

# The effects of pulse stimulation on biota – Research in relation to ICES advice – Effects on dogfish.

D. de Haan, B. van Marlen, I. Velzenboer, J. van der Heul,  
Hans van der Vis

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# Wageningen *IMARES*

Location IJmuiden

Client: Ministerie van LNV  
Directie AKV  
P.O. Box 20401  
2500 EK Den Haag

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## Assignment

The work has been commissioned by the authority Directorate of Fisheries of the Ministry of Agriculture, Nature and Food Quality of the Netherlands and conducted in cooperation with the private company Verburg-Holland Ltd. of Colijnsplaat, the Netherlands.

## Confidentiality

Details of the pulse trawl system developed by Verburg-Holland Ltd. and in particular the characteristics of the stimulus are kept confidential and therefore not revealed in this report.

## Quality Assurance

IMARES utilises an ISO 9001:2000 certified quality management system (certificate number: 08602-2004-AQ-ROT-RvA). This certificate is valid until 15 December 2009. The organisation has been certified since 27 February 2001. The certification was issued by DNV Certification B.V. The last certification inspection was held the 16-22 of May 2007. Furthermore, the chemical laboratory of the Environmental Division has NEN-AND-ISO/IEC 17025:2000 accreditation for test laboratories with number L097. This accreditation is valid until 27 March 2009 and was first issued on 27 March 1997. Accreditation was granted by the Council for Accreditation, with the last inspection being held on the 12<sup>th</sup> of June 2007.

It is foreseen to involve members of the Expert Group on Pulse Trawling, who worked on the ICES Advice, to give guidance on the proper methodology applied. In addition publishing articles in peer-reviewed magazines provides an opportunity for international critique and quality testing.

# Summary

In response to questions asked by ICES on the effects of pulse stimulation in commercial beam trawling on components of the marine ecosystem a number of preliminary studies were undertaken in the period between 1 November 2008 and 01 June 2009.

The study that we report on here involved the exposure of lesser spotted dogfish to a simulated electric pulse under laboratory conditions, and monitoring of mortality, injuries and behavioural responses, in particular feeding response. The electric pulse simulator was made available by Verburg-Holland Ltd. with pulse characteristics similar to the commercial Verburg pulse system.

On 4 December 2009, three groups of 16 fishes with similar lengths (0.3 – 0.65 m) were exposed to the electric stimulus, with each group in one of three distance ranges:

1. A “far field” range with the fish exposed at 0.4 m side ways of a conductor element.
2. A “above field” range with the fish exposed at 0.1-0.3 m above the center of a conductor pair;
3. A “near field” range with the fish exposed at 0.1 m from the conductor element;

Furthermore, in order to be able to monitor the effects of transfer and other unknown influences a control group of 16 fish was confined in the same way, but not exposed to the electric stimulus. Each fish was exposed four times in a row. All fish were examined for injuries directly after the end of the last stimulus. Feeding response was monitored for 14 days after. Other behavioural responses (in particular contractions, swimming patterns) were monitored during stimulus and in the 14 days period following stimulation. Finally, fish were kept in husbandry for another 9 months. Additionally long term mortality and other behaviour such as egg production were monitored.

No evidence was found of differences in feeding response or likelihood of injury or death between the exposure groups. There was no evidence that fish sustained injuries as a result of the exposures. Respectively 8 and 9 months after the experiment a single specimen of the “above field” category and “near field” category died. In the 14 days observation period after the exposures no aberrant feeding behaviour could be distinguished. Fish in all tested groups started feeding normally the same day directly after the exposures. In a period of 7 months after the exposures all exposed groups produced eggs in numbers varying between 5-39 per group. Surprisingly the control group did not produce eggs.

Regarding the other behavioural responses (mainly reflexes and muscle contractions, and post-reactions, such as a rapid body reverse, short-curved body rotations and acceleration towards the water surface), there were some clear differences between exposure groups. The responses of the fish exposed in the “far field” range, representing the fish just aside the fished area of the trawl, were minor and ignorable. However, the responses of the fish exposed in the “above field” range were more pronounced with contractions, rapid body reverses, short-curved body rotations and acceleration towards the water surface occurring. The responses of the fish exposed in the shortest possible range, the “near field” range, were the strongest with increased incidence of contractions and rapid body reverses, short-curved body rotations and acceleration towards the water surface. Although this experiment has not been set up, or designed, to investigate differences between exposure groups in terms of behavioural responses other than feeding responses, we note that a common behavioural response in the “near field” group was to ‘accelerate upward’. Since, in field situations this behaviour has been observed to lead to dogfish becoming entangled in the meshes of the top panel of the full-scale trawl this merits further investigation.

# 1. Introduction

## 1.1. Background and research carried out so far

In response to ecosystem related concerns about bottom trawling and particularly beam trawling that were raised by various scientists in the last decades of the previous century (Anon., 1988, 1995; Jennings and Kaiser, 1998; Lindeboom and De Groot, 1998; Kaiser and De Groot, 2000; Paschen *et al.*, 2000; Fonteyne and Polet; 2002; Piet *et al.*, 2000) pulse stimulation was developed as an alternative to tickler chains to enable the catch of flat fish, in particular sole (*Solea vulgaris* L.) and plaice (*Pleuronectes platessa* L.). Many studies were done in the 1970s and 1980s, but in spite of promising results commercial uptake was lacking (De Groot and Boonstra, 1970, 1974; Vanden Broucke, 1973; Stewart, 1975, 1978; Horn, 1976; Horton, 1984; Agricola, 1985; Van Marlen *et al.*, 1997). The development of pulse trawling was again taken up in the 1990s by a private company (Verburg-Holland Ltd.) in The Netherlands (Van Marlen, *et al.*, 1999; 2000; 2001a, 2001b). This led to trials over a complete year on a commercial vessel fully equipped with the new technology (Van Marlen, *et al.*, 2000, 2005a, b, 2006).

Meanwhile, questions about ecosystem effects of introducing pulse beam trawling in the Dutch flatfish fishery were raised by the European Scientific, Technical and Economic Committee for Fisheries (STECF) and the International Council for the Exploration of the Sea (ICES) and discussed at the meeting of the ICES Working Group on Fishing Technology and Fish Behaviour (WGFTFB) in 2006. These questions led to field strength measurements in situ onboard the commercial beam trawler, and research on the effects of pulse stimulation on cod (*Gadus morhua* L.), and elasmobranch fish. The initial study revealed a potential problem concerning spinal damage in cod, and suggested only weak responses and no mortality in lesser spotted dogfish (*Scyliorhinus canicula* L.) caused by the electric stimuli, but due to the strong effect of the measurement protocol on feeding behaviour it was suggested that more experimentation was needed (Van Marlen, *et al.*, 2007). This report gives the results of further experiments a representative species of elasmobranch fish, lesser spotted dogfish (*Scyliorhinus canicula* L.). The intention of the research is to give insight in the effects of the electric stimulus applied by Verburg Holland-Ltd. in terms of injuries, survival, and food intake.

# 2. Materials and Methods

## 2.1. Organisation research protocol and procedures

The research was started on 01 June 2008 and commissioned by the Directorate of Fisheries of the Dutch Ministry of Agriculture, Nature and Food Quality and in advance of the experiments several meetings with Imares staff were conducted to set up the research protocol and to develop all requirements. The pulse simulator equipment was equal to the system used in the experiment on cod in 2008 (de Haan *et al.*, 2009) with pulse characteristics equivalent to the nominal menu settings of the commercial full-scale system, which represent the average settings of the pulse properties, such as pulse width, amplitude and repetition rate. In practice these settings can be adapted to seasonal conditions and can be varied in a range of +/- 20 % of the nominal condition. In summertime the amplitude settings are normally reduced by 10 %, while in wintertime these are usually raised by 10 % above the nominal value. Technical details of the pulse equipment is illustrated in section 2.2 and Annex 1, Pulse simulation equipment.

