Tralesinidase alfa enzyme replacement therapy prevents disease manifestations in a canine model of mucopolysaccharidosis type IIIB


Supplementary Information

Supplementary Methods

PK/PD study in MPS IIIB dogs

Study design: The study design is outlined in Supplementary Table 2 and schema in Supplementary Figure 1B. The dogs used in this study were produced and maintained at ISU as described in the main text until transferred to the Northern Biomedical Research Inc., an AAALAC-accredited facility. Housing was compliant with the Guide for the Care and Use of Laboratory Animals, DHHS, and NIH guidelines. Dogs were fed Purina Certified Lab Canine Diet. Dogs were given filtered municipal water ad libitum.

Dogs of approximately 1 year of age were implanted with catheters to a lateral ventricle for dose administration and to the cervical spine for serial CSF collection. MRI was performed as described to implant the ICV catheter appropriately (Vuillemenot et al. 2014). Dogs were randomized into groups by body weight and identified with a tattoo and an implanted transponder. Dogs were administered directly into the CNS either 15 mg or 30 mg tralesinidase alfa (TA) weekly for 8 weeks.
Tralesinidase alfa administration: TA was formulated in artificial CSF (148 mM NaCl, 3 mM KCl, 0.8 mM MgCl2•6H2O, 1.4 mM CaCl2•2H2O, 0.7 mM Na2HPO4, 0.3 mM NaH2PO4, pH 7.0) at 12.2 mg/ml (15 mg dose level) or 25.1 mg/ml (30 mg dose level). Dogs were administered ~1.2 ml TA by ICV infusion over 5 minutes (~0.24 ml/min) after first removing up to 1.2 ml CSF via the IT-C port when possible. When ICV catheters were no longer patent, dogs were administered a bolus dose of TA (1.2 ml) by CM spinal tap over approximately 1.2 minutes after first removing up to 1.2 ml of CSF.

PK and antibody analysis: Pharmacokinetic samples were obtained from CSF from the lumbar before the ICV infusion start (pre-dose), immediately after the end of the dose (~2 minutes post dose), and at 0.5-, 2-, 6-, 10-, 24-, 36-, 48-, 72-, and 96-hours post-dose at Doses 1, 4, and 8. Pharmacokinetic samples were obtained from blood from a peripheral vein prior to the dose, after the end of the dose (~2 min post dose) and 0.5, 1, 2, 6, 10, 16, 24, 36, 48, 72, and 96 hours pose dose at Doses 1, 4, and 8. TA levels were measured as described (Grover et al. 2020). Total anti-drug antibody levels were analyzed in serum and CSF samples using an electrochemiluminescent immunoassay technology platform (ICON Laboratory Services, Whitesboro, NY).

Necropsy: Dogs from Group 1 were euthanized at 1-, 2-, or 4-weeks post last dose (n = 3 per time point) and dogs from Group 2 were euthanized at 1 week post last dose. CSF samples and biopsies from CNS tissues were collected and analyzed as described previously (Grover et al. 2020) and in the main text.

PD marker analysis. Tissue punches from brain regions and spinal cord from MPS IIIB dogs were collected at the time of necropsy. CNS tissue samples were pooled for each of the dogs. Analysis of HS and HS-NRE levels in CSF (ARUP Laboratories, Salt Lake City, UT) and CNS tissue (BioMarin Pharmaceutical Inc.) was conducted as described previously (Aoyagi-Scharber et al. 2017) and in the main text.