Modeling ionic mechanisms of GABA-mediated excitation through the expression of NKCC1 and KCC2

Anna Chen[1,5], Xintong Lin[2,5], Dustin Miao[3,5], Amy Sun[4,5]

La Salle College Preparatory, 3880 E Sierra Madre Blvd, Pasadena, CA 91107[1] ; Union High School, 6201 NW Friberg-Strunk St, Camas, WA 98607[2] ; The Harker School, 500 Saratoga Ave, San Jose, CA 95129[3]; Winchester High School, 80 Skillings Rd, Winchester, MA 01890[4]; Boston University, Boston, MA 02215[5]

Introduction

- Gamma-aminobutyric acid (GABA)
  - An amino acid that acts as the **major inhibitory neurotransmitter** in the central nervous system

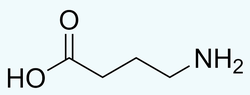


Figure 1 *The molecular structure of GABA*

- Responses to GABA are determined by cellular chloride (Cl^- levels), which are controlled by the **Na–K–Cl (NKCC1)** and **K–Cl (KCC2)** cotransporters
  - NKCC1 is a chloride "importer", while KCC2 is a chloride "extruder". Both are important for the neuron's ability to maintain ion homeostasis.
- Previous studies show that the change in GABA's effect is a result of a shift in the expression from NKCC1 to KCC2 as a neuron matures[2]
  - During early stages of development, **GABA plays an excitatory role** by depolarizing the cell due to high levels of intracellular Cl^- maintained by NKCC1
  - In mature cells, **GABA plays an inhibitory role** and hyperpolarizes the neuron due to lower levels of intracellular Cl^- levels caused by chloride extrusion by KCC2
- Our goal is to use a computational model to analyse how the expression of these transporters affects GABA's role as a neurotransmitter as a neuron develops

Methods

- Adapted the Lewin-Aksay-Clancy neuronal model that was based off of experimental data[1]
  - Simulates ion homeostasis in a post-synaptic CA1 pyramidal cell with 183 compartments
  - Ionic channels: NaCaX, NaKATPase, NKCC1, KCC2, GABAA

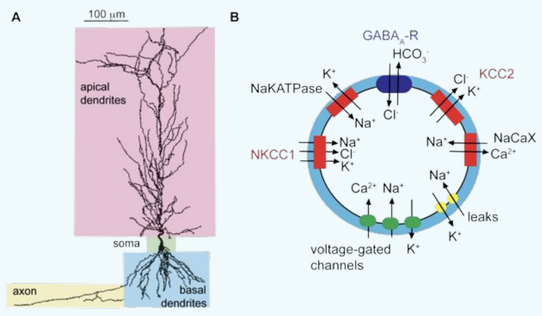


Figure 2A *CA1 pyramidal cell*
Figure 2B *Diagram of a single compartment*

  - Applies brief square pulses of GABA transmitter to simulate high-frequency electrode stimulation of CA1 interneurons
- Altered expression level of protein transporters by changing the maximum capacity to simulate neuronal development

$$c_m \frac{\partial V}{\partial t} = -(i_{Na^+} + i_{K^+} + i_{Cl^-} + i_{Ca^{2+}} + i_{HCO_3^-}) + \frac{1}{r_1} \cdot \frac{\partial^2 V}{\partial x^2}$$

Figure 2 *The cable equation*

- Tracked membrane potential over 2.5 seconds of neural activity, using the forward Euler method to approximate the Cable equation

Results

Figure 3
A 3-dimensional visualisation of the change in GABA's effect on the neuron's spike frequency

