





a Patient 0101



b Patient 0201



c Patient 0401









Fig 5

Day 7+ pep-PDC line



С





Table 1 : overall demographics and activity

										Clini		
										cal		
										resp		
					Nb of prior				Clinical	onse	Melan A	
				Stage	systemic	Prior			response	IrRC	immune	Clinical
ID	Age	sex	TNM	(AJCC 2009)	treatments	immunotherapy	Cohort	Dose	RECIST§	§	response	outcome
								(cells			(fold	
	(years))			increase)	
		_	T4N3M1c					4x10				
0101	/5	F		IV	3	-	1	0	PD	PD	1.1/ •	alive
0004		_	T3N2bM1c					4x10	55		20*	D (22)
0201	80	F		IV	1	-	1	4.10	PD	PD	30*	D (32)
0301	35	М	T4bN1bM1c	IV	2	ICI(2)	1	4X10 6	PD	PD	1.1	D (9)
			= 41					20x1				
0401	61	М	14bN3M1c	IV	1	-	2	06	PD	PD	5.4*	D (26)
								20x1				
0501 84	84	F	TZXINDIVITQ	IV	2	IFN AD	2	0 ⁶	PD	PD	NI	alive
			T3bN2bM1a					20x1				
0103	72	М	13511251111	IV	1	IFN AD, VACCIN	2	06	PD	-	NI	D (20)
			T3aN1bM1a			VACCIN(2), TIL,		60x1				
0203	0203 76	F		IV	6	ICI(2)	3	06	PD	PD	1	D (18)
			T4bN2cM1b					60x1				
0403	79	M		IV	1	-	3	06	SD	-	NI	alive
			T1xN3M1a					60x1				
0601	79	F		IV	2	-	3	06	SD	SD	1.8	alive

ICI: immune checkpoint inhibitor

IFN AD: interferon alpha adjuvant

TIL: tumor infiltrating lymphocytes

D (): death (weeks post first vaccine injection)

NI: not interpretable

*: significant increase associated with naive/memory T cell switch

*: Fold increase calculated from W1, since W0 data were missing

§: RECIST and IrRC results at the end of patient follow-up

Supplementary figures legend

Supplementary figure 1

Immunomonitoring of immune cell subsets by flow cytometry. A) gating strategy used to determine the percentages of the different cell subsets with the 3 different panels of antibodies used (see suppl table 1 for antibodies references). Analyses were performed with FlowJo software. B) Kinetic analysis of immune cell subset percentages in the different patients. C) Gating strategy for CD45RA/CCR7 staining

Supplementary Figure 2

Total number of adverse events observed during the clinical trial, according to patient group dose, with their localizations

Supplementary Figure 3

MLANA specific T cells percentages and naive/memory frequencies. Specific anti-MLANA T cells were characterized in terms of naïve/memory profile, according to their differential expression of CCR7 and CD45RA (see methods), at different time points during patients follow-up.



Suppl Fig 1, continued



Suppl Fig 1, continued

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Suppl Fig 1, continued

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Gating strategy (patient 0401, W16)



