TOOLS FOR STUDYING GENETICS

STAC Brook Trout Workshop

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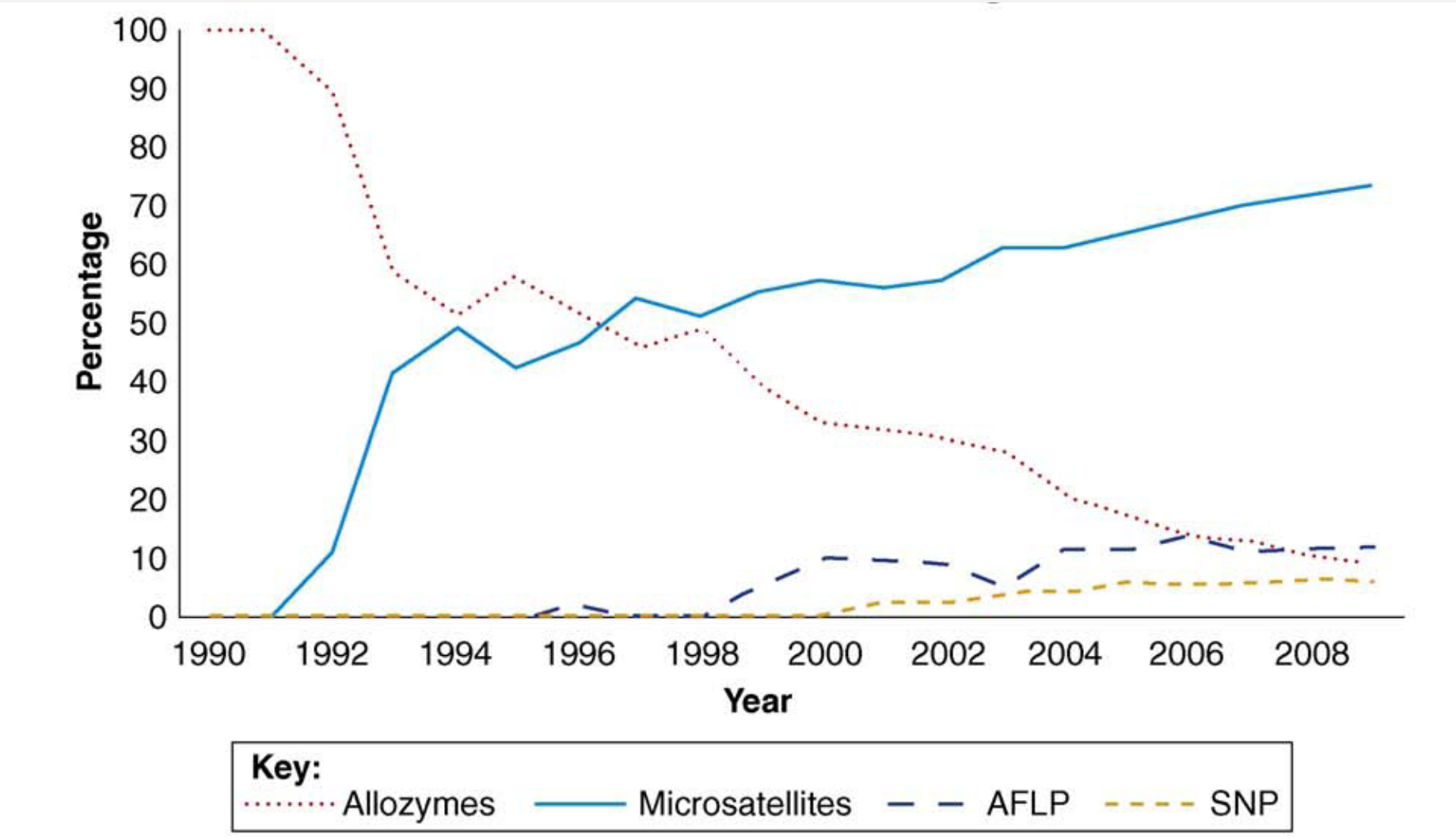
GENETIC MARKERS

USE OF MARKERS IN CONSERVATION GENETICS

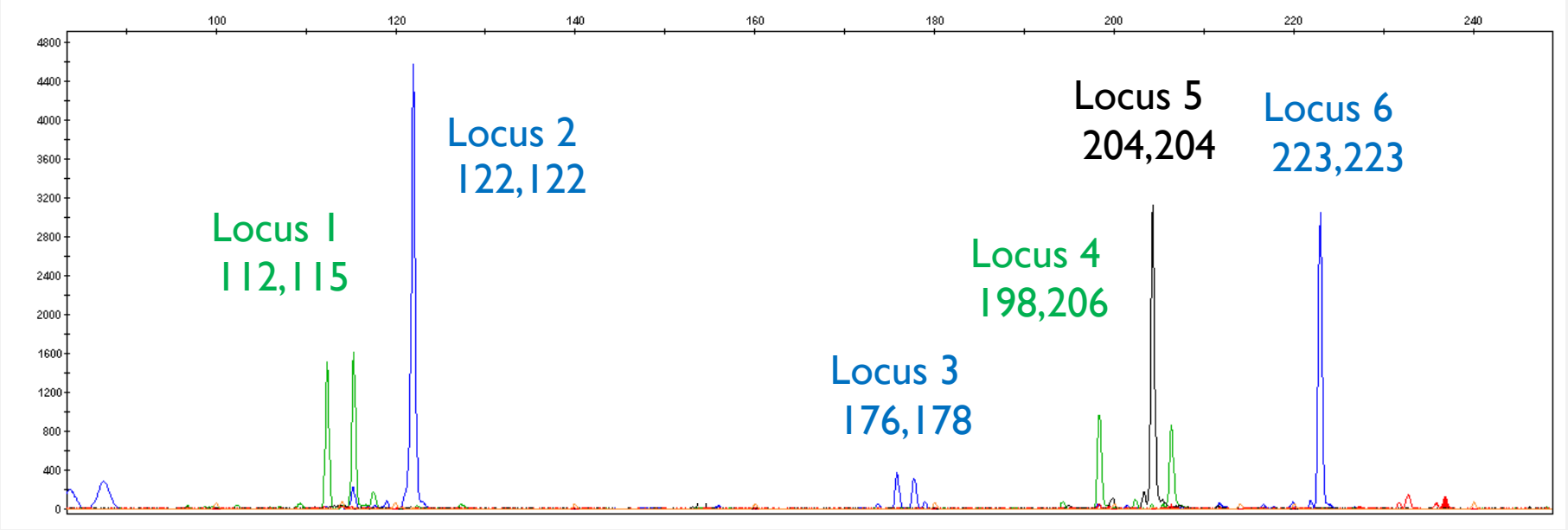


Ouborg et al. (2010)

