Figure S1. Dynamic *in vivo* HP MRSI of prostate cancer patients. Related to Figure 1 and Figure 2

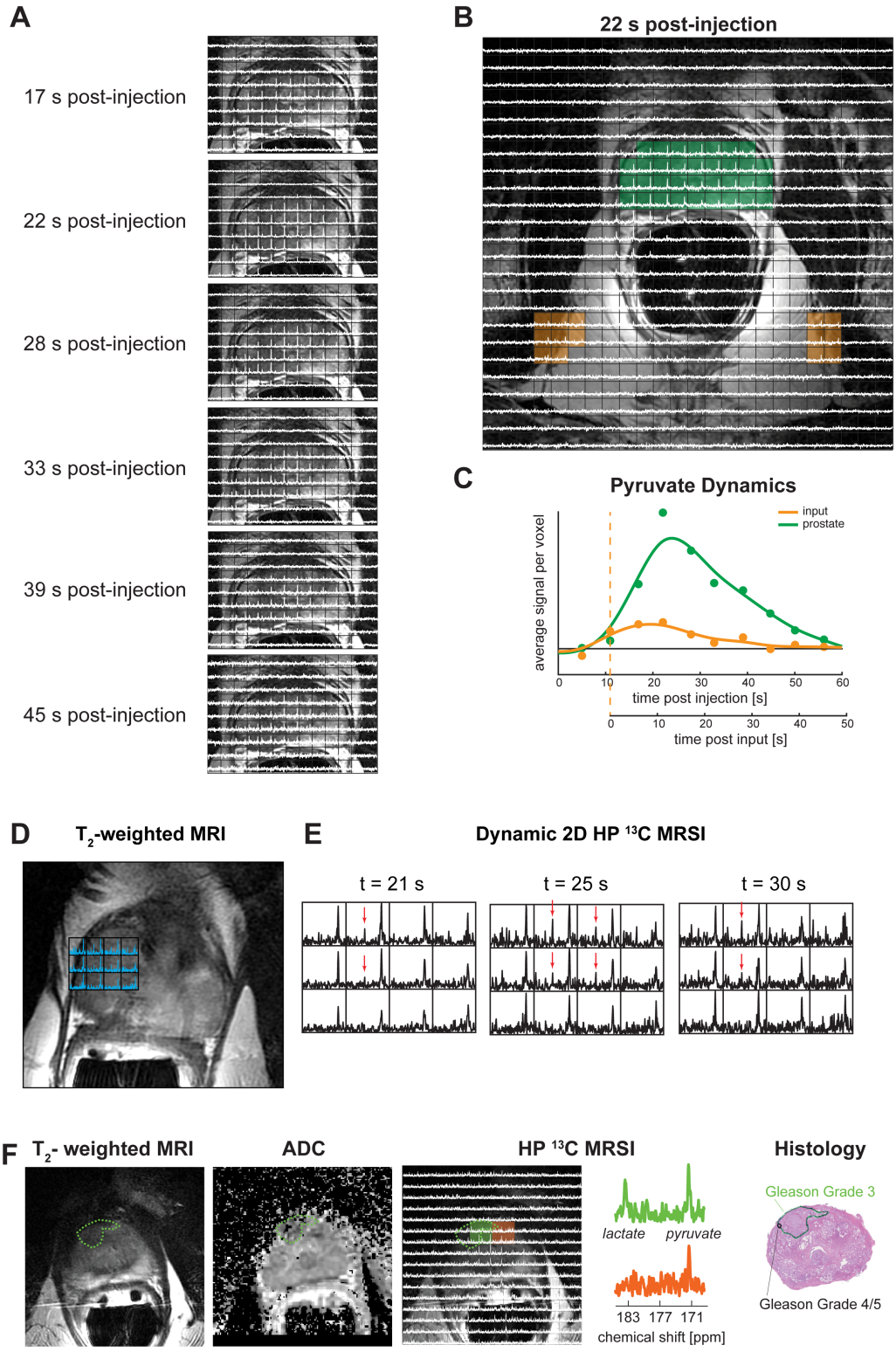

Figure S1. Dynamic *in vivo* HP MRSI of prostate cancer patients. Related to Figure 1 and Figure 2 (A) Spectra are shown overlaid on a T_2-weighted image for various time points corresponding to the data shown in **Fig. 2**. Pyruvate and lactate signal is localized in the prostate. The first time point (17 s post-injection) shows pyruvate delivery to the prostate. The maximum lactate ratio occurs at 33 s post-injection. (**B**) 2D overlay highlighting the prostate and input function voxels. (**C**) Arterial Input Correction. Pyruvate dynamics for the input function and the prostate. The first time point with a signal in the input is used to correct the timing of the dynamics, denoted time post input. **(D)** Representative T_2-weighted MRI and multiple voxels of ^{13}C data highlighting the region of interest in a patient with Gleason 4/5 prostate cancer. **(E) Detection of a HP bicarbonate in a region of prostate cancer.** Dynamic time points of the HP data at 21-30s post-injection. The resonance annotated by the red arrow corresponds to the aliased location of bicarbonate. (**F**) **Apical Tumor** The tumor region (dotted red outline) is shown on the T_2-weighted image and ADC map. Voxels with high lactate signal are shown in red overlaid on the T_2-weighted image. Voxels from the contralateral side are shown in green. Composite spectra from the two regions are also shown. The lesion is not conspicuous on the conventional images, but the region of high lactate corresponds to the tumor region on step-section pathology.

Figure S2. Clinical, pathologic and biochemical data correlations. **Related to Figure 3**

