

Gene Therapy for the CNS Manifestations of the Lysosomal Storage Disorders

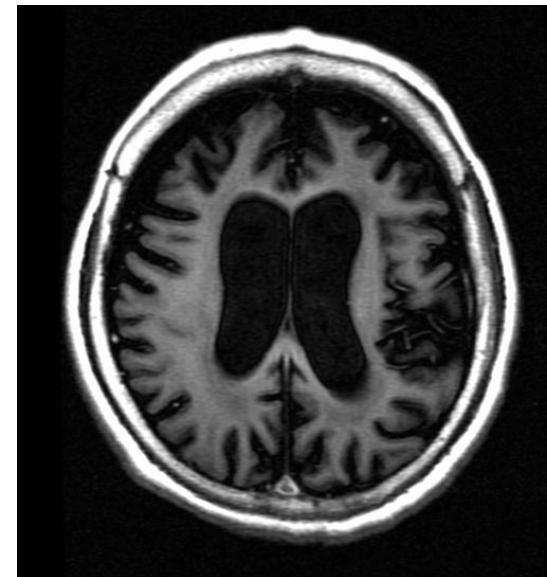
R. Crystal

Weill Cornell Medical College

11-12-10

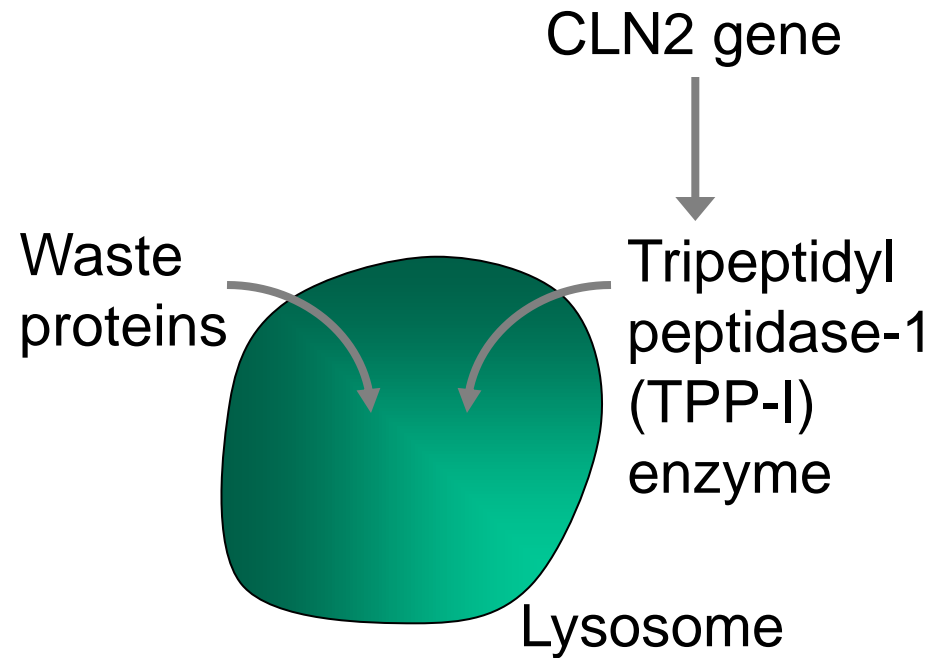
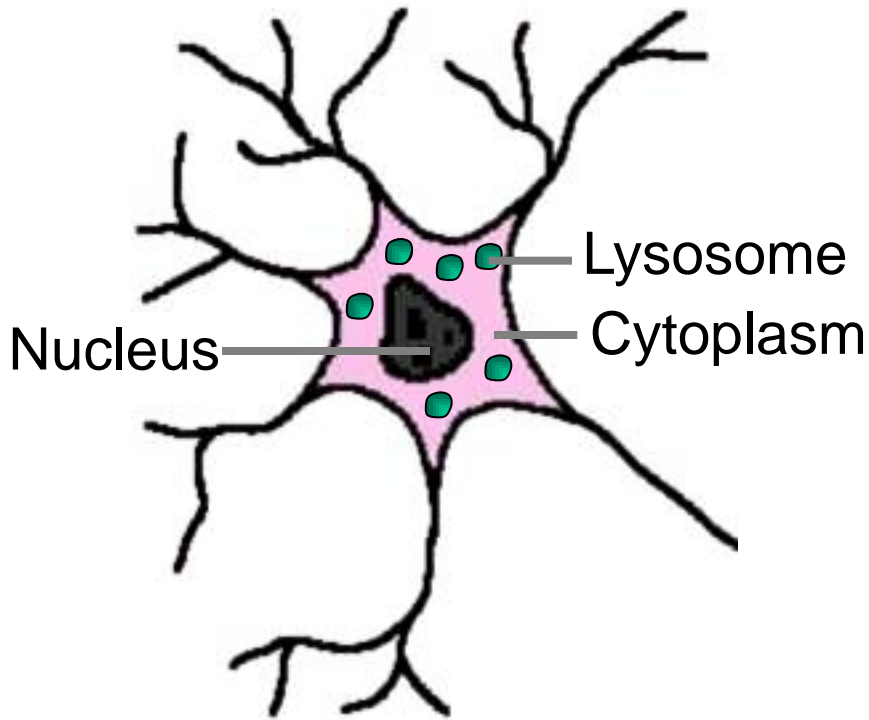
Late Infantile Neuronal Ceroid Lipofuscinoses (LINCL, Batten Disease)

- 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



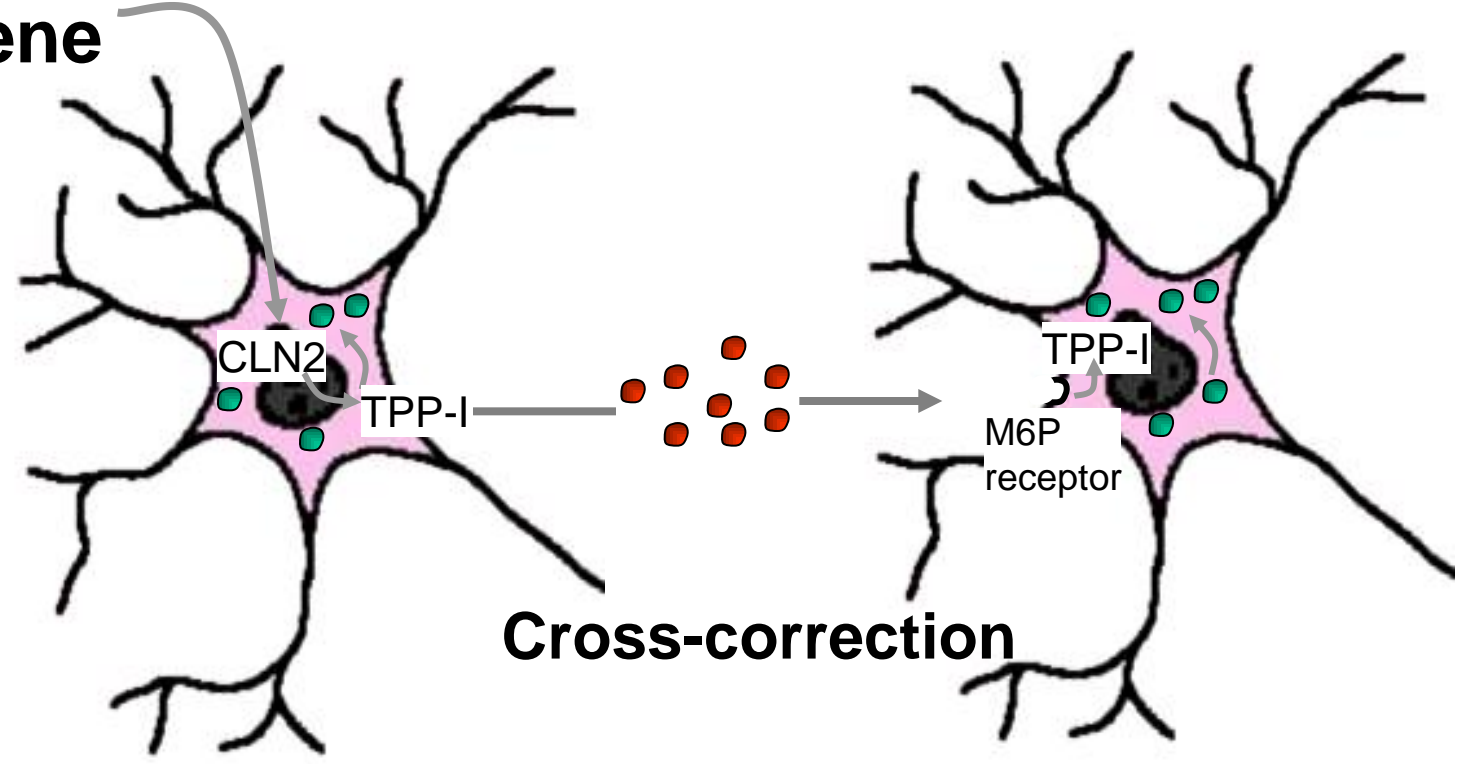
LINCL Is Caused by Mutations in the CLN2 Gene

