

Articles

Teaching Molecular 3-D Literacy

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An understanding of three-dimensional structure/function relationships is increasingly important to modern biochemistry and molecular biology. Not only are the overall “folds” that illuminate evolutionary relationships uncomprehensibly three-dimensional but so, too, are the critical details at active sites. Fortunately, most protein and nucleic acid structures are now readily available on the Internet from the Protein Data Bank [1, 2], as are good tools for displaying them on small computers such as Mage [3, 4], Rasmol [5, 6], Chime [7], and SwissPDBViewer [8, 9]. However, teaching these skills and concepts effectively is nevertheless a challenge, because the prior education of students has concentrated entirely on one-dimensional (verbal) and two-dimensional (static pictures) information; many students feel unfamiliar and uncomfortable with 3-D¹ materials.

Many biochemistry textbooks now come with supplements using molecular graphics, such as Voet *et al.* [10], Branden and Tooze [11], and Horton *et al.* [12]. There are web sites with teaching materials on protein structure, such as Eric Martz’s Chime and Protein Explorer site [7], the Protein Society’s ProTeach site [30], Robert Bateman’s undergraduate kinemage site [31], our own Kinemage Homepage [4], and a site planned for this journal. There have also been papers reporting on teaching use of these molecular graphics tools in a variety of settings and approaches [13–16]. So far, however, there is little experimental data on either the absolute or the relative effectiveness of these materials for teaching 3-D literacy and only minimal guidance about the best ways to use them in the classroom. We do not yet have hard quantitative data on effectiveness, although we are now working on that problem under a joint National Science Foundation grant with Robert Bateman. However, for many years we have been using molecular graphics in our classes and striving to improve the 3-D literacy of our students and colleagues. We have learned a number of things that don’t work and some that do, and we would like to share those lessons with other biochemical educators.

In the 1970s we learned to make “worm” drawings of protein backbones on the blackboard, but we taught

mainly from hard copy handouts of text and 2-D figures. Our first experience developing systematic teaching materials to illustrate molecular structure was over 20 years ago, when J.S.R. worked out conventions for hand-drawn ribbon schematics such as Fig. 1, aiming to do as well as possible at translating the 3-D organization of protein “folds” into a static 2-D form on paper [17, 18]. Descendants of those representations, now rotatable in three dimensions, are a standard feature of current molecular graphics. We still give our classes a set of full-page line drawings as a “coloring book” and have them hand back a few pages colored in some way that makes sense in three dimensions. Except for an occasional M.D./Ph.D. student who feels insulted by this assignment, everyone else enjoys it very much; their results are often very inventive and/or esthetic. The active involvement in decoding the 2-D picture helps them connect printed figures with their computer exercises and also helps them recognize the handedness of structural features.

Soon after the ribbon schematics, our research laboratory obtained an Evans & Sutherland PS300 graphics machine, and D.C.R. wrote a display program (Chaos) for it. We then developed an extensive series of laboratory exercises with that system for the graduate students in our advanced seminar course. This was very effective but only workable for a dozen students at a time. In the 1980s we also made several movies and a large number of slides for use in introductory classes. Movies can communicate very well for a specific example, but we never had very many, because making good ones was very difficult, they could not be modified to suit new needs, and they inherently lacked the ability of interactive graphics to zoom in and show the answer to a student question. The use of actual physical models is also an effective learning tool, but they are more expensive and require more teacher interaction than computer graphics, and they work well only for small scale pieces of structure. We still use both brass Kendrew stick models and plastic “CPK” space-filling models in our seminar class to build initial familiarity with the geometry and motions of small atomic groupings up to a few residues.

In 1989, with the help of a North Carolina Biotechnology grant, we first got a computer projector that could show interactive molecular graphics to a large group in the classroom. It was a big, expensive machine, with a special fast phosphor to allow stereo; it required a fussy alignment and convergence procedure, and we had to wheel an

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¹ The abbreviations used are: 3-D, three dimensional; 2-D, two dimensional.

