In situ target strength of bigeye tuna (Thunnus obesus) associated with fish aggregating devices

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Bigeye tuna (Thunnus obesus) is an important commercial fish species, which aggregates around fish aggregating devices (FADs) together with other tropical tuna species. Acoustics is the main technology used by fishers and scientists for the location and quantification of tunas at FADs. However, currently it is not possible to reliably discriminate between the different tropical tuna species that are found together at FADs using acoustic methods, which hampers the development of selective fishing needed to preserve some of the tropical tuna species for which overfishing is occurring. One of the prerequisites for species discrimination is to know the target strength (TS) of each species at different frequencies. This paper measures in situ TS values and explores the frequency response of bigeye tuna at FADs in the central Pacific Ocean using three different acoustic frequencies. For the range of body length caught (40–100 cm), the obtained b20 values were /C0 65, /C0 66, and /C0 72 dB for 38, 120, and 200 kHz, respectively. The decreasing frequency response pattern obtained for this swimbladder bearing species contrasts with the opposite pattern previously observed for skipjack tuna (bladder-less), the most abundant tuna species found at FADs, hence allowing the potential for discrimination between the two species.

Keywords: acoustics, bigeye, biomass, FAD, frequency response, selectivity, target strength, tropical tuna

Introduction

Bigeye tuna (Thunnus obesus) is a high value commercial species present in the subtropical and tropical areas of the Atlantic, Indian, and Pacific Oceans. Bigeye can be observed either in free schools or associated with floating objects. Juvenile bigeye are caught on the surface by a range of gears including handline, ringnet, and purse-seine and are used mainly for canning, whereas most of larger/older fish are caught by longline fishery for the sashimi market.

Juveniles of bigeye tuna are normally caught, along with skipjack (Katsuwonus pelamis) and juvenile yellowfin tuna (Thunnus albacares), associated with fish aggregating devices (FADs), which are artificial floating objects built by fishers to aggregate tuna (Kingsford, 1993; Parin and Fedoryako, 1999). Nowadays FADs are geolocated with a buoy equipped with an echosounder to provide remote estimates of the amount of tuna aggregated around the FAD (Lopez et al., 2014). Currently, catches around FADs represent around 65% of the tuna catches of purse-seiners (average for the three tropical oceans; Scott and Lopez, 2014).

In the eastern Pacific Ocean, all bigeye indicators, except for catch, show strong trends over time indicating increasing fishing mortality and reduced abundance (Xu et al., 2018). In the Western and Central Pacific Fisheries Commission, recent assessment of the bigeye stock showed a more optimistic status for bigeye, compared with prior assessments, indicating that overfishing is likely not occurring and that the stock is not being overfished. In the Atlantic Ocean, the latest stock assessment indicated that overfishing is occurring and that the stock is in an overfished state.
(Anonymous, 2018). Only in the Indian Ocean the stock of bigeye tuna is estimated to be in good condition (International Seafood Sustainability Foundation, ISSF, 2017).

Stock assessment requires a substantial amount of information. Data on retained catch, discards, catch per unit of effort, and size compositions of the catches from different fishery are typically used. Several assumptions regarding processes such as growth, recruitment, movement, natural mortality, and fishing mortality, are also done. The interpretations of stock status are strongly dependent on those assumptions.

Because of concerns about stock status of bigeye tuna in the different regions, as well as the uncertainty derived from the assumptions in stock assessment, scientist and managers are faced with the need to find new direct data sources and biomass estimates to complement current stock assessment. Direct estimates of abundance are already undertaken in other fishery and have been proven to be effective to complement traditional stock assessment and inform management (e.g. Massé et al., 2018). In the case of tropical tuna, biomass on delayed echosounder buoys data could also be used to develop direct indices of tuna abundance (as proposed by Capello et al., 2016; Moreno et al., 2016; Santiago et al., 2017).

FADs do not only aggregate bigeye tuna: they aggregate other tuna species, such as skipjack, which is the main target species of purse-seiners working with FADs, and yellowfin, so that the three species can regularly be found together in a single FAD.

