

Noninvasive Genetic Techniques and its Application in Wildlife Conservation



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Lecture Structure

- Introduction
- Sources and methods of collecting samples
- Applications
- Pitfalls
- How to avoid pitfalls

Introduction

- Historically, molecular techniques developed in the human genetics community
- Over time adopted by the wildlife community

Introduction

- Molecular studies in wildlife populations
 - Ecological
 - Evolutionary
- Assessing the genetics of wild populations 30 or 40 years
 - required large amounts of fresh tissue
 - animals were often killed
 - widely used for birds, rodents, insectivores
 - techniques fell under public scrutiny

Introduction

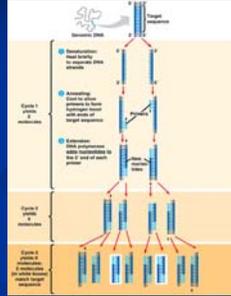
- First alternative was blood samples
 - used to study serum proteins
 - did not provide a lot of genetic information
- Recent advancement in molecular techniques offer promise for future research
 - microsatellites
 - Polymerase Chain Reaction (PCR)

Introduction

- Microsatellites
 - widely distributed in the genome making it easy to develop a suite of markers available for genetic studies
 - highly variable, which provides unique fingerprint
 - Can be amplified using PCR

Introduction

- PCR
- Tiny samples collected from hair, feathers, feces
- Uses a series of heating and cooling steps with a mixture of enzymes, base pairs, and template strands to replicate
- 20 million copies in 3 hours !



Sources and Collection Methods

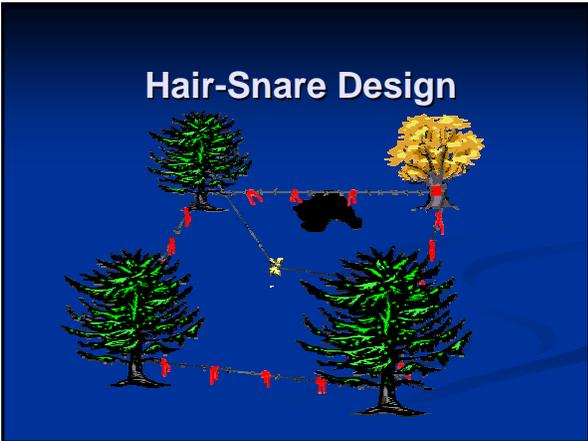
Hair-Snare Design



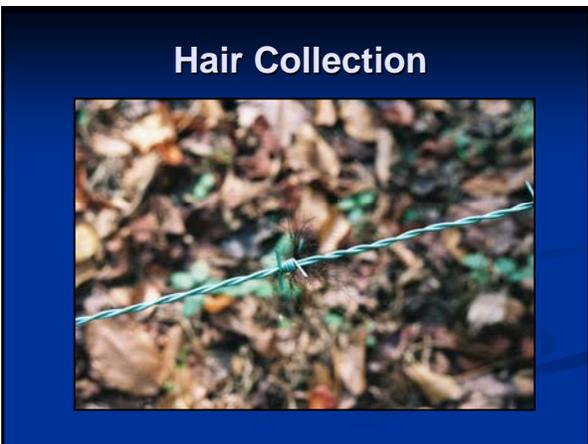
Hair-Snare Design



Hair-Snare Design



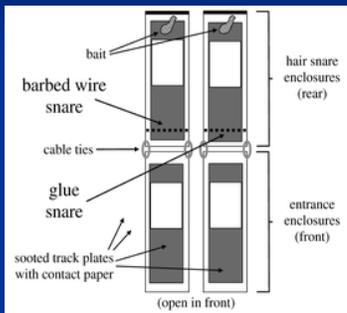
Hair Collection



Wire and Glue Hair Snares



Wire and Glue Snare



Collecting Hair – Ground Dwellers



Collecting Hair – Tree Dwellers



Feces

- Collected
 - On roads
 - In the woods
 - Edges
 - River banks



Feathers and Egg Shells



Skin



- Pods are located
- Researchers wait for activities such as breaching and tail slapping
- Results in sloughed skin

