

Rapid Neutralization Assays: Influenza Virus Particles for Flexible Readouts

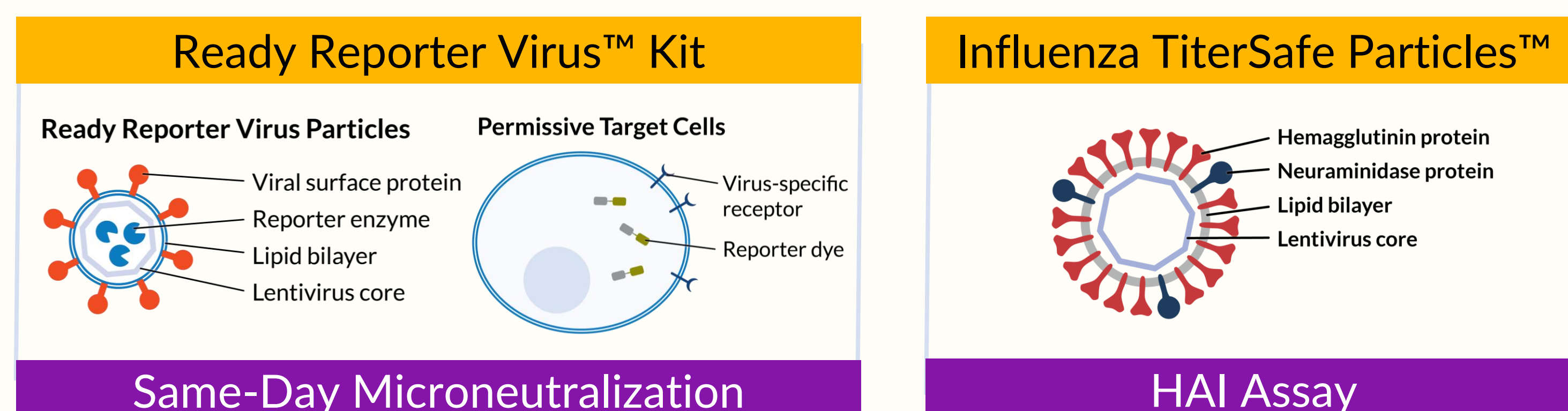
Christina K. Go, Chidananda Sulli, Nicholas Molino, Valerie Fiers, Parul Ganjoo, Lexy Rodrigues, Lewis J. Stafford, Jennifer Pfaff-Kilgore, Maya Cabot, Nathan Krump, Kyle Doolan, Benjamin Doranz

Integral Molecular, Philadelphia, PA

Non-Replicative Viral Particles with Authentic Antigen for Same Day, Simple Assays

- High-throughput functional neutralization data for antibody discovery and lead selection
- Consistent performance across lots supports QC lot release
- Multiple subtypes and strains enable rapid cross-reactivity studies from serum and mucosal sample cohorts

