Legends for supplemental figure and table

Supplemental figure 1: Choice of ataxin-3-expressing flies

A-C) Western blots of new transgenic *Drosophila* lines expressing UAS-ataxin-3 of the noted versions, driven by sqh-Gal4, which expresses UAS constructs in all fly tissues and throughout development and in adults (Franke et al., 2006; Franke et al., 2005; Todi et al., 2005). Ten one day-old flies were harvested in SDS lysis buffer, sonicated, boiled and centrifuged, then loaded on SDS-PAGE gels. Arrows indicate the lines we chose based on similarity of their protein levels to wild-type ataxin-3.

D, E) qRT-PCR from the various lines, showing higher expression of ataxin-3 transgenes that encode ataxin-3 with the Rad23 site mutated compared with wild-type ataxin-3, but whose protein levels are similar to those of wild-type ataxin-3 (panels A-C).

Supplemental table 1: *Drosophila* lines used

Comprehensive list of the fly lines that we procured from Bloomington *Drosophila* Stock Center and Vienna RNAi *Drosophila* Center for our studies. Information includes the genotype, gene target for RNAi lines, catalog number, and fly orthologue for genes without a name designation, based on our own BLASTp searches using the NCBI BLAST tool.

Supplemental References

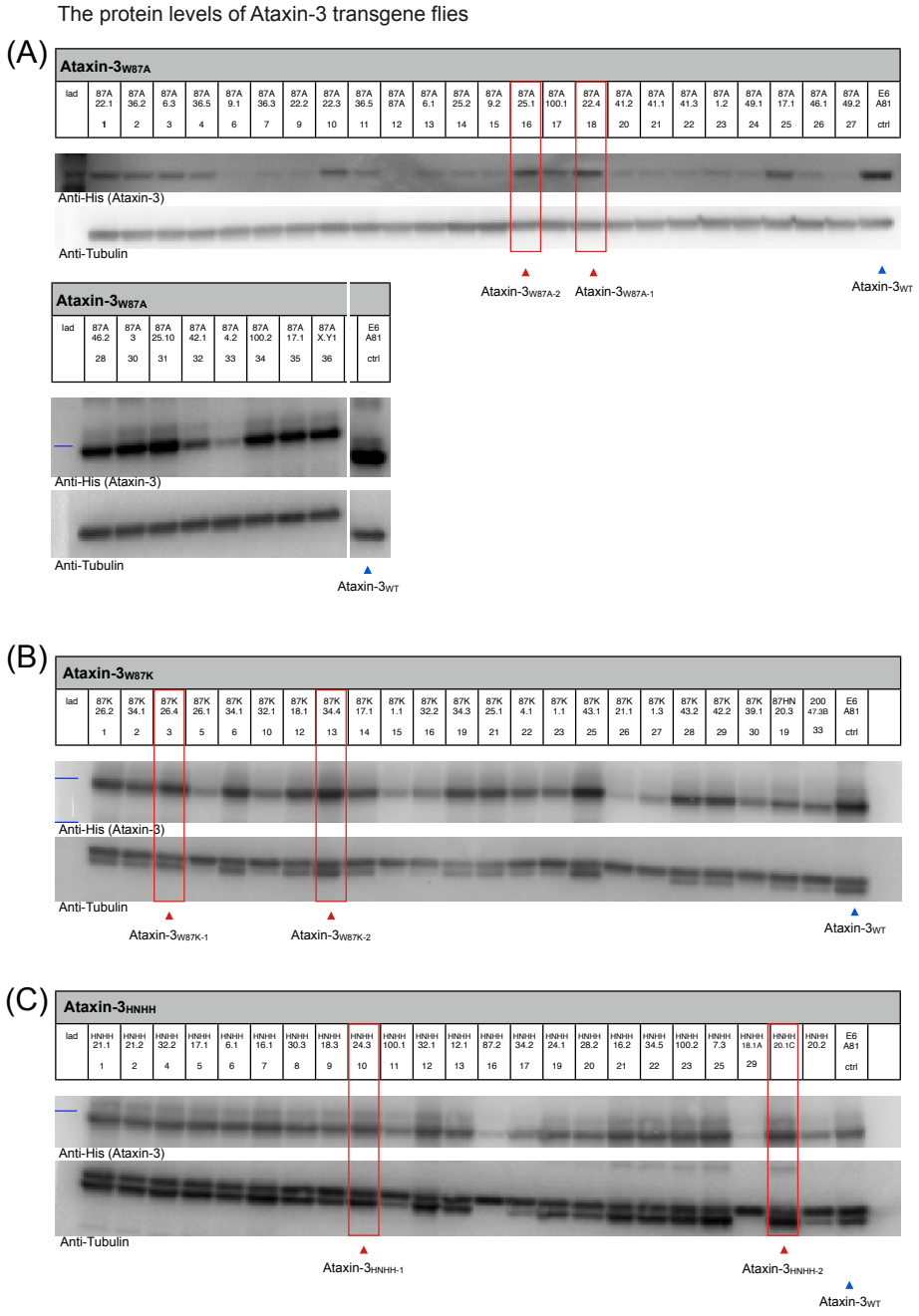
Franke, J. D., et al., 2006. Native nonmuscle myosin II stability and light chain binding in Drosophila melanogaster. Cell Motil Cytoskeleton. 63**,** 604-22.

