2014 ASH Annual Meeting
Special Scientific Symposium on RNA Therapeutics in Hematology

RNAi Therapeutics for Hemophilia and Complement-Related Diseases

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Alnylam Pharmaceuticals
RNAi Therapeutics

- Harness natural pathway
  - Catalytic mechanism
  - Mediated by small interfering RNA or “siRNA”

- Therapeutic gene silencing
  - Any gene in genome
  - Distinct mechanism of action vs. other drug classes
  - Unique opportunities for innovative medicines

- Clinically validated platform
GalNAc-siRNA Conjugates as RNAi Therapeutics

Asialoglycoprotein Receptor (ASGPR)
- Highly expressed in hepatocytes
- High rate of uptake
- Recycling time ~15 minutes
- Conserved across species

GalNAc-siRNA Conjugates (revusiran, ALN-AT3, ALN-CC5, ALN-PCSsc, other programs)
- siRNA conjugated to N-acetylgalactosamine (GalNAc) ligand
- Efficient delivery to hepatocytes following subcutaneous administration
- “Enhanced stabilization chemistry” (ESC) used with ALN-AT3, ALN-CC5, ALN-PCSsc, and other programs
  » Significantly improved potency and durability compared with revusiran
## Alnylam Development Pipeline

<table>
<thead>
<tr>
<th>Condition</th>
<th>Discovery</th>
<th>Development</th>
<th>Phase 1</th>
<th>Phase 2</th>
<th>Phase 3</th>
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<tbody>
<tr>
<td>TTR-Mediated Amyloidosis</td>
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<td>Patisiran (ALN-TTR02)</td>
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<td>ALN-TTRsc</td>
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<td>Hemophilia and Rare Bleeding Disorders</td>
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<td>ALN-AT3</td>
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<td>Complement-Mediated Diseases</td>
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<td>ALN-CC5</td>
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<td>Hypercholesterolemia</td>
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<td>ALN-PCSsc</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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<td>Hepatitis B Virus Infection</td>
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<td>ALN-HBV</td>
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<td>Beta-Thalassemia/Iron-Overload Disorders</td>
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<td>ALN-TMP</td>
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<td>Mixed Hyperlipidemia/Hypertriglyceridemia</td>
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<td>ALN-ANG</td>
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<tr>
<td>Hypertriglyceridemia</td>
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<td>ALN-AC3</td>
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<td>Hypertension/Preeclampsia</td>
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<td>ALN-AGT</td>
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<td>Primary Hyperoxaluria Type 1</td>
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<td>ALN-GO1</td>
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<tr>
<td>Additional Genetic Medicine/Other Programs</td>
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</table>

**Delivery Technology:**
- LNP (IV)
- Standard Template Chemistry (STC)-GalNAc Conjugate (SC)
- Enhanced Stabilization Chemistry (ESC)-GalNAc Conjugate (SC)
Hemophilia and Complement-Mediated Diseases

Why RNAi Therapeutics?

• RNAi human translation achieved in multiple clinical programs
• GalNAc-siRNA conjugates have solved delivery challenge to liver
• Hemophilia and complement-mediated diseases are technical and strategic fit
  » Significant unmet medical need
  » Hepatocyte-expressed, secreted plasma protein targets
  » Utilize existing delivery platform
  » Well-validated, genetically defined pathways/diseases
  » Phase 1 proof-of-concept possible
  » Clear and rapid development
RNAi to treat hemophilia and rare bleeding disorders (RBD)

- Hemophiliias are recessive X-linked monogenic bleeding disorders
  - Hemophilia A: loss of function in Factor VIII
    - >40,000 Patients in EU/US
  - Hemophilia B: loss of function in Factor IX
    - ~9,500 Patients in EU/US

- Segments of high unmet need remain
  - E.g., “Inhibitor” patients\(^1\),\(^2\)
    - 2,000 Patients in major markets; up to 6,000 WW
    - >15-25 Bleeds/year; >5 in-hospital days/year
    - ~$300,000/year avg. cost; up to $1M/year

- RBD patients\(^3\)
  - Includes deficiencies in Factors II, V, VII, X, and XI
  - ~1,000 patients WW with severe bleeding phenotype

\(^1\) WFH 2012 Global Survey; \(^2\) Antunes et al., Haemophilia. 20:65-72 (2014); \(^3\) Peyvandi et al., J Thromb Haemost; 10, 615-621 (2012)
Antithrombin (AT) is genetically defined target
- AT is key natural anticoagulant
  - Inactivates factors Xa and thrombin
  - Attenuates thrombin generation
- Human AT deficiency associated with increased thrombin generation
- Expressed in liver; circulates in plasma

Co-inheritance of thrombophilic traits in hemophilia
- Associated with milder bleeding, reduced factor requirements, fewer complications
- Includes heterozygous
  - Antithrombin deficiency
  - Factor $V_{\text{Leiden}}$
  - Protein C deficiency
  - Protein S deficiency

ALN-AT3 in clinical development
- ESC-GalNAc-siRNA for SC dosing
- Positive top-line SAD Phase 1 results
- Orphan drug status in US and EU (HA/HB)
- Phase 1 MAD study in patients ongoing

Antithrombin depletion increases peak height and delays inhibition of thrombin.

