Supplemental Figures:

Figure 1: *Vav3 and AR3 (AR-V7) are critical for proliferation of CRPC cells*

CWR-R1 cells stably expressing either plk0.1-shGFP, -shVav3, -shAR3 or -shAR were plated at 20,000 cells/well in 24 well dishes in 10% FBS. Cells were harvested and counted on three separate days. A representative experiment of three experiments was performed in sextuplicate and data are plotted as cell number (thousands) ± SEM.

Figure 2: *Vav3 and AR3 (AR-V7) drive ligand-independent AR activity in CRPC*

A, 22Rv1 or B, CWR-R1 cells stably expressing either shGFP, shVav3, or shAR3 were transfected with reporter plasmid PSA-Luc. Luciferase activity was determined 48 h after transfection. Data represent three experiments performed in triplicate and are plotted (fold induction ± SEM) with shGFP set as one.

Figure 3: *Vav3 enhances AR3 (AR-V7) transcriptional activity in a PH domain independent manner*
AR negative human prostate cancer cell line PC3 was transfected with AR or AR3, reporter plasmid ARE-Luc, a Vav3 mutant lacking the PH domain (ΔPH) or equivalent amounts of the corresponding empty vector (EV). Cells were treated with either vehicle (−) or 1nm R1881 (+). Luciferase activity was determined 48 h after transfection. Data represent one of three experiments performed in triplicate plotting the mean RLU/protein ± SEM. Significance was determined using a two-tailed Student's *t* test (*, $p<.05$).

Figure 4: *Vav3 interacts with AR3 (AR-V7) but poorly with wild type AR*
HEK293 cells were transfected with Vav3-myc and AR or AR3 and harvested 48 hours later. Co-immunoprecipitation was performed as described in *Materials and Methods* using antibodies to mouse (M), rabbit (R), IgG (control), myc, or AR (N-terminal AR, AR-N20). Equivalent volumes were immunoblotted and probed for N-terminal AR and myc. A representative blot of four independent experiments is shown.

Figure 5: *Vav3 regulates nuclear levels of AR3 (AR-V7)*

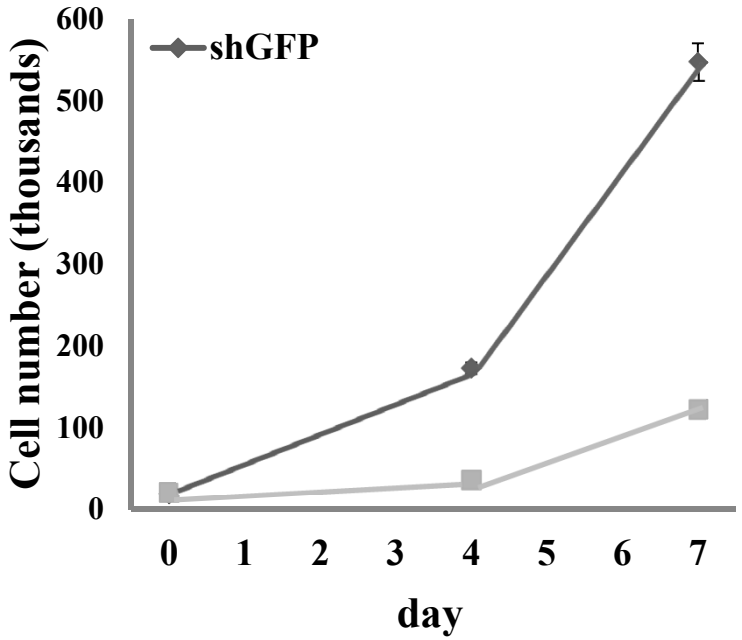
22Rv1 cells stably expressing tet-shGFP or tet-shVav3 were grown in 5% CSS ± 1ug doxycycline for 72 hours. A, Equivalent amounts of total cellular protein were immunoblotted for N-terminal AR and actin. B, Cells were fractionated and immunoblotted for N-terminal AR, histone and SOD as described in *Materials & Methods*. Protein levels were normalized to SOD (cytoplasmic fraction), histone (nuclear fraction) or actin (input). Protein levels were determined from three independent experiments and represent the ratios of doxycycline to untreated cells and are compared to the tet-shGFP treated/untreated values (set to one). Significance was determined using a two-tailed Student's *t* test comparing (treated/untreated) of the experimental group to (treated/untreated) tet-shGFP. C, 22Rv1 cells stably expressing Vav3-FLAG or FLAG were grown in 5% CSS for 72 hours. Equivalent amounts of total cellular protein were immunoblotted for N-terminal AR and actin. D, Nuclear and cytosolic fractions

