

### 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.

### ProQR development pipeline

	TARGET	DISCOVERY	NON-CLINICAL	CLINICAL	NEXT MILESTONE	ESTIMATED POPULATION
DEVELOPMENT PIPELINE						
AX-0810 for Cholestatic diseases	NTCP				CTA filing in Q2 2025	~100K patients
AX-2402 for Rett syndrome	MECP2 R270X				Candidate selection in 2025	~5K patients
AX-1412 for Cardiovascular disease	B4GALT1				Scientific update in mid 2025	~200M patients
AX-2911 for MASH	PNPLA3				Candidate selection in 2025	~8M patients
DISCOVERY PIPELINE						
<b>AX-1005</b> for CVD	Undisclosed					~200M patients
AX-0601 for obesity and T2D	Undisclosed					~650M patients
AX-9115 for rare metabolic condition	Undisclosed					
AX-2403 for Rett syndrome	MECP2 R168X					~6K patients
AX-2404 for Rett syndrome	MECP2 R255X					~5K patients
AX-2405 for Rett syndrome	MECP2 R294X					~6K patients
AX-2406 for Rett syndrome	MECP2 R133H					
AX-3875 for rare metabolic & CNS disorder	Undisclosed					
<b>AX-4070</b> for rare CNS disorder	Undisclosed					
PARTNERED PIPELINE						
10 targets (option to expand to 15)	Undisclosed	Progress undisclosed				Lilly

<sup>&</sup>lt;sup>1</sup>Approximately 100K people affected with Primary Sclerosing Cholangitis and Biliary Atresia in US and EU5. <sup>2</sup>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: Trivedi PJ, et al. Clin Gastroenterol Hepatol. 2022 Aug;20(8):1687-1700.e4; Schreiber RA, et al. J Clin Med. 2022 Feb 14;11(4):999; Tsao CW, et al. Circulation. 2022;145(8):e153-e639. World Health Organization, World Gastroenterology Organization

### **Catalyst overview**

4 trial readouts expected in 2025-2026, cash runway into mid-2027

#### **AX-0810 for Cholestatic disease**

- CTA submission Q2 2025
- Top-line data Q4 2025

#### **AX-2402 for Rett Syndrome**

- Clinical candidate selection in 2025
- Anticipated trial start and top-line data in 2026

#### **AX-1412 for Cardiovascular disease**

Non-clinical data update in mid 2025

#### **AX-2911 for MASH**

- Clinical candidate selection in 2025
- Anticipated trial start and top-line data in 2026

#### **Partnerships**

- Opportunity to earn up to \$3.75B in milestones in the Lilly partnership
- Opportunity to receive a \$50 M opt-in fee from Lilly for expansion to 15 targets
- Opportunity for other strategic partnerships





#### INNOVATIVE ADAR-ENABLED RNA EDITING SCIENCE DRIVING ADVANCEMENT OF AXIOMER<sup>TM</sup>

supported by robust IP estate



### HIGH IMPACT STRATEGIC PARTNERSHIPS

Eli Lilly, Rett Syndrome Research Trust



#### PIPELINE WITH TRANSFORMATIVE POTENTIAL FOR DISEASES WITH HIGH UNMET MEDICAL NEEDS

Across rare and common liver and CNS disease



### EXPERIENCED TEAM DRIVING EXECUTION



### RUNWAY INTO MID 2027

€ 149.4 million cash and cash equivalents as of end of 2024, providing runway into mid-2027

**ProQR - Corporate Presentation** 

5

### ProQR's Axiomer™ ADAR journey since 2014

ProQR invents oligo mediated RNA Editing recruiting endogenous ADAR

2014

Key ADAR patents get granted in EU and US

2020-2023

ProQR pivots to solely focus on ADAR editing

2022

ProQR's ADAR patents win opposition cases filed by strawmen across the world

2023-2024

2014-2018+

ProQR files key patents that protect ADAR mediated RNA editing broadly 2015-2021

ProQR optimizes the ADAR platform in stealth 2021

ProQR and Eli Lilly enter into first 5 target partnership worth \$1.25B 2022

ProQR and Eli Lilly expand partnership to 10 targets worth ~\$3.9B 2023

ProQR
demonstrates
>50% editing in
CNS and liver in
NHP and
announces
pipeline

2024

- ProQR first in the field to report a disease relevant biomarker effect using Axiomer in NHP. Initial indication of good safety profile.
- Initial clinical validation of ADAR editing

2025

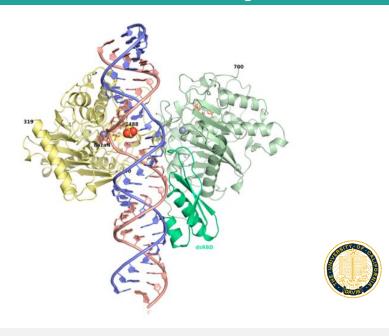
- Advance
   AX-0810
   NTCP program
   to clinical
   development
- Topline data Q4

ADARs: Adenosine deaminases acting on RNA, EONs: Editing oligonucleotides

## Axiomer™ EONs unlock cellular machinery potential to treat diseases

By attracting ADARs and allowing highly specific editing

#### ADAR (Adenosine Deaminase Acting on RNA)

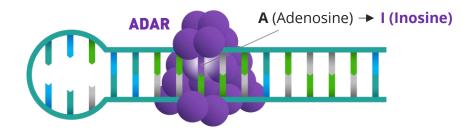


Enzyme that performs specific form of natural RNA editing, called **A-to-I editing.** During A-to-I editing an **A nucleotide** (adenosine) is changed into an I nucleotide (inosine)

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

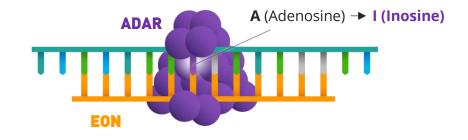
Natural ADAR editing (A-to-I)

RNA Double stranded



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

RNA+EON
Double
stranded



## 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

### Alter protein function or include protective variants

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



Variant resulting in a dominant negative effect

#### **Protein modulation**

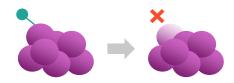


### Disrupt >400 different types of PTMs

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



Reduction of protein phosphorylation altering protein function



### Change protein interactions

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



Variant impacting protein interaction with sugar



## AX-0810 Program

Targeting NTCP to address cholestatic diseases unmet medical need at the root cause

## AX-0810 RNA editing therapy targeting NTCP for cholestatic diseases



Cholestatic diseases have high unmet medical need. Patients accumulate bile acids in liver leading to fibrosis and ultimately liver failure.



Initial indications are **Primary Sclerosing Cholangitis** affecting adults and Congenital **Biliary Atresia** affecting pediatrics early in life. Both conditions have no approved therapies and may require liver transplantation.<sup>1,2</sup>



- **Biliary Atresia** is projected to affect ~20,000 pediatric individuals in US and EU.
- **Primary Sclerosing Cholangitis** is projected to affect more than 80,000 individuals in US and EU.

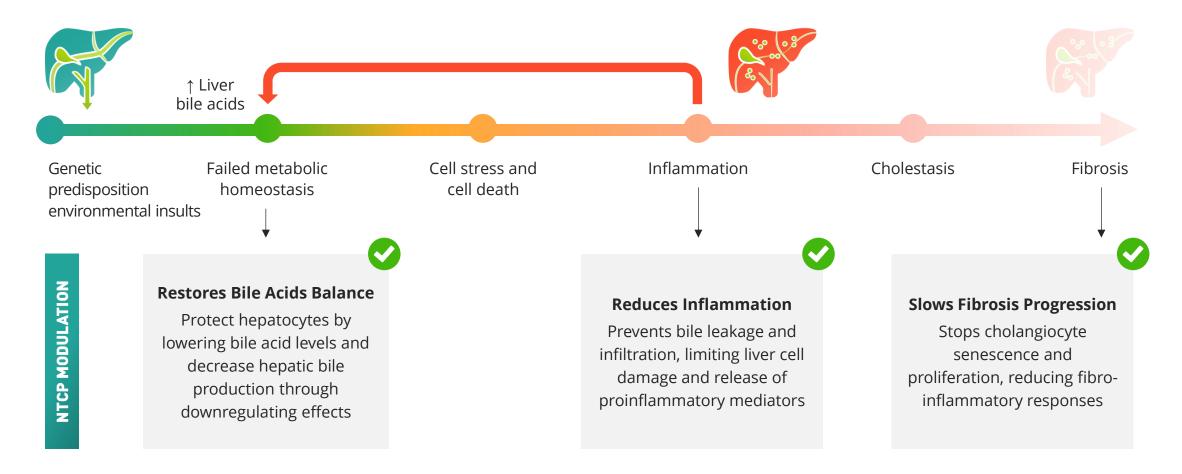


AX-0810 is a unique therapeutic approach leading to a potentially disease modifying therapy by targeting the NTCP channel which is responsible for majority of bile acid re-uptake in liver cells.



