



Supplementary figure 1. Representative TCR V β spectratyping profiles of T-cells from healthy donors and co-inhibitory receptor ligand expression on mature monocyte-derived DCs utilised during the *in vitro* antigen-stimulation of naïve and memory T-cells. (A, B) TCR V β spectratyping profiles of T-cells from healthy donors (A) S2 and (B) S4 showing skewed CDR3 lengths distribution after *in vitro* priming of naïve T-cells to SMX-NO. The expression of (C) the PD-1-ligand PD-L1 (PD-L1-PE), (D) the CTLA4-ligands CD80 (CD80-FITC) and CD86 (CD86-FITC), and (E) the TIM-3-ligand galectin-9 (Gal-9-APC*) on LPS- and TNF- α -matured DCs (Grey histogram, unstained cells; blue histogram, CD14 $^{+}$ monocyte-derived mature DCs).

Supplementary table 1. Flow cytometric analysis of TCR V β usage on naïve and SMX-NO specific memory T-cells.

TCR	Day 0 - % naive T-cells					Mean day0 ¹ (excluding S1)	SD	mean -3SD	mean +3SD	Day 11- % SMX-NO spec. memory T-cells				
	S2	S3	S4	S5	S6					S2	S3	S4	S5	S6
Vb 1	4.66	3.96	4.28	3.69	3.76	4.07	0.40	2.87	5.27	3.17	1.28	1.82	1.93	5.59
Vb 2	7.12	8.49	6.65	7.29	5.34	6.98	1.14	3.56	10.40	18.12	0.88	0.62	1.30	9.89
Vb 3	0.67	0.43	10.64	4.60	6.39	4.55	4.26	-8.23	17.32	1.24	0.57	6.81	3.57	4.11
Vb 4	0.21	0.07	0.09	0.53	0.03	0.19	0.20	-0.42	0.80	2.85	3.27	3.37	5.24	3.92
Vb 5.1	4.53	8.54	6.82	6.88	4.44	6.24	1.75	1.00	11.48	5.34	0.86	0.41	0.30	3.32
Vb 5.2	0.77	0.80	0.33	0.61	0.91	0.68	0.23	0.01	1.36	0.73	10.65	7.88	12.22	1.00
Vb 5.3	0.78	0.26	0.21	0.40	0.77	0.48	0.27	-0.34	1.31	1.16	3.88	4.25	2.88	0.82
Vb 7.1	2.91	1.95	1.04	2.59	1.62	2.02	0.75	-0.22	4.27	5.36	0.70	0.73	1.12	1.64
Vb 7.2	2.53	0.60	0.06	0.35	0.03	0.71	1.04	-2.41	3.84	3.11	1.98	1.00	1.47	0.00
Vb 8	3.55	1.76	2.34	1.92	3.52	2.62	0.86	0.03	5.21	3.72	5.00	3.89	3.30	3.74
Vb 9	0.18	0.22	0.15	0.15	0.07	0.15	0.06	-0.01	0.32	3.84	3.45	4.47	6.85	2.27
Vb 11	0.75	0.87	0.77	0.73	0.58	0.74	0.10	0.43	1.05	1.18	4.69	5.66	2.92	0.65
Vb 12	1.17	1.64	1.32	1.22	0.68	1.21	0.35	0.17	2.24	1.52	1.78	2.32	1.61	1.10
Vb 13.1	3.27	4.27	2.84	3.27	3.57	3.44	0.53	1.85	5.03	4.62	3.35	2.81	3.66	5.03
Vb 13.2	3.91	2.34	0.97	1.73	0.60	1.91	1.31	-2.01	5.83	3.92	0.49	0.57	0.51	0.42
Vb 13.6	1.57	1.62	1.29	1.79	1.61	1.58	0.18	1.03	2.12	1.45	6.02	2.75	2.93	2.59
Vb 14	2.19	0.89	2.47	0.85	0.89	1.46	0.80	-0.95	3.86	6.05	4.83	6.59	5.44	3.70
Vb 16	0.55	0.86	0.75	0.82	1.09	0.81	0.20	0.23	1.40	0.38	1.37	0.69	1.15	0.62
Vb 17	6.90	3.45	6.20	6.44	4.68	5.53	1.43	1.24	9.83	8.00	1.64	1.91	1.06	4.97
Vb 18	0.52	0.11	0.17	0.12	0.39	0.26	0.18	-0.29	0.81	0.49	10.40	7.89	8.05	1.02
Vb 20	1.33	2.18	0.96	2.14	1.43	1.61	0.53	0.01	3.21	2.68	2.35	0.67	1.93	2.25
Vb 21.3	1.63	2.54	2.67	2.46	1.82	2.22	0.47	0.82	3.62	1.72	1.55	1.69	1.93	1.70
Vb 22	3.10	3.47	4.02	1.79	4.00	3.28	0.92	0.53	6.02	1.88	1.14	1.56	0.65	3.87
Vb 23	0.79	0.67	0.29	0.57	1.30	0.72	0.37	-0.39	1.84	0.54	5.65	0.37	4.21	0.85
total Vb	55.59	51.95	57.33	52.95	49.52					83.08	77.79	70.72	76.23	65.09

¹ Donor 1 excluded from mean calculation

Supplementary Table 2. Summary of the antigen-specific proliferation of SMX-NO-primed naïve T-cells ± PD-L1/CTLA4/TIM-3 blocking antibodies.

