Additional file 2

Table S1 Spartalizumab Preclinical Characteristics

Protein binding	Mean K_D (M) *n* = 3	SD (M)
Human PD-1/Fc	8.27E-10	5.05E-10
Cynomolgus PD-1/Fc	9.29E-10	1.50E-10
Mouse PD-1/Fc	No detectable binding	
Ligand blocking	**Mean IC_{50} (nM) *n* = 3**	**SD (nM)**
PD-L1	0.94	0.15
PD-L2	1.3	0.25

Fc, fragment crystallizable; IC_{50} half maximal inhibitory concentration; K_D dissociation constant; *PD-1* programmed death 1; *SD* standard deviation

Table S2 Adverse Events (Any Grade, Occurring in ≥10% of Patients, Regardless of Study Drug Relationship) by Treatment Group

Preferred term, n (%)	1 mg/kg Q2W *n* = 16		3 mg/kg Q2W *n* = 15		10 mg/kg Q2W *n* = 11		3 mg/kg Q4W *n* = 6		5 mg/kg Q4W *n* = 10		All patients *N* = 58	
	All	Gr 3/4	All	Gr 3/4	All	Gr 3/4	All	Gr 3/4	All	Gr 3/4	All	Gr 3/4
Total	16 (100)	12 (75.0)	15 (100)	10 (66.7)	11 (100)	5 (45.5)	6 (100)	3 (50.0)	10 (100)	6 (60.0)	58 (100)	36 (62.1)
Fatigue	10 (62.5)	1 (6.3)	3 (20.0)	2 (13.3)	2 (18.2)	0	2 (33.3)	0	5 (50.0)	1 (10.0)	22 (37.9)	4 (6.9)
Nausea	7 (43.8)	0	5 (33.3)	1 (6.7)	3 (27.3)	0	3 (50.0)	0	4 (40.0)	1 (10.0)	22 (37.9)	2 (3.4)
Anemia	6 (37.5)	3 (18.8)	7 (46.7)	3 (20.0)	2 (18.2)	1 (9.1)	1 (16.7)	1 (16.7)	3 (30.0)	1 (10.0)	19 (32.8)	9 (15.5)
Dyspnea	6 (37.5)	2 (12.5)	7 (46.7)	1 (6.7)	2 (18.2)	0	0	0	4 (40.0)	1 (10.0)	19 (32.8)	4 (6.9)
Diarrhea	7 (43.8)	0	6 (40.0)	1 (6.7)	1 (9.1)	0	0	0	3 (30.0)	0	17 (29.3)	1 (1.7)
Abdominal pain	3 (18.8)	1 (6.3)	4 (26.7)	2 (13.3)	2 (18.2)	0	2 (33.3)	1 (16.7)	3 (30.0)	0	14 (24.1)	4 (6.9)
Vomiting	6 (37.5)	0	1 (6.7)	0	3 (27.3)	0	2 (33.3)	0	2 (20.0)	1 (10.0)	14 (24.1)	1 (1.7)
Decreased appetite	5 (31.3)	1 (6.3)	1 (6.7)	0	3 (27.3)	0	2 (33.3)	0	2 (20.0)	1 (10.0)	13 (22.4)	2 (3.4)
Constipation	5 (31.3)	0	5 (33.3)	0	0	0	2 (33.3)	0	0	0	12 (20.7)	0
Weight decreased	4 (25.0)	1 (6.3)	2 (13.3)	0	1 (9.1)	0	1 (16.7)	0	3 (30.0)	0	11 (19.0)	1 (1.7)
Cough	3 (18.8)	0	4 (26.7)	0	0	0	0	0	3 (30.0)	0	10 (17.2)	0
Dizziness	4 (25.0)	0	6 (40.0)	0	0	0	0	0	0	0	10 (17.2)	0
Headache	1 (6.3)	0	4 (26.7)	0	1 (9.1)	0	1 (16.7)	0	3 (30.0)	0	10 (17.2)	0
Hypothyroidism	3 (18.8)	0	3 (20.0)	0	1 (9.1)	0	0	0	2 (20.0)	0	9 (15.5)	0
Pruritus	3 (18.8)	0	5 (33.3)	0	1 (9.1)	0	0	0	0	0	9 (15.5)	0
Arthralgia	0	0	3 (20.0)	0	1 (9.1)	0	1 (16.7)	0	2 (20.0)	0	7 (12.1)	0
Dehydration	0	0	3 (20.0)	1 (6.7)	1 (9.1)	0	2 (33.3)	0	1 (10.0)	0	7 (12.1)	1 (1.7)
Hyponatremia	2 (12.5)	2 (12.5)	3 (20.0)	1 (6.7)	1 (9.1)	1 (9.1)	0	0	1 (10.0)	1 (10.0)	7 (12.1)	5 (8.6)
Pleural effusion	3 (18.8)	0	1 (6.7)	0	1 (9.1)	1 (9.1)	0	0	2 (20.0)	1 (10.0)	7 (12.1)	2 (3.4)
AST increased	2 (12.5)	1 (6.3)	1 (6.7)	0	2 (18.2)	0	0	0	1 (10.0)	1 (10.0)	6 (10.3)	2 (3.4)

