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**Perspective:**  
**Towards computer aided design of cellular structure**

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**Abstract**

Cells are complex machines with tremendous potential for applications in medicine and biotechnology. Although much effort has been devoted to engineering the metabolic, genetic, and signaling pathways of cells, methods for systematically engineering the physical structure of cells are less developed. Here we consider how coarse-grained models for cellular geometry at the organelle level can be used to build computer aided design tools for cellular structure.

**Introduction**

Cells represent the scale of organization at which life emerges from non-living matter. Although cells are sometimes viewed merely as amorphous bags of enzymes, in fact cells have highly complex and intricate structures. Cells are full of internal compartments (organelles) that serve the role of reaction vessels, analogous to the reactors inside a chemical factory. Cells are bristling with sensors for both the external world and internal conditions, and these sensors feed into a network of control systems. Far from being simple bags of enzymes, cells are intelligent nanomachines, and there is currently great interest in being able to re-engineer cell to address specific applications in medicine and biotechnology. An excellent example is the use of chimeric antigen receptor (CAR) T-cells to attack tumors (Lim and June, 2017).

Although much of the effort in engineering cells has been devoted to metabolic and signaling pathways, another aspect of cells that has received far less attention is their physical structure or geometry. In chemical engineering, the physical size and shape of the reaction vessels, columns, and pipes in the factory exert a large influence on the yield and purity of the chemical reactions taking place inside them. By paying more attention to the physical design of a cell, it may be possible to achieve better outcomes in metabolic engineering.

Here, we consider the geometry of a cell as resulting from the size and shape of its component organelles, together with their spatial arrangement inside the cell (Rafelski and Marshall 2008). The first order challenge is to be able to adjust the volume and surface area of organelles in order to optimize biochemical reactions involving that particular organelle. The underlying assumption behind such a goal

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3 is that the size and shape of an organelle can have an effect on the biochemical  
4 reactions it contains, for example by affecting flux across the bounding membrane  
5 or the capacity for storing intermediates inside the lumen. The idea that organelle  
6 size relates to function is supported by the observation that in cells specialized for  
7 particular functions, organelles involved in those functions are generally much  
8 larger than in other cell types. For example, cells specialized for secretion typically  
9 have highly enlarged endoplasmic reticulum and Golgi apparatus. A second reason  
10 to believe that organelle size and shape may affect biochemical function is the fact  
11 that most cancer cells have abnormal morphology at the organelle level. For the  
12 most part, these types of evidence are correlative, and there remains a need for  
13 clear experimental testing of the relation between organelle size and function.  
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16 Unlike the components of most human-built machines, organelles are highly  
17 dynamic structures that are constantly exchanging material with other parts of the  
18 cell. The structure of the cell that we see under the microscope is a snapshot in time  
19 of a constantly evolving dynamical system. The mechanisms by which a cell can  
20 regulate the size of such dynamic internal structures remain poorly understood  
21 (Chan and Marshall 2012). However, it has been shown that alteration in the  
22 trafficking systems that move membranes between organelles can be sufficient to  
23 change the steady state size (Chan and Marshall 2014). Although many questions  
24 still remain about organelle size control systems, we believe the time has to come  
25 start grappling with the question of how we might be able to harness these  
26 pathways to design cells with customized structures.  
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### 31 **CellCAD: Computer Aided Design for Cells**

