



**WCPFC TUNA TISSUE BANK
STEERING COMMITTEE**

ELECTRONIC MEETING

16 July 2020, from 18:00-19:30 hours Pohnpei time (UTC+11 hours)

**Appraisal of new biological sampling approaches for tropical tunas
on purse seiners**

WCPFC-SC16-RP-P35b-02

**J. Macdonald, F. Roupsard, B. Leroy, C. Sanchez, M. Hosken,
J. Hampton, G. Anderson, S. Nicol**

Contents

Executive summary..... 2

Background and objectives 3

Approach 1: VMS bag and store..... 4

 Summary 4

 Implementation 5

Approach 2: VMS Widget 12

 Implementation 12

A trial of Approach 1: VMS bag and store 13

Conclusions and next steps 15

References..... 16

Acknowledgements..... 16

Executive summary

SPC is currently undertaking work to develop and trial new biological sampling approaches that optimise purse-seine observer efforts for tuna sample collection. The primary goal is to expand sampling coverage for the WCPFC Tuna Tissue Bank (TTB) in both space and time, and in a systematic fashion, and to ensure that the highest-quality biological material is available for the scientific analyses/applications needed to meet WCPFC/SPC objectives.

This Information Paper details two new sampling approaches targeted primarily towards yellowfin and skipjack tunas aboard purse seine vessels, yet employing methods easily tuneable for use on long line vessels. These two approaches are referred to throughout as:-

Approach 1: ‘VMS bag and store’

Approach 2: ‘VMS Widget’

Both approaches make use of real-time, vessel monitoring system (VMS) data to identify a suite of purse seine vessels operating simultaneously, but in different regions across the WCPO, from which tuna samples are then collected. Both rely on effective dialogue with fisheries authorities, fishing companies and observer programmes to coordinate sample collection.

A key difference between approaches relates to where sample processing occurs. In Approach 1, fish are set aside immediately following capture and stored whole at sea, with sample processing undertaken at port. Approach 2 makes use of a tissue-sampling tool called the ‘Widget’, developed over several years by colleagues at CSIRO, Australia. The Widget permits the sampling of clean, small (i.e. 1.5 cm), contamination-free muscle tissue from fresh fish on board the vessel, which can then be frozen individually or stored in a preservative solution on board, guaranteeing sample quality and alleviating cross-contamination risk for downstream genetic analyses.

SPC notes that a proposal for a field trial of Approach 1 was formally endorsed at the 20th Regional Observer Coordinator Workshop held in Funafuti, Tuvalu, in February 2020. The roll-out of this trial has now been delayed due to COVID-19 related travel restrictions, but this has allowed SPC time to sharpen plans for a ‘design study’ for ongoing tuna stock structure work in the region, and to explore how these new sampling approaches may contribute.

We also highlight that CSIRO has granted SPC-OFP access to the Widget for collection of tissue samples from bigeye and skipjack tuna on the upcoming CP14 tuna tagging cruise through the Kiribati EEZ. This will represent the first application of the Widget to WCPO tunas.

We invite SC16 to note:-

- the progress on enhancement of biological sampling approaches for the benefit of TTB holdings, and for scientific analyses making use of TTB samples;
- that following a thorough cost-benefit analysis, SPC recommends **Approach 2: VMS Widget**, or variations on the theme, as the best strategy for achieving tuna sampling objectives in the mid- to long-term;
- that work is continuing to optimise these approaches in the context of developing routine biological sampling protocols for WCPFC needs, and for evolving work on tuna stock structure and population connectivity.

Background and objectives

This document canvasses options for new approaches to biological sampling of tunas aboard purse seine vessels. These approaches are designed to enhance and simplify observer-based sampling to meet WCPFC needs, expanding spatial and temporal sample coverage in a systematic manner and providing the repository of high-quality biological material required to address longstanding questions on tuna stock structure and population dynamics across the South Pacific Ocean. The need for a fresh approach has been identified by SPC through a tuna research directions paper recently published in the journal *Fisheries Research* (Moore et al. 2020), and in an Information Paper to SC15 (Macdonald et al. 2019), and stems from five main issues with the current sampling scheme:-

1) Sampling coverage limitations

The present lack of spatial and temporal replication in sampling coverage across the region. This constrains the breadth, scale and complexity of ecological and management-related questions that can be posed.

2) Sample contamination questions

The current biological sampling methods for muscle tissue make it difficult to guarantee high-quality, uncontaminated samples for subsequent genetic analyses.

3) Incentives and low sample returns

Challenges regarding observer incentives for onboard biological sampling. This has led to high variability in observer effort in sample collection across the region. There are typically many other compliance-related tasks required of observers aboard purse seiners, and biological sampling needs are sometimes prioritised lower. Consequently, we currently see low and inconsistent (in time and space) sample returns to the Pacific Marine Specimen Bank (PMSB).

4) Transport and logistics

The current process of sample transport, archiving and data entry requires substantial effort by SPC staff and in-country associates regarding coordination and logistics. This situation could be improved upon.

5) High costs with less than optimal outcomes

The high costs (in both time and effort) of training observers in biological sampling protocols during observer training courses sometimes outweigh the benefits in terms of later sample recovery.

In light of these issues, two meetings were convened at SPC on 24 October and 29 November 2019 to define options for new, alternative sampling strategies, with a particular focus on obtaining samples for population genetic analyses under the Pacific-European Union Marine Partnership (PEUMP) programme (<https://fame1.spc.int/en/projects/peump>). SPC staff including John Hampton, Neville Smith, Bruno Leroy, François Rouspard, Valérie Allain, Tim Park, Joe Scutt Phillips, Caroline Sanchez and Jed Macdonald attended these meetings. Several options were tabled, and a working document created that outlined the details and anticipated costs and benefits of each approach.

