The effect of pulse stimulation on marine biota – Research in relation to ICES advice – Progress report on the effects on benthic invertebrates

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Report C103/09

IMARES Wageningen UR

(IMARES – Institute for Marine Resources & Ecosystem Studies) Report number C103/09

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Fisheries Direction
P.O. Box 20401
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BO-07-002-910

Publication Date: 21/10/2009

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A_4_3_2-V7.0

Contents

Summary

Experiments were carried out in July 2009 on a range of benthic invertebrates (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 based on the Verburg-Holland stimulus.

Groups of twenty animals per species were exposed to three treatments of four 1 s bursts of electrical pulses using a pulse simulator: nearby (0.10-0.20 m distance), at medium distance (0.20-0.30 m), and further away (0.40 m) of the electrodes. A 1 s pulse burst is deemed to represent the in situ passage of the pulse field of the gear beneath a non-moving fish. A control group was used for all species to correct for handling effects. The animals were caught with methods minimizing catch effects, and kept in water quality controlled circulating sea water tanks, and regularly fed. Survival, food intake and behaviour were monitored for a period of some two weeks after the exposure. The data were analysed with generalized linear models in the SAS statistical package.

For two species (ragworm and European green crab) a 3-5% statistically significantly lower survival was found compared to the control group, when all exposures were lumped together. For the near field exposure a 7% lower survival was also found for Atlantic razor clam. For the other species (common prawn, subtruncate surf clam, common starfish) no statistically significant effects of pulses on survival were found. Surf clam seemed not to be affected at all, common prawn seemed to show lower survival in the highest exposures (near and medium field), while common starfish showed lower survival, but not for the highest (near field) exposure.

Food intake turned out to be significantly lower (10-13% less) for European green crab, except in the far field exposure for which the reduction (~5%) was non-significant. No effect at all was found for ragworm, surf clam and razor clam, lower food intake for common prawn, and higher for common starfish, but all these results were statistically non-significant.

Surf clam and starfish did not show any behavioural reaction at all, they did not move. The other species showed very low responses in the far field exposure range. In the medium and near field ranges the reactions were stronger. Food intake and behaviour recovered after exposure.

In general terms the effects of the pulse stimulus in terms of mortality and food intake can be described as low. It is therefore plausible that the effects effects of pulse beam trawling, as simulateed in this study, are are far smaller than the effects of conventional beam trawling.

This research is performed within LNV program Beleidsondersteunend Onderzoek (BO) under Ref.: BO-07-002-910, Vervolg Pulskor Bodemdieren 2, under programme: VPT “Verduurzaming Productie en Transitie”.

1 Introduction

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, b). 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 vertebral damage in cod, and suggested only weak responses and no mortality in lesser spotted dogfish (synonym: small spotted cat sharks) (*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).

Further studies were done on cat sharks (*Scyliorhinus canicula* L.) (De Haan et al., 2009), and cod (De Haan et al., 2008). This report gives the results of further experiments on benthic invertebrates.

2 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.

3 Confidentiality

Details of the pulse trawl system developed by Verburg-Holland Ltd. and in particular the characteristics of the stimulus are kept confidential for the sake of protection of knowledge and are therefore not revealed in this report. These technical details can be supplied in a confidential detailed report. This report is based on the specifications of the pulse stimulus of Verburg-Holland Ltd., and will only become available to the technical experts of IMARES and members of the ICES Expert Group, who will be asked to keep this knowledge confidential.

4 Materials and Methods

The following species were chosen for testing based on their abundance and variety in morphology: ragworm (*Nereis virens* L.), common prawn (*Palaemon serratus* L.), subtruncate surf clam (cut through shell) *(Spisula subtruncata* L.), European green crab (*Carcinus maenas* L.), common starfish (*Asterias rubens* L.), and Atlantic razor clam (American jack knife clam) (*Ensis directus* L. = *Ensis americanus*).

European green crab were caught in the Eastern Scheldt using a shrimp beam trawl (3 m wide and 8 m long) on FRV "Schollevaar" at a towing speed of 2.97 kts (5.5 km/h, 1.52 m/s). Subtruncate surf clams were caught in the same area with a mussel dredge at 2.16 kts (4 km/h, 1.11 m/s).

Figure 1: Species tested: from upper left to lower right: 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.).

The common prawn were collected by divers from the Grevelingen (at position "Dreischor") on 06/07/2009, and the starfish from the Eastern Scheldt (at "Kattendijke") on 07/07/2009. Atlantic razor clam were caught by a commercial fisherman specialized in this species using a suction dredge in the "Voordelta" at a towing speed of 0.189 kts (350 m/h, 0.097 m/s), and brought in on 30 June 2009. Ragworm (*Nereis virens* L.) were delivered by the Dutch company "Topsy Baits" on 01/07/2009.

The animals were kept in sea water circulated bins onboard, and then transferred to sea water circulated holding tanks at IMARES Yerseke (*Figure 2*). These tanks were covered with concrete tiles to keep them in the shade. Food was provided on a daily basis. The surf clam were kept in jars with a sandy sediment in which they buried. Atlantic razor clam and ragworm were kept in buckets with sand and placed in seawater tanks. The refreshment water was obtained from a known and controlled source also used by mussel hatchers.

Figure 2: Large sea water circulated holding tanks with storage compartments at IMARES Yerseke.

4.1 Data measured and equipment used

The measuring equipment consisted of a 200 MHz LeCroy WaveSurfer 24XS oscilloscope with a differential high voltage probe type ADP 305 (SN5069) and a CWT Rogowski 60B current probe (0.5 mV/A). Measurement results (screen images) were stored as JPEG images on hard disk. Environmental data of the water in the tanks such as salinity, temperature, turbidity, conductivity, and oxygen content were monitored during all experiments with a Hydrolab data sonde, type DS5A serial nr. 43975 (Hydrolab SN44909).

The electric field strength and current were measured with a CWT-current probe mentioned above before applying the stimuli to the animals. These measurements were taken either at the spots where animals were to be placed or from within the small bins used to fixate the mobile species at the location of exposure to ensure that values were representative for the *in situ* condition.

A pulse simulator consisting of two parallel electrodes with conductors was used to represent the stimulus in the fishing gear (*Figure 3*, see also Van Marlen et al., 2007).

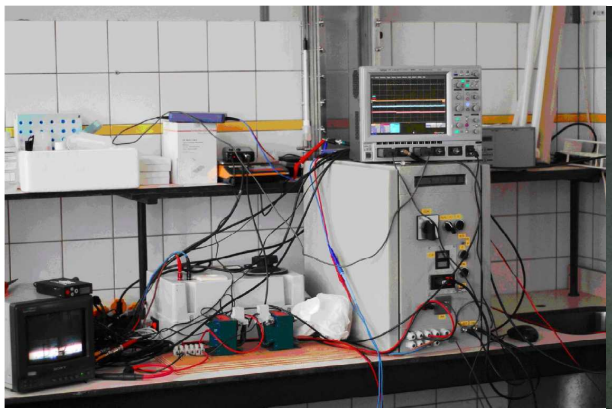


Figure 3: Pulse generator and oscilloscope used.

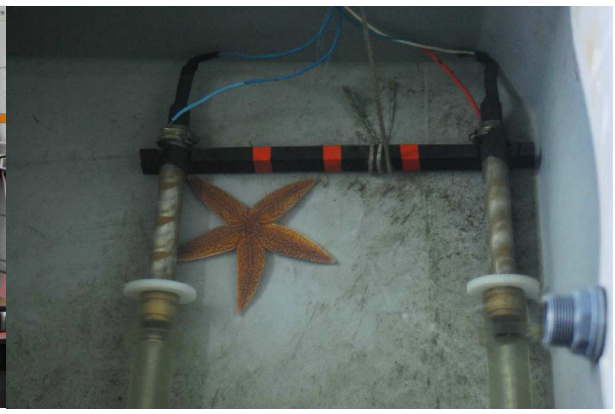


Figure 4: Experimental tank with two electrodes and a common sea star in the "near field" position.

4.2 Methods of applying stimuli

The following species were tested: ragworm, common prawn, subtruncate surf clam, European green crab, common starfish, and Atlantic razor clam (see *Figure 1*). Individual specimens were exposed to a pulse train of four pulses of 1 s duration at constant amplitude with a time interval of 1 s in between in either one of three different distance ranges (called "near field", "medium field" and "far field") from the conductor elements of the electrode pair. A 1 s pulse burst at constant amplitude is deemed to represent the passage of the pulse field of the gear beneath non-moving fish (Van Marlen et al., 2007).

Given the result of the preliminary study of invertebrates exposed in close range it was decided to locate the mobile species at certain positions from the conductors and not let them be able to move freely around the tank. Surf clams, starfish and razor clams were put on the bottom of the experimental tank in the required positions. Ragworms, and crabs were put into a small plastic container in the electric field to keep them at the position of exposure. Prawns were put in a PET bottle in the electric field to enable vertical movements, but keep them also at the position of exposure.

The animals were divided in four groups of 20 specimens to test the effects in the three treatments (distance ranges). A fourth group was kept as control reference and not electrically exposed. These animals were only subdued to the transferring operation in a similar manner to enable discrimination of the effects resulting from transfers from effects caused by exposure by the electrical stimuli.

The three exposure categories were:

1. A "near field" distance range of 0.10-0.20 m from a single conductor element with the highest field strength.
2. A "medium field" distance range of 0.20-0.30 m above the centre of the conductors with medium to low field strength.
3. A "far field" distance range of 0.40 m beside a conductor element with very low field strength.

Bins and jars containing animals were labeled as follows using tie ribbons:

- Near field: 1
- Medium field: 2
- Far field: 3
- Control: 4

A first series of exposures were conducted on 02/07/2009 on European green crab and razor clam in the "near field" position. These observations showed good food intake and burying behaviour after exposure.

Then the required number of invertebrates (20 specimens for each treatment group) was taken from the stock and transferred to one of the experimental tank for follow-up tests (*Figure 2*).

A second series of trials was carried out in the period between 06/07/2009 to 10/07/2009, followed by monitoring of survival and observation of food intake and behaviour until 24/07/2009.

4.3 Observations under stimuli

During exposure video recordings were made of the reactions of each individual. Behaviour was observed and scored on paper lists for each species at short intervals of time.

4.4 Observations after applying the stimuli

Mortality, food intake, and comments on behaviour were scored during the monitoring period for the range of species tested in the three exposure categories (far, medium, and near field), and of the control group in two weeks following the exposure (Table 14, Table 15, Table 16, Table 17, Table 18, Table 19). The survival numbers were calculated from the observed mortality and plotted in graphs (*Figure 5 - Figure 7*). The same was done for percentages of food intake (*Figure 8 - Figure 10*).

5 Results

5.1 Survival

5.1.1 Effect of exposure lumped vs. control

The first important question is: "does the exposure to electrical pulses affect the survival of the animals?"

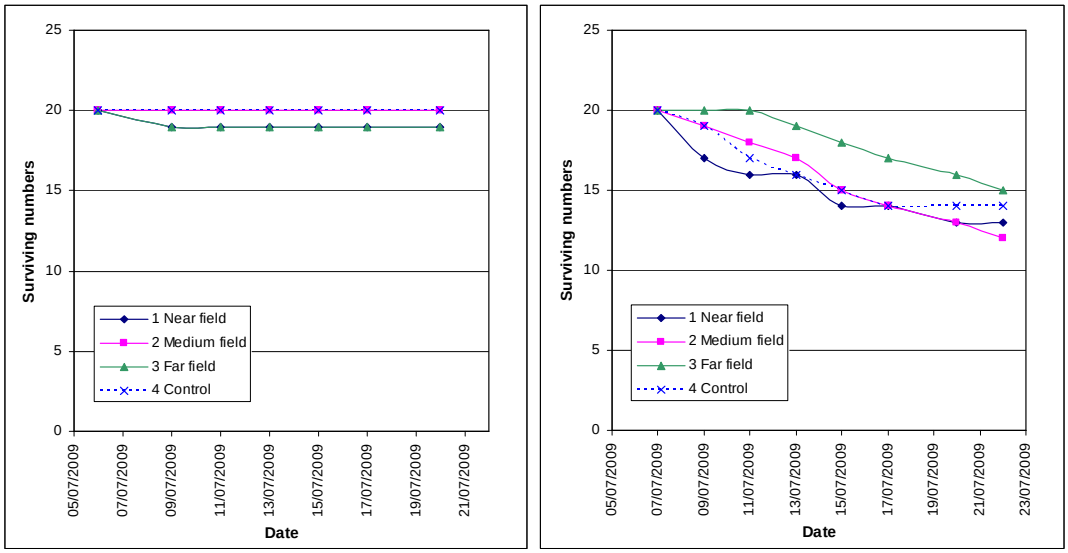


Figure 5: Survival vs. time of observation for ragworm (left) and common prawn (right) for the three exposures (near, medium and far), and the control group.

Only the far field exposure resulted in a somewhat lower survival for ragworm, while individuals of this species in the control group and in the other exposure distances all survived (*Figure 5*, left). The survival of all exposures of common prawn dropped with time most for the near field group and least for the far field group, but the same applied to the control group (*Figure 5*, right).

