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Marine Scotland Science
Freshwater Fisheries Laboratory
Faskally
Pitlochry
PH16 5LB

Behavioural Responses of Atlantic Salmon to Mains Frequency Magnetic Fields

John D Armstrong, Donna-Claire Hunter*, Rob J Fryer¹, Phil Rycroft² &
James E Orpwood

Marine Scotland Science Freshwater Fisheries Laboratory
Faskally, Pitlochry, Perthshire, PH16 5LB

1. Marine Scotland Science Marine Laboratory
Victoria Road, Aberdeen, AB11 9DB

2. Wyre Micro Design Ltd.
The Glyn, Llanerfyl, Powys, SY21 0JD

*Present address: Envirocentre, Alder House, Cradlehall Business Park
Inverness, IV2 5GH

Summary

Effects of mains frequency (50Hz) magnetic fields (MF) on behaviour of captive Atlantic salmon were examined in a large arena. Fish swam spontaneously within the arena passing from one side to the other through Helmholtz coils (arranged in four pairs) that penetrated a net barrier. When activated, a uniform intensity MF was generated between coils within a pair. Salmon were considered in two size categories: large fish (62-85 cm) and smaller post-smolts (24-41 cm). Large fish were tested in groups of six and considered at the individual level. For each fish, measurements were made of the times to approach, traverse and leave an activated coil pair (intensity = 95 μ T) on first encounter across defined distances. These times were compared with previous passage times for the same coil in a deactivated state across similar distances. There were no significant differences in approach, traverse or departure times associated with activation state of the coils. Post-smolts (18 trials of groups of six fish) were exposed to three 30-min periods of MF at 1.3, 11.4 and 95 μ T with 30-min controls before each treatment. The sequence in which the different MF levels were presented was varied sequentially. There was no evidence that the numbers of fish passing through the coils depended on the sequence in which

the MF intensities were presented ($p = 0.18$) or on the field intensity itself. Similarly, the numbers of shoals passing through coils did not depend on sequence or field intensity. There were no observations of unusual behaviours in association with MF up to $95 \mu\text{T}$.

Key words: EMF, Helmholtz coils, marine renewable energy, *Salmo salar*, swimming speed.

Introduction

The Scottish Government aims to meet its electricity demand from renewable sources by 2020 (Scottish Government, 2011). This objective is expected to be met in part by marine renewable energy developments that use offshore wind, wave and tidal power to generate electricity. For sustainable development of a marine renewable industry, it is necessary to consider effects on animals of the infrastructure needed to harness power (Gill, 2012). There is therefore interest in assessing the potential effect of magnetic fields associated with high voltage alternating current (AC) cables used to transmit electricity between adjacent generating devices. The electromagnetic field created when electrical current passes through a cable comprises an electric field and a magnetic field. Usually the electric field is shielded and therefore retained within the cable, whereas the magnetic field is detectable outside the cable. Magnetic fields generated by AC power cables (MF) are cyclical at 50 Hz (UK mains power frequency).

Atlantic salmon, *Salmo salar* L., is an important species to consider. It is both a cultural conservation icon (for example as a qualifying interest of Special Areas of Conservation) and the subject of important fisheries that have significant value for the rural economies of Scotland and other countries bordering the North Atlantic Ocean basin. Atlantic salmon spawn in fresh water, then, after a several years of growth, enter the “smolt” stage, when they metamorphose into a marine form and leave rivers. Little is known of the behaviour of salmon emigrating through coastal waters as post-smolts to feed in northern high seas. However, recent work has determined that adult fish returning to spawn are predominantly surface-dwelling but pass through a wide range of water depths (Godfrey *et al.*, 2014) and hence might encounter cables and associated MF from energy generating devices throughout the water column.

Atlantic salmon possess magnetite (ferrite particles that are influenced by magnetic fields) in the lateral line sense organ (Moore *et al.*, 1990). In the salmonid fish rainbow trout (*Oncorhynchus mykiss*), magnetite particles are arranged in chains (Diebel *et al.*, 2000) located in the nose and are linked to neural connections (Walker *et al.*, 1997). These structures might enable responses to magnetic fields. The Earth's magnetic field is largely static, directional and variable across space due to change in inclination and presence of anomalies. It therefore can provide migrating animals with information on location and direction of travel. There is evidence that a diverse range of organisms use magnetic fields for migration and orientation, including bacteria, bees and turtles (Blakemore, 1975; Wiltschko & Wiltschko, 2005).

