

The effect of electric pulse stimulation to juvenile cod and cod of commercial landing size

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Summary

Juvenile (0.12-0.16 m length) and large cod of 0.5 m length were exposed to simulated pulse stimuli representing commercially applied pulse systems with varying amplitude, frequency and pulse width at fixed positions in close range of a conductor. The juvenile fish were exposed with the highest possible amplitude in the nearest possible range of a conductor in a horizontal plane, along the conductor and perpendicular to the centre of a conductor. The field strength in the exposed position was in a range of 250-300 V/m. All juveniles (involving 12 trials and a total of 168 specimen) survived this harshest condition even after 4 exposures in a row. The post mortem research showed not a single vertebral injury or haemorrhage.

Larger cod of 0.5 m length were exposed to pulse patterns with a field strength of 40 to 100 V/m, depending on the pulse amplitude. Similar results as obtained in a pilot study of 2008 with similar length of cod with a series of 4 exposures were found in this present study using a single exposure.

Vertebral injuries occurred in 50-70 % of the fish under various pulse compositions (26 trials and a total of 262 specimen) and did not reduce when the shape of the burst was modulated according the "tooth-shape model". Injuries occurred in all three tested pulse concepts, although the Delmeco TX19 type had a lower score. This could have been related to the frequency (80 Hz), which also reduced the injuries when the HFK stimulus was tested at that frequency and probably also the shorter pulse duration and so the energy contents. We found a reduction of injuries with rising frequencies ≥ 80 Hz. At 180 Hz visible injuries reduced even to zero.

The post-mortem results did confirm the observations of injuries directly after the exposures and they show that the pulse stimuli can harm both the musculature and skeleton in cod, including fractures of the neural and hemal arch. In most cases the injuries are a combination of haemorrhages and vertebral ruptures. In a single case minor haemorrhages were found without bone fractures (Delmeco TX19, trial 19).

Vertebral injuries in cod of 0.5 m occurred when using the Delmeco TX68 stimulus in a field strength range of 40 to 100 V/m with 40 V/m as threshold value where injuries reduced from 60 to 30 % measured in the closest possible range from a conductor. The exposures with the Delmeco TX68 under nominal amplitude setting (50 V) were an overdose of 54 %. Under this condition a field strength of 91 V/m was measured in the laboratory in the exposed reference position of the fish, while under real fishing practice this rating was 59 V/m. However, the field strength measured under fishing condition is still above the threshold field strength level (38 V/m) where injuries reduced to 30 % and the fished field strength (59 V/m) is well inside the range (Figure 14) where 60 % of the fish were injured in the laboratory (Appendix B, Table 4, trial 7). Field strength measurements on the HFK pulse concept showed that the conditions measured during the experiments were similar to the field strength measured with the gear on the seabed for both juvenile and larger cod. These observations showed the laboratory experiments were conducted in a relevant range of field strengths and that injuries observed are likely to occur under full-scale practice.

As the fish became shortly paralysed in most of the cases after the exposures or showed a disorientated behaviour it is likely that most of the fish will be caught when they move through the net in close proximity of the conductors. This injured fish is of lower market value than fish that is landed without injuries. However, next to the quality issue there is also an ethical motive to find solutions to reduce such injuries. Reduced injuries can be achieved by increasing pulse frequency. The research showed that electric pulse systems did not reach their optimum in terms of electrode and conductor efficiency and this research also showed that there is room for further technical improvements.

It can be considered to conduct full-scale electric field measurements on the TX19 as with the Delmeco TX19 pulse concept the effects to large cod were lower than with other pulse concepts and to include this vessel in future monitoring field tests.

The field strength measurements at sea showed that reference measurements with the gear hanging vertically aside of the ship do not produce the same figures as in the actual fished condition with the gear on the sea bed. Reasons are the differing geometry of the electrodes in the hanging condition, and the different electric field around the conductors due to sea bed presence and conductivity. Field strength measurements as a standard inspection routine with the vessel in the harbour are not accurate and could contain an error of 10%.

Introduction

Work on reducing the drag and impact of traditional beam trawls on the ecosystem started in the sixties with adding an experimental electric stimulus to a shrimp trawl (Boonstra and de Groot, 1970). In those years pulse stimulation was also developed to replace tickler chains in flatfish beam trawling, in particular sole (*Solea vulgaris* L.) and plaice (*Pleuronectes platessa* L.). Many studies were done since in the 1970s and 1980s, also in other European countries with similar beam trawls, but in spite of promising results commercial uptake was lacking (De Groot and Boonstra, 1970, 1974; Van den Broucke, 1973; Stewart, 1975, 1978; Horn, 1976; Horton, 1984; Agricola, 1985; Van Marlen et al., 1997). Motives for using electrical stimuli other than the reduction of the impact to the seabed are the reduction of gear drag and so the use of fossil fuel and CO₂ emissions and the reduction of bycatch and discarding. 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; 2001). 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. Following the ICES Advice on Pulse Trawling on flatfish of 2006 research was carried out by IMARES, the Netherlands on cod (*Gadus morhua* L.) (de Haan et al., 2009a) and cat sharks (*Scyliorhinus canicula* L.) (de Haan et al. 2009b) and a range of benthic species (ragworm (*Nereis virens* L.), common prawn (*Palaemon serratus* L.), subtruncate surf clam (*Spisula subtruncata* L.), European green crab (*Carcinus maenas* L.), common starfish (*Asterias rubens* L.), and Atlantic razor clam (*Ensis directus* L.) under pulse stimulation of the Verburg-Holland electric pulse system (Van Marlen et al., 2009).

For cod the experiments were conducted at and in cooperation with scientists of the research station Austevoll of the Institute of Marine Research Bergen, Norway.

The experiments on dogfish and benthic species, executed at the Imares laboratories, showed hardly an effect, the strongest effects were measured in the experiments on cod. The fish exposed with nominal pulse settings in a very short range were injured and suffered haemorrhages close to the vertebral column.

In February 2010 the outcome of the three studies was discussed at the ICES WKPULSE workshop (ICES, 2010) with international experts and the group advocated continued research with the following terms of reference:

- a. To review current technical developments on electrical fishing (with main focus on marine systems).
- b. To review studies on the relationship of pulse characteristics (power, voltage, pulse shape) and thresholds in effects (mortality, injury, behavioural changes).
- c. To improve knowledge about the effects of Electrical Fishing on the marine environment (reduction of bycatch, impact on bottom habitat, impact on marine fauna, energy saving and climate related issues).
- d. To evaluate the effect of a wide introduction of electric fishing, with respect to the economic impact, the ecosystem impact, the energy consumption and the population dynamics of selected species.

- e. To consider whether limits can be set on these characteristics to avoid unwanted effects (e.g. unwanted and uncontrolled growth on catch efficiency, unwanted ecosystem effects) once such systems are allowed and used at wider scale.

WKPULSE recommended to collect more information on the effect on cod before the pulse trawl can be allowed, and among other items to:

1. Monitor the current (by)catches with the latest version of the system (TX68) and compare these with a conventional beam trawler
2. Sample cod from current (by)catches with the latest version of the system (TX68) and compare the occurrence of spinal damage with those from a conventional beam trawler
3. Investigate the effect on fish not necessarily caught in the trawl after being subjected to the pulse system and determine whether this is a source of unaccounted mortality.

After WKPULSE an ICES study group SGELECTRA was established and met in May 2011 in Reykjavik with specialists of European institutions. The findings and interim results of current pulse related projects and other relevant publication were discussed to develop and test the present guidelines and to develop other recommendations. The findings were reported and discussed at the ICES-FAO WGFTFB meeting (ICES, 2011). SGELECTRA recommended among other to:

1. conduct in situ measurements on the field strength of the two existing Dutch pulse trawling systems as a reference to the experiments of 2010 on cod in Norway,
2. review the results of the comparative fishing experiments in the Netherlands in May 2011,
3. study the effects of with pulses identified as less harmful in flatfish systems on target species (e.g. sole),
4. continue studying the effect of wider introduction of pulse trawling on target and non-targets species

This report addresses recommendations reported after the first pilot study (de Haan et al., 2009), recommendation 1 from SGELECTRA and 2, and 3 from WKPULSE.

1.1 Pulse system description

1.1.1 *New developments*

At present there are two Dutch manufacturers, who developed electric pulse system prototypes for the Dutch beam trawl fleet fishing for flatfish. The company Verburg-Holland Ltd. was taken over in 2010 by another Dutch company Delmeco Group Ltd., Goes, Netherlands and the present pulse system developed to a new concept with reduced voltage across the conductors (60 to 50 V^{0 to peak}) and increased electrode distance (from 0.325 to 0.425 m), reducing the number of electrodes and so the overall power ratings. All voltage ratings in this report refer to the peak voltage measured over the positive part of the pulse, described as zero to peak (Figure 3).

The Delmeco Group supports two basic pulse concepts (Delmeco TX68 and Delmeco TX19). The Delmeco UK153 system listed in Table 1 is not commercially applied anymore. In 2009 a second Dutch manufacturer, HFK-engineering, Baarn, Netherlands, developed a commercial electric pulse system. The pulse system is applied on a new type of beamtrawl, the so-called "SumWing" trawl (Figure 2). This new design has less contact with the seabed. The cylindrical beam is replaced by a wing-shaped foil with a runner at the centre forcing it towards the seabed. Trawl shoes are not used in this gear. The wing glides above the seabed and has a lower impact on the seabed environment and reduces fuel consumption by some 10% when using a traditional beamtrawl net behind it with tickler chains (van Marlen et al., 2009). The implementation of the pulse system to the "SumWing" trawl is called "PulseWing" and has a larger

potential (50%) for reduction of gear drag, bottom impact and fuel consumption (van Marlen et al., 2011).

1.1.2 Common characteristics

Electric pulse systems are provided with electric power from the vessel by an electric cable system additional to the warps with which the gear is towed. The electric power supplied is restricted in a current derogation to a maximum of 1.25 kW/m beam length and an electrode voltage of 15 V^{rms}. The main controls are on-board the ship and this control system communicates through the electric cable with the pulse generator equipment placed on the trawl.

A monitor is often used of the pulse system displaying the momentary condition of the electric system and power ratings and in some cases the electric parameter settings can be adapted to the fishing condition or seasonal conditions such as temperature and salinity affecting sea water conductivity or seasonal flatfish response. The basic nominal design characteristics of the pulse systems are listed in Table 1. In most cases the discharge unit of the pulse system fitted on the fishing gear is built up in modules and are shaped to the profile of the beam or wing to contribute to drag reduction. These modules are sealed and filled with resin and not accessible. Each module is connected to an electrode with isolated and conducting elements.

At present the smaller beam trawlers with 220 kW main engine power license are rigged with beams of 4 m length with 10-11 electrodes (Figure 18 , TH10 field strength measurements), while gears of larger 1470 kW licenced vessels with 12 m beam trawl length contain 25-28 electrodes of similar construction (Figure 19, OD 17 field strength measurements). All systems are designed for a towing speed of 5 knots.

Table 1 Overview of main pulse parameters of the three simulated systems with also the ratings of a smaller 220 kW licenced vessel.

Pulse concept	Power Single gear (kW)	Electrode Voltage (V ^{0 to peak})	Pulse Freq. (Hz)	Pulse width (µs)	Electrode		
					Nr	Distance (m)	Conductor (nr) (l x d (mm))
Delmeco UK153	8.5	60	40	220*	32	0.325	6 (180x26)
Delmeco TX68	5.5	50	40	220*	25	0.425	6 (180x26)
Delmeco TX19	5.5	50	80	130	25	0.425	6 (180x26)
Delmeco TH10	2.1	50	40	220*	10	0.425	6 (180x41)
HFK TX36	7	45	45	170/50	28	0.415	2 (125x27) + 10 (125x33)

*The pulse duration refers to the a single pulse period

All voltage ratings in this report refer to the peak voltage measured over the positive part of the pulse, described as zero to peak

1.1.3 *Delmeco pulse concepts*

The electrode configuration for the Delmeco-equipped fleet is similar, but the electrode distance of 0.325 m is no longer applied in commercial fishing and increased to 0.425 m. The TX19 pulse concept is basically different (Figure 3) and uses a bipolar pulse with minimum delay between the positive and the negative part and a higher pulse frequency. At present this concept is only applied on-board TX19. On-board the TX68 and all other Delmeco supplied vessels the TX68 pulse concept is applied with the increased electrode distance.

1.1.4 *HFK pulse concept*

The hardware of the HFK pulse concept has a different energy concept and range. The energy of the "Delmeco" system is directly coupled to the discharge circuitry of the electrodes, while the "PulseWing" concept is limited in size by the physical space inside the wing type of beam and comprises a relatively small energy buffer capacitor, which is charged from the main ship-based circuit and discharged over the electrode system. The size of the capacitor supplies a physical limitation in terms of peak voltage across the conductors and the discharged energy.

1.2 Objectives for this study

The first pilot study on the effects of electric pulse stimulation on larger cod carried out in 2008 (de Haan et al., 2009) was based on a single nominal setting of the Verburg-Holland UK153 pulse system (Table 1) with the intention to determine the range of pulse characteristics with which injuries to the fish occurred.

Three distance ranges were defined and tested:

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 0.2-0.3 m above the centre of a conductor pair;
3. A "near field" range with the fish exposed at 0.1 m from the conductor element.

The dissection and radiological observation showed that only fish in the "near field" group had vertebral bone fractures. In total 45 % (9 of 20) of the "near field" group had injuries, while injuries in fish of other groups were not found (de Haan et al., 2009).

This study was designed to obtain a more detailed view on the effects of the pulse characteristics and to investigate if a specific pulse parameter has a key role in the effects. Lower pulse amplitudes with longer pulse width and higher frequency could induce different effects than pulses with higher amplitudes, shorter pulse width and lower frequency. In this way the effects can be related to a specific pulse parameter and its threshold value. Another aim of major importance is that this research also carried out on the electrically exposed fish that would normally escape through the cod-end meshes with unknown longer term effects. This was already recommended after the first experiment in 2008 (de Haan et al., 2009). These two objectives follow the advices of the ICES WKPULSE (ICES, 2010).

1.2.1 *Animal welfare approval*

The final research protocol was sent to the Norwegian research animal board in September 2010 to make sure the research could be conducted under the existing Norwegian animal welfare legislation. The approval of the research on 17 November 2010 ensured the research was feasible under the proposed terms and after approval the research was prepared and equipment mobilised to the IMR laboratory in Austevoll, Norway.

