Chapter 4

Week 2 Activities and Assignments Set 2

Most of the exercises and activities are based on material that is available in short lectures and slides on our website

`http://www.math.ubc.ca/~keshet/MCB2012/`

Please check out the link t either Week 1 or Week 2. or else, in some of the daily lectures recorded at PIMS. You can get to those recoded lectures by going through a link on our main Home Page.

`http://www.math.ubc.ca/~keshet/MCB2012/`

4.1   Polymer length distributions (continued)

In Homework Set 3, you found the steady state length distribution of a polymer *when the monomer concentration is held fixed*. You did so by solving the (linear discrete) equation

$$\frac{dx_j}{dt} = k^- x_{j+1} - (k^- + ak^+)x_j + k^+ a x_{j-1}$$

assuming that k^+, k^-, a were constant. For consistency of notation in this homework set, we will use the notation p_j throughout, so our polymer equation is

$$\frac{dp_j}{dt} = k^- p_{j+1} - (k^- + ak^+)p_j + k^+ a p_{j-1} \tag{4.1}$$

Your solution for the number of polymers of length j, p_j as a function of j contained an arbitrary constant, let us call it B, as well as dependence on some

ratio of rate constants that I will call a_{crit}. Here we add some postscripts to that problem.

Exercise 4.1.1 (Polymer Length, continued)

(a) Suppose that the smallest stable configuration consists of an m-sized nucleus, meaning that m monomers must come together to form the first nucleus that can grow. Then there is a term proportional to a^m in the equation for p_m, and we will call the proportionality constant k_{init}. We also define γ as the rate at which the m-size polymer breaks apart. Write down a differential equation satisfied by the first stable nucleus, p_m.

(b) Assume that the monomer concentration is fixed, and that at steady state, $p_m = Br^m$. Use the steady state equation for p_m to find B in terms of $k_{init}, \gamma, a_{crit}, etc.$ You have thereby determined the arbitrary constant. Use it to write down the solution for p_j where $j > m$.

4.2   Simulations of polymers and/or microtubules

Do EITHER one of Exercises 2 or 3, or if you are motivated, you can do both for bonus points. XPP codes are provided in the Appendix and on the website. This has parameter values, and other useful information, even if you plan to use Matlab.

Exercise 4.2.2 (Simulating the dynamic polymer length distribution)

Here we consider the dynamics of polymer assembly when the monomer pool is being depleted, and when the smallest polymer is size m.

Let N_p be the total number of polymer pieces, any one of which can grow or shrink by polymerizing,

$$N_p = \sum p_i.$$

We take the same polymer equation (4.1). On a separate handout (see website) it is shown that when the total mass is constant, the equation satisfied by the monomer is:

$$\frac{dc}{dt} = -[mk_{init}c^m - (m\gamma + k_r)p_m] - (ck_f - k_r)N_p \qquad (4.2)$$

After a while, once there are lots of growing polymers, this can be approximated by

$$\frac{da}{dt} = (k_r - k_fc)N_p. \qquad (4.3)$$

(a) Explain the Equation 4.3.

(b) Use your favorite simulation method to simulate the growth of polymers up to size $j = 100$ (i.e. the evolving length distribution). Assume that the smallest stable size is a nucleus of 3 monomers (a tri-mer, namely size 3 polymer). Start with monomers only, and show how the distribution evolves. [NOTE: while XPP may not be the best tool for this problem, if you'd like to use it, you can watch the instructional video XPP tutorial, and use the .ode file **asizedis2.ode**, which you just have to edit slightly.]

Exercise 4.2.3 (Simulating Microtubule populations (Dogteron-Leibler))
The Dogterom-Leibler equations for microtubule (MT) populations are:

$$\frac{\partial u_g}{\partial t} = -v_g \frac{\partial u_g}{\partial x} - f_{gs} u_g + f_{sg} u_s, \tag{4.4a}$$

$$\frac{\partial u_s}{\partial t} = v_s \frac{\partial u_s}{\partial x} + f_{gs} u_g - f_{sg} u_s, \tag{4.4b}$$

Here $u_g(x,t), u_s(x,t)$ are the "densities" of growing and shrinking microtubule plus ends, $v_g, v_s > 0$ are the rates of growth and shrinking, and $f_{gs}, f_{sg} > 0$ are the rates of transition $g \to s$ and $s \to g$, respectively. We will also consider the boundary conditions

$$u_g(0,t) = P, \tag{4.5a}$$

$$v_s u_s(L,t) = v_g u_g(L,t), \tag{4.5b}$$

(a) Explain the equations (4.4) and the boundary conditions (4.5).

(b) Analytically solve the steady state equations.

(c) Use your favorite software or XPP to numerically simulate the growth of the MT population.