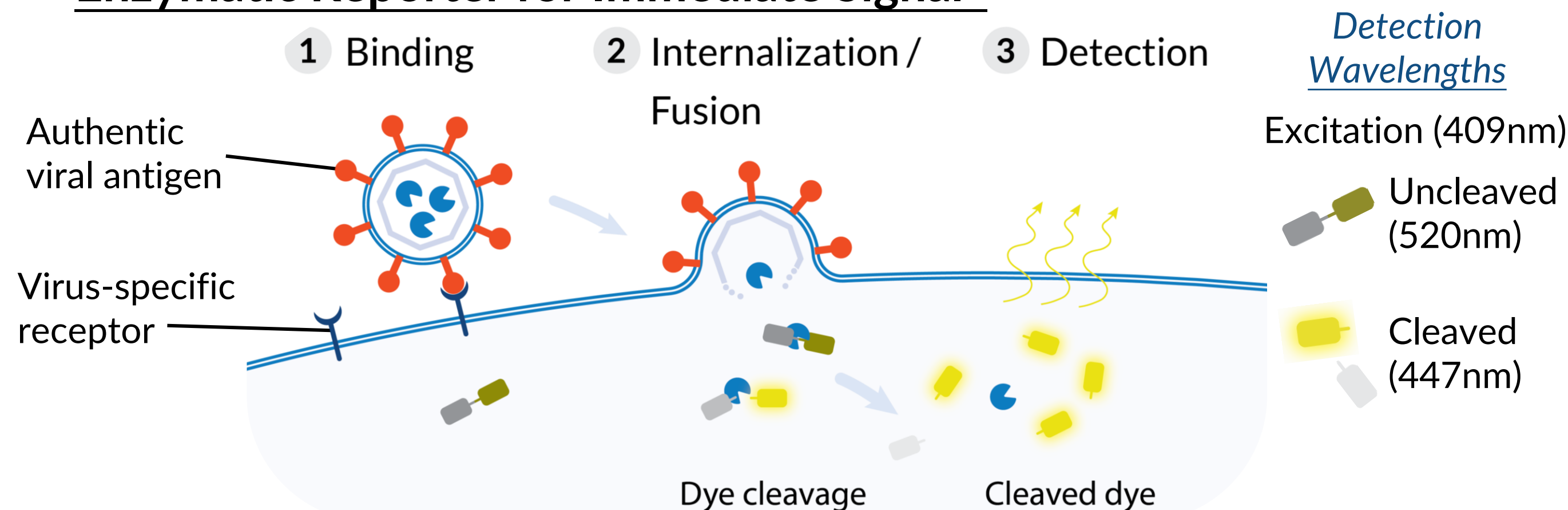
Goal: create faster, simpler neutralization assays with authentic non-replicative viral antigen



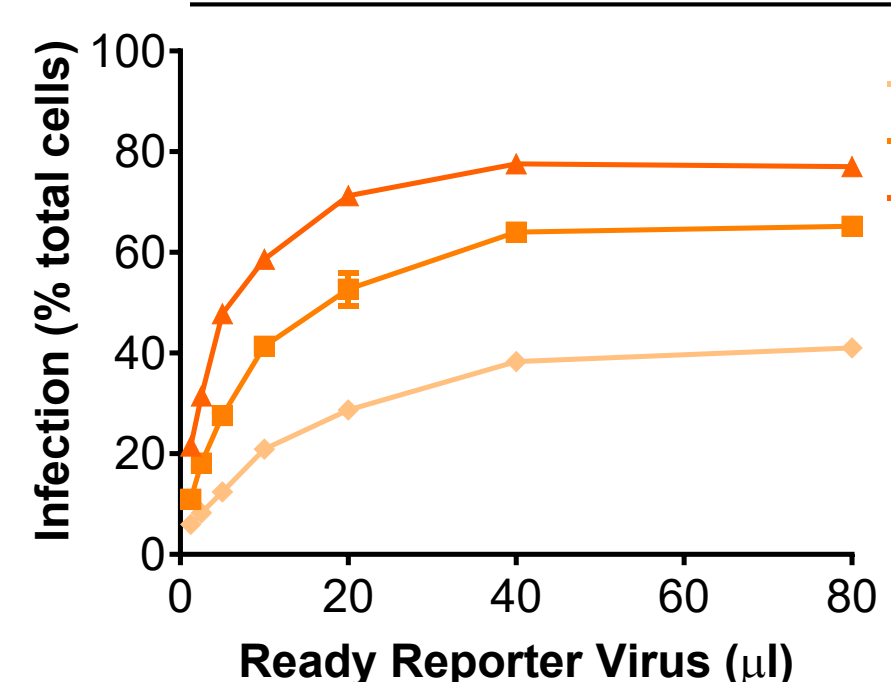
- Plasmid-defined production enables rapid response to emerging strains
- Thaw-and-use reagents minimize variability and simplify assay workflows and transfer
- Same-day neutralization assays that maintain consistency with live virus results
- Non-replicative particles enable BSL-2 study of HPAI with intact viral antigen

