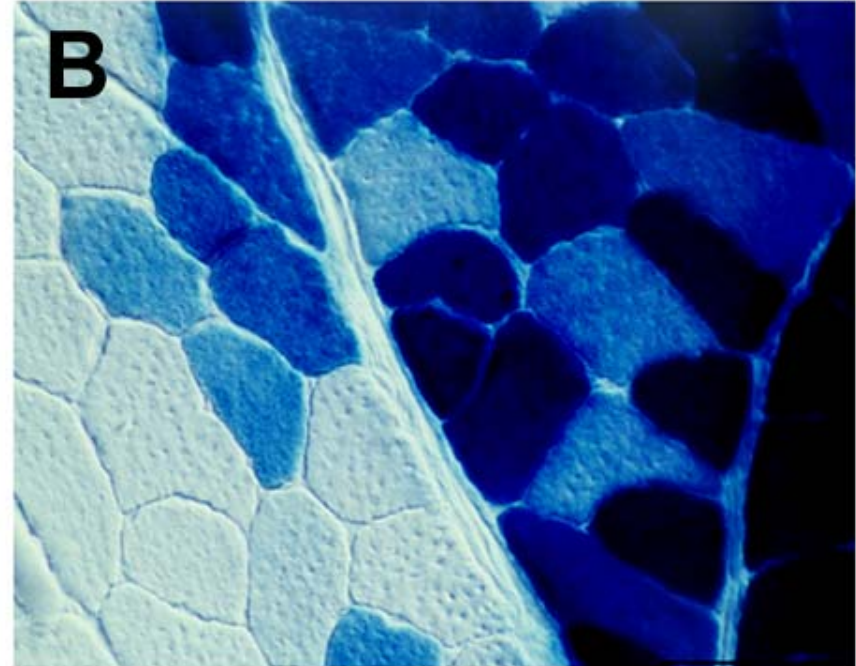
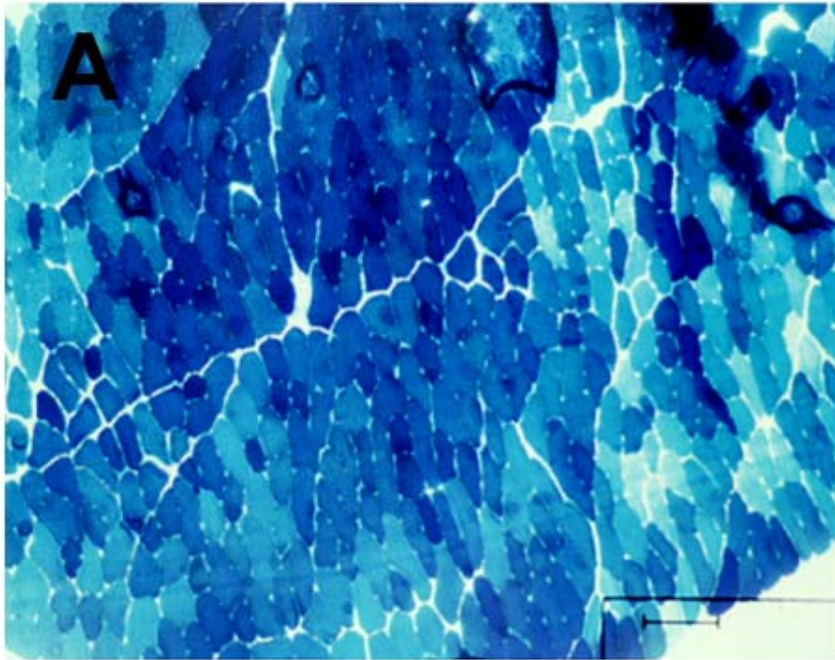


AAV delivery to muscle leads to permanent gene transfer



Kessler, et al, PNAS '96

Why Gene Therapy?

