SHORT-TERM CONTRACT FOR BFT GROWTH IN FARMS STUDY (ICCAT-GBYP 09/2019-a) OF THE ATLANTIC-WIDE RESEARCH PROGRAMME FOR BLUEFIN TUNA (ICCAT GBYP Phase 9)

BETWEEN

THE INTERNATIONAL COMMISSION FOR THE CONSERVATION OF ATLANTIC TUNAS (ICCAT)

AND

TUNIPEX, S.A. EMPRESA DE PESCA DE TUNÍDEOS (PORTUGAL)

DELIVERABLE 5:

Final Report

Prepared by:

Pedro G. Lino, Rubén Muñoz-Lechuga, Maria Nunes, Alfredo Poço, Inga Barata, Morikawa Hirofumi & Rui Coelho

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FINAL REPORT OF ACTIVITIES

1) Executive summary

ICCAT requested an update on the potential growth rates of bluefin tuna in farming/fattening facilities, with the aim of improving coherence within the growth rates derived from eBCD, as stipulated in paragraph 28 of Rec. 18-02.

Given the particular situation of the Portuguese tuna traps located along the South coast capturing the adult fraction of the bluefin tuna exiting the Mediterranean after the reproduction season, a contract was established between ICCAT and Tunipex (with IPMA as scientific sub-contractor) to fulfill the required work in Portuguese traps in the eastern Atlantic Ocean.

The Tunipex tuna trap, where the tagging operations took place, is located about two and a half nautical miles from the coast of the Algarve, between about 20-60m depth. The central location of the trap is at: Lat= 37.01332 (North); Long= -7.71035 (West).

Between 27th of June and 21st of August 2019, eighty-nine (89) adult bluefin tunas were individually weighted, measured, double tagged and returned to the cage for fattening. Deliberate harvest of tagged fish started one month after the tagging date, with all fish being weighted and measured and, whenever possible, biological samples were collected from the tagged fish.

Data regarding initial and final weight and length, feeding amounts, stereoscopical measurements and environmental parameters were collected and reported.

The overall weight increase for the harvested fish had a mean of 27.4% (varying between 0% and 54.8%), for fish that were fattened between 41 and 129 days between tagging and harvesting. It is noted that the condition factor of the fish when tagged was very low, meaning the possibility that their potential growth in weight was high in a relatively short period of time.

The growth data collected in this study and reported here was collected successfully according to the ICCAT contract. At the end of the report, we also provide some additional recommendations, specifically in terms of onboard tagging operations, that may be considered for adjusting the tagging strategy on future phases of the ICCAT/GBYP tagging project.

2) Background

During the 21st Special Meeting of the Commission, the SCRS was asked to provide an update on the potential growth rates of bluefin tuna in farming/fattening facilities, with the aim of improving coherence within the growth rates derived from eBCD, as stipulated in paragraph 28 of Rec. 18-02. Consequently, GBYP was committed to carry out a broad

study on this topic, involving ad hoc experiments in selected farms along the eastern Atlantic and Mediterranean. Such broad study has been planned within Phase 8, and included several preparatory tasks, such as elaboration and distribution of a detailed questionnaire submitted to all the operative BFT farms, and meetings with farm owners, local authorities and scientists in the five areas where the study will be developed. The implementation of the study has started in Phase 9, which involved tagging experiments to determine individual growth trajectories, intensive monitoring of representative cages, including the record of relevant environmental variables and food provided to caged fishes and seasonal measurements of their growth by means of stereo-cameras measurements, as well as the elaboration and analysis of a database including data on initial length distributions from stereo-cameras and data on final sizes and weight at the end of farming period obtained during harvesting operations.

In this sense, and given the particular situation of the Portuguese tuna traps located along the South coast capturing the adult fraction of the bluefin tuna exiting the Mediterranean after the reproduction season, a contract was established between ICCAT and Tunipex (with IPMA as scientific sub-contractor) to fulfill the required work in Portuguese traps in the eastern Atlantic Ocean.

