Molecular Tools Used in Population Structure and Tracking

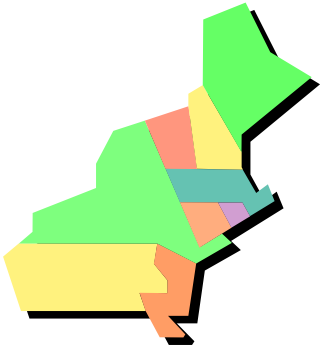
Molecular Ecology

GIS

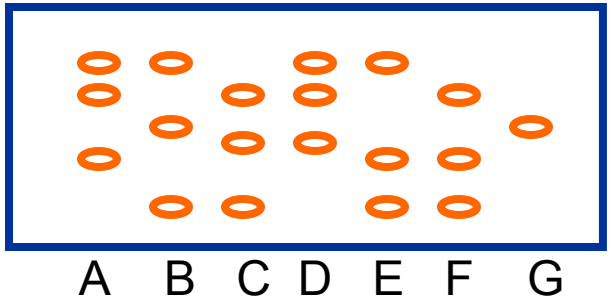
1990's: Geographic Information Systems

2000's: Genetic Information Systems

Mapping Landscapes



Mapping Genes



Concepts

The Big Picture

Genome: all of the genetic information contained in an organism.

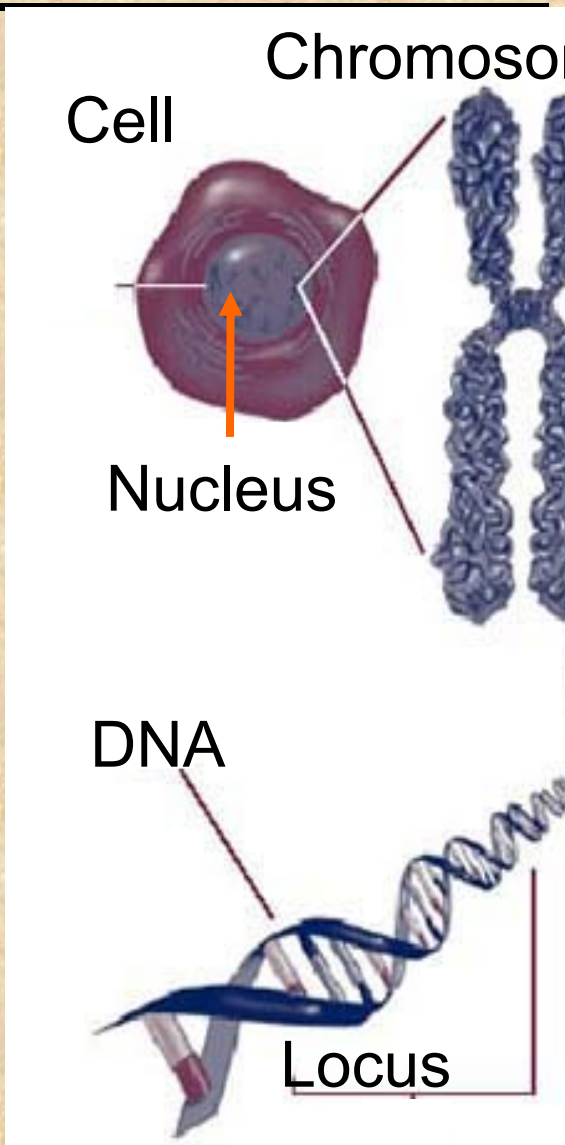
Each cell in an organism has a complete copy of the entire genome.

Genomes are made of chromosomes

Chromosomes: large physical aggregations of DNA found within cells.

Locus: specific location on a chromosome

Alleles: specific DNA sequence variants observed at loci.



