



## Abstract Genetic and Morphological Identification of Formalin Fixed Larval Fishes: How Long Is Too Long?<sup>+</sup>

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Abstract: Identification of larvae of fish is usually based on assembling a developmental series of wild-collected larvae, usually formalin preserved, using pigmentation patterns, morphology and fin meristics. However, for many species, larvae are still undescribed, or there are only limited descriptions of larvae development. Formalin fixation of larval fish was previously thought to prevent genetic sequencing compared to ethanol-preserved larvae. In this poster, we detail the results of an integrative taxonomic approach based on morphology, imaging and DNA barcoding of the mitochondrial (mtDNA) cytochrome c oxidase subunit (COI) gene. We used this approach in both cultured yellow tail kingfish, Seriola lalandi and wild-sourced fish larvae fixed in 5% formalin. DNA barcoding and genetic species identification were 100% successful in S. lalandi fixed in formalin for up to 6 months, while barcoding of wild-caught fish larvae enabled species identification of 93% for up to 8-week formalin-fixed specimens. While COI genetic identifications from the in-field experiments were patchier than the controlled experiments, our study highlights the possibility of recovering suitable DNA from formalin-fixed larvae for up to six months. This was achieved by applying DNA extraction methods that use de-cross-linking steps and species identification based on both full-length reference and mini-barcodes. Our study provides a practical framework for undertaking both morphological and genetic identification to document the larval development of previously undescribed species from historic and current formalin-fixed samples collected around southern Australia.

**Keywords:** fish larvae; southern Australia; cytochrome c oxidase subunit I (COI) barcoding; formalin fixation

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