<sup>&</sup>lt;sup>1</sup>Trivedi PJ, et al. Clin Gastroenterol Hepatol. 2022 Aug;20(8):1687-1700.e4; <sup>2</sup>Schreiber RA, et al. J Clin Med. 2022 Feb 14;11(4):999

## NTCP modulation leads to positive effect on different mechanism involved in cholestasis



Zeng J, Fan J, Zhou H. Cell Biosci. 2023 Apr 29;13(1):77; Trauner M, Fuchs CD. Gut 2022;71:194–209; Halilbasic E, Claudel T, Trauner M. J Hepatol. 2013 Jan;58(1):155-68.

### NTCP variants reduced bile acids uptake into liver in health population research

Healthy population discovered with NTCP variants that reduces bile acids uptake into liver<sup>1-4</sup>



Ethnicity-dependent Polymorphism in Na+taurocholate Cotransporting Polypeptide (SLC10A1) Reveals a Domain Critical for Bile Acid Substrate Recognition\*

Received for publication, June 2, 2003, and in revised form, December 1, 2003 Published, JBC Papers in Press, December 2, 2003, DOI 10.1074/jbc.M305782200

#### Richard H. Hots, Brenda F. Leaket, Richard L. Roberts, Wooin Leet, and Richard B. Kimt\*

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The key transporter responsible for hepatic uptake of Bile acids, synthesized from the enzymatic catabolism of bile acids from portal circulation is Na\*-taurocholate cholesterol, are the major solutes in bile, essential for the cotransporting polypeptide (NTCP, SLC10A1). This maintenance of bile flow and biliary lipid secretion (1). In transporter is thought to be critical for the maintenance addition, an important mechanism for cholesterol homeostasis of enterohepatic recirculation of bile acids and hepatooccurs through its elimination in the form of bile acids. Indeed cyte function. Therefore, functionally relevant polymorphisms in this transporter would be predicted to have an important impact on bile acid homeostasis/liver function. However, little is known regarding genetic heterogeneity in NTCP. In this study, we demonstrate the presence of multiple single nucleotide polymorphisms in NTCP in populations of European, African, Chinese, and Hispanic Americans. Specifically four nonsynonymous single nucleotide polymorphisms associated with a sig-nificant loss of transport function were identified. Cell surface biotinylation experiments indicated that the altered transport activity of T668C ( $\rm Ile^{223} \rightarrow Thr)$ , a variant seen only in African Americans, was due at least in part to decreased plasma membrane expression. Similar expression patterns were observed when the variant alleles were expressed in HepG2 cells, and plasma membrane expression was assessed using immunofluorescence confocal microscopy. Interestingly the C800T exhibited a near complete loss of function for bile acid uptake yet fully normal transport function for the non-porter responsible for the observed Na\*-dependent uptake of bile acid substrate estrone sulfate, suggesting this position may be part of a region in the transporter critical and specific for bile acid substrate recognition. Accordingly, our study indicates functionally important polymorphisms in NTCP exist and that the likelihood of

Grants GM54724 and GM31304, by the NIGMS, National Institutes uncleotides specific to Ntcp, the expressed Na+dependent tau-Grants GM54724 and GM31304, by the NIGMS, National Institutes of Health Pharmacegenetics Research Network and Database (U01GM61974) under Grant U01 IHAS982, and by an NCI, National Institutes of Health United Variable (United States and Long Research Network) and the Company of the Company COORSION, SPANSONO, AND LEDGOWSEL THE COSES OF SUBSIGIATION OF PROCEEDINGS AND PROCESSING AND PR

de novo synthesis of bile acids from cholesterol is thought to modulate the release of pancreatic secretions and gastrointestinal peptides and activate enzymes required for the absorption of lipid-soluble vitamins (2, 3). Furthermore, their detergent properties assist solubilization of cholesterol and dietary fats in the intestine. Bile salts are efficiently reabsorbed in the small intestine and are returned to the liver via the portal circulation and resecreted into bile, thus forming an enterohepatic circuit (4). The efficient enterohepatic recirculation of bile acids is maintained by polarized expression of bile acid uptake and efflux transporters in the intestine and liver (4). Moreover taurine or glycine conjugates of bile acids tend to be polar and lar uptake and efflux (5).

In the liver, it is estimated that Na+-dependent transport → Phe) variant, seen only in Chinese Americans, pathways account for greater than 80% of the hepatic uptake of conjugated bile acids such as taurocholate (6-10). The transconjugated bile salts is Na+taurocholate cotransporting polypeptide (NTCP, SLC10A1) (11-14). This bile acid uptake transporter, whose function is coupled to a sodium gradient (15), is expressed exclusively in the liver and localized to the basolateral membrane of the hepatocyte (16). The human being carriers of such polymorphisms is dependent on NTCP gene encodes a 349-amino acid protein (14) and shares 77% amino acid sequence identity with rat Ntcp (17). Hagenbuch et al. (18) demonstrated that, when Xenopus laevis oocytes \* This work was supported by United States Public Health Service were coinjected with total rat liver mRNA and antisense oligo-

One potential source of altered NTCP function may be ge

d D Viruses and Bile Salts on Molecular Determinants on Sodium

He, \*-> Bijie Ren,\* Zhiyi Jing,\* Jianhua Sui,\* Wenhui Li\*

sting polypeptide (NTCP) is responsible for the majority of sodisonger true purpose and the second of the se reaction between NTCP and the pre-S1 domain of HBV large envelope character technical value and one previous atomass on fair value curvature ones of NTCP are independent or if they interfere with each other. Here Ans or A LLF are independent of at they interacte white calls other, there \[ \begin{align\*}
 \begin{align\*}
 & \text{plocks taurocholate uptake by the receptor; conversely, some bille }
 \] success nature-nomine uptake by the receptor; conversus, somewise ons of NTCP residues critical for bile salts binding severely impair s important for sodium binding also inhibit viral infection. The a magnetisms are securing or summing and animals area inection. The physics (SNP) found in about 9% of the East Asian population, a printing the property for the case Assau population, if you the ability to support HBV or HDV infection in cell culture. analy on the aboutly to suppose the v or the v successor in consumers, ical for HBV and HDV entry overlap with that for bile salts uptake by as nor rany and trave entry oversup bean tunn for one sours appeare to, ormal function of NTCP, and bile acids and their derivatives hold

is D virus (HDV), are important human pathogens. Available theris D varus (ttDv), are importain numan parinogens, avanance unter-dinically available for HDV infection. A liver bile acids transporter stinically available and stary intection. A liver une acids transport tical for maintaining homeostasis of bile acids serves as a func-CP-binding lipopeptide that originates from the first 47 amino late transport. Some bile salts dose dependently inhibit HBV donate transport, some one same unse dependently minute 110 v. s of NTCP critical for HBV and HDV entry overlap with that for s of ATLE CRITCHIOF FLDV and FLDV entry overlap with that for TCP-mediated HBV and HDV infection in relation to NTCP's their derivatives hold potential for development into novel

epG2 cells complemented with human or treeshrew NTCP. Respace cases storage measures were numerous recentrew to the re-acting a few amino acids of crab-eating monkey (amino acids aa] 157 to 165) or mouse NTCP (aa 84 to 87) with their human reparts converted these NTCPs to functional receptors for and HDV, respectively. Thus, HepG2 cells complemented uman NTCP provide a valuable and convenient in vitro cell e system for increasing our understanding of the mecha a of viral entry and for the development of novel antiviral

uman NTCP (SLC10A1) is a multiple-transmembrane pro hat is predominantly expressed at the basolateral membrane

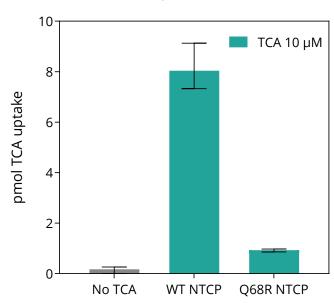
<sup>1</sup>Ho RH, et al. J Biol Chem. 2004 Feb 20;279(8):7213-22; <sup>2</sup>Vaz FM, et al. Hepatology. 2015 Jan;61(1):260-7; <sup>3</sup>Schneider AL, et al. Clin Res Hepatol Gastroenterol. 2022 Mar;46(3):101824; <sup>4</sup>Slijepcevic D, et al. Hepatology. 2018 Sep;68(3):1057-1069.

