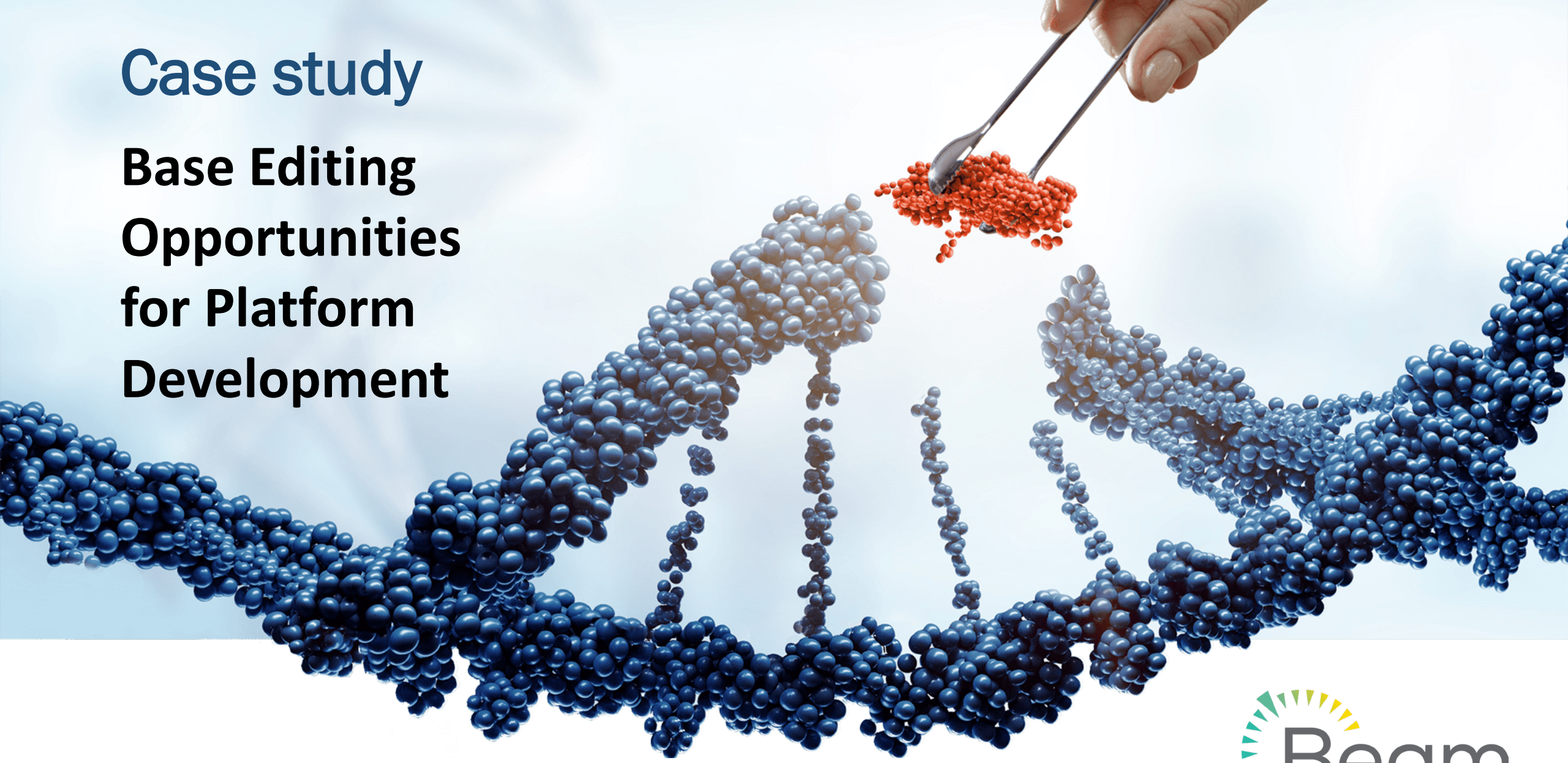
# Case study

## Base Editing Opportunities for Platform Development



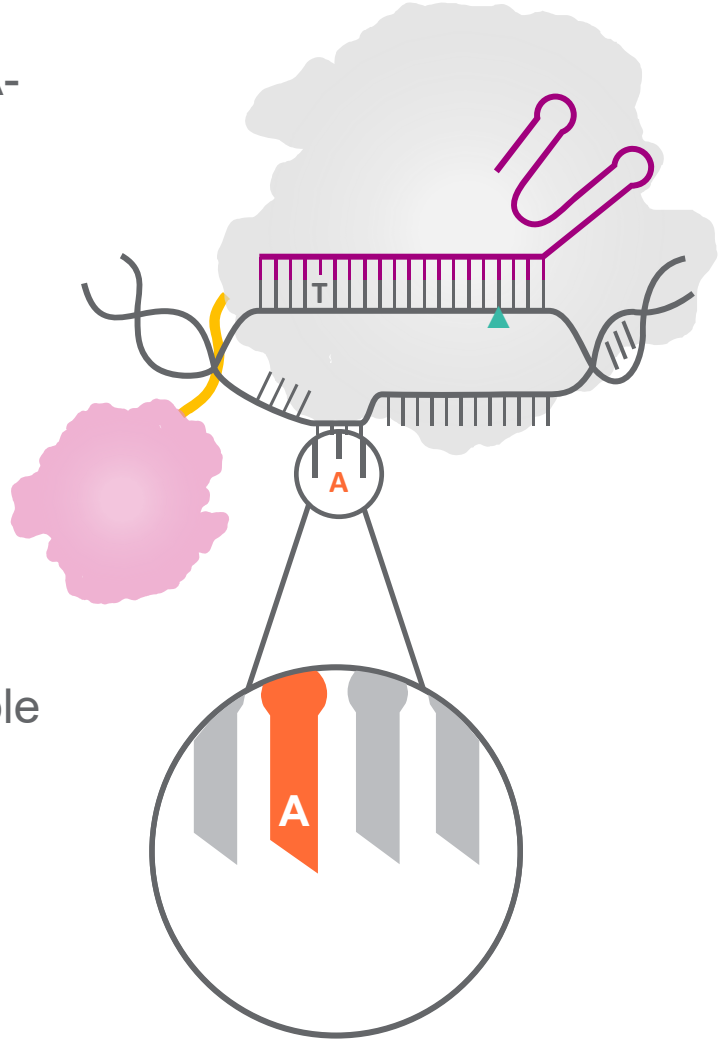
Beam THERAPEUTICS

# Base editors chemically modify target