Supporting Information

Table S1: List of antibodies used in the immunophenotyping panel.

Antibody	Fluorescence	Clone	Supplier	Volume used per tube*
CD3	FITC	SK7	BD Biosciences	1.5uL
CD8	PE-Cy7	SK1	Biolegend	0.5uL
CD16	APC-FireTM 750	3G8	Biolegend	2.0uL
CD45	PerCP	2D1	BD Biosciences	5.0uL
CD56	BV421TM	HCD56	Biolegend	1.5uL
CD57	BV510TM	QA17A04	Biolegend	1.5uL
PD-1/CD279	PE	A17188B	Biolegend	2.5uL

APC, allophycocyanin; BV, brilliant violet; FITC, fluorescein isothiocyanate; PE, phycoerythrin; PerCP, peridinin chlorophyll protein complex; PD-1, Programmed Cell Death Protein.
* 100 uL of sample

Table S2: Characterization of major circulating and peritoneal lymphoid subsets in the control group and in women with endometriosis.

Table S2	Peripheral Blood			Peritoneal Fluid		
Parameter	**Control (n=15)**	**EM (n=47)**	**p-value**	**Control (n=15)**	**EM (n=46)**	**p-value**
Percentage Values (%)						
Lymphocytes (% of CD45^{+} Leucocytes)	26.93 [19.84, 39.77]	27.65 [20.41, 33.12]	0.669	51.17 [37.26, 72.00]	47.93 [30.60, 59.06]	0.480
T cells (% of CD45^{Hi} SSC^{low} Lymphocytes)	71.66 [61.37, 78.41]	75.03 [70.97, 79.65]	0.206	84.64 [78.99, 90.90]	82.86 [75.36, 88.86]	0.456
CD16^{+} T cells (% of total T cells)	10.70 [8.63, 18.46]	10.52 [6.10, 19.33]	0.576	41.30 [27.46, 50.20]	32.98 [25.19, 45.84]	0.303
CD57^{+} T cells (% of total T cells)	8.91 [5.58, 14.71]	9.65 [5.50, 16.51]	0.863	14.93 [10.60, 17.50]	16.29 [11.72, 20.44]	0.398
PD-1^{+} T cells (% of total T cells)	29.38 [26.57, 38.78]	25.40 [20.92, 33.34]	0.101	73.44 [63.04, 82.13]	69.65 [58.10, 75.73]	0.121
CD8 T cells (% of total T cells)	32.64 [28.36, 38.29]	31.80 [27.01, 36.87]	0.588	65.83 [60.66, 72.88]	61.68 [56.68, 66.97]	0.265
CD16^{+} CD8 T cells (%)	42.18 [25.42, 55.95]	32.67 [4.26, 48.70]	0.421	27.59 [18.94, 31.98}	29.73 [25.17, 38.35]	0.113
CD56^{+} CD8 T cells (%)	24.74 [20.08, 29.06]	19.92 [13.69, 30.20]	0.500	57.26 [42.92, 63.88]	50.91 [43.34, 69.48]	0.993
CD57^{+} CD8 T cells (%)	18.81 [14.00; 37.43]	21.73 [13.23; 31.98]	0.644	18.77 [13.72; 23.89]	20.43 [14.87; 26.14]	0.498
PD-1^{+} CD8 T cells (%)	38.66 [27.06, 61.86]	31.21 [23.16, 49.00]	0.207	75.95 [61.76, 82.48]	70.14]62.22, 77.38]	0.234
CD4&DN T cells (% of total T cells)	67.36 [61.72, 71.64]	68.20 [63.13, 73.00]	0.588	34.17 [27.12, 39.34]	38.32 [33.04, 43.32]	0.265
CD16^{+} CD4&DN T cells (%)	2.17 [0.63, 3.42]	1.76 [1.21, 2.67]	0.974	19.91 [14.87, 24.80]	18.71 [12.71, 24.56]	0.821
CD56^{+} CD4&DN T cells (%)	3.34 [2.28, 6.50]	4.44 [2.89, 6.82]	0.538	4.84 [3.13, 7.01]	3.65 [2.50, 5.19]	0.096
CD57^{+} CD4&DN T cells (%)	3.21 [2.56; 5.45]	3.42 [2.25; 7.03]	0.942	10.38 [5.91; 15.41]	8.85 [6.58; 13.60]	0.586
PD-1^{+} CD4&DN T cells (%)	24.85 [20.76, 31.07]	22.30 [17.35, 26.25]	0.133	68.85 [62.06, 80.33]	68.54 [58.27, 77.44]	0.645