On 12 August 2008 the research protocol was approved by the Dutch board for animal welfare DEC (Dieren Experimenten Commissie).

## 2.2. Holding conditions and food regime

In the week of 10 November the fisheries research vessel (FRV) "Tridens" of the Ministry of Agriculture, Nature and Food Quality caught 80 lesser spotted dogfish (*Scyliorhinus canicula* L.). The fish were accustomed under ideal conditions in the onboard saltwater aquarium and transported on 13 November to the IMARES laboratories in IJmuiden. During transportation the water was oxygenated by using a bottle with compressed oxygen in combination with a diffuser that was present on the bottom of each tank. Upon arrival the fish were randomly divided over three holding tanks (2 x 2 x 0.5 m). Each group contained a similar length distribution. The length of the fish varied between 0.3 m and 0.65 m.

Before the experiments a dose of 500 grams sardine was manually divided over all three groups three times a week (Monday, Wednesday and Friday).

A week after arrival the fish deemed fully accustomed to the holding conditions and feeding regime, but of the total amount 2 dogfish died. On 3 December, a day before the actual start of the experiment, the food was reduced to a dose of 150 grams for all three groups.

The research was conducted in the IMARES indoor aquarium in four grouped square tanks, each of 2 x 2 m. In this facility the animals were kept after the exposure. The tank, in which the electric exposures were conducted, was of similar dimensions and was positioned in an adjacent room. Water was derived from the Imares standard saltwater store and circulated continuously through the tanks, except through the test tank in the period of the electrical exposures. Water in this tank was refreshed in the periods between each of the four exposure tests. The water level of the tanks during the recovery period was 1 m and during the electric exposures the water level in the test tank was reduced to 0.6 m. Conductivity, temperature and oxygen of the water in the test tank were measured after the exposures using a Hydrolab DS5 datasonde. The averaged results of 60 samples were for salinity 32.05 (ppt), temperature 16.4 °C and dissolved oxygen 56.6 %.

Each tank was provided with a video camera situated approx. 1 m above the water surface to monitor fish behaviour during and after the exposures and during the 14 days observation period. The test tank in which individual fish was exposed was provided with an additional underwater camera to observe the response of the fish to the stimulus and to trigger the exposure when the fish was within the required distance from the electrodes. The video equipment was controlled close by the tanks. During the 14 days observation period the recordings were automated on fixed periods of 15 minutes during the feeding period (each morning starting at 09:30, 5 days a week) and additionally at 17:00 (7 days a week). The first three feedings after the exposures the food dose of 180 grams was divided over all four tanks. On 9 December this amount was increased to 150 grams per group.

## 2.3. Pilot test and preparations of the electric exposures

On 3 December 2008 a pilot test was conducted in accordance with the methodology used in the cod experiment in Austevoll, Norway (de Haan et al., 2009). Three individuals were exposed to the nearest range from a conductor (0.1 m from the center of a conductor), the "near field" range. The effect of the exposures was not as strong as on cod, but stronger than the results of the pilot study on lesser spotted dogfish in 2007 (Van Marlen et al., 2007). In that experiment the fish was not exposed in close range of the conductors and the fish hardly reacted to the exposures. In this case the fish contracted during the exposures and after the pulse extinguished the fish reversed 180° in a sharp curve with rapid swimming behaviour towards the water surface. After the exposures the fish were released in a recovery tank and behaved normally. Five small sardines of 50 mm were offered in the evening after the experiment and this food appeared all eaten the morning after. This short test showed that additional measures, such as X-ray photography, were not required.

On 4 December 2008 the fish were randomly divided in four groups of 16 and exposed in three different distances ranges. A fourth group of fish was kept as control reference and not electrically exposed, but only subdued to the transferring operation in a similar manner to the other groups of fish, to enable discrimination of the effects resulting from transfers from effects caused by exposure by the electrical stimuli. The complete experiment took a period of 7 hours in total. The order of execution was "near field", "above field", "far field" and finally the transfers of the control group.

Individual fish were exposed 4 times to the electric stimulus in three different distance ranges from the conductor elements of the electrode pair. As the full-scale system contains 6 conductors in 4 m long electrodes, the



passage of commercial full-scale electrodes at a towing speed of 4 knots would induce 6 exposures in 2 seconds to a stationary fish.

The three exposure categories were:

1. A “far field” range with the fish exposed at 0.4 m side ways of a conductor element.
2. A “above field” range with the fish exposed at 0.1-0.3 m above the center of a conductor pair;
3. A “near field” range with the fish exposed at 0.1 m from the conductor element;

Field strength in the described ranges and pulse characteristics were measured the day after the experiments according the methods used on measurements sessions on the electric pulse systems (Van Marlen et al., 2007).

Shortly before the exposures, half amount of the total of each group were transferred to a mobile holding tank of 1.4x1.1x0.7 m, which was filled completely with sea water and aerated. This tank was positioned close to the entrance of the test tank facility and from this tank groups of five fishes were taken into a smaller standard fish container of 0.8x0.4x0.25 m.

From this box individual fish was manually transferred to a cage of flexible polyethylene netting (Figure 1) with a triangular profile of 0.6x0.3x0.55 m (length x width x height), also used on the experiments with cod (de Haan et al., 2009). The length of each fish was measured before or after the exposures using a ruler, which was attached on top of the tank wall (Figure 1). The polyethylene netting had a mesh size of 20 mm, wide enough to observe the caged fish by underwater camera. The cage was covered on top with nylon netting (with an opening in the middle to load the fish through) to avoid fish from jumping out the cage as a reaction to the exposures. With this cage, the fish could be accurately positioned in the required distance range, while the fish was still able to swim freely in the limited area. The sequence of four exposures lasted on average 2 minutes, and depended on the behaviour of the fish prior to the exposure and the reactions to the exposures. Each exposure was filmed underwater and 1.0 m above the water surface.

After the four exposures the cage with the exposed fish was moved through air and the fish immediately released in another mobile aerated holding tank, which was within a range of 2 m distance from the exposure tank. When half of the amount of fish was processed the exposed fish was transferred to the recovery tank and video observations were started.

## 2.4. Electric pulse simulator, pulse properties and field strength measurements

The electric stimulus was generated using the Verburg-Holland Ltd. simulator model (Annex 1 Pulse simulating equipment), of which the output parameters were similar and thoroughly checked against the full-scale electric beam trawl system used onboard “Lub Senior” UK 153 (Van Marlen, et al., 2007). The simulated electrodes (Figure 1) consisted of a single pair of two electrodes provided by Verburg-Holland Ltd. with equivalent conductors, materials, isolated sections as used in the full-scale system, but with reduced number of conductors (2 pairs instead of 6), reducing the length from 4 m to 0.96 m with an isolation extension of 0.6 m between both conductor pairs (Figure 2). The distance between electrodes was set to the nominal design value of 325 mm. Assuming a towing speed of 4 knots of the commercial pulse trawl system, the electric field around a single pair of conductors will pass a stationary fish in approx. 0.5 s. In this approach a worst case condition was simulated with 4 successive exposures of a 1 s burst of pulses of constant amplitude with pulse parameters characterized as “nominal” in the Verburg electric beam trawl system. The exposures were triggered when the fish was positioned in the required distance range. The pulse controller was set according the nominal menu settings illustrated in Annex 1 Pulse simulating equipment.

