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Acoustic comparison of a patchy Mediterranean shallow water seascape: *Posidonia oceanica* meadow and sandy bottom habitats

Ceraulo M. [1,3], Papale E. [1], Caruso F. [1,4], Filiciotto F. [2], Grammauta R. [1], Parisi I.[5], Mazzola S. [1], Farina A. [3] and Buscaino G. [1]

[1] National Research Council – Institute for Coastal Marine Environment – Bioacousticslab Capo Granitola, Via del Mare, 6 – 91021 Torretta Granitola, Campobello di Mazara (TP), Italy

[2] National Research Council – Institute for Coastal Marine Environment – Spianata S. Ranieri, 86 - 98122 Messina Italy

[3]Department of Pure and Applied Sciences (DiSPeA) – University of Urbino– Campus Scientifico "Enrico Mattei"– 61029 Urbino, Italy

[4]Woods Hole Oceanographic Institution, Biology Department, Woods Hole, MA 02543, USA

[5]eConscience, no-profit organization, via Provinciale 610 , 90046, Monreale (PA), Italy

Abstract

Soundscapes are strongly linked with the physical structure and biological features of the habitats and their study can reveal ecological processes of the underwater environment. Objective of this study is to characterize two Mediterranean habitats, the *Posidonia oceanica* meadow and the sandy bottom, and demonstrate their acoustic diversification basing on their soundscapes. Firstly, the habitats have been compared using two different acoustic metrics, the Power Spectral Density (PSD) and the Acoustic Complexity Index (ACI), measured in different frequency band. Then, the acoustic biological component of the habitats has been identified and characterized: five biological signals were described and their acoustic properties and temporal patterns were defined. Finally, the geophonical and anthropogenic components of the two habitats have been compared. In the low frequency (< 0.5 kHz) the sandy habitat showed higher values of PSD and lower values of ACI. From 0.5 to 24 kHz the greatest values of both parameters were recorded in the Posidonia habitat due to the acoustic activity of snapping shrimps and fishes. The wind speed resulted significantly correlated with PSD from 0.1 to 2 kHz for both habitats, but the correlation is less intense in Posidonia habitat suggesting a noise attenuation phenomenon. The two habitats present biophonical component belonged to different fish species and invertebrates; they showed alternated temporal pattern and different frequency allocation. The Posidonia habitat resulted acoustically richer than sandy habitat, confirming the importance of ecoacoustic method to study ecological processes. Finally, a strong acoustic impact from the anthropogenic component was revealed: it achieves 60% of daytime during the summer, especially in sandy habitat. Results demonstrated not only the possibility to discriminate habitats through the sound information but also the need to protect marine ecosystems from the human noise.

Key words: Soundscape ecology, Posidonia meadow, sandy habitat, fish signals, ACI, noise, Mediterranean Sea

1. Introduction

The sound characterizing aquatic and marine environment, or soundscape, is produced by the combination of geophonies, biophonies and antropophonies (Pijanowski et al. 2011). The geophonies are the result of sounds produced by physical agents as wind, waves and rain; the biophonies are produced by mammals, fishes and crustaceans vocalization; finally the antropophonies are originated during mechanical human activities, as ship noise, seismic prospection (air-gun), seabed drilling, etc..(Farina 2014).

In marine environment, seagrass meadows, rocky and coral reefs, are complex habitats characterized by an higher number of shelters and food opportunities (La Mesa et al. 2011), which lead the colonization of an high number of species (Giakoumi and Kokkoris 2013).

The complexity of the habitats, in terms of structure of the animal community, is connected with the complexity of its biophonical component (Kennedy et al. 2010). The food and shelters availability of the habitats can determine a different biophony, since the acoustic activity of fishes and crustaceans is related to feeding (Radford et al. 2008b), territorial and feeding competition (Myrberg 1997, Amorim & Hawkins 2000) and spawning behavior (Lugli et al. 1995, Aalbers & Drawbridge 2008). These differences can be better observed during specific periods of the day and year. The circadian cycles of the acoustic activity in marine coastal environment are regulated by the light: the acoustic emission of marine vertebrates and invertebrates increases during the night and mostly during the new moon periods (Lammers et al. 2008, Radford et al. 2008a, b, Lillis et al. 2014, Staaterman et al. 2014, Buscaino et al. 2016, Caruso et al. 2017). The acoustic activity of many fish species shows seasonal pattern following spawning and breeding periods (Amorim 2006, McCauley 2012, Buscaino et al. 2016), and androgenic factors regulate the tropic state of the sonic muscles following a strong seasonal cycle (Connaughton et al. 1997).

The physical structure of the habitats determine not only variation in biophonic components, but also in the geophonical components. In terrestrial ecosystem, it was demonstrated that wind and rain noise depends on the openness of the vegetation, the leaf area density and width and the breadth of canopy (Aylor 1972), making the spectral profile of ambient noise habitat specific (Slabbekoorn 2004). In our knowledge, specific studies of geophony propagation in marine coastal habitats have not been published yet, but the physics parameters of different marine environments determine phenomena of scattering and absorption (Hermand 2004, Knobles et al. 2008) that are commonly used by side scan sonar to differentiate the sea bottom.

The study of aquatic and marine soundscape was dealt recently using eco-acoustic indices (Lillis et al. 2014, Staaterman et al. 2014, Kaplan et al. 2015, Bertucci et al. 2015, 2016 Buscaino et al. 2016). Their use highlights the presence of biological sounds also during high background noise condition, making faster and easier the analysis and interpretation of huge amount of data (Sueur et al. 2008, Farina 2014). Harris et al. (2016), testing the correspondence between fish biodiversity and three different indices, founded that the Acoustic Complexity Index (ACI) (see Pieretti et al. 2011 for details on computation) is a good descriptor of the acoustic community in temperate marine environment.

The Mediterranean system comprises a plurality of ecosystems that allow high degree of biological diversity (Bianchi & Morri 2000). However, the coastal environment are area at high risk for sound pollution due to the increase of human pressure (Samuel et al. 2005) and different coastal habitats are exposed to different levels of human pressure, due to e.g. fishery activity, commercial shipping and recreational interests (Halpern et al. 2008). Noise increases concerns about health and fitness for all marine species from invertebrates to vertebrates (Celi et al. 2015, Filiciotto et al. 2014, 2016, Papale et al. 2015, Everley et al. 2016, Simpson et al. 2016), with influence species survival. Human disturbance of habitats soundscape reduces the orientation capacity of different species (Holles et al. 2013) because the recognition of a distinct acoustic signature for each habitat is a key mechanisms for their viability (Simpson 2005, Radford et al. 2010). The Mediterranean soundscape has been investigated only recently (Buscaino et al. 2016), and no studies describe both spatial and temporal acoustic patterns.

The Mediterranean shallow waters are characterized by an alternation of *Posidonia oceanica* meadow, sandy and rocky bottoms. *Posidonia oceanica* (a protected species for Habitat Directive 92/43/EU) is the dominant seagrass species in the Mediterranean Sea and it is the base for a crucial habitat that provides refuge, nursery and food sources for fish and invertebrate species. Instead, the sandy bottom habitats mostly provide refuge for infauna (Thiel & Ullrich 2002), and it determines peculiar speciation of organisms that become exclusive (Tunesi et al. 2006).

The purpose of this study is to distinguish two different Mediterranean habitats, the *Posidonia oceanica* meadow and the sandy bottom, basing on their soundscapes. In particular, this study aims at: 1) comparing the soundscape of the *Posidonia oceanica* meadow and sandy bottom habitats considering two different metrics, the Power Spectral Density (PSD) and the Acoustical Complexity Index (ACI) and their daily and seasonal trend; 2) evaluating if the physical agents influence in different way the background noise of the two habitats 3) identifying, describing and comparing the biotic sonic component of the two habitats; 4) analyzing the impact of anthropogenic noise on the two habitats

2. Material and Methods

2.1. <u>Study area and data collection</u>

Data collection was carried out along the south-western coast of Sicily, in an area comprised between Capo Granitola and Tre Fontane villages (Fig. 1). Here, the seascape is distinguished by an alternation of patches of sandy, rock and *Posidonia oceanica* meadows. This area is characterized by upwelling phenomena (Bonanno et al. 2014) promoting the primary and secondary production

Along the Sicilian coasts, Posidonia meadow covers about 76000 ha (Calvo et al. 2010). Thanks to the favorable ecological conditions and pristine natural state, the western side is one of the most dense and extensive beds of all the entire Mediterranean Sea (Calvo et al. 2009, 2010).

For this study, we selected three patches of sandy bottom and three patches of *Posidonia* beds on rocky bottom, alternatively distributed along the coast. Six sites were chosen (and named for *Posidonia* meadows P01, P02, P03; for sandy bottom S01, S02, and S03; Fig.1) inside the patches: the sites P01 and S01 were located to the south-western side of the coast, the sites P03 and S03 to the south-eastern and the sites P02 and S02 in the southern. The patches were selected using *Google* satellite imagines, ecosounder and visual observation.

An autonomous recorder was located for each site. They were selected considering a minimum distance from the patch boundary of 30 m. The recorders were deployed between 10 and 12 m depth at about 3 m from the bottom and 9 m from the surface, using a ballast and a buoy to maintain a vertical assessment of the hydrophone (Fig. 2). For details about site locations see Tab.1.

The autonomous recorder consisted on an omnidirectional calibrated hydrophone with a flat sensitivity response of -174.5 (± 2) dB re V/μPa from 0.1 to 100 kHz (model Benthowave Low Noise Broadband Hydrophone BII 7016 T6) and a Digital Signal Processor (model C5535 DSP-TMS320C5535) coupled with an AIC3204 audio codec (Texas Instruments).

In order to balance the limits of the data storage and the battery operating time, the instruments were set to record for 10 minutes continuously followed by 20 minutes of pause (33% of duty cycle), using a sample rate of 48 kHz at 16 bit. This configuration allowed to record for about 7 consecutive days during each deployment. The recordings took place during winter (January and February) and during summer (June, July and September). Recordings were carried out during the new moon week, to be sure to record the maximum sound activity of crustaceans and fishes (as found by Lillis et al. 2014 and by Staaterman et al. 2014). All recorders were synchronized before the deployment and data were acquired for a total of 1487 hours.

