# TUNA OCEAN RESTOCKING (TOR) PILOT STUDY - LONG-TERM GROWTH RATES AND FOOD CONVERSION RATIOS IN ATLANTIC BLUEFIN TUNA BROODSTOCK IN CAPTIVITY

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### SUMMARY

In 2018 a broodstock cage containing 48 adult Atlantic bluefin tuna (ABFT) was established 6 km off the coast of Malta from fish caught in Tunisian waters in July 2017. At transfer to a grow out cage stereo camera determinations of length were made and initial biomass calculated. Feeding was three times a week (1% body weight/day) with MSC certified baitfish. Water temperature at top and bottom of the cage were monitored from April 1st, 2018 until 4 October 2020 (30 months) when the remaining fish (43) were harvested and Standard Fork Length (SFL), Round Weight (RWT). Length weight relationships between wild fish and the broodstock held in a cage facility showed a much greater increase in weight for a given length in captive fish. Growth rates in the facility over a 30 months period averaged 7.1% of the RWT per month with an average increase in weight per fish of 213% over 30 months. From the known amount of feed and the end biomass of the fish on harvesting, a food conversion ratio (FCR) of 13,4 was calculated.

# RÉSUMÉ

En 2018, une cage de géniteurs contenant 48 thons rouges adultes de l'Atlantique (ABFT) a été mise en place à 6 km au large de la côte de Malte, provenant de poissons capturés dans les eaux tunisiennes en juillet 2017. Lors du transfert dans une cage d'engraissement, des déterminations de la longueur par caméra stéréoscopique ont été réalisées et la biomasse initiale a été calculée. L'alimentation était effectuée trois fois par semaine (1% poids corporel/jour) avec des poissons-appâts certifiés MSC. La température de l'eau dans la partie supérieure et inférieure de la cage a été surveillée du 1<sup>er</sup> avril 2018 jusqu'au 4 octobre 2020 (30 mois) lorsque les poissons restants (43) ont été mis à mort, tout comme la longueur à la fourche standard (SFL) et le poids vif (RWT). Les relations longueur-poids entre les poissons en liberté et les géniteurs maintenus dans la cage indiquaient un gain de poids bien plus important pour une longueur donnée chez les poissons en captivité. Les taux de croissance dans la cage pendant une période de 30 mois étaient en moyenne de 7,1% du RWT par mois avec un gain de poids moyen par poisson de 213% sur 30 mois. D'après le volume connu d'alimentation et de biomasse finale des poissons à la mise à mort, un coefficient de transformation des aliments (FCR) de 13,4 a été calculé.

#### RESUMEN

En 2018 se estableció una jaula de cría que contenía 48 ejemplares adultos de atún rojo del Atlántico (ABFT) a 6 km de la costa de Malta a partir de peces capturados en aguas de Túnez en julio de 2017. En el momento de la transferencia a una jaula de crecimiento se realizaron determinaciones de talla con cámara estereoscópica y se calculó la biomasa inicial. La alimentación se realizó tres veces por semana (1 % del peso corporal/día) con peces de cebo certificados por el MSC. La temperatura del agua en la parte superior e inferior de la jaula se monitorizó desde el 1 de abril de 2018 hasta el 4 de octubre de 2020 (30 meses), cuando se sacrificaron los peces restantes (43) y se determinó la longitud a la horquilla estándar (SFL), el peso en vivo (RWT). Las relaciones talla-peso entre los peces en estado salvaje y los reproductores mantenidos en jaulas mostraron un aumento de peso mucho mayor para una talla determinada de los peces en cautividad. Las tasas de crecimiento en la instalación durante un periodo de 30 meses fueron de una media del 7,1 % del RWT por mes, con un aumento medio del peso por pez del 213 % en 30 meses. A partir de la cantidad de alimento conocida y de la biomasa final de los peces en el momento del sacrificio, se calculó un índice de conversión alimentaria (FCR) de 13,4.

