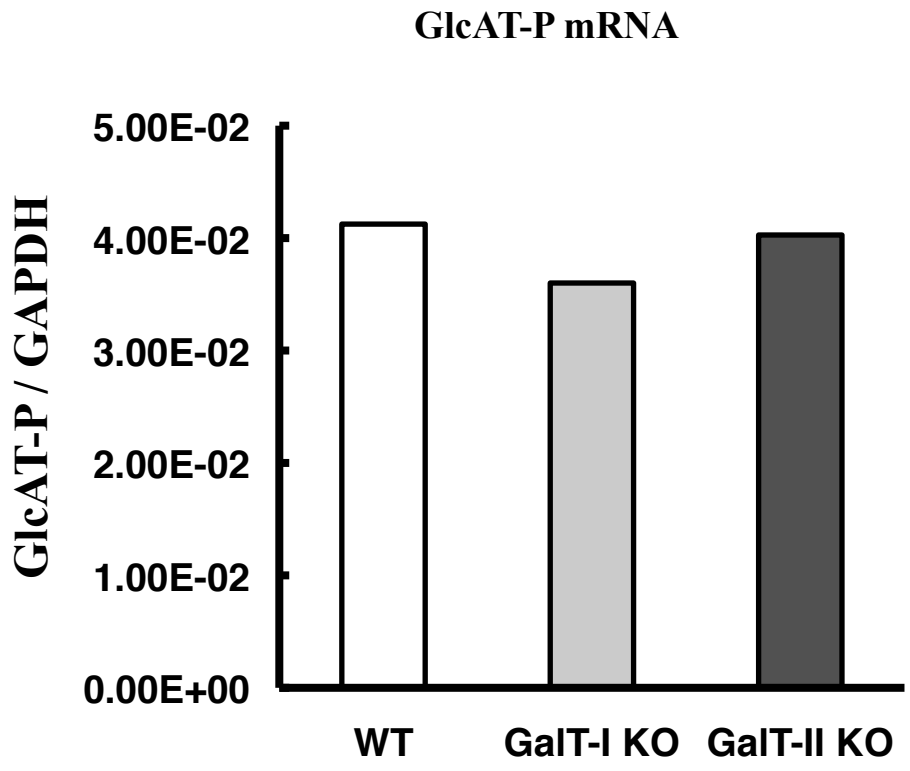
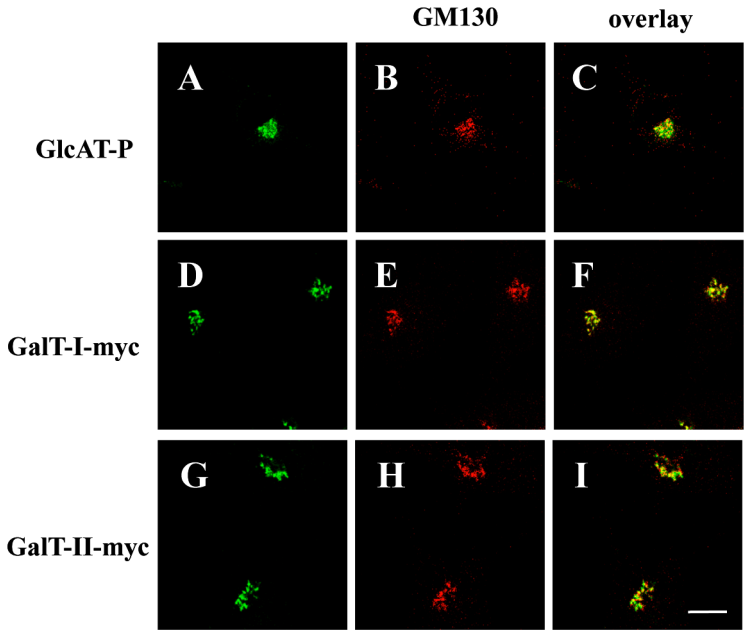
Supplemental Fig. 1



