

Riverscape genetics defines population connectivity in Golden (*Tor putitora*) and Chocolate Mahseer (*Neolissochilus hexagonolepis*) (Cyprininae: Torini) in Bhutan

Marlis Douglas, Tyler K. Chafin, Zachery Zbinden, Karma Wangchuk, Changlu, Gopal Prasad Khanal, Pema Norbu, Sangay Norbu, Sonam Dorji, Singye Tshering, Julie E. Claussen, David P. Philipp, Michael E. Douglas



University of Arkansas



Ministry of Agriculture
& Forests
Department of Livestock



Fisheries Conservation
Foundation



Golden Mahseer in Bhutan:

What is known?

- Migrate (spawning?)
- Aggregate at tributaries
- Site-fidelity

Fish Migrations

Synchronized movements of thousands of fish

- Predictable
 - When
 - Where
- Hypotheses
 - Causes
 - Consequences





Fish Dispersal

Movement of individuals to new areas (populations)

= gene flow

= connectivity

- Opportunistic
 - Unpredictable
 - Environmental response
- Hypotheses

Fish Movements: How to Measure?

Telemetry

= Fish Migration

- Tracks individuals
- Predictable movements

Genetic Analyses

= Fish Dispersal + Migration

- Tracks populations
- Over many generations

Radio Tracking



DNA

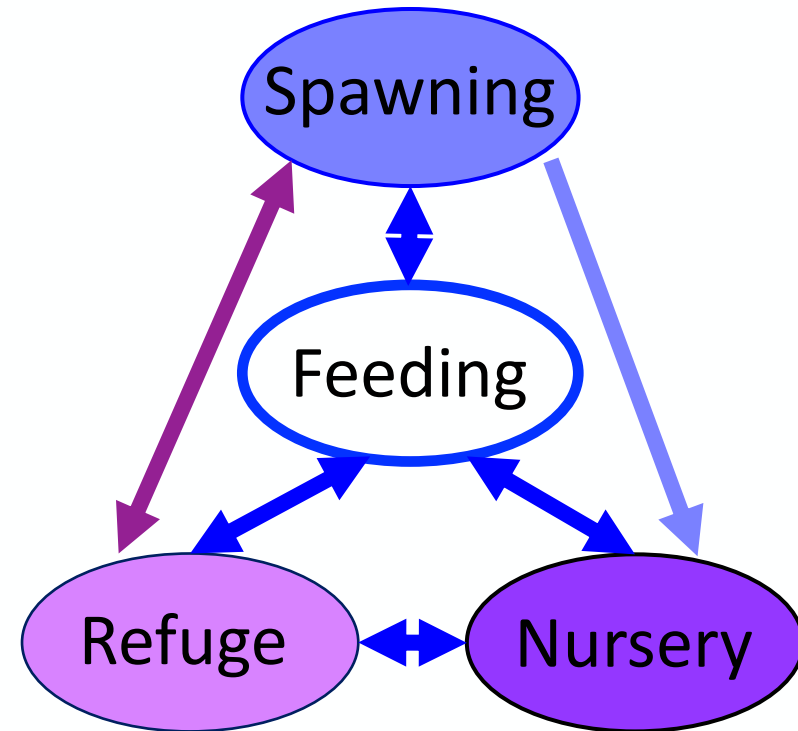
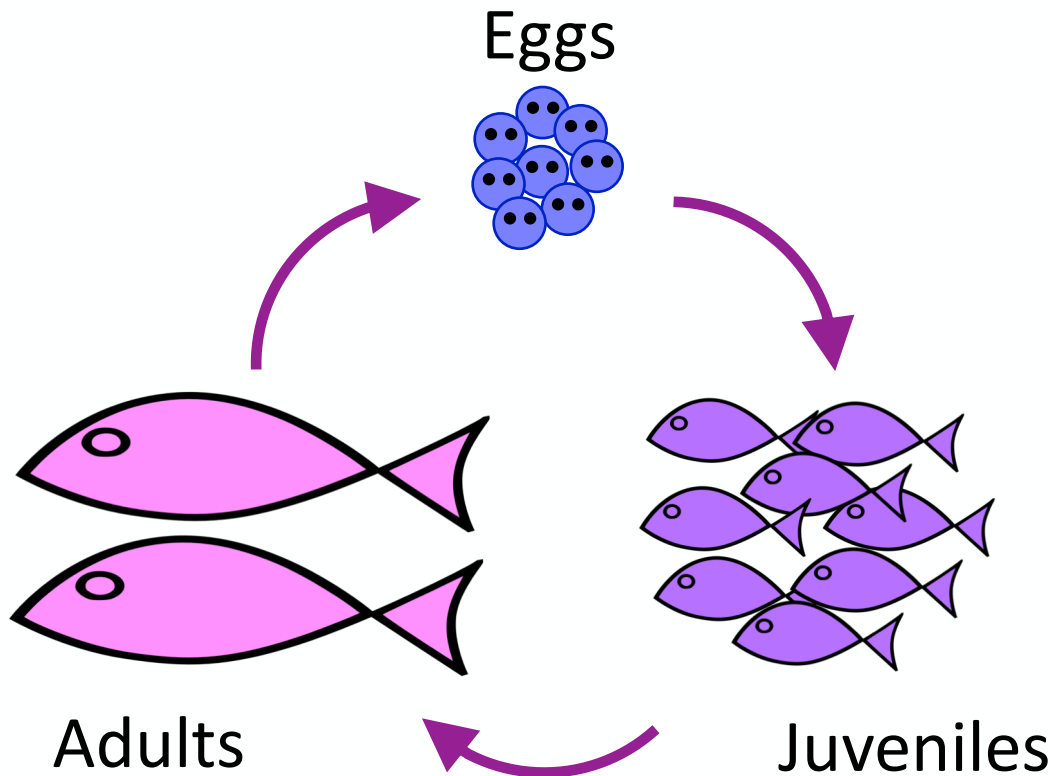


Fish Movements: Why is this Important?

Life History Stages



Different Habitats



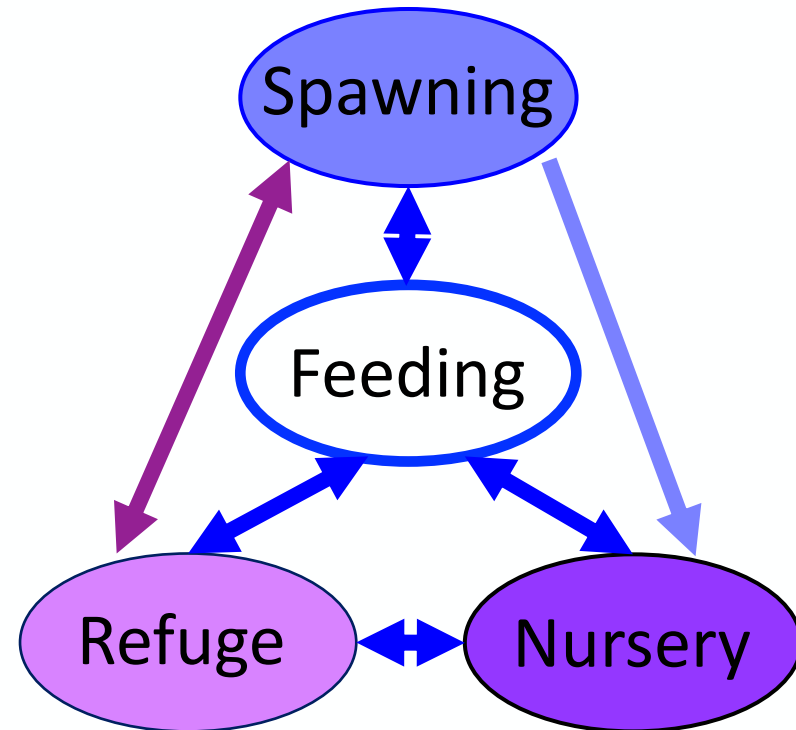
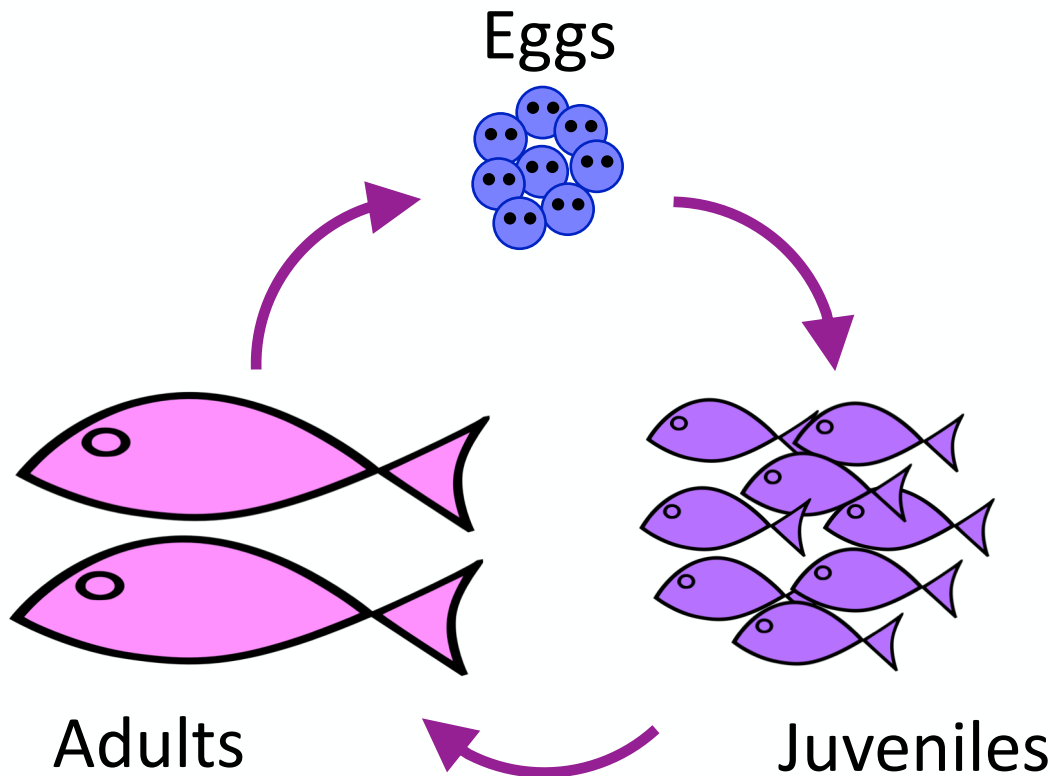
Fish Movements: Why is this Important?