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### **KEYWORDS**

*Thunnus thynnus, broodstock, aquaculture, long-term growth rates, tuna ranching, length weight relationships, size classes, feeding, food conversion ratio (FCR)* 

### 1. Introduction

For full cycle aquaculture of Atlantic bluefin tuna (Thunnus thynnus) to succeed, then information is required on the best husbandry techniques to be used when these fish are held as a broodstock in captivity. The present study therefore endeavours to look at some of the key parameters such as growth and feeding in captivity, in other words within cages. The practice of ranching or farming of Atlantic bluefin tuna is derived out of the methodology developed in Australia by Croatian immigrants for the Southern bluefin tuna (Thunnus maccoyii) and then developed back in Croatia in 1996. As of 2021 the ICCAT record of BFT farming facilities includes the following countries, Albania, Croatia, Cyprus, Spain, Greece, Italy, Malta, Portugal, Egypt, Libya, Morocco, Tunisia and Turkey with a total of 63 farms in all with a capacity of 72,585 tons. Farming or fattening activities now account for over 60% of the catch which was limited by quota in 2021 to 36,000 tons in the Eastern Atlantic and Mediterranean. The normal procedure in these farms is that the fish are fattened for 3 to 6 months before harvesting. The duration of fattening varies in different regions and may be dependent upon market forces. In Croatia where a special dispensation allows the catching of small tuna (8 kg+) two different fattening regimes are used of 18 or 30 months depending upon commercial requirements. Long-term fattening cycles are relatively rare due to the costs of providing feed, although the profit margin may increase with larger fish. The present study has been carried out with the collaboration of a commercial farm and, while not designed to measure fattening, it gives some insight into long-term growth rates in farms under different feeding regimes.

In a previous report of the SCRS (Anon, 2010) the role of standardised length weight relationships taken from wild fish and using them to back estimate the initial catch and therefore to monitor compliance to quota, have been raised. Ortiz (2015; 2018) and Ortiz *et al.* (2014) have presented some preliminary evaluations of potential growth of fattened/farmed Atlantic bluefin tuna based on harvesting data which was available up to this period including data from the regional observer program (ROP-BFT). Recently Deguara *et al.* (2021) highlighted the need for new data on growth in farms as the dependence of the compliance to quota levels and the following increasing biomass during fattening needs to be assessed and in particular maximum growth rates established and the ICCAT GBYP programme has specifically looked at growth in farms (Deguara, 2019; 2021).

Within the last three years a pilot ABFT restocking project was established in Malta and has provided millions of pre-incubated DNA tagged larvae for release into the sea, thus avoiding egg predators, before they hatch and assisting the spawning of ABFT. At the same time molecular markers were used to overcome the problems of previous mark and recapture experiments in terms of numbers and assessment of success rates. The results of this part of the study can be found in Bridges *et al.* (2020). During the present study it has been possible to examine growth in captivity over a longer term (30 months). At the same time environmental parameters were measured together with feeding regimes (developed in previous EU projects: SELDOTT, 2011; TRANSDOTT, 2014) for the broodstock fish before harvesting such that these data could be used to assess growth rates and food conversion ratios of the fish in captivity.

# 2. Material and Methods

Initially 48 ABFT were placed in a sea cage ( $\emptyset$  30 m, depth 20 m) as a broodstock at a low stocking density to support animal welfare. Broodstock management and supplementary feeding was according to proven methods during the decade of research carried out in EU DOTT projects. No antibiotics were used for the captive broodstock and during the period of three years the mortality rate was relatively low (<1 fish per 6 months).

### 2.1 Broodstock Cage

In April 2018 a pilot broodstock cage (30 m in diameter and 20 m deep) containing 48 adult Atlantic bluefin tuna was established by Malta Fish Farming Ltd (MFF) under the direction of TUNATECH at a site (**Figure 1a**) approximately 6 km off the south east coast of Malta. This cage was later modified by the removal of the railing (**Figure 1b**) which was the final configuration before harvesting. These fish were part of the total catch of 1407 fish caught in Tunisian waters and transferred from the towing cage to the growout cage on the 28<sup>th</sup> July 2017 of which 20% were used for biomass calculations using stereo camera (SC) observations. In April 2018 48 fish with

a biomass of 4985 kg (mean weight = 103,9 kg) were then transferred to the broodstock cage where the broodstock experiments began. At the end of the experimental period in October 2020 the tuna still present in the cage were harvested and their empirical Standard Fork Length (SFL) and Round Weight (RWT) were determined.

