SCIENTIFIC COMMITTEE

TWENTY-FIRST REGULAR SESSION

Tonga

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Progress Towards a Close-Kin-Mark-Recapture Application to South Pacific Albacore (Project 100c)

WCPFC-SC21-2025/SA-WP-09

SPC-OFP[1] and CSIRO[2]

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Executive Summary

SC17 established Project 100c to prepare western and central Pacific tuna fisheries for the application of close-kin mark-recapture (CKMR) methods to resolve key stock assessment uncertainties.

The project is co-funded between WCPFC and the European Union with additional support from the Oceanic Fisheries Programme of the Pacific Community (SPC-OFP) and CSIRO, Australia. The financial contribution of the European Union is sourced through their European Maritime, Fisheries and Aquaculture Fund. EMFAF funds became available in November 2022 (~10 months later than originally planned at SC17). To accommodate this delay, project milestones have been extended by 12 months (as noted in SPC-OFP 2022 [SC18/SA-IP-10]).

A working paper to SC20 (SPC-OFP and CSIRO 2024 [SC20/SA-WP-09]) listed the specific activities of Project 100c and summarised work completed up until SC20. This working paper updates SC21 on progress under each activity over the past 12 months. Specific activities are listed below, with brief updates under each in italics. Detailed updates for each activity are found in the main text.

1. Complete the foundational research needed for the application of CKMR methods to WCPFC stocks to reduce the uncertainty in stock assessments. This will include:
    i. epigenetic ageing for south Pacific albacore and Pacific bigeye using existing validated otolith age.
    CSIRO has initiated a new avenue of research in collaboration with DArT, which if successful, would significantly improve the workflows and efficiency of applications of epigenetic ageing for CKMR. We expect to be in a position to report on this in more detail to SC22.
    ii. evaluation of radiocarbon otolith age validation of southwest Pacific swordfish and epigenetic age calibration.
    To date, 13 measurements of ^{14}C have been made on 10 swordfish (SWO) aged 3-12 years by Fish Ageing Services. A follow up set of 50 SWO otoliths is currently being processed. Accessing the appropriate number and age distribution of samples to define an age calibration curve remains a challenge, particularly when a genomic-quality tissue sample from the same fish is also required.
    iii. genome resequencing of South Pacific albacore and Pacific bigeye for enhanced detection of kin-pairs.
    Collection and whole genome resequencing of bigeye samples continues. The underlying goal of selecting a subset of highly informative loci for kin-finding analyses is complete for albacore.

2. Complete CKMR feasibility and design study for South Pacific albacore.

    The design study was completed and reported to SC20. The target sample size remains 36,000-84,000 tissue samples across three years. New insights provided by bioinformatic analyses of the first 15,000 genetically sequenced samples do not indicate that refinement of that range is required.

3. Complete CKMR scoping studies for Pacific bigeye and southwest Pacific swordfish.

    CKMR scoping studies are complete within the scope of the current project. Full design studies will still be necessary should CKMR assessments of these fish stocks proceed.

Final bigeye results were reported to SC20. Results for southwest Pacific swordfish from a range of scenarios indicate that at least 10,000 samples would be required over four or five years to conduct a satisfactory CKMR assessment.

4. Develop and trial Standard Operating Procedures for the cost effective and reliable collection of tissue samples necessary for CKMR applications to WCPFC stocks.

   A core standard operating procedure has been available since 2023 and has produced satisfactory DNA quality, as confirmed by the first major round of kin-finding bioinformatics conducted in 2025.

5. Use trial samples to investigate and validate connectivity hypotheses via non-close-kin methods for South Pacific albacore in preparation for the 2024 stock assessment.

   Following the results from a phase-one population structure assessment reported to SC20, additional samples were incorporated to assess population structure at a finer geographic scale. New analyses support the original interpretation that population structure exists for South Pacific albacore within the WCPO and proposes that at least three genetic groups present in the WCPFC Convention Area.

6. Develop capacity within WCPFC to implement and evaluate CKMR applications to WCPFC stocks.

   Nine CKMR-focused training events have been held since 1 July 2024. These have trained or upskilled 67 samplers from 5 countries. In-country staff are now responsible for 73% of samples collected for CKMR assessments. Roughly 36,000 tissue samples from south Pacific albacore have been collected since February 2023.