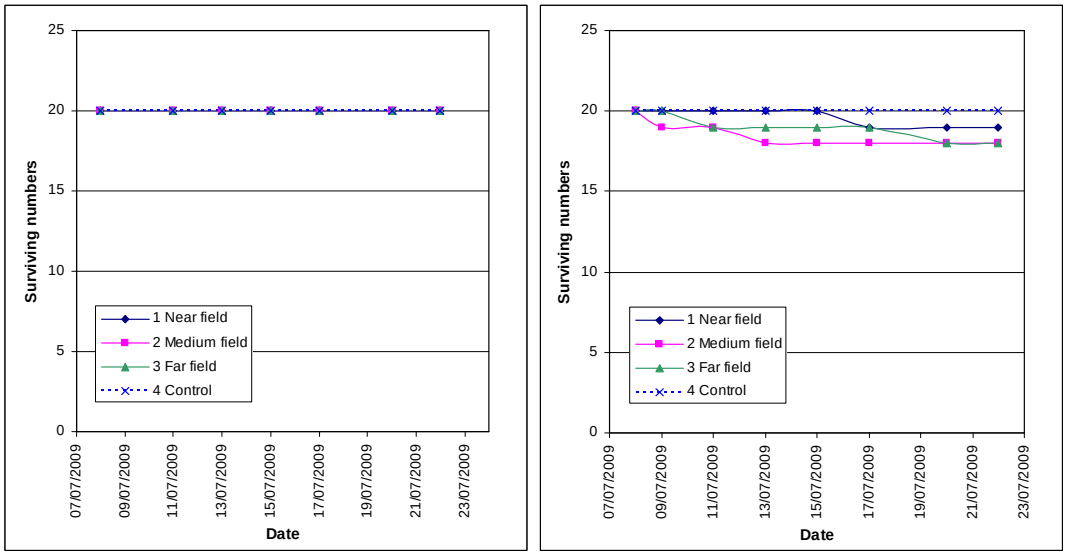


Figure 6: Survival vs. time of observation for surf clam (left) and European green crab (right) for the three exposures (near, medium and far), and the control group.

All surf clam survived irrespective of treatment (*Figure 6*, left). There was a slight drop in survival with time for green crab, but in the control group all survived (*Figure 6*, right).

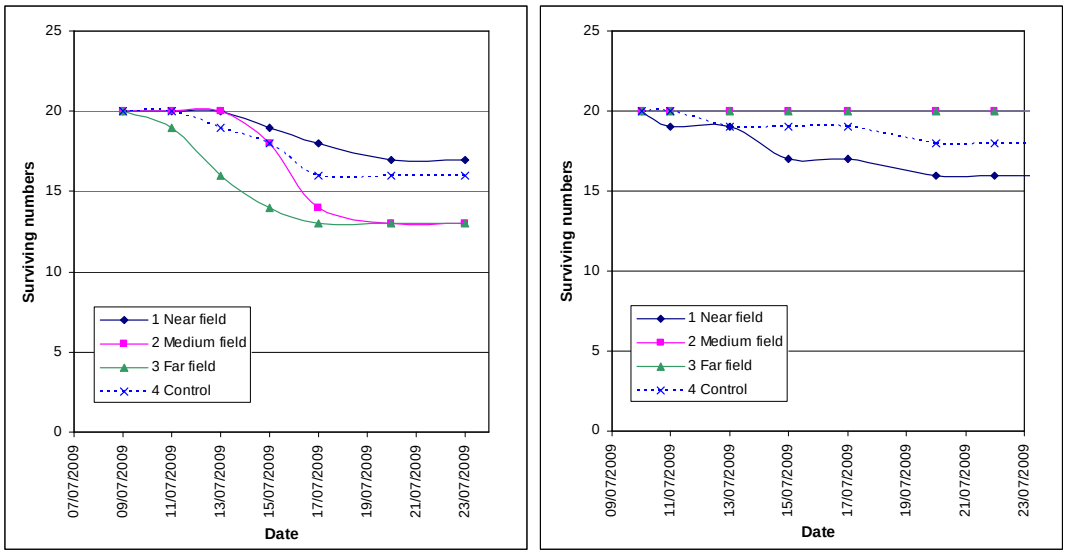


Figure 7: Survival vs. time of observation for common starfish (left) and Atlantic razor clam (right) for the three exposures (near, medium and far), and the control group.

The starfish showed the lowest survival, with the far field exposure yielding the worst result. The control group, however, also showed a decreased survival (*Figure 7*, left). The near field exposure seemed to affect the razor clam, but here too the survival of the control group was negatively affected (*Figure 7*, right).

5.1.2 Statistical analysis of lumped exposure vs. control for survival.

We have tested the dataset using a generalized linear model in the SAS Statistical Package v. 6.1 (PROC GLM). The first test was to see whether there are differences between the control groups and the exposed groups lumped together, that is without looking into details about the exposure.

We used the model:

Surviving numbers = treatment,

with values control or exposed to the electrical stimulus.

Only two cases showed significant differences, i.e. ragworm (~3% lower survival in the exposed group) and green crab (~5% lower survival in the exposed group). For common prawn, surf clam, and razor clam no significant difference was found and the survival was not lower, this was only the case for common starfish, but this result was also non-significant (*Table 1*).

Table 1: Output of SAS Procedure GLM for survival, all exposures lumped together compared to control group. Lower CL= lower confidence limit; LSMean = least square mean; UpperCL= upper confidence limit, Ratio E/C LSMean = LSMean control/LSMean exposed in %.

Effect	Species	Stimulus	Dependent	Treatment	LowerCL	LSMean	UpperCL	Probt	Sign.	Ratio E/C LSMean
Treatment	ragworm	All combined	Survivors	control	19.65	20.00	20.35	**0.0067**	s.	**97.1%**
Treatment	ragworm		Survivors	exposed	19.23	19.43	19.63			
Treatment	prawn	All combined	Survivors	control	14.27	16.13	17.98	0.7238	n.s.	102.3%
Treatment	prawn		Survivors	exposed	15.43	16.50	17.57			
Treatment	surf clam	All combined	Survivors	control	20.00	20.00	20.00		n/a	100.0%
Treatment	surf clam		Survivors	exposed	20.00	20.00	20.00			
Treatment	green crab	All combined	Survivors	control	19.49	20.00	20.51	**0.0023**	s.	**95.2%**
Treatment	green crab		Survivors	exposed	18.75	19.04	19.34			
Treatment	starfish	All combined	Survivors	control	15.74	17.86	19.97	0.4773	n.s.	95.2%
Treatment	starfish		Survivors	exposed	15.78	17.00	18.22			
Treatment	razor clam	All combined	Survivors	control	17.89	18.88	19.86	0.6055	n.s.	101.5%
Treatment	razor clam		Survivors	exposed	18.60	19.17	19.74			

Then we tested the comparison of the exposed groups split over the exposures: near field, medium field and far field with the control group.

Table 2: Output of SAS Procedure GLM for survival, near field exposure compared to control group. Lower CL= lower confidence limit; LSMean = least square mean; UpperCL= upper confidence limit, Ratio E/C LSMean = LSMean control/LSMean exposed in %.

Effect	Species	Stimulus	Dependent	Treatment	LowerCL	LSMean	UpperCL	Probt	Sign.	Ratio E/C LSMean
Treatment	ragworm	Near field	Survivors	control	19.78	20.00	20.22	**0.0001**	s.	**95.7%**
Treatment	ragworm		Survivors	nearfield	18.92	19.14	19.36			
Treatment	prawn	Near field	Survivors	control	14.33	16.13	17.92	0.5373	n.s.	95.3%
Treatment	prawn		Survivors	nearfield	13.58	15.38	17.17			
Treatment	surf clam	Near field	Survivors	control	20.00	20.00	20.00		n/a	100.0%
Treatment	surf clam		Survivors	nearfield	20.00	20.00	20.00			
Treatment	green crab	Near field	Survivors	control	19.72	20.00	20.28	0.0596	n.s.	98.1%
Treatment	green crab		Survivors	nearfield	19.35	19.63	19.90			
Treatment	starfish	Near field	Survivors	control	16.51	17.86	19.21	0.3476	n.s.	104.8%
Treatment	starfish		Survivors	nearfield	17.36	18.71	20.07			
Treatment	razor clam	Near field	Survivors	control	17.91	18.88	19.84	**0.0494**	s.	**92.7%**
Treatment	razor clam		Survivors	nearfield	16.53	17.50	18.47			

In this comparison two cases turned out to be statistically significant, namely: ragworm (~5% lower survival in the exposed group) and razor clam (~7% lower survival in the exposed group). Common prawn and green crab showed a somewhat lower survival, and common starfish somewhat higher, but these results were not statistically significant (*Table 2*).

Table 3: Output of SAS Procedure GLM for survival, medium field exposure compared to control group. Lower CL= lower confidence limit; LSMean = least square mean; UpperCL= upper confidence limit, Ratio E/C LSMean = LSMean control/LSMean exposed in %.

Effect	Species	Stimulus	Dependent	Treatment	LowerCL	LSMean	UpperCL	Probt	Sign.	Ratio E/C LSMean
Treatment	ragworm	Med field	Survivors	control	20.00	20.00	20.00		n/a	100.0%
Treatment	ragworm		Survivors	medfield	20.00	20.00	20.00			
Treatment	prawn	Med field	Survivors	control	14.11	16.13	18.14	0.9264	n.s.	99.2%
Treatment	prawn		Survivors	medfield	13.98	16.00	18.02			
Treatment	surf clam	Med field	Survivors	control	20.00	20.00	20.00		n/a	100.0%
Treatment	surf clam		Survivors	medfield	20.00	20.00	20.00			
Treatment	green crab	Med field	Survivors	control	19.59	20.00	20.41	**0.0001**	s.	**92.5%**
Treatment	green crab		Survivors	medfield	18.09	18.50	18.91			
Treatment	starfish	Med field	Survivors	control	15.61	17.86	20.11	0.5068	n.s.	94.4%
Treatment	starfish		Survivors	medfield	14.61	16.86	19.11			
Treatment	razor clam	Med field	Survivors	control	18.43	18.88	19.32	**0.0019**	s.	**106.0%**
Treatment	razor clam		Survivors	medfield	19.55	20.00	20.45			

Now we find green crab and razor clam to differ significantly, green crab with 7.5% lower and razor clam with 6% higher survival in the exposed group. Common prawn and starfish showed a small (unsignificant) reduction, while no effect at all was found for ragworm and surf clam (*Table 3*).

Table 4: Output of SAS Procedure GLM for survival, far field exposure compared to control group. Lower CL= lower confidence limit; LSMean = least square mean; UpperCL= upper confidence limit, Ratio E/C LSMean = LSMean control/LSMean exposed in %.

Effect	Species	Stimulus	Dependent	Treatment	LowerCL	LSMean	UpperCL	Probt	Sign.	Ratio E/C LSMean
Treatment	ragworm	Far field	Survivors	control	19.78	20.00	20.22	**0.0001**	s.	**95.7%**
Treatment	ragworm		Survivors	farfield	18.92	19.14	19.36			
Treatment	prawn	Far field	Survivors	control	14.48	16.13	17.77	0.0862	n.s.	112.4%
Treatment	prawn		Survivors	farfield	16.48	18.13	19.77			
Treatment	surf clam	Far field	Survivors	control	20.00	20.00	20.00		n/a	100.0%
Treatment	surf clam		Survivors	farfield	20.00	20.00	20.00			
Treatment	green crab	Far field	Survivors	control	19.59	20.00	20.41	**0.0022**	s.	**95.0%**
Treatment	green crab		Survivors	farfield	18.59	19.00	19.41			
Treatment	starfish	Far field	Survivors	control	15.80	17.86	19.91	0.0934	n.s.	86.4%
Treatment	starfish		Survivors	farfield	13.38	15.43	17.48			
Treatment	razor clam	Far field	Survivors	control	18.43	18.88	19.32	**0.0019**	s.	**106.0%**
Treatment	razor clam		Survivors	farfield	19.55	20.00	20.45			

Statistically significant were the lower survival for ragworm (~5% lower survival in the exposed group), and green crab (~5% lower survival in the exposed group), and the 6% higher survival for razor clam in the exposed group. Higher survival was found for common prawn, lower for common starfish, while no effect at all for surf clam, but all these results were statistically non-significant (*Table 4*).

The signal arising from field strength is not very clear, and the near field exposure does not score very much worse. A slight drop in survival is restricted to ragworm and green crab, as was found before.

5.2 Food intake

A second question is: “does the exposure to the electric pulses affect the intake of food by the animals?”

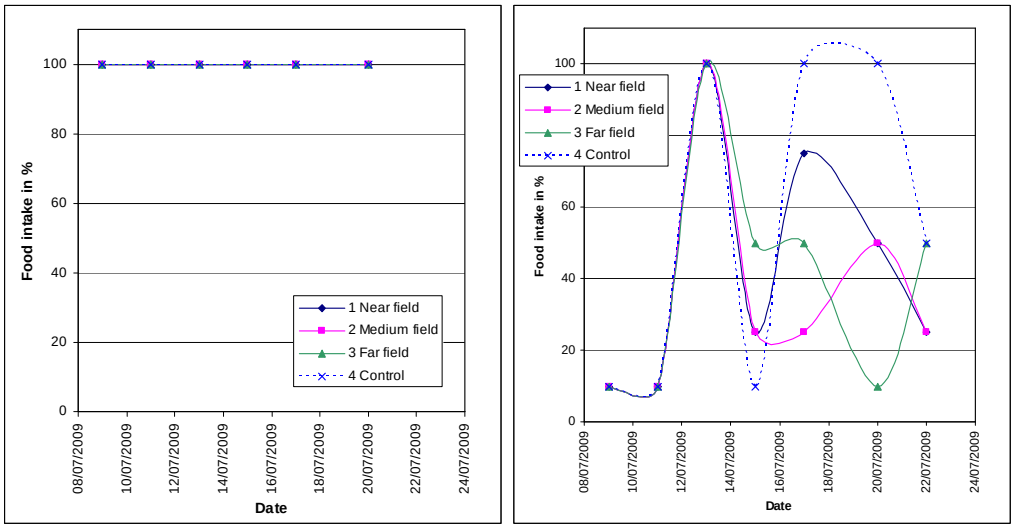


Figure 8: Food intake vs. time of observation for ragworm (left) and common prawn (right) for the three exposures (near, medium and far), and the control group.

