FIGURE S1

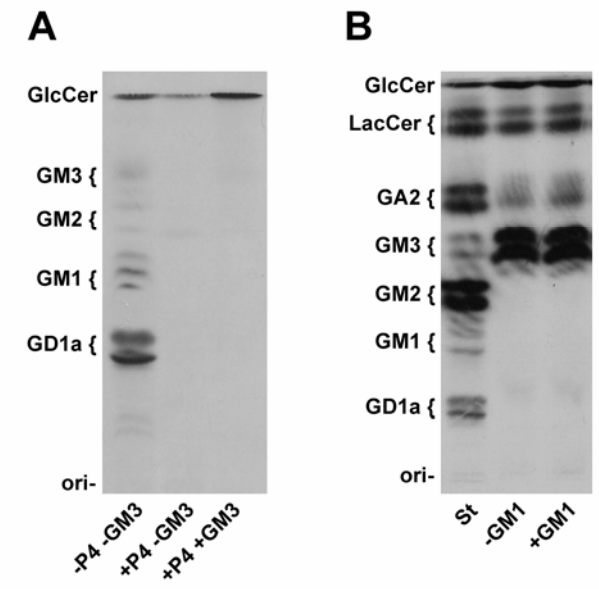

Figure S1: Exogenous GM3 and GM1 gangliosides are not suitable substrates for Golgi-associated glycosyltransferases in CHO-K1 cells. (A) CHO-K1$^{GalNAc\text{-}T+/Gal\text{-}T2+}$ (clone 4) cells were grown without P4 (-P4) or with P4 (+P4) for 4 days. Then, cells were incubated (+GM3) or not (-GM3) with 100 μM GM3 for 4 h, washed, and metabolically labeled with 40 μCi/ml [^{14}C]-galactose for 12 h. Next, lipid extracts were purified, resolved by HPTLC, and visualized as indicated in Experimental Procedures. The positions of radioactive glycolipid are indicated on the left of plate. GM1, GD1a, GD1b and GT1b were also co-chromatographed and visualized by exposing the plate to iodine vapor. (**B**) Wild-type CHO-K1 cells (predominantly expressing the ganglioside GM3) were incubated (+GM1) or not (-GM1) with 100 μM GM1 for 3 h, washed, and metabolically labeled with 40 μCi/ml [^{14}C]-galactose for 12 h. Next, lipid extracts were purified, resolved by HPTLC, and visualized as indicated in Experimental Procedures. The positions of co-chromatographed radioactive glycolipid standards (St) are indicated on the left of the plate. Lipids migrate as multiple bands on the HPTLC plate because of the heterogeneity of the fatty acyl chains of the molecules.

Overall, these results rule out the possibility that exogenously incorporated GM3 or GM1 arrive to the Golgi complex to be used as an acceptor substrate for ganglioside glycosyltransferases.