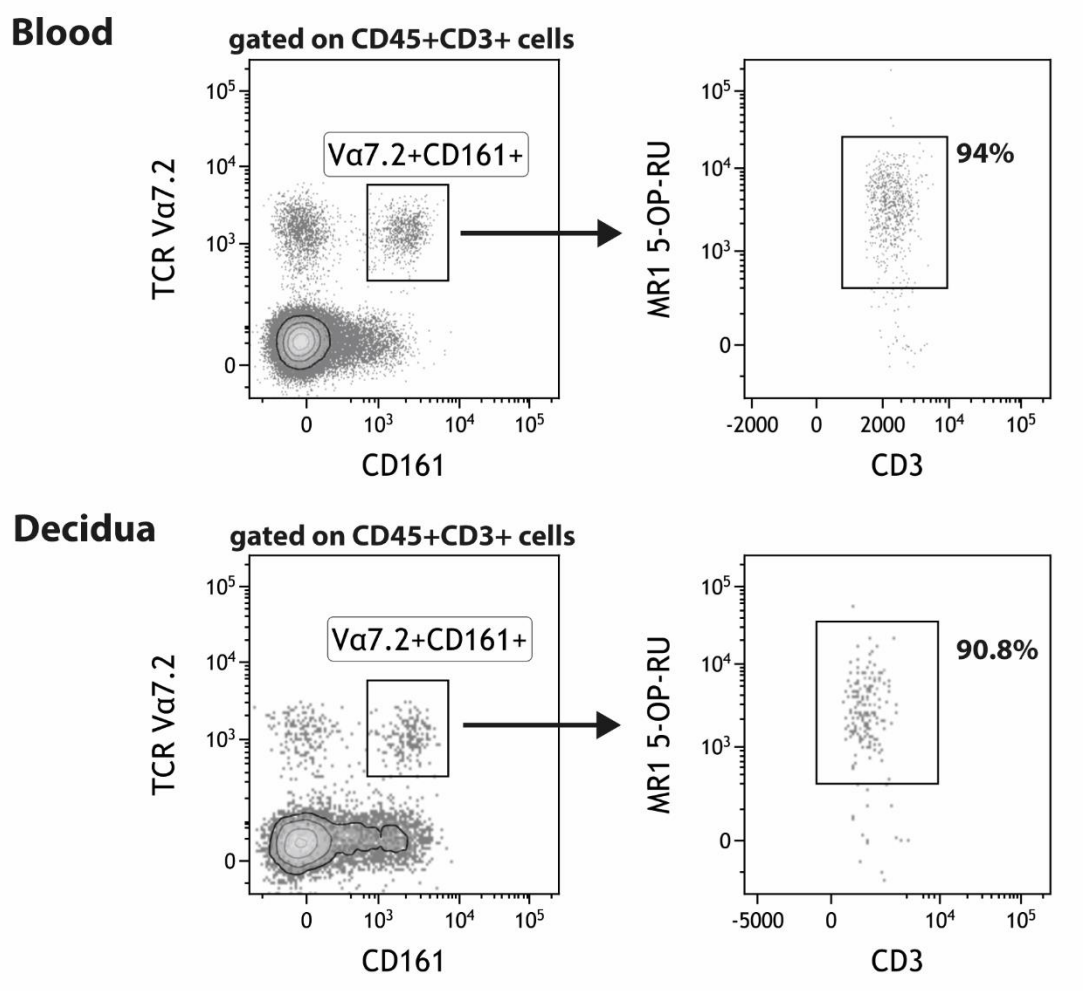
Supplementary Material
Supplementary Figures and Supplementary Table

