



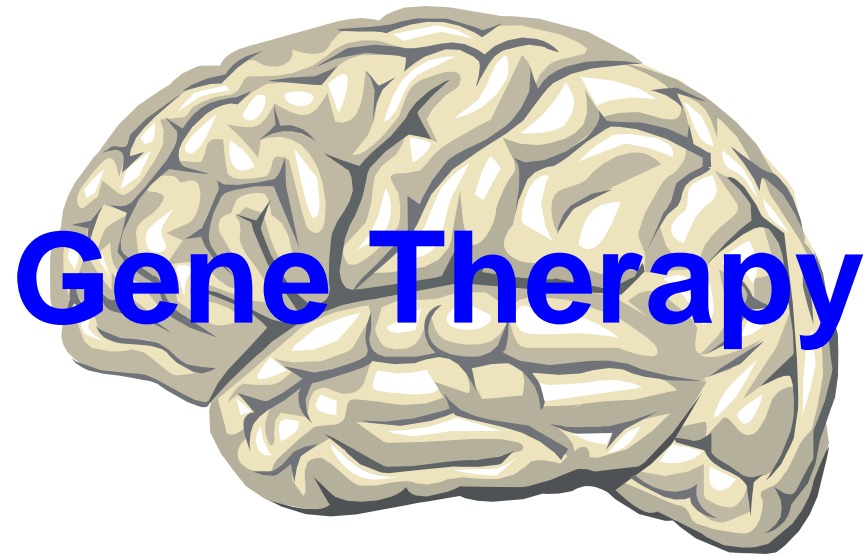
Disclaimer:

Presentation slides from the Rare Disease Workshop Series are posted by the Kakkis EveryLife Foundation, for educational purposes only. They are for use by drug development professionals and statisticians, and are not to be used to guide the prescribing or use of any of the drugs mentioned in the slides. To obtain information on a particular drug, refer to the drug labeling. Do not reproduce or distribute the slides (full set or any portion of) without the permission of the author.

Gene Therapy for CNS Manifestations of the Lysosomal Storage Disorders

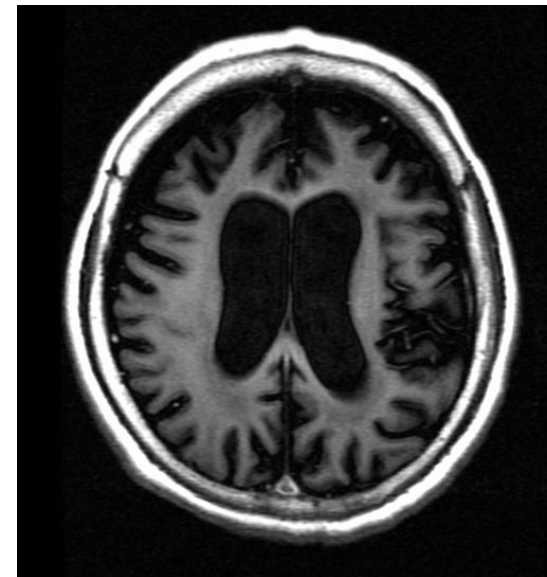
**Rare Disease Workshop
Clinical Evaluation of Rare Disease
Treatment
Washington, DC**

**R. Crystal
Weill Cornell Medical College
6-15-11**



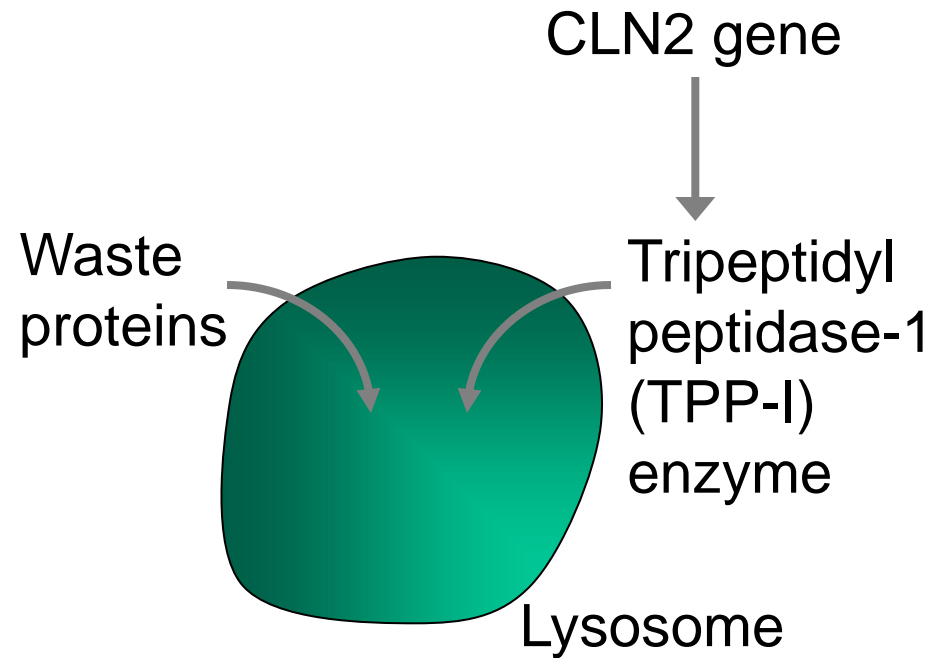
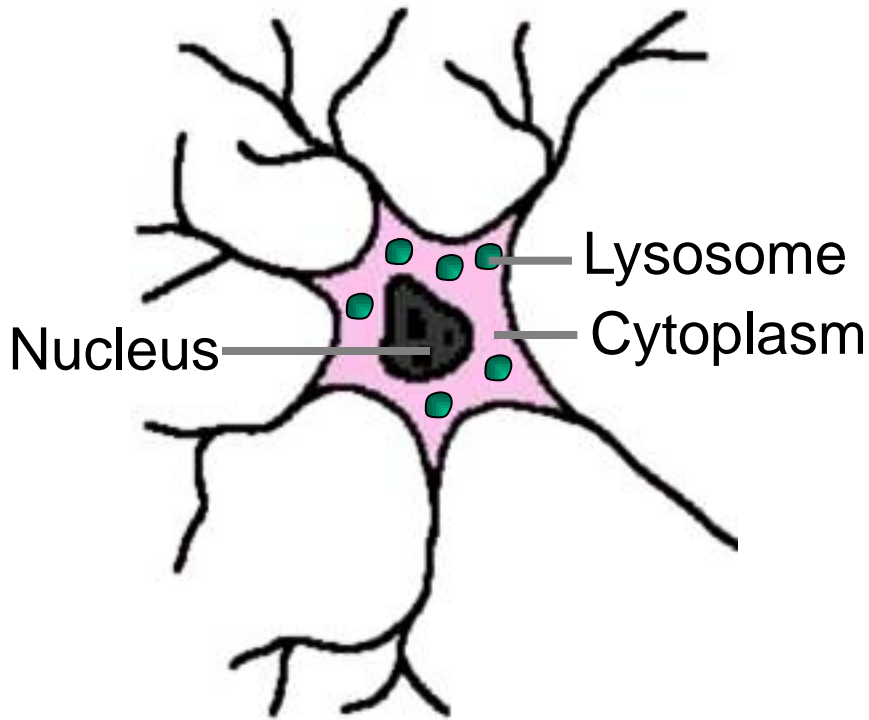
Late Infantile Neuronal Ceroid Lipofuscinoses (LINCL, Batten Disease)

- Autosomal recessive, ~ 200-600 cases worldwide
- Disease onset ages 2-4
- Cognitive impairment, visual failure, seizures, and deteriorating motor development, leading to a vegetative state and death by ages 8-12



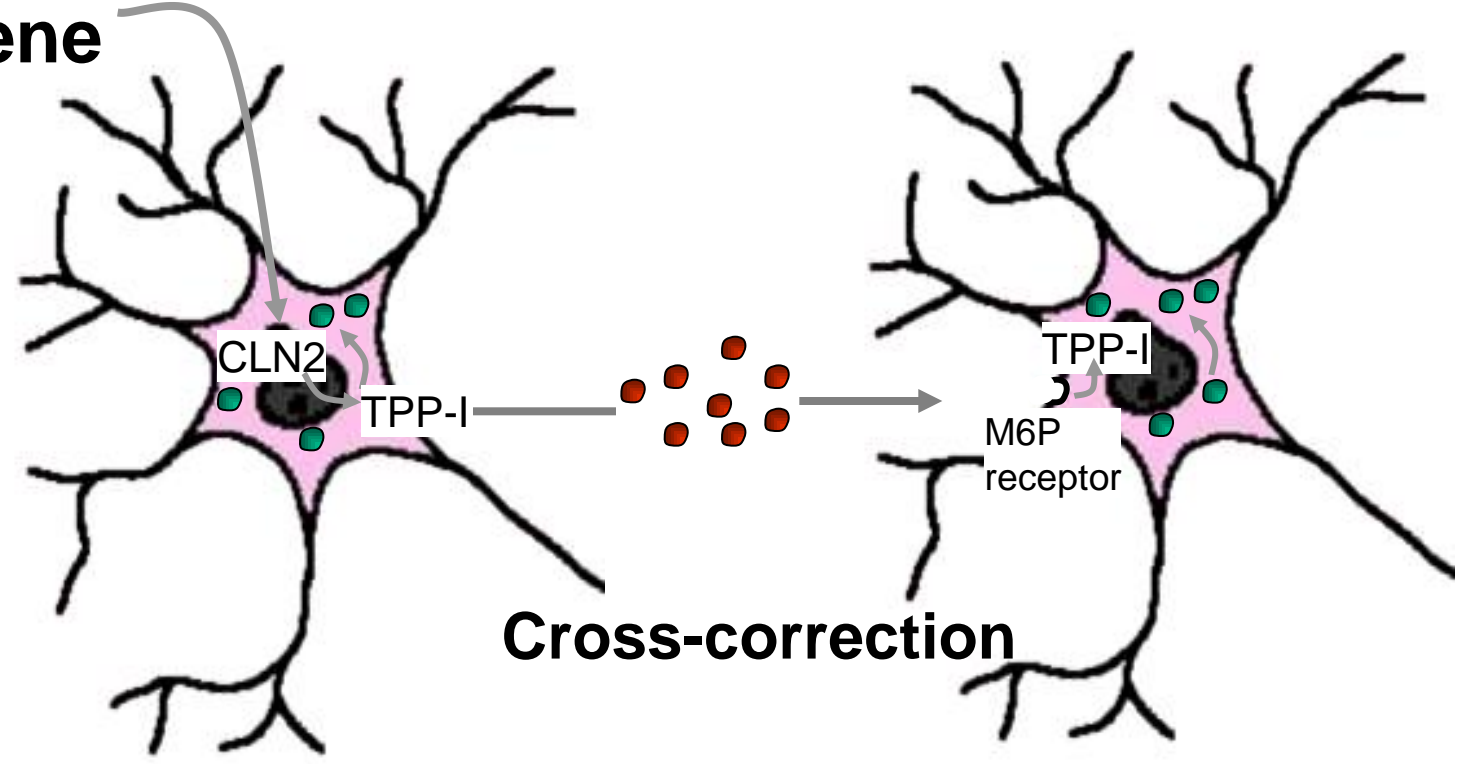
LINCL Is Caused by Mutations in the CLN2 Gene

