Mitochondrial-targeted reactive oxygen species scavenger JP4-039 prevents oxidative stress in cerebral cortex and striatum of glutaryl-CoA dehydrogenase-deficient mice

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Introduction

Glutaric acidemia type I (GA I; OMIM #231670) is an inherited autosomal recessive neurometabolic disease caused by mutations in the gene encoding the mitochondrial enzyme glutaryl-CoA dehydrogenase (GCDH) of the catabolic pathway of the amino acids lysine (Lys), hydroxylysine and tryptophan¹,².

The estimated worldwide frequency of GA I is between 1:30,000 to 1:100,000 newborns, being one of the most prevalent organic acidurias. Patients present at birth with macrocephaly and frontotemporal cortical atrophy and develop acute striatal degeneration mostly during encephalopathy crises. These crises are generally triggered by fever, infections or prolonged fasting and occur in two third of affected patients during the first 3 years of life¹.

Since the pathophysiology of cerebral cortex and striatum damage in GA I is not totally established and treatment is limited, the present study evaluated the effects of JP4-039, a mitochondrial-targeted reactive oxygen species (ROS) scavenger, on ROS production, antioxidant defenses and synaptophysin levels in cerebral cortex and striatum of GCDH-deficient (Gcdh−/−) mice.

Methods

Thirty-day-old wild type and Gcdh−/− mice received a high Lys chow (4.7%) for 3 days. During this period, the animals also received three intraperitoneal injections of JP4-039 (5 mg/kg/day; 1 injection per day) or vehicle (PBS+5% DMSO). After this, mice were euthanized, and cerebral cortex and striatum were dissected and used for the measurement of biochemical parameters³.
Results

Figure 1. Malondialdehyde (MDA) levels (A), 2’,7’-dichlorofluorescein (DCFH) oxidation (B), and catalase (CAT) (C), glutathione S-transferase (GST) (D), and superoxide dismutase (SOD) (E) activities in cerebral cortex of Gcdh+/+ and Gcdh−/− mice on a high lysine (4.7% Lys) diet in cerebral cortex of Gcdh+/+ and Gcdh−/− mice. Results are represented as mean ± standard deviation for three independent experiments (animals) per group. *P < 0.05, **P < 0.01, compared to Gcdh−/− mice injected with PBS+DMSO (Two-way ANOVA).

Figure 2. Immunoblot and densitometric analysis for heme oxygenase-1 (HO-1) (A) and synaptophysin (B) in cerebral cortex of Gcdh+/+ and Gcdh−/− mice on a high lysine (4.7% Lys) diet. β-actin was used as endogenous control. Results are represented as mean ± standard deviation for three independent experiments (animals) per group (Two-way ANOVA).
Results

Figure 3. Malondialdehyde (MDA) levels (A), 2’,7’-dichlorofluorescein (DCFH) oxidation (B) and glutathione S-transferase (GST) activity (C) in striatum of Gcdh+/+ and Gcdh−/− mice. Results are represented as mean ± standard deviation for three independent experiments (animals) per group. *P<0.05, **P<0.01, ***P<0.001, compared to Gcdh+/− mice injected with PBS+DMSO (Two-way ANOVA).

Figure 4. Immunoblot and densitometric analysis for heme oxygenase-1 (HO-1) in striatum of Gcdh+/+ and Gcdh−/− mice on a high lysine (4.7% Lys) diet. β-actin was used as endogenous control. Result are represented as mean ± standard deviation for three independent experiments (animals) per group (Two-way ANOVA).
Summary

Our results demonstrated that JP4-039 prevented the increase in MDA levels, DCFH oxidation and GST and CAT activities observed in cerebral cortex of Gcdh\(^{-/-}\) mice. Augmented SOD activity was only mitigated by JP4-039. In contrast, synaptophysin and HO-1 levels were not modified in cerebral cortex of Gcdh\(^{-/-}\) mice.

We also verified elevated MDA levels, and increased DCFH oxidation and GST activity in striatum of Gcdh\(^{-/-}\) mice and that these alterations were prevented by JP4-039. HO-1 content was not significantly modified in Gcdh\(^{-/-}\) mice.

These findings indicate that lipid peroxidation, ROS overproduction and disturbances in antioxidant defenses underlie cerebral cortex and striatum abnormalities observed in GA I patients. In addition, since JP4-039 mitigated all alterations observed in brain of Gcdh\(^{-/-}\) mice, we suggest that this molecule may be a promising strategy for adjuvant therapy of GA I.
References


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