from the same cells were immunoblotted for N-terminal AR, Histone, and SOD. Densitometry was performed on blots from three independent experiments.

Figure 6: *Vav3 regulates AR3 recruitment to UBE2C enhancer 2*
22Rv1 cells stably expressing shGFP, shAR3, or shVav3 were grown in 5% CSS phenol red free media for 72 hours. Chromatin immunoprecipitation was performed as described in Materials and Methods. Data represent three independent experiments and are plotted as (% input relative to IgG control ± SEM).

Supplemental Figure 1

A



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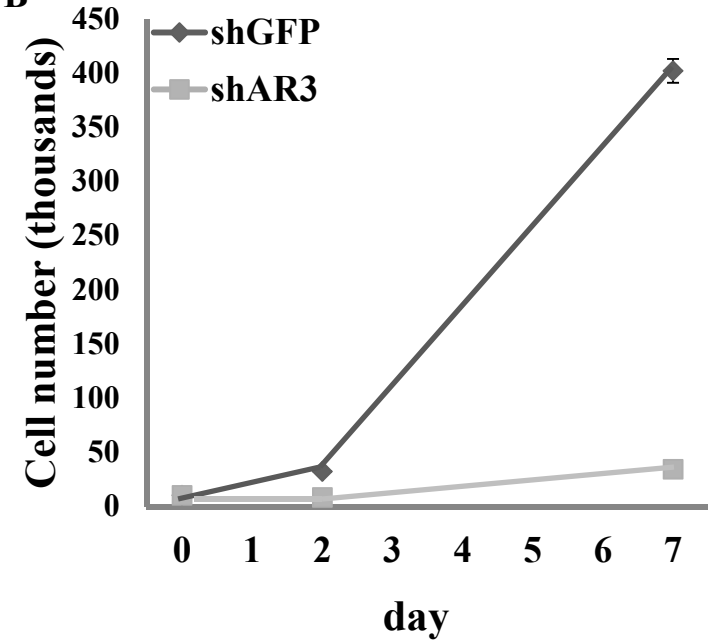

