

Supplementary Online Content

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This supplementary material has been provided by the authors to give readers additional information about their work.

**eTable 1. Overall Toxicities Possibly Related to Ipilimumab for Run-In
Phase 1 and Phase 2**

1A. Clinical toxicities

Adverse Events	All Grade n pts (%)	Grade \geq3 n pts
Fatigue	16 (38.1%)	2
Anemia	11 (26.2%)	4
Nausea	11 (26.2%)	1
Anorexia	10 (23.8%)	1
Vomiting	7 (16.7%)	1
Dehydration	5 (11.9%)	2
Edema limbs	4 (9.5%)	0
Dyspnea	3 (7.1%)	1
Abdominal pain	3 (7.1%)	0
Weight loss	2 (4.8%)	1
Back pain	2 (4.8%)	0
Pain	2 (4.8%)	0
Lung infection	1 (2.4%)	1

1B. Biological Toxicities

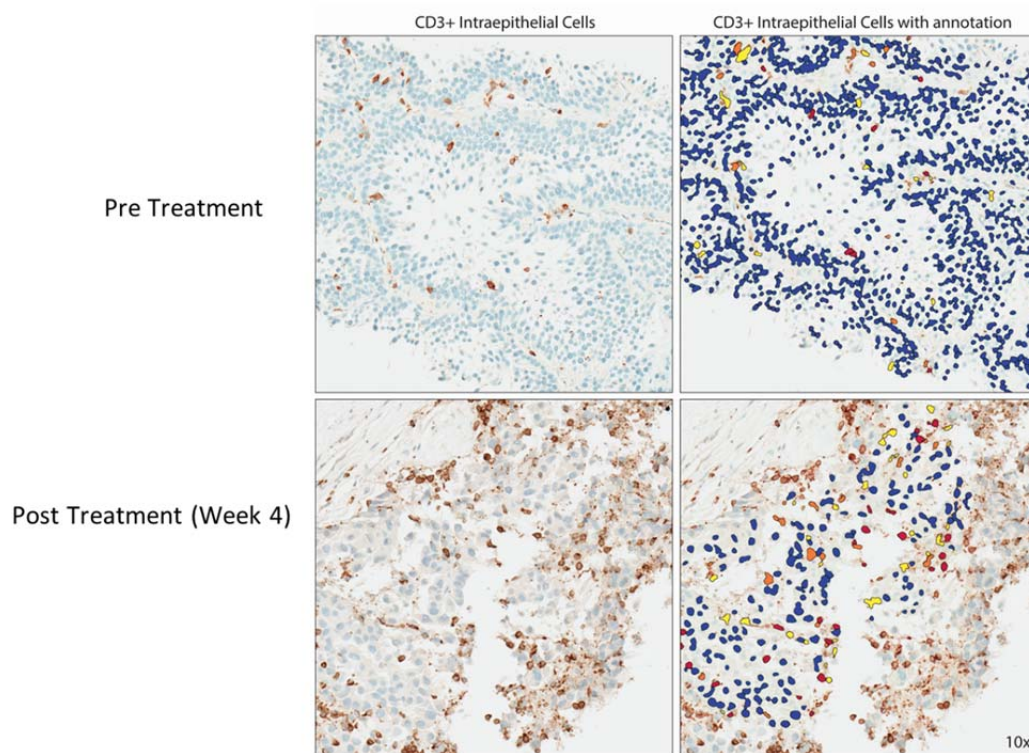
Adverse Events	All Grade n pts (%)	Grade \geq3 n pts
Lymphocyte count decreased	7 (16.7%)	1
Hypo albuminemia	7 (16.7%)	1
Hypomagnesemia	5 (11.9%)	1
Hyponatremia	4 (9.5%)	3
Alkaline phosphatase increased	4 (9.5%)	0
Hypokalemia	2 (4.8%)	1
Hypo phosphatemia	2 (4.8%)	2
Hypocalcemia	2 (4.8%)	1
Hypercalcemia	1 (2.4%)	1

eTable 2. DAB-Stained for Immune Markers as Part of the Trial.

