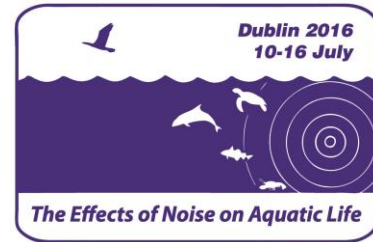




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## An investigation of inland water soundscapes: which sonic sources influence acoustic levels?

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Acoustic recordings were carried out in two different glacial lakes (i.e. Lough Na Fooey, Ireland and Windermere, U.K) using different Passive Acoustic Monitoring approaches. At Lough Na Fooey, a vessel-based survey over pre-established sampling stations covering the entire lake surface (together with a bottom survey) was carried out, while a moored sampling was carried out around the clock at selected sites in the shallow, gravel littoral shores of Windermere. Lough Na Fooey soundscape lacked both the biophony and anthrophony component. Night-time recordings from Windermere were characterized by biophony sources, such as invertebrate (family Corixidae) and fish air passage sounds. Day-time acoustic recordings from Windermere were characterized by consistent boat traffic noise. Classification models were used to investigate which sonic sources contributed to the detected noise levels. The results indicate anthropogenic noise as an important factor ruling freshwater soundscapes. Based on the results obtained, it is recommended that further studies focus on a wider geographical and temporal range in order to start filling the knowledge and legislative gaps regarding anthropogenic noise monitoring in inland waters.



## 1. INTRODUCTION

Anthropogenic noise is proven to elicit a wide range of physiological, perceptual and behavioral effects on aquatic life (e.g. Slabbekoorn et al., 2010), where some studies have investigated behavioral and physiological effects of boat noise on freshwater species. Altered nesting behavior was reported for the longear sunfish *Lepomis megalotis* (Mueller, 1980), while Graham & Cooke (2008) reported a dramatic increase in heart rate and a slight decrease in stroke volume in the largemouth bass (*Micropterus salmoides*). Wysocky et al. (2006) demonstrated that ship noise elicited a cortisol stress response in the common carp (*Cyprinus carpio*), the gudgeon (*Gobio gobio*) and the perch (*Perca fluviatilis*), regardless of their hearing sensitivities.

Fresh water makes up only 0.01% of the water on Earth and covers approximately 0.8% of the planet's surface, yet this small area of aquatic habitat supports almost 6% of all described species (Dudgeon et al., 2006). The biological communities inhabiting freshwater habitats constitute a valuable natural resource in economic, cultural, aesthetic, scientific and educational terms; however, inland freshwater habitats are experiencing far greater declines in biodiversity than terrestrial ecosystems (Dudgeon et al., 2006). Although it is well recognized that the biological communities inhabiting inland aquatic habitats currently face unprecedented threats from human activities (Winfield, 2013), and anthropogenic pressures often act in a multimodal fashion (Halfwerk & Slabbekoorn, 2015), anthropogenic noise pollution has been rarely measured and reported in inland water ecosystems (Amoser et al., 2004; Seppänen & Nieminen, 2004; Wysocki et al., 2007; Bolgan et al., 2016a). Furthermore, fewer studies have described inland water soundscapes in comparison to marine soundscapes (e.g. Stoiber, 1969; Nystuen, 1986; Lugli & Fine, 2003; Amoser et al., 2004; Seppänen & Nieminen, 2004; Lugli & Fine, 2007; Amoser & Ladich, 2005; Wysocki et al., 2007; Amoser & Ladich, 2010).

To date, a wide range of mathematical models are applied to acoustic data. Modelling of underwater sound propagation has been an established practice for decades; several modelling approaches have been developed, each with different suitability according to frequency range, computational requirements and ability to account for spatial variability (Farcas et al., 2016). On the other hand, the incorporation of acoustic data into models to calculate animal density and abundance is still a relatively young field (Cholewiak et al., 2013). Different modelling approaches have been developed for calculating animal density and abundance on the basis of the rate of animal vocalisations (Dawson & Efford, 2009; Efford et al., 2009; Marques et al., 2012; Cholewiak et al., 2013) or for investigating the relationships between measured acoustical data and geospatial data, with the scope of predicting acoustical conditions in environments with an unknown and potentially innumerable amount of acoustic sources (Mennitt et al., 2014). To the best of our knowledge, classification models have never been applied to investigate which environmental features may contribute to determine acoustic levels in freshwater soundscapes. The aims of this study were to apply classification models to i) investigate which environmental characteristics might influence noise level in a glacial lake in which anthropogenic noise was effectively absent (i.e. Lough Na Fooley, Ireland) and to ii) investigate the contribution of anthropogenic noise to the noise levels of a large multi-use glacial lake with numerous powered boats (i.e. Windermere, U.K.).