Ragworm showed a 100% food intake, irrespective of the treatment (exposure or not, *Figure 8*, left), and prawn showed strong fluctuations in food intake (*Figure 8*, right).

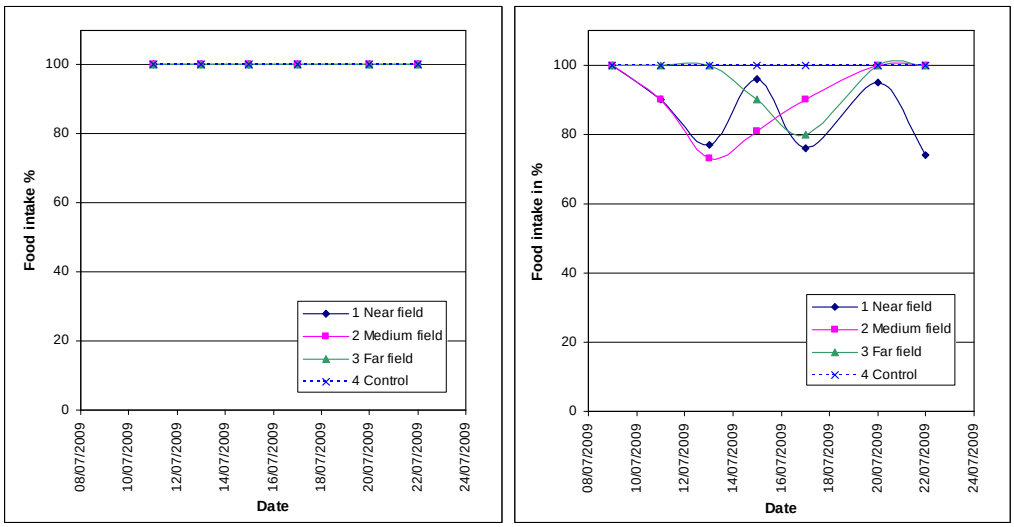


Figure 9: Food intake vs. time of observation for surf clam (left) and European green crab (right) for the three exposures (near, medium and far), and the control group.

Surf clam also showed a 100% food uptake (*Figure 9*, left), and green crab showed great variability in food uptake in the exposure treatments, while the control group took all food (*Figure 9*, right).

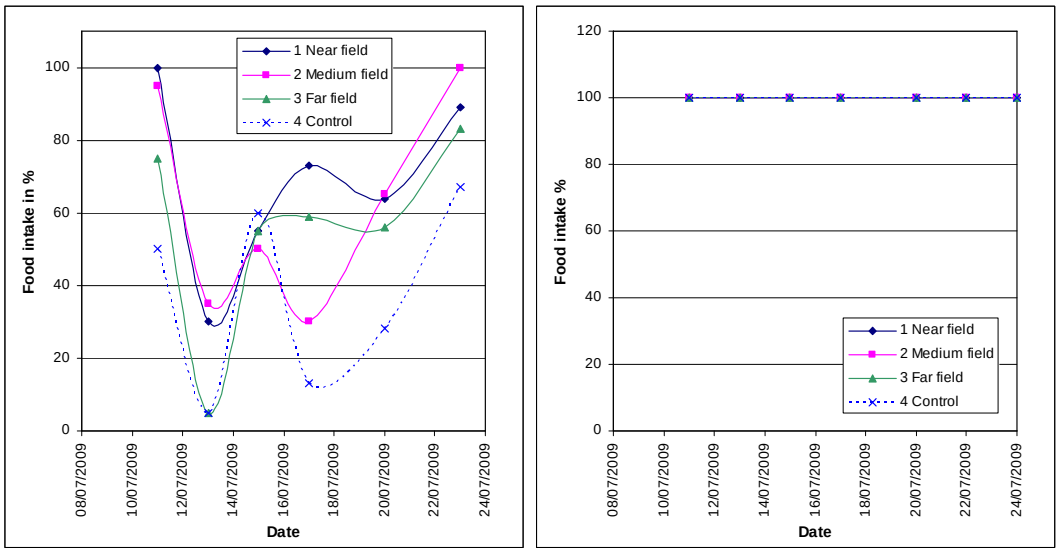


Figure 10: Food intake vs. time of observation for common starfish (left) and Atlantic razor clam (right) for the three exposures (near, medium and far), and the control group.

A fluctuating picture for both the exposed groups and the control group was found for starfish (*Figure 10*, left), while razor clam demonstrated a 100% food uptake (*Figure 10*, right).

5.2.1 Statistical analysis of lumped exposure vs. control for food intake

We have tested the dataset using again the generalized linear model in SAS (PROC GLM). The first test was to see whether there are differences between the control groups and the exposed groups lumped together, that is without looking into details about the exposure.

In this case we used the following model:

Food intake in % = treatment, with values control or exposed to the electrical stimulus.

Table 5: Output of SAS Procedure GLM for food intake, all exposures lumped together compared to control group. Lower CL= lower confidence limit; LSMean = least square mean; UpperCL= upper confidence limit, Ratio E/C LSMean = LSMean control/LSMean exposed in %.

Effect	Species	Stimulus	Dependent	Treatment	LowerCL	LSMean	UpperCL	Probt	Sign.	Ratio E/C LSMean
Treatment	ragworm	All combined	Feeding	control	100.00	100.00	100.00		n/a	100.0%
Treatment	ragworm		Feeding	exposed	100.00	100.00	100.00			
Treatment	prawn	All combined	Feeding	control	26.99	54.29	81.59	0.3296	n.s.	71.9%
Treatment	prawn		Feeding	exposed	23.29	39.05	54.81			
Treatment	surf clam	All combined	Feeding	control	100.00	100.00	100.00		n/a	100.0%
Treatment	surf clam		Feeding	exposed	100.00	100.00	100.00			
Treatment	green crab	All combined	Feeding	control	93.14	100.00	106.86	**0.0283**	s.	**91.0%**
Treatment	green crab		Feeding	exposed	87.09	91.05	95.01			
Treatment	starfish	All combined	Feeding	control	15.03	37.17	59.30	0.0548	n.s.	167.3%
Treatment	starfish		Feeding	exposed	49.39	62.17	74.95			
Treatment	razor clam	All combined	Feeding	control	100.00	100.00	100.00		n/a	100.0%
Treatment	razor clam		Feeding	exposed	100.00	100.00	100.00			

Of all species tested, only green crab showed a significant difference between the control and the exposure treatments, with a 9% lower food intake in the exposed group. This signal coincides with the higher survival found. For all the other species no significant differences were found between control and lumped treatment groups. No effect at all was found for ragworm, surf clam and razor clam. Lower food intake was found for common prawn and higher for common starfish, but these latter results were statistically non-significant (*Table 5*).

Again we tested the comparison of the exposed groups split over the exposures: near field, medium field and far field with the control group.

Table 6: Output of SAS Procedure GLM for food intake, near field exposure compared to control group. Lower CL= lower confidence limit; LSMean = least square mean; UpperCL= upper confidence limit, Ratio E/C LSMean = LSMean control/LSMean exposed in %.

Effect	Species	Stimulus	Dependent	Treatment	LowerCL	LSMean	UpperCL	Probt	Sign.	Ratio E/C LSMean
Treatment	ragworm	Near field	Feeding	control	100.00	100.00	100.00		n/a	100.0%
Treatment	ragworm		Feeding	nearfield	100.00	100.00	100.00			
Treatment	prawn	Near field	Feeding	control	21.25	54.29	87.32	0.5817	n.s.	77.6%
Treatment	prawn		Feeding	nearfield	9.10	42.14	75.18			
Treatment	surf clam	Near field	Feeding	control	100.00	100.00	100.00		n/a	100.0%
Treatment	surf clam		Feeding	nearfield	100.00	100.00	100.00			
Treatment	green crab	Near field	Feeding	control	93.65	100.00	106.35	**0.0078**	n.s.	**86.9%**
Treatment	green crab		Feeding	nearfield	80.51	86.86	93.20			
Treatment	starfish	Near field	Feeding	control	14.16	37.17	60.17	0.0575	n.s.	184.3%
Treatment	starfish		Feeding	nearfield	45.49	68.50	91.51			
Treatment	razor clam	Near field	Feeding	control	100.00	100.00	100.00		n/a	100.0%
Treatment	razor clam		Feeding	nearfield	100.00	100.00	100.00			

For the near field exposure compared to the control group the reduction in food intake (~13% less) for green crab stands out even stronger, while all other species showed no statistically significant effects, again with no effect at all was found for ragworm, surf clam and razor clam, and lower food intake for common prawn and higher for common starfish (*Table 6*).

Table 7: Output of SAS Procedure GLM for food intake, medium field exposure compared to control group. Lower CL= lower confidence limit; LSMean = least square mean; UpperCL= upper confidence limit, Ratio E/C LSMean = LSMean control/LSMean exposed in %.

Effect	Species	Stimulus	Dependent	Treatment	LowerCL	LSMean	UpperCL	Probt	Sign.	Ratio E/C LSMean
Treatment	ragworm	Med field	Feeding	control	100.00	100.00	100.00		n/a	100.0%
Treatment	ragworm		Feeding	medfield	100.00	100.00	100.00			
Treatment	prawn	Med field	Feeding	control	22.24	54.29	86.33	0.3721	n.s.	64.5%
Treatment	prawn		Feeding	medfield	2.95	35.00	67.05			
Treatment	surf clam	Med field	Feeding	control	100.00	100.00	100.00		n/a	100.0%
Treatment	surf clam		Feeding	medfield	100.00	100.00	100.00			
Treatment	green crab	Med field	Feeding	control	93.86	100.00	106.14	**0.0357**	n.s.	**90.6%**
Treatment	green crab		Feeding	medfield	84.43	90.57	96.71			
Treatment	starfish	Med field	Feeding	control	11.90	37.17	62.43	0.1453	n.s.	168.2%
Treatment	starfish		Feeding	medfield	37.23	62.50	87.77			
Treatment	razor clam	Med field	Feeding	control	100.00	100.00	100.00		n/a	100.0%
Treatment	razor clam		Feeding	medfield	100.00	100.00	100.00			

Again only a significant reduction for green crab with ~10% less food intake, with no effect at all for ragworm, surf clam and razor clam, lower food intake for common prawn and higher for common starfish (*Table 7*).

Table 8: Output of SAS Procedure GLM for food intake, far field exposure compared to control group. Lower CL= lower confidence limit; LSMean = least square mean; UpperCL= upper confidence limit, Ratio E/C LSMean = LSMean control/LSMean exposed in %.

Effect	Species	Stimulus	Dependent	Treatment	LowerCL	LSMean	UpperCL	Probt	Sign.	Ratio E/C LSMean
Treatment	ragworm	Med field	Feeding	control	100.00	100.00	100.00		n/a	100.0%
Treatment	ragworm		Feeding	farfield	100.00	100.00	100.00			
Treatment	prawn	Med field	Feeding	control	21.71	54.29	86.86	0.5120	n.s.	73.7%
Treatment	prawn		Feeding	farfield	7.43	40.00	72.57			
Treatment	surf clam	Med field	Feeding	control	100.00	100.00	100.00		n/a	100.0%
Treatment	surf clam		Feeding	farfield	100.00	100.00	100.00			
Treatment	green crab	Med field	Feeding	control	95.42	100.00	104.58	0.1751	n.s.	95.7%
Treatment	green crab		Feeding	farfield	91.13	95.71	100.30			
Treatment	starfish	Med field	Feeding	control	13.14	37.17	61.20	0.2570	n.s.	149.3%
Treatment	starfish		Feeding	farfield	31.47	55.50	79.53			
Treatment	razor clam	Med field	Feeding	control	100.00	100.00	100.00		n/a	100.0%
Treatment	razor clam		Feeding	farfield	100.00	100.00	100.00			

None of the differences is significant in the far field exposure, but the tendency is the same as in the medium field exposure, resulting in a reduction for green crab, no effect at all for ragworm, surf clam and razor clam, lower food intake for common prawn and higher for common starfish (*Table 8*).

5.3 Behaviour

The behaviour of the six different species during the exposure in the three distance ranges (near, medium and far field) was recorded on log sheets for the four pulses that every individual animal received.

The strongest reactions were seen in prawn and common crab, and weaker reactions in rag worm and razor clam. Prawns jump up when stimulated, crab usually stiffens and razors can exhibit quite strong reactions using their foot and siphon, often enough to propel them away. A graphical representation of behaviour is given in *Table 9 - Table 12* below, and detailed descriptions are given in Table 20 - Table 37 in Appendix A. Generally the near field reactions are strongest, followed by the medium and far field ranges. No reactions were seen in surf clam and starfish irrespective of exposure. These species were not graphically represented therefore.

Table 9: Graphical representation of behaviour of individual ragworms, strongest behaviour is darkest.