$CD56^+$ T cells (% of total T cells)	11.87 [7.48, 14.49]	9.26 [7.30, 15.12]	0.669	14.00 [11.96, 14.92]	15.23 [12.46, 17.14]	0.125
$CD57^+$ $CD56^+$ T cells (%)	55.63 [28.24, 71.82]	55.93 [31.57, 68.60]	0.961	19.53 [13.93, 28.09]	21.54 [12.62, 28.69]	0.604
PD-1^+ $CD56^+$ T cells (%)	36.26 [31.00, 51.88]	32.65 [24.17, 45.30]	0.190	63.79 [50.28, 74.44]	57.69 [48.77, 68.44]	0.336
NK cells (% of $CD45^{Hi}SSC^{low}$ lymphocytes)	16.26 [11.81, 23.45]	11.75 [9.29, 17.77]	0.081	13.90 [5.76, 19.94]	13.17 [7.54, 21.15]	0.731
$CD8^+$ NK cells (% of total NK cells)	48.48 [39.22, 62.10]	50.75 [43.42, 60.33]	0.645	24.64 [19.88, 31.76]	27.96 [19.15, 36.75]	0.498
$CD57^+$ NK cells (% of total NK cells)	49.92 [35.66, 61.27]	43.74 [30.89, 62.50]	0.618	16.26 [8.96, 29.71]	22.16 [11.03, 32.82]	0.303
PD-1^+ NK cells (% of total NK cells)	0.48 [0.21, 1.49]	0.73 [0.28, 1.35]	0.382	1.36 [0.83, 2.47]	1.46 [0.99, 3.24]	0.738
$CD56^+$ $CD16^+$ NK cells (% of total NK cells) *	-	-	-	14.00 [10.46, 17.06]	15.63 [9.76, 19.57]	0.530
$CD57^+$ $CD56^+$ $CD16^+$ NK cells (%)	-	-	-	13.40 [3.52; 25.55]	14.92 [7.65; 27.00]	0.609
PD-1^+ $CD56^+$ $CD16^+$ NK cells (%)	-	-	-	1.65 [0.00; 2.34]	3.00 [1.36; 6.67]	**0.021**
$CD56^+$ $CD16^{Hi}$ NK cells (% of total NK cells)	95.15 [93.98, 96.78]	93.78 [91.17, 95.73]	0.073	44.92 [28.89, 50.26]	45.73 [30.93, 60.93]	0.303
$CD8^+$ $CD56^+$ $CD16^{Hi}$ NK cells (%)	50.47 [40.25, 63.80]	52.96 [44.96, 60.95]	0.681	35.48 [23.14, 40.01]	42.34 [32.97, 51.87]	**0.029**
$CD57^+$ $CD56^+$ $CD16^{Hi}$ NK cells (%)	51.01 [36.60; 64.07]	43.90 [31.57; 63.73]	0.618	28.00 [12.24; 40.54]	31.27 [19.11; 50.64]	0.212
PD-1^+ $CD56^+$ $CD16^{Hi}$ NK cells (%)	0.35 [0.20, 1.21]	0.57 [0.30, 1.16]	0.377	0.75 [0.13, 2.82]	0.87 [0.39, 2.34]	0.847
$CD56^{Hi}$ $CD16^-$ NK cells/NK cells (%)	4.46 [3.21, 5.31]	5.75 [4.51, 8.77]	**0.019**	49.02 [41.78, 65.77]	45.84 [33.36, 64.13]	0.353
$CD8^+$ $CD56^{Hi}$ $CD16^-$ NK cells (%)	41.16 [33.18, 51.06]	44.17 [30.33, 51.82]	0.834	17.21 [12.41, 20.52]	18.11 [13.01, 22.07]	0.621
$CD57^+$ $CD56^{Hi}$ $CD16^-$ NK cells (%)	15.58 [6.11, 40.00]	26.12 [6.88, 47.98]	0.711	15.24 [4.82, 21.43]	16.53 [7.58, 23.72]	0.967
PD-1^+ $CD56^{Hi}$ $CD16^-$ NK cells (%)	0.69 [0.00, 1.50]	0.85 [0.10, 1.97]	0.672	0.92 [0.64, 1.75]	1.51 [0.88, 2.95]	0.170

