Available online at www.sciencedirect.com

ScienceDirect

Current Opinion in Structural Biology

Bringing macromolecular machinery to life using 3D animation

Janet H Iwasa

Over the past decade, there has been a rapid rise in the use of three-dimensional (3D) animation to depict molecular and cellular processes. Much of the growth in molecular animation has been in the educational arena, but increasingly, 3D animation software is finding its way into research laboratories. In this review, I will discuss a number of ways in which 3d animation software can play a valuable role in visualizing and communicating macromolecular structures and dynamics. I will also consider the challenges of using animation tools within the research sphere.

Address
Department of Biochemistry, University of Utah, United States

Corresponding author: Iwasa, Janet H (jiwasa@biochem.utah.edu)

Current Opinion in Structural Biology 2015, **31**:84–88

This review comes from a themed issue on **Macromolecular machines and assemblies**

Edited by **Katrina T Forest** and **Christopher P Hill**

http://dx.doi.org/10.1016/j.sbi.2015.03.015

Introduction

As molecular biologists, our energies are often focused on obtaining a deep and thorough understanding of a single or a small number of macromolecules (your favorite proteins, or YFPs). Why is YFP functionally important for the cell or an organism? Where does YFP localize within the cell, and how does it get there? What other macromolecules does YFP interact with, and how do those interactions impact YFP's role? What does YFP look like, and how might mutations or other alterations affect its structure and function? In this reductionist view, we can think of biologists as storytellers who use experimental evidence to weave together a rich narrative of the life and times of these specific macromolecules within the cell. This narrative can be condensed into a simplified visual depiction, the model figure, which often graces the final pages of a research manuscript. More often than not, however, the model figure fails to convey the richness of the original narrative it was meant to illustrate [1••]. How molecules move in 3D space and over time, for example, cannot be readily shown using a two-dimensional line drawing.

The rise in the popularity of 3D animation within the research sphere can be largely credited to its ability to visually capture and readily relate a rich and complex molecular narrative or hypothesis. The animation can be utilized in a number of outlets, including research seminars, course lectures, laboratory websites and supplemental figures. Studies on the use of molecular animation in educational contexts have shown that animation can have a positive impact on student learning, and particularly on retention [2,3], and related effects are probably experienced in seminar audiences as well.

That molecular animations can successfully depict a complex molecular process (particularly when compared to simplistic line drawings) is rarely contested. My own personal interest in 3D animation, however, lies largely in understanding how animation tools can be used by members of the research community to guide and refine their hypotheses.

Visualizing dynamic molecular machines

Some of the earliest examples of molecular movies depicted proteins that undergo significant conformational changes as part of their catalytic cycle. In 1995, Clemens Vonrhein and colleagues aligned 17 structures of nucleoside-monophosphate kinases solved by X-ray crystallography in order to create a 40-frame movie that illustrated a kinase moving from a closed to an open conformation [4]. In their concluding statements, the authors stated, 'Just like Daguerre's static pictures from the middle of the last century learned how to 'run' by the end of that century, we expect to see more and more static proteins becoming lively actors in the future.'

Indeed, movies of 'morphing' molecules soon became increasingly common with the development of research tools and databases that could readily provide interpolations between two homologous structures [5,6]. Around this time, other individuals and groups began looking towards 3D animation software from the entertainment industry to create detailed visualizations of molecular movement. Some examples of sophisticated movies from the late 1990s to the early 2000s that utilized molecular structures include Drew Berry's animations of the central dogma [7], animations of the bacterial flagellum from Keiichi Namba's group at ERATO [8], and Graham Johnson's animation of kinesin, completed in collaboration

with Ron Vale and Ron Milligan [9]. These visualizations proved to be powerful tools for describing complex and 3D molecular processes to diverse audiences.

