An inducible glycogen synthase-1 knockout halts but does not reverse Lafora disease progression in mice

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Supporting information – material included:

Fig. S1
Fig. S2
Fig. S3
Fig. S4
Fig. S5

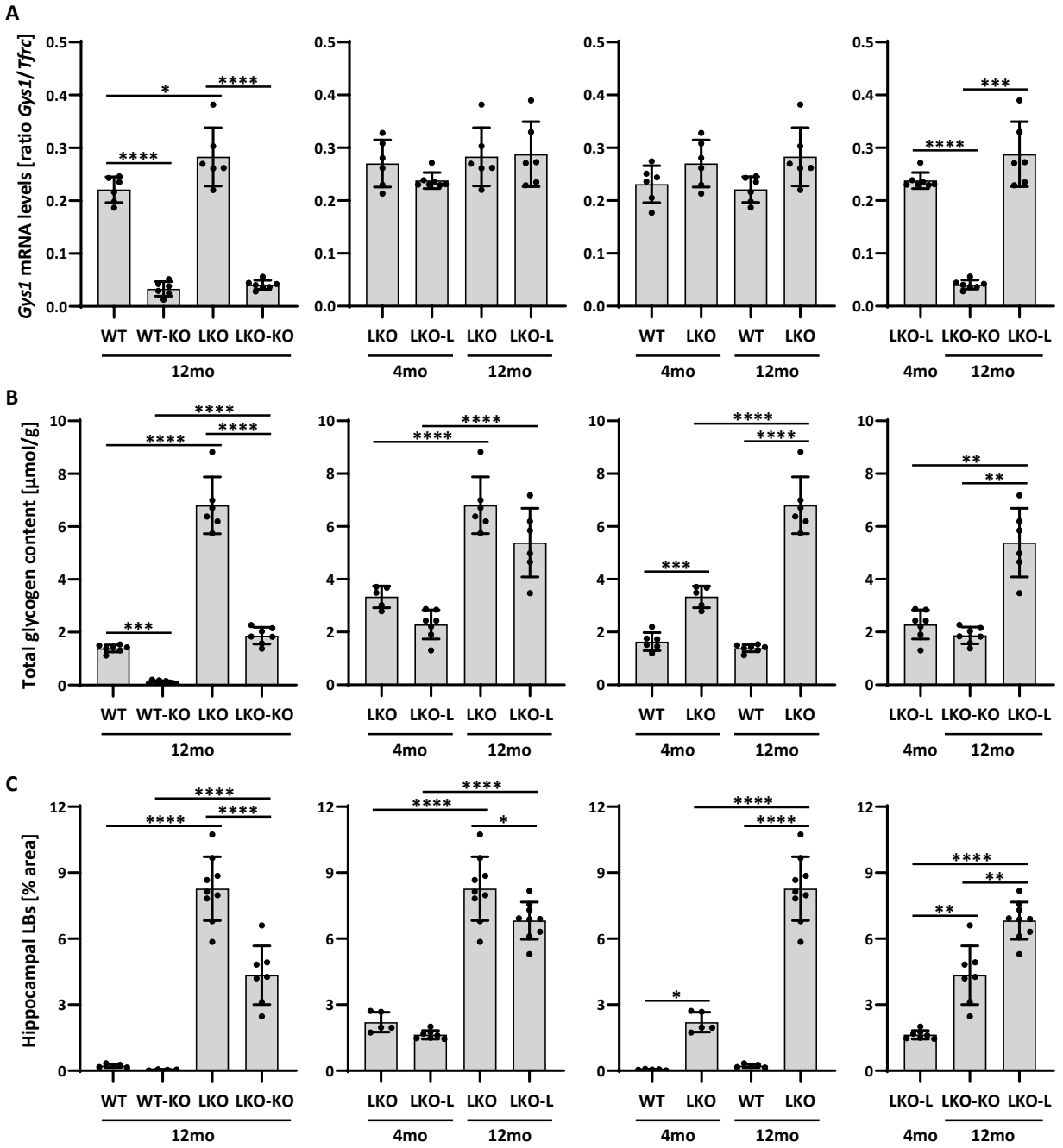
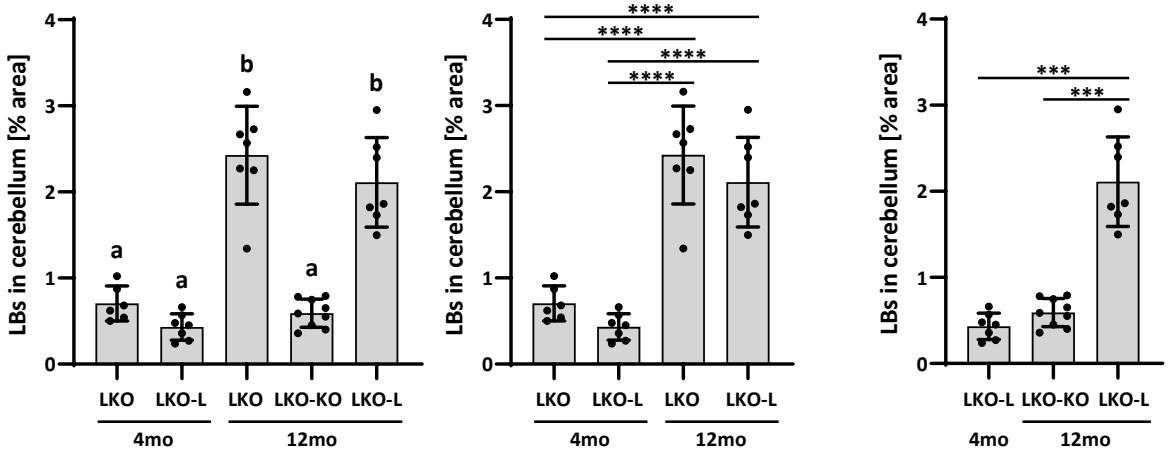
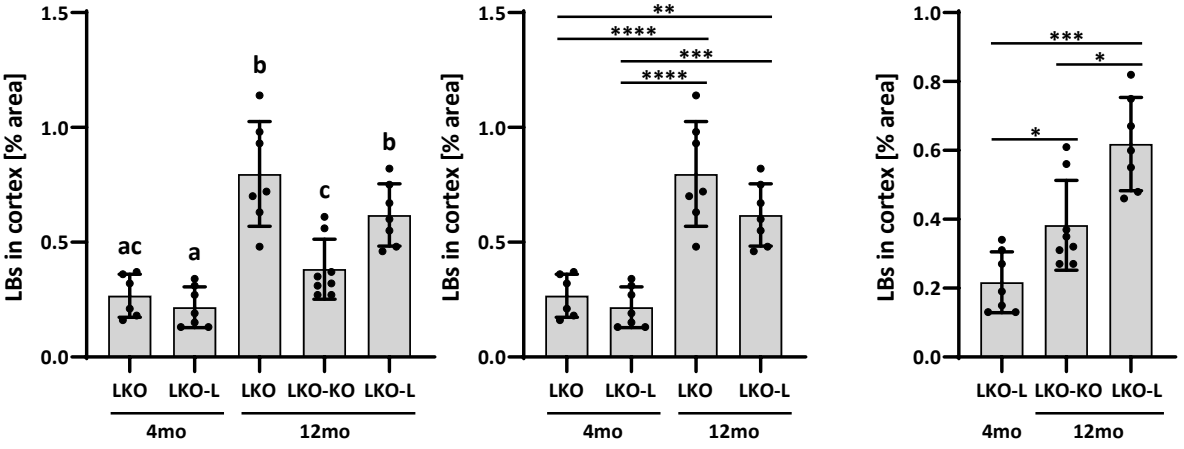