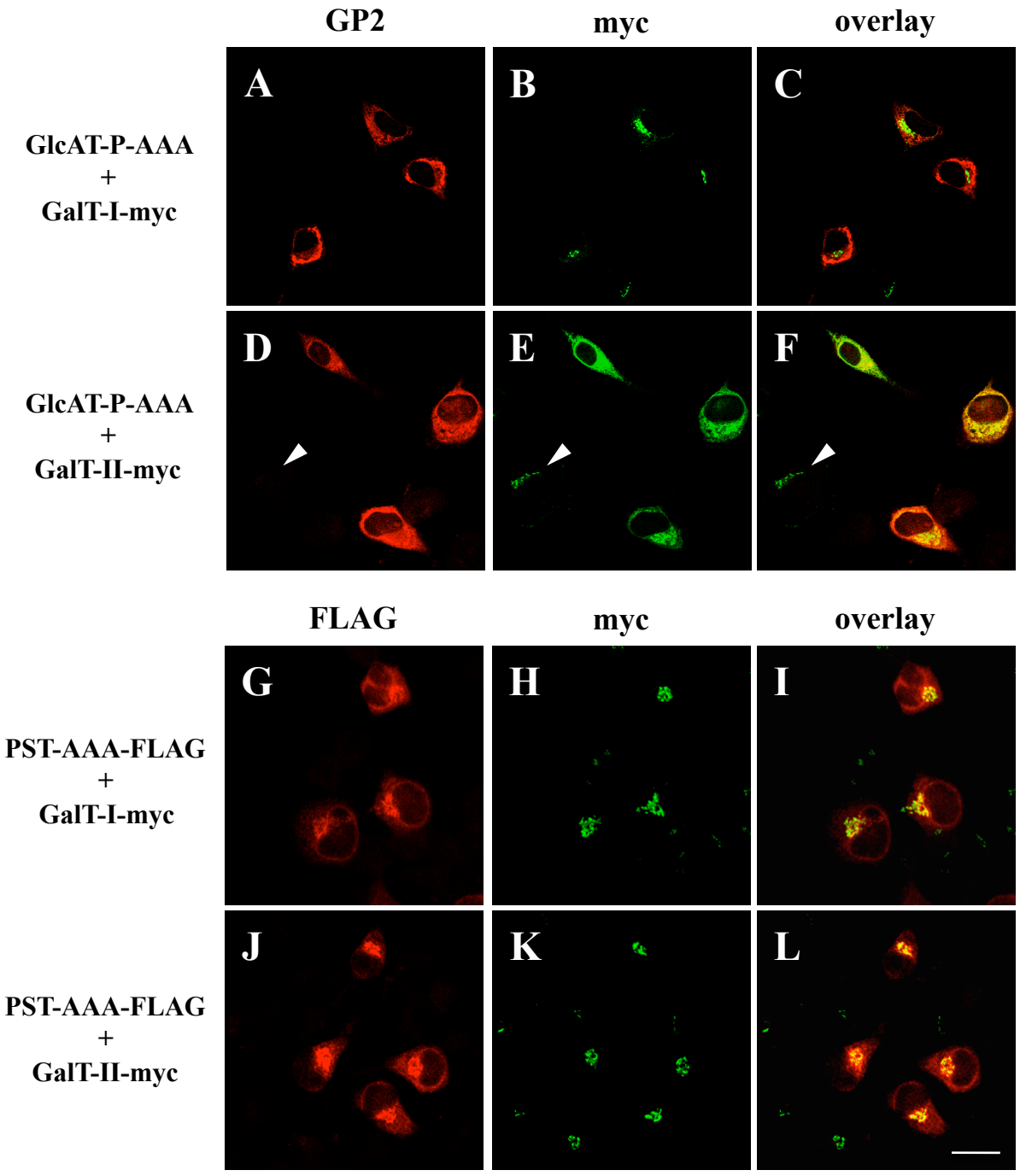
Supplemental Fig. 1 **Expression of GlcAT-P transcripts in mouse brain.** Levels of GlcAT-P mRNA were examined by quantitative real-time PCR. Total brain RNA was extracted from 10-11-week old C57BL/6 wild type (WT), β4GalT-I -/- (GalT-I KO) and β4GalT-II -/- (GalT-II KO) mice (WT, n=2; GalT-I KO, n=1; GalT-II KO, n=3). The cDNA was reverse-transcribed by SuperScript III reverse transcriptase (Invitrogen) according to the instructions of the supplier. The expression of GlcAT-P mRNA was estimated using the SYBR green reagent (SYBR Premix Ex Taq II; TAKARA BIO) with optimal thermal cycle in Thermal Cycler Dice Real Time System Single (TAKARA BIO). GAPDH mRNA was amplified from all samples to normalize the expression level. The following primers were used: GlcAT-P, 5'-AAAGACCAGAGAGCTGGGGATTCCG and 5'-CAGCTCTGAGGAAGAGCAGGTGTGA; GAPDH, 5'-ATGAATACGGCTACAGCAACAGG and 5'-CTCTTGCTCAGTGTCCTTGCTG.

Supplemental Fig. 2



Supplemental Fig. 2 **Distribution of GlcAT-P, β4GalT-I-myc, and β4GalT-II-myc in N2a cells.** N2a cells were transiently transfected with GlcAT-P, β4GalT-I-myc, and β4GalT-II-myc. GlcAT-P was detected with GP2 pAb and β4GalT-myc (I and II) was detected with anti-myc pAb (A, D, G). Cells were immunostained with anti-GM130 mAb (B, E, H). The right panels show overlaid images (C, F, I). Bar, 10 μm.

Supplemental Fig. 3



Supplemental Fig. 3 **ER retention assay using GlcAT-P-AAA and PST-AAA-FLAG.** A-F, N2a cells were transiently co-transfected with GlcAT-P-AAA and β4GalT-I-myc or β4GalT-II-myc. At 24 h post-transfection, cells were washed with PBS and fixed with ice-cold methanol for 10 minutes at -20°C. Then, cells were incubated with primary antibodies followed by Alexa Fluor 546 (*red*) and 488 (*green*)-conjugated secondary antibodies. GlcAT-P-AAA was detected with GP2 pAb (A, D) and β4GalT-myc (I and II) was detected with anti-myc mAb (B, E). C and F, overlaid images. Note that β4GalT-II showed normal Golgi-localization in the GlcAT-P-AAA-negative cell (*arrowheads* in D, E, and F). G-L, N2a cells were transiently co-expressed with PST-AAA-FLAG and β4GalT-I-myc or β4GalT-II-myc. Cells were immunostained as described above. PST-AAA-FLAG was detected with anti-FLAG pAb (G, J) and β4GalT-myc (I and II) was detected with anti-myc mAb (H, K). I and L, overlaid images. Bar, 20 μm.