bases, permanently and predictably

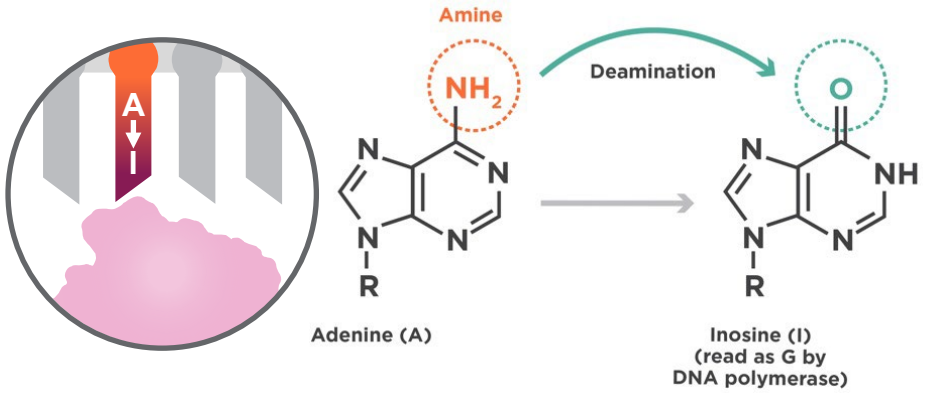
**CRISPR** – established guide RNA-driven DNA targeting:

- Opens 4-5 base single strand DNA window
- Modified to not cause double stranded breaks

