

Grant conference

SMP, DRS, TSO and STH

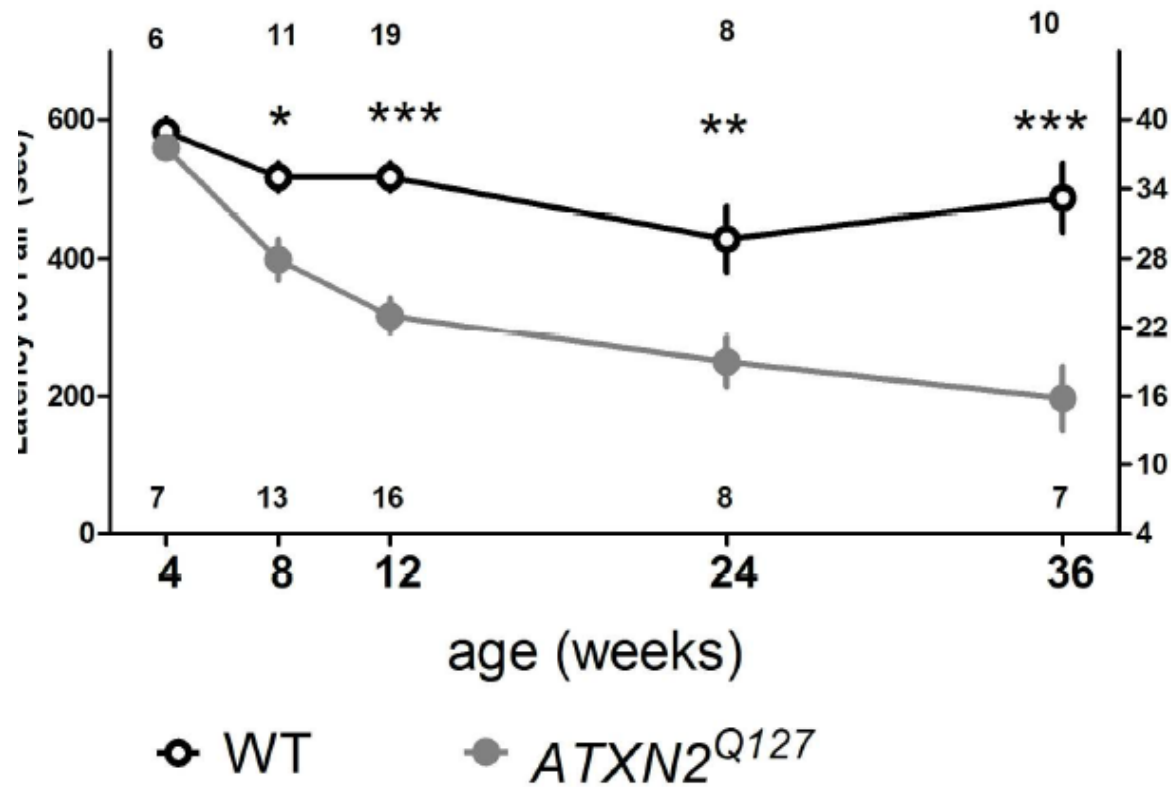
9/12/2012

Background

Mouse models:

