Supplemental Data
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IDS Crossing of the Blood-Brain Barrier

Corrects CNS Defects in MPSII Mice

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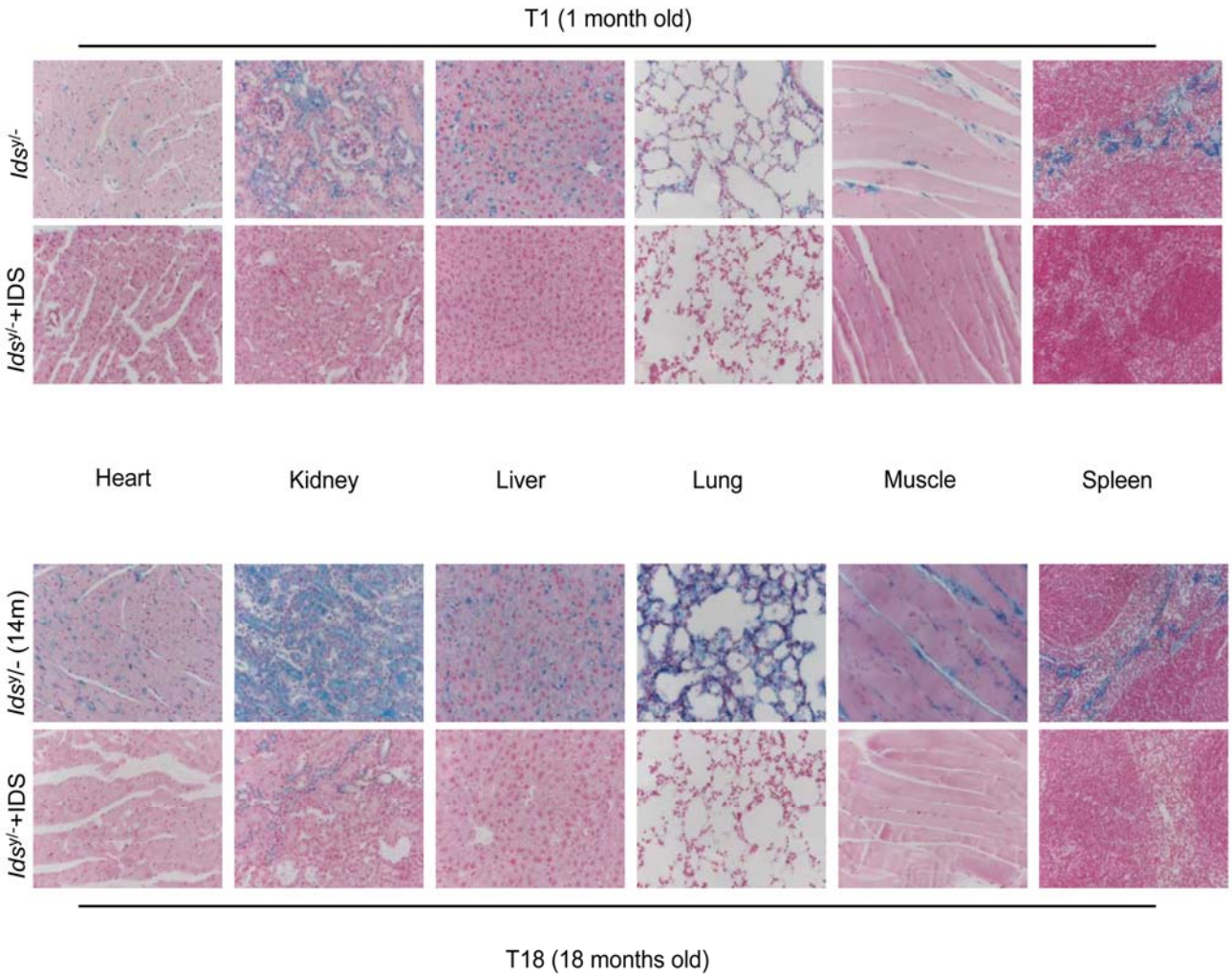

Figure S1. Clearance of GAG Accumulation in all Tissues of *Ids*$^{y/-}$+IDS (T1) and *Ids*$^{y/-}$+IDS (T18) Injected Mice

Qualitative GAG accumulation measured by Alcian Blue staining of sections of all tissues of *Ids*$^{y/-}$ (T1-T14) and *Ids*$^{y/-}$+IDS (T1-T18) mice. Magnification, 20x.