Figure S1. Follow-up statistical testing for *Gys1* mRNA levels (A), total glycogen content (B), and Lafora body (LB) quantification (C) in the brain. Two-way ANOVA for *Gys1* and *Epm2a* genotype (column 1), potential tamoxifen (TAM)-independent Cre recombination (Cre leakage) and age (column 2), and *Epm2a* genotype and age (column 3) as well as Welch's one-way ANOVA for TAM effect in isogenic mice (column 4). Error bars indicate SD. $p < 0.05$ *, $p < 0.01$ **, $p < 0.001$ ***, $p < 0.0001$ ****. Corresponding figure in the main text is Fig. 2.

A


LBs in cerebellum [% area]
a
a
b
a
b
LKO
LKO-L
LKO
LKO-KO
LKO-L
4mo
12mo
****
****
****
****
***
***

B


LBs in cortex [% area]
ac
a
b
c
b
LKO
LKO-L
LKO
LKO-KO
LKO-L
4mo
12mo
**
****
***
****
***
*
*

C

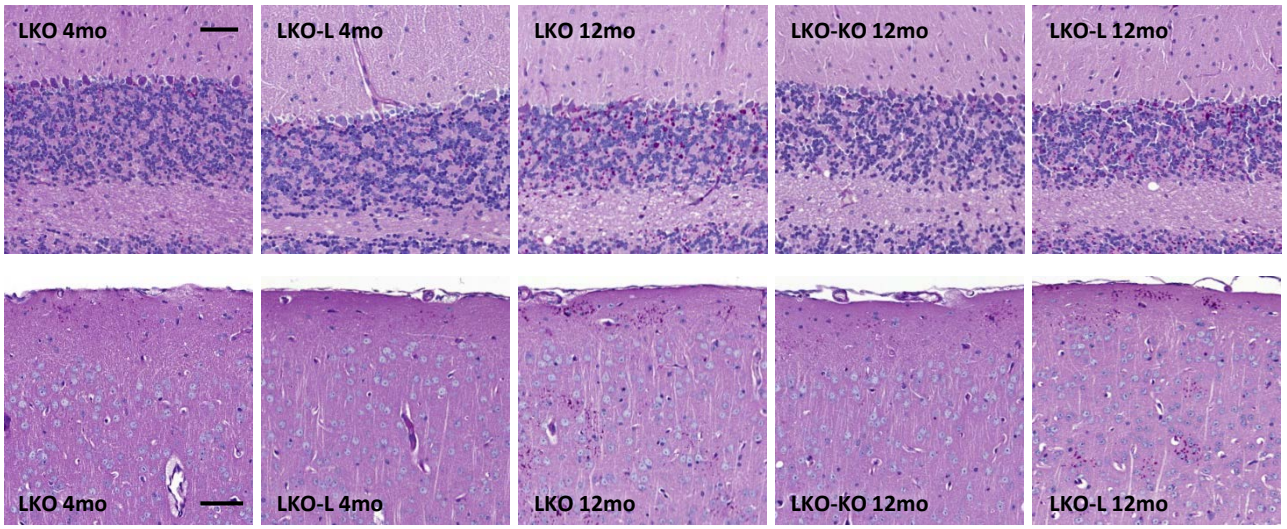

LKO 4mo
LKO-L 4mo
LKO 12mo
LKO-KO 12mo
LKO-L 12mo
LKO 4mo
LKO-L 4mo
LKO 12mo
LKO-KO 12mo
LKO-L 12mo

Figure S2. Lafora body quantification in cerebellum (A) and cortex (B). Error bars indicate SD. Statistical significance ($p < 0.05$) is denoted by different letters (column 1, Welch's one-way ANOVA), while lack of significance is reflected by at least one shared letter. Where applicable, subsets of experimental groups underwent secondary follow-up statistical testing, using two-way ANOVA for potential tamoxifen (TAM)-independent Cre recombination (Cre leakage) and age (column 2) and Welch's one-way ANOVA for TAM effect in isogenic mice (column 3). $p < 0.05$ *, $p < 0.01$ **, $p < 0.001$ ***, $p < 0.0001$ ****. C, Representative images of PASD stained cerebellum (top) and cortex (bottom). Scale bar: 50 µm.

