Applications of In-Solution DNA Hybridization Capture at the Center for Conservation and Evolutionary Genetics

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CCEG laboratory @NZP

Robert Fleischer

Jesús Maldonado

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Applications

- Ancient DNA (aDNA)
- Population/Conservation genomics
- Species ID
  - Environmental DNA (eDNA)
  - Disease ecology (pathogen detection)
  - Dietary analysis
- Noninvasive DNA

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Molecular Markers

- UCEs
- Mitochondrial genomes
- Custom SNPs
- Specific genes of interest
- Exon capture

Ancient DNA (aDNA)

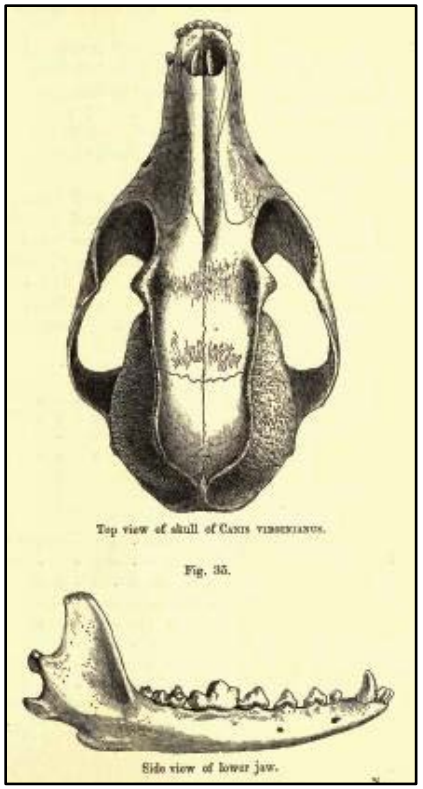
Benefits:

- More cost/time efficient than PCR
- Increase proportion of endogenous DNA
- Heterospecific capture possible
- Multiplex multiple samples/dilute probes

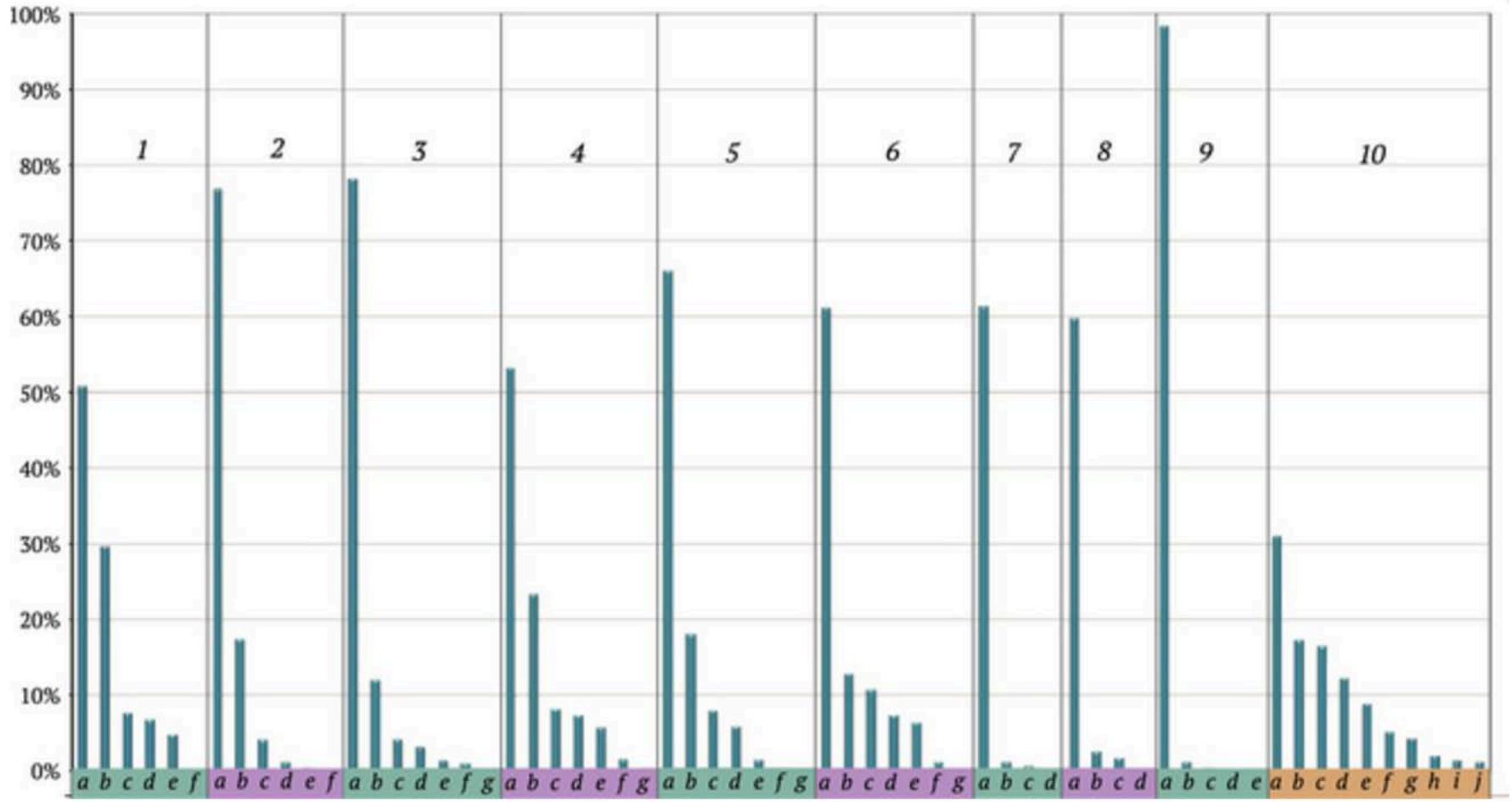
Ancient DNA (aDNA)

