

Genomic Sequencing across a Zone of Secondary Contact Uncovers Complex Demographic History and Admixture between Cryptic Gray Fox Lineages

Sophie Preckler-Quisquater¹, Elizabeth Kierepka², Dawn M. Reding³, Antoinette J. Piaggio⁴, Seth Riley⁵ and Ben Sacks¹

(1) Mammalian Ecology and Conservation Unit, Veterinary Genetics Laboratory, School of Veterinary Medicine, University of California, Davis, Davis, CA;

(2) North Carolina State University

3) Luther College

4) USDA/APHIS National Wildlife Research Center

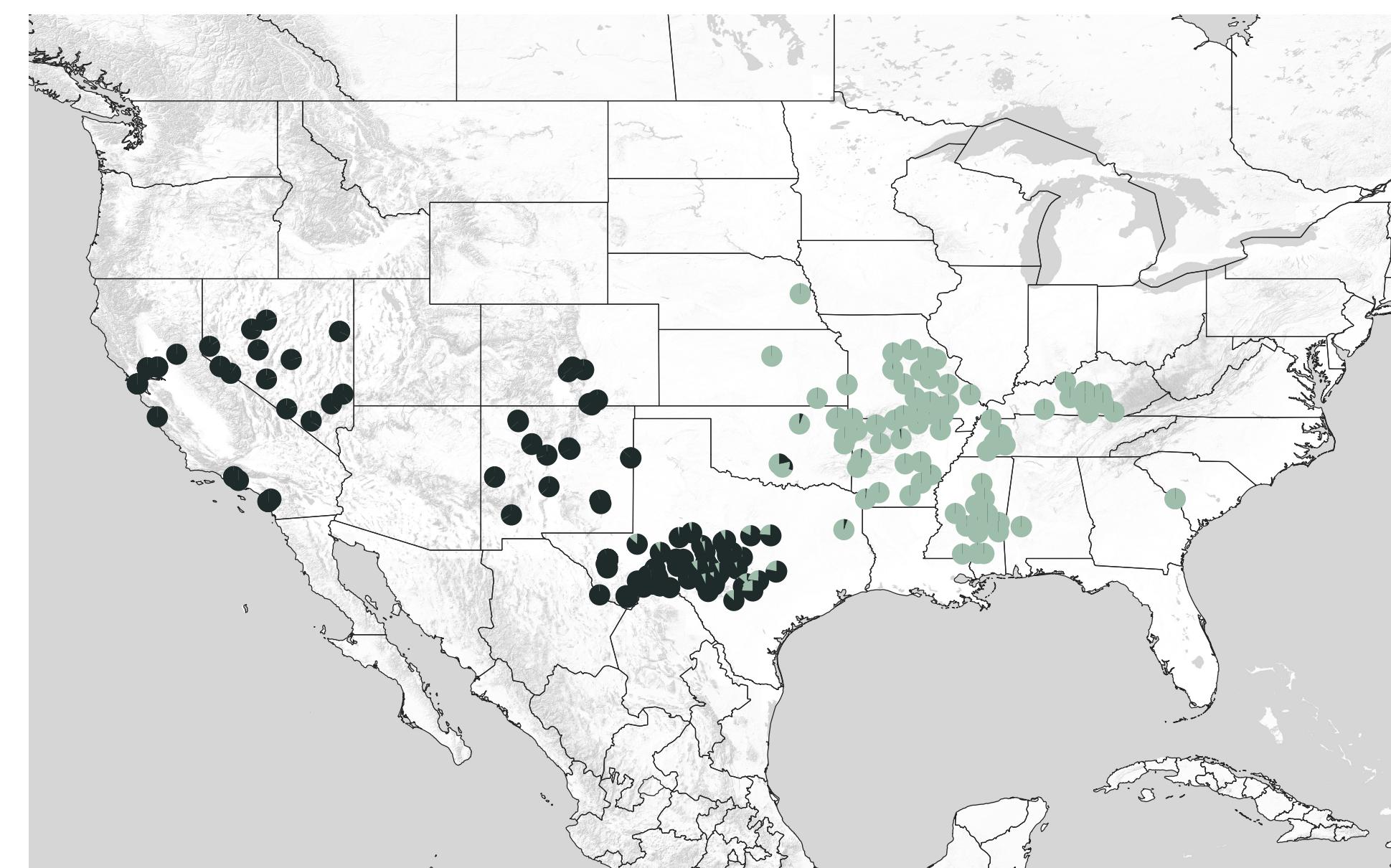
5) United States National Park Service, Los Angeles, CA