# 2.2 In situ Temperature Monitoring

Measurements weere carried out using the standardised "Onset Hobo" data loggers as shown below (**Figure 2**) which were attached the top and bottom of the cage itself and then at specific intervals read by a diver-assisted readout system (see Bridges *et al.*, 2019 for details). Recordings were made throughout the whole of the year such that careful monitoring of environmental conditions could be made. During feeding periods extra temperature measurements were made by the divers who were monitoring the feeding of the fish.

# 2.3 Feeding

Fish were fed on a regular basis (see results section) with amounts varying from 180 - 550 kg which mainly consisted of baitfish from MSC sources. As can be seen in the figure (**Figure 1a, b**) a feeding cage was used into which the thawing frozen baitfish were placed (**Figure 3a**) and consumed by the fish underwater (**Figure 3b**). Feeding was observed by divers and excessive overfeeding was avoided. Winter feeding was maintained at lower levels and in the summer between the months of April to July the diet was supplemented with squid from 100 to 300 kg / week.

### 2.4 Harvesting

Fish were harvested in October 2020 by Malta Fish Farming Ltd using normal ICCAT procedures with the fish being euthanised underwater and then transferred to a support vessel for further elaboration. At this time individual RWT in kilograms and SFL in centimetres were determined. In a separate operation from a normal fattening cage of MFF, which contained fish which had been fed for a period of approximately three months, a sample of 12 fish of different sizes were taken and RWT and SFL determined as well. At the same time DNA samples were also taken from all fish in the present study for later analysis of sex and parental designation of spawning activity.

### 3. Results

### **Temperature**

Daily temperature measurements were made at intervals of 15 minutes using data loggers installed at both the top (within 1 m of the surface) and at the bottom of the cage (**Figure 2**). The results (**Figure 4a and 4b**) indicate a winter temperature of around 15 to  $16^{\circ}$  and a maximum temperature in summer of up to  $28^{\circ}$ C. There is discrepancy in temperature between the top and bottom of the cage - which is at 20 m - of approximately  $2^{\circ}$ C in winter. In summer, this depends very much on the formation of a thermocline, which can be dramatic as seen in in 2020 with a  $4^{\circ}$ C difference between top and bottom of the cage and large swings in bottom temperature. It must be remembered that these cages are 6 km offshore in depths of water which are over 100 m deep. The surface temperature increases continually from March onwards, but there is a rapidly decline from a bottom temperature of  $19^{\circ}$ C in October to  $15^{\circ}$ C in December/January. The data for 2019 is not shown as there were some problems with the loss of data loggers in stormy weather and the recordings were incomplete. However, they showed a similar temperature pattern to those experienced in 2018 and 2020.

# Feeding

Over the whole experimental period ABFT were fed on 420/903 days (46%) and daily recordings were made of the amounts of fish or squid which were given to the fish. When squid was fed during the reproductive buildup from April to June this was done on days when no baitfish was used. In total baitfish was fed on 359 days compared with 61 days for squid over a 30 month period from April 2018 until October 2020. The cumulative total feed as shown in the diagram (**Figure 5**) shows a comparison of the feeding strategy over the 30 month period. As this is a broodstock the feeding regime is different to that of a normal fattening farm when fish would be fed every day if possible, dependent upon the weather. In the present study 46% feeding rate appears to be sufficient to maintain the fish and also produce gametes during the reproductive season. The overall feed diagram (**Figure 6**) indicates clearly that squid is only a small proportion of the total feed which consisted mainly of MSC baitfish from a known source. The total amounts of feed per year were roughly equivalent throughout the study and feeding was

performed on a continual basis with only small decreases in the winter months. In a farming/fattening situation fish are only fed for approximately four months intensively every day and if carryover fish are to be held over the winter months, then feeding is decreased.