Acoustics used by fishers may represent one of the key tools not only to obtain direct indices of tropical tuna abundance, independent from catch data, but also to discriminate tuna species found at FADs before the net is set. One of the prerequisites to discriminate tuna species and assess their biomass is knowing the target strength (TS; dB re 1 m²), TS-length (L; cm) and TS-frequency (f; kHz) for the three species found at FADs. Of the three species, the frequency response of skipjack tuna, a bladder-less species, has been recently published (Boyra et al., 2018), whereas bigeye and yellowfin tuna, swimbladder bearing species, have been less studied. For yellowfin and bigeye, although there are published TSL relations, those are preliminary because of the small number of observations (Bertrand and Josse, 2000) and TS(f) relations are not available in the literature.

ISSF with the aim of reducing undesired tuna mortality has supported a series of research in collaboration with AZTI to develop acoustic methodologies to help discrimination of tropical tuna species around FADs (Moreno et al., 2019). The objective of this study is to obtain in situ TS measurements, TS-length relationship, and frequency response of bigeye tuna found associated with FADs. The acoustic characteristics of bigeye tuna will represent another step towards the objective of achieving discrimination and direct abundance estimation of the main tuna species caught at FADs.

Methods
Data collection
The cruise took place in May 2014 in the central Pacific Ocean aboard the Albatrun 3, a 115 m and 4406 GT Spanish-flagged purse-seiner. The cruise started on Christmas Island (Kiribati) on 3rd May and ended in Tarawa Island (Kiribati) on 31st May. Meteorological and oceanographic information was obtained from the CLS (Collecte Localisation Satellites, France, https://www.cls.fr). During the selected sets the conditions were 7–10 knot of wind (Beaufort 3) and 1–1.5 m of swell. Acoustic data were registered during the purse-seine sets at 20 different FADs, followed by intensive spillover sampling (Lawson, 2009) to compare acoustic data and species composition caught. The strategy to collect the data from the purse-seiner is thoroughly detailed in Boyra et al. (2018). Here, in this second work, in contrast to the previous one, no recorded sonar data were available, so we only used echosounder data.

Acoustic data collection
About 10 min before the start of each of the purse-seine sets, a workboat was attached to the drifting FAD and slowly towed it to maintain it separated from the net and the vessel. The workboat was equipped with acoustic sensors and, during the ~60 min duration of the sets, it registered TS and volume backscattering strength (SV; dB re 1 m⁻¹) data from 5 to 200 m depth. Acoustic data were collected with a Simrad EK60 echosounder with 38, 120, and 200 kHz split-beam transducers, focused vertically (Figure 1) and working with a pulse duration of 0.512 ms (Table 1). The calibration was done at the beginning of the survey, following the standard target procedure (Demer et al., 2015) with a tungsten carbide sphere of 38.1 mm.

Catch data collection
Purse-seine sets, performed with a 1800 m length × 310 m height gear, were followed by intensive sampling of the catch (between 1 and 2 tonnes per set) once the aggregation was lifted on-board. Fish samples were selected randomly to avoid bias. Species were identified and each fish in the sample measured to the nearest centimetre on flat measuring boards. The weights of sampled individuals were estimated using length–weight relations available for each species (Parks et al., 1982; Caverivière, unpublished data). These proportions by weight were then extrapolated to the total tonnage of each set, as estimated by the fishing master. The sets with more than 90% in weight (about >75%) of bigeye (Table 2) were selected for acoustic analysis to obtain TS-length relationship and acoustic frequency response.

Data analysis
Acoustic data were processed from the beginning of the set until the moment in which the net was visible in the echogram. To reduce echoes from bycatch fish species, SV and TS data were excluded if shallower than 25 m (Muir et al., 2012; Forget et al., 2015). Then, the TS echograms at each frequency were processed using a single target detection algorithm (Simrad, 1996; Soule et al., 1997) with the following settings: minimum threshold = −80 dB; normalized pulse durations = 0.9 to 1.5; maximum off-axis angles = 3°; and maximum standard deviations of phase = 0.6°.

A series of target selection filters (a school masking, a fish tracking, and a high-density filter) was applied to retain single targets of tuna and remove the echoes attributed to plankton and micronekton. The single target detection algorithm, the school processing, and the fish tracking were applied using Echoview (Myriax Inc.) software. The remaining data processing was carried out on the exported csv files using R (R Core Team, 2014).