Neuron

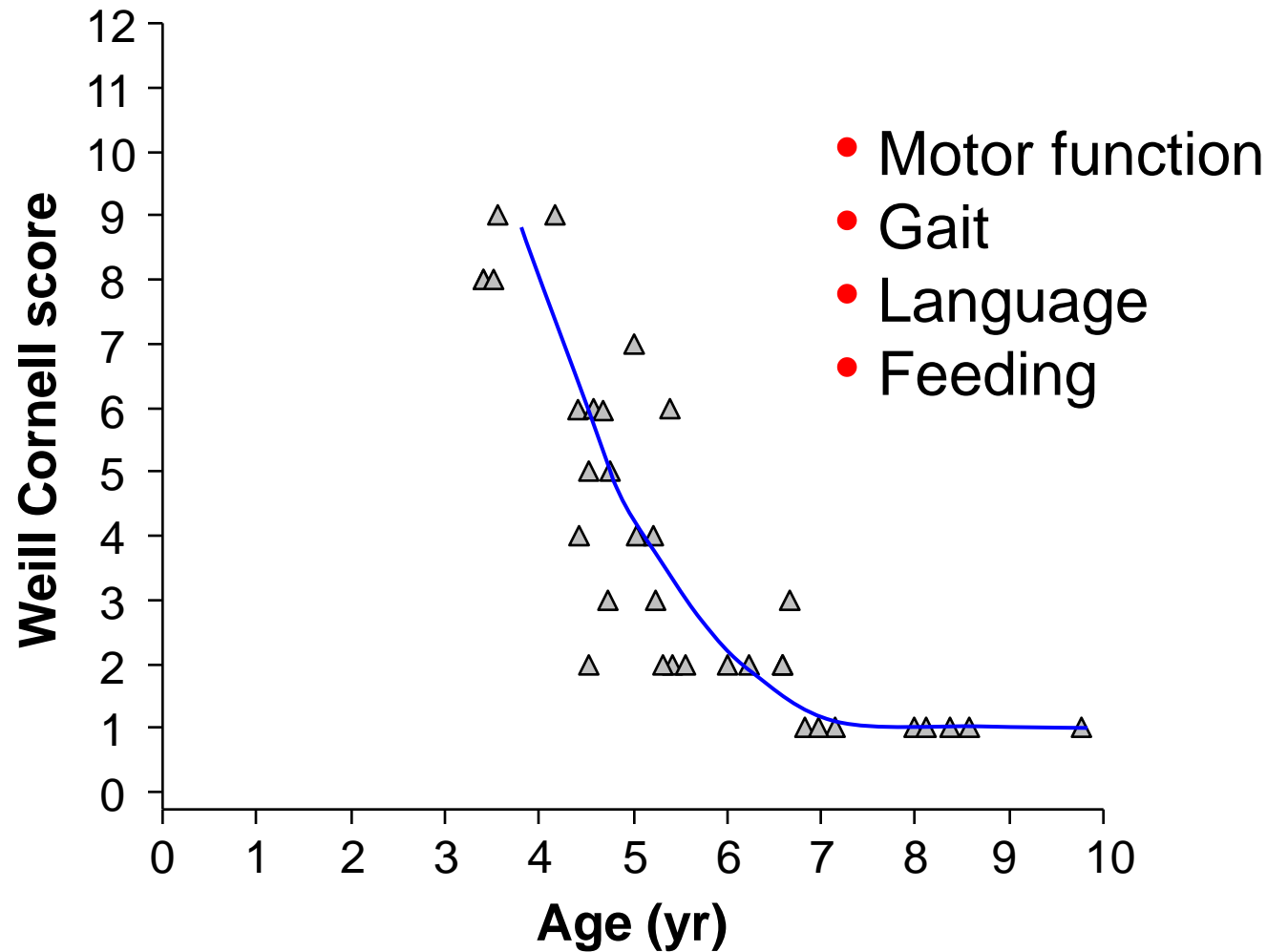


Cross-correction of CNS Cells via the Mannose-6-phosphate Pathway

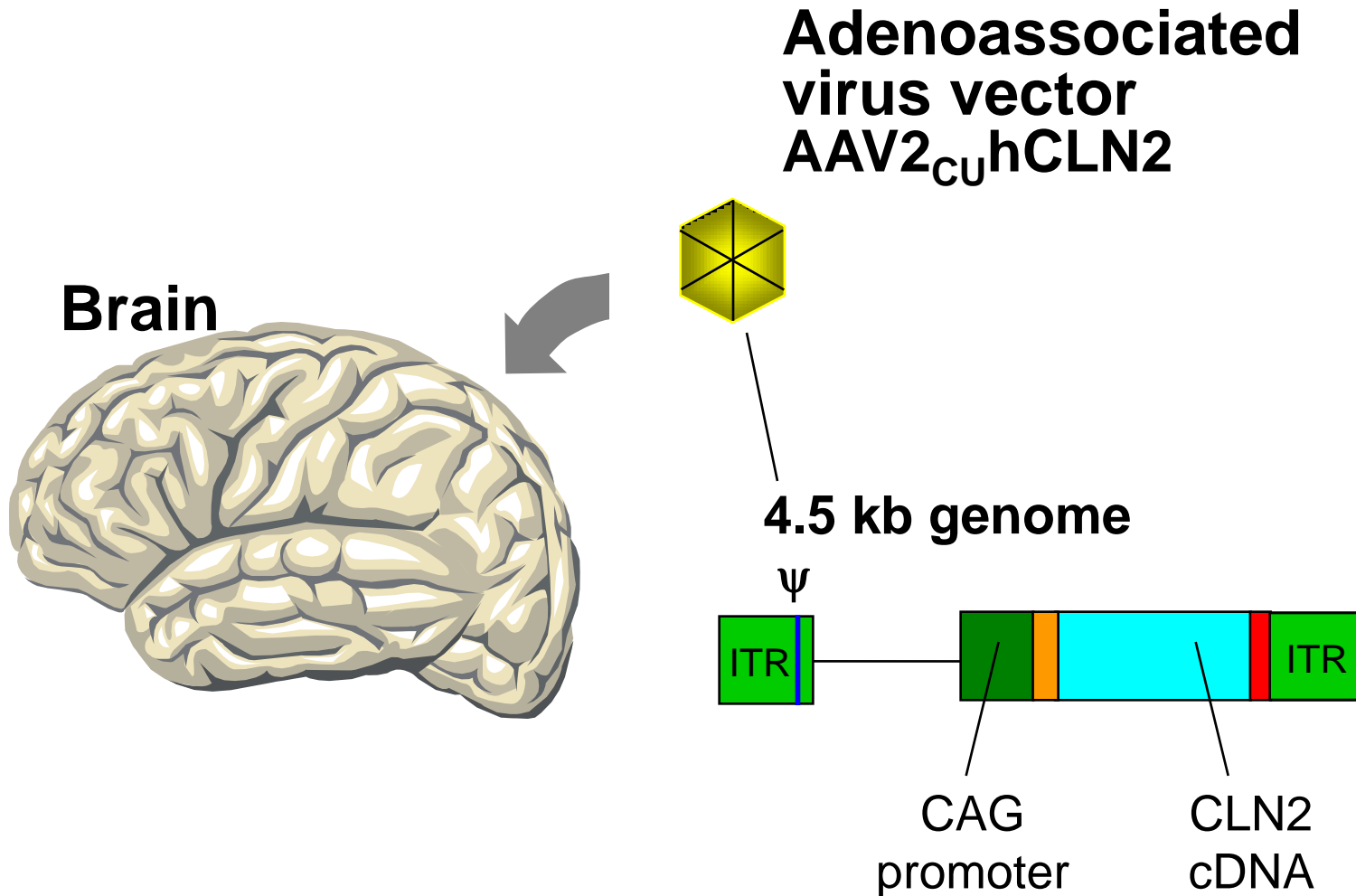
CLN2 gene



LINCL Age-dependent Changes in the Weill Cornell LINCL Score

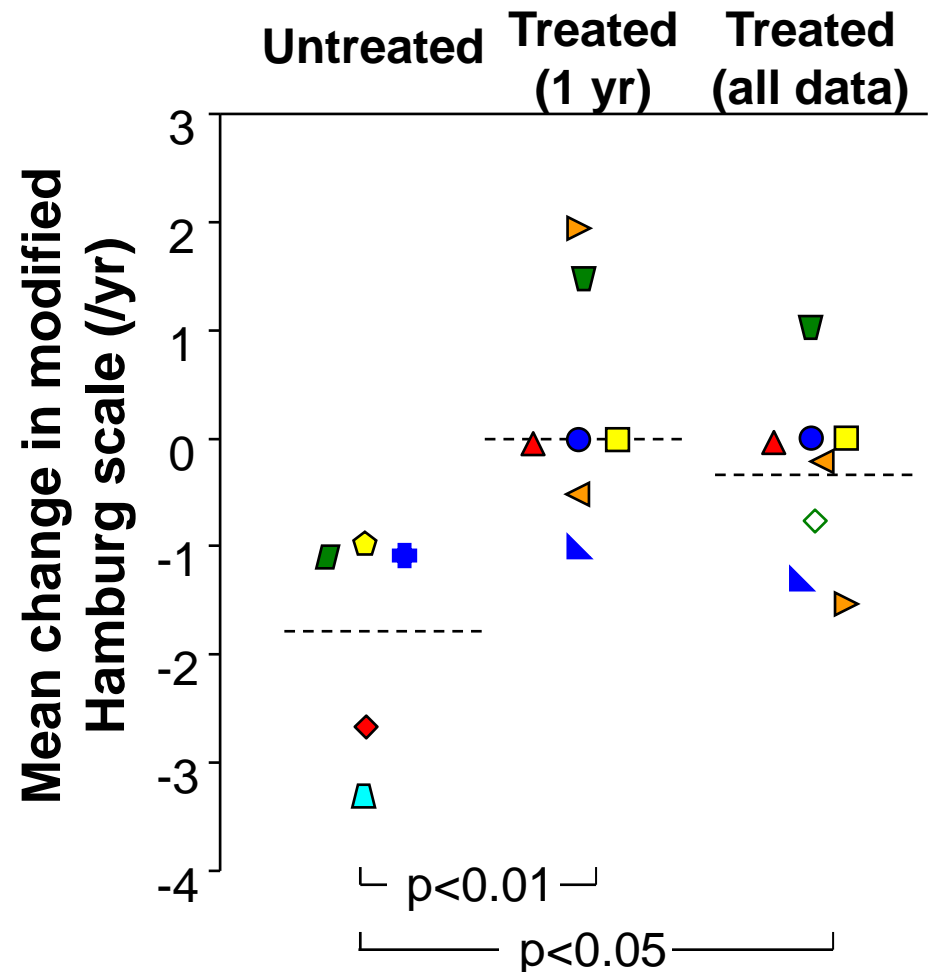


Gene Therapy for LINCL

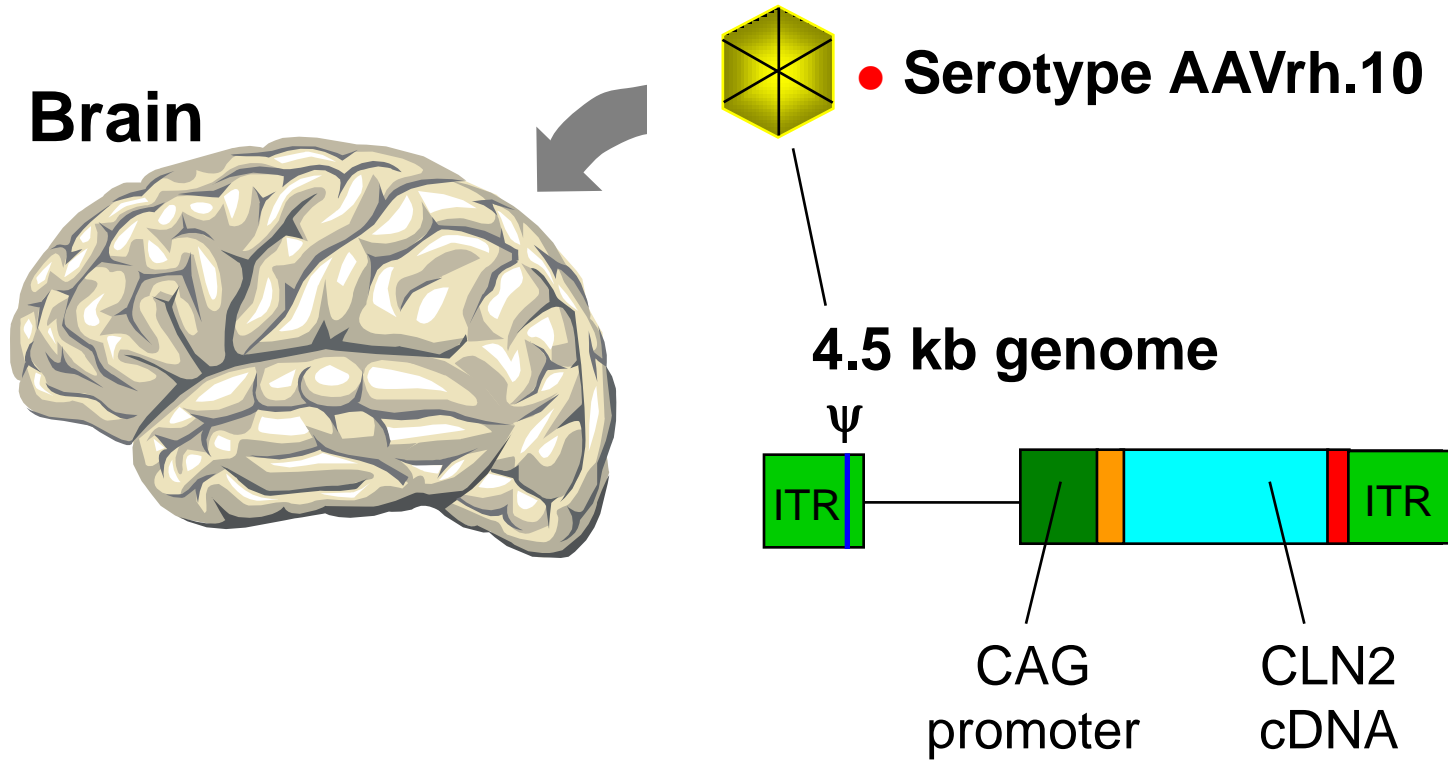


AAV2hCLN2 Gene Therapy Trial for the CNS Manifestations of LINCL

- 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

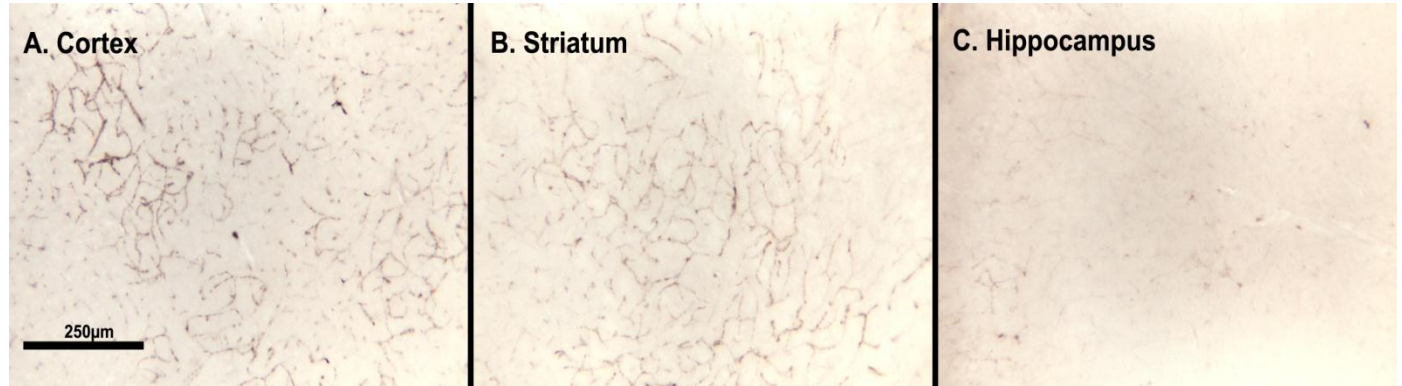