3) Objectives

The objective of this document is to provide Deliverables 2, 3 and 4 (Update of Work and Draft Final Report) of the Project ICCAT/GBYP - Phase 9 - Short-term contract for BFT growth in farms study (ICCAT/GBYP 09/2019-a) of the Atlantic-wide Research Programme for Bluefin tuna. The details included in this Report, as requested in the signed contract, include a Scientific report (this report) containing:

a) a full description of the initial conditions of the monitored cages (origin of the caged fishes, date and area of capture, characteristics of the monitored cage, initial number and biomass of caged fishes, etc.);

b) a detailed description of all the methodologies and protocols applied for monitoring environmental variables, biological sampling of dead fishes and taking measurements of live fishes - both directly and through stereoscopic cameras-, as well as tagging operations (how protocols have been applied, any departure from the protocol, difficulties encountered, etc.);

c) files containing videos and raw data from stereoscopic camera measurements of tagged fishes carried out after the first official stereoscopic camera measurements at caging of the whole catch;

d) detailed tables and graphs including:

- length and weight of any fish dead in the monitored cages due to causes other than harvesting operations, as well relevant data on biological samples from these fishes, if any,
- weekly records of environmental parameters (T, S, DO2) in the monitored cages,
- daily quantities and types of feed given to the trial cage/s,
- length and weight of tagged fishes at tagging and at harvesting, as well as information of deployed tags and biological samples taken from these fishes,
- length and weight of each fish harvested from the monitored cages (specifying date of harvesting),

e) an Executive Summary of the final report.

4) Full description of the work carried out

4.1) Methodology

Tagging for the Growth study in the Tuna trap owned by company Tunipex and located off the South coast of the Algarve province (Portugal) started on the 27th of June and was completed on the 21st of August, with 80 fish tagged as contracted.

In order to compensate for a higher than expected mortality, on the last day of tagging an additional 9 fish were tagged, therefore making a total of 89 fish tagged for this contract.

All fish were migrating in an East to West route, exiting the Mediterranean after the spawning season.

Fish were held in a temporary cage (PP11) before being tagged and transferred to the monitoring and fattening cage (PP18, officially named by ICCAT as Farm Cage PRT903). Both cages are of the same dimensions: LxWxH 120m x 55m x 33m (H is limited by the sea floor, so it is the same as water depth).

Fish to be tagged were isolated on a knot-less net to minimize damage to the fish before hauling onboard on a stretcher (Figure 1).

Figure 1- Fish being individually captured in a knot-less net before hauling on board on a stretcher. Mr. Alfredo Poço (center with white t-shirt), the coordinator for the tagging activities, is supervising the net setup.

Fish were individually hauled on the stretcher with a digital scale and weighted (Figure 2). The stretchers used were individually identified (to subtract the weight of the stretcher to the measured weight) and had a mesh along the middle of the whole stretcher length to guarantee that all water was purged before weighting. Furthermore, fish weight was registered immediately before hauling back into the water (not at hauling on board) to ensure that water was purged to the maximum extent possible.

Figure 2- Digital scale used to individually weight the tagged fish.

All tagged fish were individually weighted, measured with a tape (SFL - Straight Fork Length) and double tagged with conventional tags provided by ICCAT (Figure 3 and Annex 1). All tagged fish were individually monitored with a stereoscopic-camera system (Annex 2) immediately after tagging, noting that it was only possible to obtain length measurements from 69 fish. Measurements and weights were double checked using the GOPRO video recordings of the tagging events. As such, any possible data errors during the tagging events can be attributed to human error due to the movement of the fish.

