

Signaling networks in influenza virus assembly and budding

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Assembly and budding of influenza A virions is a tightly regulated process orchestrated by matrix protein M1. We showed that phosphorylation of M1 at Y132 drives efficient virus assembly and budding. Here, we identify signaling pathways involved in M1-induced assembly and budding and aim to pin-point signaling networks decisive for virion morphology. M1 phospho-site mutants were generated via reverse genetics. Virus budding phenotypes and morphological changes were evaluated using atomic force microscopy (AFM) and cryo-electron tomography (cryo-ET), respectively. To examine host cell signaling, kinase activity profiling of cells infected with M1 mutant viruses or wild type strains of spherical, filamentous or mixed morphology was performed using PamGene technology. M1 mutant virions changed to a more filamentous morphology or retained a predominant spherical shape with slight differences in size. Kinome activity analysis revealed a strong activation of S/T kinases by filamentous and viruses of mixed morphology during budding that was not observed with spherical strains and cannot be attributed to virus replication efficiencies. Overall, activity of Y kinases was increasingly reduced during late stages of budding independent of virion morphology. However, single M1 mutations were sufficient to induce very low if any activity changes of Y kinases compared to uninfected cells. Our data indicate that M1 possibly determines virion morphology via orchestrating S/T kinase signaling.

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