Journal of the West
ISSN: 0022-5169 / **E-ISSN:** 1930-0115
https://journalofthewest.com/jw

Molecular Phylogenetics and Evolutionary Relationships Among Freshwater Fish Populations

Prof. Ricardo Almeida
Department of Climate Finance, São Paulo Global University, Brazil

Submission Date: 10.06.2025 | Acceptance Date: 01.09.2025 | Publication Date: 19.02.2026

Abstract
Understanding evolutionary relationships among freshwater fish populations is essential for biodiversity assessment, conservation planning, and taxonomic clarification. Traditional morphological classification has often been limited by phenotypic plasticity and convergent evolution, leading to uncertainties in species boundaries and lineage divergence. Molecular phylogenetics provides a powerful framework for resolving these ambiguities by analyzing genetic variation at the DNA level. the application of molecular markers, including mitochondrial DNA sequences such as cytochrome b and cytochrome oxidase I, along with nuclear gene markers, to reconstruct phylogenetic relationships among selected freshwater fish populations. Comparative sequence analysis and phylogenetic tree construction reveal patterns of genetic divergence, historical dispersal, and population structuring across different river basins. The results indicate significant genetic differentiation among geographically isolated populations, suggesting the influence of vicariance events, habitat fragmentation, and historical climatic fluctuations on evolutionary trajectories. Molecular data further highlight instances of cryptic speciation and previously unrecognized lineages within morphologically similar taxa. Genetic distance measures and haplotype diversity analyses provide insights into population connectivity and gene flow. These findings underscore the importance of integrating molecular tools with ecological and morphological data to achieve accurate taxonomic resolution.
Keywords: Molecular Phylogenetics, Freshwater Fish, Genetic Diversity, Mitochondrial DNA

Introduction
Freshwater ecosystems support a remarkable diversity of fish species, many of which exhibit complex evolutionary histories shaped by geological events, climatic fluctuations, and hydrological changes. Rivers, lakes, and drainage basins often function as natural barriers, restricting gene flow and promoting population divergence. As a result, freshwater fish populations provide valuable models for studying speciation, adaptation, and evolutionary processes. Accurate understanding of their evolutionary relationships is essential not only for taxonomy but also for conservation and sustainable management. Traditional classification of freshwater fishes has largely relied on morphological characteristics such as body shape, fin structure, scale patterns, and coloration. While morphology remains important, it may not always reflect true evolutionary relationships. Phenotypic plasticity, environmental influences, and convergent evolution can lead to misidentification or underestimation of species diversity. In many cases, morphologically similar populations may represent genetically distinct lineages,

often referred to as cryptic species. Molecular phylogenetics has revolutionized evolutionary biology by enabling researchers to analyze DNA sequence variation to infer lineage relationships. Genetic markers, particularly mitochondrial DNA regions such as cytochrome b and cytochrome oxidase I, along with nuclear genes, have become standard tools for reconstructing phylogenetic trees and assessing genetic diversity. These molecular approaches allow for precise estimation of divergence times, historical dispersal patterns, and population connectivity across geographic regions. Freshwater fish populations are especially vulnerable to habitat fragmentation, pollution, overexploitation, and climate change. Molecular data provide critical insights into population structure and gene flow, helping identify evolutionarily significant units for conservation. By integrating genetic, ecological, and morphological evidence, researchers can achieve a more comprehensive understanding of biodiversity patterns.

Genetic Markers Used in Phylogenetic Studies

Genetic markers are fundamental tools in molecular phylogenetics, allowing researchers to trace evolutionary relationships, assess genetic diversity, and understand population structure. In freshwater fish studies, both mitochondrial and nuclear DNA markers are widely employed because they provide complementary information. The choice of marker depends on the research objective, whether it involves deep evolutionary divergence, recent population differentiation, or fine-scale genetic variation.

Mitochondrial DNA Markers (Cytochrome b, COI)

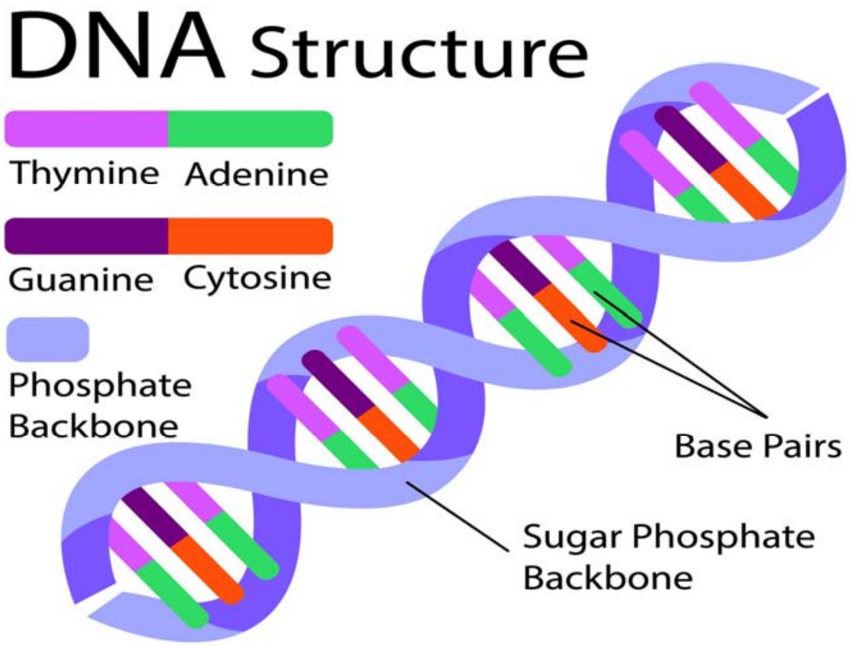
Mitochondrial DNA (mtDNA) is one of the most commonly used genetic markers in phylogenetic research. It is maternally inherited, lacks recombination, and evolves relatively rapidly compared to nuclear DNA, making it suitable for studying species-level relationships and population divergence.

