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Author

Fletcher, Daniel A

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"Bottom-up biology: Harnessing engineering to understand nature"

Dan Fletcher

UC Berkeley

Abstract

Engineering as a field has fundamentally different goals than biology, but the perspective engineers take – that systems can be designed and built – is helping to motivate and equip efforts to understand and construct biological systems from the bottom up.

Engineering could be viewed as the antithesis of nature. Engineers want control, reproducibility, and specific outcomes, as do all of us who rely on the bridges, planes, and computers they build. Structures and systems constructed by humans are intentionally different from those cultured or grown from the primordial soup. Engineered systems are designed to perform well-defined roles using well-defined rules based on well-defined materials. Nature produces marvels of chemistry and physics whose roles, rules, and materials we are far from understanding at the same level of detail. But there is at least one clear point of connection. Both biological systems and engineered systems are redesigned and replaced when they don't perform, albeit one by evolution and the other by customer dissatisfaction (remember the Apple Newton?).

So what contributions can an engineering perspective make to biology? Quite simply, it is the idea that biology can be built. No doubt all who have marveled at a *Xenopus* embryo dividing in synchrony or watched the speedy growth of a bacterial culture realize that biology is built by something. Namely by itself. We are also aware of our power to change bits and pieces of cells through genetic modifications or environmental conditions, as well as to bias organism traits through selection. An engineering perspective offers something different – the idea that biological systems can be built by us.

This notion is not surprising to the early enzymologists and biochemists who were able to reproduce in vitro the same reactions cells use to synthesize molecules in vivo. Nor is it surprising to the biophysicists who study in great detail each and every step of a motor protein as it carries cargo along a microtubule track or slides two actin filaments relative to one another. In theory, it is an idea that all biologists subscribe to at some level if they believe in the central dogma. Breaking down biological systems into their parts, and characterizing each one, has been and continues to be a critical part of the enterprise of biological research. Such an approach is referred to as 'reductionism' and has inspired a reductionist agenda to understand the molecular basis of cell behavior (Pollard, 2003).

Yet for some, reductionism is by its very nature missing the point. The tissues and organisms that we seek to understand, particularly in fields like cell and developmental biology, are not

necessarily revealed in all their glory by the better understanding of an enzyme or two in dilute solution in a plastic tube at room temperature. Something more remarkable occurs when all the parts of a system, be it a gastrulating embryo or a fusing muscle cell, come together in a spatially organized way to produce a function not obvious from the smallest parts.

The importance of the whole was highlighted by cell and developmental biologist Paul A. Weiss (1898–1989) in a talk entitled “From Cell to Molecule” (Weiss, 1962). As molecular biology was vitalized by its successes with the structure of DNA and enzymology, he made the point that going the other direction – from molecule to cell – was the harder and more important goal of biology. To illustrate this, he showed three vials, one containing a whole chick embryo, the next a homogenized chick embryo, and the third a homogenized and fractioned chick embryo (the original image is worth a look). Weren’t the contents of the three vials – whole, homogenized, and fractioned – the same, and yet the biology completely different? Isn’t it the hierarchical spatial organization that turns molecules into organisms? How do we understand that process?

The answer is not to abandon reductionism. Going against the idea that biology can be understood from its parts is to tread uncomfortably close to vitalism, the old idea that something special occurs when the parts that make up living organisms come to life. Without doubt the interactions between molecules, between cells and their environment create the possibility for different physical and chemical processes that give rise to collective behavior. However, the study of complex systems is stymied if those systems are both irreducible and unbuildable.

This is where an engineering perspective can help. The piston in a car engine is no more an explanation of the car than an actin filament is an explanation of a motile cell. But in the car, the link between the part and the system is clear and precise because the properties of the piston are well known, and mathematical models accurately predict its movement and connection to other parts of the car. And, importantly, because we can build cars to test our models.

