

The following questions consider binding of a transcription factor protein, denoted  $X$ , to a gene promoter with  $N$  binding sites. The number of transcription factors bound is denoted  $n$ , and in the general case

$$n \in \{0, 1, 2, 3, \dots, N\}.$$

The number of copies of the gene with  $n$  sites bound is denoted  $G_n$ . The total number of copies of the gene is the constant

$$G_T = \sum_{n=0}^N G_n,$$

and the fraction of copies in state  $n$  is  $G_n/G_T$ . The transcription rate for a copy of the gene with  $n$  transcription factors bound is denoted  $v_n$ , and the total transcriptional rate is

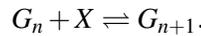
$$\beta = \sum_{n=0}^N v_n G_n.$$

We will assume that  $v_0 = 0$  and that increased binding gives increased transcription, which implies that the maximum possible transcription rate occurs when  $G_N$  is the only occupied state. Thus, the maximum transcription rate is  $\beta_{\max} = v_N G_T$ . The concentration of transcription factor is sufficiently large that changes in the value of unbound  $X$  need not be considered. All questions assume that binding has reached equilibrium.

Cooperativity at a particular value of  $X$  is defined as  $(d/d \ln X) \ln[\beta/(\beta_{\max} - \beta)]$ , usually evaluated at the value of  $X$  giving half-maximal transcription,  $\beta = \beta_{\max}/2$ . Note that  $d/d \ln X$  is  $X d/dX$  by the chain rule.

1. Fully cooperative binding. The standard model for fully cooperative binding is that only the two states  $G_0$  and  $G_N$  exist,  $G_0 + NX \rightleftharpoons G_N$ , with forward rate constant  $k_f$  and backward rate constant  $k_b$ .
  - (a) Show that  $\beta$  as a function of  $X$  is proportional to  $(X/K)^N/[1 + (X/K)^N]$ . Provide  $K$  as a function of model parameters.
  - (b) Provide  $\beta/(\beta_{\max} - \beta)$ .
  - (c) Show that the cooperativity is  $N$  regardless of the value of  $X$ .
2. Fully independent and additive binding. Suppose each site is bound independently. Each site has two states,  $u$  (unbound) and  $b$  (bound), with  $u + X \rightleftharpoons b$ . The forward rate constant is  $k'_f$ , and the backward rate constant is  $k'_b$ . Because this expression is in terms of a single site,  $u + b = 1$ . When  $n$  sites are bound, the transcription rate is additive,  $v_n = n\Delta v$ .
  - (a) What is the probability  $\theta$  that a particular site is bound?
  - (b) In terms of  $\theta$ , what is the probability  $G_n/G_T$  that  $n$  of the  $N$  sites are bound for a particular copy of the gene?

- (c) Show that the transcription rate is  $\beta = G_T \Delta v \sum_{n=0}^N n C(N, n) \theta^n (1 - \theta)^{N-n}$ , where  $C(N, n)$  is the combinatorial factor  $N!/n!(N-n)!$ .
  - (d) Define  $\tilde{\theta}(s) = \sum_{n=0}^N e^{-sn} C(N, n) \theta^n (1 - \theta)^{N-n}$ . Show that  $\tilde{\theta}(s) = (1 - \theta + e^{-s} \theta)^N$ . Use this result to calculate  $\langle n \rangle$  as  $-(d/ds) \ln \tilde{\theta}(s)$ .
  - (e) Show that  $\beta$  for a particular value of  $X$  is  $\beta_{\max} \theta$ .
  - (f) What is the cooperativity for independent binding and additive transcription? How does the cooperativity depend on the transcription factor concentration?
3. Sequential binding. Here we model a system that is intermediate between independent binding and cooperative binding. In this model, the transcription factors bind in spatial order with transitions possible only between neighboring states:



The forward rate constant is  $k'_f$  and the backward rate constant is  $k'_b$ .

- (a) Define  $\lambda = G_1/G_0$ . Provide  $\lambda$  in terms of model parameters  $k'_f$ ,  $k'_b$ , and  $X$ .
- (b) Provide the general term  $G_n/G_0$  in terms of  $\lambda$ .
- (c) Use the normalization  $\sum_{n=0}^{\infty} G_n = G_T$  to provide  $G_n/G_T$  in terms of  $\lambda$  without  $G_0$ .
- (d) At what value of  $\lambda$  are all the states equally occupied,  $G_0 = G_1 = \dots = G_N = G_T/(N+1)$ ?
- (e) Suppose that the transcriptional activation is additive,  $v_n = n\Delta v$ . Show that  $\beta = G_T \langle n \rangle \Delta v$ , where

$$\langle n \rangle = \frac{\sum_{n=0}^N n \lambda^n}{(\lambda^{N+1} - 1)/(\lambda - 1)}.$$