ProQR®

ANALYST & INVESTOR

AXIOMER® TECHNOLOGY R&D EVENT

March 29, 2023, Virtual

Agenda

1. Welcome and Agenda
Sarah Kiely

2. Strategy overview
Daniel A. de Boer

3. Introduction to ADAR
Gerard Platenburg

4. ADAR RNA editing
Peter Beal, PhD

5. Axiomer® platform overview
Gerard Platenburg

6. IP overview and Partnering strategy
René Beukema

7. Pipeline overview
Gerard Platenburg

8. Summary and Milestones
Daniel A. de Boer

9. Q&A
Daniel A. de Boer
Gerard Platenburg
René Beukema

10. Closing
Daniel A. de Boer

Speakers

Sarah Kiely
VP Investor Relations & Corporate Affairs

Daniel A. de Boer
Founder & CEO

Gerard Platenburg
Chief Scientific Officer

René Beukema
Chief Corporate Development Officer

Peter Beal, PhD
Professor, UC Davis; ProQR Scientific Advisory Board member

Forward-looking statements

This presentation contains forward-looking statements. All statements other than statements of historical fact are forward-looking statements, which are often indicated by terms such as "anticipate," "believe," "could," "estimate," "expect," "goal," "intend," "look forward to", "may," "plan," "potential," "predict," "project," "should," "will," "would" and similar expressions. Such forward-looking statements include, but are not limited to, statements regarding our strategy and future operations, statements regarding the potential of and our plans with respect to our technologies and platforms (including Axiomer®), our preclinical model data, our pipeline targets, our other programs and business operations, our current and planned partnerships and collaborators and the intended benefits thereof, including the collaboration with Lilly and the intended benefits thereof, including the upfront payment, equity investment, and milestone and royalty payments from commercial product sales, if any, from the products covered by the collaboration, as well as the potential of our technologies and product candidates; our updated strategic plans and the intended benefits thereof, our plans to seek strategic partnerships for our ophthalmology assets, and our financial position and cash runway. Forward-looking statements are based on management's beliefs and assumptions and on information available to management only as of the date of this presentation. Our actual results could differ materially from those anticipated in these forward-looking statements for many reasons, including, without limitation, the risks, uncertainties and other factors in our filings made with the Securities and Exchange Commission, including certain sections of our annual report filed on Form 20-F. These risks and uncertainties include, among others, the cost, timing and results of preclinical studies and other development activities by us and our collaborative partners whose operations and activities may be slowed or halted due to shortage and pressure on supply and logistics on the global market; our reliance on contract manufacturers to supply materials for research and development and the risk of supply interruption from a contract manufacturer; the ability to secure, maintain and realize the intended benefits of collaborations with partners, including the collaboration with Lilly; the possible impairment of, inability to obtain, and costs to obtain intellectual property rights; possible safety or efficacy concerns that could emerge as new data are generated in research and development; general business, operational, financial and accounting risks; and risks related to litigation and disputes with third parties. Given these risks, uncertainties and other factors, you should not place undue reliance on these forward-looking statements, and we assume no obligation to update these forward-looking statements, even if new information becomes available in the future, except as required by law.

Strategy overview

Daniel A. de Boer, Chief Executive Officer

ProQR Therapeutics

Overview

Focus on Axiomer®

Exclusively focused on the development of proprietary Axiomer® RNA editing platform across multiple therapeutic areas; initial focus on liver and CNS diseases

Novel Mechanism of Action

Axiomer® was discovered in ProQR labs in 2014 and uses well-proven modality of oligonucleotides to recruit a novel mechanism of action

Validated across multiple genes

Preclinical data demonstrate Axiomer® is broadly validated across multiple genes

ADAR

Axiomer® is ADAR-mediated RNA editing, recruiting endogenous adenosine deaminase acting on RNA (ADAR)

Two pillars underly strategy

ProQR developing wholly owned pipeline with initial targets in liver-originated diseases

Selectively enter into partnerships: initial partnership with Lilly in September 2021, expansion announced December 2022

RNA editing – a new class of medicines

Developing the RNA field

Era of **genetic** medicine

Focusing on developing **next generation approaches** with RNA

Potential to become a **new class** of RNA therapeutics

Developing Axiomer®

Originating from **human** genetics

Inform **Axiomer® RNA editing platform** and **pipeline development**

Axiomer® RNA-editing platform technology

Versatile

- Ability to target multiple organs and a wide range of diseases with numerous applications
- Potential to include protective variants
- Designed to target a variety of RNA species (mRNA, miRNA, lncRNA)

Safety

- No permanent changes
- No irreversible DNA damages and less risk of permanent side effects

High specificity

- Highly targeted therapeutic with potential to minimize off-target effects and reduce the risk of adverse reactions

Transient

- Provide a long-lasting therapeutic effect that does not require frequent dosing
- Potential to target diseases for which permanent changes would be deleterious

No viral vector

- No risk of immunogenicity or capacity limitation due to the vector
- Efficient development and faster production increase the chance to reach market

Endogenous ADARs

- Leverage body's potential to treat disease
- Less risk of off-target effect vs. exogenous ADARs

ADAR: Adenosine deaminase acting on RNA, mRNA: messenger RNA, miRNA: microRNA, lcRNA: long non-coding RNA

Axiomer® leading the field of RNA editing since 2014

1980s
Discovery of the ADAR family

1990s
ADAR1 purified and ADAR gene cloned

2012
ProQR founded

Principle of therapeutic RNA editing using exogenous ADAR
1995

2014
Invention of Axiomer®
First experiments *in vitro* using EONs recruiting natural and endogenously expressed ADARs leading to IP filing

2019
Systematic improvement stability through chemistry

Multiple targets edited using transfection and gymnosis
2017

2022
Editing and functionality in multiple targets *in vivo*

2023
Full modification yielding stable and efficacious molecules

Current status
Axiomer® ProQR pipeline - clinical target selection

2024+
Full integration of optimal design and delivery technologies

Development of a highly specific, safe, and efficacious platform that is capable of efficient *in vivo* editing for therapeutic uses

ADARs: Adenosine deaminases acting on RNA, EONs: Editing oligonucleotides. References: Bass and Weintraub, Cell 48: 607–613, 1987; Rebagliati and Melton, Cell 48: 599–605, 1987; Bass and Weintraub, Cell 55: 1089-1098, 1988; Wagner et al. Proc Natl Acad Sci USA 86: 2647-2651, 1989; Hough and Bass, J Biol Chem 269: 9933–9939, 1994; Kim et al. J Biol Chem 269: 13480–13489, 1994; O'Connell and Keller, Proc Natl Acad Sci USA 91: 10596–10600, 1994; Woolf et al. Proc Natl Acad Sci USA 92: 8298–8302, 1995

Value creation strategy

ProQR will develop its own pipeline and selectively enter into partnerships

ProQR Pipeline

- Build **in-house pipeline** based on Axiomer® RNA editing technology platform
- Initial focus on **liver originated diseases**

Partnerships

- Largely unencumbered platform, ProQR may **selectively enter partnerships**
- **Lilly partnership** with expansion announced December 2022 – total potential value of ~$3.9B

R&D strategy yielding our initial Axiomer® pipeline programs

- Pipeline programs informed by **human genetics**
- **Liver as primary organ**: limited delivery risks and accessibility to RNA therapeutics
- **Preclinical models** available with strong translatability into the clinic
- Timely insight on **safety and target engagement** by conducting early clinical programs on healthy individuals using validated biomarkers
- **Clinical programs** with disease specific endpoints for regulatory approval

ProQR development pipeline

	TARGET	DISCOVERY	NON-CLINICAL	CLINICAL	GUIDANCE	ESTIMATED POPULATION
PROQR PROGRAMS						
CHOLESTATIC DISEASES	**AX-0810** *for* **NTCP**				Entry into clinical trials in late 2024 / early 2025	~ 100K[1]
CARDIOVASCULAR DISEASES	**AX-1412** *for* **B4GALT1**				Entry into clinical trials in late 2024 / early 2025	~ 200M[2]
	AX-1005 *for CVD*					
METABOLIC DISEASES	**AX-2911** *for NASH*					~ 16M
	AX-0601 *for obesity and T2D*					~ 650M
	AX-9115 *for rare metabolic condition*					~ 20K
RARE NEURO DISEASES	**AX-2402** *for neurodegenerative condition*					~ 30K
OTHERS	*Multiple targets in discovery pipeline*					
PARTNERED PROGRAMS						
Lilly	*Initial **5** undisclosed targets*	*Progress undisclosed*				
	*Next **5** undisclosed targets*	*Progress undisclosed*				
	*Up to **5** potential additional targets*					

[1]Approximately 100K people affected with Primary Sclerosing Cholangitis and Biliary Atresia in US and EU5. [2]Approximately 200 million people suffer from too high a level of cholesterol in US and EU5. *SLC10A1* is the gene that encodes for NTCP protein.
CVD: Cardiovascular Diseases, NASH: Nonalcoholic steatohepatitis, T2D: Type 2 Diabetes.
References: Boonstra K, Beuers U, Ponsioen CY. J Hepatol. 2012 May;56(5):1181-1188; Karlsen TH, et al. J Hepatol. 2017 Dec;67(6):1298-1323; Dyson JK, et al. Lancet. 2018 Jun 23;391(10139):2547-2559; Sundaram SS, et al. Liver Transpl. 2017 Jan;23(1):96-109. Raghu VK, et al. Liver Transpl. 2021 May;27(5):711-718; NORD, 2019. Tsao CW, et al. Circulation. 2022;145(8):e153–e639. World Health Organization, World Gastroenterology Organization

Well positioned to advance Axiomer®

Science

- Deep understanding of basic science – ADAR, oligos
- Optimization of editing oligonucleotides (EONs) for therapeutic development

Axiomer® has broad applicability

- Large number of potential therapeutic applications
- *In vivo* POC established in nervous system, liver

Advancing toward the clinic

- Extensive translational and developmental expertise with oligo modality
- AX-0810 and AX-1412 initial pipeline targets

Leading IP position

- Axiomer® is protected by 10 published patent families
- Continuously investing in expanding IP estate

Strategic partnership strategy

- Lilly collaboration
- Selectively form additional partnerships
- Optionality and multiple value creating opportunities

Experienced leadership

- Deep RNA, corporate finance, and business development expertise across Management Team, Supervisory Board, and Scientific Advisory Board

Strong balance sheet

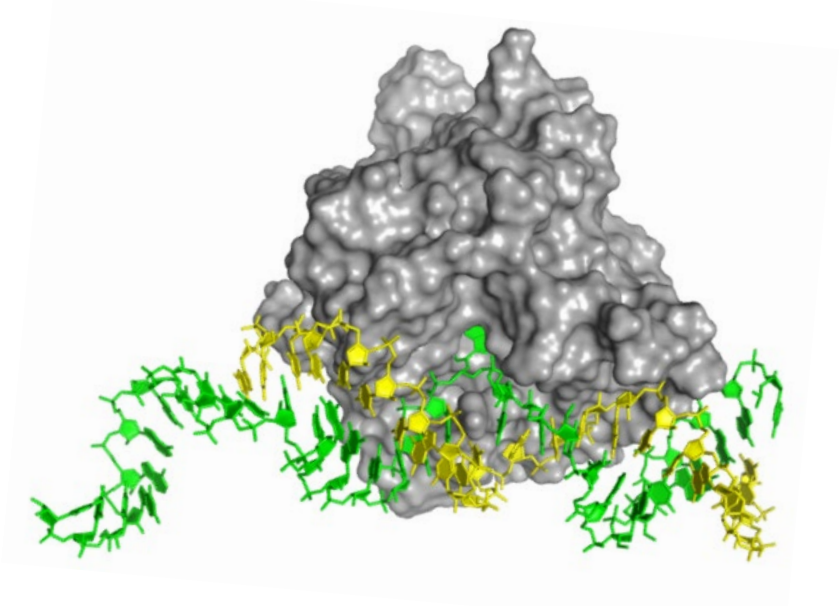
- 2022 YE cash €94.8M, plus $60.0M from Lilly partnership expansion
- Cash runway to mid-2026, excluding potential for additional BD-related upside

Introduction to ADAR

Gerard Platenburg, Chief Scientific Officer

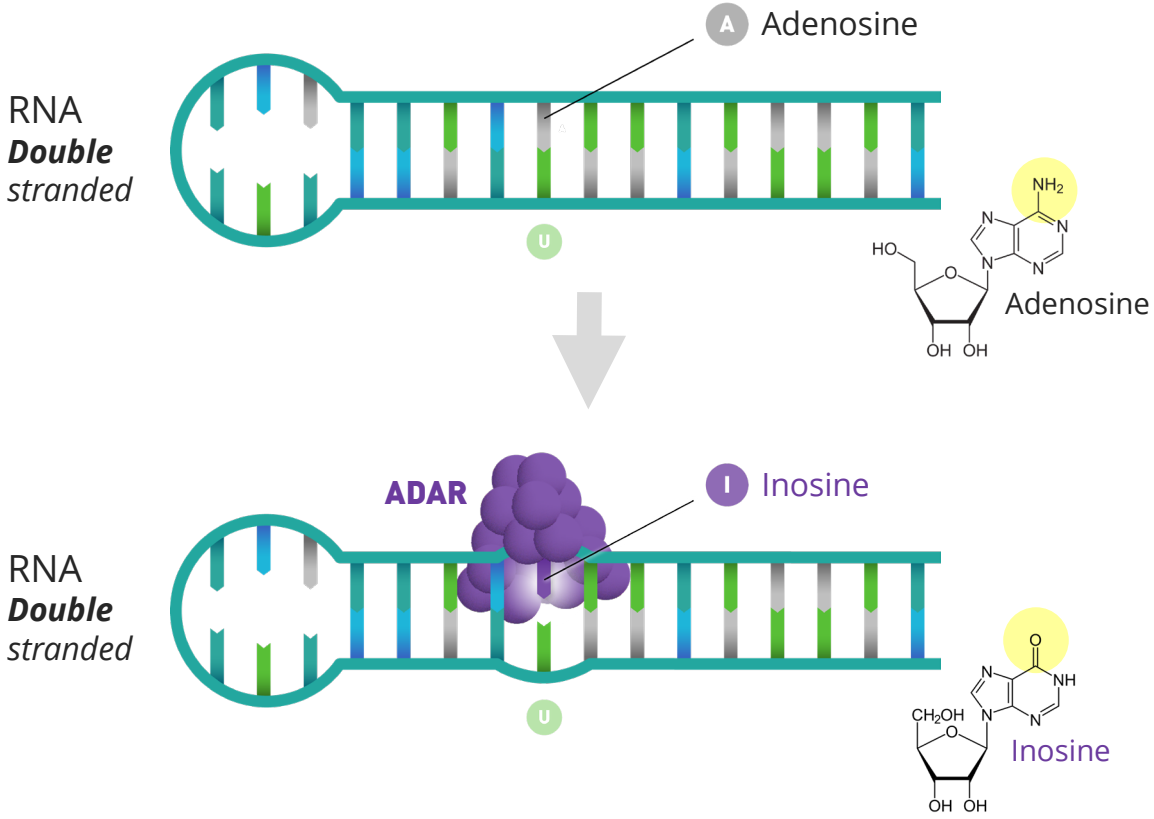
What is ADAR editing?

ADAR *(Adenosine Deaminase Acting on RNA)*



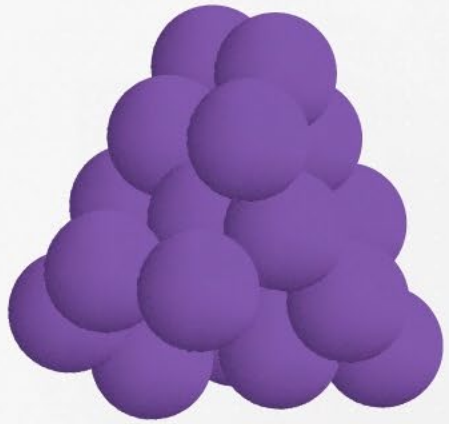
Enzyme that performs specific form of natural RNA editing, called **A-to-I editing.** During A-to-I editing an **A nucleotide (adenosine)** is changed into an **I nucleotide (inosine)**

Natural ADAR editing *(A-to-I)*



HOW DOES ADAR WORK?

Explained in 4 minutes



Peter Beal, PhD

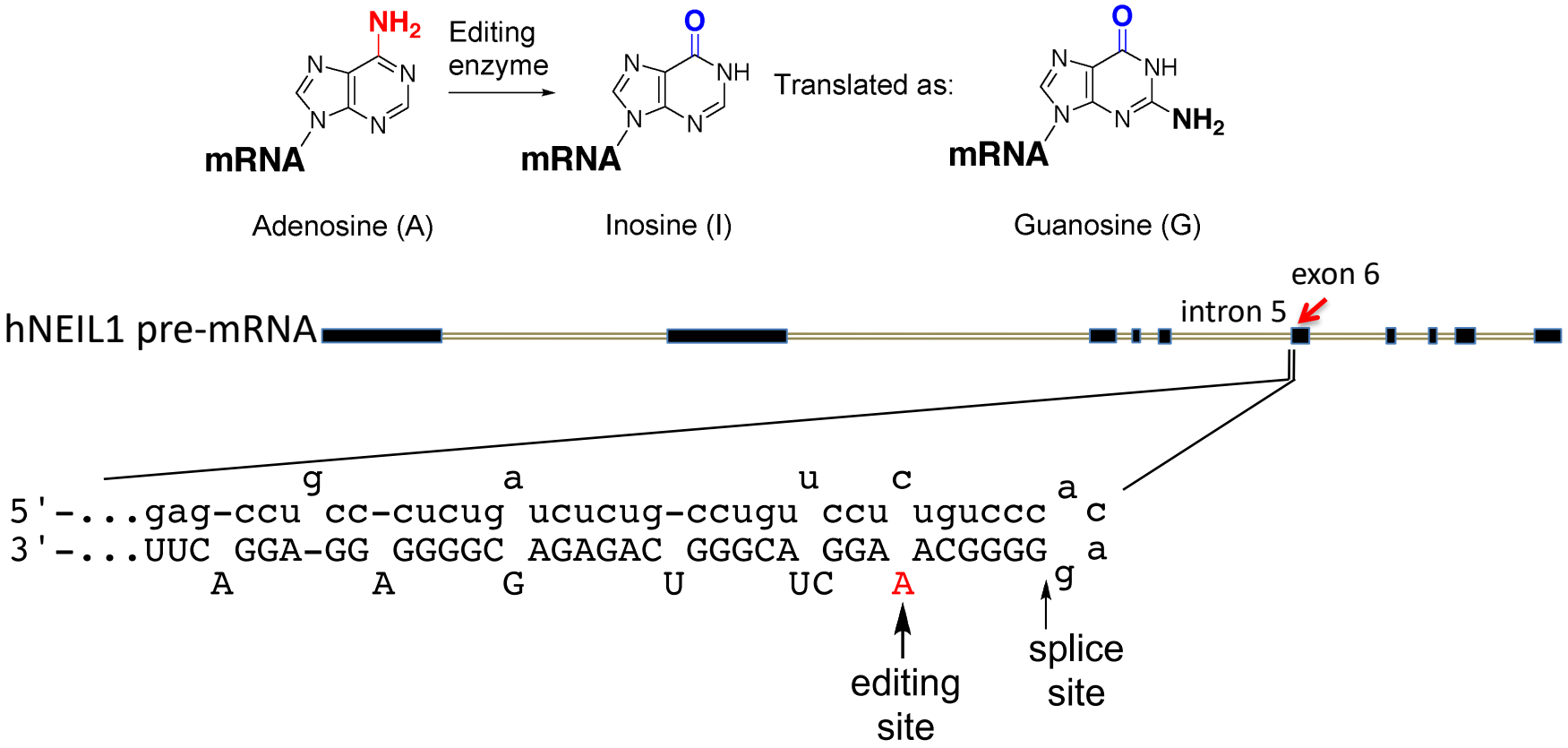
UC Davis, ProQR Scientific Advisory Board member

- Professor in the Department of Chemistry at the University of California at Davis and Director of the NIH-funded UC Davis Chemical Biology Graduate Program
- Advanced understanding of the structures and mechanism of action for the ADAR enzymes responsible for adenosine to inosine RNA editing in humans
- Led in the development of structure-guided methods for optimizing chemically modified oligonucleotides for recruitment of RNA-binding proteins including ADARs
- Teaches organic chemistry at the undergraduate level and several classes in nucleic acids chemistry and chemical biology at the graduate level
- Over 100 peer-reviewed publications in the field of RNA chemical biology and mentored over 50 Ph.D. and M.S. degree students
- Disclosure: ProQR Scientific Advisory Board

RNA editing by the ADARs

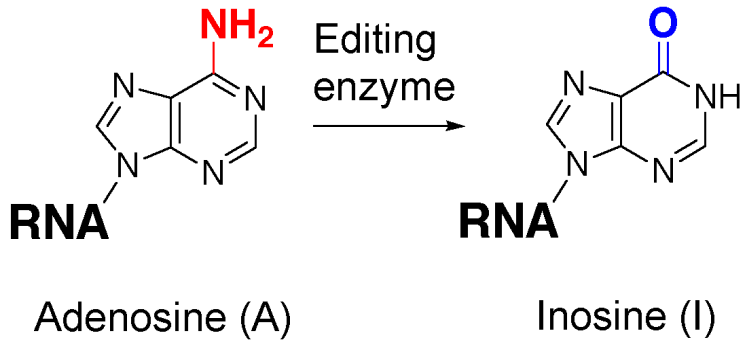
PETER BEAL Department of Chemistry, University of California, Davis

Adenosine to Inosine RNA editing

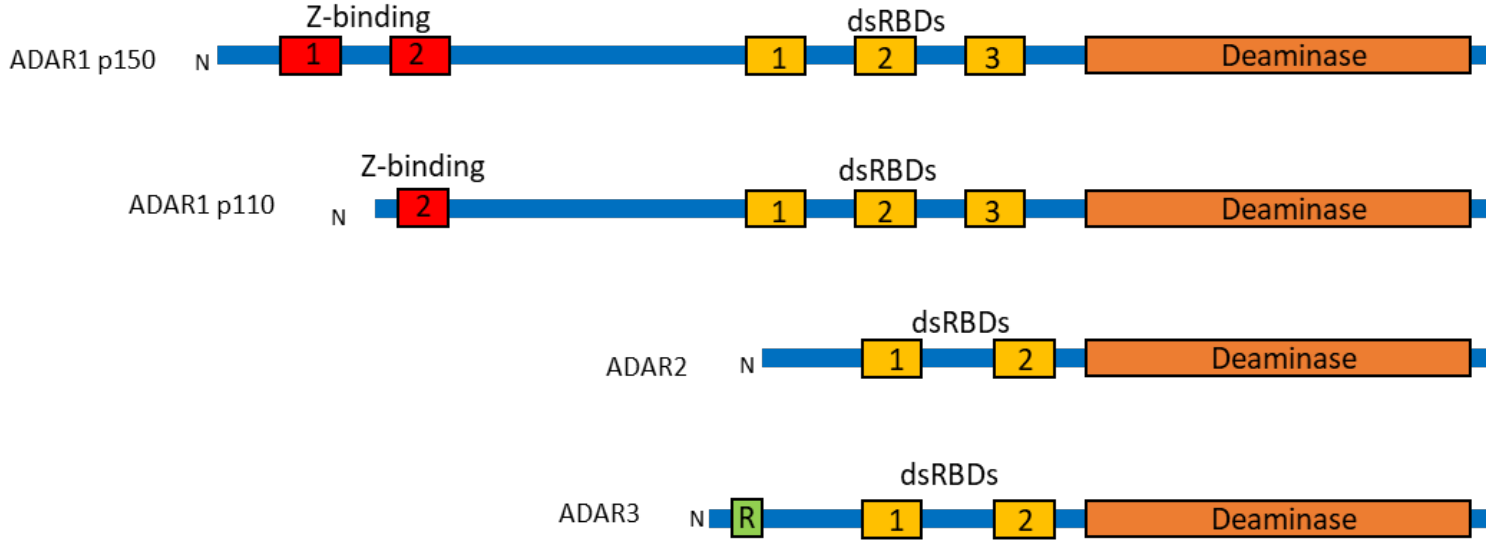


•Yeo, J.; Goodman, R.; Schirle, N.; David, S.S.; Beal, P.A. *Proc. Natl. Acad. Sci. U.S.A.* **2010**, 107, 48, 20715.

