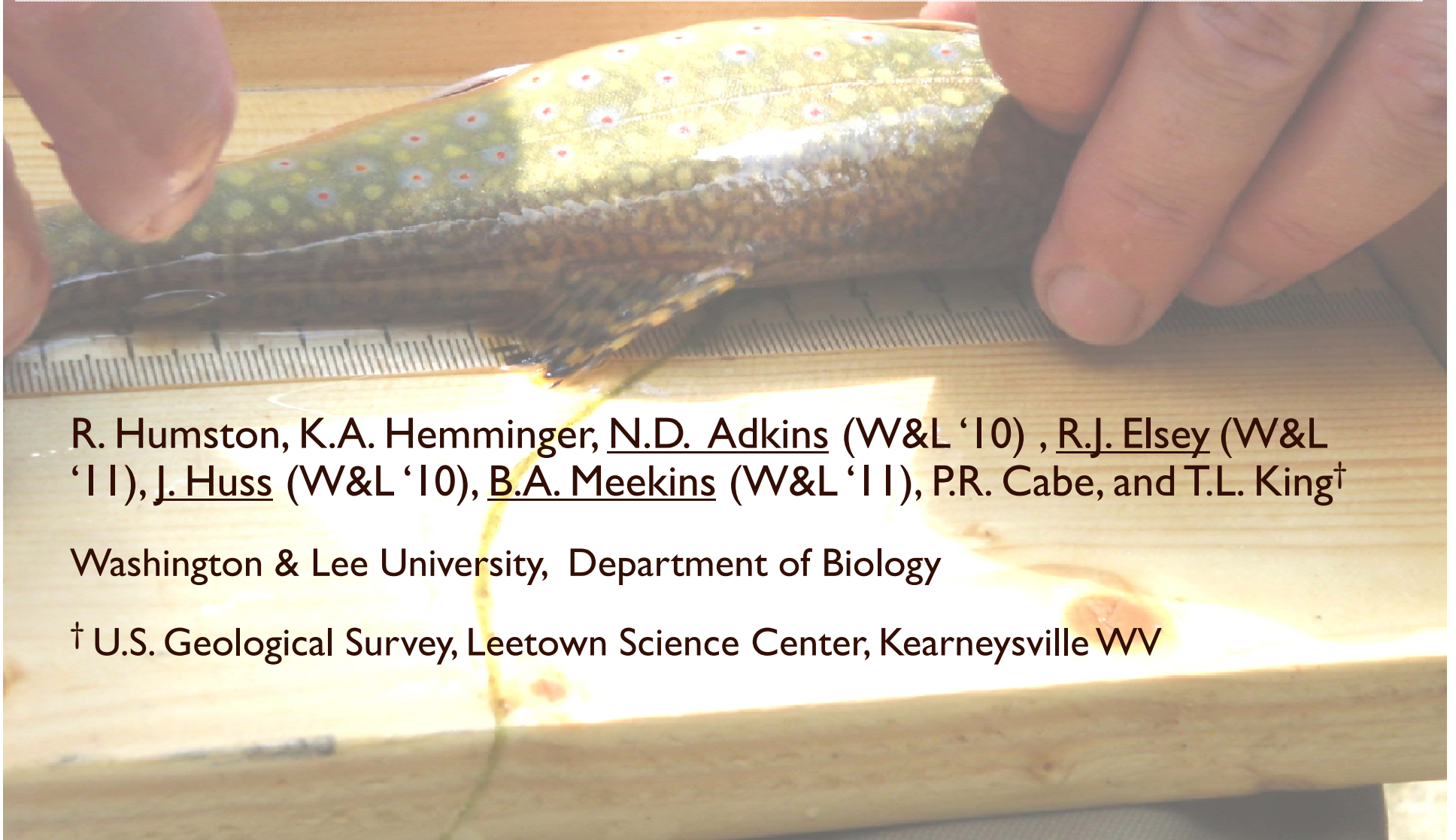


# Stocking Brook Trout in Virginia's reservoirs: dispersal, naturalization, and introgression with native trout populations



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# Stocking Hatchery Trout





# Stocking Hatchery Trout

- **Positives:**

- Reduces fishing pressure on wild (native) populations.
- Restore populations where extirpated.
- More cost effective than translocation of fish from intact populations.

- **Negatives:**

- Non-native species / strain introductions
- Suppression of native stocks and genetic impacts



# Genetic impacts of hatchery supplementation

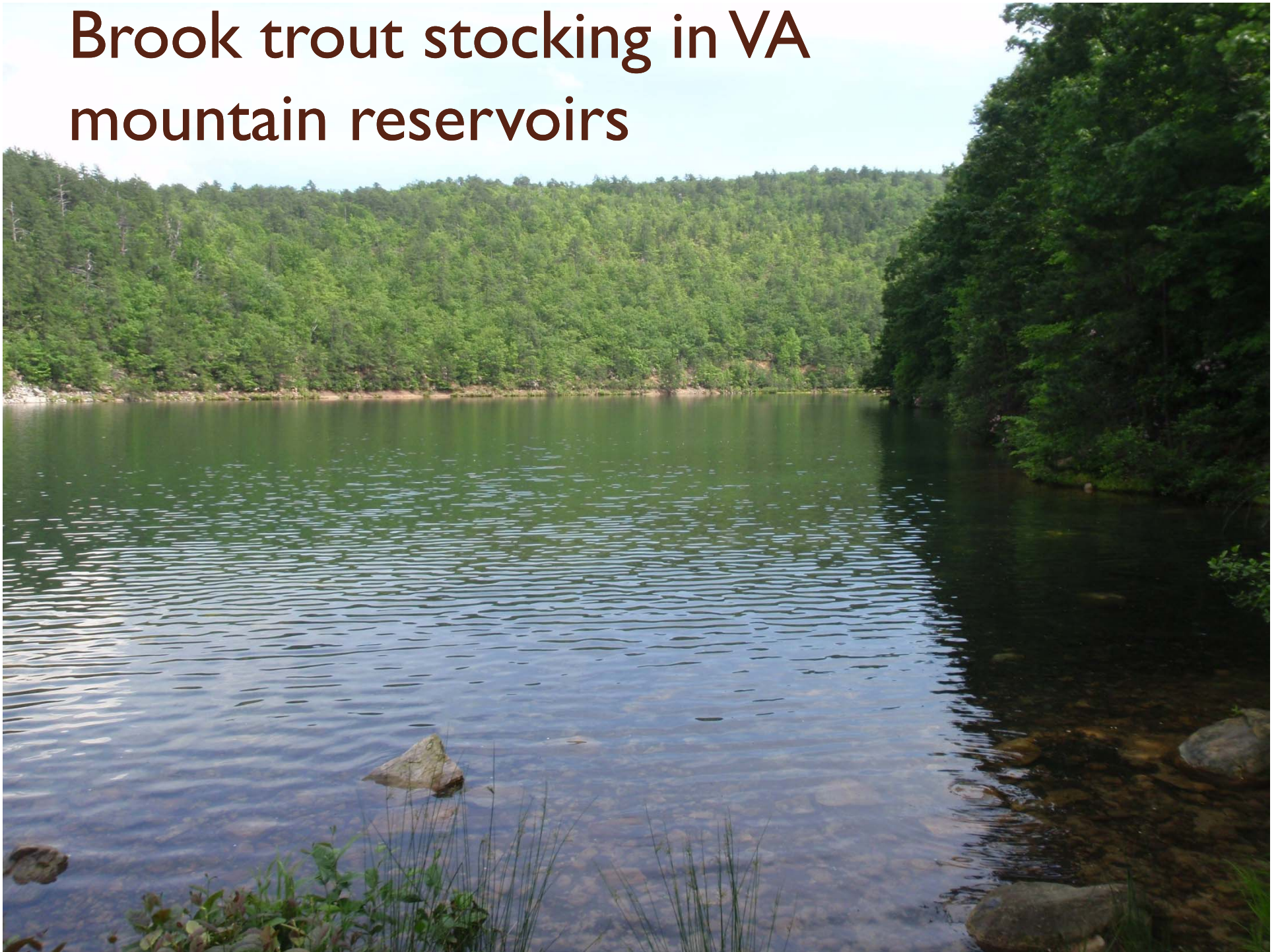
- **Poor fitness** of hatchery lineages
- **Homogenization** of genetic structure among populations.
- Loss of **functional diversity** in native / wild stocks (e.g. local adaptation).
- Potentially can hinder recovery, persistence of wild stocks



