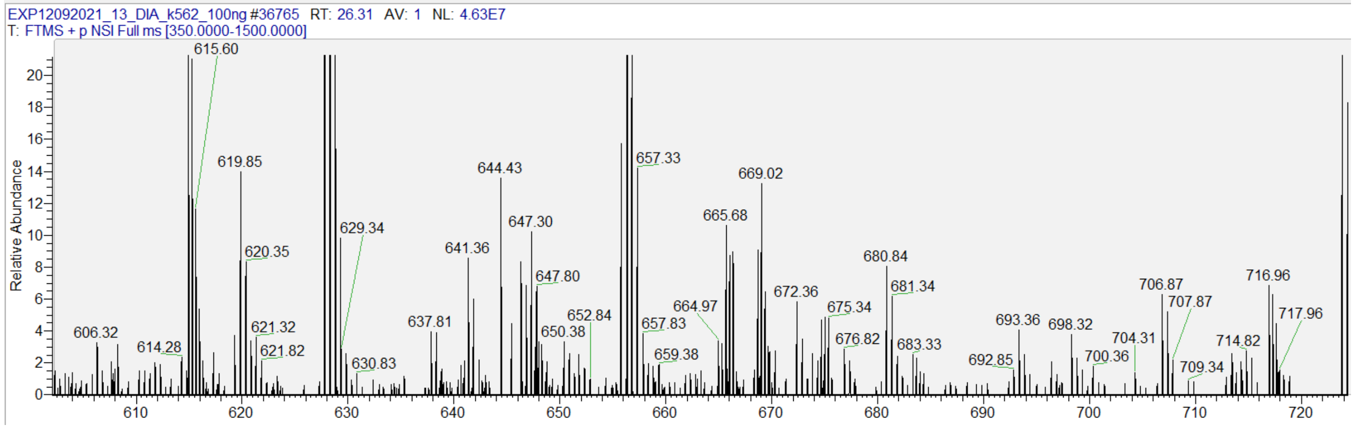
Complex Mixtures (10^3) of proteins are digested using a protease into peptides (10^6)

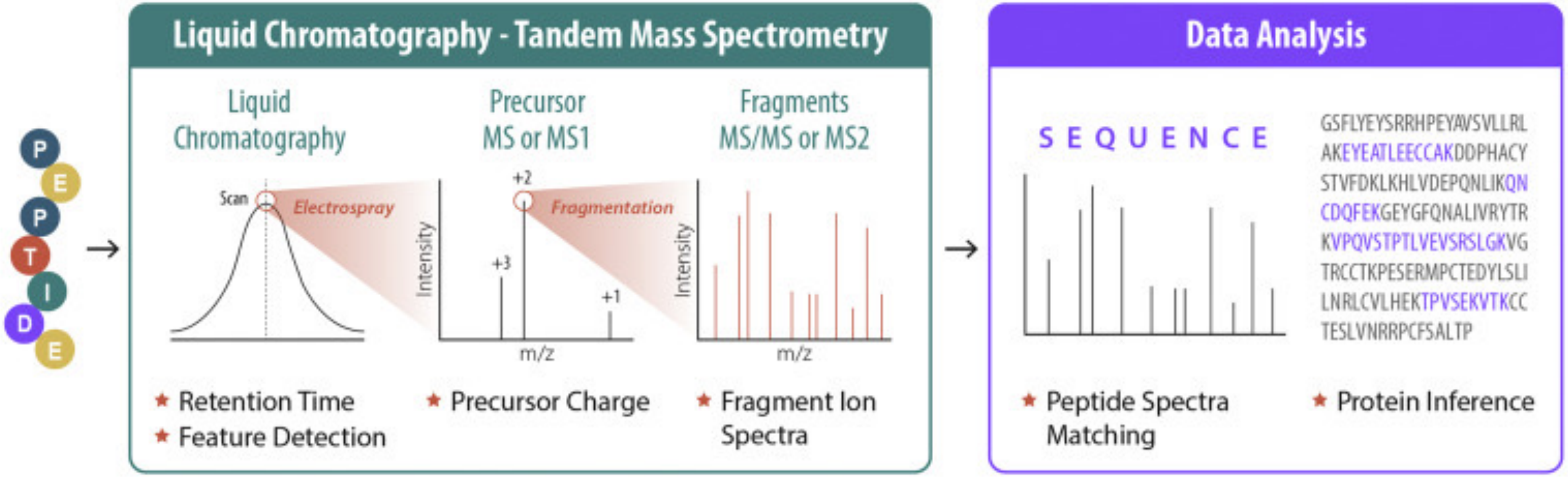


Peptides are Separated and fed into a Mass Spectrometer

Zoomed in. Tons of peptides!!







Liquid Chromatography - Tandem Mass Spectrometry

Liquid Chromatography

Precursor MS or MS1

Fragments MS/MS or MS2

- ★ Retention Time
- ★ Feature Detection
- ★ Precursor Charge
- ★ Fragment Ion Spectra

Data Analysis

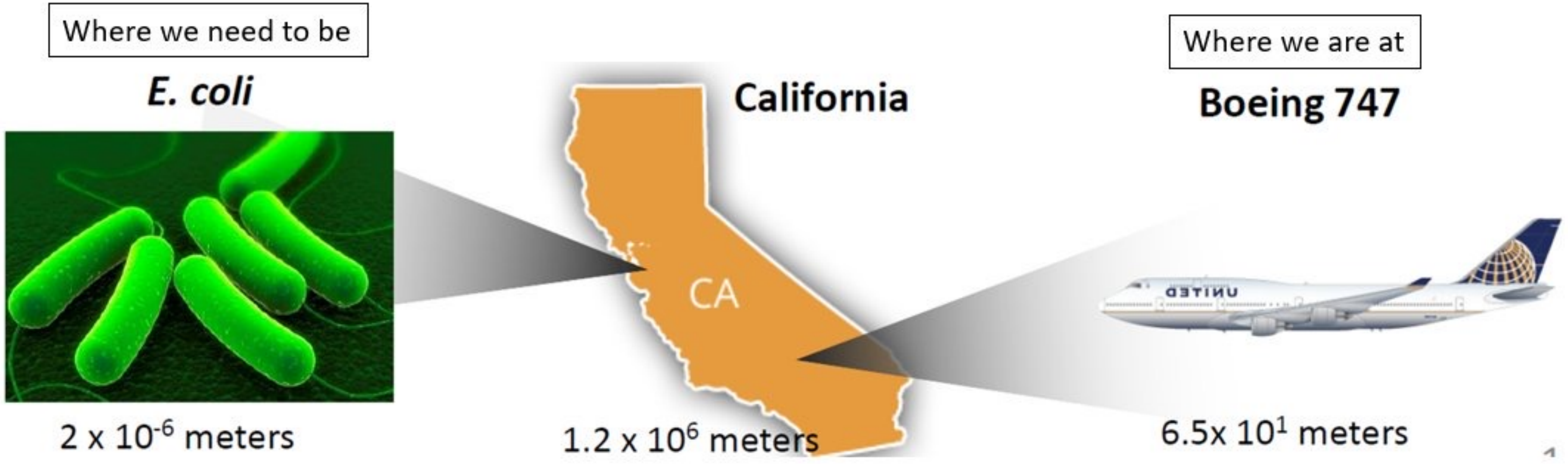
SEQUENCE

GSFLYEYSRRHPEYAVSVLLRL
AKEYEATLEECCAKDDPHACY
STVFDKLKHLVDEPQNLIKQN
CDQFEKGEYGFQNALIVRYTR
KVPQVSTPTLVEVSRSLGKVG
TRCCTKPESERMPCTEDYLSLI
LNRLCVLHEKTPVSEKVTKCC
TESLVNRRPCFSALTP

- ★ Peptide Spectra Matching
- ★ Protein Inference

From : https://www.sciencedirect.com/science/article/pii/S2667237521000035

Courtesy Mike MacCoss

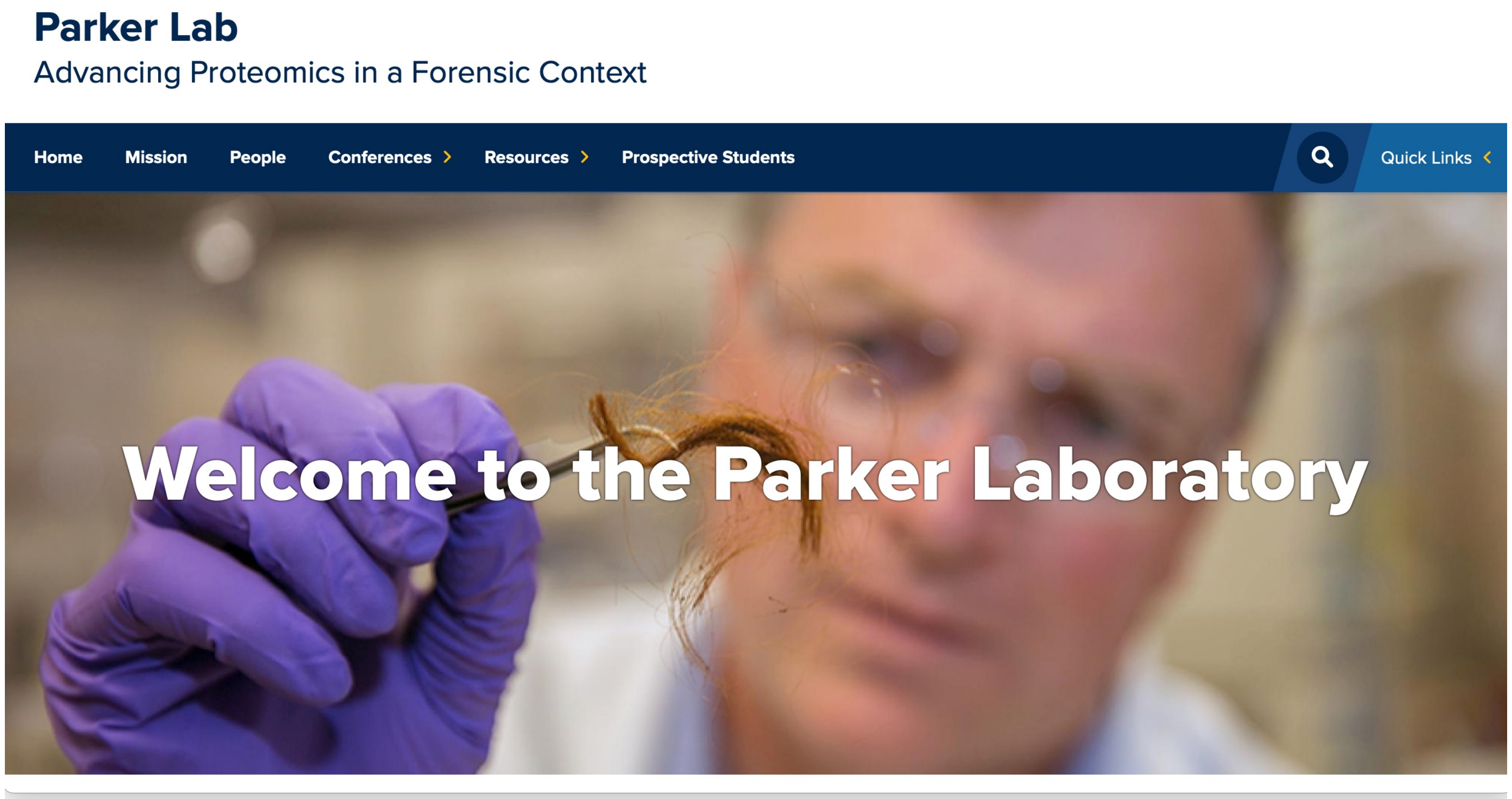


If the most abundant protein represents the length of California, the least abundant proteins represent the length of *E. coli*. Most analytical methods only have 10^4 to 10^5 dynamic range. Representing the size of a 747.