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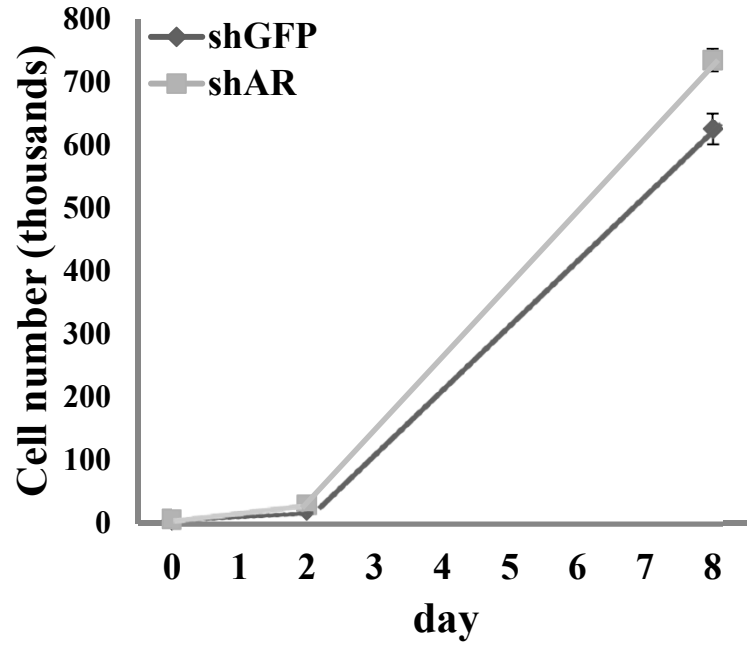
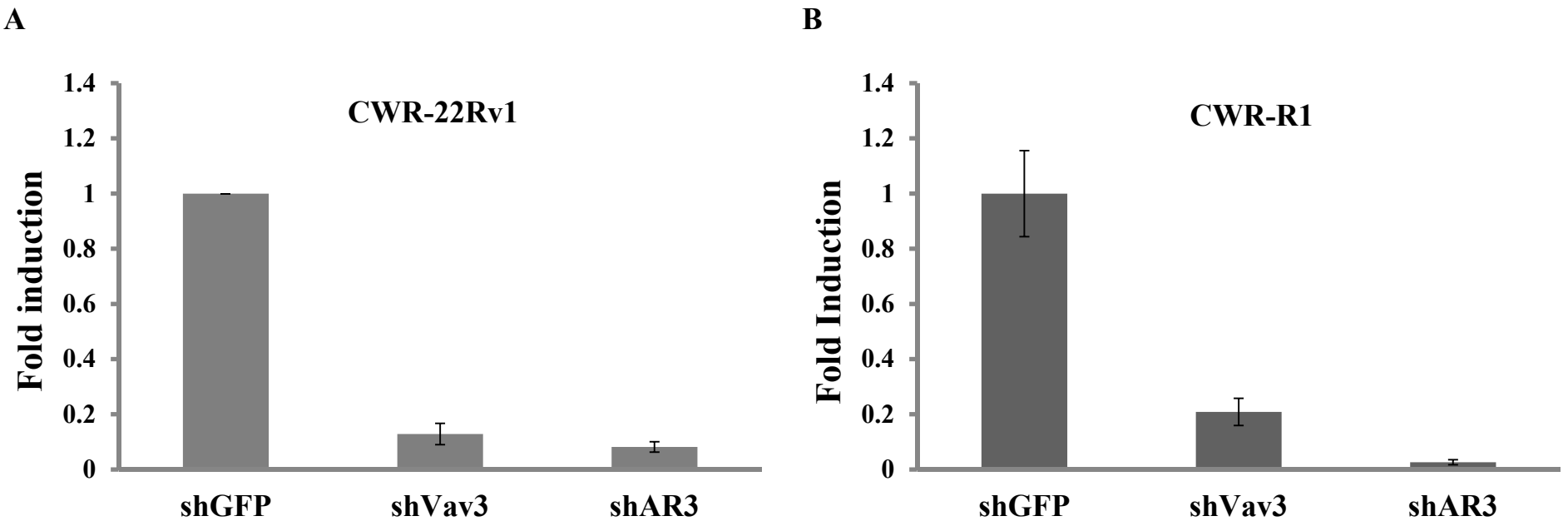
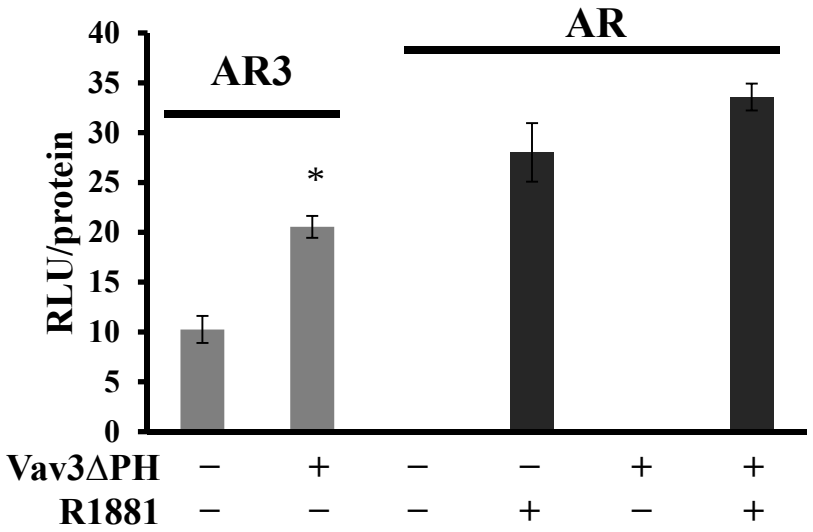