2nd Generation Gene Therapy for LINCL

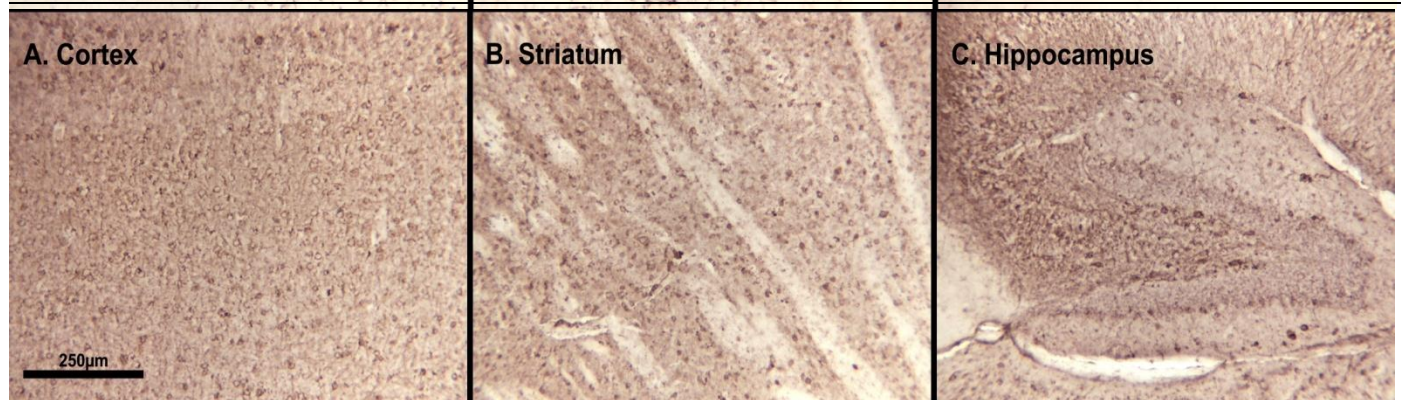


CNS TPP-I Expression Following AAVrh.10hCLN2 Gene Transfer to Day 2 CLN2 ^{-/-} Mice and Assessment at 1 yr

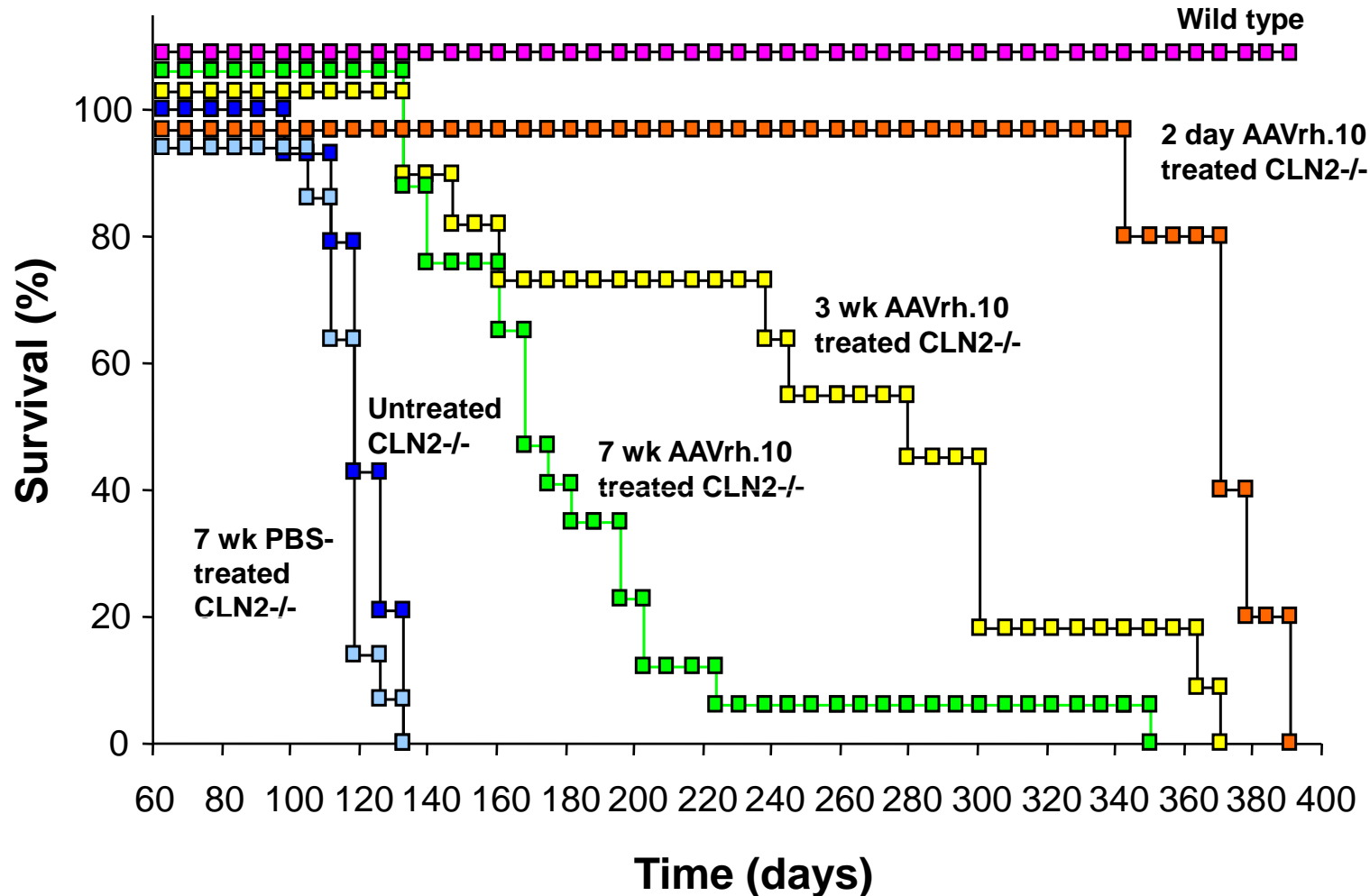
PBS control



AAVrh.10hCLN2



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

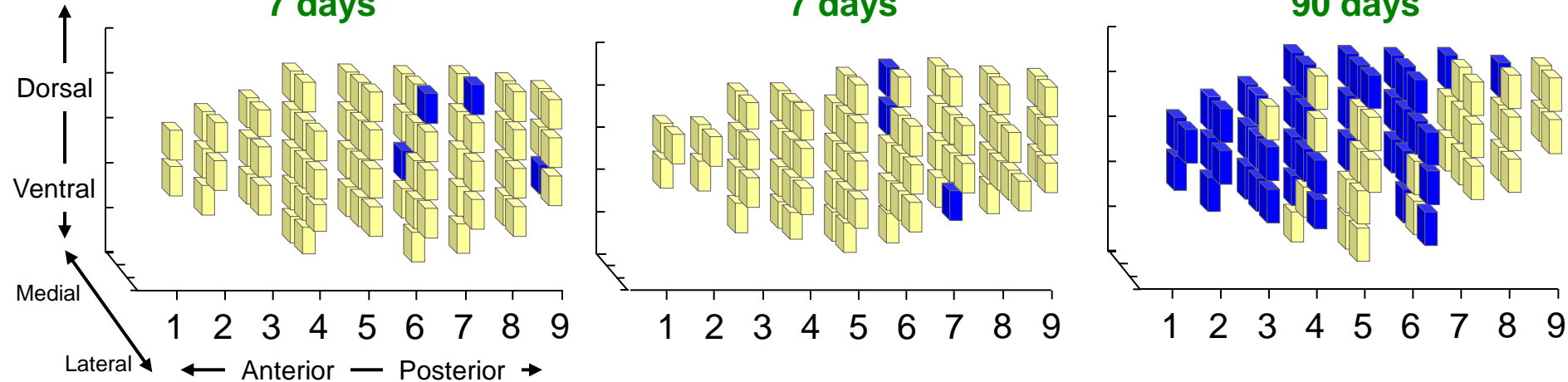




Distribution of TPP-I Activity in the CNS of Non-human Primates Following CNS Administration of AAVrh.10_{CU}hCLN2