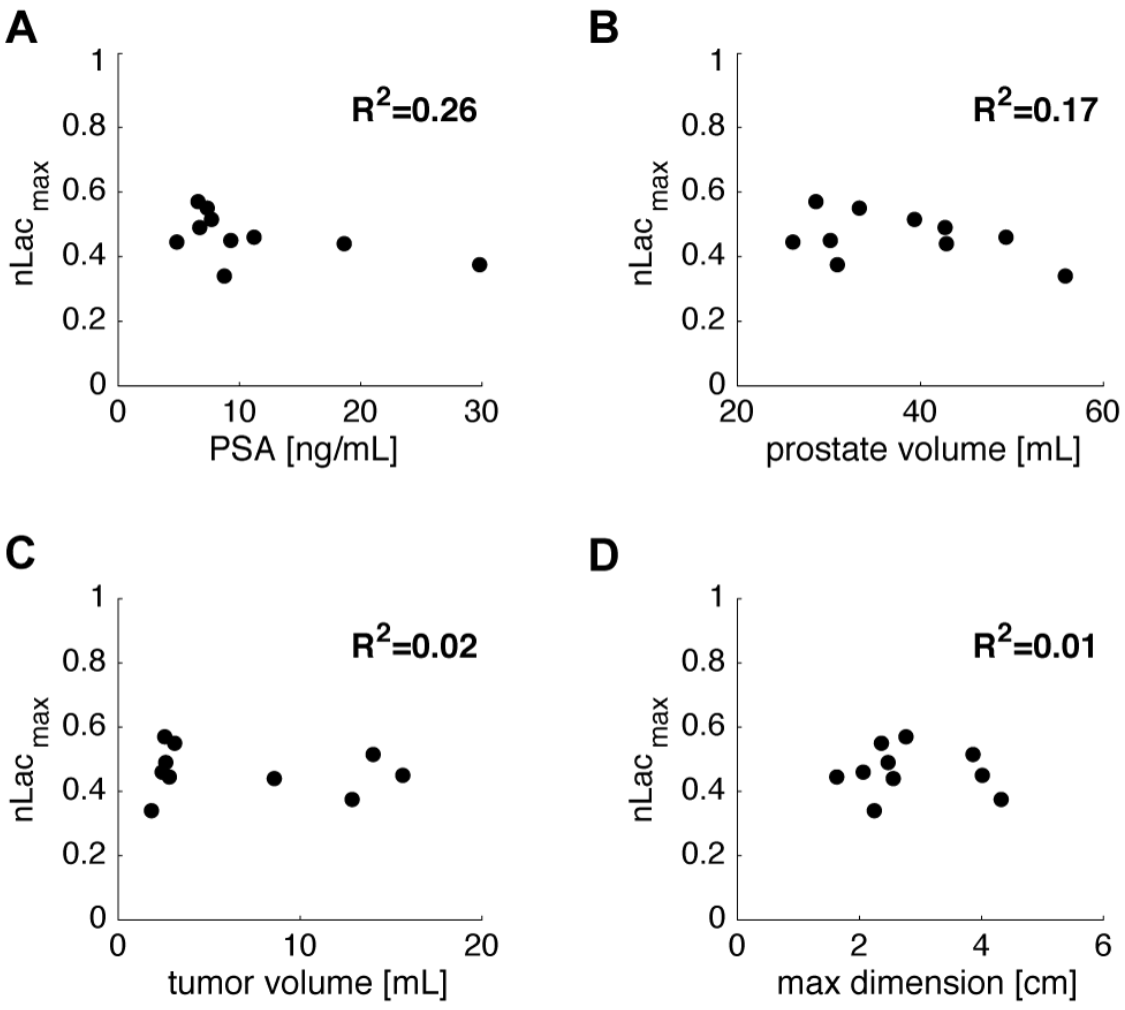

Figure S2. Clinical, pathologic and biochemical data correlations. Related to Figure 3 The maximum normalized lactate in a tumor region is weakly correlated with various clinical parameters, including (**A**) serum prostate-specific antigen, (**B**) prostate volume, (**C**) total tumor volume, and (**D**) maximum tumor dimension.

Figure S3. Gene expression data of LDH isoenzymes and MCT1/4 across tumor and normal prostate samples. Related to Figure 4

