

Supplemental Information

**AAV9 Gene Therapy Increases Lifespan and Treats
Pathological and Behavioral Abnormalities
in a Mouse Model of CLN8-Batten Disease**

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Supplemental Table 1. Death and euthanasia descriptions.

Treatment Group	Sex	Age at Death	Morbidity Notes
WT	F	16	Found dead - likely intestinal tumor leading to rupture of blood vessel and internal hemorrhaging
WT	F	17	Euthanized - significantly underweight, labored breathing, lateral recumbancy, hunched
WT	F	18	Euthanized - labored breathing; hunched
WT	F	21	Unknown
WT	F	21	Unknown
WT	F	22	Euthanized - labored breathing; moribund
WT	F	22	Euthanized – eye ulcer, unthrifty
WT	F	23	Euthanized – prolapsed rectum
WT	F	23	Euthanized – severe lesion on ear, distended abdomen
WT	F	23	Euthanized – hunched, mass
WT	F	24	Euthanized – actively bleeding rectum
WT	F	24	n/a - retired from study
WT	F	24	n/a - retired from study
WT	F	24	n/a - retired from study
WT	F	24	n/a - retired from study
WT	M	22	Euthanized – ruptured abscess
WT	M	22	Euthanized – mass on tail, abscess in abdomen that drained, unthrifty
WT	M	24	Euthanized – labored breathing, abdominal mass
WT	M	24	Euthanized – lesion due to ruptured abscess
WT	M	24	Euthanized – distended abdomen, mass
WT	M	24	n/a - retired from study
WT	M	24	n/a - retired from study
WT	M	24	n/a - retired from study
WT	M	24	n/a - retired from study
WT	M	24	n/a - retired from study
WT	M	24	n/a - retired from study
WT	M	24	n/a - retired from study
WT	M	24	n/a - retired from study
WT	M	24	n/a - retired from study
WT	M	24	n/a - retired from study
WT	M	24	n/a - retired from study
<i>Cln8^{mynd}</i> + AAV9	F	17	Euthanized - prolapsed/bleeding rectum
<i>Cln8^{mynd}</i> + AAV9	F	18	Euthanized - Significant growth appearing on lateral abdomen, emaciation, impaired movement due to growth
<i>Cln8^{mynd}</i> + AAV9	F	19	Unknown

<i>Cln8^{mnd}</i> + AAV9	F	21	Euthanized – prolapsed rectum
<i>Cln8^{mnd}</i> + AAV9	F	22	Euthanized – prolapsed rectum
<i>Cln8^{mnd}</i> + AAV9	F	22	Euthanized – prolapsed rectum, masses in abdomen
<i>Cln8^{mnd}</i> + AAV9	F	24	Euthanized – hunched, labored breathing, bleeding rectum
<i>Cln8^{mnd}</i> + AAV9	F	24	n/a - retired from study
<i>Cln8^{mnd}</i> + AAV9	F	24	n/a - retired from study
<i>Cln8^{mnd}</i> + AAV9	F	24	n/a - retired from study
<i>Cln8^{mnd}</i> + AAV9	F	24	n/a - retired from study
<i>Cln8^{mnd}</i> + AAV9	F	24	n/a - retired from study
<i>Cln8^{mnd}</i> + AAV9	F	24	n/a - retired from study
<i>Cln8^{mnd}</i> + AAV9	F	24	n/a - retired from study
<i>Cln8^{mnd}</i> + AAV9	F	24	n/a - retired from study
<i>Cln8^{mnd}</i> + AAV9	M	21	Unknown
<i>Cln8^{mnd}</i> + AAV9	M	22	Unknown
<i>Cln8^{mnd}</i> + AAV9	M	23	Euthanized – abscess in abdomen
<i>Cln8^{mnd}</i> + AAV9	M	23	Euthanized - abdominal mass; mobility issues
<i>Cln8^{mnd}</i> + AAV9	M	23	Euthanized - abdominal mass
<i>Cln8^{mnd}</i> + AAV9	M	24	Euthanized - abdominal mass, labored breathing
<i>Cln8^{mnd}</i> + AAV9	M	24	Euthanized - abdominal mass
<i>Cln8^{mnd}</i> + AAV9	M	24	n/a - retired from study
<i>Cln8^{mnd}</i> + AAV9	M	24	n/a - retired from study
<i>Cln8^{mnd}</i> + AAV9	M	24	n/a - retired from study
<i>Cln8^{mnd}</i> + AAV9	M	24	n/a - retired from study
<i>Cln8^{mnd}</i> + AAV9	M	24	n/a - retired from study
<i>Cln8^{mnd}</i> + AAV9	M	24	n/a - retired from study
<i>Cln8^{mnd}</i> + AAV9	M	24	n/a - retired from study
<i>Cln8^{mnd}</i> + AAV9	M	24	n/a - retired from study
<i>Cln8^{mnd}</i>	F	7	Euthanized - severe ulcerative dermatitis
<i>Cln8^{mnd}</i>	F	9	Euthanized - seizures, severely impaired ambulation
<i>Cln8^{mnd}</i>	F	9	Euthanized - seizures, severely impaired ambulation
<i>Cln8^{mnd}</i>	F	9	Found dead
<i>Cln8^{mnd}</i>	F	9	Euthanized - extreme weight loss
<i>Cln8^{mnd}</i>	F	9	Euthanized - humane end point
<i>Cln8^{mnd}</i>	F	9	Found dead