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

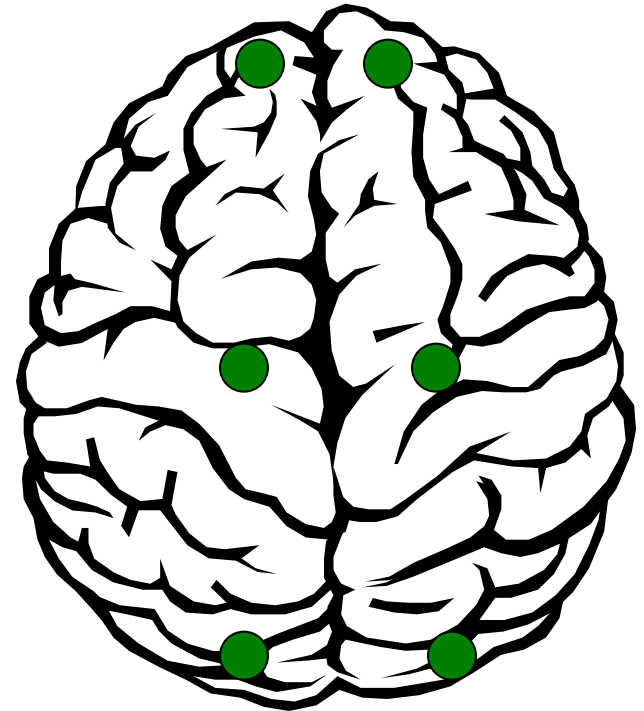
Total dose 1.8×10^{12} gc

Assess efficacy
parameters at 6, 12, 18
months

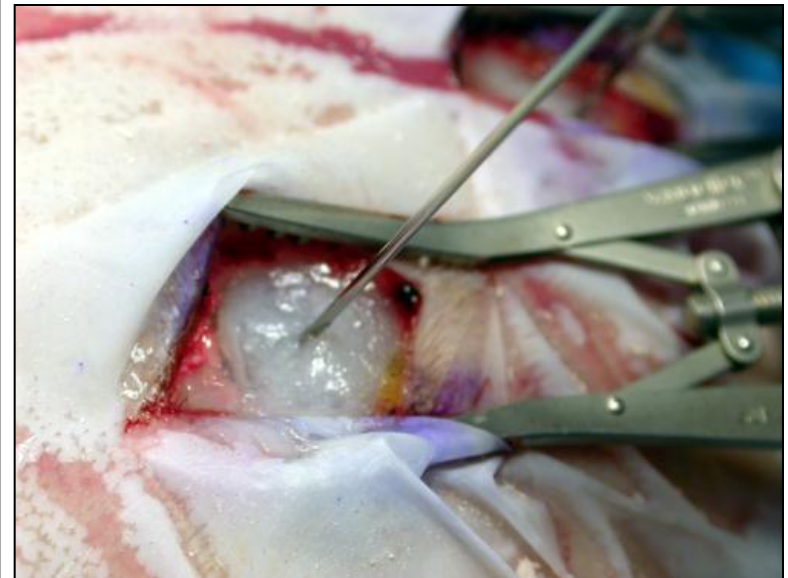
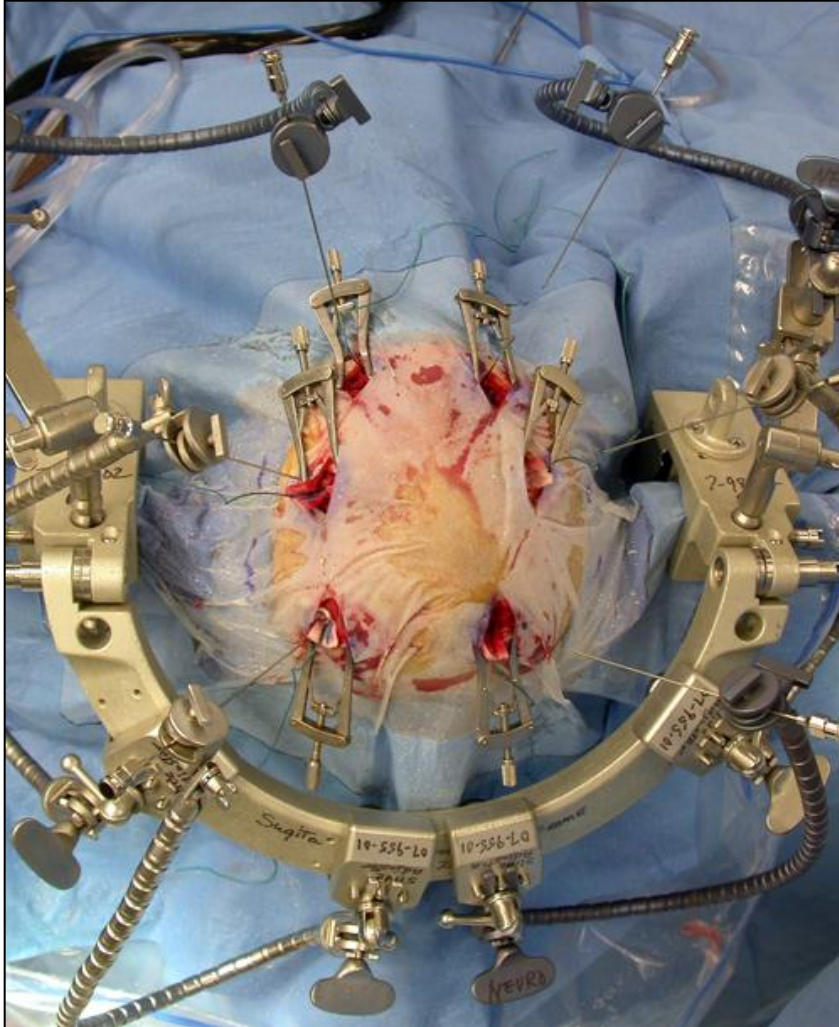
Decision by 4 faculty, representing
3 departments, independent of PI
LINCL mild-moderate

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



Outcome Measures

Primary

- Weill Cornell LINCL score¹
- Quantitative MRI
 - % Ventricular volume
 - % Grey matter volume
 - Cortical apparent diffusion coefficient

Secondary

- Mullen score
- CHQ Quality of Life questionnaire

¹ Video, blinded assessment by 3 independent pediatric neurologists

Status of the AAVrh.10 Trial

- 1st child treated
- 7 yr 3 months
- CLN2 genotype G3556C homozygote
- Weill Cornell LINCL score 6.3
- 7.5×10^{10} genome copies/site, 12 sites, total dose 9×10^{11} genome copies
- No serious adverse events (10 wk post-rx)

Critical Path in Developing Therapies for Rare Disorders of the CNS

PHENOTYPE

PHENOTYPE

PHENOTYPE

Critical Need – To Develop Quantitative Phenotypes to Assess Efficacy

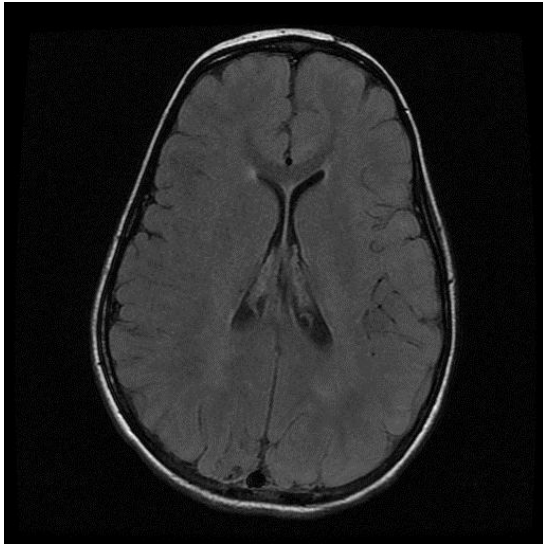
Clinical scales

- Quantitative clinical scales are, at best, integrated measures of overall CNS function
- To limit observer variability, use 3 blinded observers to assess videotapes

