Systemic AAV gene therapy with CNS-targeted engineered capsids achieves significant GCase activity increases in the primate brain to support potential treatment of PD-GBA

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Introduction

Biallelic mutations in the GBA1 gene cause autosomal recessive Gaucher's Disease, the most common lysosomal storage disorder. More recently, epidemiological studies have revealed a role for mutations in this gene in the development of Parkinson's disease (PD), and current evidence suggests that heterozygous mutations within the GBA1 gene are present in up to 15% of PD patients, making it the most significant genetic risk factor[1].

The GBA1 gene encodes a lysosomal hydrolase enzyme, glucocerebrosidase (GCase), which metabolizes glycolipids within the lysosomal compartment. Mutations in this gene result in decreased GCase activity, leading to accumulation of glycolipid species which are thought to contribute either directly or indirectly to α-synuclein pathology, the pathological hallmark of PD[2, 3]. While certain aspects of the pathogenesis underlying PD-GBA remain unclear, substantial preclinical evidence suggests that interventions targeted to restore GCase activity in the brain could slow or stop the progression of PD-GBA[4, 5, 6].

Capsida is developing a gene supplementation therapy candidate (CAP-003) to be administered as a single intravenous (IV) infusion to PD-GBA patients. CAP-003 consists of an engineered AAV capsid that is designed to deliver the functional human GBA1 gene broadly across the CNS while de-targeting the liver and DRGs, which have been shown to be associated with safety issues in previous gene therapy programs. Using a loss-of-function mouse model we provide proof of concept pharmacology demonstrating that administration of the therapeutic cargo using a surrogate capsid results in dose-dependent increases in GCase activity that coincide with dose-dependent decreases in glycolipid accumulation. In non-human primates using the clinical candidate, we show that administration of CAP-003 at low to moderate doses results in robust expression across the entire CNS along with increases in levels of GCase protein and activity which show strong correlations with similar measurements in biofluids. Given that a ~30% decrease in brain GCase activity is expected in the patient population[7], the observed increases are expected to normalize activity levels, reduce glycolipids, and potentially slow or stop the progression of PD-GBA.

Methods and Materials

GBA1 LOF mouse model: GBA D409V KI (Jax Strain# 019106) and WT mice (Jax Strain# 005304) received a retroorbital IV injection at 8-weeks of age. After 4-weeks cargo DNA and RNA were assessed using PCR. GCase protein, activity, and glycosphingolipid content were assessed in CNS tissue using mass spectrometry. A surrogate capsid (CAP.B10) was used to deliver the therapeutic cargo enabling assessment of target engagement in CNS tissue achieved by a capsid that crosses the BBB in mice.

NHPs: The CAP-003 development candidate was administered IV to WT male and female cynomolgus macaques at ~33 months of age. After 6-weeks, DNA biodistribution and mRNA expression were assessed using ddPCR. GCase protein, GCase activity, and glycosphingolipid content were measured via mass spectrometry in both tissue as well as plasma and CSF to allow for early assessment of planned clinical target engagement biomarkers.

NHP-Driven Capsid Engineering Platform

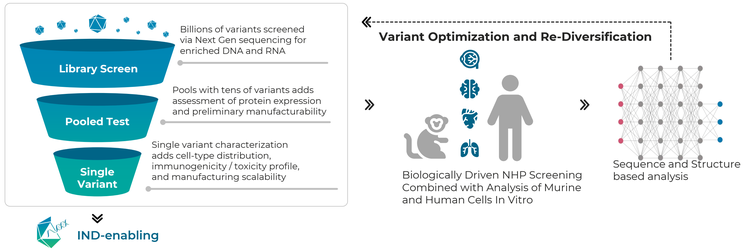

Figure 1. NHP-Driven Capsid Engineering Platform utilized to identify Capsid for CAP-003 for PD-GBA

Capsida's high-throughput screening process in NHPs identifies capsids that target desired tissues and cell types while de-targeting undesired tissues such as liver and dorsal root ganglion which have been associated with adverse events

Results

IV Administration of CAP-003 in NHPs shows expression >200-fold higher in the brain and shows significant de-targeting of the liver and dorsal root ganglion (~20-fold) relative to systemically administered AAV9