(i) School masking. A school detection algorithm (Lawson et al., 2001) was used to retain the main aggregation. The rejected echoes from outside the aggregation were likely echoes of plankton and/or micronekton. After smoothing by a 5 × 5
convolutions, “schools” (i.e. the main aggregations around the FAD) were selected using: minimum total school length and height \( = 0.2 \text{ m} \); minimum candidate length and height \( = 0.1 \text{ m} \); and maximum vertical and horizontal linking distances \( = 5 \) and 20 m, respectively. The school detection was applied on TS echograms, and data from within the schools were attributed to tuna (Figure 2).

(ii) Fish tracking. A fish tracking analysis (Blackman, 1986), which consists in grouping targets according to their mutual spatial and temporal proximity, considering that they are successive detections of the same fish in a track, was applied to the TS single targets. The fish tracking was set up retaining only fish that had been spotted for at least three times in three different pings, allowing five missing pings in a track. The sensitivity to unpredicted changes in position (alpha) and velocity (beta) were left as default (0.7 and 0.5, respectively). The exclusion distances along the major and minor axis were set to 0.4 m. Missed ping expansion percentage was left as default (zero). The weight along the major and minor axis applied for use in target to track assignment was 30 and 40 along the vertical axis. For each track, mean TS values were calculated in linear scale.

(iii) High-density (HD) filter. We filtered those tracks located in areas of the echogram with higher densities of fish individuals, thus with higher probability of failure in detecting multiple targets of the single target detection algorithm. For this, the echogram was divided into a grid of regularly spaced cells (20 pings \( \times 10 \text{ m} \)) and the number of fish per echo integrated volume in each cell was determined as:

\[
N_V = \left( \frac{s_v}{\sigma_p} \right) V, \tag{1}
\]

where \( s_v \) is the volume backscattering coefficient, \( \sigma_p \) the backscattering cross section (Maclennan et al., 2002) and \( V \) is the sampled volume corresponding to each cell, calculated as the volume of the ideal conic section of sphere described by the beam in each ping multiplied by the number of pings \( N_p \) in each cell:

\[
V = \frac{2\pi}{3} N_p (1 - \cos(\theta/2))(z_{\text{max}}^3 - z_{\text{min}}^3), \tag{2}
\]

where \( \theta \) is the beam angle of the transducers, and \( z_{\text{min}} \) and \( z_{\text{max}} \) are the depth limits of the cell. In order to provide an objective criterion for establishing a fish density threshold (and avoid the circularity implicit in the application of a density threshold before you know the TS value that allows you to calculate the actual density) the rule by Gauthier and Rose (2001) was applied, based on the comparison of the number of fish in a given volume, \( N_V \), and the number of single targets detected by the algorithm, \( T_V \), in the same volume. We represented \( T_V \) against \( N_V \) and the threshold was chosen at the point where an increase in \( N_V \) was not accompanied by a corresponding increase in \( T_V \), i.e. when the number of single targets reached saturation because of increasing probability of detecting multiple targets (Figure 3). As the method is

![Figure 1. Sample echogram of set number 6 showing \( S_v \) (in dB) for the three frequencies, 38, 120 and 20 kHz from left to right. Each panel shows a window of \( \sim 500 \text{ pings} \times 100 \text{ m} \). Upper panels show the raw echograms and lower panels show the echograms after plankton/micronekton filtering.](https://academic.oup.com/icesjms/article/76/7/2446/5537349)

### Table 1. Configuration of the acoustic equipment and calibration parameters.

| Frequency (kHz) | 38 | 120 | 200 |
|-----------------|----|-----|-----|
| Pulse duration (\( \mu \text{s} \)) | 512 | 512 | 512 |
| Power (W)       | 2000 | 250 | 150 |
| Gain (dB)       | 26.16 | 25.96 | 27.09 |
| Gain Correction (dB) | 0.86 | -0.39 | -0.34 |
| Ath. beam angle (deg) | 6.92 | 6.38 | 6.43 |
| Along beam angle (deg) | 6.94 | 6.39 | 6.37 |
| Ref. target TS (dB) | -42.3 | -40 | -39.9 |
| TS deviation (dB) | 5 | 5 | 5 |
| RMS beam model  | 0.19 | 0.18 | 0.20 |
| RMS polynomial model | 0.16 | 0.16 | 0.15 |
insensitive to the actual $\sigma_{b20}$ used to estimate $N_{V}$ (Gauthier and Rose, 2001), a preliminary value (the mean of the unfiltered $\sigma_{b20}$ values) was used; then, after the filtering process, the $N_{V}$ values were rescaled according to the corrected TS values for consistent visualization. A windowing smoothing process was applied by grouping the fish densities, $N_{V}$, by ranges to help highlighting the pattern (Figure 3). Once estimated the $N_{V}$ threshold to define HD areas, an HD mask was created, and those tracks totally or partially overlapping HD areas were removed (Figure 4).

