

Supplemental Figure 1: Expression of Vav3 DH domain inhibits growth of AR-expressing cells only, and does not decrease total levels of FL-AR, AR-V7, Vav3 or Vav2.

A) PC3 cells were transfected with Vav3 or Vav3 Δ DH, harvested, and immunoblotted using anti-Vav3 antibody. B) CWR-R1 and the AR-null ALVA 31 cell lines stably expressing DH-FLAG or control vector were harvested and immunoblotted with anti-FLAG-antibody. C) 22Rv1, CWR-R1 or ALVA31 cells stably expressing DH-FLAG or EV control (EV-FLAG) were harvested and counted at three distinct time points using a hemocytometer. Data represent three independent experiments performed in triplicate plotted as cell number (thousand) \pm SD. D) PC3 cells stably expressing DH-FLAG or EV control (EV-FLAG) were harvested and counted using a hemocytometer. Data represent two independent experiments performed in triplicate and normalized to the EV control. E) 22Rv1 stably expressing DH-FLAG or EV control were immunoblotted using FL-AR or AR-V7 antibody, and actin as a control. F) HEK293 cells were transfected with AR-V7, Vav3 or empty vector, and DH-FLAG or EV control; and were subjected to immunoblotting using anti-Vav3, anti-AR-V7, and anti-actin antibodies. G) 22Rv1 stably expressing DH-FLAG or EV control were immunoblotted using anti-Vav2 and anti-actin antibodies.

Supplemental Figure 2: Disruption of coactivator interactions with FL-AR or AR-V7 increases cell death through apoptosis.

A) 22Rv1, CWR-R1, or ALVA31 cells stably expressing DH-FLAG or EV control (EV-FLAG) were plated at 20,000 cells/well in 24 well dishes. Adherent and floating cells were harvested at one time point and cells that stained with trypan blue or that excluded stain were counted. Data represent three independent experiments performed in triplicate. For 22Rv1 and CWR-R1: Student's T-test, p value < 0.01; for ALVA31: Student's T-test, p value > 0.5). B) Lysates from the cell lines in panel A were immunoblotted with antibodies anti-cleaved PARP or actin as a loading control. (** p value < 0.01)