Figure S1. **O-glycosylation markers are redistributed from the Golgi apparatus under PDGF growth factor stimulation.** (A–C) HPL staining at the Golgi (Giantin) in unstimulated HeLa cells grown throughout in 10% FBS (B) or serum starved overnight (A) is redistributed out of the Golgi after PDGF treatment (50 ng/ml) for 2 h (C). (D and E) Anti-Tn staining colocalizes with HPL at the Golgi (Giantin) in unstimulated HeLa cells (D) or away from the Golgi in punctate cytoplasmic structures after PDGF treatment for 2 h (E). (F) GalNac-T1 staining colocalizes with HPL in punctate and diffuse cytoplasmic structures after PDGF treatment for 2 h. In all images nuclei were stained using Hoechst and colored blue. Bar, 10 µm.
Inhibition of transcription does not block HPL redistribution from the Golgi under growth factor stimulation. (A) HPL staining is highly enriched at the Golgi (Giantin) in serum-starved HeLa cells before growth factor stimulation. (B) Incubation of serum-starved HeLa cells with RNA polymerase II inhibitor α-Amanitin (20 µg/ml) alone for 2 h does not promote significant redistribution of HPL staining from the Golgi apparatus. (C and D) Redistribution of HPL staining from the Golgi under EGF stimulation (100 ng/ml) for 2 h (C) is not affected by cotreatment with α-Amanitin (D; 20 µg/ml). (E) Quantification of HPL enrichment at the Golgi in HeLa cells treated with EGF alone or cotreated with EGF and α-Amanitin (20 µg/ml) for indicated times. 30 cells were quantified for each sample. Error bars show SEM. Statistical significance (p) measured by two-tailed paired t test relative to mean HPL staining in EGF-only treated cells at each time point. This is a representative example from two independent experiments. In all images nuclei were stained using Hoechst and colored blue. Bar, 10 µm.
**Figure S3.** HPL staining is redistributed from the Golgi apparatus only in Src-activated SYFsrc fibroblasts and not Src-deficient SYF fibroblasts under growth factor stimulation. (A and B) HPL staining in serum-starved Src-deficient SYF cells (A) or Src-activated SYFsrc cells (B) before EGF stimulation. Image acquisition parameters were optimized for visualization of HPL staining in the Golgi (Giantin). (C and D) HPL staining remains localized exclusively to the Golgi in EGF-treated (50 ng/ml) Src-deficient SYF fibroblasts for 2 h (C) but is significantly redistributed from the Golgi in Src-activated SYFsrc fibroblasts treated in a similar manner (D). (E and F) HPL staining remains localized exclusively to the Golgi in PDGF-treated (25 ng/ml) Src-deficient SYF fibroblasts for 2 h (E) but is significantly redistributed from the Golgi in Src-activated SYFsrc fibroblasts treated in a similar manner (F). In all images nuclei were stained using Hoechst and colored blue. Bar, 10 µm.
Figure S4. GalNac-T1, -T4, and -T6 glycosyltransferases are redistributed away from the Golgi under increased Src expression. (A–C) Co-microinjection of Src(E378G)-DsRed and ManII-GFP plasmids in WI38 human fibroblasts and staining for endogenous GalNac-T1 (A), -T4 (B), and -T6 (C). Untransfected cells are highlighted by arrows and Src(E378G)-DsRed-expressing cells by arrowheads. In all images nuclei were stained using Hoechst and colored blue. Bar, 20 µm.
Figure S5. ManIC, C2GnT, and GaIT glycosyltransferases as well as the cis-Golgi matrix protein GM130 are not redistributed with HPL staining from the Golgi under increased Src expression. (A and B) Co-microinjection of HA-tagged ManIC (A) or C2GnT-DsRed (B) with active Src(E378G) in SYF mouse fibroblasts. (C) Co-microinjection of GaIT-GFP reporter with active Src(E378G) in SYF mouse fibroblasts. (D) Microinjection of active Src(E378G) in SYF mouse fibroblasts and staining for endogenous GM130 Golgi matrix protein. In all panels untransfected cells are highlighted by arrows and Src(E378G)-DsRed–expressing cells by arrowheads. Cells transfected with Src were identified in A, C, and D using an anti-Src antibody and in B using an anti-pTyr antibody (PY20) that binds to proteins containing phosphorylated tyrosines. In all images nuclei were stained using Hoechst and colored blue. Bar, 20 µm.