

# Electrical stunning and killing of Atlantic salmon

Final report



Illustration: Nofima

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## Report

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## **Preface**

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# 1 Summary

Atlantic salmon (*Salmo salar*) at various size 226-1938 g were placed in electric dry stunning either tail- or head- first and then exposed to 50 Hz, 240 V AC for 0.5 and thereafter 9.5 s of electricity. This to verify whether the animal is rendered unconscious prior to killing by immersing the animal into cold brine (-14 to -18 °C) or hypoxic water, supersaturated with CO<sub>2</sub>. In addition, salmon from 2000 to 5000 g were exposed to head stunning only. Behaviour, heart (ECG) and brain (EEG) activity were monitored until animal was classed as dead. Post the treatment, the internal temperature of the animal was measured placing loggers into the neuro and heart cavity. The electric potential across the brain was also measured. A preliminary evaluation of the EEG and ECG registrations in the individual fish showed that loss of consciousness can be induced within 0.5 second and consciousness can be prolonged without recovery when the cold brine or seawater saturated with carbon dioxide is applied as killing method. Our results also suggest that the current can be applied head to tail, which facilitates immediate stunning of Atlantic salmon in a commercial setting. We conclude that electrical stunning in combination with cold brine is effective for humane stunning and killing Atlantic salmon.

## 1.1 Sammendrag

Atlantisk laks (*Salmo salar*) i forskjellige størrelser mellom 226-1938 g ble lagt i elektrisk tørrbedøvelse enten med hale eller hode først og deretter utsatt for 50 Hz, 240 V AC i 0,5 og deretter 9,5 s elektrisitet. Dette for å verifisere om fisken er bevisstløs før avliving ved å senke fisken i kaldt saltlake (-14 til -18 °C) eller hypoksisk vann, overmettet med CO<sub>2</sub>. I tillegg ble laks fra 2000 til 5000 g kun utsatt for hodebedøvelse. Atferd, hjerte (EKG) og hjerne (EEG) aktivitet ble overvåket inntil fisken ble klassifisert som dødt. Etter behandlingen ble den indre temperaturen til fisken målt ved å plassere loggere inn i neuro og hjertehulen. Det elektriske potensialet gjennom hjernen ble også målt. En foreløpig evaluering av EEG- og EKG-registreringene hos den enkelte fisken viste at bevissthetstap kan induseres innen 0,5 sekund og bevisstheten kan forlenges uten restitusjon når den kalde saltlaken eller sjøvann mettet med karbondioksid brukes som avlivingsmetode. Resultatene våre tyder også på at strømmen kan påføres hode mot hale, noe som muliggjør umiddelbar bedøvelse av atlantisk laks i kommersielle omgivelser. Vi konkluderer med at elektrisk bedøvelse i kombinasjon med kald saltlake er effektivt for human bedøvelse og avliving av atlantisk laks.

## 2 Introduction

During rearing of Atlantic salmon (*Salmo salar*) conditions may occur that require an intervention. Conditions that require an intervention can be the occurrence of a disease, a breakdown of a life-supporting production system where compromised welfare is unavoidable or when fish are in an emaciated condition. In these cases, chemical anaesthetics are often used in particular freshwater systems on parr and smolts, while post smolt at sea electrical stunning in combination with mincing is a common. In all cases, the byproduct potential of is often significantly reduced by mincing and silage or turned into waste by the use anaesthetics. An ultimate intervention would be slaughter, which can be needed for legal, health, welfare and sustainability reasons. It is known that asphyxia, hypoxia or chilling on ice are not acceptable methods for killing conscious fish. However, when killing at a farm is required, stunning methods that are applied in a slaughterhouse are likely not feasible or practicable. Alternative methods for humane killing are, therefore, needed. A method is considered humane when consciousness is lost without any avoidable discomfort until death occurs. When electricity is used, Norwegian legislation requires that consciousness is lost within 0.5 seconds (Anon, 2006).

To protect Atlantic salmon at slaughter, the conditions applied for stunning and killing have to be underpinned by registration of brain function (EEG) in combination with registration of heart function (ECG).