Introduction

Past climatic fluctuations have heavily influenced current species distributions, generating complex evolutionary histories through periods of isolation in refugia as well as secondary contact and gene flow post-expansion. The gray fox (*Urocyon cinereoargenteus*) represents the most basal extant canid lineage and occurs only in the Americas. Previous mitochondrial analysis identified deeply divergent (up to 1 Mya) eastern and western lineages, and a major phylogeographic break between lineages along the Great Plains Suture Zone, indicating that gray foxes have likely been isolated for numerous glacial-interglacial cycles.^{1,2} However, it is still unclear whether these lineages maintained complete reproductive isolation during this time, or whether there were periods of secondary contact and admixture post-divergence. Using a combination of reduced-representation ($n = 259$) and whole-genome ($n = 42$) sequencing of gray foxes across their US range, we generated estimates of nuclear split times and assessed concordance with previously published mitochondrial estimates.^{1,2} We additionally quantified genome-wide ancestry proportions, to identify whether there was any evidence of nuclear admixture between eastern and western lineages at the previously described contact zone. Using a local ancestry inference approach, we additionally tested whether gene flow was recent (potentially due to human induced landscape changes in the last 100 years) or whether it occurred during an older post-Pleistocene expansion event. We also explored whether selective introgression of beneficial genetic variation may have played a role in the evolutionary history of these two lineages. Understanding the complexities surrounding divergence and secondary contact between these gray fox lineages will allow us to better understand the role past and future climate shifts may play in the overall diversity of species.