Where we need to be to characterize the transcriptome: 4x10^4 meters, ~marathon

Parker Lab UC Davis

https://parkerlab.ucdavis.edu



Parker Lab

Advancing Proteomics in a Forensic Context

Welcome to the Parker Laboratory

Virtual Presentations

JUNE 18TH AT 1:00PM EDT

SEXING SKELETONS WITH PROTEOMICS WEB

Presented by Dr. Glendon Parker, University of California - Davis

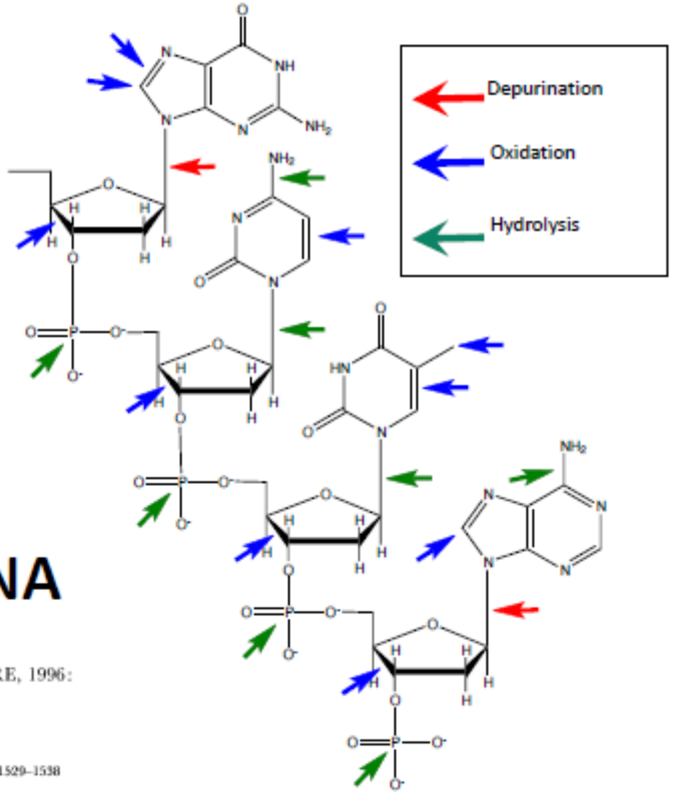
ProteoGenomics

Objectives

- To gain a basic understanding of proteomic genotyping.
- To learn how proteomic information can be useful in a forensic context.
  - Human Hair: Forensic intelligence / human ancestry
  - Human Hair: human identification
  - Fur Hair: Identification of Species Origin
- To learn about applications to Archaeology and Paleontology
  - Proteomic Sex Estimation

Advantages of Protein?

- Protein is intrinsically more stable.



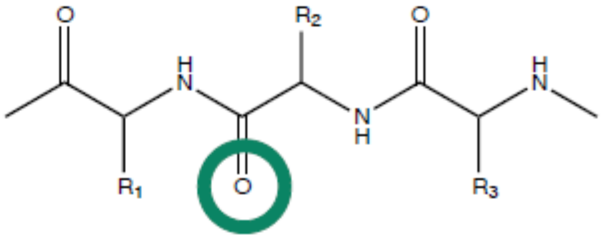
DNA

THE CROONIAN LECTURE, 1996:
Endogenous damage to DNA

T. LINDAHL

Phil. Trans. R. Soc. Lond. B (1996) **351**, 1529–1538

Protein

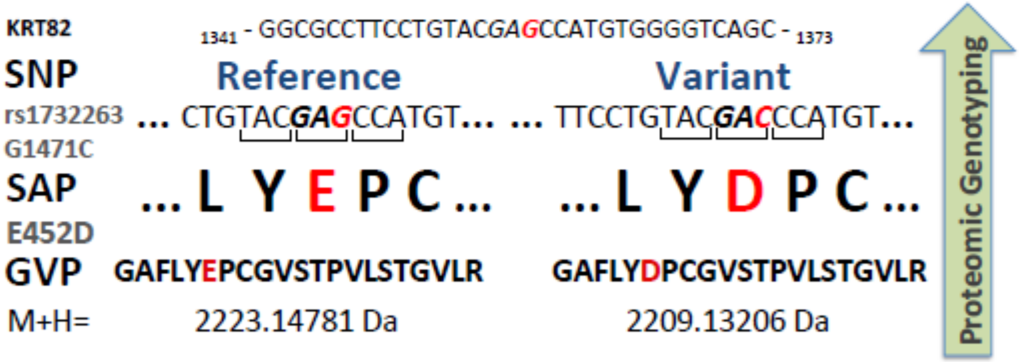


- Less Reactive
- Protein is intrinsically more stable than DNA
  - 1°>2°>3°
- “Pre-Amplified”
- Peptides persist beyond whole proteins

Protein contains genetic information: Proteomic Genotyping

- Genetically Variant Peptides
  - Contains Single Amino Acid Polymorphisms (SAPs)
  - Result of non-synonymous SNPs
  - Detection of GVPs → nsSNP genotype
  - 'Proteomic Genotyping'

  - Allele frequency changes with population (F_{ST})
  - Autosomal: Apply Product Rule
  - Discrimination: GVPs >> mtDNA



KRT82 $_{1341}$ - GGCGCCTTCCTGTAC*GAG*CCATGTGGGGTCAGC - $_{1373}$

	Reference	Variant
SNP rs1732263 G1471C	… CTGTAC***GAG***CCATGT…	… TTCCTGTAC***GAC***CCATGT…
SAP E452D	**… L Y E P C …**	**… L Y D P C …**
GVP	**GAFLYEPCGVSTPVLSTGVLR**	**GAFLYDPCGVSTPVLSTGVLR**
M+H=	2223.14781 Da	2209.13206 Da
af_{EUR}=	0.96	0.04
af_{AFR}=	0.79	0.21

- **Do genotyping when no DNA is present!**
- **Protein Stability >> DNA**
- **Mass Spectrometry sensitive down to 50 ng**

Genomic content in proteome


A global reference for human genetic variation

The 1000 Genomes Project Consortium*

1 OCTOBER 2015 | VOL 526 | NATURE | 71

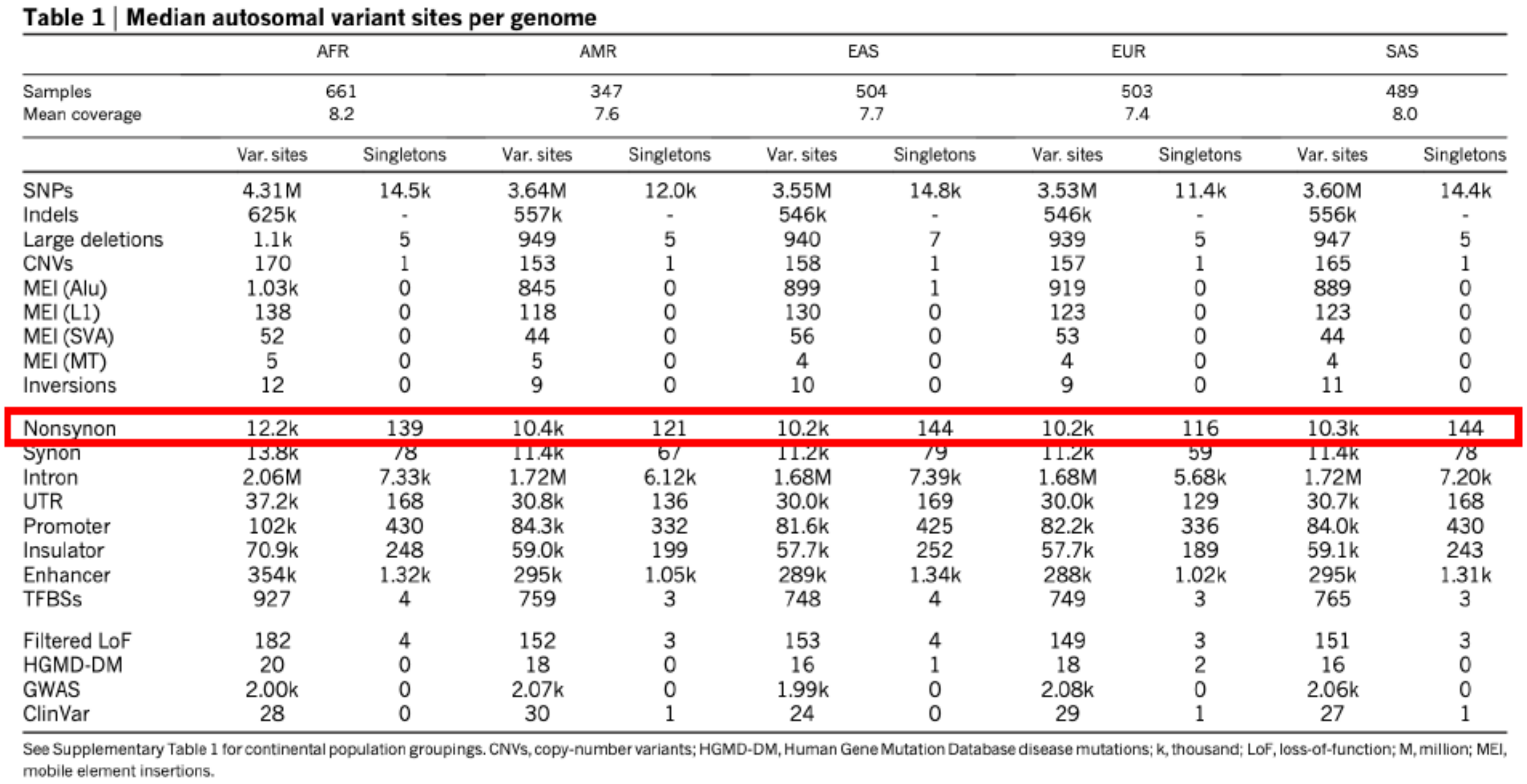


Table 1 | Median autosomal variant sites per genome

	AFR		AMR		EAS		EUR		SAS	
Samples	661		347		504		503		489	
Mean coverage	8.2		7.6		7.7		7.4		8.0	
	Var. sites	Singletons	Var. sites	Singletons	Var. sites	Singletons	Var. sites	Singletons	Var. sites	Singletons
SNPs	4.31M	14.5k	3.64M	12.0k	3.55M	14.8k	3.53M	11.4k	3.60M	14.4k
Indels	625k	-	557k	-	546k	-	546k	-	556k	-
Large deletions	1.1k	5	949	5	940	7	939	5	947	5
CNVs	170	1	153	1	158	1	157	1	165	1
MEI (Alu)	1.03k	0	845	0	899	1	919	0	889	0
MEI (L1)	138	0	118	0	130	0	123	0	123	0
MEI (SVA)	52	0	44	0	56	0	53	0	44	0
MEI (MT)	5	0	5	0	4	0	4	0	4	0
Inversions	12	0	9	0	10	0	9	0	11	0
Nonsynon	12.2k	139	10.4k	121	10.2k	144	10.2k	116	10.3k	144
Synon	13.8k	78	11.4k	67	11.2k	79	11.2k	59	11.4k	78
Intron	2.06M	7.33k	1.72M	6.12k	1.68M	7.39k	1.68M	5.68k	1.72M	7.20k
UTR	37.2k	168	30.8k	136	30.0k	169	30.0k	129	30.7k	168
Promoter	102k	430	84.3k	332	81.6k	425	82.2k	336	84.0k	430
Insulator	70.9k	248	59.0k	199	57.7k	252	57.7k	189	59.1k	243
Enhancer	354k	1.32k	295k	1.05k	289k	1.34k	288k	1.02k	295k	1.31k
TFBSs	927	4	759	3	748	4	749	3	765	3
Filtered LoF	182	4	152	3	153	4	149	3	151	3
HGMD-DM	20	0	18	0	16	1	18	2	16	0
GWAS	2.00k	0	2.07k	0	1.99k	0	2.08k	0	2.06k	0
ClinVar	28	0	30	1	24	0	29	1	27	1