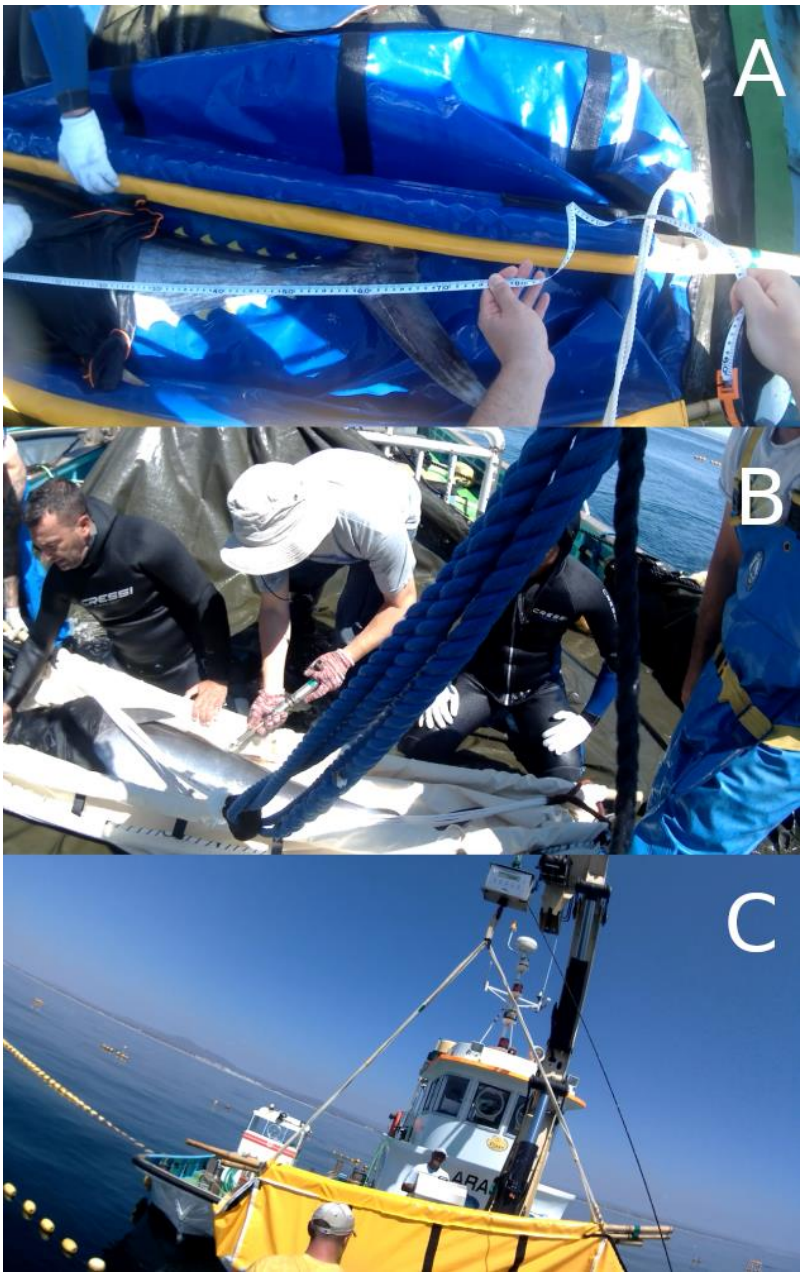

Figure 3 – A: Measurement of Straight Fork Length; B- Double conventional tagging; and C – Total live weight (scale on top of image)

All fish were moved from the capture cage (PP11) to the monitoring cage (PP18) and the amount and type of food provided to the fishes in the monitoring cage was recorded on a daily basis (Annex 3). Weights and lengths and date of harvesting for all non-tagged fish farmed together with the tagged fish is included in Annex 4.

Surface and bottom temperature, air temperature, current direction and speed, wind direction and speed, water visibility (m), wave height and direction and cloud coverage were recorded on a daily basis, most of the days during the morning and afternoon (Annex 5). Deliberate harvesting of tagged fish was scheduled to start one month after the last tagging date (21st Aug), but 4 fish were accidentally harvested before that date (due to low visibility). All harvested fish (both tagged and others not tagged) were sacrificed underwater with a "lupara" and individually weighted using the same digital scale and measured with a tape (SFL). After harvesting, biological samples of spines (23), muscle tissue for genetics (25) and otoliths (7) were collected, and these samples will be provided to ICCAT.

Differences between the two weighting methods (on board vs stereoscopic-camera) was analyzed using a Wilcoxon Signed-Rank test. The L-W relationship at tagging event was compared to the ICCAT L-W equation and Fulton's condition factor (K) at tagging for each individual was calculated using the equation (Fulton, 1904 *in* Nash, Valencia and Geffen, 2006):

K = 100*(W/L^3)

where W is the Weight in grams and L is the Length in cm

A description of number of fish that died (i.e. not deliberately harvested), and those that were harvested and were not recovered, is provided. The length and weight distribution at tagging event and at-harvesting is provided, as well as an analysis of the weight increase during caging.

All analysis was carried out using R version 3.6.3 (R Core Team, 2020)

4.2) Results and Discussion

An analysis of the weight measured on board vs the weight estimated by the stereo camera AM100 showed that for the 69 fish that it was possible to obtain a measurement with the camera, the weights obtained were not statistically different (Wilcoxon Signed-Rank test, p-value = 0.83). There was a slight and not statistically significant overestimation of the total weight by the stereo camera compared to the on board weighting by less than 3% (188kg more in a total of 6754kg). Nevertheless, there were differences in individual

length that ranged from -18.8% to +21.2% (Average±STDEV 2,2±7,3%), making the Stereo Camera measurements less suited for individual growth estimation in this area.