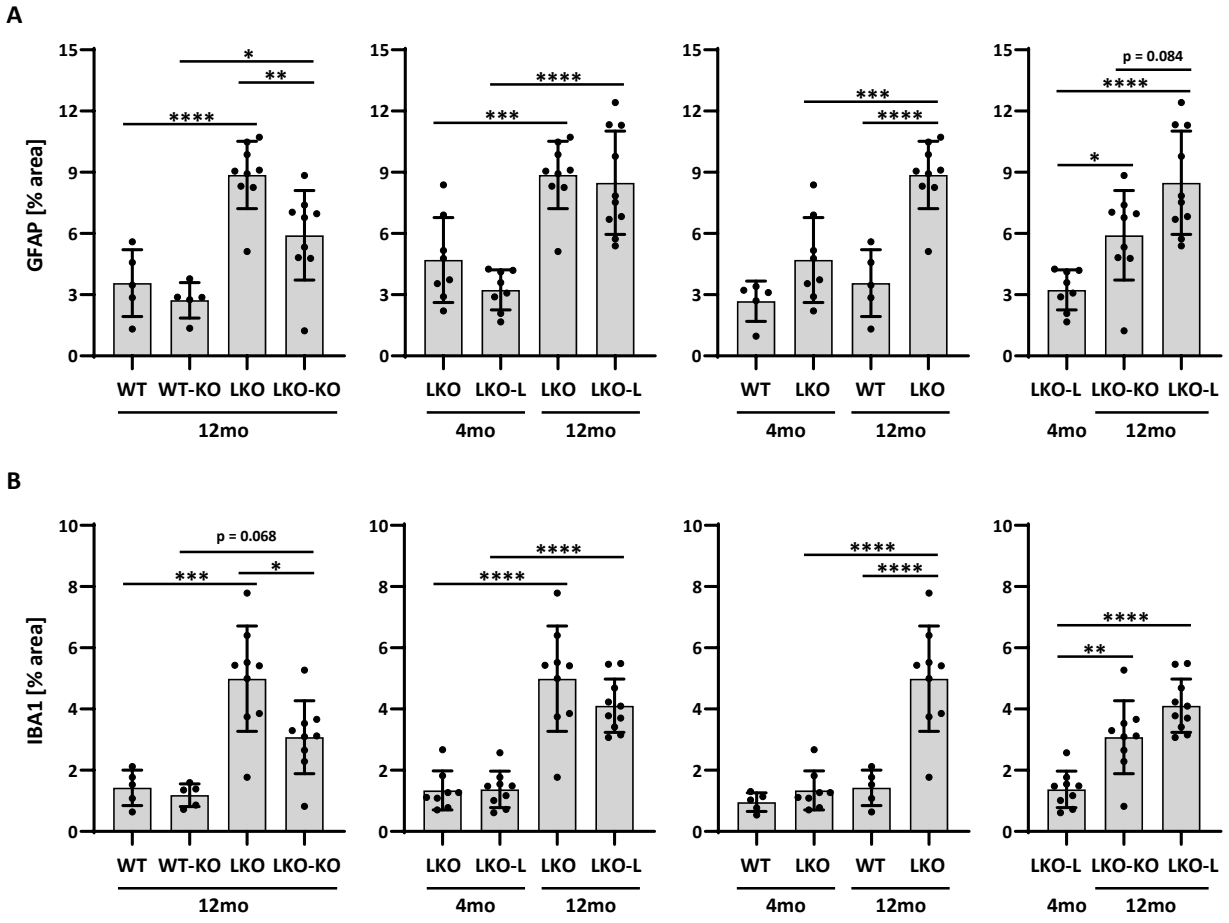


Figure S3. Follow-up statistical testing for GFAP (A) and IBA1 (B) immunohistochemical analysis in the hippocampus. Two-way ANOVA for *Gys1* and *Epm2a* genotype (column 1), potential tamoxifen (TAM)-independent Cre recombination (Cre leakage) and age (column 2), and *Epm2a* genotype status and age (column 3) as well as Welch's one-way ANOVA for TAM effect in isogenic mice (column 4). Error bars indicate SD. $p < 0.05$ *, $p < 0.01$ **, $p < 0.001$ ***, $p < 0.0001$ ****. Corresponding figure in the main text is Fig. 3.

