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Teacher Notes

The Data Dilemma© — Modeling Scientific Practice

Introduction

Modeling is an important and integral part of scientific practice. The Data Dilemma© Activity is designed to model the process of incorporating evidence into a working external representation of the data. This activity may be used to facilitate a discussion on how scientific models evolve.

Materials

Five piece tangram set (all of the same color)
One extra data piece (a different color)

Procedure

1. Hand out the first piece of the tangram (the large triangle labeled “A”). Create a scenerio for the students explaining that while investigating a topic in the lab you have developed a “triangle model”.

2. Hand out the second piece of the tangram (the second large triangle labeled “B”). Have the students build upon the first triangle model with this second piece. You will notice that some groups will continue to form a triangle while others may form a square or a parallelogram. Stacking pieces is not allowed.

Class Discussion Questions

A. How many different shapes were created with this second piece of information collected from the lab?

B. How do you suppose scientists decide which model to continue building on as their investigation progresses?

3. Tell the students that because the initial evidence acquired suggested the simple triangle model, you decide to continue developing the triangle model and not the parallelogram or square models. Hand out the next three pieces (C, D and E) and have the students continue to build a more robust triangle model using these pieces. There are at least three solutions to this puzzle. If a group quickly discovers a solution, encourage them to find other solutions.

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Modeling Scientific Practice (continued)

Class Discussion Questions

C. Why do you think your teacher directed you to continue with the triangle model?

D. Why do you think researchers would choose to pursue the simplest model that would explain the data?

4. Introduce the "rogue" data piece (F). This part of the tangram is in a different color. Instruct the students that they now have to incorporate this latest piece of data into a geometric shape with the fewest sides possible. There are at least four solutions. If a group discovers a solution quickly, encourage them to find other solutions.

Class Discussion Questions

E. What shape did your model take once the extra piece was incorporated?

F. How did your model evolve when the new piece of data was uncovered?

G. How does this tangram activity model scientific practice?

H. Why might a scientist find it difficult to let an old model go?

I. Considering that there were multiple solutions to the data dilemma, how might a scientist determine if their explanation is the right one?

Teacher Tips

A. We recommend the students work in groups of two or three for this activity. Collaboration is an important skill to develop in the field of science.

B. Many teachers may opt to use this as an opening day activity to introduce the process of science to their students.

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Extension

The Missing Piece

Teachers may want to pose the question
Can you build another model that would require an additional piece of data while still maintaining the rectangle shape?

It is possible to construct a four sided rectangle model with a missing piece of data in the middle of the model. Other models may be built with a piece of data missing from an outside edge. Teachers may want to discuss the significance of the missing piece. Once a model has been developed, the researcher may find holes in the model that may direct the path of continued research on a given project in an effort to lend credibility to the work that has already been established. This is the crux of scientific practice. As basic researching progresses and engineering provides more sophisticated tools to collect and analyze data, the basic research that follows may provide models which evolve into more robust explanations of observed phenomena.

National Framework

Connections to A Framework for K-12 Science Education
Practices, Crosscutting Concepts, and Core Ideas

Dimension 1: Scientific and Engineering Practices
1. Asking questions and defining problems
2. Developing and using models
4. Analyzing and interpreting data
6. Constructing explanations and designing solutions
7. Engaging in argument from evidence

Dimension 2: Crosscutting Concepts
1. Patterns
4. Systems and system models

Dimension 3: Disciplinary Core Ideas

Life Sciences

LS 1: From molecules to organisms: Structure and processes

Engineering,Technology and the Application of Science

ETS 1: Engineering design

ETS 2: Links among engineering, technology, science, and society

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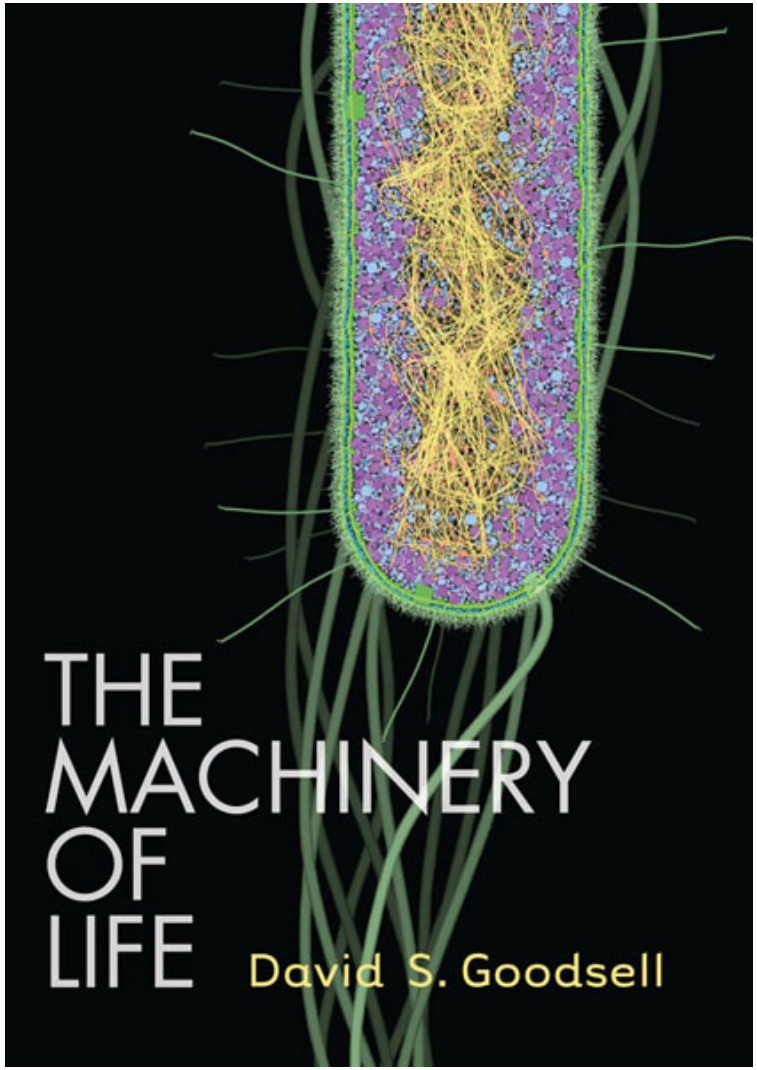
Center for BioMolecular Modeling

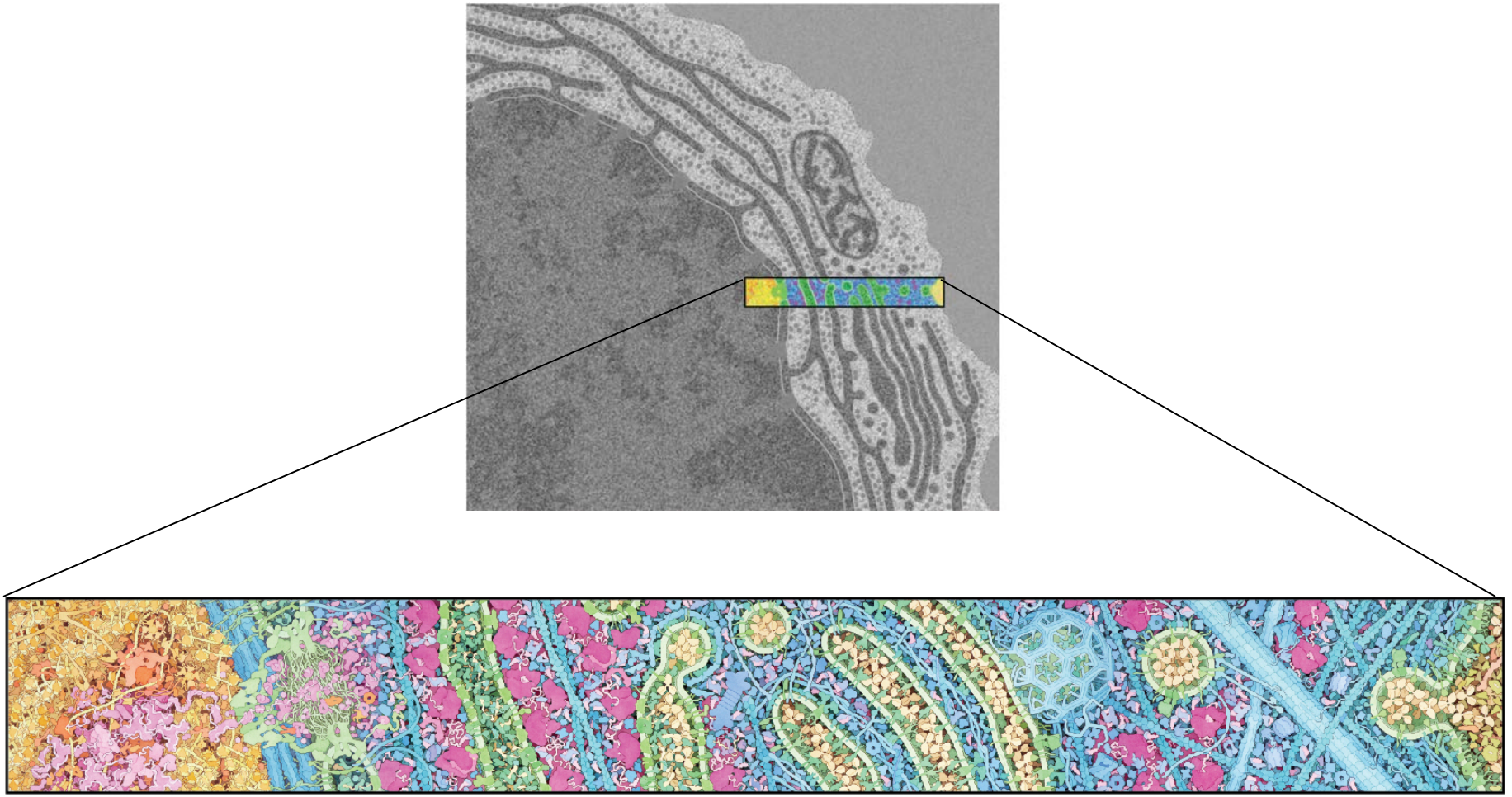
The Cellular Landscapes of David Goodsell



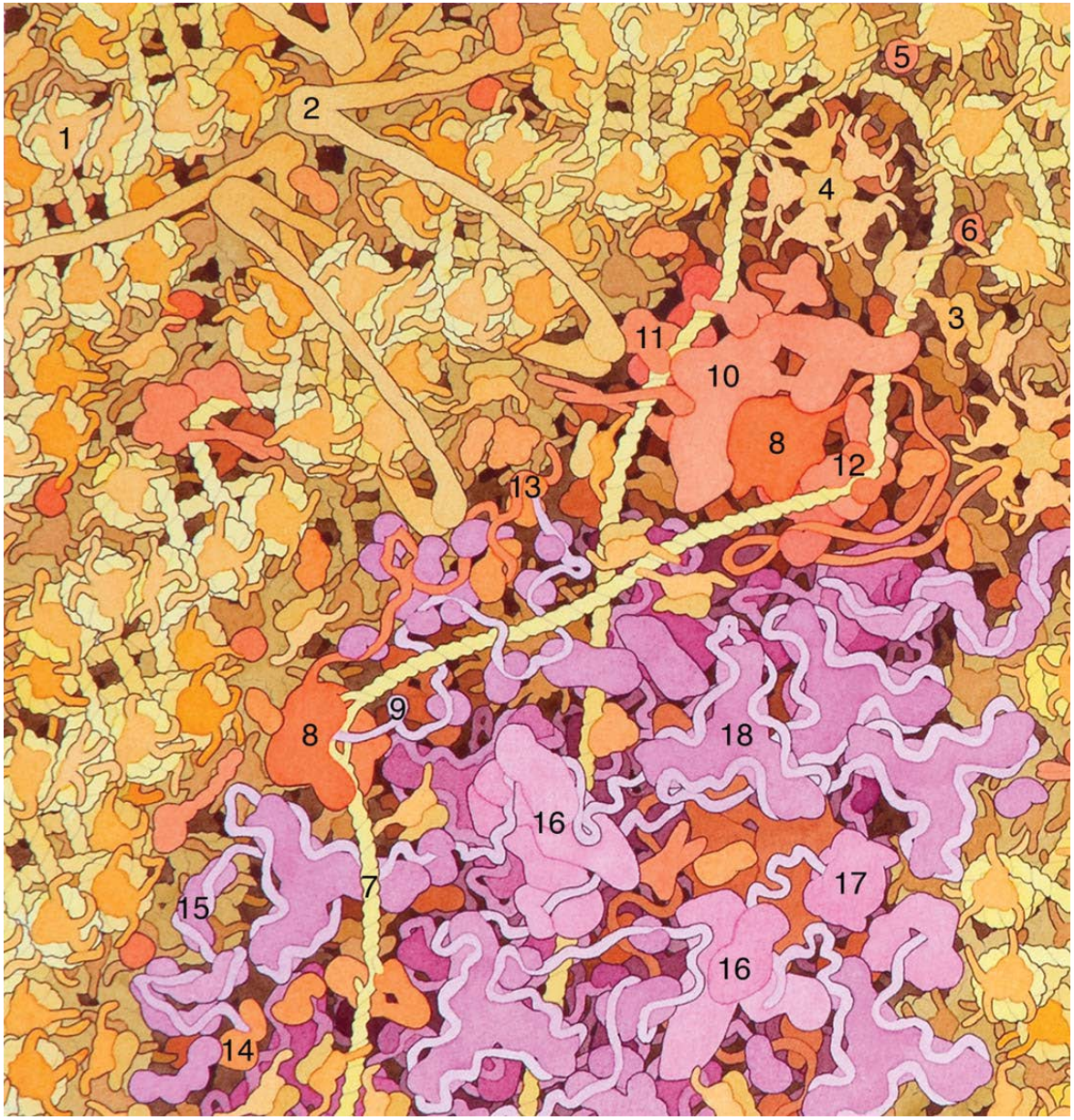
David Goodsell
Scripps Research institute

Scientist…, Author,… and Artist of all things small.

An electron micrograph of an antibody-producing B-cell



Your recent flu shot,….in action.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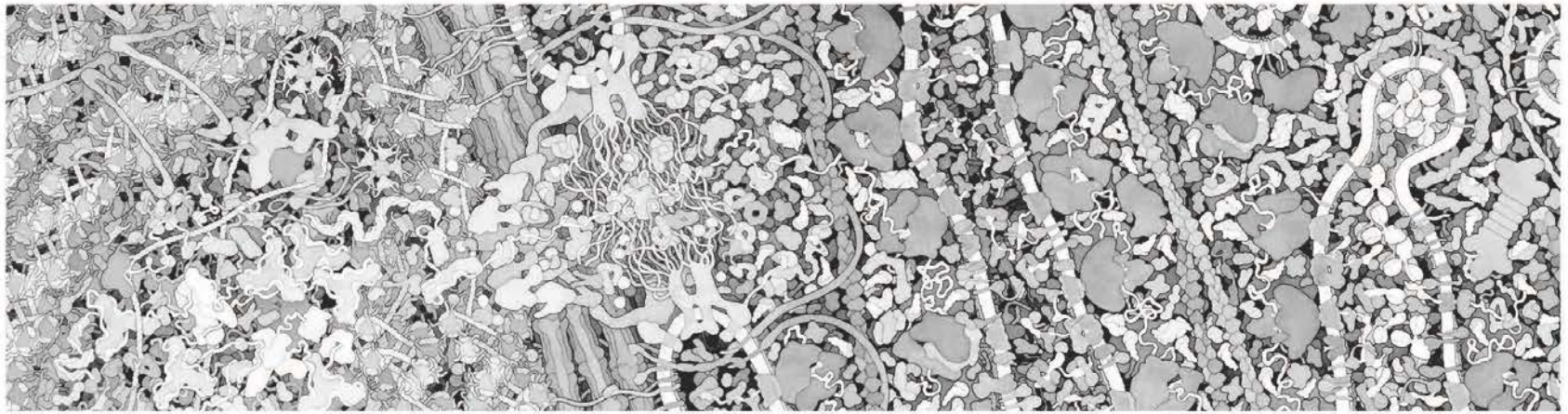
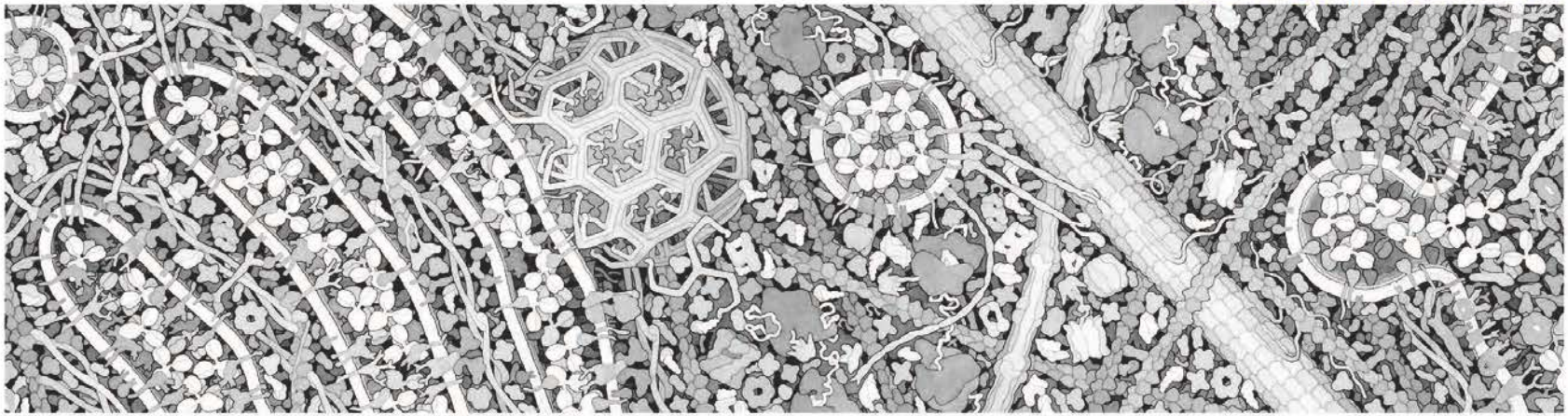
Tour of a Human Cell©



Find/Color/Label – a nucleosome
Find/Color/Label – the antibody gene
Find/Color/Label – an mRNA leaving the nucleus
Find/Color/Label – a ribosome, docked onto the E.R.
Find/Color/Label – a folded antibody protein

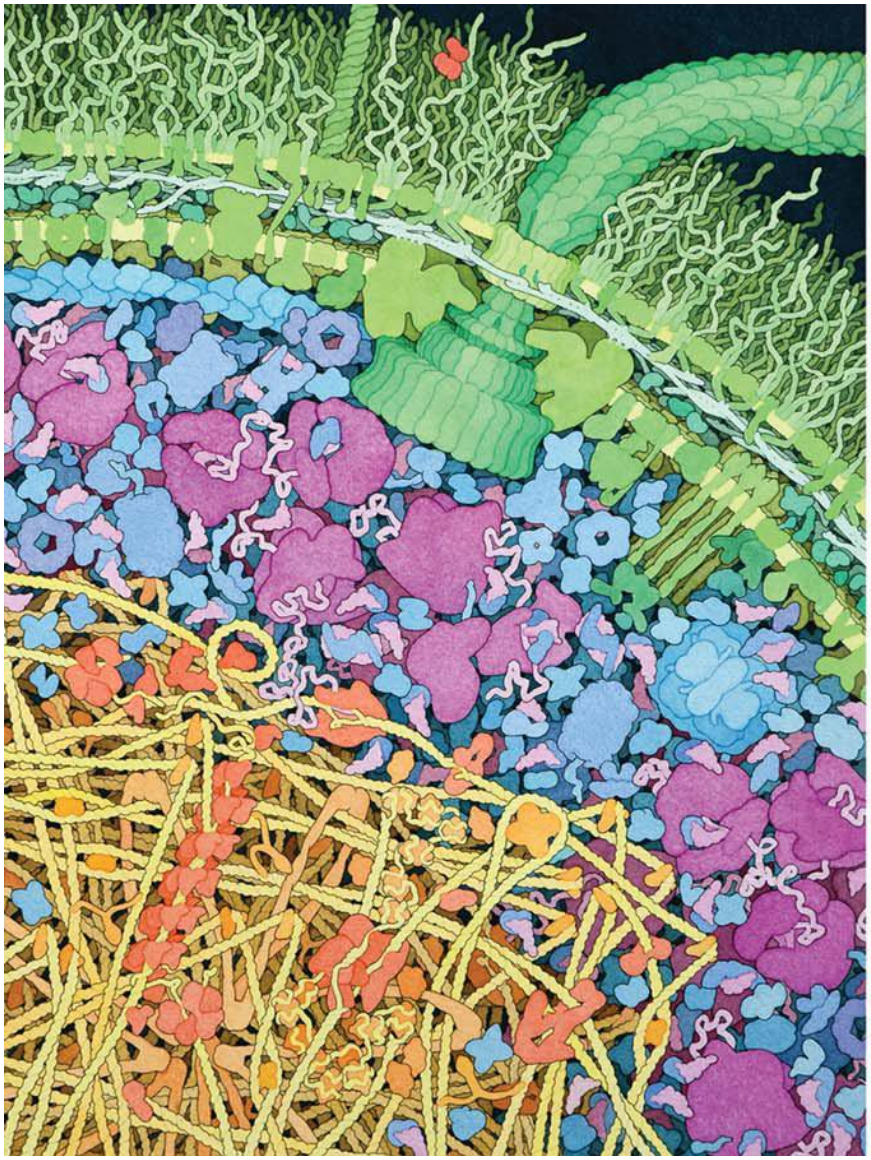
A Molecular Landscape by David Goodsell

3dmoleculardesigns.com/Education-Products/Tour-of-Human-Cell.htm

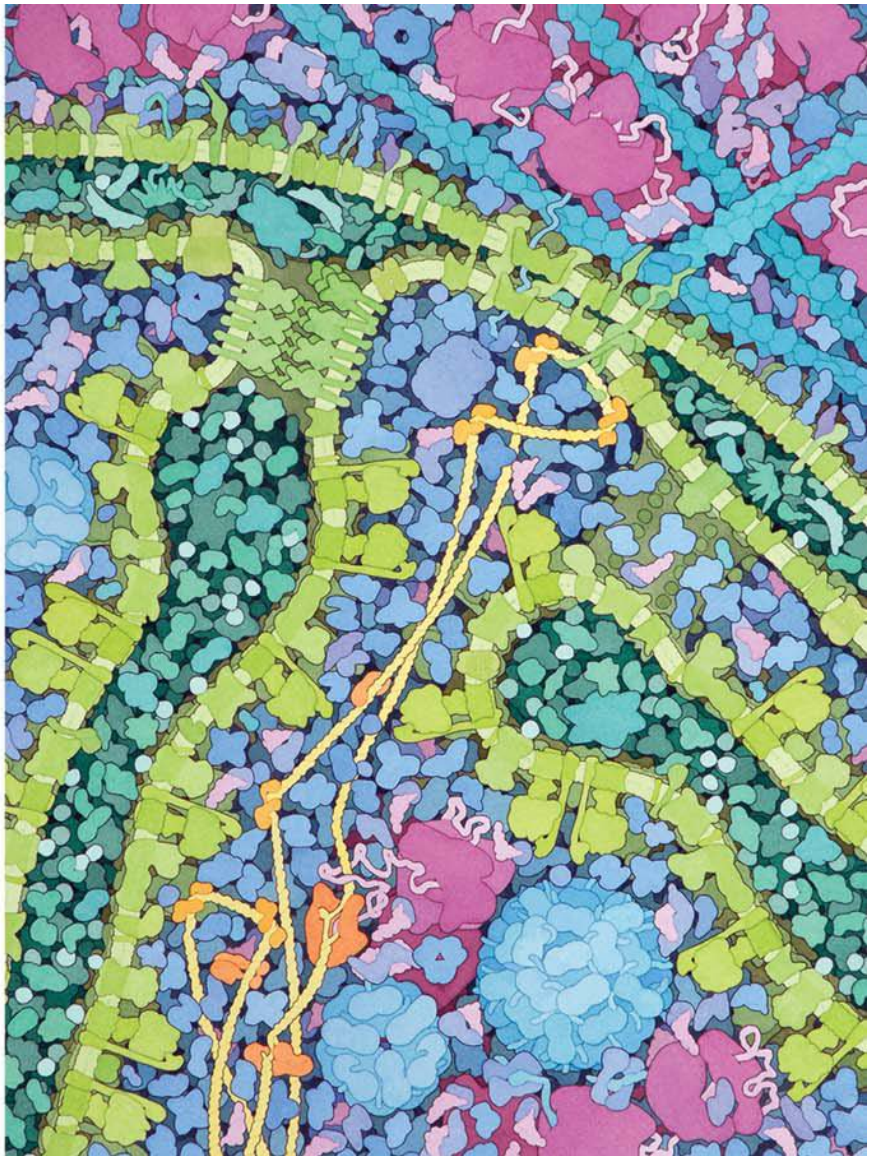


Find/Color/Label – the Golgi Apparatus
Find/Color/Label – a clathrin cage, forming an endosome
Find/Color/Label – kinesin motor proteins
Find/Color/Label – antibodies released into the circulation

E. coli



Mitochondrion



BIOCHEMISTRY AND MOLECULAR BIOLOGY EDUCATION
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Article

Miniseries: Illustrating the Machinery of Life

Eukaryotic Cell Panorama

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Diverse biological data may be used to create illustrations of molecules in their cellular context. This report describes the scientific results that support an illustration of a eukaryotic cell, enlarged by one million times to show the distribution and arrangement of macromolecules. The panoramic cross section includes eight panels that extend from the nucleus to the cell surface, showing the process of protein synthesis and export. Results from biochemistry, electron microscopy, NMR spectroscopy and x-ray crystallography were used to create the image.

Keywords: Eukaryotic cell structure, B-cell, cellular biology, molecular biology, molecular visualization, textbook diagrams, protein synthesis, nucleus, endoplasmic reticulum, Golgi.

INTRODUCTION

As part of the book, "The Machinery of Life," [1], I wanted to include a panoramic image that captures the major features of eukaryotic cells, including their many internal compartments and their extensive infrastructure for managing these compartments. Eukaryotic cells are typically too large to show in their entirety and still show individual molecules: at 1,000,000 X magnification, the entire cell would be several meters wide. So instead, I extracted a long rectangular portion that extends from the nucleus to the cell surface, and presented it on eight successive pages.

I had several goals when designing the image. The first was to include the major compartments of the cell: the nucleus, the endoplasmic reticulum, the Golgi, and the cytoplasm. I presented the mitochondria in a separate image, as they are too large to include in this compressed panoramic view. The mitochondrion image was presented in a previous article [2]. The second goal was to capture the entire process of protein synthesis and export, from DNA to final protein. Finally, I wanted to highlight a few of the unstructured proteins in the cell, and their unusual functions.

I chose a B-cell for the panorama, for several reasons. I wanted a free-living cell, so that there would not be complications with cell junctions. I also wanted a cell that synthesized and exported a familiar and recognizable protein, so that the reader could easily follow it along the process of synthesis.

This article describes the science supporting the illustration. The four letter codes presented here (1i6h, etc.) correspond to structures in the Protein Data Bank and UniProt codes (Q# or P#) are given for amino acid sequences. Full references are not given for these files, as they are available at the RCSB PDB and UniProtKB WWW sites, http://www.pdb.org and http://www.uniprot.org.

Ultrastructure

The overall layout of the panorama was based on electron micrographs of plasma cells, obtained from the electron microscopic atlas of cells, tissues and organs, on the WWW at: http://www.uni-mainz.de/FB/Medizin/Anatomie/workshop/EM/EMPlasmaZ.html.

The amount of cytoplasm separating the nucleus and cell surface is typically larger than what is depicted in the panorama, with many layers of endoplasmic reticulum, mitochondria, and Golgi. Many micrographs, however, include sections where the nucleus approaches fairly closely to the cell membrane, with only a few layers of endoplasmic reticulum, as shown in the panorama. So, the painting presents one extreme.

The panorama is broken into four sections, each with two halves to fit on two facing pages. The first section (Figs. 1 and 2) is the nucleus, with the nuclear interior on the left and the nuclear membrane on the right. The next section shows the endoplasmic reticulum, with one long compartment on the left (Fig. 3), and a budding vesicle on the right (Fig. 4). The third section shows the edge of the Golgi, with a vesicle docking on the left side (Fig. 5)

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 DOI 10.1002/bmb.20494

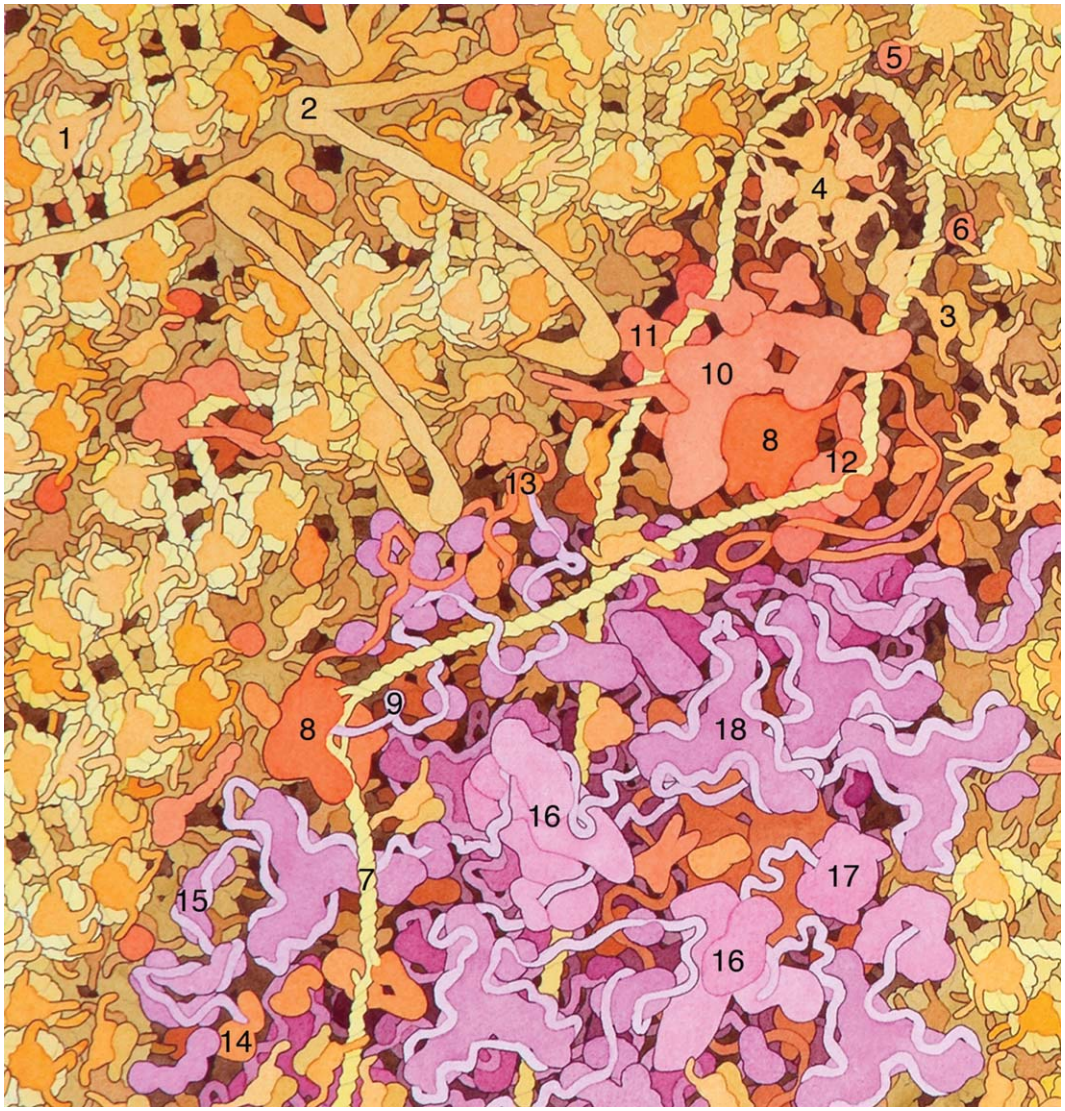

FIG. 1. **Transcription and RNA processing in the nucleus.** (1) nucleosome; (2) SMC; (3) CIA and histone; (4). nucleoplasmin and histones; (5) histone acetyltransferase; (6) histone deacetylase;. (7) DNA; (8) RNA polymerase; (9) messenger RNA; (10) Mediator; (11) enhanceosome; (12) transcription factors; (13) capping enzyme; (14). poly-A polymerase; (15) poly-A-binding protein; (16) spliceosome; (17) exosome; (18) hnRNP-C.

and another vesicle leaving at the right (Fig. 6). The final section shows a vesicle being transported along a microtubule (Fig. 7), and second vesicle fusing with the cell surface at the far right (Fig. 8).