Antibody clone	Source	Host	Product No	Dilution	Ventana XT/incubate	Control tissue	Staining pattern	Program
CD3-2GV6	Ventana/ Roche (Tucson, AZ, USA)	Rb mono	52784220 01	dispenser	CC1m/20m	spleen	membrane	P305/UV
CD4-SP35	Ventana/ Roche	Rb mono	55527370 01	dispenser	CC1s/Option 5- blocking/20m	spleen	membrane	P306/UV
CD8-SP57	Ventana/ Roche	Rb mono	59372480 01	dispenser	CC1s/Option 5- blocking/20m	spleen	membrane	P307/UV
FOXP3- 236A/E7	Abcam Inc (Cambridge, MA, USA)	Mouse mono	ab20034	1/100	CC1s/60m	spleen	nucleus	P311/UV
PD-L1- E1L3N	CST (Boston, MA, USA)	Rb mono	13684	1/100	CC1e/120m	tonsil	membrane / cytoplasm	P338/UV
IDO-1F8.2	EMD Millipore (Darmstadt, Germany)	Mouse mono	MAB1000 9	1/300	CC1s/60m	spleen	cytoplasm	P359/UV
ultraView Universal DAB detection kit	Ventana/ Roche	-	760-500	-	-	-	-	ultraView (UV)

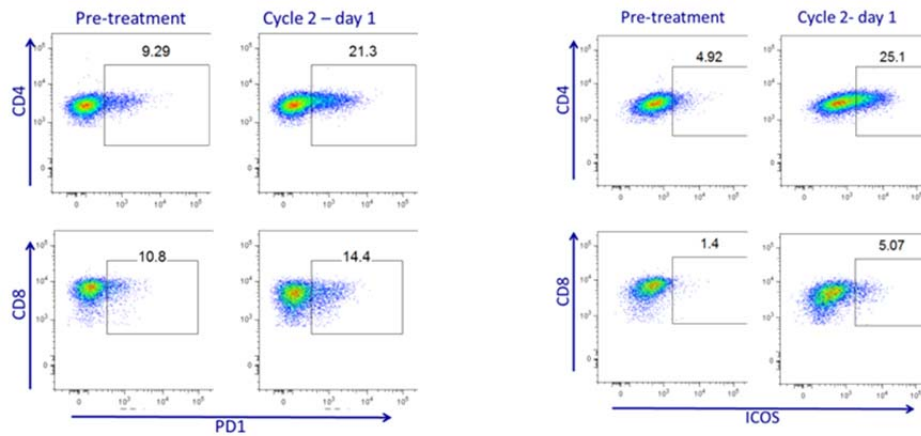
eFigure 1. Immunohistochemistry on Paired Tumor Biopsies.

Increase in CD3 staining indicate T cell infiltration post treatment.



Pre/Post treatment Biopsy: Formalin-fixed, paraffin-embedded FFPE samples were DAB-stained for immune markers using the BenchMark XT (Ventana/Roche, Tucson, AZ, USA) (eTable 2) at Princess Margaret Cancer Centre. DAB-stained slides (CD3, CD4, CD8, FOXP3) were digitized (Aperio Scanscope XT, Leica Biosystems Inc., Buffalo Grove, IL, USA) and analyzed for percentage and intensity of staining by automated image analysis (AperioTumor-rich and tumor-negative zones were annotated and analysed separately). PD-L1 and IDO expression were scored manually for percentage cells positive.

eFigure 2. Flow Cytometry Plots for PD-1 and ICOS on CD4+ and CD8+ CD3+ T Cells.



Blood Analysis: Treg Panel: CD3, CD45RA, CD194 (Biolegend); CD25, CD127, FOXP3, CD4 (Thermofisher); **Th Subset Panel:** CD4, CD8, CD161, CD196, CD183, CD194 (Thermofisher); **NK Panel:** CD314 (R&D); CD3, CD56, CD16, CD314, CD335, CD337 (Thermofisher); **T cell memory Panel** (CD3, CD8, CD45RA, CD28, CD95 (Thermofisher); CCR7, CD4 (Biolegend) and **T cell activation/exhaustion Panel:** CD4, PD1 (Biolegend); CD3, CD4, CD8, ICOS, HLA-DR, BTLA (Thermofisher). CD14 and CD19 were used for the dump channel in all the panels