Life cycle requires all habitats + connectivity

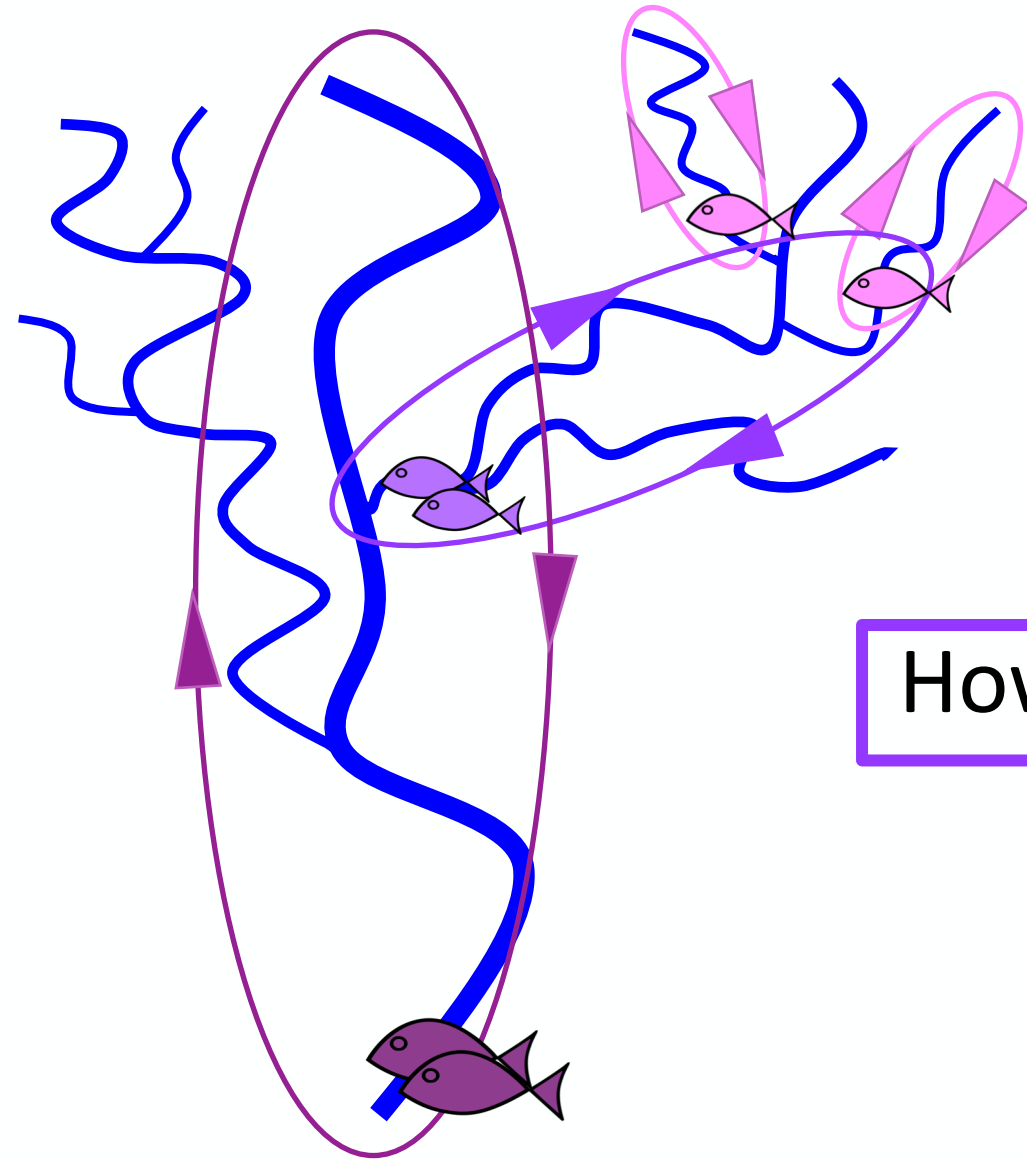
Life History Stages



Different Habitats



Fish Movements: Habitat Connectivity



Need to know
spatial extent

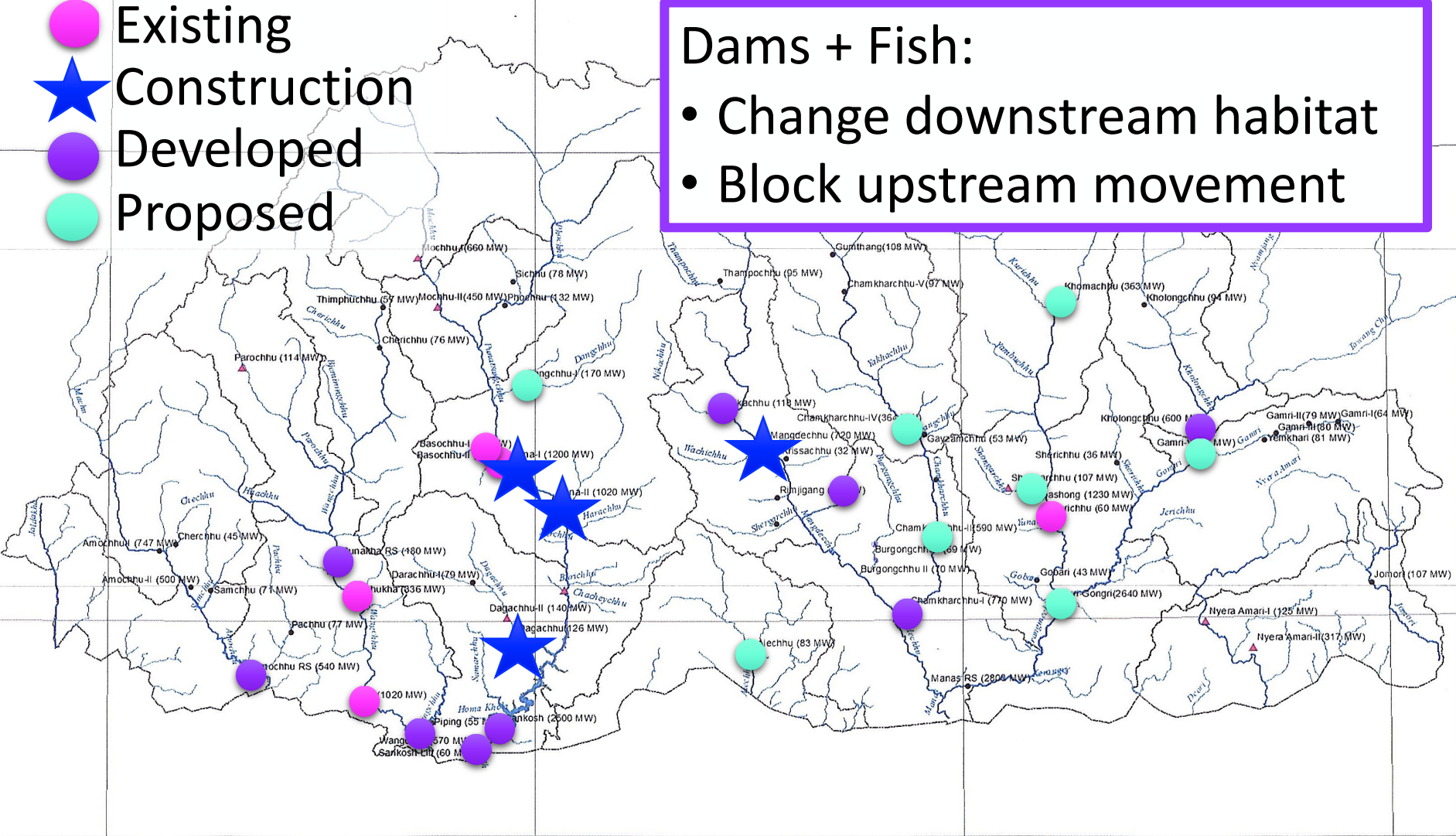
How far do fish move?

Bhutan: Hydropower Potential Sites (> 25 MW)

- Existing
- ★ Construction
- Developed
- Proposed

Dams + Fish:

- Change downstream habitat
- Block upstream movement

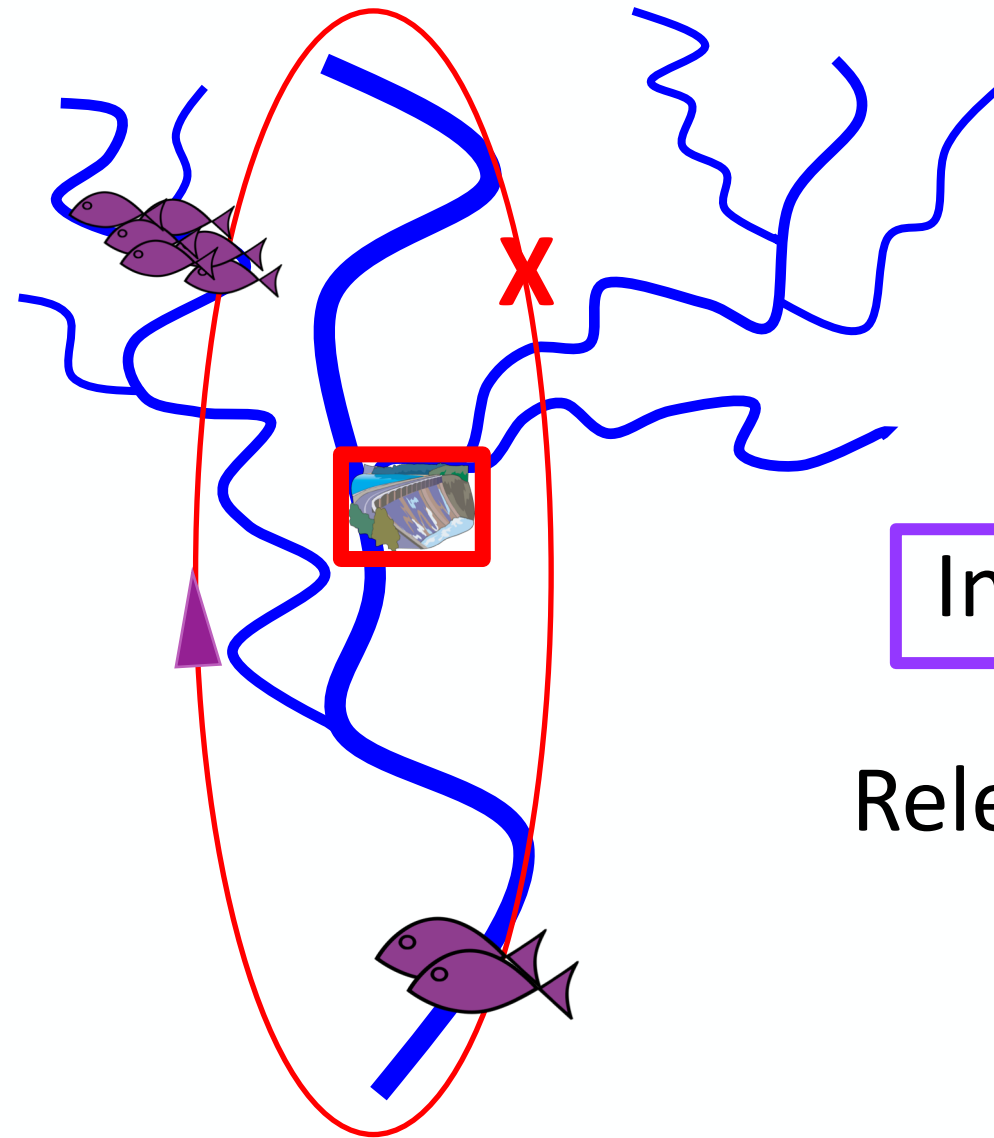


● Existing HPP	▲ Reconnaissance Stage
★ Under Construction HPP	● Balance HPP Site
● DPR Stage	— River
○ PFR Stage	

1:600,000
Kilometers

Data Source:
HPP location from PSMP 2003 & Administrative Map of Bhutan from NLCS

