Molecular Modeling Project 2014:

Structure-based drug design

Homology modeling, molecular dynamics simulations, and docking

Background: G protein-coupled receptors

G protein-coupled receptors (GPCRs) are responsible for a large part of the signaling across the cell membrane and have received considerable attention in drug development. Drug discovery against these receptors have also been remarkably successful – close to 30% of all marketed drugs target GPCRs. GPCRs share a similar topology with seven transmembrane helices, but the extracellular ligands that activate these receptors range from small molecules to proteins. Breakthroughs during the last years have resulted in structures of almost a dozen structures of GPCRs. However, there are still hundreds of pharmacologically relevant GPCRs for which there are no high-resolution structures available. In this project, you will focus on modeling of monoaminergic GPCRs, which recognize neurotransmitters such as adrenaline, histamine, and serotonin. There are crystal structures several monoaminergic GPCRs, but not all. Your task is to model a receptor of unknown structure and predict the structure of that receptor in complex with one agonist and one antagonist.

General information & rules

• The project should be completed individually. You are encouraged to discuss ideas and general approaches you use, but not share results.
• An email to me (jens.carlsson@dbb.su.se) with a brief update of your current model of the receptor (pdb) and the next steps should be sent to me every Friday (Starting the 5 Dec, but the first time you only need to tell me which receptor you have selected!). You can also ask questions about the modeling. I will inspect the models and suggest on how it can be improved.
• An important part of the project is that you should write a short report (5 pages). It should be written as a scientific paper with a brief abstract summarizing your findings, an introduction, description of the methods and choices made, results, conclusions, and references. Obviously we don’t expect a full research paper, but do your best - it is good training for your future career!
• The course grade will be based both on the written exam (75 points) and this project (25 points). Both the form of the written report and the discussion about your results will matter.
• You are encouraged to hand in a preliminary version of your report by January 13, 2014. This will not be graded, but we will offer feedback and try to help you by pointing out things that might be incorrect or can be improved. We will provide feedback quickly, which gives you the chance to adjust your report to get more points.
• The deadline for handing in the final report is January 19, 2014. If you do not hand it in on time, you the maximum number of points you can get is 20. You have to pass to complete the course.

Modeling strategy

You can use any computational tool that you want to carry out this task, but we would recommend the two web servers GPCRModSim (http://gpcr.usc.es/) and DOCKblaster (http://blaster.docking.org/). The molecular dynamics program GROMACS could also be useful if you want to further refine your models.

1. Choice of receptor

First choose which GPCR to work on. You can choose any human aminergic GPCR except those that have a known structure, which you can check in the Protein Data Bank. You can find all the different families on the following web page:
http://www.guidetopharmacology.org/GRAC/GPCRListForward?class=A
The aminergic receptors that you want to model are:
- Adrenoceptors
- Dopamine receptors
- Histamine receptors
- 5-Hydroxytryptamine receptors
- Acetylcholine receptors

Don’t forget to read some literature about the receptors! You want to model a receptor that is pharmaceutically relevant.

2. Starting sequence
You will find the starting sequences for the human receptor in the Uniprot database (www.uniprot.org). At this point email me the Uniprot code for the receptor that you have chosen.

3. GPCRModSim – Alignment, structure prediction, and molecular dynamics.
The sequence alignment can be carried out using the GPCRModSim server (http://gpcr.usc.es/).
*Please register as a user on this site and start modeling! GPCRModSim is also the focus of one computer exercise.*

The most important part of a homology modeling project is to select a good template and to modify the sequence so that we get a good model of the region that we are interested in (the orthosteric binding site). This involves looking at what the sequence identity to the other available crystal structures and select a good template with high sequence identity >35%. Document your choices carefully and include the sequence identity between the target protein (your selected GPCR) and the chosen template (an available crystal structure) in the report. Look at the resulting structure in a viewer (e.g. PyMOL and VMD, which are available in the computer room) and compare it to the template by aligning the structures.

Examples of common problems:
- The sequence identity may be <35% to the closest template and then I would suggest that you switch to a different GPCR.
- The N and C terminal of the GPCR often have to be removed because they cannot be modeled with high accuracy. Simply remove them from the sequence and remodel the protein.
- The sequence identity may be very low in the loop, so it is sometimes a good idea to rebuild the extracellular loops, in particular if they are close to the binding site.
- The sequence identity may be very low in the loop, so it is sometimes a good idea to rebuild the extracellular loops, in particular if they are close to the binding site.
- The sequence alignment may have obvious errors.

4. Further refinement of model using molecular dynamics simulations
*[NOTE: We may have to skip this step this year due to limitations of the web-server. We will know this after Christmas – please avoid running MD on the server until then]*
GPCRModSim makes it possible to setup the receptor model in explicit membrane and simulate the structure using GROMACS on a web-server. After the molecular dynamics simulation exercise, you could also decide to run a longer simulation of the protein on your own computer.

5. Molecular docking of an agonist and antagonist to the model(s)
The DOCKblaster webserver makes it possible run dock small molecules to your receptor. This is also the topic of a computer exercise. You have to dock two molecules to the receptor: the endogenous agonist and an antagonist or inverse agonist (two molecules in total). Note that you may want to use different templates for the agonist bound and antagonist bound model. You can search for ligands to dock to your structures in the literature.