Atxn2^{Q127}

BAC^{Q72}

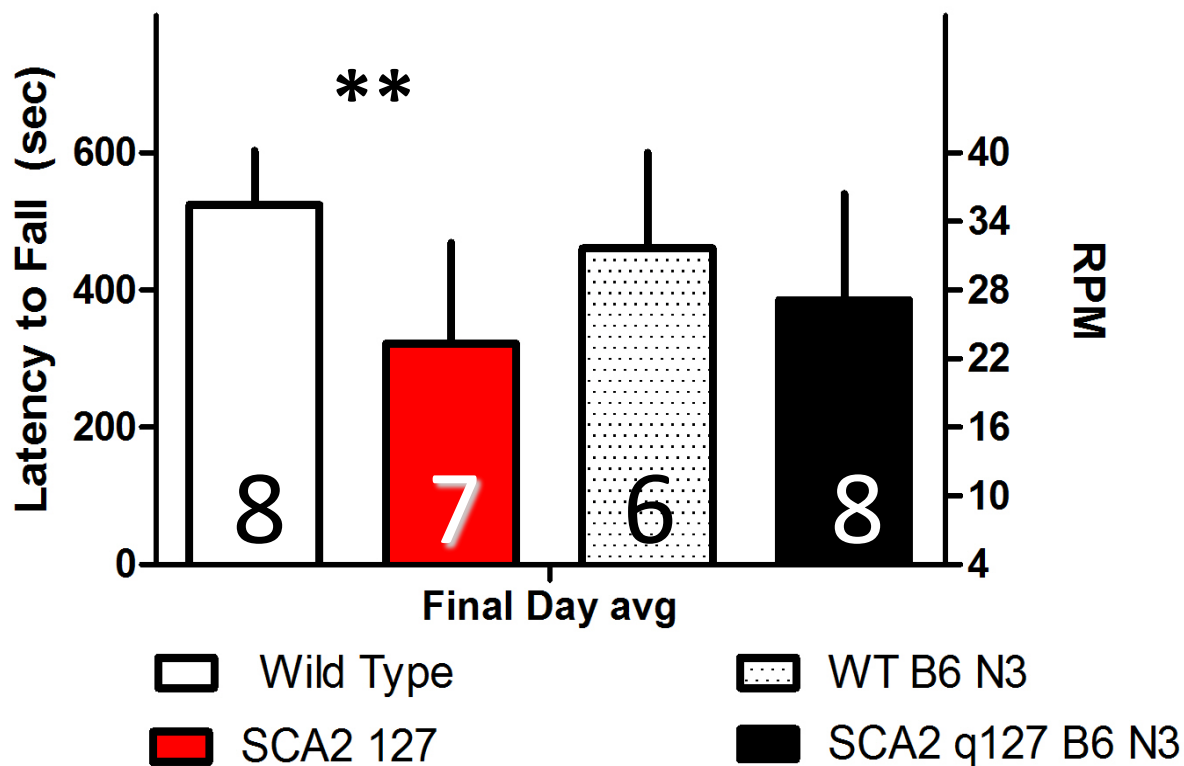


Behavioral Phenotype: accelerating rotarod assay

B6/D2 hybrid mice performance across age. Significant deficit in performance is observed in Tg mice at 8 weeks and becomes progressively worse with age.

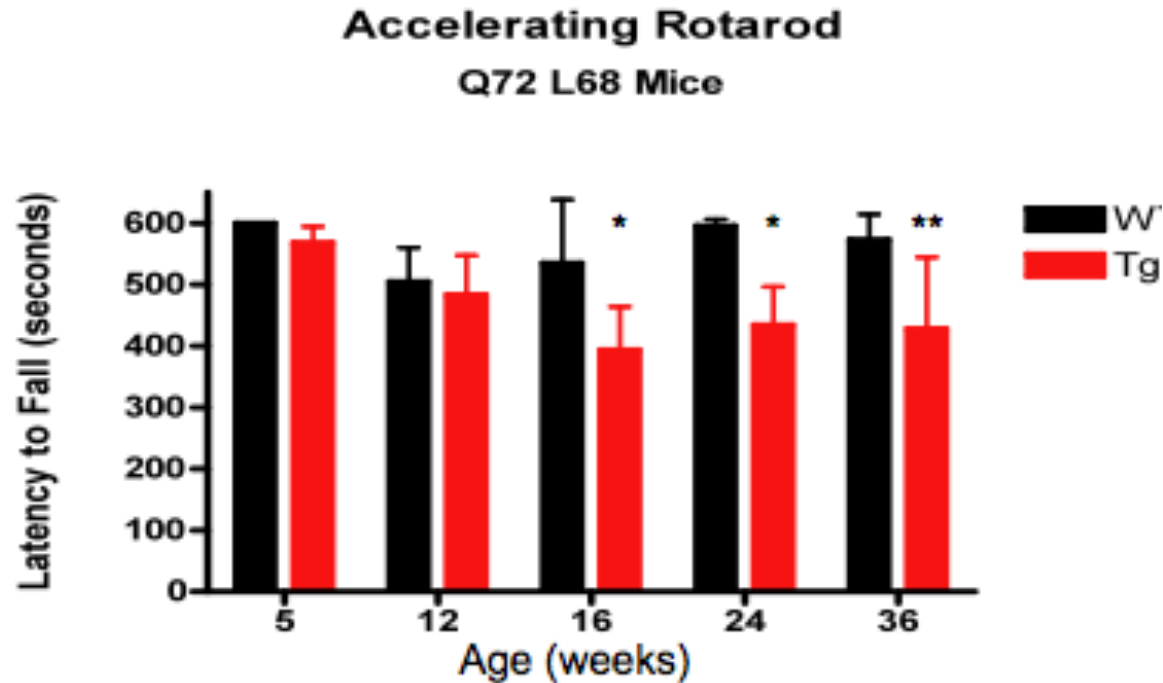
moving into B6 background

24 weeks old
Final day avg



Moving Atxn^{Q127} hybrid mice into a pure C57Bl/6J (B6) background has reduced the motor pathology and appears to delay the onset of the behavioral phenotype.

BAC Q72 Line 68 mice



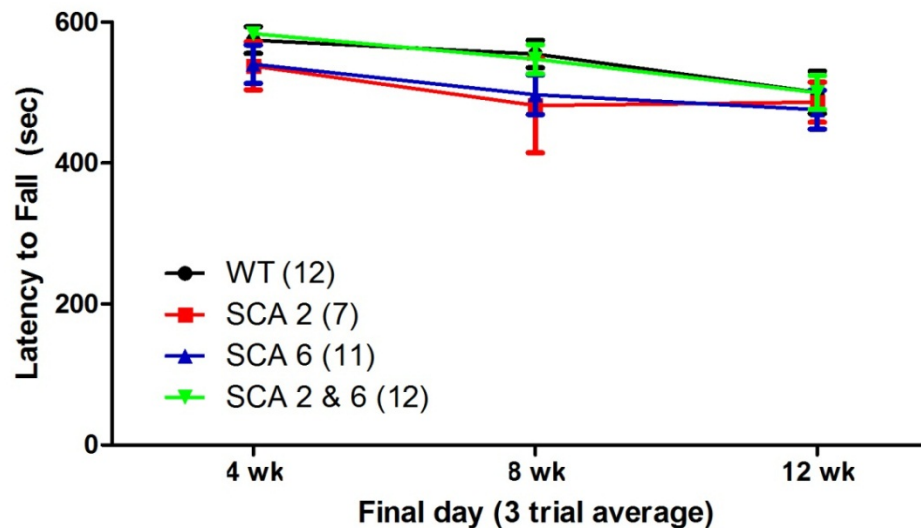
Behavioral Phenotype: accelerating rotarod assay

BAC Q72 mice performance across age. Significant deficit in performance is observed in BAC mice at 16 weeks and remains below WT performance out to 36 weeks. Controlled by the endogenous human *Atxn2* promoter these mice show a delayed onset of a behavioral phenotype.