Biologists have done extraordinarily well at characterizing biological systems and their molecular parts, especially with recent advances in fluorescence microscopy, force microscopy, mass spectrometry, and cryo electron microscopy. Many have also begun to embrace the role of mathematical modeling of systems to test hypotheses and to employ the power of gene editing to modify or remove specific parts of their biological systems. But it is the last step – the building – that has remained largely out of reach. Because of that so has the satisfying connection between parts and systems that underlies the utility and capability of engineered systems, as well as a sense of confidence that they are understood.

In practice, the idea that biology can be built by us becomes rather hazy when one goes beyond a few molecules purified from cells to consider the cells themselves and the tissues they form. Thanks to clever genome engineering strategies, we now know that the DNA of a cell can be completely replaced with a synthetic chromosome (Gibson et al., 2010). A very impressive accomplishment, but it still leaves unbuilt, at least by us, the original

compartment and organization of the cell that enables it to make use of the DNA. Would loading that minimal genome into an in vitro translation/transcription system produce a bacterium? Likely not. Swapping out the pilot of a plane without it crashing is different from understanding how planes fly.

Therein lies the ‘constructionist’ agenda. Thanks to the success of biochemical reconstitutions that proved the chemistry of biological systems can be recapitulated in vitro, we are now in a position to take the next step toward constructing spatially organized biological systems with functions like those seen in nature. The parts involved in numerous biological processes have been identified, purified, and studied. Many more have been identified in genomes and cataloged in databases.

Yet how to actually put these parts together is not clear. Self-assembly works locally to drive, for example, polymerization of actin or oligomerization of receptors. As Weiss showed, however, self-assembly cannot be relied on to put together entire organisms from a pool of parts. Order of assembly and the role of boundary conditions must be considered, as they are in any engineered system. Tests would need to be developed to know if each sub-assembly is working, and uncharacterized genes and gene products would need to be studied. The challenge is so monumental that it is not at all obvious that building a cell from scratch is even possible. But neither is it obvious why it should not be possible.

Though constructing a cell de novo may be a grand but distant goal, an especially rich vein of reconstitution research has emerged at the level of the plasma membrane. Here spatial and temporal control is proving essential – control of information transfer during signaling, control of shape during trafficking, and control of size during growth. Recent examples of reconstitution of each of these aspects show how constructing systems from known parts is providing important new insight into biological control.

In the exploration of signaling regulation at the plasma membrane, reconstitution of protein clustering associated with transmembrane signaling has revealed roles for multiple distinct mechanisms. Multivalent interactions between the cytoplasmic proteins Nck and N-WASP and membrane-bound nephrin, an adhesion molecule important for kidney function, form two-dimensional phase-separated clusters (Banjade and Rosen, 2014). This concept that has been extended in a remarkable 12-component reconstitution of T cell receptor signaling on supported bilayers (Su et al., 2016). Spatial organization of membrane proteins can also be driven by entirely passive size-based segregation at membrane interfaces (Schmid et al., 2016).

Reconstitution of membrane deformations in vitro have demonstrated how protein binding and bending are coupled. ESCRT-III polymerization on membranes is promoted by negative membrane curvature (Lee et al., 2015) and forms spirals that flex to induce membrane deformation (Chiaruttini et al., 2015). Bending of membranes can also be achieved by crowding of endocytic proteins to form tubules (Stachowiak et al., 2012).

Incorporation of compartment boundaries into reconstitutions has shown their importance for size scaling during growth. The Min system in bacteria, which oscillates between the poles of a bacterial cell, forms self-organized waves of protein density that depend on

compartment geometry and can target the prokaryotic tubulin homolog FtsZ to mark the site for cell division (Zieske and Schwille, 2014). Compartments also influences the size of *Xenopus laevis* spindles in droplets of cytoplasmic extract, with increasing droplet volume producing a longer spindle up to a plateau size (Good et al., 2013; Hazel et al., 2013).

