

EXPLORATION OF ALTERNATIVE DESIGNS FOR EASTERN BLUEFIN TUNA CLOSE-KIN MARK RECAPTURE (CKMR)

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SUMMARY

Alternative options for potential sampling designs suitable for BFT-E CKMR are explored. Seven designs are considered that differ in terms of total sample size and in the breakdown of samples across fisheries. The effect of using genetic samples collected in the Atlantic and Mediterranean in recent years is also investigated. This document extends and uses the work developed in Bravington and Fernández, 2024.

RÉSUMÉ

Des options alternatives pour de potentielles conceptions d'échantillonnage adaptées au modèle CKMR pour le BFT-E sont étudiées. Sept conceptions sont examinées et diffèrent en termes de taille totale des échantillons et de ventilation des échantillons parmi les pêcheries. L'effet de l'utilisation des échantillons génétiques collectés dans l'Atlantique et la Méditerranée ces dernières années est également étudié. Le présent document élargit et utilise les travaux élaborés par Bravington et Fernández, 2024.

RESUMEN

Se exploran otras opciones para posibles diseños de muestreo adecuados para el CKMR del atún rojo del este. Se tienen en cuenta siete diseños que difieren en cuanto al tamaño total de la muestra y al desglose de las muestras entre pesquerías. También se investiga el efecto de utilizar muestras genéticas recogidas en el Atlántico y el Mediterráneo en los últimos años. Este documento amplía y utiliza el trabajo desarrollado en Bravington y Fernández, 2024.

KEYWORDS

Close-Kin Mark-Recapture (CKMR); design; abundance estimation; bluefin tuna

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1. Introduction

This report addresses the 3 Subtasks identified in Article 1 Contract Amendment no. 1 to the Short-Term Contract of the Model-Base Sampling Design for Eastern Bluefin Tuna Close-Kin Mark Recapture (ICCAT GBYP 01/2024) of the Atlantic-wide Research Program for Bluefin (ICCAT GBYP Phase 13). Those subtasks are:

1. Based on the feedback obtained during the BFT Species Group Intersessional Meeting in April 2024, investigate alternative sampling scenarios (i.e. designs) and, in particular, focus on the possibility of using genetic samples collected in the Atlantic and Mediterranean in recent years. Furthermore, show the value of adding more years of data.
2. Implement any refinements to the CKMR model that may be relevant for the analysis and doable for July.
3. Collaborate with the BFT Technical Sub-group on CKMR, which will be coordinating the complete feasibility study that will integrate results and information on epigenetic ageing, genetic techniques, logistic considerations, costs, etc.

2. Exploration of alternative designs

This section addresses Subtask 1: “Based on the feedback obtained during the BFT Species Group Intersessional Meeting in April 2024, investigate alternative sampling scenarios (i.e. designs) and, in particular, focus on the possibility of using genetic samples collected in the Atlantic and Mediterranean in recent years. Furthermore, show the value of adding more years of data.”

We recall that in the work presented to the BFT Species Group Intersessional Meeting in April 2024, a “basic design” was used for illustrative purposes, explained in Section 4 of document SCRS/2024/053 presented at that meeting. It was based on the number of samples indicated in Tables 3 and 4 of that document, which we reproduce here as **Table 1**.

From the feedback received by the BFT Species Group and its Technical Sub-group on CKMR, it was decided that an attempt should be made to examine and use, to the extent possible, already-existing information (i.e. tissue samples of fish that have already been collected and could be appropriate for genotyping and CKMR analysis). This implies considering the use of larvae preserved in ethanol from the Balearic survey, available since 2019, and bigger fish sampled since 2020 which could be used as potential parents in POP checks with larvae from the Balearic survey and, possibly, also as potential offspring in POP checks.

Available samples from recent years

Balearic larval survey (Wlar)

Larvae from this survey since 2019 had already been incorporated in the “basic design” presented in SCRS/2024/053, although only approximate numbers of larvae were known at that time. Detailed discussions with the survey experts after the Malta (April 2024) meeting provided additional insights about the samples available from this survey, as presented below.

Table 2 indicates the number of larvae found in each survey during 2019-2023. The surveys are run on a systematic grid using Bongo 500 nets, and two identical collectors are used, with the larvae from one of the collectors preserved in formalin and the larvae from the other one preserved in ethanol. The difference in the number of sampled stations between years depends on logistic constraints, in particular, number of ship days and ship used. The total number of larvae found in each survey also depends on environmental variables. Whereas the larvae preserved in formalin have all been separated and counted, as they are used for the larval index calculation, only a limited number of the larvae preserved in ethanol (which are the ones that would be used for genetic analysis) has been separated and counted so far.

Based on this information, the following settings could be considered in the CKMR design: 2822 larvae from the 2019 survey, 2250 larvae from the 2020 survey, and up to 8000 larvae appears to be possible for each year from 2022 onward. There was no survey in 2021.

As was the case in SCRS/2024/053, for all the CKMR results shown in this report (i.e. results in terms of expected number of “meaningful” kin pairs, and precision of estimates obtained from the CKMR analysis), the number of samples from the larval survey have been multiplied by an arbitrary 0.5 factor, to account for intra-cohort sibship in larval samples. However, all the tables that present sample sizes show the actual number of larvae that would be collected and genotyped, as this is what will affect logistics and costs.

To investigate the extent of intra-cohort sibship in larval samples, an analysis is planned with approximate 4000 larvae taken from the 2022 survey. The results will provide useful insights to improve the CKMR design and statistical analysis.

West Atlantic fisheries (Canada and USA fisheries)

Table 3 summarizes the amount of samples available since 2020, as provided by relevant experts.

Although the information on the proportion of fish of Eastern origin (EBFT) for years 2020 onwards is not yet available, based on what was found in samples from preceding years (2016-2018 for Canada and 2016-2017 for USA), we assume this proportion to be 61% for USA and 47% for Canada. The age composition of the samples differs between USA and Canada, as shown in **Figure 1**.

The different age composition between USA and Canada samples is taken into account in the analyses presented here.

For 2023-2025, relevant experts suggested that about 2000 samples from USA and 1200 from Canada may be expected per year. Using the proportion of EBFT in the samples of 61% for USA and 47% for Canada, this would result in 1225 and 567 fish of EBFT, respectively.

East Atlantic fisheries

The samples available in the GBYP tissue bank were facilitated to us by relevant experts (with date 3 June 2024). From the database, we considered the samples for which either muscle or fin tissue is available, collected since year 2020, for which an estimated birth year is provided in the database, and such that the fish were of age ≥ 3 when caught.

We first considered all samples with these characteristics in the entire East Atlantic, i.e. corresponding to FAO areas 27 (Northeast Atlantic) or 34 (Central Eastern Atlantic). We then also considered specifically the available samples from Traps in the Strait of Gibraltar area (in the Atlantic) and the Norwegian PS fishery, as these fisheries had been previously identified as potential sources of adult samples.

Although a few of these fish, which were caught in the East Atlantic, will be of Western origin (WBFT), which will reduce the actual number of EBFT available for CKMR, we anticipate that the proportion "lost" will be negligible in the context of the overall EBFT CKMR model. **Table 4** summarizes the available information.

