Structural biology resources

Compiled by the bumbling biochemist (Bri Bibel), last updated 08/06/26
Link to page where you can download: https://bit.ly/structural_biology

Educational Resources

My stuff:

- Here are some key posts:
  - Structural biology overall: https://bit.ly/structural_biology
  - X-ray crystallography, start here: http://bit.ly/xraycrystallography2
  - Cryo-EM: http://bit.ly/cryoemxray
  - Using and interpreting the results of AlphaFold: https://thebumblingbiochemist.com/365-days-of-science/interpreting-alphafold-predictions/
  - Resolution: https://bit.ly/structure_resolution
  - Databases: https://bit.ly/databases_guide
  - Intro to PDB, crystal structure entries, crystal contents, resolution: https://bit.ly/pdbstructures
  - Tools for working with proteins: https://thebumblingbiochemist.com/365-days-of-science/proteintools/
  - UniProt & ProtParam: https://bit.ly/uniprotprotparam
  - Protein alignments: https://bit.ly/proteinalignments
  - HDX-MS (hydrogen-deuterium exchange mass spectrometry): http://bit.ly/hdxmassspec
  - Molecular dynamics: https://thebumblingbiochemist.com/365-days-of-science/dynamics/
  - Structure-aided design: https://bit.ly/structureaideddesign
  - H++: https://bit.ly/hplusplus

- My structural biology YouTube playlist: https://youtube.com/playlist?list=PLUWsCDtjESrGhwVxsRbTJdL-BEsN60RCs

- My x-ray crystallography YouTube playlist: https://www.youtube.com/playlist?list=PLUWsCDtjESrHIGzgUctYRWSWm-lw9_VhX

- My PDB YouTube playlist: https://youtube.com/playlist?list=PLU1mvOopp1Ik&si=LLBBOZkQfvOX-YIe

- My PyMOL YouTube playlist: https://www.youtube.com/playlist?list=PLUWsCDtjESrG-46VDvPZfYPQyyXhZbGCN

More official stuff:

If you are new to structural biology, start with PDB101!

The PDB based out of the US is the Research Collaboratory of Structural Bioinformatics (RCSB) PDB has a great website called **PDB-101**: http://pdb101.rcsb.org/

If you go to this page in the "learn" tab, you'll find this Introduction to PDB Data: http://pdb101.rcsb.org/learn/guide-to-understanding-pdb-data/introduction as well lots of pages about the basics of crystallography and crystal structures.

They also have lots of cool resources for teachers and learners, including downloadable infographics and posters and make-it-yourself paper models. I used the GFP model when I taught a summer camp lesson: https://bit.ly/gfpfunscience Structural biology can be kinda hard to get into, so they have ways to lure you in, like their "Molecule of the Month" series. Each month they feature a different molecule or class of molecules, tell you cool stuff about them, and walk you through some structures.

X-ray Crystallography

Books:

- Introduction to Macromolecular Crystallography, Second Edition, Alexander McPherson First published:11 March 2008.Print ISBN:9780470185902 |Online ISBN:9780470391518 |DOI:10.1002/9780470391518
    - I had the pleasure of sitting in on the CSHL crystallography course twice and learning directly from Dr. McPherson (and a who's who list of great crystallographers!). This book is really good and explains things in a comprehensive way that's still comprehendible! It doesn't get too into the math and instead helps you get a more intuitive sense of what's going on. Highly recommend.
- Crystallography Made Crystal Clear: A Guide for Users of Macromolecular Models, Gale Rhodes. ISBN 0080455549, 9780080455549 https://books.google.com/books/about/Crystallography_Made_Crystal_Clear.html?id=rwnR6qvaWgkC&source=kp_book_description
    - This is a more "meaty" book for those interested in really getting it down.

Articles:

Overviews of entire process:

- Wlodawer, A.; Minor, W.; Dauter, Z.; Jaskolski, M. Protein Crystallography for Aspiring Crystallographers or How to Avoid Pitfalls and Traps in Macromolecular Structure Determination. *The FEBS Journal* **2013**, *280* (22), 5705–5736. https://doi.org/10.1111/febs.12495.