Data of speed and wind direction were collected during the recording sessions by SIAS (Regione Siciliana - SIAS – Servizio Informativo Agrometereologico Siciliano). The meteorological station is located at about 7 km far from the recording sites.

2.2. Data analysis

The dataset was aurally and visually inspected trough spectrogram survey in order to obtain the preliminary identification of biological sources and to evaluate the presence/absence of ship noise in each 10-minute file. In order to obtain a good representation of the soundscape of the areas, data analysis was carried out by considering three bands:

- Low Frequency (LF): from 0.1 kHz to 0.5 kHz
- Medium Frequency (MF): from 0.5 kHz to 2 kHz
- High Frequency (HF): from 2 kHz to 20 kHz

The choice of these bands allowed us to improve the description of biological components of the marine soundscape. Generally, fish emission range up to 2 kHz (Ladich & Fine 2006, Picciulin et al. 2013) but they are extremely variable between different groups. For this reason, we considered two bands of frequency (LF and MF) as potentially used by different fish species. The third band (HF) could be occupied by invertebrates broadband pulses that extend from 2 kHz up to 120 kHz (Au & Banks 1998, Buscaino et al. 2011, Di Iorio et al. 2012) or signals (both impulsive and tonal) of *delphinidae* species (Papale et al. 2014, Buscaino et al. 2015, Caruso et al. 2017). The day time (night and day) was established basing on the solar time of each recording session using ephemeris tables (Night and Day software- Benvegnù M. and Menichelli M.)

2.2.1. Power Spectral Density Analysis

Using MATLAB code, the Power Spectral Density (PSD - dB re 1 µPa^2/ Hz) of each recording was calculated through the *welch* function (Welch 1967) (24000 points FFT, 2^{15} points Hamming

Window, 50% Overlap).The PSD values have been summarized on the three bands (LF, MF, HF bands).

2.2.2. Acoustic Complexity Index Analysis

In order to compare the biological acoustic community of the two habitats, filtering out the non-animal produced sounds, we computed the Acoustic Complexity Index (ACI) (Pieretti et al. 2011, Harris et al. 2016, Buscaino et al. 2016) on the three frequency bands.

ACI was computed through the *SoundscapeMeter* plug in of WaveSurfer platform (for details on algorithm see Pieretti & Farina 2013). In order to obtain a temporal resolution adapted to amplify the most representative fish sound emissions (temporal step of 0.064 sec), all data were resampling at 32 kHz, and a FFT of 2048 points (frequency resolution of 15.6 Hz) was used.

Moreover, the *SoundscapeMeter* permits to apply an amplitude filter (named *noise filter*) on the data before computing the calculation of the index (Farina et al. 2016). As found by Buscaino et al 2016, during the choruses of snapping shrimps and fishes, the number of signals emitted is so high that the energy between one temporal step and the subsequent is comparable. It determines lower values of the index than expected. For this reason, we decided to process the data twice each time using a different filter setting: one without using amplitude filter (ACI no flt), and one using an amplitude filter of 2000 μV2/Hz (ACI flt).

Successively, the ACI values obtained for each frequency were summed on the LF, MF and HF bands.

2.2.3. Identification and description of the biophonic component

Biological sounds of the soundscape of the habitat considered were identified. Since the certain attribution of some sounds to specific biological source was not possible, we first acoustically characterized them. Subsequently, their daily patterns were described through the count of the signals.

To characterize the sounds, a subsample, with a good signal to noise ratio, was randomly selected for each sites. Using AvisoftSASlab Pro, the data selected were processed applying different filters in order to isolate the different sounds from the other biophonic components, without interfere with the signals characterized. In detail:

- Signals within the LF band: undersampling from 48 kHz to 11.025 kHz; high pass filter of 0.1 kHz; low pass filter of 0.5 or 0.8 kHz depending from the signals.
- Signals within the MF band: undersampling from 48 kHz to 11.025 kHz; high pass filter of 0.35 kHz; low pass filter of 1.5 kHz.
- Signals within the HF band: high pass filter of 1 kHz

The characterization was carried out using the pulse train analysis of AvisoftSASlab Pro, changing the hysteresis, the threshold, the time constant and the group time according of each signal analyzed. For each signal and train of signals we measured: duration (s), peak of frequency (Hz) and bandwidth (Hz) (for single signal); number of pulses (n) and pulse rate (n/s) (for train of signals).

To describe the daily trend of the principal acoustical components, we processed files of 2 min/hour collected during the three days over the new moon day. All the signals within this subsample were counted. The count process was carried out through both visual and acoustic inspection of files and by using the pulse train analysis of AvisoftSASlab Pro for frequent signals.

2.3. Statistical Data Analysis

The two acoustic habitats were compared using two metrics:

1. PSD values measured at the three frequency bands and averaged for each recording;
2. ACI values measured at the three frequency bands and averaged for each recording;

In detail, considering PSD values as dependent variables, linear mixed models (LMEM) (Bates et al. 2012) were applied to determine if the factors "habitat" (Posidonia, Sand), "daytime" (Day, Night), and "season" (Winter, Summer) affected the acoustic levels at the three frequency bands. The factors "sites" and "month of recording" were included as random factors. We excluded the recordings with the presence of boats to consider this factor separately. The best-fit model was selected by means of model averaging based on the information criterion (AIC). Validation graphs (e.g. residuals versus fitted values, Q–Q plots, and residuals versus the original explanatory variables) were analyzed in order to control possible model misspecification and the presence of outliers.

For ACI values, we compared only the data from summer recordings, when biological emissions of fish are present in both habitats. It was not possible to apply any linear mixed models because the low variability of the index in sandy habitat compared to the variability in the Posidonia habitat violates the homogeneity of variance criterion. As a consequence, the non-parametrical Kruskal-Wallis analysis, and post-hoc multiple comparisons test were carried out to compare the values of ACI of the two habitats (for each frequency band), in relation to the daytime.

The influence of wind speed on the PSD values, measured on the three frequency bands, was investigated using the linear regression model (LRM). In order to reduce the variability produced by the different exposure of the sites along the coast, firstly we carried out a LRM - using wind velocity as independent variable and PSD measured at LF band as dependent variable - splitting data for each site and for each wind direction. Basing on these results, we carried out the LRM on PSD values of all frequency bands considering only cases when the wind direction affects all sites.

The characterization of the biophonic components of the two habitats was carried out considering the mean value of the different parameters for each signals. We statistically compared the parameters of sounds present both in sandy and Posidonia habitats. We tested their acoustic variables for normally distribution (Shapiro - Wilk test); since data was not normally distributed, the U-Mann Whitney test was performed.

The percentage of files with the presence of vessel noise was compared between the habitats and between seasons on each site. These data were tested for normally distribution (Shapiro-Wilks test for each group of data). We applied T-test for independent data between two habitats and T-test for paired data to compare the data from each site between the seasons.

3. Results

In total, we recorded 1487 hours (450.7 during the winter and 1036.5 during the summer): 765.8 hours were collected in Posidonia habitat (254 during the winter and 515 during the summer) and 717.3 hours in sandy habitat (196 during the winter and 521 during the summer). In the Fig. 3 we showed the spectrogram of three days of recording (2 min/hour) collected during the three days over the new moon day. The different components are marked.

3.1. Acoustical habitat comparison through PSD and ACI

The best model (LMEM) selected using the information criterion (AIC), included habitat, daytime, season and their interaction as independent variables. In Tab. 2 the results of the models for each frequency band are shown. Significant differences resulted between the two habitats for each frequency band, considering the season and the daytime (Fig.4). In detail, during the winter (both night and day) sandy habitat was noisier (higher level of PSD) than Posidonia, considering LF and MF bands. During the summer, sandy habitat was still noisier than Posidonia in LF band, but Posidonia habitat had higher level of PSD in MF band. The HF band was noisier in Posidonia than in sandy habitat both during winter and summer, during the day and the night time.

Considering the ACI values, the efficiency of the index in term of biophonic amplification, resulted different using or not using the filter for each band considered. The daily pattern of ACI (with and without filter) was plotted for each band (Fig. 5). At the low frequency (LF), where the continuous noise of boats, wind and wave is predominant, the use of the filter reduced the power of attenuation of these sounds. At medium and high frequency bands, where the effect of geo- anthropophonic noise is not so strong and the biophonic choruses are more intense, the use of the filter was essential to amplify the biophonic component of the soundscape. Basing on these results, we carried out the subsequent analysis using different settings of ACI in relation to the band involved. Comparing ACI values, the two habitats presented significant differences for all frequency bands both during day and night. In particular, Posidonia habitat showed higher biophonic activity than sandy habitat (Kruskal-wallis test: LF band χ^2=1673.5, df=3, p<0.001; MF band χ^2=958.71, df=3, p<0.001; HF band χ^2=444.2, df=3, p<0.001) (Fig. 6).

Considering the effect of wind speed on PSD values, we reported the results of LRM using data split for each site and for each wind direction (Tab.3). Only significant correlations have been reported in Fig.7. The highest significant angular coefficients (β) resulted for the south wind direction within each site. In Tab.4 the results of the models using the cases when the wind direction was from south, are presented. In the LF and MF bands, the PSD resulted positively correlated with the wind speed in both habitats, but the angular coefficient (β) of the regression line resulted higher in sandy habitat than in Posidonia meadow. In the HF band, the PSD was not correlated with the wind speed in both habitats.

3.2. Identification and description of the biophonic component

Through the visual and acoustic analysis of the spectrograms, different biological elements of the soundscape were identified as characterizing the two habitats. In Table 5, the acoustic features (both spectral and temporal) of signals are shown.

Considering the three bands:

- Low Frequency band (Fig. 8 D-E):

In this band, we found fish sounds (probably emitted by one or two similar species) (LF fish) made up of train of pulses. These signals were recorded both in Posidonia and in sandy habitats, but they showed some different spectral and temporal characteristics (respectively Fig. 8-D and 8-E; Tab. 5). In particular, the duration of pulses (Z=-7.85 p<0.001) and the peak of frequency

($Z=-10.11$ $p<0.001$) were significantly different, with longer duration and higher frequency peak in sandy bottoms. Other parameters did not show any differences among the habitats (Bandwidth of pulse: $Z=-1.0$ $p>0.05$; Number of pulse: $Z= -1.01$ $p>0.05$; Pulse Rate: $Z=-0.6$ $p>0.05$).