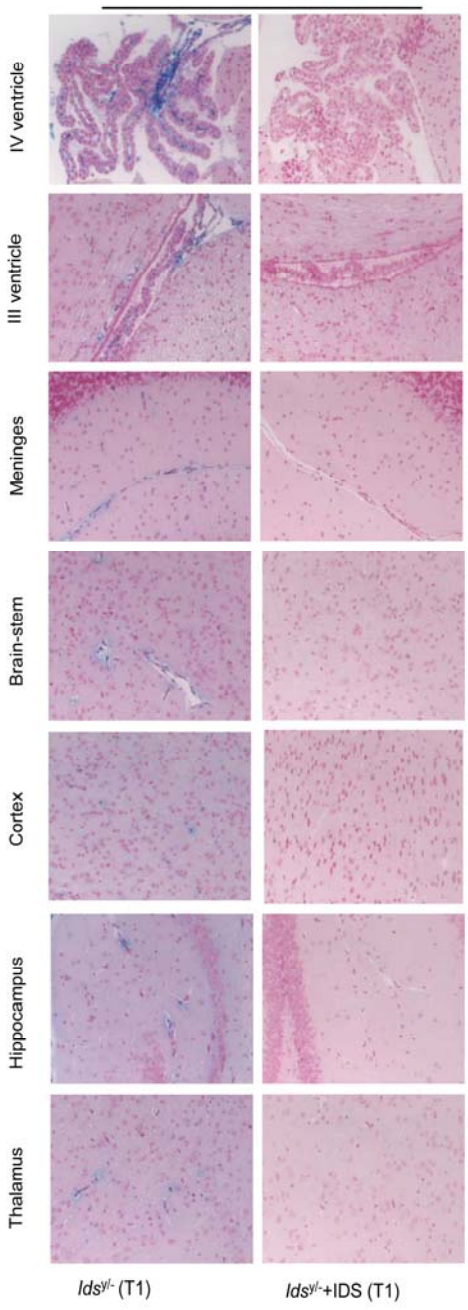

Figure S2. Clearance of GAG Accumulation in *Ids*$^{y/-}$+IDS (T1) Mice

Alcian Blue stained sections of different regions of the brain of *Ids*$^{y/-}$ (T1) and *Ids*$^{y/-}$+IDS (T1) mice. Magnification, 20x.

