

Supplemental Information for:

# Covalently Immobilizing Interferon- $\gamma$ Drives Filopodia Production through Specific Receptor- Ligand Interactions Independently of Canonical Downstream Signaling

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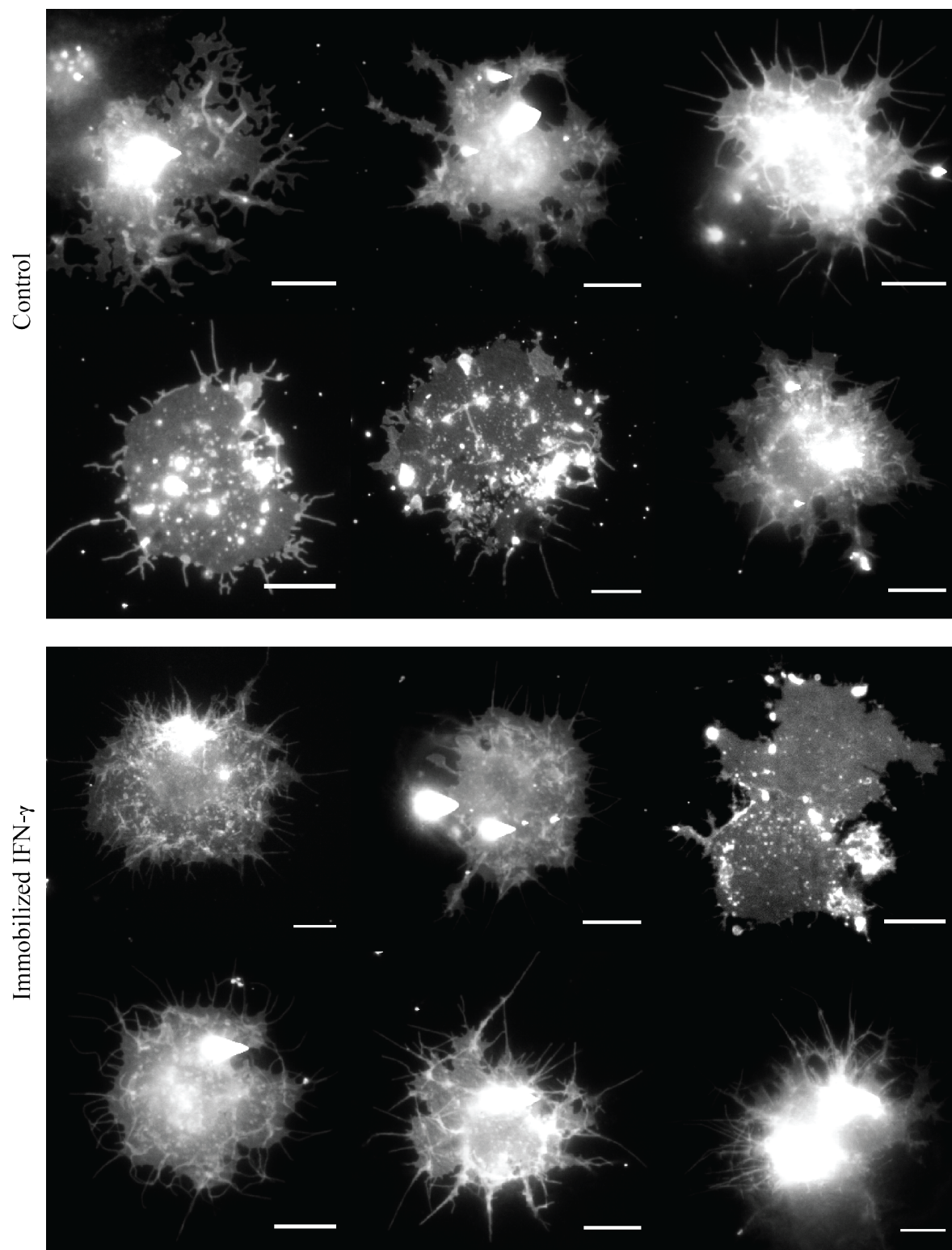


Figure S1: NSC morphology response to control and immobilized surfaces. The cell membrane was stained with DiO-C18 and imaged using a 488 nm TIRF laser. Scale bars represent 10 μm.

