Lentivirus Titer Rapid Assay Kit

Cat No.	Size
RA10089	10T
RA10089	100T
Store at room temperature, shelf life: 1 year	

1、Introduction

This product is a semi-quantitative detection card for HIV-1 P24 protein based on colloidal gold lateral flow chromatography technology, which can be used for rapid evaluation of the packaging efficiency of related lentiviral vectors and pseudoviruses (P24 protein is the most abundant marker protein in the lentiviral envelope).

The detection card consists of a chromatography membrane, sample pad, gold conjugate pad, absorbent pad, and backing plate. Goat anti-mouse polyclonal antibody (Control Line) and P24 mouse monoclonal antibody (Test Line) are immobilized on the chromatography membrane; another strain of colloidal gold-labeled P24 mouse monoclonal antibody is immobilized on the gold conjugate pad.

When the test sample is added to the sample pad, it migrates toward the gold conjugate pad via capillary action. Upon reaching the gold conjugate pad, P24 protein in the sample binds to the mouse anti-P24 monoclonal antibody on the gold conjugate pad to form a complex. The complex moves to the Test Line and is captured by another strain of P24 mouse monoclonal antibody on the Test Line. As the captured complex accumulates, a wine-red color appears at the Test Line. When the complex migrates to the Control Line, the goat anti-mouse polyclonal antibody captures the colloidal gold-labeled mouse monoclonal antibody, showing a wine-red color.

The presence of P24 antigen in the sample is determined by reading whether the Test Line and Control Line on the chromatography membrane develop color. When the concentration of P24 in the sample changes, the color depth of the Test Line is positively correlated with the P24 concentration. By comparing with the color card (example in Figure 1), the concentration of P24 protein in the test specimen can be semi-quantified, and the lentivirus titer can be roughly estimated.

Number	T1	T2	T3	T4	T5
P24	500ng/mL	100ng/mL	50ng/mL	10ng/mL	1ng/mL
LPs/ml*	6.25×10^{9}	1.25×10^{9}	6.25×10^{8}	1.25×10^{8}	1.25×10^{7}
TU/ml	$6.25\times10^{6\text{-}7}$	$1.25\times10^{6\text{-}7}$	$6.25\times10^{5\text{-}6}$	$1.25\times10^{5\text{-}6}$	$1.25\times10^{4\text{-}5}$

Note: Since this product is a semi-quantitative detection card, the color card data in Figure 1 is for reference only.

A lentiviral particle (LP) contains approximately 2000 P24 protein molecules. The number of lentiviral vector particles (LP) can be calculated using the following formula:

One LP is equivalent to: $2000\times24\times10^{3}/(6\times10^{23})$g of P24 = 8×10^{-5} pg of P24; or, 1ng P24 = 1.25×10^{7} LPs ($\approx1.25\times10^{4-5}$ TU*)

**Under normal conditions, 1 out of every 100-1000 LPs will be an infectious viral vector, i.e., 1 Transducing Unit (TU).

2、Intended Use

Detection of packaging efficiency of HIV-1-based lentiviral vectors

Regardless of the lentiviral packaging system used, packaging efficiency is the key to the success of the experiment. Traditional methods for detecting lentiviral packaging efficiency include ELISA (detecting P24 protein content) and Real-time PCR (detecting nucleic acid copy number), both of which are time-consuming, labor-intensive, and costly. Compared with these two methods, this product is easy to use and has a short detection time, allowing for real-time evaluation of transfection effects. For example, when changing the medium, aspirate a sample and add it to the detection card, and the results can be read after 10-15 minutes. Therefore, using this quick detection card can quickly evaluate transfection effects and reduce unnecessary waste.

Detection of packaging efficiency of HIV-1-based pseudoviruses

HIV-1-based pseudoviruses also contain P24 protein. After transfection, this detection card can be used to quickly evaluate the packaging effect.

3、Components

1） Detection Card;　2) Running Buffer;　3) Instructions for Use

4、Usage Method

1. Open the package, take out the detection card, and place it flat on the biosafety cabinet tabletop.
2. Pipette 20μL of cell culture supernatant with a micropipette and add it to the sample window (S); then add 50μL of buffer.
3. Read the results after standing for 10-15 minutes.

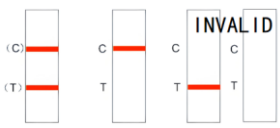
5、Result Interpretation

1. Positive: Both the Control Line (C) and Test Line (T) develop color in the detection window.

2. Negative: The Control Line (C) develops color, and the Test Line (T) does not develop color.

3. P24 content determination: Please judge the P24 content according to the color card. (If the T color is too dark, dilute the sample and retest)

4. Invalid: If the Control Line (C) does not develop color, the test is invalid regardless of whether the Test Line (T) develops color (Figure 2).



6、Troubleshooting

Phenomenon	Possible Cause	Solution
Faint color of both Test Line and Control Line	Interfering substances in the sample	Dilute with DMEM and retest
Test failed	Detection card expired	Check the shelf life and avoid moisture after opening the bag
High background	Medium pH, excessive salt concentration, or other interference	Dilute several times with PBS and retest