- Antithrombin depletion increases peak height and delays inhibition of thrombin.
**ALN-AT3 Improves Hemostasis**

**Microvessel Laser Injury Model (HA Mice)**

- **Deposition of Platelets**
- **Deposition of Fibrin**

### Table: Hemostasis Parameters

<table>
<thead>
<tr>
<th>Group</th>
<th>Animals (N)</th>
<th>Injuries (N)</th>
<th>Stable Thrombus (N)</th>
<th>Percent AT mRNA in liver</th>
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</thead>
<tbody>
<tr>
<td>WT</td>
<td>5</td>
<td>25</td>
<td>25</td>
<td>100%</td>
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<tr>
<td>HA + PBS</td>
<td>5</td>
<td>25</td>
<td>0</td>
<td>100%</td>
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<tr>
<td>HA + 1 mg/kg ALN-AT3</td>
<td>5</td>
<td>25</td>
<td>25</td>
<td>50%</td>
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<tr>
<td>HA + 30 mg/kg ALN-AT3</td>
<td>6</td>
<td>30</td>
<td>30</td>
<td>5%</td>
</tr>
</tbody>
</table>

*Akinc, ISTH, July 2013; In collaboration with Dr. Lacra Ivanciu and Dr. Rodney Camire*
ALN-AT3 Improves Hemostasis
Saphenous Vein Bleeding Model (HA Mice)

Saphenous vein bleeding model adapted from Whinna¹
- Saphenous vein exposed and transected to initiate bleeding
- Thirty seconds after cessation of blood loss, clot dislodged to re-initiate bleeding
- Number of hemostatic events in 30 min observation period recorded

Protocol details²
- Treated animals received single SC dose of ALN-AT3 to yield ~70% AT knockdown at time of model initiation
- Control animals received 25 IU/kg Advate via IV administration 15 min prior to model initiation

Akinc, WFH 2014; ¹Buyue et al., Blood; 112:3234-3241 (2008); ²In collaboration with Dr. Brian Cooley
ALN-AT3 Single-Dose Pharmacology in NHP
AT Silencing Leads to Increased Thrombin Generation

- ED50 for ALN-AT3 is ~1 mg/kg
- Nadir for AT level ~Day 14
**ALN-AT3 Repeat-Dose Pharmacology in NHP**

**Monthly, SC Dosing**

- **Animals:** Cynomolgus monkeys (N=3 per group)
- **Dose levels:** 0.5, 1 mg/kg, and 2 mg/kg (SC, monthly dosing)

![Graph showing Relative Plasma AT levels over days for different dose groups.](image)

- Dosing suspended in 2 mg/kg group after 3 doses due to AT levels <20%
ALN-AT3 Treatment Normalizes Thrombin Generation in Hemophilic NHPs with Inhibitors

**Induction of Hemophilia A**

- Pre Ab vs 4 hr Post Ab
- Relative FVIII Levels
- Saline, 0.25, 0.50 mg/kg ALN-AT3 qw
- <0.01

**Normalization of Thrombin Generation**

- Peak Thrombin (nM)
- Pre-dose, Normal, 60% AT reduction
- Saline, 0.25, 0.50 mg/kg ALN-AT3 qw
- 80% AT reduction
- **(p<0.01)**

*similar results obtained by ETP (p<0.01 at 0.50 mg/kg)*

ISTH, July 2013
ALN-AT3 Survival Benefit in HA Mice
Results from 26-Week GLP Chronic Toxicity Study

Groups
- Animals: Hemophilia A (HA) mice (B6;129S4-F8\textsuperscript{tm1Kaz})
- Dosing: Saline, 10 mg/kg, and 30 mg/kg (SC, weekly dosing)
- Group sizes: (N=70; 35 per sex) powered to account for spontaneous loss due to background sensitivity of HA mouse strain

Interim results (through Week 25):
- No adverse clinical signs; no changes in body weight, hematology or clinical chemistry
- Survival benefit noted in treated animals (p <0.0001; Log-rank, Mantel-Cox test)
ALN-AT3 Phase 1 Study
Dose-escalation Study in Two Parts

Study Design
- Randomized, single-blinded, placebo-controlled SAD study in healthy volunteers

Primary Objective
- Safety and tolerability of single doses with AT knockdown <40%

Secondary Objectives
- Assess clinical activity
  » AT knockdown

Study Design
- Open-label, MAD study in subjects with moderate to severe hemophilia A or B (N=up to 18)

Primary Objective
- Safety and tolerability of multi-dose in hemophilia subjects

Secondary Objectives
- Assess clinical activity
  » AT knockdown
  » Increase in thrombin generation