Neuron

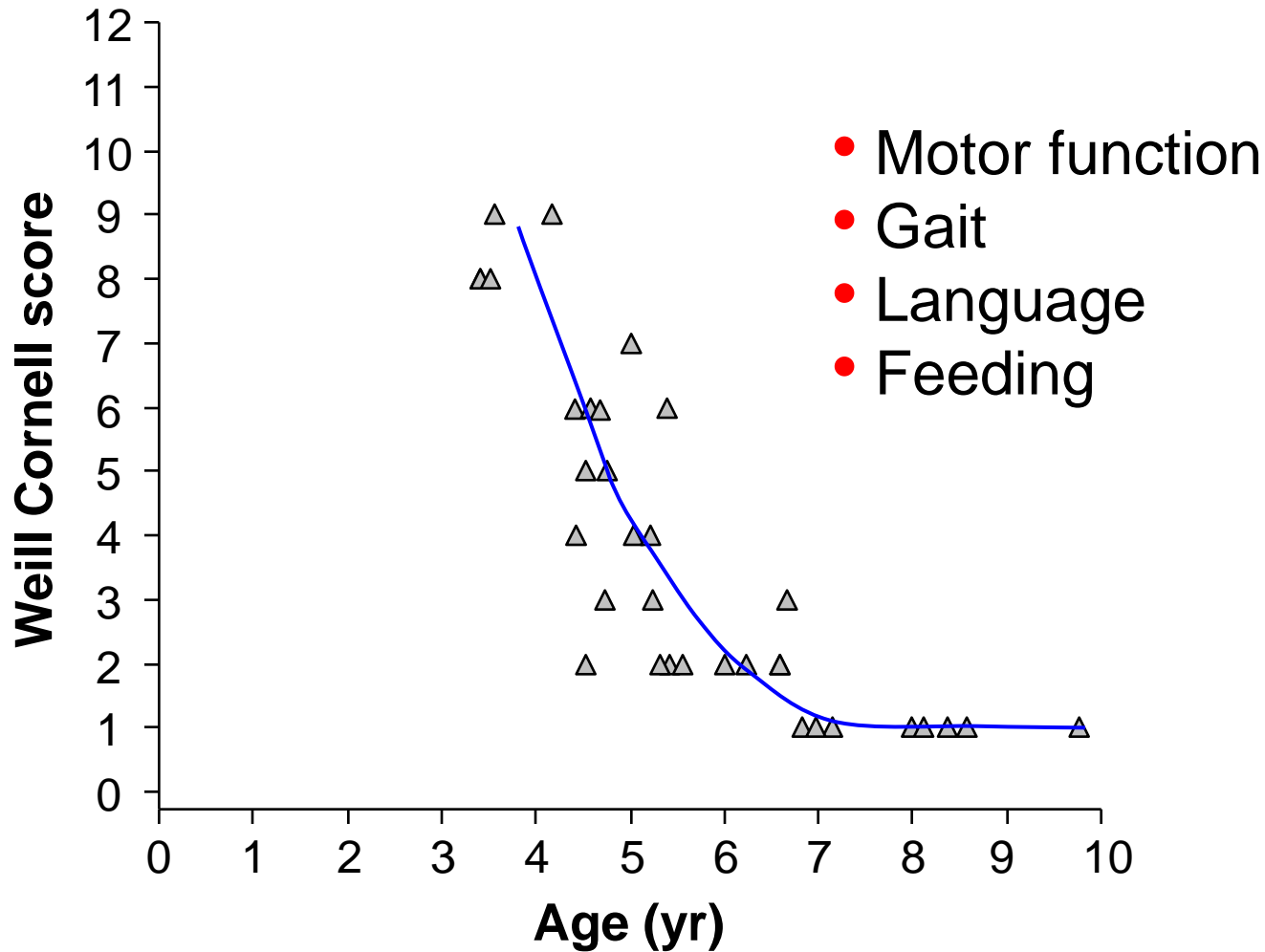


Cross-correction of CNS Cells via the Mannose-6-phosphate Pathway

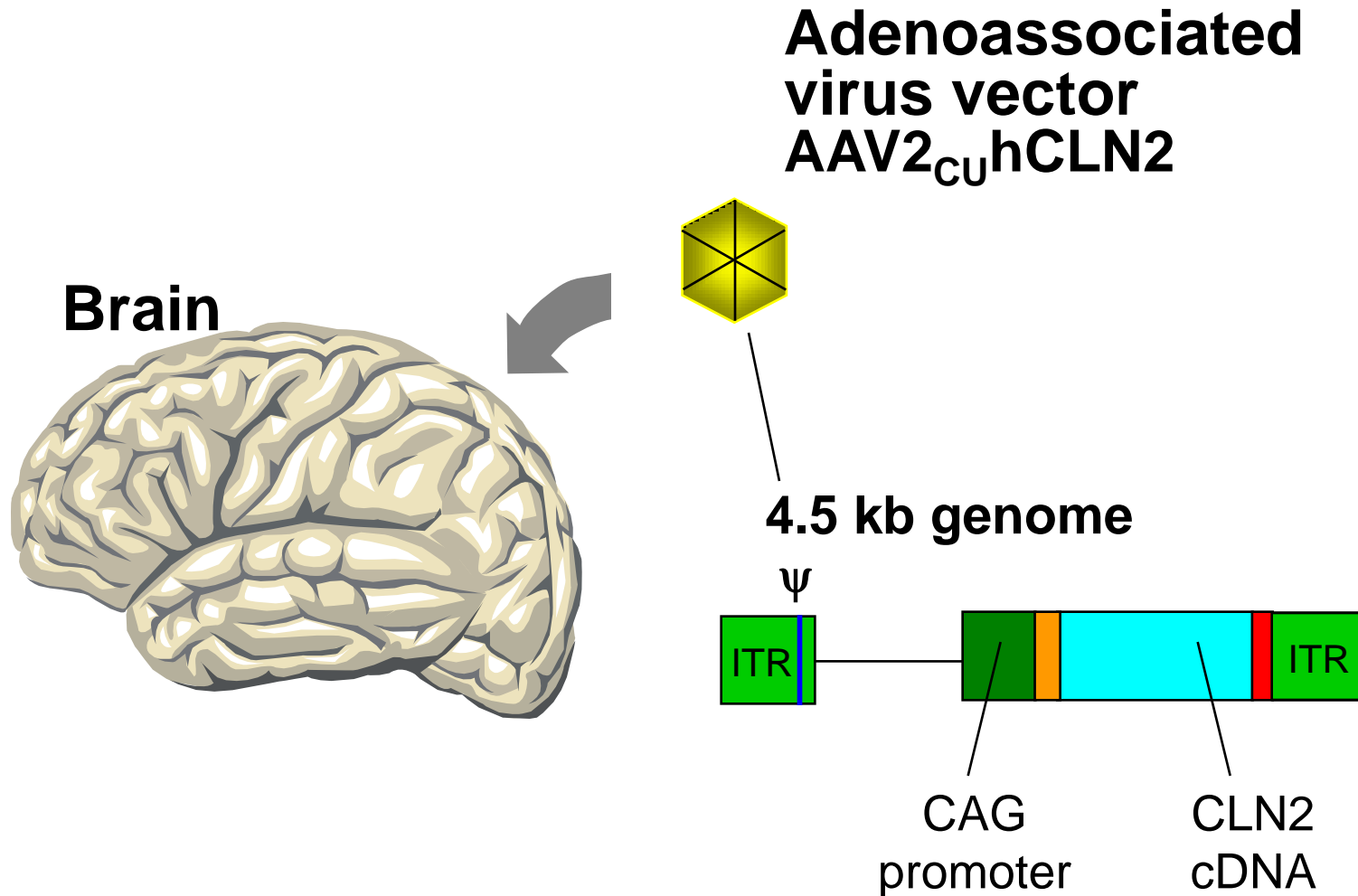
CLN2 gene



LINCL Age-dependent Changes in the Weill Cornell LINCL Score

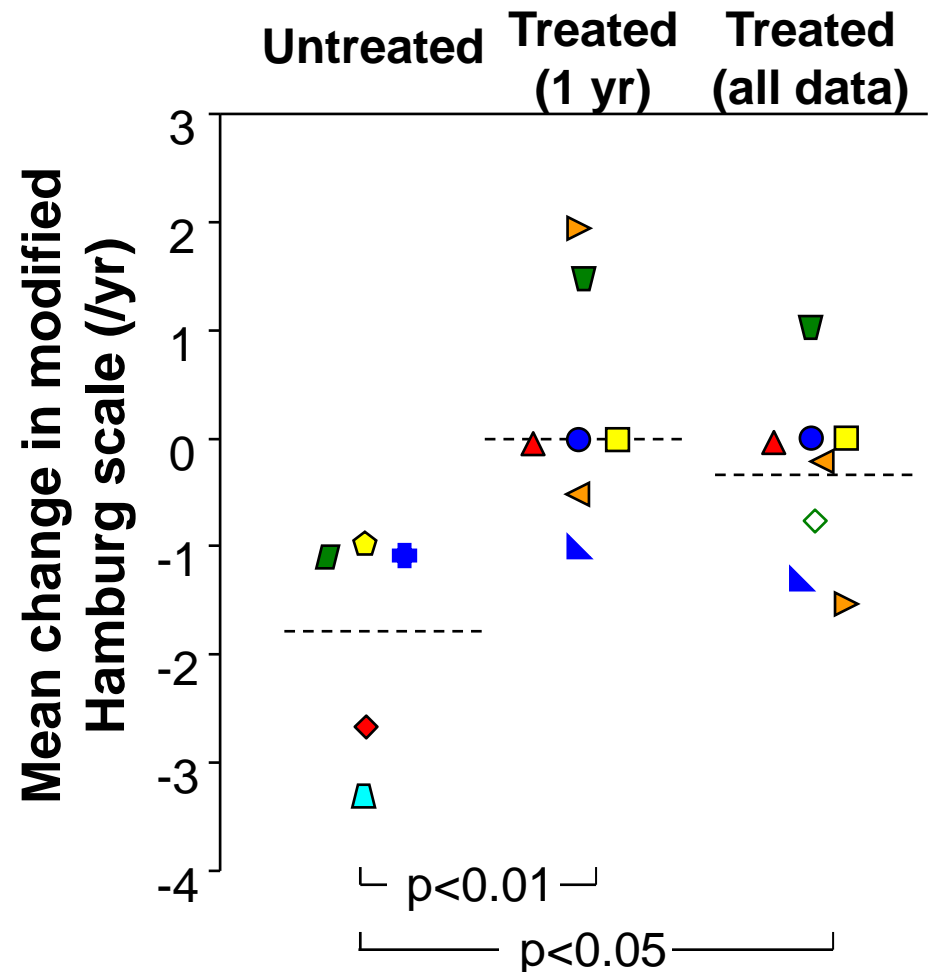


Gene Therapy for LINCL

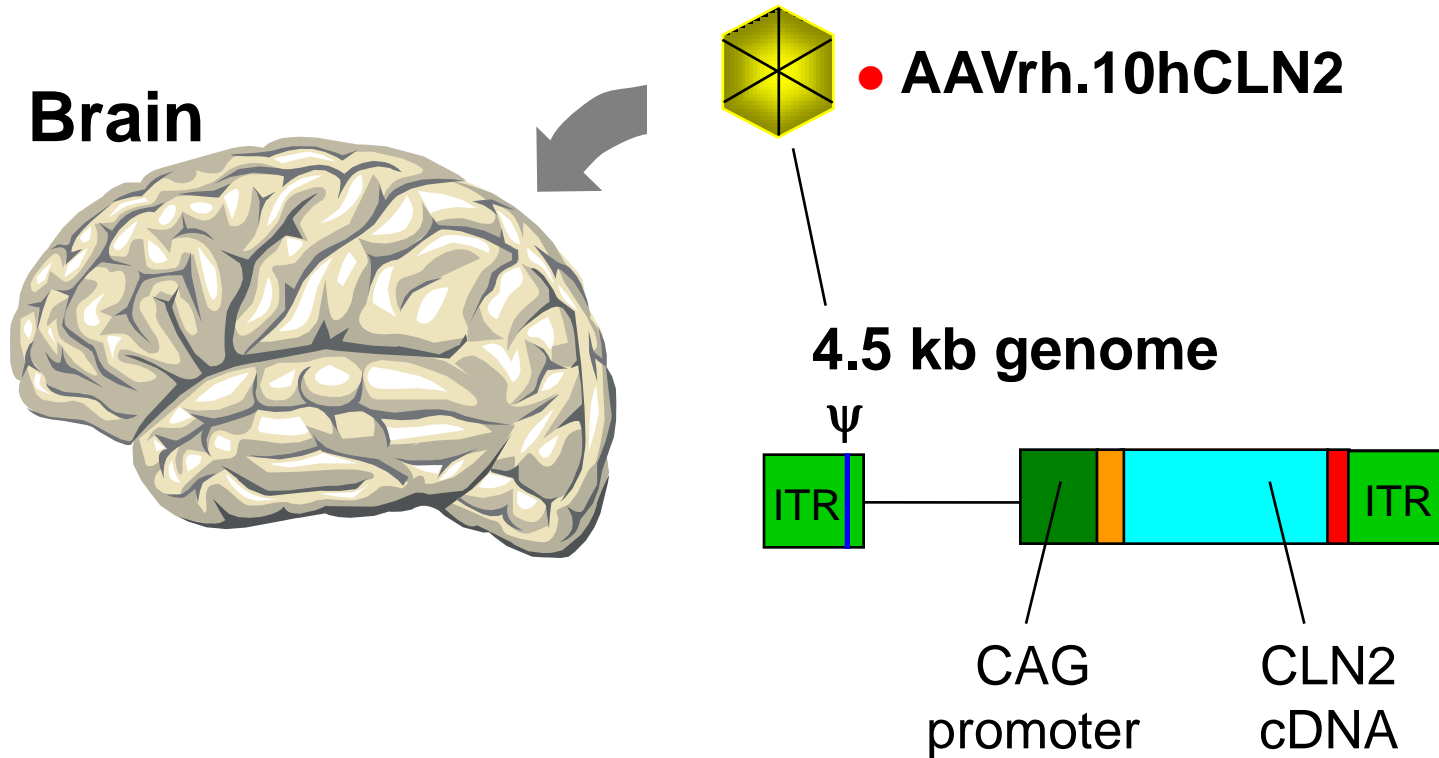


AAV2hCLN2 Gene Therapy Trial for the CNS Manifestations of LINCL