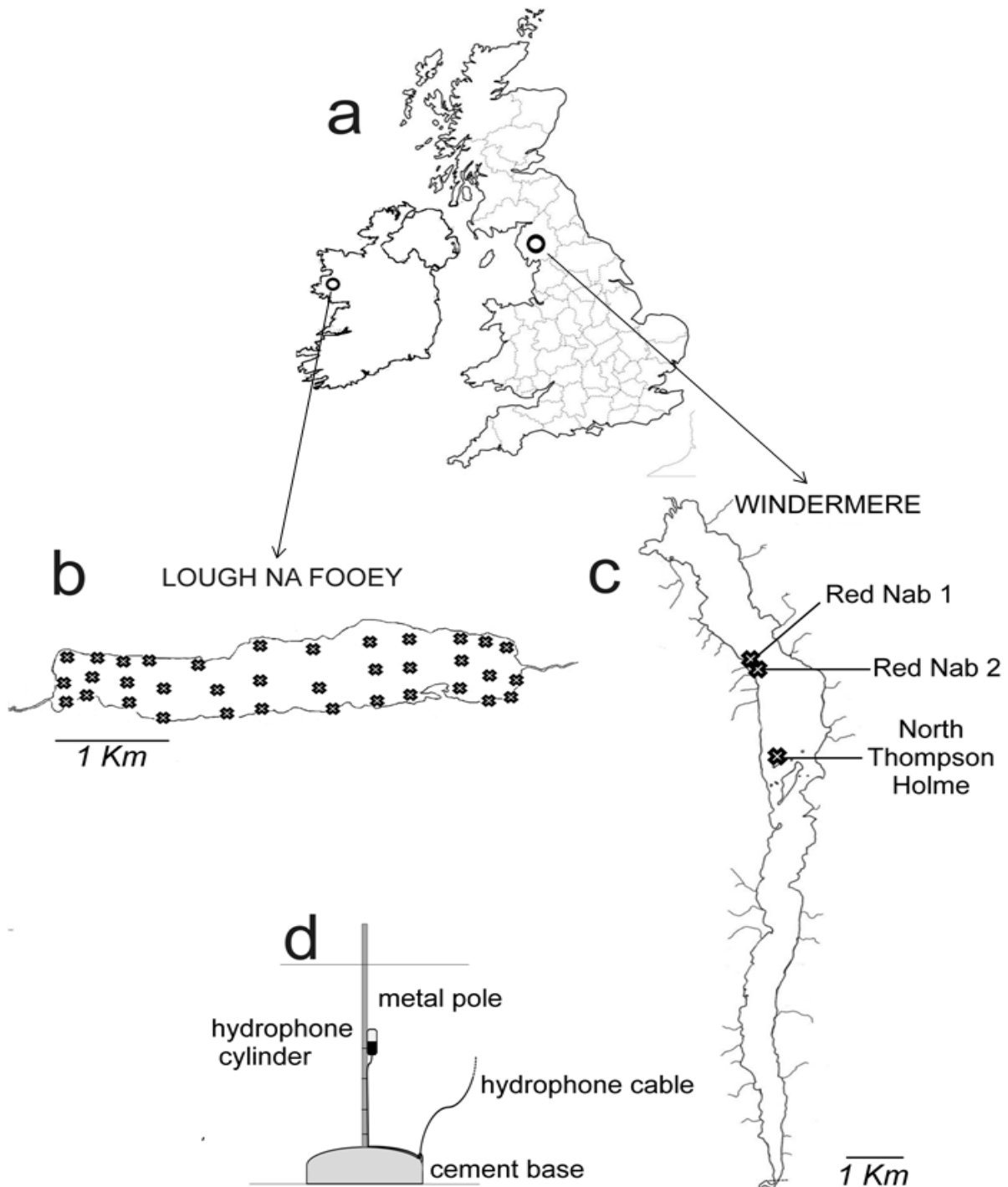
## 2. MATERIALS AND METHODS

### A. LOUGH NA FOOEY: A GLACIAL LAKE FREE OF ANTHROPOGENIC NOISE

#### i. Acoustic survey

Surrounded by Galway's Mamturk mountains to the South and Mayo's Partry mountains to the North, Lough Na Fooley (53° 34' N, 9° 32' W; altitude 24 m; 248 hectares) is a long and narrow rectangular shaped glacial lake bordering counties Galway and Mayo on the West coast of Ireland (Rooney et al., 2014) (Fig. 1a). During the acoustic sampling carried out at Lough Na Fooley, no boats were in operation on the lake (with the exclusion of the 6-m long angler fancy boat used for the survey); no harbors occur on the lakeshore, making Lough Na Fooley the perfect candidate for addressing noise levels in the effective absence of anthropogenic pressures. Thirty-six pre-defined sampling stations ensured an even coverage of the entire lake system at Lough Na Fooley (Fig. 1b). Acoustic recordings were taken at each station between October and December 2013 (1 to 2 repetitions per station). A total of 46 acoustic samples (10 minutes duration each, total of 460 minutes of recordings) were collected using an uncalibrated omni-directional hydrophone Magrec HP40 (hydrophone sensitivity -204 dB re 1V/Pa; hydrophone frequency response 10 Hz- 160 kHz). The hydrophone was connected to a waterproof stereo monitor box Magrec HP26, mounting a Magrec HP02 pre-amplifier (differential output, gain 29 dB) powered by batteries. The monitor box was connected to a ZoomH2N Handy recorder generating .wav files (sampling rate 44.1 kHz, 24-bit). At each sampling station, a 15 L manually operated Van Veen grab was used to collect a sediment sample, in order to assign each sampling station to one of four main granulometric categories (i.e. cobbles and

stones; medium sand; muddy sand; mud). Furthermore, at every sampling station, additional data recorded included; i) surface water temperature (deep sounder Garmin FishFinder 140), ii) bottom depth (deep sounder Garmin FishFinder 140), iii) sea state (visual estimation, Beaufort scale), iv) weather condition (visual estimation), v) wind speed and direction (hourly data obtained from the National Meteorological Service, measuring station 40 km distant from Lough Na Fooy).



**Figure 1-Recording sites and semi-moored PAM set-up. a) The location of Lough Na Fooy (Ireland) and Windermere (United Kingdom) is indicated; b) Lough Na Fooy: the 36 recording stations are depicted; c) Windermere; recording sites are indicated; d) semi-moored PAM set-up used at Windermere (custom-built hydrophone support).**

## ii. Acoustic analysis

All acoustic recordings were analysed for biological sound presence using Raven 1.5 for Windows (Bioacoustic Research Program, Cornell Laboratory of Ornithology, Ithaca, NY, USA), by aural and visual assessment of the spectrograms (sampling rate 44.1 kHz, 24 bit). Unwanted noise (such as cable noise, platform noise, wave slap noise) was cut from each recording prior to the frequency analysis. The frequency analysis of each acoustic recording consisted of a Fast Fourier Transformation based on a Hanning window at 4096 points (from 63 Hz to 22 kHz). The spectral values of all the recordings were converted in linear scale and then summed in wider bandwidths, discriminating for high and low frequency contents, i.e. Low sonic band (63-2000 Hz,) and High sonic band (2 kHz- 20 kHz).

## B. WINDERMERE: A LARGE MULTI-USE LAKE

### i. Acoustic survey

Windermere (54° 22' N, 2° 56' W; altitude 39 m; 1473 hectares) is the largest natural lake in England: situated in the English Lake District, UK (Fig. 1a), it is composed of a mesotrophic north basin (max depth 64 m, area 8.1 km<sup>2</sup>) and a eutrophic south basin (max depth 64 m, area 6.7 km<sup>2</sup>) (Miller et al., 2015). The lake is an important multi-use resource for the local economy, in terms of both general tourism (with an associated extensive ferry network) and recreational fishing. Windermere has consistent boat traffic during daytime hours, mainly of small recreational boats (with outboard engines) and cruise ferries (with inboard diesel engines), in addition to canoes, kayaks, rowing boats and sailing boats.