MICROSATELLITES
Short, tandem repeats

GATAGATAGATAGATAGATA

Alleles: length variants



MICROSATELLITES

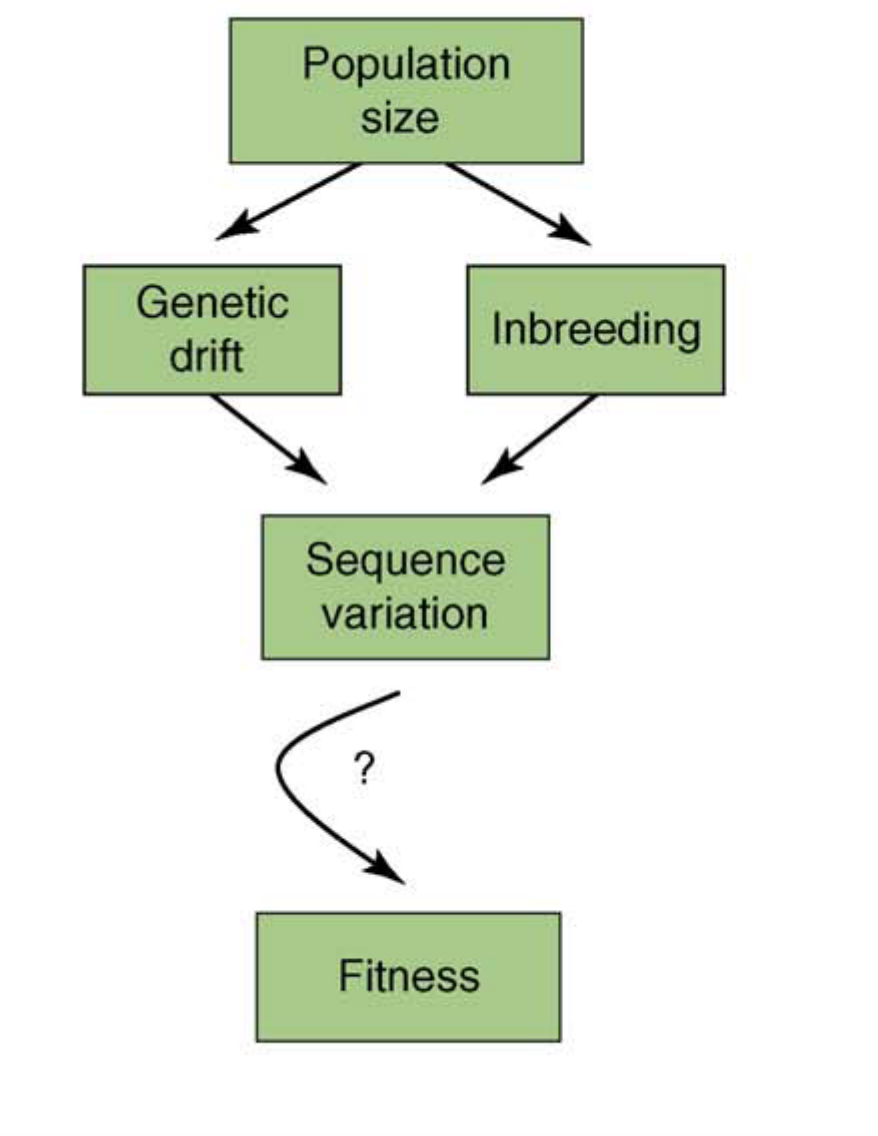
Advantages

- High variability
- Neutral (migration, drift)
- Established baselines

Disadvantages

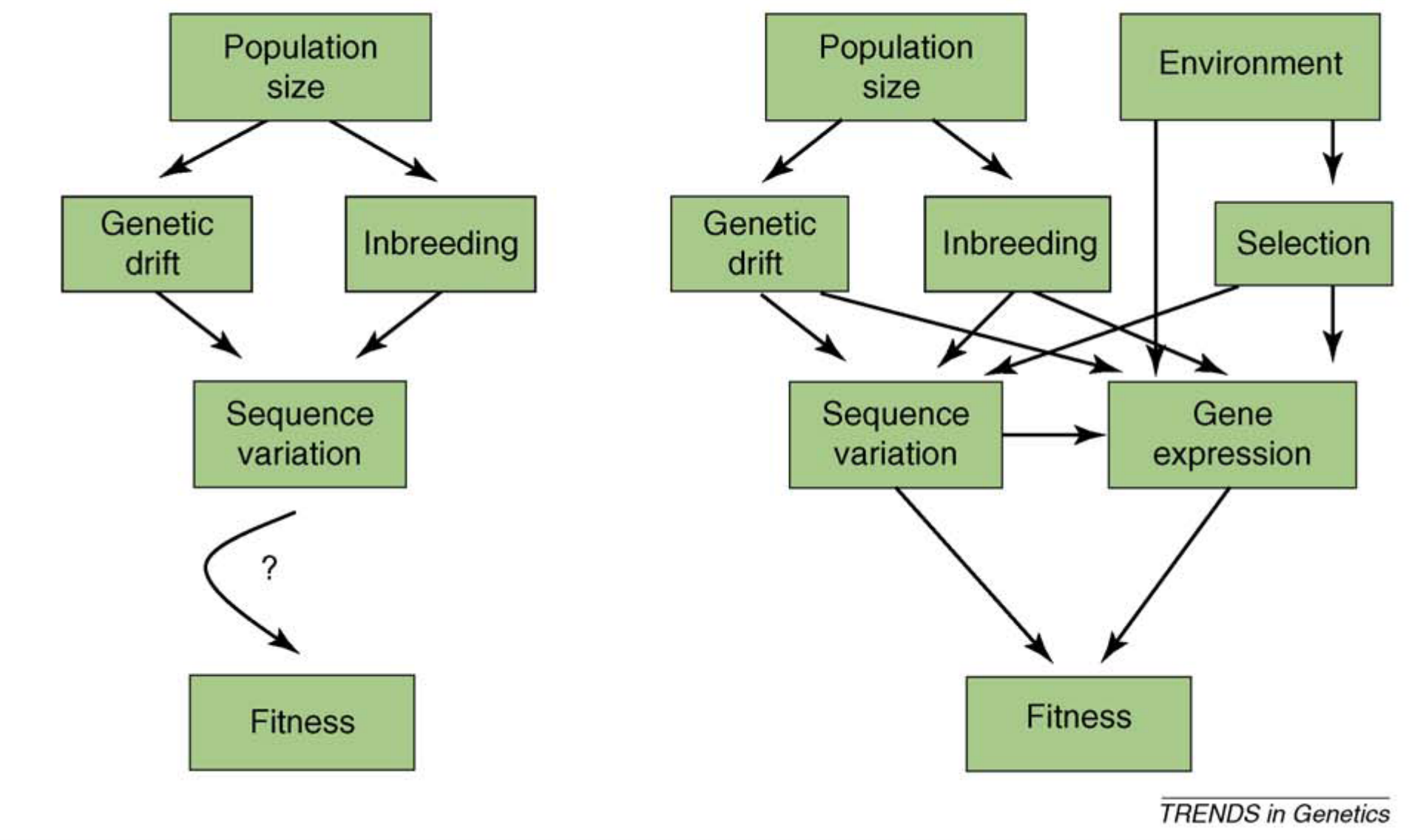
- Neutral (no selection)
- Not representative of whole genome

Conservation Genetics



Conservation Genetics

Conservation Genomics



Ouborg et al. (2010)

SNPS (SINGLE NUCLEOTIDE POLYMORPHISMS)

Advantages

- Neutral & adaptive
- Better representation of genome

Disadvantages

- Need lots of them
- SNP identification question-specific
- Can be hard adding samples to existing project