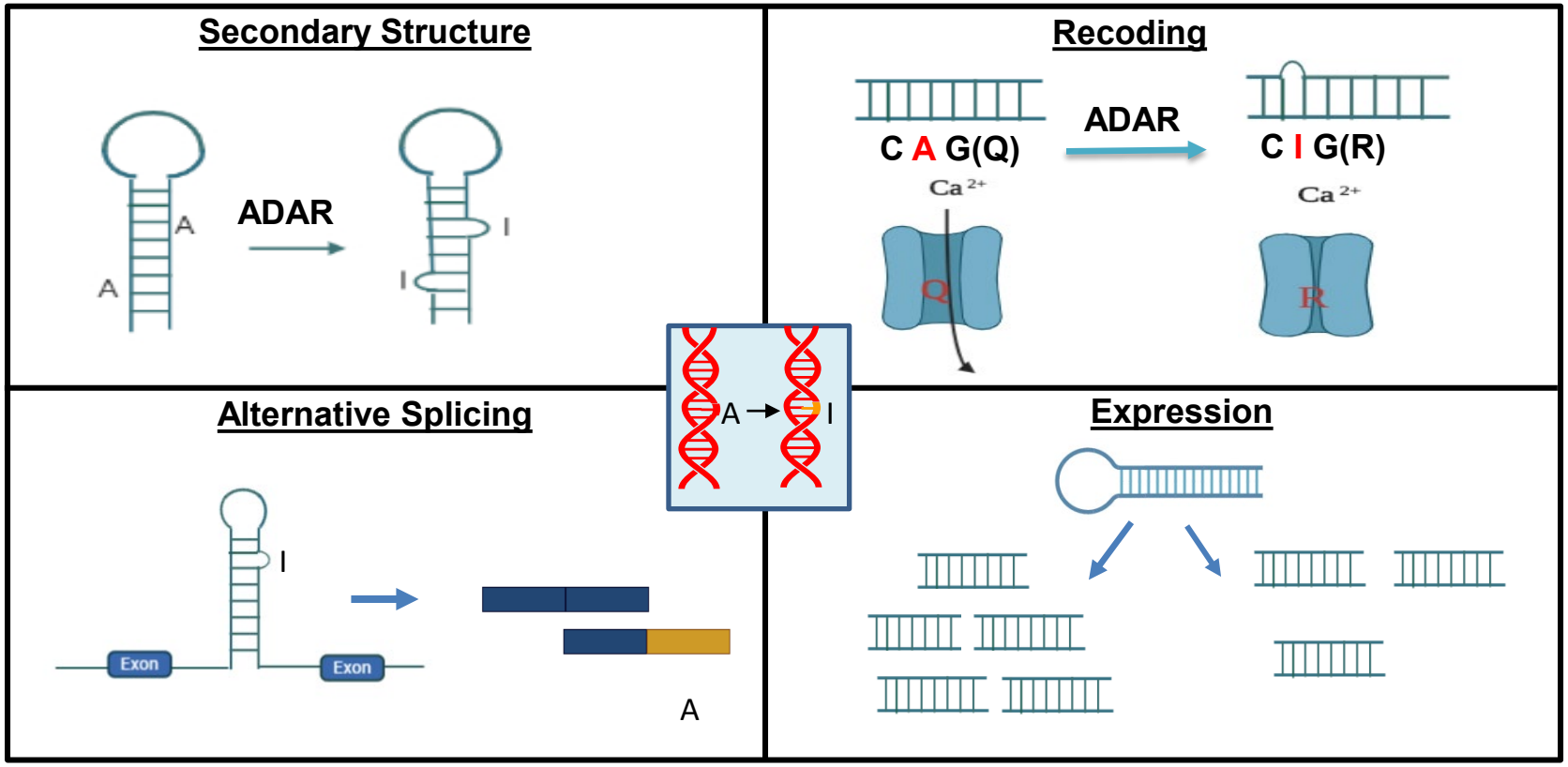
Adenosine to Inosine RNA editing



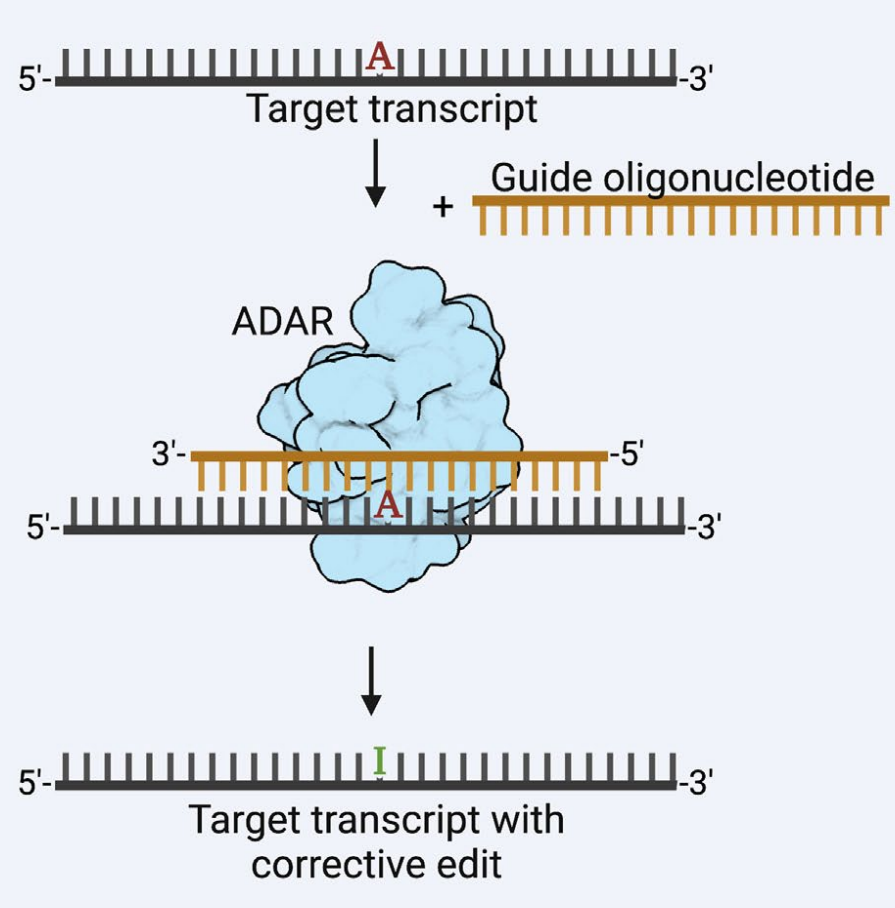
ADARs



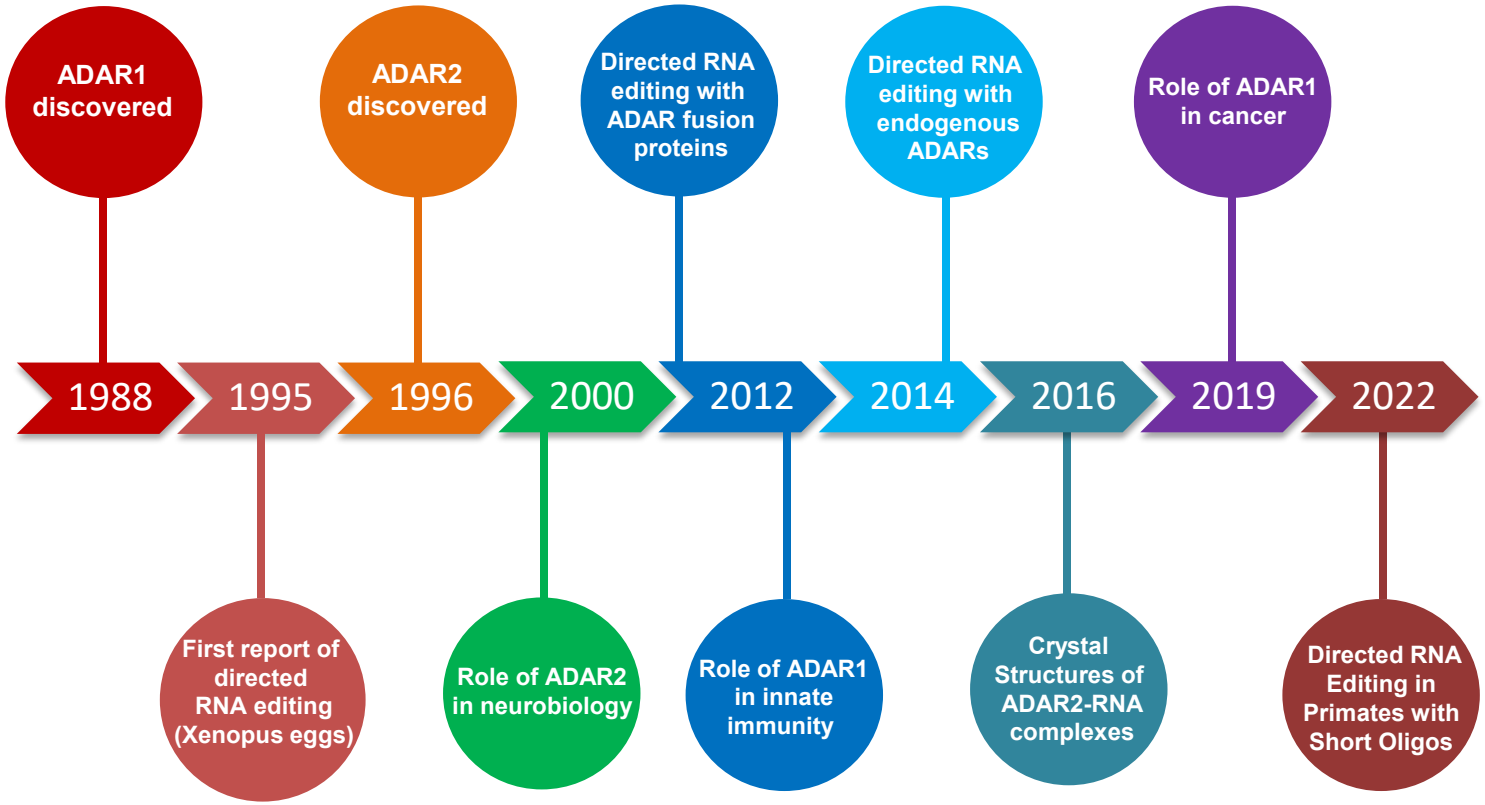
Consequences of RNA Editing by ADARs



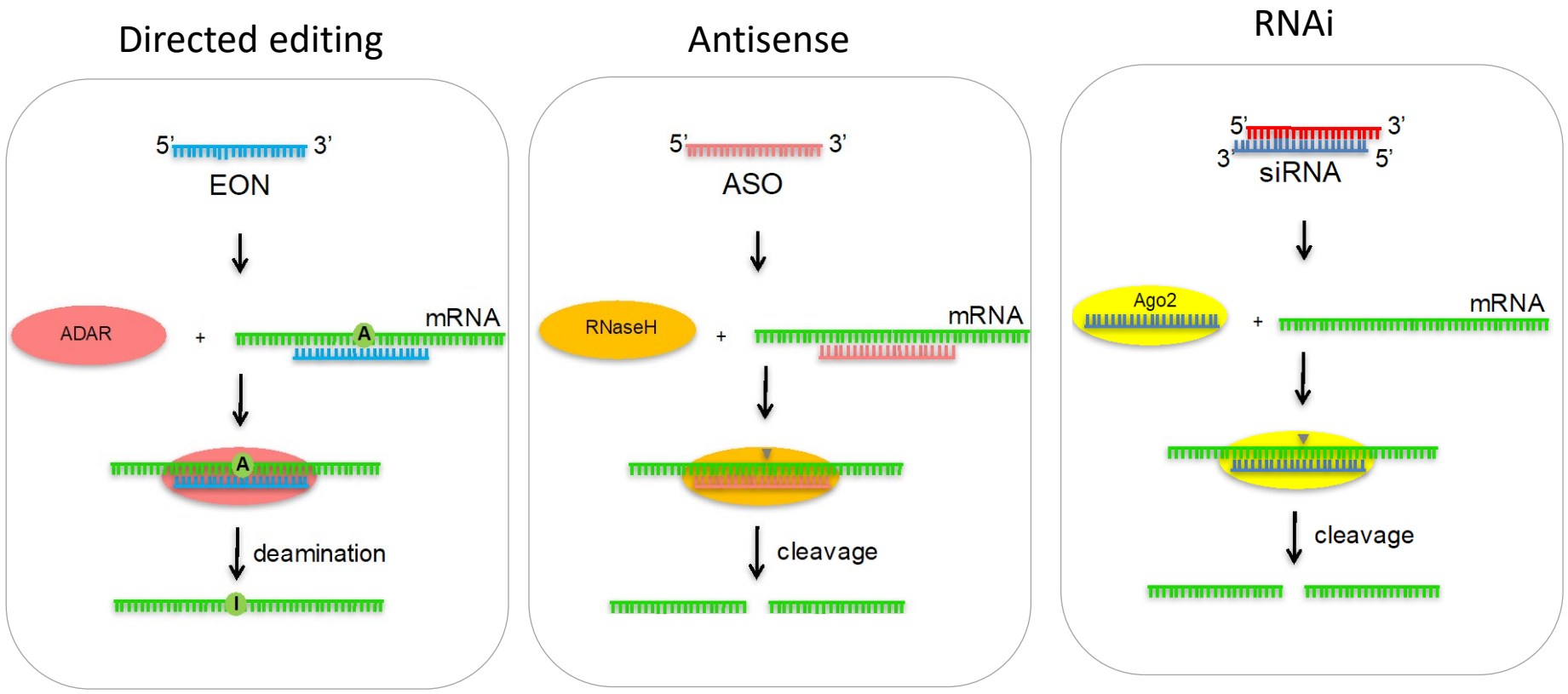
Directed RNA editing



Key Advances in ADAR research (1988-2022)



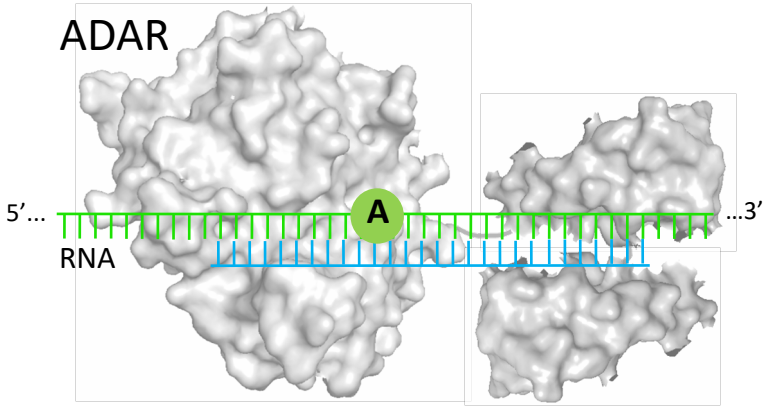
Recruiting human effector proteins by oligonucleotides

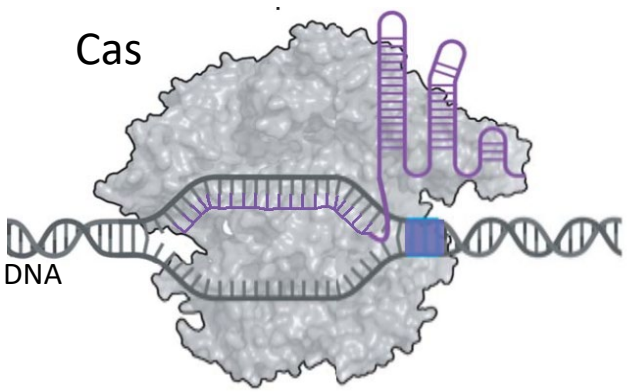


Mipomersen (Kynamro), Inotersen (Tegsedi), Volanesorsen (Waylivra)

Patisiran (Onpattro), Givosiran (Givlaari), Inclisiran (Leqvio), Lumasiran (Oxlumo)

Directed RNA Editing





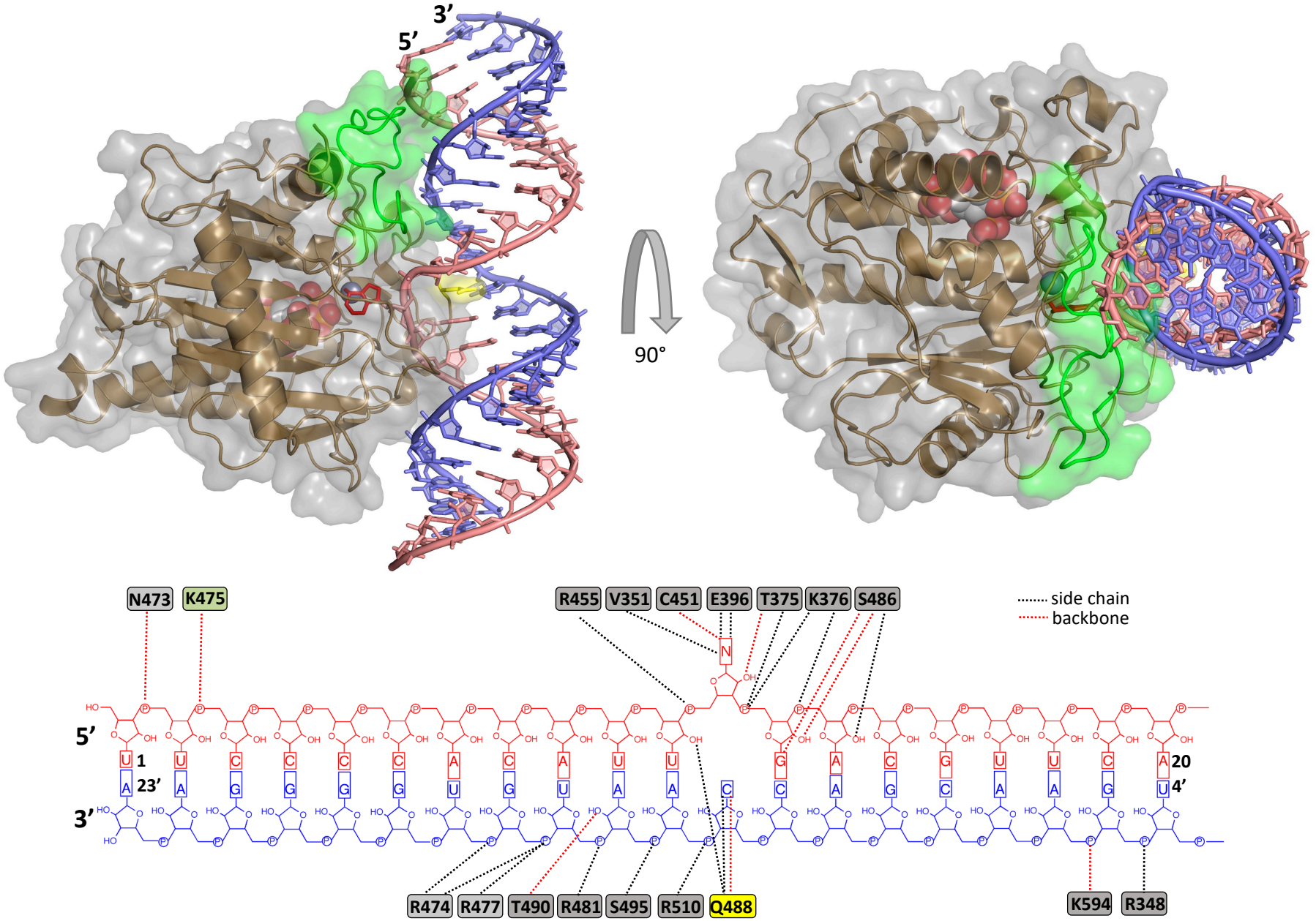
Directed RNA editing vs Genome editing

Advantages

- Effects are reversible
- No need for enzyme delivery
- Human protein vs one of bacterial origin

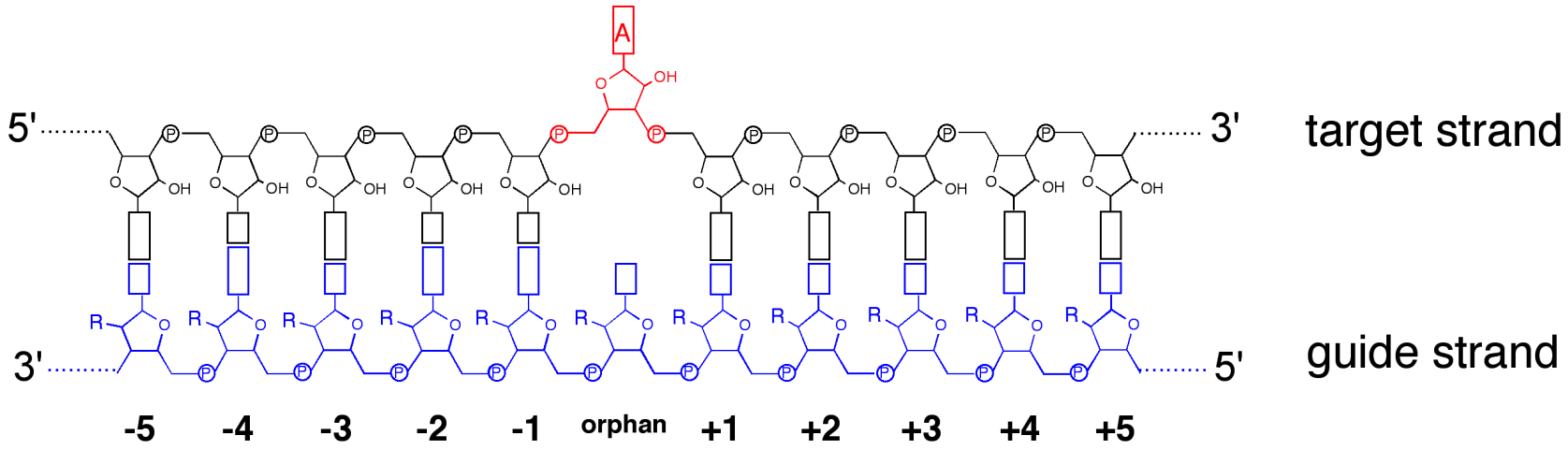
Challenges

- Continuous administration of therapeutic may be necessary
- Low editing efficiency for certain target sequences

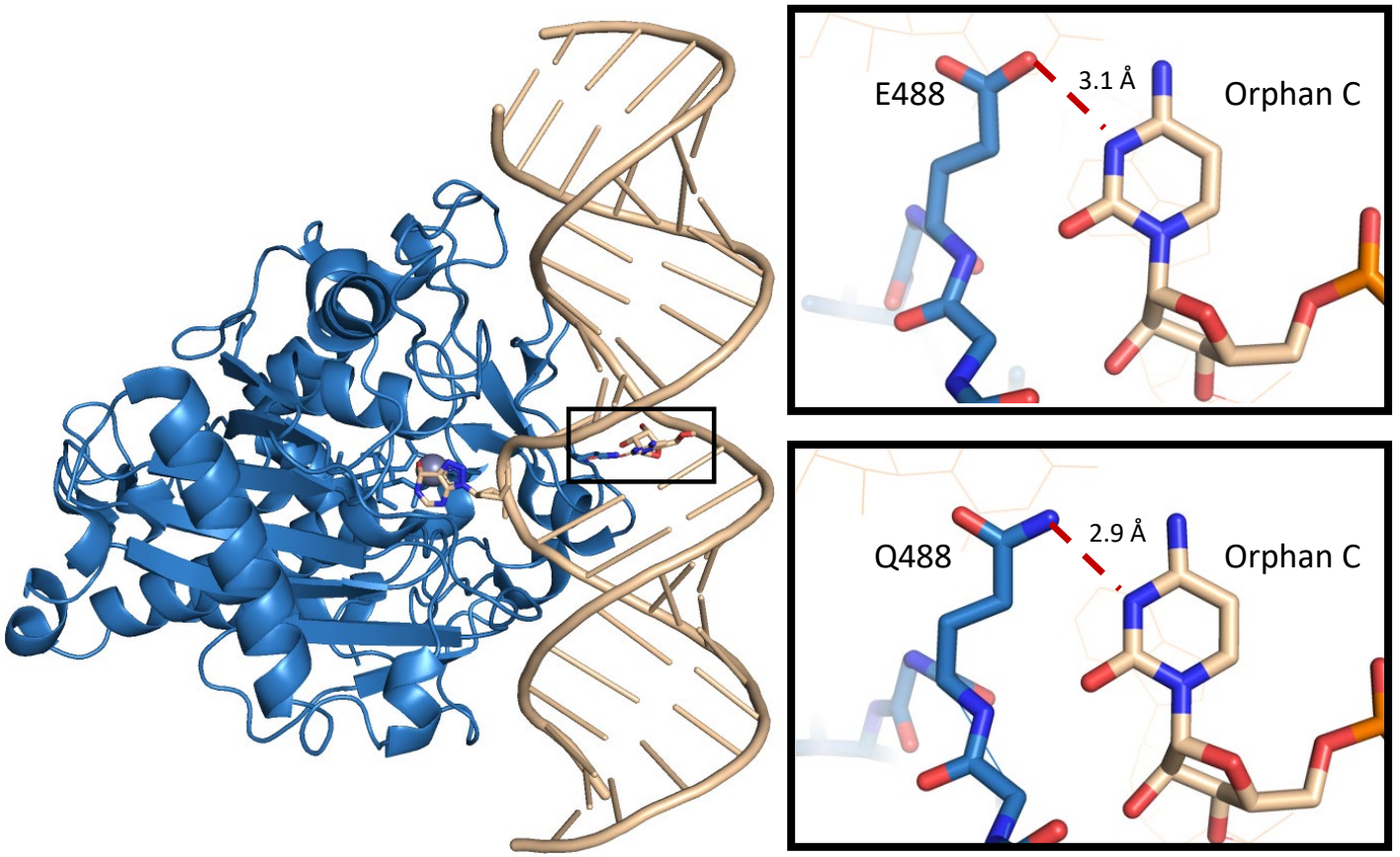


-Matthews, M.M.; Thomas, J.M.; Zheng, Y.; Tran, K.; Phelps, K.J.; Scott, A.I.; Havel, J.; Fisher, A.J.; Beal, P.A. *Nat. Struct. Mol. Biol.* **2016**, 23, 426-433.