# Length Frequency Distribution

A length frequency distribution analysis was carried out using data obtained from the stereo camera on the initial transfer from towing cage to grow out cage. On transfer (July 2017) of the initial 1407 fish to the grow-out cage 20% were sampled for length and the length frequency distribution is shown in the diagram (**Figure 7**). It would appear that there are two major cohorts one of smaller fish with a peak at 121 to 125 cm and the second cohort of larger fish with a length between 205 and 210 cm. The mean initial weight of the fish transferred to the broodstock cage (April 2018) from the grow out cage was calculated from stereo camera measurements of length which were converted to weights using the equation of Rodriguez-Marin *et al.* (2015) and indicated by the arrow in the diagram (**Figure 7**). Further analysis on harvesting used direct measurements of length to establish the frequency of different length classes empirically and shows a shift of the size classes from an initial value of 180 cm to a final value of 241 to 245 cm.

### Length/Weight Relationships

The length/weight relationships (after 30 months) of the 43 harvested broodstock fish are shown in the figure (**Figure 8a**) together with the mean and standard deviation values for SFL and RWT. The initial SC transfer data from the towing cage to farm grow out cage for the 20% length sample is also shown (**Figure 8b**). These measurements are based on stereo camera measurements of SFL and the growth equation of Rodriguez-Marin *et al.* (2015) to calculate RWT. Further estimations of the length/weight relationships in MFF harvested fish (3 months fattening) were made from both empirical measurements on length and weight gathered at harvest (**Figure 8b**). Comparative data for small Croatian tuna (2yr+) are also presented in the figure from the work of Katavic *et al.* (2018) (**Figure 8b**).

# Growth Rate and Feed

The overall biometric data for the broodstock and a summary of the feeding experiment are illustrated in the table (**Table 1**). It can be seen that the average body weight increase (BWI) was 213 % over a period of 30 months this can be used to calculate an average percentage growth in RWT per month of 7% for fish from the size range from 103 to 325 kg. The average SFL increase over 30 months was 62 cm given an average monthly increase of 2 cm or 1.1% increase in SFL per month. In total the 43 fish consumed 127 tons of feed over the 30-month period. The food conversion ratio (FCR) was therefore calculated to be 13.4 (based on a wet weight feed) this would equate to a dry feed FCR of 4.6 (assuming a conversion factor for Mackerel of 3,0; Cresson *et al.*, 2017).

# 4. Discussion

Holding broodstock in captivity is an important precursor for any successful full cycle aquaculture operation, especially for long periods of time. This is in contrast to the short fattening periods used within a commercial farm, although as stated in Croatia these captivity periods may extend up to 30 months. As stated in the introduction the farming/fattening or ranching as it is termed in some countries has expanded over the last 20 years and it is becoming increasingly important to study the growth rates of Atlantic bluefin tuna in such facilities. Numerous previous publications (Anon, 2010; Ortiz, 2018 and Deguara et al., 2021) have pointed out the need for accurate measurements in the policing of compliance regulations in terms of quota. Similar questions have been raised for the Southern bluefin tuna and considerations made of growth rates in other Tunnid species (for review see Jefferies, 2016) Although much empirical data from fisheries and from farming in terms of RWT and SFL at harvesting is available, very few studies have actually directly looked at growth rates at known feeding levels. Aguado-Giménez and Garcia-Garcia (2005) were one of the first to examine this in a farming situation using video camera techniques to determine length biomass by calculation from length weight relationships of empirical data of harvested fish. Their average growth RWT per month was 12% in 32 kg fish compared to only 2.2% in 219 kg fish. Using data from two recent specific growth studies reports (Deguara, 2019; 2021) it is possible to calculate average growth rates from average weight gains for fattened fish over a 3 to 4 months summer period in 2019 of 11.3% body weight per month for a cage with a large number of fish (2580). In a later study (Deguara, 2021) on carryover fish (156) this had decreased to 3.7% body weight per month but was over a 12 month period for larger fish and included winter months when feed amounts were reduced. This compares with 7.1% RWT per month in the present study (Table 1) for 103 to 325 kg fish kept as broodstock again with a completely different feeding regime. In the present study with the broodstock a feeding rate of approximately1% of body weight per day was used. This compares to 1,85% and 2,47% in Deguara (2019, 2021)studies on commercial fattening cages. It is quite evident that the feeding regime influences growth rates as would be expected and so direct comparisons of growth rates between different studies need to take into account these differences together with other environmental and behavioural parameters.