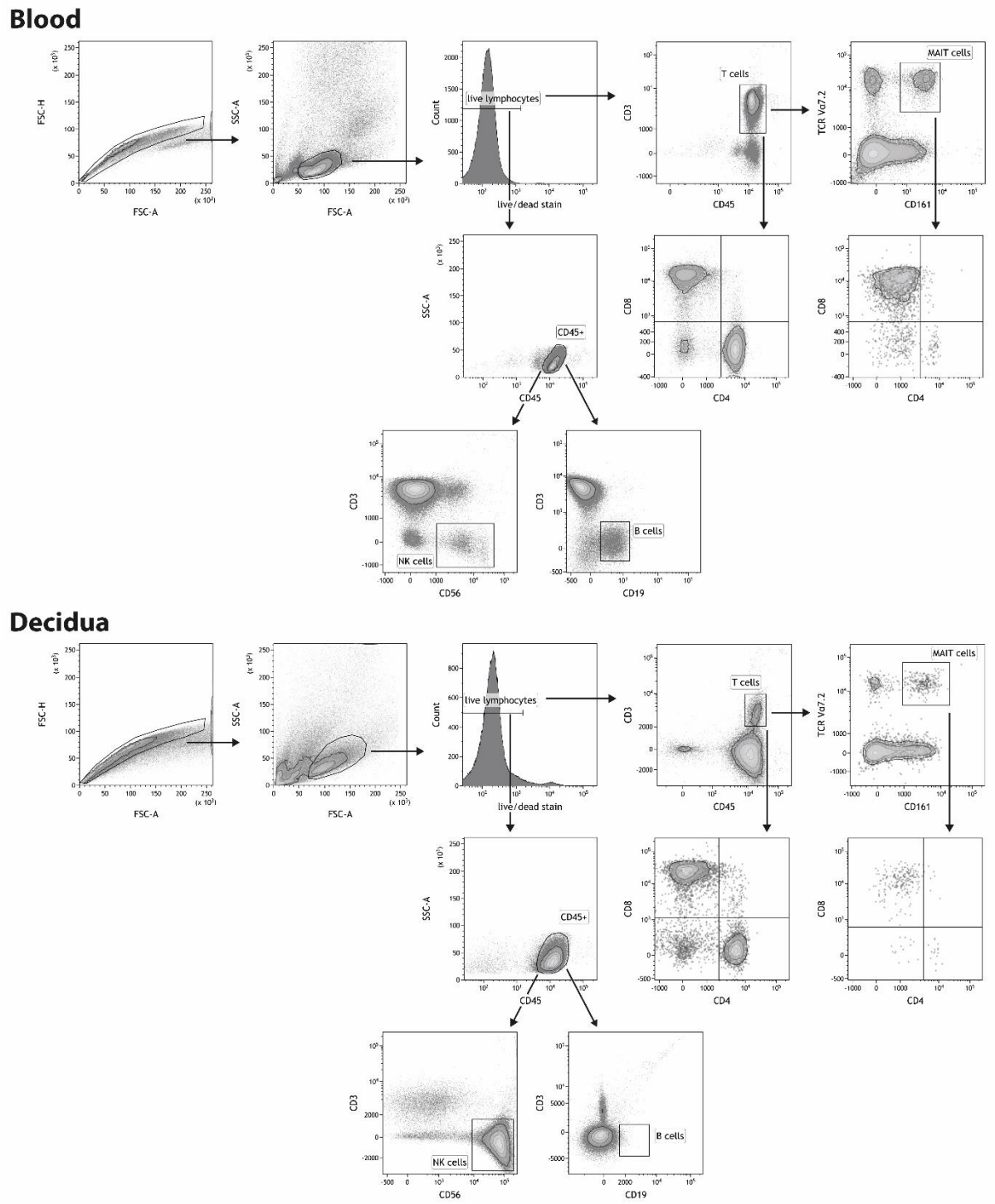
Supplementary Figure S1



Supplementary Figure S1: The majority of Vα7.2$^+$CD161$^+$ MAIT cells are positive for the MR1 tetramer

MR1 tetramer staining of paired blood and decidua samples was carried out in combination with staining for the surface markers CD45, CD3, Vα7.2 and CD161. Vα7.2$^+$CD161$^+$ MAIT cells were gated among CD45$^+$CD3$^+$ cells (left panels), and in the next step (right panels) analyzed for binding of the MR1 5-OP-RU tetramer. The MR1 6-FP tetramer was used as a negative control and to define the gate for MR1 5OP-RU-positive cells (not shown).

