

| Compound | Precursor Ion m/z (Ion Type) | Fragment Ion <i>m/</i> z Values |
|--------------------------------------|--|------------------------------------|
| $T(^{13}C_3)$ | 292.2263 [M+H] ⁺ | 100.0749, 112.0749 |
| DHT (¹³ C ₃) | 294.2419 [M+H] ⁺ | 159.1168, 218.1895, 258.2208 |
| DHEA (D_6) | 277.2433 [M-H ₂ O+H] ⁺ | 219.2014, 259.2327 |
| F (D ₄) | 367.2417 [M+H]+ | 97.0649, 121.0648 |
| Preg $({}^{13}C_2D_2)$ | 303.2562 [M-H ₂ O+H] ⁺ | 161.1325, 285.2456 |
| Prog $({}^{13}C_3)$ | 318.2419 [M+H]+ | 100.0749, 112.0749 |
| AED (¹³ C ₃) | 290.2106 [M+H]+ | 100.0749, 112.0749 |
| A5-diol (D ₃) | 276.2401 [M-H ₂ O+H] ⁺ | 159.1169, 258.2208 |
| DOC (D ₈) | 339.2770 [M+H] ⁺ | 100.0836, 113.0899 |

Supplemental Figure 2, Related to Figure 2. A) Immune repertoire of orthotopic PTE-82 tumors following treatment with Lupron depot or aCSF-1. 17 days post implant, tumors were excised and single cell suspensions were processed for flow cytometry. B) Gene set enrichment analysis (GSEA) following whole tumor RNA sequencing. The normalized enrichment score is shown comparing tumors from mice treated with Lupron versus an IgG control. Data reflects 3 tumors per group from a single experiment. C) Volcano plots of gene expression changes between aCSF-1 or Lupron and the IgG control. The decreases in Tmprss2, Abca1, and Cd36 expression are highlighted. Significant changes shared between the α CSF-1 and Lupron groups are shown in a Venn diagram to the right. D) Example extracted ion chromatograms for 9 stable isotope-labeled standards for androgens. Each ion chromatogram is labeled with the abbreviation for the androgen and the description of the stable isotopes incorporated into the standards (i.e. Carbon-13 or Deuterium). These data from parallel reaction monitoring of each precursor ion indicate the sum of ion signal intensity for the fragment ions listed in the table (lower right). E) Steroid levels including pregnenolone, progesterone, androstenedione, DHEA, 5-androstenediol, testosterone and DHT in fresh tumor tissues from each individual mouse of Fig. 2E. n=5-6 mice per group, from one of two independent experiments. Measurements were normalized by tissue weight. Significance determined by unpaired t test for DHT. *p<0.05.