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# Report of the SBT genetagging program 2022

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Report to the Commission for the Conservation of Southern Bluefin Tuna



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### 1 Abstract

The CCSBT gene-tagging program provides an estimate of the absolute abundance of the age-2 cohort, for use in the Cape Town Procedure and stock assessment models. Four estimates are available for the age-2 cohorts in 2016-2019, but the 2020 program of work was cancelled because of disruption to the field work from COVID-19. The 2021 field work recommenced with over 7000 aged-2 fish tagged. The collection of tissue samples from age 3 fish in 2022 has been completed with over 11000 fish sampled. DNA from these two sets of samples will be compared and an estimate of abundance of the age 2 cohort in 2021 will be available in early 2023. The 2022 field work was also successful with over 5000 fish tagged.

## 2 Introduction

The CCSBT gene-tagging program is designed to provide an estimate of the absolute abundance of age 2 SBT, for use in the Cape Town Procedure and stock assessment models. The program has been in operation since 2016.

This report provides the abundance of age 2 fish in 2016-2019. There is no estimate for 2020 because the field work was cancelled due to covid-19.

We also provide an update on the gene-tagging programs in 2021 and 2022, Research Mortality Allowance (RMA) usage and the RMA request for 2023.

### 3 Method

Gene-tagging SBT involves "tagging" fish by taking a very small tissue sample (Bradford et al 2016) from a large number of 2-year-old SBT, releasing the fish alive, allowing 12 months for mixing with untagged SBT, and then taking tissue samples from the catch of 3-year-old fish at time of harvest. The DNA from the two sets of samples are genotyped and then compared in order to find the samples with matching DNA; a match indicates that a tagged and released fish was recaptured. The abundance estimate is calculated from the number of samples in the release and harvest sets and the number of matches found. The design study examined sample sizes, potential sources of bias, costs and precision of estimates and integration of data in stock assessment and management procedure models (Preece et al., 2015).

The gene-tagging program follows the specifications for the pilot study as recommended in the design study. Twenty days at sea is considered the minimum viable period to achieve the desired samples size, allowing for bad weather and poor fishing days, based on previous experience with conventional SBT tagging projects. The design study recommended tagging and releasing 5,000 fish and harvest sampling 10,000 fish.

The project involved the following steps:

- 1. Tag and release: vessel charter and pole and line fishing for at-sea collection of tissue samples from age 2 fish in the Great Australian Bight during the summer of year 1.
- 2. Harvest sampling: collection of tissue samples from age 3 fish in winter (June-August) of year 2, during harvest of fish in farms, which were caught by the Australian surface fishery in Jan-Mar of year 2.
- 3. DNA extraction and genotyping of tissue samples, using CSIRO-developed SNP markers.
- 4. Data analysis and calculation of an abundance estimate, and provision of results to the CCSBT data exchange and Extended Scientific Committee for use in stock assessment models and the management procedure.

The design study noted potential extensions to the basic design adopted, which include: tagging and resampling fish from regions outside the Great Australian Bight, tagging and resampling multiple age classes, and collection of otoliths to address uncertainties in age classes of the fish sampled, given they are selected using a specified length-class. Direct ageing of otoliths and vertebrae was used in 2019 to revise length classes chosen to target 2-year-olds and 3-year-olds (Preece et al., 2019).

### 4 Results for 2016-2019

The results of the 2016-2019 gene-tagging program provide the absolute abundance estimate of the age 2 cohort in the year of tagging (Preece et al, 2021). The gene-tagging data used in the Cape Town Procedure are the number of matches, the abundance estimate, and the age and year to which the estimate applies (Hillary et al, 2020).

Table 1 The results of the gene-tagging programs 2016-2019 which provide the absolute abundance estimate for the age-2 cohort in the year of tagging.

YEAR	COHORT AGE	N RELEASES	N HARVEST	N MATCHES	ABUNDANCE ESTIMATE (MILLIONS)	CV
2016	2	2952	15389	20	2.27	0.224
2017	2	6480	11932	67	1.15	0.122
2018	2	6295	11980	66	1.14	0.123
2019	2	4242	11109	31	1.52	0.180

### 5 The 2020 gene-tagging program

In 2020, the fifth cycle of gene-tagging commenced with at-sea tagging in March 2020. The field team had difficulties finding fish, and weather conditions were not ideal. The CSIRO field team was urgently recalled back to Hobart after only 9 days of the 20-day field trip because of COVID-19 risks and border closure uncertainties at that time. Too few fish were sampled in the limited time at sea to provide a sufficient release sample size for abundance estimation, and therefore there was no harvest sampling in 2021. There is no gene-tagging estimate of abundance for the age-2 cohort in 2020.

# 6 The 2021 gene-tagging program

In 2021 the tagging field work recommenced, and 7155 fish were tagged and released, substantially more than the 5000 fish target. Harvest sampling recommence in 2022, with over 11,000 tissue samples collected. DNA extraction is underway. These data and the abundance estimate will be available in early 2023.

### 7 The 2022 gene-tagging program

In 2022, the field tagging team successfully tagged over 5000 fish, and the trip report is available at appendix A. In 2023 harvest samples will be collected to find recaptured fish, and this abundance estimate will be available in 2024.

#### 8 Research Mortality Allowance

In 2022 233kg of RMA was used. There were 26 mortalities (see trip report, Appendix A).

The request for RMA for the 2023 field trip is 2t. This is expected to be an over-estimate of the requirements, that allows for unusual and unforeseen conditions.

