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Horizontal target strength-size conversion equations for sea bass and gilt-head bream

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ABSTRACT

Horizontal hydroacoustics is a technique of remote fish detection that has proved to efficiently determine fish density and biomass in shallow or superficial waters. This non-intrusive technique could render better results than those obtained by conventional sampling when monitoring shallow waters. To apply this technique, we need equations that relate the amount of sound returned by fish, known as target strength (TS in dB), with their length (mm) or weight (g). This study presents horizontal conversion equations for the species gilt-head bream (*Sparus aurata*) and sea bass (*Dicentrarchus labrax*), two of the most important species bred in aquaculture. Moreover, this study presents a new formula to calculate a mixed equation adjusted to the percentage of distribution of those species in case of mixed populations. The results in this research have been obtained by applying a simple setup that allows for high-quality acoustic data. These new equations will enable the application of this technique in order to estimate these species' density and biomass in shallow aquatic systems such as aquaculture ponds with semi-intensive production densities.

1. Introduction

Nowadays, aquaculture as a whole produces more than half of all fish consumed in the world. In southern European countries, gilt-head bream (*Sparus aurata*) and sea bass (*Dicentrarchus labrax*) are the most produced and consumed species (Apromar, 2016).

Quantity and biomass estimations of fish in ponds are essential for preparing the production and management plan of aquaculture companies. For example, feeding constitutes >50% of production costs (Soliveres, 2015). This feeding estimation is based on the biomass of the ponds together with other factors, such as average fish size, time of year, water temperature, etc. The accuracy of these estimates allows for an optimization of the feeding processes and minimizes waste products, which improves the quality of water. In addition, the use of techniques that provide us with accurate estimates improves population management plans. This includes splitting the ponds, i.e. distributing the fish group from one pond into two different ponds in order to facilitate their growth and development as well as population monitoring.

Nowadays, in order to estimate fish biomass in shallow aquaculture ponds, managers conduct periodic samplings that usually give wrong estimates. They calculate the number of fish present in a pond as an approximation that takes into account the initial number of sowed fish and countable deaths. Biomass is estimated by multiplying this number by the average weight of fish captured by manual methods (Conti et al., 2006). In addition to not being accurate enough, this type of manual sampling causes a high level of stress in fish, which sometimes leads to pathologies and sudden deaths. Furthermore, these methods can monitor fish in normal production conditions, but they are not valid in exceptional situations where losses cannot be quantified, for example in cases of theft, predators or extensive deaths. In these cases, traditional methods cannot determine the quantity of fish remaining in the pond.

The results presented in this study belong to an extended project aimed at estimating biomass in fish production systems using non-invasive methods (PI_57052). Given that hydroacoustics is one of the most relevant existing capture-independent methods, applying it to shallow ponds would considerably improve fish's wellbeing. Unlike visual methods, hydroacoustics is also efficient when used in systems with poor visibility (Lucas and Baras, 2000; Simmonds and Maclennan, 2005; Kubečka et al., 2009) such as production ponds. However, hydroacoustics does not work properly in cases of very high densities. Therefore, it could only be used in production ponds with extensive or semi-intensive farming. Another advantage of hydroacoustics is that it studies the aquatic system at the exact moment when sampling is being performed, taking an instant picture of the situation in the pond. Therefore, it would be useful even in the exceptional cases previously mentioned.

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Horizontal hydroacoustics is a technique where sound is used to study aquatic ecosystems. Acoustic instruments (the echosounder) transmit and receive sound waves used to detect fish and other objects beyond the water surface (Simmonds and MacLennan, 2005). The key to properly interpreting hydroacoustic data is the target strength value returned by fish (TS in dB re 1m^2). This value allows one to create conversion equations to translate the sound returned by fish (TS) into biological parameters that can be measured, such as length or weight. The use of these equations allows calculating the size and quantity of fish in an ecosystem (Lucas and Baras, 2000). Moreover, in horizontal applications, it has been proven that changes in TS are related to changes in the swimming direction of fish. Therefore, horizontal conversion equations need to be developed for the three main orientations: side, oblique and head/tail (Foote, 1980; Kubecka and Duncan, 1998; Hazen and Horne, 2003; Frouzova et al., 2005; Henderson et al., 2007; Kubilius and Ona, 2012; Rodríguez-Sánchez et al., 2015).

