

GENE EXPRESSION PROFILING DURING RAINBOW TROUT ONTOGENESIS REVEALS SUCCESSIVE CHANGES GATED IN TELEOSTS IN GERM STEM CELL MOLECULAR SIGNATURE AND REGULATORY PATHWAYS.

Maouche,A. Curran, E. Goupil, A,S. Sambroni, E. Le Gac, F. Lareyre J,J

INRA UR1037, Laboratory of Fish Physiology and Genomics, Campus de Beaulieu, 35042 Rennes cedex, France. Email: jean-jacques.lareyre@inra.fr

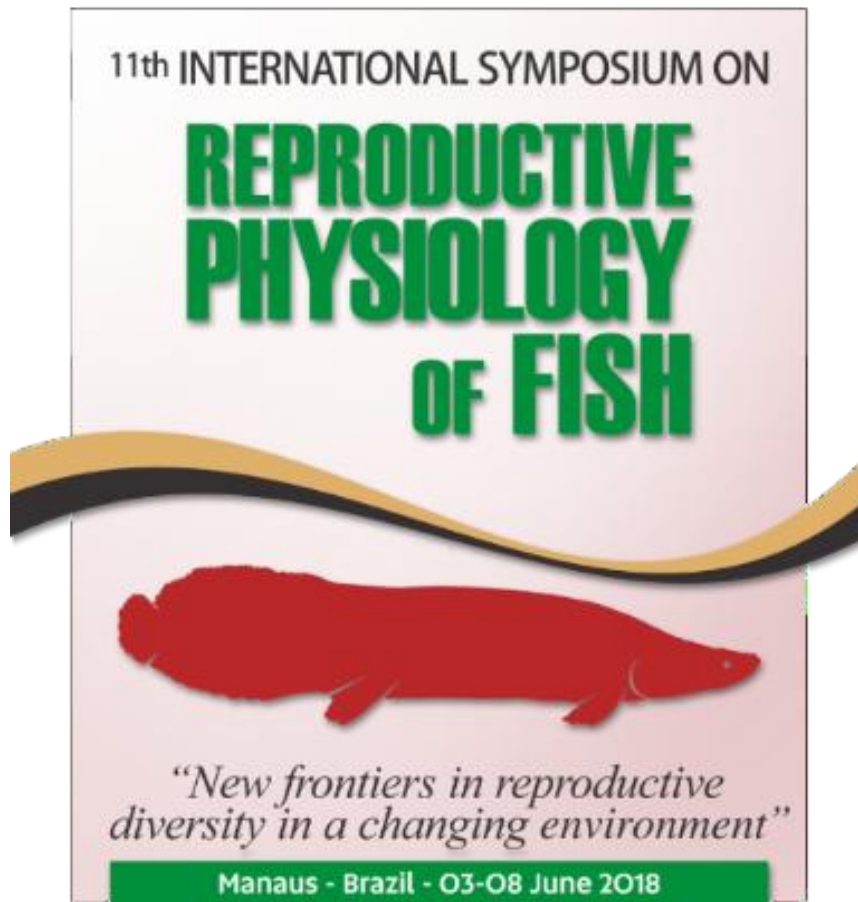
Introduction: The spermatogonial stem cells (SSCs) are adult male germ stem cells and they represent a narrowed population of undifferentiated spermatogonia. SSC have the ability to continuously self-renew and/or to differentiate ultimately into spermatozoa throughout adult male lifespan. In mammals, there is evidence that SSCs derived originally from primordial germ cells (PGCs) and both cell types are submitted to different micro-environments formed by somatic “germinal niches” that regulate their survival, migratory properties, self-renewal and/or differentiation. However, regulatory pathways that may be functional in the successive germinal niches remain poorly investigated in Teleosts. Our study was aimed to investigate potential changes in regulatory pathways that may regulate germ stem cell fate during late rainbow trout ontogenesis. The *kitlg/kitr* and Glial cell line-derived neurotrophic factor (*gdnf*)/ glial cell line-derived neurotrophic factor receptor alpha 1(*gfra1*) systems are two major regulatory pathways involved in mammalian PGCs and SSCs proliferation in embryo and adult male, respectively.

Methods: Phylogenetic reconstructions and analysis of syntenic chromosomal fragments were carried out from the genome of different vertebrate species to investigate the evolutionary conservation of the candidate genes and to establish reliable orthologous relationships. The temporal expression profiles of the candidate genes were determined in trout testes from 55 to 180 days post fertilization.

Results and Discussion: All candidate genes were conserved in cartilaginous and bony fishes but the latter showed taxon-specific gene duplications and losses. For instance, the teleost-specific *gfra1a* gene copy was further duplicated in Rainbow trout and Atlantic salmon whereas *gfra1b* gene copy was not found in both genomes and in mRNA expression databases. A different evolutive history was observed with *kitr* (duplication of the *kitra* gene copy whereas a single *kitrb* gene copy was retained in salmoninae). Temporal expression profiles showed high expression levels of paralogous *kitr* genes (*kitlgb*, *kitrb* and *kitra2*) at 55 days followed by a sharp decreased expression suggesting a role in germ stem cell fate before spermatogenesis onset. Interestingly, the *gfra1a1*, *gdnfa1* and *gdnfb* genes were barely expressed before spermatogenesis onset. Genes involved in the *gdnf/gfra1* regulatory pathways were highly expressed after 90 -120 days corresponding to spermatogenesis onset and the apparition of first progenitors. Gene expression profiling of additional germ cell specific genes indicate that the molecular signature of undifferentiated spermatogonia change during ontogenesis.

Conclusion Our study demonstrates that the germ stem cell entities and germinal environment change drastically during fish ontogenesis, well before and after spermatogenesis onset.

PROGRAM AND ABSTRACTS



**11 ISRF
2018**