2 Materials and Methods

2.1 Delmeco pulse simulation

The simulated pulse had a bipolar shape (thus with a positive and negative part) in all three cases, but with different delay times between the positive and negative sequences of the pulse (Figure 3). The Delmeco TX68 pulse concept of which the nominal settings were tested and reported in 2008 (de Haan et al., 2009) was repeated in this study as a basic reference for other pulse compositions. The equipment consisted of a pulse generator based on a microprocessor and a pair of high power Insulated-Gate Bipolar Transistors, IGBT's (Figure 5). With a controller the main parameters of the stimulus, amplitude, pulse shape, pulse width etc. were programmed with a limited range of frequency and pulse width settings.

The electrode distance for all pulse concepts was set on 0.325 m, earlier applied in the first pilot study, and used as reference to exclude effects of distance. In a single case the effects of the nominal Delmeco commercially applied electrode distance (0.425 m) and an increased conductor diameter (41 mm) was tested (materials provided by Delmeco). It was thought that the electric field density would proportionally decrease with increasing conductor diameter and so reducing the effects to cod in close range. The Delmeco stimuli (TX68 and TX19) were simulated using the projection equipment (Figure 10) used in the first pilot study (de Haan et al., 2009), including the twin set of conductors of 180 mm length and 26 mm diameter.

The pre-programmed simulated TX68 pulse was extended by the manufacturer with a second, the TX19 pulse menu. The setting of the Delmeco frequency range was limited to 30-57 Hz and the minimum pulse width to 150 μ s.

2.2 HFK pulse simulation

As the set-up of the HFK simulation was based on a waveform generator and less limited in range settings, the effects of frequency and pulse width were tested with this pulse concept. The HFK pulse simulation consisted of an electronic switching device (H-bridge MOS-FET), identical to the commercial applied components and a provisional controllable DC power source according the schematic overview of Figure 5. The electronic switch was gated using an IMARES arbitrary waveform generator (type HP 22180). Arbitrary waveforms had to be used to generate positive and negative parts of the pulse and were created in Excel and exported using the serial RS232 connection gate of the instrument for each test series at a specific frequency and pulse width combination.

The simulated HFK electrodes were a commercial copy, provided by the manufacturer, consisting of a single pair of conductors of 125 mm long and a diameter of 27 mm. The buffering capacitor had to be enlarged to 22000 μ F, which is outside the commercially applied scale to support the energy required for the 1 s burst cycle. This buffer capacity normally holds enough energy to support a single pulse. The pulse amplitude could therefore not be kept constant over the complete burst cycle. At the 60 V pulse setting the actual amplitude was higher at the start (i.e. 66 V) and drops to 53 V at the end of the burst (Figure 6). The amplitude was set to produce the referred voltage at 50 % of the burst period. The second limitation caused by the provisional buffering was the pulse width setting (single period), which was limited to a maximum of 240 μ s.

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 (not swimming along with the gear) in 0.5 s. In order to compensate for this dynamic situation the stimulus consisted of a burst of pulses of 1 s, which is

equivalent to the length used in the first study of 2008 (de Haan et al., 2009). In a single case (Table 4, trial nr 20) the burst period was reduced to 0.5 s to determine the effect of the exposure period.

Under towing conditions the electric field strength at a stationary point drops and rises dynamically with the passage of the isolation parts of the electrode, as only the conductors generate the electric field, which is therefore non-uniform. In this way the exposure of a constant peak amplitude can be regarded as an overdose and would not produce a realistic figure. To investigate the dynamic effects of this reduced electric field strength condition the Delmeco burst amplitude was modulated in a single case (Table 4, trial nr 9) according the patterns of Figure 7. Measurements results of the simulated HFK stimulus are illustrated in Figure 8 and 9.

2.3 Positioning of the fish in the electric field

2.3.1 *Juvenile cod*

Juvenile cod of length between 0.12-0.16 m were exposed in two directions on the bottom of the tank, parallel to the centre line of the conductor and 90° rotated, perpendicular to the conductor. The fish were positioned using a meshed cylindrical cage (180 x 70 mm) of synthetic mesh material. The reference distance of the fish to the conductor (taken from underwater observations during the trials) was taken at the centre axis, 35 mm away from the conductor edge and 20-35 mm above the bottom (Figure 10). This was the height range the fish took during the trials. The second exposed condition was perpendicular to the centre of the conductor with the beak 20-30 mm from the edge of the conductor. The juvenile fish could freely swim inside the cage, and thus were not completely fixated.

2.3.2 *Large cod*

The tested length of marketable cod (MLS = 0.35 m) ranged between 0.44 and 0.55 m. The fish were accurately positioned lengthwise with its centre line 55 mm aside and 80 mm above the bottom according the illustration of Figure 11. The positioning materials and cage of synthetic mesh netting, used in 2008 were also used in this experiment. In the experiment of 2008 the cod could freely swim through the cage and exposures were timed at a chosen position of the fish relative to the conductor. In this experiment the large cod were positioned more accurately 55 mm aside and 80 mm above the bottom. The fish could freely swim, but were forced downward (using a plate of equivalent synthetic mesh material) in the given position once they reached a required position and orientation. Then the pulse was triggered.

2.4 Procedures

2.4.1 *General conditions*

Large cod were transferred from the outdoor net pen to the main storage tank in the laboratory and were conditioned a week prior to the first experiment. The small cod were transferred two months before the experiment and kept in a second similar storage tank. The husbandry was kept very stable in this pre-period and at the experiments, all fish stayed alive. They were fed by an automated system at fixed times daily.

2.4.2 *Video observations*

The behaviour of the fish during the exposures were filmed from above the tank in air using an IMARES bullet camera (bullet type 1/3 Sony HQX CCD 560 TVL pal) and a second equivalent positioned underwater in close range of the conductors. The video signals were recorded using the IMR recording system. All video images were stored on hard disc in MPEG 4 format.

2.4.3 Salinity and temperature

The sea water used in the tanks was pumped from the adjacent fjord. The conductivity and temperature of this water were monitored. The average values for water temperature and salinity during the experiments were respectively 7.5 °C and 34.8 ppt ± 0.1. The salinity and water temperature were measured at sea during the field strength measurements using a Hydrolab type DS5 CTD sonde, which was attached to the trawl not measured on the other side. Samples were logged every 10 s and covered the complete measurement session.

2.4.4 Test procedure

The fish were tested in groups of 20 individuals for the first series at the start and when significance was found in groups of 10 for each following pulse setting and orientation (Appendix B, Table 4). At the start of the experiments both length groups were exposed to the dose used in 2008, a series of 4 exposures (de Haan et al., 2009) and the effects compared to a test with a single exposure. As the effects did not reduce with a single exposure, this setting was maintained thereafter.

The pulse output parameters and field strength in the defined ranges from the conductors were measured shortly before or after the exposures. Samples of measurement results were stored as JPEG images on hard disc.

Length, and weight of individual fish were measured after the exposure once the fish was dead and frozen per group in the IMR store. After the experiments the fish were transferred to the Matre station of IMR where they were X-rayed and dissected.

2.5 Pulse measurement procedure

2.5.1 Pulse measurements in the laboratory

The pulse output parameters and field strength in the defined ranges from the conductors were measured shortly before the start of the exposures using a 200 MHz LeCroy WaveSurfer 24XS oscilloscope with two types of differential probes, a high voltage type ADP 305 (SN5069) and a AP031 70 V probe for field strength measurements and a CWT Rogowski 60B current probe (0.5 mV/A) to measure the electrode current. All voltage measurements in this report are referred to the zero to peak voltage of the positive amplitude of the bipolar pulse. Samples of measurement results were stored as JPEG images on hard disc. Electric field strength was measured in the plane of the electrodes with a probe of fixed spacing of 25 mm over the tips with grid units of 32.5 mm. Samples were taken along the 35 and 55 mm axis aside of -and parallel to- the conductor, representing the axis of the bodies of juvenile and large cod (Figures 10 and 11).

2.5.2 Pulse measurements at sea

Pulse system parameters were measured on two representative Delmeco and HFK commercial pulse fishing systems. Measurements were done of field strength in three fixed positions between a conductor pair as well as the voltage across the conductors at the front and back end of the electrode pair on which field strength was measured. The Delmeco pulse parameters were measured on-board TH10 "Dirkje", and the HFK pulse parameters were taken on-board OD17 "Buis".

The field strength probes were positioned in a rigid stainless steel frame (Figure 18, 19 and 20), which was clamped on the electrodes. The field strength readings were taken at the bottom level between a conductor pair as reference in the position the 0.5 m cod were exposed in the IMR laboratory and as reference to earlier measurements taken in the laboratories of Verburg-Holland Ltd. For the bottom field strength reference the probes were positioned on the centre level of a conductor pair with the outer

probes 55 mm aside the centre of both conductors and the centre probe on the centre axis between both conductors. The average 55 mm outcome was taken as reference for the 0.5 m cod exposure and this probe was lifted 80 mm relative to the bottom level of the conductor as reference position of the exposed large cod in the IMR laboratory. To assure the measurements were taken with the fishing gear at the seabed and to observe any artefact on the position of the probes relative to the conductors a low light underwater video camera (DeepSea 1060) was added to the system. Measurements were done at sea in the Dutch coastal zone with the fishing gear hanging vertically just under the water surface with the gear moving very slowly on the bottom, while the ship drifted slowly with the tide 0.1-0.3 knots.

The measurement equipment consisted of a 200 MHz LeCroy WaveSurfer 24XS oscilloscope with 3 differential probes, a high voltage type LeCroy ADP 305 to measure the pulse amplitude across the electrodes at the front and back of the electrodes. Field strength was measured with a differential TA043 (2x) and a LeCroy AP031 70V probe (1X). All probes were battery powered during the measurements.

2.6 Physical effects of the electric stimulus, post mortem research

The post-mortem study on effects was conducted at the IMR research station Matre, Norway. Each fish was labeled and photographed to ensure a unique identification in the analysis. In the dissection stage the presence and location of haemorrhages were analysed. The vertebral column of the fish were carefully dissected, visually examined for pathology, and finally X-rayed and then evaluated for bone fractures on the radiographs. The radiological analysis involved the detection of vertebral, hemal and neural fractures. Dissected vertebral columns were radiographed by using a portable X-ray apparatus (HI-Ray 100, Eickenmeyer Medizintechnik für Tierärzte e.K., Tuttlingen, Germany) and 30x40 cm film (FUJIFILM IX 100, FUJIFILM Corp., Tokyo, Japan). The film was exposed once for 50 mAs and 68 kV, and developed using a manual developer (Cofar Cemat C56D, Arcore (MI), Italy) with Kodak Professional manual fixer and developer (KODAK S.A., Paris, France). The pictures were digitalised by scanning (Epson Expression 10000 XL, Seiko Epson Corp., Nagano-Ken, Japan). The analysis results were sorted per group and added to the overview of Table 4 Appendix B.

3 Results

3.1 Overall

The experiments were executed in the period of 28 November to 14 December 2010 and are based on 38 trials involving a total of 441 specimen, involving 26 trials with commercial landing size cod (272 specimen) of and 12 trials with juvenile cod (total number 169).

The experiments started with trials with the Delmeco TX68 pulse concept to find the reference condition determined under nominal pulse conditions in 2008. In the overview of Table 4 (Appendix B) all trials and the pulse specifications are listed in order of execution with the observed results including the post-mortem analysis .

The results show juvenile cod all survived the tests of all three pulse concepts with the highest conductor voltage available (60 V) at pulse frequencies of 30, 40, 45, 80 and 180 Hz. Large cod were injured with all three pulse concepts under conditions varying between a conductor voltage range of 30 to 60 V. Injuries did reduce by increasing the pulse frequency. At 180 Hz injuries reduced to zero.

With the Delmeco TX68 pulse concept first the threshold value for the voltage across the conductors was defined and found at 20 V. Continued experiments with other pulse variables and settings were executed with a pulse amplitude slightly above this setting to become more sensitive to influences of other pulse variables, like frequency and pulse width.

3.2 Direct responses to the exposures

3.2.1 Juvenile fish

The fish produced a very strong response on the Delmeco TX68 pulse concept during the trials with a series of four exposures with very short intervals (Table 4, trial 2). The fish fell into coma for a short period, but all recovered. The fish were kept alive over the weekend and responded normally to food. In the continued trials the muscle contraction was medium to low and the fish return to normal behaviour directly after the exposure. The post mortem research showed not a single vertebral injury or haemorrhage.

3.2.2 Large fish

Results obtained in the pilot study of 2008 with similar length of cod with a series of 4 exposures were confirmed in this present study with a single exposure. Vertebral injuries occurred with 50-70 % of the fish under various pulse compositions (26 trials and a total of 262 specimen) and did not reduce when the shape of the burst was modulated according the "tooth-shape" model (Figure 7). The threshold value for the amplitude of the conductor voltage was found at to 20 V. At this value injuries reduced from 60 to 30 %. Injuries occurred in all three tested pulse concepts, although the Delmeco TX19 type had a lower score. The large fish produced a medium to strong contraction during the exposure and produced a strong startle response after similar to the responses observed in the experiments of 2008 (de Haan et al., 2009) with controlled and disorientated behaviour and accelerated occasionally from the cage into the tank. As in 2008, vertebral injuries caused black circular patterns on the tail (Figure 12). These images disappeared after the fish died and would probably have disappeared when exposed fish is brought on deck under normal fishing practice. During the trials the sound of vertebral ruptures were recognised at the side of the tank and were recorded underwater on the final days of the experiment to determine the time lap between the exposure and the moment of vertebral rupture. It appeared that multiple ruptures did occur and also they were detected after the pulse burst extinguished.

3.3 Field strength during the experiments

3.3.1 Juvenile cod

The electric field strength was measured during the experiments as reference to the observed effects. Figure 13 and 16 show the field strength measured along the 35 mm axis (see also Figure 10) for the juvenile cod with respectively the Delmeco TX68 and HFK stimulus. On tests with juvenile cod only the maximum electrode voltage (60 V) was applied. The field strength measured across the centre of the conductor was the highest and varied between 255 and 311 V/m, depending on the vertical level. Figure 17 shows the field strength measured perpendicular to the centre of the conductor.

3.3.2 Large cod

Figure 14 and 15 show the field strength measured 80 mm above the bottom along the 55 mm axis (Figure 11) for the large cod with respectively the Delmeco TX68 and HFK stimulus. The electric field strength was measured at a conductor voltage range of 20-60 V amplitude, in steps of 32.5 mm along the conductor in a range of 38-103 V/m. The maximum HFK field strength was 17 V/m lower than the Delmeco TX68 result.