- Single gene defect with well defined pathophysiology
- Ability to provide single dose therapeutic with a systemic effect in the musculature
- Endogenous source of gene product
 - Avoidance of circulating Ab / CTL
 - Differential fiber type processing
- Gene transfer to motor neurons if involved

Clinical Development Time Line

POC: Animal model

Pre-clinical Toxicology

GMP Production

IND and Phase I



Years

Preclinical and platform studies

- Biodistribution
- Dose-related direct vector toxicity
- Mutagenesis risk
- Potential for germ line transmission

Role of Toxicology Studies in Gene Therapy Clinical Trials: Compliance

- **GLP toxicology studies facilitated by early definition of a clinical plan**
 - availability of test article
 - predictive preclinical models
 - study designs to address informative questions
- Concurrent studies of test article stability
- QA/QC with independent reporting

Phase I/II Trial of rAAV1-hAAT Vector for Treatment of Alpha-1 Antitrypsin Deficiency

- Safety of IM administration of rAAV1-CB-hAAT in AAT-deficient adults
 - General Safety
 - Biodistribution
 - Immune response
- Dose-finding pharmacokinetics of rAAV1-CB-hAAT
 - Comparison to ongoing rAAV2-CB-hAAT

Phase I/II rAAV-CB-hAAT

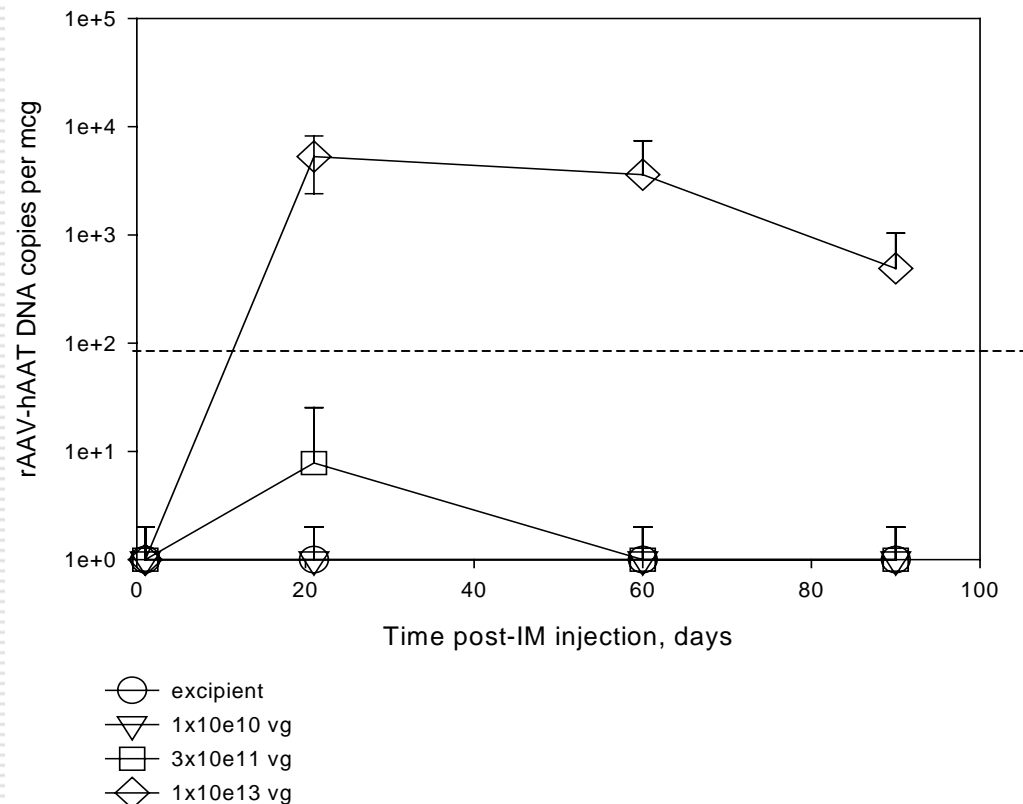
NIH: HL69877

- Single site (UF)
- Open label
- Single dose
- Dose escalation between subjects
- Intramuscular administration with ultrasound guidance to avoid vascular delivery
- N = 12 (4 cohorts of 3 subjects)