7. Provide advice to the Scientific Committee on what further research and data improvements are needed to enable best use of CKMR methods.

   This activity is scheduled for project completion.

A full report on population structure results from activity 5 is available in SC21/SA-WP-10. We also recommend SC21/SA-WP-14 for a detailed report on preliminary bioinformatic analyses referenced in activity 1iii and activity 2.

Recommendations

SC21 is invited to:

- note the progress on each activity to date;
- note progress on bioinformatic activities reported more thoroughly in SC21/SA-WP-14, particularly:
    - the design and successful roll-out of a specialised kin-finding genetic array
    - quality control and kin-finding assessments of more than 14,000 samples
- recall that all proposed project milestones are delayed by 12 months (from that endorsed at SC17) due to the EMFAF grant not commencing until November 2022;
- consider the scheduling of and resourcing for the inclusion of CKMR data in future stock assessments for south Pacific albacore, noting the positive results from initial genetic sequencing and bioinformatic efforts and the demonstrated capacity of sampling teams throughout the region to achieve the target of 36,000-84,000 tissue samples over a three-year period; and
- acknowledge the European Union for their continued support of this work.

Specific to activity 5, testing connectivity hypotheses, SC21 is invited to:

- Note the results summarised in this paper and in SC21/SA-WP-10
- Recognise the value of the evidence presented here to:
    - help inform decisions on the configuration of tuna stock assessments (sensu Hamer al. 2023); and
    - help inform CKMR sampling designs and analytical pipelines for WCPFC Project 100c
- Continue to support 2024 PAW recommendation for follow-up studies of south Pacific albacore population structure, particularly through the expansion of the study area to the EPO

Background

The successful application of close-kin mark-recapture (CKMR) is dependent on adequate understanding of a species' biology, fishery operation and consideration of sampling logistics. This includes the capacity to collect enough tissue samples, given the size of the stock, to identify sufficient numbers of kin pairs and the capacity to estimate the age of the individuals sampled. Validating our understanding of the species biology (e.g., reproductive biology, sexual dimorphism, spatial connectivity) and evaluating the logistical feasibility of sample collection, storage, transport and analysis are necessary first steps for implementing CKMR.

Project 100c was established by the WCPFC Scientific Committee (SC) at SC17 to address these questions. The specified activities of the project include:

1. Complete the foundational research needed for the application of CKMR methods to WCPFC stocks to reduce the uncertainty in stock assessments. This will include:
    i. epigenetic ageing for south Pacific (SP) albacore and Pacific bigeye using existing validated otolith age.
    ii. evaluation of radiocarbon otolith age validation of southwest Pacific swordfish and epigenetic age calibration.
    iii. genome resequencing of SP albacore and Pacific bigeye for enhanced detection of kin-pairs.
2. Complete CKMR feasibility and design study for SP albacore.
3. Complete CKMR scoping studies for Pacific bigeye and southwest Pacific swordfish.
4. Develop and trial Standard Operating Procedures for the cost effective and reliable collection of tissue samples necessary for CKMR applications to WCPFC stocks.
5. Use trial samples to investigate and validate connectivity hypotheses via non-close-kin methods for SP albacore in preparation for the 2024 stock assessment.
6. Develop capacity within WCPFC to implement and evaluate CKMR applications to WCPFC stocks.
7. Provide advice to the SC on what further research and data improvements are needed to enable best use of CKMR methods.

Project 100c is now in its fourth reporting year and has taken significant steps towards fulfilling its activities.

Project Administration

This project is supported by the European Union through its European Maritime, Fisheries and Aquaculture Fund with a budget of approximately Euro 270,000. WCPFC has allocated a further USD40,000 to support implementation. SPC-OFP and CSIRO are providing additional in-kind and operational support (to December 2025). Project review and guidance is provided by the SC.

