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Genetic analysis and comparison of segment A of aquatic birnavirus isolated from rockfish *Sebastes schlegeli* in Korea

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Introduction

The aquatic birnaviruses have a bisegmented, double stranded RNA genome (Cohen et al., 1973; Dobos et al., 1979; Dobos & Roberts, 1983). The genome segment A (approximately 3100 bp) contains 2 partially overlapping open reading frames (ORF) (Duncan et al., 1987; Havarstein et al., 1990). The large ORF encodes a major capsid protein VP2, a minor capsid protein VP3 and a non-structural protein NS. The coding order for these polypeptides within the virion genome is NH_2-pVP2-NS-VP3-COOH. The small ORF encodes a 17 kDa arginine-rich minor polypeptide VP5. The genome segment B (2784 bp) is monocistronic and encodes an internal polypeptide VP1, the putative virion-associated RNA-dependent RNA polymerase. This study was performed to characterize the virus genome on segment A. For this study, nucleotide sequence was analyzed and compared the sequence to that of the previously reported aquatic birnaviruses.

Materials and methods

The aquatic Birnavirus (GC-1) was isolated from rockfish (*Sebastes schlegeli*) reared in Gochang, North Jeonla province, Korea, characterized and maintained in our laboratory (Seo et al., 1988). The GC-1 was propagated in chinook salmon embryo cells (CHSE-214) and the genomic RNA was extracted from the concentrated and purified virus. Seven Primer pairs used for RT-PCR. The oligonucleotide sequences were deduced according to the published sequences of

the Y-6 strains (Sato et al., 1999). The RT-PCR products were cloned and sequenced. The sequences were analyzed by Dnasis program.

Results and summary

The nucleotide sequence of segment A was 3,086 bp long and contained a large open reading frame (ORF) of 2,916 bp and a small ORF of 399 bp. The large ORF of the GC-1 started at nucleotide 117 and ended with a single TAA termination codon at nucleotide 3,033 and contained 5'-VP2-NS-VP3-3' genes. The small ORF was also existed in GC-1. The nucleotide was started at nucleotide 110 and ended with a single TGA termination codon at nucleotide 511 encoding 133 amino acids.

The large ORF of the GC-1 encodes 972 amino acid precusor polyproteins with a predicted molecular weight of 106,726 daltons. The postulated cleavage sites of GC-1 is located #490-503 between VP2 and NS, and #721-729 between NS and VP3.

The homology of large ORF with Y-6 was 98.1%, with DRT and Jasper was 86.9%, and Sp and N1 was 81.5%. This result indicated that GC-1 has the nearest relationship and more similar to genogroup I (DRT and Jasper) than genogroup II (Sp and N1). The homology of small ORF with Y-6 was 95.5%, with IPNV group I, containing VR299, Jasper, and DRT was average 80.5%, with IPNV genogroup II containing Ab, Fr.21, N1 and Ca2 was average 64.0%. The percentage of relative homolgy was lower than that of the large ORF, when compared with other aquatic birnaviruses.

References

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