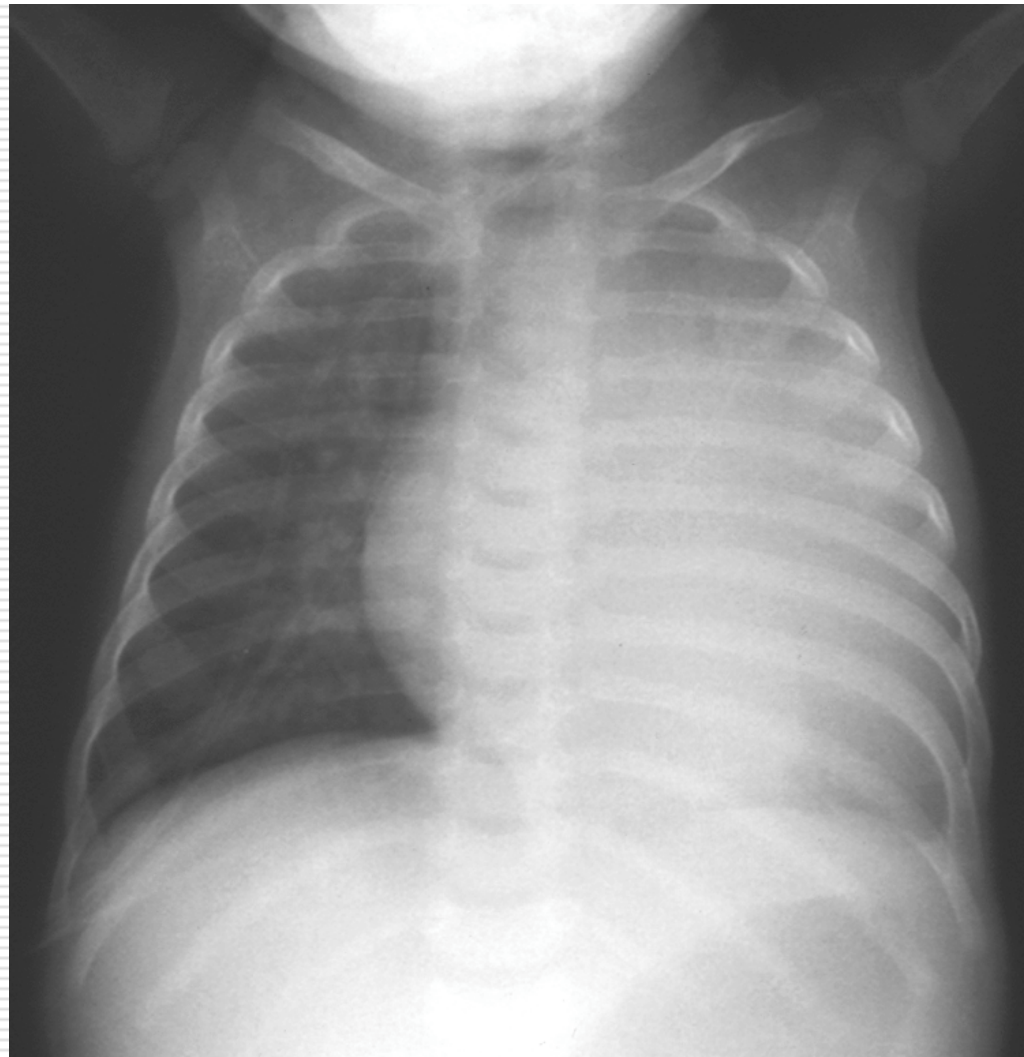
| Cohort # | 1 | 2 | 3 | 4 |
|-----------------|----------------------|----------------------|----------------------|----------------------|
| Dose, vg | 2.1x10 ¹² | 6.9x10 ¹² | 2.1x10 ¹³ | 6.9x10 ¹³ |
| N | 3 | 3 | 3 | 3 |