32 The complexity of biological systems has prompted efforts to automate the  
33 design process. CAD tools have been developed for DNA folding (Douglas 2009), for  
34 metabolic pathways (Rocha et al., 2010; Cardoso et al., 2018), and for gene  
35 expression networks (Nielsen 2016). However, no such tools exist for designing the  
36 physical structure of cells at the organelle level.  
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38 In many cases, forward genetic screens have already revealed mutations that  
39 result in alterations of organelle size. Such existing mutations can in principle be  
40 used to produce cells with altered geometry. However, such an approach has  
41 limitations. If the size and shape of the organelle desired for a particular  
42 engineering purpose happens to coincide with a size and shape that has been  
43 reported in a forward genetic screen, then of course it would be straightforward to  
44 simply use that mutation, and no further modeling or work would be required. This  
45 approach is similar to the method used to build stone walls in historical times, when  
46 a wall-builder would simply select stones that happened to be the right size and  
47 shape to fit into a wall as it is built. The respect that we feel for the artisans capable  
48 of such work is due in large part to our recognition of how difficult such an approach  
49 is. In engineering cells, it would be highly desirable to be able to pick whatever  
50 sizes and shapes we want for the organelles, and then be able to make cells that  
51 meet that specification. Such an ability would justify using the term “engineering” in  
52 the first place. A second potential limitation to directly using mutants from  
53 traditional screens is the challenges that arise when attempting to modify more than  
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3 one organelle at a time. One way to think about this issue is to ask whether different  
4 organelles are independently addressable in the sense that mutations can alter the  
5 geometry of a single organelle without affecting any others. Many instances are now  
6 known in which organelles interact physically with each other at defined contact  
7 sites (Elbaz and Schuldiner 2011). Such interactions suggest that altering one  
8 organelle could have indirect affects on others. A recent study examined a set of  
9 numerical descriptors of size and shape for several subcellular structures and used  
10 principal components analysis to identify groups of co-varying morphology features  
11 that accounted for most of the variation in the cell (Chang and Marshall 2019). One  
12 result of that analysis was that each of the principal components included  
13 substantial contributions from all cellular structures examined, providing empirical  
14 support for the idea that distinct organelles are not independently addressable. It is  
15 therefore highly likely that whenever two or more mutations are combined that  
16 were identified in screens for different organelles, the combined result of the double  
17 mutant will be difficult to predict. For these reasons, we believe that an alternative  
18 approach based on mechanistic understanding of organelle biogenesis pathways , if  
19 converted into the appropriate mathematical models, may be helpful in designing  
20 cells of a desired geometry.  
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24 We envision an approach in which coarse-grained mathematical models are used  
25 to build a computer aided design (CAD) system for cellular structure (Figure 1). The  
26 first step would be to specify the desired structure of a cell. How exactly to do this is  
27 still an open question. One could draw a cell, like a blue print, however any such  
28 drawing might only represent one exemplar of a range of cells that would, for a  
29 given purpose, still be considered to have equivalent shapes. An alternative would  
30 be to specify the volume, surface, or other morphological parameters for each  
31 organelle, without necessarily specifying their exact shapes or arrangements. Such  
32 an approach is more aligned with the ultimate purpose of the design, but may be  
33 harder for the user to visualize. In any case, once the user has specified a design, the  
34 result will be a list of numerical values, or ranges of values, that specify a position in  
35 the total space of cell morphologies. The question becomes, given that wild-type  
36 cells occupy a particular region of morphology space, what molecular alterations (to  
37 be implemented by genome engineering) are required to move the cell to the  
38 desired new region of morphology space.  
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42 Several strategies could be imagined for carrying out CellCAD. Here we consider  
43 a strategy based on coarse-grained mathematical models, in which the size and  
44 shapes of organelles are described by real variables, and the processes of organelle  
45 dynamics are represented by terms in differential equations governing each  
46 organelle feature. For some organelles, such models already exist, but for many  
47 others, modeling organelle dynamics is still a goal of current research. Each model  
48 will be characterized by a number of adjustable parameters, such as rate constants  
49 or diffusion constants. Given a particular set of values for these parameters, the  
50 differential equations can be solved (usually numerically) to produce a simulation  
51 or prediction of how the cell geometry will change over time. We are generally most  
52 interested in the steady state solution, which would describe the final geometry of  
53 the cell after it has adjusted to any change in parameters. For design purposes, the  
54 challenge is to start with the desired end-point, and work backwards to infer what  
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parameter values will, when plugged into the differential equations, yield the desired result (Figure 2). This model inference problem is extensively studied in the context of engineering and control systems, and a number of mathematical methods are being developed for solving this problem in complex biological systems (Babtie and Stumpf 2017).

In this Perspective we consider the feasibility of this approach by examining some of the challenges that CellCAD would face. Obviously a major challenge is having sufficiently reliable predictive models that include all the relevant biological details. We will consider a plug-and-play approach to building organelle dynamics models, but first we ask, given that we can build such models, would CellCAD be possible. Specifically, we consider what constraints such a program would need to satisfy. We will consider three questions that must be answered to design a CellCAD program: realizability, uniqueness of solutions, and biological implementability.

The question of **Realizability** asks, if one chooses a set of parameter values at random, how likely is that combination of parameters to yield a solution that is physically possible, i.e., one in which the contents of the cell can fit inside, and in which the surface to volume ratio of all compartments is greater than or equal to the limiting value for a sphere. Can we really just pick any combination of parameters? This question has been extensively studied and solved in the context of electrical engineering (Zemanian 1996) but we believe that cell biology presents its own challenges. **Uniqueness** describes the mapping from parameters onto geometry, i.e., if an engineer wanted to make a cell with a specific geometry (or if that geometry was selected by natural selection), is there a unique set of parameters that will give the desired geometry, or is there a large space of possible solutions? Finally, **Implementability** asks whether a set of parameter values can be brought about using realistic molecular manipulations such as gene knockouts or over-expression.

This Perspective will address these basic aspects of the cellular design space in the context of coarse-grained models of organelle dynamics, after which we will consider approaches for formalizing the model building process itself.

### **Realizability: Implications of a vesicle exchange model**

We start with a model in which there are two organelles exchanging vesicles with each other (Figure 3). In an actual cell, one of these might correspond to the Golgi apparatus and the other to a lysosome or vacuole, but here we are just considering two abstract membrane bound compartments without reference to any specific existing organelles. We represent the surface area of each organelle as  $S_1$  and  $S_2$ . We make the assumption that vesicles bud from the surface of each organelle at a rate proportional to the surface area. To our knowledge the rate of budding has not been measured as a function of organelle area, however the most reasonable assumption is that budding occurs at a rate proportional to surface area. We further assume that each organelle buds spherical vesicles with a uniform, possibly organelle-specific, radius. We let  $r_1$  and  $r_2$  be the characteristic radius of vesicles that bud from organelles 1 and 2. It is assumed that every vesicle budding from one organelle will fuse with the other organelle. In this simplest version of the model, we assume that fusion is instantaneous.

