Small molecule inhibition of glycogen synthase I restores autophagolysosomal and metabolic pathway dysfunction in a mouse model of Pompe disease

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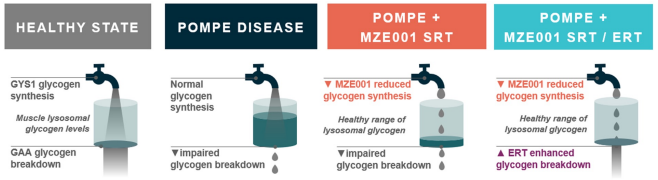
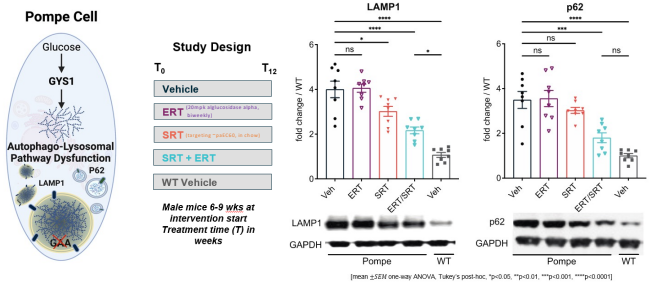
Introduction

Pompe disease is caused by loss of function mutations in the lysosomal enzyme acid alpha glucosidase (GAA), leading to pathological buildup of glycogen and ultimately cell death and muscle atrophy [1, 2]. Current standard of care is enzyme replacement therapy (ERT) with human recombinant GAA, which has been life saving for some Pompe patients, but it is not without significant limitations [3]. An alternative therapeutic approach to treating Pompe disease is to reduce glycogen synthesis by inhibiting glycogen synthase 1 (GYS1), the rate limiting enzyme for glycogen synthesis in skeletal and cardiac muscle, and preventing accumulation of glycogen in the lysosome. To evaluate the potential of this substrate reduction therapy (SRT) for Pompe disease, we developed a potent and selective small molecule inhibitor of GYS1. Our preclinical data demonstrate that SRT (alone, in combination with ERT, or after pretreatment with ERT), may be a viable therapeutic approach to treat Pompe disease.



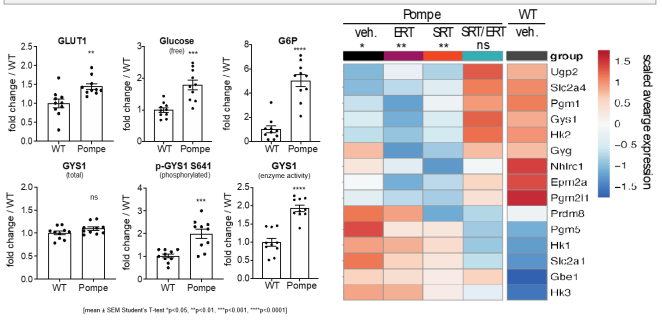
Substrate Reduction Therapy (SRT) for Pompe disease: novel mechanism of action

SRT + ERT reduces autophago-lysosomal pathway dysfunction markers in gastrocnemius muscle



Western blot quantification of gastrocnemius tissue lysate for LAMP1 and p62. Representative blots shown. Expression of LAMP1 and p62 is increased in Pompe mice compared to WT, suggesting lysosome accumulation and improper degradation of autophagosome cargo. SRT+ERT combination rescues autophagolysosomal function in Pompe gastrocnemius.

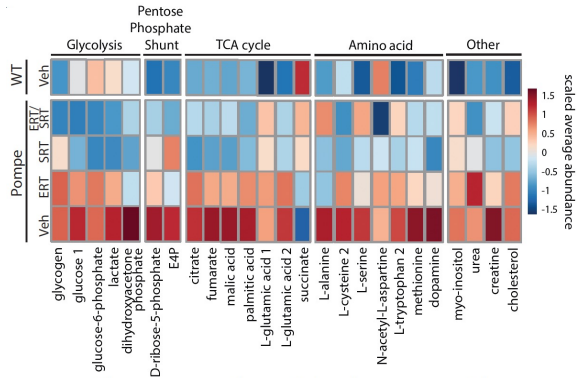
SRT + ERT normalizes glycogen biosynthetic pathway gene expression in Pompe mouse gastrocnemius muscle



Upregulation of glycogen biosynthetic pathway in Pompe. Increased GLUT1, free glucose and G6P levels, and increased GYS1 activity detected in Pompe gastrocnemius, suggesting that increased glycogen synthesis may contribute to Pompe pathophysiology.

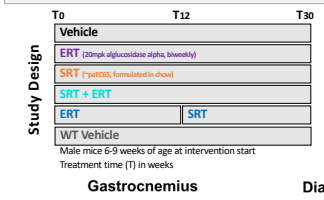
Heat map of gene transcripts relative to WT. Glycogen biosynthesis is upregulated in Pompe mouse gastrocnemius muscle and is normalized with SRT/ERT treatment.

SRT corrects global metabolic derangements in the Pompe mouse

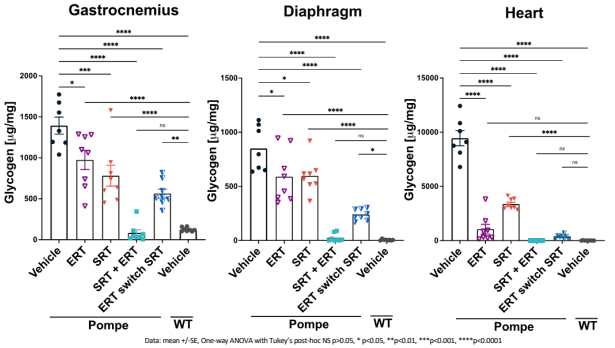


Heatmap of polar metabolites in Pompe mouse gastrocnemius relative to WT. Vehicle-treated Pompe mice exhibit dysregulation of metabolites in the central carbon and amino acid pathways. Clustering analysis reveal that ERT-treated Pompe mice most closely resemble vehicle-treated Pompe mice, Pompe mice receiving SRT, both alone and in combination with ERT, clustered more closely with WT mice.

Preclinical data support therapeutic potential of SRT in Pompe disease in multiple treatment paradigms



SRT has the potential to serve as monotherapy, combination therapy with ERT, and switch therapy post ERT to treat Pompe disease. SRT monotherapy significantly reduces tissue glycogen. SRT+ERT combination normalizes glycogen across multiple tissues to WT levels. SRT monotherapy after pre-treatment with ERT further reduces tissue glycogen.



Data: mean +/-SE, One-way ANOVA with Tukey's post-hoc NS p>0.05, * p<0.05, **p<0.01, ***p<0.001, ****p<0.0001

Conclusions

1. GYS1 SRT monotherapy in Pompe mice reduces glycogen build up across muscle tissues
2. Combination therapy of SRT and ERT normalizes tissue glycogen and corrects global metabolic derangements within 12 weeks
3. ERT treatment for 12 weeks followed by 14 weeks of SRT further reduced tissue glycogen in skeletal muscle and maintained healthy levels in heart
4. SRT may be a promising therapeutic approach as a monotherapy, combination therapy with ERT, or switch therapy post ERT for patients with Pompe disease

Acknowledgments & References

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