### 2.4.1. Pulse parameters measured

Measurements of the electrical stimulus focused on the main parameters:

- Amplitude in V;
- Pulse width in  $\mu$ s;
- Rise and fall times in  $\mu$ s;
- Repetition rate in Hz;
- Electric field strength in V/m between the electrodes.

#### 2.4.2. Field strength measurements

The pulse characteristics and field strength around the conductors were similar to the outcome of the measurements on the commercial system in the Verburg-Holland Ltd. facilities and during the first pilot experiment on dogfish in 2007 in the IMARES laboratories (Van Marlen et al., 2007). Field strength was measured on 5 December 2008 in the test tank. The measurements were conducted with the high voltage probe ADP305 and stored on LeCroy WaveSurfer 24XS oscilloscope.

Under nominal system settings, the peak field strength measured 0.1 m cm opposite the center of a conductor was 162 V/m, corresponding to the peak value of the “near field” range.

Field strength measured 0.1 m above the center of a pair of conductors was 99.6 V/m.

Field strength 0.15 m above the center of a pair of conductors was 64 V/m.

Field strength 0.2 m above the center of a pair of conductors was 48 V/m.

Field strength 0.4 m opposite the center of a conductor element was ignorable 8 V/m.

Readings measured through the cage matched the cases without cage and the outcome was in the range of the field strength measured on the cod experiment (de Haan et al., 2009).

#### 2.4.3. Measurement system

The pulse output parameters and field strength in the defined ranges from the conductors of the electrodes were measured shortly before the start of the exposures using a 200 MHz LeCroy WaveSurfer 24XS oscilloscope with 2 differential probes, a high voltage type ADP 305 (SN5069) and a AP031 70V probe for field strength measurements and a CWT Rogowski 60B current probe (0.5 mV/A) to measure the electrode current. Samples of measurement results were stored as JPEG images on hard disc of the oscilloscope. Electric field strength was measured in the plane of the electrodes with a probe of fixed spacing of 25 mm along three longitudinal rulers with grid units of 36 x 100 mm. These rulers were spaced in the center between the electrodes, and two 100 mm at either side of the center ruler. The probe was positioned with the center on each marker and tips perpendicular to the longitudinal rulers.

### 2.5. Video observations

The behaviour of the fish during the exposures and the recovery period were observed above the tanks and underwater in close range of the electrodes using two Sony HQX CCD 560 TVL bullet video cameras and a 4 channel X-SportsCam Extreme III MPEG-2 Solid-State recorder, provided with flashcard memory to store the video data. The two video recordings during the exposures were grouped in one stream of four programmable squared windows. The additional two windows were filled with images taken from the video cameras above the recovery tanks in which the exposed fish were transferred. For the complete recording cycle two flashcards of each 16 Gb were used. Sequentially the videodata were transferred to computer harddisc.

### 2.6. Analysis of the effects of the electric stimulus

#### 2.6.1. Pre-experimental observations

Standard behaviour of the fish was observed in advance of the experiment. A restricted amount of data were available as the period between transport and the execution of the tests was relatively short. However, the conditions after the catch on board FRV “Tridens” and the transport to the Imares laboratory were optimum. The fish habituated quickly to the food and only two fishes died after arrival. Normal behaviour can include passive behavior at the bottom in groups or solitarily, slowly swimming at the bottom and surfacing with the beak at the surface or even in the air. On the moment of the feeding session the fish became active one by one and this took a few minutes. Once the food was taken the fish returned to normal behaviour moments after.

#### 2.6.2. Injuries and mortality

In the 14 days observation period following the exposures, all fish were kept under observation in order to investigate whether they had sustained injuries or died.

### 2.6.3. Feeding response

For a period of 14 days the behavior of the fish of each of the four groups was filmed during the feeding session, which was scheduled daily (Monday to Friday) at 09:30 and on a second moment at 17:00 (including the weekends). The food, a dose of 140 to 170 grams per group, was offered manually the first two cycles. On the first day after the exposure two cycles of this dose was offered, a second at 16:00. From 9 December 2008 the dose was increased to 150 grams per group. All video recordings were automated to capture 5 minutes in advance of the feedings and 10 minutes from the start of the feedings. At 17:00 a period of 5 minutes was captured (including the weekends). Each tank was equipped with a video camera mounted 1 m above the center of the tanks. Video recordings were stored on flashcard memory.

### 2.6.4. Reactions to the exposures

Although this experiment has not been set up, or designed, to investigate differences between exposure groups in terms of behavioural responses other than feeding responses, it is important to monitor the direct behaviour of the fish to the stimuli. The behaviour of the fish as a post-reaction to the exposures were analysed and sorted to determine if this behaviour has relevance to the operation and by-catches of the full-scale electric gear. We categorised the effects of the electric stimulus a number of categories, of which the following states are most likely to occur:

1. Contraction. During the exposure the animal will produce strong reflexes and muscle contractions, which paralysed the fish during the 1 s pulse period. The degree of this effect will be related to the size of the fish, the exposed range and corresponding field strength.
2. Post-reaction. At the moment the pulse train extinguishes the animal will respond with a certain behaviour related to the physical impact of the exposure. The degree of the effect is related to the size of the fish, the exposed distance range and corresponding field strength. For example, a typical post-reaction was for fish a rapid body reverse, short-curved body rotations and acceleration towards the water surface.

## 3. Results

### 3.1. Injuries and mortality

No evidence was found of differences in likelihood of injury or death between the exposure groups. There was no evidence that fish sustained injuries as a result of the exposures. Respectively 8 and 9 months after the experiment a single specimen of the “above field” category and “near field” category died.

### 3.2. Behaviour after the exposures and feeding response

The fish released in the observation tanks directly after the exposures were more active than before the start of the experiment. The feeding behaviour of the fish the first day after the exposures was normal and similar to the behaviour before the experiment. Within 5-10 minutes the food was completely taken.

In the 14 days observation period after the exposures no aberrant feeding behaviour could be distinguished. Fish in all tested groups started feeding normally the same day directly after the exposures. Most of the food (160 g in total, which is 25 % of the normal dose) was taken after 5-10 minutes (Annex 3 Feeding response).

### 3.3. Reactions to stimuli

Overall, there was a tendency for fish to respond in a similar way to the four successive exposures. It therefore seemed that the response was partly determined by the condition of the animal itself at or before the moment of the exposures. For this reason, apart from presenting numbers of exposure-cases with a particular response, we also present our results in terms of numbers of animals that showed a particular behaviour at least once (in the latter case, the number of statistical significant cases reduce from 64 to 16).

It seemed that smaller fish ( $\leq 0.3$  m) reacted less pronounced to the electric exposures than the largest fishes (0.5-0.6 m). However, it should be noted that the number of small fish in the dataset were low (only 3 (Annex 2 Table 1, 2, 3)).

In the “far field” range, there were few responses of fish to the exposures (Annex 2 Table 1 and 2). Only in 5 out of 64 cases a minor reverse behaviour was noticed and in a single case an eye-closure. As the fish hardly reacted and stayed passively in position, the repetition rate of the exposures was much higher than in other categories. The shortest full cycle of 4 exposures was 5, the longest 31 s. (Annex 2 Table 1, column 3).

