

Fig. S1. The anatomy of cerebellar circuits is grossly intact in mdx mice. A. The total number of Purkinje cells (PC) in the whole mdx cerebellum (Cb) does not differ statistically from the number of Purkinje cells (PC) in the whole control cerebellum (counted in all lobules/per sagittal section; Control = 569.2 ± 32.7 , $mdx = 578.7 \pm 6.7$, two-tailed Student's t-tests, N = 3 animals per genotype, n = 3-8 sections per animal, t(4) = -0.286, p = 0.79). B-D. The total number of Purkinje cells (PC) in the *mdx* cerebellum does not differ from that of controls when analyzed by zone (Control = 256.0 ± 12.9 (anterior), $169. \pm 7.4$ (central), 143.8 ± 14.3 (posterior/nodular), mdx= 260.6 ± 3.6 (anterior), $181.\pm 5.1$ (central), 136.7 ± 2.3 (posterior/nodular), two-tailed Student's ttests, N = 3 animals per genotype, n = 3-8 sections per animal; t(4) = -0.340, p = 0.75, t(4) = -0.3401.345, p = 0.25, t(4) = 0.489, p = 0.65, respectively). E-F. The fastigial (FN) and interposed (IN) nuclear densities (no = number) are comparable between the genotypes (Control = 17.36 ± 1.61 (FN), 36.11 ± 3.12 (IN), $mdx = 21.26 \pm 1.20$ (FN), 33.19 ± 1.14 (IN), two-tailed Student's t-tests, N = 3 animals per genotype, n = 3 sections per animal; t(4) = -1.9646, p = 0.12, t(4) = 0.880, p = 0.43, respectively). G-J. The areas of the fastigial (FN) and interposed (IN) cerebellar nuclei are comparable between control and *mdx* mice (Control = $1,242.5 \pm 154.9 \,\mu\text{m}^2$ (FN), $4,051.9 \pm 385.4$ μ m² (IN), $mdx = 1,483.5 \pm 287.7 \mu$ m² (FN), $3,015.8 \pm 567.9 \mu$ m² (IN), two-tailed Student's t-tests, N = 3 animals per genotype, n = 3 sections per animal, t(4) = -0.737, p = 0.50, t(4) = 1.510, p = 0.21, respectively). The distances of the fastigial and interposed nuclei from bregma are +0.72 mm (FN) and +1.56 mm (IN), respectively. Calbindin staining (green) labels Purkinje cell soma, axons and terminals whereas Nissl (blue) is a general stain for cell nuclei. Scale bar, 50 µm.



Fig. S2. Purkinje cells in control and *mdx* mice express multiple isoforms of dystrophin. A. Schematic detailing the different isoforms of dystrophin. Gray asterisks indicate the isoforms that are expressed in the mammalian brain. The dystrophin antibody used in Fig. 1 targets the rod domain (red) while the MANDRA1 antibody used in this supplemental figure targets the c-terminus (teal), which is present in all of the isoforms. B. The MANDRA1 antibody detects dystrophin isoforms that are located near the plasma membrane since the c-terminus of the dystrophin protein interacts with glycoproteins embedded in the plasma membrane. C. The hippocampus expresses MANDRA1 in the control brain, demonstrating the expected pattern of expression. D-E. MANDRA1 expression in control (D) and *mdx* (E) Purkinje cells is identical. Representative examples of Purkinje cells are outlined. F. Examples of control Purkinje cells (PC) that express MANDRA1 (red arrows). G. Examples of *mdx* Purkinje cells (PC) that express MANDRA1 (red arrows). Scale bar (C), 200 μ m. Scale bar (D-E), 50 μ m. Scale bar (F-G), 10 μ m. Molecular layer (ml). Purkinje cell layer (pcl). N = 3 mice per genotype.

1.2

1

CS CV2

0.8

0.6

0



2

1.5

0

0.5

1

CS Hz

Fig. S3. The time between surgery and recording does not affect the Purkinje cell properties.