3.4 Measurements on full scale pulse systems

3.4.1 Delmeco TX68 pulse system

The measurements took place on 02 November 2011 and were carried out in the coastal Dutch zone just outside Scheveningen harbour at a water depth of 15 m. The Delmeco pulse system on-board TH10 comprised of a TX68 type of pulse characteristic with a pulse peak amplitude setting of 54 V (215/255 *65V) and an electrode system with six conductors (Figure 18) with an electrode distance of 0.425 m. The measurement frame was clamped over the most frontal conductor pair of the centre electrodes 5 and 6 marked as "field 5" on the ship's monitor. The voltage across the conductor was measured on this conductor pair and the conductor pair at the back end of the electrodes. Total consumed power displayed by the system monitor was 2.1 kW and an electrode current of 250 A. During the measurements the adjacent fields were switched off, to avoid triggering of the oscilloscope on adjacent fields. On the first deployment the cable connection at the back end conductors broke (the electrode distance was not limited). On the second measurement with the gear at the bottom, the speed was momentarily increased which stretched the electrodes giving a more symmetrical result (probe A and C are at similar distance from the conductor and the outcome should be equal). The average value of the salinity measured at the seabed was 33.21 ppt and at the surface 32.96 ppt. The water temperature readings were respectively 13.20 against 13.16 °C. The field strength was measured according to the overview of Table 2.

Table 2 Results of Delmeco pulse parameters measured on-board OD17 taken on the centre electrodes ("field 5"). Numbers in bold face black were selected as in-situ representative value. Blue numbers show the asymmetry with the gear vertically deployed.

Delmeco TH10						
Fishing gear position	Position Probes	Conductor 1 (V)	Conductor 6 (V)	A probe (V/m)	B probe (V/m)	C probe (V/m)
Surface vertical	Bottom level	47.7	37.3	154	54	326
Seabed	Bottom level	50	Error	158	78	155
Surface vertical	Bottom level A +80 mm	48	47	51	23	124
Seabed	Bottom level A +80 mm	51.5	51.2	59	78	155

3.4.2 HFK pulse system measurements

On 4 November 2011 the pulse characteristics and field strength of the HFK pulse system were measured on-board OD17 "Buis". The start-up of the measurement sessions was delayed due to a technical failure in the communication wiring in the main cable between the deck unit and the pulse generator at the electrode end. With the fishing gear deployed vertically aside of the ship in the harbour it appeared the system produced a pulse amplitude of 24 V with a frequency of 77 Hz. When communication fails the system software automatically addresses the last setting applied. Apparently in this case the pulse system was operating at 80 Hz. The electrode voltage measured across the first conductor pair was 24 V, which is far below the 60 V nominal open clamp supply voltage of the discharge modules. This lower result is possibly attributed to the increased number of conductors (12) instead of the 6 of the previous design, operational in the first half of 2011 on the HFK "PulseWing" fleet (Figure 20). Secondly the length of isolating pieces was shortened in this new electrode arrangement in such a way that the clamps of the measurement frame did not fit without influence to the electric field of the adjacent conductor pair, which had to be isolated by tape.

After repair the first representative measurements were taken at 16:30 h local time with the gear vertically deployed at sea just below the water surface. Under this condition the frontal pulse amplitude was 32.6 V and at the back of the electrodes 30.6 V (effective power 7.3 kW over 28 electrodes/12 conductors). With the gear deployed on the seabed the electrode voltages were respectively 38.5 and 36.6 V, and the effective electric power 6.6 kW (Table 3). The salinity at the surface condition was 31.95 ppt, water temperature was 13.2°C. Although the salinity measured at the seabed position was significantly higher (33.2 ppt) the voltage across the conductor did not reduce accordingly, but rose instead. Apparently the higher electrode voltages were attributed to lower conductance of the part of the field through the seabed. The water temperature at the bottom was in the same range, at 13.25°C. The distances measured between the tips of the probes A, B and C were respectively 25.3, 25.8, 25.2 mm and were polished prior to the measurements (to be used also for the Delmeco measurements).

After evaluation the measurements were disqualified for two reasons:

- 1) Two of the 12 conductors had to be isolated to attach the frame with the field strength probes and so the discharge characteristic expressed in the voltage across the conductors at the begin and end of the electrodes did not represent a full reference value for "PulseWing" fishing condition;
- 2) The electrodes on which the measurements took place consisted of conductors of 33 mm, while the fishing trials on-board TX36 conducted in May 2011 (van Marlen et al., 2011) were conducted with 27 mm conductors. So the results could also not be used as reference for the TX36 fishing condition.

After the consulting the ship-owner Jaczon, of Scheveningen, the Netherlands the measurements were repeated on 11 November 2011 under similar conditions. The measurements were carried out on the same gear at the starboard side, and with the inner electrodes nr 4 and 5 both rigged with 12 conductors of 27 mm (Figure 20), which is equivalent to the rigging on-board TX36 during the fishing trials together with GO4 and TX68 in the week of 5 May 2011. The gear of OD17 appeared to be a mixture of types of previous and latest design, on which conductors are clamped on an inner steel reinforcement. The measured electrode layout was rigged with 2 frontal conductors of 27 mm and 10 adjacent of 33 mm, all 125 mm long.

To minimise the influence of the metal frame the first conductor pair with the frontal 1 m connection lead was taken out and reversely connected (Figure 20). Since the measurements of 4 November the two outer modules and electrodes of the "PulseWing" gear were taken out, which brings the total to 26 electrodes. The overview of results is listed in Table 4. At the surface the supplied electric power of the pulse system was 6.8 kW and at the bottom significantly lower 6.2 kW (26 modules instead of the standard 28).

Table 3 Overview of HFK field strength measurements conducted on 11 November on-board OD17 with the reference results in bold for large cod (row 3) and juvenile cod (row 6). Numbers in bold face black were selected as in-situ representative value. Blue numbers show the asymmetry with the gear vertically deployed.

HFK OD17						
Fishing gear position	Position Probes	Conductor 1 (V)	Conductor 12 (V)	A probe (V/m)	B probe (V/m)	C probe (V/m)
Surface vertical	Bottom level	36.3	29.0			
Seabed	Bottom level	42.6	37.3	147	26	138
Seabed	Bottom level. A-probe lifted +80 mm	46.4	43.0	69	31	157
Surface	Bottom level A +80 mm	36.0	29.0	52	26	130
Surface	Bottom level A +35/35 mm	36.3	29.5	138	26	130
Seabed	Bottom level A +35/35 mm	46.4		208	28	152

The results of measurements applied on the latest HFK electrode (with 10 conductors of 33 mm diameter) and the electrode with 12 conductors of 27 mm diameter showed that the previous system resulted in a significantly higher electrode voltage, also with less distortion in the rising edges of the pulse (Figure 21 and 22).

3.5 Post mortem research

The post-mortem analysis results are in line with the estimates of injured fish observed directly after the exposures (Appendix B, Table 4 remarks). These estimates were based on indications of vertebral injuries through tail colouring as well as sound recordings on the final days of the experiments. The analysis showed that the muscular haemorrhages were most often associated with some kind of bone fracture, so we believe that this shows that the pulse stimulus can harm both the musculature and skeleton in cod. The fact that the two events, bone fracture and muscular haemorrhages, were located at the same spot, often in the anterior part of the tail region, indicated that the stimulus caused a strong musculature contraction in this body part. When strong enough this contraction may have caused the tissue damage in the musculature and skeleton. The injuries were sorted in three types and combinations of fractures, neural, hemal and vertebral, as well as haemorrhages. The first type of injuries were detected using radiological equipment, while haemorrhages were analysed through dissection. Examples are illustrated in Figure 23.

4 Discussion

4.1 Orientation of the fish in the electric field.

At WKPULSE references, including some Russian studies, were presented and discussed on the responses of fish as a function of their orientation in electric fields. These studies showed that the greatest effect is observed when the electric field lines are perpendicular to the longitudinal axis of the fish body. At longitudinal orientation of fish to the electric field lines the irritation threshold levels are lowest and the nerve crosses the maximum number of equipotential planes and so gaining the greatest potential difference on its ends (Petrov, 1935).

The species selective characteristics were well explained by McK. Bary, 1956 showing the species-specific characteristics of body shape and peculiar excitable structures of length and spatial orientation.

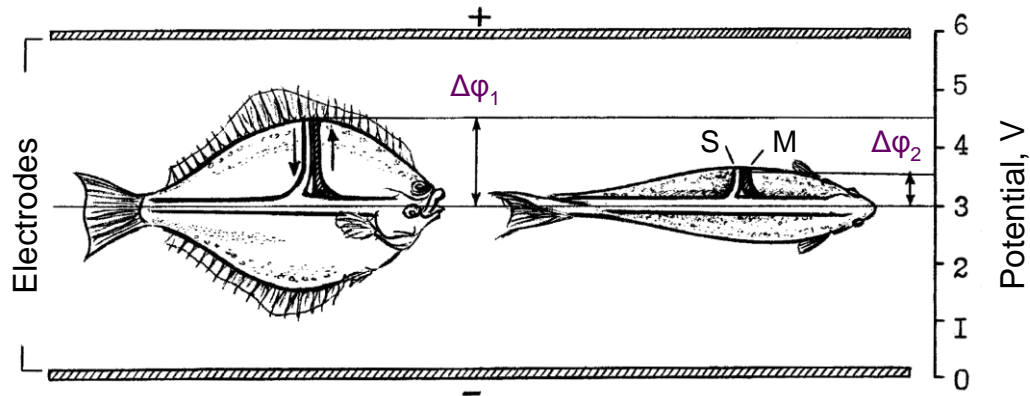


Figure 1 Potential difference on the segmental nerves in the cross electric field for the fishes differing in their body shape (McK. Bary, 1956). $\Delta\phi_1$, $\Delta\phi_2$ – potential difference. S – sensory fibers, M – motor fibers of the segmental nerves

These references point out that the orientation of the fish and their species specific shape are important variables in trials investigating the effects of electric fields. For this reason we included exposures with the fish perpendicular to the conductor.

The study of Sharber and Carothers, 1988 on trout (*Salmo gairdneri*) showed that vertebral damage is a common injury related to electro-fishing and occurred in 50 % of the 209 fish examined. Of the tested shapes (quarter-sine waves, exponential pulses, square waves) pulses based on a 60 Hz pulsed output of a 260 DC source) of quarter sine type had a slightly higher proportion than the other types, but vertebral injuries occurred with all types of pulse shapes and a relation with pulse shape could not be determined.

4.2 Post-mortem observations.

It is not possible to be sure about the significance of the different types of injuries found in the post-mortem research to the survival chances of these fish without following the fish after exposure to the pulse stimuli and investigating the long-term effects of neural, hemal and vertebral fractures.

4.3 Effect of burst period, towing speed and fish behaviour.

Reduced injuries were found on a trial with reduced burst period (Table 4, trial 20 versus trial 21). Theoretically an equivalent result can be expected by increasing towing speed by a factor two (increasing the towing speed will be practically limited and is related to the stability and performance of the wing of gliding in a fixed height above the bottom) or reducing conductor length by 50 %, which is more feasible. Another approach would be to deter the cod from the bottom at the level of the beam or wing. This could be achieved by adding a conductor pair of very short length more to the front in the area where the electrodes climb upwards to the wing. On the HFK system this would be achieved by using the connection joint at the front of the electrodes (Figure 20) and to remove the isolation. Further research on deterring cod to a higher level above the bottom will be required.

4.4 Relevance of the laboratory results

4.4.1 *The Delmeco pulse concept*

The results of field strength measurements during the experiments and at full-scale at sea showed the Delmeco TX68 exposures in the laboratory are an over-estimate of 54 %. With the Delmeco TX68 pulse concept with 50 V conductor voltage a field strength of 91 V/m was measured in the laboratory in the exposed reference position of the fish, while under real fishing practice this value was 59 V/m. Under this particular lab setting this can be considered as an overdose, however, the threshold field strength level where injuries reduced to 30 % was at 38 V/m, which shows the fished field strength (59 V/m) is well inside the range (Figure 14) where injuries did occur in the laboratory (Table 4, trial 7). Figure 24 illustrates the field strength curve measured in the laboratory in the exposed position of large cod and the down scale of the curve with the single point outcome at the centre of the conductor (grid position "0").

4.4.2 *Delmeco TX19 concept*

Full-scale reference measurements on fishing conditions were not executed in this research and there was also no information found on the landings of this vessel in relation to injured cod. It will be recommended to conduct full-scale field measurements and to include this vessel in future field test as the laboratory results show the effects to large cod were lower than with other pulse concepts.

4.4.3 *The HFK "PulseWing" concept*

On-board OD17 a field strength of 69 V/m was measured in the exposure reference position of large cod with the gear at the seabed. During the experiments in the laboratory this reference value was 57 V/m. When the difference of electrode voltages (46 V measured at sea, lab reference 40 V) is taken into account both conditions will equalise and we conclude that field strength conditions in the laboratory were realistic for this pulse concept.

Figure 25 illustrates the field strength curve measured in the laboratory and the down scale of the curve with the single point outcome at the centre of the conductor (grid position "0"). Only for the HFK pulse concept a field strength reference was also measured in the exposed position of juvenile cod (Figure 26). In the IMR laboratory a field strength of 212 V/m was measured across the centre of a conductor while under fishing conditions with the gear at the seabed a similar rating of 207 V/m was measured in the equivalent position (all at 35 mm above the bottom).

Other pulse parameters of the HFK pulse concept were not according the reference given in Table 1. The specified and tested pulse period was 380 μs , while the measured pulse width on-board OD17 was 490 μs , and the delay time appeared increased from 60 to 100 μs . These changes are opposite the direction the results of this study indicate, where a reduction in the opposite direction is recommended. Measurements at sea showed that technical process is still on-going and that these adaptations are not always an improvement are indicated by the differences found in the latest electrode performance against the previous model as expressed by the voltages measured at the front and back part of the electrodes (Figure 21 and 22). The latest design expresses higher inductive influences and produced 10 % lower electrode voltage at the front part of the electrodes. The number of conductors were increased from 6 to 12 per electrode and the configuration illustrated in Figure 20 with relatively short isolator sections of 220 mm between conductors increased the number of "hot spot" of higher field strength. Measurements at sea showed that with a series of 12 conductors the internal 60 V source is overloaded. So a reduction of the number of conductors will raise the conductor voltage accordingly and so impeding (counteract?) the reduction of "hot spots", unless the 60 V source voltage is limited electronically.

