

Efficient CRISPR gene editing in primary cells and organoids using virus-like particles

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Organisation

Name of the organisation Katholieke Universiteit Leuven (KUL)

Department Chronic Diseases and Metabolism

Specific Research Group or Service

Laboratory of Respiratory Diseases and Thoracic Surgery (BREATHE)

Country Belgium

Geographical Area Flemish Region

Name of the organisation Katholieke Universiteit Leuven (KUL)

Department Pharmaceutical and Pharmacological Sciences

Country Belgium

Geographical Area Flemish Region

Partners and collaborations

Katholieke Universiteit Leuven (KUL)

SCOPE OF THE METHOD

The Method relates to	Human health
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The Method is situated in	Basic Research, Translational - Applied Research
Type of method	In vitro - Ex vivo
Specify the type of cells/tissues/organs	several primary cell models (rectal organoids, endothelial cells, epithelial cells, iPSCs, ...)

DESCRIPTION

Method keywords

organoids
 genome engineering
 gene editing
 CRISPR-Cas9
 CRISPR/Cas
 virus-like particles
 transduction
 base editing
 prime editing
 knock out

Scientific area keywords

gene therapy
 genetics
 3D organoid models
 human diseases
 human health

human adult stem cells

Method description

Using the combination of advanced CRISPR tools including several Cas orthologs, based editors (ABE, CBE, CGBE) and prime editing technologie with efficient delivery vehicles such as LV, AAV and virus-like particles (VLPs), our lab has become experienced with introducing or replacing precise edits in several primary cell models. For gene editing, VLPs are especially well suited because they link the efficacy of viral transduction with the delivery of RNP cargo, delivering a very transient dose of gene editing cargo. We have built VLPs harbouring several gene editing enzymes (Cre, several Cas variants, several base editors, several PE strategies), as well as reporter cargos (mNeonGreen, B-galactosidase, fLuc,...).

Lab equipment

BSL2 is required to work with cells from human origin and with particles capable of entering these cells.

Method status

Still in development

Internally validated

PROS, CONS & FUTURE POTENTIAL

Advantages

Efficient gene editing in hard-to-edit cell types using ultra transient exposure and therefore limiting risks on off-target editing. VLP preps can be ordered from the Leuven Viral Vector Core (<https://gbiomed.kuleuven.be/english/corefacilities/LVVC/technology>)

Challenges

VLP production is expensive and needs to be performed by experienced staff under highly standardised SOP's. We have large libraries of VLPs available and have shared many before under academic collaboration.

REFERENCES, ASSOCIATED DOCUMENTS AND OTHER INFORMATION

Links

[Leuven Viral Vector Core](#)

Other remarks

This work was performed in collaboration with the Leuven Viral Vector Core (Contact: lvvc@kuleuven.be).

Coordinated by



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Vlaanderen
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