

**Echoes in motion:
An acoustic camera (DIDSON) as a monitoring tool in
applied freshwater ecology**

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Manuel Langkau

aus Essen

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Berichterstatter (Gutachter): Prof. Dr. Jost Borchering

Prof. Dr. Hartmut Arndt

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«Das Prinzip aller Dinge ist Wasser;
aus Wasser ist alles, und ins Wasser kehrt alles zurück.»

Thales von Milet (um 624 - 546 v. Chr.)

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Abbreviations and terms

0+	same definition as young of the year fishes, that had hatched within that year
Beam	Defined volume of water ensonified by the sonar
ARIS	Adaptive Resolution Imaging Sonar
Bull	The phrase “bulls” originates from the Garonne River and is related to the Occitan language (Cassou-Leins et al., 2000). It has been established among French scientists and describes a typical <i>A. alosa</i> mating with circling individuals.
DIDSON	Dual Frequency Identification Sonar
Echogram	A chart plotting received sonar data over time. Typically the x-axis shows the time or the pings and the y-axis the received intensities over the range.
EFA	Elliptic Fourier Analysis
Fish trace	Footprint of an object (fish) in the sonar beam plotted in an echogram over the time.
HPP	Hydro power plant
NEFD	Normalised Elliptic Fourier Descriptor
PCA	Principal Component Analysis
YOY	Young of the year fishes, that had hatched within that year

Teilpublikationen

- Langkau, M. C., Balk, H., Schmidt, M. B. & Borcharding, J. (2012). Can acoustic shadows identify fish species? A novel application of imaging sonar data. *Fisheries Management and Ecology* **19**, 313-322.
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Abstract

Aquatic environments are increasingly faced with anthropogenic impact. Rivers have been fully developed into navigable waterways resulting in a dramatic loss of habitats and longitudinal and horizontal disconnection. Human sewage polluted rivers and in addition with overfishing and a rising recreational use, most European diadromous fish species have suffered and can be graded as vulnerable. Species-conservation and re-stocking projects support to save diverse fish communities. Capable monitoring- and assessment tools are urgent, given that knowledge is the key for sustainable management. The DIDSON, a multibeam sonar, delivers video-like live images in high resolution, enabling the measurement of fish length and behavioral observations even in turbid water and by night in a non-invasive manner.

It could be shown that a special application of the sonar offers potential to discriminate fish species based on their characteristic acoustic shadows. Beside the possibility to count and measure fish and to observe their behavior this provides additional valuable information in certain monitoring applications.

New insights in the spawning behavior of *Alosa Alosa* could be revealed on a spawning site in the Garonne River, France. It could be observed that spawning events are not restricted to one couple since additional individuals join. Drifting clouds of sexual products and micro bubbles could be detected with the sonar and were consistent with the number of sound based spawning measurements (by human hearing) and thus served as an indicator for spawning activity.

A mid- term application of the sonar in front of a trash rack of a hydro power plant demonstrated the potential to gain knowledge in the field of spatial ecology of fish with high temporal resolution. An hourly fish abundance raster was chosen as a measure of fish activity to intuitively illustrate alternating diel and seasonal activity patterns at a glance. Distinct patterns and migration peaks of three groups 'fish', 'eels' and 'shoals' could be identified. The size class potentially at risk to pass the trash rack, was faced the power production data to identify time windows of higher and lower risk of entrainment and respective fish protection requirements.

Keywords: DIDSON, fish activity, fish protection, fish species discrimination, hydro acoustic, spawning activity

Introduction

Aquatic as well as terrestrial environments are facing a continuous increase of anthropologic impact, as human's experienced strong attraction since the beginning of time. With the beginning of the industrialization fish are confronted with a steady increase of man-made transformation of their environment (Limburg & Waldman, 2009). Rivers have been fully developed into navigable waterways by straightening their riverbed and applying bank reinforcements. Weirs and dams were built for drinking water abstraction and shipping, while power plants were constructed for energy production. Industrial and human sewage polluted many rivers (Limburg & Waldman, 2009).

This led to a dramatic loss of habitat diversity and lateral and longitudinal connection of rivers, which is crucial for a diverse fish community (Ward, 1989; Scharbert & Borcharding, 2013). These factors together with overfishing represent the major causes for the dramatic declines, and in some cases, the extinction of diadromous fish species in European rivers (De Groot et al., 2002; Limburg & Waldman, 2009). Additionally, today water has a big attraction as recreational area. Thus, besides shipping, sport boat traffic has potential to negatively influence fish and other aquatic animals.

Great efforts have been done to improve the conditions and to learn from mistakes made in the past. Re-introduction projects have successfully led to returning diadromous species in some systems, e.g. Houting (*Coregonus oxyrinchus*) (Borcharding et al., 2013) and European Shad (*Alosa alosa*) (Hundt et al., 2015) in the Rhine system. However, success of sustaining a diverse fish community depends on knowledge. Managers need to know exactly about the demands of a species and the developments of population status.

Powerful examination tools are required to monitor fish populations. Classical methods like netting and electro fishing can be a solution, but in some cases populations are vulnerable and non-invasive methods are needed to avoid negative impact caused by stock-assessment methods. Thus, in some areas non-invasive monitoring methods are therefore preferential (Johnson & Moursund, 2000).

Hydro acoustics represent such a less- or non-invasive method and gives the additional possibility to assess fish behavior. Depending on the task, nowadays a variety of hydro acoustic instruments are available to address different topics of fish monitoring without the need to catch and handle one fish.

Fishery acoustics

Water is not only the environment of aquatic life. It is also a suitable medium for the transmission of sound. In contrast to light and radio waves, sound waves have a comparably low attenuation under water and travel with a speed of $\sim 1450 \text{ m s}^{-1}$ (freshwater) (Simmonds & McLennan, 2005), depending on the physical characteristics of the water. Aquatic animals, e.g. marine mammals took advantage of this phenomenon by evolving echolocation and remote sensing via sound to communicate over hundreds of kilometers in the oceans (Schevill et al., 1964; Walker, 1963; Watkins et al., 1987 cited in Simmonds & McLennan, 2005). Leonardo da Vinci already documented that ships could be heard over long distances by installing a long tube in the water and listen at its end in 1490 (Urick, 1983).

The potential of transmitting and receiving sound waves (active sonar) to detect fish over long distances was recognized already in the first part of the 20th century although the main driving factor in technical development can be related to military research in the first and second World Wars (Simmonds & McLennan, 2005; Urick, 1983).

However, generally the function of acoustic instruments e.g. sonars and echosounders can be described by the emission of a pulsed pressure front (sound wave), which propagates at the local speed of sound until a part of the transmitted sound wave will be reflected and scattered in the moment it encounters another medium or target. While one part of the energy is transmitted in to the new medium the remainder is travelling back and can be received by the sonar as an echo (Simmonds & McLennan, 2005).

While the first fish school related echoes were mentioned by Rallier du Baty (1927) and Portier (1924) cited in Simmonds & McLennan (2005), the first successful acoustic detection of fish was demonstrated in an experimental setup by Kimura (1929).

The exploration of the oceans by the use of sonars has many advantages. Compared to conventional biological sampling methods, mobile surveys enable the coverage of a large volume of water per time (Simmonds & McLennan, 2005) and represent a fast remote method that is less-invasive and non-extractive, by providing a high spatial resolution (Chu, 2011).

The technological progress led to the application of acoustics in fishery, enabling an effective method for fish school detection and capture. This is true especially after the

Second World War (Simmonds & McLennan, 2005). Up to date hydro acoustic methods have reached a high quality in acoustic hardware, as well as in the understanding and post processing of acoustic data.

Especially for the study of fish populations and ecology by means of the estimation of fish abundance, distribution on time and space axis and population size structures, acoustic methods and the mobile application of scientific split-beam echosounders have established among and in combination with other fishery methods. As a result a European standard protocol has been developed to ensure the comparability of acoustic survey data (Ref. No. EN 15910:2014 E).

Multi-Beam

One of the latest milestones in the history of fishery acoustics is the development of multi-beam technology (Chu, 2011). Imaging sonars represent one special type of multi-beam sonar. In the last decade the development of these devices experienced large technological progress.

The DIDSON (Dual Frequency Identification Sonar - www.soundmetrics.com), which was used in the present study, represents a high-resolution imaging sonar using a unique system of acoustic lenses to focus the emitted sound to generate high quality images underwater (Belcher et al., 2001). It counts to the latest developments in the field of sonar imaging technology. Recently the follower ARIS (Adaptive Resolution Imaging Sonar) was released which is based on the same principle by delivering even higher resolution due to a higher operation frequency and a higher number of beams. This type of visualization sonars, often called 'acoustic camera', deliver video like sequences with high frame rates. Although it is also possible to display sonar recordings in classical echograms, plotting echo intensities over the range as a function of time, imaging sonars enable the playback of videos showing high resolution pictures of fish, including information about their shape, size and behavior. However, originally engineered for underwater surveillance, inspection and exploration new fields and old challenging hydro acoustic topics in fisheries research and fish ecology can be addressed by bridging the gap between existing fisheries-assessment sonar and optical systems (Moursund et al., 2003).

During the last 15 years a variety of research in fish ecological applications has been conducted since fish biologists started to explore the potential of DIDSON as a fish monitoring tool. Moursund et al. (2003) early evaluated the sonar, based on the

multibeam nature, to represent a robust fish monitoring tool in acoustically noisy environments commonly encountered at hydropower facilities.

Most of the studies addressed fish detection, counting and sizing due to the ability of the system to work even in turbid water. In this context examinations have been conducted in wild rivers (Crossman et al., 2011; Han & Asada, 2007; Hayes et al., 2015; MacNamara & MacCarthy, 2014; Magowan et al., 2012; Maxwell & Gove, 2007; Pipal et al., 2010; Rakowitz et al., 2013; Rand & Fukushima, 2014) as well as in aqua culture (Han et al., 2009; Zhang et al., 2014) and marine environments (Frias-Torres & Luo, 2009). Here, Han & Uye (2009) and Makabe et al. (2012) demonstrated the detection capabilities of DIDSON by assessing abundance and distribution of jelly fish, which is quite demanding regarding the low density differences between jelly fish and water.

Another major topic addressed, is the observation of fish behavior, which was very limited in turbid waters, even with optical systems and conventional hydro acoustic methods in the past. Thus, fish behavioral studies were often restricted to experimental setups in the lab where the visibility can be kept constant. Becker et al. (2013) and Xie et al. (2008) examined the impact of boat traffic on the behavior of fish in estuaries and migrating salmon respectively. Predation related behavior was examined with DIDSON by Becker & Suthers (2014) and Price et al. (2013) while O'Connell et al. (2014) used the DIDSON follower ARIS to analyze the behavior of bull sharks at a sharksafe barrier. The selectivity and the efficiency of trawls have great relevance in fishery. Naturally that several studies involved DIDSON to investigate fish behavior and trawl avoidance in the marine (Handegard & Williams, 2008; Williams et al., 2013) and even freshwater application (Juza et al., 2013; Rakowitz et al., 2012).

The reliability of the measurements based on imaging sonar data is an important point. Several studies addressed the accuracy of sizing (Burwen et al., 2010; Hightower et al., 2012; Tuser et al., 2014, Zhang et al., 2014) and counting (Holmes et al., 2006; Petreman et al., 2014) measurements to carve out the limitations of the system. In summary the authors agree that DIDSON is a valuable tool to estimate fish length with some restrictions. The accuracy can vary with the fish size, the orientation of the fish in the beam and also the morphology of the species. Tuser et al. (2014) and Hightower et al. (2012) demonstrated that the sizing error is larger for small fish compared to bigger fish and the dependence of the fish's orientation in the

beam array. A rather perpendicular angle of the fish in relation to the center of the beam array allows the most accurate measurements. Tuser et al. (2014) developed an error function to compensate for DIDSON based length measurements in an experimental setup. Hightower et al. (2012) found underestimations of the size of Atlantic Sturgeon and related this to the characteristic morphology and the constrained difficulty to see the end of the snout and tail. Probably the underestimations in the size of cultured Chinese sturgeon found by Zhang et al. (2014) represent this issue. Despite limitations and sources of inaccuracy, DIDSON length measurements were demonstrated to correlate with the total length of the fishes and hence deliver ecological relevant data extracted with a non-invasive method.

In the field of DIDSON based fish species identification research is still scarce. Mueller et al. (2008) found that eels can be identified and separated from debris by a computer driven process under certain conditions and that tail-beat-patterns in DIDSON echograms can be used to discriminate between Sockeye salmon (*Oncorhynchus nerka*) and Chinook salmon (*Oncorhynchus tshawytscha*) based on tail-beat frequency and fish length (Mueller et al., 2010). Grote et al. (2013) used a Bayesian mixture model based on the known fork length distributions of four fish species to apportion DIDSON observations by species in a fishway entrance at a dam in the Penobscot River, Maine. Target species was American shad (*Alosa sapidissima*).

Objective

This thesis was focused on contributing knowledge in three fields with high demand for research and aimed for building bridges between latest technological progress in sonar hardware and applied fish ecological questions (Fig. 1).

Species identification

Acoustic fish species identification is one of the most challenging topics (Horne, 2000) and up to date still a field that has been identified to have a high demand for further research (Pollom & Rose, 2016). Against the background of a rising anthropological influence on rivers and the need of non-invasive fish monitoring tools, a special setup of the sonar was examined to explore species specific data contained

in DIDSON recordings and to find post processing methods for fish species discrimination by analyzing acoustic fish shadows, which are strongly correlated to fishes' outer shapes.

Behaviour

Most European diadromous fish species today are classified as threatened or endangered due to anthropogenic impact (Limburg & Waldman, 2009). In the last decade, remaining populations of the anadromous Allis Shad (*Alosa alosa*) have declined seriously and are now characterized as vulnerable (Rougier et al., 2012). Due to the nocturnal spawning at sites with challenging monitoring conditions the knowledge about spawning behavior is still poor. A DIDSON was installed at a known Allis Shad spawning site to reveal details of the spawning behavior and the possibilities to use the sonar based observations as a measure for spawning activity and success.

Fish Activity

In many rivers potamodromous and diadromous fish migration is influenced by the obstruction with hydro power plants. Compared to the quite good knowledge and the existence of state of the art guidelines for the upstream transfer of fish (DWA, 2014), the knowledge about efficient downstream transfer and the demands of downstream migrating fish is still poorly assessed, although the EU Water Framework Directive (Directive 2000/60/EC) requires both, unobstructed migration up- and downstream. Knowledge of the spatial ecology of fish is the key for successful management actions and can on the other hand be a source of error when misunderstood (Cooke et al., 2016). A DIDSON was mounted in front of a trash rack of a hydro power plant between April and December. The potential to use DIDSON data to intuitively illustrate diel and seasonal fish activity patterns at a glance was evaluated and time windows with higher or lower entrainment risk and respective fish protection requirements were derived, by facing activity data of small fish with power production data.

- CHAPTER I** **Fish species:** Evaluation of discriminative properties of acoustic fish shadows and a possible post processing method.
- CHAPTER II** **Behaviour:** Evaluation of the number of Allis Shads involved in a spawning event and the possibilities to use the sonar based observations as a measure for spawning activity and success.
- CHAPTER III** **Fish activity:** Assessment of diel and seasonal fish activity patterns in front of a hydro power plant and derived time windows with higher or lower entrainment risk and respective fish protection requirements.

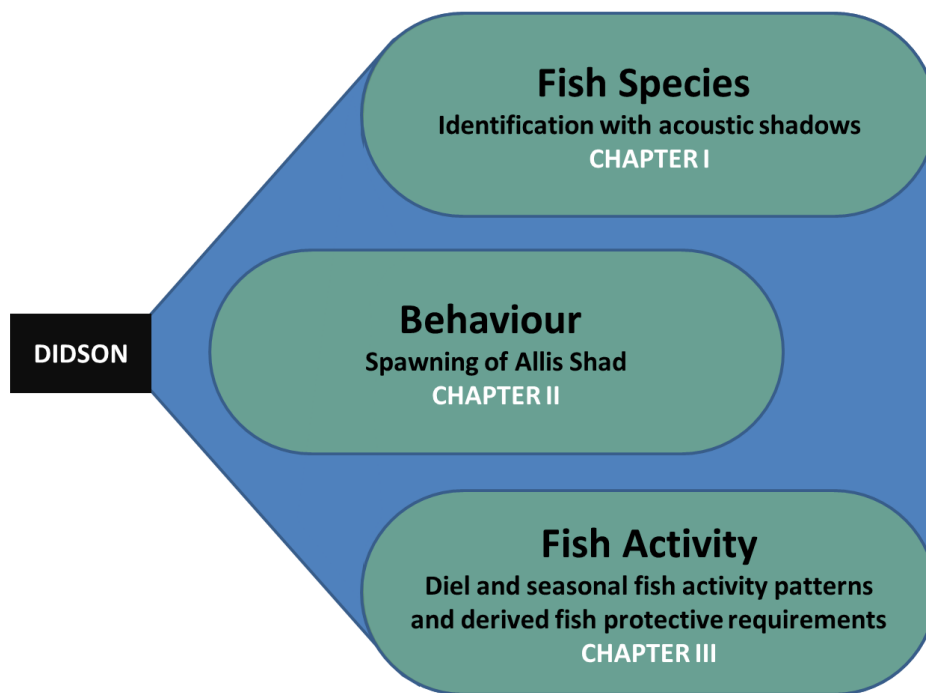


Figure 1. Overview about the three fields of research the present thesis was focused on.

Material and Methods

Underwater sound and sonar

Although multibeam imaging sonars are quite complex instruments, their general function is based on the same principles of underwater sound, which are valid for simple single beam sonars. The theory of underwater sound has been described in detail, among others by Simmonds & McLennan (2005) and Urick (1983). Most of the information given below is based on these books.

SONAR is the abbreviation for **SO**und **N**avigation **A**nd **R**anging. Sonars work after the principle of transmission and reception of sound. The transducer of a sonar system converts electrical energy into acoustic energy and emits a pulse of a certain frequency (depending on the sonar system) into the water. The sound propagates away from the transducer through the medium in a beam similar to the beam of a torch light. Targets reflect the pulse which travels back to the transducer to be received as an echo and transformed into electrical energy again. The received signal is then amplified and processed in various ways depending on the kind of system to be finally displayed in an echogram on a control screen (Simmonds & McLennan, 2005).

In comparison to light and radio waves, the attenuation of sound waves underwater is lower. Depending on the physical characteristics of the water, sound is traveling with a speed of $\sim 1450 \text{ m s}^{-1}$ in freshwater. Temperature, salinity and pressure affect the speed of sound underwater. A sound wave is characterized by advancing pressure fronts of certain frequency and wavelength. While the wavelength (λ) is defined by the distance between the pressure fronts in the wave, the frequency (f) in Hz can be described by the number of cycles per second. The speed of sound (c) can be calculated by the product of λ and f . (Simmonds & McLennan, 2005).

$$c = \lambda f$$

The frequency and the wavelength are crucial parameters affecting the spatial resolution of sonars. The smaller the wavelength or the higher the frequency the better is the resolution and thus the ability to discriminate between targets that are close together. This is for example important for the detection of fish in denser schools or fish that are close to the bottom. The detection range of sonars is also

depending on the frequency. With increasing frequency the absorption losses raise rapidly due to higher friction losses (Simmonds & McLennan, 2005). The higher the frequency the shorter is the possible detection range. As a result, the frequency of a sonar has an important influence on its application. There is a trade-off between range and resolution.

Regarding conventional sonars, frequencies between 30-200 kHz are normally used for fish detection, while higher frequencies are suitable e.g. for plankton studies. Lower frequencies are used for the detection of large targets like fish schools on long ranges in the marine environment (Simmonds & McLennan, 2005).

DIDSON (Dual Frequency Identification Sonar)

Although the DIDSON basically works with the same principles like conventional echosounders, it is necessary to describe the more complex architecture due to the high number of beams used and to explain the difference of its application in the field.

Traditionally the use of single or split-beam sonars is carried out in mobile surveys with a vertical beam orientation in order to capture the water column under the boat, including the targets of interest e.g. nekton or plankton (Fig. 2). This enables to cover a large volume of water per time.

