Alnylam interrupts preeclampsia

By Michael J. Haas, Associate Editor

Preeclampsia often involves overactivation of the renin-angiotensin system that regulates blood pressure, but inhibiting the pathway is not possible because it would be toxic to the fetus. **Alnylam Pharmaceuticals Inc.** has developed an siRNA conjugate that does not cross the placenta or affect fetal development but can block angiotensin signaling and improve symptoms in a rat model of the condition.

Preeclampsia occurs in about 5% of pregnant women and can lead to life-threatening seizures. The condition is usually diagnosed in the second or third trimester when symptoms of hypertension and proteinuria appear, but the precise etiology of preeclampsia is poorly understood.

Over the past 20 years, multiple studies have shown that preeclampsia and other forms of pregnancy-related hypertension often involve upregulation of the renin-angiotensin system caused by activating mutations in *angiotensinogen* (*AGT*)[1–3] or by agonistic autoantibodies that target a key receptor in the pathway called angiotensin II type 1 receptor (AGTR1).[4,5] AGT, a protein produced mainly in the liver, sits atop a cascade that regulates angiotensin-converting enzyme (ACE)-induced production of the hypertensive peptide angiotensin II.

Many companies market antihypertensives that inhibit AGTR1 or ACE, but the drugs' use during pregnancy is contraindicated because they can cross the placental barrier and cause fetal injury or death.

Thus, the standard of care for preeclampsia involves managing symptoms by treating hypertension with other drug classes—such as calcium channel blockers and adrenergic receptor antagonists—in addition to using magnesium sulfate to prevent seizures and steroids to promote fetal lung development before delivery. The only effective treatment is delivery of the baby by induced labor or Caesarean section.

Alnylam's siRNA conjugate—dubbed ALN-AGT—is the first disclosed program that targets the renin-angiotensin system to treat preeclampsia. ALN-AGT contains two parts: a chemically modified siRNA against human *AGT* and an *N*-acetylgalactosamine (GalNAc) ligand that binds asialoglycoprotein receptor 1 (ASGR1; CLEC4H1) on hepatocytes, thereby targeting the *AGT* siRNA specifically to the liver.

"For treating hypertensive disorders of pregnancy, the specificity of our hepatic targeting combined with the size of the molecule that prevents placental transfer provides a significant therapeutic advantage over existing drugs that target the renin-angiotensin system," said Alnylam spokesperson Cynthia Clayton.

The company tested the compound in rats engineered to express human wild-type *AGT* in addition to rat *Agt*. The idea was to overexpress the precursor and thus produce high levels of angiotensin II that would generate a preeclamptic phenotype when the rats became pregnant.

In the rats, subcutaneous injections of ALN-AGT decreased mean arterial pressure by about 20 mmHg and decreased proteinuria by more than 80% compared with no treatment. In addition, ALN-AGT decreased the activity of agonistic antibodies against Agtr1 by about 90%. ALN-AGT also reduced maternal kidney levels of placental growth factor (Pgf; Plgf) and soluble Vegf receptor 1 (sFlt1; sVegfr-1)—two proteins thought to be predictive markers of preeclampsia.[6–8]

ALN-AGT increased fetal weight, the overall weight of the utero-placental unit and the ratio of fetal liver weight to brain weight—all of which are factors that could improve fetal outcomes. The company attributed the improvements to therapy-induced enlargement of the labyrinth, the region of the placenta where nutritional exchange between mother and fetus occurs.

Moreover, whereas ALN-AGT reached significant levels in the maternal liver, its levels in fetal livers were below the limit of detection, a finding that suggested ALN-AGT did not cross the placental barrier to reach the fetus.

Alnylam presented the findings in a poster at the **American Heart Association**'s High Blood Pressure Research meeting in September.[9] The study included researchers from the **Max Delbrueck Center for Molecular Medicine**, **Charité–University Hospital Berlin** and **Helios Kliniken GmbH**.

Clayton said that the company thinks ALN-AGT could have broad utility in treating preeclampsia and other pregnancy-related hypertensive conditions.

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>
> *—Matthew Cooper, Carmenta Bioscience Inc.*

Birth of the RNAi age?

"These are really exciting data," said Matthew Cooper, founder and CEO of **Carmenta Bioscience Inc.** "I'm happy to see that a company has a serious therapeutic candidate for preeclampsia."

"And kudos to Alnylam for choosing to take on this target," he said. "AGT plays a major role in hypertension, which is the leading cause of organ damage in preeclampsia. So if you can actually regulate that hypertension in preeclampsia—even if you are not treating the underlying cause of the disease—then you are affecting one of its major causes of mortality—and that's fantastic."

He also noted that the siRNA conjugate achieved "the preeclampsia trifecta" of altering markers of the disease, inducing beneficial structural changes in the placenta and improving outcomes for the fetuses.

Carmenta has a panel of six undisclosed serum markers in clinical development as a confirmatory diagnostic for preeclampsia.

Irina Buhimschi agreed that Alnylam's approach was interesting, but she said it was not surprising that silencing the *AGT* transgene responsible for the preeclamptic phenotype in the rats reversed their disease symptoms. Moreover, she said, mice and rats do not develop preeclampsia naturally or spontaneously, so any genetic alteration that induces preeclampsia-like symptoms in animals might be unrelated to the underlying causes of the disease in patients.

Thus, before concluding that ALN-AGT could treat human preeclampsia, "it needs to be explored in an animal model of the disease that is not affected by the same *AGT* transgene–induced mechanism," she said. She cited rats with chronic inhibition of nitric oxide synthase or mice that overexpress sFlt-1 as models that Alnylam could try next.

Buhimschi is a professor of pediatrics and obstetrics/gynecology at **The Ohio State University** and director of the Center for Perinatal Research at **Nationwide Children's Hospital**.

Cooper countered that although the rat model is not perfect, "it is a good model for the hypertensive component of preeclampsia, if not preeclampsia as a whole." Moreover, "if ALN-AGT had not worked in this rat model, then you would know it was a no-go."

He added, "RNAi therapeutics are coming of age, and there are several situations where they could work really well. Preeclampsia could be one of them."

Clayton said that Alnylam's next step will be to test the approach in a non–angiotensin-driven animal model of preeclampsia. The company has not disclosed its clinical plans for ALN-AGT.

This week, Alnylam reported preliminary data from an ongoing open-label Phase II extension study of patisiran (ALN-TTR02) suggesting the compound may have halted progression of transthyretin (TTR)-mediated amyloidosis (ATTR) in patients with familial amyloidotic polyneuropathy (FAP). Alnylam and partner **Sanofi** also have ALN-TTR02 in Phase III testing to treat FAP. The compound is an RNAi therapeutic that targets *TTR* using second-generation lipid nanoparticle technology.

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COMPANIES AND INSTITUTIONS MENTIONED

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American Heart Association, Dallas, Texas
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