Phenylbutyrate may stop progression of idiopathic Parkinson's disease

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Abstract

There is no drug therapy that can stop the progression of Parkinson's disease. In 2003, Bonifati and colleagues discovered that mutations in the DJ-1 gene cause familial Parkinson's. We took that observation and searched for drugs that could increase expression of DJ-1. We found that the short chain fatty acids butyrate and phenylbutyrate can upregulate the DJ-1 gene and protein in cultures of N27 dopamine neurons and protect those cells from hydrogen peroxide and misfolded proteins. In mice expressing a mutant human form of alpha-synuclein (Y39C) under control of the Thy1 promoter, both butyrate and phenylbutyrate increase expression of DJ1 protein in the brain and protect the animals from age-related motor and cognitive decline by stopping the aggregation of alpha-synuclein in neurons. The mechanism by which DJ-1 prevents protein aggregation is by increasing lysosome and exosome activity, thereby promoting transfer of alpha-synuclein from neurons into the bloodstream where the protein is eliminated. In mice, phenylbutyrate nearly doubled plasma alpha-synuclein concentrations compared to non-treated control animals. We have given the liquid formulation of the drug, glycerol phenylbutyrate, to 20 subjects with Parkinson's disease and to 20 age-matched controls. As in the mice, glycerol phenylbutyrate increased plasma alpha-synuclein from 50 to 150 per cent of baseline values indicating that the drug improved clearance of alpha-synuclein from neurons into blood plasma. Because phenylbutyrate is a relatively expensive FDA approved drug, we are evaluating the short chain fatty acid butyrate as a potential alternative for human therapeutic use. The successful treatment of transgenic mouse models of Parkinson's disease with butyrate and phenylbutyrate combined with human data that show phenylbutyrate can increase clearance of alpha-synuclein has provided the rationale for double-blind, placebo controlled trials of butyrate and phenylbutyrate to stop progression of Parkinson's disease in humans.

Questions:

- **Is glycerol phenylbutyrate safe and well tolerated in people with idiopathic Parkinson's disease?**
- **Can glycerol phenylbutyrate increase plasma alpha-synuclein concentrations in human subjects as a biomarker for increased clearance of alpha-synuclein from brain?**

Results

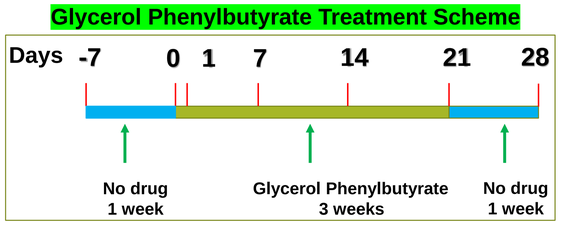

Figure 1: Phase I human clinical safety trial of glycerol phenylbutyrate. 20 subjects with recent onset idiopathic Parkinson's disease and 20 age-matched control subjects were recruited for a 28-day Phase I clinical trial. All subjects were given glycerol phenylbutyrate, 5.8 ml (6.4g), 3 times per day with meals, the maximum FDA-approved dose. Plasma and serum samples were collected at 7 times points throughout the study as indicated by the red hash marks.

Demographics

Groups	Parkinson patients	Normal subjects
Males	15	15
Females	5	5
Age	62 ± 2.4	61 ± 2.2
H&Y stage	1 or 2	N/A

Adverse Effects

Headache
Fatigue
Decreased appetite
Nausea, vomiting
Diarrhea
Skin rash

Plasma alpha-synuclein levels in Parkinson and Normal subjects

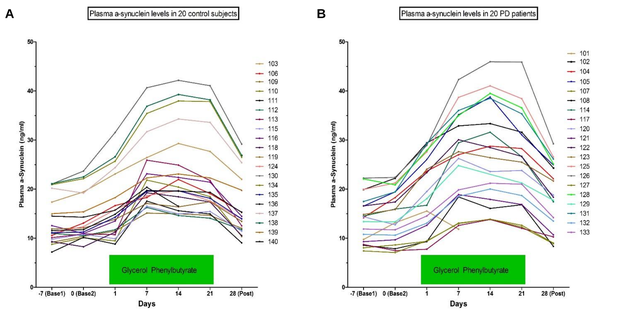

Figure 2: Baseline and peak plasma alpha-synuclein levels vary dramatically between patients regardless of PD. Plasma alpha-synuclein was assayed by ELISA . Profiles of plasma concentrations for each subject are presented in Panel A for normal subjects and Panel B for Parkinson subjects. Absolute values for each individual varied at baseline and after peak response to glycerol phenylbutyrate. Both groups showed increasing alpha-synuclein at Days 1-21 while glycerol phenylbutyrate was being administered..