4.3 Reverse Engineering a polymer reaction

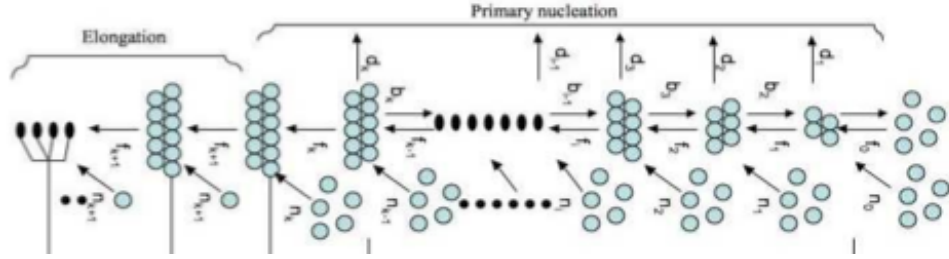**Exercise 4.3.4 (Using scaling to determine Polymerization kinetics)**

Figure 4.1: The generic polymerization mechanism. There are several oligomers stages (right to left) that are each formed by adding some number n_i of monomers to a smaller oligomer. The schematic shows forward and reverse rates and disassembly rates. Once the nucleus is formed, it is stable and grows.

The following generic model is assumed for the assembly process. The goal will be to determine various powers and coefficients in these equations.

$$\frac{dp_1}{dt} = f_0 c^{n_0} - f_1 c^{n_1} p_1 + b_2 p_2 - d_1 p_1, \tag{4.6a}$$

$$\frac{dp_i}{dt} = f_{i-1} c^{n_{i-1}} p_{i-1} - f_i c^{n_i} p_i - b_i p_i + b_{i+1} p_{i+1} - d_i p_i, \tag{4.6b}$$

$$\frac{d\nu}{dt} = f_k c^{n_k} p_k, \tag{4.6c}$$

$$\frac{dM}{dt} = f_{k+1} c \nu. \tag{4.6d}$$

where Eqn. (4.6b) holds for $2 \le i \le k$.

It will be assumed that the total mass is approximately divided between monomer and nuclei (with very little in the oligomers p_i) so that $c(0) = c(t) + M(t)$.

The observation of critical importance is that the data for many polymerization curves (each with its own starting monomer level) will collapse to a single "master curve" when the following scaling is applied:

$$t_0 \propto A_\infty^{-\gamma} \tag{4.7}$$

(a) Use the definitions in Table 4.1 to explain the terms in the equations. Fig 4.1 shows the process that the above equations represent.

(b) Rescale the equations for p_1 and p_i by defining the following dimensionless variables:

$$\hat{t} = \frac{t}{t_o}, \quad \hat{c} = \frac{c}{c_o}, \quad \hat{p}_i = \frac{p_i}{X},$$

where $c_o = c(0)$.

Variable	meaning	model
$c(t)$	monomer concentration	all
$p_i(t)$	number concentration of the ith oligomer ($i = 1 \ldots k$)	all
n_i	number of monomers added to p_i to form p_{i+1}	
$\nu(t)$	number concentration of nuclei	NDP
k	number of different oligomer species	
f_i	forward rate constant for the ith oligomer species	
f_k	forward rate constant for nuclei formation	
f_{k+1}	forward rate constant for polymer elongation	
b_i	backward rate constant for the ith oligomer species	
d_i	disintegration rate constant for the ith oligomer species	
$M(t)$	monomer mass in fibril, excluding monomers in oligomer and nuclei	NDP
$A(t)$	the measured fluorescence, assumed to be a measure of M	NDP

Table 4.1: Variables and their meanings in the various models. NDP= nucleation dependent polymerization.

(c) Now apply the fact that data scales such that

$$t_o \propto c_o^{-\gamma}, \quad \Rightarrow \quad t_o = \frac{\lambda}{c_o^{\gamma}}$$

Here λ is some proportionality constant carrying units, and needed to convert units of c_o^{γ} to time units. We want the coefficients in the scaled equations to be independent of c_o. This enforces strict constraints.

Find n_1, b_2, d_1 using this constraint, applied to the (scaled) equation for p_1. Explain what your conclusions mean about the polymerization reaction.

(d) Similarly use $t_o = \lambda c_o^{-\gamma}$ applied to the equation(s) for p_i to draw conclusions about the quantities n_i, b_i, d_i.

(e) Consider the nuclei and the mass equations, and use the scaling

$$\hat{\nu} = \frac{\nu}{\mu}, \quad \hat{M} = \frac{M}{c_o}$$

to convert these to dimensionless form. At this point it will be convenient to set $X = \mu$.

(f) Show that the same constraint then identifies the required value for n_k. Show that another conclusion reached is that $\mu = c_o^{\gamma}$. Finally, determine the value of n_0 using your results so far.

(g) Now that all the scales (and some coefficients) have been so determined, you can rewrite the dimensionless equations in a simpler form. Please do so.

(h) How many monomers are there in the first, second, third oligomer? (i.e. in p_1, p_2, p_3)? If? the first stable nucleus is represented by $\nu = p_k$ many monomers are there in the first stable nucleus (answer in terms of k).

In the next exercise we will use some other information to find the value of the constant k, i.e. the size of the first stable nucleus.