We obtain the following differential equation describing the dynamics of the surface area of each organelle

$$\frac{dS_1}{dt} = 4\pi r_2^2 a_{21} S_2 - 4\pi r_1^2 a_{12} S_1 \quad 1$$

$$\frac{dS_2}{dt} = 4\pi r_1^2 a_{12} S_1 - 4\pi r_2^2 a_{21} S_2 \quad 2$$

Each vesicle that moves between the organelles carries a fixed quantity of surface area as well as a fixed quantity of volume. We can therefore write a second pair of equations to describe the dynamics of the volume of the two organelles

$$\frac{dV_1}{dt} = \frac{4}{3}\pi r_2^3 a_{21} S_2 - \frac{4}{3}\pi r_1^3 a_{12} S_1 \quad 3$$

$$\frac{dV_2}{dt} = \frac{4}{3}\pi r_1^3 a_{12} S_1 - \frac{4}{3}\pi r_2^3 a_{21} S_2 \quad 4$$

Note that in this pair of equations, the rate of change of volume only depends on the surface areas of the two organelles, because the volume changes are due to budding and vesicle fusion, which only depend on surface area.

We now solve for the steady state solution of equations 1 and 2 to obtain

$$S_1 = \frac{r_2^2 a_{21}}{r_1^2 a_{12}} S_2 \quad 5$$

Finally, we assume that there is a fixed quantity of membrane available from which to construct the two organelles, which we call  $S_{tot}$ . With this assumption we can find the steady state values of  $S_1$  and  $S_2$  as follows

$$S_1 = \frac{S_{tot}}{1 + \frac{r_1^2 a_{12}}{r_2^2 a_{21}}} \quad 6$$

and

$$S_2 = S_{tot} \left[ 1 - \frac{1}{1 + \frac{r_1^2 a_{12}}{r_2^2 a_{21}}} \right] \quad 7$$

It is clear by inspection that any value we choose for the four model parameters  $a_{12}$ ,  $a_{21}$ ,  $r_1$  and  $r_2$ , will yield a physically realizable solution with respect to surface area, in the sense that the surface areas  $S_1$  and  $S_2$  will always be nonzero and less than  $S_{tot}$ .

What about volume? If we insert the steady state solutions for  $S_1$  and  $S_2$  into equations 3 and 4, we can ask whether this also yields a steady state solution with

respect to volume. Are there any parameter choices where this would be true? By solving equation 3 for the steady state in which V1 does not change, we obtain the criterion:

$$S_1 = \frac{S_{tot}}{1 + \frac{r_1^3 a_{12}}{r_2^2 a_{21}}} \quad 8$$

But for this to be true at the same time that equation 6 is true, it must be the case that:

$$\frac{r_1^3 a_{12}}{r_2^3 a_{21}} = \frac{r_1^2 a_{12}}{r_2^2 a_{21}} \quad 9$$

from which we derive the necessary condition

$$r_1 = r_2 \quad 10$$

Unfortunately, this criterion for existence of a steady state solution, namely that all transport vesicles have the same radius, is not, in fact, met by actual vesicles found in the cell. The largest transport vesicles tend to be clathrin-coated vesicles which are 50–100 nm diameter (Heuser 1980 Bonafacino 2003). Other types of vesicles of intermediate size include COPII-coated vesicles (60–70 nm) originating at the ER, and intra-Golgi transport vesicles (70–90 nm) (Vigers et al., 1986; Balch et al., 1994; Orci et al., 2000; Sato and Nakano 2007; Verissimo and Pepperkok 2013). At the small end of the size range are synaptic vesicles (40nm; Zhang 1998) and intracellular nanovesicles known as INV which have diameters of 30 nm (Larocque 2019). These size variations show that the size of a transport vesicle is strongly dependent on the organelle from which it originates. Even within a single Golgi stack, the diameter of COPI buds and vesicles increases from around 45 nm on the cis side to 60 nm on the trans side (Bykov et al., 2017). In many cases, these size differences result from the different proteins that mediate vesicle formation, for example via proteins that affect the curvature of the vesicle, thus fixing the radius (Bonafacino 2003; Miller 2015).

Given the size differences in vesicles moving in the two directions between the organelles, we conclude that the simplistic toy model presented here cannot result in a steady state solution for both surface area and volume. No matter how we may decide to choose parameters, the volumes will not achieve a steady state even though the surface areas may do so. Because the two organelles would continue to exchange vesicles even after the surface areas had reached steady state values, the organelle from which the smaller vesicles was budding would send less volume, for a given surface area, than it received from the other organelle, and would thus shrink until its internal volume approaches zero, potentially compromising its biochemical function. The other organelle would receive more volume per vesicle

