



## **AAVPrime™ AAV Serotype Testing Kit**

Cat. No. AA320

## **AAVPrime™ AAV Serotype Testing Kit Plus**

Cat. No. AA321

### **Datasheet**

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## AAVPrime™ AAV Serotype Testing Kit Plus

### I. Introduction

GeneCopoeia's AAVPrime™ Adeno-associated virus (AAV) Serotype Testing Kit (Cat. No. AA320) contains 9 premade GFP-expressing AAV in serotypes 1, 2, 3, 6, 7, 8, 9, DJ and DJ/8. AAVPrime™ Adeno-associated virus (AAV) Serotype Testing Kit Plus (Cat. No. AA321) contains 12 premade GFP-expressing AAV in serotypes 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, DJ and DJ/8. All AAV particles are offered at  $5 \times 10^{12}$  GC/ml, in 25  $\mu$ l stocks. These kits are ideal for testing the infection efficiency of AAV serotypes on different cell types.

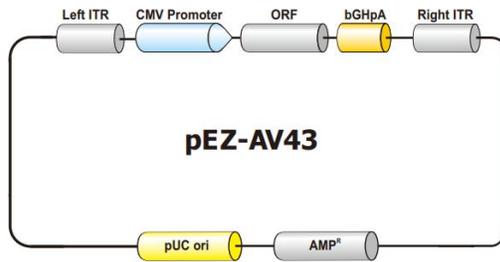
### II. Contents and Storage

AAV stocks are supplied in liquid form at the indicated titer in PBS storage solution. Store at  $-80^{\circ}\text{C}$ . If desired, aliquot viral stock upon arrival, and store the aliquots at  $-80^{\circ}\text{C}$  immediately. Avoid repeated freeze and thaw.

Catalog	Products	Titer	Quantity
AA601-025	eGFP AAVPrime™ Purified Adeno-associated Viral Particles. Serotype: 1. Promoter: CMV.	$5 \times 10^{12}$ GC/ml	25 $\mu$ l
AA001-025	eGFP AAVPrime™ Purified Adeno-associated Viral Particles. Serotype: 2. Promoter: CMV.	$5 \times 10^{12}$ GC/ml	25 $\mu$ l
AA801-025	eGFP AAVPrime™ Purified Adeno-associated Viral Particles. Serotype: 3. Promoter: CMV.	$5 \times 10^{12}$ GC/ml	25 $\mu$ l
AA074-025*	eGFP AAVPrime™ Purified Adeno-associated Viral Particles. Serotype: 4. Promoter: CMV.	$5 \times 10^{12}$ GC/ml	25 $\mu$ l
AA084-025*	eGFP AAVPrime™ Purified Adeno-associated Viral Particles. Serotype: 5. Promoter: CMV.	$5 \times 10^{12}$ GC/ml	25 $\mu$ l
AA097-025	eGFP AAVPrime™ Purified Adeno-associated Viral Particles. Serotype: 6. Promoter: CMV.	$5 \times 10^{12}$ GC/ml	25 $\mu$ l
AA107-025	eGFP AAVPrime™ Purified Adeno-associated Viral Particles. Serotype: 7. Promoter: CMV.	$5 \times 10^{12}$ GC/ml	25 $\mu$ l
AA117-025	eGFP AAVPrime™ Purified Adeno-associated Viral Particle . Serotype: 8. Promoter: CMV.	$5 \times 10^{12}$ GC/ml	25 $\mu$ l
AA016-025	eGFP AAVPrime™ Purified Adeno-associated Viral Particles. Serotype: 9. Promoter: CMV.	$5 \times 10^{12}$ GC/ml	25 $\mu$ l
AA127-025*	eGFP AAVPrime™ Purified Adeno-associated Viral Particles. Serotype: 10. Promoter: CMV.	$5 \times 10^{12}$ GC/ml	25 $\mu$ l
AA201-025	eGFP AAVPrime™ Purified Adeno-associated Viral Particles. Serotype: DJ. Promoter: CMV.	$5 \times 10^{12}$ GC/ml	25 $\mu$ l
AA401-025	eGFP AAVPrime™ Purified Adeno-associated Viral Particles. Serotype: DJ/8. Promoter: CMV.	$5 \times 10^{12}$ GC/ml	25 $\mu$ l

\* AA074-025, AA084-025, and AA127-025 are not included in AAVPrime™ AAV Serotype Testing Kit (AA320)

### III. Plasmid Map



### IV. Quality Control

Every lot is tested for performance consistency, titer, sterility and purity.

### V. Experimental Procedure

The procedure below is based on the transduction of HT1080 cells with the AAV Serotype Testing Kit Plus in 24 well plates. Other cells can also be used, but efficiencies may vary up to several orders of magnitude.

#### Day 1: Plate cells to be transduced

1. Plate ~  $1.3 \times 10^5$  cells per well in a 24-well plate 24 hours prior to viral infection. Use 1 ml of culture medium supplemented with 10% heat-inactivated fetal bovine serum for each well. Incubate the cells at 37°C with 5% CO<sub>2</sub> overnight until 80% confluent.

#### Day 2: Transduce target cells

2. (Optional) Treat cells with 0.8 μM camptothecin in the medium. Mix well by swirling, then return the plates to the 37°C incubator for 4 hours. This step is optional but can increase the transduction efficiency for many cell lines.

3. Calculate the volume of viral stock to be added to the cells according to the desired multiplicity of infection (MOI).

$$\text{MOI} = \text{AAV GC particles needed} / \text{Number of cells to be infected}$$

For an MOI = 1, the volume (in μl) of AAV particles needed = ((total number of cells per well)/(number of genome copies (GC)/ml)) x 1,000.

The MOI used is critical to achieve 100% infection of the target cells without causing major side effects. A range of MOI from 1,000 to 10,000 is suggested for most cell lines, however, up to 500,000 MOI may be needed for some cell lines with some specific serotype AAV particles.

**To determine the MOI needed** when transducing a cell line for the first time, we recommend first to transduce the target cells with the same serotype eGFP reporter AAV particles at a series of MOIs, such as MOIs of 100, 1,000, and 10,000, or even higher for some cell lines that are difficult to be transduced.

**Example:**  $3 \times 10^5$  HT1080 cells were prepared for transduction by AAV particles ( $1 \times 10^{11}$  GC/ml) one day after plating. For an MOI = 1,000, 3μl AAV particles are needed. For an MOI = 10,000, 30μl AAV particles are needed.

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4. Thaw the AAV particles at room temperature. Dilute the viral stock to the needed MOI in 200µl culture medium supplemented with 2% (V/V) heat-inactivated FBS.
5. Aspirate the medium from the camptothecin-treated (optional) cells. Add the 200 µl of diluted virus to the cells.
6. Incubate 2 hours at 37°C in a CO<sub>2</sub> incubator, swirling the plate every 30 minutes.
7. Add 200µl pre-warmed culture medium with 18% (V/V) heat-inactivated FBS, and incubate for 40-48 hours.

### **Day 4: Replace medium**

8. Remove the medium containing AAV particles from the wells and replace with 1 ml fresh pre-warmed culture medium with 10% (V/V) heat-inactivated FBS.

### **Day 5 to 14:**

9. Harvest the cells for further investigation.

GeneCopoeia is committed to providing our customers with high-quality products. If you should have any questions or concerns about any GeneCopoeia products, please contact us at (301)-762-0888.

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