Creatine in Huntington disease is safe, tolerable, bioavailable in brain and reduces serum 8OH2′dG

Abstract—In a randomized, double-blind, placebo-controlled study in 64 subjects with Huntington disease (HD), 8 g/day of creatine administered for 16 weeks was well tolerated and safe. Serum and brain creatine concentrations increased in the creatine-treated group and returned to baseline after washout. Serum 8-hydroxy-2′-deoxyguanosine (8OH2′dG) levels, an indicator of oxidative injury to DNA, were markedly elevated in HD and reduced by creatine treatment.

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Neurodegeneration in Huntington disease (HD) likely involves mitochondrial dysfunction, oxidative injury, and deleterious transcriptional alterations. Creatine is a high-energy phosphate donor that can buffer cellular energy depletion and secondarily relieve oxidative stress. Creatine treatment of transgenic mouse models of HD[1-3] delays motor dysfunction, extends survival, reduces huntingtin aggregation, reduces brain atrophy, and restores adenosine triphosphate depletion. We conducted a 16-week, randomized, double-blind, placebo-controlled study of the safety and tolerability of 8 g/day creatine in subjects with HD and assessed brain and serum biomarkers of creatine availability and activity.

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Methods. *Study design.* Sixty-four subjects were enrolled at four sites. Eligible subjects were randomized to 8 g/day of creatine monohydrate or placebo by computer-generated blocked randomization with site stratification. Treatment was administered as chewable wafers twice daily for 16 weeks. A medical monitor and independent safety committee reviewed clinical data monthly. Consent, study procedures, and case report forms were approved by the institutional review board at each site. Blood samples for analysis of serum biomarkers were obtained with consent from 30 age-appropriate individuals without neurologic illnesses.

Eligibility criteria. Eligible subjects were 18 years of age and older with a diagnosis of HD confirmed by genetic testing, a total functional capacity score of ≥5, and a caregiver to witness consent and monitor compliance. Exclusion criteria included previous creatine exposure within 30 days of baseline; underlying hematologic, hepatic, or renal disease; screening white blood cell count <3,800/mm^3; creatinine >2.0 mg/dL or alanine aminotransferase greater than twice the upper normal limit; or unstable medical/psychiatric illness.

Study procedures. Subjects were screened within 25 days of randomization (baseline visit). Screening included assessment of eligibility criteria, medical history, physical examination, Unified Huntington's Disease Rating Scale (UHDRS), EKG, DNA analysis, complete blood count, chemistry panel, and urinalysis. Subjects were on study drug for 16 weeks followed by an 8-week washout. Study visits occurred at baseline, weeks 8, 16, and 24 with telephone contacts at weeks 1, 10, and 20. Physical examination was done at screening and week 16. UHDRS scores, vital signs, adverse event assessment, and safety laboratory tests were repeated at all study visits. Blood was collected for serum creatine levels and serum 8-hydroxy-2′-deoxyguanosine (8OH2′dG), a measure of oxidative injury affecting DNA.[4-6] The latter was analyzed in the samples from two sites. Magnetic resonance spectroscopy (MRS; STEAM; TR/TE = 6000/20 milliseconds) of frontal cortex (voxel size = 56 mL) and occipital cortex (voxel size = 18 mL) performed at one site was analyzed[7] for this report. Subject compliance was assessed by wafer counts.

Statistical analyses. The primary outcome measure was tolerability. Subjects who did not complete week 16 or required more than one study drug suspension or any suspension exceeding 7 days were considered treatment failures. With 27 subjects per group, there was a 94% power to detect an absolute difference of 50% or more between the placebo group and the active treatment group assuming a 15% dropout rate. In accordance with intent to treat, all randomized subjects were included in safety and tolerability analysis. Tolerability was assessed by comparing the proportion of treatment failures in creatine and placebo groups using a