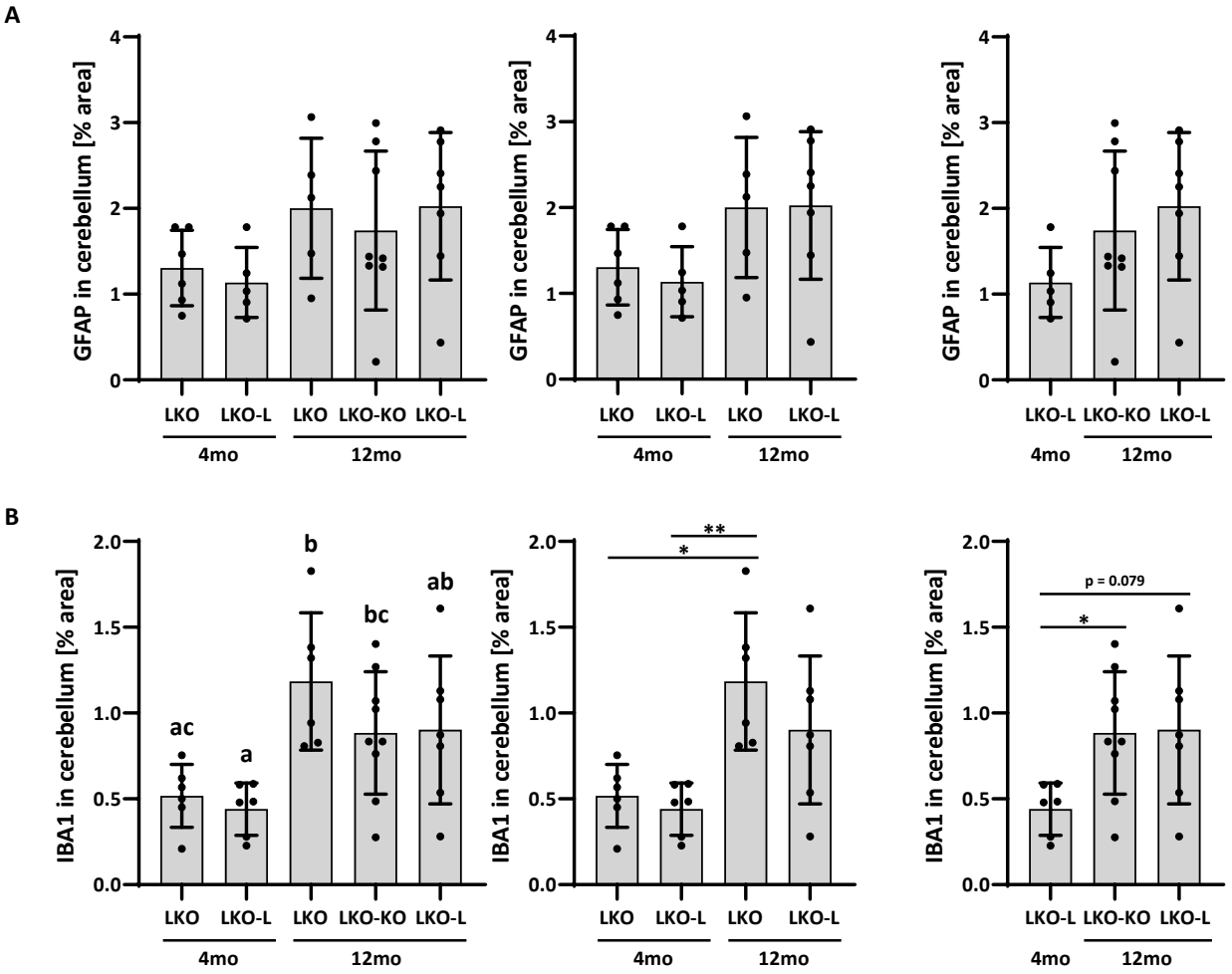


Figure S4. Quantification of GFAP (A) and IBA1 (B) immunohistochemical stain in cerebellum. Error bars indicate SD. Statistical significance ($p < 0.05$) is denoted by different letters (column 1, Welch's one-way ANOVA), while lack of significance is reflected by at least one shared letter. Where applicable, subsets of experimental groups underwent secondary follow-up statistical testing, using two-way ANOVA for potential tamoxifen (TAM)-independent Cre recombination (Cre leakage) and age (column 2) and Welch's one-way ANOVA for TAM effect in isogenic mice (column 3). $p < 0.05$ *, $p < 0.01$ **.