Determining $TS(L)$ and $TS(f)$ relationships
The relation between TS and fork length ($L$; cm) is normally assumed to be (Simmonds and MacLennan, 2005):

$$TS = a \cdot \log(L) + b,$$

(3)

In our case, it was modelled as:

$$TS = 20 \cdot \log(L) + b_{20} + \epsilon,$$

(4)

i.e. considering a fixed slope of 20 because of the small number of sets (two) and adding an error term $\epsilon$ to account for the natural variability of the TS. For each frequency, $b_{20}$ was estimated by fitting the observed TS distributions of in situ bigeye and the predicted TS distributions based on the measured $L$ from the purse-seine catches, using a curve fitting method similar to that of MacLennan and Menz (1996) or Gastauer et al. (2017).

The bigeye tuna body length distributions were converted to predicted TS distributions using Equation (4; Figure 5). For the error term, two alternative curve types were used: a simple normal function, $\epsilon = N(\mu = 0, SD)$, and a twofolded (skewed) normal, $\epsilon = N_{1}(\mu = 0, SD, s)$, included to increase the accuracy of the estimated $b_{20}$ values in cases of asymmetric observed TS distributions. An optimization process was run for the parameters of Equation (4) using sequences of $b_{20}$ (from $-80$ to $0$ in intervals of $0.1$ dB), standard deviation ($SD$; from $0$ to $20$ in intervals of $0.1$ dB) and, in the case of the twofolded normal, percentage of skewness, $s$, (from $-100\%$ -left sided- to $100\%$ -right sided- in intervals of $5\%$), being the resulting functions fitted to the observed TS distributions. For each curve type, all the combinations of parameters were computed and the one with the highest coefficient of determination was chosen. Then, the choice of curve type was based on AIC (Akaike Information Criterion, Akaike, 1973) to allow penalization for the extra parameter of the twofolded Gaussian. The curves obtained with this optimization procedure were the proposed predicted TS distributions. Standard deviations, confidence intervals of the TS distributions (Cumming et al., 2007), and coefficient of determination values of the fit between observed and predicted TS distributions were calculated to evaluate the goodness of the obtained $TS(L)$ relationships.

The TS($f$) relationship was calculated as the succession of $b_{20}$ values at the three available frequencies, for all the sets together as well as per individual set. This was done to assess the potential incidence in cases of relatively low predominance of bigeye tuna, because of the relatively weak condition to consider a set monospecific (90% in weight).

Distribution of TS values along the echogram
To test the validity of our analysis in the difficult observation environment of the FADs (concerning mainly potential incidence of bycatch and the efficiency of HD filtering) we checked the spatial distribution patterns of the data. We plotted the distribution of TS values against time and depth, to show whether they distributed randomly in the echograms or were subjected to any spatial distribution patterns, a skewed or stratified distribution potentially indicating some kind of selection.

Results
Size distribution
From the 20 sets done in the survey, only two (sets 6 and 7) had more than 90% in mass of bigeye tuna (i.e. 79%, Table 2) and were hence used for this study. The range of average bigeye tuna sizes (10 and 90% quantiles) was 57–90 and 56–89 cm, respectively for sets 6 and 7 (Table 3). In each set, the length
TS values by based on departure from monotonous increase of TV for lower frequencies, especially the 38 kHz, where the plankton tions into monomodal ones (Figure 6). The correction was higher the mean value in more than 5 dB and changing the sumably plankton and/or micronekton) were removed, increasing but after applying the school masking, the lower TS values (0.1 and 0.9 quantiles) observed after all the filtering steps were (light-grey circles) were removed from the analysis. Here and in Figure 4, the Nv estimates were obtained using the HD-corrected b20 values. This example corresponds to the 38 kHz. distributions were bimodal with each mode centred in ~62 and ~89 cm (Table 3, Figure 2).