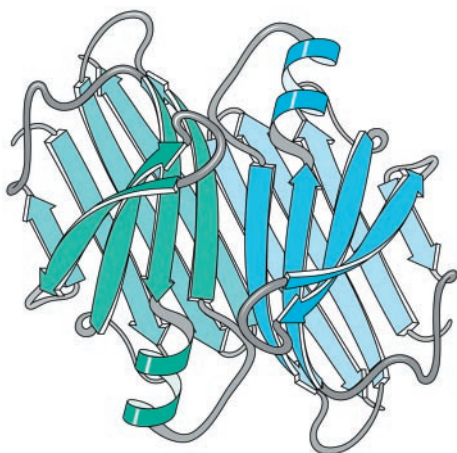


FIG. 1. **Hand-drawn ribbon schematic of a dimer of prealbumin, or transthyretin.** β -Strands are represented as *arrows*, α -helices as *spiral ribbons*, and loops as *round ropes* with *smoothed contours*. From PDB file 2PAB [22]; line drawing from Fig. 62 of Richardson [17].

entire Evans & Sutherland work station (by then an ESV) in on a cart. We were very excited about the possibilities, however, because it let us manipulate in real time for the class either stick-figures or multistrand ribbons (e.g. see Fig. 2) of the molecular structure so that the students could see the same movable images that gave us so many insights in our research. We gradually mastered the different rhetoric needed for this new medium: continual slow rotation back and forth, gradual zooms, markers at the points of interest, etc. We almost never used the stereo capability, because it turned out that motion provided better 3-D perception than stereo with much less hassle and could be seen by everyone.² The projection system had clearly taken us far beyond the static 2-D images of slides or textbook illustrations, and we got enthusiastic feedback from a number of the students; however, this was still mostly an enrichment activity that made no obvious difference to any learning that was tested on our exams. More importantly, by querying a wider range of students, we realized that the enthusiastic comments came from the ones who had already been familiar with 3-D structure and graphics, whereas the students who needed 3-D literacy training the most were as much confused as illuminated by these classroom presentations.

The answer to this problem was having students work with interactive graphics themselves, at their own pace, which for sizable classes required capable molecular graphics on small personal computers. In 1991, both for the needs of the new *Protein Science* journal and for use in teaching, we developed the concept of the kinemage (from *kinetic image*), and D.C.R. wrote the Mage display program [3], originally for the Mac but soon ported to Windows and later to Unix, Linux, and Java [19, 20]. The kinemage is a different way of organizing computer graphics that is aimed explicitly at the communication of specific ideas in 3-D. It is a two-step process with the kinemage file authored from the atomic coordinates by one person

² 10–15% of people do not actually have effective vision in both eyes, so they cannot see stereo with any type of hardware or training.

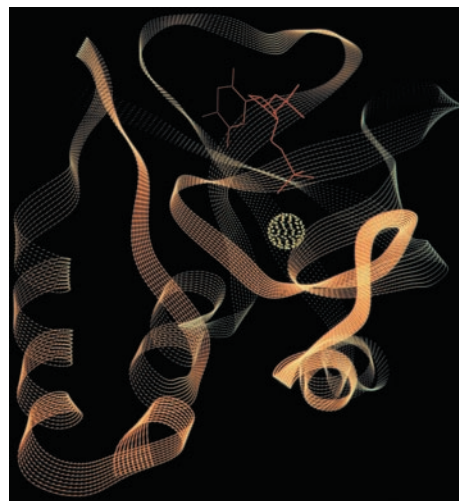


FIG. 2. **Multi-strand ribbon representation from the older graphics program Chaos, showing Staphylococcal nuclease with the thymidine diphosphate inhibitor in red and the Ca^{2+} as a yellow sphere.** From PDB file 1SNS [23].

(usually using the companion program Prekin) and the kinemage then displayed in Mage to be explored openly by that same person or by someone else (e.g. the student); thus, it is like a textbook illustration brought to life. As for any illustration medium, the critical aspect of authoring a kinemage is guiding the viewer to successfully perceive the key relationships by use of viewpoint, color, emphasis, and especially by leaving other things out. In addition to the graphics window, there is a scrollable text window with whatever explanations and instructions the kinemage author thought appropriate.