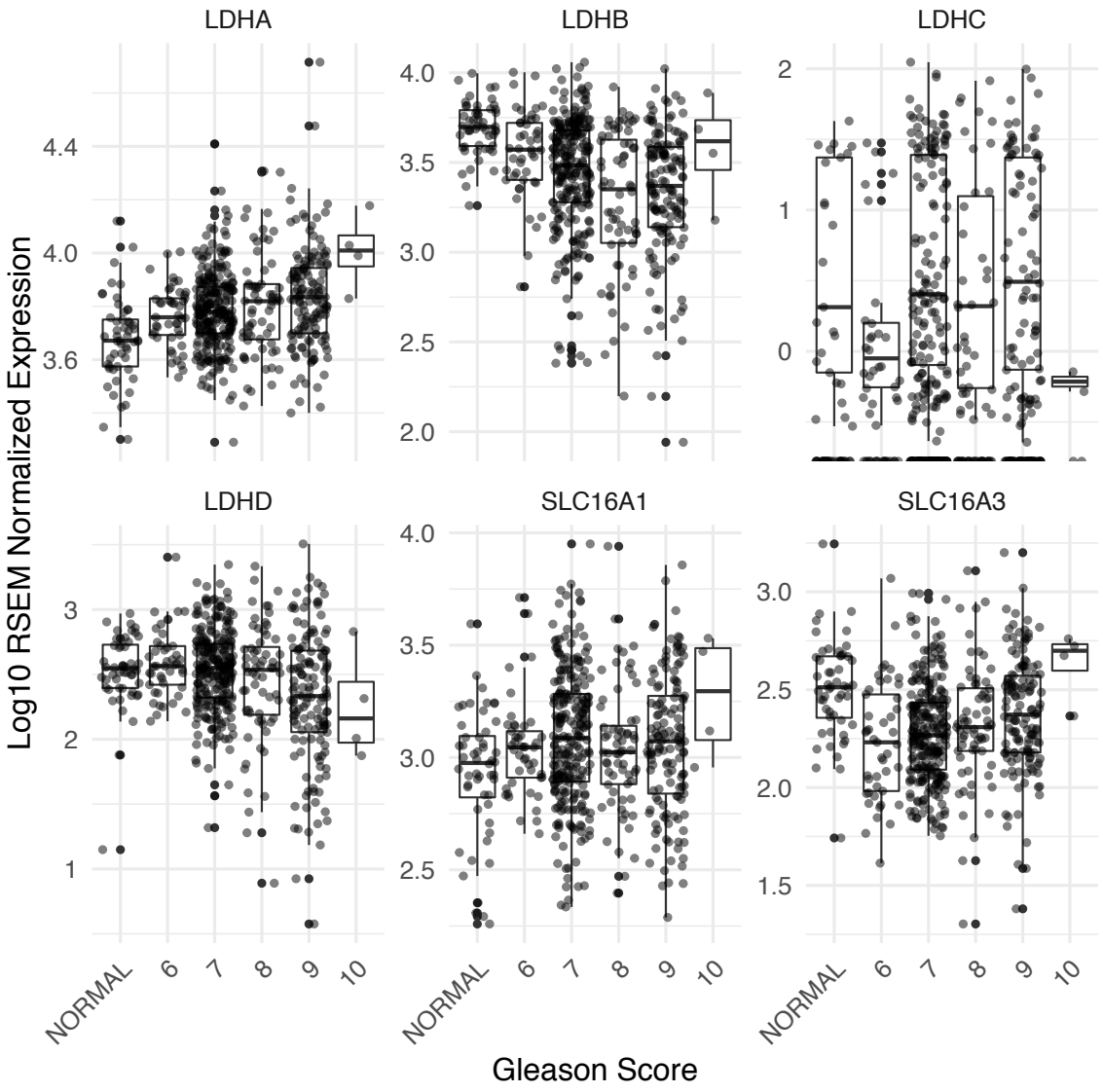

Figure S3. Gene expression data of LDH isoenzymes and MCT1/4 across tumor and normal prostate samples. Related to Figure 4 The expression of LDHA and MCT1 (SLC16A1) is elevated in tumors compared to normal tissue, and is further elevated in high Gleason grade tumors. Gleason score is determined as the sum of the primary and second Gleason grade.