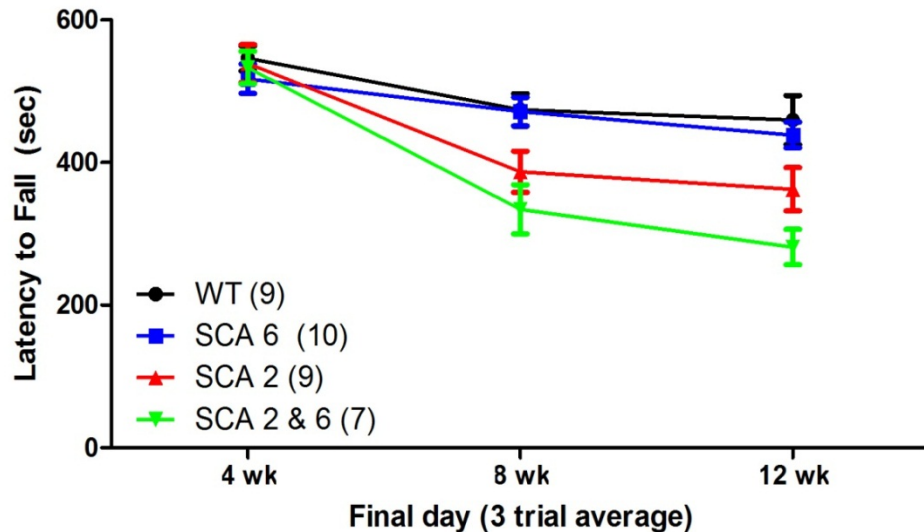
We have Q72 high and low expressing lines as well as Q22 control animals. These mice are in the Fvb pure background.

Modifiers of the Atxn^{Q127} phenotype

B6: SCA2^{Q127} x SCA6^{Q14}



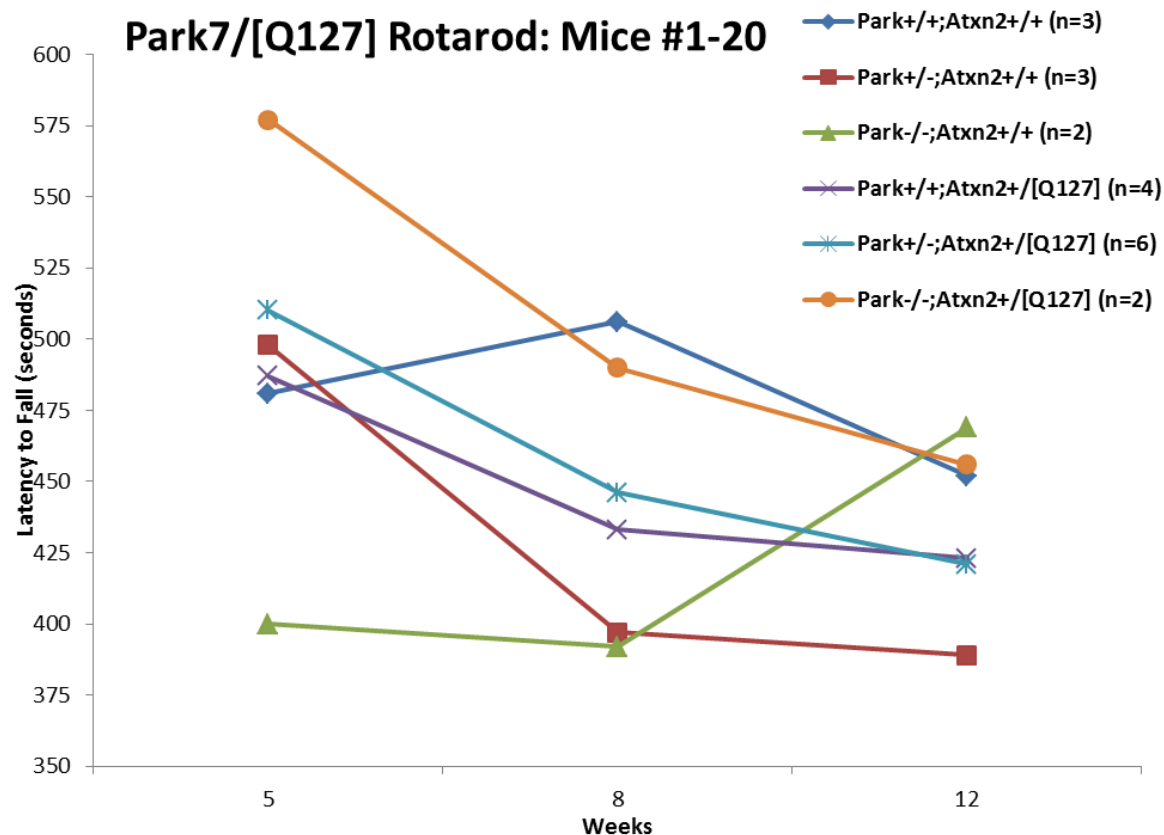
B6: SCA2^{Q27} x SCA6^{Q84}



Behavioral Phenotype: accelerating rotarod assay

Mice in the pure B6 background performance across age. Significant deficit in performance is observed in Tg Atxn2Q127 and double mutant mice at 12 weeks. Double mutant mice are more different from WT than Tg alone. SCA6^{Q14} does not appear to have an effect on phenotype.

