A Supplementary Guide to Fundamentals of Cellular and Molecular Neuroscience

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1 Introduction

Welcome to Fundamentals of Cellular and Molecular Neuroscience (Neur 385/Bioc 385/Bioc 585)! The goal of this course is to provide you with a basic understanding of what we know the nervous system can do, how we've uncovered these characteristics, and what remains unanswered as of yet. Though it may be daunting at first to those with little biological background (I had none when I took the course), there is no reason you can't succeed! It just may take a lot of extra work to play catch up with your peers. However, despite the challenge, this has been my favorite course at Rice, and I hope you find the same enjoyment I did in it too!

As you may have seen on the syllabus, our course is broken into 3 sort of "chunks", with multiple subunits within them. Each of these "chunks" will be covered on an exam — 2 midterms and one final. You may be wondering if the exams are cumulative in this course, and in a way they are. We initially cover the biochemistry and biophysics required to understand the higher level processes covered in later units. So long as you devote time to in depth understanding of the units as they are taught, you should be golden. You may be thinking "Same goes for every class", and it is definitely true that devoting a large amount of time to any course material will help with understanding. However, this course builds upon itself, again and again, and, personally, it required many hours in Fondren reading our textbook and others — namely because I was lacking necessary background, but I still found the time I invested in making sure I completely understood the material invaluable. The first "chunk" of material is the most difficult of the three by far, but if you can get through it while maintaining a reasonably high level of understanding, you should do well in the course. Plus, think of it as a sort of bonus that the later material becomes more and more interesting as time goes on! Ever wonder why things are sometimes so hot they feel cold? How about why we have blind spots in our vision? Why is it that chewing foods in differents parts of our mouth make them taste slightly different? All these questions (and more!) we'll get to after Chunk 1.

Don't get me wrong though — Chunk 1 has some pretty interesting content as well! And, if you're really interested in the biophysical basis behind neuronal communication, you should find it super cool! However, we'll save "chunk" breakdowns for the introduction to each.

The textbook for this course will be **Neuroscience 5th Edition by Purves**. Many people end up not using this book during the course, but doing the recommended readings really helped me internalize and understand the material to a high degree. It's very well written, the diagrams are great, and some exam questions *may* cover material from the readings that are not mentioned in class. Additionally, the same book is used in Fundamental Neurosystems (NEUR 380 — spring only), so purchasing the book, or renting for 2 semesters, may be a cheaper option if you plan to continue the Neuroscience track next semester.

1.1 Literature Review

As you very well may already be aware, Neuroscience is a relatively unexplored science. I make this claim due to the plethora of unanswered questions standing within the field itself, not due to any shortage of investigators (although I would argue some areas are experiencing this!). Thus, in order to give a somewhat accurate grasp of the state of the field, Dr. Caprette shall be organizing you into groups based on year, GPA and background to read *actual* research papers, current and fundamental to the state of the science. By doing so, you shall gain experience

reading and comprehending publications to a high level, even without expertise in the field — if you have not done this before, I recommend carving out a large chunk of time and annotating while reading. I've personally been reading publications for my research for a few years, and it still takes me approximately 5-6 hours to fully understand a paper, especially if I skim the cited papers to get more background.

Dr. Caprette may have mentioned in class that a given number of exam questions will cover the information you learned from reading the publications. Not to worry! He will provide you with a list of questions from which he will choose approximately 3 for the exams, so you and your group have time to discuss them beforehand. You may bring your annotated papers with you to exams for reference, so you do not have to memorize findings from the paper. *However*, there are limitations to the last 2 sentences. First, you may not get help from the TAs/Fellows/other groups for the exam questions based on the assigned publication. Treat these questions like actual exam questions — we can help you with general paper understanding but not with the questions themselves. Second, do not write your answers on the article you bring with you on exam day. You may jot down points you want to remember, but Dr. Caprette will collect your papers after the exam and check if you wrote down and just copied your answers. Utilize your group time for paper understanding to avoid even needing to do this!

1.2 Homework

You will have one homework assignment per "chunk", due a week before the exam. These homeworks make up approximately 10You may hear this often, but I highly suggest you be sure to start these somewhat early to avoid issues. Many students find they struggle with questions right before the deadline and tend to overwhelm the TAs at the last minute, making it harder for everyone to get help in the allotted office hours. I found it easiest, personally, to look at the questions as we progress through the unit, and complete ones I felt I knew how to do as soon as I could. I would then return to the questions later, seeing if I had any questions or felt the answer was different than how I initially thought.

