

Figure S1. High magnification imaging of vasculature in infected larvae.

Confocal imaging, single focal planes extracted from Fig. (3A') and (3D'). Scale bars: 50 µm.

- (A) SINV infection live imaging, from Fig. (3A'); (A') merge of green (infected cells) and magenta (endothelium) fluorescence; (A'') green (infected cells) fluorescence; (A''') magenta (endothelium) fluorescence. White and yellow arrowheads: infected areas close to but not part of the vasculature.
- (**B**) CHIKV infection, fixed sample labeled for GFP and RFP, from Fig. (**3D'**). (**B'**) merge of green (infected cells) and magenta (endothelium) fluorescence; (**B'''**) green (infected cells) fluorescence; (**B'''**) magenta (endothelium) fluorescence. White and yellow arrowheads: areas of colocalization.

(C'-C''') SINV infection, fixation at 2 dpi followed by clarity treatment and immunolabeling for GFP and RFP, lateral views; (C') Entire larva, boxes indicate the magnified areas in (C'') and (C'''); (C'') 3D rendering of the heavily infected area encompassing gills, liver, and hindbrain. See **Movie 3**, for rotation of the 3D structure; (C''') High magnification of the infected area in the tail region. White arrowheads: infected cells close to but not part of the vasculature. Scale bars: 150 μm.

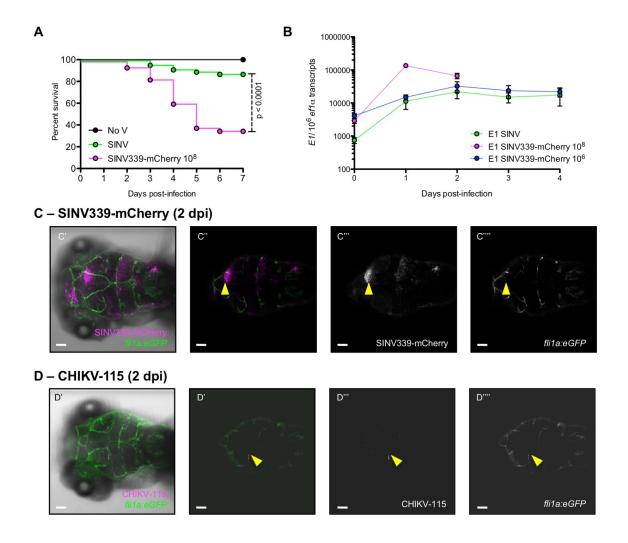


Figure S2. Infection of zebrafish larvae with other SINV and CHIKV strains.

(**A** and **B**) Characterization of SINV339-mCherry strain in zebrafish WT larvae. (**A**) Survival curves of control uninfected (No V) and SINV- or SINV339-mCherry infected zebrafish larvae. Data pooled from 5 independent experiments. N = 60 larvae per group. (**B**) Expression levels of E1 SINV gene, measured via qRT-PCR, for SINV and SINV339-mCherry (2 different concentrations). Note that at the highest concentration, all SINV339-mCherry infected larvae had succumbed at 2 dpi.

(C and D) Confocal image of WIHC processed SINV339-mCherry (C) and CHIKV-115 (D) infected fli1a:eGFP larvae, (C' and D') maximal projection. Scale bars: 50 µm. (C'-C''') SINV-infected cells in magenta; vasculature in green. (C'') merge of magenta (infected cells) and green (endothelium)

fluorescence from a single focal plane; (C''') magenta (infected cells) fluorescence; (C'''') green (endothelium) fluorescence. Yellow arrowheads: infected areas close to but not part of the vasculature. (D''') merge of magenta (infected cells) and green (endothelium) fluorescence from a single focal plane; (B'''') magenta (infected cells) fluorescence; (B'''') green (endothelium) fluorescence. Yellow arrowheads: areas of colocalization.

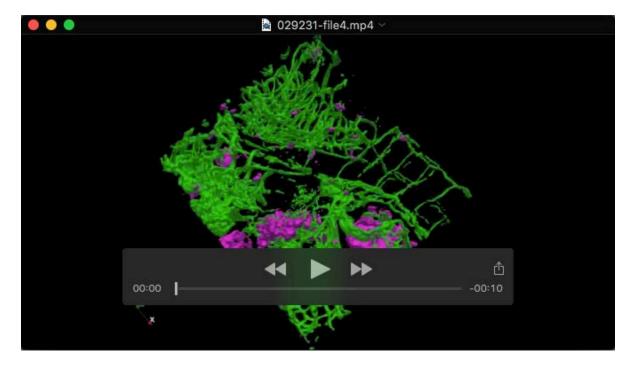
Supplementary movies



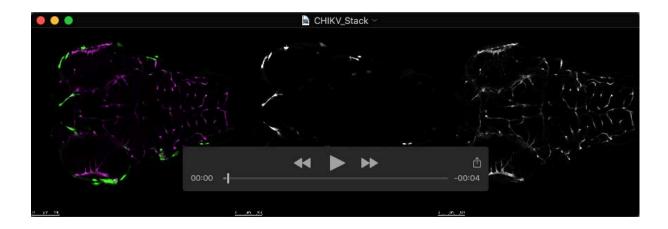
Movie 1. Movement through Z-stacks of *fli:G/U:RFP* SINV-infected larva, at 2 dpi. Related to Fig. 3B.



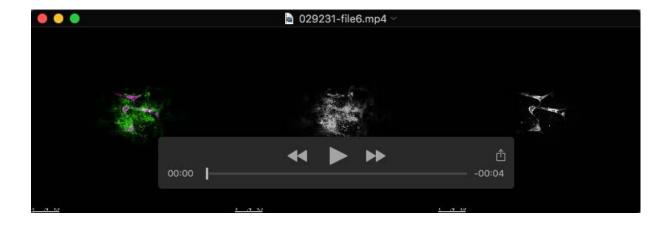
Movie 2. Movement through Z-stacks of the same *fli:G/U:RFP* SINV-infected larva, at 1 dpi. Related to Fig. **3A.**



Movie 3. 3D rendering of fli:G/U:RFP SINV-infected larva. Related to Fig. S1D.



Movie 4. Movement through Z-stacks of fli:G/U:RFP CHIKV-infected larva, at 1 dpi. Related to Fig. 3D.



Movie 5. Movement through Z-stacks of *fli: G/U:RFP* IHNV-infected larva, at 2 dpi. Related to Fig. 3F.