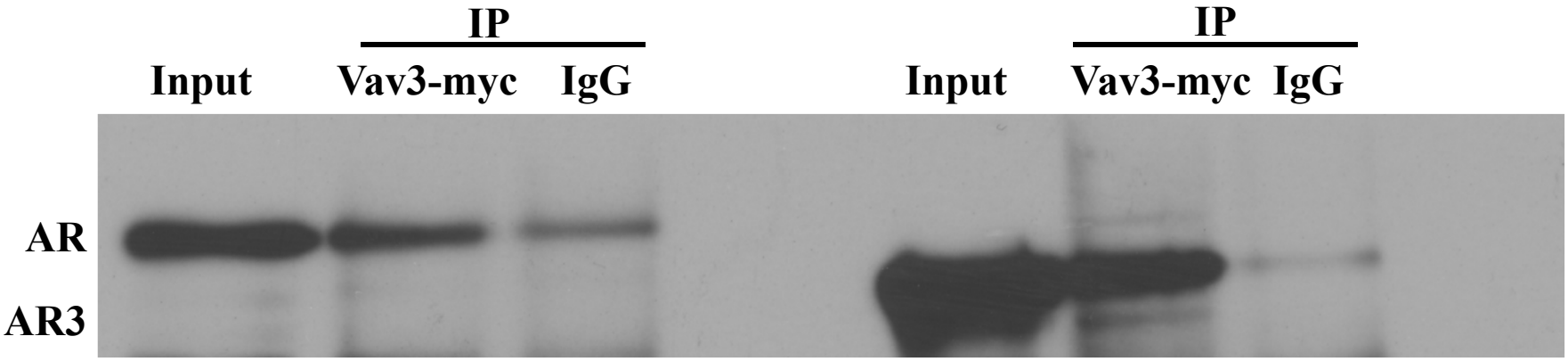
Supplemental Figure 2



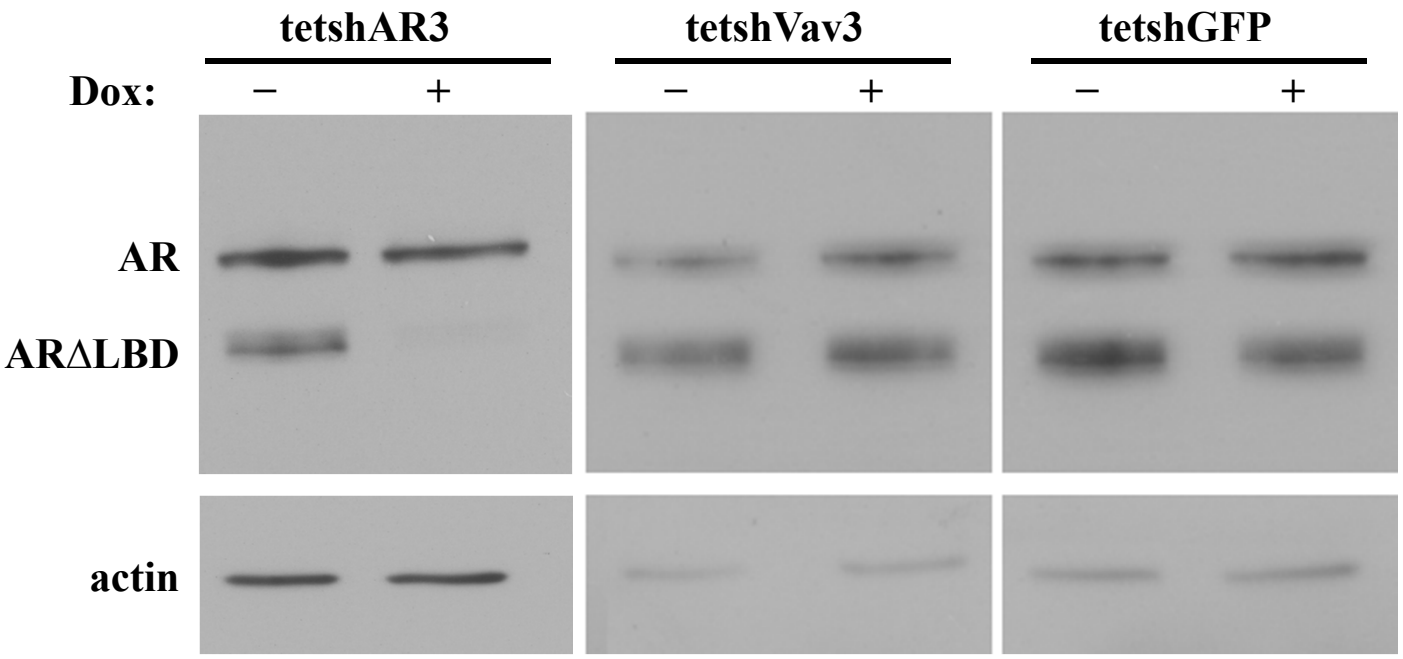
