

PRODUCT DATA SHEET

Conjugation QC Lateral Flow Dipstick Kit

Catalog Number LF-018-10

Kit Components

- 10 Conjugation QC Lateral Flow Dipsticks
- 1.5 mL Lateral Flow Running Buffer

Procedure for Testing an Antibody Conjugate After Conjugation

General Description and Test principle

Cytodiagnosics' Conjugation QC Lateral Flow Dipstick Kit Test is a rapid immunochromatographic-based test intended for quality control of antibody conjugation to gold nanoparticles.

The dipstick strips supplied in the kit have four anti-species antibodies (anti-Goat IgG, anti-Human IgG, anti-Rabbit IgG and anti-Mouse IgG) immobilized at discrete locations (test lines) on the membrane of the strip. When an antibody-conjugated gold nanoparticle migrates up the test strip, it will bind to the location with immobilized antibodies specific for the antibody species conjugated to the gold nanoparticle. As a result, a red line will appear indicating a successful conjugation reaction, figure 1. A conjugate from a failed conjugation reaction will not generate any red lines indicating that troubleshooting is required.

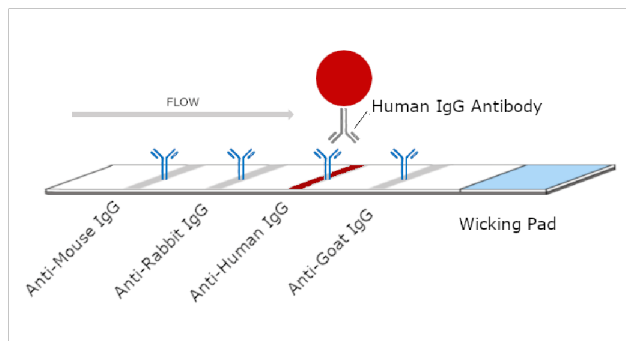


Figure 1. Schematic of the Conjugation QC Lateral Flow Dipstick and its use for evaluating conjugation of an antibody to gold nanoparticles.

Limitations

The test strips in this kit are only able to detect gold nanoparticles conjugated with antibodies from the following species and class:

- Goat IgG
- Human IgG
- Rabbit IgG
- Mouse IgG

Required Components Not Included with Kit

- 96-well plate

Storage and Stability

The dipsticks supplied in the Conjugation QC Lateral Flow Dipstick Kit should be stored between 2-30°C and the supplied Lateral Flow Running Buffer should be stored at 2-8°C. If stored properly, this kit is stable for at least 3 months.

Protocol

1. Dilute a small aliquot of your gold conjugate to be tested with 1XPBS (pH 7.4) to reach a final optical density of 10 (Absorbance=10, 1 cm pathlength) at the main absorption peak.
2. Transfer 75 μ L of Lateral Flow Running Buffer into a well of a 96-well plate.
3. Transfer 5 μ L of diluted gold conjugate into the well with Lateral Flow Running Buffer and mix well by pipetting up and down a few times.
4. Place a lateral flow dipstick into the well with conjugate and running buffer. The strip should be placed into the solution with the logo of the strip positioned up, see figure 1.
5. Incubate for 10 minutes.
6. Remove lateral flow dipstick from the well. Some solution will remain in the well and is normal.
7. Immediately record the result.

The appearance of at least one clearly visible red line in the location of the test line specific for the species of antibody conjugated to the gold nanoparticles indicates a successful conjugation, see figure 1. A failed conjugation reaction will not result in the formation red lines on the strip.

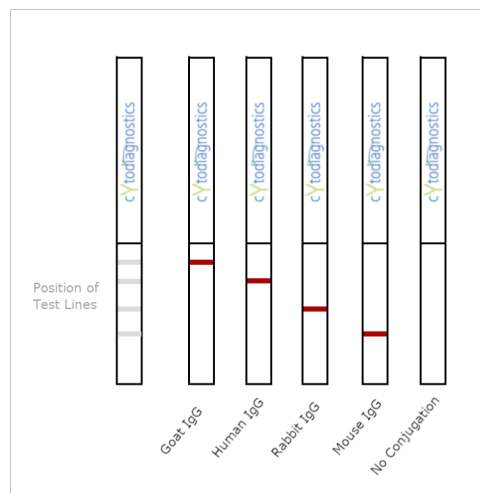


Figure 2. Positive results for NHS-activated gold nanoparticles conjugated to a goat, human, rabbit and mouse IgG antibody, respectively. The far-right strip shows a negative result indicating a failed conjugation reaction. Note that depending on the characteristics and cross-reactivity of the antibody conjugated to the gold nanoparticles, lines in addition to the specific test line may appear and is normal.