Mouse Tox/Biodistribution Studies: *rAAV1-CB-hAAT*

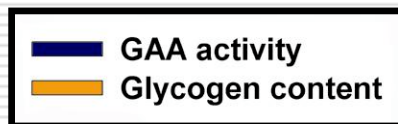
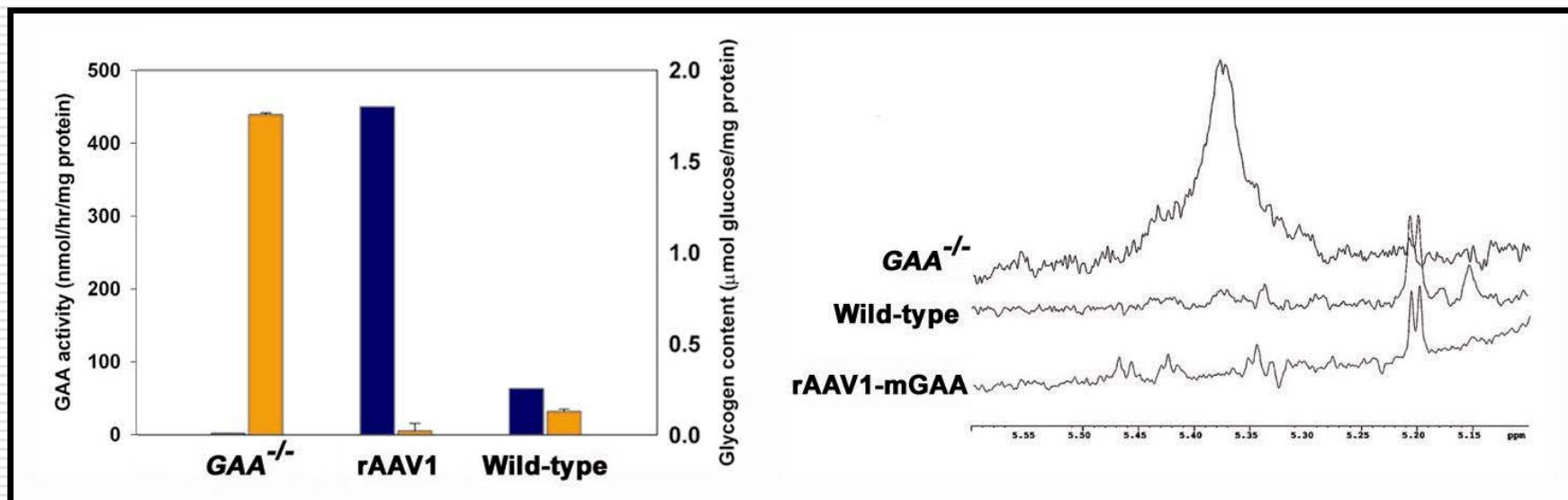
- ❑ Vector prepared in GLP lab, study performed GLP tox lab
- ❑ Dose: up to 4×10^{14} vg/kg (400X)
- ❑ No clinical, heme, chem abnormalities
- ❑ Mild local inflammation at injection site with 400X top human dose
- ❑ hAAT levels confirmed
- ❑ Biodistribution in Blood, other organs, Gonads ->