Modifiers of the Atxn^{Q127} phenotype

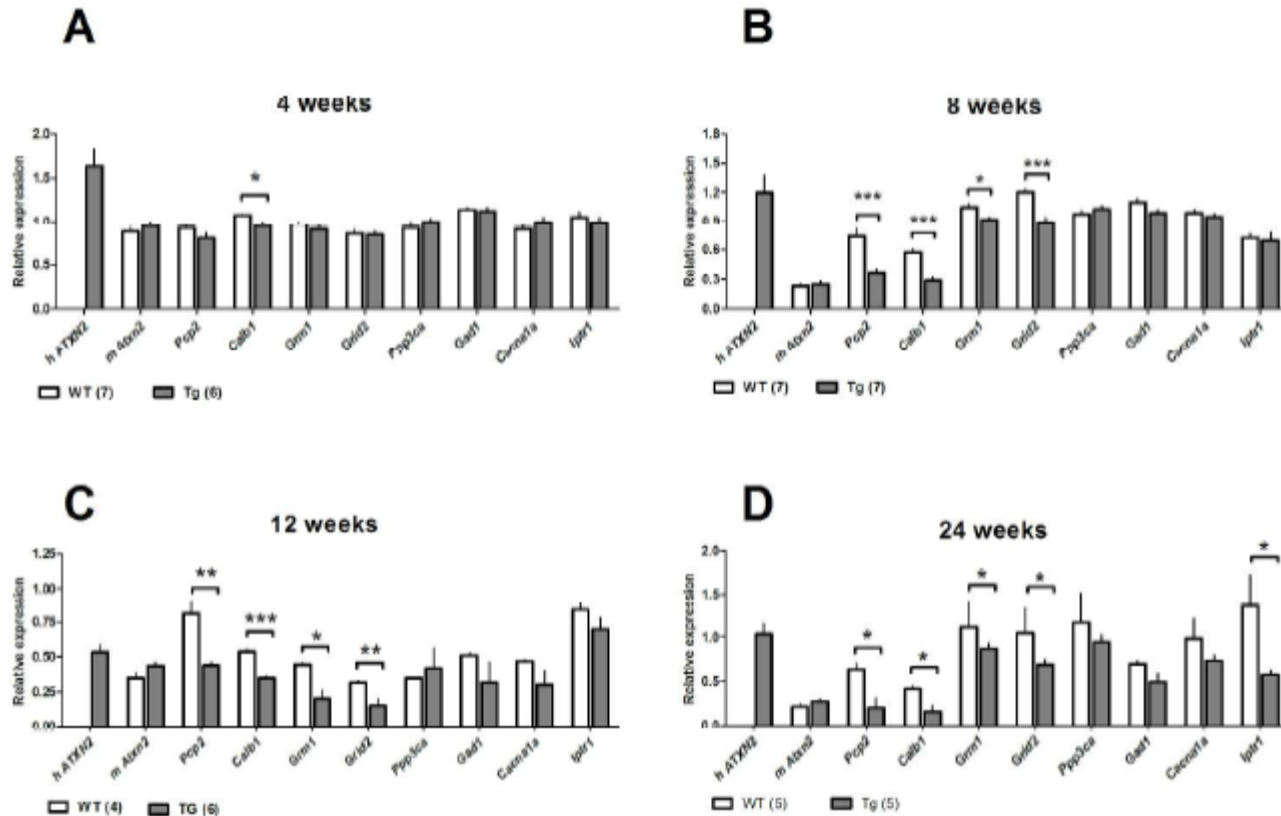


Behavioral Phenotype: accelerating rotarod assay

Pure B6 mice performance across age. Significant deficit in performance is observed in Tg mice at 8 weeks and becomes progressively worse with age.

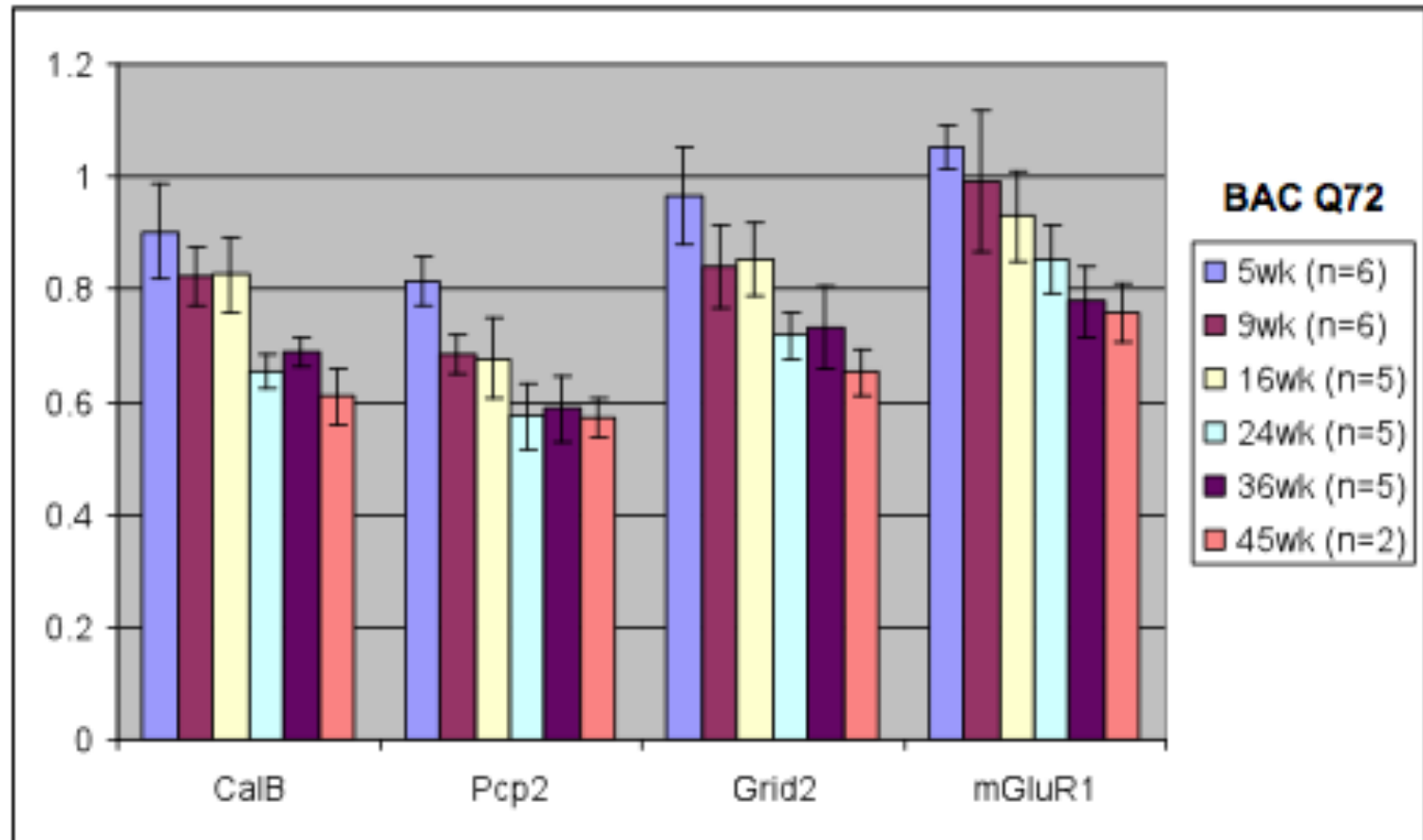
Small N size. We're continuing to breed and collect data for later ages (20wks).