Some considerations for aDNA:

- Minimize contamination (dedicated clean lab)
- Dual indexing recommended
- **Can** optimize multiplex
- Lower hybridization temperature
- Minimize PCR cycles; pool multiple rxns
- Increase sequencing effort



Ancient DNA (aDNA)



Hawkins et al. (2015) Mol. Ecol. Res.

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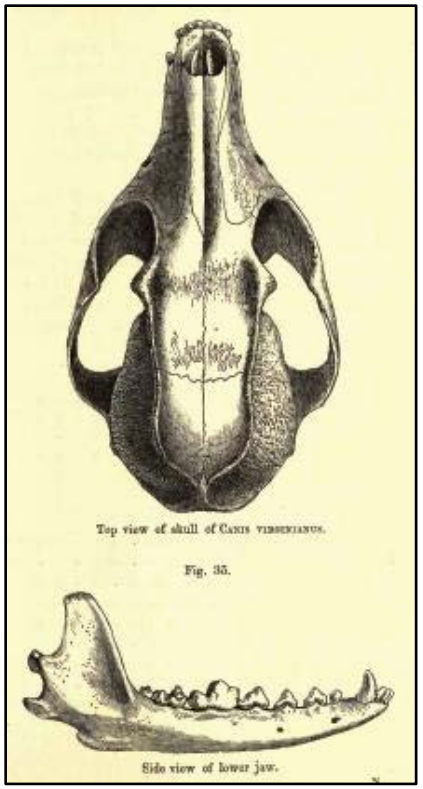
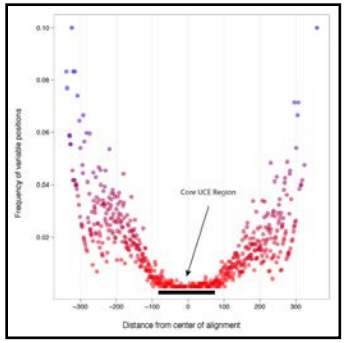
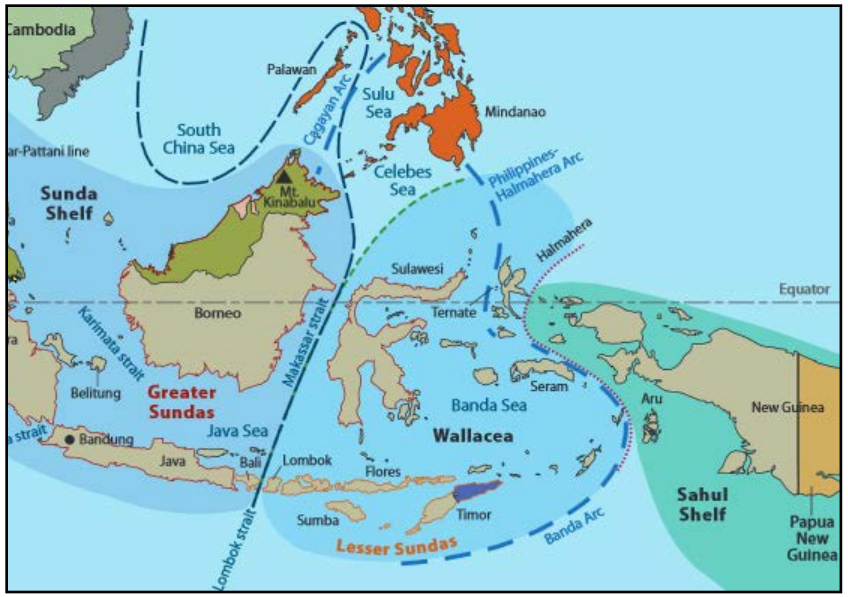
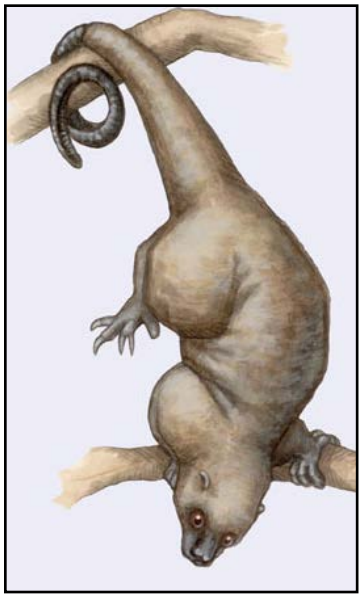


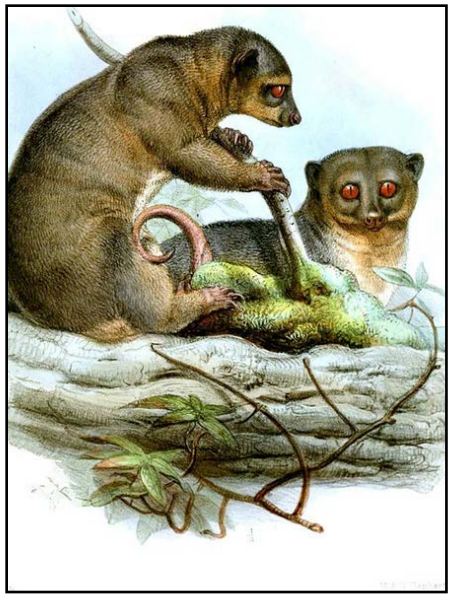
Project: Phylogenomics of the Phalangeridae
Markers: UCEs and Mitogenomes from museum specimens

