Supplemental Information

Interaction of Munc18c and Syntaxin4 facilitates invadopodium formation and extracellular matrix invasion of tumour cells

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Running Title: Munc18c regulates Stx4 function during ECM invasion

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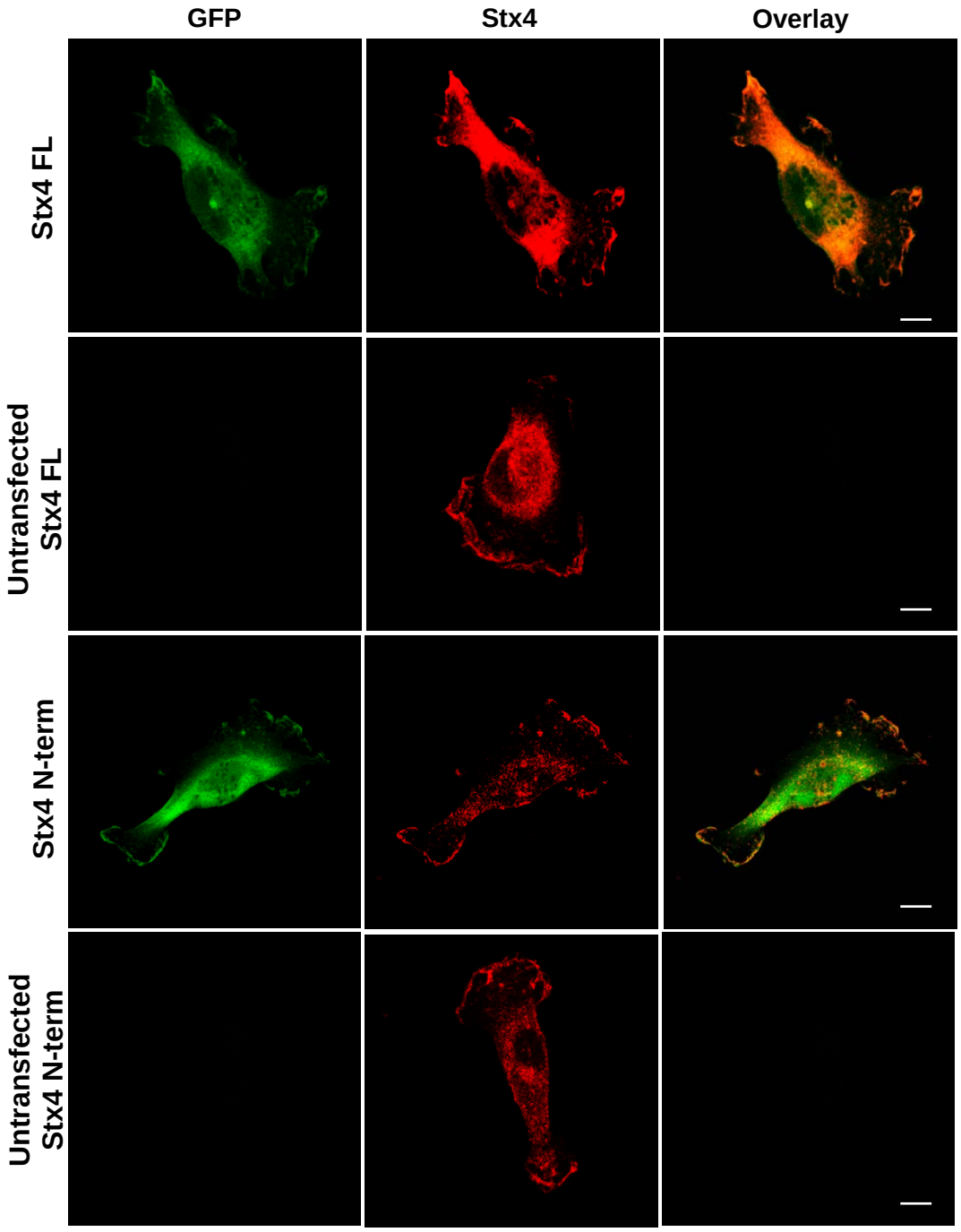
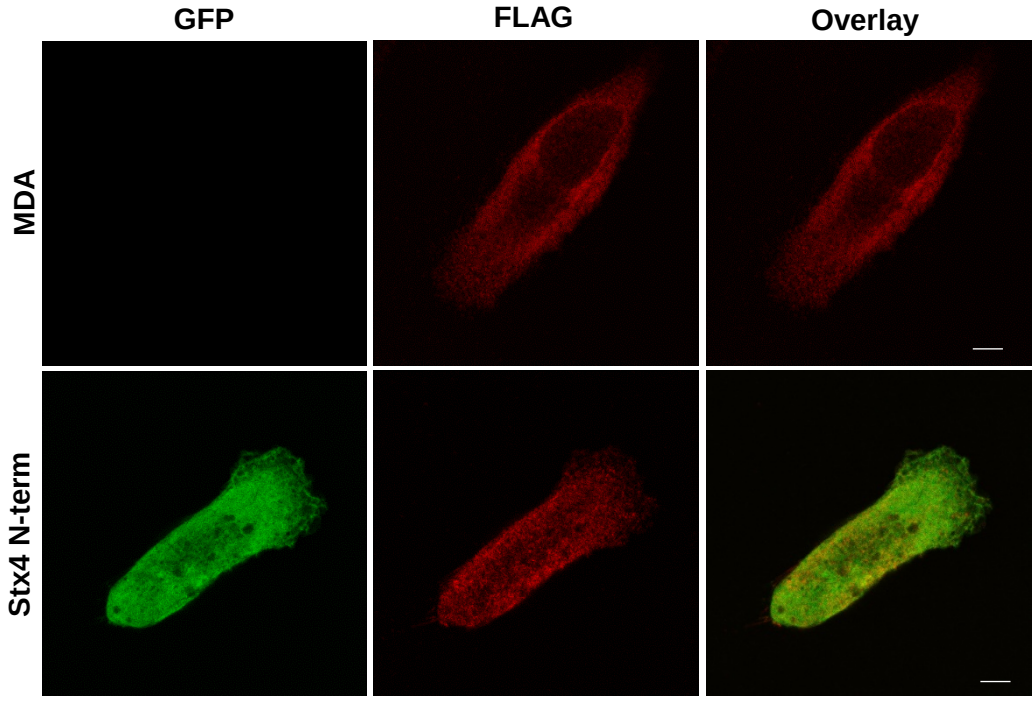
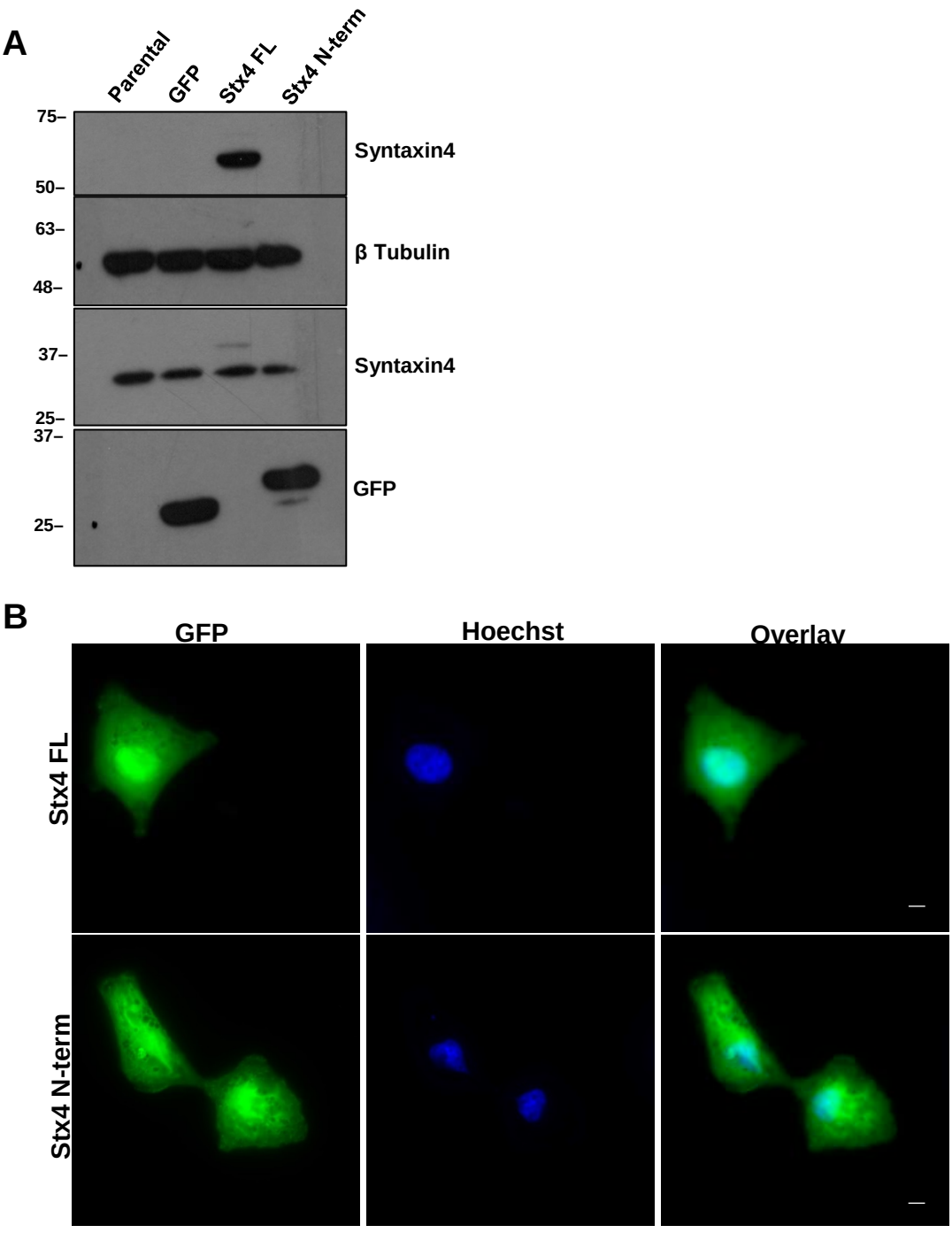