Dry mouth	3 (18.8)	0	1 (6.7)	0	1 (9.1)	0	0	0	1 (10.0)	0	6 (10.3)	0
Hypercalcemia	1 (6.3)	1 (6.3)	2 (13.3)	2 (13.3)	2 (18.2)	0	1 (16.7)	1 (16.7)	0	0	6 (10.3)	4 (6.9)
Peripheral edema	1 (6.3)	0	2 (13.3)	0	2 (18.2)	0	0	0	1 (10.0)	0	6 (10.3)	0
Pain in extremity	0	0	2 (13.3)	0	2 (18.2)	0	1 (16.7)	0	1 (10.0)	0	6 (10.3)	0
Pyrexia	2 (12.5)	0	2 (13.3)	0	1 (9.1)	0	0	0	1 (10.0)	0	6 (10.3)	0

7 *AST* aspartate aminotransferase; *Gr* Grade; *Q2W* once every 2 weeks; *Q4W* once every 4 weeks

Table S3 Adverse Events of Special Interest (Any Grade, Suspected to Be Related to Study Drug) by Treatment Group

Preferred term, n (%)	1 mg/kg Q2W *n* = 16	3 mg/kg Q2W *n* = 15	10 mg/kg Q2W *n* = 11	3 mg/kg Q4W *n* = 6	5 mg/kg Q4W *n* = 10	All patients *N* = 58
Diarrhea	4 (25.0)	4 (26.7)	1 (9.1)	0	1 (10.0)	10 (17.2)
Pruritus	3 (18.8)	5 (33.3)	0	0	0	8 (13.8)
Hypothyroidism	2 (12.5)	1 (6.7)	1 (9.1)	0	2 (20.0)	6 (10.3)
Maculopapular rash	1 (6.3)	2 (13.3)	0	0	0	3 (5.2)
Rash	1 (6.3)	1 (6.7)	0	0	0	2 (3.4)
Alopecia	0	0	1 (9.1)	0	0	1 (1.7)
ALT increased	0	0	1 (9.1)	0	0	1 (1.7)
AST increased	0	1 (6.7)	0	0	0	1 (1.7)
Autoimmune colitis	0	0	1 (9.1)	0	0	1 (1.7)
Blood TSH increased	0	0	0	0	1 (10.0)	1 (1.7)
Hyperglycemia	1 (6.3)	0	0	0	0	1 (1.7)
Lichen planus	0	0	0	0	1 (10.0)	1 (1.7)
PPE syndrome	0	1 (6.7)	0	0	0	1 (1.7)
Rash erythematous	1 (6.3)	0	0	0	0	1 (1.7)
Transaminases increased	1 (6.3)	0	0	0	0	1 (1.7)