The proportions at age in each year, and averaged over the years, are shown in **Figures 2** and **3**.

Mediterranean fisheries

Unlike samples from Atlantic fisheries, samples from juvenile and adult Mediterranean fisheries are likely not "well-mixed" with respect to spawning-ground usage (i.e. they do not equally represent all EBFT fish). Thus, they may be of less short-term value to EBFT CKMR than Atlantic samples, because they do not lead as directly to an overall EBFT abundance estimate. Mediterranean samples would nevertheless be highly informative about spatial structure between Mediterranean spawning grounds and, depending on the outcome on this issue (if investigated), they might offer a particularly cost-effective way to sample for general EBFT CKMR in the longer term. In the shorter term, though, the BFT Species Group considered that Mediterranean non-larval samples would be less of a priority than Atlantic adults and Balearic larvae.

Despite this appreciation, we checked if there were tissue samples from *Wad* or *Cad* fisheries available in the GBYP database.

In the same way done for the East Atlantic fisheries, we considered the samples for which either muscle or fin tissue is available, collected since year 2020, for which an estimated birth year is provided in the database, and such that the fish were of age ≥ 3 when caught.

Regarding the *Wad* fishery, we checked for fish caught in PS fisheries in the “BA-Balearic and Catalan Sea” area. Regarding the *Cad* fishery, we checked for fish caught in PS fisheries in the “TY- Tyrrhenian Sea” or the “SIS-South/Strait of Sicily” or the “TU-Gulf of Gabes” areas. **Table 5** summarizes the available information.

The proportions at age in each year, and averaged over the years, are displayed in **Figure 4**.

Investigating alternative sampling designs for CKMR

Based on the feedback received from the Chairs of the BFT Species Group and Technical Subgroup on CKMR, we examine designs including “historical” data (i.e. fish with tissue samples already collected, as shown above).

For future years (meaning 2025 and onwards), we first focus on designs including data that could be collected during 2025 and 2026, with the aim of having a first analysis available in 2027 in time for the MSE review. The focus at that point will be to be able to provide a first estimate (which may be rough, but hopefully useful) of total EBFT adult population size.

At a later stage, and based on the experience gained until that time (i.e. with data collected until 2026), the sampling design may be modified appropriately to continue collecting data so as to be able to improve the precision of EBFT population estimates in future years, and to learn about issues such as possible stock structure within the Mediterranean that could be relevant from a management perspective.

We have explored 6 alternative designs (Designs 1 to 6), all of them with samples until 2026. The designs differ in the number of samples from various sources used in different years (see **Table 6**). The impact of extra years, i.e. extending sampling until 2030, is covered at the end of this section. In all cases, the age composition of the historical samples has been taken from that available in the databases (explained above) and for future samples (2025 onwards) similar age compositions to those of the historical samples have been assumed for each fishery.

For comparative purposes, we have also included in **Table 6** a “Design 0”, which essentially corresponds to the “basic design” in SCRS/2024/053, presented at the BFT intersessional meeting in April, which does not include historical data from any of the fisheries (only from the Balearic larval survey) and starts sampling in Atlantic and Mediterranean fisheries in 2025. The only difference with the “basic design” presented in April is that now the age compositions of the future adult fishery samples have been assumed similar to those in the historical samples, as explained in the previous paragraph.

For years 2025 and 2026, Designs 1-4 consider only samples from the Balearic larval survey and Atlantic fisheries (**Table 6**), because it is a reasonable working assumption at this stage that Atlantic fisheries will be “well-mixed” (particularly the traps in the Gibraltar area) and, therefore, samples from these fisheries are expected to be the most efficient in terms of getting an estimate of total EBFT population size in the short-term. As noted earlier in this document, 1225 and 567 EBFT samples are expected to be available from the USA and Canada fisheries, respectively, and in the different designs assume that 1000 or 2000 samples will be available per year from East Atlantic fisheries (mainly from traps in the Gibraltar area and possibly also from Norwegian PS).

We note, however, that in the long-term, this sampling strategy, focusing exclusively on Atlantic fisheries and Balearic larvae, would be somewhat risky, as there is no guarantee that Atlantic fisheries will be well-mixed. Therefore, for the longer term, samples from Mediterranean fisheries would be desirable, starting with the Croatia juvenile fishery (*CROjuv*) that can provide samples of juvenile fish which are not exclusively from the West Med spawning ground (as is the case for the Balearic larval survey). In addition, sampling from the *Wad* and *Cad* spawning ground fisheries in the West and Central Med may be reasonably easy and efficient (through sampling in farms) and they can provide useful future insights into whether there exists, or not, population substructure within the Med. For this reason, it may perhaps be useful trying to collect data already from Mediterranean fisheries, even if there is no guarantee of ever being able to genotype them.

In all the designs explored here, the adult samples are used as potential parents in POP comparisons, but also as potential offspring, if their age at capture does not exceed 10 years and their birth year is not before 2014, which is the initial year considered in the underlying Population Dynamics Model (PDM, described in Section 2 of SCRS/2024/053). We did not consider it appropriate to start the PDM used in the CKMR analysis in an earlier

year than 2014, because the CKMR estimates of population abundance would then become more strongly dependent on assumptions made in the PDM and the precision calculated for those estimates may be higher than warranted based on the information provided by the CKMR data.

Table 6 displays the number of samples collected and genotyped under each Design 0-6, for the period 2019-2026. **Table 7** displays the expected number of “meaningful” kin pairs for each of those designs, and **Table 8** summarizes the results in terms of the Coefficient of Variation (CV) expected for the total EBFT population SSB estimates from each design. Some more detailed tables are included in **Appendix 1** at the end of this document. To facilitate comparison between the different designs, **Table 8** also includes a summary of the total sampling and genotyping effort, whereas **Figure 5** presents a summary of results in graphical form.

Further description of Designs 1-6, with data up to 2026, is provided next:

Design 1

Design 1 uses historical samples available, or expected to be available, starting from 2019 for the Balearic larval survey (assuming 8000 samples each year since 2022), and starting from 2020 for the *Wad* fishery, *Cad* fishery and Atlantic fisheries. For the Atlantic fisheries, it uses the historical samples available from Gibraltar traps, Norwegian PS, USA and Canadian fisheries, combined.

For years 2025 and 2026, Design 1 uses only samples from the Balearic larval survey and Atlantic fisheries, as already indicated. For the Balearic larval survey it assumes 8000 samples each year. For the Atlantic fisheries it assumes 1225 and 567 EBFT samples per year from the USA and Canada fisheries, respectively, and 2000 samples per year from East Atlantic fisheries (mainly from traps in the Gibraltar area and possibly also from Norwegian PS).

Design 2

In order to save genotyping resources, Design 2 investigates the possibility of using a smaller number of larval samples, in particular, 4000 instead of 8000 larvae per year during 2022-2026, therefore reducing the number of larvae to be genotyped by 20000 over the 5-year period relative to Design 1. Design 2 is identical to Design 1 in all other aspects (**Table 6**).

