

## Sequence variation of the Pantophysin (*PanI*) in Atlantic cod (*Gadus morhua*): Comparison of two locations

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Pantophysin (*PanI*) is an integral membrane spanning protein in cytoplasmic vesicles but its function is not exactly known. In Atlantic cod (*Gadus morhua*) there are two major allelic lineages known *PanIA* and *PanIB* and they have been known to show a signature of natural selection (Pogson, 2001). The alleles have been found to be highly divergent on the nucleotide level in several Atlantic populations (Pogson, 2001). Cod which was captured in springtime and wintertime living in deep sea (approx. 400 meters) North of Iceland will be compared to cod living in shallower waters at the South coast of Iceland. Genotyping will be performed by utilizing a polymorphism in the *DraI* restriction site. The DNA sequence of the gene is about 1.94 Kb but a 1051 bp long fragment of the gene is produced in a PCR reaction. All genotypes have one fixed restriction site for the *DraI* restriction enzyme which cuts the 1051 bp fragment to 773-bp fragment and 278 bp fragment. Individuals which are homozygous AA therefore produce two fragments in digestion with *DraI*. On the other hand individuals which are homozygous BB have a polymorphic restriction site in the 773-bp fragment and produce two fragments, a 495-bp long fragment and the 278 bp-fragment (Fevolden and Pogson, 1997). Pogson (2001) analysed the sequence variation in *PanI* by utilizing the fact that the restriction enzymes *BstXI* and *SacII* cut genomic DNA of one or the other allele in heterozygotes. After digestion the other allele can be amplified and sequenced. However, problems arise due to incomplete digestion which results in overlapping sequences. Another approach has been developed during the course of this project. Homozygous individuals have been selected and the whole 1.94-bp fragment has been amplified and isolated from electrophoresis gels. The fragment has been cloned in *E. coli* and the clones sequenced. The object of this project is to test for natural selection by using genotype frequencies and haplotype information from sequencing of the whole 1.94-bp fragment.