



# Transcriptome assembly, annotation, and comparative analysis for characterizing adaptive potential of the Streamside Salamander (*Ambystoma barbouri*)



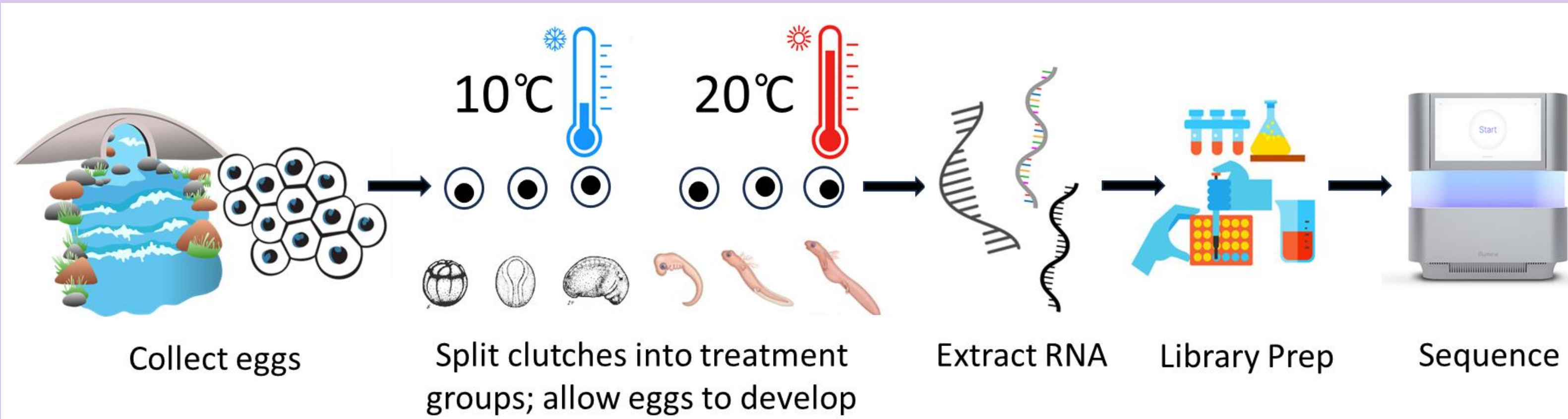
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## Introduction