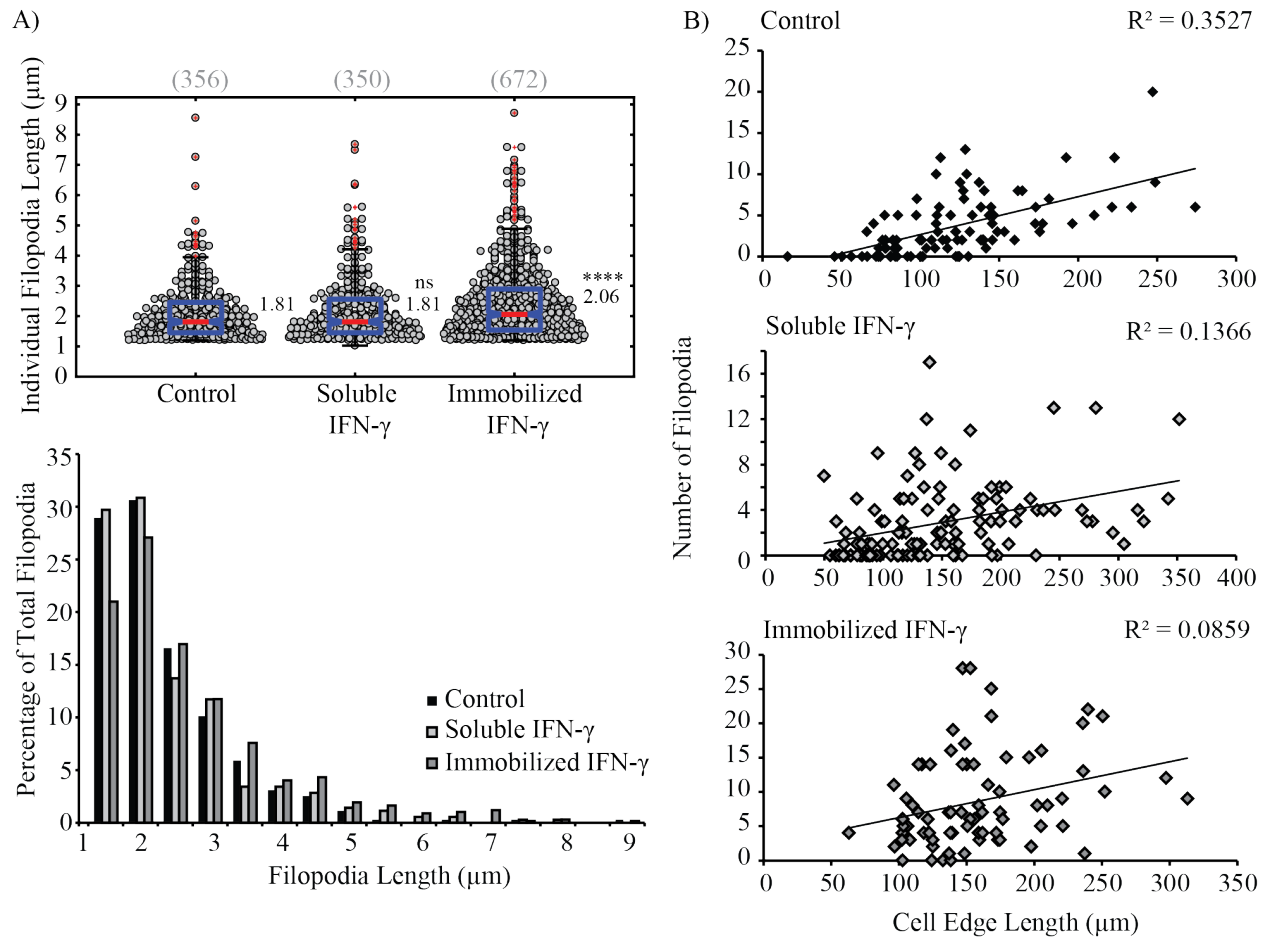


Figure S2: Additional Data Output from FiloQuant Analysis of Cos-7 A: Box plot and bee swarm plot (top) and normalized histogram (bottom) of filopodia length counts which show greater median length and a distribution shift toward longer filopodia in the immobilized IFN- $\gamma$  treatment group. The grey number above each plot shows the number of filopodia analyzed. \*\*\*\* represents  $p < 0.0001$  as determined by Student's t-test in pairwise comparisons with the control group. B: Scatter plot of number of filopodia compared