Block ¹ used	Donor number	- block drug Response (CPM)	+ block drug Response (CPM)	SI of comparative response with addition of block
PD-L1	Donor 1	weak (3549.0)	good (15214.0)	↑ 4.3 (p=0.002)
	Donor 2	weak (2543.3)	weak (12360.6)	↑ 4.9 (p=0.038)
	Donor 3	weak (13318.3)	very stong (63258.3)	↑ 4.7 (p=0.012)
	Donor 4	weak (7689.0)	good (29107.6)	↑ 3.8 (p=0.020)
	Donor 5	very stong (51009.6)	good (29463.6)	No increase
CTLA4	Donor 6	weak (5475.3)	weak (9750.3)	↑ 1.8 (p=0.043)
	Donor 7	good (19496.6)	strong (32843.3)	↑ 1.7 (p=0.027)
	Donor 8	good (15668.3)	strong (36262.0)	↑ 2.3 (p=0.023)
	Donor 9	weak (8050.0)	very strong (49669.3)	↑ 6.2 (p=0.006)
	Donor 10	very strong (50829.6)	very strong (68964.3)	↑ 1.4 (p=0.017)
	Donor 5	good (19659.0)	strong (36279.6)	↑ 1.8 (p=0.010)
	Donor 11	good (25933.6)	weak (14800.6)	No increase
	Donor 12	weak (8802.6)	weak (7135.6)	No increase
	Donor 4	strong (39129.3)	strong (38245.0)	No increase
TIM-3	Donor 13	very strong (50698.0)	very strong (54427.0)	No increase
	Donor 14	strong (43080.3)	good (23292.0)	No increase
	Donor 5	very strong (50357.3)	strong (44196.0)	No increase

¹Naïve T-cells from healthy donors were cultured with mature autologous DCs in the presence of SMX-NO (50 µM) for one week, ± PD-L1 (5 µg/ml) /CTLA4 (10 µg/ml) /TIM-3 (7.5 µg/ml) blocking antibodies before restimulation with fresh DCs and SMX-NO (20-50 µM). Strength of proliferative response was measured by [³H] thymidine incorporation and is shown as CPM above background; weak (< 15000), good (15000-30000), strong (30000-45000), very strong (> 45000). An enhanced response is shown by significant fold increase with block compared to without (*p ≤ 0.05).

Supplementary Table 3. Summary of the antigen-specific proliferation of SMX-NO-exposed memory T-cells ± PD-L1/CTLA4/TIM-3 blocking antibodies.

Block used	Donor number	- block drug Response (CPM)	+ block drug Response (CPM)	SI of comparative response with addition of block
PD-L1	Donor 15	weak (13750.6)	weak (8158.0)	No increase
	Donor 5	strong (32780.0)	good (24932.3)	No increase
	Donor 16	weak (10167)	weak (8338.6)	No increase
CTLA4	Donor 5	weak (14443.6)	good (22310.3)	↑ 1.5 (p=0.022)
	Donor 9	weak (7470.3)	weak (9981.6)	↑ 1.3 (p=0.009)
	Donor 8	weak (981.0)	weak (5666.6)	↑ 6.6 (p=0.002)
	Donor 11	weak (8032.6)	weak (6903.3)	No increase
	Donor 6	weak (8500.3)	weak (5548.0)	No increase
	Donor 10	very strong (47951.3)	good (29999.6)	No increase
TIM-3	Donor 5	strong (33874.0)	strong (34144.3)	No increase
	Donor 15	weak (13750.6)	good (19919.3)	No increase
	Donor 13	good (17515.3)	weak (13202.3)	No increase

¹Memory T-cells from healthy donors were cultured with mature autologous DCs in the presence of SMX-NO (50 µM) for one week, ± PD-L1 (5 µg/ml) /CTLA4 (10 µg/ml) /TIM-3 (7.5 µg/ml) blocking antibodies before restimulation with fresh DCs and SMX-NO (20-50 µM). Strength of proliferative response was measured by [³H] thymidine incorporation and is shown as CPM above background; weak (< 15000), good (15000-30000), strong (30000-45000), very strong (> 45000). An enhanced response is shown by significant fold increase with block compared to without (*p ≤ 0.05).