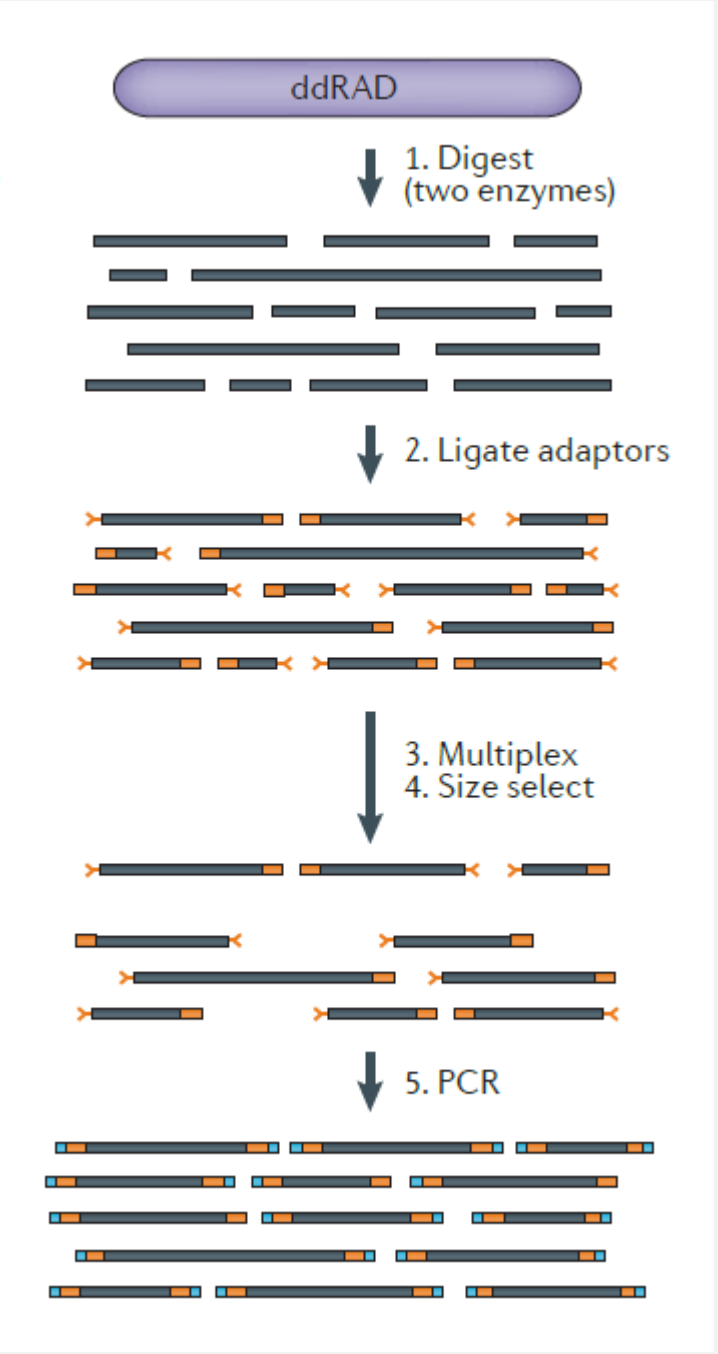
NEXT-GENERATION SEQUENCING

- DNA sequence variation throughout the entire genome
- Identify single nucleotide polymorphisms (SNPs)
- Can find SNPs under selection

Illumina video

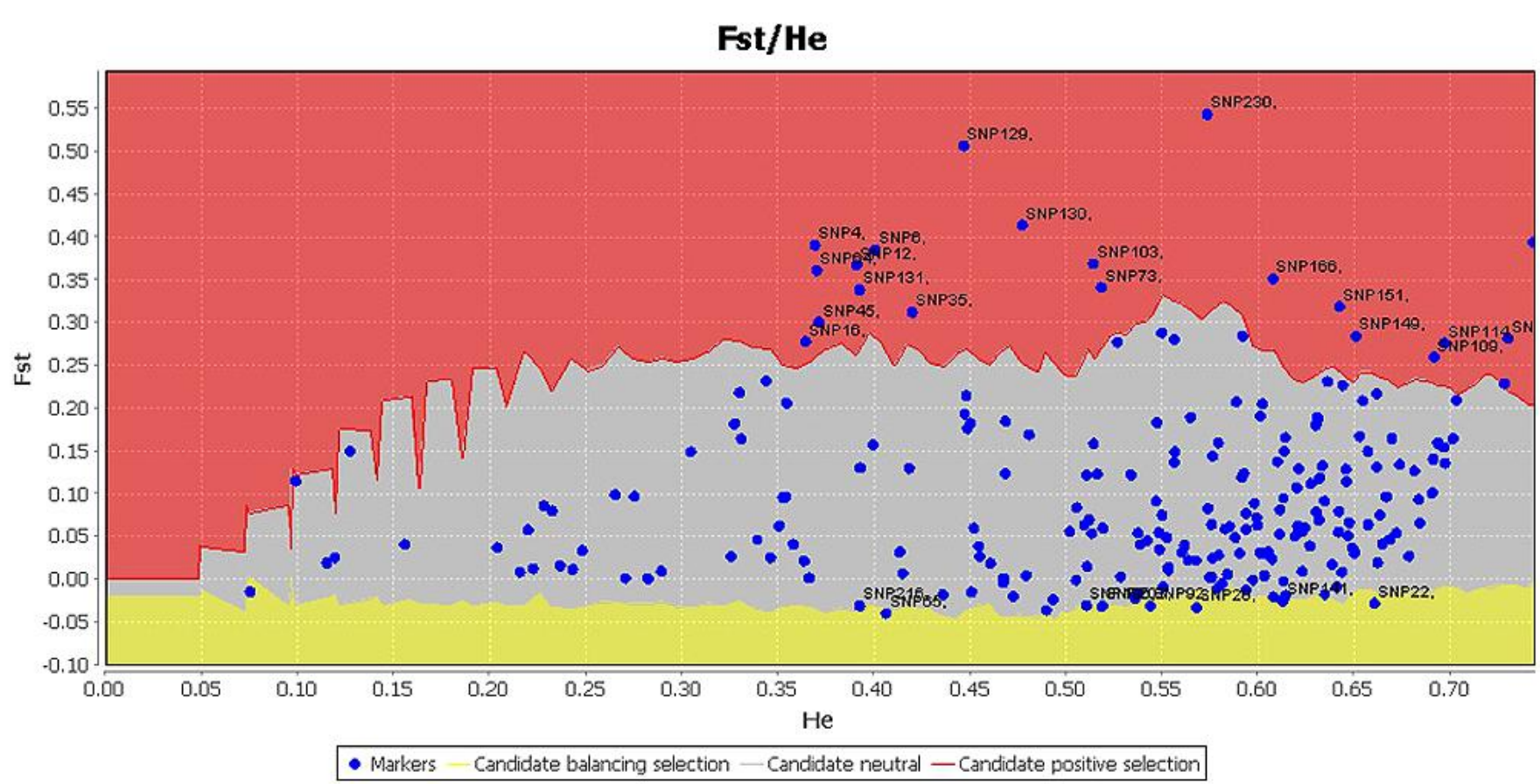
NEXT-GENERATION SEQUENCING

- RADseq (and variations)
  - reduce genome complexity using restriction enzymes
- Considerations
  - PCR based – can use extracts from previous genetic studies
  - Limited labwork – heavy on data analysis
  - Need sufficient depth