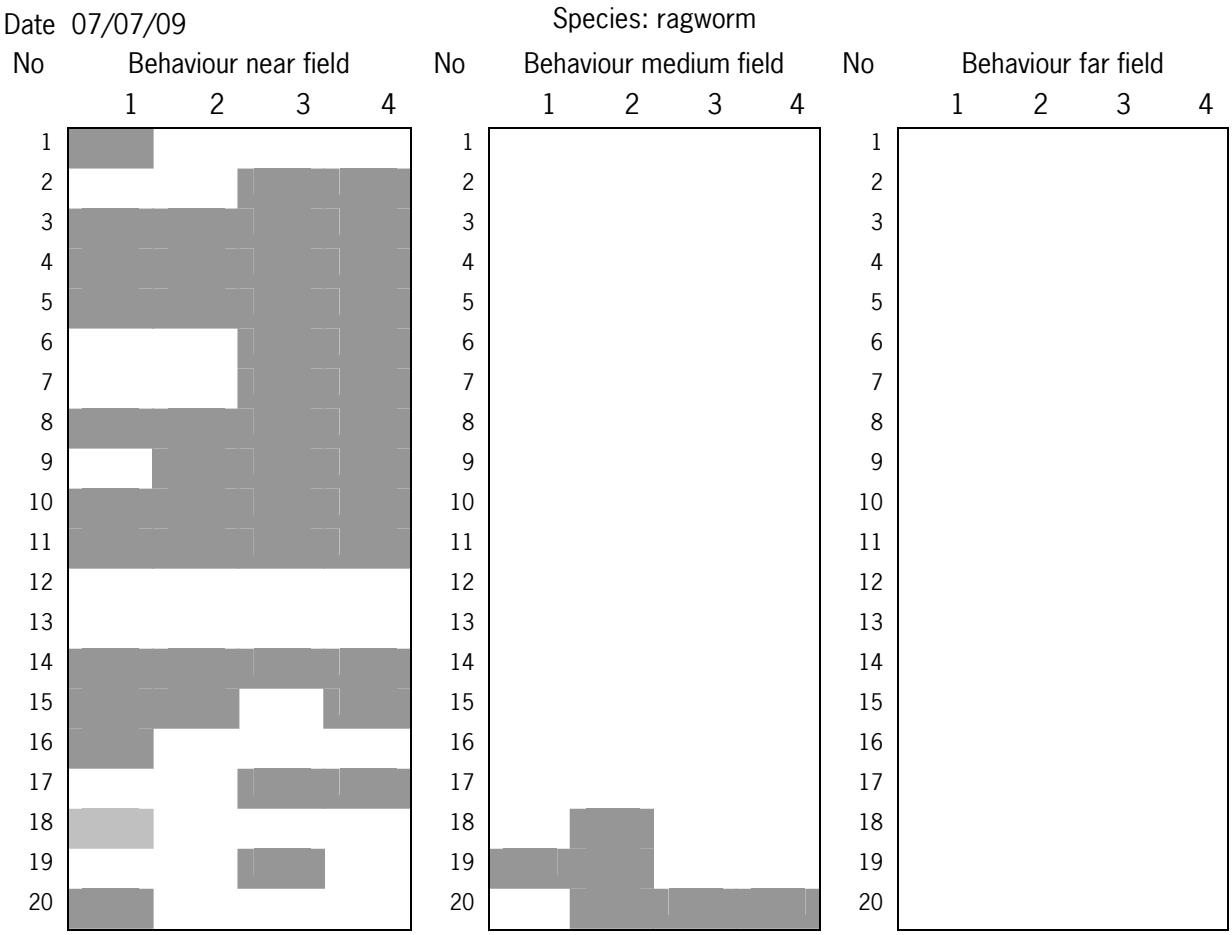


Table 10: Graphical representation of behaviour of individual common prawns, strongest behaviour is darkest.

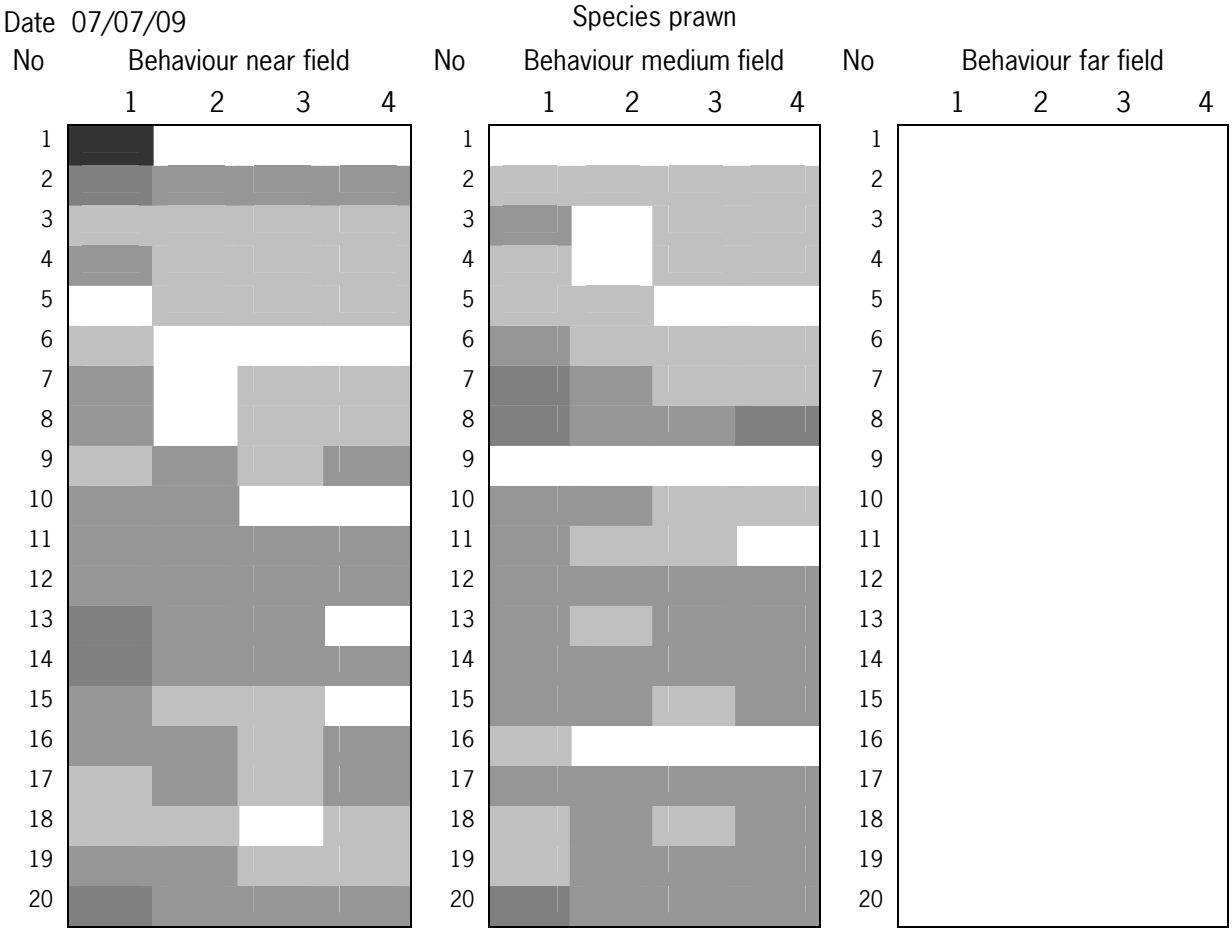
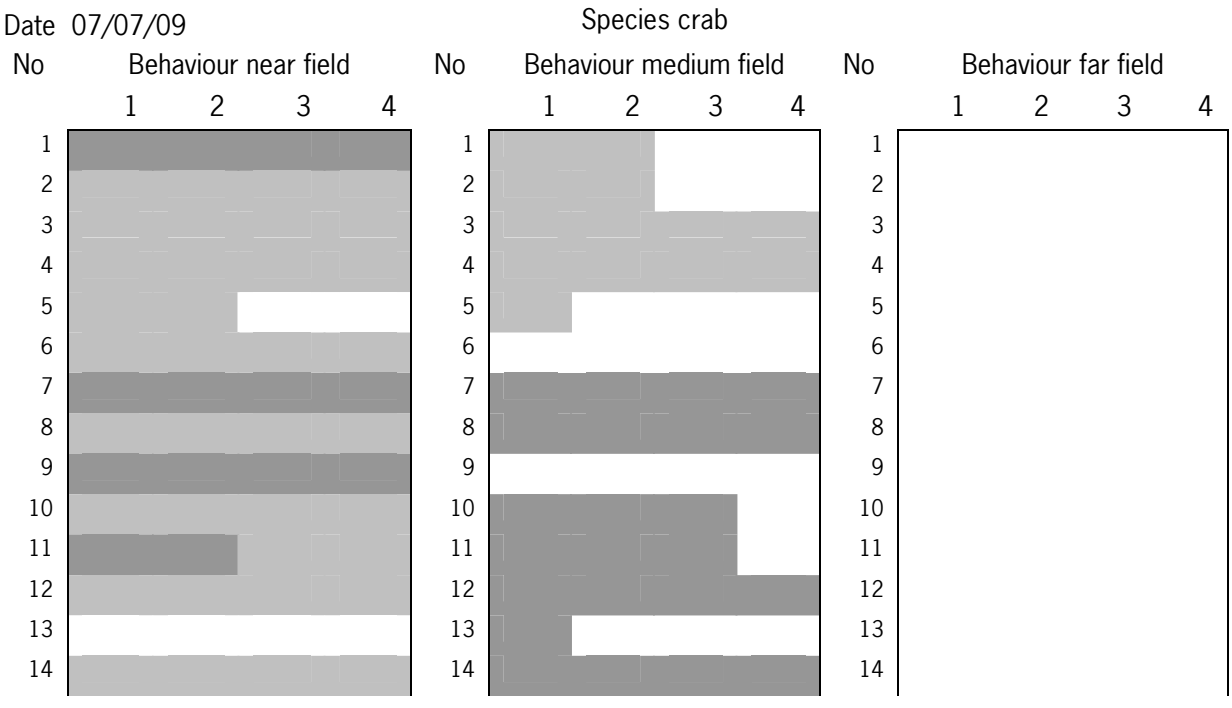


Table 11: Graphical representation of behaviour of individual green crabs, strongest behaviour is darkest.



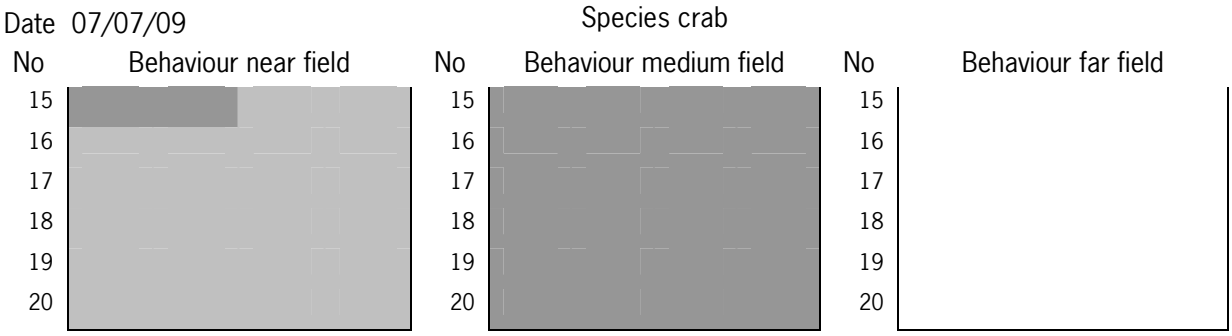
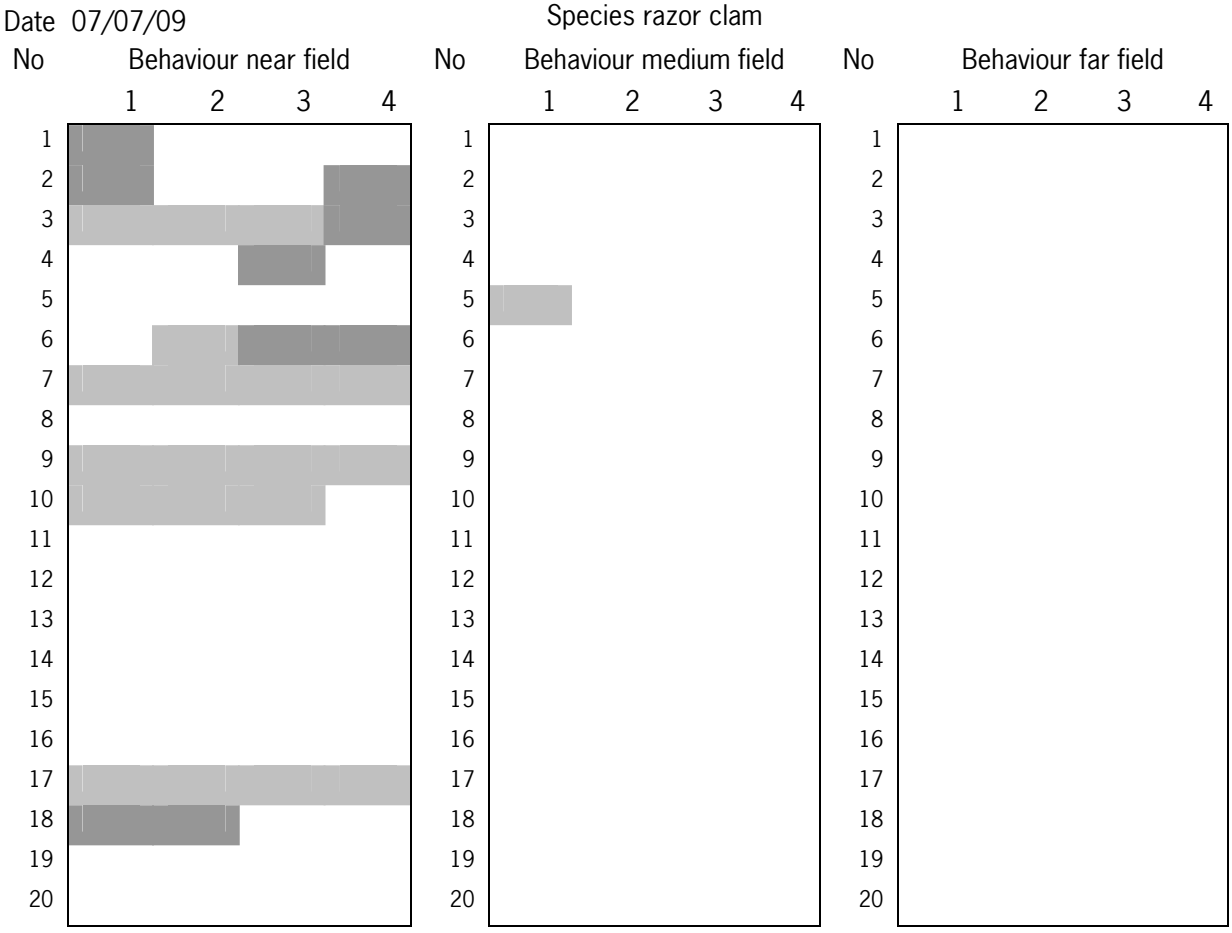


Table 12: Graphical representation of behaviour of individual razor clams, strongest behaviour is darkest.