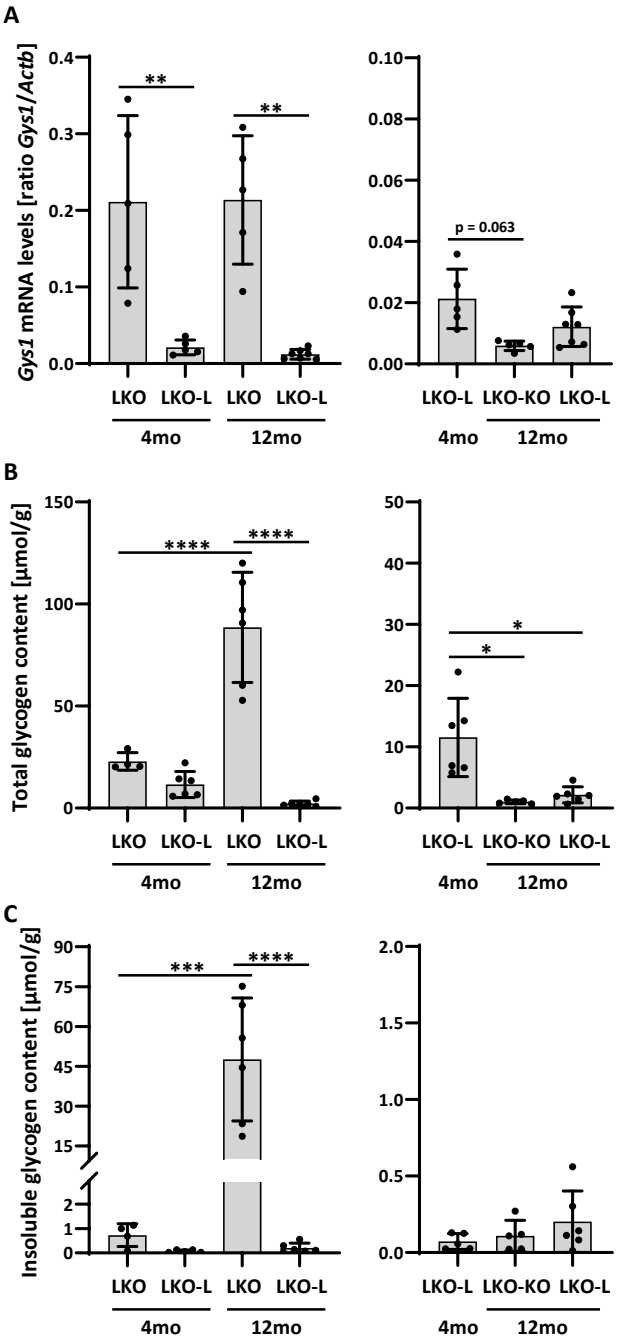


Figure S5. Follow-up statistical testing for *Gys1* mRNA levels (A), total glycogen content (B), and insoluble glycogen analyses (C) in the muscle. Two-way ANOVA for potential tamoxifen (TAM)-independent Cre recombination (Cre leakage) and age (column 1) and Welch's one-way ANOVA for TAM effect in isogenic mice (column 2). Error bars indicate SD. $p < 0.05$ *, $p < 0.01$ **, $p < 0.001$ ***, $p < 0.0001$ ****. Corresponding figure in the main text is Fig. 4.