# Direct / Indirect Genetic Impacts

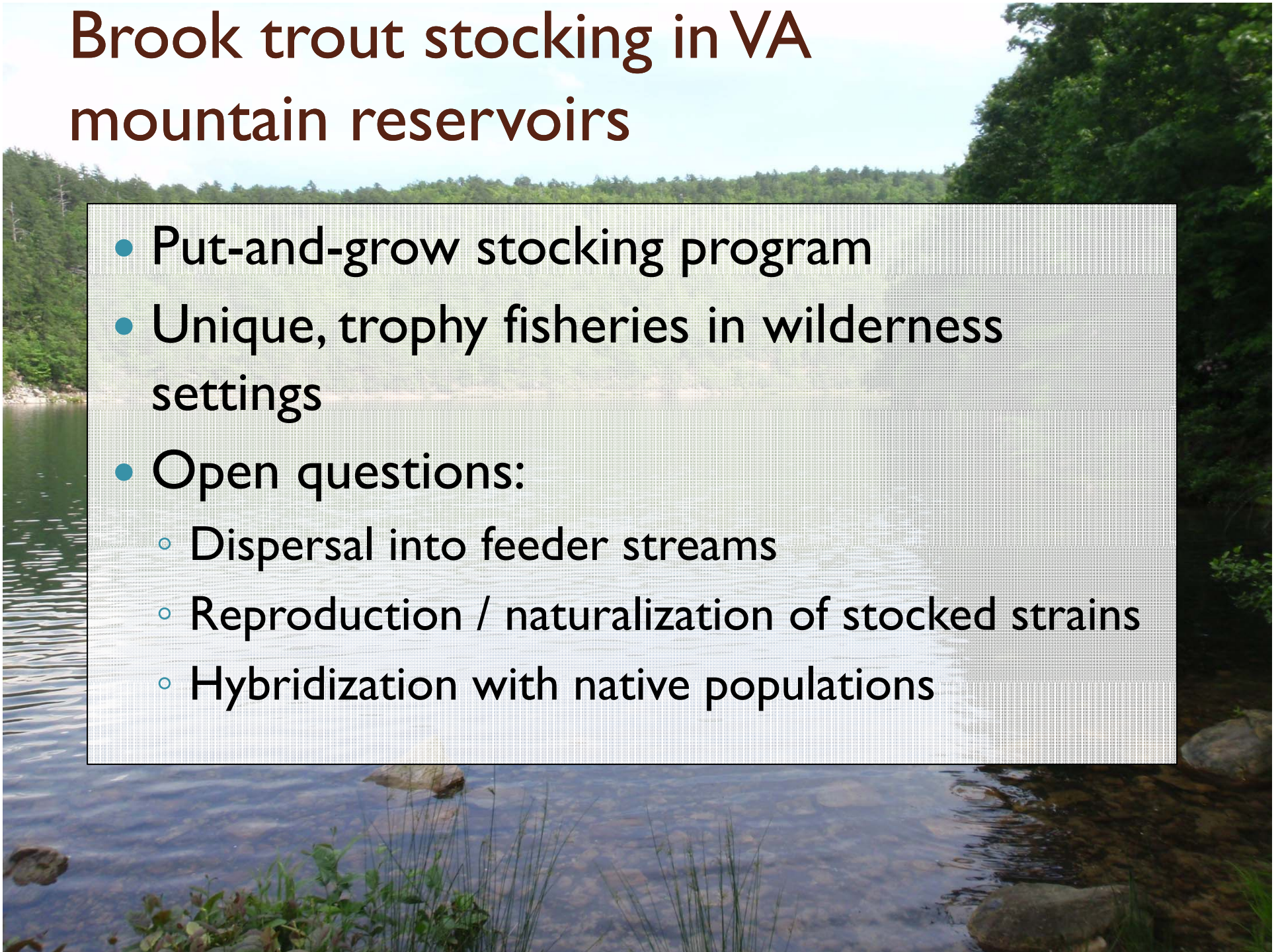
- Direct
  - Results of interbreeding
  - Reduced genetic variability among populations (*homogenization*)
  - *Outbreeding depression* and reduced fitness of subsequent generations
- Indirect:
  - Reduced *effective population size* ( $N_e$ )
  - Increased inbreeding, genetic drift

# Brook trout stocking in VA mountain reservoirs



# Brook trout stocking in VA mountain reservoirs

- Put-and-grow stocking program
- Unique, trophy fisheries in wilderness settings
- Open questions:
  - Dispersal into feeder streams
  - Reproduction / naturalization of stocked strains
  - Hybridization with native populations

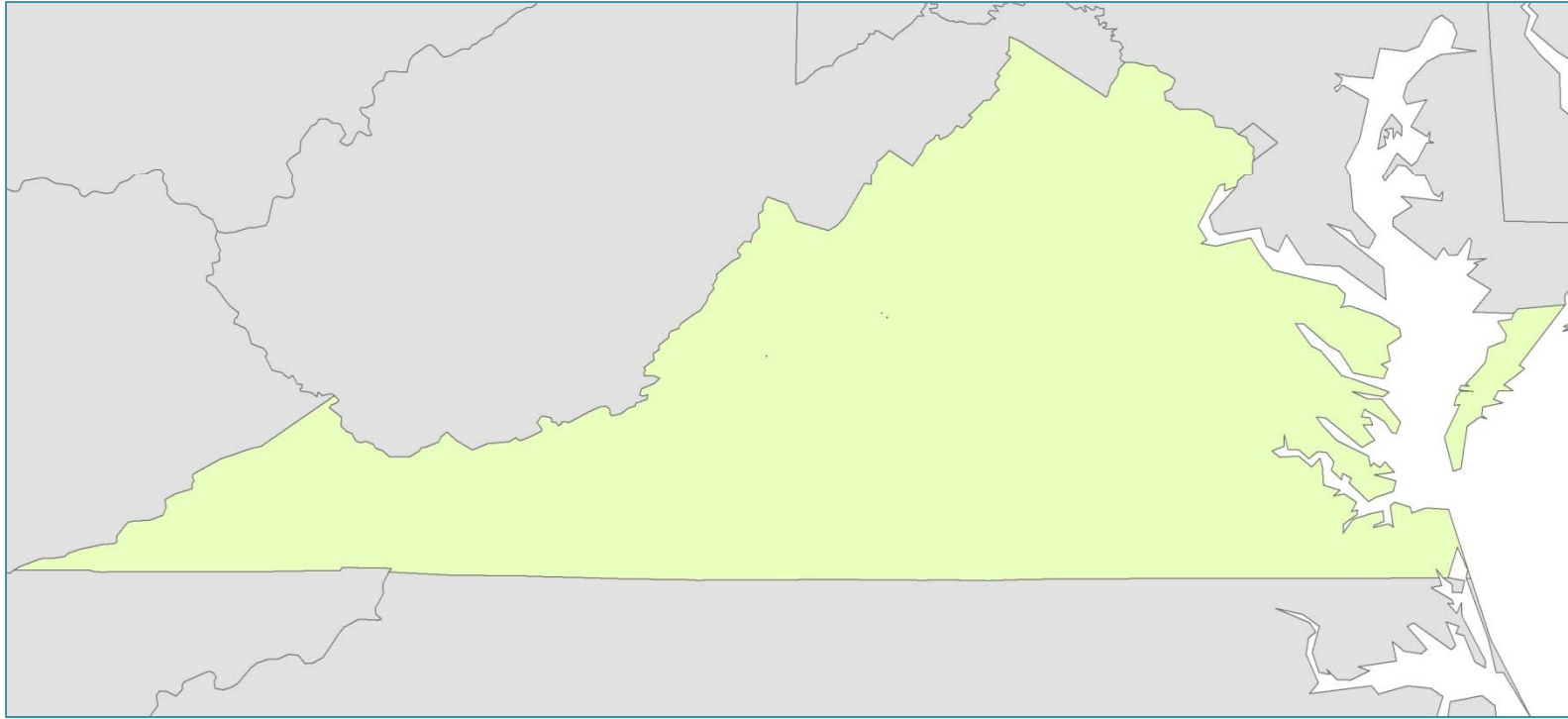




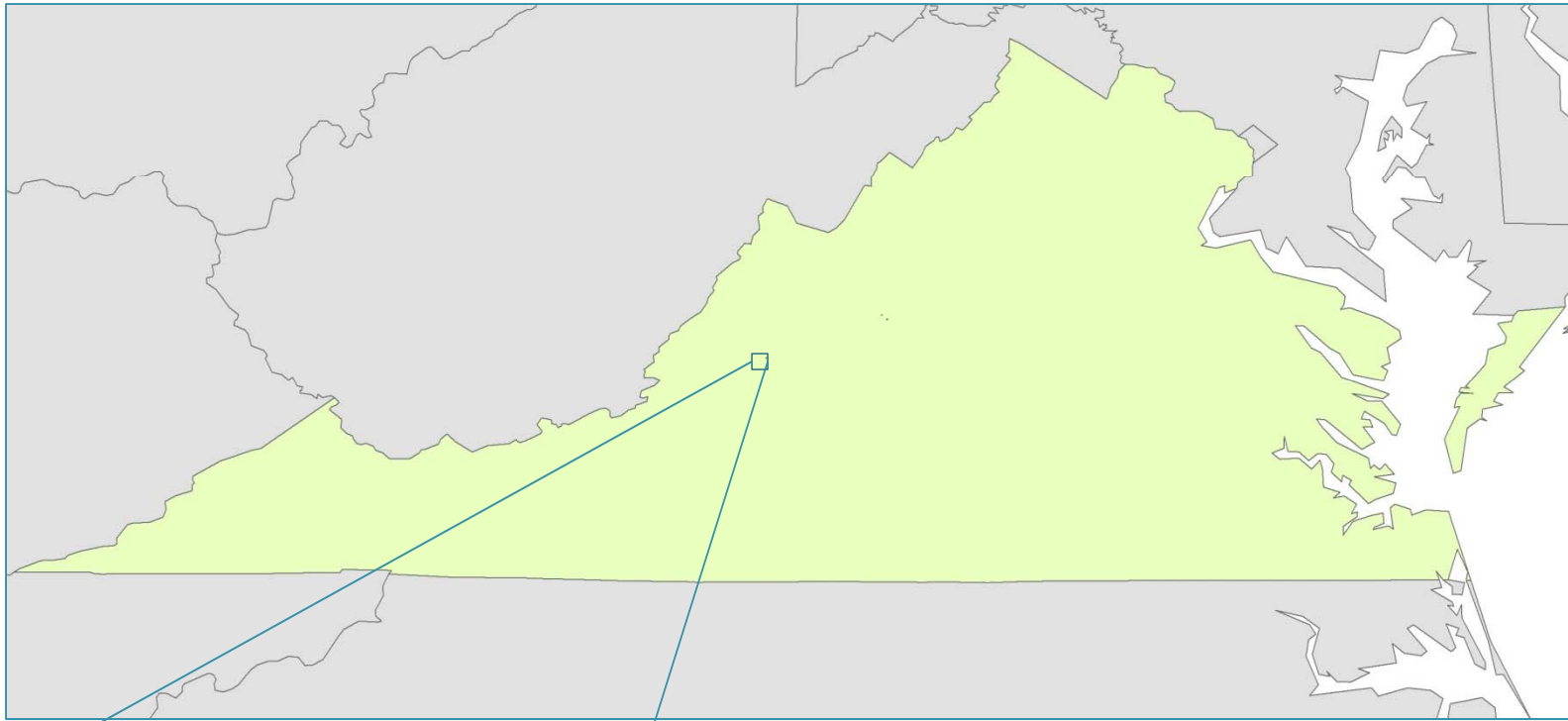
## Study Objectives:

- 1) Determine if stocked brook trout disperse into and inhabit tributaries.
- 2) Determine if natural reproduction of hatchery-origin fish occurs in tributaries.
- 3) Assess degree of introgression (interbreeding) occurring between hatchery and native strains.