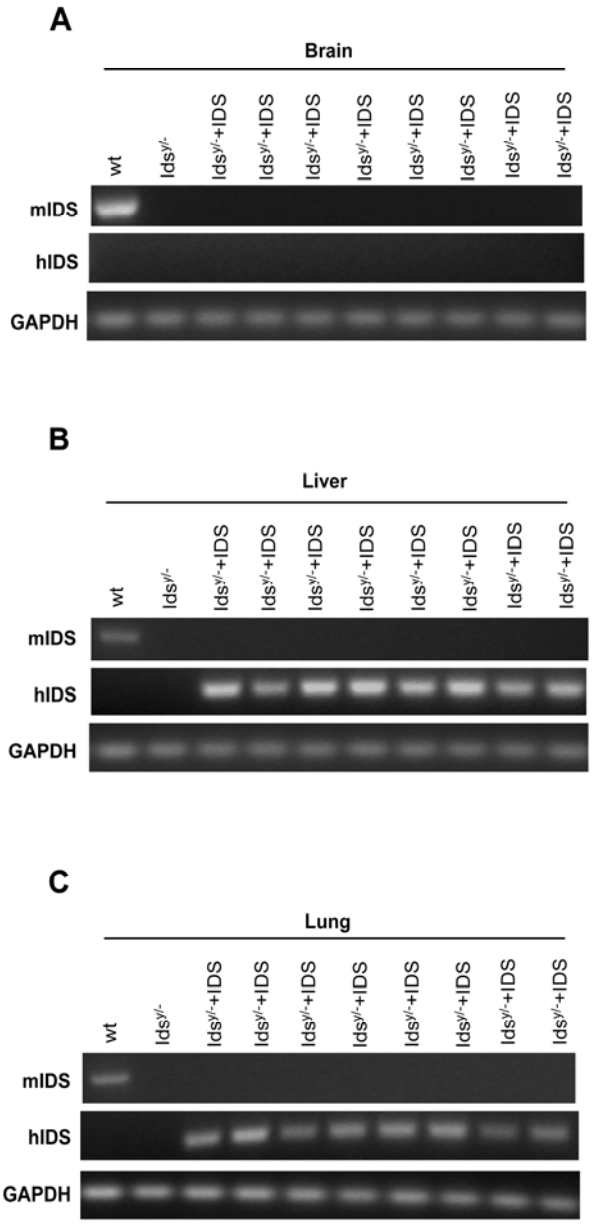

Figure S3. IDS Protein Crossing of the BBB in *Ids*$^{y/-}$+IDS (T1) Injected Mice

(A-C) RT-PCR of hIDS and mIDS mRNAs from brains, livers and lungs of wt (T1), *Ids*$^{y/-}$ (T1), *Ids*$^{y/-}$+IDS (T1) mice.

