Raman Spectroscopy Profiling of Splenic T-Cells in Sepsis and Endotoxemia in Mice

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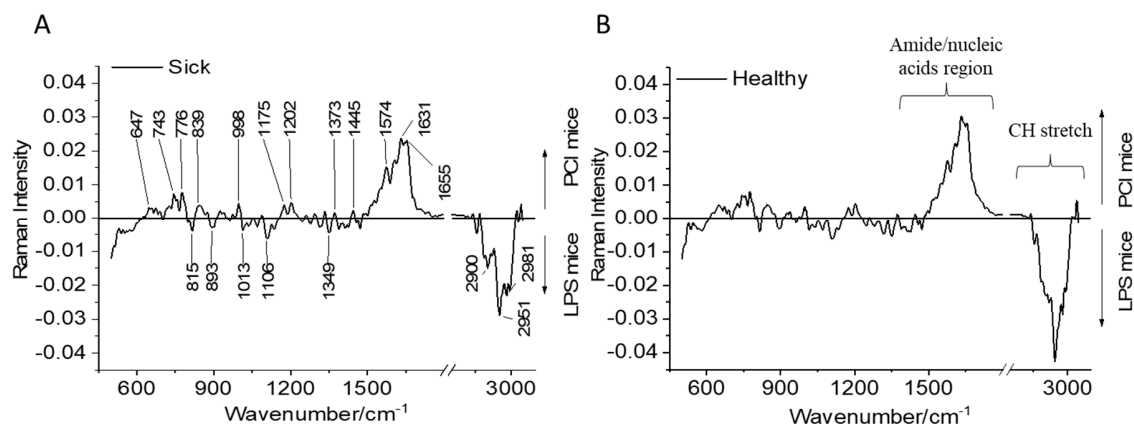
Supplement Figures

Figure S1: Raman difference spectra generated by subtracting C) mean Raman spectra of T-lymphocytes isolated from the spleen of endotoxin mice from septic mice. D) For comparison, the Raman difference spectra were generated by subtracting the mean Raman spectra of T-lymphocytes isolated from the spleen of healthy mice within the endotoxemia model and the PCI models.
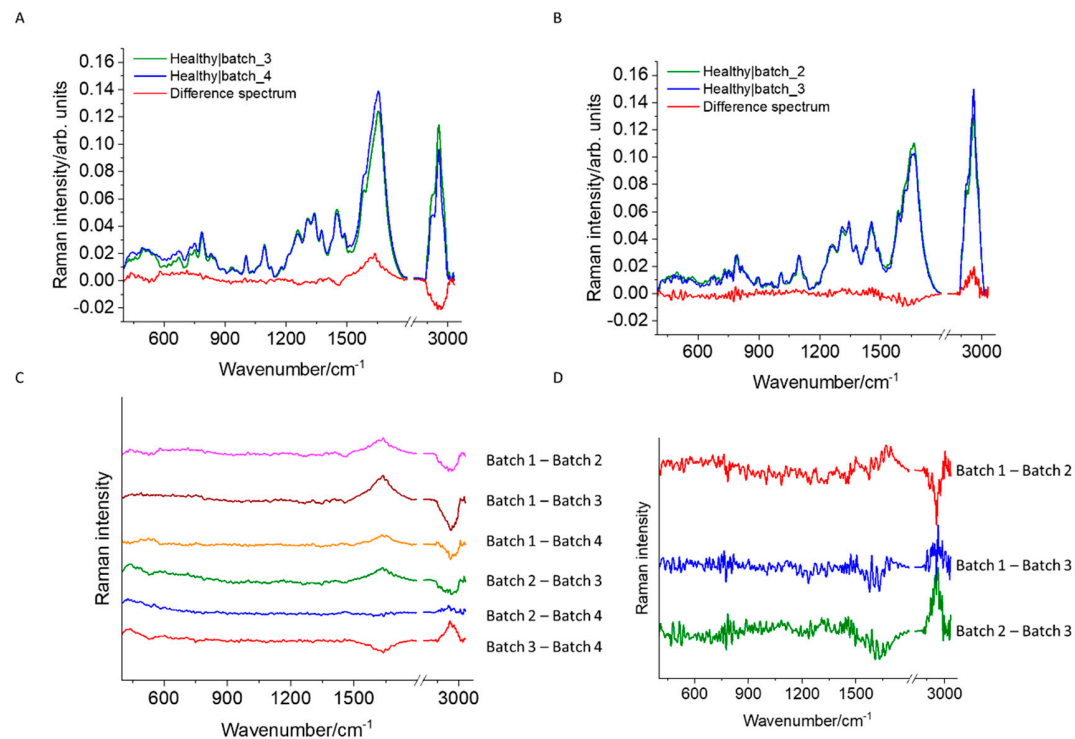
Figure S2: Raman difference spectra generated by subtracting mean Raman spectra of A) control mice from experimental batch 3 and batch 4 of the PCI model, B) control mice from experimental batch 2 and batch 3 of the endotoxemic model. Raman difference spectra for C) PCI model control mice from different experimental batches, D) endotoxemic model control mice from different experimental batches.

