Keeping the Physics in Biophysics -- and vice versa

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For these slides see:
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A dropped connection

I did my undergraduate studies, majoring in Physics, at a famous university where biophysical topics never entered the undergraduate Physics curriculum at all. In fact, there were no biophysics courses offered, even to interested students. Much later I realized that sometime around the 1950s, Physics departments largely pushed Biophysics out, saying “that’s not Physics.”

Luckily, times change. But when I was asked to teach the subject, I found myself way behind other people who had studied, say biochemistry, or physiology, or neuroscience. And when I tried to read books by those people, I found it tough going, in part because of the very different culture in those fields.

And yet--it was not always so. Before my time, life science and physical science were regarded as inseparable--as Natural Science. Advances went in both directions:

- Electrons ----> Electron microscopy (instrumentation)
- Galvani’s frogs ----> electrochemistry (fundamental mechanisms)
- Heredity+polymer chemistry ----> DNA
- etc.

Today we’re entering another golden age of two-way exchange between life science, physical science, and even engineering. It’s time for our education to reflect that.
Why do we even have classes at all?

🔹 To tell them facts? No -- facts are now free in infinite quantity. Ingesting and excreting facts may have been ok till now, but in our students' professional future it's not enough.
🔹 To tell them the latest, most trustworthy facts? No -- facts go out of date in the blink of an eye.
🔹 If research is so important, why even bother with classes?

Well -- *skills and habits* still matter a lot. When you walk into a room with your toolbag and encounter a problem you’ve never seen, which tool should you pull out of your bag? Knowing where to begin is a difficult but learnable skill.

There are a lot of poorly-paid drones out there who sit in front of a black box and do repetitive tasks.
Better to formulate *new ideas*.
Students need to develop the right skills and habits for that, but many (most?) courses don’t really help.

A class should help them do that -- in some specific context. *Biophysics is an interesting context for that purpose.*
The interesting questions in science are those where we shake our heads and ask, "How could anything like that possibly happen at all?" And biophysics is full of such questions.

Effective science needs an act of imagination, but imagination must be coupled to discipline, often in the form of a falsifiable model, a distillation of the processes thought to be relevant.

Quantitative models are the most falsifiable, and therefore the strongest when they survive repeated attempts to falsify them.

But the physical world is full of randomness which gets in the way of confronting quantitative models with data.

Far from being a mere nuisance, something that good scientists must eliminate, randomness is often unavoidable -- and sometimes even the main point. This means that we have got to get good at figuring out what quantitative conclusions can be inferred from noisy data, and how reliable those conclusions are. Physicists are good at that.
RECOMMENDATION #1.3

The principles of physics are central to the understanding of biological processes, and are increasingly important in sophisticated measurements in biology. The committee recommends that life science majors master the key physics concepts listed below. Experience with these principles provides a simple context in which to learn the relationship between observations and mathematical description and modeling.

The typical calculus-based introductory physics course taught today was designed to serve the needs of physics, mathematics, and engineering students. It allocates a major block of time to electromagnetic theory and to many details of classical mechanics. In so doing, it does not provide the time needed for in-depth descriptions of the equally basic physics on which students can build an understanding of biology. By emphasizing exactly solvable problems, the course rarely illustrates the ways that physics can be applied to more recalcitrant problems. Illustrations involving modern biology are rarely given, and computer simulations are usually absent. Collective behaviors and systems far from equilibrium are not a traditional part of introductory physics.
A course that works at Penn

Physicists are pretty comfortable teaching about molecular biophysics.

- It’s a good fit -- we like to talk about entropy, which we teach in a far more appropriate way than what they get in PChem.
- It’s still an exciting, opening field.
- A number of modern textbooks are now available.

But...

- Molecular biophysics is not so obviously connected to medicine, which many of our students plan to study.
- Every human on the planet is intrinsically interested in, e.g., their own vision.
- Today The Buzz is in “systems biology.”
- Physicists have a lot to say about Physical Models of Biological Systems, but we’re somehow ceding all the high ground to mathematicians and computer scientists.

Let’s see how a Physics department could offer an indispensable course to a wide variety of students, including the more numerate Bio majors, and the burgeoning group of Bioengineering majors.