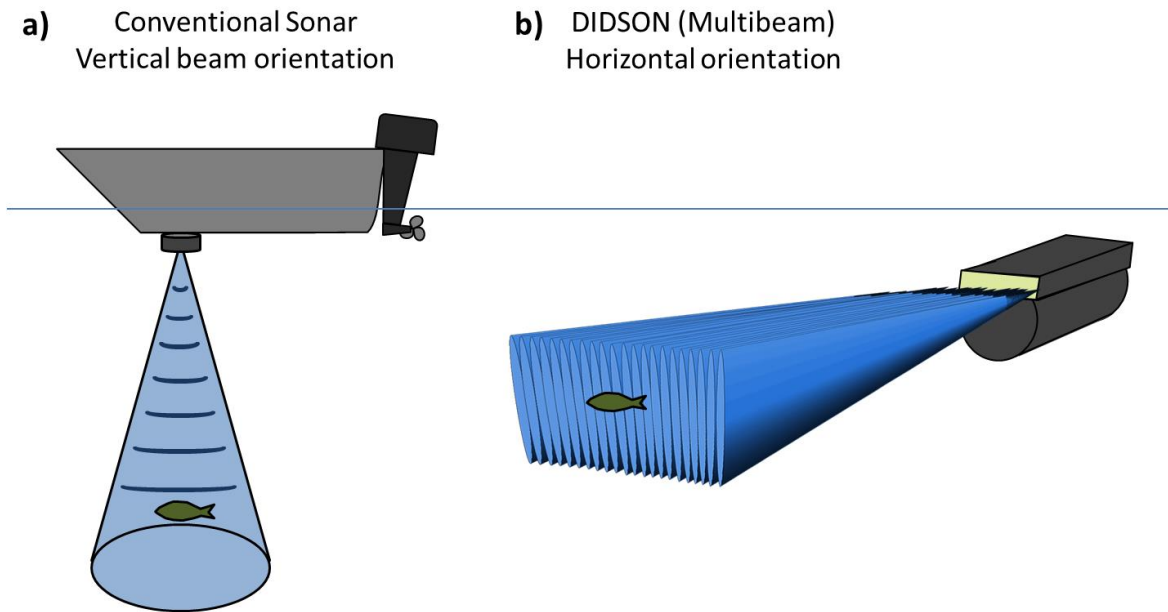


Figure 2. a) Schematic vertical beam orientation of surveys with conventional sonar echosounder application. b) Usual horizontal beam orientation of DIDSON monitoring application.

Following a different approach, the DIDSON is typically used in a fixed horizontal application, mounted on a pan-and-tilt unit to allow the adjustment of the field of view. In contrast to conventional echosounders the DIDSON can be described as an acoustic camera due to its ability to generate pictures from underwater, which are comparable to images obtained by optical systems. This is facilitated by the use of up to 96 single-beams (0.3° horizontal x 14° vertical each) forming a beam-array with an opening angle of 29° horizontally x 14° vertically (Fig. 3 a)). The close horizontal alignment of the narrow beams in the array enables the scan of the environment with a high resolution. The DIDSON can be operated in two frequencies, the detection mode at 1.1 MHz (ranges up to 40 m) and the identification mode at 1.8 MHz (ranges up to 15 m).

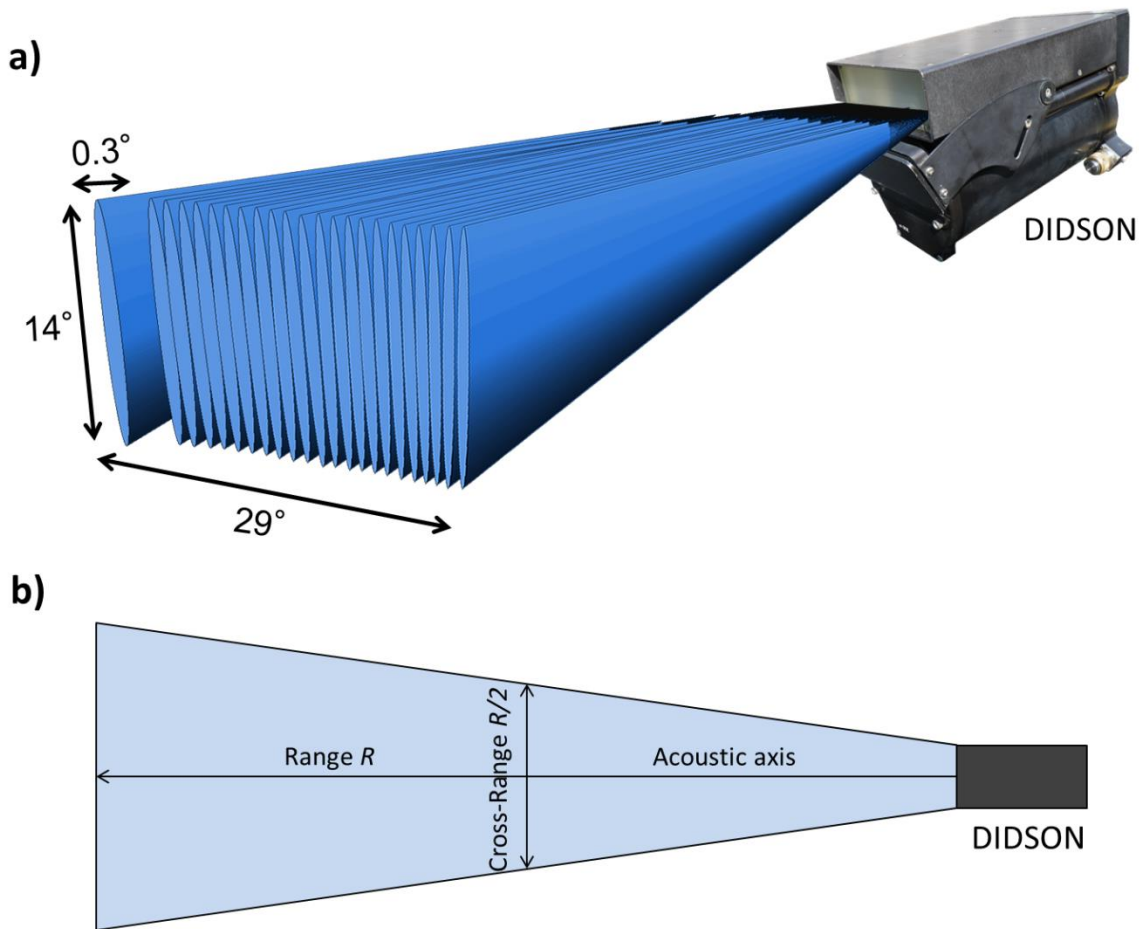


Figure 3. a) Schematic illustration of the beam-array spanning the field of view. b) Top view of the beam with range- and cross-range axis marked.

As described above such high frequencies permit a high resolution. Medical ultrasound devices work on shortest ranges, with even higher frequencies between 6-8 MHz. In the detection frequency the single beams dimensions are $0.6^\circ \times 14^\circ$ forming the same field of view with $29^\circ \times 14^\circ$ with a lower resolution. Frame rates up to 15 frames s^{-1} deliver video like sequences. Although the obtained video sequences have a 3D appearance, the sonar delivers just 2D information. The DIDSON beam is defined by the range-dimension and the cross-range dimension (Fig. 3 b)). The resolution can be calculated for both by calculating the size of the “acoustic pixels” defined by the 512 pixels in the range-dimension and the 96 beams in the cross-range- dimension. Because the number of range pixels is fixed, the height of the pixels increases with a larger range adjustment (called “window length” in the sonar topside software). Hence, the range resolution decreases with longer window length adjustments. The width of the pixels increases with the increase of the range due to

the opening of the single beams. Hence the cross-range resolution is higher in closer proximity to the emitter than far away. Closer to the sonar the beams are still thinner, allowing a higher resolution. The highest resolution can therefore be obtained by observing objects in closer range to the sonar with a small window length setting.

The formulas to calculate the sonars resolution at a certain range are as followed (Soundmetrics Corp. – www.soundmetrics.com) (Here for the example of 1.8 MHz and 96 beams):

Range resolution

Height of the acoustic pixel [cm]:

$$H = \frac{R [cm]}{512}$$

H is the acoustic pixel height. R is here defined as the range set to be covered in the sonar viewer (window length). For example a window length setting of 5 m results in a pixel height of about 1 cm ($\frac{500 \text{ cm}}{512} = 0.98 \text{ cm}$).

Cross-range-resolution

Width of acoustic pixel at a certain range [cm]:

$$W = \frac{R^{\frac{1}{2}} [cm]}{96}$$

W is the acoustic pixel width. The cross-range at a certain range is defined as $R^{\frac{1}{2}}$.

For example the width of the acoustic pixel at a range of 5 m is 2.6 cm:

$$\left(\frac{500^{\frac{1}{2}} [cm]}{96} = 2.60 \text{ cm}\right)$$

The horizontal application of the sonar results in pictures seen with a bird view perspective on the horizontal “plane” covered by the sonar beam-array similar as schematically illustrated in Fig. 3 b). One has to consider that there is no information about the exact position of a target in the 14° of the vertical domain. The position in

the horizontal (cross-range dimension) can be measured. It is defined by the angular deviation to the acoustic axis defining the center of the beam array (Fig. 3 b)).

The DIDSON possesses a specially designed ping cycle to avoid cross talk among the 96 single beams. To avoid adjacent beams to listen to pings from others, one frame (image) is established by a ping cycle where always 12 single beams, in a total of 8 bundles, are “fired” in a certain sequence resulting in a clear image (8x12 beams=96 total).

The DIDSON is more or less insensitive to turbidity and independent of light. The ability to produce high resolution live images in motion from underwater environments enables new approaches to assess freshwater ecological questions which were so far challenging to assess with optical or classical sampling methods.

DIDSON data processing (Video and echogram)

The following section gives a short description of the general data processing and analysis that was basic for all 3 chapters comprising the present dissertation. Further analyses and data processing is described in detail in the respective chapters.

A great advantage beside the video like sonar sequences is the possibility to generate echograms from DIDSON data. This can be realized with DIDSON topside software and third party software. Echograms are the standard presentation form for sonar data. They are characterized by a plot of received echo intensities over the range as a function of time. In the present study Sonar 5 Professional (Balk & Lindem, 2010) was used to convert DIDSON data into maximum intensity echograms. Maximum intensity echograms plot the intensity maximum of all beams. This kind of echogram is characterized by a sharp and detailed display of the data. Echograms have the advantage to show a larger period of recorded sonar data at a glance and are therefore suitable to screen data for fish echoes. Fish traces can be located in the echograms and real-time video analysis can be minimized by skipping data containing no targets. Fig. 4 shows a maximum intensity echogram containing fish traces together with the corresponding video data. Fish can be marked, measured and stored in a database for further data analysis. Depending on the scientific questions, a variety of other measures can be assessed from the sonar data with the Sonar 5 Professional software.

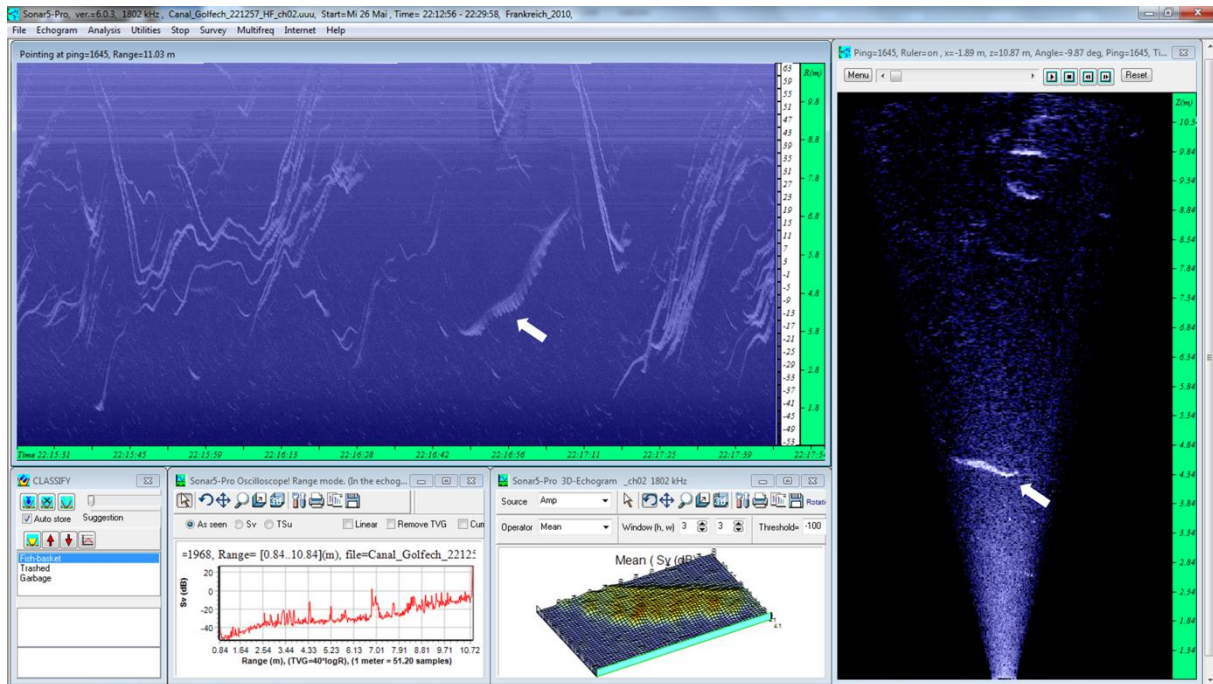


Figure 4. Workspace of Sonar 5 Professional. The upper window in the left shows a maximum intensity echogram together with the corresponding sonar video sequence (right window). The white arrows mark one fish trace in the echogram and the image of the fish in the right.

Results and Discussion

CHAPTER I

Can acoustic shadows identify fish species? A novel application of imaging sonar data

Abstract

This study addresses a fish species discrimination method based on normalised elliptic fourier descriptors applied to acoustic shadows derived by Dual-frequency Identification Sonar (DIDSON). Acoustic shadows of templates (20 cm, 30 cm, 40 cm and 50 cm) and live fish of four species (bream, *Abramis brama* (L.); barbel, *Barbus barbus* (L.); chub, *Leuciscus cephalus* (L.); trout, *Salmo trutta* L.) were projected on a plate in an experimental setup and tested on suitability for species discrimination. Twenty-centimetre templates were correctly classified in 97.5 % of the cases, indicating a size threshold. The larger templates reached values of 100 % correct classification based on cross-validated discriminant function analysis. It was also possible to classify moving fish based on screenshots of their acoustic shadows with a certainty of 83.9 %. Extended field tests are required to evaluate the method for use in practical monitoring applications in multi-species river environments.

Introduction

One of the most challenging hydroacoustic issues is remote fish species identification. The majority of related research has been carried out in the marine environment (Scalabrin et al., 1996; Simmonds et al., 1996; LeFeuvre et al., 2000; Hannachi et al., 2004; Korneliussen et al., 2009; Charef et al., 2010; Robotham et al., 2010). Horne (2000) reviewed the diversity of acoustic approaches in species identification concluding that definitive identification of aquatic animals currently is not possible by exclusive use of sound. Rogers et al. (2004) addressed the acoustic identification of freshwater fish species in Lake Huron and Lake Michigan. Although neural networking was able to deliver promising classification results from monotypic fish aggregation data, it did not perform satisfactorily for *in situ* fish applications. Successful detection and identification of Chinese sturgeons in the Yangtze River was realised using a set of descriptors provided by data from a split-beam hydroacoustic system (Tao et al., 2009). However, compared with the efforts spent on marine fish species identification, freshwater applications, especially aimed at acoustic single fish identification, still remain a poorly investigated field.

There are many studies addressing non-intrusive monitoring methods. Fishery biologists are often faced with turbid conditions in combination with deep water, where many methods are supposed to be limited or not applicable. Video-based species recognition can be successful under certain conditions (Cattoen et al., 1999; Lee et al., 2004; Zion et al., 2007), but 15 m is the maximum range that can be achieved with optical systems in clear water with appropriate lightning. Unfortunately, conditions in aquatic environments are often affected by high turbidity that can decrease the visibility to a fraction of a metre (Belcher et al., 2001). While visual identification and counting is a common fish estimation method in flat and clear waters (Locke, 1997, Holmes et al., 2006), hydroacoustic (Maxwell & Gove, 2007) and infra-red light based (Baumgardner et al., 2010) methods have also been investigated. However, tools and related methods that can be used for species discrimination under turbid conditions are needed.

DIDSON (Dual frequency IDentification SONar, www.soundmetrics.com) delivers near video-like, high quality live images almost independent of water turbidity and light (Belcher et al., 2001). Although it was originally developed for technical inspection and military purposes for underwater observation and target identification, application in fisheries and aquatic research has increased since the launching of the

system (Tiffan et al., 2004; Holmes et al., 2006; Maxwell & Gove, 2007; Frias-Torres & Luo, 2009; Han et al., 2009; Han & Uye, 2009).

Although the DIDSON delivers high quality images, general subjective fish species identification is arguable and depends on the presence of remarkable morphological features or striking echo signals. Eel exhibits special body architecture in combination with special swimming behavior (Gray, 1933; Webb, 1982) leading to recognisable patterns in the videos as well as in echograms. Mueller et al. (2008) presented a computer-driven process that classified DIDSON images of eel and debris with high accuracy.

Objects in the DIDSON beam array reflect and absorb a part of the emitted sound and, as a result, they cast acoustic shadows. These shadows are strongly related to the objects outer shape and thus should be investigated for use as a parameter for species identification. The aim of this study was to test whether it is possible to discriminate between fish species based on their specific acoustic shadows in a computer-driven process. Species identification based on DIDSON generated acoustic shadows would be a gain in data acquisition in hydroacoustic fish monitoring surveys. This study covers an experimental approach based on a four-species set of templates and an experiment involving live fish corresponding to the chosen species.

Methods

DIDSON serves as a tool to generate the acoustic shadows. It delivers video-like, high quality live images and allows *in-situ* control under challenging conditions where optical systems are limited. DIDSON features two operational frequencies. A low detection frequency (1.1 MHz) suitable for ranges up to 30 m and a high frequency identification mode (1.8 MHz) that serves ranges up to 15 m (www.soundmetrics.com). The horizontal field of view provides an opening angle of 29° consisting of 48 single beams with a width of 0.6° each at the lower frequency or 96 beams with a width of 0.3° at the higher frequency. The beam array provides a vertical opening angle of 14°. Based on this beam geometry, a cross-range resolution of 1 cm and a down-range resolution of 0.24 cm can be achieved for an object in 2-m range with a window length adjustment of 1.25 m in the high frequency mode. The down-range resolution is restricted to 512 pixels over a chosen window length.

Hence the resolution is supposed to be better when covering short distances (window length adjustment) in a preferably near distance.

Experimental setup

Two types of experiments were conducted to assess the quality of acoustic shadows to be suitable for fish species discrimination. A basic set of four morphological distinct species that can be found together in various German rivers was chosen to assess differences in acoustic shadows: bream, *Abramis brama* (L.), barbel, *Barbus barbus* (L.), chub, *Leuciscus cephalus* (L.) and trout, *Salmo trutta* L. or rainbow trout, *Oncorhynchus mykiss* (Walbaum) respectively. The two setups can be separated into an experimental setup (Fig. 1) involving 1-mm stainless steel, laser-cut templates with typical shapes of the four species (Fig. 2 a) and a mesocosm experiment in which acoustic shadows of living fish were projected on a plate with respect to the four chosen species (Fig. 1).

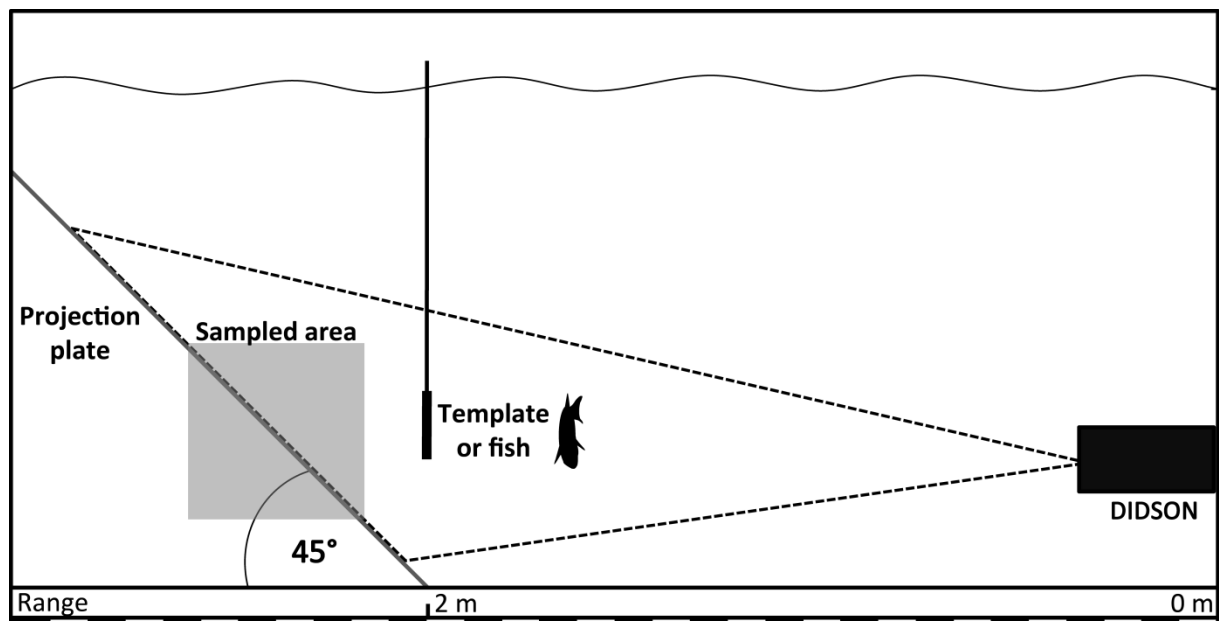


Figure 1. The DIDSON was installed parallel to the bottom and aimed at the 45° projection plate. Templates were positioned in the center of the beam array. Fish were able to cross the beam array in the same manner.

