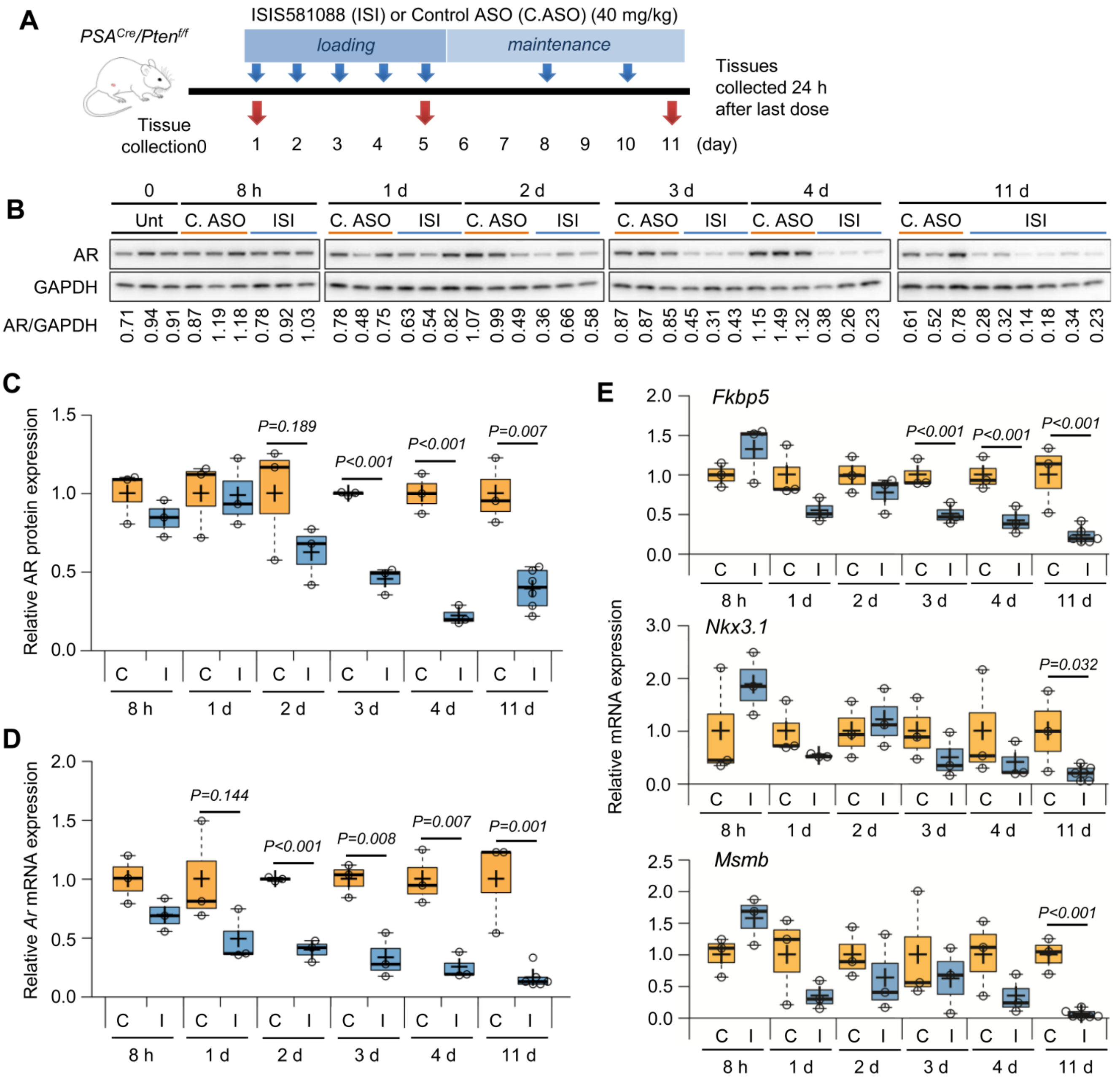
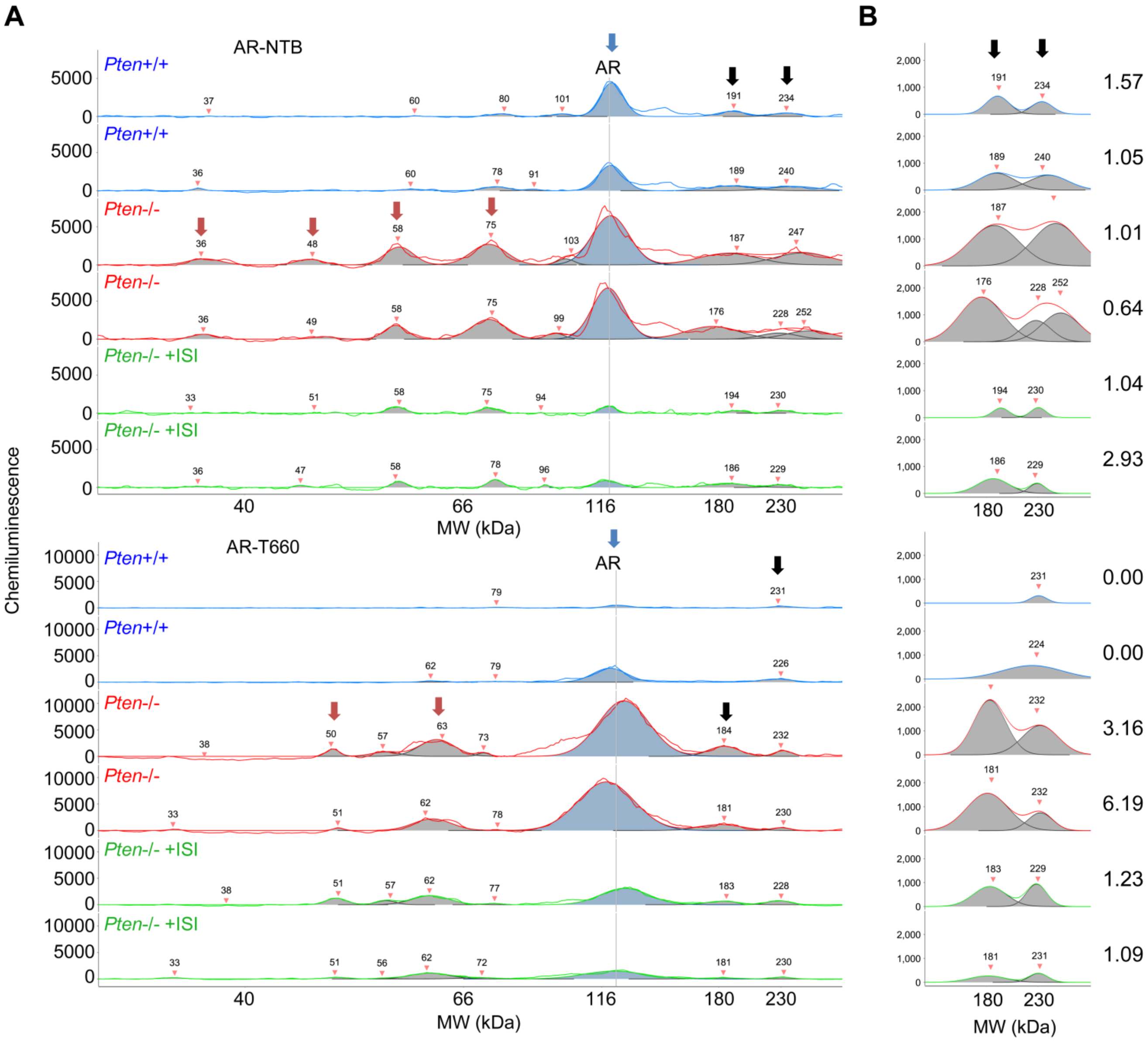
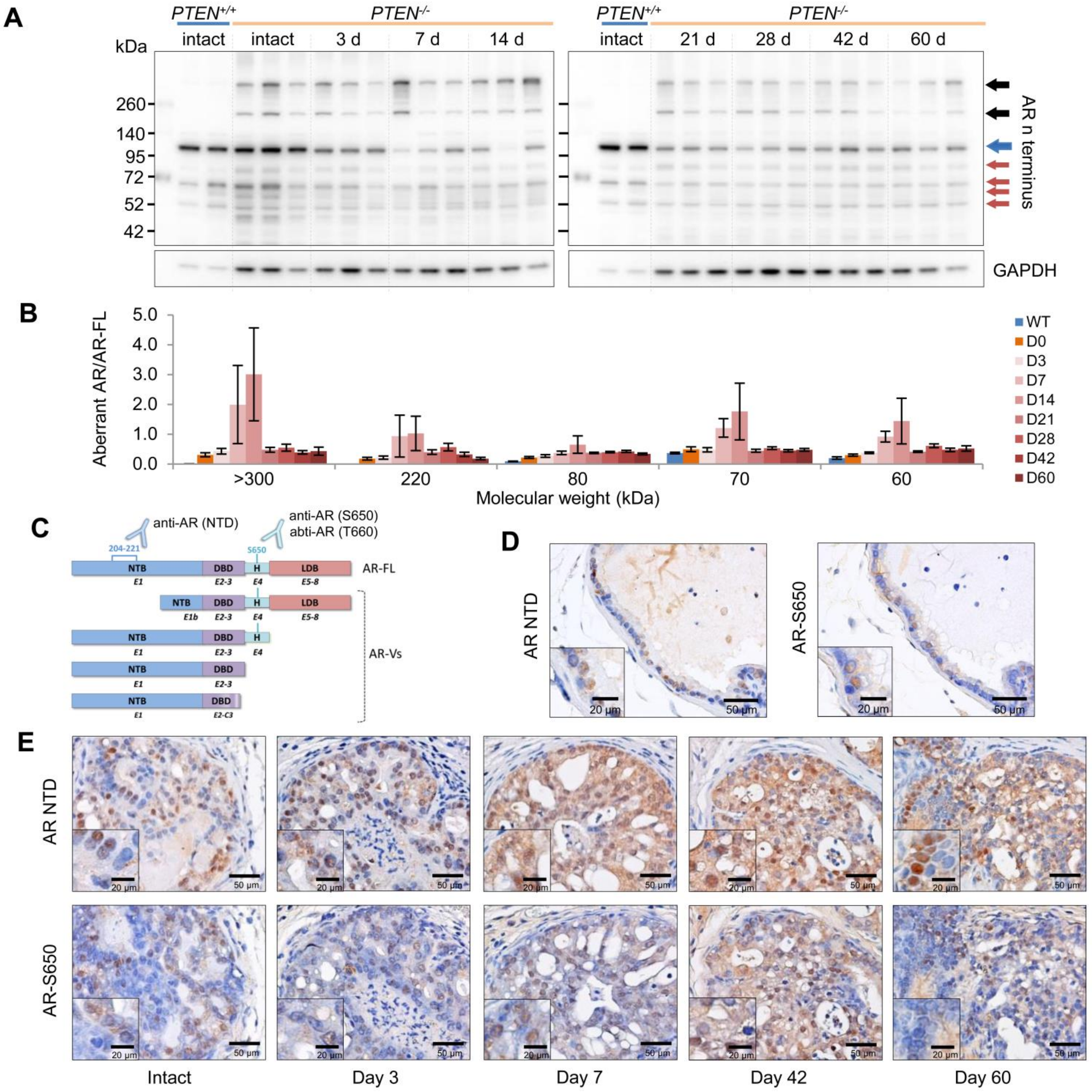