Absolute Counts						
Lymphocytes (cells/μL)	1423.88 [1200.70, 1776.19]	1628.98 [1329.38, 1889.52]	0.218	269.02 [85.94, 452.96]	296.47 [158.66, 491.89]	0.622
T cells (cells/μL)	1019.70 [907.42, 1143.81]	1188.55 [971.40, 1405.38]	0.054	187.39 [77.62, 378.67]	240.91 [145.01, 420.32]	0.530
CD8 T cells (cells/μL)	315.15 [212.10, 411.26]	367.55 [278.81, 502.11]	0.218	143.05 [49.67, 249.72]	166.84 [96.83, 238.26]	0.633
$CD16^+$ CD8 T cells (cells/μL)	91.67 [42.80, 182.26]	111.35 [52.24, 177.25]	0.974	40.55 [14.15, 66.25]	49.55 [22.95, 86.18]	0.295
$CD56^+$ CD8 T cells (cells/μL)	89.60 [38.36, 109.09]	80.14 [50.73, 126.57]	0.554	59.07 [27.67, 124.00]	69.64 [38.05, 138.13]	0.598
CD4&DN T cells (cells/μL)	693.61 [576.07, 770.44]	795.36 [675.77, 962.20]	0.100	58.19 [35.17, 129.39]	86.04 [48.19, 158.13]	0.519
$CD16^+$ CD4&DN T cells (cells/μL)	10.87 [3.67, 21.67]	14.79 [8.28, 25.64]	0.308	17.18 [6.59, 26.23]	12.65 [6.48, 29.66]	0.821
$CD56^+$ CD4&DN T cells (cells/μL)	23.12 [15.20, 41.59]	35.40 [21.94, 59.62]	0.118	4.76 [1.28, 8.37]	2.90 [1.55, 6.25]	0.541
$CD16^+$ T cells (cells/μL)	109.06 [56.49, 191.57]	119.67 [72.44, 194.52]	0.857	68.18 [29.24, 139.54]	68.48 [41.28, 140.19]	0.913
$CD56^+$ T cells (cells/μL)	107.71 [59.70, 141.04]	116.33 [88.02, 172.15]	0.366	26.83 [12.46, 50.36]	34.34 [22.77, 63.73]	0.353
Total NK cells (cells/μL)	183.10 [131.89, 316.08]	197.33 [106.70, 317.58]	0.780	41.40 [4.95, 76.97]	30.05 [23.07, 69.59]	0.887
$CD8^+$ NK cells (cells/μL)	100.76 [48.44, 153.22]	102.41 [59.77, 156.98]	0.857	11.51 [1.22, 19.629	9.16 [4.60, 15.39]	0.913
$CD56^+$ $CD16^+$ NK cells (cells/μL) *	-	-	-	4.31 [0.84, 9.07]	5.98 [2.13, 10.75]	0.621
$CD56^+$ $CD16^{Hi}$ NK cells (cells/μL)	177.28 [123.85, 305.84]	178.45 [97.98, 282.90]	0.705	16.44 [1.95, 27.81]	15.02 [6.22, 24.65]	0.731
$CD8^+$ $CD56^+$ $CD16^{Hi}$ NK cells (cells/μL)	97.37 [44.71, 154.35]	101.18 [56.08, 149.48]	0.908	4.97 [0.65, 8.22]	6.41 [2.90, 10.09]	0.280
$CD56^{Hi}$ $CD16^-$ NK cells (cells/μL)	8.12 [5.58, 10.19]	11.24 [7.24, 17.57]	**0.047**	26.48 [2.23, 49.05]	17.24 [5.55, 35.55]	0.847
$CD8^+$ $CD56^{Hi}$ $CD16^-$ NK cells (cells/μL)	3.17 [1.93, 4.86]	4.13 [2.42, 7.73]	0.147	4.88 [0.33, 5.50]	2.59 [0.98, 6.07]	0.913

