SBT-272 Improves Mitochondrial Integrity and Motility in Upper Motor Neurons with TDP-43 Pathology

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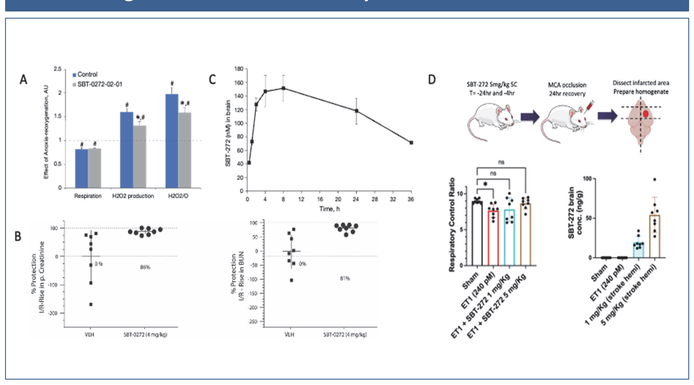
Abstract

Mitochondrial dysfunction is a major driver of pathophysiology in neurodegenerative diseases, including amyotrophic lateral sclerosis (ALS). SBT-272, a CNS penetrant compound that restores mitochondrial inner membrane structure and function under conditions of stress, is an investigational drug in phase I clinical trials. It is a rationally designed small molecule which binds to the mitochondrial phospholipid cardiolipin, preserving mitochondria function against ischemia/reperfusion in cardiac and kidney tissue. SBT-272 was neuroprotective in the SOD1^{G93A} mouse model of ALS (Keefe, et al. NEALS 2019). Since mitochondrial defects emerge as one of the major causes of upper motor neuron (UMN) degeneration with respect to TDP-43 pathology in ALS (Gautam et. Al. 2019), we investigated whether SBT-272 would improve mitochondrial integrity and the health of UMNs exhibiting TDP-43 pathology. Mixed cortical cultures were prepared from prp-hTDP-43^{A315T}-UeGFP mice, in which diseased UMNs are labeled by eGFP. Cultures were treated with (a) serum free medium, (b) SBT-272 (at 10, 100 and 1000 nM), (c) Edaravone (at 10, 100 and 1000 nM) and (d) a combination of sodium phenylbutyrate (PB, 100 uM) and tauroursodeoxycholic acid (TUDCA, 1 mM), the two components of AMX0035, respectively, for 72 hrs, prior to cellular analyses. SBT-272 significantly improved the structural integrity of mitochondria, enhanced their motility, and resulted in improved UMN axon outgrowth and arborization, indicators of improved neuronal health. While all three test compounds were active to different degrees, the effect of SBT-272 was significantly higher and well differentiated in this assay. The data supports further investigation of SBT-272 as a potential treatment for UMN degeneration in ALS with TDP-43 pathology.

Figure 1: In Vivo Pharmacodynamic effect of SBT-272



(A) Reduction of mitochondrial toxicity *in vitro* due to induced anoxia in permeabilized cardiac fiber. (B) SBT-272 normalized the plasma creatinine and BUN levels after acute kidney ischemia-reperfusion in rats. (C) SBT-272 rat brain pharmacokinetic (0.5 to 36 hr) after a single treatment (5 mg/Kg, sc). (D) SBT-272 (1.0 mg/Kg & 5.0 mg/Kg, s.c., treated 4 and 24 hours before injury) prevented the loss of mitochondrial respiratory control ratio in rat brain following cerebral ischemia-reperfusion injury induced via stereotactical delivery of 240 pM ET1 (aCSF in sham) in the piriform region. Each data point are oxygraph results (O2K, oroborous®) of individual animal. ET1 = endothelin-1 vasoconstrictive peptide (* P < 0.05 Kruskal Wallis followed by Dunn's test, # P < 0.05 compared to baseline respiration)

Results

Figure: 2: CSMN with hTDP-43 pathology can be visualized in prp-hTDP-43^{A315T}-UeGFP mice