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3 surface area than it donated to the other organelle, and would therefore continue to  
4 grow until its surface to volume ratio decreased below the minimal surface to  
5 volume ratio corresponding to a sphere, in which case it would explode. There are  
6 several potential solutions to this dilemma.  
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9 First, it is not necessarily true that organelle volume only changes due to vesicle  
10 trafficking. Lipid bilayers are not completely impermeable to water, potentially  
11 allowing excess volume to escape if an organelle became overly inflated. Water  
12 channels can also assist in this flow. In either case, an organelle operating in such a  
13 regime would assume the shape of a sphere, since water is being expelled by  
14 mechanical pressure from the membranes surface tension. So while this type of  
15 mechanism might prevent organelle explosion, it would require that organelle shape  
16 be close to spherical, which is true for some organelles but not for others.  
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20 Second, our simplified model has assumed that vesicle budding and fusion occur at  
21 rates that are independent of organelle volume. This need not be the case.  
22 Biochemical processes taking place on a membrane surface can be sensitive to the  
23 curvature of the membrane, or to membrane tension, and such regulation could  
24 allow vesicle budding or fusion events to be increased or decreased to avoid  
25 reaching a bursting point. For example, when an organelle membrane became  
26 overly stretched, further fusion of vesicles would be mechanically inhibited. One  
27 clear example is Rab-mediated membrane fusion of the yeast vacuolar membrane,  
28 which is directly sensitive to osmotic conditions (Brett and Merz 2008).  
29 Alternatively, the cell might have some way to actually measure organelle volume  
30 and adjust rates accordingly.  
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34 Finally, many organelles can undergo additional processes to alter surface area and  
35 volume that have not yet been discussed. De novo membrane biosynthesis can  
36 increase surface area without increasing volume, and could allow a swelling  
37 organelle to keep pace with the increase in volume that might be driven by vesicle  
38 trafficking. Organelles can also undergo fission into multiple smaller organelles, or  
39 else fusion to produce a smaller number of larger organelles. These whole-organelle  
40 fission and fusion processes result in instantaneous changes in the surface to  
41 volume ratio. Many organelles contain proteins in their membranes that affect local  
42 membrane curvature (Simunovic et al., 2015). Such constraints on the average  
43 curvature will restrict the organelle to a certain range of surface to volume ratios.  
44 Attempts to move the surface to volume ratio outside of this range would be  
45 opposed by the energetic cost of deforming the curvature sensing proteins.  
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50 A realistic model of organelle dynamics would thus need to include a variety of  
51 processes beyond vesicle budding and fusion, and should include the possibility that  
52 the rates of these processes may vary as a function of surface area or volume. A  
53 more realistic model would also take into account the fact that cells contain many  
54 different organelles that can exchange vesicles with each other. Each additional  
55 process that is included in the model will add more parameters to the model. Thus,  
56 a solution to the realizability problem may come at the price of reduced uniqueness.  
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## Uniqueness

Each model we write down creates a mapping between parameter space and solution space. Realizability considerations may reduce the range of parameter values that we are allowed to choose. Implementation considerations (see below) may limit the precision with which we can actually set the value of a given parameter. For this reason it is important to consider the degree to which a particular design can be achieved only by choosing a specific set of parameter values. If the solution has to be too precisely specified, it may be impossible to achieve in reality. This question of uniqueness also has interesting implications for the evolution of cellular structure. A structure that could only be achieved by a highly specific set of parameter values is less likely to occur in random variation than would be a structure that was more generic, in the sense that a larger portion of parameter space could achieve it. In the simple vesicle trafficking model discussed above, the surface areas are only determined by a single lumped parameter that involves all four model parameters:

$$\frac{r_1^2 a_{12}}{r_2^2 a_{21}}$$

Any given set of organelle surface areas would be achieved by any points on a level set within the four dimensional parameter space for which the value of this expression is a constant.

## Implementation: Once we choose parameter values, then what?

Finally, there is a question of implementation. Given a set of parameter values, do we have the necessary tools to create a cell in which those parameters have the desired values? In a typical model of any cellular process, different parameters will describe different aspects of the underlying molecules. Some parameters may describe the affinity of an enzyme for its substrate, others describe mechanical properties such as elasticity of cytoskeletal elements or diffusion constants, while other parameters may describe the quantity of a protein or its rate of expression. In a computational model, each of these parameters can be arbitrarily changed to whatever number we want, but this is not the case inside the cell. Some aspects of biology are easy to manipulate with modern molecular approaches, but others are far more difficult. The easiest change that we can make is to knock out a gene, so that a particular protein is entirely missing. Such a modification would generally cause one or more parameter values to be set to zero. If a specific non-zero value is desired, more subtle methods are available to alter the degree of gene expression (Aranda-Diaz 2017). Achieving precise control of gene expression through promoter swapping is still challenging, but by screening a collection of similar constructs it can be accomplished in most cases. A separate question is whether



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3 changing one promoter only affects expression of that particular gene, or whether  
4 many other genes may also be affected as an unintended side effect.  
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7 In the simple vesicle exchange model presented above, changing the radius of the  
8 vesicles would be extremely difficult because this is a product of the protein  
9 structures of the key proteins that organize the vesicles, such as clathrin or COPII.  
10 These proteins have been tuned by evolution to assemble into structures with  
11 particular curvature, hence alteration of the vesicles size would likely require  
12 extensive protein engineering. This is not to say that it cannot be done, just that it is  
13 relatively difficult on a routine basis. In contrast, the rates of budding can be  
14 controlled by the transcriptional levels of genes required for the two pathways. The  
15 best case scenario, from an engineering perspective, is when multiple parallel  
16 vesicles trafficking pathways exist, such that deletion mutations in one pathway  
17 result in a partial reduction of trafficking overall. Such an effect has been  
18 demonstrated in the yeast vacuole, in which loss of a pathway that brings vesicles  
19 away from the vacuole back to the Golgi causes a smaller steady state vacuole size,  
20 while loss of a pathway that brings vesicles from the Golgi to the vacuole causes and  
21 increased vacuole size, but even when the pathway to the vacuole is deleted, the  
22 vacuole does not disappear because other parallel pathways still bring membrane to  
23 the vacuole (Chan and Marshall, 2014). To the degree that other vesicle trafficking  
24 pathways are partially redundant, deletion mutants can similarly be used to achieve  
25 quantitative effects on trafficking rates.  
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### 30 **Towards a general organelle modeling framework**

