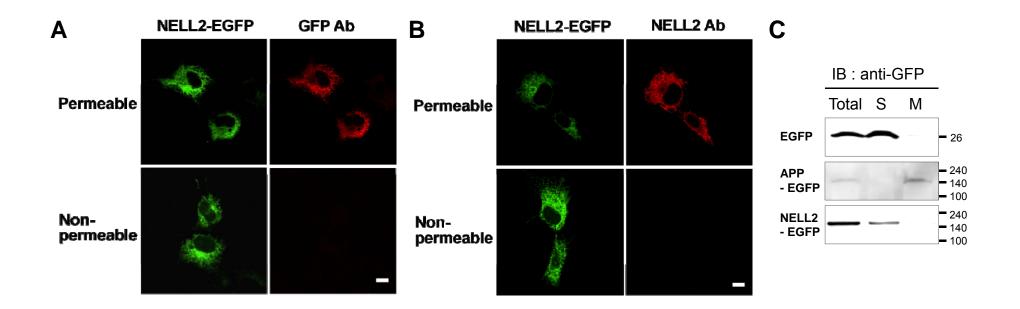


Supplemental Figure 1. (A) Effect of tunicamycin on the glycosylation of NELL2 or NELL2-EGFP. HEK293T cells expressing NELL2 or NELL2-EGFP were treated with tunicamycin at 5 μ g/ml for 18 h. The cells were harvested and then analyzed by Western blot analysis with anti-GFP or anti-NELL2 antibody. (B) Colocalization of NELL2 with ER or Golgi markers. HiB5 cells were transiently transfected with NELL2-EGFP and markers for subcellular organs such as pECFP-ER (ER), pECFP-Golgi (Golgi), and pDsRed-Mito (Mitochondria). Colocalization was observed in the Golgi and ER. Scale bar = 20 μ m.



Supplemental Figure 2. NELL2 is rarely present on the cell surface. HiB5 cells expressing NELL2-EGFP were fixed with 3.7% formaldehyde after which they were either stained directly or first permeablized with 0.25% Triton X-100. The cells were stained with either GFP antibody (A) or NELL2 antibody (B). NELL2 antibody recognizes the middle region of NELL2 and displays the typical vesicular pattern of NELL2 expression in permeabilized cells. Scale bar = $20 \ \mu m$. (C) Protein extracts from HEK293T cells expressing EGFP(soluble protein), APP-EGFP(type I transmembrane protein) or NELL2-EGFP were fractionated into soluble (S) and membrane (M) fractions by ultracentrifugation, and then analyzed by Western blot analysis with anti-GFP antibody.