Directed RNA editing by ADARs-Guide strand optimization



Orphan base contacts in ADAR2-RNA complexes



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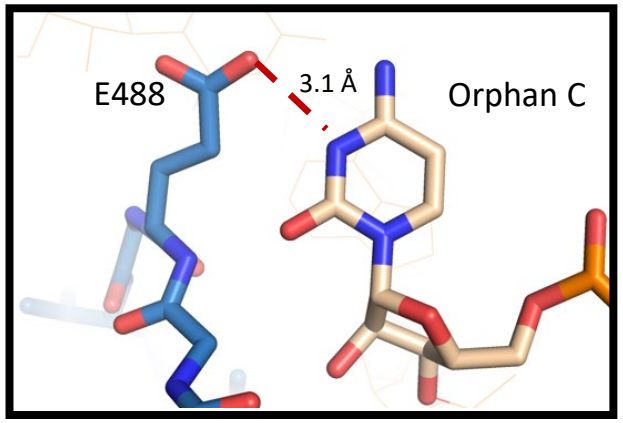
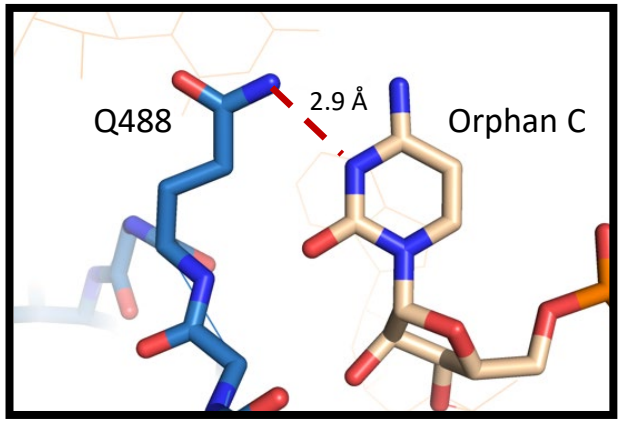
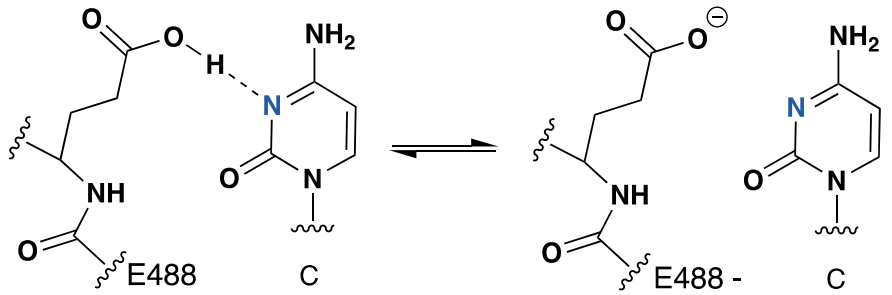
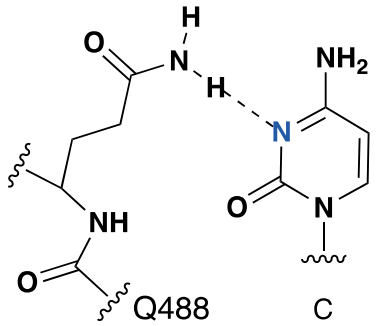




Wild Type-Cytidine
Protonation-Dependent Hydrogen Bonding

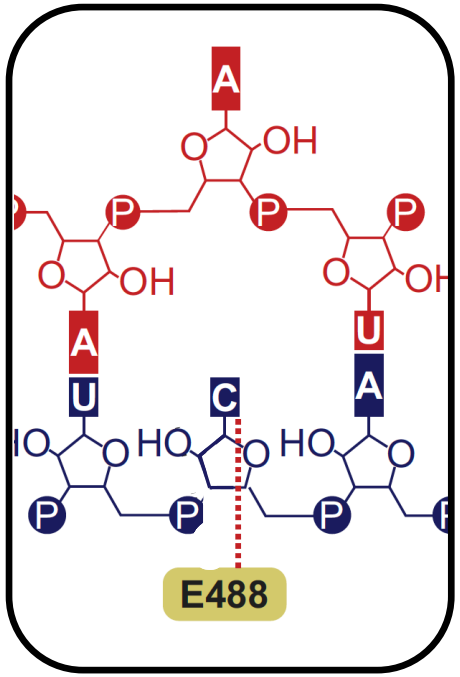


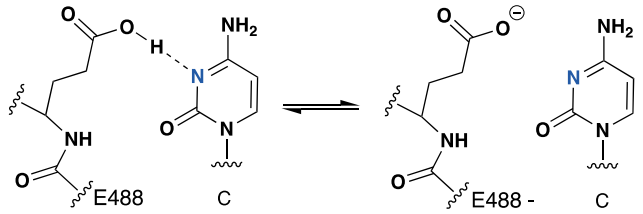
E488Q-cytidine



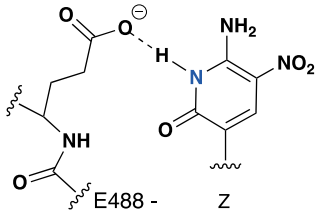
Matthews, M. M. et. al. *Nat. Struct. Mol. Biol.* **2016,** *23* (5), 426-33.

Cytidine analogs to increase editing efficiency

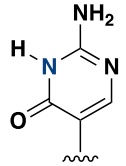




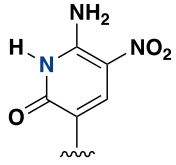
ADAR2 - cytidine



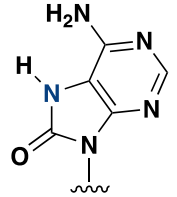
ADAR2 cytidine analog



Pseudoisocytidine
(piC)



Benner's base Z
(Z)

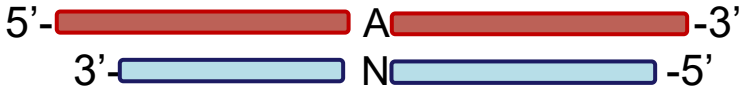


8-oxoadenosine
(8-oxoA)

cytidine analogs

Doherty, E.E.; Wilcox, X.; van Sint Fiet, L.; Kemmel, C.; Turunen, J.; Klein, B.; Tantillo, D.J.; Fisher, A.J..; Beal, P.A. *J. Am. Chem. Soc.* ***2021*** *, 143, 6865-6876.*

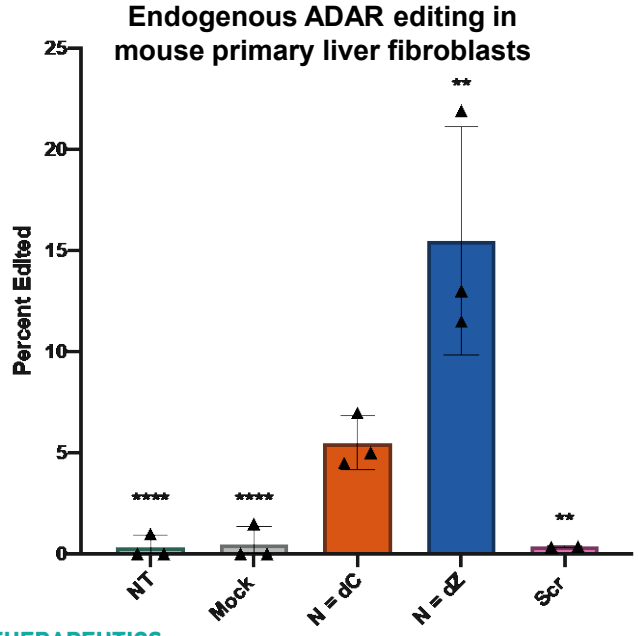
Cellular editing is enhanced by the Z base

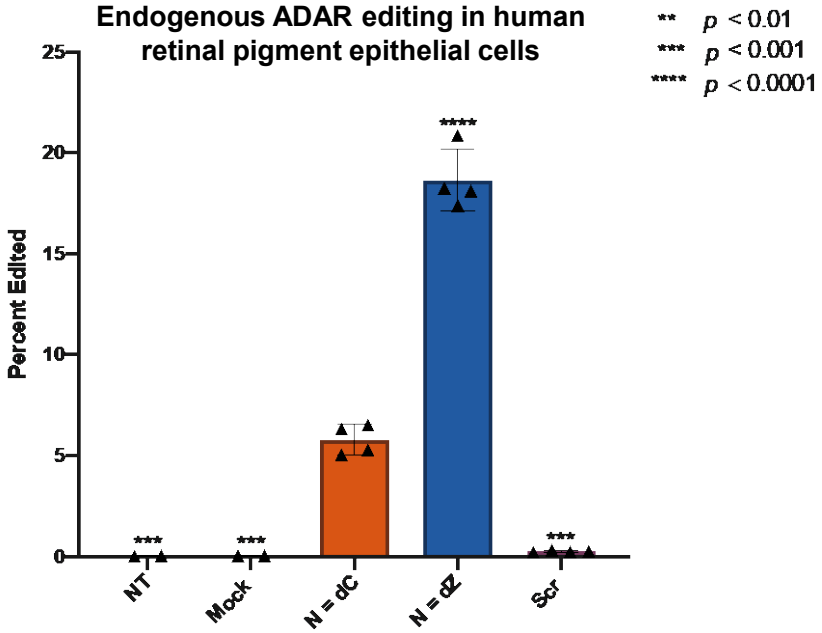


[N] DNA
N 2’-OMe
* PS

5’-G*C*C*C*C*A*GCCUUUGAG*A*C*C*U*CUGUC[NAG]AGUU*G*U*U*C*U-’3

5’-C*A*A*G*G*U*GAUGACGAU*C*A*C*U*GUCG[CNA]UGACA*A*C*A*C*C*G*C -’3





ProQR® THERAPEUTICS

Doherty, E.E.; Wilcox, X.; van Sint Fiet, L.; Kemmel, C.; Turunen, J.; Klein, B.; Tantillo, D.J.; Fisher, A.J..; Beal, P.A. *J. Am. Chem. Soc.* ***2021*** *, 143, 6865-6876.*

ADARs have 5’ nearest neighbor preference

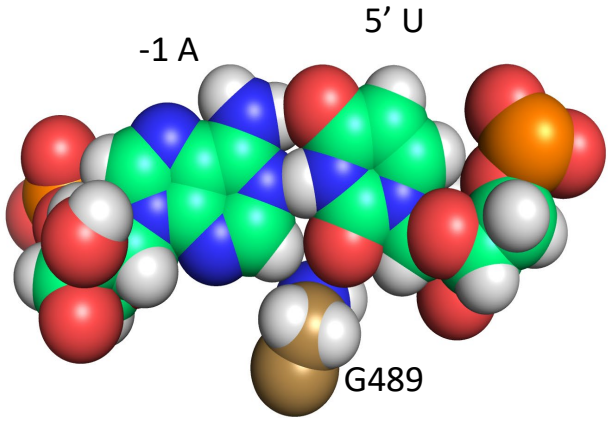
enriched

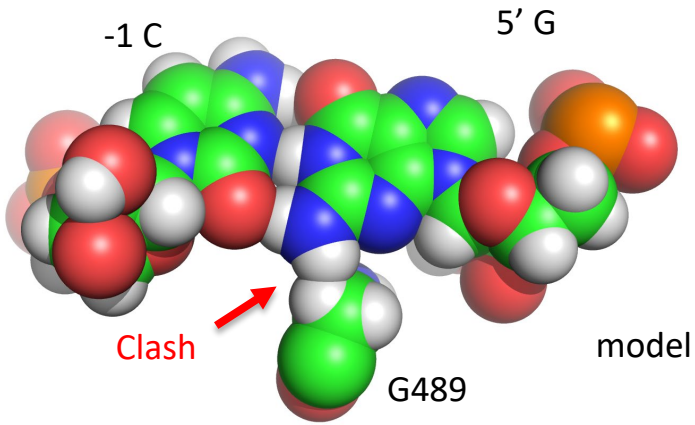
depleted

enriched

depleted

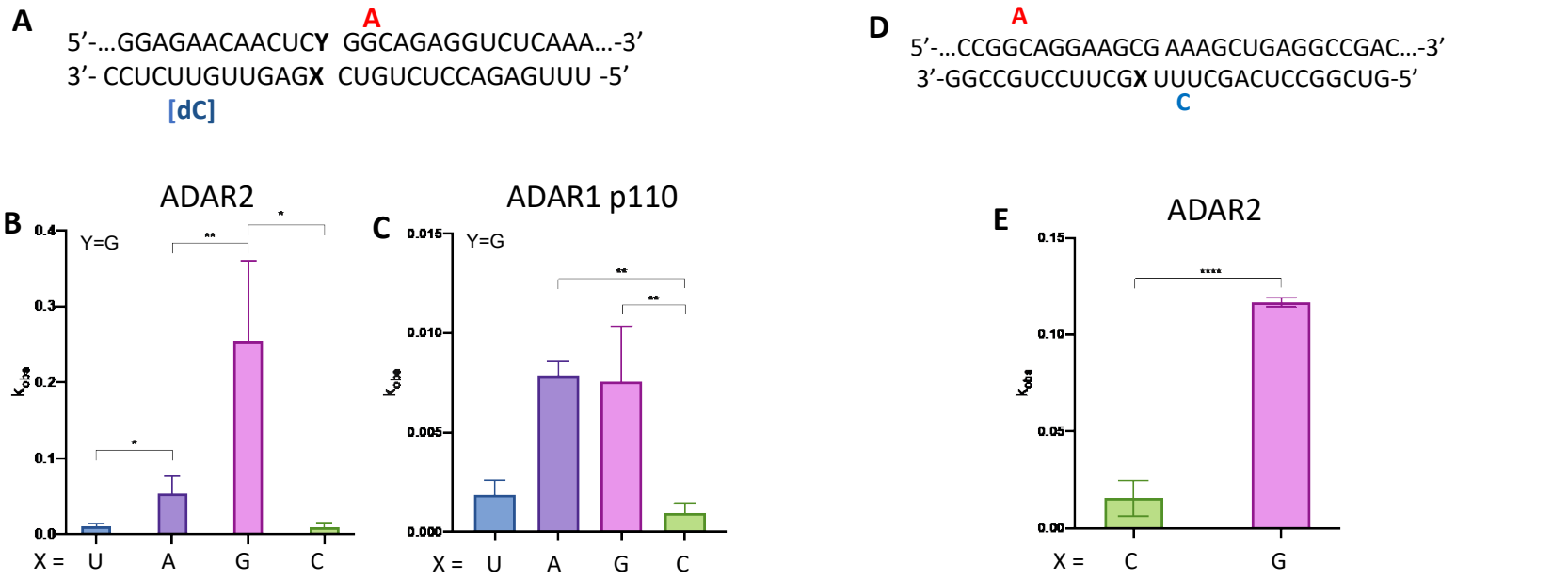
-Eggington, J.M.; Greene, T.; Bass, B.L. *Nat. Comm.* **2011**,2:319.





Nat. Struct. Mol. Biol. **2016**, 23, 426-433

G-G and G-A Pairs Enable Editing for targets with 5’ G



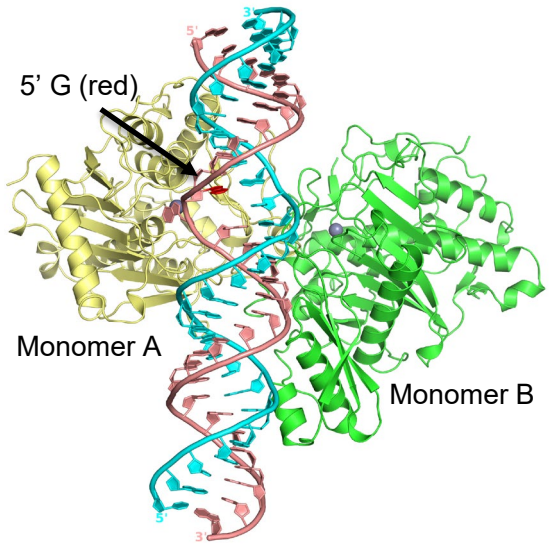
Doherty, E.E.; Karki, A.; Wilcox, X.; Mendoza, H.G.; Manjunath, A.; Jauregui-Matos, V.; Fisher, A.J..; Beal, P.A. *Nucleic Acids Res.* ***2022****, 50, 19, 10857-10868.*

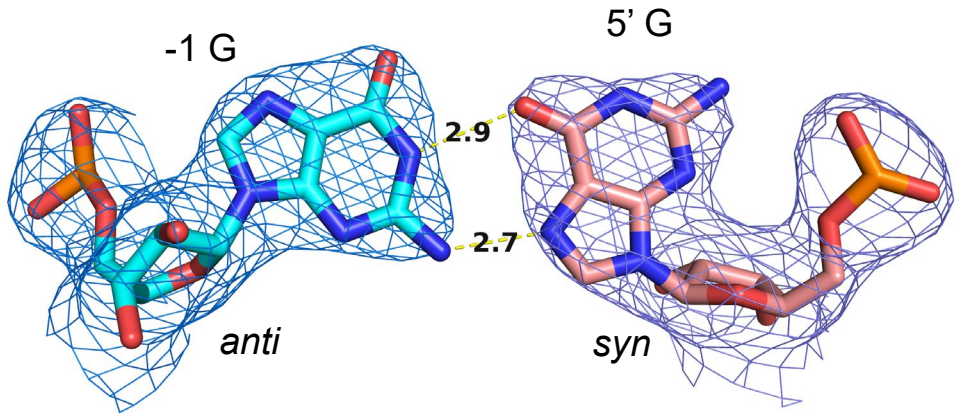
Crystal structure of ADAR-RNA complex shows G-G with a specific pairing geometry

hADAR2 RD

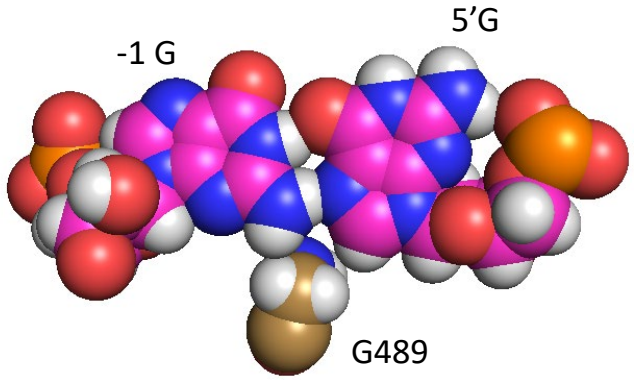
+

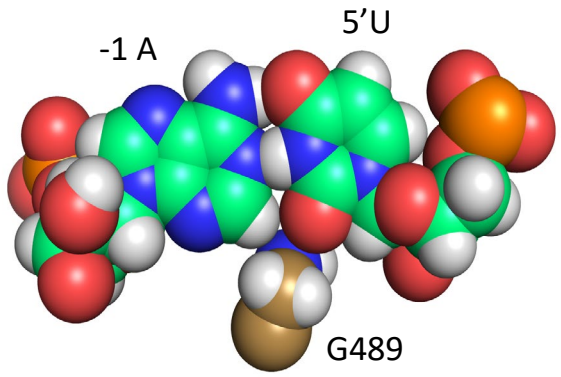
5′–GCUCGCGAUGC G NGAGGGCUCUGAUAGCUACG –3′
3′–CGAGCGCUACG G CCCCCCGAGACUAUCGAUGC –5′

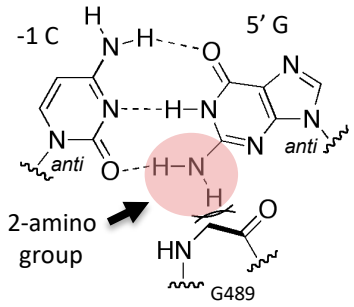


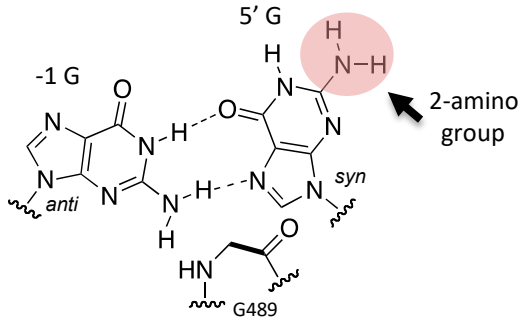


Inducing syn conformation at 5’ G solves steric problem

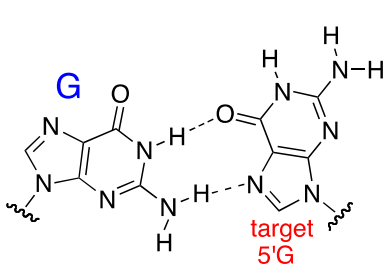


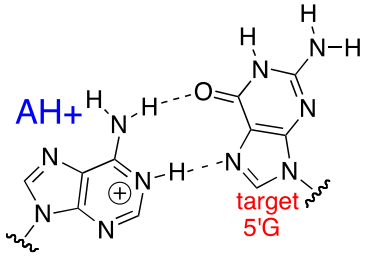


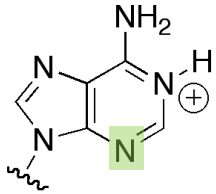




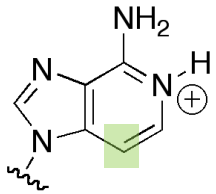
Effects of purine analogs paired with 5’ G







pKa = 3.7

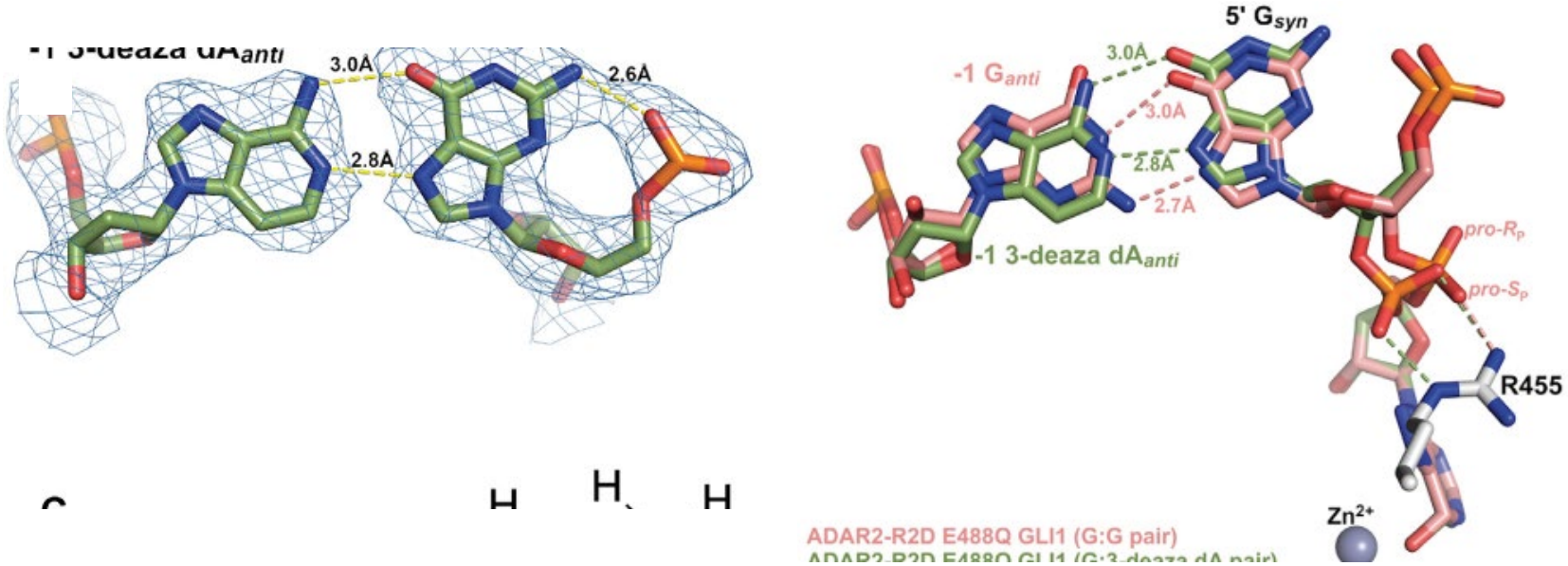


pKa = 6.8

3-deaza dA

Krishnamurthy, R. *Acc. Chem. Res.* **2012**, 45, 12, 2035-2044.