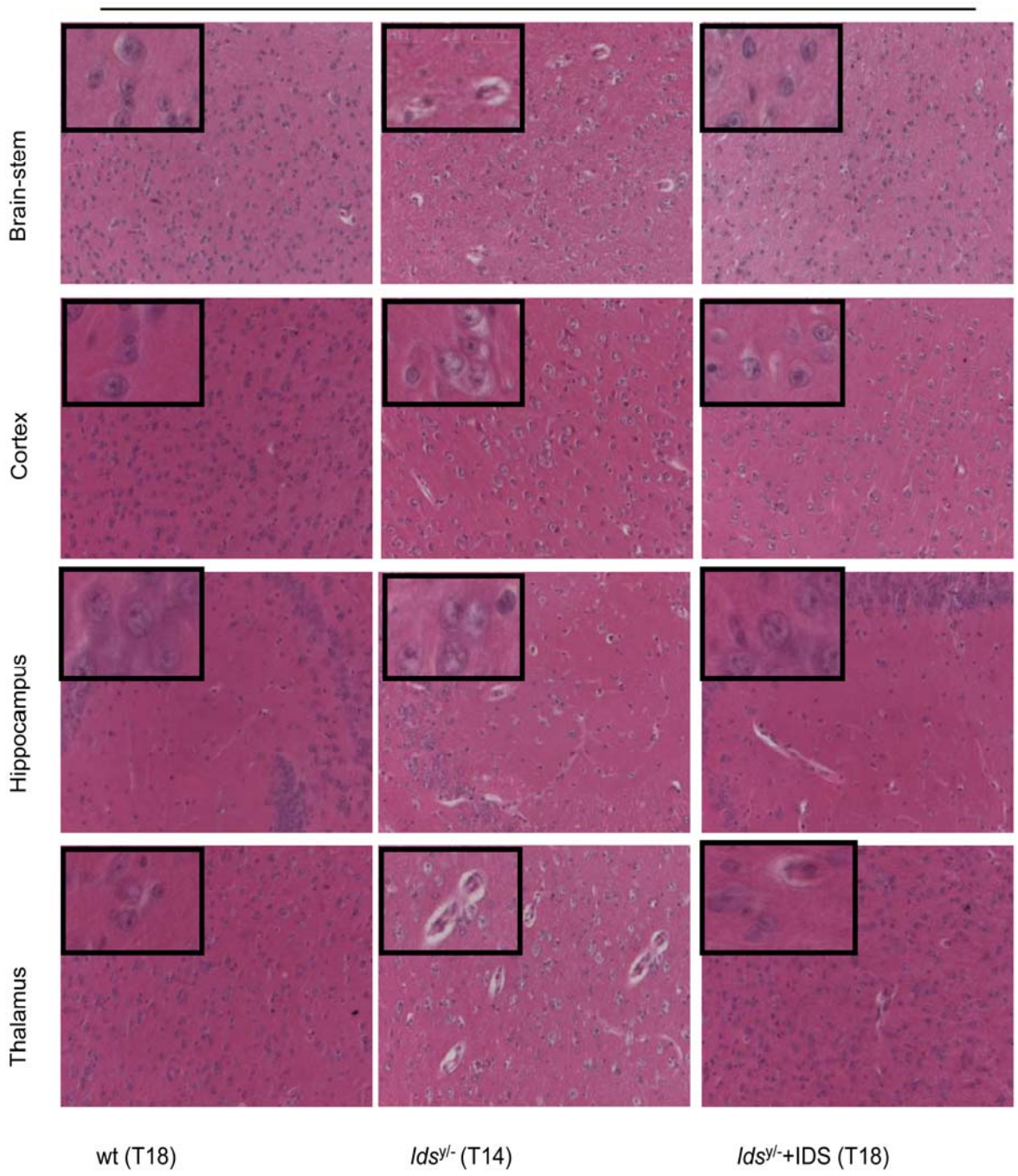

Figure S4. Clearance of Vacuolization in Brain Sections of Treated Mice

H&E stained sections of different regions of brains of wt (T18), *Ids*$^{y/-}$ (T14) and *Ids*$^{y/-}$+IDS (T18) mice. Magnification, 20x (black box, 40x).