With a single conductor, the MF is proportional to the electric current and inversely proportional to the distance from the centre of the conductor. Cable burial or other cable protection measures reduce MF by increasing distance, as does having multiple conductors in close proximity which allows positive and negative currents to cancel, as with three-phase AC. The magnetic properties of armouring can also affect MFs. All these elements frequently apply in actual cable deployments, but modelling is complex, and there is often uncertainty over what the resulting field strengths will be and more values measured in field situations would be useful (Gill *et al.*, 2014).

MF are superimposed upon the Earth's field. If detectable by Atlantic salmon, then MF might be perceived to be attractive, adverse or confusing stimuli and invoke some attraction, avoidance, stress, or alarm behaviour response. Here we test whether Atlantic salmon respond to MF in a controlled setting. Following observations of the behaviour of sturgeon, *Acipenser ruthenus* and *A. gueldenstaedti*, which do respond to MF (Basov, 1999), we postulated that salmon may respond behaviourally either by attraction (slowing of swimming and aggregation at the source), or avoidance (startle with acceleration in swimming speed, retreat from source, and perhaps subsequent reduction in activity).

Materials and Methods

Experimental Arena

Experiments were conducted in an annular tank (He & Wardle, 1988; Figure 1), which provided a circular channel (3.66 m wide, 0.98 m deep) between two concentric walls (9.78 m, internal diameter, 2.46 m, external diameter). The channel was divided into two lateral halves of equal volume by pairs of mesh (0.01 m) walls (Figure 1). Each of the walls was penetrated by two openings through mesh columns between pairs of Helmholtz coils (Figure 1) each mounted within a plastic hoop (1 m internal diameter, 0.025 m nominal bore). This arrangement meant that fish could pass from one side of the arena to the other only by swimming through a coil pair. The coils were separated face-face by 0.5 m (Figure 1). Video cameras (four in total) were positioned to allow observation of fish passage through each of the coil pairs from above and below water (Figure 1). Signals from the cameras were multiplexed together with time and date on to a single image and could be viewed and recorded in a control room adjacent to the room housing the annular tank.

Helmholtz Coils

The Helmholtz coils in a pair generated a near-uniform field between them. Faces of the coils were parallel to 20° east of magnetic north in the Earth's static field. Field strength diminished according to an approximate inverse square relationship to about 50% at 0.3 m outside the coil face. Fish approaching the field would experience an increasing field, a steady field within the coils and a reducing field as they departed. Initial calibration routines established relationships between current and magnetic field strength. Current was produced from a voltage regulator at mains frequency and could be adjusted remotely in the control room to generate pre-determined field intensities. Cooling systems within the plastic coil housings used pumped water to prevent any measureable local build-up of heat at high intensities. Following calibration, a target MF intensity could be generated by adjusting voltage and verified remotely by measuring current in the circuit. Regular tests between trials checked functionality of the system by *in situ* measurement of magnetic field strength.

Experimental Animals

Pilot studies established that wild-caught Atlantic salmon were unsuitable experimental subjects. Such animals showed common symptoms of high stress levels, including prolonged inactivity, no interest in food and darkened colouration. By contrast, Atlantic salmon that had been reared in captivity quickly settled in the arena. Two life stages were tested: large salmon and post-smolts.

The large fish had average length 76.7 cm (range 62-85 cm) and average weight 4511.9 g (range 2490-6429 g). They were transported from sea cages in an aquaculture facility and held in the Marine Scotland Science aquarium at Aberdeen prior to experiments. These fish exhibited extended periods of slow sustained swimming when settling in the experimental arena and readily moved through deactivated coil pairs.