Much inspiration from Bialek/Botstein 2004; Wingreen/Botstein 2006.
(Disclaimers)

(I realize that most of the people in this room could give a different, and equally good, talk on this subject. This is just a report on my own experience.)

(E.g., Rob will say things that are mostly orthogonal to my remarks, and they’re very important too.)
Ahem. You have to convince your department to actually offer the course. That means committing staff (you) to it, which generally means discontinuing something else. Doing it one-off isn’t enough -- it needs to continue. It’s a serious concern.

 Everywhere Engineering departments are sick of freshman Physics, and trying to wriggle out of having their students take it. We desperately need a product that our customers want.

 Engineering departments, and their students, are keenly interested in life science.

 A course on Biophysics is also essential for a major in Biophysics. You can create such a major even without a corresponding academic department. Then the course gets a base.

 Your colleagues may still say “that’s not Physics.” To avoid this, you need to make sure you keep the physics in Biophysics -- you need to include some top-drawer, indisputable Physics content. It can’t all be about bioinformatics.

 Is it worth the struggle? More on that later.
Monod found something funny in the growth of bacteria in mixed medium. He asked, "how could that possibly happen at all?" And he ended up with the operon model.

Could bacteria somehow be implementing a two-state switch like the ones that changed human civilization in the mid-20th century?
Switching, II

Students can write a model of two mutually repressing genes, make the phase-plane analysis, and find the region of bistability in Matlab.

It’s not speculation -- now the transfer functions of each element have been measured. The era of *synthetic biology* has arrived.

Figure S1: Snapshots of a typical regulator dilution experiment using the $O_{R2}^{2*}$-λ-cascade strain. Panels show the same microcolony as Fig. 1D, with greater time-resolution. Cl-YFP protein is shown in red and CFP is shown in green. Times, in minutes, are indicated on snapshots. Insets show a selected cell lineage (outlined in white).
Molecular machines

Myosin is a molecular motor that walks on actin filaments:

How can we get information about this invisibly small motor’s mechanism?
Here is where a little probability goes a long way.
Students can test two hypotheses about the gait by analyzing the statistics of the motor’s steps:

Images based on x-ray crystallography data by David Goodsell.
Dwelling time distribution of the two stepping types of myosin X.
Red bars: Type 1; Blue bars: Type 2.
Assuming the same stepping rates of the two heads of myosin X.
These distributions tell a story, if you know how to convert a hypothesis to a predicted distribution (curves).

MyoX-QD655 Dwelling Time 1 µM ATP 02/13/08

\[
\frac{1}{k_1} = 0.894 \text{ s} \\
\text{mean (dwelltime1)} = 1.144 \text{ s}
\]

\[
\frac{1}{k_1} + \frac{1}{k_2} = 1.621 \text{ s} \\
\text{mean (dwelltime2)} = 1.912 \text{ s}
\]
Luria and Delbruck noticed a statistical peculiarity in their data -- a huge "fat tail." They came up with a "Mendel, not Lamarck" model for drug resistance, and detailed quantitative predictions for such distributions that distinguished their model from the alternative. They had to work very, very hard. But now it’s trivial for students to simulate in Matlab.
Matlab simulation of Luria--Delbruck model:

Of 5000 simulated cultures, most have zero resistant mutants but a few have very many. This may be an ancient case history, but a lot of cutting-edge research is done on “fat-tail” distributions like this one. Biophysics is a good context for students to learn how to handle such things. In fact, many distributions arising in Biophysics have infinite variance.

Oh, also -- drug resistant bacteria are a very, very current problem.

And -- similar ideas are also illuminating when applied to retinoblastoma. A good physical model applies to problems beyond the one for which it was developed.
Students can simulate a random walk, then run it again and again to grasp the generic similarity of these figures despite the fact that they’re always different in detail. Then they can find the mean-square displacement to confirm the diffusive law.

We can show that the diffusive flux of oxygen to a bacterium is limited by its size. But that derivation is a bit abstract. We don’t “see” the oxygen molecules themselves.

Instead, students can simulate random walks in Matlab and find the diffusive flux to an absorbing sphere. They can then find the flux to a reflecting sphere with absorbing patches -- a calculation they cannot do analytically, and one with big implications (Berg and Purcell).
Genetic drift

Genetic drift is also a random walk -- with non-constant “diffusion constant.” Kimura had to work very, very hard to solve this model, but it led to the fundamental result that probability of fixation is proportional to $1/(\text{population})$. It’s incredibly important -- the basis of the molecular clock that gives us phylogenetic trees.