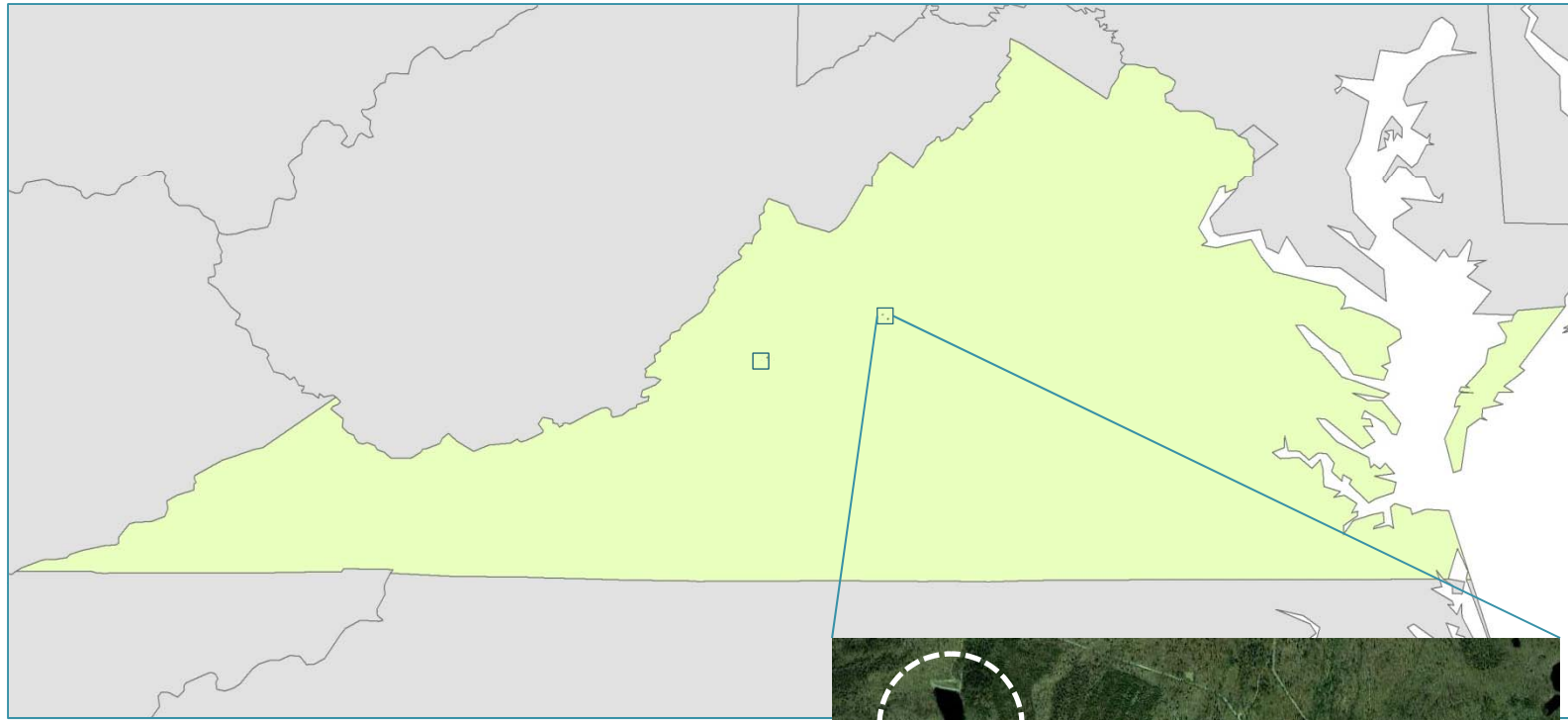




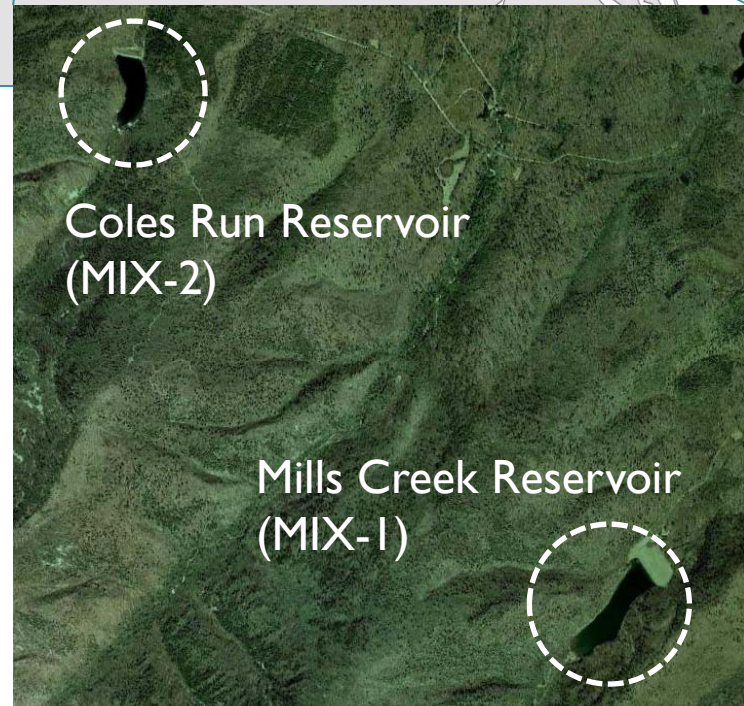
**Study Sites - 2009**

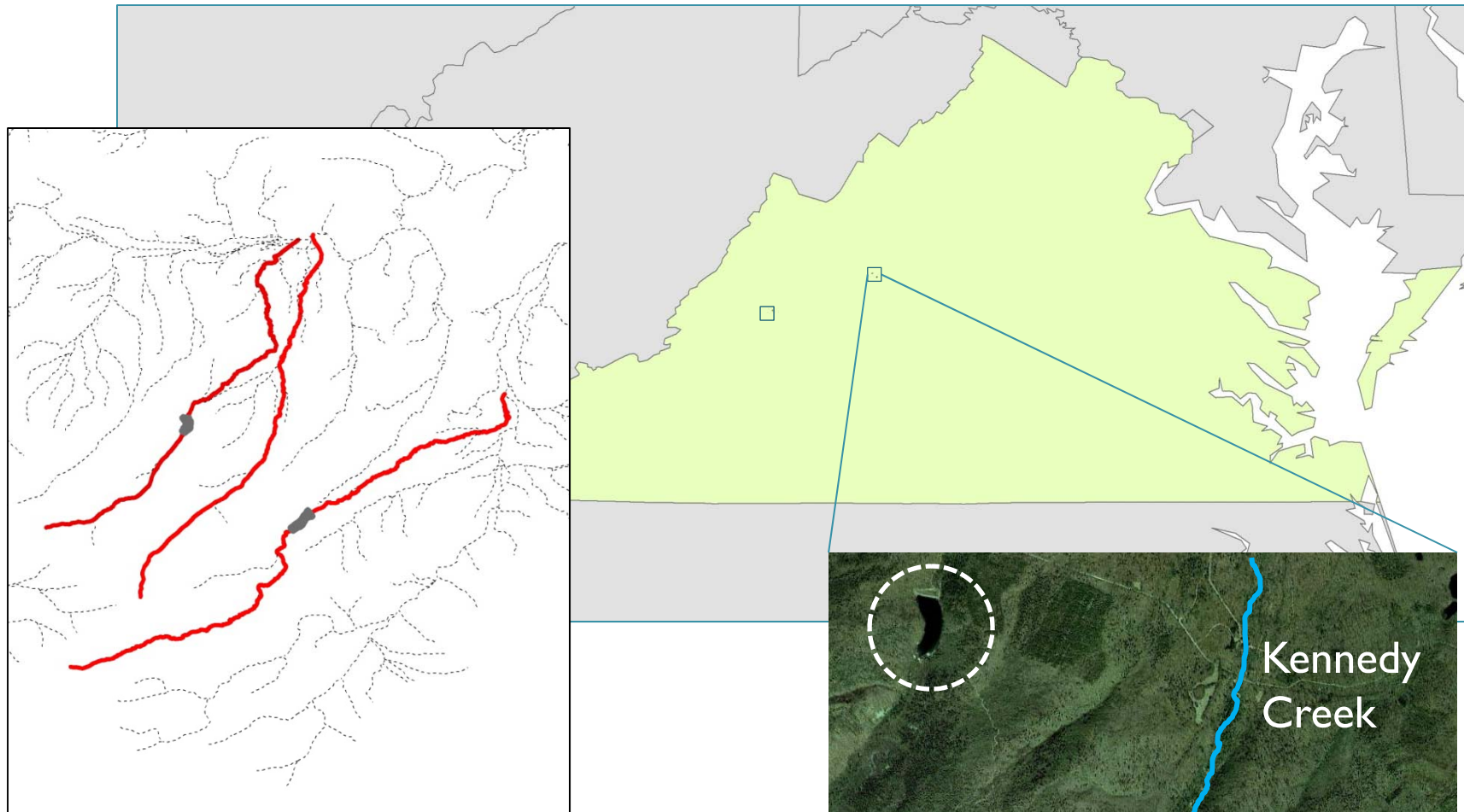


- Historic water supply impoundment for Lexington.
- Stocked with brook trout since 1984.
- **Moore Creek (feeder tributary) did not hold brook trout prior to stocking.**
- **Code name: STO**