Design 3

Since **Table 7** indicates that only a quite small number of POPs are expected involving the historical *Wad* and *Cad* samples used in Designs 1 and 2, consideration could be given to saving additional genotyping resources at this stage by not genotyping now any of the 2750 historical samples available from these fisheries (even if they could be genotyped in the future). Design 3 is identical to Design 2 in all other respects (**Table 6**).

The results in **Table 8** indicate that the expected CVs for the total EBFT SSB estimates are appreciably higher for Design 3 than for Design 1 and Design 2.

Design 4

Design 4 assumes that only 1000 samples could be collected from the East Atlantic fisheries during 2025-2026 (instead of the 2000 assumed in Designs 1-3). This could occur if, for example, there were difficulties in the initial years of a newly setup sampling scheme.

To compensate for the lower number of Atlantic samples genotyped in this design relative to Designs 1-3, the number of samples from the Balearic larval survey since 2022 is assumed to be 6000 per year (as opposed to 4000 in Design 2, although lower than the 8000 assumed in Design 1; see **Table 6**).

Design 5

Design 5 is identical to Design 4, but assumes that sampling also occurs in the Croatian farms (supplied by the *CROjuv* fishery) in 2025 and 2026. Because the sampling would occur at the time of harvesting the fish in the farm, which is usually around December - February approximately 18 months since the time of capture at sea, for this design we assume the catch years have been 2024 and 2025 (see **Table 6**, Design 5), which would correspond to sampling in the farms at harvest time at the end of 2025 (and/or beginning of 2026) and at the end of 2026 (and/or beginning of 2027), respectively for catch years 2024 and 2025.

The fish from the Croatia juvenile fishery are mainly of ages 2 and 3, including also some age 4 fish, when they are caught from the sea, and are likely to be partly-mixed fish from the three subpopulations (*W*, *C* and *E*), i.e. we cannot safely assume that they are pure *C*-origin fish, and the mixing proportions are unknown (and have to be estimated from the CKMR data, bringing in additional parameters that have to be estimated in the CKMR analysis). They nevertheless constitute a useful second source of juveniles that are not all of West Med origin and that help for the verification of the well-mixed assumption for Atlantic fisheries (as explained in SCRS/2024/053). Furthermore, in conjunction with other fisheries (including also other Med fisheries), *CROjuv* samples would let us deduce a great deal of information about spatial structure within the Med subpopulations (see Appendix of document SCRS/2024/053 for further explanation of these matters). Since the fishery catches some age-4 animals, it can also provide information about abundance in years prior to their catch year, starting from 2020 for fish caught in 2024 at the age of 4.

Design 6

Design 6 is identical to Design 5, but assuming that sampling and genotyping also occurs for the Mediterranean spawning ground fisheries (*Wad* and *Cad*) in 2025 and 2026.

Discussion on the results of Designs 0-6, sampling to 2026:

Table 8 and **Figure 5** aim to facilitate the comparison between the different designs explored.

The use of samples that have already been collected in recent years (Designs 1-6, as opposed to Design 0) allows to increase the precision (i.e. decrease the CV) of the total EBFT SSB estimates (**Table 8** and **Figure 5**). The only exception is Design 3, which uses the lowest number of samples overall (40923) and does not include any historical samples from Mediterranean fisheries. Design 3 is the only one, among Designs 1-6, that results in a higher CV than Design 0 for the estimate of SSB in 2026.

Design 2 uses the next lowest number of samples overall (43673) and results in increased precision (i.e. lower CV) of the SSB estimates relative to Designs 3 and 0. Designs 2 and 4 result in fairly similar CVs for the SSB estimates, despite the fact that Design 2 uses fewer samples overall than Design 4 (51673).

Design 5 results in substantially lower CV for the SSB estimates than Design 2. This is at the cost of using a larger sample size (55673).

Designs 1 and 6 use the largest number of samples (63673 each of them). Among these two designs, Design 6 provides slightly better results (i.e. lower CVs).

From these results, we may conclude that Design 2 is economical (in terms of total sample size) for a single "pinning-down" of EBFT abundance, with an expected CV of around 21% for the SSB in 2020 (increasing to 33% for the SSB in 2026). Design 5 results in appreciably lower CVs than Design 2, although at the cost of using larger sample sizes. Design 6 results in the lowest CVs of all designs explored, although those CVs are only very slightly lower than those obtained from Design 5.

Therefore, ignoring logistic considerations (which the coordinators of the complete feasibility study would have to consider), and assuming that costs are a linear function of the number of samples analyzed, Designs 2 and 5 (and possibly 6) appear as good candidates at this point.

If Design 2 was selected at this point, we would still consider it a good idea to collect *CROjuv* samples, if "cheap" to do, because this is a source of juveniles which do not suffer from the extra-sibship issue that affects larvae samples (and that results in a decreased precision of CKMR estimates). Furthermore, the inclusion of *CROjuv* fish helps for the verification of the well-mixed assumption for Atlantic fisheries. We note that sampling *CROjuv* fish now would not necessarily imply a commitment to genotype them at some point, but at least it would not preclude this option if later found desirable.

A similar comment applies to Balearic larvae collection, i.e. we consider it a good idea to keep collecting large numbers of those larvae (in ethanol), in case genotyping a larger number was found desirable later on.

Further efficiency gains could be achieved in future by fine-tuning the plan as we go.

Results for sampling schemes until year 2030

Here we investigate the effect of a longer sampling period, with samples collected until 2030. In CKMR, data collected in each new year can be compared (via POP and/or HSP checks) with data collected in all previous years, therefore also increasing the information value of the samples collected in previous years.

We illustrate this in the context of Design 5, although similar arguments would apply to the other designs.

Two extensions (V1 and V2) of Design 5, sampling to year 2030, are investigated (**Table 9**). In both extensions, the number of Balearic larvae and fish from Atlantic fisheries collected per year during 2027-2030 is the same as in Design 5 for 2025-2026. Furthermore, 2000 *CROjuv* fish are collected per year for the catch years 2026-2029, continuing the sampling started in Design 5 for the catch years 2024-2025. In addition, the extension V2 also uses 2000 samples per year during 2027-2030 for the *Wad* and the *Cad* spawning ground fisheries, which were not sampled under Design 5 in 2025 or 2026.

Table 9 displays the number of samples used in both extensions, and **Table 10** shows the expected number of “meaningful” kin pairs. The expected CVs for the resulting SSB estimates are presented in **Table 11**. To help understand the benefit gained by the additional years of sampling (to 2030), **Table 11** also shows the CVs expected from the original Design 5, using samples to 2026. More detailed tables are included in **Appendix 1** at the end of this document.

3. Refinements to the CKMR mathematical model

This section addresses Subtask 2: “Implement any refinements to the CKMR model that may be relevant for the analysis and doable for July.”

We made some technical adjustments to the population dynamics model and CKMR probability equations, relative to what was presented in the BFT Intersessional Meeting in Malta in April 2024. The adjustments had very little effect on final CVs for the estimates of overall abundance, but successfully resolved some unrealistic results we had previously noticed with the models, when estimating the extent of site fidelity using Mediterranean samples. The adjustments made concern how attainment of individual maturity is linked to selection of spawning ground. The final version of document SCRS/2024/053 is based on the updated model configuration, with these technical adjustments.