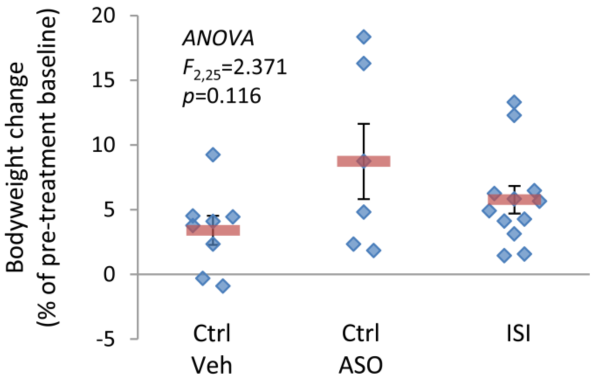
Supplemental Figure 1. Pharmacodynamic activity of ISIS581088 in mouse prostate tumors. (**A**) Schematic for ASO dosing and tissue collection. (**B**) AR protein expression analysis by western blot and relative density quantified with ImageJ. Comparison of western blot AR protein (**C**) and qRT-PCR Ar mRNA levels (**D**) and AR target genes (**E**) in prostate tumors after treatment with control ASO (C) or ISIS581088 (I). Center lines represent medians; box limits indicate the 25th and 75th percentiles; crosses represent sample means; data points are plotted as open circles, n=3-6 mice/group. Significance represents Student's t-test for comparisons between control ASO and ISIS581088.



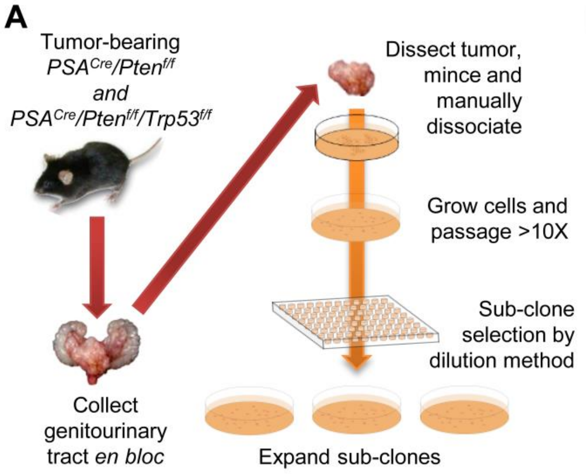
Supplemental Figure 2. Expression profiles AR protein mouse prostate. (**A**) Individual electropherograms of AR isoforms corresponding to Figure 2A-B by automated capillary electrophoresis using AR antibodies targeting the N-terminal domain (AR-NTB) or the ligand binding domain (AR-LBD). AR-FL, putative AR-Vs and high molecular weight bands are represented by blue, red and black arrows, respectively. Shaded areas in the electropherogram denote peaks after baseline correction. (**B**) Enhanced view of individual electropherograms of high molecular AR peaks showing fit baseline corrected peaks and the ratio of AUC for 180/230 kDa peaks.



Supplemental Figure 3. Expression of full length AR and putative splice variants in mouse *Pten*-deficient prostate tumors. (**A**) Analysis of full-length AR and aberrant AR in mouse prostate tumors by western blot analysis using an AR antibody targeting the n-terminal domain (AR-NTD). Prostates tumors were collected from intact 25-week-old conditional *Pten*-KO mice and following surgical castration as indicated. Normal prostate tissue from age 20-week-old wildtype *Pten* mice was used as a normal reference and GAPDH was used as loading control. The blue arrow indicated AR-FL and putative AR-Vs are marked with red arrows and high molecular weight bands with black arrows. (**B**) Aberrant AR/AR-FL ratio from densitometric analysis of the immunoblots in (**A**). (**C**) Schematic for immune detection of AR-FL and putative AR-Vs by immunohistochemistry. AR-NTD antibody detects AR-FL and AR-Vs, while AR-650 detects AR-FL but not truncated AR-Vs lacking the hinge region and ligand binding domain (LBD). Representative photomicrographs of serial sections normal mouse prostate (**D**) and intact prostate tumors and prostate tumors post-surgical castration (**E**) immunostained with AR-NTD and AR-S650 antibodies. Scale bars represent 50 µm.