Lillian Parker, Kristofer Helgen, Jesús Maldonado



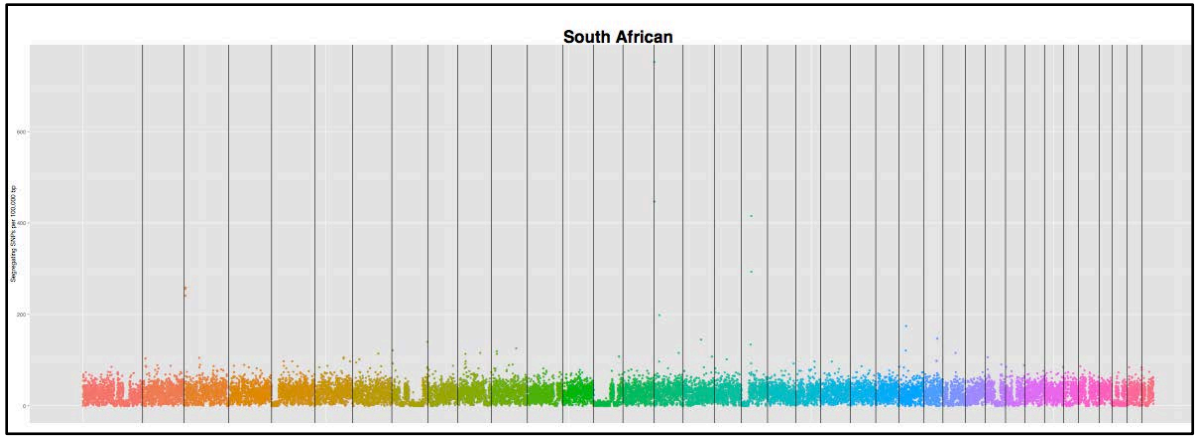


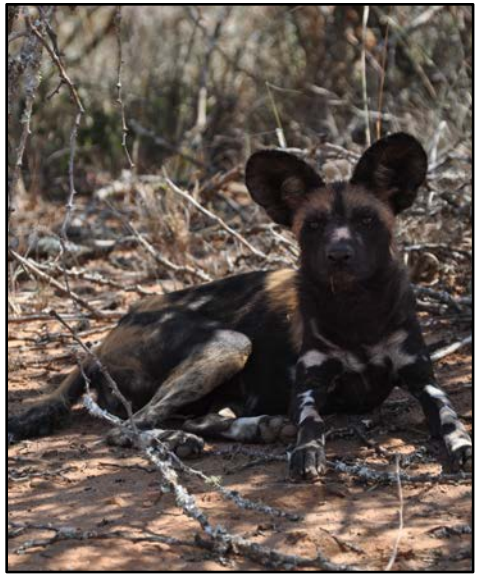




Design custom SNPs

- Use **genome reference** to design SNP bait set
  - Use heterozygous sites in single genome or multiple individuals
- Can also design probes from **RADseq data**
- Custom script by Michael Campana (CampanaM@si.edu)

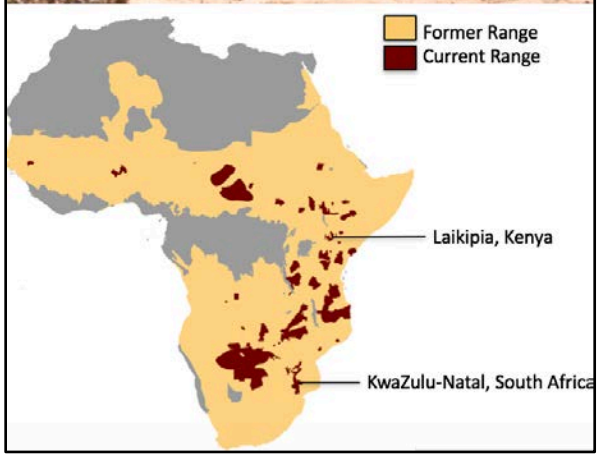




Project: Population Genomics of African Wild Dogs
Markers: Single Nucleotide Polymorphisms

Michael Campana, Robert Fleischer, Kristofer Helgen, et al.