Crystallization:

- McPherson, A.; Gavira, J. A. Introduction to Protein Crystallization. *Acta Crystallographica Section F: Structural Biology Communications* **2014**, *70* (1), 2–20. https://doi.org/10.1107/S2053230X13033141.

    - One of my favorites: it provides a comprehensive overview of crystallization in theory (solubility curves, etc.) and practice (techniques, precipitants, strategies, etc.)

- Gorrec, F. A Beginner's Guide to Macromolecular Crystallization. *Biochem (Lond)* **2021**, *43* (1), 36–43. https://doi.org/10.1042/bio_2020_108.
    - A lighter read. It has good information about mosaicity, etc.

- Calero, G.; Cohen, A. E.; Luft, J. R.; Newman, J.; Snell, E. H. Identifying, Studying and Making Good Use of Macromolecular Crystals. *Acta Crystallogr F Struct Biol Commun* **2014**, *70* (Pt 8), 993–1008. https://doi.org/10.1107/S2053230X14016574.

- Luft, J. R.; Wolfley, J. R.; Snell, E. H. What's in a Drop? Correlating Observations and Outcomes to Guide Macromolecular Crystallization Experiments. *Crystal Growth & Design* **2011**, *11* (3), 651–663. https://doi.org/10.1021/cg1013945.

- Budziszewski, G. R.; Snell, M. E.; Wright, T. R.; Lynch, M. L.; Bowman, S. E. J. High-Throughput Screening to Obtain Crystal Hits for Protein Crystallography. *J Vis Exp* **2023**, No. 193, 10.3791/65211. https://doi.org/10.3791/65211.

- Benvenuti, M.; Mangani, S. Crystallization of Soluble Proteins in Vapor Diffusion for X-Ray Crystallography. *Nat Protoc* **2007**, *2* (7), 1633–1651. https://doi.org/10.1038/nprot.2007.198.
    - This is a protocol paper with good tips and practical advice.

- Bergfors, T. Succeeding with Seeding: Some Practical Advice. In *Evolving Methods for Macromolecular Crystallography*; Read, R. J., Sussman, J. L., Eds.; NATO Science Series II: Mathematics, Physics and Chemistry; Springer Netherlands: Dordrecht, 2007; Vol. 245, pp 1–10. https://doi.org/10.1007/978-1-4020-6316-9_1.
    - For those interested in seeding methods and how to use them.

Data analysis:

- "Protein crystallography for non-crystallographers, or how to get the best (but not more) from published macromolecular structures" by Alexander Wlodawer, Wladek Minor, Zbigniew Dauter, and Mariusz Jaskolski. https://febs.onlinelibrary.wiley.com/doi/full/10.1111/j.1742-4658.2007.06178.x
    - This is probably one of my most-reread articles. It does a great job explaining the basics of how to go about critically interpreting the quality of protein crystal structures, what to keep an eye out for, and what the basic statistics mean.

- Lamb, A. L., Kappock, T. J., & Silvaggi, N. R. (2015). You are lost without a map: Navigating the sea of protein structures. Biochimica et biophysica acta, 1854(4), 258–268. https://doi.org/10.1016/j.bbapap.2014.12.02

- Shabalin, I. G.; Porebski, P. J.; Minor, W. Refining the Macromolecular Model - Achieving the Best Agreement with the Data from X-Ray Diffraction Experiment. *Crystallogr Rev* **2018**, *24* (4), 236–262. https://doi.org/10.1080/0889311X.2018.1521805.

- Fu, Z.-Q.; Geisbrecht, B. V.; Bouyain, S.; Dyda, F.; Chrzas, J.; Kandavelu, P.; Miller, D. J.; Wang, B.-C. I/σI vs {Rmerg, Rmeas, Rpim, CC1/2} for Crystal Diffraction Data Quality Evaluation. bioRxiv February 2, 2025, p 2024.12.10.627855. https://doi.org/10.1101/2024.12.10.627855.

