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Universal Lateral Flow Assay kit

Applicable to:

4300-0100

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Introduction

Innova's universal Lateral Flow Assay (LFA) kit is designed to enable the easy development of customized sandwich lateral flow assays, by combining Lightning-Link® and InnovaCoat® GOLD conjugation technologies with an immunochromatography test performed on Universal-LFA strips.

The great advantage of this kit is its adaptability to any pair of capture and detection antibodies, which permits to detect of any type of analyte, without the need to stripe the capture antibody onto LFA strips using expensive equipment.

The capture antibody is conjugated to Lightning-Link® Ulfa-Tag, and the detection antibody is conjugated to 40nm InnovaCoat® GOLD, both of which require only 30 seconds to set up.

The capture and detection antibodies are diluted and incubated with the analyte and then run on Universal LFA strips. Universal LFA strips consist of a nitrocellulose membrane containing a 'Test line' (T-line) of immobilized anti-Ulfa-Tag antibody, that binds the Ulfa-Tag conjugated capture antibody which further binds the analyte in complex with the InnovaCoat® GOLD-detection antibody. A red T-line appears when the analyte is present and the line intensity varies depending on the analyte concentration (see Figure 1). Universal LFA strips also contain a 'Control-line' (C-line) that exploit the extraordinarily high affinity of streptavidin for biotin, which shows that the test is valid, and an absorbent pad to promote and control the flow of sample through the membrane.

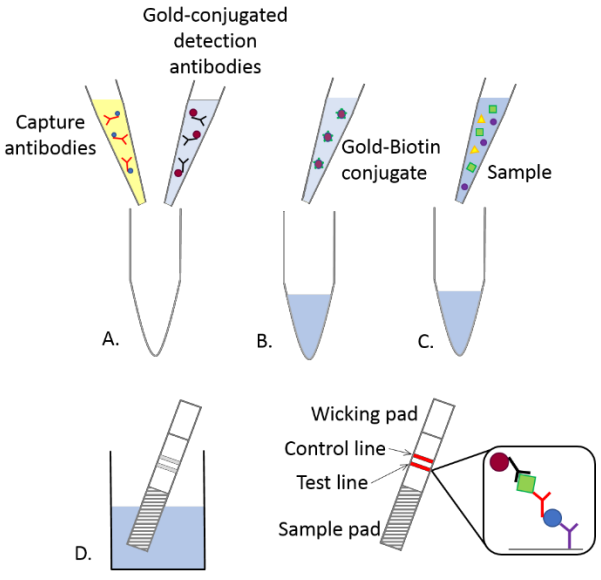

Figure 1.
This simple qualitative lateral flow assay does not require any specialized or costly equipment. The signal intensities can be qualitatively analyzed using the supplied scoring card or, for a quantitative detection, a LFA reader can be used.

Storage and shipping

The kit is shipped at ambient temperature. Upon receipt, store the pot containing the strips, the 10X Universal Running Buffer and InnovaCoat® GOLD-Biotin vials at +4°C.

The Lightning Link® Ulfa-Tag conjugation kit and 3 Reaction 40nm InnovaCoat® GOLD Mini kit should be stored at -20°C.

Kit contents

3 x 100µg vials Lightning-Link® Ulfa-Tag conjugation kit

3 x Reaction 40nm InnovaCoat® GOLD Mini kit

100 x Universal LFA strips

2 x cryovials of 10x Universal Running Buffer

1 x vial 40nm InnovaCoat®GOLD-Biotin, 10 OD

1 x scoring card

2 x 96-well clear low binding plates

Not supplied: Bovine Serum Albumin (BSA)

Instructions

1. Conjugate the capture and the detection antibodies using the Lightning-Link® Ulfa-Tag and 40nm InnovaCoat® GOLD kit respectively, following the protocols inside each kit
2. Dilute the 10x Universal Running Buffer 1:10 with distilled water and add a blocking agent (0.1% BSA final concentration) to obtain 1x Universal Running Buffer + BSA
3. Dilute the Ulfa-Tag conjugated capture antibody to 40-150µg/ml in 1x Universal Running Buffer + BSA
4. Dilute the InnovaCoat®GOLD-detection antibody to 6 OD in 1x Universal Running Buffer + BSA
5. Dilute the 40nm InnovaCoat®GOLD-Biotin 10 OD to 1 OD in 1x Universal Running Buffer + BSA (1:10 dilution)
6. Dilute your analyte in 1x Universal Running Buffer + BSA

7. We recommend testing each sample in duplicate or triplicate. For a single strip, prepare the following mix:

   5µl diluted capture Ulfa-Tag conjugated antibody

   5µl 6 OD 40nm InnovaCoat®GOLD-detection antibody

   5ul 1 OD 40nm InnovaCoat®GOLD-Biotin

   75µL analyte solution

8. Incubate for 5 minutes
9. In a 96 well plate, load 80µl of the mix and insert one Universal LFA strip in each well. Handle the strip from the wicking pad (thicker pad, made of cellulose), avoid touching the nitrocellulose, and make sure the sample pad (thinner and longer pad, made of glass fiber) is dipped into the well.
10. Run the mix for 20 minutes
11. Compare the T-line color intensity with the score reported on the scoring card. If the C-line is not visible the test is not valid and should be repeated.

Lateral Flow Assay optimization

We recommend testing different concentrations of Ulfa-Tag conjugated capture antibody between 40 and 150µg/ml, loading a fixed concentration of gold conjugate for initial testing. Too much capture antibody will result in a low signal, as free antibody may compete with immunocomplexes for binding to the T-line.
Increasing the OD of the gold-conjugates will increase the intensity of the T and C-lines but also the background.
If your sample contains biotin, it may cause a reduction in C line intensity: if it is not clear whether the test is valid, either dilute your sample in Running Buffer or increase the amount of 40nm InnovaCoat®GOLD-Biotin conjugate.
When running a LFA for the first time, we recommend trying large dilutions of sample/antigen to determine the dynamic range of the assay. The sensitivity of the test will vary based on the concentration and nature of the antigen to be detected.
We advise to keep the volume loaded on the strips at 80µl.
The 2 x 96-wells plates provided allows you to have wells in surplus to plan with ease your experiments.

Related products

Antibody labeling kits:
https://www.innovabiosciences.com/products/lightning-link-antibody-labeling-kits
Gold nanoparticles:
https://www.innovabiosciences.com/products/particles-particle-conjugation-kits
Lateral Flow Immunoassays guide
https://www.innovabiosciences.com/resources/applications/lateral-flow-immunoassay/
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Technical support

For further information or for any technical enquiries get in touch via our website at:
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