Structural Optimization of TransCon CNP - Development of a Sustained-Release Prodrug of CNP for Achondroplasia


This trial was sponsored by Ascendis Pharma A/S.

Background

Achondroplasia (ACH), the most common form of human dwarfism, is caused by a gain-of-function mutation in the fibroblast growth receptor 3 (FGFR3) gene, a key negative regulator of chondrocyte proliferation and terminal differentiation.

Via activation of natriuretic peptide receptor B (NPR-B), C-type natriuretic peptide (CNP) inhibits the effects of the FGFR3 signaling pathway and has demonstrated efficacy in longitudinal bone growth.1,2

CNP is rapidly cleared and degraded by natriuretic peptide receptor C (NPR-C) and neutral endopeptidase (NEP), respectively, resulting in a 2-minute half-life.

Vosoritide, a mutated CNP analogue with a 20-minute half-life, is in phase 3 development for use in ACH. However, its short half-life leads to high peak-to-trough ratios and limited therapeutic coverage. Humans and animals dosed with vosoritide have also demonstrated hypotension coinciding with maximum blood concentrations (Cmax), indicating a causal relationship.1,2

A potential long-acting CNP drug candidate should provide sustained exposure of CNP at therapeutic levels. Additionally, a safe and efficacious candidate should not cause adverse cardiovascular effects or undergo rapid clearance and degradation of CNP.

Methods

Through a series of in vitro assays (including a potency assay with NIH 3T3 cells expressing murine NPR-B, a binding assay with HEK cells expressing NPR-C, and a NEP incubation assay), the EC50, IC50, and degradation time, respectively, of multiple TransCon CNP candidates was compared to unmodified CNP. To avoid release of unmodified CNP in the in vitro assays, a permanently conjugated TransCon CNP molecule, in which the linker does not autohydrolyze, was used.

The present work describes the structural optimization of TransCon CNP.

Results

As designed, in the cell-based potency and binding assays, the TransCon prodrug of CNP demonstrated extremely low biological activity against both the NPR-B and NPR-C receptors. Additionally, CNP within the prodrug was protected against NEP degradation. In contrast, liberated, unmodified CNP was fully active.

In monkeys, a TransCon CNP half-life of 79 hours was confirmed:

![Graph showing the half-life of TransCon CNP in monkeys.](image)

In monkeys, no decrease in SAP was detected at TransCon CNP doses of up to 100 µg/kg compared to controls during a 48-hour monitoring period. In contrast, an equivalent dose of vosoritide led to a decrease in SAP at 0-1 hours post-dose.

![Graph showing blood pressure changes after TransCon CNP and vosoritide administration.](image)

Conclusions

- The TransCon CNP prodrug has minimal binding to the NPR-B and NPR-C receptors and improved NEP stability in vitro compared to unmodified CNP.
- In vivo TransCon CNP demonstrated the desired half-life extension without adverse hemodynamic effects.
- These data support clinical development for weekly dosing and suggest TransCon CNP may be a safe and efficacious option for children with ACH.

Context

Structural optimization of the TransCon CNP molecule provided the background for successful preclinical studies:
- TransCon CNP demonstrated efficacy in healthy juvenile cynomolgus monkeys, increasing long bone growth in a dose-dependent fashion.1
- In a murine model of ACH, TransCon CNP improved growth plate architecture and increased naso-anal and femur bone length.1

Collectively, these studies support continued development of TransCon CNP.

1 BioMarin R&D Day 2016 Presentation
2 Wendt et al. J Pharmacol Exp Ther, 2015; 353, 132-149
3 Ascendis Pharma IMPE 2017 Presentation

"Vosoritide" refers to a synthesized molecule with the amino acid sequence prepared by Ascendis Pharma.