

An Information and Communication Theoretical Analysis of the Nervous System towards Internet of Nano-Bio Things

by

Naveed Ahmed Abbasi

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**An Information and Communication Theoretical Analysis of the Nervous
System towards Internet of Nano-Bio Things**

Koç University

Graduate School of Sciences and Engineering

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Naveed Ahmed Abbasi

and have found that it is complete and satisfactory in all respects,
and that any and all revisions required by the final
examining committee have been made.

Committee Members:

Prof. Özgür Barış Akan (Advisor)

Prof. Fatih Alagöz

Prof. Alper Tunga Erdoğan

Assoc. Prof. Sinem Çöleri Ergen

Assoc. Prof. Berk Canberk

Date: _____

ABSTRACT

Amongst the various networks inside the human body, the nervous system forms the largest and the most complex nanonetwork. This network communicates with several types of internal and external stimuli on a molecular scale and interfaces with other peripheral systems of the body to help it function normally. Study of naturally existing molecular communication systems from information and communication-theoretical (ICT) perspective provides a novel approach in understanding these networks. Therefore, the main motivation of the proposed thesis is to analyze the nervous system and the communication happening within this network from an ICT perspective. In particular, we target modeling of the nervous system from a network perspective, experimental testbeds for molecular communication based on nervous system, insulin-glucose system as a peripheral systems of the nervous system and describe applications of these system in the Internet of Bio-Nano Things (IoBNT). The study of the nervous system will provide novel directions to augment the existing diagnostic and treatment techniques as well as provide a framework for artificial nanonetworks that may be based on the nervous system.

ÖZETÇE

İnsan vücudundaki iletişimci yapılar arasında, sinir sistemi en geniş ve karmaşık nano-ölçekte oluşmaktadır. Bu yapı, kimyasal ve elektriksel uyaranlarla moleküler düzeyde iletişim kurmakta ve vücudundaki evresel sistemleri ile ara yüz oluşturarak normal olarak almalara yardım etmektedir. Doğal olarak mevcut moleküler iletişim sistemlerinin bilgi ve iletişim teknolojisi (BT) perspektifiyle araştırılması, bu alanın anlayışında yeni bir yaklaşım sağlamaktadır. Bu nedenle, önerilen tezin temel motivasyonu sinir sistemi ve bu alanda gerçekleştirilen iletişimin BT perspektifiyle analiz edilmesidir. Özellikle, sinir sisteminin bir perspektifiyle modellenmesini, sinir sistemine dayalı moleküler iletişim için deneysel test düzeneklerini, sinir sisteminin evresel sistemi olarak insülin-glikoz sistemini ve bu sistemlerin Biyo-Nano Nesnelerin İnternetinde (BNN) uygulamalarının tanımlanmasını hedefledik. Sinir sisteminin araştırılması mevcut teşhis ve tedavi tekniklerinin geliştirilmesinde yeni istikametler sağlayacak, aynı zamanda, sinir sistemine dayalı olabilecek yapay nano-ölçekte yapılar için bir atak sağlayacaktır.

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Chapter 1

INTRODUCTION

With the recent advancements in nanotechnology, things are quickly moving from traditional communication architecture to micro and nano scales. Nanotechnology is one of the frontiers of the scientific research in the current age having a huge set of applications that are being realized in a variety of environments. Some of these include bio-hybrid systems [5], nanoscale sensing [6], intelligent drug delivery [7] and body area networks (BAN) [8]. Although nanotechnology promises a big revolution in the future, the implementation challenges are also huge.

Due to their simple architecture, consisting of a small number of molecules along with power and size limitations, nanomachines have little processing, memory and networking capabilities. These limitations can be overcome by dense deployment of nanomachines in networked environments, termed as *nanonetworks*. Nanonetworks is an emerging field of study where nanomachines communicate to work beyond their individual limited processing capabilities and perform complicated tasks.

The deployment of nanonetworks raises a number of questions as to which techniques can be used optimally under the challenging conditions. Different communication paradigms are suggested for the realization of nanonetworks in physical environment such as nanomechanical, acoustic, electromagnetic, however, the most promising one is molecular communication [9], where molecules are used to encode, transmit and receive information. Since the current work is related to molecular communications, we will delve into more details of this particular communication paradigm in the following sections.

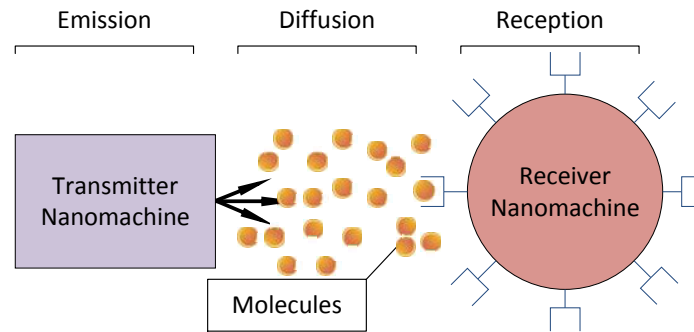


Figure 1.1: Diffusion-based molecular communication.

1.1 Molecular Communication

As stated earlier, molecular communication is found to be the most suitable method for nanoscale communication due to its compatibility with both biological and non-biological systems. Although several parallels can be drawn between traditional communication systems and molecular communication systems, this paradigm is significantly different from traditional communication systems since it uses particles to carry information instead of electromagnetic carriers. Thus, the ICT-based parameters of molecular communication paradigm are affected by physical and chemical properties of network entities such as diffusion coefficient of the channel, structure and type of molecules used as information carriers instead of the wave propagation properties. Hence, the classical information science does not suffice to define the fundamentals of molecular communication.

1.1.1 Diffusion-based Molecular Communication

Diffusion-based molecular communications form the basis of the most frequent type of communication within living entities. As an example, communication inside the human body is achieved by diffusion of molecules like for calcium signaling among living cells, neurotransmitters-based nervous communication, and intra-cellular communication between different cell body parts. A typical physical model of diffusion-based

molecular communication is depicted in Fig. 1.1. Within the scope of molecular communication, diffusion-based molecular communication is considered as the most promising paradigm for the applications of nanoscale networks especially inside living bodies.

Diffusion-based communications can be categorized into analog and digital communication similar to conventional communication networks [10]. A fundamental analog communication model is proposed in [11], where information is encoded by continuously emitting molecules with varying concentration. In digital diffusion-based communication, information is encoded into either types of molecules [12], time of release of molecules [13] or the intensity of concentration of molecules [14].

Several models of diffusion-based molecular communication are proposed in literature. One such model is proposed in [10]. Another model, where particles are released into fluid medium with drift is studied and analyzed in [13]. One of the most important issues in developing diffusion-based molecular communication is residual molecules from previous transmission which are considered as noise and various approaches are suggested to maximize the rate of information by reducing this noise in [15]. Moreover, the information capacity of the channel with residual molecules is studied in [16] by modeling it as *channel with memory*. Other external noise sources and ISI are modeled in [17].

Different digital modulation techniques for diffusion-based molecular communication are proposed in [12]. Some of them are compared with respect to their susceptibility to interference in [18]. Some receiver designs are presented in [10, 19], and [20].

Although a significant body of research exists on diffusion-based molecular communication, much work remains to be done. Some of these areas include modeling of molecular communication in subsystems of living entities as well as the effects of asynchronism between the transmitters and receivers.

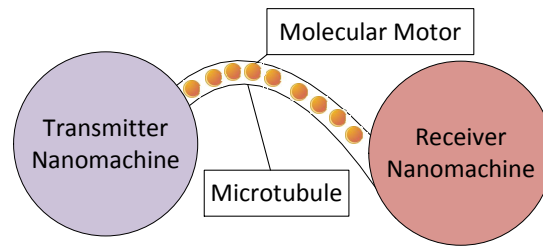


Figure 1.2: Molecular communication with wired active transport using a microtubule.

1.1.2 Wired Active Molecular Communication

Wired active molecular communication between a transmitter and a receiver nanomachine is illustrated in Fig. 1.2. Information molecules in this communication paradigm move along preset paths of microtubules or capillaries to deliver information from the transmitter to the receiver [21]. The flow of information can be self-propelled by molecular nanomotors as in Fig. 1.2 or based on the flow generated by another system such as a blood circulatory system. Based on the assumptions of independence between particles, a simple mathematical model for analyzing information rate is derived in [22]. A Markov channel model with a mathematical formula for calculating transition probability matrix of the model is proposed in [23]. Design and optimization of the shape of a rectangular channel in [24] shows that channel capacity is proportional to the perimeter. A secure wired active channel is proposed in [25], where information molecules are encapsulated inside vesicles while transporting the information. Various approaches for self-organizing microtubule networks such as polymerization/depolymerization and molecular motors are investigated in [26]. Although the effect of channel characteristics such as width, shape and length have been investigated in some studies, particle-particle interactions need to be a part modeling as well.

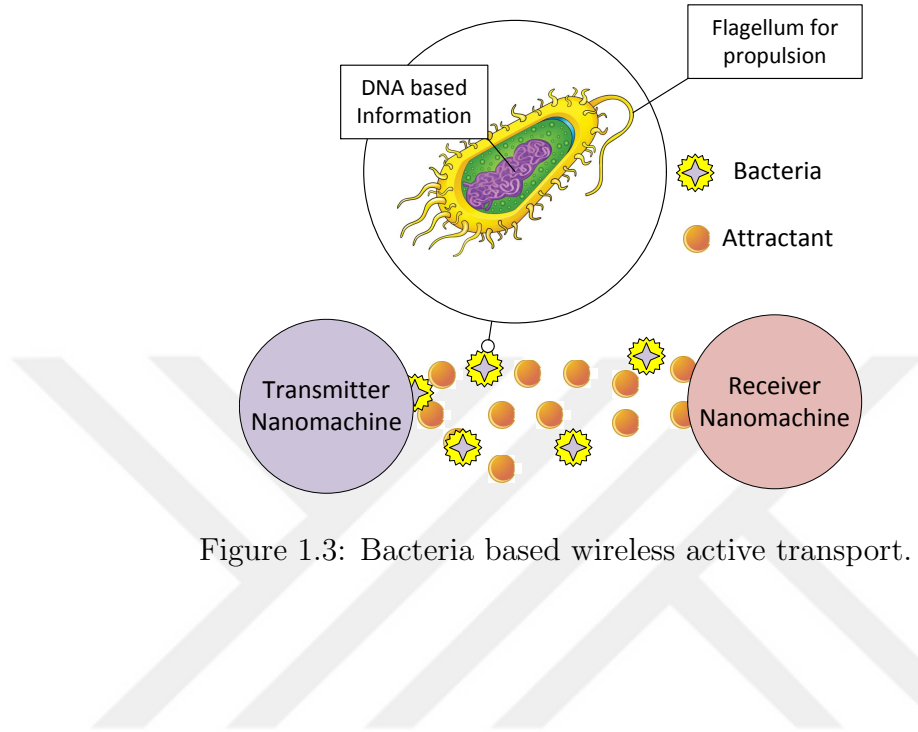


Figure 1.3: Bacteria based wireless active transport.

1.1.3 Wireless Active Molecular Communication

Bacteria-based Wireless Active Molecular Communication

A typical system of wireless active molecular communication paradigm based on bacteria is shown in Fig. 1.3. In this type of communication, first, a bacteria that only responds to a set of specific attractants is chosen as a communication vessel. A DNA-based message is then introduced in the cytoplasm of the bacteria and it is released in the environment. The bacteria propels itself towards the receiver using its flagellum in response to the release of attractant particles by the receiver as shown in Fig. 1.3 [27]. Communication range, channel capacity, throughput and end-to-end delay analysis is done in [28] shows that bacteria-based communication has huge potential. However, a definitive general model that can apply to a variety of bacteria-based communication scenarios as opposed to existing models for specific scenarios is required. A simulation tool for bacteria-based communication, BNSim, is introduced in [29] and discussed in Chapter 3.

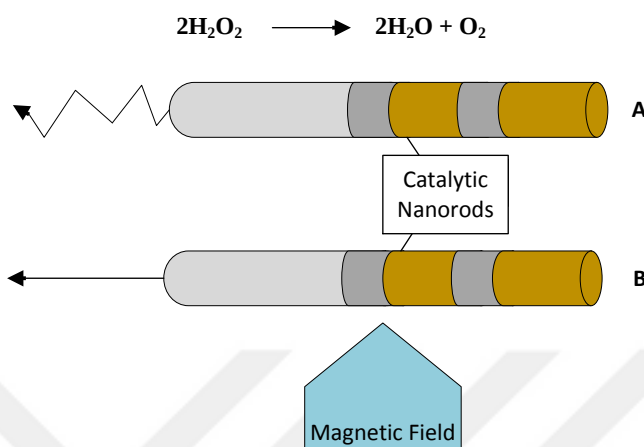


Figure 1.4: Catalytic nanorods in hydrogen peroxide solution without a magnetic field applied (A), and with a field (B).

Catalytic Nanomotor Based Wireless Active Molecular Communication

Catalytic nanomotors are usually platinum or gold nanorod particles that are able to propel themselves and small information containing objects, by catalyzing the free chemical energy present in the environment [27] as shown in Fig. 1.4. Without a controlling magnetic field, the nanorods move in arbitrary directions, however, once under the influence of a magnetic field, they can be forced to travel in specific directions. The speed of these particles depends on their dimensions as well as the atmospheric conditions [30]. Further research is required in the bio-compatibility of catalytic nanomotors since some of the materials used might be dangerous to living things

1.1.4 Physical Contact-based Molecular Communication

Nanocommunication via gap-junctions (GJs) and synapses are two possible kinds of molecular communication via physical contact. In [31], a collision-based mobile ad hoc molecular nanonetwork (MAMNET) is proposed, which uses neuro-spike communication for transmitting information between two nanomachines. It is shown that the size and speed of nanomachines have a direct relation with the average through-

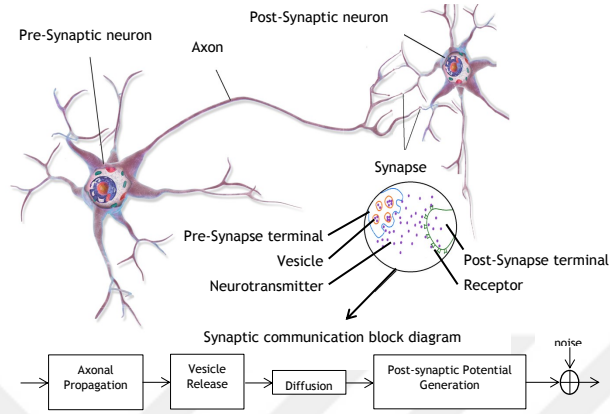


Figure 1.5: Neuro-spike communication between two neurons.

put and a reverse relation with the average message delivery delay. Neurons are used as molecular communication medium in [32]. A microplatform is designed in [33], which is able to pattern the mammalian cells into a predefined network connecting them through gap-junctions. These preliminary studies are samples of achieving the bio-inspired molecular communication networks.

1.2 Molecular Communication Inside the Human Body

The human body is an example of a very large nanoscale communication network, where individual constituents communicate by means of molecular nanonetworks. These networks follow complex cellular signaling processes that usually work on diffusion-based molecular communication principles, wired active molecular communication or through gap junctions. Brief overviews of each are discussed in the following passages.

1.2.1 Nervous Nanonetwork

Communication taking place within the nervous system is based on the arrival of stimuli to nervous cells called neurons. These stimuli are converted into electrical impulses called action potentials (AP). The sources of impulses can be external like

touch, sound and light, or they can be internal such as blood pressure, heart rate, and cardiac output. The effect these impulses is then carried through a network of neurons, called the sensory neurons, to the central nervous system (CNS), which comprises of the brain and the spinal cord. Decisions according to the received impulses made by the CNS are propagated to the muscle cells by a different set of neurons known as the motor neurons.

A neuron is composed of three main parts which are the soma, the axon and the dendrites. The soma is responsible for the spike generation in response to stimuli, the axon is the place where the spikes are conducted and the dendrites function is the reception of the excitation. Spikes or impulses are used to transmit information from one neuron to another. Hence, it is called neuro-spike communications. It can be divided into three main parts. The first one, the axonal transmission, is the propagation of the spikes along the axon. At the end of the axon, there are some specialized terminals, which are called the presynaptic terminals whose function is the release of neurotransmitter packets or vesicles to the gaps among neurons, i.e., synapses. The release depends on the plasticity of the neuron, i.e., the change in the release probability in response to its history. Release of the packets to the synapses initiates the synaptic transmission, which is the second main part of the neuro-spike communications. Since each packet contains many neurotransmitter molecules, following the release the neurotransmitters inside the packets go out and the diffusion of each neurotransmitter begins towards the output neuron. In order to get the diffused molecules, the neurons have many postsynaptic terminals at the dendrite in which the receptors are located. There are two main types of the receptors, which are the ionotropic and the metabotropic receptors. The difference between them is that the ionotropic receptors directly excite the neurons, whereas the metabotropic receptors have indirect effect such that they initiate the metabolic change in the neuron, and this change excites the neuron. At the end of the diffusion, these neurotransmitters bind to the receptors at the postsynaptic terminal. When neurotransmitters bind to the receptors, the movement of ions begins, which provides excitation, and this consti-

tutes the last part of the communications called spike generation. Moving ions excite the membrane potential of the output neuron, and lead to the Excitatory PostSynaptic Potential (EPSP) generation. The level of excitation changes from one packet to another, because of the variation in the behavior of neurons with time, which is called the trial-to-trial variability. Since neurons have some threshold value to be depolarized, the excitation level must be above the threshold. However, one receptor is not sufficient to depolarize the neuron, therefore, the response of a group of receptors belonging to the same neuron are summed. If the total summation is greater than the threshold value, the neuron has sufficiently depolarized and the spike is generated at the output, which completes the information transfer.

1.2.2 Cardiovascular Nanonetwork

Cardiovascular Nanonetwork is a kind of gap junction communication inside the human body. Two most important types of cells within the heart are the *cardiomyocytes* and the *cardiac pacemaker cells* [34]. Cardiac pacemaker cells are distributed throughout the heart and are responsible to spontaneously generate and propagate APs to cardiomyocytes, which then create the beating motion of the heart [34]. Cellular bridges form porous gap-junctions between the various types of heart cells providing a medium for AP transfer [35, 36]. Two cells connected in the GJ by means of channels called connexons are shown in Fig. 1.6. Connexons remain closed in the rest state, however, when an AP arrives, they open and yield an ion diffusion to occur towards the receiver cell, thus causing the AP to be carried forward to the next cell. Generic GJ communication is studied in [37] and [38]. Various mathematical models such as [39, 40] and 3D models such as [41, 42] describe the blood flow through the heart. Electric circuit models of the heart such as [43, 1] are developed on the basis of Hodgkin-Huxley neuron model [44] to understand the operation of the heart based on cardiac APs. GJ channel is modeled from the information theoretical perspective by [45], where a probabilistic physical channel model is provided with a probability mass function for GJ conductance considering the possibility of AP propagation failure. A

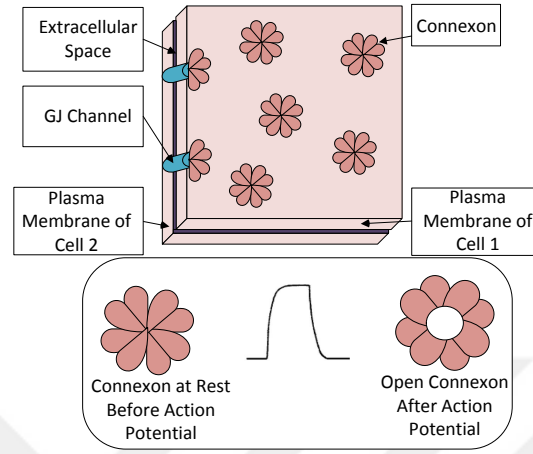


Figure 1.6: Gap-junction communication between two cells. The AP causes the connexon channels to open, starting diffusion between the cells.

closed-form expression for channel capacity and correlation between the decrease in capacity and several heart diseases is studied. Employing the various mathematical [39, 40] and 3D [41, 42] models that describe the blood flow through the heart, the ICT-based models can be improved on large scale.

1.2.3 Endocrine Nanonetwork

The endocrine system comprises a set of glands that secrete different hormones through the circulatory system to their recipient cells [46]. Transmission of hormones depends on concentrations of various materials sensed in tissues. The required amount of hormones are then added to the blood stream by the endocrine system to maintain homeostasis in body [47]. The propagation of hormones depends on blood circulation as well as their Brownian motion within the blood stream [48]. Reception of the hormones is selective because only the target cell can respond to a hormone.

Modeling of entire endocrine system from an ICT perspective is an open research area with huge implications, however, very few studies are present in this field to date. Hormones are considered as modulated molecular information carriers in the model of endocrine nanonetwork in [1]. The suggested framework is shown in Fig.

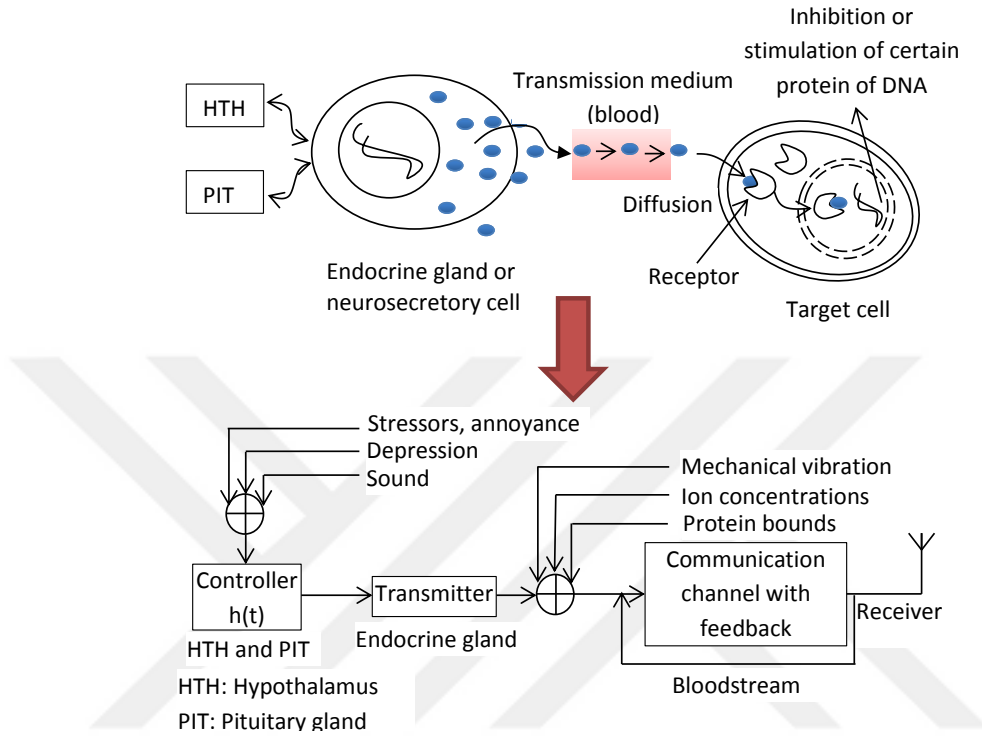


Figure 1.7: Model of the hormonal communication [1].

1.7, where various stimuli cause the Hypothalamus to signal the release of hormones to blood. A similar system is proposed in [48]. A queueing theory-based insulin mediated GLUT 4 translocation model is present in [49]. Bidirectional communication between neuroendocrine and immune systems is discussed in [50] and [51]. In [52], bacteria-to-host communication via hormones is shown. Communication by means of milk borne hormones between the mother and the suckling is proposed in [53]. Several hormonal systems act as peripherals of the nervous system, therefore, an important direction is the combination of the two systems such that both can be looked at together. This direction will be further explored Chapter 2.

1.3 Conclusion

In conclusion, we see that nanonetworks have a huge potential in terms of the current technological advances. Molecular communication is one of the most promising

techniques for nanonetwork implementation because of bio-compatibility and abundant existence in nature. However, nano-scale communication in general and molecular communication in particular have several open issue especially regarding the ICT-based modeling of the systems. These modeling techniques will be necessary to augment the current treatment and diagnostic techniques, and act as road-maps for artificial nanonetworking techniques.



Chapter 2

STUDY OF NETWORKS OF NEURONS IN THE NERVOUS SYSTEM

In this chapter of the reports, we will discuss the topic detailed for the nervous system in the abstract. These will include the work conducted on queueing model of neurons, our current work on experimental data communication through neurons as well as future work on the modeling of plasticity and networks of neurons. In the end of the chapter, we propose the construction of an artificial synapse.

2.1 ICT-based Modeling of Nervous System

Nervous nanonetwork is an ultra large-scale network of neurons gathering and transmitting information between different parts of the body. The communication between neurons takes place through chemical as well as electrical signaling referred to as neuro-spike communication [2].

As depicted in Fig. 1.5, a general neural signaling pathway consists of three basic structures; pre-synaptic neuron, post-synaptic neuron and a synaptic gap between the two neurons [2]. The information encoded in electrical impulses, passes through the axon of a pre-synaptic neuron that conveys this information by releasing neurotransmitters, special types of chemical substances, into the synapse. These neurotransmitters are then received by the post-synaptic neuron. The physical channel model of this single-input single-output (SISO) neuro-spike communication is given in [2]. The effect of inter-symbol interference on the achievable information rate of the SISO channel is studied in [54]. Since multiple pre-synaptic neurons are needed to transfer information in reality, multiple-input single-output (MISO) communication paradigm is modeled in [3]. Moreover, SISO and MISO synaptic interference channels

are investigated in [55].

Although complete neuro-spike communication channel is considered in [2] and [3], there are other studies that analyze subparts of this channel. For instance, a trade-off between information rate and energy efficient transmission is examined within the axonal channel in [56] and a realistic model for vesicle release, which involves the effects of AP width variation is proposed in [57]. Moreover, a receiver design in nervous network considering the variability in the anatomy of dendrites on post-synaptic neuron is given in [58].

Neuro-spike communication has also been analyzed in the networked environment. An optimization problem for neuronal TDMA is formulated in [59], in order to find optimum TDMA scheduling. A queueing model for nervous nanonetwork is also proposed in literature, which identifies the delay of the entire network of neurons [60].

The nervous nanonetwork is an optimum information processing system with respect to energy efficiency rather than the information capacity alone [61, 62]. The information collected from environment by intra-body nanosensor networks is encoded, processed and transmitted by nervous system to corresponding organs. Several studies have been published to estimate these encoding mechanisms, such as, considering the discrete binary and frequency coding scheme [61], interspike interval codes [63], and rate and interspike interval decoders [64]. On the other hand, in studies such as [65], stimulus response curves are fitted to input-output relationship of neuron with respect to information theoretical parameters. However, more efforts are required to reach the efficiency of neural encoding and decoding mechanism.

Moreover, information capacity of nervous nanonetwork needs to be quantified in order to implement ICT-based solutions of critical diseases of nervous system. A simple synaptic channel is proposed and analyzed from information theory perspective in [66] to derive the lower bound on capacity. In another work, the correlation among pre-synaptic terminals is suggested to improve information rate [3]. Some of the key models are discussed in more detail in the following sections to develop a motivation for further work.

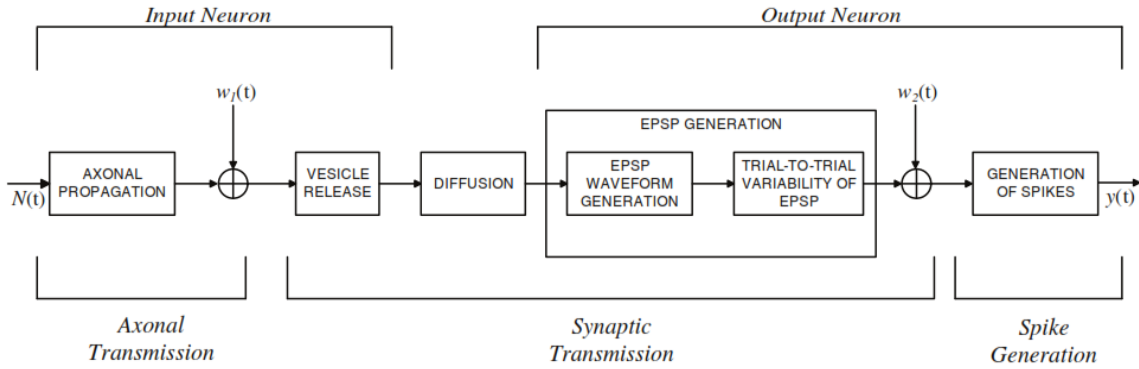


Figure 2.1: Block diagram of the neuro-spike communications. [2].