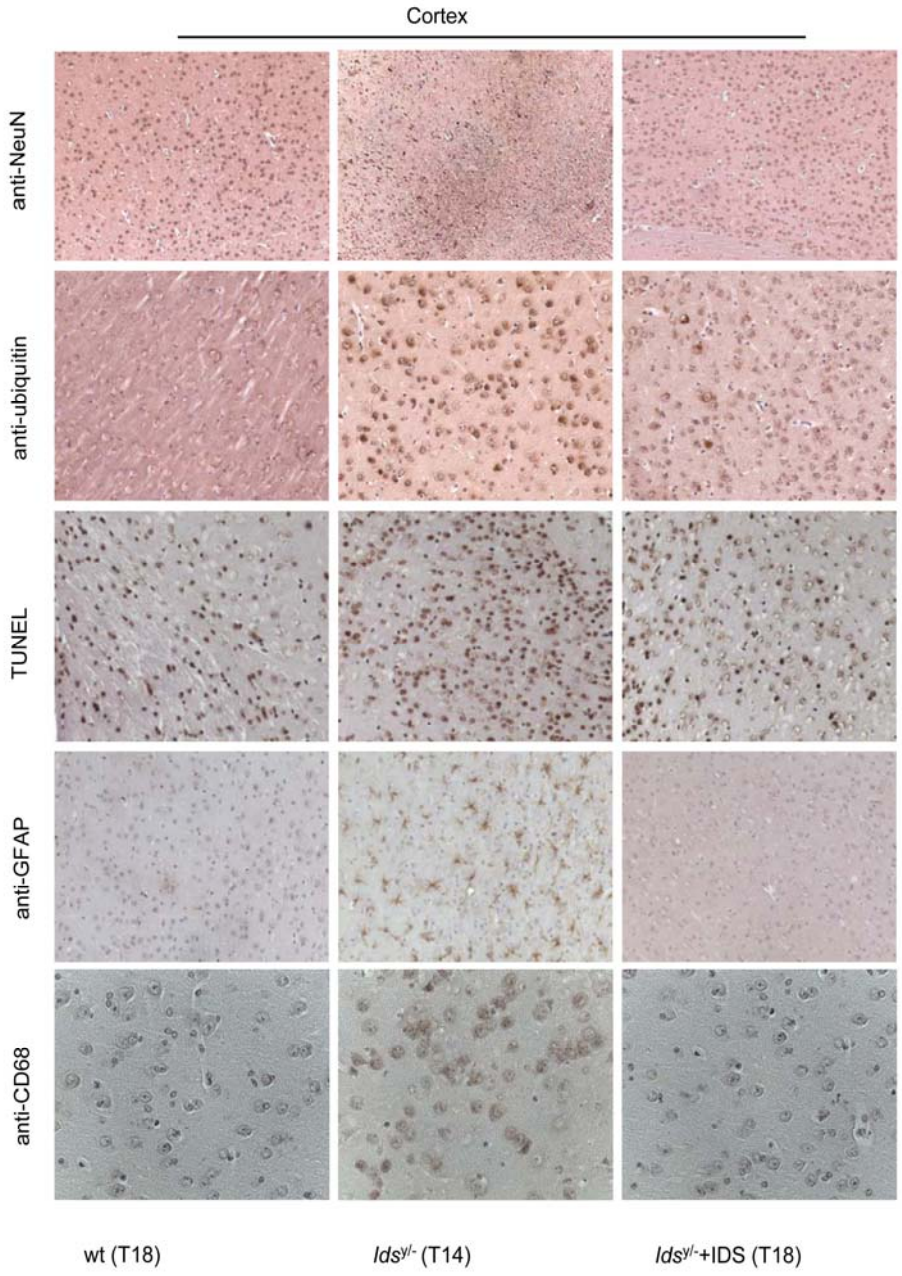

Figure S5. Rescue of Brain Defects in *Ids*$^{y/-}$ Treated Mice

Immunohistochemistry of different specific brain markers (anti-NeuN, anti-ubiquitin, TUNEL, anti-GFAP and anti-CD68) in cortex of wt (T18), *Ids*$^{y/-}$ (T14), *Ids*$^{y/-}$+IDS (T18) brains. Magnification, 10x (anti-NeuN sections); 40x (anti-CD68 sections); 20x (all others).

Figure S6. Rescue of Brain Defects in *Ids*$^{y/-}$ Treated Mice

Immunohistochemistry of different specific brain markers (anti-NeuN, anti-ubiquitin, TUNEL, anti-GFAP and anti-CD68) in brain stem of wt (T18), *Ids*$^{y/-}$ (T14), *Ids*$^{y/-}$+IDS (T18) brains. Magnification, 10x (anti-NeuN sections); 40x (anti-CD68 sections); 20x (all others).