31 A key requirement for the model-based design approach is having reliable coarse-  
32 grained models for all organelles of interest. Although each organelle is different in  
33 terms of the dynamic processes that govern its size, at an abstract level most  
34 organelles employ a limited, partially overlapping set of processes (Figure 4). Here  
35 we consider each in turn, with a focus on how they alter the number, surface area, or  
36 volume of the organelles (Table 1).  
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40 *Vesicle trafficking:* This has already been considered in the simplified model above,  
41 but it is important to note that the transfer of membrane from one organelle to  
42 another actually involves three processes. First, a vesicle must bud from the surface  
43 of one organelle. Then, it moves elsewhere in the cell, either by diffusion or more  
44 often pulled along by motor proteins. Given the importance of motors for moving  
45 vesicles, we can see that cytoskeletal organization may play a role in determining  
46 the efficiency of exchange between a particular pair of organelles. Finally, the  
47 vesicle must undergo fusion with the target organelle. These processes affect both  
48 surface and volume but do not alter the copy number of either organelle.  
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52 *Membrane biosynthesis:* Although vesicle fusion is one way for membrane to be  
53 added to the surface of an organelle, in other cases the membrane can be  
54 synthesized in situ. Enzymes responsible for producing phospholipids localize to  
55 the surface of some organelles and add to the membrane. This addition requires a  
56 suitable supply of precursor lipid. An interesting example is during the unfolded  
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3 protein response in yeast, during which the endoplasmic reticulum increases in size  
4 (Bernales et al., 2006). This requires formation of new lipid, and in fact triggering  
5 of this pathway leads to transcriptional upregulation of lipid synthesis enzymes  
6 (Cox et al., 1997). From the point of view of organelle size and shape, the primary  
7 effect of membrane synthesis is an increase of surface area without an increase in  
8 volume.  
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11 *Pumping:* As discussed above, organellar membranes can be permeable to water,  
12 and in other cases may contain water channels. One driving force for water to move  
13 across a membrane would be mechanical tension if the organelle was inflated up to  
14 the shape of a sphere. But another, probably more common driving force is osmotic  
15 force. As solutes such as proteins are concentrated inside an organelle, this will  
16 tend to drive an influx of water, thus driving an increase in volume of the lumen in  
17 two ways, by accumulation of material represented by the imported protein, and  
18 osmotically driven influx of water. From the point of view of organelle size and  
19 shape, the net effect is an increase of organelle volume without an increase in  
20 surface area.  
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24 *Fission and fusion:* Many organelles are capable of undergoing fission, producing  
25 two or more smaller versions of themselves, which can then undergo fusion to  
26 restore the original large size. Mitochondria, for example, undergo extensive fission  
27 and fusion, the net result of which is a complex interconnected network (Shaw and  
28 Nunnari 2002). Vacuoles also undergo fission and fusion, for example in response  
29 to changes in osmotic conditions (Bone et al., 1998). Fission and fusion have as  
30 their primary effect an alteration of organelle copy number, and do not generally  
31 involve a change in the total surface or volume. Obviously, however, if a large  
32 organelle undergoes fission the resulting new organelles will have decreased  
33 surface area and volume. If the fission is symmetric to produce equal sized  
34 organelles, this will not alter the surface to volume ratio, however it is possible for  
35 organelles to undergo asymmetric budding, producing one small and one large  
36 product, and in this case the resulting new organelles may have different surface to  
37 volume ratios.  
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42 *Differentiation:* Although we usually consider a given organelle to have a fixed  
43 molecular identity, there are cases where an organelle can change its molecular  
44 composition and begin to assume new functions. A commonly seen example is  
45 when chloroplasts differentiate into chromoplasts during the ripening of fruit. The  
46 photosynthetic machinery is largely eliminated and enzymes are imported that  
47 generate the pigment molecules that give ripe fruit its color. Such changes may well  
48 be accompanied by pumping of contents, vesicles trafficking with other organelles,  
49 and other dynamic processes, however to a first approximation we imagine that  
50 differentiation is primarily a matter of organelle contents, and need not alter the  
51 surface area or volume of the organelle. Differentiation will affect the organelle  
52 copy number, with each differentiation event decreasing the number of organelles  
53 of the original identity and increasing the number with the new identity.  
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3 *Autophagy*: Under conditions of stress, cells consume their internal components in  
4 order to harvest their molecular contents for survival. This process of autophagy  
5 involves formation of an internal membrane that surrounds a portion of the  
6 cytoplasm, including those organelles trapped within. In some cases this process  
7 can be highly specific for particular organelles, for example in the process of  
8 mitophagy whereby mitochondria are consumed (Youle and Narendra 2011).  
9 Autophagy results in a decrease in the number of whichever organelles are involved,  
10 but would not affect the size or shape of the organelles that were not consumed.  
11 The only potential exception to this statement is that autophagy may be size-  
12 selected, such that small organelles wind up inside the autophagic membrane while  
13 larger organelles are too big to be surround and hence do not get eliminated.  
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17 The importance of enumerating the classes of organelle dynamic processes is that  
18 each of these processes gives rise to a term in a differential equation governing the  
19 surface, volume, or copy number of an organelle. Thus, one only needs to invest the  
20 effort to develop mathematical models for each of these processes separately, and  
21 then they can be mixed and matched to produce a model for any organelle of  
22 interest.  
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25 Each of the processes shown in Figure 4 will contribute a term appearing in the  
26 appropriate equations for the organelles involved. For example, a differential  
27 equation governing the rate of change of the copy number  $N$  of a given organelle will  
28 encompass terms which will increase  $N$  (fission, differentiation from another  
29 organelle to the considered one) and terms which will decrease  $N$  (fusion,  
30 differentiation to another organelle from the considered one, autophagy) but no  
31 terms for, e.g. lipid biosynthesis, which will instead increase surface area without  
32 changing copy number. While the specific form of the equations may be tailored to  
33 specific organelles, we can imagine that a list of all such processes is possible, and it  
34 represents the basic ontology for a “plug and play” version of a general organelle  
35 model. In fact, we can even imagine that the dynamic nature of each process can  
36 codified in general, and then automatically be obtained for a new organelle by just  
37 tuning or fitting the parameters to the data. For examples, in general one can expect  
38 that fission may increase the copy number of the organelle linearly with the existing  
39 number of organelles, that is:  
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$$44 \quad \frac{dN_{fission}}{dt} \propto \phi_{fis} N$$