Furthermore, a question was raised in the BFT Intersessional Meeting in Malta about the probabilities for the fish selecting any of the 3 spawning grounds (the g vector, defined in document SCRS/2024/053), which were initially taken to be (0.4, 0.3, 0.3) for the Western, Central and Eastern Mediterranean, respectively. Based on feedback from the meeting participants, alternative values were tried, in particular (0.25, 0.5, 0.25) and (0.3, 0.4, 0.3), and provided very similar results. The results in the final version of document SCRS/2024/053 are presented for the configuration (0.3, 0.4, 0.3).

4. Collaboration with the BFT Technical Sub-group on CKMR

This section addresses Subtask 3: “Collaborate with the BFT Technical Sub-group on CKMR, which will be coordinating the complete feasibility study that will integrate results and information on epigenetic ageing, genetic techniques, logistic considerations, costs, etc.”

In this respect, we simply note that this is being done on an ongoing basis, providing information and interacting with the sub-group whenever this has been required from us. We will continue to do so as needed.

5. Discussion and conclusions

Although already previously stated (document SCRS/2024/053), we re-state some main caveats here:

- The CVs calculated here assume that the EBFT population abundance is similar to the estimates from the latest BFT group stock assessment (in 2022). But if the true population abundance is considerably higher than assumed here, then the CVs will be higher than those calculated here (e.g. if the true population was four times higher than assumed, the CVs would double those calculated here). This is a very important and unavoidable source of uncertainty in CKMR design; therefore it is crucial to plan for flexibility in sampling over time.

- The CVs calculated here might look very good (i.e. low). But it should be realized that the population estimates from CKMR are not independent annual estimates (as may be obtained from a scientific survey), but behave more like the overall output of a stock assessment, where if the population abundance estimate is wrong in one year, it is also likely to be wrong by a similar amount the next year.
- Our results are based on an assumption that Atlantic samples are well-mixed (i.e. that they represent equally well all fish from the entire Med and not preferentially the fish from a particular spawning ground). As previously agreed, this is a reasonable assumption to start with, while trying to check at a later stage if this is a justifiable approximation; only if there is good evidence against it, would it be worth developing a more complex model. As explained in SCRS/2024/053, checking this assumption requires having enough POPs to be able to ascertain with statistical significance if there are different rates of POPs between different Atlantic fisheries and a particular juvenile source, i.e. the larvae from the Balearic survey (*Wlar*), or if two juvenile sources (*Wlar* and *CROjuv*) have different rates of POPs when compared against the combined Atlantic adult samples. The number of POPs observed from data until 2026 may not be sufficient to diagnose this potential issue, but it should be feasible with the sampling schemes going to 2030. Fortunately, weak departures from the well-mixed assumption of Atlantic samples are unlikely to trouble the total EBFT estimates much anyway; and there is a reasonable a priori basis for not expecting major departures, at least between the *W* and *C* subpopulations.
- Samples from the *Wad* and *Cad* fisheries would be helpful to estimate potential subpopulation structure, including possible faithfulness and heritability to main spawning grounds in the Med, and the abundance split between the subpopulations (if structure was detected). This basic biological information is likely to be of at least qualitative importance for management

To summarize, in this document (which extends and uses the work carried out in document SCRS/2024/053), we have investigated a number of alternative designs focusing on trying to provide a first estimate (which may be rough but hopefully useful) of total EBFT adult population size in time for the MSE review in 2027.

From the results of Designs 0-6, using samples until 2026, we may conclude that Design 2 is economical (in terms of total sample size) for a single "pinning-down" of EBFT abundance, with an expected CV of around 21% for the SSB in 2020 (increasing to 33% for the SSB in 2026). Design 5 results in appreciably lower CVs than Design 2, although at the cost of using larger sample sizes. Design 6 results in the lowest CVs of all designs explored, although they are only very slightly lower than those obtained from Design 5.

Ignoring logistic considerations (which the coordinators of the complete feasibility study would have to consider), and assuming that costs are a linear function of the number of samples analyzed, Designs 2 and 5 (and possibly 6) appear as good candidates at this point.

If Design 2 was selected at this point, we would still consider it a good idea to collect *CROjuv* samples, if "cheap" to do, because this is a source of juveniles which do not suffer from the extra-sibship issue that affects larvae samples. Furthermore, the inclusion of *CROjuv* fish helps for the verification of the well-mixed assumption for Atlantic fisheries. We note that sampling *CROjuv* fish now would not necessarily imply a commitment to genotype them at some point, but at least it would not preclude this option if later found desirable.

A similar comment applies to Balearic larvae collection, i.e. we consider it a good idea to keep collecting large numbers of these larvae (in ethanol), in case genotyping a larger number was found desirable later on.

Results for some possible designs to 2030 are also presented in this document, to illustrate the value of adding more years of data. Other alternatives may be explored at a later time depending on the experience gained from the first analyses of real CKMR data.

Further efficiency gains could be achieved in future by fine-tuning the plan as we go.

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References

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Table 1. Tables 3 and 4 from SCRS/2024/053, combined into a single table.

Number of samples per year					
	<i>Larval survey</i>	<i>Juvenile fishery</i>	<i>Adult fisheries</i>		
	Balearics: <i>Wlar</i>	Croatia: <i>CROjuv</i>	West Med: <i>Wad</i>	Central Med: <i>Cad</i>	Atlantic: <i>ATLad</i>
Age at sampling		2-4	3-28	3-28	3-28
2019-2024	3000 (exc. 2021)	0	0	0	0
2025-2030	8000	2000	2000	2000	2000

Table 2. Summary of larvae available from Balearic larval survey.

Balearic larval survey					
		<i>Larvae per survey (in formalin)</i>		<i>Larvae in ethanol</i>	
<i>Year</i>	<i>Number of survey stations</i>	<i>Total number</i>	<i>Maximum number in one station</i>	<i>Total number already separated</i>	<i>Notes about the larvae in ethanol</i>
2019	108	45133	31124	2822	All larvae preserved in ethanol are from a separate adaptive grid run in the 2019 survey. There may be more larvae available from that adaptive grid. No larvae are available from the systematic grid.
2020	91	26026	4182	2250	No more larvae are available (they have been used for other purposes).
2022	124	26100	7391	7632	Most of the larvae separated so far are from Bongo 1000. More larvae are available from Bongo 500.
2023	106	55452	13381	2901	More larvae are available

Table 3. Number of fish samples since 2020, with tissue currently available from USA and Canada.

<i>Number of samples</i>	<i>Total</i>		<i>BFT-E</i>	
	USA	CAN	USA (~61%)	CAN (~47%)
2020	920	414	563	196
2021	1798	1011	1101	478
2022	1556	1060	953	501

Table 4. Number of fish samples since 2020, with tissue currently available in the GBYP database for the entire East Atlantic, traps in the Gibraltar area (in the Atlantic) and Norway PS.