Structure of ADAR-RNA complex with 3-deaza dA:G pair



Doherty, E.E.; Karki, A.; Wilcox, X.; Mendoza, H.G.; Manjunath, A.; Jauregui-Matos, V.; Fisher, A.J..; Beal, P.A. *Nucleic Acids Res.* ***2022****, 50, 19, 10857-10868.*

Summary

- ADAR-mediated RNA editing is capable of rewriting genetic information at the RNA level
- Human ADARs have important roles in neurobiology, innate immunity and cancer
- Synthetic oligonucleotides (EONs) can be used to direct ADARs to make corrective edits
- High-resolution structures of ADAR-RNA complexes, along with rigorous biochemical studies, have enabled optimization of EONs by rational design (e.g. Z at orphan base, 3-deazaA at -1 position)

Beal Group

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Bailey Wong
Sukanya Mozumder
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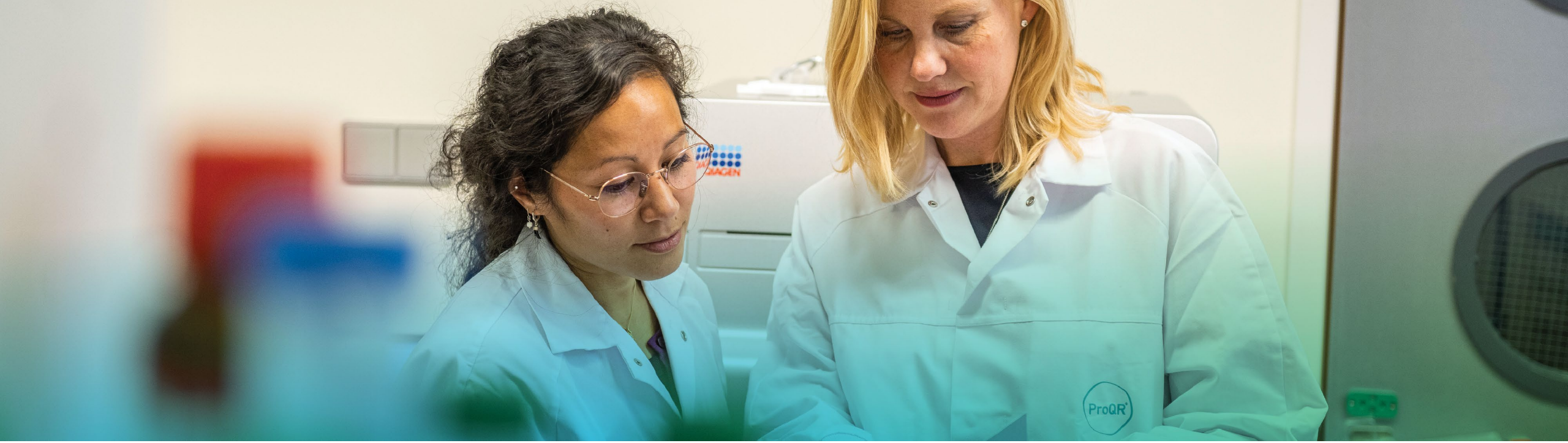
Dr. Lenka van Sint Fiet (ProQR)

NATIONAL INSTITUTES OF HEALTH

ProQR®

rett syndrome research trust



Axiomer® platform overview

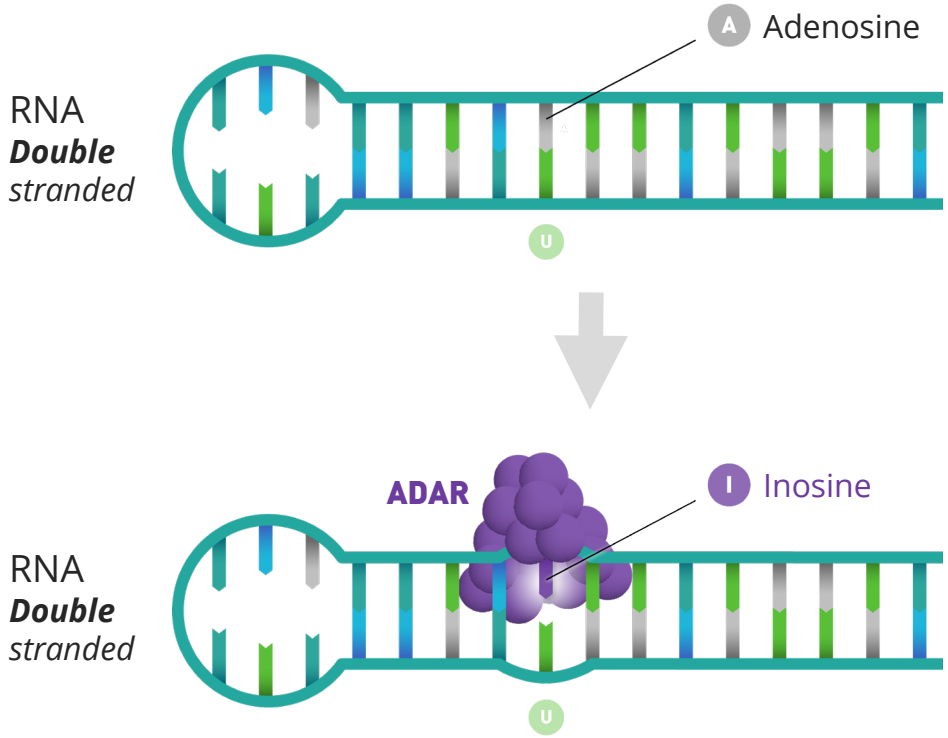
Gerard Platenburg, Chief Scientific Officer

DISCLAIMER: THIS SECTION CONTAINS DATA GENERATED IN HOUSE AND IN COLLABORATION EXPLAINING THE UNDISCLOSED TARGETS IN SOME OF THE FOLLOWING SLIDES

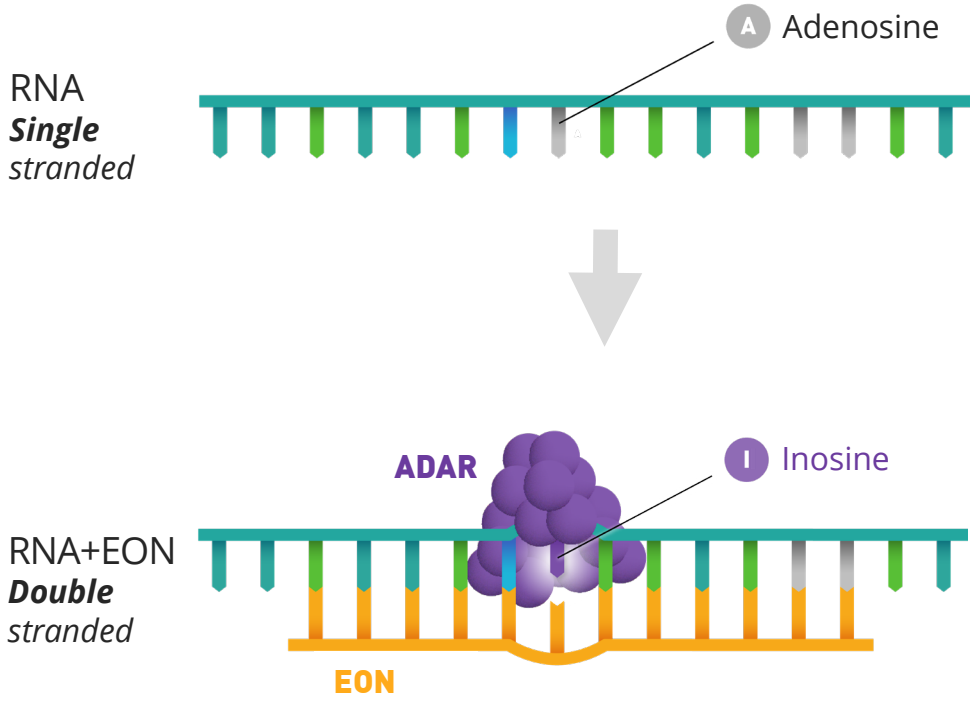
Axiomer® EONs unlock cellular machinery potential to treat diseases

By attracting ADARs and allowing highly specific editing

Natural ADAR editing *(A-to-I)*



Editing Oligonucleotide (EON)-directed therapeutic editing *(A-to-I)*

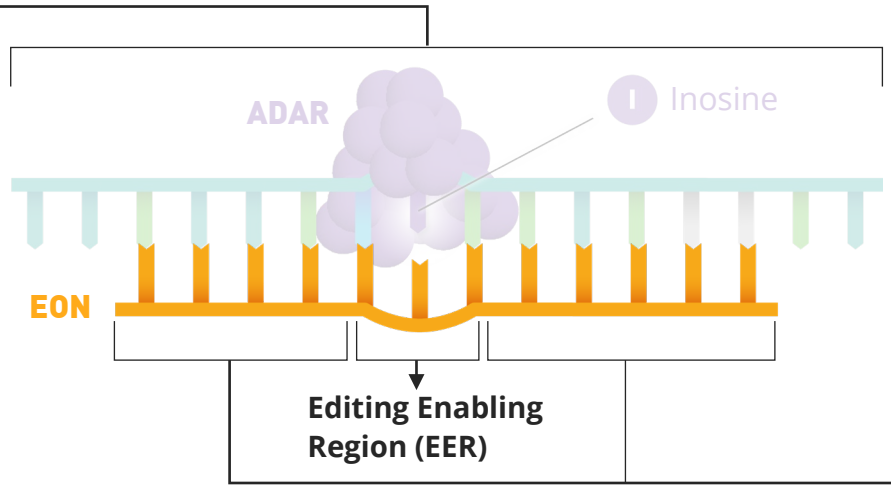


ADARs: Adenosine deaminases acting on RNA.

Driving the development of optimized EONs for therapeutic use

Sequence defines target
RNA binding and provides stability

EON total length
Between ~25 - 30 nt



ADAR-binding region (ABR)
Backbone modifications enable ADAR binding, and disable off-target editing

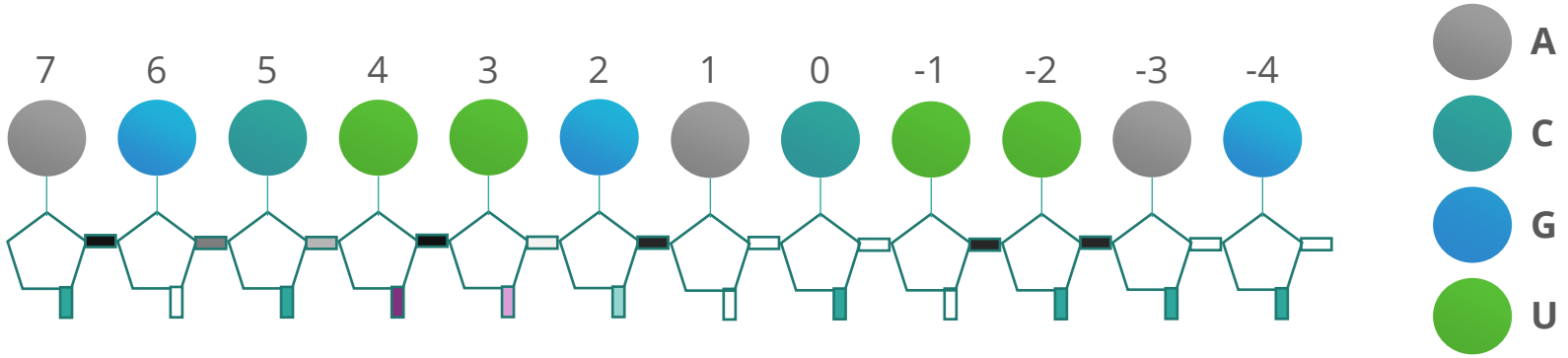
Optimized sequence and chemistry define functionality

- Increase editing efficacy
- Bring metabolic stability
- Prevent off-target ('bystander') editing
- Ensure bioavailability (cell and tissue uptake)
- Offer safety and tolerability at therapeutic doses

ADAR: Adenosine deaminase acting on RNA, EON: Editing oligonucleotide, Nt: nucleotides

Accelerating program advancement with focus on design principles

EON



Aspect	Determined by	Modifications	Effects
Base	Target RNA	Mismatches and analogs	Improved PD
Ribose modification	ADAR structure	2′-H; 2′-OMe; 2′-MOE; 2′-F; 2′-NH2, LNA, TNA, diF, 2′-FANA	Improved PK and PD
Linkage	ADAR structure	PO; PS; PN; MeP; UNA; PAc	Improved PK and PD

This work led to a portfolio of 10 published patent families

ADAR: Adenosine deaminase acting on RNA, EON: Editing oligonucleotide, PD: pharmacodynamic, PK: pharmacokinetic

Axiomer® RNA editing platform has broad potential

Consistent RNA editing

in all models evaluated in nervous system and liver, including NHP *in vivo*

Increased editing efficiency and hepatocyte uptake *in vivo*

GalNAc does not interfere with A-to-I editing and leads to editing increase

Validation of Axiomer's potential for therapeutic targets

With positive effect on protein expression

Broad applicability

With proof of concept in mutation correction and multiple forms of protein modulation

Establishing a strong platform in multiple organs, targets and models

Nervous system

Targeting CNS and PNS

Liver

Targeting liver-originated diseases

Cell models

Organoids

Mice *in vivo*

NHP *in vivo*

Model target

PoC therapeutic targets

Tool targets used for optimization

Pipeline targets

CNS: Central nervous system, NHP: Non-human primate, PNS: peripheral nervous system

Assessing RNA editing across different models and targets in the nervous system

Cell models

Cerebral Organoids

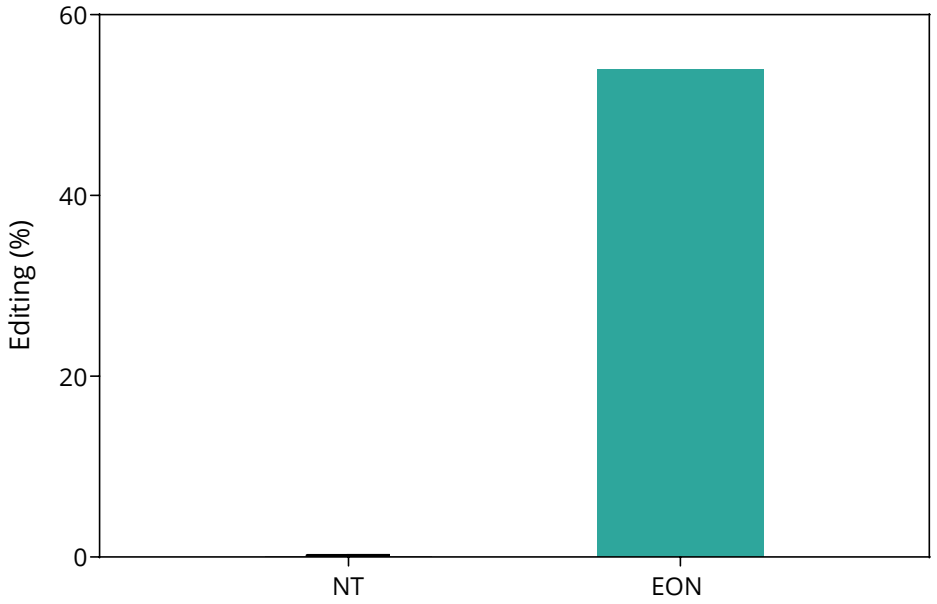
Mice *in vivo*

NHP *in vivo*

More than 50% RNA editing achieved in human iPSC derived neurons

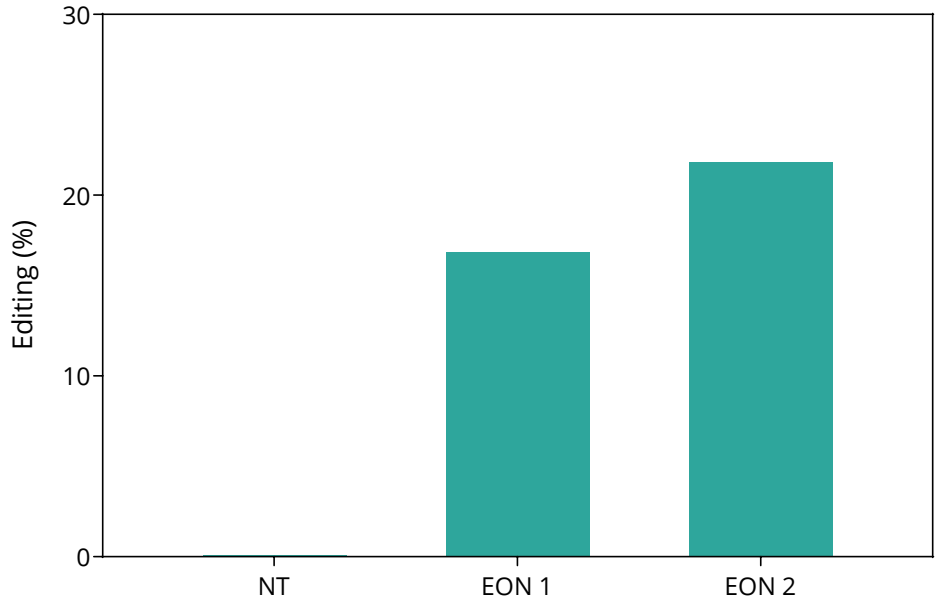
RNA editing of *ACTB*

Gymnosis, 2.5µM, single dose, n=1, 2 weeks, dPCR



RNA editing of *APP*

Gymnosis, 10µM, single dose, washout, n=1, 2 weeks, dPCR

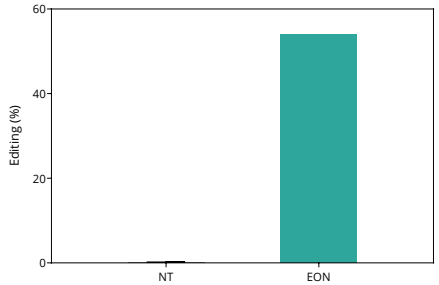


iPSC-derived neurons of 4-6 weeks of neuron maturation

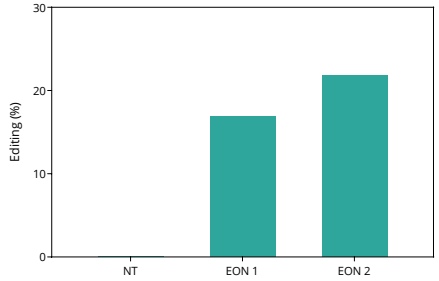
Assessing RNA editing across different models and targets in the nervous system

Cell models

More than 50% RNA editing of *ACTB* in human iPSC derived neurons



More than 20% RNA editing of *APP* in human iPSC derived neurons



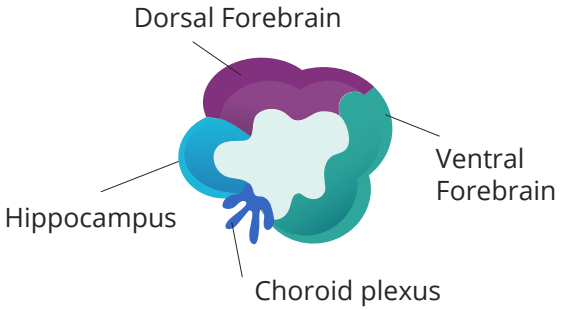
Cerebral organoids

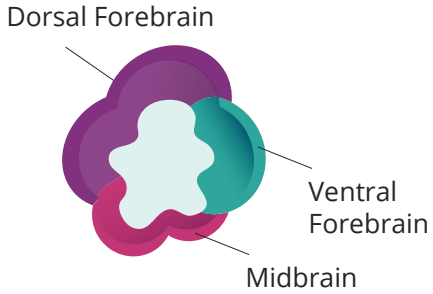
Mice *in vivo*

NHP *in vivo*

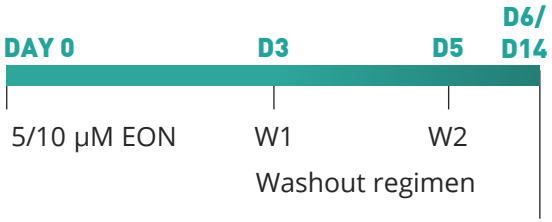
Conditions of the *ACTB* iPSC derived neurons experiment: gymnosis, 2.5µM, single dose, n=1, 2 weeks, dPCR and conditions of the *APP* iPSC derived neurons experiment: gymnosis, 10µM, single dose, washout, n=1, 2 weeks, dPCR.

Up to 65% RNA editing achieved in iPSC derived cerebral organoids





Gymnotic uptake

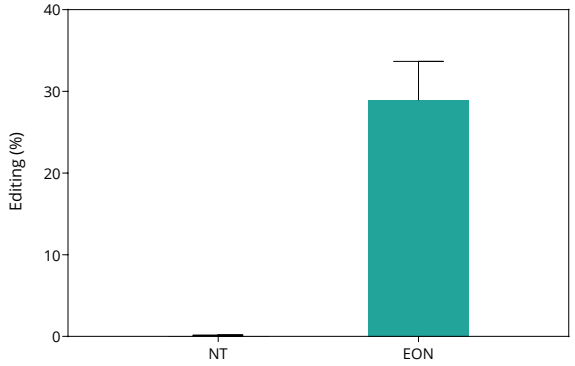


Human cerebral organoids

130-150 days

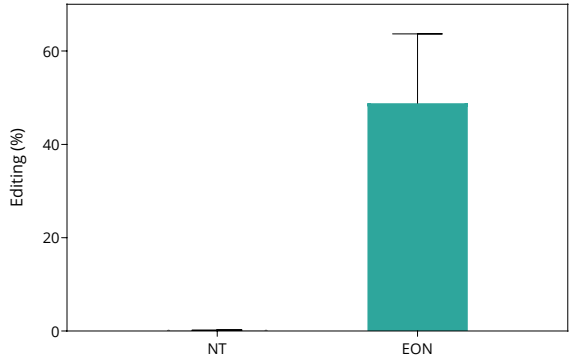
RNA editing of *ACTB*

Gymnosis, 10µM, single dose, washout, n=7, 6 days, ddPCR, mean, SD



RNA editing of *APP*

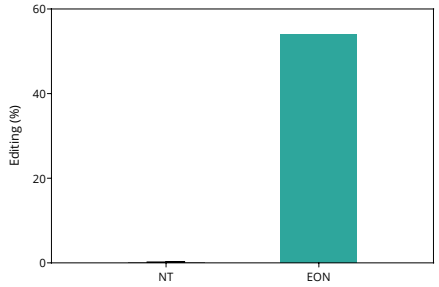
Gymnosis, 5µM, single dose, washout, n=5, 2 weeks, ddPCR, mean, SD



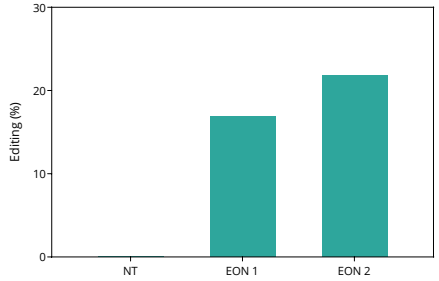
Assessing RNA editing across different models and targets in the nervous system

Cell models

More than 50% RNA editing of *ACTB* in human iPSC derived neurons

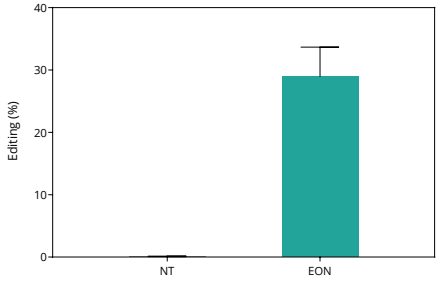


More than 20% RNA editing of *APP* in human iPSC derived neurons

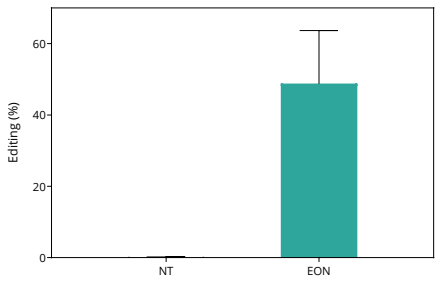


Cerebral organoids

Up to 35% RNA editing of *ACTB* in cerebral organoids



Up to 65% RNA editing of *APP* in cerebral organoids



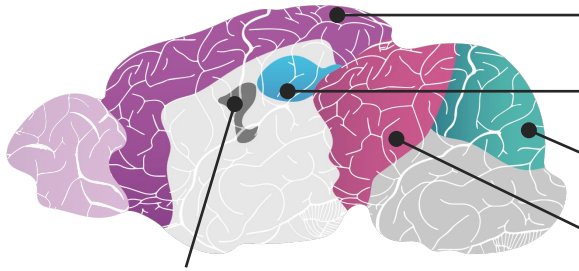
Mice *in vivo*

NHP *in vivo*

Conditions of the *ACTB* iPSC derived neurons experiment: gymnosis, 2.5µM, single dose, n=1, 2 weeks, dPCR and conditions of the *APP* iPSC derived neurons experiment: gymnosis, 10µM, single dose, washout, n=1, 2 weeks, dPCR. Conditions of the *ACTB* cerebral organoids of 130 days: gymnosis, 10µM, single dose, washout, n=7, 6 days, ddPCR, mean, SD and *APP* cerebral organoids of 150 days: gymnosis, 5µM, single dose, washout, n=5, 2 weeks, ddPCR, mean, SD

RNA editing leads to protein function recovery in brain tissues of interest *in vivo*

Mouse brain



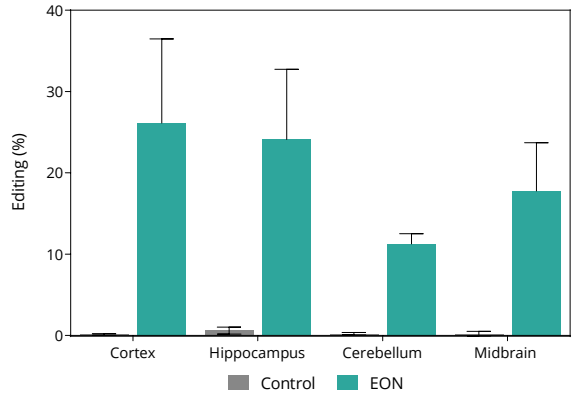
ICV Injection
(lateral ventricle)

	RNA editing (%)	Protein function (Fold change to aCSF)
Cortex	**15-40%**	**26**
Hippocampus	**15-39%**	**6**
Cerebellum	**11-14%**	**13**
Midbrain	**14-25%**	**8**

Up to 40% editing *in vivo* leading to 26-fold change in protein function recovery at 4 weeks with a single dose

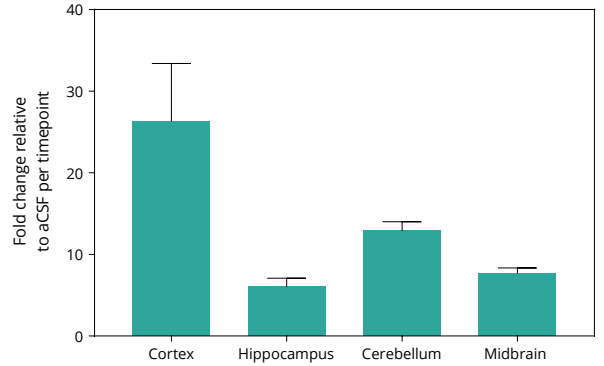
RNA editing in mice brain*

ICV, 250µg, single dose, n=6, 4 weeks, ddPCR, mean, SD



Protein function in mice brain*

ICV, 250µg, single dose, n=6, 4 weeks, western blot, mean, SEM

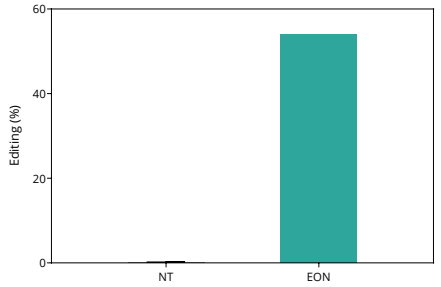


*Undisclosed target. ICV: intracerebroventricular, aCSF: artificial cerebrospinal fluid. Mouse brain (sagittal) from Allen Mouse Brain Atlas

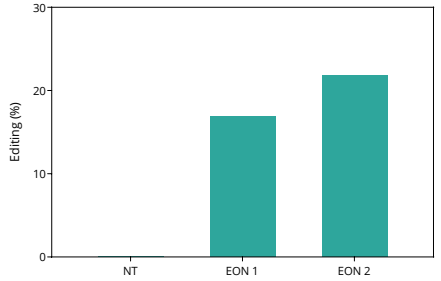
Assessing RNA editing across different models and targets in the nervous system