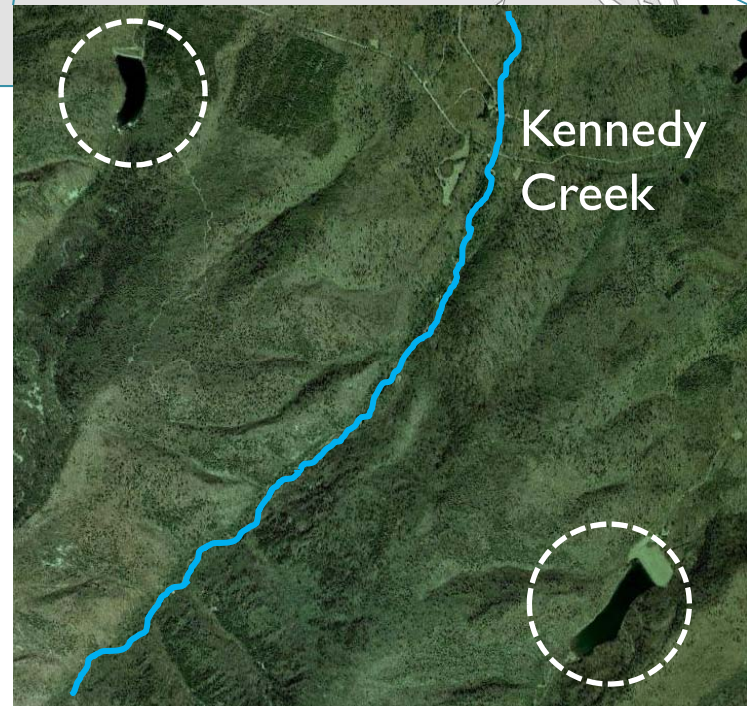


- Historic water supply & flood control impoundments.
- Stocked with brook trout since 1989.
- Wild (putative native) brook trout populations in feeder streams.
- **Code named: MIX-1 & MIX-2**





- Unstocked wild (native) trout stream.
- “Reference” native population for subwatershed.
- **Code name: NAT**





# Stocking History

- All fingerlings stocked in reservoirs from a single brood stock lineage (Paint Bank hatchery, VA).
- 
- Only exception:
    - Fingerlings stocked in 2008 derived from Rome Lab strain.
    - This cohort would not have spawned in time to contribute offspring to population by summer 2009.



## Study Objectives:

Determine if stocked fish disperse into and inhabit tributaries.

- *Sample Moore Creek (STO; no previous wild population) for trout.*
- *Use genetic data to distinguish hatchery-strain from native trout in Mills Creek (MIX-1) and Coles Run (MIX-2).*



## Study Objectives:

Determine if natural reproduction of hatchery-origin fish occurs in tributaries.

- *Compare genotypes of 2008 year class with known hatchery strains (Paint Bank, Rome Lab) to determine if Paint Bank strain naturally produced offspring in 2008.*



# Study Objectives:

Assess degree of introgression occurring between hatchery and native strains.

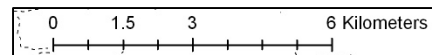
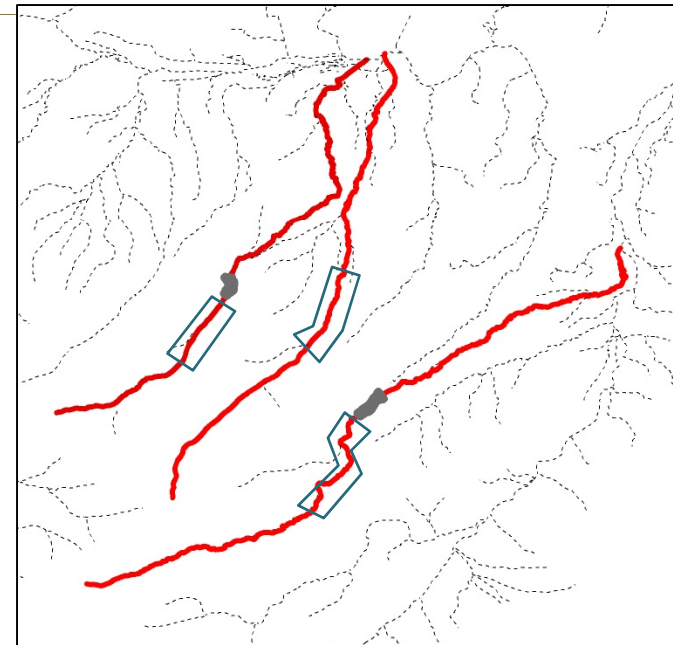
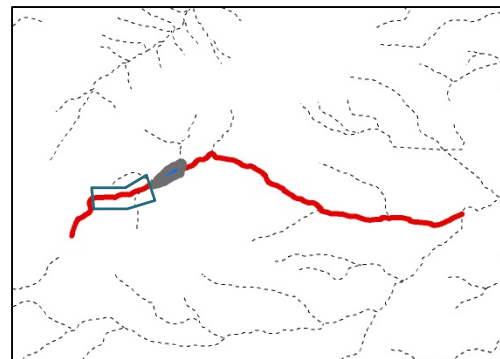
- *Use “assignment tests” to identify potential ‘hybrids,’ and examine individual genotypes to determine likelihood of hybrid status.*





# Field collections

- June-July 2009
- Single-pass electrofishing
- Pelvic fin snips
- Measure in field (TL)





# Hatchery samples

- Paint Bank brood stock and 2009 young-of-year
- Pelvic fin snips
- Rome Lab genotyped in 2005 and 2009 (T.L. King, unpublished data)





# DNA analysis

- Genotyped at ten (10) established, polymorphic **microsatellite** loci (T.L. King, unpublished data)

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  - Non-coding regions of DNA
  - Repeated sequences of 1-6 specific base pairs.
  - Alleles vary by length (number of repeats in a row).
  - Highly versatile genetic markers



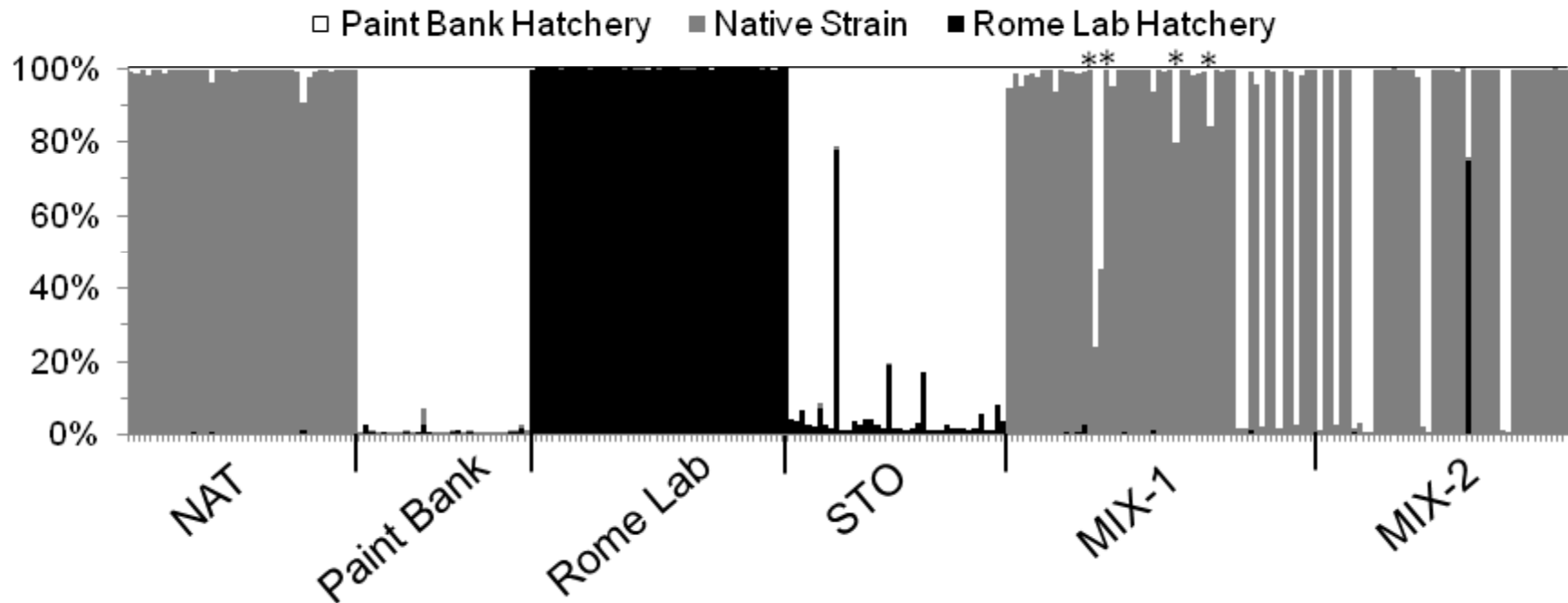
# Data Analysis – Genetic Data

- Program **Structure**
  - Groups individuals into “clusters” that best approximate distinct populations based on allele frequencies.
  - Provides *posterior probability* of population membership for individuals (population *assignment*).