4.4.4 Reference measurement conditions

The field strength measurement further showed that reference measurements at sea with the gear hanging vertically aside of the ship with the wing below the water surface do not present the field strength and electrode voltages values under normal fishing conditions and that large errors in the range of 8-10 % can be expected, and probably even higher when control routines are conducted with the vessel in the harbour. The outcome is influenced by the lower conductance with the gear at the seabed and the drag of the gear on the seabed which forces the electrodes to spread out to the designed conductor distance. The geometry is uncontrolled with the gear deployed vertically as than the back part of the trawl is still on deck. (Table 2 and 3). The electrode voltage measured at the surface is 7.8 % lower than result at the seabed and field strength measured in the centre position of the conductor pair was 31 % lower, while the salinity at the seabed was higher. This result demonstrates that measurements on the pulse gear as a standard inspection routine with the vessel in the harbour will even be worse and will lead to significant underestimates.

4.5 Effect on fish physiology, swimming performance and chances of survival

The inner part of the neural arch forms the spinal canal (canalis vertebralis) in which the spinal cord (medulla spinalis) is located. Furthermore there are several spinal nerves extending from the medulla spinalis (radix dorsalis and radix ventralis). A fracture at the basis of the neural arch may therefore most probably harm these nerves, which may result in paralysis. The inner part of the hemal arch makes out the hemal canal where the aorta and the vein vena caudalis are located. A fracture at the basis of the hemal canal may therefore probably harm these blood vessels, which again may lead to internal bleeding and reduce the capacity of the circulatory system. The vertebral column of teleost fishes is specialized to an aquatic mode of life; it is not weight bearing and movement between successive vertebral bodies is mainly limited to lateral flexion during swimming. Therefore, a fracture of vertebral bodies or a rupture of the intervertebral ligaments will reduce the swimming capacity of the fish. These are all possible primary effects of the fractures; there will most probably be many different secondary effects, especially related to the repair processes and the immune system. The effects of vertebral injuries in rainbow trout (*Onchorhynchus mykiss*) were observed over a long period of 355 days showing that fish with moderate to severe injury (vertebral misalignment and fracture), representing 28% of the total number exposed, had markedly lower growth and a worse physical condition than fish with no or little vertebral injury (Dalbey and McMahon, 1996).

A secondary effect may be that due to deterioration in swimming performance fish escaping from the gear may become more susceptible to predation.

5 Conclusions and recommendations

5.1 Juvenile cod

The results on juvenile cod showed that all fish (involving 12 trials and a total of 168 specimen) survived this harshest pulse condition even after a series of 4 exposures with short intervals. The post mortem research showed not a single vertebral injury or haemorrhage, which confirms that electric pulse fishing will not affect juvenile cod stocks.

5.2 Large cod

Results obtained in the pilot study of 2008 with similar length of cod with a series of 4 exposures were confirmed in this present study with a single exposure. Vertebral injuries occurred with 50-70 % of the fish under various pulse compositions (26 trials and a total of 262 specimen) and did not reduce when

the shape of the burst was modulated according the “tooth-shape model”. The threshold value for the amplitude of the conductor voltage was found at to 20 V. At this value injuries reduced from 60 to 30 %.

Injuries occurred in all three tested pulse concepts, although the Delmeco TX19 type had a lower score. This was probably related to the frequency (80 Hz), as at this setting the injuries of the HFK stimulus reduced (trial and probably also the shorter pulse duration and so the energy contents. We found a reduction of injuries with rising frequencies ≥ 80 Hz. At 180 Hz visible injuries reduced even to zero. The 100 Hz test (Table 4, trial 31) injuries unexpectedly increased, which could have been related to the longer delay period. A longer delay time between the positive and negative part of the pulse seems to contribute to injury as indicated by the 80 Hz trials with the delay period as tested parameter (Appendix B, Table 4, trial 37 and 38) and the Delmeco TX68 versus Delmeco TX19 trials.

It is recommended to study vertebral injuries of the landings of cod on-board TX19, which is equipped with the pulse concept with shortest delay time.

Reduced injuries were also found on a trial with reduced burst period (Appendix B, Table 4, trial 20 versus trial 21). Theoretically an equivalent result can be expected by the increase of the towing speed (which is practically limited, while trial 20 suggests that an increase of factor 2 could be needed) or a reduction of conductor length by 50 %, which could be more feasible. Another approach would be to deter the cod from the bottom.

5.2.1 Post mortem results

The post-mortem results did confirm the observations of injuries directly after the exposures and showed that the pulse stimuli can harm both the musculature and skeleton in cod. In a single case minor haemorrhages were found without bone fractures (Delmeco TX19, Appendix B, Table 4, trial 19).

5.2.2 Technical improvements and recommendations

As the larger cod paralysed for most of the cases after exposure or showed a disorientated tail flapping behaviour, it is likely that most of the fish that suffer similar exposure at sea will be caught. However, there is an ethical motive to find solutions to reduce such injuries. This can be achieved by increasing pulse frequency and as beam trawling is an operation involving two nets paired tests with nominal and adapted pulse settings can be done in one haul to show the effects to the target species of flatfish and the effect on injuries in cod.

Especially tests with increased pulse frequency are recommended or with modulated bursts of short duration instead of singles. Theoretically an equivalent result can be expected by increasing the towing speed or reducing conductor length. Another approach would be to deter the cod from the bottom.

The measurement at sea on-board OD17 showed that the total conductance using a series of 20 conductors in an electrode can be regarded as an overload condition, as the internal 60 V source dropped to 43 V, measured at the frontal conductor pair (Figure 22). This condition was the strongest (38 V) with the latest electrode lay-out with a steel re-enforced wire (Figure 21), while also the rising edge of the pulse was less steep and raised according a power of E, showing the overall inductance was apparently higher in the latest design. This demonstrates the design is not completed and progress affecting the pulse amplitude needs to be monitored.

5.3 Relevance of the laboratory results

The *in-situ* measurements of field strength at sea showed that the laboratory experiments were carried out with representative electric stimuli. Field strength measurements conducted at sea showed that the HFK stimulus applied in the laboratory was similar for the fished conditions and that the Delmeco TX68 field strength values applied in the laboratory were 54 % higher than the valued measured at sea.

In addition it was found that measurements should be done with the gear on the seabed to allow for the proper electric field build-up, which is different when the gear is hanging from the side, due to the fact that the electrodes are not parallel at equal distance in the same plane in this mode, and the differing conductivity of the bottom sediment.

It is recommended to measure field strength *in-situ* on-board TX19, due to the differences in pulse characteristics with other Delmeco systems in use and to investigate the simulation matches the commercial applied pulse stimulus

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7 Quality Assurance

IMARES utilises an ISO 9001:2008 certified quality management system (certificate number: 57846-2009-AQ-NLD-RvA). This certificate is valid until 15 December 2012. The organisation has been certified since 27 February 2001. The certification was issued by DNV Certification B.V. Furthermore, the chemical laboratory of the Environmental Division has NEN-AND-ISO/IEC 17025:2005 accreditation for test laboratories with number L097. This accreditation is valid until 27 March 2013 and was first issued on 27 March 1997. Accreditation was granted by the Council for Accreditation.

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Justification

Rapport C141/11

Project Number: 430.8101.031

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

Approved: B. van Marlen (M.Sc.)
Fishing Gear Technology Researcher

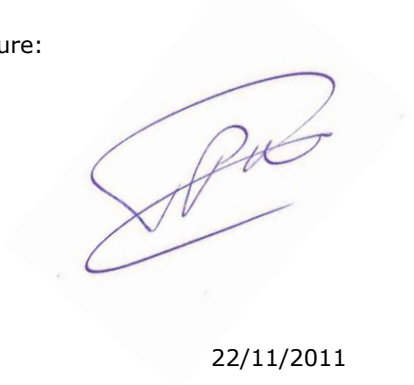
Signature:



Date: 22/11/2011

Approved: T.P. Bult (Ph.D.)
Head Fisheries Department

Signature:



Date: 22/11/2011

9 Appendix A: Figures and illustrations not in the main text



Figure 2 Typical HFK "PulseWing" system

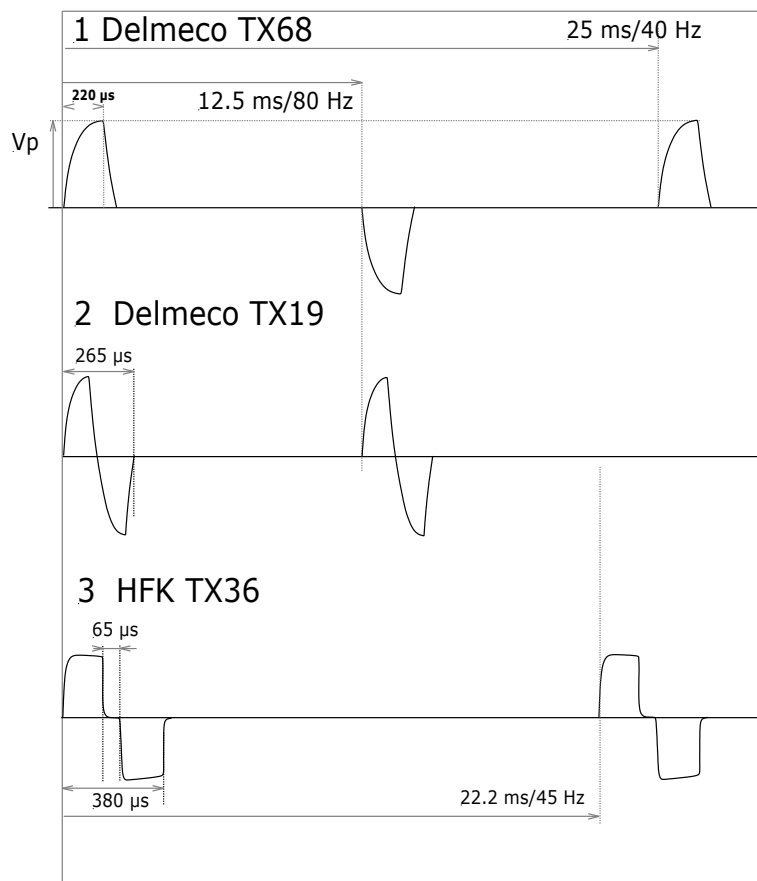


Figure 3 Nominal settings of the three tested pulse concepts

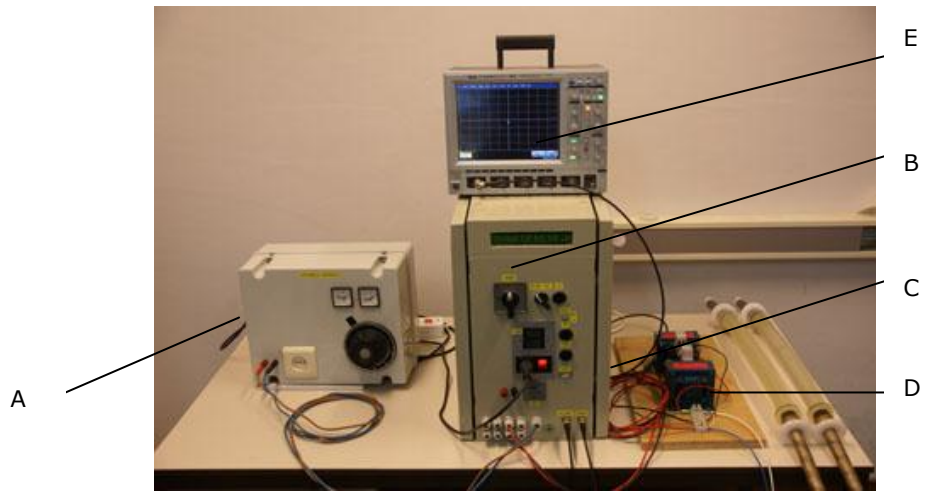


Figure 4 Simulator equipment of the Delmeco pulse concepts consisting of an adjustable power supply (A), pulse generator (B), output inductance (C), electrodes (D), oscilloscope (E).

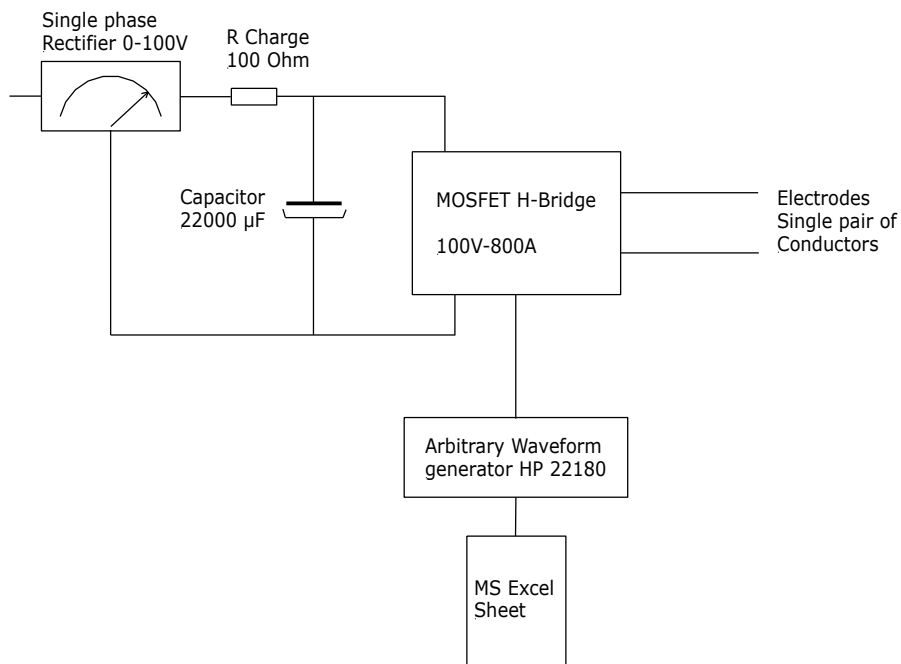


Figure 5 Schematic overview of main components of the simulated HFK pulse concept

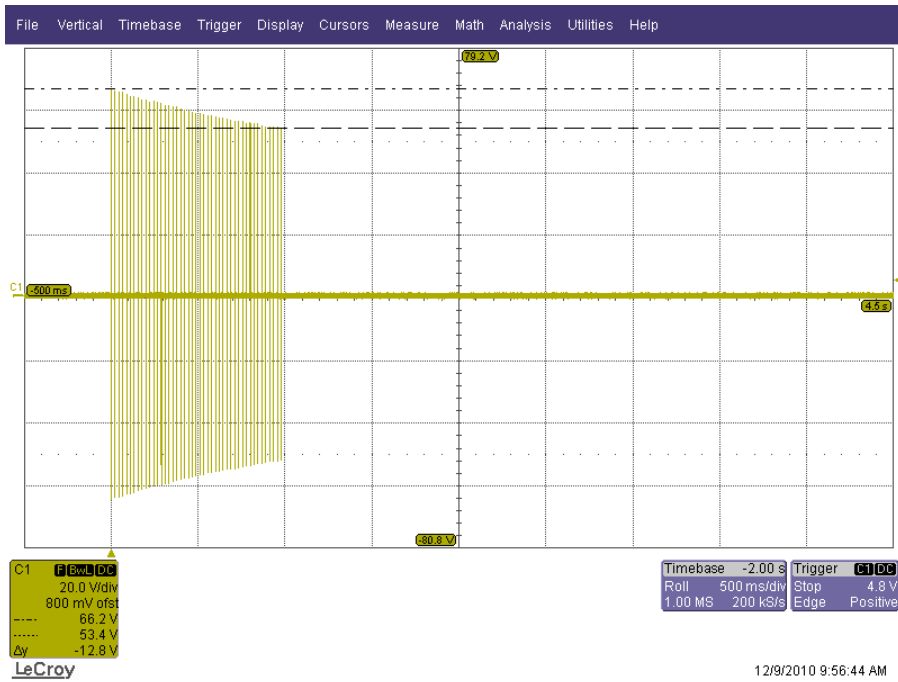


Figure 6 Simulated HFK 60 V burst of 1 s showing the amplitude voltage reducing from 66 to 53 V over the burst period.