Since then, SPC has delivered presentations on the new sampling approaches at several regional meetings and workshops, namely the Observer Debriefing and Observer Trainer workshops held in Nouméa in late 2019, the 20th Regional Observer Coordinators Workshop (ROCW 20) held in Funafuti, Tuvalu, in early February 2020, and a meeting on tropical tuna biology and ecology attended by SPC and CSIRO scientists in Hobart during late February 2020. The presentations appeared well received by meeting and workshop participants. Much useful feedback was gained, and the working document updated accordingly.

Here, we distil information from the working document and present what we consider the two most

promising sampling approaches based on options tabled to date. These approaches, hereafter referred to as ‘**Approach 1: VMS bag and store**’ and ‘**Approach 2: VMS Widget**’ are outlined below, and have much in common. Both make use of vessel monitoring system (VMS) data to identify a suite of purse seine vessels operating simultaneously, but in different areas, from which tuna samples are then collected. Both rely on open, effective and ongoing communication with fisheries authorities, fishing companies and observer programmes. A key difference between approaches relates to where sample processing occurs:-

Approach 1: VMS bag and store	Fish set aside, bagged and stored whole at sea; sample processing done in port.
Approach 2: VMS Widget	Sample processing done at sea.

We explain these processing procedures further in the next sections. Importantly, both approaches offer substantial advantages over the current tuna sampling scheme. Specifically, they will guarantee systematic sampling coverage across space and through time, increase the number and quality of samples returned to the PMSB, simplify observer duties related to tuna biological sample collection, and streamline the process of sample transport, archiving and database management.

A proposal for a field trial of Approach 1 (see [A trial of Approach 1: VMS bag and store](#) for details) was formally endorsed by ROCW 20 under Agenda Item 31:-

‘ROCW 20 supports SPC in conducting an initial trial of ‘Approach 1: VMS bag and store’ for sampling tuna on board selected purse seine vessels, as described in ROCW 20 Working Paper 7.’

The roll-out of this trial has now been delayed due to COVID-19 related travel restrictions, but this has allowed us time to sharpen plans for a ‘design study’ for ongoing tuna stock structure work in the region, and to explore how these new sampling approaches may contribute.

We note that the sampling approaches set out here are still in development, and SPC welcomes input from WPCFC regarding avenues for improvement.

Approach 1: VMS bag and store

This approach uses real-time VMS data to track purse seine vessel positions, with observers setting aside and freezing whole fish at locations and times defined by SPC. Processing of biological samples then occurs later in port, conducted by in-country port samplers and SPC staff. Below, we provide a summary of the approach and a guide for how it might be implemented, along with a breakdown of the advantages, disadvantages and estimated costs. In addition, we outline details of a field trial to test its feasibility for obtaining samples both for the PEUMP stock structure work, and for ongoing tuna sample collections across the South Pacific more broadly.

Summary

The approach takes advantage of real-time VMS data on purse-seine vessel movements to identify a set of vessels operating simultaneously, but in different areas, from which we obtain samples of whole tunas for later processing in port. The approach rests on engaging fisheries authorities, fishing companies and observers directly in the sampling process (see [Implementation](#) below). Given that this can be successfully achieved, opportunities for building industry interest in SPC’s tuna research programme and for forging collaborative sampling arrangements are many.

Implementation

The following seven steps provide a guide for implementing this approach:-

1) Define species and scales

Determine the species of interest and the spatial and temporal scales of sampling.

These scales are intended to be flexible, tunable to the species and life-history stage(s) selected, the questions posed, and environmental gradients considered important for shaping behavior, population connectivity and stock structure. A systematic, repeatable sampling approach is the main goal. As an example:-

Species: SKJ, YFT

Spatial scale: South Pacific wide. Eight $20^\circ \times 20^\circ$ areas (see Figure 1)

Temporal scale: 1-week sampling period, repeated quarterly

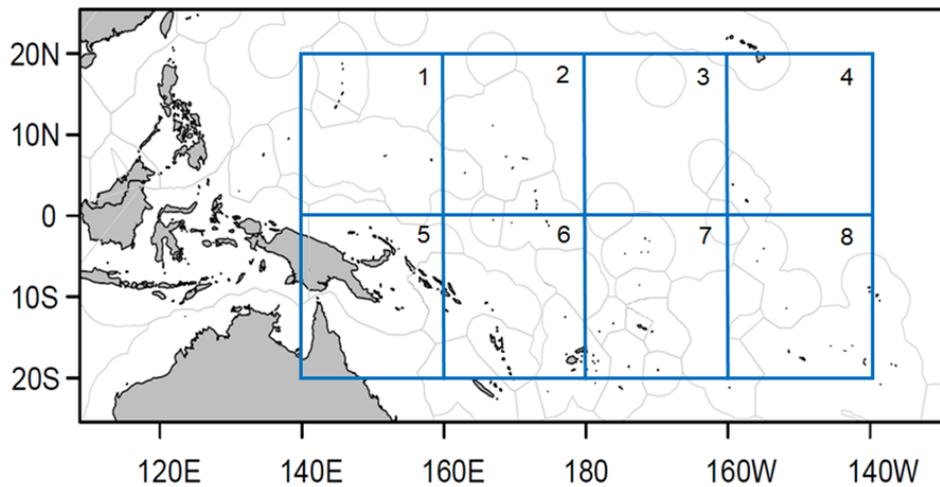


Figure 1. One possible spatial sampling grid. Numbered blue squares define the eight $20^\circ \times 20^\circ$ areas from which tuna samples are required. Note that purse seine fishing effort in the region is concentrated between 10° N and 10° S.

