Evaluation of tangential flow filtration coupled to long-read sequencing for Ostreid Herpesvirus type 1 genome assembly

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Since the 90's, the Pacific oyster *Crassostrea gigas* has suffered significant mortality events associated with the detection of the Ostreid Herpesvirus 1 (OsHV-1). Genomes of such viruses are large and complex with long direct and inverted terminal repeats. To date, the diversity of OsHV-1 has been mainly characterized via microsatellites or multi-genomic fragment analyses and more recently via high-throughput sequencing based on the short-read approach (Illumina). Despite the high accuracy of this type of sequencing, it cannot be used to identify and characterize structural variation and isoforms of such complex viral genomes. Recently, long-read sequencing techniques like nanopore sequencing from Oxford Nanopore Technologies (ONT) have been developed and offer a solution to the short-reads sequencing limitations.

Since OsHV-1 cannot be cultured, we developed a tangential flow filtration (TFF) method to enrich for viral infective particles from infected host tissues. This virus purification allowed us to extract high molecular weight and high-quality viral DNA that was processed to Illumina short-read and Nanopore long-read sequencing. Dedicated bioinformatic pipelines were developed to assemble complete OsHV-1 genomes with reads from both sequencing technologies. Nanopore sequencing was used to characterize new structural variations and major viral isomers while having 99,98% of nucleotide identity with the Illumina assembled genome.

Altogether, our results strongly suggest that the TFF-based purification method, coupled with Nanopore sequencing, is a promising approach to enable field sequencing of unculturable aquatic DNA virus.