See Supplementary Table 1 for continental population groupings. CNVs, copy-number variants; HGMD-DM, Human Gene Mutation Database disease mutations; k, thousand; LoF, loss-of-function; M, million; MEI, mobile element insertions.

nsSNPs are a small subset of all SNPs

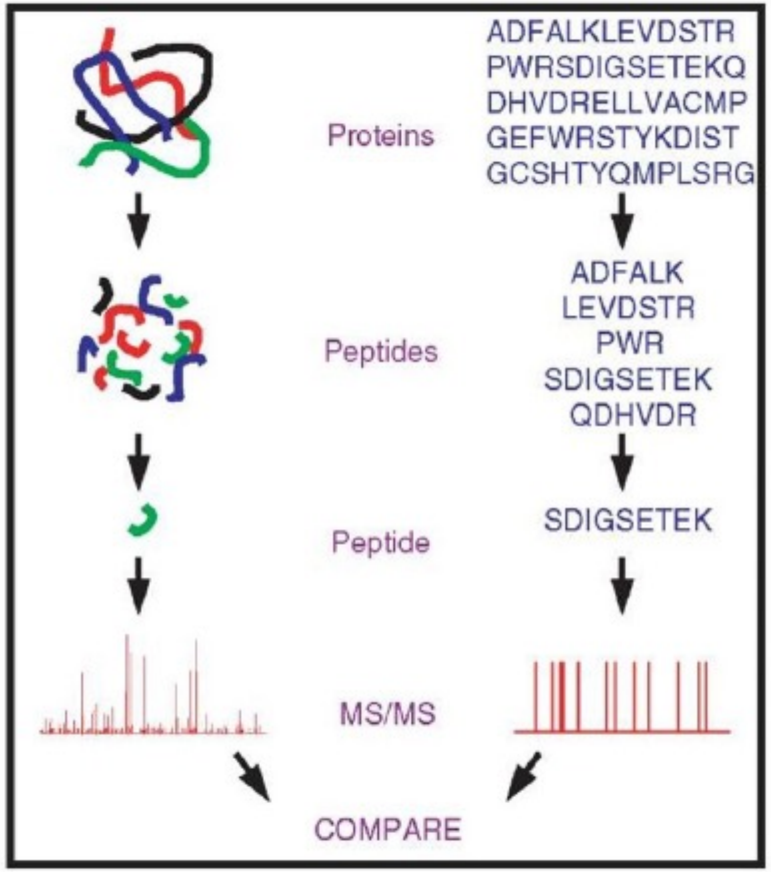
~ 1 nsSNP / 2 proteins

Tandem Mass Spectrometry

- De Novo Sequencing
- Peptide Spectra Matching
  - Match with theoretical sequence
    - Requires reference database
  - Assign a "score"
  - Calculate False Discovery Rate
  - DNA-confirmation/genotyping
- Internal Standards

Proteomics depends on genomics

Proteomics infers the presence of a gene

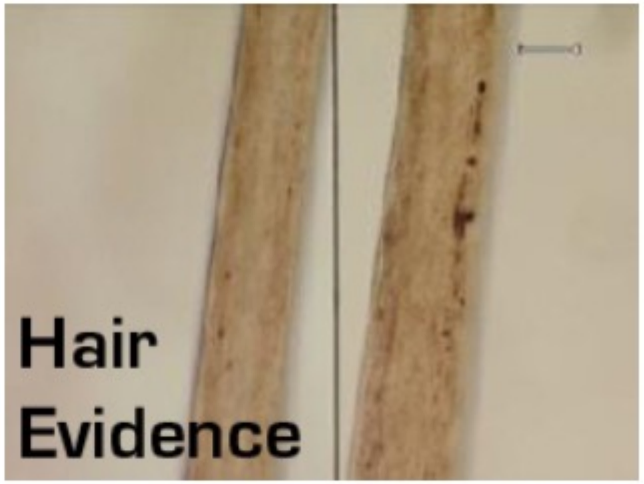


Wholistic Approach to Crime Scene Management

- What do you do when DNA is missing or degraded:
  - Hair shafts
    - DNA degradation: biological
  - Sexual Assault Evidence
    - DNA degradation
    - Multiple contributors
  - Buried remains
    - DNA degradation: environmental
  - Fingermarks
    - Low copy DNA
    - Highly transferred / mixed

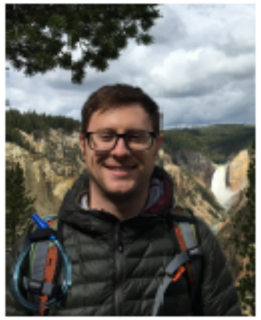
Forensic Use of Hair Shafts

- Hair is ubiquitous
  - 50 to 150 shed per day
- Investigation
  - Ancestry
  - Body Site
  - Identity
- Methods
  - Morphological
  - Analytical
  - Genetic
- **BUT** morphological comparisons are subjective / highly controversial

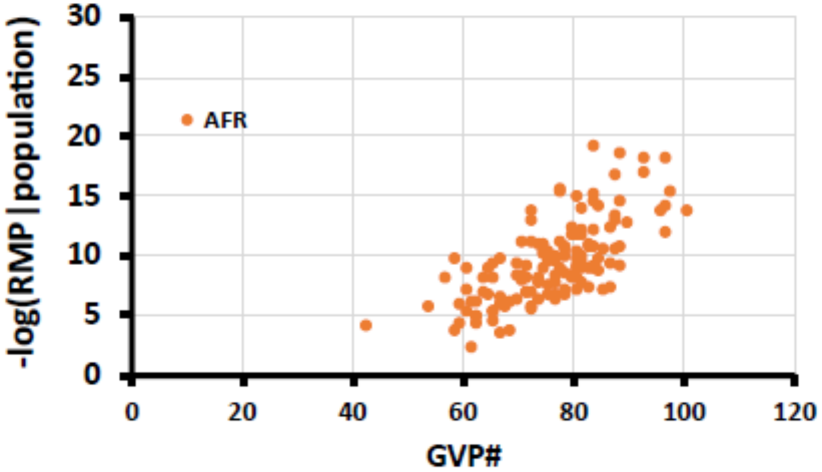


Proteomic Genotyping from Human Hair

- 508 non-synonymous SNP alleles
- Use inferred genotype to develop random match probabilities
- Optimize sample processing
- Optimize mass spectrometry platform
- Sibship and paternity tests
- Several tissues
  - Hair, Bone, Skin, Semen
- Validations
  - Body site
  - Peroxide
  - Greying hair
  - Storage time

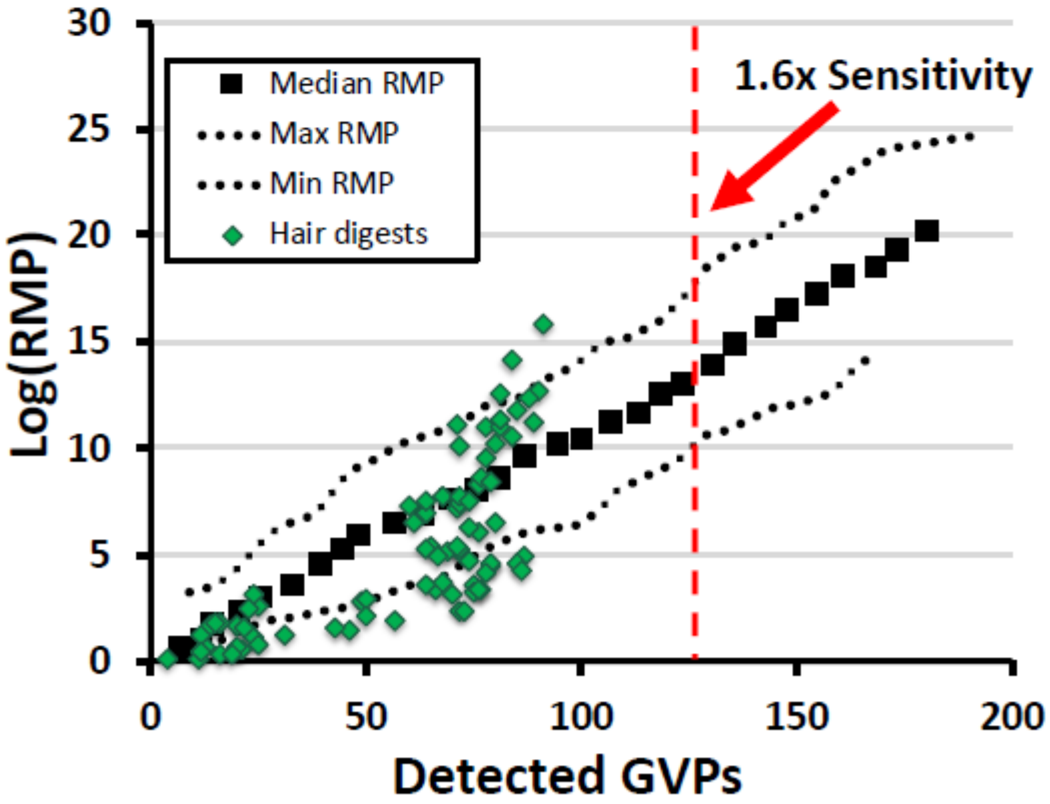


Zachary Goecker



- Hair
  - ~500 proteins, 3 - 4,000 peptides
  - Detect up to 101 GVPs per hair
  - RMPs range up to 1 in 10^{18}

Side Bar: How many GVPs do you need?

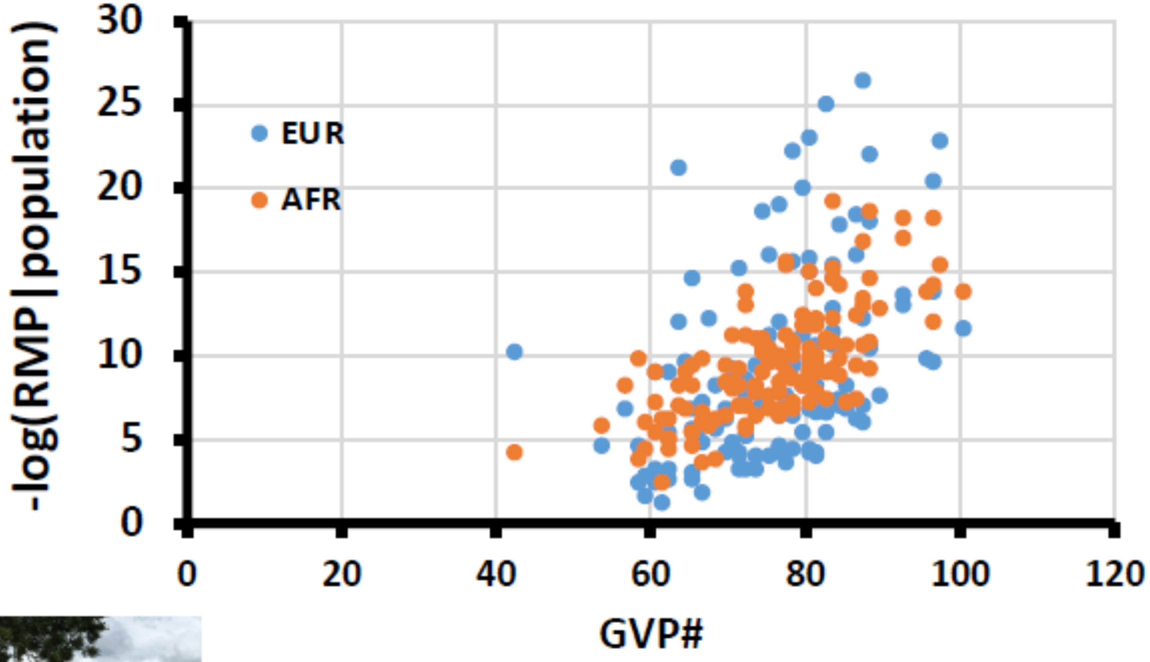


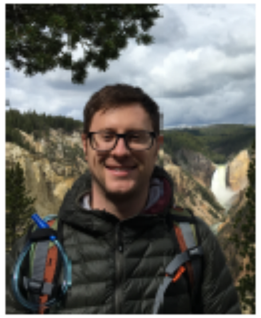
- Current
  - Average GVPs = 77
  - Median RMP = 1 in 1.1 million
- Projected with heavy-isotope peptides
  - Average GVPs = 123
  - Median RMP = 1 in 10 trillion

~10 GVPs per order of magnitude

What can we do with this information?

