



qPCR Lentivirus Titer Kit

Cat. No. LV900

Store at -20°C.

Product Description

abm's qPCR Lentivirus Titer Kit is a one-step assay that employs a rapid lysis step, followed by RT-qPCR. Designed to deliver high sensitivity and specificity, the kit ensures minimal non-specific background and provides better overall performance compared to similar kits on the market.

Product Component	Quantity	Part No.
BlasTaq™ 2X qPCR Titer MasterMix	1.25 ml	P889-1
Primer Mix	100 rxn (200 µl)	LV900-A
Standard Control DNA	50 µl	LV900-B
Virus Lysis Buffer	2 x 800 µl	LV900-C
ROX Reference Dye	15 µl	P101
Nuclease-Free H ₂ O	2 x 1.0 ml	P100

Protocol

The MasterMix contains a dye comparable to SYBR Green™ and EvaGreen™. ROX Reference Dye is provided separately from the MasterMix, making this kit universally compatible with most qPCR instruments. See **ROX Machine Compatibility** on our product page under the **Documents** tab on our website.

The recommended amount of ROX Reference Dye to be added to the MasterMix may vary depending on the qPCR machine type:

- No ROX equipment: Not needed.
- Low ROX equipment: 1 µl per 1.25 ml MasterMix.
- High ROX equipment: 11 µl per 1.25 ml MasterMix.

1. **Sample Preparation:** For purified high-titer viral samples, dilute the virus to the 10⁶IU/ml range (for best results) using 1X PBS or DMEM.
2. **Viral Lysis:** Add 2 µl of the sample preparation (from Step 1, or 2 µl of viral supernatant for low-titer preparations) to 18 µl of Virus Lysis Buffer. Incubate at room temperature for 5 minutes, followed by a brief centrifugation (snap spin). Use the lysed sample for the reaction setup in Step 4. **Note:** The viral sample is further diluted by an additional factor of 10. This total dilution factor must be taken into account when calculating the final titer, especially for purified virus sample titrations.

3. **Standard Control DNA Dilutions:** Perform four (4) 10-fold serial dilutions of the Standard Control DNA by adding 5 µl of DNA to 45 µl of Nuclease-free H₂O. The resulting 1/10 to 1/10,000 dilutions will be used to generate the standard curve.
4. **Set-up:** All reactions are recommended to be set-up on ice in **triplicates**.

Component	Volume
2X qPCR MM	10 µl
Primer Mix	2 µl
Sample, NTC, or Standard DNAs	2 µl
Nuclease-Free H ₂ O	6 µl

5. qPCR cycling conditions:

Step	Temperature	Duration	Cycle(s)
Reverse Transcription	42°C	20 min	1
Enzyme Activation	95°C	10 min	1
Denaturation	95°C	15 sec	34
Annealing/Extension	62°C	1 min	

Data Analysis

Plot Ct value (Y-axis, linear scale) vs. Virus titer (X-axis, logarithmic scale). Generate a logarithmic regression using the four (4) Standard Control DNA dilutions to determine the unknown virus sample titer using $y = m \ln(x) + b$ from the trendline equation. The R² value should be > 0.95 to justify the proper assay setup. Note to include the dilution factor of 10 plus additional diluting factor for purified viral samples in the final calculation.

$$\text{Virus titer (IU/ml)} = e^{(Ct - b)/m}, \quad \text{where } m \text{ is the slope of the line and } b \text{ is the y-intercept.}$$

Example: trendline equation is $y = -1.408 \ln(x) + 37.42$; Ct of unknown sample = 17.16
 $\text{Virus titer (IU/ml)} = e^{(17.16 - 37.42)/-1.408} = 1.77 \times 10^6 \text{ IU/ml}$

Dilution	Virus Titer (IU/ml)
1/10	1 x 10 ⁶
1/100	1 x 10 ⁵
1/1,000	1 x 10 ⁴
1/10,000	1 x 10 ³

Download the **qPCR Lentivirus Titer Calculation Form** from the product page under the **Datasheet** Tab on our website.