Progress to date on each activity

Activity 1—Complete the foundational research needed for the application of CKMR methods to WCPFC stocks to reduce the uncertainty in stock assessments.

i—epigenetic ageing for south Pacific albacore and Pacific bigeye using existing validated otolith age

As noted to SC20 (SPC-OFP and CSIRO 2024), epigenetic ageing has the potential to transform the assessment and management of tuna fisheries by being able to estimate age of individual fish directly from tissue samples. This would significantly reduce the cost and logistic challenges of collecting direct age data from otoliths. CSIRO and others (e.g., Chevrier et al., 2025) have been investing in the development of this priority area. The development of high-throughput approaches, as would be required for routine implementation of CKMR, for example, has proved challenging. CSIRO has initiated a new avenue of research in collaboration with DArT, which if successful, would significantly improve the workflows and efficiency of applications of epigenetic ageing for CKMR. We expect to be in a position to report on this in more detail to SC22.

ii—evaluation of radiocarbon otolith age validation of southwest Pacific swordfish and epigenetic age calibration

Swordfish (SWO) age estimation (age reading of otoliths) and validation using bomb radiocarbon (14C) dating is currently underway. To date, 13 measurements of 14C have been made on 10 SWO aged 3-12 years by Fish Ageing Services (FAS). A few exploratory measurements were also made on sagitta edge and lapillus material to potentially assist with 14C alignments in time. A follow up set of 50 SWO otoliths is currently being processed for core extraction with estimated ages of 0 to 21 years. All material extractions for accelerator mass spectrometry on this species are performed by hand and is very tedious for confident isolation of 100-200 ug of earliest otolith material. Preliminary 14C results from SWO from the North Atlantic indicate that the age reading of otoliths from this species is very difficult due to irregular otolith shape, very low mass, and fragile nature. Hence, we hope the findings of this study can support the determination of age in establishing an epigenetic clock that can be used *in lieu* of otoliths.

iii—Genome resequencing of south Pacific albacore and Pacific bigeye for enhanced detection of kin-pairs

Genome resequencing information is helpful to guide marker selection and determine loci to be used for CKMR assessments per species. Collection and whole genome resequencing of additional bigeye tuna continues, including on SPC's 2025 tagging cruise between Hawaii and the Line Islands in Kiribati. The tagging cruise is ongoing at the time of writing.

For albacore, work is now complete to select two sets of highly informative loci for standardised quality control and kin-finding purposes, which fulfills the purpose of genome resequencing in that species. Please see SC21/SA-WP-14 (CSIRO and SPC, 2025a) for an in-depth discussion.

Activity 2—Complete CKMR feasibility and design study for South Pacific albacore

The SP albacore design study was completed in 2024 and reported to SC20 (Tremblay-Boyer et al. 2024). The target sample size for a CKMR assessment of SP albacore remains 36,000-84,000 tissue samples across three years.

Initial genetic sequencing and bioinformatic work supports this range. Roughly 15,000 samples have been sequenced, screened for data quality and analysed for presence of close kin. Less than 4% of sequenced samples were rejected from analysis due to quality issues, including

DNA degradation, cross contamination or species misidentification. It is therefore not necessary to expand sampling goals to accommodate a higher-than-expected sample rejection rate. This metric does not account for samples that are currently deferred from sequencing due to their reduced likelihood of producing adequate results (such as those that yielded a low concentration of DNA) because there are laboratory protocols that can reclaim some of these samples (at a financial and labour cost) if eventually needed.

Among the 15,000 samples, kin-finding analyses identified 3 parent-offspring pairs and 12 half-sibling pairs. The addition of more samples will produce disproportionately more opportunities to identify a close relative. Therefore, no refinement to the number of samples collected as recommended in Tremblay-Boyer-et al., (2024) is expected to be required.

For a full commentary on initial genetic sequencing results and other bioinformatic work related to this project, see SC21/SA-WP-14 (CSIRO and SPC-OFP, 2025a).

Activity 3—Complete CKMR scoping studies for Pacific bigeye and southwest Pacific swordfish

CKMR scoping studies for Pacific bigeye tuna and southwest Pacific swordfish have been completed. The scoping studies used population parameters and abundance estimates from the most recent WCPFC stock assessments for bigeye tuna (Day et al. 2023) and southwest Pacific swordfish (Ducharme-Barth et al. 2021) to estimate the number of parent-offspring pairs (POPs) and half-sibling pairs (HSPs) that would be expected to be identified from alternative scenarios of abundance, annual sample sizes and sampling program lengths.