Cell models

More than 50% RNA editing of *ACTB* in human iPSC derived neurons

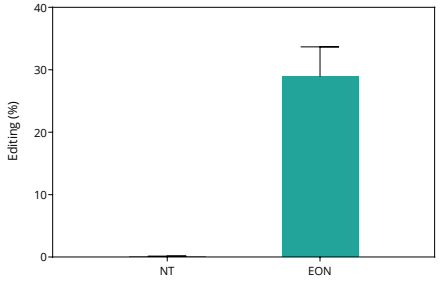


More than 20% RNA editing of *APP* in human iPSC derived neurons

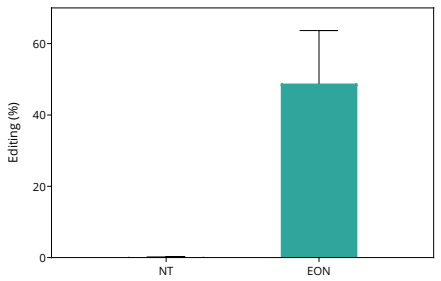


Cerebral organoids

Up to 35% RNA editing of *ACTB* in cerebral organoids

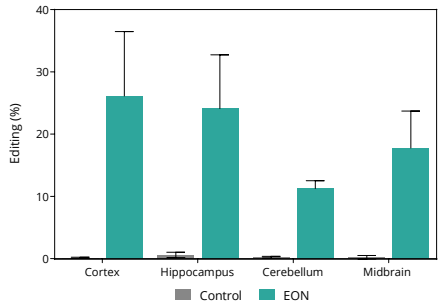


Up to 65% RNA editing of *APP* in cerebral organoids

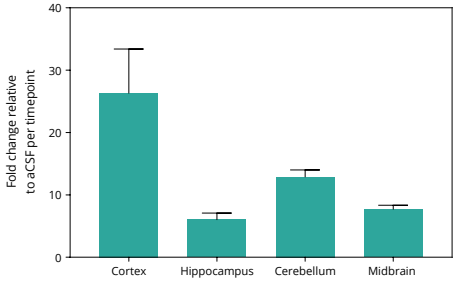


Mice *in vivo*

Up to 40% RNA editing in mice brain*



26-fold change in protein function in mice brain*



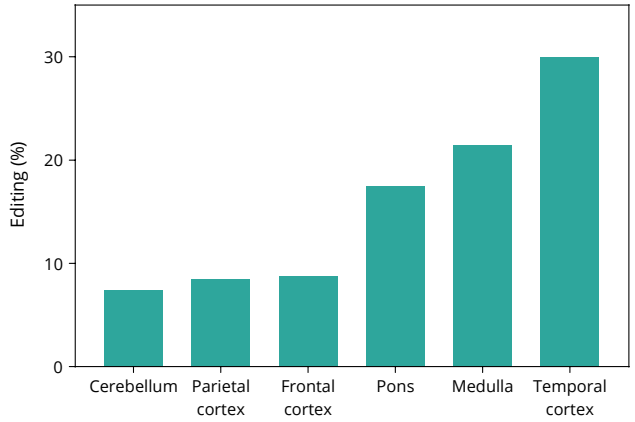
NHP *in vivo*

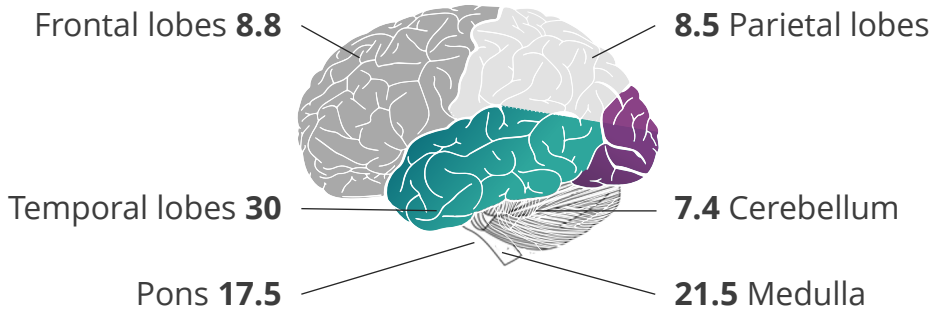
*Undisclosed target. Conditions of the *ACTB* iPSC derived neurons experiment: gymnosis, 2.5µM, single dose, n=1, 2 weeks, dPCR and conditions of the *APP* iPSC derived neurons experiment: gymnosis, 10µM, single dose, washout, n=1, 2 weeks, dPCR. Conditions of the *ACTB* cerebral organoids of 130 days: gymnosis, 10µM, single dose, washout, n=7, 6 days, ddPCR, mean, SD and *APP* cerebral organoids of 150 days: gymnosis, 5µM, single dose, washout, n=5, 2 weeks, ddPCR, mean, SD. Conditions of the mice *in vivo* experiment: intracerebroventricular (ICV), 250µg, single dose, n=6, 4 weeks, editing: ddPCR and protein function: western blot, mean, SD and SEM

Up to 30% RNA editing reported in brain and approx. 50% in spinal cord in NHP *in vivo*

RNA editing *in vivo* in NHP brain*

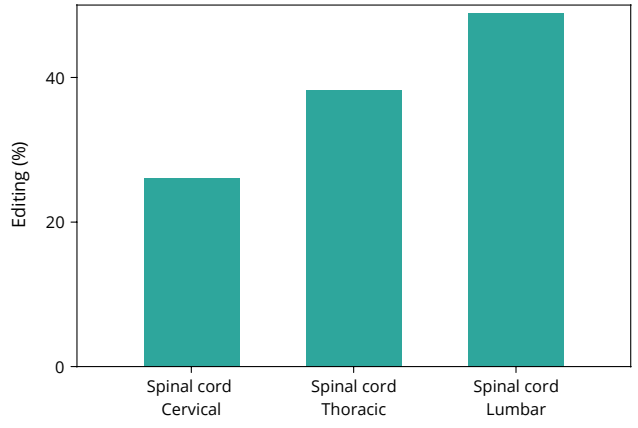
*IT administration, 12mg, single dose, n=3**, 7 days, ddPCR*

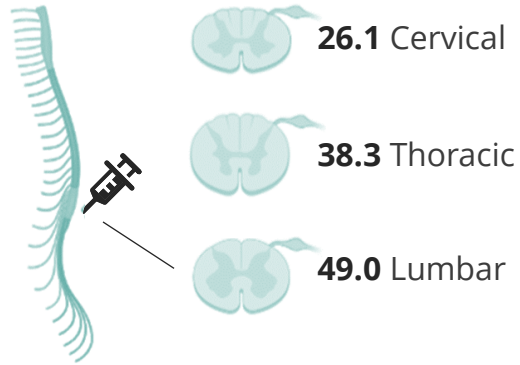




RNA editing *in vivo* in NHP spinal cord*

*IT administration, 12mg, single dose, n=3**, 7 days, ddPCR*



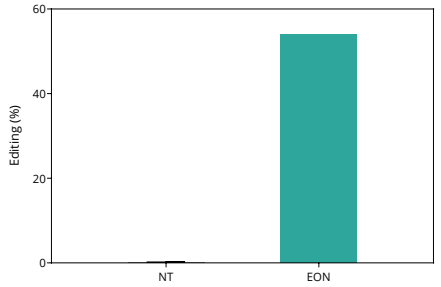


*Undisclosed target. **Data of 2 NHPs not analyzable due to human error during injection procedure. IT: intrathecal, NHP: non-human primate

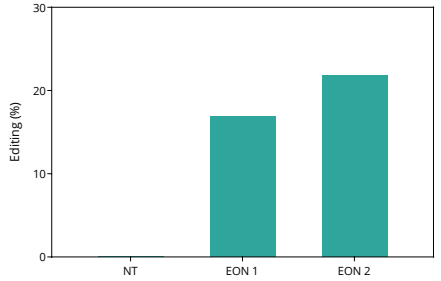
Consistent editing reported - including *in vivo* NHP - in the nervous system

Cell models

More than 50% RNA editing of *ACTB* in human iPSC derived neurons

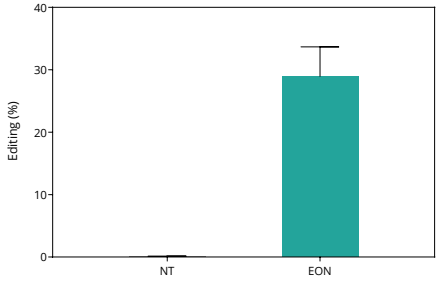


More than 20% RNA editing of *APP* in human iPSC derived neurons

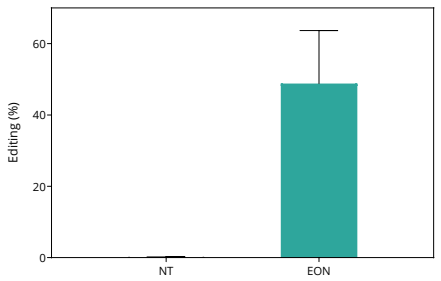


Cerebral organoids

Up to 35% RNA editing of *ACTB* in cerebral organoids

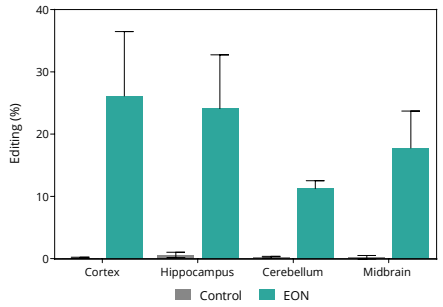


Up to 65% RNA editing of *APP* in cerebral organoids

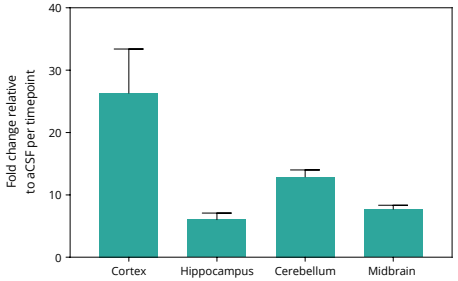


Mice *in vivo*

Up to 40% RNA editing in mice brain*

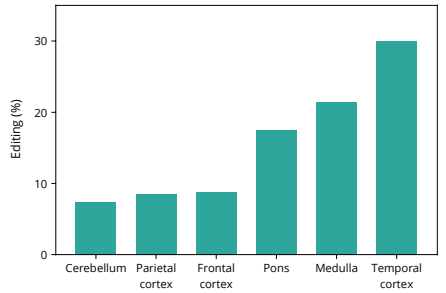


26-fold change in protein function in mice brain*

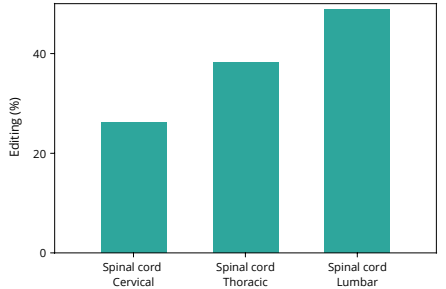


NHP *in vivo*

Up to 30% RNA editing in NHP brain*



Approx. 50% RNA editing in NHP spinal cord*



*Undisclosed target. Conditions of the *ACTB* iPSC derived neurons experiment: gymnosis, 2.5µM, single dose, n=1, 2 weeks, dPCR and conditions of the *APP* iPSC derived neurons experiment: gymnosis, 10µM, single dose, washout, n=1, 2 weeks, dPCR. Conditions of the *ACTB* cerebral organoids of 130 days: gymnosis, 10µM, single dose, washout, n=7, 6 days, ddPCR, mean, SD and *APP* cerebral organoids of 150 days: gymnosis, 5µM, single dose, washout, n=5, 2 weeks, ddPCR, mean, SD. Conditions of the mice *in vivo* experiment: intracerebroventricular (ICV), 250µg, single dose, N=6, 4 weeks, editing: ddPCR and protein function: western blot, mean, SD and SEM. Conditions of the non-human primate (NHP) *in vivo* experiment: intrathecal (IT), 12mg, single dose, n=3**, 7 days. ** Data of 2 NHPs not analyzable due to human error during injection procedure.

Establishing a strong platform in multiple organs, targets and models

Nervous system

Targeting CNS and PNS

Liver

Targeting liver originated diseases

Cell models

Organoids

Mice *in vivo*

CNS: Central nervous system, PNS: peripheral nervous system

Advancing Axiomer® development across different models and targets in the liver

Cell models

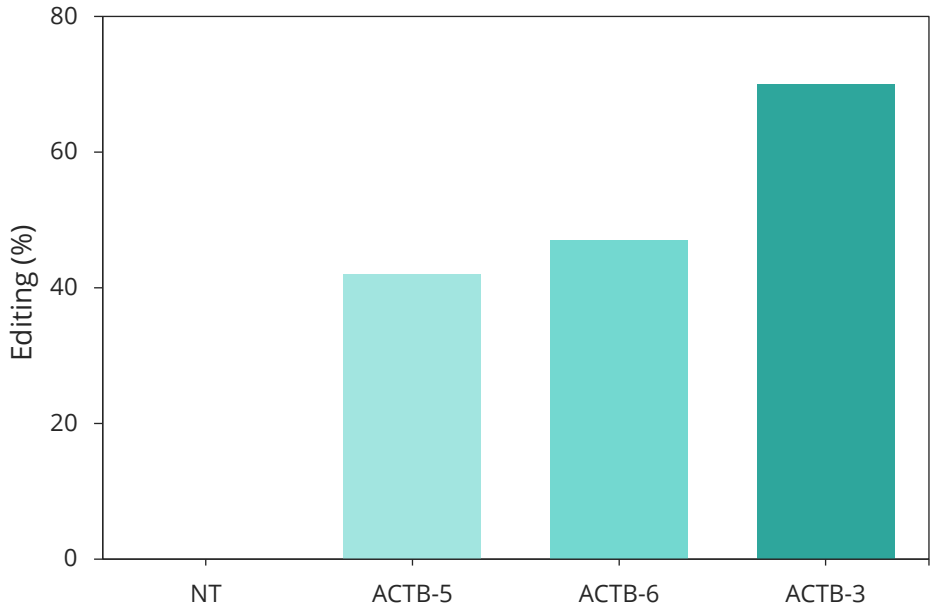
Liver organoids

Mice *in vivo*

Up to 70% RNA editing in human primary hepatocytes

RNA editing of *ACTB* in human primary hepatocytes

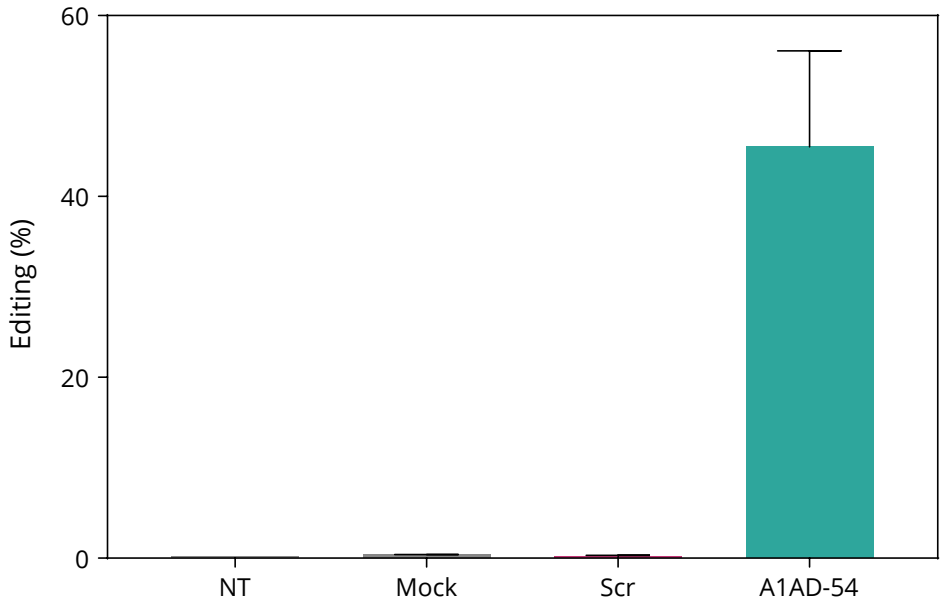
Gymnosis, 10µM, single dose, n=1, 48 hours, dPCR



Similar levels of RNA editing of *ACTB* achieved in several models of liver origin (not presented here)

RNA editing of *SERPINA1* E366K in human A1AD patient hepatocytes

Transfection, 100 nM, single dose, n=2, 47 hours, dPCR, mean, SD



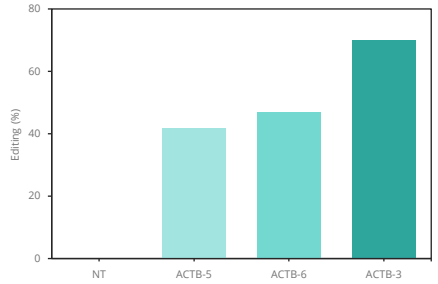
>50% RNA editing of *SERPINA1* E366K in human A1AD patient hepatocytes

A1AD: Alpha-1 antitrypsin deficiency.

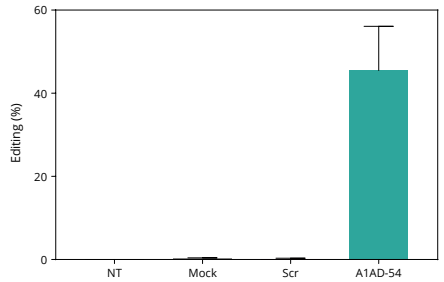
Advancing Axiomer® development across different models and targets in the liver

Cell models

Up to 70% RNA editing of *ACTB* in human primary hepatocytes



>50% RNA editing of *SERPINA1* E366K in human A1AD patient hepatocytes



Liver organoids

Mice *in vivo*

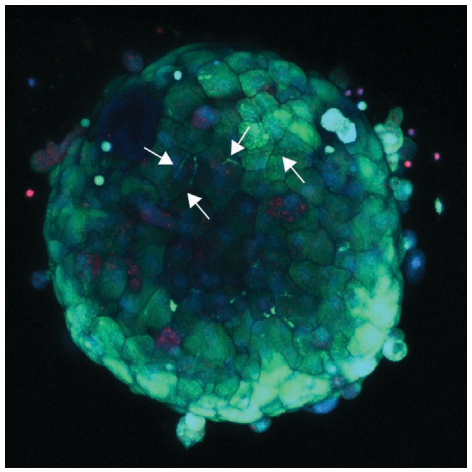
Conditions of *ACTB* editing experiment in human primary hepatocytes experiment: gymnosis, 10uM, single dose, N=1, 48 hours, dPCR; Conditions of the of *SERPINA1* editing experiment in human A1AD patient hepatocytes experiment: transfection, 100 nM, single dose, N=2, 47 hours, dPCR, mean, SD.

Editing in InSphero Human Liver microtissues (LMTs)

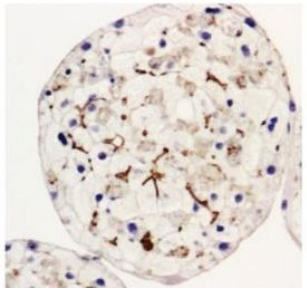
Primary hepatocytes, Kupffer cells and liver endothelial cells in 3D spheroid

Live imaging of LMT

Stained with 5-CFDA (green), PI (red) and Hoechst (blue)



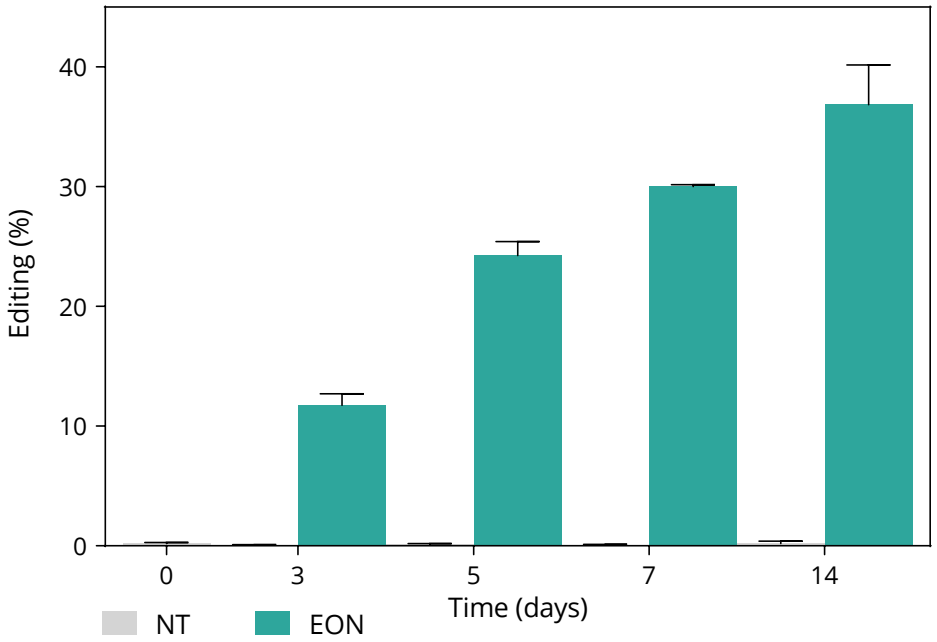
BSEP Bile Canaliculi (InSphero data)



Presence of bile channels in LMTs by day 7 fluorescent dye 5-CFDA secreted from healthy cells into bile channels (canaliculi)

Editing of *ACTB* in human LMTs

Gymnosis, 1µM, constant dose, 3 pools of 24 LMTs per condition, 14 days, dPCR, mean, SD



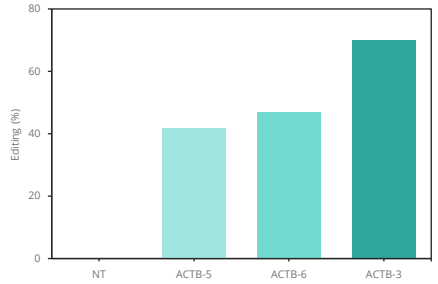
Treatment of LMTs with 1µM EON for 14 days results in up to 40% RNA editing of *ACTB*

BSEP: Bile salt export pump, LMTs: Liver Microtissues constituted of primary hepatocytes, Kupffer cells and liver endothelial cells in 3D spheroid.

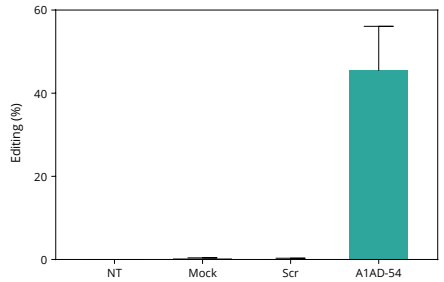
Advancing Axiomer® development across different models and targets in the liver

Cell models

Up to 70% RNA editing of *ACTB* in human primary hepatocytes



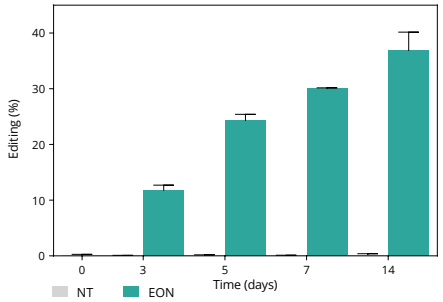
>50% RNA editing of *SERPINA1* E366K in human A1AD patient hepatocytes



Liver organoids

Up to 40% RNA editing of *ACTB* in human LMTs

Gymnosis, 1µM, constant dose, 3 pools of 24 LMTs per condition, 14 days, dPCR, mean, SD



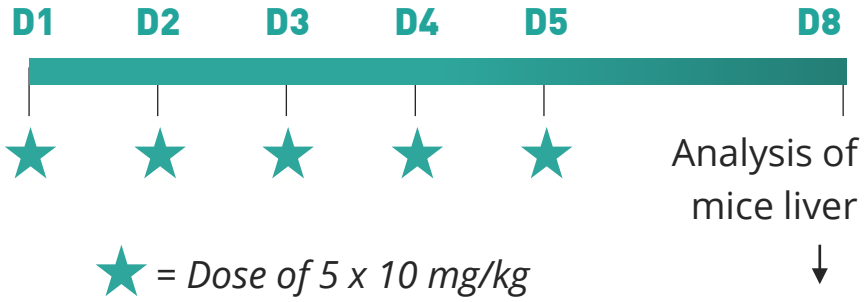
Mice *in vivo*

Conditions of *ACTB* editing experiment in human primary hepatocytes experiment: gymnosis, 10uM, single dose, N=1, 48 hours, dPCR; Conditions of the of *SERPINA1* editing experiment in human A1AD patient hepatocytes experiment: transfection, 100 nM, single dose, N=2, 47 hours, dPCR, mean, SD. LMTs: human liver microtissues.