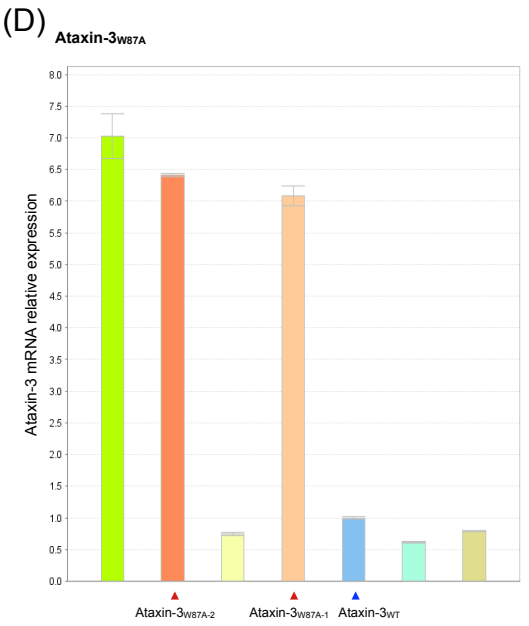
Franke, J. D., et al., 2005. Nonmuscle myosin II generates forces that transmit tension and drive contraction in multiple tissues during dorsal closure. Curr Biol. 15**,** 2208-21.

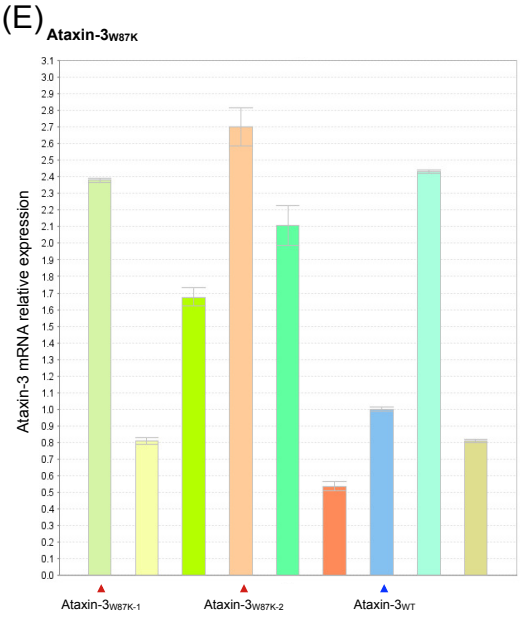
Todi, S. V., et al., 2005. Myosin VIIA defects, which underlie the Usher 1B syndrome in humans, lead to deafness in Drosophila. Curr Biol. 15**,** 862-8.

