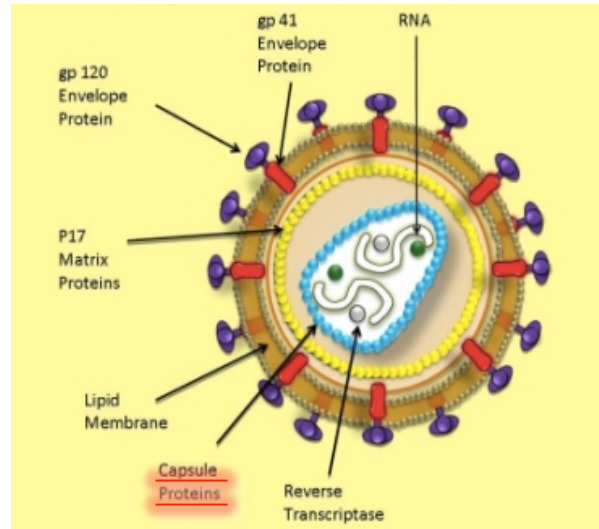


Lentivirus Titration XpressCard (ATG-LT-01)

Introduction

Antagen's Lentivirus Titration XpressCard is a rapid lateral flow immunoassay that allows quick confirmation of useful lentivirus titers* in the packaging supernatants. The test detects lentiviral capsule proteins released into the packaging supernatant as a surrogate marker to determine whether virus production yields a usable titer for subsequent infection. This assay takes only 10-15 minutes, compared to the conventional re-infection based assay that takes 2-3 days.



Protocol

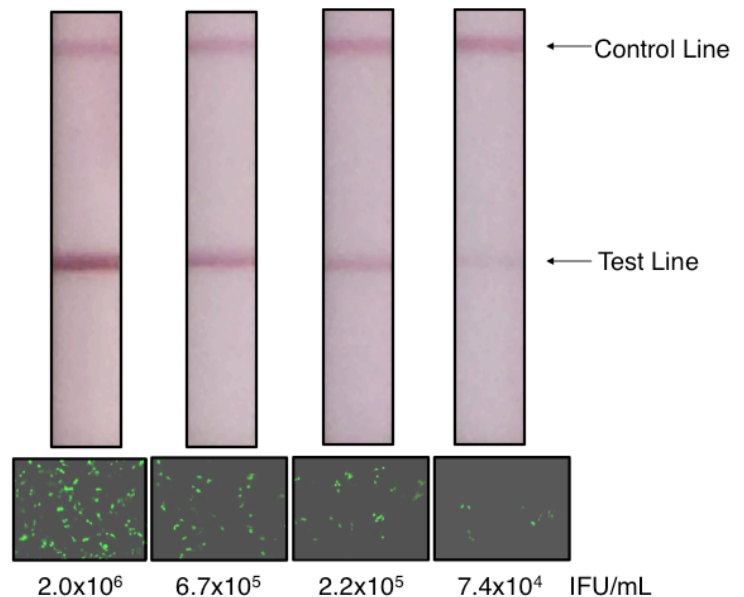
- 1) Place the card horizontally;
- 2) Add 100 μ L of packaging supernatant directly to the Sample Well;
- 3) Wait for 10-15 minutes.

A red control line will always appear in 5 minutes. Depending on the titers (a usable titer should be at least 1×10^6 IFU/mL), a dark red test line will appear within 10-15 minutes.

Storage

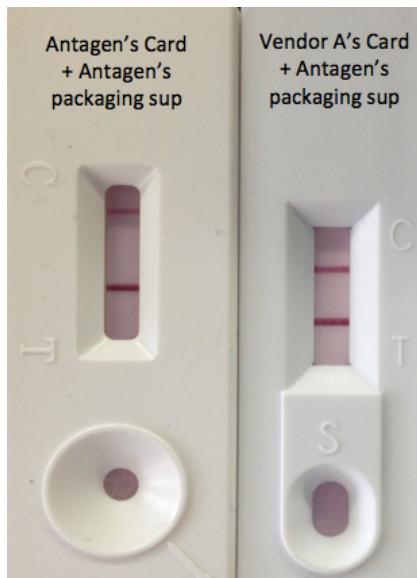
Store the cards in their original packaging at room temperature. This product is stable for up to 18 months from the certified date. Freezing the strips is not recommended.