Preliminary Results

Global Ancestry Inference



Divergence Time Estimation

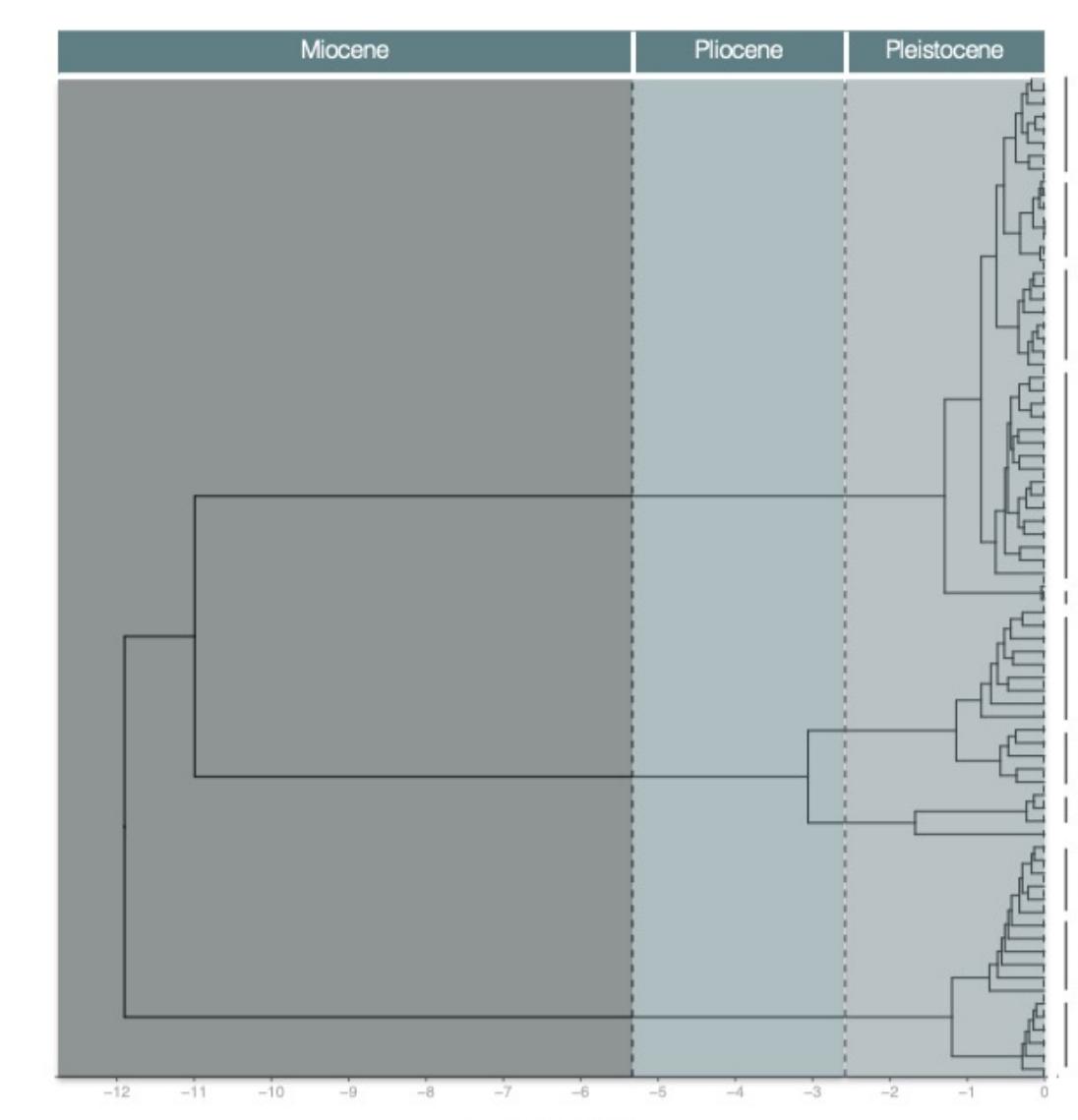


Fig 2. A time-calibrated semi-parametric phylogeny indicated that gray fox divergence likely occurred sometime between the divergence times of Eurasian/North America red fox, and arctic/kit fox. This supports a mid-Pleistocene split, in line with previously published mitochondrial estimates, and is much older than splits typically used to classify intraspecific divisions in North American carnivores.

