## Supplemental Table

Amplicons	Directions	Sequences
Cre fragment 1-465 pb	Forward	GGGGAGATTTGTGTGGGTCGACACCATGCCCAAGAAG
	Reverse	GTAACCTTGATACTTACACCTGGTCGAAATCAGTGCGTTC
Cre fragment 466-1523 pb	Forward	TCGTTCACTCATGGAAAATAGCG
	Reverse	GTACAAGAAAGCTGGGTAAAGCTTGTCCGCCACACCCAG
Intron BGH-Ig	Forward	GTAAGTATCAAGGTTACAAGACAGGTTTAAG
	Reverse	CTATTTTCCATGAGTGAACGACTGTGGAGAGAAAGGCAAAGTG
Intron Prm2	Forward	AAGTAGAGGGCTGGGCTG
	Reverse	CCATGAGTGAACGAACCTAGAAAGGTAAGAAAAGTG
Intron Prm2-AG	Forward	GTAGAGGGCTGGGCTGGGC
	Reverse	CCATGAGTGAACGAACCTAGAAAGGTAAGAAAAGTG
PSEBC	Forward	GGATCCGTCGAATTTAAATAAATCTAGCTGATATAGTGTGGC
	Reverse	ATACGAAGTTATTGCGCAGGCTGGGGAGCCTCCCCCAG
Primer set 1 (Luc)	Forward	TATCTCTTCATAGCCTTATGCA
	Reverse	GGTAAAGCCACCATGGAAGA
	Probe	AGGCCCGGCGCCATTCTATCCGCTGGA
Primer set 2 (stop cassette)	Forward	GTGCCTTCTAGTTGCCAG
	Reverse	ATAGAATGACACCTACTCAGACA
	Probe	TGCCCCTCCCCGTGCCTTCCTTGA

## Table S1: Primers for plasmid constructions and RT-qPCR

## **Supplemental Figures and Legends**

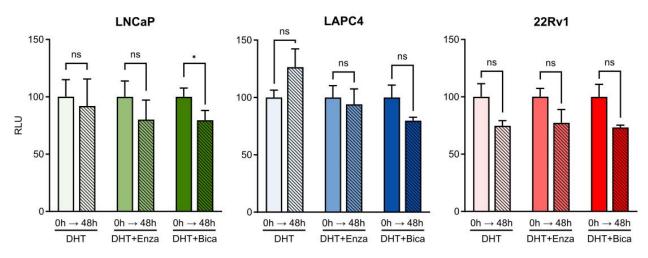
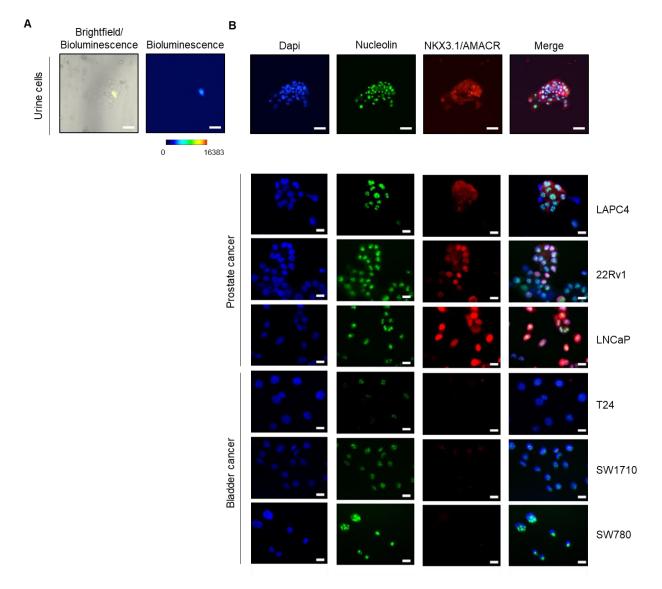


Figure S1. Bioluminescent plate reader cannot monitor dynamic prostate cancer cell line antiandrogen sensitivity after *PCA3*-Cre-*PSEBC*-ITSTA transduction. Luciferase activity was measured in the same wells, before and after 48 h of treatment (DHT, DHT + Bica or DHT + Enza) of ARAT responsive (LNCaP and LAPC4) and non-responsive (22Rv1) prostate cancer cells infected with *PCA3*-Cre-*PSEBC*-ITSTA. The luciferase activity is represented as relative activity over initial measurement (t = 0 h). The data represents mean of triplicates  $\pm$  S.D. Data were compared by paired Student's t-test. ARAT: androgen receptor-axis-targeted therapies; Bica: bicalutamide; DHT: dihydrotestosterone; Enza: Enzalutamide; ns: non significative; RLU: relative light unit.



**Figure S2. Immunofluorescence staining with multiple markers confirms the prostate cancer origin of urine cells expressing bioluminescence.** (A) Bioluminescence imaging of urine cells expressing *PCA3*-Cre-*PSEBC*-ITSTA system. (B) Immunofluorescence staining with multiple markers DAPI, Nucleolin, NKX3.1 and AMACR showed higher expression and were co-localized in prostate cancer cells. After bioluminescence imaging of cells collected from urine or 24 hours after cell line seeding, the cells were fixed in paraformaldehyde. The cells were exposed to antibodies against Nucleolin, NKX3.1 and AMACR as well as stained for DAPI. The wells were then imaged at 40X magnification using fluorescence microscope. Scale bars represent 50 µm for cells collected from urine and 20 µm for cell lines.