Procedure for Testing an Antigen Conjugate After Conjugation

General Description and Test principle

Cytodiagnostics' Conjugation QC Lateral Flow Dipstick Kit Test is a rapid immunochromatographic based test intended for quality control of antigen conjugation to gold nanoparticles.

The dipstick strips supplied in the kit have four anti-species antibodies (anti-Goat IgG, anti-Human IgG, anti-Rabbit IgG and anti-Mouse IgG) immobilized at discrete locations (test lines) on the membrane of the strip. The first step is to incubate the antigen gold conjugate with an antibody specific to the antigen. When the antibody bound antigen-conjugated gold nanoparticle is subsequently allowed to migrate up the test strip, it will bind to the immobilized antibody specific for the species of antibody used. As a result, a red line will appear indicating a successful conjugation reaction, figure 3. A conjugate from a failed conjugation reaction will not generate any red lines indicating that troubleshooting of the conjugation reaction is required.

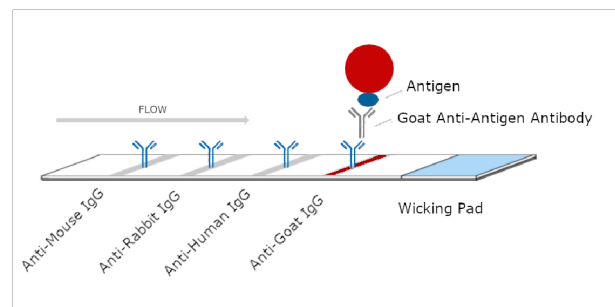


Figure 3. Schematic of the Conjugation QC Lateral Flow Dipstick and its use for evaluating conjugation of an antigen to gold nanoparticles in conjunction with an antibody specific to the antigen.

Limitations

The test strips in this kit are only able to detect gold nanoparticles conjugated to an antigen in conjunction with a detection antibody from the following species and class:

- Goat IgG
- Human IgG
- Rabbit IgG
- Mouse IgG

Required Components Not Included with Kit

- Antibody specific to the conjugated antigen from a source and class in the list above
- 96-well plate

Storage and Stability

The dipsticks supplied in the Conjugation QC Lateral Flow Dipstick Kit should be stored between 2-30°C and the supplied Lateral Flow Running Buffer should be stored at 2-8°C. If stored properly, this kit is stable for at least 3 months.

Protocol

1. Dilute a small aliquot of your gold conjugate to be tested with 1XPBS (pH 7.4) to reach a final optical density of 10 (Absorbance=10 at the main absorption peak, 1 cm pathlength).
2. Transfer 75 μ l of Lateral Flow Running Buffer into a well of a 96-well plate.
3. Dilute an antibody specific to the antigen conjugated to the gold nanoparticles to a final concentration in the range of 3 μ g/ml – 15 μ g/ml with 1XPBS (pH 7.4).
4. Transfer 10 μ l of the diluted antibody solution to the well with Lateral Flow Running Buffer.

5. Transfer 5 μ l of diluted gold conjugate into the well with Lateral Flow Running Buffer and antibody and mix well by pipetting up and down a few times.
6. Incubate for 2 minutes.
7. Place a lateral flow dipstick into the well with conjugate and running buffer. The strip should be placed into the solution with the logo of the strip positioned up, see figure X.
8. Incubate for 10 minutes.
9. Remove lateral flow dipstick from the well. Some solution will remain in the well and is normal.
10. Immediately record the result.

The appearance of a clearly visible red line in the location of the test line specific for the species of the antibody used for binding to the antigen (step 3 above) indicates a successful conjugation, see figure 4. A failed conjugation reaction will not result in the formation of red lines on the strip.

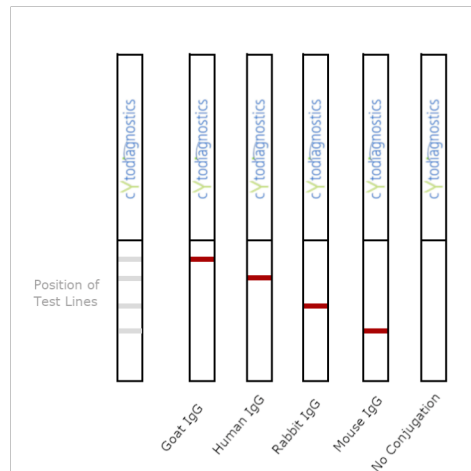


Figure 4. Positive results for NHS-activated gold nanoparticles conjugated to an antigen and incubated with a goat, human, rabbit and mouse IgG detection antibody specific for the antigen, respectively. The far-right strip shows a negative result indicating a failed conjugation reaction. Note that depending on the characteristics and cross-reactivity of the antibody used for detection, lines in addition to the specific test line may appear and is normal.