During November 2014, acoustic recordings were collected over three shallow water stations in the North basin of Windermere (i.e. 2 recording stations within the Red Nab site, and one recording station at the North Thompson Holme site) (Fig. 1c). Acoustic recordings were carried out for durations of 24 hours at each site using a semi-moored PAM configuration (Fig. 1d), with the exception of the first recording site within the Red Nab site (i.e. Red Nab 1), where recordings were conducted for 18 hours, from 15:00 to 09:00, due to technical limitations. In particular, four cycles of recordings were carried out over the 2 different sites within the Red Nab site and one 24-hour recording was carried out at the North Thompson Holme site (total of 108 hours of recordings). Bottom and hydrophone depth were the same in all recording sites (bottom depth=1.20 m; hydrophone depth=0.70 m); furthermore, all recording sites were characterized by a cobble type of substratum. All acoustic recordings were obtained using a calibrated omni-directional hydrophone Aquarian H2a (sensitivity -180 dB re 1V/Pa; frequency response 10 Hz-100 kHz) connected to a ZoomH1 Handy recorder (sampling rate 44.1 kHz, 24-bit) operating on an external power bank (Power walker N95LH, 10000 mAh) and recording .wav files. Prior to each recording, the signal was calibrated using a generator of pure wave of known voltage (100 mV RMS @1 kHz). Additional information collected for each hour of recording at each site included; i) wind speed, wind direction and water temperature (data were taken from the Centre for Ecology & Hydrology automatic monitoring buoy situated in the lake's south basin) and ii) weather conditions (i.e. no precipitation, shower and heavy rain, data obtained from the UK National Meteorological Service).

### ii. Acoustic analysis

All acoustic recordings were sectioned per hour, where acoustic analysis consisted of; i) quantification of sound sources, ii) Acoustic Complexity Index calculation and iii) spectral analysis in 1/3 octave band (dB re 1 $\mu$ Pa). In order to estimate presence and rate of biological sound sources, visual inspection was conducted using Raven 1.5 for Windows (Bioacoustic Research Program, Cornell Laboratory of Ornithology, Ithaca, NY, USA) on the first 10 min of each hour of recordings (listening period  $\tau_1 = 10$  min). Where biological sources (i.e. fish air passage sounds, see Bolgan et al. 2016b, and macroinvertebrate sounds) occurred, they were counted and categorized, while anthropogenic noise (i.e. boat noise) was quantified as the percentage of occurrence over the selected listening period  $\tau_1$  (10 min). The same 10 min listening periods ( $\tau_1$ ) were processed through the open source acoustic program Wavesurfer (v1.8), using a plug-in soundscape-meter developed by Pieretti et al. (2011) in order to calculate the Acoustic Complexity Index (ACI). The ACI was calculated across the frequency range of 63–22,000 Hz using an FFT size 512, Hanning window. Subsequently, following McWilliam & Hawkins (2013), ACI values were plotted and visually evaluated to establish the peak energy locus, in order to give a more localized sound profile. ACI values were finally averaged for the frequency band 400- 6000 Hz, for each hour of recording and for each recording cycle. Spectral analysis, on the other hand, was conducted over the entire 1 hour samples. Instantaneous Sound Pressure Level ( $L_{SP}$ , L-weighted, 63 Hz–20 kHz, RMS fast) was measured per second along each hour of acoustic samples using SPECTRA Plus 5.0 software (Pioneer Hill Software, WA, U.S.A.; windows Hanning, FFT overlap 75%, averaging fast). This software utilizes the Discrete Fast Fourier Transform algorithm to compute the frequency spectrum among the 1/3 octave bands (frequency range 63-20000 Hz). The equivalent continuous SPLs were further calculated for each hour averaging the  $L_{SP}$  over the entire one hour sample (after

linear scale conversion). In order to conduct the statistical analysis, the intensity levels characterising the 1/3 octave bands were converted to a linear scale and then summed in wider bandwidths, discriminating for high and low frequency contents i.e. Low sonic band (63-2000 Hz) and High sonic band (2 kHz- 20 kHz).

### C. STATISTICAL ANALYSIS

Statistical analysis was carried out using SPSS Modeller. Data were first inspected for type of distribution: acoustic data were found not to be normally distributed (Shapiro-Wilk,  $p < 0.01$ ). Subsequently, the outcome of different models for continuous numeric range outcomes (i.e. acoustic levels; Random Forest, CART, CHAID, linear regression and generalized linear regression) were generated and compared using the Auto Numeric function of SPSS Modeller. Models were compared based on correlation scores and relative error (McCormick et al. 2013). Chi-square Automatic Interaction Detector (CHAID), a technique created by Kass in 1980, was the model which provided the highest correlation scores and the lowest relative errors within all datasets (i.e. both Lough Na Fooley and Windermere) and was therefore used to discover the relationship between acoustic and environmental variables. CHAID analysis builds a predictive model, or tree, to help determine how variables best merge to explain the outcome in the given dependent (target) variable (McCormick et al. 2013). The CHAID node generates decision trees using chi-square statistics to identify optimal splits (McCormick et al. 2013). Unlike the CART Tree and QUEST nodes, CHAID can generate non-binary trees, meaning that some splits have more than two branches (McCormick et al., 2013). Although initially developed to deal with categorical dependent variables (Kass 1980), the CHAID models actually accommodate a variety of variables: target and input fields can be numeric (continuous) or categorical and no assumption on data distribution is made (McCarty & Hastak, 2007; McCormick et al., 2013). The CHAID procedure has been largely used in social and medical sciences (Elphinstone, 1986; Chung et al., 2004; Welte et al., 2004; Tan et al., 2005; Menendez et al., 2006) and, although less frequently employed, has also proven useful in biological studies (Schroder et al., 1992; Wolter & Menzel, 2005; Menendez et al., 2006).

The CHAID procedure was used on the Lough Na Fooley dataset in order to investigate how different environmental variables (i.e. depth, wind speed and direction, water temperature and type of bottom) might contribute to explain the detected noise levels in a freshwater environment free of anthropogenic noise and of biophonic sources. The CHAID procedure was used on the Windermere dataset considering night-time and day-time separately, in order to investigate the influence of anthropogenic noise (which was present only during day-time hours) on the detected noise levels of shallow freshwater soundscapes characterized also by biophonic sources (mainly macroinvertebrate sounds). Different CHAID analyses were carried out for each environment. For the Lough Na Fooley dataset, three CHAID models were created; the first used the broadband SPL as target variable, the second used the Low Frequency band and the third targeted the High Frequency band. In all cases, the predictors used were: weather condition, water temperature, depth, type of bottom, wind speed, wind direction. For the Windermere dataset, eight models were created, four for the night-time hours and four for the day-time hours. The target variables (for both day and night) were: SPL, Low sonic band, High sonic band and ACI. The predictors were: weather condition, water temperature, wind speed, wind direction, number of fish sounds, number of macroinvertebrate sounds and boat noise rate.