- n=10, 5 severe, 5 moderate
- Total dose 2×10^{12} particle units, 12 sites
- Outcome
 - MRI trend, not significant
 - Hamburg scale significant but with “random” controls

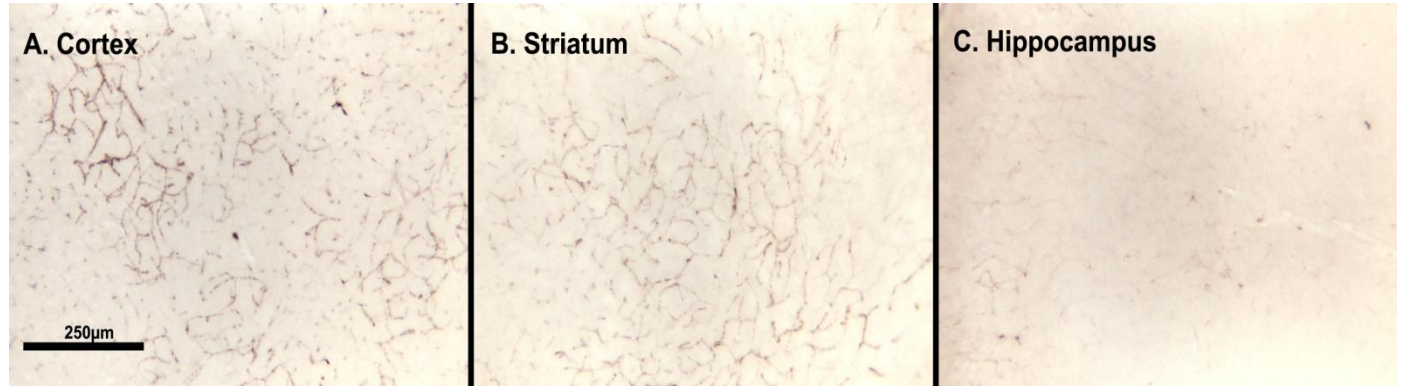


2nd Generation Gene Therapy for LINCL

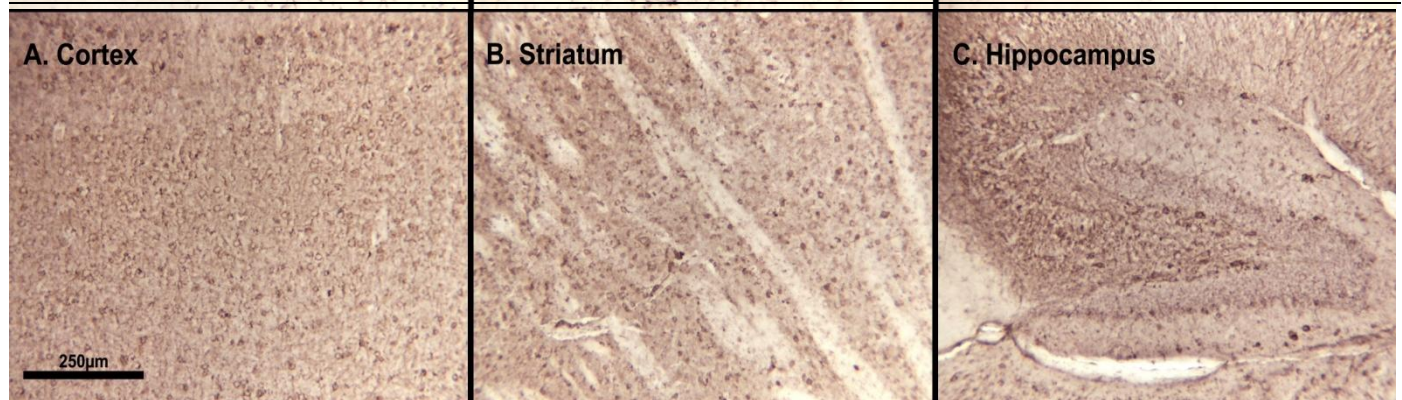


CNS TPP-I Expression Following AAVrh.10hCLN2 Gene Transfer to Day 2 CLN2 -/- Mice and Assessment at 1 yr