Concentrations

The concentration of macromolecules was by far the most difficult parameter to define for this illustration. A wide range of values are reported, for instance: 200–300 mg/mL for "cytoplasmic protein" [3], 17–26% protein by weight for "actively growing cells" [4], and 15–25% for the "average protein content of animal cells" [5]. I chose a value at the higher end of this scale, roughly 25% protein.

Nucleus

The interior of the nucleus includes DNA in chromatin as well as DNA that is being transcribed. The chromatin is shown at upper left in Fig. 1 and adjacent to the nuclear membrane in Fig. 2. The nucleosomes are based on the crystal structure (1aoi), with the unstructured tails of the histones extended to their actual length. An alternating solenoidal model is used for the chromatin fiber [6]. I have also included a structural maintenance of chromosomes (SMC) protein [7], drawn as a large star-shaped complex as observed in bacterial cells [8]. At the nuclear membrane, several proteins interact with the nucleosomes, as described below.

Nucleosomes and histones are dynamic structures, and there is a substantial infrastructure for coordinating their action. I have included several histone chaperones [9], including the protein CIA (CCG1-interacting factor A) interacting with free histone dimers and the protein nucleoplasmin associating with five histone octamers. In addition, a variety of histone acetyltransferases and deacetylases are shown modifying the tails of histones [10].

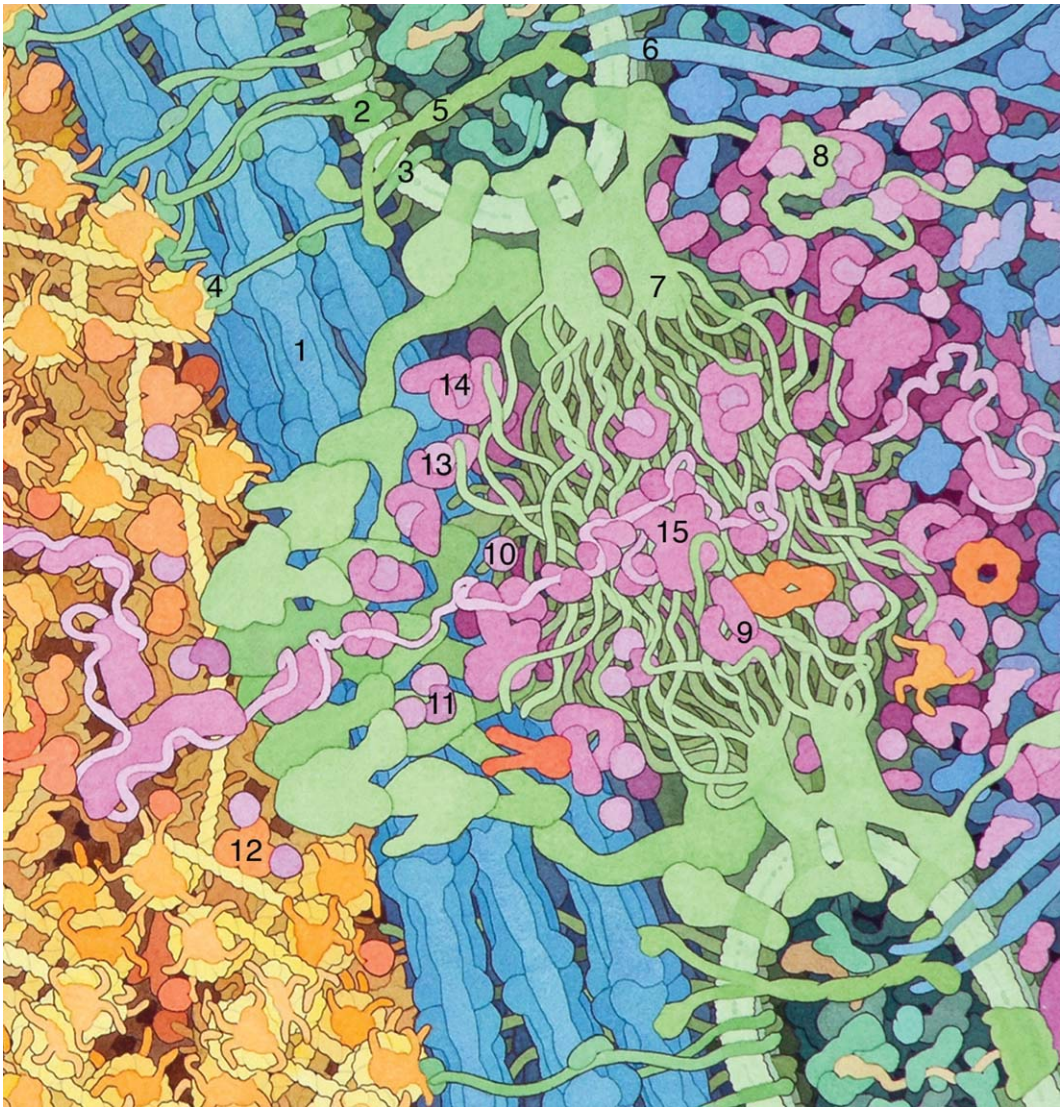

FIG. 2. **Transport through the nuclear pore.** (1) lamin; (2) LBR; (3) emerin; (4) BAF; (5) SUN; (6) nesprin; (7) nuclear pore complex; (8) Nup358 and associated proteins; (9) alpha/beta importin; (10) Ran; (11) NTF2; (12) RCC1; (13) Ran and beta importin; (14) alpha importin and CSE; (15) TAP/p15.

Two RNA polymerases are shown, one at initiation and one actively elongating an RNA transcript. The initiation complex includes a large Mediator complex based on an electron micrograph reconstruction [11], a complex of transcription factors based on crystallographic and electron microscope structures [12], and an enhanceosome taken from a structure-based model [13]. RNA polymerase is based on the crystal structure of the yeast enzyme (1i6h), with a long unstructured C-terminal tail based on the amino acid sequence (P24928). The second RNA polymerase is shown during elongation, with the transcribed RNA looping back and being processed by several capping enzymes [14]. The capping enzyme, along with other enzymes involved in modification, is shown bound to the unstructured tail of RNA polymerase [15].

RNA processing is shown at the bottom of the nuclear region in the image. At the 3′ end of the RNA chain, poly-A polymerase (1f5a) is adding the poly-A tail, which then associates with poly-A-binding protein (1cvj). Two spliceosome complexes are shown, one before splicing and one after splicing, based on structures from electron microscopy [16]. The introns are then degraded by exosomes (2nn6). The RNA is shown associating with hnRNP-C (heterogeneous nuclear ribonucleoprotein C), with the three-armed structure taken from electron micrograph structures [17]. These are shown dissociating as an RNA strand is transported through the nuclear pore.

Nuclear Membrane

The nuclear membrane, shown in Fig. 2, is composed of two lipid bilayers [18]. On the nuclear side, the membrane associates with the nuclear lamina, on the cytoplasmic side, it is continuous with the endoplasmic reticulum. The entire membrane is pierced by nuclear pores, which mediate transport into and out of the nucleus.

The nuclear lamina is composed of layers of lamin, a protein similar to cytoplasmic intermediate filaments. I have based these on electron micrograph structures of isolated intermediate filaments (described in more detail

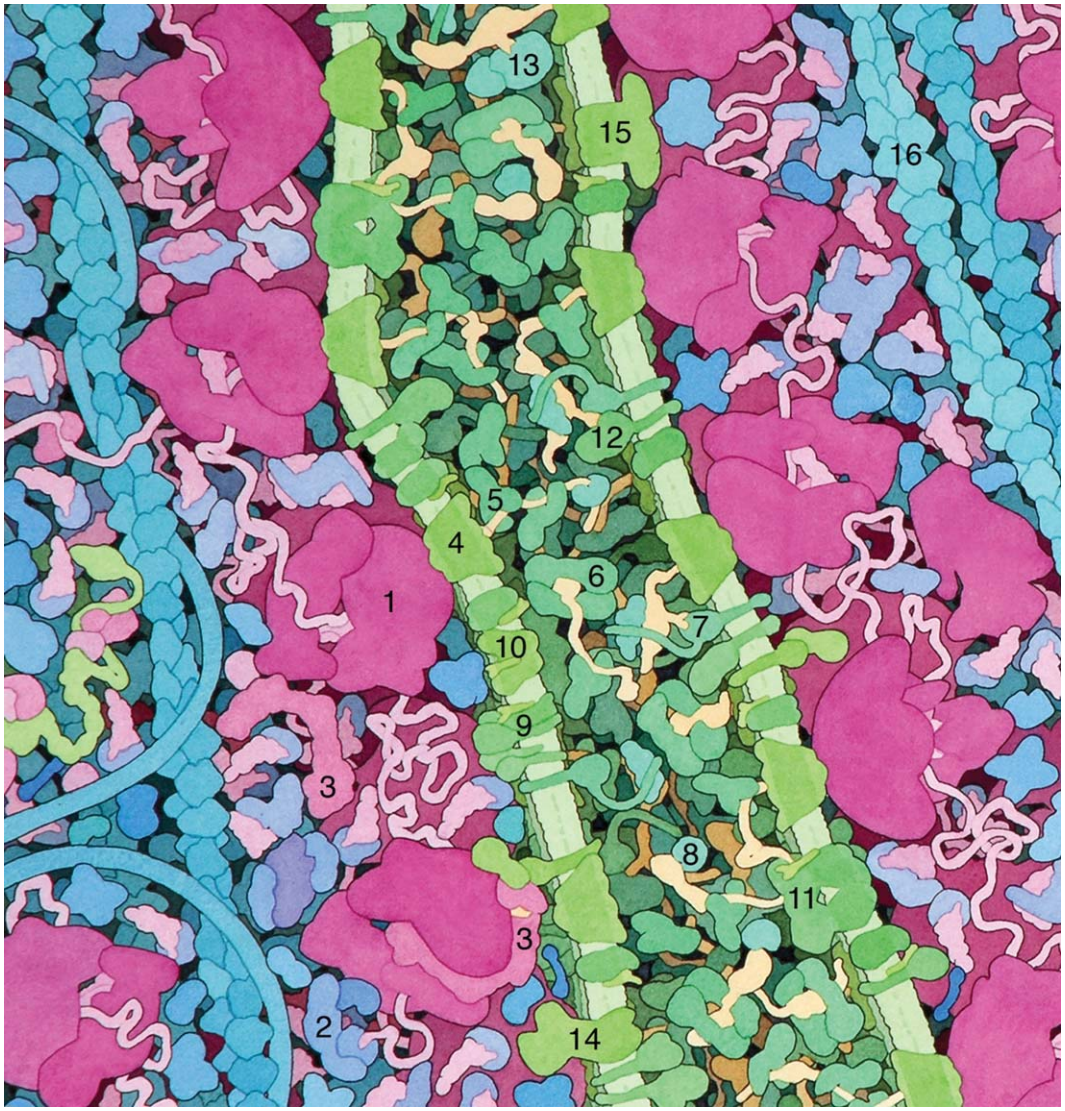

FIG. 3. **Endoplasmic reticulum.** (1) ribosome; (2) initiation factors; (3) signal recognition particle; (4) Sec61/SecY channel; (5) BiP; (6) GRP94; (7) calnexin and Erp57; (8) cyclophilin; (9) glycotransferases; (10) flippase RFT1; (11) oligosaccharide transferase; (12) GlsI; (13) GlsII; (14) calcium pump; (15) IP3R; (16) actin.

below) and electron micrographs of the lamina [19]. The lamina is connected to the membrane through a variety of membrane-bound proteins, including lamin-B receptor (LBR) and emerin. I based these structures on the amino acid sequences (Q14739 and P50402) and schematic diagrams [20]. They are drawn as unstructured proteins, with small proteins such as BAF (barrier-to-autointegration factor, 2odg) associating with the lamin and with nucleosomes.

A SUN protein (Sad1 and UNC84 domain containing protein) is shown bridging the two membranes and interacting with nesprin outside [21]. Nesprin is a long structural protein that interacts with cytoskeletal proteins [22]. I have included the chaperones and other proteins of the endoplasmic reticulum between the two membranes—these are described in more detail below.

The nuclear pore was the most exciting subject that I researched and rendered for this image. Current models of the pore include a large collection of unstructured nucleoporin proteins extending into the lumen of the pore, mediating the flow of proteins in and out [23–25]. The overall shape of the pore is based on electron micrograph reconstructions [26]. The Nup358 nucleoporin extending on the cytoplasmic side, and its interaction with transport proteins ras-related nuclear protein (Ran), Ran GTPase-activating protein (RanGAP), small ubiquitin-related modifier (SUMO), and ubiquitin-conjugating enzyme (UBE2I), is based on the annotation in UniProtKB entry P49792.

The entire transport cycle for nuclear proteins is shown [27]. This includes alpha and beta importin [28] transporting a cargo of a topoisomerase, a DNA clamp, and a repressor. Also, two copies of the Ran protein are being transported inwards by NTF2 (nuclear transport factor). Once inside, the GDP in Ran is exchanged for GTP by RCC1 (regulator of chromosome condensation, 1a12), which is shown bound to DNA [29]. Ran then associates with beta importin, and with a complex of alpha importin and CSE (importin alpha re-exporter, or chromosome segregation protein), carrying them back out through the nuclear pore. The export of RNA is also shown, with the

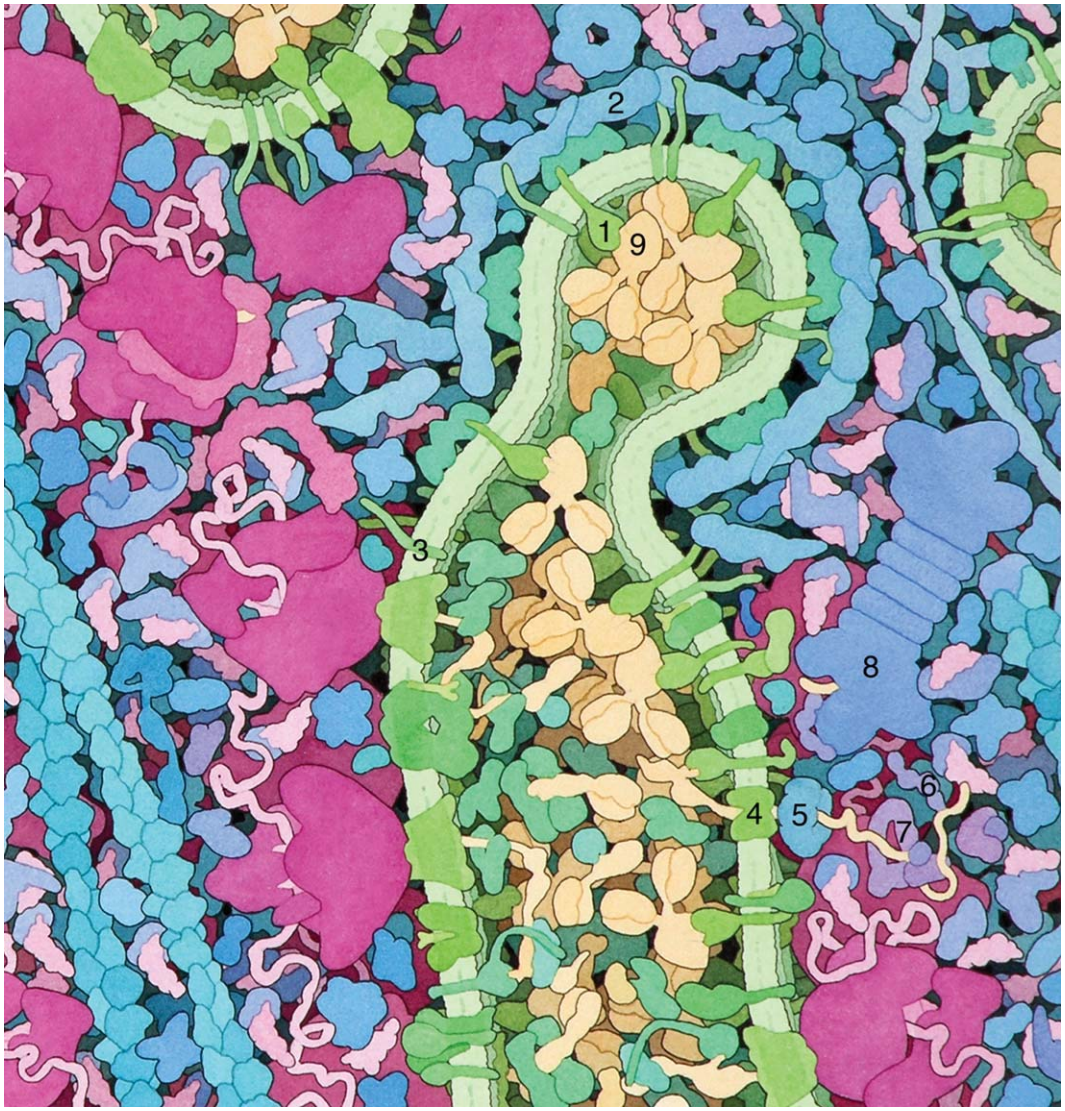

FIG. 4. **Transport from the endoplasmic reticulum.** (1) cargo receptor protein; (2) COPII; (3) SNAP; (4) EDEM; (5) AAA+ protein; (6) ubiquitin; (7) ubiquitin-conjugating proteins; (8) proteasome; (9) antibody.

complex of TAP (nuclear RNA export factor, or Tip-associated protein) and p15 proteins guiding the RNA outwards [27].

Endoplasmic Reticulum (ER)

Once the RNA leaves the nucleus, it is picked up by ribosomes and used to direct the synthesis of proteins, as shown in Fig. 3. At lower left, a ribosome has started synthesis, initiated by the complex of initiation factors [30]. A signal recognition particle (1ry1) has bound to the complex, recognizing the short signal sequence that is translated first. Interaction of the signal recognition particle with the ribosome was taken from a review [31], and eukaryotic ribosome structures were taken from electron micrograph reconstructions [32]. Binding of the signal recognition particle to its receptor delivers the ribosome to Sec61/SecY, the channel through the ER membrane, and the protein BiP (ER luminal binding protein) acts as a ratchet binding to the nascent protein inside the ER and ensuring that it stays inside [33, 34].

A variety of chaperonin proteins inside the ER assist with folding, including GRP 94 (glucose-regulated protein 94, modeled after HSP 90, 2cg9), calnexin (1jhn) [35] and protein disulfide isomerase ERp57 (2h8l), and cyclophilin (2cpl). Studies have shown that immunoglobulins are largely folded in the ER, not proceeding to the Golgi until they are in their proper oligomeric state [36, 37].

The ER also glycosylates proteins [38]. Ig-G molecules contain one N-linked site for glycosylation in the constant domain, and this glycosylation is important for structural stability, for interaction of antibodies with other serum proteins, and for transport and secretion of antibodies from the plasma cell [39]. The oligosaccharides are built on a lipid anchor by a collection of glycotransferases [40], shown here based on amino acid sequences of the yeast enzymes, taken from UniProtKB. Strangely, this process starts with the growing oligosaccharide chain facing outwards, and then the flippase RFT1 (based on the primary sequence from the yeast protein, UniProtKB P38206, which shows 13 transmembrane helices) flips the lipid toward the inside. Once the oligosaccharide is

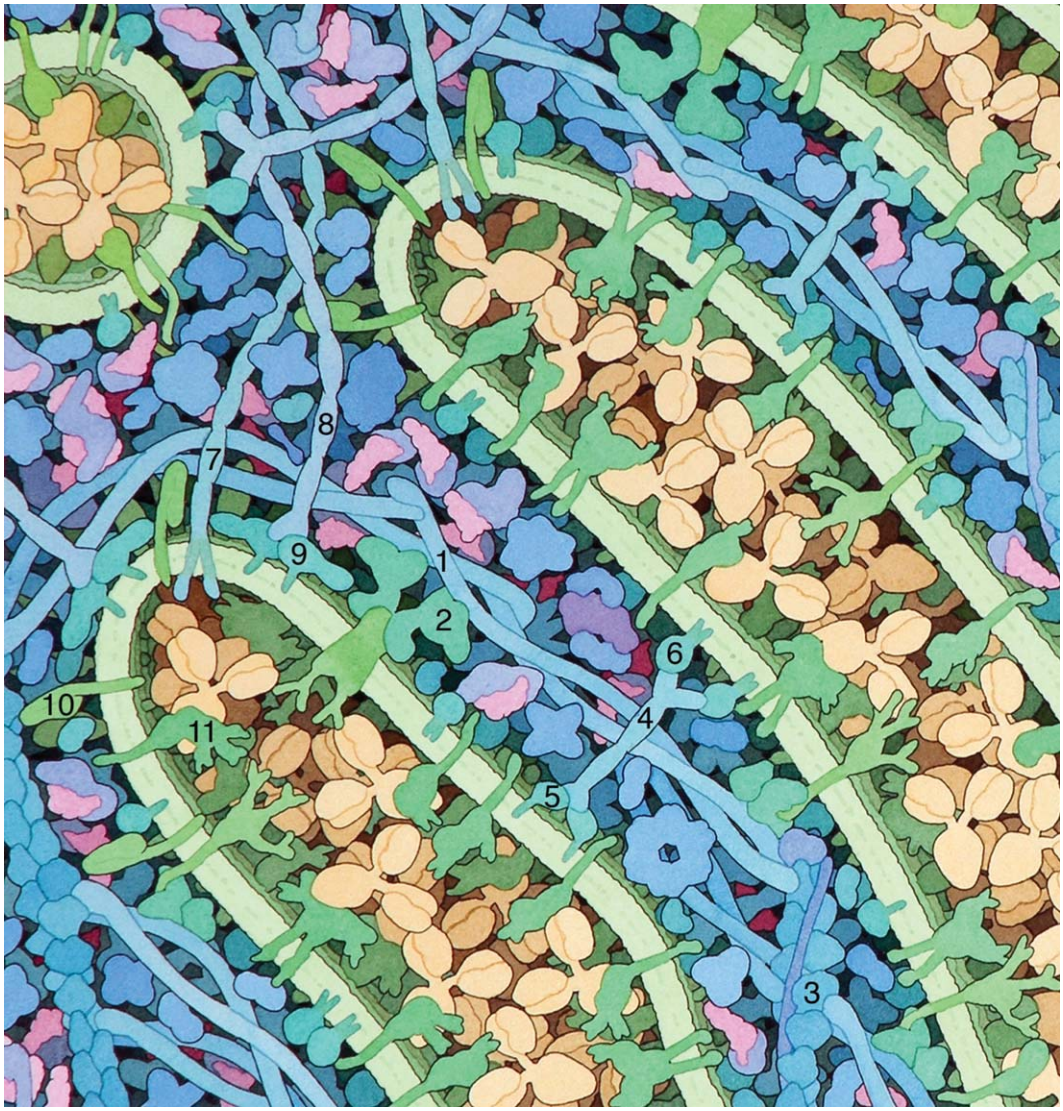

FIG. 5. **Protein sorting in the golgi.** (1) spectrin; (2) ankyrin; (3) actin; (4) golgin45; (5) GRASP55; (6) RAB2; (7) gigantin; (8) GM130; (9) GRASP65; (10) SNARE; (11) glycosyltransferase.

built, it is transferred from the lipid to the protein by oligosaccharide transferase, a large protein complex based on electron micrograph reconstructions [41]. Finally, several glucosidases, such as GlsI and GlsII [42] trim the oligosaccharides.

The ER is the major storage space for calcium ions in most cells, which is used for signaling [43]. I have included two proteins in ER membrane that are important in this process, a calcium pump (1su4), which transports calcium into the ER, and IP_3R (inositol 1,4,5-triphosphate receptor/calcium channel) [44], which releases calcium for signaling.

I found very little information on the infrastructure that is used to maintain the overall ultrastructure of the ER. There is evidence that microtubules are involved in the generation of the structure of the ER in human cells [45, 46], although actin plays this role in plant and yeast cells. However, there is also evidence that microtubules are not needed for maintaining the structure of the ER [46]. I chose to gloss over this point by simply including a few generic actin filaments in the image, without showing any specific interaction between the cytoskeleton and the ER membranes.

Figure 4 shows a vesicle being removed from the ER. Cargo receptor proteins, modeled here after ERGIC-53 (ER-Golgi intermediate compartment protein, 1r1z) [47], have captured antibodies and are interacting with COPII proteins (vesicle coat proteins) that are forming the vesicle, based on atomic structures (1m2v, 2pm7) and electron micrograph reconstructions [48]. Several SNAP proteins (synaptosomal-associated protein) are also included, which will be important for the fusion of the vesicle at its next stop, the Golgi. Finally, at lower right, I have included EDEM (ER degradation-enhancing alpha-mannosidase-like protein), a protein that recognizes faulty proteins from the ER [38]. I have shown a hypothetical transport protein powered by an AAA+ protein ejecting this protein from the ER, where it is ubiquinated and ultimately destroyed by a proteasome. Ubiquitin and ubiquitin-conjugating enzymes were based on crystal structures (1r4n, 1fxt, 1ldk, 1fqv), and the proteasome is based on an electron micrograph reconstruction [49].

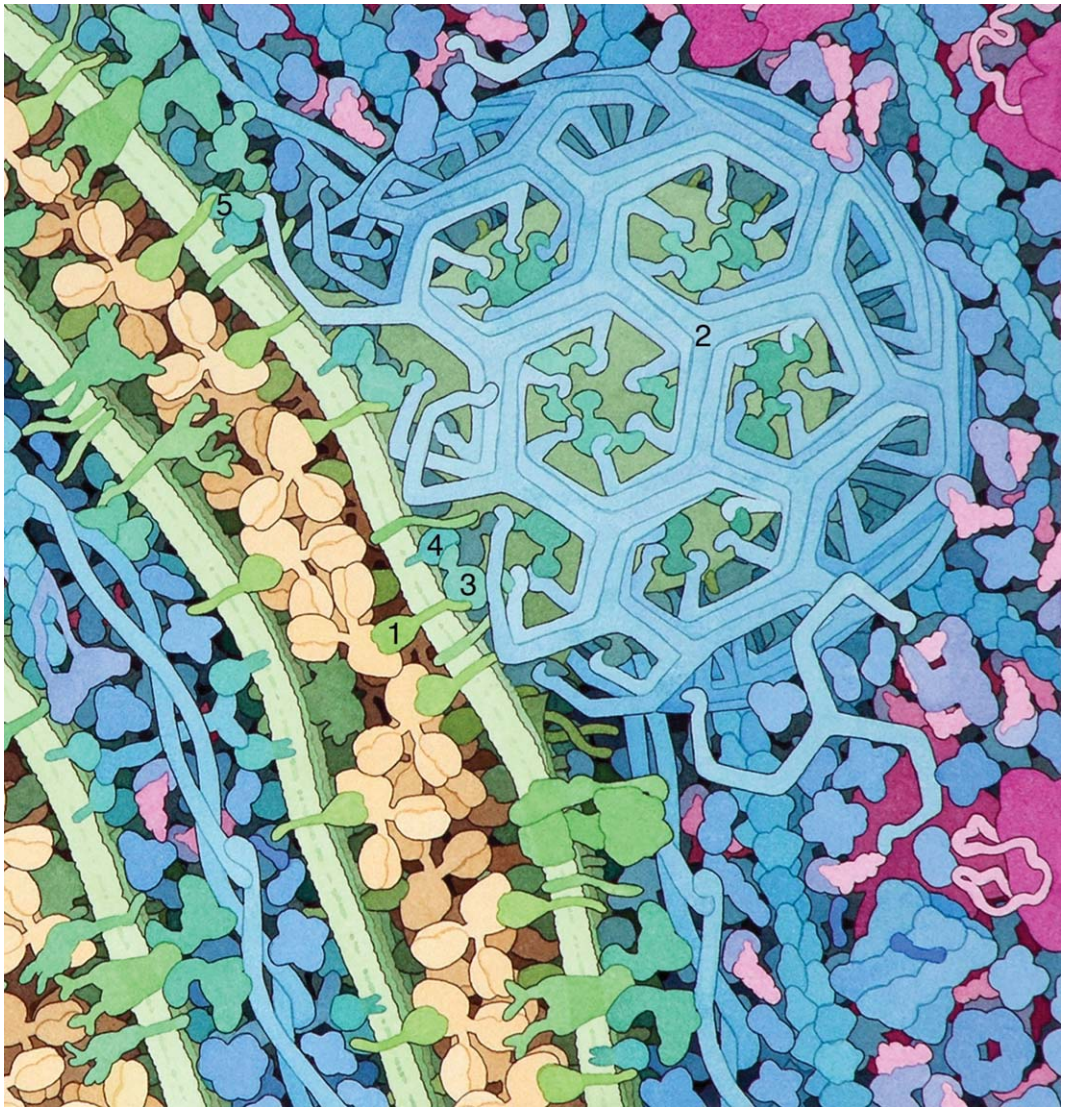

FIG. 6. **Transport from the golgi.** (1) cargo receptor protein; (2) clathrin; (3) AP2; (4) ARF; (5) GGA.

Golgi

The Golgi, shown in Figs 5 and 6, is the site of fine-tuning and sorting of proteins. I have included three layers of the Golgi, ranging from cis to trans as you move from left to right through the panorama. I have included many infrastructure proteins, based on biochemical studies and schematic diagrams. These include spectrin, bound to the Golgi membrane through ankyrin and linked through short filaments of actin [50]. The spectrin and ankyrin structures are based on a combination of primary sequence, electron microscopy and crystal structures of domains [51]. I have included a speculative complex of golgin45 protein linking between the Golgi stack, associating with GRASP55 (Golgi reassembly-stacking protein) and RAB2 (Ras-related protein) in the membrane surface [52]. Huge proteins like gigantin and GM130 (cis-Golgi matrix protein, bound to GRASP65) extend from the Golgi, acting as tethers to trap vesicles [53–56], bringing them close enough for membrane fusion by SNARE proteins [57]. Many of these proteins are characterized by long helical bundles, forming a flexible ropelike structure.