Students can trivially simulate this system, and obtain the key result, using Matlab.
If you’re looking at an unknown gene, and it resembles a lot of kinase genes, then maybe it’s a kinase. But resemblance can be hard to spot. Nonlocal sequence homologies can be important. Random walks on graphs can tease out those nonlocal aspects.

Well, that’s also the basis of the Google PageRank algorithm. Another civilization-changing discovery.

Noble et al FEBS 2005
Eyes are an ancient invention: Here is Trilobite, half a billion years old. That design was successful: Here is a modern aphid. But if you can afford to carry more weight around, here is a better design:
Either way, an eye looks like a planar array of pixels--superficially like a modern camera. About a hundred million photoreceptor cells in the human eye:
The human eye also has a lens-based focusing system, again like a camera. Already in 1625 Christopher Scheiner removed the coating from the back of an animal's eye and looked through the transparent inner wall. Through the eye he could see miniature upside-down images that were both sharp and bright.

It made sense in terms of the budding theory of optics, e.g. Snell’s 1621 result on refraction of light. Later a Who’s-Who of scientists (Descartes, Huygens, Newton, Kepler...) developed optics into a set of powerful rules. Much later, circa 1801, Young proposed that light was a wave phenomenon, then Maxwell, Fresnel, and many others showed how all the phenomena of optic, including refraction, reflection, polarization and diffraction, can be understood starting from the hypothesis that light is a wave excitation.

A key moment was the realization that the finite size of our pupils limits the resolution we can get at the retina, due to diffraction. There’s no point having a pixel size smaller than this resolution limit, and remarkably, our photoreceptor cells really are about this size.

Looks like the wave theory of light explains everything.
But what happens next? What happens in those photoreceptor cells that translates light into nerve impulses? We can detect very dim light with a photomultiplier tube, or a photodiode. Either way, light causes discrete clicks in the detector. *Dimmer light gives equally big clicks, just less frequent.* You might imagine a mechanism something like this:

![Diagram](image)

But that mechanism would give *uniformly spaced* clicks. Instead the clicks are *as random as possible* -- they are a “Poisson process.” Something about light is intrinsically random.

Moreover, when we shine dim light on several photodetectors, they *never respond in unison:* Each click comes from just one detector, even if the beam of light is spread out to cover them all.
Both digital and film cameras also expose one pixel at a time, at random:

3.6x10^6 photons

Images Albert Rose
Even classic diffraction effects turned out to be particulate in character.
All these phenomena are related to the “photoelectric effect.” Starting from its discovery by Heinrich Hertz, people found that:

- Light can discharge a negatively-charged electroscope.
- But not a positively-charged one.
- Ability of light to discharge an electroscope does not depend on the intensity (brightness) of the light, although brighter light discharged it faster.
- But it does depend on *color*: for many metals, only ultraviolet light worked.

Einstein found he could only understand these phenomena, and especially the photoelectric effect and thermal radiation, by postulating that light consists of tiny *lumps* called “quanta” or “photons.”
Hecht et al measured the probability of seeing a flash vs intensity. They found a simple physical model predicting the form of this “probability of seeing curve.” Then they were led from this information to the conclusion that single photons can excite rod cells, and that a quorum of simultaneous rod-cell firings is registered consciously. Students can fit their data in Matlab and find the critical quorum size.
Students can fit Hecht et al.’s data in Matlab and find the critical quorum size.

Above: threshold=7 fits the data well.
Modern methods

Direct measurements on single rod cells confirm that they can respond to single photons, and confirm the inherent randomness of the response.

An individual rod or cone cell’s response can be measured by gently sucking its outer segment into a pipette electrode and stimulating it with 500 nm light (green).

Scale: outer diameter of pipette about 6 micrometers.
“OK, so light comes in tiny lumps. Is that all?”

And yet, I mentioned earlier that light also shows many other properties long thought to be slam-dunk evidence of wavelike behavior, much of it critically important for the design of visual organs. How could any of that possibly happen at all in the particle picture? Einstein didn’t know.