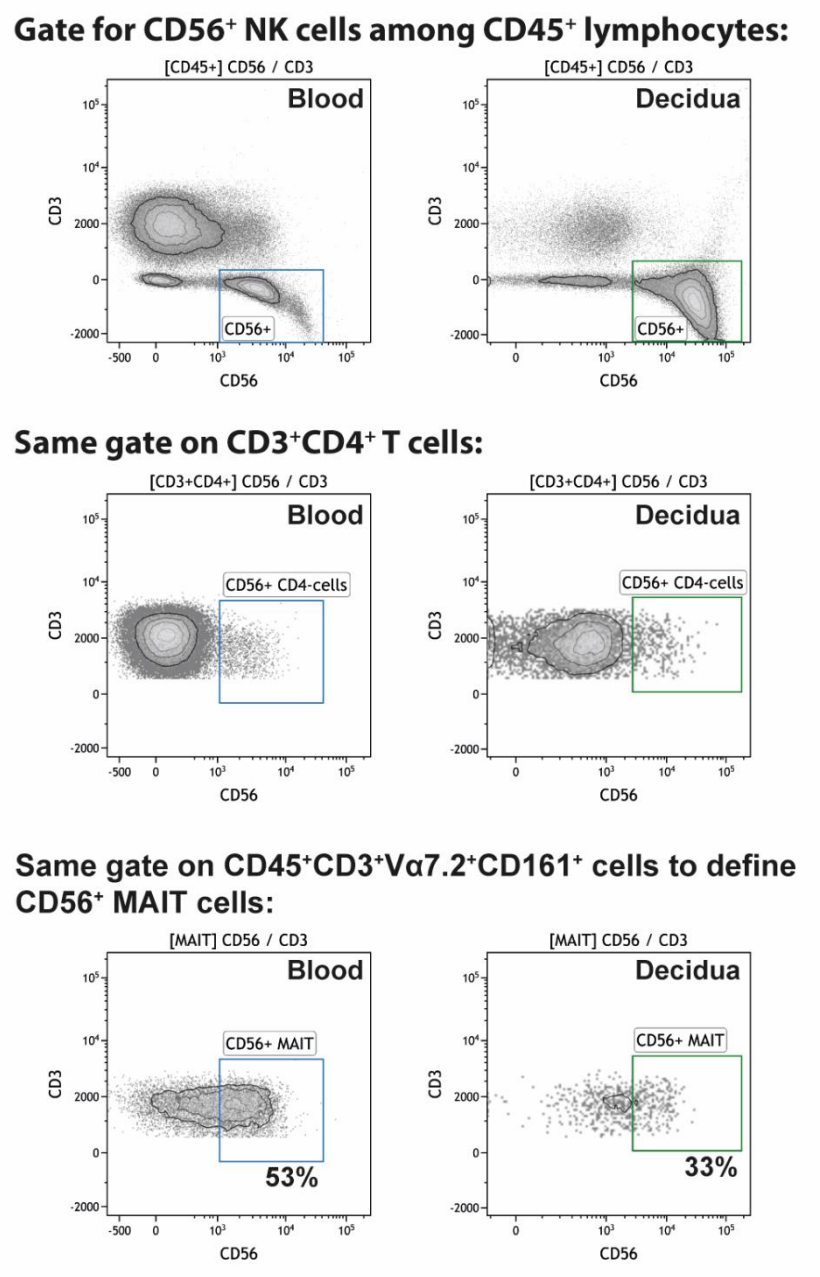
Supplementary Figure S2



Supplementary Figure S2: Gating strategies for lymphocyte subsets in blood and decidua

Peripheral blood mononuclear cells (PBMCs) and decidual cells were isolated from blood and 1st trimester decidual tissue, respectively, from the same donors, and analyzed by flow cytometry. Gating strategy for T cells, MAIT cells, NK cells and B cells, as well as for $CD8^+$, $CD4^+$ and double-negative T cells and MAIT cells are shown for one representative paired sample.

