A close-kin mark-recapture pilot study for Indian Ocean yellowfin tuna

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Ashley Williams, Laura Tremblay-Boyer, Rich Hillary, Ann Preece

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Contents

1 Summary

A close-kin mark-recapture (CKMR) design study completed in 2022 estimated that the collection of 25,000 to 30,000 samples per year from Indian Ocean yellowfin tuna, over a five-year period, would provide an estimate of absolute abundance with an acceptable level of precision. The Working Party on Methods (WPM) noted the logistical challenges in collecting this many samples and suggested a staged approach to the implementation of CKMR for yellowfin tuna. This paper outlines a proposal for the implementation of a CKMR pilot project for Indian Ocean yellowfin tuna to evaluate the logistics and feasibility of sampling, and levels of cross contamination of DNA. The WPM is invited to provide feedback on this proposal.

2 Introduction

Uncertainty in tuna stock assessments across Regional Fisheries Management Organisations (RFMOs) has motivated the exploration of alternative approaches for estimating abundance of tuna. This is particularly the case for the Indian Ocean yellowfin tuna stock, which has been estimated to be overfished and subject to overfishing since 2015, and is currently subject to a review due in part to conflicts and inaccuracies in the data inputs to the stock assessment (IOTC 2021).

Close-kin mark-recapture (CKMR) is a fisheries-independent method that can provide an estimate of absolute abundance and other key population metrics such as total mortality (Bravington et al. 2016a). Importantly, it is independent of the current data inputs and biases associated with the current stock assessment. CKMR has a proven track record in the assessment and management of Southern bluefin tuna (Bravington et al. 2016b), where the data are integrated into the stock assessment models and are used in the management procedure (e.g., Hillary et al. 2019, 2020). CKMR is being developed for a range of other species, and across the tuna RFMOs, including for Atlantic bluefin tuna (Grewe et al. 2018) and South Pacific albacore (SPC-OFP and CSIRO 2023).

CKMR was first identified as a priority research activity for yellowfin tuna in the Working Party on Tropical Tuna (WPTT) Program of Work in 2017 (IOTC 2017). In 2022, a design study for Indian Ocean yellowfin tuna evaluated a range of sampling scenarios that would provide estimates of spawning stock biomass, depletion, adult mortality and mean recruitment using CKMR (Hillary et al. 2022). The authors concluded that annual sample sizes of 25,000 to 30,000 collected from the catch of yellowfin tuna over a five-year period would provide reasonable precision in estimates of these population metrics. Furthermore, they estimated that greater precision in population parameter estimates could be achieved by sampling proportionally more juveniles than adults, specifically 70% juveniles and 30% adults.

The application of CKMR requires an estimate of age for the individuals that are sampled to determine birth year and age-specific parameters for the calculation of kinship probabilities. Otoliths currently provide the most reliable estimates of age for tunas, but it is unlikely that 25,000 to 30,000 otoliths could be collected, processed and analysed each year for yellowfin tuna. Epigenetic ageing using DNA methylation have demonstrated encouraging results across a range

of species (Mayne et al. 2020, 2021), including yellowfin tuna in the Pacific Ocean up to 10 years of age (Mayne et al. 2023). The cost of epigenetics is comparable to using otoliths to estimate age, but tissue samples for epigenetics can be processed far more quickly than otoliths. Epigenetic ageing would present a more viable option for CKMR sampling of yellowfin tuna as the same tissue sample could be used for both genotyping (to identify kin pairs) and epigenetic ageing.

In 2022, the Working Party on Methods (WPM) supported further advancing the implementation of CKMR for Indian Ocean yellowfin tuna (IOTC 2022). The WPM noted the logistical challenges in collecting the recommended number of samples, and suggested that a staged approach be used, whereby an initial year of sampling is conducted to determine whether samples in sufficient numbers and quality can be collected with DNA contamination kept to acceptably low levels. The WPM also noted the possible need for adaptive sampling, whereby the annual sample size may need to increase in subsequent years if the early sampling contains fewer parent-offspring pairs (POPs) and half-sibling pairs (HSPs) than expected based on the results of the design work.

This paper outlines a proposal for the implementation of a CKMR pilot project for Indian Ocean yellowfin tuna to evaluate the logistics and feasibility of sampling, and levels of cross-contamination of DNA.

3 Proposed pilot study

3.1 Objectives

The proposed CKMR pilot study for Indian Ocean yellowfin tuna is a 3-year project with the main objectives to:

1. Establish a network of international collaborators to coordinate the implementation of a sampling program for yellowfin tuna.

2. Develop standard operating procedures for collecting, storing and transporting yellowfin tuna tissue samples.

3. Collect up to the annual target of 25,000 to 30,000 muscle tissue samples from yellowfin tuna from across the Indian Ocean, aiming for approximately 70% juvenile and 30% adult fish.

4. Examine the level of DNA contamination among samples collected and adjust sample collection methods if necessary.

5. Calibrate an epigenetic clock to estimate the age of Indian Ocean yellowfin tuna from tissue samples (Mayne et al. 2023).

6. Evaluate the feasibility of implementing the full 5-year sampling design recommended by Hillary et al. (2022).

3.2   Schedule

Year 1: Several workshops with key international collaborators would be scheduled in the first year of the project to establish the project team and to develop standard operating procedures for the training and sampling. Training of personnel (e.g. port samplers, observers) to conduct the sampling will also commence in the first year.

Year 2: The sampling program will commence in the second year and will be reviewed after the completion of 12 months to evaluate the feasibility of collecting the target number of samples.