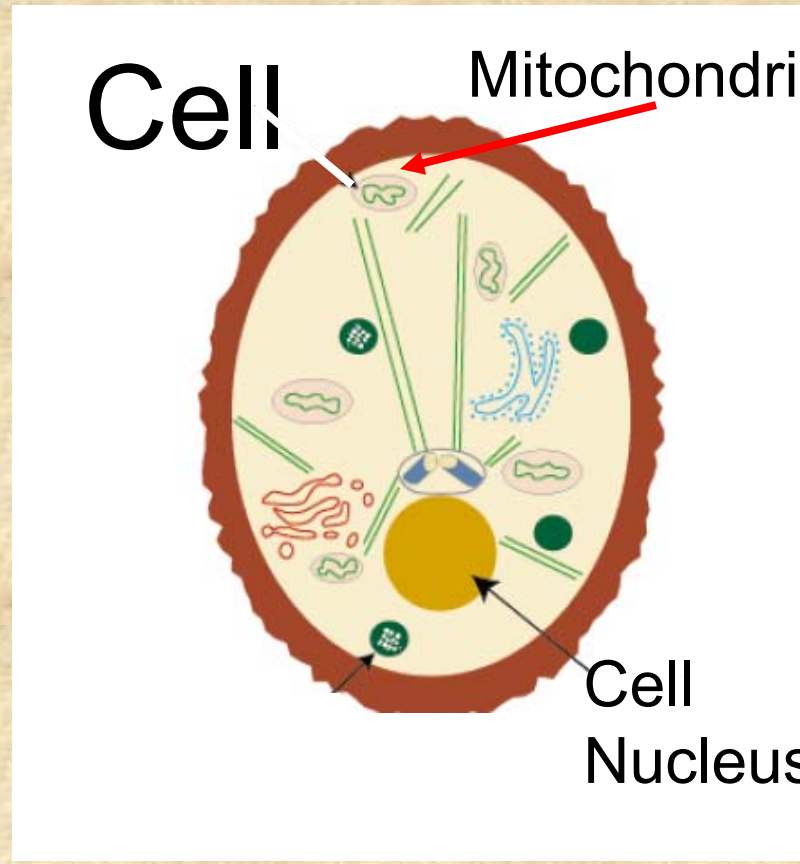
Animals Have Two Genomes

uclear

- Genomes are large: 10^6 to 10^{11}bp.
- Diploid (two alleles/locus)
- Variable number of chromosomes
- Bi-parentally inherited
- Does not evolve as quickly mtDNA

rganelle: chloroplasts or mtDNA.

- Small number of genes
- Haploid—one copy
- Only 1 chromosome
- Usually maternally inherited.
- Evolves quickly: 5x as fast as nuclear DNA.



Ultimately....

We are trying to:

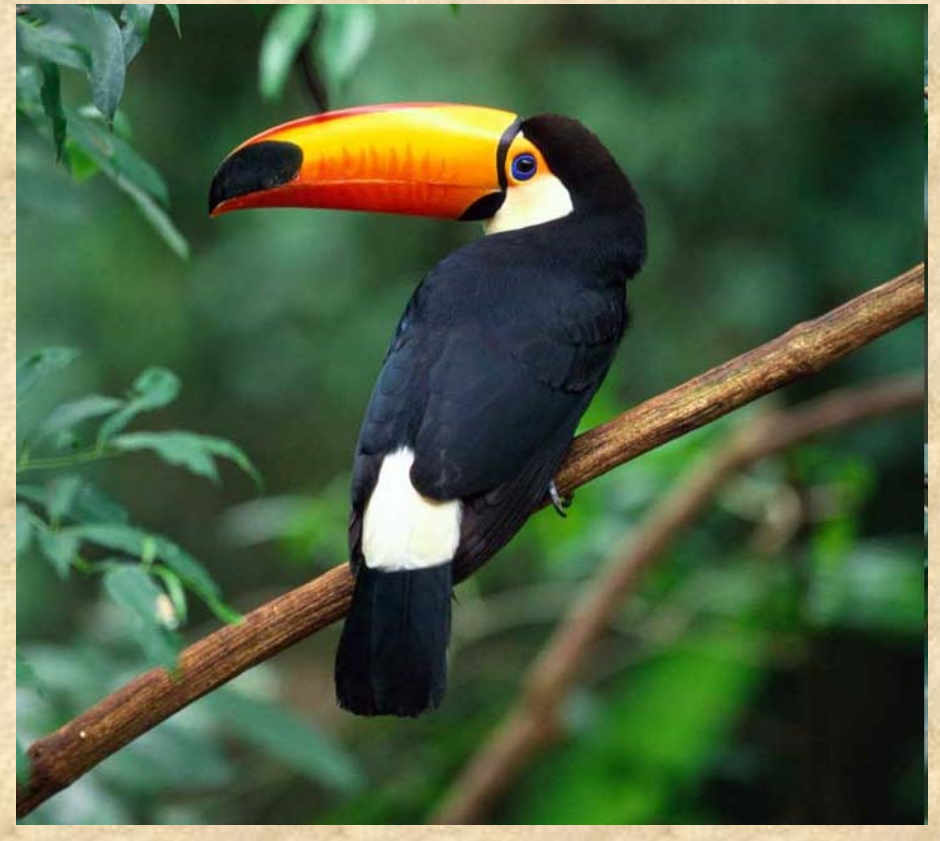
Characterize Variation Among ALLELES

At Multiple LOCI

Across Multiple CHROMOSOMES

So that we can make inferences about GENOMES

Molecular Markers



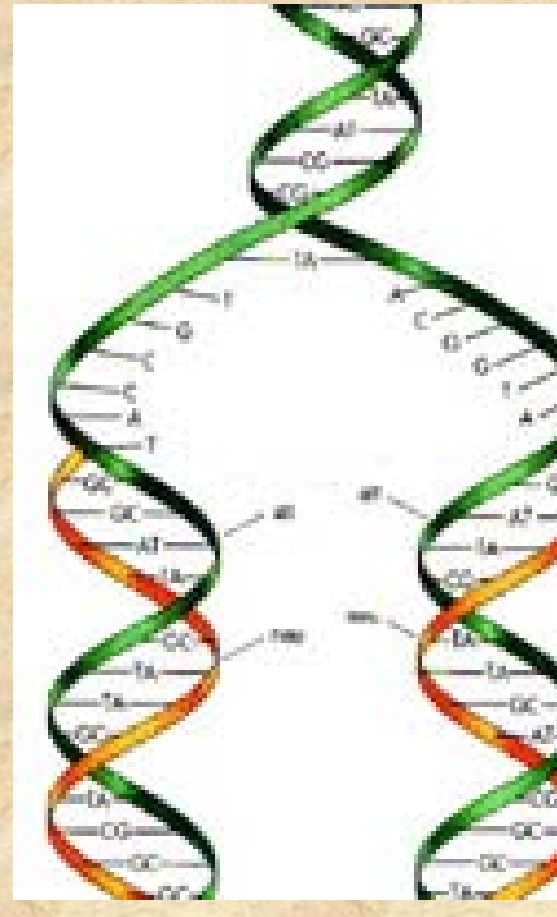
Common Molecular Markers

- **mtDNA sequences**
- **Microsatellites**
- **AFLP's: Amplified Polymorphic Loci**

Polymerase Chain Reaction

Revolutionized molecular biology (Mullis 1987).

Creates (amplifies) more copies of DNA from a targeted region of DNA so molecular analyses can be carried out.