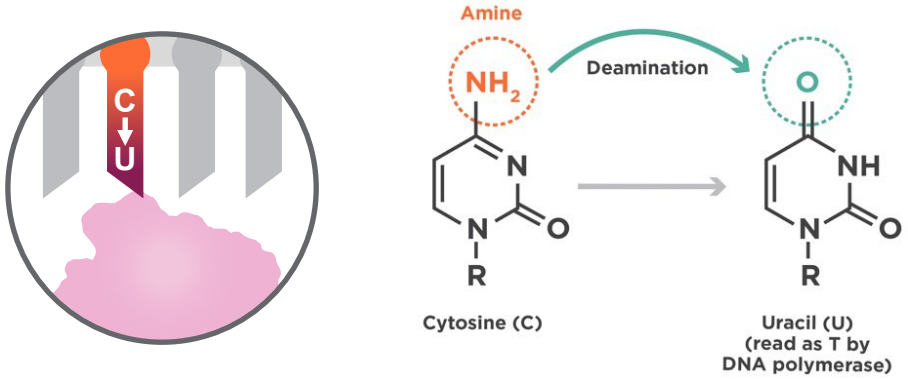
**Deaminase** – operates on single stranded DNA to completes chemical modification at predictable target DNA base




## A-to-G base editor ("ABE")

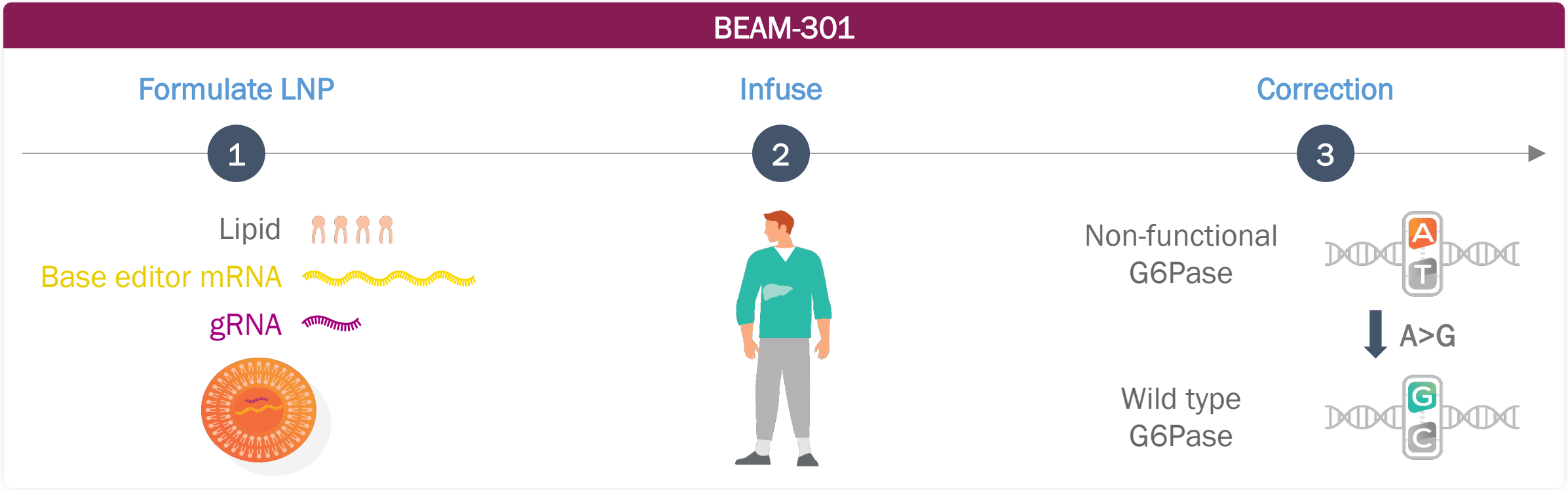



## C-to-T base editor ("CBE")



# The Precision of Base Editing Presents New Opportunities

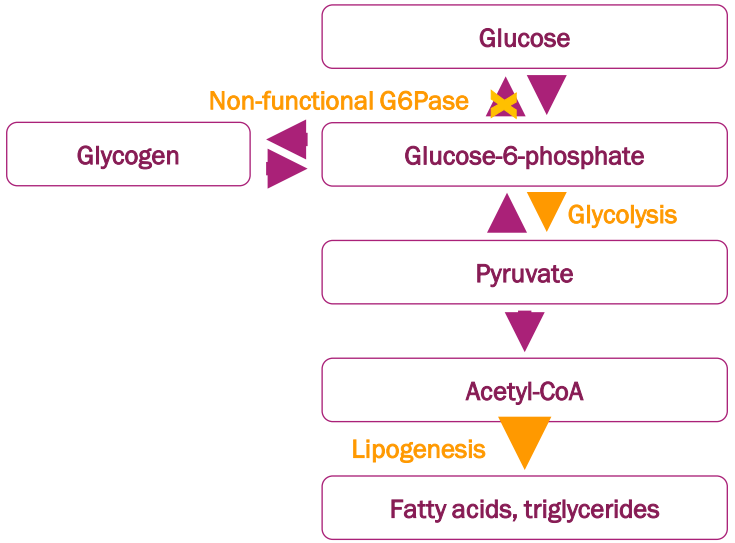
- Base editing can be used to precisely correct single nucleotide variants that cause disease
- Many inborn errors of metabolism can be corrected by gene editing in the liver through LNP-mediated delivery (e.g., BEAM-301 for glycogen storage disease 1a to correct G6Pase)

