DE NOVO ASSEMBLY AND CHARACTERIZATION OF THE OVARIAN TRANSCRIPTOME IN SWORDFISH *XIPHIAS GLADIUS* (LINNAEUS, 1758): FOCUS ON GENES DRIVING VITELLOGENIC PROCESS.

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Keywords: Swordfish, vitellogenesis, RNA-seq, differential expression

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INTRODUCTION: The swordfish *Xiphias gladius* (Linnaeus, 1758) is a large, highly migratory mesopelagic species with a global distribution and seasonal migrations. An important swordfish fishery has developed over the last 20 years but catches started to decline from the 1980s. To date, information on swordfish reproduction is lacking and does not provide comprehensive insights into gonadal development, which are necessary to determine their reproductive output. Knowledge on female gonadal development is required to establish the duration of the spawning season, the size and age of maturity and spawning patterns. The aim of the present project was a *de novo* assembly and annotation of the transcriptome of X. gladius. Moreover, a differential expression analysis was performed in order to compare pre-vitellogenic and vitellogenic ovaries, highlighting all genes driving vitellogenic process in swordfish.

METHODS: The Illumina HiSeq 2000 paired-end sequencing platform was adopted to sequence RNA isolated from 3 pre-reproductive and 3 reproductive ovaries. After quality check and trimming of low quality reads the transcriptome assembly was performed with Trinity v2.5.1. Then, the overall quality of the assembly was assessed by means of the BUSCO3 pipeline. Next, quantification of the assembled transcripts and identification of differentially expressed genes were achieved through the software Kallisto and the package NOISeq (False Discovery Rate: <=0.01), respectively. Finally, a Gene Ontology Enrichment Analysis (GOEA) and a Kyoto Encyclopedia of Genes and Genomes (KEGG) Pathway Analysis was performed. To validate the results, qPCR was carried out for a pool of genes of interest.

RESULTS: A total of 100.869 sequences with N50 of 2.037 bp were generated from cDNA libraries of pre-vitellogenic and vitellogenic ovaries and a GO annotation was assigned to 30.398 transcripts. On the other hand, we identified 25.151 unigenes of which 22.433 and 2718 were annotated in Trembl and Swissprot database, respectively. The differential expression analysis between pre-vitellogenic and vitellogenic ovaries revealed a total of 6.501 transcripts differentially expressed, with 2494 predicted to be up-regulated in pre-vitellogenic ovaries. Furthermore, GOEA highlighted that the most differentially expressed transcripts within the category “Biological Process” were related to RNA/DNA processing, cell cycle regulation, endosome organization and transport and lipid metabolism. In addition, 2862 unigenes were mapped to 361 pathways in the KEEG database revealing that the most affected pathways are RNA/DNA processing, ovarian steroidogenesis, autophagy and apoptosis processes and lysosome formation/maturation.

CONCLUSIONS: This study provides the first *de novo* transcriptome analysis currently available for X. gladius, and identify many important functional genes, GO terms and KEGG pathways involved in swordfish oocyte quality. Results of the present study will facilitate future studies on swordfish reproduction.

Funding

This work was supported by Ministry of Agriculture, Food and Forestry Policies (MIPAAF), note 6775, Art.36 Paragraph 1 Reg (UE9 n 508/2014) to O.C.