Up to 50% RNA editing of *ActB* in liver of mice

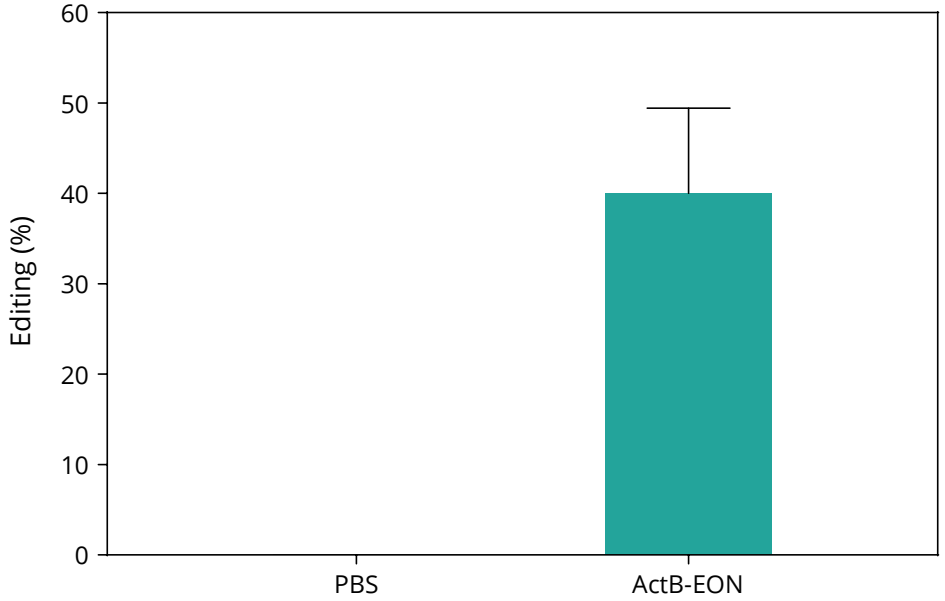
Mice treatment

In vivo



RNA editing of *ActB* in liver of mice

SC, 5 daily doses of 10 mg/kg, N=4, 1 week, ddPCR, mean, SD

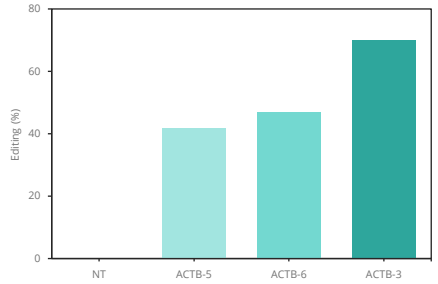


High *in vivo RNA* editing of *ActB* in the liver of mice reaching up to 50%

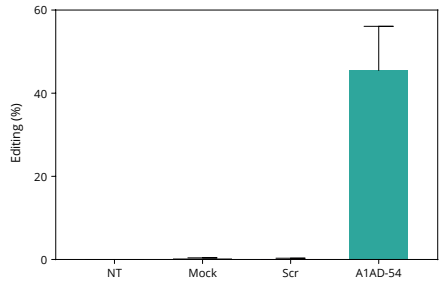
Advancing Axiomer® development across different models and targets in the liver

Cell models

Up to 70% RNA editing of *ACTB* in human primary hepatocytes



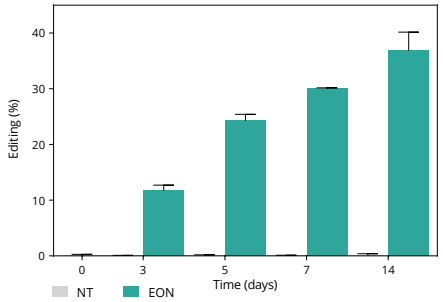
>50% RNA editing of *SERPINA1* E366K in human A1AD patient hepatocytes



Liver organoids

Up to 40% RNA editing of *ACTB* in human LMTs

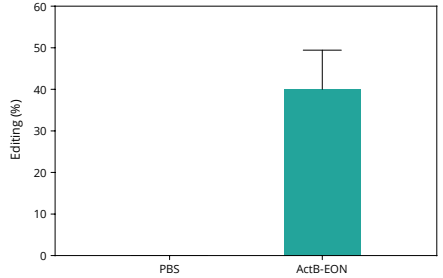
Gymnosis, 1µM, constant dose, 3 pools of 24 LMTs per condition, 14 days, dPCR, mean, SD



Mice *in vivo*

Up to 50% RNA editing of *ActB* in liver of mice

SC, 5 daily doses of 10 mg/kg, N=4, 1 week, ddPCR, mean, SD



Conditions of *ACTB* editing experiment in human primary hepatocytes experiment: gymnosis, 10µM, single dose, N=1, 48 hours, dPCR; Conditions of the of *SERPINA1* editing experiment in human A1AD patient hepatocytes experiment: transfection, 100 nM, single dose, N=2, 47 hours, dPCR, mean, SD. LMTs: human liver microtissues.

Assessing the potential of GalNAc on cell uptake and RNA editing efficiency

Liver

Targeting liver originated diseases

GalNAc

Optimizing liver delivery

BEA assay

Mice *in vivo*

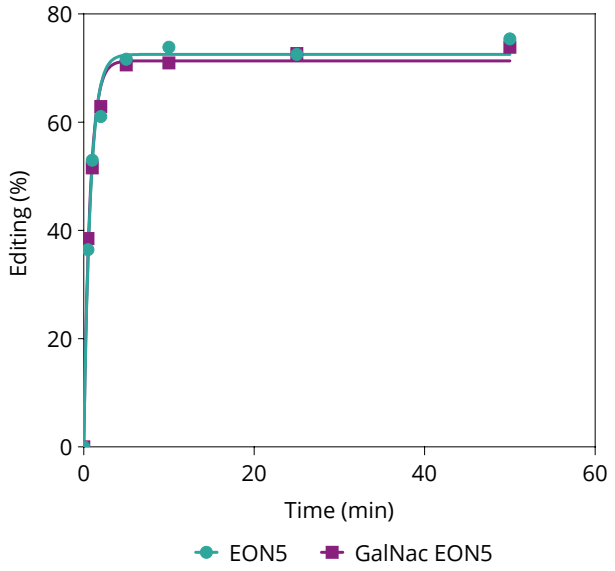
BEA, Biochemical editing assay.

Positive impact of GalNAc on cell uptake and RNA editing efficiency

BEA assay

GalNAc does not interfere A-to-I editing *in vitro*

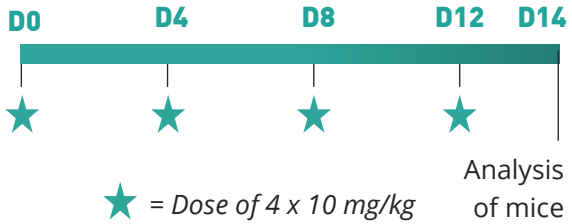
*2nM target RNA, 6nM EON and 6nM ADAR2, N=1, BEA assay**



Mice *in vivo*

Mice treatment

In vivo



★ = *Dose of 4 x 10 mg/kg*

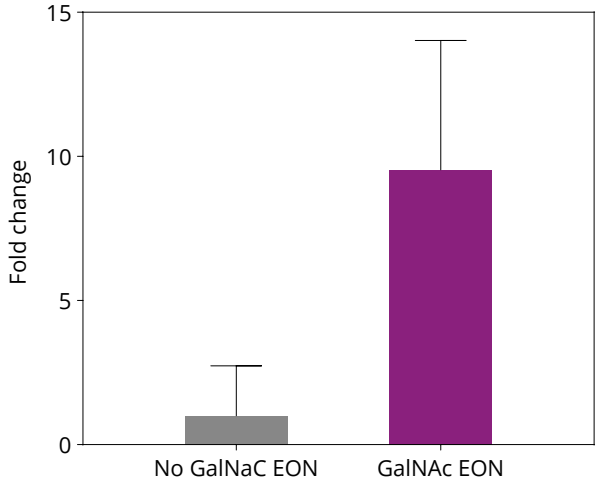
Analysis of mice liver

↓

Editing quantification using dPCR

10-fold change in editing in liver of mice**

SC, 4 doses of 10 mg/kg, N=4-5, 2weeks, dPCR, mean, SD



BEA, Biochemical editing assay; SC, subcutaneous; SD, standard deviation. *BEA assay timepoints 0, 0.5-, 1-, 2-, 5-, 10-, 25- and 50-min. **Undisclosed target.

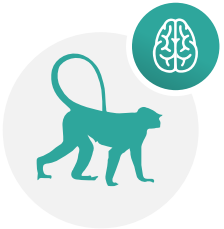
Axiomer® PoC in the nervous system and liver across multiple models including *in vivo*

Consistent RNA editing reported

in all models in nervous system and liver

Up to 40% editing reported in the nervous system of mice *in vivo*

Up to 50% editing reported in the liver of mice *in vivo*

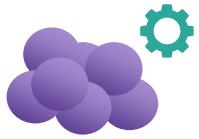
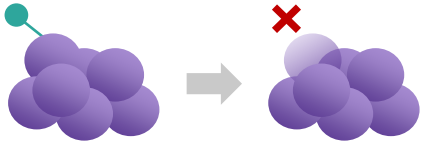


Approx. 50% editing reported in the nervous system of NHP *in vivo*

Increased editing efficiency and hepatocyte uptake *in vivo*

GalNAc does not interfere with A-to-I editing and leads to a 10-fold editing increase

Axiomer® creating a new class of medicines with broad therapeutic potential

Correction	Protein modulation		
			
Mutations correction Thousands of G-to-A mutations, many of them described in literature	**Alter protein function or include protective variants** Modified proteins achieving loss- or gain-of-functions that help addressing or preventing diseases	**Disrupt >400 different types of PTMs** Regulate protein activity, change localization, folding, preventing immune escape or slowing down degradation	**Change protein interactions** Changes localization, folding, protein function or prevents immune escape of glycosylated tumor antigens

BROAD THERAPEUTIC POTENTIAL

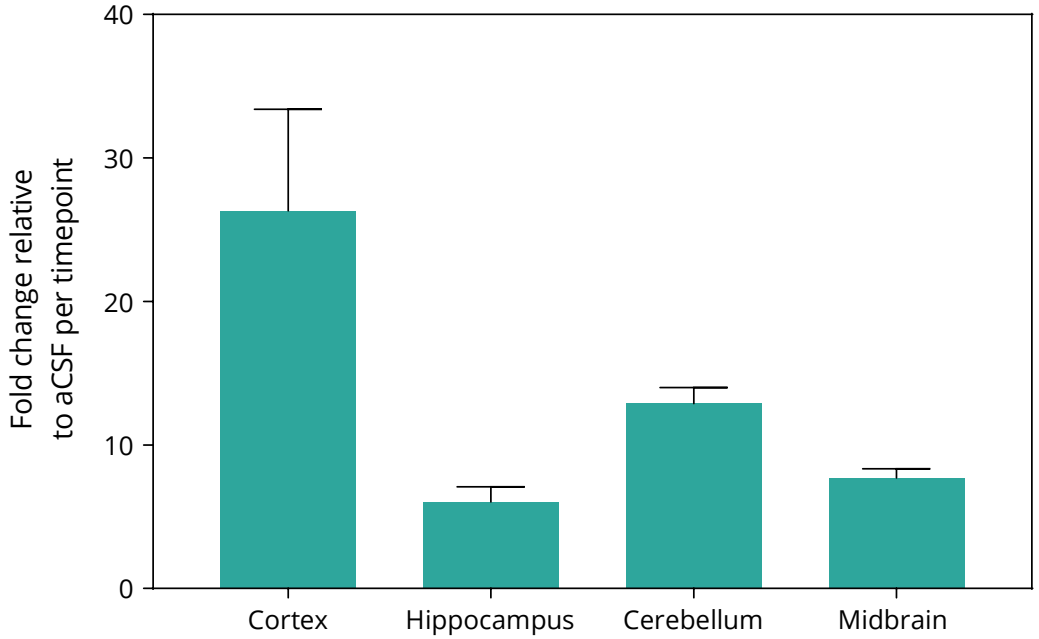
- Common diseases
- Rare diseases
- Target a wide variety of organs
- Treat so-far undruggable targets

PTMs: Post-translational modifications.

Mutation correction with Axiomer® leads to protein recovery

Protein function in mice

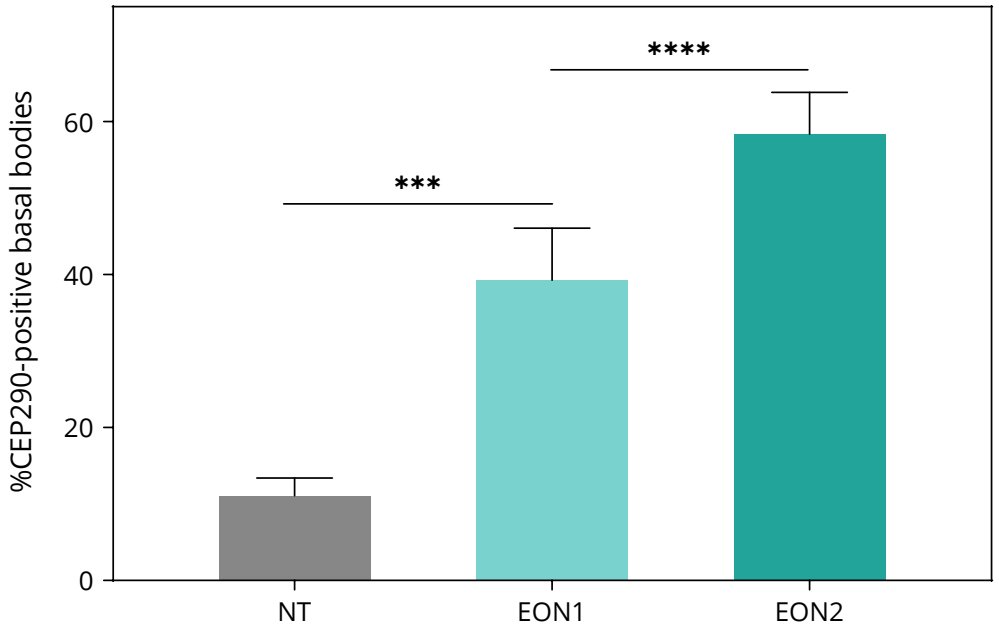
ICV, 250µg, single dose, N=6, 4 weeks, western blot, mean, SEM



In the brain, Axiomer® EONs lead to 26-fold increase in protein function in the cortex after editing

CEP290 protein recovery in organoids

Gymnosis, 10µM, single dose, N=8, 2 weeks, IF, mean, SD



Significant increase in CEP290 protein levels and intensity was detected at the basal body of LCA07-3 organoids treated with EONs after 2-weeks treatment

ICV: Intracerebroventricular injection, IMF: Immune Fluorescence; SD: standard deviation, SEM: Standard error of the mean, WT: wild type. Statistical significance was determined using Brown-Forsythe and Welch ANOVA test.

Axiomer® creating a new class of medicines with broad therapeutic potential

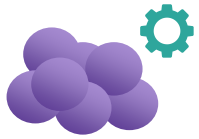
Correction

Mutations correction

Thousands of G-to-A mutations, many of them described in literature

Mutation correction leading to protein recovery

Protein modulation

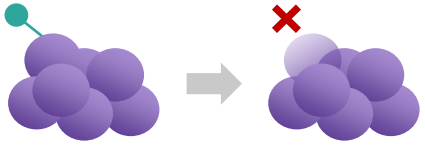


Alter protein function or include protective variants

Modified proteins achieving loss- or gain-of-functions that help addressing or preventing diseases

Disrupt >400 different types of PTMs

Regulate protein activity, change localization, folding, preventing immune escape or slowing down degradation



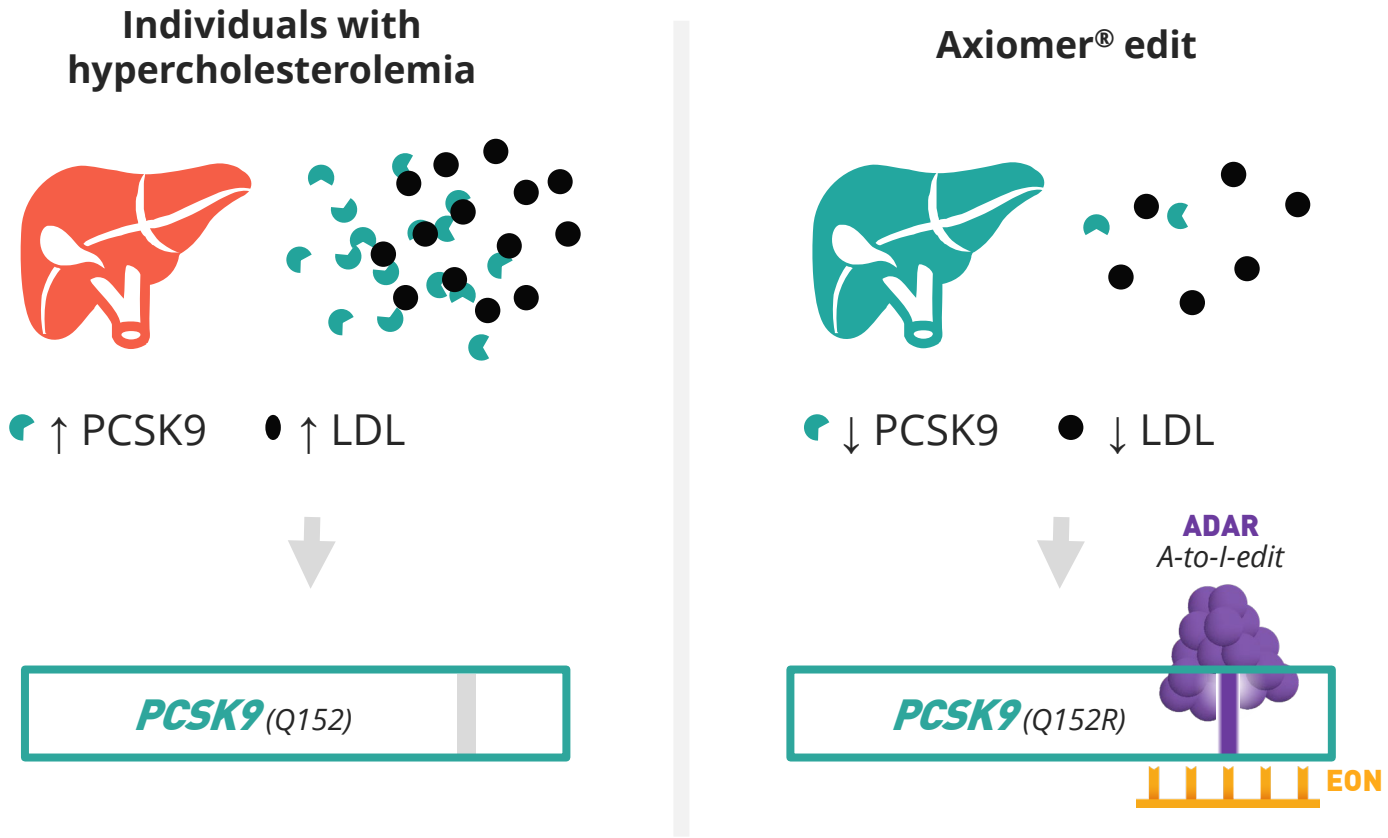
Change protein interactions

Changes localization, folding, protein function or prevents immune escape of glycosylated tumor antigens

PTMs: Post-translational modifications.

Changing the autocleavage site with Axiomer® leads to a LOF in PCSK9

Generation of a loss-of-function variant to lower PCSK9



Disruption of PCSK9 autocleavage site reduces protein in bloodstream

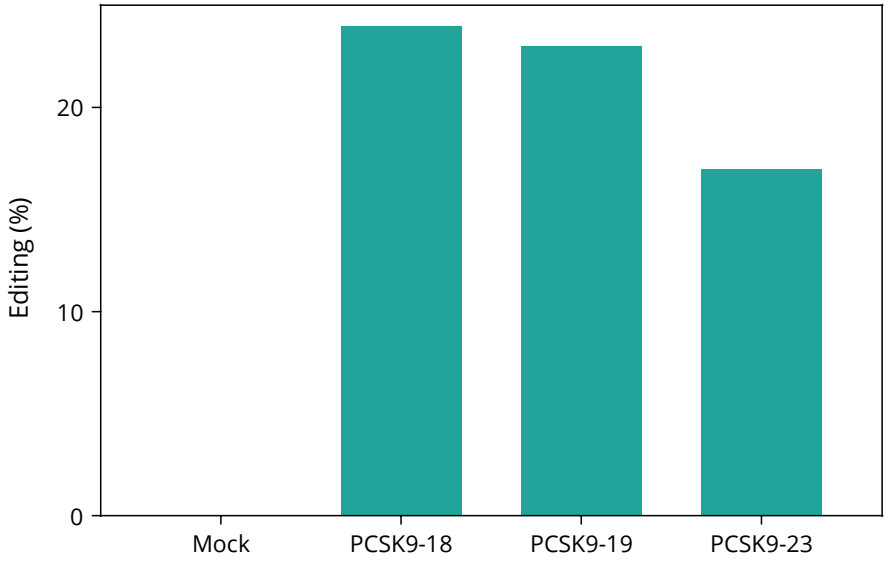
- Less PCSK9 leads to increase of LDL-R on cells, decrease of 'bad' LDL in bloodstream
- Loss-of-function *PCSK9* variant Q152H is associated with low plasma LDL cholesterol in a French-Canadian family and with impaired processing and secretion in cell culture

LDL: Low density lipoprotein, LDL-R: Low density lipoprotein receptor. LOF: Loss of function. Reference: Mayne J, et al. Clin Chem. 2011 Oct;57(10):1415-23.

Editing of *PCSK9* RNA results in a proenzyme with dominant negative properties

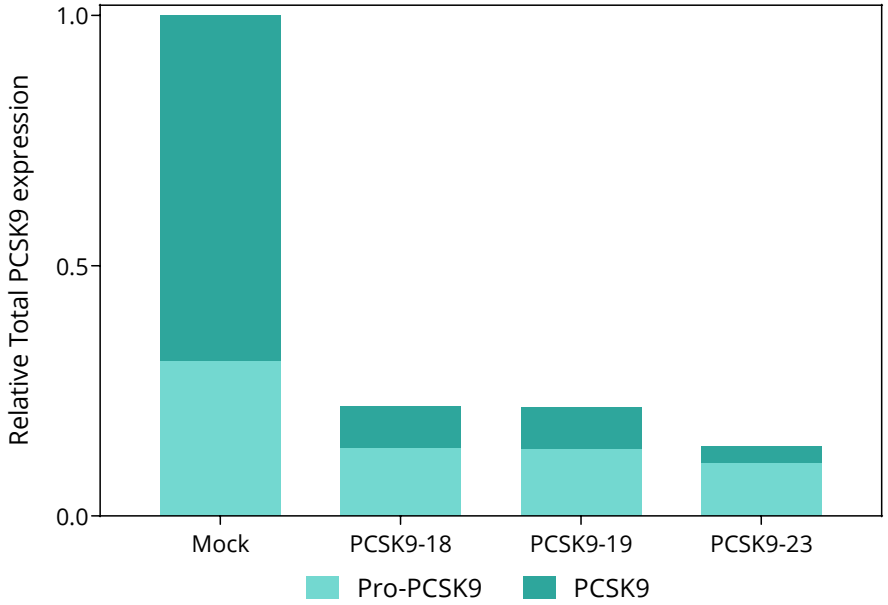
RNA editing of *PCSK9* in HeLa cells

Transfection, 100nM, single dose, N=2, 48 hours, ddPCR



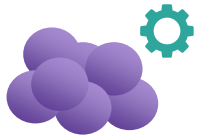
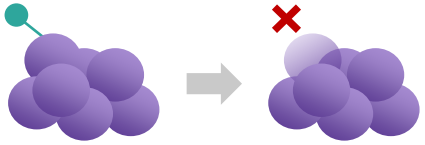
PCSK9 protein expression in HeLa cells

Transfection, 100nM, single dose, N=2, 48 hours, western blot



- Up to 25% A-to-I editing of *PCSK9* RNA detected using ddPCR assays leading up to 80% reduction of total PCSK9 protein
- Shift in the ratio cleaved to uncleaved PCSK9 observed; 70%:30% in mock to 25%:75% in treated samples
- The inability to undergo autocleavage likely retains the proenzyme in the endoplasmic reticulum where it can act as a dominant negative protein, preventing the exit of the wild-type form of PCSK9.

Axiomer® creating a new class of medicines with broad therapeutic potential

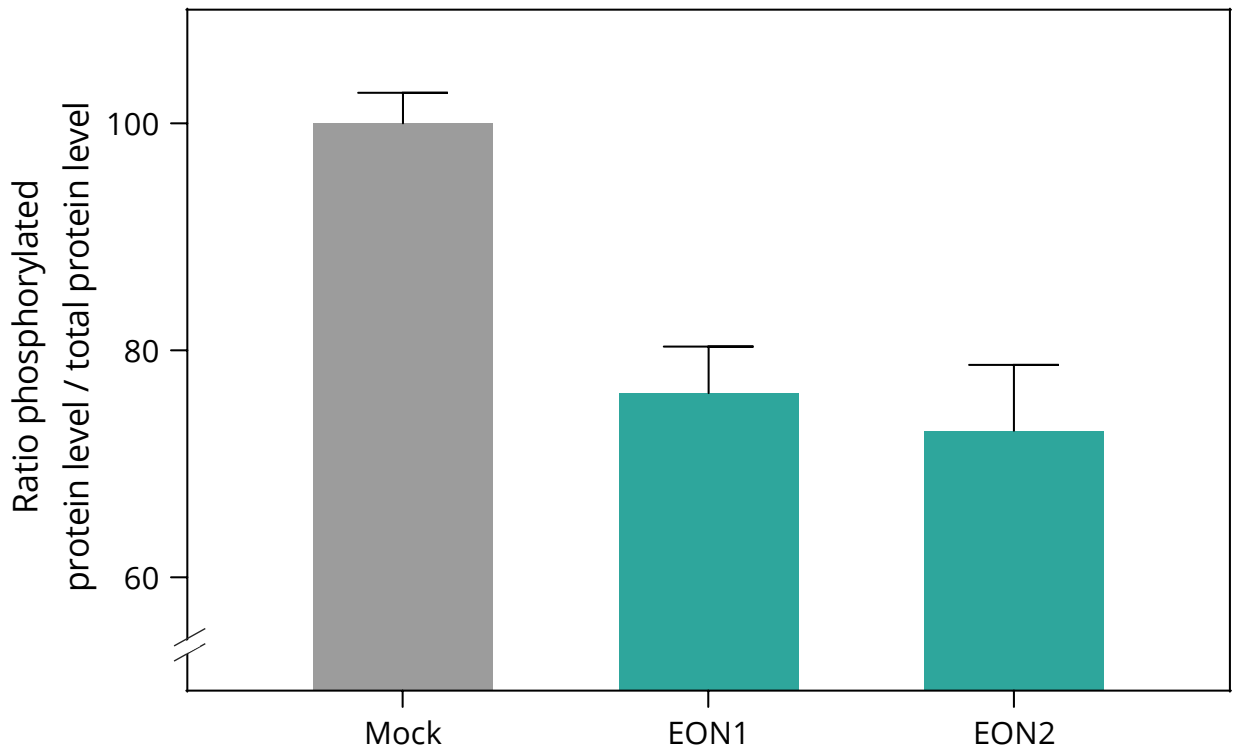
Correction	Protein modulation		
			
Mutations correction Thousands of G-to-A mutations, many of them described in literature	**Alter protein function or include protective variants** Modified proteins achieving loss- or gain-of-functions that help addressing or preventing diseases	**Disrupt >400 different types of PTMs** Regulate protein activity, change localization, folding, preventing immune escape or slowing down degradation	**Change protein interactions** Changes localization, folding, protein function or prevents immune escape of glycosylated tumor antigens
Mutation correction leading to protein recovery	Variant resulting in a dominant negative effect		

PTMs: Post-translational modifications

Changing a specific amino acid with Axiomer® reduces phosphorylation

Change in phosphorylated protein ratio*

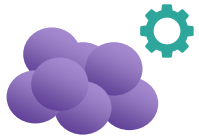
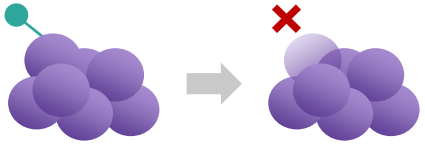
Transfection, 400nM, N=2-4, 48 hours, western blot, SD



