Quantitative Analysis of CD4+ and CD8+ T Cells Structures and Morphology Based Classification

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Hypothesis and Specific Aims

**Hypothesis**
The diffraction imaging flow cytometry method allows label-free classification of B and T cell subtypes.

**Aims**
1. Quantitative study of lymphocyte morphology through confocal imaging
2. Acquisition of Cross-Polarized Diffraction Images
3. Cell Classification by SVM with confocal and diffraction image data

Motivation of Research

- To investigate the correlation between diffraction images of lymphocytes and their 3D morphology.
- To develop an innovative label-free method for rapid and accurate assay of leukocytes.

Background and Significance: The Immune System

- First line of defense against a microbial invasion or abnormal cells.
- Adaptive and innate responses
  - Adaptive responses
    - Requires prior exposure to certain pathogens.
    - Body produces antibodies against pathogens.
    - Lifelong protective immunity to reinflection of the same pathogen.
  - Innate responses
    - Does not require prior exposure to particular pathogens.
    - Macrophages engulf pathogens.
    - Does not provide lifelong immunity to reinflection of the same pathogen.

Support Vector Machine Cell Classification

- Model is saved and evaluated with a decision function
- SVM will perform a binary classification of the data based upon the outcome of the decision function, whether 1 or 0 is predicted.
- **Classification Accuracy**

Support Vector Machine Cell Classification

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3D Morphological Feature Parameters data of CD4+ and CD8+ T cells from human spleen sample

**Support Vector Machine Cell Classification**

- 3D morphological feature parameters and the p-Diff feature parameters data is imported into an in-house developed software (SWAM) based upon the SVM algorithm.
- The software will use the SVM algorithm to recognize patterns within the feature parameters of both 3D morphological feature parameters and the p-Diff feature parameters data and perform a binary classification of the different lymphoid subtypes.
- Software will be used to obtain optimized SVM models for classification.

**Summary**

- Research plan
  - Quantitative study of lymphocyte morphology through confocal imaging and 3D reconstruction
  - Acquisition of Cross-Polarized Diffraction Images to investigate lymphocyte classification
  - Apply SVM algorithm for Cell Classification with image data
- **Goal**
  - To develop label-free “fingerprints” for rapid and morphology-based assay of human lymphocytes.
References

• Literature Cited
• 4. Y. Sa et al., “Study of low speed flow cytometry for diffraction imaging with different chamber and nozzle designs,” Cytometry A 83(11), 1027–1033 (2013)