Andrews et al. 2016
Nature Reviews Genetics 17: 81-92

IDENTIFICATION OF ADAPTIVE LOCI



ADAPTIVE LOCI

May identify same pattern as neutral markers

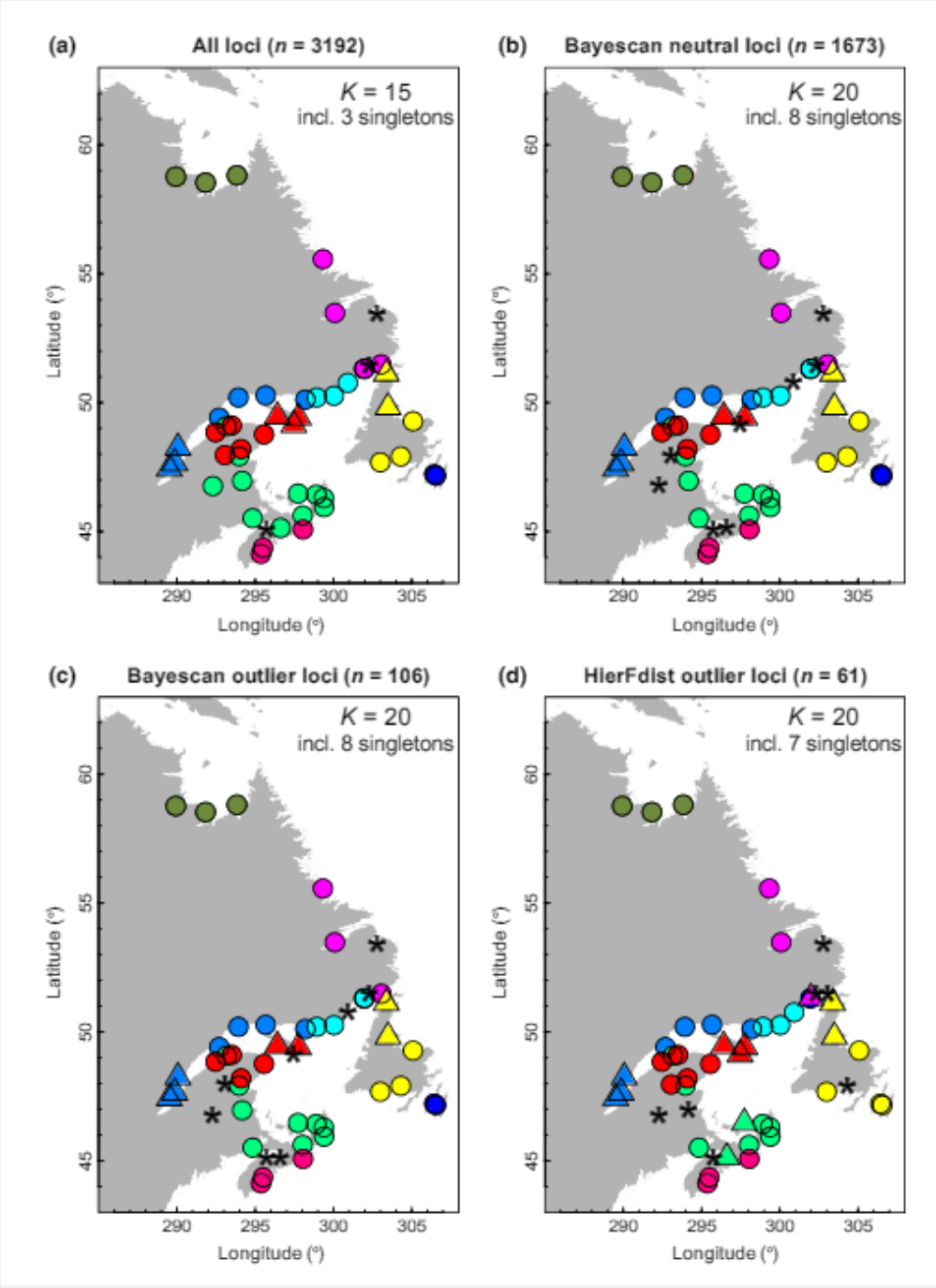
Few loci = less power

Effects of gene flow and small N_e

Many genes of small effect

Problems with pooling all outliers

Moore et al. 2014

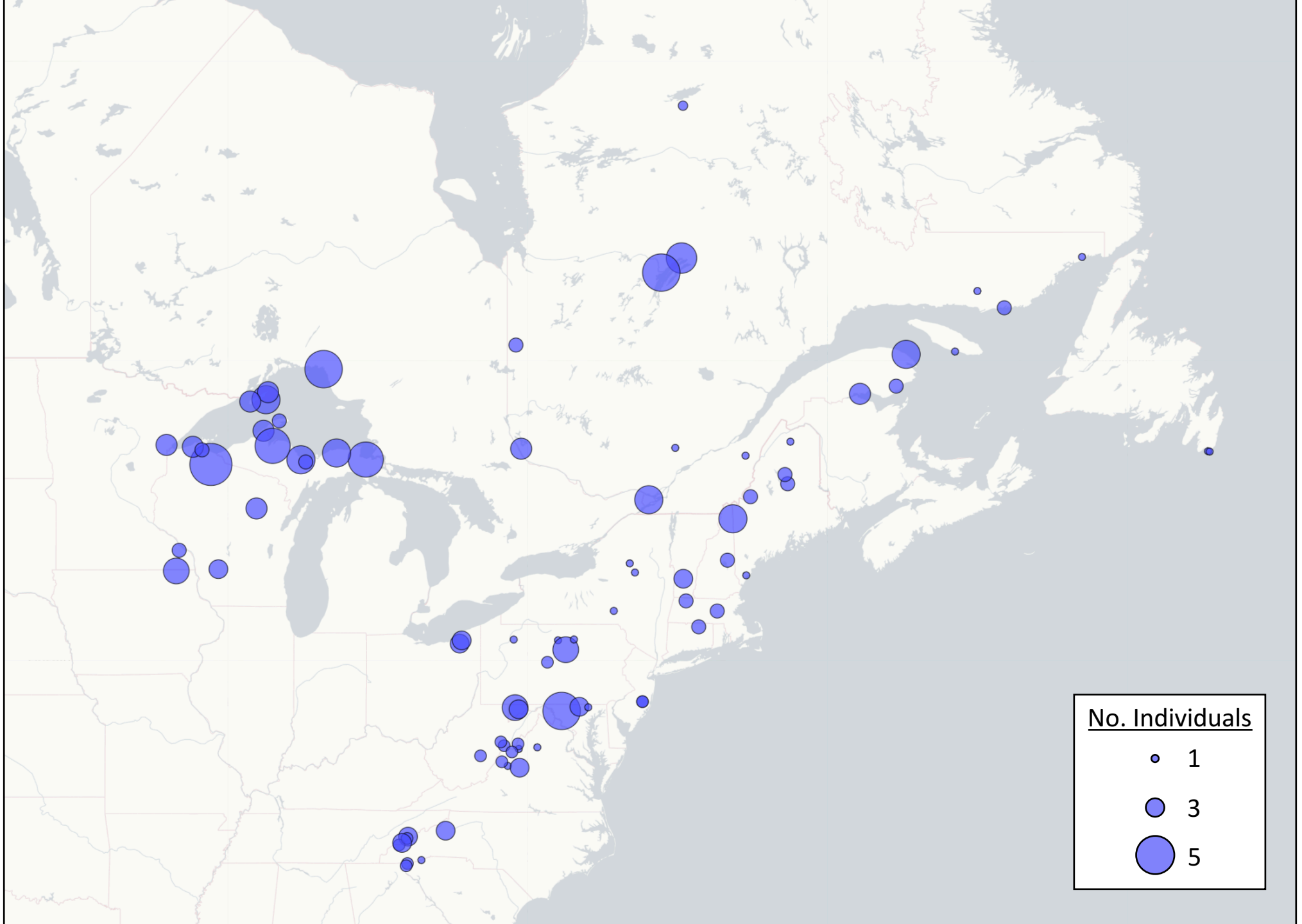