- Optimizing method for forensic lead generation (ancestry) from a single hair
  - ~500 proteins, 3 - 4,000 peptides
  - Detect up to 101 GVPs per hair
  - RMPs range up to 1 in 10^{18}
  - RMPs change with reference population (1000G)

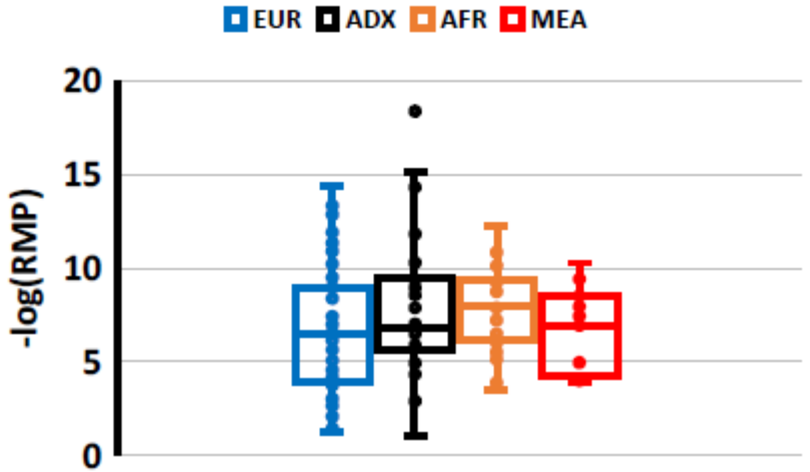




Zachary Goecker

Ancestry Estimation from Hair

- 132 subjects
  - (>75% DNA sequence, >2000 peptides)
  - ADMIXTURE analysis (% African Ancestry)
- Four populations
  - European to African axis
- Q-Exactive Plus MS
- X!Tandem & GVPFinder:
  - nsSNP profile
- 3 Approaches
  - STRUCTURE
  - PLSDA
  - Likelihood Ratios



	European	**Admixed**	**African**	**Middle Eastern**	**All**
n =	71	26	24	11	132
$-\log(\mathrm{RMP})_{max}$	14.3	18.4	12.2	10.3	18.4
$-\log(\mathrm{RMP})_{median}$	6.5	8.7	8.0	6.9	8.8
#GVPs	57 - 101	54 - 98	43 - 88	71 - 90	43 - 101

Summary

- Proteomics
  - 43-101 GVPs per hair shaft
  - RMPs up to 1 in 10^{18}
- STRUCTURE
  - Not work for proteomic genotype data
- PCA / PLS-DA
  - Single populations (AFR= 13/21; EUR= 54/65)
  - Mixed ancestry (ADX= 7/20)
- Likelihood Ratio
  - 26 orders of magnitude
  - Partitions genetic populations
  - Accuracy = 11 ± 13%
    - (67% of samples = 6.8 ± 8.3%)
  - Reference population = 1000G

  - Poorer performance with log(LR) values between -2.5 to 2.5.

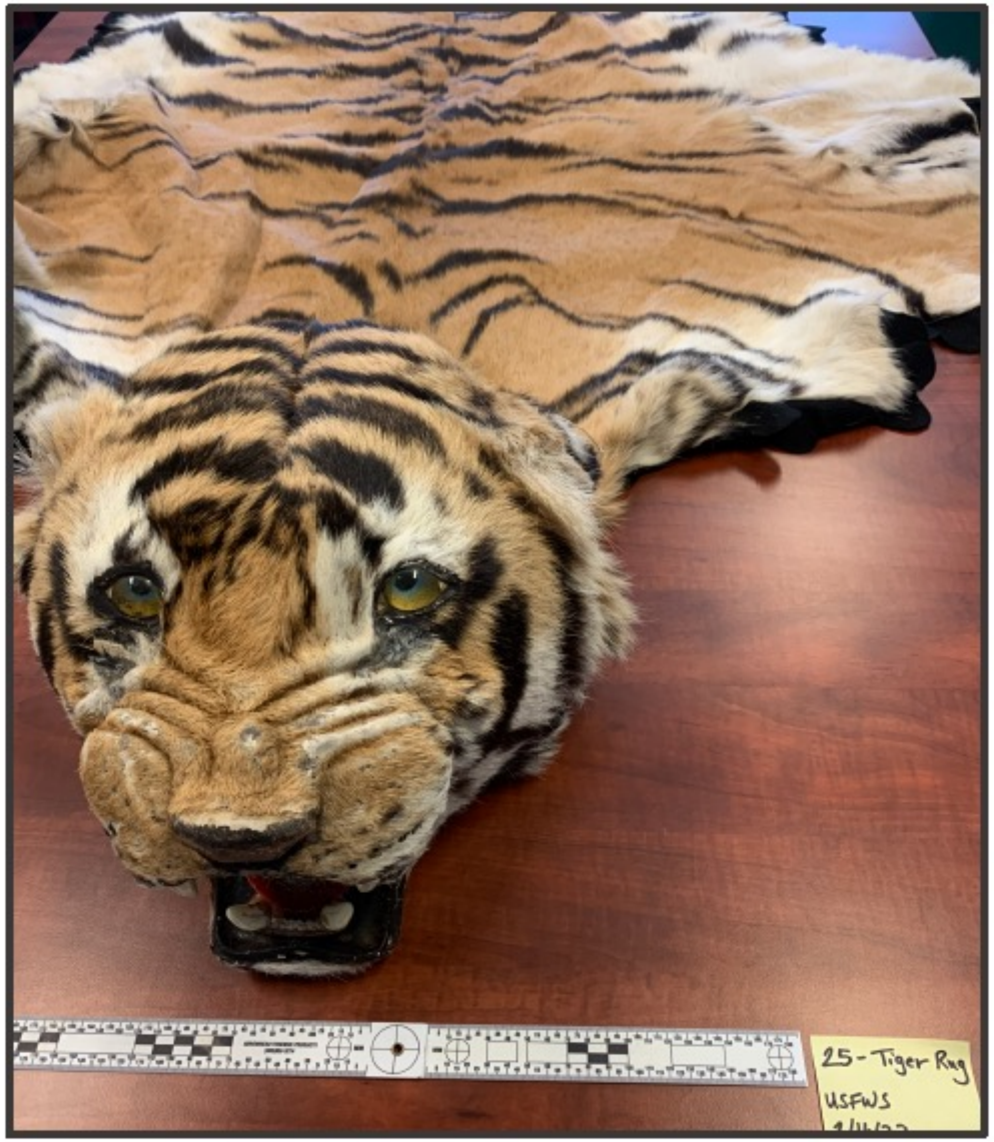
Conclusion

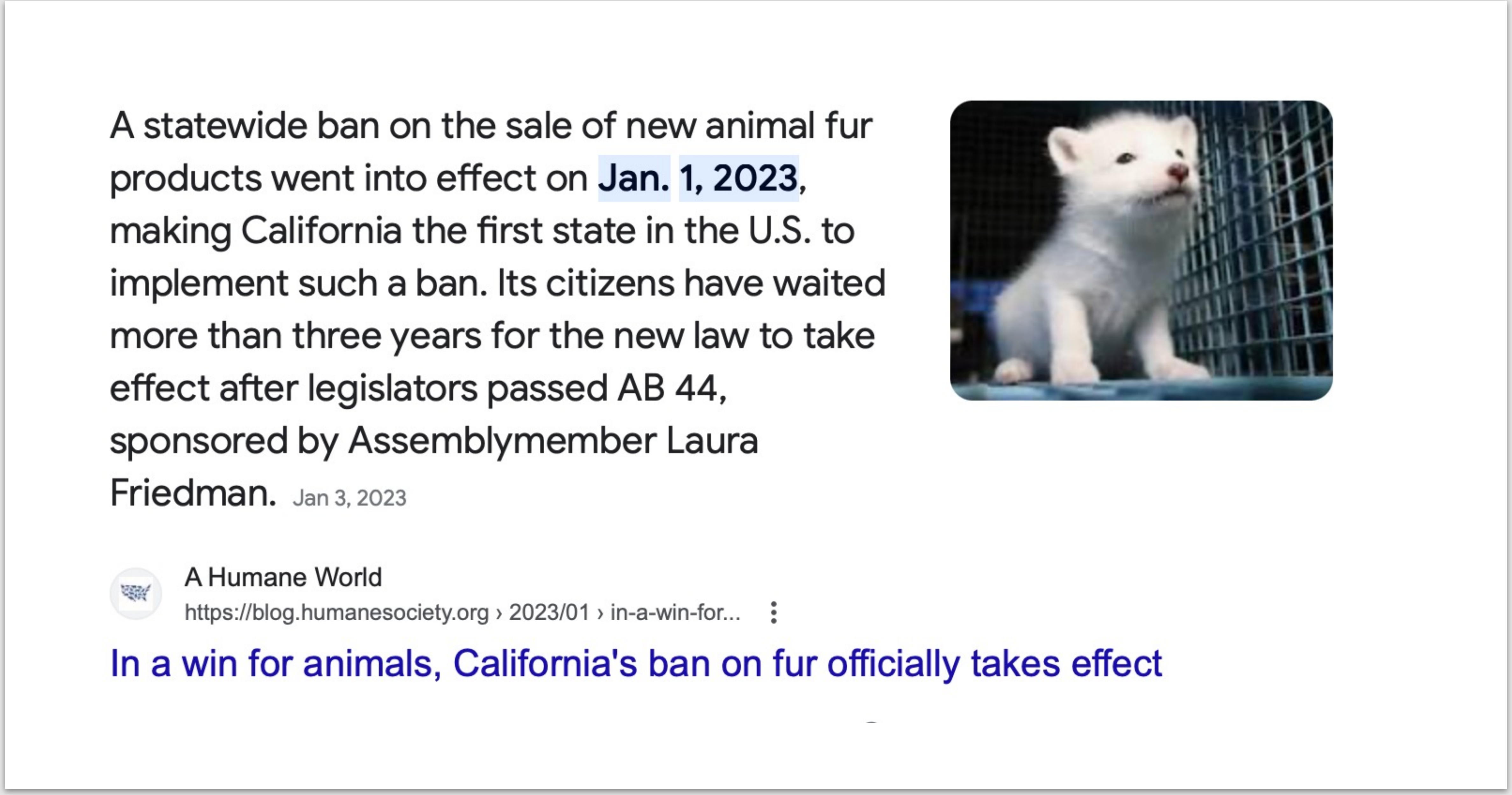
- Investigative leads
  - Actionable information
  - Data from sample alone
  - Minimize bias
    - Subjective analysis
    - Non-systematic reference population
  - Scientifically based
  - Statistically sound
  - Complement mtDNA

- Ancestral Likelihood Ratio
  - Accuracy = 11 ± 13%
    - (66% of samples = 6.8 ± 8.3%)
    - Uneven: $-2.5 < \log LR < 2.5$
  - Sample population = 1000G
- Human Identification
  - Median RMP = 1 in $10^{8.8}$

