How many fish? Comparison of two underwater visual sampling methods for monitoring fish communities (#23334)

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How many fish? Comparison of two underwater visual sampling methods for monitoring fish communities

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Background. Underwater visual surveys for monitoring fish communities are preferred than fishing surveys in certain habitats such as rocky or coral reefs and seagrass beds, and are the standard monitoring tool in many cases, especially in protected areas. However, despite their wide application there are potential biases mainly due to imperfect detectability and the responsive movement of fish.

Methods. The performance of two methods of underwater visual surveys were compared to test if they give similar results in terms of fish population density, occupancy, species richness and community composition. Distance sampling (line transects) and plot sampling (strip transects) were conducted at 31 rocky-reef sites in the Aegean Sea (Greece) through SCUBA diving.

Results. Line transects generated significantly higher values in terms of occupancy, species richness, and total fish density, compared to strip transects. For most species, density estimates differed significantly between the two sampling methods. Line transects yielded higher estimates for cryptic species and for those presenting avoidance behavior to the observer, as it accounted for imperfect detectability and utilized a larger survey area compared to the strip transect method. On the other hand, large-scale spatial patterns of species composition were similar for both methods.

Discussion. Overall, both methods presented a number of advantages and limitations, which should be considered in survey design. Line transects appear to be more suitable for surveying cryptic species, while strip transects should be preferred at high fish densities and for species of high mobility.

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Abstract

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1. Introduction

Over the last decades several sampling approaches have been developed for the assessment of fish communities in different marine habitat types. The choice of the most suitable sampling method is a crucial step during the survey design process. This decision is usually dictated by the overall research objectives, the level of accuracy needed to address scientific questions, the time and resource availability to carry out the survey, as well as the physical, ecological and behavioral characteristics of the fish and habitats under investigation (Lessions 1996; Rotherham et al. 2007). Available methods may be broadly separated into destructive, such as fishery-based methods (Thrush and Dayton 2002), and non-destructive such as Underwater Visual Survey methods (UVS; Hill and Wilkinson 2004; Andaloro et al. 2011, 2013). The former typically involve the mechanical removal of biotic or abiotic components from the natural environment, while the latter are mainly restricted to visual techniques. Non-destructive methods are usually preferred as the most adequate ones when sampling for conservation purposes, especially when assessing fish and habitat types that require protection, e.g. coral reefs, seagrass meadows, and endangered species.

UVS methods for the assessment of fish include five main quantitative or semi-quantitative methods, which can be carried out either through diving (SCUBA or free diving), or through the examination of video and photographic records. These methods include plot sampling (strip transects and point counts), distance sampling (line transects and point transects), fixed-time transects, occupancy estimation based on repetitive sampling, and rapid visual techniques (Katsanevakis et al. 2012). In this study, the first two methods (specifically, strip transects and line transects) were further analyzed and compared.

In shallow water reef fish assemblages, plot sampling, and especially strip transects, is the most widely used UVS technique (Hilborn and Walters 1992; Cheal and Thompson 1997; Watson and Quinn 1997). Strip transects is an easy to apply, low-cost technique, as it can be performed through SCUBA diving or snorkeling, depending on water visibility and depth, with minimum equipment requirements (Holmes et al. 2013). During strip transects, observations of target fish are made within a predetermined surface area (Cote and Perrow 2006). Mapstone and Ayling (1993) proposed that mid-sized strips, i.e. 50 or 75 m length and 5 or 10 m width, are suitable to obtain a representative sample of the fish community. The optimal swimming speed of the observer is usually accepted to be a compromise between a rapid constant pace (necessary to avoid implications due to fish movement) and search efficiency (Samoilys and Carlos 2000).

A crucial assumption in strip transect sampling is that detectability within the investigated area is perfect. Yet again, when assessing fish populations, there are several reasons that may lead to imperfect detectability, and subsequently, result in an underestimation of species composition and population density (Monk 2014). Several studies have shown that detectability varies considerably across fish species and is mostly affected by body size, schooling behavior, shyness and cryptic coloration or behavior (MacNeil et al. 2008b; Bozec et al. 2011). Environmental factors such as habitat complexity (Edgar and Barrett 1999) and water visibility (MacNeil et al. 2008a, b) also influence detectability. Alongside the various morphological and ecological characteristics of different species and habitats, several methodological factors, such as the selection of strip width, also affect the level of detectability (Kulbicki and Sarramégna 1999). Consequently, species richness and abundance may be substantially underestimated in strip transects (Franzreb 1981; Katsanevakis 2009).

In many cases, the problem of imperfect detectability can be addressed through distance sampling, as this method properly accounts for detection probability (Buckland et al. 2001, 2004). In the marine environment, line transects, is the most commonly used distance sampling technique (Katsanevakis et al. 2012). The sampling process followed is similar to that of strip transects, but fish observations are not restricted within a pre-defined strip width; instead, the perpendicular distance of each fish observation from the transect line is recorded. These perpendicular distances are then used to account for the detection probability (Buckland et al. 2001). Estimating the detection probability (Pa) is the most important task of the analysis related to distance sampling data (Buckland et al. 2001).

A critical assumption of distance sampling that should be ensured by the survey design and protocols is that detection on or near the line is perfect (Buckland et al. 2001; Thomas et al. 2010). In the case of violation of this assumption, a negative bias in the estimation of abundance is expected. Another important requirement is that all measurements of the distances are precise. Tape lines and laser rangefinders usually offer more precise measurements than rough estimates by eye, which may be affected by the observers' s visual ability (Thresher and Gunn 1986). Moreover, water turbidity may also affect distance estimations, as in clear waters distances are commonly underestimated, while in turbid waters they are overestimated (Kulbicki 1998).

Both methods suffer from many additional sources of bias. They both depend on the observer's ability to identify fish species *in situ* (Thompson and Mapstone 1997). Fish are assumed to be observed at their original location, before being influenced by the researcher's presence, as important bias in abundance estimation may be caused due to fish movement in response to observer's presence (Fewster et al. 2008). This largely depends on the behavior of different fish species; if individuals are attracted by the researcher the bias will be positive, while in the case of avoidance, the bias will be negative. Abundance will also be overestimated if the same individuals are recorded more than once due to their movement ahead of the observer. Biases caused by the observer are likely to be restricted by experience (Sale and Sharp 1983; Thompson and Mapstone 1997), while biases related to the distribution and behavior of individuals will differ according to the field protocols. The attitude of fish towards the observer also varies according to the different levels of fishing pressure in the area under study (Bohnsack and Bannerot 1986; Bellwood 1998). Kulbicki (1998) showed that, due to divergent fish behavior, marine protected areas would seem to have higher estimated fish densities than areas with high fishing pressure even for the same real values of density.