- Medium frequency band (Fig. 8 B-C):

Biological sounds dominated only in Posidonia habitat also in this case. Two different types of sounds were identified: tonal (MF fish Fig. 8 B) and impulsive sounds (MF pulse Fig. 8 C).

- High frequency bands (Fig. 8 A):

Snapping shrimp pulses dominated the high frequency band, only in Posidonia habitat.

In Fig. 9 the temporal distribution of biological sounds counted along the day is showed. The LF fish sounds were recorded only during the summer in both habitats, and they showed a circadian pattern, with pitches during sunrise and sunset. The MF fish sounds were recorded mostly in Posidonia habitat during the night in summer period, with the presence of chorus at sunset. During the winter, they were sporadic and no choruses were recorded. In sandy habitat, we found only few tonal signals during summer, but no evidence of chorusing either pattern was present. The MF pulse sounds were recorded only in Posidonia habitat during both winter and summer but only during the daytime. Finally, the snapping shrimp pulses were recorded only in Posidonia habitat in both winter and summer, showing pitches at sunrise and sunset. An increase during the summer was present.

3.3. <u>Habitat comparison through number of boats</u>

Focusing on the anthropogenic noise, the percentage of presence of boats (number of 10 minutes files with the presence on vessel noise on the total number of recordings) for each site during both winter and summer is shown in Fig.10. The percentage of boats counted visualizing the recordings spectrograms was lower in Posidonia than in sandy habitat ($T=-2.7$; $p<0.05$) and increased during the summer ($T=-3.2$; $p<0.05$).

4. Discussions

The main goal of this study is to analyze the differences in the soundscape of two of the most typical habitats of the Mediterranean Sea: the Posidonia and the sandy habitats. For the passive acoustic comparison, we decided to consider three different frequency bands. The two environments showed different characteristics both using PSD and ACI values and by analyzing the biological sonic component.

Focusing on the results of acoustic energy data, the comparison between the two habitats showed differences in all the frequency bands considered.

The sandy habitat presented higher values of power spectral density at the low frequency band (LF), both in summer and in winter compared to Posidonia habitat. Since this result was obtained excluding the files with the presence of boats, it could be due to both a different biotic sound activity, or to a different response to the geophonical component. The results of the Acoustic Complexity Index indicated that the biophonic component is not responsible for higher values of PSD in sandy habitat. Indeed, we found that Posidonia habitat, during the summer daytime, showed higher values of index. The analysis of geophonical component, instead, revealed a higher angular coefficient of the linear relation among PSD and wind velocity, showing a stronger noise increase at the low and medium frequency bands in sandy habitat. Therefore, our data suggested that the geophysics component have different effects on the soundscape, due to the physical structure of the two habitats. In terrestrial environment, the presence of vegetation is recognized to be an important factor to reduce noise energy (Embleton 1963, Aylor 1972, Kragh 1981). In marine environment, the seagrass photosynthetic activity produces free gas contained within the aerenchyma and bubbles on the surface of the plant tissue. This phenomenon affects the local sound propagation and backscattering (Clay & Medwin 1977, Hermand et al. 1998, Wilson & Dunton 2009, Wilson et al. 2013), determining a unique acoustic footprint, commonly used to map and characterize the submerged macrophyte (Wilson et al. 2013). As consequence the physical structure of Posidonia meadow, could attenuate also the noise produced by geophonical factors, making Posidonia habitat a potential acoustic refuge for marine species.

The power spectral density measured at MF band showed higher values of energy in sandy habitat compared to Posidonia only during the winter. During the winter, these results could be generated by geophonic components that strongly affects sandy habitat. Instead, during the summer, Posidonia soundscape is more affected by biophonic components. This result is confirmed by ACI data measured during summer: the index captures the acoustic activity of fishes during the night and the presence of impulsive signals (at low and medium frequency) during the daylight hours.

Considering all frequency bands results, the application of the Acoustic Complexity Index in this paper has demonstrated to be a useful proxy for the biotic acoustic activity. The choice to split the analysis basing on the frequency bands of the principal biologic components recorded, helped the results interpretation. Harris et al. (2016) compared different acoustic indices relating these to reef fish abundance and diversity. They found a strong correlation between the ACI and species richness and evenness. Also, Stateerman et al. (2014) and Bertucci et al. (2016) used successfully this index to study and compare different soundscapes. We found that the use of this index should take into account different settings that can strongly affect the results. The application of an intensity filter to the data or not, can help to discriminate far and fore acoustic fields, leading to amplify the strongest ecoacoustic events (Farina et al. 2016) or to attenuate the non biotic component of the soundscape. Harris et al. (2016) do not consider this parameter and the index applied on those conditions does not show these problems on his study. It could be because Harris et al. (2016) correlated the index values with other species assemblage diversity indices, not with the number of signals. Moreover, the reason of different results could be found on different habitats analyzed and in temporal resolutions used. Kaplan et al (2015) and Buscaino (2016) found that ACI values result to be lower than expected when the density of calling activity is too high. Through a differential frequency band approach, we decided to adapt the computation of the index using the filter only in those bands not strongly affected by geophonical component. This method allowed to amplify the chorus of MF fishes and HF snapping, but determined the attenuation of the less frequent and less intense pulses in the MF band. Until now, in marine environments single metrics were separately considered to describe habitat complexity. In this study, more methodologies have been carried out and developed together for the first time, to unroll the acoustic complexity of different habitats.

Focusing on the sonic biotic component, different sounds of marine animals have been identified as principal elements of soundscape of these habitats. They occupy differently the acoustic spectrum, reducing the overlap of signals along time and/or frequency dimensions.

At LF band, two types of signals have been recorded and characterized. They showed typical acoustic properties and daily patterns belonging to fishes of the *Ophididae* family, in particular *Ophidion rochei* (Parmentier et al. 2010). Within the vocalizations of this species, the acoustic features (in amplitude and frequency band), the pulse rate and the peaking during dusk and dawn are distinctive. *Ophidion rochei* is a Mediterranean species that typically lives in the sand, but its presence in Posidonia habitat has been also recorded (Keskin 2007). In this study, we obtained that the frequency characteristics of these fish sounds are different between the two habitats: even if the bandwidth is comparable, the peak of frequency in sandy habitat is higher of about 20 Hz. The MF band (over 0.5 kHz) is not used by other species in sandy habitat, while in Posidonia is totally saturated by the presence of other fish sounds. Therefore, we can hypothesize that the differences found in the frequency characteristics could be due to an acoustic adaptation to the two habitats. However, since this variation, a certain attribution to one or more species needs further studies.

The MF band was occupied by biological signals only in the Posidonia environment. We obtained acoustic presence of pulses along all daytime and of fish tonal sounds during summer night (with peak at the dawn). The short pulses were present irregularly during the daytime in winter and summer. They totally disappeared during night. The correct attribution of these sounds is still not clear. In our knowledge, a fish species that emit sounds following this seasonal and daily pattern have not been described before. Also, we can exclude invertebrates as source of these signals, since generally, their acoustic activity, even if present along all the year, shows increase during the night and peaks during dusk and dawn (Radford et al. 2008b, 2010, Buscaino et al. 2011, 2016). The daily pattern of these pulses suggests an association with a phenomenon linked to presence of light. Therefore, one possible explanation could be the connection with photosynthetic or decomposition phenomena within Posidonia meadows. Hermand (2004) suggests the production of photosynthesis bubbles from Posidonia leaves, and their collapse could produce impulsive sounds (Versluis et al. 2000, Pettit et al. 2015). Further studies are necessary to prove this correlation.

Regarding fish tonal sounds in MF band, as far as we are aware, these signals have still not been described. They show frequency characteristics and daily pattern similar to *Therapon theraps* as recorded in the Great Barrier Reef in Australia and Arabian Sea (Mahanty et al. 2015, McCauley & Cato 2000). *Therapon theraps* is typical from Indo-West Pacific and Australian waters, but individuals of this species and of *Therapon jarbua*, have been reported in the Mediterranean Sea along the Aegean (Minos et al. 2012), Adriatic (Lipej et al. 2008) and Israeli coasts (Golani & Appelbaum-Golani 2010). They are considered invasive species, probably arrived in Mediterranean through a Lessepsian migration. The intense and regular occurrence of these broadband sounds, along summer days, suggests a stable presence of this species along the Mediterranean coasts. Invasive species could determine changing in the acoustic community acting as selective pressures on the native species (Farina et al. 2013). This could lead to variation in communication features of local species in order to improve the transmission of information (Acoustic Adaptation Hypotesis, Morton 1975). The absence of visual census monitoring in this area does not allow confirming the presence of this specie in this part of the Mediterranean Sea. However, our results draw attention to the importance of the acoustic method to reveal any rapid changes within an ecosystem.

The HF band is occupied by impulsive signals of snapping shrimps only in Posidonia habitat. They follow circadian and seasonal rhythms as already described along all temperate habitats (Radford et al. 2008b, Bohnenstiehl et al. 2016, Buscaino et al. 2016). Their acoustic activity is connected to different abiotic factors, such as dissolved oxygen concentration (Watanabe et al. 2002) and water temperature (Bohnenstiehl et al. 2016). Regarding the seasonal activity, the summer increase in the number of snaps occurs during the night and both dawn and dusk chorus, but not during the rest of the day. Comparing this result with the number of snapping shrimps counted in Lampedusa area (Buscaino et al. 2016), a different trend of the snapping activity during the winter season is evident. Buscaino et al. (2016) in Lampedusa area, showed that the number of snapping signals recorded during January and February did not present a strong circadian trend as shown here. This could be due to the presence of the upwelling wind, typical of the northern sector of Sicilian Channel (Patti et al. 2010), that determines less variation of water temperature along the year. However, we cannot exclude that the differences found among these two areas of the Sicily Channel could also be due to a different species composition, since the habitat monitored around Lampedusa Island is more heterogeneous due to the presence of a mix of *Posidonia oceanica* patches, sand and rocks.

Mostly in Posidonia habitat where different fish sounds share the same temporal dimension, all the biological components recorded partition different frequency spaces, supporting the idea of the evolutionary adaptive function of the acoustic niche (Krause 2012, Farina 2014).

