GLOBAL AND LOCAL METHYLATION PATTERN IN TRIPLOID AND DIPLOID ATLANTIC SALMON

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Summary

Background: Induction of triploidy is a common way to induce sterility in aquaculture. Recently protocols for this technology have also been adapted to industrial scale Atlantic salmon production. The main explanation for late commercial implementation is the reduced culture performance of triploids observed in previous studies. Problems related to triploidy in Atlantic salmon has been indentified such as low tolerance to high temperature in seawater, susceptibility to skeletal deformities, cataracts and heart deformations. Little is known about what may cause these phenotypes in triploids. The answer might be found in how they regulate their additional alleles. It is further unknown how triploid fish use or regulate the extra chromosomal copies. What is known is that the extra chromosomal copies do not seem to significantly cause an additive gene expressional response in fish. Likewise in plants where ploidisation is a common breeding technique, gene expression upon ploidsation is often non-additive. Plant studies have revealed epigenetic mechanisms behind this dosage compensation of gene expression including hypermethylation of DNA, histone acetylation and micro-RNA. It is unknown if such epigenetic mechanisms are initiated upon triploidsation in fish, but novel molecular techniques has enabled us to investigate if methylation of the genome could contribute to non-additive gene expressional regulation in triploids.

Aims: To investigate whether nonadditive gene expression upon triploidy in Atlantic salmon could be regulated by increased methylation of the genome.

Results: First we examined gene expression level of several genes in triploid and diploid Atlantic salmon, none of the investigated genes showed differential expression between triploid and diploids. Also we measured the DNA:RNA ratio and as expected it was significantly lower in triploid compared to diploid fish. To explore local methylation effects, we measured the CpG methylation level of the housekeeping gene elongationfactor 1α in whole 700 day˚ embryos, but no differences could be detected between triploid and diploid samples (n=10 per group Fig1). This result pointed at no general change in methylation level between triploid and diploid Atlantic salmon. To search for methylation differences on a global level we performed RRBS (Reduced Representation Bilsulfite Sequencing) on samples from whole diploid and triploid embryos (700 day˚). Obtained sequence material was mapped upon an early release of the salmon genome assembly. Initial results showed that global methylation level was similar between triploid and diploid Atlantic salmon. CpG islands and gene prediction revealed that in the sequenced material, only 1395 genes showed representative CpG islands. Of the 1395 genes 364 genes were more methylated (above 30% more) in triploid than diploids while 139 genes showed the opposite. In the group of overmethylated genes in triploids, genes related to protein turnover, heart formation, bone/cartilage formation and micro RNA regulation were detected.

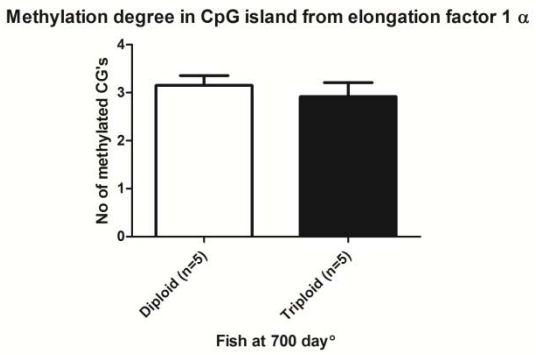

Fig1. Methylation level of promoter

Conclusions: Triploid Atlantic salmon shows non-additive gene expression. Further the general mechanism of non additive expression does not seem to be due to hypermethylation of the whole genome. Although some specific changes in methylation seems to occur possibly related to protein turnover heart formation, bone/cartilage formation and micro RNA regulation.

Keywords: epigenetic, ploidy, RRBS, methylome, CpG islands