Despite the fact that some conversion equations already exist for several fish species (Burwen and Fleischman, 1998; Lilja et al., 2000; Frouzova et al., 2005; Boswell and Wilson, 2008; among others), these are not sufficient to meet the needs of hydroacoustics. General equations could be used to convert acoustic data. However, given that TS values are species-dependent, the results will be more accurate if species-specific equations are used when converting data. Consequently, this article is aimed at developing horizontal conversion equations for gilt-head bream (*Sparus aurata*) and European sea bass (*Dicentrarchus labrax*). Sometimes, ponds are composed by a mixture of these two species with known proportions. To calculate density and biomass in these situations, the equation of the species with a highest representation in the system is normally used. This paper also presents a simple method to create a mixed equation adapted to the percentage of these

two species when surveying mixed ponds. All these equations will allow the application of this technique for routine estimations of density and biomass in shallow waters such as aquaculture ponds.

2. Material and methods

2.1. Experimental set-up

The experimental design to obtain the equations was conducted in the facilities of the company Pesquerías Isla Mayor S.A. (PIMSA, Spain), specifically in one of the tanks of the fattening room in its hatchery facilities. The experimental tank was a rectangular pool ($2 \times 8 \times 2$ m). A calibration test was performed before the start of the experiment in order to ensure that there were no significant echo returns coming from the pool. The test entailed performing a horizontal calibration and collecting recordings from the calibration sphere, which was a 13.7 mm copper sphere (TS = -45 dB) placed at seven different distances with respect to the transducer (meter by meter, from 1 to 7 m away from the transducer). The mean deviation in the expected TS was lower than 0.6 dB at every distance.

TS data was recorded with a Simrad EK60 scientific echosounder (Simrad Kongsberg Maritime AS, Horten, Norway) equipped with a 200 kHz circular split-beam transducer (ES200-7C). The transducer was placed on a stainless-steel clamping frame on one side of the pool. It was aimed horizontally, parallel to the water surface (Fig.1). The acoustic axis was located 1 m from the surface. The acoustic unit was calibrated with the calibration copper sphere according to the standard calibration method (Simrad, 2004). Echosounder settings are listed in Table 1.
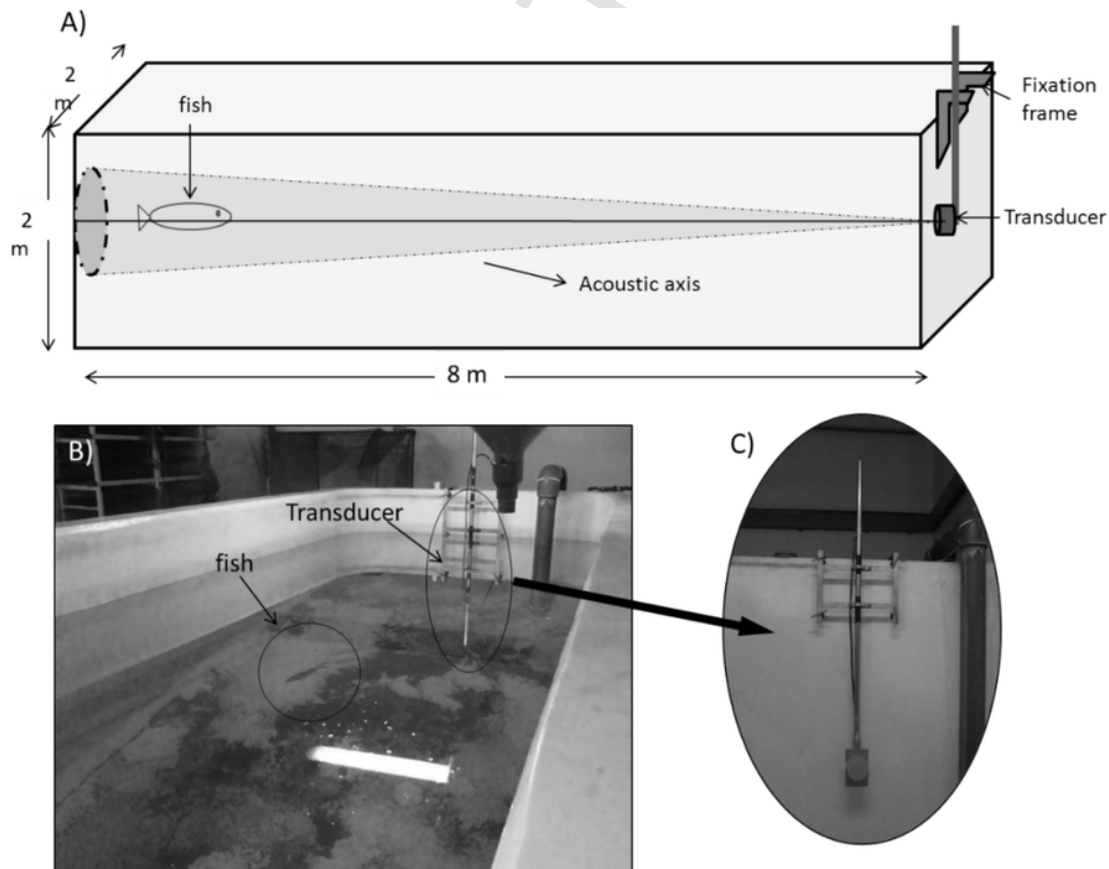