In his review of various species Jeffriess (2016) pointed out that in many studies for the Mediterranean Atlantic bluefin tuna growth in farms was much higher than in the wild especially in smaller fish. From the data of Tzoumas *et al.* (2010) calculated average growth RWT increases per month of 4.5-5.4 for fish between 205 and 295 cm SFL over a 6-to-7-month period can be calculated. On plotting SFL against RWT a clear difference was seen by these authors between fattened and wild fish as in the present study. It should be noted (**Figure 8b**) that the initial transfer data for wild fish from the stereo camera measurements indicate that a fish of 240 cm plus SFL would have a RWT of approximately 250 kg. The broodstock in the present study grew over a period of 30 months to an average SFL of 241 cm but with an average RWT of 325 kg indicating a 30% greater increase in the broodstock fish compared to the wild fish. The reason for this is quite clear. In the wild tuna must forage for their prey continuously to maintain their high metabolic rate and maintain their various behavioural activities. In a fattening cage food is provided on a regular basis to satiation and therefore the metabolic costs of obtaining food are much lower. A comparison of the MFF harvested length weight relationship for the production cage fish held for three months from July until October (**Figure 8b**) indicates that the broodstock harvested fish lie on the same exponential trend line but obviously within a limited size and weight range (**Figure 8a**).

It is clear that in most farming situations environmental and husbandry details are important in the present study is shown by the temperature monitoring there can be large differences even within a confined caged environment especially when these are offshore (6 km+). In the present study a small cage (30 m) with a relatively small number of fish (46) were fed with a broodstock feeding regime, commercial cages may be 90 to 100m in diameter and contain over 2500 fish (Deguara, 2019).

As feed and feeding play such an important role in the maintenance of a broodstock it is important to look at the food conversion ratio for captive fish. To ensure that the welfare of broodstock is an important part of husbandry, "A healthy fish will produce healthy eggs". In the present study with our feeding regime an average FCR of 13.4 was obtained over 30 months (including two winters) for fish between 100 and 325 kg. Estimates for FCR values in tuna vary considerably (Otolengi *et al.*, 2004; Mrčelić *et al.*, 2020) with values as high as 30 being reported and as low as 7 (Jeffriess, 2016). In their original study. Aguado-Giménez and Garcia-Garcia (2005) calculated values of 24.8 for 255 kg fish and 15.3 for 63 kg fish over a nine-month period from February till October. Again, using the data from the reports of (Deguara, 2019; 2021) one can estimate and FCR of 5.5 for 225 kg fish over a period of four months and 9.0 for 320 kg carryover fish for a period of one year. It would appear that the FCR rates are dependent upon fish size and duration of feeding. Aguado-Giménez and Garcia-Garcia (2005) also converted the FCR's to dry weights with values of 7.4 and 4.6 respectively which concurs with the value of 4,6 in the present study. In one of the few studies carried out with a dry feed, Jeffriess (review 2016) reports values from Japanese studies on Pacific bluefin tuna (*Thunnus orientalis*) with FCR's of 3.8 and 3.7 for small tuna.