The aim of this study was to quantitatively compare the performance of the strip and line transect methods, for the assessment and monitoring of Mediterranean rocky reef fish in a non-destructive manner, and to investigate potential differences in the outputs of the two methods.

2. Methodology

2.1 Study area

The study area comprises the Greek territorial waters of the Aegean Sea. The study was conducted from July to October 2016 and included 31 rocky reef sites (Fig. 1). At each site but one (due to lack of appropriate substratum) two stations were surveyed.

2.2 Sampling methods & target species

Every station was assessed using both strip and line transects, each one covering a total distance of 75 m length. All transects were conducted on rocky reef habitats, at a water visibility of at least 20 m, while the exact location of the transects was randomly selected. For strip transects the transect width was 5 m (2.5 m on either side of the transect line). In order to minimize disturbance, fish recording and transect deployment were done simultaneously by the observer. In line transects, the perpendicular distance of individual fish (or cluster of fish) from the line was measured using a measuring tape, for fish detected up to 8 m on either side of the central line. When individuals were observed in schools (clusters), the distance of the center of the school was estimated as well as the number of individuals in the school. The survey targeted twenty fish taxa (Table 1).

2.3 Estimating population densities

In strip transects, the population mean density was estimated by the formula:

$$\hat{D} = n/\,2wL = n/A_c$$

n: number of individuals

2w: total width of the transect

L: length of the transect

A_c: total covered (sampled) area

Bootstrap (bias-corrected and accelerated with 1000 permutations) was applied to estimate, for each species, the unconditional standard error (Efron and Tidshirani 1993), as well as the 95%

bootstrap-based unconditional confidence interval of the mean density, using R version 3.2.3 (R Development Core Team 2015).

For line transect data, the mean density is given by

$$\hat{D} = n/(A_c P_a)$$

where P_a is the detection probability, given by:

$$P_a = \frac{\int_0^w g(y)dy}{w}$$

where *w* is the half-width of the line transects and *g(y)* is the detection function, representing the probability of detecting an individual that is at a distance *y* from the transect line (Buckland et al. 2001).

The function *g(y)* was estimated from the distance data (grouped data, right truncated at width that varied from 1.2 m to 8 m, depending on the dataset of each species to exclude outliers) with a semi-parametric approach, according to Buckland et al. (2001), using the software DISTANCE 6.2 (Thomas et al. 2010). Specifically, the detection function was modeled in the general form:

$$g(y) = \frac{key(y)[1 + series(y)]}{key(0)[1 + series(0)]}$$

where *key(y)* is the key function and *series*(*y*) is a series expansion used to adjust the key function. The uniform function *key(y)* = 1/*w* (0 parameters), the one parameter half normal function $key(y) = \exp\left(-y^2 / 2\sigma^2\right)$ and the two –parameter hazard-rate function $key(y) = 1 - \exp\left[-\left(y/\sigma\right)^{-b}\right]$ were considered as key functions; three series expansions were considered: the cosine series $\sum_{j=1}^{m} a_j \cos(j\pi y/W)$, simple polynomials of the form $\sum_{j=1}^{m} a_j \left(\frac{y}{W} \right)^{2j}$ and hermite polynomials of the form $\sum_{j=2}^{m} a_j H_{2j}(y/\sigma)$, where σ and a_j are the best-fit parameters (Buckland et al. 2001).

Six models were considered for *g(y)*: uniform key with cosine or simple polynomial series expansions, the half normal key with cosine or hermite polynomial series expansions and hazard-rate key with cosine or simple polynomial series expansions, as proposed by Buckland et al. (2001). Model selection was based on the Akaike's Information Criterion (AIC) (Akaike 1973). The number *j* of parameters in each series expansion was also defined using AIC between models of increasing order. The model with the smallest AIC value (AIC_{min}) was selected as the 'best' among the models tested.

2.4 Comparing occupancy, species richness and density estimates between strip and line transects

The occupancy of each species (percentage of stations in which the species were recorded), species richness and population density estimates, based on the two different sampling methods were compared. Occupancy was estimated for each of the 20 species per method separately. This resulted into two distinct datasets, each consisting of 20 occupancy values; one for the line transect method and one for the strip transects method. The set of differences between line and strip transects (i.e. line transects minus strip transects) was then subjected to bootstrapping. Similarly, the comparison of species richness values obtained by the two different methods was achieved through the bootstrapping technique. Initially, species richness was estimated for each station and method separately. Consequently, two datasets of 61 species richness values each were obtained for the two methods. The set of differences when subtracting the second dataset from the first, was the actual dataset that was bootstrapped. A similar procedure was followed for the comparison of the density estimates between the two methods. For the comparison of the 'overall densities', the mean density of each species over all stations was estimated by each method. The differences between the two datasets (comprising of the 20 mean densities of distinct species) were bootstrapped, while stations in which the species was not found were excluded, as the aim was to test for differences in the estimates of densities between the two methods when a species was actually present. Additionally, the density for each species at each station was also estimated. Therefore, two datasets (one for each method) with 61 values, corresponding to the number of stations, were created. The differences by subtracting the dataset of strip transects from the dataset of line transects were bootstrapped to estimate the confidence interval of the differences and test if it differed from zero. Again, stations in which a species was not recorded were excluded from the analysis of the specific species.

2.5 Species composition

To investigate potential differences in species composition between the two sampling methods, a Bray-Curtis similarity matrix was generated based on a square-root transformation of fish density data, which was then used to carry out cluster analysis and construct a non-metric multidimensional scaling (nMDS) plot. In this case, fish density data (by both methods) derived only from one of the two stations of each site (31 stations in total) were used, in order to improve clarity. Moreover, in the respective plots different colors were used for the visual depiction of the station geographical position; stations marked with cold colors (shades of blue) refer to areas of the northern Aegean, stations marked with warm colors (yellow/orange/red) are located in the southern Aegean, while green colors denote stations found in the central Aegean Sea. The species composition analysis was carried out with PRIMER 6 software (Clarke 1993).

