

## KILR<sup>®</sup> H322 Cell Pool

**Catalog Number:** 97-1003P020

**Lot Number:**

See Vial

**Contents:** 1 x 10<sup>6</sup> cells per vial in 1 mL

### Background

KILR cell lines are engineered to express an enhanced Prolabel (ePL) tagged housekeeping gene and may sometimes overexpress an untagged version of a receptor. Once the cells have been lysed the ePL-tagged protein is released into the media. Addition of enzyme acceptor (EA) will cause the complementation of the  $\beta$ -galactosidase enzyme fragments, EA and ePL. The resulting functional enzyme will hydrolyze its substrate to generate a chemiluminescent signal.

### Product Information

**Cell Background:** H322

**Cell Line Species:** Human

**Cell Line Source:** ATCC

**Cell Type:** NSCLC

**Culture Mode:** Adherent

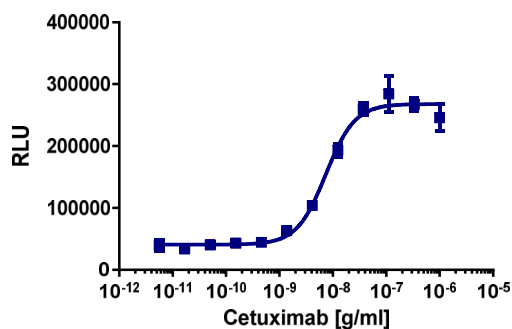
**ADCC Validation:** Cetuximab

**CDC Validation:** N/A

**Storage:** Short term (<24 h): Store at -80°C; Long term (>24 h): Store in vapor phase of liquid nitrogen.

### ADCC Assay Performance

Cells were plated in a 96-well plate and incubated at 37°C and 5% CO<sub>2</sub> for the indicated amount of time. Antibody was added and opsonized for the indicated time (below). Effector cells were added and the plate was incubated at 37°C/5% CO<sub>2</sub> using the assay conditions described below. Target cell death was detected using the KILR detection reagent according to the recommended protocol. Additional reagents needed are noted on this document.



<b>Target Cell Number/Well:</b>	10,000
<b>Effector Type:</b>	primary PBMC's (frozen)
<b>Effector Species:</b>	Human
<b>Effector Cell Number/Well:</b>	250,000
<b>Effector to Target Ratio:</b>	25:1
<b>Control Antibody:</b>	Cetuximab
<b>Cell Seeding Time (minutes):</b>	30
<b>Antibody Incubation Time (minutes):</b>	30
<b>Antibody Incubation Temperature (°C):</b>	37
<b>Assay Incubation Time (minutes):</b>	180
<b>Assay Incubation Temperature (°C):</b>	37
<b>Incubation with KILR detection reagent (hours):</b>	1
<b>EC<sub>50</sub> for Antibody:</b>	7.4
<b>Signal:Background Ratio:</b>	8.4
<b>Max % Lysis:</b>	9

**Recommended Culture Conditions:** Split cells when monolayer reaches 80% confluency. Recommended seeding density:  $1-3 \times 10^5$  cells/ cm<sup>2</sup>; Recommended Split ratio: 1:3 to 1:6; Medium renewal: Every 2 to 3 days.

**Note:** Higher % ADCC is observed when primary NK cells (vs PBMCs) are used as effectors (e.g. at 10:1 ratio of NK: target cells; 1h opsonization with antibody).

### Passage Stability

This cell line has been confirmed to be stable through 15 passages with no significant drop in assay window or change in EC<sub>50</sub>.

### Mycoplasma Testing

This lot was tested and found to be free of mycoplasma contamination. Data available upon request.

### Required Materials

The following additional materials are required but not provided:

Product Use*	Product Description	Catalog Number
Detection	KILR <sup>®</sup> Detection Kit	97-0001M
Cell Culture	AssayComplete™ Cell Culture Kit-101	92-3101G
Cell Plating	AssayComplete™ Cell Plating 39 Reagent	93-0563R39A
Cell Detachment	AssayComplete™ Cell Detachment Reagent	92-0009
Cell Thawing	AssayComplete™ Thawing Reagent T6	92-4106TR
Cell Freezing	AssayComplete™ Freezing Reagent F5	92-5105FR
Ligand Dilution	AssayComplete™ Protein Dilution Buffer	92-0023

\*Please inquire about our cell line-specific AssayComplete Starter Packs to get you started with your cell culture needs.

### Required Antibiotics

Antibiotic Name	Concentration (µg/mL)	Catalog Number
AssayComplete™ Puromycin	Not Applicable	Not Applicable
AssayComplete™ Hygromycin B	Not Applicable	Not Applicable
AssayComplete™ G418	300	92-0030

### Additional Ligand Information

**Control Compound:** Cetuximab

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