TS filtering steps
The initial TS distributions had multiple modes at all frequencies but after applying the school masking, the lower TS modes (presumably plankton and/or micronekton) were removed, increasing the mean value in more than 5 dB and changing the TS distributions into monomodal ones (Figure 6). The correction was higher for lower frequencies, especially the 38 kHz, where the plankton layers were denser (Figure 1). The fish tracking step did not reduce further the mean TS value, but it decreased variability of the TS values by ~2 dB. The HD filtering threshold was set at 30 fish, based on departure from monotonous increase of TV against Nv (Figure 3) and decreased the mean TS by ~2 dB at all frequencies. Overall, the TS filtering steps reduced the number of single targets by more than one order of magnitude at all frequencies. The ranges of TS values (0.1 and 0.9 quantiles) observed after all the filtering procedures were (−39 dB, −24.5 dB), (−37.5 dB, −25 dB) and (−39.5 dB, −29 dB) for 38, 120, and 200 kHz, respectively (Table 3).

TS-L and TS-f relationships
The fit between modelled and observed TS distributions had coefficients of determination well over 80% (Table 3; Figure 6). The fitted TS-length relationship in Equation (1) had intercepts of −65, −66, and −72 dB respectively at 38, 120, and 200 kHz, with uncertainties of ~5.5, 4.5, and 4 dB. The frequency response was flat or slightly decreasing between 38 and 120 kHz and declined over 6 dB at the highest frequency 200 kHz (Figure 8). When the two sets were considered individually, set 7 yielded results ~1 dB higher at all frequencies and virtually the same relative frequency response as set 6.

Distribution of TS values along the echogram
The TS vs. time scatterplot showed that the mean TS value did not show trends along time (Figure 9). The TS vs. depth showed that the TS values were spread over the full depth range of the aggregation (Figure 10). The HD filtering removed the highest TS values, but there was no selection pattern according to depth. The filter was able to remove those areas with higher density while leaving unaltered nearby areas with lesser number of individuals (Figures 4 and 10).

Discussion
This work presents TS(L) and TS(f) relationships for bigeye tuna associated with FADs in a scientific study conducted from a fishing vessel during its regular commercial activity. There are both advantages and disadvantages in performing in situ measurements of TS following this procedure. The option of working from a fishing vessel during commercial activity facilitates the encounter with FADs and represents a cost-effective way to study these highly migratory species that can be found spread over huge oceanic areas of all oceans. The estimation of TS is done in situ, meaning that it provides measurements in the same conditions in which we intend to estimate their abundance. According to Simmonds and MacLennan (2005), there are three main potential problems when measuring TS of individual fish in situ (i) representativity of biological samples, (ii) representativity of acoustic targets, and (iii) discrimination between single and multiple acoustic targets.

Representativity of biological samples
In this work we benefitted from the high degree of certainty of the species and size composition of the measured targets, given that the aggregation is normally completely caught after the acoustic measurements (Boyra et al., 2018). There were only two sets where there were predominant catches of bigeye tuna, and there were still appreciable amounts of skipjack tuna in set 7 (Figure 2 and Table 2). Nevertheless, the relative frequency response was the same in both sets despite the larger proportion of skipjack in set 7 (Figure 7), thus suggesting that the incidence of skipjack in the measured frequency response of bigeye tuna was not appreciable. In our opinion, given the consistency between results of the two sets, both are reliable and should be considered for the final estimation of this analysis and this is what we propose in Table 3. Nevertheless, results from both independent sets are also provided, and the reader can compare them.

One of the challenges we faced is the wide length distribution of tuna found typically at each FAD, which causes dispersion in the observed TS distributions (Figure 7, Table 3) and, hence, uncertainty in the b20 estimation. We deal with this limitation by applying a curve fitting method to compare the observed and predicted TS distributions (as in MacLennan and Menz, 1996; Gastauer et al., 2017), a method in which all size and TS classes contribute to the fitting, in contrast to other methods that fit central values (i.e. the mean length against the averaged TS value in the linear domain, as in Clay and Castonguay, 1996; Ona, 2003; Madirolas et al., 2016 among many other examples). The application of the curve fitting method allowed providing measurements of goodness of the adjustment (R2 over 80% in all cases, higher at all frequencies and virtually the same relative frequency response as set 6. Distribution of TS values along the echogram
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We also provided confidence intervals for the estimated values (Table 3).