45 while the term for fusion, assuming the limit of homogenous mixing, may be written  
46 as:  
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$$49 \quad \frac{dN_{fusion}}{dt} \propto -\phi_{fus} N(N - 1)$$

50 where the rates are to be obtained from the data. Similar considerations can be  
51 used to develop formal expressions for the other processes in Figure 4. Additional  
52 external conditions may be included or folded into parameters values, if their effect  
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3 on the limited set of considered processes is known. Once a model of this kind has  
4 been written, the usual approaches of dynamical systems and control theory can be  
5 brought to bear on the problem. Parameter sweeps and steady state analysis can  
6 generate predictive hypotheses about which process would have the most impact on  
7 a desired design.  
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10 As such models start to be analyzed, it will be of great interest to see how the  
11 different processes interact with each other. We have seen above that in the course  
12 of achieving a stable steady state solution for surface area, a vesicle trafficking  
13 system creates a problem for regulating volume. When more processes are  
14 combined, if each one perturbs the others, it is not obvious what will happen.  
15 Investigating such models may be able to teach us lessons about why cells are put  
16 together the way they are.  
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### 19 **Conclusions and Prospects**

20 The growing understanding of organelle dynamics at a quantitative, predictive level,  
21 means that we should soon be able to use models as design tools to create new cell  
22 geometries never before seen. In order for this type of approach to become routine,  
23 we will need software to assist with model generation, to determine regions of  
24 parameter space that achieve a desired design, and to guide the construction of  
25 mutant strains in order to reach those parts of parameter space. Together, such  
26 algorithms would constitute a computer aided design system for cells. Outstanding  
27 questions remain, for example, what is the most natural or effective language in  
28 which to describe the desired cell geometry, or what is the role of data-driven  
29 modeling and machine learning in the complex process of cellular design. We  
30 believe that the effort to achieve computer aided cell design has the potential to  
31 uncover new fundamental aspects of cell biology by forcing us to clarify  
32 assumptions. This represents a highly interdisciplinary goal that we hope will  
33 motivate computer scientists and engineers to join forces with cell biologists in the  
34 future.  
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## Figure Legends

**Figure 1.** Computer-aided design of cells. Starting with a conceptual idea for a cellular structure that might be optimal for some specific purpose, the structure is specified within a computer, either by drawing a blueprint or by listing design specifications in terms of individual organelle size and shape parameters. This design information is then used to create a representation of the design target as a point in a multi-dimensional cell morphology space.

**Figure 2.** Model-driven CellCAD. Starting with a mechanistic model, formulated in terms of the molecular and cellular processes that underlie organelle dynamics, a coarse grained mathematical model is formulated in the form of a system of differential equations, which can then be solved to predict how the system will behave over time in response to various inputs or signals. The steady state solution of this model for organelle dynamics will be a prediction of the size and shape of the corresponding organelles, thus defining a position in the possible morphology space of the cell. This process can also be reversed, by starting with a desired solution, corresponding to a desired cell structure, and estimating parameters of the coarse grained model that yield the desired solution. This results in a model-based design tool.

**Figure 3.** Vesicle exchange model, showing two organelles in two different colors, each characterized by a surface area  $S$  and a volume  $V$ . Each organelle generates vesicles from its surface, indicated by the small spheres in the same color as the parent organelle. These vesicles then move to the other organelle and fuse with it, increasing its surface and volume while decreasing that of the parent organelle. In this model, each organelle donates vesicles to the other, with rates that are proportional to the parent organelle surface area with the rate constants  $a_{12}$  and  $a_{21}$ .

**Figure 4.** The fundamental operations of organelle dynamics. A. Vesicle trafficking, in which a pair of organelles exchange vesicles via a mechanism similar to the model shown in Figure 3. B. Membrane biogenesis, in which phospholipids are synthesized on the organelle surface, leading to an increase in surface area. C. Pumping, in which the import of proteins into an organelle lead to an increase in volume. D. Fission and fusion. In fission, an organelle splits into two disconnected organelles, which can also fuse back together. E. Differentiation, in which an organelle turns into a different type of organelle by changing its molecular composition. F. Autophagy, in which an organelle is consumed by the cell. The effects of these six fundamental operations on organelle number, surface area, and volume, are enumerated in Table 1.