Websites:

- Resources for Readers of Crystallography Made Crystal Clear https://spdbv.vital-it.ch/TheMolecularLevel/CMCC/index.html
    - This goes along with that book, but you don't need the book to appreciate it. It has links to TONS of useful software programs and articles for learning about crystallography

- Protein Crystallography Course, Randy Read. https://www-structmed.cimr.cam.ac.uk/course.html
    - Clear visuals & explanations.

- Hampton Research Crystal Growth 101 https://hamptonresearch.com/crystal-growth101.php
    - Great practical advice for growing quality crystals. You can download individual PDFs covering specific techniques or a compilation of them all. All free.

- Terese Bergfors tutorials https://xray.teresebergfors.com/tutorials/
    - Lots of examples of different outcomes of crystallization experiments and how to interpret them and make use of them.

Lectures:

- If you want to get hard-core into the nitty gritty of protein crystallography, Dr. Andrea Thorn has a great YouTube series of videos, "Basics of Macromolecular Crystallography": https://youtube.com/playlist?list=PLHHBmgJ8vFm6xZPlWlRGuBaoOM3OGlN5T
    - It's by teachers of CSHL's crystallography course and is great

- SBGrid is a consortium that labs can join that provides structural biology software. They also have weekly lectures that are free to watch on YouTube. They often have talks by software developers that can be very useful. https://www.youtube.com/channel/UCbo1TnKiXGtkE_R5b526JmQ

Other:

- The CCP4 bulletin board (CCP4bb) is a great place to keep up to date and make connections through the listserv, as well as ask questions and search through answers to questions people have already asked. https://www.ccp4.ac.uk/the-ccp4-bulletin-board/

- CSHL Crystallography Course. This hands-on course brings in experts from the field as incredibly comprehensive. It is competitive and expensive, however.

Cryo-electron microscopy (cryo-EM)

Virtual course:

- EM-University has hours and hours of content and resources. Basically a free full course (minus the hands-on stuff of course) with lectures by Prof. Grant Jensen (Caltech): the lectures are also on YouTube: https://em-learning.com/?CID=2019-LS-SPA-EMLearning&utm_source=Comms-Blog&utm_medium=EM+Blog&utm_campaign=2019-LS-SPA-EMLearning&utm_term=speak-to-an-expert-2

Articles:

- Milne, J. L., Borgnia, M. J., Bartesaghi, A., Tran, E. E., Earl, L. A., Schauder, D. M., Lengyel, J., Pierson, J., Patwardhan, A., & Subramaniam, S. (2013). Cryo-electron microscopy--a primer for the non-microscopist. *The FEBS journal*, *280*(1), 28–45. https://doi.org/10.1111/febs.12078
    - Highly recommend this review article: some of it's a little dated now, but it does a great job of explaining lots of things

- Cheng, Y., Grigorieff, N., Penczek, P. A., & Walz, T. (2015). A primer to single-particle cryo-electron microscopy. *Cell*, *161*(3), 438–449. https://doi.org/10.1016/j.cell.2015.03.050
    - Another good review

- Cheng Y. (2018). Single-particle cryo-EM-How did it get here and where will it go. *Science (New York, N.Y.)*, *361*(6405), 876–880. https://doi.org/10.1126/science.aat4346
    - For a historical perspective

- Lyumkis D. (2019). Challenges and opportunities in cryo-EM single-particle analysis. *The Journal of biological chemistry*, *294*(13), 5181–5197. https://doi.org/10.1074/jbc.REV118.005602

- D'Imprima, E., & Kühlbrandt, W. (2021). Current limitations to high-resolution structure determination by single-particle cryoEM. Quarterly reviews of biophysics, 54, e4. https://doi.org/10.1017/S0033583521000020

- Beckers, M., Mann, D., & Sachse, C. (2021). Structural interpretation of cryo-EM image reconstructions. Progress in biophysics and molecular biology, 160, 26–36. https://doi.org/10.1016/j.pbiomolbio.2020.07.004
    - Great at explaining how resolution is determined for cryo-EM