2) Vessel selection

- a. Using the VMS database, extract the list of purse seine vessels operating in each of the eight proposed 20° areas over the previous calendar year (or other relevant timeframe).
- b. Map fishing activity, departure port and arrival port for each vessel per area, during that year, by collecting the following fields:-
 - Vessel name
 - No. of trips (total per year)
 - No. of sets (total per year)
 - Area % (% sets made inside area)
 - Dep. port % (% trips departing from a particular port)
 - Arr. port % (% trips arriving at a particular port)
 - Dep. = Arr. (Departure port is the same as Arrival port) (Y/N)
- c. Use this information to rank each vessel's suitability for our sampling needs. Highest ranked vessels will display high 'Area %', 'Dep. port %' and 'Arr. port %', ideally with 'Dep. = Arr.' = Y.

- d. Select a subset of high-ranking vessels for biological sampling. Targeting specific vessels will simplify observer coordination, sampling logistics, and sample storage and transport processes.

We analysed all purse seine trips conducted in 2019 across the eight areas covered by the spatial sampling grid in Figure 1. Results from this analysis are presented in Table 1. High-ranking vessels are highlighted in yellow, with the location of vessel tracks and fishing activity from the highest-ranked vessel per area (marked with an * in Table 1) shown in Figure 2.

Vessels suitable for our sampling needs were found in most areas, particularly areas 1, 5, 6 and 7, yet our results also suggest that obtaining samples in some areas might be easier than others (Table 1). For some areas (i.e. areas 2, 3, 4 and 8), we will likely need to select >1 vessel per area to obtain adequate samples, as entries in the key field of ‘Area %’ are low (see Table 1).

Table 1. Summary of 2019 purse seine vessel trips across the spatial sampling grid in Figure 1. The top four ranked vessels for each of the areas 1 to 8 are highlighted in yellow. Vessel names have been removed for the purposes of this exercise. *, highest-ranked vessel per area.

2019	Vessel name	No. trips	No. of sets	Area %	Dep. port %	Arr. port %	Dep. = Arr.
Area 1	Anon.	1	181	98	AT SEA: 100%	AT SEA: 100%	Y
	Anon.	1	181	76	AT SEA: 100%	AT SEA: 100%	Y
	Anon.	No info.	No info.	No info.	No info.	No info.	No info.
	* Anon.	4	182	64	SHIMIZU: 25%	YAIZU: 50%	N
	Anon.	7	179	61	YAIZU: 42%	YAIZU: 42%	Y
	Anon.	7	139	58	SHIZUOKA: 42%	SHIMIZU: 57%	N
	Anon.	6	107	57	SHIMIZU: 33%	YAIZU: 50%	N
	Anon.	11	196	55	POHNPEI: 72%	POHNPEI: 63%	Y
	Anon.	6	149	55	YAMAGAWA: 83%	YAMAGAWA: 66%	Y
Area 2	Anon.	9	169	55	YAIZU: 33%	YAIZU: 66%	Y
	Anon.	10	203	42	TARAWA: 60%	TARAWA: 50%	Y
	Anon.	8	219	38	MAJURO: 87%	MAJURO: 87%	Y
	Anon.	9	157	34	MAJURO: 88%	MAJURO: 88%	Y
	*Anon.	8	237	34	MAJURO: 100%	MAJURO: 87%	Y
	Anon.	10	171	33	MAJURO: 90%	MAJURO: 60%	Y
	Anon.	6	199	33	MAJURO: 100%	MAJURO: 100%	Y
	Anon.	16	231	33	TARAWA: 87%	TARAWA: 81%	Y
	Anon.	7	153	33	MAJURO: 85%	MAJURO: 85%	Y
Area 3	Anon.	13	170	32	MAJURO: 30%	TARAWA: 30%	N
	Anon.	10	136	32	MAJURO: 70%	MAJURO: 70%	Y
	*Anon.	12	166	33	MAJURO: 66%	MAJURO: 66%	Y
	Anon.	6	113	32	BUSAN: 16%	TARAWA: 33%	N
	Anon.	8	225	30	MAJURO: 50%	MAJURO: 62%	Y
Area 3	Anon.	8	162	30	PAGO PAGO: 37%	PAGO PAGO: 37%	Y
	Anon.	7	188	27	MAJURO: 57%	MAJURO: 57%	Y