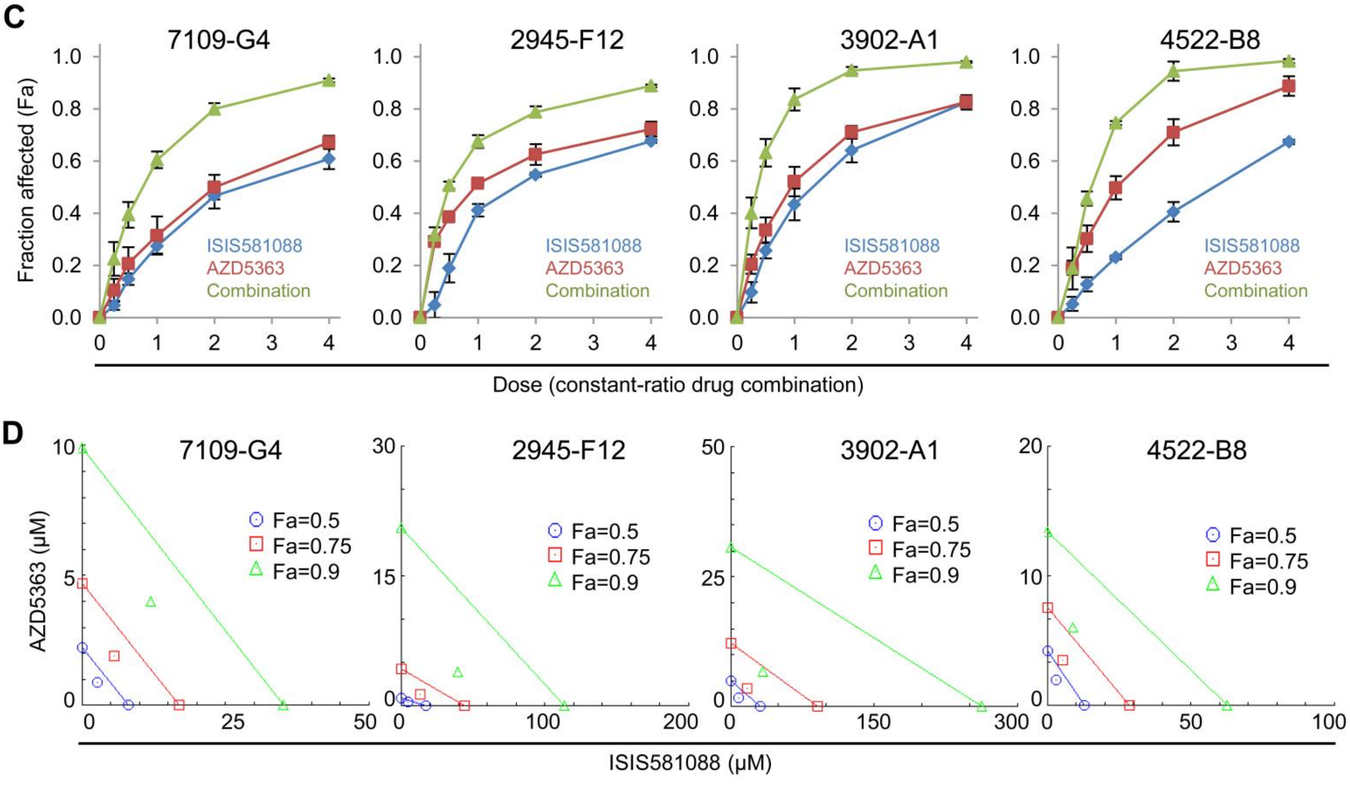


Supplemental Figure 4. Safety and tolerability of *Ar*-ASO administration in conditional *Pten*-KO mice. Mouse bodyweight changes after treatment with vehicle (*n*=8), control ASO (*n*=6) or ISIS581088 (*n*=12). Control ASO and ISIS581088 were administered i.p. for four weeks (40 mg/kg/d for first week loading followed by three weeks of maintenance dosing, 40 mg/kg 3X/week), mice four weeks after surgical castration (Orch, *n*=8). Horizontal bars represent mean ± s.e. and diamonds represent individual mice.