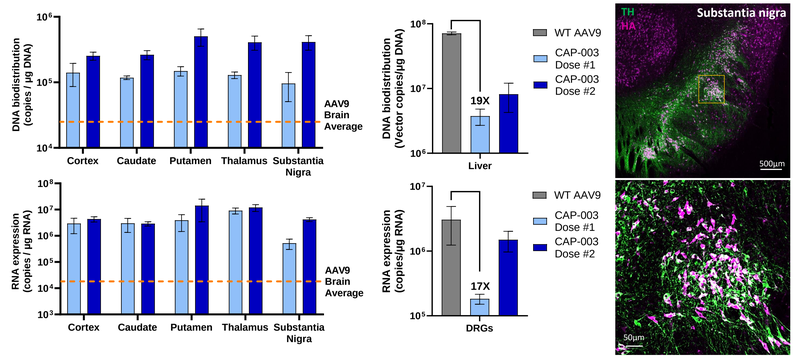

Figure 2. IV administration of CAP-003 in primates results in robust DNA biodistribution and mRNA expression across key areas of interest in the CNS while de-targeting peripheral organs including the liver and DRGs reducing safety risks associated with WT AAV9. AAV9 was IV delivered at a similar low dose. Representative histology in the substantia nigra was derived from a study that administered an epitope tagged version of CAP-003 to enable histological quantification of transduced cells.

In vivo target engagement in a GBA1 LOF mouse model following administration of CAP.B10-hGBA1 shows robust target engagement and significant reductions in GluSph

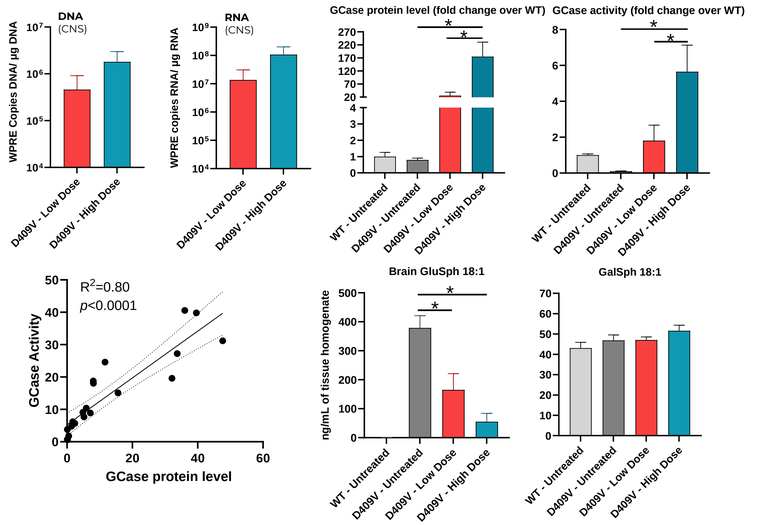

Figure 3. AAV treatment results in dose-dependent increase in GBA1 protein and GCase activity. These increases coincide with significantly reduced GluSph levels in the CNS and validate target engagement of the hGBA1 cargo. GluSph 18:1 = Glucosylsphingosine, GalSph 18:1 = Galactosylsphingosine

Results

Increases in GCase protein and activity in the NHP brain significantly surpass expected therapeutic threshold and correlate with key biomarkers

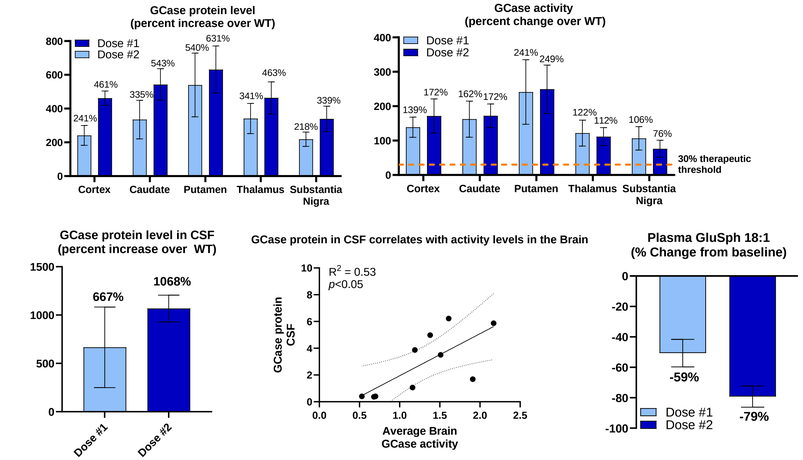

Figure 4. Administration of CAP-003 in primates results in up to 6-fold increases in GCase protein level and up to 3-fold increases in GCase activity in therapeutically relevant brain regions, exceeding levels that are expected to be clinically meaningful. Measurement of key fluid biomarkers show strong relationship to GCase activity in the brain across the doses tested.