	Anon.	4	122	25	MAJURO: 100%	MAJURO: 75%	Y
	Anon.	10	160	25	MAJURO: 40%	MAJURO: 40%	Y
	Anon.	11	232	24	TARAWA: 63%	TARAWA: 54%	Y
	Anon.	12	191	24	NELSON: 33%	NELSON: 25%	Y
	Anon.	6	160	24	PAGO PAGO: 83%	PAGO PAGO: 66%	Y
Area 4	*Anon.	6	209	36	POSORJA: 83%	POSORJA: 66%	Y
	Anon.	7	163	32	PAGO PAGO: 100%	PAGO PAGO: 71%	Y
	Anon.	7	181	30	MANTA: 28%	PANAMA: 42%	N
	Anon.	5	161	30	LA UNION: 60%	LA UNION: 40%	Y
	Anon.	5	196	30	POSORJA: 100%	POSORJA: 100%	Y
	Anon.	6	157	29	POSORJA: 66%	POSORJA: 66%	Y
	Anon.	6	179	26	PAGO PAGO: 33%	PANAMA: 33%	N
	Anon.	16	160	26	TARAWA: 56%	TARAWA: 56%	Y
	Anon.	6	166	25	MANTA: 66%	MANTA: 50%	Y
	Anon.	5	209	23	MANTA: 80%	MANTA: 80%	Y
Area 5	Anon.	4	101	100	LAE: 100%	LAE: 100%	Y
	Anon.	4	157	100	LAE: 100%	LAE: 75%	Y
	Anon.	32	217	92	NORO: 59%	NORO: 65%	Y
	*Anon.	20	207	92	NORO: 90%	NORO: 75%	Y
	Anon.	22	155	90	NORO: 50%	NORO: 63%	Y
	Anon.	10	202	90	LAE: 90%	LAE: 90%	Y
	Anon.	13	225	89	WEWAK: 46%	WEWAK: 46%	Y
	Anon.	26	172	89	NORO: 53%	NORO: 65%	Y
	Anon.	11	208	89	LAE: 54%	LAE: 54%	Y
	Anon.	9	129	88	LAE: 33%	LAE: 33%	Y
Area 6	Anon.	9	193	81	TARAWA: 100%	TARAWA: 100%	Y
	Anon.	12	219	80	TARAWA: 66%	TARAWA: 66%	Y
	Anon.	5	115	78	MAJURO: 80%	MAJURO: 60%	Y
	Anon.	13	162	75	TARAWA: 46%	TARAWA: 46%	Y
	*Anon.	12	222	73	TARAWA: 100%	TARAWA: 91%	Y
	Anon.	13	228	72	TARAWA: 76%	TARAWA: 76%	Y
	Anon.	8	128	72	MAJURO: 37%	MAJURO: 37%	Y
	Anon.	7	166	72	MAJURO: 100%	MAJURO: 85%	Y
	Anon.	11	207	71	MAJURO: 45%	MAJURO: 45%	Y
		Anon.	12	224	70	TARAWA: 100%	TARAWA: 91%
Area 7	*Anon.	6	143	63	PAGO PAGO: 100%	PAGO PAGO: 83%	Y
	Anon.	4	141	62	PAGO PAGO: 100%	PAGO PAGO: 75%	Y
	Anon.	11	176	59	PAGO PAGO: 45%	PAGO PAGO: 36%	Y
	Anon.	12	163	56	KIRITIMATI: 41%	KIRITIMATI: 33%	Y
	Anon.	7	91	53	HONIARA: 28%	HONIARA: 28%	Y

	Anon.	13	193	52	KIRITIMATI: 23%	KIRITIMATI: 15%	Y
	Anon.	15	202	47	KIRITIMATI: 20%	TARAWA: 20%	Y
	Anon.	5	108	46	PAGO PAGO: 40%	PAGO PAGO: 40%	Y
	Anon.	6	209	44	MAJURO: 50%	MAJURO: 33%	Y
	Anon.	12	216	44	TARAWA: 41%	TARAWA: 41%	Y
Area 8	Anon.	13	211	30	KIRITIMATI: 38%	TARAWA: 38%	N
	Anon.	8	157	24	MAJURO: 50%	MAJURO: 62%	Y
	Anon.	4	154	23	PAGO PAGO: 100%	PAGO PAGO: 75%	Y
	Anon.	4	116	22	BUSAN: 25%	ZHOUSHAN: 50%	N
	Anon.	5	212	22	LA UNION: 80%	LA UNION: 60%	Y
	Anon.	11	241	21	TARAWA: 36%	KIRITIMATI: 27%	N
	*Anon.	6	123	21	PAGO PAGO: 83%	PAGO PAGO: 66%	Y
	Anon.	12	209	20	TARAWA: 41%	TARAWA: 41%	Y
	Anon.	12	233	19	KIRITIMATI: 41%	TARAWA: 50%	N
Anon.	24	228	18	TARAWA: 37%	TARAWA: 37%	Y	

