

## **SUPPLEMENTARY FIGURE LEGENDS:**

**Supplemental figure S1. Disulfiram induces prostate cancer cell apoptosis.** VCaP cells were incubated with vehicle, DSF, copper alone or DSF in combination with copper. The effects of the copper chelation (BCS) as well as the antioxidants (NAC and LGR) were evaluated. Following 24 hr. drug treatments cells were harvested, stained with Annexin V-Alexa Fluor® 488 and Sytox red dead cells staining, and analyzed by flow cytometry. Percentage of cell population stained with Annexin V and/or Sytox under different treatments is represented.

**Supplemental figure S2. The effect of Disulfiram on prostate cancer cell apoptosis is more pronounced at 72hr.** VCaP cells were incubated with either vehicle or DSF for 72 hr. The effect of the copper chelation (BCS) was also evaluated. Cells were then harvested, stained with Annexin V-Alexa Fluor® 488 and Sytox red dead cells staining, and analyzed by flow cytometry. The populations of Annexin V<sup>-</sup>/Sytox<sup>-</sup>, Annexin V<sup>+</sup>/Sytox<sup>-</sup>, and Annexin V<sup>+</sup>/Sytox<sup>+</sup> correspond to live cells, early apoptotic cells, and late apoptotic cells respectively. Representative plots of one set of triplicate experiments are presented.

**Supplemental figure S3. Disulfiram induces 22RV1 prostate cancer cell apoptosis.** 22RV1 cells were incubated with vehicle, DSF, copper alone or DSF in combination with copper. The effects of the copper chelation (BCS) as well as the antioxidants (NAC and LGR) were evaluated. 24 hr. later, cells were harvested, stained with Annexin V-Alexa Fluor® 488 and Sytox red dead cells staining, and analyzed by flow cytometry. The populations of Annexin V<sup>-</sup>/Sytox<sup>-</sup>, Annexin V<sup>+</sup>/Sytox<sup>-</sup>, and Annexin V<sup>+</sup>/Sytox<sup>+</sup> correspond to live cells, early apoptotic cells, and late apoptotic cells respectively. Representative plots of one set of triplicate experiments are presented.

**Supplemental figure S4.** VCaP cells were pretreated for 1 hr. with either vehicle or 0.1 µg/ml cycloheximide followed by vehicle (Veh) or 10 nM R1881 for 12 hr. and the mRNA expression of CTR1, ATP7B, STEAP4 or INSIG1 were evaluated using qPCR. Results as fold induction over vehicle-treated cells ±SE (n=3).

**Supplemental figure S5.** RWPE-1 Empty Vector control (RWPE-1 EV) or RWPE-1 overexpressing AR (RWPE-1 AR) cells were treated overnight with either vehicle or increasing concentration of R1881 and the mRNA expressions of CTR1 (**A**) and ATP7B (**B**) using qPCR were evaluated. RWPE-1 EV and AR cells (**C**) or LNCaP cells (**D**) were treated overnight with either vehicle or 10 nM R1881 and their whole-cell extracts were subjected to Western blot analysis. Blots were probed for AR, CTR1 or GAPDH, loading control. **E.** LNCaP control (LNCaP EV) or LNCaP overexpressing AR (LNCaP AR) cells were treated overnight with either vehicle or increasing concentration of R1881 and the mRNA expressions of ATP7B using qPCR was evaluated.

**Supplemental figure S6.** VCaP Cells were incubated with 5 $\mu$ Ci/ml  $^{64}$ CuCl<sub>2</sub> for 30-120 min at 37°C in the presence of either vehicle or 1 $\mu$ M DSF.  $^{64}$ Cu uptake was quantified and normalized to protein concentrations of cells lysates.