Increasing the scope of genetic proteomics

- Other forensic tissues
  - Sexual Assault Evidence
  - Degraded Skeletons
  - Fingermarks
- Wildlife Forensics
  - Identify Species of Origin
- Proteomic Sex Estimation

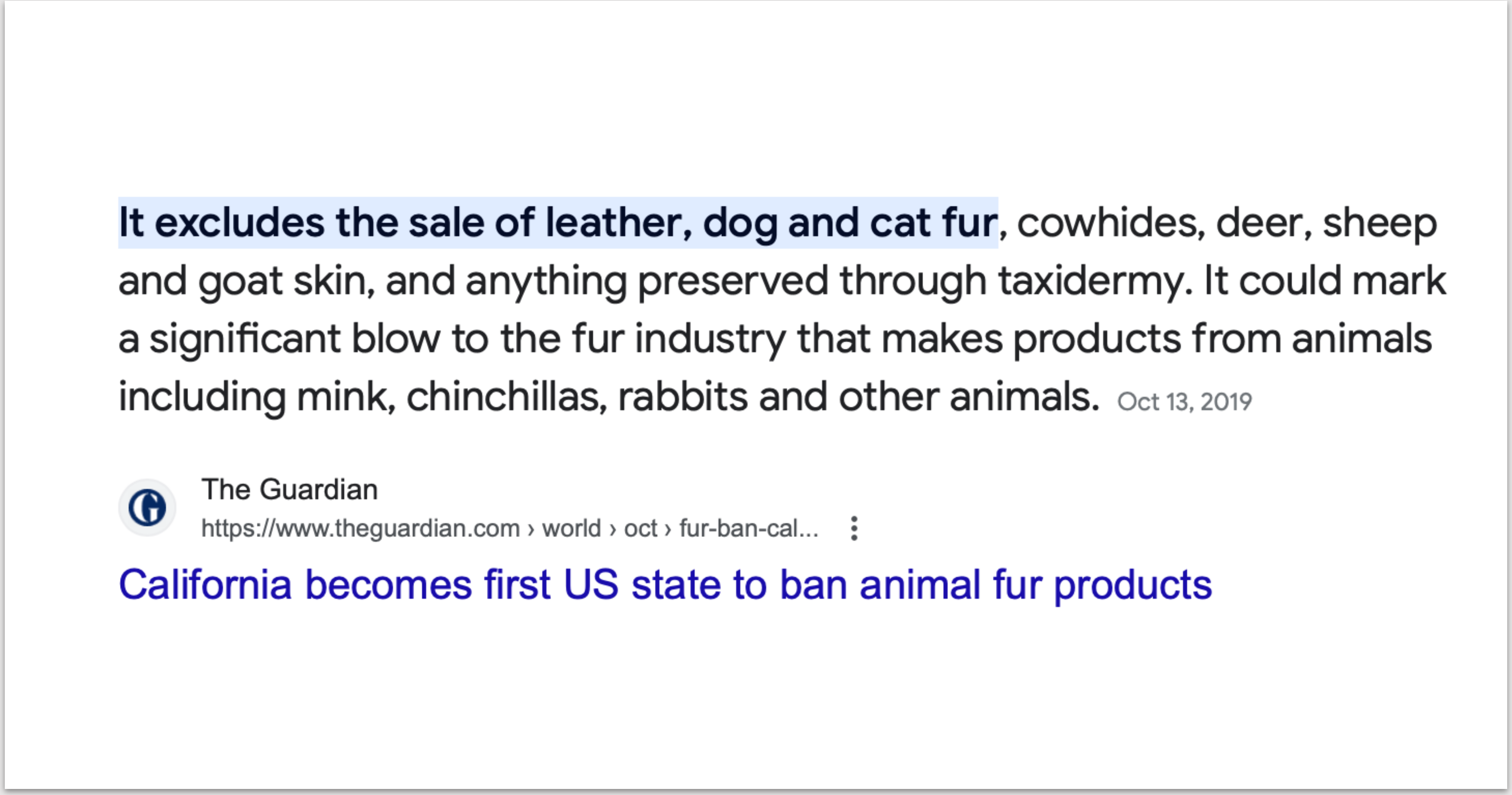




A statewide ban on the sale of new animal fur products went into effect on **Jan. 1, 2023**, making California the first state in the U.S. to implement such a ban. Its citizens have waited more than three years for the new law to take effect after legislators passed AB 44, sponsored by Assemblymember Laura Friedman. Jan 3, 2023

A Humane World
https://blog.humanesociety.org › 2023/01 › in-a-win-for...

In a win for animals, California's ban on fur officially takes effect



It excludes the sale of leather, dog and cat fur, cowhides, deer, sheep and goat skin, and anything preserved through taxidermy. It could mark a significant blow to the fur industry that makes products from animals including mink, chinchillas, rabbits and other animals. Oct 13, 2019

The Guardian
https://www.theguardian.com › world › oct › fur-ban-cal...

California becomes first US state to ban animal fur products

Hybrid cat

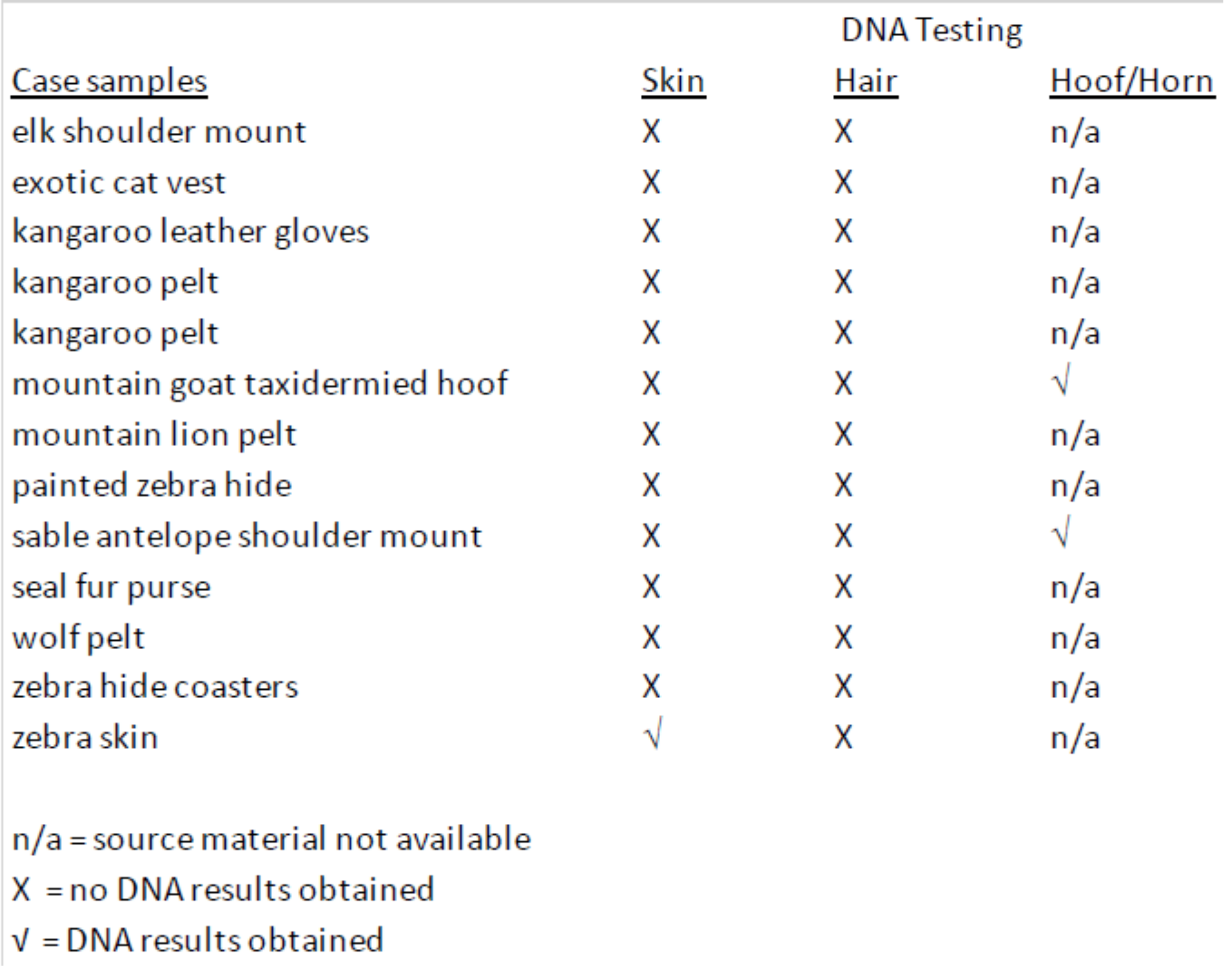
(B) “Fur product” does not include any of the following:

(i) **A dog or cat fur product**, as defined in Section 1308 of Title 19 of the United States Code, as that section read on January 1, 2020.

(ii) An animal skin or part thereof that is to be converted into leather, which in processing will have the hair, fleece, or fur fiber completely removed.

Statement of Problem: Phylogenetic Proteomics

- Fur is a major challenge in wildlife forensics.
- Easier to traffic.
- Manufacture requires
  - High temperatures
  - Low pH
  - ± formaldehyde
  - ± peroxide, bleach
- High Failure rate for DNA bar coding! (<3%)



	DNA Testing		
Case samples	Skin	Hair	Hoof/Horn
elk shoulder mount	X	X	n/a
exotic cat vest	X	X	n/a
kangaroo leather gloves	X	X	n/a
kangaroo pelt	X	X	n/a
kangaroo pelt	X	X	n/a
mountain goat taxidermied hoof	X	X	√
mountain lion pelt	X	X	n/a
painted zebra hide	X	X	n/a
sable antelope shoulder mount	X	X	√
seal fur purse	X	X	n/a
wolf pelt	X	X	n/a
zebra hide coasters	X	X	n/a
zebra skin	√	X	n/a

n/a = source material not available
X = no DNA results obtained
√ = DNA results obtained

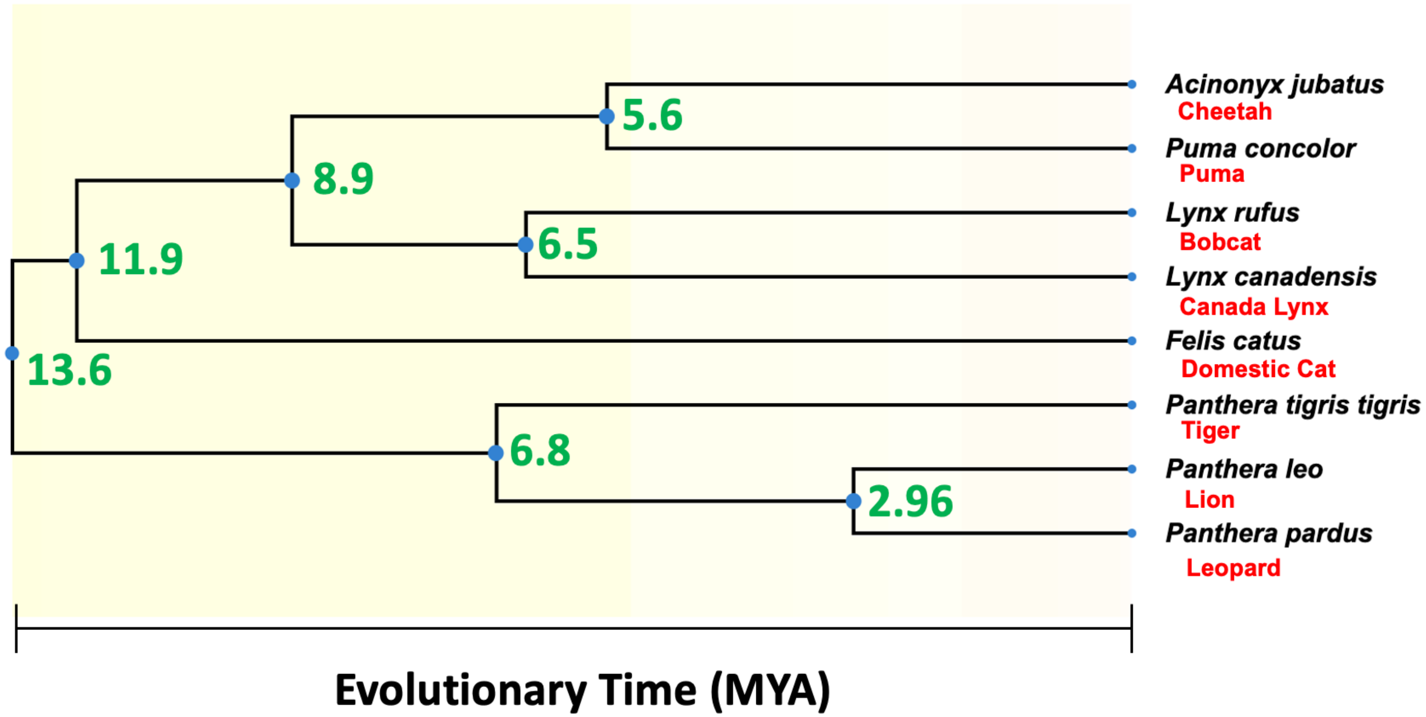
Proteomics on Non-Model Organisms

- Theory: Peptide Spectra Matching Efficiency
  - Efficiency down with evolutionary distance
  - nsSNPs
    - Individual
    - Common Minor Allele
    - Major Allele
    - Population Specific
    - Species-Specific