Transcriptional Changes in Atxn2^{Q127}



Hybrid mice changes in a RNA transcription as a function of Tg expression. Significant deficit in Calbindin 28k is observed in Tg mice at 4 weeks and becomes progressively worse with age. At 8 weeks Calbindin, Pcp2, Grid2 and mGluR1 are down regulated.

Transcriptional Changes in BAC^{Q72}



qPCR assay

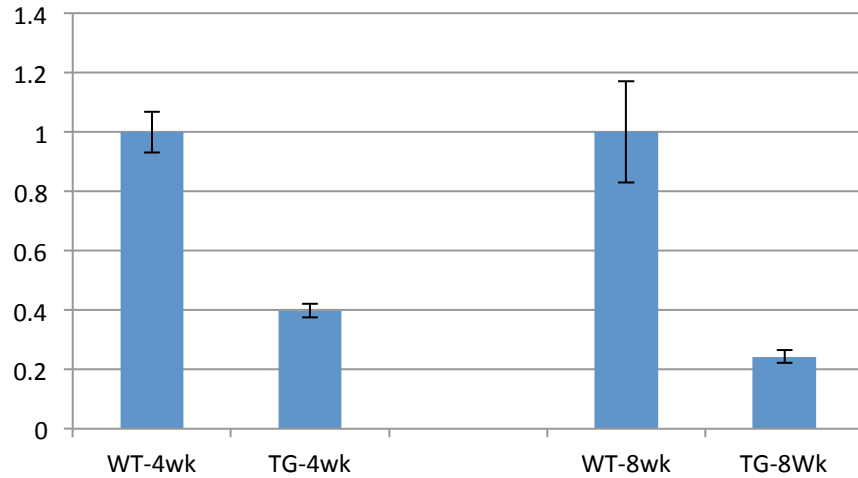
FVB mice changes in a RNA transcription as a function of Tg expression.

Transcriptional Changes in Atxn2^{Q127}

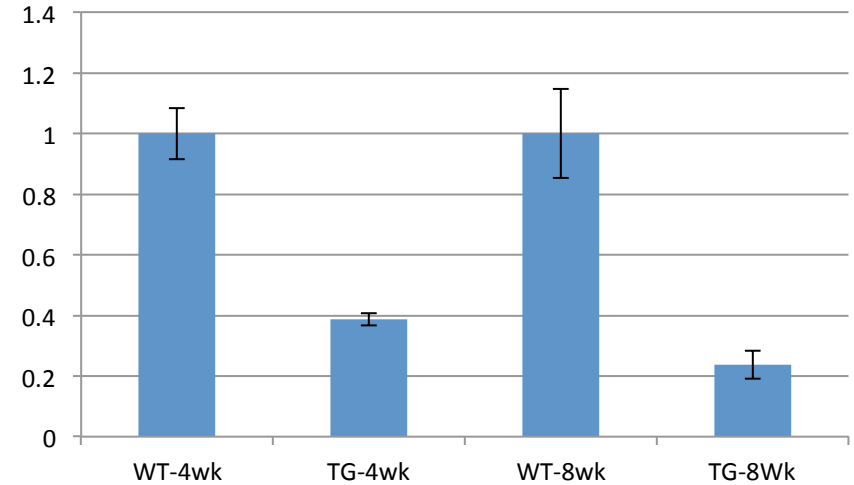
- RNA sequencing:
 - Hybrid B6/D2
 - Comparing WT and Tg: 4 and 8 weeks of age
 - Identified many candidate genes for markers of cellular pathology and receptor pathology
 - Rgs8, Fam107B, Gpr63 etc.
 - Confirmed with qPCR