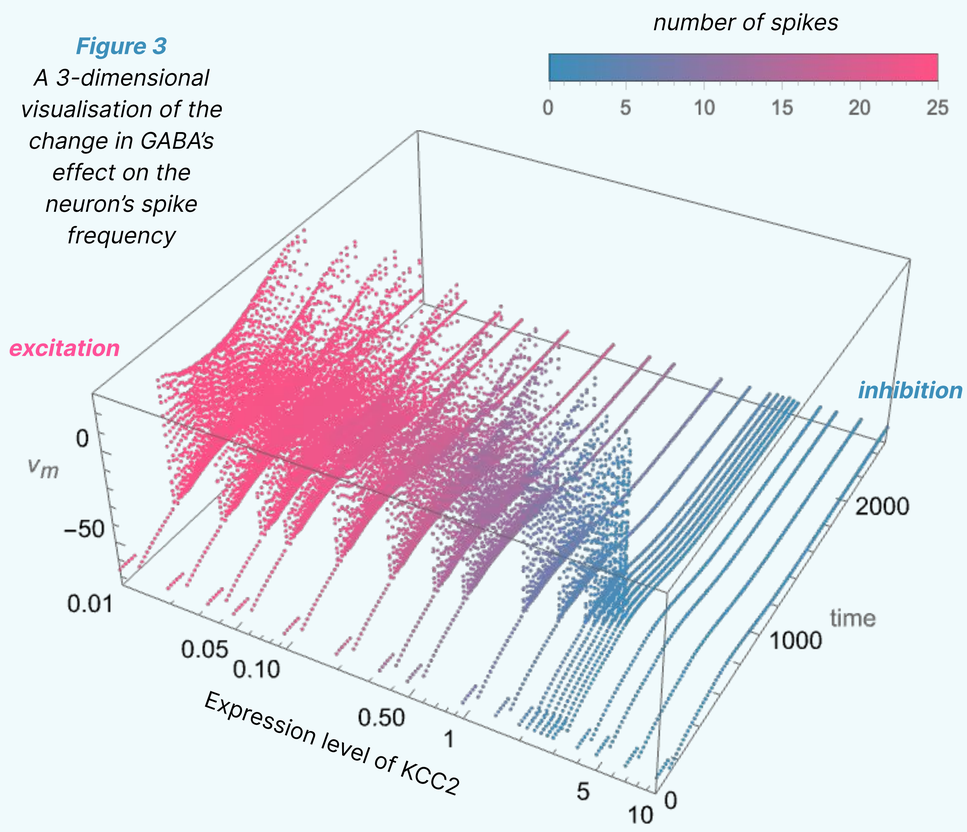


Figure 4A
Changing expression of NKCC1

Figure 4B
Changing expression of KCC2

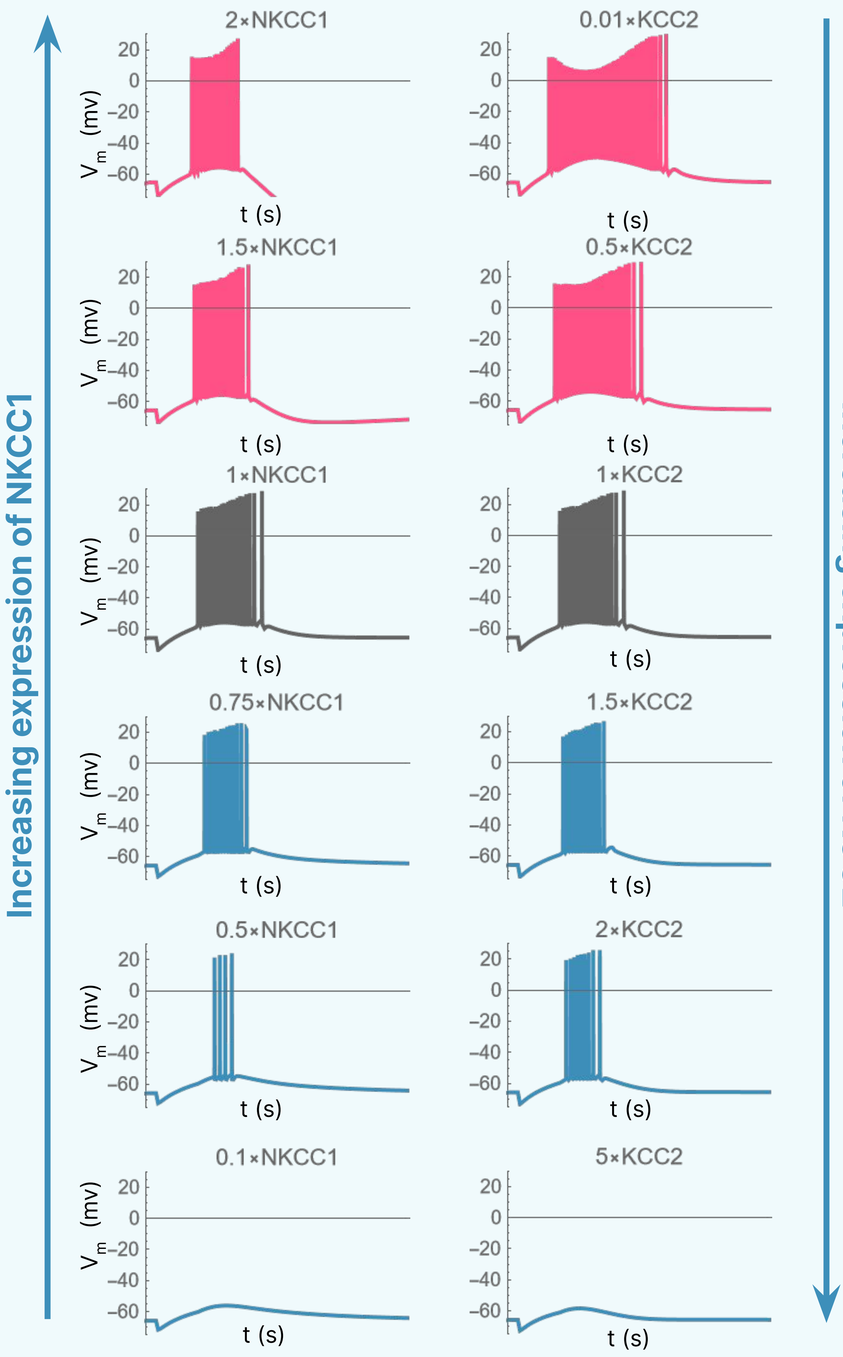


Figure 5A

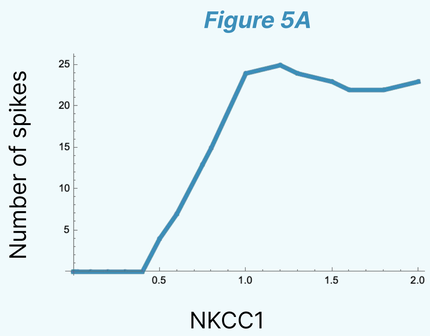
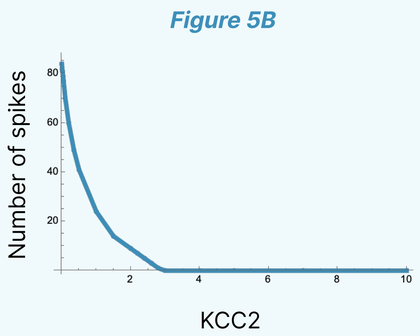


Figure 5B



Discussion

- In **Fig. 4A**, the maximum transport rate of NKCC1 was multiplied by 2, 1.5, 0.75, 0.5, and 0.1
  - When increased (2x, 1.5x), the resulting simulation showed the neuron spiking more frequently compared to the control (1x)
  - When decreased (0.75x, 0.5x, 0.1x), the spikes were less frequent, until eventually the neuron did not spike at all (0.1x)
- In **Fig. 4B**, the maximum transport rate of KCC2 was multiplied by 0.01, 0.5, 1.5, 2, and 5
  - When decreased (0.01x, 0.5x), the neuron spiked more frequently compared to the control (1x)
  - When increased (1.5x, 2x, 5x), the spikes were fewer, and at 5x, the neuron did not spike at all
- **Fig. 5A** and **5B** plot the number of times the neuron spikes against the expression level of each transporter on a logarithmic scale
  - As NKCC1 is expressed more, a higher internal Cl^- concentration causes the GABA stimulation to depolarize the cell, leading to more action potentials. **Immature cells express a higher level of NKCC1.**
  - On the other hand, as KCC2 is expressed more, the neuron has a lower internal Cl^- concentration, and GABA acts as an inhibitor, preventing the cell from spiking.

Limitations

Our model simulated and recorded the action of a single neuron, so the data does not represent GABA's effect on the entire neural system. A multi-neuron cluster will require greater optimization and computing power.

Further Research

Modeling the progression of GABA's effect on neuronal development can provide insights into its role in governing brain development and function, such as during epileptic seizures in the developing brain. Additionally, a sequel to this project could focus on finding the exact threshold for the swap between excitatory and inhibitory GABA, which has significant applications within the medical field in regards to manipulation of Cl^- dynamics in favor of a particular GABA effect.

References

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