1.3 Office Hours and Reviews

Please please please go to office hours! Don't just wait until the last minute to ask your questions — we're here because we love the course and *want* to help you! The sooner we can catch you up on material, the easier the next units will be for you to understand! This course is not one in which you want to fall behind. It will just become harder and harder to get back up to speeed the longer you wait. I also highly recommend attending Dr. Caprette's office hours if you feel you are really struggling, or just for conversation. He's probably one of the friendliest professors I've had at Rice, and thus you should take advantage of having a resource who will go out of his way to make himself available to you. This is not the case for all professors at Rice (or any institution, really), so I suggest you utilize him while you can!

Oftentimes, the TAs will host approximately 2 review sessions prior to an exam. I used to go to both of these, since different TAs would explain concepts in different ways each night, but I recommend you go to one. Definitely take advantage of the time after the review session when you can ask one-on-one questions. We will cover the concepts we believe you need to have an understanding of in order to do well on the exam. However, keep in mind we don't see answer keys for your homeworks or even the blank exam, so trying to get a correct answer out of us is useless. We are just here to guide your thinking, and help steer you in the right direction — attempting to get us to do your work for you is pointless (you'd be surprised how many students come to office hours just repeating a short answer question on the homework instead of asking conceptual questions).

1.4 One Last, Quick Note

At the cellular and molecular level, conditions are nearly never as ideal as they are presented to be in courses. Dr. Caprette has an appreciation of this fact, and, in an effort to prepare you to the fullest for your future in cellular and molecular neuroscience, he has a tendency to present complex situations. Thus, wording becomes a notable player in the determination of your success in the course — both on his behalf, and yours. Even if an answer is correct, Dr. Caprette will penalize for what he deems to be improper wording, or too long of explanations. It sounds daunting, but this is not too difficult. Pay close attention to the way he presents concepts in class, try

thinking about when these processes would fail, and so on. Ensure that your answers to questions are never too lengthy — this is good practice for the real world, and makes grading easier (which is more favorable for you).

This guide shall aim to provide you with resources, practice problems, and additional explanations of topics taught in class. In no way does this replace class — Dr. Caprette provides great explanations and very useful feedback, which I strongly encourage you to take advantage of. Thus, I hope this guide eases your transition, and I welcome you to reach out to me or any of the other TA's (whose contact info you may find on the canvas site) with any questions you may have. My email is Ariel [dot] K [dot] Feldman [at] rice [dot] edu.

2 Chunk 1: The Sciences

I affectionately refer to the first chunk of this course as the science chunk, since the material provides you with a strong physics/biology/chemistry background in context of understanding the fundamentals of neural communication and processing.

2.1 Biological Basis of Behavior

In order to put the rest of the course into perspective, it is imperative that we cover the findings that sparked to life the field we today know as Neuroscience. We may as well start at the beginnings of the field, with the now very outdated pseudoscience of phrenology. Franz Gall, its founder, firmly believed that the measurements of one's skull could provide telling information regarding one's personality. Though Gall's intentions were genuine (and he had the *gall* to spread them [insert laughing track here]), the pervasive nature of phrenology led to a delayed acceptance of the brain working as a functional system on a larger scale — preferring a view of clear, delineated functions for distinct areas of the brain. He did, however, have one very accurate theory: the brain is composed of functional regions. Though his assertion that there were discrete functional regions proved false — your brain is much more elastic than that would imply — he *was* the first to connect behavior and biology.

J. Hughlings Jackson followed Gall's impact, but took a different approach to uncovering the basis of neurology. Rather than focusing on exactly what specific areas did in the brain, Jackson was concerned with the effect disabling certain regions would have on human behavior. So, what were some of his contributions to what we know as the field of Neuroscience today? Perhaps his most influential contribution derives from his work with partial (Jacksonian) seizures, resulting from irregular electrical activity within a specific brain regions. Noting a correlation between injured region and behavioral impact, Jackson hypothesized the existence of functional regions within the brain. Specifically, within the cortex.

2.2 Studying the Nervous System

Neural circuits are diverse subsets over the totality of neurons. They make up the primary components of neural systems, processing specific types of information. Remember Gall and Jackson's theories on the functionality of specific regions in the brain? Rather than devote *regions* to processes, such circuits of interconnected neurons arise to perform specialized functions. What's the distinction? Think of it as though many circuits can be intertwined with each other (your brain looks like a big mess of axons and somas — the vast majority of neurons are greatly intertwined), and performing different functions depending on what other neuron their next point of contact is. On the contrary, a region would indicate that the brain is segmented into discrete chunks by function. We're a lot more malleable than that!

