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COMPARATIVE ANALYSIS OF SEQUENCES FROM PT 2013

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Abstract:

Every year The European reference Laboratory offers two proficiency tests for all national Reference Laboratories in Europe as well as any other country that wants to participate. In 2013 43 laboratories participated in at least one of the two proficiency tests that cover all the listed fish diseases in Europe.

As part of the EURL proficiency test for fish diseases it is required to sequence any RANA virus isolates found in any of the samples. It is also highly recommended to sequence the ISA virus to determine whether it be HPRΔ or HPR0. Furthermore, it is recommended that any VHSV and IHNV isolates be genotyped.

As part of the evaluation of the proficiency results it was decided this year to look into the quality and similarity of the sequence results for selected viruses.

Ampoule III in the proficiency test 2013 contained an EHNV isolate. The EURL received 43 sequences from 41 laboratories. All but one sequence mapped to the MCP gene while the last sequence mapped to the Neurofilament gene. Approx. half of the sequences contained no errors while the rest differed with 88-99 percent similarity with most having 99% similarity. One sequence, when BLASTed, showed most similarity to European Sheatfish and not EHNV.

Generally, mistakes occurred at the ends of the sequences. This can be due to several factors. One is that the sequence has not been trimmed of the sequence primer sites. Another is the lack of quality control of the chromatogram. Finally, sequencing in just one direction can result in unclear determination of nucleotides at places with a bad quality score.

This talk will present some of the problems that can occur with sequencing as well as discuss potential pitfalls.