In the “above field” range, fish commonly showed some type of response to the exposures (Annex 2 Table 3 and 4). A total of 55 cases were available for analysis (9 cases are disqualified (trial 5, 4 faulty triggers and a missing case (trial 16))). The results showed that in this range contractions still occurred with a share of 33 out of 55 cases  $\approx 60\%$ . Among these 9 cases with special reactions, in 6 out of 9 cases tail-bends and/or body-curles were observed. Additionally in 3 cases an eye-closure was observed during the exposure.

A rapid body reverse was observed as a post-reaction when the electric stimulus extinguished. This phenomenon occurred in 29 out of 55 cases and was observed in two broad classes; rapid sharp reverses, and a slower action with the same effect (moderate reverse). In a number of cases the body reverses ended in a reaction upward due to the restriction of the physical dimensions of the cage against the body length of the fish. In 2 out of 55 cases a moderate reverse was observed.

A minor reaction upward was observed in 6 out of 55 cases. In most cases this reaction was followed by swimming with the head at or above the surface.

Of the total number of exposures (64) in the “near field” range 61 were available for analysis (Annex 2 Table 5 and 6). Three cases were cancelled due to a faulty trigger (1) or abortion (2) by failed positioning of the fish (continuous surfacing). A complete cycle of 4 exposures took either 36 seconds as minimum or 8min36s as maximum. In most cases a body contraction was observed when the electric stimulus was activated (55 out of 61 cases  $\approx 90.1\%$ ). The body shape during contraction was slightly bent with a occasionally typical bend in the tail and in some cases the beak was opened (not always visible). A series of successive video frames, showing the contraction and post-reaction (reverse) of a dogfish in the “near field” range, is illustrated in Figure 3. After the pulse extinguished the animal responded with:

- A reverse (38 out of 61 cases= $62.3\%$ ) either followed by swimming upward or passive behaviour at the bottom;
- Swimming upward rapid or slower (14 out of 61 cases);
- Some turns with body curled up ending in an unfolded reversed position (7 out of 61 cases). In 4 out of 4 cases the fish arrived curled up and did not unfold after the exposure (Figure 4).
- A weak reaction in 3 out of 65 cases, concerning a small fish of 0.3 m length.

The numbers of cases of upward reactions per fish are given in Table 7, as well as.

The numbers of cases with the fish positioned horizontally on the bottom (between brackets). The chance of a reaction upward increased when the animal was exposed with the head pitched upward. Since there were only a few instances when fish were positioned horizontally, no firm conclusion can be drawn on this aspect.

Table 7 Numbers of cases of upward reactions per fish as observed in the three exposed distance ranges. Numbers of cases with the fish positioned horizontally on the bottom are given in brackets.

Number of fish reactions upward			
Trial (nr)	Near field (nr)	Above (nr)	Field (nr)
1		3 (0)	
2	3 (1)	4 (1)	
3	3 (0)	4 (1)	
4		4 (0)	
5	3 (1)		
6		2 (0)	
7	2 (2)	2 (1)	
8			
9		4 (1)	
10		4 (0)	
11			
12		1 (1)	
13	3 (2)		
14		3 (0)	
15		4 (0)	
16		3 (0)	
Total	11 (6)	38 (5)	0

### 3.4. Additional long-term observations

Next to the mortality scores over a long period (9 months) egg production was monitored as a standard routine within the Imares husbandry procedures. In a period of 7 months after the exposures all exposed groups produced eggs in numbers varying between 5-39 per group. Surprisingly the control group did not produce eggs.

Table 8 Overview of egg production in the three exposure ranges ("above field", "far field" and "near field" and the control group (Ref)). Given are the cumulative numbers of eggs over time.

	Above	Far	Near	Ref
12-01-2009	1	10	12	
26-01-2009		16	14	
23-02-2009	2	29		
16-03-2009	4	37	19	
12-05-2009				
15-07-2009	5	39	21	

## 4. Discussion

No evidence was found of differences in feeding response or likelihood of injury or death between the exposure groups. No fish were found to have injuries. Respectively 8 and 9 months after the experiment a single specimen of the “above field” category and “near field” category died. It is not possible to link these deaths to the experiment, as these deaths could have easily occurred as a result of natural causes. In the 14 days observation period after the exposures no aberrant feeding behaviour was observed.

We did not find evidence of a relationship between the degree of exposure and the production of eggs (which is an important indicator for physical welfare of the fish). Surprisingly the fish of the control group that were not exposed to the electric stimulus, did not produce eggs. A possible explanation for this could be that the control group consisted of a high number of males. However, we did not measure the sex contribution per group.

Regarding the other behavioural responses (mainly reflexes and muscle contractions, and post-reactions, such as rapid body reverses, short-curved body rotations and acceleration towards the water surface), there were some clear differences between exposure groups. The responses of the fish exposed in the “far field” range, representing the fish just aside the fished area of the trawl, were minor and ignorable. However, the responses of the fish exposed in the “above field” range were more pronounced with contractions, rapid body reverses, short-curved body rotations and acceleration towards the water surface occurring. The responses of the fish exposed in the shortest possible range, the “near field” range, were the strongest with increased incidence of contractions and rapid body reverses, short curved body rotations and acceleration towards the water surface. Although this experiment has not been set up, or designed, to investigate differences between exposure groups in terms of behavioural responses other than feeding responses, we note that a common behavioural response in the “near field” group was to accelerate upward. However, we note that no conclusions with regard to this particular behavioural aspect can be drawn on the basis of this study, since this study was not designed to investigate this. For example, there were many instances in which fish were positioned upwards, making an upward reaction possibly more likely (Table 7). Nevertheless, a tendency to accelerate upward in response to the exposures may be relevant since, in field situations, dogfish have been observed to become entangled in the meshes of the top panel of the full-scale pulse trawl (pers.com Tammo Bult). This may well have been caused by a tendency to accelerate upwards in response to electric exposure.

A study on the survival rate of lesser spotted dogfish in Spanish trawler fisheries (Rodríguez- Cabello et al., 2005) showed that commercially discarded lesser spotted dogfish have a survival rate of 78 %. Combined with the results of the current study,(no evidence of increased likelihood of injury or mortality as a result of exposure to the stimuli) this indicates that lesser spotted dogfish caught in the pulse trawl have a reasonable chance of survival as long as the fish do not become entangled in top panel meshes. It may be possible to make some adaptations to the gear to avoid this from happening, such as enlarging top panel meshes. This aspect merits further investigation.

It seemed that smaller fish (0.3 m) reacted less pronounced to the electric exposures than the largest fishes (0.5-0.6 m). However, it should be noted that there were only 3 small fish ( $\leq 0.3$  m) in the dataset (Annex 2, Table 1, 3, 5). A theoretical study on the effects of field strength in relation to fish length (Stewart, 1975) shows that fish reactions to electric fields are length dependent with lower effects for smaller fish. Smaller fish need to be positioned more accurately at spots of highest field strength to evoke similar effects. However, our study included an insufficient number of small fish. A practical constraint is that the amount of small fish in the catch at sea cannot be controlled. Consequently the effects on smaller fish remain uncertain and may need further attention.

## 5. Conclusions

No evidence was found of an effect of the degree of the electric exposure on feeding response or likelihood of injury or death. There was no evidence that fish sustained injuries as a result of the exposures. Respectively 8 and 9 months after the experiment a single specimen of the “above field” category and “near field” category died. In the 14 days observation period after the exposures no aberrant feeding behaviour could be distinguished. Fish in all tested groups started feeding normally the same day directly after the exposures. In a period of 7 months after the exposures all exposed groups produced eggs in numbers varying between 5-39 per group.