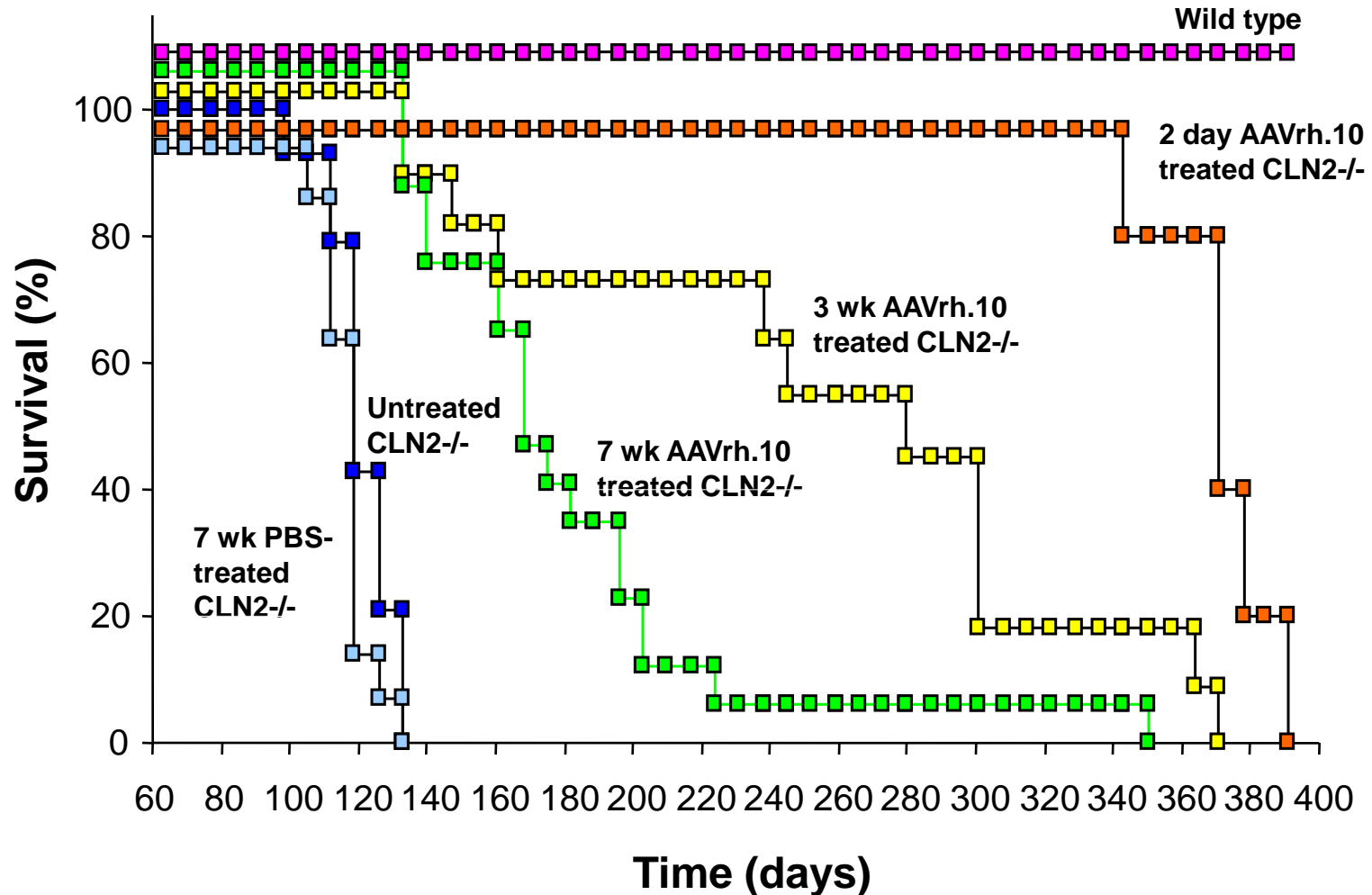
PBS control



AAVrh.10hCLN2



Survival of CLN2^{-/-} Mice Treated at Different Times with AAVrh.10hCLN2

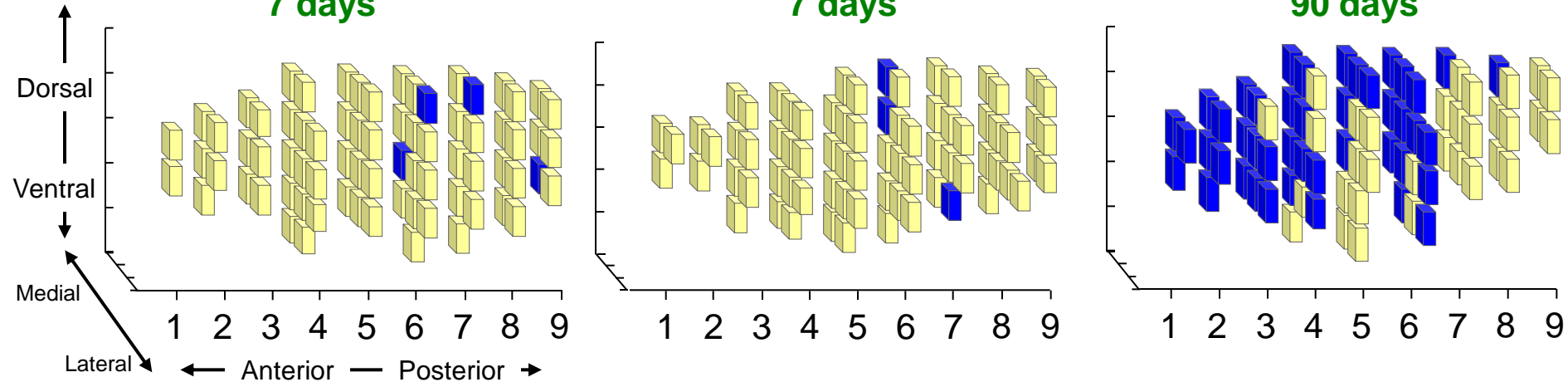




Distribution of TPP-I Activity in the CNS of Non-human Primates Following CNS Administration of AAVrh.10_{CU}hCLN2

NHP Y132
PBS
7 days

NHP 066
AAVrh.10_{CU}hCLN2
7 days

NHP 029
AAVrh.10_{CU}hCLN2
90 days



 < 2 Standard deviations (SD)  > 2 SD

Overall Trial Design

Subject with LINCL

Screening protocol
(5 genotypes)

Not eligible

Eligible

Family given choice to continue in
screening protocol or enter
treatment protocol

Consent process
independent of PI, includes
CTSC patient advocate

Untreated
n=16

Assess efficacy parameters
at 18 months

Treatment with AAVrh.10hCLN2
n=16

1st dose cohort
n=8

7.5×10^{10} gc/site
12 sites

Total dose 9.0×10^{11} gc

Assess efficacy
parameters at 6, 12, 18
months

2nd dose cohort
n=8

1.5×10^{11} gc/site
12 sites

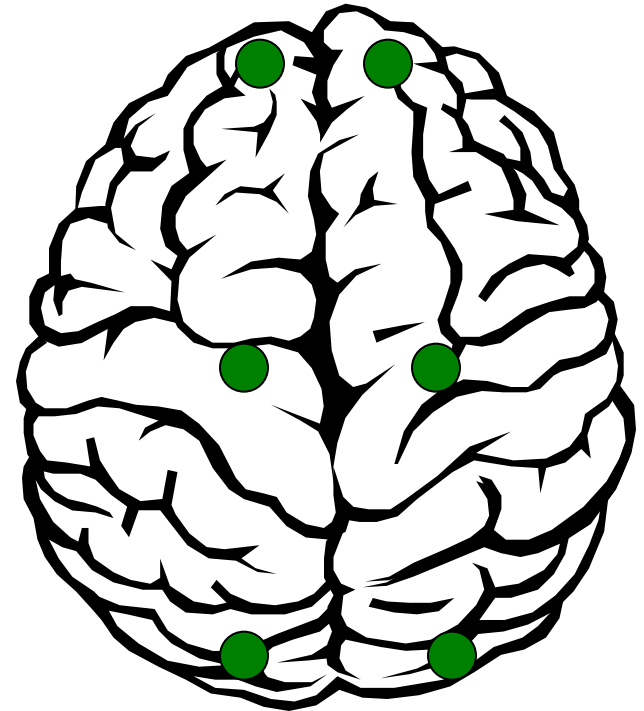
Total dose 1.8×10^{12} gc

Assess efficacy
parameters at 6, 12, 18
months

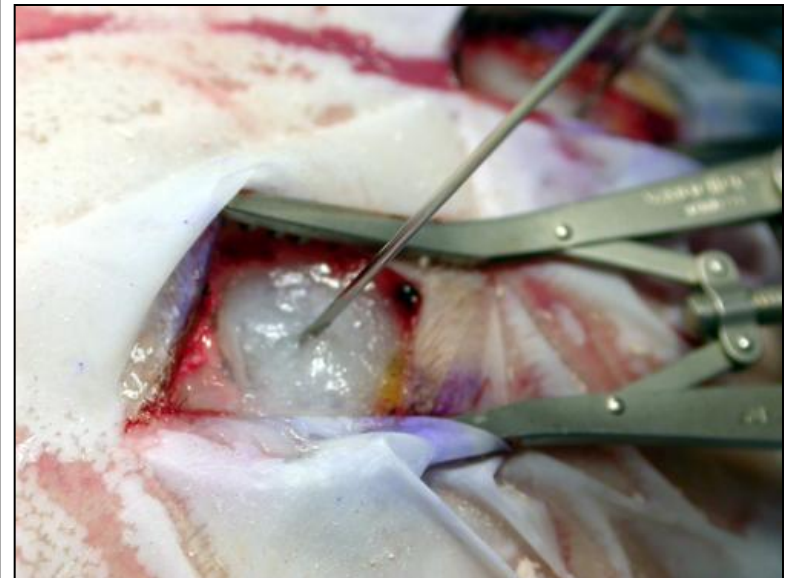
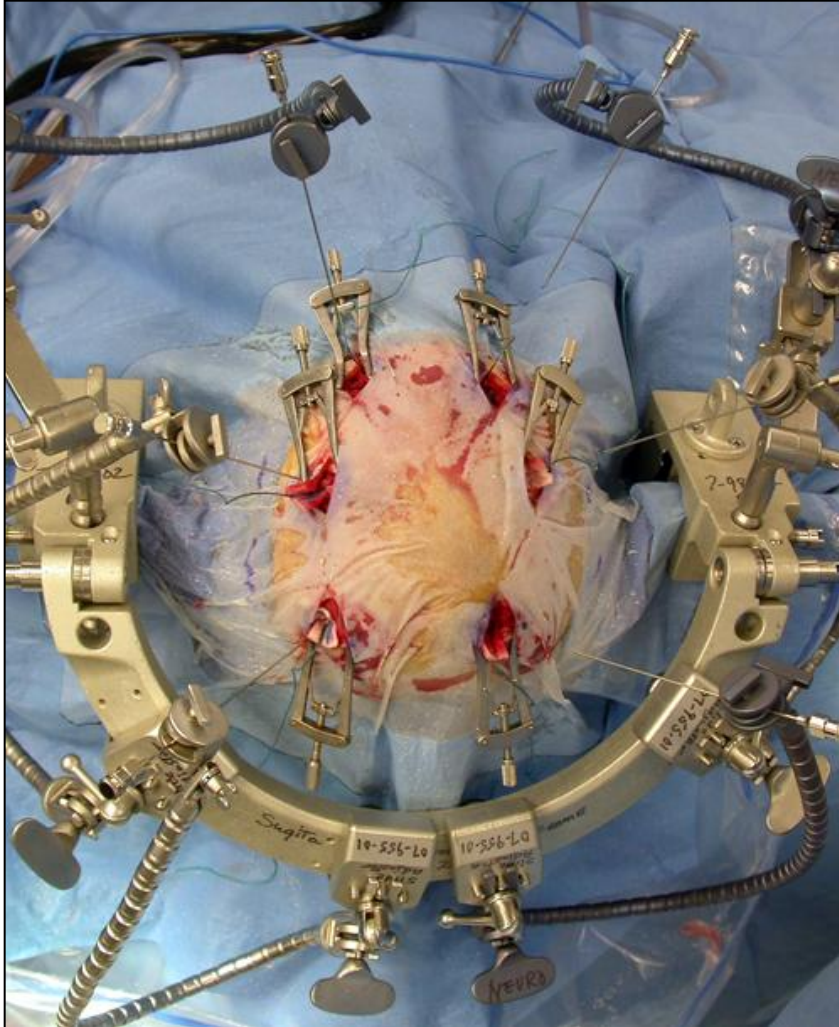
Decision by 4 faculty, representing
3 departments, independent of PI
LINCL mild-moderate