## 3. RESULTS

### A. LOUGH NA FOOEY

#### i. Wideband

The strongest predictors of the broadband sound pressure levels at Lough Na Fooley were bottom depth, wind speed and wind direction (Fig. 2). In particular, bottom depth appears as the main factor regulating the broadband sound pressure levels at Lough Na Fooley. In depths greater than 4 m wind speed was the main predictor of the detected SPL, while in shallower stations, wind direction also appeared to contribute.

#### ii. Low sonic band

The strongest predictors of the low frequency (i.e.  $< 2$  kHz) sound pressure levels at Lough Na Fooley were bottom type, wind speed and bottom depth (Fig. 2).

#### iii. High sonic band

The strongest predictors of the high sonic band levels (i.e.  $> 2$  kHz) at Lough Na Fooley were wind speed and wind direction. Bottom depth also appeared to influence the high sonic band, especially in conditions of high wind speed (i.e. winds stronger than 12 mph) (Fig. 2).

## **B. WINDERMERE DURING NIGHT-TIME HOURS**

### **i. SPL**

The presence or absence of rain appears to be the main factor affecting the broadband sound pressure levels in Windermere shallow waters during night-time hours (i.e. when no anthropogenic pressure in terms of boat traffic is present). In fact, the strongest predictors of the broadband sound pressure levels were weather condition (i.e. rain or not rain), wind direction, number of macroinvertebrate sounds and wind speed (Fig. 3). In the absence of rain, the presence of macroinvertebrate calls (i.e. presumed *Corixidae* spp.), together with wind characteristics (speed and direction) were the main factors influencing the broadband sound pressure levels.

### **ii. Low sonic band**

The presence or absence of rain appears to be the main factor ruling the low frequency band (i.e.  $< 2$  kHz) sound pressure level in Windermere shallow waters during night-time hours (Fig. 3).

### **iii. High sonic band**

The strongest predictors of the high frequency (i.e.  $> 2$  kHz) sound pressure levels in Windermere shallow waters during night-time hours were weather conditions, wind direction and number of macroinvertebrate sounds (Fig. 3). In particular, in the absence of rain, macroinvertebrate sounds appear to be the main factor ruling the high frequency band levels. The macroinvertebrate sounds detected in the shallow stations of Windermere were compared in their sound features (i.e. period between sounds of the same burst, period between sound bursts, sound peak frequency, pulse period, pulse peak frequency) with *Corixidae* cleaning sounds recorded in controlled conditions (i.e. aquaria, N=60 sounds kindly provided by Kevin French, unpublished data). Sound features were found to be similar (Mann Whitney test  $p$ -value  $> 0.05$ ); therefore the macroinvertebrate sounds recorded in Windermere were considered as belonging to the *Corixidae* family. The pulse peak frequency of these sounds recorded in Windermere was  $3639 \pm 80$  Hz.

### **iv. ACI**

The strongest predictors of the ACI were number of macroinvertebrate and fish sounds, water temperature and wind characteristics (Fig. 3). In particular, the main factor influencing the ACI in Windermere shallow waters during night-time hours was the presence of macroinvertebrate sounds. Furthermore, when water temperature was lower than  $8.5$  °C fish sounds also appeared to influence the ACI (Fig. 3).

## **C. WINDERMERE DURING DAY-TIME HOURS**

### **i. SPL**

During day-time hours, Windermere is characterized by frequent boat traffic, mainly of small recreational boats (with outboard engines) and cruise ferries (with inboard diesel engines), in addition to canoes, kayaks, rowing boats and sailing boats. Boat noise appears to be the main factor affecting the broadband sound pressure levels in Windermere shallow waters during day-time hours. Under conditions of high boat traffic (i.e. boat noise rate  $> 80\%$ ), the presence of rain also appeared to influence the broadband sound pressure levels, while wind speed contributed to the broadband levels under conditions of lower boat traffic (i.e. boat noise rate  $< 80\%$ ) (Fig. 4).

### **ii. Low sonic band**

The low frequency bands sound pressure levels are mainly influenced by the rate of boat traffic. When boat noise is present in more than  $80\%$  of the acoustic sample, this is the only factor determining the low frequency bands pressure levels. When boat noise is present in less than  $80\%$  of the acoustic sample, wind speed and direction also appear to contribute to the low frequency pressure levels (Fig. 4).

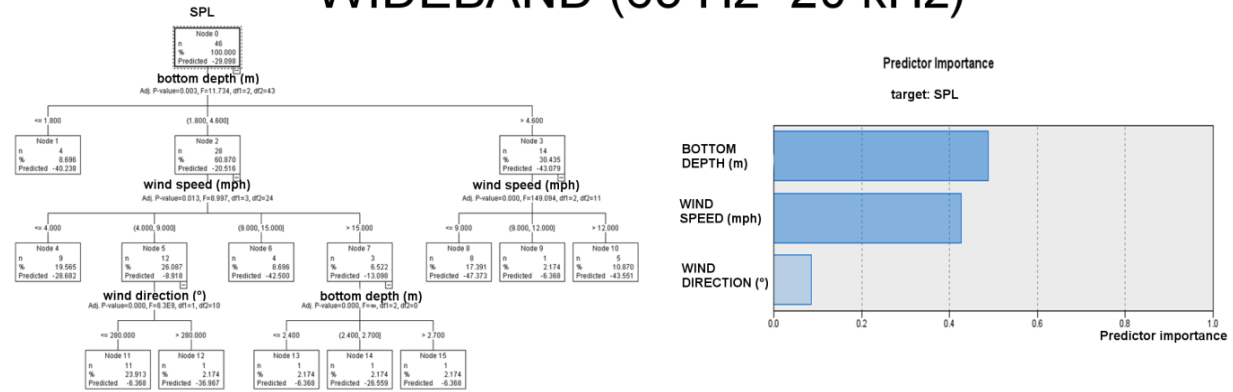
### **iii. High sonic band**

The high frequency bands levels were mainly influenced by the rate of boat traffic. When boat noise is present in more than  $80\%$  of the acoustic sample, this is the only factor determining the high frequency bands pressure levels. When boat noise is present in less than  $80\%$  of the acoustic sample, wind speed and direction also appear to contribute to the high frequency pressure levels (Fig. 4).

