



Short Term Scientific Mission (STSM) Report

- Reference code: COST-STSM-FA1205-34403
- STSM applicant: Ainara Valencia, PhD student
- Home institution: Cell Biology in Environmental Toxicology Research Group, Department of Zoology and Animal Cell Biology, University of the Basque Country UPV/EHU, Spain
- Host: Dr. Rüdiger W. Schulz, Associate Professor
- Host institution: Reproductive Biology Research Group, Division Developmental Biology, Department Biology, Science Faculty, Utrecht University, The Netherlands
- Period: 26/04/2016-23/07/2016

Work during the STSM period

- Testis tissue culture technique using *Danio rerio* zebrafish: 4 days zebrafish testis tissue culture using basal conditions was performed using testis from mature male zebrafish. Skills on dissection of zebrafish testis, preparation of basal medium and the agar cylinders used for the culture were obtained.
- Histological analysis of zebrafish testis: after the 4 days testis tissue culture, testis were collected and processed for histological analysis, in order to confirm the adequate maintenance of the testis morphology and cell composition and structure after the culture period. Skills on identification of different germ cell types in testis were obtained. The proportion of the area occupied by each germ cell type was calculated based on the abundance of each germ cell per testis tissue. In addition, sections from testis that had been treated with the cell proliferation marker 5-bromo-2'-deoxyuridine (BrdU) for 6 hours were also analyzed to calculate the mitotic index, by identifying and counting specific cells that had or had not incorporated the BrdU marker.
- In situ hybridization technique: in situ hybridization (ISH) was used for the detection and localization of spermatogenesis related genes in adult zebrafish testis. Riboprobes of the selected genes were synthesized and used in two different procedures; the first one using cryosections of the testis and the second one using paraffin embedded sections. Protocols were optimized to the tissue and experimental conditions.
- Histological analysis of *Chelon labrosus* thicklip grey mullet testis: sections of mullet testis, brought from the home institution were analyzed to identify the different germ cells present in the testis of mature males. Special interest was put on the identification of the different spermatogonial germ cells.



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Future work

The techniques learned in the host research group will be applied at the home institution using thicklip grey mullet testis, which will be helpful for a better understanding of mullet gametogenesis and especially for the understanding of endocrine disruption processes affecting the spermatogenesis in this species, such as the intersex condition.

Confirmation by the host institution of the successful completion of the STSM

Dr. Rüdiger W. Schulz from the University of Utrecht certifies that Ainara Valencia has completed a STSM partially funded by COST Action FA1205 AQUAGAMETE during the period 26/04/2016-23/07/2016 in the University of Utrecht, The Netherlands.

Rüdiger W. Schulz