Now, in physics we often put a box around a set of issues and say, “We can't understand that today,” and move on. But this is an intolerable contradiction. It's too big to put a box around it. We have to understand it before we have any business moving on.

Generally professors say, “You’re not ready for that. You’ll understand that some day.” (They really mean, “Shut up.”)

Is that really an adequate response? Students would have to wait till they were halfway through a PhD in high-energy particle physics (which they’re not going to do anyway) before we’d get around to telling them.

Can’t we tell them something we actually believe is true? Can’t we have them do a calculation for themselves that illuminates this apparent paradox?
So, OK -- our eyes respond to photons. So what about that intolerable contradiction? How can little bullets display the diffraction and refraction needed to explain physiology (and much more)?

*Lumpy-light hypothesis, continued:*

- The probability to observe a photon is the length-squared of a certain complex number $\psi$.
- If there are multiple routes (or processes) by which a photon could make the trip, and we don’t directly observe which one was taken, then they all make additive contributions to the total amplitude. These contributions are all complex numbers with equal lengths, but different angles, so they may reinforce or cancel.
- The angle (phase) of any one contribution equals the angular frequency of the light (related to its color) times the transit time for the path. In vacuum, light travels at the fixed speed $c$, so transit time may be written as $(\text{path length})/c$.
- In a transparent material, it gets complicated by all those electrons, but for some purposes it’s a good approximation to say that in water, etc. the speed is reduced to $c/n$, where $n$ is the “index of refraction.”

*That’s it.*

See Feynman, *QED*
So how does that help?

It’s incredible, but with that additional info we can reproduce all of the familiar classical optics results, including focusing by the lens of the eye. And you don’t need to trust some authority figure on that -- **students can do the calculations for themselves.**

The key question is, *Why does light (usually) (seem to) go on (pretty) straight lines?*

After all, a penny held in the sunlight casts a sharp shadow. And yet our Lumpy-light Hypothesis says that photons take *all possible paths* between source and detector!

We need a digression to see how, and when, this familiar behavior emerges from the Hypothesis.

In the pictures below, **students approximate the integrals in Matlab** as sums, drawing little arrows to represent each term in the sum. The full integral (red arrow) is the vector from one end of the chain to the other end, times $dx$.

\[
\int_{-5}^{5} e^{ix^2} \, dx
\]

\[
\int_{-1}^{9} e^{ix^2} \, dx
\]

A similar integral whose range of integration contains *no* stationary-phase point will have a small total value:

\[
\int_{1}^{11} e^{ix^2} \, dx
\]

Again: Near $x = 0$ the arrows point mainly to the right and they add up to something significant. If the range of integration contains that stationary-phase point, we’ll get a large total (long red arrow). Otherwise, we won’t.
The concept we found -- "stationary phase" -- explains why we sometimes get sharp edges, other times not.

Left: wide slit, sharp edges. Lower left: medium slit, medium edges. Lower right: narrow slit, fuzzy edges.

That gives us the "diffraction limit" on resolution (how well we can see) of an optical system. You can now understand other optical phenomena (like focusing) with similar principles.
I guess I believe that what everybody says is possible. We learned about the true character of light, we reconciled its wave and particle aspects... Is that all? Can’t we learn something new about biophysics once we’ve got this master key? I don’t want to learn physics just for the (alleged) fun of it!”

Yes. Somehow or other, the reception of a single photon gets converted to a neural impulse. By the time that impulse reaches the optic nerve, it has become a spike.

Spiking neurons, e.g. in the optic nerve, have limited dynamic range. They represent their signal by the times of individual spikes, each of which is exactly like any other. There are upper and lower bounds on this rate. The eye must make the best possible use of this limited range of representing the intensity of light at each pixel.

And the problem is even more acute than that: you have $10^8$ photoreceptors, but only $10^6$ optic nerve fibers!
At high illumination, truncation to 1-bit depth destroys a lot of detail (top right). But we can do much better than this, at the same high level of compression. To do so, students can apply a filter (bottom). The filter is called “center/surround;” it is the difference of two concentric Gaussians. And by the way, it’s a convolution -- an idea we met in a very different context in the start of the course.

Truncating in this way vastly reduces the image size, or the bandwidth needed to transmit it in a given amount of time. (In this case we went from 8 bits per pixel to just one.)
“Well, cool, but... *Do our eyes really do that?*” First look at psychophysical clues.

