BACKGROUND

Human genetic analysis has identified that individuals with loss-of-function mutations in either apolipoprotein-C3 (APOC3) or angiopoietin-like protein 3 (ANGPTL3) have very low plasma levels of triglycerides (TGs) and in the setting of ANGPTL3 deficiency, low-density lipoprotein (LDL-C). Both conditions are associated with a reduced risk of cardiovascular disease.

OBJECTIVE

ANGPTL3 and APOC3 are primarily expressed in hepatocytes. An RNA interference (RNAi) based therapy using Arrowhead Pharmaceuticals’ TRiM™ platform to reduce APOC3 or ANGPTL3 production by gene silencing may be an effective approach to treat dyslipidemias and metabolic diseases (AHA 2018).

METHODS

Highly potent and specific RNAi conjugates were identified targeting human and non-human primate (NHP) APOC3 transcripts (ARO-APOC3) or ANGPTL3 transcripts (ARO-ANG3). Rodent or NHP (high fructose diet-fed rhesus macaques) dyslipidemic animal models were used to study pharmacodynamic effects in target protein reduction and reductions in TGs and LDL-C.

RESULTS

ARO-ANG3 studies

ARO-ANG3 was evaluated in LDL receptor knockout (LDLr KO) mice, diet-induced obese (DIO) mice, as well as a fructose-fed dyslipidemic NHP model. In all animal models, maximum serum reductions in ANGPTL3 of 95% were achieved and persisted for at least 8 weeks. Reductions in TGs and LDL-C were also observed.

ARO-APOC3 in Dyslipidemic Rhesus

- Reductions in serum APOC3 (up to 80%) and TGs (up to 90%)
- Magnitudes of reductions correlated to the severity of dyslipidemia

CONCLUSIONS

- Our results support an RNAi therapeutic targeting APOC3 or ANGPTL3 as a treatment for dyslipidemia
- ARO-ANG3 may also provide metabolic benefits in the liver and impact LDL-C in familial hypercholesterolemia
- Both development candidates can be used to target specific patient populations depending on underlying genetic and metabolic profiles
- ARO-APOC3 and ARO-ANG3 have recently entered human clinical trials

ACKNOWLEDGEMENTS

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ARO-ANG3 Studies in Mouse Models

**Dose response in LDLr KO mice, N = 5-6, single SQ dose on Day 1**

- Serum ANGPTL3
- Serum TGs
- Serum LDL-C

**ARO-ANG3 with atorvastatin in LDLr KO mice, N = 6-7**

- LDLr KO mice on Western diet
- Atorvastatin has no effects on ANGPTL3 expression
- Additive effects of ARO-ANG3 + atorvastatin

**Improvement of glucose tolerance and reduction in hepatic steatosis in DIO mice**

- Glucose Tolerance Test
- Serum ANGPTL3

- Mice on high fat diet (60% kcal% fat), N = 7
- Two doses of 3 mg/kg ARO-ANG3 on Day 1 and 22
- Glucose Tolerance Test on Day 41, necropsy Day 44

**Reduction in hepatic Steatosis (H&E)**

- Saline (animals a and b)
- ARO-ANG3 (animals c and d)
Tumor targeting ligand (TTL) facilitates receptor-mediated tumor uptake of a HIF2α RNAi trigger

• Animals on fructose diet for 6 weeks before dosing
• Variable diet-induced dyslipidemia
• Over 95% maximum reductions in serum ANGPTL3 protein levels
• 80% maximum mean reductions in TGs
• 20-60% max reductions in LDL-C (not shown)

External condition

Reductions in Serum ANGPTL3: group average N = 2 (Saline) or 4 (ARO-ANG3)

Reductions in Serum TGs: group average N = 2 (Saline) or 4 (ARO-ANG3)

Reductions in Serum TGs: individual

Fructose-diet mediated changes in ANGPTL3 and TGs

<table>
<thead>
<tr>
<th>Animal</th>
<th>Serum ANGPTL3 (ng/dL)</th>
<th>TGs (mg/dL)</th>
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<tr>
<td></td>
<td>Pre-diet</td>
<td>Pre-dose</td>
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<tr>
<td>ARO-ANG3-4</td>
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</table>
A RO-APOC3 Studies in Mice and Fructose Diet-Fed Dyslipidemic Rhesus Monkeys

Dose response in human APOC3 transgenic mice
N = 5-6, single SQ dose on Day 1

- Animals on fructose diet for 6 weeks
- Variable diet-induced dyslipidemia
- Maximum mean serum APOC3 reduction of 67% (range: 60-80%), which likely represents complete hepatocyte knockdown
- Small intestinal production still intact
- Maximum mean TGs reduction of 60% (range: 40-90%)
- 20-60% max reductions in LDL-C (not shown)
- ARO-APOC3 (N=4), Saline (N=2)