* Users are encouraged to correlate T line intensity with functional titers in their own infection system, as different cell types have different susceptibility to lentivirus. In the above example, HEK293 cells were infected with serially diluted lentivirus packaging supernatants. After 72 hrs, ZsGreen1-expressing cells were observed under fluorescence microscope. A dark red test line was generated by a dilution containing $\sim 2 \times 10^6$ IFU/ml (as measured by flow cytometry of transduced HEK293 cells).



FAQ

1. How is Antagen's XpressCard compared with similar product on the market?



When loaded with the same packaging supernatant, Antagen's XpressCard showed the T line quickly in about 30 sec, much faster than Vendor A's, which showed the T line around 2 min. When observed at 10 min, Antagen's XpressCard showed the T line at somewhat stronger or at least equivalent intensity as Vendor A's. Therefore, Antagen's XpressCard is more sensitive than similar product on the market.

2. What is the sensitivity of the assay?

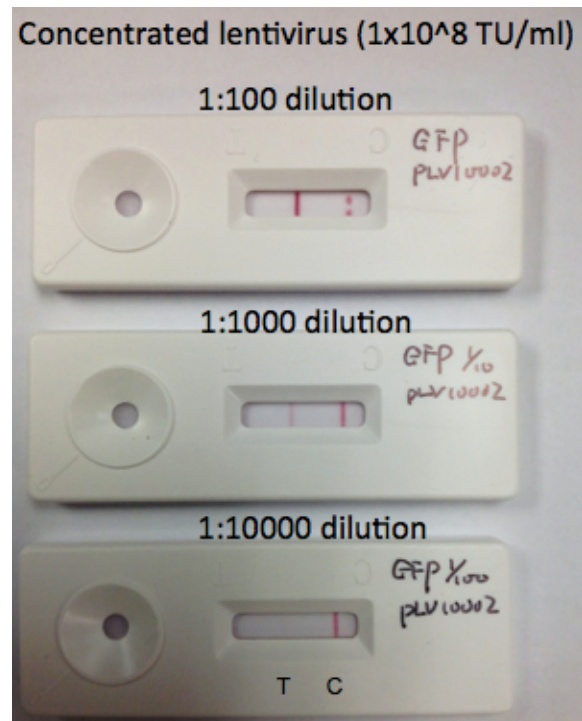
We tested XpressCards using serial dilution of purified concentrated lentivirus stock with known titres (Cellomics Technology, MD USA). The results showed that the detection limit is at 1×10^5 TU/ml or 1×10^4 TU (in 100 μ L). According to the literature, one lentivirus particle carries about 2,000 p24 molecules, which is the analyte of the XpressCard:

$$1 \times 10^5/\text{ml} \times 2000 \div (6.23 \times 10^{23}) \times 24 \times 10^3 \text{ g} \\ = 2 \times 10^8 \div (6.23 \times 10^{23}) \times 24 \times 10^3 \times 10^{12} \text{ pg/ml} \\ = 7.7 \text{ pg/ml}$$

As a comparison, Vendor A's "Lenti-X™ p24 Rapid Titer Kit" ELISA kit has a sensitivity of 12.5 pg/ml, whereas Vendor B's "QuickTiter Lentivirus Titer Kit" has a sensitivity of 1 ng/ml. These assays are all ELISA based, which need 3-4 hours. Therefore, Antagen's XpressCard can reach ELISA sensitivity but with only 10-15 min on hand time.

2. Can purified lentivirus particles be used in this assay?

As shown in the above case, both packaging supernatant and purified lentivirus particles can be used, as the sample pad contains detergent to efficiently lyse viral particles and release p24 protein during flow migration.



LentiTitre™ Cassette (ATG-LT-02):

To assist more accurate titration in real-time, we have launched a Lentivirus Titration Kit with a 4x multi-lane cassette and purified lentivirus standards with known titres, which can allow running the sample and standards side-by-side at the same time. Please visit our website for details.