2.1.1 Physical Channel Model for a Two Neurons

In [2], the authors provide a physical channel model for neuro-spike communication between two neurons of the Cornu Ammonis (CA) of the brain. The block diagram of the spike communications between the input neuron and the output neuron at CA region is given in 2.1. Where $N(t)$, $w_1(t)$, $w_2(t)$ and $y(t)$ are spike train, axonal noise, synaptic noise, and output of the post-synaptic neuron respectively. The model covers axonal transmission and the fundamental physiological events during the synaptic transmission, which are the vesicle release of the neuron in response to a spike, diffusion of the neurotransmitters through the synaptic cleft, the Excitatory postsynaptic potentials (EPSP) generated due to the vesicle release in the postsynaptic neuron. The trial-to-trial variability of EPSP, generation of spikes at the output and the noise in the channel generated during the axonal and synaptic transmission are also included in the model. The physical channel is analyzed based on its delay and probability of error in spike detection at the output.

Based on the experiments on the CA region of the Hippocampal [8], spikes with low frequency of firing rate propagate through the axon without amplitude distortion, and nearly no failure occurs. But as frequency of firing rate increases, the amplitude of the spikes decreases logarithmically and the time delay increases. Thus, in this model axonal propagation is modeled as a modified version of a second order Butterworth

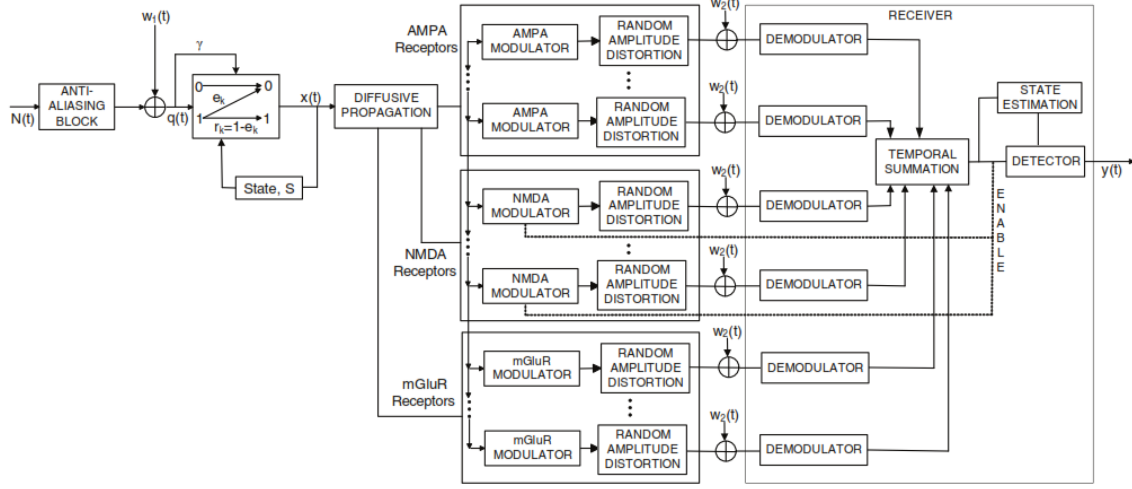


Figure 2.2: The physical channel model of a neuro-spike communications. [2].

filter. The filter is modified, because the magnitude response jumps to zero, which is the consequence of the gap time between two consecutive spikes, called refractory period. Its normalized sample transfer function is given as

$$H(j\omega) = \frac{a}{(-\omega^2 + jbw + a)}[u(j\omega) - u(j\omega - c)] \quad (2.1)$$

where $u(\cdot)$ is the unit step function and the constants a, b, c depend on the type of experiment being conducted.

Vesicle release is random in nature and it depends on initial probability of vesicle release and previous released vesicles. Hence, it is modeled as a finite-state Markov chain with input-dependent transition matrix. Once vesicles are released to the synapse, neurotransmitters will diffuse to reach post synaptic receptors. To analyze the channel propagation, it is considered that diffusion occurs in one dimension and it is modeled using the Green's function that exhibits Gaussian diffusion in response to particle release. Considering x_n as the n th vesicle containing j neurotransmitters,

$$x_n = [x_{n1}, x_{n2}, \dots, x_{nj}] \quad (2.2)$$

This can also be written as

$$x_n = \sum_{i=1}^j x_{ni} \delta(n - ni) \quad (2.3)$$

The diffusion in one dimension from a point R_i to a final point R_i^f at time t can be found by the Green's function as

$$G(R_i^f - R_i, t) = \frac{1}{4\pi t} \exp\left(-\frac{(R_i^f - R_i)^2}{4t}\right) \quad (2.4)$$

The diffusion of each particle can thus be found as

$$x_{ni}(R_i^f, t) = \int_{-\infty}^{\infty} x_{ni}(R_i, t) G(R_i^f - R_i, t) dR_i \quad (2.5)$$

For generation of EPSP waveform, because the response of metabotropic receptors occurs in a longer time than others and the metabotropic receptors are rare in the Hippocampal cortical neurons, Amino-3-Hydroxy-5-Methyl-4-Isoxazole-propionate (AMPA) and N-Methyl-D-Aspartate (NMDA) receptors are considered for the model only. The response of both AMPA and NMDA receptors are alpha functions with this difference that time to peak for NMDA receptors is more than the time to peak for AMPA receptors. Trial to trial variability of EPSP is modeled as random distortion of amplitude. Considering multiple synapses between the presynaptic and postsynaptic neuron, the complete physical channel model of Fig. 2.2 is proposed in [2]

Based on the proposed channel model, the analysis reveals that an efficient communication can be obtained for low incoming signal rate to the presynaptic terminal, higher number of NMDA receptors at the output and low synaptic noise. Moreover, having smaller threshold value and higher expected value of the random variable contribute to the channel performance. Furthermore, input frequency must be lower to have a smaller time delay.

2.1.2 Multiple-Input-Single-Output (MISO) Neuron Model

In reality, multiple neurons are required to fire a single neuron and each neuron can receive thousands of synaptic inputs through its dendrites and send information to multiple neurons. In this part, we introduce an existing multiple-access model for a MISO neuron model as shown in Fig. 2.3 [3] where neuro-spike multiple-access channel between hippocampal-cortical neurons is investigated. The proposed model is depicted in Fig. 2.4.

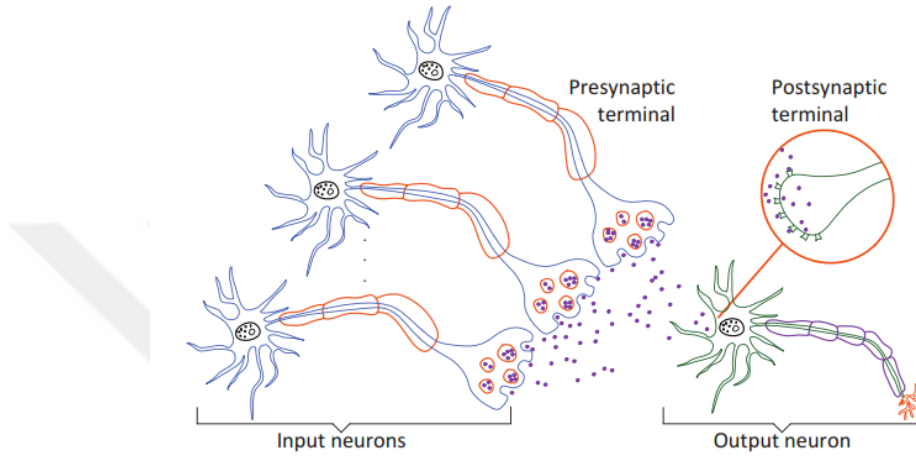


Figure 2.3: Multiple-access synaptic communication channel model. [3]

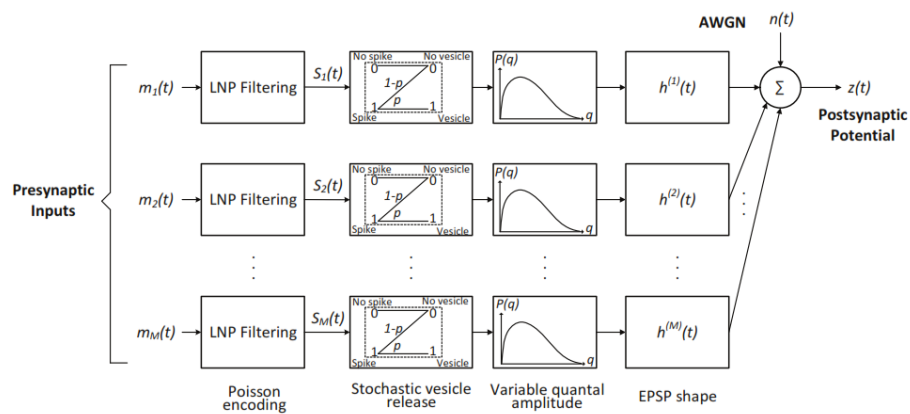


Figure 2.4: Multiple-access channel model between presynaptic and postsynaptic neuron terminals. [3]

In this model, presynaptic terminals generate their own stochastic impulse train and according to the SISO channel model of [2], each spike train passes through stochastic vesicle release and variable quantal amplitude to reach the synapse. It is considered that although there is one postsynaptic terminal, each presynaptic terminal has a distinct synapse with the postsynaptic terminal. Further, it is assumed that EPSP characteristics are identical for all channels. However, each synaptic channel is characterized by random amplitude.

The authors observed effect of multiple-access to channel on the total information rate. Moreover, they analyzed the contribution of first-order correlation on the information rate at the postsynaptic neuron terminal.

As mentioned earlier, each neuron in nervous system receives information from multiple neurons. So communication among neurons suffers from interference. It is shown that with actual adaptive synaptic weight update mechanism, performance is close to the maximum achievable rate.

2.1.3 Multiple-Input-Multiple-Output (MIMO) Synaptic Model

Fig. 2.5 shows the MIMO synaptic channel proposed in [4]. In the model, it is assumed that the inputs to the synaptic channels affect each other since the synaptic paths allow them to mix. Hence, assumptions cannot be made that the synaptic channels between the input and output terminals are parallel channels. Instead, the inputs interfere with each other to generate the channel response at the output terminals, and the performance investigation of a MIMO neuro-spike communication is thus similar to classical communication problems based on the physiological principles of neurons. Ergodic capacity is derived for the MIMO system. A lack of experimental results affects many of these studies since synaptic data is hard to come by a gap remains between the theoretical and the actual processes.

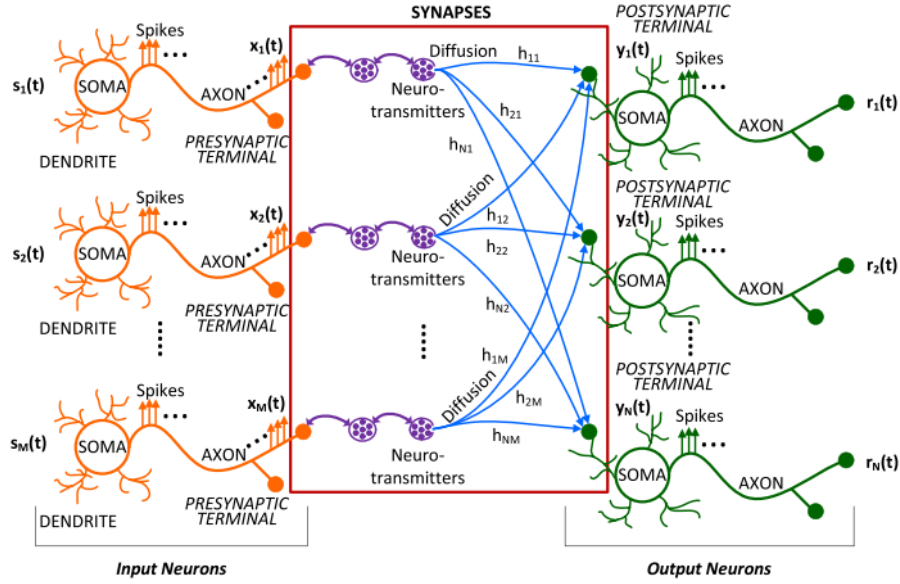


Figure 2.5: Realistic channel model for nanoscale MIMO neuro-spike communication.[4]

2.2 Networks of Neurons

As discussed earlier, the nervous system forms a big network working on top of individual neurons. While an individual neuron might just act as a transmission path, a complex network of these connections makes complex processes such as learning and memory possible. Network level behavior and modeling of neurons is very important to augment the single neuron models where the systems of switching and routing are constantly happening in the CNS on a large scale. Therefore, it is very important to develop networking models of the nervous system and study the models for network parameters such as routing, switching and see how individual neurons come together to perform complex tasks. The study of these models may give unique insight in the working of the nervous system and future artificial nanonetworks created on the basis of these studies. The motivation for such a model lies in a variety of applications from drug delivery to nervous disorder diagnosis. For example, in case of drug delivery, the nervous system can be viewed as the wired network of the body and target-specific

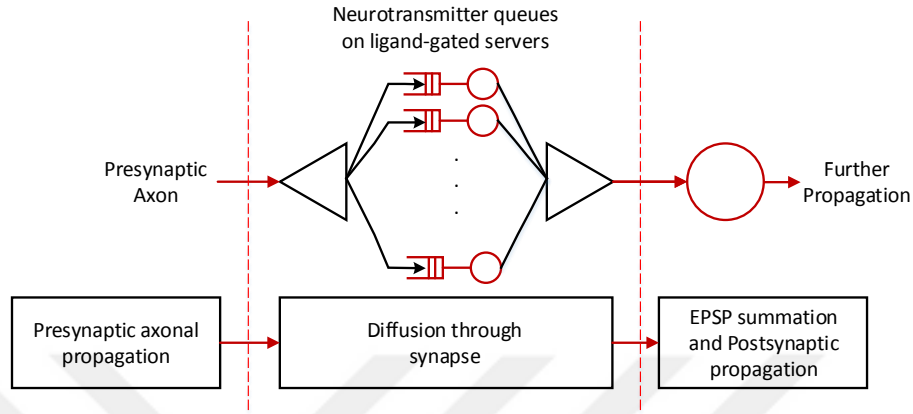


Figure 2.6: Single-input single-output neuron queue model.

drug delivery can be triggered by means of nerve impulses and neurotransmitters. Parameters such as the time required for a particular drug delivery, switching mechanisms and optimal routing through such a network as well as optimal rates of impulses to produce these scenarios can be viewed as network parameters. Additionally, the diagnostics of nervous disorders can benefit from such models.

2.3 Queueing-Theoretical Delay Model for Networks of Neurons

The advantage of using queueing analysis for biological networks lies in the fact that big networks such as central nervous system and the neural cortex can be viewed on the whole as a single network, thus making us able to study the collective behavior of these networks. In [60], we present a queueing-theoretical delay model for the nervous system. Queueing models for the network might be very essential for diagnostics of nervous disorders, where reflex latencies help in diagnosis. Practitioners can move from using basic reflexes such as patellar reflex to more complex responses to improve disease diagnostics.

To view the neuron from a queueing perspective, we need to break down the neuron model into queueing constituents namely, the servers and the buffers/queue. Fig. 2.6 illustrates a diagram of the proposed model. We assume that there are two major

constituents of the system. The first constituent of the system is a forkjoin queueing network that deals with neurotransmitter release, synaptic diffusion, dendritic propagation and EPSP generation. The number of branches of this fork-join synaptic queue is equal to the number of ligand-gated channels available for neurotransmitters attachment. To simplify the process, we assume that all the services of the synaptic communication are performed at the ligand-gated channels and each such receptor channel acts as a server. The synapse would thus act as the buffer of the system. Identification of the synapse with a queue is natural because when one neuron is trying to communicate with another, its respective neurotransmitters remain or buffer in the synapse while a previous impulse is being serviced by a neuron. In case that a single presynaptic neuron is in communication with multiple postsynaptic neurons, the synapse will still act as a queue. Each neuron will have its own fork join queue before its ligand-gated ion servers further serving their own axonal server. There is no condition on how the synaptic queue is shared between these servers as long as the number of available neurotransmitter is greater than the number of ligand-gated channels which is usually the case.

The second constituent is composed of just a single server. This server models the propagation of neuro-spikes through the postsynaptic neuron axon. The absence of a queue in the network relates to the fact that an axon can serve only one impulse at a time and does not contain any buffer where another incoming impulse can queue. It is, however, possible for a physical axon to have more than one impulse coursing through it at a time. This is because once the Na^+ absolute refractory period is over, a new impulse may be generated in the axon by a new somatic EPSP summation. This is especially true for longer axons in bigger animals where refractory periods are a fraction of the total propagation time. The reason for still keeping a single server in the axonal communication is that if we look at the system from the perspective of an incoming impulse, it will still be served by the single axonal server after being served by the synaptic queue. Any impulse that comes during the refractory period would not be entertained. The discipline of this axonal server is first come first serve

(FCFS).

The summation of the EPSP in the soma may or may not result in further spike generation. Thus, depending on a threshold it might end up in a sink or might be transmitted through the axonal body. The probability of failure p_f to go beyond the threshold voltage is usually quite low because neural communication is quite reliable but it is an essential parameter for any reliable model of communication [3]. In case there is a failure, further axonal propagation does not happen. Therefore, the second network is composed of single server queues with FCFS discipline queues in parallel.

The service time of the fork-join synaptic queue network involves several processes such as the time taken for neurotransmitter release for vesicles, time take for diffusion through the synapse and the dendritic propagation. In such scenarios, we often assume network service time distributions to be some unknown general distribution. Therefore under the analysis presented, we can say that the fork-join synaptic queue is a network of parallel M/G/1 queues.

Both the axonal server and the fork-join synaptic queue are in series, therefore, the overall mean response time of a neuron to a single impulse is the summation of the mean response times of the axonal transmission and the synaptic transmission.

$$R = R_S + R_A \quad (2.6)$$

Synaptic Transmission

The total response time of synaptic transmission R_S is a summation of the average vesicle release time R_{Ves} , average dendritic and somatic transmission time R_{DS} and the average response time for diffusion through the synapse R_N

$$R_S = R_S + R_{Ves} + R_N \quad (2.7)$$

It is difficult to model fork-join queues analytically and in fact, to date, analytical results exist for only two server systems. Usually for more than two queues, approximations for mean response time exist in case of homogeneous servers. This suits the current analysis because the servers we are considering here are homogeneous in

nature. We assume that the parallel queues of neurotransmitters in the synapse are independent and identically distributed (i.i.d.). In [67], the authors have presented an approximation of the mean response time of a set of i.i.d. fork-join queues with M/G/1 queues in parallel service as

$$R_N \approx R_1 + \sigma_1 F_N \alpha_N \quad (2.8)$$

where R_1 , σ_1 and α_N are the mean response time and standard deviation, respectively, for one M/G/1 queue with no forkjoin properties. F_N is a constant which scales according to the service time distribution of the servers and α_N is a scaling factor that helps scale simulation results according to the results of a physical experiment.

The probability density function (PDF) of neurotransmitter diffusion from a presynaptic to a postsynaptic neuron is given by the Green's function as

$$G(x, t) = \frac{1}{\sqrt{4\pi t}} \exp\left[-\frac{(x - x')^2}{4t}\right] \quad (2.9)$$

under the normalization condition

$$\int_{-\infty}^{\infty} G(x, t) = 1 \quad (2.10)$$

If we consider a unit distance between the two neurons through the synapse, the PDF of neurotransmitter diffusion given in 2.9 can give us a service time distribution of neurotransmitter service for unit distance case. It should also be mentioned here that a normalization constant according to 2.10 should be multiplied so that the PDF does not exceed the unit area under the curve condition. Thus, the distribution of service time for a single M/G/1 neurotransmitter service is given as

$$G(t) = \frac{1}{\sqrt{4\pi t}} \exp\left[\frac{-1}{4t}\right] \exp\left[\frac{-t}{2.57\pi}\right] \quad (2.11)$$

where the normalization constant is approximated numerically.

Since the failure rate of neurotransmitters generating an AP from their individual EPSP is quiet low in case of a valid stimulation to the presynaptic neuron, there must be a high degree of synchronization in their diffusion though the synapse. In other words, this means that the mean response times of each of the fork-join queues is

quite similar. Thereby, the standard deviation term from 2.8 can be neglected for successful transmission case resulting in

$$R_1 \approx R_N = \int_0^\infty tG(t)dt \quad (2.12)$$

Axonal Transmission

To determine the response time of an axon, we should first look at what constitutes the service of an axon. After an impulse enters an axon, as long as it is propagating within an axon, we consider it under service. Additionally, after an impulse leaves, the axon cannot have any further impulses pass through it until the Na^+ ion channels return from their inactivation state. Thus, the total response time of an axon is a summation of the Na^+ ion inactivation period and the response time for propagation through the axon body.

$$R_A = R_{Na} + R_{AP} \quad (2.13)$$

where R_A is the average response time of an axon, R_{Na} is the average time it takes for Na^+ ion channels to become reactivated and R_{AP} is the average response time for impulse propagation through the axon.

Considering that the axons of a particular neuron type have an average length of $E[L_A]$, and conduction speed s , R_{AP} is given as

$$R_{AP} = \frac{E[L_A]}{s} \quad (2.14)$$

Using the cable theory of axonal transmission, the conduction speed of any axon is given as

$$s = \sqrt{\frac{\partial^2 V(x, t)}{\partial t^2} \frac{d}{4\rho I_m}} \quad (2.15)$$

where $V(x, t)$ is the voltage of the action potential, d is the diameter of the axon, ρ resistivity of the axoplasm and I_m is the membrane current density. For an unmyelinated neuron 2.15 can be modified as

$$s_{nm} = \sqrt{\frac{d}{8\rho c^2 r^*}} \quad (2.16)$$

where c is the membrane capacitance per unit area and r^* is the resistance per unit area. Therefore, 2.14 can be modified as

$$R_A = R_{Na} + \frac{E[L_A]}{\sqrt{\frac{\partial^2 V(x,t)}{\partial t^2} \frac{d}{4\rho I_m}}} \quad (2.17)$$

$$R_{A-nm} = R_{Na} + \frac{E[L_A]}{\sqrt{\frac{d}{8\rho c^2 r^*}}} \quad (2.18)$$

Thus, the overall response time is given by 2.6 can be modified as

$$R = R_{ves} + \int_0^\infty tG(t)dt + R_{Na} + \frac{E[L_A]}{\sqrt{\frac{\partial^2 V(x,t)}{\partial t^2} \frac{d}{4\rho I_m}}} \quad (2.19)$$

This shows that the mean response time of a neuron depends on the diffusion characteristics, ion channel inactivation periods and the physical parameters of the neuronal structure. These characteristics match the results of previous neural communication models and experimental results.

Case study of a nervous circuit: the knee-jerk reflex arc

One of the key advantages of using a queueing theory is the ease with which it can look at complex networks to find the average delays faced by packets. Under this motivation, we study a simple reflex action of the human body, known as the knee-jerk reflex, which is often used by physicians to test nervous diseases of the lower spinal cord. The knee-jerk reflex is an example of a monosynaptic nervous circuit in the body. This reflex is a reflex of proprioception which helps maintain posture and balance, allowing to keep ones balance with little effort or conscious thought. The process of eliciting a knee-jerk reflex is detailed in Fig. 2.7.

Striking the ligament of the knee with a reflex hammer just below the patella stretches the muscle spindle in the quadriceps muscle. Muscle spindles are sensory receptors present inside a muscle which detect changes in the length of the muscle. These spindles then produce a signal which travels back to the spinal cord via a sensory neuron and synapses in the L4 spinal segment.

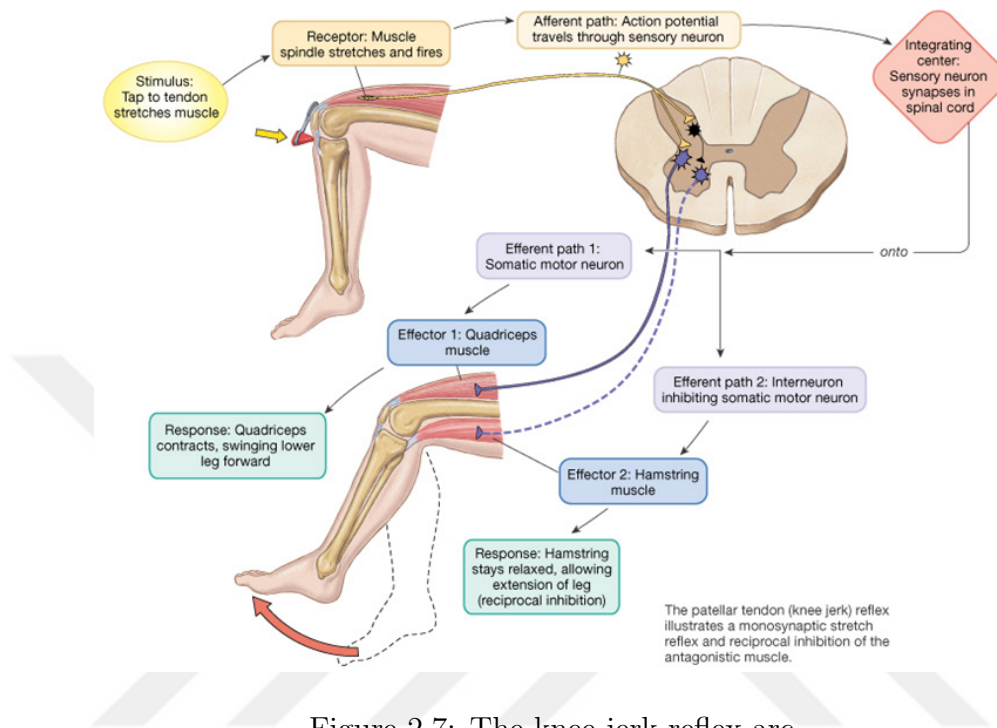


Figure 2.7: The knee-jerk reflex arc.

The sensory neuron synapses with two motor neurons. A femoral motor neuron then conducts the impulse back to the quadriceps femoris muscle, triggering its contraction. Through an interneuron, a tibial motor neuron carries the signal to the hamstring muscle to relax it. This contraction, coordinated with the relaxation of the hamstring muscle causes the leg to kick.

Since the knee-jerk response is a very simple and efficient way to detect nervous diseases, it has been the center of various scientific studies for more than a hundred years. Many studies are present where the latencies from the time of stimulation to the time of response have been conducted. In [68], the knee-jerk reflex latencies are found and further divided as reflex latency, which is the time taken by the nervous components of the reflex arc, and the reflex motor time which is the time required by the muscles to move after receiving signals from motor neuron. The results of the study for a set of normal people are presented in Table 2.1 and some key axonal parameters are provided in Table 2.2.

Table 2.1: Knee-jerk reflex parameters.