Inside the Golgi, a variety of glycosyltransferases modify the oligosaccharides on the antibodies [58]. I have drawn them as bound to the Golgi membrane by a transmembrane segment [59]. In the trans compartment, cargo receptor proteins capture antibodies and prepare them for transport to the surface. I have shown a clathrin-based mechanism for creation of vesicles, based on schematic diagrams from a review article [60], although other mechanisms may be important [61]. The clathrin coat is composed of three-armed triskelions based on the crystal structure (1xi4). The adaptor protein AP2 (1gq5, 1ky7, 2g30) links the clathrin coat to the cytoplasmic portion of the receptor, with the help of the regulatory protein ARF (ADP-ribosylation factor, 1rrf). GGA (Golgi-localized gamma-ear-containing ARF-binding protein) is also shown in the process of recruiting the receptor, AP2 (adapter protein) and ARF to the clathrin coat. The clathrin coat then disassembles after formation of

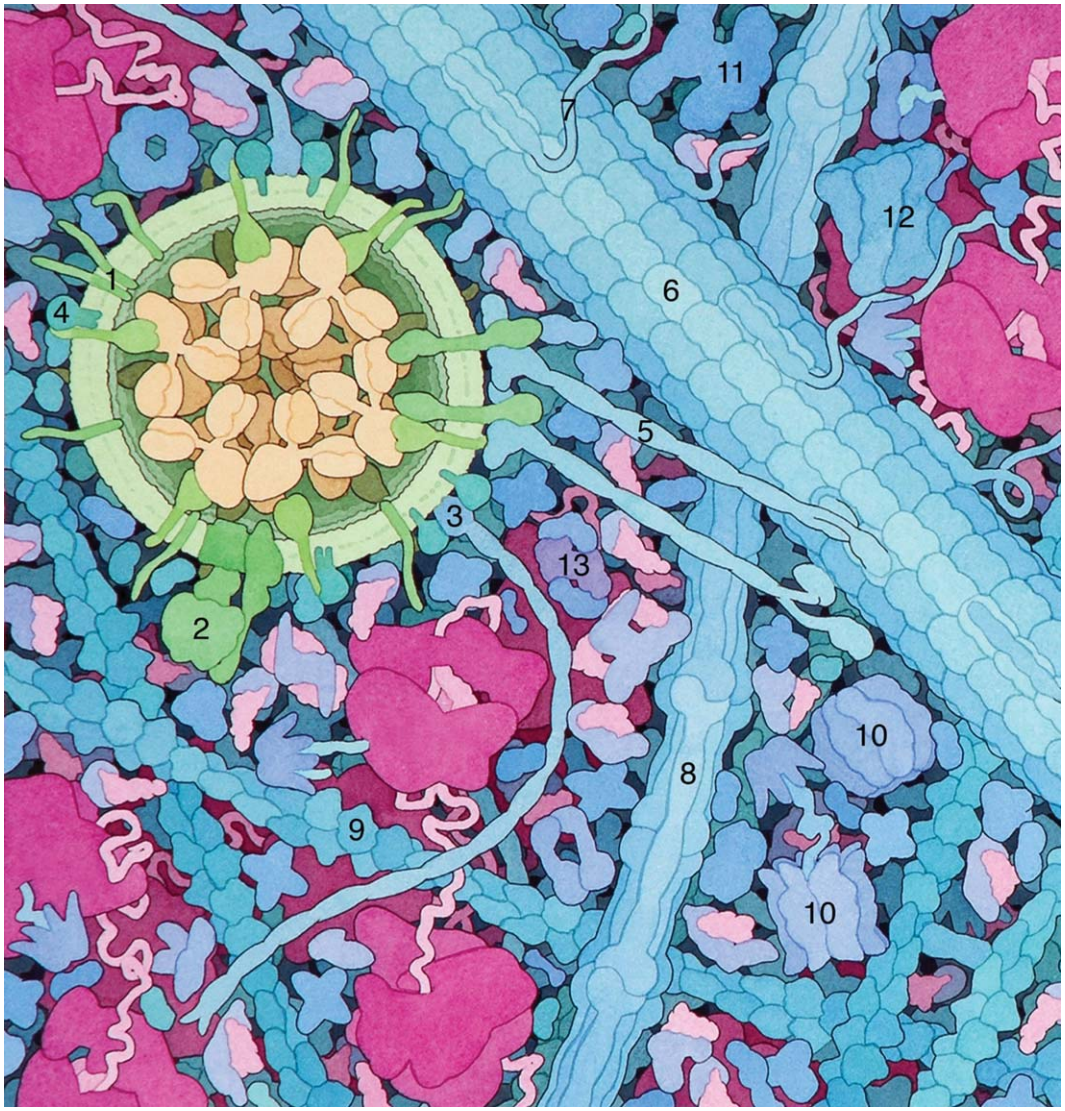

FIG. 7. **Transport of a vesicle through the cytoplasm.** (1) SNARE; (2) vacuolar ATPase; (3) golgin; (4) Rab; (5) kinesin; (6) microtubule; (7) MAP; (8) intermediate filament; (9) actin; (10) TRiC/CCT chaperonin; (11) fatty acid synthase; (12) calcium/calmodulin-dependent protein kinase II; (13) caspase 7 and XIAP.

the vesicle, but there was not room in the panorama to show this process.

Cytoplasm and Cell Surface

Figure 7 shows the journey of a vesicle to the cell surface. The vesicle includes SNARE proteins [62], a vacuolar ATPase (based on ATP synthase, 1c17 and 1e79), and several long golgin tether proteins connected to the vesicle through ARL (ADP-ribosylation factor-like protein) [54]. Small regulatory Rab proteins are bound to the vesicle surface with lipid anchors (3rab). Two kinesin proteins are transporting the vesicle along a microtubule (3kin) [63].

Three types of filaments from the cytoskeleton are shown. The microtubule is based on the crystal structure of a tubulin dimer fit to a cryo-EM map of the intact filament [64], shown here associated with unstructured MAP proteins, also based on electron microscopy [65]. The intermediate filament is based on electron micrographs and structural models of the coiled-coil regions [66], and the actin filaments are based on the crystal structure of the subunit and electron micrographs of the filament [67]. Several Arp2/3 (actin-related protein) junctions are shown in the actin network [68], which connect actin filaments with a characteristic 70 degree angle. The overall arrangement of filaments is based roughly on results from electron microscopy [69].

Many enzymes are shown in the cytoplasm. These include familiar enzymes of protein synthesis, glycolysis and other metabolic tasks, described in more detail in the previous article on the *Escherichia coli* illustration [70]. A few of the most showy examples include the TRiC/CCT chaperonin (TCP1-ring complex or chaperonin containing TCP1) in its open and closed forms (3iyg) [71], fatty acid synthetase (2cf2), and calcium/calmodulin-dependent protein kinase II [72] with calmodulin (3cln). Also included are caspase 7 bound to XIAP (X-linked inhibitor

FIG. 8. **Export of proteins across the cell membrane.** (1) Arp2/3; (2) c-Abl; (3) gelsolin; (4) spectrin; (5) ankyrin; (6) Fas receptor; (7) magnesium transporter; (8) immunoglobulin M; (9) interleukin receptor; (10) Src tyrosine kinase; (11) exocyst; (12) Rab; (13) Rho; (14) SNARE complex; (15) NSF.

of apoptosis, 1i4o), oncogene c-Abl tyrosine kinase (1opk), and gelsolin [73].

The cell membrane, shown in Fig. 8, is braced on the inside by a spectrin network, attached to the membrane through ankyrin. Cell surface proteins include Fas receptor (P25445), a magnesium transporter (based on bacterial MteE, 2yvy) and a membrane-bound form of immunoglobulin M (1igt). The large signaling complex is an interleukin receptor (3bpl) and Src tyrosine kinase (2src). Interleukins and similar signals are important for the survival of plasma cells [74].

A vesicle is shown midway through the process of fusion. The exocyst complex, bound to the membranes through Rab and Rho, acts as a tether to bring the vesicle to the membrane. The structure is based on electron microscopy [75]. The SNARE complexes shown on either side of the vesicle power the fusion of membranes [62]. The NSF (N-ethylmaleimide-sensitive factor) protein is shown separating a SNARE complex after membrane fusion (1d2n).

Aesthetics and Pedagogy

The design of this illustration was highly constrained, both by the need to be consistent with other illustrations in the book [1] and previous articles in BAMBED [2, 70, 76], and by the size of the book. The colors, magnification, and style are all similar to the other illustrations, so readers can compare the structure of the eukaryotic cell with the structure of a simpler bacterial cell. The consistent scheme, for instance, highlights the similar function of the bacterial nucleoid and the eukaryotic nucleus.

Each illustration is presented at 1,000,000× magnification in the book, and slightly reduced here. A simplified shape is shown for each macromolecule, since atoms are too small to be visible at this magnification. Molecules in the nucleus are colored yellow and orange and proteins in the cytoplasm are colored blue. Molecules with RNA, and many of the enzymes that process and transport RNA, are colored magenta. Membranes are colored green to highlight the different compartments in the cell, and mole-

cules inside the ER and Golgi are also colored green. The antibodies being produced are colored tan to set them apart from the cellular macromolecules.

The layout of the illustration is designed to tell the story of protein synthesis and export in a series of page-sized panels, while still connecting into a continuous panoramic cross-section. Each pair of panels presents a particular compartment on the left-hand page, and a mechanism of transport from the compartment on the right-hand page. In this way, readers are drawn through the panorama as they follow the transcription, translation, processing and export of antibodies.

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Miniseries: Illustrating the Machinery of Life

Mitochondrion*

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Diverse biological data may be used to create illustrations of molecules in their cellular context. I describe the scientific results that support a recent textbook illustration of a mitochondrion. The image magnifies a portion of the mitochondrion by one million times, showing the location and form of membranes and individual macromolecules, revealing the molecular basis of its role in energy metabolism and apoptosis. Results from biochemistry, electron microscopy, and X-ray crystallography were used to create the image.

Keywords: mitochondria, cellular biology, molecular biology, molecular visualization, textbook diagrams, energy metabolism, apoptosis, programmed cell death, endosymbiont hypothesis.

Mitochondria are organelles with many intriguing aspects [1]. They play a familiar role in energy metabolism, housing the machinery of ATP synthesis and harnessing chemical, electrical, chemiosmotic, and mechanical energy transformations in the process. They also play a major role in apoptosis (programmed cell death), amplifying the signal that ultimately leads to the death of the cell. In addition, mitochondria live their lives as more-or-less autonomous symbiotic units that live and reproduce inside our cells, which has been taken as evidence that mitochondria evolved from endosymbiotic cells that took up residence in eukaryotic cells early in the evolution of life [2, 3]. For the new edition of "The Machinery of Life" [4], I wanted to create an illustration that captured these many aspects of mitochondrial structure and function. Figures 1 and 2 show the illustrations used in the book, and Fig. 3 is a key to Fig. 2. I present here the scientific support for the molecular and ultrastructural details of the illustration, as well as some of the aesthetic and pedagogic choices that I made when designing the illustration.

MORPHOLOGY

The traditional model of mitochondria, inferred primarily from electron micrographs of thin sections, has a smooth outer membrane and a folded inner membrane, folded either into plate-like invaginations or tubules. The numerous folds of the inner membrane create a large surface area that is filled with the membrane-bound molecules of electron transport and ATP synthesis. However, more recent models based on electron tomography of mitochondria have shown that the traditional model may be too simplistic [5]. These studies suggest that the inner membrane folds to form two compartments that do not share soluble proteins. A portion of this membrane lies immediately inside the outer membrane, defining the intermembrane space between them. Another portion of the inner membrane surrounds the cristae, forming the intercristal space. The two compartments are connected by small (or no) openings that connect them, and the two compartments have a different complement of proteins.

I have drawn a speculative model that has the two spaces connected by narrow connections that are constricted by the protein OPA1 [6]. This protein is similar to dynamin, which forms helical assemblies that are important for pinching off membranes during the process of budding. I based the structure on a combined crystallographic/EM structure of dynamin [7]. There is also evidence for regions where the outer and inner membranes are closely opposed [8, 9], allowing transfer of small molecules and proteins between the cytoplasm and the matrix space. There are also connections between the infrastructure of the mitochondrion and the cytoskeleton. I have shown one such connection that links actin filaments in the cytoplasm with proteins in the mitochondrial membrane and inside to link to mitochondrial DNA, based on schematic diagrams from a review article [10]. To generate a model of this complex, the sizes and locations of intermembrane regions in the complex of MDM proteins (mdm10, mdm12, mmm1, mdm31, and mdm32) were taken from protein sequences at the ExPASy proteomic server (http://ca.expasy.org), and the Arp2/3 protein at the actin junction is taken from electron micrograph reconstruction [11].

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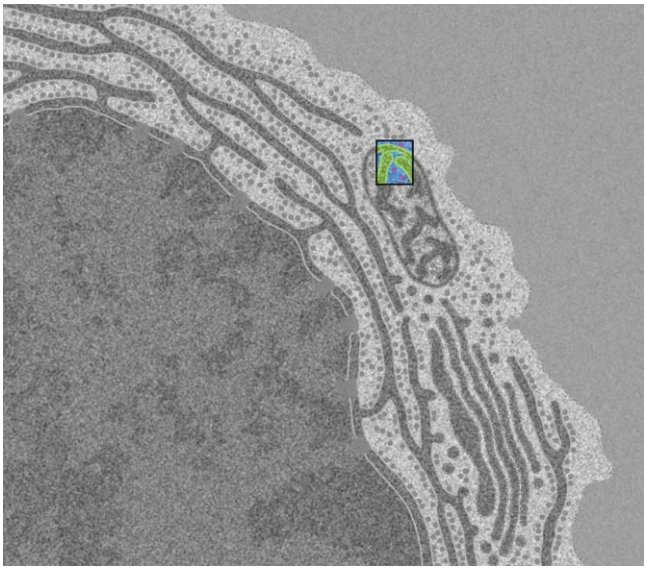

FIG. 1. **Simulated cross section through a eukaryotic cell, showing the location of the enlarged portion in Figure 2.**

The innermost compartment of the mitochondrion is termed the matrix. The matrix is one of the most densely packed portions of the cell; some estimates place it at greater than 50% protein [12]. It includes the enzymes of the tricarboxylic acid cycle and enzymes for fatty acid utilization, as well as an entire set of protein synthesis machinery, including DNA, polymerases, ribosomes, and transfer RNA, all described in more detail later.

ATP SYNTHESIS

The major task of the mitochondrion is the production of ATP. The enzymes of the tricarboxylic acid cycle are found in the matrix, along with a collection of enzymes for utilization of fat and other energy-rich molecules. These include two large multiprotein complexes, pyruvate dehydrogenase complex and alpha-ketoglutarate dehydrogenase complex, both drawn based on results from crystallography and electron microscopy [13, 14]. I have included little tails to represent the lipoic acid cofactors that are involved in substrate transfer between the subunits. Other tricarboxylic acid cycle enzymes are drawn based on crystallographic structures: citrate synthase (1cts), aconitase (1aco), isocitrate dehydrogenase (1lwd), succinyl-CoA synthetase (2fp4), succinate dehydrogenase (1zoy), fumarase (1yfm), and malate dehydrogenase (1mld). (4-letter codes included in this paper are accession codes for atomic structures at the Protein Data Bank, http://www.pdb.org.)

The electron transport chain is found in the membranes of the cristae. Decades ago, it was discovered that the large protein complexes of the electron transport chain are not arranged in a structured chain—rather, electrons are transferred by random diffusion of small carrier molecules between randomly-placed protein complexes in the membrane [15, 16]. More recently, however, there has been growing evidence that the proteins form a supercomplex in some organisms [17]. For this illustration, I have chosen to depict them as separate complexes in the membrane. NADH dehydrogenase complex is based on an electron micrograph reconstruction [18], and the other large complexes are based on atomic structures: cytochrome reductase (1bgy) and cytochrome oxidase (1oco). I have also included coenzyme q in the membrane and cytochrome c in the intercristal space (3cyt). Cytochrome c peroxidase (2pcc), also in the intercristal space, may play a role in detoxifying peroxide that leaks from the electron transport chain. ATP synthase (one of the wonders of the biomolecular world) is modeled after several crystal structures (1c17, 1e79, 1l2p, 2a7u).

I have also included a number of other enzymes involved in energy metabolism. I identified these using results from 2D gel electrophoresis [19] and by searching for "mitochondrial matrix human" in ExPASy. In the outer membrane, I included the membrane-linked enzyme monoamine oxidase (1gos). In the intermembrane and intercristal spaces, these include: creatine kinase (1qk1), adenylate kinase (1ak3), nucleotide diphosphate kinase (1ndl), and sulfate oxidase (based on molecular weight from ExPASy). In the matrix, I included manganese superoxide dismutase (1ja8) and copper-zinc superoxide dismutase (2sod), pyruvate carboxylase (2qf7), acyl-CoA dehydrogenase (3mdd), ornithine transcarbamoylase (1fb5), ornithine aminotransferase (2can), and glutamate dehydrogenase (1aup). I also included two forms of glycerol-3-phosphate dehydrogenase (1x0x), GPD1 in cytoplasm and GPD2 in the mitochondrion, which together form a shuttle.

APOPTOSIS

The mitochondria also play an important role in apoptosis (programmed cell death). Cytochrome c is a "moonlighting" protein that plays a secondary role entirely unrelated to its primary function as an electron transport protein. If it leaks into the cytoplasm, it initiates the cascade of apoptosis. A complex set of cellular machinery receives apoptotic signals and then ruptures the outer mitochondrial membrane, thus releasing cytochrome c and other apoptotic proteins.

In this illustration, I included many of the mitochondrial proteins of apoptosis, including several in the outer membrane and in the intermembrane space. In the outer membrane, BID (2bid) and Bcl-2 (1g5m) are ready to initiate an apoptotic cascade [20]. Fzo1 protein (based on a schematic diagram of its domain structure [21]) may play a role in fusion of mitochondria. In the intermembrane space, I included Smac (1few), which plays a role in activating caspases, and the apoptotic ribonuclease EndoG (3ism) and serine protease HtrA2/Omi (1lcy). I also included several cytoplasmic apoptotic proteins, including caspase-7 in an inactive complex with XIAP (1nw9) and Apaf-1 (1z6t), the protein that associates with cytochrome c when it is released from the mitochondrion, triggering the apoptotic cascade.

I created a separate illustration to show a rupturing mitochondrion in the process of losing its cytochrome c, and the consequences for the cell. A detail is included in

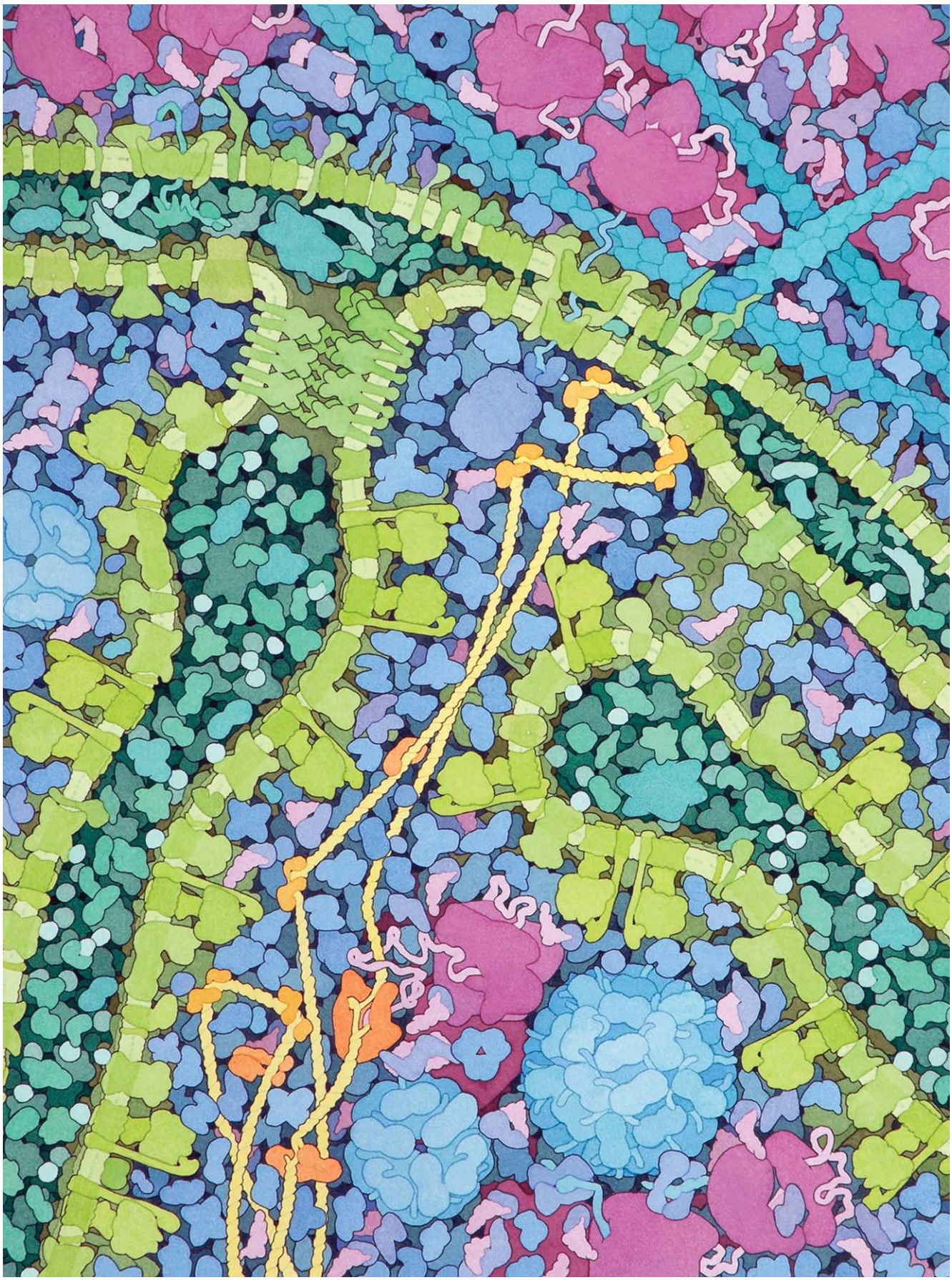

FIG. 2. **Cross section through a mitochondrion at 1,000,000× magnification.** All macromolecules and membranes are shown, but small molecules, ions and water are omitted for clarity. The cellular cytoplasm is at the top, and the mitochondrion fills most of the lower portion of the image. Soluble proteins are shown in shades of blue, and membranes and membrane-bound proteins are shown in green. RNA is shown in pink and ribosomes in magenta. DNA and DNA-associated proteins are shown in yellow and orange.

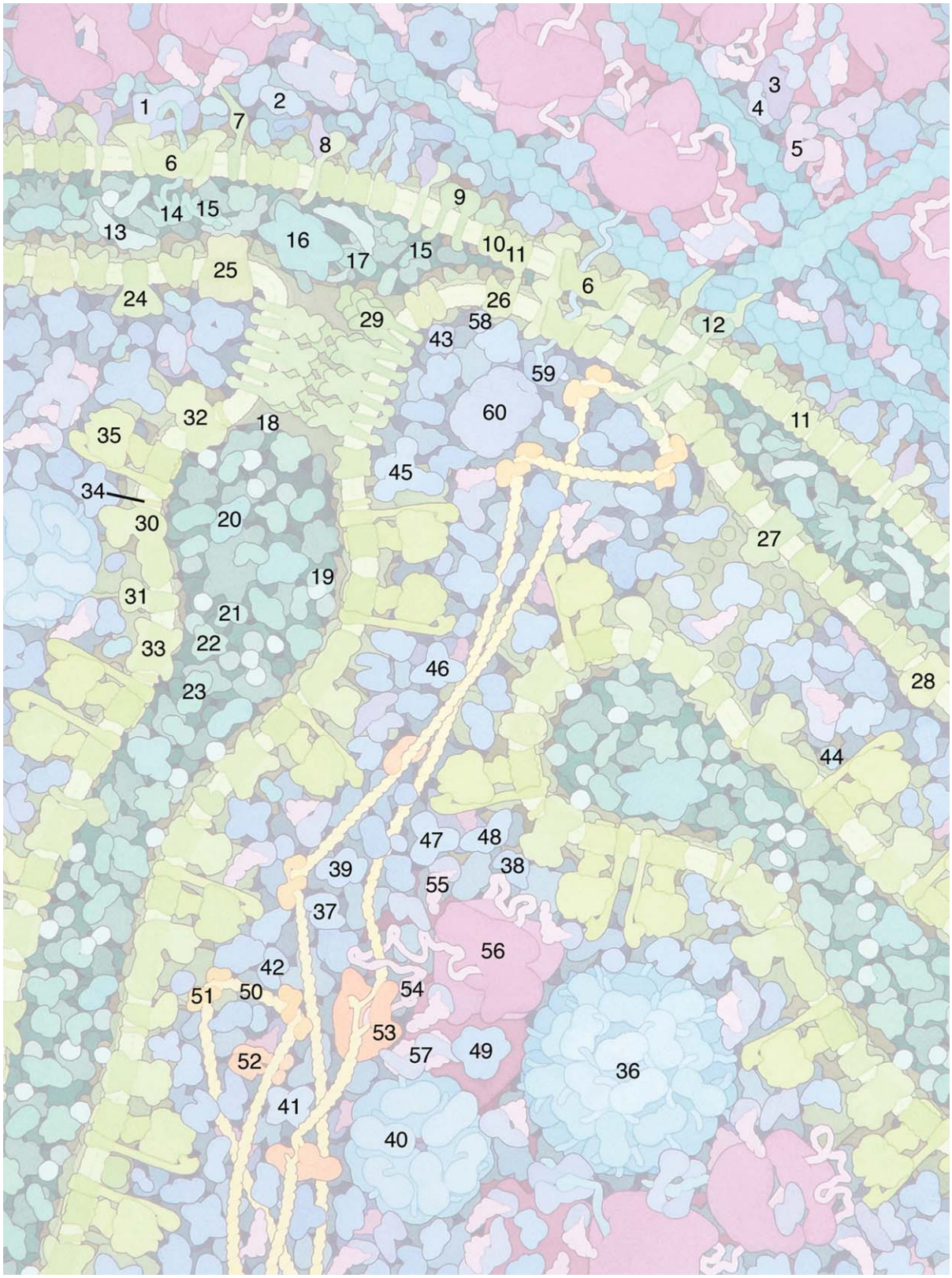

FIG. 3. **Key to Figure 2.** *Cytoplasm:* 1. Hsp90; 2. glycerol-3-phosphate dehydrogenase 1; 3. caspase-7; 4. XIAP; 5. Apaf-1. *Outer Membrane:* 6. protein transporter; 7. Fzo1; 8. BID/Bcl-2 complex; 9. monoamine oxidase; 10. PBR; 11. VDAC; 12. MDM complex bound to Arp2/3 and actin. *Intermembrane Space:* 13. Smac; 14. TIM9/10; 15. HtrA2/Omi; 16. creatine kinase; 17. EndoG. *Intercristal Space:* 18. cytochrome c; 19. cytochrome c peroxidase; 20. glycerol-3-phosphate dehydrogenase 2, 21. adenylate kinase; 22. nucleotide diphosphate kinase; 23. sulfate oxidase. *Inner Membrane:* 24. magnesium transporter; 25. RyR1; 26. ADP/ATP carrier; 27. potassium channel; 28. ABC-type transporter; 29. Opa1; 30. NADH dehydrogenase; 31. succinate dehydrogenase; 32. cytochrome bc1 reductase; 33. cytochrome oxidase; 34. coenzyme q; 35. ATP synthase. *Matrix: TCA enzymes:* 36. pyruvate dehydrogenase complex; 37. citrate synthase; 38; aconitase; 39. isocitrate dehydrogenase; 40. alpha-ketoglutarate dehydrogenase complex; (31. succinate dehydrogenase); 41. fumarase; 42. malate dehydrogenase; *Other enzymes:* 43. manganese superoxide dismutase; 44. copper-zinc superoxide dismutase; 45. pyruvate carboxytransferase; 46. acyl CoA dehydrogenase; 47. ornithine transcarbamoylase; 48. ornithine aminotransferase; 49. glutamate dehydrogenase; *Protein synthesis:* 50. DNA; 51. TFAM; 52. steroid receptor; 53. RNA polymerase; 54. messenger RNA; 55. transfer RNA; 56. ribosome; 57. aminoacyl-tRNA synthetase; 58. cyclophilin D; 59. MPP, 60. Hsp60.

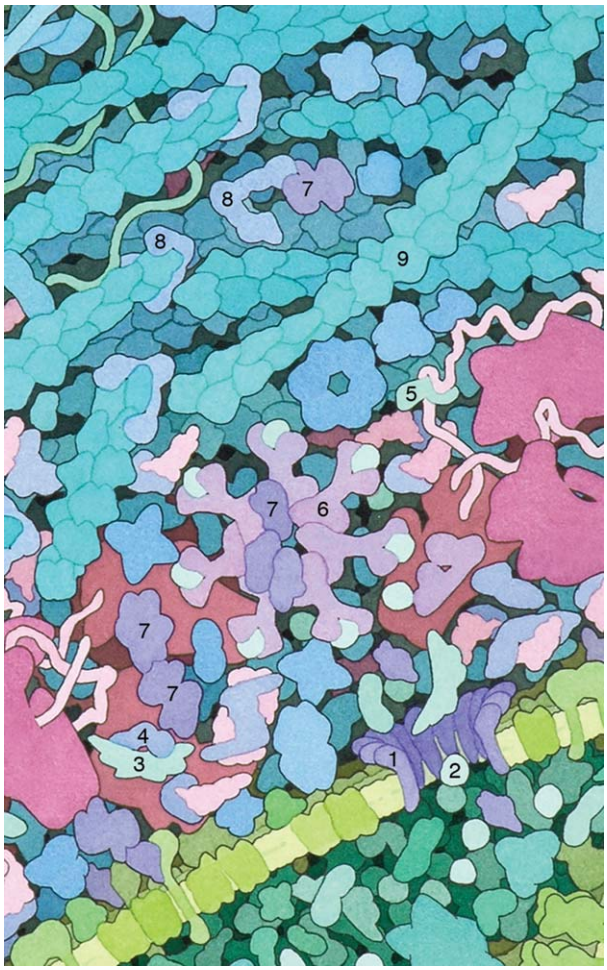

FIG. 4. **Apoptosis (detail)**. The outer membrane of the mitochrondrion has been ruptured, releasing mitochondrial proteins that trigger and assist with apoptosis. 1. BAX; 2 cytochrome c; 3. Smac; 4. XIAP; 5. EndoG; 6. Apaf-1; 7. caspase; 8. gelsolin; 9. actin.

Fig. 4. The BAX protein has formed a speculative pore through the membrane [22], and presumably the intercristal space is also breached, allowing cytochrome c, EndoG [23], HtrA2/Omi [24], and Smac to exit. Cytochrome c associates with Apaf-1 to form a beautiful seven-membered apoptosome [25], which then activates caspases and starts the destruction of proteins throughout the cell. Smac removes XIAP from caspases, further assisting the activation. EndoG attacks messenger RNA molecules, halting translation of new proteins, and caspases activate enzymes such as gelsolin [26], which disassembles actin filaments.

MITOCHONDRIAL DNA

The mitochondrial matrix includes an entire mechanism for protein synthesis, different and separate from the protein synthesis machinery in the cytoplasm. Moreover, the mitochondrial ribosomes, polymerases and other protein synthesis molecules are very similar to those found in bacteria. The presence of this bacteria-like protein synthesis machinery is one of the main clues that mitochondria evolved from endosymbiotic bacteria. This machinery is an evolutionary artifact, however, and is currently used to make only 13 proteins involved in the electron transfer chain and ATP synthase, as well as 22 mitochondrial transfer RNA and two ribosomal RNA [27]. The 700 or so other mitochondrial proteins [28], including the bacteria-like polymerases and translation factors, are made in the normal way by cytoplasmic ribosomes, and then imported into the proper compartment in the mitochondrion [29].

The mitochondrial ribosomes, transfer RNA, polymerases and other protein synthesis machinery are more similar to their bacterial counterparts than to the ribosomes and other machinery found in the cytoplasm of the cell. I have included many of these molecules in the illustration, including ribosomes modeled after the ones in *Thermus thermophilus* (1yl3, 1yl4), bacterial elongation factors EFTu (1ttt) and EFG (1dar), transfer RNA (1ttt), aminoacyl-tRNA synthetases (1asz, 1ffy, 1gax, 1euq, 1eiy, 1qf6), RNA polymerase (2e2i), and DNA. The chaperone Hsp60 was modeled after bacterial GroEL (1aon). The mitochondrial transcription factor TFAM is based on HMG-domain structures (1qrv, 2gzk), and the steroid receptor is based on the nuclear vitamin D receptor (1db1,1kb6).

TRANSPORT

Since mitochondria are surrounded by two membranes, there are potential challenges with transport. Like the outer membrane in *Escherichia coli*, the outer membrane of the mitochondrion is leaky. It is filled with voltage-dependent anion channel (VDAC), a protein similar to bacterial porins that forms a pore through the membrane. This pore is roughly 20–30 Å in size, large enough for small molecules like ATP and glucose to pass, but small enough to exclude larger molecules like proteins. VDAC is the most abundant protein in the outer membrane, and was found in large, densely packed clusters in a high-resolution atomic force microscopy study [30], where each pore was spaced by about 53 Å apart. I have modeled the structure after PDB entry 2k4t. I also included the PBR (peripheral-type benzodiazepine receptor), a protein involved in transport of cholesterol and other molecules [31]. I have shown it interacting with VDAC and the ADP/ATP carrier at the contact site shown at the center of membrane in Fig. 2 [9].

The inner membrane, however, must be sealed to allow generation of proton gradients to power ATP synthase. A large collection of transporters facilitate movement of molecules across this membrane. These include at least 49 different specific transport proteins of the mitochondrial carrier family [32]. The most abundant is the ADP/ATP carrier (1okc) that transports nucleotides in and out of the mitochondrion [33]. Other members of the mitochondrial carrier family, which have similar pore-like structures, transport pyruvate and other important metabolites, cofactors, and inorganic molecules. I have also included several other classes of transporters and channels [34], including the magnesium transporter (2bbj), RyR1 (ryanodine receptor, a possible calcium transporter), a potassium channel (1f6g) and several ABC-type transporters modeled after the vitamin B12 transporter system (2qi9).