Confirmation of RNA-seq with qPCR in *Atxn*^{Q127} mice

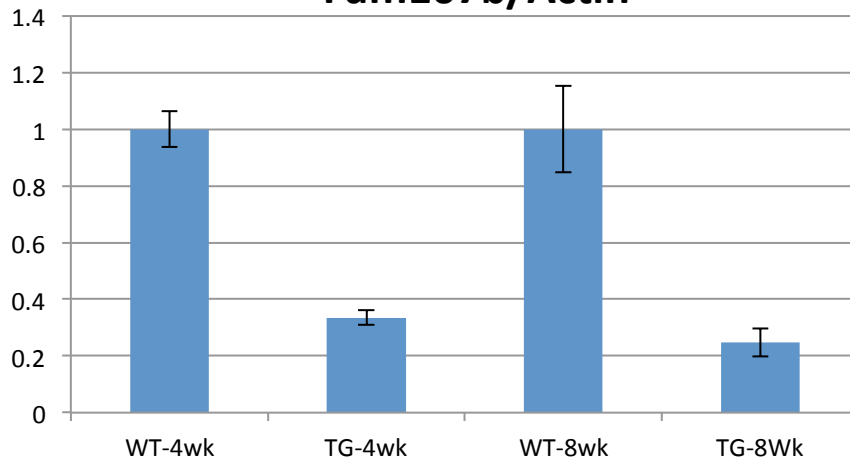
Gpr63/Actin



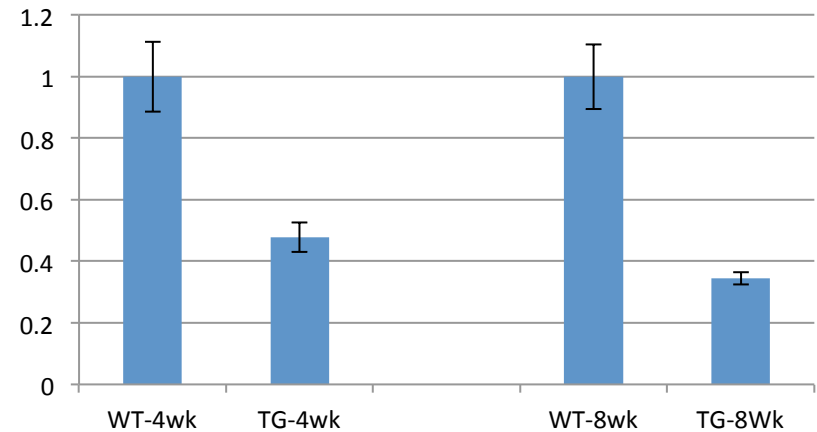
Rgs8/Actin



Fam107b/Actin

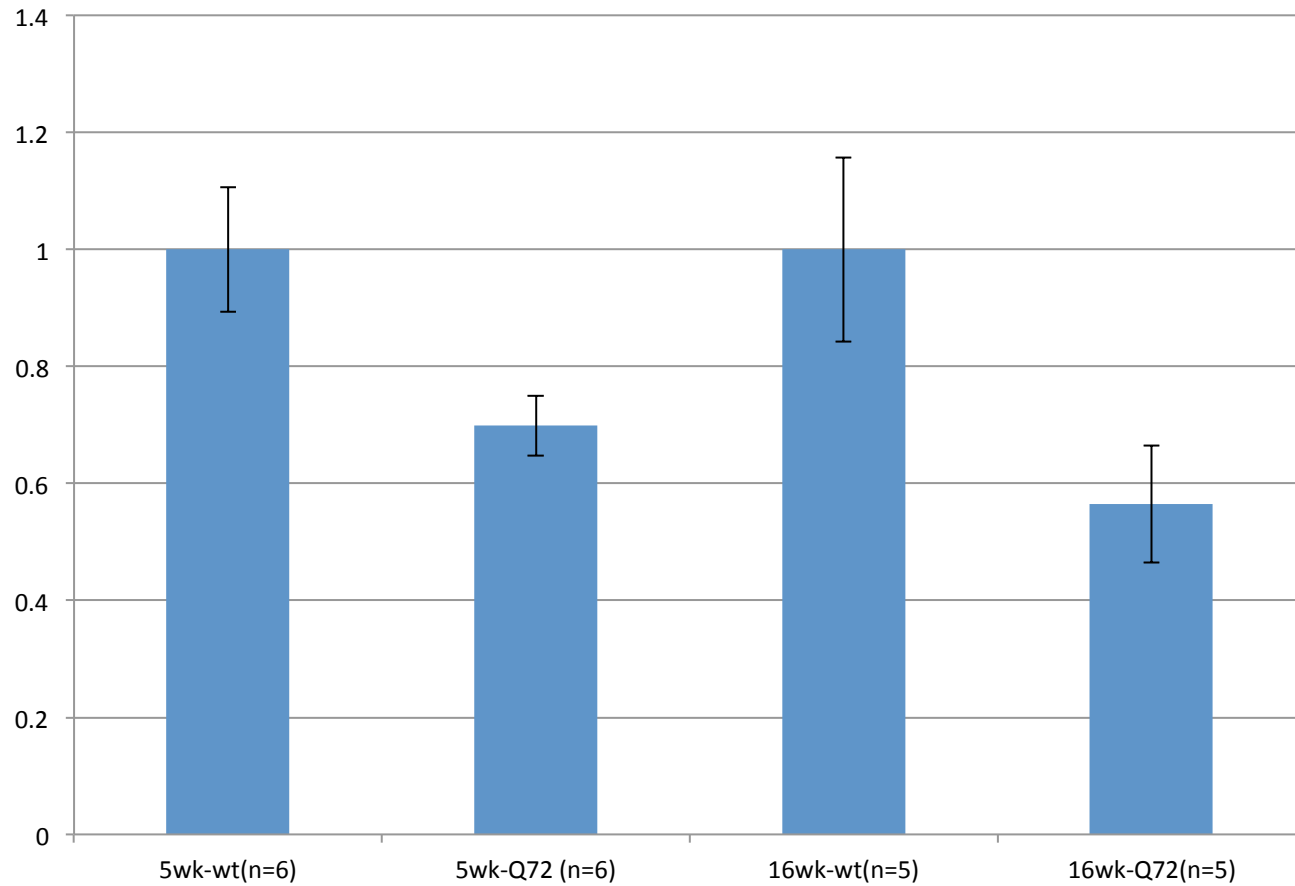


Cep76/Actin



qPCR in BAC mice

Rgs8/Actin



