

M.Sc ZOOLOGY

SEMESTER 2

PAPER CC 8

MOLECULAR-TAXONOMY

Dr.Anjali Gupta

Associate professor

Department of zoology

H.D.Jain College

ARA

Molecular taxonomy

The classification of organisms on the basis of the distribution and composition of chemical substances in them for the use of molecular genetics, to study the evolution of relationships among individuals and species. It involves comparing the sequences of functionally homologous molecules from each organism to determine the number of difference is between them. The greater the number of differences the most distantly related the organisms are likely to be.

Molecular techniques in the field of biology have helped to established genetic relationship between the members of different taxonomic categories

Molecular Phylogenetic

The study of evolutionary relationship among biological entities (individuals, population, species for higher taxa), by using a combination of molecular data (such as DNA and protein sequences, presence or absence of transposable elements and gene order data) and statistical techniques

Fitch and Margoliash, (1967) made first phylogenetic tree based on molecular data

Phylogenetic tree

- This tree was close to the already established phylogenetic tree
- The taxonomist realized significance of molecular data and this made them understand that other traditional methods are all though important but molecular evidences could be final or confirmatory evidences
- Phylogenetic studies assess the historical processes which effect relationships and phylogeographic studies asses the geographical distribution.
- These two studies started with the introduction of **mDNA markers** in population genetics analysis.

Objectives of molecular taxonomy

- Reconstruct the correct Genealogical ties among biological entities.
- To estimate the time of divergence between biological entities
- Chronicle the sequence of entities along evolutionary lineage

Tools of molecular taxonomy

Molecular taxonomy uses a variety of techniques to derive phylogenetic trees.

- **Genetic markers** are used to make inferences about relationship between environment and morphology, well as physiology and behaviour.
- **Polymerase chain reaction (PCR)** is used to investigate variations of DNA on large scale.
- Genetic amplification is also fundamental two new approaches to **DNA fingerprinting**.
- **DNA Barcoding:** PCR amplification and sequencing of genetic marker.
- Chromosome pairing.

Molecular markers: It can be categorized as type 1 and type II marker

1. **Type 1** markers are associated with genes of known function.
2. **Type II** markers are associated with genes of unknown function.

Allozyme: These are type 1 marker as the protein they encode are associated with same function. Allozyme electrophoresis is a method which can identify genetic variation at the level of enzymes that are directly encoded by DNA. Protein variants called Allozyme originate from allelic variants and they will differ slightly in electric charge. Allozyme are codominant markers having been expressed in heterozygous individuals in a Mendelian way.

Mitochondrial DNA marker

- Non nuclear DNA i.e. mitochondrial DNA in the cells having located within mitochondria in cytoplasm. It is maternally inherited with a haploid genome.
- The entire genome undergoes transcription as one single unit. They are not subjected to recombination and thus they are homologous markers.

Microsatellites: It is a simple DNA sequence which is repeated several times across various points in the DNA of the organism. These repeats are highly variable and these loci can be used as markers

Single nucleotide polymorphisms (SNPs):

SNPs arise due to single nucleotide substitution (transversion/ transition) or single nucleotide insertion /deletion. These point mutations give rise to two different alleles with alternative bases at a particular nucleotide position. They are most abundant polymorphism in the genome coding or non coding of any organisms. These can be detected using PCR, microchip array or fluorescence technology

Arbitrary nuclear DNA markers

These are used when we target a segment of DNA of unknown function. The widely used method of amplifying unknown regions or RAPD(random amplified polymorphic DNA) and AFLP(amplified fragment length polymorphism DNA).

Specific nuclear DNA marker:

- **VNTR (Variable Number of Tandem Repeat)** is a segment of DNA that is repeated tens or even hundreds to thousands of times in nuclear genome. They repeat in tandem, vary number in different loci and differently in individuals.

There are two main classes of repetitive and highly polymorphic DNA

1. **Minisatellite DNA** referring to genetic loci with repeats of length 9-65 bp.
2. **Microsatellite DNA** which repeats of length 2-8 bp long.

Microsatellites are much numerous in the genome of vertebrates then Minisatellite.

- **ESTs (Expressed Sequence Tags)**

ESTs are single-pass sequences which were generated from random sequencing of cDNA clones full stop it can be used to to identify genes and analyze their expression by means of expression analysis. These are most valuable for linkage mapping.

Applications of molecular taxonomy

It helped in cleaning up many taxonomical problems.

SPECIES	TAXONOMICAL PROBLEM	TECHNIQUE
1. Amphiprion sebae	Ratified taxonomic status	RAPD
2. Four species of Clown fishes	Revealed phylogenetic relationship	RAPD
3. Indian Mackerel Population	Revealed genetic Homogeneity	Multi technique approach
4. Dolphins, Porpoise, whales and Dugony	Developed the capability of accurate identification	Mitochondrial DNA sequence

Advantages and disadvantages of molecular taxonomy

Advantages: Using molecules is advantageous for the following two reasons

1. Closer to actual level of heredity (especially if DNA sequences is used).
2. The number of independently varying characters is huge. Each nucleotide position can be considered a character and assumed independent theoretically. The DNA of any given organism has millions to billions of nucleotide positions

Disadvantages:

1. Homoplasy likely to occur at a higher rate in nucleotide sequences than in morphological characters.
2. Homology among characters (nucleotides) is sometimes not easily assessed.
3. They also have high cost and intensive training line.