Fig. 1. A) Diagram of the experiment. B) Photograph of the experiment C) Photograph of the tranducer's frame.

Table 1

Settings of the echosounder used to measure horizontal target strength (TS).

Parameters	
Type of transducer	Simrad ES200-7C
Transducer shape	Circular
Transmission frequency (kHz)	200
Transmitting power (W)	150
Pulse length (ms)	0.128
Ping rate (ping•s^{-1})	10
Minimum threshold (dB)	−70

The sound records of a total of 15 individual sea bass and 15 individual gilt-head bream of different sizes. All selected fish were measured (mm) in total, fork and standard length and weighed (g) before being placed in the experimental pool. The different sizes were classified based on an artificial order depending on the weight range of the fish (Table 2).

Fish were individually placed in the tank to be insonified. Data was stored on a PC and later processed with the Sonar5 Prov.6.0.1 analysis software (Balk and Lindem, 2011). Raw data (.raw) was converted with the 40logR function. The echosounder's single echo parameters were set so that we obtained as many echoes as possible from the recorded fish. The minimum threshold was set to −60 dB; minimum and maximum echo durations were 0.80 ms and 1.6 ms (rel. to pulse lengths). The maximum gain compensation was −3 dB (one-way) and the maximum phase deviation was 5. TS values compensated for angular location in the beam (TS, dB re 1 m^2) were used for the analysis. Once the echograms were obtained, insonified fish's tracks were manually classified and registered. The near field of the largest fish was calculated following the equation $D = r^2/\lambda$, where r is the half length of the fish (SL) and λ is the length of the sound wave (Medwin and Clay, 1998). Tracks were stored at a distance of 3.5–7 m from the transducer,

avoiding the possible TS deviation caused by the effect of the near field of the fish and the transducer (Tichy et al., 2003; Dawson et al., 2000; Rodríguez-Sánchez et al., 2016). Only linear swimming tracks with at least five echoes were stored. Linear tracks are defined as the group of echoes drawn by the line described by a fish moving without changing its swimming direction (Fig. 2).

A total of 300 linear tracks were classified for each fish, i.e. 100 tracks for each main orientation. Following the methodology used in Rodríguez-Sánchez et al., 2015, the angles between 180° to 360° were converted to their specular angles, i.e. angles of 120° were equal to angles of 90°. Only the swimming tilt of fish was considered, not its swimming direction. For each fish, the selected tracks were divided into 18 categories of 10° according to their aspect angle (0–180°). Subsequently, the mean TS of each category was calculated and used for the creation of the equations. Three angular regions were established to describe swimming orientation: head and tail orientation (H&T) (with angles between 0 and 20° and 160–180°), oblique orientation (with angles between 20 and 70° and 110–160°) and lateral orientation (70–110°).