6 Discussion

This work addresses some of the critiques on the earlier preliminary study. For each species a total of 20 individuals were exposed to three treatments (three distances) of electrical pulses: close (0.10-0.20 m), medium (0.20-0.30 m) and far (>0.40 m). A control group that received the same treatment as the test groups, but without exposure to electrical pulses, was used to correct for husbandry conditions. The number of individuals we chose is based on experience in testing animals, for which experts mentioned to use a range of 20-25 individuals in case the variance is not known (*pers. comm. Dr. J.W. van der Vis, IMARES*). In addition the water quality was regularly monitored. The stimulus was chosen to represent the in situ exposure in a beam trawl using the pulse technique of Verburg-Holland.

The study showed some effects, but not in all species to the same magnitude. The reactions and effects were not very strong and for the species studied and can be compared similar to the one found by Polet (2004) and Polet et al., (2005). Not all of the species we tested live on the sea bed, some usually spend considerable time buried in the sediment (ragworm, surf clam and razor clam). We tested the animals lying on the bottom of the tank or placed inside a container put on the bottom. In the case of a gear passage animals buried in the sediment will not receive a stimulus as high as when placed under our test conditions as the electric field does not penetrate the sediment and the effect will be smaller.

One should compare these effects of pulse trawling with those of conventional beam trawls, which affect the marine ecosystem by favouring short lived over long lived species (Lindeboom and de Groot, 1998). Pulse beam trawling was shown to result in significantly 40-50% lower bycatches of benthic invertebrates, and gave indications ($p = 0.09$) of lower direct trawl path mortality for many species compared to conventional tickler chain beam trawling (median for 15 abundant benthic species: 24% for the pulse trawl vs. 36% for the conventional trawl, see Van Marlen et al., 2001).

We found mortalities that were only 3-7% higher for ragworm, green crab and razor clam due to these relatively high stimuli, and hardly no effect for other species. The simulator was based on a 1 s pulse burst representing the passage of the pulse field of the gear beneath non-moving fish (Van Marlen et al., 2007), and here we used a sequence of four times 1 s pulse bursts at constant amplitude with 1 s time lapse in-between. Food intake and behaviour recovered after exposure (Table 14 - Table 19). We believe that it is therefore plausible that the effects of pulse beam trawling, as simulated in this study, are far smaller than the effects of conventional beam trawling.

7 Conclusions

Exposure to pulse stimuli as used in the Verburg-Holland system does not seem to severely affect the benthic species tested (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.)).

The survival of three species was lowered by 3-7%, i.e. ragworm, green crab and razor clam. Food intake might be affected as found for green crab (lowered by 5-15%) which may have contributed to their lower survival. The other species (common prawn, surf clam and starfish) showed no significant effects (*Table 13*).

Surf clam and starfish did not show any behavioural reaction at all, they did not move. The other species showed very low responses in the far field exposure range. In the medium and near field ranges the reactions were stronger. The behaviour depends on species, prawn often jump up, crab stiffens; ragworm gave jerky movement as response, while razor clam sometimes uses its foot and siphon to move away.

Conventional beam trawling with heavy tickler chains affect the marine ecosystem by favouring short lived over long lived species (Lindeboom and de Groot, 1998). Pulse beam trawling was shown to result in significantly lower bycatches of benthic invertebrates compared to conventional tickler chain beam trawling, and gave indications of lower trawl path mortality (24% vs. 36% median) for 15 abundant benthic species (Van Marlen et al., 2001).

Mortality increase, if at all, was low (3-7% for ragworm, green crab and razor clam), and food intake and behaviour recovered after exposure. It is therefore plausible that the effects effects of pulse beam trawling, as simulated in this study, are are far smaller than the effects of conventional beam trawling.

Table 13: Indicative summary of effects of electrical pulses on the species tested

Species/Effect	Mortality	Food intake	Behaviour
Ragworm	++	-	+
Common prawn	+	+	++
Subtruncate surf clam	-	-	-
European green crab	++	++	++
Common starfish	+	+	-
Atlantic razor clam	++	-	+

++ = noticeable effect; + = indicative effect; - = no effect

8 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. 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. The contents of this report were reviewed by dr. Oscar Bos.

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Justification

Rapport C103/09
Project Number: 430.15000.01

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

Approved: dr. O.G. Bos
Researcher

Signature: 

Date: 23 October 2009

Approved: dr. ir. T.P. Bult
Head of Fisheries Department

Signature: b/a 

Date: 23 October 2009

Number of copies: 15
Number of pages 53
Number of tables: 37
Number of graphs: 10
Number of appendix attachments: 1

Appendix A. Additional tables

Table 14: Scores of mortality, food intake, and comments on behaviour during the monitoring period for common prawn. Each group started with 20 individuals.

	1 Near field				2 Medium field				3 Far field				4 Control			
Date x/07/2009	Dead	% food cons.	Food	Behaviour	Dead	% food cons.	Food	Behaviour	Dead	% food cons.	Food	Behaviour	Dead	% food cons.	Food	Behaviour
7 start	0		2 half mussels	lively, jumpy	0		2 half mussels	lively, jumpy	0		2 half mussels	lively, jumpy	0		2 half mussels	lively, jumpy
9	3	<10	No only replaced. 2 half mussels	lively, jumpy	1	<10	No only changed 2 half mussels	lively, jumpy	0	<10	No only changed 2 half mussels	lively, jumpy	1	<10	No only changed 2 half mussels	lively, jumpy
11	1	<10	No	lively, jumpy	1	<10	No	lively, jumpy	0	<10	No	lively, jumpy	2	<10	No	lively, jumpy
13	0	100	2 half mussels	lively	1	100	2 half mussels	Lively	1	100	2 half mussels	lively	1	100	2 half mussels	lively
15	2	25	No	lively	2	25	No	Lively	1	50	2 half mussels	lively	1	<10	No	lively
17	0	75	2 half mussels	lively	1	25	2 half mussels	Lively	1	50	2 half mussels	lively	1	100	2 half mussels	lively
20	1	50	2 half mussels	lively	1	50	2 half mussels	Lively	1	<10	2 half mussels	lively	0	100	2 half mussels	lively
22 (last day)	0	25	-	lively	1	25	-	Lively	1	50	-	lively	0	50	-	lively

Table 15: Scores of mortality, food intake, and comments on behaviour during the monitoring period for Atlantic razor clam. Each group started with 20 individuals.

	1 Near field				2 Medium field				3 Far field				4 Control			
Date x/07/2009	Dead	food cons. indicative	Food	Behaviour	Dead	food cons. indicative	Food	Behaviour	Dead	food cons. indicative	Food	Behaviour	Dead	food cons. indicative	Food	Behaviour
10 start	0		Algae on sediment	Burying quickly	0		Algae on sediment	Burying quickly	0		Algae on sediment	Burying quickly	0		Algae on sediment	Burying quickly
11	**1**, Resting on sediment	Algae eaten around siphon	Algae on sediment	Inside sediment, 1 Resting on sediment	0	Algae eaten around siphon	Algae on sediment	Inside sediment, Algae eaten around siphon.	0	Algae eaten around siphon	Algae on sediment	Inside sediment, Algae eaten around siphon.	0	Algae eaten around siphon	Algae on sediment	Inside sediment, Algae eaten around siphon.
13	0	Algae eaten around siphon	Algae on sediment	Inside sediment, 2 Resting on sediment	0	Algae eaten around siphon	Algae on sediment	Inside sediment, Algae eaten around siphon.	0	Algae eaten around siphon	Algae on sediment	Inside sediment, Algae eaten around siphon.	**1**, Resting on sediment	Algae eaten around siphon	Algae on sediment	Inside sediment, Algae eaten around siphon.
15	2	Algae eaten around siphon	Algae on sediment	Inside sediment, 1 Resting on sediment	0	Algae eaten around siphon	Algae on sediment	Inside sediment, Algae eaten around siphon.	0	Algae eaten around siphon	Algae on sediment	Inside sediment, Algae eaten around siphon.	0	Algae eaten around siphon	Algae on sediment	Inside sediment, Algae eaten around siphon.
17	0	Algae eaten around siphon	Algae on sediment	Inside sediment, 1 Resting on sediment	0	Algae eaten around siphon	Algae on sediment	Inside sediment, Algae eaten around siphon.	0	Algae eaten around siphon	Algae on sediment	Inside sediment, Algae eaten around siphon.	0	Algae eaten around siphon	Algae on sediment	Inside sediment, Algae eaten around siphon.
20	0	Algae eaten around siphon	Algae on sediment	Inside sediment, 1 Resting on sediment	0	Algae eaten around siphon	Algae on sediment	Inside sediment, Algae eaten around	0	Algae eaten around siphon	Algae on sediment	Inside sediment, Algae eaten around	0	Algae eaten around siphon	Algae on sediment	Inside sediment, Algae eaten around

	1 Near field				2 Medium field				3 Far field				4 Control			
								siphon.				siphon.				siphon.
22	1	Algae eaten around siphon	Algae on sediment	Inside sediment.	0	Algae eaten around siphon	Algae on sediment	Inside sediment, Algae eaten around siphon.	0	Algae eaten around siphon	Algae on sediment	Inside sediment, Algae eaten around siphon.	**1**	Algae eaten around siphon	Algae on sediment	Inside sediment, Algae eaten around siphon.
24 Last day	0	Algae eaten around siphon	Algae on sediment	Inside sediment.	0	Algae eaten around siphon	Algae on sediment	Inside sediment, Algae eaten around siphon.	0	Algae eaten around siphon	Algae on sediment	Inside sediment, Algae eaten around siphon.	0	Algae eaten around siphon	Algae on sediment	Inside sediment, Algae eaten around siphon.

Table 16: Scores of mortality, food intake, and comments on behaviour during the monitoring period for subtruncate surf clam. Each group started with 20 individuals.

	1 Near field				2 Medium field				3 Far field				4 Control			
Date x/07/2009	Dead	food cons. indicative	Food	Behaviour	Dead	food cons. indicative	Food	Behaviour	Dead	food cons. indicative	Food	Behaviour	Dead	food cons. indicative	Food	Behaviour
8 start	0		Algae on sediment	Burying	0		Algae on sediment	Burying	0		Algae on sediment	Burying	0		Algae on sediment	Burying
11	0	Algae eaten around siphon	Algae on sediment	Inside sediment, 2 resting on sediment	0	Algae eaten around siphon	Algae on sediment	Inside sediment, 1 resting on sediment	0	Algae eaten around siphon	Algae on sediment	Inside sediment.	0	Algae eaten around siphon	Algae on sediment	Inside sediment.
13	0	Algae eaten around siphon	Algae on sediment	Inside sediment, 2 resting on sediment	0	Algae eaten around siphon	Algae on sediment	Inside sediment, 1 resting on sediment	0	Algae eaten around siphon	Algae on sediment	Inside sediment.	0	Algae eaten around siphon	Algae on sediment	Inside sediment.
15	0	Algae eaten around siphon	Algae on sediment	Inside sediment,	0	Algae eaten around siphon	Algae on sediment	Inside sediment.	0	Algae eaten around siphon	Algae on sediment	Inside sediment.	0	Algae eaten around siphon	Algae on sediment	Inside sediment.
17	0	Algae eaten around siphon	Algae on sediment	Inside sediment,	0	Algae eaten around siphon	Algae on sediment	Inside sediment.	0	Algae eaten around siphon	Algae on sediment	Inside sediment.	0	Algae eaten around siphon	Algae on sediment	Inside sediment.
20	0	Algae eaten around siphon	Algae on sediment	Inside sediment,	0	Algae eaten around siphon	Algae on sediment	Inside sediment.	0	Algae eaten around siphon	Algae on sediment	Inside sediment.	0	Algae eaten around siphon	Algae on sediment	Inside sediment.
22 Last day	0	Algae eaten around siphon	Algae on sediment	Inside sediment,	0	Algae eaten around siphon	Algae on sediment	Inside sediment.	0	Algae eaten around siphon	Algae on sediment	Inside sediment.	0	Algae eaten around siphon	Algae on sediment	Inside sediment.