Study Design

- 6 burr holes
- Total doses – 9.0×10^{11} and 1.8×10^{12} genome copies
- 2 injections (different levels) per burr hole
- 2 $\mu\text{l}/\text{min}$ to minimize damage, 300 $\mu\text{l}/\text{burr hole}$ (150 μl at each of 2 sites)



AAV Vector CNS Administration



Critical Path in Developing Therapies for Rare Disorders of the CNS

PHENOTYPE

PHENOTYPE

PHENOTYPE

Outcome Measures

Primary

- Weill Cornell LINCL score¹
- Quantitative MRI

Secondary

- Mullen score
- CHQ Quality of Life questionnaire

¹ Video, blinded assessment by 4 independent pediatric neurologists

Critical Need – To Develop Quantitative Phenotypes to Assess Efficacy

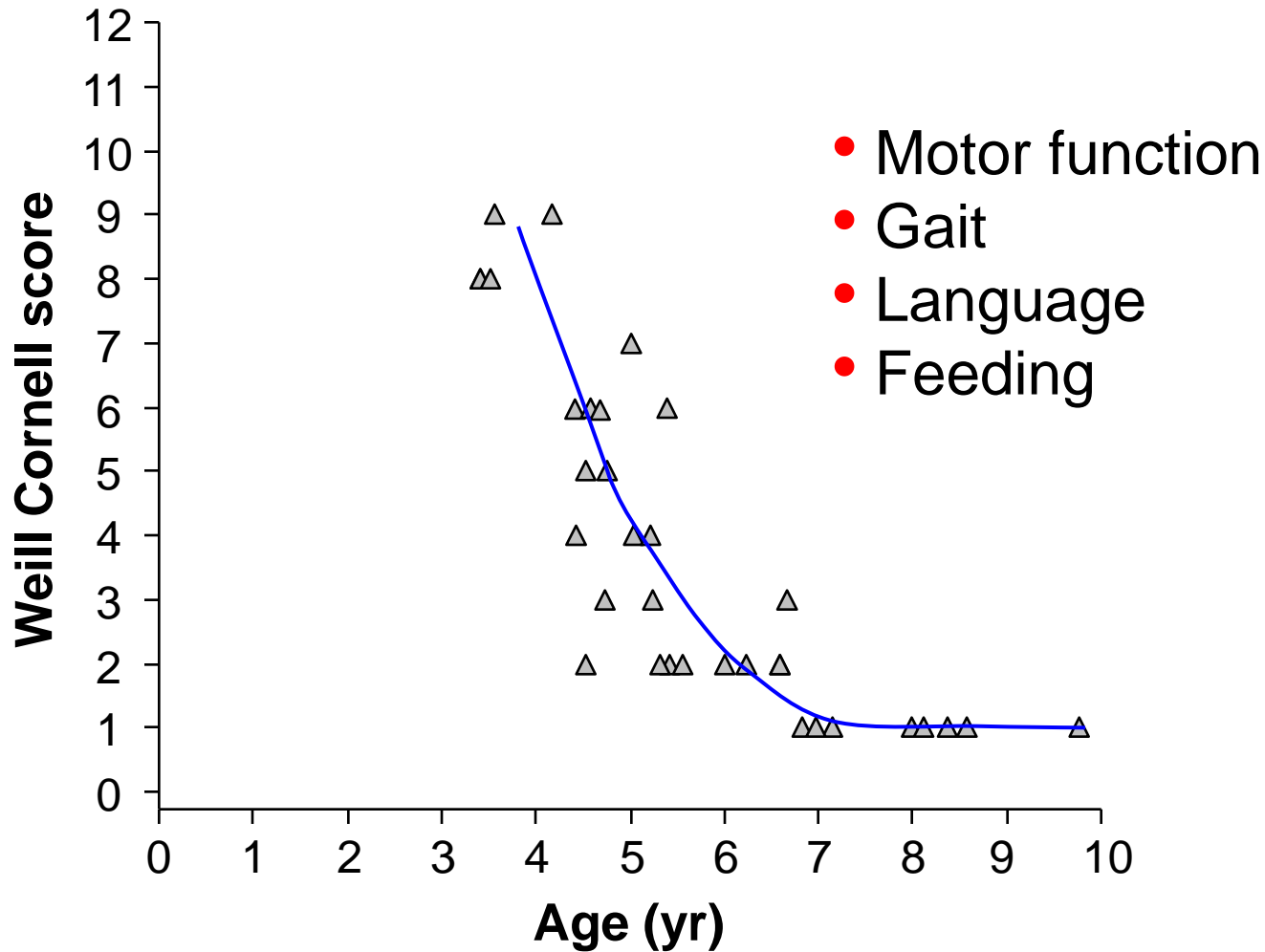
Clinical scales

- Quantitative clinical scales are, at best, integrated measures of overall CNS function
- To limit observer variability, use 4 blinded observers to assess videotapes

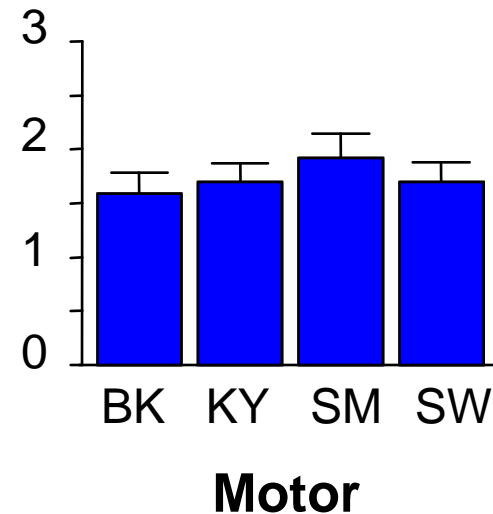
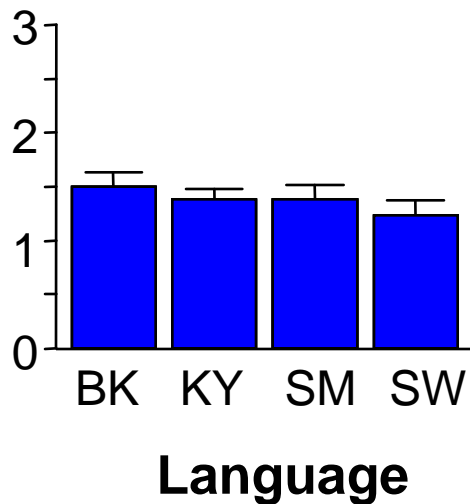
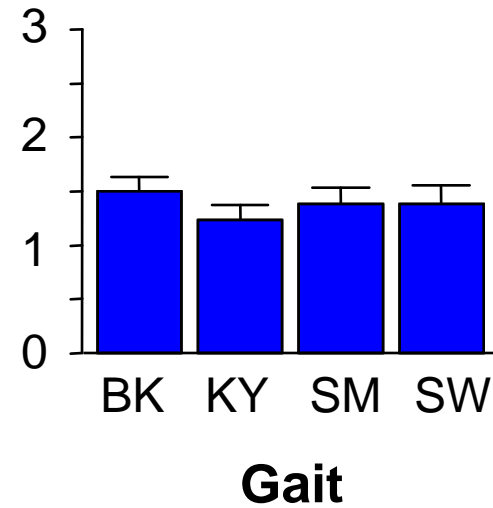
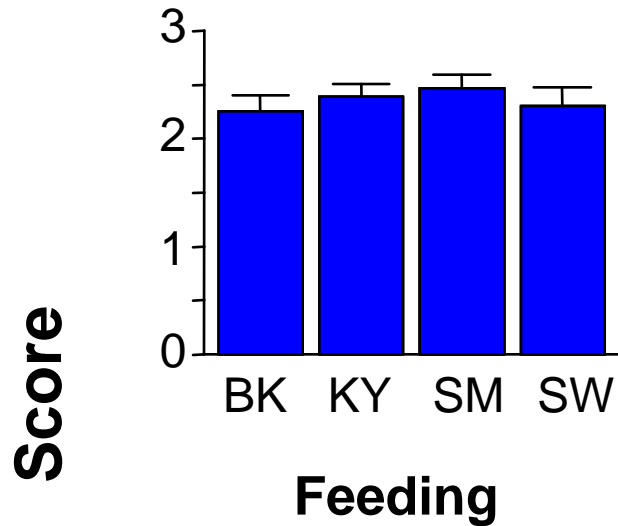
Quantitative MRI

