SYNOPSIS

In the pursuit to understand information processing in the brain, a general question that is often posed is: What is the neuron *encoding*? This is addressed by assessing *feature selectivity* in sensory neurons, which informs us of the specific features of the stimulus that the neuron is selectively responding to. In this respect, a quantity that has been widely used is the *spike triggered average* (STA), an unbiased estimate of the *spike-triggering features* in the stimulus. From the single neuron perspective, the STA provides us with a direct link connecting the biophysical properties of the neuron and its response dynamics in a network with its *encoding* schema. While this approach has been used extensively for sensory modalities, it is a slightly more complex problem for multimodal areas such as the hippocampus where the information itself is in the form of complex spatiotemporal pattern of synaptic inputs.

The hippocampus is a brain region that has been implicated in navigation, learning and encoding of several forms of context-dependent and episodic memory. Pyramidal neurons in the CA1 subfield of the hippocampus receive synaptic inputs from the entorhinal cortex and the CA3 pyramidal neurons. These inputs are spatially segregated on the dendritic arbour of CA1 pyramidal neurons as well as containing information in spectrally parsed streams. Specifically, inputs from the entorhinal cortex exhibit high power in the fast gamma frequency range (60–100 Hz), whereas inputs from the CA3 exhibit high power in the slow gamma frequency range (30–60 Hz). The spectral segregation of inputs has important ramifications for encoding of specific, behaviourally relevant information about the space and contextual cues in an ambulatory animal. Moreover, these neurons exhibit a form of multiplexed coding where the firing rate while expressing coherence with different gamma frequency bands also demonstrate phase-locking with an ongoing slower oscillation in the theta (4–10 Hz) frequency range. Research spanning the past two decades has resulted in extensive literature on the active and passive properties of the CA1 pyramidal neuronal dendrites, their somato-dendritic transfer properties and the physiological roles of the heterogeneous distribution of voltage-gated ion channels along their somato-dendritic arbour. In addition, location-dependence of several physiological properties including subthreshold theta resonance and phase preference in these neurons, as well as activity-dependent plasticity in ion channel expression profiles

and subsequent plasticity in their excitability and impedance characteristics have also been established in these neurons. Juxtaposed against this extensive literature on active dendritic physiology and plasticity with specific reference to hippocampal pyramidal neurons, we asked the following questions.

1. What feature selectivity does the spike initiation dynamics of CA1 pyramidal neurons exhibit? Specifically, these neurons are known to exhibit theta-frequency *subthreshold* resonance. But does this subthreshold resonance also translate to spectral selectivity in the suprathreshold regime manifesting as theta-frequency selectivity in their spike initiation dynamics?
2. Given the intraneuronal gradients in various ion channel properties and in physiological properties regulated by these ion channels, is there also a location-dependent gradient, or an intraneuronal functional map, in the spike initiation dynamics of these neurons?
3. Do CA1 pyramidal neurons exhibit coincidence detection of excitatory synaptic inputs in the gamma frequency range? Given the spatial segregation of the spectrally parsed afferent inputs along the CA1 apical dendrites, is the coincidence detection window expressed by these neurons equipped to detect these differences in afferent frequencies in a location-dependent manner?
4. How do *plastic active* dendrites regulate location dependent spike initiation dynamics and coincidence detection windows? How do active properties of the dendrites and plasticity therein alter the location dependence of spike initiation dynamics?
5. How do the somato-dendritic ion channels regulate the specific *spike-triggering features* in CA1 pyramidal neurons? How do individual channels and interactions among them alter the location dependence of spike initiation dynamics and coincidence detection windows?
6. Are there correlations between the frequency selectivity in spike initiation dynamics and subthreshold resonance frequency? Under what conditions do these correlations hold and when does the correlation disintegrate? Are there specific ion channels that mediate correlations between the frequency selectivity in the sub- and supra-threshold regimes?

In the pursuit to answer these questions, we employed a combination of computational and electrophysiological methods to assess the spike initiation

dynamics and coincidence detection windows in CA1 pyramidal neurons using the STA and various STA-derived quantitative metrics that we developed as part of this thesis.

As a first step, we employed a single compartmental model of a CA1 pyramidal neuron with only the spiking conductances (transient sodium and delayed rectifier potassium channels) and the hyperpolarization-activated cyclic nucleotide-gated (HCN) nonspecific cation channel, an established mediator of subthreshold resonance (f_R) in these neurons. We injected zero-mean Gaussian white noise (GWN) current with a fixed standard deviation adjusted to elicit ~1–2 Hz average firing. We measured the STA as the average current in a 1 s time window preceding a spike computed over ~1000 spikes obtained in response to the GWN current. We computed the STA for various densities of HCN channels and observed that the STA transitions from class I in the absence of HCN channels to class II in their presence. This was evidenced by an increase in the depth of the negative lobe as well as a progressive sharpening/narrowing of the spike-proximal positive lobe (SPPL) with increasing density of HCN channels.