The fish captured presented a low Condition Factor (Average±STDEV 1.65±0.17), which is expected given that they are spent and migrating to the feeding grounds. In fact, the estimated value of K for fish migrating into the Atlantic (Non fatted low fattening condition) obtained from the equation by Santos et al. (2003) is 1.4.

After the final round of harvesting in cage/pool PP11 where fish were concentrated, from the originally tagged 89 fish, 34 tagged fish were harvested while 18 were not recovered and 37 were found dead after tagging (Table 1 and Figure 4). This represented a mortality of nearly 42%. Out of these only 4 were reported as Scientific Quota (with all RMAs sent to ICCAT/GBYP within the 24h period) with a summed total weight of 321kg, while the remaining 33 dead fish were included in the Tunipex quota.

Table 1 - Number of fish Harvested, Dead and Unrecovered per 10cm size class

SFL Class	Harvested	Dead	Unrecovered	Total
110	0	2	2	4
120	0	2	4	6
130	3	1	3	7
140	4	2	3	9
150	6	3	0	9
160	6	7	0	13
170	0	5	1	6
180	3	4	1	8
190	1	3	0	4
200	3	1	1	5
210	6	2	1	9
220	0	3	0	3
230	2	2	1	5
240	0	0	1	1
Total Result	**34**	**37**	**18**	**89**

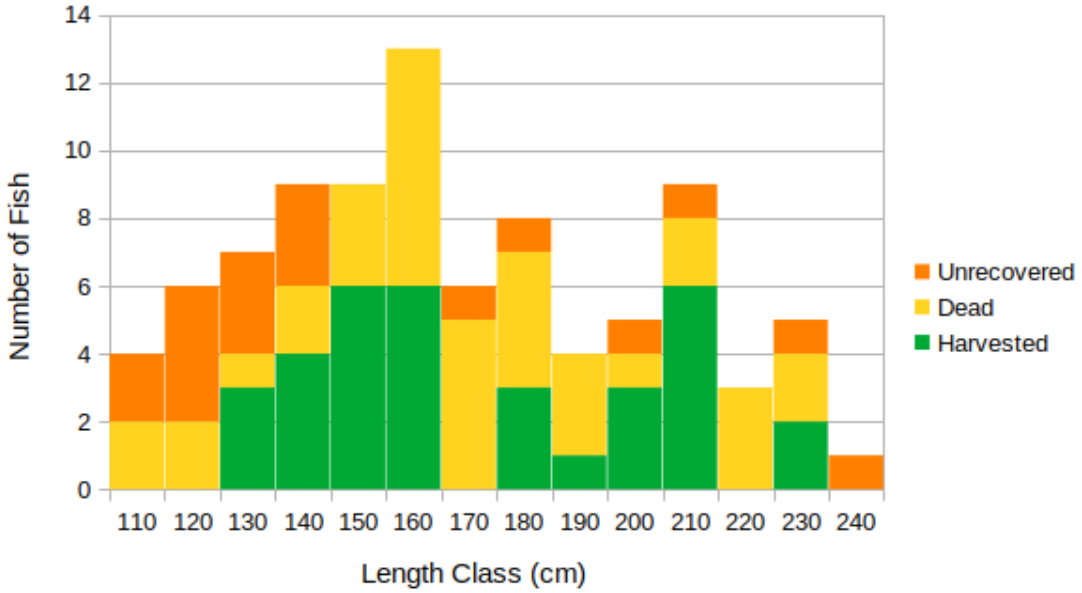

Figure 4- Number of fish tagged per 10cm size class with Number of Fish Harvested (Green), Dead (Yellow) and Unrecovered (Orange).

The unexpected high mortality could have been caused by a combination of the tagging stress and the handling of the fish. As mentioned before, these fish were in low condition as can be observed from Figure 5. The majority of fish showed a lower RWT than the weight estimated using ICCAT's L-W equation for BFT and closer to the local equation estimated by Santos et al. (2003) for Unfattened fish migrating to the Atlantic.