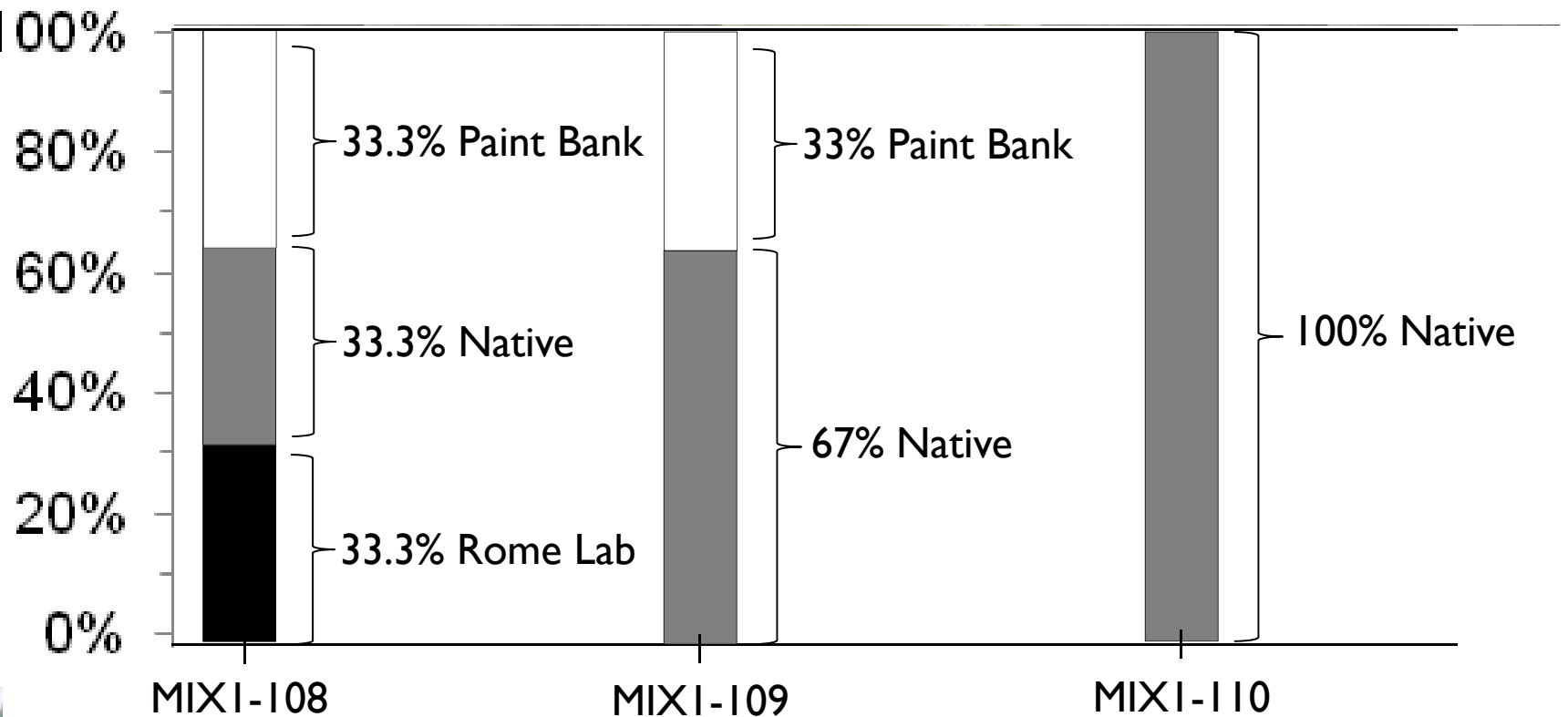
# How to read Structure output

The probability of membership to each population is represented in a 'stacked' bar chart with a bar for each fish.



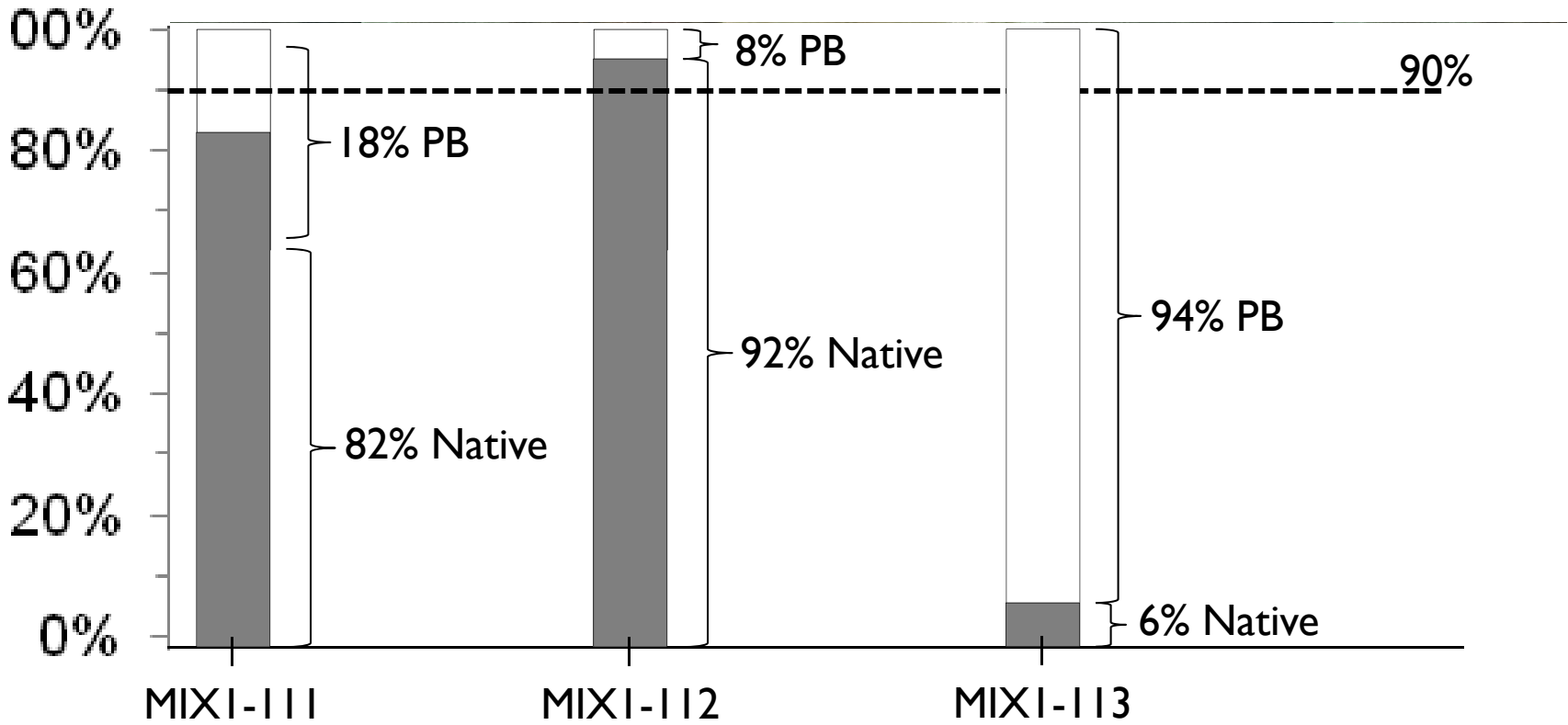
# How to read Structure output

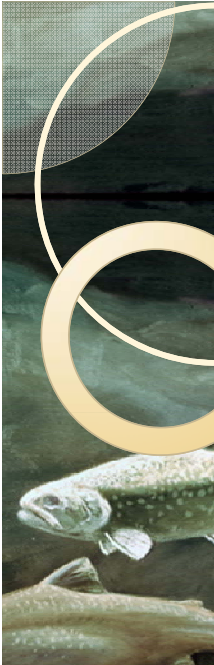
- Paint Bank Hatchery strain
- Rome Lab Hatchery strain
- Native strain



# Assignment "Rules"

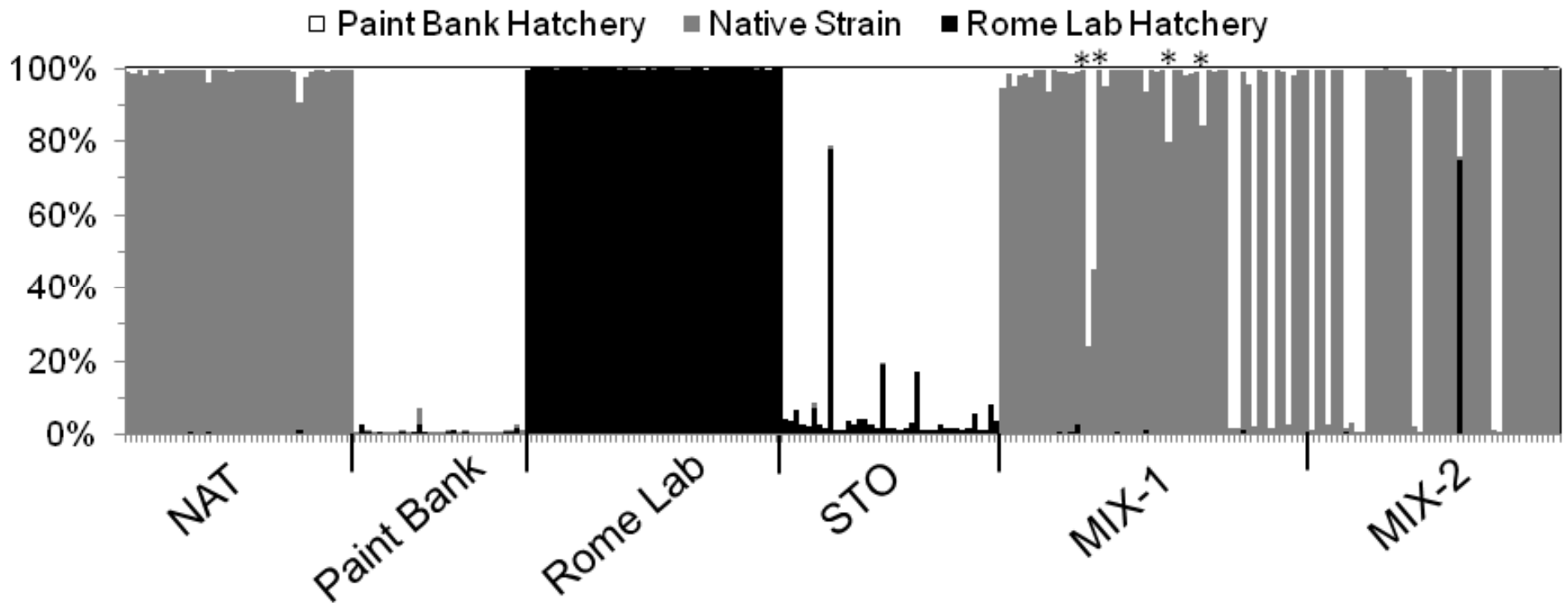
- Paint Bank Hatchery strain
- Rome Lab Hatchery strain
- Native strain