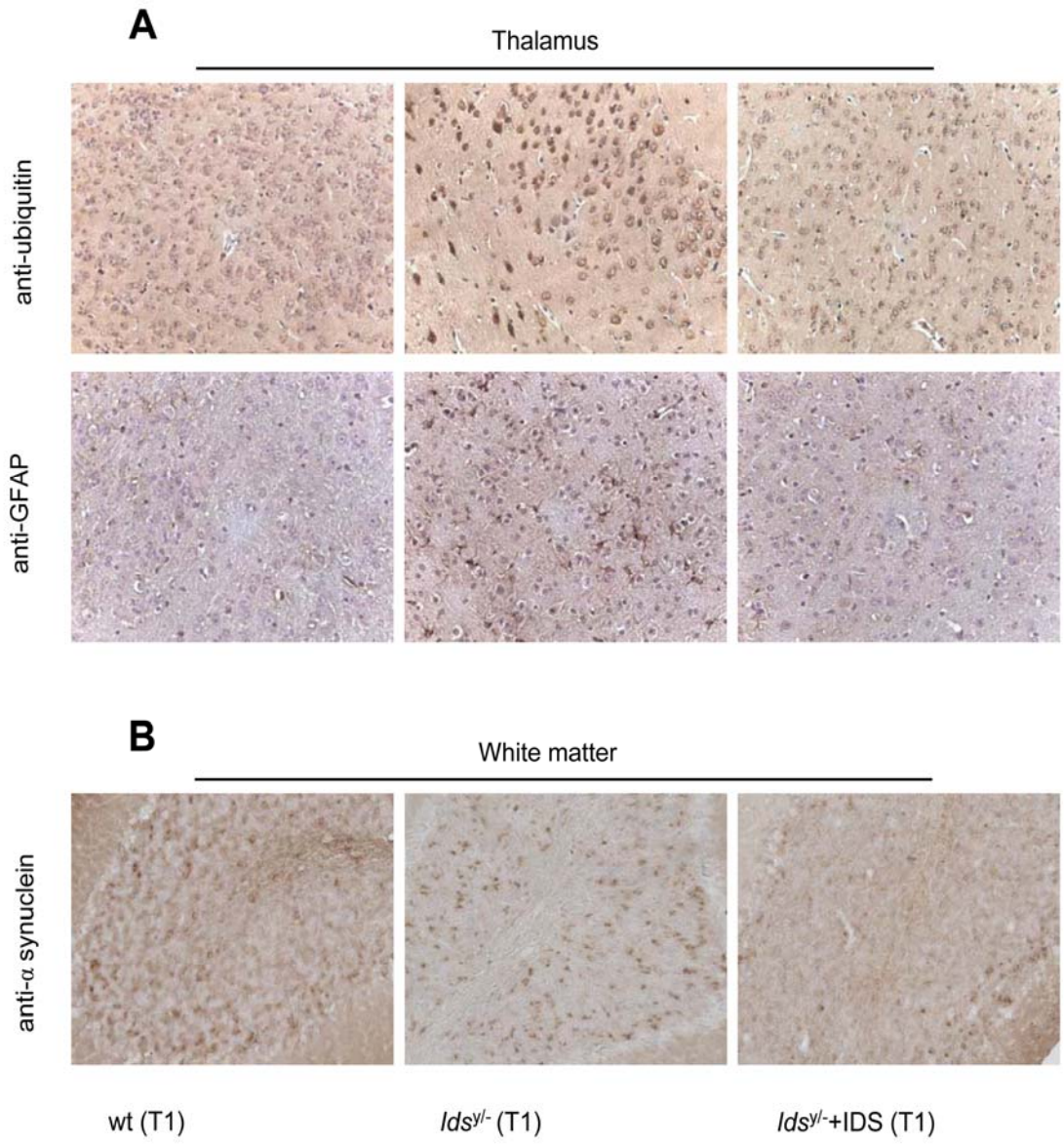

Figure S7. Rescue of Brain Defects in *Ids*$^{y/-}$ Treated T1 Mice

(A) Immunohistochemistry of specific brain markers (anti-ubiquitin, anti-GFAP) in thalamus of wt (T1), *Ids*$^{y/-}$ (T1) and *Ids*$^{y/-}$+IDS (T1) brain sections. **(B)** Immunohistochemistry of anti-α-synuclein in white matter of the same groups of mice. Magnification, 20x.