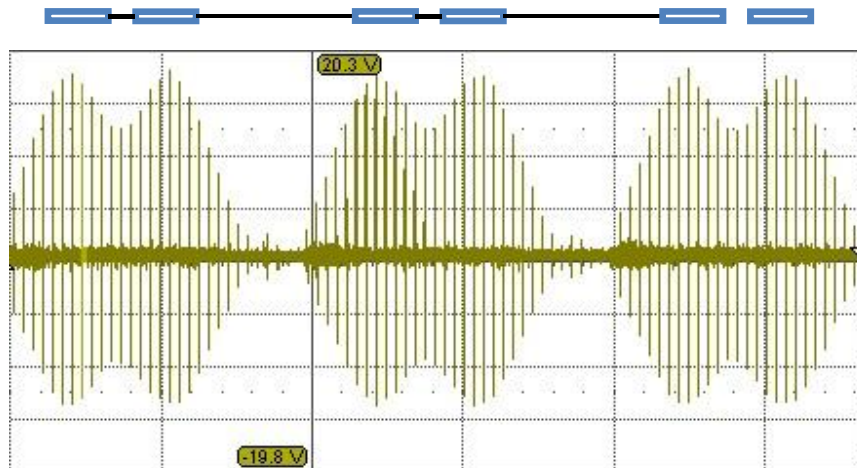


Figure 7 Modulated burst pattern of the electric field with a total length of 1 s simulating the dynamic peak effects of the conductor hot spots to a fish in static position on the passage of a series of 6 conductors (blue) isolated electrode parts (black).

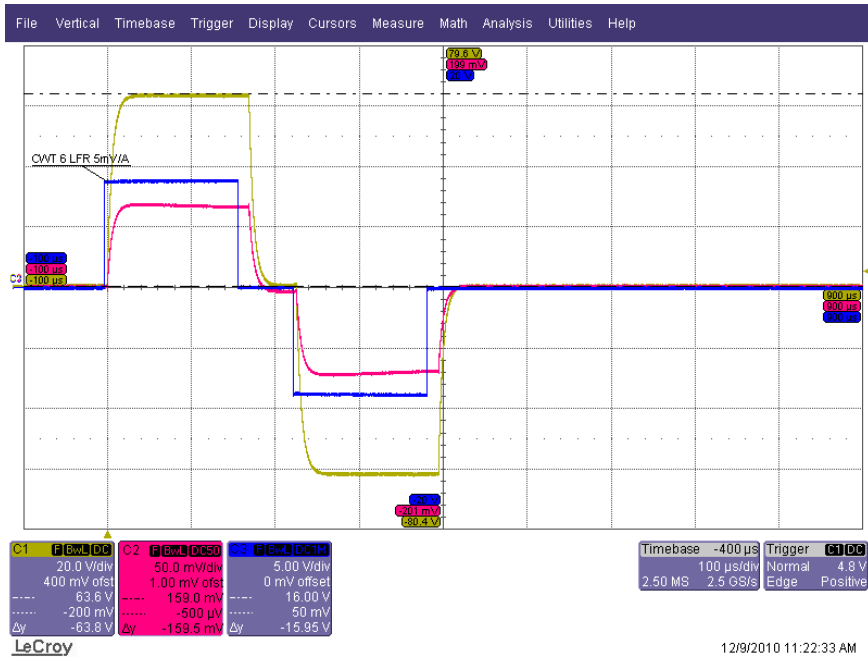


Figure 8 HFK simulated nominal pulse concept with 60 V electrode voltage at 45 Hz, total pulse duration 380 μ s, single pulse width 170 μ s, delay time 170 μ s. The first channel is the voltage across the conductors, Ch 2 the conductor current (106 A). The blue signal (Ch 3) is the trigger waveform to the H-bridge.

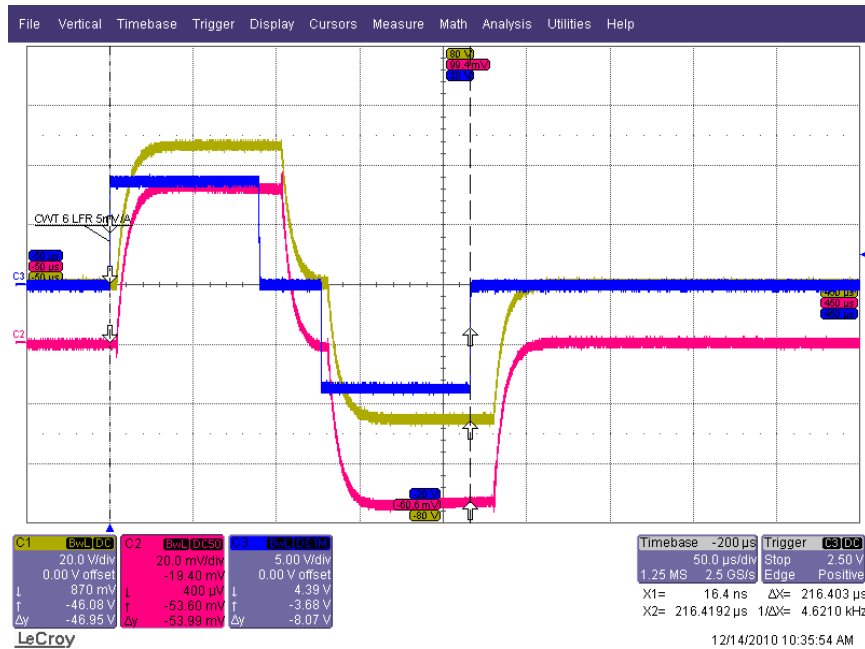


Figure 9 HFK simulated pulse concept with 46 V/106 A voltage (Ch 1)/current (Ch 2) amplitude at 80 Hz, total pulse duration 250 μ s, single pulse width 100 μ s, delay time 50 μ s. Ch 3 is the gate signal to the H-bridge.

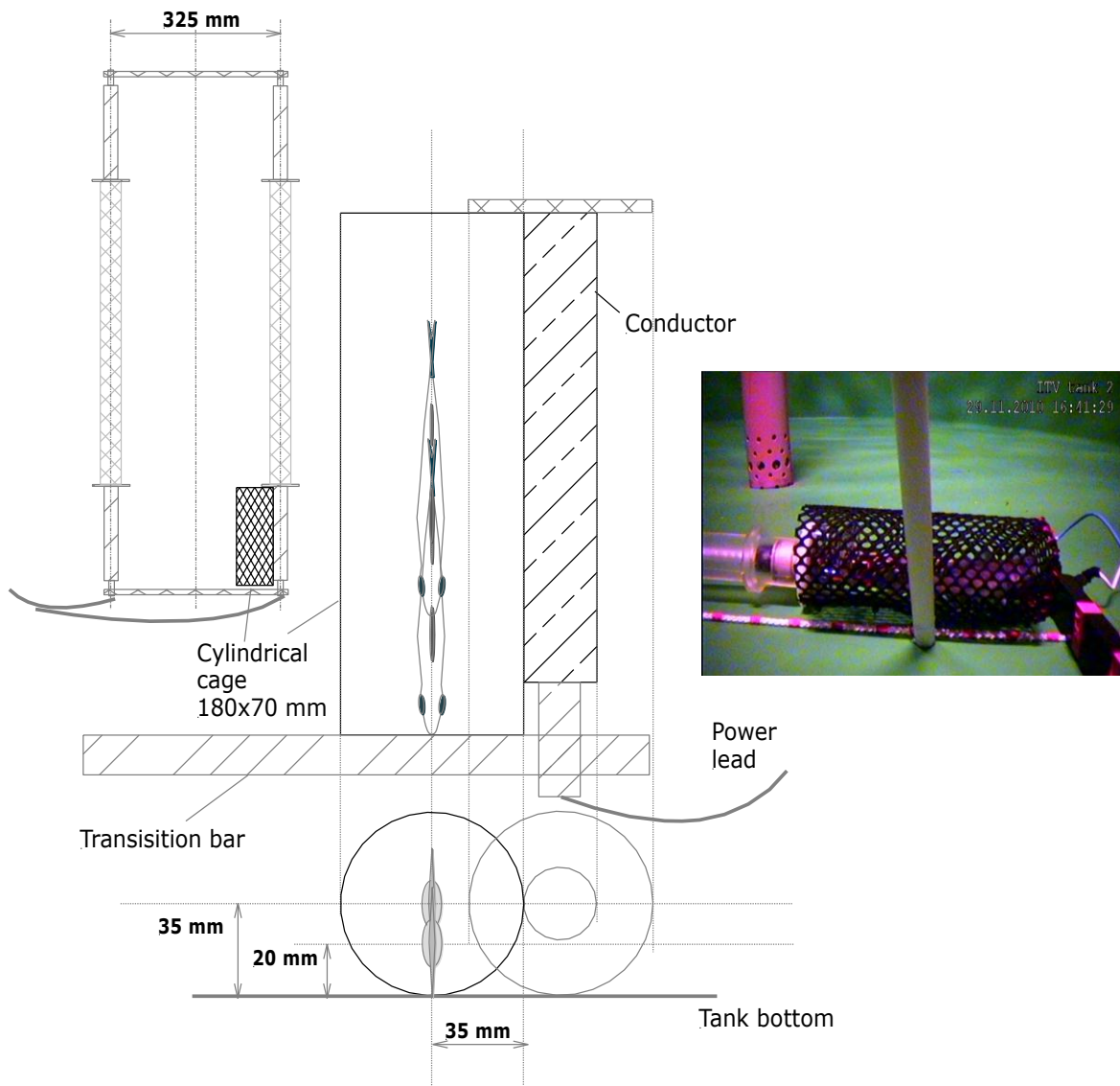


Figure 10 Longitudinal position of juvenile cod relative to the conductor and a picture of the actual positioning during the experiments. The marked cord is the reference on which field strength measurements were taken.

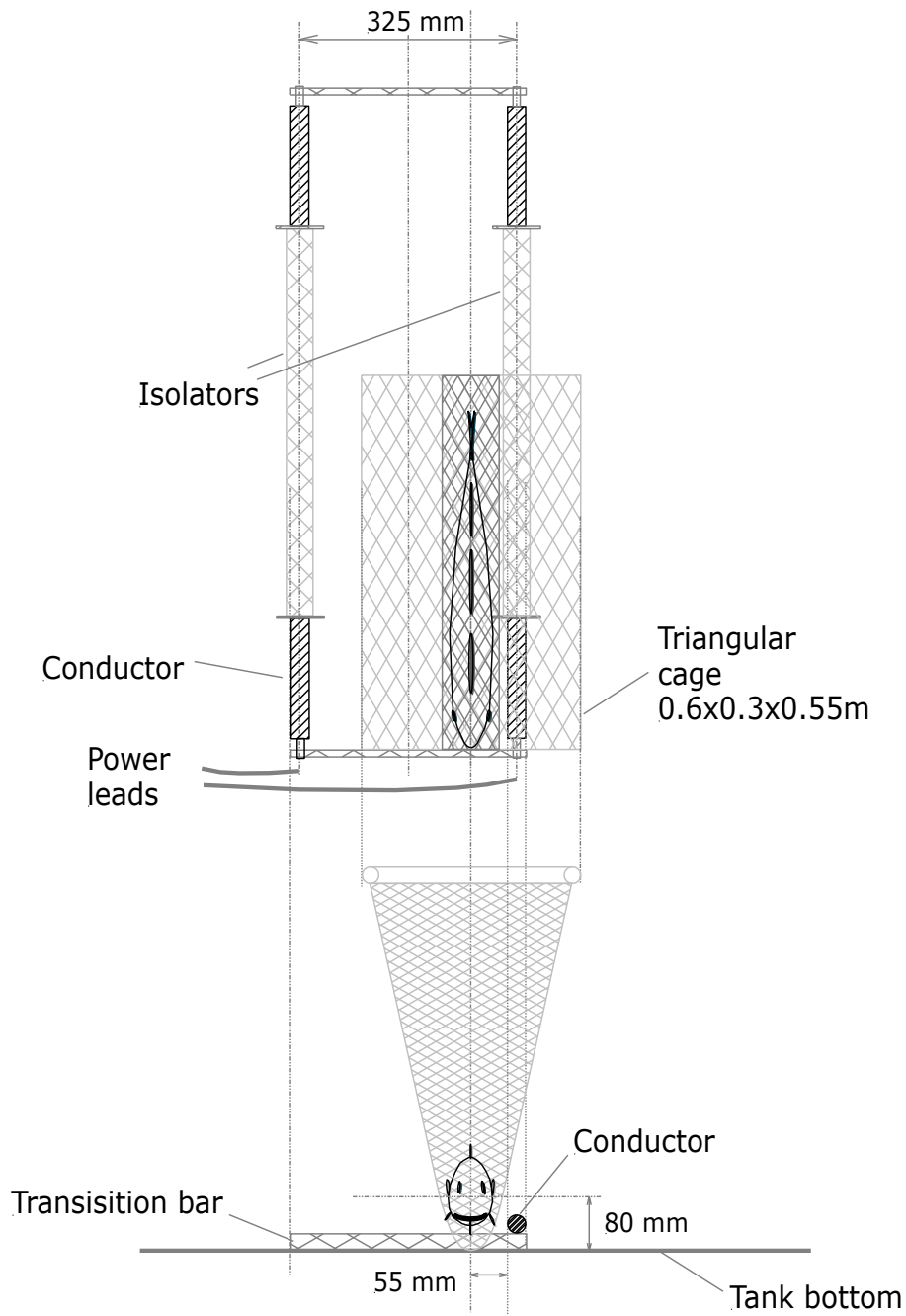


Figure 3 Position of the large cod relative to the conductor during the exposures and the reference position/level where the field strength was measured.