Fish were anaesthetised with clove oil (García-Gómez et al., 2002) and laterally X-rayed after the sound measurements. Since not all insonified fish preserved their swim bladders in optimal conditions, the radiographic information was completed with several not insonified individual fish which belonged to the same fish farm. A portable Diagnostic X-ray Unit, EcoRayOrange 1060 HP, was used to perform the radiographs at an intensity of 4 mAs and 55 kv during 1.2 s. the lateral area of the swim bladder (A) and the lateral area of the body (B) for each fish was calculated using the public domain Java image processing and analysis program IJ 1.46r (Rasband, 1997–2014) (Fig. 3). The lateral area of the fish was considered to be only from the mouth to the root of the tail (standard length) because the fish tail scatters sound very weakly (Kubilius and Ona, 2012).
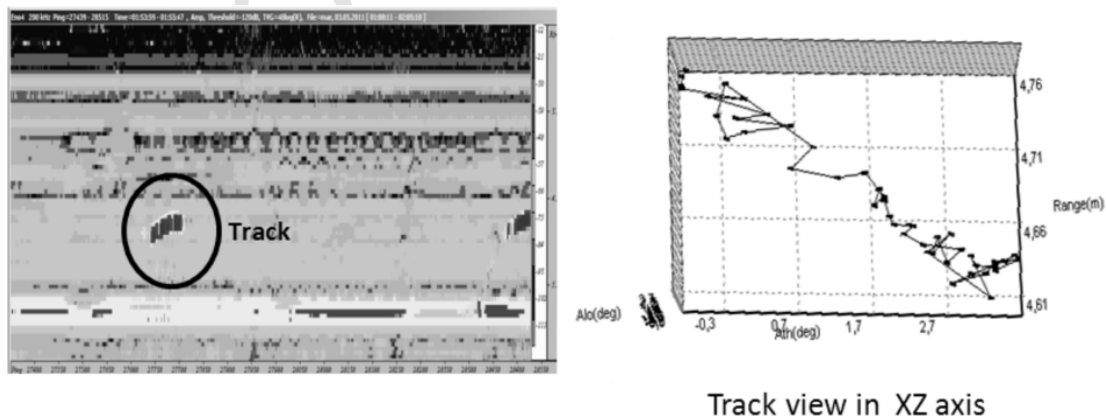
Table 2

Spaurus aurata and *Dicentrarchus labrax* size classification.

	Spaurus aurata						*Dicentrarchus labrax*					
Size class	Range (g)	SL	S.D.	W	S.D.	N	Range (g)	SL	S.D.	W	S.D.	N
1	0–60	127.5	2.8	50.0	10.7	3	0–50	137.6	6.0	49.3	8.3	3
2	100–150	161.0	5.3	112.0	2.1	3	50–120	168.0	3.6	90.6	1.1	3
3	150–200	172.3	8.7	171.5	6.5	3	120–200	195.0	5.0	150.3	0.5	3
4	500–700	255.0	7.1	573.0	63.3	3	600–750	340.0	10.0	699.3	37.1	3
5	700–900	286.6	10.4	778.0	75.9	3	750–950	371.6	2.8	906.0	25.1	3

SL Standard length (mm); W, weight (g); S.D., Standard deviation; N, number of individuals