The session day following surgery in which analyzed awake Purkinje cells were recorded did not affect simple spike (**A**) or complex spike (**B**) summary statistics when comparing neurons recorded 10 days or fewer following surgery (n = 25) versus those recorded more than 10 days after surgery (n = 23). For days post-surgery as a predictor variable, recorded cells were divided into two groups, high and low, both relative to the median value. The median value was 10, but with two cells recorded exactly 10 days after surgery. Please refer to **Table S1** for a list of the statistical tests used in this figure.





Fig. S4. The depth of the recordings does not affect overall Purkinje cell properties. The depth of recorded awake Purkinje cells did not affect simple spike (A) or complex spike (B) summary statistics when comparing neurons recorded dorsal to 1650 μ m (n = 24) and those recorded ventral to 1650 μ m (n = 24). The depth of recorded neurons was calculated based on where we found the recording features at optimal signal to noise ratio with respect to the surface of the cerebellum at the site of the craniotomy. The recorded cells were recorded into two groups, high and low, relative to the median value. Therefore, as a population the dorsoventral depth indicates neurons that fall in the upper half versus the lower half of the analyzed distance. Please refer to **Table S1** for a list of the statistical tests used in this figure.



Fig. S5. Potential predictor variables apart from genotype show no effect on spiking characteristics in cerebellar nuclear neurons. A. The length of time between craniotomy surgery and recording date did not significantly affect rate, CV, or CV2 (one-way MANOVA p = 0.79). B. Anteroposterior recording position showed no difference in spiking statistics between the anterior and posterior half of recorded cells (p = 0.25). C. Mediolateral distance was not a significant factor influencing cerebellar nuclear firing (p = 0.13). D. Dorsoventral recording depth did not affect spiking statistics (p = 0.18). E. Putative recorded nuclei showed no significant effect on spiking statistics (p = 0.18).



Fig. S6. Dystrophin is weakly expressed in a subset of cells in the cerebellar nuclei of control and *mdx* mice. A-B. A small number of cells in the fastigial (A) and interposed (B) cerebellar nuclei are weakly immunoreactive for the Dp427 isoform in control mice. C-D. Anti-dystrophin staining in the fastigial (C) and interposed (D) cerebellar nuclei of *mdx* mice reveals an identical staining pattern to that of control mice. The expression of Dp427 is weaker in specific regions of the interposed nuclei of both genotypes. In many cells within both cerebellar nuclei, the expression of Dp427 is barely beyond the level of background staining. Scale bar (A-D), 200 μ m. N = 3 mice per genotype.

Comparison	Statistical	Ν	Statistic	Degrees	Likelihood	BH-
	Test			of		corrected
				freedom		significance
Overall cerebellar area by	Two-tailed	6	t = 0.280	4	p = 0.7936	0
genotype	t-test				1	
Molecular layer thickness	Two-tailed	6	t = -0.623	4	p = 0.5668	0
by genotype	t-test					
Purkinje cell number by	Two-tailed				0.7000	0
genotype, whole	t-test	6	t = -0.286	4	p = 0.7888	0
cerebellum						
Purkinje cell number by	Two-tailed	6	t = -0.3401	4	p = 0.7509	0
genotype, anterior zone	t-test				1	
Purkinje cell number by	Two-tailed	6	t = -1.3452	4	p = 0.2498	0
genotype, central zone	t-test					
Purkinje cell number by	Two-tailed	6			0.6500	0
genotype, posterior-	t-test	6	t = 0.4894	4	p = 0.6502	0
nodular zones						
Fastigial nucleus area by	Two-tailed	6	t = -0.7372	4	p = 0.5019	0
genotype	t-test					
Fastigial nucleus density	Two-tailed	6	t = -1.9457	4	p = 0.1236	0
by genotype	t-test					
Interposed nucleus area by	Two-tailed	6	t = 1.5096	4	p = 0.2056	0
genotype	t-test				-	
Interposed nucleus density	Two-tailed	6	t = 0.8796	4	p = 0.4287	0
by genotype	t-test				-	
Anesthetized simple spike	One-way	04	1 - 0.0028	1.02	n = 0.02(1)	1
response variables by	MANOVA	94	$\lambda = 0.9028$	1,92	p – 0.0261	1
genotype						
Anesthetized simple spike	Wilcoxon	48/46	z = 1.99	93	p = 0.0471	0
firing rate by genotype	rank sum					
Anesthetized simple spike	Wilcoxon	48/46	z = 1.76	93	p = 0.0787	0
CV by genotype	rank sum					
Anesthetized simple spike	Wilcoxon	48/46	z = -1.48	93	p = 0.1392	0
CV2 by genotype	rank sum					
Anesthetized complex	One-way	94	$\lambda = 0.0020$	1.02	n = 0.8859	0
spike response variables	MANOVA	74	k = 0.7727	1,72	p = 0.0057	0
by genotype						
Awake simple spike	One-way	18	3 - 0.8266	1.46	n = 0.0371	1
response variables by	MANOVA	40	$\lambda = 0.8200$	1,40	p = 0.0371	1
genotype						
Awake simple spike firing	Wilcoxon	24/24	z = 2.53	47	p = 0.0115	1
rate by genotype	rank sum					
Awake simple spike CV	Wilcoxon	24/24	z = -0.03	47	p = 0.9753	0
by genotype	rank sum					