The kinemage format works well for seeing the overall shape of a macromolecular fold using either $\text{C}\alpha$ backbone or ribbons, plus a few orienting details (Fig. 3, A and B). Kinemages really shine, however, at animating motion (not shown here, of course) and at making clear the complex details at an active or binding site (Fig. 3, C and D). In an exercise, for instance, some bonds to a ligand could be shown initially, whereas students are asked to identify, measure, and draw in others. Side chains, atom types, H-bonds, or contact dots are easily shown in a region of interest without including them for everything. Kinemages are easily modified later, either using the on-screen editing tools in Mage or editing directly in the plain-text kinemage file, where all atoms and residues are labeled, and the keywords are in English (e.g. color= gold). The reader functions in Mage are very simple and intuitive so that students can almost immediately begin interacting with the molecules rather than investing time in learning the program. Because of these advantages we have almost exclusively used kinemages in our own teaching. However, most of the lessons we describe here could also be set up using other software, although with varying degrees of ease for teacher and for student.

The most central function of molecular graphics in biochemistry teaching is to have the students explore in 3-D the overall molecular organization of proteins and nucleic acids, the structural details that underlie that organization, and the critical relationships at binding and active sites. For that purpose, we first demonstrate the use of Mage in

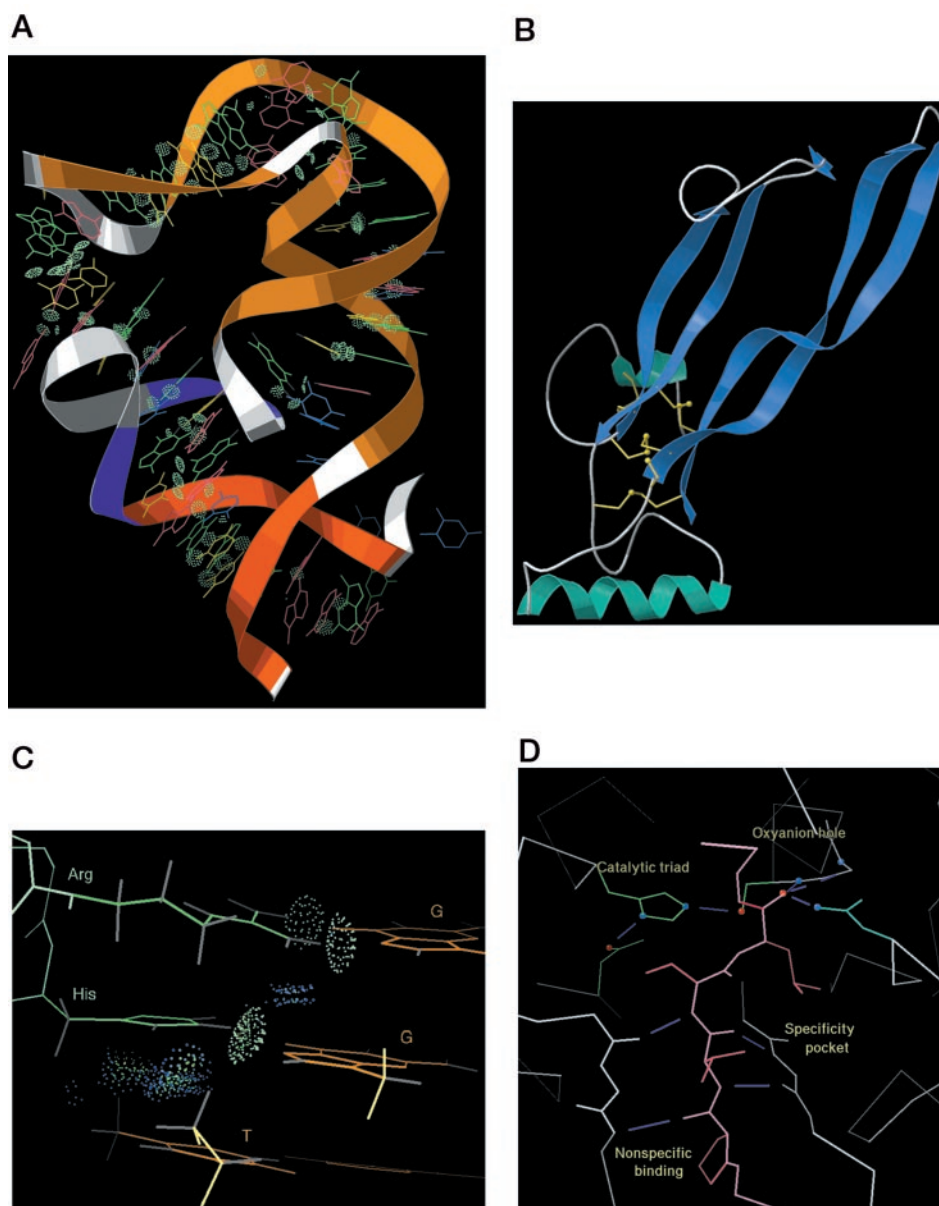


FIG. 3. **Screen-image snapshots from kinemages displayed in Mage.** *A*, helix 7 region of the ribosomal 23 S RNA from PDB file 1FFK [24], with backbone ribbon colored in the double-helical sections, bases colored by type, and *pale green lenses* of contact dots showing the base pair hydrogen bonds (output from Mage as PostScript). *B*, ribbon schematic of transforming growth factor β , with disulfide bonds in *yellow* (from PDB file 2TGI [25]; output rendered in Raster3D). *C*, interactions between the central zinc finger of Zif268 (PDB file 1AAY [26]) and DNA, with H-bonds shown as *pale green lenses* of contact dots and van der Waals contacts in *green* and *blue*; at the *top* is a typical Arg-G double H-bond, and at the center the His ring H-bonds to a G base and stacks with an T base. Hydrogens were added by Reduce [27], and all-atom contacts were by Probe [28]. *D*, selected details at the active site of subtilisin file 1CSE [29], with H-bonds and non-carbon atom balls for the catalytic triad, the oxyanion hole, and some substrate binding interactions. Parts *C* and *D* are from the Kinemage Supplement to Branden and Tooze [11], output as screen-image bitmaps. Of course, these snapshots convey much less information than the moving image, which is the whole point of using interactive graphics.