The post-smolts were hatchery-reared and the progeny of wild sea-run adult Atlantic salmon sourced from the River Don, Aberdeenshire, Scotland. These fish had average length 33.1cm (range 24-41cm) and average weight 408g (range 143 to 732g). Eggs were fertilized and hatched at the Marine Scotland Aultbea fish rearing facility in 2010 before being transferred to the Marine Scotland Marine Laboratory, Aberdeen, in May 2011. The fish were held under standard hatchery conditions, fed on commercial salmon pellets of a size appropriate to their developmental stage, and subject to regular disturbance during cleaning of tanks. These fish were tested at the post-smolt stage in full sea water, having metamorphosed from the freshwater to the more silvery, elongated marine form. The post-smolts were first generation progeny of wild-caught parents whereas the period of hatchery selection of the larger salmon was not known. Salmon were held on *ad libitum* rations at ambient temperatures prior to experiments. Food was withheld in the experimental arena.

Experimental Protocols

Marking

Large salmon were lightly anaesthetised (in MS-222), weighed and measured the day before being introduced to the experimental arena. They were marked with external floy tags comprising a subcutaneous anchor and filaments of various

shades. These tags were visible on the camera systems and could be used for individual identification. It was not possible to devise a reliable method for externally tagging small smolts such that they could be differentiated by the video system. Other internal tagging methods, such as using passive integrated transponders, were rejected because oscillations of such metallic devices could be induced within the field and hence affect the behaviour of the fish. As a consequence, large salmon could be considered as individuals, but post-smolts were considered at the group level. Hence, experimental designs, response variables and statistical analyses differed to some extent between these categories of fish.

Large Salmon

These fish were tested in six trials each of six fish between 3 and 21 May 2013 to establish if the swimming speed when approaching, traversing and leaving a coil pair depended on whether the coils were activated. The fish were introduced as a group between 16:00 and 17:00 h and allowed to settle overnight. Tests began between 10:15 and 12:30 h on the following day and proceeded according to a common protocol. The video recording system was activated and recorded spontaneous activity of salmon within the arena over 30 min (Period 1). The coil pair on either one side of the arena or the other (chosen sequentially with trial number) was then activated at 95 μT for 30 min (Period 2). Subsequent analysis of the recordings measured the passage time of each fish through three zones: approaching (over 1 m), traversing (over 0.5 m) and departing (over 1 m) the activated coil pair. Each of these variables was measured from video recordings of the first occasion the fish encountered the activated coil pair in Period 2 and the first time it encountered the same coil pair in Period 1.

Post-smolts

In total, 18 trials were carried out between 28 May and 17 July 2012. At approximately 16:00 h on the day before each trial, six fish were introduced to the annular tank. The following day, the video cameras were activated and fish movements were recorded between 11:00 and 14:00 h. Fish were exposed to three 30 min periods of MF in which both coil pairs were activated at 1.3, 11.4 and 95 μT , with 30 min controls before each treatment. The sequence in which the different MF levels were presented was varied sequentially between

control-1.3-control-11.4-control-95 or control-11.4-control-95-control-1.3 or control-95-control-1.3-control-11.4 (all values in units of μT). Thus, 6 replicates of each presentation order were carried out over the course of the experiment. Subsequent analysis identified the numbers of fish and shoals of fish passing through both coil pairs in each period of the experiment. Also records were made of the passage times of the first fish to traverse either of the coil pairs were also recorded, again through in relation to three zones (as in adults): approaching (over 1 m), traversing (over 0.5 m) and departing (over 1 m) the first activated coil pair. Analogous measures were made of the first fish to traverse in relation to the same coil when in its prior inactivated state in the previous control period (although because it was not possible to individually identify the smolts, these might have been different fish).

Statistical Analysis

Passage Times of Large Fish

There were six measurements of passage time for each fish: the times taken for the fish to approach, traverse and depart a coil pair when the coil was inactive, and the corresponding times after activation of the coil. The times were log transformed to achieve a more symmetric distribution and then modelled by a linear mixed model, written informally as

$$\text{passage time} \sim \text{zone} + \text{zone} : \text{activation} + (1 \mid \text{trial} / \text{fish} / \text{activation}) \quad (1)$$

The fixed effects were zone (approach, traverse, departure) and the interaction between zone and coil activation (off/on). The interaction is the term of interest since it measures the change in mean passage time in each zone after activating the coil pair. The random effects were trial, fish (nested within trial) and activation (nested within fish), where this last effect allows for random variation in the basal swimming speed of each fish between its passage through the inactive and the activate coil pair that is not been accounted for by the interaction between zone and activation.