Finally, we found that the anthropogenic activity affects strongly both these coastal habitats but in a different way. The percentage of boats recorded in sandy habitat is higher than in Posidonia and it increases during the summer. The explanation could be found in the use of the area by recreational boats. Clear and bright waters with sandy bottom result to be more attractive than the dark waters of Posidonia bottom. This is supported by the fact that both habitats located near the more touristic area present the highest percentage of boats. Anyway, considering all the daytime, in summer period, the presence of boats was identified in the 30% of the recordings. It means that, considering the general diurnal vessel activity, it can reach the 60% during the daylight summer days.

Noise pollution can affect marine organisms' acoustic communication through auditory masking (in which the perception of one sound is affected by the presence of another sound) (Clark et al. 2009).

In particular, the noise generated mainly by boats is able to determine this occurrence. The sounds produced by engines and related vibrating accessories elements of the boats, in fact, occupy the acoustic space reserved for the acoustic communication among fish (Brumm & Slabbekoorn 2005, Wysocki & Ladich 2005, Vasconcelos et al. 2007, Codarin et al. 2009) and, probably, crustaceans. Our results evidence how the presence of boats is able to 'camouflage' the transmission systems of intra- and inter-specific information between these organisms, limiting or preventing some fundamental bio-ecological processes during the animal life.

5. Conclusion

The soundscape analysis confirmed to be a key approach to understand ecological processes and habitat discrimination. The acoustic information transmitted by different communities can be received by species helping them on orientation processes.

Our work demonstrated that the Posidonia habitat is not only richer than sandy one in term of number of species, but it is also acoustically richer in term of biophonic components. The presence of shelters and food opportunities makes this habitat crucial for species survival and the acoustic activities of sonic species revealed the importance of this environment. The bare sandy bottom creates a particular habitat where only few vocalizing species are adapted to live and reproduce. The human pressure, in term of noise, in both coastal habitats is very alarming mostly during summer period when the recreational boats traffic increases and the resulting noise pollution determines an almost constant disturb along all days. Anthropogenic noise impact negatively on the marine organisms in different way (Clark et al. 2009, Slabbekoorn et al. 2010, Filiciotto et al. 2016) and further studies should be conducted in posidonia and sand shallow water Mediterranean ecosystems to quantify and manage this negatively effects on marine organisms.

Therefore, the acoustic monitoring method could represent a useful and not-invasive tool for the evaluation of the human pressure on the bio-ecological and conservation factors in the marine ecosystems, in order to achieve a Good Environmental Status (GES) defined by the Marine Strategy Framework Directive 2008/56 CE. Posidonia soundscape can be considered as a cue for the conservation status of this habitat, and must be monitored and evaluated for management purposes, especially when new anthropogenic activities are planned in the area.

6. Acknowledgements

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7. Foundings

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8. Author contributions statement

M.C. conceived the study, collected data, performed analysis and paper writing; F.C., F. F., E. P., G.B. and R.G., took part to the data collection, participating to the paper writing and PSD analysis. I. P. performed acoustic processing to the counting processes; G.B. and A.F. conceived, founded and guided the study, the results interpretation and participating to the paper reader.

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10. Figures

Figure 1 Study area. In green Posidonia patches, in light yellow sandy patches

Figure 2 Schematic representation of the deployment system

Figure 3 Three continuous days of recordings (2 minutes/ hour) during both season, in Posidonia habitat (left side) and sandy habitat (right side). The dashed white lines indicate the night time. The letters indicate the different components of the soundscapes. In detail, SS: Snapping shrimps impulses; MF: MF fish choruses; LF: LF fish choruses; I: MF impulses; W: Waves; R: Rain; B: Boat noise. N.B. The lower intensity band between 3500 and 5500 Hz is due to non-linear response of Digital Acquisition Card.

Figure 4 Box plot (central line: median; box limits: first and third quartile; whiskers: minimum and maximum) of PSD measured on two habitats, during daytime of the different seasons. The star represents the significant differences ($p<0.05$) between habitats.

Figure 5 Mean ± Standard Error of ACI values, measured without using an amplitude filter (dark green - posidonia habitat; dark yellow sandy habitat) and using an amplitude filter (light green - posidonia habitat; light yellow sandy habitat)

Figure 6 Box plot (central line: median; box limits: first and third quartile; whiskers: minimum and maximum) of ACI (without amplitude filter at LF band, and with amplitude filter in MF and HF bands) measured on two habitats, during summer daytime. The star represents the significant differences ($p<0.05$) between habitats.

Figure 7 Scatterplot of PSD values and wind speed, when the direction of wind was from south (dark green - posidonia habitat; dark yellow sandy habitat) and from south-western (light green - posidonia habitat; light yellow sandy habitat)

Figure 8 Spectrogram, waveform and amplitude spectrum of the principal biological signals recorded. HF Band: A. Impulsive signals of snapping shrimps; MF band: B. Tonal fish sound C. Impulses; LF band: D. train of fish impulses recorded in Posidonia Habitat E. train of fish impulses recorded in Sandy Habitat. N.B. The waveform and the amplitude spectrum are computated on isolated sounds, see the MS for details of filters applied.

Figure 9 Mean and standard error of number of signals counted during three days of recording in all sites for both season. HF Band: averaged number of impulsive signals of snapping shrimps; MF band: averaged number of tonal fish sound (barr) and impulses (point with connection line); LF band: averaged number of train of fish impulses

Figure 10 Percentage of boats measured on each site of recordings during both season. The star represents the significant differences ($p<0.05$) between seasons (for each site) and habitats.

11. Tables

Table 1 Characteristics of the recording sites.

Table 2 Results of selected LMEM models using PSD values as dependent variables and including habitat, daytime, season and their interaction as independent variables. Sites and moths of recordings were included as random factors.

Table 3 Results of regression linear model using wind velocity as independent variable and PSD measured at LF band as dependent variable, splitting data for each site and for each wind direction. In bold the significant models.

Table 4 Results of regression linear model using southern wind velocity as independent variable and PSD measured at LF, MF and HF bands as dependent variables.

Table 5 Mean ± SE of signals features measured for the principal biological sounds individuated on each frequency band

Figure 01

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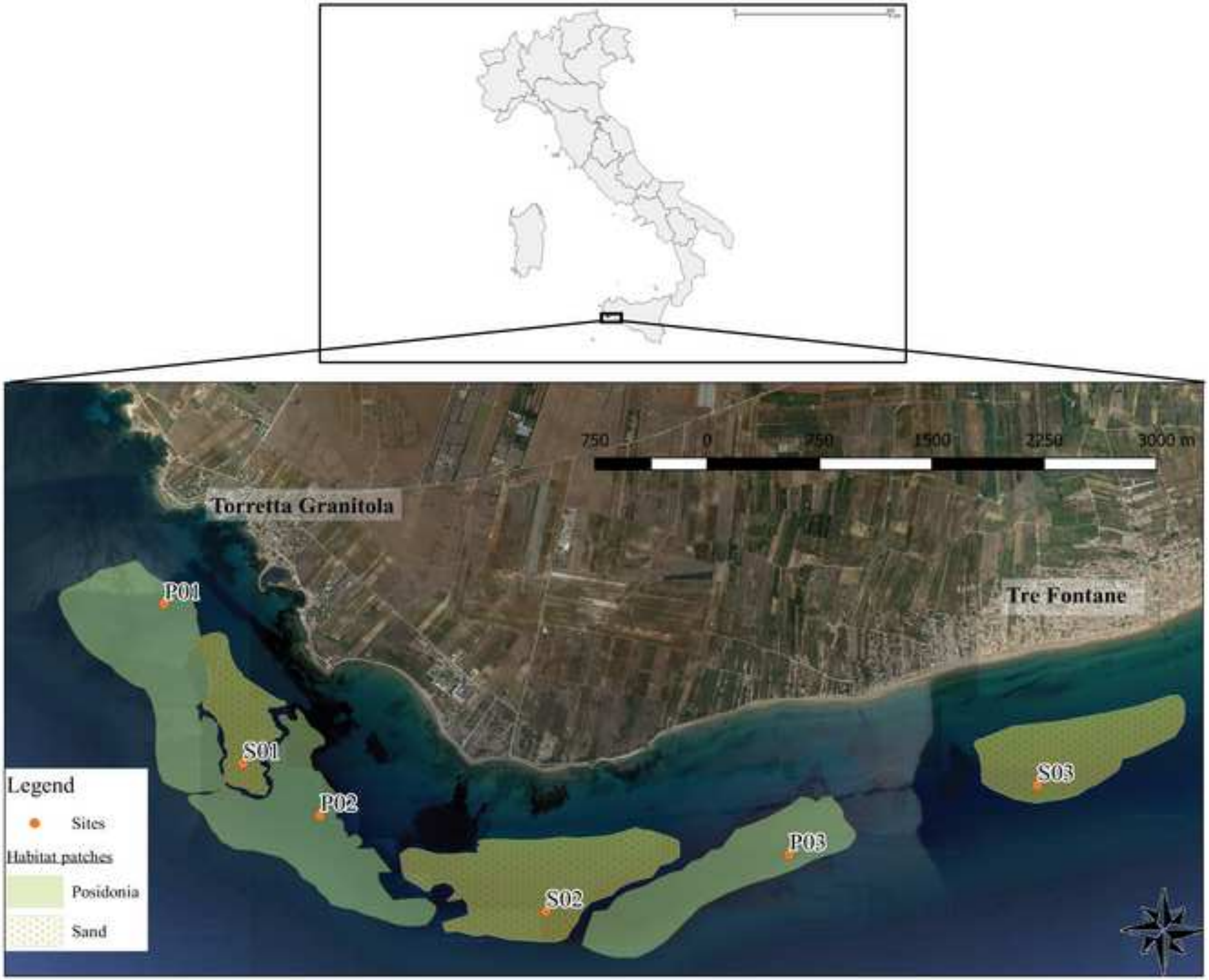