$CD56^{+}CD16^{Hi}$ NK cells / $CD56^{Hi}$ $CD16^{-}$ NK cells ratio	21.08 [17.79, 30.17]	15.98 [10.47, 21.20]	0.019	0.88 [0.41, 1.15]	1.05 [0.49, 1.93]	0.311
T cells / NK cells ratio	4.27 [2.57, 6.91]	6.40 [3.97, 8.21]	0.100	6.01 [3.96, 15.69]	6. 35 [3.58, 12.09]	0.657

Results for all the subsets characterized in this study, including both absolute counts and percentages for each population within the parent population. All results are presented as Median [IQC] values. P-values were calculated by means of Wilcoxon rank-sum test.

* $CD56^{+}CD16^{+}$ NK cells were only visible in the Peritoneal Fluid samples.

DN – Double negative; EM – Endometriosis; n – number of observations; IQC – interquartile range.

Table S3: Characterization of circulating and peritoneal lymphoid subsets in control group and in EM women with different clinical manifestations

Dysmenorrhea				
PF	**Control (n=15)**	**EM Dysmenorrhea (n=44)**	**EM No Dysmenorrhea (n=3)**	**p-value**
PD-1$^+$ NK cells	1,36 [0,83; 2,47]	1,60 [0,99; 3,45]	1,02 [0,38; 2,08]	0,447
PD-1$^+$ CD56$^+$ CD16$^+$ NK cells	1,65 [0,00; 2,34]	3,06 [1,36; 6,67]	2,78 [0,78; 10,00]	0,069
PD-1$^+$ CD56$^+$ CD16^{Hi} NK cells	0,75 [0,13; 2,82]	0,84 [0,39; 3,25]	1,28 [0,26; 1,69]	0,938
PD-1$^+$ CD56^{Hi} CD16$^+$ NK cells	0,92 [0,64; 1,75]	1,64 [0,96; 3,14]	0,77 [0,45; 1,50]	0,149
PF	**Control (n=15)**	**EM Severe Dysmenorrhea (>=7) (n=35)**	**EM None/Mild/Mod Dysmenorrhea (<7) (n=11)**	**p-value**
PD-1$^+$ NK cells	1,36 [0,83; 2,47]	1,31 [0,92; 2,66]	2,08 [1,02; 5,10]	0,530
PD-1$^+$ CD56$^+$ CD16$^+$ NK cells	1,65 [0,00; 2,34]	2,04 [1,36; 5,41]	5,66 [1,11; 10,00]	**0,028**
PD-1$^+$ CD56$^+$ CD16^{Hi} NK cells	0,75 [0,13; 2,82]	0,65 [0,35; 1,90]	1,85 [0,75; 4,26]	0,228
PD-1$^+$ CD56^{Hi} CD16$^+$ NK cells	0,92 [0,64; 1,75]	1,51 [0,88; 2,82]	1,50 [0,77; 4,14]	0,352
Dyspareunia				
PF	**Control (n=15)**	**EM Dyspareunia (n=27)**	**EM No Dyspareunia (n=19)**	**p-value**
PD-1$^+$ NK cells	1,36 [0,83; 2,47]	2,08 [1,10; 3,67]	1,02 [0,64; 2,32]	**0,035**
PD-1$^+$ CD56$^+$ CD16$^+$ NK cells	1,65 [0,00; 2,34]	3,48 [1,38; 6,95]	1,87 [0,78; 5,13]	**0,029**
PD-1$^+$ CD56$^+$ CD16^{Hi} NK cells	0,75 [0,13; 2,82]	1,34 [0,53; 3,70]	0,45 [0,13; 1,63]	0,102
PD-1$^+$ CD56^{Hi} CD16$^+$ NK cells	0,92 [0,64; 1,75]	1,65 [1,10; 2,95]	1,09 [0,63; 3,18]	0,215
PB	**Control (n=14)**	**EM Dyspareunia (n=28)**	**EM No Dyspareunia (n=19)**	**p-value**
PD-1$^+$ T cells	29,38 [26,57; 38,78]	28,81 [24,56; 33,74]	21,60 [19,60; 28,82]	**0,021**
PD-1$^+$ CD8$^+$ T cells	38,66 [27,06; 61,86]	38,70 [23,84; 50,32]	25,92 [21,48; 42,42]	0,162
PD-1$^+$ CD8$^-$ (CD4$^+$&DN) T cells	24,85 [20,76; 31,07]	25,07 [17,34; 29,43]	20,55 [19,51; 22,39]	**0,049**
PD-1$^+$ CD56$^+$ T cells	36,26 [31,00; 51,88]	35,35 [26,28; 48,91]	26,72 [17,38; 40,38]	0,100
PD-1$^+$ NK cells	0,48 [0,21; 1,49]	0,91 [0,60; 1,95]	0,33 [0,22; 0,91]	0,052