Figure S4. Allele-specific copy number analysis indicates PTEN deletion in a subset of samples. Related to Figure 4

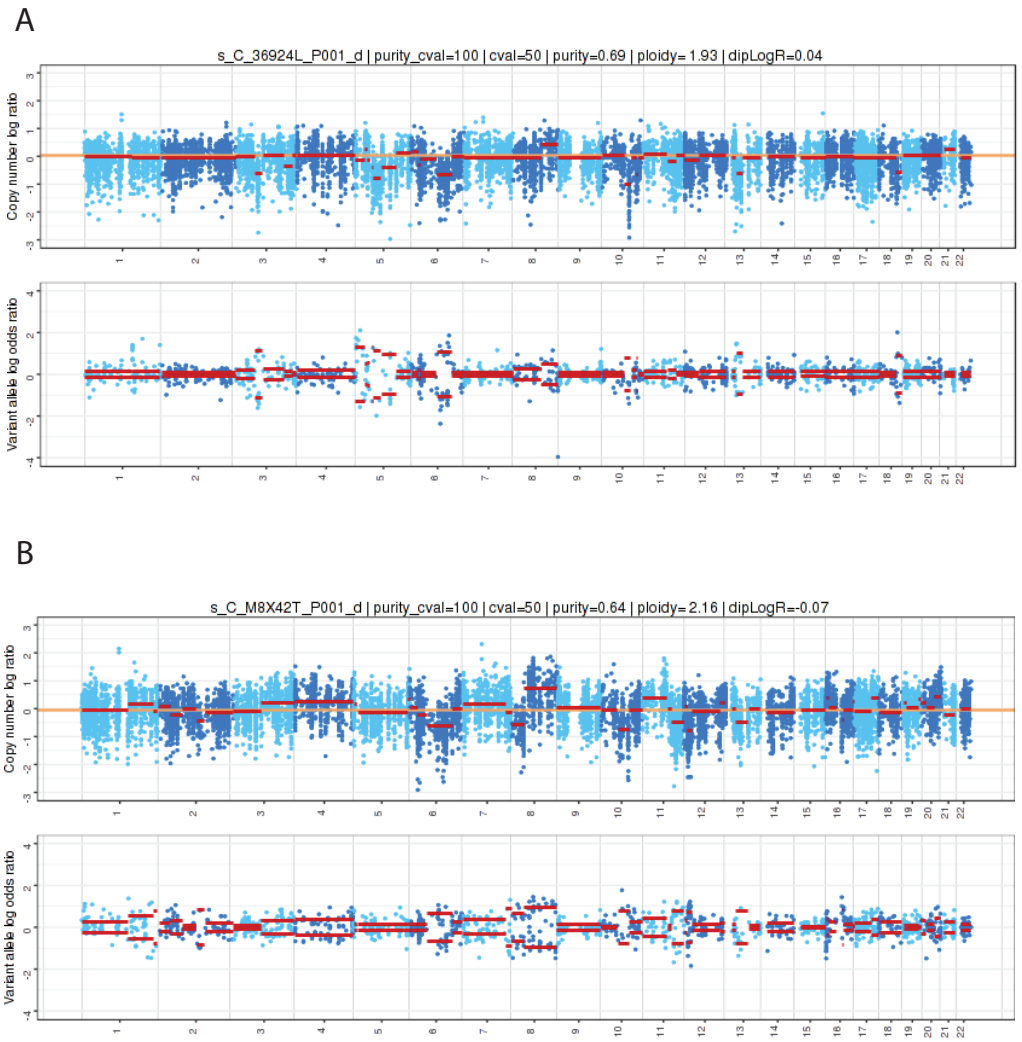

Figure S4. Allele-specific copy number analysis indicates PTEN deletion in a subset of samples. Related to Figure 4 Depicted are two cases demonstrating deep (homozygous) deletion of the *PTEN* locus on chromosome 10q.