### NTCP modulation validated in vitro, vivo and clinic

Reducing liver bile acids toxic overload via NTCP modulation is a key driver for hepatoprotective effects

#### BAs uptake (TCA) in vitro

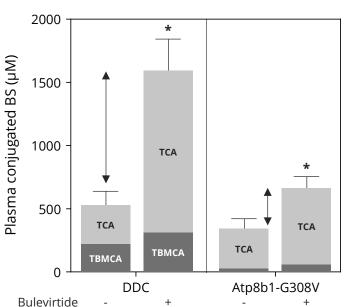
n=3, mean±SEM



Q68R NTCP variant leads to modulation of bile acids re-uptake

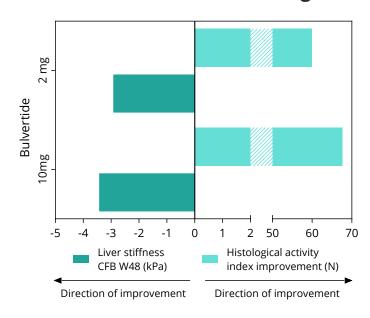
3-fold increase 2-fold increase

in conjugated BA in conjugated BA



NTCP modulation demonstrated effectivity in cholestatic disease model<sup>1</sup>

#### Bulevirtide 2 and 10 mg



Clinical PoC with bulevirtide in Ph3
Hepatitis D trial. **Improvement** occur in patients, **even without virologic response**<sup>2-4</sup>

Bulevirtide (Hepcludex) is a daily SC injected NTCP inhibitor approved for Hepatitis D. NTCP channel is a known transporter for bile acids and hepatitis virus from bloodstream to the liver.

1. Slijepcevic D, et al. Hepatology. 2018 Sep;68(3):1057-1069; 2. Wedemeyer H, et al. N Engl J Med. 2023 Jul 6;389(1):22-32; 3. Wedemeyer H, J Hepatol. 2024 Oct;81(4):621-629.; 4. Dietz-Fricke C, JHEP Rep. 2023 Mar 15;5(4):100686.

### NTCP modulation approach broadly validated

Reducing liver bile acids toxic overload via NTCP modulation is a key driver for hepatoprotective effects



#### **HUMAN GENETICS**

Healthy population discovered with NTCP variants that reduces bile acids uptake into liver<sup>1-3</sup>



#### **IN VITRO**

NTCP variant leads to an 8-fold decrease of bile acids re-uptake in vitro



#### IN VIVO

NTCP modulation demonstrated effectivity in mouse cholestatic disease model, with 2- to 3-fold change in conjugated bile acids<sup>4-5</sup>



#### **IN CLINIC**

Clinical PoC with bulevirtide in Ph3 Hepatitis D trial, for which liver improvement occur in patients, even without virologic response<sup>6-8</sup>



Vol. 279, No. 8, Issue of February 20, pp. 7213-7222, 2004 Printed in U.S.A.

Ethnicity-dependent Polymorphism in Na+taurocholate Cotransporting Polypeptide (SLC10A1) Reveals a Domain

Richard H. Hotes, Brenda F. Leaket, Richard L. Roberts, Wooin Leet, and Richard B. Kimt\*

From the †Division of Clinical Pharmacology, Departments of Medicine and Pharmacology, Vanderbilt Univer-From the Livission of Clinical Patermicology, Registrolistic of societies and Patermicology, Valuetrean University Department of Patellaries, Vanderbelt University Medical Center, Nashidis, Transcase 27223-2861, and the Master Department of Pathology, Vanderbilt University Medical Center, Nashidis, Transcase 27223-2861, and the Master of Science in Clinical Investigation Program, Vanderbilt University School of Medicine, Nashidis, Transcase 27223-2861, and the Spatial Control of Science in Clinical Investigation Program, Vanderbilt University School of Medicine, Nashidis, Transcase 27222-2861, and the Spatial Control of Science in Clinical Investigation Control of Science in Clinical Investigation, and Control of Science in Clinical Investigation Control of Science in Clinica

> occurs through its elimination in the form of bile acids. Indeed account for nearly half of the daily elimination of cholestero from the body (1). In the gastrointestinal tract, bile acids also erties assist solubilization of cholesterol and dietary fats in (4) The efficient enterohenatic recirculation of bile acids i maintained by polarized expression of bile acid uptake and efflux transporters in the intestine and liver (4). Moreover taurine or glycine conjugates of bile acids tend to be polar and hydrophilic, thus dependent on transporter proteins for cellular uptake and efflux (5). pathways account for greater than 80% of the hepatic uptake of

porter responsible for the observed Na -dependent untake of polypentide (NTCP 1 SLC10A1) (11-14). This bile acid uptak sporter, whose function is coupled to a sodium gradien (15) is expressed exclusively in the liver and localized to the eral membrane of the hepatocyte (16). The human NTCP gene encodes a 349-amino acid protein (14) and share strated that, when Xenopus laevis o were coinjected with total rat liver mRNA and antisense olig nucleotides specific to Ntcp, the expressed Na+dependent ta rocholate transport activity was reduced by 95%. This finding take of bile acids. Accordingly, the extent of its expression o patic circulation of bile acids and directly affect cellular signal ing pathways importantly involved in cholesterol hom

Bulevirtide (Hepcludex) is a daily SC injected NTCP inhibitor approved for Hepatitis D. NTCP channel is a known transporter for bile acids and hepatitis virus from bloodstream to the liver. 14 o RH, et al. | Biol Chem. 2004 Feb 20;279(8):7213-22; 2Vaz FM, et al. Hepatology. 2015 Jan;61(1):260-7; 3Schneider AL, et al. Clin Res Hepatol Gastroenterol. 2022 Mar;46(3):101824; 4Slijepcevic D, et al. Hepatology. 2018 Sep;68(3):1057-1069; 5Salhab A, et al. Gut. 2022 Jul;71(7):1373-1385; \*Wedemeyer H, et al. N Engl J Med. 2023 Jul 6;389(1):22-32; \*Wedemeyer H, J Hepatol. 2024 Oct;81(4):621-629.; \*Dietz-Fricke C, JHEP Rep. 2023 Mar 15;5(4):100686.

## Human genetics validates NTCP modulation as strategy for cholestatic disease

### LIVER WITH CHOLESTATIC DISEASE

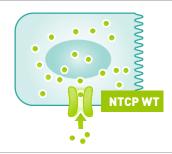
High concentration of bile acids in hepatocytes

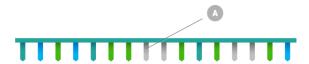


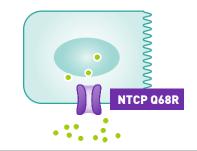
#### AX-0810 STRATEGY FOR DISEASED LIVER

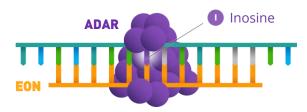
AX-0810 modifies the NTCP channel to limit bile acids uptake while preserving all other functions of the channel











- The AX-0810 program introduces a variant in individuals with cholestatic disease to lower bile acids concentration in hepatocytes by a single A-to-I change
- The AX-0810 program is designed to be a disease modifying treatment
  - To alleviate symptoms in PSC and BA
  - To limit inflammation and fibrosis linked to bile acid toxicity
  - To prevent or delay the development of cirrhosis, organ failure and need for transplant

BA, Biliary atresia; PSC, Primary Sclerosing Cholangitis

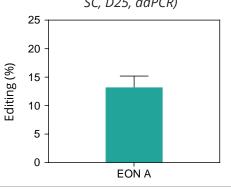
## EON mediated editing demonstrates consistent editing of NTCP and impact on biomarker in vivo