**Table 1** A non-exhaustive list of organelle dynamic processes and the physical quantity they may impact.

<b>Process</b>	<b>Copy number</b>	<b>Surface area</b>	<b>Volume</b>
<i>Lipids biosynthesis</i>	No	Yes	No
<i>Pumping</i>	No	No	Yes
<i>Fission</i>	Yes	Yes	Yes
<i>Fusion</i>	Yes	Yes	Yes
<i>Vesicle trafficking</i>	No	Yes	Yes
<i>Differentiation</i>	Yes	No	No
<i>Autophagy</i>	Yes	No	No

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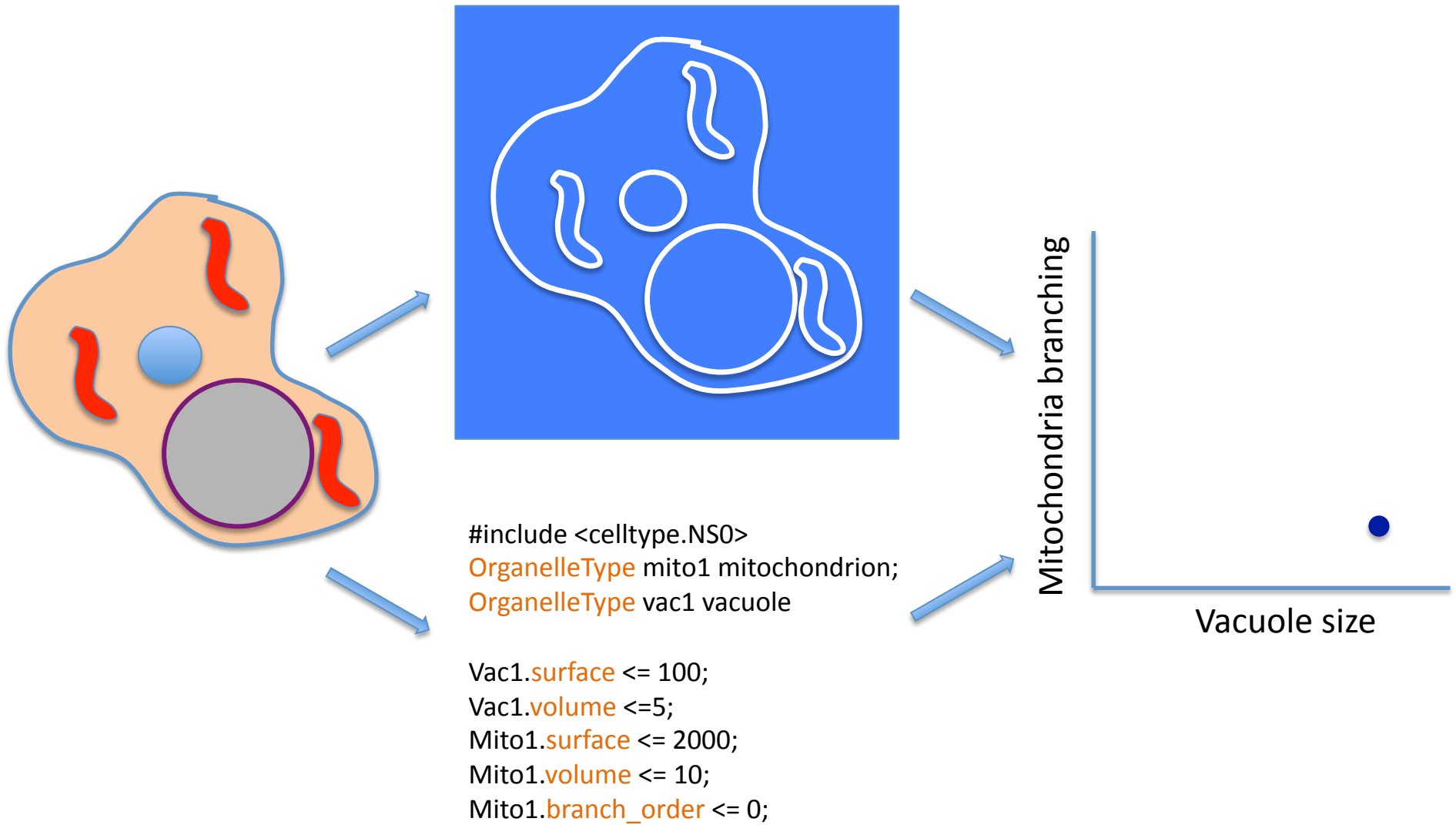
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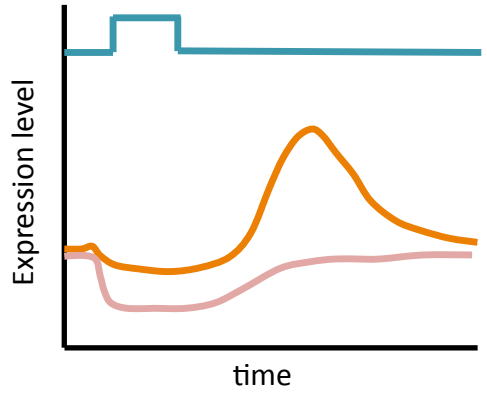
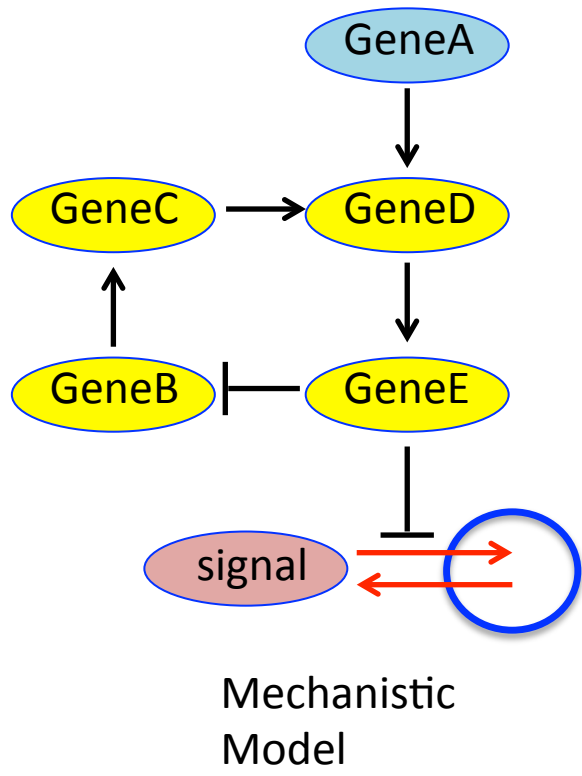
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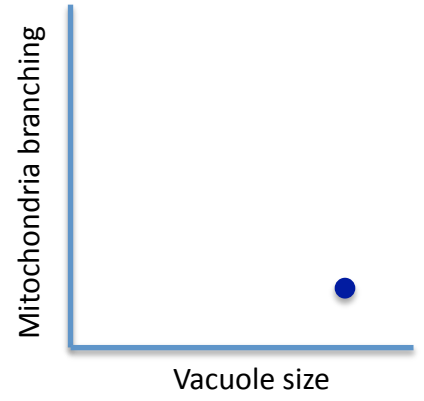