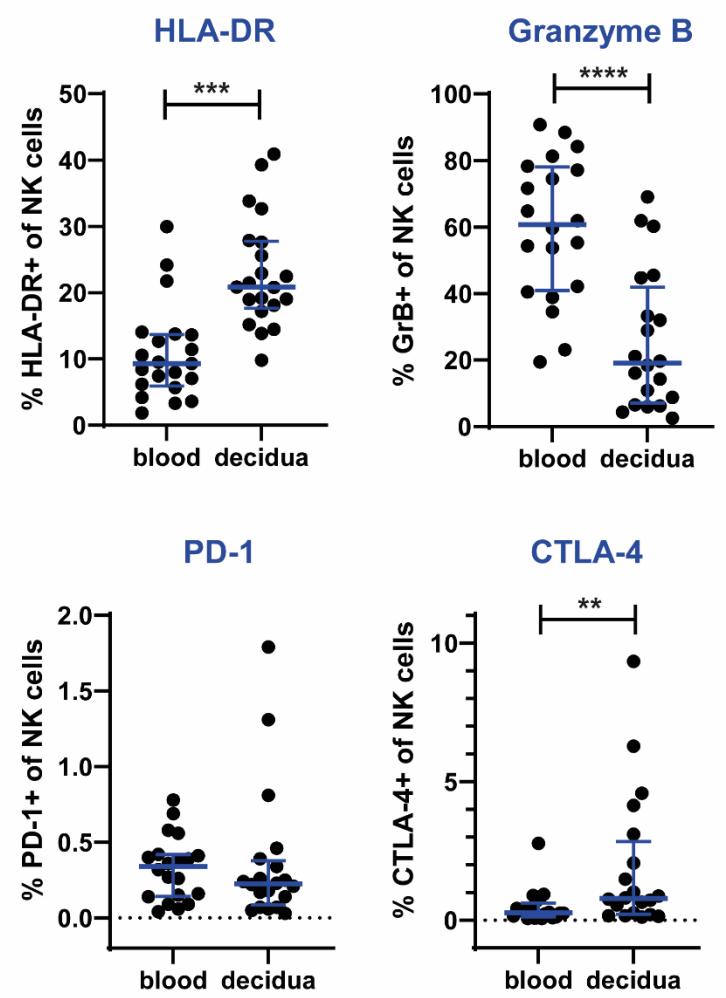
Supplementary Figure S3:



Supplementary Figure S3: Gating strategy for CD56$^+$ MAIT cells in blood and decidua

Peripheral blood mononuclear cells (PBMCs) and decidual cells were isolated from blood and 1st trimester decidual tissue, respectively, from the same donors, and analyzed by flow cytometry. CD56$^+$ lymphocytes (NK cells, upper panels) and CD4$^+$ T cells (middle panels, chosen because they are mostly CD56$^-$) were used to set the gate for CD56$^+$ MAIT cells (lower panels). Percentages of CD56$^+$ MAIT cells are presented in **Figure 2C**.