Table 1 *Baseline subject characteristics*

Variable	Creatine, n = 32	Placebo, n = 32
Gender (F:M)	12:20	14:18
Age, y (range)	44.7 (19–62)	47.9 (27–73)
Duration of illness, per rater, y	7.9 (3.5)	6.1 (5.4)
CAG repeat number	44.8 (2.6)	45 (5.5)
Weight, kg	76.5 (15.3)	77.0 (15.9)
Total functional capacity	9.0 (2.3)	9.3 (1.7)
Independence score	81.4 (11.4)	83.1 (9.2)
Total chorea score	13.3 (5.7)	13.4 (5.2)
Total motor score	46.8 (16.9)	42.8 (16.5)
Symbol Digit Modalities Test*	21.0 (8.5)	28.3 (10.9)
Verbal Fluency Test*	17.56 (9.1)	22.47 (12.6)
Stroop Interference	23.8 (1.4)	27.1 (11.7)

Values are means (± SD) or ratio. Scale ranges (normal to most severe) include total functional capacity (13 to 0), independence score (100 to 10), total chorea scores (0 to 28), total dystonia score (0 to 20), total motor score (0 to 124). For cognitive tests, higher scores are better: Symbol Digit (number of correct responses in 90 seconds), Verbal Fluency (number of correct responses in 3 minutes), Stroop Interference (number of correct responses in 45 seconds).

CAG = cytosine-adenine-guanine trinucleotide repeat numbers.

* $p < 0.05$.

Table 2 *Tolerability and adverse events*

Variable	Creatine, n = 32	Placebo, n = 32
Study withdrawal before wk 24	2	3
Subjects reporting AEs		
All AEs: mild, moderate, and severe, no. (%)	26 (86.9)	29 (90.7)
Serious AEs	3	3
AE		
Depression	4	5
Fall	3	3
Irritability	3	0
Lethargy	2	0
Dizziness	1	1
Increased movements	1	1
Vomiting	1	2
Dysphagia	1	0
Apathy	1	1
Cough	1	1
Hematuria	1	1

Values are frequencies and percentages. Differences between groups are not significant.

AEs = adverse events.

χ^2 test with continuity correction. As the primary interest was in detecting intolerability in the active group, a one-sided test was used (significance = 0.05). This was also applied to assess adverse events and laboratory abnormalities. The laboratory score changes from screening were analyzed by repeated-measures analysis of variance (ANOVA), and the differences between visits were analyzed by paired *t* test. Demographic and baseline variables were summarized for each group, and comparisons made using Fisher's exact test and continuous variables were compared using *t* test. Changes in UHDRS motor, cognitive, behavioral, and functional component subscores were measured at baseline and weeks 8, 16, and 24 and analyzed by mixed-model ANOVA.

Results. *Enrollment and baseline characteristics.* Sixty-four of 69 subjects screened were enrolled, 32 per treatment group (for a subject flowchart, see figure E-1 on the *Neurology* Web site at www.neurology.org). There were no differences between groups (table 1), with the exception of a higher rate of depression in the placebo group ($p < 0.02$). Normal controls donating blood for 8OH2′dG determinations consisted of 19 men and 11 women with an average age of 43.6 and a range of 21 to 78, indistinguishable from the subjects with HD.

Tolerability of creatine. Creatine at a dose of 8 g/day was well tolerated. Treatment assignment did not affect the likelihood of study completion (one-sided Fisher's test, $p = 0.50$). Of five subjects who terminated the study, two were on placebo (fall, difficulty with travel) and three on creatine (no specified reason, dysgeusia, dysphagia). There were six serious adverse events in three subjects in the placebo group (fall, vomiting, incorrect study drug) and three in the creatine group (fall, depression requiring hospitalization, suicidal ideation). The incidence of adverse events was similar between groups (table 2), and differences in the occurrence of any particular adverse event were insignificant. Compliance was comparable between treatment groups (90% vs 89%). Body mass index and UHDRS subscores were compared with baseline using paired *t* test and did not change with treatment (see table E-1 on the *Neurology* Web site at www.neurology.org). There were no significant laboratory abnormalities with the exception of mild reversible serum creatinine and alkaline phosphatase elevations (see table E-2). Serum creatinine (0.88 ± 0.19 to 1.04 ± 0.24, $p < 0.01$) increased by week 8 in the creatine group, remained elevated at week 16, and returned to baseline at washout. These elevations were statistically but not clinically significant and are consistent with creatinine being the primary metabolite of creatine.