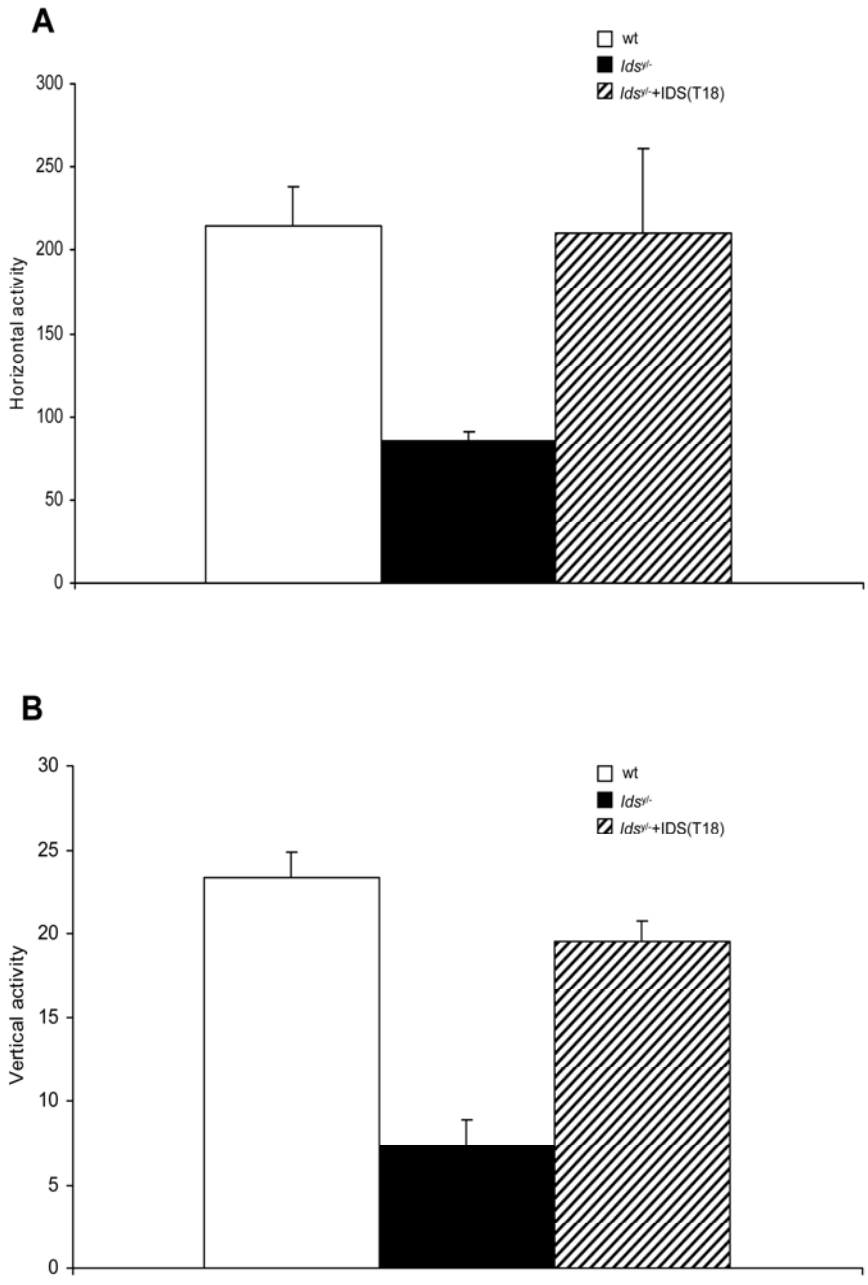

Figure S8. *Ids*$^{y/-}$ Treated Mice Underwent the Open-Field Test

(A, B) Horizontal and vertical activities measured in *Ids*$^{y/-}$+IDS (T18, *n* = 4) and control *Ids*$^{y/-}$ (T14, *n* = 3) mice. The error bars indicate standard deviations. *P* <0.05 (Student's *t*-test).

Table S1. GAG Accumulation in the Urine

	GAG Accumulation (mg GAG/mg creatinine)			
	T1	T6	T12	T18
wt	16.0 ±0.6	17.0 ±1.0	19.0 ±1.0	19.0 ±1.1
Ids $^{y/-}$	34.0 ±1.1	47.0 ±2.0	51.0 ±1.2	
Ids $^{y/-}$+IDS	22.0			
Ids $^{y/-}$+IDS	21.5			
Ids $^{y/-}$+IDS	22.3			
Ids $^{y/-}$+IDS	24.0			
Ids $^{y/-}$+IDS	23.0			
Ids $^{y/-}$+IDS	24.4			
Ids $^{y/-}$+IDS	21.0			
Ids $^{y/-}$+IDS	22.8			
Ids $^{y/-}$+IDS	21.0	22.5	22.0	25.0
Ids $^{y/-}$+IDS	22.0	24.0	27.0	28.0
Ids $^{y/-}$+IDS	24.0	25.1	23.0	29.0
Ids $^{y/-}$+IDS	23.2	24.5	24.0	26.0

GAG accumulation in the urine measured at different times after therapy (T1-T6-T12-T18) (1-6-12-18 months after the injection, respectively) of wt (n = 3), *Ids*$^{y/-}$ (n = 3) and *Ids*$^{y/-}$ AAV2/5CMV-hIDS-injected mice. P<0.05 (Student's t-test). GAG concentrations were normalized against urine creatinine contents.