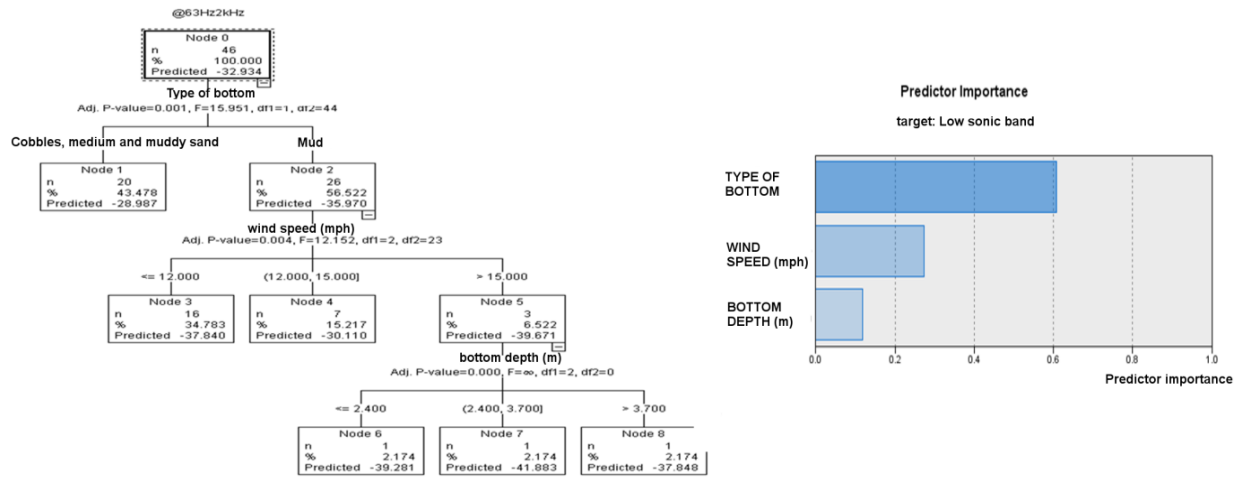
### **iv. ACI**

The strongest predictors of ACI values detected during day-time hours in Windermere were number of macroinvertebrate and fish sounds, wind speed, boat traffic and water temperature (Fig. 4).

## WIDEBAND (63 Hz- 20 kHz)



## LOW FREQUENCY BAND (63 Hz- 2kHz)



## HIGH FREQUENCY BAND (2-20 kHz)

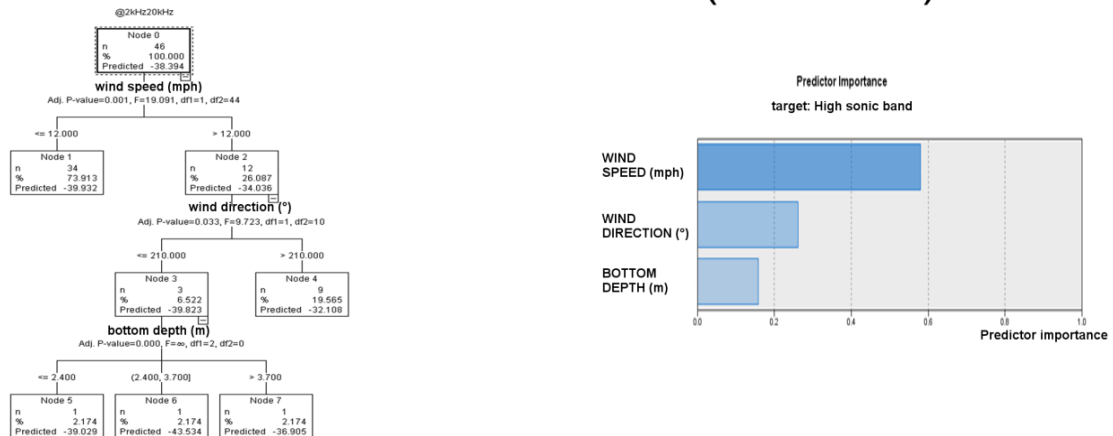
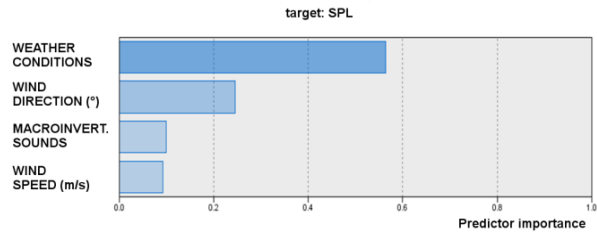
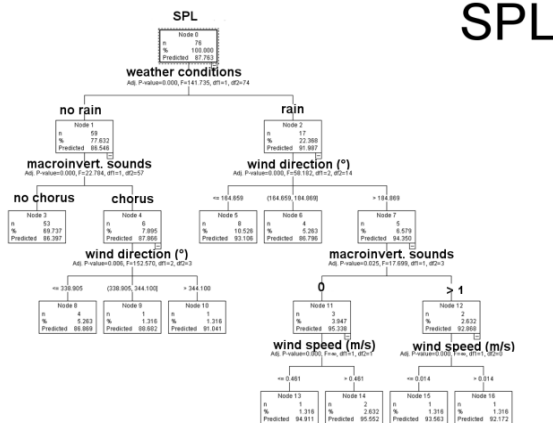
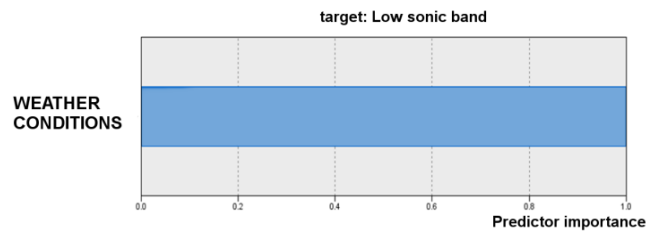
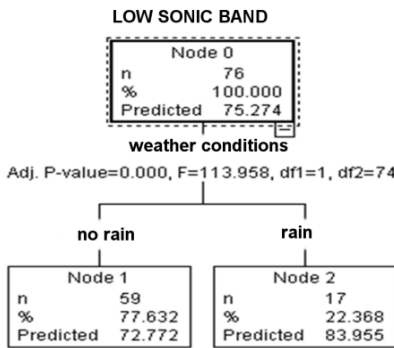


Figure 2- CHAID trees and predictor importance of the wideband, low frequency and high frequency band levels at Lough Na Fooey.



