Didactically suitable VMD presentations with support of Button control windows.

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VMD– presentations with radio button or text explanation buttons are included, that are particularly suitable for science fairs and for teaching.
Due to the utilization of buttons to control VMD – Presentations, the usability in such an environment is faciliated. The concept can also be applied in an e–learning environment. It is similar to the now outdated internet –plug–in Chime or Javascript or WebGl –based online viewers such as *Mol, JSMol or NGL , which are often used in a analogue manner. However the graphical presentation and the usage of VMD are superior to those viewers and the composition of the presentations is much more efficient. Furthermore, VMD is platform –independent and not bound to particular operating systems or even browsers, as it is the case with Chime.
For questions concerning VMD, you may consult the operating manuals [Caddigan et al.(2005), Humphrey et al.(1996), Stone et al.(2001)], which can be downloaded from the VMD -homepage. As far as the programming of the scripts and their installation is concerned, there are brief manuals available from my homepage [Schellenberg(2010)].

The particular protein presentations described here are part of the whole package `vmdscriptger.tar.gz` resp. `vmdscriptger.zip`, which contain many more scripts apart from the ones used here. For installation of the scripts, please read the installation text. Under Linux, there is also the script `vmdlehre.sh` available, that starts a button control window, from which you can load the belowmentioned presentations. Otherwise, you can start a presentation by loading the respective VMD –script in the program.

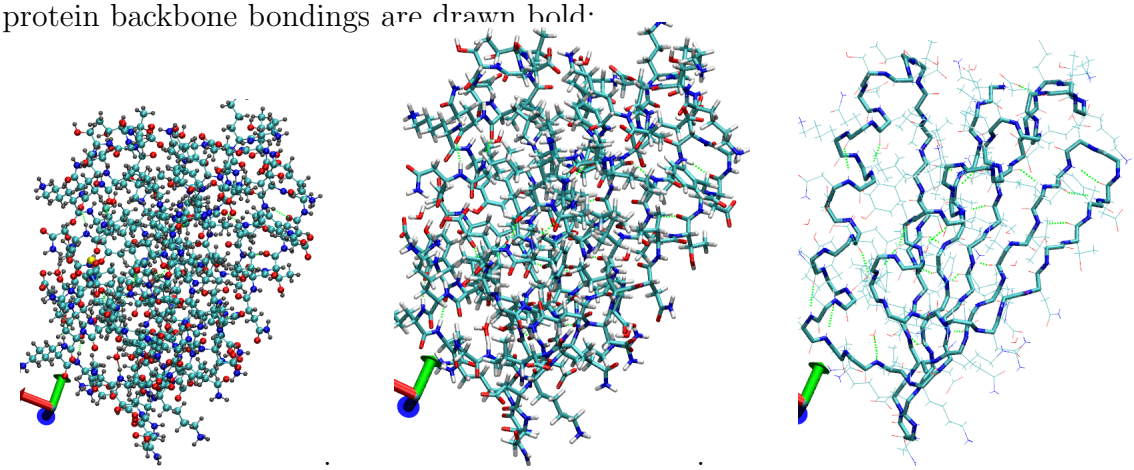
First, an introduction into abstract protein presentations is given on the example of a simple structured proteins prior to showing some particularly important protein classes. These are by name antibodies, which play an important role in the immune system, a vision pigment, the photosynthetic oxygen evolving system PSII and the ribosome, that translates the genetic conde intos the corresponding protein sequence.