class (as part of an introductory and motivational presentation of some especially elegant, intriguing, and/or timely structures) and then assign a set of kinemage exercises to be done as homework. In these exercises, each program function is thoroughly explained in the kinemage text window the first time it is needed, from the simplest operations such as rotating by dragging or identifying a point by clicking on it, to more advanced features such as how to measure dihedral angles or how to construct a hypothetical $C\beta$ onto a glycine. In addition, the students are advised to consult the explanatory Demo5_4a.kin file if they are having any trouble with the mechanics. We have used

this format most extensively in first-year graduate Biochemistry and interdepartmental courses at Duke University but also in a shorter version for undergraduate courses, workshops at meetings, and guest lectures at other institutions. We use a longer and somewhat different format for an advanced seminar-laboratory course; in addition to graduate students in biochemistry and related fields, this course often attracts undergraduates, post-docs, or faculty, as well as graduate students from computer science, chemistry, and now bioinformatics.

Our initial teaching kinemages for proteins show the polypeptide backbone and sidechains (with rotatable an-

gles and optional H atoms) and illustrate backbone ϕ, ψ and sidechain rotamer preferences. Next, H-bonding, secondary structures, motifs, and domains are shown, leading to examples of the common folds and their organizational principles, and finally, structure/function relationships are studied for DNA binding and enzyme active sites. Along the way, the following two other aspects of molecular 3-D literacy are emphasized: 1) handedness preferences at all size scales, with practice cases, and 2) atomic packing interactions inside and between molecules, shown with all-atom contact dots (e.g. see Fig. 3C) to counteract both the impression of emptiness given by stick or ribbon models and the hiding of interactions by space-filling models. We also include a lecture and exercise on motions in proteins, which we consider almost as important as the equilibrium structures. Motion is covered relatively poorly in most textbooks but can be shown really well by animation in kinemages. Most of this subject matter is incorporated in the kinemage supplement to the second edition of Branden and Tooze [11], which is available on CD from the publisher or on our web site [4]. However, the supplement acts mainly as an enhanced version of the textbook illustrations and their captions and only occasionally incorporates explicit exercises with questions to be answered.