*Cardiac Hypertrophy in Pompe Disease:
Most severe form of myopathy in infancy*

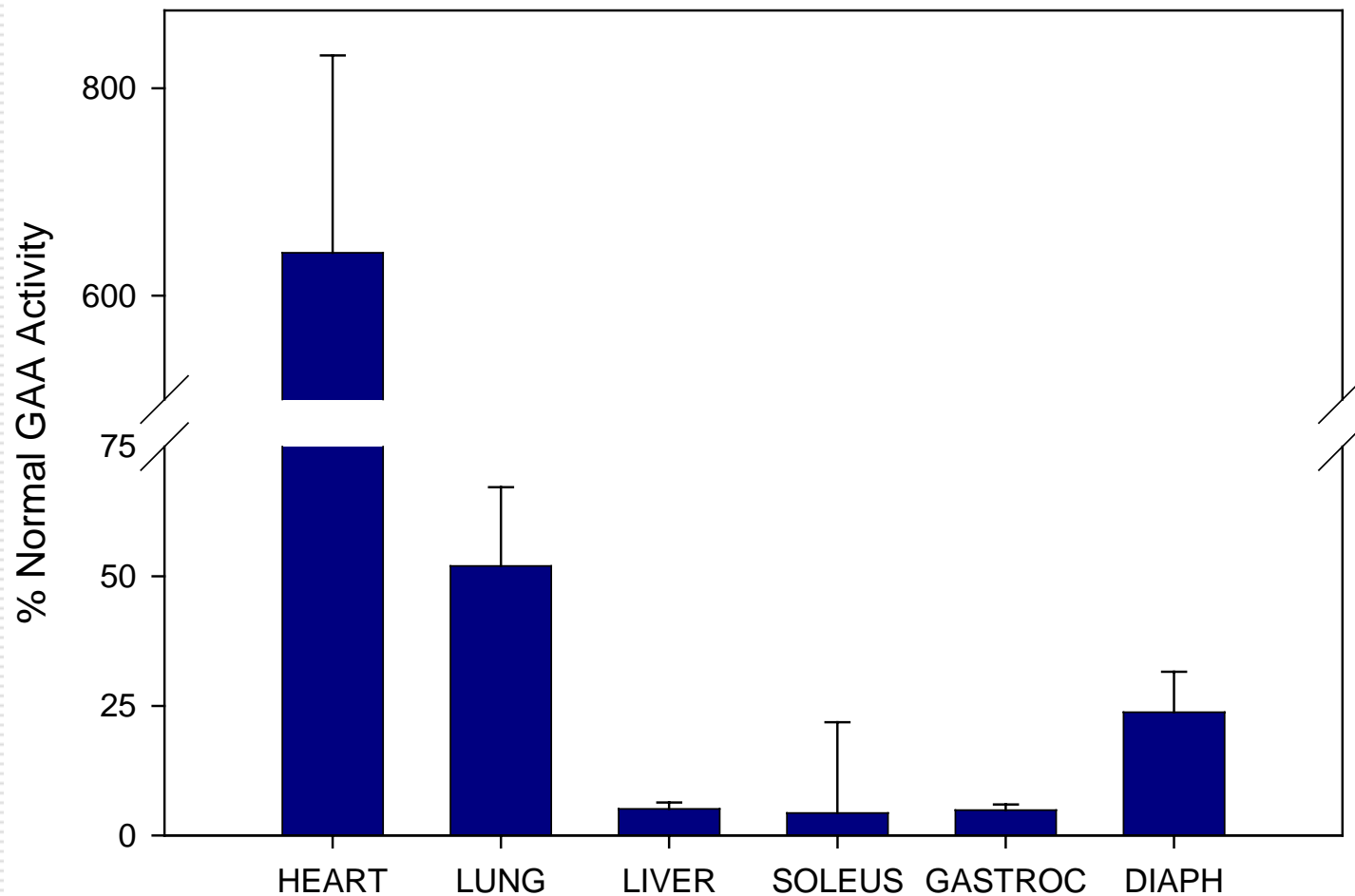


Direct muscle transduction leads to glycogen clearance

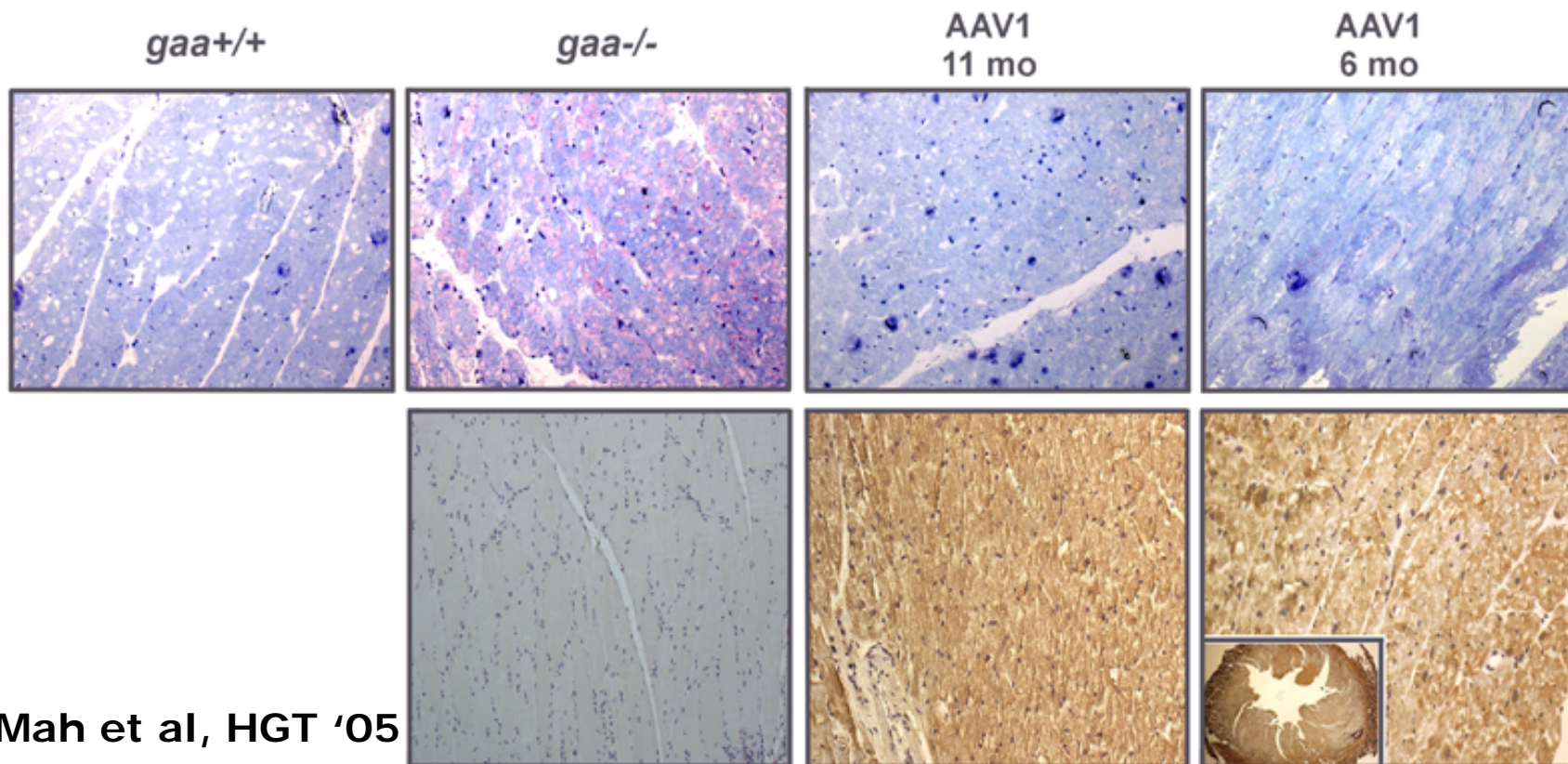