To systematically quantify these changes, we developed specific metrics based on the shape of the STA. First, the peak of the positive lobe of the STA was quantified and was found to increase with increasing HCN conductance suggesting an inverse relationship between this peak and the excitability of the neuron. Next, we performed Fourier transform on the STA, which revealed spectral selectivity in the STA defined by a distinct band-pass structure in the frequency domain. We quantified the frequency at which the |STA(f)| reached its maximum as the STA characteristic frequency (f_{STA}) and found this to be in the theta frequency range in the presence of HCN channels, with increase in the f_{STA} on increasing HCN conductance. Furthermore, the strength of this frequency selectivity (Q_{STA}), quantified as the ratio |STA(f)|/ |STA(0.5 Hz)| also increased with HCN density.

We reasoned that the SPPL of the STA reflects the temporal window over which the neuron integrates coincident inputs and so quantified the total duration of the SPPL as the total coincidence detection window (T_{TCDW}). Additionally, to account for the shape of the STA that underwent a change on altering HCN conductance, we computed the effective coincidence detection window (T_{ECDW}) as the STA-weighted measure of the SPPL. Both the total and effective CDW underwent a reduction on increasing the HCN channel density and the effective coincidence detection window

was in the gamma frequency range. Together, these results demonstrated that the HCN channel alone was sufficient to confer coincidence detector properties on the neuron in the gamma frequency range as well as theta frequency selectivity in the spike initiation dynamics. This also implied that graded expression of HCN channels was sufficient to effectuate a transition in the STA along the integrator-coincidence detector (*I-CD*) continuum. We confirmed that such a transition could not be elicited simply by altering the passive properties of the membrane, in particular the leak conductance, and was specifically mediated by HCN channels.

Further analysis in the single compartmental model suggested that the f_{STA} was correlated to the f_R in the presence of HCN channels. However, in their absence, there was delta frequency selectivity in the STA, which was critically dependent upon the density and kinetics of the spiking conductances. Importantly, although subthreshold resonance was completely abolished in the absence of HCN channels, the transient sodium channels and delayed rectifier potassium channels mediated delta frequency selectivity in the STA, thereby providing lines of evidence on a dissociation between subthreshold resonance and STA spectral selectivity.

Next, to assess location dependence of the STA, we built models of increasing complexity from a ball-and-stick model to a morphologically realistic model and measured the STA (with reference to somatic spike timings) by injecting GWN at various locations along the dendrite. We imposed a gradient of HCN channels on these models that matched electrophysiological data on gradients in input resistance and resonance frequency and assessed the distance-dependent variation in STA measurements. We found that in the ball-and-stick model with a non-spiking dendrite, the f_{STA} was normalized with distance from soma and global plasticity in HCN channel density altered f_{STA} values across locations in a distance-invariant manner. Introduction of spike-generating conductances into the dendritic compartment resulted in enhancements of f_{STA} and Q_{STA}, with distance-invariance perturbed at distal dendritic locations owing to dendritic spike initiation. A morphologically realistic model, on the other hand, exhibited a clear functional map in the STA with both f_{STA} and Q_{STA} increasing with distance, suggesting that the STA was dependent on the location of inputs along the somato-dendritic axis.

Thus far, our focus was on the HCN channel and its interactions with the spike generating conductances in regulating STA measurements. In assessing the impact of other channels that are expressed by CA1 pyramidal neurons, we first employed

single compartmental models to study the effect of kinetic interactions between these ion channels. These analyses revealed that the co-presence of another resonating conductance, the *T*-type calcium (CaT) channel, further increased the f_{STA} and Q_{STA} in the presence of HCN channels while also reducing the CDW measures. However, the correlation between the f_{STA} and f_R was reduced in the presence of CaT channels. On the other hand, the co-presence of HCN channels with a subthreshold restorative conductance, mediated by *A*-type potassium channels, reduced the f_{STA} and Q_{STA} but broadened the CDW. A regenerative conductance, the persistent sodium channel increased the Q_{STA} significantly without significantly altering f_{STA}. These observations clearly dissected the differential effects of various ion channels and interactions therein on STA and CDWs. They also demonstrated that a clear correlation between sub- and supra-threshold frequency selectivity existed only when HCN channels were the sole subthreshold channels present in the model. However, when multiple ion channels came into play or when HCN channels were absent, a clear dissociation between these forms of selectivity was observed.

To assess the role of active plastic dendrites spanning several somatodendritic ion channels, we incorporated gradients in HCN, CaT and KA into a morphologically realistic model and tuned several measurements to match electrophysiological data. In this model, we observed the emergence of location dependent theta frequency selectivity in STA showing strong class II characteristics. Importantly, a gradient in CDW measures was also observed, with the effective CDW decreasing from slow gamma range in the proximal dendrites to fast gamma range in the distal dendrites. Removal of all active conductances resulted in a transition to class I STA with significant broadening of the integration window, and the presence of uniform resonating conductances in the dendrites resulted in class I STA with a narrow integration window. These results demonstrated the emergence of location-dependent theta frequency selectivity in the STA and the presence of stratified gamma-range CDW that is essential for detecting frequency-multiplexed inputs afferent onto different regions of the dendritic arbor. These observations also emphasized the importance of gradients in ion channels in maintaining functional maps of spike initiation dynamics and CDW, and suggested that local or global plasticity in any or all of these ion channels would alter feature selectivity and coincidence detection in hippocampal pyramidal neurons.