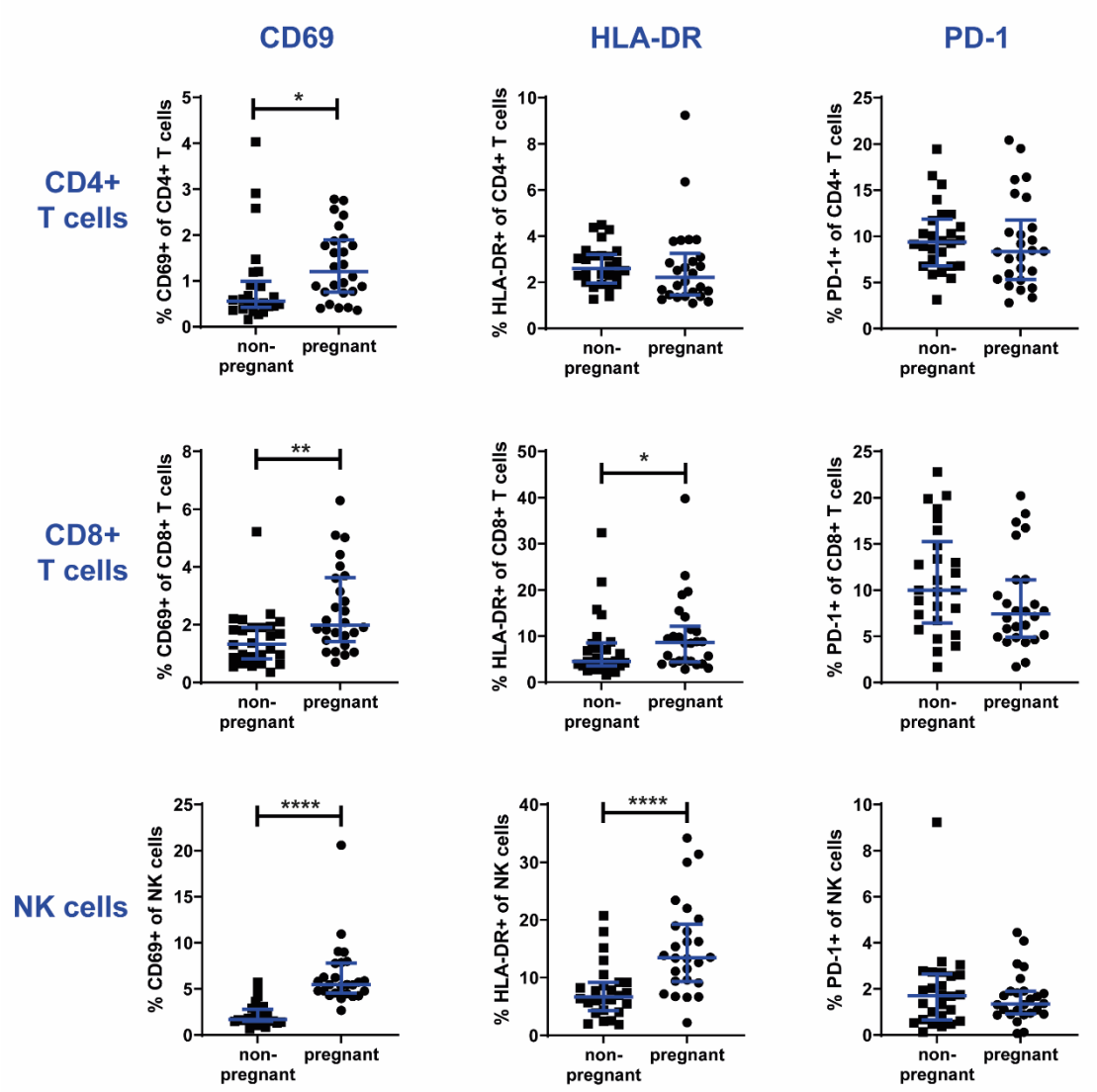
Supplementary Figure S4



Supplementary Figure S4: Expression of phenotypic markers by circulating and decidual NK cells

Peripheral blood mononuclear cells (PBMCs) and decidual cells were isolated from blood and decidual tissue, respectively, from the same donors, and analyzed by flow cytometry for expression of HLA-DR, granzyme B, PD-1 and CTLA-4 by NK cells. HLA-DR and PD-1 were analyzed on the cell surface, whereas granzyme B and CTLA-4 were stained intracellularly. Bars show median and interquartile range (IQR) from 20-21 donors. Statistical comparisons were performed with Wilcoxon signed-rank test. ** $p<0.01$, ***$p<0.001$, **** $p<0.0001$.