- Evolving methodology that requires more data and verification

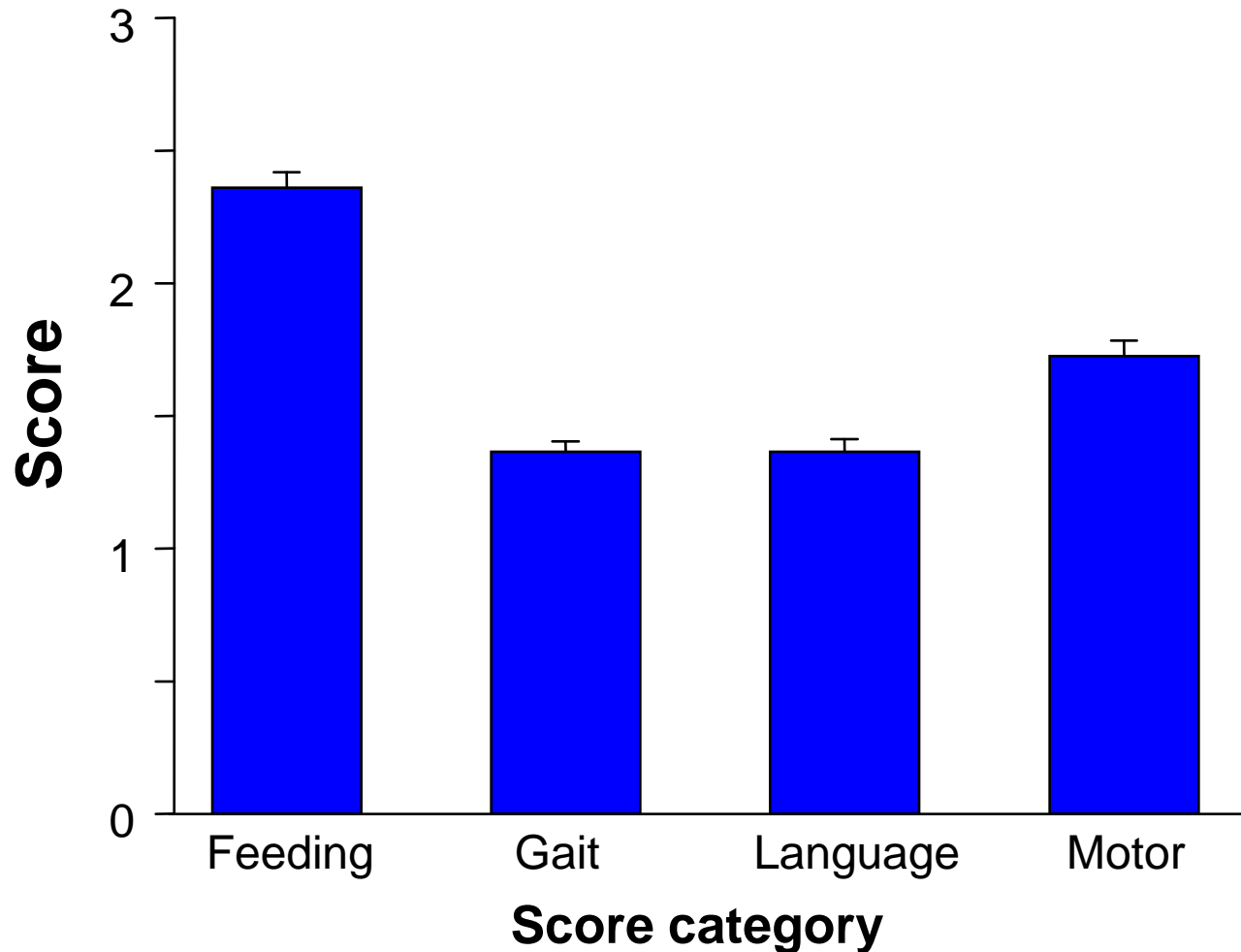
LINCL Age-dependent Changes in the Weill Cornell LINCL Score



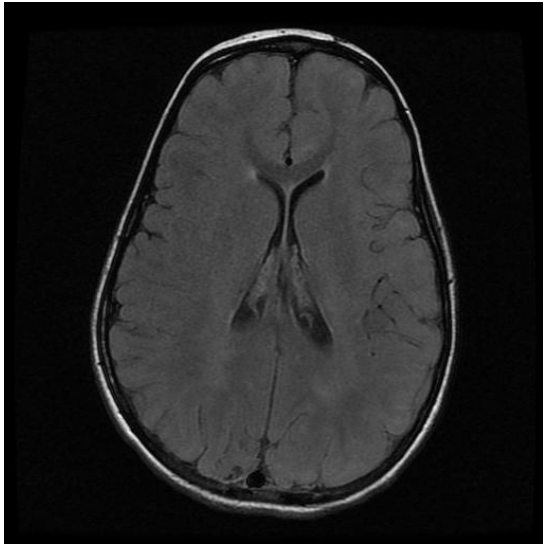
Variation in LINCL Individual Category Scores Among Four Pediatric Neurologists



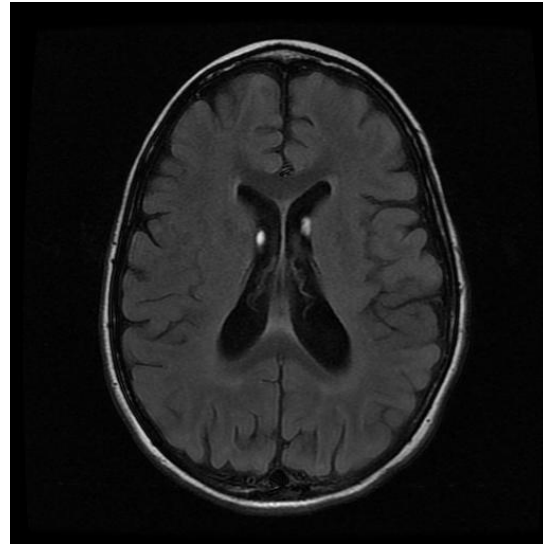
Mean and Average Standard Deviation for LINCL Category Score of Four Pediatric Neurologists for Each Score Category