If you fixate on one intersection, other intersections appear gray, darker than the streets.
“Oh, please...

...Spare us the psychophysics. Is there some objective data on this?”
Meet *Limulus*:

![Horseshoe crab](image)

**Fig. 4.** The discharge of impulses from a single receptor unit in response to a simple "step" pattern of illumination in various positions on the retinal mosaic. The pattern of illumination was rectangular, covering an area 1.65 mm. × 1.65 mm. on the eye. It was obtained by projecting the demagnified image of a photographic plate on the surface of the eye. The insert shows the relative density of the plate along its length as measured, prior to the experiment, by means of a photomultiplier tube in the image plane where the eye was to be placed. The density of the plate was uniform across its entire width at every point. The measurements illustrated were made over the central 1.5 mm. of the image on the eye.

The upper (rectilinear) graph shows the frequency of discharge of the test receptor, when the illumination was occluded from the rest of the eye by a mask with a small aperture, minus the frequency of discharge elicited by a small “control” spot of light of constant intensity also confined to the facet of the test receptor. Scale of ordinate on the right.

The lower (curvilinear) graph is the frequency of discharge from the same test receptor, when the mask was removed and the entire pattern of illumination was projected on the eye in various positions, minus the frequency of discharge elicited...
Students can find a simple solution to this model, on a grid of just 8 points:

\[ a=0.7; \quad b=a^4; \quad c=a^9; \]

\[
M1 = \begin{bmatrix}
    a & b & c & 0 & 0 & 0 & 0 & 0 & 0 & 0 \\
    b & a & b & c & 0 & 0 & 0 & 0 & 0 & 0 \\
    c & b & a & b & c & 0 & 0 & 0 & 0 & 0 \\
    0 & c & b & a & b & c & 0 & 0 & 0 & 0 \\
    0 & 0 & c & b & a & b & c & 0 & 0 & 0 \\
    0 & 0 & 0 & c & b & a & b & c & 0 & 0 \\
    0 & 0 & 0 & 0 & c & b & a & b & c & 0 \\
    0 & 0 & 0 & 0 & 0 & c & b & a & b & c \\
    0 & 0 & 0 & 0 & 0 & 0 & c & b & a & b \\
    0 & 0 & 0 & 0 & 0 & 0 & 0 & c & b & a \\
\end{bmatrix}
\]

\[ E = [0 \ 0 \ 0 \ 0 \ 0 \ 1 \ 1 \ 1 \ 1 \ 1 \]';

\[ F = (\text{eye}(10) + .5*\text{M1})\backslash E; \]

Hartline and Ratliff’s model was a bit more elaborate than that -- but not a lot more. We have exploited an analogy to a physics subject: antiferromagnetism.
“Is that all? Is anything new going on?”
Oh, yes. Perhaps you are tired of the diffraction limit? Perhaps you want to image things smaller than the wavelength of light? Perhaps you want to invent a method that gets named “Method of the year” by Nature Methods?

Understanding the statistical character of light has led to subdiffraction microscopy methods like PALM, STORM, FIONA... Now we can see.
It turned out we could not understand vision at all without some top-drawer ideas from fundamental physics (like quantum theory). Other cool ideas entered too (stationary-phase, antiferromagnetism...). In fact, I’d say that biophysical contexts are a compelling way to introduce many of these ideas to any student, interested in Biophysics or not.

Now let’s step back from those specifics.

There is a cohort of students who are very interested in Physics, but who don’t pursue it. They (or their parents) think of “physicist” as something akin to “professional poet.” What would you do with that?? They feel they must study something like Biochemistry or Medicine if they want to be employed some day.

★ If we can meet these students halfway, show them a little bit of what’s really happening in Biophysics, everyone can win. For example, we can use biophysical ideas to introduce statistical physics, or even quantum physics. We need to keep the biophysics in Physics.

★ But we shouldn’t be ashamed of, or downplay, fundamental physical ideas. These students after all, love the Big Ideas, and those ideas really matter in the latest biomedical research.

★ What’s more, physical models are weirdly, unreasonably effective in stripping away the inessential from a biological system. We need to keep the physics in Biophysics.
Thanks

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For a syllabus and course materials, please contact me:
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