to cell edge length in control (top) and experimental (center, bottom) groups. Filopodia number increases as cell size increases for the control group and less so for the soluble treated group, with no such trend seen for the immobilized IFN- $\gamma$  group.

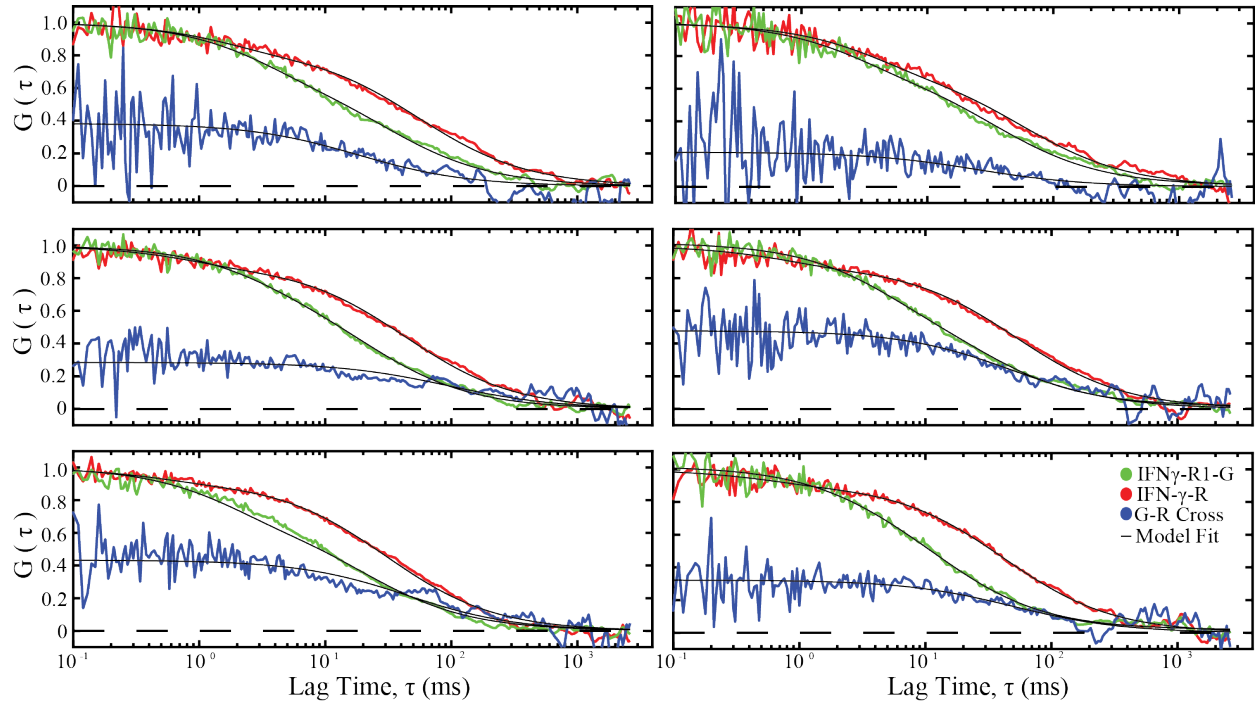


Figure S3: Typical auto- and cross-correlation data and model fits for IFN $\gamma$ R1-GFPSpark and soluble IFN $\gamma$  interaction in COS7 cells. Receptor auto-correlation is shown in green, ligand auto-correlation is shown in red, and cross-correlation is shown in blue.