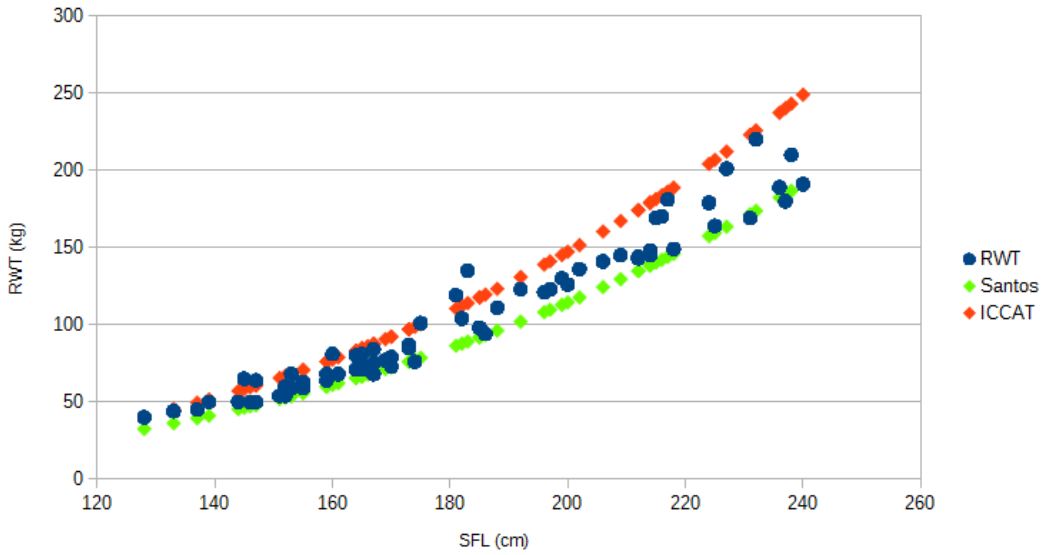

Figure 5- Relation between round weight (RWT) and straight fork length (SFL) at tagging compared to estimated weight at the same length using the ICCAT L-W equation (red dots) and the Santos et al (2003) (green dots).

Regarding the unrecovered fish, sorting data by tagging date (Table 2 and Figure 6), it can be observed that the majority (78%) of the unrecovered fish occurred from the first day of tagging.

Table 2 - Number of fish Harvested, Dead and Unrecovered per Tagging date

Tagging date	Harvested	Dead	Unrecovered
27-06-2019	3	3	14
02-07-2019	8	4	2
09-07-2019	7	10	0
10-07-2019	2	3	1
26-07-2019	6	13	1
21-08-2019	8	4	0
Total Result	**34**	**37**	**18**

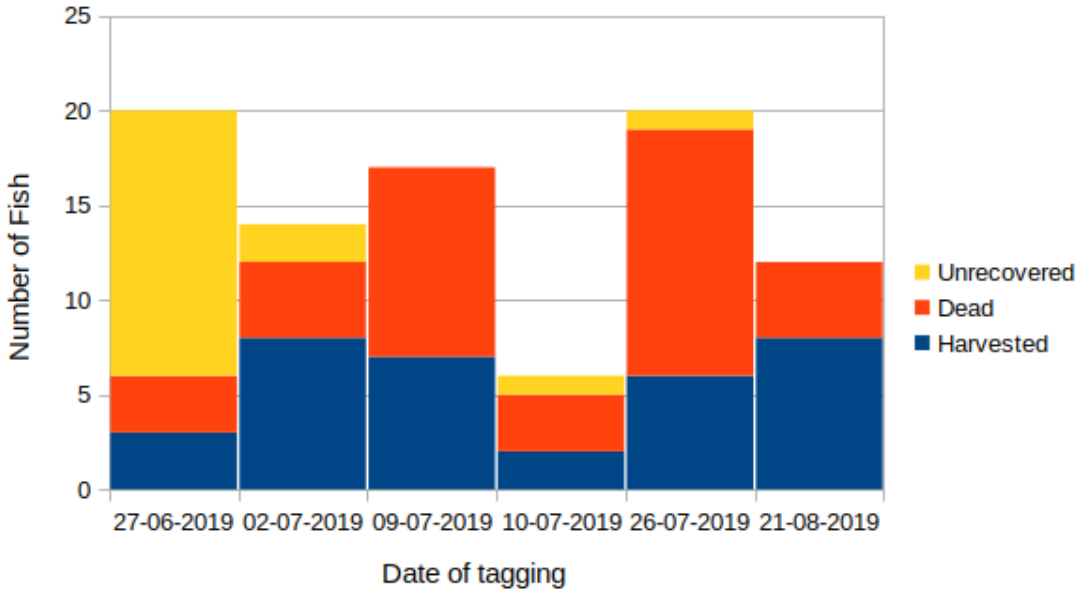

Figure 6 - Number of fish tagged per date with indication of Harvested, Dead and Unrecovered.