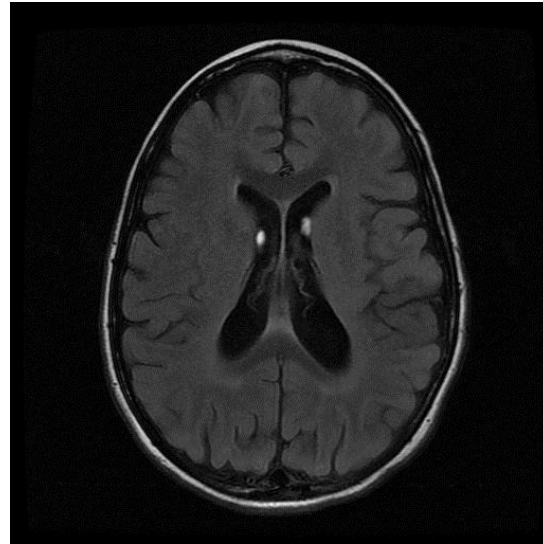
Quantitative MRI

- Evolving methodology that requires more data and verification

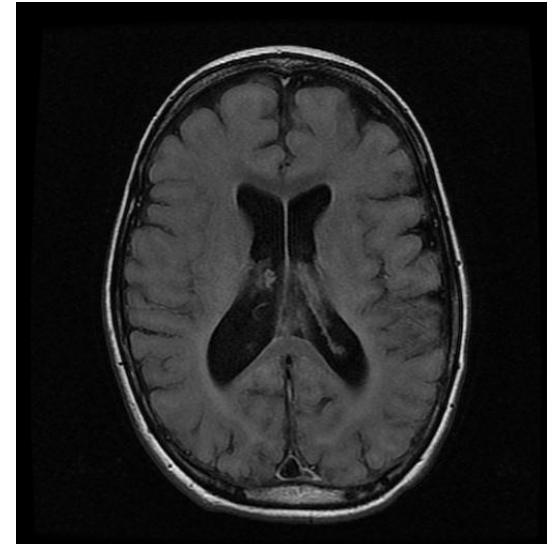
Magnetic Resonance Imaging



LINCL 9

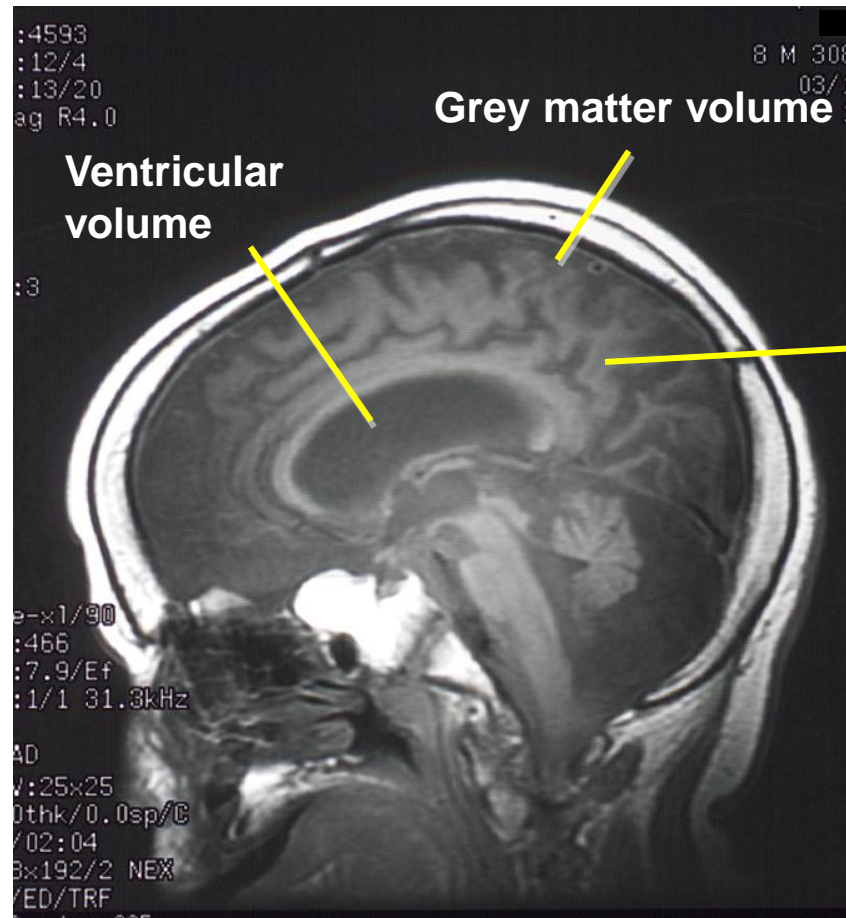


LINCL 6



LINCL 3

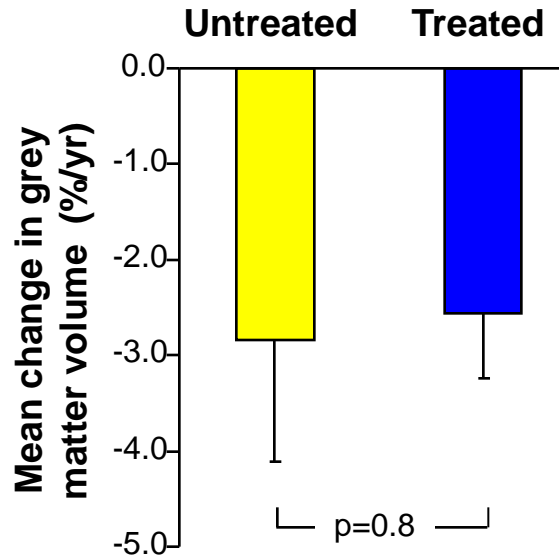
Quantitative MRI Parameters



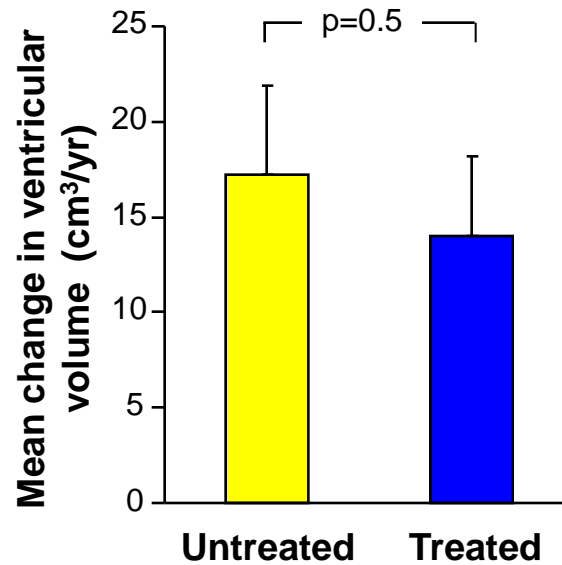
Cortical
apparent
diffusion
coefficient

2^o Variables – Quantitative MRI

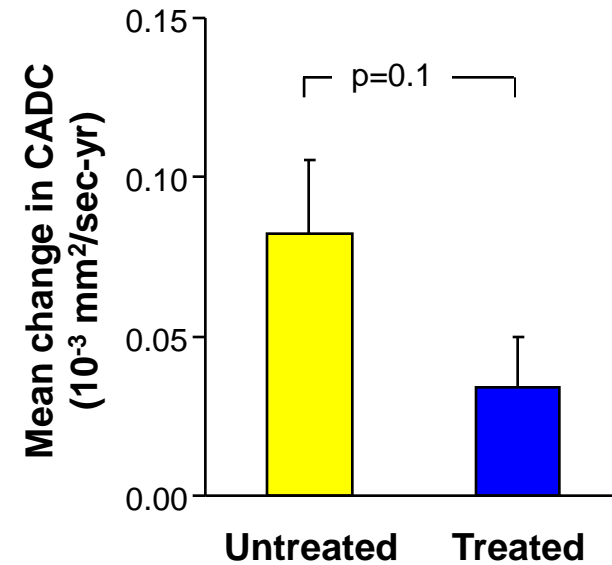
A.



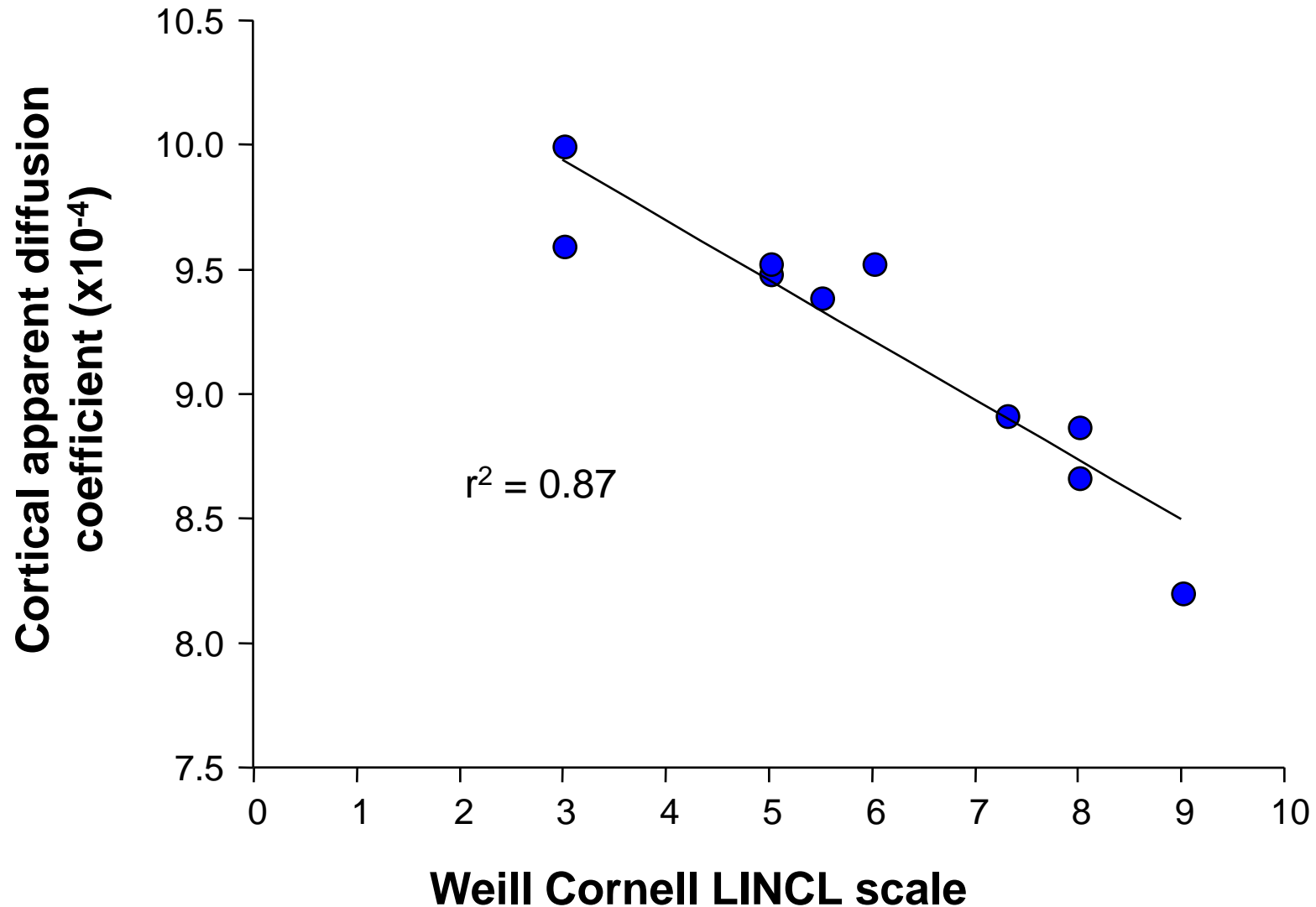
B.



