

POSTER 32**THE LYPSL MOTIF ACTS AS A SECONDARY LATE DOMAIN FOR ROUS SARCOMA VIRUS**

Kari A. Dilley¹, Devon Gregory², Marc C. Johnson², and Volker M. Vogt¹

¹Cornell University, Ithaca, NY 14850; ²University of Missouri, Columbia, MO 65211

The efficient release of newly assembled retrovirus particles from the plasma membrane requires the recruitment of a network of cellular proteins (the ESCRT machinery) normally involved in the biogenesis of multivesicular bodies (MVBs). It is known that retroviruses, as well as other enveloped viruses, recruit this ESCRT machinery through short motifs termed late domains. Retroviruses use three classes of late domains as docks for the ESCRT machinery; PT/SAP, PPXY, and LYPX_nL. The late domain of Rous sarcoma virus (RSV) has been mapped to a PPPY motif in Gag that binds members of the Nedd4-family of ubiquitin ligases. RSV Gag also contains a second putative late domain motif, LYPSL, positioned five amino acids downstream of PPPY. LYPX_nL motifs have been shown to support budding in other retroviruses by binding Alix/AIP1. To investigate the role of this YPSL motif in RSV budding we measured budding rate, spreading rate, and examined budding phenotypes with SEM of various PPPY and LYPSL mutants in the context of infectious RSV. We found that mutating the LYPSL motif alone had no consequences on the budding or spreading rate of RSV. However, the role of the LYPSL motif in budding and spreading LYPDL motif acts as a stronger late domain than the LYPSL. Furthermore, the over-expression could be uncovered when the primary PPPY late domain was fully or partially mutated. In this same context we also showed that the related EIAV of Alix partially rescued RSV budding and spreading rate of PPPY mutants in a LYPSL-dependent manner. Taken together these results support a model of RSV budding in which the LYPSL motif acts as a secondary late domain via its interaction with Alix.