Protein structure

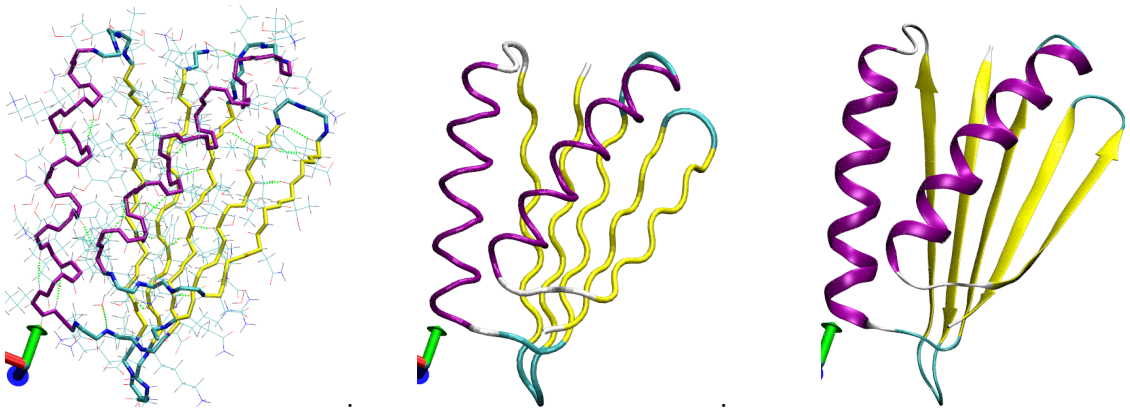
Skriptfile: designerproteinhypertxtengl.vmd

The structure of this protein had been calculated in advance, prior to producing the primary structure genetically. So far for the name designer protein. The theoretical predictions turned out to be surprisingly accuratate. Taking this simple and straightforward structured protein, it is demonstrated, how to get from the atomic structure model via the bonding model to the abstract protein presentations regularly used in biochemistry, such as tubes, ribbons and cartoons.

The left representation shows the atomic structure model, in which the atomic coordinates are taken from the coordinates in the pdb -file. In the middle this is altered to the bonding lines presentations, and on the right as a first step towords more abstract presentations, the protein backbone bondings are drawn bold:



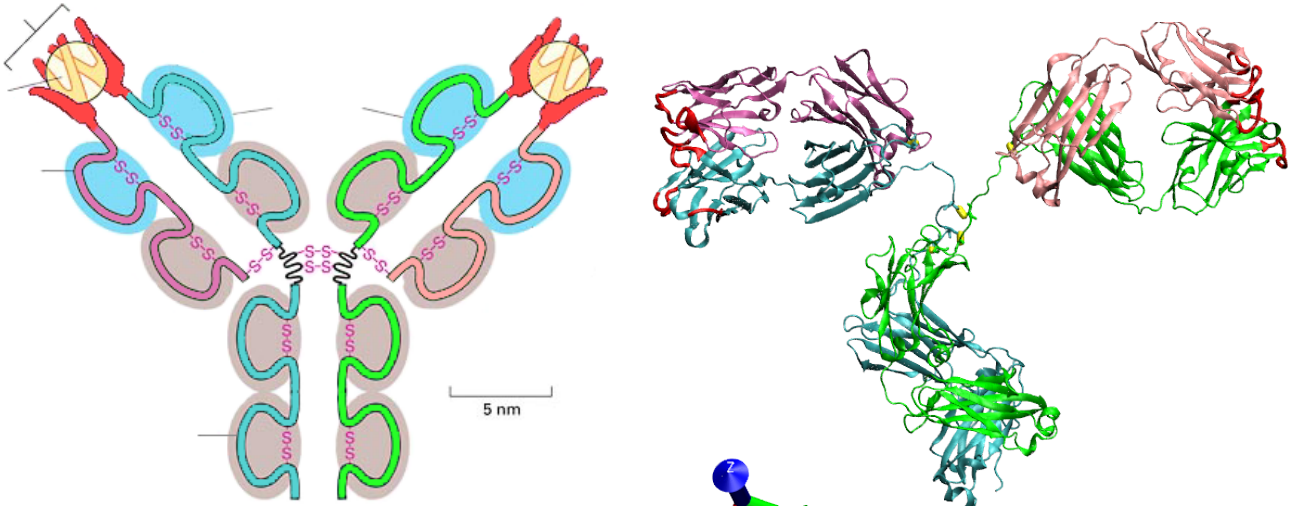
The increasingly abstract models are based on the backbone structure, with alpha –helix (violet), beta –sheet (yellow) and turns (cyan). The nonstructured parts are shown in white.



