

Development of an Isavirus minigenome system to study the function of the pocket RNA-binding domain of the viral nucleoprotein (NP) in salmon cells

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The Isavirus is an orthomyxovirus with a genome composed of eight segments of negative single-strand RNA (ssRNA). It has been proposed that the eight genomic segments of the Isavirus are organized as a ribonucleoprotein (RNP) complex called a minigenome, which contains all the viral RNA segments, a viral heterotrimeric polymerase and multiple copies of the viral nucleoprotein (NP). Here, we develop an Isavirus minigenome system and show the importance of the formation of active RNPs and the role of viral NP R189, R194, R302 and K325 residues in the NP RNA-binding domain in the context of RNPs. The results indicate it is possible to generate a minigenome in salmon cells, a composite ISAV RNPs with EGFP-based chimeric vRNA with heterotrimeric polymerase (PB1, PB2, PA) and NP protein using CMV-based auxiliary plasmids. It was also shown that NP R189, R194, R302 and K325 residues are important to generate viral mRNA from the constituted RNPs and a detectable reporter protein. This work is the first salmon cell-based minigenome assay for the Isavirus, which was evaluated by a bioinformatic and functional study of the NP protein in viral RNPs, which showed that correct NP-vRNA interaction is key to the

infectious salmon anaemia virus

minigenome assay

nucleoprotein

orthomyxovirus

salmon

messenger RNA

virus nucleoprotein

ribonucleoprotein

viral protein

animal cell

Article

assay

Isavirus

nonhuman

plasmid

RNA-binding domain

salmonine

virus genome

animal

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Isavirus

RNA-binding domain

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Animals

Genome, Viral

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Isavirus

Ribonucleoproteins

RNA-Binding Motifs

Salmo salar

Viral Proteins