Ready Reporter Virus™ (RRV): Instantaneous Signal Upon Virus Fusion

Enzymatic Reporter for Immediate Signal



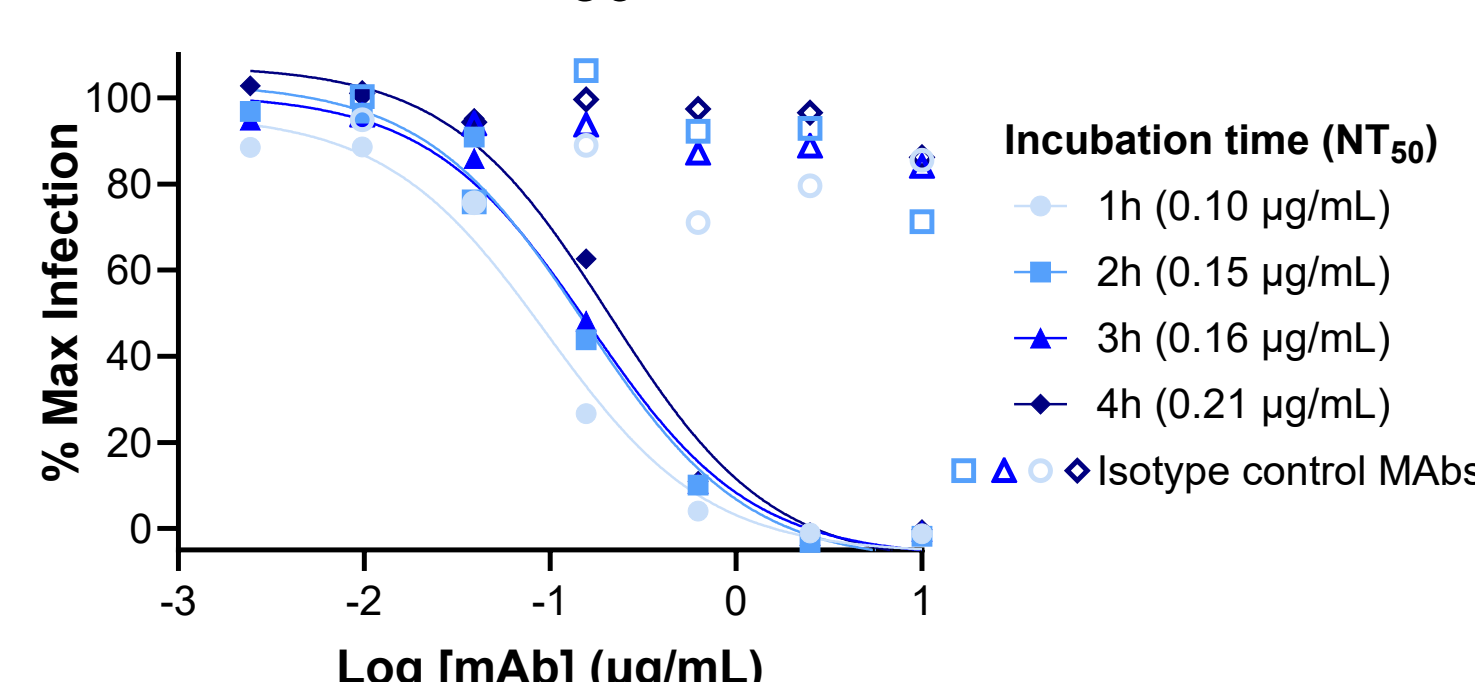
Results in <4 Hours



Infection (H5N1)

- Infection by influenza A/Texas/37/2024 RRV particles was measured at 1-, 2-, and 3-hours post-infection