Unlike molecular visualization software, like Chimera [6] and Pymol [10], 3D animation applications were not designed to display or analyze molecular structures. Rather, most animation software was created for the game and entertainment industries, and the software capabilities reflect the needs of this target audience [11]. Tools are often separated into numerous categories within the 3D animation application, and typically include modeling tools (for generating a mesh or geometry in the shape of an object or character), animation tools (for the dynamic manipulation of the mesh, such as warping and stretching) and shading tools (for providing a specific texture or pattern to a mesh). To create a molecular animation, the first step often is to import a molecular structure as a mesh. This can be done by exporting geometry from molecular visualization software and importing it into the 3D animation software, or more directly through the use of a plug-in, such as ePMV [12•], Molecular Maya [13•] and BioBlender [14]. At this point, the molecular model is a mesh that can be treated as any other mesh — free to be manipulated and animated, much like a character in an animated film. The 3D animation software provides a sandbox of sorts, allowing the user to freely explore and 'play' with one or more molecular structures in an unrestrained way that few other scientific software tools allow.

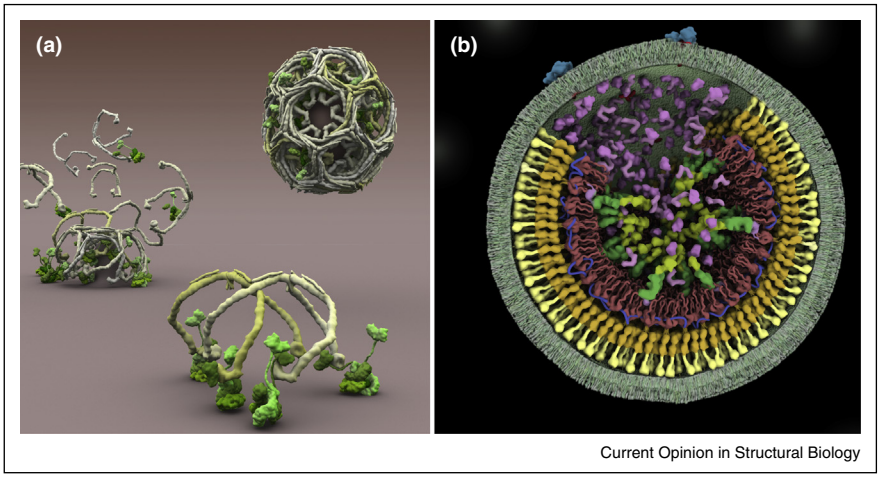
Envisioning molecular assemblies

One of the strengths of 3D animation software is its ability to handle a large number of molecular meshes, which can then be animated forming (or disassembling from) a larger multi-subunit complex or assembly. A number of my own animations fall into this category, including animations of clathrin-mediated endocytosis (Figure 1a, http://biochem.web.utah.edu/iwasa/projects/clathrin.html) and HIV maturation (Figure 1b, http://scienceofHIV.org) [15,16]. For these types of animations, one of the major challenges is building the larger macromolecular assembly (such as the immature HIV virion, or the clathrin-coated endocytic vesicle) out of a large assortment of proteins from many different data sets. The process of building an assembly often involves numerous steps of 'filling in the gaps' and exploring how molecules fit (or unexpectedly do not fit) together.

Complete structures may be known for proteins within the assembly, but for others, only a partial structure may be solved. For partial or unknown structures, a stand-in structure can be made using modeling tools, imported meshes from homologous proteins, or a mix of both. For example, a protein that has two solved domains but misses a long linker region can be made into a single protein model by attaching the two solved portions with a flexible tube of the correct length. Likewise, flexible tails, such as those of tubulin and histones, or other elements that are difficult to crystallize, such as glycans, can be added in a similar manner to create a more complete protein model.

Creating a large molecular assembly from protein components can be a tedious process if done individually by hand, especially considering the thousands of protein components found in assemblies such as a viral particle.