We finally tested our computational predictions using acute rodent hippocampal slices and performed electrophysiological measurements of somatic STA from CA1 pyramidal neurons. Our primary goals were to (i) confirm theta frequency selectivity and gamma-range CDW in the STA of CA1 pyramidal neurons as predicted from our models; (ii) test the quantitative prediction that the blockade of HCN channels would reduce f_{STA} from theta- to delta-frequency ranges; and (iii) assess the efficacy of the novel STA-derived metrics against other physiological measurements of excitability and impedance profiles from the same neurons. Our results unveiled theta-frequency selectivity in the STA of hippocampal pyramidal neuron somata, coupled with gamma-range CDW, thereby validating our computational predictions. Furthermore, we confirmed that the correlation between the f_{STA} and f_R was weak across cells, corroborating our model prediction on the dissociation between the two forms of spectral selectivity in the presence of multiple ion channels. We observed strong correlations between the peak STA current and excitability measures such as input resistance and impedance amplitude, as well as strong correlations between the CDW measures and f_R suggesting that neurons resonant at higher frequencies could detect coincident inputs at higher frequencies. We also demonstrated the dependence of the STA measurements on the membrane potential with hyperpolarization increasing the f_{STA} akin to increase in HCN-dependent f_R, but the Q_{STA} increased on depolarization. Our results also revealed the adaptability of the somatic STA and its quantitative measurements to the input statistics. Finally, we pharmacologically tested the impact of blocking HCN channels on somatic STA of identified hippocampal pyramidal neurons. We found, consistent with our model predictions, that f_{STA} decreased from theta frequency to delta frequency upon blockade of HCN channels, although subthreshold resonance was completely abolished with HCN-channel blockade. Our electrophysiological analyses allowed us to confirm many of our model predictions, apart from demonstrating HCN-channel dependent theta-frequency selectivity in spike initiation dynamics and gamma-range coincidence detection windows in CA1 pyramidal neurons.

Together, the results of our computational and electrophysiological studies unveiled the critical role of several voltage-gated ion channels in regulating spectral selectivity in spike initiation dynamics as well as in mediating sharp coincidence detection windows. Our computational analyses expounded the role of plastic active dendrites in mediating a functional map in the STA in CA1 pyramidal neurons, with

multiple, degenerate mechanisms involving interactions between ion-channels and a state-dependent modulation of the STA. These results identified explicit roles for plastic active dendrites in neural coding and strongly recommend a dynamically reconfigurable multi-STA model to characterize location-dependent input feature selectivity in pyramidal neurons. Importantly, they also showed that the presence of resonating and spike-generating conductances serve as a mechanism underlying the emergence of stratified gamma-range coincidence detection in the dendrites of CA1 pyramidal neurons, enabling them to perform behaviour- and state-dependent gamma-frequency multiplexing. Our electrophysiological experiments confirmed many of our model predictions including the critical role of HCN channels in mediating theta-frequency selectivity and paved the way for future studies involving dendritic ion channels and plasticity therein.

The thesis is organized into a total of eight chapters with Chapter 1 providing a general introduction and laying the motivations for the thesis. Chapter 2 is an overview of literature on the physiology of the hippocampus that served as the cornerstone for the pursuits of this thesis. Chapter 3 is a brief discourse on the theoretical principles and computational models of single neurons that were paramount to synthesizing this thesis. Chapter 4 is the first of the results chapters and discusses the various STA-derived metrics that were used to quantify spectral selectivity and coincidence detection windows in the STA of model hippocampal pyramidal neurons along with dissecting the specific role of resonating conductances in mediating these forms of feature selectivity. Chapter 5 explores the role of dendrites in mediating location dependence in the STA, focusing specifically on theta frequency selectivity in STA and interactions between HCN channels and the spiking conductances. The chapter also establishes specific correlations and dissociations between sub- and supra-threshold theta frequency selectivity. Chapter 6 first delves into the kinetic interactions between various dendritically expressed subthreshold conductances in a single-compartmental model. The latter part of this chapter assesses spatio-temporal interactions between these conductances in a morphologically precise model towards concerted regulation of location-dependent STA and coincidence detection windows. Chapter 7 presents the results of electrophysiological experiments on rodent acute hippocampal slices, demonstrating theta frequency selectivity and gamma-range coincidence detection window in the somatic STA of CA1 pyramidal neurons. The chapter also confirms computational predictions on the reduction of

STA frequency selectivity from theta to delta ranges upon pharmacological blockade of HCN channels. Finally, Chapter 8 presents the broad implications of results presented here and posit some future directions stemming from this thesis.