Table 17: Scores of mortality, food intake, and comments on behaviour during the monitoring period for European green crab. Each group started with 20 individuals.

	1 Near field				2 Medium field				3 Far field				4 Control			
Date x/07/2009	Dead	% food cons.	Food	Behaviour	Dead	% food cons.	Food	Behaviour	Dead	% food cons.	Food	Behaviour	Dead	% food cons.	Food	Behaviour
8 start	0		20 half mussels	lively, lying against each other, not touching each other	0		20 half mussels	lively, lying against each other, not touching each other	0		20 half mussels	lively, lying against each other, not touching each other	0		20 half mussels	lively, lying against each other, not touching each other
9	0	100	20 half mussels	lively, lying against each other, not touching each other	1	100	20 half mussels	lively, lying against each other, not touching each other	0	100	20 half mussels	lively, lying against each other, not touching each other	0	100	20 half mussels	lively, lying against each other, not touching each other
11	0	90	20 half mussels	lively, lying against each other, not touching each other	0	90	20 half mussels	lively, lying against each other, not touching each other	1	100	20 half mussels	lively, lying against each other, not touching each other	0	100	20 half mussels	lively, lying against each other, not touching each other
13	0	77	20 half mussels	lively, lying against each other, not touching each other	1	73	20 half mussels	lively, lying against each other, not touching each other	0	100	20 half mussels	lively, lying against each other, not touching each other	0	100	20 half mussels	lively, lying against each other, not touching each other
15	0	96	19 half mussels	lively, lying against each other, not touching each other	0	81	15 half mussels	lively, lying against each other, not touching each other	0	90	18 half mussels	lively, lying against each other, not touching each other	0	100	20 half mussels	lively, lying against each other, not touching each other
17	0	76	19 half mussels	lively, lying against	0	90	18 half mussels	lively, lying against	0	80	19 half mussels	lively, lying against each	0	100	20 half mussels	lively, lying against

	1 Near field				2 Medium field				3 Far field				4 Control			
				each other, not touching each other				each other, not touching each other				other, not touching each other				each other, not touching each other
20	1	95	19 half mussels	lively, lying against each other, not touching each other	0	100	18 half mussels	lively, lying against each other, not touching each other	0	100	19 half mussels	lively, lying against each other, not touching each other	0	100	20 half mussels	lively, lying against each other, not touching each other
22 Last day	0	74	-	lively, lying against each other, not touching each other	0	100	-	lively, lying against each other, not touching each other	1	100	-	lively, lying against each other, not touching each other	0	100	-	lively, lying against each other, not touching each other

Table 18: Scores of mortality, food intake, and comments on behaviour during the monitoring period for ragworm. Each group started with 20 individuals.

	1 Near field				2 Medium field				3 Far field				4 Control			
Date x/07/2009	Dead	% food cons.	Food	Behaviour	Dead	% food cons.	Food	Behaviour	Dead	% food cons.	Food	Behaviour	Dead	% food cons.	Food	Behaviour
6 start	0		Algae on sediment	Burying quickly	0		Algae on sediment	Burying quickly	0		Algae on sediment	Burying quickly	0		Algae on sediment	Burying quickly
9	1 Resting on sediment		20 grains. Appr. 0.30 gr.	Inside sediment. Active after feeding.	0		20 grains. Appr. 0.30 gr.	Inside sediment. Active after feeding.	1 Resting on sediment		20 grains. Appr. 0.30 gr.	Inside sediment. Active after feeding.	0		20 grains. Appr. 0.30 gr.	Inside sediment. Active after feeding.
11	0	100	20 grains. Appr. 0.30 gr.	Inside sediment. Active after feeding.	0	100	20 grains. Appr. 0.30 gr.	Inside sediment. Active after feeding.	0	100	20 grains. Appr. 0.30 gr.	Inside sediment. Active after feeding.	0	100	20 grains. Appr. 0.30 gr.	Inside sediment. Active after feeding.
13	0	100	20 grains. Appr. 0.30 gr.	Inside sediment. Active after feeding.	0	100	20 grains. Appr. 0.30 gr.	Inside sediment. Active after feeding.	0	100	20 grains. Appr. 0.30 gr.	Inside sediment. Active after feeding.	0	100	20 grains. Appr. 0.30 gr.	Inside sediment. Active after feeding.
15	0	100	20 grains. Appr. 0.30 gr.	Inside sediment. Active after feeding.	0	100	20 grains. Appr. 0.30 gr.	Inside sediment. Active after feeding.	0	100	20 grains. Appr. 0.30 gr.	Inside sediment. Active after feeding.	0	100	20 grains. Appr. 0.30 gr.	Inside sediment. Active after feeding.
17	0	100	20 grains. Appr. 0.30 gr.	Inside sediment. Active after feeding.	0	100	20 grains. Appr. 0.30 gr.	Inside sediment. Active after feeding.	0	100	20 grains. Appr. 0.30 gr.	Inside sediment. Active after feeding.	0	100	20 grains. Appr. 0.30 gr.	Inside sediment. Active after feeding.
20 Last day	0	100	20 grains. Appr. 0.30 gr.	Inside sediment. Active after feeding.	0	100	20 grains. Appr. 0.30 gr.	Inside sediment. Active after feeding.	0	100	20 grains. Appr. 0.30 gr.	Inside sediment. Active after feeding.	0	100	20 grains. Appr. 0.30 gr.	Inside sediment. Active after feeding.

Table 19: Scores of mortality, food intake, and comments on behaviour during the monitoring period for common starfish. Each group started with 20 individuals.

	1 Near field				2 Medium field				3 Far field				4 Control			
Date x/07/2009	Dead	% food cons.	Food	Behaviour	Dead	% food cons.	Food	Behaviour	Dead	% food cons.	Food	Behaviour	Dead	% food cons.	Food	Behaviour
9 start			20 half mussel	Move to the side of the basket.			20 half mussel	Move to the side of the basket.			20 half mussel	Move to the side of the basket.			20 half mussel	Move to the side of the basket.
11	0	100	20 half mussel	On basket surface, some clustered.	0	95	20 half mussel	On basket surface, some clustered.	1, arms separated with suction caps still working.	75	10 half mussel	On basket surface, some clustered.	0	50	10 half mussel	On basket surface, some clustered.
13	0	30	20 half mussel	On basket surface, some clustered.	0	35	20 half mussel	On basket surface, some clustered.	3, arms separated with suction caps still working.	5	15 half mussel	On basket surface, some clustered.	1, half decayed	5	10 half mussel	On basket surface, some clustered.
15	0	55	20 half mussel	On basket surface, some clustered.	0	50	20 half mussel	On basket surface, some clustered.	1, arms separated with suction caps still working.	55	15 half mussel	On basket surface, some clustered.	0	60	19 half mussel	On basket surface, some clustered.
17	1, arms separated with suction caps	73	19 half mussel	On basket surface, some clustered.	2, arms separated with suction	30	20 half mussel	On basket surface, some clustered.	1, arms separated with suction caps	59	14 half mussel	On basket surface, some clustered.	1, arms separated with suction caps	13	18 half mussel	On basket surface, some clustered.

	1 Near field				2 Medium field				3 Far field				4 Control			
	still working.				caps still working.				still working.				still working.			
20	1, arms separated with suction caps still working.	64	16 half mussel	On basket surface, some clustered.	4, arms separated with suction caps still working.	65	14 half mussel	On basket surface, some clustered.	1, arms separated with suction caps still working.	56	13 half mussel	On basket surface, some clustered.	2, arms separated with suction caps still working.	28	14 half mussel	On basket surface, some clustered.
23	1, arms separated with suction caps still working.	89	-	On basket surface, some clustered.	1, arms separated with suction caps still working.	100	-	On basket surface, some clustered.	0	83	-	On basket surface, some clustered.	0	67	-	On basket surface, some clustered.

Table 20: Recorded behaviour of ragworm under pulse stimulation in the near field range.

Date 06 07 09		Species ragworm	Range nearfield		
Time	Sample (nr)	Effects exposures and comments			
		Pulse 1	Pulse 2	Pulse 3	Pulse 4
16.05	1	Jerky movement	No reaction	No reaction	No reaction
16.06	2	No reaction	No reaction	Slightly Jerky movement	Jerky movement
16.07	3	Slightly Jerky movement	Slightly Jerky movement	Slightly Jerky movement	Slightly Jerky movement
16.08	4	Jerky movement	Jerky movement	Jerky movement	Jerky movement
16.09	5	Slightly jerky movement	Slightly jerky movement	Slightly jerky movement	Slightly jerky movement
16.10	6	No reaction	No reaction	Slightly jerky movement	Slightly jerky movement
16.11	7	No reaction	No reaction	Slightly jerky movement	Slightly jerky movement
16.12	8	Jerky movement	Jerky movement	Jerky movement	Slightly jerky movement
16.13	9	No reaction	Slightly jerky movement	Slightly jerky movement	Slightly jerky movement
16.14	10	Slightly jerky movement	Slightly jerky movement	Slightly jerky movement	Slightly jerky movement
16.15	11	Slightly jerky movement	Slightly jerky movement	Slightly jerky movement	Slightly jerky movement
16.17	12	No reaction	No reaction	No reaction	No reaction
16.18	13	No reaction	No reaction	No reaction	No reaction
16.19	14	Jerky movement	Jerky movement	Jerky movement	Jerky movement
16.20	15	Slightly jerky movement	Slightly jerky movement	No reaction	Slightly jerky movement
16.21	16	Slightly jerky movement	No reaction	No reaction	No reaction
16.22	17	No reaction	No reaction	Slightly jerky movement	Slightly jerky movement
16.24	18	Movement until start of pulse	No reaction	No reaction	No reaction
16.25	19	No reaction	No reaction	Slightly jerky movement	No reaction
16.27	20	Slightly jerky movement	No reaction	No reaction	No reaction

Table 21: Recorded behaviour of ragworm under pulse stimulation in the medium field range.

Date 06/07/09		Species ragworm	Range medium field		
Time	Sample (nr)	Effects exposures and comments			
		Pulse 1	Pulse 2	Pulse 3	Pulse 4
17.11	1	No reaction	No reaction	No reaction	No reaction
17.13	2	No reaction	No reaction	No reaction	No reaction
17.14	3	No reaction	No reaction	No reaction	No reaction
17.15	4	Jerks its head	No reaction	No reaction	No reaction
17.16	5	No reaction	No reaction	No reaction	No reaction
17.18	6	Moving first but at rest after pulse	No reaction	No reaction	No reaction
17.19	7	No reaction	No reaction	No reaction	No reaction
17.21	8	No reaction	No reaction	No reaction	No reaction
17.22	9	No reaction	No reaction	No reaction	No reaction
17.24	10	No reaction	No reaction	No reaction	No reaction
17.26	11	No reaction	No reaction	No reaction	No reaction
17.27	12	No reaction	No reaction	No reaction	No reaction
17.28	13	No reaction	No reaction	No reaction	No reaction
17.30	14	No reaction	No reaction	No reaction	No reaction
17.32	15	No reaction	No reaction	No reaction	No reaction
17.34	16	No reaction	No reaction	No reaction	No reaction
17.35	17	No reaction	No reaction	No reaction	No reaction
17.36	18	No reaction	Slightly jerky movement	No reaction	No reaction
17.38	19	Jerky movement	Jerky movement	No reaction	No reaction
17.39	20	No reaction	Slightly jerky movement	Jerky movement	Slightly jerky movement

Table 22: Recorded behaviour of ragworm under pulse stimulation in the far field range.

Date 06/07/09		Species ragworm	Range far field		
Time	Sample (nr)	Effects exposures and comments			
		Pulse 1	Pulse 2	Pulse 3	Pulse 4
18.11	1	No reaction	No reaction	No reaction	No reaction
18.13	2	No reaction	No reaction	No reaction	No reaction
18.14	3	No reaction	No reaction	No reaction	No reaction
18.15	4	No reaction	No reaction	No reaction	No reaction
18.16	5	No reaction	No reaction	No reaction	No reaction
18.17	6	No reaction	No reaction	No reaction	No reaction
18.19	7	No reaction	No reaction	No reaction	No reaction
18.21	8	No reaction	No reaction	No reaction	No reaction
18.22	9	No reaction	No reaction	No reaction	No reaction
18.23	10	No reaction	No reaction	No reaction	No reaction
18.25	11	No reaction	No reaction	No reaction	No reaction
18.26	12	No reaction	No reaction	No reaction	No reaction
18.27	13	No reaction	No reaction	No reaction	No reaction
18.28	14	No reaction	No reaction	No reaction	No reaction
18.29	15	No reaction	No reaction	No reaction	No reaction
18.30	16	No reaction	No reaction	No reaction	No reaction
18.31	17	No reaction	No reaction	No reaction	No reaction
18.33	18	No reaction	No reaction	No reaction	No reaction
18.34	19	No reaction	No reaction	No reaction	No reaction
18.35	20	No reaction	No reaction	No reaction	No reaction

Table 23: Recorded behaviour of common prawn under pulse stimulation in the near field range.

