BACHELOR'S THESIS

(Arbeitstitel)

submitted to the

Institute of
Analysis and Scientific Computing
TU Wien

under the supervision of

Assistant Prof. Dr. Andreas Körner

by

Ida Hönigmann
Matriculation number: 12002348

Vienna, June 22, 2024

Acknowledgement

Eidesstattliche Erklärung

Ich erkläre an Eides statt, dass ich die vorliegende Bachelorarbeit selbstständig und ohne fremde Hilfe verfasst, andere als die angegebenen Quellen und Hilfsmittel nicht benutzt bzw. die wörtlich oder sinngemäß entnommenen Stellen als solche kenntlich gemacht habe.

Wien, am 22. Juni 2024

Ida Hönigmann

Contents

1 Introduction

2 T-Cells, Calcium Concentration

Lymphocytes form a key component of the immune system. T cells are a type of lymphocyte and are responsible for responding to viruses, fungi, allergens and tumors. Different subtypes of t cells exist, that fulfill various responsibilities. They are transported throughout the body via the lymphatic system and blood.[KCF18]

Precursor cells are formed in the bone marrow. Once they are transported to the thymus they undergo maturation and selection to become t cells. Each cell forms receptors, called t cell receptors (TCR), that respond to one perticular out of many ($10^6 - 10^9$) possible short pieces of proteins, called peptides. These peptides are attached to the major histocompatibility complex (MHC) present on antigens and antigen presenting cells (APC). Important aspects of the selection are ensuring that the t cells react to foreign peptides, but not to those present on the body's own cells.[AH24]

In positive selection cells in the thymus present peptides on their MHC. If a t cell is unable to bind, it will undergo apoptosis, a type of cell death. T cells which were able to bind recieve survival signals. Negative selection verifies that t cells will not attack the body's own cells. This is done by only selecting t cells which only bind moderatly to the peptides presented, as a strong bond sugessts that these t cells would have a high likelihood of being reactive to own cells.[Hag18] If a t cell passed both the positive and negative selection it is transported to the periphery.

There are multiple types of peripheral t cells. Native t cells respond to new antigens. Cytotoxic t cells kill cells which present peptides on their MHC compatible with the t cells TCR. Helper T cells activate other parts of the immune response. Memory t cells shorten the reaction time when the same antigen is encountered again at a later point in time. Suppressor t cells moderate the immune response.[Gan97]

2.1 Components of a T-Cell

2.2 Activation

Activation is necessary for t cells to divide and perform their functions.[Gan97]

When a native t cell encounters an APC that is compatible, a bond is formed between the TCR on the t cell and the peptide-MHC complex on the APC. This recognition can be triggered by less than ten molecules of foreign substance and is therefore described as near perfect. Sufficiently long contact is nececcary between the APC and the t cell in order for the t cell to activate. The role of contact time in t cell activation is modelled by Morgan et.al.[ML23].

The presence of co-stimulatory molecules is needed for proper activation. The bond between the co-stimulatory molecules on the t cell and APC plays a role in signaling.

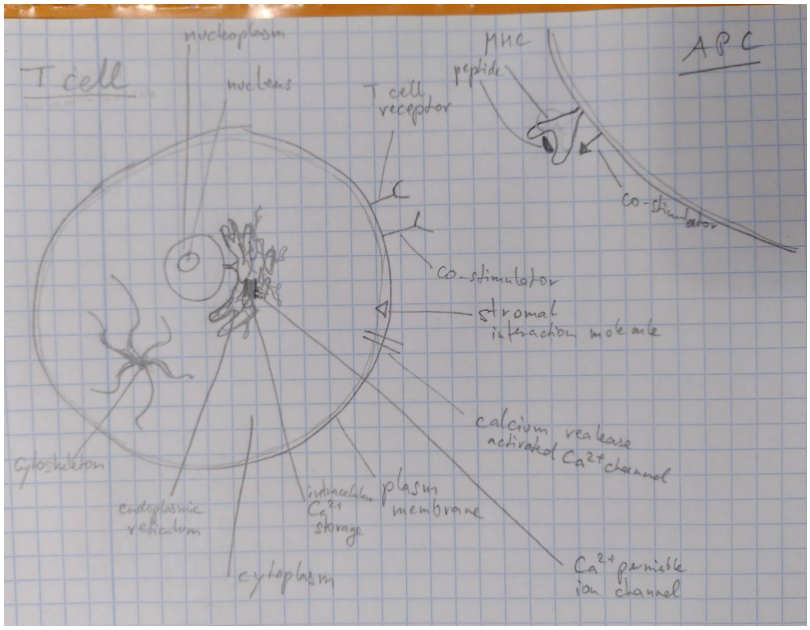

Figure 2.1: Skematic view of a t cell and antigen presenting cell, with all relevant components.