Parameter	Value	Standard Deviation
Number of test subjects	10	-
Mean Age	23.3 years	3.1 years
Total reflex time	77.7ms	16ms
Reflex latency	23.4ms	1.3ms
Reflex motor time	54.2ms	16.3ms

Table 2.2: Axon parameters for standard adult (*age > 20 years*)

Type	Myelin	Diameter(mm)	Propagation Speed (m/s)
Tibial motor neurons	Yes	13-20	41-53
Femoral motor neurons	Yes	13-20	35-50
Sural sensory neurons	Yes	13-20	40-58
Preganglionic neurons	Yes	1-5	3-15
Postganglionic neurons	No	0.2-1.5	0.5-2

Based on our discussion of the knee-jerk reflex, we present the queueing model for the circuit in Fig. 2.8. Application of an external impulse to the knee produces sensations in the muscle spindle. The muscle spindles synapse with a sensory neuron making the first fork-join synaptic queue. This queue feeds the resultant summed EPSP signals to the Sensory neuron server. The sensory neuron is further connected with two fork-join queues, one for the motor neuron of quadriceps and the other with an inhibitor interneuron. We can consider two independent fork-join queues for each of the nervous connections because the amount of neurotransmitter released is much higher than number of gates for either of the connections. The interneuron is further connected with the hamstring muscle through another fork-join queue. Each of these neurons has an axonal server as well. We now move on to simulate our knee-jerk reflex

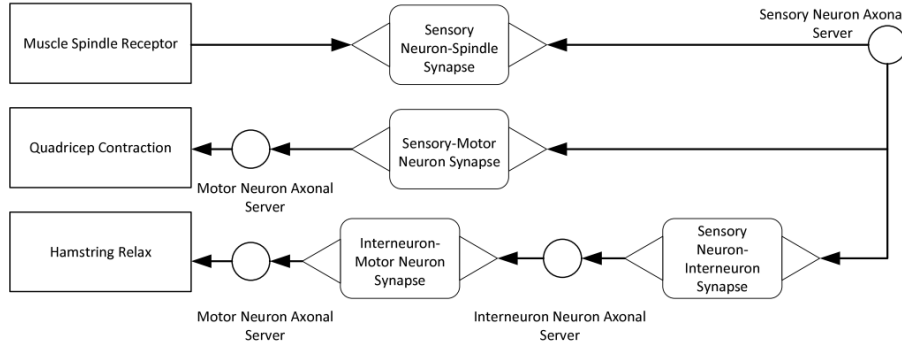


Figure 2.8: Queueing model of the knee-jerk reflex arc.

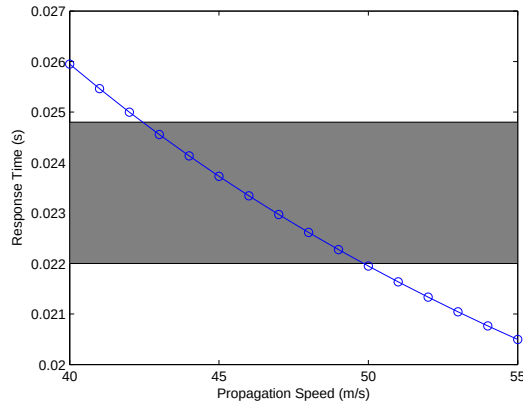


Figure 2.9: Simulation of knee-jerk reflex over a range of propagation speeds.

arc model. The average axonal lengths of the sensory and motor neurons for the knee-jerk reflex have not been estimated in any study, therefore, we have to approximate these. Since the axons of the sensory and the motor neurons run the length of the thigh to the base of the spinal cord near the hip, a good approximation would be to use the average distance between the hip and knee joint. According to US Center for Disease Control (CDC) statistics, the average upper leg length for males and females aged 2029 is approximately 40cm. Using 2.19 we simulate for the entire reflex arc over the conduction speeds for the range of values provided in Table 2.2 and show the results in Fig. 2.9. The shaded region of the graph represents the range of values for reflex latency as described by Table 2.1. We see that a significant number of

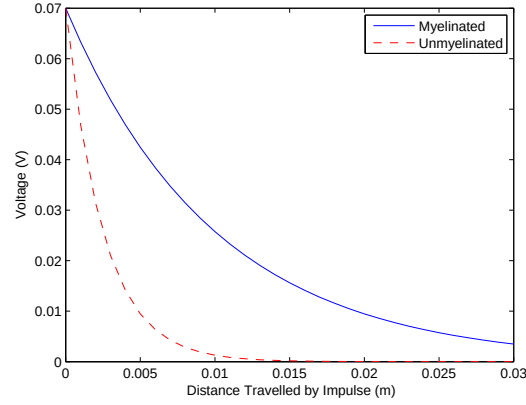


Figure 2.10: Voltage decay along a neuron; myelinated vs. unmyelinated neurons.

result data points fall within this range. This shows that application of the current model to the knee-jerk reflex nervous circuit gives similar results to those found by experimental studies. The results can be further improved if axonal lengths and the latencies are measured from the same population or studies such as [68] are conducted on more subjects.

Voltage decay along myelinated and unmyelinated neurons

The Human nervous system contains both myelinated and unmyelinated neurons. Myelinated neurons are typically found in sensory and motor neurons while non-myelinated neurons are found in the brain and spinal cord.

Voltage decay along the axonal length is plotted in Fig. 2.10. We assume that the AP was at a maximum potential of 70mV at the start of two neurons, one myelinated and one unmyelinated. For a myelinated neuron, the value membrane resistance is about 10 – 2000 times that of an unmyelinated neuron depending on the thickness of the sheath of myelin outside the axon. For our current result, we use a value of 20 times. The resulting figure shows that over a similar length, the signal of an unmyelinated neuron decays more quickly. This was expected because as membrane resistance increases, the length constant λ increases and the signal can travel longer

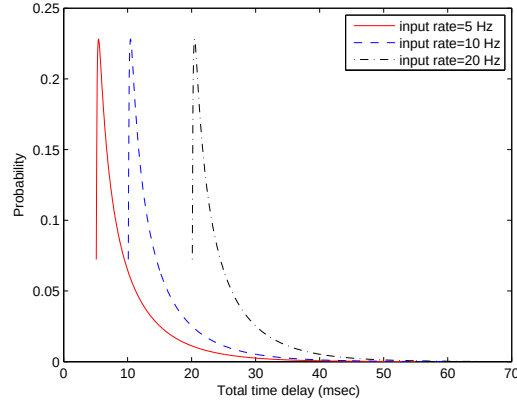


Figure 2.11: Effect of frequency on response time distribution.

distances. An interesting fact here is that as the membrane resistance increases for myelinated neurons, the membrane capacitance decreases. This keeps the value of the time constant τ , given by $\tau = rc$, nearly unchanged. The result agrees with experimental results that larger animals, needing slower AP decay, usually have myelinated neurons which operate at higher propagation speeds. Similarly, sensory and motor neurons have myelination because they have to carry impulses over longer distances as compared to impulses carried over much smaller distances carried by the unmyelinated neurons in the human brain.

Effect of frequency

The frequency of the input to a neuron is directly proportional to the internal resistance and results in a decrease in the length constant. The results of Fig. 2.11 are generated using the PDF provided by for a unit diffusion distance. Our results agree with the results of [2] which state that as the frequency of the input signal to a neuron increases, the response time increases as well.

2.4 Neuroplasticity and Networks of Neurons

Neuroplasticity is wide field that describes how lasting changes to the brain happen throughout an animal's life course. This term gained prominence in the latter half of the 20th century, when new research showed many aspects of the brain remain changeable even into adulthood [69]. This was contrary to the previous scientific consensus that the brain develops during a critical period in early childhood, then remains relatively unchangeable.

Neuroplastic change can occur at small scales, such as physical changes to individual neurons, or at whole-brain scales, such as cortical remapping in response to injury; however cortical remapping only occurs during a certain time period for example if a child were injured and it resulted in brain damage then cortical remapping would most likely occur, however, if an adult was injured and it resulted in brain damage, then cortical remapping would not occur since the brain has made the majority of its connections [70]. Even then some changes might happen over the course of life and previously used brain regions might go on to control other tasks. Behavior, environmental stimuli, thought, and emotions may also cause neuroplastic change, which has significant implications for healthy development, learning, memory, and recovery from brain damage. Neuroplasticity has two important types namely synaptic plasticity and non-synaptic plasticity.

Synaptic Plasticity

Synaptic plasticity is the ability of synapses to strengthen or weaken over time, in response to increases or decreases in their activity. Since memories are postulated to be represented by vastly interconnected networks of synapses in the brain, synaptic plasticity is one of the important neurochemical foundations of learning and memory. Two key molecular mechanisms for synaptic plasticity involve the NMDA and AMPA glutamate receptors. Opening of NMDA channels, which relate to the level of cellular depolarization, leads to a rise in post-synaptic Ca^{2+} concentration and this has been linked to long-term potentiation (LTP). Strong depolarization of the post-synaptic

cell completely displaces the Mg^{2+} ions that block NMDA ion channels and allows Ca^{2+} ions to enter a cell causing LTP, while weaker depolarization only partially displaces the Mg^{2+} ions, resulting in less Ca^{2+} entering the post-synaptic neuron and lower intracellular Ca^{2+} concentrations in turn activating protein phosphatases and inducing long-term depression (LTD) [71].

These activated protein kinases serve to phosphorylate post-synaptic excitatory receptors (e.g. AMPA receptors), improving cation conduction, and thereby potentiating the synapse. Also, these signals recruit additional receptors into the post-synaptic membrane, stimulating the production of a modified receptor type, thereby facilitating an influx of calcium. This in turn increases post-synaptic excitation by a given pre-synaptic stimulus. This process can be reversed via the activity of protein phosphatases, which act to dephosphorylate these cation channels [72].

The second mechanism depends on a second messenger cascade regulating gene transcription and changes in the levels of key proteins at synapses such as CaMKII and PKAII. CaMKII and PKAII are protein kinases that regulate by the Ca^{2+} and K^{+} . These are involved in many signaling cascades and are thought to be an important mediator of learning and memory. Activation of the second messenger pathway leads to increased levels of CaMKII and PKAII within the dendritic spine. These protein kinases have been linked to growth in dendritic spine volume and LTP processes such as the addition of AMPA receptors to the plasma membrane and phosphorylation of ion channels for enhanced permeability [73]. Localization or compartmentalization of activated proteins occurs in the presence of their given stimulus which creates local effects in the dendritic spine. Calcium influx from NMDA receptors is necessary for the activation of CaMKII. This activation is localized to spines with focal stimulation and is inactivated before spreading to adjacent spines or the shaft, indicating an important mechanism of LTP in that particular changes in protein activation can be localized or compartmentalized to enhance the responsivity of single dendritic spines. Individual dendritic spines are capable of forming unique responses to presynaptic cells. This second mechanism can be triggered by protein phosphorylation but takes

longer and lasts longer, providing the mechanism for long-lasting memory storage [74]. The duration of the LTP can be regulated by breakdown of these second messengers. Phosphodiesterase, for example, breaks down the secondary messenger cAMP, which has been implicated in increased AMPA receptor synthesis in the post-synaptic neuron. Long-lasting changes in the efficacy of synaptic connections between two neurons can involve the making and breaking of synaptic contacts. These processes are regulated by a certain group of genes. Further discussion on this topic is currently beyond the scope of this research.

The number of ion channels on the post-synaptic membrane affects the strength of the synapse [75]. Research suggests that the density of receptors on post-synaptic membranes changes, affecting the neurons excitability in response to stimuli. In a dynamic process that is maintained in equilibrium, NMDA and AMPA receptors are added to the membrane by exocytosis and removed by endocytosis [76, 77]. These processes, and by extension the number of receptors on the membrane, can be altered by synaptic activity [76, 77]. Experiments have shown that AMPA receptors are delivered to the synapse through vesicular membrane fusion with the postsynaptic membrane via the protein kinase CaMKII, which is activated by the influx of calcium through NMDA receptors. CaMKII also improves AMPA ionic conductance through phosphorylation.

Non-Synaptic Plasticity

Nonsynaptic plasticity is a form of neuroplasticity that involves modification of ion channel function in the axon, dendrites, and cell body that results in specific changes in the integration of EPSP and inhibitory postsynaptic potentials (IPSPs). Nonsynaptic plasticity is a modification of the intrinsic excitability of the neuron. It interacts with synaptic plasticity, but it is considered a separate entity from synaptic plasticity. Intrinsic modification of the electrical properties of neurons plays a role in many aspects of plasticity from homeostatic plasticity to learning and memory itself. Nonsynaptic plasticity affects synaptic integration, subthreshold propagation,

spike generation, and other fundamental mechanisms of neurons at the cellular level. These individual neuronal alterations can result in changes in higher brain function, especially learning and memory. However, as an emerging field in neuroscience, much of the knowledge about nonsynaptic plasticity is uncertain and still requires further investigation to better define its role in brain function and behavior.

In comparison, nonsynaptic plasticity is a less well known and somewhat new and ongoing field of research in neuroscience. It is manifested through changes in the characteristics of nonsynaptic structures such as the soma, the axon, or the dendrites. Nonsynaptic plasticity can have short-term or long-term effects. One way these changes occur is through modification of voltage-gated channels in the dendrites and axon, which changes the interpretation of excitatory or inhibitory potentials propagated to the cell. For example, axonal nonsynaptic plasticity can be observed when an action potential fails to reach the presynaptic terminal due to low conduction or buildup of ions. Nonsynaptic and synaptic plasticity have been shown to work concurrently in a variety of ways to produce stimulating effects in the neuron. This includes spike generation, a product of nonsynaptic regulation of potassium and other presynaptic ion channels, which increase the response of the excitatory postsynaptic potential through neurotransmitter release and augmentation of the action potential. Thus modeling of nonsynaptic plasticity will involve making the models of axons and soma flexible enough to incorporate and change according to inputs as opposed to current static models for these entities.

Neuroplasticity and Hebbian Theory

One of the fundamental principles of how neuroplasticity functions is linked to the concept of synaptic pruning, the idea that individual connections within the brain are constantly being removed or recreated, largely dependent upon how they are used. This concept is captured in the aphorism which states that neurons that fire together, wire together or neurons that fire apart, wire apart summarizing Hebbian theory [78]. If there are two nearby neurons that often produce an impulse simultaneously, their

cortical maps may become one. This idea also works in the opposite way, i.e. that neurons which do not regularly produce simultaneous impulses will form different maps.

Cortical organization, especially for the sensory systems, is often described in terms of maps. For example, sensory information from the foot, projects to one cortical site and the projections from the hand, target another site. As the result of this the cortical representation of the body resembles a map. Many researchers have observed in their experiments that if the cortical map is deprived of its input it will become activated at a later time in response to other, usually adjacent inputs. In [79], the authors mapped the cortex of an owl monkey hands before and after amputation of the third digit. Before amputation, there were five distinct areas, one corresponding to each digit of the experimental hand. Sixty-two days following amputation of the third digit, the area in the cortical map formerly occupied by that digit had been invaded by the previously adjacent second and fourth digit zones. The areas representing digit one and five are not located directly beside the area representing digit three, so these regions remained, for the most part, unchanged following amputation. This study demonstrates that only those regions bordering a certain area will invade it to alter the cortical map. In [80, 81], the authors went on to use cortical implants to study the evolution of plasticity in both the somatosensory and auditory systems. Both systems show similar changes with respect to behavior. When a stimulus is cognitively associated with reinforcement, its cortical representation is strengthened and enlarged. In some cases, cortical representations can increase two to threefold in 1-2 days at the time at which a new sensory motor behavior is first acquired, and changes are largely finished within at most a few weeks.

Since the adult brain is not entirely hard-wired with fixed neuronal circuits, there are many instances of cortical and subcortical rewiring of neuronal circuits in response to training as well as in response to injury. There is solid evidence that birth of brain cells occurs in the adult, mammalian brain and such changes can persist well into old age. The evidence for neurogenesis is mainly restricted to the hippocampus

and olfactory bulb, but current research has revealed that other parts of the brain, including the cerebellum, may be involved as well. The specific details of how this process occurs at the molecular and macro levels are topics of active neuroscience research. The manner in which experience can influence the synaptic organization of the brain is also the basis for a number of theories of brain function. Since these processes occur as a result of plasticity in networks of neurons, a networking study of these changing topologies is very necessary. Therefore, in this thesis first we will model plasticity in a single neuron where it will be organization of neurotransmitter receptors in response to specific impulses. Then we will continue to model this effect in a network of neurons and thereby approach the concept of memory in neurons.

2.5 Open Issues in Networks of Neurons

In this chapter, we discussed the neuron queueing model and discussed how neuroplasticity and networks of neurons work together to form the basics of memory and learning. For a detailed study of networks of neurons, it is imperative to learn the principles of plasticity in the nervous system. Key open issues related to networks of neurons that will be investigated as part of this thesis include the following:

- Improvement of neuron queueing model based on further physiological works and improving synaptic models.
- Improvement of existing nervous models to include models of synaptic plasticity.
- Investigation and modeling of nonsynaptic plasticity.
- Simulation based analysis of Medium access and routing mechanisms for networks of neurons for memory organization.

Chapter 3

EXPERIMENTAL DATA COMMUNICATION THROUGH NERVOUS SYSTEM

3.1 Introduction

With steady miniaturization of scientific applications over the last century, we are at the dawn of nanotechnology. Many interesting applications of nanotechnology such as nano-switches, actuators, intelligent drug delivery systems, nanoscale sensing and bio-hybrid systems are being proposed and realized [9]. Most nanotechnology applications require many nanomachines to operate in synchrony among themselves or with other devices. Therefore, nanomachines have to communicate with other nanomachines and macroscale devices. However, nanomachines face a number of constraints in terms of power, processing, limited memory resources and simple networking that limit their capabilities.

Many paradigms such as nanomechanical, acoustic, electromagnetic, chemical and molecular communications are suggested for nanocommunications, however, the most promising one is molecular communication [48]. Molecules are used to encode, transmit and receive information in this form of communication. The key reason for selection of molecular communication over other schemes of nanocommunication is that in this system we can take advantage of molecular communication constructs already available in nature that have been optimized over millions of years of evolution [1]. Natural molecular communication networks are all around us and within us, therefore, they have the added advantage of being bio-compatible and have lower power requirements.

Huge nanonetworks in large living entities such as human beings communicate

among themselves and on macroscopic scale based on molecular communications on cellular and intercellular level [82]. Understanding the dynamics of molecular communication in living entities not only guides the development of new systems, it also gives new directions to study ourselves and figure out solutions to the problems our bodies face. Many diseases are actually failures of certain types molecular communications within our cells. Hence, by exploring these systems, we take an important step forward.

The nervous system is one of the most important and complex nanonetwork of an animal body. This system is based on nerve cells known as *neurons*. Neurons are connected to sensory elements in animal bodies from where they carry information about external stimuli in the form of impulses known as action potentials or nerve spikes, to the central nervous system (CNS) that is also composed of neurons. The CNS neurons take decisions based on the received impulses and the resulting decisions are then communicated by a separate chain of neurons to muscle cells for further action [83]. Individual neurons or their small networks operate on small information capacities, however, larger networks based on these units such as the human brain are able to achieve huge memories and are able to perform very complex decision making tasks.

Many emerging medical applications such as body area networks (BAN) and brain-machine interfaces are targeting wearable electronics that communicate directly with the nervous system [84]. Targeted drug delivery is another important research field where a lot of research is being conducted [85]. Since the nervous system acts a wired network that is spread throughout the body, it can be used to reliably control various devices connected to the body. Bio-computers based on actual neurons are also envisioned as alternative computing paradigm [86]. Additionally, diseases due to failures of nanocommunication in the nervous system such as Alzheimer's disease, Schizophrenia and Parkinson's disease are key challenges in the current age [82]. Therefore, nanocommunication in the nervous system is the target for a number of important research efforts.

Several classical models for neurons, their networks and neural coding based on circuit models of neurons are provided in [83]. A number of contrasting theories exist on the information content of nervous signals and neural codes [83, 87]. However, none of these theories and models are based on information and theoretical (ICT) modeling of the nervous system and, therefore, cannot be applied to ICT behavior of the nervous system. This type of analysis and modeling has only been targeted more recently. A physical channel model for neuro-spike communications in neurons is provided in [2]. Communication-theoretical analyses and models of synaptic multiple-access channel for hippocampal-cortical neurons and cortical neurons are presented in [3, 4]. In [60], a queueing-theoretical delay model for the nervous system is described. Despite several models, to date there has been no attempt to communicate data through the nervous system experimentally. In this regard, we are working towards a unique test bench of nervous system that achieves data communication through living neurons.

The first step in the development of an experimental data communication system based on the nervous system is the selection of an appropriate neuron type and testing animal such that the results of the experiments can be extrapolated to other animals. The selected animal should suit the experimental setup and be easily available as well. The vast majority of animals including humans are bilaterians. A fundamental bilaterian body form consists of a tube containing major nerves in a hollow cavity running from the brain to throughout the body with each side of the body being identically served by nerves that shoot from the central nerves. Worms are amongst the simplest bilaterian animals making them a sample bilaterian case [88]. Nerve cords runs along the length of the worm body and merges at the mouth and the tail. The central nerve cords are connected by other nerves like the rungs of a ladder throughout the body. Two ganglial nervous tissues at the head function similar to a simple brain. Worm neurons are covered by sheaths similar to myelin coating of the CNS in higher animals like humans [88]. Although this covering of neurons is not as good as the myelin sheath of human neurons, it offers significant speed advantages

to worm axonal communication compared to other invertebrates such as cockroaches and brings their model system near to that of vertebrates. The size of worm neurons is also large as compared to other invertebrates making them easy to clamp into by means of external probes. Apart from the similar structure, the ease of availability of common worm types and their easy upkeep makes them the ideal animals for nervous system experiments.

Earthworms are common segmented worms found in soil. They are one of the most developed species among invertebrates in terms of the nervous system. The nervous system of an earthworm comprises of a ventral nerve cord and peripheral nerves. Since their neurons are not truly myelinated, to increase conduction velocity, they have large diameter axons creating giant fibers [88]. Common earthworms have one medial and two lateral giant fibers all being bilateral in conduction [88]. However, when stimulation occurs from the anterior end of the worm only the medial giant fiber responds and only lateral fibers respond to stimulation from the posterior end [89]. Therefore, the median giant fiber channel and the lateral giant fibers channels have two different set of inputs and can be separated spatially by applying inputs to different parts of the earthworm body. The two kinds of fibers are also distinguishable by their conduction velocities. Lateral fibers conduct action potentials slower than the medial fiber [89].

Extracellular recording of earthworm spike signals by means of external electrodes is easy because of the thin body wall and muscles [90]. The nervous system of earthworms is sensitive to both mechanical and electrical stimuli. Responses can be recorded from intact, awake worms in which the muscle potentials are recorded beside action potentials due to body movements. However, the peripheral sensory cells and muscles can be inactivated with proper level of anesthesia providing records only from the giant fibers that are still active thus isolating the nervous system of the animal for experiments with external data signals [90].

In this study, we implement a data communication system between macroscopic instruments and a nervous channel. We first model the nervous channel, which is

composed of a network of neurons in the nerve cord of an earthworm, and identify the required parameters for communication system. Data translated as an amplitude and a frequency is then modulated onto the network of neurons to be received on the other end of the nerve cord. The strategies for modulation and symbol assignment are also investigated. The unique contributions of this work can be summarized as follows

- To the best of our knowledge, this is the first demonstration of a controlled information transfer through a nervous system based channel.
- This is the first example of a macro-to-nano scale communication using biological nanonetworks in multicellular animals and among the very few examples of experimental molecular communications. This study will impact not only as a test bench for molecular and nervous communication, it also has the potential to act as a test bench for artificial neurons and synapses.
- The results and analysis of our experiments provide a method to model networks of neurons, identify optimum parameters of input on such networks such as the frequencies, amplitudes and the modulation schemes. This can pave way for the design, testing and implementation of future nanonetwork applications based on the nervous system.
- We show that the information modulated onto the nervous channel is stored in the number of neural spikes per pulse of input signal.

The remainder of this chapter is organized as follows. In Section 2, we present the experimental setup and methodology to model and implement the nervous system based nanocommunication channel. In Section 3, we discuss the results of our experiments. Finally, we conclude the work in Section 4.

3.2 Methods

Our experiments can be divided into two categories on the basis of experimental setups. The first setup is used to evaluate the parameters for the operation of the physical channel, calculate the channel propagation delay and modeling of the network of neurons in the nervous channel. A second similar but more elaborate setup is for data communication through the nervous channel. The experiments for modeling and data communication are conducted for the medial giant fibers in most cases by stimulating an earthworm at its anterior end and receiving the signals at the posterior end. Similar results are expected for the lateral giant fibers, however, due to their slower conduction speeds and larger activation potentials, lower data rates and higher propagation delays are expected. For each setup, a complete reading from one earthworm is considered if an earthworm gives values for all the test points. Any cases where an experiment could not be completed is not considered in the results and analysis afterwards. For cogent results, readings from the same earthworm are not used twice in the analysis. After giving a brief discussion about the earthworms used in these experiments, we take a look at each of the setups.

3.2.1 Animal Selection and Important Measures

The experiments for this work started with two earthworm species, namely, *Lumbricus Terrestris* and *Eisenia veneta*. Both of these earthworms are very common and can be found easily in moist soils. *Eisenia Veneta* or compost worms are physically bigger and their body wall is stripy as compared to *Lumbricus Terrestris* or the common earthworm [91]. The identification and discrimination of the two species are based on physical features described in [91] and the operation of CNS for both the worm species is as described in the previous section. Common earthworms exhibit a better signal quality and lower noise levels than compost worms due to a thinner muscle layer, however, they frequently broke during stimulation experiments and rendered the data of incomplete sessions useless. Therefore, the experiments reported here are taken from our results of the compost worm. Mature worms were chosen intentionally

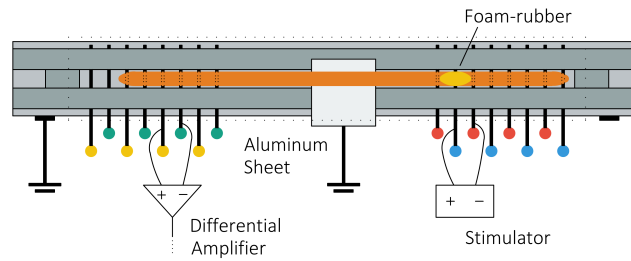


Figure 3.1: Experimental setup for the modeling of a network of neurons.

for the experiments because of their larger sizes where the worm maturity is recognized by the presence of clitellum, which is non-segmented region toward the anterior end of the worm [88]. The worms used for the experiments are kept inside a moist soil drum in our lab that matches their natural habitat. Worms on which an experiment is conducted are subsequently separated from others so that they may not be used in an experiment again inadvertently. The worms are not harmed by the experiments and all worms used in the experiments survived and recovered very quickly afterwards.

3.2.2 Experimental Setup For Modeling of Network of Neurons

To measure the characteristics of the nervous channel and to model the network of neurons present in it, we use the setup detailed in Fig. 3.1. Earthworms are anesthetized before an experiment using an alcohol solution to reduce their movement and associated muscle potential. Nervous signals are not recoverable in case the earthworm is moving because the muscle potentials being several orders of magnitude larger than action potential, make the detection of valid nervous spikes very difficult. After being washed and dried, an anesthetized worm is placed on the setup as shown in Fig. 3.1. A grounded aluminum sheet is placed underneath the earthworm to isolate the transmitter and the receiver by ensuring that the stimulator signal does not flow directly through the worm skin. Pin electrodes may be used to connect the stimulator and the differential amplifier at two points each by either putting the electrodes on the worm skin or by inserting the electrodes inside the worm body. The

second method is preferred because it keeps the worms firmly in place during the experiments and provides better nervous signals because of higher proximity to the worm nerves. An alcohol solution may be poured over the worm several times during the course of an experiment to keep it anesthetized.

At the start of each experiment, we first test the setup by applying a direct current (DC) signal of appropriate voltage at the stimulator electrodes and observe the corresponding action potentials. Since the nervous spikes of a particular earthworm are the same shape and voltage, the shape and voltage of the spikes thus measured are used as a benchmark for the rest of the experiment. If similar waveforms are generated by applying alternating current (AC) waveforms, we proceed with the experiment since it is confirmed that there is no leakage between the input and the output.

Stimulator

The stimulator should be an isolated unit of electrical stimulation that is capable of producing single or multiple electrical pulses up to 10 V amplitude with different shapes and stimulus frequencies over the range of at least $1 - 100Hz$. We used GW Instek SFG-2004 synthesized function generator that provides the required signals for our experiments.