- Practice: Potential confounding effects
  - Some species closely related
    - Lion and leopard ~3-4 million yrs
  - Some proteins do not change much with evolution.
  - Protein modifications confused for species specific amino acids
  - Not all proteomes are equal

Sidebar: Felidae Phylogenetics

- Forensic Challenge
  - Related species
    - Eg. Lion and Leopard
  - How good can you resolve a population with proteomics?

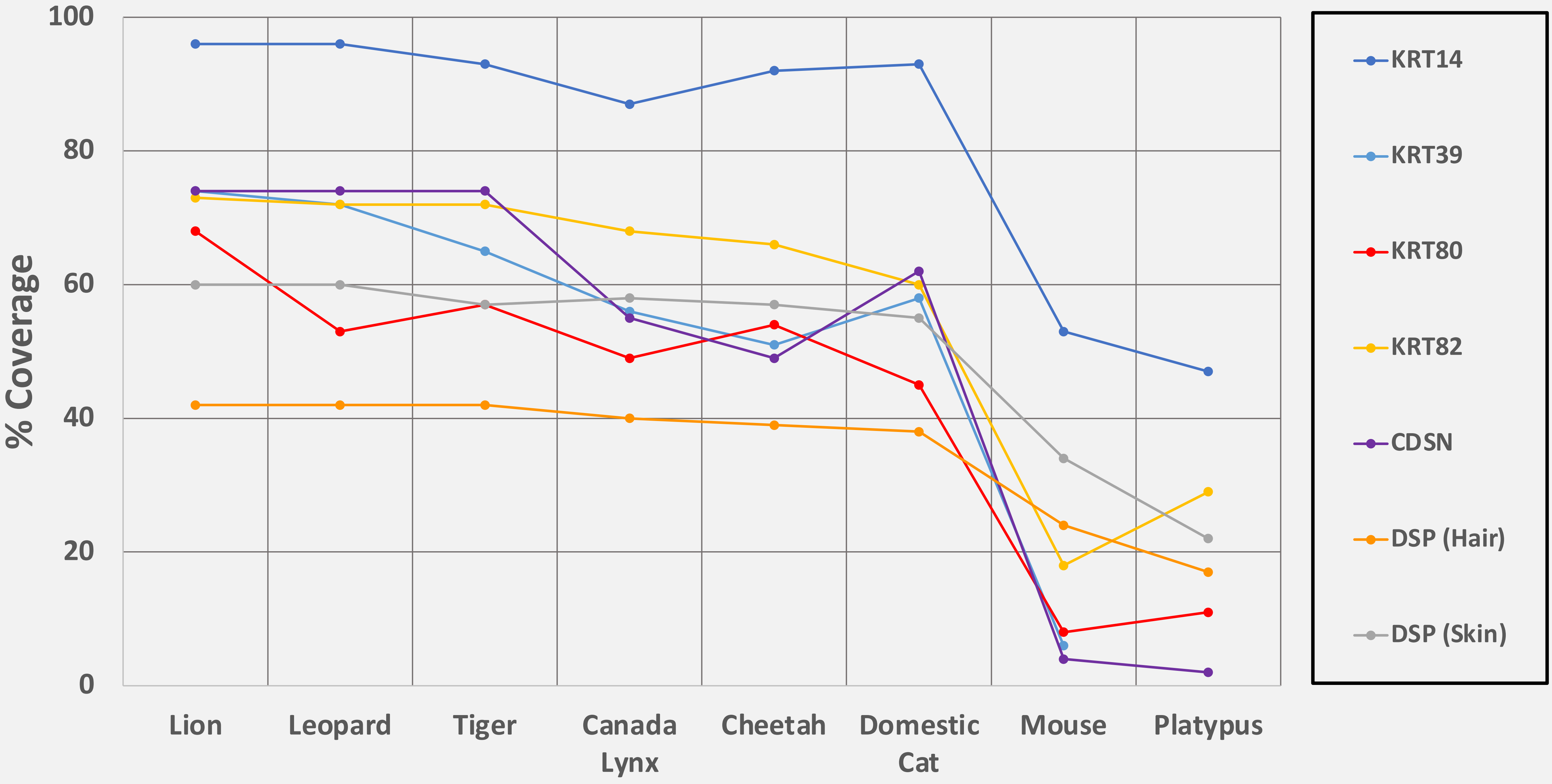


Identify Species Origin of Felidae Fur

- 5 fur Species Samples
  - Panthera leo (tiger)
  - Panthera pardus (leopard)
  - Panthera tigris (tiger)
  - Puma concolor (puma)
  - Acinonyx jubatus (cheetah)
- 6 samples
  - 3 skin
  - 3 fur hair

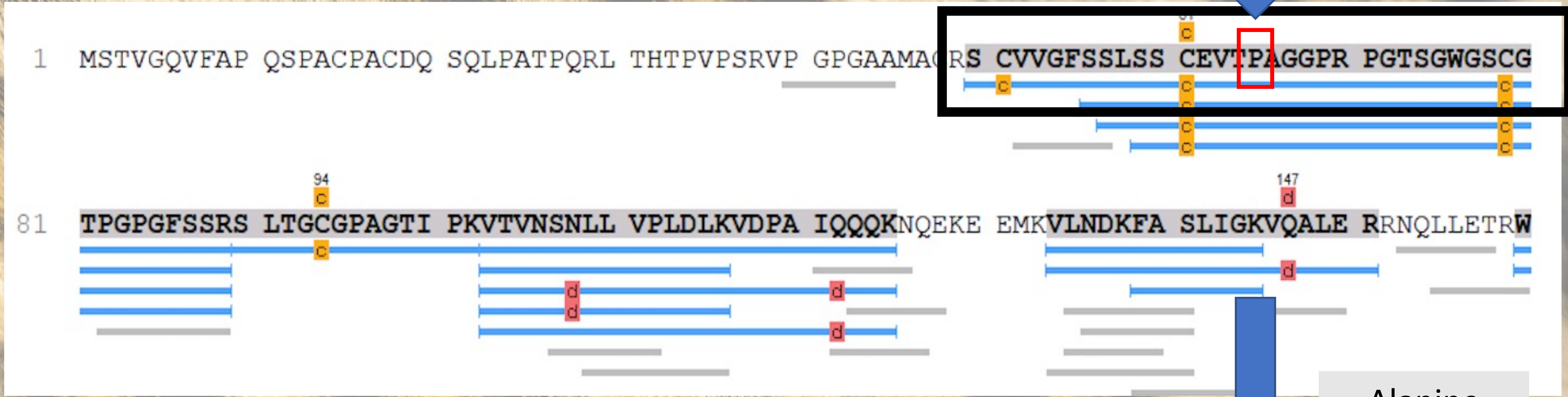
1. Process using an optimized hair protocol.
2. Proteomic mass spectrometry
3. Search raw data with species-specific reference protein databases
4. Measure PSM