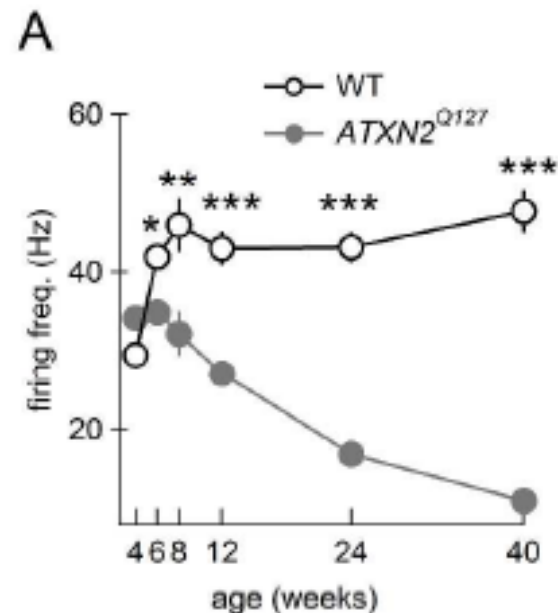
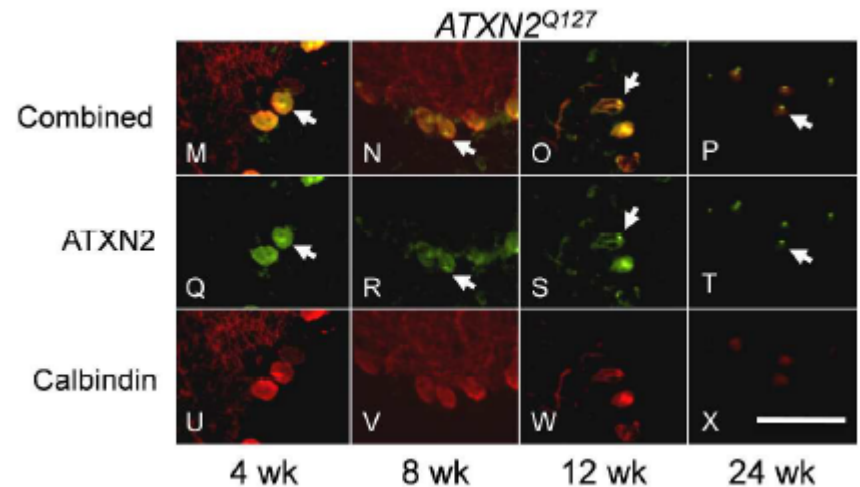
RNA seq with iPS

- iPS dedifferentiation time course
 - Do we have RNA seq data on iPS comparing patient vs. normal?

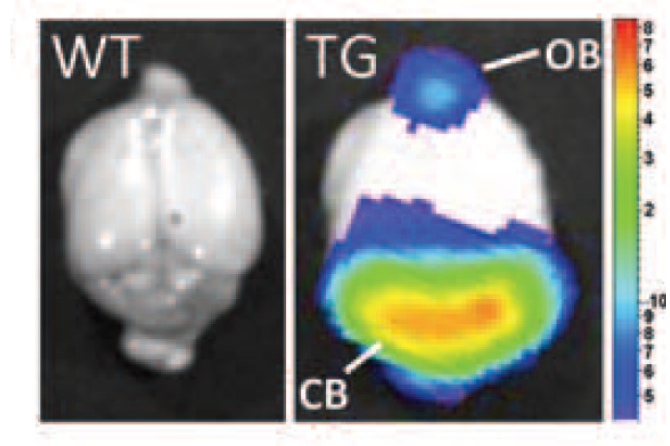
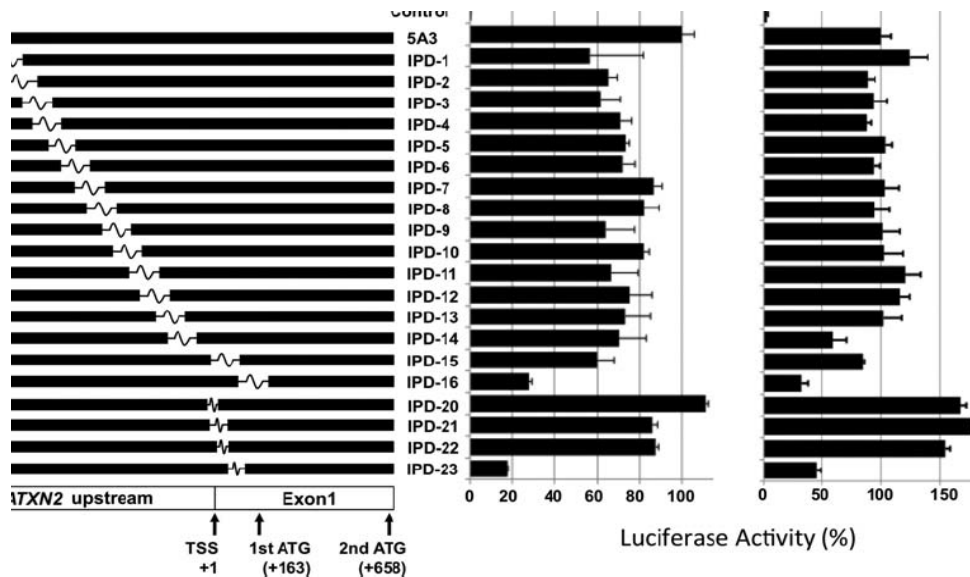
Work leading to publication