Probably the most important and simplest lesson we have learned about teaching with molecular graphics is that there must be individual student homework involving worksheets with explicit questions to be answered and that, at least in lower level classes, those worksheets must be handed in. There are three quite different reasons for this. The most intellectually interesting reason is that when studying new and complex data one needs some specific aim or question to focus the attention and conceptually organize perception of the details, even if the original question sometimes acts mainly to suggest another, better question. We have certainly found this essential in our research on protein structure. For student exercises, we have found that most of the questions should be very simple and straightforward (the students are very tolerant of answering even quite trivial questions in this context where they are looking at something interesting), leading up to some questions that are deeper and more complex and which address issues of real functional importance.

The other two reasons are practical. Your class competes with all the other demands on students' time; for the molecular graphics to enhance learning, though, they must not only be done but must be done at the right time to instantiate the material from one set of lectures and give the background for the next set. In particular, if these exercises are optional or are not monitored, then they will often be done only by those students who need them least. In our experience, however, it is not actually necessary to assign grades for the exercises, only to collect them, check off that they were done, and hand them back with some comments. In our advanced seminar class, we simply hand out an answer sheet the following week. The third reason is so the teacher can monitor whether the exercises are at the right level for the class and are actually working. Questions and explanations can then be modified where many students were confused. Our exercises (and the program, as well) have been much improved over the

years by feedback from patterns of wrong answers and also from two questions we ask at the end: Where did you have trouble, and how could it be made clearer? What part did you find most interesting or helpful? For instance, we discovered that way that some of our students did not initially know much at all about hydrogen bonds. We also found that many students were intrigued to learn about the structural roles of the amino acids [21] and how they can lead to surprising mutational patterns (e.g. that sometimes Gly is a more conservative replacement for Asn than is Gln), and we have since developed a lecture and an exercise focused on that topic. We hand out the worksheets on paper; text files from which they can be printed, as well as the kinemage files they use, can be found on our web site [4] in the folders for various courses (most recently CMB 258 and BCH222).

After the students have done one or two kinemage exercises on their own, then interactive graphics can be used effectively in lecture or discussion sessions, with essentially everyone following successfully if movements are done slowly and smoothly. Computer projectors are now small, simple, and crisply focused, although we hope the color rendition continues to improve. Mage is easy for the instructor to use in class, because all functions are on-screen (but often with keyboard shortcuts, such as "a" for animation). Once started, rotation by mouse motion behaves consistently no matter where the cursor travels, so the lecturer can pay attention to the class rather than the computer screen. Mage has a number of features specifically designed for presentation, such as optional side-to-side rocking motion, markers on the last two points picked (so students don't miss out if they look down at their notes), the capability to flash those markers larger and smaller for emphasis, and optional enlargement of the fonts of labels and of the display line that shows atom identity and distance (either by selecting from the pulldown Display menu or by just hitting the "w" key). An initial exposure to interactive graphics in the classroom is, of course, needed to show students what to expect when they use the program themselves. After that, classroom graphics can enhance the realism and specificity with which any molecular structure and function can be taught. Once students have acquired molecular 3-D literacy, usually in introductory structural biology material, then interactive graphics can be used wherever desirable for other topics, instructors, or courses with very little start-up effort.

There are several practical and strategic issues of concern in assigning molecular graphics homework exercises. We find that the most persistently awkward one, although for only a relatively small percentage of cases, is simply downloading the program in a form that runs successfully (the kinemage files, on the other hand, are plain ASCII text and almost always download fine). Mage is maintained on almost all currently common hardware and operating systems, and in the days when we used either a Mac or a PC diskette to hand out the program and files (which are quite compact), there were very few problems. Now the transfers are all done over the Internet, which is very convenient and modern but occasionally fails because of the idiosyncratic interactions of particular browsers, web servers, and

operating system setups. Therefore, as the assignment for the first day, we ask the students to download Mage and the demo file, check that they can open the kinemage, and report back. For the few with a problem, there is then time for troubleshooting or alternative provisions. These difficulties are gradually lessening as browsers and servers improve and as we learn more tricks for improving our website, but the process is certainly not as foolproof as one would like it to be. Fortunately, once Mage is up and running, the students have very few difficulties and usually require no other extra help from us.