Estimating Timing of Admixture Pulses

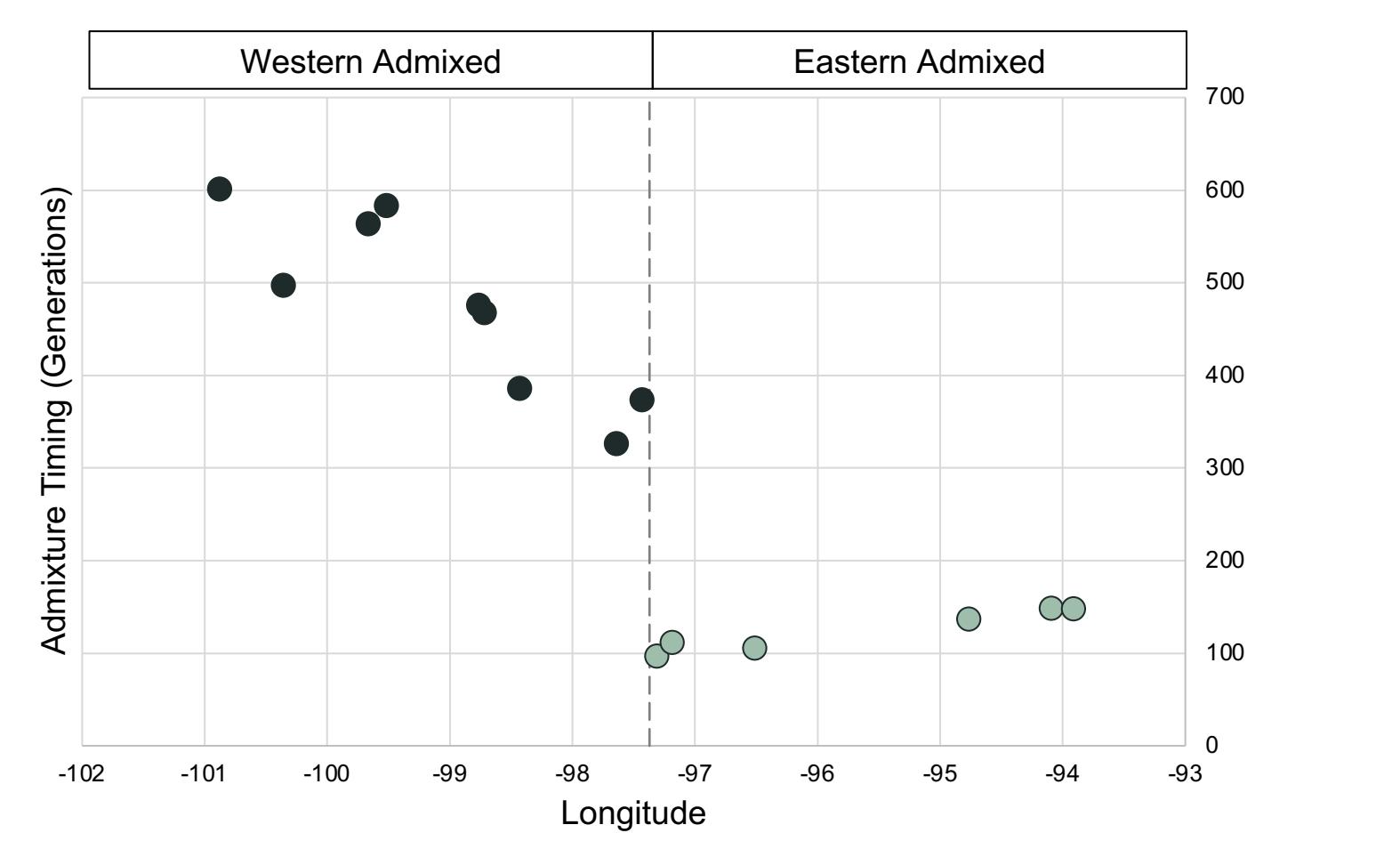


Fig 4. Preliminary analyses identified distinct admixture pulses in the western (left; dark green) and eastern (right; light green) populations. The western admixed population appears to have experienced an older pulse of eastern gene flow (300 – 600 generations ago), while the eastern admixed population experienced a more recent pulse of western gene flow (100 – 150 generation ago).