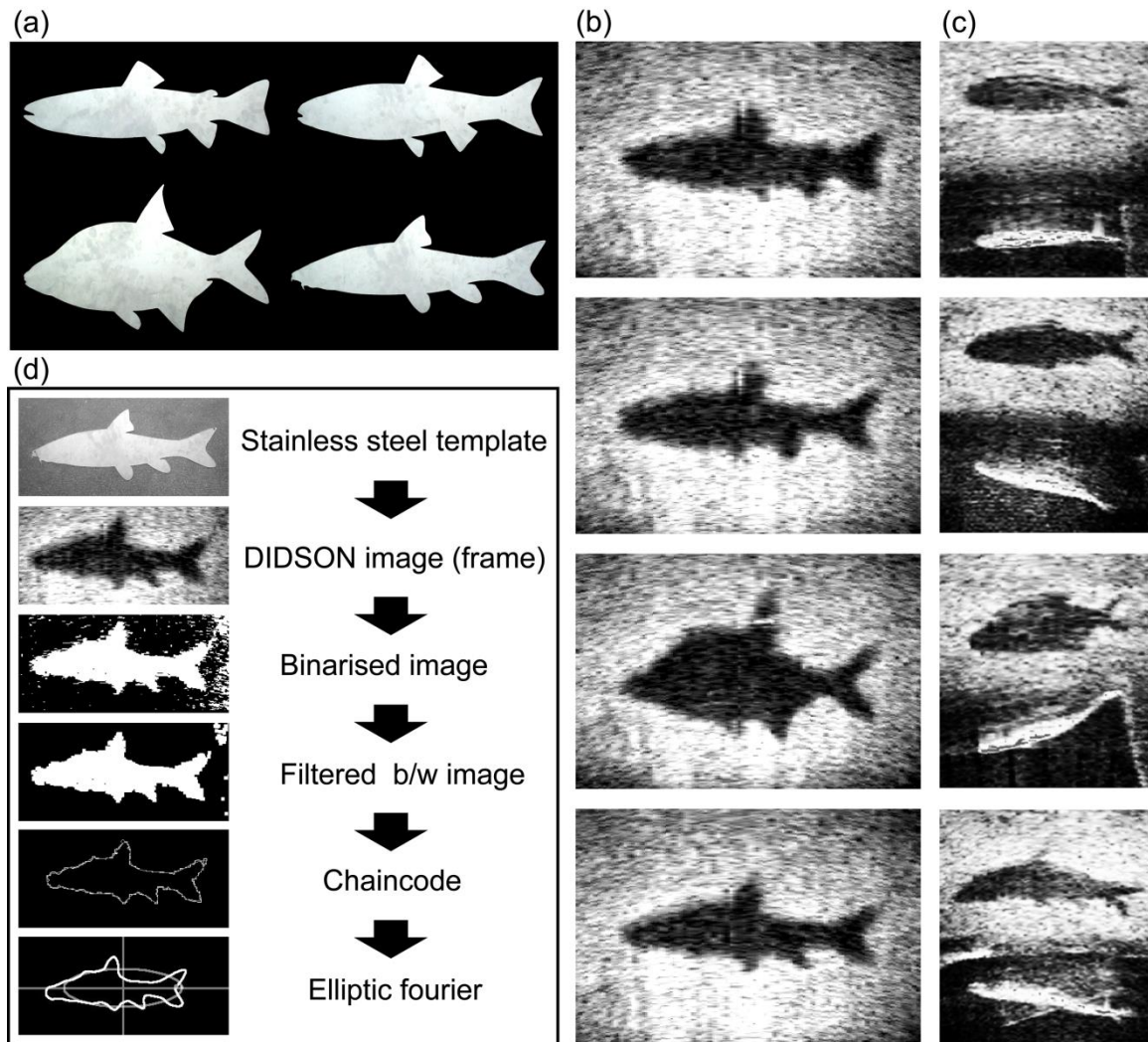


Figure 2. (a) Stainless steel templates of the four species (b) Acoustic shadows of 50-cm stainless steel templates (trout, chub, bream, barbel (top down)) (c) Acoustic shadows of living fish (trout, chub, bream, barbel (top down)) (d) Image processing with SHAPE Ver. 1.3 (Iwata & Ukai, 2002).

The experiments were carried out in tanks equipped with a wooden projection plate that was installed at an angle of 45° relative to the horizontal plane. This adjustment provided unbiased shadows. Deviation away from the 45° angle naturally lead to either vertically compressed or extended shapes because of the beam geometry and the translation of acoustic wave rays into a two-dimensional image. The DIDSON was mounted on a remote controlled pan and tilt unit aimed parallel to the bottom (Pan/Tilt: $0^\circ/0^\circ$). Four sizes of templates (20, 30, 40 and 50 cm) per species were placed in the centre beam of the array between the DIDSON and the projection plate (Fig. 1). The distance between the emitter and the projection plate was 2 m

(measured to the bottom touching side of the plate). All templates were exposed to the same range.

Fish were stocked into the tank for the live fish experiment. Each species was recorded separately. Fish differed in size between 30 and 49 cm. A projection plate distance of 2 m was maintained to serve as an applicable range for future field tests, e.g. in front of a fish path. Data were recorded in high frequency mode (1.8 MHz) and with a window length adjustment of 1.25 m to obtain the highest possible resolution. The window start was set to 2.08 m. For live fish this value was set to 1.25 m to get distance information of the free swimming fish.

Shape analysis: template experiments

A total of 16 templates corresponding to the four species and the four different sizes were used. Data with echoes and shadows from each of the 16 templates were recorded for about 60 s using the DIDSON data acquisition software (V5.21.01) (Sound Metrics Corp.). DIDSON raw files were imported into Sonar5-Pro hydroacoustic data post-processing software (Balk & Lindem, 2010) and 20 sample frames were then chosen randomly from each of these 16 recorded files by capturing the screen in the paused videos. Images were saved in full-color, 24-bit bitmap format in MS Paint.

Image binarisation, Elliptic Fourier Analysis (EFA) and a Principal Component Analysis (PCA) were conducted using SHAPE analysis software (SHAPE Ver. 1.3, Iwata & Ukai, 2002; Fig. 2), and further statistical analysis was conducted in MS Excel and SPSS 18. The bitmap images of the shadows were opened in ChainCoder, which is part of the SHAPE software package. The sector of interest (Fig. 2) was cut out in the first step to discard undesired shades leading to artifacts during the subsequent edge detection. The red color channel of the 8-bit RGB (red, green, blue) image was chosen. In the next step, the image was converted into a black and white image (binarisation). The binarisation threshold was applied automatically. ChainCoder features cleaning algorithms (erosion and dilution filtering) that were used to remove noise. Finally, edge detection was applied to find the shadow contours. The closed contour was labeled and saved as chain code (Freeman 1974) in plain text format.

EFA, a method to approximate closed, two-dimensional contours of arbitrary shapes (Iwata & Ukai, 2002), was conducted with CHC2NEF, which is also part of the SHAPE software. The output contour file containing the chain-codes of the shadows was opened and the first 20 harmonics elected. The chain-code was then converted into Normalised Elliptic Fourier Descriptors (NEFDs), normalised with respect to size, rotation and starting point (Kuhl & Giardina, 1982). NEFDs were stored in a text file.

Principal Component Analysis (PCA) was used to focus the explanatory power into a few features because the NEFDs are numerous and thus difficult to use directly for classification. SHAPE offers the possibility to conduct a PCA by feeding the program PrinComp with the CHC2NEF-output files containing the NEFDs. PCA was applied and the output stored in text format.

To verify the output from the EFA, the variation explained by the principal components was visualised by drawing the principal component contours with the program PrinPrint (included in SHAPE 1.3).

In a next step, the PCA output was imported into MS Excel and prepared in spreadsheets. The PC scores, which represented the different features that have been recognised by the PCA to serve best as discriminant variables (between 4 to 6 significantly contributing PC scores with respect to the 4 template sets), were used in a cross-validated Discriminant Function Analysis (DFA) in SPSS 18 to classify the four species. A leave-one-out cross-validation was applied, where each case was classified by the functions derived from all cases other than that case.

The DIDSON images were relatively inhomogeneous respecting the unequal power allocation in the beam array. The highest backscatter was received from the middle of the projection plate because the bigger part of the power is emitted in the centre beam. This resulted in a DIDSON image with a bright central area, fading out to the borders of the beam. A maximum contrast between the object of interest (foreground) and the background was required to achieve best results because SHAPE uses binarisation to find an object's silhouette.

Shape analysis: live fish mesocosm experiments

Statistics were performed with 459 shadows (95 barbel, 120 bream, 125 chub and 119 trout). Shadows differed in size in relation to the position of the fish in the beam array. The image processing, Fourier analysis and PCA were conducted in SHAPE 1.3, in the same way as with the template data. Fish swimming more distant to the DIDSON unit produced smaller shadows than those swimming very near, analogous to objects in a beam of a torch light. Fish distances between 1.5 m and 2 m were included in the analysis. The bigger part of distances measured around 1.75 m. Thus, normalization played a larger role in this case as it equalized shadow sizes. This issue also plays an important role in the field. Based on the PC scores, a cross-validated DFA was performed in SPSS 18 to classify the shadows according to the correct species. Therefore the dataset was randomly mixed and split up into two subsample datasets. The first dataset served as a training dataset to calculate the discriminant functions. The second dataset, which was excluded to the discriminant model calculation and thus was unknown, was applied as a validation dataset to the discriminant functions derived from the first dataset to validate the classification power. Prior to the DFA, the extracted feature variables (PC scores) were tested on normal distribution by applying Kolmogorov-Smirnov and Shapiro-Wilk tests in SPSS 18. Because not all 12 variables exhibited normal distribution according to the tests, the error of deviation from a normal distribution was calculated in MS Excel and considered as negligible. DFA is considered to be relatively robust against infraction as long as the number of cases is high (McGarigal et al., 2000; McCune et al., 2002).

Results

Template experiments

The stainless steel templates proved to be well suited to project the fish silhouettes on to the projection plate. The quality of the shadows depended on template size. The 20-cm shapes were hard to distinguish by the human eye because of blurred contours. Only the deep-bodied bream shadows were identifiable. However, the four species could be identified by statistical analysis. For the remaining template sizes (30 cm, 40 cm, 50 cm), species-specific features of the shadows were more obvious (Fig. 2b) and revealed statistically significant differences between the species. PCA

revealed between 4 and 6 principal components that significantly explained the largest amount of difference in the 4 sizes of templates. In all cases the first two principal components cumulatively explained between 81.1 % and 86.5 % of the variance between the 4 species. The cumulative values ranged between 91.6 % and 95.2 % for all significant contributing PC scores (Table 1). PCA separated the shape data into four distinct clusters.

A cross-validated discriminant function analysis was fed with the principal component scores of each size set of the four species. In all cases separation of the four data clusters was observed. The clusters of barbel and trout were less distant for the 20, 30 and 40-cm templates. The results for the 50-cm templates showed a greater similarity for the chub and barbel shadows. The cluster of the bream-related shadows separated far away from the other species clusters in all cases (Fig. 3). The discriminant function analysis showed a correct classification rate of 100 % for all sizes of templates, except the 20-cm set that reach a value of 97.5 % (Table 2).

Table 1. Outputs of principal component analysis of various templates

Principal component	Eigenvalue	Proportion (%)	Cumulative (%)	> 1/77
20-cm template				
Prin1	2.56E-02	69.0	69.0	*
Prin2	4.49E-03	12.1	81.1	*
Prin3	1.37E-03	3.7	84.8	*
Prin4	9.81E-04	2.6	87.4	*
Prin5	9.03E-04	2.4	89.9	*
Prin6	6.46E-04	1.7	91.6	*
30-cm template				
Prin1	1.41E-02	69.1	69.1	*
Prin2	3.56E-03	17.3	86.5	*
Prin3	8.34E-04	4.1	90.1	*
Prin4	3.90E-04	1.9	92.5	*
Prin5	3.32E-04	1.6	94.1	*
40-cm template				
Prin1	1.47E-02	74.3	74.3	*
Prin2	1.95E-03	9.8	84.2	*
Prin3	1.59E-03	8.0	92.2	*
Prin4	4.18E-04	2.1	94.3	*
50-cm template				
Prin1	0.0072	60.5	60.5	*
Prin2	0.0026	22.1	82.7	*
Prin3	0.0009	7.8	90.5	*
Prin4	0.0004	3.2	93.7	*
Prin5	0.0002	1.5	95.2	*
Prin6	0.0001	0.9	96.1	*

* = significant

Canonical Discriminant Functions

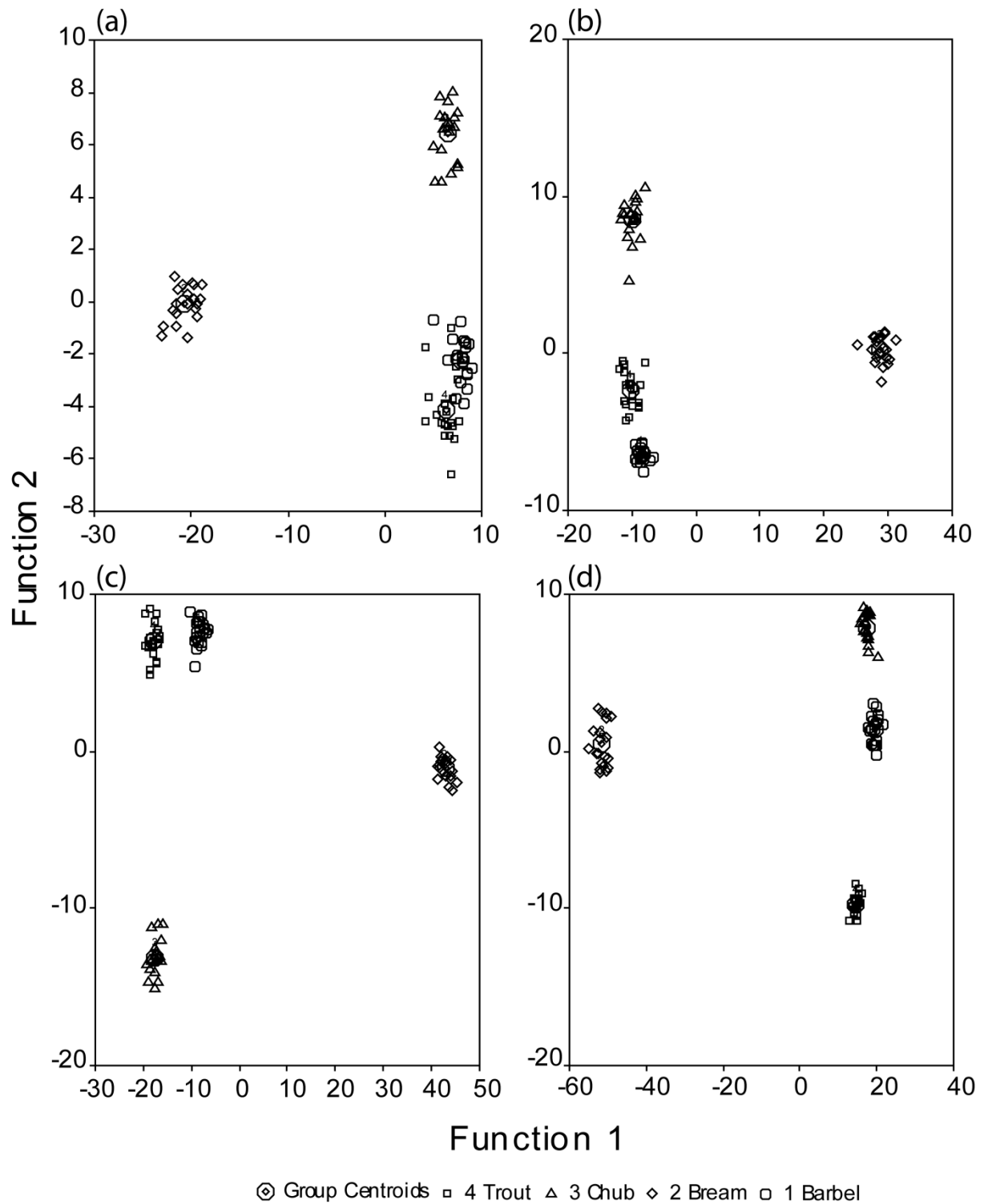


Figure 3. Discriminant function scatterplots corresponding to the four sizes of templates (a) 20-cm templates (b) 30-cm templates (c) 40-cm templates (d) 50-cm templates

Table 2. Classification results of discriminant function analysis of the 20-cm templates. In the cross validation, each case is classified by the functions derived from all cases other than that case. 100.0 % of original grouped cases were classified correctly. 97.5 % of cross-validated grouped cases were correctly classified.

		Predicted Group Membership					
		GROUP	1	2	3	4	Total
Original	Count	1	20	0	0	0	20
		2	0	20	0	0	20
		3	0	0	20	0	20
		4	0	0	0	20	20
	%	1	100	0	0	0	100
		2	0	100	0	0	100
		3	0	0	100	0	100
		4	0	0	0	100	100
Cross-validated	Count	1	20	0	0	0	20
		2	0	20	0	0	20
		3	0	0	20	0	20
		4	2	0	0	18	20
	%	1	100	0	0	0	100
		2	0	100	0	0	100
		3	0	0	100	0	100
		4	10	0	0	90	100

Live fish mesocosm experiments

Despite the relatively close borders inside the mesocosm, the DIDSON delivered good quality images with the chosen setup. In accordance with the given power allocation of the beam array, the image of the projection plate was brightest in the centre and faded to the borders. Thus, the processing of the projected shadows was affected in the matter of a higher artifact rate during binarisation when silhouettes crossed a wide range of the brightness gradient. However, the shapes of the four species could intuitively be identified by the observer. Subjective species recognition was easier when watching the videos than the screenshots. Trout exhibited compact silhouettes, and fins were rarely observable in the acoustic shadows. Only a V-shaped tail fin shadow was visible (Fig. 2 c). Chub silhouettes were also relatively compact but differed from trout because of visible dorsal, anal and ventral fins. The tail fin appeared larger and possessed a deeper V-shape (Fig. 2 c). Bream produced

striking silhouettes. Dorsal, anal, and ventral fins were observable as well as the deep notched tail fin. The high body depth was well projected on the plate and identified as bream in all cases (Fig. 2 c). The barbel shadows also exhibited a special shape. Silhouettes were characterised by a beaked head, a slender streamlined body, and visible dorsal, tail, anal and ventral fins (Fig. 2 c).

PCA revealed 12 principal components significantly contributing to the separation of the four species. The first three principal components explained 24.6, 16.8 and 11.3 % of the variance. Cumulatively, all significant PC scores contributed 85.3 % (Table 3). Each principal component is related to a characteristic variation in the shape. Fig. 5 visualizes the morphological variation corresponding to the first three PC scores.

Table 3. Outputs of Principal Component Analysis for live fish

Principal Component	Eigenvalue	Proportion (%)	Cumulative (%)	> 1/77
Prin1	5.36E-03	24.6	24.6	*
Prin2	3.67E-03	16.8	41.4	*
Prin3	2.46E-03	11.3	52.7	*
Prin4	1.65E-03	7.6	60.3	*
Prin5	1.26E-03	5.8	66.1	*
Prin6	8.95E-04	4.1	70.2	*
Prin7	8.50E-04	3.9	74.1	*
Prin8	8.06E-04	3.7	77.8	*
Prin9	5.50E-04	2.5	80.3	*
Prin10	4.66E-04	2.1	82.4	*
Prin11	3.30E-04	1.5	84.0	*
Prin12	2.93E-04	1.3	85.3	*
Prin13	2.80E-04	1.3	86.6	

* = significant

Similar to the results of the template experiments, the bream cluster separated furthest from the remaining three clusters. The clusters of barbel, trout and chub were partially overlapping. A subsequently applied cross-validated discriminant function analysis achieved a much better separation (Fig. 4). The crucial features obtained from the PCA (PC scores) served as the dataset. Bream and barbel clusters

claimed distinct regions in the plot with less overlapping. Trout and chub clusters partly shared the same room. However, the cluster formed by the chub data was comparably compact (Fig. 4). The cross-validated discriminant function analysis classified 83.9 % of the shadows to the correct species based on the unknown data set (Table 4 and Fig. 4).