Antibodies

Scriptfile: antibodyhypertxtengl.vmd

Antibodies are the central proteins of the immune system. The protein consists of two heavy and two light chains, which are connected by disulfide bonds. On the tip of the antibody wings, the hypervariable reagions are located, which bind to certain structures (Epitopes) of the binding partner (Antigene) specifically. In the following pictures, a schematic drawing of the antibody structure is shown (left), and the corresponding abstract structure presentation in the same colors produced with VMD. The heavy chains are green and cyan, the light chains are violet and pink, and the loops of the hypervariable regions are depicted in red.

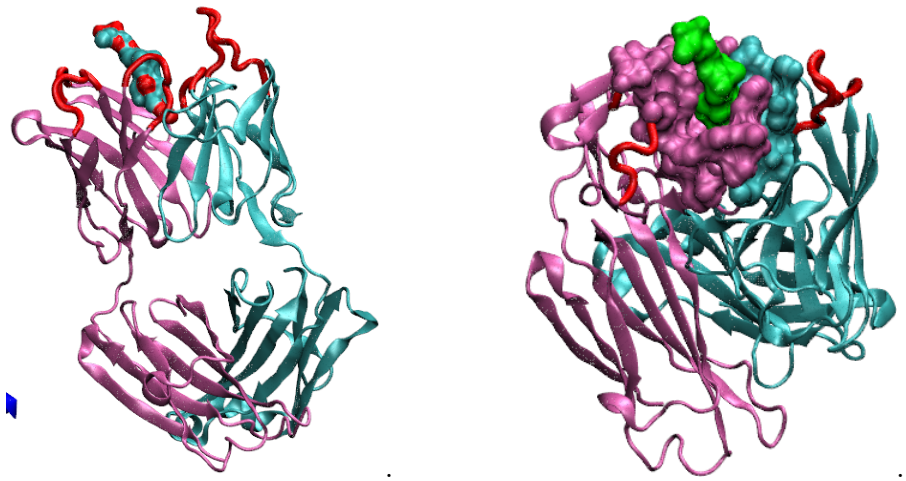


One can enzymatically cut the protein parts with the binding regions without loss of functionality. Thes protein parts are called FAB's. The disulfid bridges are also shown. In a further presentation, they are enhanced and the bridges in the hinge region, at the base of the FABs and in the individual fragments are distinguished by color.

The immunesystem produces antibodies against larger indruders like viruses and bacteria. The antibodies bind to particular locations of the intrudor, for example to certain enzymes on the surface of viruses. Small molecules or proteins are usually not attacked. However, one can produce antibodies against these systems for analytical purposes or for research applications. In this case, the molecule is bound to a large particle, that is intravenously utilized to an animal. The immunsystem identifies this particle as an intrudor and produces antibodies to structures on the surface of the particle, e.g. the bound molecules. This mechanism may also play a role in the occurance of allergic reactions. In this case the increase of microparticles in the environment due to civilisatory effects may be crucial. These particles may adsorb otherwise harmless substances from the environment.