Consistent NT₅₀ Across Infection Times

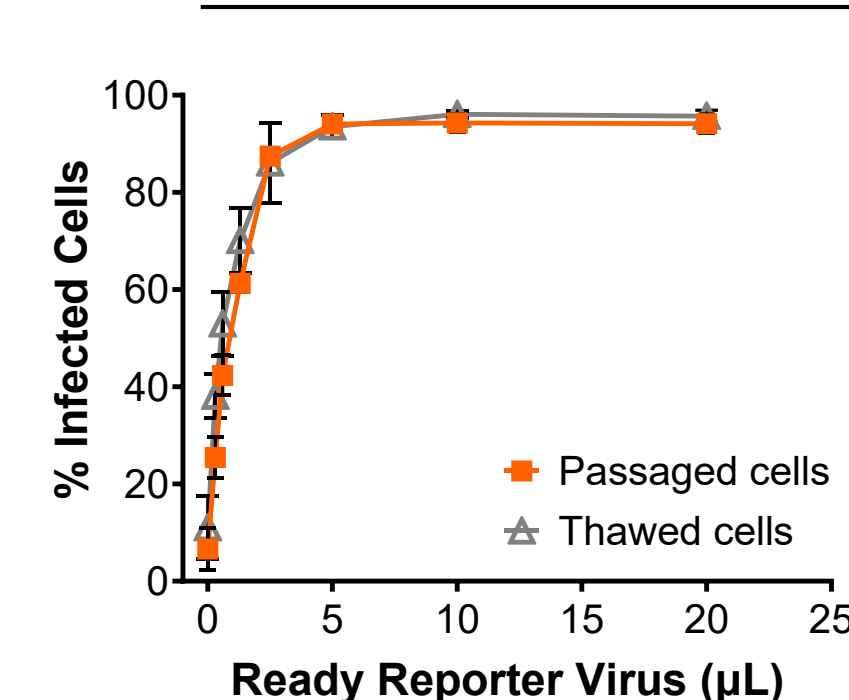


Neutralization (SARS-CoV-2)

- Readout of SARS-CoV-2 RRV neutralization at 1-4-hours post-infection of hsACE2-293T target cells

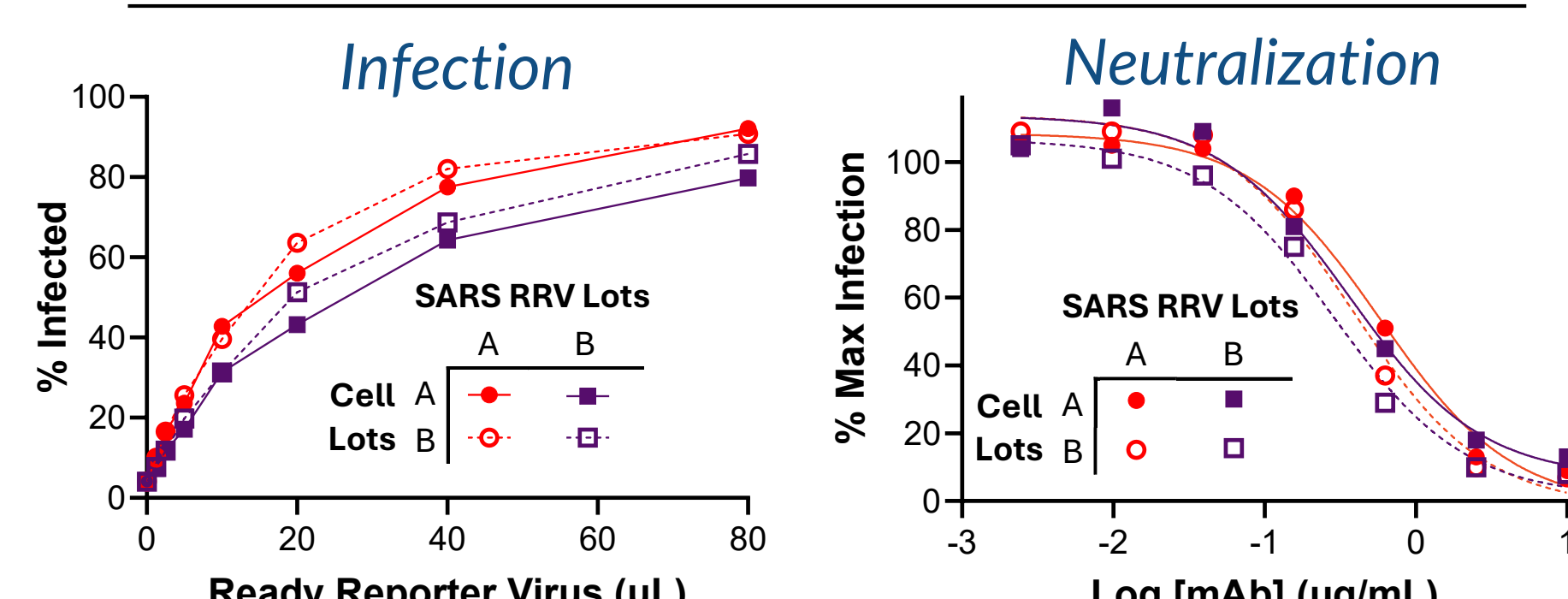
Thaw-and-Use RRV Kit Workflows for Assay Reproducibility and Transferability