Local Ancestry Inference

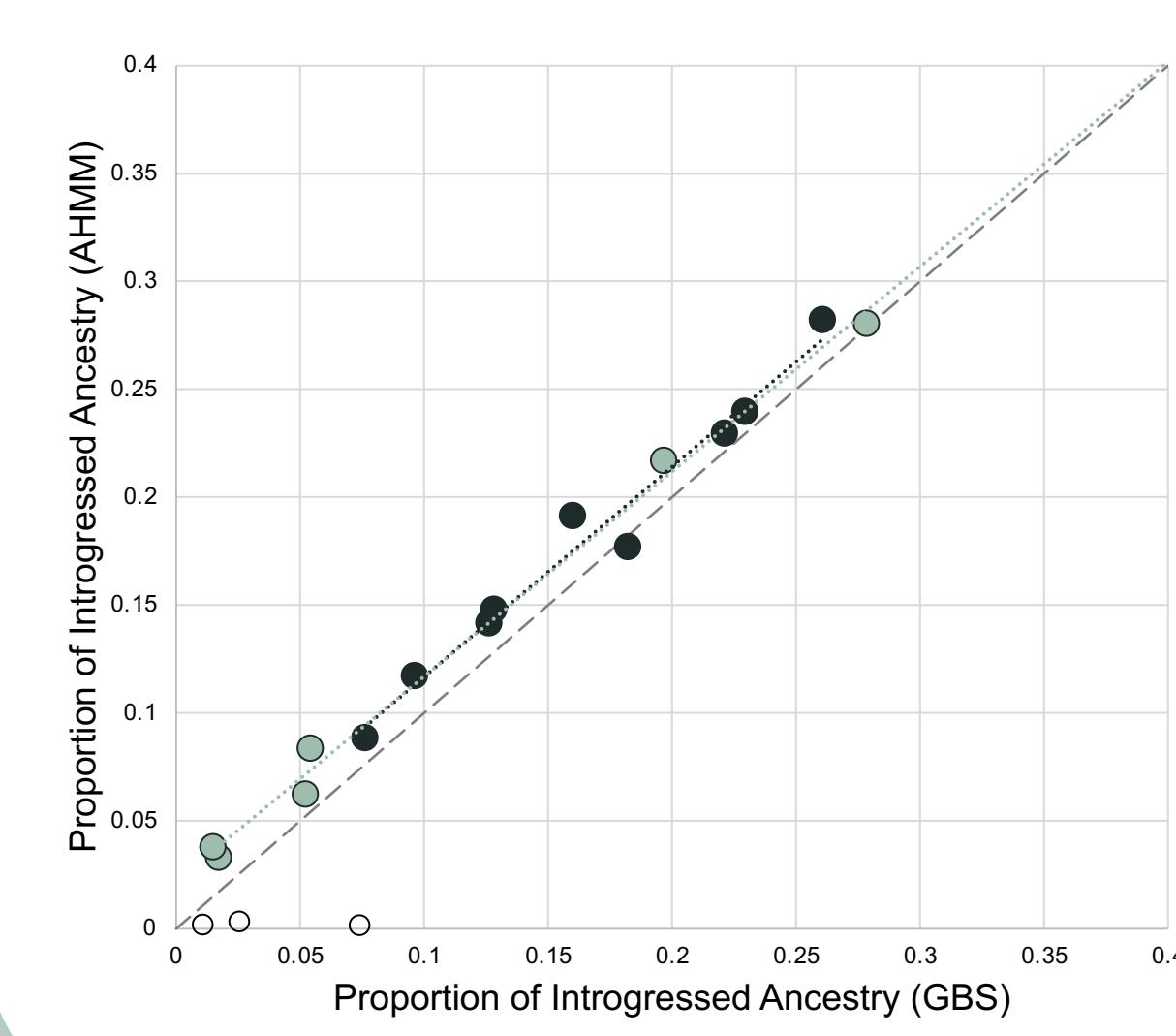


Fig 3. Relative proportions of introgressed ancestry were highly concordant for both western (dark green) and eastern (light green) admixed individuals when comparing our global ancestry approach (44,931 GBS loci + Bayesian clustering approach), and our local ancestry inference (500k AIM loci + Hidden Markov Model). Open circles indicate three samples that were identified as admixed in our global ancestry approach but had <1% introgression in our local ancestry inference and were therefore removed from downstream analyses.