Skin



Saliva



Regurgitates

- Found at roosting sites of owls



Urine

- Wolf and wolverine studies



Applications

Abundance Estimation

- Obtaining reliable results from mark-recapture methods based on live trapping is challenging
 - low capture probabilities
 - capture biases
- Unique genotypes can be used in traditional mark-recapture studies
- Advantages
 - increased capture probability
 - tag permanency
 - decreased intrusiveness
 - reduced bias

Abundance Estimation

- Romain-Bondi et al. (2004) studied endangered grizzly bear population in Washington
 - 6 bears (90% CI =1-27)
 - implications are natural recovery is very unlikely
 - recommend population augmentation



Genetic Diversity

- Important factor in long term population persistence
- Allows species to respond to environmental change
- Small, isolated populations may experience inbreeding depression
 - cheetah
 - Florida panther

Genetic Diversity

- Eastern Slopes Grizzly Bear Project
- Genetic variation in the Bow River Watershed
- Found little maternal diversity compared to the Northern Continental Divide population
- Overall genetic variation was good
- Suggests males travel far enough to provide variation

Gene Flow

- Gene flow- movements of individuals from one location to another and their subsequent integration into the gene pool of their new locality
- increasingly isolated populations may experience reduced gene flow, thus decreased genetic variation

Gene Flow



Demographic History

- Patterns of population expansion and decline
- Culver et al. 2008
 - Population size for Florida panther mid 20th century?
 - Genetic variation 1890s vs. 1990s
 - Found threefold decrease in genetic diversity
 - Used models to estimate effective population required to account for the decline



Population of Origin

- Assignment test - Uses allele frequencies to assign individuals to their most likely source population
- Blanchong et al. (2002)
 - Used genotypic data to confirm that Michigan deer infected with tuberculosis were harvested in area assumed to be free of the disease

Hybridization

- Hybridization can be a problem, especially when populations are small and are in close proximity to a similar species.
- If hybrids are fertile, continued hybridization could lead to genetic swamping

Hybridization

- 1973 red wolf received protection under the Endangered Species Act
- Red Wolf Recovery Program recognizes hybridization as the primary threat to recovery of the species



Hybridization

- Adams et al. (2003) – used fecal DNA to monitor hybridization between red wolves and coyotes
 - gave a better understanding of the amount of hybridization and the spatial distribution of wolves and hybrids
 - provided an important tool for curbing hybridization in the future

Others

- Phylogeography
- Identification of Sex
- Parentage
- Population Structure
- Landscape Genetics
- Dispersal and Immigration
- Migratory Movements
- Forensics
- Dietary Studies
- Documenting Rare Species
- Conservation Genetics

Pitfalls

- Amount of DNA collected from free ranging animals is often very low
- Allelic dropout is the amplification of only 1 of 2 alleles in a heterozygote pair
 - produces false homozygotes
 - may interpret as new captures
 - can significantly bias estimates

Pitfalls

- False alleles are a result of artifacts created during amplification
 - can be misinterpreted as true alleles
 - homozygous individual may be recorded as a heterozygote and identified as a new capture
 - McKelvey and Schwartz (2004) found allelic dropout caused a 5.5 fold increase in wolf abundance estimates

Pitfalls

- Shadow effect – when 2 or more individuals share the same genotype at the same loci
 - misclassification of unique individuals
 - negatively bias abundance estimates
 - occurs when too few loci are examined
 - when low heterozygosity loci are used

How to Avoid Pitfalls

- First thing first
 - differences exist among species
 - wolf feces provides much more DNA than bear feces
 - generalizations can be hazardous
 - thoroughly research literature
- Do trial test before diving into experiment
 - Determine what method provides the best results to reach your scientific goal

Questions ?