Live Cell Infectivity



- Live target cells were thawed immediately before 3-hour infection with RRVs

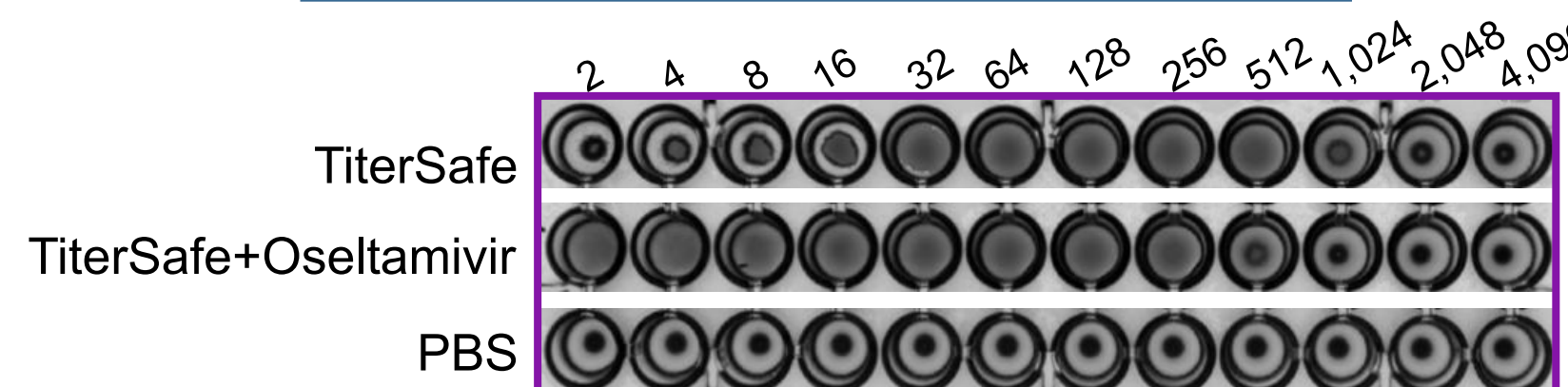
Uniform Results Across Manufactured Lots



- Infectivity and neutralization tested at 3 hours post-infection across two production lots of RRVs and target cells

Plasmid-Based Design Enables Assay Development for New Strains and Viruses within Weeks

TiterSafe HA Assay H5N1 Influenza A/Texas/37/2024

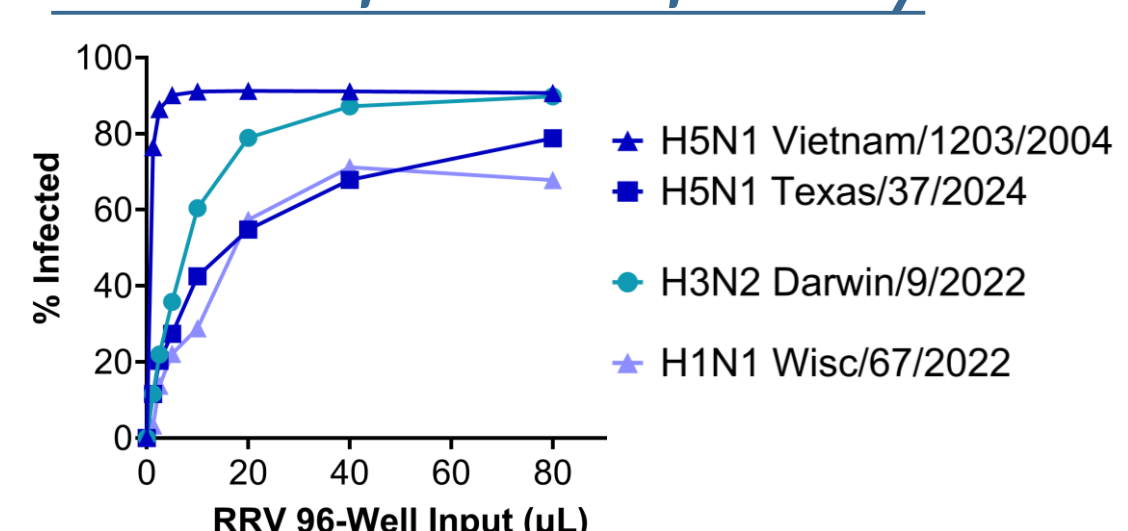


- HA assay performed with 1% guinea pig RBCs.
- Neuraminidase-mediated cleavage of hemagglutination is inhibited by oseltamivir