Identifying Selectively Introgressed Regions

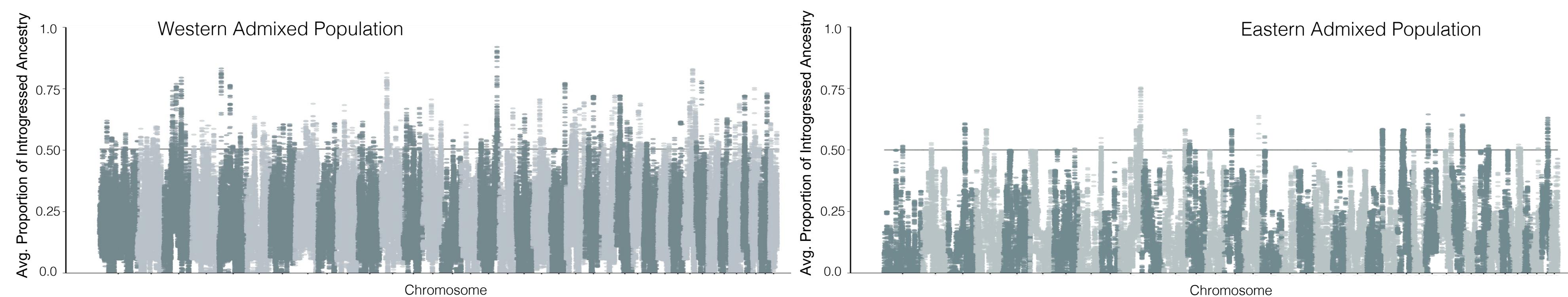
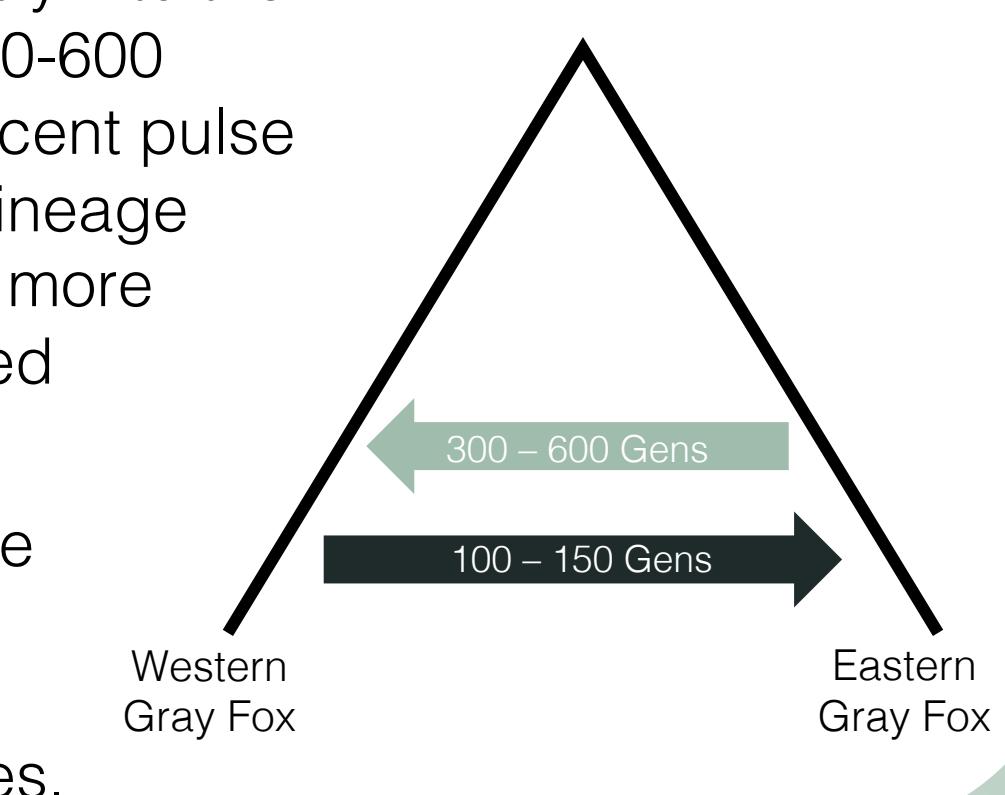


Fig 5. Proportion of eastern and western introgressed ancestry at each SNP averaged across all individuals within the western (left panel; $n=9$) and eastern (right panel; $n=6$) admixed populations, respectively. We identified the top 1% outliers, denoted by the gray horizontal line, for the western (>0.504) and eastern (>0.499) admixed populations based upon the distribution of the average introgression proportions for all loci across the genome.

Conclusions



- A zone of secondary contact with nuclear gene flow between eastern and western gray fox lineages was identified in the southern Great Plains region.
- Divergence estimates correspond to the Early-Middle Pleistocene Transition (1.4 – 0.4 mya) in the Irvingtonian land mammal age, which is substantially older than those typically characterizing intraspecific divisions for most North American carnivores.^{8,9}
- We identified an older pulse of eastern ancestry into the western gray fox population approximately 300-600 generations ago. We also identified a more recent pulse of western ancestry into the eastern gray fox lineage approximately 100-150 generations ago. This more recent pulse may coincide with human-induced landscape changes facilitating migration.
- Several regions may have undergone selective introgression across the zone of secondary contact, however further analyses will be required to identify any putative functional roles.



References

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