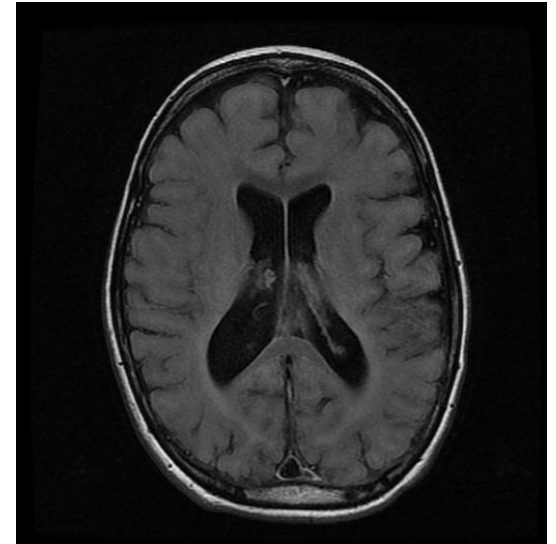
Magnetic Resonance Imaging



LINCL 9

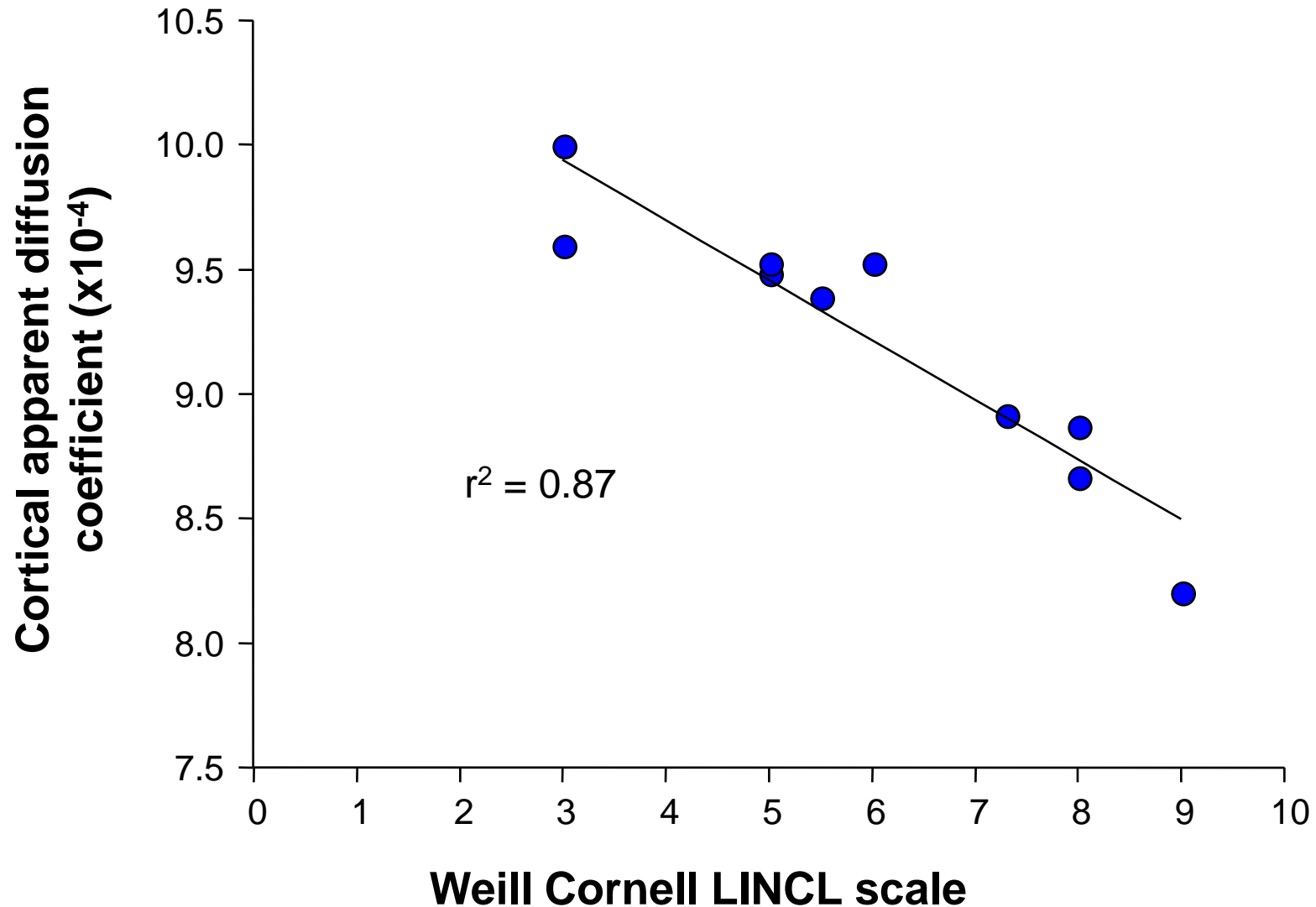


LINCL 6

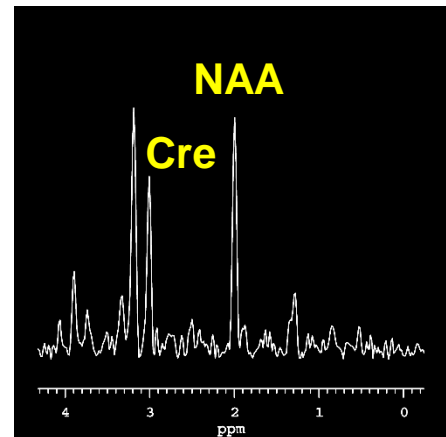
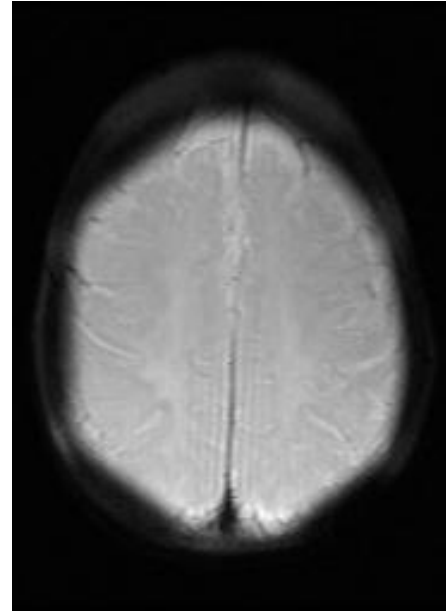
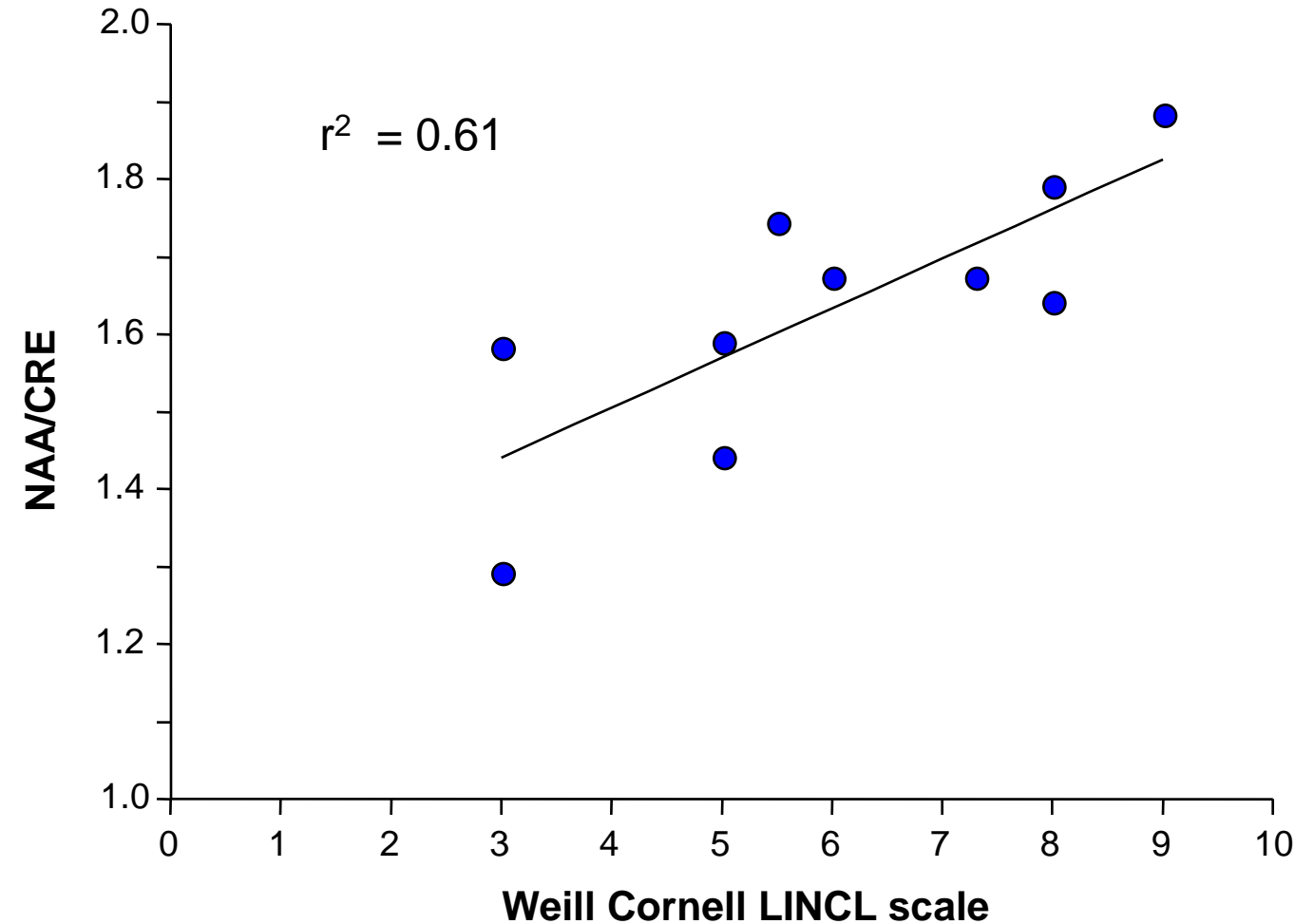


LINCL 3

Cortical Apparent Diffusion Coefficient in LINCL as a Function of Disease Severity

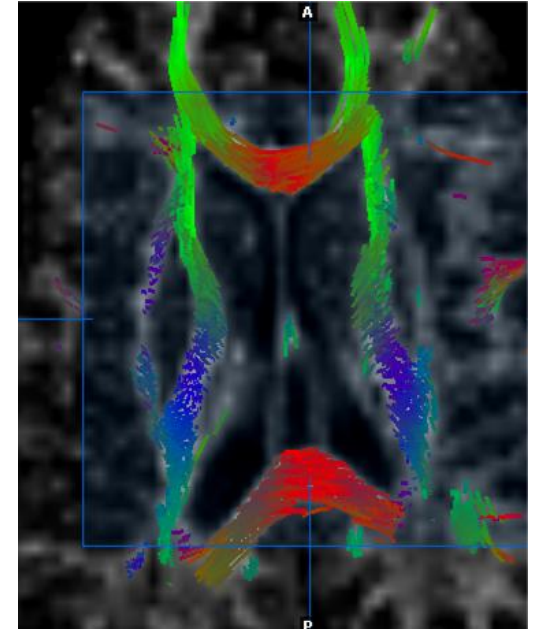
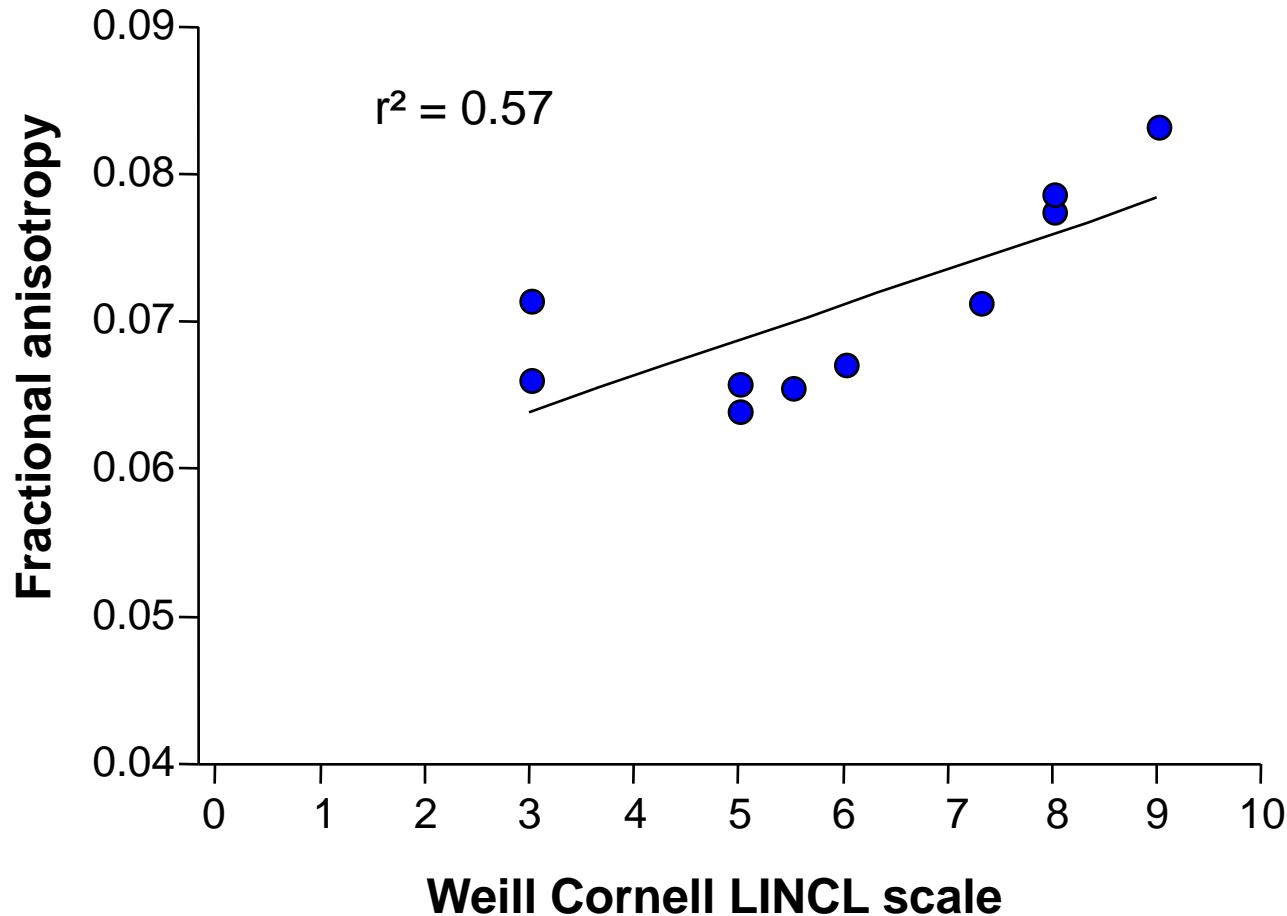