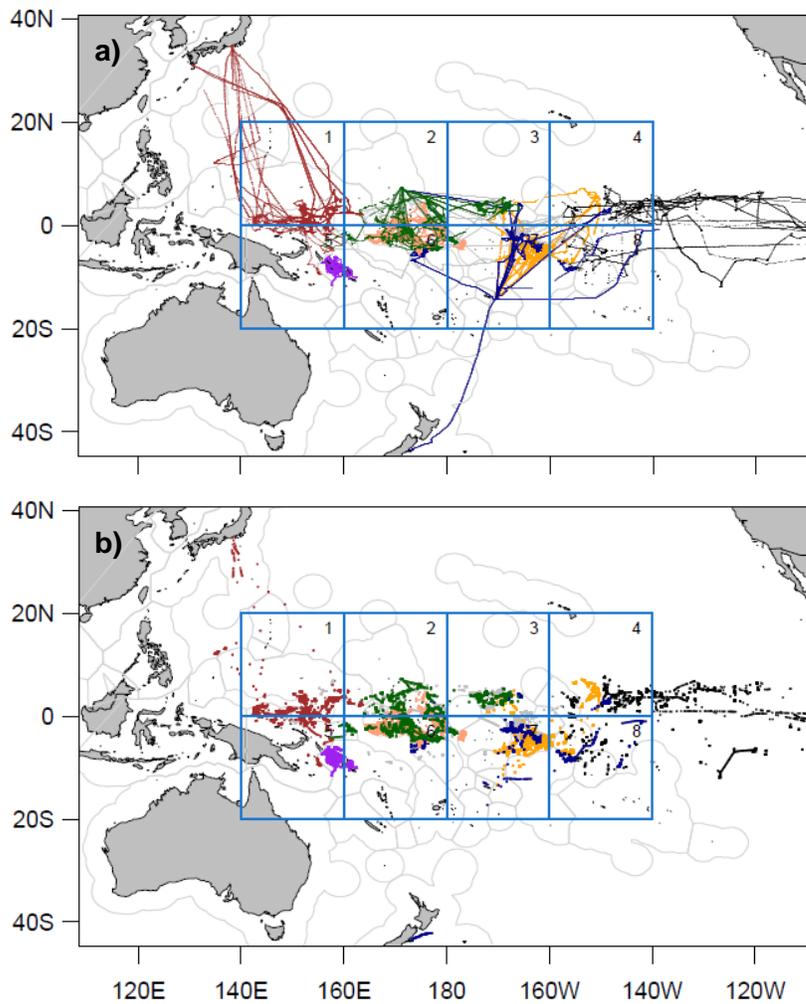


Figure 2. 2019 VMS tracks from the eight highest-ranking purse seine vessels (marked with an * in Table 1), with the spatial sampling grid of Figure 1 overlaid. Each vessel's track is represented by different coloured symbols, with panel a) denoting positions of all activities (i.e. transit, fishing, and in-port) at 30-min to 1-hour intervals in 2019, and panel b) showing positions of fishing activities only. The highest ranked vessel for area 1 is shown in brown; area 2 - dark green; area 3 - grey; area 4 - black; area 5 - purple; area 6 - light salmon; area 7 - orange; area 8 - dark blue.

3) Engage relevant fisheries authorities and fishing companies

After identifying the subset of high-ranking vessels from step 2, contact relevant fisheries authorities, fishing companies and national observer programmes (NOPs).

- a. Explain the research and sampling plans, the intended use of the data and SPC's expectations of the fishing authorities and companies.
- b. May need to establish formal contracts (ToRs or MOUs) between SPC and the fisheries authorities and fishing companies, with incentives for participating in the project (see Table 2).
- c. Observer coordinators, observer debriefers and observers must be also be briefed on the approach and incentives for participation mapped out.
- d. All of the above points must be completed well ahead of time.

4) Inspect VMS tracks

Once relevant parties have been contacted and agreements signed, inspect VMS tracks during the defined 1-week sampling period to determine vessel positions.

5) Contact fishing vessels

When vessel positions align with the intended spatial scale of sampling (see step 1) SPC then makes contact with the relevant fishing vessels by phone and email and gives the word to collect samples from the next set.

Ideally, the observer debriefer and/or the on-board observer would be contacted directly via email, potentially sent to hand-held GPS devices distributed to each observer, as described in Agenda Item 13 at ROCW 20.

6) Sample collection and storage

Once contact has been confirmed, then:-

- a. The observer sets aside 30 SKJ and/or 30 YFT whole, from the next purse seine set.
- b. Fish size is not critical, but ideally the length range should be representative of the range captured in that set.
- c. Place fish in large, sealable plastic bags (separated by species) along with labels recording basic metadata (i.e. species, position, date and time, vessel name, observer name).
- d. Place bags in freezer immediately, preferably in 'dry' freezers or alternatively in a brine well, and store frozen for the remainder of the fishing trip.
- e. Upon reaching port, samples are offloaded with the observer, coordinated by the observer debriefer, and housed frozen in a storage facility on shore awaiting later processing.

7) Port sampling and shipment

- a. Following the offload and storage of fish at port, SPC staff, in collaboration with in-country port samplers, visit said port and process whole fish on site.
- b. All standard sample types (e.g. muscle and fin clips for genetics, otoliths for ageing, chemistry and shape analyses, stomachs, gonads, liver, dorsal spines) will be obtained from each fish.
- c. At the conclusion of port sampling, SPC staff will arrange transport of samples, ideally carrying samples back with them to Nouméa for storage, archiving and future analyses.

Table 2. Some advantages and disadvantages of implementing Approach 1: VMS bag and store.

Advantages	Disadvantages
<ul style="list-style-type: none"> • <u>General</u>: Systematic, repeatable, cost- and time- efficient sampling approach. • <u>Sampling and logistics</u>: Spatial and temporal consistency in sample collection guaranteed at the scales we need for the ongoing stock structure work, and for other ecosystem-related studies. This level of sampling precision not possible to date. Removes current issues with patchy sample coverage and low sample return rates to the PMSB. For e.g., in 2018, 351 fish were sampled by observers on PS or LL during 386 days at sea, which is less than 1 fish sampled per day. All possible sample types (e.g. muscle and fin clips for genetics, otoliths for ageing, chemistry and shape analyses, stomachs, gonads, liver, dorsal spines) are obtained from each fish. The number of samples needed is pre-defined, making it simpler to coordinate logistics and budgeting. Sampling approach can be tailored to different applications and projects. • <u>Sample quality</u>: Sample quality, and importantly, consistency in quality among regions, is ensured. Onshore sampling has many advantages over sampling at sea in terms of obtaining high quality, undamaged, uncontaminated samples in a fast and efficient manner. • <u>Motivation and workloads</u>: Current challenges regarding observer motivation for biological sampling and resultant low sample recovery rates become non-issues. Simplifies tuna sample collection on purse seine vessels. Achieved through removing the need for at-sea biological processing of tuna samples. We continue to utilise the established network of observers for biological sampling, but this approach 	<ul style="list-style-type: none"> • <u>Logistics</u>: A new approach, so potential for unforeseen logistical issues. Limited freezer space on board vessels and in ports. Possible bottleneck during offloading in some ports. How best to get samples from the ship to the storage facility in good condition? Change in protocol for purse seine observers. Questions around how best to make this happen and to communicate our objectives. Risk of having messages to observer not relayed, misunderstood or ignored. Verification of fish sample collection on board re: date and time is essential. Photographic evidence needed (can be obtained from camera, tablet, or phone). Note that similar issues exist with the current sampling protocol. Transport of potentially a large amount of frozen samples (might not be possible/expensive with certain airlines); potential problems with storage in quarantine in transit countries. Uncertainty over Nagoya Protocol regulations in terms of obtaining samples for genetic analyses. • <u>Samples</u>: Blood samples not available from frozen fish. Gonads will be slightly defrosted when collected in port which may impact sample quality.

