Development of an Investigational RNAi Therapeutic Targeting Glycolate Oxidase for the Treatment of Primary Hyperoxaluria Type 1

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September 5, 2015
Statement of Conflict of Interest: The speaker is an employee of Alnylam Pharmaceuticals.
RNAi Therapeutics

A New Class of Innovative Medicines

- Harness the natural pathway, mediated by small interfering RNA or “siRNA”, to therapeutically silence any gene
  - Distinct mechanism of action vs. other drug classes
    - Catalytic gene silencing
- GalNAc-siRNA conjugates for efficient delivery to hepatocytes through ASGPR
  - Permits subcutaneous dosing with a wide therapeutic index
  - “Enhanced stabilization chemistry” used to significantly improved potency and durability
- Clinically validated platform with human Proof-of-Concept in multiple programs

Adapted from Essentials of Glycobiology (2009)
## Development Pipeline

### Genetic Medicines

<table>
<thead>
<tr>
<th>Condition</th>
<th>Discovery</th>
<th>Development</th>
<th>Phase 1</th>
<th>Phase 2</th>
<th>Phase 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>TTR-Mediated Amyloidosis</td>
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<td></td>
<td>Patisiran</td>
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<tr>
<td>Hemophilia and Rare Bleeding Disorders</td>
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<td>ALN-AT3</td>
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<td>Revisiran</td>
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<td>Complement-Mediated Diseases</td>
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<td>ALN-CC5</td>
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<td>Hepatic Porphyrias</td>
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<td>ALN-AS1</td>
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<td>Alpha-1 Antitrypsin Deficiency</td>
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<td>ALN-AAT</td>
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<tr>
<td>Primary Hyperoxaluria Type 1</td>
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<td>ALN-GO1</td>
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</table>

### Cardio-Metabolic Diseases

<table>
<thead>
<tr>
<th>Condition</th>
<th>Discovery</th>
<th>Development</th>
<th>Phase 1</th>
<th>Phase 2</th>
<th>Phase 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hypercholesterolemia</td>
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<td></td>
<td></td>
<td>ALN-PCSsc</td>
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<tr>
<td>Hypertriglyceridemia</td>
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<td>ALN-AC3</td>
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<tr>
<td>Mixed Hyperlipidemia/Hypertriglyceridemia</td>
<td></td>
<td>ALN-ANG</td>
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<tr>
<td>Hypertension/Preeclampsia</td>
<td></td>
<td>ALN-AGT</td>
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</tbody>
</table>

### Hepatic Infectious Diseases

<table>
<thead>
<tr>
<th>Condition</th>
<th>Discovery</th>
<th>Development</th>
<th>Phase 1</th>
<th>Phase 2</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hepatitis B Virus Infection</td>
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<td>ALN-HBV</td>
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<td>Hepatitis D Virus Infection</td>
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<td>ALN-HDV</td>
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<td>Chronic Liver Infection</td>
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<td>ALN-PDL</td>
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</table>

Additional Genetic Medicine Programs

Additional Cardio-Metabolic Programs

Additional Hepatic ID Programs
Primary Hyperoxaluria Type 1 (PH1)

Summary

- Rare, devastating disease due to excessive overproduction of oxalate by the liver
  - Deficiency of alanine-glyoxylate aminotransferase (AGT)
  - Prevalence of 6-7 per million, higher in some populations globally
  - Calcium oxalate crystals deposit in kidneys, inflammation & fibrosis, nephrolithiasis
- Chronic kidney disease develops, leading to end-stage-renal-disease for the majority (nearly 100% lifetime risk)
  - Median age at diagnosis of 5 yrs; onset of ESRD - 24 yrs for males, 27 for females
- Once renal function is compromised, progressive systemic oxalate deposition can then lead to severe illness and death
  - Heart, CNS, skin, retina, joints, bone marrow
- The only effective treatment is dual liver-kidney transplantation
  - No approved pharmacological interventions

“As the liver is the only organ responsible for glyoxylate detoxification by AGT, the excessive production of oxalate will continue as long as the native liver is left in place.” - OxalEurope
Glycolate Oxidase (GO) Knockdown to Starve Substrate for Oxalate Synthesis in PH1

- Human GO deficiency well tolerated \(\rightarrow\) provides validation through increased glycolate excretion
  - 8-yr old boy with homozygous GO loss-of-function identified by Dr. Yaacov Frishberg
  - 20x increase in glycolate, normal oxalate, normal kidneys, no nephrocalcinosis
- GO deficient mice also validate therapeutic approach (Dr. Eduardo Salido)
  - Breeding with PH1 disease mice (AGT deficient) substantially resolves Uox levels

![Diagram of metabolic pathways involving GO and Uox levels](image)

Am J Physiol 2004; 287:C1359-65
J Med Genet 2014; 51:526-9
11th Annual PH Workshop, June 27-29, 2014, Chicago, IL
ALN-GO1 in Normal and Diseased Mice
Potent mRNA Silencing, Substantial Efficacy with Durability

Normal mice (single dose)

PH1 mice (single dose)

Urine Oxalate
(mg/g Creatinine/24 hr)

Urine Glycolate
(mg/g Creatinine/24 hr)

Serum glycolate (μM)

ALN-GO1 (mg/Kg)

day 10 sac

In collaboration with University of Alabama, Birmingham
ALN-GO1 Substantially Lowered Urinary Oxalate in Rat PH1 Model

Oxalate decreased up to 98% following weekly dosing

![Graph showing Urine Oxalate (mg/g Creatinine/24 hr) over days 0 to 28 for different ALN-GO1 doses (0, 0.3, 1, 3 mg/kg) compared to Vehicle (Veh)].

**Note:** >95% HAO1 mRNA silencing at all doses
Single, Low Dose ALN-GO1 in PH1 Rats
1:1 Relationship of Oxalate Lowering to HAO1 mRNA Silencing

Demonstrates substantial potential for efficacy
ALN-GO1 in Non-human Primates (ongoing)
Potent mRNA Silencing, Expected Increases in Serum Glycolate

<table>
<thead>
<tr>
<th>Group #</th>
<th>Dose Level (mg/kg)</th>
<th>Dose Frequency</th>
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</thead>
<tbody>
<tr>
<td>1</td>
<td>Veh</td>
<td>qMx6</td>
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<tr>
<td>2</td>
<td>0.25</td>
<td>qWx8</td>
</tr>
<tr>
<td>3</td>
<td>1</td>
<td>qWx8</td>
</tr>
<tr>
<td>4</td>
<td>1</td>
<td>qMx6</td>
</tr>
<tr>
<td>5</td>
<td>2</td>
<td>qMx6</td>
</tr>
<tr>
<td>6</td>
<td>4</td>
<td>qWx4 → qMx5</td>
</tr>
<tr>
<td>7</td>
<td>2 → 1</td>
<td>qWx4 → qMx5</td>
</tr>
</tbody>
</table>

Up to 99% silencing of HAO1 mRNA in non-human primates
ALN-GO1
Summary, Next Steps, & Acknowledgments

• Pre-clinical data summary
  ◦ Potent, durable silencing of HAO1 mRNA across species
    – Translates into expected increases in serum glycolate in healthy animals
  ◦ Profound lowering of urinary oxalate in animal models of PH1
    – 1:1 relationship of oxalate lowering to mRNA silencing

• Next steps
  ◦ Plan to file CTA in late 2015 and start Phase 1 study in early 2016 to study safety, along with impact on glycolate and oxalate metabolism
    – Pre-clinical durability supports monthly, and potentially quarterly, subcutaneous dosing

• Acknowledgments
  
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