Differential Amplifier

Having a myelin like neuron coating offers a challenge since tapping into the nervous signals in such cases requires high quality amplifiers with features such as low noise and high gain. This can be achieved by a number of extracellular amplifiers available in the market. We use AD instruments AM 17 extracellular amplifier to amplify the received signal 1000 times for our experiments. Since extracellular amplifiers are very sensitive to external noise, the setup is placed inside a Faraday's cage to cancel external noise sources. The Faraday's cage is connected to the system ground connection so that all the system stays on the same ground level. If the extracellular amplifier includes filters, they may also be used to condition the signal further. We

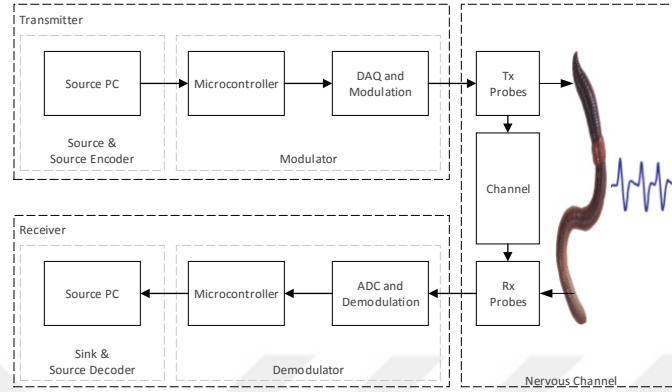


Figure 3.2: Experimental setup for data communication through the nervous channel.

used filters to cancel high frequency noise for our measurements.

Display and Signal Aquisition

The equipment for visualization and recordings of the nervous signal may consist of an oscilloscope. We use Rhode and Schwarz RTO1024 digital oscilloscope for data acquisition and storage. The data processing on the recorded signals is further achieved in MATLAB.

3.2.3 Experimental Setup For Data Communication

The channel setup of data communication is similar to the first setup. At the start of a data transmission, an anesthetized earthworm is placed on the experimental setup as shown in Fig. 3.1 with both the probes of the transmitter (at simulator position) and the detector (at the extracellular amplifier position) inserted inside its body. The block diagram for the communication system is given in Fig. 3.2. We take a look at the major blocks of this setup below.

Channel Equalization

Before the start of a transmission session, the transmitter sends a pre-known sweep of different frequency pulses with different amplitudes across the channel and the

receiver stores the number of nerve spikes generated by each pulse. This helps the receiver generate an estimate of the channel and also generate a symbol conversion formula so that the number of pulses received may be converted to a valid symbol afterwards. This step is important because there is a trial-to-trial variation in natural systems even between different organisms of the same species.

Source and Source Encoder

The information to be transmitted through the channel is prepared in the form of a random binary sequence at the transmitter personal computer (PC). Other forms of data with a suitable encoding and conversion may also be transmitted.

Modulator

The transmitter PC passes the data bits to a microcontroller that acts as a modulator by means of a serial port (RS232) connection. The directives for the modulation scheme are also sent by the PC to the microcontroller. These directives define the voltage levels and the frequencies the microcontroller has to generate for each symbol. Afterwards, the microcontroller breaks the bit sequence into bits required per symbol and transmits each symbol with the specific voltage and frequency for that symbol. Any microcontroller with a digital to analog converter may be used as the modulator. We use Arduino UNO to generate pulse of various frequencies and duty cycles. To vary the amplitude of the signal, external resistors are used.

Transmission and Detection

The pulses of each symbol are applied by the stimulator probes. The sequence of action potentials generated as a result then travels across the worm to be received by the receiver, which has to amplify the really small received signal. Although not shown as a part of Fig. 3.2, an extracellular amplifier takes the input from the receiver probes and passes them to the demodulator microcontroller after amplification.

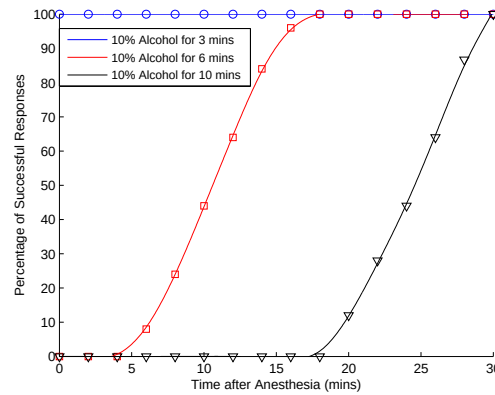


Figure 3.3: Percentage of successful responses as a result of different alcohol concentrations in anesthesia.

Demodulator

The demodulator microcontroller works as a pulse counter and counts the pulses in an interval equal to the just above half of the interval for the smallest symbol frequency on one of its analog input ports. This ensures that different frequency symbols that are sent one after the other from the transmitter are reliably received. Based on the equalization data received at the start, the number of pulses is then converted to a symbol. The demodulator is also arduino UNO based that is connected to the receiver computer through a serial port.

Source Decoder and Sink

The source decoding and analysis of the system are performed at the receiver PC that decodes the received signal and generates the estimated bits to recover the transmitted information.

3.3 Results and Discussions

3.3.1 Physical Layer Parameter Optimization

The first step in preparing the grounds for data communication is the estimation of physical layer parameters that provide an ideal scenario for spike generation. These parameters include the time for anesthesia, the voltage levels necessary to generate spike signals and the type of transmission waveform. Knowing these details and selecting the best parameters ensures that the physical layer operates effectively throughout the experiments. All the results of the section are averaged over 30 different specimens. To make the analysis more straightforward, solid lines in the results of this section are fitted over data points to show a trend.

Anesthesia Time

To easily differentiate between our desired signals, which are the nervous action potentials, the worms should be properly anesthetized. Muscle potentials are usually so large that any action potentials that are being generated while an earthworm is moving are very hard to distinguish. The anesthesia is done by placing worms in a solution of ethyl alcohol and water for a certain amount of time before the start of an experiment. Care must be taken in the anesthesia, since too much of it can cause the nerve cords to become unresponsive and correspondingly the earthworm responds to the input signals after a long while or may not give any signal at all or even die. To avoid such problems, our first experiment focuses on the appropriate amount of anesthesia required to achieve high reliability signals from a worm.

Each reading for this experiment is taken 2 *minutes* apart with electrical stimulation using a DC voltage of 3 *V* that is applied momentarily on the stimulator while keeping the distance between the stimulator probes as 1 *cm* and the percentage of successful responses is noted. The results of the experiment are shown in Fig. 3.3. We see that with a solution of 10% alcohol and 3 *minutes* of anesthesia, the earthworms are ready for the experiment right away. With longer times in anesthesia

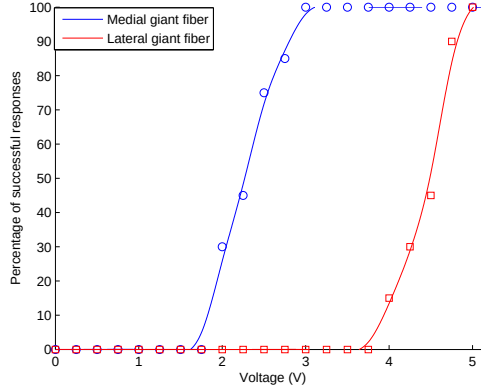


Figure 3.4: Percentage of successful responses for different transmission signal voltages.

or higher concentrations of alcohol, the worms take considerable time to be ready for the experiment. Higher concentrations of alcohol are not recommended since the process damages the worms irreversibly. Since the process of anesthesia depends on the health, age and type of worms, a small trial-to-trial variation may be expected in the results.

Transmission Signal Level

The optimal voltage level of the transmission signal to create action potentials is a very important parameter for the physical layer to operate properly. For this experiment, we sweep a DC voltage in the range of 0–5 V with a resolution of 0.25 V and averaged our results for both the medial and the lateral giant fibers to generate Fig. 3.4. With the distance between the stimulating electrodes fixed at 0.5 cm, momentary pulses (0.5 seconds and 10 seconds apart) of DC voltage were given. The results shows that action potentials are easily generated on a smaller voltage levels in the medial giant fibers which are thicker than the lateral giant fibers. For the medial giant fibers, a voltage above 3 V is necessary for reliable transmission of the signal whereas for the lateral giant fibers, a voltage of at least 4.5 V is necessary.

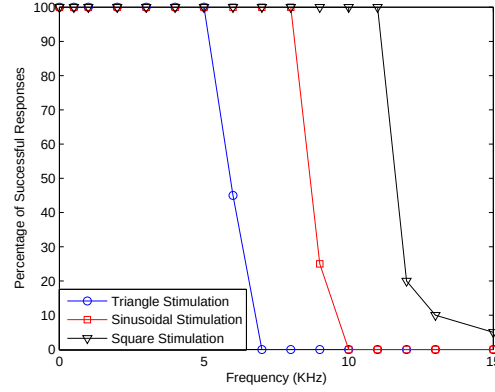


Figure 3.5: Effect of transmission waveform on percentage of successful responses.

Transmission Waveform

The transmission signal waveform is very essential to reliably transmit a signal over the nervous channel. In this set of experiments, three different types of waveforms, namely, the square wave, the sinusoidal wave and the triangular wave, are applied to the medial giant fiber at different frequencies and the percentage of successful responses from the receiver were noted. The peak-to-peak voltage values were set at 3 V, which is usually enough to generate action potentials. Each waveform was applied to the anterior end of the worm for 0.5 *seconds* and the corresponding action potential generation was noted. The results are shown in Figure 3.5.

The results show that at lower frequencies, all types of transmission waveforms perform very reliably, however, once the frequency increases, the square wave performs the best, followed by the sinusoidal and the triangular wave. The reason for this lies in the fact that amongst the waves applied, square wave has the highest root mean square (RMS) voltage and the effective current through the worm body to create transmission pulses decreases with frequency. Therefore, square pulse based transmission signals provide the best results on the physical layer. Since the waveform analysis is independent of the type of neuron, a similar result is expected for the lateral giant nerves and they are therefore omitted from this analysis.

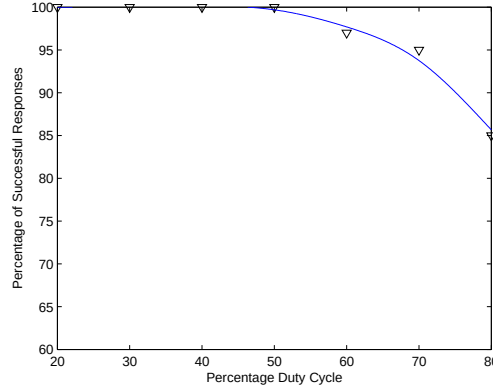


Figure 3.6: Effect of pulse width on successful response generation.

Transmission Pulse Width

Now that the square pulse is found as the best waveform for generating the transmission pulses, the pulse width becomes an important parameter. The minimum pulse width that can generate an impulse can be used to reduce the power requirements of the system while ensuring minimum damage to the worms. In this regard, by using signal frequency of 10 Hz , signals of different pulse widths were applied to the transmission end of the medial giant fiber and the resultant percentage successful responses are shown in Fig. 3.6.

The results show that even with a pulse width of 20%, very reliable communication can be achieved. Using a pulse width of 20% with a square pulse reduces the power requirements of the transmitter so this value is selected for subsequent experiments.

3.3.2 Channel Propagation Delay

The overall channel propagation delay of a network of neurons depends on the axonal conduction velocity and diffusive delays along the length of the network [60]. Since diffusive delays cannot be singled out with the knowledge of the exact physiology, they are considered as a part of the channel conduction velocity. The overall channel

propagation delay for a network of linearly connected neurons by [60] is

$$R = \frac{L}{v_{avg}}, \quad (3.1)$$

where L is the length of the channel and v_{avg} is the measured conduction velocity. Our results show that the average conduction velocity for the medial giant fibers is 13.196 m/s and 5.48 m/s for the lateral giant fibers. Our results agree with previous studies and show that the lateral medial fibers have higher propagation delays [89].

3.3.3 Modeling of Network of Neurons

In order for a communication system to be established, we should know how the channel translates an input to an output. Certainly, a neural coding mechanism is converting the information we provide to the channel to spikes that then travel through the network. Therefore, the most important question is how information is modulated onto the channel as a result of different inputs. A number of neural coding theories try to answer this question by describing information content of nervous signals by various metrics [83] but the exact mechanism for our scenario can only be understood once a system model is developed.

To model the network of neurons present in the nervous channel of an earthworm, we consider it as a black box and analyze the inputs and outputs of the system to find a relationship between them. Black box modeling can be efficiently conducted for the analysis of networks of neurons because neurons respond to a limited range of inputs that may easily be replicated under laboratory conditions. To achieve the black box modeling, we swept signals with various frequencies and amplitudes in an orderly manner by applying each frequency and amplitude signal for at least 10 seconds as the input of the nervous channel and recorded the outputs from the extracellular amplifier. A square pulse with a duty cycle of 20% was used to this end. The ordering of the tests was shuffled to prevent any biases in the data. Once the data was collected, we tested for several schemes of neural coding to evaluate which among them best described the information we passed onto the channel. Schemes such as average spike rate, instantaneous spike rates, spike width analysis and interspike delay were tested,

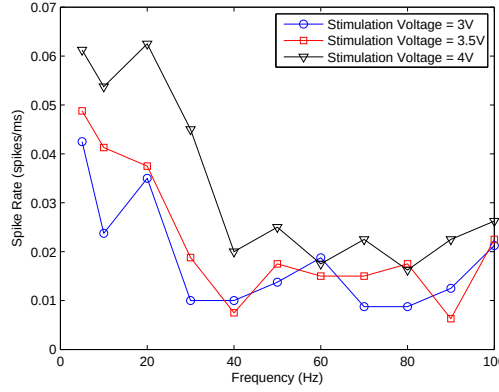
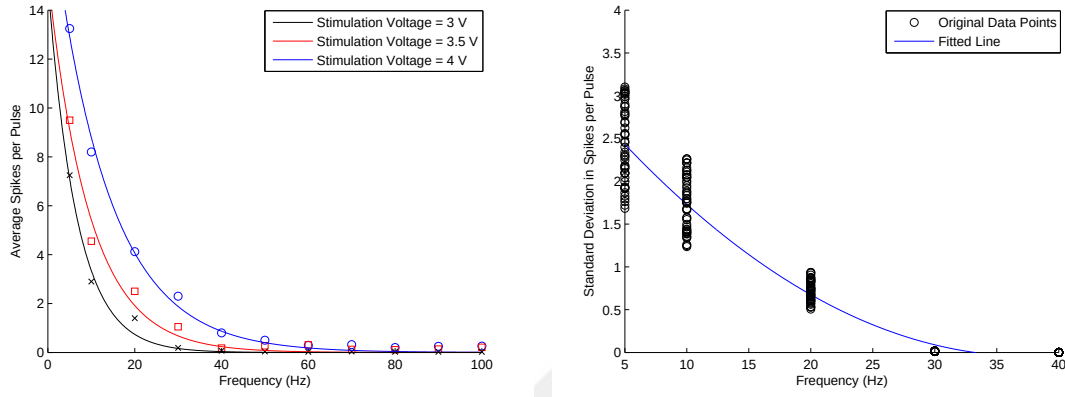


Figure 3.7: Effect of frequency on average spike rate.

however, none of these metrics truly represented the information we modulated. For instance, Fig. 3.7 shows effects of changing input voltage and frequency on the average spike rate. We see that the spike rate does decrease in general as the frequency increases, however, the relationship is not straight forward and at multiple points, the data does not describe the unique input we gave. The same holds for an instantaneous spike rate case. Further, our analysis reveals that there is no information in the interspike delays that are randomly distributed following a log-normal distribution with a mean of 1 *ms* and a stand deviation of 0.1 *ms*. The spike width also stays nearly unchanged over a specimen having mean values of around 2 *ms*. We then tested spikes per input pulse as a measure of the information content. The results of these experiments for 50 worms are summarized in Fig. 3.8. We see that the resultant data describes the information we gave to the channel. Fig. 3.8 (a) shows how the number of spikes per input pulse reduce as the frequency increases. Using an exponential function we fitted the data points by minimizing the sum of squared errors (SSE) as

$$Y(x) = ae^{-Xb}, \quad (3.2)$$

where Y represents the number of spikes per pulse generated by a frequency X . a and b are constants that change from worm to worm. Our experiments show that



(a) Average number of spikes per pulse. (b) Standard deviation of number of spikes per pulse.

Figure 3.8: Analysis for number of spikes per input pulse.

data from all the earthworms can be fitted by this function for different values of the constants a and b . The rationale behind suggestion of an equalization block in the data communication is to communicate the values of these parameters to the receiver for a particular worm. The number of spikes per pulse varies from its mean value randomly with a standard deviation term σ shown in Fig. 3.8 (b). We can observe that for lower frequencies, where the number of spikes per pulses may be higher, σ is higher. σ is modeled by minimizing SSE for a second degree polynomial function as

$$\sigma(x) = p_1 x^2 + p_2 x + p_3, \quad (3.3)$$

where the average values of the constants are $p_1 = 2.34 \times 10^{-3}$, $p_2 = -1.756 \times 10^{-1}$ and $p_3 = 3.252$. Thus, the network of neurons for earthworm medial giant can be modeled by using (1) and (2). This black box modeling approach for can be used with any network of neurons available in an experiment.

3.3.4 Data Communication through Network of Neurons

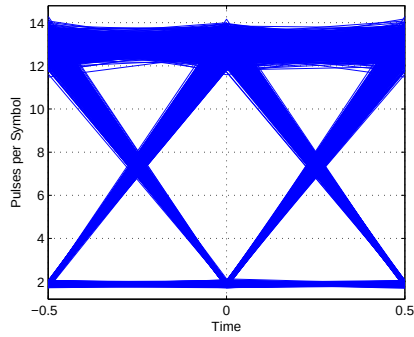
From the results of Fig. 3.8 (a), we see that a number of modulation schemes can be employed for data communication through the nervous system of an earthworm. We

may employ on-off keying (OOK), amplitude shift keying (ASK) or frequency shift keying (FSK). For instance, for OOK we can use any frequency in the response range of the network with an amplitude above $3V$ and select the absence of a signal as the off case. Similarly, for ASK we may select a frequency of input signal and send different amplitudes as different symbols. This is especially true for lower frequencies where the difference of pulses between two amplitudes is higher. Similarly, keeping a constant amplitude, different frequencies may be used to generate the symbols for FSK. ASK operates at lower data rates than FSK because it performs better at lower frequencies and might not be feasible at higher frequencies. Taking FSK as a sample case of modulation, we perform our analysis in the rest of the section accordingly.

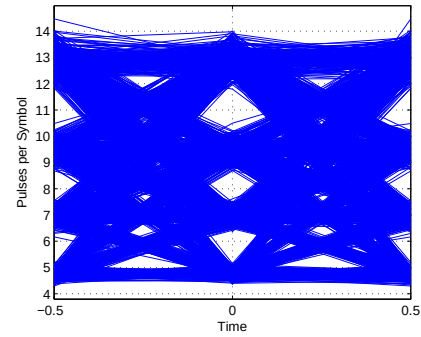
Simulation Model

In order to analyze, compare and improve the performance of a communication system, simulations before the actual implementation are standard practice. Thus, we designed a simulation of the system that generates 10000 bit randomly that are then modulated using (3.2) and (3.3) with log-normally distributed interspike delay and a fixed spike width of 2 ms as described in the modeling section.

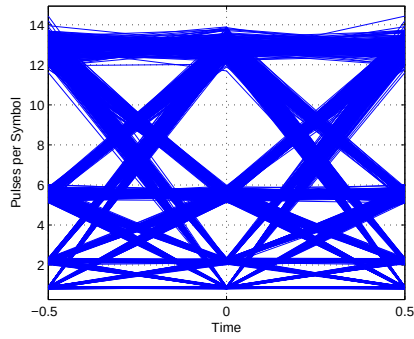
To analyze the results of the simulations, we use eye diagrams. Several eye diagrams generated by the simulation are shown in Fig. 3.9. These diagrams show how the symbols having different frequencies are translated into number of spikes by the channel. Since the change is governed by a random process, the number of spikes changes around its average value. The opening of an eye in this case shows the margin a receiver has for decisions. Smaller eye opening thus stand for a higher bit error rate (BER) and vice versa. Fig. 3.9 (a) shows the results for a modulation scheme using two FSK symbols. We see that the eye opening is very wide and the margins for decision at the receiver are very big. Having a decision boundary at around 7 pulses, the receiver can easily distinguish between the symbols. We also see that since at lower frequencies the spike number per symbol varies much more, the spikes generated by the 5 Hz symbol are spread over a large area. In Fig. 3.9 (b), (c) and (d), we see



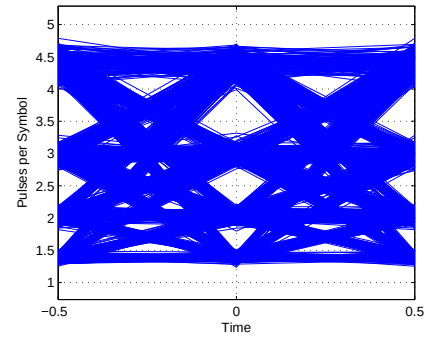
(a) Symbol frequencies of 5, 30 Hz.



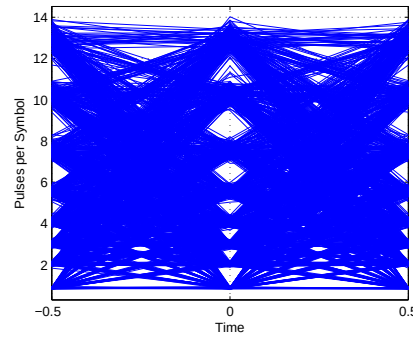
(b) Symbol frequencies of 5, 9, 13, 18 Hz.



(c) Symbol frequencies of 5, 16, 28, 40 Hz.



(d) Symbol frequencies of 19, 24, 29, 34 Hz.



(e) Symbol frequencies of 14, 16, 18, 22, 26, 30, 35, 40 Hz.

Figure 3.9: Eye diagrams for FSK with different number of symbols.

FSK assignments for four symbols. The biggest eye opening are seen in Fig. 3.9 (c) where the symbols are distributed as far as the bandwidth permits. This points to the lowest BER for this case. In Fig. 3.9 (b) and (d), the eye openings are much smaller suggesting a higher BER with better comparative performance for Fig. 3.9 (d) with slightly bigger eye openings. This is because of the fact that spike variability reduces at higher frequencies. Finally, in Fig. 3.9 (e), we present the case for an eight symbol FSK arrangement. The frequency assignment is done in such a way that the separation at lower frequencies is larger than at higher frequencies. The BER for this case is expected to be higher than all other cases.

Experimental Results

After completing the simulation modeling, we performed experiments of data communication where 1000 randomly generated bits of data were passed through an earthworm. We ensured that all the symbols of each symbol set were equally represented in the transmission data. The results are averaged over 50 earthworms and shown in Table 3.1. We see that, as expected, when we use symbol sets that have higher frequencies (case 3), the BERs are lower as compared to symbol sets with lower frequencies (case 2). This is because of smaller variability in spike number at higher frequencies as shown by Fig. 3.8 (b). Lowest BERs are possible when the symbol set is distributed such that the symbols are at a maximum distance from each other (case 1). However, we also notice that the best data rates are achieved when symbol sets of higher frequencies were used (case 3). A result for FSK with eight symbols is also shown (case 4). We see that the data rate is further increased in this case, however, the BER is the highest. Thus, the results of our implementation of data communication through the nervous system agree with our simulations.

Further improvements may be achieved by taking advantage of source encoding schemes that assign symbols with lower probability to lower frequencies that have more variability and reduce the over all data rate. Such improvements may increase the data rate and improve the BER performance further. Since the worms nerves

Table 3.1: BER and data rates for various symbol sets.

(#)	(Symbols)	(BER)	(Data Rate (bps))
1	[5, 16, 28, 40] Hz	2×10^{-5}	24.7514
2	[5, 9, 13, 18] Hz	4.6×10^{-4}	18.0346
3	[19, 24, 29, 34] Hz	7.2×10^{-4}	52.6646
4	[14, 16, 18, 22, 26, 30, 35, 40] Hz	6.8×10^{-3}	66.6102

run in two directions with different points of stimulation, a study on bi-directional communication through the nervous system can be an interesting future work.

The data rates for a nervous system based communication system may seem nominal in comparison with contemporary communication system, however, we should note that for animals like humans, higher information processing data rates exist due to naturally existing diversity. Organs like the human brain have billions of neurons working in parallel and such system therefore have much higher information capacity than many contemporary systems although their fundamental units have similar performance.

3.4 Conclusion

In this work, we realised the first data transfer through the nervous system as a communication channel on nanoscale. In our system, macroscale devices are practically connected by nanocommunications existing in the nervous system. Physical layer parameters, channel propagation delay and the system model are investigated. The use of eye diagrams for system evaluation in simulations is also evaluated and our results for the physical communication agree with our simulation results.

We believe that the experimental results obtained in this work are very encouraging for researchers to develop novel applications based on information exchange with the nervous system. The experimental setup is quite easy-to-implement as an educational tool, a model biological communication system and may be used as a

test bench for future applications in the nervous system. The study of bi-directional channels and effects on blocks such as source coding are left as future studies.



Chapter 4

ICT-BASED MODELING OF INSULIN-GLUCOSE SYSTEM

Among the different communication paradigms suggested for the realization of nanonetworks in physical environment such as nanomechanical, acoustic, electromagnetic, chemical and molecular communications, the most promising one is molecular communication [48]. Molecules are used in this type of communication to encode, transmit and receive information. These systems are based on bio-nanomachines that can be found all around and within us [1]. Naturally existing bio-nanomachines form complex molecular nanonetworks to communicate among themselves and with external environment at intra and inter-cellular level. Understanding this molecular signaling among living cells from information and communication theoretical (ICT) perspective provides novel nanonetworking paradigms as well as a deep insight into the ICT-based fundamentals of biological systems [1]. Several ICT models exist in literature for key molecular communication systems of the human body such as the nervous system [2, 82] and the gap junction communication between cardiomyocytes [45] among others.

The endocrine system is a set of glands that produce hormones to control important functions of the body such as metabolism, growth, development, reproduction and emotions, among other functions. Molecular concentrations of various chemicals are sensed in tissues that are then communicated to the endocrine system. Hormones required to regulate these chemicals are then added by the endocrine system into the blood stream to achieve homeostasis. The propagation of hormones from the transmitter to a receiver depends on blood circulation as well as their Brownian motion of the hormones [48]. Since a generic biochemical model that fits all hormones is

not possible, several works on various individual hormones are present in literature. However, due to its sheer importance in metabolism, a huge body of literature focuses on the insulin-glucose endocrine system. The key function of this system is the regulation of sugar or glucose in the blood stream that is digested from food. Insulin is the hormone that regulates blood glucose by assisting its uptake by the body cells. Specialized cells in pancreas known as the '*beta cells*' produce insulin. In cases when the pancreas does not produce enough insulin or when the body cannot effectively use the insulin it produces, a set of conditions known as diabetes occur. According to World Health Organization (WHO) statistics, 9% of adult population in the world is affected by diabetes as of 2014[92]. With such high prevalence rates of diabetes, the study of insulin-glucose systems is very important.

Taking a brief overview of the literature, we see that some mathematical models for the biochemical processes involved in the insulin-glucose system are presented in [93]. In [94, 95], the authors discuss meal simulation model of insulin-glucose system and the prevalent intercellular communication. Insulin communication within the pancreatic islets is further explored by [96]. Gap junction communication for insulin release within pancreatic islets is presented in [97]. In [49], insulin mediated GLUT 4 translocation modeling is provided based on queueing theory. Apart from physiology and models, applications such as insulin management for diabetic patients by means of subcutaneous insulin pumps are already realized [98, 99]. Currently, insulin is released in these pumps at preprogrammed times or according to the local glucose concentration of the body area surrounding the pump. Since most diagnostic, management and treatment applications of this system are based on molecular communication, it is essential to model this communication so that the efficiency of the entire system may increase. For instance, future applications may communicate directly with pancreatic islets and release the exact insulin concentration in the body through a subcutaneous pump, thus, improving the pump efficiency [100]. Additionally, the insulin-glucose system is interconnected with other networks of the body such as the nervous system [101]. A complete ICT model of body requires the ICT

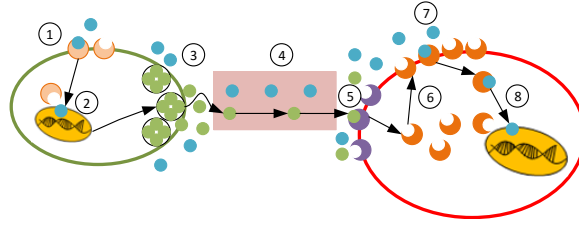


Figure 4.1: Insulin-glucose system involving the following key steps: 1. Uptake of glucose by a beta cell, 2. Metabolism of glucose by the beta cell, 3. Insulin vesicle exocytosis starts as a result of glucose metabolism, 4. Insulin and glucose diffuse through the circulatory system to be transported throughout the body, 5. Insulin binding to insulin receptors at a muscle cell, 6. Glucose transporters are pushed to the cell membrane because of insulin binding, 7. Glucose binding with glucose transporters and 8. Glucose transport and metabolism by the muscle cell.

models of each its constituent nanonetworks.