A possible explanation for this is that although tags were implanted in the muscle below the second dorsal fin to a depth of 5cm as usual (the same as the dual barbel tags in double tagging experiments), this could be too shallow. At the Tagging workshop (4-5th of July) it was noted that inserting the PSATs, as well as the conventional tags, a bit deeper could be beneficial for tag recovery. As such, from the third tagging day onward tags were inserted to a depth of 8cm. Since extreme tag loss was not recorded in the second day of tagging (where tags were still inserted to a depth of 5cm), tag loss from the fish tagged on the first day could also be attributed to unknown factors other than tag insertion depth.

Another important point to take note is that the fish tags were reported by the Trap Coordinator to be covered with algae growth upon harvesting requiring the tag to be cleaned to distinguish from parasites. This happened in most of the tags even the ones that were only a few months in the water. In the Trap Coordinator's opinion this algal growth could have caused increased drag that promoted tag shedding.

At the final stage of harvesting several fish were observed with double scars at the position of the tagging, indicating the tag loss, but unfortunately this was not fully documented by the harvesters. Even so, it is a qualitatively important observation that shows that tag shedding took place in some of the tagged tunas.

For the harvested fish, a brief analysis of the weight data shows (Figure 7) that weight increase does not start immediately. The overall weight increase of the harvested BFT varied between 0% and 54,8% (mean = 25.5, SD = 13.2), noting that those values corresponded to fish that were harvested between 41 and 129 days after tagging. Weight increase up to 129 days exceeded 50% only for a single harvested fish.

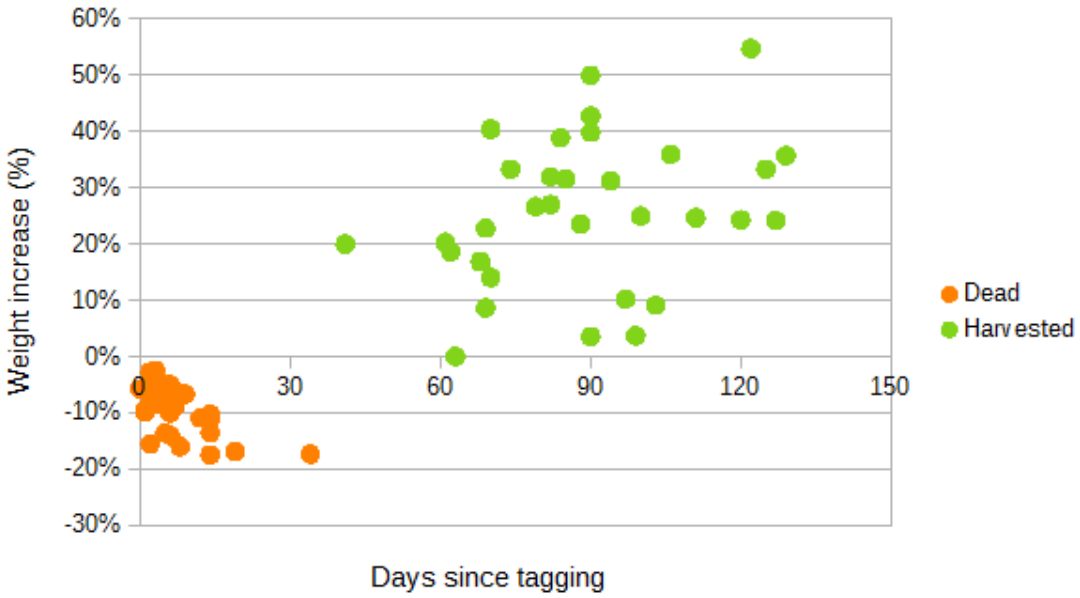

Figure 7- Weight increase as a proportion of initial weight with time after tagging.