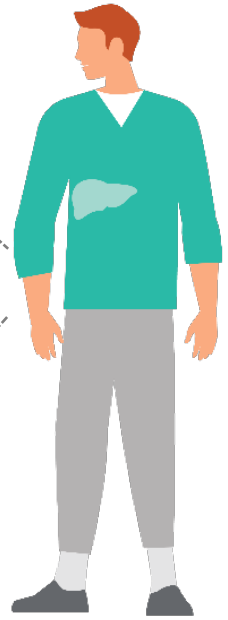


# Base-editing strategy to treat a severe pathogenic variant underlying Glycogen Storage Disease Type Ia


Wild type G6Pase


Non-functional G6Pase



Acute drops in blood glucose levels can result in fasting hypoglycemia, seizures, and death

No treatment beyond dietary management with slow-release glucose

Metabolic and long-term complications include,

- Elevated cholesterol, triglycerides, lactic and uric acid
- Enlarged livers (glycogen, fats)
- Hepatic adenomas, HCC
- Growth impairment

- GSDIa is a genetic disease caused by mutations in the G6PC gene encoding G6Pase, a predominantly liver-expressed enzyme vital to glucose metabolism
- G6PC-p.Arg83Cys (R83C) is a prevalent pathogenic variant associated with severe manifestations of GSDIa
- Beam's base-editing technology has the potential to permanently correct this mutation and restore glucose metabolism

# The Precision of Base Editing Also Presents New Challenges in Drug Development

- Monogenic diseases often have a diverse mutational spectrum
- GSD1a has ‘founder’ mutations which vary by ethnic background and region
- Within ethnicities there is also a long tail of rare variants

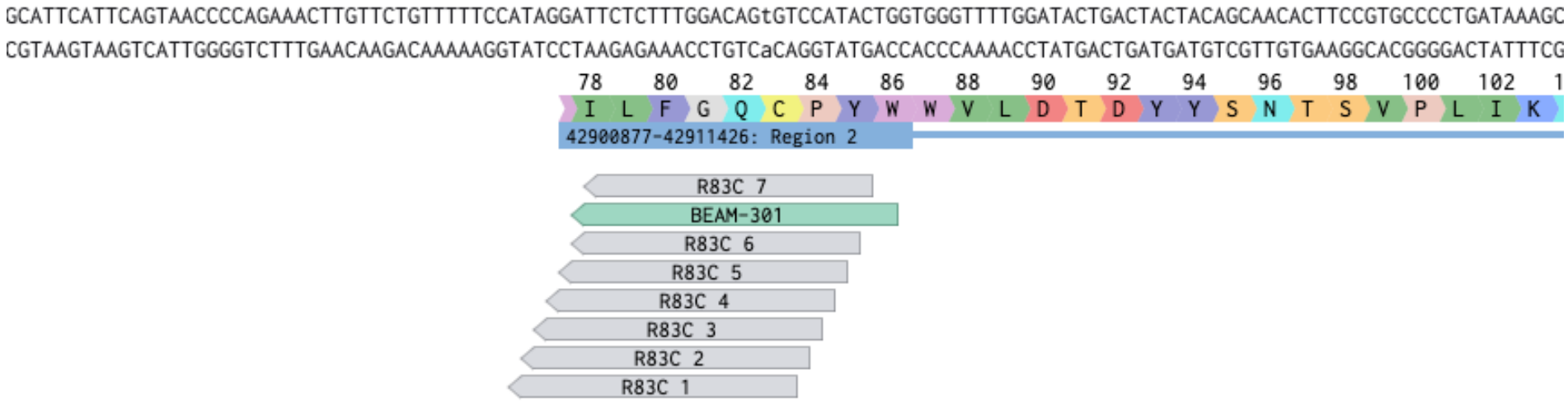
| Caucasian | Chinese | Japanese | Korean | Hispanic | Caucasian Ashkenazi Jewish |
|---|---|---|---|---|---|
| p.R83C (33%) | p.R83H (26%) | c.648G>T (91%) | c.648G>T (75%) | c.380_381insTA (54%) | p.R83C (98%) |
| p.Q347X (18%) | c.648G>T (54%) | | | p.R83C | p.Q347X (2%) |

Hum Mutat. 2008 Jul; 29(7): 921–930.