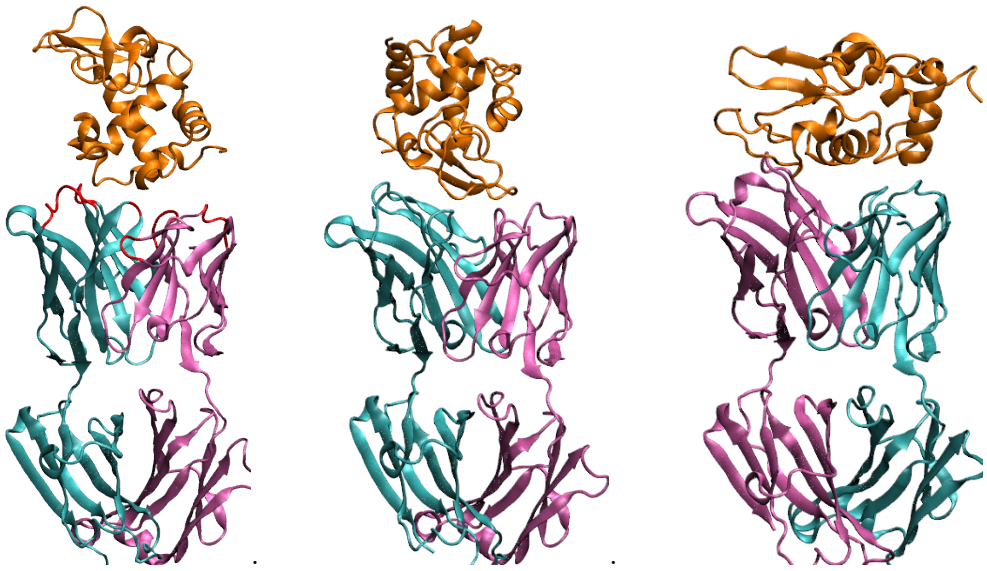
The presentations show antibody –antigen pairs produced in this manner. By far most of the analysed structures only deal with FAB –fragments, usually with the respective antigens. As an example for a small antigen, digoxigenin is chosen, which is a therapeutically

active steroid. This fit into a binding pocket, that is shaped by the hyervariable regions. Different presentations illustrate the shape of the binding pocket and the position of the digoxigenin.



As an immune response of the body, multiple different structures against one antigen are produced, first with low binding constant and specificity. In the course of the immune response, antibody forming cells are selectively activated that produce more specific and better binding antibodies. (clonal selection theory). Even at that stabe different antibodies exist, that bind differently to the antigen. In principle, every antibody producing cell (B –lymphocyt) forms an unique antibody. This is adventageous, since the indrudor is recognized via different positions, and can not become resistant due to an alteration in one position. The heterogenous mixture is referred to as polyclonal antibody. A standardization of such a mixture is difficult, since the immune response is individually specific for each host animal. I would be advantageous for many experiments to have a homogenous single antibody. For structure determination, this is crusial, since differently formed hypervariable regions and the differently binding antigens would lead to an overlap of the different structures, that would prevent structure determination in the crucially interesting parts of the antibody. In the 1980's a method was developed, how an individual antibody forming cell could be bred, therefore producing a large amount of an individual antibody (monoclonal antibody). To this end a B –lymphocyte is fusioned with a myeloma cell, and this hybridoma cell is selected and bred. In this manner, a large amount of monoclonal antibody can be produced. Since the cells can be frozen, one can produce the same type of antibody any time later.

As an example for an antibody for a large antigen, three different monoclonal antibodies to the protein lysozyme are shown. The lysozyme is colored in orange, the two chains of the antibody are cyan and violet. One can clearly distinguish the different epitopes, of lysozyme to which the antigen binds. An overlap of the three structures is also shown.



Apart from the naturally occuring principal structure of the antibodies, several other proteins where modified in a way to emulate antibodies. Particularly proteins, that already fulfill a binding function to specific molecule in nature, for example maltose binding protein or lipocalins, are genetically modified to bind to molecules of interest. As an example the script anticalin.vmd is included.