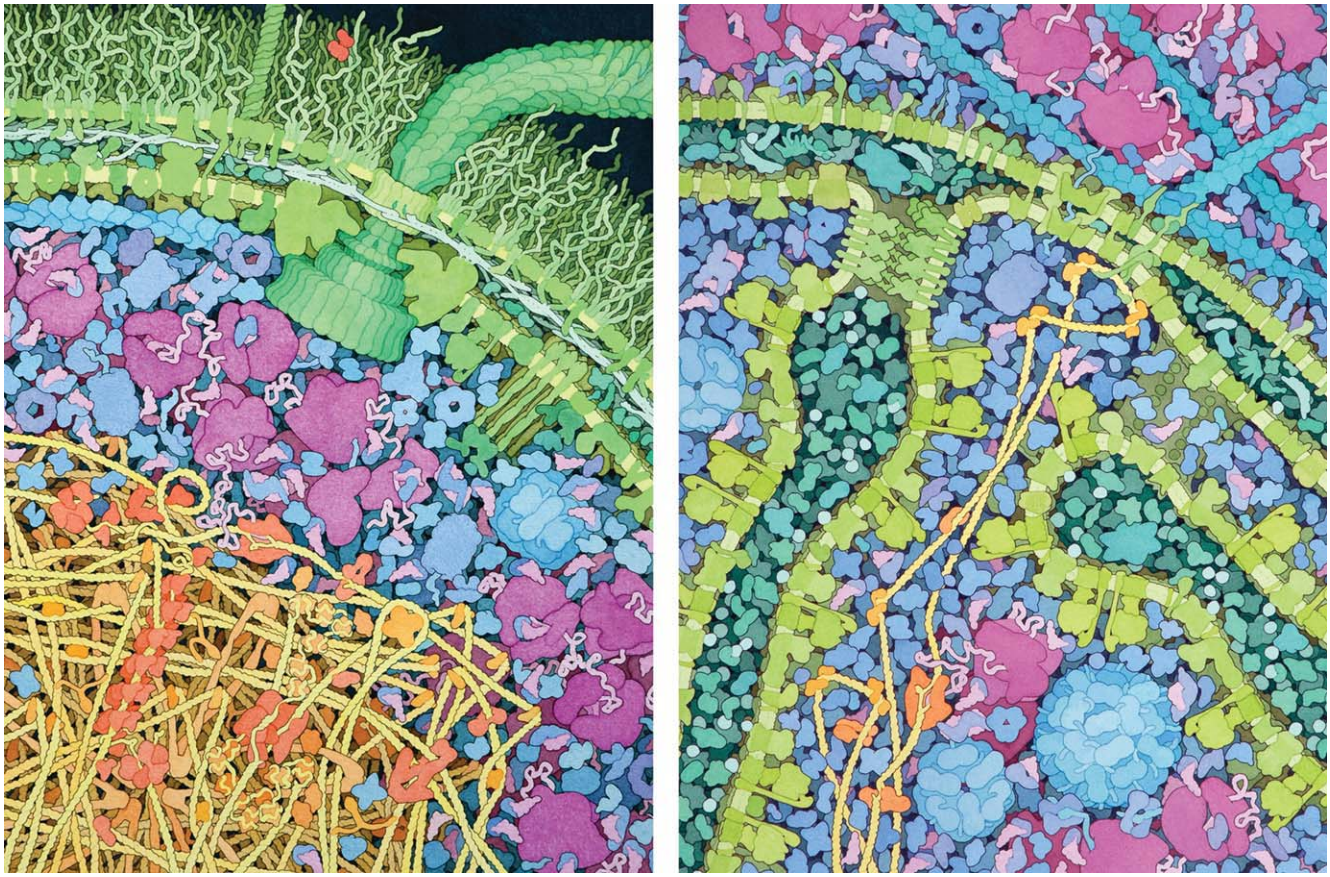

FIG. 5. **Illustrations of cross sections of *Escherichia coli* (left) and the mitochondrion (right) are designed to show structural and functional similarities.**

There are also several specific transporters for importing proteins into the intermembrane space or into the matrix [29]. I included two separate protein transporters. The one on the left in Fig. 2 is shown transporting an unfolded protein from Hsp90 (2cg9) in the cytoplasm and delivering it to the intermembrane space, where it is picked up by the chaperonin TIM9/10 (2bsk) and possibly HtrA2/Omi (1lcy). The one on the right is shown transporting the protein through both membranes into the matrix, where it is processed by the mitochondrial-processing peptidase MPP (1hr6).

AESTHETIC AND PEDAGOGIC CONSIDERATIONS

I designed the layout of this painting to match the layout of the *Escherichia coli* picture that was presented in an earlier chapter in the book (Fig. 5), to highlight the evolutionary relationship between mitochondria and bacteria [2, 35], and to show their many structural and functional similarities [3]. As with the other illustrations, a cross-section at 1,000,000× magnification is depicted, with the section chosen to place the membranes roughly perpendicular to the plane of the cut. The level of magnification is a compromise between two pedagogic goals: to show the entire subject in one comprehensive illustration, but still be able to see the shape and form of each macromolecule. This magnification allows display of a section of the mitochondrion that is large enough to see the major ultrastructural features as well as the molecular details of its major functions.

The colors are chosen to match the colors used for illustrations throughout the book. This scheme is designed to highlight the functional compartments of the cell/organelle, as described in my earlier article [36]. In this scheme, soluble proteins are shown in shades of blue, and membranes and membrane-bound proteins are shown in green. RNA is shown in pink and ribosomes in magenta. DNA and DNA-associated proteins are shown in yellow and orange. This consistent scheme integrates the illustrations throughout the book, allowing readers to compare the features presented in the different scenes, but it also poses a few problems. For instance, the mitochondrial matrix is rendered with similar colors as the cytoplasm. This has the advantage of showing the similarities between the two compartments (e.g., that both compartments are performing protein synthesis with similar molecular machinery), but it introduces the disadvantage of making it difficult to distinguish the mitochondrion from the cytoplasm. The consistent coloring scheme also required one unfortunate choice: after much vacillation, I chose to color cytochrome c in turquoise to match the other molecules in the intermembrane space, rather than coloring it its actual red color.

Acknowledgments— This manuscript 20555 is from the Scripps Research Institute.

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Basic Lesson Plans

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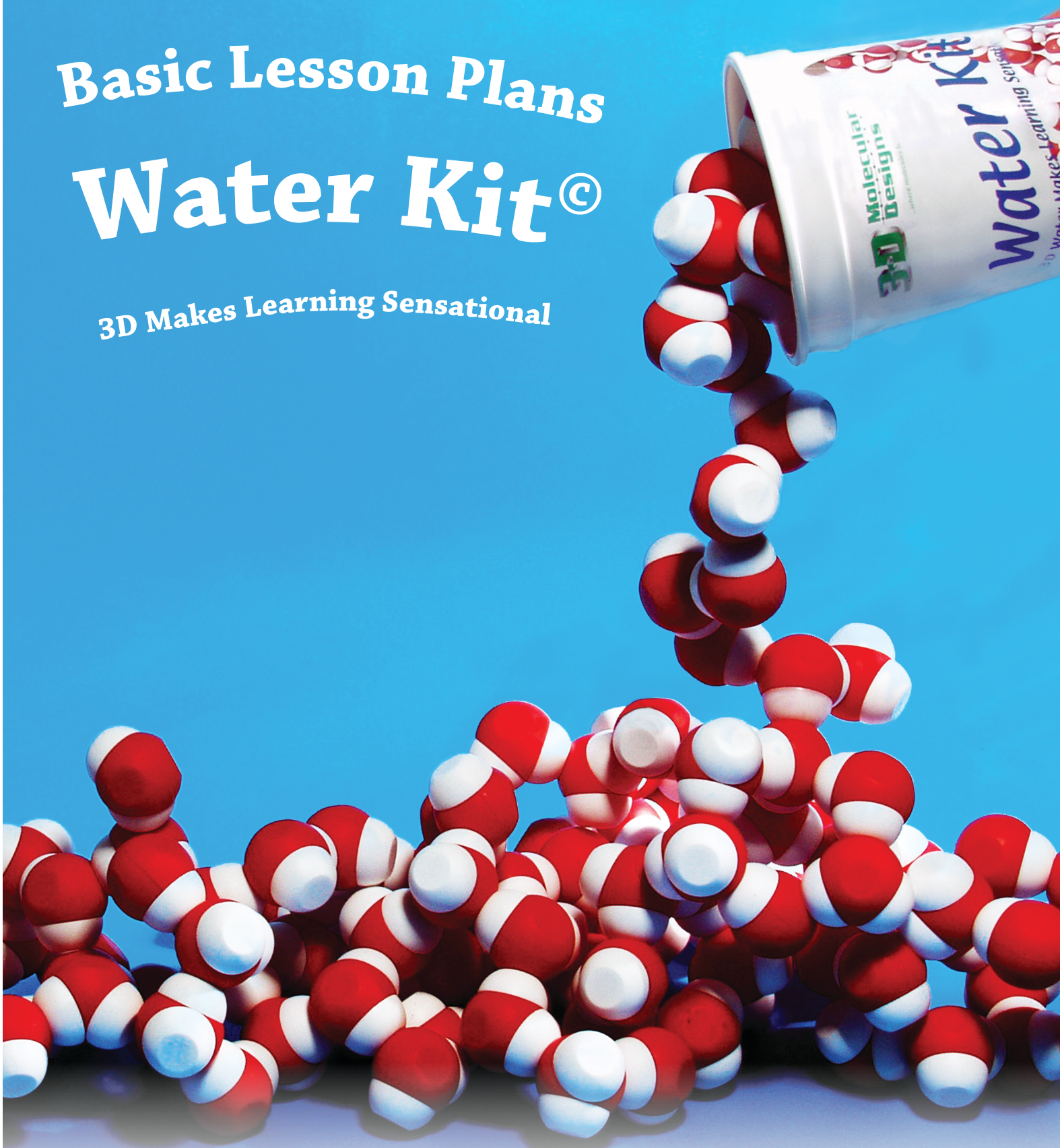

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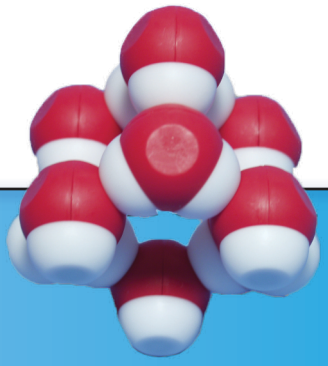

Table of Contents

Project WET (Water Education for Teachers)

3D Molecular Design's Water Kit© can be used to teach many of the concepts in Project WET's K-12 Curriculum Guide. Project WET is a nonprofit water education program that facilitates and promotes awareness, appreciation, knowledge and stewardship of water resources through the dissemination of classroom-ready teaching aids. Teachers can use the Water Kit© as part of many Project WET lessons or as an extension to the lessons. Please see the document Project WET and the Water Kit©. The chart shows which Project WET lessons the Water Kit© can be used with to enhance understanding.

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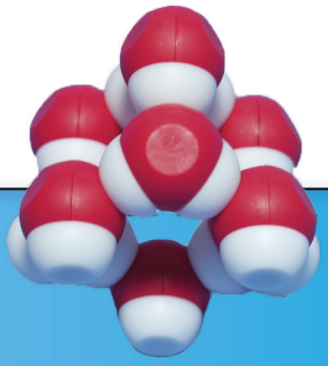
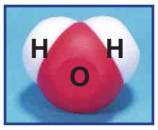
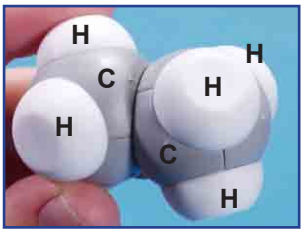


Contents of Water Kit©

Water Kit© Contents Per Cup

24 Oxygen* Pieces (Used to Assemble 12 Water Molecule Models)
24 Hydrogen* Pieces (Used to Assemble 12 Water Molecule Models)
1 Chloride* (Chlorine) Model
1 Sodium* Model
2 Carbon Pieces (Used to Assemble 1 Ethane Model)
6 Hydrogen** Pieces without Magnets (Used to Assemble 1 Ethane Model)
1 Post (Used to Assemble 1 Ethane Model)
1 Hydroxyl* Group Model (Preassembled)





Ethane molecule (CH_3CH_3)

Ethanol molecule (CH_3CH_2OH)

**North and south poles on embedded magnets simulate the partially positive and negative charges of oxygen and hydrogen atoms, and chloride and sodium ions.*

***The hydrogen pieces for the ethane model do not include embedded magnets, since ethane is nonpolar.*

Misconception Caution – Although the use of magnets to represent the partial positive and negative charges that exist on the hydrogen and oxygen atoms of water is a powerful teaching tool, it is important to explain to students that atoms are not magnets.

⚠WARNING:
SMALL MAGNETS — This product contains small magnets. Swallowed magnets can stick together across intestines causing serious infections and death. Seek immediate medical attention magnets are swallowed or inhaled.

⚠WARNING:
CHOKING HAZARD — This product contains small parts and should be kept out of the reach of children under the age of 3, because the parts or their pieces may present a choking hazard to small children.

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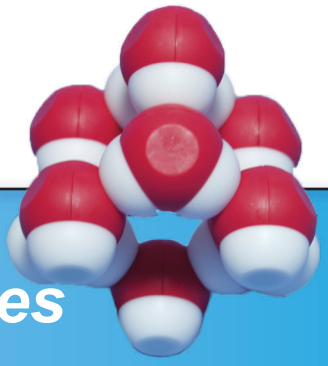
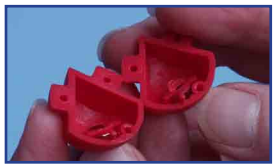


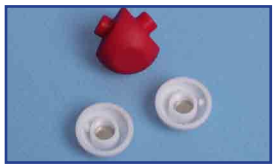
Assembling 3D Water Molecules

Start by selecting two red oxygen pieces and two white hydrogen pieces.



Take one red piece in each hand. You will see one post and one hole on the inside rim of each red piece.

Position the post of one piece into the hole of the other piece. Place your thumbs on the flat surface of each oxygen piece and push the two pieces together until they fit tightly and you no longer see a space between the pieces.



The red oxygen atom will now roughly resemble a sphere with two knobs sticking out of each half.

Take the red oxygen atom in one hand and one white hydrogen piece in the other and place the open side of the hydrogen piece onto the knob on the oxygen. Push the two together until the hydrogen fits tightly onto the oxygen, with no space between the two pieces.

Repeat with a second white hydrogen piece.

You now have one complete water molecule. Repeat steps 1 through 6, until 12 water molecules are assembled.

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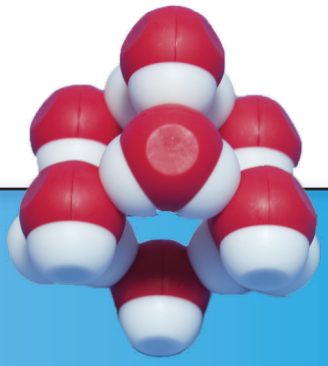


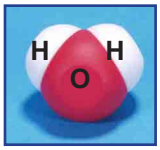
Basic Lesson Plans

Polarity

General Information

Understanding the unique characteristics of water begins with understanding polarity.

Polarity refers to the partial positive charge (+) and partial negative charge (-) that a molecule has when electrons are unequally shared between two or more atoms. Molecules that have partial charges are polar molecules. Water molecules are **polar molecules**.



In a water molecule, each hydrogen atom has a partial positive charge and the oxygen atom has two partial negative charges.

Some molecules do not have unequal regions of charges and therefore do not interact with polar molecules. These are **nonpolar molecules**. Oil and ethane are examples of a liquid and a gas composed of nonpolar molecules.

Water Kit©

The water molecules in the Water Kit© have been embedded with magnets to help students experience what positive (+) and negative (-) charges feel like as they interact with other polar molecules. Through manipulation of the 3D water molecule models, it becomes easy for students to understand the physical and chemical properties of water.

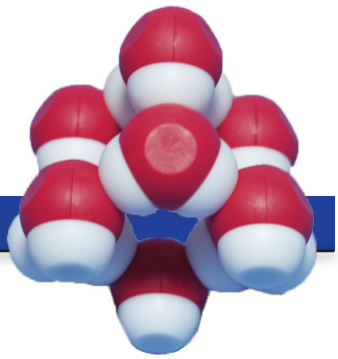
Embedded magnets are positioned at 105º from each other.

There are two magnets embedded in the oxygen at 105º degrees from each other. These represent the two partial negative (-) charges.

One magnet in each of the hydrogen atoms represents the partial positive (+) charges. The hydrogen molecules are also positioned 105º degrees apart.

The location of these four magnets (representing charges) makes it possible for your students to create a *tetrahedral* structure. A tetrahedral structure is formed when one water molecule interacts with four other water molecules.

Tetrahedral Structure



In the Classroom

Hand out one 3D water molecule model to each student.

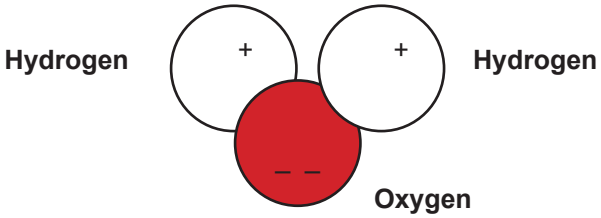
- What is the chemical formula of water?
  H_2O

Have your students discuss what the H and the O represent. Refer to the periodic table of elements.

- What does the 2 between the hydrogen and oxygen stand for?
  When a number # is placed after a symbol, it means there is more than one atom of that element and when there is no number after a symbol, it means there is only one atom of that element.

Have your students identify which parts of the model represent the hydrogen and which the oxygen. Clarify that the colors on the model are used by chemists to represent hydrogen (white) and oxygen (red), but water, as a real molecule, is not colored. Emphasize that it is impossible to see a single water molecule with the naked eye or even with the most powerful light microscope.

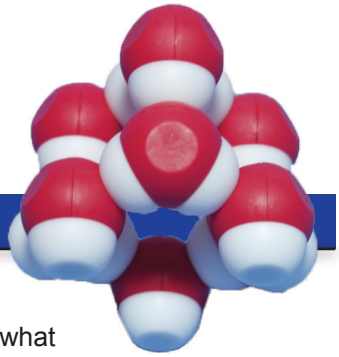
Have your students draw and label a water molecule. Remind your students that each hydrogen atom has a partial positive charge and the oxygen atom has two partial negative charges. Explain that they can show this on their drawing by placing a + symbol on each hydrogen and two – symbols on the oxygen.



Your students can now predict, what will happen when water molecules interact. Hand out one more 3D water molecule model to each student and let him or her investigate whether his or her prediction is correct.

- What will happen when two water molecules bump into each other?
  - When the oxygen atoms of 2 different water molecules come together, they repel.
  - When the hydrogen atoms of 2 different water molecules come together, they repel.
  - When an oxygen atom and a hydrogen atom from two different water molecules come together, they attract.

Have the students draw and label what they have discovered. Then, have your students write about what they have observed.



(In the Classroom Continue)

Difference Between Magnetism and Polarity

Display two large horseshoe-shaped, or other, magnets and ask your students what they know about magnets. List and diagram the student's answers on the board.

- What are the similarities and differences between magnets and water molecules?
  Atoms are not really magnetic. However, the north and south poles of magnets can be used to effectively model the partial positive and partial negative charges that exist on the polar water molecule.

Water Facts

- How many water molecules are in a single drop of water?
  There are 3 x 1021 of molecules in one drop of water (3,000,000,000,000,000,000,000). This number can be read, "three times ten to the twenty first power."

- At the scale of the water molecule models, how large would a water drop be?
  Clarify that it is impossible to see a single water molecule with the naked eye or even with the most powerful microscope. A water drop made of 3 x 1021 of 3D water molecule models would be ***larger*** than the earth.

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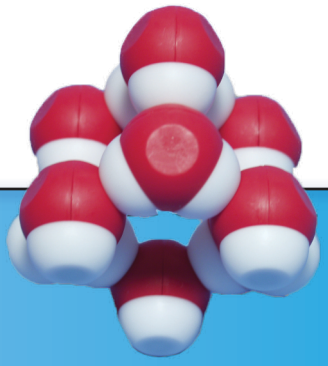


Hydrogen Bonding

General Information

A **covalent bond** is formed when two atoms share two electrons. A covalent bond is an ***intramolecular*** bond within one molecule. Covalent bonds can be either polar (which have partially charged atoms) or nonpolar (without charged atoms).

Hydrogen bonds are intermolecular forces between *two* molecules where a positively charged hydrogen atom interacts with a negatively charged fluorine, nitrogen or oxygen atom in a second molecule.

• ***Water fact:*** *A hydrogen bond is about 1/20 as strong as a covalent bond.*

An **ionic bond** is the complete transfer of an electron between two atoms resulting in one positively and one negatively charge atom. *Ionic bonds are intramolecular bonds within one molecule.*

Ions are charged atoms that have gained or lost electrons as a result of an ionic bond.

Water Kit©

The magnets in the 3D water molecule models simulate the intermolecular force of two polar water molecules forming a hydrogen bond. All of the bonds between multiple water molecules are hydrogen bonds. The bonds between water molecules and the OH group on the ethanol are hydrogen bonds.

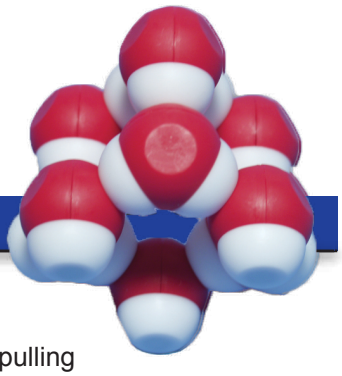
In each individual 3D water molecule models, the bonds between the hydrogen atoms and the oxygen atom are covalent bonds. Covalent bonds are also formed between all of the atoms that form ethane and ethanol. These are all intramolecular bonds. Water molecules and ethanol molecules are polar covalent molecules while the ethane is a nonpolar covalent molecule.

Ionic bonds are formed between the sodium and chloride ions. This is an ***intra***molecular bond.

The attraction between the negative charge of the oxygen and the positive charged of the sodium ion is an ***inter***molecular force.

Intramolecule force

Partially positively charged hydrogen atoms (see water and ethanol molecules) will interact with negatively charged chloride ions. ***This is not called a hydrogen bond because the chloride ion is not one of the three elements listed above (fluorine, nitrogen or oxygen)***. This is another type of intermolecular force that is covered in the Solubility Section.



Hydrogen Bond Activity and Questions

Give each student two 3D water molecule models.

Have your students *break* the hydrogen bond between the water molecules by pulling them apart. Next, have them compare this to the relative strength of a covalent bond by pulling a hydrogen atom off a water molecule. (Use a fingernail to pry the hydrogen off the oxygen if needed.)

- What is the intermolecular force that holds these two water molecules together?
  Hydrogen bond.

- What is the intramolecular bond that holds the hydrogen atoms and oxygen atom within a water molecule?
  Covalent bond.

- Is a water molecule a polar covalent molecule, or a nonpolar covalent molecule? Why?
  Water molecules are polar covalent molecules because electrons are shared within the molecule and the sharing of the electrons is unequal which results in partially positively charged hydrogen atoms and a partially negatively charged oxygen atom.

Ethane

Give each group of students an ethane molecule.


Ethane CH_3CH_3

- What is ethane?
  Ethane is a short hydrocarbon. Hydrocarbons are combinations of carbon and hydrogen atoms. Ethane is an odorless, colorless gas that can be used as a fuel, a freezing agent, and in making other chemicals.

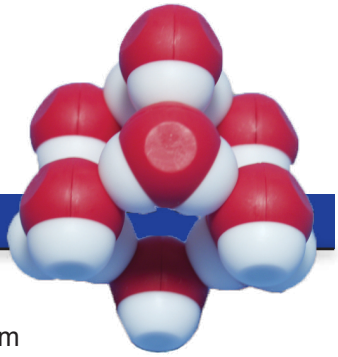
- What do the different colors on the ethane molecule represent? What is the chemical formula of ethane?
  White represents hydrogen atoms and the gray represents carbon. CH_3CH_3.

- What is the intramolecular bond that holds the hydrogen and carbon atoms within an ethane molecule?
  Covalent bonds.

- Is an ethane molecule a polar covalent molecule, or a nonpolar covalent molecule? Why?
  Ethane molecules are nonpolar covalent molecules because electrons are equally shared within the molecule. This equally charged molecule is nonpolar.

- Will ethane form a hydrogen bond with water? Why or why not?
  No, ethane will not form a hydrogen bond, or ionic bond with water or any other polar molecule because it is nonpolar. Ethane does not have any partial positive or negative charges.

- Will two ethane molecules form bonds with each other? Why or why not?
  No. Ethane molecules will not form covalent, ionic or hydrogen bonds with each other. Ethane molecules have small intermolecular forces and will interact under the certain temperature and pressure conditions to form liquids and solids.



Hydrogen Bond Activity and Questions (continued)

Give each student group an OH (hydroxyl) group* and have them replace the detachable hydrogen with the OH group. They have just changed the model from ethane to ethanol.

• What do the different colors represent? What is the chemical formula of ethanol?
White represents hydrogen atoms, the gray represents carbon atoms, and the red represents oxygen atoms. CH_3CH_2OH.

• Will ethanol and water interact? How?
The water molecules will form hydrogen bonds with the oxygen atom and the single polar hydrogen atom on the ethanol.

• Will ethanol molecules interact with each other? Why?
Yes. Ethanol will form hydrogen bonds with other ethanol molecules.

•What is sodium chloride?
Show a sodium chloride molecule to your class.

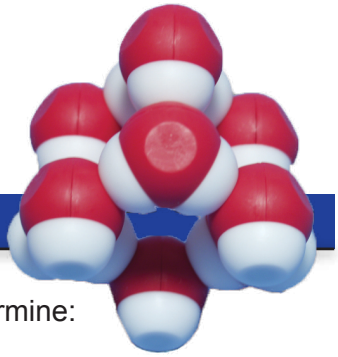
Sodium chloride is often called table salt or rock salt.

• What do the colors represent? What is the chemical formula of sodium chloride?
The blue atom represents sodium, the green chlorine. NaCl.

• The *intra*molecular bond that holds these ions together is ionic. What does this mean?
Ionic bonds are formed when an electron is completely transferred from one atom to another. This results in a positively charged ion (the atom that looses an electron) and a negatively charged ion (the atom that gains an electron).

• Will a water molecule interact with sodium chloride? Why?
Yes, water molecules and sodium chloride ions will interact. The partially charged water molecule will interact with the fully charged sodium chloride. These are intermolecular forces.

* The OH group, also called a hydroxyl group, is a functional group of the ethanol molecule. It is a way to refer to the part of the molecule that is polar – the oxygen atom and the hydrogen atom that is covalently bonded to the oxygen atom.



Hydrogen Bond Activity and Questions (continued)

Give each group of students the sodium chloride ions. Ask them to determine:
- which ion the hydrogen atoms interacts with.
- which ion the oxygen atom interacts with. Have your students record their findings.

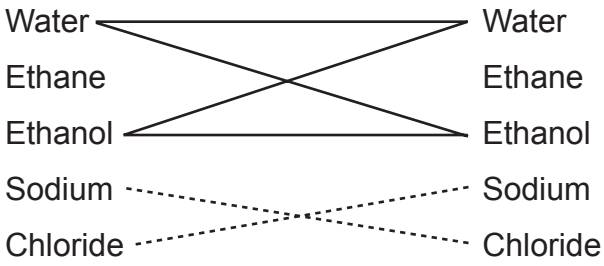
- Are chloride ions positively or negatively charged? Why?
  Chloride ions interact with hydrogen atoms in water molecules. Since hydrogen atoms are partially positively charged, the chloride ions are negatively charged.

- Do hydrogen atoms and chloride ions form hydrogen bonds? Why?
  No, the intermolecular force between the hydrogen atom and chloride ion is not a hydrogen bond because hydrogen bonds are only formed between hydrogen and fluorine, nitrogen and oxygen atoms.

- Are sodium ions positively or negatively charged? Why?
  Sodium ions interact with oxygen atoms in water molecules. Since oxygen atoms are partially negatively charged, the sodium ions are positively charged.

- Will sodium chloride interact with ethane? With ethanol? Why or why not?
  Sodium chloride will not interact with ethane because ethane is nonpolar. Sodium chloride will interact with the ethanol.

Have your students list water, ethane, ethanol, sodium and chloride in two columns. First, have them draw solid lines between the molecules that will form hydrogen bonds. Second, have them draw dotted lines between the ions that will form ionic bonds.



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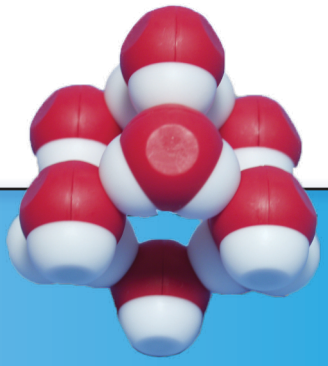


Adhesion, Cohesion & Capillary Action

General Information

The attraction between two like molecules is **cohesion**.

The attraction between two unlike molecules is **adhesion**.

Adhesion and cohesion are intermolecular forces between two molecules. These forces are only called hydrogen bonds when a hydrogen atom and a fluorine, nitrogen, or oxygen atom are attracted to each other. Capillary action is the spontaneous rising of a liquid in a capillary (small diameter) tube.

Capillary action in plants is a good example of adhesion and cohesion. The inner surface of the xylem, the cell wall of a plant, contains positive and negative charges. Water forms hydrogen bonds with the xylem. This is called adhesion. As water creeps up the sides of the xylem (adhesion) the water molecules in the middle connect to other water molecules because of cohesion. The water moves up as the water molecules at the top of the xylem enter the leaves and evaporate (move out of the stomata in the leaf). When a water molecule leaves the leaf, the molecule behind it moves up causing a general movement of the water up the tree.

Tip: *Adhesion* **adds** *a different molecule to the substance.*

Water Kit©

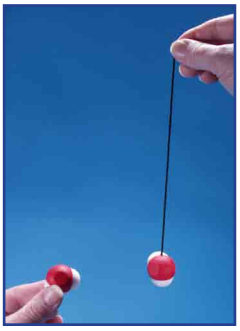
When 3D water molecule models are attracted to another substance, the force is adhesion.

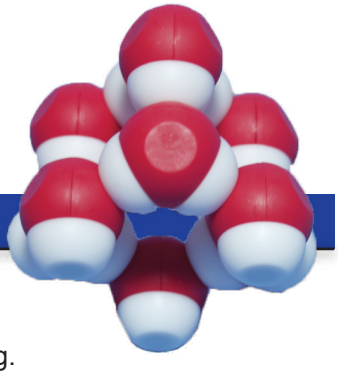
When 3D water molecule models are attracted to each other, the force is cohesion.

Activity

Demonstrate adhesion and cohesion by tying a string (dental floss or fishing line works well) around a 3D water molecule models. Hold the string in one hand and another 3D Water Molecule your other. Slowly bring the second 3D Water Molecule toward the one on the string. Students should see the hanging molecule move toward the molecule in your hand. Explain that this attraction is cohesion.

Next, bring an ethanol molecule toward the hanging 3D water molecule models.

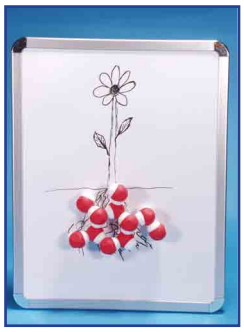




Adhesion, Cohesion & Capillary Action

- What is the attraction between these molecules?
  The force is called adhesion because two different molecules are attracting.
  Test your students' grasp of adhesion and cohesion by hanging an ethanol molecule. Bring a second ethanol molecule toward the first.

- What is the attraction between these molecules?
  The force is called cohesion because two like molecules are attracting.



Demonstrate capillary action by drawing a plant on a whiteboard. If your whiteboard is not magnetic, use a small magnet white message board. Place 11 of the 3D Water Molecules at the root of the outlined plant. Pull the molecules up the roots, through the stem and out the leaves of your plant with the molecule attached to the string. Note: Some of the water molecules will bond with the board and some to other water molecules.

Explain that this is capillary action. Water molecules form hydrogen bonds with cellulose in the xylem. The cellulose is composed of carbon atoms with hydroxyl groups (sugars). In addition, water molecules also form hydrogen bonds with each other.