Year 3: If collecting the target number of 25,000-30,000 samples looks feasible, sampling will continue in the third year of the project with the aim of supporting the full implementation of CKMR for yellowfin tuna.

An epigenetic clock for Indian Ocean yellowfin tuna will be developed in the third year of the project from a subsample of the tissue samples collected. The level of cross-contamination of DNA in the samples will be tested for a subsample of fish collected from each major source of sampling. If levels of contamination are found to be high, adjustments to the sampling methods will be made for future sampling.

Results from the CKMR pilot study for Indian Ocean yellowfin tuna will be reported back to the IOTC WPM and Scientific Committee at the conclusion of the project, where a decision to support the continuation of CKMR for this species will be made.

If a full CKMR study was to proceed following the pilot study, the samples collected therein could be used in the full study. Adequate storage for all samples collected would be included as part of the project.

3.3   Budget

The cost of implementing this CKMR pilot study for Indian Ocean yellowfin tuna is substantially less than the full implementation of CKMR, as only 1 -2 years of samples will be collected, as opposed to 5 years, and most of these samples will not be genotyped (at the pilot program stage). However, there are still significant costs in coordinating the collection of a large number of samples across the Indian Ocean. Therefore, it will be necessary to source funds external to the IOTC science budget. An approximate budget for the implementation of this CKMR pilot study for Indian Ocean yellowfin tuna is USD$1.2M over a period of three years. A more detailed budget will be developed for a formal funding proposal if supported by the WPM.

3.4   Sampling considerations

Hillary et al. (2022) showed that with uniform sampling across the four stock assessment regions and two size classes (<50 cm and >75cm), the required number of samples (up to 30,000 per year with 70% juveniles and 30% adults) could be achieved with around 2,250 large yellowfin tuna (>75cm) sampled each year from each region, and 10,500 small yellowfin tuna (<50cm) from Region 1 and Region 4 (as very few juveniles are likely to be sampled from Region 2 and Region 3). The proposed workshops for the CKMR pilot project will bring together expert knowledge of the fisheries operating across the Indian Ocean and of the potential sampling opportunities (e.g., port

sampling, observers, etc.). Key locations and fisheries will be identified for the collection of the samples.

The development of standard operating procedures for sampling will be an important product developed during the proposed workshops in the first year of the project. These standard operating procedures will outline the protocols for samplers to use when identifying species (as juvenile yellowfin tuna are difficult to differentiate from juvenile bigeye tuna), selecting which fish to sample, collecting tissue samples, recording ancillary data (including accurate length measurement), storing tissue samples and shipping samples to the laboratory for DNA extraction and potential sequencing. The standard operating procedures are particularly important to ensure that cross-contamination of DNA is minimised, as contamination increases the rate of false positives in the kin-finding process of CKMR.

3.5   Sample processing

Samples collected during the first year of sampling will be sent to a laboratory at CSIRO in Hobart, Australia for storage. DNA will be extracted for a subsample of 50 fish collected from each major location of sampling and used to test for cross-contamination as well as for epigenetic ageing. The level of cross-contamination of DNA in the samples will be examined to identify sampling methods, locations or samplers for which contamination might be unacceptably high. If levels of contamination are found to be high, adjustments to sampling methods will be made for future sampling.

An epigenetic clock has been developed for yellowfin tuna in the Pacific Ocean, but it is unclear whether it is appropriate to apply this to Indian Ocean yellowfin tuna. Therefore, in the third year of the project, an epigenetic clock will be developed using a subsample of 500 otolith and tissue samples from Indian Ocean yellowfin tuna, which would then be available to estimate the age of all samples collected in the full implementation of CKMR for Indian Ocean yellowfin tuna.

3.6   Collaboration

An informed and substantial sampling program will be required to collect a target of 25,000-30,000 samples across regions and size classes throughout the Indian Ocean. However, this level of sampling intensity is already being achieved by existing length data collection programs implemented by IOTC member countries (Hillary et al. 2022). These existing programs could be expanded to include the collection of tissue samples for CKMR. To achieve this would require strong collaboration and cooperation from member nations, institutes, organisations and, most importantly, fishers, observers, and port samplers. The proposed workshops in the first year of the CKMR pilot study would establish a network of experts and a project team to collaborate on designing and implementing the sampling program.

4   Discussion

This paper details a possible pilot study to support the implementation of close-kin methods for unbiased estimation of population abundance and other management metrics of yellowfin tuna in

the Indian Ocean. It responds to concerns by the WPM that the number of samples required annually might be logistically challenging.

The approach outlined here would lead to an improved understanding of logistical hurdles in the large-scale collection of tissue samples for yellowfin tuna in the Indian Ocean. It would allow the WPM to make an informed decision when evaluating whether to pursue a full CKMR study. If a full CKMR study was to proceed following the pilot study, the samples collected therein could serve as the first 1-2 years of the required samples. Conversely, if the WPM decided not to support a full CKMR study, this project will have provided a calibrated epigenetic clock to facilitate the ageing of yellowfin tuna individuals, as well as enhanced regional capacity for large-scale sampling of this stock through training, network development and documentation of sampling opportunities. Given ongoing issues in the quality of data inputs to the current stock assessment, such opportunities for large-scale sampling have applications beyond CKMR.

As such, the authors seek support and feedback from the WPM on the proposed pilot study (noting funding to be sourced externally to IOTC), as well as expressions of interest for collaboration in the development of international networks to conduct large-scale sampling for yellowfin tuna.

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