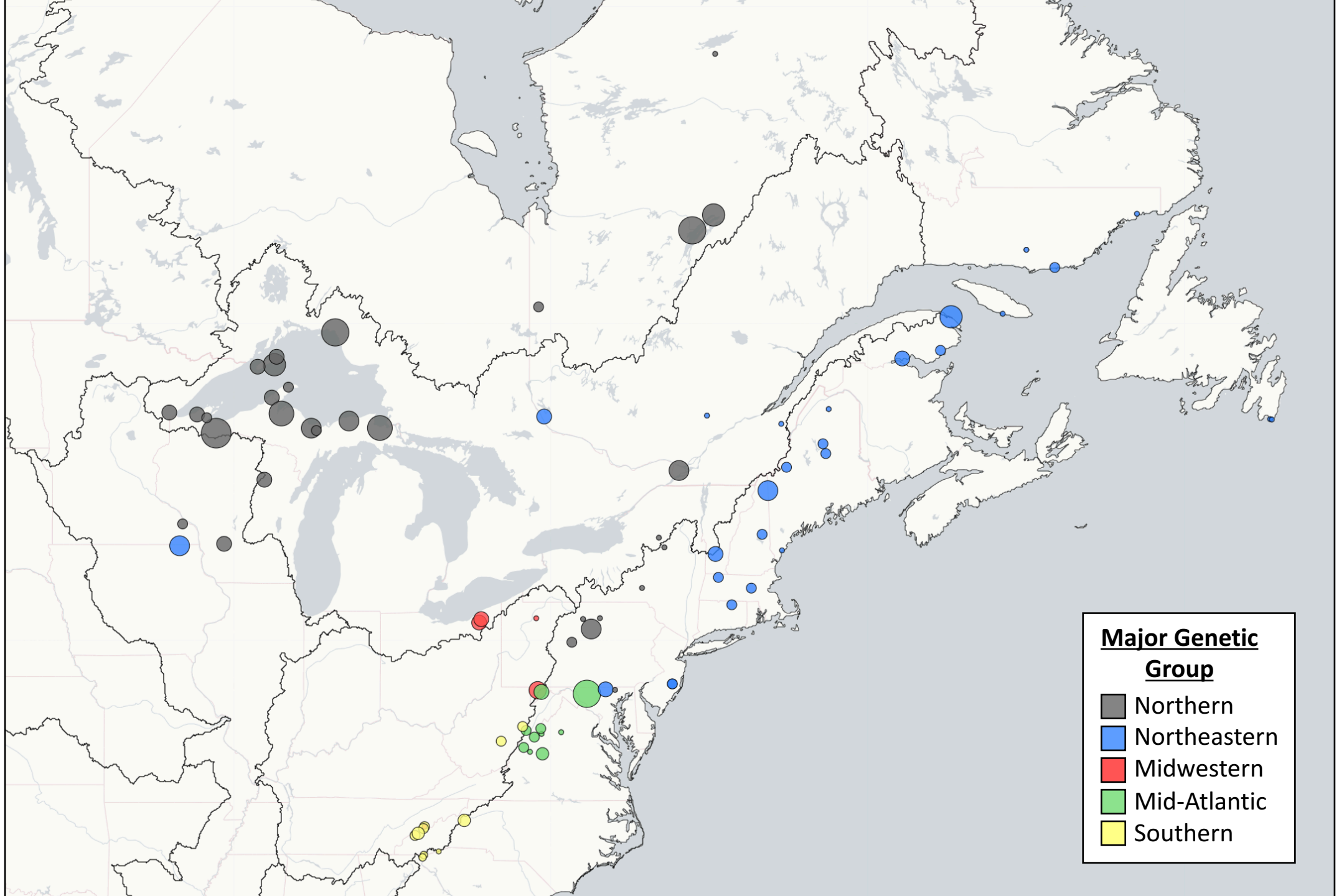
Goal: Develop a genomic resource that enables standardized surveys of genetic diversity in native populations of brook trout

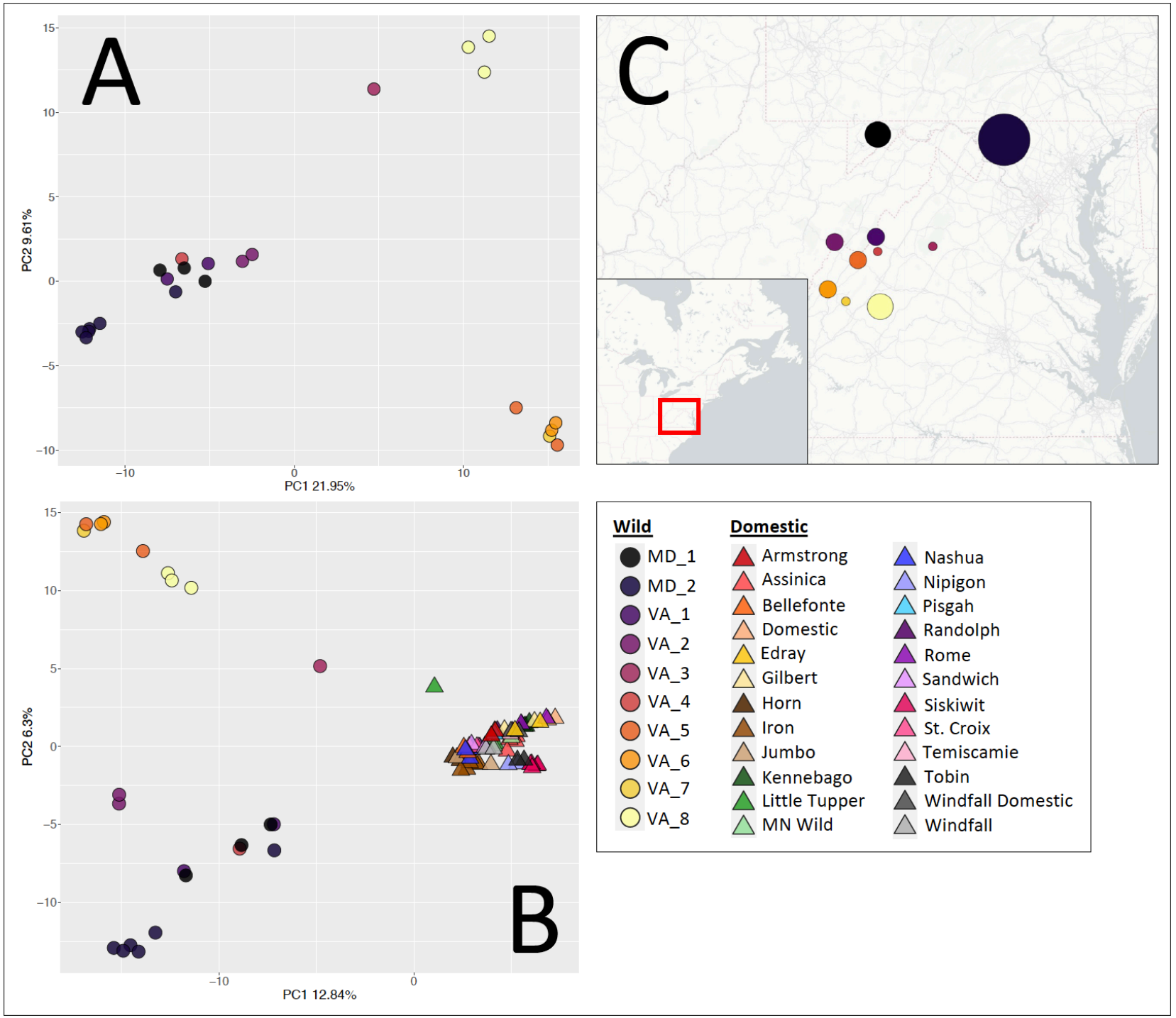
Objectives

1. Identify a suite of genome-wide SNPs representative of range-wide genetic diversity
2. Demonstrate the ability of SNPs to resolve lineages at local to continental scales

