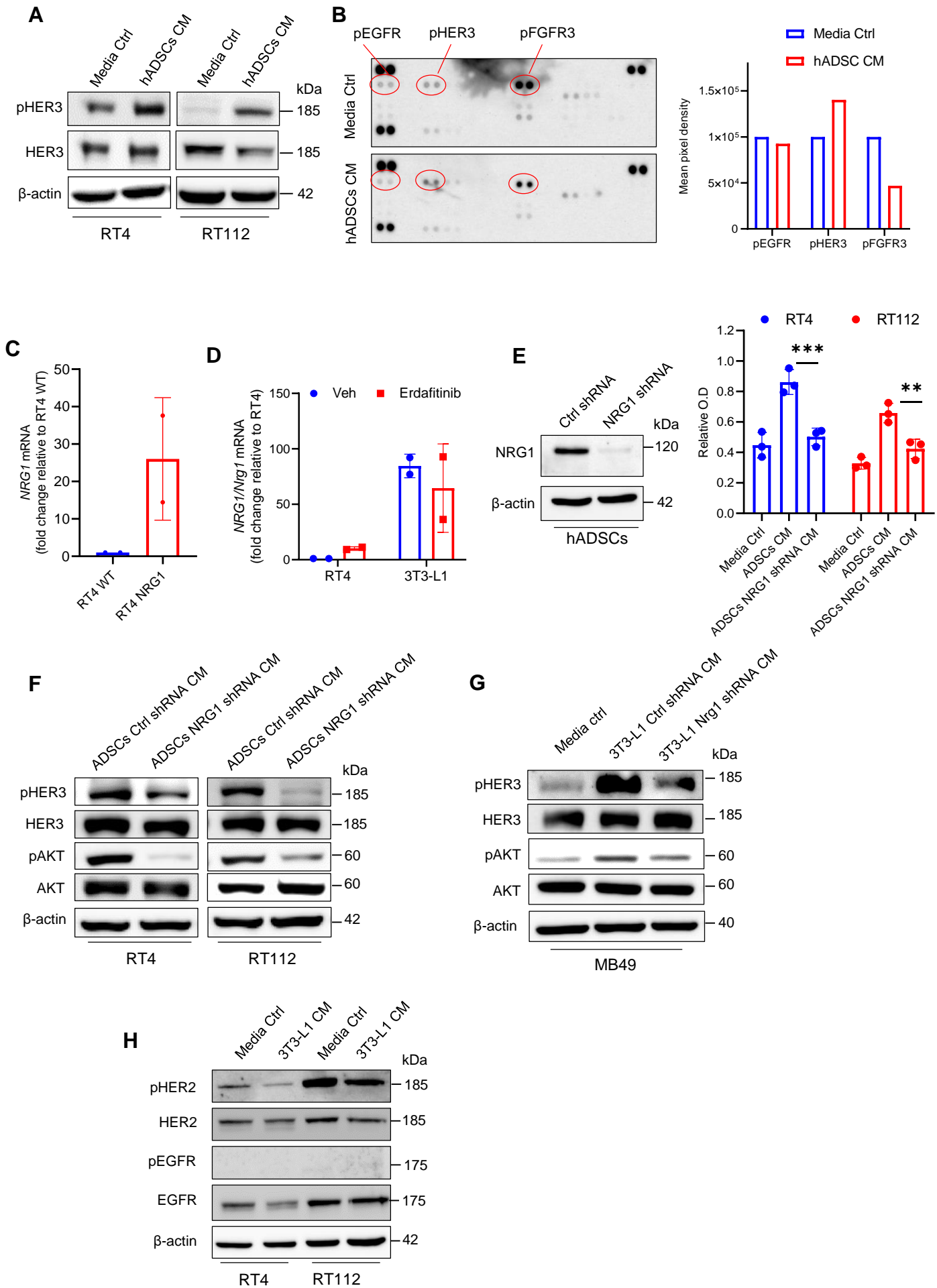


# Supplementary Figure S2.



**Supplementary Figure S2: NRG1 is secreted from pre-adipocytes and induces erdafitinib resistance in RT4 and RT112 bladder cancer cells by activating HER3.** (A) Western blot analysis of (p)HER3 in RT4 and RT112 cells treated with media control (ctrl) or conditioned media (CM) of human adipose-derived stem cells (hADSCs) for 16 hr.  $\beta$ -actin serves as a loading control. (B) Left: Receptor tyrosine kinase (RTK) array on RT4 cells cultured in media ctrl or CM of hADSCs for 16 hr. Right: Quantification of mean pixel density of the RTK array shown on the left. Data are represented as mean of the duplicate dots per kinase. (C) qRT-PCR analysis of *NRG1* in RT WT and RT4 *NRG1* cells. Expression levels are normalized to *GAPDH*. Data are represented as mean  $\pm$  SD. (D) qRT-PCR analysis of *NRG1* in RT4 and 3T3-L1 cells treated overnight with Vehicle (Veh) or 10 nM erdafitinib. Expression levels are normalized to *GAPDH*. Data are represented as mean  $\pm$  SD. (E) Left: Western blot analysis of *NRG1* in hADSCs transduced with lentivirus encoding ctrl shRNA or *NRG1* shRNA.  $\beta$ -actin serves as a loading control. Right: Proliferation analysis of RT4 and RT112 cells treated with 10 nM erdafitinib in media ctrl or CM of hADSCs ctrl/*NRG1* shRNA. Crystal violet staining was performed on day 7. Data are normalized to cells treated with vehicle. "O.D": optical density. Three biological replicates were performed. Data are represented as mean  $\pm$  SD. Two-way ANOVA was used for statistical analysis. \*\*\* $p < 0.001$ , \*\* $p < 0.01$ . (F) Western blot analysis of (p)HER3 and (p)AKT in RT4 and RT112 cells treated with CM of hADSCs ctrl/*NRG1* shRNA for 16 hr.  $\beta$ -actin serves as a loading control. (G) Western blot analysis of (p)HER3 and (p)AKT in MB49 cells treated with media ctrl or CM of 3T3-L1 ctrl/*Nrg1* shRNA for 16 hr.  $\beta$ -actin serves as a loading control. (H) Western blot analysis of (p)HER2 and (p)EGFR in RT4 and RT112 cells treated with CM of 3T3-L1 cells or media ctrl for 16 hr.  $\beta$ -actin serves as a loading control.