Effect of creatine on serum and brain biomarkers. Average serum creatine levels in the treatment group rose from 46.4 μmol/L (SD = 35.7) at baseline to 382.5 μmol/L (SD = 259.9) at week 16 and back to 51.8 μmol/L (SD = 29.5) at washout; creatine concentrations were unchanged in the placebo group at each time point. By MRS, absolute creatine concentrations, estimated from the pool of total metabolites,[7] increased by 13% ($p < 0.05$) in the occipital cortex and by 7.5% in the frontal cortex in the ($p < 0.05$) by week 16 in subjects on creatine and returned to baseline at week 24. Brain creatine was unchanged in the placebo group.

Serum 8OH2′dG levels were significantly ($p < 0.0001$) elevated at baseline in subjects with HD (45.29 pg/mL, SD = 14.62) compared to normal controls (13.5 pg/mL, SD = 4.04). Treated subjects had an average reduction of 8OH2′dG of 9.11 pg/mL (SD = 13.93), whereas placebo subjects averaged an increase of 5.87 pg/mL (SD = 11.52)

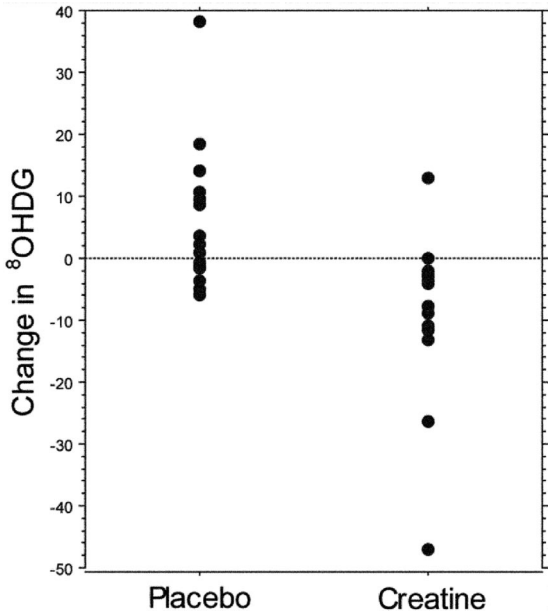

Figure. Changes in serum 8-hydroxy-2′-deoxyguanosine (8OH2′dG) levels in placebo and creatine treated subjects. Scatterplots showing individual changes in 8OH2′dG levels (pg/mL) at week 16 in the 32 subjects from two sites. The difference in the changes between treatment and placebo groups was significant ($p < 0.0042$).

(figure). The reduction from baseline to 16 weeks was significant in the treated group compared to the placebo group ($p < 0.0042$).

Discussion. Creatine, at a dose of 8 g/day, was well tolerated by subjects with HD, complementing previous studies using 3 to 10 g[8,9] daily. Clinical (UHDRS) measures were unchanged over the treatment course. Adverse events occurred with equal frequency in creatine and placebo groups. Reversible but mild increases in serum creatinine and alkaline phosphatase were seen by week 8 in subjects on creatine, returning to baseline levels at the washout visit. Serum and brain creatine concentrations rose significantly during treatment and returned to baseline levels after washout, indicating systemic and brain bioavailability of ingested creatine. A recent MRS study using 20 g/day creatine demonstrated similar increases in brain levels.[10] This is the first report of plasma 8OH2′dG elevations in HD, which were much higher than elevations reported in ALS[5] and Friedreich ataxia.[6] This finding is consistent with oxidative injury to DNA being an indicator of disease activity and contributing to pathogenesis. The improvement of elevated 8OH2′dG observed in this study suggests reduced oxidative injury in subjects with HD on creatine treatment and establishes 8OH2′dG as a promising peripheral biomarker.

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