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Lectin-assisted Enzyme Replacement Therapy for GM1 Gangliosidosis

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In this project, a fusion protein of the human β-gal enzyme with the tag RTB lectin (β-gal:RTB) was produced in a plant-based protein production system. The purified protein was administered to *β-gal*$^{-/-}$ mice to determine if active enzyme reaches the cells of the brain. This research is at the basis of an NIH-funded project focused on the development, optimization, and biochemical characterization of the β-gal:RTB product and an assessing its efficacy in reducing or ameliorating disease pathogenesis in the GM1 gangliosidosis mouse model, following intravenous administration. The ultimate scope of this research project is to translate it to the clinic.

Results and Conclusions:

We have successfully produced plant-based β-gal:RTB and purified it to the degree required for its administration in mice. Several trials have been carried out in *β-gal*$^{-/-}$ mice ranging in age from 5 to 9 weeks. Different doses of β-gal:RTB were administered twice a week for 6 weeks and β-gal activity quantified in various organs 24 hours after the last injection. Although a clear dose response curve of the enzyme was measured in all visceral organs, increasing the amount of the therapeutic protein did not produce a linear increase in β-gal activity in different brain regions. However, we could detect low levels of the recombinant protein in some brain regions, which was accompanied by a slight increase in enzyme activity and reduction of the storage product. To better assess the ability of the recombinant protein to be internalized by affected neural cells and to spread throughout the brain parenchyma, we have also carried out direct injections into the ventricular system of the brain. Increased β-gal activity was measured up to a week post injection in all brain regions tested. Surprisingly, significant β-gal activity was also detected in several visceral organs of the brain-injected mice, suggesting that the recombinant β-gal:RTB was mobilized into the circulatory system and subsequently routed to the visceral organs. Of significance, our experimental results with both administration routes indicate that the RTB delivery module is a suitable system to mobilize β-gal enzyme across the blood-brain-barrier.

GM1-gangliosidosis: how can we better understand disease pathogenesis and move towards a clinical trial?

- ERT vs gene therapy – combinatorial approaches that could help tackling the disease at different cellular and organ sites
- Are monitoring enzyme activity and substrate reduction sufficiently accurate parameters to assess restored functionality?
- Understanding disease pathogenesis with the scope of identifying [novel] pathways that could be exploited therapeutically