Table 4. Classification results of discriminant function analysis for live fish. 91.3% of selected original grouped cases were correctly classified; 83.9% of unselected original grouped cases (validation dataset) correctly classified

		GROUP	Predicted Group Membership				Total
			1	2	3	4	
Cases Selected (Training dataset)	Count	1	45	0	5	3	53
		2	0	59	0	0	59
		3	0	0	49	9	58
		4	1	0	2	56	59
	%	1	84.9	0	9.4	5.7	100
		2	0	100	0	0	100
		3	0	0	84.5	15.5	100
		4	1.7	0	3.4	94.9	100
Cases Not Selected (Validation dataset)	Count	1	32	0	6	4	42
		2	3	55	3	0	61
		3	0	0	58	9	67
		4	1	0	11	48	60
	%	1	76.2	0	14.3	9.5	100
		2	4.9	90.2	4.9	0	100
		3	0	0	86.6	13.4	100
		4	1.7	0	18.3	80	100

Canonical Discriminant Functions

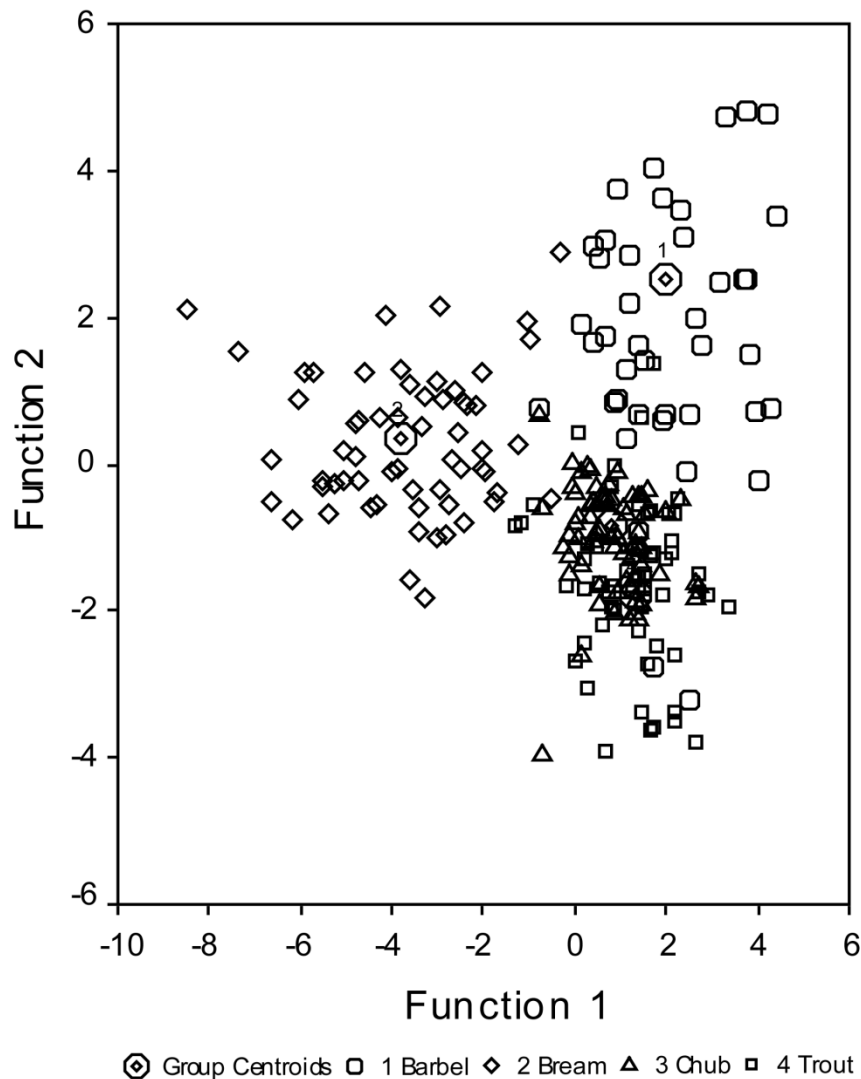


Figure 4. Discriminant function scatterplot of living fish data showing the clusters of the four different species.

Discussion

The results suggest the quality of acoustic template and fish shadows are appropriate for fish species discrimination. Template shadows were classified correctly in almost all cases. The classification rate was only 97.5 % in the 20-cm templates, (Table 2) indicating a size threshold, thus, templates smaller than 20 cm are supposed to produce inferior classification results. Template shadows appeared to be accurate replications of the stainless steel models. Fins were projected correctly for the larger templates (30 cm, 40 cm, 50 cm), but a loss in detail was observed with declining template sizes. Differences in fin exposure influenced

projected shadows of the live fish. Stagnophilic species like bream exposed a larger part of their fins than rheophilic fish like trout, which exhibited the most compact shadows. Presence or absence of fins appeared to be a characteristic feature. Body depth is another feature because of the highest separation of bream in both experiments. However, the mesocosm experiments demonstrated that it is possible to classify moving fish based on screenshots of their acoustic shadows.

Despite the heterogeneous brightness reflected by the projection plate and fish angle related bias, data quality was good enough to reach a correct classification rate of 83.9 % for the unknown validation data set (Table 4). The type of morphological variation remains uncertain because the PCA is based on chain code derived NEF descriptors rather than on manually measured shape descriptors. However, the morphological meaning of the variation could be evaluated by analysing the reconstructed shapes corresponding to each PC score (SHAPE: PrinPrint software). Although this analysis has a subjective component, PC1 seemed to be associated with body depth of the live fish. Variation in dorsal and ventral body shape was explained by PC2 and tail fin variations appeared to be related to PC3 (Fig. 5). This interpretation explained the striking separation of bream shadows because of the high body depth, visible dorsal and ventral fins and the deep notched tail fin (Fig. 2 c).

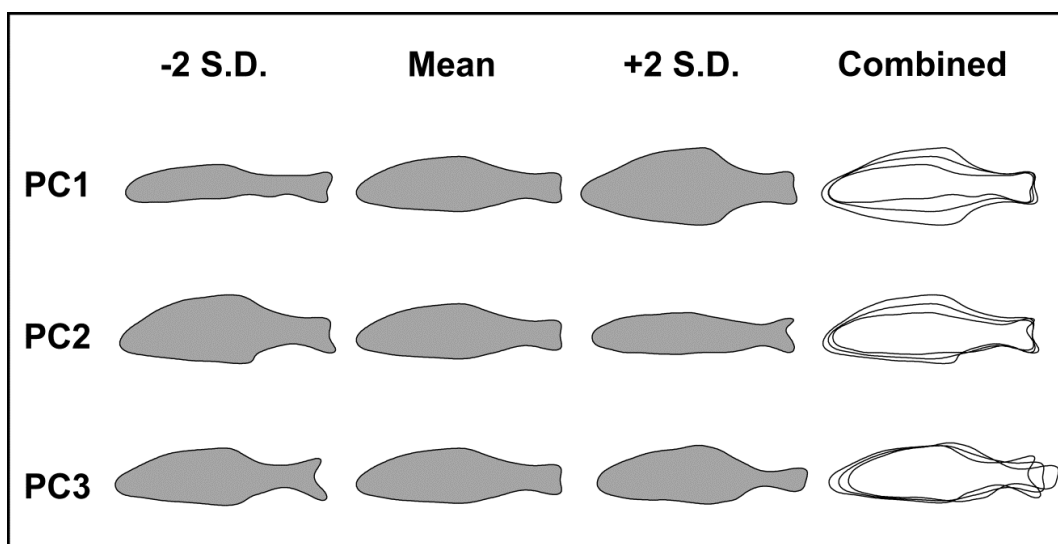


Figure 5. Shape variations corresponding to the PC scores.

Fourier methods serve as a powerful tool in a wide field of disciplines to discriminate and classify objects (Wallace & Wintz, 1980), species of plants and animals or other biological structures (e.g. Iwata et al., 2002; Hiraoka & Kuramoto, 2004; Truong et

al., 2005) based on their outer shape. Hence, the linkage of high resolution sonar images with the SHAPE software or other elliptic Fourier methods is an important step into semi-automated processes of individual fish species identification. Promising results in classification of DIDSON images were made earlier by applying neural network-based image processing methods to acoustic shadows of different underwater objects. (Kim et al., 2008). Compared with classical fisheries methods, e.g. fyke netting or electric fishing, acoustic methods are a non-invasive way of monitoring fish. This is a reason why research and application of sonar based observations in fisheries is important and gaining increasing attention. DIDSON has proved to be a powerful tool for accurate fish counting and sizing applications (Maxwell & Gove, 2007; Burwen et al., 2010). However, among counting and sizing, the setup used in this study is appropriate to acquire additional important data, e.g. species identification.

The drawback of missing 3D information in DIDSON data can be compensated by using acoustic shadows to calculate depth of migrating fish (Balk & Lindem, 2010; Sonar5-Pro: Help files). Hence, in further research an *in-situ* field test (e.g. in front of a fish pass or at other comparable bottlenecks) is necessary. Assessing the number and length of species accepting a fish pass or migration bypass is necessary to evaluate their functionality. However, in a practical monitoring application new challenges and problems will arise. For example because for the given sonar resolution, the discrimination method is limited to larger fish; in this study the smallest templates measured was 20 cm. Manual, individual discrimination of this size was not feasible. The computer-driven process was, however, still able to distinguish the four species at 97.5 % accuracy for the templates. The classification rate for live fish of this size is expected to be less exact because of the declining potential of fin detection in the shadows with the current sonar resolution. Furthermore, the higher number of different species and the possibility of merged shadows in the case of schooling fish have to be considered in a natural environment. Compared with the selectivity of different classical fisheries sampling methods, limits can be expected even for the approach presented in this study.

A low frequency application of the setup was supposed to be less successful and thus has not been conducted. However, because of the unexpected high classification rate of the 20-cm templates a test with large species at far ranges could

be challenging. To improve and enhance the species classification ability, it would be useful to create a database with DIDSON acoustic fish shadows of different species. A combination of other classification features (e. g. size and swimming behavior) extracted from DIDSON data is an additional alternative. Variation in tailbeat frequency is another potential discriminant feature (Mueller et al., 2010).

Acknowledgements

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CHAPTER II

Spawning behaviour of Allis shad *Alosa alosa* in the Garonne River - new insights based on imaging sonar data

Abstract

Spawning behaviour of *Alosa alosa* was observed by high resolution imaging sonar. Detected clouds of sexual products and micro bubbles served as a potential indicator of spawning activity. Peak spawning time was between 0130 h and 0200 h at night. Increasing detections over three consecutive nights were consistent with sounds of mating events (“bulls”) assessed in hearing surveys in parallel to the hydro acoustic detection. In 70 % of the analysed mating events there were no additional *A. alosa* joining the event whilst 70 % of the mating events showed one or two *A. alosa* leaving the cloud. However, in 31 % of the analysed mating events three or more *A. alosa* were leaving the clouds, indicating that matings are not restricted to a pair. Imaging sonar is suitable for monitoring spawning activity and behaviour of anadromous clupeids in their spawning habitats.

Introduction

Allis shad *Alosa alosa* (L. 1758) is an anadromous clupeid species with an original distribution range from Iceland to Morocco, covering the western European coasts (Aprahamian et al., 2003). Like most European migratory fish species, formerly abundant, *A. alosa* populations suffered under increasing commercial fishing pressure, water pollution, habitat degradation and construction of dams (Limburg & Waldman, 2009). In the middle of the 20th century populations experienced dramatic declines (De Groot, 2002). As a protected species, *A. Alosa* is listed in appendices of the European community directive on the conservation of natural habitats and wild fauna and flora and the Bern Convention. In the last decade, remaining populations especially in the Gironde system have declined seriously and are now characterized as vulnerable (Rougier et al., 2012).

A reintroduction program (EU-Life) according to the guidelines of the International Union for Conservation of Nature (IUCN) was established in 2007 to resettle *A. alosa* in the Rhine system (LIFE 06 NAT/D//000005). Individuals from the Gironde-System served as the basis for breeding *A. alosa* larvae for a restocking program (Klinger, 2011). Although *A. alosa* is the focus of recent research, detailed knowledge of individual spawning behaviour is still lacking.

Monitoring *A. alosa* spawning activity and behaviour is challenging. Counting of mating events (bulls) by hearing surveys has become a common method to determine spawning activity and estimate population developments over the years (Cassou-Leins & Cassou-Leins, 1981; Boisneau et al., 1990; Acolas et al., 2006; Caut et al., 2006; S. Gracia & I. Caut, unpublished data). First assessments with direct hearing of mating related sounds were made by Cassou-Leins & Cassou-Leins (1981) while the first automatic system was used by J.F. Trouilhet (unpublished data). Scientists acoustically record the environment close to known spawning grounds or count the loud sounds generated by each mating *in-situ*. This method delivers a measurement index for spawning activity. However, the method lacks information concerning the individual behaviour of *A. alosa* and the number of individuals involved in a mating event.

Gaining detailed information about spawning behaviour in the water column at night is complicated. Optical cameras depend on sufficient ambient light and have a relatively limited range under water. Telemetric methods, however, can deliver further information about individual fish distribution in space and time. Acolas et al. (2004)

revealed information about how often male and female *A. alosa* participated in mating events and their residency times on a spawning ground using acoustic tracking. For their study period, observed females and males were estimated to be involved individually in 0-2 and up to 60 mating events, respectively. A maximum number of 16 participations in one night was measured for a male individual. However, it remains difficult to assess how *A. alosa* interact with each other and how many individuals are involved in one mating event. Cassou-Leins et al. (2000) described how *A. alosa* ascend close to the surface to spawn. *Alosa alosa* then exhibit a specific swimming behaviour by rapidly moving in circles keeping their flanks close to each other in order to release sperm and eggs into the water column. To gain a more detailed picture about *A. alosa* behaviour under the surface, hydro acoustic observations were conducted at a known spawning site in the Garonne River (France). A Dual-Frequency-Identification-Sonar (DIDSON - Soundmetrics Corp. [<http://www.soundmetrics.com>]) served for data acquisition. It is independent of light and insensitive to turbidity by delivering video like sequences (Belcher et al., 2001) with monitoring ranges up to 30 m. Imaging sonar technology has been used successfully in a variety of studies to analyse fish behaviour (Tiffan et al., 2010; Rakowitz et al., 2012; Jůza et al., 2013; Price et al., 2013; Becker & Suthers, 2014; O'Connell et al., 2014). In the present study the imaging sonar was tested for its suitability to monitor *A. alosa* spawning by means of detection and quantification of mating events.

Material and Methods

Data sampling

The study was conducted in the Garonne River (Golfech) in France close to a known *A. alosa* spawning site. The area was situated approximately 1 km downstream of a hydro power plant [Canal de Golfech; latitude: 44° 06' 45.61" N; 0° 50' 32.26" E., altitude: 54 m]. In this section the width of the canalised river measured approximately 80 m.

Parallel to the hydro acoustic observations, nocturnal hearing surveys were carried out over the reproduction period of *A. alosa* in order to estimate spawner stocks (Carry & Tauzin, 2011). The species specific sounds generated by each couple when circling at the surface during the mating event were manually heard and counted.

Sound data were used to compare with the results delivered by the sonar. Water temperature and discharge were measured at the power plant.

The hydro acoustic study was conducted between 31 May 2010 – 3 June 2010. Dual-frequency-identification-sonar (<http://www.soundmetrics.com>) features high resolution acoustic images, flexibility in frequency and range settings and the capability to analyse raw data with third party post processing software (Langkau et al., 2012). The field of view was characterized by a horizontal opening angle of 29° and a vertical opening angle of 14°. At the high frequency (1.8 MHz) the array was formed by 96 single beams with a width of 0.3° and at low frequency (1.1 MHz) by 48 beams with a width of 0.6° respectively.

The sonar was fixed on a pan-and-tilt unit and installed on a stable steel shore mount for a number of hours during the 3 nights. The mounting featured a walking bridge to overcome the macrophyte belt adjacent to the shore line. The sonar was mounted in a distance of approximately 3 m from the shore at a depth of 1.5 m. Tilt was adjusted to cover the water volume close to the surface to include the important proportions of the water column where *A. alosa* spawning was expected to occur. The slope of the shore allowed the tilt to be set close a horizontal alignment ($0^\circ \pm 5^\circ$) without recording bright static bottom echoes. Thus, no background subtraction algorithm had to be applied. Although the DIDSON was placed at a spawning site, mating activity peaked at different ranges and areas around the mounting. The study was focussed on both the spawning activity and related behaviour of *A. alosa*. Therefore the pan was adjusted *in-situ* in order to increase amount of data for a numerical analysis of individuals participating in mating events.

Recordings were carried out between approximately 2230 h to 0230 h. Although the majority of the recordings were made in a window length of 20 m and a window start of 5 m in low frequency mode (1.1 MHz at 7 frames s⁻¹) some data were collected in high frequency mode (1.8 MHz at 6 frames s⁻¹) with window length of 10 m and a window start of 5 m. Further observations were made in a maximum range setting with a window length of 40 m and a window start of 5 m in low frequency mode at a frame rate of 4 frames s⁻¹.

Data analysis

Hearing data were cumulated by adding up the nightly numbers of estimated mating events and percentage was calculated. The DIDSON raw data were converted into maximum intensity echograms in the post processing software Sonar 5 Professional (Balk & Lindem, 2010). An echogram is a chart, plotting received sonar data over time. Typically the x-axis shows the time or the pings and the y-axis the covered range of the sonar system (Fig. 1 (a)). DIDSON videos typically show underwater live images in a bird view perspective when the sonar is mounted horizontally. Echograms were screened for mating related events, as they were visible in both echograms and video data. The authors discriminated between three phenomena: mating events “bulls”, clouds originated by “bulls” and mating attempts. The phrase “bulls” originates from the Garonne River and is related to the Occitan language (Cassou-Leins et al., 2000). It has been established among French scientists and describes a typical *A. alosa* mating with circling individuals. It was used here as a definition for a successful mating ending up with a cloud of sexual products visible in the sonar. Drifting clouds were defined as an observation related to a successful mating that has happened upstream the active field of view. Mating attempts described a behaviour of two *A. alosa* circling for a short time and parting without generating a cloud. Fig. 2 gives an overview of the basic phenomena in a schematic drawing. All detected events from the three categories were tracked in Sonar 5 Professional and stored with a time stamp.

Spawning activity and peak time were assessed by analysing the number of observations with respect to the defined categories. Data were divided into half hour time samples and pooled from the 3 nights. Metric measurements of the clouds were made with the polyline ruler tool in Sonar 5 Pro from a subset of mating events ($n=91$) where the end of the mating was visible in the beam. Diameter and perimeter (contour length) of the clouds were measured at the moment when the *A. alosa* were leaving the cloud by drawing a line through and a contour line around the clouds, respectively.

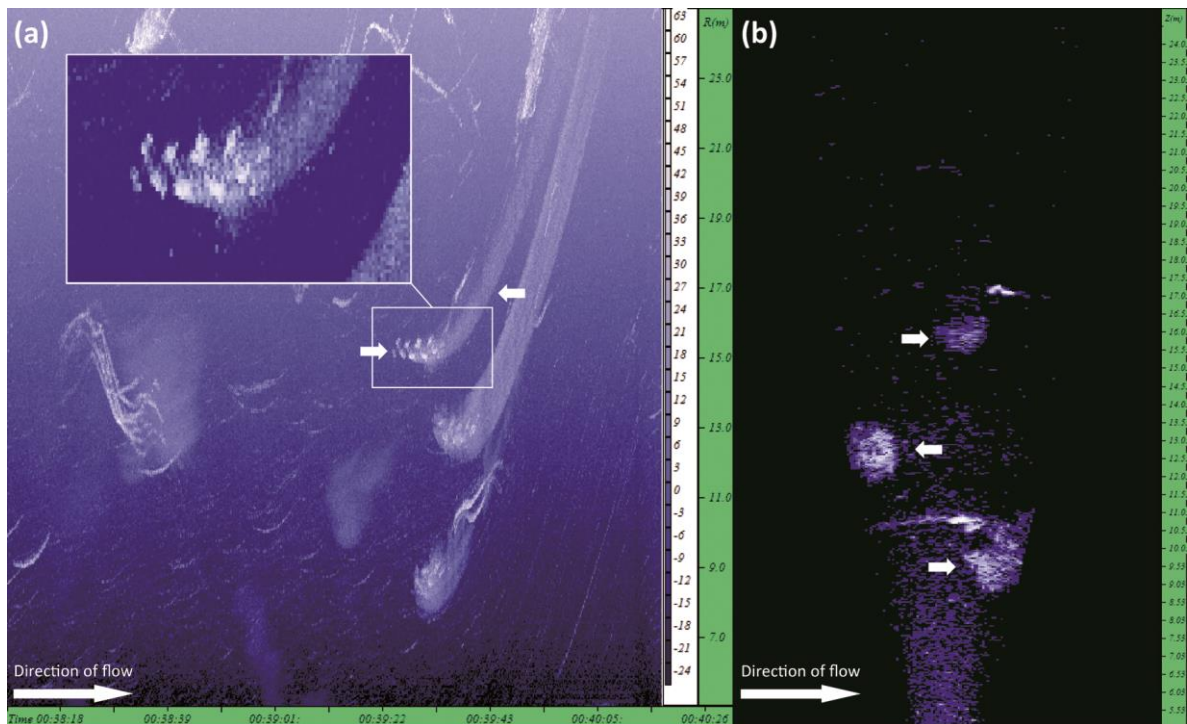


Figure 1. (a) Echogram of spawning *Alosa alosa*. The two arrows mark the typical zig-zag traces of the circling *A. alosa* and the corresponding cloud originating from the event (window length 20 m / window start 5 m / low frequency mode) The white rectangle shows the zoom image of the trace. (b) Corresponding video screenshot showing three clouds and individuals next to the upper and lower cloud.