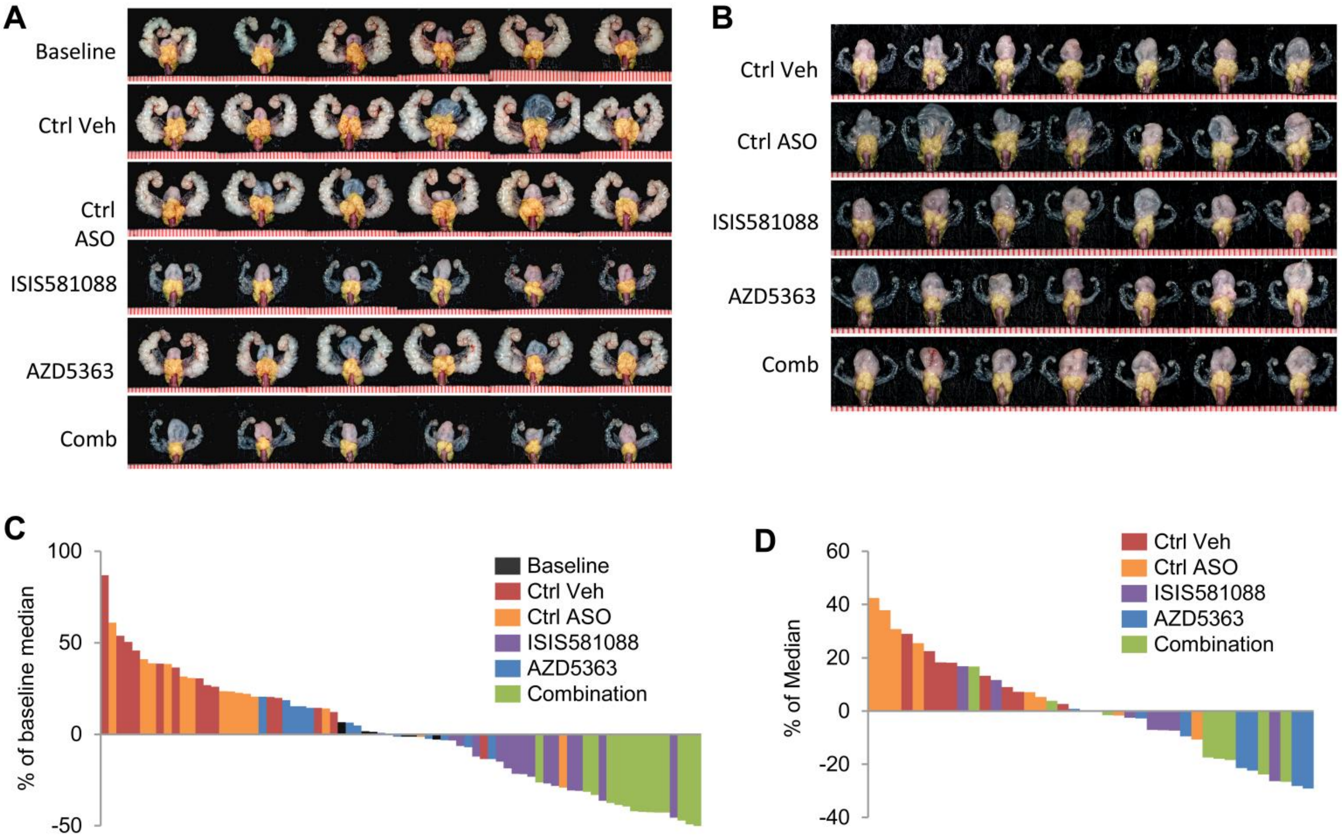


B

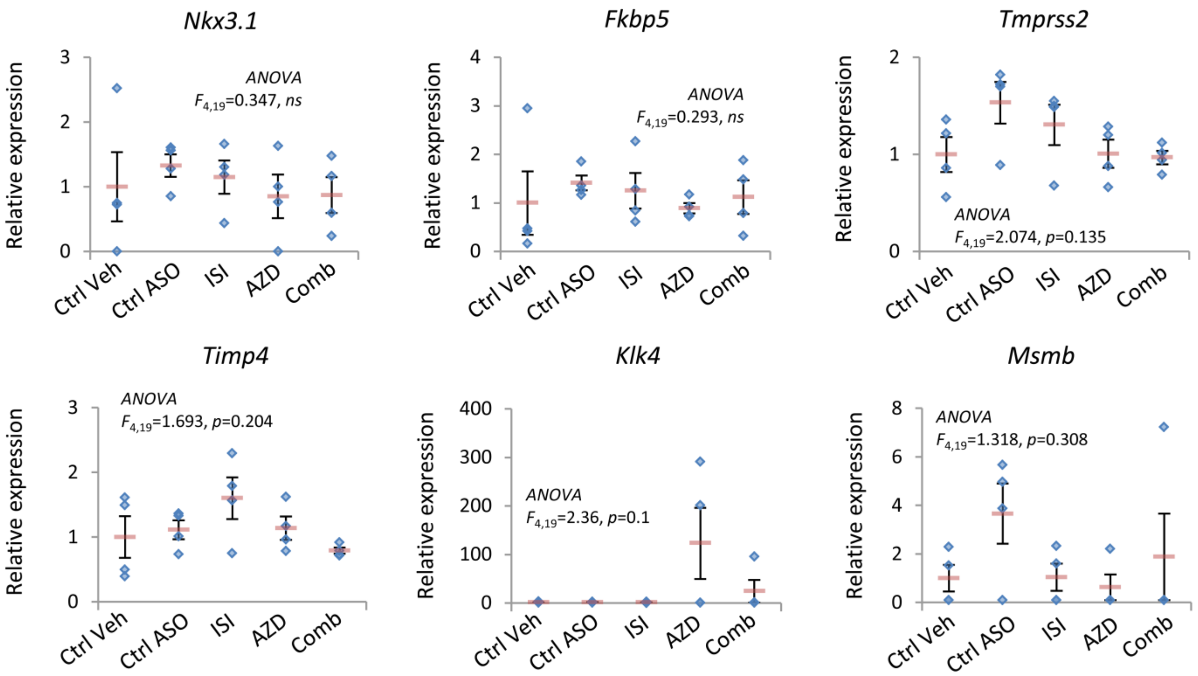
Cell line	Origin	Castratiion status	Mean ED_{50} (s.e.) ISI	Mean ED_{50} (s.e.) AZD 5363	Mean CI at ED_{50} (s.e.)	Combination effect
7109-G4	87 wk-old *Pten*$^{-/-}$	CNPC Intact	7.25 (0.85)	2.06 (0.40)	0.66 (0.11)	Synergistic
2945-F12	83 wk-old *Pten*$^{-/-}$	CRPC 12 wks post castration	17.84 (1.13)	0.98 (0.13)	0.85 (0.10)	Moderately synergistic
3902-A1	66 wk-old *Pten*$^{-/-}$/ *Trp53*$^{-/-}$	CNPC Intact	25.99 (2.68)	3.70 (0.62)	0.63 (0.03)	Synergistic
4522-B8	60 wk-old *Pten*$^{-/-}$/ *Trp53*$^{-/-}$	CRPC 13 wks post castration	24.96 (0.99)	4.74 (0.94)	0.81 (0.05)	Moderately synergistic



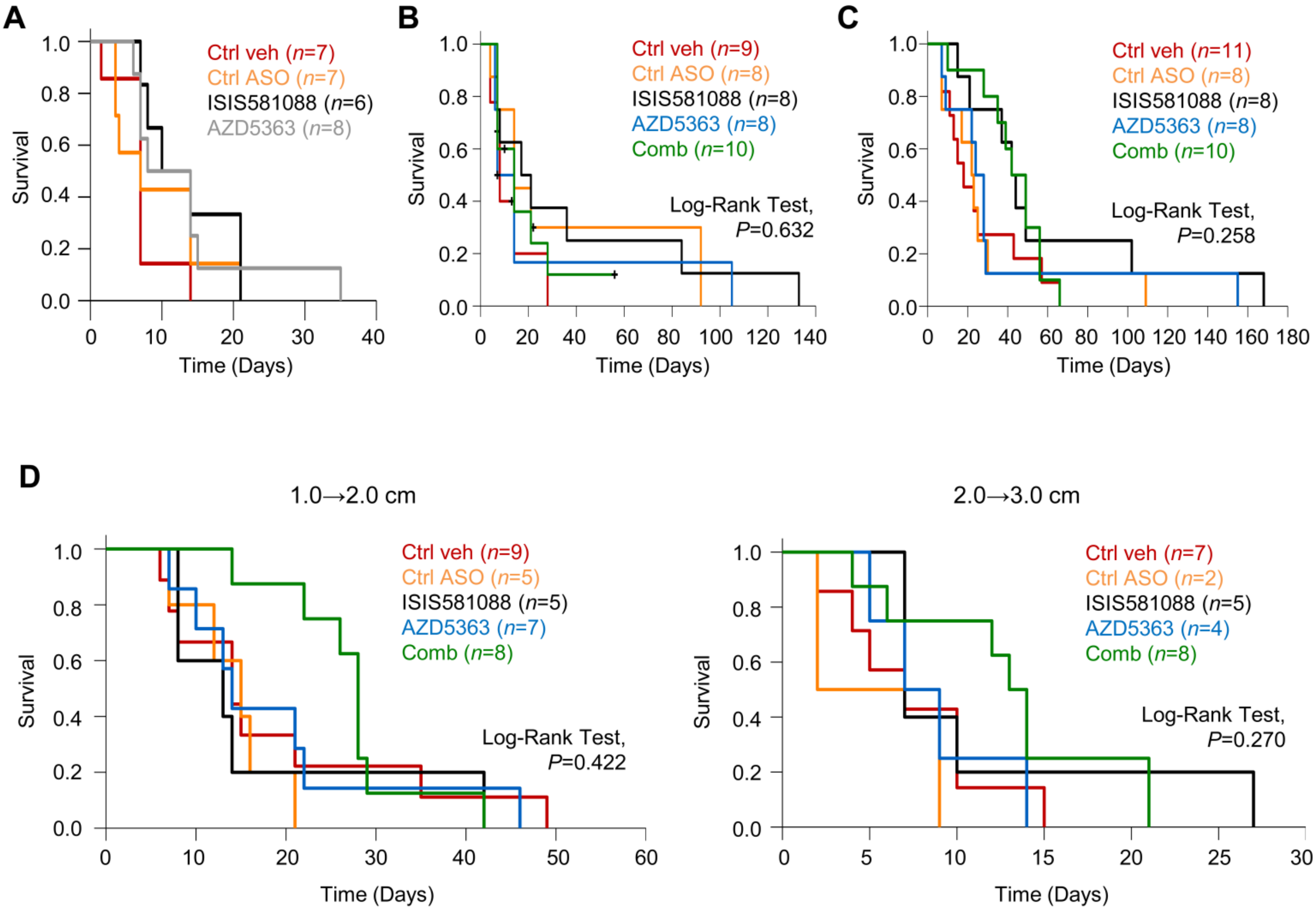
Supplemental Figure 5. *In vitro* efficacy determination of combination therapy with ISIS581088 and AZD5363 mouse *Pten*-deficient prostate cancer lines. (**A**) Schematic for the establishment of mouse prostate cancer cell lines. (**B**) Summary of drug sensitivity (effective dose requiring 50% growth inhibition (ED_{50})) and synergy measured by the Chou-Talalay combination index (CI). Dose-effect curves of ISIS581088 and AZD5363 combination therapy in mouse prostate cancer cell lines (**C**) and representative isobolograms for the constant-ratio design (**D**). Data represent mean values of three independent experiments performed in quadruplicate.



Supplemental Figure 6. Efficacy determination of *in vivo* combination therapy with ISIS581088 and AZD5363 in mouse *Pten*-deficient prostate cancer models. Representative images of genitourinary tracts (**A**) and waterfall plot (**C**) to the indicated treatments of mice in Figure 7A are shown. Representative images of genitourinary tracts (**B**) and waterfall plot (**D**) to the indicated treatments of mice in Figure 7D are shown. Prostate tumors are highlighted by a yellow mask, scale represents mm. Waterfall plot of individual treatment response based on tumor area normalized to median baseline or vehicle control.



Supplemental Figure 7. mRNA expression of AR-responsive genes in CRPC after combination therapy with ISIS581088 and AZD5363. qRT-PCR analysis of mRNA of core AR-responsive genes in prostate tumors from Figure 8D. Horizontal bars represent mean ± s.e. and diamonds represent individual samples, *n*=4 mice/group.



Pairwise multiple comparisons (Holm-Sidak method)	*P Value*
Ctrl ASO vs. Comb	0.0170

Supplemental Figure 8. Combined AR and PI3K/AKT blockade suppresses cancer growth and improves survival in a mouse model of advanced castration-resistant prostate cancer (CRPC). Kaplan-Meier plots showing progression-free survival curves for mice in Fig. 10 after the indicated treatment in castration-naive (**A**) and CRPC (**B**) settings. (**C**) Kaplan-Meier plots showing overall survival after treatment randomization of mice in Fig. 10c. (**D**) Kaplan-Meier plots showing time to tumor growth in the CRPC setting (palpable tumor size from 1 to 2 cm and 2 to 3 cm).