- Marques, M. A., Purdy, M. D., & Yeager, M. (2019). CryoEM maps are full of potential. Current opinion in structural biology, 58, 214–223. https://doi.org/10.1016/j.sbi.2019.04.006
    - This helps explain some of the similarities and differences between cryo-EM & crystallography data

- Ortiz, S.; Stanisic, L.; Rodriguez, B. A.; Rampp, M.; Hummer, G.; Cossio, P. Validation Tests for Cryo-EM Maps Using an Independent Particle Set. *Journal of Structural Biology: X* **2020**, *4*, 100032. https://doi.org/10.1016/j.yjsbx.2020.100032.
    - This has information on how/why FSC cut-offs are made

Lectures:

- "Single Particle Cryo-EM", Yifan Cheng, iBiology https://www.ibiology.org/biophysics/single-particle-cryo-em/

- Prof. Grant Jensen, "Getting Started in cryo EM", Caltech: YouTube playlist with hours and hours of content: goes really deep into the details: https://youtube.com/playlist?list=PL8_xPU5epJdctoHdQjpfHmd_z9WvGxK8-

- SBGrid has lots of great ones too: https://www.youtube.com/channel/UCbo1TnKiXGtkE_R5b526JmQ

Websites:

- *The FSC and Gold-Standard Refinement | CryoSPARC Guide*. https://guide.cryosparc.com/the-fsc-and-gold-standard-refinement (accessed 2026-07-26).
    - This is really helpful for understanding FSC curves.

- EMDB Validation Analysis Help: https://www.ebi.ac.uk/emdb/va/help/

Software:

- **THE PDB**!!!! This is where all the structures are deposited. https://www.rcsb.org/
    - More about the PDB and structures: https://bit.ly/pdbstructures
    - A Guide to PDB Structures and the RCSB PDB (a chapter of a free online book I wrote and talk about more below): https://utexas.pressbooks.pub/molviz-education/chapter/a-guide-to-pdb-structures-and-the-rcsb-pdb/

- **SBGrid** is a consortium that labs can join that provides structural biology software. They also have weekly lectures that are free to watch on YouTube. They often have talks by software developers that can be very useful. https://www.youtube.com/channel/UCbo1TnKiXGtkE_R5b526JmQ
    - One of my favorites is on using ChimeraX's augmented reality features! SBGrid Consortium talk, "Making Augmented Reality Videos with ChimeraX", by Tom Goddard, University of California, San Francisco, Resource for Biocomputing, Visualization, and Informatics https://youtu.be/g80hxLO1SVk and you can see my post on that for more: blog: https://bit.ly/chimeraxwebcam ; YouTube: https://youtu.be/E42aImuftCw

- For visualizing and making pretty graphics of protein structures, there are 2 main tools, ChimeraX & PyMOL. ChimeraX is fully free for academic use. PyMOL has a free version with slightly limited functionality and academic labs often have a license for the full thing.

    - **PyMOL**: https://PyMOL.org/ (paid (incentive) version) – free for educational use & https://github.com/schrodinger/pymol-open-source (totally free (but more limited) open-source version)
        - More here: https://bit.ly/PyMOLintro & https://youtu.be/CeTTmm5DI_k & https://www.youtube.com/playlist?list=PLUWsCDtjESrG-46VDvPZfYPQyyXhZbGCN
        - Consult their wiki for help: https://pymolwiki.org/

    - **ChimeraX:** https://www.cgl.ucsf.edu/chimerax/
        - Consult their wiki for help: https://www.rbvi.ucsf.edu/chimerax/tutorials.html

    - Note: less powerful, but convenient, browser-based alternatives include iCN3D & Mol*
        - **iCN3D:** https://www.ncbi.nlm.nih.gov/Structure/icn3d/full.html
        - **Mol*:** https://molstar.org/
        - It has a stand-alone browser version and is also integrated into the PDB and UniProt

    - Here's an OER I helped write showing how to do different basic tasks in the different software tools: Acevedo, R., & Procko, K. (Eds.). (2025). Seeing the Invisible: Learning to Teach with Biomolecular Visualization. The University of Texas at Austin. https://doi.org/10.15781/m4zd-7691