See Sorensen, Oral Abstract 0693; 322. Disorders of Coagulation or Fibrinolysis: Novel Hemostatic Therapies and Assays; Mon Dec 08 6:45-7:00PM.
Complement Disease Program
Unmet Need and Program Opportunity

Complement-Mediated Diseases
- Excessive complement activity drives disease pathophysiology in many indications
  » Paroxysmal nocturnal hemoglobinuria (PNH)
  » Atypical hemolytic uremic syndrome (aHUS)
  » Neuromyelitis optica (NMO)
  » Myasthenia gravis
  » Many others
- Soliris™ (eculizumab) is blockbuster drug
  » >$1.5B in reported 2013 sales
  » >$2.0B in forecasted 2014 revenue

New therapeutic options needed
- Consistent level of efficacy
- SC delivery for more tolerable treatment regimen
- Reduce access barriers to treatment
Complement C5 and ALN-CC5 Program

**Complement C5 is genetically validated target**
- Key component of terminal pathway
  - C5 cleavage releases C5a; initiates membrane attack complex (MAC) formation
- C5 deficiency associated with minimal complications
  - Susceptibility to increased *Neisserial* infections
  - Many C5 deficient mouse strains
- Majority expressed in liver; circulates in plasma

**Complement C5 is clinically validated target**
- Eculizumab is anti-C5 Mab
- Approved in PNH and aHUS
  - In PNH, >80% inhibition of hemolytic activity associated with clinical benefit\(^1\)
- Potential advantages of synthesis inhibition vs. protein binding approach

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*ALN-CC5 CTA filed*
- ESC-GalNAc-siRNA for SC dosing
- Efficacy in pre-clinical animal models
- CTA filed; initial data mid ’15


**See Borodovsky Poster Abstract 1606; 508. Bone Marrow Failure: Poster I; Sat Dec 06 5:30-7:30**
Robust and Sustained C5 Knockdown in NHP

Robust knockdown of serum C5 with SC dosing in NHP for >7 months
- Q2W Regimen: Every other week dosing (5 mg/kg, qw x 8, q2w thereafter)
- QM Regimen: Every month dosing (5 mg/kg, qd x 5, qw x 8, 10 mg/kg qm thereafter)
- 2xW Regimen: 5 mg/kg 2xw x 8

Up to 99.2% knockdown of serum C5
- 98.4 ± 0.7% knockdown as group average
- Low inter-animal variation
- Q2W Regimen provides optimal C5 knockdown results in NHP
  » Expect QM dosing regimen in humans based on translation of ESC-GalNAc-siRNA conjugates

Serum C5

Individuals (Q2W and QM)
Both Classical and Alternative Pathways Strongly Inhibited

- Up to 96.9% inhibition of alternative pathway (CAP) activity (mean 95.1 ± 0.93%), and up to 96.2% inhibition of hemolysis (mean 88.0 ± 6.1%)
  - Q2W Regimen provides optimal inhibition of complement activity in NHP but monthly likely in humans based on translation of ESC-GalNAc conjugates
- In line with results observed in published reports on eculizumab
C5 Knockdown Reduces Proteinuria
Rat Membranous Nephropathy Model

~90% reduction in urinary albumin levels with C5 knockdown in rat model of Passive Heymann Nephritis (PHN)

- Nephritis induced by injection of sheep anti-rat kidney fraction antiserum (anti-Fx1A)
  - Cobra venom factor (CVF) treated rats receive daily injections
- Similar reduction in urinary albumin excretion to CVF complement depletion
Glomerular MAC Deposition is Prevented with C5 Silencing

- No glomerular C5b-9 deposition with C5 siRNA or CVF treatment
- Equivalent glomerular sheep IgG deposition regardless of treatment, as expected
C5 Knockdown Results in Equivalent Activity to Anti-C5 Ab

Mouse CAIA Model

Circulating liver-derived C5 key driver of pathology in mouse CAIA

- Little or no role for locally produced C5
ALN-CC5 Phase 1 Study

ALN-CC5 CTA filed

Phase 1/2 study expected to start in early ’15; Initial data expected in mid ’15

- ALN-CC5 with SC dosing
- Parts A/B: SAD/MAD in up to 60 normal healthy volunteers
  » Randomized, double-blind, placebo-controlled study
  » Assess safety, tolerability, PK/PD, and clinical activity
- Part C: Multi-dose in up to 8 PNH patients
  » Open-label study
  » Assess safety, tolerability, PK/PD, clinical activity, and LDH reduction
Summary

• RNAi therapeutics continue to progress in clinical trials
• ALN-AT3 and ALN-CC5 are Alnylam’s first programs in hemophilia and complement-mediated disease, respectively
  » Robust AT and C5 target knockdown observed with monthly SC dosing in NHPs
  » Pharmacodynamic activity observed in multiple preclinical models
• ALN-AT3 currently in Phase 1 in hemophilia patients, and ALN-CC5 will be in Phase 1/2 in early 2015
Thank You