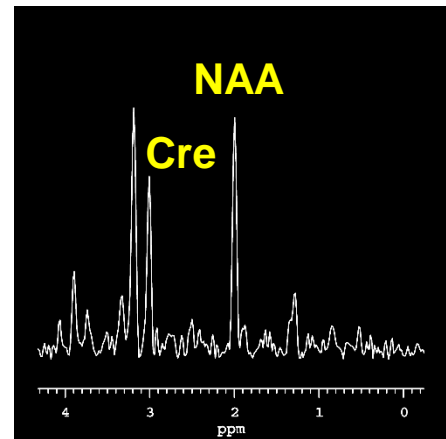
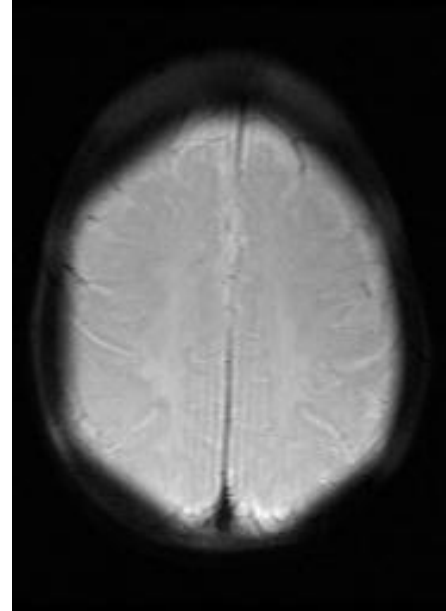
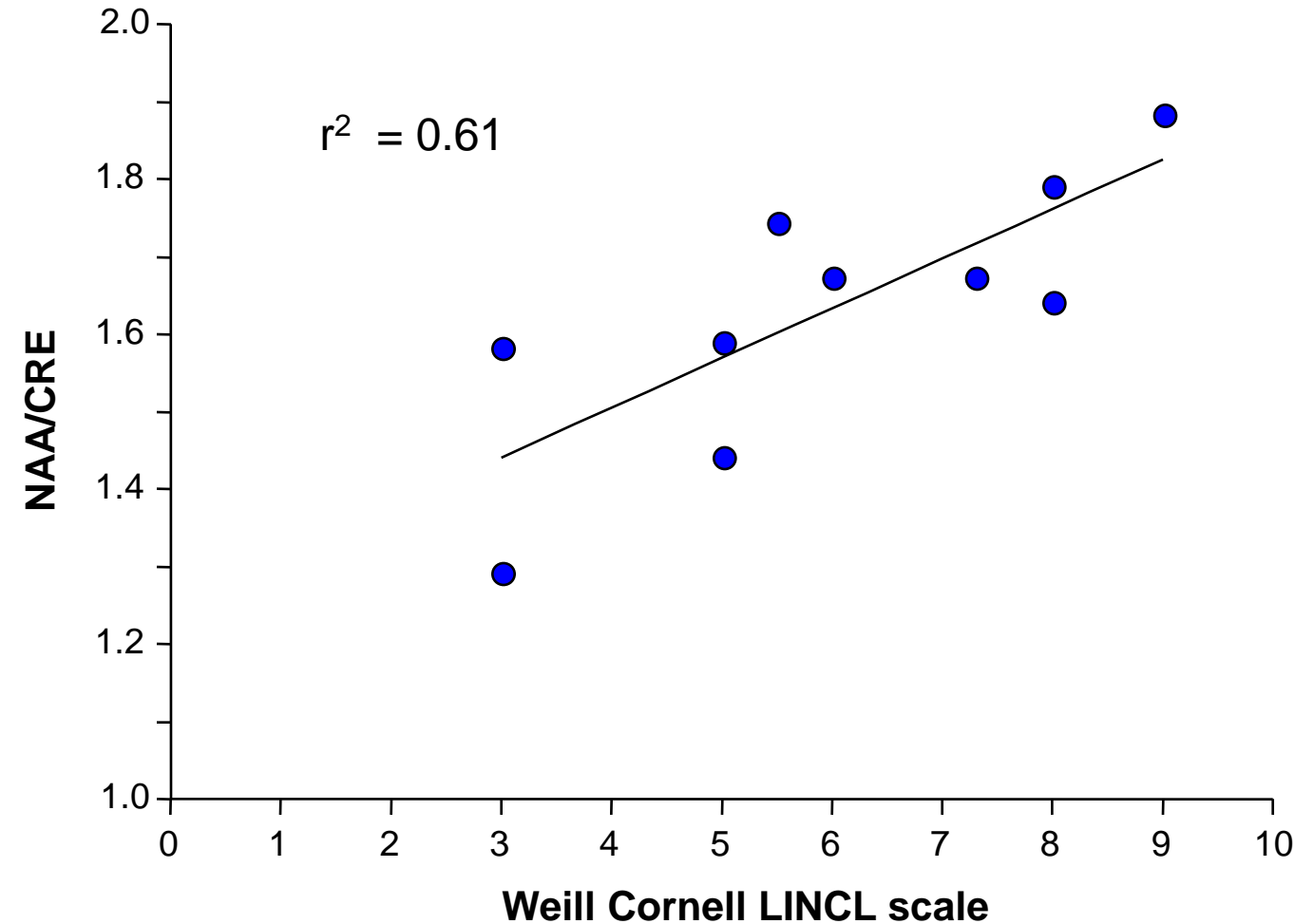
C.



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

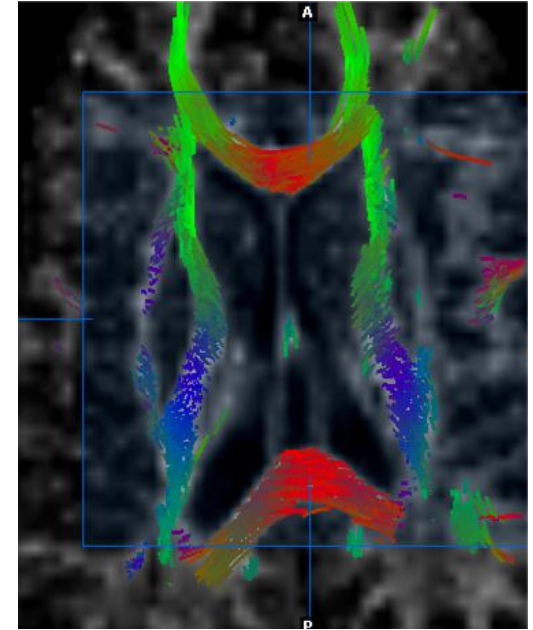
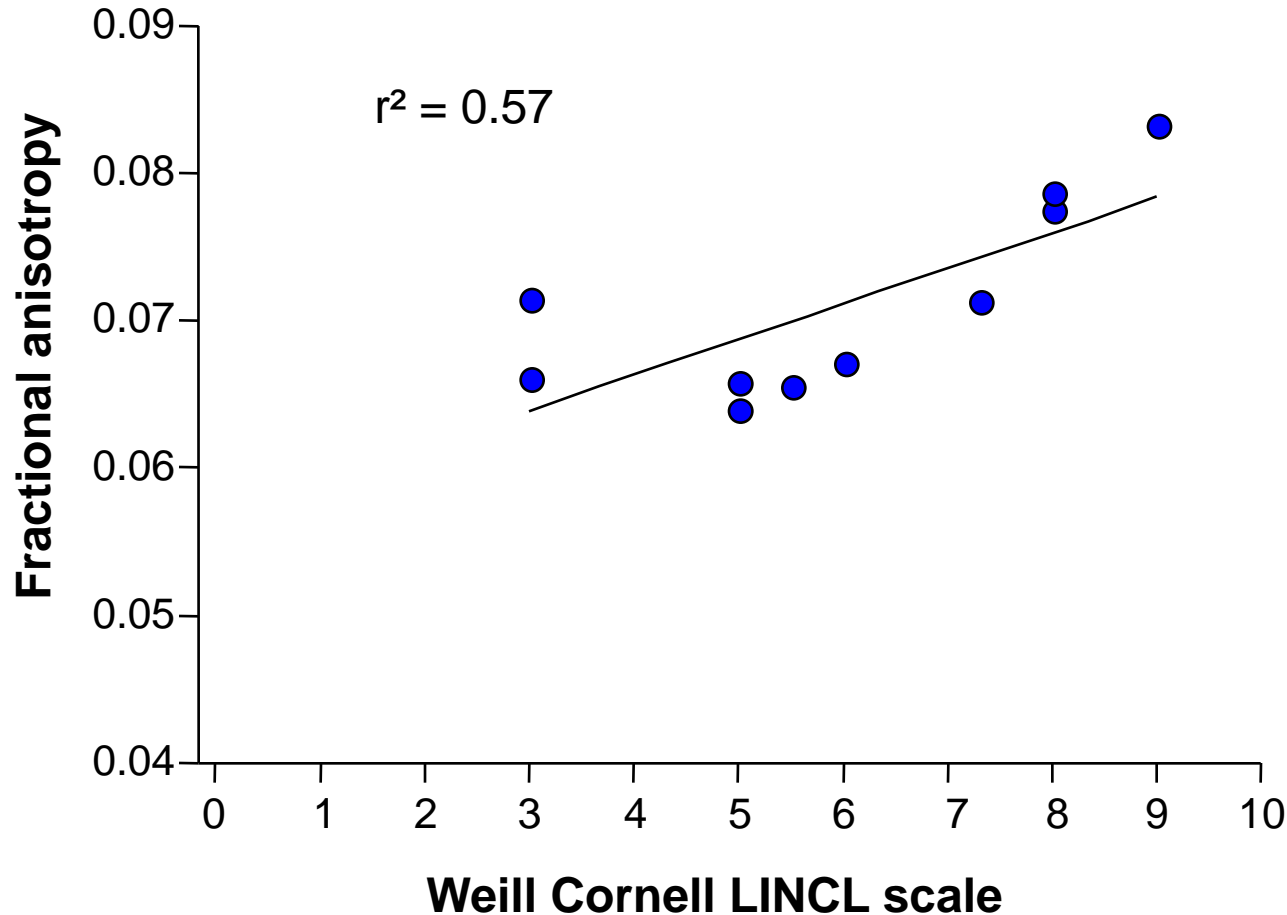


