

BMEN E4001x: Quantitative Physiology I / Molecular and Cellular Systems
Notes 13 – Motors and muscles

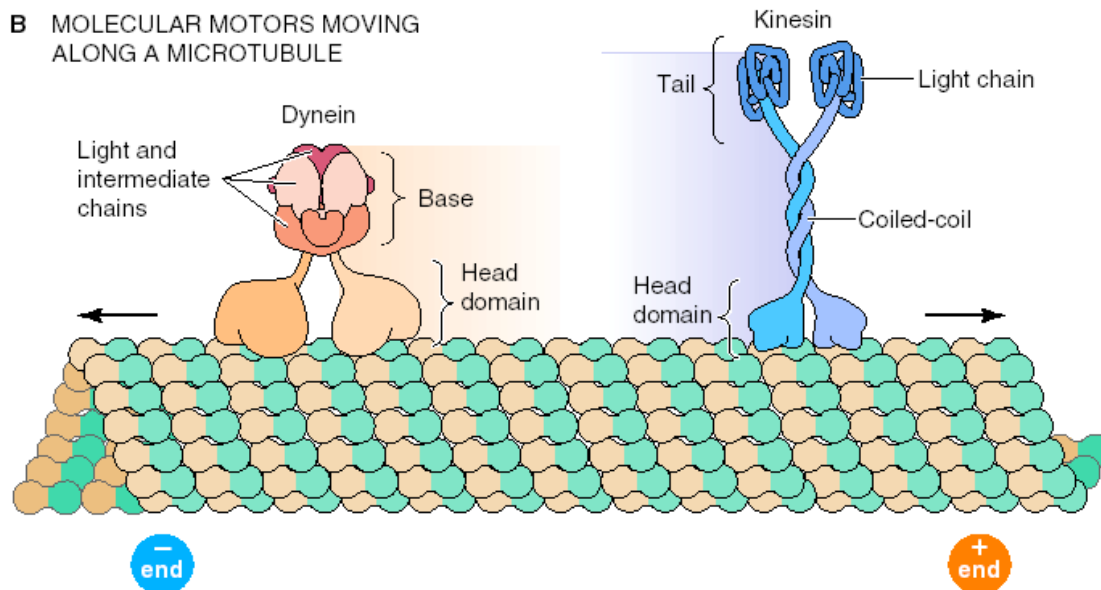
Sources for this section:

B&B: Chapter 9

Nelson: Section 10.4

Tubulin - kinesin:

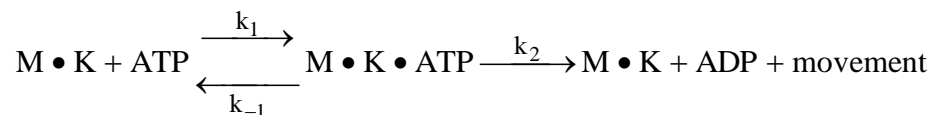
(We'll focus on kinesin, as it is better understood)



With ATP, kinesin moves one step along microtubule (~8nm), even with large loads. Suggests tight coupling between motor protein and microtubule. As a chemical reaction, can expressed as

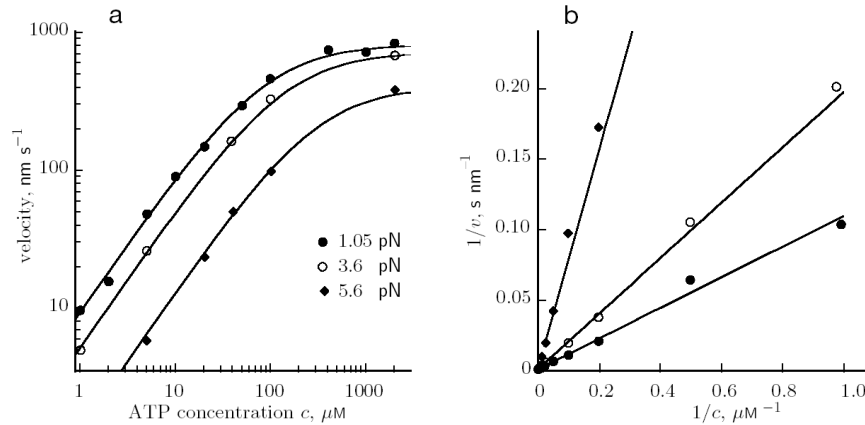


and in this respect, is an enzymatic reaction. Let's look a bit closer, first by rewriting as:



ATP binding is purely chemical, but there is a power stroke that accompanies hydrolysis.

This would suggest a MM-type response with respect to ATP, and indeed, it is superficially seen to be true.