#### **EDITING EFFICIENCY**

# MICE in vivo

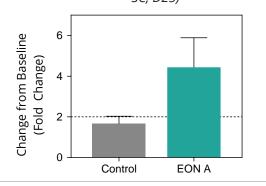
NHP in vivo

#### NTCP RNA Editing in Humanized Mice (N=4, 20mg/kg, 6 doses, GalNAc conjugation, SC, D25, ddPCR)



#### **PLASMA TOTAL BILE ACIDS**

#### Plasma TBA in Humanized Mice (N=4, 20mg/kg, 6 doses, GalNac conjugation, SC, D25)



#### NTCP RNA Editing in NHP (N=1, 1-4mg/kg, 4 doses, LNP formulation,

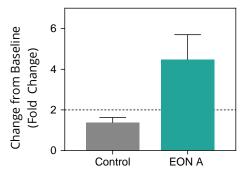
IV, up to D46, ddPCR)

20 
(%) 8u 10 
10 -

EON A

#### Plasma TBA in NHP

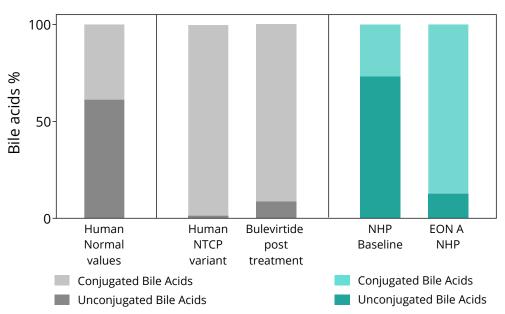
(N=1, 1-4mg/kg, 4 doses, LNP formulation IV, up to D39)



- EON A results in consistent editing data in humanized mouse model and NHP in vivo with approx. 15% editing reaching expected NTCP modulation
- Reaching >2-fold changes in biomarkers - expected impact on plasma bile acids levels following NTCP EON treatment

## PoC in NHP on bile acid profile and TUDCA elimination

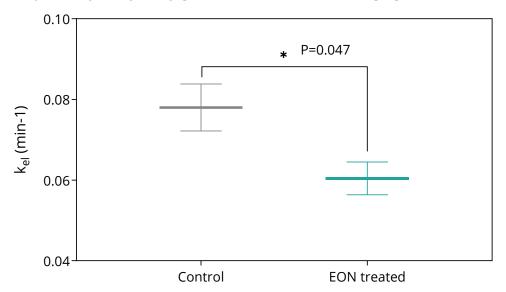
#### **Change in Plasma BA Profile**



- Conjugated bile acids are transported by NTCP back to the liver, change in plasma BA profile confirms NTCP specific modulation
- High confidence on NTCP EON treatment to positively impact BA toxic load in the liver

#### **TUDCA elimination rate from plasma in NHP**

Exploratory study, early generation EON, n=5-7, 10mg/kg, 4 doses, SC, D51



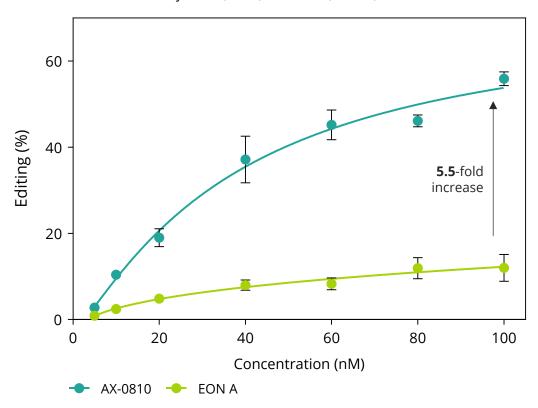
- TUDCA is a Tauro-conjugated bile acid specifically transported by NTCP from the plasma to the liver
- Decrease in TUDCA plasma clearance kinetics further confirm NTCP target engagement for EON treated NHP

Conditions in the NHP experiment on the left: N=1, 1-4mg/kg, 4 doses, LNP formulation, IV, up to D42, LC-MS/MS. Mao F, et al. J Biol Chem. 2019 Aug 2;294(31):11853-11862; Haag M, et al. Anal Bioanal Chem. 2015 Sep;407(22):6815-25.; Wedemeyer H, et al. N Engl J Med. 2023 Jul 6;389(1):22-32.

## AX-0810 clinical candidate selected with enhanced potency and stability profile

### AX-0810 clinical candidate has an enhanced potency profile over EON A in PHH

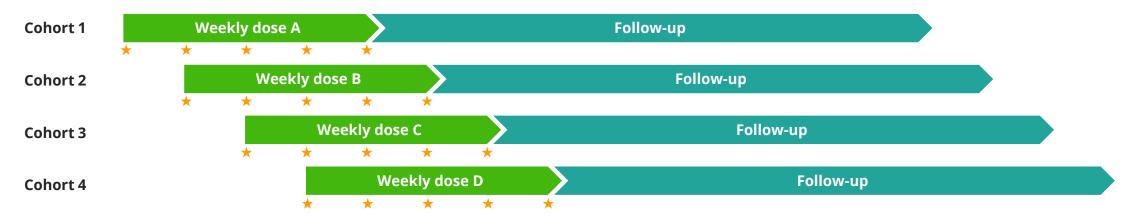
Transfection, n=3, 72 hours, dPCR, mean±SEM



- AX-0810 clinical candidate is a GalNAc conjugated EON
- 5.5-fold increase in potency over early generation NTCP editing oligonucleotide
- Improved stability profile in vitro
- Confirmed class safety, with no hepatotoxicity or immunostimulatory score

## First in human trial of AX-0810 to establish target engagement

#### Integrated single/multiple ascending dose study design



#### **Treatment**

AX-0810 GalNAc conjugated editing oligo-nucleotide

#### **Objectives**

- Confirm target engagement as measured by biomarkers
- Assess safety, tolerability, and PK of AX-0810

#### **Trial design**

- Combined single and multiple ascending dose
- ≥60 heathy volunteers, 4 weeks dosing phase followed by 12 safety weeks follow-up
- 5 weekly subcutaneous injections
- Baseline and placebo-controlled design
- Standardized conditions for assessment of bile acids at multiple timepoints
- DMC safety reviews before proceeding to next dose and dose escalation

#### **Key endpoints**

- Change in bile acids levels and profile in plasma and urine, liver biomarkers
- Circulating RNA as exploratory endpoint

CTA submission in Q2 2025

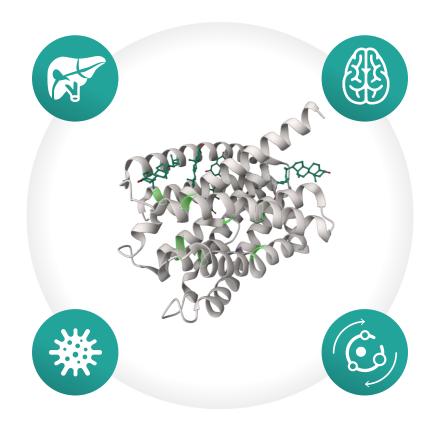
Top-line data in Q4 2025

## NTCP and bile acids are involved in a variety of therapeutic areas

Providing opportunity across multiple indications

#### **Cholestatic diseases**

- Primary Sclerosing Cholangitis (PSC)
- Biliary Atresia
- Primary Biliary Cholangitis (PBC)
- Alagille syndrome
- Dubin-Johnson Syndrome
- Progressive Familial Intrahepatic Cholestasis (PFIC)
- Drug-Induced Cholestasis
- Alcoholic Liver Disease
- Secondary Biliary Cirrhosis
- Rotor syndrome
- Neonatal cholestasis



#### Neurological diseases

- Multiple Sclerosis
- Amyotrophic Lateral Sclerosis
- Neurological diseases
- Epilepsy
- Parkinson's Disease

#### Infectious disease

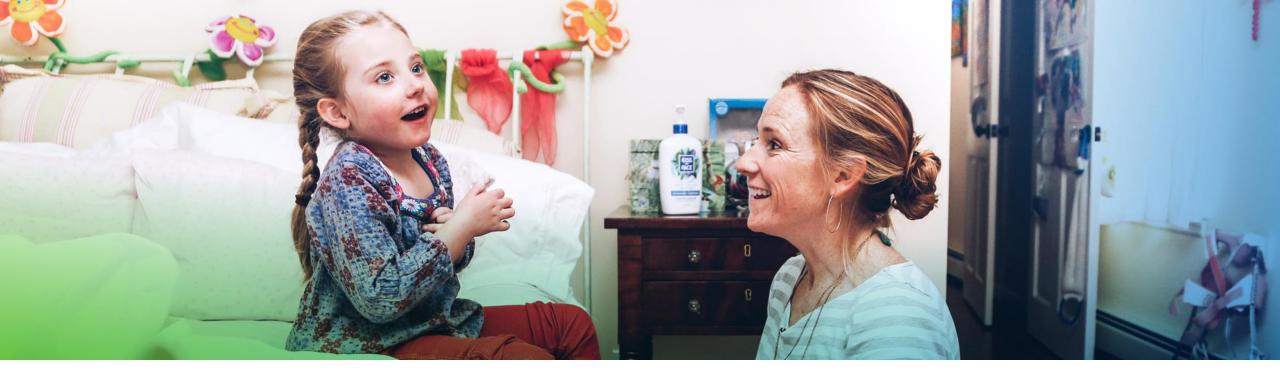
- Parasitic Infections
- Sepsis-Associated Cholestasis
- Viral Hepatitis: Hepatitis A, B, C, D, E