Date 07/07/09		Species common prawn	Range near field		
Time	Sample (nr)	Effects exposures and comments			
		Pulse 1	Pulse 2	Pulse 3	Pulse 4
9.31	1	Jumps out of container			
9.38	2	Jumps about 15 cm	Jumps about 10 cm	Jumps about 10 cm	Jumps about 10 cm
9.41	3	Jumps about 5 cm	Jumps about 5 cm	Jumps about 5 cm	Jumps about 5 cm
9.44	4	Jumps about 10 cm	Jumps about 5 cm	Jumps about 5 cm	Jumps about 5 cm
9.47	5	Jumps about 2 cm	Jumps about 5 cm	Jumps about 5 cm	Jumps about 5 cm
9.49	6	Jumps about 5 cm	Jumps about 2 cm	Jumps about 2 cm	Jumps about 2 cm
9.54	7	Jumps about 10 cm	Jumps about 2 cm	Jumps about 5 cm	Jumps about 5 cm
9.55	8	Jumps about 10 cm	Jumps about 2 cm	Jumps about 5 cm	Jumps about 5 cm
9.58	9	Jumps about 5 cm	Jumps about 10 cm	Jumps about 5 cm	Jumps about 10 cm
10.00	10	Jumps about 10 cm	Jumps about 10 cm	Jumps about 2 cm	Jumps about 2 cm
10.02	11	Jumps about 10 cm	Jumps about 10 cm	Jumps about 10 cm	Jumps about 10 cm
10.21	12	Jumps about 10 cm	Jumps about 10 cm	Jumps about 10 cm	Jumps about 10 cm
10.25	13	Jumps about 15 cm	Jumps about 10 cm	Jumps about 10 cm	No reaction
10.27	14	Jumps about 15 cm	Jumps about 10 cm	Jumps about 10 cm	Jumps about 10 cm
10.29	15	Jumps about 10 cm	Jumps about 5 cm	Jumps about 5 cm	Jumps about 2 cm
10.31	16	Jumps about 10 cm	Jumps about 10 cm	Jumps about 5 cm	Jumps about 10 cm
10.33	17	Jumps about 5 cm	Jumps about 10 cm	Jumps about 5 cm	Jumps about 10 cm
10.34	18	Jumps about 5 cm	Jumps about 5 cm	Jumps about 2 cm	Jumps about 5 cm
10.36	19	Jumps about 10 cm	Jumps about 10 cm	Jumps about 5 cm	Jumps about 5 cm
10.39	20	Jumps about 15 cm	Jumps about 10 cm	Jumps about 10 cm	Jumps about 10 cm

Table 24: Recorded behaviour of common prawn under pulse stimulation in the medium field range.

Date 07/07/09		Species common prawn	Range medium field		
Time	Sample (nr)	Effects exposures and comments			
		Pulse 1	Pulse 2	Pulse 3	Pulse 4
11.05	1	Jumps aside	Jumps aside	Jumps aside	Jumps aside
11.07	2	Jumps about 5 cm	Jumps about 5 cm	Jumps about 5 cm	Jumps about 5 cm
11.09	3	Jumps about 10 cm	Moving	Jumps about 5 cm	Jumps about 5 cm
11.11	4	Jumps about 5 cm	Moving	Jumps about 5 cm	Jumps about 5 cm
11.13	5	Jumps about 5 cm	Jumps about 5 cm	Moving	Moving
11.15	6	Jumps about 10 cm	Jumps about 5 cm	Jumps about 5 cm	Jumps about 5 cm
11.17	7	Jumps about 15 cm	Jumps about 10 cm	Jumps about 5 cm	Jumps about 5 cm
11.20	8	Jumps about 15 cm	Jumps about 10 cm	Jumps about 10 cm	Jumps about 15 cm
11.22	9	Jumps about 2 cm	Jumps about 2 cm	Jumps about 2 cm	Jumps about 2 cm
11.24	10	Jumps about 10 cm	Jumps about 10 cm	Jumps about 5 cm	Jumps about 5 cm
11.26	11	Jumps about 10 cm	Jumps about 5 cm	Jumps about 5 cm	No reaction
11.27	12	Jumps about 10 cm	Jumps about 10 cm	Jumps about 10 cm	Jumps about 10 cm
11.29	13	Jumps about 10 cm	Jumps about 5 cm	Jumps about 10 cm	Jumps about 10 cm
11.34	14	Jumps about 10 cm	Jumps about 10 cm	Jumps about 10 cm	Jumps about 5 cm
11.36	15	Jumps about 10 cm	Jumps about 10 cm	Jumps about 5 cm	Jumps about 10 cm
11.38	16	Jumps about 5 cm	Jumps about 2 cm	Jumps about 2 cm	Jumps about 2 cm
11.40	17	Jumps about 10 cm	Jumps about 10 cm	Jumps about 10 cm	Jumps about 10 cm
11.42	18	Jumps about 5 cm	Jumps about 10 cm	Jumps about 5 cm	Jumps about 10 cm
11.43	19	Jumps about 5 cm	Jumps about 10 cm	Jumps about 10 cm	Jumps about 10 cm
11.45	20	Jumps about 15 cm	Jumps about 10 cm	Jumps about 10 cm	Jumps about 10 cm

Table 25: Recorded behaviour of common prawn under pulse stimulation in the far field range.

Date 07/07/09		Species common prawn	Range far field		
Time	Sample (nr)	Effects exposures and comments			
		Pulse 1	Pulse 2	Pulse 3	Pulse 4
12.05	1	No reaction	No reaction	No reaction	No reaction
12.06	2	No reaction	No reaction	No reaction	No reaction
12.08	3	No reaction	No reaction	No reaction	No reaction
12.09	4	No reaction	No reaction	No reaction	No reaction
12.10	5	No reaction	No reaction	No reaction	No reaction
12.12	6	No reaction	No reaction	No reaction	No reaction
12.13	7	No reaction	No reaction	No reaction	No reaction
12.14	8	No reaction	No reaction	No reaction	No reaction
12.15	9	No reaction	No reaction	No reaction	No reaction
12.16	10	No reaction	No reaction	No reaction	No reaction
12.17	11	No reaction	No reaction	No reaction	No reaction
12.18	12	No reaction	No reaction	No reaction	No reaction
12.20	13	No reaction	No reaction	No reaction	No reaction
12.21	14	No reaction	No reaction	No reaction	No reaction
12.22	15	No reaction	No reaction	No reaction	No reaction
12.25	16	No reaction	No reaction	No reaction	No reaction
12.26	17	No reaction	No reaction	No reaction	No reaction
12.30	18	No reaction	No reaction	No reaction	No reaction
12.31	19	No reaction	No reaction	No reaction	No reaction
12.33	20	No reaction	No reaction	No reaction	No reaction

Table 26: Recorded behaviour of subtruncate surf clam under pulse stimulation in the near field range.

Date 08/07/09		Species Spisula subtruncata	Range near field		
Time	Sample (nr)	Effects exposures and comments			
		Pulse 1	Pulse 2	Pulse 3	Pulse 4
15.37	1	No reaction	No reaction	No reaction	No reaction
15.40	2	No reaction	No reaction	No reaction	No reaction
15.41	3	No reaction	No reaction	No reaction	No reaction
15.42	4	No reaction	No reaction	No reaction	No reaction
15.43	5	No reaction	No reaction	No reaction	No reaction
15.44	6	No reaction	No reaction	No reaction	No reaction
15.45	7	No reaction	No reaction	No reaction	No reaction
15.46	8	No reaction	No reaction	No reaction	No reaction
15.47	9	No reaction	No reaction	No reaction	No reaction
15.50	10	No reaction	No reaction	No reaction	No reaction
15.51	11	No reaction	No reaction	No reaction	No reaction
15.51	12	No reaction	No reaction	No reaction	No reaction
15.52	13	No reaction	No reaction	No reaction	No reaction
15.53	14	No reaction	No reaction	No reaction	No reaction
15.54	15	No reaction	No reaction	No reaction	No reaction
15.54	16	No reaction	No reaction	No reaction	No reaction
15.55	17	No reaction	No reaction	No reaction	No reaction
15.56	18	No reaction	No reaction	No reaction	No reaction
15.56	19	No reaction	No reaction	No reaction	No reaction
15.57	20	No reaction	No reaction	No reaction	No reaction

Table 27: Recorded behaviour of subtruncate surf clam under pulse stimulation in the medium field range.

Date 08/07/09		Species Spisula subtruncata	Range medium field		
Time	Sample (nr)	Effects exposures and comments			
		Pulse 1	Pulse 2	Pulse 3	Pulse 4
16.57	1	No reaction	No reaction	No reaction	No reaction
16.58	2	No reaction	No reaction	No reaction	No reaction
16.58	3	No reaction	No reaction	No reaction	No reaction
16.59	4	No reaction	No reaction	No reaction	No reaction
17.00	5	No reaction	No reaction	No reaction	No reaction
17.00	6	No reaction	No reaction	No reaction	No reaction
17.01	7	No reaction	No reaction	No reaction	No reaction
17.02	8	No reaction	No reaction	No reaction	No reaction
17.02	9	No reaction	No reaction	No reaction	No reaction
17.03	10	No reaction	No reaction	No reaction	No reaction
17.04	11	No reaction	No reaction	No reaction	No reaction
17.04	12	No reaction	No reaction	No reaction	No reaction
17.05	13	No reaction	No reaction	No reaction	No reaction
17.05	14	No reaction	No reaction	No reaction	No reaction
17.11	15	No reaction	No reaction	No reaction	No reaction
17.11	16	No reaction	No reaction	No reaction	No reaction
17.12	17	No reaction	No reaction	No reaction	No reaction
17.13	18	No reaction	No reaction	No reaction	No reaction
17.19	19	No reaction	No reaction	No reaction	No reaction
17.19	20	No reaction	No reaction	No reaction	No reaction

Table 28: Recorded behaviour of subtruncate surf clam under pulse stimulation in the far field range.

Date 08/07/09		Species Spisula subtruncata	Range far field		
Time	Sample (nr)	Effects exposures and comments			
		Pulse 1	Pulse 2	Pulse 3	Pulse 4
17.46	1	No reaction	No reaction	No reaction	No reaction
17.47	2	No reaction	No reaction	No reaction	No reaction
17.47	3	No reaction	No reaction	No reaction	No reaction
17.48	4	No reaction	No reaction	No reaction	No reaction
17.48	5	No reaction	No reaction	No reaction	No reaction
17.49	6	No reaction	No reaction	No reaction	No reaction
17.50	7	No reaction	No reaction	No reaction	No reaction
17.51	8	No reaction	No reaction	No reaction	No reaction
17.52	9	No reaction	No reaction	No reaction	No reaction
17.53	10	No reaction	No reaction	No reaction	No reaction
17.55	11	No reaction	No reaction	No reaction	No reaction
17.55	12	No reaction	No reaction	No reaction	No reaction
17.56	13	No reaction	No reaction	No reaction	No reaction
17.56	14	No reaction	No reaction	No reaction	No reaction
17.56	15	No reaction	No reaction	No reaction	No reaction
17.57	16	No reaction	No reaction	No reaction	No reaction
17.58	17	No reaction	No reaction	No reaction	No reaction
17.58	18	No reaction	No reaction	No reaction	No reaction
18.05	19	No reaction	No reaction	No reaction	No reaction
18.06	20	No reaction	No reaction	No reaction	No reaction

Table 29: Recorded behaviour of green crab under pulse stimulation in the near field range.

Date 08/07/09		Species European green crab	Range near field		
Time	Sample (nr)	Effects exposures & comments			
		Pulse 1	Pulse 2	Pulse 3	Pulse 4
9.49	1	Jerky movement of legs	Jerky movement of legs	Jerky movement of legs	Slightly jerky movement of legs
9.52	2	Stiffening of legs	Stiffening of legs	Stiffening of legs	Stiffening of legs
9.54	3	Stiffening of legs	Stiffening of legs	Stiffening of legs	Stiffening of legs
9.57	4	Stiffening of legs	Stiffening of legs	Stiffening of legs	Stiffening of legs
9.59	5	Stiffening of legs	Stiffening of legs	Stretching of legs	Stretching of legs
10.01	6	Stiffening of legs	Stiffening of legs	Stiffening of legs	Stiffening of legs
10.04	7	Jerky movement of legs	Jerky movement of legs	Jerky movement of legs	Jerky movement of legs
10.07	8	Stiffening of legs	Stiffening of legs	Stiffening of legs	Stiffening of legs
10.10	9	Jerky movement of legs	Jerky movement of legs	Jerky movement of legs	Jerky movement of legs
10.15	10	Stiffening of legs	Stiffening of legs	Stiffening of legs	Stiffening of legs
10.17	11	Jerky movement of legs	Jerky movement of legs	Stiffening of legs	Stiffening of legs
10.19	12	Stiffening of legs	Stiffening of legs	Stiffening of legs	Stiffening of legs
	13	No data			
10.24	14	Stiffening of legs	Stiffening of legs	Stiffening of legs	Stiffening of legs
10.25	15	Jerky movement of legs	Jerky movement of legs	Stiffening of legs	Stiffening of legs
10.28	16	Stiffening of legs	Stiffening of legs	Stiffening of legs	Stiffening of legs
10.29	17	Stiffening of legs	Stiffening of legs	Stiffening of legs	Stiffening of legs
10.32	18	Stiffening of legs	Stiffening of legs	Stiffening of legs	Stiffening of legs
10.35	19	Stiffening of legs	Stiffening of legs	Stiffening of legs	Stiffening of legs
10.36	20	Stiffening of legs	Stiffening of legs	Stiffening of legs	Stiffening of legs

Table 30: Recorded behaviour of green crab under pulse stimulation in the medium field range.