Influenza TiterSafe Particles

- Reagents and protocols optimized for:
 - Influenza A strains (H5N1, H3N2, H1N1)
 - Influenza B (Yamagata, Victoria lineages)

3-Hour Influenza Infectivity



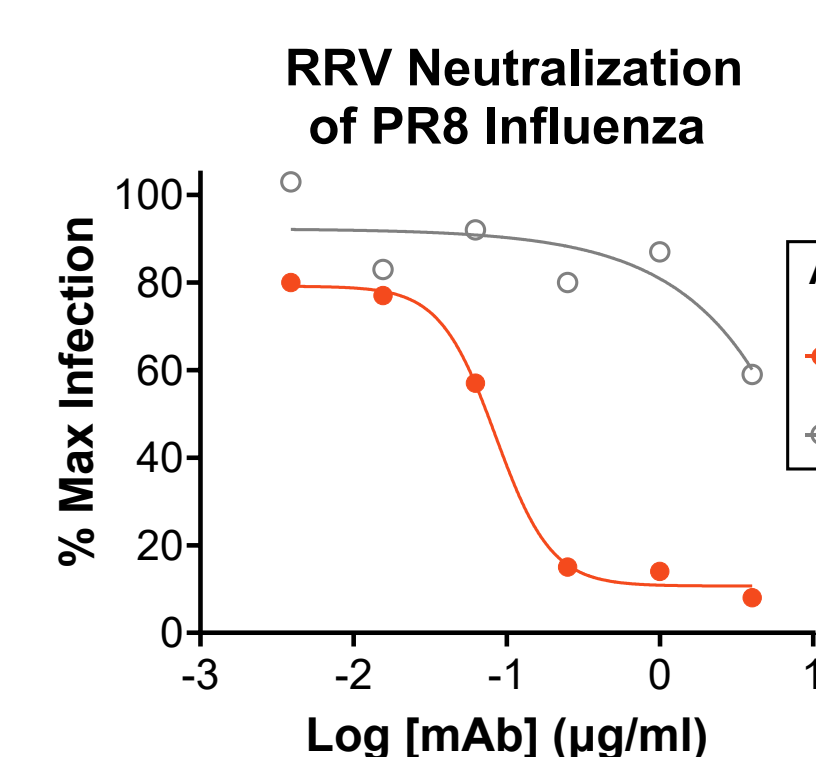
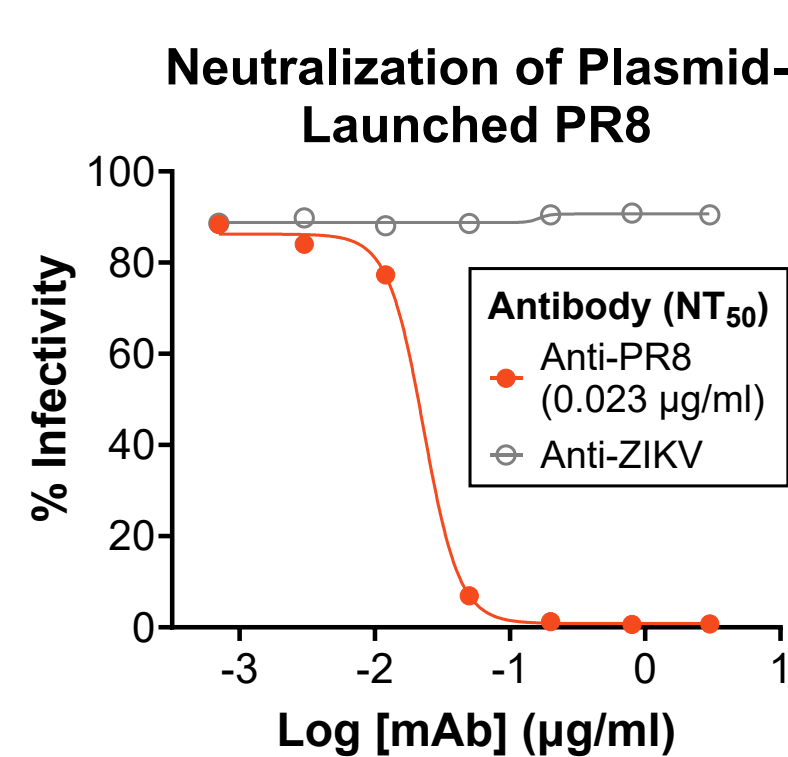
- Ready Reporter Virus particles were developed for multiple influenza A subtypes
- Infectivity at 3 hours post-infection