Mamoozadeh, N., Whiteley, A., Letcher, B., Kazyak, D., Tarsa, C., & Meek, M. Evaluating genomic relationships across spatial and temporal scales to guide conservation and management of imperiled species. *Submitted to Molecular Ecology Resources.*


No. Individuals
1
3
5


Major Genetic Group
Northern
Northeastern
Midwestern
Mid-Atlantic
Southern


A
PC2 9.61%
PC1 21.95%
B
PC2 6.3%
PC1 12.84%
C
Wild
MD_1
MD_2
VA_1
VA_2
VA_3
VA_4
VA_5
VA_6
VA_7
VA_8
Domestic
Armstrong
Assinica
Bellefonte
Domestic
Edray
Gilbert
Horn
Iron
Jumbo
Kennebago
Little Tupper
MN Wild
Nashua
Nipigon
Pisgah
Randolph
Rome
Sandwich
Siskiwit
St. Croix
Temiscamie
Tobin
Windfall Domestic
Windfall

Application	Key Questions
Identifying biologically appropriate management units	a) Which individuals comprise demographically independent management units (e.g., populations)? b) Which populations comprise adaptive groups? c) On what spatial and temporal scales do populations and adaptive groups occur?
Conservation prioritization	a) Which adaptive groups exhibit the greatest mismatch with predicted future conditions? b) Which adaptive groups exhibit the greatest adaptive potential? c) Are there populations or adaptive groups that harbor unique genetic variation warranting special protection, including variation associated with distinct phenotypic traits?

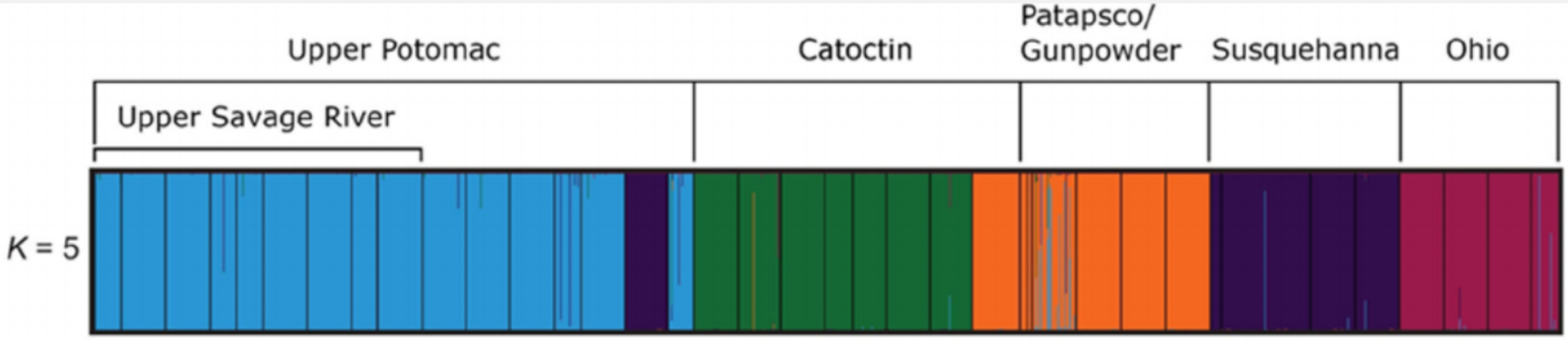
Mamoozadeh, N., Whiteley, A., Letcher, B., Kazyak, D., Tarsa, C., & Meek, M. Evaluating genomic relationships across spatial and temporal scales to guide conservation and management of imperiled species. *Submitted to Molecular Ecology Resources.*

GENETIC MEASURES

MEASURES OF GENETIC DIFFERENTIATION

Bayesian clustering

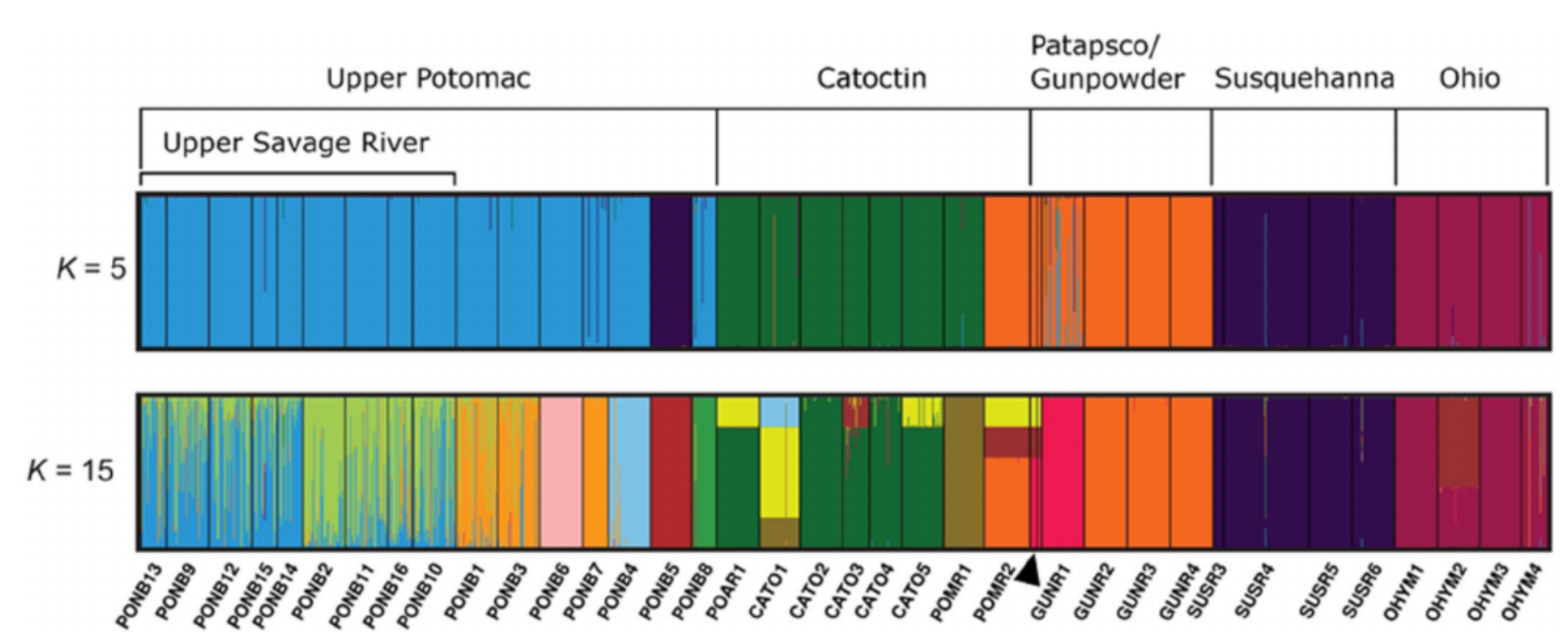
- STRUCTURE
- Identify most likely number of populations based on genetic data
- Clusters in HWE
- Determine individual membership to each cluster