ALT alanine aminotransferase; *AST* aspartate aminotransferase; *PPE* palmar-plantar erythrodysesthesia; *Q2W* once every 2 weeks; *Q4W* once every 4 weeks; *TSH*, thyroid-stimulating hormone

12 **Table S4** Baseline and On-treatment Biomarker Analyses, Per Patient

Diagnosis	CD8+ Staining, by IHC, %		PD-L1+ Cells, by IHC, %		BOR, RECIST v1.1
	Baseline	On-treatment	Baseline	On-treatment	
HCC	1.61	–	0	–	PD
Ovarian cancer	0.7	C2D1 2.81	0	C2D1 5	SD
Liposarcoma	0.38	C2D1 0.21	–	C2D1 0	SD
Liposarcoma	0.23	C2D1 0.16	0	–	SD
Neuroendocrine carcinoma	0.31	C2D1 0.03	2	C2D1 1	PD
mRCC	0.41	–	0	–	UNK
Sarcoma	0.01	–	0	–	NCRNPD
Neuroendocrine carcinoma	0.02	C2D1 0.23	0	C2D1 1	SD
Sarcoma	0.14	–	0	–	UNK
Prostate cancer	0.03	C2D1 0.07	0	–	NCRNPD
Other	0.01	C2D1 1.12	0	C2D1 5	SD
Other	11.81	C2D1 25.88	40	C2D1 100	SD
mRCC	2.24	C2D1 1.71	95	C2D1 60	PD
HCC	0.75	C3D1 0.88	0	C3D1 0	PD
Testicular cancer	0.0029	–	0	–	PD
H&N cancer	0.69	C2D1 0.75	10	C2D1 5	SD
mRCC	1.49	C3D1 0.67	0	C3D1 0	PD
SCLC	1.49	–	80	–	UNK
Cholangiocarcinoma	0.17	C2D1 0.52	0	–	PD
Urothelial carcinoma	0.35	C2D1 0.83	10	C2D1 1	PD
Liposarcoma	0.02	C3D1 18.77	0	C3D1 80	SD
Sarcoma	1.53	C2D1 2.19	1	C2D1 3	SD

ATC	1.51	C2D1	15.7	30	C2D1 75	SD
Carcinoid	5.61	C2D1	37.55	0	–	PR
H&N cancer	0.68	–		0	–	UNK
Sarcoma	0.01	–		0	–	SD
Other	0.51	C2D1	0.24	1	C2D1 1	SD
Mesothelioma	2.49	–		2	–	PD
Sarcoma	0.85	–		0	–	PD
TNBC	1.62	C2D1	2.5	0	C2D1 10	SD
NSCLC	0.41	C2D1	0.42	0	C2D1 0	PD
SCLC	0.75	–		0	–	UNK
mRCC	0.36	C3D1	1.98	0	C3D1 0	PD
Anal cancer	0.46	C2D1	2.74	10	C2D1 50	PR
Sarcoma	0.1	–		20	–	UNK
Merkel cell carcinoma	0.0147	–		0	–	UNK
Anal cancer	1.23	–		50	–	UNK
Cholangiocarcinoma	0.63	–		0	–	PD
Breast cancer	1.46	C3D1	2.1	0	C3D1 5	PD
Sarcoma	1.84	C2D1	0.5	0	C2D1 0	SD
Esophageal cancer	0.57	–		0	–	PD
Cutaneous melanoma	–	–		–	C3D1 0	SD
Sarcoma	0.24	–		–	–	SD
mRCC	2.37	C2D1	0.96	0	C2D1 10	PD
Urothelial carcinoma	1.72	–		0	–	SD
Esophageal cancer	0.57	C2D1	0.96	0	C2D1 1	PD