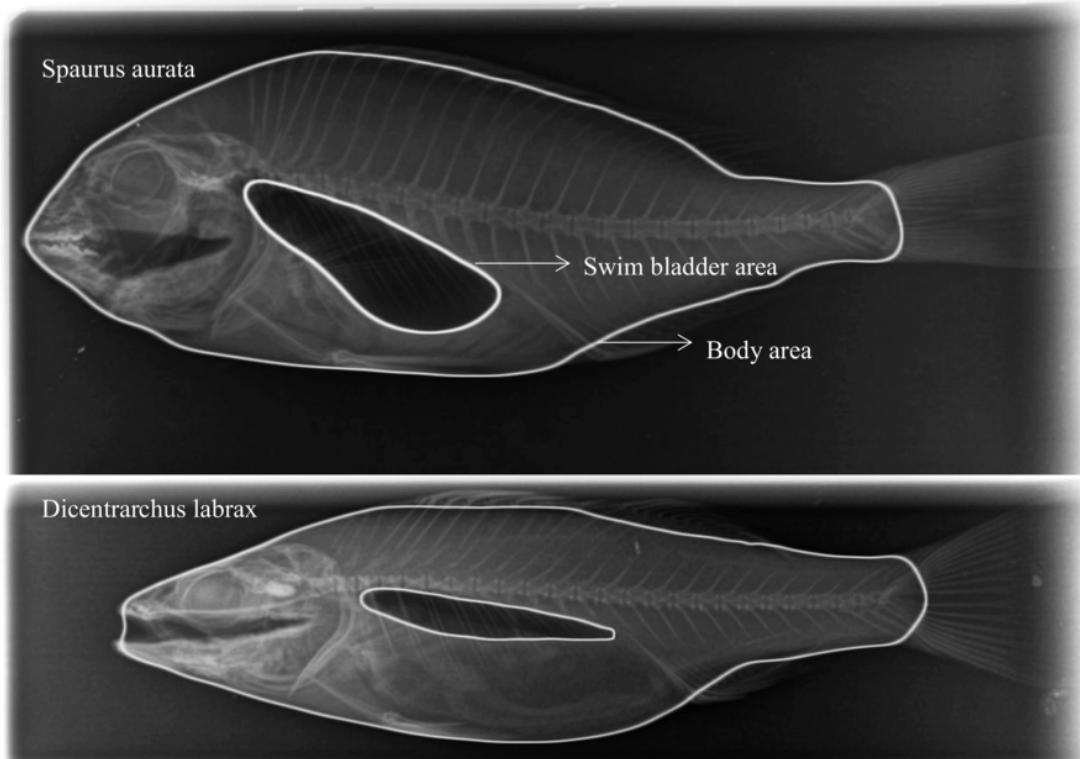
Track view in XZ axis

Fig. 2. Linear track.

Fig. 3. Radiographs of two individuals of *Spaurus aurata* and *Dicentrarchus labrax* (size class 3) used in this study. The swim bladder area and the body area are highlighted.

2.2. Statistical analysis

The acoustic data collected was statistically analyzed by SPSS 20.0. (IBM Corp., 2011). A Kolmogorov-Smirnov goodness-of-fit test was used to test the normality of the TS values by species, orientation and size classes. For each species and orientation, the mean TS values calculated for each 10°-category were regressed against the logarithm of the standard length and weight of the individual fish using the following relationship:

1) $Y = a \cdot LogX + b,$

where Y is the target strength (TS) in dB, X is the length or weight of the fish and a and b are regression constants.

Then, the specific equation for sea bass (species 1) and gilt-head bream (species 2) were calculated:

$$TS_1 = a_1 \cdot logX + b_1$$

$$TS_2 = a_2 \cdot logX + b_2$$

By definition:
$TS = 10 \cdot log\sigma$ and, $\sigma = 10^{TS/10}$.

Then, the TS mixed values (dB) were calculated following the next formula:

$$TS_{mixed} = TS_{\alpha_1, \alpha_2}$$
$$= 10 \cdot log \left[\alpha_1 \cdot 10^{\frac{TS_1}{10}} + \alpha_2 \cdot 10^{\frac{TS_2}{10}} \right] \quad ; \quad 0$$
$$\leq \alpha_i$$
$$\leq 1$$

where TS_1 and TS_2 are the TS values derived from the species-specific equations and α_1 and α_2 are the distribution percentages of each species in a system. In this study, the mixed equation was calculated considering that the species were equally distributed, i.e. 50% of fish were sea-bass and 50% of fish were gilt-head beam.

The calculated TS_{mixed} values were used to create the mixed equation using formula 1).

An analysis of variance (ANOVA) was conducted for each studied species in order to test the TS differences in relation to the individuals' lengths. In order to study the variation of TS depending on the orientation, a one-way analysis of covariance (ANCOVA, $\alpha = 0.05$) was conducted with the standard length as the covariate. In order to study the variation of the swim bladder area depending on the species and size class, a one-way analysis of covariance (ANCOVA, $\alpha = 0.05$) was conducted using the body area as the covariate.

3. Results

TS data for each species, orientations and size class followed a normal distribution (Kolmogorov-Smirnov test, $p > 0.05$). Fig. 4 shows the TS returned by sea bass of three size classes in each of the main orientations. As can be seen, TS values increase as fish become bigger. This TS-size correlation has proven to be a direct one both for sea bass (ANOVA, $F_{14,1325} = 23.230$; $p < 0.001$) and gilt-head bream (ANOVA, $F_{14,1298} = 10.019$; $p < 0.001$). Furthermore, we can see that TS values vary depending on the studied orientation. These variations occur both in sea bass (ANCOVA, $F_{2,1137} = 127.351$; $p < 0.001$) and gilt-head bream (ANCOVA, $F_{2,1253} = 69.061$; $p < 0.001$). In both species, head-tail orientations produce the lowest TS values (-46.81 dB, Std. E. 0.79 for sea bass and -43.63 dB, Std.E. 0.60 for gilt-head bream) and lateral orientations give the highest ones (-39.50 dB, Std.E. 0.95 for sea bass and -38.66 dB, Std.E. 1.93 for gilt-head bream).