Software programs for when actually solving structures:

- **Phenix:** Great for x-ray crystallographic phasing, model building and automated refinement. http://www.phenix-online.org/ They also have some great tutorials on YouTube: https://www.youtube.com/c/phenixtutorials
- **Coot:** This is the software I use for manual refinement https://www2.mrc-lmb.cam.ac.uk/personal/pemsley/coot/ This cheatsheet is a definite must! https://www2.mrc-lmb.cam.ac.uk/personal/pemsley/coot/web/docs/crib-sheet.pdf
- Those are integrated with each other, along with **MolProbity** which tells you about things like clashes and awkward geometry https://molprobity.biochem.duke.edu/
    - MolProbity is also used as part of PDB Validation
- **CryoSparc:** for some of the steps of cryo-EM processing; they also have great tutorials https://cryosparc.com/

Tools for predicting structures and/or structural features, designing constructs, etc.

- **AlphaFold:** Predicts the structure of proteins, nucleic acids, and complexes. The server has more limited options than the stand-alone (GitHub) and ColabFold (Python notebook-based) versions, but uses the latest version of AlphaFold https://alphafoldserver.com/
    - More here: https://thebumblingbiochemist.com/365-days-of-science/interpreting-alphafold-predictions/
    - And for more, I encourage you to check out EMBL-EBI's online tutorial, "AlphaFold: A practical guide" by Paulyna Gabriela Magana Gomez and Oleg Kovalevskiy: https://www.ebi.ac.uk/training/online/courses/alphafold/
- **Institute for Protein Design Software** including **RFdiffusion** – can design new protein backbones (basically, the skeletons of proteins); **ProteinMPNN** – can predict sequences likely to fold into specific shapes (such as those predicted by **RFdiffusion**); **RoseTTAFold** – "classical" structure from sequence; **FoldIt** – crowdsourced protein folding game https://www.ipd.uw.edu/software/
- **SWISS-MODEL:** I used this for homology modeling in undergrad. It's good for when there's a solved structure of a highly similar protein to one you want to know the structure of (such as if there's a structure of the mouse one but you're interested in the human one) https://swissmodel.expasy.org/
- **HawkDock:** Models protein-protein interactions and analyzes strength of interactions at a residue level basis (for either predicted structures or already-prepared PDB files) https://cadd.zju.edu.cn/hawkdock/
- **MPI Bioinformatics Toolkit:** this has lots of tools for structural prediction, homology finding, etc. https://toolkit.tuebingen.mpg.de/
- **Crystallisation Construct Designer2 (CCD^2):** helps you design protein constructs to attempt to crystallize - good for figuring out where to make truncations & stuff (e.g. identify domain boundaries & remove predicted disordered regions at the end) https://ccd.rhpc.nki.nl/tutorial
- **CHARM-GUI:** for molecular dynamics (MD) https://www.charmm-gui.org/

    - Check out their “lecture” series of tutorials for help: https://www.charmm-gui.org/?doc=lecture

- **3D bio notes:** you put in the PDB code for a structure, then this site integrates a ton of information about the protein from various sources and lets you see sites of mutations, etc. in 3D. Also good for a quick overview or on the go look when you don’t want to open PyMol. http://3dbionotes.cnb.csic.es/ws

- **PDBSum:** Easy way to automatically generate protein topology & domain diagrams & investigate interfaces https://www.ebi.ac.uk/thornton-srv/databases/pdbsum/
    - Just upload or fetch a PDB file and it'll generate a bunch of things including: domain architecture diagram, secondary structure diagrams, topology map, diagram of and information about ligand-protein and protein-protein interactions (distances, types, etc.), information about pockets, tunnels, and channels.
    - *Note: Their server is down as of this writing, but there’s a stand-alone version you can download from GitHub.*
    - More here: https://youtu.be/AFmN4CKU1uE
- **HawkDock:** Models protein-protein interactions and analyzes strength of interactions at a residue level basis (for either predicted structures or already-prepared PDB files) https://cadd.zju.edu.cn/hawkdock/