Whole Brain Spectroscopic Analysis



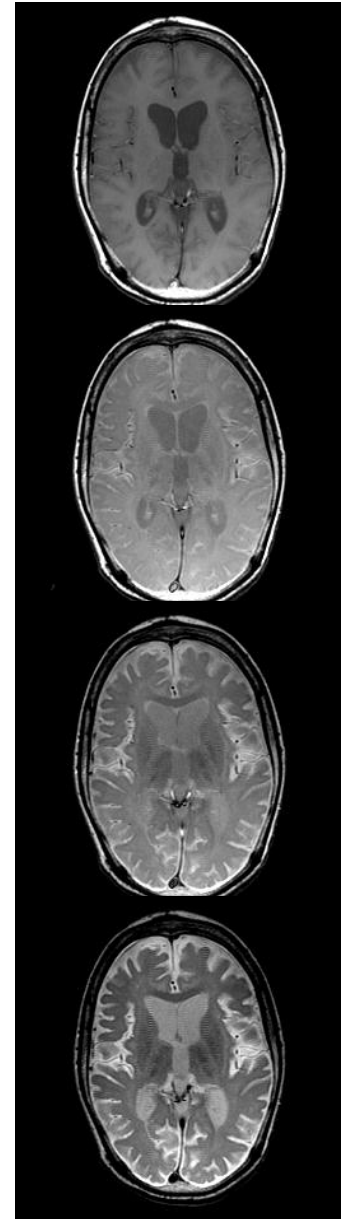
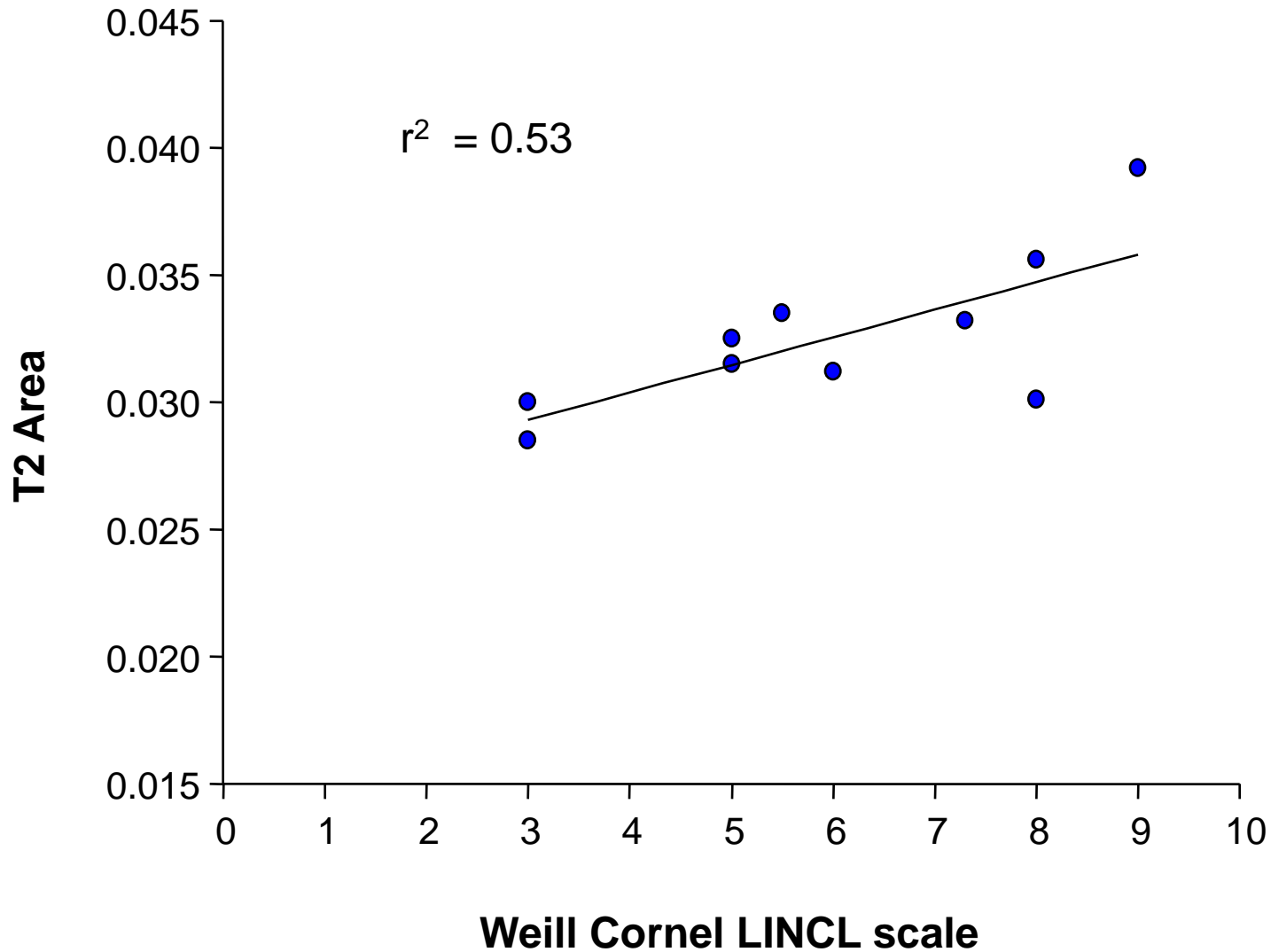
- NAA = N-acetyl-aspartate (marker of neuronal health)
- CRE = creatine (energy metabolic indicator)

Whole Brain Diffusion Tensor Imaging

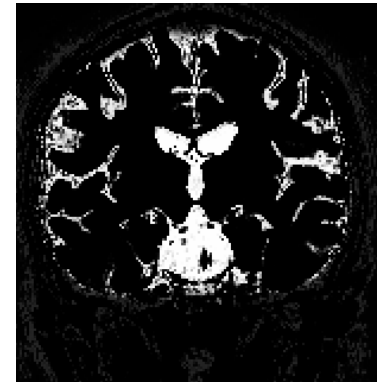
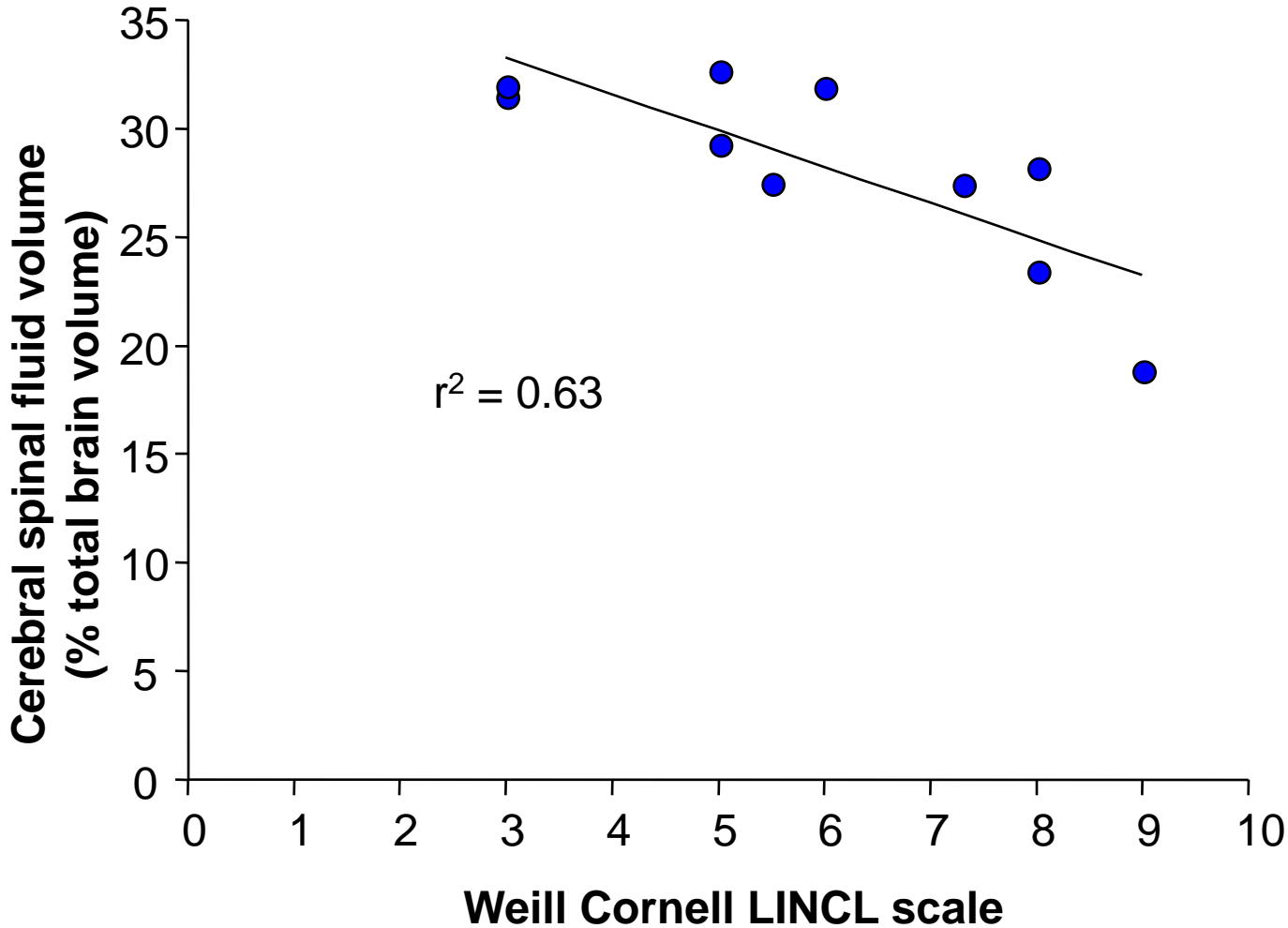


- FA =0 has no preferred diffusion direction (isotropic)
- FA =1 is preferentially aligned in 1 specific direction (anisotropic)