Figure 1



Examples of large, complex molecular assemblies built in 3D animation software. Both images represent still images from a 3D animation. **(a)** Different stages of clathrin-mediated endocytosis. Pale yellow proteins are clathrin, and AP2 is shown in green. Created in collaboration with Tom Kirchhausen, Harvard Medical School. Please view animation online at: http://biochem.web.utah.edu/iwasa/projects/clathrin.html; **(b)** immature HIV particle, where gag protein is shown in yellow, orange and red. Gag-pol protein additionally includes green segments. Purple proteins are non-structural viral proteins derived from a CellPACK model. Please view this model and animations at: http://scienceofHIV.org.

Fortunately, most 3D animation software allow users to write programs that can carry out tasks, such as duplicating a mesh and moving and rotating the mesh within defined parameters. This code could be used to create microtubules from individual tubulin dimers, or to position viral proteins along a membrane surface. Scripts can also be used to import data from outside sources, for example, for importing coordinates of molecular movement from light microscopy based tracking data or from a molecular simulation. These coordinates could then be used to animate a molecular mesh in the animation software.

Creative use of built-in animation tools can also aid in the development of large molecular assemblies. Meshes can be duplicated as 'instances' where a change to one mesh will change all of the duplicates — a particularly useful feature for animating radially symmetric protein complexes such as the opening and closing of pores.

Providing cellular context

Providing biological context for macromolecular interactions is another area that 3D animation software can excel compared to standard scientific visualization tools. Modeling tools allow users to create models of large structures, such as organelles and cells, which can be used as a visual backdrop to provide context to a molecular animation. Animation tools can also be used to provide a magnification effect to provide greater context. For example, a specific subcompartment of the cell can be magnified starting from a whole cell (or even whole body) view, which can help contextualize where a molecular event occurs.

Many molecular animators seek to provide a vision of the 'cellular mesoscale,' beautifully demonstrated in David Goodsell's watercolor paintings, where protein structures (derived mainly from crystallography structures) are visible within a subcellular context (Figure 2) [17]. Particularly notable in Goodsell's paintings is the crowdedness of the cellular environment. To simulate this type of crowding in a 3D scene, plug-ins such as cellPACK can be used (Figure 3) [18••]. CellPACK utilizes a user-defined 'recipe' to pack molecular meshes in specified ratios into containers, which represent a biological compartment such as a vesicle or organelle, and can also be used to populate membrane surfaces. The resulting model can then be analyzed and animated using standard animation tools.

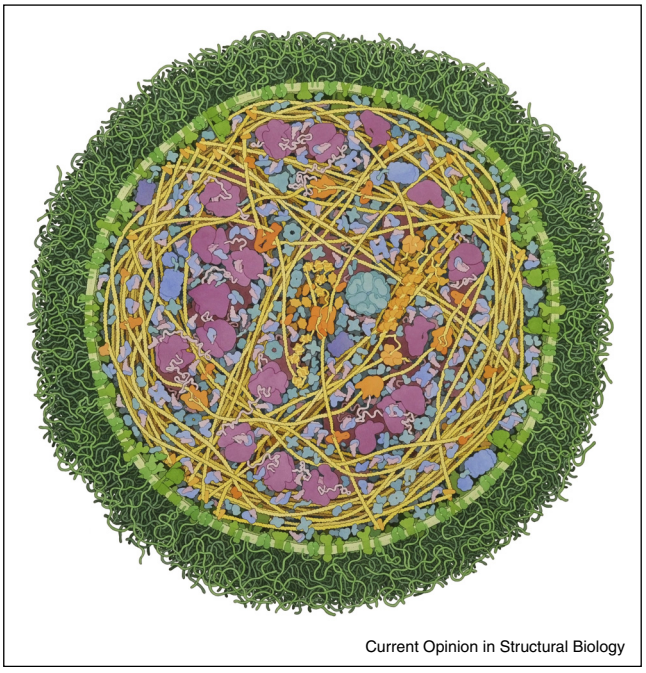
Challenges and future directions

As predicted 20 years ago, molecules are indeed increasingly becoming 'lively actors' in our scientific movies. However, some barriers need to be overcome before we can expect to see animation becoming a more widespread tool.