#### **Metabolic diseases**

- Hyperlipidemia
- Lysosomal
- Hypertension
- storage diseases

MASH

- Hyper-
- Obesity
- cholesterolemia
- Diabetes
- ASCVD



## AX-2402 Program

Targeting MECP2 to restore protein functionality in Rett Syndrome, a severe neurodevelopmental disorder

## AX-2402 RNA editing therapy targeting MECP2 for Rett Syndrome





Rett Syndrome is a **devastating and progressive neurodevelopmental disorder** caused by variants in the transcription factor Methyl CpG binding protein 2 (*MECP2*). There is a **high unmet need for a disease modifying therapy**.



Nonsense variants lead to **severe phenotypes.** They represent more than one third **of Rett Syndrome** cases and are projected to affect **20,000 individuals** in US and EU.<sup>1,2</sup>



Rett Syndrome is **not a neurodegenerative disorder** and restoring levels of the MECP2 protein has shown to **reverse symptoms** in mice.<sup>3</sup>



Axiomer has the potential to **restore the precise level of MECP2 protein regulatory function**, which is lacking in Rett Syndrome, and become a disease modifying therapy.



Rett Syndrome Research Trust partnership includes \$9.2 M in funding; collaboration established in January 2024, expanded in December 2024



<sup>1</sup>Krishnaraj R, et al. Hum Mutat. 2017 Aug;38(8):922-93; <sup>2</sup>RSRT 2023 conference; <sup>3</sup>Guy J, et al. Science. 2007 Feb 23;315(5815):1143-7.

## Axiomer<sup>™</sup> has the potential to restore physiological levels of functional MECP2

AX-2402 correcting MECP2 R270X into WT-like R270W

GGGGC**C>UGA**AAGCCG

**EXON 3** 

**EXON 4** 



#### **RETT syndrome**

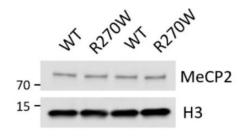
Postnatal microcephaly, stereotypic hand movements, ataxia, abnormal breathing, and growth retardation, social withdrawal, loss of speech, seizures

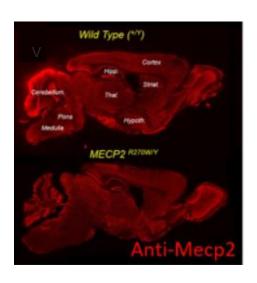


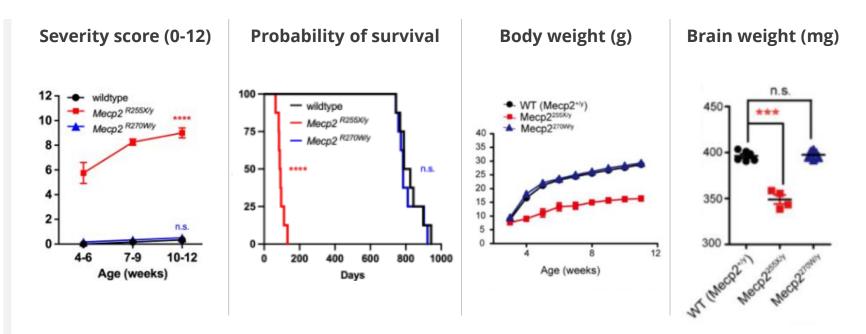
#### WT like phenotype

- MeCP2 protein restoration/recovery
- MeCP2 R270W (Arg > Trp) mouse model indistinguishable from wild type mice

### R270W variant demonstrates wild-type like profile







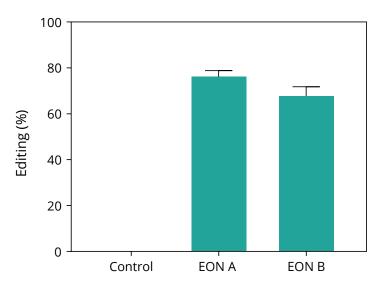
AX-2402 can restore physiological levels of functional MECP2 potentially reverting Rett syndrome into a WT like phenotype<sup>1</sup>

<sup>1</sup>Colvin, S. (2023) thesis. Massachusetts Institute of Technology. Figures adapted from: Colvin, S. (2023) thesis. Massachusetts Institute of Technology

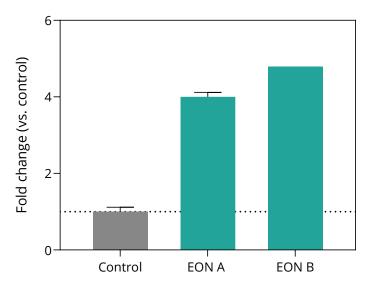
## EON mediated editing in patient's cells increases mRNA levels and restores protein expression

PTC recoding leading to absent NMD mediated RNA degradation

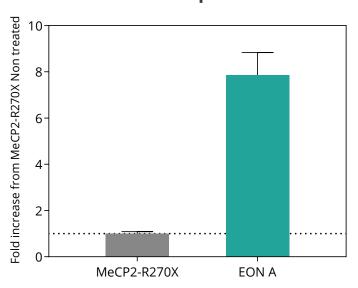




#### **MECP2 RNA levels**



#### **R270W MECP2 protein levels**



Up to 80 % editing of R270X MECP2 in patient fibroblasts

Increased MECP2 RNA levels due to PTC recoding and NMD inhibition

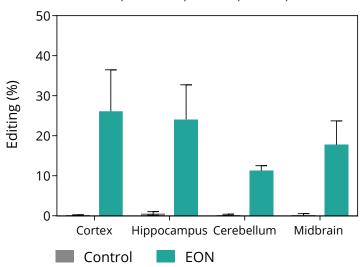
Increased R270W MECP2 protein levels

EON, Editing oligonucleotide; NT, Non-treated; TF, transfection, Conditions panel on the left and middle: 100 nM EON, transfection, 48h, N=2, mean±SEM. Conditions panel on the right: MeCP2-R270X-NanoLuc activity; 100 nM EON, transfection, 48h, N=8, mean±SEM.

## Consistent CNS editing demonstrated across species

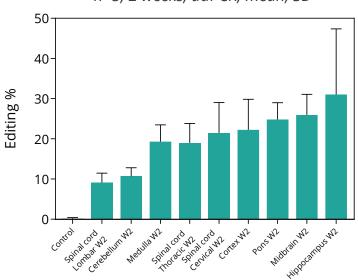
Mice in vivo

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



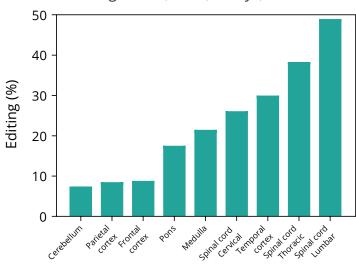
#### Rat in vivo

*ICV,* 500μg, APP, single dose, n=5, 2 weeks, ddPCR, mean, SD



#### NHP in vivo

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

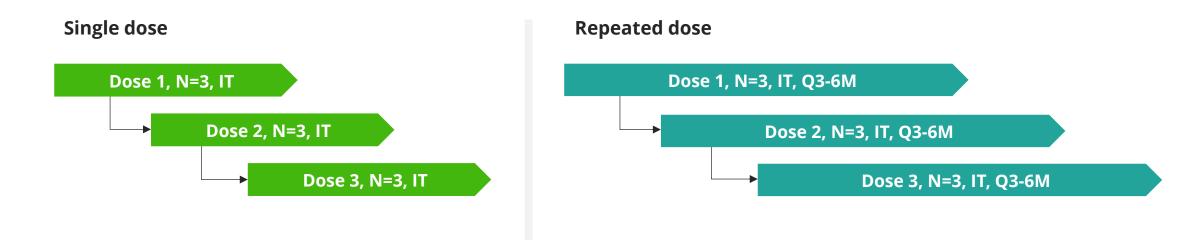


- Up to 40% editing in vivo leading to 26-fold change in protein function recovery in brain tissues of interest at 4 weeks with a single dose in mice model
- In rat, Axiomer EONs demonstrated up to 50% editing *in vivo* with sustained editing between W2 and W4 after single dose
- Up to 30% RNA editing reported in brain and approx. 50% in spinal cord in NHP in vivo

<sup>\*</sup> Data of 2 NHPs not analyzable due to human error during injection procedure.