Morgan et al. (2021)

MEASURES OF GENETIC DIFFERENTIATION

- Detects highest level of clustering first
- Poor job at detecting weak structure

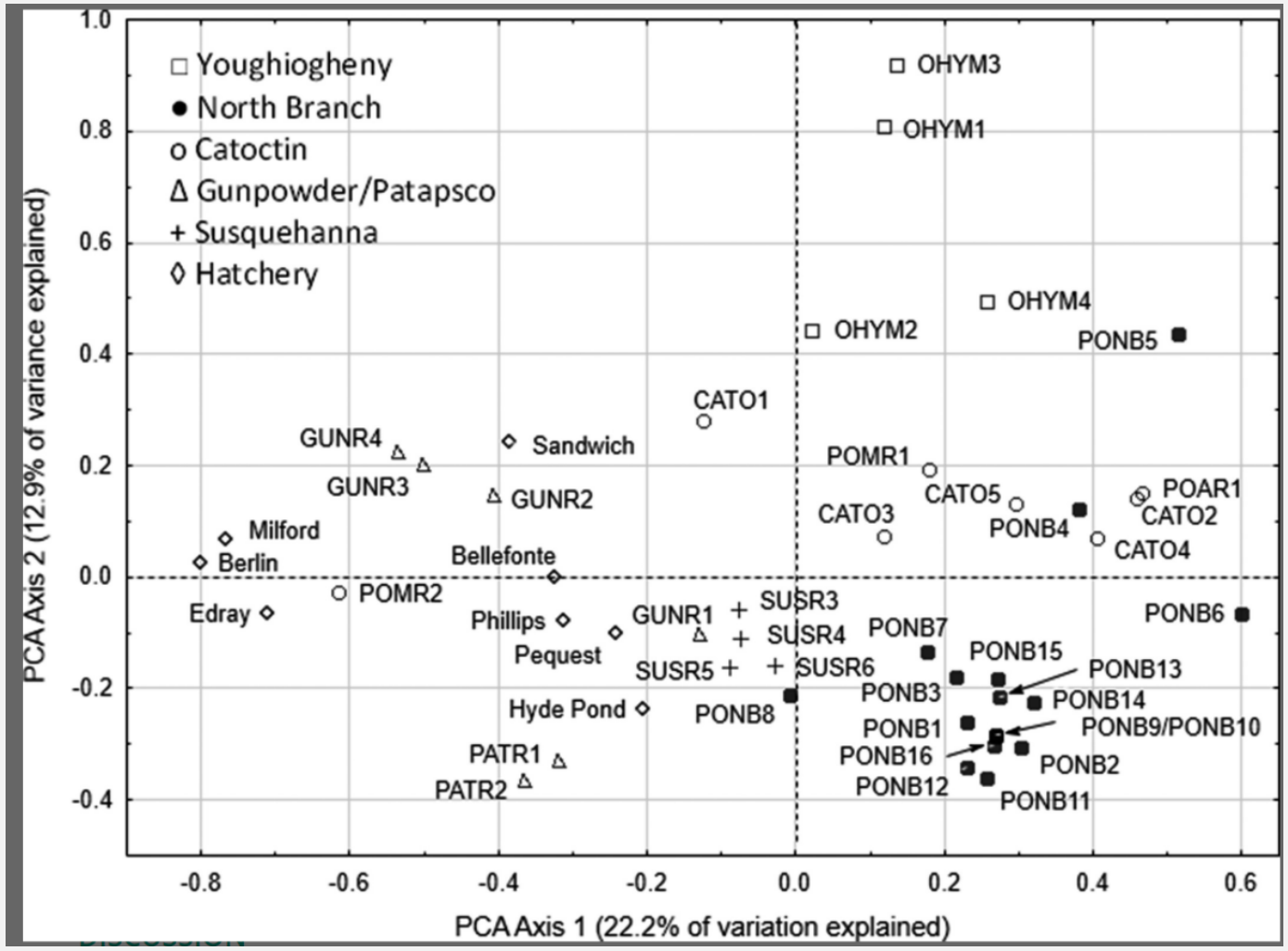


Morgan et al. (2021)

MEASURES OF GENETIC DIFFERENTIATION

PCoA, PCA, DAPC

- Based on allele frequencies
- Maximal variation on minimal axes



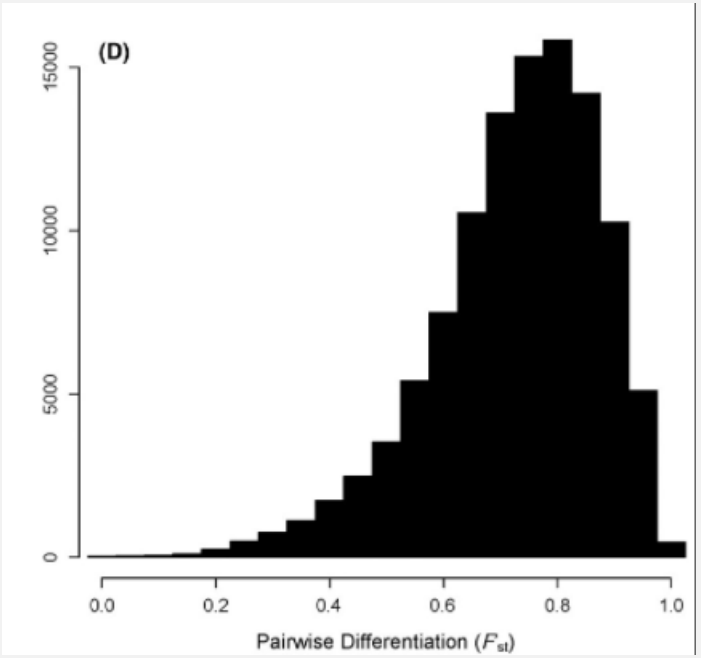
Morgan et al. (2021)

MEASURING GENETIC DIFFERENTIATION

$$F_{ST} = 1 - \frac{H_S}{H_T}$$

Ranges from 0 (the same) to 1 (completely different)

	Dillons_2020	Dillons	Edwards	Hawk	New Creek	Himmelwright	Trout Pond	Waites	Capon	Linton	Mill	Reymann	VA_DR	VA_CC	Edray
Dillons	0.044														
Edwards	0.150	0.132													
Hawk	0.168	0.145	0.222												
New Creek	0.137	0.125	0.167	0.190											
Himmelwright	0.186	0.127	0.219	0.149	0.201										
Trout Pond	0.153	0.095	0.243	0.252	0.164	0.245									
Waites	0.123	0.082	0.132	0.115	0.119	0.131	0.154								
Capon	0.163	0.161	0.253	0.195	0.143	0.225	0.227	0.164							
Linton	0.358	0.316	0.317	0.461	0.266	0.417	0.391	0.334	0.338						
Mill	0.255	0.259	0.326	0.314	0.207	0.346	0.292	0.263	0.240	0.504					
Reymann	0.066	0.016	0.179	0.196	0.162	0.197	0.088	0.137	0.206	0.370	0.285				
VA_DR	0.195	0.204	0.313	0.264	0.183	0.301	0.197	0.226	0.194	0.421	0.278	0.231			
VA_CC	0.177	0.195	0.228	0.224	0.139	0.266	0.220	0.167	0.181	0.354	0.228	0.232	0.157		
Edray	0.286	0.260	0.249	0.348	0.201	0.348	0.297	0.281	0.273	0.377	0.382	0.310	0.314	0.283	
Paint Bank	0.184	0.168	0.209	0.260	0.120	0.260	0.206	0.178	0.225	0.333	0.278	0.203	0.210	0.181	0.236