Figure 4 Black tail patterns on large cod directly after the exposures indicating vertebral injuries.

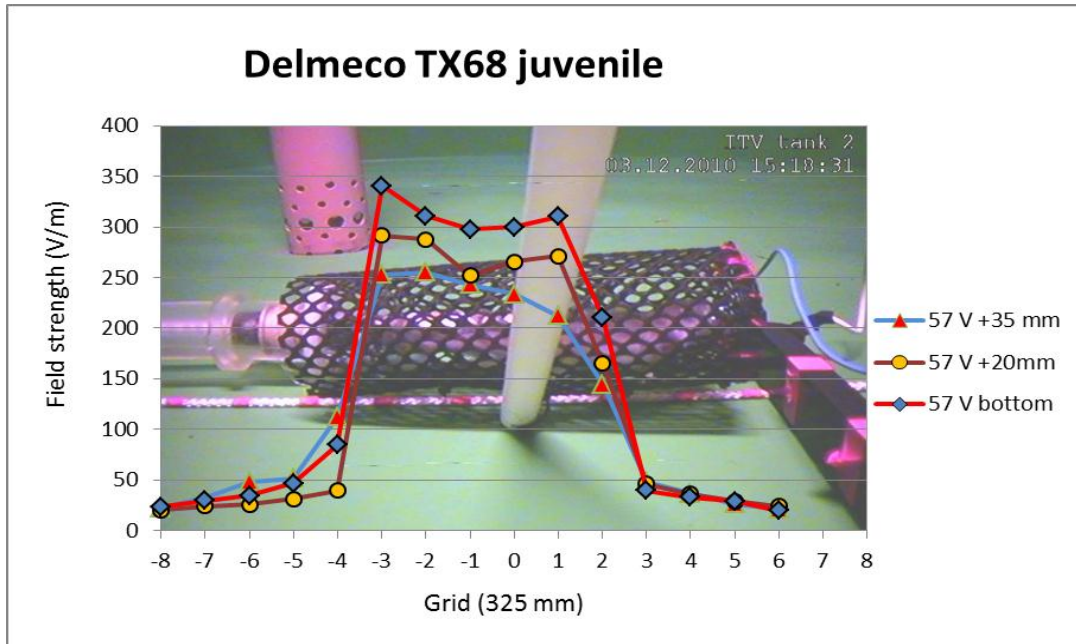


Figure 5 Field strength reference of Delmeco stimulus (TX68) for juvenile cod measured along the 35 mm axis at the bottom and two levels above the bottom (25 and 35 mm) in steps of 32.5 mm with the "zero" value representing the centre of the conductor. Across the centre the field strength ranged between 255 and 311 V/m.

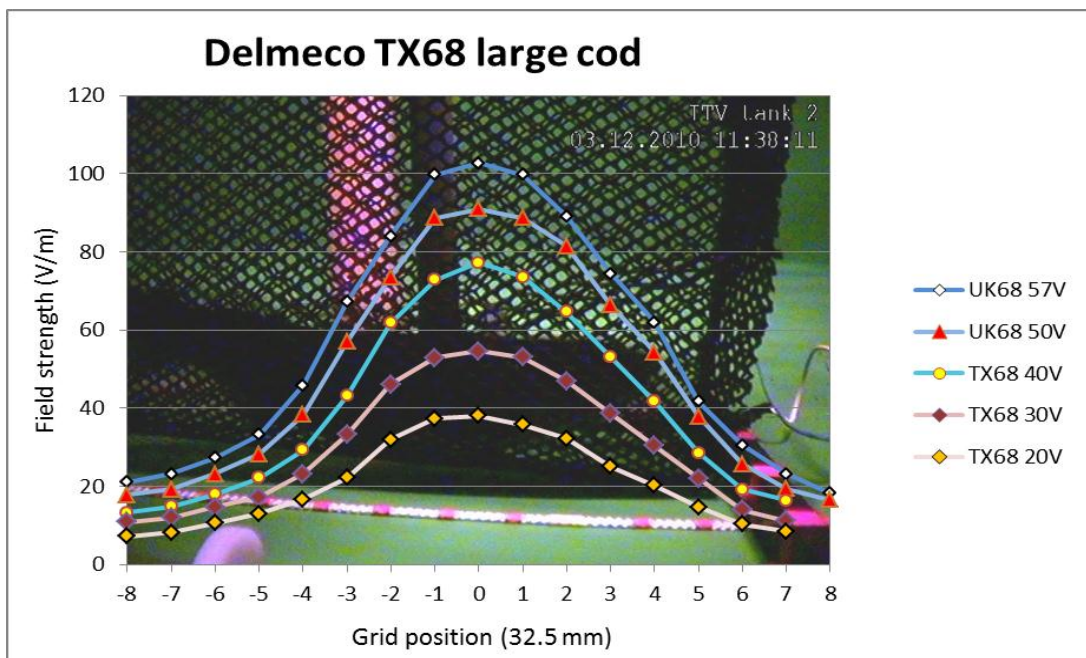


Figure 6 Field strength reference of one of the Delmeco stimulus type TX68 for 0.5 m cod measured at a level of 80 mm above the tank bottom along the 55 mm axis in steps of 32.5 mm with the "zero" value representing the centre of the conductor. At 20 V amplitude a threshold condition was found where the injuries reduced from 60 to 30 %. In the position across the centre of the conductor a field strength of 38-103 V/m was measured with an voltage amplitude range of 20 to 57 V measured across the conductors.

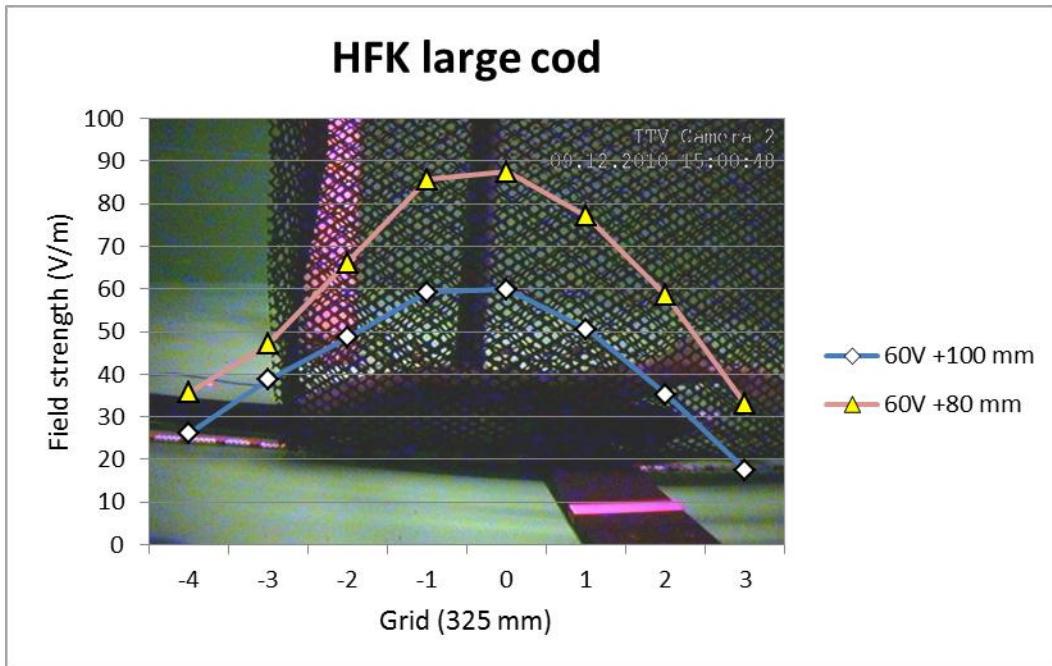


Figure 15 Field strength results measured with the HFK stimulus with nominal settings in the reference position of large cod as well as 20 mm above this level. The outcome at +80 mm measured opposite the centre of the conductor is 10 V/m lower than the Delmeco outcome under similar conductor voltage. Across the centre the field strength at a level of 80 mm was 86 and at 100 mm 60 V/m.

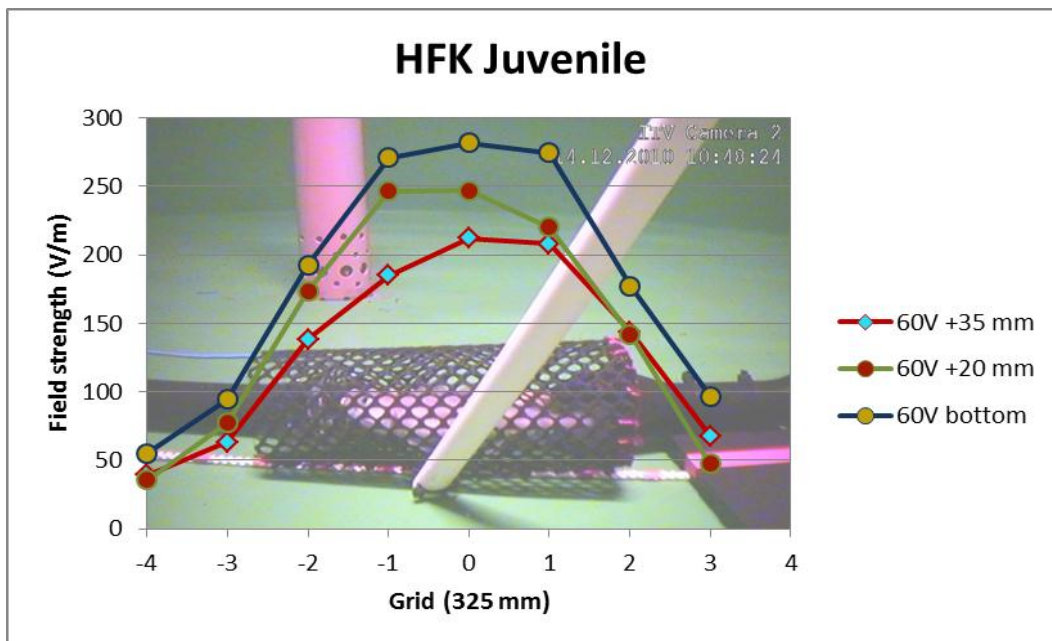


Figure 16 Field strength reference of HFK stimulus with the maximum amplitude setting of 60 V (0-peak) for juvenile cod measured along the 35 mm axis at the bottom and two levels above the bottom (25 and 35 mm) in steps of 32.5 mm with the "zero" value representing the centre of the conductor. Across the centre the field strength ranged between 212 and 282 V/m.

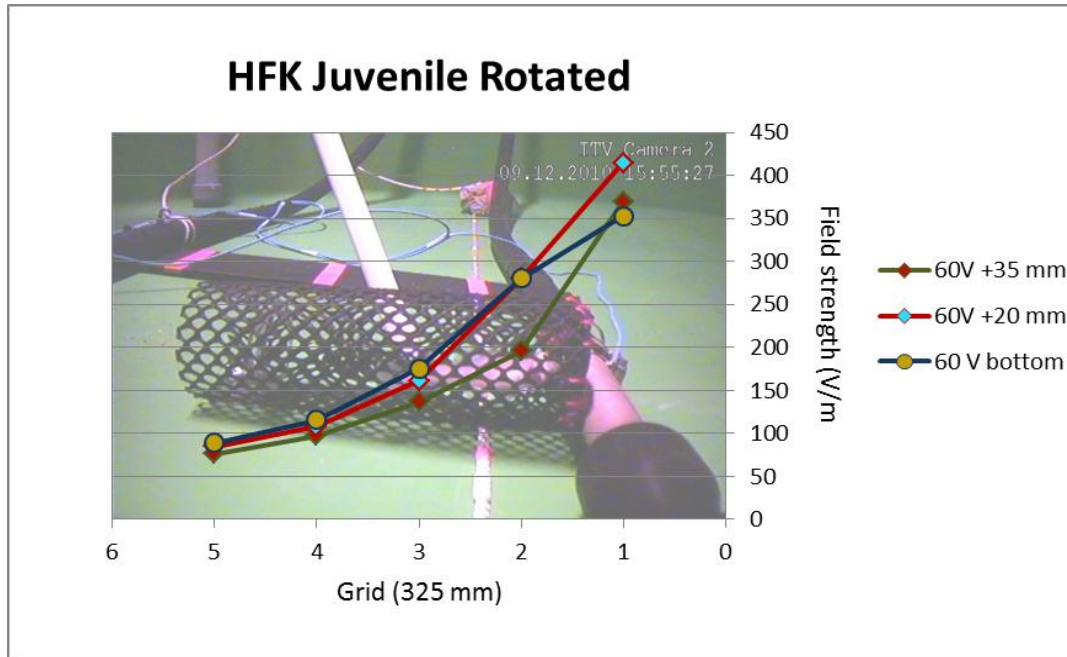


Figure 17 Field strength ratings measured perpendicular to the centre of a conductor in the height range of the animal's position relative to the bottom during the exposure. This reference is linked to session 24 conducted on 09-12-2010. The first position is taken with the probe tip against the conductor. The highest field strength was measured at a height of 20 mm above the bottom with fits to the longitudinal centre. Field strength was in the range of 76 to 415 V/m.