### Preliminary clinical trial design



- Preliminary Phase 1/2 SAD & MAD design
- Up to 18 subjects with the R270X mutation
- Primary objective: safety, tolerability and pharmacokinetics
- Secondary objectives: target engagement and biomarkers
- Financially supported by \$8.2 M funding provided by Rett syndrome Research Trust
- Clinical candidate selection in 2025
- Top-line data expected in 2026



## AX-1412 Program

Targeting B4GALT1 to reduce the risk of cardiovascular diseases

## AX-1412 RNA editing therapy targeting B4GALT1 for cardiovascular diseases



Leading causes of death in the world ~18 million people die from CVDs every year (32% of all global deaths) Despite therapies, the unmet medical need remains.



AX-1412 is designed to provide people with a protective genetic variant of B4GALT1 that is associated with **36%¹** reduction in the risk of cardiovascular disease.

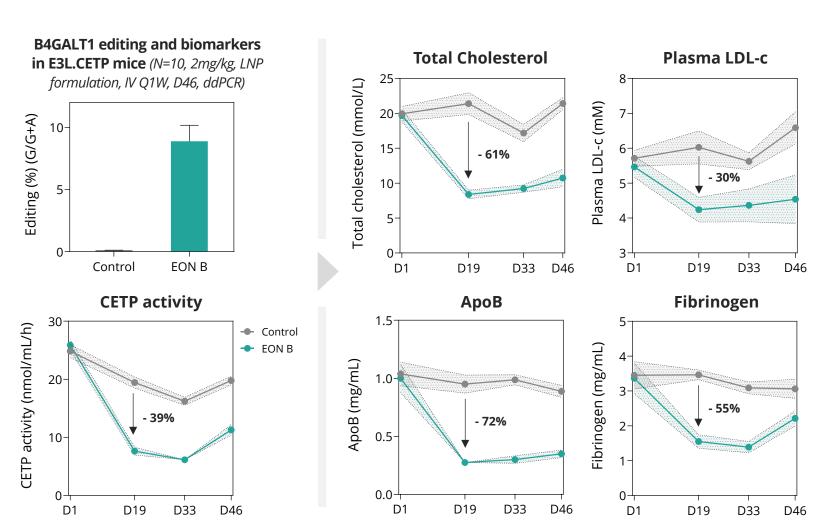


AX-1412 may become a **stand-alone cardiovascular therapy** that may also work **synergistically with standard of care** to further reduce risk of CVDs.



<sup>1</sup>Montasser ME, et al. Science, 2021 Dec 3:374(6572):1221-1227

## EON-mediated editing of B4GALT1 leads to meaningful effect on key biomakers in E3L.CETP Mice



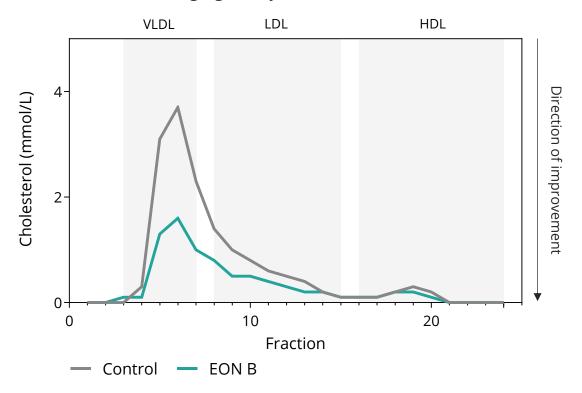
Following treatment
with EON B, a marked
reduction in total
cholesterol, ApoB, and
LDL-c by observed
already at Day 19
confirms our
approach to address
cardiovascular
diseases

## **B4GALT1 EON leads to a positive shift in lipoprotein profiles**

Specifically targeting atherogenic lipoproteins

### Impact on lipoprotein profile following editing of B4GALT1 in E3L.CETP mice

(N=10, 2mg/kg, LNP formulation, IV Q1W, D46)



- Treatment with EON B significantly decreases VLDL and LDL cholesterol compared to control
- These lipoproteins are associated with increased cardiovascular risk due to their role in atherosclerotic plaque formation
- HDL cholesterol which supports reverse cholesterol transport and is associated with reduced cardiovascular risk, remains unchanged





## **EON-MEDIATED RNA EDITING OF B4GALT1**

leads to the required reduction in galactosylation, reflecting the human genetics observed effect



## LNP-DELIVERED EON EDITING OF B4GALT1

leads to editing and meaningful changes in biomarker effect on LDLC, CEPT, cholesterol and fibrinogen in an industrystandard in vivo disease model



## FURTHER OPTIMIZATION OF A GALNAC DELIVERED EON ONGOING

to achieve a TPP desirable for CVD



## UPDATE ON THE GALNAC OPTIMIZATION EFFORTS

expected in mid 2025



## **AX-2911 Program**

Targeting PNPLA3 to address unmet medical needs in MASH

AX-2911 RNA-editing therapy to address Metabolic dysfunction associated steatohepatitis (MASH)



MASH and subsequent stages of liver disease are very prevalent and still on the rise worldwide. MASH individuals have a high unmet medical needs due to the progressive nature of the disease (cirrhosis and hepatocellular carcinoma) and limited therapeutic options available<sup>1</sup>



PNPLA3 (patatin-like phospholipase domain-containing 3) I148M is a variant **commonly reported** in the MASH population worldwide (20-60% of the patients) and is known as **associated risk factor**.<sup>2,3</sup> Approximately 8 million individuals in US and EU are homozygous for the 148M variant.



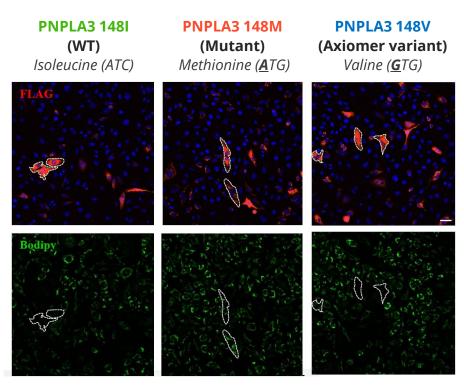
Axiomer EONs have the potential to change the Methionine into a Valine bringing the **PNPLA3 protein back to a WT-like functional conformation**.



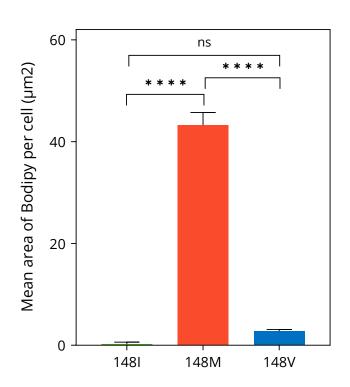


## Axiomer™ creates a PNPLA3 protein with WT-like functionality

1481 and 148V reports equivalence in lipid droplet sizes



Hoechst (nuclei), Bodipy (Lipids) and M2 anti-flag (PNPLA3)



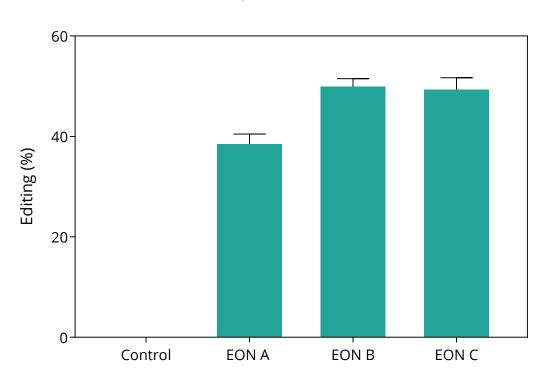
- The wild-type 148I shows smaller lipid droplets, reflecting normal lipid metabolism
- The 148M variant induces significantly larger lipid droplets, consistent with its pathogenic role in lipid metabolism disorders
- The corrected variant 148V results in wild-type like droplet sizes, suggesting a corrective effect on lipid accumulation, similar to 148I

Treatment conditions: HeLa cells, plasmid, transfection, 250uM linoleic acids, 24h, cell lipase activity by IF One-way ANOVA, \*\*\*\*, P<0.0001; Mean, SEM.