Photosynthesis

Skriptfile: psIIhypertxtengl.vmd

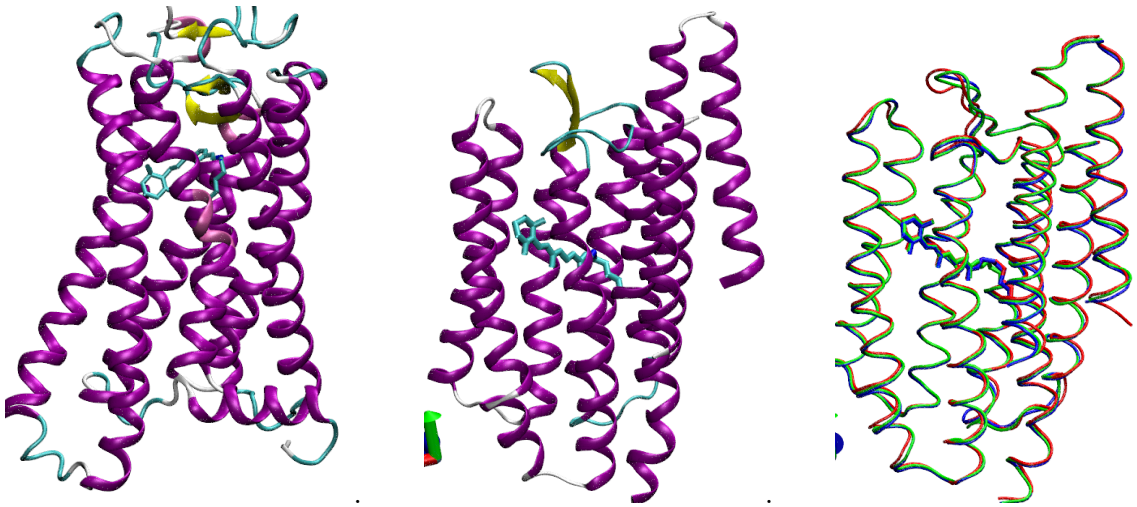
Photosystem II of blue algae (cyanobacteria) and of green plants is responsible for water splitting and as a consequence for oxygen evolution on earth. By far most of the atmospheric oxygen originates from this source. Since amino acids do not absorb in the visible range of the spectrum, chromophores like chlorophylls (green) carotenoids (orange) and pheophytins (blue) are bound to the protein. The protein complex consists of light harvesting proteins (CP-43, CP-47 etc.) and as a central part of the reaction center. In plants, the protein is embedded in the tylacoid membran which can be clearly seen from the many alpha–Helixes spanning the membrane.

The light harvesting pigments absorb the light and transfer the excitation energy to the heart of thew protein complex, the reaction center, whose pigments are drawn bold. The reaction center absorbs the light directly or gets the excitation energy from the antenna pigment. Following the excitation of the chlorophyll dimer (Special Pair, violet) an electron is transferred via a chlorophyll and a pheophytin chromophor to two quinones on the other side of the tylacoid membran. This electron flow produces an electrical potential accross the membrane, which can be used to drive biochemical reactions. However, to reduce water to oxygen, four redox equivalents are required, but only one redox equivalent is produced per excitation of the special pair. The manganese –oxygen –cluster serves as a redox depot by successive oxydation of the cluster from oxidation state zero to +IV. The four successive excitations of the special pair. The distance between the Mn–O –cluster and the special pair is in principal too large for an electron transfer, but a tyrosin from the protein backbone serves as transient electron carrier.

Vision proteins

Skriptfile: sensoryrhodopsinhypertxtanimation.vmd

Thze sensory proteins are responsible for the primary processes of vision Since the light reception has to happen with visible light, the protein contain a carotenoid chromophore, the opsin, which is connected kovalently to the protein backbone via a nitrogen (Schiff's base). The chromophore contains conjugated double bonds to shift the absorption to the visible. and reacts to light absorption with a photophysical process, a cis–trans isomerization around one of the double bonds. As in photosynthetic systems, the protein is embedded in a membran, and consists dominantly of alpha –helixes. The protein indroduced here is not the vision protein of mammals, but instead a similarly built light detection protein from an archaebacterium, the sensory rhodopsin. It is responsible for phototaxis (orientation of the bacterium to or away from light) This system is used here, since X–ray structures exist of transition states formed following excitation. By aligning the ground state structure and the transition state structures an animation is created that illustrates the principle of the functionality In a further presentation, the differently colored structures are superimposed. Although the protein is from an organism that even belongs to a different kingdom, the similarity to the vision pigments of mammals is surprising. In the VMD script collection you also find a structure for the vision pigment from bovine: bovinerhodopsin.vmd.



In the figures, the vision pigment of bovine is depicted on the left, followed by a comparable picture of the sensory rhodopsin and the superimposed structures of the ground state (green) and two transition states of sensory rhodopsin.