Table S1. Patient Overview. Related to STAR Methods Experimental Model and Subject Details

Patient	Number of injections	Age at HP MRI	PSA [ng/mL]	Dominant prostatectomy Gleason Score	Tumor volume (G3/G4+) [cm^3]	Time since biopsy [days]	Time to prostatectomy [days]
1	2	69	11.2	4	0.26/2.15	36	29
2	2	51	9.28	4	0.02/15.63	65	228
3	1	72	6.05	N/A	N/A	70	N/A
4	1	57	8.75	3	1.79/0.03	108	18
5	1	68	18.63	4	0.13/8.45	60	6
6	1	55	6.58	4	<0.01/2.55	50	11
7	2	66	4.83	3	2.71/0.10	81	6
8	1	62	4.92	3	0.47/0.34	95	6
9	1	62	6.72	3	2.32/0.29	82	2
10	2	66	7.34	4	0.54/2.6	111	29
11	2	51	29.8	4	0.09/12.77	38	3
12	2	60	7.69	4	13.4/0.63	102	1
Mean		62	10.1		0.87/4.70	75	31
SD		7	7.2		1.09/6.03	26	66

Table S2. Dose Overview Related to STAR Methods Method Details

Dose	Patient	Pyruvate concentration [mM]	pH	EPA concentration [mM]	Temperature [°C]	Time to injection [s]	Dose volume [mL]	Polarization [%]	Polarization time [min]
1	1	244	7.3	3.3	35.9	59.8	32	10.7	141
2	1	244	6.3	2.2	34.9	37.2	32	21.4	315
3	2	245	7.6	0.8	31.9	35.6	34	15.1	358
4	2	247	6.6	1.2	35.7	40.7	34	22.2	420
5	3	247	7.5	0.5	36.1	48.7	25	23.5	310
6	4	246	7.8	0.9	34.9	80.9	35	45.9	
7	5	244	7.9	0.5	33.2	99.8	38	42.0	503
8	6	244	7.5	1.0	31.7	106.5	29	23.6	421
9	7	252	7.3	0.7	26.6	76.5	37	40.1	260
10	7	244	7.7	0.8	30.5	93.2	37	22.6	305
11	8	245	7.5	0.6	32.9	73.7	34	31.2	199
12	9	246	7.5	0.4	33.7	71.0	33	18.1	227
13	10	241	7.8	0.4	29.1	81.9	38	31.5	280
14	10	246	7.7	0.6	31.6	100.1	38	12.9	410

15	11	248	7.6	0.7	30.7	74.7	34	36.0	206
16	11	260	7.3	0.6	29.7	73.9	34	10.9	244
17	12	237	8.0	0.5	30.7	76.8	33	35.2	239
18	12	260	7.3	0.6	32.2	80.1	33	22.6	259
Mean		247	7.5	0.9	32.3	72.8	34	25.9	300
SD		**6**	0.4	0.7	2.6	21.3	3.3	10.8	96

Table S3. ROI Analysis. Related to Figure 4

ROI	Grade	Maximum Lactate Ratio	Time to maximum lactate ratio [s]	Cumulative Lactate Ratio
1	High	0.28	33	0.2332
2	Low	0.4313	45	0.1815
3	Mixed	0.4364	45	0.2602
4	High	0.3838	45	0.1753
5	Benign	0.2288	39	0.1388
6	High	0.301	45	0.1116
7	Low	0.2373	45	0.0885
8	High	0.2449	28	0.2437
9	Benign	0.3218	56	0.2062
10	High	0.4569	56	0.1862
11	Low	0.4285	45	0.247
12	(redundant section)			
13	Mixed	0.6383	40	0.412
14	Mixed	0.5559	40	0.3044

15		(redundant section)		
16	High	0.2907	56	0.1032
17	High	0.5462	56	0.162
18	Mixed	0.289	33	0.2491
19		(redundant section)		
20	Benign	0.1485	45	0.1485
21	Low	0.175	45	0.1730
22	Mixed	0.4268	39	0.4200
23	Mixed	0.442	61	0.2419
24	High	0.3243	56	0.2034
25		(redundant section)		
26	High	0.2831	28	0.2213
27	Benign	0.218	33	0.1018
28	High	0.3579	45	0.1829