Lentivirus Production Report

Produced by | P.I. | Date produced

Generation ☐ 2nd ☐ 3rd

Transgene

☐ virus is oncogenic

Plasmids
(indicate vector backbone and envelope and packaging plasmids)

Storage Location

Titre

Describe titring method

Replication competent Lentivirus testing

Complete this section if you wish to use your virus in a level II facility. The test must be done only once for each plasmid you will use to make virus, unless your virus contains an oncogene, in which case every cell line made must be tested before it can be removed from the virus core facility.

A
Percent transduction
(Attach data. Transduction must be >50%)

Date of transduction

Date sups were harvested

B ELISA results
(Attach data)

	[p24] (ng/ml)
positive control	
negative control	
test cells	

Procedure

1. Plate 30,000 HEK 293T cells in a 24-well plate in 500 ul media (IMDM or DMEM with 10% FCS and 2mM L-glutamine). Allow to adhere 3 hours. Add virus particles in 500 ul media at an MOI of 5:1 to 10:1.
2. 5-7 days post-transduction, check transduction efficiency and record in Box A. At least 50% transduction efficiency is required to continue with the assay.
3. Culture cells for 3 weeks, splitting as necessary.
4. After 3 weeks, count the cells and plate 60,000 cells in a 24-well plate in a total volume of 1 ml fresh media. Incubate for 48 hours and then collect supernatant. Freeze at -80 or continue with step 5.
5. Detect p24 by ELISA using the QuickTiter Lentivirus Kit by Cell Biolabs (Cedarlane catalogue #VPK-107). Positive control is virus diluted in media (add Triton X to inactivate the live virus as per the instructions in the kit). Negative control is media alone. Report results in Box B and attach ELISA data.