3. Results

3.1 Distance sampling analysis

For each species, the best model, based on AIC, was used for inference (Table 2). An empirical minimum of observations to model the detection function is 30 observations (Buckland et al. 1993). However, a number of species did not fulfill this requirement. These species were *Dentex dentex, Epinephelus marginatus, Muraena helena, Sciaena umbra* and *Spondyliosoma cantharus.* The highest detectability values (excluding species with very low number of observations <30) were recorded for *Epinephelus costae* (0.84) followed by *Siganus luridus* (0.73). The lowest detectability values were recorded for *Scorpaena* spp. (0.32) and *Serranus cabrilla* (0.41) followed by *Mullus surmuletus* (0.49). The estimated detection probability curves corresponded to different fish behaviors (Fig. 2) according to Kulbicki (1998). Species that presented shy behavior (i.e. avoiding the observer) were *Diplodus annularis, D. puntazzo, D. sargus, D. vulgaris, Oblada melanura, Sparisoma cretense, S. luridus* and *Siganus rivulatus.* Species that presented neutral behavior were *M. surmuletus, E. costae, Serranus scriba, S. cabrilla* and *Sarpa salpa* while species with cryptic behavior were *Scorpaena* spp. for which a rapid decrease in detectability was obvious within the first 0.4 m (Fig. 2A).

3.2 Species Occupancy

Across all sites, *D. vulgaris* was the most commonly occurring species, as it was recorded in 58 stations by both methods, while *Dicentrahus labrax* was never recorded (Fig. 3). Occupancy estimates for the target species varied between the two methods; line transects gave higher estimates in 12 cases, strip transects gave higher estimates in 4 cases, while for three species they gave the same estimates (Fig. 3). The highest observed differences were for *Scorpaena* spp., with an estimated occupancy of 0.64 by line transect sampling and 0.10 by strip transect sampling. The bootstrap method conducted to compare occupancy estimates (expressed in percentages) between the two methods showed significant differences (mean: 5.7, 95% Confidence Interval (CI):1.3, 11.3).

3.3 Species Richness

Species richness (i.e. the number of species per station) was estimated significantly higher in 36 stations by line transects and in 11 stations by strip transects, while in 14 stations no significant differences were detected between the two methods (Fig. 4). According to the bootstrap method, the mean difference of species richness was 0.98 [CI: 0.57, 1.40], thus indicating significantly higher species richness estimates in line transects than strip transects.

3.4 Density estimates

Fish density per station (i.e. number of individuals per hectare) was highly variable both among species and between methods. The most abundant species was *D. vulgaris*, which presented the highest density with a mean value of 702.9 individuals per hectare for the line transects and

567.8 individuals per hectare for the strip transects. Species that also presented high-density values were *S. salpa, O. melanura* and *S. luridus* (Table 3, Fig. 5). The least abundant species was *S. umbra* which presented a mean density of 1.56 individuals per hectare in the line transects, while no individuals were recorded in the strip transects. Other species that presented low-density values were the *M. helena, D. dentex* and *E. marginatus* (Table 3, Fig. 5).

The mean difference of the overall fish density was significantly higher for line transects than for strip transects (50.5 individuals per hectare; CI [18.0, 85.7]). However, when examining density estimates for each species separately results varied (Table 4). For *D. sargus, D. vulgaris, D. dentex, Scorpaena* spp., *S. cabrilla, S. scriba, S. luridus* and *S. rivulatus* the line transects estimates were significantly higher than strip transects, while the opposite was found for *E. costae* and *S. cantharus.* No statistically significant differences between the two methods were found for *D. annularis, D. puntazzo, E. marginatus, M. surmuletus, M. helena, O. melanura, S. cretense* and *S. salpa*. No comparison was possible *D. labrax* and *S. umbra* due to lack of data.

3.5 Comparing species composition between sampling methods

To investigate the similarity of species composition among stations and between methods, the analysis was restricted to one station from each site to improve clarity; otherwise, the resulting MDS plot and dendrogram were too crowded (with 122 points-61 stations x 2 methods). The same analysis with the other half stations gave quite similar results (not shown here). In the majority of cases, the two methods presented similar species composition within the stations. Stations 13, 27 and 19 presented the highest resemblance, showing a similarity of 84%, 82% and 80% respectively (Figs. 6, 7). However, in some stations the resulting similarity in species composition between the two methods was low (Figs 6, 7). Specifically, stations 48, 50 and 9 demonstrated the lowest similarity in species composition between the two methods (i.e. 22%, 30%, and 45% respectively). A clear separation between distinct geographical regions (North and South Aegean Sea) was obvious in both methods.

4. Discussion

Statistically significant differences were detected between line and strip transects in the estimates of occupancy and species richness. The higher overall estimates of occupancy and species richness by the line transect method are mainly attributed to the narrower strip transect width, and thus, the lower surveyed surfaces when using the latter method. The use of narrow strips is dictated by the need to satisfy the assumption of perfect detectability, which is the main assumption of strip transects (Katsanevakis et al. 2012). On the contrary, in line transects perfect detection is required only "on the line"; this allows to expand the width of the transects in order to survey larger surfaces, and increases the probability to record infrequent species. Furthermore, the reaction of fish to the presence of the observer can be crucial for the detection of a species. Many 'shy' species may react to the divers' presence by fleeing away at distances greater than the fixed width of the strip transect, and hence remain undetected. Bozec et al. (2011) indicated that 'shy' species display a clear avoidance behavior towards the diver, while the distance that fish may flee from the observer increases with fish size. The appropriate width of the strip transect to ensure species detection may differ even for closely related species (Kulbicki and Sarramégna 1999), or even when considering the same species but in a different habitat (Smith and Nydegger 1985; Einsing et al. 1995; Cheal and Thompson 1997). By extending the surveyed width through the use of line transects, these sources of error can be reduced.

With regards to overall fish density, line transects again led to a higher estimate than strip transects. This difference is partly related to fish behavior (Bozec et al. 2011; Pais and Cabral 2017). Kulbicki (1998) pointed out that fish are not motionless items and in most cases, they will be either scared or attracted by the observer, while these reactions may change from site to site. 'Shy' species records peak at distances >0 as they tend to keep a distance from the observer. The frequency distribution of distances for the majority of the species in the present study followed the pattern of 'shy' species. In these cases, the peak of the distance frequency distribution of fish observations was at distances between 0.7 – 2.2 m from the line. Fish behavior is therefore, a possible reason why line transects, which utilized a wider surface area (i.e. 8 m on either side of the transect), yielded higher overall density estimates compared to strip transects (i.e. 2.5 m on either side of the transect), as some 'shy' fish could have moved beyond the limit of the transects and thus were not recorded. Nevertheless, many species seemed to flee at distances <2.5 m and thus were not missed in the strip transects.

Furthermore, another important factor which may lead to a potential underestimation of abundance in strip transects, especially for cryptic species, is imperfect detectability (Franzreb 1981; Kulbicki 1998). A transect with a narrow width may yield poor population estimates both for the more mobile species (Samoilys and Carlos 2000), such as *Siganus* spp., but also for the small cryptic species (Bozec et al. 2011), such as *Scorpaena* spp. The results from DISTANCE analysis showed that a sharp decline in detectability is obvious at distances >2.5 m from the transect line for the majority of the surveyed species. Bozec (2011) also stated that a progressive rise in diver's avoidance up to 3 m from the transect line was apparent with increased fish size.