Table S1: A) Experimental details of T-cells isolated from the spleen of control mice, septic mice (PCI), and endotoxemic mice (LPS) showing the number of biological replicates, and gender of mice.

Mice with LPS insult			
Biological replicates	**9 to 12 mice/ batch**	**Post-LPS insult time**	**Gender of mice**
Batch 1	Pooled	One day	Females and Males
Batch 2	Pooled	One day	Females and Males

Batch 3	Pooled	One day	Females and Males
Mice with PCI insult			
Biological replicates	**Mice/batch**	**Post PCI insult time**	**Gender of mice**
Batch 1			
PCI_73646 PCI_73647 Sham_73670 Sham_73671	4 mice (2 control and 2 septic)	One day	Females
Batch 2			
PCI_73648 PCI_73666 Sham_73678 Sham_73679	4 mice (2 control and 2 septic)	One day	Females Males Males Males
Batch 3			
PCI_73667 PCI_73668 Sham_73677 Sham_73680	4 mice (2 control and 2 septic)	One day	All males
Batch 4			
PCI_73669 PCI_73681 Sham_73682 Sham_73683	4 mice (2 control and 2 septic)	One day	All males
Batch 5			
PCI_73739 PCI_73740	2 septic mice	One day	All females

Table S2: Batch-wise confusion matrix of Raman models for differentiation of T-lymphocytes from sham and septic/inflammation mice.

CLASSIFICATION MODELS using 10-fold cross-validation	Balanced accuracy*/Cohen's kappa					
	PCI mice model			LPS mice model		
	PCA-LDA	PCA-RF	PCA-SVM	PCA-LDA	PCA-RF	PCA-SVM
BATCH 1	0.85/0.71	0.86/0.72	0.85/0.71	0.60/0.19	0.60/0.19	0.62/0.23
BATCH 2	0.67/0.33	0.76/0.52	0.77/0.53	0.82/0.64	0.83/0.66	0.87/0.74
BATCH 3	0.72/0.43	0.76/0.52	0.81/0.60	0.71/0.42	0.71/0.42	0.72/0.44
BATCH 4	0.84/0.67	0.83/0.66	0.82/0.63	-	-	-
*Balanced accuracy is the mean sensitivity						

Table S3: Confusion matrix obtained from PCA-LDA classification model built to differentiate T-lymphocytes from sham and infected mice from PCI model.

PCA-LDA classification model (batches 2 to 5) for PCI using batch-wise cross-validation								
	Confusion matrix for individual aggregated spectra				Majority vote			
True/Predicted	Sham	Infected	Sens	Spec	Sham	Infected	Sens	Spec
Sham	356	264	0.57	0.75	1	0	1	1
Infected	174	519	0.75	0.57	0	1	1	1
Balanced accuracy* /Cohen's kappa	0.67/0.33				1/1			
*Balanced accuracy is mean sensitivity, Sens: sensitivity, Spec: specificity								

Table S4: Confusion matrix obtained from PCA-LDA prediction of T-lymphocytes from septic mice.

PCA-LDA prediction model (batch 1) for PCI								
	Confusion matrix for individual aggregated spectra				Majority vote			
True/Predicted	Sham	Infected	Sens	Spec	Sham	Infected	Sens	Spec
Sham	134	49	0.73	0.85	1	0	1	1
Infected	23	130	0.85	0.73	0	1	1	1
Balanced accuracy* /Cohen's kappa	0.79/0.57				1/1			
*Balanced accuracy is mean sensitivity, Sens: sensitivity, Spec: specificity								

Table S5: Confusion matrix obtained from PCA-LDA classification model built to differentiate T-lymphocytes from sham and infected mice from LPS model.

PCA-LDA classification model (batches 1 and 3) for LPS using batch-wise cross-validation								
	Confusion matrix for individual aggregated spectra				Majority vote			
True/Predicted	Sham	Infected	Sens	Spec	Sham	Infected	Sens	Spec
Sham	1535	33	0.98	0.04	1	0	1	0
Infected	1398	51	0.04	0.98	1	0	0	1
Balanced accuracy* /Cohen's kappa	0.51/0.02				0.50/0			
*Balanced accuracy is mean sensitivity, Sens: sensitivity, Spec: specificity								

Table S6: Confusion matrix obtained from PCA-LDA prediction of T-lymphocytes from LPS mice.

PCA-LDA prediction model (batch 2) for LPS								
	Confusion matrix for individual aggregated spectra				Majority vote			
True/Predicted	Sham	Infected	Sens	Spec	Sham	Infected	Sens	Spec
Sham	78	595	0.12	0.68	0	1	0	1
Infected	230	497	0.68	0.12	0	1	1	0
Balanced accuracy* /Cohen's kappa	0.4/-0.21				0.50/0			
*Balanced accuracy is mean sensitivity, Sens: sensitivity, Spec: specificity								