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## 7. Figures and Illustrations



*Figure 1 Video image of the of the research tanks with top left the underwater side view on the exposed area with the cage of flexible polyethylene netting positioned between the conductors and top right the view from above with a dogfish being measured. Bottom left is the “near field” recovery tank and bottom right the “above field” recovery tank (the date information is invalid (recorder config bug).*



*Figure 2 Underwater detail of the conductors fixed at a distance of 325 mm and the isolated extension of 0.6 m.*



Figure 3 A series of successive video frames, showing the position of the animal at the bottom, top left, and the contraction (top right) and post-reaction (reverse), bottom left and right, of a dogfish exposed in the “near field” range.



Figure 3 Sequence of a dogfish exposure in the “near field” range. The fish arrived with a body curl (top picture left), after the exposure (top right picture (note the two LED indicators activated at the left hand of the operator). The fish unfolded as a post-reaction (bottom picture), but adopted the original body curl shortly after.

## 8. Annex 1 Pulse simulating equipment

The pulse generating equipment consists of the following main parts:

An adjustable power supply (A);  
Pulse generator (B);  
Output inductance (C);  
Electrodes (D);  
Oscilloscope (E).

Adjustable power supply (A)

Power supply output provides a DC intermitting voltage of 0-200 V.

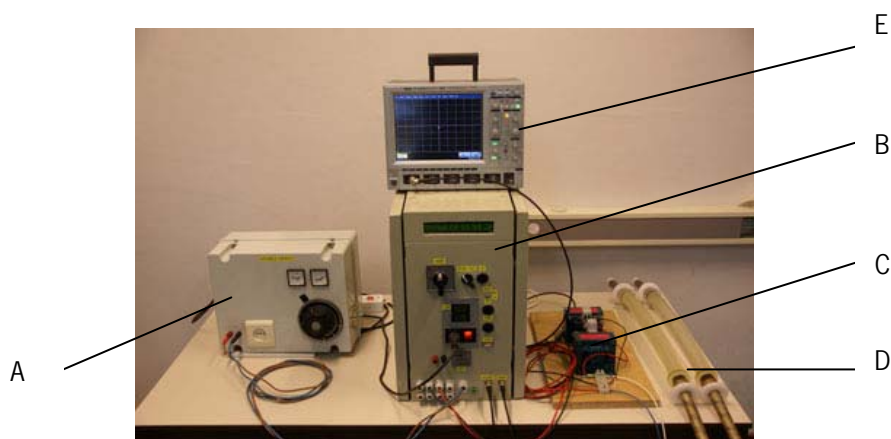
Pulse generator (B)

The pulse generator consists of a microprocessor and a pair of IGBT's. With the controller the main parameters of the stimulus, amplitude, pulse shape, pulse width etc. are programmed.

### Menu settings

Pulsevorm	(Pulse shape)	P1;
Length T3	(Burst length)	1.0 s;
Period T2	(Pulse period)	nominal;
Pulse T1	(Pulse width)	nominal;
Powervorm	(Power shape)	P0;
Cont/single	(pulse out)	s single;
Number T4		Overruled by T3;
Aantal	(number)	1
Powerinst.	(power setting)	Amplitude 88 %

Equipment layout



## 9. Annex 2 Analysis of the behavioural responses

*Table 1 Overview of behavioural responses of dogfish exposed in the "far field" category.*

Dogfish "far field" category (0.4 m beside the center of a conductor)							
Trial (nr)	File Time	Exp Time (m:ss)	Body length (m)	Exposures and observed behavioural effects			
1	14:44 14:46	0:16	0.6	No reaction	No reaction	Minor reaction (eyes)	No reaction
2	14:46 14:48	0:24	0.5	Weak reverse	No reaction	No reaction	No reaction
3	14:49 14:50	0:07	0.4	No reaction	No reaction	No reaction	No reaction
4	14:50 14:52	0:31	0.55	Weak reverse	No reaction	Weak reverse	No reaction
5	14:53 14:56	0:15	0.6	No reaction	No reaction	No reaction	No reaction
6	14:56 14:58	0:06	0.6	No reaction	No reaction	No reaction	No reaction
7	14:59 15:00	0:06	0.65	No reaction	No reaction	No reaction	No reaction
8	15:01 15:03	0:29	0.4	No reaction	No reaction	No reaction	No reaction
9	15:06 15:06	0:06	0.55	No reaction	No reaction	No reaction	No reaction
10	15:07 15:09	0:31	0.5	No reaction	No reaction	No reaction	Weak reverse
11	15:13 15:15	0:08	0.5	No reaction	No reaction	No reaction	No reaction
12	15:17 15:19	0:32	0.55	Weak reverse	No reaction	No reaction	No reaction
13	15:19 15:22	0:05	0.55	No reaction	No reaction	No reaction	No reaction
14	15:23 15:23	0:05	0.6	No reaction	No reaction	No reaction	No reaction
15	15:24 15:24	0:05	0.55	No reaction	No reaction	No reaction	No reaction
16	15:25 15:28	0:40	0.25	No reaction	No reaction	No reaction	No reaction

Table 2 Additional comments on the observed behaviour in the "far field" category.

Case (nr)	Additional behavioural observations "far field" category
1	Animal remained passive on the bottom during all 4 exposures. No physical reactions other than a weak eye closure on the 3 <sup>rd</sup> exposure.
2	The 1 <sup>st</sup> and 2 <sup>nd</sup> exposure the animal was at 0.3 m above the bottom. On the 3 <sup>rd</sup> and 4 <sup>th</sup> at the bottom.
3	The animal was lowering during the exposure towards the bottom. The 1 <sup>st</sup> exposure was at 0.1 m above the bottom, on the other exposures the animal was passively at the bottom.
4	The 1 <sup>st</sup> exposure was at the bottom, the animal reversed upward after the exposure, the 2 <sup>nd</sup> and 4 <sup>th</sup> exposure the animal was at 0.2 m above the bottom. On the 3 <sup>rd</sup> exposure at 0.1 m above the bottom.
5	The animal was exposed 0.1 m above the bottom, the 2 <sup>nd</sup> at 0.2 m, the 3 <sup>rd</sup> at 0.1 m and the 4 <sup>th</sup> at the bottom.
6	During the 4 exposures the animal lowered passively horizontally from 0.1 m above the bottom to zero with no visible reaction.
7	The 1 <sup>st</sup> exposure was 0.1 m above the bottom, the other exposures at the bottom.
8	Animal was exposed 0.3 m above the bottom, on the 2 <sup>nd</sup> , 3 <sup>rd</sup> and 4 <sup>th</sup> at 0.1 m above the bottom.
9	Animal completely passive on the bottom of the cage.
10	Animal reacted only on the 4 <sup>th</sup> exposure with a weak reverse.
11	Animal completely passive on the bottom of the cage.
12	On the 1 <sup>st</sup> and 2 <sup>nd</sup> exposure the animal was exposed at 0.2 m above the bottom.
13	Animal completely passive on the bottom of the cage.
14	Animal completely passive on the bottom of the cage.
15	Animal completely passive on the bottom of the cage.
16	Animal completely passive on the bottom of the cage.