<p>would allow a refocus towards other priorities (e.g. bycatch, discards).</p> <p>Ease observer debriefer and coordinator workloads. As motivation for sampling is largely removed from the equation under this approach, this reduces time and energy required for coordination efforts regarding sample collection and shipping.</p> <p>Importantly, setting aside of fish from one set would constitute the observers' only tasks in relation to tuna sampling from that trip.</p> <ul style="list-style-type: none"> • <u>Training and capacity building:</u> Opportunities for SPC to build new relationships and collaborations with industry and fisheries authorities around research that will ultimately benefit these actors, and to improve upon existing relationships. • <u>Data recording:</u> The 'Onshore' app could be used for data entry (SPC team are currently finalising on a new version that includes gears other than LL). Simplifies data entry and validation procedures - much easier/faster (linked to Tufman2). May be possible to have a debriefing plug-in for Onshore. • <u>Equipment:</u> Simplifies purchasing and coordination, and lowers costs. No need for special labels (e.g. Hallprint), with standard paper labels ok. Most of the equipment (i.e. bags for fish storage) can be purchased locally, so no need for purchase and distribution coordination. As sampling is standardised and pre-planned, equipment needs can be estimated and gear purchased in bulk well ahead of time. • <u>Finance and incentives:</u> The new approach is more efficient financially, ensuring a higher sample return rate per dollar spent than the current strategy. <p>At present, one day at sea = 5 USD bonus per observer. In 2018, with 351 fish sampled from 386 days at sea, the cost was 5.50 USD per fish.</p>	
---	--

<p>A possible break down of costs under the new approach could something like:-</p> <ol style="list-style-type: none"> 1) Per trip payment to fishing companies for purchase of fish (i.e. 100 fish @ 5 USD per fish). 2) Bonus for the observer on board to secure the samples (e.g. 40 USD per trip for successfully setting aside and storing whole fish). 3) Payment per fish processed by the port sampler (e.g. 4 USD per fish). This is easier to manage, allows work to be done in the port sampler's own time. <p>See Table 3 below for initial cost estimates for a proposed trial of the approach, originally scheduled to take place in early 2020, but now delayed due to the COVID-19 situation.</p> <ul style="list-style-type: none"> • <u>Nagoya Protocol</u>: Is purchasing the fish from the fishing company a viable workaround regarding Nagoya protocol regulations? 	
--	--

Approach 2: VMS Widget

This approach involves at-sea biological sampling, making use of a tissue-sampling tool called the 'Widget', developed over a numbers of years by CSIRO, Australia (see Bradford et al. 2016 for details of early testing). The Widget allows us to obtain clean, small (i.e. 1.5 cm), contamination-free muscle tissue samples from fresh fish on board the vessel, which can then be frozen individually or stored in a preservative solution on board, guaranteeing sample quality for downstream genetic analyses.

This approach may require the procurement of a cadre of experienced 'super observers' to receive a short, specialised training in the use of the Widget and to conduct at-sea biological sampling aboard purse seine vessels.

Implementation

The approach closely follows implementation steps 1 to 5 as outlined for Approach 1: VMS bag and store. It differs from Approach 1 primarily in how observers collect samples at sea, and where sample processing occurs. We explain the implementation process and these differences in a–f below.

- a. Following vessel and port selection (step 2), just prior to the defined sampling times (i.e. 1-week, quarterly), SPC in collaboration with NOPs send a suite of eight trained observers to selected ports – observers well briefed on SPC's sampling needs, and each armed with CSIRO Widgets.
- b. These observers then board the selected vessels and, when contacted by SPC (step 5 in Approach 1), go ahead and sample 30 SKJ and/or 30 YFT from the next successful set. As in Approach 1, fish size is not critical, but ideally the length range should be representative of the range captured in that set.

- c. The observer is then tasked with spending one full day on biological sampling and processing fish from that set, collecting muscle samples from each fish using the Widget, in addition to all other biological samples needed for scientific use (i.e. otoliths, gonads, stomachs, dorsal spines, liver).
- d. Standard metadata (i.e. species, position, date and time, vessel name, observer name) would be required for each sample.
- e. Muscle samples are then stored frozen, or in preservative solutions such as RNAlater® (<https://www.sigmaaldrich.com/catalog/product/sigma/r0901?lang=en®ion=NC>) or DNAgard® (<https://www.sigmaaldrich.com/technical-documents/protocols/biology/dnagard-tissue.html>) on board and offloaded with the observer at port, with samples shipped to SPC as per existing biological sample transport protocols.
- f. Importantly, the above tasks would constitute the observers' only tasks in relation to tuna sampling from that trip.

