Krabbe disease: a pre-clinical study of nanoparticle-based enzyme replacement therapy in the twitcher mouse

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Disease Models & Mechanisms

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Background

Krabbe disease (KD) is a fatal pediatric neurodegenerative lysosomal storage disorder (LSD) caused by deficient activity of the enzyme galactosylceramidase (GALC). This condition leads to severe demyelination and consequent neurodegeneration of both the central nervous system and peripheral nervous system. Despite the disease gravity, the current standard of care is mostly supportive only. The enzyme replacement therapy has gained broad interest thanks to the effective results achieved in other LSD. However, applications of such therapy to KD still do not exist.

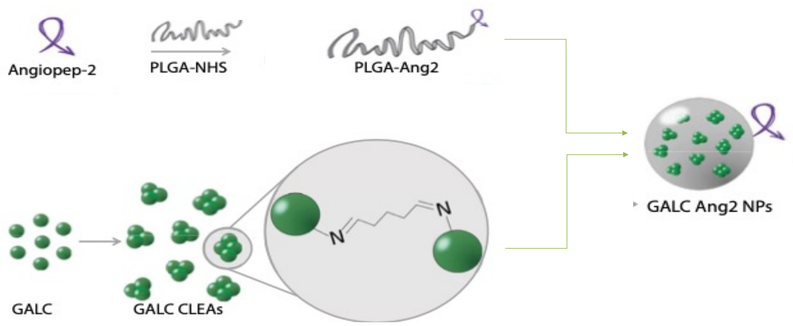
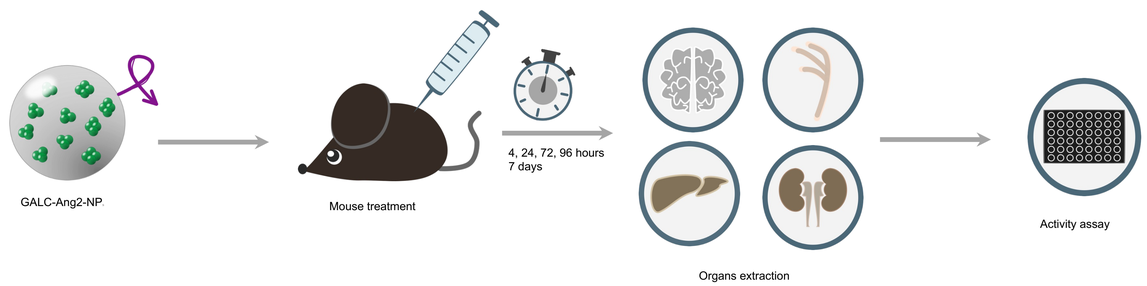


Figure 1. Brain targeted GALC-Ang2-NPs [Del Grosso et al., Scientific Reports, 2019].

Aim of the study

Here, an enzyme delivery system based on the encapsulation of cross-linked enzyme aggregates (CLEAs) into poly-(lactide-co-glycolide) (PLGA) nanoparticles (NPs) functionalized with brain targeting peptide (Ang-2) (GALC-Ang2-NPs) (Figure 1) is investigated for the treatment of the natural murine model of KD, called Twitcher (TWI).

Materials and methods



TWI mice were treated with GALC-Ang2-NPs, or with the free rm-GALC (GALC). After 4, 24, 72, 96 hours and up to 7 days later, mice were euthanized, and GALC activity was assayed in extracted brain, sciatic nerves,, kidneys, and liver by HMU-bGal assay.

Acknowledgements

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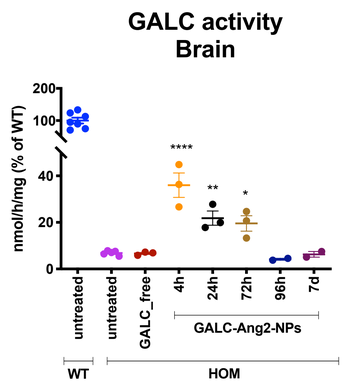
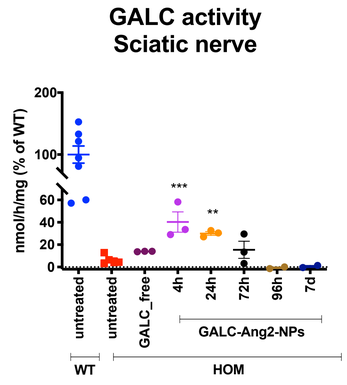
Results

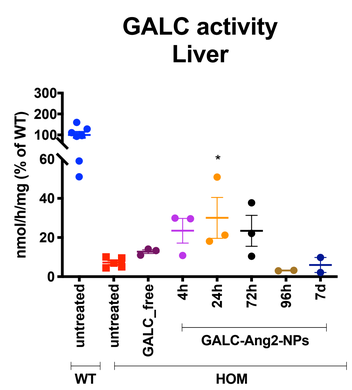


Regarding the brain, we found a significant increase of enzymatic activity in the brain of HOM mice treated with GALC-Ang2-NPs after 4, 24 and 72 hours from the treatment. These findings confirm the capability of our NP to cross the blood-brain barrier (BBB) and to deliver enzymatically active GALC into the brain.

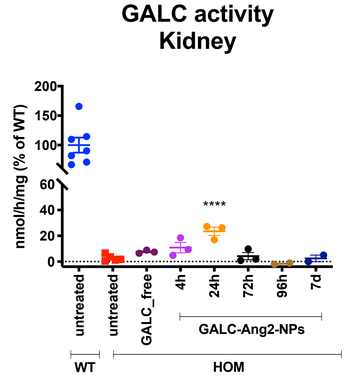


Enzymatic activity of HOM treated mice was significantly increased after 4 and 24 hours also in sciatic nerves.

**** P < 0.0001; *** P < 0.001; ** P < 0.01; * P < 0.05 vs HOM untreated ANOVA, Dunnett's multiple-comparisons test.



In the accumulation organs, we found an increase of enzymatic activity in the liver of HOM mice treated after 24 hours from the treatment in the liver.



Regarding the kidneys, we found data similar to the liver. enzymatic activity after 24 hours was significantly increased in respect to untreated TWI mice.

Conclusions

We report a significat increase of GALC enzymatic activity measurements in the nervous system and in accumulation districts upon intraperitoneal injections, demonstrating activity recovery in the brain up to levels of clinical interest. Thus, given the established ability of our developed drug to pass the blood-brain barrier and release the active enzyme until 3 days, experiments of prolonged treatment have currently been ongoing. Together, these results open new therapeutic perspectives for all LSDs with major central nervous system-involvement.

References

Del Grosso A, et al. Brain-targeted enzyme-loaded nanoparticles: A breach through the blood-brain barrier for enzyme replacement therapy in Krabbe disease. 2019, Sci Adv. 20;5(11):eaax7462. doi: 10.1126/sciadv.aax7462.