## 3 Objectives

The objective was to see if the application of alternating electrical current after dewatering of farmed Atlantic salmon in combination with immersion into a cold brine is a humane stunning and killing method for farmed Atlantic salmon. An alternative killing method of electrically stunned salmon was also studied; this was immersion of the unconscious fish in seawater that is saturated with carbon dioxide.

We studied whether tail-first stunning consciousness in Atlantic salmon could be lost within 0.5 seconds. For this purpose the configuration of the electrodes in the dry stunner was adjusted. Tail-first stunning is of interest for practical application, as orientation of the fish prior to entering the stunner is then no longer required.

## 4 Material and methods

### Preparation of fish

In March 2023, a total of 50 Atlantic salmon with mean (SD) size of 1523 (1205,4) g (ranging 276 to 5320 g) were selected from 3 different production tanks at Matre research station, Matre, Norway. All fish were held under seawater conditions at 8-9 °C and fed daily. Before the experiment one fish at the time was captured from the tank with a dipnet and transported in a transfer tank to the experimental facilities. Oxygen was at all times above 80% saturation. Prior to neurophysiological measurements (EEG and ECG), each fish was then fixed in a net and kept in a holding tank with water of appropriate temperature, salinity, and oxygen concentration. Then, with the fish in the water, the EEG and ECG electrodes are invasively applied under local anaesthesia, using injectable 2% lidocaine (Ross and Ross, 2008). This procedure does not require a period of recovery prior to registration of an EEG and ECG in fish.

### Stunning and killing

In a laboratory setting, the specifications for immediate stunning, i.e. within 0.5 second, Atlantic salmon without recovery until death were established, using registration of the electrical activity in the brains (EEG) and heart (ECG). We assessed whether a transfer of the stunned fish to a cold brine of -14 to -18 °C or seawater of 8 °C supersaturated with carbon dioxide (< pH 5.2) prevented recovery thus killing.

To assess whether an immediate stun in Atlantic salmon could be achieved, each individual fish was exposed to 220 V 50 Hz AC for 0.5 s in dry stunner (Stansas, Optimar, Norway). Please, note that for the 0.5-second exposure to the current, the duration of unconsciousness is too short to kill a fish in an unconscious state (Van de Vis and Lambooj, 2016). Hence, after a time interval of approx. 10 seconds, which is too short for recovery, a long-duration exposure of at least 5 seconds was applied to prevent recovery prior to the application of one of the two killing methods. This time interval of 10 seconds was sufficient to observe changes on the EEG and ECG caused by the expose to the 0.5-second stun, which is needed to determine whether consciousness was lost.

After a 10 second stun in total each individual salmon was placed in the cold brine to kill it in an unconscious state. For the seawater that was saturated with carbon dioxide we used individual Atlantic salmon that was exposed for in total 5.5 seconds to the electricity. These exposure of 9.5 and 5 seconds, respectively, were maintenance stuns which were needed to prolong the period of loss of consciousness in each individual Atlantic salmon prior to killing it the cold brine or seawater saturated with carbon dioxide, respectively. It is known that the time interval between stunning and the application of one of the two killing methods is crucial. For each individual stunned fish, the time interval between stunning and the application of a killing method was approx. 10-15 seconds.

### EEG and ECG registrations

EEG and ECG registrations were carried out in individual Atlantic salmon. This implies that the procedure was repeated for each fish. The fish were not re-used.

Immediately after the electrodes were placed, a baseline for both the EEG and ECG was measured; this involved the fish being in the tank of water. Then, each fixed fish was placed between the stunning electrodes and the current was applied head to tail. Based on changes in the EEG and ECG, we determined if unconsciousness was indeed induced within 0.5 second. EEG and ECG registrations continued for at least 15 min after a maintenance stun followed by the application of one the two killing methods.



### **Voltage, temperature measurements**

For measuring the potential difference across the brain. The EEG electrodes were placed in a fixed position 10 mm from centre to centre. Connected to the electrodes, in parallel to EEG equipment was oscilloscope (PXI multimeters). The during recording, the peak electric potential difference (V) was recorded within 0.5 sec was measured. For temperature logging in brine. Fish were after classed as dead, re-thawed to ambient temperature (from brine studies) or directly after CO<sub>2</sub> experiments placed into the brine having temperature loggers placed into the neuro and heart cavity until passing freezing temperature for the carcass (below -3 °C).

