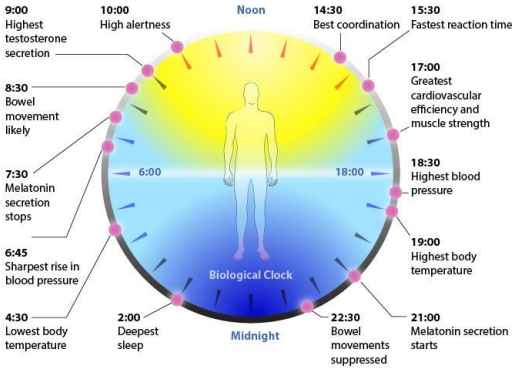
BCS410. Practical Class 2 & Homework 2 [Due: April 07]

Caution: You must submit the code you wrote and used to solve each problem. When submitting, name the folder with your student ID and submit each problem's code file as 'ProblemX_sol' (e.g., 'Problem1_sol'). You are free to use Python libraries. However, you are not allowed to ask AI to generate the entire code for you. If detected, you will receive a score of zero. You must provide your answers in a descriptive format for each problem.

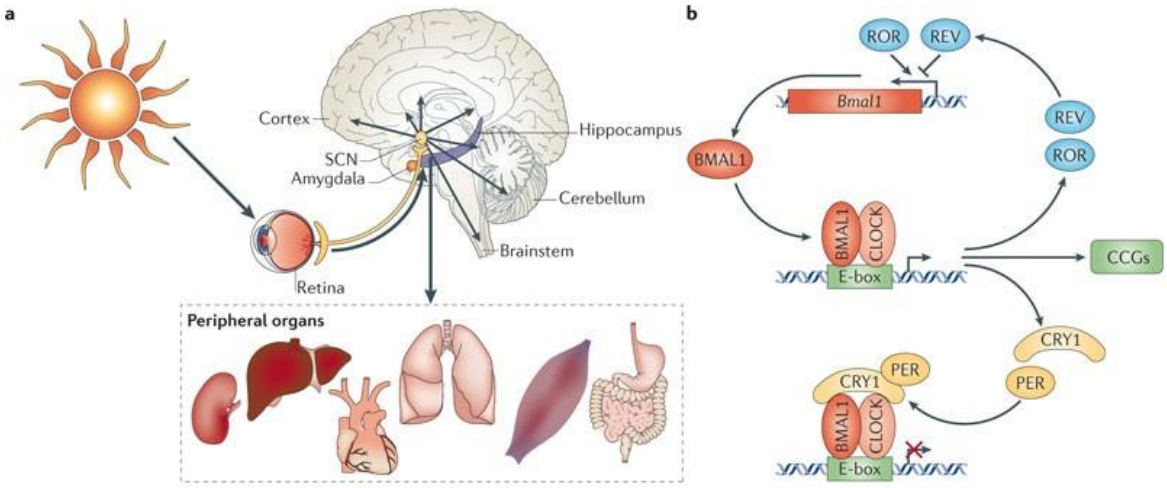
Background:

1. Overview of Circadian Rhythms: Circadian rhythms are endogenous biological rhythms that follow an approximately 24-hour cycle and regulate various physiological and behavioral processes in mammals. These rhythms are influenced by external cues, primarily the light-dark cycle, and are controlled by an internal timekeeping system known as the circadian clock. The term "circadian" originates from the Latin words circa (around) and diem (day), reflecting their daily rhythmicity.



2. The Suprachiasmatic Nucleus (SCN) as the Central Clock: The suprachiasmatic nucleus (SCN) located in the anterior part of the hypothalamus serves as the central circadian clock in mammals. This small but critical structure, located above the optic chiasm, synchronizes peripheral clocks in different tissues and organs. The SCN receives direct input from the retina via the retinohypothalamic tract, enabling it to adjust to environmental light conditions. The SCN maintains circadian rhythms through a network of oscillatory neurons that communicate via neurotransmitters (e.g., GABA, glutamate) and neuropeptides (e.g., vasoactive intestinal peptide, VIP). Even in the absence of external light cues, the SCN can sustain rhythmic activity due to its endogenous oscillatory properties.