Supplementary Table 1. ASO uptake in surrounding tissues

Group	Time	Dose (mg/kg)	No. of mice	Normal prostate glands		Anterior prostate (Coagulating gland)	Urothelium	Connective tissue		Skeletal muscle	Peri-urethral gland	Seminal vesicle
				Dorsal lobe	Ventral lobe			Macro phages	Fibro blasts			
Baseline	0	0	4	-	-	-	-	-	-	-	-	-
Ctrl ASO	24	40	2	-	+/-	+/-	+	+++	++	+/-	++	+
Ctrl ASO	96	40	2	+/-	+	-	+	+++	++	-	+	+
ISIS581088	2	40	2	-	-	-	+/-	+++	+	+/-	np	+
ISIS581088	4	40	2	-	+/-	-	-	+++	++	+/-	+/-	+
ISIS581088	8	40	2	-	+	-	+/-	++	++	-	np	+
ISIS581088	24	40	2	-	+	-	+	+++	++	-	np	+
ISIS581088	48	40	2	-	+/-	-	+	+++	+	-	+/-	+
ISIS581088	76	40	2	-	+	-	+	+++	++	-	+/-	np
ISIS581088	96	40	2	+/-	+	-	++	+++	++	-	+	+

np (not present), - negative , +/- slight, + minimal ++ mild +++ moderate

Supplementary Table 2. One way ANOVA analysis for qRT-PCR-based panel Ar , core AR responsive, AR stimulating, proliferation and apoptosis genes.

Gene	BL mean	BL sd	Veh mean	Veh sd	ISI mean	ISI sd	Cast mean	Cast sd	Enz mean	Enz sd	Apa mean	Apa sd	Abi mean	Abi sd	CMA mean	CMA sd	F ratio	Sum of squares (groups)	Sum of squares (error)	degrees of freedom (groups)	degrees of freedom (error)	Raw p-Value
Ar Ex5-Ex6J	0.424	0.326	-0.043	0.206	-2.171	0.072	0.513	0.627	0.808	0.405	-0.149	0.423	0.874	0.337	-0.256	0.670	20.988	26.647	4.353	7.0	24.0	8.74E-09
Ar-Ex2	-0.074	0.279	-0.264	0.119	-1.965	0.079	0.704	0.236	1.075	0.712	-0.268	0.342	0.750	0.701	0.041	0.877	13.957	24.886	6.114	7.0	24.0	4.41E-07
Ar-Ex7	-0.100	0.179	-0.162	0.031	-2.112	0.087	0.603	0.314	0.965	0.538	-0.007	0.416	0.759	0.691	0.054	0.873	15.808	25.475	5.525	7.0	24.0	1.38E-07
Casp3	0.962	0.860	-0.266	0.523	-1.195	0.598	-0.404	0.535	0.214	0.595	0.129	0.633	-0.637	0.331	1.195	1.353	4.709	17.939	13.061	7.0	24.0	1.93E-03
Mki67	-0.785	0.190	0.298	0.809	1.260	0.632	1.384	0.718	-1.141	0.143	0.037	0.479	-0.859	0.161	-0.194	0.264	14.733	25.148	5.852	7.0	24.0	2.67E-07
Myc	1.104	0.604	-1.110	0.250	-1.124	0.464	-0.175	0.372	1.154	0.828	-0.112	0.589	0.717	0.844	-0.455	0.241	10.274	23.244	7.756	7.0	24.0	6.57E-06
Nkx3.1	0.709	0.812	0.553	0.863	-0.503	0.616	-1.033	0.032	0.783	0.814	-0.718	0.227	0.545	1.617	-0.336	0.577	3.079	14.668	16.332	7.0	24.0	1.84E-02
Stat3	1.246	0.574	-0.889	0.428	-1.205	0.374	-0.224	0.487	1.179	0.765	-0.597	0.441	0.503	0.358	-0.013	0.863	10.498	23.368	7.632	7.0	24.0	5.48E-06
Fkbp5	0.769	0.684	0.843	0.421	-1.073	0.080	-1.147	0.082	1.371	0.704	-0.862	0.160	0.364	0.558	-0.265	0.220	19.581	26.381	4.619	7.0	24.0	1.74E-08
Igf1r	1.319	0.659	-0.669	0.388	-1.053	0.156	0.110	0.164	0.724	1.273	-0.679	0.478	0.234	0.901	0.013	1.113	4.383	17.395	13.605	7.0	24.0	2.95E-03
Msmb	0.037	0.612	1.022	0.917	-0.529	0.185	-0.736	0.000	1.388	1.733	-0.450	0.096	-0.237	0.405	-0.496	0.419	4.272	17.197	13.803	7.0	24.0	3.42E-03
Timp4	0.711	0.317	1.666	1.233	-0.874	0.038	-0.899	0.027	0.605	0.468	-0.699	0.252	0.166	0.405	-0.677	0.060	13.658	24.780	6.220	7.0	24.0	5.37E-07
Tmprss2	0.897	0.345	0.103	0.883	-1.282	0.315	-0.799	0.160	1.255	0.914	-0.757	0.478	0.580	0.533	0.004	0.719	8.835	22.333	8.667	7.0	24.0	2.28E-05
Bcl2	-0.236	0.816	-0.652	0.783	0.442	1.166	0.837	1.409	0.551	0.881	-0.902	0.737	0.529	0.435	-0.570	0.266	2.283	12.391	18.609	7.0	24.0	6.22E-02
Klk4	0.609	1.916	-0.303	0.039	-0.342	0.022	-0.265	0.144	0.629	1.903	-0.284	0.084	-0.313	0.037	0.269	1.133	0.689	5.186	25.814	7.0	24.0	6.81E-01
Igf1	0.117	0.624	-0.395	0.405	-0.116	1.079	0.079	1.467	0.164	0.313	-0.679	0.170	0.668	1.414	0.162	1.675	0.597	4.599	26.401	7.0	24.0	7.52E-01

Supplementary Table 3. Fold changes of mRNA expression of AR-dependent genes in mouse *Pten*-deficient prostate cancer

	CNPC				CRPC			
	Linear fold change vs Ctrl Unt		p-Value vs Ctrl Unt		Linear fold change vs Ctrl Unt		p-Value vs Ctrl Unt	
Gene	Ctrl ASO	ISIS 581088	Ctrl ASO	ISIS 581088	Ctrl ASO	ISIS 581088	Ctrl ASO	ISIS 581088
Abcc4	1.033	0.890	0.903	0.669	0.856	0.799	0.663	0.549
Abo	1.389	0.198	0.380	0.001	0.244	0.250	0.412	0.528
Actg2	0.757	0.469	0.687	0.308	0.461	0.197	0.279	0.051
Aldh1l2	0.425	0.360	0.032	0.013	0.504	0.572	0.088	0.173
Alox12	0.660	0.914	0.337	0.833	0.960	0.639	0.950	0.494
Ank1	0.569	0.624	0.194	0.272	0.972	0.516	0.950	0.153
Ar	0.696	0.068	0.257	0.000	0.865	0.157	0.575	0.000
B4Gal5	0.561	0.505	0.050	0.023	0.719	0.748	0.170	0.223
C1qc	0.643	1.163	0.379	0.740	0.788	0.924	0.806	0.932
Camkk2	0.880	0.923	0.844	0.894	0.388	0.449	0.211	0.229
Chka	1.064	0.788	0.770	0.269	0.911	0.864	0.624	0.443
Crabp1	1.076	0.194	0.862	0.001	0.322	0.357	0.051	0.072
Creld2	0.856	0.580	0.552	0.051	0.870	0.912	0.573	0.708
Cxcl11	0.846	2.594	0.778	0.127	0.742	0.458	0.481	0.128
Cxcl17	0.584	1.145	0.479	0.858	0.954	0.508	0.944	0.322
Cyp2f2	0.877	1.260	0.604	0.366	0.866	0.666	0.712	0.305
Dbi	0.950	0.600	0.900	0.218	0.922	0.897	0.689	0.591
Derl3	0.562	0.266	0.122	0.002	0.883	0.693	0.582	0.121
Dhcr24	0.871	0.449	0.710	0.044	0.985	0.918	0.948	0.717
Dhcr7	0.846	0.634	0.302	0.011	0.968	0.992	0.895	0.975
Dnajc3	1.010	0.544	0.978	0.100	0.890	0.866	0.584	0.499
Doc2b	1.256	0.236	0.500	0.001	0.858	0.816	0.602	0.491
Epas1	0.713	0.516	0.305	0.056	0.848	0.841	0.535	0.513
Epn2	1.122	0.556	0.710	0.107	0.880	0.860	0.619	0.576
Erp44	1.061	0.807	0.733	0.230	0.967	0.935	0.862	0.730
Fas	0.724	0.831	0.321	0.563	0.983	0.961	0.896	0.769
Fkbp11	0.852	0.587	0.502	0.037	0.902	0.961	0.717	0.889
Fkbp5	0.964	0.248	0.891	0.000	0.821	0.795	0.326	0.256
Gmpr	0.723	0.936	0.187	0.782	0.886	0.877	0.531	0.496
Gnmt	0.391	0.174	0.004	0.000	0.862	0.568	0.547	0.055
Grb10	0.641	0.579	0.057	0.023	0.728	0.657	0.284	0.162
Greb1	0.178	0.112	0.002	0.000	0.946	0.948	0.892	0.896
Gsdma1	0.093	0.208	0.000	0.000	0.903	0.968	0.806	0.937
Hapln4	0.912	0.266	0.822	0.005	0.893	0.962	0.777	0.922
Herc3	0.760	0.716	0.191	0.116	0.941	0.711	0.807	0.183
Hif1a	0.796	0.572	0.428	0.064	0.976	0.999	0.876	0.995
Hmgcr	0.973	0.596	0.879	0.011	0.836	0.801	0.462	0.364
Hmgcs1	0.536	0.535	0.063	0.076	0.920	0.774	0.861	0.593
Insig2	0.911	1.543	0.524	0.008	0.866	0.680	0.658	0.243
Iqgap2	0.824	0.377	0.547	0.007	0.937	0.977	0.790	0.924
Ldha	1.104	1.011	0.633	0.959	0.866	0.948	0.441	0.775
Limk2	0.791	0.795	0.375	0.386	0.955	0.804	0.831	0.325
Map2k4	0.982	1.261	0.956	0.493	0.947	0.952	0.777	0.796