One way of avoiding the download problem is to have the students work together in a computer laboratory or cluster with the software and files already installed. That system has other advantages, as well; it ensures timely work on the exercises and makes it easy for the instructor to be there to answer questions. We have usually used such a setup when visiting other institutions. However, there are also disadvantages, which we feel on the whole outweigh the assets. No matter what the level of care, we have essentially never had all the machines set up equivalently, so that troubleshooting is still needed. Notably, it seems to cause problems if the program is run from a central server rather than installed on each machine. After such a lab session, the student has not been effectively empowered to use the graphics easily on their own computers for other purposes. Most importantly, when the class works as a group, their very different levels of prior familiarity and aptitude are glaringly obvious, so that both timing and psychology make it difficult for those students who can benefit the most to spend the time they need and to feel good about what they have learned. These can be some of the best students in other ways, with a very thorough understanding of the verbal and even mathematical aspects of biochemistry; if they successfully work through the three-dimensional material at their own pace, they often integrate it into a very powerful overall knowledge. We feel these are the people for whom 3-D literacy makes the most difference, and therefore we try to tailor our teaching methods for them as well as we are able.

For further empowerment, we want students to learn how they can use molecular graphics for their own interests in the future. Therefore, we now always include some level of exercise in which they locate relevant coordinate files and make their own kinemages (always done after they have become familiar with pre-made kinemages). In an introductory course where structure is a minor part, this may be a brief exercise where they go to the Protein Data Bank web site [2], use the search tools to find, for instance, a file for α -hemolysin that includes all seven subunits, download it, use Prekin to make one kinemage of the backbone or a ribbon of the whole molecule and another kinemage using the "lots" script on a single subunit, and then answer a few questions about the structure. If other bioinformatics skills, such as sequence matching, are also being taught, then integrating those with molecular graphics enhances both. At the other extreme, in our advanced seminar course, the students spend half of the semester working on an individual project studied and reported in kinemage form, with a progress report shown in class every other week and a final presentation at the end. A

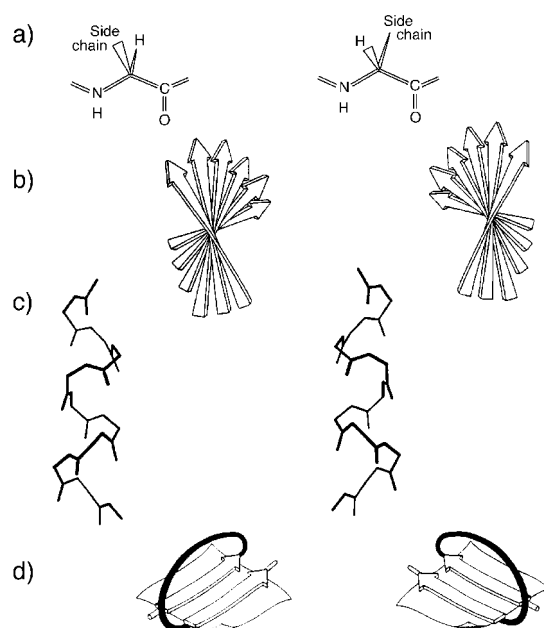


FIG. 4. A sample exam question testing ability of the students to recognize the correct biological handedness of various molecular structures, from α -carbons through secondary structures and motifs. The correct answers are as follows: a, left; b, right; c, left; d, left.

report done in kinemage format (which can be much shorter and simpler than the one described above for the advanced seminar and can be handed in electronically rather than presented in class) makes a very effective way to focus the individual learning of a student about a specific biological structure issue of interest to them, and it is a much more revealing medium than either a written report or a standard oral presentation for allowing the teacher to evaluate the depth of their understanding and their level of original insight. Our most important overall criterion for judging the reports is whether the student has explicitly shown in the kinemage enough of the right details to independently judge the correctness of what is said in the papers, with appropriate explanations in the kinemage text window and/or in their presentation. If they find something to criticize or something new to show, then all the better.