Fish Movements: Habitat Connectivity



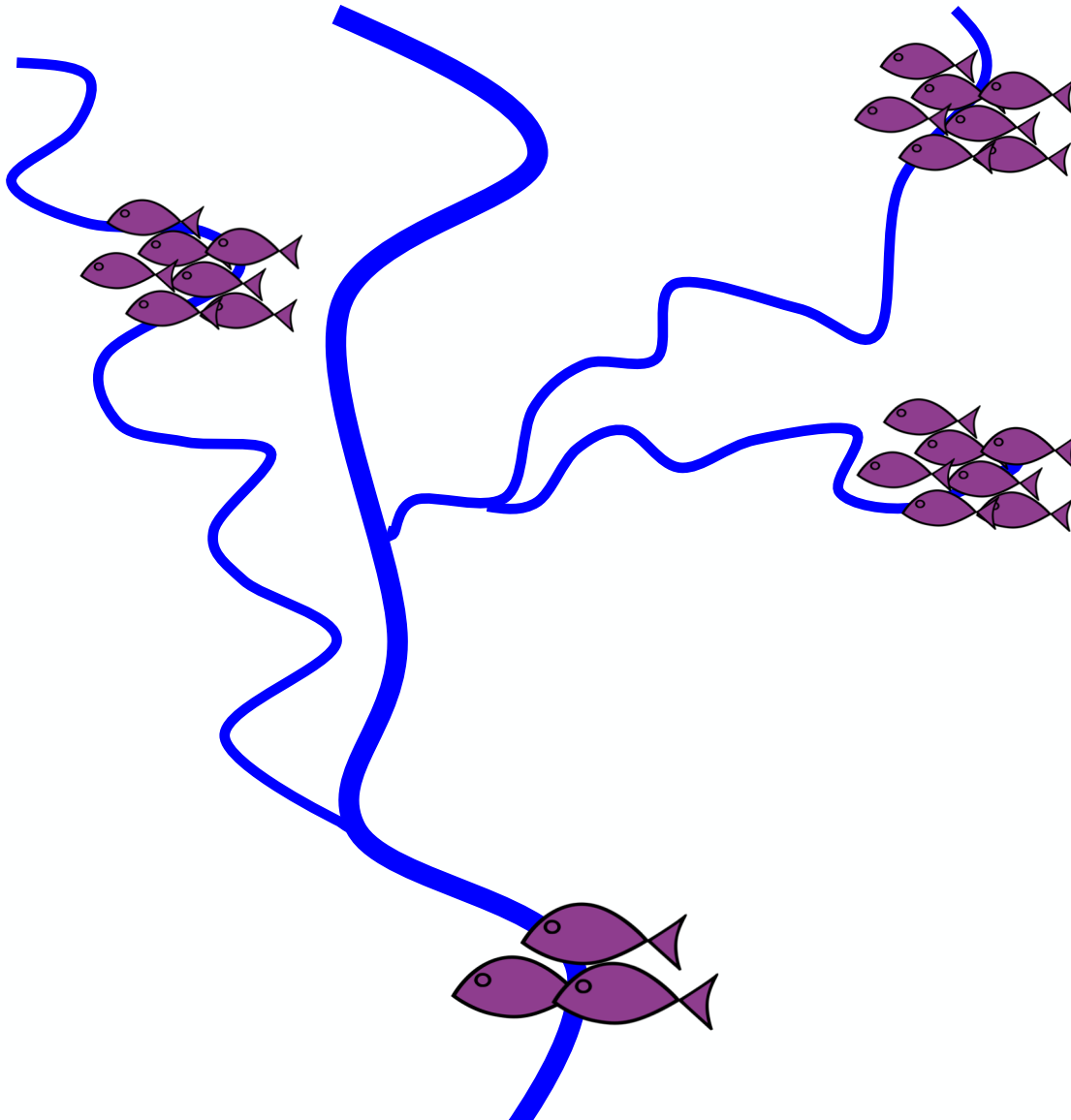
Need to know
spatial extent



Inform Policy Decisions

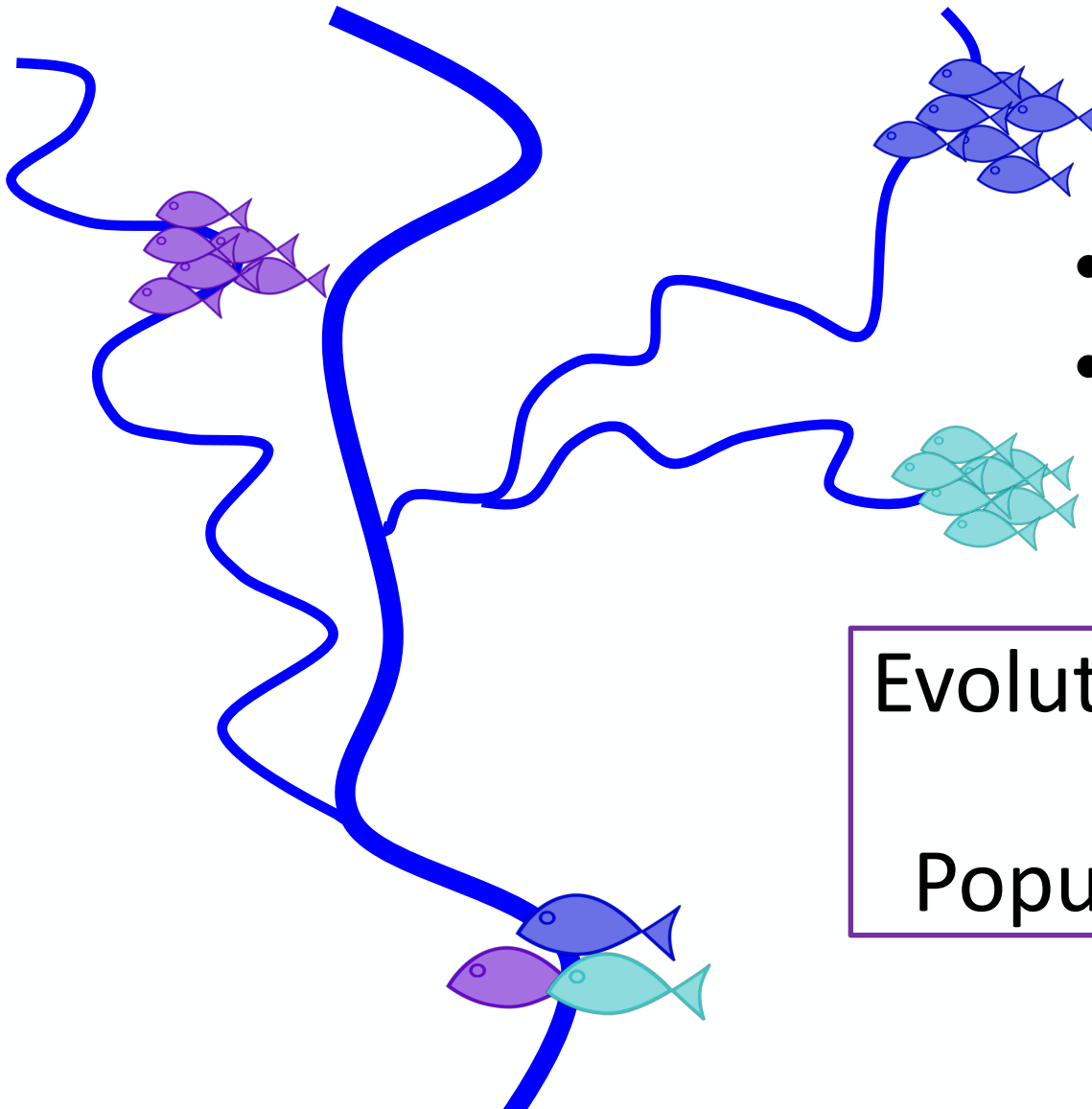
Relevant for conservation
+ management

Mahseer Movements: Site fidelity



Philopatry
Homing

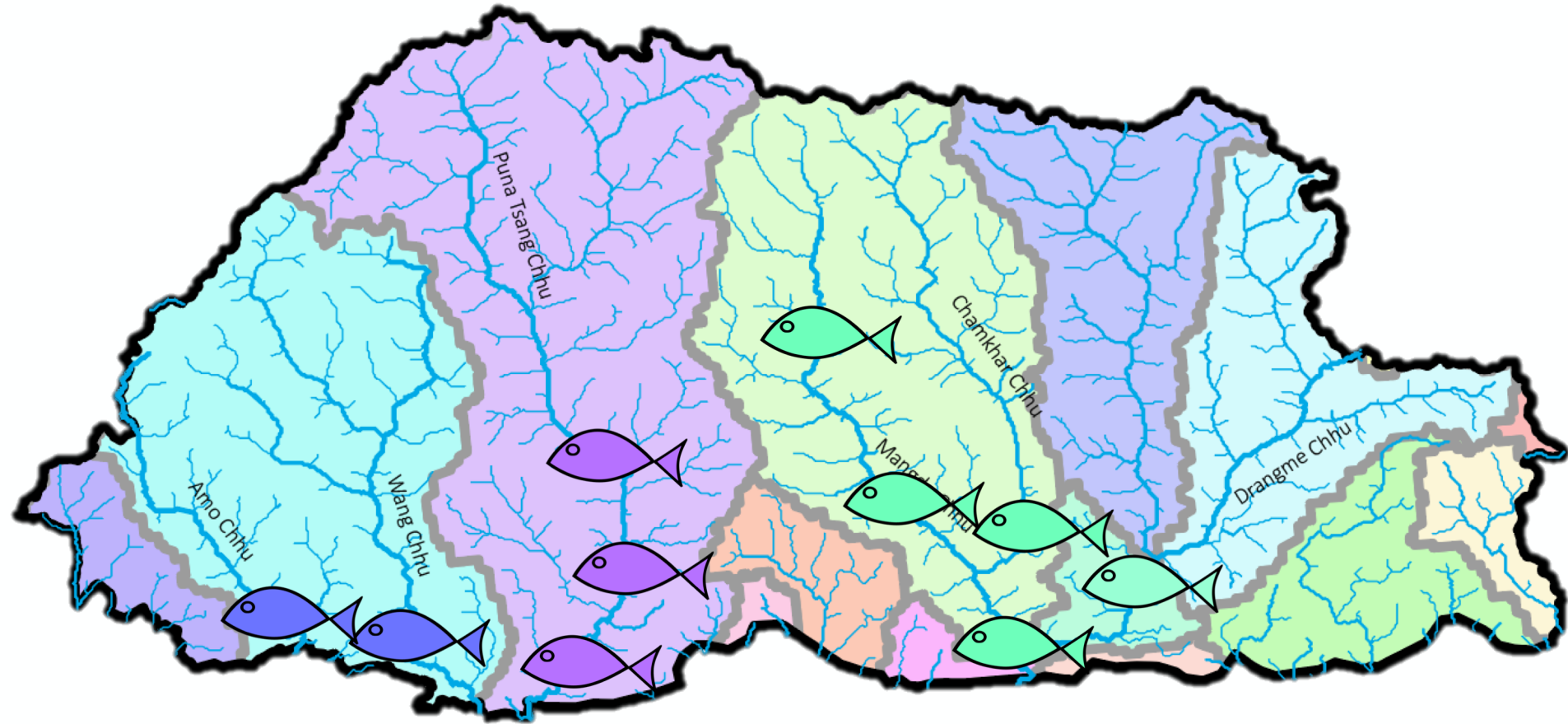
Mahseer Philopatry: Genetic Stocks



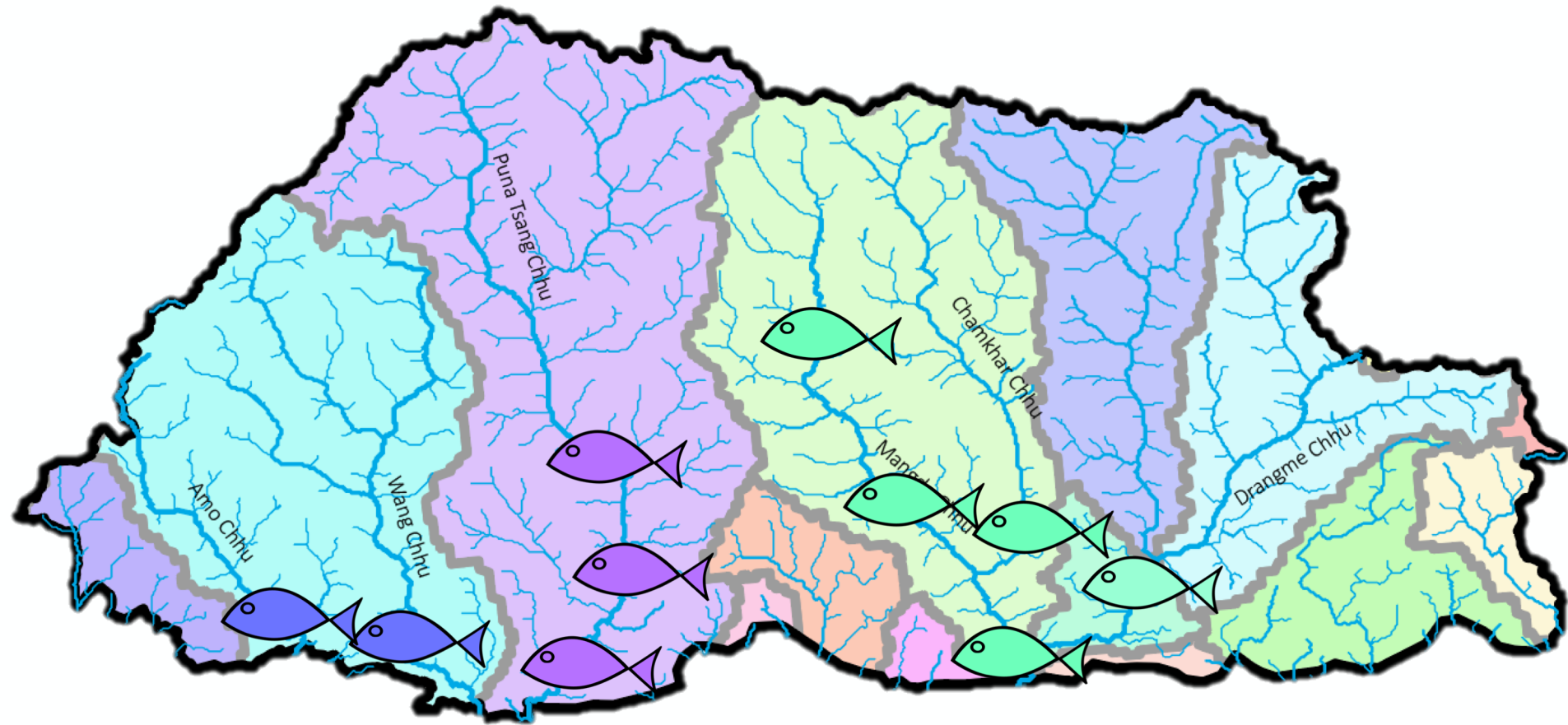
- Genetic drift
- Local adaptation

Evolutionary Processes
↓
Population Structure

Mahseer in Bhutan: Genetic Stocks?

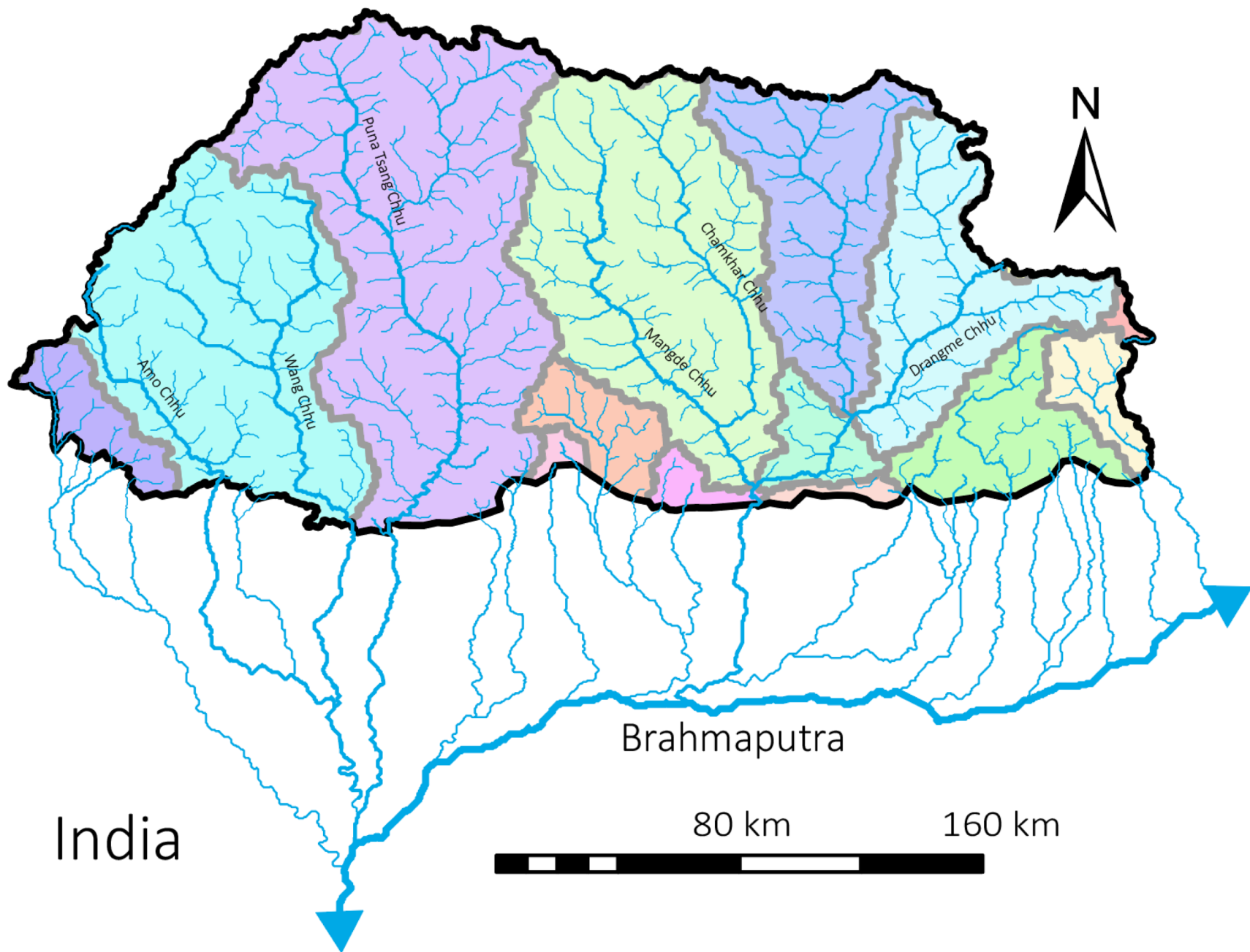


Mahseer in Bhutan: Population Connectivity?



Evolutionary Processes = a very long time