A common and valid question about such reconstituted systems is whether they are really the same as wild type biology. Certainly not. As is said about mathematical models, all reconstitutions are wrong, but some may be useful. When they are useful, they reveal mechanisms behind emergent behavior that were obscured by the bounty of emergence that is a live cell. When they are not useful, they can end up as aggregates at the bottom of a tube, quickly to be forgotten. But even those failures tend to tell us something about our ability to construct biological systems in the same way as engineered systems – we are missing something. Perhaps a part, a boundary condition, or an essential technique is still needed to build the system.

Compared to any engineering manufacturing plant, we do lack many of the tools needed for building biological systems. Indeed, the equipment in most cell and molecular labs is there to help tear things apart – blenders, centrifuges, chromatography systems – not put them back together. Mixing solutions with pipettes works so long as self-assembly is all that is needed, but that does not appear to do the trick.

Cells, it should be noted, cheat at construction. De novo assembly must have happened at least once in history, but subsequent building of new cells takes advantage of templating by parent cells. No need to assemble a brand new plasma membrane from parts on your own when you can steal mom and dad’s to make your new home. Exactly when, where, and how that assembly takes place – templated or not – and how assembly of sub-systems connects to construction of the whole system, is the question at hand.

Techniques originally from other fields are being used to add spatial and mechanical control to in vitro reconstitutions. Lithography techniques originally developed for the semiconductor industry are enabling spatial control of actin filament nucleation (Reymann et al., 2010). Optical tweezers first developed to manipulate microparticles are being used to uncover the mechanics of the ribosome (Goldman et al., 2015), and atomic force microscopy originally developed as a surface profiling tool is enabling control of forces on reconstituted actin networks (Bieling et al., 2016).

New technologies are still needed. Defining the hierarchical organization needed for a biological system to function will require temporal as well as spatial control of reactions that balance self-assembly and self-organization, the energy-consuming non-equilibrium cousin of equilibrium self-assembly. Maybe one day we will have a machine, not unlike a flow cytometer, that takes in soluble proteins, membrane proteins, metabolytes, DNA, and RNA, mixes them in just the right ways to kick off metabolism to pop out self-replicating cells. Yes, much like cells do now, but with us at the controls.

Building biological systems has long been the domain of science fiction and more recently the pursuit of synthetic biology. The prospect of Mary Shelley’s Victor Frankenstein conjuring his creature to life is frightening although unlikely, but the possibility that

synthetic organisms could have catastrophic impacts on society is a more recent and ongoing concern raised by genetic engineering and synthetic biology. The potential of technology to be good and bad, be it bombs or viruses, is an age-old risk of a world run by humans. More power to control biology is inevitably associated with more possibility for misuse and responsibility for careful thought.

Is building biological systems really a grand goal for biology? Perhaps a few modifications are all that is needed to safely cure a disease or create an organism that efficiently produces fuel. Such advances would represent a tremendous success for biology and important contribution to society, and they are rightly the focus of many researchers' efforts.

But the fundamental workings of any system, whether a watch or a water bear, will remain an irresistible curiosity for those who aim to make sense of nature – not only what biological systems do, but how exactly do they do it. And with better understanding of fundamentals will come better abilities to construct and control, exactly the sequence of events that gave rise to the useful engineered systems in our lives.

Of course, an engineering perspective on biology should not be taken too far. The quasi-static and equilibrium assumptions built into many engineered systems don't map well to the dynamic and non-equilibrium reality of biological systems. Even culturing cells is sometimes mysterious enough that we resort to anthropomorphizing them as being "happy" or "sad" (imagine an "unhappy" airplane). But we have far from exhausted the data we can collect, the experiments we can conduct, and the models we can build to better understand the subjects of our research.

Ultimately, whether we can reconstitute functional biological systems more complex than a dozen or so molecules remains to be seen. The constructionist agenda may be a short one, limited by the overwhelming complexity and inherent variability of systems that mix soft matter physics and biochemistry with impossible goals. Maybe unforeseen limits will prevent us from defining, building, or knowing that we have built a biological system like one produced by nature. Perhaps reconstitution will become a Sisyphean task pursued only by naïve graduate students and post-tenure professors.