According to the Tunipex staff it is normal that captured fish do not feed for 3 to 4 days after capture, so the initial weight decrease is normal. This could also be an effect of water/blood loss after harvesting which would be more relevant initially, before somatic weight growth is measurable.

Nevertheless, the increase in weight for the tagged fish was within the expected values since their weight at harvesting is within the interval of untagged fish (Figure 8). Fitting linear regressions to the logaritmized values of SFL and RWT for tagged and untagged fish, returns respectively a = -11,35 b= 3,08 r2=0,9481 and a = -11,203 b=3,053 r2=0,9597

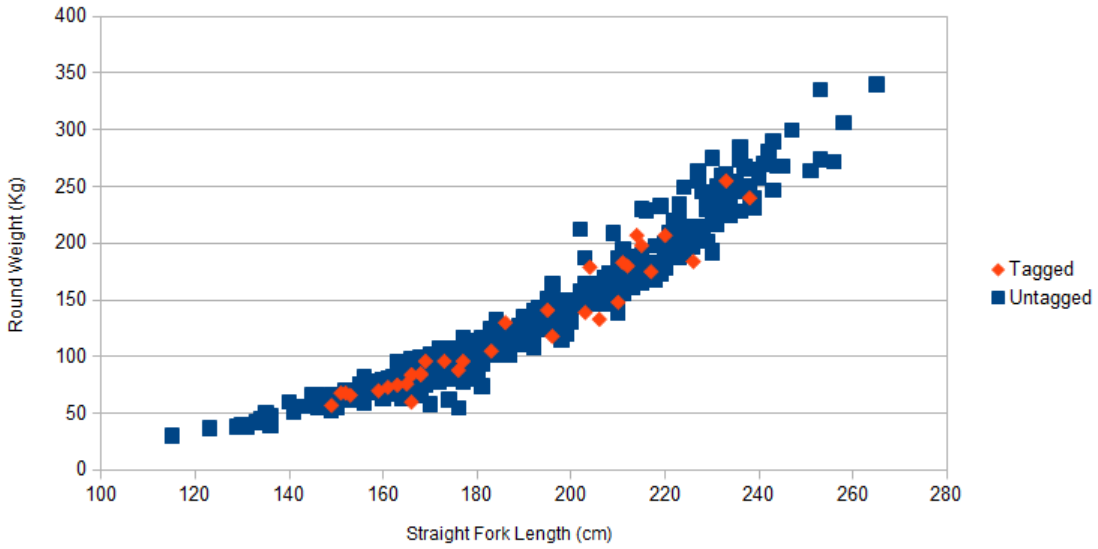

Figure 8- Straight Fork Length vs Round Weight of all fish in the monitored cage harvested after September 5th (date the first tagged fish was harvested).

An ANCOVA test shows that the factor Tag is not affecting the relation between length and weight of harvested fish (p-value > 0.95)

5) Conclusions and Recommendations

- The overall weight increase for the harvested fish varied between 0% and 54.8%, with a mean of 25.5% (one outlier with -44% change was not considered). Those fish were fattened in the BFT farm between 41 and 129 days between tagging and harvesting. We note that the condition factor of those fish when tagged was low, possibility meaning that their potential growth in weight was high in a relatively short period of time.

- The mortality of tagged fish over this project was relatively high. As stated in the previous point, the Fulton's condition factor of those BFT (migrating after spawning) was low, which may have contributed to such high tagging mortality.

- Tag shedding was also relatively high during the project. We note that the conventional tags used showed a very high algae growth after just some weeks or few months of tagging. That additional drag produced can have contributed to tag shedding. We would recommend to use tags with some anti-fouling to prevent algae growth in the future.

- Although very high tag shedding occurred on a single date, this could be related to tag insertion depth. As a precautionary measure we recommend that tag insertion in the muscle is done to at least 8cm depth.

- Tagging and handling does not seem to have affected weight gain significantly since at harvesting tagged fish weight was not significantly different from non-tagged fish of the same length.

-To deepen in the causes of the high observed variability and for getting more representative and conclusive results, we recommend to continue this work with additional fish tagged in future BFT fishing seasons.

6) Acknowledgements

The authors would like to thank all the staff from Tunipex and in particular to the divers for capturing and placing/removing on/from the stretcher the very lively tunas and assisting the onboard tagging.

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7) References

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