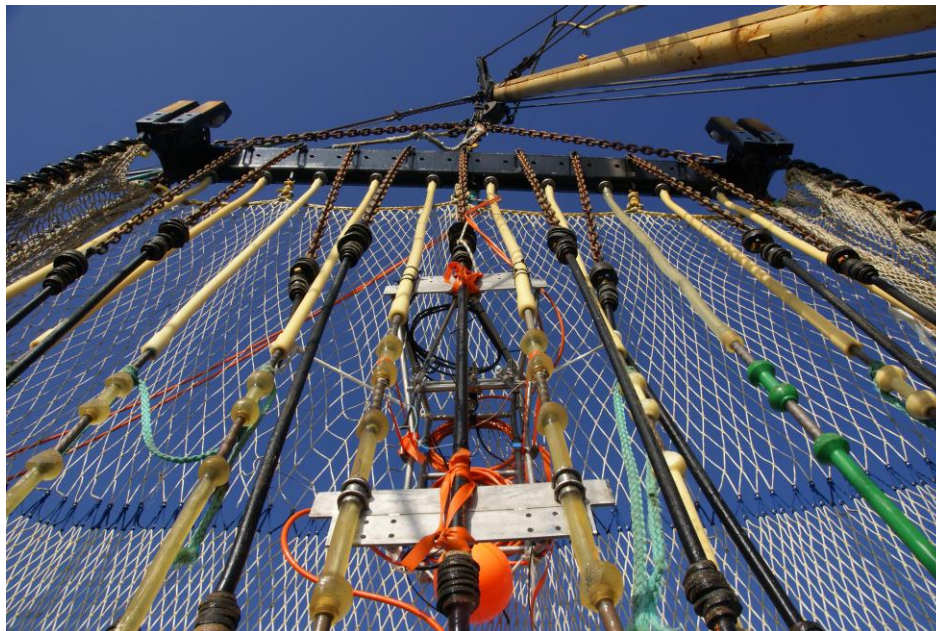


Figure 18 The rigid frame carrying three field strength probes and an DeepSea 1060 underwater video camera clamped on the first conductor pair of field 5 of the Delmeco pulse system on-board TH10 (220 kW licenced vessel).



Figure 19 The rigid frame carrying three field strength probes and an DeepSea 1060 underwater video camera clamped on the first conductor pair of field 5 and 6 of the HFK PulseWing system on-board OD17 (1470 kW licenced vessel).

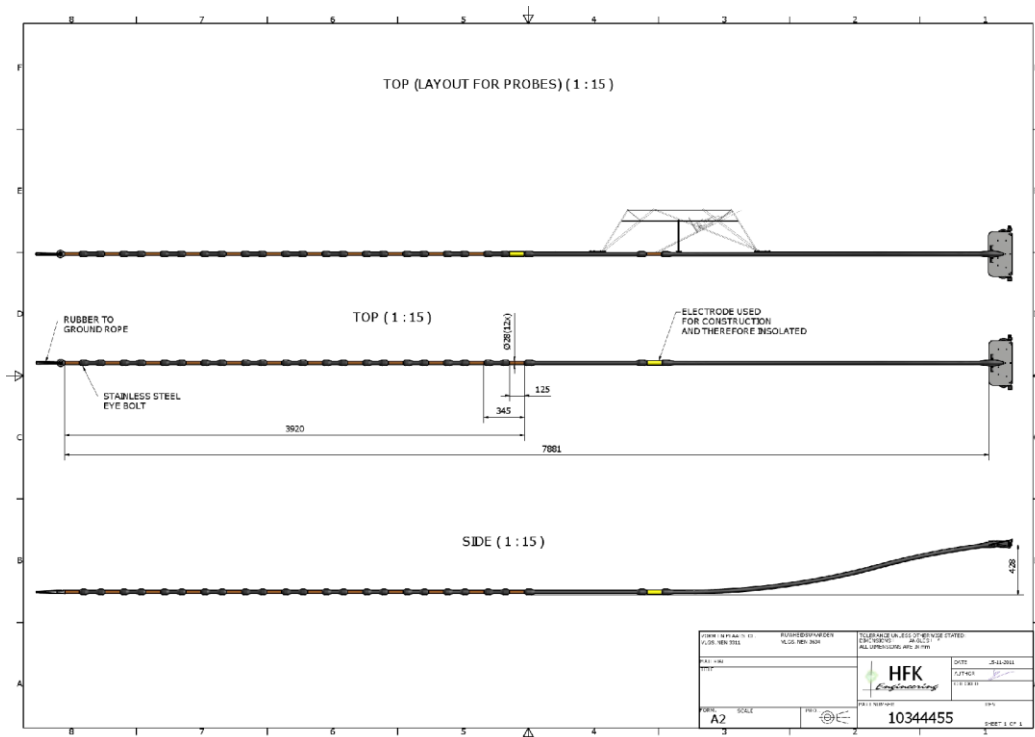


Figure 20 HFK Electrode design with 12 conductors 27 mm diameter and 125 mm length. For the field strength measurements the first conductor/isolator string was reversely connected (top part of the drawing with the side view of the attached position of the measurement frame) to minimise the influence of the metal frame carrying the field strength probes to the electric field.

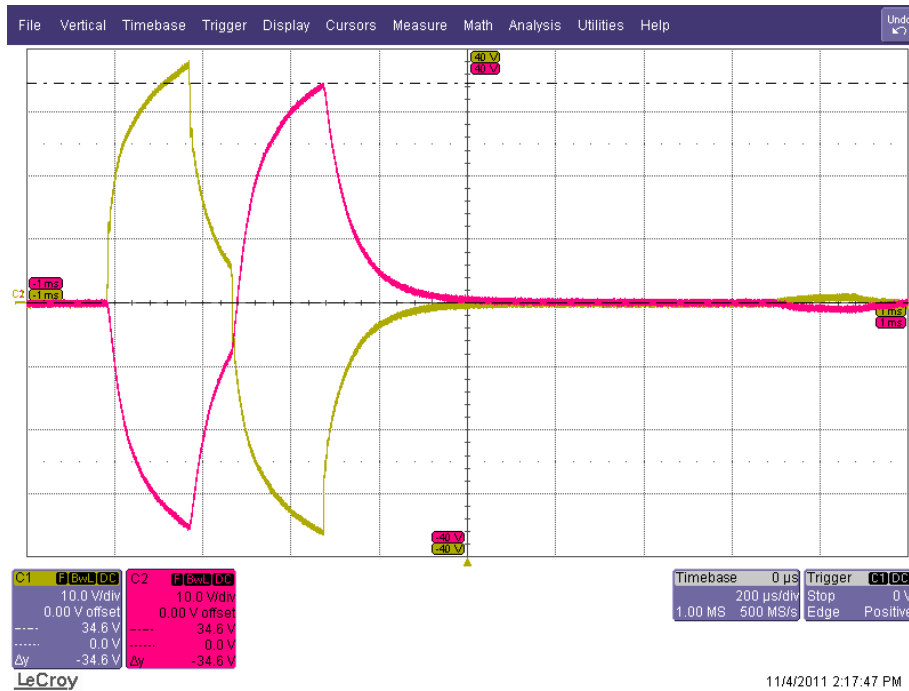


Figure 21 Electrode voltage measured on 4 November 2011 on-board OD17 on the **latest** HFK electrode design across the most frontal 27 mm conductor (electrode is built of 2 conductors of 27 mm and 10 conductors of 33 mm diameter). Channel 1 represent the pulse across the frontal conductors, Channel 2 the pulse at the back end of the electrode, respectively 38 and 34 V.

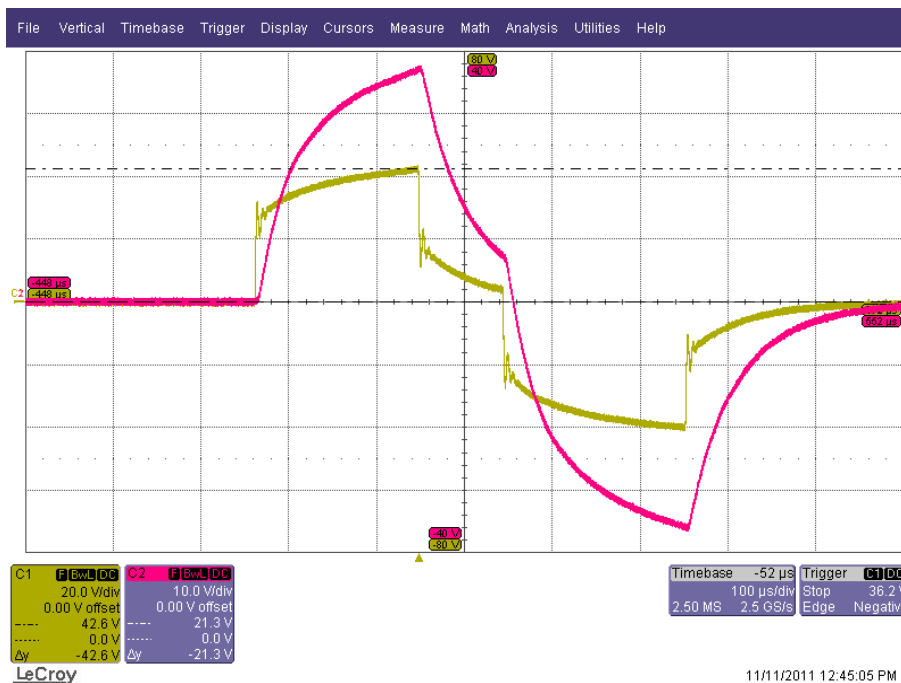


Figure 22 Electrode voltage measured on 11 November 2011 on-board OD17 on the **previous** HFK electrode design across the most frontal 27 mm conductor (electrode is built of 12 conductors of 27 mm). Channel 1 represent the pulse across the frontal conductors, Channel 2 the pulse at the back end of the electrode, respectively 42.6 and 37 V.

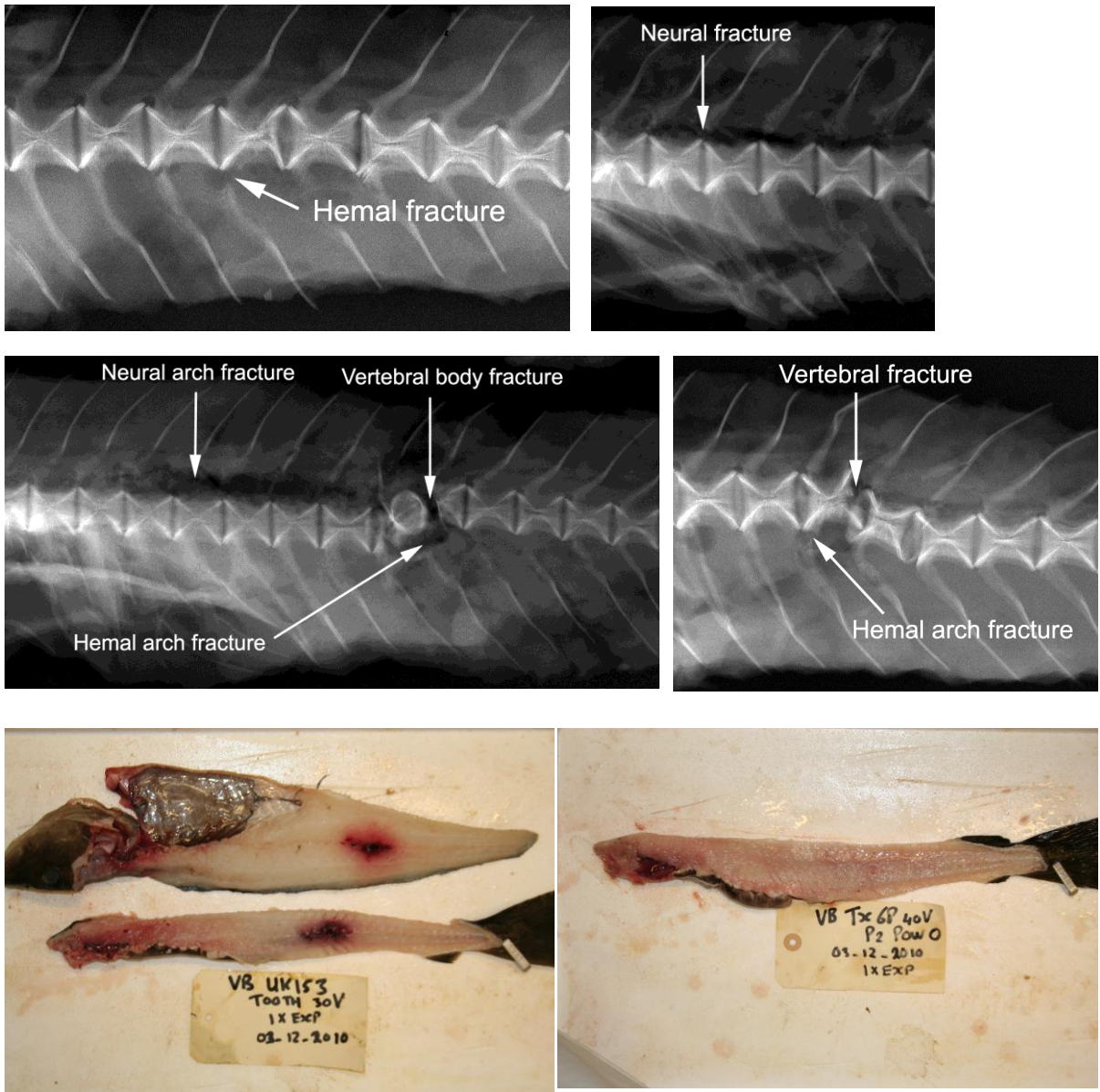


Figure 23 Examples of injuries in larger cod detected by radiological analysis and dissection

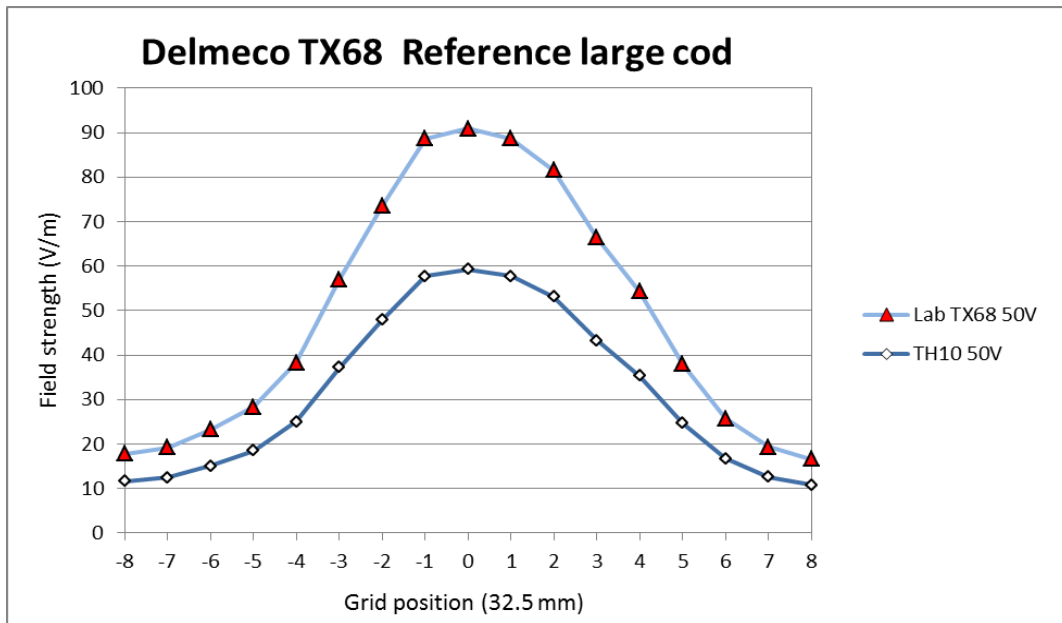


Figure 24 Field strength reference in the exposed position of large cod for the Delmeco TX68 pulse concept applied in the IMR laboratory and the down scaled result derived from the real fishing condition measured on-board TH10 (59 V/m). The TH10 curve is the downscaled result from this single point result at the centre of the conductor. The field strength in other grid positions was calculated from the measured curve in the laboratory.

