## **Supplementary Files**

**Movie S1.** NCLSs transport in VP24<sup>wt</sup> -expressing cells. Huh-7 cells were transfected with plasmids encoding NP, VP35, VP30-GFP, and VP24<sup>wt</sup>. Time-lapse microscopy analysis was started at 20 h p.t. Acquisition: Sequence displays 3 min, one frame was taken every 2-3 s. Scale bar, 10 µm.

**Movie S2.** NCLSs transport in VP24<sup>AxxL</sup> -expressing cells. Huh-7 cells were transfected with plasmids encoding NP, VP35, VP30-GFP, and VP24<sup>AxxL</sup>. Time-lapse microscopy analysis was started at 20 h p.t. Acquisition: Sequence displays 3 min, one frame was taken every 2-3 s. Scale bar, 10 µm.

**Movie S3.** NCLSs transport in VP24<sup>YxxA</sup> -expressing cells. Huh-7 cells were transfected with plasmids encoding NP, VP35, VP30-GFP, and VP24<sup>YxxA</sup>. Time-lapse microscopy analysis was started at 20 h p.t. Acquisition: Sequence displays 3 min, one frame was taken every 2-3 s. Scale bar, 10 µm.

**Movie S4.** NCLSs transport in VP24<sup>AxxA</sup> -expressing cells. Huh-7 cells were transfected with plasmids encoding NP, VP35, VP30-GFP, and VP24<sup>AxxA</sup>. Time-lapse microscopy analysis was started at 20 h p.t. Acquisition: Sequence displays 3 min, one frame was taken every 2-3 s. Scale bar, 10 µm.

Movie S5. NCLSs transport of VP24<sup>wt</sup>-expressing, si-Ctrl treated cells. Huh-7 cells were

treated with si-Ctrl prior to transfection of NP, VP35, VP30-GFP, and VP24<sup>wt</sup>. Time-lapse microscopy analysis was started at 20 h p.t. Acquisition: Sequence displays 2 min, one frame was taken every 1 s. Scale bar, 10 μm.

**Movie S6.** NCLSs transport in VP24<sup>wt</sup> – expressing cells, treated with si-Alix . Huh-7 cells were treated with si-Alix prior to transfection of plasmids encoding NP, VP35, VP30-GFP, and VP24<sup>wt</sup>. Time-lapse microscopy analysis was started at 20 h p.t. Acquisition: Sequence displays 2 min, one frame was taken every 1 s. Scale bar, 10 µm.

**Movie S7.** NCLSs transport in VP24<sup>AxxA</sup> –expressing cells, treated with si-Ctrl. Huh-7 cells were treated with si-Ctrl prior to transfection of plasmids encoding NP, VP35, VP30-GFP, and VP24<sup>AxxA</sup>. Time-lapse microscopy analysis was started at 20 h p.t. Acquisition: Sequence displays 2 min, one frame was taken every 1 s. Scale bar, 10 µm.

**Movie S8.** NCLSs transport of VP24<sup>AxxA</sup> –expressing cells, treated with si-Alix. Huh-7 cells were treated with si-Alix prior to transfection of plasmids encoding NP, VP35, VP30-GFP, and VP24<sup>AxxA</sup>. Time-lapse microscopy analysis was started at 20 h p.t. Acquisition: Sequence displays 2 min, one frame was taken every 1 s. Scale bar, 10 µm.