

## Certificate Of Analysis

### Product Description

KILR<sup>®</sup> Retroparticles for Adherent Cells (Hygromycin B) contains MMLV particles, that can transduce a wide variety of adherent cells, resulting in high level expression with a housekeeping gene tagged with  $\beta$ -gal reporter fragment, ProLabel<sup>®</sup> (ePL). This results in a high level of expression of the fusion protein inside the target cells. Target cell death in a cytotoxicity assay results in the release of the ePL-tagged protein into the medium. Addition of KILR detection reagent, containing the complementing  $\beta$ -gal reporter fragment, Enzyme Acceptor (EA), results in complementation of the two enzyme fragments (EA and ePL). The resulting functional enzyme hydrolyzes a substrate to generate a chemiluminescent signal.

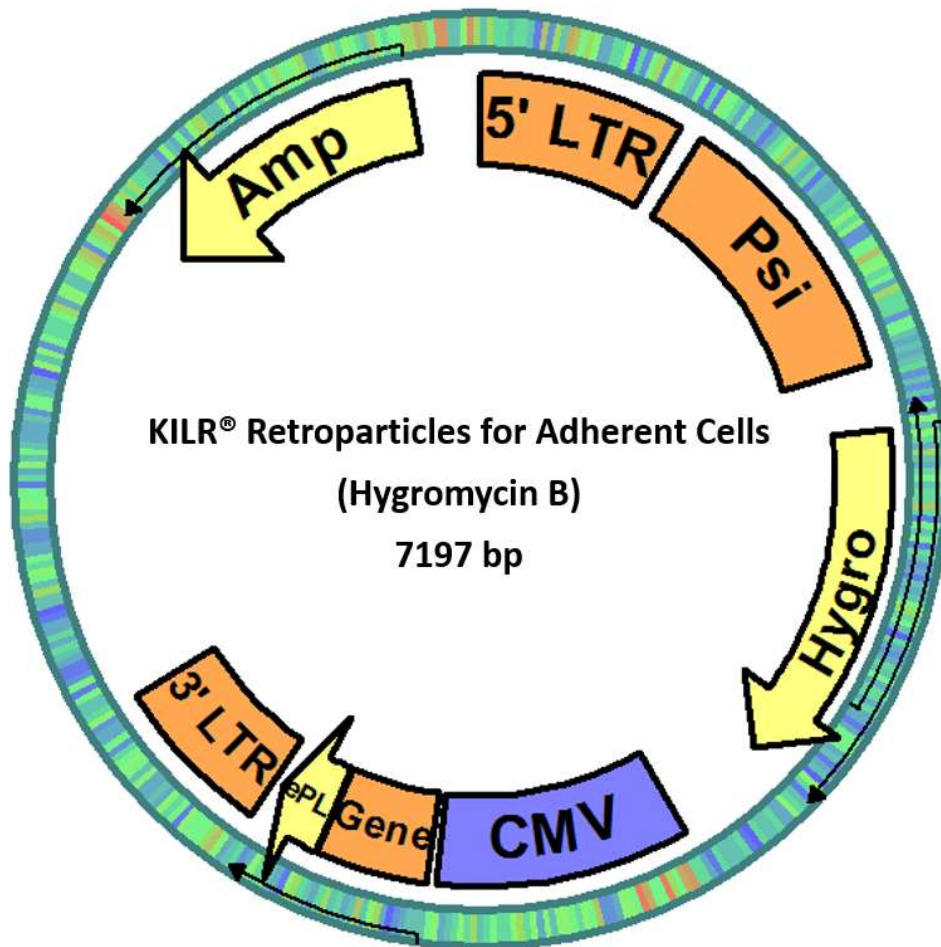
Product Information	
Product Name	KILR <sup>®</sup> Retroparticles for Adherent Cells (Hygromycin B)
Catalog Number	97-0005
Cryovial Label	KILR <sup>®</sup> Retroparticles (HYGRO) Adherent Part #30-565
Cryovial Part Number	30-565
Lot Number	21J1612
Vial Contents	0.5 mL

Shipping and Storage Information	
Storage Conditions	Store at -80°C. Do not freeze/thaw.
Shipping Conditions	Dry ice (-80°C)
Expiration Date	12/28/2023

**IMPORTANT SAFETY NOTE:** Replication-defective retroviral particles, such as provided in this product are not known to cause any diseases in humans or animals. However, retroviral particles can transduce, express protein and/or integrate into human cells. Accordingly, this material is in Risk Group 2 and should be handled under BSL2 controls as defined by the US Public Health Service. Please refer to the CDC Biosafety Manual: <http://www.cdc.gov/biosafety/publications/bmb15/index.htm> for details.

Use and Handling	
Biosafety Level	2 (Biosafety classification is based on US Public Health Service guidelines)
Product User Manual	KILR <sup>®</sup> Retroparticles for Cytotoxicity Assays; part # 70-373
Single Use	For one time use only. Repeated freeze/ thaw will result in loss of activity.
Recommended Use	Transduction of Adherent cells to generate KILR cell lines for use in cytotoxicity assays.
Acceptable Use	Research Use Only. Not for use in Humans.

Retroparticles Vector Information		
Vector	pMLV backbone	Vector identification was confirmed by sequencing
Viral Elements	5' and 3' LTRs	
Viral Replication Status	Replication incompetent retroviral particles - helper virus free. Retrovirus can only infect dividing cells.	
Antibiotic Resistance	Hygromycin B	Viral LTR
Reporter	Proprietary Housekeeping Protein	CMV
Viral Pseudotype	VSV-G envelope	Suitable for infecting all mammalian cell types



### Quality Control Data

Titer: The titer of viral particles was determined by colony formation at limiting dilution on adherent U2OS cells after 7 days under selection with the appropriate antibiotic concentration. Titer (colony forming units/mL, abbreviated pfu/mL) was calculated by multiplying the number of colonies per well by the dilution factor divided by the volume (in mL) used in the experiment. Additional details of QC tests available upon request.

Analytical QC Tests	
Viral Titer	2 x 10 <sup>2</sup> cfu/mL
Mycoplasma	Passed
Sterility	Passed

Functional: To confirm transduction by these retroviruses, expression of the encoded protein with the EFC reporter fragment tag was functionally assessed. Following transduction and antibiotic selection for 7 days, the target cells were treated ± addition of the complementary EFC fragment (as indicated below) plus lysis buffer and PathHunter FLASH Detection Reagent (DiscoverX, Cat. # 93-0247). A Signal:Background (S:B) ratio [(RLU + complementary EFC fragment) / (RLU - complementary EFC fragment)] >1 (typically >20) is reflective of transduction by the retroviruses.

Functional Test				
Cell Line	Average RLU (-ED)	Average RLU (+ED)	S:B Ratio	Days in Selection
U2OS	430	66270	154	7

### Signatures

Signature: \_\_\_\_\_

Documented by Rene Hoffman  
Staff Scientist

Date: 2/28/2022

Signature: \_\_\_\_\_

Approved by Paul Shapiro, Ph.D.  
Group Leader, Assay and Product Development

Date: 2/28/2022