Pharmacological Inhibition of Glucocerebrosidase Impairs Memory and Motor Phenotypes in an Alpha-Synuclein Transgenic Mouse Model.

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Abstract

Mutations in the gene encoding glucocerebrosidase (GBA1) have been associated to synucleinopathies, Parkinson’s Disease (PD) and Lewy Body Dementia (LBD). Decreased glucocerebrosidase (GCase) activity has been noted in brain and CSF from PD patients. However, the relevance of the GCase activity in the development of the disease is still unknown. In the present study, we investigated the effects of a pharmacological inhibitor (CBE, 100mg/kg, 3 x week) on the behavioral responses in wild-type and alpha-synuclein mice. Treatment with CBE impaired cognitive and motor responses in the alpha-synuclein mice, suggesting that decreased GCase enzymatic activity can exacerbate behavioral deficits. These studies confirm the relevance of GCase activity in the development of synucleinopathies and suggest that disease severity can be exacerbated by decreased GCase activity as seen in PD patients with GBA1 mutations. Next steps include completing biochemical and histological analysis.

Introduction

Synucleinopathies: a group of neurodegenerative diseases characterized by an abnormal accumulation of α-syn in Lewy body neurites.

Parkinson’s Disease

2.2M patients in US and Europe

Loss of dopaminergic neurons

Progressive movement impairment

Lewy Body Dementia

1.4M patients in US

Loss of cortical neurons

Progressive Cognitive decline

Glucocebroside (GBA1) gene mutations are the most common genetic risk-factor for PD and LBD

Gldhcerosis (PND and LBD)

Gialcerebrosidase (GBA1) gene mutations

- Parkison’s disease exhibit 3-fold increased prevalence of PD

- Carrier of GBA1 mutations 1 in 10 in US present 3-fold increased prevalence of PD

- Gaucher patients exhibit classical CNS pathlogy (Levoy bodies) and accumuilation of α-synuclein depositions

A53T Alpha-Synuclein Transgenic Mouse Model

- Overexpress human α-syn throughout the CNS

- Develop a lethal movement disorder between 8-16 months of age

- Exhibit α-syn aggregates resembling neuronal inclusions in humans

- Show reduced GCase enzyme activity in the brain

Subjects and Groups

<table>
<thead>
<tr>
<th>Genotype</th>
<th>Dose</th>
<th>Injection</th>
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<tbody>
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<tr>
<td>Wild Type</td>
<td>CBE (100mg/kg)</td>
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<tr>
<td>A53T α-syn</td>
<td>CBE (100mg/kg)</td>
<td>3x/week</td>
<td>12</td>
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Materials and Methods

Novel Object Recognition: Mice were placed in an arena, as shown below, and the number of target investigations were recorded.

Pole Test: Mice were placed at the top of a vertical pole. The time it took to orient their body downwards and descend the pole was recorded. Four trials per animal were performed in succession.

Results

Nest Building: Nests were scored on a 1-5 scale approximately 18 hours after mice received nesting material. The remaining unused material was weighed.

Rotarod: After training, mice were placed on an accelerating rotarod (5-60 RPM) for 5 minutes. The time at which they fell from the rod (latency) was recorded.

Fear Conditioning: Mice were placed in a chamber for 6 minutes, during which they experienced three shocks, each paired with a tone. The following day, mice were placed back into the same box (context) for 5 minutes and cumulative freeze percentage was measured. Next, mice were placed into a novel context for six minutes, during which the associated tone was played for the entirety of the trial and cumulative freeze percentage was recorded.

Motor Function

C) A53T-α-syn outperform wild-type mice in the rotarod test. A53T-α-syn mice showed an increased latency to fall from the accelerating rotarod. In addition, GCase inhibition in WT and α-syn mice had no effect on rotarod performance, suggesting that gross motor skills are not affected by this treatment. All data represents the mean ± SEM (**P < 0.01). Bars marked with different letters are significantly different from each other (P < 0.01)

D) A53T-α-syn mice present deficits in fine motor skills that are exacerbated by GCase inhibition. A53T-α-syn mice exhibited an impaired ability to build a nest. GCase inhibition further diminished this ability as determined by nest scores or unused nesting material. All data represents the mean ± SEM (*P < 0.05).

E) A53T-α-syn mice present deficits in fine motor skills that are exacerbated by GCase inhibition. A53T-α-syn mice displayed a motor deficit in both orienting downwards and descending the pole. GBA1 inhibition with CBE increased this deficit in both tasks. All data represents the mean ± SEM (**P < 0.05).

Summary and Future Goals

Summary:

- The behavior of A53T-α-synuclein transgenic mouse model was further characterized using the aforementioned behavioral tests.

- A53T-α-syn mice exhibited deficits in memory function and fine motor skills. Gross motor function was enhanced in this transgenic animal.

- Pharmacological inhibition of GCase accentuated the memory impairment and fine motor deficits in α-syn mice.

Conclusion:

These studies confirm the relevance of GCase activity in the development of synucleinopathies and suggest that disease severity can be exacerbated by decreased GCase activity.

Future Steps:

- Next steps include completing biochemical and histochemical analysis to further validate the behavioral findings.

- Evaluate if these behavioral abnormalities seen in A53T α-syn transgenic mice can be ameliorated by GCase augmentation via AAV-GBA1.

References/Acknowledgements

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