Supplementary Table 3. Fold changes of mRNA expression of AR-dependent genes in mouse *Pten*-deficient prostate cancer (continued)

Gene	CNPC				CRPC			
	Linear fold change vs Ctrl Unt		p-Value vs Ctrl Unt		Linear fold change vs Ctrl Unt		p-Value vs Ctrl Unt	
	Ctrl ASO	ISIS 581088	Ctrl ASO	ISIS 581088	Ctrl ASO	ISIS 581088	Ctrl ASO	ISIS 581088
Mapre2	0.831	0.988	0.409	0.957	0.931	0.821	0.725	0.341
Mat2a	0.652	0.284	0.107	0.000	0.990	0.760	0.971	0.320
Mfsd2	0.598	0.130	0.099	0.000	0.828	0.549	0.468	0.033
Mki67	0.697	0.348	0.138	0.000	0.838	0.642	0.300	0.018
Mme	0.866	0.227	0.514	0.000	0.874	0.365	0.664	0.005
Mthfd2	0.612	0.139	0.236	0.000	0.956	0.939	0.900	0.867
Myc	1.036	0.763	0.919	0.464	0.863	0.543	0.742	0.186
Ndgr1	0.572	0.817	0.086	0.515	0.751	0.647	0.524	0.336
Nelf	0.697	0.646	0.065	0.029	0.978	0.808	0.947	0.527
Nkx3-1	0.321	0.122	0.000	0.000	0.590	0.724	0.340	0.568
Parp1	0.710	0.707	0.200	0.196	0.948	0.983	0.740	0.913
Pdia4	0.837	0.472	0.362	0.001	0.945	0.908	0.846	0.738
Pdk4	0.507	0.593	0.170	0.285	0.903	0.811	0.751	0.514
Pdlim2	0.869	0.374	0.267	0.000	0.952	0.714	0.904	0.412
Pfkfb4	0.751	0.428	0.337	0.010	0.860	0.772	0.406	0.165
Pgam1	0.776	1.018	0.651	0.974	0.870	0.600	0.842	0.489
Phf19	0.227	0.105	0.019	0.002	0.939	0.611	0.911	0.452
Pkd2l1	1.010	1.042	0.981	0.924	0.700	0.781	0.224	0.413
Pkm2	0.237	0.341	0.002	0.011	0.946	0.834	0.893	0.672
Ppara	0.685	0.799	0.338	0.565	0.843	0.821	0.633	0.598
Ppargc1a	0.666	0.871	0.187	0.645	0.921	0.800	0.630	0.204
Psmd1	0.952	0.878	0.889	0.709	0.988	0.892	0.960	0.642
Psph	1.435	1.665	0.413	0.253	0.834	0.772	0.646	0.513
Rasgrf1	0.446	0.323	0.060	0.016	0.868	0.819	0.575	0.452
Scd4	0.543	0.231	0.319	0.025	0.507	0.550	0.323	0.430
Sgk1	1.482	0.424	0.177	0.008	0.932	0.740	0.750	0.189
Sgk3	0.680	0.780	0.273	0.475	0.812	0.787	0.328	0.262
Slc2a4	0.395	0.425	0.004	0.009	0.701	0.339	0.619	0.161
Slc38a5	0.584	0.121	0.153	0.000	0.724	0.491	0.726	0.472
Slc40a1	0.745	1.057	0.387	0.869	1.000	0.900	0.999	0.584
Sqle	1.047	0.530	0.886	0.064	0.903	0.832	0.723	0.526
Stk39	0.724	0.426	0.452	0.059	0.861	0.760	0.748	0.558
Tax1bp1	0.762	0.988	0.290	0.961	0.854	0.873	0.195	0.259
Timp4	1.332	0.232	0.421	0.001	0.861	0.949	0.375	0.751
Tmem45a	0.642	0.766	0.275	0.508	0.901	0.891	0.644	0.609
Tmem97	0.538	0.531	0.012	0.011	0.928	0.752	0.675	0.122
Tmprss2	0.751	0.607	0.059	0.003	0.991	0.825	0.976	0.503
Tpd52	1.804	2.958	0.703	0.465	0.854	0.632	0.786	0.453
Twist1	0.599	1.110	0.293	0.835	0.956	0.486	0.904	0.082
Ube2c	0.378	0.479	0.020	0.078	0.775	0.538	0.699	0.374
Uso1	1.045	0.506	0.879	0.032	0.887	0.988	0.652	0.964
Wwtr1	0.924	0.770	0.905	0.693	0.720	0.671	0.391	0.379
Zfp36l1	0.702	0.858	0.134	0.501	0.891	0.707	0.676	0.219

Supplementary Table 4. Summary of mouse characteristics and treatment responses for CNPC treated mice

Description	Ctrl veh	Ctrl ASO	ISIS581088	Orch.	*P*-value
No. of Mice	8	7	7	9	
Median start age (w)	56.9	52.9	58.4	49.9	*0.957*
range	*(39-83.9)*	*(42.4-61.4)*	*(43.4-66.4)*	*(39.4-67.1)*	
Median initial bodyweight (g)	35.9	35.9	35.7	34.9	*0.817*
range	*(31.3-44.2)*	*(31.9-49.0)*	*(32.4-41.4)*	*(28.5-57.3)*	
Median bodyweight change (%)	-13.9	-10.3	-12.2	-6.7	*0.187*
range	*(-26.3-17.5)*	*(-17.8-8.8)*	*(-20.2-9.9)*	*(-29.2-4.2)*	
Median GUT weight (g)	2.5	2.7	3.7	3.2	*0.853*
range	*(1.2-9.3)*	*(2.4-8.9)*	*(1.3-7.3)*	*(1.3-7.0)*	
Metastasic incidence (%)	22.2	40	20	33.3	*0..915*
frequency	*(2/7)*	*(2/5)*	*(1/5)*	*(3/9)*	
Median GUT weight (g)	3	3	2	2	*0.365*
range	*(2-3)*	*(1-3)*	*(1-3)*	*(1-3)*	