- **Each disease variant requires its own drug product**
  - Common LNP composition
  - Unique gRNA (required)
  - Customized base editor mRNA (maybe…)

# Challenges at the top of the funnel: lead discovery and optimization

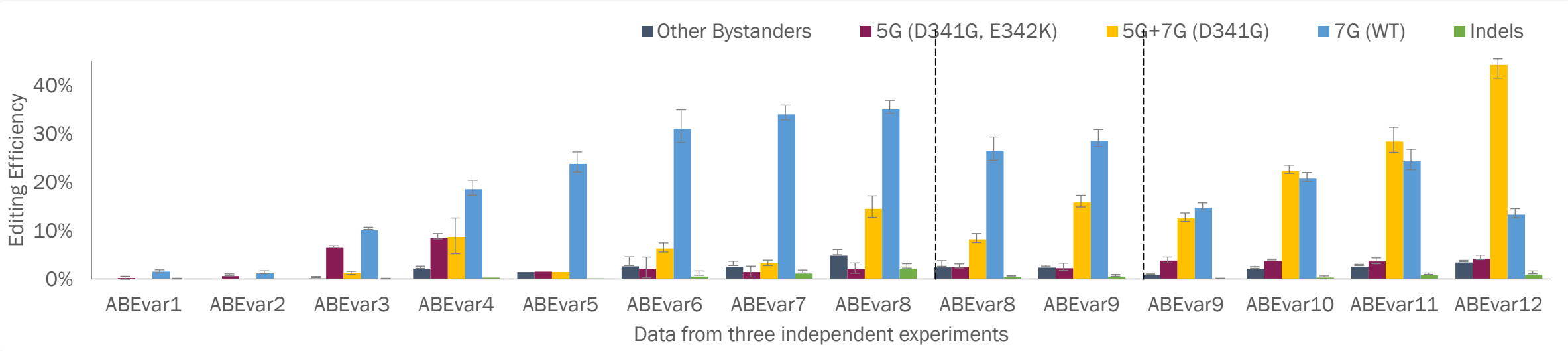
- gRNA screens are limited to the sequence immediately surrounding the disease causing SNV
- This densely tiled gRNA screen requires use of Cas orthologues and variants that accept alternative PAM motifs




- Preliminary hits are often evaluated with a panel of editors with varying deaminases and linkers
  - Optimizing for editing of the target base while minimizing undesirable bystander edits.
  - **Thus far, Beam's development stage programs each use a unique base editor**

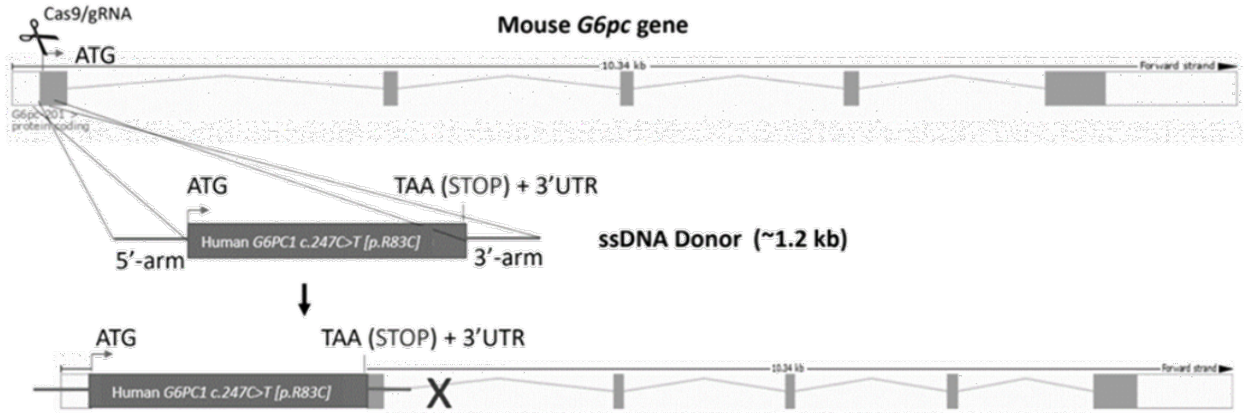
# Case Study: Optimizing a Base Editor to Correct the PiZ mutation in AATD