Moreover, numerous studies have also shown that an obvious decline in detectability is observed at approximately 3 m distance from the transect (Harmelin-Vivien et al. 1985; Smith and Nydegger 1985; Fowler 1987; McCormick and Choat 1987; Cheal and Thompson 1997; Kulbicki and Sarramégna 1999). According to the above, the 2.5 m width on each side of the strip transects used in the present study should be sufficient for the detection of the majority of the target species. However, there were several exceptions, such as *Scorpaena* spp. (Fig. 2), *S. cabrilla*, and *S. scriba*, which presented a substantial decline in detectability at distances <2.5 m. For these latter species the density estimates by line transects were substantially higher than by strip transects.

Although, in most cases line transects yielded higher estimates, some sources of bias are yet to be mentioned. An important assumption in distance methodology is that fish should be recorded prior to any movement as a response to the observer's presence. A potential violation of this basic assumption is known to lead to a negative bias in abundance estimates of 'shy' species (Buckland et al. 1993). Moreover, the additional time needed to carry out the distance measurements and the actual deployment of a tape-measure, may further augment the fleeing response of more mobile fish, and hence lead to an underestimation of their numbers during line transects. Yet, this source of bias is considered to be more intense in areas of high fish densities (Watson et al. 1995).

The multivariate analysis of the species composition indicated an overall high resemblance between the two methods. In most stations the majority of the species recorded by one method were also recorded by the second method, and at similar densities. These results suggest that the choice of a specific method (either plot sampling or distance sampling) should not significantly affect the overall outcome regarding the spatial patterns of species composition, especially in large scale studies.

Unfortunately, as is the case in most field studies, the real density values of the fish species in the areas under study were not known. Therefore, it is not easy to determine which is the 'best' method by providing precise estimates of the biases related to each method per species. According to several studies, distance sampling appears to be advantageous in many cases. Kulbicki and Sarramégna (1999) have proposed that the use of distance sampling method in UVC surveys could potentially improve estimates by yielding values closer to the true values. Similarly, Einsing et al. (1995) showed that distance sampling, compared to quadrat sampling and strip transects, produced density estimates that were closer to true densities, while Thresher and Gunn (1986) proposed that distance sampling should be preferred for the assessment of cryptic species.

5. Conclusion

Both methods have several specific advantages and limitations, and both are prone to biases. Strip transects suffer from imperfect detectability and the related necessity of narrow transect widths, which may cause underestimation of densities, occupancy, and species richness. In line transect sampling, detection probability is properly taken into account, but still the assumption that all individuals are detected at their initial position is difficult to be satisfied especially for fish of high mobility. Line transect sampling is expected to provide much more accurate estimates than strip transect sampling in the case of cryptic species of low mobility. An additional advantage of the line transect method is that it provides a way to assess fish behavior through the analysis of distance frequency graphs. On the contrary, in the case of mobile species with neutral or close to neutral behavior, and especially at high fish densities, strip transects would probably be more efficient, as line transects are time-consuming and the disturbance of fish would be higher due to the distance measurements. The choice of the best method to apply needs careful consideration and depends on the aims of each study, the target species, and the peculiarities of the study area. One benefit of line transect sampling is that it provides a way to assess fish behavior through the analysis of distance frequency graphs. Joint application of both methods could be considered, with line transects applied by one observer for cryptic and large fish, and strip transects by another observer for the bulk of medium-sized mobile fish. Further research is needed to improve the performance of both methods and reduce their biases.

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Table 1(on next page)

Fish taxa surveyed (according to Horton et al. 2017)

Species	Family	Authority
Diplodus annularis	Sparidae	(Linnaeus, 1758)
Diplodus puntazzo	Sparidae	(Walbaum, 1792)
Diplodus sargus	Sparidae	(Linnaeus, 1758)
Diplodus vulgaris	Sparidae	(Geoffroy Saint- Hilaire,1817)
Dentex dentex	Sparidae	(Linnaeus, 1758)
Dicentrachus labrax	Moronidae	(Linnaeus, 1758)
Epinephelus costae	Serranidae	(Steindachner, 1878)
Epinephelus marginatus	Serranidae	(Lowe, 1834)
Mullus surmuletus	Mullidae	Linnaeus, 1758
Muraena helena	Muraeninae	Linnaeus, 1758
Sparisoma cretense	Scaridae	(Linnaeus, 1758)
Scorpaena spp.	Scorpaenidae	Linnaeus, 1758
Oblada melanura	Sparidae	(Linnaeus, 1758)
Sarpa salpa	Sparidae	(Linnaeus, 1758)
Sciaena umbra	Scianidae	Linnaeus, 1758
Serranus cabrilla	Serranidae	(Linnaeus, 1758)
Serranus scriba	Serranidae	(Linnaeus, 1758)
Siganus luridus	Siganidae	(Rüppell, 1829)
Siganus rivulatus	Siganidae	Forsskål & Niebuhr, 1775
Spondyliosoma cantharus	Sparidae	(Linnaeus, 1758)

Table 2 (on next page)

Best fit model, maximum width of line transect after truncation (w) and value of detectability (Pa) of the DISTANCE analysis for each species.

Species	model	*Wmax (m)*	P_a
Diplodus annularis	Hazard rate, simple polynomial of order 2	6.9	0.57
Diplodus puntazzo	Uniform, cosine of order 1	6.9	0.60
Diplodus sargus	Hazard rate, cosine of order 2	6.8	0.64
Diplodus vulgaris	Uniform, cosine of order 2	7.0	0.66
Dentex dentex *	Uniform	7.3	1.00
Disentrachus labrax	-	-	-
Epinephelus costae	Hazard rate, hermite of order 2	6.5	0.84
Epinephelus marginatus *	Uniform, cosine of order 1	7.0	0.58
Mullus surmuletus	Hazard rate, simple polynomial of order 2	8.0	0.49
Muraena helena *	Half normal, cosine of order 1	4.2	0.99
Oblada melanura	Hazard rate, simple polynomial of order 2	7.6	0.66
Sparisoma cretense	Hazard rate, simple polynomial of order 2	6.0	0.78
Sciaena umbra *	Half normal, cosine of order 1	1.4	0.99
Sarpa salpa	Hazard rate, simple polynomial of order 2	6.0	0.68
Scorpaena spp.	Half normal, cosine of order 2	1.2	0.32
Serranus cabrilla	Hazard rate, simple polynomial of order 3	5.0	0.41
Serranus scriba	Half normal, hermite of order 1	6.0	0.54
Siganus luridus	Hazard rate, simple polynomial of order 2	6.0	0.73
Siganus rivulatus	Uniform, cosine of order 1	6.3	0.56
Spondyliosoma cantharus *	Uniform	6.7	1.00

Table 3 (on next page)

Mean population densities and 95% confidence intervals for all species per sampling method (line or strip transects).