Supplementary Table 5. Summary of mouse characteristics and treatment responses

Description	Ctrl veh	Ctrl ASO	ISIS581088	AZD5363	ISIS581088 + AZD5363	*P*-value
No. of Mice	11	8	8	8	10	
Median start age (w)	55.4	61.6	49	63.4	60.1	*0.167*
range	*(45.9-78.9)*	*(42.4-65.4)*	*42.4-70.9)*	*52.6-73.9)*	*(48.4-70.9)*	
Median initial bodyweight (g)	34.3	33.5	35.3	34.2	35.7	*0.847*
range	*(32.1-38.0)*	*(30.6-43.6)*	*(33.4-41.9)*	*(32.0-38.5)*	*(31.3-38.9)*	
Median bodyweight change (%)	0.6	-3.3	-3.3	0.1	-7	*0.369*
range	*(-11.7-17.1)*	*(-18.2-18.0)*	*(-18.2-10.0)*	*(-9.6-12.7)*	*(-24.3-5.7)*	
Median genitourinary tract (GUT) weight (g)	3.8	2.2	5.4	2.7	3.8	*0.264*
range	*(1.2-11.9)*	*(0.7-7.1)*	*(1.1-11.8)*	*(0.9-5.9)*	*(0.3-8.7)*	
Metastasic incidence (%)	36.4	50	37.5	37.5	50	*0.945*
frequency	*(4/11)*	*(4/8)*	*(3/8)*	*(3/8)*	*(5/10)*	
# of metastatic lesions/mouse	1.5	1.5	3.3	1.7	1.6	*0.449*
range	*(1-3)*	*(1-2)*	*(1-7)*	*(1-2)*	*(1-3)*	
Mean performance score	2.2	2.3	1.6	2.5	1.8	*0.377*
s.e.	*0.3*	*0.4*	*0.4*	*0.3*	*0.2*	

Supplementary Table 6. Multi-cluster gene functional enrichment analyses for mouse CNPC and CRPC after *Ar* gene silencing with ISIS581088

			pValues		Gene List	
Category	ID	Title (or Source)	CNPC - logP	CRPC - logP	CNPC	CRPC
GO: Biological Process	GO: 0055114	oxidation-reduction process	10.0000	5.8407	*AKR1B1 ALDH1L2 DHCR24 DHCR7 GNMT GRB10 HIF1A HMGCR MTHFD2 PDK4 PFKFB4 PKM SCD5 SQLE*	*AKR1B1 ALDH1L2 ALOX12 CYP2F1 GNMT GRB10 MYC PGAM1 SCD5 TWIST1*
GO: Biological Process	GO: 0051186	cofactor metabolic process	10.0000	5.6186	*AKR1B1 ALDH1L2 ANK1 GNMT HIF1A HMGCR MAT2A MTHFD2 PDK4 PFKFB4 PKM SCD5 TSPO*	*AKR1B1 ALDH1L2 ANK1 GNMT MYC PGAM1 SCD5*
GO: Biological Process	GO: 0006695	cholesterol biosynthetic process	10.0000		*DHCR24 DHCR7 HMGCR HMGCS1 SQLE*	
GO: Biological Process	GO: 0009628	response to abiotic stimulus	10.0000		*AKR1B1 DNAJC3 EPAS1 HIF1A HMGCR HMGCS1 MKI67 MME NKX3-1 NSMF PKM SGK1 SLC2A4 STK39 TSPO*	
GO: Biological Process	GO: 0032787	monocarboxylic acid metabolic process		10.0000		*ALOX12 CRABP1 CYP2F1 GNMT INSIG2 MYC PGAM1 SCD5 TWIST1*
GO: Biological Process	GO: 0016126	sterol biosynthetic process	10.0000		*DHCR24 DHCR7 HMGCR HMGCS1 SQLE*	
GO: Biological Process	GO: 0006732	coenzyme metabolic process	10.0000		*ALDH1L2 GNMT HIF1A HMGCR MAT2A MTHFD2 PDK4 PFKFB4 PKM SCD5*	
GO: Biological Process	GO: 0044283	small molecule biosynthetic process	10.0000		*AKR1B1 DHCR24 DHCR7 GNMT HIF1A HMGCR HMGCS1 MTHFD2 PDK4 PKM SCD5 SQLE*	
GO: Biological Process	GO: 0009725	response to hormone	10.0000		*AKR1B1 DHCR24 GRB10 HIF1A HMGCS1 MAT2A NKX3-1 NSMF PDK4 PKM SGK1 SLC2A4 TIMP4 TSPO*	
GO: Biological Process	GO: 1902653	secondary alcohol biosynthetic process	10.0000		*DHCR24 DHCR7 HMGCR HMGCS1 SQLE*	
GO: Biological Process	GO: 0006694	steroid biosynthetic process	10.0000		*AKR1B1 DHCR24 DHCR7 HMGCR HMGCS1 SQLE TSPO*	
GO: Biological Process	GO: 0019752	carboxylic acid metabolic process		5.8130		*ALDH1L2 ALOX12 CRABP1 CYP2F1 GNMT INSIG2 MYC PGAM1 SCD5 TWIST1*
GO: Biological Process	GO: 0010243	response to organonitrogen compound	5.7496		*AKR1B1 GRB10 HIF1A HMGCS1 MAT2A NSMF PDK4 PKM SGK1 SLC2A4 TIMP4 TSPO*	
GO: Biological Process	GO: 0018931	naphthalene metabolic process		5.6299		*AKR1B1 CYP2F1*
GO: Biological Process	GO: 0090420	naphthalene-containing compound metabolic process		5.6299		*AKR1B1 CYP2F1*
GO: Biological Process	GO: 0006730	one-carbon metabolic process	5.5342		*ALDH1L2 GNMT MAT2A MTHFD2*	
GO: Biological Process	GO: 1901617	organic hydroxy compound biosynthetic process	5.5211		*AKR1B1 DHCR24 DHCR7 EPAS1 HMGCR HMGCS1 SQLE*	
GO: Biological Process	GO: 0046165	alcohol biosynthetic process	5.4391		*AKR1B1 DHCR24 DHCR7 HMGCR HMGCS1 SQLE*	

Supplementary Table 6. Multi-cluster gene functional enrichment analyses for mouse CNPC and CRPC after *Ar* gene silencing with ISIS581088 (continued)

			pValues		Gene List	
Category	ID	Title (or Source)	CNPC -logP	CRPC -logP	CNPC	CRPC
GO: Biological Process	GO: 0043436	oxoacid metabolic process		5.4020		*ALDH1L2 ALOX12 CRABP1 CYP2F1 GNMT INSIG2 MYC PGAM1 SCD5 TWIST1*
GO: Biological Process	GO: 0019318	hexose metabolic process	5.2283		*AKR1B1 GNMT GRB10 HIF1A PDK4 PFKFB4 PKM*	
GO: Biological Process	GO: 1901657	glycosyl compound metabolic process	4.7909		*AKR1B1 GNMT HIF1A HMGCR MAT2A PFKFB4 PKM TSPO*	
GO: Biological Process	GO: 0005975	carbohydrate metabolic process		4.5789		*ABO AKR1B1 DERL3 GNMT GRB10 MYC PGAM1 SLC2A4*
Pathway	SMP 00023	Steroid Biosynthesis	10.0000		*DHCR24 HMGCR HMGCS1 SQLE*	
Pathway	1270037	Cholesterol biosynthesis	10.0000		*DHCR24 DHCR7 HMGCR HMGCS1 SQLE*	
Pathway	142269	superpathway of cholesterol biosynthesis	10.0000		*DHCR24 DHCR7 HMGCR HMGCS1 SQLE*	
Coexpression	M2571	Genes dn-regulated during prostate cancer progression in the JOCK1 model due to inducible activation of FGFR1 gene in prostate.	10.0000	5.7103	*ACTG2 CRABP1 GNMT GREB1 GSDMA MAT2A MTHFD2 PHF19 SLC2A4 TIMP4*	*ACTG2 CRABP1 GNMT NDRG1 PHF19 SLC2A4*
Coexpression	12734205-TableS4	Human Ovarian Wood03 142genes		10.0000		*ACTG2 MME MYC SCD5 TWIST1*
Coexpression	M5908	Genes defining response to androgens.	10.0000		*DHCR24 FKBP5 HMGCR HMGCS1 IQGAP2 NKX3-1 SGK1 STK39 TMPRSS2*	
Coexpression	M2672	Genes up-regulated by everolimus [PubChem = 6442177] in prostate tissue.	10.0000		*DHCR24 DHCR7 GSDMA HMGCS1 SQLE TMEM97 UBE2C*	
Coexpression	12606954-table1b	Human Prostate Bigler03 11genes	5.7837		*FKBP5 IQGAP2 TMPRSS2*	
Coexpression	16707422-tableS1a	Human Prostate Hendriksen06 163genes	5.3977		*DHCR24 FKBP5 HMGCR STK39 TMPRSS2*	
Coexpression	20386565-TableS1	Human Prostate Zhao10 1841genes	5.0856		*ACTG2 DHCR7 GREB1 HIF1A IQGAP2 MTHFD2 PDIA4 TMEM97 UBE2C*	