### **Ethics**

The experiment was approved by the Norwegian Food Safety Authority in accordance with application id FOTS no 29074.

## 5 Findings, discussion, and conclusion

A preliminary evaluation of the EEG and ECG registrations in the individual fish show that loss of consciousness can be induced within 0.5 second and consciousness can be prolonged without recovery when the cold brine or seawater saturated with carbon dioxide is applied as killing method of the stunned fish. Our results also suggest that the current can be applied head to tail, which facilitates immediate stunning of Atlantic salmon in a commercial setting. To prevent that fish are exposed to pre-shocks prior to an immediate loss of consciousness the first row of electrodes was set as the opposite polarity than the second row allowing electricity to pass from tail to head.

Figure 1A shows an example of the registration of the EEG and ECG in an individual Atlantic salmon prior to electrical stunning and after a 0.5 second exposure to the electricity. The right side in Fig 1A shows the induced epileptiform insult, which is indicative for an immediate loss of consciousness, after the 0.5 second stun, is presented (green line on the right side).

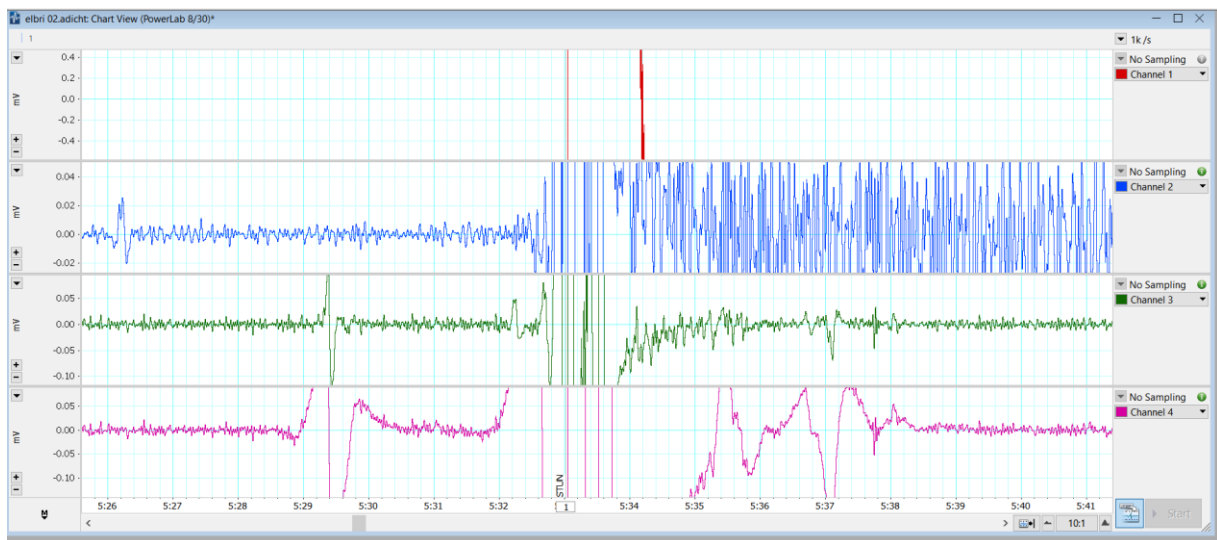


Figure 1A: EEG and ECG in a conscious Atlantic salmon (**left side only**) and the induced general epileptiform insult (unconscious fish) (**right side only**) after a 0.5 s stun.

