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ViroMag CRISPR transduction reagent

Protocol

For CRISPR/CAS9 viral vectors

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PolyMag CRISPR Reagent User Guide

Package contents	**VMC50200:** 200µl of ViroMag CRISPR reagent **VMC51000**: 1mL of ViroMag CRISPR reagent **KVC50100** : 100µL of ViroMag CRISPR reagent + 1 Super Magnetic plate
Storage Conditions	Store at 4°C upon receipt
Product Description	ViroMag CRISPR reagent is the only magnetic viral transduction enhancer for CRISPR/Cas9 viruses (adenovirus, lentivirus, retrovirus...) for gene editing applications.
Important guidelines	This reagent has to be used with a magnetic plate For Research use only. Not for use in diagnostic procedures

BEFORE YOU BEGIN:

• The viral vector solution and ViroMag CRISPR Should be used at room temperature and be gently vortexed prior to use.

• All the complexes must be prepared in medium without serum and supplement.

• It is not recommended to use RPMI during complex preparation, prefer DMEM or PBS.

• For sensitive cells, medium can be replaced with fresh complete culture medium 4 to 6h after transfection.

Tissue Culture Dish	Cell Number per well	ViroMag CRISPR volume per well	Total transduction volume per well
96-well	0.5 – 2.0 x 10^4	1.5 µL	0.2 mL
24-well	0.5 – 1.0 x 10^5	6 µL	0.5 mL
12-well	1.0 – 2.0 x 10^5	12 µL	1.0 mL
6-well	2.0 – 4.0 x 10^5	30 µL	2.0 mL
60 mm dish	0.5 – 1.0 x 10^6	60 µL	4.0 mL
90-100 mm dish	1.0 – 2.0 x 10^6	150 µL	8.0 mL
T75 flask	2.0 – 5.0 x 10^6	150 µL	12.0 mL

Table 1: Recommended cell number, ViroMag CRISPR volumes and transduction volume per well.

PROTOCOL STEPS

The following protocol is given for a single well from a 24-well tissue culture plate containing ~$1x10^5$ cells/well in 400 µL complete culture serum. If a different culture plate format is used, adjust cell number and reagent amounts according to the table 1.

NOTES: ViroMag CRISPR should be stored at +4°C. Use 6 µL of ViroMag CRISPR per well. Viral particles will be diluted in 100 µL of medium without any supplement (DMEM) and added onto ViroMag CRISPR magnetic nanoparticles.

1. Viral particle solution

The total MOI may vary depending on viral constructions and cell type: MOI of 3 for HEK-293 cells, and MOI of 60 for iPS. We recommend using a MOI of 10 to begin.

Prepare a viral suspension* for a final MOI=10 in 100 µL of DMEM without any supplement.

2. ViroMag CRISPR solution

Add 6 µL of ViroMag CRISPR in a new tube.

3. Complexes preparation

A. Add the viral solution onto ViroMag CRISPR nanobeads, mix gently by carefully pipetting up and down.

B. Incubate the mixture for 20 min. at room temperature.

4. Infection

A. Add the magnetic complexes dropwise onto the cells and homogenize by gently rocking the plate side to side to ensure a uniform distribution of the mixture.

B. Place the cells on the magnetic device and incubate at 37°C for 30 min.

C. After 30 min of incubation, remove the magnetic plate.

D. Incubate the cells under your standard culture conditions for 24 to 72h.

OPTIONAL:

Perform a medium change. Keep the magnetic plate beneath the cell culture dish, withdraw the transfection medium and add fresh growth medium. Then, remove the magnetic plate.

* Single virus containing both Cas9 & gRNA or multiple viral particles coding for Cas9 and gRNA.

Additional products for CRISPR Cas9 experiments:

- PolyMag CRISPR for Genome editing using expression plasmids
- RmesFect CRISPR for mRNA transfection
- ProDeliverIN CRISPR for Cas9 protein delivery

Purchaser Notification

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