In conclusion, it can be said that the maintenance of a captive broodstock for Atlantic bluefin tuna can be achieved with the present commercial practices available. Growth rates and food conversion ratios will be dependent on a number of physical parameters such as fish numbers, cage size, temperature and season. Furthermore, feeding amounts and feeding regimes (number of days feeding) together with the size of fish kept within a cage (behavioural components) will influence growth rates. It is quite clear that growth in captive fish does not represent growth in wild fish since wild fish must continuously, actively forage for pelagic species whereas in a caged situation this activity falls away, therefore the energy budget for growth increases.

# 5. Acknowledgments

The authors would like to thank the divers, boat crews and technical staff of MFF who made this study possible. Dr. Simeon Deguara and the AquaBiotech Group kindly supplied length/weight measurements and DNA samples from the MFF production cage. MARS Petcare Europe have sponsored this project and we are grateful to them for their continued support.

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Parameter	Initial	Final	Comment
Feeding Period	01.04.2018	09.10.2020	30 months
Number of Broodstock	48	43	Mortality (<1 fish per 6 months)
Total Biomass	4985 kg	13992 kg	Initial through SC and final harvest weight
Avg. SFL (+/- SD)	180 cms*	241,6 +/- 8,8 cms	*Calculated from SC
Avg. RWT (+/- SD)	103,8 kg*	325,4 +/- 6,3 kg	*Calculated from SC
Avg. % BWI / Fish	0	213,3	Over 30 months
% Growth RWT/Month	0	7,1	For fish size range 103 -325 kg
Avg. SFL Increase	0	61,6 cms	Over 30 months
Avg. SFL Incr. /Month	0	2,05 cms	1,1 % increase in SFL/month
Total Feed Consumed	0	127360 kg	Includes all types of feeds
Biomass Loss	0	5x103,8 =519 kg	Through mortality
Total Biomass Gain	0	9526 kg	For 43 fish
Food Conversion Ratio	0	13,4	Feed as wet weight

**Table 1.** Showing initial biometric data for broodstock fish and final data at harvesting together with total feed quantites and growth estimates and food conversion ratios.



**Figure 1.** a) Shows the offshore broodstock cage at the MFF facility in Malta. The cage dimensions were 30 m diameter and 20 m deep. b) Shows the final configuration the MSC fish feeding box can be see in the centre.



**Figure 2.** The cage temperature monitoring system conssisting of a "Onset HOBO" light and temperature sensor attached to the cage bottom – Sensors were attached to both top and bottom of the cage and recorded light an temperature at 15 minute intervals.



**Figure 3. a.** Feeding broodstock with MSC certified herring placing a "big bag" and lowered into the fish feeding box. **b**. ABFT feeding on thawing frozen fish within the cage.





**Figure 4. a.** Data logger seawater temperatures in broodstock cage top and bottom for 2018. Data is only shown between April and August. **Figure 4.b.** Data logger seawater temperatures in broodstock cage for 2020. Data is only shown between April and August. Winter temperatures remain relatively constant around 15 to 16°C.



**Figure 5.** Cumulative total feed per year from April 2018 till October 2020. During the months from April till July tuna were fed a supplement of squid to enhance reproductive capacity.



**Figure 6.** Overall quantities of feed per year showing bait fish and squid amounts separately and total feed per year in kilograms. The number of days feeding per experimental period is shown in brackets.



**Figure 7.** Length frequency data for all fish at transfer from towing cage to grow out cage and length frequency data for harvested broodstock after 30 months in the farm. Initial calculated weight in April 2018 from stereo camera determinations.



**Figure 8 a.** Standard fork length versus round weight relationship for the harvested broodstock in October 2020. The mean values for length and weight for all 43 fish are also shown



**Figure 8b.** Actual measured round weight (RWT) and standard fork length (SFL) at harvesting of the broodstock after 30 months compared with MFF harvesting of fish after 3 months. These are empirical data of actual measurements. Comparative calculated data from known equations from the literature for small Croatian tuna (Katavic *et al*, 2018) small fish are shown. The calculated average initial broodstock length and weight from SC measurements is also shown ( $\bullet$ ). Data from SC measurements of the initial transfer from towing cage to grow out cage in 2017 using the equation of Rodriguez-Marin *et al.* (2015) are also shown.