- Sequenced genomes for 2 individuals
- Designed custom SNP baits (~20K) from genome alignment
- 32 extant Kenyan wild dogs
- 17 historic wild dogs



Species Detection

Applications:

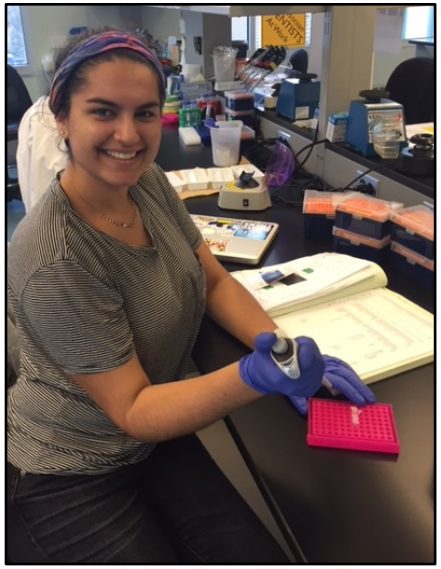
- Diet analysis
- Disease ecology (ectoparasite, host and pathogens)
- Environmental DNA (metagenomics)

Project: Recreating Paleoecosystems using eDNA from Archeological Sites

Markers: Mitochondrial genes (COI, Cytb)

Sabrina Shirazi, Courtney Hofman, Torben Rick and Jesús Maldonado

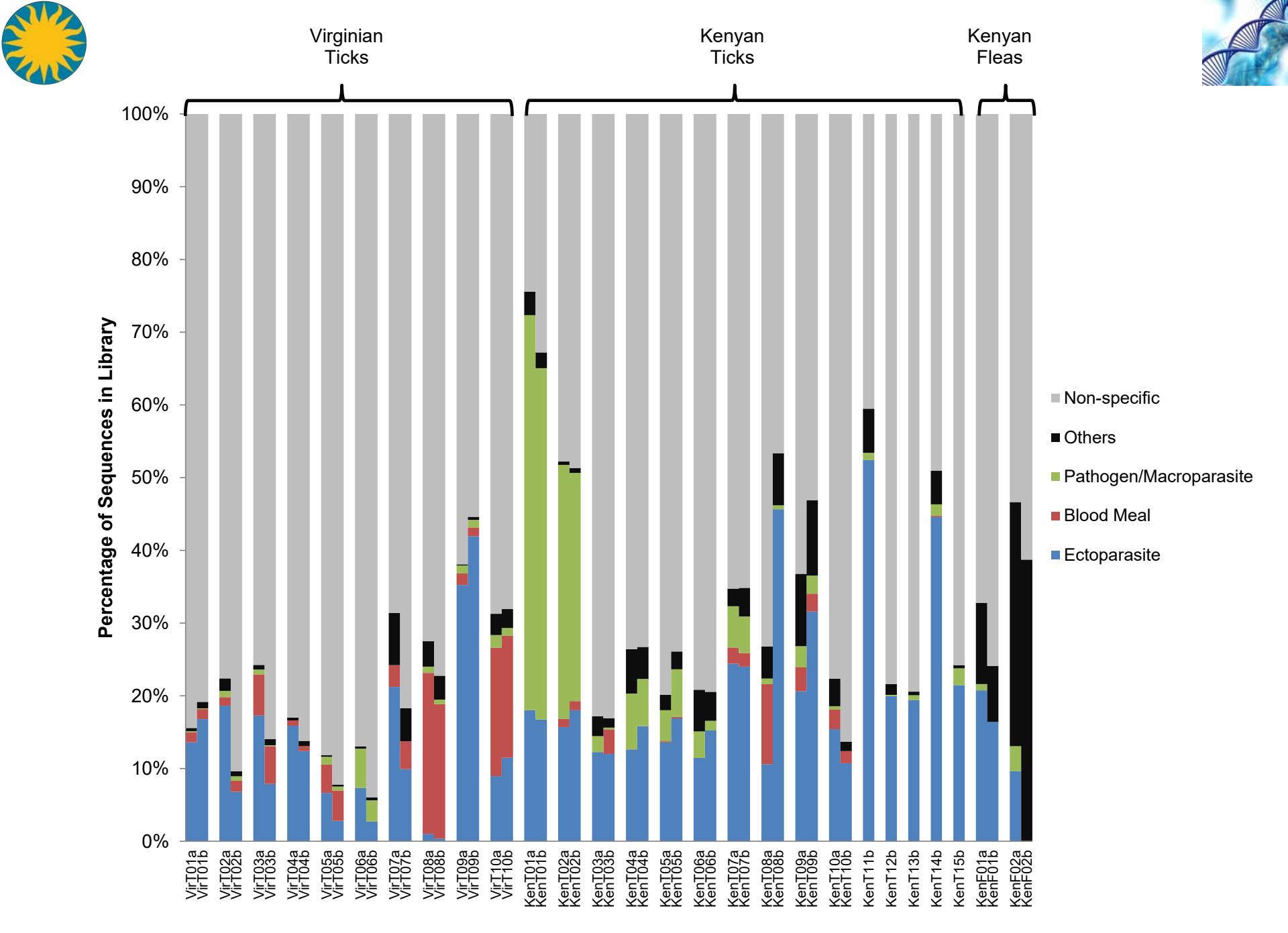
- Baits designed from mtDNA sequences of birds and mammals
- Extract DNA from soil at archeological sites
- Applying method to samples from Santa Cruz Island