Characterization of Atxn^{Q127} mouse

- Early transcriptional changes: calbindin (4wk), Pcp2, Grid2, mGluR1 (8wk)
- Histology: Gross changes in the molecular layer (12wk) and detection of inclusion bodies (4wk).
- Behavioral deficits detected as early as 8 weeks
- Electrophysiology deficits at 6 weeks

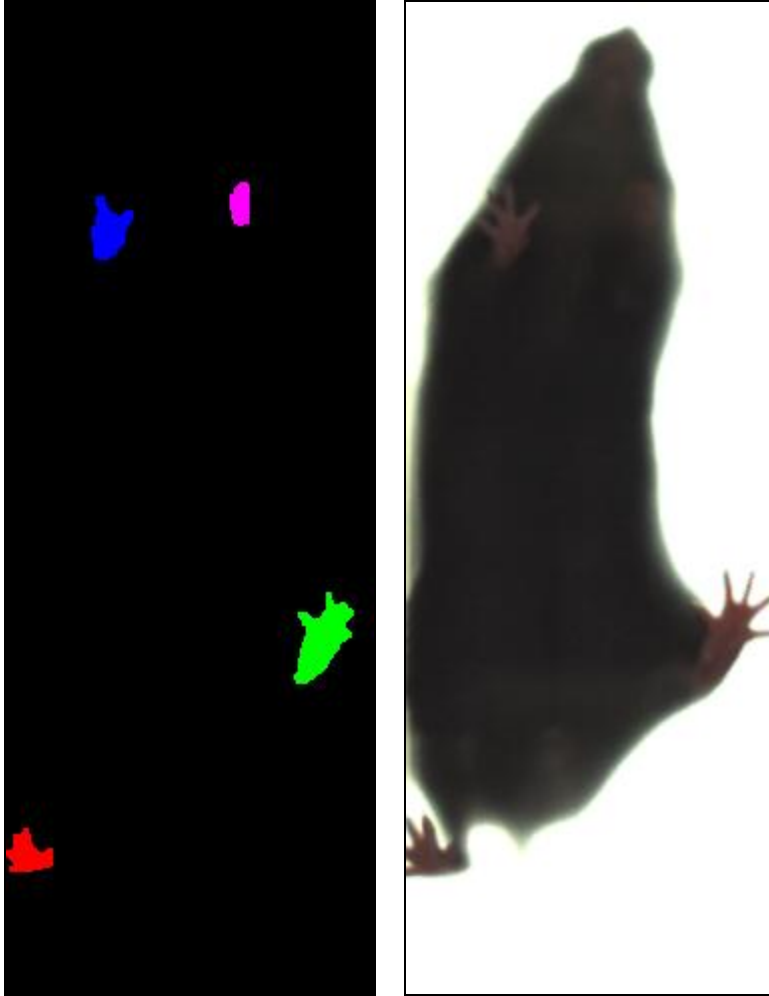


Promoter analysis and Luciferase mouse



- Advanced understanding on ATXN2 regulatory control ETS1
- Start codon
- No RNA translation
- LncRNA identification Luciferase mouse created using endogenous Atxn2 promoter in mouse
- Advanced our understanding on ATXN2 localization: Cerebellum, Olfactory bulb, Epididymis

DigiGait automated treadmill assay



- Study comparing performance of B6 and 129 mouse strains using the treadmill assay under conditions of EtOH induced ataxia.
- Evaluation of the instrument's utility
- Ability to detect ataxia in mice under extreme conditions of EtOH intoxication
 - Poor at detecting modest ataxia
- Mouse strain affects results

Manuscripts

- Hansen *et al.* (2012) Changes in Purkinje cell firing and gene expression precede behavioral pathology in a mouse model of SCA2. *Human Molecular Genetics* (Accepted—mostly)
- Scoles *et al.* (2012) ETS1 regulates the expression of *ATXN2*. *Human Molecular Genetics* (Accepted)
- Response to ethanol induced ataxia between C57Bl/6J and 129X1/SvJ mouse strains using a treadmill based assay *Pharmacology Biochemistry and Behavior* (Favorable initial reviews, resubmitted 9/2/2012)
- Complex spike behavior in *Atxn2*^{Q127} mouse PC (Otis?).

Manuscripts in Progress

- RNA translation (Lance Pflieger)
- LncRNA (Dan Scoles)
- BAC mouse (Warunee Danisthong)
- SCA2 x SCA6 mouse (Steve Hansen)

Work in Progress

- Laser Capture Microscopy: Lance working on
- RNA sequencing: B6 Atxn2^{Q127} and BAC mouse
 - Younger age points: 1day, 2 weeks.
- Endophenotype with candidate genes identified with RNA seq
- E-phys (Otis)
- Complete characterization of Atxn2^{Q127} crossed with Park KO, SCA6 KO and SCA6 KI