Suppl Fig 2



Suppl Fig 3



Suppl Table 1

Product	Supplier	Réferences	
Monitoring immune cell subsets:			
VA TRANK (CDD) FITC (CD16 DE + CD56 DE (CD46 DecCD C-6 E)			
KIT IBNK (CD3 FITC / CD16 PE + CD36 PE / CD45 PerCP-Cy5.5 /	BD-Biosciences	337166	
CD4 PE-CY// CD19 APC/ CD8 APC-CY/)			
HLA-DR-BV421	BD-Biosciences	562804	
Lin-FITC	BD-Biosciences	340546	
CD123 -PE	BD-Biosciences	370545	
CD11c-PE Cy5-5	BD-Biosciences	551077	
		+	
HLA- ABC PE-C7	BD-Biosciences	561349	
PDCA3 ADC	1110	130,000,005	
BDCA2-APC	MALS .	150-090-905	
ULA DR RVA21	BD-Biosciences	561211	
HLA-DR-BV421	BD-Biosciences	562804	
CD45-V500	BD-Biosciences	560777	
Kit T reg (CD4 -FITC/CD25-PE-Cy7/CD127-Alexa Fluor* 647)	BD-Biosciences	560249	
CD3-PE	Beckman-Coulter	A07747	
CD45-V500	BD-Biosciences	560777	
Monitoring Specific T cells:			
Dex-control FITC	IMMUDEX	WB2666-FITC	
Dex-control PE	IMMUDEX	WB2666-PE	
Dex HLA-A*0201 (Melan-A: ELAGIGILTV)- FITC	IMMUDEX	WB2162-FITC	
Dex HLA-A*0201 (Mage3: FLWGPRALV)-PE	IMMUDEX	WB3415-PE	
Dex HLA-A*0201 (gp100 :IMDQVPFSV)-FITC	IMMUDEX	WB2158-FITC	
Dex HLA-A*0201 (Tyrosinase: YMDGTMSQV)-PE	IMMUDEX	WB3245-PE	
Dex HIA-A*0201 (Elu: GILGEVETL)-EITC	IMMUDEX	WB2161-EITC	
Dev MIA-A*0201 (BMIE-1: GICTIVAMI) DE	IMMUDEX	W82130-PE	
CDASDA-ADC-U7	RD-Biosciences	560674	
CCP3.BarCPO/5.5	BD-Biosciences	560074	
CCR/-PercPcys-s	BD-Biosciences	501144	
CD3 PE-Cy7	Beckman-Coulter	737657	
CD8 APC	Beckman-Coulter	IM2469	
Drug product characterization:			
CD11C APC CLONE B-ly6	BD Biosciences	559877	
ANTI HLA DR PE CLONE B8.12.2	Beckman coulter	IM0464	
ANTI HLA A2 FITC CLONE BB7.2	BD Biosciences	551285	
CD303 PE (BDCA-2) CLONE AC144	MILTENYI BIOTEC	130-090-511	
CD56 APC CLONE B159	BD Biosciences	555518	
CD4 APC CLONE SK3	BD Biosciences	345771	
CD304 -APC CLONE ADS-17F6	MILTENYI BIOTEC	130-090-900	
CD123 PE CLONE 9F5	BD Biosciences	340545	
CD40-PE CLONE MAB89	Beckman coulter	IM1936	
CD86 FITC CLONE 2331 FUN-1	BD Biosciences	555657	
CD80 FITC (CLONE MAB104)	Beckman coulter	IM1853	
Tumor characterization			
Day MLA-A*0201 (Malan-A: ELAGIGILTI/), EITC	IMMUDEV.	WB2162-EffC	
Dev HLA-A80201 (an100 JMDD0//DES/2 DE	IN ANALINEY	WB2158.0E	
CDR ResCe CrE E CLONE EK1	INIMODEX BD Bioseir	2410E0	
CD8-PERCP-Cy5.5 CLONE 5K1	Bu-Biosciences	341050	
CD3 PE-Cy7 CLONE UCHT1	Beckman-Coulter	/3/65/	
CD2/4 (PD-L1) APC CLONE MIH1	BU Biosciences	303/41	
CD45-APC-H7 CLONE 2D1	BD Biosciences	5601718	
CD11c-BV421 CLONE B-ly6	BD Biosciences	562561	
CD279 (PD-1)-BV510 CLONE EH12.1	BD Biosciences	563076	
Immunohistochemistry			
CD3 polyclonal (1 :150)	Dako	A452	
CD8 CLONE C8/144B (1 :100)	Dako	M7103	
PD-1 CLONE NAT105 (1:75)	Abcam	ab52587	
PD-L1 CLONE 22C3 (1:30)	Dako	M3653	
HLA-ABC CLONE B9-12-1 (1:7500)	Abcam	ab70328	
CD163 CLONE 10D6 (1 :100)	Abcys	AbC10-C067	
PD-1 expression on melanoma patients' PBMC			
Day HI A-A*0201 (Melan-A: ELACICITITAL ELTC	IMMUDEX	WB2162-507C	
Description operation interest and an an an and an	IN MODEX	WB2152-PF	
CPR PLACE OF S CLONE SKI	INIMODEX	102130'PE	
CD8-Percp-cy5.5 CLONE 5K1	BD-Biosciences	341050	
CD279 (PD-1)-PC7 CLONE EH12.1	Beckman-Coulter	PN-A78885	
CD3 BV421 CLONE UCHT1	BD Biosciences	562426	

Suppl Table 2

HLA typing of PDC lines cell and melanoma patients

	A A		ВВ		DR DR		DQ DQ		Nb of mismatches			
PDC line	A*02:01	A*02:01	B*07:02	B*44:02	DR*01:03	DR*08:01	DQ*04:02	DQ*05:01	HLA-I	HLA-II	Total	
ID Patient												
0101	A*02:01	A*02:01	B*07	B*51	DR*04:05	DR*11	DQ*03	DQ*03	1/4	4/4	5/8	
0201	A*02:01	A*01	B*08	B*44	DR*03	DR*11	DQ*02	DQ*03	2/4	4/4	6/8	
0301	A*02:01	A*25	B*18	B*51	DR*04	DR*08:03	DQ*03	DQ*03	3/4	4/4	7/8	
0401	A*02:01	A*32	B*18	B*57	DR*07	DR*11	DQ*03	DQ*03	3/4	4/4	7/8	
0501	A*02:01	A*02:01	B*37	B*37	DR*08	DR*11	DQ*03	DQ*04	2/4	2/4	4/8	
0103	A*02:01	A*02:01	B*15	B*51	DR*01	DR*04	DQ*03	DQ*05	2/4	2/4	4/8	
0203	A*02:01	A*01	B*35	B*37	DR*01	DR*10	DQ*05	DQ*05	3/4	1/4	4/8	
0403	A*02:01	A*31	B*15	B*49	DR*03	DR*11	DQ*02	DQ*03	3/4	4/4	7/8	
0601	A*02:01	A*24	B*39	B*44	DR*04	DR*11	DQ*03	DQ*03	3/4	4/4	7/8	