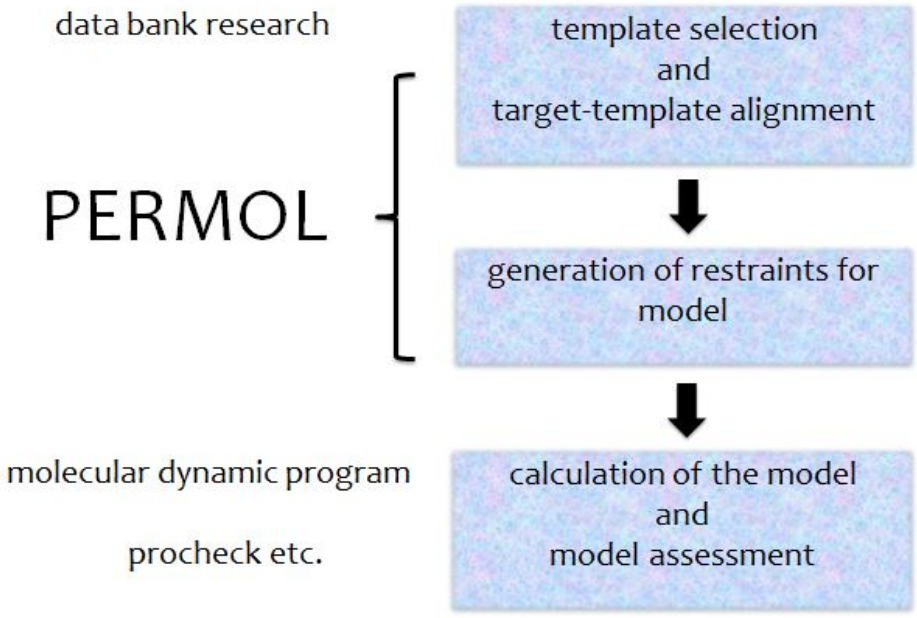
PERMOL

The program PERMOL [1,2] which is part of the software package AUREMOL was developed to extract information about the conformation of a known protein in the form of restraints – specifically distances between atoms, dihedral angles and H-bonds - and then transfer this information to an unknown protein. These restraints can be used for molecular dynamics as restrictions of the configuration space.

Workflow of Homology Modeling with PERMOL

PERMOL was originally developed for homology modeling, which is based on the observation that two proteins share the same folding pattern even if they match in only 20 % of their sequence. This technique implicates three steps: First alignment of the unknown protein to one or more models, then the generation of the restraints – here the user has a lot of possibilities e.g. the selection of the atom types or the upper und the lower limit of the distance between the atoms, which should be considered at generation, or the selection of only parts of the sequence - and finally calculation of the new structure with the molecular dynamic program CNS.



Workflow of Homology Modeling

Possible applications

We used PERMOL successfully to estimate the three dimensional structure of single protein domains and for complexes [3], too, in terms of homology modeling, but also for structure improvement [4] and for analysis of structural changes under variation of temperature and/or pressure. In addition PERMOL is a practical tool to find hydrogen bonds or to analyse atomic distances of a given structure.

Who to create restraints with PERMOL?

Step 1:

Use AUREMOL or download the standalone version of PERMOL! Start it!

Step 2:

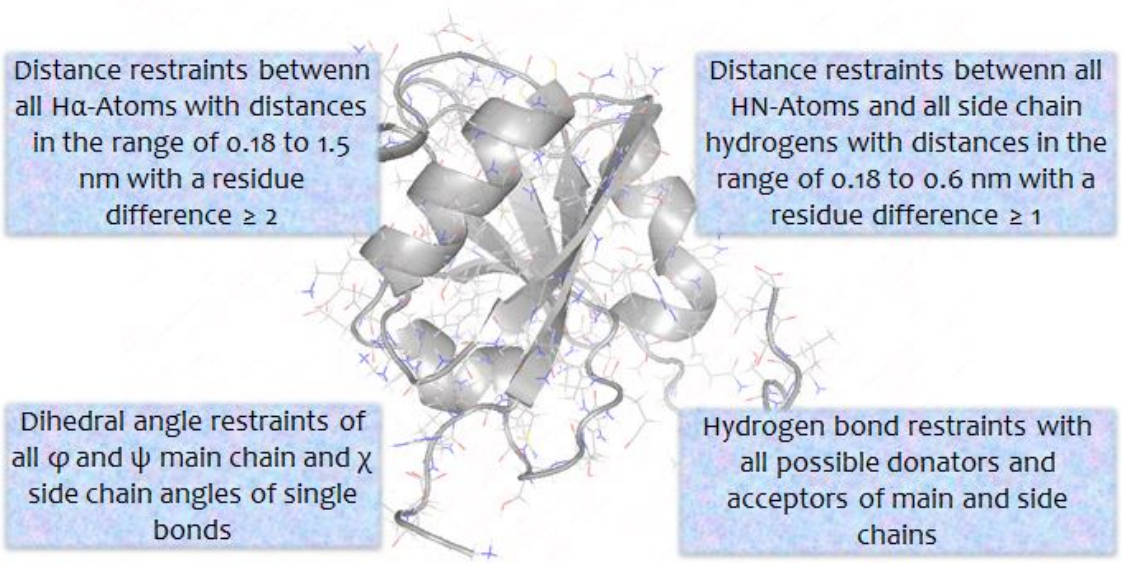
Feed target sequence(s) of the protein(s) you will get the model: One-letter-code or an AUREMOL sequence file or the possiblity to extract it from a PDB file.

Step 3:

Prepare the PDB files with your homologous proteins from which the informations will be extracted: Add hydrogens (if you will use XRAY-structures) and use IUPAC names for the atoms! Check the stereochemical nomination, too, because if there are mistakes you get structures with a lot of violations und big terms of energy!

Step 4:

By the generation of restraints you choose between the standard set of restraints (show picture) or a user defined one!



Proved and tested set of restraints

Step 5:

Choose a path for output!

Step 6:

After you have clicked the OK-Button the alignment will be shown. This alignment can be edited, but the sequences cant't variated at all: You can shift the gaps (-). Be careful that at the end of your editing the sequences must have the same length: The signs + at the beginning and the end of the sequences must align. In addition you have the option to exclude parts of the sequences from information generation.

Step 7:

Then the restraints will be generated.

Test Dataset

To test the restraints generation by PERMOL you can use our simple test dataset: Copy the sequence (one-letter-code) of the target (target.txt) into the dialog "Add sequence" (Step 2). Use model.pdb as homologous structure (Step 3).

And now?

Now you can use these restraints to calculate structures with CNS [5] (http://structure.usc.edu/cns/main/frame.html). Thereby you have to pay attention the following points:

- Because of the big number of generated restraints (much more than by experimental data analysis) you have to adapt the possible parameters in file *readdata* (NMR module) to read in all restraints. By the final waterrefinement according to Linge et al. [6] you have to change the parameter *nrestraints*.
- To optimize the weighting between the potentials which represent the a priori knowledge of proteins and these potentials which use the informations from the with PERMOL generated restraints by structure calculation you must decrease the parameters *md.hot.noe*, *md.hot.cdih*, *md.cool.noe*, *md.cool.cdih*, *md.cart.noe*, *md. cart.cdih*, *md.pow.noe* and *md.pow.cdih* in script *anneal.inp*. In the waterrefinement script these parameters have to change with the parameter *scale*. The right choice of parameters can be detected by small energies - especially of the energies which represent the physical and chemical properties of proteins like that of binding length between atoms or Van-der-Waals energies of the generated structures, what means that the structures will be formes, but not deformed by the PERMOL restraints.
- The time steps *md.hot.ss*, *md.cool.ss*, *md.cart.ss* and *md.pow.ss* must be smaller and - adapted to this - the number of iteration steps *md.hot.step*, *md.cool.step*, *md.cart.step* and *md.pow.step* have to increase. Elsewise it is possible that atoms „get lost“ by simulation, if the forces of these are too strong because of the big number of restraints, and then in one of the steps of iteration their new place could be out of the possible volume.

The new version of PERMOL unifies ideas from: Andreas Möglich, Daniel Weinfurtner, Till Maurer, Wolfram Gronwald, Josef Scheiber, Konrad Brunner, Carolina Cano, Michael Ebel, Bärbel Kieninger and Hans Robert Kalbitzer.

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Literature

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