Ca^{2+}signals play a vital part in t cell activation.

An increase of Ca^{2+}in t cells during activation is caused by the stimulation of Ca^{2+}permiable ion channel receptors on the ER membrane. $Ca^{2+}$is released from the ER into the cytoplasm. Additionally this decrease in $Ca^{2+}$is sensed by STIM, which leads to an influx of Ca^{2+}through plasma membrane CRAC channels.[SKJ09]

As the intracellular Ca^{2+}concentration is dependent on the interaction between Ca^{2+}sources and sinks, a variety of different forms in Ca^{2+}concentration have been observed. Examples are infrequent spikes, sustained oscilations and plateaus.[Lew01]

Intercellular Ca^{2+}increase together with other signals lead to a redistribution of receptors, signaling molecules and organelles.[JRB14]

3 Data

calcium concentration shows activatedness of t cells (reference chapter t cells), relativly easy to measure

3.1 Structure of Data

what format is the data in? which columns are present + datatypes

Name	Data Type	Description
x	float64	Position of cell in pixels along the horizontal axis
y	float64	Position of cell in pixels along the vertical axis
frame	int32	Number of frame, with frame rate of 1 frame per second
mass short	float64	Brightness of cell in 340nm channel
bg short	float64	Background in 340nm channel
mass long	float64	Brightness of cell in 380nm channel
bg long	float64	Background in 380nm channel
ratio	float64	Calculated as mass short divided by mass long
particle	int32	Identification for each particle

Table 3.1: Description and data type of all columns present in the data matrix.

3.2 How it was generated

exprimental setup, what types of t cells where used?, apc layer, explain steps in experiment

- Date: 18/12/23
- Cells: Jurkat wt labelled with Fura-2
- Sample: PDMS coated with OKT3 (positive control)
- Imaging: SDT3, ratiometric Ca imaging, 340nm & 380 nm, Total cycle time 1000ms (-¿ 1 frame per sec in sum/ratio image)
- pixel size: 1.6 um / px

3.2.1 Measuring Calcium Concentration

how is the calcium concentration measured? different wavelengths and then ratio between them, show example video frame

3.2.2 Processing

tracking of particles (in sum of two images), numbering them, removing bad ones (too out of focus, too short)

4 Optimization Algorithm

objective, mathematical formulation of problem

4.1 Algorithm Name

algorithm description
  pseudo code for algorithm
  [proof of convergence, if applicable]

5 Results

6 Conclusion

Bibliography

[AH24] K Maude Ashby and Kristin A Hogquist. "A guide to thymic selection of T cells". In: *Nature Reviews Immunology* 24.2 (2024), pp. 103–117.

[Gan97] William F. Ganong. "Circulating Body Fluids". eng. In: *Review of medical physiology*. 18. ed. Stamford, Conn: Appleton & Lange, 1997, pp. 486–488. ISBN: 9780838584439.

[Hag18] Kimberly Hagel. *Positive and Negative Selection of T Cells*. 2018. URL: https://immunobites.com/2018/08/20/positive-and-negative-selection-of-t-cells/ (visited on 06/21/2024).

[JRB14] Noah Joseph, Barak Reicher, and Mira Barda-Saad. "The calcium feedback loop and T cell activation: how cytoskeleton networks control intracellular calcium flux". In: *Biochimica et Biophysica Acta (BBA)-Biomembranes* 1838.2 (2014), pp. 557–568.

[KCF18] Brahma V Kumar, Thomas J Connors, and Donna L Farber. "Human T cell development, localization, and function throughout life". In: *Immunity* 48.2 (2018), pp. 202–213.

[Lew01] Richard S Lewis. "Calcium Signaling Mechanisms in T Lymphocytes". In: *Annual Review of Immunology* 19.Volume 19, 2001 (2001), pp. 497–521. ISSN: 1545-3278. DOI: https://doi.org/10.1146/annurev.immunol.19.1.497. URL: https://www.annualreviews.org/content/journals/10.1146/annurev.immunol.19.1.497.

[ML23] Jonathan Morgan and Alan E Lindsay. "Modulation of antigen discrimination by duration of immune contacts in a kinetic proofreading model of T cell activation with extreme statistics". In: *PLOS Computational Biology* 19.8 (2023), e1011216.

[SKJ09] Jennifer E Smith-Garvin, Gary A Koretzky, and Martha S Jordan. "T cell activation". In: *Annual review of immunology* 27 (2009), pp. 591–619.