	<i>All fisheries in East Atlantic</i>		<i>Traps in Gibraltar (Atlantic)</i>		<i>Norway PS</i>	
	<i>Number of samples</i>	<i>Mean age</i>	<i>Number of samples</i>	<i>Mean age</i>	<i>Number of samples</i>	<i>Mean age</i>
2020	641	10.3	0	-	385	12.2
2021	415	10.6	41	11.6	168	11.7
2022	284	11.2	87	10.7	65	12.2
2023	298	11.2	0	-	115	12.8
2024	35	10.8	30	10.9	0	-

Table 5. Number of fish samples, since 2020, with tissue currently available in the GBYP database for the *Wad* PS and *Cad* PS fisheries.

	<i>PS fisheries in "BA-Balearic and Catalan Sea"</i>		<i>PS fisheries in "TY- Tyrrhenian Sea" or "SIS-South/Strait of Sicily" or "TU-Gulf of Gabes"</i>	
	<i>Number of samples</i>	<i>Mean age</i>	<i>Number of samples</i>	<i>Mean age</i>
2020	310	11	671	10.3
2021	333	11.5	627	9.6
2022	334	11.6	470	10.7
2023	2	9	4	12.2
2024	0	-	0	-

Table 6. Designs 0 to 6, to 2026: number of samples used per catch year (i.e. the year the fish are extracted from the sea). Historical samples (already collected or expected to be collected until year 2024) are in brackets. West Atlantic samples are from USA + Canada fisheries. For East Atlantic fisheries, historical samples are from traps in Gibraltar + Norway PS, and future ones are mainly from traps in Gibraltar and also, possibly, from Norway PS.

	<i>Number of samples collected, and genotyped, per catch year</i>								
	<i>Year</i>	<i>Total number of samples</i>	<i>Larvae</i>		<i>Juveniles</i>		<i>Adult fisheries</i>		
			<i>Balearic survey: Wlar</i>	<i>Croatia fishery: CROjuv ages 2-4</i>	<i>West Med: Wad ages 3-28</i>	<i>Central Med: Cad ages 3-28</i>	<i>Atlantic: ATLad ages 3-28</i>		
							<i>Total</i>	<i>East</i>	<i>West</i>
Design 0	2019	(3000)	(3000)	0	0	0	0		
	2020	(3000)	(3000)	0	0	0	0		
	2021	(0)	(0)	0	0	0	0		
	2022	(3000)	(3000)	0	0	0	0		
	2023	(3000)	(3000)	0	0	0	0		
	2024	(3000)	(3000)	0	0	0	0		
	2025	16000	8000	2000	2000	2000	2000	500	1000+500
	2026	16000	8000	2000	2000	2000	2000	500	1000+500
TOTAL	47000	31000	4000	4000	4000	4000			
Design 1	2019	(2822)	(2822)	0	(0)	(0)	(0)	(0)	(0)
	2020	(4374)	(2250)	0	(310)	(670)	(1144)	(0+385)	(563+196)
	2021	(2748)	(0)	0	(333)	(627)	(1788)	(41+168)	(1101+478)
	2022	(10410)	(8000)	0	(334)	(470)	(1606)	(87+65)	(953+501)
	2023	(9913)	(8000)	0	(2)	(4)	(1907)	(0+115)	(1225+567)
	2024	(9822)	(8000)	0	(0)	(0)	(1822)	(30+0)	(1225+567)
	2025	11792	8000	0	0	0	3792	2000	1225+567
	2026	11792	8000	0	0	0	3792	2000	1225+567
TOTAL	63673	45072	0	979	1771	15851			
Design 2	2019	(2822)	(2822)	0	(0)	(0)	(0)	(0)	(0)
	2020	(4374)	(2250)	0	(310)	(670)	(1144)	(0+385)	(563+196)
	2021	(2748)	(0)	0	(333)	(627)	(1788)	(41+168)	(1101+478)
	2022	(6410)	(4000)	0	(334)	(470)	(1606)	(87+65)	(953+501)
	2023	(5913)	(4000)	0	(2)	(4)	(1907)	(0+115)	(1225+567)
	2024	(5822)	(4000)	0	(0)	(0)	(1822)	(30+0)	(1225+567)
	2025	7792	4000	0	0	0	3792	2000	1225+567
	2026	7792	4000	0	0	0	3792	2000	1225+567
TOTAL	43673	25072	0	979	1771	15851			
Design 3	2019	(2822)	(2822)	0	0	0	(0)	(0)	(0)
	2020	(3394)	(2250)	0	0	0	(1144)	(0+385)	(563+196)
	2021	(1788)	(0)	0	0	0	(1788)	(41+168)	(1101+478)
	2022	(5606)	(4000)	0	0	0	(1606)	(87+65)	(953+501)
	2023	(5907)	(4000)	0	0	0	(1907)	(0+115)	(1225+567)
	2024	(5822)	(4000)	0	0	0	(1822)	(30+0)	(1225+567)
	2025	7792	4000	0	0	0	3792	2000	1225+567
	2026	7792	4000	0	0	0	3792	2000	1225+567
TOTAL	40923	25072	0	0	0	15851			

Table 6. Continued.

Design 4	2019	(2822)	(2822)	0	(0)	(0)	(0)	(0)	(0)
	2020	(4374)	(2250)	0	(310)	(670)	(1144)	(0+385)	(563+196)
	2021	(2748)	(0)	0	(333)	(627)	(1788)	(41+168)	(1101+478)
	2022	(8410)	(6000)	0	(334)	(470)	(1606)	(87+65)	(953+501)
	2023	(7913)	(6000)	0	(2)	(4)	(1907)	(0+115)	(1225+567)
	2024	(7822)	(6000)	0	(0)	(0)	(1822)	(30+0)	(1225+567)
	2025	8792	6000	0	0	0	2792	1000	1225+567
	2026	8792	6000	0	0	0	2792	1000	1225+567
TOTAL	51673	35072	0	979	1771	13851			
Design 5	2019	(2822)	(2822)	0	(0)	(0)	(0)	(0)	(0)
	2020	(4374)	(2250)	0	(310)	(670)	(1144)	(0+385)	(563+196)
	2021	(2748)	(0)	0	(333)	(627)	(1788)	(41+168)	(1101+478)
	2022	(8410)	(6000)	0	(334)	(470)	(1606)	(87+65)	(953+501)
	2023	(7913)	(6000)	0	(2)	(4)	(1907)	(0+115)	(1225+567)
	2024	(7822)+2000	(6000)	2000	(0)	(0)	(1822)	(30+0)	(1225+567)
	2025	10792	6000	2000	0	0	2792	1000	1225+567
	2026	8792	6000	0	0	0	2792	1000	1225+567
TOTAL	55673	35072	4000	979	1771	13851			
Design 6	2019	(2822)	(2822)	0	(0)	(0)	(0)	(0)	(0)
	2020	(4374)	(2250)	0	(310)	(670)	(1144)	(0+385)	(563+196)
	2021	(2748)	(0)	0	(333)	(627)	(1788)	(41+168)	(1101+478)
	2022	(8410)	(6000)	0	(334)	(470)	(1606)	(87+65)	(953+501)
	2023	(7913)	(6000)	0	(2)	(4)	(1907)	(0+115)	(1225+567)
	2024	(7822)+2000	(6000)	2000	(0)	(0)	(1822)	(30+0)	(1225+567)
	2025	14792	6000	2000	2000	2000	2792	1000	1225+567
	2026	12792	6000	0	2000	2000	2792	1000	1225+567
TOTAL	63673	35072	4000	4979	5771	13851			

Table 7. Designs 0 to 6, to 2026: Expected number of “meaningful” kin pairs.