Cardiac Gene Transfer – Importance in Muscular Dystrophy

Mah et al, HGT '05

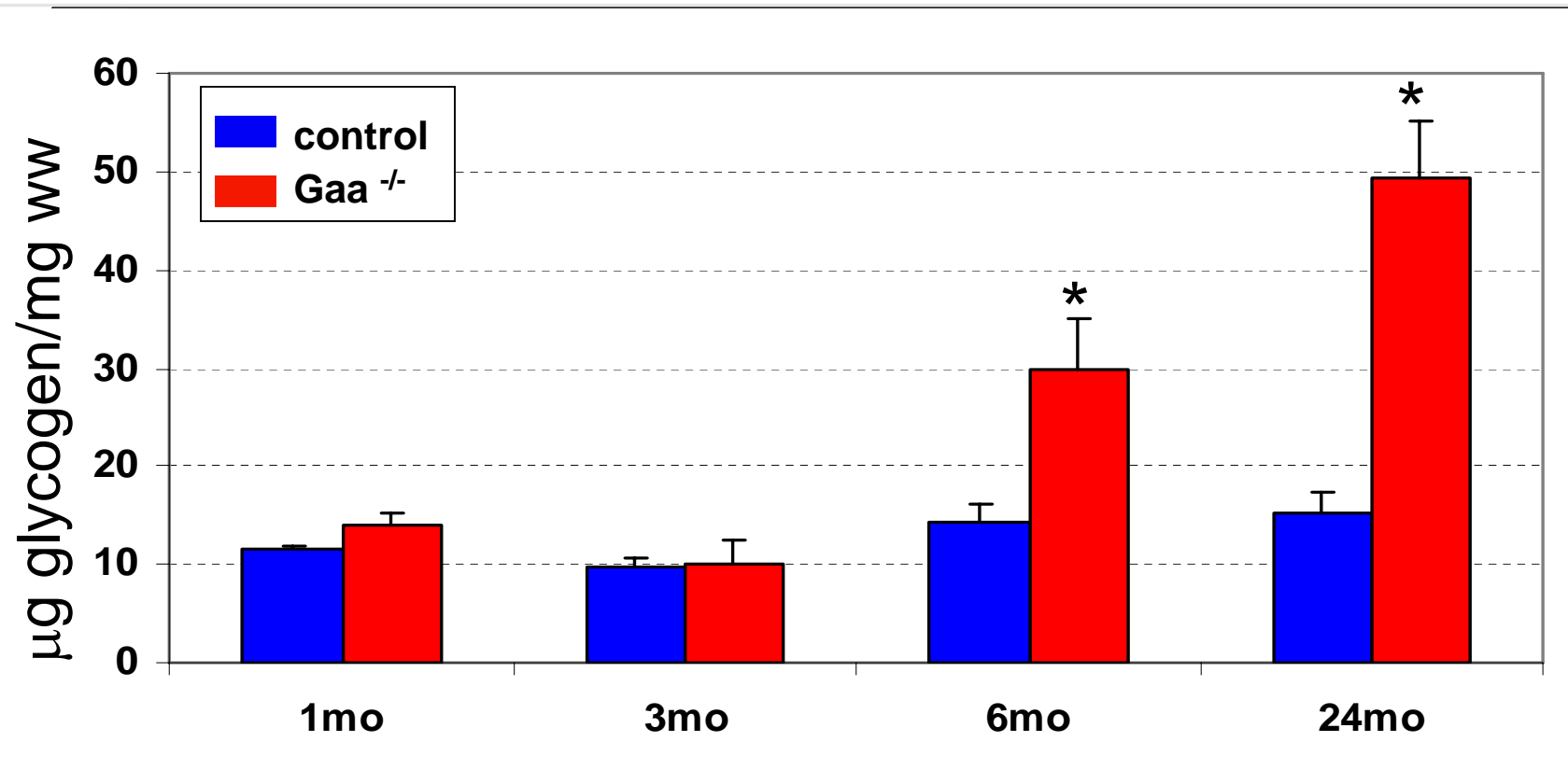


Restoration of GAA and glycogen clearance rAAV1-treated Pompe mice after 11 months