Project: EctoBaits
Markers: Published genes for species ID

Michael Campana, Lauren Henson, Jesús Maldonado, Robert Fleisher et al.

- DNA extracted from ectoparasite
- Probe set with baits for **ectoparasite, blood meal, and pathogen**
- Metagenomic analysis
- Applications for disease ecology

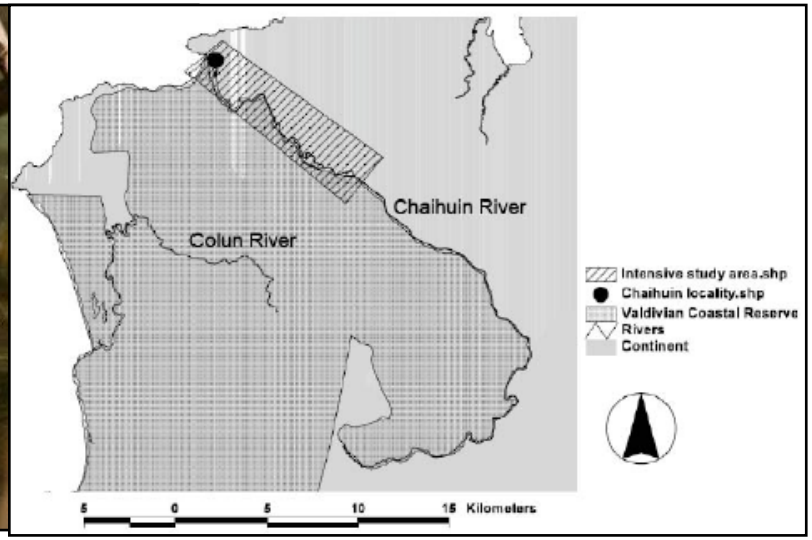

Virginian Ticks
Kenyan Ticks
Kenyan Fleas
Percentage of Sequences in Library
100%
90%
80%
70%
60%
50%
40%
30%
20%
10%
0%
VirT01a VirT01b
VirT02a VirT02b
VirT03a VirT03b
VirT04a VirT04b
VirT05a VirT05b
VirT06a VirT06b
VirT07a VirT07b
VirT08a VirT08b
VirT09a VirT09b
VirT10a VirT10b
KenT01a KenT01b
KenT02a KenT02b
KenT03a KenT03b
KenT04a KenT04b
KenT05a KenT05b
KenT06a KenT06b
KenT07a KenT07b
KenT08a KenT08b
KenT09a KenT09b
KenT10a KenT10b
KenT11b
KenT12b
KenT13b
KenT14b
KenT15b
KenF01a KenF01b
KenF02a KenF02b
Non-specific
Others
Pathogen/Macroparasite
Blood Meal
Ectoparasite

Project: Assessing the impact of invasive mink on Southern river otters in Chile (Noninvasive DNA)

Markers: Mitochondrial genomes

Lilly Parker, Jesús Maldonado, et al.