	Expected number of POPs							Expected number of HSPs (MHSPs & FHSPs)		
	Total POPs	Potential Parent	Potential Offspring					Wlar Wlar	CROjuv Wlar	CROjuv CROjuv
			Wlar	CROjuv	Wad	Cad	ATLad			
Design 0	91	Wad	45	5	1	1	1	104	15	3
		Cad	3	11	0	1	1			
		ATLad	14	7	1	1	1			
Design 1	106	Wad	4	0	0	0	3	237	0	0
		Cad	0	0	0	0	3			
		ATLad	73	0	0	0	21			
Design 2	75	Wad	4	0	0	0	3	71	0	0
		Cad	0	0	0	0	3			
		ATLad	43	0	0	0	21			
Design 3	64	Wad	0	0	0	0	0	71	0	0
		Cad	0	0	0	0	0			
		ATLad	43	0	0	0	21			
Design 4	75	Wad	4	0	0	0	3	142	0	0
		Cad	0	0	0	0	3			
		ATLad	46	0	0	0	18			
Design 5	91	Wad	4	0	0	0	3	142	15	3
		Cad	0	0	0	0	3			
		ATLad	46	15	0	0	18			
Design 6	194	Wad	69	5	2	1	5	142	15	3
		Cad	4	9	1	3	4			
		ATLad	46	15	4	8	18			

Table 8. Designs 0 to 6, to 2026: overall sampling and genotyping effort, expected number of POPs, and expected CV (percentage) of total EBFT SSB estimates for several years.

Design	Total number of samples collected and genotyped from 2019-2026						Expected number of POPs		CV (%) of total EBFT SSB estimates				
	Total	Wlar	CROjuv	Wad	Cad	ATLad			2014	2017	2020	2023	2026
0	47000	31000	4000	4000	4000	4000	91	84	54	36	31	34	
1	63673	45072	0	979	1771	15851	106	31	19	18	21	26	
2	43673	25072	0	979	1771	15851	75	32	19	21	26	33	
3	40923	25072	0	0	0	15851	64	51	32	32	31	40	
4	51673	35072	0	979	1771	13851	75	33	20	21	25	31	
5	55673	35072	4000	979	1771	13851	91	32	19	17	20	26	
6	63673	35072	4000	4979	5771	13851	194	27	17	16	19	24	

Table 9. Design 5 extended to 2030 in two alternative ways (V1 and V2): number of samples used per catch year (i.e. the year the fish were extracted from the sea).

	Number of samples collected, and genotyped, per catch year									
	Year	Total number of samples	Larvae	Juveniles	Adult fisheries					
			Balearic survey: Wlar	Croatia fishery: CROjuv ages 2-4	West Med: Wad ages 3-28	Central Med: Cad ages 3-28	Atlantic: ATLad ages 3-28			
							Total	East	West	
Design 5 To 2030 V1	2019	(2822)	(2822)	0	(0)	(0)	(0)	(0)	(0)	(0)
	2020	(4374)	(2250)	0	(310)	(670)	(1144)	(0+385)	(563+196)	
	2021	(2748)	(0)	0	(333)	(627)	(1788)	(41+168)	(1101+478)	
	2022	(8410)	(6000)	0	(334)	(470)	(1606)	(87+65)	(953+501)	
	2023	(7913)	(6000)	0	(2)	(4)	(1907)	(0+115)	(1225+567)	
	2024	(7822)+2000	(6000)	2000	(0)	(0)	(1822)	(30+0)	(1225+567)	
	2025	10792	6000	2000	0	0	2792	1000	1225+567	
	2026	10792	6000	2000	0	0	2792	1000	1225+567	
	2027	10792	6000	2000	0	0	2792	1000	1225+567	
	2028	10792	6000	2000	0	0	2792	1000	1225+567	
	2029	10792	6000	2000	0	0	2792	1000	1225+567	
	2030	8792	6000	0	0	0	2792	1000	1225+567	
	TOTAL	98841	59072	12000	979	1771	25019			
Design 5 To 2030 V2	2019	(2822)	(2822)	0	(0)	(0)	(0)	(0)	(0)	
	2020	(4374)	(2250)	0	(310)	(670)	(1144)	(0+385)	(563+196)	
	2021	(2748)	(0)	0	(333)	(627)	(1788)	(41+168)	(1101+478)	
	2022	(8410)	(6000)	0	(334)	(470)	(1606)	(87+65)	(953+501)	
	2023	(7913)	(6000)	0	(2)	(4)	(1907)	(0+115)	(1225+567)	
	2024	(7822)+2000	(6000)	2000	(0)	(0)	(1822)	(30+0)	(1225+567)	
	2025	10792	6000	2000	0	0	2792	1000	1225+567	
	2026	10792	6000	2000	0	0	2792	1000	1225+567	
	2027	14792	6000	2000	2000	2000	2792	1000	1225+567	
	2028	14792	6000	2000	2000	2000	2792	1000	1225+567	
	2029	14792	6000	2000	2000	2000	2792	1000	1225+567	
	2030	12792	6000	0	2000	2000	2792	1000	1225+567	
	TOTAL	114841	59072	12000	8979	9771	25019			

Table 10. Design 5 extended to 2030 in two alternative ways (V1 and V2): Expected number of “meaningful” kin pairs.

	Expected number of POPs							Expected number of HSPs (MHSPs & FHSPs)		
	Total POPs	Potential Parent	Potential Offspring					Wlar	CROjuv	CROjuv
			Wlar	CROjuv	Wad	Cad	ATLad			
Design 5 To 2030 V1	255	Wad	4	0	0	0	4	393	81	34
		Cad	0	0	0	0	5			
		ATLad	124	66	0	0	50			
Design 5 To 2030 V2	553	Wad	168	21	5	3	11	393	81	34
		Cad	12	36	1	7	9			
		ATLad	124	66	14	25	50			

Table 11. Design 5, to 2026, and two possible extensions to 2030 (V1 and V2): Expected CV (percentage) of total EBFT SSB estimates for several years.