	Limitations of Investigational Therapies	CAP-003 Differentiators
Neuronal Transduction	— Low neuronal transduction, especially in substantia nigra	+ Up to 70% of neurons transduced (57% in substantia nigra)
GCase Elevation	— Limited GCase elevation	+ GCase increases > levels needed to treat PD-GBA; reach 172% in cortex & 249% in putamen
Delivery	— Direct injection to the brain or CSF is invasive and results in inconsistent expression	+ Non-Invasive IV delivery limits risks and allows for broad coverage across the CNS
Safety	— Liver and DRG toxicity risks	+ No adverse histopathology findings in surveyed NHP organs, including liver and DRGs

Conclusions

- Capsida's engineering platform in NHPs has identified novel capsids with significant improvements in CNS penetrance and peripheral de-targeting relative to AAV9; CAP-003 is well tolerated and shows a favorable safety profile
- Proof-of-concept pharmacology in a GBA1 LOF mouse demonstrates dose-dependent increases in GCase protein levels and activity following administration of the hGBA1 cargo. These increases coincide with dose-dependent decreases in GluSph and indicate successful target engagement
- CAP-003 achieves broad CNS distribution in non-human primates, including high expression in regions impacted by PD-GBA pathology and increases in GCase activity expected to normalize levels in the PD-GBA patient population. Importantly, exploratory biomarkers show strong relationships to GCase activity in the brain which sets expectations for clinical outcomes in similar measures
- Capsida has initiated IND-enabling studies with the PD-GBA development candidate (CAP-003) with plans for IND filing in 1H 2025.

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References

1. Sidransky, E., Nalls, M. A., Aasly, J. O., Aharon-Peretz, J., Annesi, G., Barbosa, E. R., ... & Ziegler, S. G. (2009). Multicenter analysis of glucocerebrosidase mutations in Parkinson's disease. New England Journal of Medicine, 361(17), 1651-1661.
2. Smith, L., & Schapira, A. H. (2022). GBA variants and Parkinson disease: mechanisms and treatments. Cells, 11(8), 1261.
3. Migdalska-Richards, A., & Schapira, A. H. (2016). The relationship between glucocerebrosidase mutations and Parkinson disease. Journal of neurochemistry, 139, 77-90.
4. Sardi, S. P., Clarke, J., Viel, C., Chan, M., Tamsett, T. J., Treleaven, C. M., ... & Shihabuddin, L. S. (2013). Augmenting CNS glucocerebrosidase activity as a therapeutic strategy for parkinsonism and other Gaucher-related synucleinopathies. Proceedings of the National Academy of Sciences, 110(9), 3537-3542.
5. Rocha, E. M., Smith, G. A., Park, E., Cao, H., Brown, E., Hayes, M. A., ... & Isacson, O. (2015). Glucocerebrosidase gene therapy prevents α-synucleinopathy of midbrain dopamine neurons. Neurobiology of disease, 82, 495-503.
6. Sucunza, D., Rico, A. J., Roda, E., Collantes, M., González-Aseguinolaza, G., Rodríguez-Pérez, A. I., ... & Lanciego, J. L. (2021). Glucocerebrosidase gene therapy induces alpha-synuclein clearance and neuroprotection of midbrain dopaminergic neurons in mice and macaques. International journal of molecular sciences, 22(9), 4825.
7. Gegg, M. E., Burke, D., Heales, S. J., Cooper, J. M., Hardy, J., Wood, N. W., & Schapira, A. H. (2012). Glucocerebrosidase deficiency in substantia nigra of parkinson disease brains. Annals of neurology, 72(3), 455-463.