Table 3 Overview of behavioural responses of dogfish exposed in the “above field” category

Dogfish “above field” category (0.10-0.30 m above the center of a conductor pair)							
Trial (nr)	File Time	Exp Time (m:ss)	Body length (m)	Exposures and observed behavioural effects			
1	12:54 12:56	0:40	0.6	Sharp reverse up	Faulty trigger	Minor Acceleration	Minor reaction upward
2	12:58 12:59	1:08	0.6	Minor reaction upward	Minor reaction upward	Contraction acceleration up	Contraction rapid reverse up
3	13:00 13:06	4:56	0.5	Contraction Sharp slow reverse	Contraction	Faulty trigger	Contraction Sharp slow reverse Contraction Slow reverse
4	13:06 13:08	0:44	0.6	Rapid reaction upward	Rapid reaction upward	Reverse strong reaction upward	Reverse strong reaction upward
5	Not Rec	n.a.	0.6	Moderate reaction	Moderate reaction	Moderate reaction	Moderate reaction
6	13:14 13:15	0:26	0.45	Contraction reverse	Faulty trigger	Reverse rapid up	Reverse rapid up Contraction sharp reverse
7	13:16 13:22	2:10	0.6	Contraction rapid up (eyes)	Contraction rapid reverse	Reverse rapid up	Contraction rapid up
8	13:30 13:31	0:29	0.6	Contraction curl unfold full turn	Faulty trigger	Contraction strong reverse	Contraction curl unfold full turn Contraction curl unfold full turn
9	13:37 13:40	1:49	0.5	Contraction rapid reverse upward	Contraction rapid reverse upward	Rapid reverse upward	Rapid reverse upward
10	13:41 13:43	0:35	0.55	Reverse rapid upward	Rapid reverse upward	Rapid reverse	Contraction rapid reverse upward (eyes)
11	13:44 13:54	9:33	0.5	Contraction	Rapid reverse	Rapid reaction upward	Rapid reaction upward
12	13:55 13:57	0:27	0.5	Contraction curl full turn	Contraction reaction upward	Contraction rapid reverse (eyes)	Contraction remaining curled
13	13:58 13:06	0:42	0.5	Contraction rapid reverse bottom	Contraction rapid reverse bottom	Contraction reverse bottom	Contraction full turn bottom
14	14:00 14:02	0:28	0.5	Contraction reaction upward	Moderate reverse	Contraction rapid reserve at the bottom	Contraction rapid reaction upward
15	14:03 14:04	0:43	0.33	Minor reaction upward	Moderate reverse	Minor reaction upward	Minor reaction upward
16	14:05 14:06	0:43	0.54	Contraction head arched 90° rapid up	Contraction reaction upward	Contraction semi-curl reaction upward	Contraction semi-curl reaction upward

Table 4 Additional comments on the observed behaviour in the "above field" category.

Trial (nr)	Additional behavioural observation "above field" category
1	On the 1 <sup>st</sup> exposure the animal was at 0.1 m above the bottom, 2 <sup>nd</sup> , 3 <sup>rd</sup> and 4 <sup>th</sup> exposure the animal was 0.3 m above the bottom.
2	On 1 <sup>st</sup> and 2 <sup>nd</sup> exposure the animal was with at 0.2 m above the bottom beside the other conductor pair. On the 3 <sup>rd</sup> exposure the animal was pitched up ward with the head 0.1 m above the bottom. On the 4 <sup>th</sup> exposure the animal was at the bottom.
3	The 1 <sup>st</sup> exposure at the bottom, 2 <sup>nd</sup> exposure the animal was pitched upward with the head 0.2 m above the bottom. The 3 <sup>rd</sup> exposure was triggered with the head of the animal not between the conductors, the animal reacted with a reverse. The 4 <sup>th</sup> and 5 <sup>th</sup> exposure was with the animal 0.2 m above the bottom.
4	The 1 <sup>st</sup> , 3 <sup>rd</sup> and 4 <sup>th</sup> exposure the animal was 0.2 m above the bottom, on the 2 <sup>nd</sup> the animal was pitched up with the head 0.2 m above the bottom.
5	Video recording failure.
6	On the 1 <sup>st</sup> exposure the animals was at the bottom, on the 2 <sup>nd</sup> 0.1 m on the 3 <sup>rd</sup> the animal was at the bottom, on the 4 <sup>th</sup> 0.2 m above the bottom.
7	The 1 <sup>st</sup> and 4 <sup>th</sup> exposure the animal was pitched up with the head 0.2 m above the bottom. On the 1 <sup>st</sup> exposure eye closure was observed. On the 3 <sup>rd</sup> exposure the animal was pitched downward with the head at the bottom.
8	The 2 <sup>nd</sup> exposure was triggered with the animal between the conductors and the head at the surface. The 3 <sup>rd</sup> exposure the animal was 0.2 m above the bottom. The 4 <sup>th</sup> and 5 <sup>th</sup> exposure the animal was pitched up with the head 0.1 m above the bottom.
9	On the 1 <sup>st</sup> exposure the animal was pitched downward with the beak at the bottom. On the 2 <sup>nd</sup> exposure the animal was pitched downward with the head at the bottom (tail at 0.1 m). On the 3 <sup>rd</sup> and 4 <sup>th</sup> the animal was pitched up was the head at 0.3 m above the bottom.
10	The 1 <sup>st</sup> and 2 <sup>nd</sup> exposure was triggered with the animal 0.2 m above the bottom, the 3 <sup>rd</sup> with the animal 0.3 m above the bottom. On the 4 <sup>th</sup> exposure the animal was pitched up with the head 0.2 m above the bottom, the animal closed eyes during the exposure.
11	The 1 <sup>st</sup> exposure the animal was pitched up with the head 0.2 m above the bottom. The 2 <sup>nd</sup> exposure the animal was horizontally at 0.1 m above the bottom. The 3 <sup>rd</sup> and 4 <sup>th</sup> exposure the animal was pitched up with the head at 0.3 m above the bottom. Long session, the animal swam mostly with the head at the surface and lowered occasionally.
12	All exposures with the animal at the bottom. On the 1 <sup>st</sup> exposure the animal reacted with a full turn and a reverse. On the 2 <sup>nd</sup> and 4 <sup>th</sup> exposure a tail bending was observed. On the 3 <sup>rd</sup> exposure the animal closed eyes during the exposure. On the 4 <sup>th</sup> exposure the animal remained curled for a few seconds.
13	All exposures with the animal at the bottom. The reverses were very rapid and all with a body curl. The final exposure was responded with a full body turn. The contractions were strong and with tail bends.
14	The 1 <sup>st</sup> exposure was triggered with the animal at 0.2 m above the bottom. On the 2 <sup>nd</sup> exposure the animal was at 0.3 m above the bottom. On the 3 <sup>rd</sup> exposure the animal was pitched downward with the beak at the bottom. On the 4 <sup>th</sup> exposure the animal was pitched upward with the head at 0.3 m above the bottom.
15	The 1 <sup>st</sup> exposure the animal was pitched upward with the head 0.3 m above the bottom. On the 3 <sup>rd</sup> exposure the animal was at 0.2 m above the bottom. On the 2 <sup>nd</sup> at 0.1 m. On the 4 <sup>th</sup> exposure the animal was pitched upward with the head at 0.3 m above the bottom.
16	Only three exposures. The 1 <sup>st</sup> and 2 <sup>nd</sup> exposure was triggered with the animal at 0.1 m above the bottom. On the third the animal was pitched downward with the head at the bottom.

Table 5 Overview of behavioural responses of dogfish exposed in the “near field” category.