## EON mediated PNPLA3 editing leads to over 50% RNA editing and change in lipid droplet

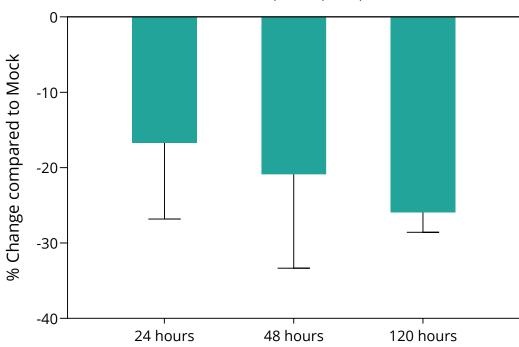
#### **Editing of PNPLA3 in PHH**

100nM EON, transfection, 72h, dPCR, mean, SEM, n=3



### **Change in intracellular lipid droplets post PNPLA3 148V EON treatment**

Bodipy/DAPI stainings, 5μM EON, transfection, exposure to linoleic acid, mean, SEM, n=2







## CLINICAL CANDIDATE SELECTION

Final optimization of AX-2911 EONs ongoing for clinical candidate selection in 2025



#### **DEVELOPMENTAL ACTIVITIES**

to start in 2025



## SUBCUTANEOUS GALNAC-DELIVERY

expected with 3-6 monthly dosing interval



#### **CLINICAL TRIAL**

to start in 2026

**ProQR - Corporate Presentation** 

37

## Well positioned

to advance Axiomer™





#### **CLINICAL TRIAL RESULTS EXPECTED**

across 4 trials in 2025 and 2026

- Clinical PoC data of NTCP trial in 2025
- Up to 4 clinical trials with data readouts in 2025/2026



#### **RICH DISCOVERY PIPELINE**

with potential for broad pipeline expansion

- Large number of potential therapeutic applications in discovery pipeline
- Broad applicability beyond current discovery pipeline



#### **LEADING IP POSITION**

- Axiomer™ is protected by >20 published patent families
- Continuously investing in expanding IP estate



### VALIDATING STRATEGIC PARTNERSHIPS

- Eli Lilly collaboration valued up to \$3.9B, with opportunity for near-term milestones
- Rett Syndrome Research Trust cofinancing of AX-2402 program
- Selectively form additional partnerships



#### STRONG BALANCE SHEET

- € 149.4 million cash and cash equivalents as of end of 2024
- Cash runway to mid-2027, excluding potential for additional BD-related upside





## Resource slides



# HOW DOES ADAR WORK?

Explained in 5 minutes

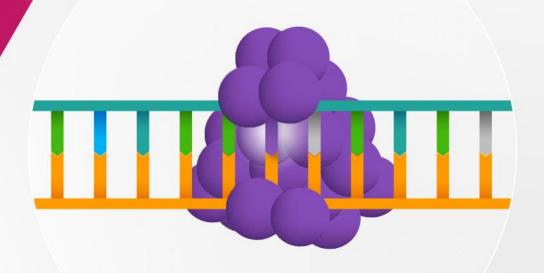






# WHAT IS AXIOMER™?

Explained in 5 minutes





### **ProQR Leadership Team**

#### **Management Team**



**Daniel de Boer** Founder & CEO, Board Executive Director



**Gerard Platenburg** Chief Scientific Officer, Board Executive Director





PHARMING



**Dennis Hom** Chief Financial Officer

ProQR THERAPEUTICS













Cristina Lopez Lopez, MD, PhD

Chief Medical Officer Johnson Johnson





#### **Board of Directors**



James Shannon, MD Chair







**Alison Lawton** 



#### **Key Advisors**



John Maraganore, PhD Board advisor

2 Alnylam



Begoña Carreño







Martin Maier, PhD







Peter A. Beal, PhD ProQR Chief ADAR Scientist





**Bart Filius** 





**Dinko Valerio** 





Phillip D. Zamore, PhD Scientific Advisory Board





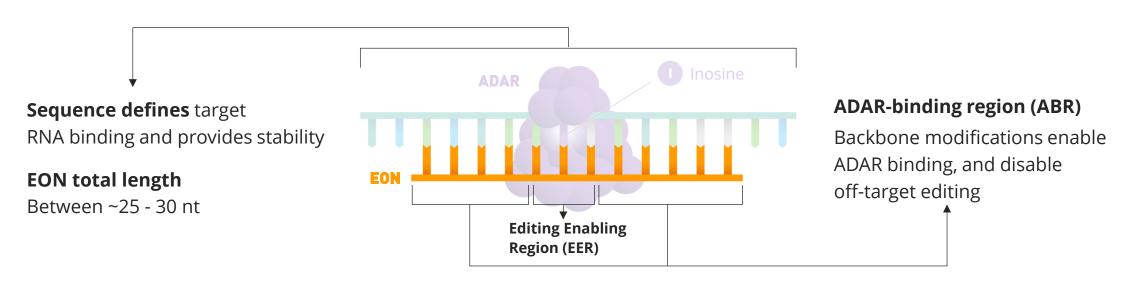


**Theresa Heggie** 

2 Alnylam FREELINE



## Driving the development of optimized EONs for therapeutic use



#### 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

## Leading IP supporting ADAR-mediated RNA editing platform technology

- Axiomer™ IP strategy commenced in 2014 with first patent application filings
- Currently 25 published patent families, comprising 33 national/regional patents
- Axiomer™ IP portfolio is constantly expanding
- Oppositions/appeals and several Third-Party Observations have been filed against a variety of applications and patents in the Axiomer™ IP portfolio, all by strawmen

## ProQR Axiomer<sup>™</sup> leading IP estate for ADAR-mediated RNA editing

- ProQR's Axiomer™ IP contains 3 early RNA editing platform patent families covering single-stranded oligonucleotides that recruit endogenous ADAR
- Oppositions/appeals and Third-Party Observations have been filed throughout these three patent families
- First (2014): oligonucleotides with a complementary (**targeting**) and a stem-loop (**recruiting**) portion
- Second (2016): oligonucleotides without a stem-loop structure but with one or more mismatches and chemical modifications
- Third (2016): oligonucleotides **without a stem-loop structure** but with specific chemical modifications in the '**Central Triplet**'

## Overview of Axiomer™ related patents

Docket	Priority	Feature	Status	Remarks
1 (0004)	17DEC2014	Targeted RNA Editing using endogenous ADARs	Granted AU BR <u>CA CN EP</u> IL IN <u>JP</u> NZ <u>US US</u> ZA	Platform IP
2 (0013)	22JUN2016	Short EONs with wobble and/or mismatch base pairs	Granted <u>AU</u> IL <u>JP KR US US US</u>	Platform IP
3 (0014)	01SEP2016	Chemically modified short EONs	Granted AU <u>CN EP</u> IL <u>JP KR</u> NZ <u>US US US</u> ZA	Platform IP
4 (0016)	19JAN2017	EONs + protecting SONs (heteroduplex formation)	Granted <u>US</u>	Platform IP
5 (0023)	18MAY2018	PS linkages / chiral linkages (e.g., PS, PN)	Published	Platform IP
6 (0025)	28JAN2019	Editing of PTC in exon 61 USH2A	Published	Target
7 (0026)	11FEB2019	Phosphonacetate linkages / UNA modifications	Published	Platform IP
8 (0029)	03APR2019	MP linkages	Published	Platform IP
9 (0031)	24APR2019	Editing inhibition	Published	Platform IP
10 (0032)	13JUN2019	Benner's base (dZ)	Published Granted CN ZA	Platform IP – with UC Davis (P Beal)
11(0035)	23DEC2019	Editing in exon 35 of ABCA4 for Stargardt disease	Published	Target
12 (0039)	23JUL2020	Split EONs	Published	Platform IP
13 (0045)	14FEB2022	PCSK9	Published	Target
14 (0046)	15JUL2022	5'-GA-3' editing	Published	Platform IP – with UC Davis (P Beal)
15 (0048)	15JUL2022	diF modification	Published	Platform IP
16 (0051)	21OCT2022	Heteroduplex Editing Oligonucleotide (HEON) complexes	Published	Platform IP
17 (0052)	24NOV2022	HFE	Published	Target
18 (0053)	09DEC2022	B4GALT1	Published	Target
19 (0054)	01DEC2022	ALDH2	Published	Target
20 (0055)	20JAN2023	AG1856 + (H)EONs	Published	Platform IP – with FU Berlin (A Weng)
21 (0057)	20FEB2023	ANGPTL3	Published	Target
22 (0058)	24MAR2023	KCC2	Published	Target
23 (0059)	24MAR2023	PNms linkages	Published	Platform IP
24 (0060)	27MAR2023	NTCP	Published	Target
25 (0061)	16JUN2023	RELN	<u>Published</u>	Target