Species	Method					
	Line			Strip		
	Mean (N of individuals/he)	CI		Mean (N of individuals/he)	CI	
Diplodus annularis	100.7	72.5	132.7	87.8	55.9	124.6
Diplodus puntazzo	39	26.7	53.1	33.5	23.6	44.1
Diplodus sargus	158.5	121.8	196.7	72.5	56.8	89.1
Diplodus vulgaris	703.5	606.9	803	568.6	472.5	672.3
Dentex dentex	8.5	1.8	17	1.2	0	3
Disentrachus labrax	0	0	0	0	0	0
Epinephelus costae	13.1	6.1	20.8	21.3	10	33.6
Epineph. Marginatus	4.8	2.5	7.6	5.2	1.7	10.4
Mullus surmuletus	48.5	35.3	62.5	44.9	31	61.2
Muraena helena	2.3	1	4.1	0.8	0	1.7
Oblada melanura	312.8	241.5	382.2	319.7	197.5	456.4
Sparisoma cretense	252.3	192.5	317	243	177.4	312.6
Sciaena umbra	1.6	0	3.9	0	0	0
Sarpa salpa	421.1	321.4	525.3	381.7	290.2	478.7
Scorpaena spp.	178	127.4	234.1	4.3	1.3	7.8
Serranus cabrilla	85.2	60.7	110.3	63.4	44.1	85.6
Serranus scriba	232.1	184.2	282.5	167.1	129.3	208.9
Siganus luridus	529.9	380.5	662.3	281.4	198	372.5
Siganus rivulatus	189.1	102.3	281.3	40.7	15.7	69.5
Spondyliosoma cantharus	3.5	1.6	5.8	40	11.8	79.1

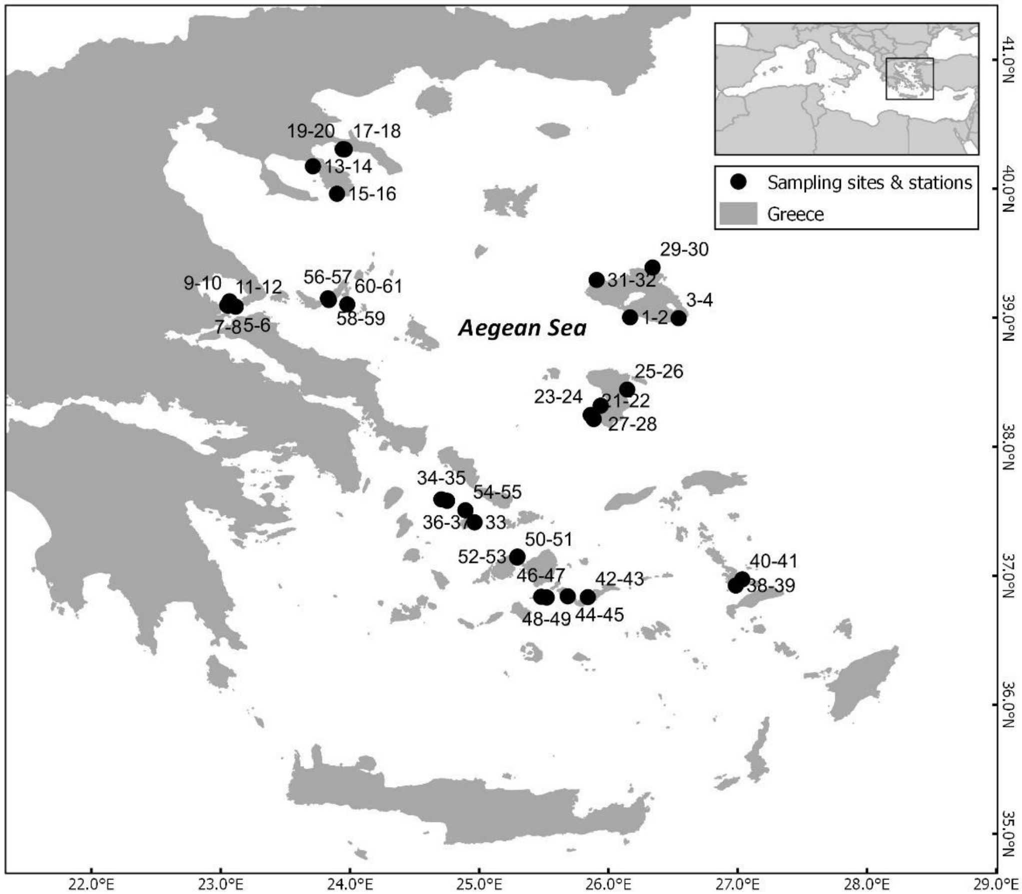
Table 4(on next page)

Mean differences (line transects - strip transects) of overall density estimates per species. 95% confidence intervals have been estimated by bootstrapping.

Species	Mean (N of individuals/he)	95% Confidence interval	
Overall	50.5	18.0	85.7
Diplodus annularis	18.9	-32.0	74.2
Diplodus puntazzo	7.9	-17.6	31.0
Diplodus sargus	92.7	45.9	140.1
Diplodus vulgaris	138.6	3.3	269.6
Epinephelus costae	-25.6	-55.4	-1.5
Epinephelus marginatus	-1.9	-20.0	13.2
Dentex dentex	73.7	29.6	145.3
Disentrachus labrax	0.0	0.0	0.0
Mullus surmuletus	6.2	-21.8	32.3
Muraena helena	10.0	-4.2	22.9
Oblada melanura	-11.4	-205.4	166.8
Sciaena umbra	1.54	0.0	3.12
Sparisoma cretense	9.4	-63.6	79.4
Sarpa salpa	50.6	-85.6	181.5
Scorpaena spp.	347.0	263.6	444.0
Serranus cabrilla	35.1	5.3	66.5
Serranus scriba	71.9	24.3	116.9
Siganus luridus	506.3	304.2	697.9
Siganus rivulatus	609.7	339.7	884.9
Spondyliosoma cantharus	-137.5	-300.1	-24.6

Figure 1(on next page)

Map of the sampling area depicting the different sites and the code numbers of sampling stations.



