A photograph of a green leopard frog with dark spots and stripes, sitting on a concrete ledge next to a body of water. The frog is facing right. The background shows a blurred view of the water and some greenery.

A Genetic Assessment of the Northern Leopard Frog in Northern New Mexico

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- *Lithobates pipiens*
 - *Widespread Decline Across the western United States*
 - *Enigmatic Decline (climate change, disease, pesticides, habitat loss)*
- Why this specific species of frog?
 - Native to northern NM
 - Morphological Intergrades between species in this area
 - Hybridization..factor for decline?



Causes of Declines

- Habitat degradation
 - Widespread problem
 - Caused by different factors
 - (deforestation, urbanization, drought)
- Can affect animal populations
 - Limit Gene Flow
 - Isolate populations
 - Increase hybridization?



Habitat and Morphological Differences



L. pipiens

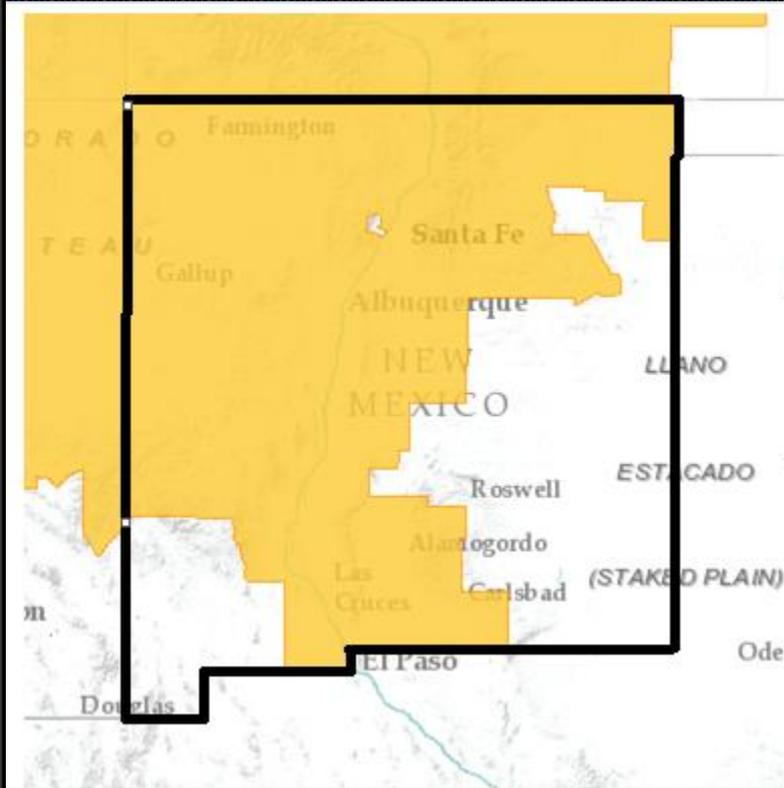
- Larger elevation range



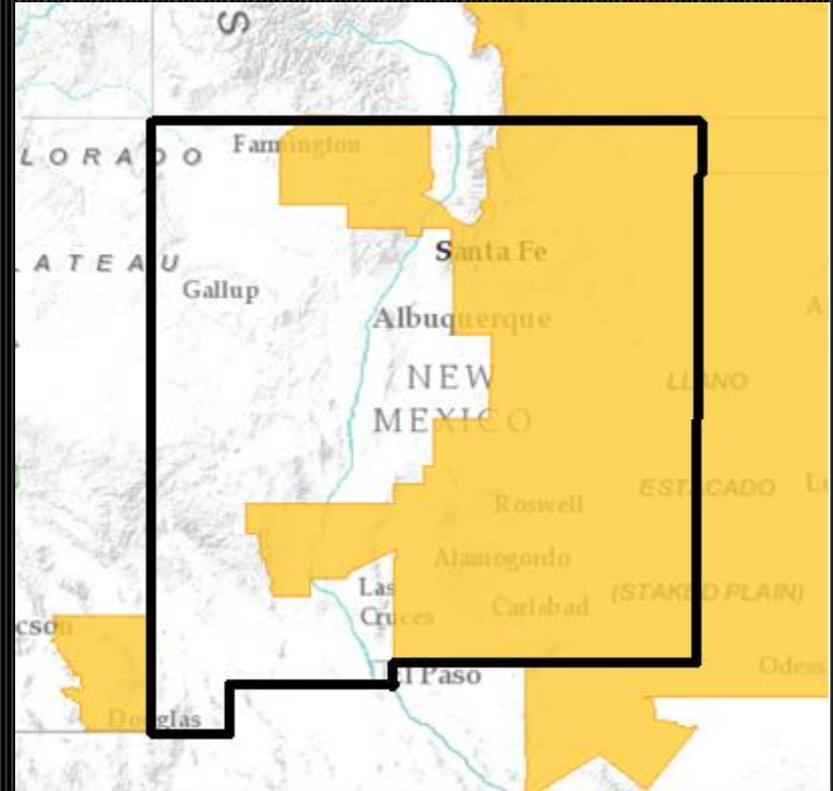
L. blairi

- Lower elevation plains

Habitat Overlap



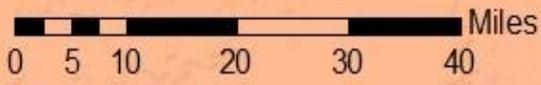
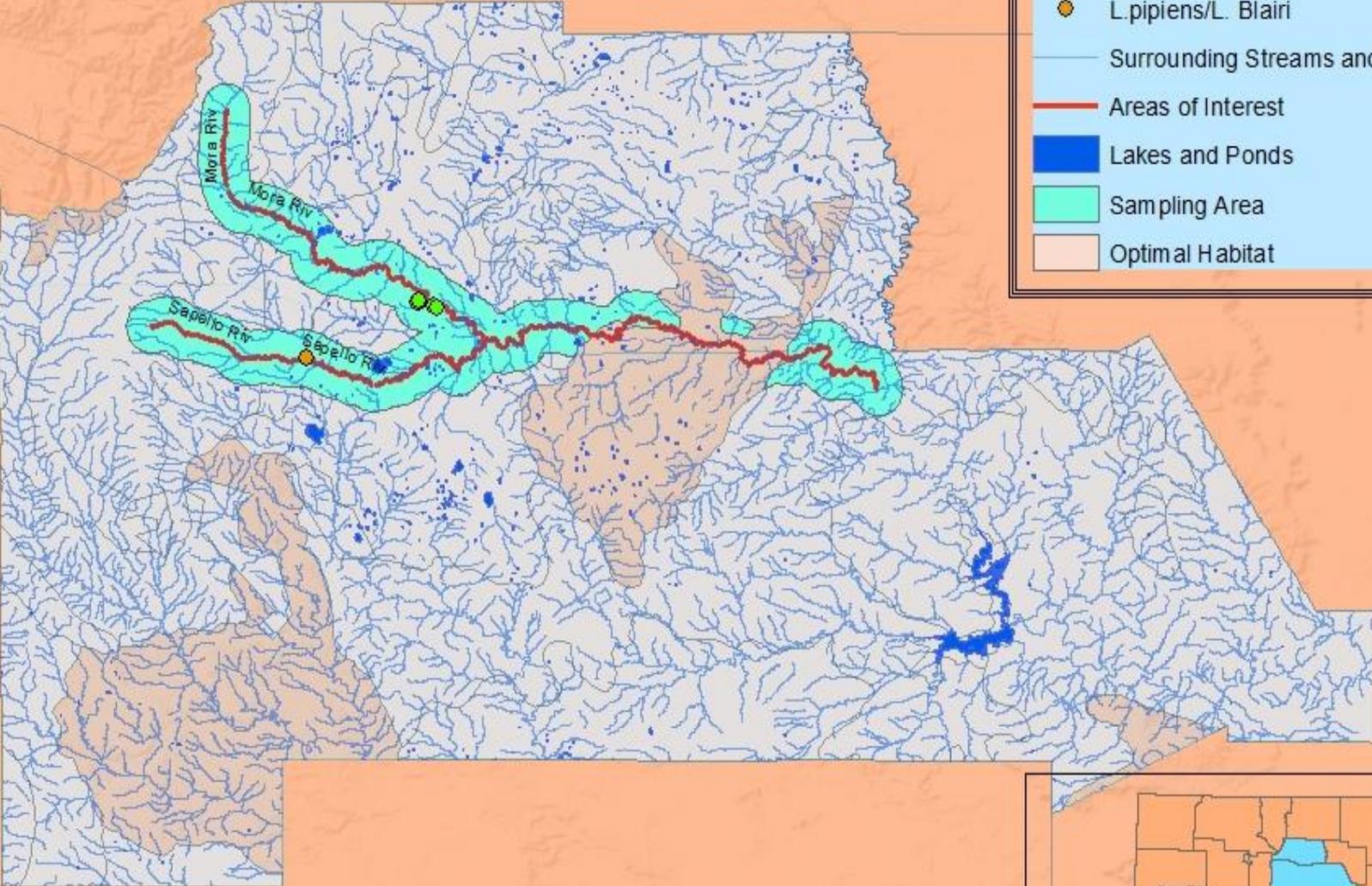
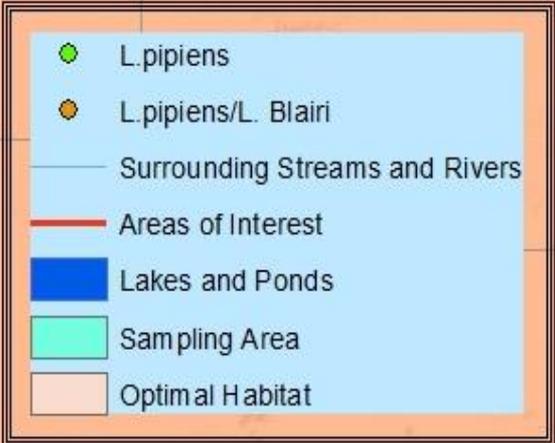
L. pipiens