<i>Cln8^{mnd}</i>	F	10	Found dead
<i>Cln8^{mnd}</i>	F	10	Euthanized - did not recover from severe seizures
<i>Cln8^{mnd}</i>	F	10	Found dead
<i>Cln8^{mnd}</i>	F	10	Euthanized - ataxia, inability to right from back, weight loss, little control of back legs
<i>Cln8^{mnd}</i>	F	10	Found dead
<i>Cln8^{mnd}</i>	F	10	Euthanized - humane end point
<i>Cln8^{mnd}</i>	F	11	Euthanized - ataxia, significant weight loss, extreme difficulty in movement
<i>Cln8^{mnd}</i>	M	9	Found dead
<i>Cln8^{mnd}</i>	M	9	Found dead
<i>Cln8^{mnd}</i>	M	9	Euthanized - seizures, severely impaired ambulation
<i>Cln8^{mnd}</i>	M	9	Found dead
<i>Cln8^{mnd}</i>	M	10	Euthanized - humane end point
<i>Cln8^{mnd}</i>	M	10	Found dead
<i>Cln8^{mnd}</i>	M	10	Found dead
<i>Cln8^{mnd}</i>	M	10	Euthanized - humane end point
<i>Cln8^{mnd}</i>	M	10	Euthanized - humane end point
<i>Cln8^{mnd}</i>	M	10	Euthanized - humane end point
<i>Cln8^{mnd}</i>	M	10	Euthanized - humane end point
<i>Cln8^{mnd}</i>	M	10	Euthanized - ataxia, difficulty getting food and water, dropping weight, rough coat
<i>Cln8^{mnd}</i>	M	11	Euthanized - unable to right itself, twitchy and hunched appearance
<i>Cln8^{mnd}</i>	M	12	Euthanized - humane end point
<i>Cln8^{mnd}</i>	M	12	Euthanized - hunched rough coat, priapism, general dyskinesia
<i>Cln8^{mnd}</i>	M	12	Euthanized - hunched rough coat, priapism, general dyskinesia

Supplemental Table 2. Detailed Ns.

N = # of Animals		WT								<i>Cln8^{mnd}</i>								<i>Cln8^{mnd}</i> + AAV9							
Age (Months)		2	4	6	8	10	12	18	24	2	4	6	8	10	12	18	24	2	4	6	8	10	12	18	24
qPCR	Cerebral Cortex	4	5	6	5	6	5	4	6	5	6	4	5	-	-	-	-	7	6	5	6	7	6	5	5
	Cervical SC	3	6	6	6	6	5	5	5	5	6	5	6	-	-	-	-	5	6	5	5	4	6	6	6
	Thoracic SC	1	6	6	6	6	6	4	6	1	5	5	6	-	-	-	-	1	5	5	5	7	5	6	6
	Lumbar SC	3	6	6	4	6	6	3	5	2	6	5	3	-	-	-	-	2	6	5	4	6	6	5	6
	Kidney	4	6	5	3	6	4	5	4	4	5	5	4	-	-	-	-	4	6	4	6	7	5	7	6