PB	Control (n=14)	EM Severe Dyspareunia (n=17)	EM None/Mild/Mod Dyspareunia (n=30)	p-value
PD-1^{+} T cells	29,38 [26,57; 38,78]	31,08 [26,92; 33,87]	24,56 [19,89; 29,68]	**0,038**
PD-1^{+} CD8^{+} T cells	38,66 [27,06; 61,86]	43,62 [35,54; 52,66]	25,59 [22,84; 42,42]	0,061
PD-1^{+} CD8^{-} (CD4^{+}&DN) T cells	24,85 [20,76; 31,07]	24,51 [17,35; 29,13]	20,70 [19,51; 25,60]	0,200
PD-1^{+} CD56^{+} T cells	36,26 [31,00; 51,88]	42,13 [33,63; 49,06]	26,79 [18,29; 40,38]	**0,029**
PD-1^{+} NK cells	0,48 [0,21; 1,49]	1,04 [0,80; 2,18]	0,58 [0,25; 0,92]	0,074
Dyschesia				
PF	**Control (n=15)**	**EM Dyschesia (n=16)**	**EM No Dyschesia (n=31)**	**p-value**
PD-1^{+} T cells	73,44 [63,04; 82,13]	76,34 [65,56; 80,38]	69,29 [56,78; 74,39]	**0,048**
PD-1^{+} CD8^{+} T cells	75,95 [61,76; 82,48]	76,31 [65,07; 83,92]	68,42 [59,05; 74,31]	0,052
PD-1^{+} CD8^{-} (CD4^{+}&DN) T cells	68,85 [62,06; 80,33]	70,99 [63,55; 75,96]	68,09 [55,30; 77,44]	0,854
PD-1^{+} CD56^{+} T cells	63,79 [50,28; 74,44]	62,31 [51,17; 75,67]	55,37 [45,91; 66,56]	0,250
Rectorrhagia				
PF	**Control (n=15)**	**EM Rectorrhagia (n=4)**	**EM No Rectorrhagia (n=43)**	**p-value**
PD-1^{+} NK cells	1,36 [0,83; 2,47]	2,97 [1,67; 5,13]	1,31 [0,92; 2,77]	0,435
PD-1^{+} CD56^{+} CD16^{+} NK cells	1,65 [0,00; 2,34]	1,42 [0,64; 2,53]	3,08 [1,38; 6,78]	**0,021**
PD-1^{+} CD56^{+} CD16^{Hi} NK cells	0,75 [0,13; 2,82]	1,79 [0,74; 3,66]	0,80 [0,39; 2,34]	0,883
PD-1^{+} CD56^{Hi} CD16^{+} NK cells	0,92 [0,64; 1,75]	1,89 [0,48; 2,98]	1,51 [0,88; 2,95]	0,361
Pelvic pain				
PF	**Control (n=15)**	**Pelvic pain (n=19)**	**No pelvic pain (n=28)**	**p-value**
PD-1^{+} NK cells	1,36 [0,83; 2,47]	2,09 [1,02; 3,24]	1,25 [0,74; 3,45]	0,474
PD-1^{+} CD56^{+} CD16^{+} NK cells	1,65 [0,00; 2,34]	2,78 [1,27; 6,67]	3,06 [1,38; 6,78]	0,057
PD-1^{+} CD56^{+} CD16^{Hi} NK cells	0,75 [0,13; 2,82]	0,90 [0,45; 3,25]	0,76 [0,35; 2,34]	0,956
PD-1^{+} CD56^{Hi} CD16^{+} NK cells	0,92 [0,64; 1,75]	1,65 [0,88; 3,36]	1,43 [0,63; 2,47]	0,320

