evotec

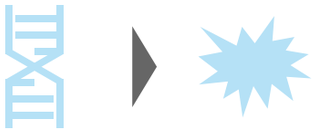
#RESEARCHNEVERSTOPS

Evotec Gene Therapy

Adding value to our partners' research – in vitro overview

Areas of gene therapy expertise at Evotec GT

Innovative and flexible solutions from target identification to IND



Promoter Regulatory elements

- Constitutive and tissue-specific promoter / enhancers
- Ongoing development effort towards proprietary synthetic promoters

Capsid

- Common natural serotypes
- Research licenses for engineered capsids
- Ongoing development of proprietary capsids through internal research and co-development partnerships

Analytics

- Vector quantification and characterisation
- *In vitro* assays: infectivity & *in vitro* biopotency
- Payload specific functional assays
- Immunological assays

Gene Therapy & Gene Editing

Payload

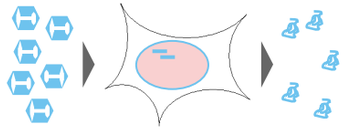
- Codon-optimized transgenes
- Vectorised antibodies
- Antisense approaches (e.g. shRNA, miRNA)
- Genome editing: *in vivo* models to be built

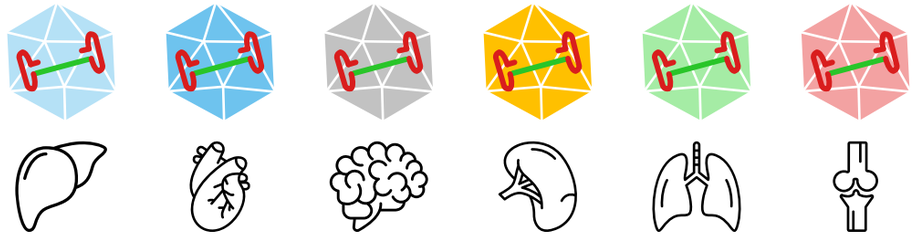
CMC

- Research material (<10^{14} vg)
- Based on HEK293 cell lines (suspension & adherent)

Translation & Nonclinical Dev.

- Pharmacology & biodistribution in humanized or disease models
- Safety pharmacology (integration & off-target analyses)
- Behavioural and functional phenotyping (cognition, physical tests)





Natural Serotypes and Capsid Engineering

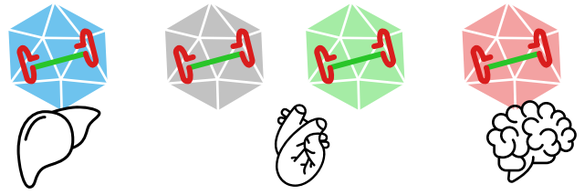
We apply established AAV gene ferries and flexible designs to novel AAV variants

Non-engineered AAV serotypes

- AAV1, AAV2 - 6, AAV7, AAV8, AAV9 or customer provided variants[1)]
- Natural limitations of selective tissue targeting

Selections are informed by

- AAV tropism for targeted organ and cell
- Comparative *in vitro* and in *in vivo* studies
- Biodistribution & transduction studies
- Preexisting Immunity and Immunogenicity
- Manufacturability considerations



Applying known natural transduction profiles

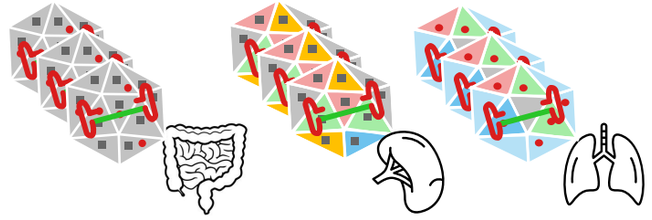
Engineered AAV capsids

- Refined AAV capsids with specific targeting profiles[2)]
  - High-efficiency transduction with broad tissue tropism (AAV-DJ)
  - Increased penetration to brain tissues (AAV-DJ/8)
  - Transduction of murine und human hepatocytes (AAV-LK03), plus human ß-cells (AAV-KP1)
  - Human hepatocyte tropism combined with reduced sero-prevalence (NP40 and NP59)

AAV vector systems with improved attributes

AAV capsid discovery and screening

- Experienced in co-development of improved capsids
  - Directed molecular evolution and rational design with renowned partners
    - Capsid shuffling
    - Peptide insertion
    - Combinatorial point mutations
- We offer co-development of novel AAV capsids fitting customer needs
  - AAV library design and screening



New engineered variants & novel attributes

[1)] Certain Licences may apply
[2)] Research License applies

Gene Editing

Overview of core activities

End to end integrated discovery

Selection of suitable editing tools (ZFN, TALEN, CRISPR) and designs, and delivery systems that fit project needs

