Desired needs

- Develop a more physiologically relevant human model for studying APOE biology in sporadic Alzheimer's disease compared to traditional animal or 2D cell culture systems.
- Create an isogenic platform capable of isolating the effects of different APOE alleles (ε2, ε3, ε4) on Alzheimer's pathology.
- Establish a scalable and reproducible brain organoid system for future mechanistic studies and therapeutic screening.

Constraints

- Long-term organoid culture is vulnerable to microbial contamination, necrotic core formation, and culture instability.
- Batch-to-batch variability in organoid differentiation may affect reproducibility of APOE-associated phenotypes.
- Brain organoid development is labor-intensive and time-consuming, limiting high-throughput scalability.

Engineering Standards

- Organoid Standards Initiative (OSI) common guideline v1.0 informed reproducibility, organoid quality assessment, and endpoint validation.
- ANSI/ATCC ASN-0002-2022 standards guided iPSC authentication and cell line traceability.
- BSL-2 laboratory standards constrained handling of human-derived biological materials.
- Imaging and protein quantification workflows followed standardized immunofluorescence microscopy and western blot analysis procedures.
- Future organoid engineering standards may emerge from this work for genotype-specific neurodegenerative disease modeling.

Ethical, Environmental, and Societal concerns

a) **Ethical**
   - Human stem cell-derived tissue models require responsible handling, ethical sourcing, and accurate reporting of experimental limitations.

b) **Environmental**
   - Long term organoid culture consumes significant laboratory resources and energy.

c) **Societal**
   - Because organoid models cannot fully recapitulate the complexity of the human brain, responsible communication of research limitations is important to maintain public trust.

Active Teamwork and Leadership

- We contributed expertise in organoid differentiation, imaging, pathology analysis, and data interpretation.
- We appreciate and incorporated constructive feedback from our mentor and lab senior members into experimental design refinement.
- As the leader of this project, I coordinated experimental planning, delegated responsibilities, and guided the overall scientific direction of the team.

Motivating factors

- Approximately 1 in 4 people carries at least one APOE ε4 allele, and carrying two ε4 alleles can increase the risk of Alzheimer's disease by 8–12 fold. Yet the biology behind APOE-driven pathogenesis remains unclear, and there is still no effective treatment. The hope that human brain organoids may help us better understand Alzheimer's disease continually motivated me throughout this project. Even through failed experiments, long timelines, and repeated troubleshooting, I never wanted to give up because I believe this work can contribute, even in a small way, to the future of Alzheimer's research and therapy.