Figure S1. Meta-analysis of AR cistromes in LNCaP-1F5 and VCaP cells and FoxA1 cistromes in LNCaP-1F5 and LNCaP cells.

(A) Overlap analysis of the DHT-AR cistrome in LNCaP-1F5 cells (FDR <2%) vs. the DHT-AR cistromes in LNCaP cells by ChIP-on-chip (Wang et al, 2009) and the R1881-AR cistrome by ChIP-seq (Massie et al, 2011).

(B) Overlap of the FoxA1 cistrome in LNCaP-1F5 cells (2 replicates, FDR <2%, Sahu et al, 2011) with the FoxA1 cistromes in LNCaP cells by ChIP-seq (Tan et al, 2012, and Robinson et al, 2011).

(C) Overlap of the DHT-AR cistrome (FDR <2%) with the R1881-AR cistrome in VCaP cells (Massie et al, 2011), both analyzed by ChIP-seq.
Figure S2. AR-binding events and transcript levels in LNCaP-1F5 cells with exposed to different ligands. (A) Distribution of ARBs in response to DHT, CPA or RU486. (B) Relative levels of selected mRNAs in presence of 100 nM DHT, 1 µM CPA or 1 µM RU486 in LNCaP-1F5 cells treated for 24 h and measured by qRT-PCR. Mean+S.E.M. values are shown for three biological replicates. (C) Top scoring cis-elements revealed by de novo motif analyses for the ARBs mapped to genes up-regulated by DHT, CPA or RU486. (D) Top scoring cis-element by de novo motif search for the ARBs mapped to genes down-regulated by DHT.
Figure S3. (A) Histogram showing the relative distance between AR- and FoxA1-binding peak summits for the ARBs unique to LNCaP-1F5 cells, with the median distance of 36 nt being shown by the red line. Previously published FoxA1 ChIP-seq data (Sahu et al., 2011) have been used for this analysis. (B) Immunoblot on FoxA1 protein levels in LNCaP-1F5 and VCaP cells. (C) Immunoblot on AR protein levels in LNCaP-1F5 and VCaP cells.
Figure S4. AR-binding sites unique to LNCaP-1F5 cells vanish upon FoxA1 depletion and are dependent on prior FoxA1 binding. (A) Depletion of FoxA1 in LNCaP-1F5 cells treated for 72 h with FoxA1-specific siRNA (siFoxA1) or control siRNA (parental). (B) Six selected ARBs unique to LNCaP-1F5 cells (sites LU1–LU6) were examined by quantitative ChIP assays in parental and FoxA1-depleted LNCaP-1F5 cells. (C) Binding of FoxA1 to the LU1–LU6 sites in parental and FoxA1-depleted LNCaP-1F5 cells prior to and after a 2-h DHT exposure, as examined by direct ChIP assays. (D) Comparison of AR binding to the LU1–LU10 sites in LNCaP-1F5 and VCaP cells before and after DHT exposure for 2 h. (E) FoxA1 binding to the LU1–LU10 sites in VCaP cells in comparison to that in LNCaP-1F5 cells. The results in panels B–E are expressed as percent of input and represent mean±SEM values from two biological replicate experiments.
Figure S5. (A) Enrichment of selected AR-binding events unique to LNCaP-1F5 cells by FAIRE analysis in LNCaP-1F5 and VCaP cells. Mean±SEM values are shown for three biological replicates (*p < 0.001). (B, C) Direct ChIP assays for H3K4me2 and the histone H2A.Z levels in LNCaP-1F5 and VCaP cells at selected AR-binding sites unique to LNCaP-1F5 cells.
Figure S6. Snapshots of selected gene loci (TSHR, CRAMP1L, GDAP2, SFT2D2, RPL32, SERINC5, PPP2R2A, and NIPSNAP3B) in which depletion of the central nucleosome, marked by reduced H3K4me2 signal, is associated with concomitant AR- and FoxA1-binding events in LNCaP-1F5 cells, but there is no AR binding at the corresponding loci in VCaP cells. The data on FoxA1 and H3K4me2 marks are from Sahu et al. (12).
Figure S7. AR and GR loading onto selected chromatin loci in LNCaP-1F5 cells after exposure to 100 nM DHT or 100 nM Dex for 2 h. Direct ChIP assays were performed using receptor-specific antibodies (α-AR and α-GR, respectively), and PSA and FKBP5 enhancer and PGC promoter regions as the loci for receptor binding. Mean ± S.E.M. values for three biological replicate experiments are shown.
Figure S8. (A) Examples of androgen- and glucocorticoid-regulated genes with shared ARBs and GRBs and the presence of RNA Pol II over the transcription units of three genes of the kallikrein cluster (KLK3, KLK2, and KLKP1), FKBP5, and ELL2. (B) Location of GRBs and ARBs, and the presence of RNA Pol II over the transcription units of the PER1, RHOB and CST3 genes that are regulated by glucocorticoid only. The numbers in red and blue within brackets refer to fold increases in transcript levels after a 24-h exposure to 100 nM DHT or 100 nM Dex, as measured by microarray in three separate experiments and shown on log₂ scale.
Figure S9. Snapshots of representative LNCaP-1F5 cell loci showing AR- and GR-binding events and RNA Pol II occupancy for genes (SOX4, C1orf116, and RASSF3) that are up-regulated only by androgen. Fold increases in mRNA accumulation, as determined by microarray experiments, are shown in parentheses for DHT in red and for Dex in blue.
Figure S10. Examples of Dex-upregulated genes in LNCaP-1F5 cells that are over-expressed in GR+ prostate cancer.
(from Kilpinen et al. Systematic bioinformatic analysis of expression levels of 17,330 human genes across 9,783 samples from 175 types of healthy and pathological tissues. Genome Biol 2008;9:R139.)
Figure S11. Ten selected GR-binding sites (VU1–VU10) unique to VCaP cells show reduced GR binding upon FoxA1 depletion, as examined by quantitative ChIP assays in parental (siControl) and FoxA1-depleted VCaP cells. (A) Binding of FoxA1 to the VU1–VU10 sites in parental and FoxA1-depleted VCaP cells after a 2-h Dex exposure. (B) Binding of GR to the VU1–VU10 sites in parental and FoxA1-depleted VCaP cells after a 2-h Dex exposure. (C) The extent of FoxA1 depletion in VCaP cells treated for 72 h with siRNA specific for FoxA1 mRNA (siFoxA1) or control siRNA (parental), as analyzed by immunoblotting.
**Figure S12.** Examples of two gene loci (*EBF4* and *CCDC3*) showing unique GR-binding events only in VCaP cells and two loci (*CROCC* and *TET3*) depicting GR-binding events shared by LNCaP-1F5 and VCaP cells. There is no AR binding at the corresponding loci in either LNCaP-1F5 or VCaP cells.
Figure S13. Fourteen selected GR-binding sites (LG1–LG14) unique to LNCaP-1F5 cells identified by ChIP-seq by using a monoclonal antibody against rat GR (see Fig. 3) were validated by quantitative ChIP assays using another GR antibody, a monoclonal antibody against human GR (BuGR2), in LNCaP-1F5 vs. VCaP cells. The BuGR antibody was included in the antibody cocktail used for ChIP-seq analysis of GR-binding events in VCaP cells. The cells were exposed to 100 nM Dex for 2 h prior to ChIP. Mean ± SEM values are shown for two biological replicates.