Supplemental Figure 3

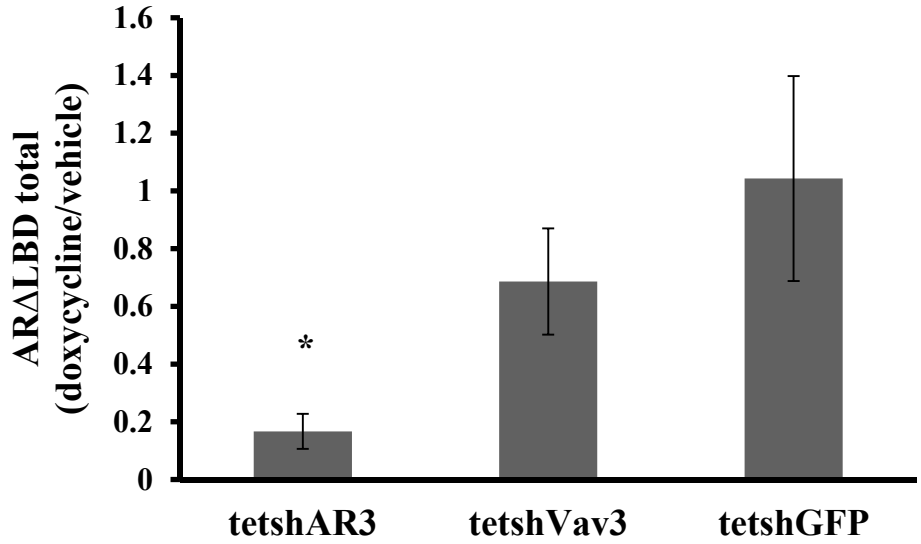


Supplemental Figure 4