- Does capillary action involve adhesion or cohesion?
  Capillary action involves both adhesion and cohesion. When water molecules are attracted to the xylem (as drawn on the board), adhesion is involved. When the water molecules are attracted to each other, cohesion is involved.

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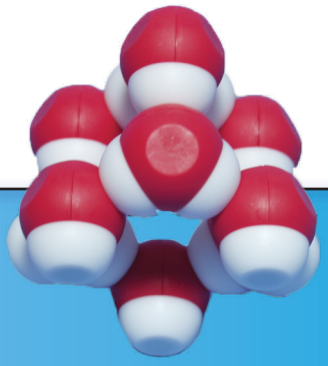


Surface Tension

General Information

Surface tension is due to the cohesion between molecules at the surface of a liquid. In a liquid, molecules are pulled in all directions by intermolecular forces. At the surface of a liquid, the molecules are only pulled downward and toward the sides. Surface tension is the amount of energy required to stretch or increase the surface of a liquid by a unit of measure.

Water Kit©

Water molecules are polar and have a relatively strong attraction to each other, called hydrogen bonds. This attraction (explained in the section labeled Hydrogen Bonding) is responsible for surface tension. Surface tension explains why water forms rounded drops on nonpolar surfaces like the waxy surface of an apple or a recently waxed car.

Activity

Have your students place drops of water on a variety of substances to test surface tension. Drops can be placed on paper, wax paper (or draw a solid rectangle with crayon on paper to simulate wax paper), paper towels, glass, plastic, fabric, plant leaves, etc. Have your students record the substance and draw the relative height of the drop.

Office Paper

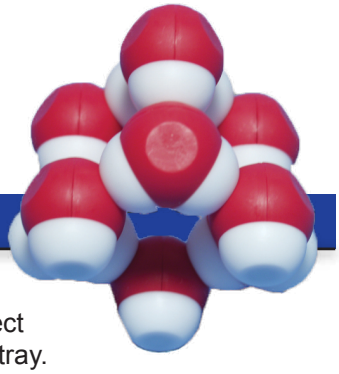
Wax Paper

Paper Towel

- Why does water bead up on some substances, like wax, and soak into others, like paper?
  Water does not interact with nonpolar substances. Wax is a nonpolar substance; the water molecules are only being pulled toward each other, forming a sphere. Office paper has a coating which is more polar than wax, but less polar than paper towels. This results in a dome-shaped drop. Paper towels are more polar than office paper. Paper towels actually break the surface tension of water to absorb it.

- Can anything walk on water?
  Yes, some insects, like the water strider, can walk on water.

- Can you explain how an insect does this?
  The hydrogen bonds are strong enough that certain insects can't break them and therefore can walk on the water molecules.



Surface Tension

Have your students pool 3D water molecule models together on a tray. Place a small toy insect on top and have your students gently shake the tray. The insect will stay on the molecules. Explain that this is surface tension.

- Why can't all animals walk on water?
  Their total mass is sufficient to break the intermolecular force of the hydrogen bonds between water molecules and they sink.

- What would happen if an insect tried to walk on a nonpolar liquid (molecules that do not have charges)? Record your hypothesis.

Pool several ethane molecules on a tray. Place a plastic insect on top and gently shake the tray. The insect will fall off.

- Why did the insect fall through the surface of the nonpolar molecules?
  There are no hydrogen bonds (cohesion) between the nonpolar molecules, therefore the insect falls between the molecules. While there are weak intermolecular forces between the ethane molecules, they are not strong enough to support the insect.

- Can you predict what would happen if an insect tried to walk on a slightly polar liquid?
  Have your students change the ethane to ethanol and repeat the activity. Results will vary with the weight of the insect and the number of hydrogen bonds formed by the ethanol. Now, have your students predict what will happen with actual liquids. Give each group a glass of water and one of vegetable oil representing 3D water molecule models and ethane, respectively. In addition, give each group a fork and several small metal paperclips.

- Predict what will happen when you lay a paperclip on top of each surface. Why? Record your hypothesis and reasons.
  Have students carefully place a small metal paper clip on the surface of a glass of water. Note: Students may have to practice this several times before they are successful. Using a fork to place the paperclip on the water can help. Paperclips should be dried thoroughly before trying to place them on a liquid again.

- What is keeping the paperclip on top of the water? Is it floating?
  No, the paperclip is not floating. The paperclip is resting on top of the water because the surface tension is stronger than the weight of the paperclip.

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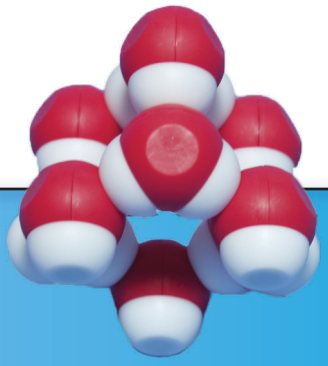


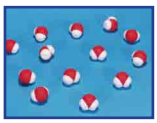
States of Water

General Information

Like other compounds, water can convert between solid, liquid, and gas phases. The remarkable property of water is that it is a liquid over a very large temperature range.

Water Kit©

3D water molecule models can be used to **demonstrate** the different phases of water as shown below.



Gas – 12 molecules separated. In the gas state individual water molecules are moving too fast and are too far apart to form hydrogen bonds.

Liquid – 12 molecules clumped together. In liquid water, the hydrogen bonds between water molecules are very short-lived. They are constantly forming, breaking, and reforming between other molecules.

Solid – 12 molecules connected in a repeating pattern or formation that form ice. In ice, the hydrogen bonds between water molecules are more stable and longer-lived.

Scientists have described 12 different structures of ice, many of which can be constructed with the Water Kit©. See pages 23 and 24 for directions on making hexagonal ice.

Activity

Demonstrate the three states of water as shown above.

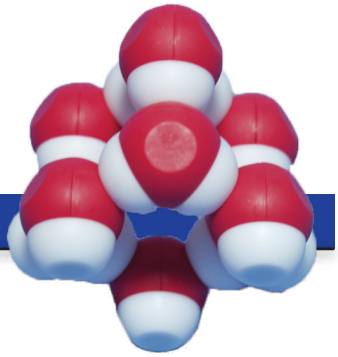
Give each group of students twelve 3D water molecule models. Challenge them to form an ice cube, an enclosed, stable ice lattice. After a few minutes, give students the hint that snowflakes, or ice crystals have six sides – they may want to try rings of six.

If your students are have trouble forming ice, print pages 23 and 24 for each group.

Display and discuss twelve 3D water molecule models in the gas, liquid, and solid states.

- What are the differences between these groups of molecules? How does their volume differ?
  Water vapor, the gaseous state, has the greatest volume. The water vapor condenses to form liquid water. Then the water molecules expand to form the solid form of water, ice.

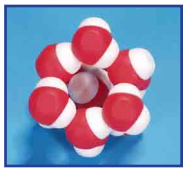
- Are these volume changes unique to water?
  All gases have greater volumes than liquids. Water, H_2O, is the only compound that expands as it freezes.



States of Water

Have your students discover how water expands as it freezes. They should form a lattice with 12 molecules and place it in a clear container, such as a bird feeder tube. Have your students measure and record the volume of water in this solid state. Next, they should gently push down on the molecules to break the hydrogen bonds until all the molecules are at the bottom of the container. Have you students measure and record the volume of the molecules in this liquid state.

Demonstrate that a 13th molecule can fit in the middle of an ice cube. Explain that when water freezes, the empty space, or void between the molecules increases.



Pool the ethane and ethanol molecules in separate containers and show them to your students.

- At room temperature, which substance is a gas and which substance is a liquid? Why?
  Ethane is a gas at room temperature because it is a nonpolar molecule which has a weak intermolecular force. It does not have a partial charge that would attract the molecules to each other. Ethanol is a liquid at room temperature because it is a polar molecule. The OH has a stronger intermolecular force, called hydrogen bonding, which attracts the molecules to each other.

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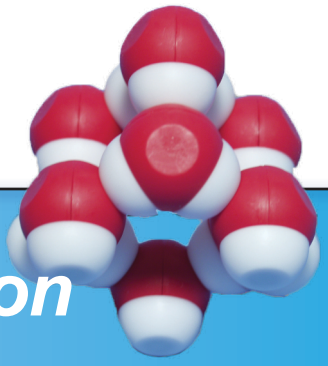


Evaporation & Condensation

General Information

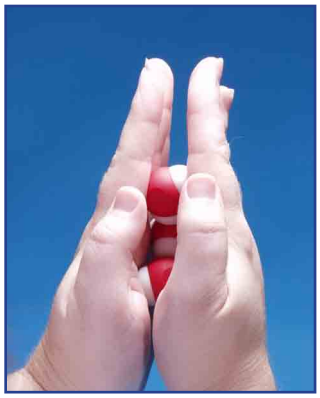
Evaporation is the transformation from a liquid to a gas. This occurs when a liquid absorbs enough heat, increasing the movement of the molecules, to cause the intermolecular forces between molecules to break the surface tension. This allows individual molecules to escape into the air. As water evaporates, the heat it absorbs breaks the hydrogen bonds and the liquid water becomes water vapor.

Condensation is the transformation from a gas to a liquid. This occurs when a gas is cooled enough that the molecules slow down to form stronger intermolecular forces with each other. As water condenses, the molecules slow down and form hydrogen bonds with each other as the water vapor cools.

Changes in pressure can also influence evaporation and condensation. As pressure increases, gases condense into liquids. As pressure decreases, liquids evaporate as gasses

Activity

- Have you ever tried to walk barefoot on hot sand, or black pavement in the summer? What was it like? What did you do?
  Tried to walk faster, keep my feet off the ground, find a cooler place to walk, put on shoes…



- When water molecules heat up they move faster as well. What happens when you heat, or boil, water?
  It evaporates.

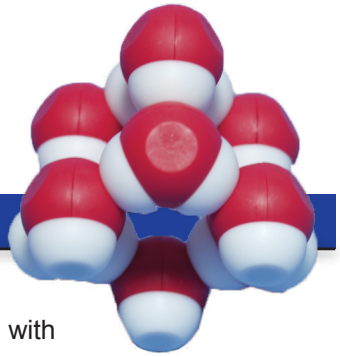
Have your students rub their hands together to create friction.

- What are you producing with your hands?
  Heat and/or energy.

Give each student three 3D water molecule models to hold between their hands. Have your students slowly move their hands together.

- What is happening to the water molecules (and the hydrogen bonds)?
  The molecules are moving around and slowly forming and breaking hydrogen bonds with each other.

This represents water in its liquid form. Have your students slowly increase the speed (heat) of their hands.



- What is happening to the water molecules now?
  The water molecules are moving fast and are not forming hydrogen bonds with each other.

This represents evaporation and water in its gas state. To simulate condensation, have your students slow down their hands, drawing heat away from the molecules.

To help your students understand evaporation, have them place one drop of water and one drop of rubbing alcohol 4 cm apart on their forearms. Start a timer. Your students should observe and record findings.

- What did you observe?
  The water forms a rounded drop due to its strong surface tension, while the rubbing alcohol flattens.

The rubbing alcohol evaporates within 2 minutes. Water takes much longer.

Explain to your students that body heat provides enough energy for the rubbing alcohol molecules to move fast enough to leave the surface of your skin. It takes about 290 cal per gram of rubbing alcohol for the molecules to evaporate, whereas, it takes 580 cal per gram of water to break hydrogen bonds between water molecules (calories are measurements of energy). Your body temperature at rest is not high enough to evaporate water rapidly.

Water fact: *580 cal of heat are needed to evaporate one gram of water at room temperature.*

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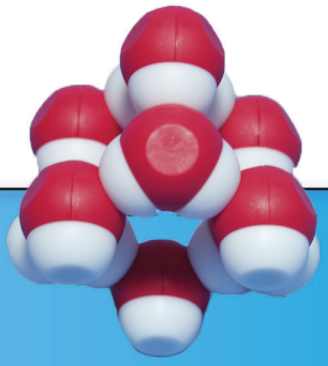


Solubility

General Information

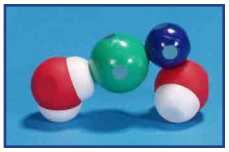
In the presence of water, salt dissolves. Water molecules always hydrate the sodium chloride.

Ions are charged atoms that have gained or lost electrons as a result of an ionic bond.

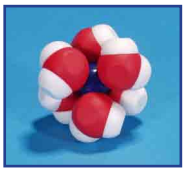
Water Kit©

The Water Kit© comes with a blue sodium (Na) ion model and a green chloride (Cl) ion model. These two ions form an ionic bond to make sodium chloride (NaCl), often called table salt.

When water dissociates sodium chloride, the hydrogen atom interacts with the negatively charged chloride ion and the oxygen atom interacts with the positively charged sodium ion.

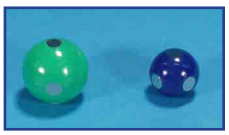


Note: The Water Kit© can demonstrate the principle behind hydration, but it cannot simulate the true chemical reaction. Many more water molecules would be needed.



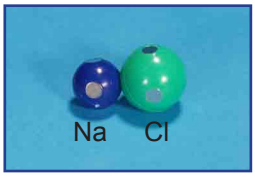
Activity

Hand out a sodium ion and a chloride ion to each group.
Have them explore what happens when they are brought together.

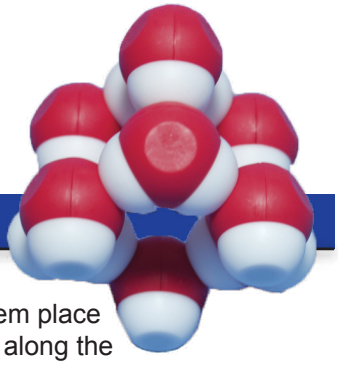


- Why did the sodium and chloride ions come together?
  Sodium has a positive (+) charge and chloride has a (-) negative charge. They form an ionic bond.

- What is the chemical formula for this? What is the compound?
  NaCl. NaCl is often called table salt.



- What happens when you put salt in a cup of water?
  It dissolves.



Solubility

Give each student group twelve 3D water molecule models and a tray. Have them place the sodium chloride in the center of the tray and the 3D water molecule models along the edges. Your students should gently shake the tray.

- Using your twelve 3D water molecule models and salt ions, can you explain what happens when you put salt in water?
  The sodium, a positively charged ion, is attracted to the oxygen of a water molecule and chloride, a negatively charged ion, is attracted to the hydrogen of the water molecule. Water molecules surround the entire sodium and chloride, breaking them apart.

- What happens when you keep adding salt to a glass of water? Why does this happen?
  Eventually, salt begins to form at the bottom of the glass when there are not enough water molecules to surround the salt.

- What is this called?
  This is a supersaturated solution.

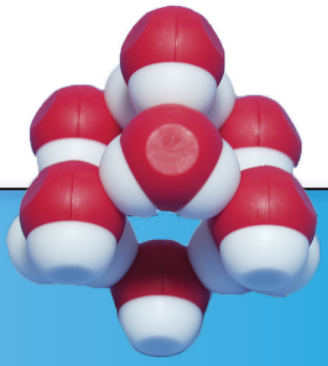
- What happens to a glass of salty water when it is heated to boiling?
  The water evaporates leaving the sodium and chloride to bond to form salt again.

Give each group of students an ethane molecule. Have them put it in the center of the tray and the 3D water molecule models along the edge. Have your students gently shake the tray.

- Will water dissolve ethane? Why or why not?
  No. Water cannot dissolve ethane because ethane is nonpolar. Water cannot form hydrogen bonds with ethane.

3-D Molecular Designs

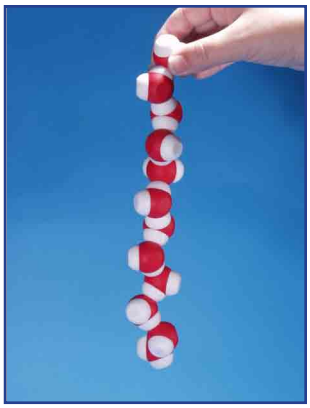
. . .where molecules become real™



Appendix

Just for Fun

The Water Kit© can be fun as well as educational! Here are some fun activities to do with your students.



Chain of Life

Have students guess how many 3D water molecule models can be strung on a chain before the chain breaks. Test their predictions.

Keep Score with Water!

Hang one 3D water molecule model from the ceiling. As each student correctly answers a water-related question, have them add a molecule to the chain. After enough students have correctly answered questions – and the chain breaks, declare No Homework!

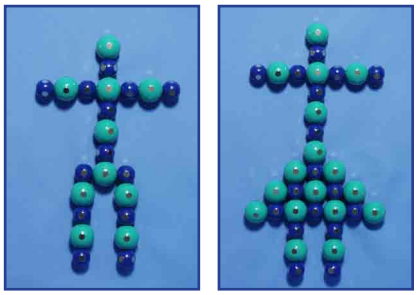
Building with Molecules

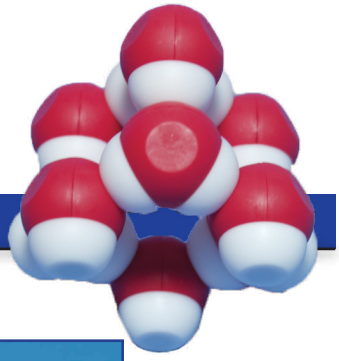
Try to build shapes with 3D water molecule models!

Snowflakes

Pyramid

NaCl

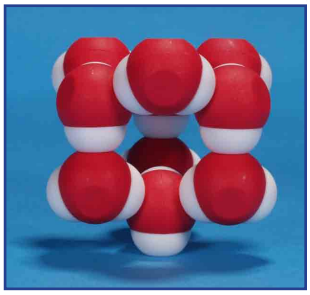




Just For Fun Activities

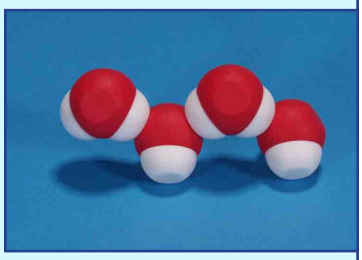
Cubic Ice

Cubic ice, (Ice 1c) is one of the twelve structures of ice identified by scientists. Cubic ice cubes can be created with as few as ten molecules, while hexagonal ice requires twelve. Check the 3D Molecular Designs website for more information on ice and how to construct some of the twelve different structures of ice. To create a Cubic Ice cube, follow the directions for either the *Step Method* or the *Pattern Method* (on the next page).

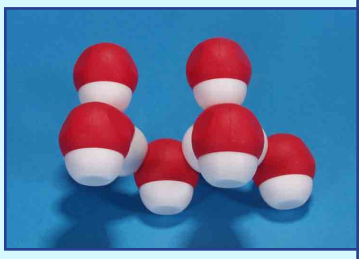


Step Method

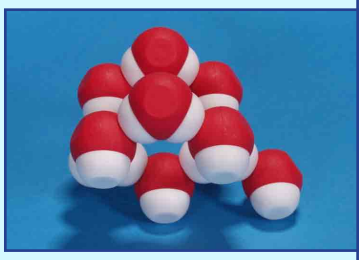
1. Create the body of a dragon as shown in the first picture.



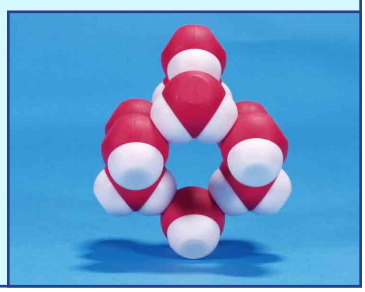
2. Add ears and wings to the dragon as shown in the second picture.
   Note: *the hydrogen atoms should point down on both the ears and wings.*

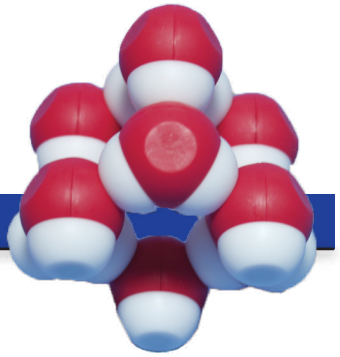


3. Connect each ear to a wing as shown in the third picture.



4. Remove the tail and place the final piece to create the cube as shown in the fourth picture.

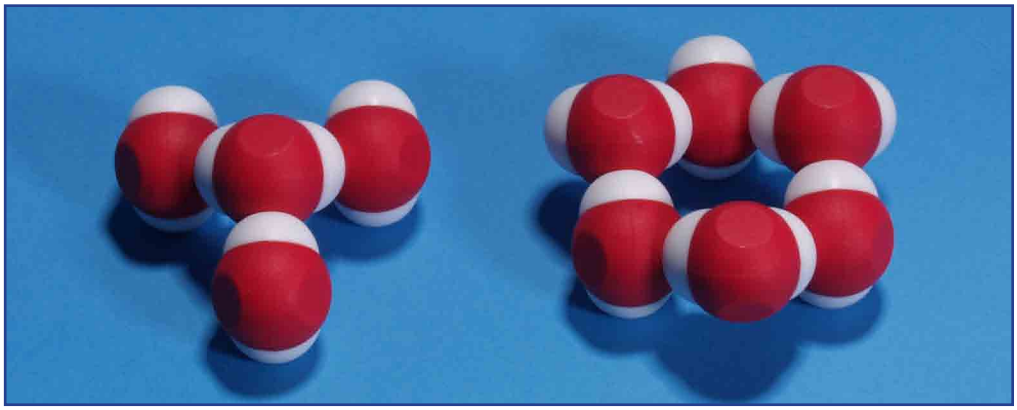


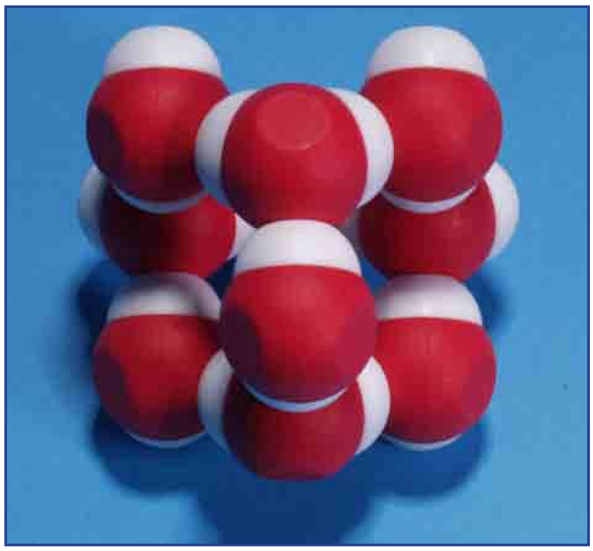


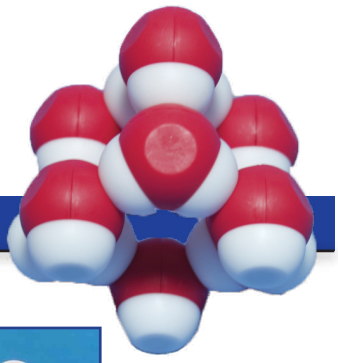
Just For Fun Activities (continued)

Pattern Method

Create a Y and a ring using the pattern below. Without rotating the Y, place it on top of the ring.



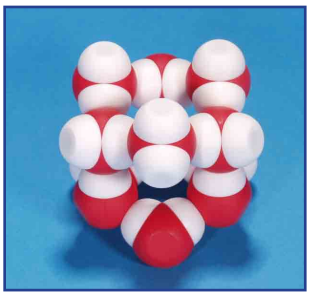




Just For Fun Activities (continued)

Hexagonal Ice

Scientists have described twelve structures of ice, many of which can be constructed with the Water Kit©. Check the 3D Molecular Designs website for more information on ice and how to construct some of the twelve different structures of ice. To construct Ice 1h, hexagonal ice, follow the directions for the *Step Method* or the *Pattern Method* (on the next page).

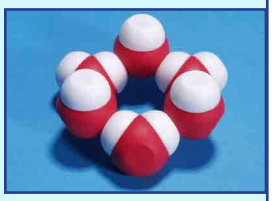


Step Method

1. Hold one molecule horizontally in front of you with the hydrogen atoms to the sides.

2. Add two vertical (hydrogen atoms pointing up and down) molecules to the two hydrogen atoms from the first step. *See the first picture on the right.*

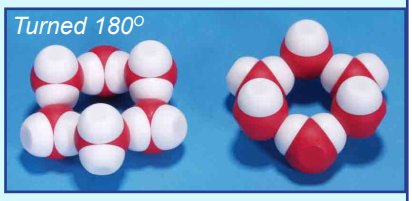
3. Add a horizontal molecule to the lower hydrogen on each molecule added in step 2. *See the 2nd picture to the right.*



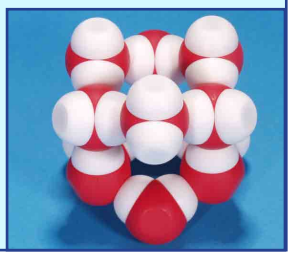
4. Add one vertical molecule to connect the hydrogen atoms from the molecules added in step 3. *See the 3rd picture to the right.*

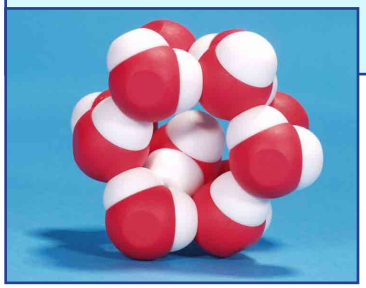
5. Create a second hexagonal ring following steps 1-4.

6. Orient the two hexagonal rings the same way – then rotate one ring 180 degrees.

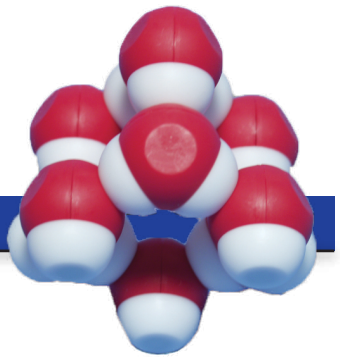


7. Place one hexagonal ring on top of the other. Do not flip one ring over – the hydrogen atoms on both rings should point the same way.





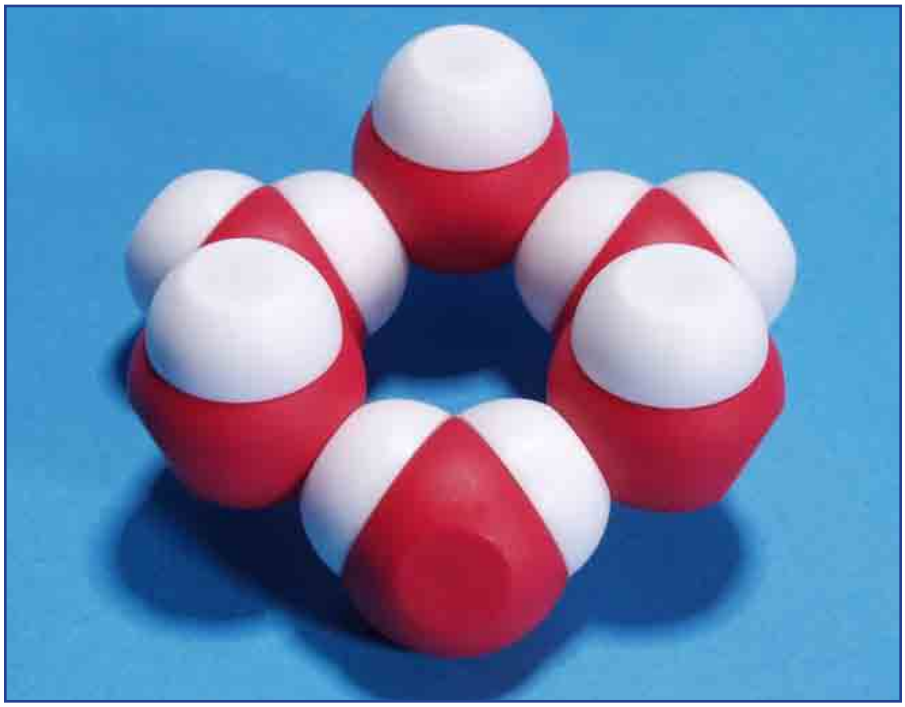
You have now formed a model of an ice cube. Note that it is possible to place a 13th water molecule into the hole formed by this lattice. Now you can see why ice floats. Ice has empty spaces in it. The same volume occupied by 12 water molecules in a solid ice lattice, can contain more water molecules in its liquid form.

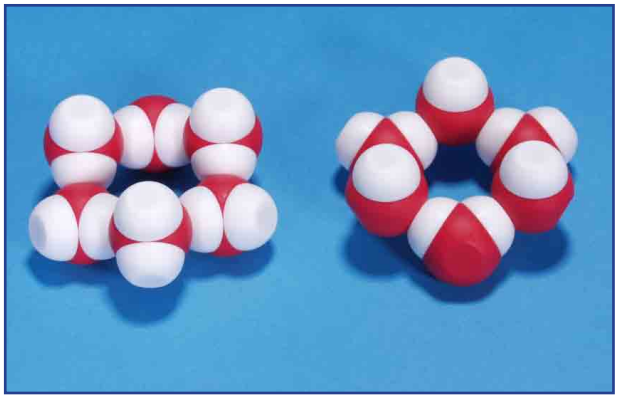


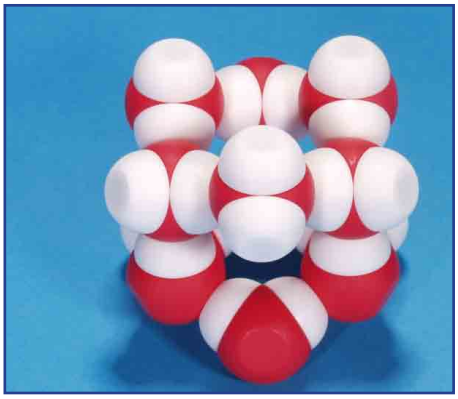
Just For Fun Activities (continued)

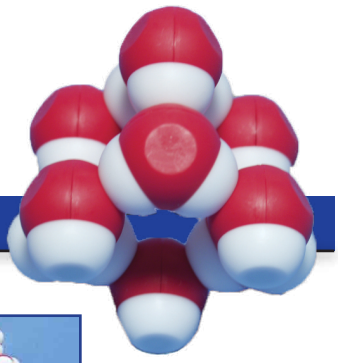
Pattern Method

Use this pattern to construct hexagonal Ice (Ice Ih). Create two rings, turn one 180 degrees, and stack one ring on top of the other.





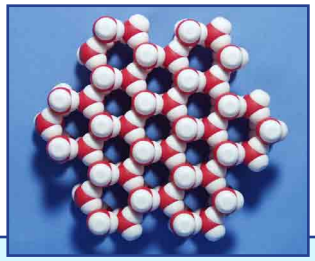




Just For Fun Activities (continued)

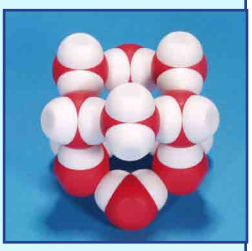
Snowflakes

Scientists have described twelve structures of ice, many of which can be constructed with the Water Kit©. To form a six-sided snowflake, you will need 6 or 7, 1-Cup Water Kits©.



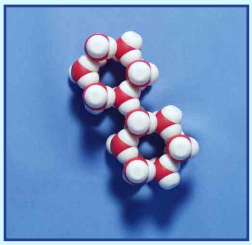
Hexagonal Snowflakes

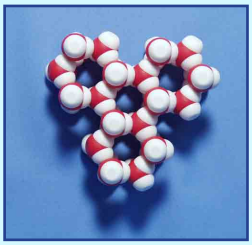
Make 6 or 7 ice cubes following the hexagonal ice instructions. Your cubes should look like the picture on the right.

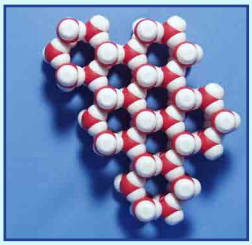


Connect your Hexagonal Ice cubes to form a hexagon. Each cube will be oriented the same way.

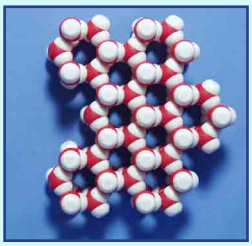
If you have problems creating a hexagon, then try to connect six cubes around a center cube.