Date 08/07/09		Species European green crab	Range medium field		
Time	Sample (nr)	Effects exposures and comments			
		Pulse 1	Pulse 2	Pulse 3	Pulse 4
11.18	1	Legs slightly stiffening	Legs slightly stiffening	No reaction	No reaction
11.19	2	Legs slightly jerking	Legs slightly jerking	No reaction	No reaction
11.21	3	Legs slightly jerking and producing air bubbles	Legs slightly jerking	Legs slightly jerking	Legs slightly jerking
11.23	4	Legs slightly jerking, and producing air bubbles	Legs slightly jerking	Legs slightly jerking	Legs slightly jerking
11.25	5	Legs slightly jerking, and producing air bubbles	Hardly any movement	Hardly any movement	Hardly any movement
11.27	6	No reaction	No reaction	No reaction	No reaction
11.29	7	Legs stiffening	Legs stiffening	Legs stiffening	Legs stiffening
11.31	8	Legs stiffening	Legs stiffening	Legs stiffening	Legs stiffening
11.32	9	No reaction, producing air bubbles	No reaction	No reaction	No reaction
11.34	10	Legs stiffening, producing air bubbles	Legs stiffening	Legs stiffening	No reaction
11.36	11	Legs stiffening	Legs stiffening	Legs stiffening	No reaction
11.37	12	Jerky movement	Stiffening, producing air bubbles	Legs stiffening	Legs stiffening
11.38	13	Jerky movement	No reaction	No reaction	No reaction
11.39	14	Jerky movement	Jerky movement	Jerky movement	Jerky movement
11.41	15	Slightly jerky movement, producing air bubbles	Slightly jerky movement	Slightly jerky movement	Slightly jerky movement
11.42	16	Slightly jerky movement, producing air bubbles	Slightly jerky movement	Slightly jerky movement	Slightly jerky movement
11.44	17	Jerky movement	Jerky movement, and producing air bubbles	Jerky movement	Jerky movement
11.46	18	Jerky movement, and producing air bubbles	jerky movement, and producing air bubbles	Jerky movement	Jerky movement
11.47	19	Slightly jerky movement	Slightly jerky movement	Slightly jerky movement	Slightly jerky movement
11.48	20	Jerky movement, and producing air bubbles	Jerky movement	Jerky movement	Jerky movement

Table 31: Recorded behaviour of green crab under pulse stimulation in the far field range.

Date 08/07/09		Species European green crab	Range far field		
Time	Sample (nr)	Effects exposures and comments			
		Pulse 1	Pulse 2	Pulse 3	Pulse 4
12.57	1	No reaction	No reaction	No reaction	No reaction
12.58	2	No reaction	No reaction	No reaction	No reaction
13.00	3	No reaction	No reaction	No reaction	No reaction
13.02	4	No reaction	No reaction	No reaction	No reaction
13.03	5	No reaction	No reaction	No reaction	No reaction
13.04	6	No reaction	No reaction	No reaction	No reaction
13.05	7	No reaction	No reaction	No reaction	No reaction
13.06	8	No reaction	No reaction	No reaction	No reaction
13.07	9	No reaction	No reaction	No reaction	No reaction
13.09	10	No reaction	No reaction	No reaction	No reaction
13.10	11	No reaction	No reaction	No reaction	No reaction
13.11	12	No reaction	No reaction	No reaction	No reaction
13.13	13	No reaction	No reaction	No reaction	No reaction
13.14	14	No reaction	No reaction	No reaction	No reaction
13.15	15	No reaction	No reaction	No reaction	No reaction
13.16	16	No reaction	No reaction	No reaction	No reaction
13.17	17	No reaction	No reaction	No reaction	No reaction
13.22	18	No reaction	No reaction	No reaction	No reaction
13.23	19	No reaction	No reaction	No reaction	No reaction
13.24	20	No reaction	No reaction	No reaction	No reaction

Table 32: Recorded behaviour of starfish under pulse stimulation in the near field range.

Date 09/07/09		Species common starfish	Range near field		
Time	Sample (nr)	Effects exposures and comments			
		Pulse 1	Pulse 2	Pulse 3	Pulse 4
12.06	1	No reaction	No reaction	No reaction	No reaction
12.07	2	No reaction	No reaction	No reaction	No reaction
12.08	3	No reaction	No reaction	No reaction	No reaction
12.09	4	No reaction	No reaction	No reaction	No reaction
12.09	5	No reaction	No reaction	No reaction	No reaction
12.10	6	No reaction	No reaction	No reaction	No reaction
12.11	7	No reaction	No reaction	No reaction	No reaction
12.12	8	No reaction	No reaction	No reaction	No reaction
12.13	9	No reaction	No reaction	No reaction	No reaction
12.14	10	No reaction	No reaction	No reaction	No reaction
12.20	11	No reaction	No reaction	No reaction	No reaction
12.21	12	No reaction	No reaction	No reaction	No reaction
12.25	13	No reaction	No reaction	No reaction	No reaction
12.25	14	No reaction	No reaction	No reaction	No reaction
12.26	15	No reaction	No reaction	No reaction	No reaction
12.26	16	No reaction	No reaction	No reaction	No reaction
12.27	17	No reaction	No reaction	No reaction	No reaction
12.27	18	No reaction	No reaction	No reaction	No reaction
12.28	19	No reaction	No reaction	No reaction	No reaction
12.29	20	No reaction	No reaction	No reaction	No reaction

Table 33: Recorded behaviour of starfish under pulse stimulation in the medium field range.

Date 09/07/09		Species common starfish	Range medium field		
Time	Sample (nr)	Effects exposures and comments			
		Pulse 1	Pulse 2	Pulse 3	Pulse 4
13.20	1	No reaction	No reaction	No reaction	No reaction
13.21	2	No reaction	No reaction	No reaction	No reaction
13.22	3	No reaction	No reaction	No reaction	No reaction
13.23	4	No reaction	No reaction	No reaction	No reaction
13.24	5	No reaction	No reaction	No reaction	No reaction
13.25	6	No reaction	No reaction	No reaction	No reaction
13.26	7	No reaction	No reaction	No reaction	No reaction
13.27	8	No reaction	No reaction	No reaction	No reaction
13.29	9	No reaction	No reaction	No reaction	No reaction
13.29	10	No reaction	No reaction	No reaction	No reaction
13.36	11	No reaction	No reaction	No reaction	No reaction
13.37	12	No reaction	No reaction	No reaction	No reaction
13.39	13	No reaction	No reaction	No reaction	No reaction
13.40	14	No reaction	No reaction	No reaction	No reaction
13.41	15	No reaction	No reaction	No reaction	No reaction
13.41	16	No reaction	No reaction	No reaction	No reaction
13.42	17	No reaction	No reaction	No reaction	No reaction
13.43	18	No reaction	No reaction	No reaction	No reaction
13.43	19	No reaction	No reaction	No reaction	No reaction
13.43	20	No reaction	No reaction	No reaction	No reaction

Table 34: Recorded behaviour of starfish under pulse stimulation in the far field range.

Date 09/07/09		Species common starfish	Range far field		
Time	Sample (nr)	Effects exposures and comments			
		Pulse 1	Pulse 2	Pulse 3	Pulse 4
14.02	1	No reaction	No reaction	No reaction	No reaction
14.02	2	No reaction	No reaction	No reaction	No reaction
14.03	3	No reaction	No reaction	No reaction	No reaction
14.03	4	No reaction	No reaction	No reaction	No reaction
14.04	5	No reaction	No reaction	No reaction	No reaction
14.06	6	No reaction	No reaction	No reaction	No reaction
14.05	7	No reaction	No reaction	No reaction	No reaction
14.06	8	No reaction	No reaction	No reaction	No reaction
14.06	9	No reaction	No reaction	No reaction	No reaction
14.07	10	No reaction	No reaction	No reaction	No reaction
14.18	11	No reaction	No reaction	No reaction	No reaction
14.18	12	No reaction	No reaction	No reaction	No reaction
14.19	13	No reaction	No reaction	No reaction	No reaction
14.20	14	No reaction	No reaction	No reaction	No reaction
14.20	15	No reaction	No reaction	No reaction	No reaction
14.20	16	No reaction	No reaction	No reaction	No reaction
14.21	17	No reaction	No reaction	No reaction	No reaction
14.21	18	No reaction	No reaction	No reaction	No reaction
14.21	19	No reaction	No reaction	No reaction	No reaction
14.24	20	No reaction	No reaction	No reaction	No reaction

Table 35: Recorded behaviour of razor clam under pulse stimulation in the near field range.

Date 10/07/09		Species razor clam	Range nearfield		
Time	Sample (nr)	Effects exposures and comments			
		Pulse 1	Pulse 2	Pulse 3	Pulse 4
10.01	1	Foot out of shells	Foot remains out of shells, No reaction	Foot remains out of shells, No reaction	Foot remains out of shells, No reaction
10.03	2	Foot sticking out of shells	No reaction	No reaction	Foot forcefully out of shells after pulse
10.06	3	Foot slightly sticking out of shells	Foot slightly sticking out of shells	Foot slightly sticking out of shells	Foot sticking out of shells
10.07	4	No reaction	No reaction	Foot sticking out of shells	No reaction
10.08	5	No reaction	No reaction	No reaction	No reaction
10.09	6	No reaction	Foot slightly sticking out of shells	Foot forcefully out of shells driving the animal away	Foot forcefully out of shells driving the animal away
10.11	7	Foot slightly sticking out of shells	Foot slightly sticking out of shells	Foot slightly sticking out of shells	Foot slightly sticking out of shells
10.15	8	No reaction	No reaction	No reaction	No reaction
10.17	9	Foot slightly sticking out of shells	Foot slightly sticking out of shells	Foot slightly sticking out of shells	Foot slightly sticking out of shells
10.18	10	Siphon spits out some sand	Foot and siphon sticking out of shells	Foot slightly sticking out of shells, animal turning	No reaction
10.20	11	Air bubbles from foot	No reaction	No reaction	No reaction
10.21	12	Air bubbles from foot	No reaction	No reaction	No reaction
10.23	13	Air bubbles from foot	No reaction	No reaction	No reaction
10.24	14	Air bubbles from foot	No reaction	No reaction	No reaction
10.25	15	Air bubbles from foot	No reaction	No reaction	No reaction
10.26	16	No reaction	No reaction	No reaction	No reaction
10.27	17	Foot slightly sticking out of shells	Foot slightly sticking out of shells	Foot slightly sticking out of shells	Foot slightly sticking out of shells
10.29	18	Foot sticking out of shells	Foot sticking out of shells	No reaction	No reaction
10.31	19	Air bubbles from foot	No reaction	No reaction	No reaction
10.35	20	No reaction	No reaction	No reaction	No reaction

Table 36: Recorded behaviour of razor clam under pulse stimulation in the medium field range.

Date 10/07/09		Species razor clam	Range medium field		
Time	Sample (nr)	Effects exposures and comments			
		Pulse 1	Pulse 2	Pulse 3	Pulse 4
11.16	1	No reaction	No reaction	No reaction	No reaction
11.17	2	No reaction	No reaction	No reaction	No reaction
11.18	3	No reaction	No reaction	No reaction	No reaction
11.19	4	No reaction	No reaction	No reaction	No reaction
11.20	5	Foot and siphon moved prior to pulse and were retreated	No reaction	No reaction	No reaction
11.22	6	No reaction	No reaction	No reaction	No reaction
11.24	7	No reaction	No reaction	No reaction	No reaction
11.26	8	No reaction	No reaction	No reaction	No reaction
11.27	9	No reaction	No reaction	No reaction	No reaction
11.27	10	No reaction	No reaction	No reaction	No reaction
11.28	11	No reaction	No reaction	No reaction	No reaction
11.28	12	No reaction	No reaction	No reaction	No reaction
11.30	13	No reaction	No reaction	No reaction	No reaction
11.31	14	No reaction	No reaction	No reaction	No reaction
11.31	15	No reaction	No reaction	No reaction	No reaction
11.32	16	No reaction	No reaction	No reaction	No reaction
11.32	17	No reaction	No reaction	No reaction	No reaction
11.33	18	No reaction	No reaction	No reaction	No reaction
11.38	19	No reaction	No reaction	No reaction	No reaction
11.38	20	No reaction	No reaction	No reaction	No reaction

Table 37: Recorded behaviour of razor clam under pulse stimulation in the far field range.

Date 10/07/09		Species razor clam	Range far field		
Time	Sample (nr)	Effects exposures and comments			
		Pulse 1	Pulse 2	Pulse 3	Pulse 4
12.47	1	No reaction	No reaction	No reaction	No reaction
12.48	2	No reaction	No reaction	No reaction	No reaction
12.48	3	No reaction	No reaction	No reaction	No reaction
12.49	4	No reaction	No reaction	No reaction	No reaction
12.49	5	No reaction	No reaction	No reaction	No reaction
12.50	6	No reaction	No reaction	No reaction	No reaction
12.50	7	No reaction	No reaction	No reaction	No reaction
12.51	8	No reaction	No reaction	No reaction	No reaction
12.51	9	No reaction	No reaction	No reaction	No reaction
12.52	10	No reaction	No reaction	No reaction	No reaction
12.52	11	No reaction	No reaction	No reaction	No reaction
12.53	12	No reaction	No reaction	No reaction	No reaction
12.53	13	No reaction	No reaction	No reaction	No reaction
12.55	14	No reaction	No reaction	No reaction	No reaction
12.55	15	No reaction	No reaction	No reaction	No reaction
12.56	16	No reaction	No reaction	No reaction	No reaction
12.57	17	No reaction	No reaction	No reaction	No reaction
12.57	18	No reaction	No reaction	No reaction	No reaction
12.58	19	No reaction	No reaction	No reaction	No reaction
12.58	20	No reaction	No reaction	No reaction	No reaction