L. blairi

Sampling Area

- Area of Interest
 - Northern New Mexico along the Mora and Sapello Rivers
- Why the specific sampling area?
 - Opportunity to add to existing data from other areas of the United States
 - Useful to study behavior and populations under changing conditions
 - Drought issues in New Mexico



Purpose

- Assess the population structure of *L. pipiens* at sites along the Mora and Sapello Rivers and investigate the presence of hybrid individuals.



Areas Sampled



Sapello River



Mora River

Leopard Frog Samples from Different Habitats

Sapello River

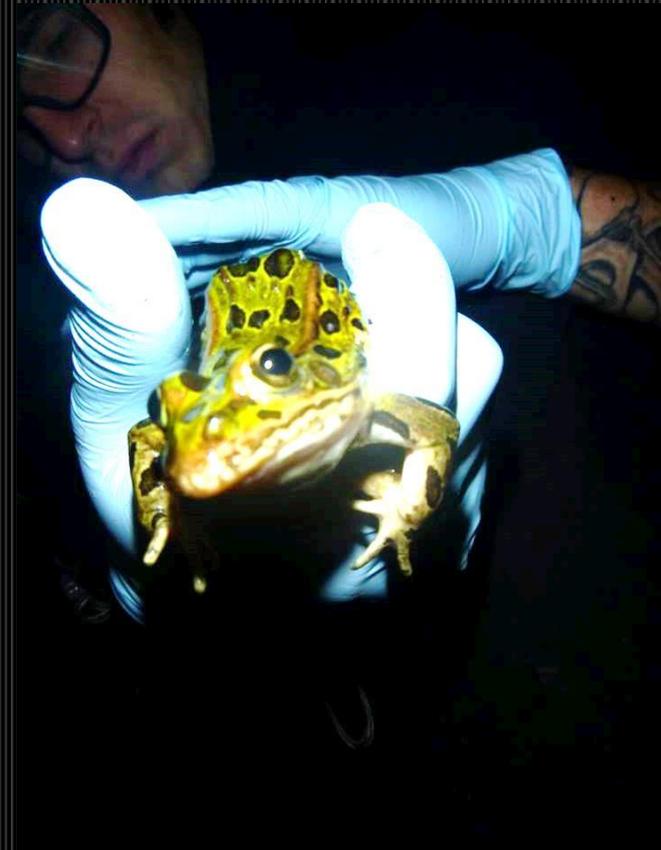


Mora River



Sampling Methods

- Sampling
 - Mora and Sapello Rivers
 - Opportunistic catch and release
 - GPS to mark location
- Genetic Analysis
 - Buccal Swabbing for DNA
 - DNA Extraction
 - PCR
 - Sequencing/Genotyping