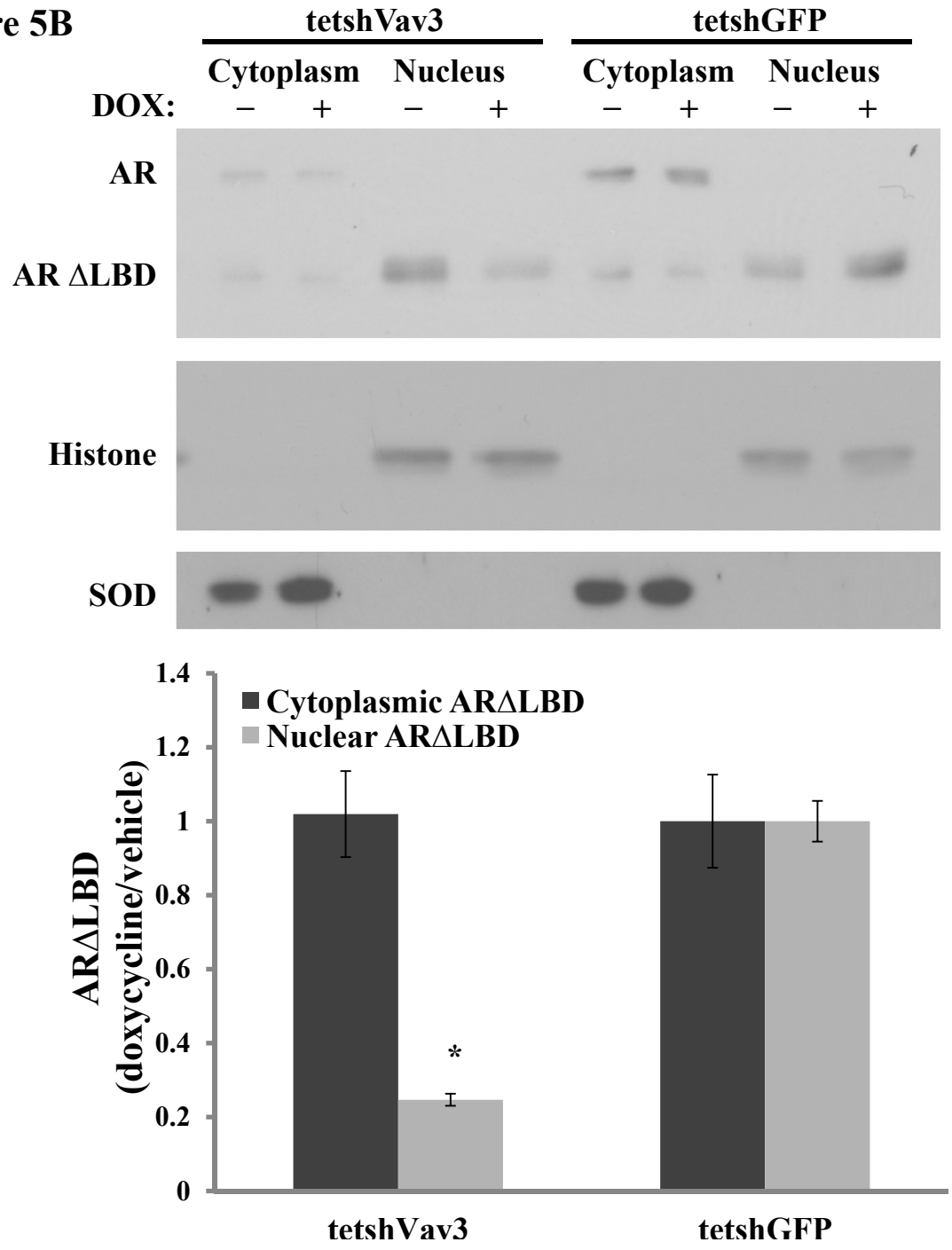


Supplemental Figure 5A

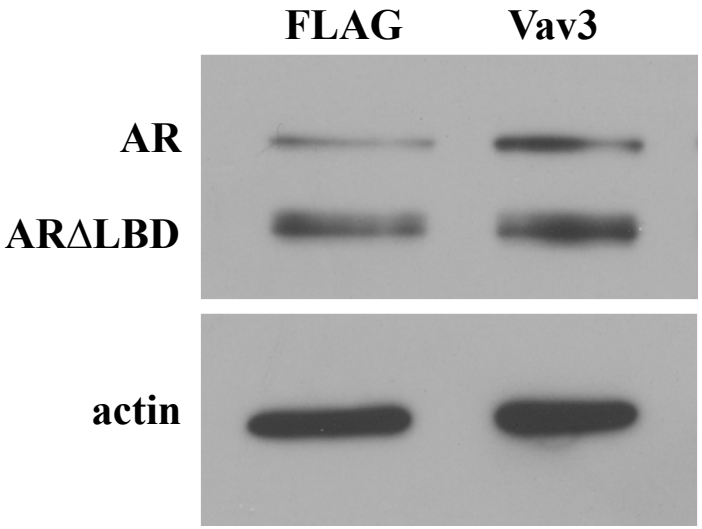