China



India

Brahmaputra

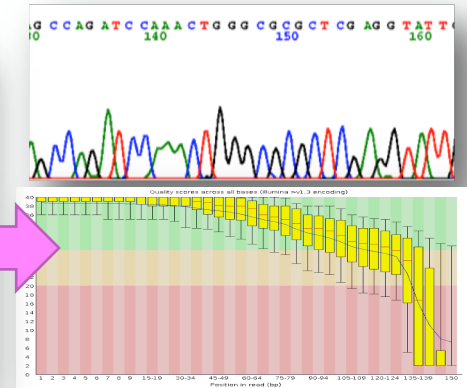
80 km

160 km

Genetic Approaches

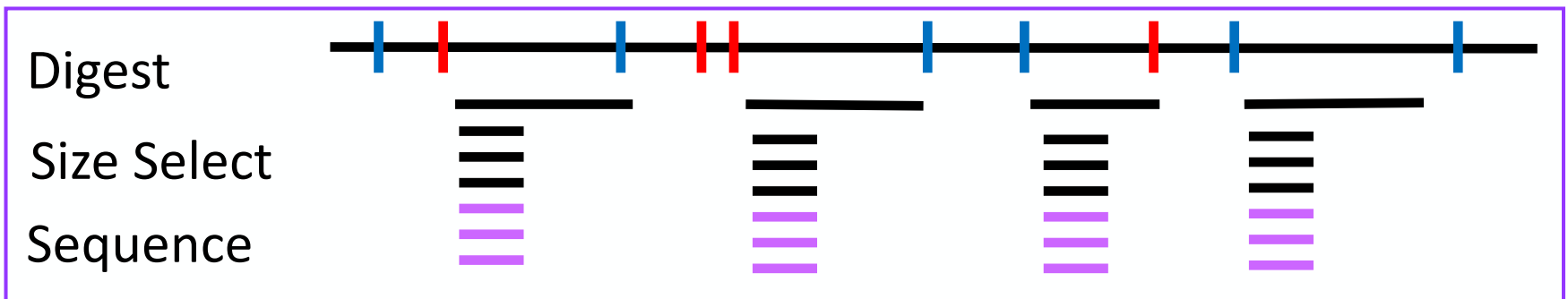


- Sample fish
- Collect fin clips
- Extract DNA from fin clips
- Genotype each sample
- Analyze genetic variation



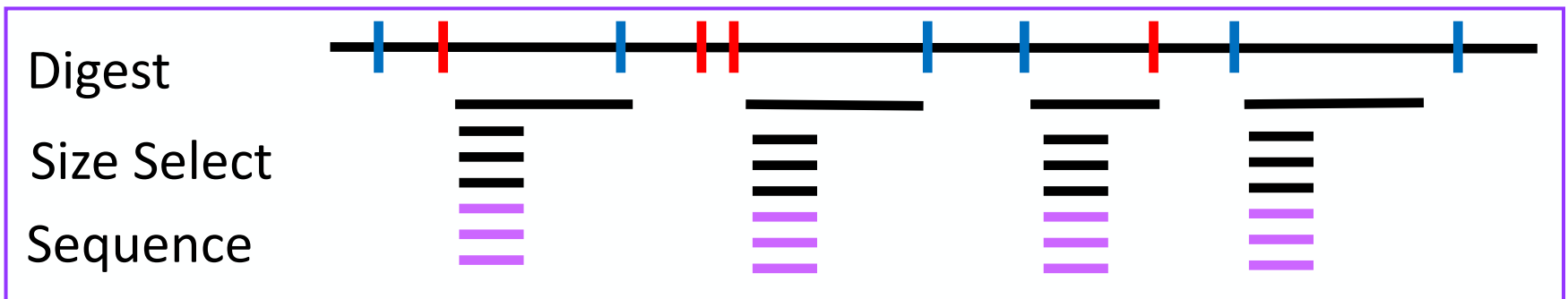
Methods: SNP Genotyping (ddRAD)

- SNPs: Single Nucleotide Polymorphism (variation)
- ddRAD: double-digest Restriction-site Associated DNA
→ reduced genomic representation



Methods: SNP Genotyping (ddRAD)

- Thousands of loci, hundreds of individuals
- Subsampling genome consistently
- Massive amount of data > computationally demanding