Whole Brain Spectroscopic Analysis



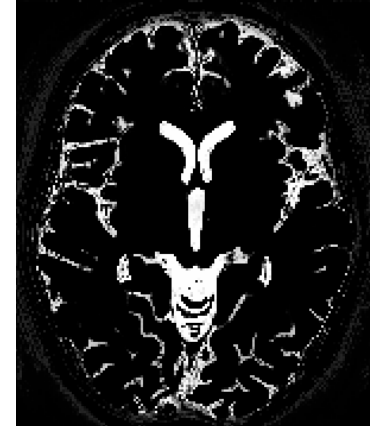
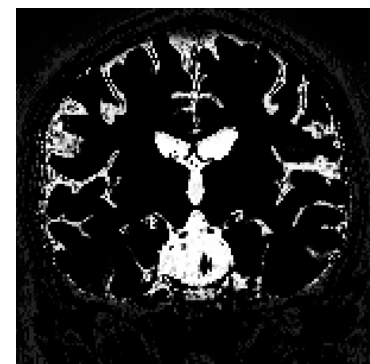
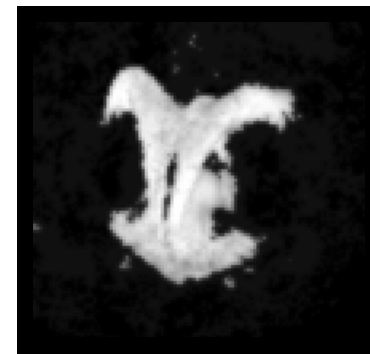
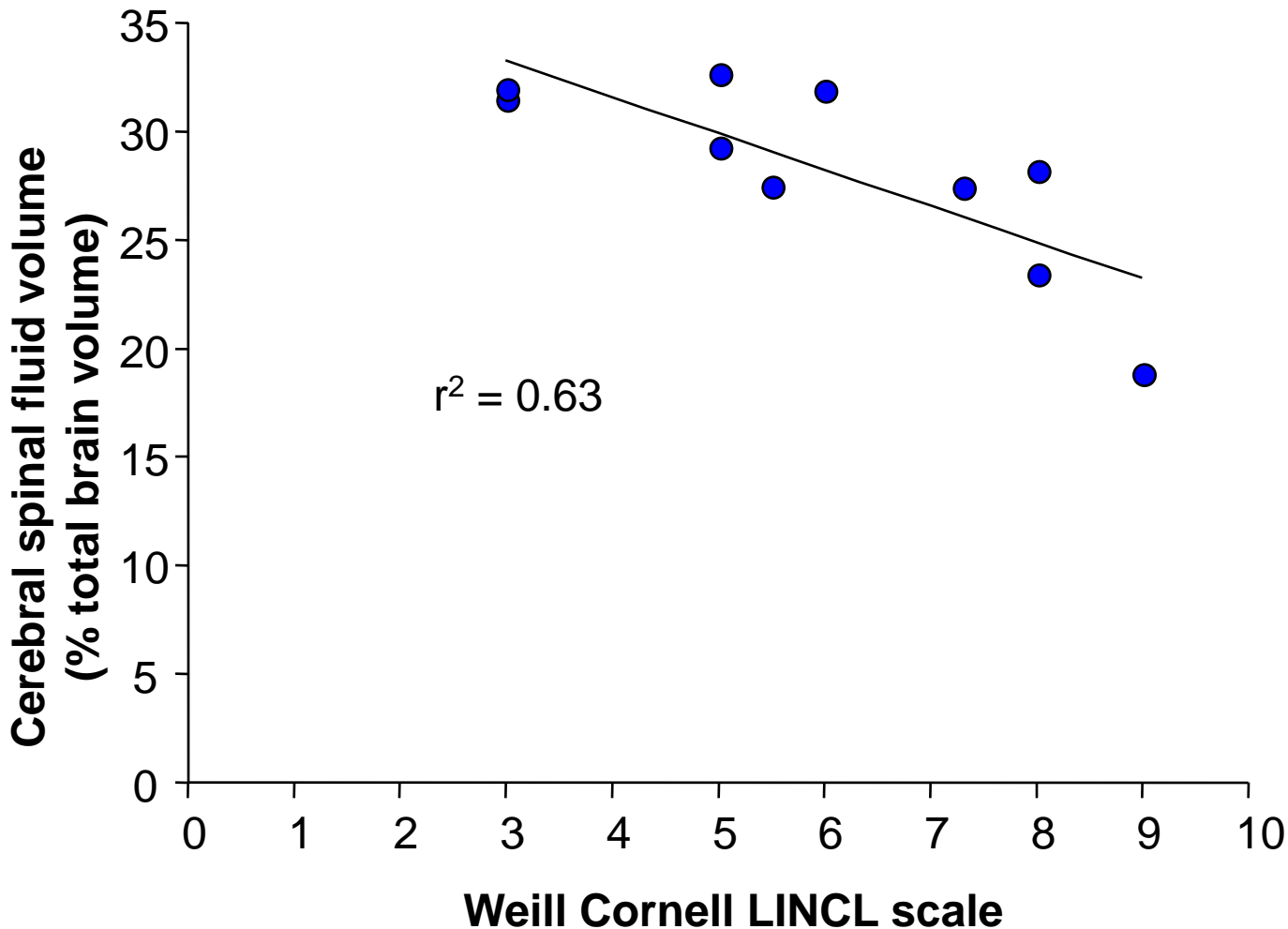
- NAA = N-acetyl-aspartate (marker of neuronal health)
- CRE = creatine (energy metabolic indicator)

Whole Brain Diffusion Tensor Imaging

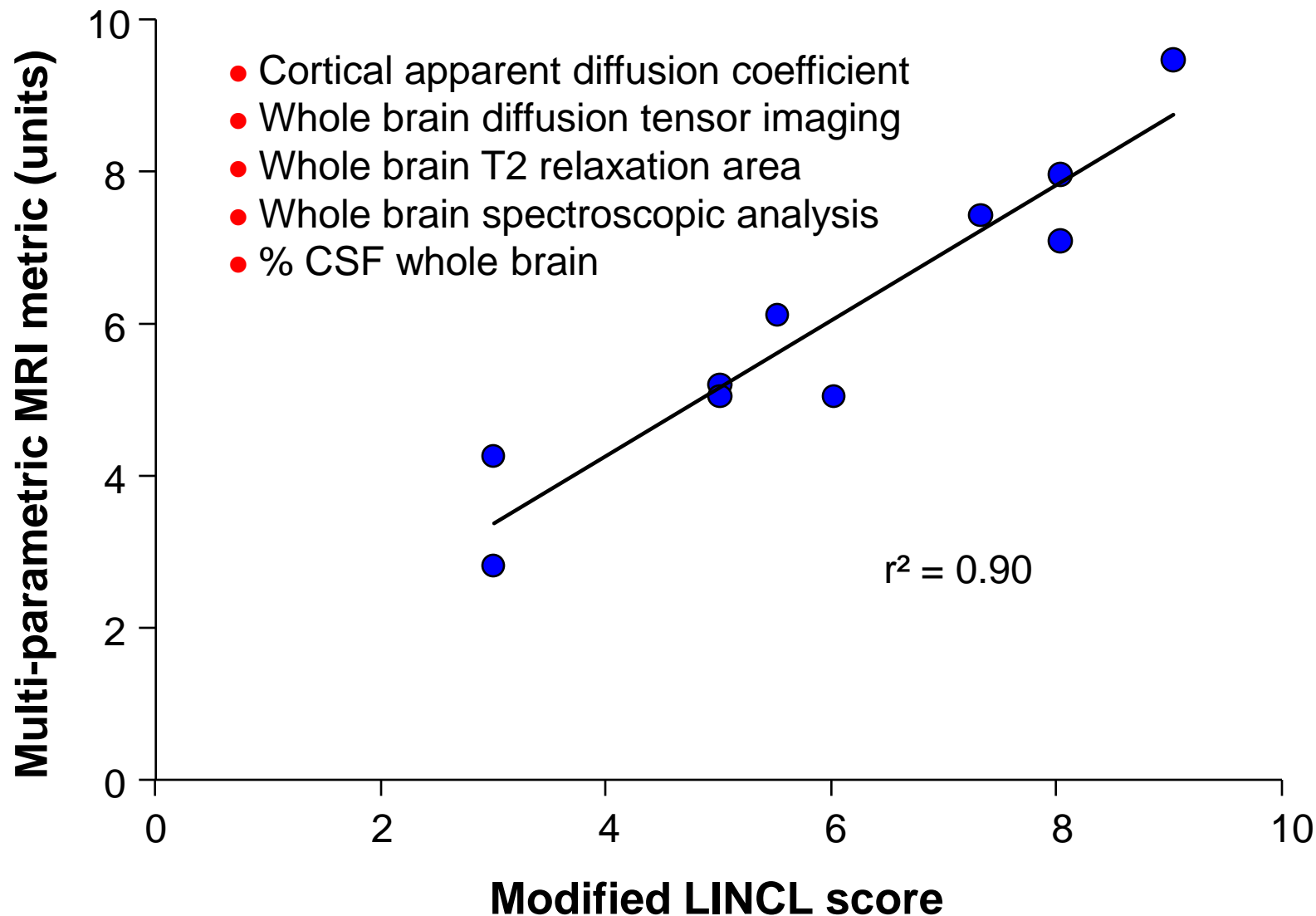


- FA =0 has no preferred diffusion direction (isotropic)
- FA =1 is preferentially aligned in 1 specific direction (anisotropic)

Cerebral Spinal Fluid Volume (% total brain volume)



Multi-parametric MRI Metric to Assess the CNS Manifestations of LINCL



Status of the AAVrh.10hCLN2 LINCL Trial*

Group	Subject	Age	Average LINCL score	Time since therapy (days)
Treated	BDrh-03-C-I	7 yr 3 mo	6.3	283
	BDrh-12-F-B	3 yr 6 mo	9.5	143
	BDrh-13-U-O	4 yr 6 mo	7.0	94
	BDrh-14-T-L	4 yr 8 mo	7.0	59
	BDrh-15-B-G	2 yr 10 mo	9.7	31
Control	BDrh-05-CR	4 yr 9 mo	6.0	—
	BDrh-07-OF	4 yr 11 mo	7.0	—
	BDrh-11-UC	5 yr 3 mo	7.3	—

* 7.5×10^{10} genome copies/site, 12 sites, total dose 9×10^{11} genome copies