3. Molecular Mechanisms of Circadian Rhythms: The molecular basis of circadian rhythms is governed by a transcription-translation feedback loop (TTFL), which involves a set of core clock genes and proteins that interact in a self-sustained cycle. The primary components include: ***1. Positive Limb***: CLOCK and BMAL1 form a heterodimer that binds to E-box sequences in the promoter regions of target genes, activating transcription. ***2. Negative Limb***: PER (Period) and CRY (Cryptochrome) proteins accumulate in the cytoplasm, then translocate to the nucleus to inhibit CLOCK-BMAL1 activity, thereby closing the loop. ***3. Regulatory Feedback****:* Other factors like REV-ERBα/β and ROR proteins help fine-tune the cycle by modulating BMAL1 expression.



4. A simple mathematical model of a genetic circadian clock: A simple genetic circadian clock model based was developed to understand intracellular circadian oscillation of clock molecules. It consists of five variables, M, D_A, P, F, and D_R. Specifically, the transcription of mRNA (M) is proportional to the concentration of DNA promoter sites that are free of the repressor protein (D_A). The mRNA is translated into cytoplasmic protein (P). The cytoplasmic protein enters into the nucleus, i.e., the free repressor protein in the nucleus (F) is produced at a rate proportional to the concentration of P. The free repressor can bind to a promoter site and change the DNA to its repressed state (D_R). All species, except for DNA, are subject to degradation (β_i), with the bound and free repressors degrading at the same rate. Note that total DNA concentration ($D_T = D_A + D_R$) is conserved. See the chemical reaction network, model diagram below for graphical illustration. Please see Table 1 below for the descriptions and values of parameters.

Chemical reaction network

$$D_A \xrightarrow{\alpha_M} D_A + M \qquad M \xrightarrow{\alpha_P} M + P \qquad P \xrightarrow{\alpha_F} P + F \qquad D_A + F \xrightarrow{k_f} D_R \qquad D_R \xrightarrow{k_b} D_A + F$$

$$M \xrightarrow{\beta_M} \cdot \qquad P \xrightarrow{\beta_P} \cdot \qquad F \xrightarrow{\beta_F} \cdot \qquad D_R \xrightarrow{\beta_F} D_A$$

Model diagram

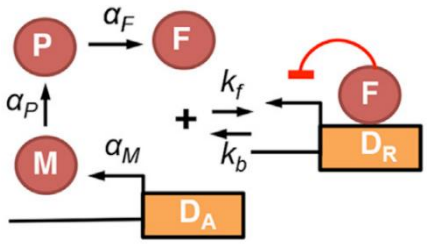


Table 1. Parameters of the genetic negative feedback loop model

Name	Description	Value
α_M	Transcription rate constant for M	15.1745/hr
α_P	Translational rate constant for P	1/hr
α_F	Production rate constant for F	1/hr
β_M	Degradation rate constant for M	1/hr
β_P	Degradation rate constant for P	1/hr
β_F	Degradation rate constant for F	1/hr
k_f	Binding rate constant for F and D_A	200/nM hr
k_b	Unbinding rate constant for D_R	50/hr
D_T	The concentration of total DNA	164.75nM

Problem 1: Please write a system of ordinary differential equations (ODEs) for the model described above and simulate (i.e., numerically solve) it using the parameters in Table 1 to check whether they generate oscillations in M and P. In other words, please plot the time courses of M and P. Please use the following initial condition: $D_A = 164.75\text{nM}$, $D_R = 0\text{nM}$, $M = 0\text{nM}$, $P = 0\text{nM}$, and $F = 0\text{nM}$.

Problem 2: Please examine how the solution trajectory changes as the dissociation constant $K_d := k_b/k_f$ increases from 0.4 to 1 by increasing k_b from 50/nM hr to 200nM hr.