Ready Reporter Virus Kits

- Reagent kits and protocols optimized for:
 - Influenza A (H5N1, H3N2)
 - Lassa virus
 - SARS-CoV-2
 - VSV
 - Marburg virus

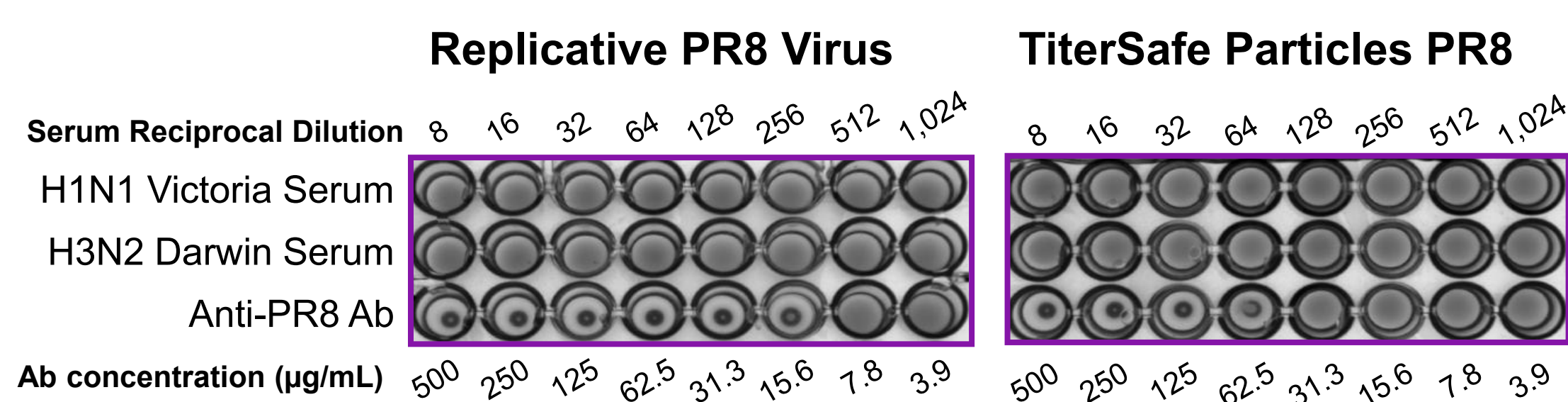
Virus Particles Show Consistent Results with Live Virus in Neutralization and HAI Assays

Ready Reporter Virus™ concordance with live virus neutralization titers



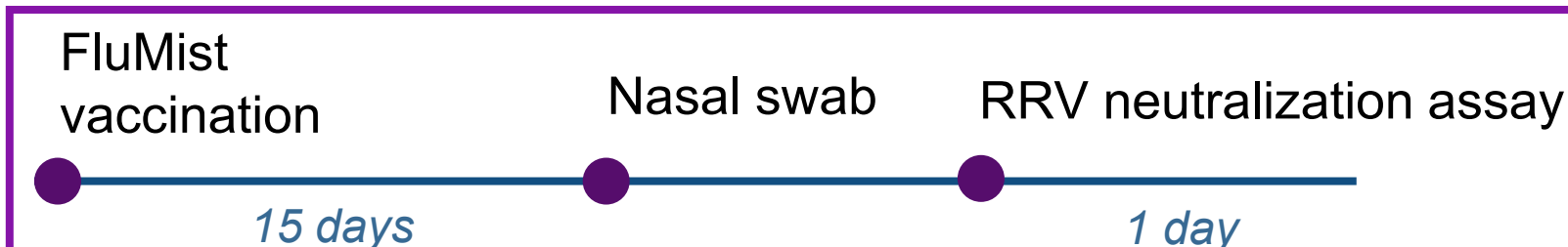
- Live H1N1 A/PR/8/34 virus and corresponding RRVs were neutralized by anti-PR8 Ab (NR-48783) with similar titers