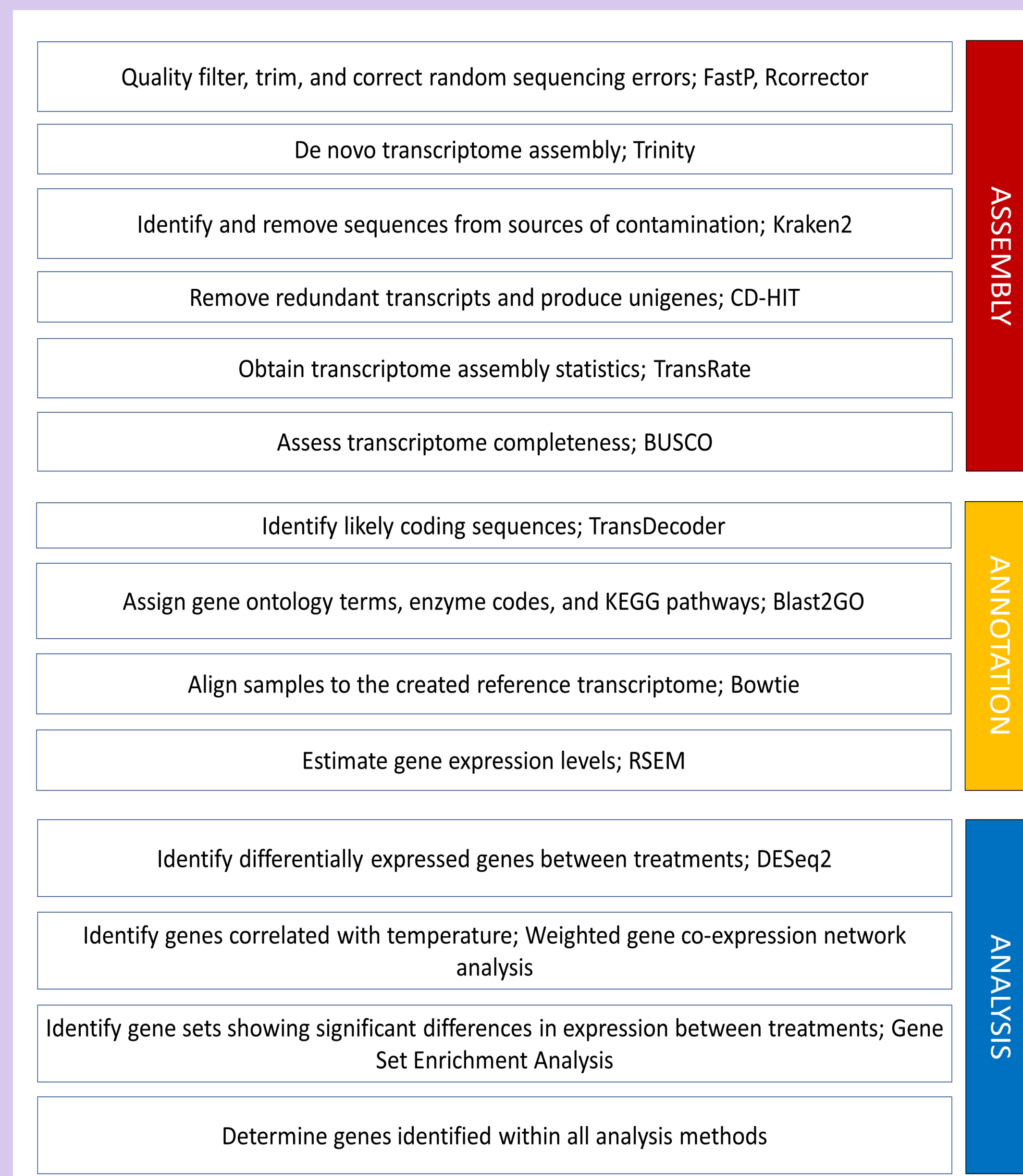
- Habitat loss and climate change are some of the greatest threats to amphibian biodiversity
- The persistence of species in the face of climate change depends on underlying genetic variation that allows populations to adapt
- The transcriptome is the complete set of mRNA molecules expressed within a cell
- Transcriptome studies characterize protein-coding genes to detect genes and pathways involved in responses to environmental conditions and understand the molecular mechanism of specific physiological processes
- Through transcriptome sequencing, adaptively significant gene regions can be identified and population-level gene expression can be compared. This information can then be used to predict population responses to future environmental conditions

## Methods

### Common Garden Experiment



### Bioinformatic Pipeline



## Results

Species	Total transcripts	Mean length	N50	GC content (%)
<i>A. barbouri</i> average	162358	810	1740	0.46
<i>Plethodon cinereus</i>	117812	1084	2345	
<i>Salamandra salamandra</i>	1146571		1529	44.70
<i>Hyla sarda</i>	1295741		914	44.00
<i>Bombina pachypus</i>	896992	616	1082	39.87

Table 2. Assembly statistics generated by TransRate for the averaged *A. barbouri* individual assemblies and published assemblies of other amphibian species

	Complete (%)	Fragmented (%)	Missing (%)
<i>A. barbouri</i> de novo reference	93.8	2	4.2
Average of individual assemblies	67.4	7.4	25.2
Minimum of individual assemblies	43.2	4.4	13.3
Maximum of individual assemblies	82.2	12	45.4

Table 3. Busco scores to assess the completeness of transcriptome assemblies

## Discussion and Next Steps

- Assemblies possess quality similar to those of related species in published literature (Table 2)
  - Suggests successful wet lab, sequencing, and bioinformatic protocol
  - Provides confidence in moving forward with annotation and analysis
- Due to the individual assembly average Busco completeness scores, data from individuals in the same clutch-treatment group should be merged to provide more complete transcriptome assemblies

## Management applications

- Monitor adaptation to climate change and assess ability of populations to acclimate to future conditions
- Guide temperature-related land management actions
- Prioritize populations with valuable adaptive genetic variation
- Provide insight into population response to future climatic conditions and assess long-term viability of populations

## Acknowledgements

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Fig. 1. Streamside Salamander