Problem 3: Please apply the standard Quasi-Steady State Approximation (sQSSA) to the model (i.e., take $\frac{dD_A}{dt} = 0$ and $\frac{dD_R}{dt} = 0$), and derive a reduced ODE model with three variables M, P, and F (You should write the equations of the reduced model in your report).

Problem 4: Simulate the reduced model derived in Problem 2 and examine whether its solution trajectories are similar with the solution trajectories of the full model in Problem 1 (i.e., examine whether sQSSA is accurate).

Problem 5: You might find that the sQSSA is inaccurate in Problem 2. The inaccuracy of the sQSSA results from treating F as a slow variable, even though it is affected by both slow (production and degradation) and fast (binding and unbinding to DNA) reactions. This problem can be solved by introducing the total amount of repressor, $R \coloneqq F + D_R$, instead of F. As a result, the model only depends on slow reactions. Please write a system of ODEs with variables M, P, R, D_A, and D_R.

Problem 6: Solve the QSS equations ($\frac{dD_A}{dt} = 0$ and $\frac{dD_R}{dt} = 0$) and obtain the equilibrium values of $D_A(R)$ and $D_R(R)$ in terms of R (i.e., apply the total QSSA (tQSSA) to the model). Then, using these QSS solutions, derive a reduced ODE model with three variables M, P, and R.

Problem 7: Simulate the reduced model derived in Problem 3 and examine whether its solution trajectories are similar with the solution trajectories of the full model in Problem 1 (i.e., examine whether tQSSA is accurate).

Bonus problem: So far, we considered a single-cell model of the circadian clock within the SCN. However, the individual rhythms of each neuron are synchronized through intercellular signaling. For example, Vasoactive Intestinal Polypeptide (VIP), released by SCN neurons, acts as an intercellular signaling molecule that helps synchronize the individual circadian clock. To investigate the intercellular coupling among multiple cells, the following mathematical model was proposed:

$$\frac{dM_i}{dt} = f(R_i) - M_i + \frac{\mu}{N}\sum_{i=1}^{N} V_i,\ \frac{dR_{ci}}{dt} = M_i - R_{ci},\ \frac{dR_i}{dt} = R_{ci} - R_i,\ \frac{dV_i}{dt} = \tau(f(R_i) - V_i) \quad \text{(Eq. 1)}$$

where $f(R_i) = \frac{A - R_i - K_d + \sqrt{(A - R_i - K_d)^2 + 4AK_d}}{2A} \approx Max\left[1 - \frac{R_i}{A}, 0\right]$, $A = 0.0659$, $K_d = 10^{-5}$, N denotes the number of cells, each indexed by $i = 1, \ldots, N$. In this model, each cell releases VIP into the extracellular space at a rate proportional to the activity of the promotor, $f(R_i)$. Similar to previously developed models, we assume that VIP in the extracellular space enters each cell at an equal rate because the VIP diffusion is sufficiently fast. Once it has entered the cell, VIP promotes the transcription of the repressor gene. The parameter μ and τ describe the coupling strength and the timescale of intercellular coupling, respectively. Here, we use $\tau = 20$ to describe the fast coupling process.

The question we address in this problem is how the periods of individual cells change in the presence of coupling. To start, we consider a pair of cells (i.e., $N = 2$) with different intrinsic periods. Following previous studies, we scale time differently in two copies of a model cell (Eq. 1) to achieve a difference in periods. Specifically, all production and degradation rates are divided by a rescaling factor, 1 and 1.2, respectively, resulting in periods that differed by 20%.

In this setting, please simulate the model by varying the coupling strength μ from 0 to 0.3 (0, 0.05, 0.1, 0.2, 0.3) and examine the periods of the two cells (or the frequency of the two cells) and varying initial conditions of the model. Feel free to use any methods (e.g., a fast Fourier transform) to estimate frequencies.

Model diagram