| Editor ID | TadA Architecture | TadA* variant | PAM variant |
|---|---|---|---|
| ABEvar1 | Dual | 7.10 | 1 |
| ABEvar2 | Dual | 7.10 | 2 |
| ABEvar3 | Dual | 7.10 | 3 |
| ABEvar4 | Dual | A | 3 |
| ABEvar5 | Single | A | 3 |
| ABEvar6 | Single | B | 3 |
| ABEvar7 | Single | C | 3 |
| ABEvar8 | Single | D | 3 |
| ABEvar9 | Single | E | 3 |
| ABEvar10 | Single | E | 4 |
| ABEvar11 | Single | E | 5 |
| ABEvar12 | Single | F | 5 |



# Overcoming barriers to achieving proof-of-concept

- Base editing correction require models that harbor the disease-causing variant of interest
- *In vitro* models may exist in biorepositories or can be engineered (CRISPR, BE, PE, lentivirus)
- *In vivo* models rarely exist but unique knock-in mouse model can be generated, characterized and used to support an IND-enabling nonclinical data package





GSD Mouse Model

- Mice compound heterozygous for mutation A and B may better reflect patient population and permit study of up to two products in a common model species
- This approach to *in vivo* models is not scalable, and at a certain point may not be necessary?
  - We expect models of the same disease to behave similarly despite having different mutations
  - *In vitro* potency assays could be developed to compare potency of distinct drug products

# Streamlining the Nonclinical Toxicology Package

| | Study | Species | Purpose |
|---|---|---|---|
| **LNP Platform Studies** | Non-GLP Tolerability Studies | NHP and rodent | Non-GLP Tolerability Studies: Initial exploratory toxicity and DRF studies confirming rodent is most sensitive species for LNP safety assessment and establish tolerability across wide dose range |
| | Single dose GLP Toxicology study | Rodent | Single dose GLP Toxicology study: LNP safety in wild-type animal (may or may not contain editable gene) |
| | GLP Cardiovascular safety pharmacology study | NHP | GLP Cardiovascular safety pharmacology study: CV safety assessment for novel lipids |
| | GLP *In vivo* micronucleus study | Rat | GLP *In vivo* micronucleus study: *In vivo* genetox assessment for novel lipids |
| | GLP Ames | *In vitro* | GLP Ames: Mutagenicity assessment for novel lipids |
| | GLP *in vitro* micronucleus assay | *In vitro* | GLP *in vitro* micronucleus assay: Clastogenicity assessment for novel lipids |
| | GLP Progeny and Fertility Study | Rodent | GLP Progeny and Fertility Study: Show lack of germline transmission of edit and fertility assessment |
| **Product Specific Studies** | GLP Pharmacology/Toxicology Study | Disease model | LNP and PD-related safety assessment in pharmacologically-relevant (containing editable gene) model |

# Streamlining Nonclinical Pharmacokinetics and Biodistribution

| | Study | Species | Purpose |
|---|---|---|---|
| LNP Platform Studies | Non-GLP Biodistribution Study | Rat | Tissue and cellular (mRNA and editor protein) distribution of mRNA, novel lipid, and gRNA distribution |
| | Non-GLP PK Study | NHP | Systemic and liver mRNA and lipid PK in higher order species to enable FIH dose prediction modeling |
| Product Specific Studies for some diseases that could impact product uptake and/or clearance | Non-GLP Biodistribution Study | Disease model | Tissue distribution of on-target editing in pharmacologically relevant model and limited, confirmation of mRNA and/or novel lipid distribution |
| | Non-GLP PK Study | Disease model | Systemic and liver mRNA and lipid PK in pharmacologically relevant model to enable FIH dose prediction modeling |