% Coverage of Proteins in Lion Paw



Example of Protein Coverage

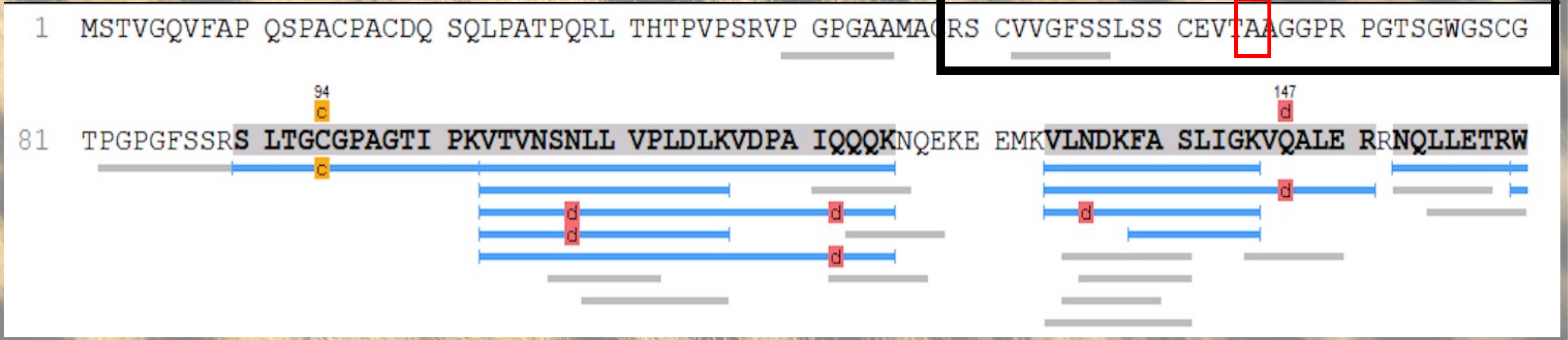
Proline



1 MSTVGQVFAP QSPACPACDQ SQLPATPQRL THTPVPSRVP GPGAAMACRS CVVGFSSLSS CEVTPAGGPR PGTSGWGSCG

81 TPGPGFSSRS LTGCGPAGTI PKVTVNSNLL VPLDLKVDPA IQQQKNQEKE EMKVLNDKFA SLIGKVQALE RRNQLLETRW

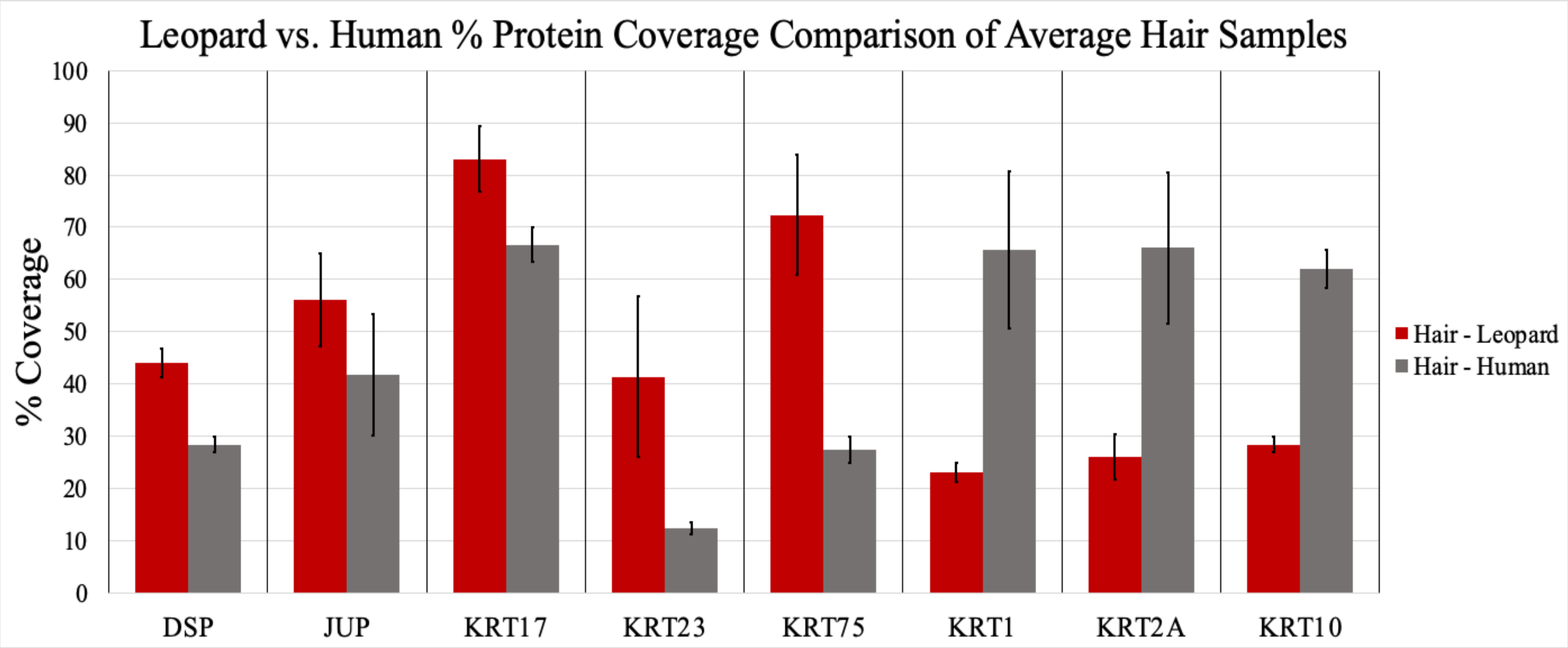
Alanine



1 MSTVGQVFAP QSPACPACDQ SQLPATPQRL THTPVPSRVP GPGAAMACRS CVVGFSSLSS CEVTAAGGPR PGTSGWGSCG

81 TPGPGFSSRS LTGCGPAGTI PKVTVNSNLL VPLDLKVDPA IQQQKNQEKE EMKVLNDKFA SLIGKVQALE RRNQLLETRW

Identify Species Origin of Felidae Fur



Other Factors?

- More stringent searches = no effects
- Protein coverage vs total peptides = no effect
- Human contamination = has an effect

Summary: Phylogenetic Proteomics

- Resolve Lion and Leopard (just)
- Changes can be more pronounced at the protein level
- Species specific peptides can be identified and detected!

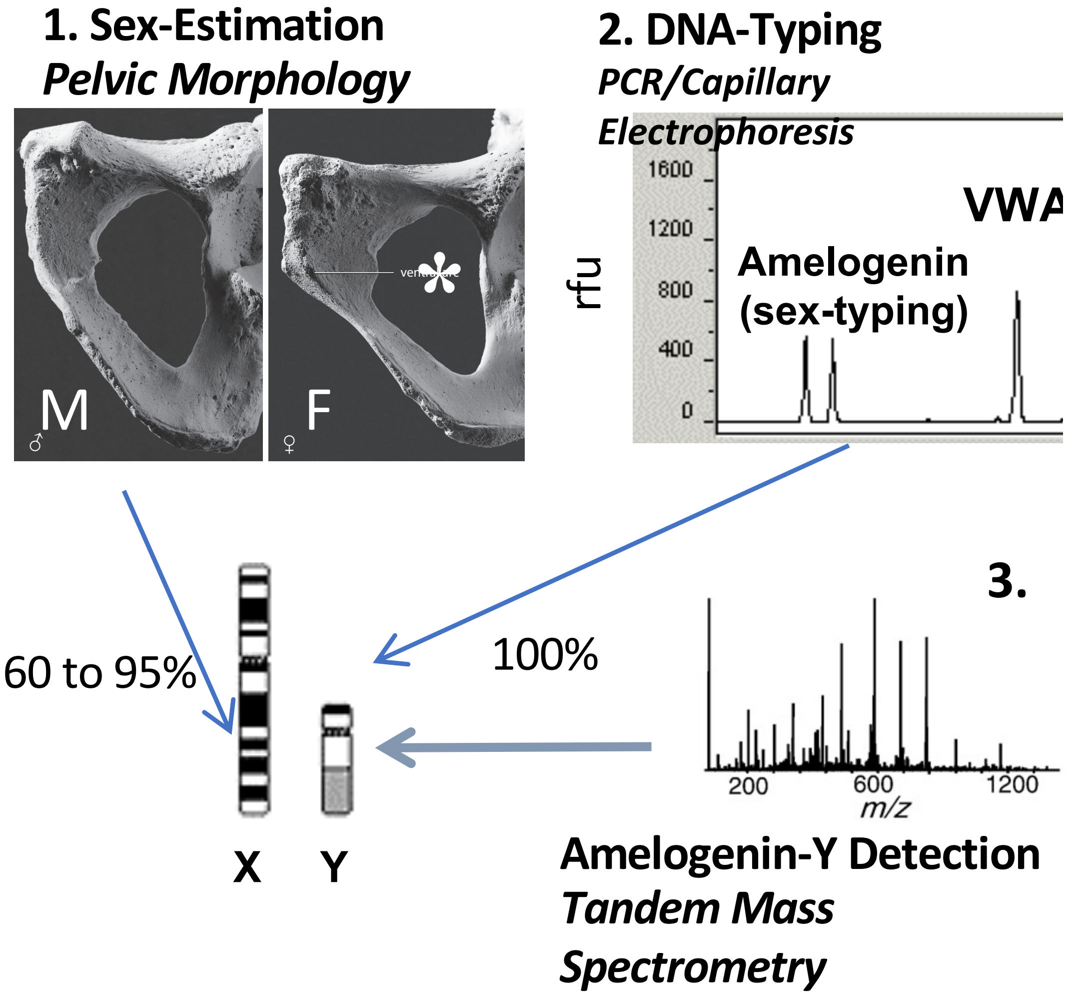
- Peptide Sequence Matching is reliable for faster analysis, with no need for development of targeted assays.

- Future work
  - Targeted assays
  - Species specific QQQ assay.

  - More unknown samples!

Statement of Problem: Proteomic Sex Estimation

- Skeletons mostly do not have sexually dimorphic markers
  - Non-adults: male/female skeletons alike
  - Degraded: pelvis bone is fragile, other markers ambiguous.
  - Together most skeletons cannot be sexed.
- DNA:
  - X-/Y-chromosome Markers
  - Sensitive, but DNA is often missing
  - Low copy number → error rate



1. Sex-Estimation
Pelvic Morphology

2. DNA-Typing
PCR/Capillary Electrophoresis

Amelogenin (sex-typing)

3.

Amelogenin-Y Detection
Tandem Mass Spectrometry

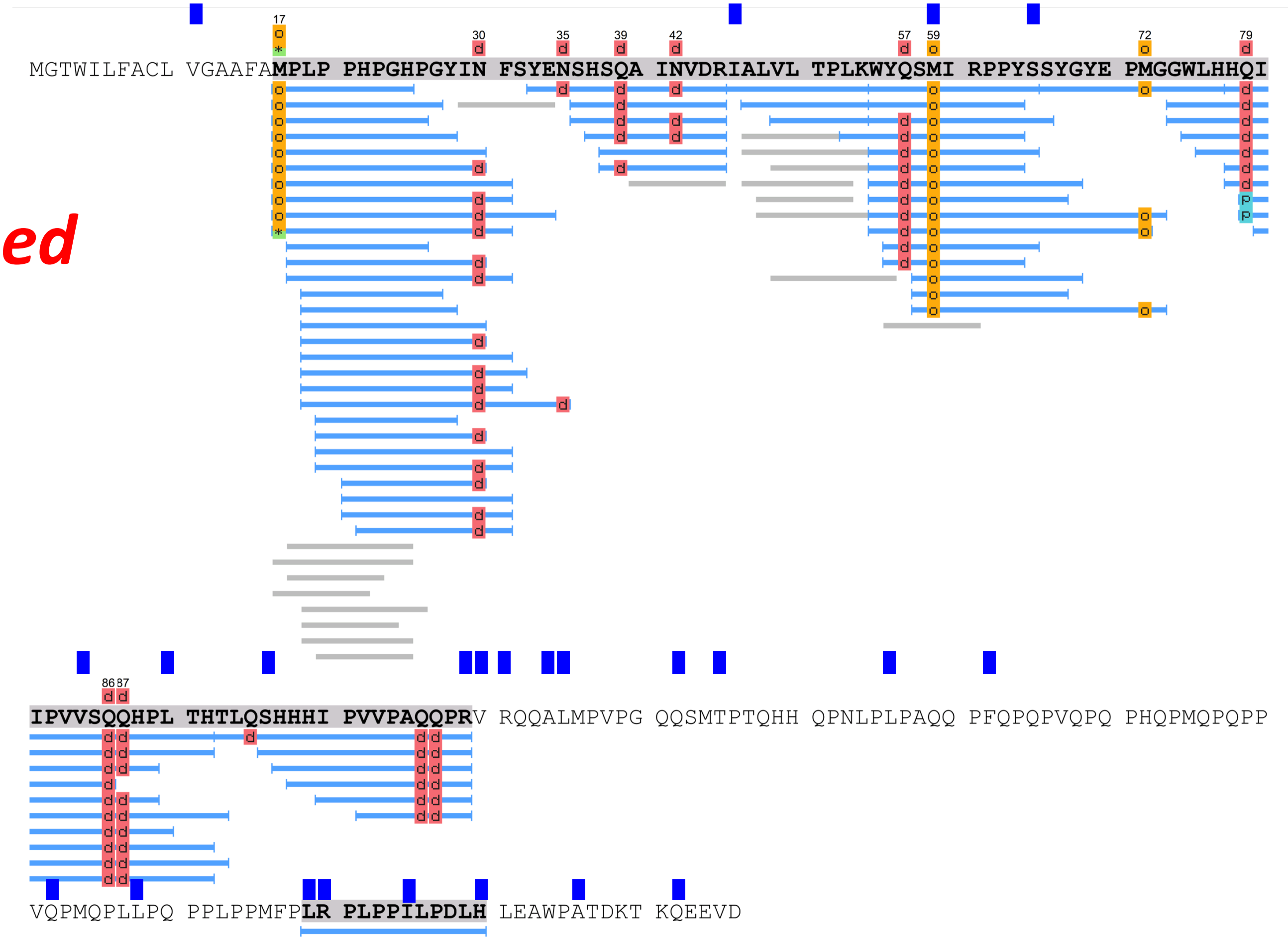
60 to 95%

100%

Amelogenin Genes are Expressed in Enamel

The most characterized sex-chromosome markers are expressed in the most robust tissue!