Sampling sites & stations
Greece
Aegean Sea
19-20
17-18
13-14
15-16
9-10
11-12
7-8
5-6
56-57
60-61
58-59
29-30
31-32
3-4
1-2
25-26
23-24
21-22
27-28
34-35
54-55
36-37
33
50-51
52-53
46-47
42-43
48-49
44-45
40-41
38-39
41.0°N
40.0°N
39.0°N
38.0°N
37.0°N
36.0°N
35.0°N
22.0°E
23.0°E
24.0°E
25.0°E
26.0°E
27.0°E
28.0°E
29.0°E

Figure 2 (on next page)

Typical distance distributions of three different fish behaviors.

(A) cryptic behavior, (B) shy behavior and (C) neutral behavior (Kulbicki, 1998). The estimated detection probability function is shown with the red line (forced to be monotonically decreasing).

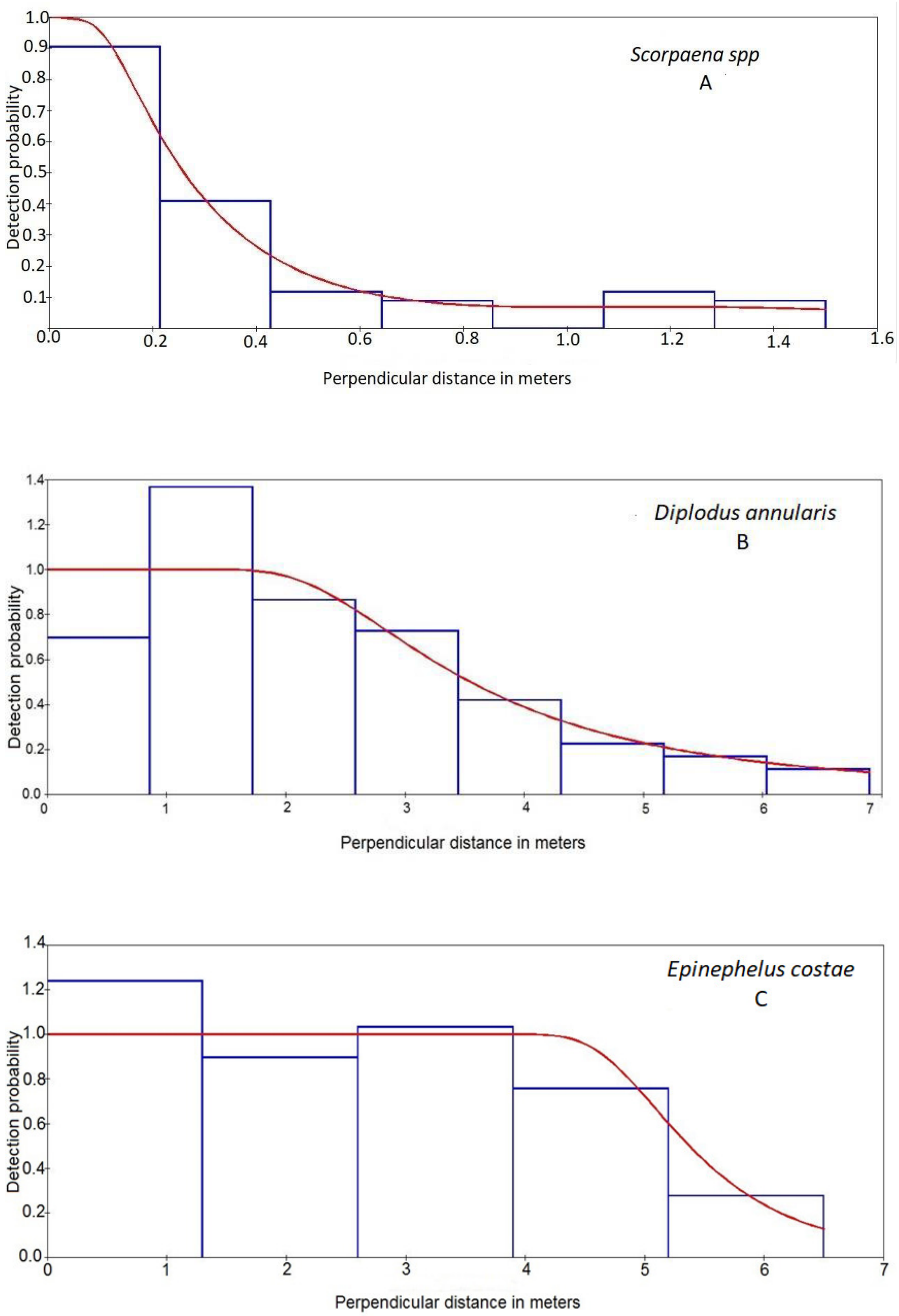

Scorpaena spp
A
Detection probability
Perpendicular distance in meters
Diplodus annularis
B
Detection probability
Perpendicular distance in meters
Epinephelus costae
C
Detection probability
Perpendicular distance in meters

Figure 3(on next page)

Total number of stations where different species were observed through the line and strip transects methods.