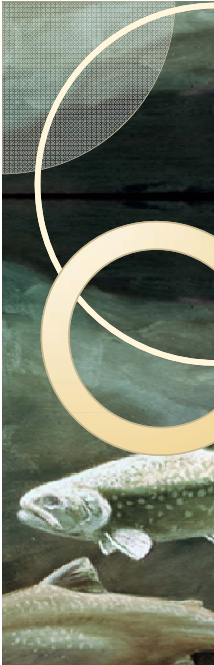


# Results: STRUCTURE

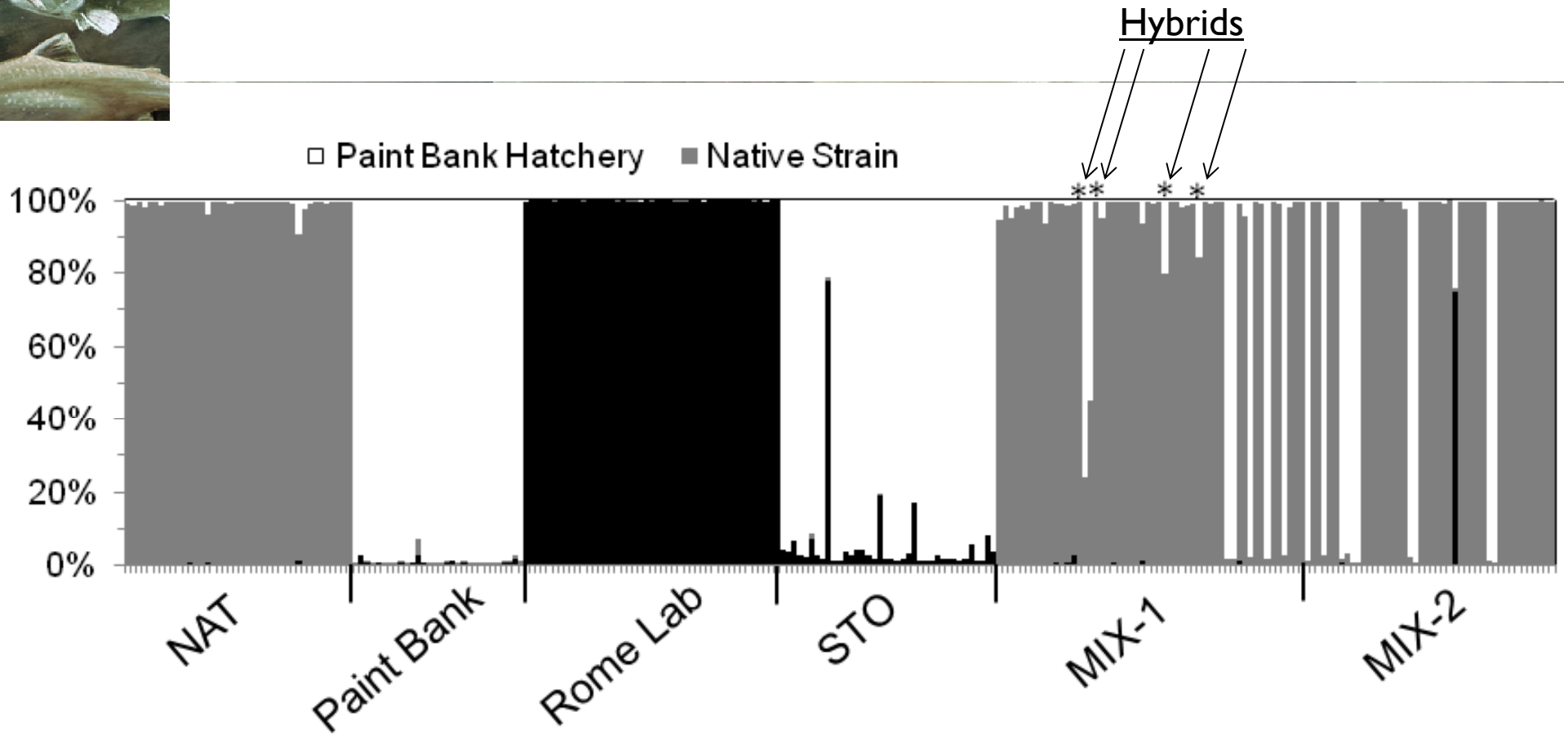
- Best solution = 3 populations in collection