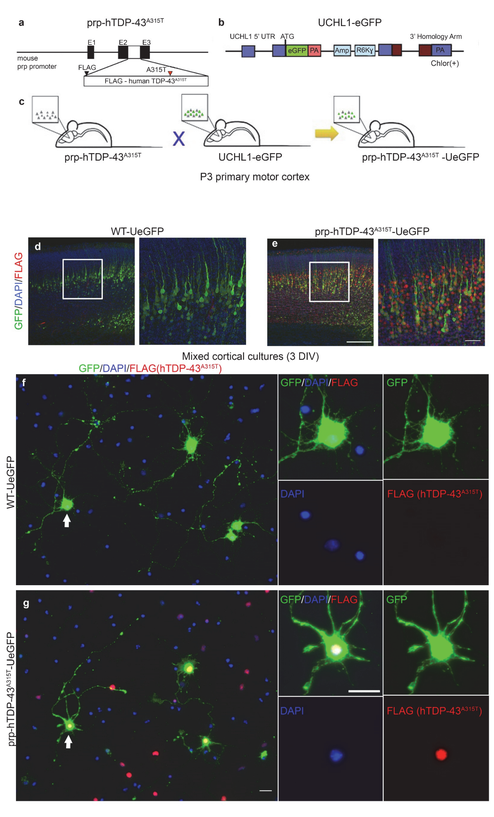


Figure: 4: SBT-272 treatment improved mitochondrial motility in hTDP-43+ CSMN

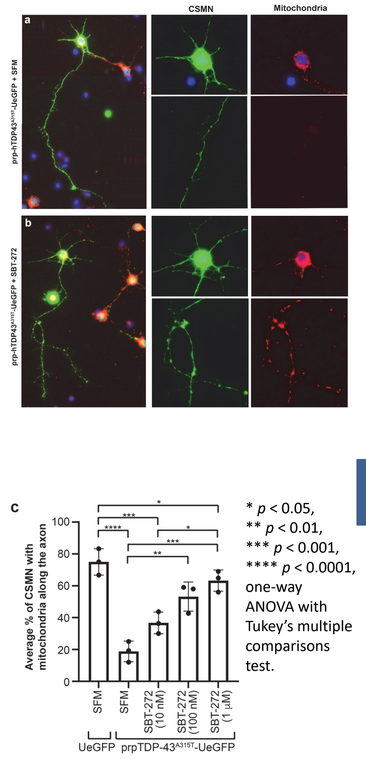


Figure: 5: SBT-272 had pronounced effect on axon outgrowth of diseased CSMN compared to Edaravone and AMX0035 at therapeutically meaningful concentrations

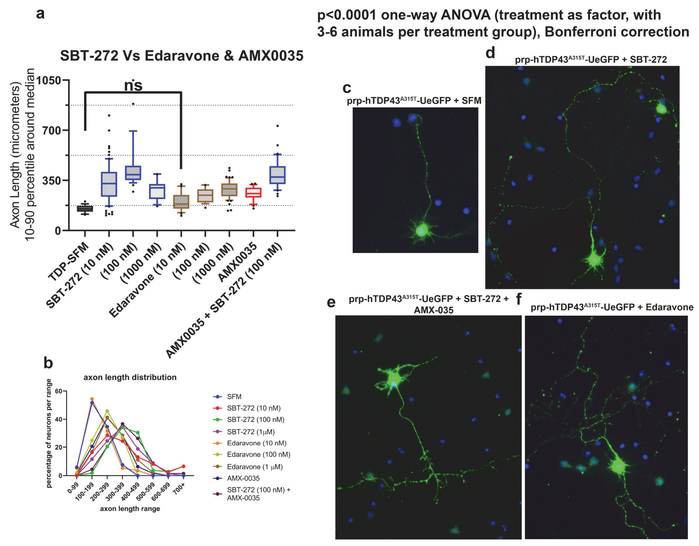


Figure: 6: SBT-272 treatment improves ultrastructural integrity of mitochondria in CSMN that are diseased due to TDP-43 pathology