The effect of activating the coil on passage time was assessed by comparing the fit of model (1) with that of a model in which passage time did not depend on coil activation:

passage time ~ zone + (1 | trial / fish / activation) (2)

The test statistic was the log-likelihood ratio between the two models. However, as the distribution of passage times was only roughly normal and there was the occasional outlier, the significance of the test statistic was assessed by a bootstrap procedure that made no parametric assumptions about the distribution of the data. In essence, the passage times were adjusted so that they satisfied the null hypothesis of no activation effect; the adjusted passage times were resampled by non-parametric bootstrapping of fish within trials; models (1) and (2) were fitted to the bootstrapped data, giving the log-likelihood ratio. Repeating this procedure 1000 times created a bootstrap reference distribution of the test statistic under the null hypothesis. The significance of the observed test statistic was then the proportion of the reference distribution that exceeded the observed test statistic; see e.g. Efron and Tibshirani (1993) for more details. Confidence intervals on the parameter estimates from model (1) were also obtained by non-parametric bootstrapping of fish within trials, this time using the unadjusted data.

Passage Times of Post-smolts

A simplified analysis of passage times was carried out for the post-smolts. The fish could not be individually identified, so there was one set of times for each trial. Further, the measurements when the coil pair was inactive were not necessarily made on the same individual fish as when the coil pair was activated. Consequently, model (1) was reduced to

passage time ~ zone + zone : activation + (1 | trial / activation) (3)

It was also necessary to assume that the different field intensities had the same effect on passage time, as there were too few data to model them separately. The rest of the analysis was analogous to that for the adults.

Movement of Post-smolts

The numbers of fish passing through the coil pair in each 30 min period were modelled using a generalised linear mixed model assuming Poisson errors and a log link. A full model of the form

$$\text{number of fish} \sim \text{sequence} : \text{period} + (1 \mid \text{trial} / \text{period}) \quad (4)$$

was first fitted to the data. The fixed effects were sequence (i.e. the three different field intensity sequences (μT) presented to the fish ((0, 1.3, 0, 11.4, 0, 95), (0, 11.4, 0, 95, 0, 1.3) and (0, 95, 0, 1.3, 0, 11.4)) and period (1 through 6). By fitting the interaction of the two categorical variables, the model estimated a different mean for each combination of sequence and period. The random effects were trial, period (nested within trial) to account for any over-dispersion in the data, and the Poisson variation typically associated with count data. This model was compared to two simpler models in a backwards stepwise procedure:

$$\text{number of fish} \sim \text{field intensity} + (1 \mid \text{trial} / \text{period}) \quad (5)$$

in which the number of fish depended only on the field intensity (0, 1.3, 11.4 and 95 μT) and not the sequence in which it was presented to the fish; and

$$\text{number of fish} \sim 1 + (1 \mid \text{trial} / \text{period}) \quad (6)$$

in which field intensity had no effect on the number of fish. The models were compared by standard log likelihood ratio tests. The numbers of fish shoals passing through the coils in each observation period were modelled in the same way.

Results

There was no evidence that activating a coil pair changed the passage times of the adults or the post-smolts in any of the three zones ($p = 0.12, 0.14$ respectively). The estimates of change in each zone, with pointwise 95% confidence intervals, are shown in Figure 2. These have been back-transformed from the log scale to give the % change in passage time after activating the coil. On a pointwise basis, the confidence intervals suggest that the passage time of post-smolts in the exit zone increased after activating the coil. However, this effect is non-significant when the number of comparisons is taken into account. The confidence intervals indicate the values of % change in passage time compatible with the data. Thus, although non-significant, the passage time of

adults in the coil could have decreased by as much as 11% or increased by as much as 35% with 95% confidence.

There was no evidence that the numbers of post-smolts passing through the coil depended on the sequence in which the MF intensities were presented ($p = 0.18$) or the field intensity itself ($p = 0.31$) (Figure 3). Similarly, the numbers of shoals passing through coils did not depend on sequence or field intensity ($p = 0.09$, 0.28 respectively).