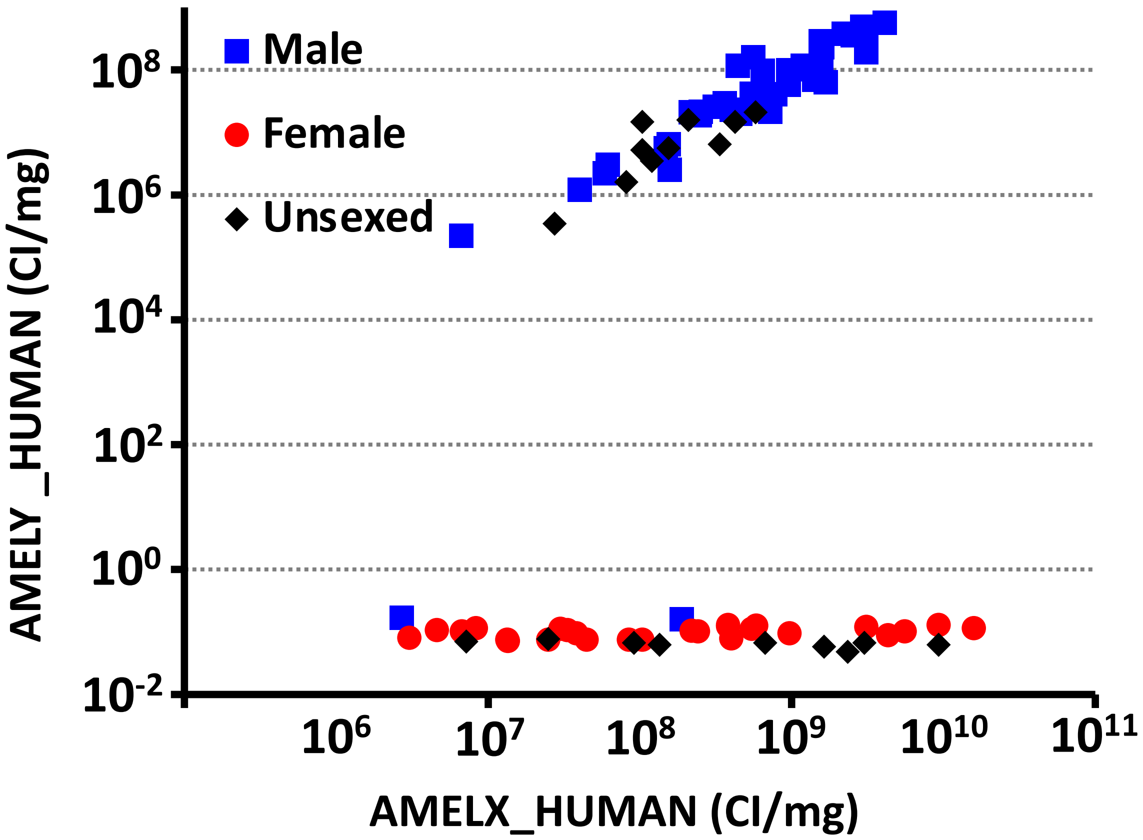
- Amino acid differences occur between them
  - AMELX_HUMAN
  - AMELY_HUMAN
- Amelogenin peptides can be extracted from enamel tissue



- AMELY_HUMAN
- CA-ALA-554 B85D, ~1000 BP

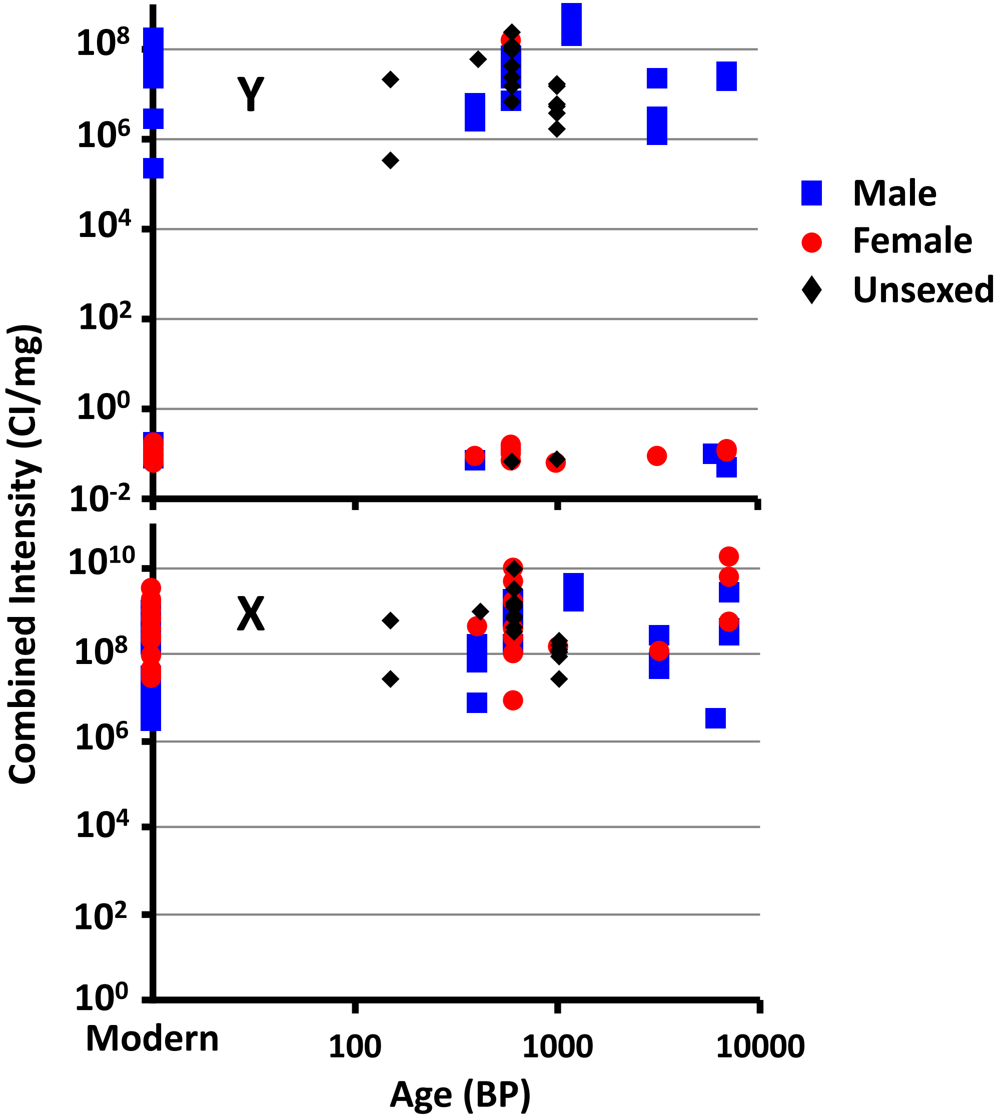
Amelogenin Genes are Expressed in Enamel

- Peptides can be measured
  - Total Ion Current (TIC)
- Combine all peptide signals that are specific to AMELX_HUMAN or AMELY_HUMAN
- Male (■) and Female (●) teeth separate into two populations.
  - Signal ranges over 2 orders of magnitude
  - AMELY is about 10% of AMELX
  - 2 outliers
  - Unsexed samples partition as well



Amelogenin Genes are Expressed in Enamel

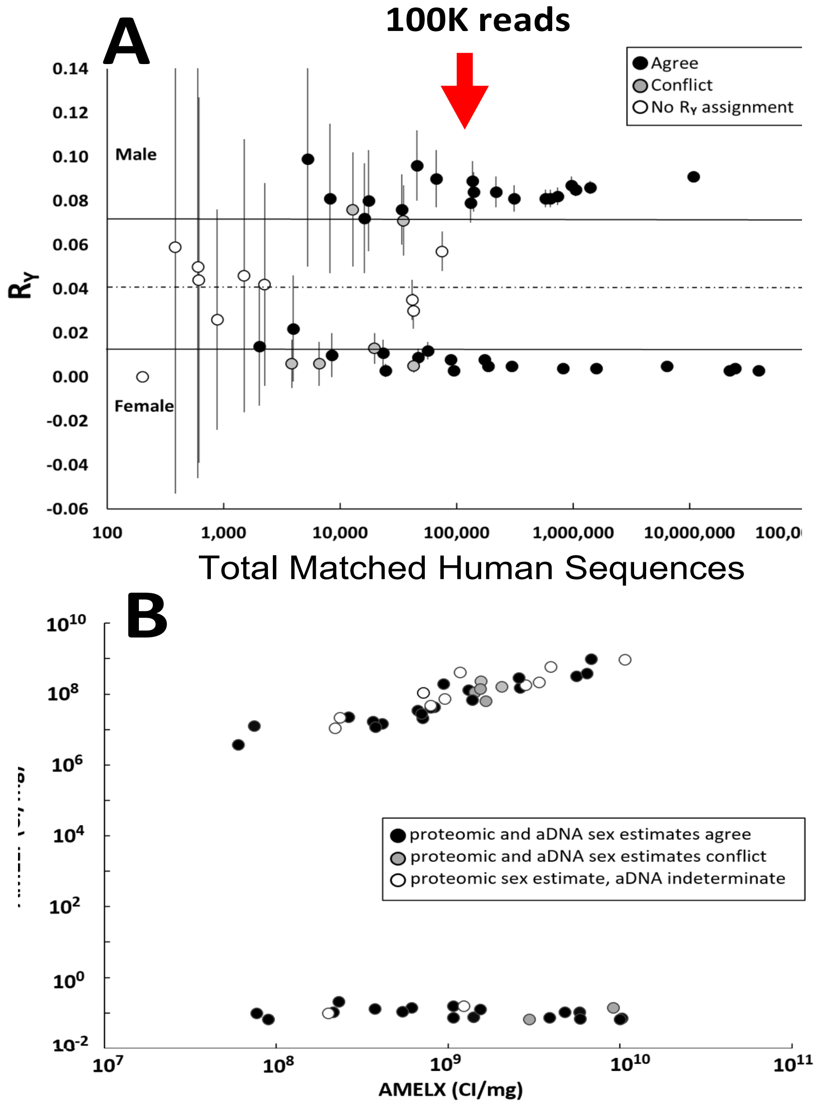
- Peptides are very stable
- Over 10,000 years no change in signal!
- Male (■) and Female (●) teeth separate into two populations.
  - AMELY is about 10% of AMELX
  - 2 outliers
  - Unsexed samples partition as well



Amelogenin Genes are Expressed in Enamel

- How does Proteomic Sex Estimation compare to other methods?
- Genomic Sex Estimation contradicts some Proteomic Sex Estimates
    - Who is at fault?: Protein, DNA or Both
    - Contradictions only occur in low quality DNA samples!
    - no pattern in protein quality!
- Therefore when estimating sex
    - Proteomic sex estimation > High read (>100K) genomic estimation > Osteology sex estimation.

Genomic Sex Estimation is NOT reliable < 100K reads



Summary: Proteomic Sex Estimation

- Pros:
  - Partitions male and female samples
  - Highly sensitive and stable
    - No change over 10K years
  - More reliable than DNA methods, where DNA read < 100K.
  - Potentially much cheaper!

- Cons:
  - DNA genotyping is being conducted anyway.
  - If >100K ancient reads, no difference.

  - Osteological sex estimation
    - Only good for ~50% of samples
    - BUT very cheap and fast
    - confident estimates are reliable (>95%).

Comparative Study > Sci Rep. 2020 Jul 17;10(1):11897. doi: 10.1038/s41598-020-68550-w.

A comparison of proteomic, genomic, and osteological methods of archaeological sex estimation

Tammy Buonasera [1] [2], Jelmer Eerkens [3], Alida de Flamingh [4], Laurel Engbring [5], Julia Yip [6], Hongjie Li [7], Randall Haas [3], Diane DiGiuseppe [8], Dave Grant [8], Michelle Salemi [9], Charlene Nijmeh [10], Monica Arellano [10], Alan Leventhal [10] [11], Brett Phinney [9], Brian F Byrd [5], Ripan S Malhi [4] [7] [12], Glendon Parker [13]

Affiliations + expand

PMID: 32681049 PMCID: PMC7368048 DOI: 10.1038/s41598-020-68550-w

Free PMC article

NEWS > SCIENCE

9000-year-old grave shows women tackled big game

UC Davis study challenges age-old ‘man-the-hunter hypothesis

An illustration of female hur
hunters who may have appe
9,000 years ago. MATTHEW
DAVIS IET ACADEMIC TECH

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Zachary Goecker