A numerical analysis was carried out with $n=59$ videos taken from the subset. The criteria for this selection was a complete mating event in the continuous frame sequence or video. Data were analysed on the number of *A. alosa* initiating the mating event, the number of *A. alosa* joining each mating after the start and the number of individuals leaving the cloud. Therefore the mating event was separated into three steps: Step one: Start - *A. alosa* initiate the *mating event*., step two: Main phase: *A. alosa* spawn (with potentially joining individuals) and step 3: End: All *A. alosa* leave the mating event (Fig. 2 (a)). The maximum number of joining *A. alosa* and the maximum number of *A. alosa* involved in one mating event was derived. However, individuals entering or leaving the beam in the vertical direction were likely to remain undetected due to the smaller opening angle (14°) and lacking information of position in this domain.

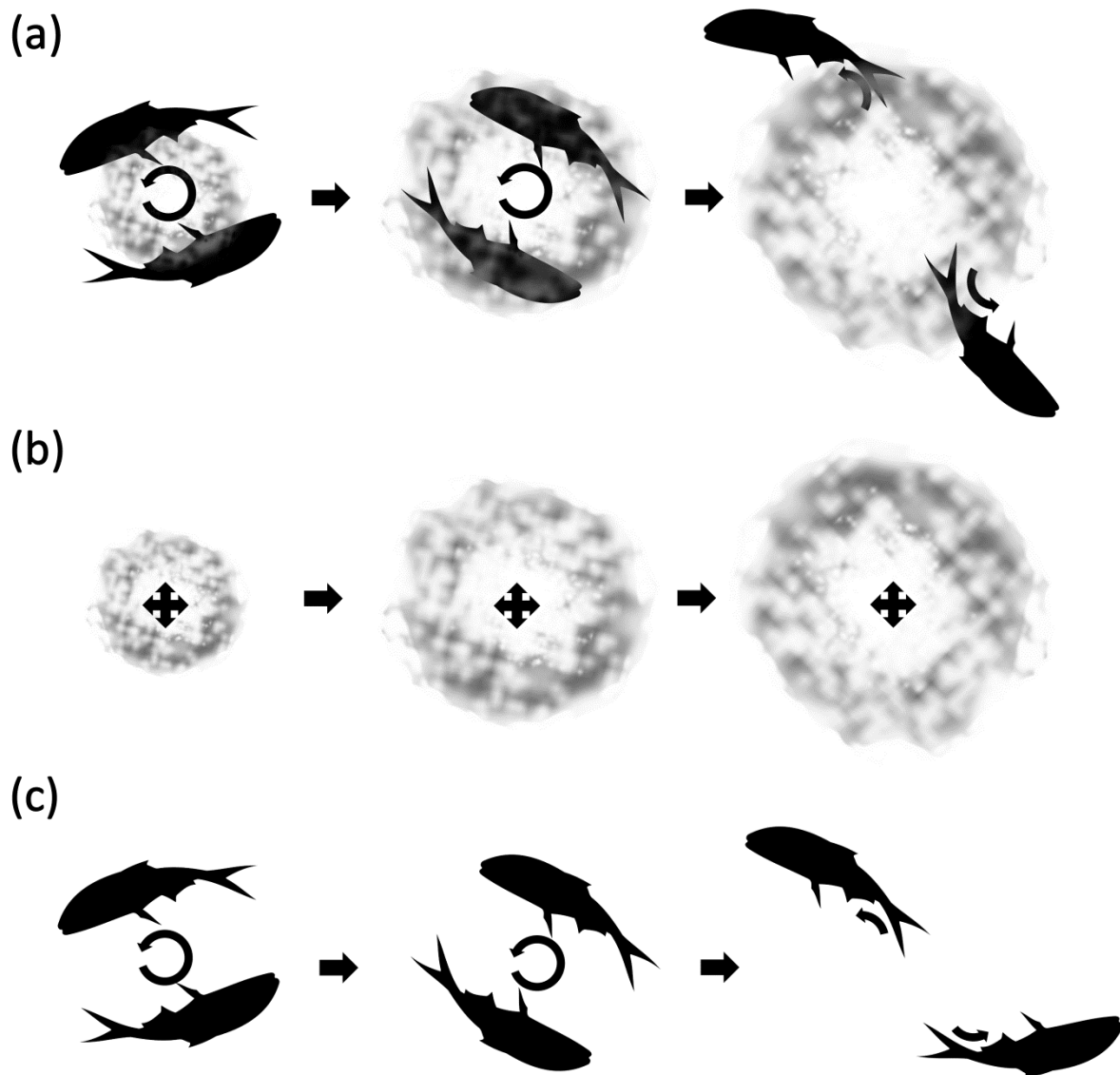


Figure 2. Schematic drawing of documented spawning phenomena. (a) Typical mating event performed by one male and one female. Individuals swim in a circle close to each other, release their sexual products in a growing cloud and leave. (b) Drifting cloud originated in a successful spawning (c) 'Mating attempt': Two *A. alosa* swim in a circle for a short time and part without generating a cloud.

Results

Based on the cumulative data of counted mating events at the spawning site (sound data), the DIDSON-study was carried out in the middle of the reproductive phase, at a progression level of approximately 50 % (Fig. 3). The mean water temperature during the hydro acoustic assessments was 17.5 ± 0.4 °C, while mean \pm S.D. discharge was 371 ± 27 m³ s⁻¹.

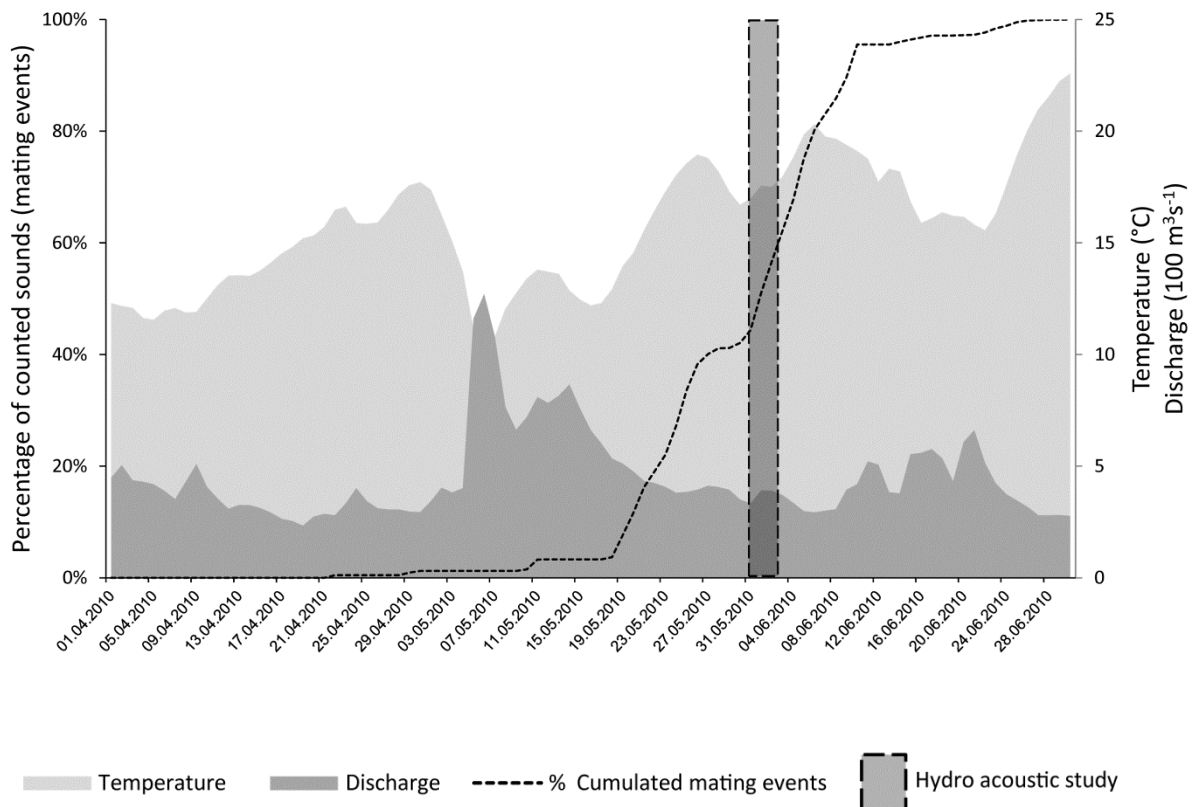


Figure 3. Period of the hydro acoustic study in relation to the level of the spawning season 2010 based on the cumulative percentage of mating events estimated by hearing surveys. Change in temperature ($^{\circ}\text{C}$) and discharge ($100 \text{ m}^3 \text{ s}^{-1}$).

The first mating events were detected after 2300 h. Pairs of *A. alosa* circled around each other rapidly for several seconds while releasing their sexual products into the water. Growing clouds of semen and eggs originating from the mating pairs could be visualized and were evaluated as successful spawning events (Fig. 1 (a,b)) and [supplementary video]. Mating events and clouds were detectable on all tested window length adjustments. Although short range settings delivered a higher resolution and hence revealed more details, it was still possible to observe individuals and mating clouds with a window length adjustment of 40 m and a window start of 5 m in low frequency mode (Fig. 4). This allowed the coverage of a larger area. Mating events were characterized by unique echo patterns in DIDSON maximum intensity echograms. Depending on the aspect angle the circling *A. alosa* produced 'zig-zag' traces resulting in a cloud. A pan adjustment perpendicular to the current produced the most striking patterns (Fig. 1 (a)).

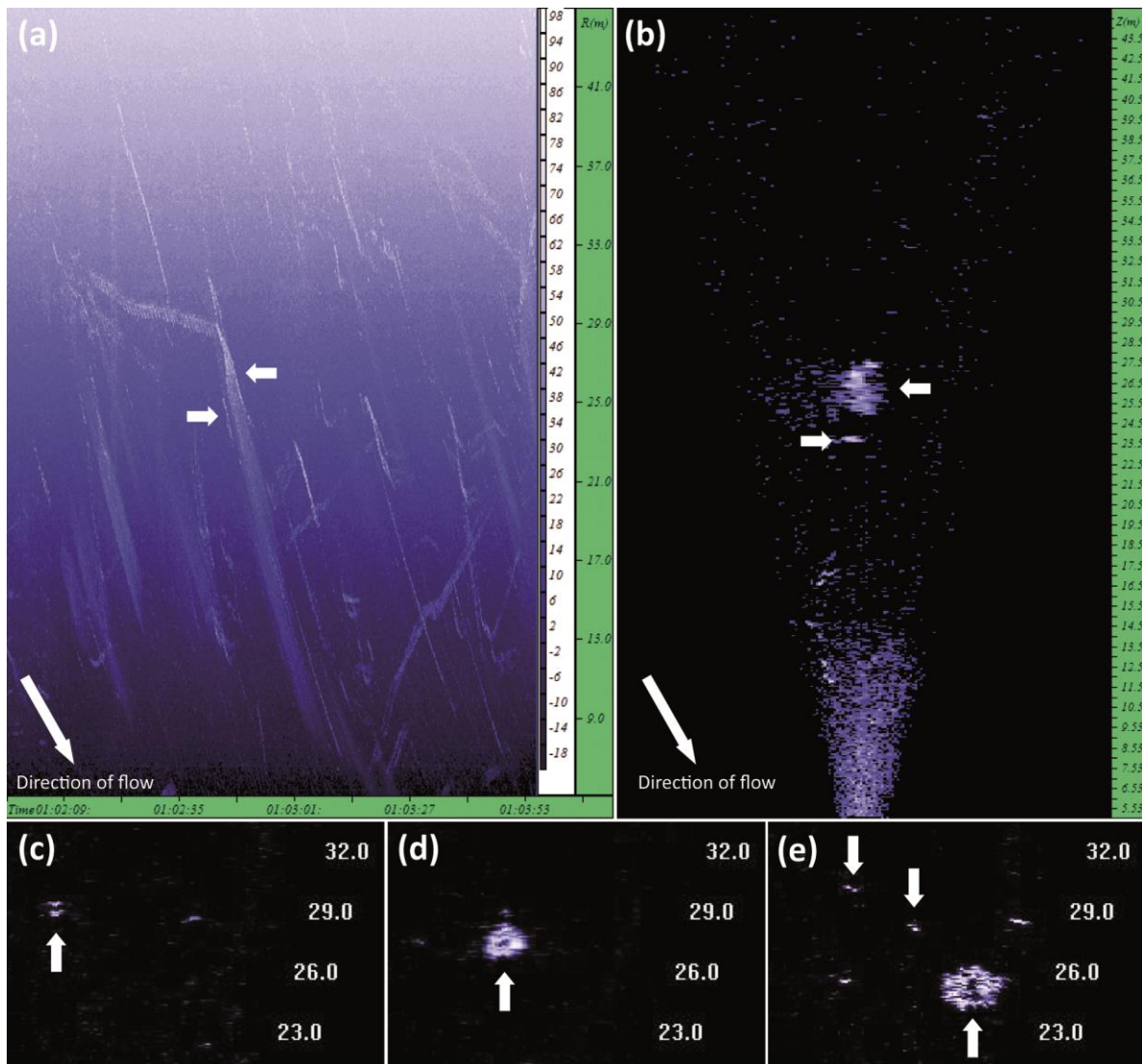


Figure 4. (a) Echogram with 40 m window length. White arrows mark a mating cloud and a single *A. alosa*. (b) Screenshot from DIDSON video with 40 m window length. Video screenshot corresponding to Fig. 2 (a). (c) – (e) Sequence of video screenshots of a mating at 40 m window length and 5 m window start adjustments at low frequency. (c) Two *A. alosa* starting to circle. (d) Growing cloud. (e) The arrows mark the drifting cloud and two individuals leaving. There are two more individuals which were not involved.

A total number of 171 mating events, 374 mating clouds and 47 mating attempts were detected during three consecutive night time observations. The spawning activity peaked between 0130 h and 0200 h (Fig. 5). The highest number of mating events (62) and clouds (120) and a low number of mating attempts (2) was observed during this period (Fig. 5). The count of mating attempts was comparatively low and reached its maximum (14) between 0100 h and 0130 h (Fig. 5).

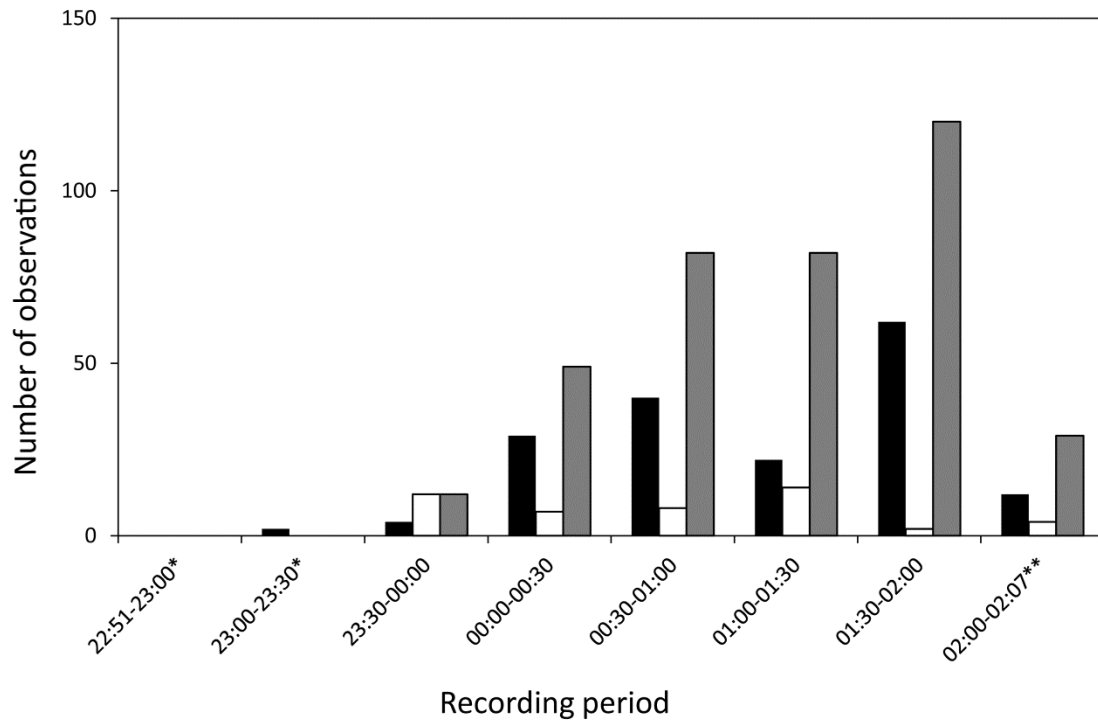


Figure 5. Number of observations (3 nights pooled) on mating events (■), attempts (□) and clouds (▣) in relation to the time of day (30 min recording periods) recorded between the 31 May and 03 June 2010 (*Period not recorded on 01.06.2010-02.06.2010 ** Period not recorded on 31.05.2010-01.06.2010).

During the 3 nights of hydro acoustic assessment the spawning activity increased, measured by the number of counted mating events and clouds in the sonar videos. This was consistent with the rise in activity depicted by the mating sounds, although the highest number of mating events was measured in the second night. (Fig. 6).

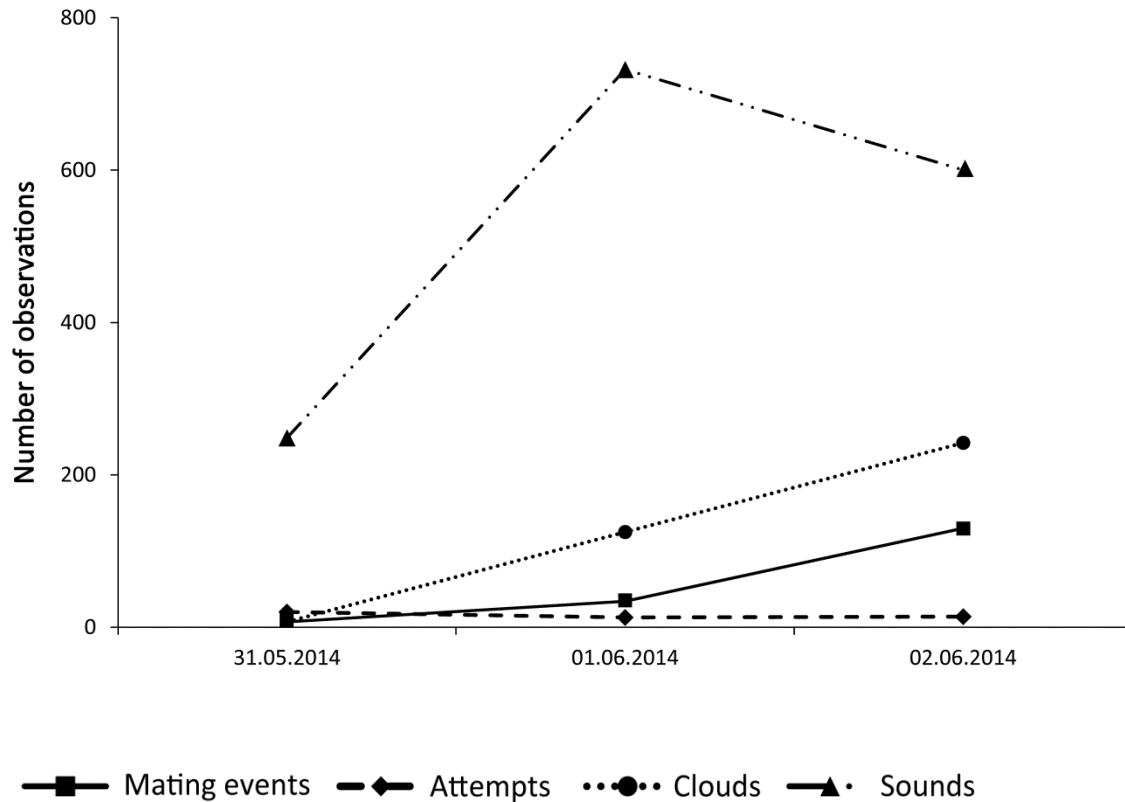


Figure 6. Trend of spawning activity based on sound assessments, and activity indices derived by hydro acoustic observation.