# Off-target Assessments Have Product-Specific Components

| Study | Experimental Assays | What is assessed? |
|---|---|---|
| gRNA-independent off-target editing | RNA-seq<br>scWGS | Editor itself (pairing with a different gRNA should not change result) |
| Genomic rearrangements | UDiTaS | gRNA/editor pair |
| gRNA-dependent off-target editing | ONE-seq<br>Digenome-seq<br>rhAmpSeq | gRNA/editor pair |
| gRNA-dependent off-target risk assessment | RT-qPCR<br>Targeted RNAseq | Individual off-target sites/edits |

# Maintaining product quality while maximizing CMC efficiencies across related drug products

## Manufacturing Process

- Multiple products (variants of same disease or different indication), could be developed using same or highly similar manufacturing processes
- Efficiencies could be gained through qualification and validation of one manufacturing process for multiple products using a platform approach

## Analytical Methods

- Where possible, analytical method development and validation can be assessed to use a platform approach
- Platform Assay Validations
  - Purity
  - Safety
- Product Specific Assays
  - Potency
  - Identity

## Stability

- Products will be evaluated to determine stability indicating methods
- Stability programs will be developed with scientific intent and compliance but maximizing efficiency
- In-use stability may be another area of efficiency

# Clinical considerations, precision medicine via base editing

Opportunity to *treat* rare and ultra rare diseases, correct disease-causing mutation in a targeted, biologically-rationale fashion, enabling genetic outcome approvable endpoints

Potential for large clinical effects that could not be explained by chance variability, enabling a reduction in patient numbers required to demonstrate efficacy and/or potentially eliminating need for placebo cohort

Potential for biomarker serving as basis of accelerated or full approval given correction of disease pathophysiology at it's most proximal and root cause, the DNA mutation

Potential for umbrella trial to treat one disease which can be caused by a variety of mutations with the corresponding based editing therapy

Opportunity to leverage knowledge of components of platform technology to understand dosing, safety, benefit-risk profile and enable more efficient study designs

Opportunity for Integrated Summary of Safety across different platform products to generate a larger safety database

Opportunity to have one long-term follow up study across multiple mutations for one disease

# Leverage synergies to support clinical development package and understanding of safety and dosing

| Drug component | Contribution to potential safety profile | Safety | Study design (stagger) | Clinical pharmacology to inform dosing |
|---|---|---|---|---|
| **LNP (delivery)** | IRR, increase LFTs, immunogenicity | Since same LNP, understanding risks from 1 indication will inform safety monitoring, mitigation and characterization in other indications(s) | Timing of acute toxicity in 1 indication can inform stagger duration for other indications; no need to wait the standard 28d period if acute tox seen within 14d | BioD is determined by delivery method<br>PK from one indication can inform modeling and dose selection.<br>Use safety and PK data from one indication to enable higher starting dose in a different indication; shorter staggering; limit number of dose levels to evaluate |
| **mRNA encoding for base editor** | guide-independent off-targets, immunogenicity | If same mRNA, safety profile from 1 indication can be used to support the other indications(s) | Doesn't contribute to acute toxicity so would not impact stagger | |
| **gRNA directing editor to precise genetic location** | guide-dependent off-targets | Unique gRNA – dependent risks need to be considered separately for different indications | Doesn't contribute to acute toxicity so would not impact stagger | Unique potency; need to generate PD data to understand dose-response or efficacy. |

# Clinical synergies achievable via a master protocol

Same investigators/sites involved (disease-driven)

Same inclusion/exclusion criteria (except for genotype)

Same endpoints for safety (LNP-mediated)

Same endpoints for efficacy (disease-driven)

Same schedule of assessments (disease-driven)

- PK will require methods appropriate to the drug substance(s) that the patient received

# Highlighted challenges

- Base editing therapies have the potential to correct a wide range of mutations that are the cause of monogenic diseases. However, **correcting each mutation typically requires the development of bespoke gene editing reagents.**

- **Developing preclinical in vivo models for every mutation may not be feasible.** Transitioning to in vitro models for additional mutations could facilitate preclinical development once proof-of-concept is established for a disease.

- LNPs with the same base components but different payloads can benefit from leveraging platform nonclinical studies, analytical methods, and manufacturing processes. **Being unable to leverage platform efficiencies significantly raises the development burden to address additional mutations.**

- Significant efficiencies are possible in clinical development, which can help achieve a faster understanding of safety and dosing by leveraging data across LNPs targeting different mutations. **Not leveraging potential efficiencies can significantly lengthen clinical development timelines.**