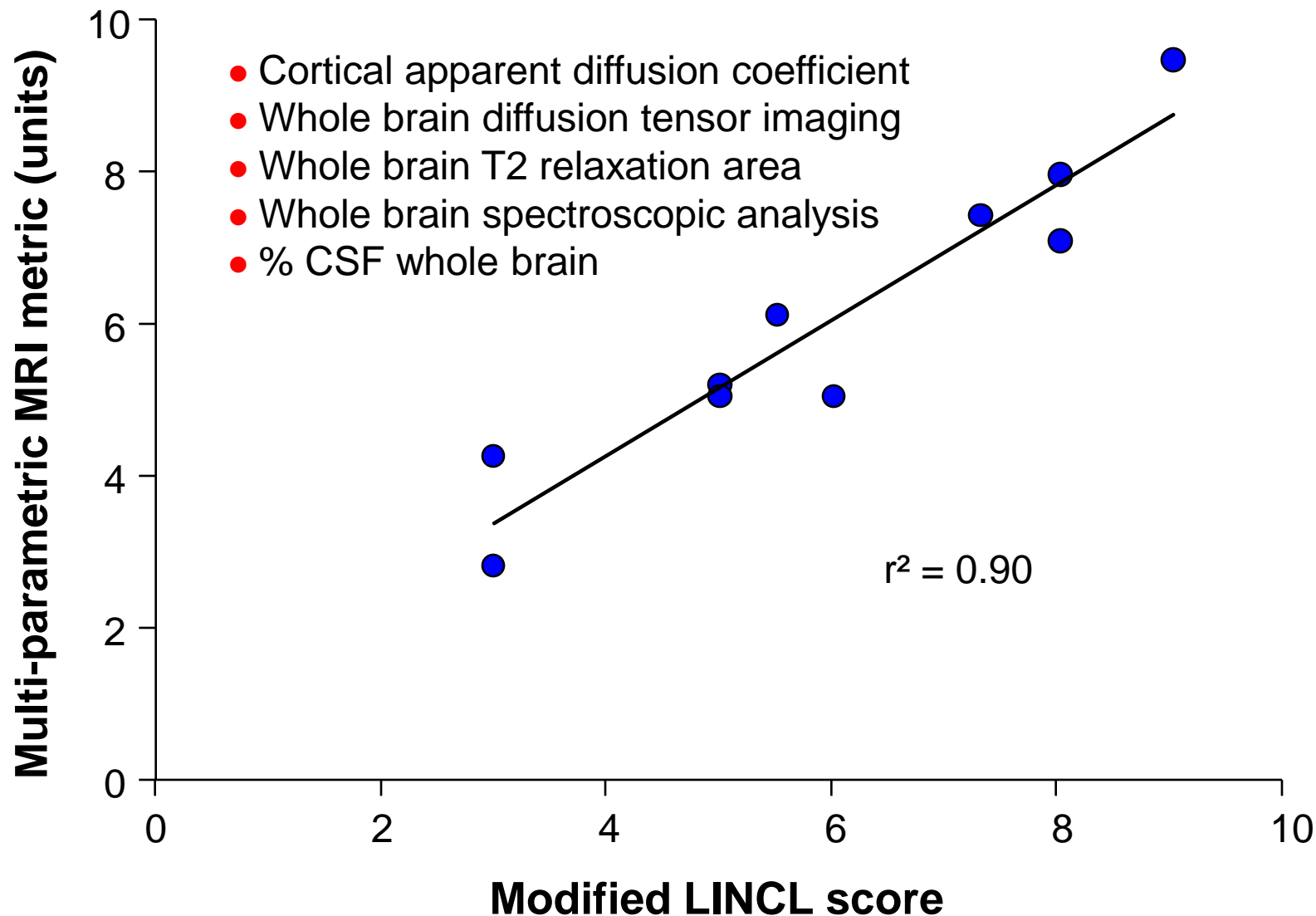
Whole Brain T₂ Relaxation Area



Cerebral Spinal Fluid Volume (% total brain volume)



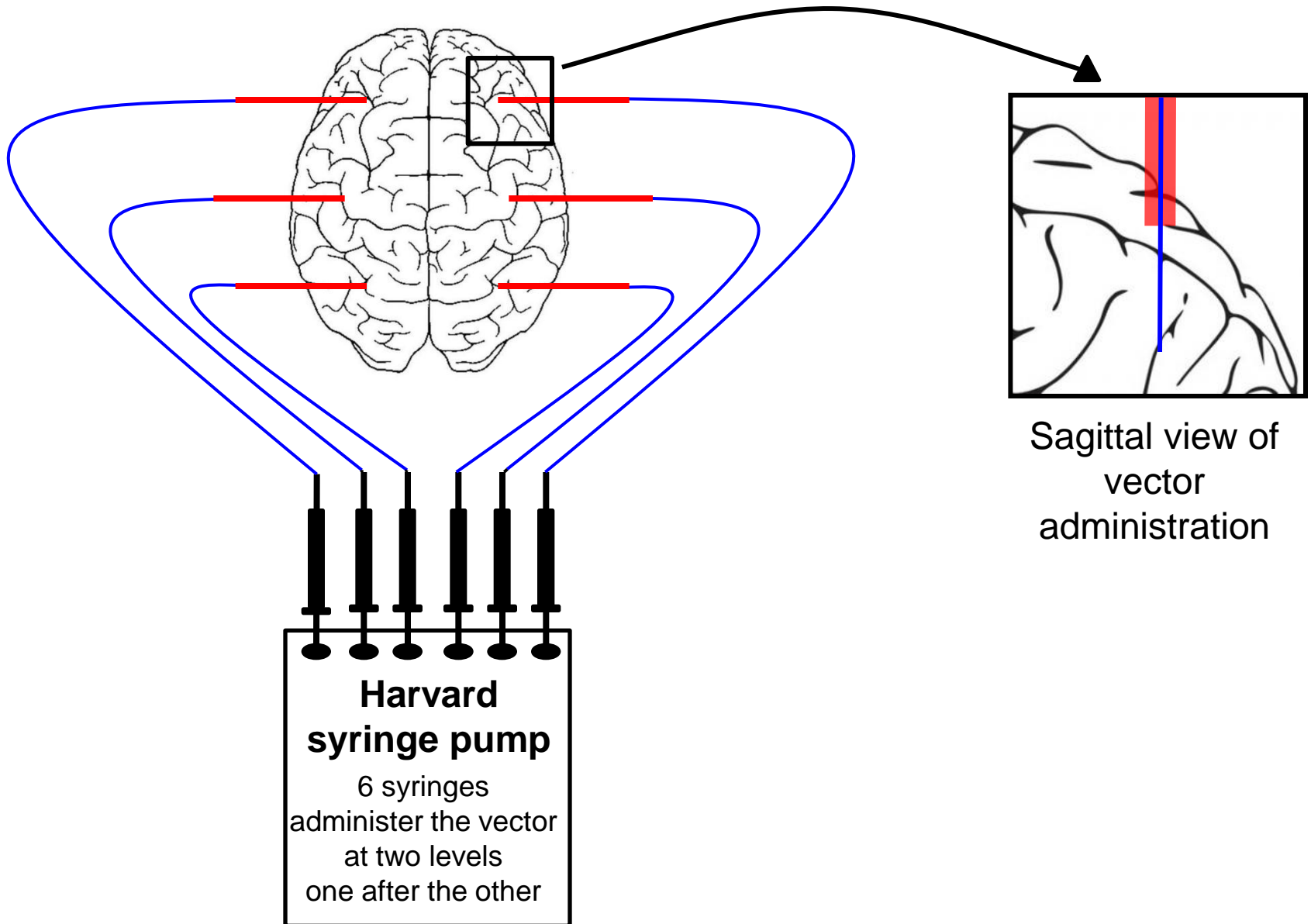
Multi-parametric MRI Metric to Assess the CNS Manifestations of LINCL



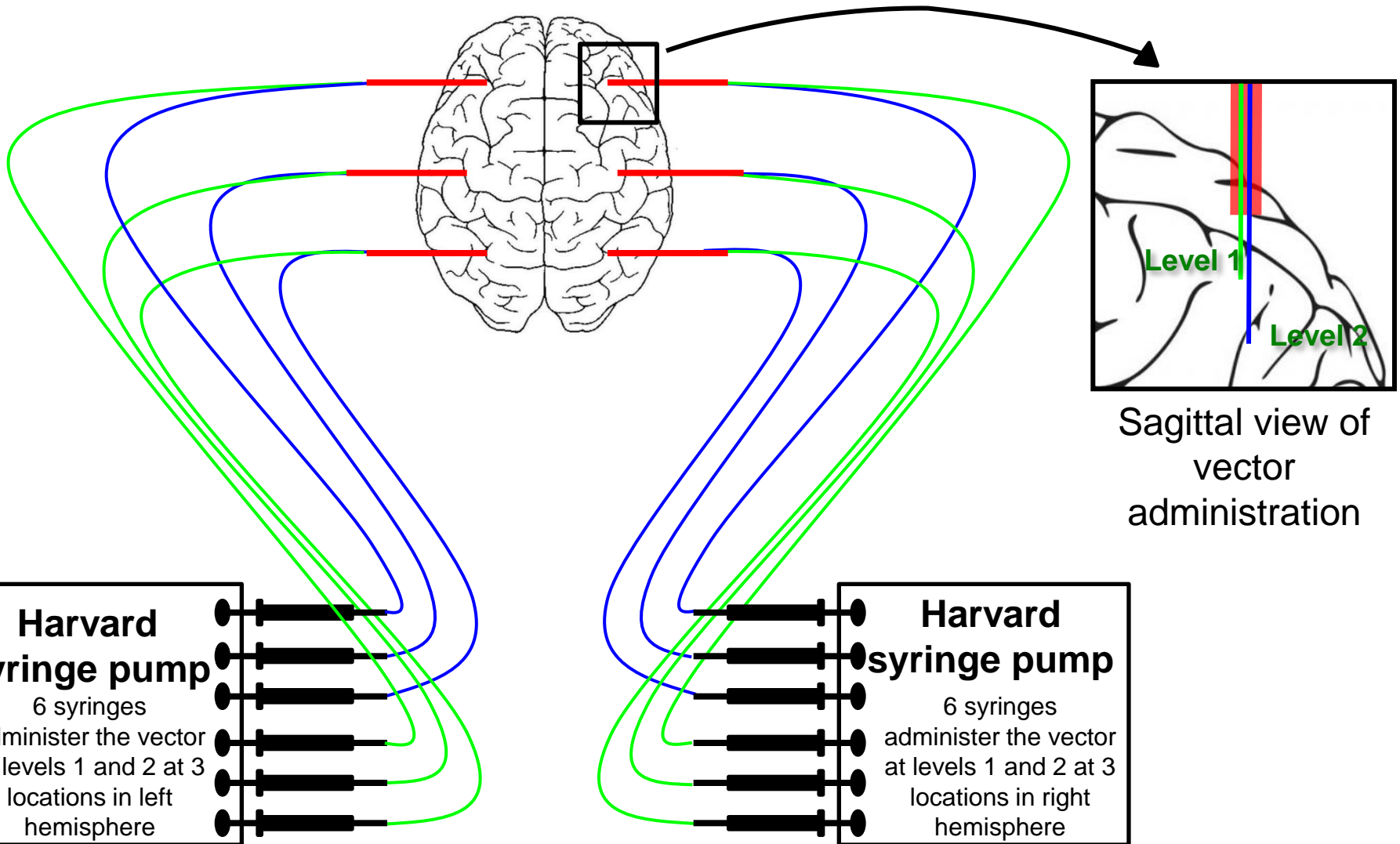
Question

- Can the administration procedure be improved?

Current Administration Protocol



Parallel Administration



Genetic Modification of the CNS