Influenza TiterSafe Particles™ concordance with live virus HAI titers



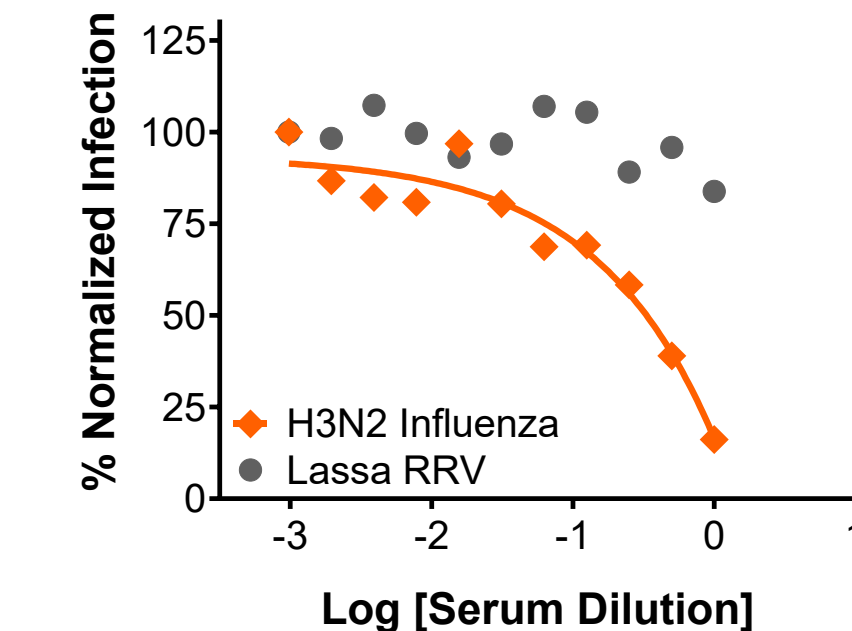
- Live H1N1 influenza A/PR/8/34 virus and TiterSafe equivalent both showed HA inhibition by anti-PR8 Ab, and not by sera recognizing H1N1 Victoria or H3N2 Darwin

Ready Reporter Virus Assays Detect Endogenous Neutralizing Responses in Mucosal Samples

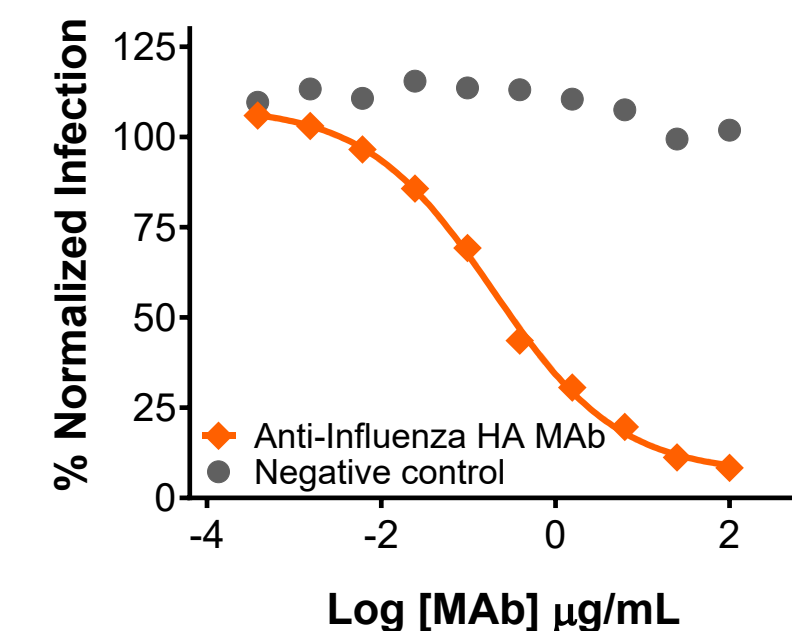


- H3N2 flu-specific neutralization measured in mucosal samples collected 15 days post-vaccination

Mucosal Response



H3N2 Influenza RRV



Lassa RRV