Discussion

When alarmed, fish frequently exhibit startle behaviour, during which locomotion mode changes from slow sustained (aerobic) to burst (anaerobic) swimming. This switch can result in a four- to ten-fold change in speed of coho salmon depending on fish size (Brett, 1964). There was no evidence of any such increase in swimming speeds of post-smolts or adults in association with MF in this experiment. Furthermore, there was no evidence that the MF significantly slowed or obstructed movements of the salmon, as would be indicated by a reduction in the passage time through the Helmholtz coils or evasion behaviour. Moreover, observations of post-smolts suggested no effect of repeated exposures to MF on activity levels. Hence, the experiments provide no evidence for startle, which in a natural situation may have negative effects such as making fish more vulnerable to attack from predators. Neither was there retardation in progress, which, in a natural context, may cause delay in migration. The possibility that there were changes in behaviour that were too small to detect is plausible. However, the experiment had sufficient power to identify about a two-fold change, which is much less than expected if the fish were startled. Furthermore, it seems unlikely that any such minor effects would have major ecological consequences in terms of impinging migration or increasing mortality risk.

Certain species of salmonid fishes have a physiological constitution that may equip them to detect magnetic fields. Magnetite is present in Atlantic salmon (Moore *et al.*, 1990) and the Earth's DC field might be involved in facilitating their long-range migrations to and/or from oceanic feeding grounds. If so, it is possible that the physiological systems that enable salmonids to detect DC magnetic fields might not respond sufficiently rapidly to detect oscillating AC

magnetic fields above a threshold frequency. Even if Atlantic salmon transduce MFs, then the signals may not be interpreted in any meaningful way by the neural system. The absence of any measurable change in swimming does not imply that salmon fail to detect MF, only that they do not necessarily exhibit an associated behavioural reaction. Conditioning experiments, as used by Rommel & McCleave (1973), might be used to determine whether MF are registered. In this case, a conditioning period is used to allow the animal to associate the test stimulus with a positive or negative experience. When the test stimulus is then presented, the animal exhibits anticipation of the associated experience, such as change in heart rate. Using this approach, Rommel & McCleave found no evidence that Atlantic salmon registered sustained directional shifts in DC magnetic field of a magnitude equivalent to the Earth's field (about 50 μT).

Lake sturgeon, *Acipenser fulvescens*, responded to MF at intensities above 1000-2000 μT by increased incidence of a range of activities: pectoral fin flare, slowing or gliding, body spasms, attraction to the magnet, sudden stops, burst swimming (C starts), thrashing and tail spasms (Bevelheimer *et al.*, 2013). Atlantic salmon were not observed to make any such behaviours in the present study. Indeed, to human observers there was no change in swimming behaviour associated with activated compared with control coils. Westerberg & Lagenfelt (2008) used acoustic tracking to study swimming speed of European eels crossing a 130 kV AC power cable in the Baltic Sea with an associated MF of about 200 μT at 1 m (Olsson *et al.*, 2010). Acoustic receivers were arranged in four transects to create three intervals, the central interval where the cable was situated, and two adjacent intervals, one each to the north and south of the cable. It was found that the swimming speed of European eels was significantly slower when crossing the cable than through the adjacent intervals. However, reductions in swimming speed tended to occur at periods of higher current and hence very high MF.

It is notable that in the cases of both lake sturgeon and eels, responses to MF are at higher intensities compared to those that fish might typically be expected to encounter in association with offshore marine renewable developments. For example, Olsen *et al.* (2010) predicted a peak field strength of 35 μT using modelled examples of typical power cable specifications. The intensities used in the present study to challenge Atlantic salmon encompass and exceed these predicted field strengths. We cannot rule out the possibility that Atlantic salmon

might respond to very high intensities or that they might respond to unprotected extremely high voltage cables such as tested using eels by Westerberg & Lagenfelt (2008).