Urinary Tract EM				
PF	**Control (n=15)**	**Urinary Tract EM (n=5)**	**No Urinary Tract EM (n=42)**	**p-value**
PD-1^{+} NK cells	1,36 [0,83; 2,47]	3,89 [3,12; 5,44]	1,31 [0,92; 2,38]	**0,042**
PD-1^{+} CD56^{+} CD16^{+} NK cells	1,65 [0,00; 2,34]	5,13 [1,74; 6,87]	2,86 [1,36; 6,30]	0,056
PD-1^{+} CD56^{+} CD16^{Hi} NK cells	0,75 [0,13; 2,82]	1,93 [0,31; 4,23]	0,87 [0,34; 2,10]	0,957
PD-1^{+} CD56^{Hi} CD16^{+} NK cells	0,92 [0,64; 1,75]	2,34 [0,93; 3,09]	1,47 [0,88; 2,95]	0,373
PB	**Control (n=14)**	**Urinary Tract EM (n=5)**	**No Urinary Tract EM (n=42)**	**p-value**
PD-1^{+} T cells	29,38 [26,57; 38,78]	33,34 [32,27; 39,86]	24,71 [20,20; 31,08]	**0,029**
PD-1^{+} CD8^{+} T cells	38,66 [27,06; 61,86]	51,64 [37,00; 78,26]	28,25 [22,84; 43,91]	**0,037**
PD-1^{+} CD8^{-} (CD4^{+}&DN) T cells	24,85 [20,76; 31,07]	24,51 [16,75; 28,66]	21,52 [18,34; 26,21]	0,318
PD-1^{+} CD56^{+} T cells	36,26 [31,00; 51,88]	48,76 [39,83; 56,96]	28,92 [22,60; 42,13]	**0,036**
PD-1^{+} NK cells	0,48 [0,21; 1,49]	2,18 [0,73; 3,31]	0,70 [0,27; 1,10]	0,177

The table presents results for the comparisons performed for different subsets, according to different clinical manifestations in EM patients. Results are presented as percentages of PD-1^{+} cells within the respective parent population. All results are presented as Median [IQC] values. P-values were calculated by of a Kruskal-Wallis with post-hoc Dunn's test for paired comparison.
DN – Double negative; EM – Endometriosis; n – number of observations; IQC – interquartile range.