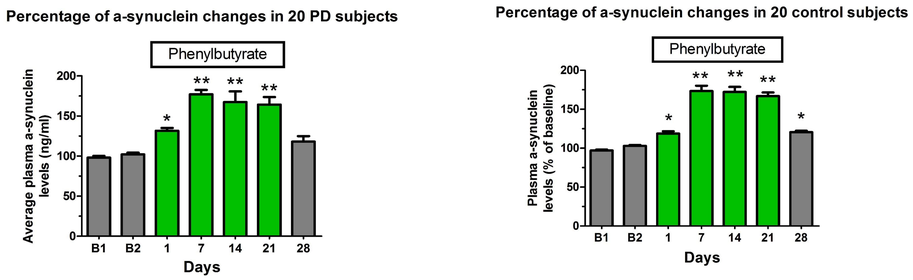

Figure. 3 Phenylbutyrate treatment increases plasma alpha-synuclein in Control and PD subjects. Plasma samples from each of the subjects taking glycerol phenylbutyrate were collected and analyzed for alpha-synuclein by a human α-synuclein ELISA kit (Invitrogen). Baseline measurements were made on two days prior to drug treatment (labeled "B1" and "B2,"). Drug was started, and plasma alpha-synuclein was measured after Day 1, 7, 14, and 21 of drug administration Drug was discontinued after Day 21, and a final alpha-synuclein level was measured at Day 28. Plasma alpha-synuclein concentrations were significantly increased by glycerol phenylbutyrate n both Parkinson and normal control subjects and remained significantly elevated 7 days after drug discontinuation (Day 28) indicating a 3-day half life for alpha-synuclein(*p<0.05, **p<0.01).

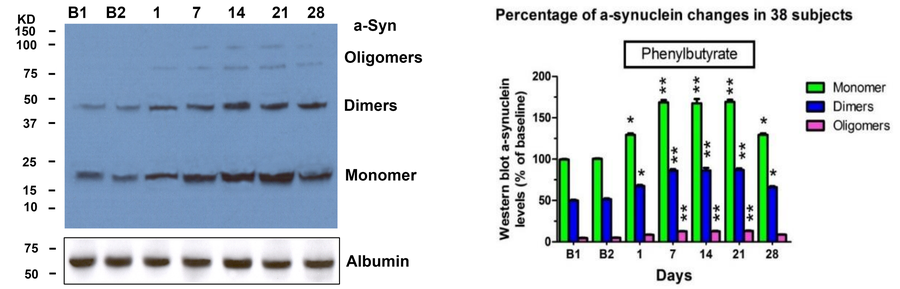

Figure 4: Percent plasma alpha-synuclein monomers, dimers, and oligomers increase in response to phenylbutyrate treatment. Western blot using α-synuclein primary antibody (Syn-1) was performed in 38 subjects. Protein levels were normalized with albumin as a loading control. Images were quantified using ImageJ software, and values were normalized to each individual's baseline plasma monomer α-synuclein concentration. Data show significant increases in monomeric, dimeric, and oligomeric alpha-synuclein in all groups. (*, $p < 0.05$; **, $p < 0.01$; n = 38).

Conclusions

- **Oral glycerol phenylbutyrate significantly increased the plasma concentrations of a-synuclein in both Parkinson patients and healthy age-matched controls, suggesting that the drug can increase clearance of a-synuclein from the brain of humans.**
- **Glycerol phenylbutyrate was generally well tolerated . Adverse effects including gastrointestinal complaints and skin rash were seen in about 20% of subjects.**
- **To test if glycerol phenylbutyrate can stop the progression of Parkinson's disease in humans as it has done in transgenic mice will require a double-blind, placebo controlled clinical trial.**

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