The success of engineering provides at least some motivation for continued work. Though we do not hold in our heads exactly how all the transistors and logic of an integrated circuit interact to carry out complex tasks, we do know how the individual parts work and have models that link the parts with the whole. And we built it.

Although we are a long way from addressing the challenge that Weiss presented to rebuild a chick embryo from its parts, this is an exciting time to apply an engineering perspective to biology. Perhaps understand biological systems so well that we can engineer them from the bottom up is the ultimate, if unintentional, goal of biology.

References

Banjade S, and Rosen MK (2014). Phase transitions of multivalent proteins can promote clustering of membrane receptors. eLife 3.

Bieling P, Li T-D, Weichsel J, McGorty R, Jreij P, Huang B, Fletcher DA, and Mullins RD (2016). Force Feedback Controls Motor Activity and Mechanical Properties of Self-Assembling Branched Actin Networks. Cell 164, 115–127. [PubMed: 26771487]

Chiaruttini N, Redondo-Morata L, Colom A, Humbert F, Lenz M, Scheuring S, and Roux A (2015). Relaxation of Loaded ESCRT-III Spiral Springs Drives Membrane Deformation. Cell 163, 866–879. [PubMed: 26522593]

Gibson DG, Glass JI, Lartigue C, Noskov VN, Chuang R-Y, Algire MA, Benders GA, Montague MG, Ma L, Moodie MM, et al. (2010). Creation of a bacterial cell controlled by a chemically synthesized genome. Science 329, 52–56. [PubMed: 20488990]

Goldman DH, Kaiser CM, Milin A, Righini M, Tinoco I, and Bustamante C (2015). Ribosome. Mechanical force releases nascent chain-mediated ribosome arrest in vitro and in vivo. Science 348, 457–460. [PubMed: 25908824]

Good MC, Vahey MD, Skandarajah A, Fletcher DA, and Heald R (2013). Cytoplasmic volume modulates spindle size during embryogenesis. Science 342, 856–860. [PubMed: 24233724]

Hazel J, Krutkramelis K, Mooney P, Tomschik M, Gerow K, Oakey J, and Gatlin JC (2013). Changes in cytoplasmic volume are sufficient to drive spindle scaling. Science 342, 853–856. [PubMed: 24233723]

Lee I-H, Kai H, Carlson L-A, Groves JT, and Hurley JH (2015). Negative membrane curvature catalyzes nucleation of endosomal sorting complex required for transport (ESCRT)-III assembly. Proc. Natl. Acad. Sci. U. S. A 112, 15892–15897. [PubMed: 26668364]

Pollard TD (2003). The cytoskeleton, cellular motility and the reductionist agenda. Nature 422, 741–745. [PubMed: 12700767]

Reymann A-C, Martiel J-L, Cambier T, Blanchoin L, Boujemaa-Paterski R, and Théry M (2010). Nucleation geometry governs ordered actin networks structures. Nat. Mater 9, 827–832. [PubMed: 20852617]

Schmid EM, Bakalar MH, Choudhuri K, Weichsel J, Ann HS, Geissler PL, Dustin ML, and Fletcher DA (2016). Size-dependent protein segregation at membrane interfaces. Nat. Phys advance online publication.

Stachowiak JC, Schmid EM, Ryan CJ, Ann HS, Sasaki DY, Sherman MB, Geissler PL, Fletcher DA, and Hayden CC (2012). Membrane bending by protein-protein crowding. Nat. Cell Biol 14, 944–949. [PubMed: 22902598]

Su X, Ditlev JA, Hui E, Xing W, Banjade S, Okrut J, King DS, Taunton J, Rosen MK, and Vale RD (2016). Phase separation of signaling molecules promotes T cell receptor signal transduction. Science 352, 595–599. [PubMed: 27056844]

Weiss P (1962). From cell to molecule In Molecular Control of Cellular Activity. (Allen JH, editor). McGraw-Hill, New York.

Zieske K, and Schwille P (2014). Reconstitution of self-organizing protein gradients as spatial cues in cell-free systems. eLife 3.