N = # of Images		WT								<i>Cln8^{mnd}</i>								<i>Cln8^{mnd}</i> + AAV9							
Age (Months)		2	4	6	8	10	12	18	24	2	4	6	8	10	12	18	24	2	4	6	8	10	12	18	24
ASM	S1BF	8	20	44	41	33	24	28	44	20	22	48	44	-	-	-	-	21	21	47	45	53	20	32	40
	VPM/VPL	8	19	47	43	33	24	32	48	20	24	47	48	-	-	-	-	24	18	48	41	55	20	32	40
SubC	S1BF	33	54	68	57	37	43	42	52	24	41	68	72	-	-	-	-	35	33	69	64	50	66	68	72
	VPM/VPL	35	52	69	56	42	38	38	61	15	44	68	72	-	-	-	-	26	32	62	66	56	67	68	62
GFAP	S1BF	48	50	63	62	60	60	45	68	38	51	72	72	-	-	-	-	40	48	69	68	36	72	68	72
	VPM/VPL	46	50	68	63	58	58	46	62	39	48	64	72	-	-	-	-	35	48	69	68	36	66	72	68
CD68	S1BF	54	53	69	72	46	57	48	63	51	34	72	72	-	-	-	-	45	24	68	68	56	68	62	65
	VPM/VPL	54	54	70	72	48	57	48	68	45	33	63	72	-	-	-	-	42	27	72	68	60	67	68	60

N = # of Animals		WT								<i>Cln8^{mnd}</i>								<i>Cln8^{mnd}</i> + AAV9							
Age (Months)		2	4	6	8	10	12	18	24	2	4	6	8	10	12	18	24	2	4	6	8	10	12	18	24
Pole Climb	Rotarod	20	20	19	20	20	20	19	11	21	21	20	20	5	-	-	-	20	20	20	20	20	20	20	11
	Down	20	19	20	19	20	20	19	11	18	18	19	17	9	-	-	-	18	18	20	20	20	20	19	12
	Turn	20	20	20	20	20	20	18	11	20	20	20	19	8	-	-	-	20	20	18	18	20	20	19	12
	Falls	20	20	20	20	20	20	19	11	21	21	20	20	8	-	-	-	20	20	19	20	20	20	19	12
Water Maze	Time	19	19	20	19	19	20	19	10	15	15	16	10	-	-	-	-	15	15	15	15	12	10	9	6
	Speed	20	20	20	20	20	20	19	10	17	61	16	10	-	-	-	-	15	14	15	14	12	10	9	6
	Reversal Time	-	-	18	-	-	20	19	9	-	-	16	-	-	-	-	-	-	-	15	-	-	10	9	6

	Reversal Speed	-	-	20	-	-	20	18	10	-	-	16	-	-	-	-	-	-	15	-	-	10	9	6
Coordination	Clasping	20	20	20	20	20	20	19	11	21	21	21	20	6	-	-	-	20	20	20	20	20	20	13
	Ledge	20	20	20	20	20	20	19	11	21	21	21	20	6	-	-	-	20	20	20	20	20	20	13
	Gait	20	20	20	20	20	20	19	11	21	21	21	20	6	-	-	-	20	20	20	20	20	20	13
	Combined Score	20	20	20	20	20	20	19	11	21	21	21	20	6	-	-	-	20	20	20	20	20	20	13
Force Plate	Male Weight	8	10	10	10	10	10	10	7	10	10	10	10	5	-	-	-	11	10	10	10	10	10	6
	Female Weight	10	10	10	10	10	10	9	4	10	11	11	10	1	-	-	-	9	9	10	10	10	10	6
	Total Distance	19	20	20	20	20	20	19	11	20	21	21	20	6	-	-	-	20	20	20	19	19	19	12
	Bouts Low Mobility	19	20	20	20	20	20	19	11	20	21	21	20	6	-	-	-	20	20	20	20	20	20	12
	Total Area	19	20	20	20	19	20	19	11	20	21	21	19	6	-	-	-	20	20	20	18	19	19	12
	Focused Stereo	19	20	20	19	20	20	18	11	20	21	20	20	6	-	-	-	20	19	19	18	20	18	12
	Tremor 5-10Hz	20	20	20	20	20	20	19	11	21	21	21	20	6	-	-	-	20	20	20	20	20	20	12
	Tremor 10-15Hz	20	20	20	20	20	20	19	11	21	21	21	20	6	-	-	-	20	20	20	20	20	20	12
	Tremor 15-20Hz	20	20	20	20	20	20	19	11	21	21	21	20	6	-	-	-	20	20	20	20	20	20	12
	Tremor 20-25Hz	20	20	20	20	17	20	19	11	21	21	21	20	6	-	-	-	20	20	20	20	20	20	12