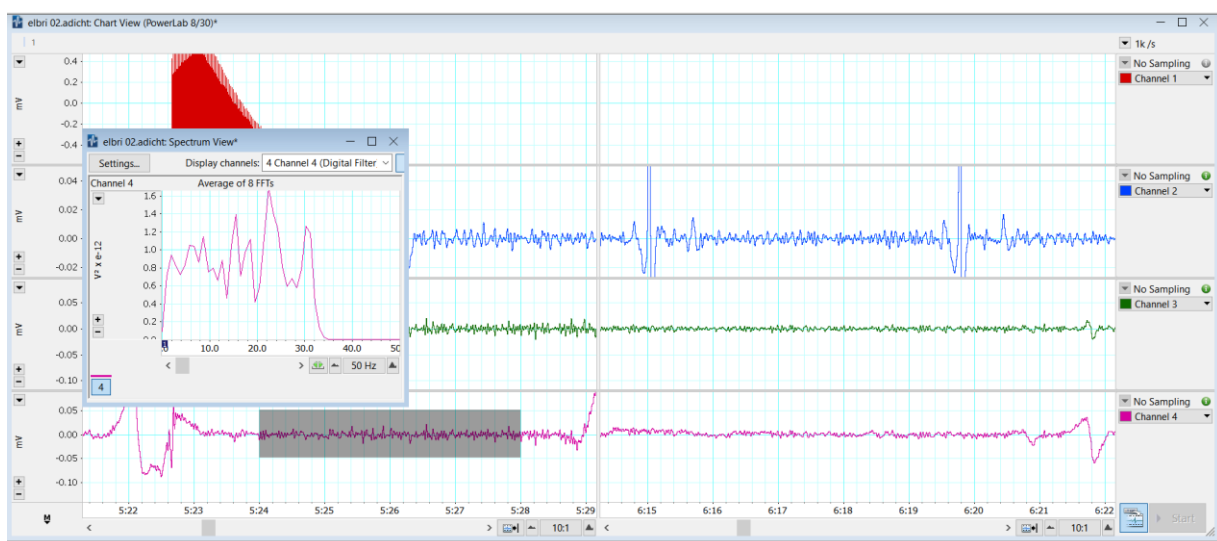


Figure 1B: Power analysis of EEG of a conscious Atlantic salmon (**frame in the left side**) and EEG and ECG in a stunned Atlantic salmon after immersion in the cold brine (**right side only**)

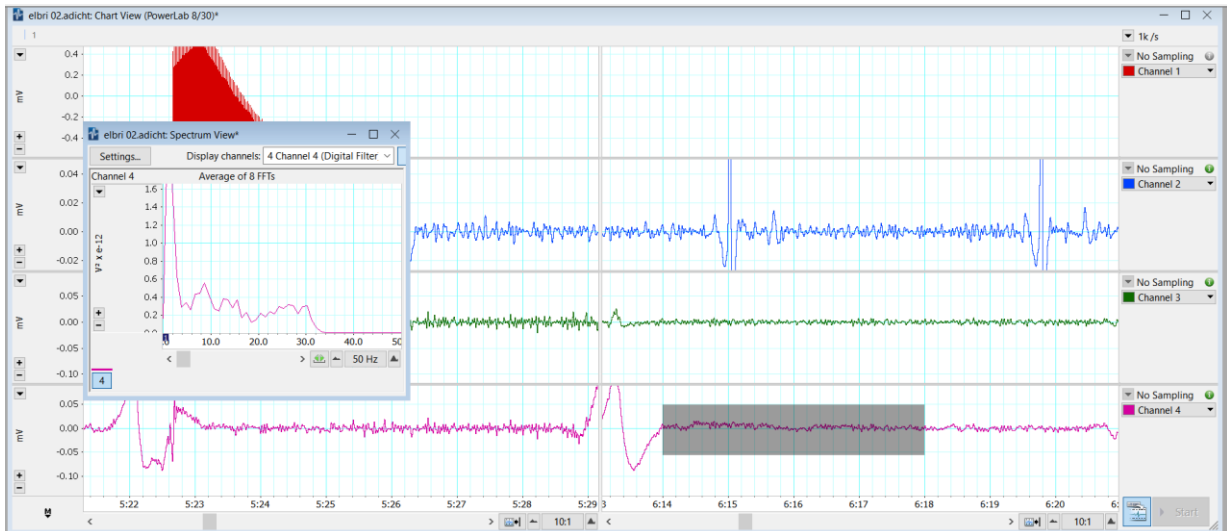


Figure 1C: EEG and ECG in a conscious Atlantic salmon (**left side**) after immersion in the cold brine (**right side**)

In all these figures the red line represents the raw EEG data, the blue line ECG data (bandpass filter of 4-25 Hz) and the green line the EEG data (bandpass filter 4-32 Hz). The purple line also represents filtered EEG data (bandpass filter 1-32 Hz).