Golden Mahseer

Tor putitora

rare



Chocolate Mahseer

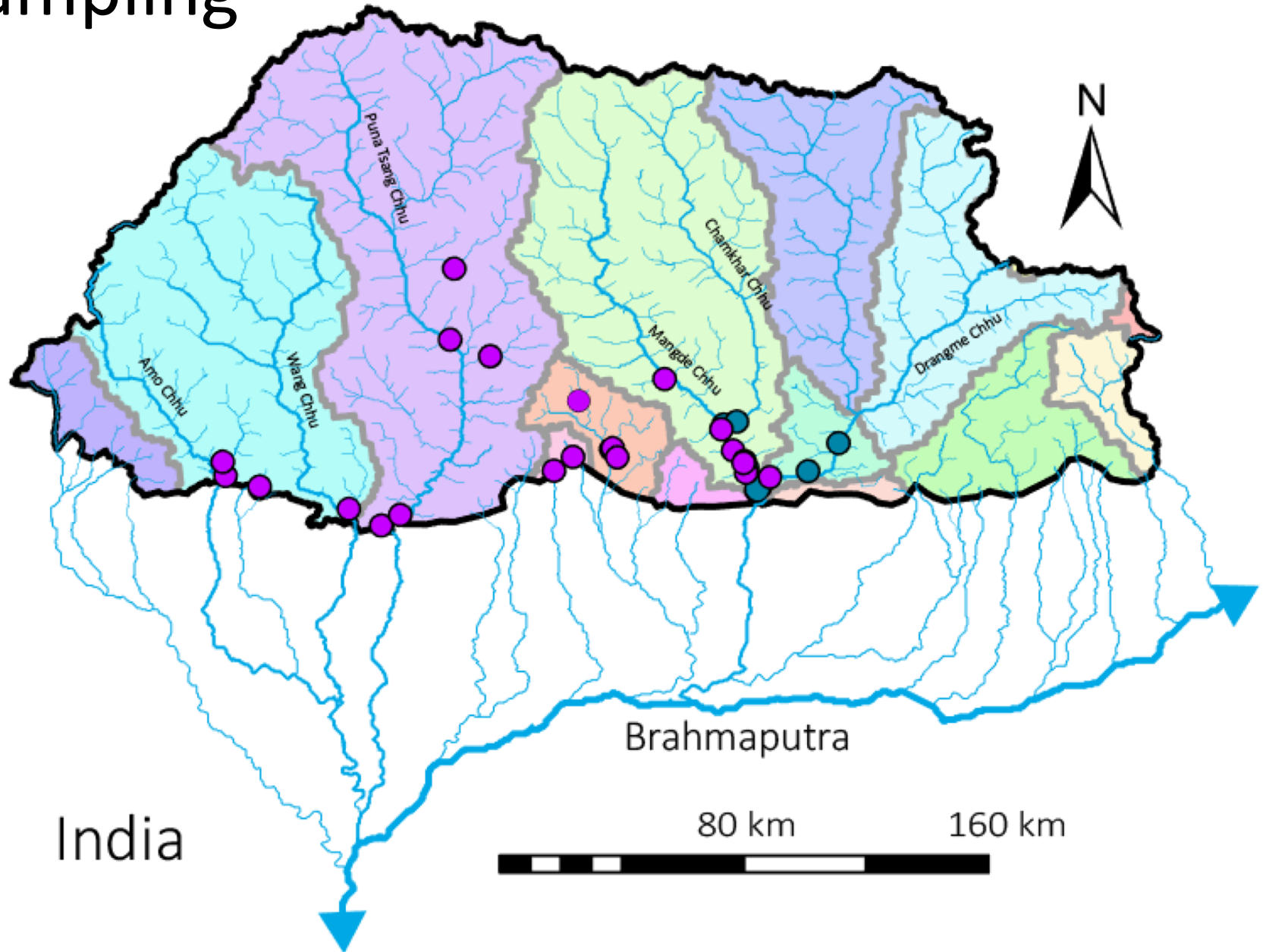
Neolissochilus hexagonolepis

common > surrogate



Sampling

China



India

Brahmaputra

80 km

160 km

Genetic Data

Golden Mahseer

N = 37 individuals

Data: ~22,400 loci

2,241,478 total nucleotides

123,662 polymorphic

80,114 parsimony informative

Single-gene Sanger sequencing

- 1-4 loci
- 1,000-6,000 nucleotides

Chocolate Mahseer

N = 144 individuals

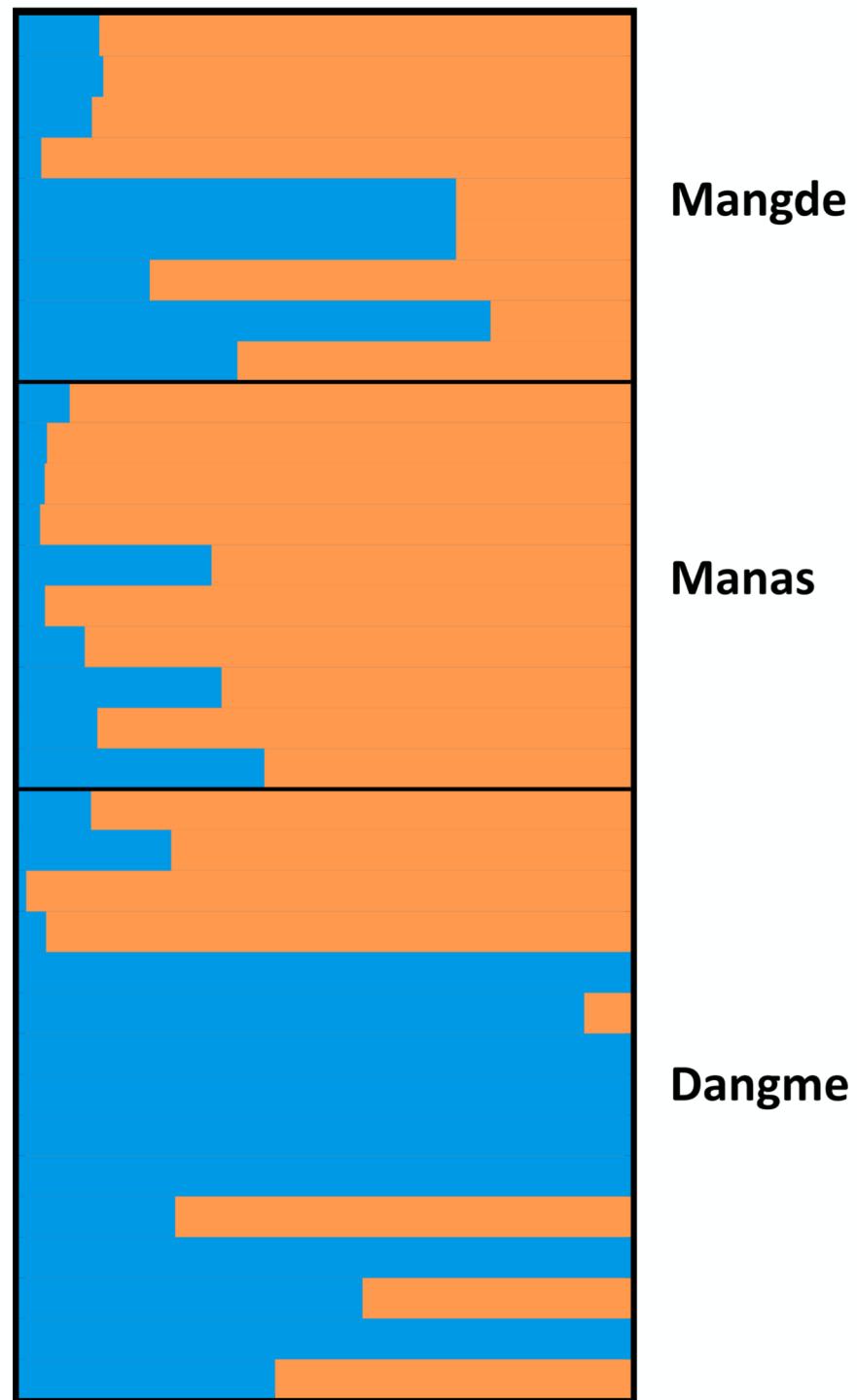
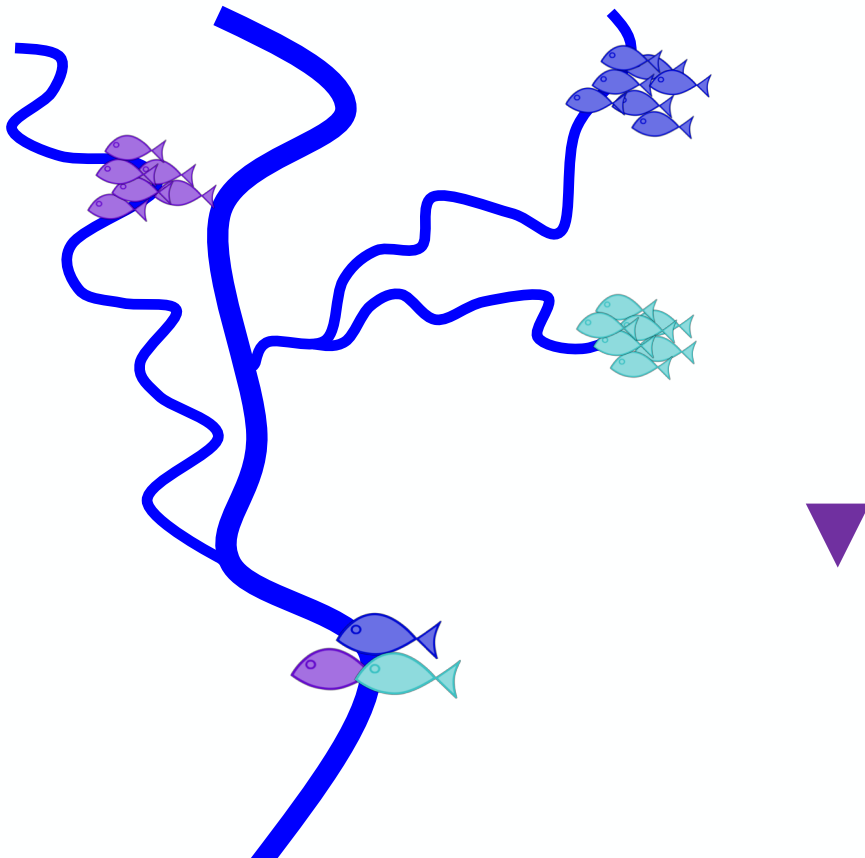
A lot of data

Variation among individuals
requires large sample sizes
to statistically infer patterns

How many genetically distinct populations?