- Introduced mink and endangered otter, habitat overlap
- Designing baits to capture mitochondrial genomes for all *Lontra* species
- Apply capture technique to DNA extracted from scats



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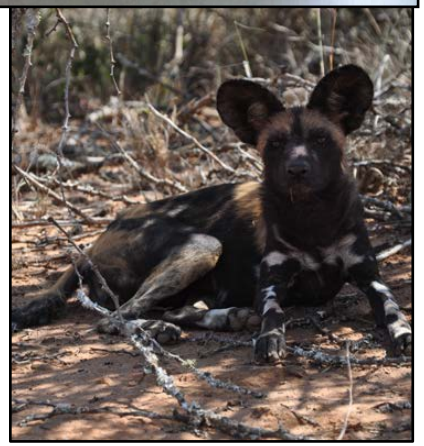

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Thank you! Questions?

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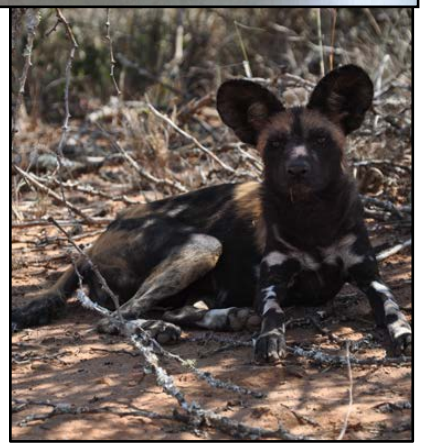
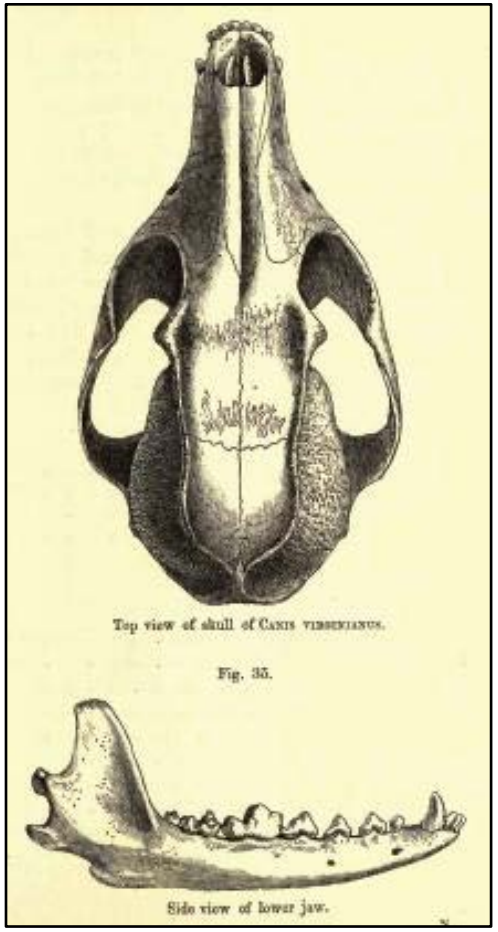


Project: Phylogenomics of *Urocyon cinereoargenteus*
Markers: UCEs and Mitogenomes from museum specimens

Susette Castañeda Rico and Jesús Maldonado

- Range from southern Canada to Venezuela and Colombia
- 16 described subspecies
- Sample 7 subspecies and investigate history in Mexico and Central America



Project: Conservation genomics of the Mariana crow Markers: Single Nucleotide Polymorphisms

Nandadevi Cortes-Rodriguez, Michael Campana, Robert Fleischer

- Mariana Crow found only on Guam and Rota Islands
- Conservation concern: threatened by loss of habitat and predation by brown tree snake
- Designed custom SNPs from *de novo* assembled genome
- Capture & sequence SNPs from many individuals
- Genetic diversity, population structure, sex ratios, Ne