### LOW FREQUENCY BAND (63 Hz- 2 kHz)



### HIGH FREQUENCY BAND (2- 20 kHz)

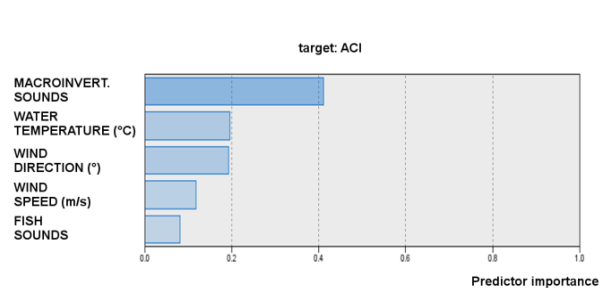
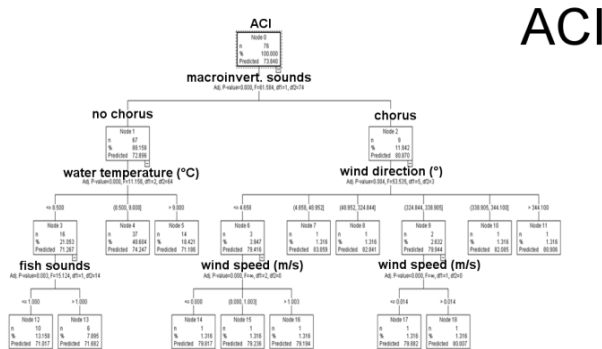
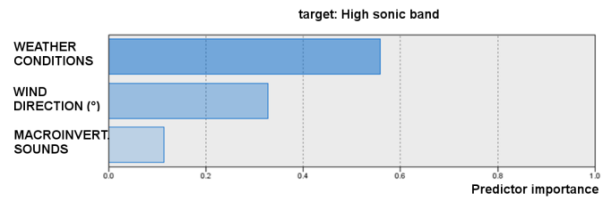
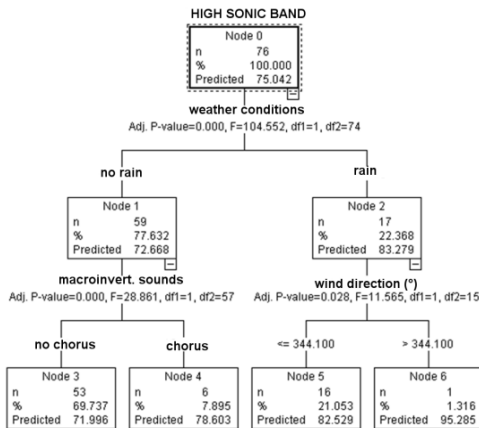
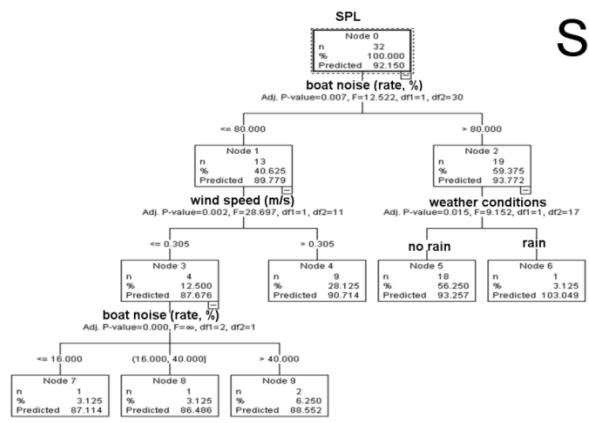
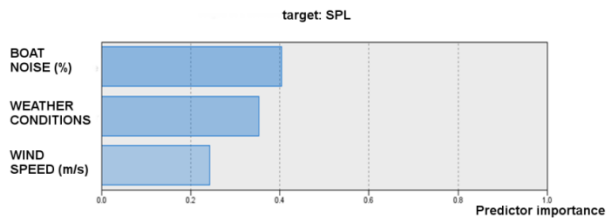


Figure 3- CHAID trees and predictor importance of the broadband, low frequency and high frequency band levels in Windermere during night-time hours.

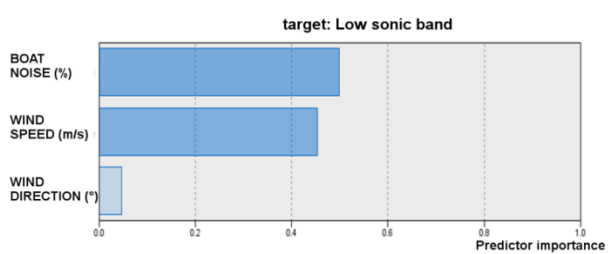
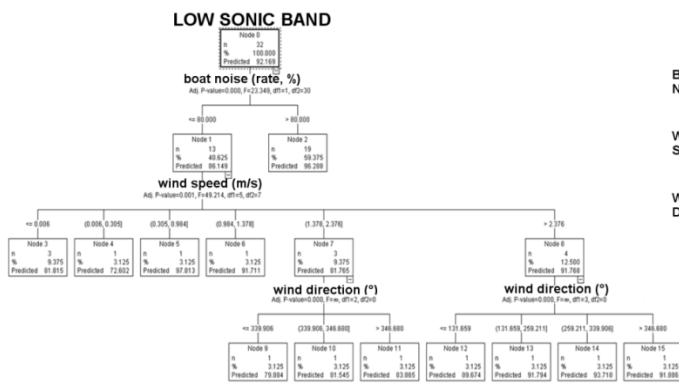