Supplemental figure 1

The protein levels of Ataxin-3 transgene flies







Supplemental Table 1

Genotype	Source	FlyBase ID	Stock Number	Associated alleles	Description	Shown in the figures
w[1118]; +; +	Bloomington	FBal0018186	3605		Isogenic host strain for the Ataxin-3 line	Yes
w[*]; +; P{w[+mC]=longGMR-GAL4}3	Bloomington	FBst0008121	8121		gmr-Gal4	Yes
w[*]; P{w[+mC]=longGMR-GAL4}2; +	Bloomington	FBst0009146	9146		gmr-Gal4	Yes
w[*]; P{w[+mC]=UAS-Hsap\MJD.tr-Q78}c211.2; +	Bloomington	FBst0008150	8150	Ataxin-3	UAS-PolyQ^{78}	Yes
w[*]; P{w[+mC]=UAS-Hsap\MJD.tr-Q78}c37.3/TM3, Sb[1]	Bloomington	FBst0008141	8141	Ataxin-3	UAS-PolyQ^{78}	Yes
w[*]; P{w[+mC]=UAS-ataxin-3-WT}E6.2; +	Todi Lab			Ataxin-3	UAS-Ataxin-3-WT	Yes
w[*]P{w[+mC]=UAS-ataxin-3-WT}E2.1;+ ; +	Todi Lab			Ataxin-3	UAS-Ataxin-3-WT	Yes
w[*]; P{w[+mC]=UAS-ataxin-3-C14A}E12.2; +	Todi Lab			Ataxin-3	UAS-Ataxin-3-C14A	Yes
w[*]; P{w[+mC]=UAS-ataxin-3-HNHH}E1101.2; +	Todi Lab			Ataxin-3	UAS-ataxin-3-HNHH$_{-1}$	Yes
w[*]; P{w[+mC]=UAS-ataxin-3-HNHH}E1102.2; +	Todi Lab			Ataxin-3	UAS-ataxin-3-HNHH$_{-2}$	Yes
w[*]; P{w[+mC]=UAS-ataxin-3-W87K}E1007.2; +	Todi Lab			Ataxin-3	UAS-ataxin-3-W87K$_{-1}$	Yes
w[*]; +; P{w[+mC]=UAS-ataxin-3-W87K}E1004.3	Todi Lab			Ataxin-3	UAS-ataxin-3-W87K$_{-2}$	Yes
w[*]; +; P{w[+mC]=UAS-ataxin-3-W87A}E908.3	Todi Lab			Ataxin-3	UAS-ataxin-3-W87A$_{-1}$	Yes
w[*]; +; P{w[+mC]=UAS-ataxin-3-W87A}E911.3	Todi Lab			Ataxin-3	UAS-ataxin-3-W87A$_{-2}$	Yes
y,w[1118]; +; +	VDRC		VDRC60000		Isogenic host strain for VDRC library	Yes
y[1] v[1]; P{y[+t7.7]=CaryP}attP2	Bloomington	FBst0036303	36303		Isogenic host strain for attP2	Yes
y[1] v[1]; P{y[+t7.7]=CaryP}attP40	Bloomington	FBst0036304	36304		Isogenic host strain for attP40	Yes
P{KK107826}VIE-260B	VDRC	FBst0476212	v104354	CG1836	Rad23 RNAi	Yes
w[1118]; P{GD4245}v30498	VDRC	FBst0458518	v30498	CG1836	Rad23 RNAi	No
y[1] sc[*] v[1]; P{y[+t7.7] v[+t1.8]=TRiP.HM05268}attP2/TM3, Sb[1]	Bloomington	FBst0031875	31875	CG7425	UbcH5 (*effete*) RNAi	Yes
P{KK109208}VIE-260B	VDRC	FBst0476336	v104478	CG7220	CG7220(Ube2W) RNAi	Yes
