

## **STSM Scientific Report**

### **1 Purpose of the STSM**

Study of temperature effect on European eel males along maturation. Analysis of 11 Ketotestosterone (11KT), estradiol (E2) and 17 $\alpha$ ,20 $\beta$ -dihydroxy-4-pregnen-3-one (17,20b-P) levels in blood plasma.

### **2 Description of the work carried out during the STSM**

In order to analyze the temperature effect on European eel male maturation, 300 animals were subjected to 3 different temperature regimes. Each week, samples from blood plasma were collected to analyse the plasma concentration of the following steroids: 17,20b-P, 11KT and E2. They have been measured by means of radioimmunoassay (RIA). Briefly, free (i.e., not conjugated) steroids were extracted from 250  $\mu$ l plasma with 4 ml diethyl ether under vigorous shaking for 4 min. The aqueous phase was frozen in liquid nitrogen, whereas the organic phase was transferred to a glass tube, evaporated in a water bath at 45 °C and then reconstituted by addition of 600  $\mu$ l assay buffer and then assayed for each steroid. To validate steroid recovery from plasma in the eel assay, plasma pools were spiked with 45 ng 17,20b-P, 11KT and E2 per ml of plasma and then subjected to ether extraction as described above. Products resulting from the different treatments were then assayed by RIA. A plasma steroid dilution curve parallel to that of the assay standard curve was established. The inter- and intra-assay coefficients of variation (CV) for the each steroid assay were calculated.

### **3 Description of the main results obtained**

The temperature is an essential environmental factor in teleost reproduction. In European eel, fish reared at higher temperatures have showed steroids levels with earlier and narrow peaks, whereas males at 10 °C showed delayed and wider peaks. Taking a count these results, the temperature is shown as important factor for gonad development, and it should be considered in the future to obtain a successful reproduction under captivity in this species.

### **4 Future collaboration with host institution**

Currently, both institutions we are partners in the European project: Reproduction of European eel: towards a self-sustained aquaculture (PRO-EEL). This stay intensifies this cooperation, opening the possibilities to new collaboration in future projects.

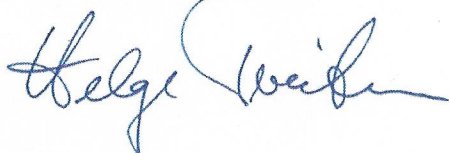
### **4 Foreseen publications/articles resulting or to result from the STSM**

Putting together these results with the gene expression of steroidogenic enzymes under three thermal regimes, we consider that the experiment will be finished and ready to be published in a journal with high scientific level: PLoS ONE (IF: 3.730; Q1).

## 5 Confirmation by the host institution of the successful execution of the STSM

Dr. Helge Tveiten, senior scientist from Nofima research centre (Tromsø, Norway) certifies that Dr. David Sánchez Peñaranda visited us as a guest researcher from May 18th to June 28th of 2013 thanks to a grant awarded by Short Term Scientific Mission, COST Action FA1205. Dr. Peñaranda has been trained and performed the analysis for the sex steroid quantification in eel plasma by RIA methodology, demonstrating maturity as an independent researcher.

Signed: Helge Tveiten

A handwritten signature in blue ink, appearing to read 'Helge Tveiten', with a stylized, flowing script.

Signed: David S. Peñaranda

A handwritten signature in blue ink, appearing to read 'David S. Peñaranda', with a stylized, flowing script.