CuII(atsm) protects against peroxynitrite-induced nitrosative damage and prolongs survival in an amyotrophic lateral sclerosis mouse model

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Supporting Online Material

Table S1: Mouse cohorts used in the Cu^{II}(atsm) treatment survival study

Group	Pre-symptomatic (140 days)	Symptomatic (200 days)
Wild-type: **Vehicle** **Cu^{II}(atsm)**	 n=5 (3m,2f) n=5 (1m,4f)	 not included
$SOD1^{G93A}$ **Vehicle** **Cu^{II}(atsm)**	 n=18 (10m,8f) n=14 (7m,7f)	 n=14 (6m,8f) n=13 (5m,8f)

Table S2. In vivo concentration of Cu^{II}(atsm) 2 hr after gavage

Mouse Strain	Plasma (ng/ml)	Brain (ng/g)	Spinal cord (ng/g)
WT (n=6)	303.6 (21.2)	69.2 (26)	18.8 (9.9)
$SOD1^{G93A}$ (n=3)	331.9 (9.5)	67.2 (13.5)	15.1 (3.2)

Data are expressed as mean with % standard deviation. No detectable Cu^{II}(atsm) was found in mice gavaged with the vehicle.

Supplementary figure legend

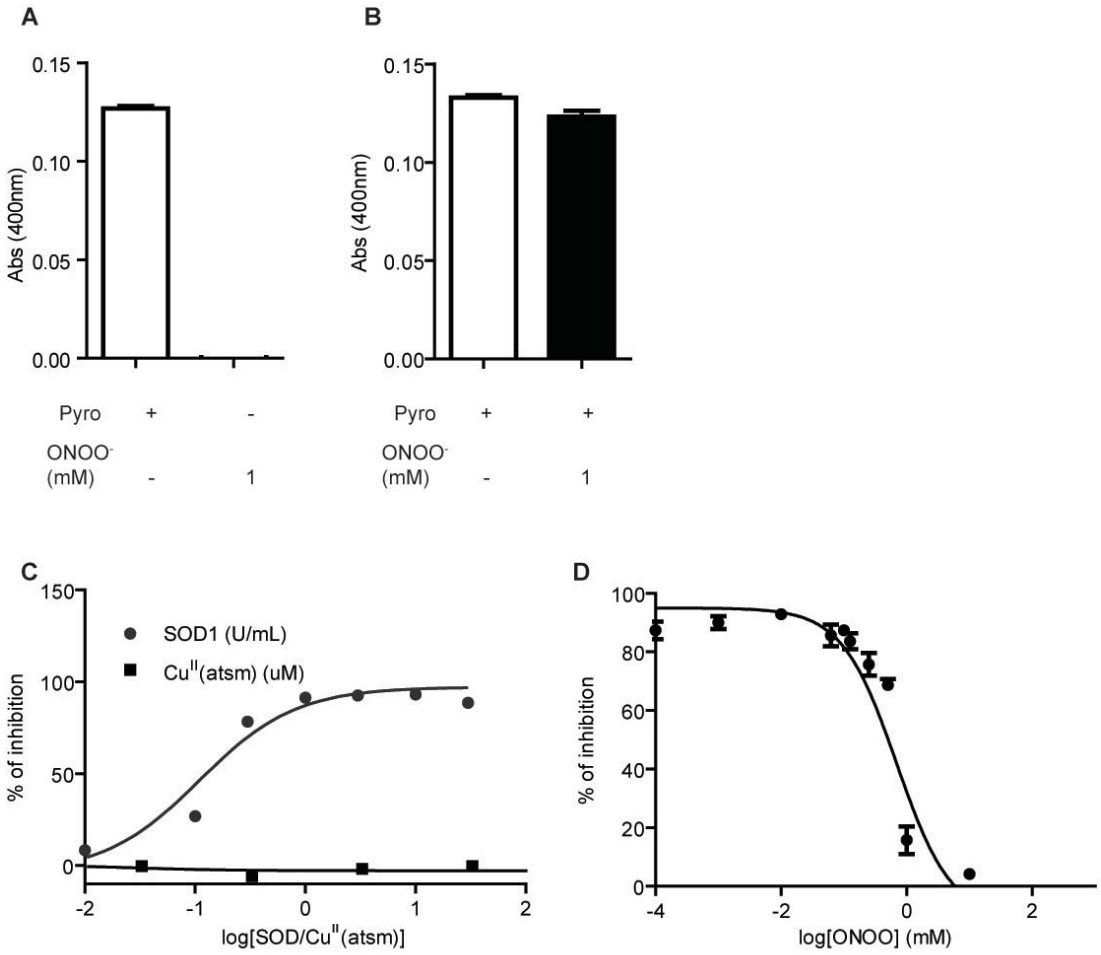
Figure S1. $ONOO^-$ has neither any interference at 400 nM (absorbance of pyrogallol) **(A)** nor pyrogallol oxidation itself **(B)**. SOD activity was measured as the ability to inhibit pyrogallol oxidation determined by absorbance at 400 nm after one hr incubation as in

Fig 1. Cu^{II}(atsm) also does not have any effect on pyrogallol oxidation **(C)**. $ONOO^-$ was able to inhibit the action of SOD1 (10 U/mL) in a dose dependent manner **(D)**.