To date there is no work on ICT-based modeling of the insulin-glucose system. Therefore, in this thesis, the molecular communication between a beta cell and muscle cell connected by means of a capillary is modeled and analyzed based on the information theory. Expressions for channel capacity and propagation delay are derived and the effects of insulin resistance on these communication parameters is discussed. The model presented in here may help in investigation, diagnosis and treatment of diabetes as well as modeling the connections of various nanonetworks such as the nervous system and the insulin-glucose system. Furthermore, the model can be utilized in the design and engineering of similar communication systems for nanodevices.

4.1 Insulin-Glucose System

In the insulin-glucose system, communication between a pancreatic beta cell and a muscle cell occurs by means of the circulatory system between them. Key steps of this process are illustrated in Fig. 4.1. Insulin and glucose molecules diffuse through the capillary membrane into the blood stream and are then circulated in

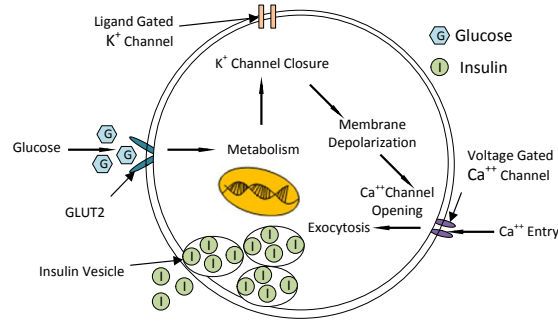


Figure 4.2: Insulin secretion as a result of glucose uptake by a beta cell.

the body along with other types of blood cells. With the increase in blood sugar concentration after a meal, more and more glucose molecules attach with glucose transporters present on the membranes of beta cells. Insulin is then released into the blood stream, which in turn, help the muscle cell uptake of glucose from the blood and lowers the concentration of blood glucose. The reduced glucose concentration ultimately reduces the insulin secretion and the body ends up in a state of homeostasis. In the remaining part of this section, we discuss these processes further to get a detailed picture.

4.1.1 Insulin Secretion

Various types of cellular channels are present on the cell membrane of a beta cell such as specialized glucose transporters called ‘*GLUT2*’ that act as glucose sensors, adenosine triphosphate (ATP)-sensitive potassium ion (K^+) channels and the voltage-gated calcium ion (Ca^{++}) channels [102]. The K^+ channels are normally open whereas the GLUT2 and the Ca^{++} channels are normally closed. The open K^+ channels cause an outflow of potassium from the cell and bring the membrane potential below with respect to the outside at about $-60mV$ [102].

The complete process of insulin secretion from a beta cell is detailed in Fig. 4.2. A small number of glucose insulin molecules are always circulating in the body even in the absence of any meal, however, when food is digested, a higher number of

glucose molecules start to circulate. This increases the probability of glucose molecule reception ($P(B_G)$) by the beta cells through their GLUT2 glucose sensors [102]. Once received, the glucose is transported inside the cell to be metabolized, which increases the concentration of ATP. The increase in ATP concentration causes the ligand-gated K^+ channels to shut down and the membrane potential starts to increase since K^+ ions are not flowing out anymore. This, in turn, causes the Ca^{++} channels to open and a flow of Ca^{++} ions to occur within the cell. Ca^{++} ions cause vesicles containing insulin to move toward the cell membrane where insulin is released by exocytosis [103]. All the processes described above are governed by chemical kinetics. The intracellular reaction dynamics are generally well understood and modeled with the values of most kinetic constants already known [104].

Some of the insulin produced by the processes discussed above is constantly being removed from the system to avoid an overload. Insulin clearance is an integral component of insulin metabolism and the liver is the primary site of insulin clearance where approximately 80% of endogenous insulin is removed while the remainder is cleared by the kidneys and muscles [105]. The overall change in concentration of insulin in the blood stream is a sum of average clearance rate of insulin from the blood, average rate of insulin release by a beta cell independent of the presence of glucose and the net increase of rate due to the presence of glucose [106]. The change of insulin concentration with respect to time can be described by

$$\frac{dI}{dt} = -l_1 I + l_2 + l_3 G, \quad (4.1)$$

where $l_1 I$ represents the average rate of insulin clearance independent of glucose with I as the insulin concentration, l_2 is the average rate of basal insulin release by the beta cell independent of glucose and $l_3 G$ is the net increase in the rate of insulin release as a result of increase in glucose concentration G .

The probability of glucose reception at a beta cell, $P(B_G)$, is directly proportional to the glucose concentration G as well as to the number of GLUT2 transporters present on the surface of the beta cell. To simplify the modeling of $P(B_G)$, we consider that glucose concentration in the blood is limited between a maximum and

a minimum level. The minimum concentration of glucose, G_0 , can be calculated by a fasting glucose test whereas G_{max} representing the maximum concentration of glucose supported by the body calculated by a glucose tolerance test. $P(B_G)$ can be expressed as

$$P(B_G) = \frac{G - G_0}{G_{max} - G_0} \times \frac{N_{act2}}{N_{GLUT2}}, \quad (4.2)$$

where N_{act2} is the number of unbound GLUT2 transporters on the surface of a beta cell and N_{GLUT2} represents the total number of GLUT2 receptors in a beta cell. After a meal, G increases above G_0 causing an increase in $P(B_G)$. The same happens if the number of GLUT2 receptors on the surface of a beta cell increase to fulfill the metabolic requirements of the beta cell. I depends on $P(B_G)$ since the release of insulin depends on the glucose sensing by the GLUT2 transporters.

4.1.2 Circulation

Blood circulation provides the mechanism for insulin and glucose delivery to the beta and the muscle cells. This transport occurs through various arteries and veins of the body that are then connected by networks of capillaries. Capillaries are the smallest type of blood vessels ranging from 5 to 40 μm in diameter with characteristic high surface area to volume ratio that maximizes the potential for blood-tissue exchange [107]. Considering the case of two specific cells connected through a capillary, this particular method of communication is classified as wired active transport. In this type of molecular communication, information molecules move along a fixed physical channel based on an active agent. The capillary exists as a fixed connection between transmission and reception sides, and the blood circulation pressure due to cardiac contractions and relaxations acts as the active agent. There are several studies in literature regarding this type of communication [22, 24, 107]. Based on these studies, we observe that the channel capacity of a capillary channel depends on the channel dimensions as well as the type of flow in the channel.

For a beta cell connected to a muscle cell by means of a capillary, there are three important components of transport for a molecule as shown in Fig 4.1. The first is the

diffusion from beta cell to the capillary, the second is transport through the capillary and finally, there is diffusion again from the capillary to the muscle cell. Diffusion of insulin molecules is characterized by the linear diffusion equation given as

$$\frac{\partial I(x, t)}{\partial t} = D \nabla^2 I(x, t), \quad (4.3)$$

where $I(x, t)$ denotes the concentration and D denotes the diffusion coefficient. Considering a Gaussian diffusion, the solution of (4.3) in an n -dimensional space from a position x is calculated by the Green's function as

$$Gr(x, t) = (4\pi Dt)^{-n/2} e^{-\frac{(x-x')^2}{4Dt}}. \quad (4.4)$$

Equation (4.4) tells us how a point of probability density initially at point x diffuses over n -dimensional space and time existing under the condition of normalization given as

$$\int_{-\infty}^{\infty} Gr(x, t) dx = 1. \quad (4.5)$$

Inside the capillary, insulin molecules are transported by the blood flow. Reynold's number is used to help predict flow patterns in different fluid flow situations with low Reynold's numbers characterizing laminar flows and high Reynold's numbers representing turbulent flow.

$$Re = \frac{\rho v_{avg} D_H}{\nu}, \quad (4.6)$$

where Re is the Reynold's number, ρ is the fluid density, v_{avg} is the mean fluid velocity, D_H is the hydraulic diameter and ν represents the fluid viscosity. The Reynold's number for capillaries is very low (0.001) meaning that there is laminar flow within capillaries [107]. Laminar flow causes flow in parallel layers with no mixing between the layers and the flow at the vessel wall is vastly reduced due to friction with the stationary vessel wall while the velocity increases towards the centre of the vessel in a parabolic manner [108]. This kind of flow assists the ready diffusion of molecules from the capillary wall to the target cell. In addition to laminar flow, Brownian motion causes random movements of particles within the capillary based on diffusion

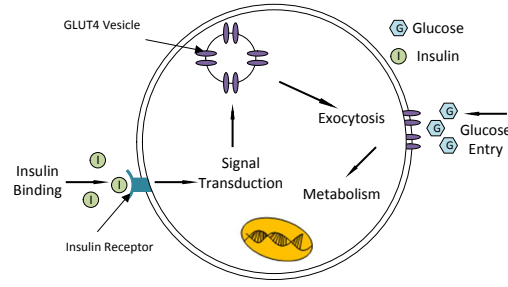


Figure 4.3: Glucose uptake by a muscle cell as a result of insulin binding.

coefficient. However, this Brownian motion within the capillary can be neglected on a microscale in comparison to the laminar flow forces [109].

Therefore, the average propagation time of an insulin molecule from a beta cell through a capillary with length L to a muscle cell is given as

$$R_C = \frac{L}{v_{avg}} + \int_0^\infty tGr_b(x, t)dt + \int_0^\infty tGr_m(x, t)dt. \quad (4.7)$$

Here $Gr_b(x, t)$ represents the normalized Green's function diffusion from the beta cell to the capillary and $Gr_m(x, t)$ represents the normalized Green's function diffusion from the capillary to the muscle cell.

4.1.3 Glucose Uptake

GLUT1 and GLUT3 glucose transporters are available throughout the cells of muscles and tissues to maintain a basal uptake of glucose [110]. However, since for the current study we are looking at the insulin mediated glucose uptake, they are neglected from Fig. 4.3, which shows the process of glucose uptake by a cell. Muscle cells begin glucose uptake when insulin molecules bind to their receptors present on the cell. Insulin receptors are protein molecules present on the cell membrane of a cell that can bind to smaller insulin ligands that act as agonist, making the receptor to function. Until this happens, 'GLUT4', the major insulin dependent glucose transporter, remains in vesicles inside the cell. As soon as the insulin binds, signals are sent to the GLUT4 vesicles and exocytosis of the vesicles starts [103]. This, in turn, allows

GLUT4 to transport glucose inside the cell that is used for metabolism. Outside the cell, due to this uptake, overall glucose concentration decreases, which then reduces the production of insulin from beta cells [106].

Some of the glucose added to the system as a result of a digestion event is constantly being removed as it is used by other parts of the body. Similarly, to maintain homeostasis, some glucose is also constantly added to the blood stream. The overall change in concentration of glucose is then a sum of average removal rate of glucose from the blood, average rate of glucose release by the body independent of a digestion event, insulin mediated glucose uptake and a digestive event that leaves the system with a big concentration of glucose [106]. This can be expressed as

$$\frac{dG}{dt} = -l_4G + l_5 - l_6I + \delta(t), \quad (4.8)$$

where l_4G represents the average rate of glucose removal independent of insulin, l_5 is the average rate of basal glucose release by the body, l_6I is the net increase in the rate of glucose uptake due to the presence of insulin and $\delta(t)$ represents the sudden event of glucose release in the circulatory system due to a digestion event at time zero. The probability of insulin reception at the muscle cell, $P(M_I)$, is directly proportional to the insulin concentration I as well as the number of insulin receptors present on the cell. Therefore, $P(M_I)$ can be written as

$$P(M_I) = \frac{I - I_0}{I_{max} - I_0} \times \frac{N_{ub}}{N_t}. \quad (4.9)$$

Here I_0 is the minimum and I_{max} is the maximum concentration of insulin. Additionally, N_{ub} is the number of unbound insulin receptors and N_r represents the total number of insulin receptors on the cell membrane of a muscle cell.

The authors in [106] use (4.1) and (4.8), to give a solution for glucose concentration G as

$$G = G_0 + Ae^{-\alpha t} \sin(\omega t) \quad (4.10)$$

where A is the undamped amplitude, $\alpha = \frac{1}{2}(l_1 + l_4)$ represents the independent clearance rates of glucose and insulin, and $\omega^2 = \omega_0^2 - \alpha^2$ with the natural response $\omega_0^2 = l_1l_4 + l_3l_6$ representing the body's response.

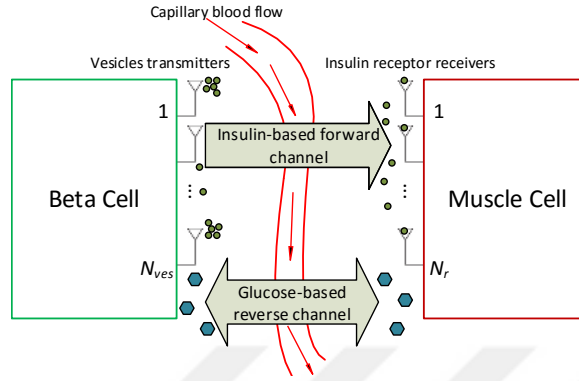


Figure 4.4: ICT model of insulin-glucose system.

4.2 Information Theoretic Model of Insulin-Glucose System

For the information theoretical model of the insulin-glucose system shown in Fig. 4.4, we consider insulin and glucose as information molecules. Although these molecules occupy the same physical channel (capillary), these molecules operate in two different channels as illustrated in Fig. 4.4. The overall system is a multiple-input-multiple-output (MIMO) system.

In the forward channel, we consider that the insulin molecules are released by the vesicle transmitter antennas into the capillary based on the feedback of glucose concentration by the GLUT2 transporters. These molecules then traverse through the blood stream, pumped by the blood circulation and diffusion, to reach the insulin receptors at the muscle cell. Each insulin molecule is considered as an independent transmission and each receiver acts as a separate antenna. The receiver as a result uptakes glucose from the blood stream by exocytosis of GLUT4.

In the feedback channel, depending on the uptake of glucose by the muscle cell, glucose concentration changes. Thus, glucose molecules constitute the information carriers of the feedback channel. The purpose of this feedback channel is to communicate the channel state information (CSI) with both the transmitter (beta cell) and the receiver (muscle cell).

4.2.1 Channel Capacity

The insulin-glucose system forms a closed-loop MIMO system, which utilizes glucose concentration as CSI. In most cases, only partial CSI is available at the transmitter because of the limitations of the feedback channel because of the small number of GLUT2 CSI sensors at the transmitter. Each vesicle is considered as an independent antenna that releases a certain number of insulin molecules. If $E(N_{I/ves})$ is the average number of insulin molecules per vesicle, the total number of insulin molecules is given as

$$N_t = N_{ves} \times E(N_{I/ves}). \quad (4.11)$$

Since each of the N_t insulin molecules constitutes an independent transmission, the maximum spatial multiplexing order for N_r insulin receptors on the muscle cell is given by

$$N_s = \min(N_t, N_r). \quad (4.12)$$

The input-output relationship of the closed-loop MIMO insulin-glucose system can be described as

$$y = Hs + n, \quad (4.13)$$

where $s = [s_1, s_2, \dots, s_{N_t}]^T$ is the $N_t \times 1$ vector of the transmitted insulin molecules from the N_{ves} separate vesicle transmitter antennas, y and n are the $N_r \times 1$ vectors of the received symbols and the noise for N_r insulin receptors on the muscle cell respectively, H is the $N_r \times N_t$ channel matrix. N_s is usually much smaller than N_t because the insulin molecules are much greater in number than the receptors. CSI communication is governed by (4.1) and (4.8), thus, controlling the number of transmitted insulin molecules based on glucose concentration, G .

Since all the transmitter antennas transmit the same information that can only bind to a single receptor at the receiver, the channel matrix H reduces to an identity matrix for the receptor antennas in use and contains zero columns for unused receptor

antennas. This is because a single insulin molecule can independently bind to only one of the receiver antennas and any other antennas in the vicinity do not receive any power from that signal. The instantaneous channel capacity of the insulin-glucose system in such cases is equal to the number of insulin-insulin receptors bindings at any time. Thus, the change of channel capacity with respect to time is equal to receptor binding rate of insulin at the receiver muscle cell governed by the kinetic equation

$$r_C = \frac{\partial C}{\partial t} = k_f I N_{ub} - k_r C, \quad (4.14)$$

where C is the instantaneous capacity of the insulin-glucose system representing the number of insulin-receptor pairs bound, k_f is the forward binding rate of insulin and k_r is the reverse binding rate with the values of k_f and k_r already provided in literature [104]. This shows that insulin concentration and receptor density at the receiver both contribute towards a higher binding rate and thus, a higher channel capacity.

At equilibrium, the rate of change of capacity is zero ($\frac{\partial C}{\partial t} = 0$) and we have the equilibrium channel capacity as

$$C_{eq} = \frac{N_{eq} I_{eq}}{k_D}, \quad (4.15)$$

where N_{eq} is the number of unbound receptors at equilibrium, I_{eq} is the equilibrium concentration of insulin and $k_D = k_r/k_f$ is disassociation equilibrium constant. k_D typically has very small values so that the capacity of the cell is very high and it can detect very small concentrations of insulin. The total number of receptors, N_r , is a sum of bound receptors, C_{eq} and the unbound receptors, N_{eq} . Therefore, (4.15) can be modified as

$$C_{eq} = \frac{(N_r - C_{eq}) I_{eq}}{k_D}, \quad (4.16)$$

which can be solved for equilibrium capacity as

$$C_{eq} = \frac{N_r I_{eq}}{k_D + I_{eq}}. \quad (4.17)$$

In order to find a maximal bound for the channel capacity of insulin-glucose system, we consider the case of negligible noise and attenuation faced by insulin molecules in the channel. Such a maximal bound may actually be achieved since insulin molecules face usually very limited attenuation in the channel because of very small degradation rates that can be neglected [105]. This results in the receivers acting as parallel channels and the maximum channel capacity equals the least number of antennas on the transmitter or the receiver or the maximum number of insulin molecules that can be contained in a capillary provided the receptors and transmitters are infinite.

$$C_{max} = \min(N_t, N_r, N_{max}), \quad (4.18)$$

where N_{max} represents the maximum number of insulin molecules that can be contained in the capillary. The diameter of capillaries is several orders of magnitude higher than that of an insulin molecule, therefore, usually $C_{max} = N_s$. As stated earlier, the number of insulin receptors is normally much less than the number of insulin molecules to facilitate the easy binding of insulin, therefore, under normal cellular operation $C_{max} = N_r$. This, however, may not always be the case because insulin pools present in the beta cell may be spent as a result of disease or persistent glucose impulses. In such cases, $C_{max} = N_t$.

4.2.2 Channel Propagation Delay

The average channel propagation delay for an insulin molecule to traverse from the beta cell to an insulin receiver on the muscle cell is a sum of the time taken for vesicle release R_{ves} , time taken for capillary propagation R_C and the time required for binding with the insulin receptors at the muscle cell R_{bind} . The average delay increases in case either of $P(B_G)$ or $P(M_I)$ reduce [104, 95]. Therefore, total average delay R is given as

$$R = \frac{1}{P(B_G) \times P(M_I)} (R_{ves} + R_C + R_{bind}). \quad (4.19)$$

The probability product, $P(B_G) \times P(M_I)$, plays the most important part in increasing the propagation delay. Physiological diseases cause either one or both of these

parameters to decrease, increasing the overall channel propagation delay.

The R_C term of (4.19) can be the delay for a single capillary or the transport through various arteries and capillaries. In the case of a larger network, the complete path from a beta cell to a muscle cell may be divided into various sections, each with a different length and average speed, and the total R_C can be expressed as a summation of all individual terms for each section. It should be noted that vesicle release and insulin binding are usually on the order of a few hundred milliseconds whereas the transport delay through a capillary and the diffusion from the capillary usually takes up several seconds [111, 112]. Therefore, for a capillary with a sufficiently long length or for a large network of blood vessels, R_{ves} and R_{bind} may be neglected.⁵

4.3 Numerical Analysis

In this section, we present a numerical analysis of the insulin-glucose model parameters namely the channel capacity, insulin transmission rate and the propagation delay to show how these parameters change with the properties of the system.

For the numerical analysis, we consider a single beta cell connected to a muscle cell through a capillary channel as shown in Fig. 4.1. The distance between the capillary and either of the cells is considered as unit to simplify the diffusion analysis. Average speed of blood flow in a capillary and the value of disassociation constant are considered at physiological values of $v_{avg} = 1.14mm/sec$ and $k_D = 3nm$, respectively [112, 113].

4.3.1 Channel Capacity

Using (4.17), we can evaluate equilibrium channel capacity, C_{eq} , for various insulin concentrations. I_{eq} values between 4–12mU/mL are selected from human physiology and results are generated for different number of insulin receptors (N_r). The various values of N_r used for the simulation are provided for mammalian cell physiology in [114].

The result of the simulation is shown in Fig. 4.5. We can observe that C_{eq}

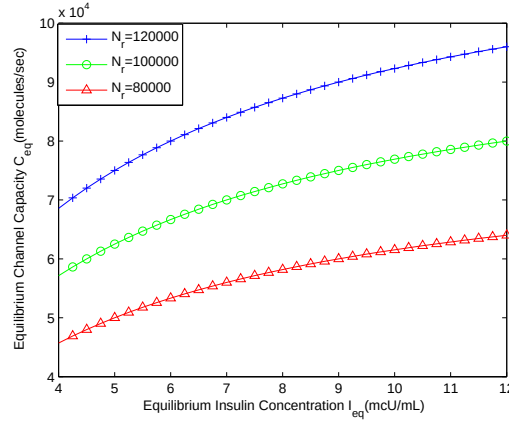


Figure 4.5: Effect of insulin concentration on equilibrium channel capacity for different number of insulin receptors.

increases with an increase in the insulin concentration, however, this increase slows down at higher concentrations. The result also shows that C_{eq} is proportional to the total number of insulin receptors present on the cell surface but the maximum limit of capacity from (4.17) for each of the cases may be achieved only at very high concentrations of insulin. Such concentrations are beyond the levels of normal insulin concentration in the body. This means that under normal conditions, the insulin-glucose operates much below its theoretical maximal capacity limits, thus, ensuring a reliable information transfer across the system. This behavior is seen in many biological molecular communication systems, which operate much below the maximal capacity bounds to ensure reliable information transfer [1].

4.3.2 Insulin Transmission Rate

In order to observe the impact of capacity requirements on the insulin concentration, which acts as a measure of insulin transmission rate, Fig. 4.6 was generated using (4.15). Firstly, the result shows that N_{eq} is inversely proportional to the I_{eq} required to achieve a certain capacity. Secondly, we observe that as capacity increases, the insulin concentration required to achieve it increases in an unbounded manner. This

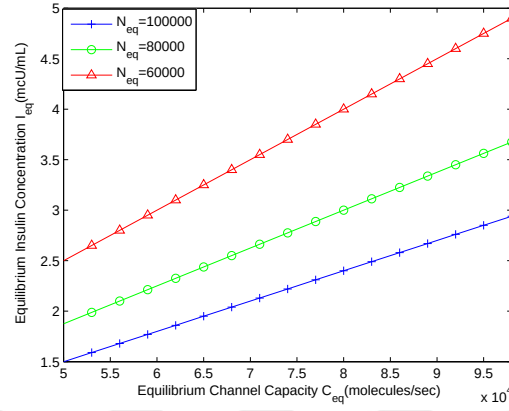


Figure 4.6: Effect of channel capacity on insulin concentration for different number of equilibrium receptors.

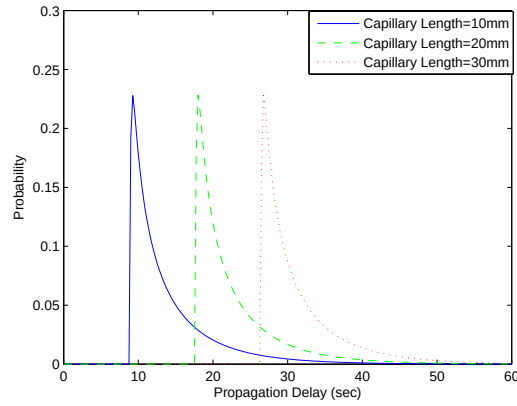


Figure 4.7: PDF of average propagation delay through capillaries of various lengths.

is not be possible for the pancreatic beta cell because the insulin pools in a beta cell can provide insulin up to a certain level before depleting the reserves and any regeneration may need further time. This also drives the beta cell in an overdrive mode to produce more and more insulin, reducing the overall cell life.

4.3.3 Channel Propagation Delay

Considering 1-dimensional diffusion, the probability density functions (PDF) of R_C , the average propagation delay through capillaries of various lengths, is shown in Fig. 4.7 using (4.4), (4.5) and (4.7). R_C is proportional to capillary lengths, however, it can be observed that delays due to diffusion are also quite significant. This means that diffusive delays cannot be neglected for analysis of capillary based transport systems.

The effects of probability product $P(B_G) \times P(M_I)$ on the total propagation delay is shown in Fig. 4.8. The time for vesicle release (R_{ves}) and the time for insulin binding (R_{bind}) are considered as $100msec$ based on [111]. The result shows that as the probability product increases, the propagation delay decreases. In the absence of a glucose impulse, $P(B_G)$ reduces and thus the product term increases the overall average propagation delay. Similarly, if insulin concentration is reduced, $P(M_I)$ increases the average propagation delay.

4.4 Insulin Resistance and Insulin-glucose System Parameters

As of 2014, among the population that suffers from diabetes, 90% cases suffer from Type 2 Diabetes that is characterized by insulin resistance [92]. Insulin resistance is a physiological condition where the body cells fail to respond to insulin and reduce the uptake of glucose leading to a high blood sugar level [115]. Beta cells subsequently increase their production of insulin, further contributing to a high blood insulin level. Since the body cells cannot uptake glucose for metabolism, the energy production of the body decreases [115]. We will take a look at the impact of insulin resistance on various insulin-glucose system ICT parameters in this section.

4.4.1 Effects on Channel Capacity

Due to insulin resistance, a significant number of insulin receptors do not activate in response to insulin binding [116]. This causes a failure on the receiver and slows down the glucose uptake. From the perspective of the model presented, this means

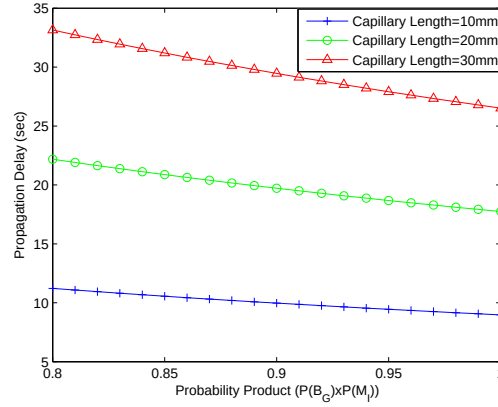


Figure 4.8: Effect of probability product on average propagation delay.

a reduction in the number of effective receiver antennas. From (4.17) and (4.18), we see that this effects the channel capacity since the bound of channel capacity for the insulin-glucose system is the maximum number of receptor antennas present. Therefore, the channel capacity of the insulin-glucose system decrease as a result of insulin resistance. This inference is also possible from Fig. 4.5 where we see that the number of insulin receivers is directly proportional to channel capacity.