<i>CV (%) of total EBFT SSB estimates for Design 5, To 2026 and to 2030 (V1 and V2)</i>						
	<i>2014</i>	<i>2017</i>	<i>2020</i>	<i>2023</i>	<i>2026</i>	<i>2029</i>
To 2026	32	19	17	20	26	-
To 2030, V1	26	15	10	10	12	13
To 2030, V2	24	13	9	9	11	12

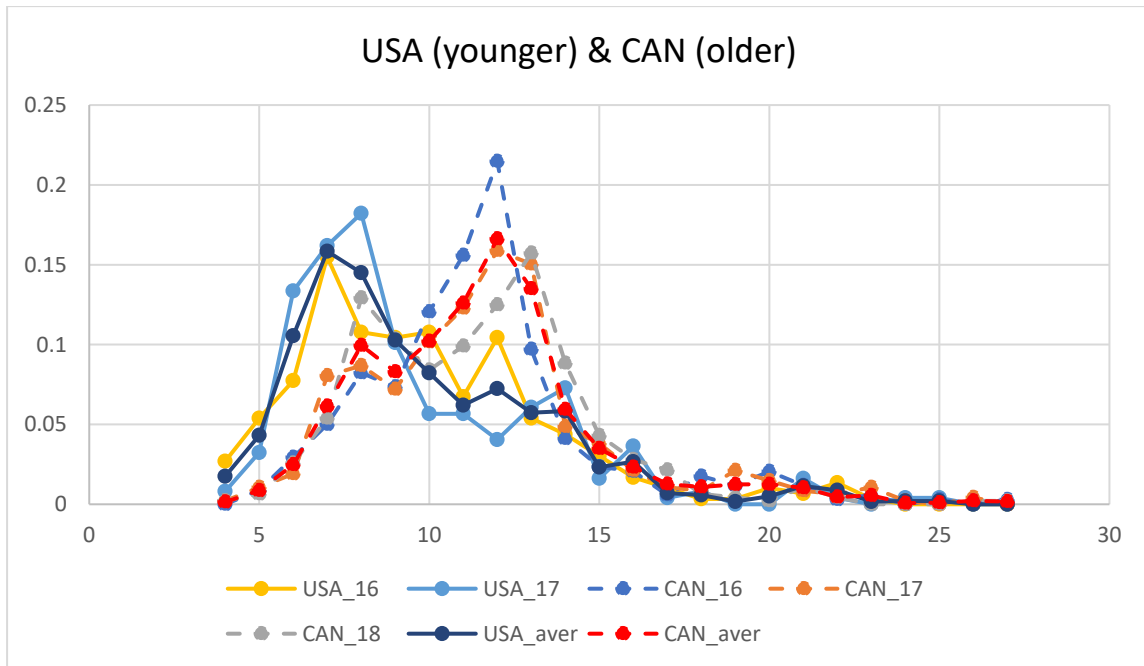


Figure 1. Age composition of samples available from USA (years 2016-2017 and average) and Canada (years 2016-2018 and average).

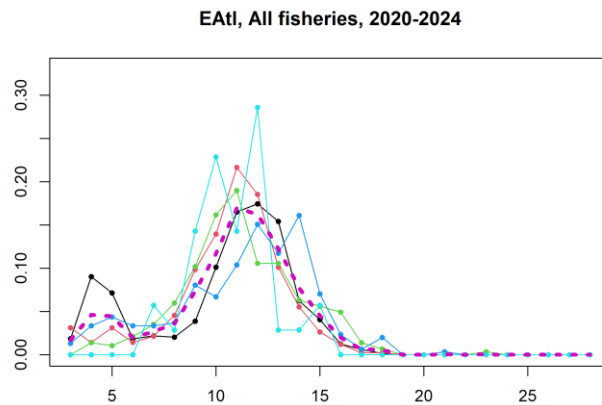


Figure 2. Age composition of the samples available in the GBYP database for the entire East Atlantic. The continuous lines represent different years (2020-2024) and the dashed line the average over the years.

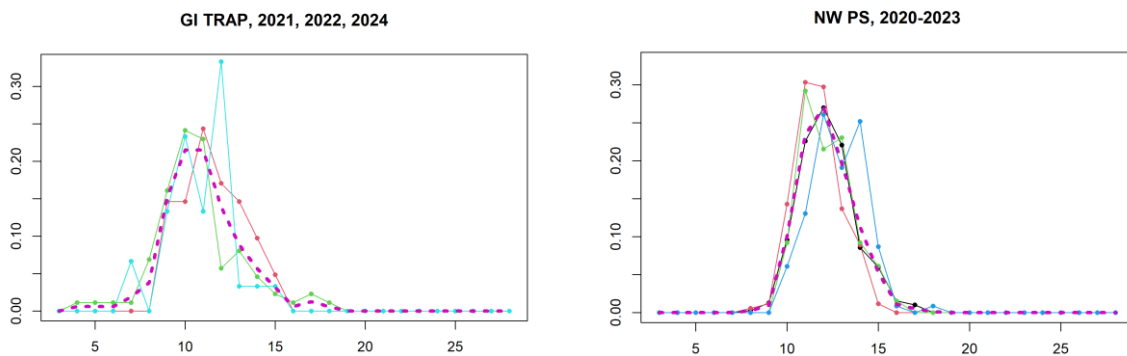


Figure 3. Age composition of samples available in the GBYP database for Traps in Gibraltar (Atlantic; left panel) and Norway PS (right panel) fisheries. The continuous lines represent different years (2021, 2022, 2024 for Traps in Gibraltar; 2020-2023 for Norway PS) and the dashed line the average over the years.

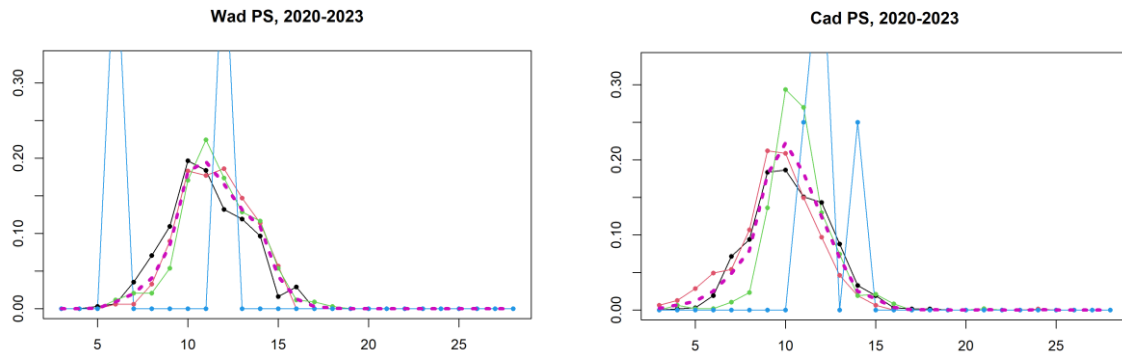


Figure 4. Age composition of samples available in the GBYP database for the *Wad* PS (left panel) and *Cad* PS (right panel) fisheries. The continuous lines represent different years (2020-2023) and the dashed line the average over the years.