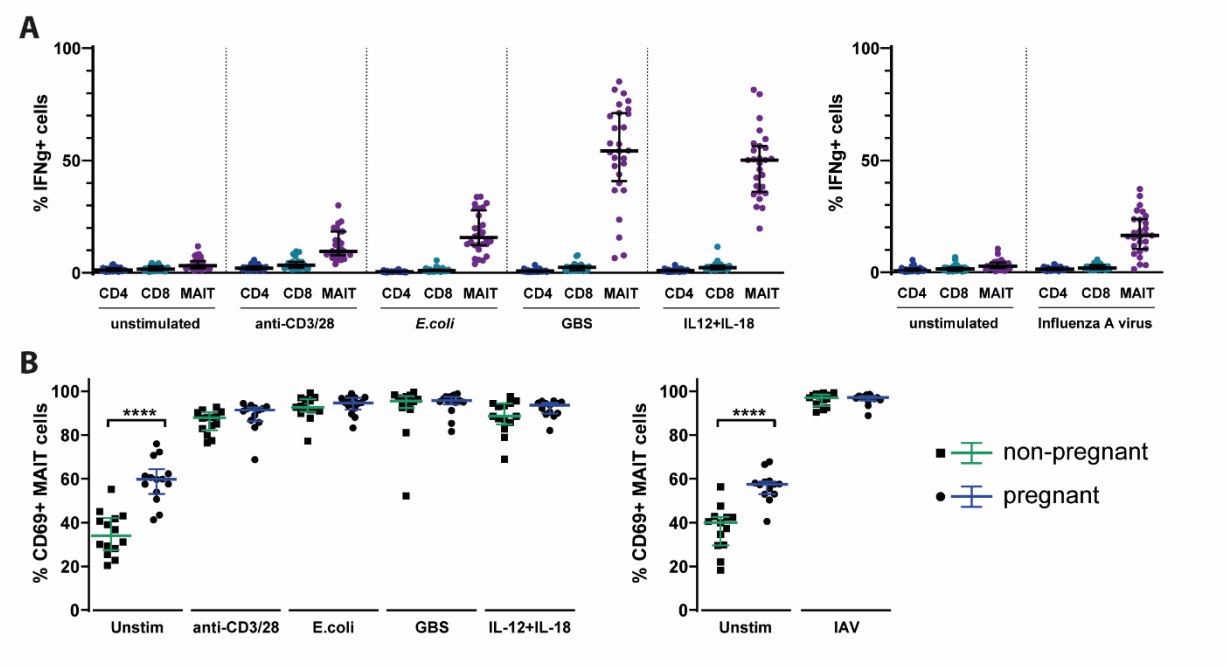
Supplementary Figure S5



Supplementary Figure S5: Expression of phenotypic markers by circulating CD4$^+$ and CD8$^+$ T cells and NK cells in the 3rd trimester of pregnancy

Peripheral blood mononuclear cells (PBMCs) from women pregnant in the 3rd trimester (n=26) and from non-pregnant, age-matched controls (n=26) were analyzed by flow cytometry for the expression of CD69, HLA-DR and PD-1 by CD4$^+$ and CD8$^+$ T cells, as well as by NK cells. Bars show median and IQR and statistical comparisons were performed using Mann-Whitney test. *p<0.05, **p<0.01, **** p<0.0001.