Dogfish “near field” category (0.10-0.20 cm from a conductor)							
Trial (nr)	Time	Exp time (m:ss)	Body length (m)	Exposures and observed behavioural effects			
1	10:09 10:14	3:11	0.4	Contraction sharp reverse	Contraction sharp reverse	Contraction sharp reverse	Contraction sharp reverse
2	10:15 10:21	5:35	0.4	Contraction sharp reverse, acceleration up	Contraction strong reaction upward	Faulty trigger	Contraction sharp reverse Contraction reaction upward
3	10:23 10:33	8:35	0.65	Contraction sharp slow reverse	Contraction, weak reaction upward	Contraction, weak reaction upward	Contraction, weak reaction upward
4	10:34 10:35	1:06	0.6	Contraction sharp reverse	Contraction strong curl reaction	Contraction strong curl reaction	Faulty trigger Contraction, strong curl reaction
5	10:37 10:50	8:36	0.6	Contraction Strong reaction upward	Contraction weak reaction upward	Contraction weak reaction upward	Aborted, fish kept swimming at the surface
6	10:53 10:56	0:49	0.6	Body curl did not unfold. Minor reaction	Body curl did not unfold. Minor reaction	Body curl did not unfold. Minor reaction	Body curl did not unfold. Moderate reaction
7	11:03 11:04	0:29	0.6	Contraction reverse slow upward	Contraction reverse slow upward	Contraction Reverse slow upward	Contraction reverse slow upward
8	11:05 11:07	2:05	0.4	Contraction rapid reverse	Contraction rapid reverse	Contraction rapid reverse	Contraction rapid reverse
9	11:15 11:17	1:27	0.6	Contraction strong reverse	Contraction strong reverse	Contraction strong reverse	Contraction strong reverse
10	11:18 11:19	0:33	0.5	Contraction reverse	Faulty trigger	Contraction in position	Contraction reverse bottom
11	11:20 11:22	1:29	0.57	Contraction sharp curl reverse bottom	Contraction sharp curl reverse bottom	Contraction sharp curl reverse bottom	Contraction sharp curl reverse bottom
12	11:23 11:24	0:36	0.58	Contraction, sharp curl reverse bottom	Contraction, rapid reverse bottom	Contraction, rapid reverse bottom	Contraction, sharp curl reverse bottom
13	11:24 11:25	0:36	0.6	Contraction upward open beak	Contraction, upward	Contraction, reserve bottom/up	Contraction, reserve bottom/up
14	11:25 11:41	9:35	0.27	Contraction reverse bottom	Reverse bottom	Reverse bottom	Aborted, fish at the surface
15	11:59 12:02	0:37	0.5	Contraction folded turns reverse	Contraction folded turns folded 13 s	Contraction folded 16 s	Contraction folded turns folded 18 s
16	12:04 12:06	1:06	0.6	Contraction, reverse bottom	Contraction reverse bottom	Contraction strong reverse bottom	Contraction strong reverse bottom



Table 6 Additional comments on the observed behaviour in the “near field” range

Trial (nr)	Additional behavioural observations “near field” category
1	The animal remained passive at the bottom on all 4 exposures.
2	On the 2 <sup>nd</sup> exposure the animal was pitched upward with the head 0.2 m above the bottom. The 3 <sup>rd</sup> exposure was triggered too late with the animal out of range. The 5 <sup>th</sup> exposure the animal was pitched up with the head 0.2 m above the bottom.
3	The 1 <sup>st</sup> exposure the animal was at the bottom. The 2 <sup>nd</sup> exposure the animal was pitched up with the head 0.2 m above the bottom. The 3 <sup>rd</sup> exposure the animal was pitched up with head 0.1 m above the bottom. On the 4 the animal was pitched up with the head at 0.2 m above the bottom.
4	The animal was exposed at the bottom on all 4 exposures. On the 2 <sup>nd</sup> , 3 <sup>rd</sup> and 5 <sup>th</sup> exposure the animal reacted with a strong body curl.
5	Animal arrived very active in the cage and became more active after the exposures with the head near the surface and the body in vertical position. On the 1 <sup>st</sup> and 3 <sup>rd</sup> exposure the animal was pitched up with the head 0.2 m above the bottom. On the 2 <sup>nd</sup> exposure the animal was at the bottom. The 4 <sup>th</sup> exposure was cancelled for this reason.
6	The animal arrived with the body curled up. The animal maintained this condition and reacted to the stimulus by slight unfolding with the strongest reaction on the 4 <sup>th</sup> exposure. The animal maintained curled condition after the exposures.
7	On the 1 <sup>st</sup> and 2 <sup>nd</sup> exposure the animal was at the bottom, on 3 <sup>rd</sup> exposure the animal was pitched up with the head 0.1 m above the bottom. On the 4 <sup>th</sup> exposure the animal was pitched downward with the head at the bottom.
8	On the 1 <sup>st</sup> , 2 <sup>nd</sup> and 3 <sup>rd</sup> exposure the animal was at the bottom and reversed after the exposure, on 4 <sup>th</sup> exposure the animal was pitched up with the head 0.1 m above the bottom. Only after the 3 <sup>rd</sup> exposure the animal swam upward.
9	On all 4 exposures the animal was at the bottom and reversed sharply after the exposure. On the 3 <sup>rd</sup> exposure the animal also rotated 180° during on contraction.
10	On the 2 <sup>nd</sup> exposure the animal was not well positioned (reversed with the head along the isolated electrode part). After the first two exposures the animal avoided the area around the conductor. On the 3 <sup>rd</sup> exposure the animal was pitched vertically with the beak at the bottom and contracted in that position without reversing. On the 4 <sup>th</sup> contraction the animal curled and reversed when the pulse extinguished.
11	Same behaviour on all 4 exposures, strong contraction, the animal curled in a sharp bend during the contractions and reversed along the bottom in all 4 cases and remained passively after.
12	Same behaviour on all 4 exposures, strong contraction, the animal curled in a sharp bend during the contractions and reversed along the bottom and remained passively after.
13	The animal was pitched up with the head 0.3 m above the bottom, during the contraction the animal opened the beak, strong contraction, on the 2 <sup>nd</sup> contraction the animal was pitched up 0.1 m and reacted upward. On the 3 <sup>rd</sup> and 4 <sup>th</sup> exposure the animal reversed along the bottom and upward.
14	After the first exposure the animal remained passive in position and a second was triggered after 2 seconds, after which the animal reversed. After the 2 <sup>nd</sup> exposure the animal kept on swimming at the surface for 9.5 minutes. On the 3 <sup>rd</sup> exposure (36:22) exposure the animal reacted 1-2 seconds after the exposure with a reverse along the bottom. As the animal kept swimming at the surface the trial was aborted 4 minutes later.
15	Animal arrived curled up but unfolded shortly after. The animal contracted on the 1 <sup>st</sup> exposure and responded with some multiple turns and reversed after along the bottom. On the 2 <sup>nd</sup> exposure this behaviour was repeated, but this time the animal did unfold after 13 seconds at the bottom inside the exposed area. After the 3 <sup>rd</sup> exposure the animal unfolded in the exposed area. On the 4 <sup>th</sup> exposure identical behaviour the animal unfolded after 19 s and stayed passively at the bottom.
16	The animal contracted and reversed at the bottom maintaining passive behaviour.

## 10 Annex 3 Feeding response

### 10.1 Pre-observations

On arrival a number of 80 dogfish were divided randomly over tank A, B and D. Feedings once per day at 09:30.