There are 3 main types of systems we deal with in Neuroscience: sensory systems, motor systems and associational systems. Sensory systems provide information about the state of the organism and its environment, while motor systems organize and generate actions. Associational systems provide a sort of communication between sensory and motor components, providing a basis for higher level cognition. Simple enough, right?

Let's go through the cellular components of the nervous system, especially for those of you with a weaker biology background. One early, incorrect theory of nervous system structure supported by Golgi was the idea of a reticulum. No, not the endoplasmic reticulum you may remember from any lower level biology course. Rather, this

reticulum is the idea of a continuous nerve cell network, connected by protoplasmic links. However, Cajal (he'll keep popping up in your future Neuroscience courses) maintained the idea that the nervous system is composed of many discrete entities — what we know today to be neurons. He supported the idea of communication between different entities where their connection is not continuous (synapses). Cajal was correct in his assertion, though there is a type of synapse called a *gap junction* in which cytoplasmic connectivity is maintained across two discrete entities. However, we will cover synapses in a later section.

Glial cells are another important piece to the neural puzzle — not only do they insulate axons by wrapping around them, but they repair damaged areas of the nervous system. They can be broken down into 2 types: macroglia and microglia. You should not be asked to discriminate between the two, but, since they serve different purposes, I shall make the distinction regardless. Microglia are related to your immune system, and thus are responsible for immune responses in the brain. They're scavenger cells — basically glorified garbage collectors, removing debris from the environment surrounding neurons. Macroglia encompass Astrocytes and Ogliodendrocytes, and strike fear into the hearts of students trying to memorize their names. Once again, don't worry about names, just what glial cells can do in general. Macroglia are restricted to the central nervous system (CNS), and serve different functions. The main purpose you would need to be aware of is their role in maintaining the chemical environment surrounding the neurons. Ogliodendrocytes wrap myelin sheaths around axons in the CNS, which is the role of Schwann cells in the peripheral nervous system (PNS). Sometimes, macroglia can act as stem cells and impact neural regeneration — encouraging where useful, discouraging where harmful.

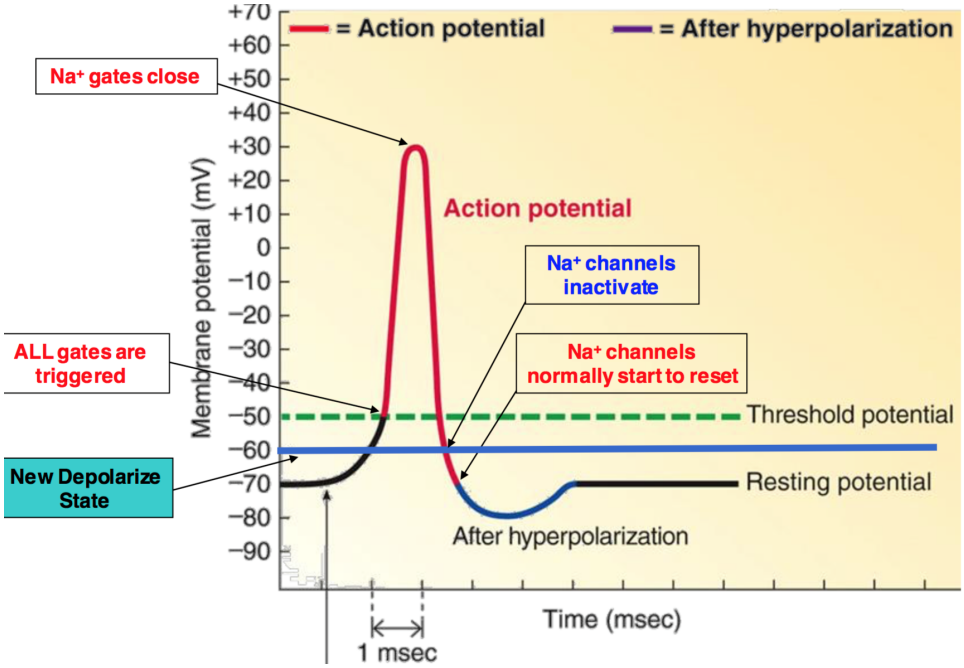
2.3 Animal Electricity

It is fair to say that modern Neuroscience is founded upon 2 key concepts: Cajal's neuron doctrine and the specialization of cortical function. From these two concepts, the rest of the field builds upon itself into a more recognizable version of the Neuroscience you *likely* enrolled to study. Familiar with the idea of shocking certain parts of the brain to directly impact (or even control) behavior? Perhaps that sounds like something straight from a sci-fi novel or movie to you — but, due to the electrical nature of neural signals, this is all too possible (see my research for more *self plug*).