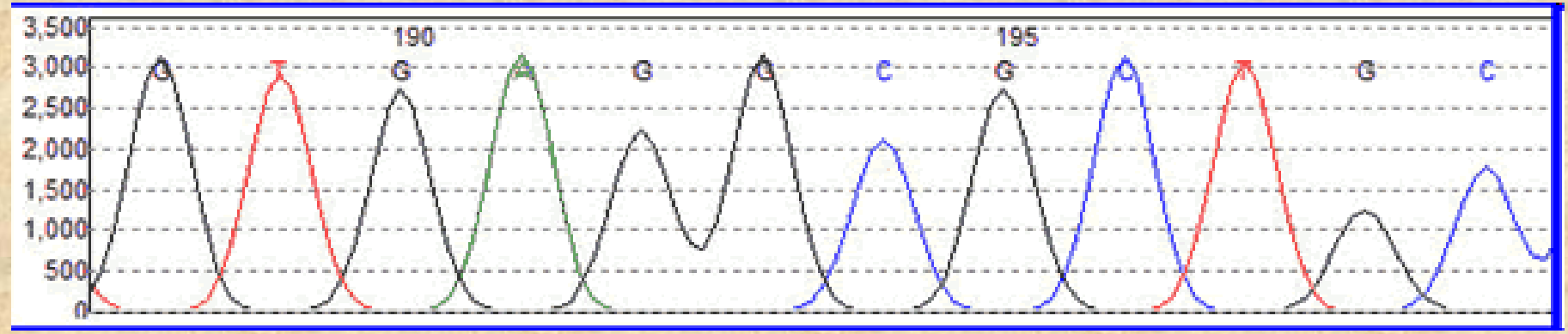
Four steps:

1. Extract: DNA
2. Denature: split strands of DNA by heating.
3. Anneal: an enzyme is used to make a complementary strand of DNA from each original strand (see orange).
4. Primer extension: complimentary strands are synthesized so there are now 2 copies of DNA. Process is repeated thousands of times.

mtDNA Sequencing

Directly sequence fragments of DNA—ultimate approach for characterizing genetic variation.

Data come as chromatograms which provide information on the order of nucleotides in a given allele: ACTG…



Can be used intra or interspecifically: phylogenetics, population structure, phylogeography.

Control region: population level

Cytochrome b: interspecific level

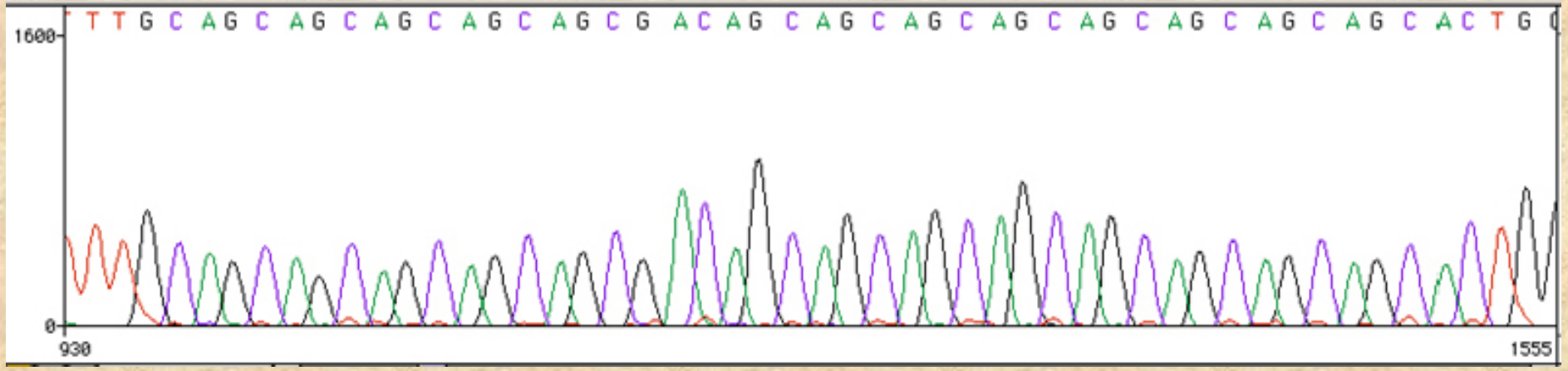
Microsatellites

Used to look at parentage, pedigrees, population structure, forensics.

They are simple sequence repeats (SSR's) of 1-6 bp where variation in number of repeats = locus heterogeneity.

GCGCGCGC vs GCGCGC

Variable loci can be problematic to isolate in some taxa (i.e., birds) --need species specific primers.