SPC recommends that **Approach 2: VMS Widget** represents the best strategy for achieving tuna sampling objectives in the mid- to long-term. However, given uncertainties around the timeframe for availability of the CSIRO Widget on a large scale¹, the best interim approach may be to begin with **Approach 1: VMS bag and store**, gauge its success and feasibility, then transition to Approach 2, or a combination of approaches, when the Widget becomes more widely available.

A trial of Approach 1: VMS bag and store

Following the meetings on 24 October and 29 November 2019, all parties identified the need for a thorough trial of Approach 1 before considering rolling it out as a standard sampling scheme. The main objectives of this trial are to test the feasibility of the method across different regions, ports and fishing companies, expose any flaws, bottlenecks and unforeseen issues, and highlight areas where further work is required.

After presenting our plans at both observer training workshops in Nouméa in November 2019 and receiving feedback from workshop attendees, it was resolved that we should run the trial after gaining approval from the regional observer coordinators at ROCW 20. As mentioned, the trial was formally endorsed by ROCW 20, and members' feedback on both the oral presentation and working document has been incorporated into this information paper.

One possible plan and timeline for the trial is outlined below. The plan follows the seven steps set out in the **Implementation** section for Approach 1 (see above).

¹ Note that CSIRO has granted SPC-OFP access to the Widget for collection of tissue samples from bigeye and skipjack tuna on the upcoming CP14 tuna tagging cruise through the Kiribati EEZ (see SPC-OFP 2020 for further details). This will represent the first application of the Widget to WCPO tunas.

Plans and timeline

Species: SKJ
Spatial scale: 3 locations: areas 2, 5, 7 (see Figure 3)
Temporal scale: Initially, planned for a 1-week period in May 2020, but now delayed due to COVID-19 restrictions.
Numbers: 30 fish per location

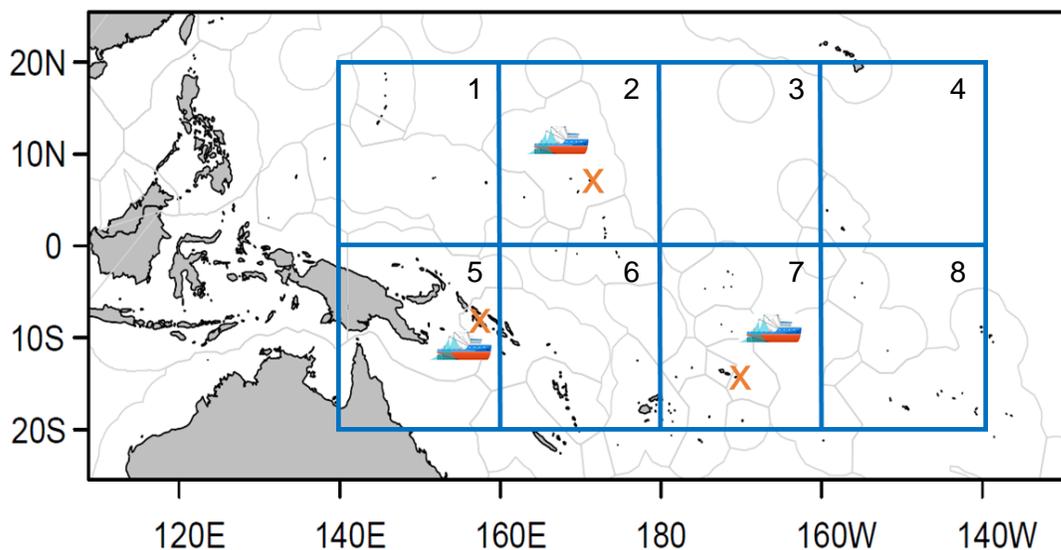


Figure 3. Proposed spatial sampling grid and possible purse seine set locations (boats) in areas 2, 5 and 7 during the trial sampling period (i.e. 1 week, following resumption of normal observer duties on purse seiners). Orange crosses in areas 2, 5 and 7, denote the intended offloading ports in Majuro, Republic of the Marshall Islands (RMI), Noro, Solomon Islands, and Pago Pago, American Samoa, respectively.

For the trial, we have selected the following three ports and three vessels to test the success of the sampling, offloading and storage procedures (Figure 3). These selections were based on results presented in Table 1 and existing relationships between SPC and fishing companies operating out of these ports. Note that ports and vessels may change in the lead up to the trial subject to fishing company agreements.

Area 2: Port = Majuro, RMI; Vessel: see * in Table 1, dark green VMS tracks in Figure 2

Area 5: Port = Noro, Solomon Islands; Vessel: see * in Table 1, purple VMS tracks in Figure 2

Area 7: Port = Pago Pago, American Samoa; Vessel: see * in Table 1, orange VMS tracks in Figure 2

Estimated costs

There are still several details to work through regarding payments to fishing companies for agreeing to participate and incentives for observers to set aside samples. Table 3 provides a coarse cost estimate for implementing the proposed trial of Approach 1.

Table 3. Estimated costs of trial (excl. flights, salaries and fishing company engagement).

Item	Quantity	Cost (USD)	Total (USD)
Fishing company engagement	3 companies	TBD	TBD
Flights	Nou-Noro-Maj-Pago-Nou	TBD	TBD
Salaries	2 SPC staff	In kind	In kind
Observer bonus	3 trips	40/trip	120
Fish purchase	90 SKJ	5/fish	450
Port sampling	90 SKJ	4/fish	360
Shipment	3	250/shipment	750
Total			1680

Conclusions and next steps

There is a clear need for new sampling approaches to answer questions on South Pacific tuna stock structure and population dynamics at the scales needed for effective management of regional tuna resources. Observers aboard purse seiners are critical to this process, and any tuna biological sampling programme should ideally be designed to maximise benefits to science from the large efforts made by the observers, observer debriefers and observer coordinators in obtaining samples.