Supplemental Figure 1. Transiently transfected cells expressing Stx4-FL and Stx4 N-terminal peptide display co-localization of GFP and Syntaxin4. Cells were transfected with either Stx4-FL or Stx4 N-terminal peptide for 24 hours, seeded onto coverslips for 12 hours, then fixed, permeabilized, and stained for Stx4. Cells were analyzed by confocal microscopy. A representative section from the ventral region of a cell is shown for cells that were GFP positive, as well as an untransfected cell in the same sample. Scale bar, 10 μm.



Supplemental Figure 2. Parental and stable cells expressing Stx4 N-terminal peptide display equal levels of Stx4-FL-3xFLAG expression. Parental MDA-MB-231 cells and stable cells expressing Stx4 N-terminal peptide were transfected with Stx4-FL-3xFLAG for 24 hours. Cells were then seeded onto coverslips for 12 hours, and fixed, permeabilized, and stained for FLAG. Cells were analyzed by confocal microscopy with equal exposure to verify equal exogenous protein expression. Scale bar, 10 μm.



Supplemental Figure 3. Stable cell lines expressing GFP, Stx4-FL, and Stx4 N-terminal peptide display equal levels of expression. (A) Parental MDA-MB-231 cells and stable cells lines expressing GFP, Stx4-FL, and Stx4 N-terminal peptide were lysed and analyzed by Western Blot. Protein levels of Syntaxin4, β Tubulin, and GFP were compared between the cell lines. (B) Stable cells expressing Stx4-FL and Stx4 N-terminal peptide were seeded onto coverslips for 12 hours, fixed, and stained with Hoechst. Cells were analyzed by epifluorescent microscopy with equal exposure to verify equal exogenous protein expression. Scale bar, 10 μm.