The first man to discover this capability, Galvani, did so by passing current through the sciatic nerve of deceased frog legs, causing reanimation in the form of kicks. It's no wonder he served as the inspiration for Dr. Frankenstein — in his time, such an event would be attributed to activity of an animal's spirit. Thus, to the laiety, in a sense Galvani was reviving the dead, which you can only imagine did not go over well with the Church. However, we are here to discuss the specifics of his work. By simply touching a cut sciatic nerve to either the muscle it served and to another nerve, producing pulsations of kicks. How, though, is this possible if the animal is dead, and the nerve is severed? Think of Volta! So long as you have two dissimilar substances that possess the ability to conduct electricity, simply touching the two together will cause a detectable (important in general, but not necessarily for this course) current to flow.

2.4 Voltage-Dependent Membrane Permeability

A basic overview of what we shall discuss in this section is as follows: The use of the voltage clamping technique provided substantial evidence indicating action potentials are generated by neurons, and arise from changes in membrane permeability. Leading up to an action potential, a neuron experiences a rapid increase in the permeability of the membrane to Sodium (Na^+), followed by a prolonged rise in Potassium (K^+) permeability.



What do we consider as the initiation of an action potential (AP)? Once the voltage across the membrane surpasses (becomes more positive than) the threshold value (hence the name), we consider an action potential as being initiated. A change in the voltage across the membrane drives a change in membrane permeability to Na^+. For those of you lacking an electrical engineering background, you may not be familiar with the idea of capacitive current. In the context of neurons, it may help to think of this as an initial response to the redistribution of charged ions across the cell membrane. When a neuron is in a hyperpolarized state, the axonal current is essentially the capacitive current. However, the neuron must depolarize to produce an AP. Thus, as the voltage across the membrane reaches 0 mV, the current within the axon rapidly increases, creating an inward ionic current. A delayed, slow rising outward current is also generated by this depolarization of the neuron. It is important to remember that, unlike the circuits you are taught in physics courses, current in neurons is driven by the movement of positive charges!

So how did voltage clamping come into the picture? Voltage clamping allows for simultaneous control of the voltage across the membrane, and recording of the change in said potential. But why is this important? Being able to vary the V_m allows us to accurately predict E_{ion} for the ions that contribute to V_m. Another important note about the voltage across the membrane is that, when less than resting membrane potential ($V_m < E_m$), no appreciable ionic current is flowing. We can break the currents observed during depolarization into 2 types: early current and late current. Very creative, I know — I did not name them, but I must admit their names are quite memory efficient. And, as a computer scientist, I appreciate memory efficiency. Regardless, let's review these two concepts, as they are imperative to your understanding of the forces that drive an AP.

Early current is known to have a somewhat U-shaped dependency on the V_m. Essentially, what this means is that the current increases as V_m approaches 0 mv, and decreases as the membrane depolarizes further. This is more of a lower case n-shaped relationship than a U-shaped one, but you should take away that there is a hyperbolic relationship between early current and V_m. On the other hand, late current increases *monotonically* with an increase in V_m — i.e., the two are directly related.

Let's jump back to voltage-clamping to get a better understanding of early and late currents. Since we know that voltage-clamping is a tool that can be used to deduce which ions are contributing to what electrical signature, we can voltage-clamp to determine the cause of the early current. When we clamp at +52 mV, the E_{Na^+} value, we notice there is no early current. So, what does this indicate about Na^+'s role in generating the early current? From this observation, we can (correctly) hypothesize that the early current is the result of an influx of Na^+ ions entering the axon during depolarization. However, we're all burgeoning scientists here — we know a mere observation is not enough to make a claim. So let's test it!

We can test this by removing all Na^+ outside of the neuron, and noting the impact this has on the early current.

What do you expect will happen? We know that E_{Na^+} becomes negative, since there is now *no* Na^+ outside the cell — meaning that if membrane permeability to Na^+ is increased, the few Na^+ ions already in the axon will flow outwards, following their concentration gradient. It follows, then, that the current driven by Na^+, I_{Na^+}, should be directed outwards. Thus, we may conclude that the early current is driven by an influx of Na^+ ions in the axon.