#### 9 Summary

The CCSBT gene-tagging program has successfully completed four full cycles (2016-2019), however in 2020 the field work was interrupted by COVID-19 travel restrictions and the remainder of that cycle did not proceed. The 2016-2019 abundance estimates will be used in 2022 in the management procedure for recommending the total global allowable catch.

In 2021 tagging recommenced with over 7000 fish tagged and samples were collected from over 11,000 fish during the harvest in 2022. DNA will be extracted from all suitable tissue samples. The 2021 abundance estimate will be available in early 2023 for use in the next stock assessment.

The 2022 tagging program has successfuly sampled tissue from over 5000 fish.

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# Appendix A: 2022 Field Trip Report

#### CCSBT-CSIRO Southern Bluefin Tuna Gene Tagging – March 2022

CSIRO Personnel: Emma Westlake, Russell Bradford

Trip dates: 10-30 March 2022.

The southern bluefin tuna (SBT) gene tagging project aims, on an annual basis, to catch, obtain a tissue sample (gene tag), and release 5,000 southern bluefin tuna (SBT) within the Great Australian Bight. The 2022 gene tagging trip was the seventh such trip to tag live SBT. With COVID-19 remaining a risk to the field team, it was decided to reduce the field staff to only two for 2022. Despite pre-boarding Rapid Antigen Tests (RAT) being taken by the majority of the crew and tagging staff, a confirmed COVID case emerged during the course of the second leg, reducing the crew to six. At the completion of field work, no other crew member nor the tagging staff tested positive for COVID.

Commercial fishing operations ceased around 10 March allowing the gene tagging field work to commence earlier than in the previous two years. The Yasmin departed port on the evening of 10 March; aiming to begin fishing operations in the region to the south and southeast of Kangaroo Island the following morning. At the start of fishing operations, the wind was up with a swell of approx. 3-4 m in height. Over the course of the next few days the weather settled somewhat. The first leg of the field work was approximately 11 fishing days, with the second leg accounting for the remainder of the 20 allocated sea days.

The Yasmin traversed a minimum of 5,000 km during the 2022 field work (Figure 1), including areas at both the western and eastern extremes of previous fishing efforts. The western regions around the Nuyts Archipelago and Cannan Reef have not been fished for a number of years. However, the successful tagging operations during the first leg allowed the team to investigate these regions again. No SBT were tagged at Nuyts (no SBT were observed schooling), and only a small number at Cannan Reef. The majority of SBT from Cannan Reef down to west of Ward Island were below the target size range.



Figure 1. The track of the Yasmin during the 2022 Gene Tagging field work.

In total, 4,493 SBT were tagged during the first leg and a further 935 during the second leg, for a grand total of 5,428 SBT gene tagged in 2022. The total number tagged is roughly in line with the previous six years (Table 1).

Table 1. Number of samples from live fish (age-2) collected at sea with number of corresponding harvest samples (age-3 fish). The genetic profiles of age-3 samples were compared against the live samples of age-2 fish from the previous year. \* No harvest samples were collected in 2021 due to the lack of field samples in 2020.

Year – Vessel	Days at Sea	Age-2 Tagged	Harvest Samples
2016 – Celtic Rose	20	3,768	0
2017 – Yasmin	20	7,633	15,908
2018 – Yasmin	20	8,200	15,088
2019 – Yasmin	20	4,631	11,318
2020 – Yasmin	8 (COVID-19 affected)	66	12,144
2021 – Yasmin	20	7,155	0*
2022 – Yasmin	20	5,428	

The target fork length in 2022 was between 75 and 85 cm (FL). The fork length of SBT tagged peaked at 76 cm (Figure 2). The number of SBT caught and returned without sampling (i.e., those outside the target FL) was 3,242 and skewed to fish smaller than the target range (Figure 2).



Figure 2. Length frequency histogram of pole-caught SBT during the 2022 gene tagging field season. Blue = tagged SBT; orange = caught and released without tagging ("discards").

A total of 75 SBT were caught on the troll lines while searching for schools of SBT. Of these, one was killed because of injury sustained from the troll line; the remaining 74 were returned after being measured and weighed. The length-weight relationship for the troll caught SBT is provided in figure 3.





Twenty-six SBT sustained damage from the poling/trolling operations that was deemed fatal. These fish were immediately humanely killed and biological samples obtained from them. Additionally, these fish were used to train in archival tag insertion and suturing techniques. Total mortalities amounted to approx. 233 kg of SBT. The length-weight relationship for the SBT mortalities is provided in figure 4.



Figure 3. Fork Length-Weight relationship for SBT mortalities during 2022 with trend line (Excel) and R-squared.

A refinement on previous years was the addition of crushed ice, compared to ice blocks alone, to the esky used to store samples while tagging is underway (Figure 4). As samples were placed in the esky, they were pushed down into the crushed ice to speed the chilling process. At the completion of each box of samples (92 samples), the vials were placed into the -20 °C freezer. If the time to complete a box of samples extended beyond approximately 40 minutes, the samples in the esky were transferred to the -20 °C freezer; with the remaining samples added later.



Figure 4. Plan view of the on-deck esky with the addition of crushed ice to speed the chilling of the tissue samples.

Also refined this year was the marking of the 'sweet' zone on the chute. Last year this zone (75-85 cm) was marked in automotive green paint. The paint rubbed off fairly quickly. Prior to the second leg of the current field work, this zone was polished with a steelo pad to make it brighter and easier to see (Figure 5). Green paint was used on the sides where less rubbing by fish resulted in a clearly defined zone. This increased measurement ease and efficiency and assisted the tagger in ensuring measurements were taken correctly.



Figure 5. The 'sweet zone' (75-85 cm) clearly marked on the chute to aid in quick and correct measurements.

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