ETS1 Regulates *ATXN2* Expression

Daniel R. Scoles, Lance T. Pflieger, Steven T. Hansen, Khan K. Thai, Stefan-M. Pulst

Department of Neurology, University of Utah, Salt Lake City

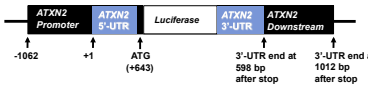
Introduction

SCA2 is an autosomal dominant cerebellar ataxia characterized by progressive degeneration of the cerebellum and brain stem. SCA2 is caused by polyglutamine expansion in the SCA2 protein ataxin-2, causing gain-of-function. We hypothesize that reduction of *ATXN2* expression will be therapeutic for SCA2, based on reversible phenotypes in polyQ models and lack of neuro-degeneration in SCA2 knockout mice.

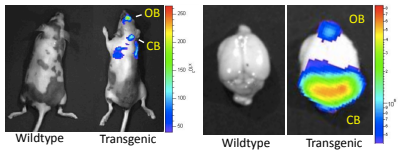
Objectives

To characterize the *ATXN2* promoter to gain understanding on how to control its expression.

ATXN2-luc reporter construct

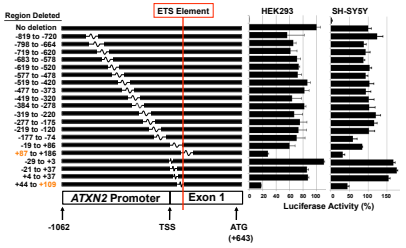


Similar to endogenous *ATXN2*, *ATXN2-luc* is expressed in cerebellum (CB) and olfactory bulb (OB)

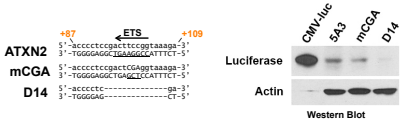


Images display photons/sec/cm^2/sr determined by *in vivo* imaging on a Xenogen IVIS 100.

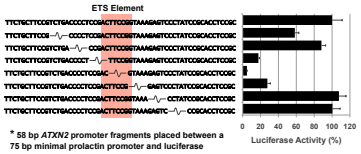
Deletions harboring an ETS element in the *ATXN2* promoter between +87 to +109 most potently inhibit *ATXN2-luc* expression



The *ATXN2* ETS site CCGGAAGT is on the minus strand and has 100% match with the consensus for ETS1 binding. A 3 bp substitution reduced expression and 14 bp deletion abolished expression of *ATXN2-luc*

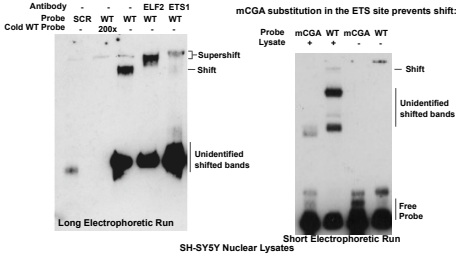


5 bp deletions in *ATXN2* reveal lowest expression with ETS core elimination

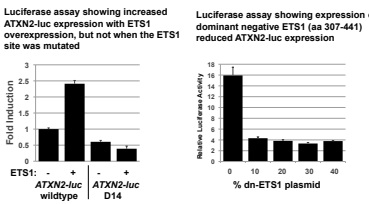


* 58 bp *ATXN2* promoter fragments placed between a 75 bp minimal prolactin promoter and luciferase

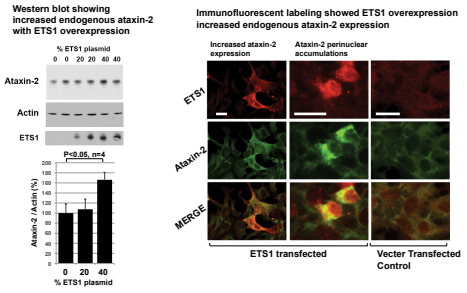
ETS1 & ELF2 bind a 20 bp *ATXN2* probe (WT in electromobility supershift assays)



ETS1 activates ATXN2-luc (HEK293)



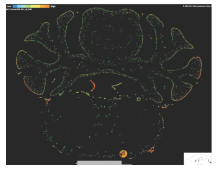
ETS1 activates endogenous ataxin-2 (HEK293)



Conclusions

•*ATXN2-luc* expression mimicked predictions for endogenous *ATXN2* (seen in Allen Brain Atlas), suggesting this *ATXN2* fragment possesses promoter elements required for tissue-specific expression.

- **_Ets1_ in the Allen Brain Atlas shows high expression in Purkinje cells, suggesting ETS1 may support *ATXN2* expression in this cell type.**



***Ets1 in situ* hybridization.** Reproduced with permission of the Allen Mouse Brain Atlas [Internet]. Seattle (WA): Allen Institute for Brain Science. ©2009. Available from: http://mouse.brain-map.org.

•The ETS element in the *ATXN2* promoter supports the majority of *ATXN2* expression.

•ETS1 may represent a therapeutic target for treating SCA2.

•Since the majority of *ATXN2* expression is supported by this small promoter region, *ATXN2* expression could be targeted by decoy oligonucleotides containing the *ATXN2* ETS1 site and short flanking sequences.

Acknowledgements

We thank Barbara Graves for advice on ETS transcription factors and Steven Lessnick for assistance with IVIS imaging. This work was supported by grants 1RC4NS073009 & 5R01NS033123 from NINDS.