Supplementary Table 6. Multi-cluster gene functional enrichment analyses for mouse CNPC and CRPC after *Ar* gene silencing with ISIS581088 (continued)

			pValues		Gene List	
Category	ID	Title (or Source)	CNPC - logP	CRPC - logP	CNPC	CRPC
Coexpression	M8124	Genes up-regulated in basal subtype of breast cancer samples.		4.9268		*ACTG2 CRABP1 CXCL11 MKI67 MYC NDRG1 WWTR1*
Coexpression	15899800-TableS1	Human Breast Martens05 117genes		4.8879		*AKR1B1 MKI67 MYC NDRG1*
Drug	C053541	bicalutamide	5.3787		*FKBP11 GREB1 MKI67 MME NKX3-1 TMPRSS2*	
Disease	umls: C0282612	Prostatic Intraepithelial Neoplasias	5.6325	5.6099	*MKI67 MME NKX3-1 SGK1 TMPRSS2 TSPO*	*ALOX12 MKI67 MME MYC NDRG1*
Disease	umls: C0001430	Adenoma		5.7515		*AKR1B1 ALOX12 CRABP1 MKI67 MME MYC NDRG1 TPD52 TWIST1 UBE2C*
Disease	umls: C1134719	Invasive Ductal Breast Carcinoma		5.6668		*ABO INSIG2 MKI67 MYC TWIST1 UBE2C*
Disease	umls: C0033578	Prostatic Neoplasms	5.2857		*ABO DNAJC3 FKBP5 GNMT GREB1 HIF1A MME NKX3-1 SQLE TIMP4 TMPRSS2 TSPO USO1*	
Disease	umls: C0677886	Epithelial ovarian cancer		5.2539		*ABO AKR1B1 ALOX12 GRB10 MKI67 MYC SLC2A4 TWIST1 UBE2C WWTR1*
Disease	umls: C0023903	Liver neoplasms	5.1997		*CRABP1 EPAS1 GNMT HIF1A HMGCR HMGCS1 IQGAP2 MAT2A PDK4 PKM RASGRF1 SGK1 SLC2A4 SQLE*	
Disease	umls: C0005695	Bladder Neoplasm		4.8533		*ABO MKI67 MME MYC NDRG1 TWIST1*
Disease	umls: C0023453	Leukemia, Lymphocytic, Acute, L2		4.7606		*MME MYC*
Disease	umls:C0746787	Cancer of Neck	4.5686		*EPAS1 HIF1A*	
Disease	umls: C0345904	Malignant neoplasm of liver		4.3702		*ABO AKR1B1 CAMKK2 CXCL11 GNMT MYC PGAM1*
Disease	umls: C1511306	Breast Diffuse Large B-Cell Lymphoma		4.3636		*MKI67 MME*

Supplementary Table 7. Antibody List

Antibodies for western blot assays

Marker	Source	Catalogue number	Predicated Band size	Dilution	2° antibody	Reference
AR (NTD)	Thermo	RB-9030	110	1:1000	Rabbit	
AR (T660)	R&D Systems	MAB58761	110	1:1000	Mouse	
Cleaved Caspase-3	Cell Signaling	9661	17,19	1:1000	Rabbit	https://www.antibodypedia.com/gene/1222/CASP3/antibody/108063/9661
Akt	Cell Signaling	9272	60	1:1000	Rabbit	https://www.antibodypedia.com/gene/135/AKT1/antibody/166716/9272
P-Akt	Cell Signaling	4060	60	1:1000	Rabbit	https://www.antibodypedia.com/gene/135/AKT1/antibody/106995/4060
P-PRAS40	Cell Signaling	13175	80	1:1000	Rabbit	https://www.antibodypedia.com/gene/32195/AKT1S1/antibody/1286783/13175
P-S6	Cell Signaling	2211	32	1:1000	Rabbit	https://www.antibodypedia.com/gene/3412/RPS6/antibody/105615/2211
GAPDH	Cell Signaling	2118	37	1:10000	Rabbit	https://www.antibodypedia.com/gene/3923/GAPDH/antibody/105543/2118

Antibodies For IHC

Marker	Source	Catalogue number	Antigen retrieval	Dilution	2° antibody	Reference
ASO	Ionis Pharmaceuticals	na		1:100	Rabbit	See reference no. 27
AR (NTD)	Thermo Fisher Scientific	RB-9030	Dako ARS	1:100	Rabbit	
AR (S650)	Abcam	AB-47570	Dako ARS	1:100	Rabbit	
Cleaved Caspase-3	Cell Signaling	9661	Dako ARS	1:400	Rabbit	https://www.antibodypedia.com/gene/1222/CASP3/antibody/108063/9661
Ki67	Thermo Fisher Scientific	RB-9043	Dako ARS	1:250	Rabbit	https://www.antibodypedia.com/gene/741/MKI67/antibody/630324/PA5-16446

DAKO ARS: 20 min steam heating in DAKO Target retrieval solution (S1699, Agilent, Santa Clara, CA)

Supplementary Table 8. List of PCR primers used for qRT-PCR

Target	Strand	Sequence	Source
Ar Exon 2	Forward	TTGGACAGTACCAGGGACCA	PrimerBlast
	Reverse	GAGAGCTCCGTAGTGACAGC	
Ar Exon 5/6 junc	Forward	GGATGCTCTACTTTGCACCTGA	PrimerBlast
	Reverse	AGGTGCCTCATCCTCACACA	
Ar Exon 7	Forward	CCAGTGGATGGGCTGAAAAAT	PrimerBlast
	Reverse	CCTTGAGCAGGATGTGGGATT	
Bcl2	Forward	AGGGTCTTCAGAGACAGCCA	qPrimerDepot
	Reverse	AGTACCTGAACCGGCATCTG	
Casp3	Forward	GACTTGCTCCCATGTATGGTC	qPrimerDepot
	Reverse	ATCAAAGCGCAGTGTCCTG	
Fkbp5	Forward	CCAACAACGAACACCACATC	qPrimerDepot
	Reverse	CGAGGGATACTCAAACCCAA	
Gapdh	Forward	CGAACGGGAAGCTCACTGGCAT	qPrimerDepot
	Reverse	TCAGATGCCTGCTTCACCACCT	
Igf1	Forward	CACTCATCCACAATGCCTGT	qPrimerDepot
	Reverse	TGGATGCTCTTCAGTTCGTG	
Igf1r	Forward	CAGCACTCGTTGTTCTCGGT	qPrimerDepot
	Reverse	AGCCCATGTGTGAGAAGACC	
Klk4	Forward	CTTGTATGATCCGGCTGCTG	qPrimerDepot
	Reverse	AGCCAACATGATGGTCACTG	
Mki67	Forward	CCATCTGAGGCAGGGCTATC	PrimerBlast
	Reverse	TTTCATATTTATTGCCAAGATGGGG	
Msmb	Forward	ACAGGTCTTCCCTGGGTTCT	qPrimerDepot
	Reverse	CGTCAATCACCTGCTGTACC	
Myc	Forward	TGAAGTTCACGTTGAGGGG	qPrimerDepot
	Reverse	AGAGCTCCTCGAGCTGTTTG	
Nkx3.1	Forward	CACTTGCTAAGTCCCCTGGA	qPrimerDepot
	Reverse	AGTATCCGGCATAGCCCC	
Stat3	Forward	CTGCTCCAGGTAGCGTGTGT	qPrimerDepot
	Reverse	CTCAGCCCCGGAGACAGT	
Timp4	Forward	GGGCTCAATGTAGTTGCACA	qPrimerDepot
	Reverse	AGAAACCAACAGTCACAAGCA	
Tmprss2	Forward	CTCCCGTAGCTCTCACTCCA	qPrimerDepot
	Reverse	CCCAACGGAGAAGATGAGAA	