Because of its steep learning curve, it is unlikely that commercial 3D animation software will enjoy widespread use in the research community. Many software applications require weeks to months of training and regular use to create a basic molecular animation (which itself may take weeks to complete). Although plug-ins simplify the import of molecular meshes into the 3D animation software, the software must often still be mastered in order to create an animation. On the horizon, however, are new animation software packages designed for molecular researchers in mind. Standalone applications, such as Molecular Flipbook [19••] and SketchBio [20], have been designed specifically for creating and animating molecular models. Molecular Flipbook uses a pared-down user interface, an intuitive slide-based animation tool and online video tutorials to allow researchers to create a simple molecular animation within minutes of being introduced to the software. In addition to these new software, existing molecular viewing software, most notably UCSF Chimera, has been steadily adding animation tools into their software suite [21•].

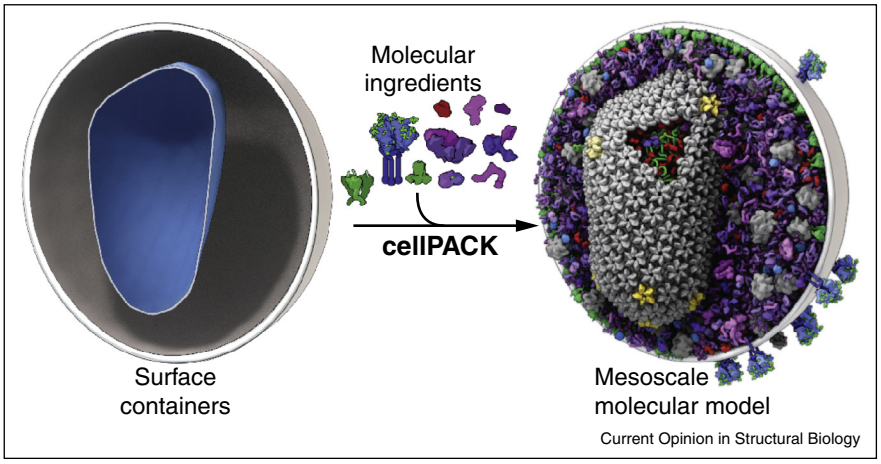
One of the major strengths of molecular animation — its ability to synthesize data from diverse sources, including molecular structures, interactions and dynamics — has also been seen as a weakness [22]. When passively viewing an animation, how can a viewer tell what data sources were used to generate it? Is the visualization showing a consensus model of a process, or a controversial

Figure 2



Watercolor painting of a *Mycoplasma mycoides* cell at mesoscale resolution.
Courtesy of David S. Goodsell, Scripps Research Institute.

Figure 3



CellPACK, a 3D animation software plugin, uses user-defined recipes to create mesoscale molecular models. Figure courtesy of Graham Johnson, UCSF.

finding, or a bit of both? How is an animation vetted, if at all? Providing greater transparency into data sources and artistic decisions, such as David Goodsell provides for a number of his molecular landscape paintings [23•], and perhaps providing a mechanism for reviewing animations will probably allow molecular animations to gain further acceptance and use within the research community.

Acknowledgements

I thank Adam Douglass for critical reading of the manuscript. Support was provided by the NIH (Supplement to P50GM082545) and the NSF (DBI1326378).

References and recommended reading

Papers of particular interest, published within the period of review, have been highlighted as:

- • of special interest
- •• of outstanding interest

1. •• Iwasa JH: **Animating the model figure**. *Trends Cell Biol* 2010, **20**:699-704 http://dx.doi.org/10.1016/j.tcb.2010.08.005 (Epub 2010 Sep 9).
This paper discusses why 3D animation should replace the 2d model figure, and provides examples of how animation tools were successfully used within cell biology research.

2. O'Day DH: **The value of animations in biology teaching: a study of long-term memory retention**. *CBE Life Sci Educ* 2007, **6**: 217-223.