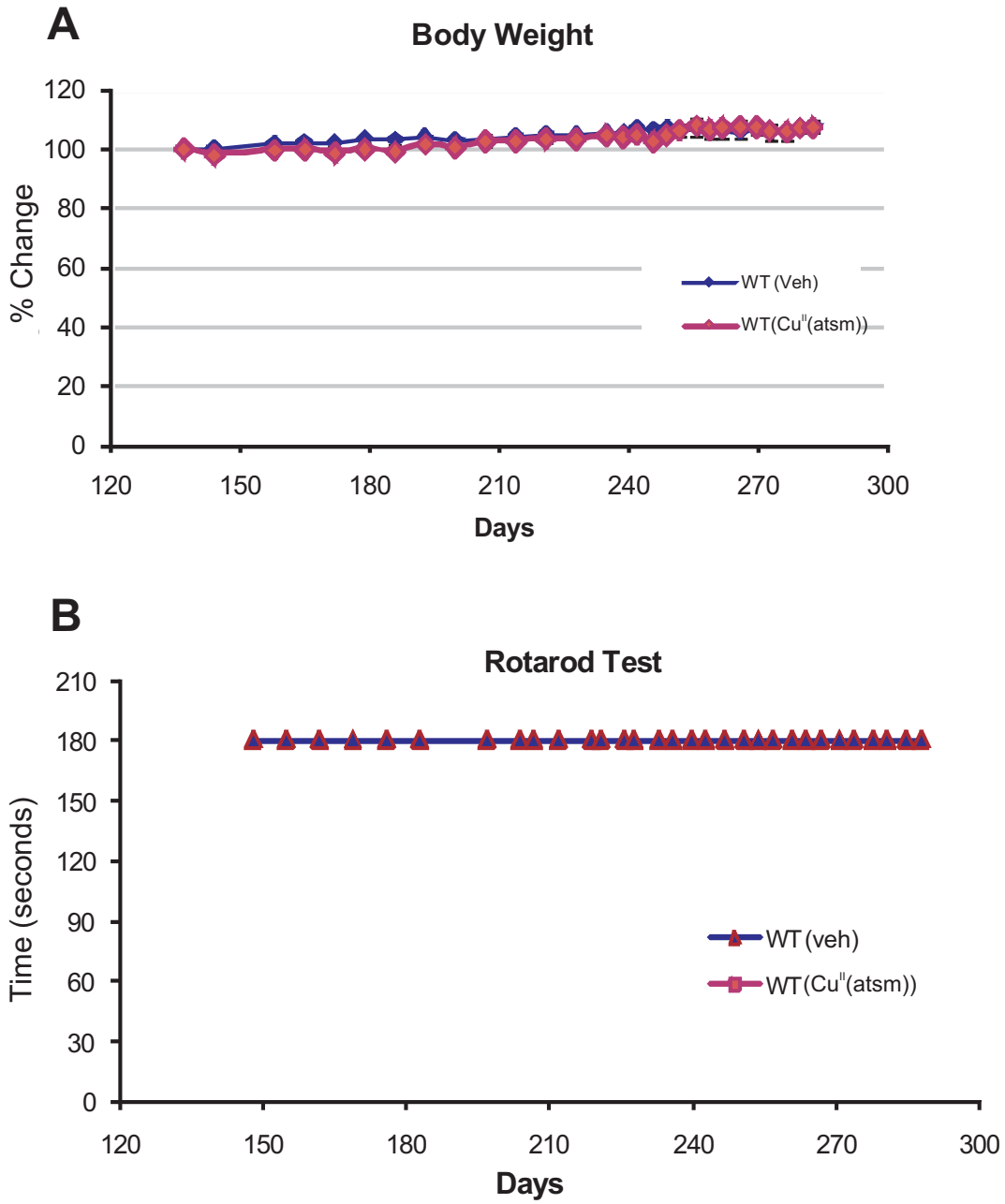
Figure S2. Effect of Cu^{II}(atsm) on the wild-type (WT) mice dosed at 30mg/kg from 140 days of age till the end of the $SOD1^{G93A}$ mice trial as Fig 2. (A). Percentage of weight changes over initial weight. (B). Rotarod test over the period of the trial. There is no difference between the vehicle and Cu^{II}(atsm) treated mice, indicating that the Cu^{II}(atsm) is well toleranced.

Figure S3. (A) A western blot of SOD1 in the 230 days mouse spinal cord homogenates. (B) Quantitation of the SOD1 level. No change in the SOD1 levels was detected between vehicle and Cu^{II}(atsm) treated $SOD1^{G93A}$ mice.

Suppl Figure 1



Suppl Figure 2



Supplementary Figure 3