For bigeye tuna, two alternative abundance scenarios examined which included i) the estimate of numbers-at-age from the terminal year of the diagnostic model of the 2023 stock assessment, and ii) twice the numbers-at age from this model as an extreme estimate of the real population size. For southwest Pacific swordfish, we used all models from the 2021 model ensemble to provide a range of alternative abundance scenarios. For simplicity, we assumed the ratio of adult to juvenile sampling was 50:50 for both species.

Results for bigeye tuna (Table 1) indicate that under the assumption that abundance estimates from the 2023 stock assessment are accurate, at least 60,000 samples of bigeye tuna (e.g. 15,000 in each of 4 years, or 20,000 in each of 3 years) would be required to detect at least 100 kin pairs (POPs and HSPs combined), which is considered the minimum number required to achieve acceptable levels of precision (~15% CV) in estimates of abundance. Alternately, if the real abundance was much larger (i.e. twice the 2023 stock assessment estimate) then approximately 100,000 samples would be required to detect at least 100 kin pairs.

Results for southwest Pacific swordfish (Table 2) indicate that at least 10,000 samples of southwest Pacific swordfish (e.g. 2,000 in each of 5 years, or 2,500 in each of 4 years) would be required to detect at least 100 kin pairs (POPs and HSPs combined), considering the median number of kin pairs across the range of abundance scenarios. Alternately, if the lower quartile of kin pairs from the abundance scenarios were closer to reality, then at least 12,000 samples would be required to detect at least 100 kin pairs.

A proper design study, such as the study recently completed for South Pacific albacore (Tremblay-Boyer et al. 2024), that represents the biology, population and fisheries dynamics more accurately, and considers alternative abundance assumptions and sampling options, should be completed for both species to provide more reliable estimates of the sampling requirements to achieve the required precision in population estimates. Estimates of the

expected precision in population metrics of interest given various sampling options would also be produced from a proper design study.

Table 1. Results from the CKMR scoping study for Pacific bigeye tuna.

Abundance scenario	Annual samples	Sampling years	POPs	HSPs	Total samples	Total Kin
Stock assessment	10000	3	5	22	30000	27
Stock assessment	10000	4	10	40	40000	50
Stock assessment	10000	5	16	63	50000	78
Stock assessment	15000	3	12	50	45000	62
Stock assessment	15000	4	22	90	60000	112
Stock assessment	15000	5	35	141	75000	176
Stock assessment	20000	3	20	89	60000	110
Stock assessment	20000	4	39	160	80000	198
Stock assessment	20000	5	63	250	100000	313
Stock assessment	25000	3	32	140	75000	172
Stock assessment	25000	4	60	249	100000	310
Stock assessment	25000	5	98	391	125000	489
2x Stock assessment	10000	3	3	11	30000	14
2x Stock assessment	10000	4	5	20	40000	25
2x Stock assessment	10000	5	8	31	50000	39
2x Stock assessment	15000	3	6	25	45000	31
2x Stock assessment	15000	4	11	45	60000	56
2x Stock assessment	15000	5	18	70	75000	88
2x Stock assessment	20000	3	10	45	60000	55
2x Stock assessment	20000	4	19	80	80000	99
2x Stock assessment	20000	5	31	125	100000	156
2x Stock assessment	25000	3	16	70	75000	86
2x Stock assessment	25000	4	30	125	100000	155
2x Stock assessment	25000	5	49	195	125000	244

Table 2. Results from the CKMR scoping study for southwest Pacific swordfish.

				POPs			HSPs		
Kin type	Annual samples	Sampling years	Total samples	25th Quantile	Median	75th Quantile	25th Quantile	Median	75th Quantile
POPs	2000	3	6000	24	35	48	6	7	11
POPs	2000	4	8000	39	58	79	11	13	20
POPs	2000	5	10000	57	85	115	17	20	30
POPs	2500	3	7500	37	55	74	10	12	17
POPs	2500	4	10000	61	91	123	17	21	31
POPs	2500	5	12500	90	133	180	26	31	48
POPs	3000	3	9000	53	79	107	14	17	24
POPs	3000	4	12000	88	132	178	25	30	44
POPs	3000	5	15000	129	192	259	38	45	69

Activity 4—Develop and trial Standard Operating Procedures for the cost effective and reliable collection of tissue samples necessary for CKMR applications to WCPFC stocks

Standard operating procedures for genetic sampling for CKMR are in place since 2023 and are demonstrably working well, based on the rate of return of high-quality sequencing data referenced in Activity 2.