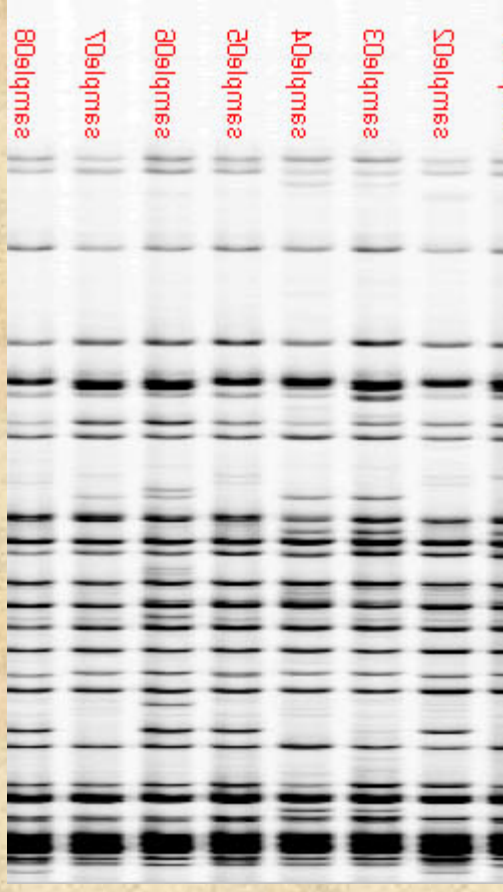
Amplified Fragment Length Polymorphism

AFLP's are fast, cheap, efficient DNA fingerprints.

They are Dominant markers used in population structure, species identification questions.

Use random primers, can generate lots of bands.

But…primers are not species-specific: So you have to be sure you know whose DNA you are sampling—contamination can be a problem.



Molecular Sampling

Tissues to Sample: Direct vs Indirec

Sex Your Birds!!

The benefits to your research are **HUGE!**

Sending samples to a lab for molecular sexing is:

A. cheap: $15-20/sample
B. fast: ~3 days
C. easy: mail blood or DNA to:

Zoogen Incorporated
Davis, CA
www.zoogen.biz

Molecular Markers Used in Conservation

	Parent	Ped	Pop. str	Pop. Diff.	Intersp.
licrosatellites	X	X	X	X	
FLP			X	X	X
tDNA			X	X	X

General Sampling Guidelines for Genetic Screening

nique	Parentage/Pedigree 20+ complete families	Pop. structure/Pop. Diff. 20 ind./pop, 5+ pop.	Hybrid/Phyloge 5-10+ind./group/ou
crosatellites	5 -10 variable loci	5 -10 variable loci	
LP	20+ variable bands	20+ variable bands	20+ variable bands
DNA	N/A	400-800+ base pairs	400-800 base pairs

Approximate Costs and Time Involved with Various Molecular Techniques

Technique	Cost	Time Commitment
DNA extraction	$3/sample	Short--week
AFLPs	$5/sample/primer	Medium--months
mtDNA	$20/sample	Medium--months
Microsatellites	$3/sample/locus[2]	Can be long--many months

[1]These costs do not include the much more expensive screening process and personnel.

[2]Must be multiplied by # loci.

Tracking Birds

Population Identification

	Pop A	Pop B
1.	10	0
2.	10	0
3.	0	10
4.	0	10
5.	10	0
6.	0	10

Population/Taxon specific markers

	Pop A	Pop B
1.	8	4
2.	10	2
3.	7	8
4.	2	8
5.	1	10
6.	3	8

Assignment Test

Prob (A): 0.8
Prob (B): 06

Hudsonian Godwit

Semipalmated Plovers

Willow Warblers

Stefan Bensch et al.

Summary

Genetics and Molecular Tools are now an every day aspect of our lives. Thus, we need to have at least a working understanding of them.

Using molecular markers to track birds can be problematic.

Careful screening of markers can yield better success in Tracking birds than traditional screening.