Table 3 shows the results of the regressions of horizontal conversion TS-standard length and weight for each of the main orientations in sea
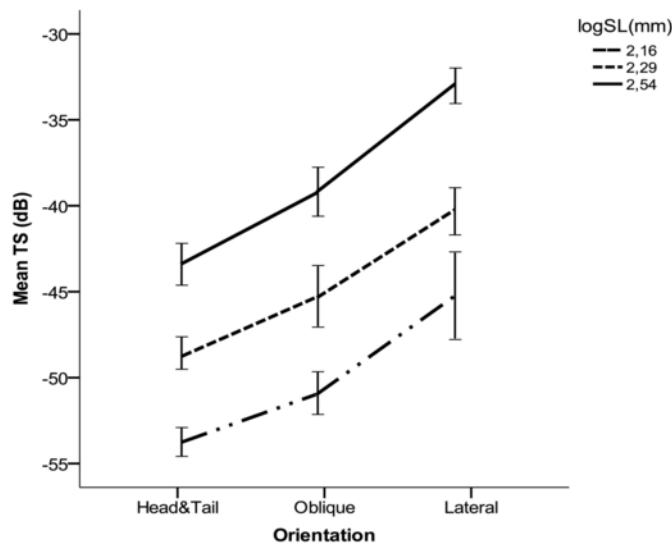
Fig. 4. Results of mean TS by orientation obtained after insonifying three sea bass of different sizes.

bass and gilt-head bream derived from acoustic data. Table 4 presents the results of the regressions of horizontal conversion TS-length and weight for the pool of species sea bass-gilt-head bream mixed at a ratio of 50% derived from the mixed equation. All regressions are meaningful ($p < 0,001$) and present a positive correlation between the acoustic signal increase and fish length. The equations obtained present a good adjustment for all studied orientations.

Fig. 5 represents TS-length lateral conversion equations for sea bass (solid line), gilt-head bream (dashed line) and the calculated mixed equation at a ratio of 50% (dashed grey line). As can be seen, the length estimated from each equation is different even though the TS is the same. If the conversion equation for sea bass is used, a TS value of -35 dB corresponds to a sea bass of 275 mm. Contrastingly, if the conversion equation for gilt-head bream is used, the same TS value of -35 dB corresponds to a gilt-head bream of 250 mm. The mixed equation at 50% falls in the middle of those two. Besides that, we can see that the equations of sea bass and gilt-head bream do not present the same slopes and the regressions intersect at one point. In lengths smaller than 225 mm, gilt-head breams present a weaker TS than that of the sea bass. In longer lengths (corresponding to a TS of -38 dB), gilt-head breams present higher TS values.

Table 5 presents the measurements of the lateral swim bladder area and the lateral body area calculated from the radiographs conducted on the studied species. In both species, the area occupied by the gaseous swim bladder becomes more and more important as the size of

the fish increases. The results of the ANCOVA performed on the swim bladder area data using the body area as the covariate prove the existence of a significant interaction between the species and size class factors ($F_{4,35} = 9.304$; $p < 0.001$). The lateral swim bladder area was different between the studied species for the size class 2 and 5 ($F_{1,35} = 12.268$; $p < 0.001$ for size class 2 and $F_{1,35} = 34.723$; $p < 0.001$ for size class 5). In both cases, the swim bladder areas of gilt-head bream were bigger than these calculated for sea bass. No significant differences were found for the rest of size classes.

4. Discussion

Numerous studies have proved that the TS returned by fish is directly related to their length and weight. This allows us to use hydroacoustic devices to discover the number of fish and estimate their size and biomass in the studied aquatic systems (Love, 1977; Kubecka and Duncan, 1998; Lilja et al., 2000, Simmonds and MacLennan, 2005; among others). However, establishing TS-length or TS-weight conversion equations for the studied species is not easy. Having an appropriate and simple system to obtain data would greatly ease this task. In this sense, the setup conducted in this experiment is recommended because, on the one hand, it simplifies data acquisition and, on the other hand, the data quality is high enough to create conversion equations.