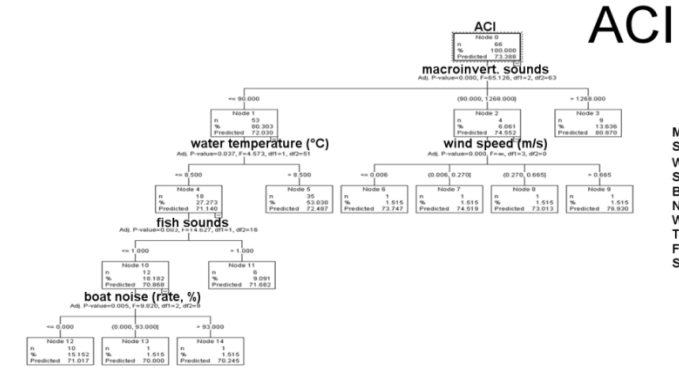
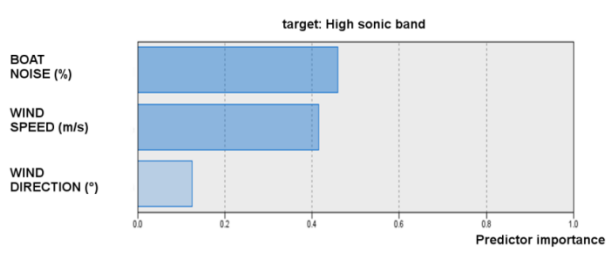
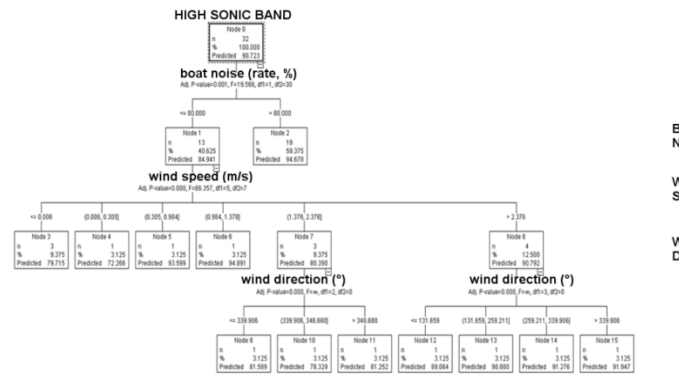
SPL



## LOW FREQUENCY BAND (63 Hz- 2 kHz)



## HIGH FREQUENCY BAND (2- 20 kHz)



ACI

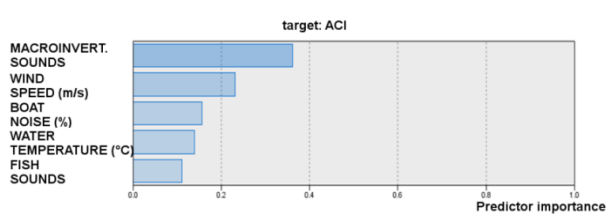


Figure 4- CHAID trees and predictor importance of the broadband, low frequency and high frequency band levels in Windermere during day-time hours.

## 4. DISCUSSION

Acoustic data were collected at different depths and over different bottom types at Lough Na Fooley during day-time hours and revealed a geophony dominated soundscape, i.e. both biophony and anthrophony sources were not detected. CHAID procedures identified depth as the main factor regulating the broadband sound pressure levels, with wind characteristics also contributing. In particular, in depths greater than 4 m, wind speed was the main predictor of the SPL, while in shallower stations wind direction also appeared to contribute. These results might be explained considering that, at Lough Na Fooley, the shallowest stations were located in closer proximity to the lake shore. The influence of wind direction on the sound pressure levels of these shallower, littoral stations might be explained considering the potential influence of mountainous formations, such as the Mamturk and the Partry mountains, on the wind energy that could actually reach the water column. The benthic habitat appears to be the strongest factor influencing the low frequency bands levels at Lough Na Fooley. The influence of the benthic habitat on the detected low frequency levels can be explained considering the cut-off phenomenon, i.e. the critical frequency below which the shallow-water channel ceases to act as a waveguide, causing acoustic energy to propagate directly into the bottom (Jenson et al., 2011). In shallow-water environments, sediments cause effective attenuation of sounds originating and propagating in the water column through both compressional wave absorption and the excitation of shear waves (Officier, 1958; Rogers & Cox, 1988). The effective attenuation of a particular sediment on the sound wave is the total transmission loss resulting from both intrinsic attenuation, scatter attenuation and energy conversion process. Different sediment types are characterized by different attenuation properties, where less coarse sediments have stronger attenuation properties, i.e. higher cut-off frequencies (Kibblewhite, 1989). The results of this first, preliminary application of classification models show that, in the absence of biophony and anthrophony sources, the low frequency component of inland waters soundscapes is influenced by the type of bottom, especially in shallow areas. Results highlighted by the CHAID procedures suggests that the information about the type of substrate may be encoded in the low frequency component of inland lakes' soundscapes. Further investigations are required both to validate this hypothesis and to investigate whether animals might be able to discriminate and rely on this information to locate important areas for their biological cycle.

In Windermere, acoustic data were collected at the same depth and over the same bottom type during night-time and day-time hours. Night-time acoustic recordings from Windermere were characterized by biophonic sources, such as macroinvertebrate (family Corixidae) and fish air passage sounds (see Bolgan et al., 2016b for a description of these sounds in a captive setting). Both weather conditions, wind characteristics and the presence of biophonic sources appeared to influence the soundscape of shallow gravel littoral environments during night-time hours and in the absence of boat traffic. The strong influence of rain on the detected noise levels, especially in the low frequency band, can be explained considering both the limited depth at which these recordings were collected (less than 1.5 m depth) and the (known) spectral characteristics of rain noise. Rain droplets hitting the air-water surface have been demonstrated to produce noise with most energy mainly below 5 kHz (Urlick, 1984), thus explaining the strong influence of weather conditions on the low sonic levels highlighted by the CHAID procedure on the Windermere night-time hours dataset. In particular, rain droplets have been shown to produce low frequency noise by means of three main processes; i.e. the impact itself, the oscillations of the air-water surface and the oscillations of the entrained air carried by the rain droplet below the water surface) (Urlick, 1984). On the other hand, the influence of macroinvertebrate calls on the night-time sound pressure levels, especially in the high sonic band, can be explained considering both quantity and spectral characteristics of macroinvertebrate sounds. Macroinvertebrate sounds were detected across all sites (Red Nab 1 and 2 and North Thompson Holme) and in some cases, a constant chorus was detected. As these sounds overlapped, the precise number could not always be determined, but more than 1300 sounds across the 10 minute listening period ( $\tau_1$ ) were calculated. The peak frequency (i.e. the most energetic frequency) of the pulses characterising macroinvertebrate sounds recorded in Windermere was  $3639 \pm 80$  Hz, thus explaining the influence of this type of biophonic source on the high frequency bands levels. Macroinvertebrate sounds were found as the main predictor of the ACI during night-time hours. The ACI, originally developed by Farina & Morri (2008), is an algorithm designed to produce a direct quantification of complex biotic sound patterns by calculating the variability of the acoustic intensity of audio-recordings. In the last 5 years, some studies have begun to investigate the possible applications of terrestrial ecoacoustic indices such as ACI to the marine environment (Harris et al. 2016). In both terrestrial and marine soundscapes, the ACI was found to be positively correlated to both number of vocalizations and traditional species diversity indexes (such as Pielou's Evenness and Shannon's index) (Harris et al., 2016). The results of this first application of classification models to inland waters soundscape seems to suggest the ACI as an effective acoustic metric to detect macroinvertebrate calls in shallow, littoral areas. Furthermore, the CHAID procedure highlighted the influence of water temperature and fish air passage sounds on the ACI. This might be explained considering