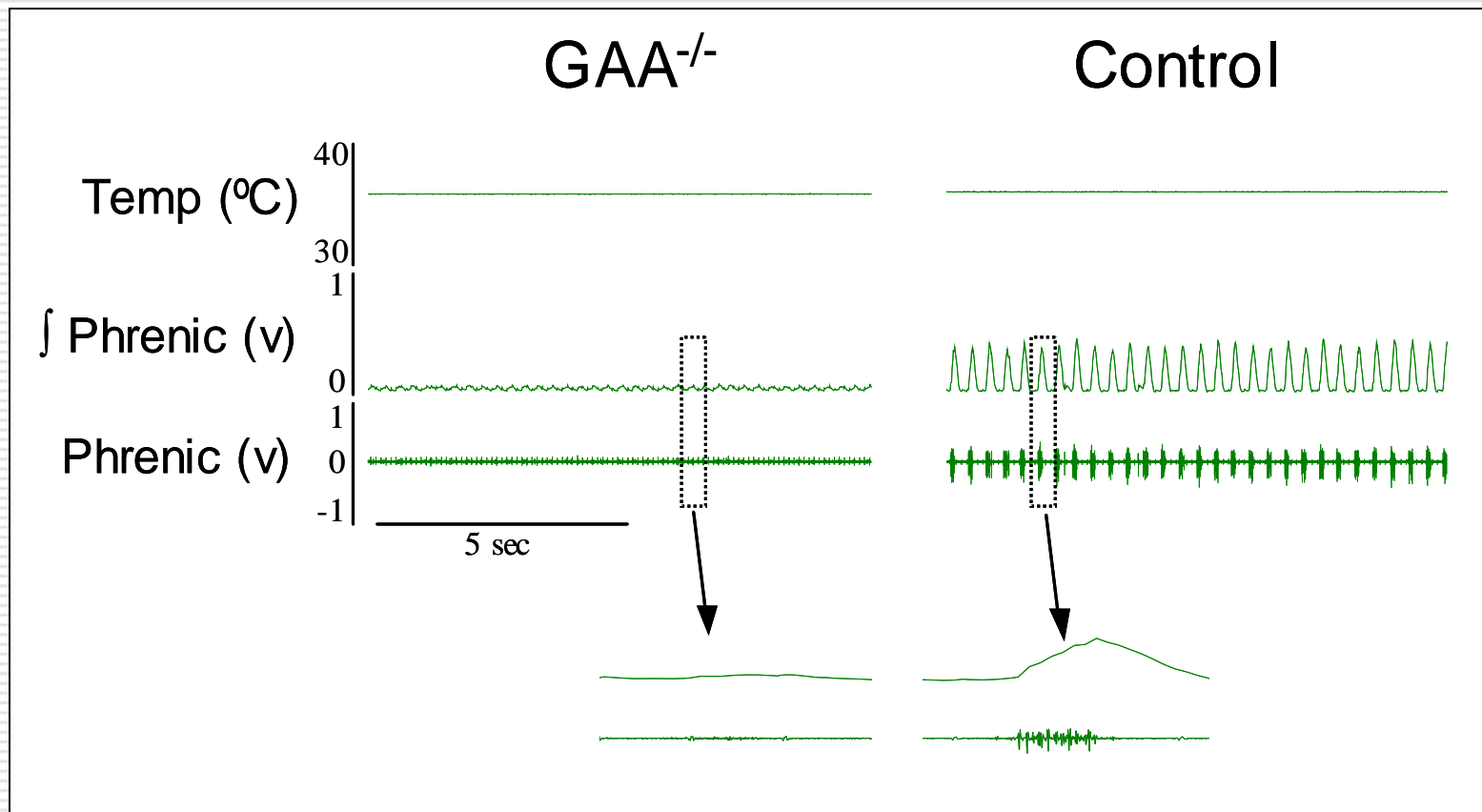
Mah et al, HGT '05

Spinal Cord Glycogen Quantification

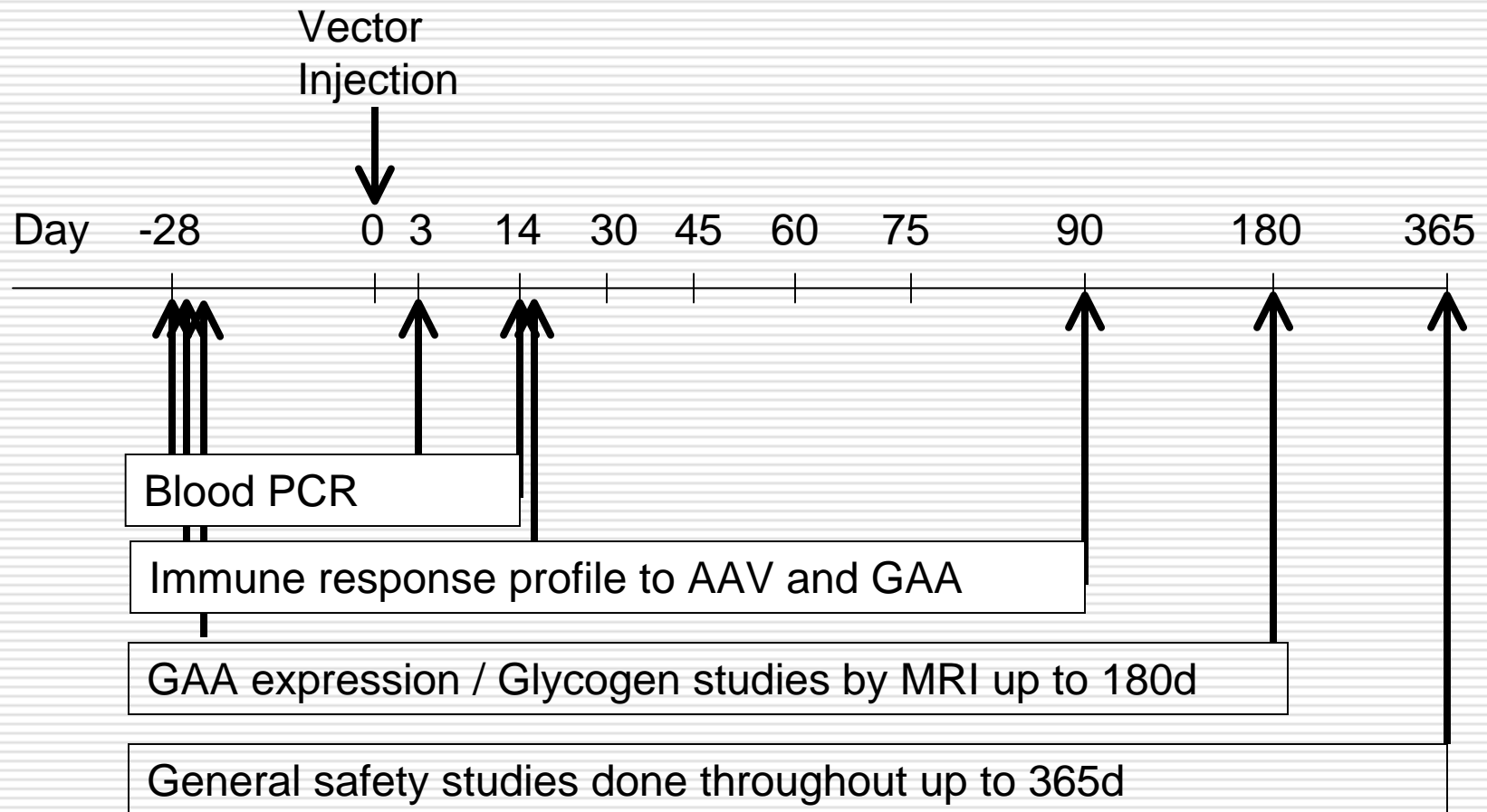


MEAN±SEM

Phrenic Inspiratory Burst Amplitude is Reduced in $GAA^{-/-}$ mouse vs. control



Time Line: Phase I/II rAAV1-GAA



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