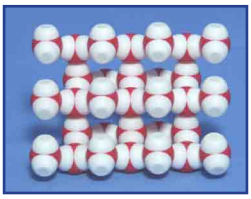
If you only have six cups, then remove the center cube and rotate it to complete the outer hexagon.

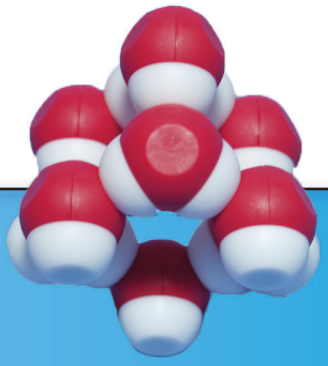


Without a center cube, which requires the seventh cup of the Water Kit©, this structure is not very strong.

Other Ice Crystals

If you do not create uniform cubes, it may be difficult to connect several cubes, and you will be unable to produce a six-sided snowflake. It is possible to create larger ice crystals by connecting several of the cubic ice cubes and some of the other twelve forms of ice described by scientists. Information on constructing other forms of ice will be posted on the 3D Molecular Designs website as they become available.





Glossary

Adhesion is the attraction between two unlike molecules.

Capillary action is the spontaneous rising of a liquid in a capillary (small diameter) tube.

Cohesion is the attraction between two like molecules.

Condensation is the transformation from a gas to a liquid. This occurs when a gas is cooled enough that the molecules slow down to form stronger intermolecular forces with each other. As water condenses, the molecules slow down and form hydrogen bonds with each other as the water vapor cools.

Covalent bonds are formed when two atoms share two electrons. A covalent bond is an intramolecular bond within one molecule. Covalent bonds can be either polar (which have partially charged atoms) or nonpolar (without charged atoms).

Evaporation is the transformation from a liquid to a gas. This occurs when a liquid absorbs enough heat, increasing the movement of the molecules, to cause the intermolecular forces between molecules to break the surface tension. This allows individual molecules to escape into the air. As water evaporates, the heat it absorbs breaks the hydrogen bonds and the liquid water becomes water vapor.

Hydrogen bonds are an intermolecular force between the two molecules where a positively charged hydrogen atom interacts with a negatively charged fluorine, nitrogen or oxygen atom in a second molecule.

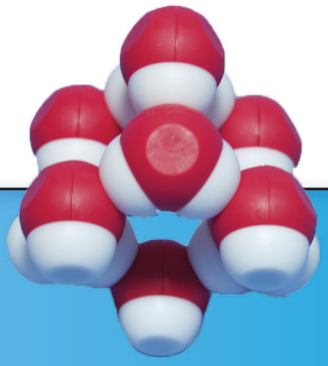
Ionic bonds are the complete transfer of an electron between two atoms resulting in one positively and one negatively charge atom. Ionic bonds are intramolecular bonds within one molecule.

Ions are charged atoms that have gained or lost electrons as a result of an ionic bond.

Nonpolar molecules do not have unequal regions of charges and therefore do not interact with polar molecules. Oil and ethane are examples of a liquid and a gas composed of nonpolar molecules.

Polar molecules have partial charges. Polarity refers to the partial positive charge (+) and partial negative charge (-) that a molecule has when electrons are unequally shared between two or more atoms. Water molecules are polar molecules.

Surface tension is the amount of energy required to stretch or increase the surface of a liquid by a unit of measure. Surface tension is due to the cohesion between molecules at the surface of a liquid. In a liquid, molecules are pulled in all directions by intermolecular forces. At the surface of a liquid, the molecules are only pulled downward and toward the sides.



National Standards

Connections to: A Framework for K-12 Science Education

Practices, Crosscutting Concepts, and Core Ideas*

Dimension 1: Scientific and Engineering Practices

1. Asking questions (for science) and defining problems (for engineering)
2. Developing and using models
5. Using mathematics and computational thinking

Dimension 2: Cross Cutting Concepts

1. Patterns
2. Cause and effect: Mechanism and explanation
3. Scale, proportion, and quantity
4. Systems and system models
5. Energy and matter: Flows, cycles, and conservation
6. Structure and function
7. Stability and change

Dimension 3: Disciplinary Core Ideas:

Physical Sciences

PS1: Matter and Its Interactions
PS1.A: Structure and Properties of Matter
PS1.B: Chemical Reactions
PS2: Motion and Stability: Forces and Interactions
PS2.A: Forces and Motion
PS2.B: Types of Interactions
PS2.C: Stability and Instability in Physical Systems

Life Sciences

LS1: From Molecules to Organisms: Structures and Processes
LS1.A: Structure and Function
LS2: Ecosystems: Interactions, Energy, and Dynamics
LS2.A: Interdependent Relationships in Ecosystems
LS2.B: Cycles of Matter and Energy Transfer in Ecosystems
LS2.C: Ecosystem Dynamics, Functioning, and Resilience

Earth and Space Sciences

ESS2: Earth's Systems
ESS2.A: Earth Materials and Systems
ESS2.C: The Roles of Water in Earth's Surface Processes
ESS2.D: Weather and Climate
ESS3: Earth and Human Activity
ESS3.A: Natural Resources
ESS3.D: Global Climate Change

**The NSTA Reader's Guide to A Framework for K-12 Science Education, National Research Council (NRC), 2011. A Framework for K-12 Science Education: Practices, Crosscutting Concepts, and Core Ideas. Washington, D.C.: National Academies Press.*

The Magic Disk Trick

--- or ---

A Macroscopic Demonstration of Hydrophobic vs. Hydrophilic Interactions.

We are always looking for ways to connect the macroscopic world in which kids live with the invisible world of molecules. For example, how can we help them construct an understanding of how hydrophobic and hydrophilic forces play a major role in determining the folded structure of a protein? Here is an activity that does just this.

Materials:

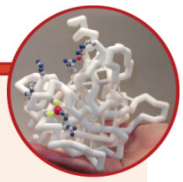
1. Hexane (~10 ml) (Lighter Fluid works as an alternative)
2. Water (~10 ml)
3. Paper….the cheap stuff, not shiny /glossy paper.
4. A number 2 graphite pencil
5. A capped test tube or vial (plastic or glass) that will hold ~20 ml of liquid.

Procedure:

1. Take the raw paper --- which is made of cellulose, and is therefore hydrophilic --- and scribble on one side with the # 2 pencil. This will coat one side of the paper with carbon --- which is hydrophobic.
2. Using a single hole punch, punch out 5 disks. One side of each disk is black --- and hydrophobic (graphite is pure carbon), while the other side is white --- and hydrophilic (paper is made from cellulose, which has a lot of OH groups).
3. Add both the water and the hexane to the test tube or vial. Cap and shake vigorously. These two solvents will quickly separate into two distinct phases, with the less dense hexane on top.
4. Add the bi-colored disks to the vial, cap and shake.
5. Observe how the disks orient themselves at the interface between the hydrophobic hexane and the hydrophilic water.

What can go wrong? Almost nothing, except:

1. You can use glossy paper….that has been treated with clay and other materials that decrease the hydrophilic nature of the cellulose surface.
2. You can scribble only very lightly, and therefore not completely cover the hydrophilic cellulose with hydrophobic carbon.
3. You can use a dirty or soapy test tube/vial….that will confound the disks.
4. You can handle the paper with oily/greasy fingers --- in which case the hydrophilic cellulose will become somewhat hydrophobic, confounding the disks.
5. You can fail to test all your reagents in advance, in which case this experiment is guaranteed …. not to work.



Membrane Starter Kit

Student Copy

A Message to Teachers

Thank you for using our tools to help your students visualize the molecular world! The lesson/activity guide that accompanies the plasma membrane kit is intended to help you consider different ways in which you may use these materials. It is not the intent of the Center for BioMolecular Modeling to require you to work through the entire lesson from start to finish (although you may do so if you wish). We do **encourage** (and perhaps insist on) your **modification** of these lessons and activities to meet the learning objectives of your specific students or to accommodate the physical limitations of the environment in which you teach. Enjoy!

Objectives: You will use the model pieces in the kit to:

- Examine the general amphipathic structure of a phospholipid.
- Compare and contrast various models of phospholipids.
- Explore the interaction between phospholipids and water.
- Construct a phospholipid monolayer, micelle and bilayer and relate it to plasma membrane structure.
- Identify and demonstrate the function of the various types of channel proteins involved in membrane transport.

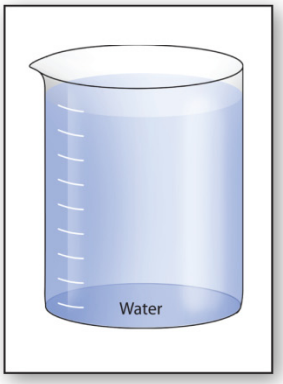
Introduction:

The **plasma membrane** is the structural boundary that separates the cell from its surroundings and controls what substances move into and out of the cell it surrounds. As only some substances are allowed to cross the membrane, the plasma membrane demonstrates the property of **selective permeability**. In particular, the plasma membrane of mammalian red blood cells (erythrocytes)

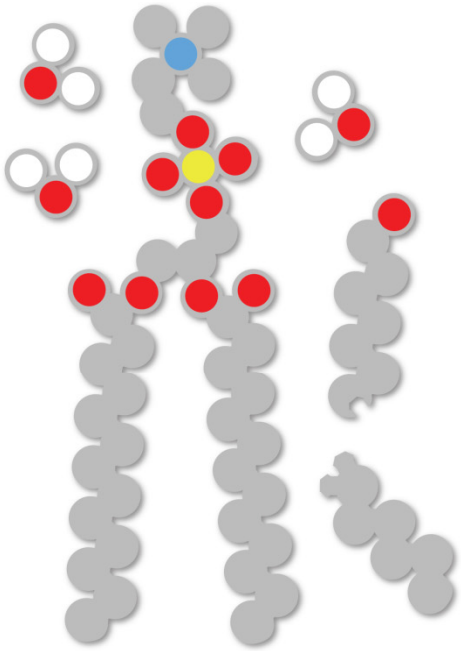
has been the focus of cell membrane study because these cells do not contain nuclei or internal membranes. As such, they represent a source from which a pure plasma membrane may be easily isolated for analysis. In 1925, the two Dutch scientists Evert Gorter and his research assistant F. Grendel extracted lipids from the membranes of a known number of red blood cells which corresponded to a known surface area of plasma membrane. The surface area occupied by a monolayer of the extracted lipid spread out at an air/water interface was determined. The results of their experiment showed that the surface area of the lipid monolayer was twice that occupied by the erythrocyte plasma membrane, leading to the conclusion that the plasma membrane consists of **lipid bilayers**. The most abundant lipids in most membranes are **phospholipids**. The ability of phospholipids to spontaneously form membranes is inherent to their **amphipathic** (meaning they have both a hydrophilic region and a hydrophobic region) nature.

Materials Needed:

Bag of Mini-Phospholipid Foam Models



Beaker Diagram



Large Foam Phospholipids

Large Molymod Phospholipids (optional)

Part 1: Focus on Phospholipids

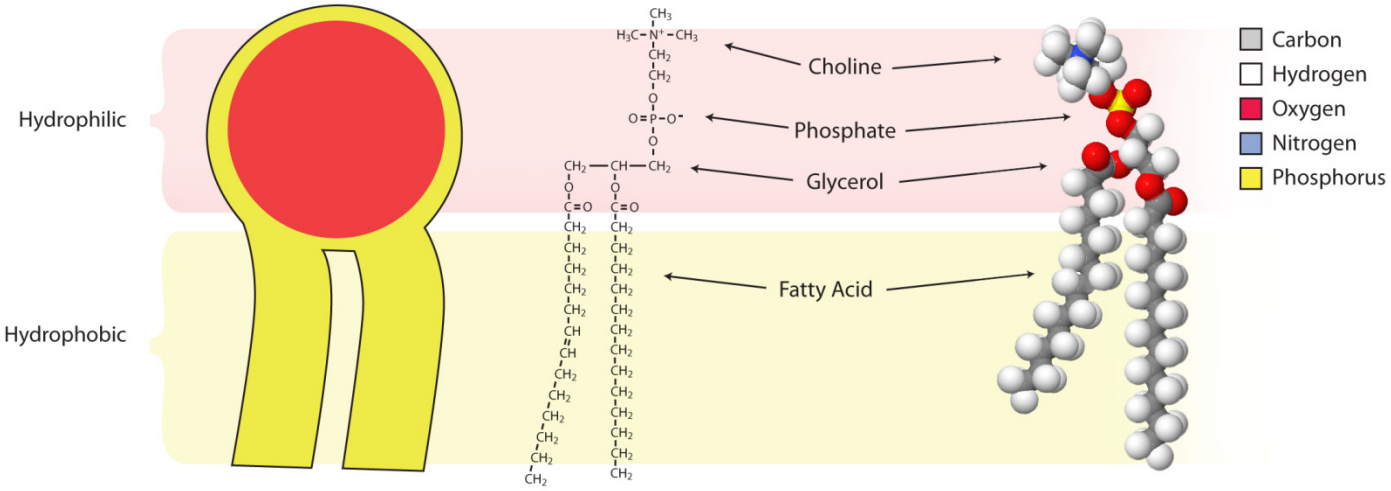
1a. Sketch the specific structural formal of the model you have been asked to examine in the space provide below. Label the hydrophilic and hydrophobic regions of your structure.

1b. Compare your structure to that of the other groups in the room. Record any similarities you observe in these phospholipid structures.

1c. Based on these similarities a simplified representation may also be used to indicate phospholipid structure. Sketch this simplified model in the space below. Label the hydrophobic and hydrophilic portions of this simplified model.

1d. Record any differences in the specific structures you have observed between these phospholipids.

The general structure of a phospholipid is most often represented by the phosphatidylcholine structure:



Part 2: The Kessler Membrane Activity

2a. Obtain ten of the simplified representations of a phospholipid. Using the diagram of the beaker of water, arrange the ten phospholipids demonstrating correct hydrophobic/hydrophilic interactions. Sketch your result below.

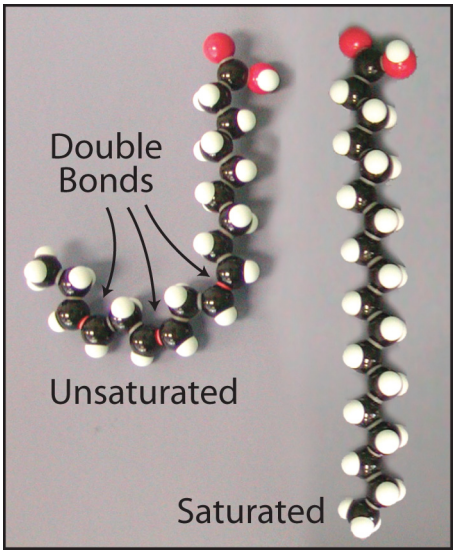
2b. Using the same ten phospholipids, rearrange them in the beaker so that they are submerged in the water while still maintaining the correct hydrophobic/hydrophilic interactions. Sketch your result below.

The resulting structure is referred to as a **micelle.** Micelles can act as emulsifiers that will allow a compound that is normally insoluble (in the solvent being used) to dissolve.

2c. Your next challenge is to construct a structure that is BOTH submerged in the water AND contains water on the inside. You may use as many of the phospholipids in your kit as you wish to complete the task. Sketch the resulting structure below.

2d. Based on the structure of a phospholipid, explain the reasoning for your arrangement.

The resulting structure is referred to as a **liposome**. Liposomes are artificially prepared vesicles that may be used as a vehicle for administration of nutrients or pharmaceutical drugs.

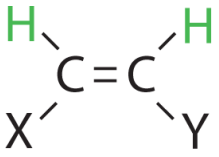


Upon closer examination of the hydrophobic tails, you may notice that in one of the tails, all of the carbons are connected with single bonds. These hydrocarbon tails are referred to as **saturated**. In the second tail there is a red connector, indicating that a double bond connects these two adjacent carbons. The presence of a double bond creates a "kink" in the structure of this hydrocarbon chain. The hydrocarbon tails containing a double bond or bonds between adjacent carbons are referred to as **unsaturated**.

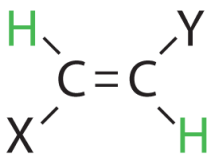
2e. Develop an explanation for the necessity of having "kinks" in the hydrophobic tail of the phospholipids that make up cell membranes.

Note to Teachers - Optional Activity:

Further comparison shows a difference in these double bonds. If the hydrogens associated with the double bonded carbons are on the same side, the configuration is said to be "cis". If the hydrogens associated with the double bonded carbons are on opposite sides, the configuration is referred to as "trans". (See illustrations below.) Teachers may also opt to include a discussion of "trans fats" using these models.



cis



trans

2f. Which configuration produces the bigger “kink” in the structure of the hydrophobic tail?

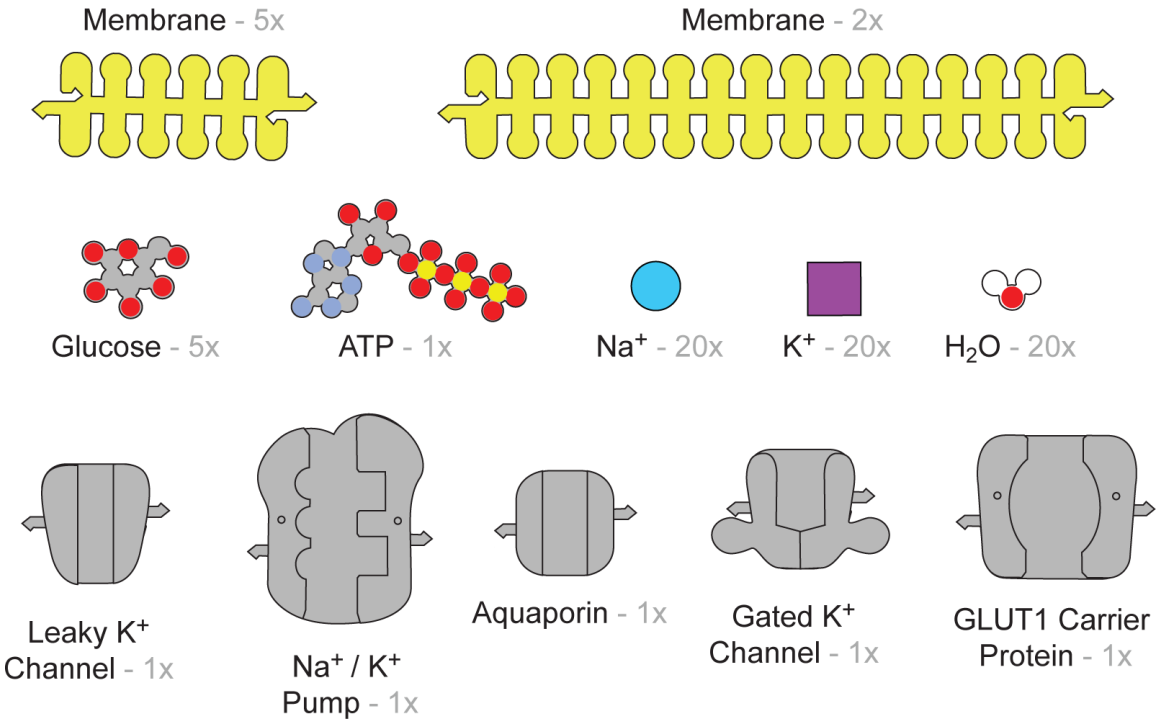
2g. How might this configuration contribute to the fluidity of the membrane?

Part 3: Exploring Membrane Permeability

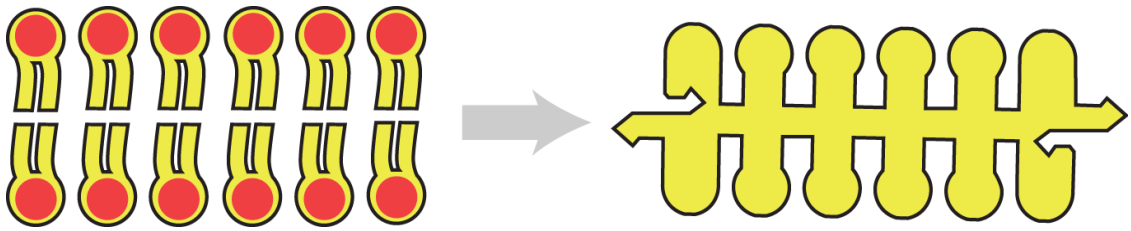
Introduction:

The phospholipid bilayer is only one aspect of the gatekeeper system responsible for the plasma membrane’s **selective permeability**. Membrane bound proteins play a key role in regulating the transport of ions and molecules through the plasma membrane.

Materials:



We will use a slightly different and simplified representation of the phospholipid bilayer in the next activity.



3a. Label the hydrophilic head and hydrophobic tail in the model pictured above.

3b. Nonpolar molecules, such as hydrocarbons, CO_2 and O_2 are hydrophobic. Explain why these molecules can easily cross the plasma membrane without the aid of proteins.

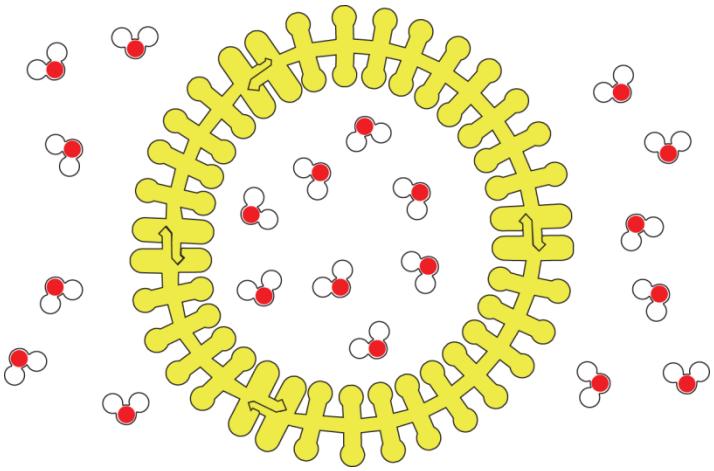
A variety of polar molecules and can't move through the plasma membrane on their own. Contact with hydrophobic lipid bilayer may be avoided by these hydrophilic substances as they cross the plasma membrane with the help of **transport proteins**.

Part 4: Channel Proteins

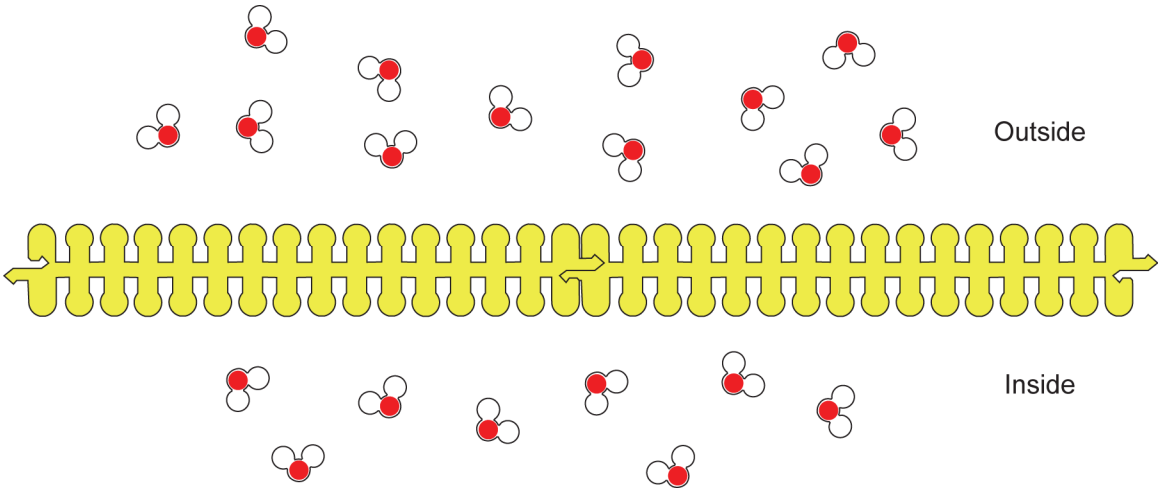
Some transport proteins, referred to as **channel proteins**, function by having a hydrophilic channel that certain molecules or ions use to cross the plasma membrane. There is a common misconception that a polar water molecule can easily cross the hydrophobic bilayer of the plasma membrane. Simple diffusion of water across the membrane does not occur at a rate fast enough in order to meet the survival needs of a cell. Channel proteins known as **aquaporins** allow entry of up to three billion (3×10^9) water molecules per cell per second!

4a. Construct a physical representation (a model) of the passage of water through a plasma membrane.

Use two short pieces and two long pieces of the simplified phospholipid membrane. Connect the pieces to form a "cell". We suggest placing 8 of the water molecules inside the cell (Intracellular) and 12 water molecules outside the cell (extracellular).

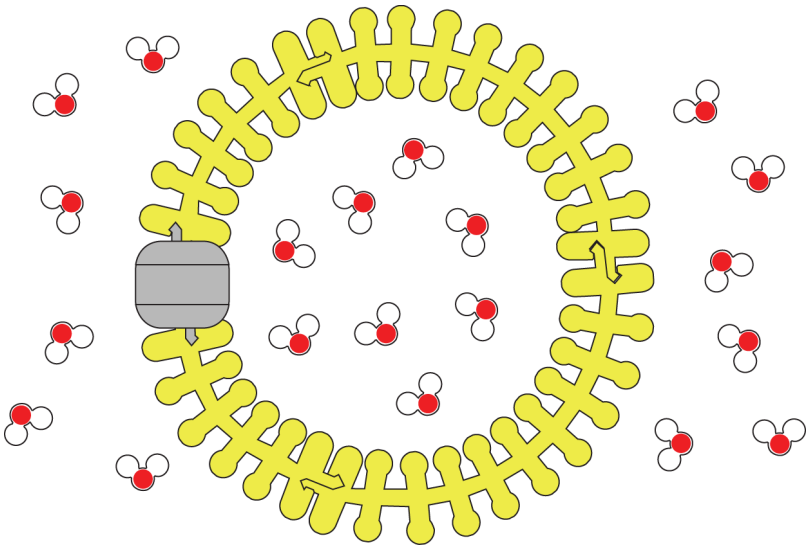


Note to Teachers - You may opt to set up the membrane in a linear fashion (shown below) if more conducive to your teaching environment.



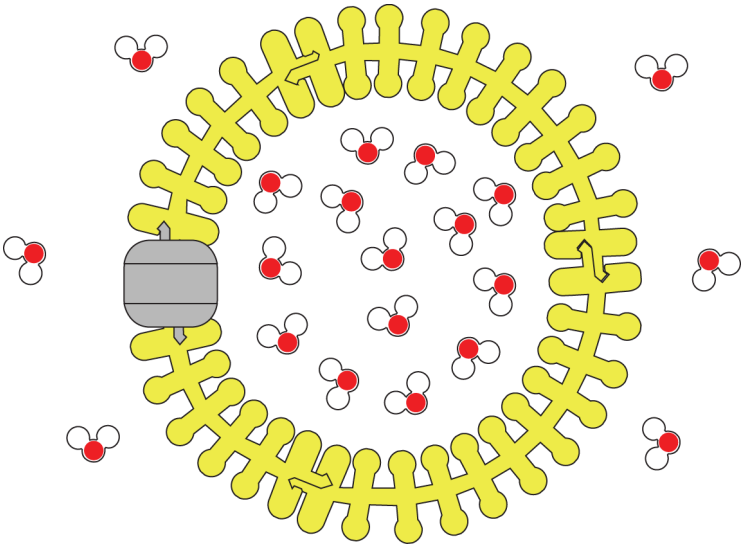
4b. Explain why water would have a difficult time diffusing across the cell membrane. Keep in mind the structure of water in your answer.

A substance will generally diffuse from where it is more concentrated to where it is less concentrated. In other words, the substance will diffuse down its **concentration gradient**. Insert a channel protein model, in this case representing **aquaporin**, into the cell membrane you have constructed. Aquaporin is an example of a channel protein found in the plasma membrane which transports water (see diagram at right).



4c. Move the water molecules to indicate the net flow of water in this system. What direction did the water molecules move?

4d. Construct a system where intracellular water molecule concentration is higher than extracellular water molecule concentration. Sketch your model in the space below and indicate the net flow of water.



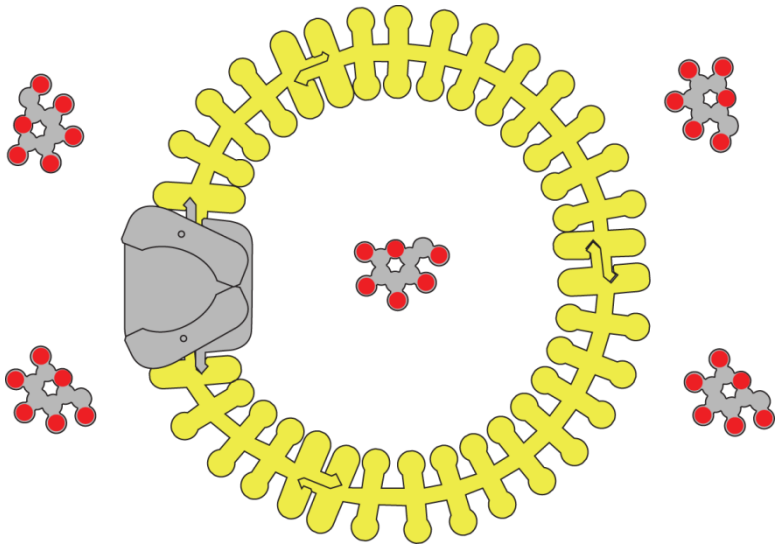
Note to Teachers -
You may choose to use the kit to introduce the students to the terms hypertonic, hypotonic and isotonic at this time.

4e. Predict what will happen to the cell due to the movement of the water.

In order for water move across the cell membrane in the above cases, the cell did **not** have to expend cellular energy (usually in the form of ATP) to move water across the plasma. Movement

of a substance across a membrane without the expenditure of energy is referred to as **passive transport**. Likewise, when the cell must expend energy to move a substance against its concentration gradient the process is referred to as **active transport**. Additionally, the channel protein aquaporin assisted in passively moving water across the membrane. This occurrence may be referred to as **facilitated diffusion**.

Part 5: Carrier Proteins



Remove the water molecules and aquaporin representation from the model cell you have constructed. Insert the carrier protein model into the plasma membrane model. Distribute the glucose molecules so that there are more extracellular glucose molecules than intracellular glucose molecules (see diagram).

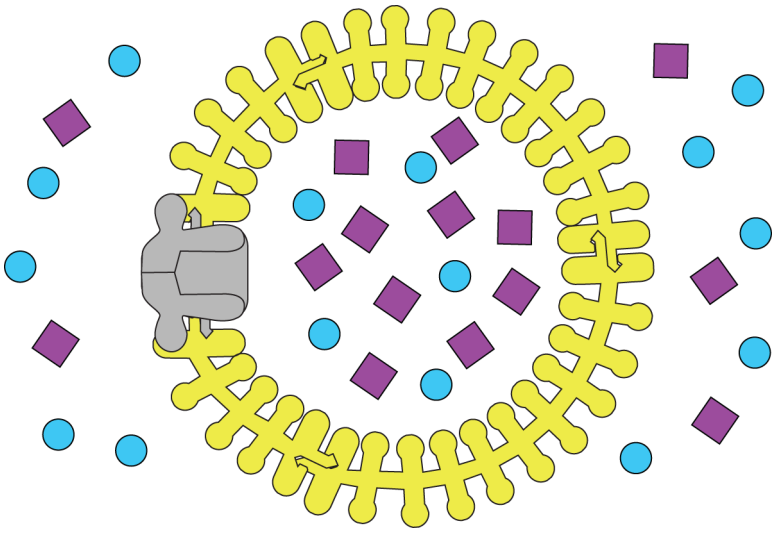
A **carrier protein** binds a solute molecule on one side of the membrane, undergoes a shape change (conformational change) and deposits the solute molecule on the other side of the membrane. GLUT 1 is an example of a protein channel frequently found in the plasma membrane of red blood cells that facilitates the movement of glucose across the cell's plasma membrane.

5a. Use the model to demonstrate the movement of glucose across the cell membrane. Sketch your model in the space below.

5b. Is this an example of passive or active transport? Explain your choice.

Part 6: Gated Channels

Remove the glucose molecules and carrier protein from the model cell you have constructed. Insert the gated channel protein model into the plasma membrane model (see diagram). Place 5 sodium ions (**round**) and 10 potassium ions (**square**) inside of the cell to simulate the intracellular environment ion concentrations. Place 10 sodium ions and 5 potassium ions outside of the cell to simulate the extracellular environment ion concentrations.



