

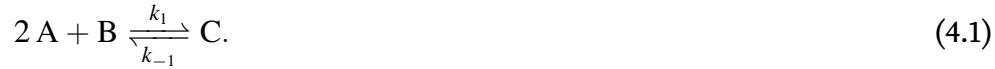
BE 25 Winter 2024

Homework #4

Due at 9 AM PST, February 1, 2024

**Problem 4.1** (Time scale of relaxation experiment, 45 pts).

You wish to study the kinetics of a reaction



- Derive an expression for the relaxation time  $\tau$  of the relaxation experiment after a T-jump perturbation. Write your expression in terms of the rate constants  $k_1$  and  $k_{-1}$  and the steady state concentrations of A, B, and C.
- What set of relaxation experiments could you do to obtain good estimates for  $k_1$  and  $k_{-1}$ ?

**Problem 4.2** (HIV protease inhibitors and pH dependence, 55 pts).

*This problem is based on problem 4.10 of WTHS.* Some enzymes, such as HIV protease, exhibit pH-dependence on their catalytic activity. As a simple example, imagine an enzyme that can bind substrate in its protonated state, but not in its unprotonated state. That is, it has the following reaction scheme.



- Derive an expression for the reaction velocity,

$$v_0 = \frac{dc_P}{dt}. \quad (4.4)$$

This should be an analytical expression, and you will need to make approximations to derive it. Be sure to clearly state which approximations you use. It should be written in terms of  $c_E^0$ ,  $c_S$ , and  $c_{H^+}$ . Does the resulting expression match a Michaelis-Menten form? If so, what are the effective  $k_{cat}$  and  $K_M$ ?

- In the presence of an inhibitor, such as HIV protease inhibitors used in some treatments, the situation gets more interesting. In an inhibitor could also bind the enzyme in either the protonated or unprotonated form, giving additional reactions





The inhibitor-bound unprotonated enzyme may also be protonated.



though, for reasons we will learn about later in the course, this last reaction is dispensible.

For this inhibited scheme, derive an expression for the reaction velocity, again making appropriate approximations. It should be written in terms of  $c_E^0$ ,  $c_S$ ,  $c_{\text{H}^+}$ , and now also  $c_I$ . Does the resulting expression still match a Michaelis-Menten form?

- c) How do the effective  $k_{\text{cat}}$  and  $K_M$  you found in part (b) depend on pH, if at all?
- d) Does it matter whether the inhibitor binds more readily to the unprotonated or protonated state of the enzyme? Explain.