Figure 02

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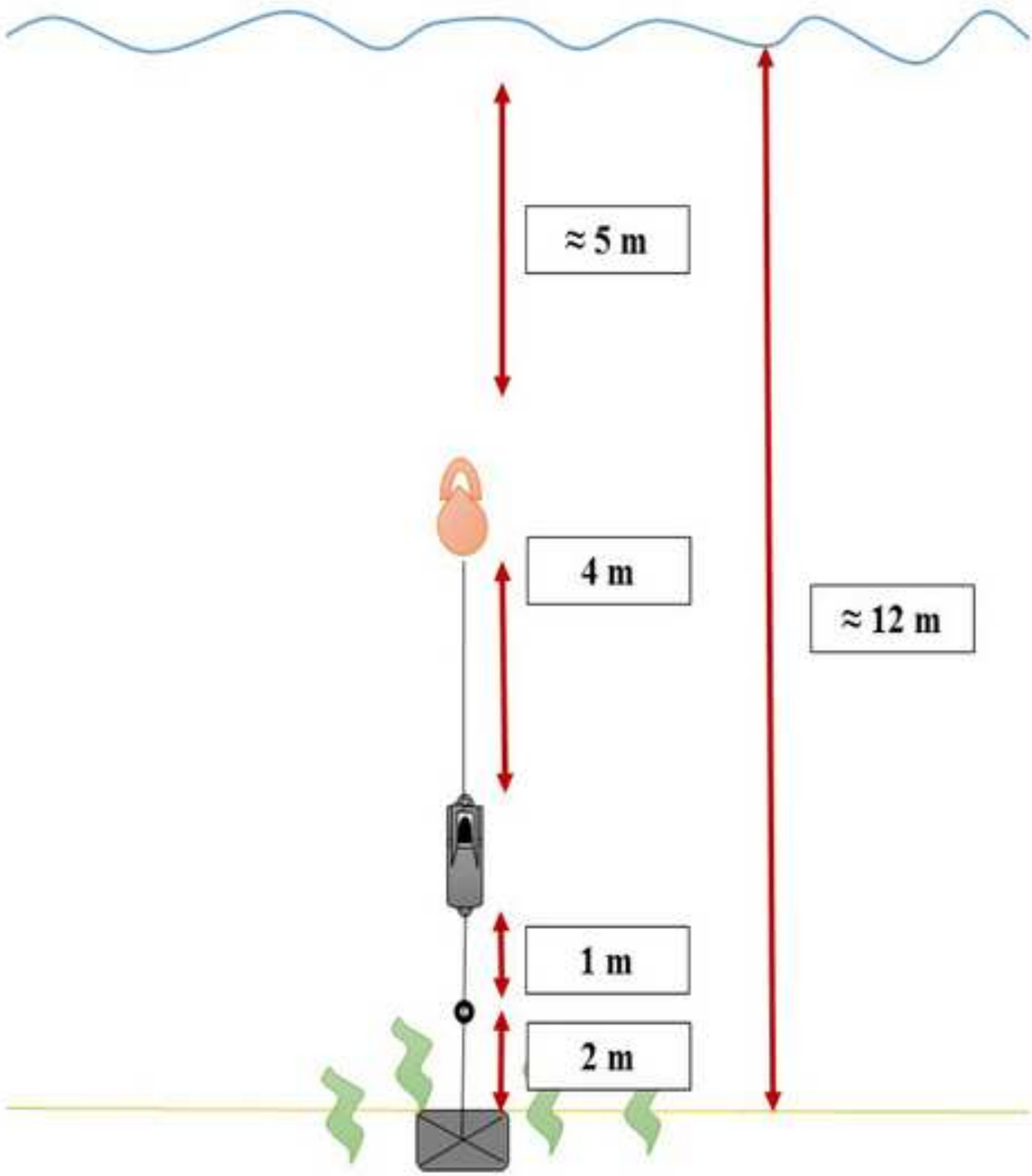

Figure 03

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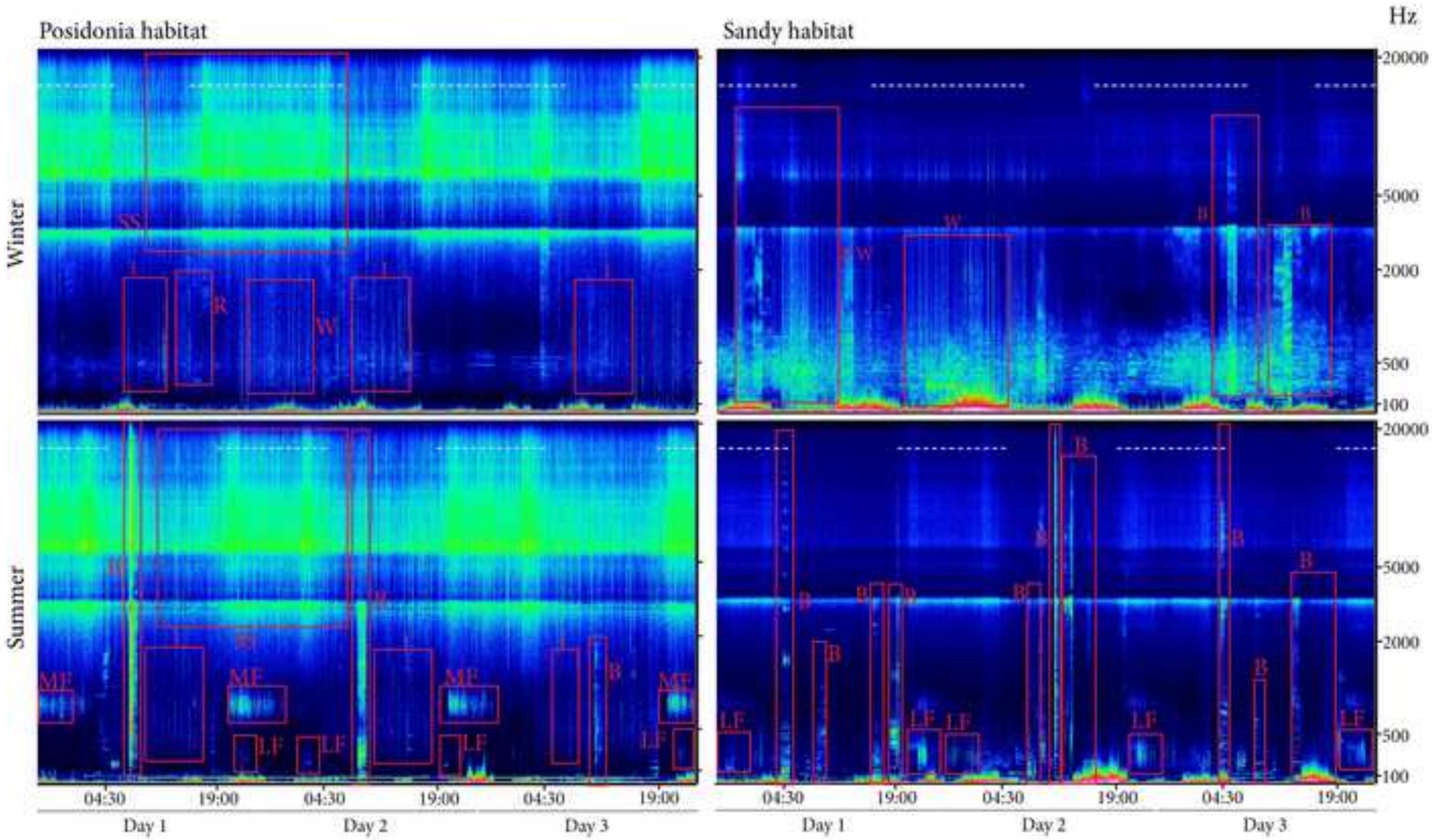

Figure 04
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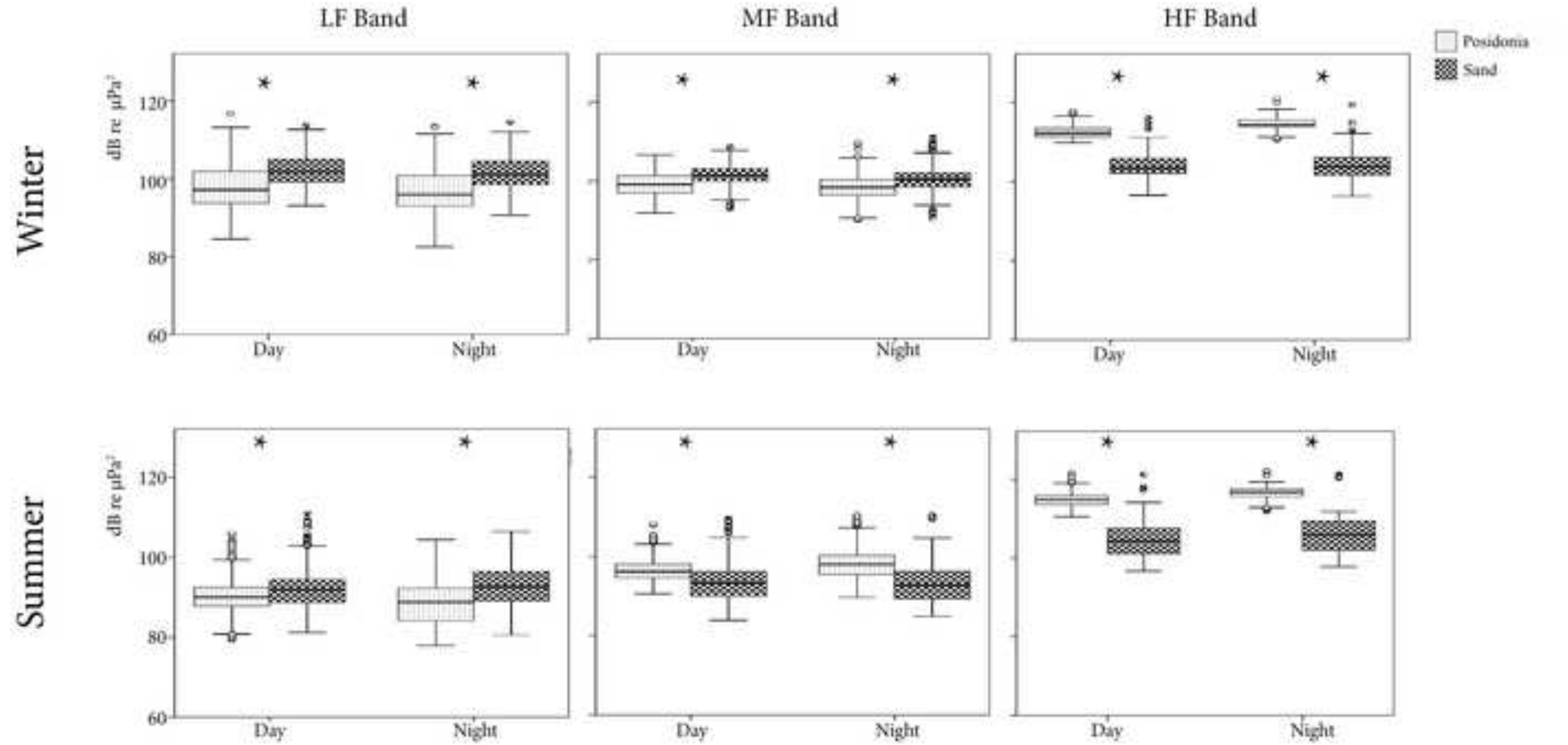

