Annex 14

SEMINAR: CLOSE-KIN MARK RECAPTURE AS A TOOL FOR ESTIMATING SPAWNING BIOMASS IN PACIFIC BLUEFIN TUNA

International Scientific Committee for Tuna and Tuna-like Species in the North Pacific Ocean

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Operationalizing the estimation of absolute spawning stock abundance for southern Bluefin tuna using close-kin genetics

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A summary of the general details, and the specifics of the southern bluefin tuna (SBT) example relating to operationalizing the close-kin mark-recapture (CKMR) approach in a fisheries context were presented to the ISC. Using the simple "cartoon" paradigm, the basic ideas relating to the abundance estimator, the associated precision, and how this relates to the sample sizes of both juveniles and adults were outlined. For the SBT application, the sampling design was explained given the spawning dynamics; then the number of parents and offspring pairs (POPs) found and how the stand-alone estimate was constructed; and, finally, how these data were incorporated into the SBT stock assessment and what were the implications in relation to updated stock status.

Discussion: Clarification was made that fecundity at length for adults was estimated from a stand-alone model rather from assumptions made in the model. Questions were raised regarding the frequency of the testing to get reasonable estimates and cost to obtain the samples. The importance of effect of maturity ogives in the estimates of POPs was pointed out. It was noted that the sampling scheme for spawners of Pacific bluefin tuna would have to consider all spawning grounds to obtain random samples. In terms of dedicated personnel, experience from the CKMR projects for SBT suggests the need for two to three geneticists, two to three modelers, and many samplers.

Progress in a Japanese Close-kin project in 2014-2015

Dr. Nobuaki Suzuki
National Research Institute of Far Seas Fisheries, Fisheries Research Agency, Japan

In 2014 Japan initiated preliminary attempts to implement the Close-kin genetics to estimate the absolute value of PBF SSB (spawning stock biomass). In this seminar, progress during this past year in the project was described. First, project members and institutes involved were identified and their tasks were

explained. Then sampling schemes and future plans around Japanese waters were shown geographically, and a simple procedure for a sampling operation using disposable biopsy punch and specific DNA stabilizing solutions were presented. RAD (restriction site-associated DNA) sequencing attempts were successful in sequencing a suitable quantity of the PBF genome and then ddRAD (double-digest restriction-site associated DNA sequencing) enabled to screen SNPs adequate for the Close-kin POP findings. Given the level of allelic variability for SNP- (single-nucleotide polymorphism) based on the results of ddRAD sequencing, computational simulations were performed to validate how many SNP markers should be analyzed through a virtual ideal reproductive process. According to both estimates from identifying or misidentifying parentages, 100 SNP markers were identified as sufficient to find POPs accurately. The project progressed more quickly than expected in 2014-2015, and continues to achieve our goal of getting robust estimate of PBF SSB.

Discussion: The importance of sampling design for adults used in close-kin analysis was discussed. The authors noted that designing the sampling program will be difficult but their design includes fish from the Sea of Japan (30-50kg) as well as the larger fish found in waters off Taiwan. A participant noted that the ISC is an ideal organization to develop and facilitate sample collection. In response to a question, the authors noted that cost for this project have been roughly 100,000 USD with approximately 50 USD per individual for the genetic processing. It was noted that the costs of developing the appropriate markers are a one-time cost and may be useful to other bluefin tuna stocks. Participants discussed the difference between this study and the CSIRO study on southern bluefin tuna. The authors clarified that they use SNP markers rather than microsatellite markers. It was noted that SNP markers are more consistent than microsatellite markers between laboratories, which facilitates collaboration.

Close-kin Mark Recapture as a Tool for Estimation of Spawning Biomass in Pacific Bluefin Tuna (PBF): Outcomes from a Workshop on Developing CKMR techniques for Pacific Bluefin Tuna

Dr. Russ Vetter
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A review of the results of a workshop held at the Southwest Fisheries Science Center (La Jolla, USA) on May 27-29, 2015 was presented. The workshop accomplished three goals: 1. It evaluated the theory and promise of Close Kin Mark Recapture (CKMR) population estimation; 2. It reviewed the known and unknown aspects of PBF life history that could influence sampling design; and, 3. It identified a sampling design and sampling program that would build on: currently monitored fisheries; existing fisheries sampling programs; current modeling approaches and the existing management structure of national and international fisheries organizations. The seminar detailed the resulting sampling design and a sampling plan for PBF-CKMR research designed to produce a preliminary estimate in three years and a more precise estimate in five years of SSB. The overall plan has three components: biological sampling; high throughput genetic screening; and population modeling. This seminar focused on the biological sampling required based on expectations of stock size derived from the 2014 PBF stock assessment and known aspects of PBF life history. The contents of the report (posted on the ISC website), and the proposed next steps should be considered as a possible way forward.