4.4.2 Effects on Insulin Transmission Rate

Since the effective number of insulin receivers decreases as a result of insulin resistance, we observe from (4.15) and Fig. 4.6 that the production of insulin also increases. This results in high blood insulin, which is a major symptom of Type 2 diabetes [94]. Additionally, since the beta cell is forced to produce higher amounts of insulin, it has to work more and this eventually causes the cell life to reduce. This often happens in late diabetes cases where starting from insulin resistance, the beta cells are lost as well, compounding the problems of a diabetic patient [92].

4.4.3 Effects on Channel Propagation Delay

Since the concentration of glucose and insulin both increases as a result of insulin resistance, it may seem like an increase in both $P(B_G)$ and $P(M_I)$. This should reduce the average channel propagation delay, however, that is not the case. Although, $P(B_G)$ remains high because of high glucose level and appropriate number of GLUT2 receptors on the beta cell, the effective number of insulin receivers reduces at the muscle cell. This reduces $P(M_I)$ by (4.9) and causes a reduction in the corresponding number of active GLUT4 transporters at the muscle cell membrane. The overall effect is an increase in the average channel propagation delay of the insulin-glucose system.

4.5 Conclusion

Therefore, we provide and analyze a molecular communication model of the insulin-glucose system from the ICT perspective. Insulin and glucose molecules are considered as modulated molecular information carriers. These two molecules operate simultaneously in the system in a single physical channel, making a forward and a feedback channel operating on insulin and glucose molecules respectively. The channel capacity, propagation delay and insulin transmission rate are analyzed for a two-cell network between one pancreatic beta cell and a receiving muscle cell, connected through a capillary. Reduction in the number of receptor elements due to insulin resistance is also investigated. The results of analyses point out a correlation between an increase in insulin resistance and a decrease in the channel capacity, an increase in the transmission rate and an increase in the propagation delay. This model may help in the investigation, diagnosis, treatment and management of insulin resistance by means of novel diagnostic methods based on ICT-based metrics as well as provide means to integrate the insulin-glucose system with ICT models of other body networks such as the nervous system.

Chapter 5

SPINAL CORD INJURIES AND TREATMENT PROPOSALS

5.1 Introduction

The nervous system is one of the most important networks of the body that forms the basis of human intellect and stores our experiences as memories. It is composed of networks of neurons connected by means of synapses to receive, transmit and process the information of the body. The synapses themselves can either be electrical or chemical. The manipulation of information by the nervous system is still not clearly understood and an information and communication theoretical (ICT) framework is required to evaluate the relationships between various entities in the system and then address the problems of this system [117]. Several theoretical models of the nervous system from various aspects are provided in our previous works where we target the nervous system from an ICT perspective to find important parameters such as the channel capacity, rate regions and network delays from the perspective of single and multiple neurons [82, 2, 3, 60, 118, 57, 119, 120].

The communication between brain and spinal cord is maintained by ascending and descending spinal cord pathways [121]. Each spinal cord pathway constitutes of neurons having long axons projecting between the brain and spinal cord. In patients with spinal cord injuries (SCI), advanced amyotrophic lateral sclerosis (ALS) or brain-stem strokes, the brain is functional, however, its signals are not transmitted to the muscles due to interruption in the transmission pathways.

According to the recent statistics [122], approximately 282000 persons suffer from SCI and each year 17000 new SCI cases occur in U.S. Incomplete tetraplegia is the

Table 5.1: Types of SCI and Corresponding Percentages

Category	Percentage
<i>Incomplete Tetraplegia</i>	45%
<i>Incomplete Paraplegia</i>	21.3%
<i>Complete Paraplegia</i>	20%
<i>Complete Tetraplegia</i>	13.3%

most frequent type of SCI [122] as shown in Table 5.1.

Two main approaches exist for reestablishing the brain-spinal cord connection in SCI patients, namely, biological methods and ICT-based treatment techniques. The later one is the main interest to us since it has provided more encouraging solutions. ICT-based treatment techniques of SCI can be achieved by, (i) use of Neural interface systems (NIS), and (ii) employment of artificial neurons (ANs). NIS were first developed for animals [123, 124, 125, 126, 127, 128] and then utilized for human patients to extract control signals from nervous system activities and operate assistive devices including computer cursors [129, 130, 131, 132, 133], robotic prostheses [134, 135, 136, 137], and more importantly, apply electrical stimulations to nervous system or muscles for restoring control of muscles [138, 139, 140, 141, 142, 143], which can also induce partial neurological recovery [144, 145]. On the other hand, employment of artificial neurons targets the replacing of the injured neurons with engineered artificial neurons.

Here, we first discuss the mechanisms of SCI to give an insight into the progression of these injuries. We then present the treatment techniques that exist in the literature which include biological, neural interface and artificial neuron based approaches. Specifically, for the neural interface and the artificial neuron, where ICT techniques may benefit existing designs, we also identify the key characteristics a successful treatment technique should have. Based on the preceding findings and discussions, we then

present two comprehensive approaches that may be able to treat SCI in their own different manners. The first approach is based on NIS where we identify key areas for improvement in the existing NIS setups. We also identify important areas such as feedback which are as yet unexplored. The second proposal is for the development of an artificial neuron based on nanomachines that act as a bridge to move neural signals across the injury site. The proposed artificial neuron will involve plasticity along with self-organization and will thus be able to adapt in the injury area to make new nervous connections. Additionally, the machines are proposed to include energy harvesting so that they are truly independent in nature. Apart from SCI treatment, the proposed schemes can be used in a variety of Internet of Bio-Nano Things (IoBNT) applications where neural interfaces are required and may assist in the development of artificial structures that process information in ways similar to the human brain.

5.2 Spinal Cord Injury Mechanisms

Following spinal trauma, the affected neurons experience the effects of the injury in three consecutive phases, namely, the primary, secondary and chronic injury phases.

- *Primary injury* refers to the condition within minutes after the injury happens. The injury immediately disrupts axonal pathways and damages cell membranes depending on the physical trauma [146]. The ischaemia, i.e. toxic chemical release from damaged membranes, initiates the secondary injury [146].
- *Secondary injury* constitutes the time interval within hours to weeks after the injury [147]. Following events such as edema, ischemia and excitotoxicity, the toxic environment results in the death of neurons and their supportive cells (oligodendrocytes) [148]. Deaths of oligodendrocytes cause demyelination, which is the loss of myelin insulator that surrounds axons [149]. Distorting and even blocking axonal propagation, demyelination eventually disconnects (partly or wholly) the brain and spinal cord from each other [149]. This manifests itself in sensory and motor deficits. Moreover, the formation of glial scar, which is a

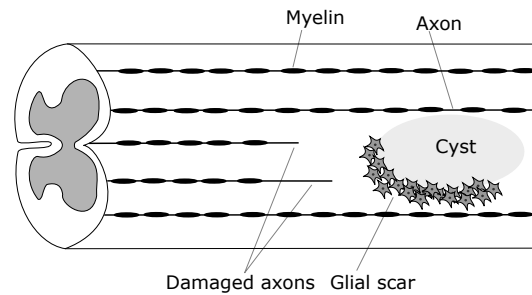


Figure 5.1: SCI and the formation of glial scars.

tissue that barriers axonal regeneration as shown in Fig.5.1, also occurs.

- *Chronic injury* phase covers the days to years following the injury [147]. In this phase, injured axons experience a special kind of degeneration called Wallerian degeneration that disconnects the distal parts of the axons from their cell bodies while the glial scar continues to grow [150]. Since the injury area is stabilized in the chronic phase, most treatment techniques aim to target this particular phase.

The overall result of SCI mechanisms is either a sensory or a motor loss or both at the same time [146]. Reestablishing the brain-spinal cord connection is important to maintain sensory and motor function and too address this goal, several approaches are proposed from different perspectives that are presented in the next section.

5.3 Treatment Techniques

To address the problem of SCI treatment, a great deal of research has focused on recovering the injured spinal cord and several clinical trials have been performed [151]. However, to date, a successful method for complete recovery still does not exist. Present treatment approaches fall in two main categories, namely, the biological treatments and the ICT-based treatment techniques. ICT-based techniques can further be divided into NIS and artificial neuron systems.

5.3.1 Biological Treatment Approaches

Biological treatment approaches have proven successful to some extent in laboratory experiments. However, this is not the case for most of the clinical trials. In addition, using any single one of these approaches seems to maintain only partial recovery and a complete recovery may only be achieved with the combination of various approaches. We discuss some of these techniques below.

Drug Delivery

Delivering drug onto the injury site mainly aims to support regeneration of the injured axons, to protect surviving neurons from post injury toxicity and to improve the conduction on demyelinated axons [152]. Nanoparticles, nanowires and carbon nanofibers are used in several experimental studies as drug delivery tools because they can pass bloodspinal cord barrier to reach the injury sites [153]. Drug delivery can also be performed via transplanted cells and gene transfer [154, 155].

Several growth factors have been employed to help injured axons to recover. Among these, most known is neurotrophins, which can improve nerve regeneration, synaptic transmission and plasticity [156].

In drug delivery for neuroprotection purposes, inhibitory molecules that block the neural signaling activity causing apoptosis are delivered to the injury site to protect the surviving neurons [157]. While the above studies have shown some improvement regarding neuroprotection and regeneration, drug delivery is able to sustain limited recovery only. The limitation is that besides protection, injured neurons also need supportive environment (scaffolding) in which they can regrow.

Cell Therapy

Cell therapy efforts are mostly based on grafting of peripheral nerve cells and transplantation of stem cells. In addition, different types of cells are used in studies to achieve axonal growth and remyelination such as olfactory ensheathing and Schwann cells [158, 159]. Transplantation of neural crest and mesenchymal stem cells in adult

bone marrow are widely investigated as a cell based therapy [160, 161], since stem cells have differentiation capabilities that allow them to change into neurons and glia.

Schwann cells, which are responsible for producing myelin sheaths in peripheral nerves system, are shown to be capable of improving remyelination as well as axonal growth by secreting neurotrophic factors [154, 162, 158]. Although recovery in terms of axonal regeneration and remyelination are observed, some issues on cell transplantation regarding immune rejection and tumor formation exist [163, 164].

Tissue Engineering

Tissue engineering aims to promote regeneration of injured nerves. After the injury, natural scaffolding for organs and tissues is damaged and the lack of supporting environment discourages nerve regeneration. Re-maintaining supporting function, tissue engineering benefits from the materials that can mimic natural scaffolding such as hydrogels, nanofiber scaffolds [165, 166] and hybrid applications [167, 168]. The chosen materials should have mechanical properties that are similar to that of the real tissues and a porous structure that enables axon growth and cell transplantation [169].

Hydrogels Hydrogels are biocompatible networks of polymers which are similar to the tissue within the spinal cord. Either bridging or filling the injury site, they make the environment proper for neural regrowth [150, 170, 171, 172]. Furthermore, that promote regrowth can be introduced in hydrogels with relative ease [169].

Nanofiber scaffolds Nanofiber scaffolds not only provide supportive scaffolding for damaged cells but also are able to deliver drugs and support transplanted cells [173, 174].

Hybrid applications Various combinations of hydrogels, nanofibers, neurotrophic factor or drugs have been shown to be promising for recovery after the injury [175, 168, 176].

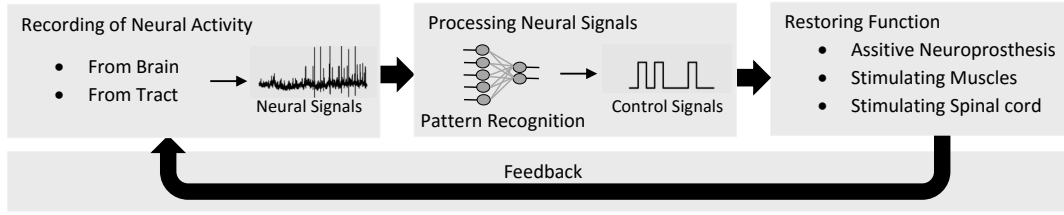


Figure 5.2: Components in a neural interface system.

5.3.2 Neural Interface Systems

While aforementioned biological techniques aim to provide treatments for this signal transmission problem, NISs are more successful in utilizing brain signals to restore motor function in patients. Main components of a NIS to reach this aim are depicted in Fig. 5.2. Each of these components are discussed in the following.

Nervous Recordings

Neural signals can be recorded from brain [177, 178, 129, 142, 145, 137, 179] or spinal cord [127]. While the first one is more common in the literature, the later one can potentially provide signals richer with volitional information in a smaller anatomical region. The important factors in selecting a recording technique in a NIS include efficacy, safety (particularly with regards to surgery), reliability, longevity, required training and support, cost and availability. In the following, we review signal recording techniques from both of these sources and discuss their advantages and limitations.

Recording from the Brain Brain signals can be recorded noninvasively by techniques such as electroencephalography (EEG), magnetoencephalography or functional magnetic-resonance imaging. These techniques have low temporal or spatial resolution, low signal-to-noise ratio and/or poor sensitivity to high-frequency changes. Hence, they are insignificant for investigating the short-lived spatio-temporal dynamics of many brain processes [180]. However, the feasibility of these techniques in detecting motor intent to some extent is demonstrated in studies such as [137, 145],

where robotic exoskeletons are operated with use of control signals extracted from EEG and electrooculography recordings. In contrast, recordings with exceptionally high signal-to-noise ratio, less sensitivity to artifacts than EEG, and high spatial and temporal resolution can be conducted by invasive recording techniques such as electrocorticography (ECoG) and intracortically electrode placements [180]. In ECoG, electrodes are implanted subdurally on the surface of the brain, hence, they are able to record activity of groups of neurons and provide movement-related field potentials. The implantation of electrodes in the cortex is required to directly record neuronal action potentials or local field potentials from the brain, which results in significant clinical risks due to infection or the formation of scar tissues. Hence, the electrodes require a very small cross sectional area to minimize the damage to the tissue. Moreover, this recording technique requires sophisticated signal processing and computationally intensive algorithms to interpret neural activity and reliably separate signals of different neurons [181]. The long-term stability of the implanted electrodes in an individual with tetraplegia is demonstrated in [133], where point-and-click control is successfully achieved 1000 days after implantation of a 100-electrode array.

Recording from Tracts While recording electrical signals from brain is studied intensively in the literature, many technical challenges still exist in usage of micro-electrodes to reach stable recording of individual cell activities. The most important one is formation of a layer of activated astrocytes around the recording electrode that makes long-term recording of single spikes very difficult. Hence, recording motor control signals through cross sectional area of corticospinal tract is investigated in [127, 128]. The main drawbacks of using this recording source for a NIS can be listed as bellow.

- Designing a mechanical stable array of electrodes is challenging.
- The range of electrode array is limited and number of neural sources are far more than the array recording electrode contacts.

- The control signal is not received in corticospinal tract in patients with tetraplegia or ALS.

Signal Processing

The real-time processing of recorded neural data is required after the recording to derive the movement trajectory. This processing is seen as a pattern recognition system that contains three main parts: (i) feature extraction, (ii) dimension reduction and feature selection, (iii) classification or regression. First, the data is processed to derive its features, i.e., discriminative information, from the properties of the signals. Since the brain signal is highly noisy and is made from many simultaneous sources, extracting an appropriate feature set is a challenging task. Moreover, these data can contain undesirable components, called artifacts, that should be removed to improve the performance of NIS. Furthermore, not all of the recorded signals through multiple channels are relevant for understanding the phenomena of interest. Hence, dimension reduction and feature selection methods are used to remove irrelevant and redundant information. In the last step, the feature set is used in a classification/regression algorithm to recognize the user's intention. In Table. 5.2, a list of methods used for each step of a pattern recognition system are provided [182].

Restoring Function

The extracted control signals from recorded activities of nervous system is used in different NISs such as brain computer/machine/spine interface [125, 142, 137, 126, 130, 131, 132, 133, 134, 135, 136, 138, 139, 140, 141, 143, 145, 182], or spinal cord computer interface [127, 128] to restore or replace natural function of a paralyzed or lost limb in one of following manners.

Operating Assitive Neuroprosthesis Real-time control of the robotic prosthesis or exoskeleton finds its applications in practical rehabilitation, replacing the lost motor function and the support of daily actions. Studies on using intracortical recordings

Table 5.2: A list of some methods used in a pattern recognition system

Process	Method
<i>Feature extraction</i>	AutoRegressive component
	Wavelet transform
	Common spatial pattern
	Matched Filtering
<i>Dimension reduction</i>	Principal component analysis
	Independent component analysis
<i>Feature selection</i>	Genetic algorithm
	Sequential selection
<i>Classification</i>	Linear discriminant analysis
	Support vector machine
	Bayesian Statistical Classifier
	K-nearest neighbor classifier
	Artificial neural network

of the brain to control robotic prosthesis [134, 135, 136] demonstrate that paralyzed patients are able to recreate multidimensional control of complex devices even years after injury. This ability is utilized in [137] to control a hand exoskeleton which helps patients to restore full independent daily living activities. Moreover, it is shown that patients achieve the ability to perform voluntary motor control in key muscles below the spinal cord injury level with sufficient trainings and use of EEG-controlled robotic actuators [145].

Direct Stimulation of Muscles Directly stimulating muscles by signals extracted from intracortically recordings is first studied on monkeys with transiently paralyzed arm [138, 140, 139] and then applied to a paralyzed human [143]. In these studies, the exact control signal for each of the muscles included in an activation are extracted and then used to stimulate the muscles to achieve the ability of performing that action. Results depict that intracortically recordings can be linked in real-time to muscle activation, which enables the control of muscles using the activity of neurons in the motor cortex. This process essentially bypasses the spinal cord and restores voluntary control of the paralyzed muscles.

Spinal Cord Stimulation Epidural spinal cord stimulation and intraspinal microstimulation are two existing method in the literature for inserting motor control signals back to the motor circuitries. In the first one, the stimulating electrodes are placed in the epidural space of the spinal cord [144], while electrodes are implanted within the ventral gray matter [183, 184] in the later one.

The sensory feedback from body, which is processed by neurons in spinal cord, is important in controlling the motor commands [185]. Hence, the idea of stimulating spinal cord just enough to make it sensitive and responsive to sensory input is studied in [144], where an array of 16 electrodes is implanted in epidural space of the spinal cord of an individual with a clinically motor complete spinal cord injury. The epidural stimulation in [144] resulted in locomotor-like patterns and full weight-bearing standing with assistance provided only for balance. Using brain signals to control

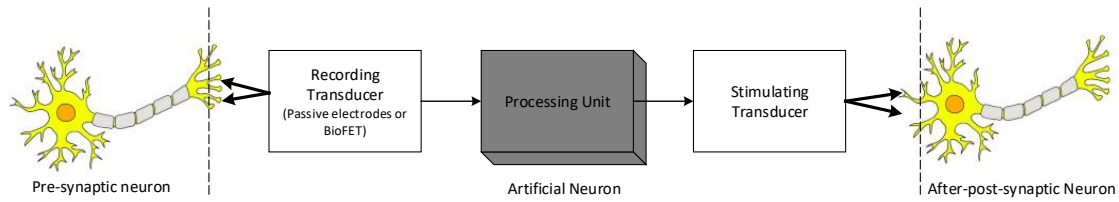


Figure 5.3: The general architecture of an artificial neuron.

the epidural stimulation is further studied in [186, 142], where locomotor movements like walking and climbing are achieved in paralyzed monkey and rat, respectively. In [141], intraspinal microstimulation is used to control hand movement in monkeys whose hands were temporary paralyzed. This stimulation method provides more natural recruitment order of motor units and reduces number of required electrodes and controllers compared to epidural stimulation. However, higher risk of tissue damage exist due to implantation of electrode into the spinal tissue, implanted electrodes retain surrounding scar tissue and long-term stability and safety of implanted electrodes are challenging issues [187].

Feedback

It is shown that visual monitoring of limb's motion can partially correct the shortcomings in movement of patients with large-fiber sensory neuropathy, who face slow and uncoordinated movement as a result of importance of somatosensory feedback for normal motor control [188]. Hence, patient's vision provides the first feedback for a NIS system. However, the effect of visual feedback on movements is not as fast as proprioceptive system, which causes instability in movement [189]. Thus, utilizing proprioceptive feedback or a feedback from spatiotemporal characteristics of limb's in a NIS can assist patients to perform more natural movements and also feel stimulated limbs or neuroprosthesis [186, 190]. As an example, stimulating peripheral afferent nerves in the limb's stump with intrafascicular electrodes [191, 192] or cuff-like electrodes [193] is studied in the literature for providing the natural sensation of missing

limbs in amputee subjects. However, these methods are not applicable in paralyzed patients who has also lost sensory pathway. In such cases, the tactile and proprioceptive feedback can be provided by artificially stimulating the central sensory system [189].

5.3.3 Artificial Neuron

Another potential direction for treatment of SCI could be the use of artificial neurons that can replace injured biological neurons. If perfectly implemented, such an approach can potentially reverse the loss of any function as a result of SCI, however, the associated challenges are not small by any means either. The general architecture of an artificial neuron (AN) is shown in Fig. 5.3 where a presynaptic neuron connects to a processing unit by means of a transducer. The AN performs necessary processing similar to a biological neuron and conducts the signal back to the biological systems.

Some key characteristics required from artificial neuron systems that may one day replace biological neurons are briefly discussed below:

- The artificial neuron should be able to change synaptic weights in a plastic manner since plasticity is forms the basis of memory and cognitive functions of the nervous system.
- The scale of implementation decides the level on which the system behaves similar to a biological neuron. From the perspective of a single neuron, the scale can either be synaptic or neuronal. On synaptic scale, individual synapses can be identified and interfaced to synapses from other neurons, whereas on neuronal scale, the neuron as a whole may be interfaced. Both the systems require a different set of interfacing techniques but the complexity and control available in artificial neuron systems will increases as we decrease the scale.
- The artificial neuron should be bio-compatible. While electronics have been implanted inside the body for quite some time in various forms, here bio-

compatibility should extend to the bio-interfaces of the artificial neuron within the body [194].

- The sizes of the devices should be small enough to be on the scale similar to biological neurons. The scale of design discussed above is critical in this respect. Synaptic clefts on average are 20 *nm* in length whereas spinal neurons are significantly larger in size.
- Energy efficiency is another major issue for any implanted system. Long lasting batteries may not be viable solutions because of replacement requirements. This energy can be harvested from the body heat or blood pressure but other novel methods can be proposed. For instance, artificial neurons may be constructed such that they can harvest energy directly from the body and use that energy to produce biological molecules that may further be used to communicate with neurons of the body.
- Processing of biological inputs may require the use of amplifications since the transmission needs to be carried over some distance and the typical energy of signals is on the order of μJ . Such amplifiers should be low noise, low power and high precision.
- The implant processes are also very critical since any implant is as good as the implant process. On scales as small as neurons, surgery by human surgeons is not a viable solution. The implant technologies may require the use of robotic surgeries that are capable of taking decisions on their own. Apart from the robotic arms and surgery tools, this requires significant machine learning and artificially intelligent systems. Such problems can be mitigated by using systems that deploy themselves by means of self-organization.

Although the challenges are very significant, the benefits of such systems are also unlimited. Apart from treatment of SCI, such systems may be used to develop computing systems similar to the human brain possibly creating a revolution in machine

learning and artificial intelligence research. Additionally, these systems may be used as bio-cyber interfaces for IoBNT applications targeting the nervous system.

To date, no work exists that may be considered as a replacement for a natural neuron, therefore, we discuss some current works in similar directions. Two major directions, namely, neuromorphic and biomimetic neuron approaches exist in literature. Here we review existing literature on these approaches and propose future research based on these direction.

Neuromorphic Neurons

Neuromorphic neurons are based on electronic circuits that mimic the neuro-biological architecture. These circuits are particularly used to target neuromorphic computing where very-large-scale integration (VLSI) of individual neurons or synapses are used to compute in a manner similar to the human brain [195].

The biggest advantage of neuromorphic systems is that they include plasticity as the basis of their models, however, implementation of artificial neurons by neuromorphic methods faces several bottlenecks. These include the large size, high power requirements and interfacing issues. Some devices such as memristors use relatively low power and have smaller sizes, however, the interfacing issues remain [196].

Biomimetic Neurons

Biomimetic circuits mimic biochemical processes using synthetic materials [197]. Since most communication between biological entities occurs by means of molecule exchange and biochemical reactions of these molecules, biomimetic circuits are ideal candidates for direct interfacing with biological systems. Biomimetic systems are used to realize applications such as artificial muscles, smart membranes, biological transducers and brain-machine interfaces [197].

The basic function of a neuron is realized by means of enzyme-based amperometric biosensors and organic ion pumps in a biomimetic fashion by [198]. Although the work is bio-compatible and the detection of bio-molecules is achieved on scales similar to

those of actual neurons, many of the characteristics expected from an implantable artificial neuron described in this section are not met. For instance, there is no plasticity in the system and the size of the hardware required for [198] is very big involving entire computers that are used to process the signals. Additionally, the system is tested in vitro without any interface to actual neurons.

Similar works that involve plasticity and interface directly with biological neurons may be the first step towards the development of organic biomimetic artificial neurons. Size reductions and in vivo testing may follow afterwards. Another direction could be the use of plasticity designs from neuromorphic systems based on biomimetic implementations. This may perhaps be the approach that is most successful and has the least time-to-market.

5.4 ICT-based Treatment Techniques

5.4.1 Neural Interface Systems

The main technical challenges to provide an ICT-based treatment for spinal cord injuries can be itemized as follows.

- Discovering the degree of freedom that should be handled by the system for the intended task and selecting the recording method that provides enough independent control signals.
- Stably and reliably recording of neural signal.
- Realization of a fully implantable system.
- Providing high bandwidth for transmitting recorded signal while considering the power consumption.
- Selecting appropriate stimulation method.
- Providing somatosensory feedback.

In this section, we first propose a framework for realizing a NIS that addresses these challenges. Our proposed NIS utilizes the neural signals recorded from brain and stimulates the spinal cord for restoring the function in paralyzed patients.

Recording Cite

While the action potential or local field potential recordings with intracortically implanted electrodes provide the most detailed information about the activity of the neurons, placements of these electrodes causes damages to the brain tissue and the long term viability of these implants is not approved yet. On the other hand, ECoG recordings are robust over long periods [199] and higher spatial resolutions can be achieved with utilizing micro-ECoG grids compared to clinical ECoG grids [200]. Moreover, ECoG signal is utilized for offline decoding of seven degrees of freedom for arm movements in Monkeys and online decoding for individual finger movements in human [201, 202], which demonstrates that it can provide enough independent control signals for simple loco-motor task. Hence, we select the minimally invasive implantation of micro-ECoG grids for our NIS, which also decreases the complexity of recording system compared to action potential recording methods and accelerates the realization of a fully implantable System.

The recording device is implanted together with a transmitter, which uses wireless transmission for transferring recorded signals to a processing unit. The wireless transmission is chosen because it offers more reliable system compared to wired transmission for patient comfort without the risk of inflammation[203]. The bandwidth demand of the wireless transmission channel between the brain implant and external processing unit is correlated with the number of recording channels employed. For better estimation of brain commands, we need densely distributed recording sites requiring high data rates (on the order of Mbps)[204]. On the other hand, the transmitter should dissipate relatively low energy because thermal heat produced by power consumption can damage neural tissues[205]. Considering these limitations, Ultra-wide band(UWB) (3.1-10.6 GHz) transmitter, which can provide high data rate within

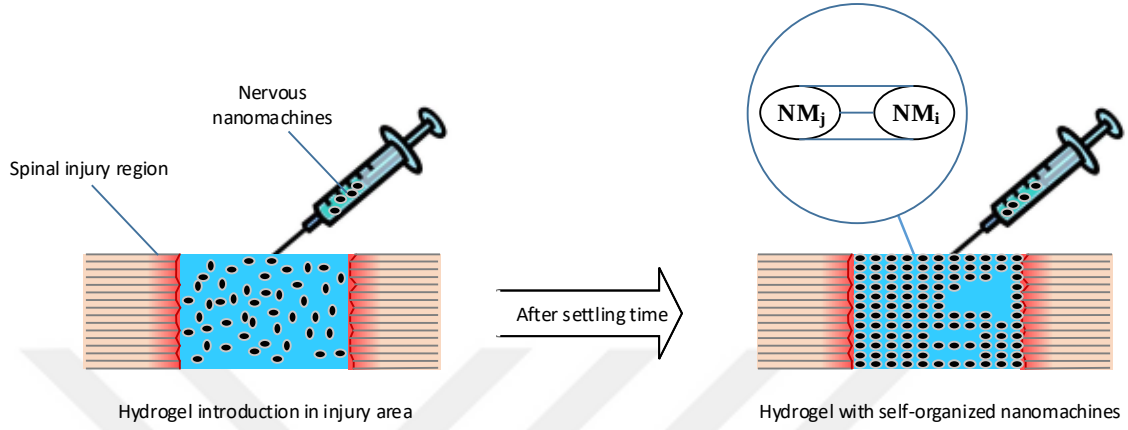


Figure 5.4: Diagram of the self-organizing artificial neurons suspended in a hydrogel.

allowable power dissipation range with minimal chip area[206], is the most suitable solution[207]. Therefore, the communication interface between the brain implant-processing unit of our NSI will consist of a simple UWB transmitter at the brain implant and a more complex UWB receiver at the processing unit[208]. Furthermore, compression techniques with efficient implementation such as the one proposed in [209] can be utilized to reduce data rate without affecting complexity so much.