In this work, the fish length distributions were not only wide but also bimodal for bigeye (Figure 2), whereas the observed TS distributions were monomodal (Figure 7). There are two possible explanations for this: (i) the two length classes were fused into a single mode of observed TS values because of the natural variability of the TS or (ii) only one of the two length classes (typically the large one, supposed to have more chances to pass the single target detection criteria) contributed to the distribution of observed TS. In this case, we think that the first explanation is the most probable (as we justify below) and this work assumes it is true.

For the first explanation to be true, the natural variability of the observed TS must be high enough to dissolve the separations between individual length modes, which requires these distances

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**Figure 4.** Example of application of the high-density (HD) filtering on the 38 kHz echogram in set 6. About 40 min of data are shown. For each depth and ping number, the following items are represented: (a) $TV$, the number of detected single targets per cell; (b) $NV$, the estimated number of fish per cell; (c) categorization of cells according to the HD condition (true or false); and (d) categorization of fish tracks (good = false and bad = true) according to the HD condition. In (d), each circle represents the centre of a track.
to be less than twice the minimal standard deviation of the individual distributions (Behoodian, 1970; as we illustrate with the following interactive simulation [https://aztiapps.shinyapps.io/overlapping_distributions/]). In our case, the standard deviation of all the observed TS distributions (5.5, 4.5, and 4 dB for 38, 120, and 200 kHz, respectively; Table 3) were higher than both the directly converted TS values based on Equation (1; \( \sim 1.6 \) dB, Figure 5) and the distance between individual modes of the directly converted predicted TS values (\( \sim 3.5 \) dB, Figure 2). This suggests that the first explanation is at least possible, and consistent with our observations. The larger variability of the observed TS values with respect to the directly converted ones agrees also with the known large variability of TS of even a single fish because of behaviour (Foote, 1980; Simmonds and MacLennan, 2005).

The alternative explanation (the contribution of only one of the two length modes to the observed TS distribution) was the option taken by MacLennan and Menz (1996) in their earlier work, where they found another case of a bimodal length distribution leading to a monomodal TS distribution. However, there are considerable differences between their work and the present study. In their case they were measuring rather small fish (\( \sim 4 \) and \( \sim 16 \) cm length modes, respectively), with more than threefold (i.e. 300\%) difference in size between both species, whereas we are dealing with much larger individuals (\( \sim 62 \) and \( \sim 89 \) cm) and a much smaller difference (about 30\%) between them. In addition, they were using a transducer with a rather wide beam angle (6.5° \( \times 17^\circ \)), i.e. sampling larger volumes than in our case (7\° \( \times 7^\circ \)). Therefore, in their case it made sense ignoring the lowest mode because it would have a much lower probability of passing the single target detection algorithm. In our case, the assumption of equal probability of detection is much safer than removing the lower mode of \( \sim 50\% \) of tuna individuals of 62 cm; the alternative assumption would be considerably stronger and thus, in our opinion, less likely to be correct.

### Acoustic single targets’ discrimination and representativity

As not only tuna but also other non-tuna fish species (bycatch) as well as plankton and micronekton can be found at FADs (Bertrand et al., 1999a), a filter was applied to try to isolate tuna targets. The bycatch filter was based on observations done in acoustic tagging of non-tuna species at FADs (Muir et al., 2012), as has been applied by other authors (e.g. Lopez et al., 2016). The plankton or micronekton filter worked based on the aggregation structure of the echoes, removing those originated in scattering layers (Figure 1). This filter was the same as the plankton filter applied by Boyra et al. (2018) for skipjack tuna aggregations at FADs, and it seemed to be effective also in this case as it was able to remove the lowest modes of the TS distribution (Figure 5).

![Table 3. Summary of the results of the least square adjustment procedure.](https://academic.oup.com/icesjms/article-lookup/76/7/2446/5537349)