Gated channels are channel proteins that open or close in response to a stimulus. In nerve cells, a stimulus opens the gated sodium channel to allow a stream of sodium ions to enter the cell. Other gated channels open or close when a substance, different from the one to be transported, binds to the channel.

Begin the simulation with the sodium gated channel closed. After a stimulus the gates swing open.

6a. Based on what you know about concentration gradients, what direction will the sodium ions move through the channel?

6b. Does this gated channel demonstrate passive or active transport? Explain your answer.

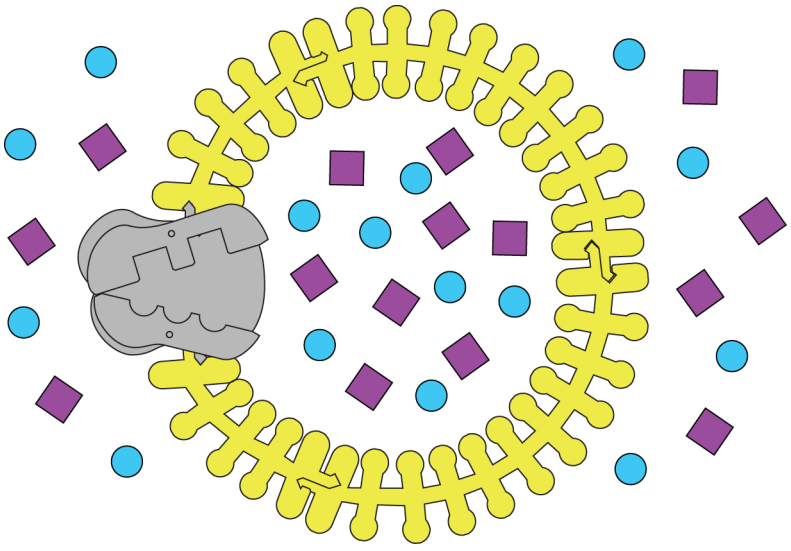
6c. Why may sodium ions move through the sodium channel while potassium ions typically do not?

6d. Speculate what other stimului may affect the operation of channel proteins.

6e. Devise a question you might have about the operation of this channel protein.

Part 7: Active Transport - The Sodium-Potassium Pump

Remove the gated channel from the model cell you have constructed. Instead, insert the sodium-potassium pump protein into the membrane of the model. (See diagram below.) Place 7 sodium ions (**round**) and 8 potassium ions (**square**) inside of the cell to simulate the intracellular environment ion concentrations. Place 8 sodium ions and 7 potassium ions outside of the cell to simulate the extracellular ion concentrations.



Transport proteins that move solutes against their concentration gradients are all carrier proteins. The sodium-potassium pump is a special carrier protein that moves sodium ions against their gradient OUT of the cell and potassium ions against their gradient IN to the cell.

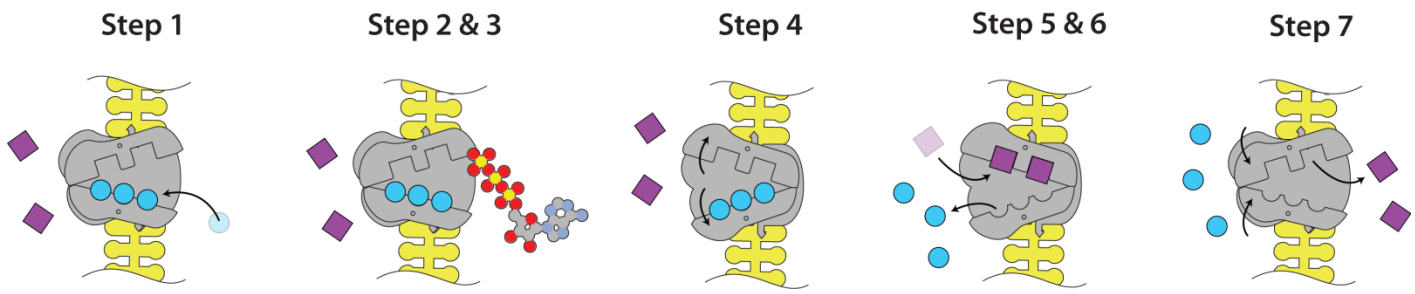
Because these ions are moving against their concentration gradients, the cell must expend energy to do the work resulting in **active transport** of these ions.

A typical animal cell has a much higher concentration of potassium ions (K^+) and a much lower concentration of sodium ions (Na^+) on the inside of the cell than the outside. The sodium-potassium pump uses energy in the form of ATP to move these ions against their concentration gradients and establish the "normal" intracellular ion concentrations. We will demonstrate the action of the sodium-potassium pump in the following simulation:

Set the sodium-potassium pump so that it is open to the inside of the cell as shown in the diagram on the previous page (page 14).

Record the initial ion concentrations in the table provided below:

Ion Types	Initial Amount	Amount After First Cycle	Amount After Second Cycle
Intracellular Na^+			
Intracellular K^+			
Extracellular Na^+			
Extracellular K^+			



Step 1: Bind three three intracellular sodium ions to the appropriate spots in the protein.

Step 2: Bring the ATP in close proximity to the pump.

Step 3: Sodium ion binding stimulates phosphorylation of the pump protein by ATP. In other words, a phosphate group is added to the sodium-potassium pump from the ATP molecule. (You will not be able to demonstrate this step with the model).

Step 4: Phosphorylation causes a change in the shape of the protein. You can demonstrate this by "swinging" the sides of the protein so that it opens to the outside of the cell.

Step 5: The shape change reduces the protein's binding affinity for sodium ions and increases the binding affinity for potassium ions. Remove the sodium ions from the protein and deposit them outside the cell and bind two potassium ions to the appropriate spots in the protein.

Step 6: Potassium ion binding triggers the release of the phosphate group from the protein. (Again, you will not be able to demonstrate this step with the model).

Step 7: Loss of the phosphate group results in the restoration of the protein's original shape which then releases the potassium ions. Swing the sides of the protein back so that they open to the inside of the cell and deposit the potassium ions.

Step 8: Repeat this process one more time.

Record the ion concentrations after completing the first cycle of the action of the sodium-potassium pump.

7a. What is the initial overall positive charge inside the cell compared to the outside the cell?

7b. Compare the total intracellular positive charge to the total extracellular positive charge after one cycle of the sodium-potassium pump.

Record the ion concentrations after completing the second cycle of the action of the sodium-potassium pump. Compare the total intracellular positive charge to the total extracellular positive charge after the second cycle of the action of the sodium-potassium pump.

7c. Where is the sodium ion concentration highest at the beginning of the sodium-potassium pump cycle?

7d. Where is the potassium ion concentration highest at the beginning of the sodium-potassium pump cycle?'

7e. What is the initial overall charge of the inside of the cell compared to the outside?

7f. Why is ATP required in this process?

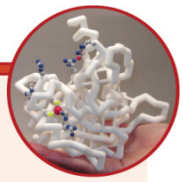
7g. After one cycle of the sodium-potassium pump, compare the overall charge of the inside of the cell to the outside? Explain how the distribution of ions changed.

7h. Is the sodium-potassium pump a channel protein or a carrier protein? Explain your answer.

7i. Devise a question you might have about the function of the sodium-potassium pump.

Fun Fact: In nerve cells, the sodium-potassium pump helps to reestablish the resting ionic concentrations after the nerve cell has fired.

Reference:
The Cell, 2nd edition
A Molecular Approach
Geoffrey M Cooper.
Boston University
Sunderland (MA): Sinauer Associates; 2000.
ISBN-10: 0-87893-106-6



Membrane Starter Kit

Teacher Copy

A Message to Teachers

Thank you for using our tools to help your students visualize the molecular world! The lesson/activity guide that accompanies the plasma membrane kit is intended to help you consider different ways in which you may use these materials. It is not the intent of the Center for BioMolecular Modeling to require you to work through the entire lesson from start to finish (although you may do so if you wish). We do **encourage** (and perhaps insist on) your **modification** of these lessons and activities to meet the learning objectives of your specific students or to accommodate the physical limitations of the environment in which you teach. Enjoy!

Objectives: You will use the model pieces in the kit to:

- Examine the general amphipathic structure of a phospholipid.
- Compare and contrast various models of phospholipids.
- Explore the interaction between phospholipids and water.
- Construct a phospholipid monolayer, micelle and bilayer and relate it to plasma membrane structure.
- Identify and demonstrate the function of the various types of channel proteins involved in membrane transport.

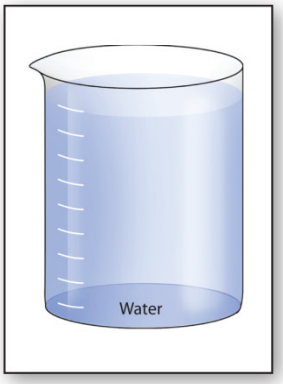
Introduction:

The **plasma membrane** is the structural boundary that separates the cell from its surroundings and controls what substances move into and out of the cell it surrounds. As only some substances are allowed to cross the membrane, the plasma membrane demonstrates the property of **selective permeability**. In particular, the plasma membrane of mammalian red blood cells (erythrocytes)

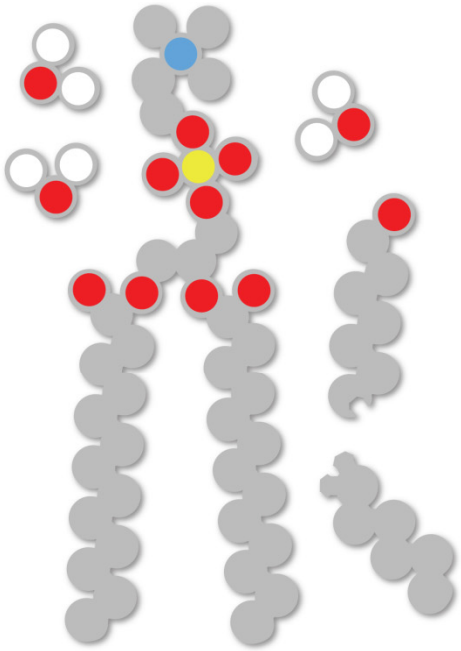
has been the focus of cell membrane study because these cells do not contain nuclei or internal membranes. As such, they represent a source from which a pure plasma membrane may be easily isolated for analysis. In 1925, the two Dutch scientists Evert Gorter and his research assistant F. Grendel extracted lipids from the membranes of a known number of red blood cells which corresponded to a known surface area of plasma membrane. The surface area occupied by a monolayer of the extracted lipid spread out at an air/water interface was determined. The results of their experiment showed that the surface area of the lipid monolayer was twice that occupied by the erythrocyte plasma membrane, leading to the conclusion that the plasma membrane consists of **lipid bilayers**. The most abundant lipids in most membranes are **phospholipids**. The ability of phospholipids to spontaneously form membranes is inherent to their **amphipathic** (meaning they have both a hydrophilic region and a hydrophobic region) nature.

Materials Needed:

Bag of Mini-Phospholipid Foam Models



Beaker Diagram



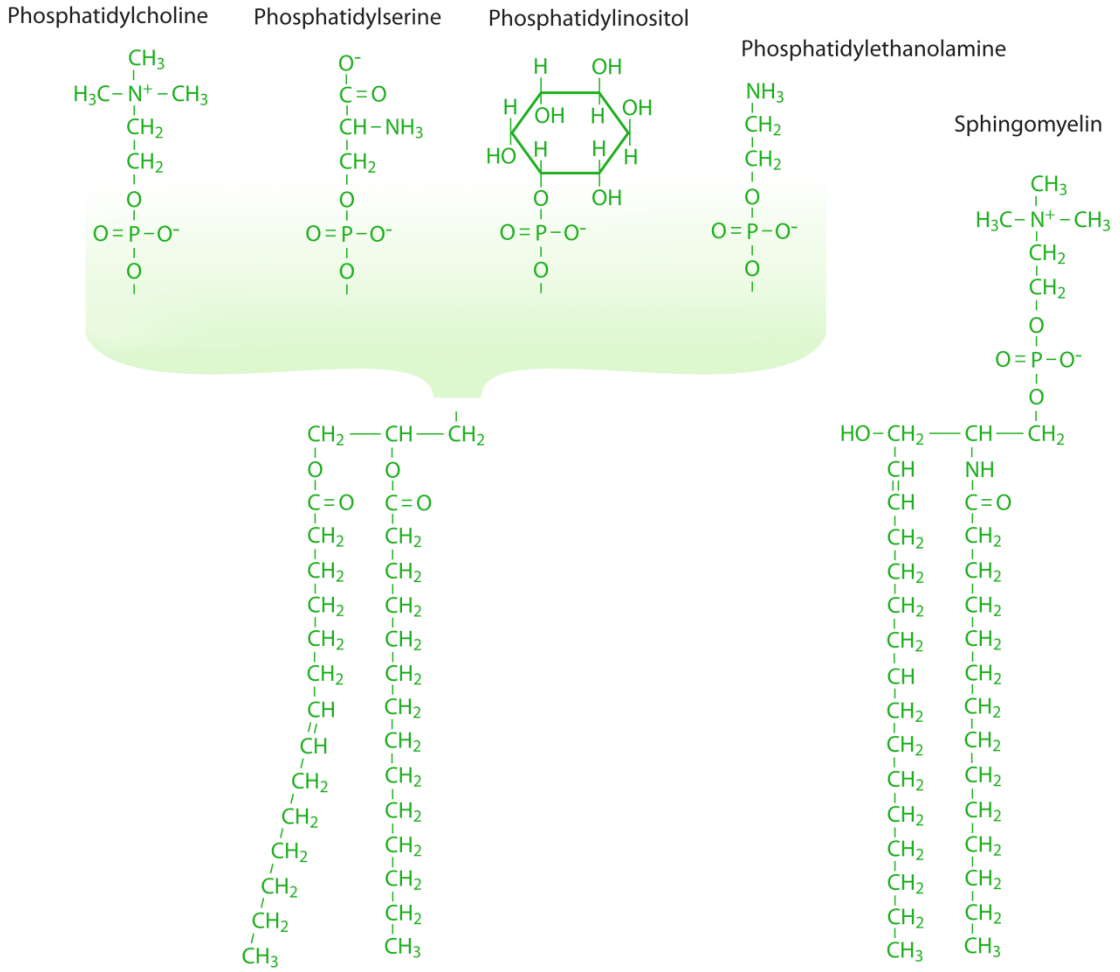
Large Foam Phospholipids

Large Molymod Phospholipids (optional)

Part 1: Focus on Phospholipids

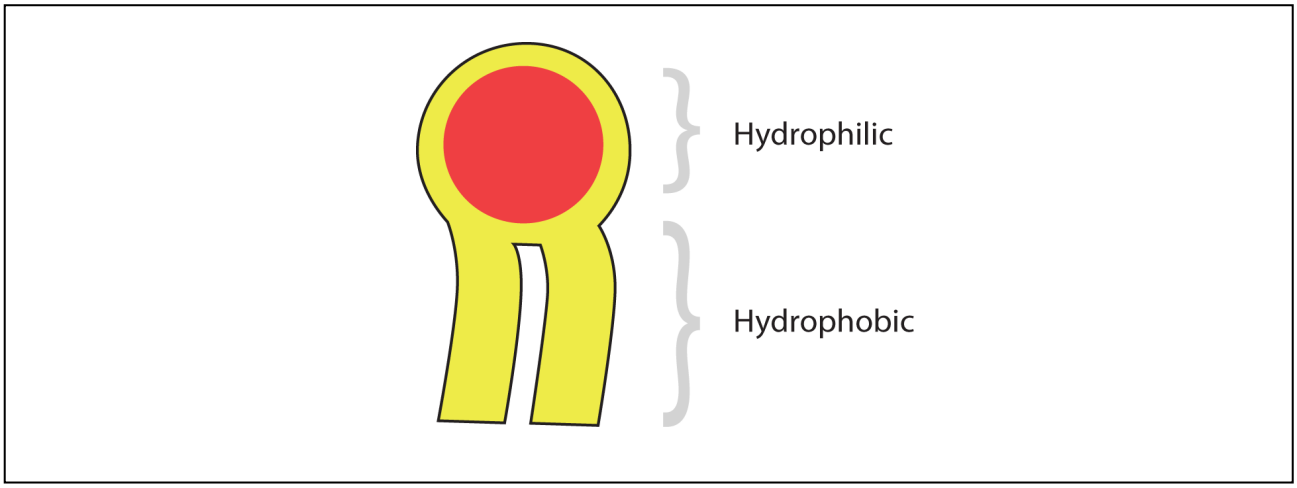
1a. Sketch the specific structural formal of the model you have been asked to examine in the space provide below. Label the hydrophilic and hydrophobic regions of your structure.

Answers will vary but should include one of the following:



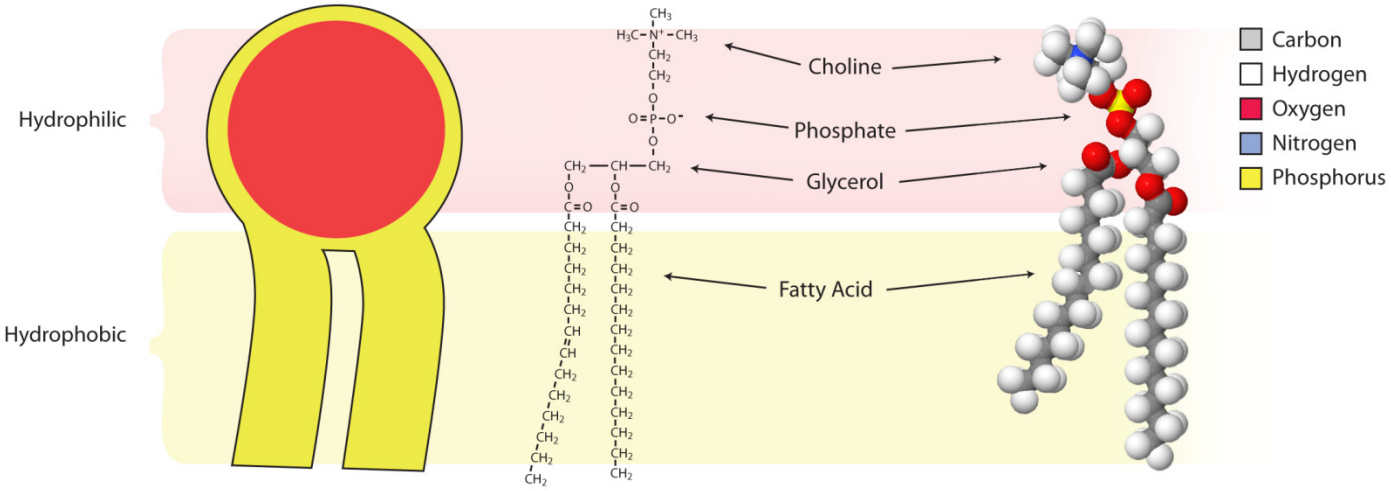
1b. Compare your structure to that of the other groups in the room. Record any similarities you observe in these phospholipid structures.
(All of the phospholipids have a hydrophilic head that contains a phosphate group and a glycerol. In addition, there are two hydrophobic tails in each phospholipid illustrated.)

1c. Based on these similarities a simplified representation may also be used to indicate phospholipid structure. Sketch this simplified model in the space below. Label the hydrophobic and hydrophilic portions of this simplified model.



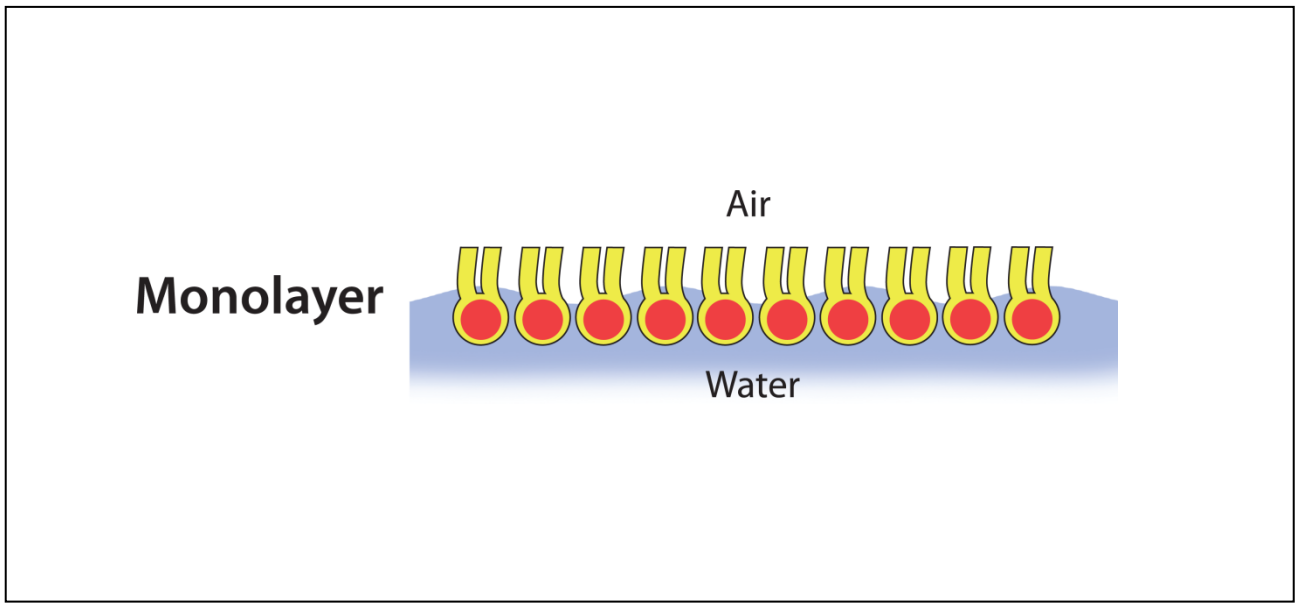
1d. Record any differences in the specific structures you have observed between these phospholipids.
(The hydrophilic head contains some compositional differences. There are four major phospholipids that comprise the plasma membrane. Phosphatidylcholine and sphingomyelin make up the outer leaflet layer of the membrane while phosphatidylethanolamine and phosphatidylserine make up the inner leaflet layer of the membrane. A fifth phospholipid, phosphatidylinositol is also found in the inner leaflet layer of the plasma membrane. Although phosphatidylinositol is a minor membrane component, it plays a major role in cell signaling.)

The general structure of a phospholipid is most often represented by the phosphatidylcholine structure:

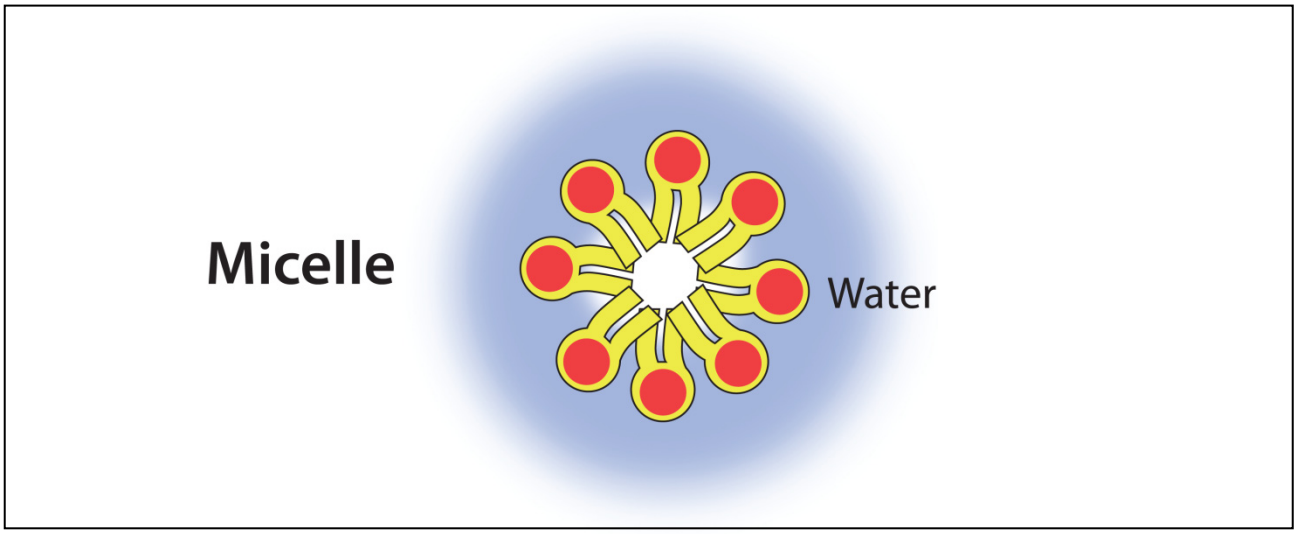


Part 2: The Kessler Membrane Activity

2a. Obtain ten of the simplified representations of a phospholipid. Using the diagram of the beaker of water, arrange the ten phospholipids demonstrating correct hydrophobic/hydrophilic interactions. Sketch your result below.

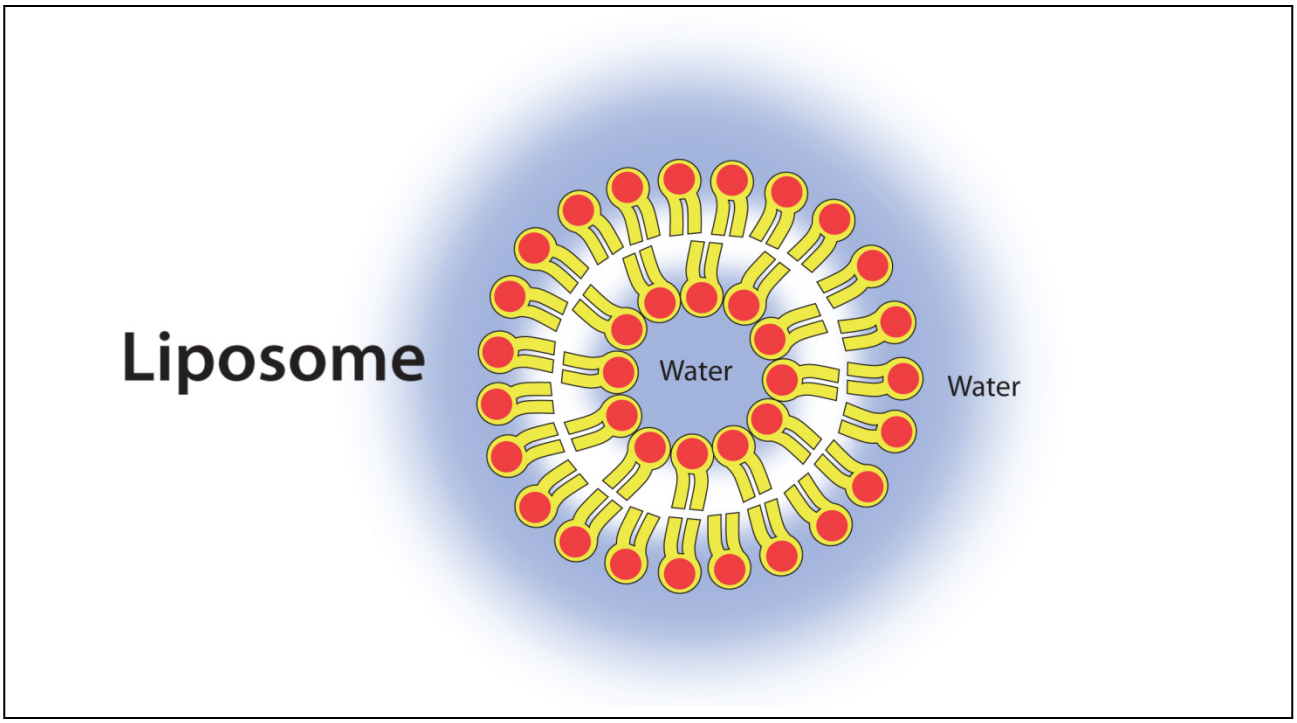


2b. Using the same ten phospholipids, rearrange them in the beaker so that they are submerged in the water while still maintaining the correct hydrophobic/hydrophilic interactions. Sketch your result below.



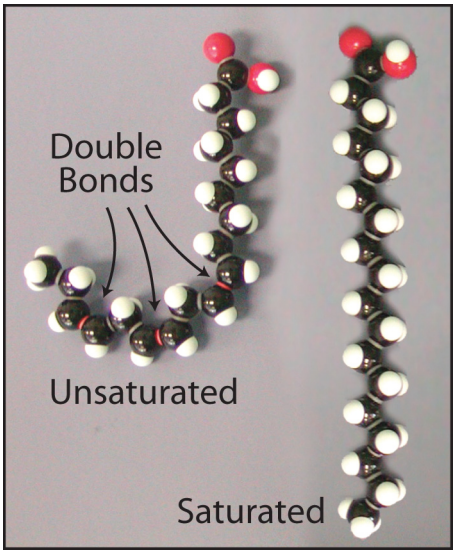
The resulting structure is referred to as a **micelle.** Micelles can act as emulsifiers that will allow a compound that is normally insoluble (in the solvent being used) to dissolve.

2c. Your next challenge is to construct a structure that is BOTH submerged in the water AND contains water on the inside. You may use as many of the phospholipids in your kit as you wish to complete the task. Sketch the resulting structure below.



2d. Based on the structure of a phospholipid, explain the reasoning for your arrangement.
(Because a phospholipid is amphipathic, this molecular arrangement shelters the hydrophobic tails of the phospholipids from water while exposing the hydrophilic heads to water.)

The resulting structure is referred to as a **liposome**. Liposomes are artificially prepared vesicles that may be used as a vehicle for administration of nutrients or pharmaceutical drugs.

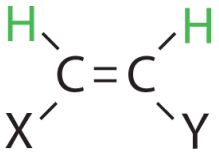


Upon closer examination of the hydrophobic tails, you may notice that in one of the tails, all of the carbons are connected with single bonds. These hydrocarbon tails are referred to as **saturated**. In the second tail there is a red connector, indicating that a double bond connects these two adjacent carbons. The presence of a double bond creates a "kink" in the structure of this hydrocarbon chain. The hydrocarbon tails containing a double bond or bonds between adjacent carbons are referred to as **unsaturated**.

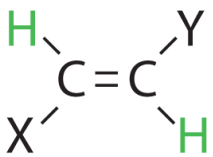
2e. Develop an explanation for the necessity of having "kinks" in the hydrophobic tail of the phospholipids that make up cell membranes.
(Because of the kinks in the tails where double bonds are located, unsaturated hydrocarbon tails cannot pack together as closely as saturated hydrocarbon tails contributing to the membrane fluidity.)

Note to Teachers - Optional Activity:

Further comparison shows a difference in these double bonds. If the hydrogens associated with the double bonded carbons are on the same side, the configuration is said to be "cis". If the hydrogens associated with the double bonded carbons are on opposite sides, the configuration is referred to as "trans". (See illustrations below.) Teachers may also opt to include a discussion of "trans fats" using these models.



cis



trans

2f. Which configuration produces the bigger “kink” in the structure of the hydrophobic tail?
(cis configuration)

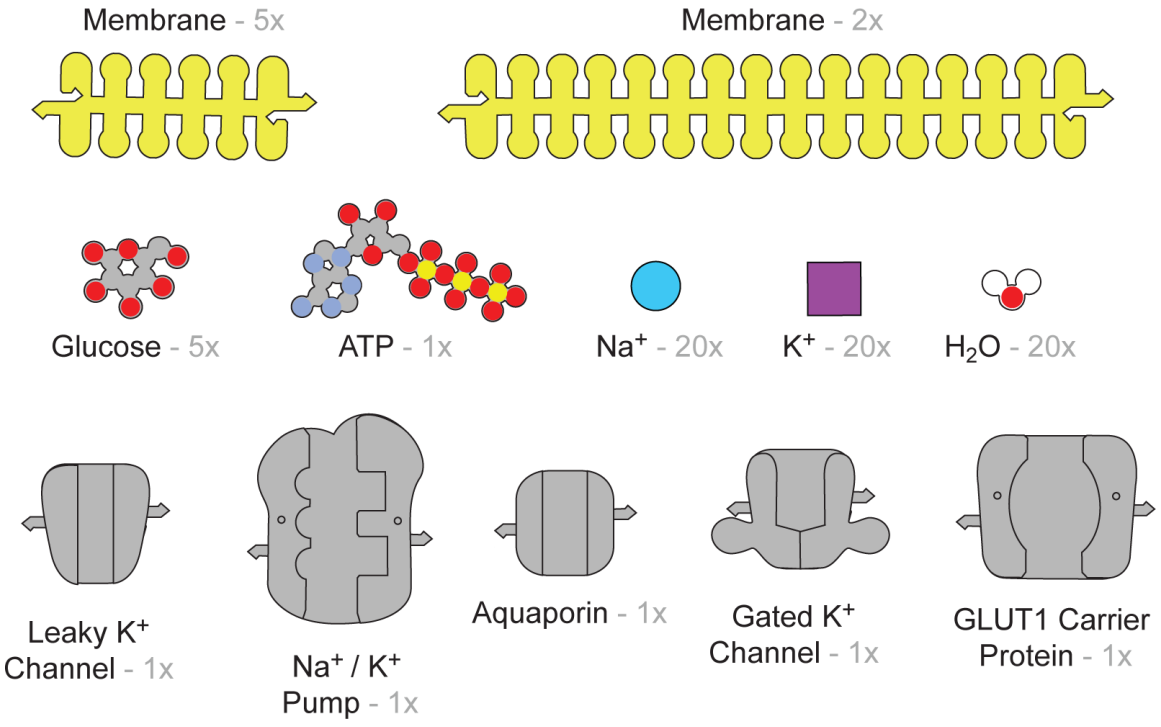
2g. How might this configuration contribute to the fluidity of the membrane?
(The “bend or kink” in the hydrophobic tail prevents these phospholipid tails from packing tightly together. The cis configuration would increase the fluidity of the membrane.)