3. McClean P, Johnson C, Rogers R, Daniels L, Reber J, Slator BM, Terpstra J, White A: **Molecular and cellular biology animations: development and impact on student learning**. *Cell Biol Educ* 2005, **4**:169-179.

4. Vonrhein C, Schlauderer GJ, Schulz GE: **Movie of the structural changes during a catalytic cycle of nucleoside monophosphate kinases**. *Structure* 1995, **3**:483-490.

5. Krebs WG, Gerstein M: **The morph server: a standardized system for analyzing and visualizing macromolecular motions in a database framework**. *Nucleic Acids Res* 2000, **28**:1665-1675.

6. Pettersen EF, Goddard TD, Huang CC, Couch GS, Greenblatt DM, Meng EC, Ferrin TE: **UCSF Chimera — a visualization system for exploratory research and analysis**. *J Comput Chem* 2004, **25**:1605-1612.

7. Berry, D: *DNA Central Dogma*. http://www.wehi.edu.au/wehi-tv/dna-central-dogma-part-1-transcription, http://www.wehi.edu.au/wehi-tv/dna-central-dogma-part-2-translation.

8. Namba K, et al.: *Protonic Nanomachine Project*. http://www.fbs.osaka-u.ac.jp/labs/namba/npn/index.html.

9. Vale RD, Milligan RA: **The way things move: looking under the hood of molecular motor proteins**. *Science* 2000, **288**:88-95 Movie 2.

10. *PyMOL, a molecular visualization system*. http://www.pymol.org.

11. Commonly used 3D animation software applications include Maya (http://www.autodesk.com/products/maya/), Cinema4D (http://www.maxon.net/products/cinema-4d-studio/), and Blender (http://www.blender.org/).

12. • Johnson GT, Autin L, Goodsell DS, Sanner MF, Olson AJ: **ePMV embeds molecular modeling into professional animation software environments**. *Structure* 2011, **19**:293-303.

13. • Molecular Maya Toolkit. http://www.molecularmovies.com/toolkit/
ePMV and Molecular Maya provide a means to directly import PDB data into 3D animation software.

14. Andrei RM, Callieri M, Zini MF, Loni T, Maraziti G, Pan MC, Zoppè M: **Intuitive representation of surface properties of biomolecules using BioBlender**. *BMC Bioinform* 2012, **13**:S16.

15. Iwasa JH, Kirchhausen T: *Clathrin-mediated endocytosis*. http://www.cellimagelibrary.org/images/12257.

16. Iwasa JH: *The Science of HIV*. http://scienceofHIV.org.

17. Goodsell DS: *The Machinery of Life*. edn 2. Göttingen, Germany: Copernicus; 2009.

18. •• Johnson GT, Autin L, Al-Alusi M, Goodsell DS, Sanner MF, Olson AJ: **cellPACK: a virtual mesoscope to model and visualize structural systems biology**. *Nat Methods* 2015, **12**:85-91.
This software plug-in utilizes 3D animation software to make crowded molecular environments based on user-defined 'recipes.'

19. •• Molecular Flipbook. http://molecularflipbook.org
Molecular Flipbook is an intuitive standalone software that allows users to readily create basic 3D animations of molecules.

20. Waldon SM, Thompson PM, Hahn PJ, Taylor RM 2nd: **SketchBio: a scientist's 3D interface for molecular modeling and animation**. *BMC Bioinform* 2014, **15**:334.

21. • **Bio-cinema verité? Editorial**. *Nat Methods* 2012, **12**:1127.
This editorial provides insight into some of the challenges facing the use of 3D animation in research.

22. Bromberg S, Chiu W, Ferrin TE: **Workshop on molecular animation**. *Structure* 2010, **18**:1261-1265.

23. • Goodsell DS: **Illustrating the machinery of life: viruses**. *Biochem Mol Biol Educ* 2012, **40**:291-296.
This manuscript is one of a series by Goodsell that describes how the scientific literature informs his intricate molecular landscapes.