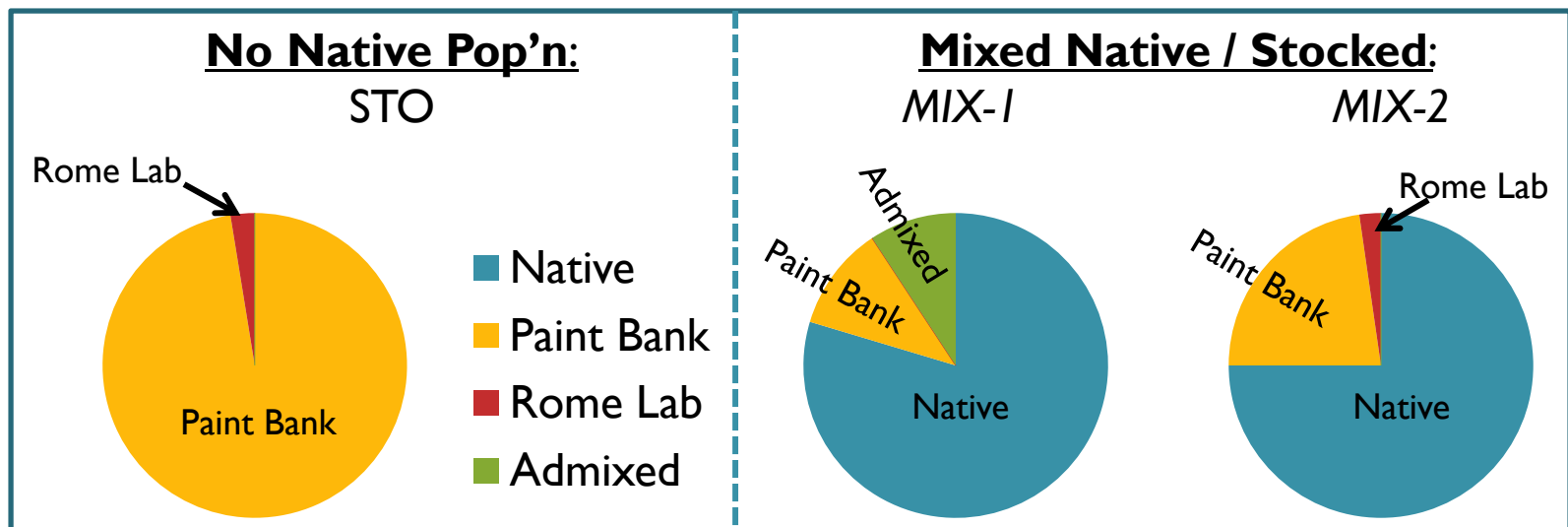


# Results: STRUCTURE



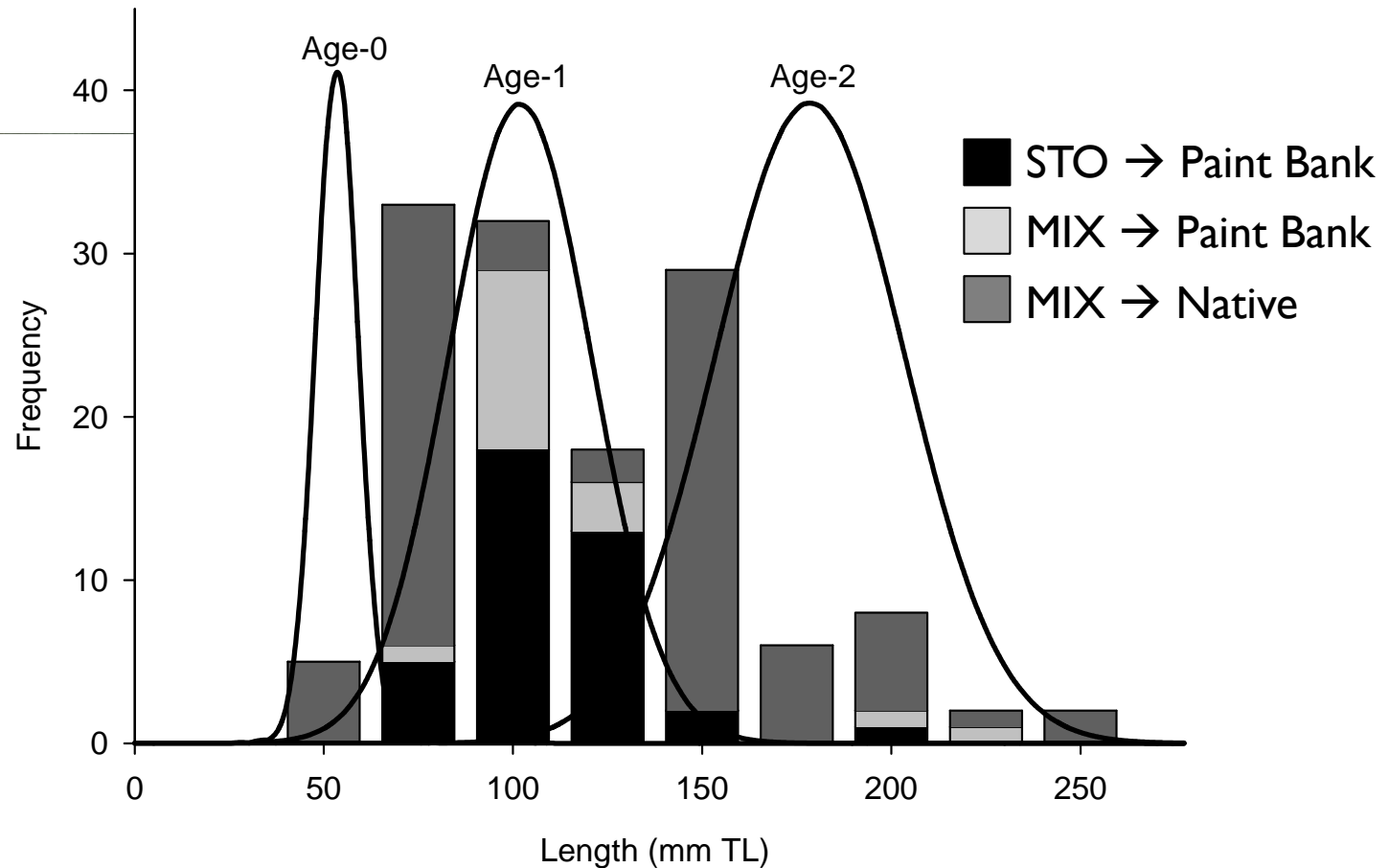
# Results: Dispersal into Tributaries

Sample Location	<i>N</i>	Size Range (mm TL)	Native	Paint Bank	Rome Lab	Hybrids
STO	38	66-180	<b>0</b>	<b>38</b>	<b>1</b>	<b>0</b>
MIX-1	54	47-235	<b>43</b>	<b>7</b>	0	<b>4</b>
MIX-2	44	46-245	<b>33</b>	<b>10</b>	<b>1</b>	<b>0</b>
NAT	40	41-216	100%			
Paint Bank	30	74-321		100%		
Rome Lab	45	NA			100%	
<b>Total</b>	<b>251</b>	<b>41-321</b>				



# Results: Hatchery Strain Spawning

*All age-1 stocked fish should be Rome Lab strain.*





# Conclusions

- Hatchery fish do disperse into streams from reservoirs, even where native populations exist.
    - Not a big surprise.
-



# Conclusions

- Hatchery fish do reproduce in streams even where native populations exist, and can establish ‘naturalized’ populations.

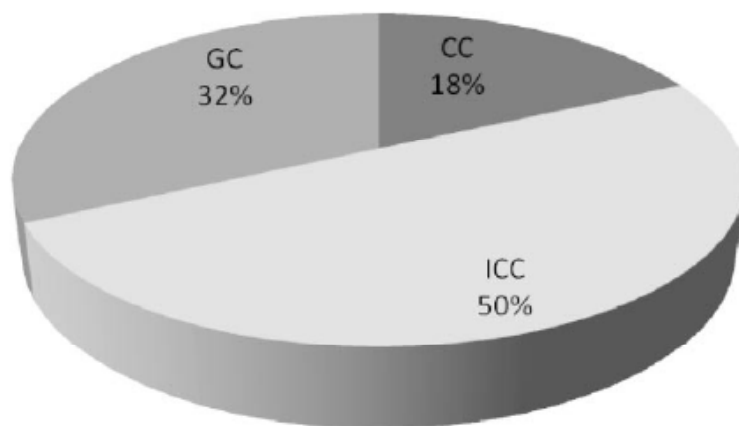
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- Promising for re-establishment of extirpated stocks.
- Suggests fitness of hatchery stocks may not be an impediment to interbreeding.



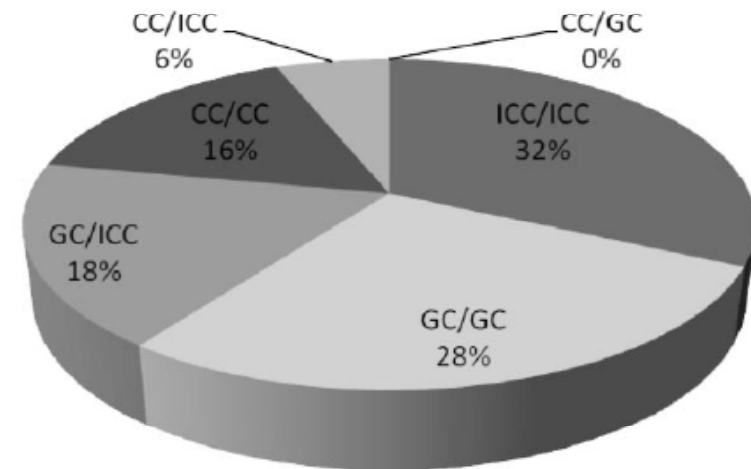
# Conclusions

- Introgression has been limited over the 20+ year stocking history in these systems.
  - Assortative mating?

*Richards et al. 2008*



**Figure 2.** Pie chart depicting the assignment of LeConte Creek brook trout to each of three source populations (CC, Cosby Creek; GC, Greenbrier Creek; ICC, Indian Camp Creek).



**Figure 3.** Percentage of LeConte Creek population assigned to each parental lineage combination (i.e., both parents from the same source population) (CC, Cosby Creek; GC, Greenbrier Creek; ICC, Indian Camp Creek).



# Conclusions

- Good news scenario:
  - Direct impacts to “native” genotype has been minimal.

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- Potential issues:
  - Naturalized hatchery strains could lead to future introgression in native populations.



## New Questions:

- Stream residents or lacustrine migrants?
- Timing of upstream dispersal?
- Ratio of dispersers : stayers?
- Exclusionary processes?
  
- Indirect genetic effects?
- Environmental factors, e.g. stream pH?





# Acknowledgements

- Larry Mohn, Paul Bugas, Jason Hallacher, Aaron Coffman (VA DGIF)
- Dawn Kirk (USFS)
- Barb Lubinski (USGS)
- Eric Hallerman, Joanne Davis, Jamie Roberts (VA Tech Fisheries & Wildlife)
- Funding from Hess Scholars Program (W&L), Mellon Foundation