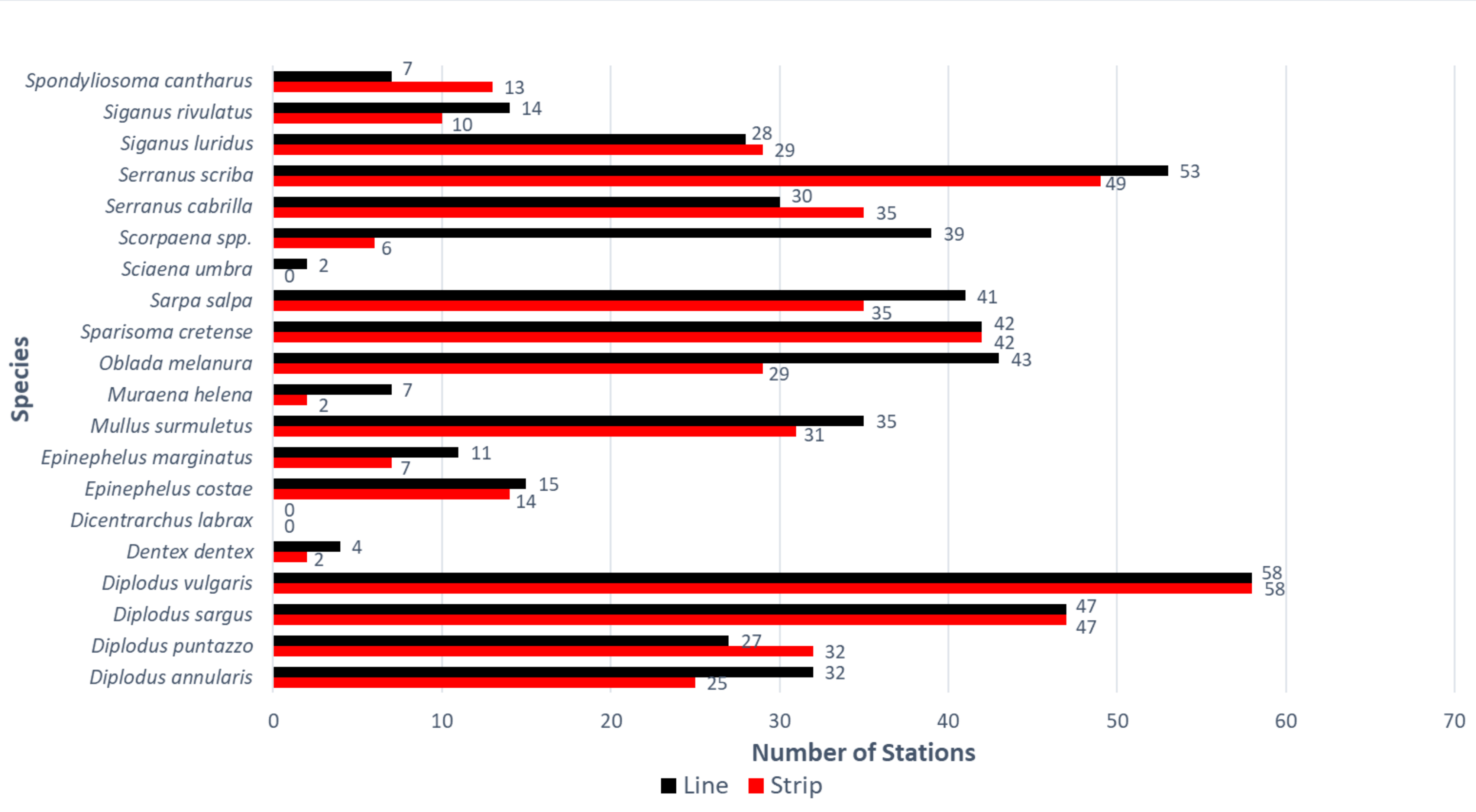

Spondyliosoma cantharus
Siganus rivulatus
Siganus luridus
Serranus scriba
Serranus cabrilla
Scorpaena spp.
Sciaena umbra
Sarpa salpa
Sparisoma cretense
Oblada melanura
Muraena helena
Mullus surmuletus
Epinephelus marginatus
Epinephelus costae
Dicentrarchus labrax
Dentex dentex
Diplodus vulgaris
Diplodus sargus
Diplodus puntazzo
Diplodus annularis
Species
7
13
14
10
28
29
53
49
30
35
39
6
2
0
41
35
42
42
43
29
7
2
35
31
11
7
15
14
0
0
4
2
58
58
47
47
27
32
32
25
0
10
20
30
40
50
60
70
Number of Stations
Line
Strip

Figure 4(on next page)

Histogram of the differences in estimated species richness by the line and strip transects methods.

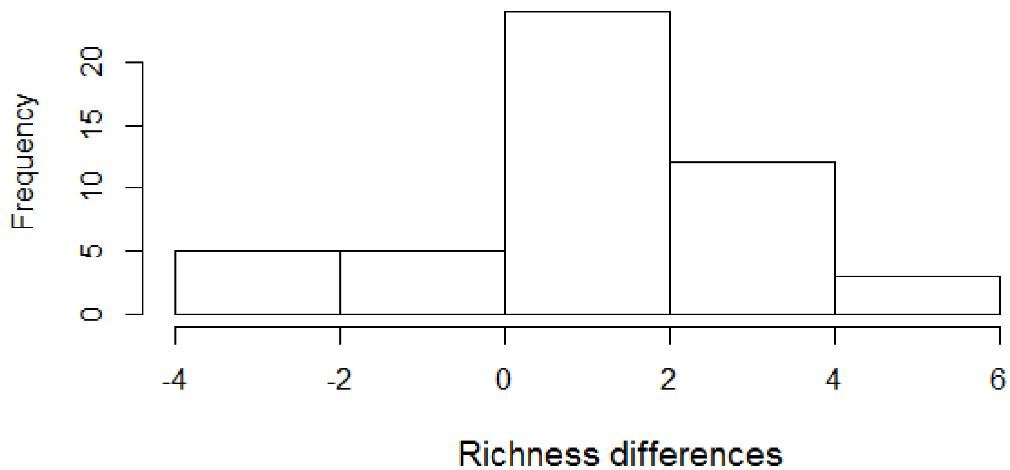

Frequency
0
5
10
15
20
-4
-2
0
2
4
6
Richness differences

Figure 5 (on next page)

Mean density, and standard error of mean, per fish species obtained through line and strip transects.