| Set | \( f \) (kHz) | \( L_{10} \) (cm) | \( L_{90} \) (cm) | \( TS_{10} \) (dB) | \( TS_{90} \) (dB) | \( N \) | \( b_{10} \) (dB) | \( SD \) (dB) | \( CI \) (dB) | \( R^2 \) (%) |
|-----|---------------|-----------------|-----------------|-----------------|-----------------|------|------------|-----------|-----------|-----------|
| 6   | 38            | 57              | 90              | -39.5           | -24             | 2,760| 55         | -         | -         | 91        |
| 7   | 38            | 56              | 89              | -38             | -25             | 2,922| 64.5       | 5.5       | -         | 85        |
| All | 38            | 56              | 90              | -39             | -24.5           | 5,682| 65         | 5.5       | -65.1     | 64.9      |
| 6   | 120           | 57              | 90              | -38             | -25             | 3,315| 45         | -         | -         | 92        |
| 7   | 120           | 56              | 89              | -36.5           | -25.5           | 3,451| 65.5       | 4.5       | -         | 86        |
| All | 120           | 56              | 90              | -37.5           | -25             | 6,766| 66         | 4.5       | -66.1     | 65.9      |
| 6   | 200           | 57              | 90              | -40             | -29             | 3,357| 73         | 4         | -         | 91        |
| 7   | 200           | 56              | 89              | -39             | -29             | 3,506| 71.5       | 4         | -         | 88        |
| All | 200           | 56              | 90              | -39.5           | -29             | 6,863| 72         | 4         | -72.1     | 71.9      |

*“f” stands for acoustic frequency, “L” for fork length, “TS” for target strength quantiles (the sub-indices 10 and 90 for the corresponding probabilities in %), “N” for number of single targets, “\( b_{10} \)” for the intercept in the TS-length relation in Equation (4), “SD” for standard deviation, “CI” for 95% confidence interval, and “\( R^2 \)” for adjusted coefficient of determination. In “All” we present the quantiles of the mixture distribution for \( L \) and TS; for \( b_{10} \) we present the lineazered average of the two values in the table.*
Perhaps the biggest challenge in this work was the high-density conditions because of the aggregative nature of tuna at FADs. It is a normal practice in TS analysis to avoid areas of high density whenever possible to avoid the risk of unresolved multiple targets (Soule et al., 1995, 1996; Barange et al., 1996; Ona, 1999). So perhaps one could think that, instead of focusing on the main aggregation, it would have been safer to remove the main aggregation and focus at its periphery. The problem is that it has been repeatedly reported that the periphery of FAD aggregations tends to be unrepresentative of the main catches: the upper part of the aggregation is frequently made of bycatch species (Muir et al., 2012; Forget et al., 2015) whereas at the bottom part are concentrated large individuals of yellowfin and bigeye that are not representative of the main tuna sizes found associated with FADs (as described by Moreno et al., 2008; Govinden et al., 2010; Muir et al., 2012; Lopez et al., 2016). The possibility of un-representativeness of the periphery of an aggregation was also pointed out by Simmonds and MacLennan (2005). Consequently, as most of the regular-sized tuna are located in the bulk of the aggregation, we focused our analysis in this area.

In order to avoid potential unresolved multiple echoes bias, a combination of two filters were used: a fish tracking (Blackman, 1986) plus a high-density filtering (Sawada et al., 1993; Gauthier and Rose, 2001). The HD filtering method, rather than discarding the full aggregation, allowed working on it while providing an objective and repeatable way of discarding selectively the parts with higher probability of multiple targets (Figure 4). The additional application of the fish tracking in the lowest density cells allowed obtaining the associated benefits of this technique (namely reducing the variability) while avoiding the risk of appearance of spurious tracks because of excessive proximity between targets. In an early version of this work, a different approach was applied to deal with potential multiple target bias, a bi-frequency simultaneity detection requirement (Conti et al., 2005). This was the same approach as that followed by Boyra et al. (2018) for skipjack tuna associated with FADs. The results with this early approach were
very similar ($b_{20} = -64, -65.5, \text{ and } -72 \text{ dB at } 38, 120, \text{ and } 200 \text{ kHz, respectively, i.e. within } \sim 1 \text{ dB at all frequencies}) to those presented here, but finally we opted by the new approach, combining HD and fish tracking, because of concerns related to potential effect of the depth on the efficiency of the bi-frequency method because of higher directivity of higher frequencies. Also, the fish tracking used in the new approach allowed reducing stochasticity. Similar approaches as this, combining various methods for mitigating multiple targets bias, have been recently applied in TS analysis (Scoulding et al., 2016; Gastauer et al., 2017). The observed TS distributions show no patterns with depth (Figure 10), which is consistent with the expected TS response for a phycologist species as bigeye tuna (Magnuson, 1973; Bertrand et al., 1999b), able to compensate the pressure induced changes in swimbladder volume to maintain buoyancy at different depths.