Exercise 4.3.5 (Number of steps from monomer to fibril)

Close to the beginning of the reaction, $c(t) \approx c(0) = c_o \approx$ constant, and $\hat{c} = c/c_o \approx 1$, $p_1(0) = p_2(0) = \ldots p_k(0) = 0$. At that very early stage, we can approximate the equation for p_1 and use that to get a rough estimate of the time course of $p_1(t)$ for a short time period:

$$\frac{dp_1}{dt} \approx \lambda f_0 \cdot 1^{2\gamma} \quad \Rightarrow \quad p_1(t) \approx \lambda f_0 t,$$

Using this idea on the other equations will allow us to estimate the values of k (the number of steps needed to form a stable fibril).

(a) Write down a similar set of approximate equations for $p_2, p_3, ..P_k, \nu, M$ close to the beginning of the reaction. By integrating these simple equations, show that you obtain an approximation of the dimensionless variable M as a function of (dimensionless) time in the form

$$M(t) = t^{(\text{some power})}$$

and find that power in terms of the number of steps k.

(b) Recall that $A(t)$, which is the experimentally measure of polymerization is an indicator of $M(t)$, the mass in polymer form in the reaction. You are given a plot of $\log(A(t)/A_\infty)$ versus $\log(t/t_0)$, and you find that this plot is well-fit by a straight line of slope 8.01. How many steps does it take to assemble a fiber? [Recall that $M(t) \equiv (A(t)/A_\infty)$.]

4.4 Appendix with XPP codes

These codes are also available on our website as plain text.

4.4.1 Polymer Size Distribution

This file has been corrected for the right monomer equation and for the fact that dimers are the smallest size class.

```
# asizedis2.ode
# polymer size distribution
# simple polymerization
# May 12, 2012

# xj= number of fibers of length j
# a= conc of monomers
# kf, kr = forward and reverse rate constants
# Set adepl=1 for constant total amt, 0 for fixed monomer

par kf=10, kr=1, kinit=0.005, gamma=1, adepl=1

# Set the initial monomer level
a(0)=20


#***********************************************
#at each stage, sum up the total number of polymer pieces
# (starting from largest down to smallest)

sm100=x100
sm[99..2]=sm[j+1]+x[j]
Np=sm2

#sm2 is then the total number of pieces after having
# added them all
#***************************************************

#eqn for monomers:
# Set adepl=1 for constant total amt, 0 for fixed monomer level
#a' = adepl*(-kf*a*(sm2-x100+x2)+kr*(sm2+x2))
# The above old equation for monomers was wrong.
# For mass conservation use:

a'=-2*kinit*a^2+(2*gamma+kr)*x2-(kf*a-kr)*Np

# initialization from dimers
x2'= kinit*a*a-(a*kf+gamma)*x2+kr*x3
x[3..99]'= kf*a*(x[j-1]-x[j])+kr*(x[j+1]-x[j])
x100' = -kr*x100+kf*a*x99

@ bounds=10000000, meth=gear, dt=0.05, total=100
done
```

To run this code: bring up XPP and type Initial conditions Go (I G)

To view the line plots of p_j versus j for various times:

File Transpose (F T). Fill in the table:

Column 1: x1

Ncols: 100 (*for the number of size classes*)

Colskip: 1 (*to display every size*)

Row 1: 1 (*start at t=0*)

NRows: 2000 (*Because there are 2000 time steps*)

RowSkip: 1 (*to see the earliest time behaviour*)

click OK

Bring up the IC's panel (top left button of your XPP window. click on each box at the left hand side of the panel (next to the quantities $A, X1, X2, ..$ up to $X9$. Click on xvst at the bottom of that panel. You should get a lovely colorful plot of the polymerization size classes. To see the later time behaviour repeat the above but use

RowSkip: 200 (*to see the plots every 200'th time step*)

4.4.2   Microtubule populations

```
# microtubulesDog.ode


# XPP code for simulating growing and shrinking microtubule
# plus ends according to Dogterom & Leibler PRL paper


# upwind basic Euler scheme on spatial domain
# discretization:
par h=0.1
#L is the domain length, set to 10 micro-meters
L=h*100.0



# here ug is the density of growing plus ends of MT.
# and us is the density of shrinking plus ends
# fgs=transition rate from ug to us
# fsg = transition rate from us to ug
#vg= growth rate
#vs=shrink rate



ug[1..100]'= vg*(ug[j-1]-ug[j])/h+ fsg*us[j]-fgs*ug[j]
us[0..99]'= vs*(us[j+1]-us[j])/h- fsg*us[j]+fgs*ug[j]

# equate fluxes for growing and shrinking tips at x=L
us[100]=ug[100]*vg/vs
# plus ends nucleated at x=0 so as to be at constant level there
ug0=1


# param values from Dogterom et al PNAS 1995
```

```
# in units of micro meter/min for veloc and sec^-1 for transitions
# (for this reason, need factor of 60 to convert f's to per min)
# vg=10,vs=15,fgs=60*0.012,fsg=60*0.02
param vg=10,vs=15,fgs=0.72,fsg=1.2

# param values from the Phys Rev Let paper footnote [9] are:
# vg=2,vs=20,fgs=60*0.004,fsg=60*0.05
# param vg=2,vs=20,fgs=0.24,fsg=3

# Note that a transition in the behaviour should take place
# when vs*fgs=vg*fsg  currently 0.18 vs 0.2

@ dt=.01,total=10
done
```