Genetic Analysis

- Fibrinogen Intron 7 (FIBI7)
- Microsatellites
- ND1

Indels in FIB Sequences

380 390 400 410 420 430

CATCTCTATATCTCAATGCATTACATCTCTATATCTCTCTTTGAGCTATATAAAATATAC
CATCTCT-----GAGCTATATAAAATATAC
CATCTCTATATCTCAATGCATTACATCTCTATATCTCTCTTTGAGCTATATAAAATATAC
CATCTCTATATCTCAATGCATTACATCTCTATATCTCTCTTTGAGCTATATAAAATATAC
CATCTCTATATCTCAATGCATTACATCTCTATATCTCTCTTTGAGCTATATAAAATATAC
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CATCTCT-----GAGCTATATAAAATATAC



Lp12a

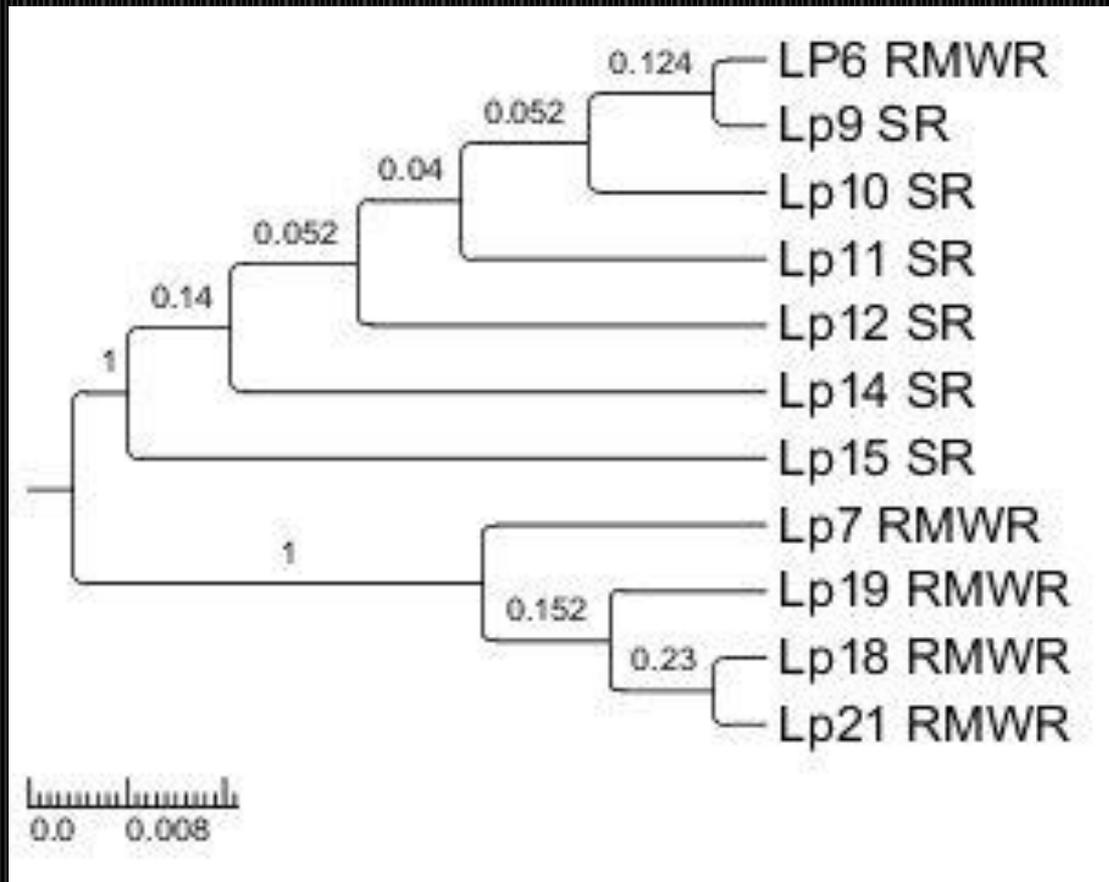
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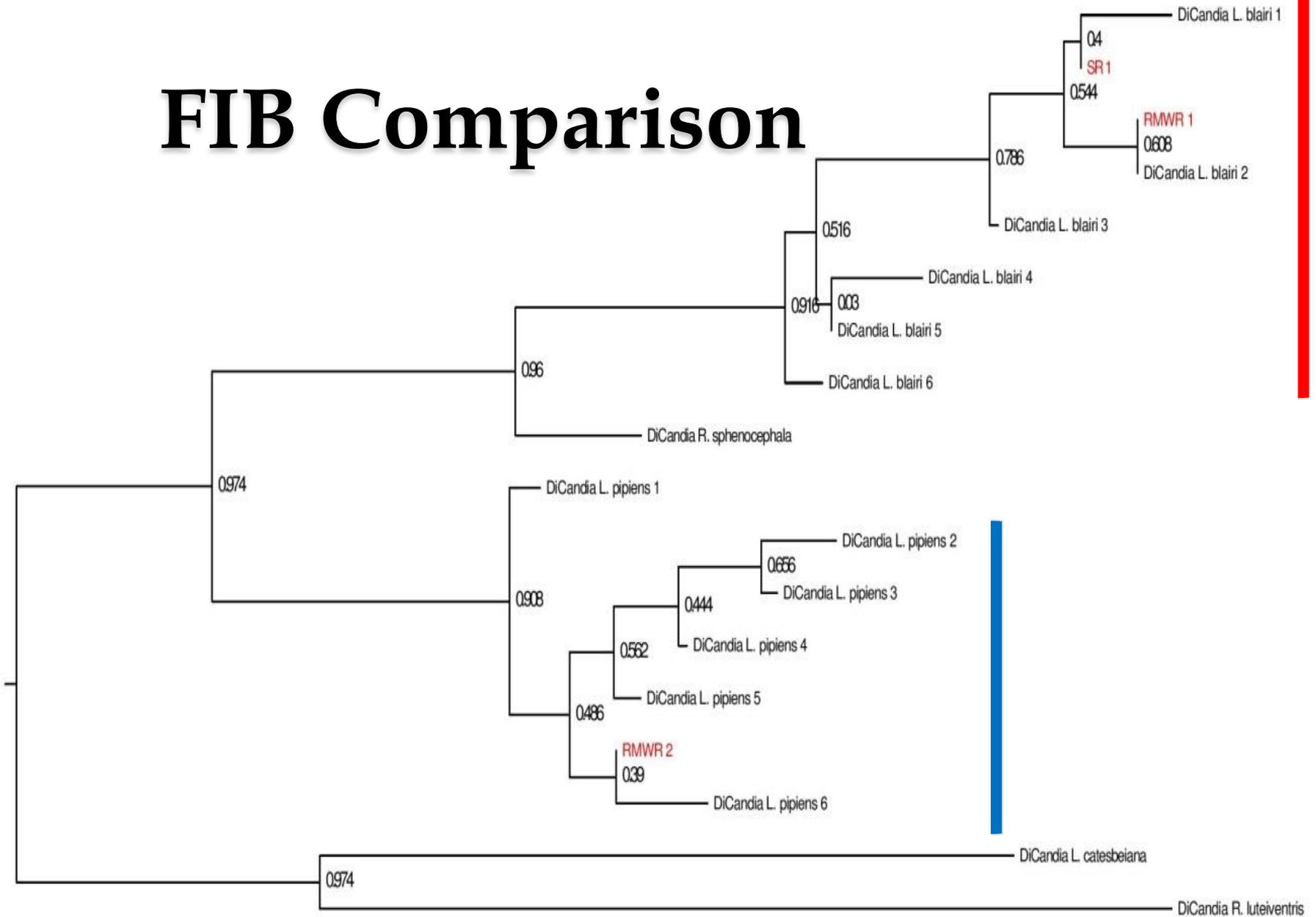
Lp19a