Previous studies where horizontal hydroacoustic techniques were applied have proved that TS varies depending on the fish's swimming orientation. These studies demonstrate that it is necessary to establish conversion equations where each one of the main orientations is separately considered (Ona, 1990; Simmonds and MacLennan, 2005; Jech, 2011). In our study, we can see that TS variations based on orientation are indeed meaningful. In order to obtain accurate results in TS-length or weight conversions, equations must be conducted for each one of the main orientations as the more accurate we are when converting TS to length or weight, the better quality our results will have.

Fig. 5 shows lateral conversion equations for each studied species, as it is shown length values vary depending on the conversion equation that is used even if the TS is the same. Sea bass' and gilt-head bream's regressions present a behavior adjusted to the typical growth of each species. Sea bass' growth tends to be isometric during its whole life. However, although gilt-head bream presents a similar growth at the beginning of its life, it presents a more allometric growth once it reaches a certain length. These differences in growth are reflected in the variations that we find in the slopes of the regression models for both species. Furthermore, these differences in the slopes of the line presented by both studied species also match the results of the lateral area occupied by the swim bladder in relation to the lateral body area of the fish. Previous studies highlight the important role that the swim bladder plays in the fish's response to sound. It is estimated that 95% of a fish's response to sound depends on its swim bladder (Foote, 1980;

Table 3

Horizontal linear regressions of target strength (TS, dB) vs. standard length (SL, mm) and weight (W, g) for *Dicentrarchus labrax* and *Spaurus aurata* created from the fish insonified with a split-beam systems operating at 200 kHz.

	Orientation	Variable	a	Std.E.	b	Std.E.	R^2	p
Dicentrarchus labrax	H/T	SL	26.96	2.12	-111.42	5.05	0.834	<0.001
	Oblique	SL	29.22	2.08	-111.69	4.9	0.719	<0.001
	Lateral	SL	27.10	1.49	-101.23	3.61	0.926	<0.001
	H/T	W	8.83	0.65	-68.33	1.57	0.851	<0.001
	Oblique	W	9.93	0.68	-65.94	1.66	0.734	<0.001
	Lateral	W	9.27	0.49	-58.98	1.23	0.932	<0.001
Spaurus aurata	H/T	SL	43.97	6.24	-152.85	14.90	0.756	<0.001
	Oblique	SL	42.91	3.80	-143.31	8.86	0.659	<0.001
	Lateral	SL	46.56	3.36	-146.67	7.95	0.936	<0.001
	H/T	W	15.51	2.12	-89.75	5.74	0.769	<0.001
	Oblique	W	12.78	0.93	-74.94	2.37	0.754	<0.001
	Lateral	W	15.02	0.73	-75.83	1.93	0.970	<0.001

Table 4

Horizontal linear regressions of target strength (TS, dB) vs. standard length (SL, mm) and weight (W, g) derived from the species-specific conversion equations created for a pool of sea bass and gilthead bream mixed at a ratio of 50%.

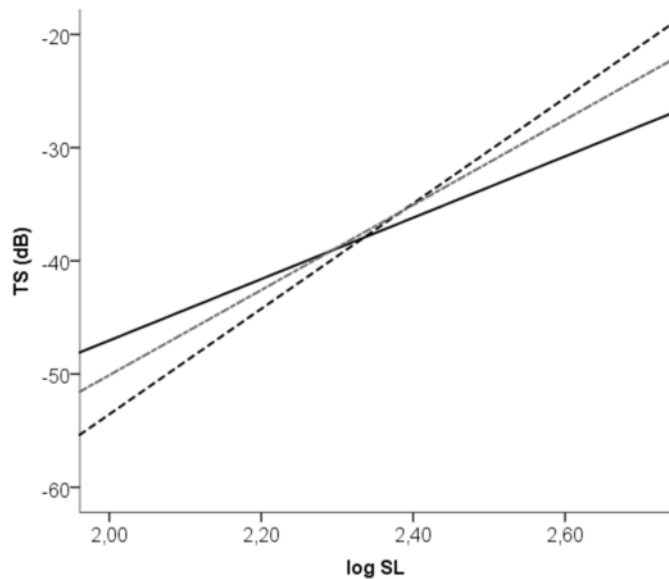|	Orientation	Variable	*a*	*b*
Mixed equation	H/T	SL	32.96	−125.63
	Oblique	SL	35.50	−125.75
	Lateral	SL	35.41	−119.82
	H/T	W	10.11	−73.26
	Oblique	W	10.94	−69.31
	Lateral	W	10.95	−64.22

Fig. 5. Lateral regressions TS–length for *Dicentrarchus labrax, Spaurus aurata* and mixed equation.