Processing Unit

The processing unit performs two tasks, (i) decoding the intent of the person from neural signals, and (ii) generating control signals for spinal cord stimulation. To perform the earlier one, we propose the utilization of a deep neural network (DNN), which can be trained by a training set of recordings. The advantage of using a DNN compared to other pattern recognition methods is that it extracts the features by itself, which is a complex task since the recorded neural data are high-dimensional and not well-known. Moreover, the DNN predicts the non-linear systems more accurately [210].

For the second task, we need to extract the patterns of spinal cord stimulations corresponding to each intended task by the patient. In [142], the extensor and flexor

hotspots corresponding to different brain signals are identified in intact monkeys, then the monkeys are paralyzed. Since we do not have access to such training data in paralyzed patients, experiments on healthy subjects are needed to detect whether these hotspots are almost the same in different subjects. Even if these hotspots are placed in different locations for each subject, we can utilize the epidural stimulation method of [144] and use a machine-learning algorithm that gets the recorded data from healthy subjects to choose stimulation parameters [144]. Identification of the extensor and flexor hotspots is needed to be done one time for a patient, then this information can be stored in a look up table that is used for stimulating the spinal cord according to each output of the DNN.

The processing unit will be placed in a rechargeable and mobile device. Hence, it needs a wireless transmitter to transfer the stimulation parameters to the site of spinal cord stimulation. The transmission between the processing unit and spinal cord neural stimulator can be performed at lower data rates compared to the brain implant-to-processing unit link since the dimensionality of the transmitted signal reduces. On the other hand, the receiver at the neural stimulator must operate with low energy [205]. Transmitting at low signal energy and covering minimal antenna area makes UWB transmission suitable for this link. Moreover, it is also advantageous in terms of interference [211].

Stimulation Site

It is shown in [144, 142] that epidural spinal cord stimulation provides enough control over lower body muscle to restore loco-motor function. For this aim, the electrodes should be placed over the identified extensor and flexor hotspots and a pulse generator is needed to control the stimulation parameters. The pulse generator receives these parameters, which are sent to it by processing unit, through its receiver.

Feedback

A wearable sensor system (WSS) is proposed to monitor the muscle activities and collect the tactile information. This information will then be processed to provide the stimulation parameters for stimulation of somatosensory cortex based on its topographically matching locations.

The sensory information collected by WSS will then be transmitted to the processing unit to generate the parameters for somatosensory cortex stimulation. For this aim, the responsible regions of somatosensory cortex for sensory feedback from each leg should be identified. This task can be done by using the proposed WSS for collecting tactile and muscle activities in healthy subjects and recording the corresponding activities in somatosensory region during walking. Then, a DNN can be trained by these information to estimate the activity of somatosensory cortex for each output of WSS. Since we consider the SCI injuries in which the sensory pathway is also damaged, our NIS will use these information to generate stimulation parameters that are used to provide same somatosensory active regions in patients. For the data transmission, Bluetooth Low Energy (LE) will be utilized, since it is one of the most suitable technologies for such sensor network to device communication links operating at low data rates with low power requirements[212]. In next step, the processing unit transmits the stimulation parameters to the controller unit that is surgically placed over pia mater. The transmission will be performed through UWB communication interface.

The stimulation should also be done with a minimally-invasive method for long-term brain implants.

5.4.2 Artificial Neuron

Although the preceding neural interface is relatively easy to implement and may return a high degree of motion to patients, the movements are not accurate. The major reason for this is that although the system capacity increases by the use of a neural interface approach solution, the capacity level is not as high as before the injury. Before an SCI, being normally wired, the system has a maximum capacity,

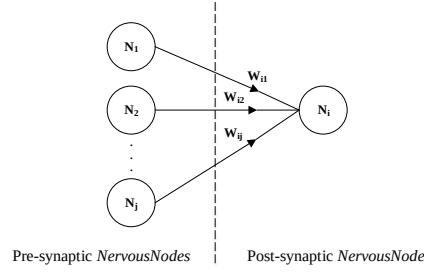


Figure 5.5: A number of presynaptic neurons connected to a postsynaptic AN, each having its own synaptic weight.

however, once the loss of wiring occurs and instead we have to collect information from expected cortical regions and correspondingly, stimulate some estimated areas of the spinal cord for the movement, an information loss occurs, thereby reducing the system capacity. These expected regions come in play since various functions of the body are not truly localized to specific regions of the brain and as such cannot be clearly differentiated. On the other hand, localization of the signals is not an issue in the spinal cord since the collective signals from the brain are routed from different regions and sent through the spinal cord in a specific manner.

As discussed in the neural interface section, the system capacity may be improved by improving the data recoding and stimulation electrodes, however, another solution of this problem could be the retrieval of the lost connections at the injury cite. The capacity could be maximized (at least theoretically) to the pre-injury levels by employing artificial nervous connections across the injury area where the signals are already localized in specific positions. Therefore, we propose a system of artificial neurons (AN) that have neuromorphic properties and while they are suspended in a hydrogel, they can form bridges across the injury cite and make new nervous connections with biological and artificial neurons. Such a system is shown in Fig. 5.4. We discuss the major properties of the proposed AN in the sections below.

Plasticity

In order that the AN is adapts to its environment, plasticity should be introduced in the system and realized by utilizing neuromorphic designs. The firing rate of each AN depends on the signals it receives from synapses with other neurons that may be both biological or artificial. Each synapse of the AN is associated to a variable *Weight* that models plasticity of the synapse and retains the weight of a particular synapse. Consider the case shown in Figure 5.5, where a number of presynaptic neurons are connected to the AN, N_i . Each neuron is associated with its own unique synaptic weight that changes over time as a result of firing patterns of the pre- and postsynaptic neurons [83]. Considering the particular case of a presynaptic neuron N_j connected to N_i , and $S_j(t) = \sum_f \delta(t - t_j^{(f)})$ and $S_i(t) = \sum_f \delta(t - t_i^{(f)})$ are pre- and postsynaptic action potential trains, the change of synaptic weight is given by [83] as

$$\begin{aligned} \frac{d}{dt} w_{ij}(t) = a_0 + S_j(t) & \left(a_1^{pre} + \int_0^\infty a_2^{pre,post}(s) S_i(t-s) ds \right) \\ & + S_i(t) [a_1^{post} + \int_0^\infty a_2^{post,pre}(s) S_j(t-s) ds], \quad (5.1) \end{aligned}$$

where the parameters a_0 represents a constant decrease in the weight of a synapse irrespective of the presence of a spiking signal on pre and postsynaptic terminal, a_1^{pre} and a_1^{post} represent the non-Hebbian long term potentiation at the pre- and postsynaptic terminals, respectively. $a_2^{pre,post}(s)$ is a kernel that represents weight change if a presynaptic spike arrival comes after a delay of s with respect to a postsynaptic action potential and $a_2^{post,pre}(s)$ represent a vice versa case. The kernels here describe a learning window in which if a presynaptic spike comes before a postsynaptic action potential, the synapse gets strengthened by a weight increase and the weight decreases if a postynaptic pulse comes before a presynaptic pulse [83]. This is in line with the Hebbian learning principle that states that 'neurons that fire together wire together and neurons that fire apart wire apart'. This experimentally verified principle is the basis of many learning rules [83] and as should be the basis of the plasticity implementation of the proposed AN.

Self-Organization

As discussed earlier, the implantation of AN systems propose a major challenge since surgery on the nano-scale itself an open problem. This problem can be bypassed by considering systems that use self organization to create the AN bridge across the nervous injury. Such a system is shown in Fig. 5.4 where AN bridges form across the injury area over time.

Self-organization is a hot research area where local interactions between individual entities give rise to an overall structure of the system. The simplest approach towards this goal is the use of electric or magnetic fields. Since the action potentials of the nervous system are themselves electrical signals, the self-organization can be based on its use as the attractor field. Graphene nanosolenoids discussed in [213] may provide the ideal implements in this regard both in terms of size required, low energy requirements and the magnetic properties. The nanosolenoids, however, are not be the only implement required for self-organization because the nervous signals are not continuous in nature and the AN bridge may fall apart if a signal is absent for some time. To ensure longterm sustenance of the structures, we either need mechanical structures such as hinges [214] or by the use of magnetic fields external to the body [215].

Energy Harvesting

For the system to be completely independent, the AN devices need to harvest energy. This energy can be harvested from within the body by means of vibrational energy or body heat [216] or from external sources using wireless power transfer. If an external source is selected for the wireless power transfer, the same may be used for the self-organization of the AN devices and as a readout interface of the AN network as well.

5.5 Applications of SCI Treatments for IoBNT

By revisiting the current treatment techniques for SCI from an ICT perspective, we can provide rigorous optimization frameworks for the improvement of these techniques. However, apart from the treatment of SCI, such systems can have applications in a number of scenarios specifically for the IoBNT. In this section, we discuss some of the other applications that are realizable as byproducts of SCI treatments. Since any SCI treatment technique will involve the interfacing with the nervous system with external sources, a number of applications can be based on such methods apart from SCI treatments.

- Large amount of data can be retrieved from the nervous system by using the interfaces of the proposed SCI treatments. This data can then be used to analyze the fundamental characteristics of the nervous signals and may help reveal the information processing techniques used by the brain.
- We can study nervous diseases such as Alzheimer's disease, multiple sclerosis, Parkinson's disease in an intimate way using SCI treatments as interfaces to the central nervous system. This may lead to novel disease diagnosis and treatment techniques as well as the identification of key factors that cause such conditions.
- Since the proposed system are online in structure, they may be used for remote health monitoring. Applications such as drug delivery can be monitored and controlled using the nervous system as a wired system of the body.
- The development of the AN may benefit the development of processors that process information similar to the human brain. This may be the next major milestone in terms of processor technologies and may assist in the development of true artificial intelligence capable of feats similar to the human brain.
- The development of extrasensory or motor applications for the integration of man and machines under the IoBNT framework may benefit from the proposed

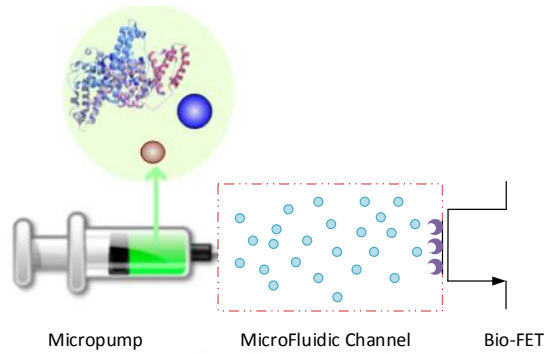


Figure 5.6: Proposed test setup for an artificial synapse.

treatments as well. Interesting application in such directions may include extrasensory feedbacks to achieve new sensations, mechanical body suits that allow superhuman strength, direct integration of gaming in the human body, among others.

5.6 Proposed Architecture of Artificial Synapse

In the approach of Fig. 5.6, the transmission of the biological molecules into a microfluidic channel is controlled by means of a micropump. Once pumped in, biological molecules will diffuse towards the receiver through an aqueous medium similar to that of an actual synapse and be received on bio-field effect transistors (bio-FET). A bio-FET is a field effect transistor that has biological receptors on its gate terminal and changes its conduction based on the number of molecules that bind to these receptors. In the next few subsections, we will give some detail about all these system constituents.

5.6.1 Micropumps

Although any kind of small pump is often referred to as micropump, a more accurate definition restricts this term to pumps with functional dimensions in the micrometre range. Such pumps are of special interest in microfluidic research, and have become

available for industrial product integration in recent years. Their miniaturized overall size, potential cost and improved dosing accuracy compared to existing miniature pumps fuel the growing interest for this innovative kind of pump.

Within the microfluidic world physical laws change their appearance, for example, volumetric forces, such as weight or inertia become negligible, whereas surface forces can dominate fluidical behaviour. Thus, most micropumps rely on micro-actuation principles, which can be scaled up only to micro sizes.

Micropumps can be grouped into mechanical and non-mechanical devices [217]. The mechanical systems contain moving parts, which are usually actuation and valve membranes or flaps. The driving force can be generated by utilizing piezoelectric, electrostatic, thermo-pneumatic, pneumatic or magnetic effects. Non-mechanical pumps function with electro-hydrodynamic, electro-osmotic, electrochemical or ultrasonic flow generation, just to name a few of the actuation mechanisms that are currently studied [217].

Micropumps have a great potential for bio-inspired applications, for example in a flexible electromagnetic micropump using Magnetorheological elastomer to replace lymphatic vessels or their proposed use in artificial synapses.

5.6.2 Microfluidic Channel

The behavior of fluids at the microscale can differ from macrofluidic behavior in that factors such as surface tension, energy dissipation, and fluidic resistance start to dominate the system. In particular, the Reynolds number, which compares the effect of momentum of a fluid to the effect of viscosity, can become very low and as consequence, when side-by-side, fluids do not necessarily mix in the traditional sense, as flow becomes laminar rather than turbulent [218]. Molecular transport between and inside the fluids occurs through diffusion.

Microfluidic channels are a very popular area of research and have a huge set of applications from inkjet printing to DNA testing, bio-sensors and optics to name a few [219]. The fabrication processes for such channels are relatively well established,

however, they vary from application to application and a new design will be required for the proposed setup.

5.6.3 Bio-FET

Bio-FETs are field-effect transistors that are gated by biological molecules. When biological molecules bind to the FET gate, they can change the gate charge distribution resulting in a change in conductance of the FET channel. Bio-FETs are extensively used in biological sensor applications [220, 221]. A bio-FET consists of two main compartments: one is the biological recognition element and the other is the field effect transistor [221].

A Bio-FET system consists of a semiconducting transducer separated by an insulator layer of SiO_2 from the biological recognition element such as ligand receptors. A bio-FET also consists of a transducer immobilized to the surface which contains the target molecule called analyte. Once the analyte binds to the recognition element, the charge distribution at the surface will change, triggering a change in the electrostatic potential of the semiconductor [222]. The transistor controls the electron flow in the channel using two electrodes, which are called source and drain just like an ordinary FET [222] where the current flow between the two terminals is controlled by the voltage across the gate terminal. As a result of ligand binding at the gate, the conductance of the semiconductor changes, which can be measured and it is directly related to the concentration of the received molecules [223].

5.7 Open Issues in Design of Artificial Synapse

In this chapter, we looked at the basics of the proposed artificial synapse and how it may be developed. The construction of an artificial synapse will not only aid as a platform for nervous repair and man-machine interfaces but also act as a test bench for molecular communications in general. This will open a host of opportunities for developing models of learning and memory that are still not quite well understood by neuroscience and the experimental work conducted will help in combating various

nervous conditions. Some of the directions to pursue the creation of an artificial synapse that will be a part of this thesis will include the following:

- Modeling and simulations of the artificial synapse in multiphysics softwares like COMSOL for parameter and design optimization.
- Emulation of synaptic plasticity in the artificial synapse.
- Once an artificial synapse is successfully modeled, experimental fabrication and validation of designed channel models can be conducted. This will include the following key steps:
 - Selection of injection micropumps.
 - Selection of biological communication molecules and respective receptor pairs.
 - Microfluidic channel and bio-FET fabrication.

Some future directions may include:

- Neuron-artificial synapse interface will be the first step towards the actual implantation of the artificial synapses. The experimental setup for nervous data communication explained earlier may be a good candidate to test such systems.
- Design of biomedical intrabody, environmental and industrial applications.

Chapter 6

MOLECULAR COMMUNICATION SIMULATORS

The unconventional nature of molecular communication necessitates contribution of from a host of fields such as ICT, biology and medicine. This tools that are used to design and analyze such systems are thus very important since they have to work for a host of different research directions. Apart form the mathematical modeling, since the complexity of molecular communication are usually very complex, most problems require the use of simulators tailored to the specific biological environments. These simulators will find immense application in disease diagnosis and treatment studies. Virtual organs simulating the processes of a specific body part and the effects of various pathogens or medicines will form the basis of future research in human biology.

Parameters such as particle size, shape, interaction with the surrounding environment as well as other particles and timescales of different processes have to be considered as important parameters for the design of any molecular communication simulator. For this reason, various research groups around the world are implementing different simulators tailored the their research needs. In the next sections we will provide a brief overview of some of established simulators and give directions for future work related to this thesis.

6.1 Nanonetwork Simulators

Since a variety of nanonetwork simulators have been proposed, several simulators have been designed to deal with nanonetworks in general without taking into account their biological or chemical specifics. Such simulators target the nanonetworking in general and try to emulate their behaviors from aspects such as diffusion or electromagnetic communication. We discuss a few of the important nanonetwork simulators below.

6.1.1 NanoNS

NanoNS is a molecular communication tool developed on top of the widely used network simulator (NS-2) [224]. The main objective of the framework is to provide a simulation tool to create a better understanding of nanonetworks and facilitate the development of new communication techniques as well as the provide a platform for validation of theoretical results. It incorporates the simulation modules for various nanoscale communication paradigms based on a diffusive molecular communication channel based on Brownian motion in aqueous medium and a consequence of the constant thermal motion of atoms, molecules, and their chemical reactions.

NanoNS is implemented in NS-2, which is an object-oriented simulator which is written in C++, and an object-Tcl (OTcl) interpreter. C++ is generally utilized in complicated protocol implementations due to its fast execution and OTcl is used for simulation configuration since it can be changed very quickly. An OTcl library, called ns-mol.tcl, is developed for a new node structure called NanoNode, which is based on the OTcl standalone class of Node. The library along with new nanonetwork components, parameters and methods for molecular communication provides the scheduling of the simulation, settings of the network topology and configuration of nanonetwork parameters.

Data is encoded in a Molecule class, which has the features of the carrier molecules such as proteins, ions or DNA and is then transmitted from a NanoNode sender. A Diffusion class is used to manage the diffusion of the molecules. The environment is assumed to be composed of a virtual lattice structures, necessitating a Lattice class for lattice sides, and every object is relocated according to lattices. A MolPosition class acts as the coordinate system of the unit lattice element. Reception of molecules on the receiver NanoNode is managed by Reaction and ReactionChannel classes.

The input of the simulator is an OTcl script. NS-2 has a command line interface, which can be used to generate a simulation trace file. The trace file can then be used with other tools such as animation tools and graph plotting tools for further analysis.

Simulations require a single machine, which has an installation of NS-2. The

requirements include a system with a C++ compiler and an OTcl interpreter. Granularity is user defined as with the event driven architecture of NS-2.

6.1.2 N3Sim

N3Sim [225] is another simulation framework for diffusion-based molecular communications, which allows the evaluation of the communication performance of molecular networks with several transmitters and receivers in an infinite space with a given concentration of molecules. Transmitters encode the information by releasing particles into the medium, thus varying the concentration rate in their vicinity. The diffusion of particles through the medium is modeled again as Brownian motion, taking into account particle inertia and collisions among particles. Finally, receivers decode the information by sensing the local concentration in their neighborhood. The benefits of such a simulator are multiple: the validation of existing channel models for molecular communications and the evaluation of novel modulation schemes are just two examples.

A three-layer architecture has been designed where the user interface layer interacts with the user to read the input data for the simulation, while the data layer writes the simulation results to files while the domain layer contains the molecular communication model. N3Sim is a Java-based simulator; therefore, it can be used in different operating systems with the appropriate Java virtual machine. The simulation parameters are determined by means of a text configuration file editable by the user, which defines the values of all possible variables in the simulation. These are separated into four groups of variables for clarity: simulation, space, transmitters and receivers. In addition, N3Sim allows to automate the execution of multiple simulations in a simple manner by means of user-defined scripts.

N3Sim is designed to be executed on a single machine with Java 1.6 or higher on Linux and Windows operating systems. The scalability of the simulator can be improved by selecting a higher simulation time step at the expense of the accuracy or by deactivating the collisions among molecules in scenarios with a low molecular

concentration. The time granularity of a simulation is defined by the user by choosing the simulation time step in the configuration file. The simulation space can be either bounded or unbounded, and both 2-dimensional and 3-dimensional configurations are supported in COMSOL Multiphysics. Since the COMSOL package supports cluster computing, once the simulation is at the COMSOL level, clustered computers can be used as well.

6.1.3 Nano-Sim

Nano-Sim is a nanonetwork simulator that specifically targets wireless nanosensor network on a NS-3 based architecture [226]. NS-3 is a discrete-event network simulator for Internet systems and is the advanced version of NS-2. Although, NS-3 is not developed for molecular communication its flexible structure allows implementation of basic elements of molecular communication. Just like NS-2, NS-3 is organized in different software libraries that can work together. User programs, written in C++ or Python programming languages, can adapt these libraries or be linked with them. External animators and data analysis and visualization tools are available. Nevertheless, in order to exploit the full potentials of the NS-3 the command line interface is suggested.

Nano-Sim considers routing and MAC protocols for wireless nanosensor networks with specific application targets in simulations of health monitoring systems. The simulator is designed to be executed on a single machine. The minimal requirements to execute basic simulations are a gcc or clang compiler and Python interpreter. The simulation granularity is defined by users. Event design can be adapted to any granularity and are synchronized in the simulator implementation processing loop.

6.2 Molecular Simulators

Simulations of molecular communications at a molecular level can be conducted by a variety of tools such as Large-scale atomic/molecular massively parallel simulator (LAMMPS) [227]. These general purpose classical molecular dynamics softwares can

be used for simulations of biology, materials science, granular, mesoscale etc., however, they pose a host of problems when nanonetworks are targeted by them. The chief rationale for domain-specific simulators over general purpose tools lies in the promise of improved conceptual control, and the possibility of exploiting the structure of model equations for the sake of computational robustness, accuracy, and efficiency. In this context, several simulators have been designed that target specific molecular communications paradigms. Some of the key simulators are discussed in the remaining section.

6.2.1 *Biological and Nano-Scale communication simulator (BiNS)*

BiNS is a simulation package for molecular communication systems developed at the University of Perugia [228] with a customizable design and a set of tools that allows the creation of objects modeling the behavior of biological entities, which can be regarded as either nodes (transmitters, receivers), carriers, or surrounding objects (e.g. vessel walls). It is possible to configure the properties of the simulated communication channel (e.g. the blood stream or the environment of an in vitro experiment) with the desired accuracy.

It has been implemented in Java. Each object belongs to a generic type of software object, named Nano-Object. Nodes and carriers are specific implementations of the nano-object, and, although they share its general features, they can expose very different functions. In particular, for any scenario, it is possible to differentiate the node object type at any time, thus obtaining a multitude of different node objects. The same properties are available for modeling carriers as nano-objects.

The list of simulated nano-objects are split into smaller lists, which can be handled in parallel by a thread pool. The suitable number of threads and the workload distribution over them depends on the hardware configuration. The simulator uses a fine grained approach for handling collisions between objects. A collision can produce either a bounce or an assimilation. The latter happens only when a carrier collides with a compliant receptor on the surface of a node. The receptors can implement a

non-negligible absorption time, thus could be found by a compliant particle in either busy or free state. The simulated environment can be either unbounded or bounded by a surface of custom shape, referred to as simulation domain.

The simulation is organized in discrete time steps. Each step consists of a number of phases, in which software objects are triggered in order to execute the operations associated with their specific behavior. The main phases are: transmission phase, reception phase, information processing phase, motion phase, object destruction phase during which objects are removed due to lifetime expiration or because they exited from the area of interest, and collision management phase, which implements the elementary interaction between particles.

6.2.2 CalComSim

CalCcomSsim is calcium signaling-based molecular communication system simulator design for both synthetic and natural cellular communication inside 3D human tissues [229]. An integration of the biological models for such mechanism along side with an stochastic solver is used to handle multiple and parallel reactions in each cell of the tissue. Three different types of tissues are implemented, which can be classified as: non-excitable (tissues which cannot propagate electric current), excitable (tissues which can propagate electric current) and hybrid (tissues capable of communicating with both excitable and non-excitable tissues). Real data was used to model the following example of those different tissue types: epithelial (non-excitable), smooth muscle cells (excitable) and astrocytes (hybrid). Gap junctions were also stochastically modeled.

This entire framework enables proper analysis of such communication system based on completely validated models fitted with real data. This software thus can be used by, not only telecommunication engineers and biology scientists, but also pharmaceutical researcher that, in a combined effort, can design new drugs and treatments for diseases.

CalCcomSsim has been implemented in Python, and requires the following depen-

dencies: Numpy, Scipy, Pypy, Cpython. The simulator is controlled by command.

6.2.3 BNSim

BNSim is multithread java simulator of Bacteria-based networks [29]. These networks interconnect engineered bacteria that communicate at nanoscales. BNSim integrates three simulation methods: (i) the Gillespies stochastic simulation algorithm (SSA), which solves the chemical master equation in order to determine the time evolution of a chemical system; the extremely small granularity of events makes this approach unpractical for population-level problems; (ii) stochastic differential equations, used to model large-scale chemical system with a controlled level of approximation; (iii) a hybrid algorithm which integrates both the SSA and the approximated methods based on differential equations.

The approximated methods based on stochastic differential equations are used to model the behavior of species that are present in large quantities and generate scalability issues. In addition, a specific thread scheduler is implemented to balance the workload for simulating the intracellular dynamics and interactions of entities.

6.3 Nervous Simulators

The central importance of the nervous system dictates the necessity of having efficient and comprehensive nervous simulators. Several such simulators exist in the literature.

It requires in some cases to use detailed biophysical representations of the neurons, for example when intracellular electro-physiological measurements are to be reproduced. In this case, one uses conductance-based (COBA) models, such as the Hodgkin and Huxley (HH) [44] type of models. In other cases, one does not need to realistically capture the spike generating mechanisms, and simpler models, such as the integrate-and-fire (IF) model are sufficient, which are much faster.

There are two families of algorithms for the simulation of neural networks: synchronous or ‘clock-driven’ algorithms, in which all neurons are updated simultaneously at every tick of a clock, and asynchronous or ‘event-driven’ algorithms, in which neu-

rons are updated only when they receive or emit a spike. Hybrid algorithms also exist in the literature. Each of the strategies has its own advantages and disadvantage based on a host of factors. In the remaining of this section we take a brief look of some of these simulators.

6.3.1 NEURON

NEURON is a simulation environment for creating and using empirically-based models of biological neurons and neural circuits. It is well-suited for COBA models of cells with complex branched anatomy, including extracellular potential near the membrane, and biophysical properties such as multiple channel types, inhomogeneous channel distribution, ionic accumulation and diffusion, and second messengers. Over the past two decade it has undergone many enhancements that make the construction and simulation of large-scale network models easier and more efficient. A huge body of works exists that is based on NEURON. These include works as simple as single neurons to works where thousands of neurons exist in a network.

Key attributes of biological neurons and networks have direct counterparts in NEURON. For instance, NEURON users specify the gating properties of voltage- and ligand-gated ion channels with kinetic schemes or families of HH style differential equations. Another example is that models may include electronic circuits constructed with the LinearCircuitBuilder, a GUI tool whose palette includes resistors, capacitors, voltage and current sources, and operational amplifiers. Synapse and artificial cell models accept discrete events with input stream specific state information. It is often extremely useful for artificial cell models to send events to themselves in order to implement refractory periods and intrinsic firing properties; the delivery time of these ‘self events’ can also be adjusted in response to intervening events.