• CATCTCT-----GAGCTATATAAAATATAC

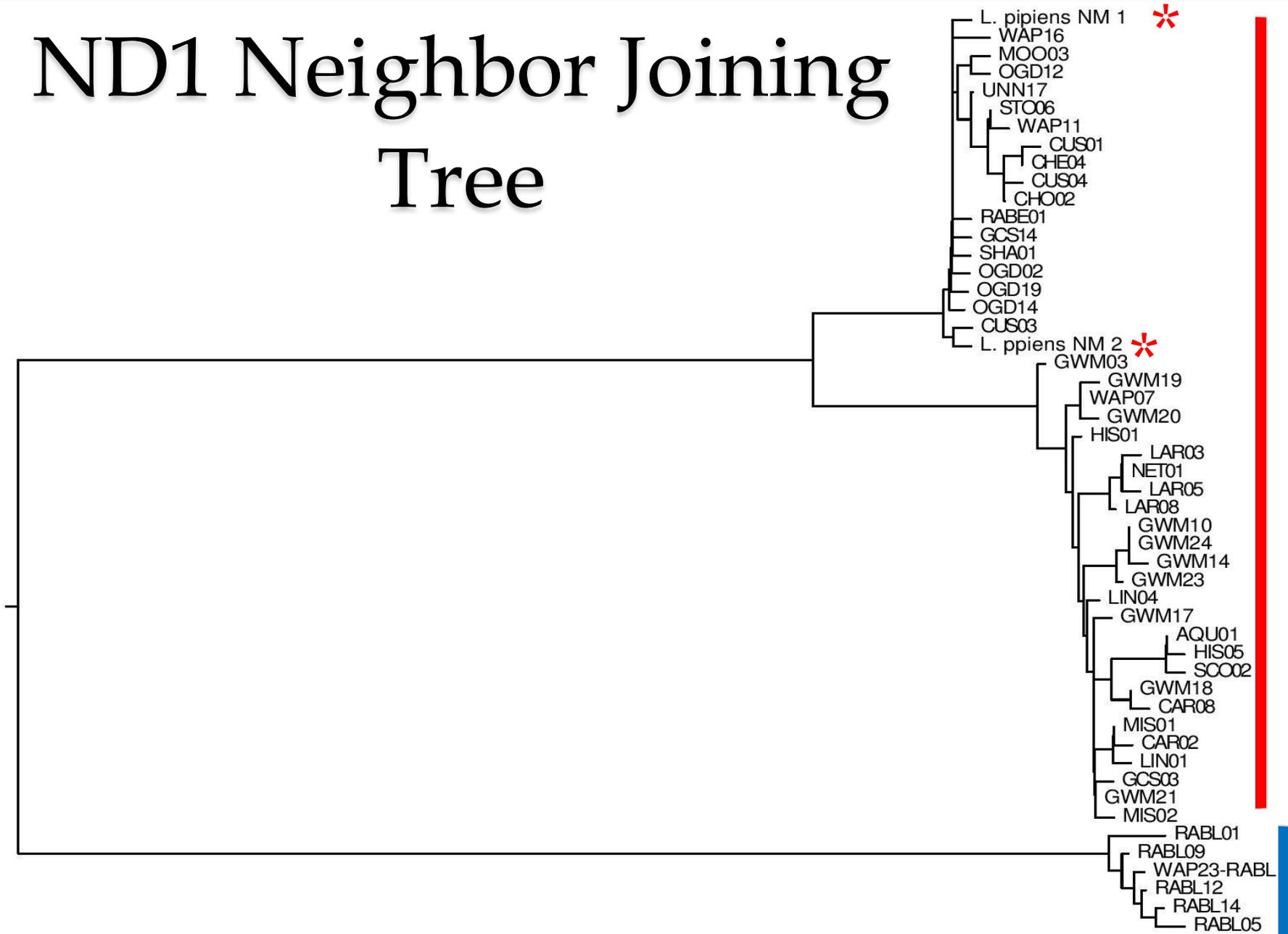
FIB Neighbor Joining Tree Results



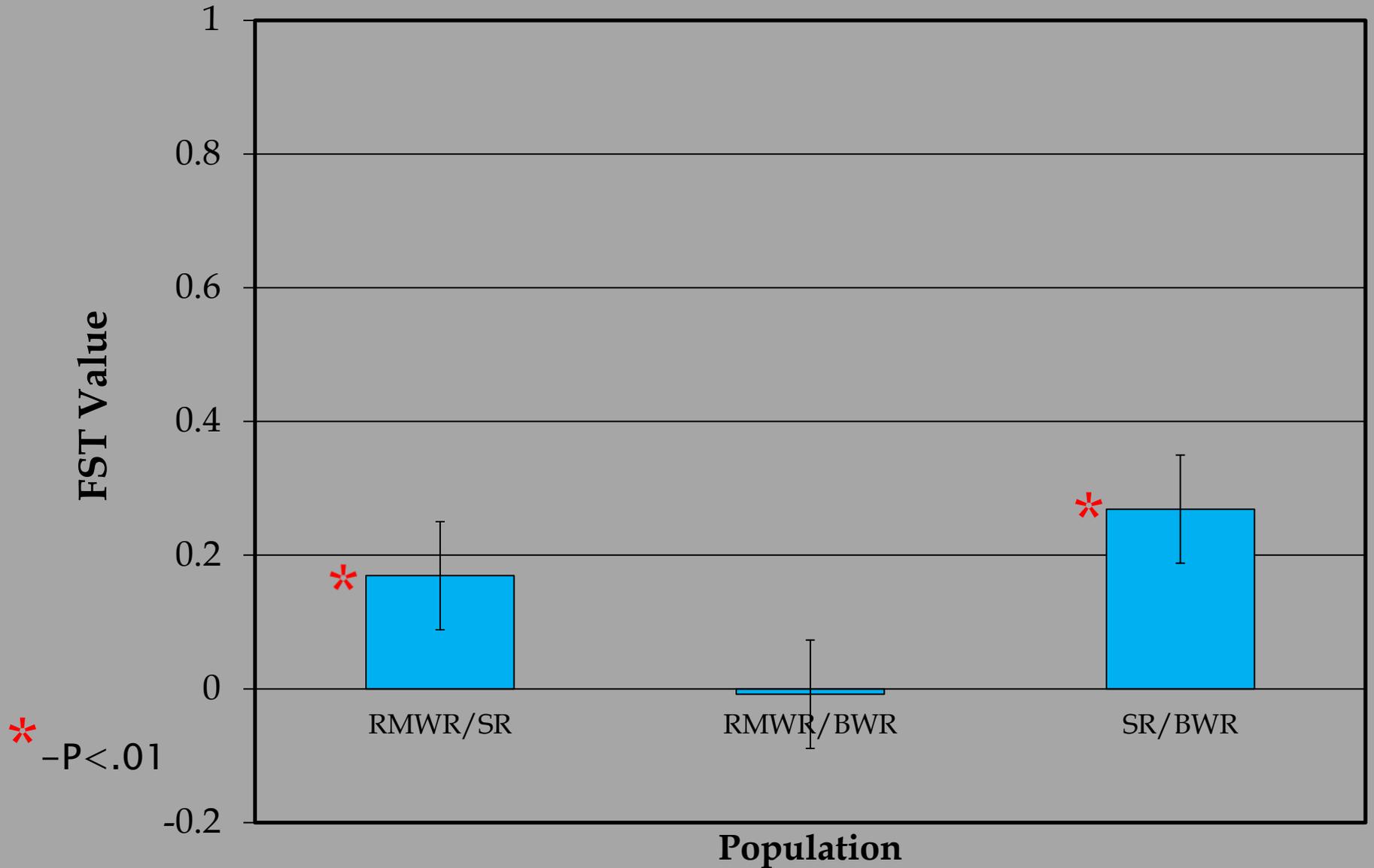
FIB Comparison



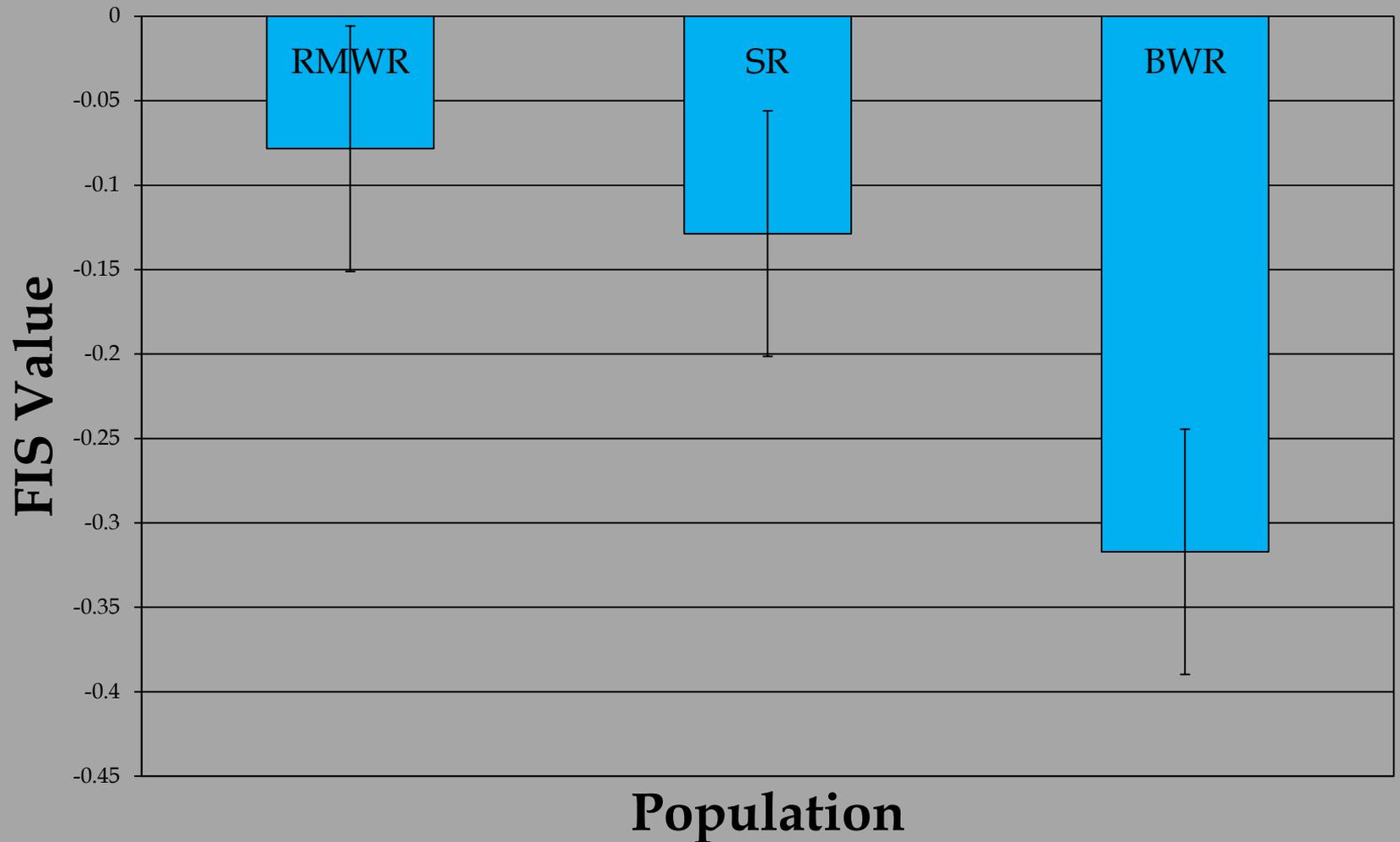
ND1 Neighbor Joining Tree



Microsatellites-Population Structuring



Inbreeding (FIS) of Populations



Conclusion

- In the samples presented, morphological classification does not match results of the FIBI7 marker.
- ND1 results illustrate discordance between nuclear and mitochondrial areas.
- Strong population structure between all samples along the Mora River compared to Sapello indicate the possibility of a barrier inhibiting complete panmixia.

- Based on the discordance between nuclear and mitochondrial markers, a hybrid zone is present in the Sapello River.
- Two rivers that are connected still do not provide a guaranteed corridor for dispersal.
- Even though populations were structured, within population isolation is not to the point of inbreeding.



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Significance

- The results of this research indicate that hybridization may be occurring more than previous literature has suggested.
- Outbreeding depression may be an indicator of the inability to disperse and breed with conspecifics.
- If the climatic trend we are seeing persists, these results may begin occurring at a larger scale which will only add to the many stressors present to leopard frog populations.



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Questions?