### ProQR Axiomer™ IP

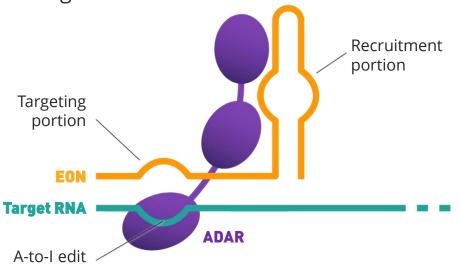
#### Broad coverage

- Axiomer<sup>™</sup> patent claims are broad and cover:
  - Any type of chemically modified oligonucleotide aimed at RNA editing of any possible target and any possible disease using endogenous ADAR
  - Specific targets, including SERPINA1 (A1AT deficiency), IDUA (Hurler syndrome),
     LRRK2 (Parkinson's disease)
  - Oligonucleotides with chirally-controlled linkages
  - Oligonucleotides with all sorts of chemistries (also in the 'Central Triplet'), including **DNA**
- To note: claims directed to chemically modified oligonucleotides do not cover viral delivery of the oligonucleotide

## Overview of key claims - 1

Granted claims in the 1<sup>st</sup> Axiomer<sup>™</sup> patent family relate to (chemically modified) oligonucleotides that comprise:

- A targeting portion for binding to a target RNA incl. target adenosine
- A recruitment portion (hairpin structure) for recruiting endogenous ADAR to edit the target adenosine



EP 3 234 134 B1	Granted; appeal pending
<u>US 10,676,737</u>	Granted
<u>US 11,781,134</u>	Granted

Claim 17 (US 11,781,134):

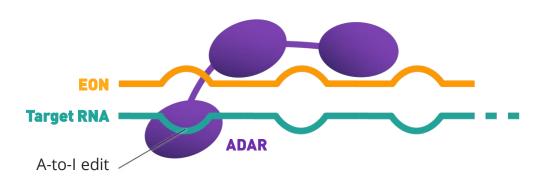
A method for making a change in a target RNA sequence in a human cell, comprising the steps of:

- introducing into the cell an oligonucleotide construct that is sufficiently complementary to bind by nucleobase pairing to the target RNA sequence, wherein the target RNA sequence comprises a target adenosine;
- allowing the formation of a double-stranded structure of the oligonucleotide construct with the target RNA sequence upon base pairing;

- allowing the double-stranded structure of the oligonucleotide and the target RNA sequence to recruit an hADARI or hADAR2 enzyme naturally present in the cell;
- allowing the hADARI or hADAR2 enzyme to perform deamination of the target adenosine to an inosine in the target RNA sequence.

## Overview of key claims - 2

Granted claims in the 2<sup>nd</sup> Axiomer™ patent family relate to oligonucleotides that do **not** have a hairpin structure, but instead have one or more wobbles and/or mismatches, and chemical modifications in the base, ribose sugar and/or linkage to increase stability and are still able to recruit **endogenous** ADAR to edit the target adenosine.



<u>US 10,988,763</u>	Granted
<u>US 11,649,454</u>	Granted
<u>US 12,018,257</u>	Granted

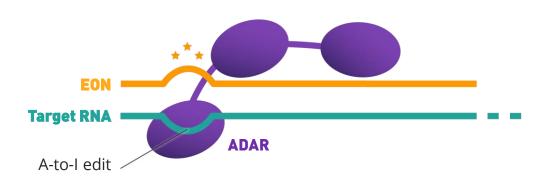
#### Target-specific claims are directed to:

- An AON capable of forming a double stranded complex with a target RNA in a cell, wherein: the target RNA encodes alpha1- antitrypsin (A1AT), LRRK2, or the target RNA is encoded by the IDUA gene
- The AON is complementary to a target RNA region comprising a target adenosine
- The AON comprises one or more nucleotides with one or more sugar modifications
- The AON does <u>not</u> comprise a portion that is capable of forming an intramolecular

- stem-loop structure that is capable of binding an ADAR enzyme
- The AON is shorter than 100 nucleotides
- The AON optionally comprises 1, 2, 3, 4, 5, 6, 7, 8, 9 or 10 mismatches, wobbles and/or bulges with the complementary target RNA region, and, wherein formation of the double stranded complex between the AON and the target RNA results in the deamination of the target adenosine by an ADAR enzyme present in the cell

## Overview of key claims - 3

Granted claims in the 3<sup>rd</sup> Axiomer™ patent family relate to oligonucleotides that do **not** have a hairpin structure, but have **chemical modifications** in the base, ribose sugar and/or linkage to increase stability and are still able to recruit **endogenous** ADAR to edit the target adenosine.



EP 3 507 366 B1	Granted; appeal pending
<u>US 10,941,402</u>	Granted
<u>US 11,851,656</u>	Granted
US 12,203,072	Granted

Claim 1 (US 11,851,656):

An antisense oligonucleotide (AON) comprising a Central Triplet of 3 sequential nucleotides, wherein

- the AON is capable of forming a double stranded complex with a target RNA molecule in a cell comprising a target adenosine;
- the nucleotide directly opposite the target adenosine is the middle nucleotide of the Central Triplet;
- 1, 2 or 3 nucleotides in the Central Triplet comprise a sugar modification and/or a base modification to render the AON more stable and/or more effective in inducing

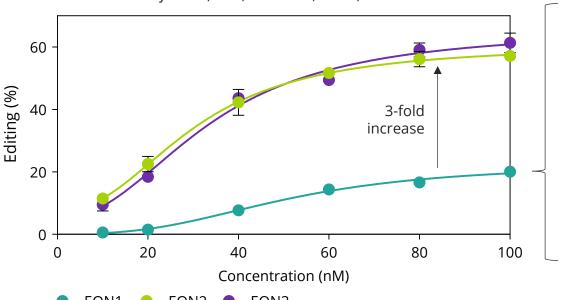
- deamination of the target adenosine; with the proviso that the middle nucleotide does <u>not</u> have a 2'-O-methyl modification;
- the AON does <u>not</u> comprise a 5'terminal O6-benzylguanosine;
- the AON does <u>not</u> comprise a portion that is capable of forming an intramolecular stem-loop structure that is capable of binding a mammalian ADAR enzyme present in the cell; and
- the AON can mediate the deamination of the target adenosine by the ADAR enzyme.

## Axiomer™ EON treatment led to NTCP Q68R variant in WT hepatocytes

Editing of NTCP RNA modulates BAs reuptake in a dose dependent fashion

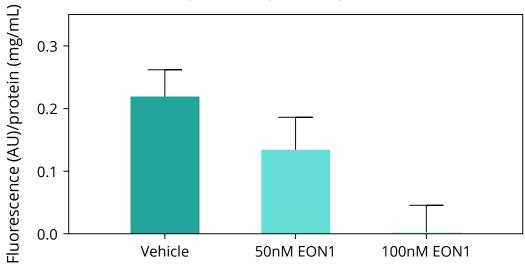
### **EONs targeting NTCP RNA optimization in PHH**

Transfection, n=3, 72 hours, dPCR, mean±SEM



NTCP-mediated BAs uptake in HepaRG cells with Axiomer EON treatment

*n*=3, 50-100*nM*, 72 hours, mean±SEM



Leveraging expertise in EONs optimization, including adjustment of sequence and chemistry, lead to increased potency of EONs targeting NTCP RNA.

Early generation of EONs (EON1) induces a dose-response inhibition of BAs in vitro confirming its mediation by NTCP

BAs: Bile acids, NTCP: Na-taurocholate cotransporting polypeptide, BAs mentioned in this experiment are specifically Tauro-nor-THCA-24-DBD. SLC10A1 is the gene that encodes for NTCP protein. Reference: Cnubben, N. et al. (2024) ASGCT 27th Annual meeting abstracts, Molecular Therapy, Volume 32, Issue 4, 1 – 889 (Abstract 705, p. 355)