In the absence of a detailed individual report, however, there remains the problem of evaluating the learning that has taken place in the molecular-graphics component of a biochemistry class. We use a combination of traditional written exam questions and a take-home examination in which the questions refer to one or more kinemage files for structures the students have not seen before (new or relatively obscure proteins or occasionally ones that we have redesigned for this purpose) but with functional similarities to examples studied in class. Our most often-used question, usually presented on paper, is to distinguish the correct biological handedness of paired examples from amino acids up through motifs or folds; Fig. 4 shows an example of such a question. Our students do very well (average, 94% correct) after the kinemage exercises, in contrast to trial results on the first day of class, which were only barely above random (average, 56% correct). For more usual written questions that test biochemical knowledge the way it is more traditionally taught, the use of

molecular graphics in the course can make some contribution but not a major one. On the other hand, exams done in the graphics medium can never fairly compare learning with *versus* without the use of molecular graphics, because the control group is unfamiliar with the medium. However, it is the individual student's acquisition of a working 3-D literacy for macromolecules that is our main concern, and those skills and concepts can themselves be tested readily in the graphics medium. Any teacher wishing examples of our exam materials in the kinemage format should send E-mail to jsr@kinemage.biochem.duke.edu.

A questionnaire asking students whether they thought the graphics component was worthwhile and interesting always gets a positive response, because the molecules are elegant and the exercises are fun to do. Of more significance is the fact that many students, especially those previously unfamiliar with molecular graphics, tell us that these exercises help them greatly in understanding papers or seminars with a structural-biology component.

In summary, we do believe that the use of interactive molecular graphics makes a unique and important contribution to student learning of biochemistry and molecular biology at any level. For best effectiveness, however, the graphics should primarily be worked through by individual students at their own pace, answering questions whose timely completion is monitored although not necessarily graded. Molecular graphics use in biochemistry teaching has two complementary goals: to enhance learning of the subject matter and to develop analytical skills of a new sort. The relative emphasis between those two goals shifts depending on the level and timing of a course, but both should always be present. With the variety and quality of graphics-based teaching materials, software, and web-sites available now, it is not hard to put together presentations and exercises suitably tailored to the needs and subject matter of a particular class.