that the fish air passage sounds recorded in Windermere were air passage sounds likely emitted by Arctic charr (*Salvelinus alpinus*) during its spawning season (see Bolgan et al., submitted) over littoral spawning grounds. In Windermere, Arctic charr is known to enter the lake shore margins to spawn during November, when temperature generally drops below 8 °C (Miller et al., 2015). This might explain the influence of this biophonic source on the ACI in relation to water temperature (i.e. in warmer littoral waters, these sounds are unlikely to occur). Even if the emission of macroinvertebrate calls was most consistent during night-time hours, it has to be noted that these calls were emitted also at sunrise and sunset, thus influencing the ACI also during day-time hours. Also boat noise influenced the ACI in Windermere during day-time hours. These results seem to confirm the ACI as an effective metric to detect biophonic sources in contrast to anthropogenic sources in inland shallow water environments, where lower values of ACI correspond to intense boat traffic and higher values of ACI indicate the presence of biophonic sources (in accordance with literature, Pieretti et al., 2011). Concluding, the ACI appears as an effective metric to characterize the biophonic and the anthropogenic component of inland water soundscapes.

During day-time hours, Windermere is characterized by frequent boat traffic, mainly of small recreational boats (with outboard engines) and cruise ferries (with inboard diesel engines), in addition to canoes, kayaks, rowing boats and sailing boats. This preliminary application of classification models to freshwater soundscapes seems to indicate that when anthropogenic noise is present, this represents an important factor ruling the acoustic environments of the biological communities inhabiting these environments. In fact, CHAID procedures identified boat noise as the main contributor to the broadband pressure levels in Windermere shallow water stations during day-time hours. In particular, both the low and the high frequency sound pressure levels were mostly influenced by the rate of boat traffic. When boat noise was present in more than 80% of the acoustic sample, this was the only factor ruling both the low and the high frequency bands sound pressure levels. When boat noise was present in less than 80% of the acoustic sample, wind speed and direction also appeared to contribute to the low and high frequency sound pressure levels. Noise energy generated by shipping traffic is known to be mainly concentrated below 1 kHz (Nakahara, 1999), however, the additional contribution of boat noise to the high frequency bands (i.e. >2kHz) in the shallow waters of Windermere during day-time hours can be explained considering both the type of noise sources (i.e. type of boat and propeller) and the extent of environmental filtering. Seppänen & Nieminen (2004) found that, in a Finnish Lake, inboard diesel-powered boats produced most of their noise at high frequencies (1000-4000 Hz) with SPLs of 133 dB re 1µPa. Furthermore, outboard engines were the loudest, producing noise with SPL of 140 dB re 1µPa at 50-100 m distance and with the most energy centred to high frequencies (above 1000 kHz). The results of Seppänen & Nieminen (2004) are comparable to the values recorded in Windermere (Bolgan et al., 2016a). The intense traffic of both cruise ferries (i.e. with inboard engines) and small recreational boats (i.e. with outboard engines) in Windermere could therefore explain the overall high frequency content of boat noise recorded at this lake. Finally, regarding environmental filtering, it should be noted that this study was conducted in very shallow waters (less than 1.5m deep). The cut-off phenomenon could therefore contribute to the relatively reduced amount of low frequency energy detected in Windermere.

Anthropogenic noise is a complex and challenging source to quantify as it varies in duration, amplitude and frequency content, and as it can also be modified by the medium through which it travels (Shannon et al. 2015). It has to be noted that this present study is strongly limited by the extremely restricted sample size (i.e. models were built on ca. 115 hours of recordings versus the over 270,000 hours used in previous acoustic modelling studies, e.g. Mennitt et al., 2014). Furthermore, although aquatic spectral and temporal soundscape composition have been proved to vary over relatively short geographical and time scales (e.g. Radford et al., 2010), this study did not involve simultaneous long-term recordings over different sampling stations. This study therefore lacks significant temporal and spatial resolution, failing to account for the potential, consistent variation of sonic sources occurring over relatively small areas and short periods of time. Considering these limitations, this study can be considered as a restricted, preliminary implementation of classification models to inland water soundscapes. However, it nevertheless represents the first application of such models to inland water soundscapes. Furthermore, it highlights for the first time the strong influence of anthropogenic noise pollution on the soundscape of a large multi-use lake in which frequent shipping, recreational fisheries and the nationally rare Arctic charr (Winfield et al., 2008) co-exist. Considering that the results of this study suggest that when anthropogenic noise is present, it constitutes the main factor ruling the acoustic environments of the biological communities inhabiting inland water environments, further studies addressing noise levels, sources and effects in freshwater environments are recommended across a wider geographical, temporal and taxonomic range. On a regulatory level, it might be advisable to consider expanding freshwater environmental legislation to include underwater noise levels as an indicator of inland water quality and ecological status, using a similar legislative approach to that adopted under the Marine Strategy Framework Directive (MSFD) of the European Union (Commission Decision 2010/477/EU). The MSFD requires European Member States to develop strategies in order to achieve and maintain Good Environmental Status (GES) in European Seas (European Commission 2008). Two indicators for underwater noise are used to describe the GES. In particular, Indicator 11.2.1 focuses on low frequency ambient noise, with the main

contributor given by commercial shipping noise. It requests monitoring of the yearly trends of underwater noise level within the 63 and the 125 Hz 1/3 octave bands (centre frequency), measured in different observation stations. In the case of freshwater environments, the first step toward a possible amendment of the Water Framework Directive of the European Union (WFD; 2000/60/EC) and corresponding legislation elsewhere in relation to noise, would involve year-round monitoring in important, multi-use systems in order to identify which frequency bands are most indicative of the actual levels of noise in inland waters. Once the appropriate frequency bands (i.e. indicators) are identified, acoustic monitoring of inland waters noise pollution could be carried out across wide geographical scales using a standardized approach. Ultimately, potential mitigation measures such as the definition of noise-free areas (i.e. fish spawning grounds, essential fish habitats), and seasonal restriction of noisy activities during sensitive biological periods should be considered (Shannon et al., 2015).

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