= Gene pools

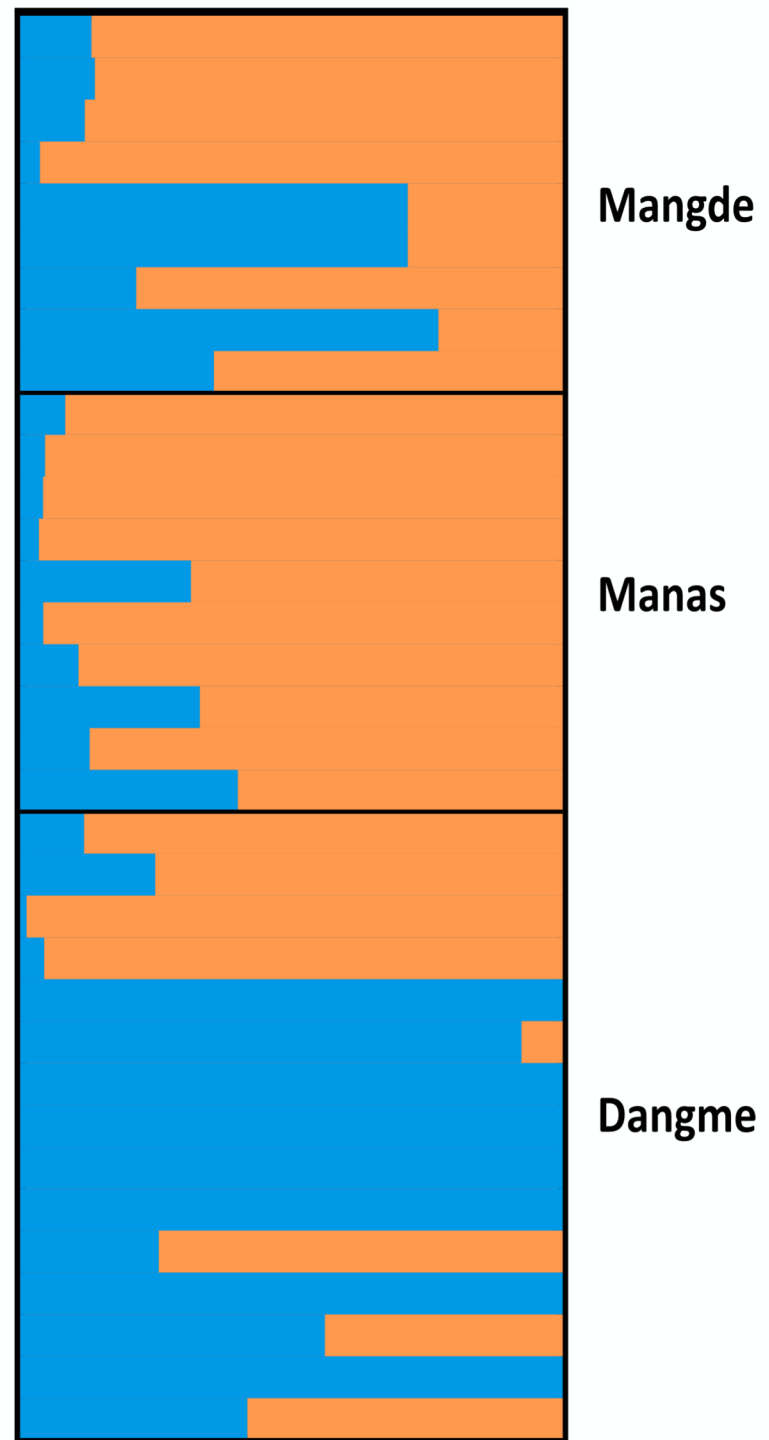
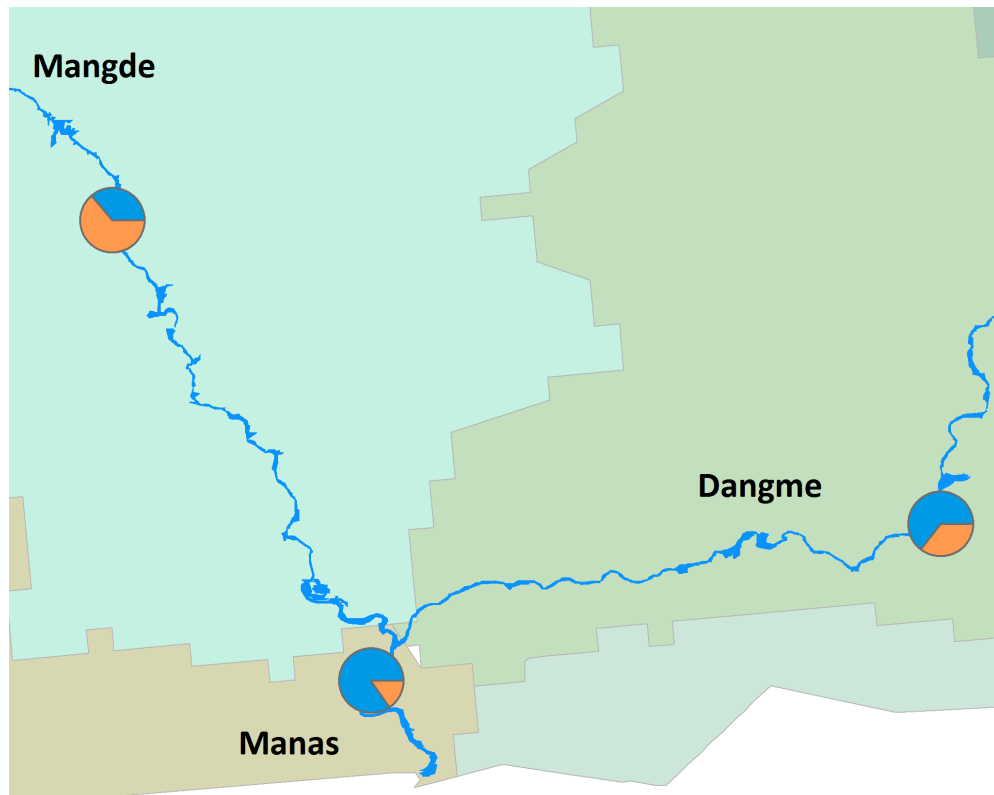
Bayesian Assignment Test



Golden Mahseer

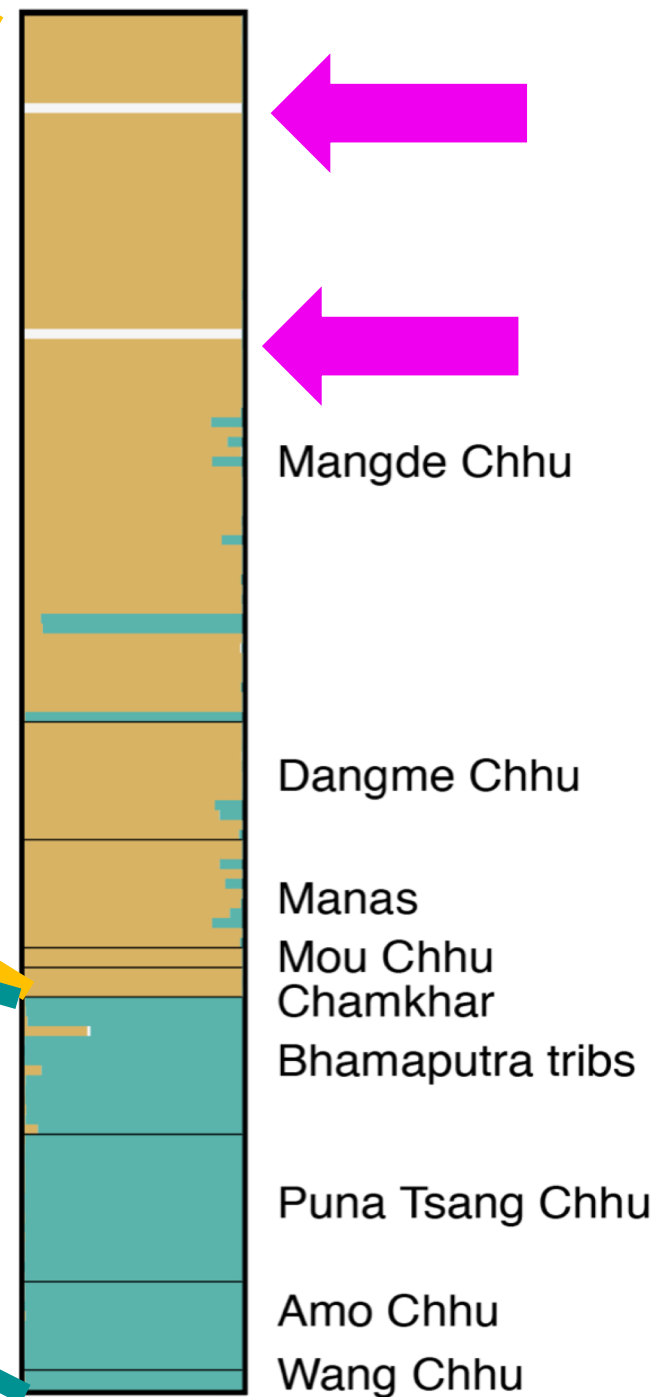
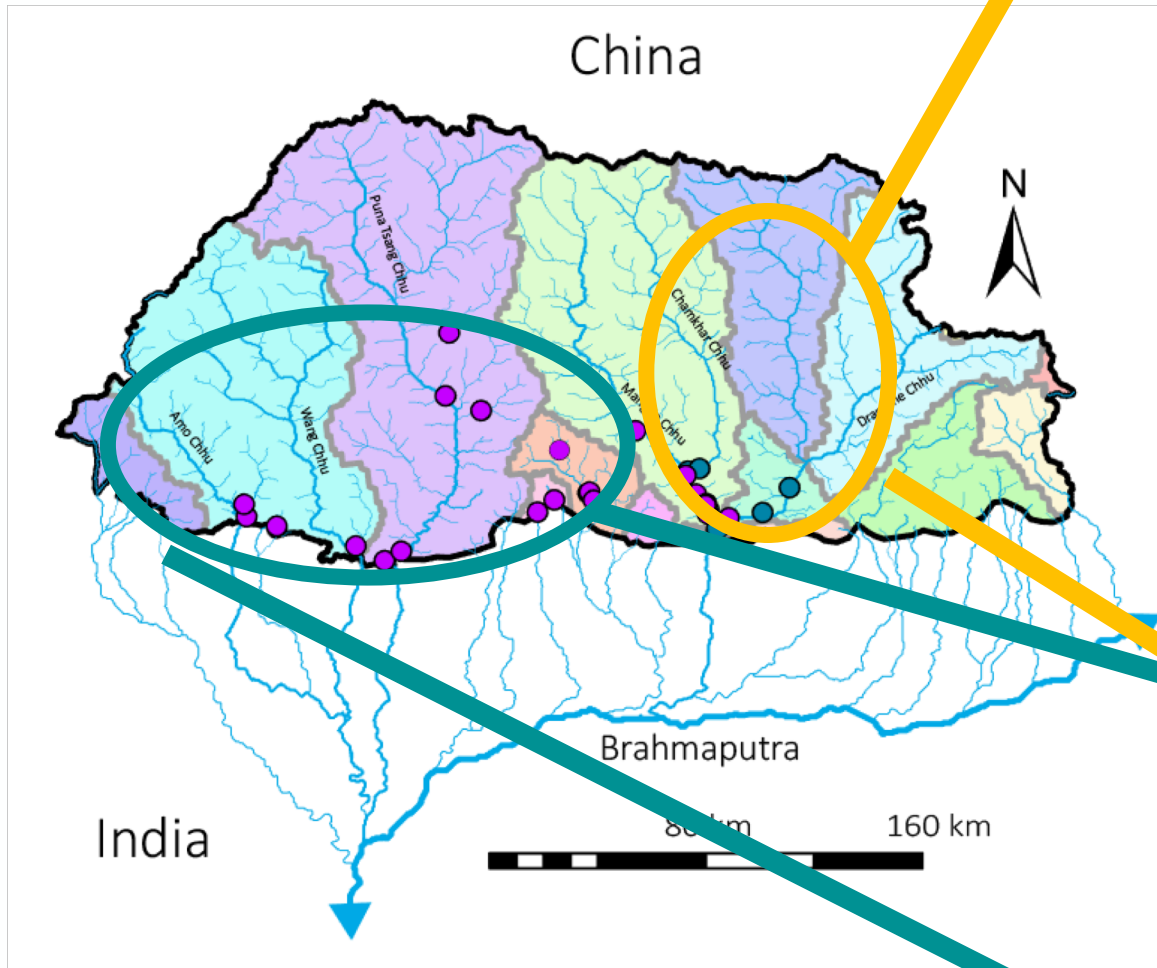
= 2 populations