Part 3: Exploring Membrane Permeability

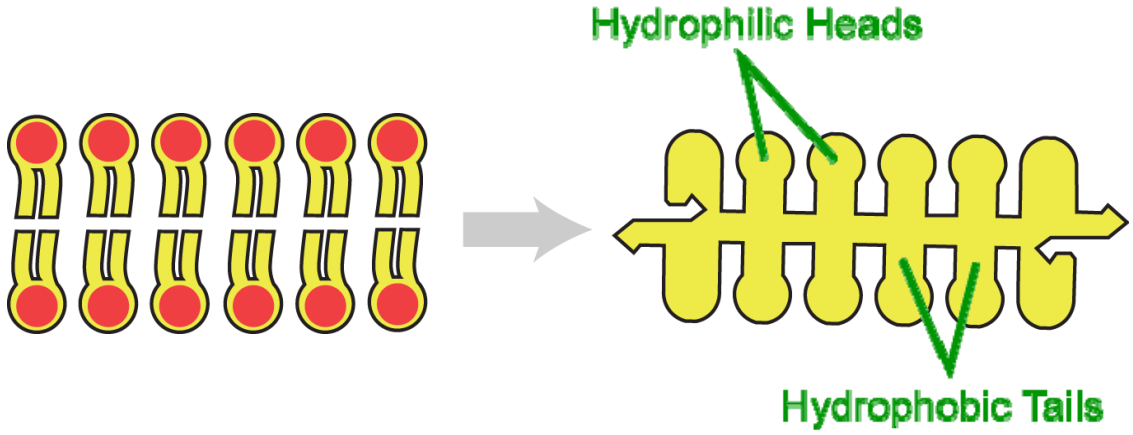
Introduction:

The phospholipid bilayer is only one aspect of the gatekeeper system responsible for the plasma membrane’s **selective permeability**. Membrane bound proteins play a key role in regulating the transport of ions and molecules through the plasma membrane.

Materials:



We will use a slightly different and simplified representation of the phospholipid bilayer in the next activity.



3a. Label the hydrophilic head and hydrophobic tail in the model pictured above.

3b. Nonpolar molecules, such as hydrocarbons, CO_2 and O_2 are hydrophobic. Explain why these molecules can easily cross the plasma membrane without the aid of proteins. (The hydrophobic nature of the interior of the membrane allows these hydrophobic molecules to cross the plasma membrane. Additionally, CO_2 and O_2 are small molecules that may diffuse across the membrane.)

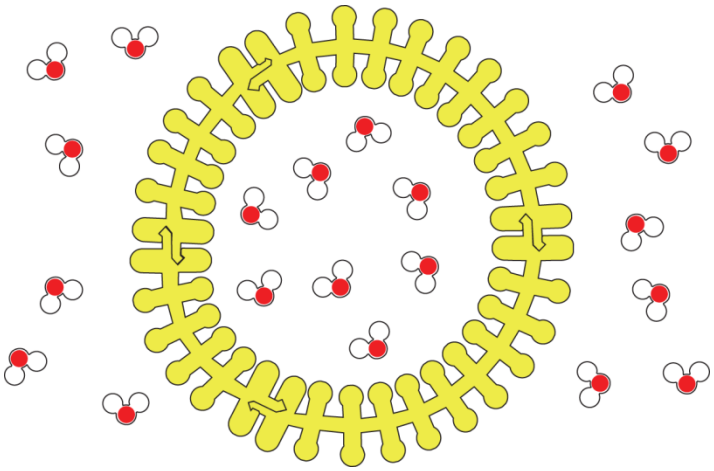
A variety of polar molecules and can't move through the plasma membrane on their own. Contact with hydrophobic lipid bilayer may be avoided by these hydrophilic substances as they cross the plasma membrane with the help of **transport proteins**.

Part 4: Channel Proteins

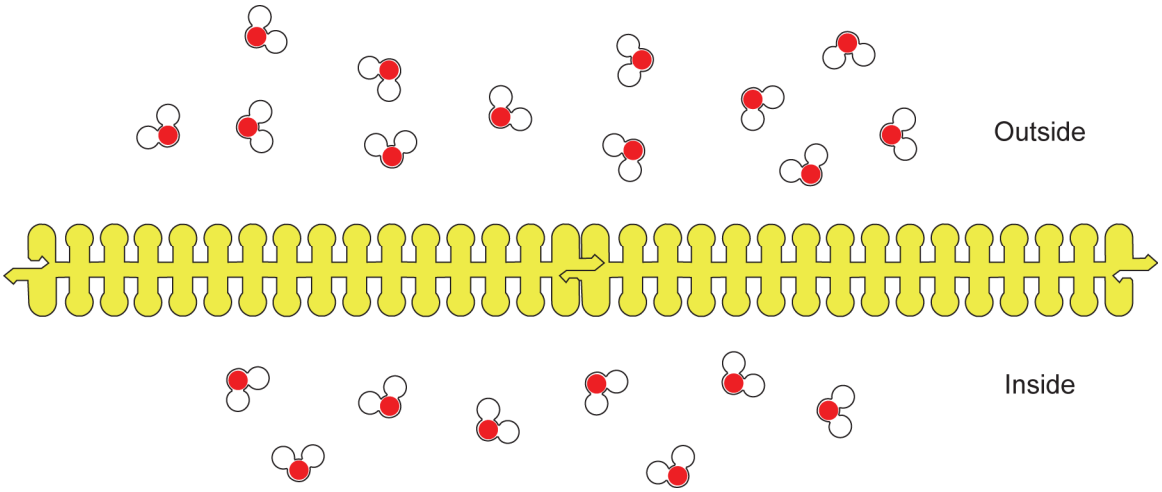
Some transport proteins, referred to as **channel proteins**, function by having a hydrophilic channel that certain molecules or ions use to cross the plasma membrane. There is a common misconception that a polar water molecule can easily cross the hydrophobic bilayer of the plasma membrane. Simple diffusion of water across the membrane does not occur at a rate fast enough in order to meet the survival needs of a cell. Channel proteins known as **aquaporins** allow entry of up to three billion (3×10^9) water molecules per cell per second!

4a. Construct a physical representation (a model) of the passage of water through a plasma membrane.

Use two short pieces and two long pieces of the simplified phospholipid membrane. Connect the pieces to form a "cell". We suggest placing 8 of the water molecules inside the cell (Intracellular) and 12 water molecules outside the cell (extracellular).

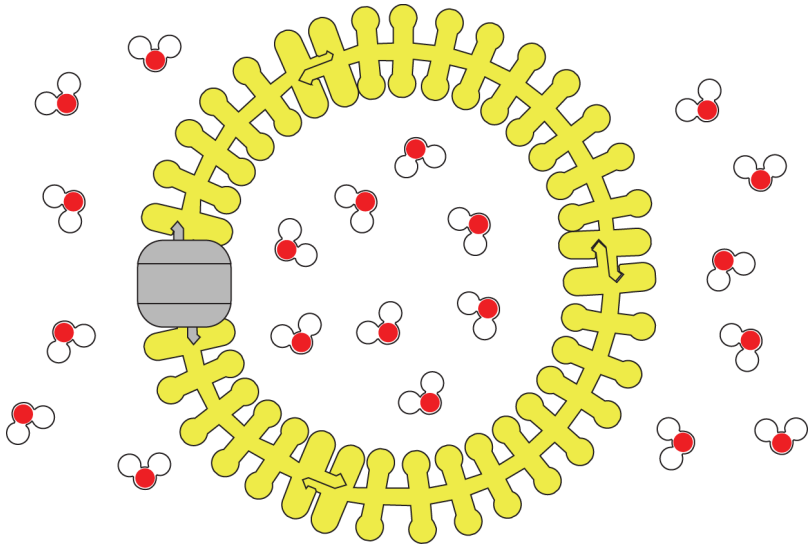


Note to Teachers - You may opt to set up the membrane in a linear fashion (shown below) if more conducive to your teaching environment.



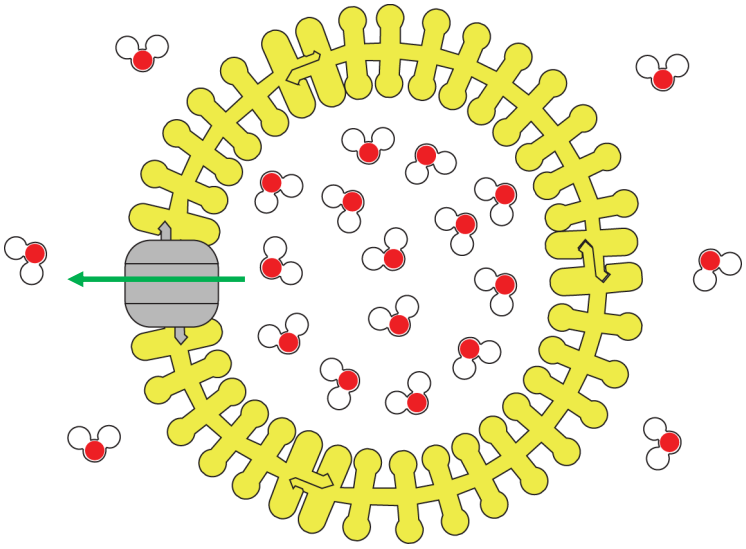
4b. Explain why water would have a difficult time diffusing across the cell membrane. Keep in mind the structure of water in your answer.
(The polar nature (slight negative charge on the oxygen end and slight positive charge on the hydrogen end) of water is not conducive to interaction with the hydrophobic phospholipid bilayer of a cellular membrane.)

A substance will generally diffuse from where it is more concentrated to where it is less concentrated. In other words, the substance will diffuse down its **concentration gradient**. Insert a channel protein model, in this case representing **aquaporin**, into the cell membrane you have constructed. Aquaporin is an example of a channel protein found in the plasma membrane which transports water (see diagram at right).



4c. Move the water molecules to indicate the net flow of water in this system. What direction did the water molecules move?
(In this configuration water molecules will move to the inside of the cell, moving from an area of high water molecule concentration to an area of low water molecule concentration.)

4d. Construct a system where intracellular water molecule concentration is higher than extracellular water molecule concentration. Sketch your model in the space below and indicate the net flow of water.

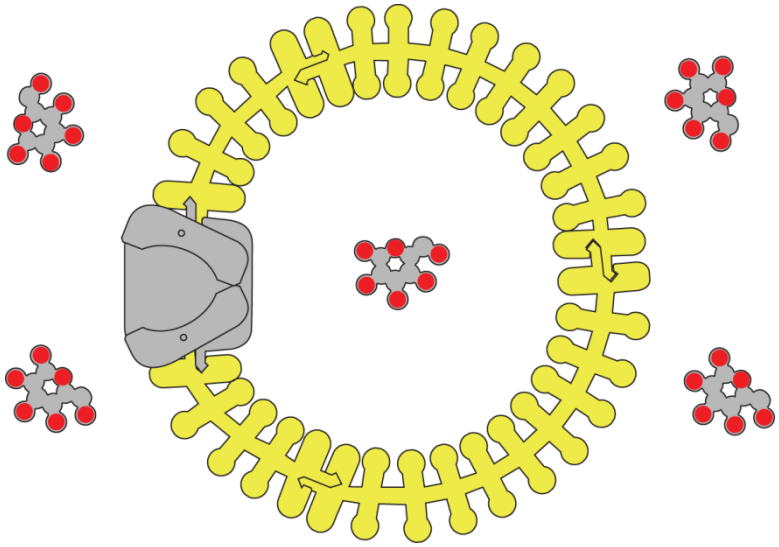


Note to Teachers -
You may choose to use the kit to introduce the students to the terms hypertonic, hypotonic and isotonic at this time.

4e. Predict what will happen to the cell due to the movement of the water.
(Since water is leaving the cell, the cell will shrink in size.)

In order for water move across the cell membrane in the above cases, the cell did **not** have to expend cellular energy (usually in the form of ATP) to move water across the plasma. Movement of a substance across a membrane without the expenditure of energy is referred to as **passive transport**. Likewise, when the cell must expend energy to move a substance against its concentration gradient the process is referred to as **active transport**. Additionally, the channel protein aquaporin assisted in passively moving water across the membrane. This occurrence may be referred to as **facilitated diffusion**.

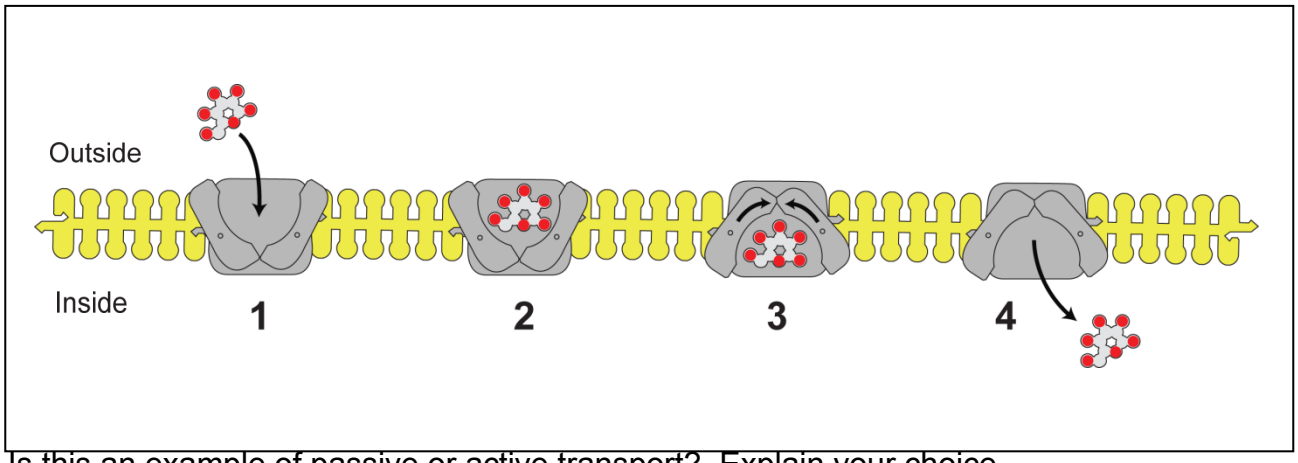
Part 5: Carrier Proteins



Remove the water molecules and aquaporin representation from the model cell you have constructed. Insert the carrier protein model into the plasma membrane model. Distribute the glucose molecules so that there are more extracellular glucose molecules than intracellular glucose molecules (see diagram).

A **carrier protein** binds a solute molecule on one side of the membrane, undergoes a shape change (conformational change) and deposits the solute molecule on the other side of the membrane. GLUT 1 is an example of a protein channel frequently found in the plasma membrane of red blood cells that facilitates the movement of glucose across the cell's plasma membrane.

5a. Use the model to demonstrate the movement of glucose across the cell membrane. Sketch your model in the space below.

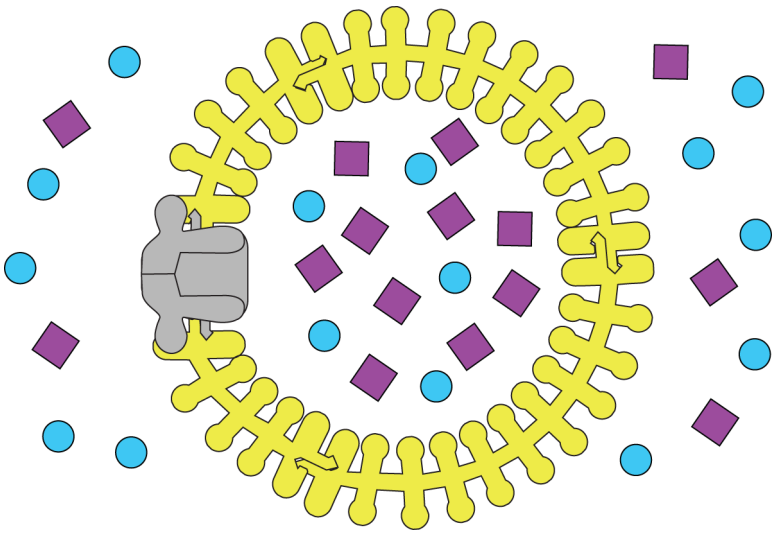


5b. Is this an example of passive or active transport? Explain your choice.

(Movement of glucose in this case is an example of passive transport because the cell does not need to expend energy to move the molecule against its concentration gradient.)

Part 6: Gated Channels

Remove the glucose molecules and carrier protein from the model cell you have constructed. Insert the gated channel protein model into the plasma membrane model (see diagram). Place 5 sodium ions (**round**) and 10 potassium ions (**square**) inside of the cell to simulate the intracellular environment ion concentrations. Place 10 sodium ions and 5 potassium ions outside of the cell to simulate the extracellular environment ion concentrations.



Gated channels are channel proteins that open or close in response to a stimulus. In nerve cells, a stimulus opens the gated sodium channel to allow a stream of sodium ions to enter the cell. Other gated channels open or close when a substance, different from the one to be transported, binds to the channel.

Begin the simulation with the sodium gated channel closed. After a stimulus the gates swing open.

6a. Based on what you know about concentration gradients, what direction will the sodium ions move through the channel?
(There are less sodium ions on the inside of the cell than the outside. Sodium ions will tend to move into the cell.)

6b. Does this gated channel demonstrate passive or active transport? Explain your answer.

(Sodium ions are moving from high concentration to low concentration. This is an example of passive transport because no energy is expended pushing them against their concentration gradient.)

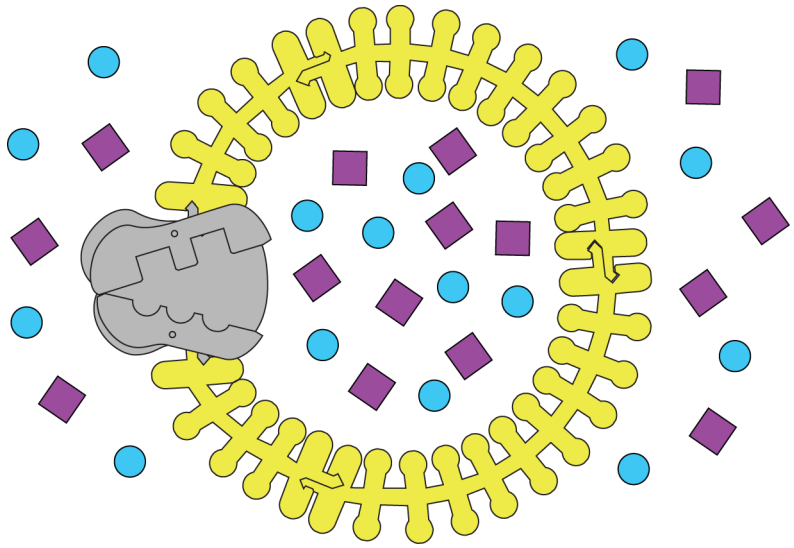
6c. Why may sodium ions move through the sodium channel while potassium ions typically do not?
(The sodium channel protein structure is such that sodium ions are allowed passage while potassium ions are not. Sodium ions are hydrated when they pass through the channel while potassium ions are not hydrated when passing through their channel.)

6d. Speculate what other stimului may affect the operation of channel proteins.
(Ligands may bind to a protein to trigger their opening or closing.)

6e. Devise a question you might have about the operation of this channel protein.
(Various answers)

Part 7: Active Transport - The Sodium-Potassium Pump

Remove the gated channel from the model cell you have constructed. Instead, insert the sodium-potassium pump protein into the membrane of the model. (See diagram below.) Place 7 sodium ions (**round**) and 8 potassium ions (**square**) inside of the cell to simulate the intracellular environment ion concentrations. Place 8 sodium ions and 7 potassium ions outside of the cell to simulate the extracellular ion concentrations.



Transport proteins that move solutes against their concentration gradients are all carrier proteins. The sodium-potassium pump is a special carrier protein that moves sodium ions against their gradient OUT of the cell and potassium ions against their gradient IN to the cell.

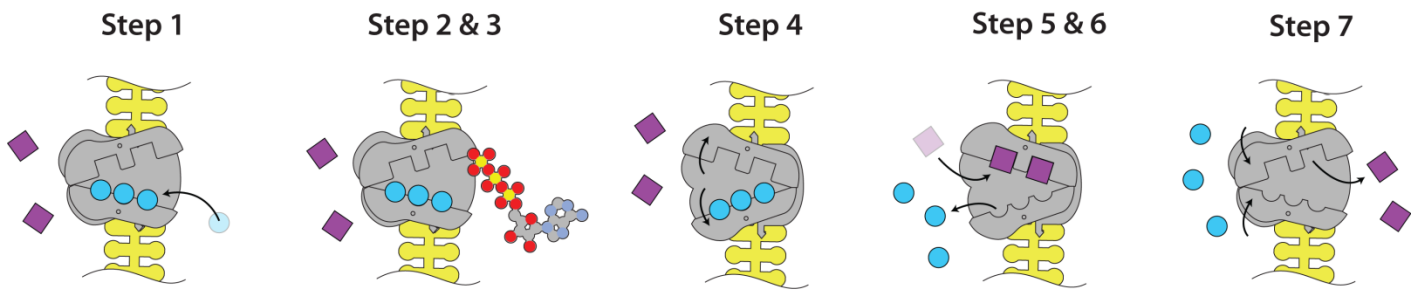
Because these ions are moving against their concentration gradients, the cell must expend energy to do the work resulting in **active transport** of these ions.

A typical animal cell has a much higher concentration of potassium ions (K^+) and a much lower concentration of sodium ions (Na^+) on the inside of the cell than the outside. The sodium-potassium pump uses energy in the form of ATP to move these ions against their concentration gradients and establish the "normal" intracellular ion concentrations. We will demonstrate the action of the sodium-potassium pump in the following simulation:

Set the sodium-potassium pump so that it is open to the inside of the cell as shown in the diagram on the previous page (page 14).

Record the initial ion concentrations in the table provided below:

Ion Types	Initial Amount	Amount After First Cycle	Amount After Second Cycle
Intracellular Na^+	**7**	**4**	**1**
Intracellular K^+	**8**	**10**	**12**
Extracellular Na^+	**8**	**11**	**14**
Extracellular K^+	**7**	**5**	**3**



Step 1: Bind three three intracellular sodium ions to the appropriate spots in the protein.

Step 2: Bring the ATP in close proximity to the pump.

Step 3: Sodium ion binding stimulates phosphorylation of the pump protein by ATP. In other words, a phosphate group is added to the sodium-potassium pump from the ATP molecule. (You will not be able to demonstrate this step with the model).

Step 4: Phosphorylation causes a change in the shape of the protein. You can demonstrate this by "swinging" the sides of the protein so that it opens to the outside of the cell.

Step 5: The shape change reduces the protein's binding affinity for sodium ions and increases the binding affinity for potassium ions. Remove the sodium ions from the protein and deposit them outside the cell and bind two potassium ions to the appropriate spots in the protein.

Step 6: Potassium ion binding triggers the release of the phosphate group from the protein. (Again, you will not be able to demonstrate this step with the model).

Step 7: Loss of the phosphate group results in the restoration of the protein's original shape which then releases the potassium ions. Swing the sides of the protein back so that they open to the inside of the cell and deposit the potassium ions.

Step 8: Repeat this process one more time.

Record the ion concentrations after completing the first cycle of the action of the sodium-potassium pump.

7a. What is the initial overall positive charge inside the cell compared to the outside the cell?
(There are 15 positive charges inside the cell as compared to 15 positive charges outside the cell.)

7b. Compare the total intracellular positive charge to the total extracellular positive charge after one cycle of the sodium-potassium pump.
(There are 14 positive charges inside the cell while there are 16 positive charges outside of the cell.)

Record the ion concentrations after completing the second cycle of the action of the sodium-potassium pump. Compare the total intracellular positive charge to the total extracellular positive charge after the second cycle of the action of the sodium-potassium pump.

7c. Where is the sodium ion concentration highest at the beginning of the sodium-potassium pump cycle?
(Sodium ion concentration is highest outside of the cell.)

7d. Where is the potassium ion concentration highest at the beginning of the sodium-potassium pump cycle?'
(Potassium ion concentration is highest inside the cell.)

7e. What is the initial overall charge of the inside of the cell compared to the outside?
(There are an equal amount of positive charges inside and outside the cell.)

7f. Why is ATP required in this process?
(In order for the cell to move sodium and potassium against their concentration gradients, energy must be expended by the cell.)

7g. After one cycle of the sodium-potassium pump, compare the overall charge of the inside of the cell to the outside? Explain how the distribution of ions changed.
(The inside of the cell has fewer positive ions than the outside of the cell. The sodium-potassium pump moves three sodium ions out of the cell for every two potassium ions in resulting in a redistribution of charge.)

7h. Is the sodium-potassium pump a channel protein or a carrier protein? Explain your answer.
(The sodium-potassium pump is a carrier protein because it is binds a substance, undergoes a shape change and deposits the substance on the other side of the plasma membrane.)

7i. Devise a question you might have about the function of the sodium-potassium pump.
(Various)

Fun Fact: In nerve cells, the sodium-potassium pump helps to reestablish the resting ionic concentrations after the nerve cell has fired.

Reference:
The Cell, 2nd edition
A Molecular Approach
Geoffrey M Cooper.
Boston University
Sunderland (MA): Sinauer Associates; 2000.
ISBN-10: 0-87893-106-6

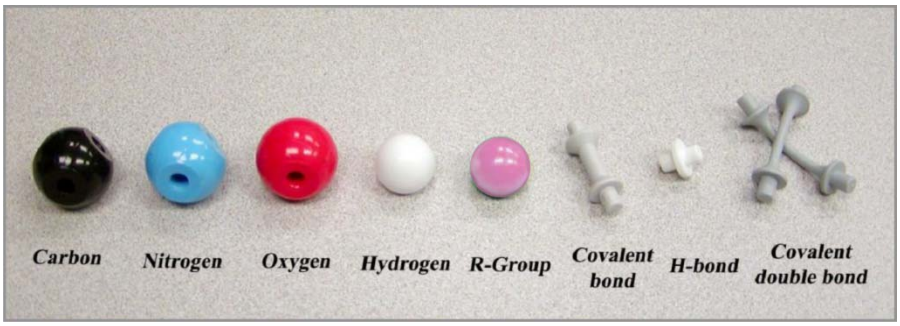
Center for BioMolecular Modeling

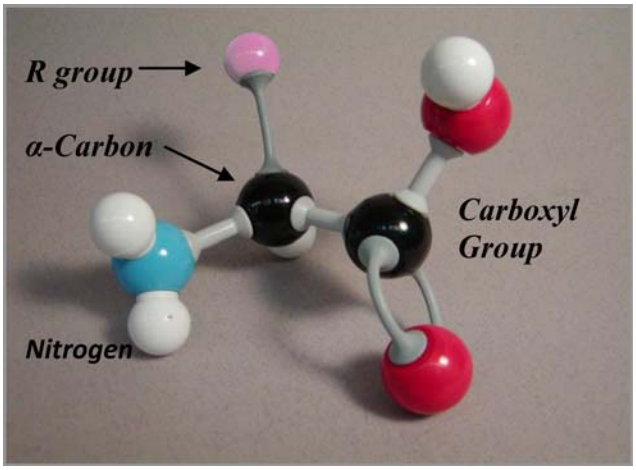
Amino Acids are the Building Blocks of Proteins

The purpose of this activity is to introduce students to the structure of an amino acid displayed in a ball and stick model format. Each atom is represented by a colored sphere; covalent bonds and hydrogen bonds are represented by "sticks". Students will build an amino acid and identify the atoms and parts of an amino acid. Students will build a dipeptide and identify components of the dipeptide. After this activity, students should be able to recognize an amino acid and identify the atoms. This knowledge is necessary to master specific commands in RasMol.

Molymod® Kit Contents:

4	Carbon (black)
4	Oxygen (red)
2	Nitrogen (blue)
2	R-groups (green), representing sidechains
10	Hydrogen (white)
6	Covalent bonds-single (thick, short, gray)
6	Covalent bonds-double (thin, long, gray)
10	Hydrogen bonds (short, white)
1	Molymod® link remover



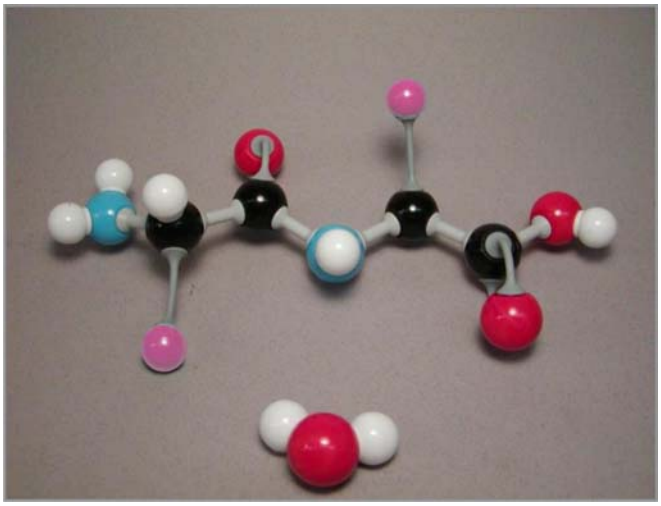


Activity:

1. Construct two separate amino acids using the Molymod® atoms, covalent bonds, and hydrogen bonds.
   a. Identify the following components: amino group, carboxyl group, the R-group or sidechain, α-carbon, carboxyl carbon, nitrogen. *(see labeled diagram above)*

b. Compare the two amino acids that have been built. Are they identical? How might two amino acids be different? *Amino acids are identical because they share the same "core" structure of NH_3-CHR-COOH. Amino acids are different because the composition of the "R-group" is different for each of the 20 amino acids. A second way that the amino acid structures may be different is their stereochemistry. The arrangement of atoms around the α-carbon may be "right-handed" or "left-handed" to form a D-amino acid or an L-amino acid. The L-amino acids are the naturally occurring form used to make proteins.*

2. Two amino acids can be chemically linked by a reaction called "condensation" to form a peptide bond linking two amino acids. A chain of amino acids linked by peptide bonds is called a polypeptide. Using the two amino acids built in step 1, create a dipeptide.



   a. What are the products of the condensation reaction? *The products are a dipeptide and a molecule of water.*
   b. Identify the following components of the dipeptide: amino groups, amino terminal end, carboxyl groups, carboxyl terminal end, carbonyl group, peptide bond, R-groups or sidechains, α-carbon, carbonyl carbon.

Teaching Points:

- Amino acids are the building blocks of protein
- Amino acid structure
  a. Identify atoms: nitrogen, oxygen, carboxyl carbon, α-carbon, oxygen, hydrogen
  b. Identify groups: amino terminal, carboxyl terminal, R-group or sidechain,
- There are twenty different amino acids
  a. Amino acids are identical because they share a core structure
  b. Amino acids are different because they have unique R-groups or sidechains
- Linear chain of amino acids is a polypeptide
- Primary sequence of protein is the linear sequence of amino acids