w[1118]; P{GD10597}v34199	VDRC	FBst0460510	v34199	CG7220	CG7220(Ube2W) RNAi	No
y[1] sc[*] v[1]; P{y[+t7.7] v[+t1.8]=TRiP.HMS00986}attP2	Bloomington	FBst0034017	34017	CG5203	Chip RNAi	Yes
y[1] sc[*] v[1]; P{y[+t7.7] v[+t1.8]=TRiP.HMS00889}attP2	Bloomington	FBst0033938	33938	CG5203	Chip RNAi	No
y[1] v[1]; P{y[+t7.7] v[+t1.8]=TRiP.JF02691}attP2	Bloomington	FBst0027540	27540	CG9934	Ube4b RNAi	Yes
y[1] sc[*] v[1]; P{y[+t7.7] v[+t1.8]=TRiP.HMS01651}attP40	Bloomington	FBst0037509	37509	CG10523	Parkin RNAi	Yes
y[1] v[1]; P{y[+t7.7] v[+t1.8]=TRiP.JF01200}attP2	Bloomington	FBst0031259	31259	CG10523	Parkin RNAi	No
y[1] sc[*] v[1]; P{y[+t7.7] v[+t1.8]=TRiP.HMS01800}attP2/TM3, Sb[1]	Bloomington	FBst0038333	38333	CG10523	Parkin RNAi	No
w[1118]; P{GD3145}v6870	VDRC	FBst0470477	v6870	CG1937	Hrd1 RNAi	Yes
P{KK102436}VIE-260B	VDRC	FBst0478883	v107060	CG1937	Hrd1 RNAi	No
w[1118]; P{GD8888}v19272	VDRC	FBst0453458	v19272	CG18174	PSMD14 RNAi	Yes
w[1118] P{GD8888}v19273	VDRC	FBst0453459	v19273	CG18174	PSMD14 RNAi	No
P{KK101319}VIE-260B	VDRC	FBst0480383	v108573	CG3416	PSMD7 RNAi	Yes
w[1118] P{GD10960}v26183	VDRC	FBst0456281	v26183	CG3416	PSMD7 RNAi	No
Prosbeta6[1] pb[1] p[p]/TM3, Sb[1] Ser[1]	Bloomington	FBst0006182	6182	CG4097	Proteasome β6 subunit T18I. (DTS5)	Yes
w[1118]; P{w[+mC]=UAS-Prosbeta6[1].B}2B; P{UAS-Prosbeta2[1]}1B	Bloomington	FBst0006787	6787	CG4097, CG3329	Prosbeta2&6; dominant negative proteasome subunits.	Yes
w[1118]; P{w[+mC]=UAS-Prosbeta6[1].B}2B	Bloomington	FBst0006786	6786	CG4097	Prosbeta6; dominant negative proteasome subunits.	No
P{KK105338}VIE-260B	VDRC	FBst0480005	v108193	CG10369	P62 RNAi	Yes
y[1] sc[*] v[1]; P{y[+t7.7] v[+t1.8]=TRiP.HMS00938}attP2	Bloomington	FBst0033978	33978	CG10369	P62 RNAi	No
y[1] sc[*] v[1]; P{y[+t7.7] v[+t1.8]=TRiP.HMS00551}attP2	Bloomington	FBst0036111	36111	CG10369	P62 RNAi	No
w[1118]; P{GD11671}v45558	VDRC	FBst0466207	v45558	CG5489	Atg7 RNAi	Yes
w[1118]; P{GD11671}v45560/TM3	VDRC	FBst0466209	v45560	CG5489	Atg7 RNAi	No
y[1] v[1]; P{y[+t7.7] v[+t1.8]=TRiP.JF02787}attP2	Bloomington	FBst0027707	27707	CG5489	Atg7 RNAi	No

