Circadian clocks regulate the day-night biological cycle in living organisms, including plants, animals, fungi, and some bacteria. Mammalia have similar endogenous circadian gene regulatory networks located in the suprachiasmatic nuclei (SCN) of the anterior hypothalamus, so the study of their clocks is generally conducted as a class. Henry Minsky (2009) developed a mathematical model of Mus musculus single-cell rhythms by describing 21 biological species with 132 parameters in a system of ordinary differential equations (ODEs), using standard biochemical equations: Michaelis-Menten, Hill, and mass action enzyme kinetics. The model consists of two regulatory loops, which we name the primary positive feedback loop and the ancillary negative feedback loop. We perturbed the model in MATLAB (MathWorks, Inc) in separate sensitivity and bifurcation analyses. We found that the ancillary loops relatively insensitive to parameter perturbation, while the primary loop is highly sensitive to perturbations. We also confirmed that parameters related to cellular function, such as gene transcription rates, were more sensitive than those exclusive to the model, such as protein-specific rates of dimerization. These data suggest that the positive regulatory loop is primarily responsible for maintaining oscillations in vivo and that the negative regulatory loop confers non-robustness-related characteristics to the model. Further research will perform similar analysis on multi-cellular tissue models whose parameter vectors are currently being discovered.

**Abstract**

Circadian clocks regulate the day-night biological cycle in living organisms, including plants, animals, fungi, and some bacteria. Mammalia have similar endogenous circadian gene regulatory networks located in the suprachiasmatic nuclei (SCN) of the anterior hypothalamus, so the study of their clocks is generally conducted as a class. Henry Minsky (2009) developed a mathematical model of Mus musculus single-cell rhythms by describing 21 biological species with 132 parameters in a system of ordinary differential equations (ODEs), using standard biochemical equations: Michaelis-Menten, Hill, and mass action enzyme kinetics. The model consists of two regulatory loops, which we name the primary positive feedback loop and the ancillary negative feedback loop. We perturbed the model in MATLAB (MathWorks, Inc) in separate sensitivity and bifurcation analyses. We found that the ancillary loops relatively insensitive to parameter perturbation, while the primary loop is highly sensitive to perturbations. We also confirmed that parameters related to cellular function, such as gene transcription rates, were more sensitive than those exclusive to the model, such as protein-specific rates of dimerization. These data suggest that the positive regulatory loop is primarily responsible for maintaining oscillations in vivo and that the negative regulatory loop confers non-robustness-related characteristics to the model. Further research will perform similar analysis on multi-cellular tissue models whose parameter vectors are currently being discovered.

**Perturbation Analyses**

**Sensitivity Analysis**

- Uses single-parameter, percent-based perturbations to compare state values of the system when running nominal versus perturbed parameter vectors

**Bifurcation Analysis**

- Uses larger parameter perturbations to discover the stability of the "bifurcation point" (i.e. state change) in system behavior

**Systems Biology**

**What we do**

- Model biological systems using data collected from cultured cells and tissue samples
- Work in collaboration with scientists from other disciplines to gain a comprehensive understanding of biology

**How we do it**

- Propose and test enzymatic rate velocities defined using standard biochemical equations: Michaelis-Menten, Hill, and mass action kinetics
- Optimize parameters of enzymatic rate velocities to fit observed data using iterative algorithms based on nature’s innovation of evolution

**Why This Model?**

Single-cell vs. multi-cell oscillators: In SCN tissue, many oscillating cells are sending and receiving signals. These signals allow cells to oscillate in synchrony and with a high amplitude. When cells are isolated from the network, their oscillations drift out of phase and lose amplitude (some become damped, and some even become arrhythmic). Knockout experiments (e.g. cry1-/-) show that intercellular signaling should confer robustness to the system (without communication there are no oscillations, but with communication there are synchronized oscillations).

**Results**

Our single cell model nearly reproduces these data. However, our related multi-cell model fails. We want to know why. We investigate this through parameter-based sensitivity and bifurcation analyses.

**Conclusions**

Mean parameter sensitivity data do not show correlations or clear clustering. Groupings designed to cluster by parameter type also do not show strong correlation of sensitivity. From this, we conclude one of two possibilities:

(a) The system is too robust to parameter perturbations using the current parameter set, or
(b) We can not analyze the model using the parameter grouping ideas of Stelling et al (2004).

**Future Goals**

Our next goal is to refit parameters so that wild type model is less robust (i.e. it is easy to get a population of damped/arrhythmic/sustained oscillators) and so that the Cry1 knockout is damped, rather than arrhythmic. This should show proper multi-cell entrainment behavior.

**References**


**Acknowledgements**

The authors would like to thank their advisor and professor, Stephanie Taylor, for teaching them the ins-and-outs of Systems Biology and for giving them an opportunity to do this research. They would also like to thank the tireless workings of Randall Downer for assistance with cluster operations. Finally, they would like to acknowledge their classmates in CS441 who have given feedback on their continued project.