NEURON source code and installers are provided free of charge and users can extend it by writing new functions and biophysical mechanism specifications in the NMODL language. Along with coding options, a GUI is also present that has a large number of tools which can be used to construct models, exercise simulations, and

analyze results, so that no knowledge of programming is necessary for the productive use of NEURON. NEURON also supports parallel processing and can work on a host of operating systems.

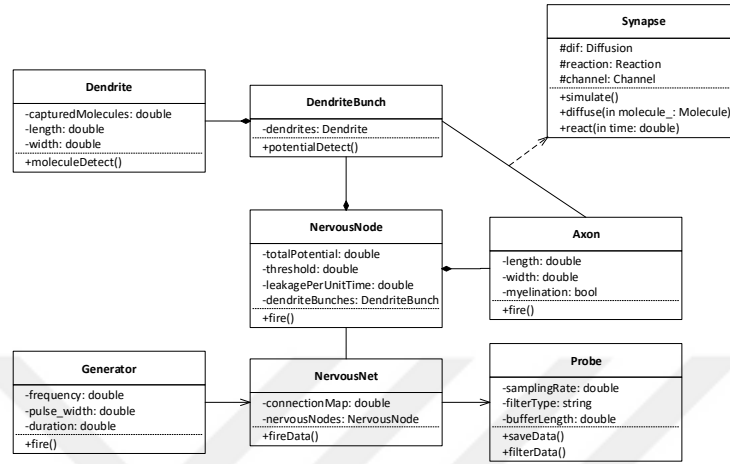
6.3.2 General Neural Simulation System (*GENESIS*)

GENESIS is an extensible general simulation system for the realistic modeling of neural and biological systems [230]. Typical simulations that have been performed with GENESIS range from subcellular components and biochemical reactions to complex models of single neurons, simulations of large networks, and systems-level models. Much like NEURON, GENESIS is also based only on the empirical HH and IF models.

GENESIS uses an interpreter and a high-level simulation language to construct neurons and their networks. This use of an interpreter with pre-compiled object types, rather than a separate step to compile scripts into binary machine code, gives the advantage of allowing the user to interact with and modify a simulation while it is running, with no sacrifice in simulation speed. Commands may be issued either interactively to a command prompt, by use of simulation scripts, or through the graphical interface.

GENESIS provides a variety of mechanisms to model calcium diffusion and calcium-dependent conductances, as well as synaptic plasticity. There are also a number of ‘device objects’ that may be interfaced to a simulation to provide various types of input to the simulation (or measurements.

GENESIS has a well-documented process for users themselves to extend its capabilities by adding new user-defined GENESIS object types (classes), or script language commands without the need to understand or modify the GENESIS simulator code. Parallel GENESIS (PGENESIS) is an extension to GENESIS that runs on almost any parallel cluster, supercomputer, or network of workstations where serial GENESIS itself is runnable.

Figure 6.1: Class diagram of a nervous network in N^4Sim .

6.4 Toward N^4SIM Nervous Nanonetwork Simulator

6.4.1 Motivation for Nervous Nanonetwork Simulator (N^4SIM)

For large-scale nervous networks, the central importance of the nervous system dictates the necessity of a dedicated simulator that can analyze the system in an efficient and comprehensive manner.

Network simulators are required for the analysis of large networks since the evaluation of closed-form solutions for network parameters using traditional analytical tools is not possible. From a molecular communication perspective, parameters such as particle size, shape, interaction with the surrounding environment and other particles, and timescales of different processes are important for the design of any simulator. Although general purpose molecular dynamics softwares such as Large-scale atomic/molecular massively parallel simulator (LAMMPS) [227], may be used to simulate nanonetworks, they are not scalable to set up large networks because of high computational overheads. Domain-specific simulators are more preferred because they are generally more robust, accurate and efficient. Therefore, various research groups around the world are implementing different simulators tailored to their research needs. In this regard, *NanoNS* and *N3Sim* are general purpose simulation frame-

works for diffusion-based nanonetworks [224, 225]. *Nano-Sim* is another nanonetwork simulator that specifically targets wireless nanosensor networks [226]. *BiNS* is a Java-based simulation package for molecular communication systems that allows the creation of objects that model the behavior of biological entities [228]. *CalComSim* is a calcium signaling simulator for both synthetic and natural cellular communication [229]. *BNSim* is multithread java simulator of bacteria-based networks [29]. Apart from molecular communication simulators, *NEURON* and *GENESIS* are two nervous simulators based on the empirical Hodgins-Huxley (HH) and integrate-and-fire (IF) models [231, 230]. These simulators are scalable from single to multi neuron structures, however, they ignore molecular communication in the system and instead use basic circuit models of neuron for simplicity. Therefore, these simulators are inadequate for a detailed understanding and evolution of molecular communication in the synapse.

The discussion above provides the motivation for a nervous simulator that represents the realistic behavior of molecular communication in the nervous nanonetwork and is efficient to allow simulations of large scale neuronal networks. Therefore, in this work, we propose Nervous NaNoNetwork Simulator, *N⁴Sim*, to mitigate the issues present in the current simulators and assist in the ICT analysis of the nervous system. Since synaptic communication is one of the most complex processes in the design of any simulator, we describe an efficient way to solve this problem and make our simulator scalable.

6.4.2 *N⁴Sim Model*

In developing *N⁴Sim*, our main aim is to develop a tool that is most helpful to researchers in understanding the principles of nervous communication and synaptic function. To achieve this goal, *N⁴Sim* is envisioned as open-source. The simulator is to be designed such that it is modular in nature and different modules can work independently, thus, providing an opportunity for parallel processing. We want to ensure that the simulator is multi-scale in time and space such that it works not only

on single synapses but it is also possible to conduct large-scale simulations of a small cerebral region or the entire brain. The first step towards these goals is the selection of nervous simulation technique.

Simulation Algorithm

There are two families of algorithms for the simulation of neural networks: synchronous or ‘clock-driven’ algorithms, in which all neurons are updated simultaneously at every tick of a clock, and asynchronous or ‘event-driven’ algorithms, in which neurons are updated only when they receive or emit a spike. The plasticity of a synaptic connection is either event dependent, i.e. the weights increase or decrease as a result of action potentials from connected neurons, or decays over time if no event occurs. Although, the latter seems to be a continuous process, it is usually modeled by an exponential decay, which implies that the calculation depends only on the time between two events [83]. Therefore, an event-driven design is applicable with the advantage of reduced computational complexity for N^4Sim .

Programming Platforms

C++ is generally utilized in complicated protocol implementations for simulators due to its fast execution whereas higher level languages such as Python are used for simulation configuration as they can be modified very quickly. Therefore, the design of N^4Sim is object-oriented where major class definitions and libraries are to be written in C++ and simulation parameters are to be provided by a higher language shell. Users can write scripts creating objects which can then interact as matrices with predefined objects of the simulator.

Major Classes in N^4Sim

Three main classes of objects are planned for the network design in N^4Sim . These are the *Generator* class objects that are responsible to act as user inputs of a nervous network, a *NervousNet* class which includes neurons defined by the *NervousNode*

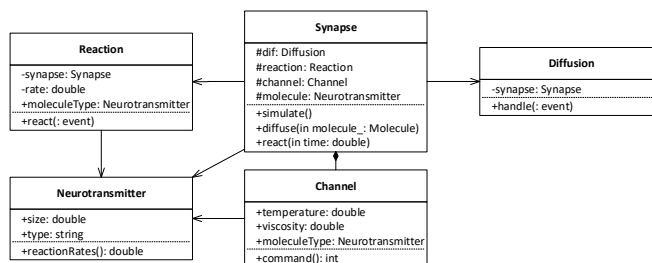


Figure 6.2: Class diagram of diffusive synapse.

class, and a *Probe* class to export outputs of the system to other formats such as MAT, CSV, TSV, NetCDF, XPORT or plain text format. A class diagram of a nervous network is shown in Figure 6.1. Using the attributes of frequency, pulse width and input duration, the *Generator* class generates input signals for the network. The topology of *NervousNode* connections within a *NervousNet*, the number of points where an input signal can be connected to a *NervousNet* and the positions of the outputs can be controlled by the user and defined as a connection map. Connection maps are matrices that contain a record of all connections across the network. Since it is not possible for users to write connection maps for large scale networks such as the human brain, we envision a *Randomizer* class that can generate random network topologies after receiving an input on the number of neurons in the system.

The *NervousNode* class is designed in order to simulate individual neuron nodes. Each *NervousNode* can have several associated *DendriteBunches* and an *Axon*. The *DendriteBunch* classes are further made up of *Dendrite* class, which depending on number of captured molecules and the geometry of the dendrite, generate a potential from each dendrite under the method *moleculeDetect*. This potential is summed and sent over to the *NerveNode* which decides to fire action potentials or not based on attributes such as the threshold. The axonal transmission depends on the geometry of the axon and its myelination.

The *Synapse* class defines the molecular channel of a chemical synapse. The axonal and dendritic communication is dependent on the synaptic communication.

The class diagram of a diffusive synapse is presented in Figure 6.2. The entire system is based on the *Neurotransmitter* class which defines the molecules to be used for the communication as well as the reaction scheme for those particular neurotransmitters. A *Synapse* can include several channels in it under the class *Channel* with different attributes for various neurotransmitter molecules. A *Reaction* class oversees the reactions in the *Synapse* whereas a *Diffusion* class performs diffusion. It should be noted that the methods *react()* and *handle()* are event based because of the type of simulator we selected.

Efficient Resource Management

Since we target network-level simulations with *N⁴Sim*, processing resources become an important parameter for our design. To tackle this, we plan to employ a multiple pronged approach where we will use parallel computing while also minimizing complex computations. It should be noted that users that utilize graphical processing units (GPU) for *N⁴Sim* may get additional benefits since GPUs outperform general purpose processors (GPP) because of their specialized architecture. The cores of a GPU communicate faster than those of a CPU, significantly reducing the communication overhead in the execution time. This is a desirable property when the calculations of different cores depend on each other, which typically is the case for neuronal networks.

We aim to use the open source OpenMP application programming interface (API) to implement parallelization. OpenMP provides a number of benefits that are essential to our vision for *N⁴Sim* such as incremental parallelism, automatic data layout and decomposition, scalability compared to other systems, and the property to handle both coarse-grained and fine-grained parallelism. Additionally, even if a sequential compiler is used by a user, no bugs will be introduced in the system because OpenMP directives are considered as comments in such cases.

To reduce complex computations in the simulator, the modeling of molecular communication in regions such as synapses requires special attention because of extensive mathematical calculations for molecular dynamics and diffusion processes. In this re-

gard, we envision to utilize libraries based on pre-existing look-up-tables (LUT) that are computed using efficient numerical analysis methods from simulations of individual system components. This will reduce cumbersome computations to simple array indexing operations saving a significant number of processing operations.

As discussed earlier, synaptic communication is the biggest bottleneck in terms of processing for the entire system. If an efficient method is available to model the synapse, input sets to different synapses could be exhausted to help us create LUTs of the outputs. A user of N^4Sim in that scenario does not need to simulate a particular type of synapse for which a LUT is already available and instead will pick up the corresponding output values directly from the table. For input values that don't exist in a LUT, desired output values can be calculated via interpolation on the existing ones. Therefore, in the next section we present a method to solve synapses efficiently by making use of synaptic geometry and effective boundary conditions. An efficient synapse calculation can assist in the design of a nervous simulator that does not ignore the molecular communication happening in the system.

6.4.3 Synapses in N^4Sim

In this section, we look at the mechanisms to deal with synapses in N^4Sim . We first provide a brief description of a deterministic algorithm, which calculates the average molecular dynamics in a chemical synapse such that LUTs for synapses can be created in an efficient manner and then describe the construction of LUTs for synapses.

Efficient Synaptic Simulation Algorithm

We work with a glutamatergic synapse, in which transmission of information is achieved via diffusion of glutamate molecules released from the presynaptic membrane. Receptor molecules, specifically α -amino-3-hydroxy-5methyl-isoxazolepropionic acid receptors (AMPA) and N -methyl-D-aspartate receptors (NMDARs), located in the postsynaptic density (PSD) on the postsynaptic membrane react with the diffusing glutamate molecules. This causes ion channels to open and facilitates a

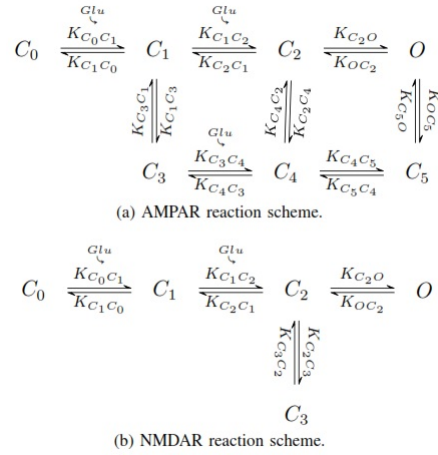


Figure 6.3: Reaction schemes for the assumed kinetic models.

consequent ion flow through the membrane, which is the input to the postsynaptic neuron.

The reaction dynamics of glutamate molecules with AMPARs and NMDARs, see Figure 6.3, are models derived from experimental data where AMPAR and NMDAR have 7 and 5 states, respectively [232, 233]. In both cases, C_O is the open state, i.e. ion channel opens, and C_0 is the initial unoccupied state. To reach the open state receptors must capture 2 glutamate molecules, during which they pass through states C_1 and C_2 . The states on the lower half of the reaction schemes are called the desensitized states. The reaction rate constants between the states are presented in TABLE I.

We assume a simple geometry for the synapse as shown in Figure 6.4. The synaptic cleft is modeled as a rectangular box, which is bounded by the pre- and postsynaptic membranes from its top and bottom. The cleft is open to the extracellular space from its sides.

The diffusion process inside the cleft is modeled by the celebrated diffusion equation

$$\partial_t u(x, t) = D \Delta u(x, t), \quad x \in \Omega, t > 0, \quad (6.1)$$

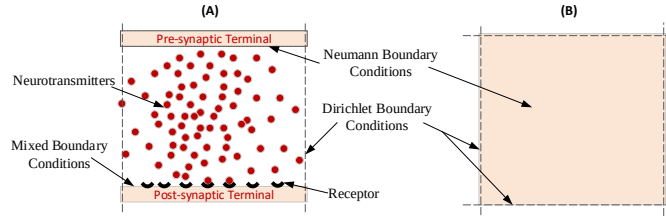


Figure 6.4: Synaptic geometry (A) Side view and (B) Top view.

where D is the diffusion rate constant, Ω corresponds to inside of the cleft, ∂_t stands for the partial derivative with respect to time, and u denotes the concentration of freely diffusing glutamate molecules. With the assumption that, molecules diffusing out of the cleft from its sides never return back to the cleft, the boundary conditions supplementing (6.1) become Dirichlet boundary conditions, i.e., $u = 0$, on the sides. This assumption is realistic because in the body these neurotransmitters are taken up by the nearby glial cells [83].

The boundary conditions on the presynaptic membrane are taken to be Neumann boundary conditions $\nabla u \cdot \vec{n} = 0$, where $\vec{n}$ is the outward normal unit vector, which corresponds to the case of reflective surface with no absorption. The boundary conditions on the postsynaptic membrane include the effects of receptor-ligand binding, and they correspond to a mixed type boundary condition. In principle, this boundary condition is obtained by first considering the boundary condition for a single receptor, and then averaging this condition to the whole postsynaptic membrane by the help of the distribution function, f_R , of the receptor. The resulting boundary condition on the postsynaptic membrane is

$$-D\nabla u \cdot \vec{n} = \sum_R \left(\sum_{ads} - \sum_{rel} \right) (A_R)_{ij} P_{R,j} f_R, \quad (6.2)$$

where, $P_{R,j}$ is the probability that the receptor R is in j^{th} state, the first summation is over the various receptors, and the other two are over adsorbing and releasing state transitions. Respectively, adsorbing and releasing state transitions correspond to state transitions in which the receptor R captures or releases a glutamate molecule,

indicated in Figure 6.3 by the Glu above the corresponding transition, with the direction of the curly arrow underneath indicating the adsorbing direction. For a given receptor R with r states, $(A_R)_{ij}$, $i, j = 1, \dots, r$, are entries of the $r \times r$ state transition matrix A_R , obtained from the reaction schemes in Figure 6.3. In essence, in case $i \neq j$, $(A_R)_{ij}$ quantifies the rate at which the receptor R transits from j^{th} -state to i^{th} -state, and it is in units s^{-1} . Thus, $(A_R)_{ij} = 0$ if there is no transition from j^{th} - to i^{th} -state. If there is one, then it is either the value given in TABLE I, or that value multiplied by the concentration near the receptor, where the latter is the case only for adsorbing transitions. Finally, $(A_R)_{ii}$, $i = 1, \dots, r$, quantifies the rate at which i^{th} -state transits to all other states, and is equal to the negative of the sum of all the corresponding transition states.

With the notation $P_R(t) := (P_{R,1}(t), \dots, P_{R,r}(t))^T$, the receptor state occupancy probabilities evolve according to the ordinary differential equation

$$\frac{d}{dt}P_R(t) = A_R(t)P_R(t), \quad t > 0. \quad (6.3)$$

We note that, for any possible state diagram, the corresponding state transition matrix with the above description always has columns that add up to zero. In view of (6.3), this fact is equivalent to the relation

$$\frac{d}{dt} \sum_i 1^r P_{R,i}(t) = 0,$$

and guarantees that the probability of finding the receptor in some state remains unity, as it should be, provided that it is initially so. Let us also note that, the dependency of the state transition matrix, A_R , on the concentration near the receptor and (6.3) show that the boundary condition (6.2) is a highly nonlinear one.

Our algorithm consists of implementing a finite difference discretization scheme on a rectangular grid to numerically approximate the solution of the diffusion equation (6.1) under the boundary conditions described above.

Synaptic LUT Construction

After simulating a synapse under some specified conditions using the algorithm described above, we receive the numerical values of receptors in various states with respect to time in the form of arrays. At any instance, the postsynaptic potential is directly proportional to the number of receptors in the open state and the amount charge flow per receptor per unit time. Since the charge flow is constant for a particular synapse type, the number of receptors in open state remains as the main variable for measuring postsynaptic potential and we aim to provide this variable in the LUTs of *N⁴Sim*.

The parameter space for synapse in *N⁴Sim* will include variables such as size of quantal release, receptor densities and their distributions, synapse dimensions and frequency of input signals etc. Each parameter will be defined a different dimension and the entire LUT will come together in the form multi-dimensional array structures. Searching in a large LUT becomes a very complex task as the size of a table increases. To avoid such a scenario, we aim to create the LUTs such that their indexes are meaningful. Therefore, the indexes and associated time will be related. Additionally, solutions to intermediate data points not covered by the discrete time index in the LUT may be computed using linear interpolation between the nearest time indexes.

6.4.4 Results and Validation

In order to validate our algorithm, we compared its predictions to the average of several results of an ensemble of Monte Carlo random walk and reaction experiments. Both approaches were run with the same nominal values of synaptic parameters presented in Table II. These values are in accordance with those of a real glutamatergic synapse [234]. The glutamate molecules are introduced into the cleft from the center of the presynaptic membrane instantaneously at time $t = 0$, and all receptors are initialized in the unoccupied state C_0 . We keep track of the evolution of the receptor states. The predictions of our algorithm and the average results of the Monte Carlo approach are overlaid in Figure 6.5 and 6.6 for comparison, and they show nearly an

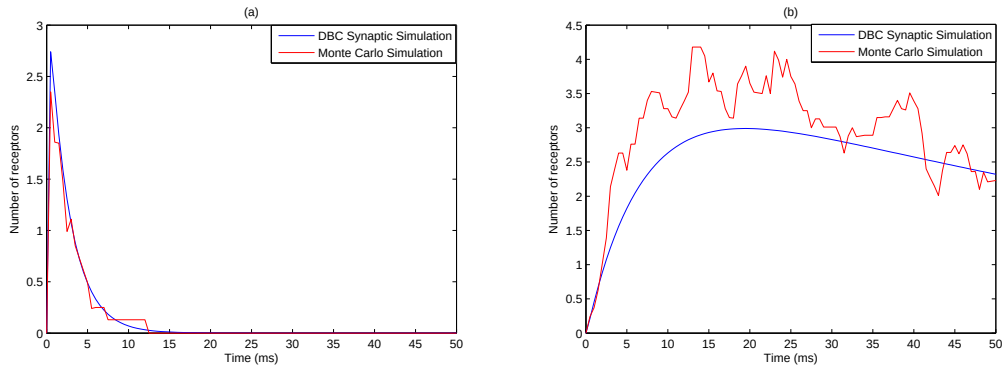


Figure 6.5: Timecourse of (A) AMPAR and (B) NMDAR open state occupancy. The results of our algorithm is compared to the average of 100 Monte Carlo simulations.

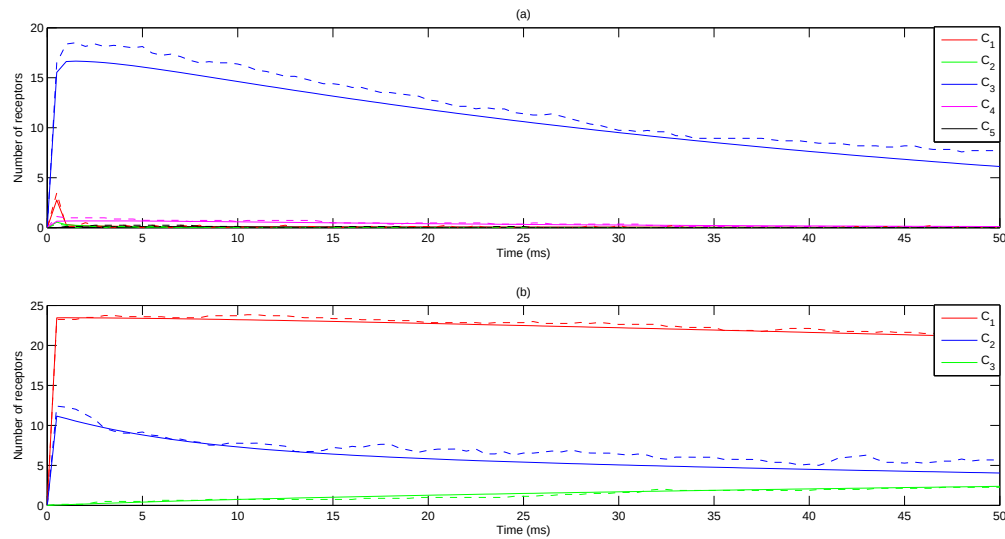


Figure 6.6: Timecourse of (A) AMPAR and (B) NMDAR state occupancies. The results of our algorithm (solid lines) and the average of 100 Monte Carlo simulations (dotted lines) are overlaid for comparison.

exact match for all the states. Also note that the results of 100 Monte Carlo simulations have not smoothed for several cases where even further simulation runs are required to reach a true mean value.

However, when we compare the performance of our approach with the Monte Carlo approach, we see that our method requires significantly less computational resources. It should note that for Monte Carlo approach requires multiple experiments to run over the scenario and an overall result is the average of all the single experiments. For our experiments, a single Monte Carlo experiment running on $2.26GHz$ *AMD64* machine requires 9.14 hours on average. In comparison, using the presented approach without any parallelization on the same machine, the calculation lasted only 1.46 Hours on average. Now consider that n experiments are needed for a Monte Carlo approach result, our results requires $6.48 \times n$ times less time for the equivalent result. In pursuit of determining the input-output relations of various synapses, this improvement is crucial for the exhaustion of the parameter space to be possible. This, we believe, is a necessary step to achieve a simulator, which is not only fast, but also realistic in terms of molecular communication dynamics.

6.4.5 Future Work

After initially creating a set of libraries, a graphical user interface (GUI) will also be constructed to easily construct simulations and conditions. While the GUI will be open source as well, it will employ complete error handling so that the software does not hang or give processing errors. Apart from import and export options, the GUI will also provide some graphical windows where the operation of the network can be observed or some graphs can be plotted.

Additionally, a website will also be launched to initially provide detailed user manuals and examples of scenarios modeled by the software. User forums will be created so that users can discuss their problems, experiences and solutions to various scenarios. A page for users to post and share their designs will also be created. As the open source software grows, newer versions will be periodically released to improve the software and remove any bugs that remain. Assisted by other users of *N⁴Sim*, the library for LUTs of synaptic data can keep growing, thus, keeping the simulator up-to-date with novel scenarios.

Table 6.1: Rate constants for the kinetic models.

Receptor	Symbol	Value
<i>AMPAR</i> [232]		
	$K_{C_0C_1}$	$4.59 \times 10^6 \text{M}^{-1}\text{s}^{-1}$
	$K_{C_1C_0}$	$4.26 \times 10^3 \text{s}^{-1}$
	$K_{C_1C_2}$	$2.84 \times 10^3 \text{M}^{-1}\text{s}^{-1}$
	$K_{C_2C_1}$	$3.26 \times 10^3 \text{s}^{-1}$
	K_{C_2O}	$4.24 \times 10^3 \text{s}^{-1}$
	K_{OC_2}	900s^{-1}
	$K_{C_1C_3}$	$2.89 \times 10^3 \text{s}^{-1}$
	$K_{C_3C_1}$	39.2s^{-1}
	$K_{C_3C_4}$	$1.27 \times 10^6 \text{M}^{-1}\text{s}^{-1}$
	$K_{C_4C_3}$	45.7s^{-1}
	$K_{C_2C_4}$	172s^{-1}
	$K_{C_4C_2}$	0.727s^{-1}
	$K_{C_4C_5}$	16.8s^{-1}
	$K_{C_5C_4}$	190.4s^{-1}
	K_{OC_5}	17.7s^{-1}
	K_{C_5O}	4.0s^{-1}
<i>NMDAR</i> [233]		
	$K_{C_0C_1}$	$2 \times 5.0 \times 10^6 \text{M}^{-1}\text{s}^{-1}$
	$K_{C_1C_0}$	4.7s^{-1}
	$K_{C_1C_2}$	$5.0 \times 10^6 \text{M}^{-1}\text{s}^{-1}$
	$K_{C_2C_1}$	$2 \times 4.7 \text{s}^{-1}$
	K_{C_2O}	46.5s^{-1}
	K_{OC_2}	91.6s^{-1}
	$K_{C_2C_3}$	8.4s^{-1}
	$K_{C_3C_2}$	1.8s^{-1}

Table 6.2: Nominal values of synaptic parameters.

Parameter	Value
<i>Base area of synapse</i>	$0.5 \times 0.5 \mu\text{m}^2$
<i>Height of synaptic cleft</i>	20 nm
<i>Diameter of the PSD</i>	350 nm
<i>Number of AMPARs</i>	80
<i>Number of NMDARs</i>	20
<i>Quantal size</i>	3000
<i>Diffusion rate constant, D</i>	$0.2 \mu\text{m}^2/\text{ms}$

Chapter 7

CONCLUSION

The main motivation of the proposed thesis was to analyze the nervous system and the communication happening within this network from an ICT perspective and its potential applications in IoBNT. To this end, the thesis investigated the following three main areas:

- *Modeling of the nervous system from a ICT perspective* - Investigation of neuron networks based on queueing theory and the analysis of nervous circuits for reflex arcs was done. These were used to develop a simple and efficient delay measurement system.
- *Experimental testbeds for molecular communication based on nervous system* - This first-of-a-kind testbed can be used to help shed light on the underlying nervous coding in such systems.
- *Insulin-glucose system as a peripheral systems of the nervous system* - ICT-based modeling of the insulin-glucose system was investigated. The link between the nervous system and the homeostasis control of glucose is an interesting future direction.
- *Nervous Nanonetwork Simulator* - Based on the experimental data and the developed models, we created a dedicated simulator for molecular communication in the nervous system.

The study of the nervous system will provide novel directions to augment the existing diagnostic and treatment techniques as well as provide a framework for artificial nanonetworks that may be based on the nervous system.

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