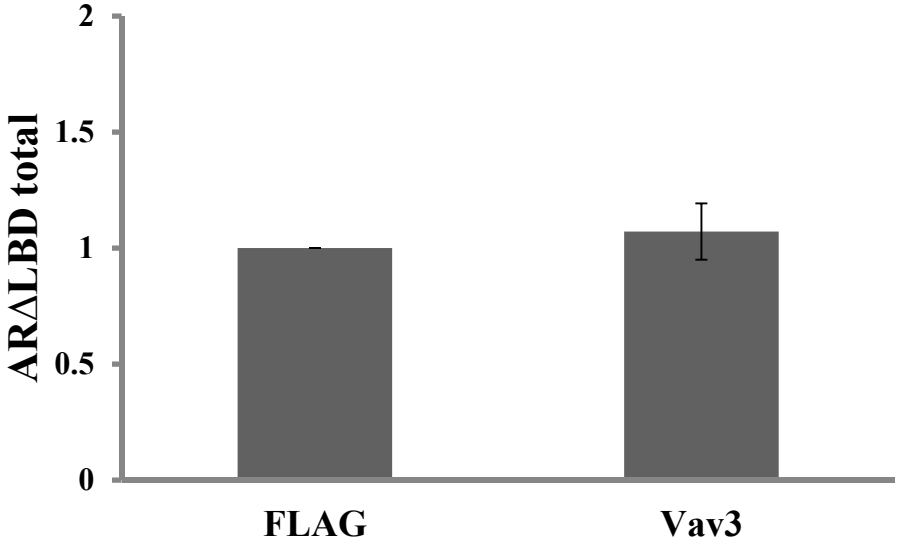


Supplemental Figure 5B

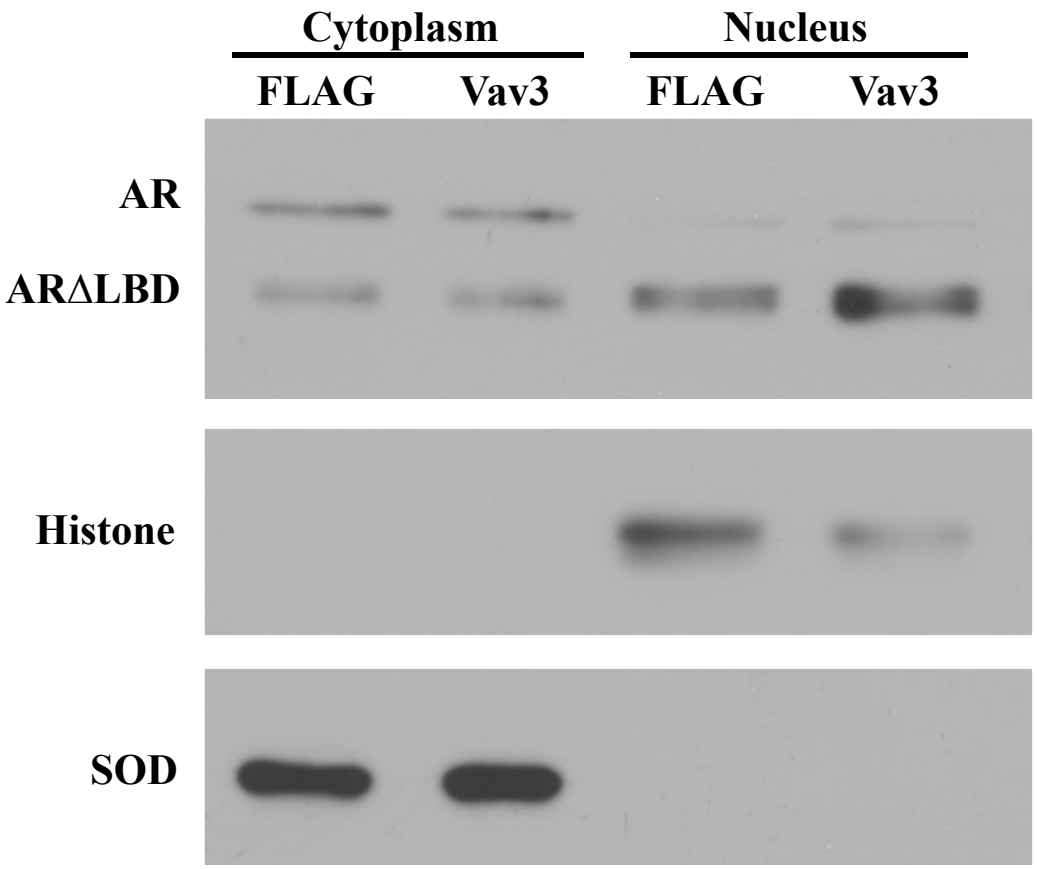