Week nr. 46		Date:14-11-2008
Tank	Food dose (g)	Remarks
A		13-11 dogfish arrived
B		
D		

Week nr. 47		Date: 21-11-2008
Tank	Food dose (g)	Remarks
A		For all tanks: dogfish are eating well
B		1 dogfish swims odd
D		

Week nr. 48		Date: 25-11-2008
Tank	Food dose (g)	Remarks
A	total: 419	Dose for 3 tanks, all food eaten
B		
D		

Week nr. 48		Date: 26-11-2008
Tank	Food dose (g)	Remarks
A	total: 297	Dose for 3 tanks, all food eaten
B		
D		

Week nr. 48		Date: 26-11-2008
Tank	Food dose (g)	Remarks
A	total: 297	Dose for 3 tanks, all food eaten
B		
D		

Week nr. 48		Date: 27-11-2008
Tank	Food dose (g)	Remarks
A	total: 735	Dose for 3 tanks, all food eaten
B		1 dogfish died
D		

Week nr. 49		Date: 02-12-2008
Tank	Food dose (g)	Remarks
A	172	1 dogfish died
B	162	
D	132	

Week nr. 49		Date: 02-12-2008
Tank	Food dose (g)	Remarks
A	50	
B	50	
D	50	

Week nr. 49		Date: 04-12-2008
Tank	Food dose (g)	Remarks
A	none	This day exposures were executed
B	none	
D	none	

## 10.2 Post-observations

Tank	Week nr. 49	Date: 04-12-2008		Time: 18:30		Remarks
	Food dose (g)	before feeding		during feeding		
A	40	passive:		passive:		Day the exposures took place
		active:		active:		
B	40	passive:		passive:		Majority of all groups activated. In 5-10 min. all food was eaten
		active:		active:		
C	40	passive:		passive:		
		active:		active:		
D	40	passive:		passive:		
		active:		active:		

Tank	Week nr. 49	Date: 05-12-2008		Time: 11:27		Remarks
	Food dose (g)	before feeding		during feeding		
A	42	passive:		passive:	3	For all tanks: in 5 minutes all food eaten
		active:		active:	13	
B	42	passive:		passive:	3	
		active:		active:	13	
C	42	passive:		passive:	6	
		active:		active:	10	
D	42	passive:		passive:	2	
		active:		active:	14	

Tank	Week nr. 49	Date: 05-12-2008		Time: 16:00		Remarks
	Food dose (g)	before feeding		during feeding		
A	37	passive:		passive:	3	For all tanks: in 5 minutes all food eaten
		active:		active:	13	
B	37	passive:		passive:	3	
		active:		active:	13	
C	37	passive:		passive:	5	
		active:		active:	11	
D	37	passive:		passive:	1	
		active:		active:	15	

Tank	Week nr. 50	Date: 08-12-2008		Time: 09:30		Remarks
	Food dose (g)	before feeding		during feeding		
A	45	passive:	16	passive:	2	1 egg
		active:	0	active:	14	
B	45	passive:	15	passive:	3	For all tanks: in 5 minutes all food eaten
		active:	1	active:	13	
C	45	passive:	11	passive:	5	2 eggs
		active:	5	active:	11	
D	45	passive:	12	passive:	1	
		active:	4	active:	15	

Tank	Week nr. 50	Date: 09-12-2008		Time: 09:30		Remarks
	Food dose (g)	before feeding		during feeding		
A	45	passive:	15	passive:	2	For all tanks: in 5 minutes all food eaten
		active:	1	active:	14	
B	45	passive:	15	passive:	3	Response as before exposures
		active:	1	active:	13	
C	45	passive:	15	passive:	5	2 eggs
		active:	1	active:	11	
D	45	passive:	16	passive:	1	
		active:	0	active:	15	

Tank	Week nr. 50	Date: 10-12-2008		Time: 09:30		Remarks
	Food dose (g)	before feeding		during feeding		
A	136	passive:	16	passive:	1	
		active:	0	active:	15	
B	145	passive:	15	passive:	1	
		active:	1	active:	15	
C	145	passive:	16	passive:	5	Least response
		active:	0	active:	11	
D	145	passive:	15	passive:	1	
		active:	1	active:	15	

Tank	Week nr. 50	Date: 11-12-2008		Time: 09:30		Remarks
	Food dose (g)	before feeding		during feeding		
A	148	passive:	15	passive:	0	In 5 minutes all food eaten
		active:	1	active:	16	
B	150	passive:	15	passive:	0	In 5 minutes all food eaten
		active:	1	active:	16	
C	149	passive:	11	passive:	13	In 8 minutes 5 fish left, eventually all food eaten
		active:	5	active:	3	
D	152	passive:	9	passive:	0	In 5 minutes all food eaten
		active:	7	active:	16	

Tank	Week nr. 50	Date: 12-12-2008		Time: 09:30		Remarks
	Food dose (g)	before feeding		during feeding		
A	144	passive:	16	passive:	1	Fast response
		active:	0	active:	15	
B	146	passive:	14	passive:	1	Fast response
		active:	2	active:	15	
C	145	passive:	15	passive:	1	Respond slower, longer active
		active:	1	active:	15	
D	144	passive:	10	passive:	2	Slowest group, in 8 min. all food eaten
		active:	6	active:	14	

Tank	Week nr. 51	Date: 15-12-2008		Time: 09:30		Remarks
	Food dose (g)	before feeding		during feeding		
A	166	passive:	15	passive:	1	
		active:	1	active:	15	
B	171	passive:	10	passive:	0	
		active:	6	active:	16	
C	166	passive:	12	passive:	1	Later on all active
		active:	4	active:	15	
D	169	passive:	7	passive:	1	Later on all active
		active:	9	active:	15	

Tank	Week nr. 51	Date: 16-12-2008		Time: 09:30		Remarks
	Food dose (g)	before feeding		during feeding		
A	142	passive:	10	passive:	1	
		active:	6	active:	15	
B	142	passive:	11	passive:	1	
		active:	5	active:	15	
C	144	passive:	10	passive:	1	The smallest not active
		active:	6	active:	15	
D	141	passive:	10	passive:	1	
		active:	6	active:	15	

Tank	Week nr. 51	Date: 17-12-2008		Time: 09:30		Remarks
	Food dose (g)	before feeding		during feeding		
A	155	passive:	16	passive:	1	Fast response All food taken directly
		active:	0	active:	15	
B	153	passive:	13	passive:	2	2 eggs, fast response All food taken directly
		active:	3	active:	14	
C	155	passive:	8	passive:	0	Fast response All food taken directly
		active:	8	active:	16	
D	154	passive:	16	passive:	0	Fast response All food taken directly
		active:	0	active:	16	

Tank	Week nr. 51	Date: 18-12-2008		Time: 09:30		Remarks
	Food dose (g)	before feeding		during feeding		
A	155	passive:		passive:		No appetite. 11:50 some food left
		active:		active:		
B	155	passive:		passive:		Appetite in 3 min all food taken
		active:		active:		
C	154	passive:		passive:		2 eggs, fast response Higher activity than others
		active:		active:		
D	156	passive:		passive:		No appetite. 11:50 some food left
		active:		active:		

Tank	Week nr. 51	Date: 19-12-2008		Time: 09:30		Remarks
	Food dose (g)	before feeding		during feeding		
A	143	passive:	0	passive:	0	All active no appetite
		active:	16	active:	16	
B	145	passive:	2	passive:	1	In 10 min. all food taken
		active:	14	active:	15	
C	145	passive:	2	passive:	0	All food eaten directly
		active:	14	active:	16	
D	143	passive:	0	passive:	1	In 10 min. all food taken
		active:	16	active:	15	

# Justification

Rapport C105/09  
Project Number: 4301300601

The scientific quality of this report has been peer reviewed by the a colleague scientist and the head of the department of Wageningen IMARES.

Approved: Ir. PhD S. M. Bierman  
Quantitative biologist

Signature:



Date:

9 November 2009

Approved: Dr. Ir. T. P. Bult  
Head of Department Fisheries

Signature:



Date:

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