Notwithstanding the absence of effects of magnetic fields observed from experimental manipulations, it is useful to consider briefly the degree to which Atlantic salmon might be exposed to MF in the marine environment. Transient noise from localised MF would generally be expected to be relatively small in relation to the Earth's field of approximately 50 μ T. Furthermore, the fish would be exposed to such field strengths for a distance of only a few metres if passing close to the cable. It may in any case be rare for migrating salmon to experience such fields on the seabed if post-smolts near Scotland's coasts are as surface oriented as they are believed to be further offshore (Shelton *et al.*, 1997) and because returning adults seem to spend little time in the benthic coastal zone (Godfrey *et al.*, 2014) at least until they approach rivers. They may however be more likely to encounter surface cabling from wave generators, particularly, perhaps, near the mouth of their home river when they would be near the shoreline, and perhaps over a wider range depending on migration routes and strategies.

Field strength diminishes rapidly with distance from the cable (Normandeau, 2011). Therefore, burial or armouring (typically covering with rocks) would reduce the field intensities that may be experienced by fish by an order of magnitude and more. Cables might be most exposed near landfall and when associated with pelagic devices.

The main finding of this study was no identifiable behavioural response of Atlantic salmon to MF at intensities of 95 μ T and below. The study animals were reared in the laboratory rather than the wild. This may have affected their response to MF. However, this rearing history ensured that they were relaxed in the experimental arena and hence probably more likely to respond to a stimulus than wild-reared fish that would already be stressed and potentially not responsive. It would be interesting to observe responses of salmon in the wild to cables *in situ*, but very challenging to attain a good degree of control, standardisation, replication and accuracy in measurement of behaviours. In the absence of such data, the results of the present study provide some reassurance that salmon are unlikely to be seriously adversely affected by MF under many circumstances.

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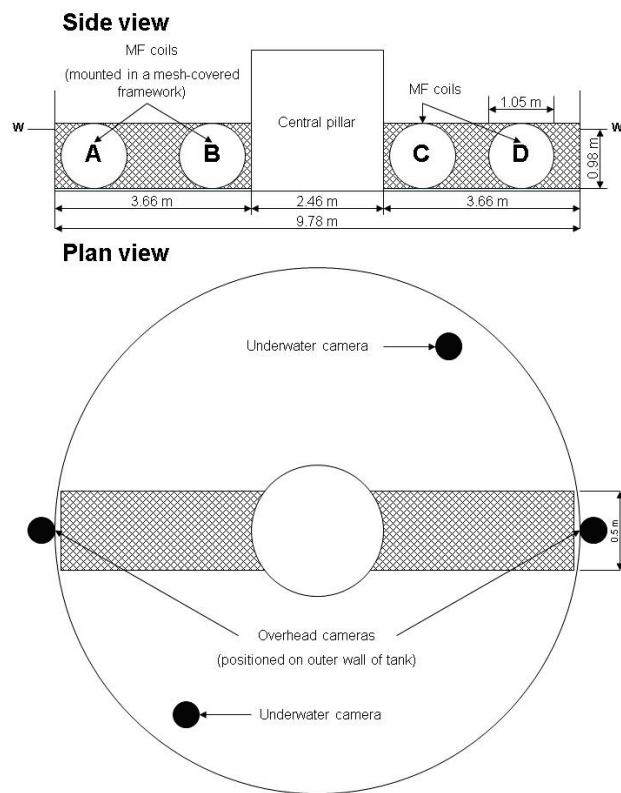
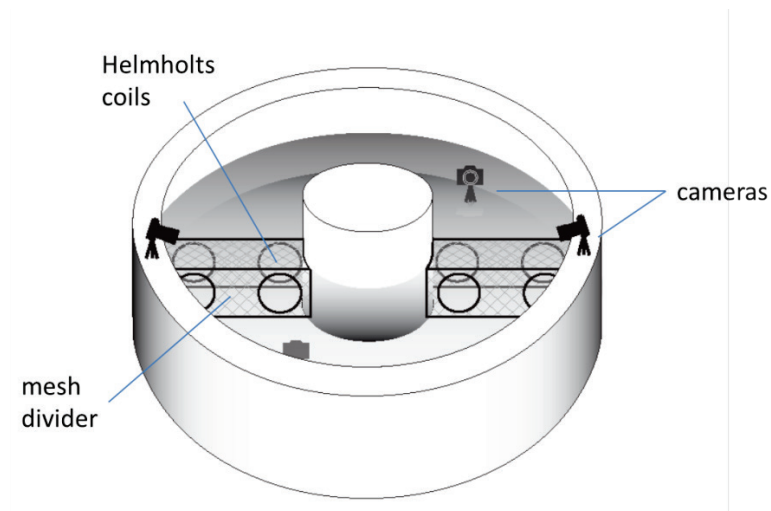


Figure 1 The annular tank used to tests effects of MF on Atlantic salmon. General elements are shown in an artist's impression (top frame); the cylinder of netting between each pair of Helmholtz coils is omitted for clarity. Details of dimensions are drawn to scale in the side (middle frame) and plan (lower frame) views.

