# Oceanit Lateral Flow Assay (LFA) Protocol

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## ABSTRACT

This experiment uses infectious WA1 strain of SARS-CoV2 and is conducted in a qualified BSL3 facility at the University of Hawaii John A Burns School of Medicine (Honolulu, Hawaii).

## MATERIALS

- RNase A (10 mg/mL) Thermo Fisher Scientific Catalog #EN0531
- Triton X-100 VWR Scientific Catalog #9002-93-1
- BSA Sigma Aldrich Catalog #A2153
- Gold Nanospheres - Bare (Citrate) 80nm Nanocomposix Catalog # AUCN80
- TBSTween Sigma Aldrich Catalog #9039-10PAK
- Sample Pad Cytiva Catalog #8134-2250

## Protocol status: In development

We are still developing and optimizing this protocol.
Prepare Serial Dilution

This experiment uses infectious WA1 strain of SARS-CoV2 and is conducted in a qualified BSL3 facility at the University of Hawaii John A Burns School of Medicine (Honolulu, Hawaii).

Aliquot diluent solution into centrifuge tubes

Diluent solution is cell growth media supplemented with 10% FBS.

Prepare lysis buffer with LFA capture molecule

Lysis buffer containing LFA capture molecule is Oceanit’s proprietary formulation.

Virus stock (4 x 10e7 pfu/mL)

Virus stock 10-fold serial dilutions.

10-fold serial dilutions and control tubes

a. Tube 1 – Neat (4e7 pfu/mL); pfu = plaque forming units
b. Tube 2 – 10e-1
c. Tube 3 – 10e-2
d. Tube 4 – 10e-3
e. Tube 5 – 10e-4
f. Tube 6 – 10e-5
g. Tube 7 – 10e-6
h. Tube 8 – 10e-7
i. Control – no virus

Add lysis buffer to dilutions
Add prepared lysis buffer to labeled individual LFA cassettes. Test and control lines are Oceanit’s proprietary molecules.

7 Incubate 5 minutes

8 Add to labeled individual LFA cassettes.
Add 250uL of the corresponding sample to individual LFA cassettes.

9 Record results with photography:

A.Amount:
B.Concentration: Virus stock and dilution range from 4 x 10^7 pfu/mL to 0 virus added.
C.Temperature: Virus stocks are stored at -80°C; assay is performed at room temperature.
D.Duration of the experiment: 1 hour elapsed time.
E.Equipment: BSL3 containment, Class II biological safety cabinet, autoclave, pipette and disposable tips, disposable tubes, dedicated camera.
F.Reagents: Vero E6 cell culture growth media supplemented with 10% FBS, Oceanit lysis buffer + capture molecule (proprietary), LFA embedded test and control line molecules (proprietary)