Discussion: Initial discussions focused on the total number of samples needed and the corresponding years over which the samples would be collected. Approximately 7000-8000 samples would be collected on an annual basis for a total of five years. A first estimate of SSB would be available after 3 years. Although the project duration is currently five years, there was general agreement that the project was valuable and should be continued beyond five years so as to provide a time-series of information rather than a three-year window of information in time. Collecting samples on a yearly basis was preferred to sampling intermittently. Given Japan's current progress in collecting samples (approximately 1000 have been collected) the CKMR program proposed samples sizes should be coordinated with the sampling from the Japanese program.

The number of samples could be expected to decline over time if SSB estimation remained the sole objective. The reason for this was that juvenile samples would continue to be useful provided those juveniles survive to adults and spawn. However, it was noted that the project allows for adjustments in sample size depending on objectives and funding. Sample size could remain at 7000-8000 per year if objectives in addition to SSB estimation (e.g. half-sibling analysis) are included. In contrast, sample size could be reduced if funding is limited. An example of collecting from only one of the multiple juvenile pools was provided as a means to reduce cost, but it was noted that this would reduce the capability to determine stock structure.

The role of ISC in the CKMR program was discussed at length. The ISC would be an obvious avenue from which this program is implemented, particularly via the PBFWG. Meeting to discuss progress and planning within the scope of the ISC makes sense. The organization chart is important for how the project moves forward. It was suggested that the ISC Chair develop a road map on working with other research organizations with complementary or similar research.

Other possible collaborations were discussed. The European Union has provided Euro 100,000 to WCPFC for a tissue bank, and therefore could be a source of storage. Alternatively, governmental agencies can serve valuable roles for tissue repository. Lastly, the involvement of stock assessment scientists in the early phases of this project was emphasized to ensure project results have high utility for stock assessments.

Discussions on Next Steps for CKMR-PBF: Sampling, Project Management and Funding

During the La Jolla workshop, a preliminary organizational chart for the CKMR-PBF proposal was developed, and the ISC Chair thought that this was a good framework from which to work from. The CKMR work was divided into 3 parts: 1) Sampling, 2) Genetics and 3) Modeling.

Based on the previous discussions, it was agreed that the ISC should take a leading role in CKMR-PBF, especially in providing the samples for the work. The Chair proposed to the Plenary that the Chair form a small advisory group on CKMR, which is allowed by the ISC operations manual, in the next 2 to 3 months to develop a sampling program with the appropriate sample sizes and the sampling protocols. The Chair also proposed to inform the NC in 2015 about the developments on CKMR.

The Plenary unanimously supported the formation of the CKMR advisory group and agreed that the Chair should report on CKMR to the NC. The Plenary suggested that much of the CKMR work should go through the PBFWG because much of the expertise is already in the PBFWG, and it is important to include modelers throughout the project. The Chair agreed and will ensure that the expertise in the PBFWG will be fully utilized. The Plenary noted that given there is no funding from the ISC to perform

the CKMR work, it maybe be easier for individual countries to collect samples and perform the genetic analysis, before sharing the data with the ISC. The Chair agreed that this may be how CKMR proceeds but emphasized that the data must be fully shared with the other countries. Adequate storage facilities will need to be developed and subsamples need to be shared with other countries to perform cross-validation and quality control. The Plenary noted that in addition to reporting to the NC, the Chair should also inform the IATTC about the CKMR proposal. The Chair agreed with this and noted that IATTC staff participated in the CKMR workshop in La Jolla.

The Plenary recommended that mechanisms for moving forward on the genetics and modeling portions of CKMR be developed at the ISC through future discussions. It was recognized that there is substantial expertise on CKMR genetics methods within the ISC, and noted that Japan has already begun developing markers for the work. However, it is important that genetic markers used be fully shared. If possible, the sampling of adults and juveniles should begin in the Spring (April-May) of 2016.

The Chair concluded by thanking the Plenary for their support. The Chair will move forward in the next several months to form the small advisory group on CKMR; and also inform the NC and IATTC on CKMR.

Appendix 1

Seminar Agenda

9:30-10:00 *Operationalising the estimation of absolute spawning stock abundance for southern bluefin tuna using close-kin genetics*

Rich Hillary, Commonwealth Scientific and Industrial Research Organization, Australia.

Break: 10:30-11:00

11:00-11:45 *Progress in a Japanese Close-kin project in 2014-2015*

Nobuaki Suzuki, National Research Institute of Far Seas Fisheries, Japan

11:45-12:30 *Close-Kin Mark Recapture as a Tool for Estimation of Spawning Biomass in Pacific Bluefin Tuna: Outcomes from a Workshop on Developing CKMR techniques for Pacific Bluefin Tuna.*

Russ Vetter, Southwest Fisheries Science Center, NOAA Fisheries, USA

Lunch: 12:30-1:30

1:30-3:00 Discussion of Next Steps for CKMR-PBF: Sampling, Project Management, Funding