



# **RCL (VSVG) Gene Copy Number Detection Kit**

RCL (VSVG) Gene Copy Number Detection Kit is designed for precise quantification of RCL (VSVG) gene copy numbers in biological products.

Catalog number: ARG83103

Package: 100 tests

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### **INTRODUCTION**

RCL (VSVG) Gene Copy Number Detection Kit is designed for the quantitative detection of RCL gene copy number in the genome of CAR-T cells prepared by using HIV-1 lentiviral vector technology.

RCL (VSVG) Gene Copy Number Detection Kit adopts the fluorescent probe method and multiplex PCR method to detect the DNA sequence related to integration or expression function on the transfer plasmid, and the VSVG gene copy number in the sample can be calculated. The kit is a rapid, specific and reliable device.

### **PRINCIPLE OF THE ASSAY**

RCL (VSVG) Gene Copy Number Detection Kit utilizes quantitative polymerase chain reaction (qPCR) technology to accurately detect and quantify RCL gene copy numbers.

RCL (VSVG) Gene Copy Number Detection Kit includes a set of primers and probes specifically designed to amplify and detect the targeted DNA sequences associated with RCL genes. By employing qPCR, which combines amplification and detection in real-time using a fluorescent dye, this kit offers high specificity and sensitivity. It is user-friendly and well-suited for laboratory applications.

### MATERIALS PROVIDED & STORAGE INFORMATION

Store the unopened kit at -20 °C. Use the kit before expiration date.

Component	Quantity	Storage information
20X RCL Standard	50 µl	-20°C
RCL Primer & probe mix	550 µl	-20°C (protect from light)
2x qPCR Reaction Buffer	1.1 ml	-20°C (protect from light)
DNA Dilution buffer	3 x 1 ml/vials	-20°C
ROX (High)	50 µl/vials	-20°C (protect from light)
ROX (Low)	50 µl/vials	-20°C (protect from light)

### MATERIALS REQUIRED BUT NOT PROVIDED

- PCR machine
- Pipettes and pipette tips
- DNase/RNase-Free Water
- PCR tube

### TECHNICAL HINTS AND PRECAUTIONS

- Wear protective gloves, clothing, eye, and face protection especially while handling blood or body fluid samples.
- Store the kit at -20°C at all times.
- All reagents must be kept on ice during the entire experiment.
- Once the assay has been started, all steps should be completed without interruption.
- It is highly recommended that the standards and samples be assayed in triplicates.
- Change pipette tips between the addition of different reagent or samples.

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### REAGENT PREPARATION

- **Standards:** Dilute 20X RCL Standard with DNA Dilution buffer to yield S6. The DNA Dilution buffer serves as zero standard (0 pg/ml), and the rest of the standard 10-fold serial

Dilute RCL DNA standard as according to the table below:

Standard	DNA Conc. (copies)	µl of DNA Dilution buffer	µl of standard
S6	1 X 10 <sup>6</sup>	95 µl	5 µl (stock)
S5	1 X 10 <sup>5</sup>	90 µl	10 µl (S6)
S4	1 X 10 <sup>4</sup>	90 µl	10 µl (S5)
S3	1 X 10 <sup>3</sup>	90 µl	10 µl(S4)
S2	1 X 10 <sup>2</sup>	90 µl	10 µl(S3)
S1	1 X 10 <sup>1</sup>	90 µl	10 µl(S2)
S0	0l	0 µl	100µl

### ASSAY PROCEDURE

- 1 Prepare qPCR mix buffer:

2x qRCR Reaction Buffer	10µl
RCL Primer & probe mix	4.6µl
ROX *	0.4µl
<b>Total</b>	<b>15µl (1 wells)</b>

- \* Choose the appropriate ROX (High or Low) to match the PCR machine. If the PCR machine does not require ROX, adjust the volume with DNase/RNase-free water to obtain a final volume of 15µl.
- 2 Mix 15µl qPCR mix buffer with 5µl diluent standard / sample / blank in PCR tube. The final volume should be 20 µl.
  - 3 Initial denaturation: 95°C, 2 min
  - 4 PCR cycle:
    - Denaturation: 95°C, 15 sec
    - Elongation: 60°C, 15 sec, for **45 cycle, 20 µl.**