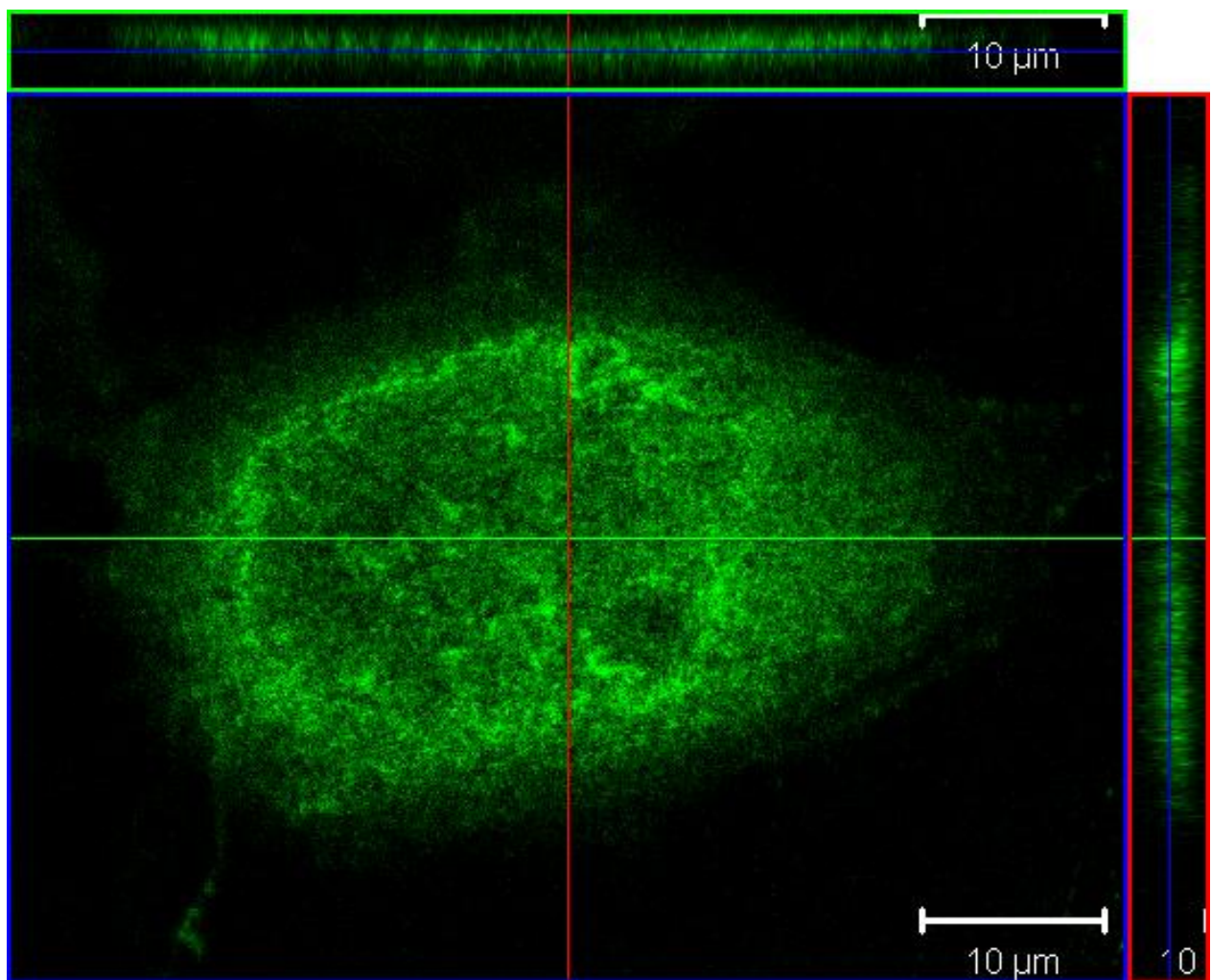


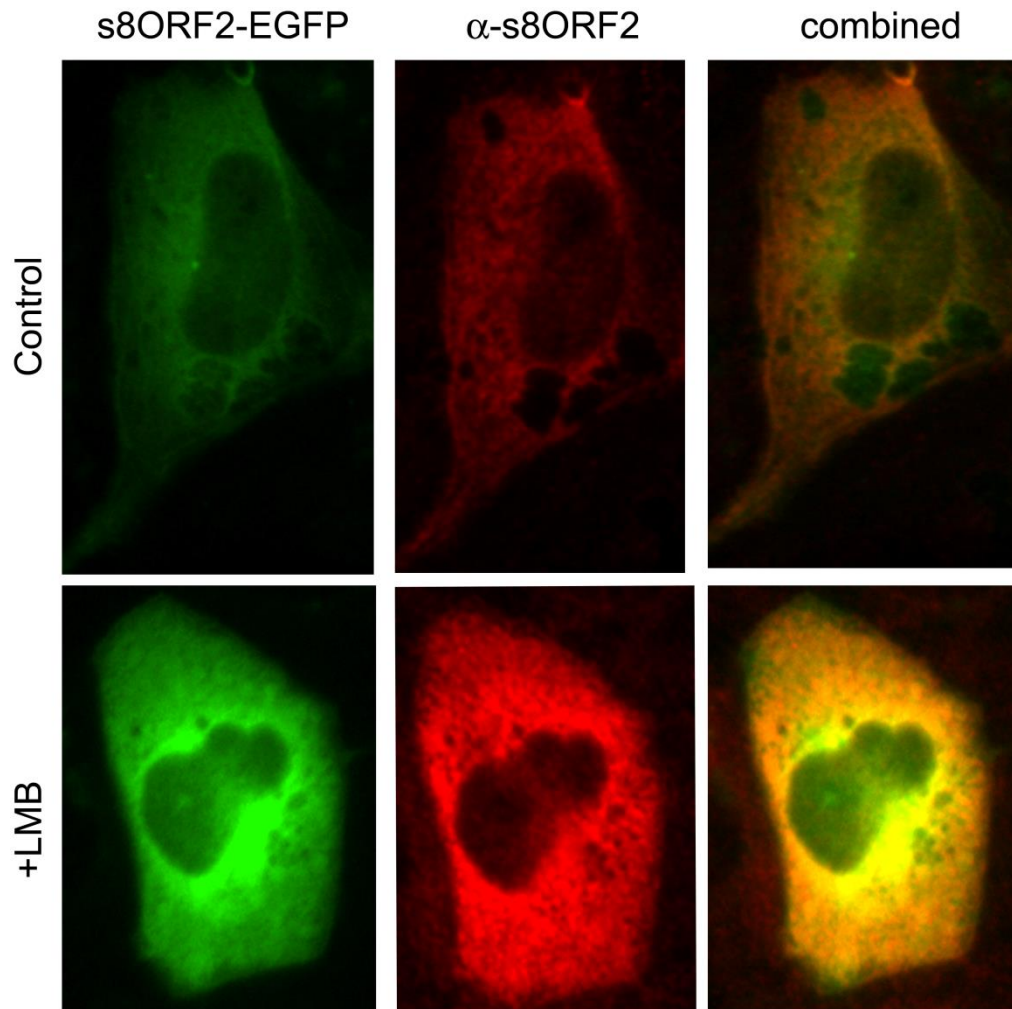
# Supplementary Materials: Infectious Salmon Anaemia Virus (ISAV) RNA Binding Protein Encoded by Segment 8 ORF2 and Its Interaction with ISAV and Intracellular Proteins

Christel M. Olsen <sup>1</sup>, Turhan Markussen <sup>1</sup>, Bernd Thiede <sup>2</sup> and Espen Rimstad <sup>1,\*</sup>

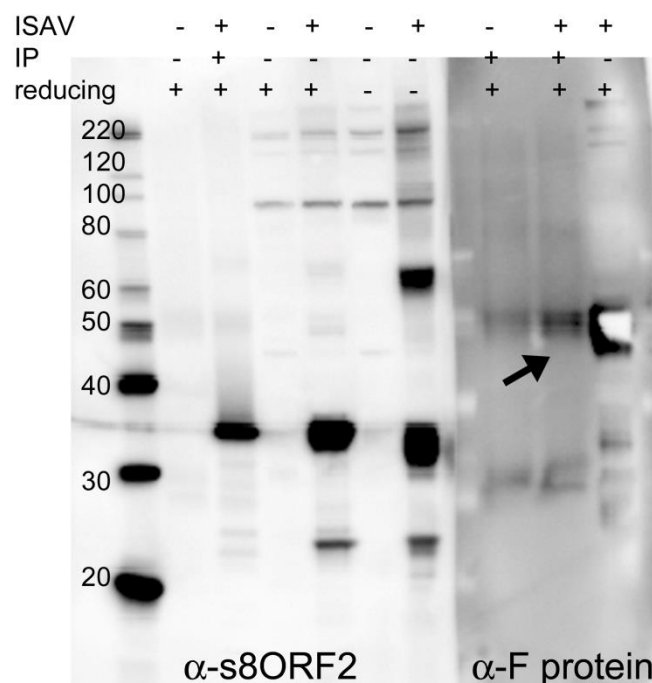
**Supplementary Materials:** The following are available online at [www.mdpi.com/link](http://www.mdpi.com/link), Figure S1: S8ORF2 protein is present in both the cytosol and nucleus of some ISAV infected ASK cells, Figure S2: Nucleo-cytoplasmic shuttling is not affected when s8ORF2-EGFP transfected cells are treated with Leptomycin B to inhibit CRM-1 dependent nuclear export, Figure S3: Western blot of co-immunoprecipitation (IP) assays with s8ORF2 and F protein.



**Figure S1.** S8ORF2 protein is present in both the cytosol and nucleus of some ISAV infected ASK cells. Picture shows x,y,z dimensions of confocal Z-stack of one ASK cell immunostained with anti-s8ORF2 antiserum 72 hpi. (Z = 3.71 μm). Picture was taken at 63× magnification in a confocal microscope.



**Figure S2.** Nucleo-cytoplasmic shuttling of S8ORF2-EGFP protein is not affected when CRM-1 dependent nuclear export is inhibited by Leptomycin B. Chinook salmon embryo cells (CHSE-214) were transfected the same way as described for EPC cells with s8ORF2-EGFP (control) or treated for 4 h with 40  $\mu$ M LMB before immunostaining with anti-s8ORF2 antiserum 24 h post transfection. Pictures were taken in inverted fluorescence microscope at 40 $\times$  magnification and show EGFP (green), anti-s8ORF2 (red) and combined signals (yellow).



**Figure S3.** Western blots of co-immunoprecipitation (IP) assays with s8ORF2 and ISAV protein F. The anti-s8ORF2 blot is the same as in Figure 1, and the membrane developed by anti-F antiserum indicate that when using anti-s8ORF2 in pull-down this also pulls down proteins in the non-infected cell with same molecular weight as F-protein (arrow). Compared to the direct lysate (-IP) small amounts of F protein is pulled down with anti-s8ORF2 (arrow), however the interpretation of this result is disturbed by the non-specific signal with the same MW in IP from non-infected cells.



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