Small protocol tweaks in the past year have been made to sample vial labels and storage requirements with intent to improve efficiency of sample handling in the laboratory. Other efforts to facilitate high-quality and high-volume sample collection involved changes outside the standard operating procedure (SOP), itself. Most notably, SPC-OFP is developing a new length conversion factor to associate the distance between the finlet length (length from the anterior insertion point of the 2nd finlet to the fork in the tail on either the dorsal or ventral side of the fish) to the total fork length. This enables sampling of frozen tail stumps saved from fish that were processed at sea, rather than requiring whole fish to achieve a length-to-age estimate. See SC21/ST-IP-03, Appendix 2, for a more complete report about the new length metric. While tails-only sampling changes nothing in the sample collection SOP, itself, it has expanded the applicability of the protocol by **tens of** thousands of samples.

Another focus of extra-SOP developments is the design of various quality checks that are in the process of rolling out. These include a checklist of protocol details to be confirmed by any visiting, trained SPC or regional staff to a sampling event, and a standardized feedback report to samplers whose samples have undergone sequencing. The intention is to flag deviation from the established SOP as soon as possible, to avoid wasting resources on collecting and processing non-conforming samples.

Activity 5—Use trial samples to investigate and validate connectivity hypotheses via non-close-kin methods for south Pacific albacore in preparation for the 2024 stock assessment

In 2022, a study was designed to address uncertainty about population structure in South Pacific albacore in the WCPO, included a small-scale phase one that was reported to SC20 (Macdonald et al 2024). Results from phase one, which contrasted genetic and otolith data from 55 albacore caught in each of French Polynesia and New Caledonia within the span of a month, validated the expansion of the study. This expansion utilised genetic markers only and incorporated almost 1000 additional samples from multiple sampling locations with both longitudinal and latitudinal variation to assess finer scale spatiotemporal patterns in population structure of South Pacific albacore.

A full discussion of the results is available in SC21/SA-WP-10 (CSIRO and SPC-OFPb, 2025). The data identifies the presence of at least three distinct genetic groups within the WCPO. This population structure will be important to incorporate into CKMR models. Existing sampling infrastructure should be sufficient to distribute sampling effort across the described subpopulations.

Activity 6—Develop capacity within WCPFC to implement and evaluate CKMR applications to WCPFC stocks

Between 1 July 2024 and 30 June 2025, SPC-OFP staff have led 9 training events that included CKMR sampling. These events included induction of new samplers and refresher/upskilling

courses for existing samplers. In total, these events were attended by 69 port samplers, fisheries observers, government and industry staff from 6 countries. Across the life of the project, that translates to 151 individuals in 14 countries who are properly trained to take samples. See Table 3, below, for a breakdown of training event attendance in the last year (July 1 2024-July 1, 2025), noting the discrepancy between number of participants and trained individuals because some individuals have participated in more than one training.

Table 3. Summary of training events for the duration of Project 100c

Training	Date	Trainees
Fiji, Suva	Jan-24	19*
Tonga, Nuku'alofa	Feb-24	13
Marshall Islands, Majuro	Mar-24	9
Samoa, Apia	Mar-24	11
Solomon Islands, Noro	Apr-24	17
Federated States of Micronesia, Kosrae	May-24	5
Federated States of Micronesia, Pohnpei		15
New Caledonia, Noumea	Jul-24	2
Marshall Islands, Majuro	Aug-24	7
Federated States of Micronesia, Kosrae	Nov-24	7
Marshall Islands, Majuro	Mar-25	8
Papua New Guinea, Lae	Apr-25	9
Papua New Guinea, Madang		8
Papua New Guinea, Rabaul		8
Papua New Guinea, Wewak		9
Kiribati, Tawara	Jun-25	11
Grand Total		158

*Staff who attended this training in Fiji received theoretical training modules. However, further practical demonstrations were conducted with these participants at a later stage.

