PROJECT SUMMARY

The mammalian brain accounts for complex sensory, motor, and cognitive abilities by processing environmental and internal information within neuronal networks. A fundamental property of neuronal networks is synaptic plasticity, a process under which the strength of synapses between the elements of a network, and thereby the functional properties of that network, can change over time. Although much is known about plasticity at chemical synapses, recent research has revealed that gap junctional (electrical) synapses exhibit a high degree of plasticity on several time scales, making critical contributions to sensory adaptation and learning. Several recent studies, in particular in the retina and hypothalamus, have highlighted the control of this plasticity by circadian clocks, a type of endogenous oscillator with a period close to 24 hrs. However, there is a fundamental gap in understanding exactly how circadian clocks control gap junctions and neuronal network activity. Continued existence of this gap represents an important problem because, until it is filled, understanding of the mechanisms that link clock malfunction and many behavior disorders will remain largely incomplete. Our long-term goal is to establish circadian plasticity and its functional consequences in a specific neuronal network: the network of photoreceptors in the retina, a tractable model for the rest of the brain. The objective of this project is to determine how a circadian clock within photoreceptors controls electrical coupling and signal processing in the photoreceptor network during the day and night. Our central hypothesis is that a circadian clock in the cones—and not in the rods—uses a pathway that includes melatonin and dopamine to control photoreceptor gap junctional coupling. Circadian control of the photoreceptor light responses results from the modulation of electrical coupling but also from intracellular messengers that are yet to be identified and their diffusion within the cones or through the gap junction channels from cones to rods. Our central hypothesis has been formulated on the basis of our own preliminary data and recent publications in the field. The rationale for the proposed research is that by genetically silencing the clock mechanism in the cones, we will be able to link this specific clock cell to clock pathways associated with photoreceptor behavior. We will pursue three specific aims: 1) Identify candidate genes and signaling pathways under the control of the cone clock; 2) determine the effects of the cone clock and its putative effectors on photoreceptor electrical coupling and/or dynamic behavior; and 3) establish a computational model of the network of photoreceptors in mouse retina and simulate the actions of the clock. Under the first aim, we will combine single cell sorting of cones with RNA sequencing and analysis to identify genes whose expressions differ between mutant and control mice and that will be candidates for the circadian control of photoreceptor function. Under the second aim, a novel electrophysiological technique—perforated patch-clamp recording of photoreceptors in mouse retina—will be combined with pharmacological and genetic approaches to reveal a cone-specific clock pathway. Under the third aim, we will develop a comprehensive Resistive-Capacitive-Inductance (RCL) model of the mouse photoreceptor array. We expect to identify the signaling pathway(s) that links the cone clock and the plasticity and behavior of the photoreceptor network. The proposed research is significant because it is expected to vertically advance and expand understanding of how circadian clocks control photoreceptor network dynamics and function. Ultimately, such knowledge has the potential to increase our understanding of the general rules governing the activity of neuronal circuits in the CNS and of the events leading to their malfunction and subsequent behavior disorders.

RELEVANCE

The proposed research is fully in line with the objectives of the 2015 UT BRAIN Seed Grant Program: it is an interdisciplinary and seminal collaborative endeavor that aims at capitalizing on recent technological and analytical progress made in the PI’s, co-PIs’ and co-I’s laboratories for elucidating the relationship between neurons, networks, and behavior. Quantitative analysis of the cone transcriptome will include generating a transcriptome database for cones by RNA sequencing and using a sensitive algorithm to detect alternative splicing events and regulatory non-coding RNAs. These techniques are routinely performed in the laboratory of Dr. Wu (co-PI). Patch-clamp recording of single or pairs of mouse photoreceptors is routinely performed in the laboratory of Dr. Ribelayga (PI). Finally, Dr. Masson (co-I) is an expert in mathematical modeling and simulations of RCL circuits and will bring his expertise to building a model of the mouse photoreceptor network. We intend to submit a BRAIN Initiative R01 application sometime in spring 2016. Besides the scientific significance of this project, the complementary expertise each of us brings to it is a strength. If funded by a UT BRAIN Seed Grant, the preliminary data that we will gather will further strengthen this proposal and increase our chances to secure funding from the NIH.