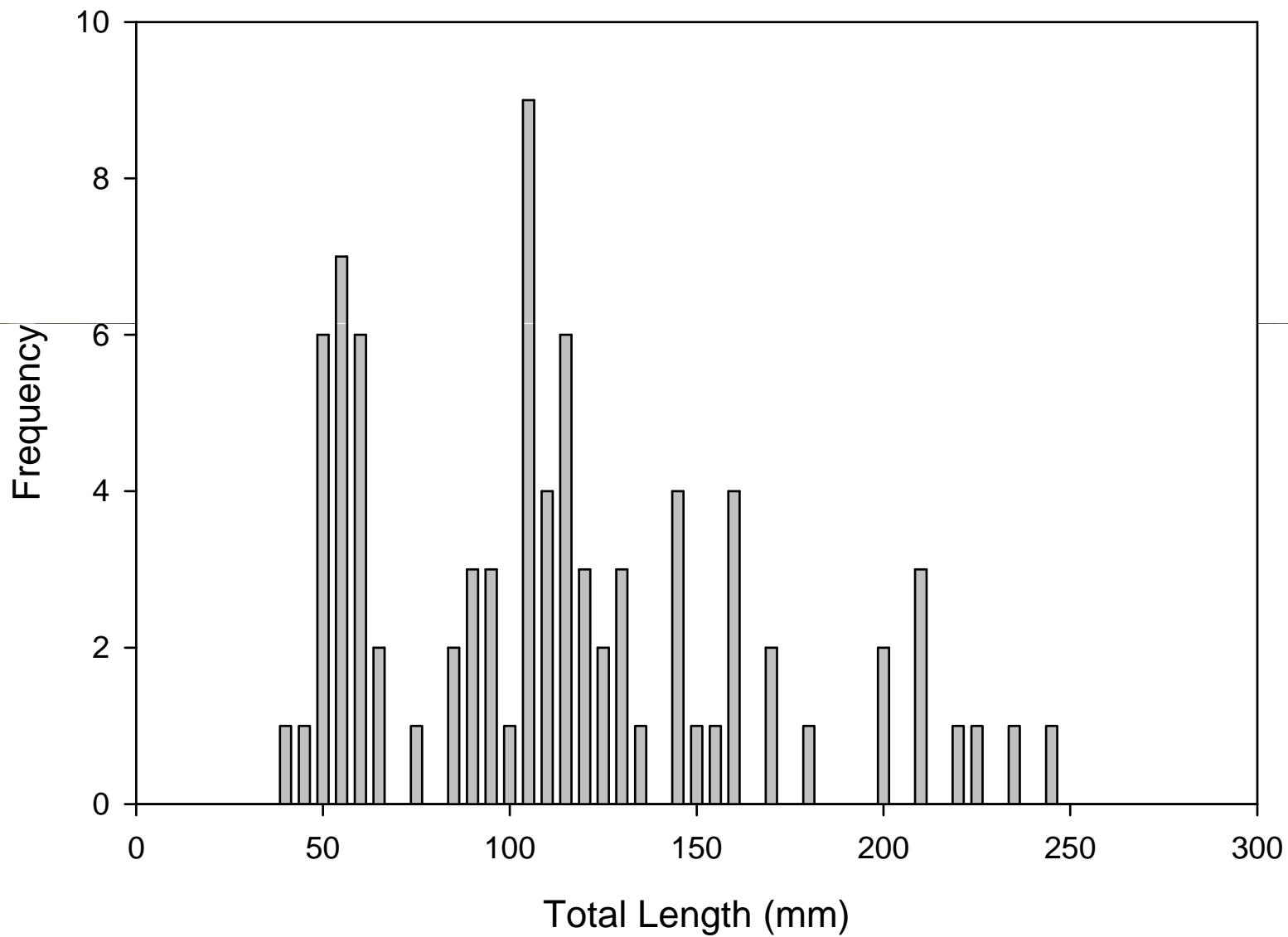


# Data Analysis – Length Data

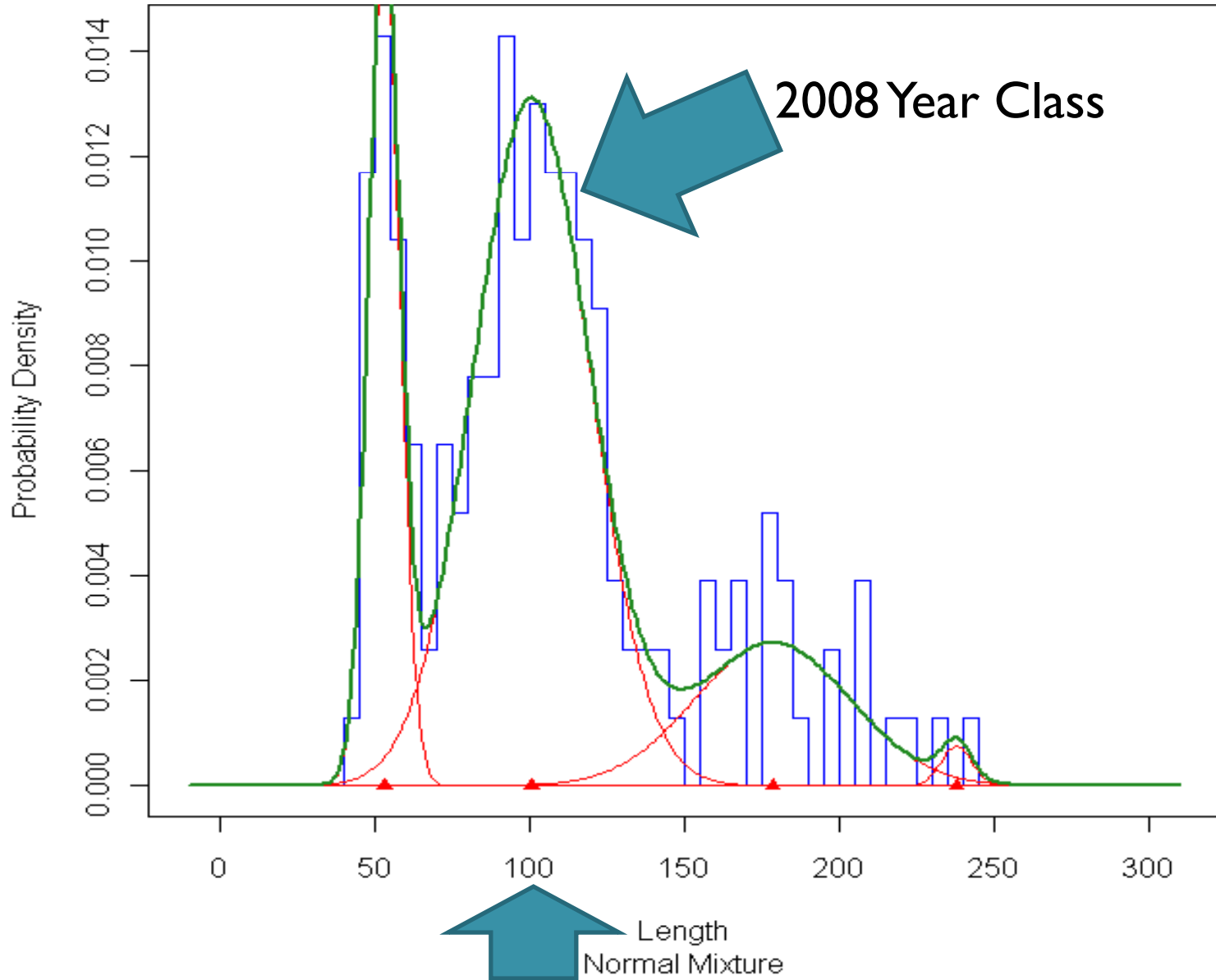
- MIXDIST package in R
  - Age structure and age-specific size distributions.
  - Estimates average size-at-age and standard deviation from collective length data.
  - Assumed 4 age classes in collection (0 – 3)



# Histogram of Lengths in Collections



# Results from MIXDIST





# Power Analysis

- Use data from known (NAT, PB) strains to generate hypothetical populations:
  - Parent generation
  - F1, F2 generations
  - back-crossings
  - HYBRIDLAB, Nielson et al. 2001
- Analyze simulated populations using Structure, determine frequency of missed assignments.



# Power Analysis

- Correct identification of parent generations (NAT, PB strains): 100%

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- Correct identification of F1, F2, 1<sup>st</sup> B-C generations as hybrids: ~100%
- With our sample sizes, we would detect introgression as low as 3% of population with 95% percent accuracy.





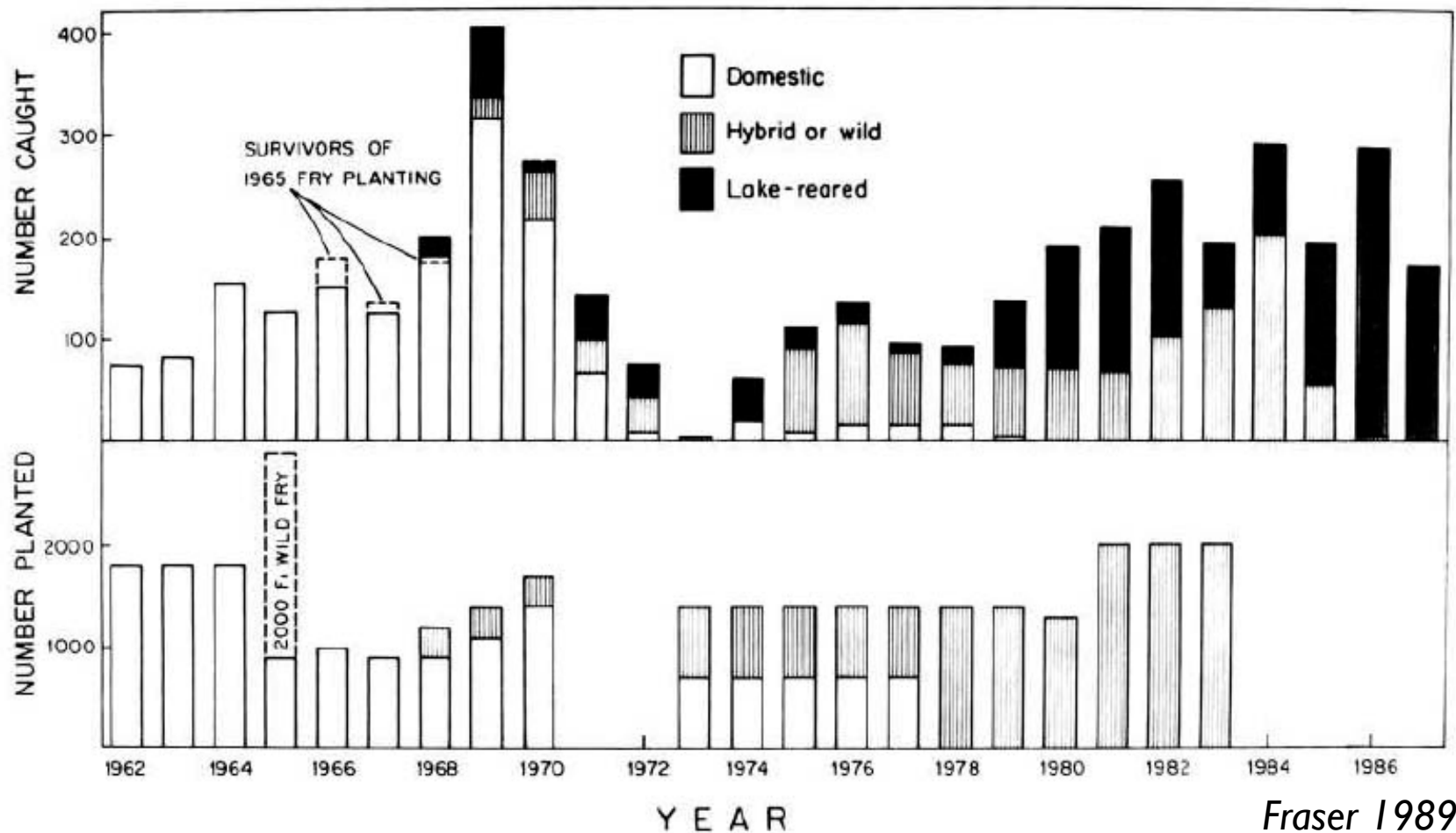
# Hatchery supplementation and native brook trout

- Previous studies mixed, but generally suggest levels of introgression between native and hatchery brook trout are low.
  - Possible exception: hybridization between northern / southern strains, Great Smokey Mountains



# How to explain low introgression despite widespread stocking?

- Low fitness of hatchery stocks



Fraser 1989



# How to explain low introgression despite widespread stocking?

- Genetic differences between native and hatchery lineages may be small (difficult to distinguish strains)
- Technology may be inadequate to detect introgression (poor “markers”)
- Context-dependence; different strains, different systems