Figure 05

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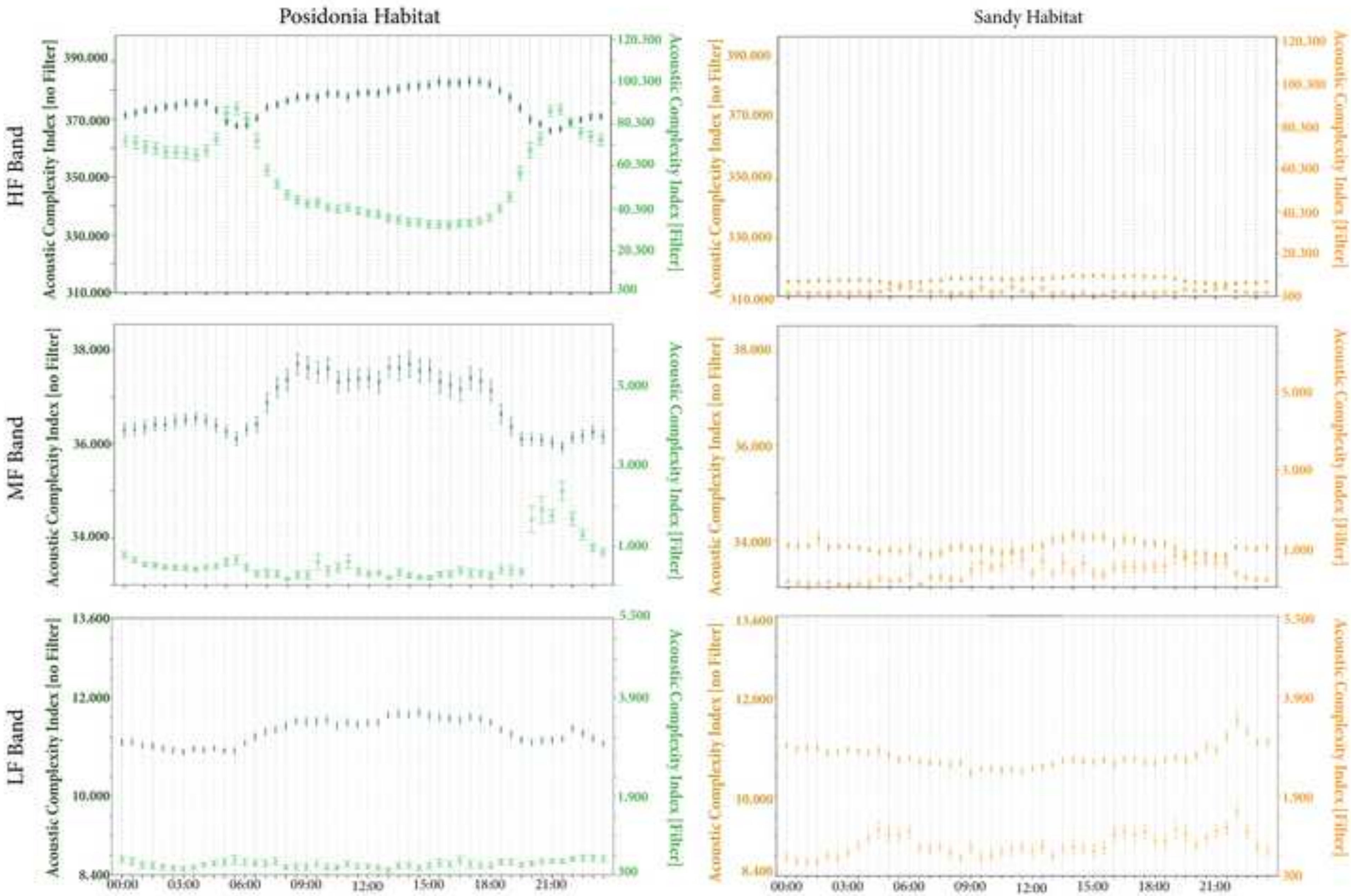

Figure 06
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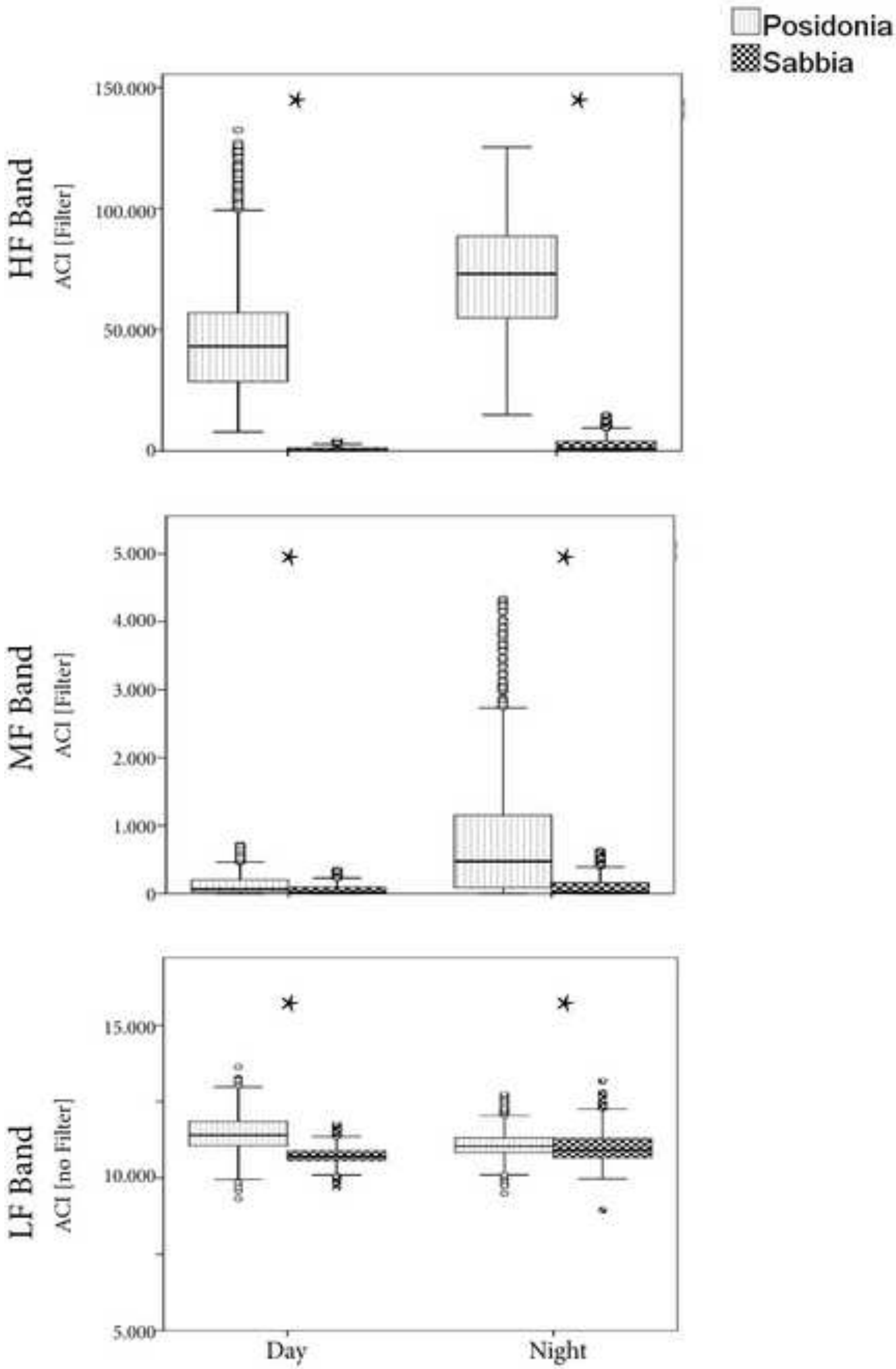