- Specific A-to-I editing achieved with Axiomer® EONs changes protein post-translational modification
- Reduction of protein phosphorylation alters protein function
- Approximatively 25% reduction in the phosphorylated protein vs. total protein level achieved with 2 EONs

*Undisclosed target. EONs: Editing oligonucleotides

Axiomer® creating a new class of medicines with broad therapeutic potential

Correction	Protein modulation		
			
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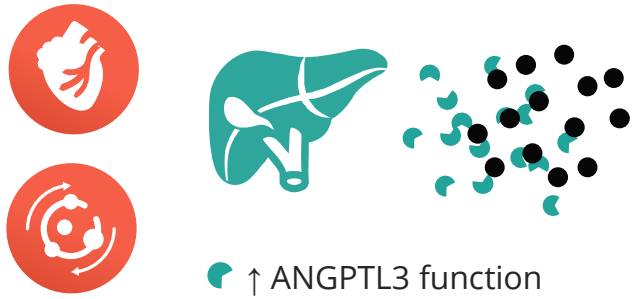
Mutation correction leading to protein recovery	Variant resulting in a dominant negative effect	Reduction of protein phosphorylation altering protein function	

PTMs: Post-translational modifications

Changing a protein binding site with Axiomer® leads to a LOF in ANGPTL3

Generation of a loss of function variant to activate lipoprotein lipases

Individuals with increased CVD risk



↑ ANGPTL3 function

↑ LDL + triglycerides

Axiomer® edit

↓ ANGPTL3 function

↓ LDL + triglycerides

Wildtype ANGPTL3

AAAGACTTTGTCCAT**AAG**ACGAAGGGCCAAATTAAT

-K--D--F--V--H--**K**--T--K--G--Q--I--N-

Edited ANGPTL3

AAAGACTTTGTCCAT**GAG**ACGAAGGGCCAAATTAAT

-K--D--F--V--H--**E**--T--K--G--Q--I--N-

= Heparin-binding motif

ANGPTL3 is an angiopoietin-like factor that inhibits lipoprotein lipases (LPL)

- Increase triglyceride, cholesterol, and non-esterified fatty acids in plasma leading to an increased risk of CVD

Reported loss of function variant of ANGPTL3

- Significantly decreased triglycerides, LDL-cholesterol, and total cholesterol
- Significantly decreased odds ratio for coronary artery disease

Heparin binding was shown to be essential for proper ANGPTL3 function

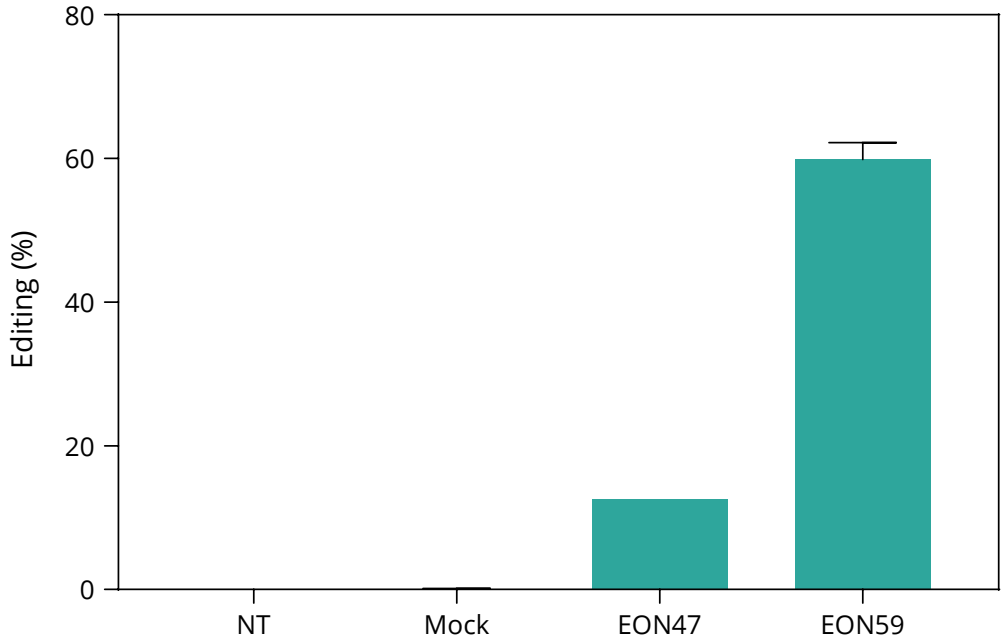
- Disruption of the heparin binding site is highly likely to abrogate LPL inhibition, ultimately leading to lipid lowering in the serum

CVD; cardiovascular disease. LDL: low density lipoprotein, LOF: Loss of function. References: Ono M et al. J Biol Chem. 2003 Oct 24;278(43):41804-9; Romeo S et al. J Clin Invest. 2009 Jan;119(1):70-9; Dewey FE et al. N Engl J Med. 2017 Jul 20;377(3):211-221.

ANGPTL3 variant disrupting essential protein binding site

More than 60% RNA editing of *ANGPTL3* in primary human hepatocytes derived spheroids

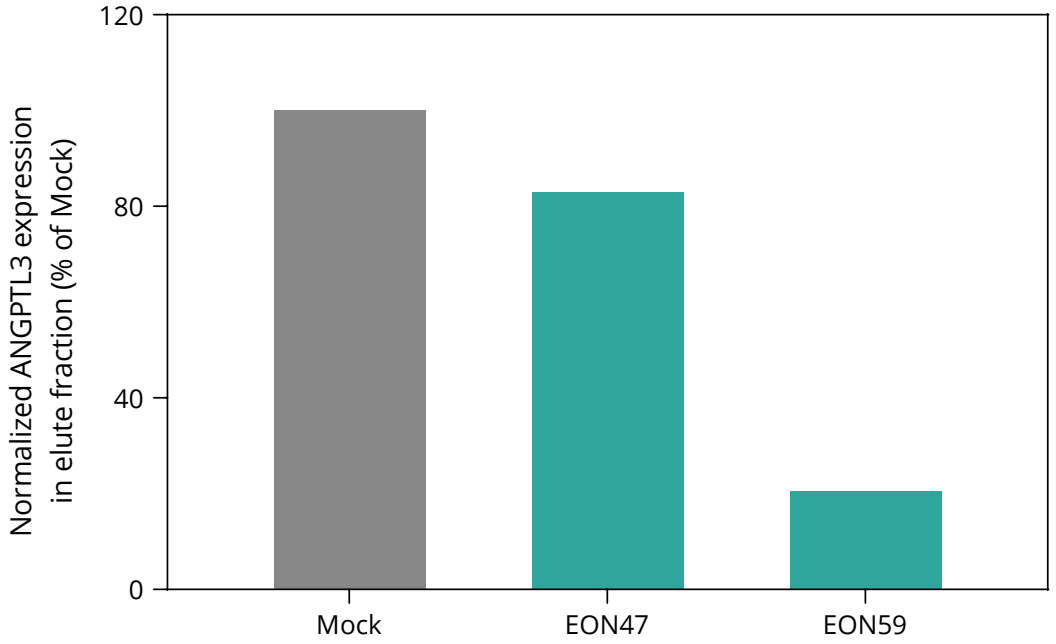
Gymnosis, 1µM, single dose, N=1 or 2, 5 days, dPCR, mean, SD



More than 60% RNA editing of *ANGPTL3* in primary human hepatocytes derived spheroids

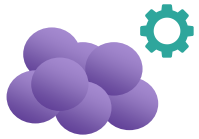
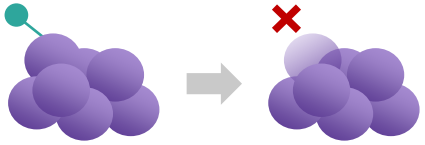
Up to 80% decrease in heparin binding in Huh-7 cells

Gymnosis, 1µM, single dose, N=1, 72 hours, western blot



Up to 80% decrease in heparin binding in Huh-7 cells

Axiomer® creating a new class of medicines with broad therapeutic potential

Correction	Protein modulation		
			
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Mutation correction leading to protein recovery	Variant resulting in a dominant negative effect	Reduction of protein phosphorylation altering protein function	Variant impacting protein interaction with sugar

Axiomer® RNA editing platform has broad potential

Consistent RNA editing

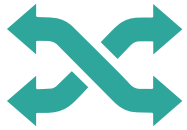
in all models evaluated in nervous system and liver, including NHP *in vivo*

Increased editing efficiency and hepatocyte uptake *in vivo*

GalNAc does not interfere with A-to-I editing and leads to editing increase

Validation of Axiomer's potential for therapeutic targets

With positive effect on protein expression



Broad applicability

With proof of concept in mutation correction and multiple forms of protein modulation

IP overview and Partnering strategy

René Beukema, Chief Corporate Development Officer

Overview of Axiomer® related patents

Docket	Priority	Feature	Status
1 (0004)	17DEC2014	Targeted RNA Editing using endogenous ADARs	Granted CA CN EP IL JP NZ RU US ZA
2 (0013)	22JUN2016	Short EONs with wobble and/or mismatch base pairs	Granted IL JP KR US
3 (0014)	01SEP2016	Chemically modified short EONs	Granted CN EP JP KR NZ US ZA
4 (0016)	19JAN2017	EONs + protecting SONs (heteroduplex formation)	Granted US
5 (0023)	18MAY2018	PS linkages / chiral linkages (*e.g.,* PS, PN)	Published
6 (0026)	11FEB2019	Phosphonacetate linkages / UNA modifications	Published
7 (0029)	03APR2019	MP linkages	Published
8 (0031)	24APR2019	Editing inhibition	Published
9 (0032)	13JUN2019	Benner's base (dZ)	Published Granted ZA
10 (0039)	23JUL2020	Split EONs	Published

In addition to the above, numerous patent applications are pending but have not yet been published.
ProQR expands its Axiomer® IP portfolio continuously.

Leading IP supporting ADAR-mediated RNA editing platform technology

- Axiomer® IP strategy commenced in 2014 with first patent application filings
  - Today 10 published patent families, currently comprise 22 patents
- Continuing to invest in and expand our IP estate
- March 2023 successful defense of key Axiomer® patent protecting ADAR-mediated RNA editing (EP 3234134 B1)
- Oppositions filed in February 2021 with the European Patent Office (EPO) by two separate strawmen against ProQR's granted patent EP 3434134 B1, which is related to targeted RNA editing using endogenous ADARs
- Following public hearing, EPO ruled in favor of ProQR

Partnering pillar of our strategy provides significant optionality and upside potential

- Partnerships bring resources, capabilities, and funding to further advance our pipeline programs as well as a new class of medicines based on our Axiomer® RNA editing technology platform
  - ProQR brings deep RNA editing and oligonucleotide development expertise, IP
- Initial Axiomer® partnership with Lilly September 2021, with expansion in December 2022 – total potential value of $3.9 B, plus potential royalties for a total of up to 15 targets
  - ProQR and Lilly to develop editing oligonucleotides for ten targets; Lilly has an option for an additional five targets for $50 M opt in fee
  - ProQR has received $125 M consisting of an upfront payments and equity investments
- Partnering ophthalmology assets (does not use Axiomer®)
- Selectively enter additional partnerships

Axiomer® has the potential to target a broad range of diseases

Metabolic disorders

- Type 2 diabetes
- Obesity

Lung diseases

- Cystic Fibrosis
- Primary ciliary dyskinesia
- Surfactant Metabolism Dysfunction
- ABCA3 deficiency
- Familial Pulmonary Fibrosis

Liver disorders

- Alpha-1 antitrypsin deficiency
- Cholestatic disorders
- Factor V Deficiency
- Hemophilia B
- Hereditary hemochromatosis
- Hurler Syndrome
- Ornithine transcarbamylase deficiency
- Non-alcoholic steatohepatitis
- Non-alcoholic fatty liver disease
- Pompe Disease
- Porphyrias
- Transthyretin-related hereditary amyloidosis
- Wilson disease

CNS and Neuromuscular disorders

- Parkinson's Disease
- Spinocerebellar Ataxia
- Alzheimer's Disease
- Huntington's Disease
- Pain disorders

Ophthalmology

- Inherited retinal disorders
- Fuchs Endothelial Corneal Dystrophy
- Primary Congenital Glaucoma

Oncology

- KRAS driven tumors
- P53 driven tumors

Blood / Cardiovascular system disorders

- Hypercholesterolemia
- Thrombophilia
- Alpha/Beta thalassemia
- Progeria

Kidney

- Polycystic kidney disease

Immunological disorders

- Paroxysmal nocturnal hemoglobinuria

Pipeline

Gerard Platenburg, Chief Scientific Officer

Millions of known sites within the RNA where ADARs perform A-to-I editing

Wide ADAR expression

RNA editing is naturally occurring with wide ADAR expression

Broad therapeutic potential

with the possibility to target multiple organs

Unlocking the therapeutic potential of Axiomer® through the liver

Enhanced delivery and available biomarkers

High editing in the liver

High expression of ADARs in the liver

GalNAc liver targeting

- 85% of the hepatocytes present the Asialoglycoprotein receptor
- GalNAc does not interfere with ADARs

Derisked delivery

Proven accessibility to RNA therapeutics through subcutaneous injection

Validated biomarkers

That correlate with the type and severity of liver diseases

Axiomer® can address numerous liver-originated diseases

Pipeline programs informed by human genetics

High metabolic activity & influence on other organs

- Bile production and excretion
- Lipid production and metabolism
- Plasma proteins synthesis
- Glucose regulation
- Micronutrients
- Detoxification

Numerous liver-originated diseases

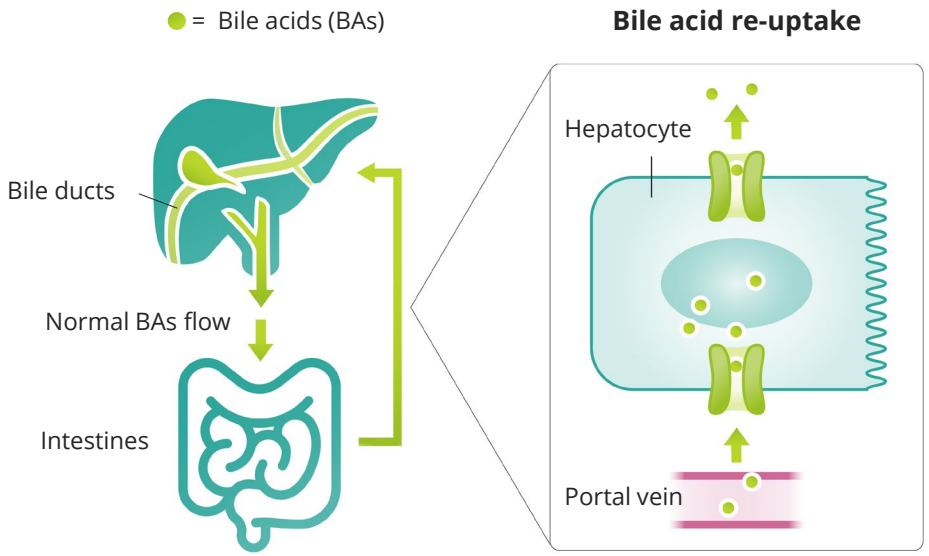
- Cholestatic diseases
- Metabolic disorders
- Cardiovascular diseases
- Storage diseases
- Coagulation disorders
- Many others…

AX-0810 for Cholestatic Diseases

Excessive accumulation of bile acids in the liver leads to cell stress and damage

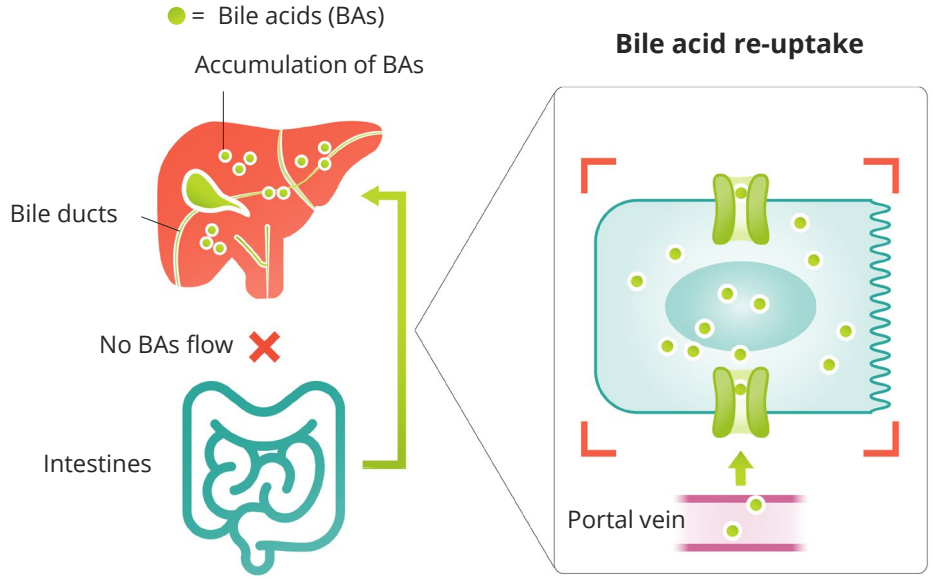
Healthy individuals

Normal BA levels in the hepatocytes



Individuals with cholestatic diseases

Excessive BA levels in the hepatocytes



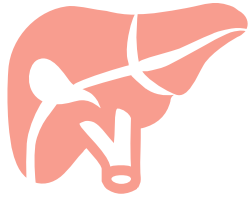
Cell stress and damage lead to activation of the immune system

Dysfunctional bile ducts lead to a toxic buildup of bile acids in the liver of individuals with cholestatic diseases

References: Cai SY, et al. JCI Insight. 2017;2(5):e90780 and Cai SY, Boyer JL. Ann Transl Med. 2021 Apr;9(8):737

Cholestatic diseases remain a leading cause of liver transplantation

Liver inflammation and fat-soluble vitamin deficiencies



Liver fibrosis – cirrhosis and portal hypertension

Liver failure and malignancy

Progression leads to poor life prognosis and need for transplantation

References: Arndtz K, Hirschfield GM. Frontline Gastroenterol. 2017 Oct;8(4):260-266; Cheung AC, et al. Dig Dis Sci. 2016 Jun;61(6):1692-9; Le M, Reinshagen K, Tomuschat C. J Pediatr Surg. 2022 Dec;57(12):934-946; Carbone M, Neuberger J. Clin Res Hepatol Gastroenterol. 2011 Jun;35(6-7):446-54; Sundaram SS, et al. Liver Transpl. 2017 Jan;23(1):96-109.

High unmet medical needs remain in Primary Sclerosing Cholangitis and Biliary Atresia

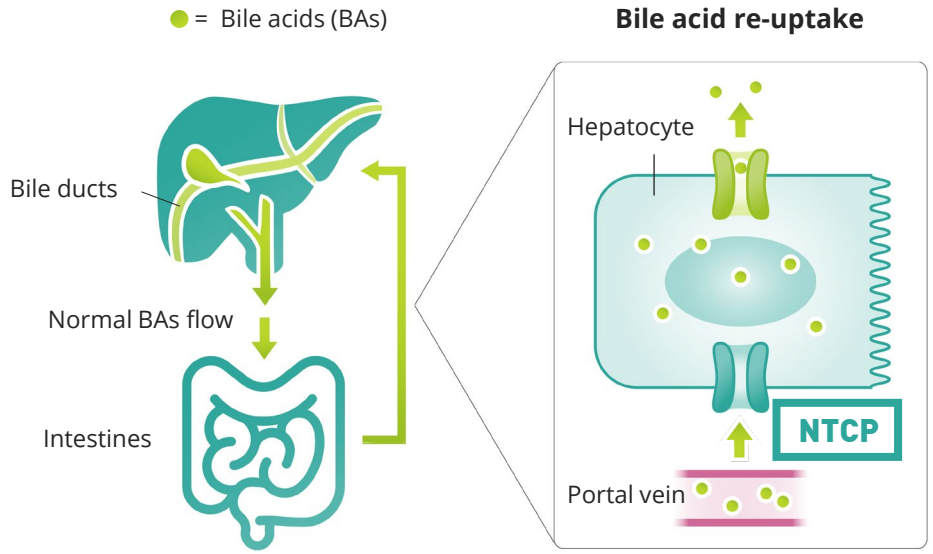
	Primary Sclerosing Cholangitis (PSC)	Biliary Atresia (BA)
Diagnosis	Adult : ~30-40 years, 2/3 men	Pediatric: first weeks of life
Population*	≈ 80K individuals	≈ 24K individuals
Patho-physiology	Bile duct strictures due to fibrosis and sclerosis	Absent or defective bile ducts
Symptoms	Pruritus, fatigue, pain, weight loss, recurrent bacterial infection, inflammatory bowel disease (70%)	Jaundice, poor weight gain, pale stool, dark urine
Progression	Progression to liver cirrhosis and increased risk of cancer	Rapid progression to cirrhosis and portal hypertension early in life
Standard of Care	**No approved therapy**	**No approved therapy Hepatoportoenterostomy****

*Patient population estimation in the international Group of Seven countries (G7) and based on current population data and literature,. **Surgery removing the extrahepatic biliary tree and linking the liver directly to a loop of intestine to enable bile to enter the bowel. References: Boonstra K, Beuers U, Ponsioen CY. J Hepatol. 2012 May;56(5):1181-1188; Karlsen TH, et al. J Hepatol. 2017 Dec;67(6):1298-1323; Dyson JK, et al. Lancet. 2018 Jun 23;391(10139):2547-2559; Neuberger J. 2003 Apr;6(2):113-121; Sundaram SS, et al. Liver Transpl. 2017 Jan;23(1):96-109. Raghu VK, et al. Liver Transpl. 2021 May;27(5):711-718; NORD, 2019. Hirschfield G, et al. The Lancet. 2013. 382, 9904, 1587 – 1599. Japanese Biliary Atresia Society. Japanese Biliary Atresia Registry (JBAR). https://jbas.net/en/national-registration/.

Blocking bile acids re-uptake into the liver reduces excessive and toxic accumulation

- NTCP (*SLC10A1* gene): Sodium (Na+)-taurocholate cotransporting polypeptide
  - 95 % of liver bile acid are recycled from the intestine back to the liver via NTCP
- In the human genetics, loss of function (LOF) variants in NTCP naturally occur in some people exhibiting a mild phenotype
- Pharmacological modulation of NTCP improves outcomes of cholestasis reducing liver damage and inflammation in a mouse model

NTCP is the main transporter involved in bile acids re-uptake from the portal circulation to the liver

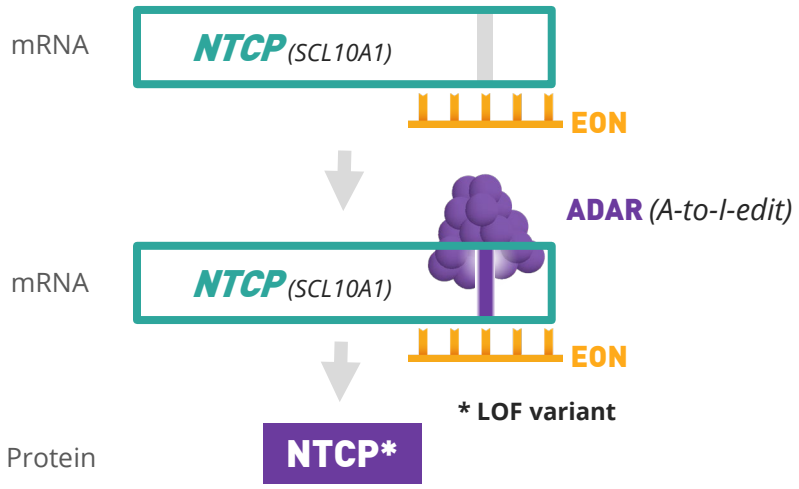


NTCP: Na-taurocholate cotransporting polypeptide. *SLC10A1* is the gene that encodes for NTCP protein. References: Ho RH, et al. J Biol Chem. 2004 Feb 20;279(8):7213-22; Vaz FM, et al. Hepatology. 2015;61(1):260-267; Vaz FM, et al. Dig Dis. 2017;35(3):259-260; Slijepcevic D, et al. Hepatology. 2018 Sep;68(3):1057-1069; Mao F, et al. J Biol Chem. 2019;294(31):11853-11862.

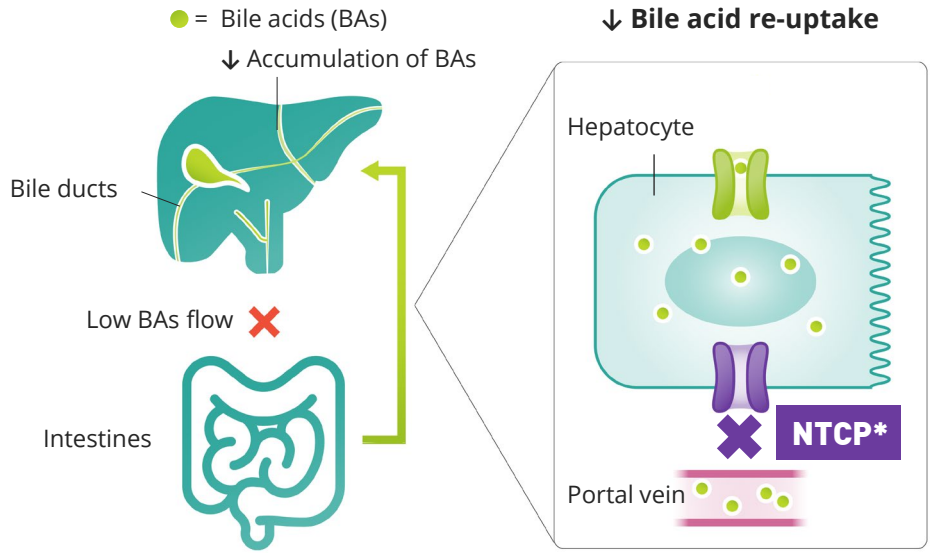
AX-0810 is designed to reduce bile acids re-uptake into the liver

RNA editing to a loss of function variant of NTCP can improve liver function

AX-0810 therapy for cholestatic diseases



Reduced BA levels in the hepatocytes



- AX-0810 is a novel and "on target" approach reducing bile acid re-uptake into the hepatocytes
  - Transient and controlled approach introducing a loss of function of NTCP
- AX-0810 can reduce bile acid load in the liver
  - To alleviate associated pathology and symptoms in PSC and BA
  - To prevent or delay the development of cirrhosis, organ failure and need for transplant

BA: Bile acids, NTCP: Na-taurocholate cotransporting polypeptide, PSC: Primary Sclerosing Cholangitis. *SLC10A1* is the gene that encodes for NTCP protein.