- **H++:** Free web-based tool that takes a pdb file of a protein and predicts the protonation state of each amino acid residue at a pH of your choosing http://newbiophysics.cs.vt.edu/H++/index.php

- **Crystallisation Construct Designer2:** helps you design protein constructs to attempt to crystallize - good for figuring out where to make truncations & stuff (e.g. identify domain boundaries & remove predicted disordered regions at the end) https://ccd.rhpc.nki.nl/tutorial

- **MPI Bioinformatics Toolkit:** this has lots of tools for structural prediction, homology finding, etc. https://toolkit.tuebingen.mpg.de/

- **SWISS-MODEL:** I used this for homology modeling in undergrad. It’s good for when there’s a solved structure of a highly similar protein to one you want to know the structure of (such as if there’s a structure of the mouse one but you’re interested in the human one) https://swissmodel.expasy.org/

If you want to **find proteins that are similar**:
- *In sequence:*
    - **BLAST (specifically blastp)**: This searches protein databases for protein sequences. and takes into account both sequence identity (are the amino acids in the sequences identical at a given location) and similarity (are the amino acids in the sequences biochemically similar at a given location)
        - You can access it directly or through UniProt (“Tools” → “BLAST”) https://blast.ncbi.nlm.nih.gov/Blast.cgi?PROGRAM=blastp&PAGE_TYPE=BlastSearch&LINK_LOC=blasthome
- *In structure:*
    - **Foldseek**: It compares structures against one another to find similar ones. What’s even cooler is that it searches both experimentally-determined structures (housed in the PDB) and AI-predicted (AlphaFold Protein Structure DataBase (AFDB)) structures.
        - You can access the web version of it here: https://search.foldseek.com/search

        - Or the open-access code to run it in command line is here: https://github.com/steineggerlab/foldseek
        - It's also integrated into the AlphaFold database so you can get results there too https://alphafold.ebi.ac.uk/
    - **PDB Advanced Search Query Builder** https://www.rcsb.org/search/advanced
    - **ENDscript** https://endscript.ibcp.fr/ESPript/ENDscript/index.php
        - This will find sequences to align and also get their PDB codes and structural information

- If you're looking to create and/or visualize (interactively or for figures) **protein *sequence* alignments**:
    - For **creating sequence alignments:**
        - **Clustal Omega:** https://www.ebi.ac.uk/jdispatcher/msa/clustalo
            - You can also access it through the "Tools" → "Align" button in UniProt: https://www.uniprot.org/
    - For **visualizing sequence alignments interactively**:
        - **JalView**JS (web version) or JalView desktop version: https://www.jalview.org/jalview-js/JalviewJS/
        - For quickly comparing sequences in a clustal alignment and finding what number in one sequence corresponds to what number in another sequence, etc. here's a simple Jupyter/GoogleColab notebook: https://github.com/bribibel/clustal_comparer/blob/ab4cd7391b9add2857e20798d2da60c2f5caacfd/clustal_alignment_comparer.ipynb
    - For **making nice pretty sequence alignment figures**, including with overlying secondary structural information, etc.:
        - **ESPript** https://espript.ibcp.fr/ESPript/ESPript/index.php
    - More here: https://bit.ly/proteinalignments

- If you're looking to create and visualize **3D protein structural alignments**:
    - **RCSB PDB Pairwise Structure Alignment Tool** https://www.rcsb.org/alignment
        - You can choose from a variety of alignment methods based on how similar the proteins &/or their structures are. Choose a rigid method to directly superimpose the structures without altering them (good if very similar) or a flexible method (which breaks the structure up and aligns parts) for ones with large structural differences.
        - This will also allow you to download mmCIF files of them overlaid.
        - More here: https://youtu.be/MOBMBuAIAKo
    - **FoldMason MSA:** Unlike a pairwise alignment tool, FoldMason MSA does Multiple Sequence Alignment, finding the best alignment for a group of sequences, rather than comparing each structure to the same reference structure https://search.foldseek.com/foldmason

Hope that helps!