Figure 07

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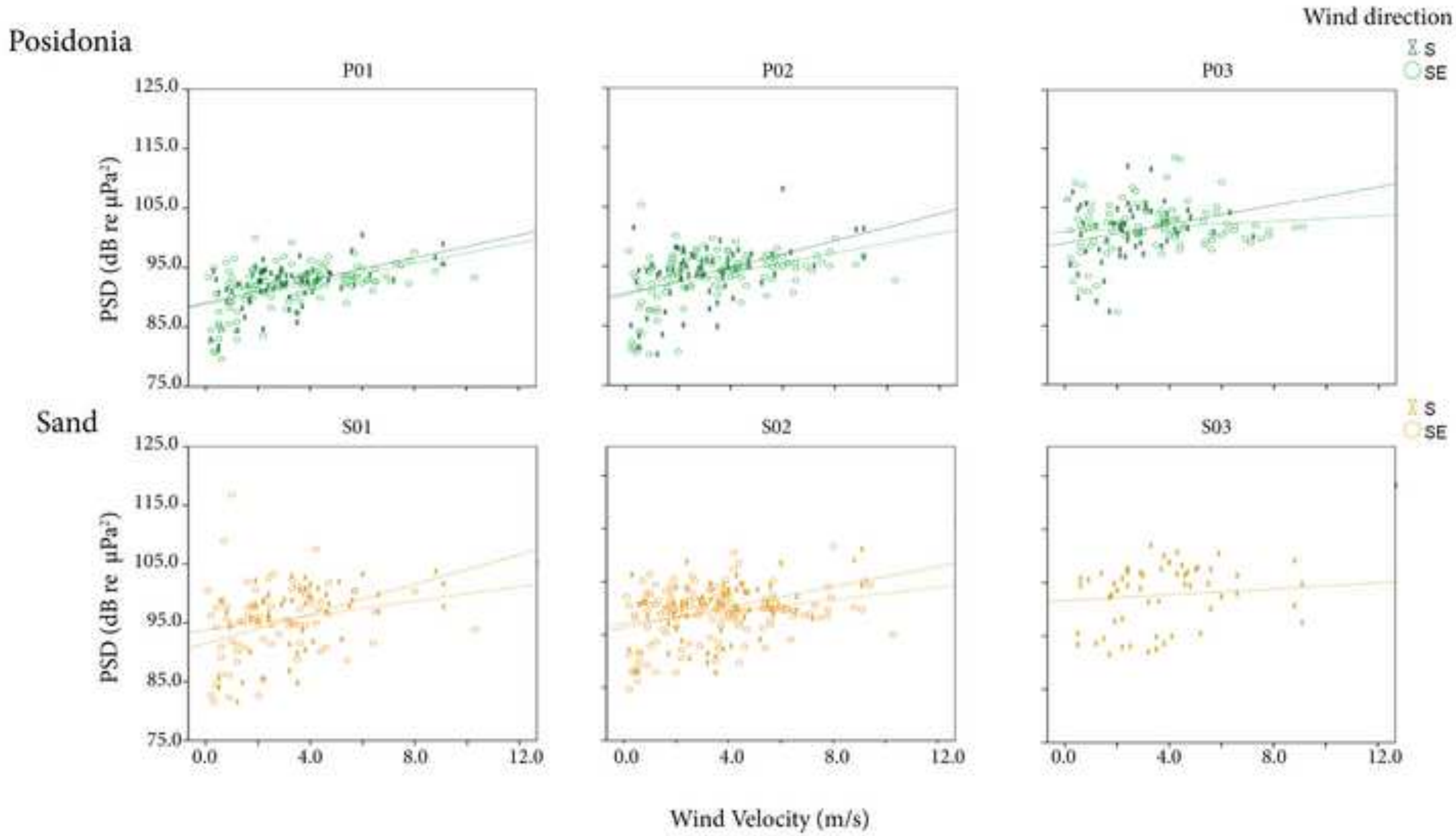

Figure 08
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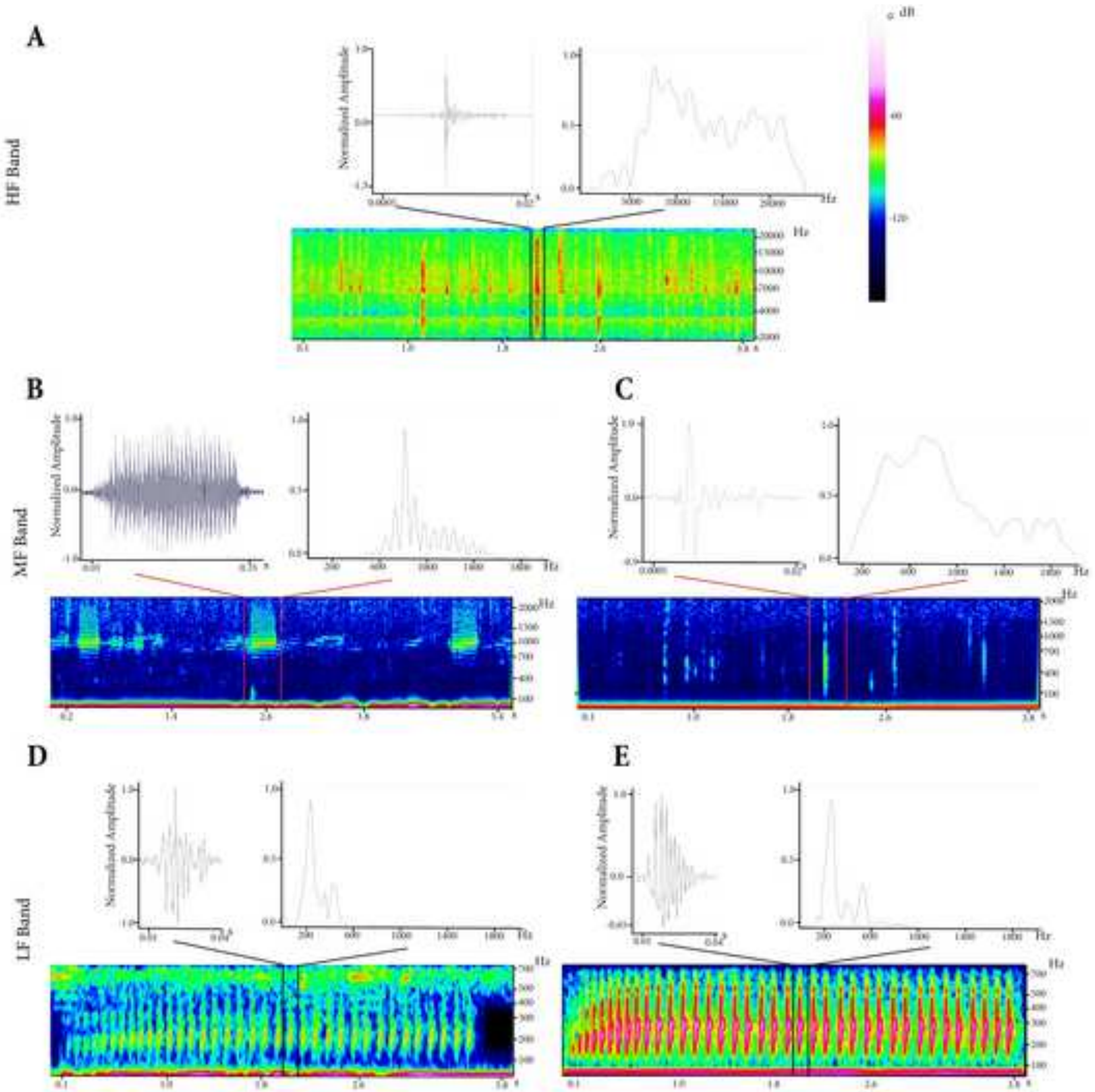

Figure 09
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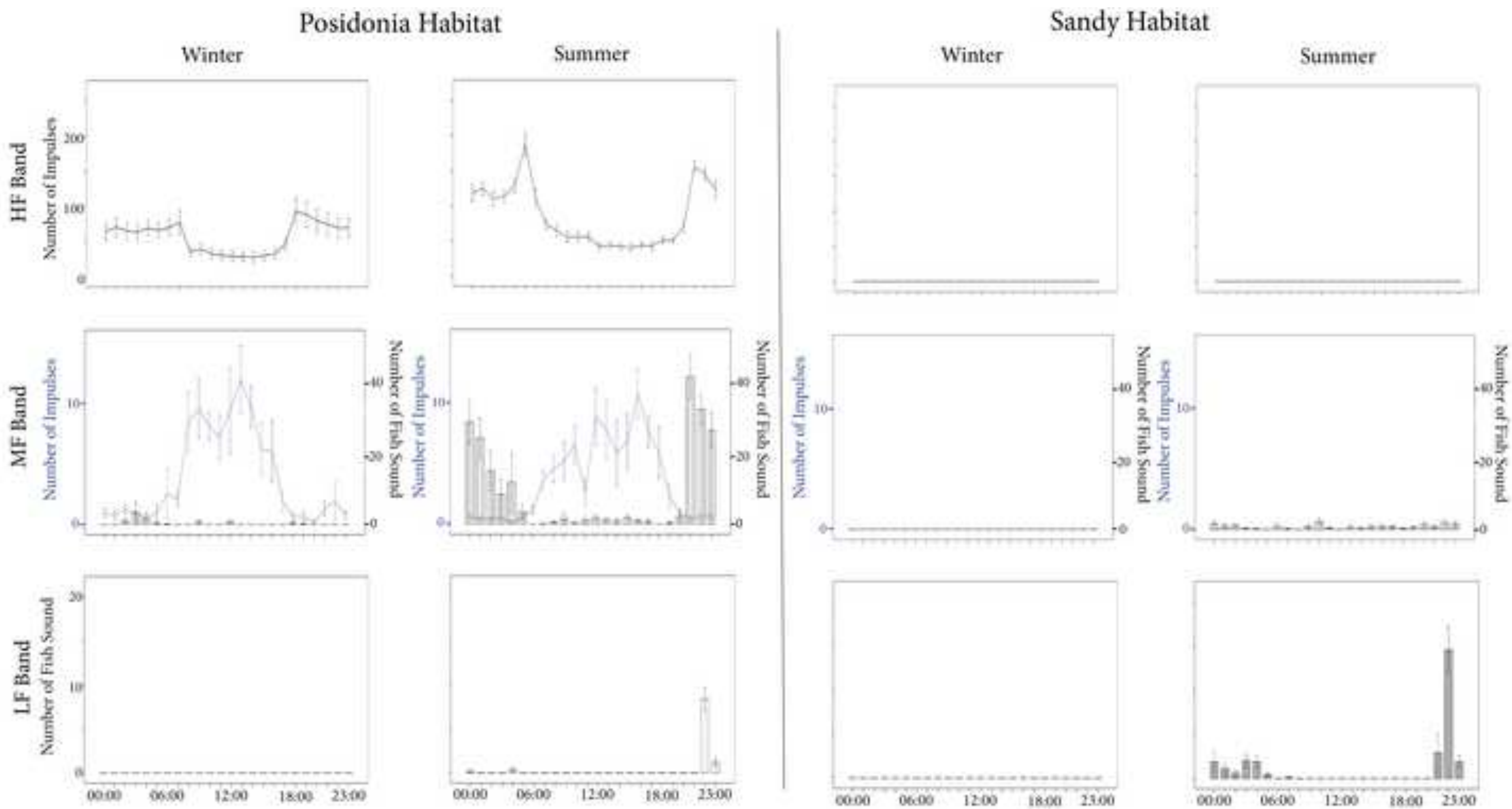