**Obtained TS values and TS-length relations**

The obtained $b_{20}$ values were $-65, -66, \text{ and } -72 \text{ dB at } 38, 120, \text{ and } 200 \text{ kHz (Table 3, Figure 8). Oshima (2008) obtained a 1.5 dB higher value ($b_{20} = -63.5 \text{ dB}$) measuring isolated
individuals of bigeye tuna in a cage at 38 kHz. On the other hand, based on two previous experiments (Bertrand et al., 1999b; Josse and Bertrand, 2000), Bertrand and Josse (2000) modelled $TS = 24.3\log(L) - 73.3$ for a range of bigeye fork lengths from 50 to 130 cm at the same frequency. Forcing the slope of that relationship, for comparability, to the value of 20 and using the intermediate length of the range, 90 cm, it would represent a $b_{20}$ of $-64.7$ dB, thus very similar to the value obtained in this study. Therefore, our result is in accordance with previously measured $TS$ values at 38 kHz.

Because the volume within the purse-seine net decreases with time along the evolution of the net hauling manoeuvre, the density of the fish aggregation increases correspondingly. During our acoustic measurements in the purse-seine net, there was concern about whether this might have an impact on the measured $TS$ values. However, the analysis of the evolution of the filtered $TS$ values with time showed no clear pattern in this respect (Figure 9) thus indicating similar efficiency of the multiple targets filtering procedure along the duration of the set.

It is recommended that, whenever possible, $TS$-length relationships are measured without forcing the slope to 20 (Mcclatchie et al., 2003). In our case we had enough $TS$ values (>5000 detections at each frequency, Figure 6) to correctly estimate a $b_{20}$ in all cases. However, the availability of only two sets to make the regression between $TS$ and $L$ prevented us from trying to estimate the slope from these data (two points are clearly not enough to make a linear regression model). It is thus desirable that further work is done to try to establish the $TS$-length of this species without forcing the slope to 20.

In this work, we also obtained $TS$ and $b_{20}$ values at 120 and 200 kHz, resulting in a response that decreases with frequency (Table 3, Figure 6), which is typical of swimbladder bearing species (e.g. Fernandes et al., 2006). The fact that autumn from 120 to 200 kHz is steeper than that predicted by the typical models for swimbladdered species is not understood yet, but the discrepancy could be caused by target size/distance constraints of most models (Medwin and Clay, 1998), that are normally focused on small pelagic species and might thus not be valid for large fish sizes as those of tuna. In the near future, we intend to explore the

![Figure 8](https://example.com/image8.png)

*Figure 8.* $TS$-frequency response of bigeye tuna (black), triangles representing set 6 and circles set seven. In grey squares, $TS$-frequency response of skipjack tuna, taken from Boyra et al. (2018). Error bars represent standard deviation.

![Figure 9](https://example.com/image9.png)

*Figure 9.* Evolution of mean $TS$ values per track with time (ping number) along the set duration. The lines represent loess smoothing to highlight the tendencies.
interpretation of these results by application of modelling using the method of Fundamental Solutions, that has been successfully applied to interpret TS measurements of other large pelagic species (Pérez-Arjona et al., 2018). Given that the trend in the frequency response obtained for bigeye is opposite to that of skipjack tuna (Figure 7) this provides the means to discriminate between these two species using a frequency response mask (e.g. Korneliussen and Ona, 2003).

This work is part of a series of initiatives to achieve species and sizes discrimination of tropical tuna species associated with FADs (Moreno et al., 2019). As part of this initiative, TS-length and TS-frequency relationships for skipjack tuna around FADs have been recently published (Boyra et al., 2018). Most FADs normally have simultaneous presence of the three main tropical tuna species, i.e. bigeye, skipjack, and yellowfin, therefore, the next step would comprise obtaining TS-f and TS-L of the third one, i.e. yellowfin tuna (and, if necessary, of other potentially abundant bycatch species). Currently, there are only published TS-L values for yellowfin tuna at 38 kHz (Bertrand et al., 1999b) and therefore, next step should be obtaining the TS-frequency and TS-length for yellowfin tuna, to build proper acoustic masks to try to distinguish between the three main tropical tuna species. Given the difficulty of finding isolated or even predominant yellowfin tuna aggregations at FADs, the planned strategy with yellowfin will likely involve ex situ measurements in a cage.

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