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Exploring Spatio-temporal Organization of SCN Circuits

The daily rhythms of mammalian behavior and physiology are regulated by an internal clock located in an area of the brain known as the suprachiasmatic nucleus (SCN). In the past, the SCN was viewed as a single unit of weakly coupled circadian oscillator cells which synchronized each other through spontaneous self-organization. More recent models present the SCN as structurally differentiated into two distinct regions: the "shell" of oscillating cells, and the "core" of non-oscillating cells through which the nucleus receives light signals. The stability of the cellular signals is highly dependent on the presence of the core region; the interaction between the core and shell is important in sustaining many different types of behaviors, including entrainment, phase resetting in response to light signals, free-running rhythms, and the recovery of rhythms after hibernation.

This study seeks to explore the structural and functional relationships within the SCN by tracing spatial and temporal patterns in PER2 gene expression in explant tissue slices from the coronal *in vitro* slice preparations of wild type and knockout mice. The resulting datasets consist of image stacks displaying the distribution of PER2 expression in the nucleus over periods of circadian time. Using ImageJ, PER2 gene expression was then measured by calculating the average grayscale pixels in dorsal to ventral and medial to lateral subregions, giving quantitative bioluminescence values. Graphing these values over time indicates that the activity of the oscillator cells depends on their locations within the nucleus, presumably due to their functional circuitry connections with surrounding cells. In slices containing both the core and the shell, rhythmic activity was evident in all regions, whereas in coreless slices, bioluminescence activity was less synchronous. We conclude that the core plays a crucial role in organizing the patterns of oscillations in the SCN tissues.

Another statistical model incorporates a Mathematica program to identify physiologically significant subregions based on deciles of pixel brightness across the SCN images. This program produces graphs and subplots based on the spatial progression of activation in SCN regions over time. Analysis has revealed that the SCN has a tidal pattern of activation, where regions activate and deactivate in opposite orders, rather than a wave pattern, where activation and deactivation retains the same chronology. The results of these *in vitro* slices will later be compared to the spatiotemporal rhythms of *in vivo* slices to assess how accurately the *in vitro* slices report the temporal changes in the living animal. These experiments are significant in understanding the SCN's contributions to circadian mechanisms by exploring the broader range of oscillatory and rhythmic phenomena in biological systems.