Kazyak et al. (2021)

MEASURING GENETIC DIVERSITY

Microsatellites

- (Polymorphic loci)
- Observed & expected heterozygosity
- Allelic richness
- F_{IS} & relatedness
- Effective population size (N_e)

SNPs

- Polymorphic loci
- Observed & expected heterozygosity
- (Allelic richness) Nucleotide diversity
- F_{IS} & relatedness
- Effective population size (N_e)

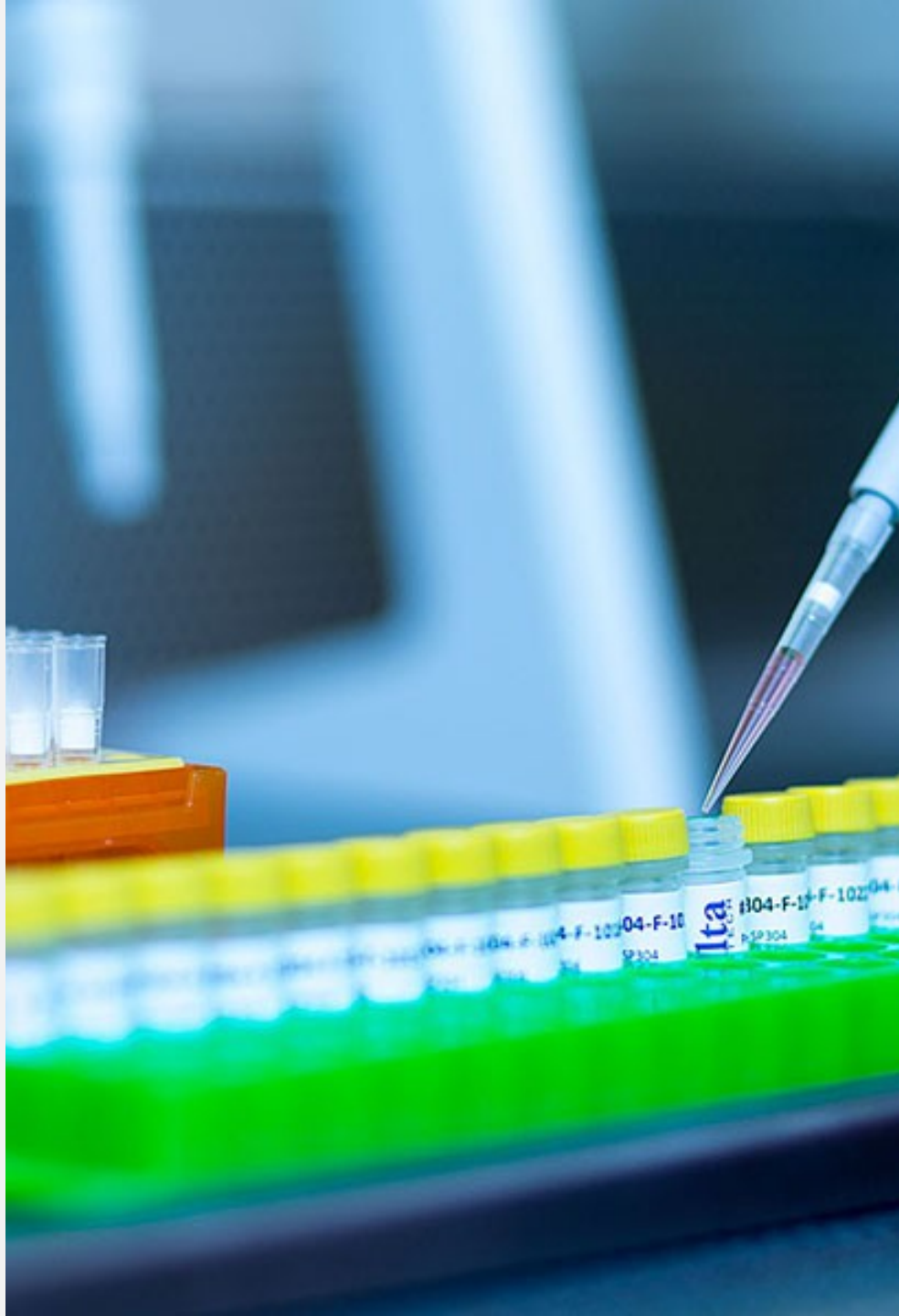
OTHER GENOMIC TOOLS

POPULATION TRANSCRIPTOMICS (RNA-SEQ)

- Gene expression
- Transcriptome: expression pattern of all transcribed elements in the genome
- Measuring differences in copy numbers of genes

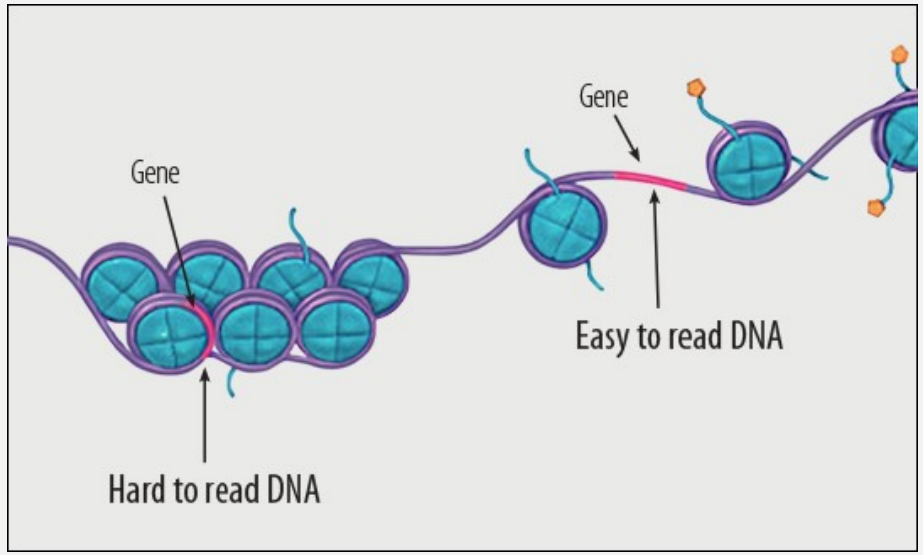
RNA-SEQ CHALLENGES

- Need high-quality samples that are carefully preserved
- Gene expression can vary depending on tissue type
- May need to take invasive tissues
- Laboratory practices can result in “gene expression” differences
- Gene expression changes with time (management implications?)



EPIGENETICS

- (Potentially) heritable gene expression changes
- No DNA sequence change
- Modifications to the genome (e.g., DNA methylation)
- Hypomethylation – expression turned on
- Hypermethylation – expression turned off



Considerations:

- Can vary depending on tissue type
- Indirect link to gene expression
- More stable than RNA-Seq