Supplementary Figure S6



Supplementary Figure S6: MAIT cells exhibit a stronger functional response to microbial and inflammatory stimuli than CD4⁺ and CD8⁺ T cells in the same cultures, and a high proportion of stimulated MAIT cells expresses the activation marker CD69

Peripheral blood mononuclear cells (PBMCs) isolated from the blood of 3rd trimester pregnant women and from non-pregnant, age-matched controls were stimulated overnight with plate-bound anti-CD3 and anti-CD28 antibodies, fixed *E. coli*, fixed group B streptococci (GBS), a combination of IL-12 and IL-18 (left panels), and with influenza A virus (IAV, right panels) or left unstimulated (Unstim). IAV stimulations were carried out in a separate plate, together with a separate unstimulated control. **(A)** shows the expression of IFNγ by CD4⁺ and CD8⁺ T cells and MAIT cells in unstimulated and stimulated samples. For this graph, the data from the pregnant and non-pregnant women are combined and presented as a total; n=27-28. Bars show median and IQR. **(B)** shows the expression of CD69 upon stimulation, for non-pregnant and pregnant women. The data for expression of IFNγ, granzyme B, PD-1 and CTLA-4 in the same experiments are shown in **Figure 5**. N=13-14 for both groups. Bars show median and IQR from 20-21 donors. Statistical comparisons in (B) were performed with Mann-Whitney test. **** p<0.0001.

Supplementary Table S1. Flow cytometry antibodies and panels used in this study

x indicates that the antibody was used for surface staining on intact cells. **xx** indicates that this marker was stained intracellularly, after staining with antibodies for surface markers (x), fixation and permeabilization of the cells. PBMC, peripheral blood mononuclear cells.

					1st trimester samples; PBMCs and decidua				3rd trimester PBMCs	
Marker/Antibody	**Clone**	**Host species and isotype**	**Fluorochrome**	**Manufacturer**	**Surface markers**	**Surface and intracellular markers**	**B cell panel**	**Tetramer staining**	**Phenotyping of freshly isolated PBMCs**	**Stimulations of PBMCs**
					Figure 1-3, S2, S3, S4	**Figure 3, S4**	**Figure 1, S2**	**Figure S1**	**Figure 4, S5**	**Figure 5, S6**
Aqua LIVE/DEAD stain	NA			Molecular Probes	x	x	x	x	x	x
CD45	2D1	mouse IgG1	APC-H7	BD	x	x	x	x		
CD3	UCHT1	mouse IgG1	Alexa Fluor 700	BD	x	x	x	x	x	x
CD4	RPA-T4	mouse IgG1	PerCP-Cy5.5	BD	x	x		x	x	x
CD8	SK1	mouse IgG1	BV605	BD	x	x		x		
CD8	SK1	mouse IgG1	APC-H7	BD					x	x
CD56	NCAM16.2	mouse IgG2b	BV711	BD	x	x	x	x	x	x
TCR Vα7.2	3C10	mouse IgG1	PE	Biolegend	x	x		x	x	x
CD161	DX12	mouse IgG1	APC	BD	x	x		x	x	x
isotype control	27-35	mouse IgG2b	FITC	BD	x					
HLA-DR	L243	mouse IgG2a	FITC	BD	x				x	
isotype control	MOPC-21	mouse IgG1	PE-Cy7	BD	x					
PD-1	EH12.1	mouse IgG1	PE-Cy7	BD	x					
isotype control	G155-178	mouse IgG2a	BV421	BD		**xx**				
CTLA-4	BNI3	mouse IgG2a	BV421	BD		**xx**				
isotype control	MOPC-21	mouse IgG1	FITC	BD		**xx**				
Granzyme B	GB11	mouse IgG1	FITC	BD		**xx**				**xx**
isotype control	X40	mouse IgG1	BV605	BD					x	
PD-1	EH12.1	mouse IgG1	BV605	BD					x	x
CD19	SJ25C1	mouse IgG1	PerCP	BD			x			
MR1/6-FP	Tetramer		PE	NIH Tetramer Core Facility				x		
MR1/5-OP-RU	Tetramer		PE	NIH Tetramer Core Facility				x		
IFNγ	4S.B3	mouse IgG1	BV421	BD						**xx**
CD69	FN50	mouse IgG1	BV786	BD						x
CTLA-4	BNI3	mouse IgG2a	PE-Cy7	Biolegend						**xx**