Although the majority of mating events were performed by one pair each (approximately 70 %), the numeric analysis revealed that there were more than two *A. alosa* involved in a mating event in some cases (Fig. 7 (a)). Almost 70 % of the matings showed one or two *A. alosa* leaving the mating event. However, a noticeable number of mating events were detected with three or even up to five *A. alosa* joining (approx. 31 %) (Fig. 7 (a)) and three or up to seven *A. alosa* leaving (approx. 31 %) one mating cloud (Fig. 7 (b)). Hence, the maximum number of *A. alosa* involved in one mating event was seven. The mean cloud diameter, measured when the *A. alosa* left the event, was 202 ± 61 cm and the mean perimeter was 956 ± 301 cm ($n=91$).

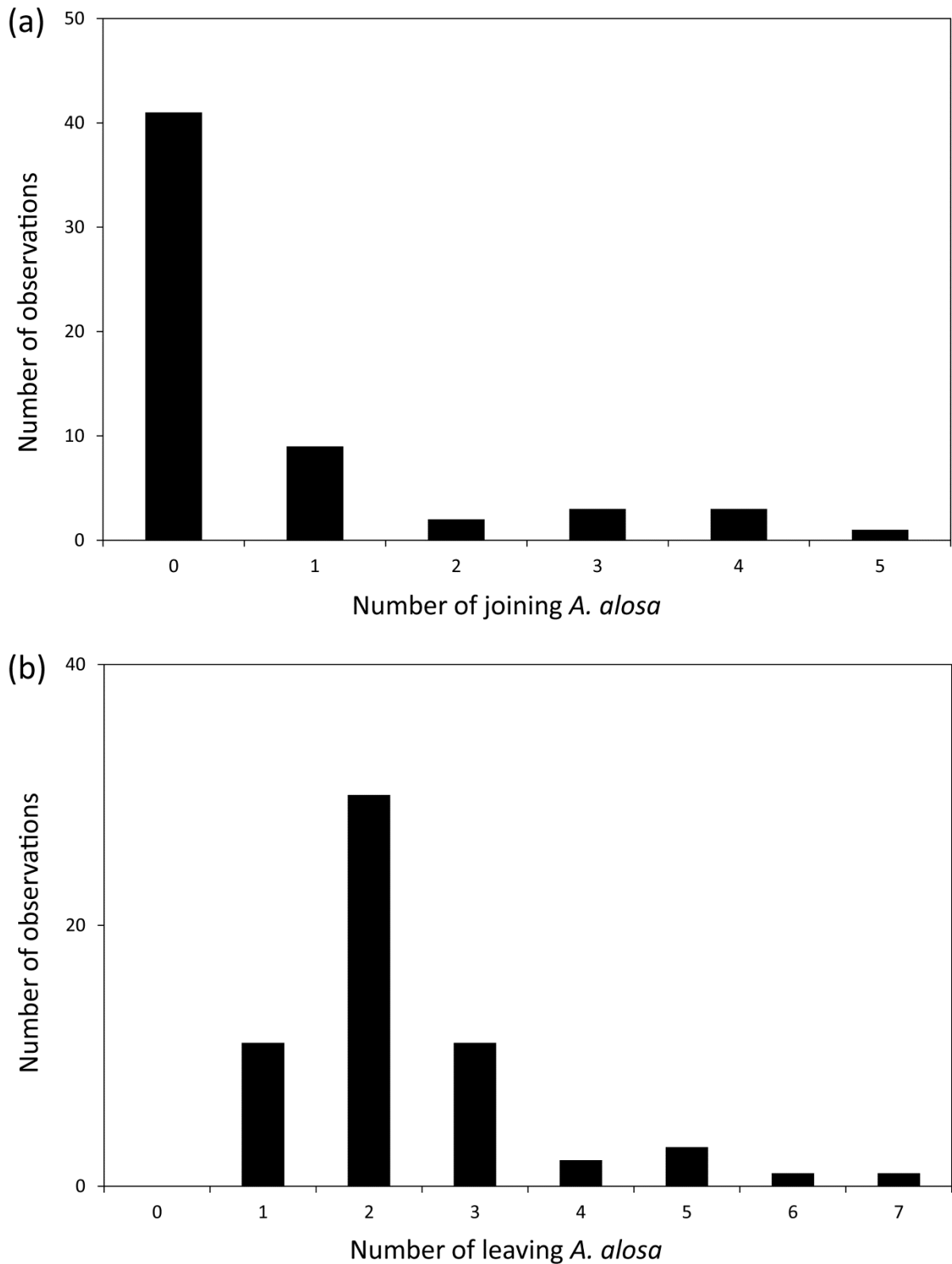


Figure 7. (a) Number of observed *A. alosa* joining a started mating event based on high quality subset of matings (sonar data) ($n=59$). (b) Number of observed *A. alosa* leaving a mating event based on high quality subset of matings (sonar data) ($n=59$).

Discussion

The present study shows the potential of imaging sonar techniques to monitor *A. alosa* spawning activities. With respect to declining populations this is an important point given the urgency of quantifying the extent of returning spawner stocks, spawning success and the need to increase knowledge of spawning behaviour.

During the 3 nights of observation an increasing trend in the spawning activity was seen with both, the sound based index and the hydro acoustic indices. Although the number of mating events recognized in the sonar data was lower due to the smaller area covered, the system was capable of displaying the increasing tendency. The peak spawning time observed between 0130 h and 0200 h fits into the time windows reported in the literature (Cassou-Leins & Cassou-Leins, 1981; Boisneau et al., 1990; D. Fatin & J. Dartiguelongue, (unpublished data) cited in Aprahamian et al., 2003). During the observation period a number of mating attempts was documented. This phenomenon might be explained as part of mating habits by means of a partner check.

In contrast to the sound based spawning activity assessment, the hydro acoustic method revealed additional information about the interaction of individuals involved in one mating event. Although there are indications that there are more than two *A. alosa* involved in a mating event (Boisneau et al., 1990), usually one male and one female are supposed to perform the mating event (Cassou-Leins et al., 2000). F. Cassou-Leins & J.J. Cassou-Leins, unpublished data) assumed that there was only one female involved in one mating event and that each female was only spawning once a night. López et al. (2011) described the participation of several individuals for *Alosa fallax* (Lacepède 1803) assessed by direct visual observation. Boisneau et al. (1990) observed participation of a second couple of *A. alosa* in one mating event in 21.1 % of cases. In only 0.7 %, three or more couples were involved. Furthermore, 78.2 % of the mating events were performed by one couple only. Based on the analysed hydro acoustic data collected in the Garonne River in the present study, the majority of mating events occurred with just a pair of *A. alosa* involved. In a proportion of the pairings (approx. 31 %), however, a higher number of participators was observed. The analysis revealed even and odd numbers of joining *A. alosa*. The missing positional information in the vertical domain of the beam might have contributed to this phenomenon, but the video analysis did not give reason to

assume that the joining *A. alosa* were always pairs. The results presented here support the findings of Boisneau et al. (1990), however, they also give indications that mating events with more than two individuals might not be restricted only to a higher number of couples.

Clupeids are described as group spawners (Mank & Avise, 2006). Alternative reproductive tactics are common (Gross, 1984; Oliveira et al., 2001; Oliveira et al., 2008), and a variety of alternative tactics are known for fishes (Taborsky, 1994). With regards to present knowledge and the results of this study it would be an interesting question to see if the percentage of joining *A. alosa* reflects just a group spawning habit or if this phenomenon could also be related to the existence of parasitic males ('sneakers'). Including the results presented here, there are still only few publications providing quantitative information on the number of participating *A. alosa* and the way individuals interact during the spawn. This is true especially with large rivers and underlines the importance of this study.

In the late evenings, *A. alosa* seemed to recognize and respond to the sound source of the sonar. However, for some reason this was not always the case. Pairs and small groups of *A. alosa* seemed to avoid swimming very close to the emitter by changing their direction away from the DIDSON and crossing the acoustic field further away. Gregory et al. (2007) observed a similar behaviour for *A. fallax* under certain conditions in the River Wye. Wilson et al. (2008) proposed that the response threshold of *A. alosa* may reflect a trade-off between energy expenditure for escapement and other activities. This is in agreement with the observation that the *A. alosa* mostly showed avoidance reactions before the peak spawning time but did not seem to be affected during the mating itself. There are numerous studies giving evidence for responses of alosines to certain ultrasound frequencies. Gregory & Clabburn (2003) revealed a sensitivity of *A. fallax* at a frequency of 200 kHz but showed that *A. alosa* remained unaffected at the high frequency of 420 kHz. Higgs et al. (2004) showed an enhanced sensitivity of American shad *Alosa sapidissima* (Wilson 1811) to a frequency of 90 kHz, which is also in agreement with the findings of Plachta & Popper (2003) and Wilson et al. (2008). However, the main DIDSON operating frequencies at 1.1 and 1.8 MHz are far from the frequency ranges shown to be most crucial for behavioural responses. Nevertheless, further research in the field of sensitivity of alosines to DIDSON emitted frequencies is needed.

Although the method has been shown to be capable of monitoring *A. alosa* spawning activity by using the number of mating events as a direct and the number of clouds as an indirect index, it possesses also some limitations. Compared to the sound assessments the sonar covers a smaller operational range. A permanent fixed installation of the system, without *in-situ* adjustment, appears difficult, especially in large rivers with respect to the high areal fluctuations of spawners around a spawning ground. Further investigations on the longevity and detectability limits of clouds in the river flow are necessary. Under the prevailing conditions the clouds were detectable and persistent over the range of the sonar when followed with the pan-and-tilt unit, indicating a good basis for quantification under adequate signal to noise conditions. Nevertheless, the method is a powerful tool to reveal detailed information about the individual behaviour of anadromous clupeids at the final stage of their migratory journey, even by using comparably simple methods of quantification as has been shown in this study.

In conclusion, the study highlights the monitoring capabilities, but especially the potential of high resolution imaging sonars to assess *A. alosa* spawning behaviour under the challenging conditions prevailing at their spawning grounds at night.

Acknowledgements

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CHAPTER III

Sonar based diel and seasonal fish activity in front of a hydro power plant and chances for flexible fish protection.

Abstract

The assessment and prediction of fish activity and abundance is challenging due to complex multifactorial causal relationships associated with fish migration. The present study demonstrates the potential of horizontally fixed imaging sonars to gain knowledge in the field of spatial ecology of fish with high temporal resolution. An hourly fish abundance raster over the span of a year was chosen as a measure of fish activity to intuitively illustrate alternating diel and seasonal activity patterns at a glance. Three groups were analysed, 'fish' (N = 47604), 'eels' (N = 767) and 'shoals' (N = 526). Diel fish activity altered from an even activity over the 24 hours in spring and early summer to a distinct nocturnal activity in late summer and autumn. Eels showed strict nocturnal activity by exhibiting low abundances in spring and summer and increasing numbers in autumn. Peak migration was measured in December. Fish shoals exhibited a distinct diurnal activity. Shoals were rare in the first half of the year but frequent in autumn, showing a peak in November. As the sonar was mounted in front of the trash rack of a run-of-the-river hydro power plant, activity data of the potential risk group, able to pass through the bar interspaces, was faced the power production data to identify time windows of higher and lower risk of entrainment and respective fish protection requirements. The study underlines the urgent need to use modern fish monitoring tools to expedite flexible fish protective management by meeting ecological and economical demands.

Introduction

On a large scale the distribution of fish in space and time is influenced by their species specific life-cycle and the framing biotic and abiotic conditions. Different life-history-stages are often constrained with habitat shifts (Brönmark et al., 2014, Larinier & Travade, 2002). These shifts can comprise short- or large-scale migrations. While diadromous fish show the largest distances travelled, as some species e.g. catadromous American eel *Anguilla rostrata* (Lesueur, 1817) and European eel *Anguilla Anguilla* (Linnaeus, 1758) migrate up to several thousand kilometers, comprising fresh and saltwater habitats (Aarestrup et al., 2009; McCleave et al., 1987; Righton et al., 2016), potamodromous species move significant distances in the river (Benitez et al., 2015; Benitez & Ovidio, 2017; Lucas & Baras, 2001; Rakowitz et al., 2008). These life cycle dependent movements comprise annual, seasonal or daily cycles to find spawning or feeding grounds, to disperse or shelter (Brönmark et al., 2014; Larinier & Travade, 2002). Life stage based migration in many cases results in a more or less complete presence or absence of a certain age-class in certain parts of the waterbody or system (e.g. salmon smolts leaving the freshwater system). Biotic and abiotic conditions, like predation pressure, availability of food organisms or light and river discharge, affect fish activity, abundance and spatial distribution on a smaller local scale within the current live stage related habitat. Young-of-the-year (YOY) European perch *Perca fluviatilis* (Linnaeus, 1758) perform an ontogenetic habitat shift from the pelagic zone into the littoral zone when shifting diet from zooplankton to benthos (Persson & Greenberg, 1990). The influence of biotic and abiotic factors (feeding opportunities, light intensity and predation risk) on the distribution of YOY burbot *Lota lota* (Linnaeus, 1758) was demonstrated in Lake Constance when larvae were present in the pelagic zone from May until August, undertaking diel vertical migrations by ascending to the surface after sunset and descending back into the hypolimnion after sunrise (Probst & Eckmann, 2009).

Knowledge of the spatial ecology of fish is the key for successful management actions and can on the other hand be a source of error when misunderstood (Cooke et al., 2016). The assessment and prediction of fish activity and abundance is challenging because of the complex multifactorial causal relationships and limited, depending on the applied method (Cooke et al., 2016). A variety of modern assessment tools like telemetry, population genetics, hydroacoustics, otolith

microchemistry and stable isotope analysis exists for the study of spatial ecology but are often not considered and integrated in management plans and actions (Cooke et al., 2016).

While the upstream migration of fish is well examined (Lucas & Baras, 2001) and standard rules for the construction of fish ways have been derived (DWA, 2014), the downstream migration of fish and how to guide them still needs more research (Larinier & Travade, 2002). Most of the studies concerning downstream migration of fish in front of hydro power plants have been carried out on long distance migratory fish species. Knowledge about migration and activity patterns of riverine potamodromous species is comparably lagging behind (Brönmark et al., 2014; Northcote, 1997). Hydro acoustic methods deliver fish abundance data in a high temporal resolution and are therefore a good tool to gather long-term data.

Especially, horizontal-fixed applications are suitable to study diel fish behavior and movement (Lucas & Baras, 2001). Kubecka & Duncan (1998) analysed the diurnal fish behavior and movement between the littoral and the mid-river with horizontal fixed hydro acoustic methods in the River Thames and revealed a higher nocturnal fish biomass in the shallow littoral zone. In the last decade the monitoring of fish movements by means of the horizontal application of modern imaging sonars gained increasing attention (Han & Asada, 2007; MacNamara & McCarthy, 2014; Magowan et al., 2012; Maxwell & Gove, 2007; Pipal et al., 2010). However, long-term studies are still rare.

Today most European rivers are impacted by man-made changes. Fish have to cope with longitudinal and horizontally disconnected rivers, water pollution, massive habitat losses due to bank reinforcements for shipping and the consequences of overfishing (Limburg & Waldman, 2009). A major factor affecting the migration of fishes in rivers, are dams and hydro power plants, disconnecting the river longitudinally. Hence, these constructions have influence on the distribution and abundance of fish, as they can delay or completely prevent fish migration (Drouineau et al., 2017; DWA, 2005; Larinier & Travade, 2002; Nyqvist et al., 2017a; Nyqvist et al., 2017b). However, delay of migration caused by HPPs can depend on site specific constructions and individual fish behavior (Økland et al., 2017). Especially equipped with hydro power plants, dams are a potential source of mortality (Dadswell & Rulifson, 1994; DWA, 2005; Nyquist et al., 2017b; Larinier & Travade, 2002; Martins

et al., 2014). Modern fish protection systems have potential to prevent turbine mortality and to facilitate downstream migration (Økland et al., 2017).

Depending on the bar interspacing trash racks can prevent certain size classes of fish from entering the turbine (DWA, 2005). Others will be able to pass through the gaps of the screen (Larinier & Travade, 2002). The size-class of fish which is able to pass through the screen represents a potential risk-group with a certain mortality rate typical for the site specific turbine (DWA, 2005; Larinier & Travade, 2002).

The present study was aimed on revealing general fish activity patterns by assessing high resolution diel and seasonal fish abundance data in front of a hydro power plant with a multi beam imaging sonar (DIDSON) installed upstream the trash rack for several months within one year. Activity was analysed for fish, eels and fish schools. Data of the size class '< 25 cm', as potentially able to pass the screen and endangered for being entrained into the turbine, was faced to the power production data of the turbine in order to demonstrate the importance and chances to connect knowledge of fish abundance with management actions, meeting ecologic and economic needs.

Material & Methods

Data sampling

The study was conducted in a tributary river of the Rhine River in Germany with a mean discharge of $44.1 \text{ m}^3 \text{ s}^{-1}$. A Dual-Frequency Identification Sonar (DIDSON) was installed upstream of a hydro power plant (HPP) in a distance of approximately 5 m to the trash rack. The bar interspace of the screen measured 20 mm. The facility was equipped with a fish way and eel bypass openings located under the trash rack, which ended up in one pipe facilitating one potential way of eel downstream migration.

The sonar delivers video-like high quality images even under turbid and dark conditions. The field of view was characterized by an opening angle of 29° in the horizontal and 14° in vertical domain and was established by an array of 96 single beams (0.3°) in identification mode at 1.8 MHz or 48 single beams (0.6°) in detection mode at 1.1 MHz. The high frequency delivered a higher resolution. The sonar was installed horizontally (tilt= 0°) on a steel mounting in the middle of the water column in a depth of approx. 2.5 m. The sonar was operated in high frequency mode at 1.8

MHz. The width of the small turbine inlet canal was covered by the sonar with a standard range setting of 5 m and a window start setting of 1.25 m. The frame rate was between 8 to 10 frames/s.

The study was conducted in 2012 in a time window between April and December. Due to technical problems of the HPP, there were recurrent periods with no power production and hence no current in the turbine inlet canal. The water was spilled over the weir. No data was recorded and used from these times. The DIDSON was first deployed on 4th of April and demounted on 14th of December. The Sonar had to be maintained continuously once a week or every two weeks to clean out sediment freights inside the lens compartment and to avoid biofilm growth. A repair of the sonar led to a delayed redeployment on June the 11th after the first repair of the HPP.

Data analysis

DIDSON raw data were converted into maximum intensity echograms and further processed in Sonar 5 Professional (Balk & Lindem, 2010). Echograms plot the received echo intensities over the range against the time. While the corresponding sonar videos show a bird view perspective in a horizontal mounting position of the sonar and need to be processed in real-time, echograms show a larger time period of recorded data at a glance. This enabled a faster screening for fish traces in the dataset. The whole dataset was screened for fish contacts and each was then length measured and classified into one of three categories. Fish contact, eel contact or fish shoal contact. Species identification in DIDSON videos is limited and depending on the sonar setup (Langkau et al., 2012). One exception is the identification of eels, which is intuitive due to their special shape and anguilliform swimming behavior. Fish aggregations were counted as shoals when they were dense with a typical shoal shape and the number of individuals was too high to count and to distinguish from each other. Each contact entering the field of view was counted. It could not be excluded that individuals leaving the sonar beam and reentering the field of view later again were counted again. All contacts were stored in the internal database in Sonar 5 Professional called the 'fishbasket' and exported into Excel (Microsoft) for further processing.

Some hours with massive fish abundance on October the 15th and November the 16th were analysed in subsamples. Fish contacts of three minutes out of half an hour

were counted and number of fish per minute was calculated and extrapolated on 30 minutes. During the peak fish measured between 6-10 cm.

Data for seasonal activity and power generation of the HPP were prepared in Excel (Microsoft) and charts were finally processed and generated in Surfer 14 (Golden Software).

Abiotic complementary data

Fig. 1 gives an overview of the annual river discharge and water temperature. Data was measured approx. 16 km upstream of the study site at the next monitoring station (Source: LANUV).

