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DNA TYPING ACROSS TEN TILAPIA SPECIES USING CYTOCHROME C OXIDASE SUBUNIT I (COI)





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INTRODUCTION

The tilapias are a group of African and Middle Eastern cichlid fish that are widely cultured in both developed and developing countries (major producers include China, Egypt, Indonesia, Philippines, Thailand and Brazil), with total world aquaculture production of 4.5 million t and total value of 7.6 billion USD in 2012 (FAO, 2014). Of this, 3.8 million t was O. miloticus, representing 84.13% of the total. With many different species and sub-species of tilapia, and extensive use of interspecies have been superiorated in aquaculture and wild populations where productions have occurred. Mitochondrial DNA cytochrome c oxidase subunit 2 (1) sequence is widely used as a "barcode of life" for species identification. The conserved sequence of the 5' region of the mitochondrial gene cytochrome oxidase subunit 1 (COI or Cox1) has been widely used for distinguishing, for example Australian fish (Ward et al., 2005), marine fishes (McCusker et al., 2013) and tilapia species (Wu ft Yang, 2012).

OBJECTIVES

- To confirm species authentication in tilapia using cytochrome c oxidase subunit (COI).
- To construct a gene tree based on COI nucleotide sequences between tilapia species

MATERIALS AND METHODS

This study involved ten species of tilapia from all three common genera, and included more than one population and/or subspecies from the major commercial species. It was undertaken in parallel to research to develop species-specific markers from nuclear DNA using double-digest restriction site associated DNA sequencing (ddRADseq).



ten different tilapia species. The O. niloticus samples consisted of two sub-species (O. n. miloticus and 0. n. cancellatus) from two populations in the former case; O. aureus, O. mossambicus and Tilapia zillii (Gervais) comprised samples from two populations each, while 0. karongae (Trewavas), O. urolepis hornorum (Norman; originally from Tanzania), O. andersonii, O. macrochir, Sarotherodon galllaeus (Linnaeus) and S. melanotheron consisted of one population each.

METHODOLOGY



Collection of Samples Purification and Sequencing



Polymerase Chain Reaction

Data Analysis

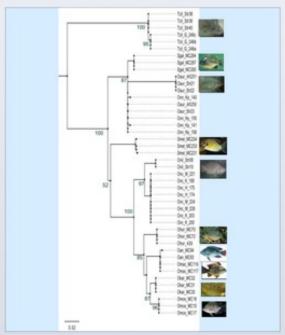
A 655 bp of the CO-I gene from mitochondrial DNA were amplified with primer pairs from Ward et al. 2005.

ACKNOWLEDGMENTS

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RESULTS

The COI partial sequences of the tilapia 13 es that were retrieved varied between 395-631 bp and agreed with those in the Barcode of Life Data System (BOLD) and the NCBI GenBank Database. The COI gene tree indicated a large discrimination between tilapia genera (Tilapia, Sarotherodon and Oreochromis). The largest group consisted of most of the Oreochromis species. West African O. niloticus (Onn_Kp and Onn_Ny) exhibited COI haplotypes typical of O. aureus, as previously reported in Rognon & Guyomard (2003), although nuclear markers clearly indicated the differences between these two species. While the last group consisted of the two populations of T. zillii being the most distant species from the Oreochromis genus.



DISCUSSION

The COI gene tree generally agrees with previous publications using allozymes (Sodsuk & McAndrew, 1991; Pouyaud & Agnèse, 1995) and the mitochondrial control region (Nagl et al., 2001), in that Sarotherodon species were not clearly separated from Oreochromis (unlike our ddRAdseq study using >600 SNP, in which all three genera were separated). We also could not separate O. andersonii and O. macrochir or West African O. nilloticus from O. aureus using COI sequence. These issues seem from the COI being only a single, maternally

SUMMARY

The COI DNA barcode is able to resolve most, but not all, of the species involved in this study. As a single, maternally inherited marker it is of limited use in analysing cases of hybridization/introgression. However, it is still likely to be useful in combination with multiple nuclear DNA markers.

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