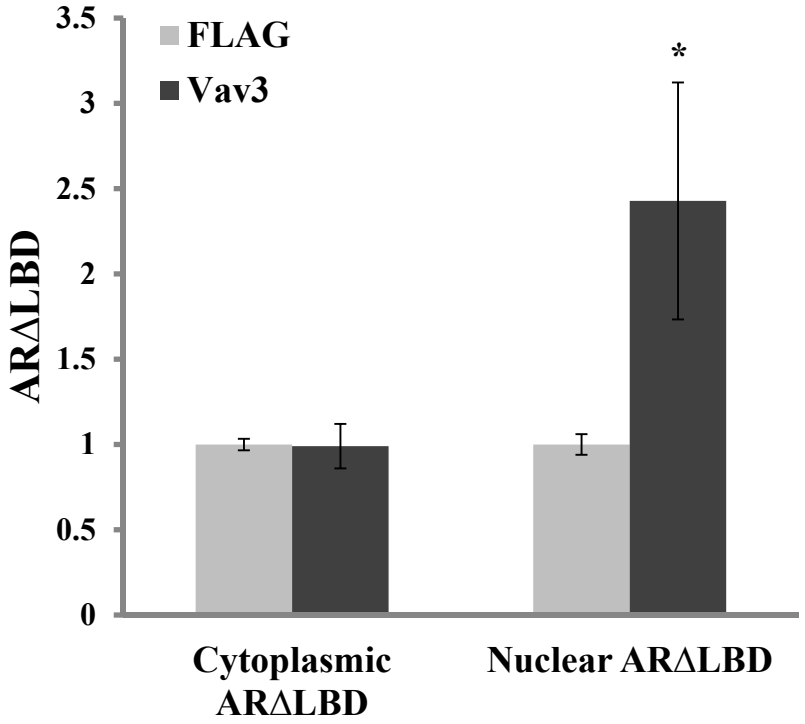
Supplemental Figure 5C





Supplemental Figure 5D





Supplemental Figure 6