A major structural reorientation such as a cis–trans Isomerization of the chromophores involved and accordingly a strong reorganization of the protein lattice is important for light detection, since that creates a strong signal in the environment. In nature, there are multiple classes of chromophores that act correspondingly besides carotenoids, for example open chain tetrapyrolls in the plant and cyanobacterial light sensor phytochrome. Due

to its simplicity the Photoactive Yellow Protein (PYP), that contains a cinnematic acid derivative, is particularly well researched and gave useful information for lgiht detection in nature (see pypanimation.vmd). Another mechanism, that is correlated to a large change of the chromophore bonds, is the plant light sensing protein phototropin, that contains a flavine molecule, see . Ein anderer Mechanismus, der mit einer starken Änderung der Farbstoffbindungen einhergeht, see phototropinanimation.vmd.
In contrast to the strong rearrangements of the chromophore environment, it is crucial in photosynthesis to use up as little energy as possible. Therefore the favored chromophores of photosynthesis are rigid ring systems such as chlorophyll and pheophytin, whose structure is barely altered upon excitation.

Ribosome

Skriptfile: ribosomebutton.vmd

the ribosome is a protein -RNA -complex, which is responsible for the translation of the genetic code into the amino acid sequence of the protein The complex consists of protein parts (blue) as well as of ribosomal RNA, (r-RNA), namely the 16s r-RNA (olive), the 24s r-RNA (ocre) and the 5s-RNA. Not only the protein acts as a catalyst, but the r-RNA (ribozyme) as well. The ribosome consists of two subunits. Similar to the function of a tape recorder, the messenger –RNA (m-RNA) is read by transfer –RNA (t-RNA) while slipping through the gap between the two subunits. In the presentation the two subunits as well as three t-RNAs and a piece of m-RNA about to be translated are shown. There exists at least one t-RNA for each amino acid, which is responsible for the translation of the nucleotid codon corresponding to the amino acid into the peptid sequence. The nucleic acid triplett is located on one end of the t-RNA, while on the other end, the RNa is loaded with the respective aminoacid, that is bound to the end of the emerging peptid chain.

Unlike the the usually plain structure of DNAs, r-RNA as well as t-RNA show a complex three dimensional structure, which is mostly due to the additional –OH group on the ribose sugar. The structure of ribosomes and of t-RNA as well as the genetic code is universal, which suggests, that these systems were also present in the last common ancester of all living beings, prior the divergence into Archae, Bacteria and Eucariotes. The r-RNA is an important part of the ribosome complex, in structural as well as catalytic respect. Such an RNA is also called a Ribozyme. It is assumed that the present function of DNA (information storage) and protein (maschinery) was first both fulfilled by RNA (RNA –world).

For a detailed view of t–RNA, there are separate scripts available: ElongFactortRNAbutton.vmd and aspartylsynthetase.vmd as well as the script DielsAlderribozymebutton.vmd as an example for an artificially produced and selected Ribozyme.

Acknowledgement

The work extensively uses the program 'Visual Molecular Dynamics' from the University of Illinois at Urbana-Champlain, and is available free of charge for science and education. I am grateful to the team that develops VMD and to the participants of the VMD mailing list, in particular to John Stone for support.

Many thanks to those who tested the scripts, made contributions or otherwise supported my teaching activities, by name César Bernardo, Christopher Bruhn, Wolfgang Fritzsche, Hugo Gonçalves, Matthias Görlach, Karl Otto Greulich, Paulius Grigaravicius, Ayman Al Remawi und Sven Peters. I am grateful for the feedback, that the feedback from the students of the lecture 'Biomolecules: Databanks, visualizations and computations, which help much to improve the lecture.

References and Notes

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[Stone et al.(2001)] John Stone, Justin Gullingsrud, Paul Grayson, & Klaus Schulten (2001). 'A system for interactive molecular dynamics simulation'. In John F. Hughes & Carlo H. Séquin (eds.), *2001 ACM Symposium on Interactive 3D Graphics*, pp. 191–194, New York. ACM SIGGRAPH.