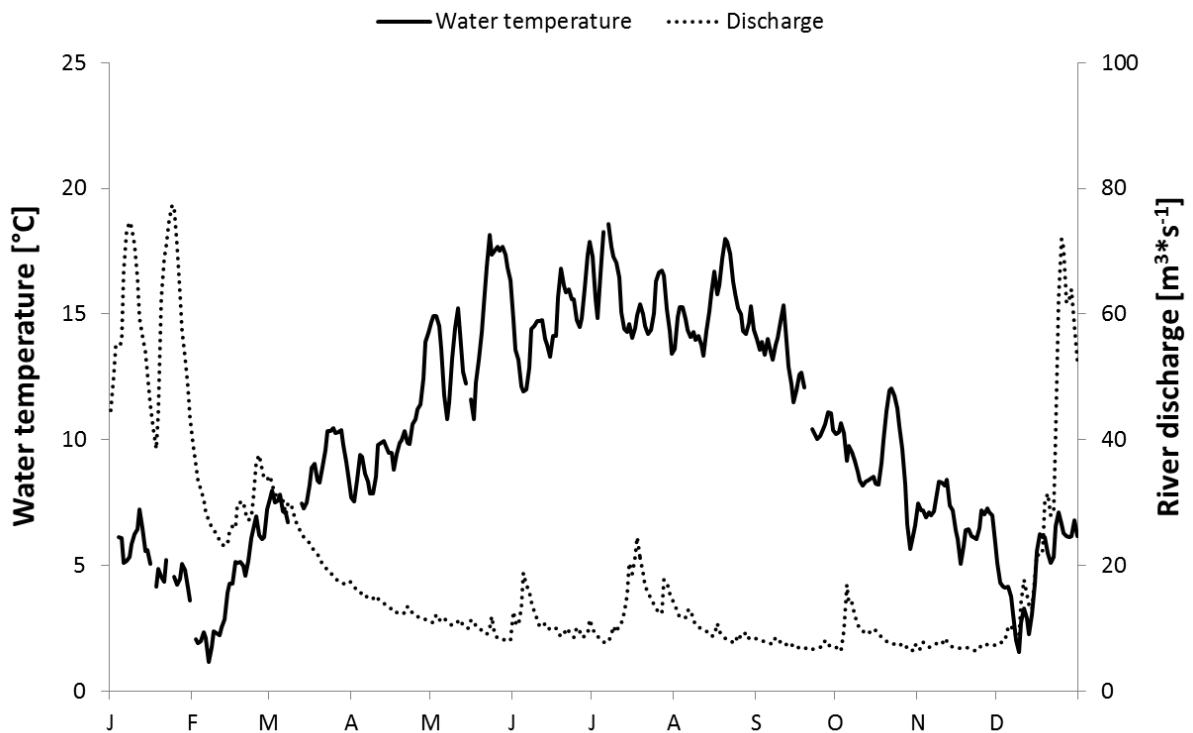


Figure 1. Annual water temperature and river discharge.

Results

Fish activity

Due to recurrent outage of the HPP there were large data gaps over the year. Despite this circumstance trends in a changing fish activity were visible over the seasons. Fig. 2 shows the diel and seasonal activity of fish, eels and fish shoals in front of the HPP screen.

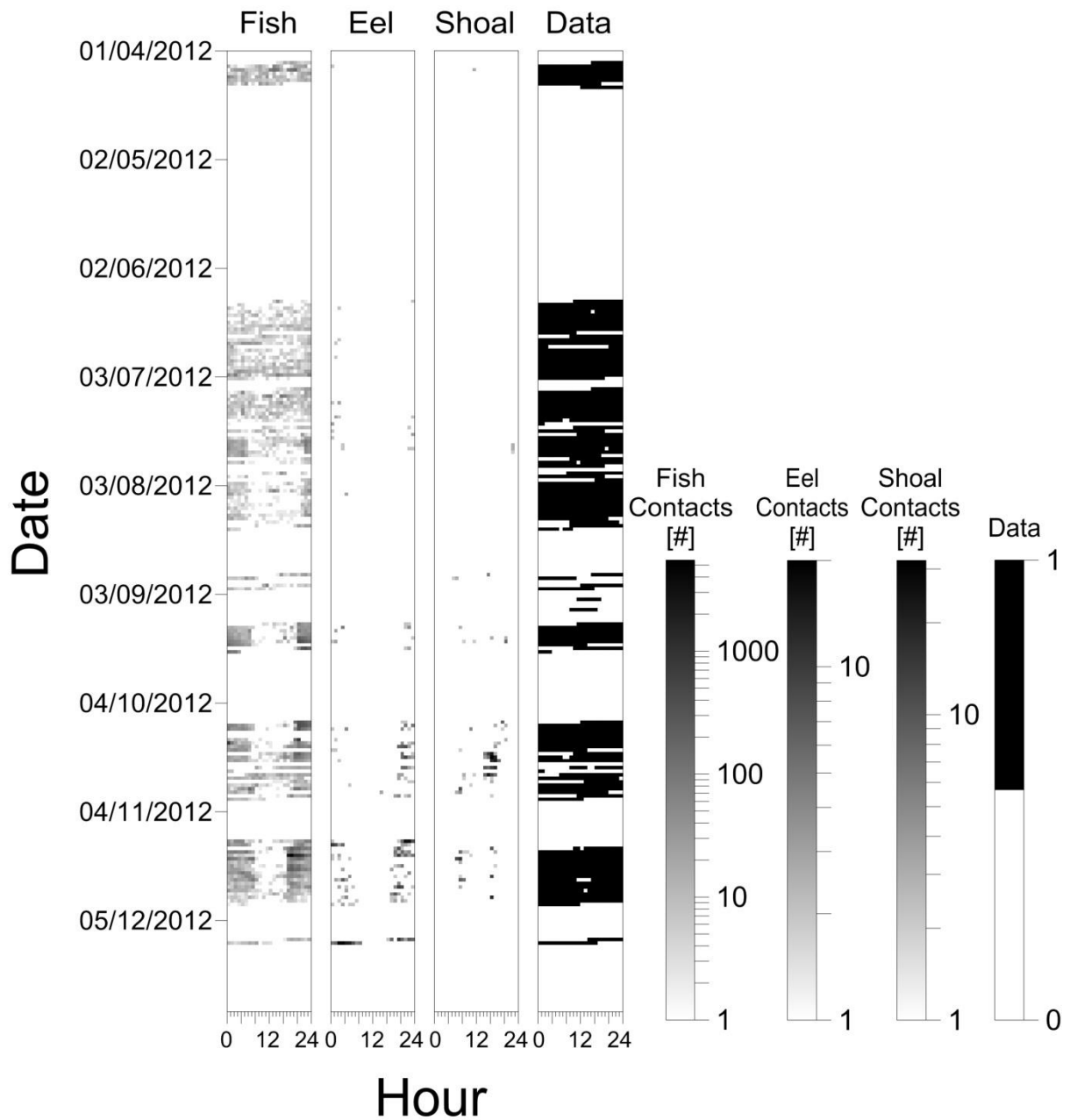


Figure 2. Diel and seasonal activity of fish, eels and fish shoals in front of a HPP screen as measured contacts per hour. Each grayscale bin represents one hour of the year. Color scales for number of fish, eel and shoal contacts are logarithmic. Black color in the right matrix 'data' represents analysed data, white color data gaps.

A total number of 47604 fish contacts were counted. In the first half of the year, approximately until mid of July, the fish activity showed a rather even distribution over the day. Especially in the dataset in April this phenomenon was present with even high fish activity during the daytime. This trend started to alter by the end of July. A decreasing diurnal and increasing nocturnal fish activity with a higher fish presence around dusk especially from September until November could be observed. The high nocturnal fish abundance reflects the borders between light and dark conditions and illustrates the advancing shortening of the daytime in the second half of the year (Fig. 2). The highest mean daily number of fish contacts could be recognized in November (Fig. 3).

A total number of 767 eel contacts were counted. The number of recognized eels increased continuously over the year (with except of August) and peaked in December though just two days were analysed here (Fig. 2 and 3). A strong nocturnal activity was observed here throughout the year. Especially in October and November the activity was higher in the first half of the nights beginning with dusk (Fig. 2). The December peak coincided with a rise in water temperature and discharge after a minimum in water temperature and a period with low discharge (Fig. 1).

A total number of 526 shoal contacts were counted. In the first half of the year the number of recognized shoal contacts was comparably low (Fig. 2). The mean daily number of contacts started slowly to increase in July and peaked in October (Fig. 3). It is striking that in comparison to the single fish detections the shoals could be observed almost exclusively during the daytime and around the transition times between light and dark (Fig. 2).

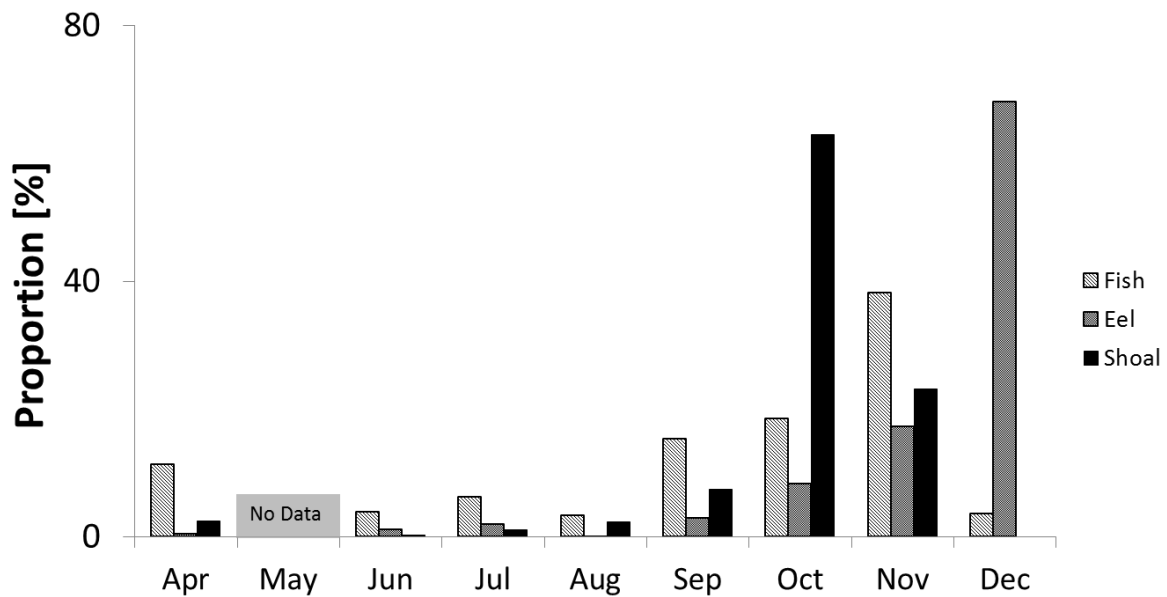


Figure 3. Mean daily number of contacts. Calculated from the monthly total number of contacts (per category) divided by the number of days with analysed data. This mean number of contacts per day was then calculated as percentage of total count per category.

Fish < 25 cm and HPP power production

Fig. 4 shows the proportions of fish contacts < 25 cm over the year. This group defines the category of individuals which are potentially able to pass through the screen of the HPP. The percentage of this group was the lowest in April with 23 % and increased over the following month to establish on a level with values over 90% from August until November. The percentage was lower again in December with a value of 38 %.

Fig. 5 shows the number of measured fish contacts < 25 cm per hour in relation to the hourly power production. The highest power production was observed in the first half of the year, showing the highest values in April when the proportion of fish contacts < 25 cm reached the lowest measured value of the year (Fig. 4). The fish activity showed an evenly distributed pattern with no special preference of day time. Power production in June and July was lower compared to April, but still on medium level. The group of fish < 25 cm was more than doubled and apportioned 61 and 69% respectively. The fish activity pattern still showed evenly distributed contacts over the

24 hours until mid of July when the diel activity pattern changed and fish activity tended to shift into the dark phase.

This pattern progressively established within the next month until November and the fish activity pattern reflected the increasing shortening of the daytime. Whether this pattern was still present in December could not be shown due to the lack of data in this month. The power production further decreased after August, with the lowest production in September and then ranged on lower level throughout the rest of the year. The proportion of fish < 25 cm was over 90% from August until November and decreased in December.

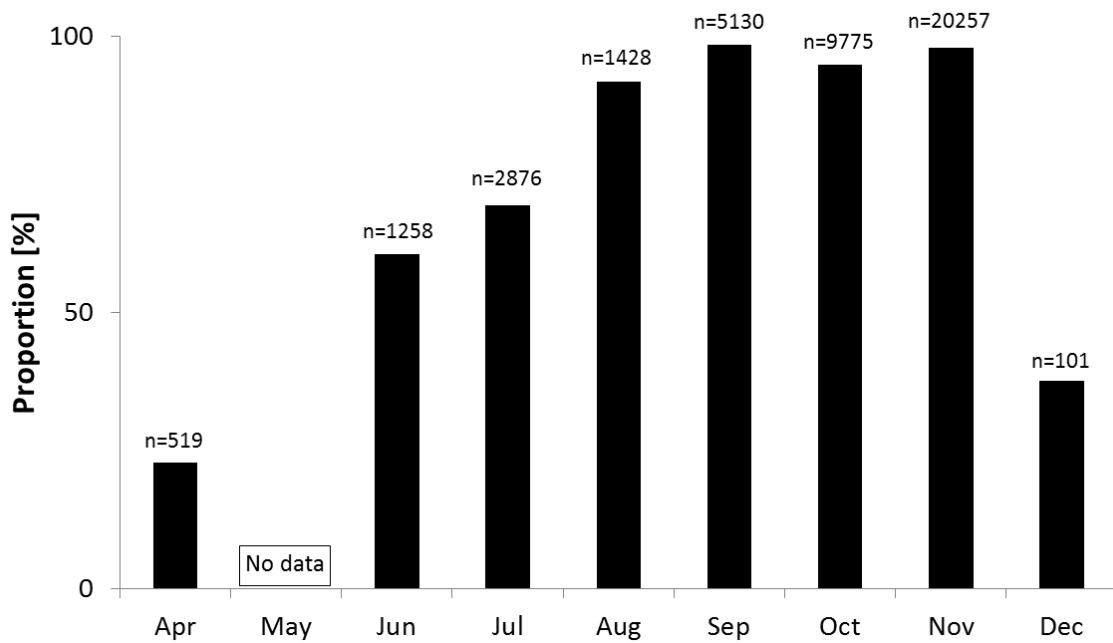


Figure 4. Percentage of fish contacts < 25 cm per month.

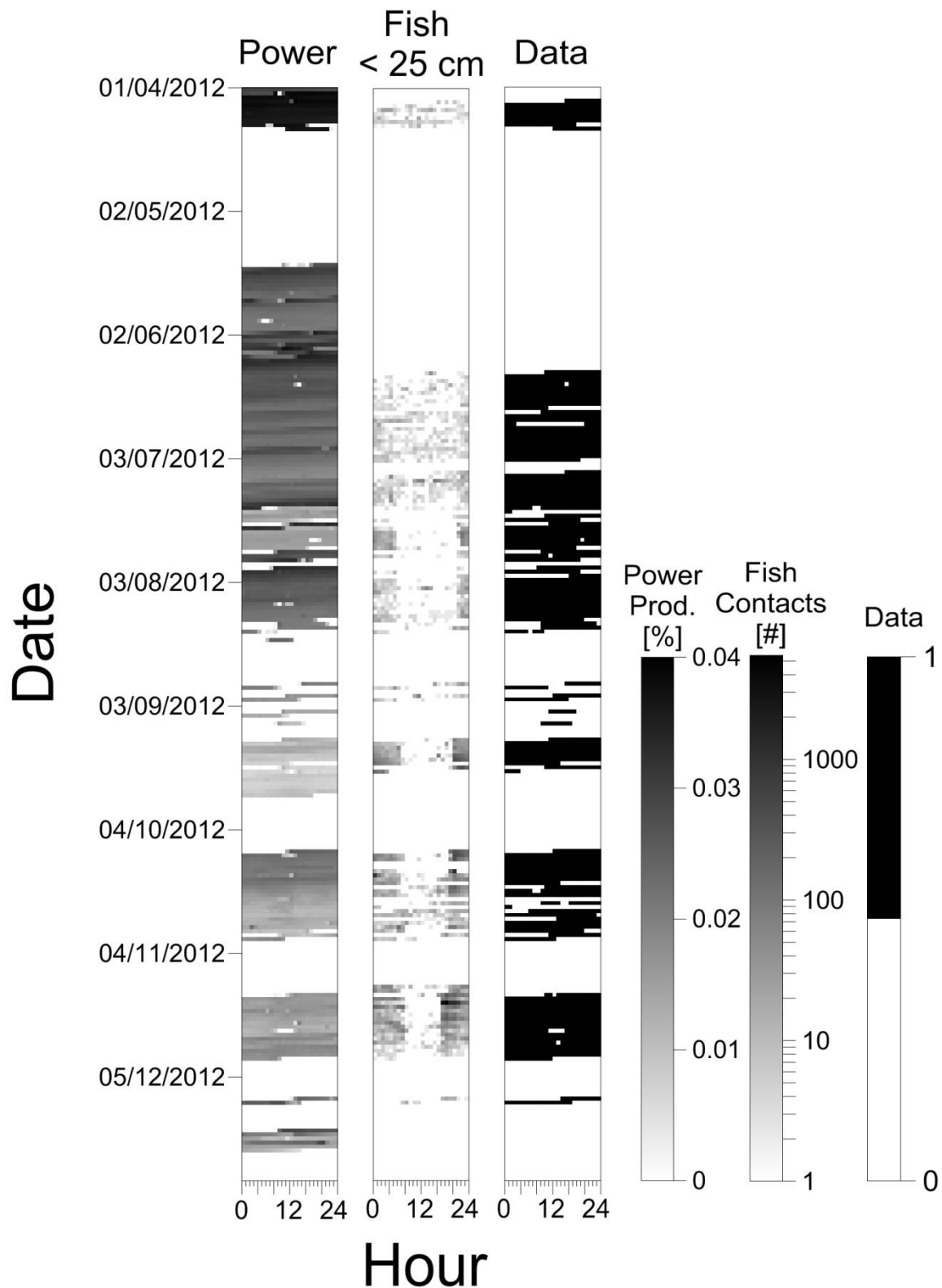


Figure 5. Diel and seasonal activity of fish < 25 cm and power production of the HHP as number of fish contacts per hour and percentage from annual total power production respectively. Each grayscale bin represents one hour of the year. Color scale for number of fish contacts is logarithmic. Black color in the right matrix 'data' represents analysed data, white color data gaps respective the fish contact data.

Discussion

The present study demonstrates the potential of horizontally fixed imaging sonars to gain knowledge in the field of spatial ecology of fish with high temporal resolution. An hourly fish abundance raster over the span of a year was chosen as a measure of fish activity to intuitively illustrate alternating fish activity patterns at a glance.

Despite of larger data gaps, the study revealed diel, as well as seasonal activity patterns of three defined groups 'fish', 'fish shoals' and 'eels'. Fish showed a shift in activity from a more evenly distribution over the 24 hours in spring, to an increased activity in the transition times and the dark phase in the second half of the year (Fig. 2). The mean daily number of fish contacts was higher in fall compared to spring and summer (Fig. 3), while the proportion of fish < 25 cm rose continuously on values over 90 % since April (Fig. 4). December was not representative for 'fish' and 'shoal' because of only two days of data. However, it is of value for the activity of eels as the highest numbers of contacts were measured here (Fig. 3). It is likely that the risen proportions of fish < 25 cm and the higher mean number of fish contacts in fall included a large number of 0+ fish from the recent cyprinid and percid spawning period. It might therefore be possible that the distinct diel activity cycle was mainly dominated by small fish.

Fish shoals were rare in spring and summer, but became numerous in fall when the peak was measured in October (Fig. 2 and 3). The observed distinct diel pattern was contrary to that of single fish and eel, given that shoals were predominantly measured by daytime and in dusk and dawn (Fig. 2). The increased number of fish and proportion of smaller fish in combination with the frequent formation of shoals in fall could reflect a beginning migration to winter habitats and predator prevention tactics respectively. Brönmark et al. (2008) and Skov et al. (2008) found massive migrations of cyprinids, dominated by roach *Rutilus rutilus* (Linnaeus, 1758), from the lake into the streams occurring in fall. The migration was driven by a trade-off between predator avoidance and growth. Although in our study area there were no connected lakes, temporarily and continuously connected floodplain waterbodies and oxbows were present, which are likely to be used as a habitat during the growth phase. Pavlov & Mikheev (2017) suggest intraspecific interactions, such as shoaling, to be promoted by spatial and temporal peaks of migration.

Considering that acoustic species identification is still one of the most challenging hydro acoustic topics (Horne, 2000), limited by specific sonar technique and setups

(Langkau et al., 2012) and up to date still a field that has been identified to have a high demand for further research (Pollom & Rose, 2016), the setup in the present study allowed the identification of eels, due to their specific intuitively recognizable anguilliform body shape and swimming behavior.