In addition, three individuals in New Zealand have received full training outside the formal workshop environment and are likewise qualified to lead port-based tissue sampling events. See Table 4 for a by-country breakdown of fully trained staff. Upwards of 10 SPC-Noumea based staff have also been formally or informally trained as part of their engagement with Project 100c, which allows them to aid in sampling or provide feedback about protocol adherence to sampling teams during port visits (as mentioned in Activity 4).

Table 4. Summary of trained samplers in participating countries

Country	**Staff trained**		
	pre-July 2024	**July 2024-June 2025**	**Total**
Cook Islands	1	0	1
Federated States of Micronesia	22	7	29
Fiji	20	0	20
French Polynesia	2	0	2
Kiribati	3	9	12

Marshall Islands	1	16	17
Nauru	1	1	2
New Caledonia	2	0	2
New Zealand	3	0	3
Papua New Guinea	0	34	34
Samoa	3	0	3
Solomon Islands	19	0	19
Tonga	13	0	13
Tuvalu	2	0	2
Vanuatu	2	0	2
Grand Total	**94**	**67**	**161**

It should be acknowledged that only around 14% of individuals who participate in training events have gone on to formally collect samples. Complicating factors include quick turnover in the industry and the nomination process within each country. However, samplers trained at these events are responsible for collecting roughly 73% of all CKMR samples (in contrast to SPC or CSIRO staff, New Zealand samplers who received training from CSIRO staff, etc). The investment in regional capacity building is demonstrably successful and essential to the progress of the project.

Based on sampling data validated in BioDaSys, roughly 25,000 SP albacore samples were collected between 1 July 2024 and 30 July 2025, for a project total of almost 36,000 since sampling began in 2023 (see table 5 for a precise breakdown). The past year's sampling volume tracks positively with the updated modelled sampling requirements from SC20 (36,000-84,000 tissue samples over three years which to achieve CVs of 15% or less; Tremblay-Boyer et al. 2024). Additional sampling capacity is also upcoming in American Samoa via the Starkist Cannery, which processes tens of thousands of tonnes of albacore per year (noting that some of the volume also comes from outside the WCPFC Convention Area).

Table 5. Summary of samples collected and validated in BioDaSys up to 1 July 2025, as queried on 7 July 2025. Note that samples are organised by port of sampling, but some samples come from surrounding EEZ's (particularly true for the Solomon Islands, which includes fish sampled in PNG and Indonesian waters; and Samoa, where fish originated from the Cook Islands EEZ)

Port	Pre-July 2024	July 2024-July 2025	Total
Fiji	503	5755	6258
French Polynesia	0	5319	5319
Marshall Islands	0	21	21
New Caledonia	3	4749	4752
New Zealand	7161	4088	11249
Samoa	245	0	245
Solomon Islands	1362	4902	6264
Tonga	1268	614	1882
Total	**10542**	**25448**	**35990**

Activity 7—Provide advice to the SC on what further research and data improvements are needed to enable best use of CKMR methods

This activity is scheduled for project completion.

Recommendations

SC21 is invited to:

- note the progress on each activity to date;
- note progress on bioinformatic activities reported more thoroughly in SC21/SA-WP-14, particularly:
    - the design and successful roll-out of a specialised kin-finding genetic array
    - quality control and kin-finding assessments of more than 14,000 samples
- recall that all proposed project milestones are delayed by 12 months (from that endorsed at SC17) due to the EMFAF grant not commencing until November 2022;
- consider the scheduling of and resourcing for the inclusion of CKMR data in future stock assessments for south Pacific albacore, noting the positive results from initial genetic sequencing and bioinformatic efforts and the demonstrated capacity of sampling teams throughout the region to achieve the target of 36,000-84,000 tissue samples over a three-year period; and
- acknowledge the European Union for their continued support of this work.

Specific to activity 5, testing connectivity hypotheses, SC21 is invited to:

- Note the results summarised in this paper and in SC21/SA-WP-10
- Recognise the value of the evidence presented here to:
    - help inform decisions on the configuration of tuna stock assessments (sensu Hamer al. 2023); and
    - help inform CKMR sampling designs and analytical pipelines for WCPFC Project 100c
- Continue to support 2024 PAW recommendation for follow-up studies of south Pacific albacore population structure, particularly through the expansion of the study area to the EPO

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