- Transfection, electroporation, AAV, Plasmids, RNA, RNP formats

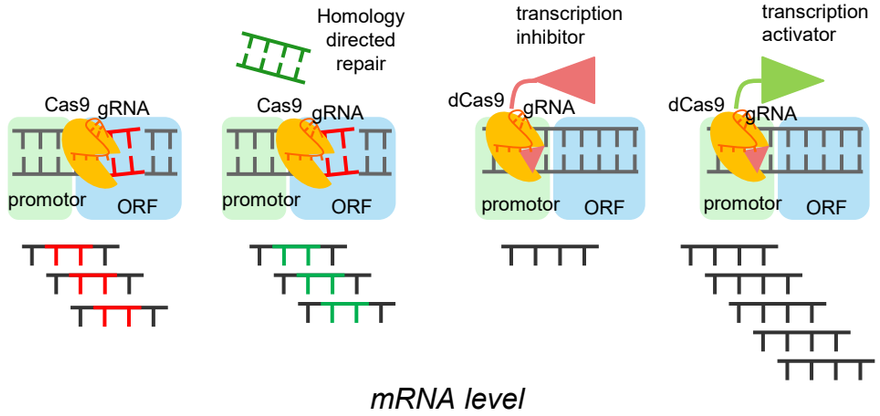
Optimization of editing components and efficiency in various cellular assays tailored to project needs

- Evaluation of on- and off-targets
- Applying specialized technology as needed (e.g. MS, proteomics, RNAseq)

Integration of *in vitro* and *in vivo* areas of expertise

- Optimization of transduction & editing efficiencies
- On/Off target editing analyses
- Assessment of gene editing efficacy in animal models of disease

CRISPR toolbox for genetic approaches



CRISPR, clustered regularly interspaced short palindromic repeats; ZFN, Zinc finger nuclease; TALEN, Transcription activator-like effector nuclease; MS, Mass spectrometry.

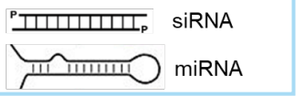
Evotec GT Non-viral LNP Capabilities

Integrated preclinical drug development platform

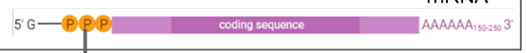
Formulation & Payloads

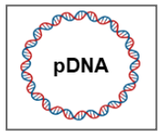
Gene editing/correction payloads

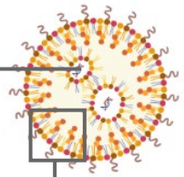
Gene modulation payloads



Gene augmentation/supplementation & vaccination payloads

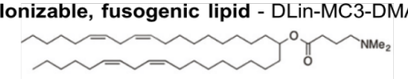




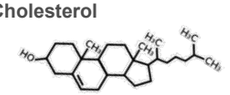


Lipid Nanoparticle (LNP)

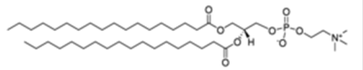
Ionizable, fusogenic lipid - DLin-MC3-DMA



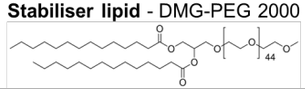
Cholesterol



Zwitterionic, quaternized lipid - DSPC



Stabiliser lipid - DMG-PEG 2000



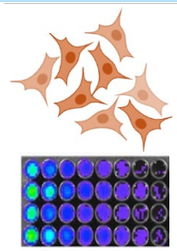
Production platforms & principles

Continuous Solvent-Antisolvent Precipitation

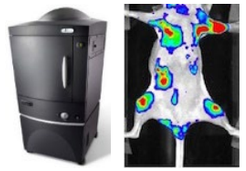
Microfluidics
Laminar mixing

Commercial laminar flow platform

In vitro & *in vivo* testing

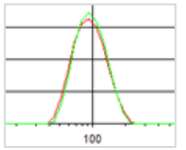


In vitro **potency testing**

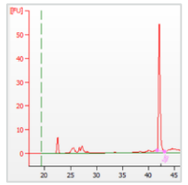


In vivo **PoC, kinetics, efficacy & biodistribution testing**

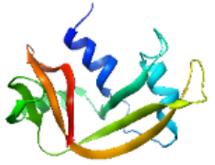
Analytics & QC



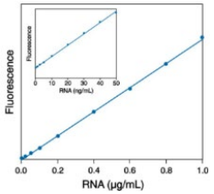
DLS characterization (size distribution, PDI)



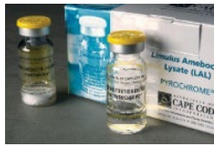
Bioanalyzer platform (payload integrity & stability)



RNAse contamination



Encapsulation efficacy % API concentration



Endotoxin (LAL) assay