y[1] sc[*] v[1]; P{y[+t7.7] v[+t1.8]=TRiP.HMS01358}attP2/TM3, Sb[1]	Bloomington	FBst0034369	34369	CG5489	Atg7 RNAi	No
y[1] sc[*] v[1]; P{y[+t7.7] v[+t1.8]=TRiP.HMS01328}attP2	Bloomington	FBst0034340	34340	CG32672	Atg8a RNAi	Yes
y[1] v[1]; P{y[+t7.7] v[+t1.8]=TRiP.JF02895}attP2 e[*]/TM3, Sb[1]	Bloomington	FBst0028989	28989	CG32672	Atg8a RNAi	No
y[1] v[1]; P{y[+t7.7] v[+t1.8]=TRiP.JF02704}attP2	Bloomington	FBst0027552	27552	CG18402	Atg12 RNAi	Yes
y[1] sc[*] v[1]; P{y[+t7.7] v[+t1.8]=TRiP.HMS01153}attP2	Bloomington	FBst0034675	34675	CG18402	Atg12 RNAi	No
w[1118]; P{GD15230}v29791/CyO	VDRC	FBst0458151	v29791	CG18402	Atg12 RNAi	No
w[*]; P{w[+mC]=UAS-DnaJ-1.K}3	Bloomington	FBst0030553	30553	CG10578	UAS-DnaJ-1	Yes
y[1] sc[*] v[1]; P{y[+t7.7] v[+t1.8]=TRiP.HMS00688}attP2/TM3, Sb[1]	Bloomington	FBst0032899	32899	CG10578	DnaJ-1 RNAi	Yes
y[1] sc[*] v[1]; P{y[+t7.7] v[+t1.8]=TRiP.HMS00778}attP2	Bloomington	FBst0032978	32978	CG10578	DnaJ-1 RNAi	No
P{KK108978}VIE-260B	VDRC	FBst0473405	v101532	CG5001	CG5001 RNAi	Yes
y[1] sc[*] v[1]; P{y[+t7.7] v[+t1.8]=TRiP.HMS00386}attP2	Bloomington	FBst0032392	32392	CG5001	CG5001 RNAi	No
w[1118]; P{GD15063}v39126	VDRC	FBst0462845	v39126	CG8448	mrj RNAi	Yes
w[1118]; P{GD2776}v44395	VDRC	FBst0465559	v44395	CG7130	CG7130 RNAi	Yes
P{KK111794}VIE-260B	Bloomington	FBst0482096	v110526	CG7130	CG7130 RNAi	No
y[1] v[1]; P{y[+t7.7] v[+t1.8]=TRiP.JF02415}attP2/TM3, Sb[1]	Bloomington	FBst0027070	27070	CG5748	Hsf RNAi	Yes
y[1] v[1]; P{y[+t7.7] v[+t1.8]=TRiP.GL00698}attP2	Bloomington	FBst0041581	41581	CG5748	Hsf RNAi	No
w[1118]; P{GD16368}v48692	VDRC	FBst0468066	v48692	CG5748	Hsf RNAi	No
P{KK100723}VIE-260B	VDRC	FBst0480645	v108851	CG5748	Hsf RNAi	No
net[1] cn[1] Hsf[1]/CyO	Bloomington	FBst0005491	5491	CG5748	Hsf mutant allele	No
y[1] v[1]; P{y[+t7.7] v[+t1.8]=TRiP.JF03136}attP2	Bloomington	FBst0028709	28709	CG4264	Hsc70-4 RNAi	Yes
y[1] sc[*] v[1]; P{y[+t7.7] v[+t1.8]=TRiP.HMS00152}attP2/TM3, Sb[1]	Bloomington	FBst0034836	34836	CG4264	Hsc70-4 RNAi	No
y[1] sc[*] v[1]; P{y[+t7.7] v[+t1.8]=TRiP.GLV21049}attP2	Bloomington	FBst0035684	35684	CG4264	Hsc70-4 RNAi	No
w[126]; P{w[+mC]=UAS-Hsc70-4.D206S}E	Bloomington	FBst0005844	5844	CG4264	Hsc70-4 mutant allele	No
w[126]; P{w[+mC]=UAS-Hsc70-4.K71S}G	Bloomington	FBst0005845	5845	CG4264	Hsc70-4 mutant allele	No
y[1] sc[*] v[1]; P{y[+t7.7] v[+t1.8]=TRiP.HMS00397}attP2	Bloomington	FBst0032402	32402	CG4147	Hsc70-3 RNAi	Yes
w[1118]; P{GD6484}v14882	VDRC	FBst0451655	v14882	CG4147	Hsc70-3 RNAi	No
P{KK109452}VIE-260B	VDRC	FBst0473639	v101766	CG4147	Hsc70-3 RNAi	No
P{KK104957}VIE-260B	VDRC	FBst0472742	v100869	CG10973	CG10973 RNAi	Yes
w[1118]; P{GD7153}v41463	VDRC	FBst0464110	v41463	CG10973	CG10973 RNAi	No
y[1] sc[*] v[1]; P{y[+t7.7] v[+t1.8]=TRiP.HMS00807}attP2	Bloomington	FBst0033007	33007	CG4466	Hsp27 RNAi	Yes
y[1] sc[*] v[1]; P{y[+t7.7] v[+t1.8]=TRiP.HMS00867}attP2	Bloomington	FBst0033922	33922	CG4466	Hsp27 RNAi	No
P{KK107896}VIE-260B	VDRC	FBst0472570	v100697	CG12101	Hsp60 RNAi	Yes
w[1118]; P{GD7481}v18738	VDRC	FBst0453228	v18738	CG12101	Hsp60 RNAi	No
y[1] sc[*] v[1]; P{y[+t7.7] v[+t1.8]=TRiP.HMS01209}attP2/TM3, Sb[1]	Bloomington	FBst0034729	34729	CG12101	Hsp60 RNAi	No
y[1] sc[*] v[1]; P{y[+t7.7] v[+t1.8]=TRiP.HMS00796}attP2	Bloomington	FBst0032996	32996	CG1242	Hsp83 RNAi	Yes
y[1] sc[*] v[1]; P{y[+t7.7] v[+t1.8]=TRiP.HMS00899}attP2	Bloomington	FBst0033947	33947	CG1242	Hsp83 RNAi	No