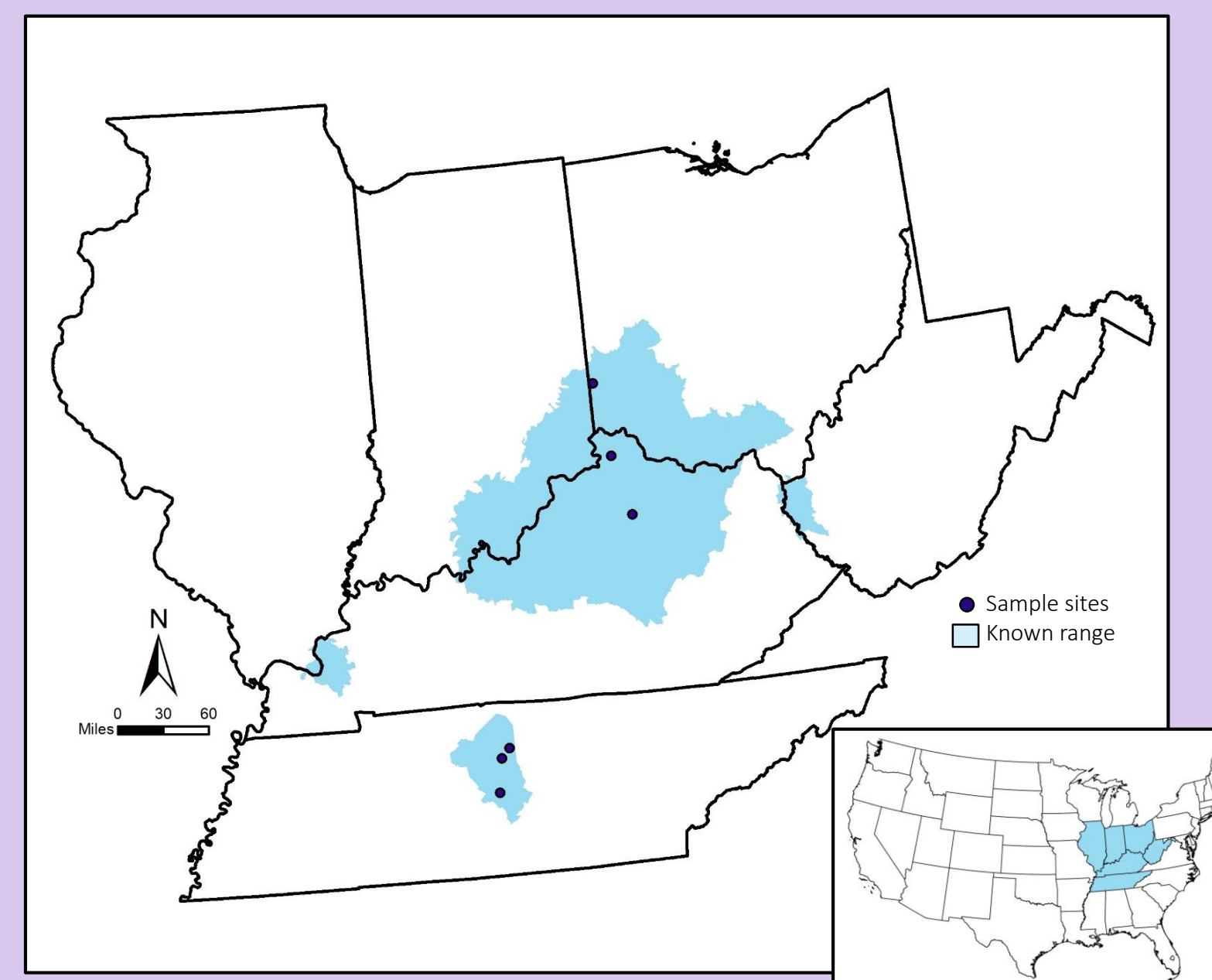


Fig. 2. *A. barbouri* distribution and collection sites

### Study species: *Ambystoma barbouri* (Fig. 1)

- Near threatened with declining populations
- Currently under review for updated status

**Sample sites:** Represent the latitudinal expanse of the species' distribution (Fig. 2)

### Overarching Goals

- Create conservation genomic resources for downstream application
- Quantify variation temperature-mediated gene expression to understand population level responses to environmental conditions

State	County	Site	10°C	20°C	Total
Ohio	Preble	Hueston Woods	6	6	12
Kentucky	Scott	J. Whitfield	6	6	12
	Boone	Beaver Road	6	6	12
Tennessee	Wilson	Fair Grounds	9	9	18
	Wilson	Williams Farm	6	6	12
	Rutherford	Leconte Court	7	7	14