by genotyperank sumrAwake complex spike response variables byOne-way MANOVA 48 $\lambda = 0.7772$ $1,46$ $p = 0.0106$ 1	
Awake complex spike response variables byOne-way MANOVA48 $\lambda = 0.7772$ 1,46 $p = 0.0106$ 1	
response variables by MANOVA 48 $\lambda = 0.7772$ 1,46 $p = 0.0106$ 1	
genotype	
Awake complex spikeWilcoxon $24/24$ $z = -0.55$ 47 $p = 0.5848$ 0	
firing rate by genotype rank sum	
Awake complex spike CVWilcoxon $24/24$ $z = 3.17$ 47 $p = 0.0016$ 1	
by genotype rank sum	
Awake complex spikeWilcoxon $24/24$ $z = 0.75$ 47 $p = 0.4517$ 0	
CV2 by genotype rank sum	
Awake CN response One-way 51 $\lambda = 0.7494$ $1,49$ $p = 0.0033$ 1	
variables by genotype MANOVA	
Awake CN firing rate byWilcoxon $28/23$ $z = -1.47$ 50 $p = 0.1424$ 0	
genotype rank sum	
Awake CN CV by Wilcoxon $28/23$ $z = -0.71$ 50 $p = 0.4778$ 0	
genotype rank sum	
Awake CN CV2 by Wilcoxon $28/23$ $z = -2.36$ 50 $p = 0.0184$ 1	
genotype rank sum	
Awake simple spikeOne-way48 $\lambda = 0.9613$ 1.46 $p = 0.6250$ 0	
response variables by days MANOVA	
Awake simple spike One-way to be a solta of the solution	
response variables by MANOVA 48 $\lambda = 0.9943$ 1,46 $p = 0.9954$ 0	
dorso-ventral position	
Awake complex spikeOne-way48 $\lambda = 0.8533$ 1,46 $p = 0.0701$ 0	
response variables by days MANOVA	
Awake complex spike One-way to a costa to the costa	
response variables by MANOVA 48 $\lambda = 0.9943$ 1,46 $p = 0.9682$ 0	
dorso-ventral position	
Recording depth byWilcoxon $24/24$ $z = 0.58$ 47 $p = 0.56$ 0	
genotype rank sum	
Awake CN response One-way 51 A 0.0702 1.10 0.702	
variables by days post- MANOVA 51 $\lambda = 0.9785$ $1,49$ $p = 0.7933$ 0	
surgery	
Awake CN response One-way 51 A 0.0154 1.40 0.0515	
variables by anterior- MANOVA 51 $\lambda = 0.9174$ $1,49$ $p = 0.2515$ 0	
posterior position	
Awake CN response One-way 51 $\lambda = 0.8891$ 1.49 $p = 0.1337$ 0	
variables by medio-lateral MANOVA	
Awake CN responseOne-way 51 $\lambda = 0.9022$ 1.49 $p = 0.1800$ 0	
variables by dorso-ventral MANOVA	
Awake CN response One-way 51 A control of the	
variables by putative MANOVA 51 $\lambda = 0.9019$ 1,49 $p = 0.1791$ 0	
lobule	

Table S1. Post-hoc statistical analyses employed throughout the study. A tabulated version of the data showing the values that were reported for the post-hoc statistical analyses.