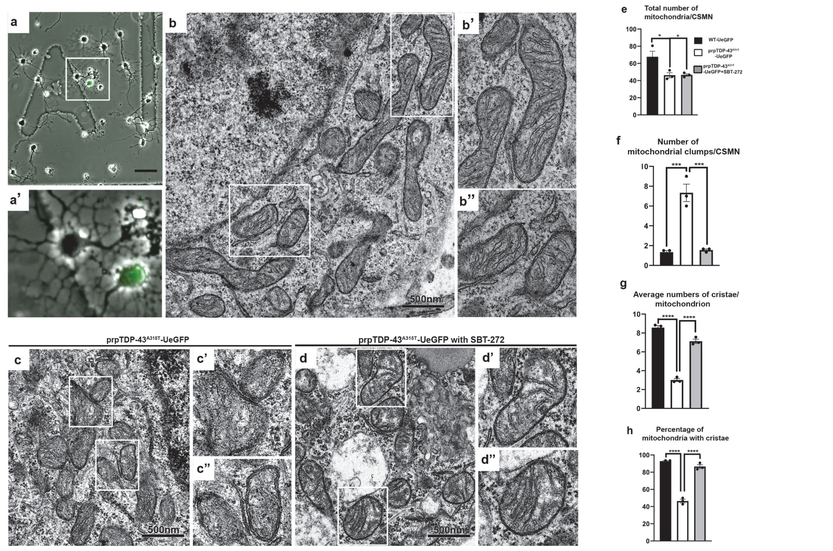


Figure: 3: Average axon length and the extent of branching/arborization are outcome measures to quantitatively assess CSMN health

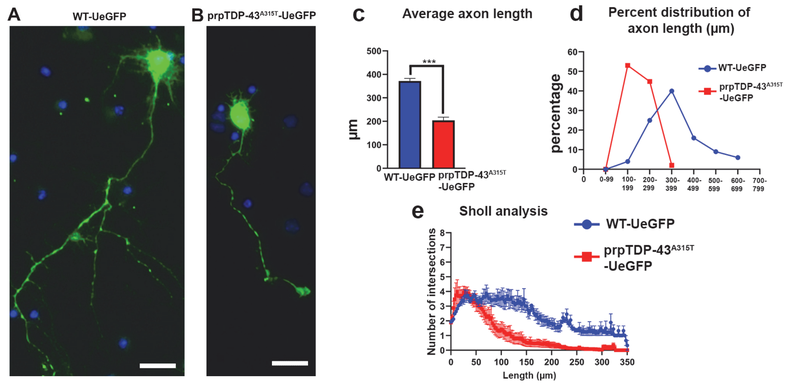
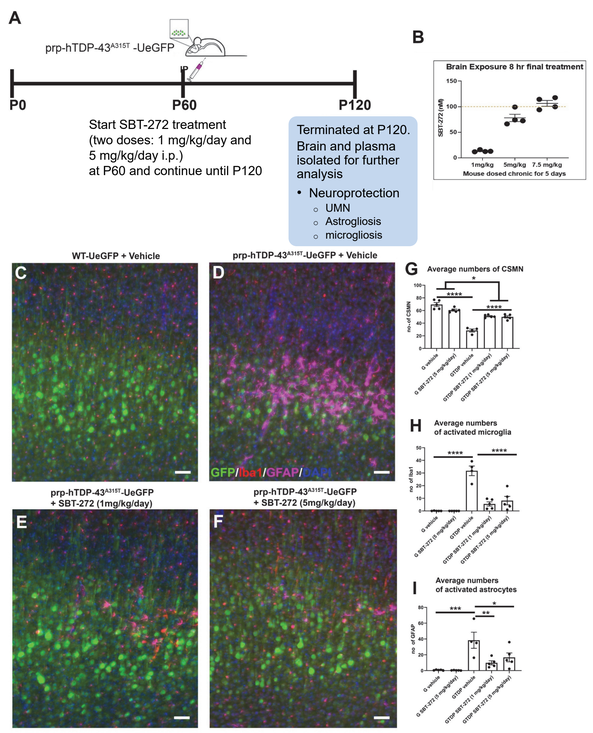


Figure 6-5: in vivo



Conclusions

TDP43 inclusions has emerged as the prominent clinical histopathological finding in sporadic ALS and ALS/FTLD patients. It has been shown to directly contribute to upper and lower motor neuron death through impact on mitochondrial structure and function. Here, by using a novel reporter line for UMNs diseased with TDP-43 pathology, we demonstrated that SBT-272, which is known to prevent the excess generation of mitochondrial reactive oxygen species (ROS) and to respiration through increased oxidative phosphorylation under conditions of cellular stress, protected and improved neuronal health of CSMN that is diseased due improve mitochondrial to TDP-43 pathology. These data support further investigation of SBT-272 for the potential treatment of motor neurons that become diseased due to TDP-43 pathology.

Future directions / Ongoing work

Based on improvement of diseased UMNs health and mitochondrial structural integrity by SBT-272 in an *in vitro* model of ALS, we are encouraged to investigate the effect of SBT-272 *in vivo* in prp-hTDP-43^{A315T}-UeGFP mouse model of ALS.

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References

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