Table 5

Measurements of body area and swim bladder area (Sb) in cm^2 of the studied species classified by size class.

Size class	N	Body area	Std. error	Sb area	Std. error
Sea bass					
1	3	40,50	1,01	2,30	0,40
2	3	64,54	3,00	2,93	0,02
3	4	83,13	2,78	4,36	0,82
4	4	212,81	10,39	15,28	1,65
5	4	264,98	9,87	20,19	0,93
Gilt-head bream					
1	4	49,25	14,26	2,67	0,28
2	3	69,93	10,23	4,74	0,32
3	5	110,89	19,76	5,16	0,41
4	4	219,35	15,99	14,69	1,28
5	4	262,67	15,38	23,76	1,70

Hazen and Horne, 2003). If we have a look at the calculations of the area occupied by the swim bladder and the results of the comparison performed between species at different size classes, it becomes easier to understand this relation and how the changes in the swim bladder influence the fish's acoustic response. Taking this into account, we can see that the area occupied by the swim bladder for sea bass and gilt-head bream is similar at small sizes when their growth shows a similar behaviour. However, this changes from a particular size on, which leads to the intersection of the regression lines. Gilt-head bream is the

species which presents the highest TS for the larger size classes. In any case, the equations developed for these species are different, but these comparisons help us understand the nature of the TS in relation to the fish's size and the importance of the swim bladder in the acoustic response.

Likewise, we can see how the mixed equation, which considers that each species makes up for 50% of the total, falls somewhere in between both species' specific relationships. This mixed equation will vary depending on the percentage of each species in the system, i.e. it will be more similar to the sea bass one when this species makes up for >50% of the total and vice versa, and the differences will change the conversion results. Although the differences derived from using a conversion equation for species other than those studied might seem unimportant, these translate into relevant variations in biomass, particularly in species such as sea bass and gilt-head bream (an increase of 100 mm can be the equivalent of 300 g in sea bass and >500 in gilt-head bream). This proves that choosing the right TS-size conversion equation is crucial to accurately estimate fish's size and biomass (Boswell and Wilson, 2008; Boswell et al., 2008). Likewise, in mixed systems, applying a specific equation to the proportion of known species will undoubtedly improve the results of length and biomass derived from acoustic signals.

As proven by other previous studies (Kubecka and Duncan, 1998; Burwen and Fleischman, 1998; Frouzova et al., 2005; Lilja et al., 2000; Rodríguez-Sánchez et al., 2015), not only choosing the right equation can significantly influence the final result of the TS-size relation, but also choosing the right method to calculate the equation. This is particularly important in horizontal samplings because lateral detection implies a high loss of echoes coming from head/tail orientations and a higher variability in TS due to fish movement, etc. Therefore, a high-quality and specific conversion equation can compensate the defects produced in detections and also improve accuracy in biomass and density estimates.

Therefore, when performing hydroacoustic studies where fish's length and biomass estimations are important, the equations used must be specific to the studied species whenever possible. The TS-length relations developed in this study for *Dicentrarchus labrax* and *Spaurus aurata* using split-beam systems operating at 200 kHz present a good adjustment and will allow us to calculate biomass of fish production systems where fish are bred. In addition, this study presents an equation that allows us to create our own mixed equation for those situations where more than one species is present. Based on specific equations, we will be able to easily create our own mixed equation adjusted to the percentages corresponding to each species in the system. As shown in this article, the studied species present differences in their acoustic response, which directly translates into variations in the estimated biomass. Therefore, this new equation will certainly improve biomass estimates in ponds with more than one species.

Applying hydroacoustic techniques to monitor size and quantity of fish in shallow semi-intensive production aquaculture ponds will allow us to obtain accurate estimates of fish biomass. In fact, the equations presented in this article have been used in the previously mentioned project (PI_57052) to calculate fish biomass in farming ponds with sea bass and gilt-head breams. The surveys conducted in this project have produced high-quality results valid to study the biomass of these two species in inland aquaculture ponds with semi-intensive production densities. These results indicate that the equations created in this study can be used in routine control studies in fish production companies providing us with quality results. This creates huge expectations regarding the future application and use of hydroacoustic techniques in the field of inland aquaculture farming management.

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