1	Sialic acid and PirB are not required for viral targeting of
2	neural circuits by neurotropic mammalian orthoreovirus
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17	Running Head: Reovirus neurotropism in the central nervous system
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regional relative infection density. (A) Nissl-stained sagittal brain slice from the Allen

24 Developing Mouse Brain Reference Atlas (Allen Developing Mouse Brain Reference

25 Atlas P14, developingmouse.brain-map.org and atlas.brain-map.org/) overlaid with regions of interest (ROI). Identical ROIs were individually overlaid onto all imaged 26 sections according to brain landmarks (1,2). (B) ROIs (green outlines) overlaid on a 27 Hoechst-dye-stained brain slice from a mock-infected animal. Brain regions are 28 indicated. DPall, dorsal pallium; MTt, collicular midbrain tectum; MPall, medial pallium; 29 PH, pontine hindbrain; PPH, prepontine hindbrain; Th, thalamus; Thy+PHy, 30 hypothalamus; SPall, subpallium. Some regions (such as DPall, MPall, and MTt) are 31 quantified from multiple ROIs. 32

33	Supplemental	Movie S1A.	Imaged v	olume of	reovirus-i	nfected	brain ti	issue
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visualized using MiPACT-HCR. Two-day-old WT mice were inoculated IC with 1000

³⁵ PFU T3SA+, and right-brain hemispheres were processed for MiPACT-HCR as

described in FIG 2. Tissue was imaged using MesoSPIM microscopy and processed

- into a video using Imaris software (Oxford Instruments). Reovirus RNA is depicted in
- green. Movie of sagittal optical sections across entire brain hemisphere. Scale bar, 1000

39 µm.

Supplemental Movie S1B. Imaged volume of reovirus-infected brain tissue visualized using MiPACT-HCR. Two-day-old WT mice were inoculated IC with 1000 PFU T3SA+, and right-brain hemispheres were processed for MiPACT-HCR as described in FIG 2. Tissue was imaged using MesoSPIM microscopy and processed into a video using Imaris software (Oxford Instruments). Reovirus RNA is depicted in green. Max projection of z-stacks formatted for movie demonstrating reovirus RNA staining at cell resolution in 3D. Scale bar, 50 µm.

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