**Sanfilippo syndrome** is an autosomal recessive disorder characterized by the accumulation of sugar chain molecules called glycosaminoglycans (GAGs) in the lysosomes [1]. Children affected by Sanfilippo syndrome experience progressive neurodegeneration, developmental delays, severe dementia, immobility, and death before adulthood [2]. The most prevalent type of Sanfilippo syndrome, type A, is caused by mutations in the **SGSH** gene. This gene encodes enzyme sulfamidase, which is involved in the breakdown of heparan sulfate GAGs [3]. Mutations in SGSH results in a lack or deficiency of sulfamidase and thus a buildup of heparan sulfate, which ultimately prevents the lysosomal trafficking from working properly [4]. Despite the extensive research in this area, *it is unknown why GAGs accumulation in the lysosomes mostly affects the nervous system in Sanfilippo syndrome patients.*

My **objective** is to better understand how the mutation in SGSH and the accumulation of heparan sulfate in lysosomes can cause neurodegeneration in Sanfilippo syndrome type A patients. I will use mouse models, as the heparan sulfate biosynthetic pathway is highly conserved between mice and humans. Additionally, the brain complexity of mice and the ability to study their lifespan make them suitable model organisms. I **hypothesize** that mutated SGSH gene will result in decreased lysosomal degradation and GAGs accumulation in the lysosomes of neurons, ultimately leading to neurodegeneration as models age. My **long-term goal** is to provide insights into the neuropathology of Sanfilippo syndrome and ultimately gain new understanding of potential effective treatments.

**Aim 1: Identify conserved amino acids in SGSH that are important for GAGs accumulation in neurons**

**Rationale:** Deletion of conserved amino acids and then comparing the GAGs level in the neurons of mutants and wildtype mice models can indicate SGSH’s importance in lysosomal trafficking and neurodegeneration. Using mutants in different age groups can give insights to the impact of mutation on brain development.

**Approach:** Use NCBI Blast to determine homologs of the SGSH gene. Utilizing ClustalOMEGA, align all the sequences and identify conserved amino acids regions. Then use CRISPR/Cas9 to create mutant knockout mice. Lastly, develop a GAG assay to detect the GAGs level in neurons of wildtype and mutant mice aging 28-70 days old, which is equivalent to humans aged 2-15 years old [5,6].

**Hypothesis:** Mice with mutations in the conserved amino acid region of SGSH gene will show higher GAGs level in the lysosomes of neurons than the wildtype, and the accumulation increases with age.

**Aim 2: Identify small molecules that rescue neurodegeneration in SGSH mutants**

**Rationale:** There are currently no approved drugs to cure this disease. Treatment of SGSH mutant mice with small molecules can identify molecules that are able to restore the phenotypes of Sanfilippo syndrome and can be used as possible drug therapies.

**Approach:** Utilize a diverse chemical library on the wildtype and mutant mice to screen for small molecules associated with brain development and lysosomal degradation pathways. Identify chemical hits and screen for phenotype rescues by assessing the GAGs level.

**Hypothesis:** Small molecules associated with upregulation of lysosomal degradation and brain development will rescue the phenotype and reduce GAGs levels in mutant mice.

**Aim 3: Quantify proteins associated with neurodegeneration and lysosomal degradation**

**Rationale:** Identifying other proteins interactions and comparing their levels in wildtype and mutant mice can help elucidate the link between lysosomal GAGs accumulation and neurodegeneration.

**Approach:** Isolate brain tissues from wildtype and SGSH mutant mice. Use iTRAQ and mass spectrometry to determine protein relative abundancy. Sort the proteins of interest in Gene Ontology.

**Hypothesis:** Proteins involved in brain development and lysosomal degradation will be more abundant in the wildtype than mutant mice. These protein levels will also decrease with age in the mutant mice.

References

[[1]](https://curesanfilippofoundation.org/what-is-sanfilippo/)Cure Sanfilippo Syndrome Foundation. Retrieved from: <https://curesanfilippofoundation.org/what-is-sanfilippo/>

[[2]](https://www.ncbi.nlm.nih.gov/pubmed/25345095) Gilkes JA, Heldermon CD. Mucopolysaccharidosis III (Sanfilippo Syndrome)- disease presentation and experimental therapies. (2014). Retrieved from: <https://www.ncbi.nlm.nih.gov/pubmed/25345095>

[[3]](https://ghr.nlm.nih.gov/condition/mucopolysaccharidosis-type-iii?_ga=2.266501518.1071217777.1580939228-804763989.1579724316#statistics) Genetics Home Reference. Retrieved from: <https://ghr.nlm.nih.gov/condition/mucopolysaccharidosis-type-iii?_ga=2.266501518.1071217777.1580939228-804763989.1579724316#statistics>
[[4]](https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4664539/) Fedele A. O. (2015). Sanfilippo syndrome: causes, consequences, and treatments. Retrieved from: https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4664539/

[5] Kubaski F, Osago H, Mason RW, et al. (2017). Glycosaminoglycans detection methods: Applications of mass spectrometry. Retrieved from: <https://www.ncbi.nlm.nih.gov/pmc/articles/PMC5477676/>

[6] Dutta, S., & Sengupta, P. (2016). Men and mice: relating their ages. Life sciences, 152, 244-248.