The different lines indicate different loads. Each load exhibits MM-type behavior, as seen from both types of plots. (slope= K_M/v_{max}). Something is wrong here when we look at the inclusion of motion, however. Let's start with some actual values:

load, pN	v_{max} , nm/s	K_M , μM
1.05	813 ± 28	88 ± 7
3.6	715 ± 19	140 ± 6
5.6	404 ± 32	312 ± 49

One would expect that increased load will decrease speed. All engineers should recognize this from working with motors of any sort. Indeed, v_{max} decreases.

A fair model would be that this affects k_2 , slowing down that progress.

Going back to the Michaelis-Menten model,

$$K_M = \frac{k_{-1} + k_2}{k_1}$$

Decreasing k_2 should decrease K_M ; contrary to observations. More complex situation, it seems.

Discussion from Nelson, section 10.4, based on mid-1990's analysis from the Block lab (while still at Princeton).

Facts about kinesin, ATP, ADP, and myosin:

Double-headed kinesin is a little too short to span one monomer, but can with some induced strain.

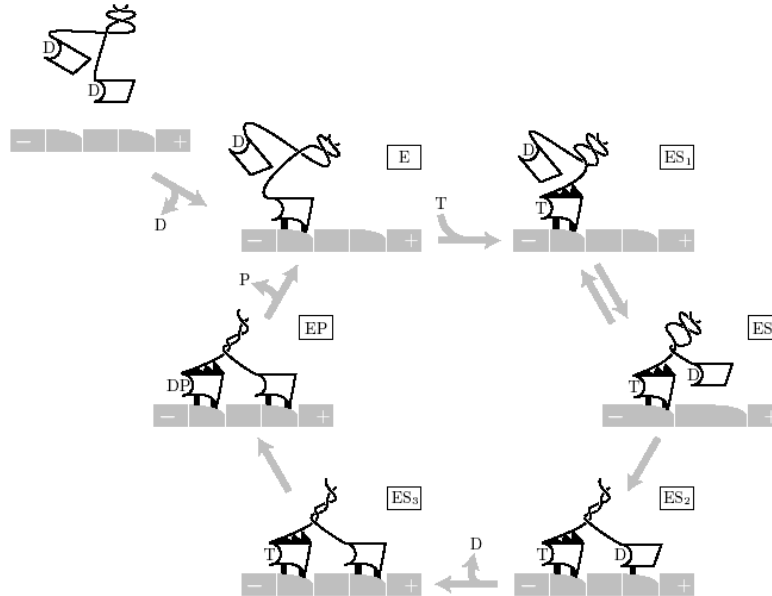
At the interface of the linkers and each head, binding site for nucleotide; in either unoccupied or ADP bound, the link is floppy. With ATP bound, it is strained and pushes molecule towards “+” end of microtubule.

Kinesin binds ATP strongly, hydrolyses quickly.

Kinesin without bound nucleotide binds microtubules strongly

microtubule-kinesin-ADP complex only weakly bound; steric hindrance or allosteric inhibition of simultaneous microtubule and ADP binding.

Oddly, kinesin with 2 ADPs bound binds readily with microtubule, but only releases half of the ADP. On addition of ATP, the other half of ADP is released. This also works if the analogue AMP-PNP is used instead of ATP; hydrolysis has nothing to do with the ADP release.



Flowchart and descriptions as in Nelson.

Note that ATP really comes in between the stages labeled E and ES₁. Thus, the transition k_1 from before corresponds to E to ES₁ transition. Consequentially, moving through ES₁' through ES₂ and ES₃, all of which are unidirectional and fast, corresponds to going through k_2 .

Consider ES₁ and ES₁' as a single composite state.

The argument is that ES₁ and ES₁' are nearly in equilibrium. Thus, more time spent in ES₁ lowers k_2 (leaving composite state). More time spent in ES₁' lower probability of reverse reaction, k_{-1} .

Modification of k_{-1} , allowed in this model in absence of a power stroke, may allow altering of both v_{max} and K_M in the manner suggested by the data. That is, if k_{-1} is lowered more than k_2 is raised, v_{max} can go up, and K_M can go down.

The driving force for this equilibrium is sort of a power stroke, the energy needed to go between the ES₁ and ES₁', which includes force over distance. Namely, $\Delta G = (\text{isomerization energy}) + F \cdot l$. with an isomerization energy of $-5k_B T$, F as dictated by experiment, and $l = 4\text{nm}$, the shown fits are achieved.

This model draws a link between tight binding and the two headed-ness of kinesin. This tight coupling is not always needed; in fact, 1 headed kinesin will also move along a microtubule, but exhibits some slippage. This motion is based on the idea of a Brownian ratchet, discussed in a number of texts, including Nelson. For those students looking to go into biophysics, very important idea, but I'll leave it out of this class.

Actin:

The molecular motor for actin actually does exhibit this slippage! This is myosin. The slides template how myosin/actin interact – crossbridge theory.

(no additional material here; directly from B&B)