REFERENCES

- [1] H. M. Berman, J. Westbrook, Z. Feng, G. Gilliland, T. N. Bhat, H. Weissig, I. N. Shindyalov, P. E. Bourne (2000) The Protein Data Bank, *Nucleic Acids Res.* **28**, 235–242.
- [2] Research Collaboratory for Structural Bioinformatics Protein Data Bank: www.rcsb.org/pdb.
- [3] D. C. Richardson, J. S. Richardson (1992) The kinemage: a tool for scientific communication, *Protein Sci.* **1**, 3–9.
- [4] Richardson Laboratory: kinemage.biochem.duke.edu.
- [5] R. Sayle (1992) Proceedings of the 10th Eurographics UK 1992 Conference, Abingdon Press, York.
- [6] H. J. Bernstein: www.bernstein-plus-sons.com/software/rasmol.
- [7] E. Martz: www.umass.edu/microbio/chime.
- [8] N. Guex, M. C. Peitsch (1997) SWISS-MODEL and the Swiss-Pdb Viewer: an environment for comparative protein modeling, *Electrophoresis* **18**, 2714–2723.
- [9] N. Guex, A. Diemand, M. C. Peitsch, T. Schwede: www.expasy.ch/spdbv.
- [10] D. Voet, J. G. Voet, C. W. Pratt. (1999) Fundamentals of Biochemistry, John Wiley & Sons, Inc., New York.
- [11] C. I. Branden, J. Tooze. (1999) Introduction to Protein Structure, 2nd ed., Garland Publishing, Inc., New York.
- [12] H. R. Horton, L. A. Moran, R. S. Ochs, J. D. Rawn, K. G. Scrimgeour. (1996) Principles of Biochemistry, Prentice Hall, Inc./Simon & Schuster, Upper Saddle River, New Jersey.
- [13] S. L. Weldon, M. A. Jones (1995) Kinemages as a visualization tool for biochemistry classes, *Biochem. Educ.* **23**, 208–212.
- [14] C. E. Sansom, D. A. Waller, A. J. Geddes (1996) Use of graphics workstations to illustrate protein and nucleic acid structure: a description of three modelling experiments carried out by second-year undergraduates, *Biochem. Educ.* **24**, 32–35.
- [15] S. W. Weiner, P. F. Cerpovicz, D. W. Dixon, D. B. Harden, D. S. Hobbs, D. L. Gosnell (2000) RasMol and Mage in the undergraduate biochemistry curriculum, *J. Chem. Educ.* **77**, 401–406.
- [16] C. S. Tsai J. (2000) Microcomputer applications in biochemistry, *Chem. Educ.* **77**, 219–221.
- [17] J. S. Richardson (1981) Anatomy and taxonomy of protein structures, *Adv. Prot. Chem.* **34**, 167–339.
- [18] J. S. Richardson (2000) Early ribbon drawings of proteins, *Nat. Struct. Biol.* **7**, 624–625.
- [19] D. C. Richardson, J. S. Richardson (1994) Kinemages-simple macromolecular graphics for interactive teaching and publication, *Trends Biochem. Sci.* **19**, 135–138.
- [20] J. S. Richardson, D. C. Richardson, in M. G. Rossmann, E. Arnold, Eds. (2001) International Tables for Crystallography, Vol. F, Kluwer Academic Publishers, Dordrecht, The Netherlands, pp. 727–730.
- [21] D. C. Richardson, J. S. Richardson (1989) in G. D. Fasman, Ed., Prediction of Protein Structure and the Principles of Protein Conformation, Plenum Press, New York, pp. 1–98.
- [22] C. C. Blake, M. J. Geisow, S. J. Oatley, B. Rerat, C. Rerat (1978) Structure of prealbumin: secondary, tertiary and quaternary interaction determined by fourier refinement at 1.8 Å, *J. Mol. Biol.* **121**, 339–356.
- [23] A. A. Arnone, C. J. Bier, F. A. Cotton, V. W. Day, E. E. Hazen, Jr., D. C. Richardson, J. S. Richardson, A. Yonath (1971) A high resolution structure of an inhibitor complex of the extracellular nuclease of *Staphylococcus aureus*, *J. Biol. Chem.* **246**, 2302–2316.
- [24] N. Ban, P. Nissen, J. Hansen, P. B. Moore, T. A. Steitz (2000) The complete atomic structure of the large ribosomal subunit at 2.4 Å resolution, *Science* **289**, 905–920.
- [25] S. Daopin, K. A. Piez, Y. Ogawa, D. R. Davies (1992) Crystal structure of transforming growth factor- β 2: an unusual fold for the superfamily, *Science* **257**, 369–373.
- [26] M. Elrod-Erickson, M. A. Rould, L. Nekludova, C. O. Pabo (1996) Zif268 protein-DNA complex refined at 1.6 Å: a model system for understanding zinc finger-DNA interactions, *Structure* **4**, 1171–1180.
- [27] J. M. Word, S. C. Lovell, J. S. Richardson, D. C. Richardson (1999) Asparagine and glutamine: using hydrogen atom contacts in the choice of side-chain amide orientation, *J. Mol. Biol.* **285**, 1735–1747.
- [28] J. M. Word, S. C. Lovell, T. H. LaBean, H. C. Taylor, M. E. Zalis, B. K. Presley, J. S. Richardson, D. C. Richardson (1999) Visualizing and quantifying molecular goodness-of-fit: small-probe contacts dots with explicit hydrogen atoms, *J. Mol. Biol.* **285**, 1711–1733.
- [29] W. Bode, E. Papamokos, D. Musil (1987) The high-resolution X-ray crystal structure of the complex formed between subtilisin Carlsberg and eglin c, and elastase inhibitor from the leech *Hirudo medicinalis*. Structure analysis, subtilisin structure and interface geometry, *Eur. J. Biochem.* **166**, 673–692.
- [30] Protein Society: www.faseb.org/protein/ProTeach.
- [31] R. Bateman: orca.st.usm.edu/~rbateman/kinemage.