Cytochrome b is a protein-coding gene widely used in fish phylogenetics due to its moderate evolutionary rate. It provides valuable information on genetic distances and lineage divergence among closely related species.

Cytochrome oxidase I (COI) is extensively used in DNA barcoding initiatives to identify species and detect cryptic diversity. The standardized COI region enables comparison across broad taxonomic groups and facilitates accurate species identification in biodiversity assessments.

Mitochondrial markers are particularly effective in reconstructing maternal lineages and understanding historical dispersal patterns across river systems.

Nuclear DNA Markers



Nuclear DNA markers complement mitochondrial data by providing biparentally inherited genetic information. Unlike mtDNA, nuclear genes undergo recombination and often evolve more slowly, making them useful for resolving deeper phylogenetic relationships and detecting hybridization events.

Examples include ribosomal RNA genes and various protein-coding nuclear genes. Nuclear markers help confirm evolutionary hypotheses derived from mitochondrial data and reduce biases associated with single-locus analysis. They are particularly important in cases where introgression or incomplete lineage sorting may complicate phylogenetic interpretation.

Microsatellites and SNPs

Microsatellites and single nucleotide polymorphisms (SNPs) are highly informative markers for population-level studies.

Microsatellites consist of short, tandemly repeated DNA sequences that exhibit high levels of polymorphism. Their variability makes them ideal for assessing genetic diversity, parentage analysis, and fine-scale population structure.

SNPs represent single base-pair variations across the genome and are abundant throughout nuclear DNA. High-throughput sequencing technologies have enabled large-scale SNP analysis, providing detailed insights into gene flow, local adaptation, and demographic history. These markers are particularly valuable for conservation genetics, as they help identify distinct population units, measure genetic connectivity, and inform management decisions.

mitochondrial DNA markers, nuclear genes, microsatellites, and SNPs offer a comprehensive toolkit for investigating evolutionary relationships among freshwater fish populations. Integrating multiple marker types enhances phylogenetic accuracy and provides a deeper understanding of biodiversity and evolutionary processes in aquatic ecosystems.

Conclusion

Genetic markers have become indispensable tools in molecular phylogenetic research, particularly in the study of freshwater fish populations. Mitochondrial DNA markers such as cytochrome b and COI provide valuable insights into species identification, lineage divergence, and historical biogeographic patterns. Their relatively rapid rate of evolution makes them especially useful for resolving relationships among closely related taxa and detecting cryptic diversity. Nuclear DNA markers complement mitochondrial data by offering biparental inheritance patterns and greater resolution for deeper evolutionary relationships. They help clarify complex phylogenetic scenarios, including hybridization events and incomplete lineage sorting, which cannot be fully resolved through single-locus analysis alone. Microsatellites and SNPs further enhance our understanding of fine-scale population structure, gene flow, and genetic diversity. These highly polymorphic markers are particularly important in conservation genetics, where identifying distinct management units and assessing genetic health are essential for sustainable resource management. integrating multiple genetic markers strengthens phylogenetic inference and improves the accuracy of evolutionary interpretations. Such comprehensive molecular approaches are vital for clarifying taxonomy, understanding diversification processes, and guiding conservation strategies in freshwater ecosystems that face increasing environmental pressures.

Bibliography

Avise, J. C. (2000). *Phylogeography: The History and Formation of Species*. Harvard University Press.

Avise, J. C. (2004). *Molecular Markers, Natural History and Evolution* (2nd ed.). Sinauer Associates.

Hebert, P. D. N., Cywinska, A., Ball, S. L., & deWaard, J. R. (2003). Biological identifications through DNA barcodes. *Proceedings of the Royal Society B: Biological Sciences, 270*(1512), 313–321. https://doi.org/10.1098/rspb.2002.2218

Kocher, T. D., Thomas, W. K., Meyer, A., Edwards, S. V., Pääbo, S., Villablanca, F. X., & Wilson, A. C. (1989). Dynamics of mitochondrial DNA evolution in animals: Amplification and sequencing with conserved primers. *Proceedings of the National Academy of Sciences, 86*(16), 6196–6200. https://doi.org/10.1073/pnas.86.16.6196

Nei, M., & Kumar, S. (2000). *Molecular Evolution and Phylogenetics*. Oxford University Press.

Sunnucks, P. (2000). Efficient genetic markers for population biology. *Trends in Ecology & Evolution, 15*(5), 199–203. https://doi.org/10.1016/S0169-5347(00)01825-5

Tamura, K., Nei, M., & Kumar, S. (2004). Prospects for inferring very large phylogenies by using the neighbor-joining method. *Proceedings of the National Academy of Sciences, 101*(30), 11030–11035. https://doi.org/10.1073/pnas.0404206101

Ward, R. D., Zemlak, T. S., Innes, B. H., Last, P. R., & Hebert, P. D. N. (2005). DNA barcoding Australia's fish species. *Philosophical Transactions of the Royal Society B: Biological Sciences, 360*(1462), 1847–1857. https://doi.org/10.1098/rstb.2005.1716