- related individuals
- ~1st cousin to half-sibling relationship



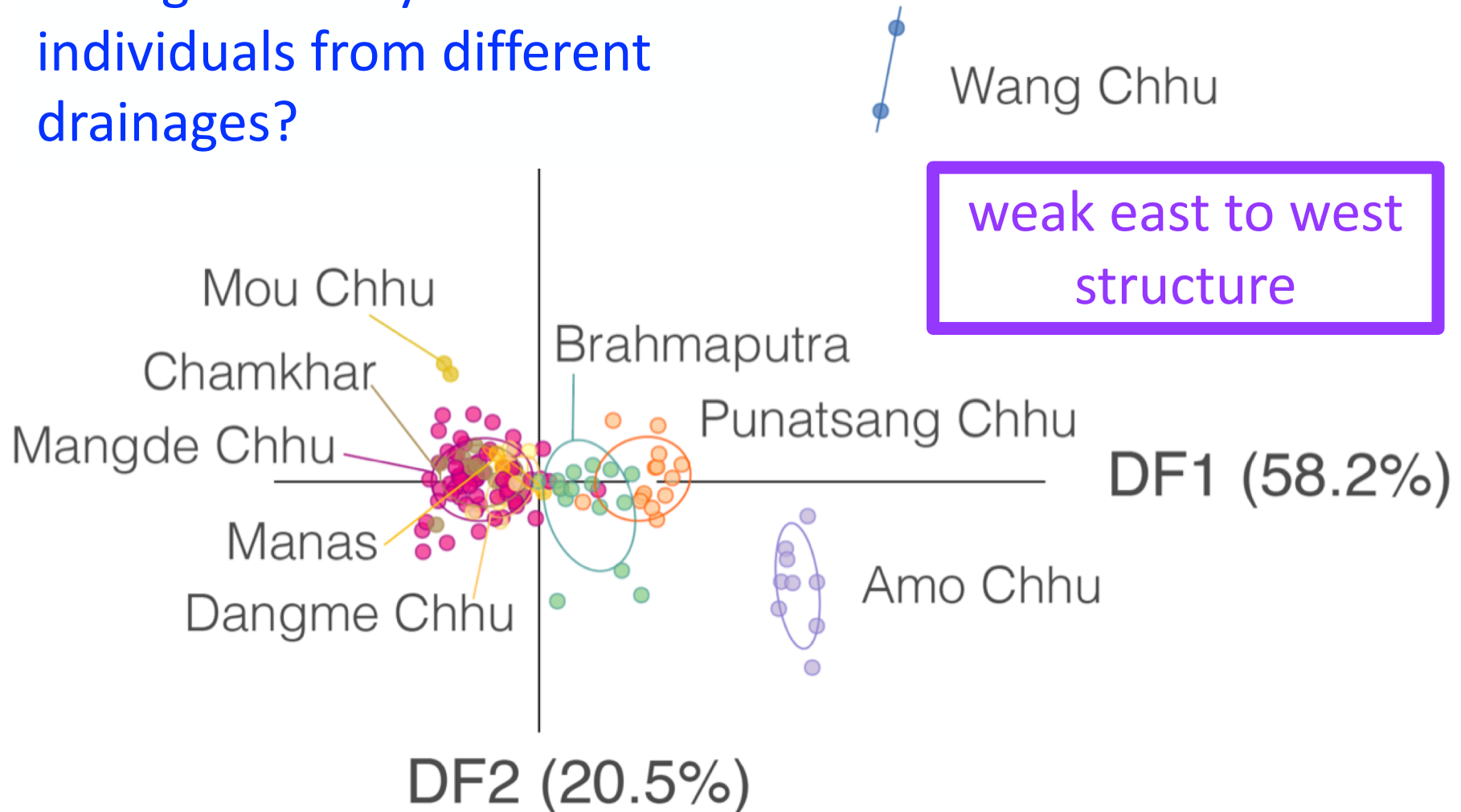
Chocolate Mahseer

= 3 populations



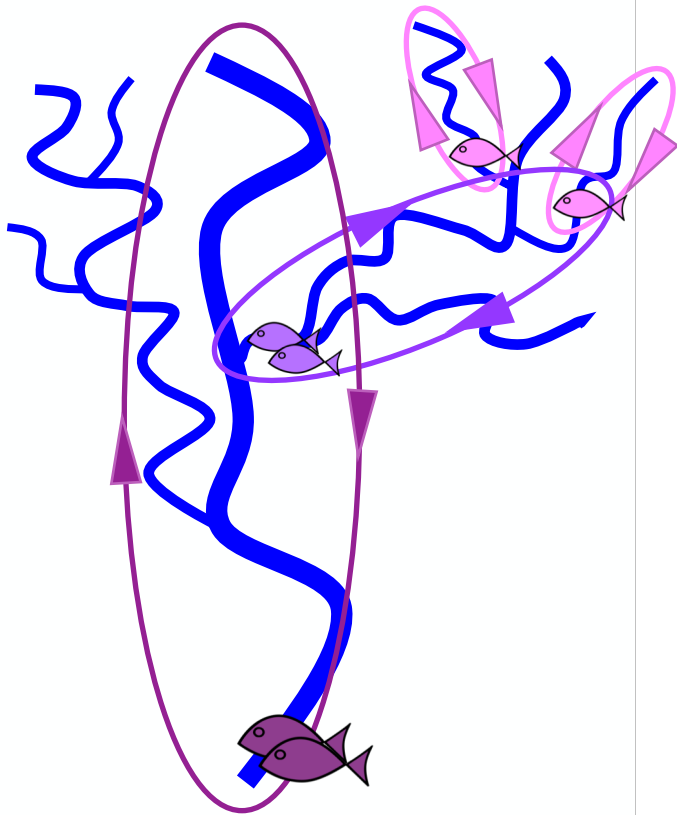
Chocolate Mahseer: DAPC

How genetically distinct are individuals from different drainages?



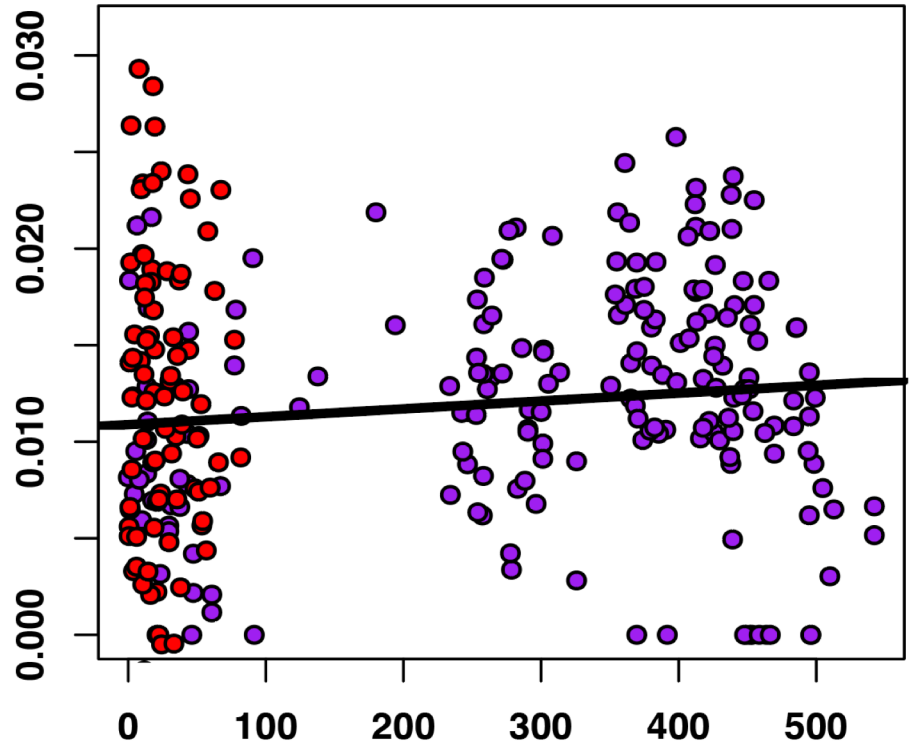
Chocolate Mahseer: Isolation by Distance

Does geographic distance influence connectivity?



Linearized Jost's D ($D/1-D$)

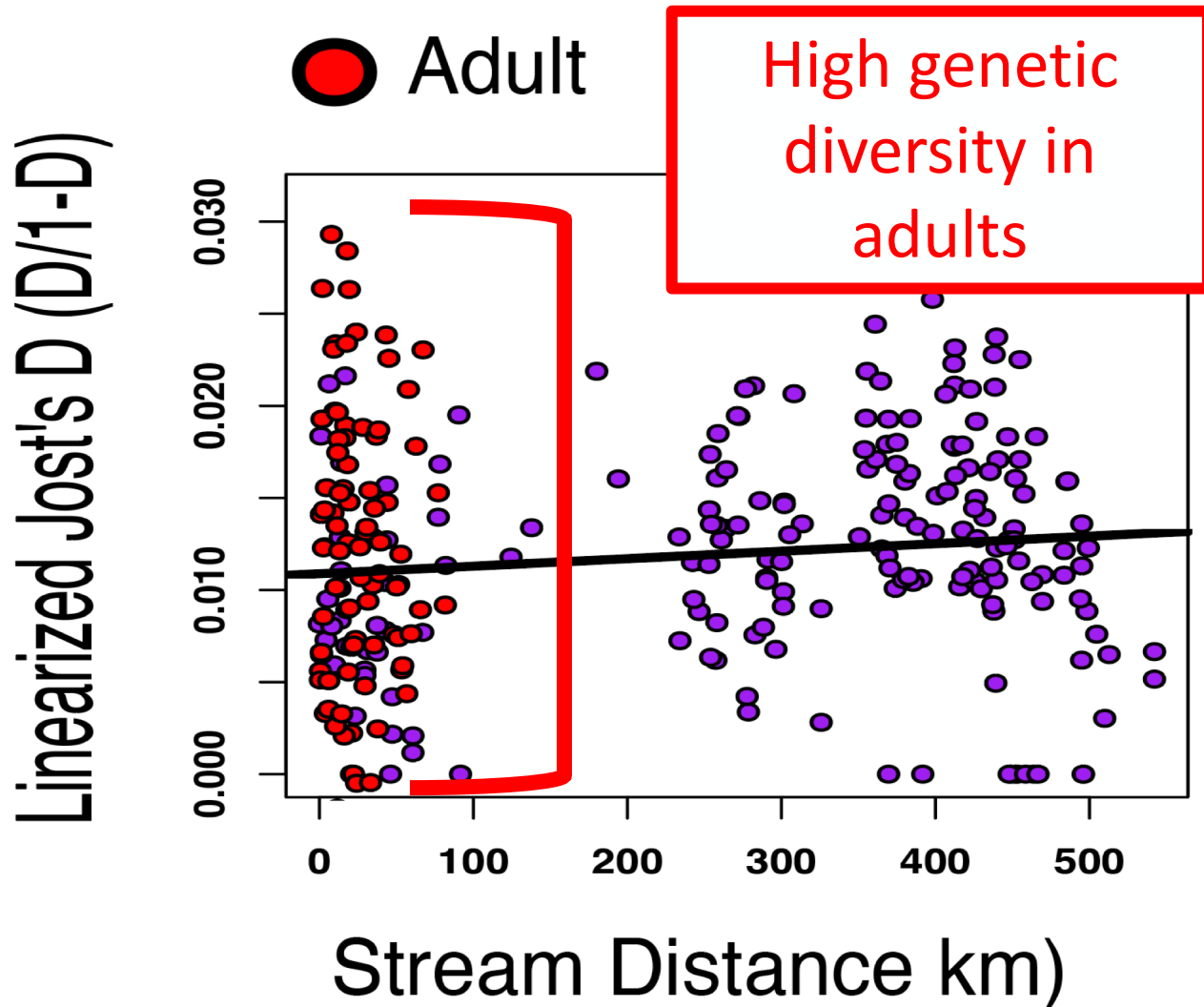
● Adult ● Juveniles



Stream Distance km)

Chocolate Mahseer: Isolation by Distance

Does geographic distance influence connectivity?

