Datasheet for ABIN2345178

HIV ELISA Kit

25 Publications

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Overview

Quantity:	96 tests
Target:	HIV
Reactivity:	Lentivirus
Method Type:	Sandwich ELISA
Application:	ELISA

Product Details

Brand:	QuickTiter™
Sample Type:	Cell Samples
Analytical Method:	Quantitative
Detection Method:	Colorimetric
Characteristics:	QuickTiter™ HIV Lentiviral Quantitation Kit (HIV p24 ELISA) is an enzyme immunoassay developed for detection and quantitation of the HIV-1 p24 core protein. A mouse monoclonal antibody to HIV-1 p24 is coated onto strip wells of microtiter plate. The quantity of HIV p24 antigen is determined by comparing its absorbance with that of known recombinant p24 antigen standard curve. The kit has a detection sensitivity limit of 1 ng/mL HIV p24, or about 10,000 to 100,000 TU/mL VSVG-pseudotyped lentivirus samples3-5. Each kit provides sufficient reagents to perform up to 96 assays including standard curve and unknown samples. The kit is suitable for both viral supernatant and purified virus. The QuickTiter™ HIV Lentiviral Quantitation Kit is intended for research use only, and not for diagnostic applications. Related Products 1. LTV-100: 293LTV Cell Line 2. LTV-200: ViraDuctin™ Lentivirus Transduction Kit 3. LTV-300: GFP Lentivirus Control 4. VPK-107: QuickTiter™ Lentivirus Titer Kit (Lentivirus-Associated HIV p24) 5.

Product Details

	VPK-112: QuickTiter™ Lentivirus Quantitation Kit 6. VPK-211-PAN: ViraSafe™ Universal Lentivirus Expression System, Pantropic 7. VPK-211: pSMPUW Universal Lentiviral Expression Vector (Promoterless)
Components:	Box 1 (shipped at room temperature) 1. Anti-p24 Antibody Coated Plate : One strip well 96-well plate. 2. FITC-Conjugated Anti-p24 Monoclonal Antibody : One 20 µL vial. 3. HRP-Conjugated Anti-FITC Monoclonal Antibody : One 20 µL vial. 4. Assay Diluent : One 50 mL bottle. 5. Triton X-100 Solution : One 15 mL bottle containing 5% Triton X-100 in TBS. 6. 10X Wash Buffer : One 100 mL bottle. 7. Substrate Solution : One 12 mL amber bottle. 8. Stop Solution (Part. No. 310808): One 12 mL bottle. Box 2 (shipped on blue ice packs) 1. Recombinant p24 Standard : One 100 µL vial of 10 µg/mL heat inactivated recombinant HIV1 p24 antigen in TBS plus BSA.
Material not included:	1. Viral Sample: purified virus or unpurified viral supernatant 2. Cell Culture Centrifuge 3. 0.45 µm filter 4. 10 µL to 1000 µL adjustable single channel micropipettes with disposable tips 5. 50 µL to 300 µL adjustable multichannel micropipette with disposable tips 6. Multichannel micropipette reservoir 7. Microplate reader capable of reading at 450 nm (620 nm as optional reference wave length) 3

Target Details

Target:	HIV
Abstract:	HIV Products
Target Type:	Virus
Background:	Lentivirus vector based on the human immunodeficiency virus-1 (HIV-1) has become a promising vector for gene transfer studies. The advantageous feature of lentivirus vector is the ability of gene transfer and integration into dividing and non-dividing cells1-2. The pseudotyped envelope with vesicular stomatitis virus envelope G (VSV-G) protein broadens the target cell range. Lentiviral vectors have been shown to deliver genes to neurons, lymphocytes and macrophages, cell types that previous retrovirus vectors could not be used. Lentiviral vectors have also proven to be effective in transducing brain, liver, muscle, and retina in vivo without

Target Details

toxicity or immune responses. Recently, the lentivirus system is widely used to integrate siRNA efficiently in a wide variety of cell lines and primary cells both in vitro and in vivo. Lentivirus particles are produced from 293T cells through transient transfection of 3 or 4 plasmids that encodes for the components of the virion. Viral medium containing viral particles produced by packaging cells within 48-72 hr can be harvested. To ensure that pseudoviral medium is viable, and to control the number of copies of integrated viral constructs per target cell, the viral titer needs to be determined before proceeding with transduction experiments. Viral titer can be determined by transduction of HT-1080 or HeLa cells, and followed by antibiotic selection of stable clones. However, it takes weeks to generate sizable stable cell colonies for counting and calculating the titer results.

