

Reconfiguring the ambi-sense S genome segment of Rift Valley fever virus.

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Rift Valley fever virus (RVFV, family *Bunyaviridae*) is a mosquito-borne pathogen of both livestock and humans, found primarily in Sub-Saharan Africa and the Arabian Peninsula. The RVFV genome comprises 3 segments of RNA designated L, M and S. The L and M segments are conventional negative-sense RNAs. The S segment is ambi-sense, encoding N protein in the negative-sense and a non-structural protein, NSs (the viral interferon antagonist), in the positive-sense. The ambi-sense strategy was originally suggested as providing temporal control over gene expression as NSs would be translated relatively late in the infectious cycle after genome replication had commenced. We are investigating the consequences of swapping the N and NSs coding sequences on the S segment by directly substituting N into the NSs locus and NSs into the N locus. A recombinant virus was recovered by reverse genetics and called S-SWAP. In both mammalian and insect cells S-SWAP is attenuated and has a small plaque phenotype. Analyses of viral protein and RNA synthesis suggest that the correct temporal expression of NSs is required for efficient replication, and further that genes expressed from the NSs locus of RVFV may be targeted by the RNAi response of insect cells.

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