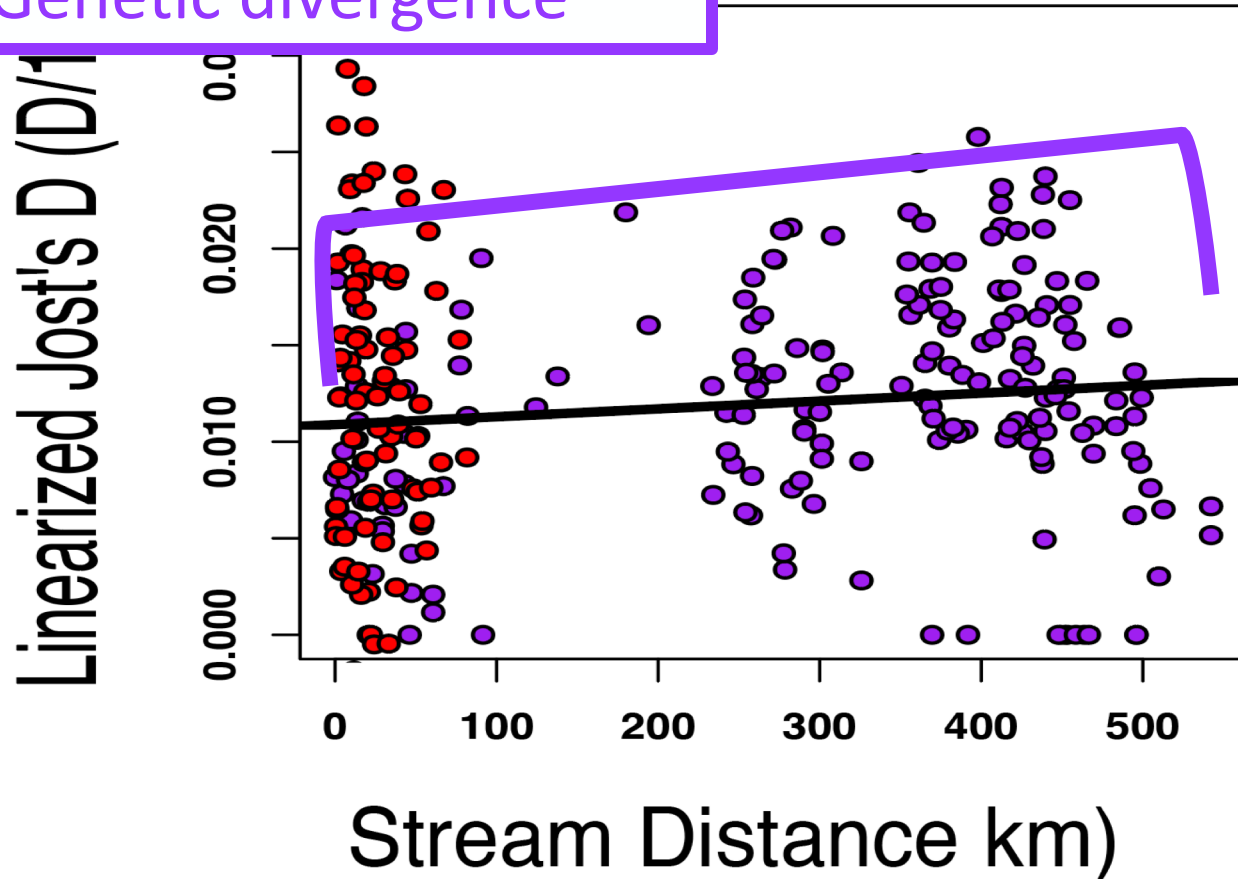


Chocolate Mahseer: Isolation by Distance

Does geographic distance influence connectivity?

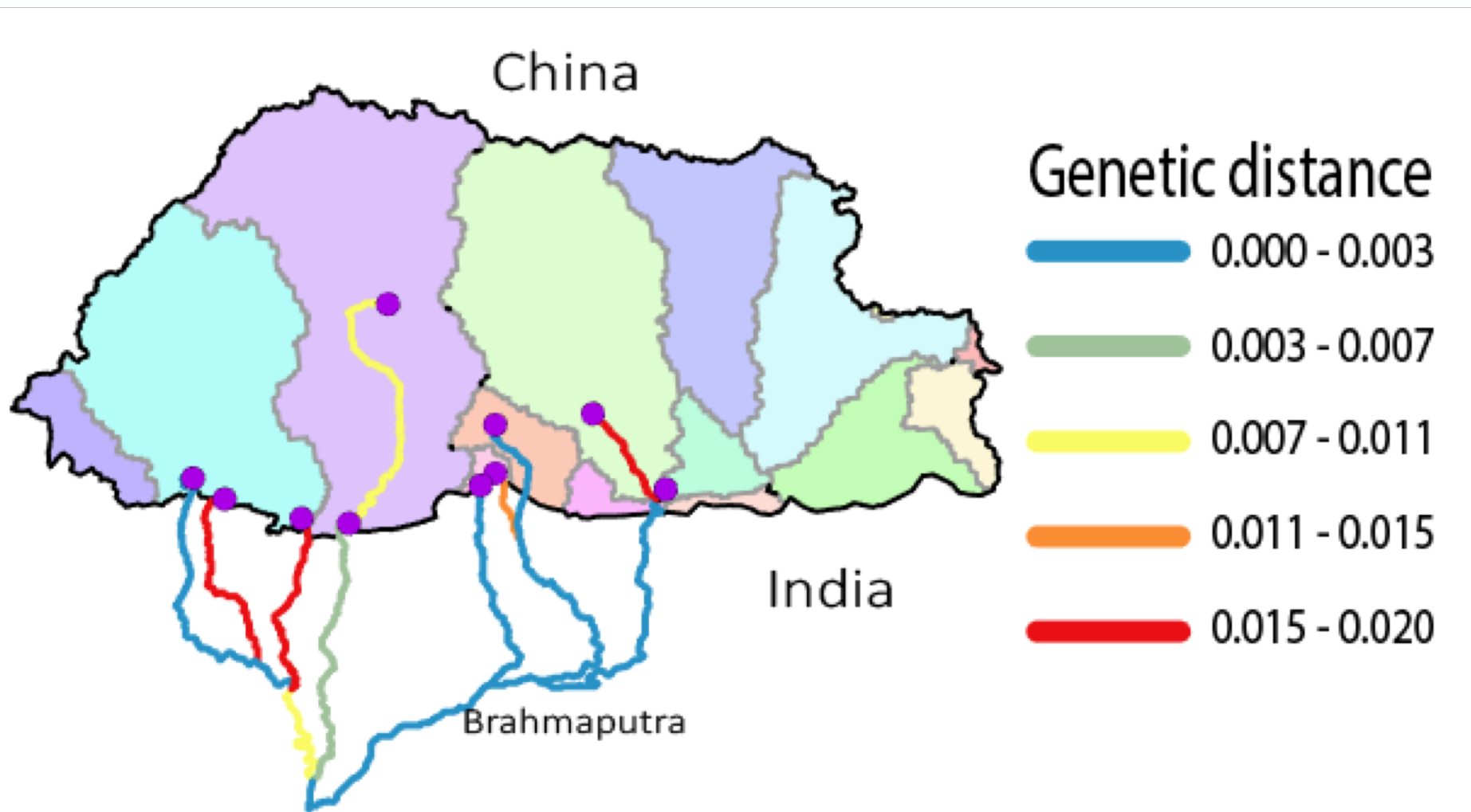
Geographic distance =
Genetic divergence

● Juveniles



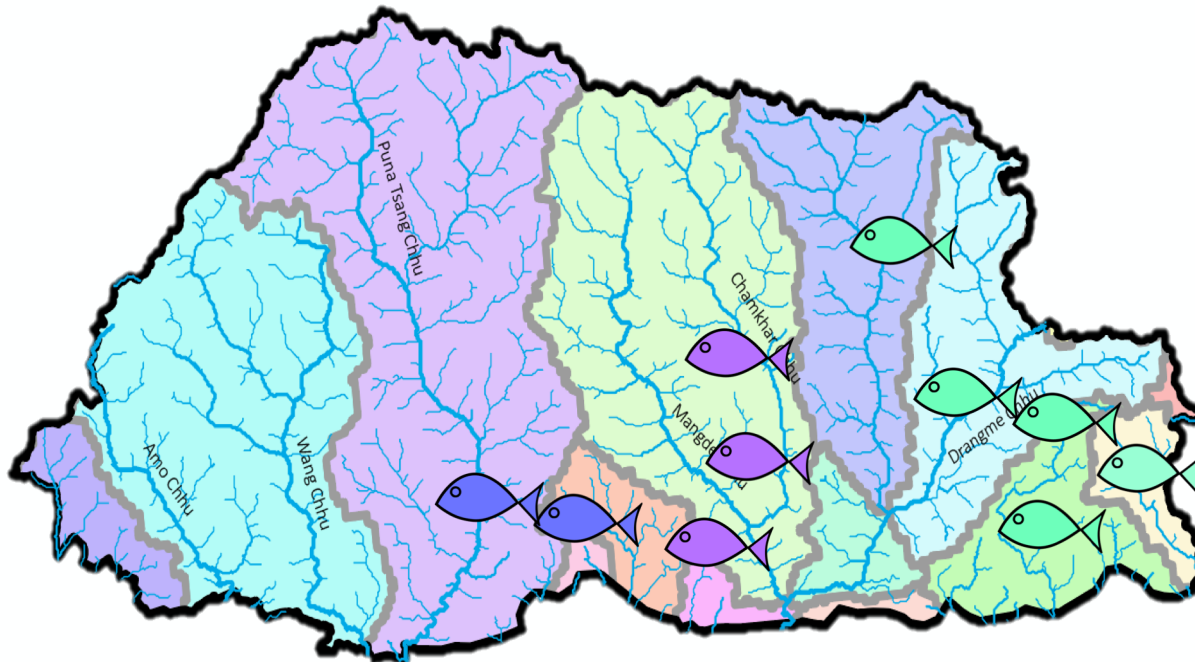
Chocolate Mahseer: StreamTree Analysis

Do landscape features influence population connectivity?



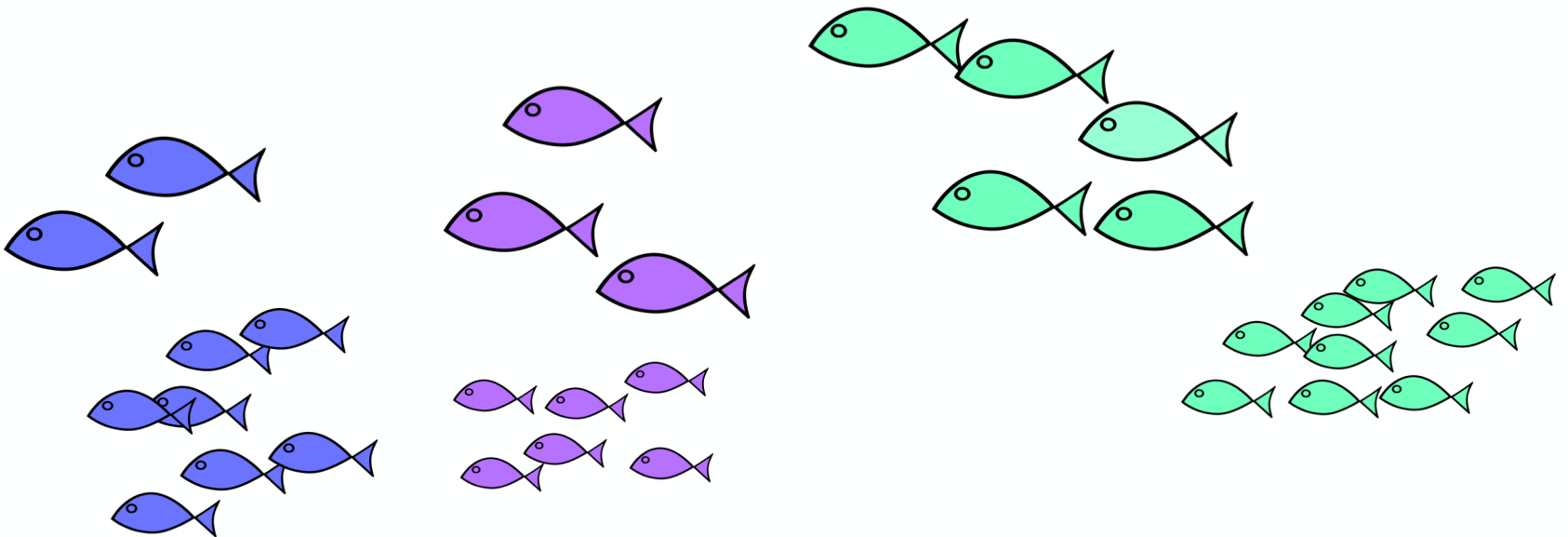
Conclusions: Preliminary

- Key Findings
 - Genetically distinct populations
 - Distinct genetic stocks in each basin
 - Riverscape determines population connectivity



Conclusions: Preliminary

- Future research
 - Expand study across all basins
 - Increase sample size → finer resolution
 - Population genetic parameters
 - Effective population size N_e
 - Effective number of breeders N_b



Thank you!