What, then, about the late current? When performing the same testing as above, no change was observed in the late current. What does this mean? The late current is, in fact, driven by a different ion than NA^+. Contributing to this idea of two different ions driving the early and late currents is the time delay between them. Such a delay indicates two different ionic permeability mechanisms being triggered by changes in V_m. Additionally, the amount of K^+ leaving the axon closely correlates to the magnitude we observe of the late current. All of these imply a different ionic trigger for the two currents, and thus we shall assume this to be true.

Perhaps you heard Dr. Caprette mention some neurotoxins in class — remember them! He will likely only evaluate your knowledge on ones he mentioned in class. Tetrodotoxin blocks Na^+ current w/o affecting K^+ current. On the other hand, Tetraethylammonium ions block K^+ currents w/o affecting Na^+ current. If Dr. Caprette did not mention these neurotoxins by name, then don't worry about memorizing them. It is just important you know the methods by which these hypotheses may be tested, to give you a more realistic understanding of the field.

2.5 Testing The Bernstein Hypothesis

2.6 Discovering The Diffusion Potential

Before we move forward in this course, it is crucial you understand the implications of the resting potential of a neuron — what it is, how it works, and why it tends to be the values it is. What do we know about the ions in a neuron? To start, we know the ions that populate the axon mainly consist of sodium (Na^+), Potassium (K^+), and Chlorine (Cl^-).

2.7 Ionic Basis Of An Action Potential

2.8 Conduction Of The Nerve Impulse

2.9 Ion Channels and Transporters

This section will be a bit dense, so I'll a quick overview of what I expect you to take away from it. First, you should understand that active transporters are membrane proteins that create and maintain ion gradients. Though active transport regulates many ions (Cl^-, H^+, etc.), perhaps the most famous of them would be the Sodium-Potassium pump. This will appear again and again throughout your neuroscience career — thus, I hope you have an accurate grasp of active transporters after this. Second, you should understand what an ion channel is, and the role they play in maintaining concentration gradients. An ion channel is a transmembrane protein that gives rise to selective changes in ion permeability — i.e., they will let more of specific ions into the cell, and barr others from entering. There are 3 types of ion channels: ionotropic, metabotropic, and electro-chemical. We will further detail these types in a moment. Finally, you should better understand how transporters create concentration gradients, and thus generate electrical signals by causing ion fluxes through the ion channels in the cell membrane. If you are confused at all about these topics, don't hesitate to reach out to me for clarification!

Seeing as we've got a lot to unpack here, let's dive right in and discuss the ion channels underlying APs. To begin, we will revisit our old friends Hodgkin and Huxley. If you recall, they predicted ion channels would possess the followin characteristics:

1. allow ions to cross the membrane at high rates.
2. make use of the electrochemical gradient across the membrane.
3. could discriminate between Na^+ and K^+, which meant they would allow only one ion to flow across the membrane under relevant conditions.

4. could sense V_m, opening only at the appropriate levels.

How did they support these claims?

Patch clamping allowed Hodgkin and Huxley to provide the first *direct* evidence of a voltage-sensitive, ion-selective channel. Whereas voltage-clamping would allow study of changes in aggregated current, patch clamping allowed for much more individualized study — they were able to access specific channels, as opposed to overall changes. This is where we must make the distinction between microscopic and macroscopic currents. Microscopic currents measure that across a single channel, typically in the range of 1-2 pA for I_{Na^+}. However, this is *still* measuring the transfer of thousands of ions per seconds. Macroscopic currents, on the other hand, are the summation of the microscopic currents of a large number of ion channels, distributed over a large region. It may be helpful to see it as such:

$$I_{macro} = \sum I_{micro}$$

Let's take a closer look at microscopic current (ha!) by specifically investigating microscopic I_{Na^+}.

As I stated earlier in this guide, current in the neuron is carried by the positively charged ions in the neural model. Thus, we know Na^+ "carries" the current, and we also know that Na^+ moves into the axon from the environment when $E_{Na^+} > V_m$.

2.10 Channel Function

2.11 Discovering The Sodium Pump

2.12 Studying Channel Structure

2.13 A Diversity Of Ion Channels

2.14 Introduction to Synaptic Transmission

3 Chunk 2: The Communication

3.1 Pre-Synaptic Mechanisms: Role Of Calcium Ions

3.2 Quantal Release And The Vesicle Hypothesis

3.3 Postsynaptic Membrane Potentials

3.4 Synaptic Plasticity In Aplysia