Eels showed a distinct diel activity pattern with a preference of the nocturnal hours throughout the year (Fig. 2). The diel activity pattern was similar to the pattern found by Stein et al. (2016), though they also observed seasonal activity in spring. During this study mean daily numbers were low in spring and summer, but increased in fall with a migration peak in December (Fig. 3). This reflects the starting autumnal downstream migration of silver eels and is in accordance with autumn migratory windows observed in other studies (Haro, 2003; Lowe, 1952; Sandlund et al., 2017; Stein et al., 2016). It is important to mention that only two days of data were existent in December. However they reflect the highest measured daily mean numbers of eel contacts within the study, suggesting that data were likely to be acquired during a main eel migration peak. The December maximum coincided with a rise in water temperature and discharge after a minimum in water temperature and a period with low discharge (Fig. 1). Many environmental factors, such as water discharge, water temperature, photoperiod, turbidity, light intensity, moon phase, atmospheric pressure or precipitation are discussed in the literature to potentially influence eel migration behavior in terms of timing, intensity or speed. Hence, the regulation of silver eel migration is complex. Different studies identified several of these factors to significantly influence eel migration. While increased water discharge played a role in various studies (Barry et al., 2015; Lowe, 1952; Vøllestad et al., 1986; Vøllestad et al., 1994; Feunteun et al., 2000), the other environmental factors were shown to have influence under certain conditions in different locations as well (Cullen & McCarthy, 2003; Stein et al., 2016; Vøllestad et al., 1994). However, the presence of HPPs, takes effect as fish migration obstacles, can cause migration delay (Drouineau et al., 2017; Nyqvist et al., 2017a; Nyqvist et al., 2017b) as well as mortality by turbines (Larinier & Travade, 2002).

As 96 % ($n = 101$) of the eels during the peak in December were larger than 60 cm (mean length 78 ± 10 cm) (data not shown) the 20 mm trash rack should have prevented turbine entrainment of this group of individuals, however impingement effects remain unknown. At the study site an existent eel bypass pipe, temporal spill over the weir and a fish path offered three potential migration routes. Økland et al.

(2017) found that modern fish protection systems in combination with capable downstream pathways can facilitate silver-eel migration with low mortality.

The group of smaller fish (< 25 cm) that was potentially at risk to pass the trash rack and become endangered by turbine entrainment, showed a very similar diel and seasonal activity pattern like the combined 'fish' group (Fig. 2 and 5). This underlines the dominance of this length class in terms of number and observed behavior.

Seasonal consideration

Comparing the monthly percentage of fish < 25 cm with the seasonal power production of the HPP at a glance (Fig. 4 and 5), the power production in spring and summer was high, but the proportion of fish < 25 cm was lower (especially with the lowest values in April) compared to the late summer and fall. The lower proportions of this group may be due to the absence of young-of-the-year fish by three potential causes. First, especially valid for April, the spawning season was not yet finished and therefore only few 0+ fish of early spawning species present. Second, regarding June and July, when proportions already increased, larger numbers of fish inhabited growth habitats e.g. oxbows and connected floodplain waterbodies. Third, very small 0+ individuals (fish larvae) were present, but not detectable with the sonar under the prevailing acoustic conditions. Based on these data, regarding the monthly low mean numbers of fish (Fig. 3) and the low proportion of the risk group (Fig. 4), spring and early summer appeared to represent a time period with a lower entrainment risk (Fig. 5). However, the large data gaps due to technical problems with the HPP and the sonar between April and June lack valuable further information.

The power production in August was still higher than in the subsequent month and although the mean daily number of fish was low, the proportion of the risk group was already high (Fig. 3, 4 and 5), indicating an unfavorable combination.

In September the mean daily number of fish increased continuously and peaked in November (Fig. 3). In the same period the proportions of the risk group ranged between 95-98 % (Fig. 4) indicating the highest probabilities to potentially pass the trash rack. However, the power production in fall was lower than in spring time (Fig. 5). On a seasonal scale, the necessity of fish protective management actions at the site was highest in fall.

Diel consideration

The diel patterns of the risk group showed a more even activity throughout the 24 hours in spring. By the mid of July this behavior altered and a progressive establishment of nocturnal activity could be observed (Fig. 5) indicating a higher entrainment risk in the dark phase. This was especially true in October and November, when the highest number of fish were present in combination with risk group proportions of over 90% in front of the trash rack due to a potential migration peak (Fig. 3). On a diel perspective the necessity of fish protective management actions changed within the year. While during times of even diel fish activity in spring and early summer, no preferred time windows for management actions could be identified, the nocturnal fish activity in fall implied the need for fish protections by night.

Conclusions and perspectives

The present study demonstrated the potential of high resolution imaging sonars to gain knowledge in the field of the spatial ecology of fish by monitoring abundance as a measure of activity. The high image resolution allowed the identification of eels and fish shoals, which could be used to reveal diel and seasonal migration behavior. It was shown that DIDSON has the potential to be applied as a long-term fish monitoring tool, though labour for manual analysis remained immense. The ability to measure fish length allowed the analysis of a certain size class, potentially at risk of entrainment, and to derive diel and seasonal critical time windows. In contrast to classical assessment methods like gill nets or electro fishing, which deliver temporal snapshots, imaging sonars are non-invasive and provide data with high temporal resolution. Because species discrimination is still limited, the combination with other assessment methods is an option, depending on the task.

Against the background of climate change and the increased promotion of renewable energy sources, the power generation by HPPs is topical. Under consideration of ecologic and economic needs it is urgent to identify time periods which are favorable for fish protective actions by minimizing power losses. The annual activity charts give a good basis to derive rough time windows, but a precise flexible management should be based on a model fed with fish activity and power production data derived by long-term studies over several years. However such a model would still be based

on migration patterns of the past and did not consider to its extents the complexity of fish migration processes. A real-time analysis of fish abundance is necessary and could be implemented to allow in-situ management actions. Recent studies on the sonar based automation of fish counting and sizing (Han et al., 2009) and the semi-automated identification of eels (Mueller et al., 2008) have been conducted. Further research in this field is necessary to reduce the financial and temporal bottleneck of manual analysis labour and to expedite the development of real-time sonar systems capable to facilitate additional flexible fish protective management actions.

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Conclusions

In the first chapter, we could demonstrate that DIDSON can be used to discriminate fish species with high correct classification rates by extracting specific descriptors from sonar derived template- and live fish shadows, as they are strongly related to the outer shapes. Although, still delivering high correct classification, template experiments indicated detail losses with declining fish lengths at a size threshold of 20 cm. Against the background that acoustic species identification is one of the most challenging hydro acoustic topics (Horne, 2000) and up to date still a field that has been identified to have a high demand for further research (Pollom & Rose, 2016), Chapter I contributes an important part of research in this field by suggesting to consider novel sonar setups in future monitoring concepts e.g. at fish ways or other kind of suitable bottlenecks. However it has to be considered that applications in the field are constrained with potentially unfavourable acoustic conditions, multi species environments and challenges arising by merged shadows of schooling fish. Hence, further research is needed to develop capable image processing algorithms, intelligent discrimination methods and automation processes allowing *in-situ* processing of sonar data.

In Chapter II we could show the capabilities of imaging sonars to observe fish behavior under challenging conditions and to assess spawning activity of endangered Allis shad (*Alosa alosa*) even on long ranges. Spawning related clouds of sexual products and micro bubbles could be detected with the sonar and be used as a measure of spawning activity, as they were consistent with sounds of mating events (bulls) assessed parallel in hearing surveys. New behavioural aspects could be revealed by observing that matings were not restricted to one pair of spawners only, which was a common opinion in the literature (Cassou-Leins et al., 2000; Cassou-Leins & Cassou-Leins, 1981). The recent further declines in the local populations of the Garonne River, with dramatically reduced spawning activities (verbal information of EU-Life project manager) already impeding the assessment of important knowledge, underline the importance of this study.

Chapter III highlights the potential of imaging sonars to serve as long-term monitoring tools to assess diel and seasonal activity patterns of fish with high temporal resolution and to point out chances for improved fish protection in front of hydro

power plants. Distinct alternating patterns for three groups ('fish', 'eels' and 'shoals') could be revealed by intuitively illustrating hourly fish abundance over the span of one year at a glance. Knowledge of the spatial ecology of fish is the key for successful management actions and can on the other hand be a source of error when misunderstood (Cooke et al., 2016). Hence, the study underlines the importance of assessing and integrating fish activity data into modern flexible fish protection systems. Data of small fish, being at risk of entrainment, was faced power production data to derive time windows with high or low risk and constrained fish protective requirements. As a future research task, a model, based on long-term data over several years, should be calculated to improve predictions of entrainment risk. However, as already mentioned in Chapter I, the development of automated fish counting and measuring algorithms is needed to allow the initiation of flexible management actions meeting ecological and economical demands.

The three chapters demonstrate potential and limitations of high resolution imaging sonars in applied freshwater ecology. DIDSON was shown to facilitate new opportunities for the analysis of challenging tasks in the fields of fish species identification, fish behavior and fish activity monitoring in a non-invasive manner.

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Supplementary Material - Chapter III

Length frequency distribution

Length frequency distribution of all measured fish and eel contacts altered throughout the year. Mean length in April was 32 ± 12 cm and length distribution was unimodal (Fig. 6). On an annual perspective the mean length showed a decreasing trend until December when the main peak of eel migration was observed (Fig. 2, 3 and 6). In June there was a bimodal distribution with peaks in the length classes >10-20 cm and >50-60 cm. The unimodal distribution progressively altered into a right-skewed distribution over the year until December. The smallest size class >0-10 cm represented the largest proportion from July until November (Fig. 6).

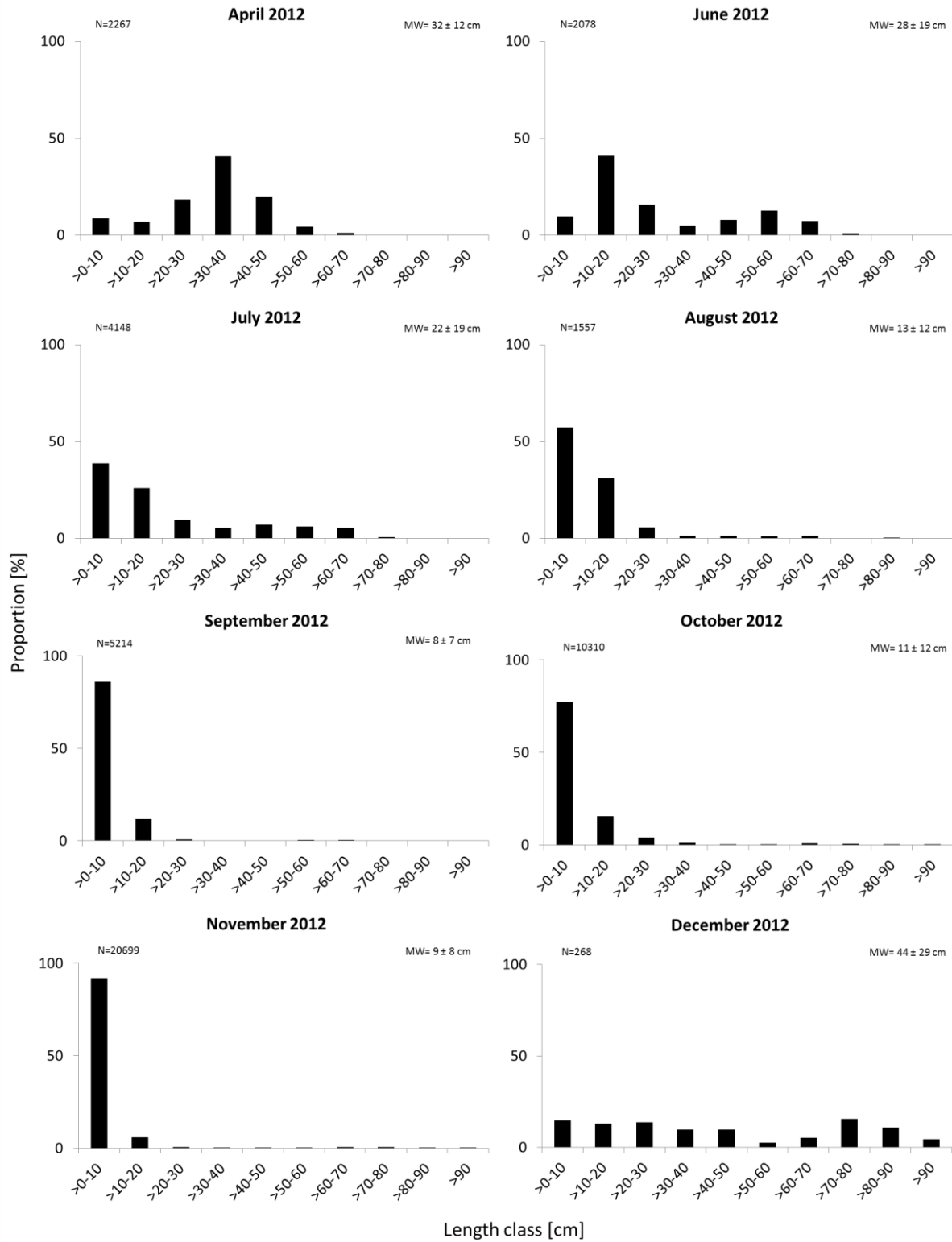


Figure 6. Monthly relative length frequency distribution of all measured fish and eel contacts. Note: No data for May available.

KURZZUSAMMENFASSUNG

Angesichts ihrer seit jeher großen Anziehungskraft auf den Menschen, erleben aquatische Ökosysteme, insbesondere seit Beginn der Industrialisierung, eine zunehmende Beeinflussung durch anthropogene Veränderungen. Flüsse wurden zum Zwecke der Schiffbarkeit begradigt, mit Querbauwerken zur Energiegewinnung aufgestaut und zunehmend durch Abwässer chemisch und organisch verschmutzt. Ein drastischer Verlust von Habitatdiversität und der Verlust der longitudinalen und horizontalen Durchgängigkeit und Vernetzung sind die Folgen. Überfischung der Bestände und wachsende freizeitliche Nutzung führten darüber hinaus zu stark schrumpfenden Beständen und zum Aussterben der meisten europäischen diadromen Fischarten. Artenschutz- und Wiederansiedlungsprojekte helfen die bedrohten Arten und diverse Fischartengemeinschaften zu erhalten. Essentiell für das erfolgreiche Management und die Umsetzung geeigneter Maßnahmen ist ein diffundiertes Wissen über Populationsentwicklungen, Verhalten und ökologische Ansprüche der Arten in allen Phasen ihrer Lebenszyklen. Hierfür sind geeignete Monitoring- und Erhebungsmethoden grundlegend. Angesichts zunehmend fragiler werdender Bestände einzelner Arten sind nicht-invasive Methoden von großer Bedeutung, da sie die bestehenden Populationen nicht in Mitleidenschaft ziehen und die Entnahme und Beeinträchtigung von Individuen z.B. für Besanderungen vermeiden. Hydroakustische Systeme, insbesondere Imaging-Sonare (DIDSON), bieten eine solche Methode durch den gezielten Einsatz von Schall zur Echtzeit-Visualisierung der Geschehnisse Unterwasser. Das DIDSON liefert auch in trübem Wasser und bei Dunkelheit Bilder und die Möglichkeit Fische zu zählen, zu vermessen und ihr Verhalten zu beobachten.

In einer speziellen Anwendung des Sonars konnten Fischarten anhand ihrer charakteristischen akustischen Schatten im Sonarbild, basierend auf der Anwendung von „Normalised Elliptic Fourier Descriptors“ und einer Diskriminanzanalyse voneinander unterschieden werden. Versuche mit Fischschablonen vier verschiedener Fischarten (Brassen, *Abramis brama* (L.), Barbe, *Barbus barbus* (L.), Döbel *Leuciscus cephalus* (L.) und Bachforelle *Salmo trutta* (L.)) in vier verschiedenen Größen (20 cm, 30 cm, 40 cm, 50 cm), zeigten eine Größenschwelle von 20 cm an, ab der die Zuverlässigkeit der Artentrennung abnahm. Auch Mesokosmosversuche mit lebendigen Fischen der vier Arten führten zu Ergebnissen

mit einer hohen korrekten Unterscheidungsrate. Standbilder ihrer akustischen Schatten dienten hier als Grundlage.

An einem Maifischlaichplatz (*Alosa Alosa* (L.)) in der Garonne in Frankreich konnten mittels DIDSON Wolken aus Geschlechtsprodukten und Mikroluftbläschen detektiert werden, die aus dem charakteristischen Laichverhalten der Maifische resultieren. Die Fische schwimmen während des Laichaktes in großer Geschwindigkeit eng aneinandergeschmiegt im Kreis. Dabei durchbrechen sie die Wasseroberfläche und erzeugen charakteristische Geräusche und geben die Geschlechtsprodukte in die Wassersäule ab. Der nächtliche Peak der Laichaktivität konnte, basierend auf den Sonaraufnahmen, ermittelt werden. Die Anzahl der Wolken diente als Indikator für die Laichaktivitäten und war im Trend übereinstimmend mit den Auswertungen der gehörten Laichakte, die traditionell in Frankreich als Maß für die Laichaktivität dienen.

Es konnten auch neue Erkenntnisse zum Laichverhalten der Maifische gewonnen werden. So konnte gezeigt werden, dass die Laichakte nicht auf jeweils ein Pärchen beschränkt sind. In etwa einem Drittel der Fälle konnte beobachtet werden, dass drei oder mehr Fische aus einem Laichakt hervorgingen.

In einem Zufluss des Rheins wurde das Sonar oberhalb des Rechens einer Wasserkraftanlage über einen Zeitraum von April bis Dezember installiert, um Veränderungen tageszeitlicher und saisonaler Fischaktivitätsmuster, basierend auf stündlicher Abundanz dreier definierter Gruppen („Fisch“, „Aal“ und „Schwarm“), intuitiv erfassbar in Aktivitätsdiagrammen, darzustellen. Aufgrund der Diagramme konnten klare Muster und Migrationspeaks dokumentiert werden. Anhand einer Gegenüberstellung der Aktivität der Risikogruppe kleinerer Fische, die potentiell gefährdet sind den Rechen zu durchdringen und der Energieproduktionsdaten der Anlage, konnten Zeiträume mit höherer oder niedrigerer potentieller Gefahr, sowie die einhergehende Notwendigkeit von Fischschutzmaßnahmen abgeleitet werden.

ERKLÄRUNG

Münster, den 04.05.2018

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Curriculum vitae

Persönliche Angaben

Name	Dipl. Biol. Manuel Langkau
Geburtsdatum/-ort	03.07.1980 in Essen
Anschrift	Von-Esmarch-Str. 10, 48149 Münster
Email	langkau@lfv-westfalen.de
Eltern	Diemut Langkau geb. Vatter und Wolfgang Langkau
Familienstand	ledig
Staatsangehörigkeit	deutsch

Ausbildung

1987-1991	Fischlaker Schule Essen
1991-2000	Goethe-Gymnasium Essen
Jun 2000	Prüfung zur allgemeinen Hochschulreife (Gesamtnote 2,3)
2002-2008	Diplomstudiengang im Fachbereich Biologie
2002-2004	Grundstudium im Fachbereich Biologie an der Heinrich-Heine Universität, Düsseldorf
2004-2008	Hauptstudium im Fachbereich Biologie an der Universität zu Köln Hauptfach: Zoologie – Aquatische Ökologie Nebenfächer: Entwicklungsbiologie, Biochemie
Mai-Jul 2006	Anstellung als studentische Hilfskraft am Zoologischen Institut der Universität zu Köln: Analyse der Eizahlen von <i>Corbicula fluminea</i> auf dem „Rheinlabor“.
Mrz 2008	Abschluss der Diplomprüfung (Gesamtnote 1,0) Thema der Diplomarbeit: Piscivore versus planktivore: How does morphological change influence ecological performance? An intraspecific survey.
Seit Jan 2009	Promotionsstelle beim Landesfischereiverband Westfalen und Lippe e.V. und der Ökologischen Außenstelle der Universität zu Köln zum Thema: Echoes in motion:

An acoustic camera (DIDSON) as a monitoring tool in applied freshwater ecology. Finanziert durch das Ministerium für Klimaschutz, Umwelt, Landwirtschaft, Natur- und Verbraucherschutz des Landes Nordrheinwestfalen aus Mitteln der Fischereiabgabe.
Seit 2012 Wissenschaftlicher Mitarbeiter in der LFV Hydroakustik GmbH

Zusätzliche Arbeitserfahrung

2000-2001 Zivildienst im Altenheim St. Ludgeri, Essen-Werden
2002 Praktikum im Aquazoo – Löbbecke Museum, Düsseldorf
2002 Praktikum in der Kleintierpraxis Kai-Sven Herrmann, Essen
2005, 2006, 2007 Hilfstätigkeiten in der Firma LimnoPlan: Elektrobefischungen und Fischhandling bei Aalmarkierungen
2006 Volontär im Projekt zum Schutz des Afrikanischen Chamäleons (Management : Hellenic Ornithological Society)

Münster, den 04.05.2018

Manuel Langkau