Ovarian cancer	1.12	–		0	–		PD
Sarcoma	0.16	C2D1	0.14	0	C2D1	0	SD
mRCC	4.95	C2D1	2.14	20	C2D1	0	PD
SCC of skin	0.15	–		10	–		PD
Sarcoma	0.17	C3D1	0.34	80	C3D1	60	PD
H&N cancer	0.03	C2D1	0.17	0	C2D1	0	PD
Sarcoma	0.01	C3D1	0.01	0	C3D1	0	SD
Solid tumor	0.09	–		0	–		PD
Merkel cell carcinoma	4.22	–		0	–		NCRNPD
Breast cancer	2.5	–		0	–		PD
Sarcoma	0.01	C2D1	0.07	0	C2D1	1	PD
Sarcoma	0.16	C2D1	0.61	0	C2D1	0	UNK

ATC anaplastic thyroid cancer; *BOR* best overall response; *C2D1* Cycle 2 Day 1; *C3D1* Cycle 3 Day 1*; CD8* cluster of differentiation 8; *CR* complete response; *H&N* head and neck; *HCC,* hepatocellular carcinoma; *IHC,* immunohistochemistry; *mRCC* metastatic renal cell carcinoma; *NCRCPD* non-CR/non-PD; *NSCLC* non-small cell lung cancer; *PD progressive disease; PD-L1* programmed death-ligand 1; *PR* partial response; *RECIST* Response Evaluation Criteria in Solid Tumors; *SCC* squamous cell carcinoma; *SCLC* small cell lung cancer; *SD* stable disease; *TNBC* triple negative breast cancer; *UNK* unknown.

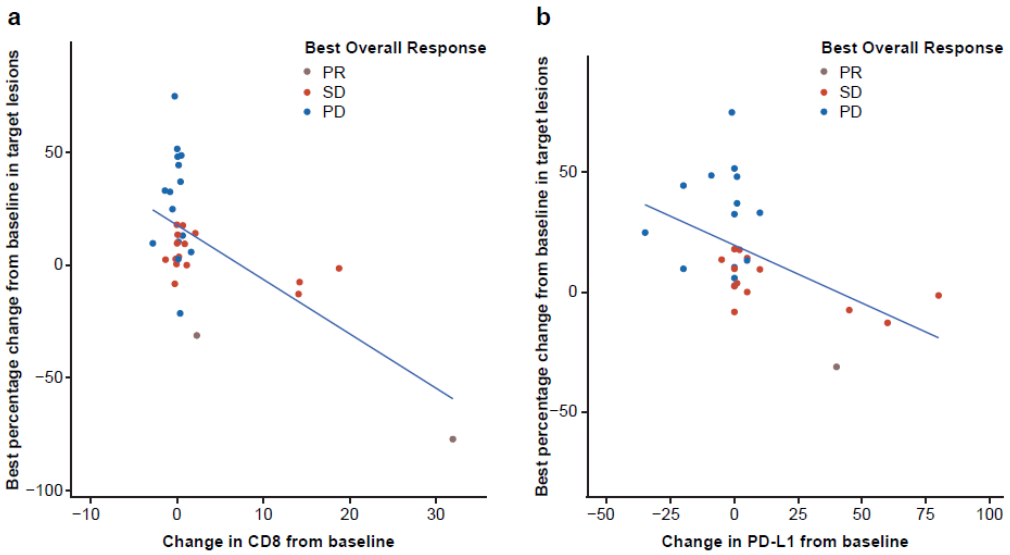

Fig. S1 Best Percentage Change From Baseline in Target Lesions, by Change From Baseline in Biomarker Levels. (**a**) Best percentage change in target lesions vs change in percentage of CD8+ staining in tumor biopsy samples. Data were available for n=32 patients; Spearman coefficient = -0.42. (**b**) Best percentage change in target lesions vs change in percentage of PD-L1+ cells in tumor biopsy samples. Best overall response is shown for each patient according to RECIST v1.1. Data were available for n=28 patients; Spearman coefficient = -0.49.
CD8, cluster of differentiation 8; PD, progressive disease; PD-L1, programed death ligand 1; PR, partial response; RECIST, Response Evaluation Criteria In Solid Tumors; SD, stable disease; UNK, unknown.