

Supplemental_Fig_S2

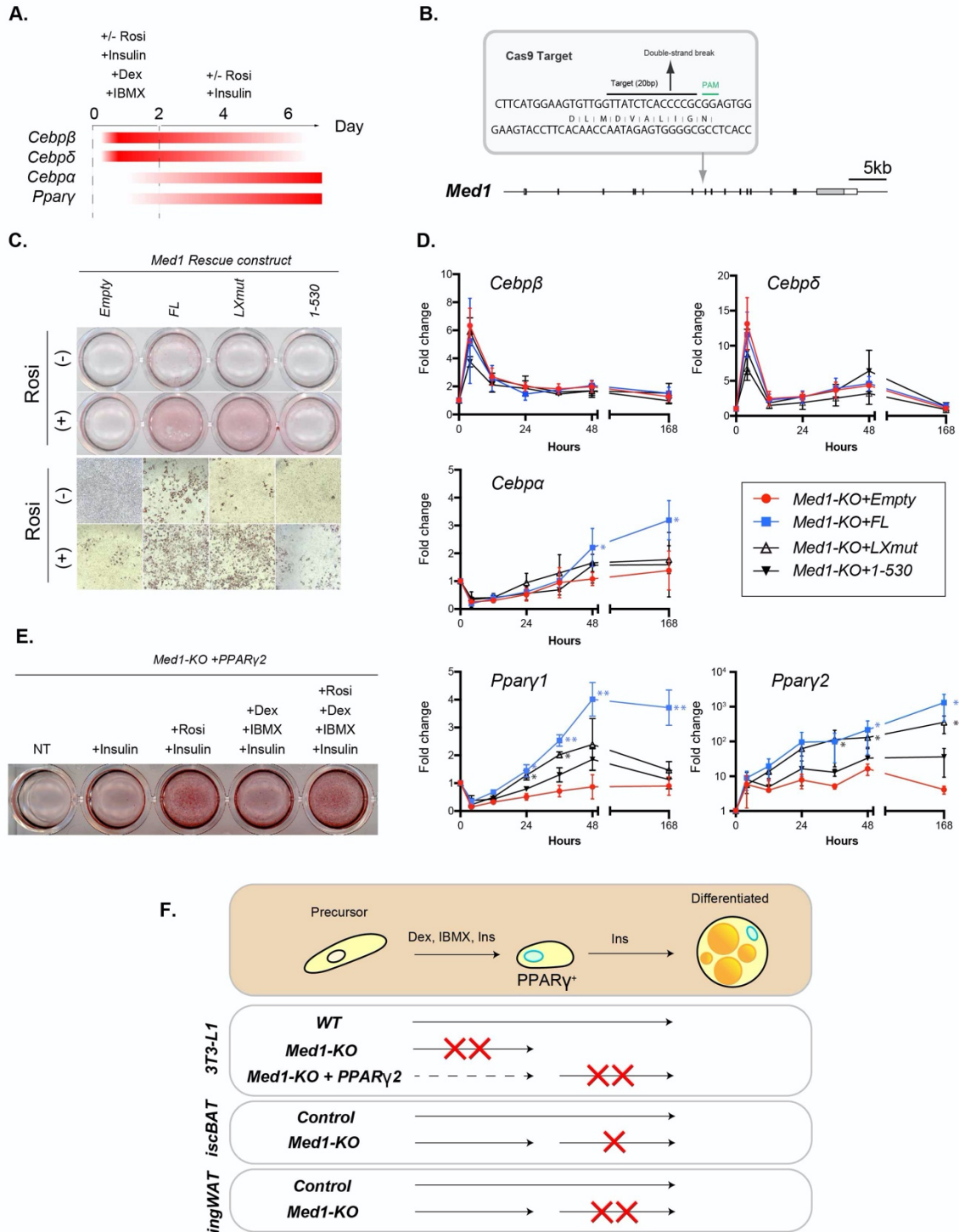


Figure S2: MED1 is required at distinct steps in 3T3-L1 differentiation

A) Diagram of the expression pattern of adipogenic transcription factors during differentiation. **B)** Design of *Med1*-knockout by CRISPR/Cas9. sgRNA is designed at exon 9. **C)** Oil-red-O staining of differentiated *Med1*-KO 3T3-L1 cells expressing different forms of MED1 in the absence or presence of Rosi. Rosi efficiently rescues the adipogenesis defect in cells re-expressing full length and LX-mutant forms of MED1. **D)** qRT-PCR of adipogenic transcription factors at 0, 12, 24, 36, 48, and 168 hours of differentiation of 3T3-L1 cells expressing different forms of MED1 in conventional culture. 36B4 (RPLP0) mRNA expression was used as internal control, and expression levels at 0 hours are set as 1 for each condition (mean, \pm SD, n=3, *p<0.05 (T-test): compared to *Med1*-KO NC). Representative data from KO Clone#15 is presented. **E)** Oil-red-O staining of *Med1*-KO 3T3-L1 cells ectopically expressing PPAR γ 2 under different culture conditions. Rosi addition allows efficient differentiation in the absence of MED1 as long as PPAR γ 2 is expressed. **F)** Summary of the *in vitro* cell culture experiments under conventional culture conditions without the use of Rosiglitazone. XX and X indicate strong and weak blocks, respectively, with loss of MED1.