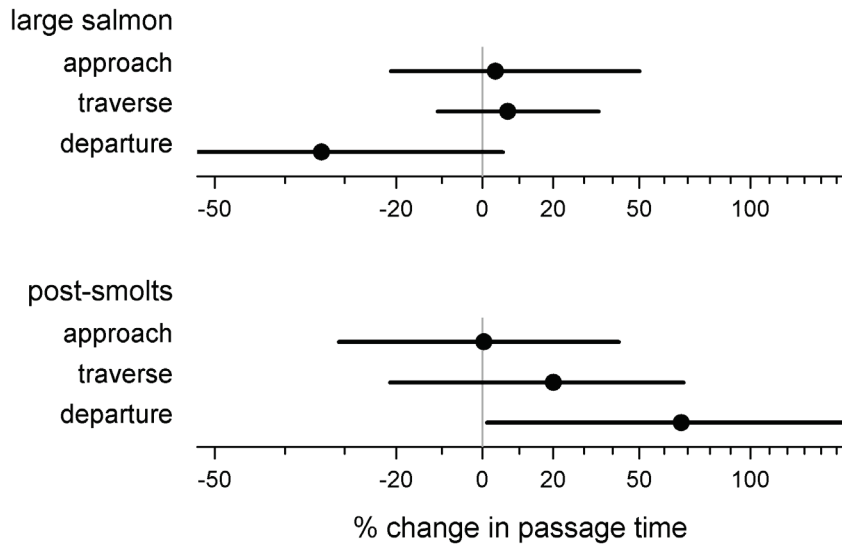


Figure 2 The percentage change in passage time when the coil-pair is activated when approaching, traversing and departing the coil-pair: estimates (dots) with pointwise 95 % confidence intervals (horizontal lines).

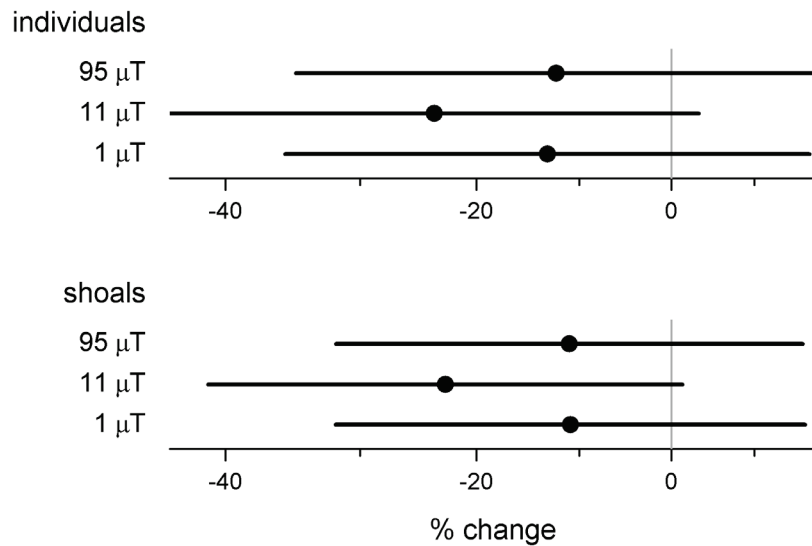


Figure 3 The percentage change in the number of individuals and shoals traversing the coil-pair when the coil-pair is activated for three different field intensities: estimates (dots) with pointwise 95 % confidence intervals (horizontal lines). These estimates are based on model (5) in which the order in which the intensities were presented is assumed to have no effect

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Marine Scotland Science
Freshwater Fisheries Laboratory
Faskally
Pitlochry
PH16 5LB

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