Well-defined development path for AX-0810

PRECLINICAL STAGE	EARLY CLINICAL	LATE CLINICAL
Preclinical models available with strong translatability into the clinic	*Early insight on safety and target engagement using validated biomarkers*	*Clinical programs with disease specific endpoints for regulatory approval*
Translational models available • Organoids models • Animal models **Proof of mechanism measures in animal models** • Serum levels of ALP and γ-GT • Total bile acids in serum and liver • Hepatic inflammation and fibrosis	**Program with Phase 1 on healthy volunteers** **Validated biomarkers in cholestatic diseases** • Bile acids in serum, urine and feces • Liver enzymes • Serum cholesterol **Disease specific biomarkers in preparation for next trials:** • ALP for PSC • Bilirubin for BA	**Primary Sclerosing Cholangitis** Co-primary endpoint for regulatory approval: • Reduction in ALP and • Histological liver evaluation **Biliary atresia** • Time to liver transplantation • Mean change in total serum bilirubin levels, liver enzymes, bile acid levels, blood platelets and serum albumin

γ-GT: γ-glutamyl transferase; ALP, Alkaline phosphatase; BA, biliary atresia; BDL, Bile duct ligation; LMT, Liver microtissues; NTCP, Na-taurocholate cotransporting polypeptide; PSC, Primary Sclerosing Cholangitis

AX-0810 early evidence generation approach on safety and target engagement

Phase 1 on healthy volunteers for cholestatic diseases

Objectives

- Assess safety, tolerability, PK and PD of AX-0810 without interference by concomitant pathological conditions
- Establish target engagement by biomarkers

Trial design

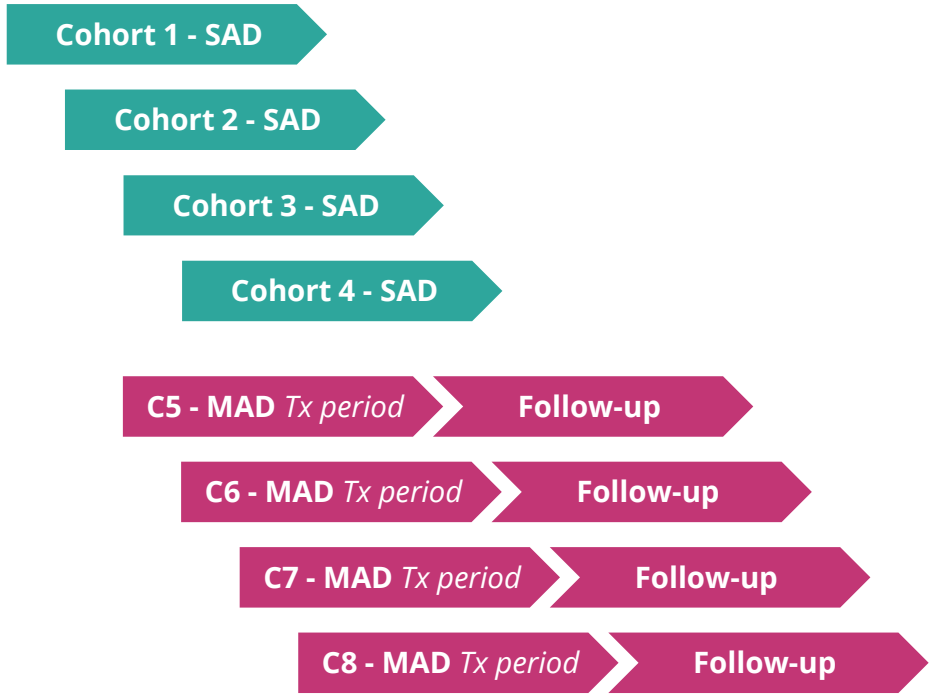
- Single and multiple dose ascending trial
- Single trial site: timely recruitment and data generation

Endpoints will include

- Safety, tolerability, PK and PD of AX-0810
- Change in bile acids in serum, urine and feces, liver enzymes and serum cholesterol
- Change in disease specific biomarkers: ALP and bilirubin
- Measure RNA editing in circulating exosomes in plasma

Entry into clinical trials in late 2024 / early 2025

Preliminary study design



ALP, Alkaline phosphatase; MAD, multiple ascending dose; PD, Pharmacodynamic; PK, Pharmacokinetics; SAD, single ascending dose.

Summary and next steps for AX-0810

High unmet medical need in cholestatic diseases

with PSC and BA being leading indications for liver transplantation

A rigorous approach to increase probability of success

from preclinical to the clinical stage, including validated biomarkers and a clear regulatory pathway

AX-0810 is a novel and “on target” approach

- Originated from human genetics
- Reducing bile acid re-uptake into the liver via NTCP loss of function

Next steps

- Generate new data on hit and lead selection
- Entry into clinical trials in late 2024 / early 2025

BA, biliary atresia; NTCP, Na-taurocholate cotransporting polypeptide; PSC, Primary Sclerosing Cholangitis

Axiomer® can address numerous liver-originated diseases

Pipeline programs informed by human genetics

High metabolic activity & influence on other organs

- Bile production and excretion
- Lipid production and metabolism
- Plasma proteins synthesis
- Glucose regulation
- Micronutrients
- Detoxification

Numerous liver-originated diseases

Cholestatic diseases

Metabolic disorders

Cardiovascular diseases

Storage diseases

Coagulation disorders

Many others...

AX-1412 for Cardiovascular Diseases (CVD)

Despite current therapies, CVD remains the highest cause of disability and death in the world

Leading causes of death in the world

~18M people die from CVDs every year (32% of all global deaths)

With projected increased number of patients

By 2035, >130 million adults in the US are projected to have some form of CVD with a total costs of $1.1 trillion

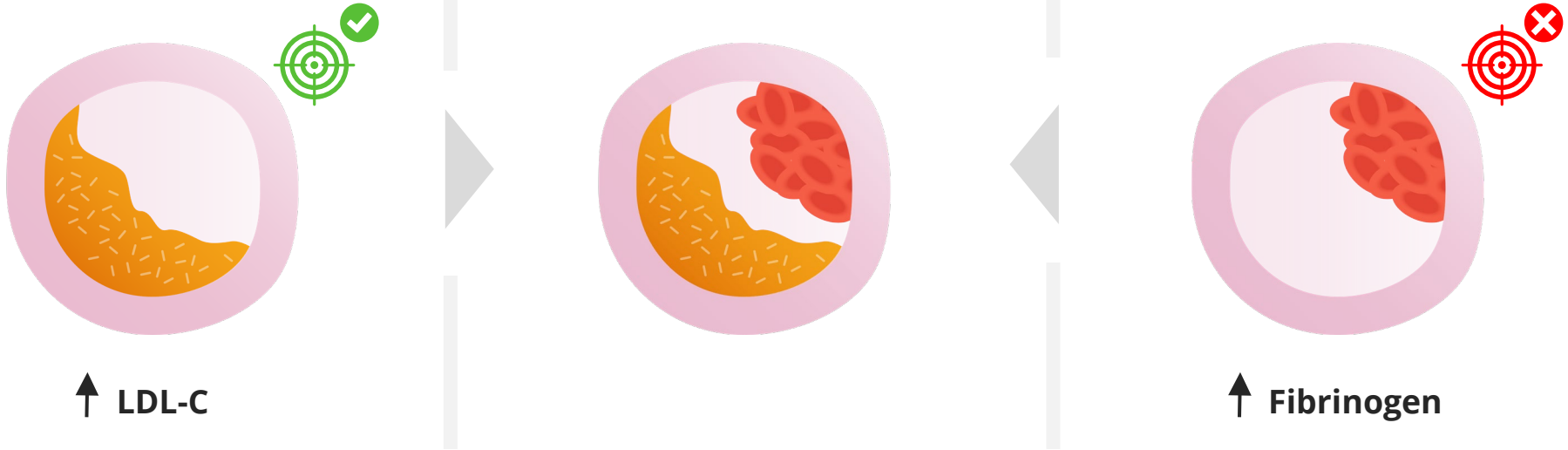
Unmet medical need remains

Current standard of care includes lipid lowering therapies and hypertension medications. Despite therapies, the unmet medical need remains

- Residual risk despite SoC
- Low treatment adherence
- Tolerability issues

CVDs: cardiovascular diseases. References: WHO report 2021, Heart Disease and Stroke Statistics—2018 Update: A Report From the American Heart Association; BMJ 2017;357:j1956; Hill MF, Bordoni B. Hyperlipidemia. 2022 Aug 8. Treasure Island (FL): StatPearls; Thompson PD, et al. J Am Coll Cardiol. 2016 May 24;67(20):2395-2410.

Two independent risk factors involved in CVD may have negative synergistic effect



LDL-c and fibrinogen are two **independent risk factors** involved in cardiovascular diseases, atherosclerosis and thrombotic events

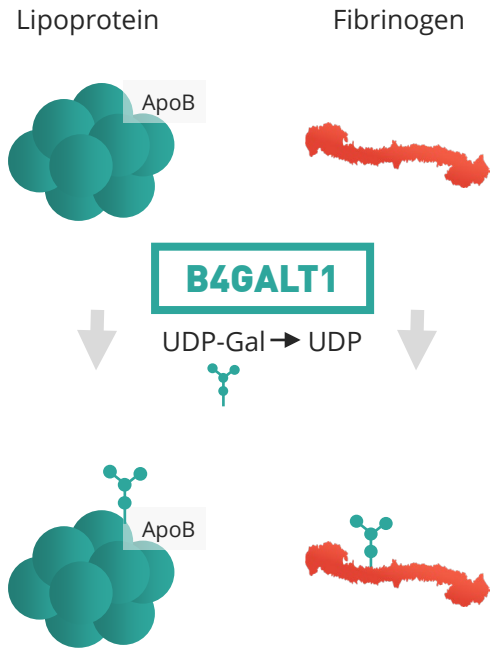
ASCVDs, atherosclerotic cardiovascular diseases; CVDs, cardiovascular diseases; LDL-c, Low-density lipoprotein cholesterol. References: Linton MF, et al. Endotext. South Dartmouth (MA): MDText.com, Inc., January 3, 2019; Vilar R et al. 2020, Haematologica 105, 284–296; Zabczyk, M et al. Clinical Outcomes. J. Clin. Med. 2021, 10, 2999; Surma S et al. 2022 Int. J. Mol. Sci. 23, 193; Ząbczyk M, et al. J Clin Med. 2021;10(13):2999.

Genetic and functional evidence links a missense variant in *B4GALT1* to lower LDL-C and fibrinogen

B4GALT1 plays a role in transferring galactose to CVD risk factors

Beta-1,4-galactosyltransferase 1 (B4GALT1)

B4GALT1 transfers galactose from uridine diphosphate galactose (UDP-Gal) to specific glycoprotein substrates

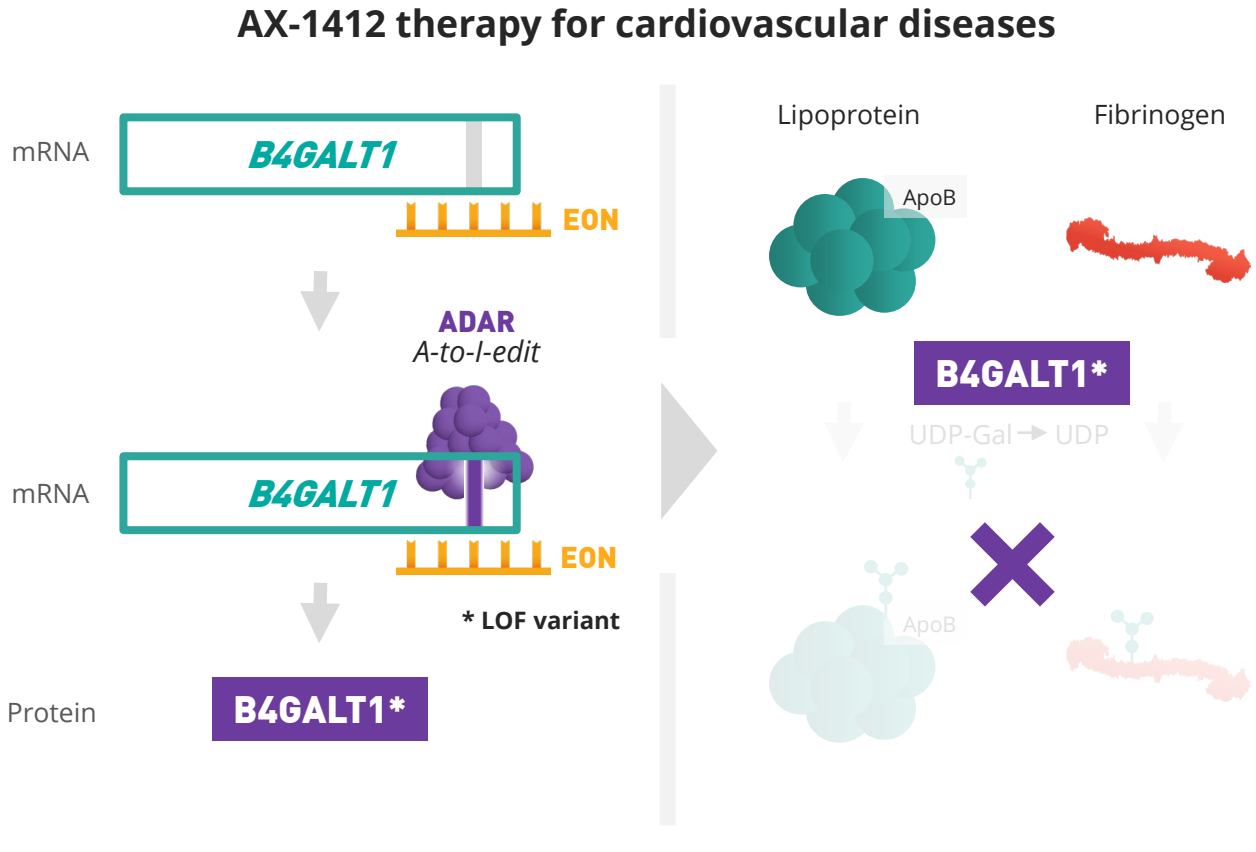


- Reported **Old Order Amish-enriched missense variant (p.Asn352Ser)** in a functional B4GALT1
  - Associated with lower serum LDL-C and lower plasma fibrinogen
  - 50% decrease in glycosylation efficiency
- B4GALT1 acts on apolipoprotein B100, fibrinogen, which are known drivers of increased risk of CVD
- *B4GALT1* **loss of function associates with decreased coronary artery disease** in gene–based analysis

ApoB: Apolipoprotein B, CVD: cardiovascular disease, LDL-C: Low-density lipoprotein cholesterol. Reference: Montasser ME. et al., 2021 Science 374(6572):1221-1227

AX-1412 brings a novel approach to reduce residual risk for a potential cardiovascular event

RNA editing to a loss of function variant of B4GALT1 can have pleiotropic effect targeting two CVD risk factors



***B4GALT1* p.N352S protective allele**

- Leads to hypo-galactosylation of apolipoprotein B100, fibrinogen

AX-1412 is a novel and unique approach to address CVD

- Pleiotropic effects for cardiovascular protection
- Not suitable for knockdown technologies, as leads to semi-lethality and severe development abnormalities in mouse studies

AX-1412 can lower LDL-C and fibrinogen levels to reduce residual risk in cardiovascular diseases

- Prevent or delay the development of cardiovascular events

ADAR: adenosine deaminase acting on RNA, ApoB: Apolipoprotein B, CVDs: cardiovascular diseases, LDL-C: Low-density lipoprotein cholesterol. Reference: Montasser ME. et al., 2021 Science 374(6572):1221-1227.

Well-defined development path for AX-1412

PRECLINICAL STAGE	EARLY CLINICAL	LATE CLINICAL
Preclinical models available with strong translatability into the clinic	*Early insight on safety and target engagement using validated biomarkers*	*Clinical programs with disease specific endpoints for regulatory approval*
Organoids models for CVD • Blood-derived myeloid cells and THP-1 cells • Cell-laden microtissue spheroids **Animal models** • The Apoe−/− mouse model **Proof of mechanism measures in animal models** • Serum lipid levels • Atherosclerotic lesion area • C-reactive protein (CRP) and Interleukin 6 (IL-6) • Endothelial function	**Programs with Phase 1 on healthy individuals** • Reduce potential signal-to-noise ratio as CVD patients have many comorbidities **General CVD biomarkers** • non-HDL-C • Triglycerides • Apoliprotein B **Target specific biomarkers** • LDL-C • Fibrinogen	**Primary endpoints** 1. All-cause mortality and fatal CVD events or 2. Composite endpoints (incl. fatal and non-fatal CVD events) **Secondary endpoints** • Could consider using biomarkers as surrogate endpoints to reasonably predict treatment effects on outcome.

Apoe: Apolipoprotein E, CVD: cardiovascular diseases, HDL-C: High-density lipoprotein cholesterol; LDL-C: Low-density lipoprotein cholesterol, THP-1: human monocytic cell line

AX-1412 early evidence generation approach on safety and target engagement

Phase 1 on healthy volunteers for CVD

Objectives

- Assess safety, tolerability, PK and PD of AX-1412 without interference by concomitant pathological conditions
- Establish target engagement of AX-1412

Trial design

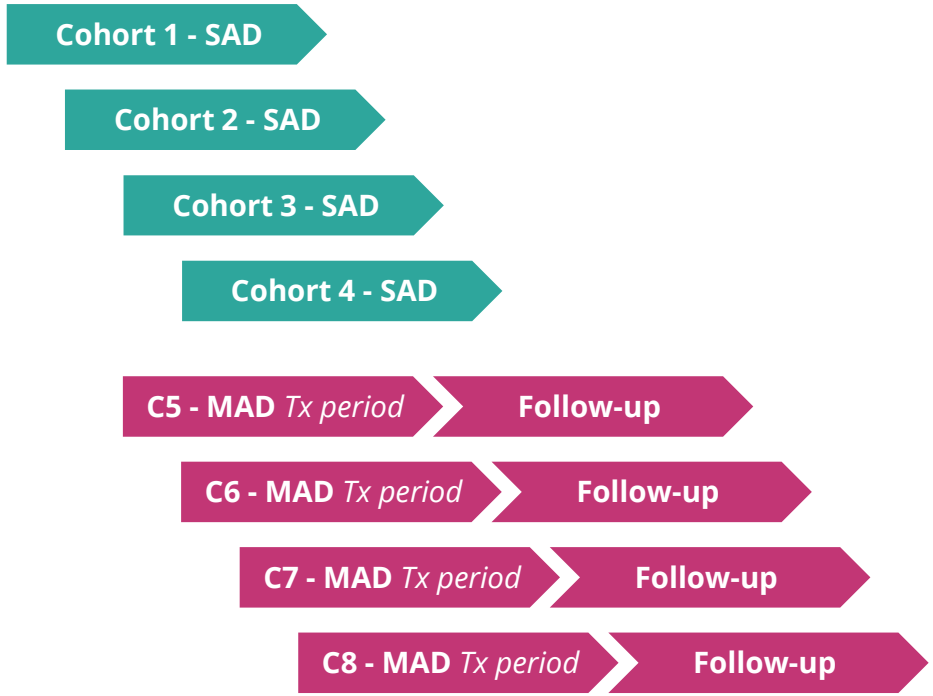
- Single and multiple dose ascending trial
- Single site trial

Endpoints will include

- Safety and tolerability
- Biomarkers measuring target engagement: LDL-C, fibrinogen, non-HDL-C, triglycerides, apolipoprotein B
- Measure RNA editing in circulating exosomes in plasma

Entry into clinical trials in late 2024 / early 2025

Preliminary study design



HDL-C: High-density lipoprotein cholesterol, LDL-C: Low-density lipoprotein cholesterol, MAD: multiple ascending dose, PD: Pharmacodynamic, PK: Pharmacokinetics, SAD: single ascending dose.

Summary and next steps for AX-1412

CVD remains the highest cause of disability and death

LDL-C and fibrinogen are two independent risk factors involved in cardiovascular diseases

AX-1412, a novel and unique approach

- Originated from human genetics
- Pleiotropic effects for cardiovascular protection
- Not suitable for knockdown technologies

A rigorous approach to increase probability of success

from preclinical to the clinical stage, including validated biomarkers

Next steps

- Generate new data on hit and lead selection
- Entry into clinical trials in late 2024 / early 2025

CVD: cardiovascular disease, LDL-C: Low-density lipoprotein cholesterol

ProQR development pipeline

	TARGET	DISCOVERY	NON-CLINICAL	CLINICAL	GUIDANCE	ESTIMATED POPULATION
PROQR PROGRAMS						
CHOLESTATIC DISEASES	**AX-0810** *for* **NTCP**				Entry into clinical trials in late 2024 / early 2025	~ 100K[1]
CARDIOVASCULAR DISEASES	**AX-1412** *for* **B4GALT1**				Entry into clinical trials in late 2024 / early 2025	~ 200M[2]
	AX-1005 *for CVD*					
METABOLIC DISEASES	**AX-2911** *for NASH*					~ 16M
	AX-0601 *for obesity and T2D*					~ 650M
	AX-9115 *for rare metabolic condition*					~ 20K
RARE NEURO DISEASES	**AX-2402** *for neurodegenerative condition*					~ 30K
OTHERS	*Multiple targets in discovery pipeline*					
PARTNERED PROGRAMS						
Lilly	*Initial **5** undisclosed targets*	*Progress undisclosed*				
	*Next **5** undisclosed targets*	*Progress undisclosed*				
	*Up to **5** potential additional targets*					

[1]Approximately 100K people affected with Primary Sclerosing Cholangitis and Biliary Atresia in US and EU5. [2]Approximately 200 million people suffer from too high a level of cholesterol in US and EU5. *SLC10A1* is the gene that encodes for NTCP protein.
CVD: Cardiovascular Diseases, NASH: Nonalcoholic steatohepatitis, T2D: Type 2 Diabetes.
References: Boonstra K, Beuers U, Ponsioen CY. J Hepatol. 2012 May;56(5):1181-1188; Karlsen TH, et al. J Hepatol. 2017 Dec;67(6):1298-1323; Dyson JK, et al. Lancet. 2018 Jun 23;391(10139):2547-2559; Sundaram SS, et al. Liver Transpl. 2017 Jan;23(1):96-109. Raghu VK, et al. Liver Transpl. 2021 May;27(5):711-718; NORD, 2019. Tsao CW, et al. Circulation. 2022;145(8):e153–e639. World Health Organization, World Gastroenterology Organization

Summary and Milestones

Daniel A. de Boer, Chief Executive Officer

Summary

Pipeline

- Initial programs focused on liver-originated diseases and will address Cholestatic Diseases targeting NTCP and Cardiovascular Diseases targeting *B4GALT1*
  - Initiation of clinical trials anticipated in late 2024/early 2025

Platform proof of concept

- Validation of Axiomer® across multiple preclinical in vitro, organoid, and in vivo models:
  - Up to 40% editing reported in the nervous system of mice in vivo leading to 26-fold change in protein function recovery
  - Up to 50% editing reported in the liver of mice in vivo
  - Up to 50% editing reported in the nervous system of NHP in vivo
- Broad applicability of Axiomer® demonstrated with ability to correct mutation, modulate protein function by altering protein function, disrupting post-translational modifications, and changing protein interaction in preclinical models

Multiple upcoming value-creating milestones

Platform

- Multiple platform updates over the next 12 months including NHP data in liver
- Plans to scale up discovery efforts ongoing
- Multiple scientific presentations and peer-reviewed publications in 2023 and 2024

Pipeline

- AX-0810 for cholestatic diseases
  - Presentation of non-clinical proof of concept data over next 12 months
  - Update on translational data over the next 18 months to enable progression into CTA
  - Entry into clinical trials in late 2024 / early 2025
- AX-1412 for CVD
  - Presentation of non-clinical proof of concept data over next 12 months
  - Update on translational data over the next 18 months to enable progression into CTA
  - Entry into clinical trials in late 2024 / early 2025

Partnerships and BD

- Eli Lilly $3.9 B partnership
  - Potential option exercise for expansion of deal to 15 targets, with $50 M opt-in payment to ProQR
  - Other milestone payments
- Potential additional multi-target discovery partnership
- Potential outlicensing of ophthalmology portfolio

Intellectual property

- Continued expansion of IP portfolio

Financial

- Cash position of €94.8M, plus $60.0M from Lilly partnership expansion at YE 2022 provides cash runway to mid 2026, beyond multiple clinical data readouts

Well positioned to advance Axiomer®

Science

- Deep understanding of basic science – ADAR, oligos
- Optimization of editing oligonucleotides (EONs) for therapeutic development

Axiomer® has broad applicability

- Large number of potential therapeutic applications
- *In vivo* POC established in nervous system, liver

Advancing toward the clinic

- Extensive translational and developmental expertise with oligo modality
- AX-0810 and AX-1412 initial pipeline targets

Leading IP position

- Axiomer® is protected by 10 published patent families
- Continuously investing in expanding IP estate

Strategic partnership strategy

- Lilly collaboration
- Selectively form additional partnerships
- Optionality and multiple value creating opportunities

Experienced leadership

- Deep RNA, corporate finance, and business development expertise across Management Team, Supervisory Board, and Scientific Advisory Board

Strong balance sheet

- 2022 YE cash €94.8M, plus $60.0M from Lilly partnership expansion
- Cash runway to mid-2026, excluding potential for additional BD-related upside

Q&A

Closing

Daniel A. de Boer, Chief Executive Officer

ProQR®

IT'S IN OUR RNA