A comparison of the power analysis of EEG prior to stunning (Fig 1B frame left side) and after immersion of the stunned fish in the cold brine (Fig 1 C frame left side) revealed a substantial decrease in the amplitude of frequencies that are characteristic for loss of consciousness, which is in accordance with e.g. data presented in a review on assessment of stunning methods of livestock (Verhoeven et al., 2015).

As demonstrated in Figure 2, measuring the electric potential difference across the brain is significantly correlated with the size of the fish, whilst doing a head stun ( $P < 0.05$ ,  $R = 0.64$ ; correlation). As expected, the relation between size and potential difference is logarithmic, whereas smaller fish with have higher potential difference across the brain due to lower resistance, not only size, but lower resistance due to thinner skin and lower fat content.

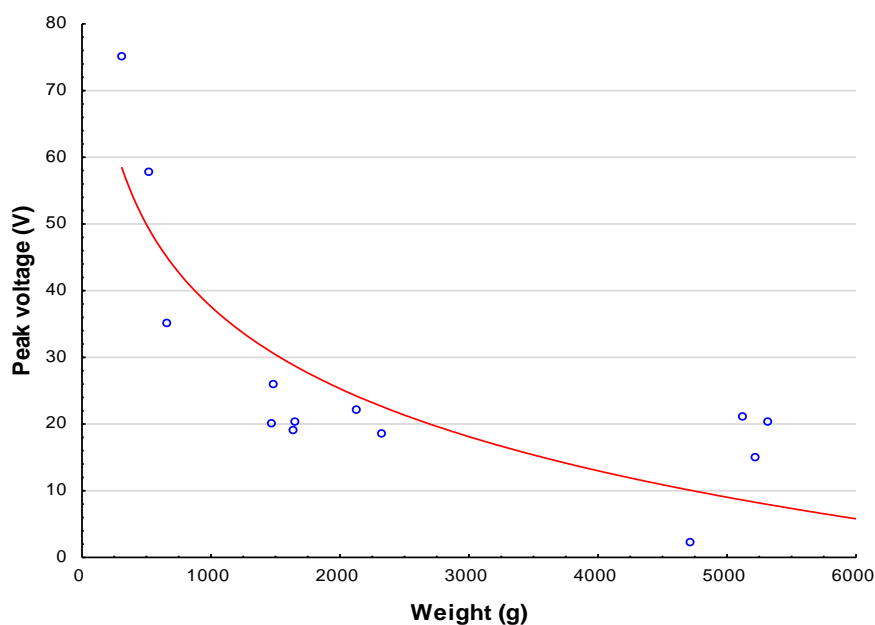


Figure 2: Potential difference across the brain (9 mm) as a function of size of Atlantic salmon

Placing fish in a cold brine of  $-14$  to  $-18$  °C will result in death, as the blood circulation will halt almost immediately as the gills and blood will freeze, then the brain and heart. As shown in Figure 2, the internal temperature both in the heart and brain cavity of smaller salmon rapidly drops to below  $-1$  °C until reaching the freezing temperature around  $-1.5$  to  $-1.8$  °C within 6 min (Figure 2). The time before freezing is highly dependent on size as salmon above 5kg will require almost 20 min before the brain starts to freeze. By that time EEG do show that the fish is already brain dead.