Figure 10
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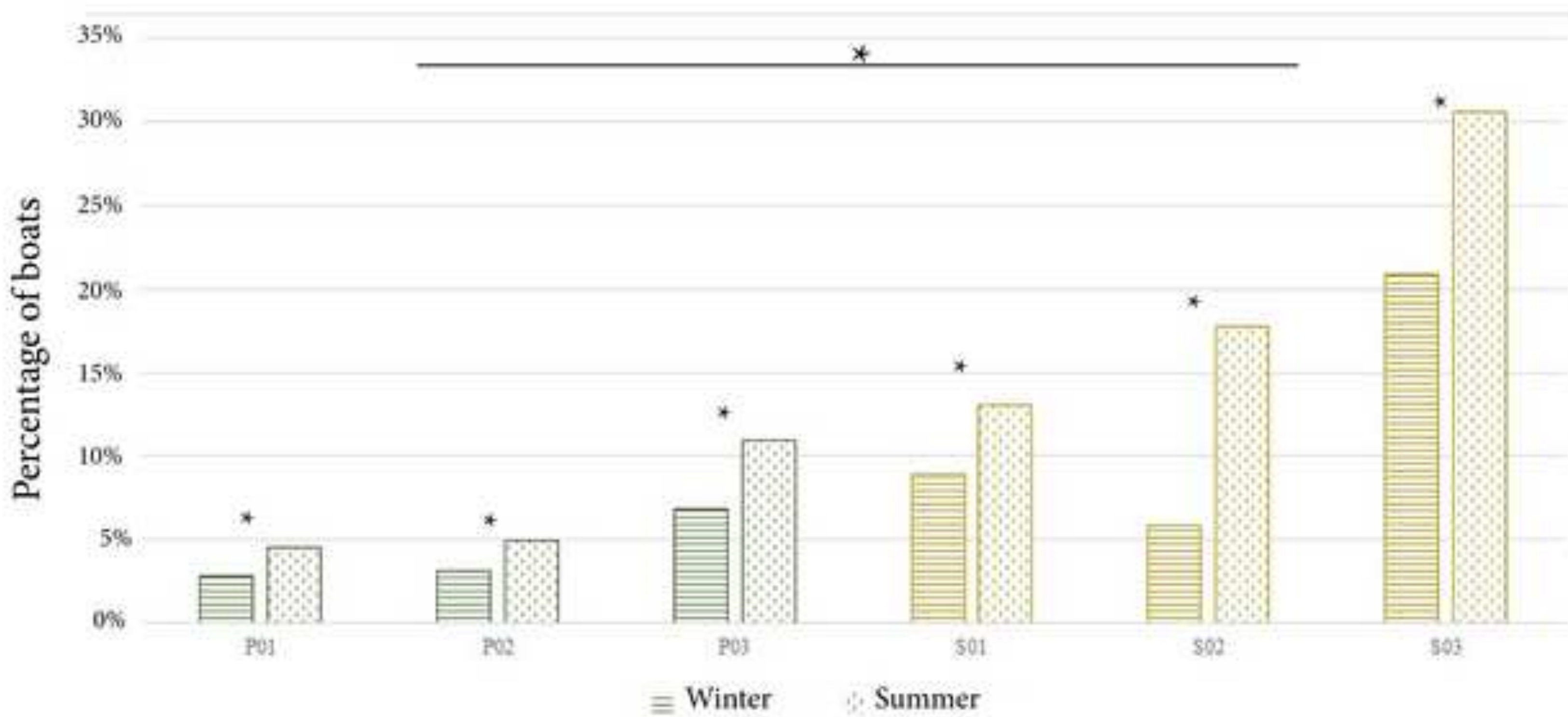

Table 01

Habitat	Site	Latitude	Longitudine	Bathymetry (m)	Distance from the coast (m)
Posidonia	P01	37°34'218 N	12°38'976 E	12,9	530
Sand	S01	37°33'611 N	12°39'357 E	12,4	450
Posidonia	P02	37°33'400 N	12°39'672 E	12,5	973
Sand	S02	37°33'058 N	12°40'723 E	11,9	930
Posidonia	P03	37°33'259 N	12°41'839 E	12,9	725
Sand	S03	37°33'542 N	12°42'967 E	12,1	722

Table 02

		Estimate	Std.Error	df	t value	p
PSD - HF band	Intercept	114.68	1.67	4.0	68.79	<0.001
	Habitat (Sand Vs Posidonia)	-10.64	2.36	4.0	-4.52	<0.05
	Daytime (Night VS Day)	1.77	0.06	7298.0	27.39	<0.001
	Season (Winter VS Summer)	-2.24	0.12	7.0	-17.95	<0.001
	Habitat* Daytime	-0.44	0.10	7303.0	-4.44	<0.001
	Habitat*Season	2.23	0.15	93.0	15.13	<0.001
	Season*Daytime	0.40	0.12	7302.0	3.41	<0.001
	Habitat*Season*Daytime	-1.59	0.18	7302.0	-9.02	<0.001
PSD - MF band	Intercept	96.39	0.89	5.0	108.63	<0.001
	Habitat (Sand Vs Posidonia)	-3.59	0.65	4.0	-5.56	<0.01
	Daytime (Night VS Day)	1.71	0.12	7300.0	13.72	<0.001
	Season (Winter VS Summer)	3.37	1.22	3.0	2.77	0.06
	Habitat* Daytime	-1.55	0.19	7300.0	-8.14	<0.001
	Habitat*Season	4.87	0.32	6486.0	15.30	<0.001
	Season*Daytime	-2.57	0.22	7300.0	-11.52	<0.001
	Habitat*Season*Daytime	1.27	0.34	7300.0	3.75	<0.001
PSD - LF band	Intercept	90.97	1.96	6.0	46.34	<0.001
	Habitat (Sand Vs Posidonia)	0.76	2.39	4.0	0.32	0.77
	Daytime (Night VS Day)	-1.86	0.15	7300.0	-12.27	<0.001
	Season (Winter VS Summer)	7.60	1.60	3.0	4.75	<0.05
	Habitat* Daytime	3.12	0.23	7300.0	13.42	<0.001
	Habitat*Season	1.63	0.39	6681.0	4.20	<0.001
	Season*Daytime	0.87	0.27	7300.0	3.19	<0.01
	Habitat*Season*Daytime	-2.94	0.41	7300.0	-7.15	<0.001

Table 03

	Site	Wind Direction	β	R^2	t	p		Site	Wind Direction	β	R^2	t	p
Posidonia	P01	N	.238	.021	1.62	.109	*Sand*	S01	N	.243	.011	.72	.475
		NE	.093	.001	.19	.850			NE	-1.373	.026	-1.01	.318
		E	-.232	.004	-.39	.702			E	.320	.003	.30	.767
		SE	**.204**	**.043**	**2.30**	**.023**			**SE**	**.572**	**.090**	**2.65**	**.010**
		S	**.529**	**.214**	**4.30**	**<0.001**			**S**	**.894**	**.237**	**3.86**	**<0.001**
		SW	.623	.067	1.22	.235			SW	.613	.021	.50	.624
		W	-.302	.014	-.73	.468			W	-.700	.018	-.63	.535
		NW	-.097	.003	-.56	.575			NW	-.654	.018	-.66	.514
	P02	N	.301	.032	1.87	.064		S02	N	.524	.038	1.82	.072
		NE	-.172	.002	-.32	.748			NE	-1.820	.050	-1.54	.130
		E	-.284	.005	-.44	.660			E	-.606	.007	-.52	.609
		SE	**.250**	**.060**	**2.73**	**.007**			**SE**	**.885**	**.231**	**6.49**	**<0.001**
		S	**.665**	**.233**	**4.51**	**<0.001**			**S**	**.993**	**.227**	**4.09**	**<0.001**
		SW	.369	.018	.61	.547			SW	-2.742	.170	-1.87	.079
		W	.176	.006	.47	.639			W	-.190	.001	-.12	.907
		NW	-.165	.006	-.79	.432			NW	.063	.000	.17	.863
	P03	N	.140	.004	.50	.616		S03	N	.738	.046	1.88	.063
		NE	-.602	.018	-.95	.348			NE	-2.646	.026	-.94	.355
		E	.427	.003	.35	.731			E	-.771	.010	-.58	.564
		SE	**.497**	**.184**	**4.49**	**<0.001**			SE	.441	.036	1.80	.076
		S	**.817**	**.121**	**2.75**	**.008**			**S**	**1.033**	**.130**	**2.89**	**.005**
		SW	-.130	.001	-.13	.901			SW	.000	.000	.00	1.000
		W	1.779	.098	1.68	.106			W	-1.046	.016	-.55	.592
		NW	.402	.022	1.21	.229			NW	.383	.016	.94	.349

Table 04

	Habitat	β	R^2	t	p
HF band	*Posidonia*	-0.09	0.06	-1.12	0.26
	Sand	0.11	0.05	0.92	0.35
MF band	*Posidonia*	0.55	0.11	5.00	< 0.001
	Sand	0.99	0.18	5.90	< 0.001
LF band	*Posidonia*	0.71	0.05	3.24	< 0.001
	Sand	0.96	0.10	4.36	< 0.001

Table 05

		HF Band	MF Band		LF Band
		HF Snapping Impulses	*MF Fishes*	*MF impulses*	*LF Fishes*
Posidonia	*Duration of pulse (sec.)*	0.0025 (± 0.00009)	0.15 (± 0.08)	0.0034 (± 0.0004)	0.0054 (± 0.0016)
	Peak frequency (Hz)	8971.81 (± 4347.48)	840 (± 75.37)	724.14 (± 315.64)	210.50 (± 36.41)
	Bandwidth of pulse (Hz)	21788.87 (± 2253.97)	762.85 (± 266.2)	2088.92 (± 313.96)	400.14 (± 112.04)
	Number of pulse of train (N)	-	-	-	30.0 (± 8.33)
	Pulse rate (N/sec)	-	-	-	9.74 (± 1.53)
Sand	*Duration of pulse (sec.)*	-	-	-	0.0061 (± 0.0016)
	Peak frequency (Hz)	-	-	-	231.63 (± 28.93)
	Bandwidth of pulse (Hz)	-	-	-	402.60 (± 140.56)
	Number of pulse of train (N)	-	-	-	26.08 (± 7.0)
	Pulse rate (N/sec)	-	-	-	9.74 (±1.80)