Application Details

Comment:	• Measures the p24 core protein of recombinant HIV-1 based lentivirus • Lentivirus quantitation is performed on a standard microplate reader • p24 Standard included
Plate:	Pre-coated
Protocol:	An anti-HIV p24 monoclonal coating antibody is adsorbed onto a microtiter plate. p24 antigen present in the sample or standard binds to the antibodies adsorbed on the plate, a FITC-conjugated mouse anti- p24 antibody is added and binds to p24 antigen captured by the first antibody. Following incubation and wash steps, a HRP-conjugated mouse anti-FITC antibody is added and binds to the FITC conjugated anti-p24. Following unbound HRP-conjugated mouse anti-FITC antibody is removed during a wash step, and substrate solution reactive with HRP is added to the wells. A colored product is formed in proportion to the amount of p24 antigen present in the sample. The reaction is terminated by addition of acid and absorbance is measured at 450 nm. A standard curve is prepared from recombinant HIV-1 p24 protein and sample p24 concentration is then determined.
Assay Procedure:	1. Prepare and mix all reagents thoroughly before use. 2. Each lentiviral sample, HIV p24 standard, blank, and control medium should be assayed in duplicate. 3. Add 110 µL of inactivated sample or p24 antigen standard to anti-p24 antibody coated plate. 4. Cover with a Plate Cover and incubate at 37 °C for at least 4 hours or 4 °C overnight. 5. Remove Plate Cover and empty wells. Wash microwell strips 3 times with 250 µL 1X Wash Buffer per well with thorough aspiration between each wash. After the last wash, empty wells and tap microwell strips on absorbent pad or paper towel to remove excess 1X Wash Buffer. 6. Add 100 µL of the diluted FITC-Conjugated Anti-p24 Monoclonal Antibody to each well. 7. Cover with a Plate Cover and incubate at room temperature for 1 hour on an orbital shaker.

Application Details

8. Remove Plate Cover and empty wells. Wash the strip wells 3 times according to step 5 above.
9. Add 100 µL of the diluted HRP-Conjugated Anti-FITC Monoclonal Antibody to all wells.
10. Cover with a Plate Cover and incubate at room temperature for 1 hour on an orbital shaker.
11. Remove Plate Cover and empty wells. Wash microwell strips 3 times according to step 5 above. Proceed immediately to the next step.
12. Warm Substrate Solution to room temperature. Add 100 µL of Substrate Solution to each well, including the blank wells. Incubate at room temperature on an orbital shaker. Actual incubation time may vary from 2-30 minutes. Note: Watch plate carefully, if color changes rapidly, the reaction may need to be stopped sooner to prevent saturation.
13. Stop the enzyme reaction by adding 100 µL of Stop Solution into each well, including the blank wells. Results should be read immediately (color will fade over time).
14. Read absorbance of each microwell on a spectrophotometer using 450 nm as the primary wave length. 5

Restrictions:	For Research Use only

Handling

Precaution of Use:	Remember that your samples contain infectious viruses before inactivation, you must follow the recommended NIH guidelines for all materials containing BSL-2 organims.
Storage:	4 °C/-20 °C
Storage Comment:	Upon receiving, aliquot and store recombinant HIV-1 p24 Standard at -20°C and avoid freeze/thaw. Store all other components at 4°C until their expiration dates.

Publications

Product cited in:	Oh, Chang, Song, Rhee, Joe, Lee, Yi, Lee: "Combined Nurr1 and Foxa2 roles in the therapy of Parkinson's disease." in: **EMBO molecular medicine**, Vol. 7, Issue 5, pp. 510-25, (2015) (PubMed). Lin, Gao, Zhang, Ma, Shen, Hu, Zhang, Zhao, Lan, Liu: "Use of a Novel Integrase-Deficient Lentivirus for Targeted Anti-Cancer Therapy With Survivin Promoter-Driven Diphtheria Toxin A." in: **Medicine**, Vol. 94, Issue 31, pp. e1301, (2015) (PubMed). Haqqani, Marek, Kumar, Davenport, Wang, Tilton: "Central memory CD4+ T cells are preferential targets of double infection by HIV-1." in: **Virology journal**, Vol. 12, pp. 184, (2015) (PubMed). Yi, He, Rhee, Park, Takizawa, Nakashima, Lee: "Foxa2 acts as a co-activator potentiating

expression of the Nurr1-induced DA phenotype via epigenetic regulation." in: **Development (Cambridge, England)**, Vol. 141, Issue 4, pp. 761-72, (2014) (PubMed).

Lucera, Tilton, Mao, Dobrowolski, Tabler, Haqqani, Karn, Tilton: "The histone deacetylase inhibitor vorinostat (SAHA) increases the susceptibility of uninfected CD4+ T cells to HIV by increasing the kinetics and efficiency of postentry viral events." in: **Journal of virology**, Vol. 88, Issue 18, pp. 10803-12, (2014) (PubMed).

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