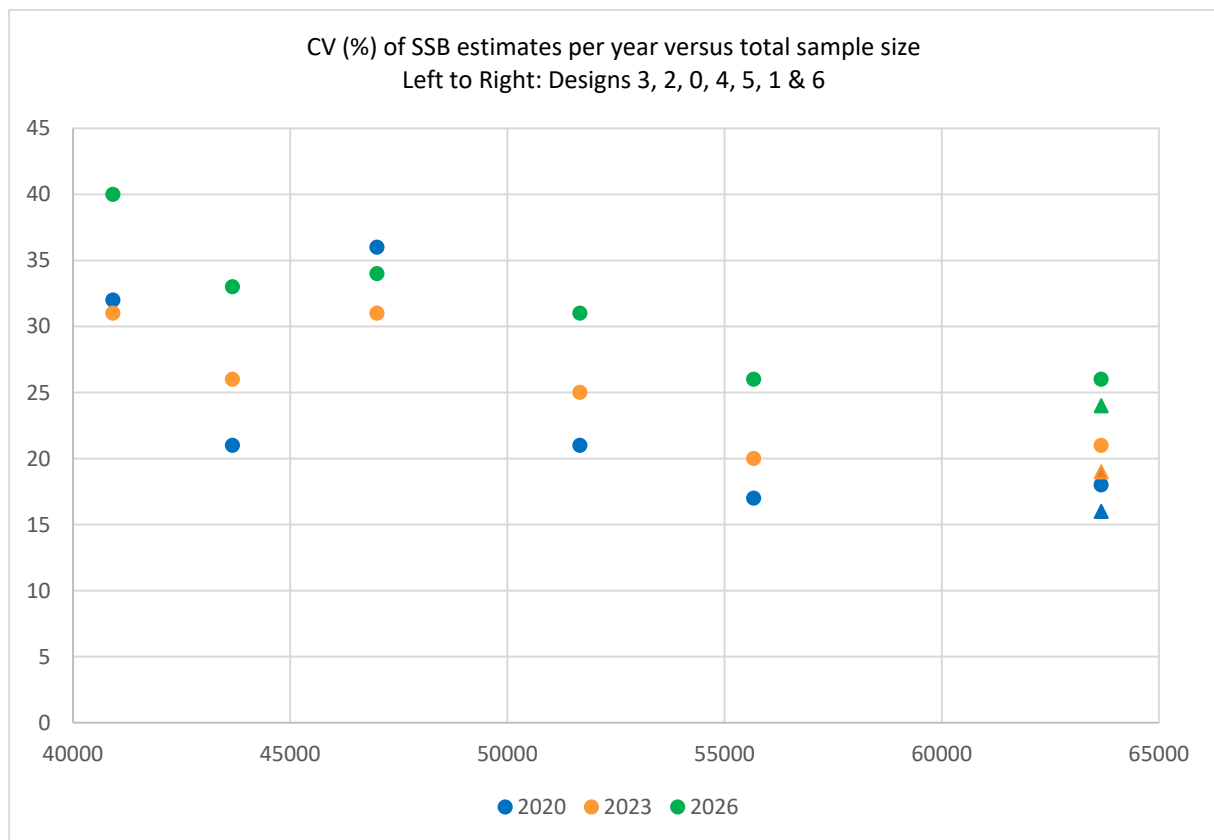


Figure 5. Results from Designs 0-6. For each design, the expected CV of the total EBFT SSB estimates in recent years (2020, 2023 and 2026) is plotted versus the total sample size used in the design. From left to right, the designs are: Design 3, 2, 0, 4, 5, 1 and 6. Designs 1 and 6 have the same total sample size. To differentiate the results from Designs 1 and 6 in the graph, they are plotted with circles and triangles, respectively.

More detailed results for Designs 0 to 6

Table A.1. Designs 0 to 6, to 2026: Expected percent CV of total EBFT abundance estimates. Panels (a), (b) and (c) refer to Spawning Stock Biomass (SSB), Biomass of ages 5 and older, and Total Reproductive Output (TRO, expressed as equivalent number of 10-year old fish), respectively.

(a) CV (%) of total EBFT SSB estimates by year													
	2014	2015	2016	2017	2018	2019	2020	2021	2022	2023	2024	2025	2026
Design 0	84	73	63	54	46	40	36	33	31	31	31	33	34
Design 1	31	26	22	19	17	17	18	19	20	21	23	24	26
Design 2	32	26	22	19	19	19	21	23	24	26	28	30	33
Design 3	51	41	35	32	32	32	32	31	31	31	32	35	40
Design 4	33	27	23	20	19	19	21	22	24	25	27	29	31
Design 5	32	27	22	19	17	16	17	18	19	20	22	24	26
Design 6	27	23	19	17	16	15	16	17	18	19	20	22	24

(b) CV (%) of total EBFT Biomass(5+) estimates by year													
	2014	2015	2016	2017	2018	2019	2020	2021	2022	2023	2024	2025	2026
Design 0	87	76	66	56	47	41	36	33	32	31	32	33	35
Design 1	33	28	23	20	18	17	18	19	20	21	23	24	26
Design 2	34	28	24	20	19	19	21	22	24	26	28	31	34
Design 3	57	45	38	33	32	32	32	31	31	31	33	36	41
Design 4	35	29	25	21	19	19	20	22	24	25	27	29	31
Design 5	34	29	24	20	17	17	17	18	19	21	22	24	27
Design 6	29	24	21	18	16	16	16	17	18	19	20	22	24

(c) CV (%) of total EBFT TRO (as equivalent number of 10-year old fish) estimates by year													
	2014	2015	2016	2017	2018	2019	2020	2021	2022	2023	2024	2025	2026
Design 0	88	78	68	59	51	45	40	36	34	33	32	33	35
Design 1	33	28	24	21	20	19	20	20	21	22	23	24	26
Design 2	34	28	24	22	21	21	22	24	25	26	28	30	33
Design 3	54	44	37	34	33	33	33	32	31	31	32	35	40
Design 4	35	30	25	23	21	21	22	23	24	25	27	29	31
Design 5	34	29	24	21	19	18	18	19	20	21	22	24	26
Design 6	29	24	21	19	17	17	17	18	18	19	20	22	24

Table A.2. Design 5, to 2026, and two possible extensions of it to 2030 (V1 and V2): Expected CV of total EBFT abundance estimates. (a) Spawning Stock Biomass (SSB); (b) Biomass of ages 5 and older; (c) Total Reproductive Output (TRO, expressed as equivalent number of 10-year old fish).

CV (percentage) of total EBFT estimates, Design 5, to 2026 and to 2030 (V1 and V2)									
	(a) SSB			(b) Biomass(5+)			(c) TRO (equivalent no. 10-year old fish)		
	To	To 2030		To	To 2030		To	To 2030	
	2026	V1	V2	2026	V1	V2	2026	V1	V2
2014	32	26	24	34	28	25	34	27	24
2015	27	22	20	29	24	21	29	23	20
2016	22	18	16	24	20	17	24	19	17
2017	19	15	13	20	16	14	21	16	14
2018	17	12	11	17	13	11	19	14	12
2019	16	11	10	17	11	10	18	12	10
2020	17	10	9	17	10	9	18	11	10
2021	18	10	9	18	10	9	19	11	10
2022	19	10	9	19	10	9	20	11	10
2023	20	10	9	21	10	9	21	11	10
2024	22	11	10	22	11	10	22	11	10
2025	24	11	10	24	11	10	24	11	10
2026	26	12	11	27	12	11	26	12	11
2027		12	11		12	11		12	11
2028		13	12		13	12		13	12
2029		13	12		14	12		13	12
2030		14	13		14	13		14	13