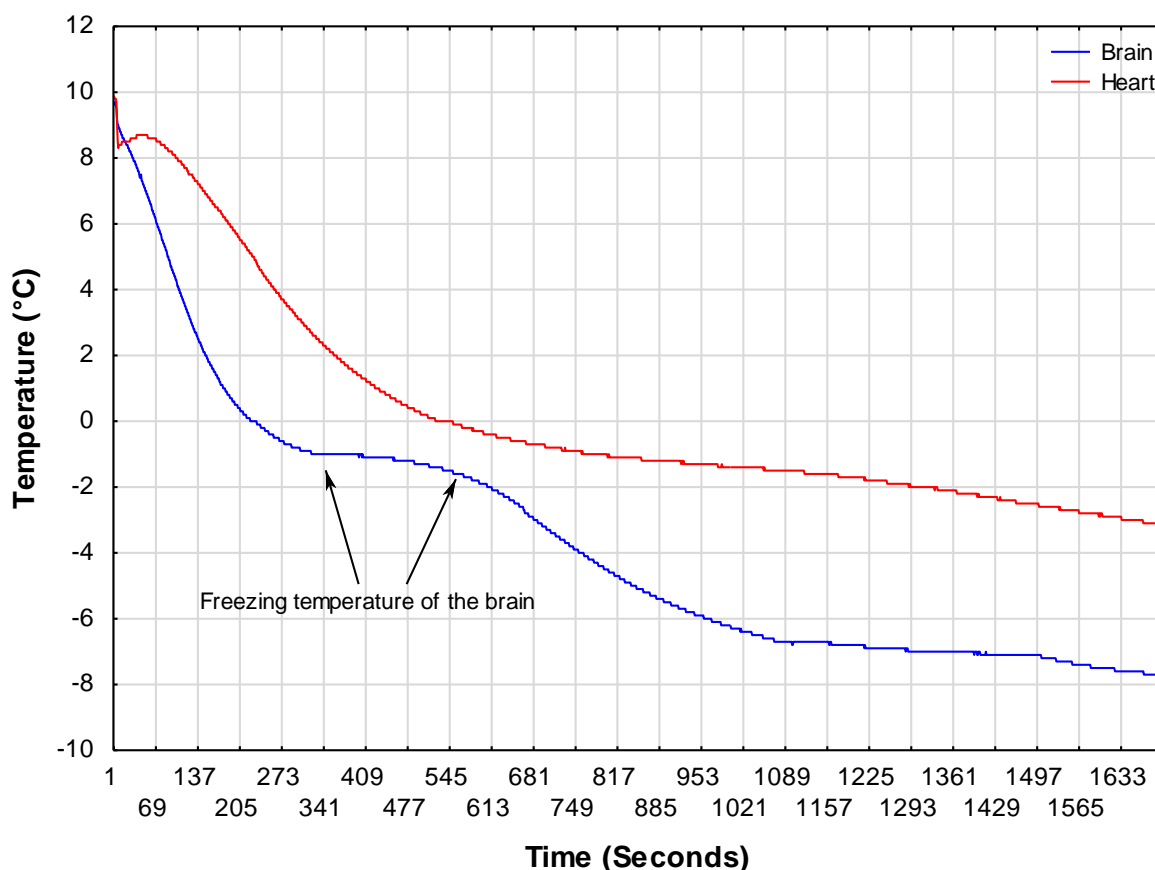


Figure 2: Internal temperature of the brain and heart cavity of small salmon placed in  $-18$  °C brine

## 5.1 Conclusion

The preliminary conclusion is that stunning Atlantic salmon with electrical dry stunning on from tail to head will result in unconsciousness within 0.5 seconds. Immersing unconscious fish into cold brine or water supersaturated with  $\text{CO}_2$  will result in permanent insensibility. Measuring the potential difference across the brain during stunning is a promising method to determine minimum requirements for stunning animals depending on size.

## 6 Main findings (should be written in both Norwegian and English)

- Changing the electric polarity on the first row, salmon can enter tail first in an electric stunner and still stunned unconscious within 0.5 second at 220 V, 50 Hz AC.
- Above 2 kg the fish must be entered head first.
- There is a negative correlation between size of the fish and electric potential difference across the brain.
- After stunning fish was efficiently killed using cold brine or seawater saturated with CO<sub>2</sub>

### 6.1 Hovedfunn

- Ved å endre den elektriske polariteten på den første raden, kan laksen gå med halen først inn en elektrisk bedøver og være bevisstløs innen 0,5 sekund ved 220 V, 50 Hz AC.
- Fisk over 2 kg skal føres med hodet først.
- Det er en negativ korrelasjon mellom størrelsen på fisken og elektrisk potensialforskjell over hjernen.
- Etter bedøvelsen ble fisken effektivt avlivet med kald saltlake eller sjøvann mettet med CO<sub>2</sub>.

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