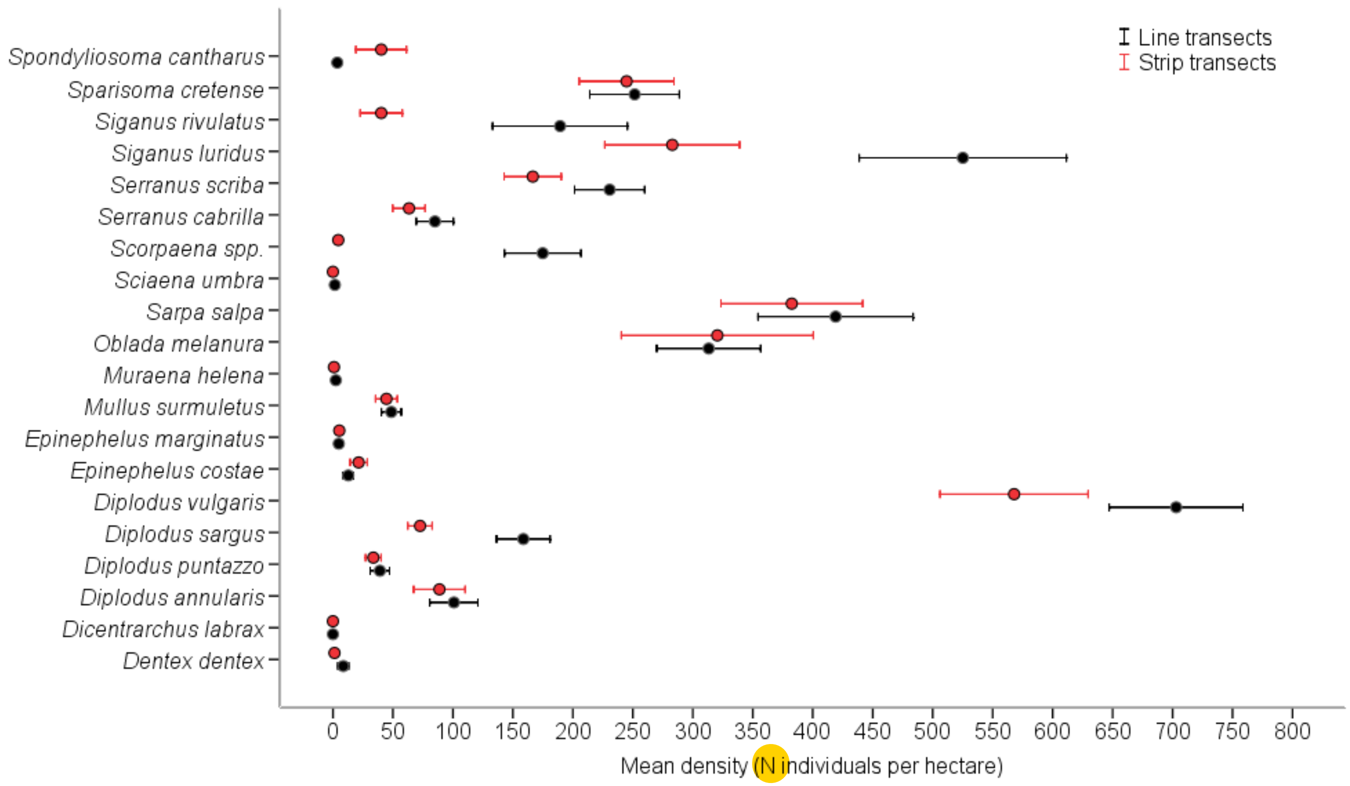

Spondyliosoma cantharus
Sparisoma cretense
Siganus rivulatus
Siganus luridus
Serranus scriba
Serranus cabrilla
Scorpaena spp.
Sciaena umbra
Sarpa salpa
Oblada melanura
Muraena helena
Mullus surmuletus
Epinephelus marginatus
Epinephelus costae
Diplodus vulgaris
Diplodus sargus
Diplodus puntazzo
Diplodus annularis
Dicentrarchus labrax
Dentex dentex
Line transects
Strip transects
0 50 100 150 200 250 300 350 400 450 500 550 600 650 700 750 800
Mean density (N individuals per hectare)

Figure 6(on next page)

Two dimensional non metric multidimensional scaling ordination (MDS) for 31 paired-by-method stations, based on square root-transformation density data and a Bray-Curtis similarity matrix.

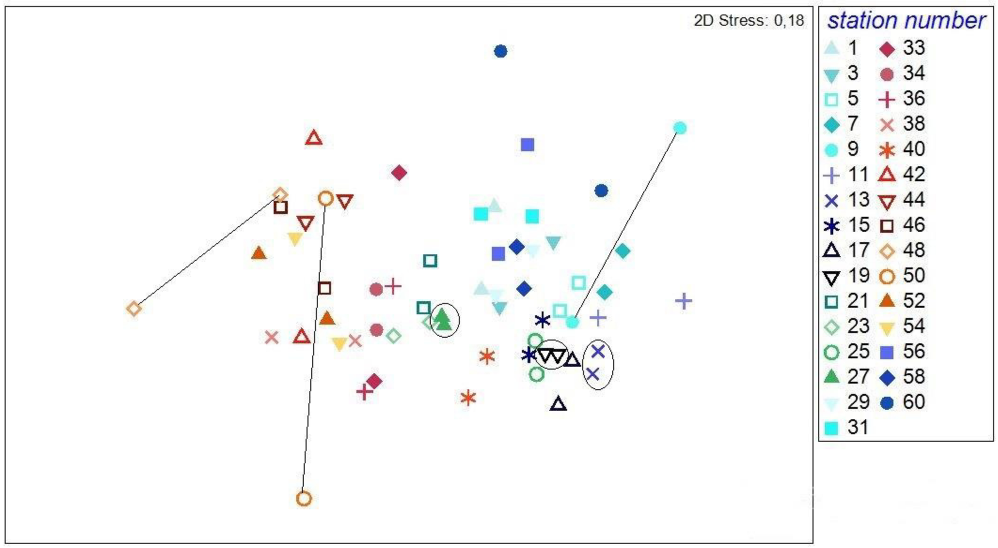
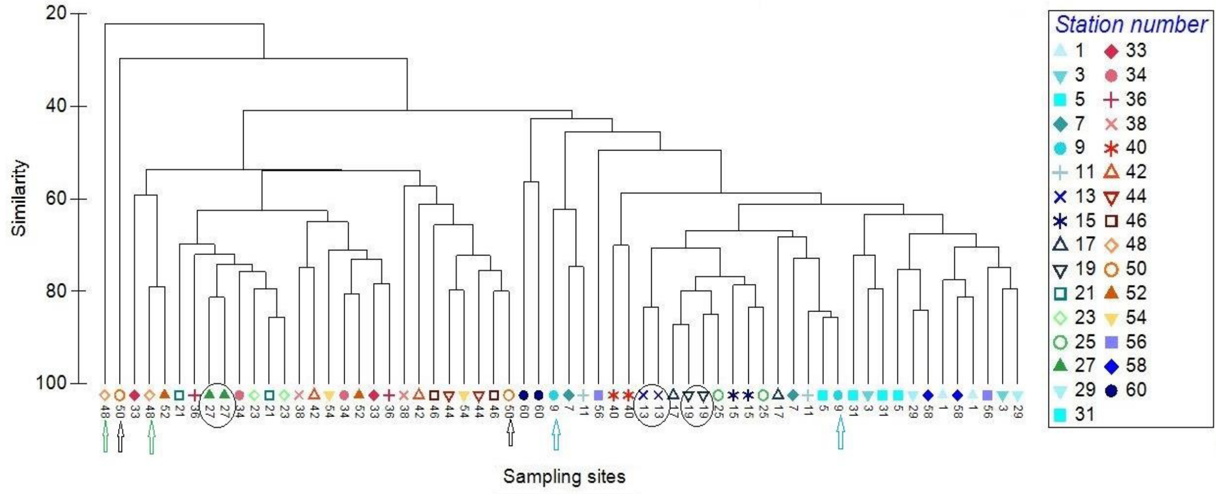

2D Stress: 0,18
station number
1
3
5
7
9
11
13
15
17
19
21
23
25
27
29
31
33
34
36
38
40
42
44
46
48
50
52
54
56
58
60

Figure 7 (on next page)

Cluster analysis of the paired–by-method stations' similarity, based on square root-transformation density data and a Bray-Curtis similarity matrix.


Similarity
20
40
60
80
100
Sampling sites
Station number
1
3
5
7
9
11
13
15
17
19
21
23
25
27
29
31
33
34
36
38
40
42
44
46
48
50
52
54
56
58
60