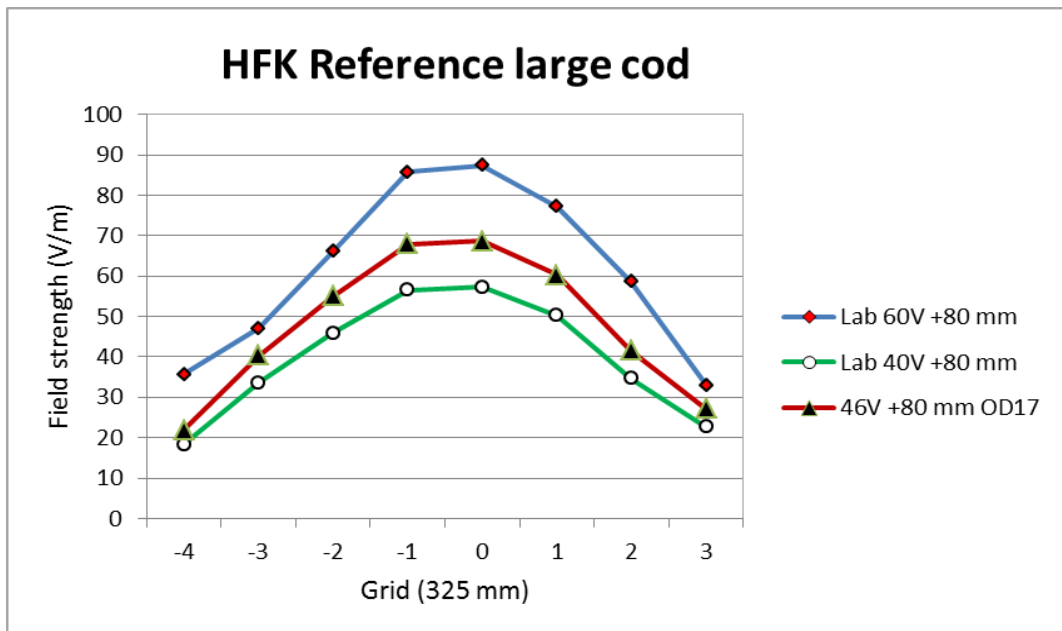


Figure 25 Field strength reference in the exposed position of large cod for the HFK pulse concept applied in the IMR laboratory and the down scaled result measured HFK gear at the seabed on-board OD17 (69 V/m). The OD17 curve is the downscaled result from the single point outcome at the centre of the conductor. The field strength in other grid positions was calculated from the measured curve in the laboratory.

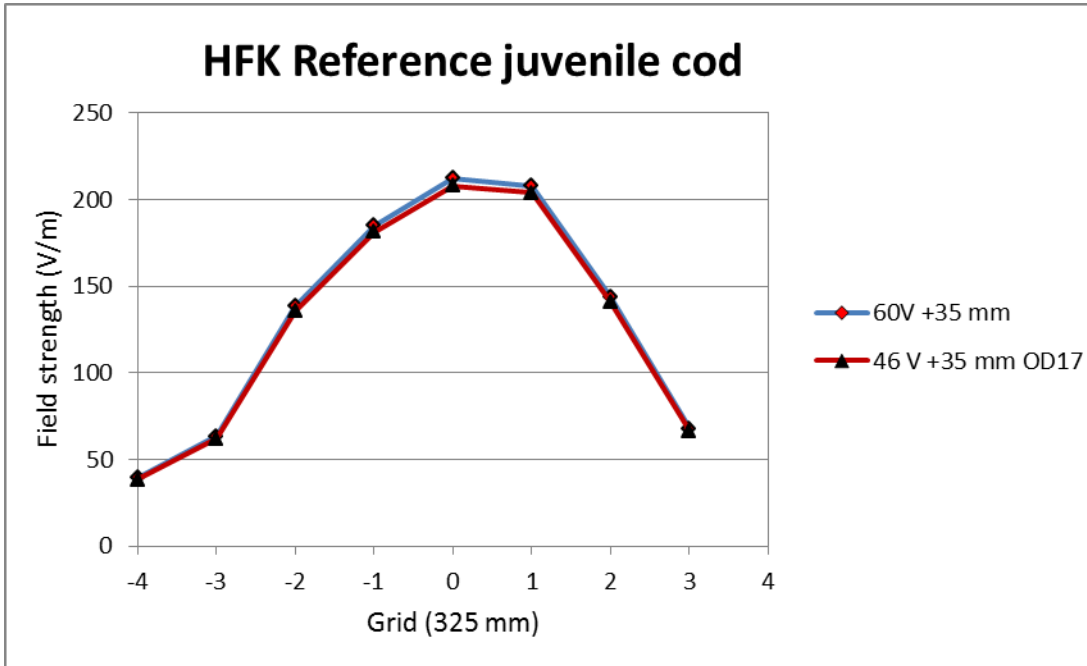


Figure 26 Field strength reference in the exposed position of juvenile cod for the HFK pulse concept applied in the IMR laboratory (212 V/m across the centre of a conductor) and the down scaled result measured HFK gear at the seabed on-board OD17 (207 V/m across the centre of a conductor). The OD17 curve is the downscaled result from this single point outcome. The field strength in other grid positions was calculated from the measured curve in the laboratory.

10 Appendix B: Table with major results not in the main text

Table 4 Overview of trials in December 2010, pulse settings and responses as a function of execution (green = no effect ; pink = strong effect; white = threshold to less effect; and blue = not analysed). All voltage ratings in this report refer to the peak voltage measured over the positive part of the pulse, described as zero to peak.

Trial (nr)	Date	Fish group length in class (m)	Nr of Fish in group (nr)	Pulse ID	Ampl. (V)	Freq (Hz)	Pulse width (μ s)	Response	Post-Mortem				
									Hemor	Bone fract	Neural	Vert	Hemal
1	01	0.1	20	VB-TX68 P1-POW0	57	40	220	Paralysed during and some seconds after. No dead fish, no injuries observed	0	0	0	0	0
2	01	0.1	20	VB-TX68 P1-POW0 4 Exp.	57	40	220	Paralysed, short coma during and after exposure, stopped breathing, all recovered no injuries observed. Normal behaviour after	0	0	0	0	0
3	01	0.1	20	None REF				Reference group for X-ray analysis	0	0	0	0	0
4	01	0.5	10	VB-TX68 P1-POW0	57	40	220	At least 6 fish with vertebral injury	6	6	4	5	2
5	02	0.5	10	VB-TX68 P1-POW0	50	40	220	At least 4 fish with vertebral injury	4	4	3	3	0
6	02	0.5	10	VB-TX68 P1-POW0	40	40	220	At least 7 fish with vertebral injury	9	8	8	7	2
7	02	0.5	12	VB-TX68 P1-POW0	30	40	220	At least 7 fish with vertebral injury. Two taken out and examined. One had vertebral injury	8	7	6	5	3
8	02	0.5	10	VB-TX68 P1-POW0	20	40	220	One fish with immediate vertebral injury. Fish stayed overnight. The next day two others showed vertebral injury	3 (3)	1	1	1	0
9	03	0.5	10	VB-TX68 P1-POW6 tooth shape	30	40	220	At least 6 fish with vertebral injury. With this result arguments against the un-adapted pulse pattern can no longer be maintained.	5	5	3	3	3
10	03	0.5	10	VB-TX19 P2-POW0	22	80	220	No injuries observed	(1)	0	0	0	0
11	03	0.5	10	VB-TX19 P2-POW0	40	80	220	At least one fish with vertebral injury	1	1	0	0	1
12	03	0.5	10	VB-TX19 P2-POW0	50	80	220	At least 5 fish with vertebral injury	3	4	4	3	0
13	03	0.1	21	VB-TX19 P2-POW0	50*	80	220	Lower voltage simulator at 60 V >100%. No injuries observed.	0	0	0	0	0

Trial (nr)	Date	Fish group length in class (m)	Nr of Fish in group (nr)	Pulse ID	Ampl. (V)	Freq (Hz)	Pulse width (μ s)	Response	Post-Mortem				
									Hemor	Bone fract	Neural	Vert	Hemal
								Significant lower paralysing response. Fish kept alive over the weekend					
14	06	0.5	10	VB-TX68	30	57.5	150	Less contraction power sensed. Noise heart which could be the sync of vertebral injury. At least 6/7 injured fish	6	6	3	3	5
15	06	0.5	10	VB-TX68	30	30.6	220	Noise sync relates to cracking of the vertebrae soon after black tail marks indicating vertebral injury. At least 7 injured fish. Sound seems to occur during the exposure.	5	4	4	2	1
16	06	0.5	10	VB-TX68	30	30.6	150	Sound recorded underwater. Five sounds determined and related to 5 injuries expressed in black tail patterns.	6	6	6	5	2
17	06	0.1	20	VB-TX68 90° rotation	57	40	220	Fish exposed with head perpendicular to centre of the conductor. Fish paralysed after exposure for 10-60s. All fish recovered and were still alive the next day.	0	0	0	0	0
18	07	0.1	8	VB-TX68	57	40	220	Repeat of 1st to confirm the reported effect. Medium to low contraction, incidental after effect of 10 s after exposure.	n.a.				
19	07	0.5	10	VB-TX19 P2-POW0	30	80	220	One fish (first) with unknown injury last fish deadly injured. Medium to incidental strong contraction, mostly immediate recovery	1 (6)	1	1	0	0
20	07	0.5	10	VB-TX68 P1-POW0- 0.5 s	30	40	220	Pulse period reduced with 50 %. Two fishes deadly injured, a third (nr 8) suspect	2	2	2	2	1
21	08	0.5	10	VB-TX68 P1-POW0 Electrode 425-41mm	30	40	220	Six out of ten fishes with cracking sound, six fish with black patterns on tails. Fishes with no sound produced a weak response.	6	6	4	4	2
22	08	0.5	10	HFK	30	80	100/ 50	Two fishes deadly injured, one is small. Two sound refs and black tail marks. The other eight fishes were still alive next day & did feed	2	1	1	1	1

Trial (nr)	Date	Fish group length in class (m)	Nr of Fish in group (nr)	Pulse ID	Ampl. (V)	Freq (Hz)	Pulse width (μ s)	Response	Post-Mortem				
									Hemor	Bone fract	Neural	Vert	Hemal
23	09	0.1	10	HFK	60	45	165/50	All fish responded weak, normal behaviour directly after exposure	0	0	0	0	0
24	09	0.1	10	HFK-90°rotation	60	45	165/50	Low response, only the 10 th fish produced a very heavy response near coma. All the fish transferred to recovery tank to stay overnight.	0	0	0	0	0
25	09	0.5	10	HFK	40	45	165/50	Strong contraction, 7 fishes injured and identified during and after exposure in sound and colouring of the tail.	6	6	4	3	4
26	10	0.5	10	HFK	40	45	165/50	Cage lifted with 50 mm. Four fatal injuries. It seemed the smaller fish were more sensitive to injury!	4	4	3	2	2
27	10	0.1	10	HFK	60	180	50/50	Indication smaller fish respond heavier to higher frequencies. The response was low/medium, fish contracted, but recovered immediately after the pulse. Group stayed alive over the weekend	0	0	0	0	0
28	10	0.1	10	HFK-90°rotation	60	180	50/50	No significant difference observed. Response medium to low. Fish of 27 & 28 were? healthy and alive next Monday	0	0	0	0	0
29	13	0.5	10	HFK	40	180	50/50	1st case with zero injury. Response low/medium, normal behaviour after. Faulty trigger with injury rejected as the animal was out position with the tail opposite the conductor	1	1	0	0	1
30	13	0.5	10	HFK	40	180	50/50	As trial 29 tail projected opposite the conductor (centre cage (shifted forward with 210 mm). No injuries so the single case of 29 could have been caused by bended vertebrae.	0	0	0	0	0
31	13	0.5	10	HFK	40	100	80/110	H4N Zoom installed with pulse sound & external microphone?. Three injuries and a fourth suspected from sound recording. Fifth fish is tagged 00030. It seemed the smaller	3(3)	3	2	3	1

Trial (nr)	Date	Fish group length in class group (m)	Nr of Fish in group (nr)	Pulse ID	Ampl. (V)	Freq (Hz)	Pulse width (μ s)	Response	Post-Mortem				
									Hemor	Bone fract	Neural	Vert	Hemal
								fish are more sensitive to injury.					
32	13	0.5	10	HFK	40	30	240/380	Four fishes with injury. H4N recorder switched off at 4 th fish. One crack at the end of the burst. The fish seemed to respond late in some cases. Is this the lower F boundary for injury?	4	4	1	1	2
33	14	0.1	10	HFK	60	30	240/380	Zero injuries. Low to very low response with direct normal behaviour after exposure. Some dizzy behaviour after.	0	0	0	0	0
34	14	0.1	10	HFK 90°rotated	60	30	240/380	Zero injuries. Low to very low response. Fish was capable of turning in the narrow cage.	0	0	0	0	0
35	14	0.5	10	HFK	40	45	165/50	Contraction medium to strong with four fatal injuries and a fifth suspect (case 7 tagged 00031).	4	4	2	2	2
36	14-2011	0.5	10	HFK	40	30	240/380	Three clear injured fish. 237 μ s Behaviour varied between very strong and a weak response. H4N Zoom recording	3 (3)	3	2	1	1
37	14	0.5	10	HFK	40	80	100/50	Contraction seemed > 1s. Crack of case 10 could underline this. Most of the contraction were strong and lasted long. 3 of 10 injured. F-threshold?	3	3	2	2	0
38	14	0.5	10	HFK	40	80*	100/1140	*negative part delayed to 1.22 ms. This session could show the effects of an asymmetric pulse pattern. Injuries increased from three to 5 audible/visible cases. Contraction varied from medium to strong.	5	5	2	2	3
39	??	0.5	10	None				Reference group	0	0	0	0	0

Remark: Only Distinct haemorrhages were scored, in some cases when the numbers of smaller haemorrhages were significant or relevant the numbers were added in brackets