Coarse grained model

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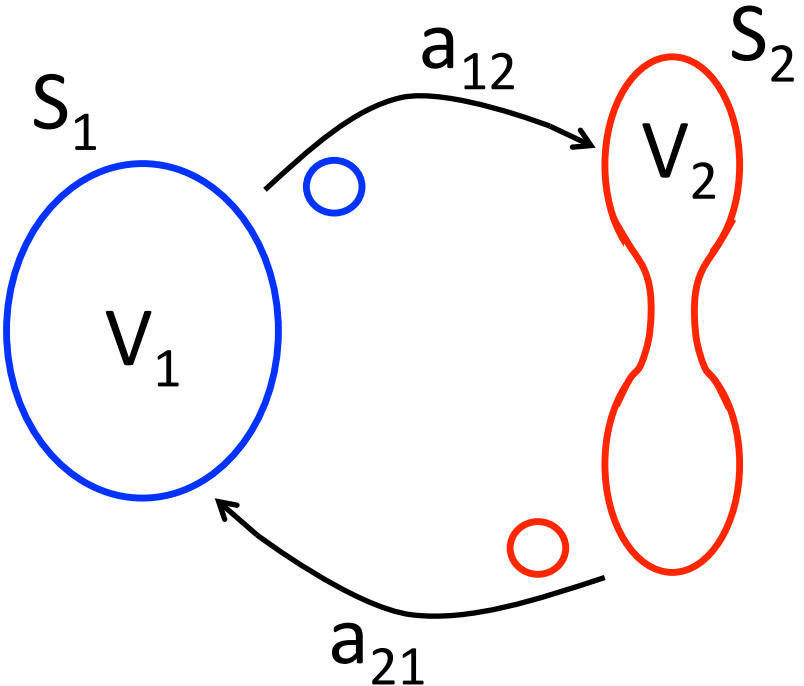
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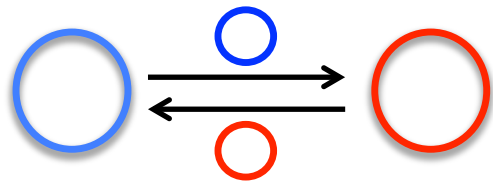
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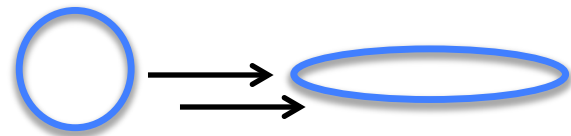


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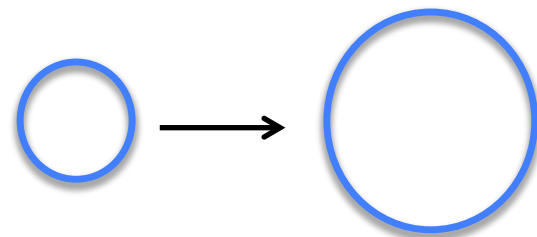
## A. Vesicle trafficking



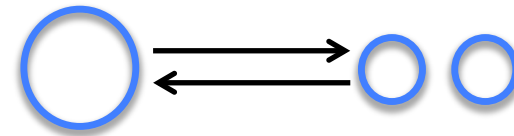
## B. Membrane biogenesis



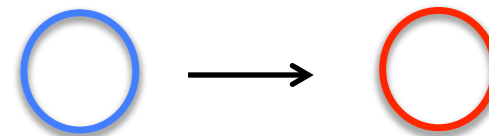
## C. Pumping



## D. Fission and fusion



## E. Differentiation



## F. Autophagy