The two new sampling approaches we outline here are designed with this in mind. They both offer improvements upon the current sampling scheme in terms of addressing biological and ecological knowledge gaps for tunas. This is achieved by ensuring systematic sampling coverage and replication across space and through time, increasing the number and building confidence in the quality of samples returned to the PMSB, simplifying observer duties related to tuna biological sample collection and streamlining sample transport, archiving and database management processes.

We stress that Approaches 1 and 2 can complement each other, and could be used interchangeably depending on vessel-, area- and port-specific factors. They can also add to, rather than replace existing biological sampling protocols.

SPC's view is that these new approaches are simply an avenue for enhancing and simplifying tuna biological sampling to meet WCPFC requirements, and for augmenting tuna sample holdings in the WCPFC Tuna Tissue Bank, embedded within the PMSB (SPC-OFP 2020). With regard to addressing tuna stock structure questions per se, these approaches must be considered in light of a broader 'design study' currently underway (see below), which will help guide final sampling decisions and protocols. One idea might be to use the PEUMP programme as a test case to assess the utility of the approaches in the first instance, and if successful, then at some point in the future the approach may be adopted as the standard way for observers to collect tuna samples on purse seine vessels. This strategy is open for further discussion.

In terms of planned actions, there are several tasks to progress over the coming months. These include:-

- Continue work on a 'design study' to define the analytical tools, sampling coverage and investment required to resolve longstanding stock structure and population connectivity questions for these species (see Anderson et al. 2020). A key component of this work involves simulating data for the analytical tools (e.g. genetic and otolith markers) we plan to employ that reflect plausible stock structure hypotheses as outlined in Moore et al. (2020) for the species of interest. This, in turn, provides us quantitative benchmarks for guiding later sample collections, both onboard vessels and in port.

- In conjunction with the design study, develop a tissue sampling and processing protocol that eliminates concerns about cross-contamination of samples and allows for highest possible confidence in inferences made using genetic and other molecular analyses.
- Shaped by the outcomes of the design study, continue to hone strategies to inform fisheries authorities, fishing companies, observer coordinators, trainers and debriefers and in-country port samplers about the new approaches, their new responsibilities within it, and to gain continuous feedback.
- Develop appropriate TORs, MOUs and payment strategies for fishing company engagement, fish purchasing, observer duties and port-sampler payments. Table 3 lists some ballpark figures, but these need refinement.
- Further investigation is needed into sampling permit and Nagoya Protocol requirements.
- Extending / reworking the sampling approaches for longline vessels would be useful. Approach 2 may be feasible with some fine-tuning.

References

- Anderson, G., Hampton, J., Macdonald, J., Nicol, S. and Scutt Phillips, J. 2020. The application of genetics and genomics to Pacific Fisheries by SPC and implications for the WCPFC Tuna Tissue Bank. WCPFC-SC16-2020/SC-IP-?? presented to the Sixteenth Regular Session of the Scientific Committee of the Western and Central Pacific Fisheries Commission. Online meeting.
- Bradford, R.W., Hill, P., Davies, C. and Grewe, P. 2016 A new tool in the toolbox for large-scale, high-throughput fisheries mark-recapture studies using genetic identification. *Mar. Freshwater Res.* 67, 1081–1089. <https://doi.org/10.1071/MF14423>
- Macdonald, J., Moore, B. and Smith, N. 2019. Stock structure considerations for Pacific Ocean tunas. Information paper WCPFC-SC15-2019/SA-IP-03 presented to the Fifteenth Regular Session of the Scientific Committee of the Western and Central Pacific Fisheries Commission, Pohnpei, Federated States of Micronesia.
- Moore, B.R., Adams, T., Allain, V., Bell, J.D., Bigler, M., Bromhead, D., Clark, S., Davies, C., Evans, K., Faasili Jr. U., Farley, J., Fitchett, M., Grewe, P.M., Hampton, J., Hyde, J., Leroy, B., Lewis, A., Lorrain, A., Macdonald, J.I., Marie, A.D., Minte-Vera, C., Natasha, J., Nicol, S., Obregon, P., Peatman, T., Pecoraro, C., Phillip Jr. N.B., Scutt Phillips, J., Pilling, G.M., Rico, C., Sanchez, C., Scott, R., Stockwell, B., Tremblay-Boyer, L., Usu, T., Williams, A.J. and Smith, N. 2020. Defining the stock structures of key commercial tunas in the Pacific Ocean II: Sampling considerations and future directions. *Fish. Res.* <https://doi.org/10.1016/j.fishres.2020.105524>
- SPC-OFP 2020. Project 35b: WCPFC Tuna Tissue Bank. Research Paper WCPFC-SC16-2020/RP-P35b-01 presented to the Sixteenth Regular Session of the Scientific Committee of the Western and Central Pacific Fisheries Commission. Online meeting.

Acknowledgements

We thank all participants at the observer debriefer, trainer and coordinator workshops, and Pierre Feutry, Karen Evans, Jess Farley, Peter Grewe and Campbell Davies (CSIRO) for their interest and input on the new sampling approaches and for their feedback on this document. Funds for travel and workshop attendance in Funafuti and Hobart were provided under PEUMP KRA 1.4 – ‘Research to address biological uncertainties in tuna stock assessment models’.