IAP/Belspo P7/44 project : Integrative Protein Science (iPROS)

Structure predictions of membrane domains of proteins from the Divisome and BlaR

J.M. Crowet[1], N. Dony[2], B. Joris[2], R. Brasseur[1], L. Lins[1]

[1]*Centre de Biophysique Moléculaire Numérique, Gembloux Agro-Bio Tech, University of Liège, Gembloux, Belgium*
[2]*Centre d'Ingénierie des Proteins, Institut de Chimie B6a, University of Liège, Sart-Tilman, 4000 Liège, Belgium*

Abstract

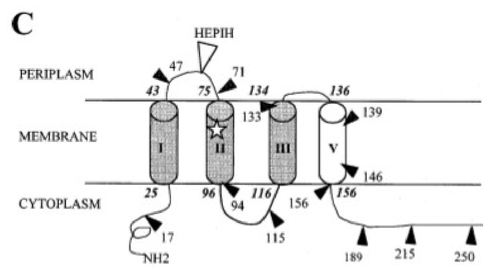
Except for MraY, for which a structure has been resolved in 2013, structures of the membrane domains of the proteins from the Divisome and BlaR are not known and there is no homolog proteins of known structure to build homolgy models. Although the structure prediction of membrane proteins seems easier than for globular proteins, the *ab initio* prediction of membrane protein structure remains a difficult task. Only few methods have been used and validated on experimental pdb structures. By using the MARTINI or Bond coarse grain representation, the multimerization of transmembrane helix has been carried out by molecular dynamics for the Glycophorin A and Influenza M2, and the structure of membrane protein has been predicted by the Rosetta and BCL packages. Here, the BCL Fold method for membrane proteins from the Meiler lab is used to predict the structure of the membrane embedded part of the politopic proteins from the divisome (FtsW, FtsK, FtsX and MraY) and BlaR.

1. Introduction

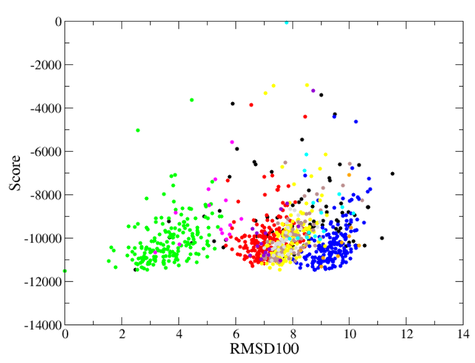
The BCL::MP-Fold method (Weiner 2013) rapidly assemble secondary structure elements (SSEs) based on a Monte Carlo protocol. 1000 structures are generated through a 6 stages process. Models are built into a static membrane object and scored according to a knowledge-based scoring function which account for the membrane environment. SSEs are generated using JUFO (Meiler 2001) and PSIPRED (Jones 1999) and TMs predicted by SPOCTOPUS are also used (Viklund 2008). A symmetry folding mode can predict homomultimers. This method has been shown to sample the correct topology in 34 of the 40 membrane protein tested. Here, as the native structure is not known, the RMSD scatter plot are computed regarding the model with the lowest score. Clusters are defined with the gromos method of the g_cluster tool (Lindahl 2001). In Weiner *et al*. 2013, they show that in most of the cases, the correct topolgy is sampled when the RMSD100 is lower than 8Å. Here, the cluster cutoff was lowered to 5Å. Moreover, in our computations, TMs experimentally determined topologies have been used instead of SPOCTOPUS because it gives us more accurate predictions and narrowed RMSD scattering plots. Good models have been predicted for BlaR and FtsK with this method.

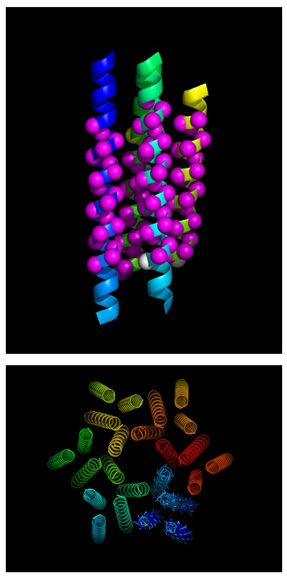
2. FtsK

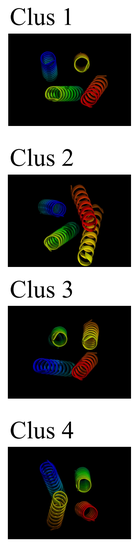
FtsK has 4 TMs (25-43, 75-96, 116-134, 136-156) whose topology has been validate experimentally and forms hexamers (Dorazi 2000). For the BCL prediction, the model 828 has the lowest score (-11532) and has been used to draw the RMSD scatter plot. The clustering show 4 clusters that represents 75% of the sampled structures and the middle structures have an average of 3.5 Å with other structures of the clusters. These are then good models for the FtsK structure. The experimental topology is represented by the magenta spheres and the hexameric form of the cluster 1 is also presented. The 4 clusters present different topologies but several interactions between helices are conserved (H3-H4, H2-H3, H1-H2, ...).

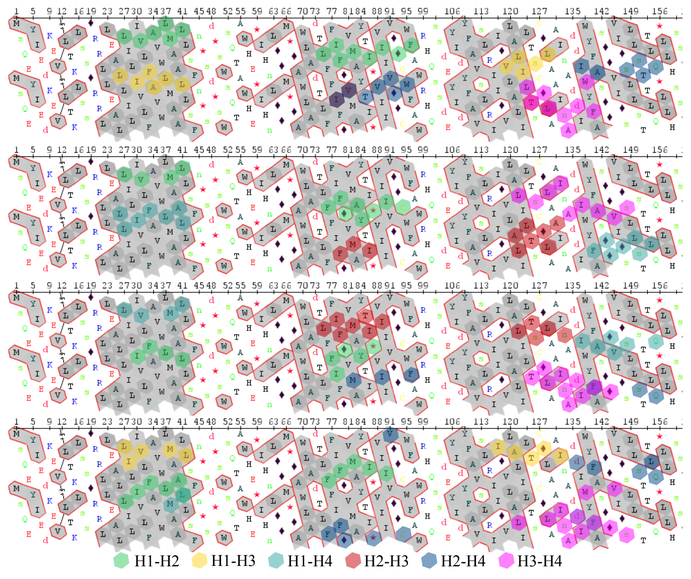


Clusters	Model N°	Nb (/1000)	Middle RMSD
Clus 1	29	202	3.63 Å
Clus 2	863	185	3.54 Å
Clus 3	198	184	3.47 Å
Clus 4	945	176	3.26 Å



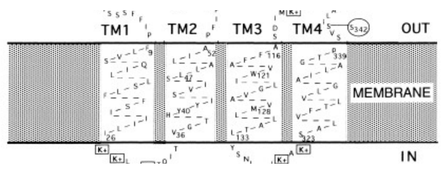




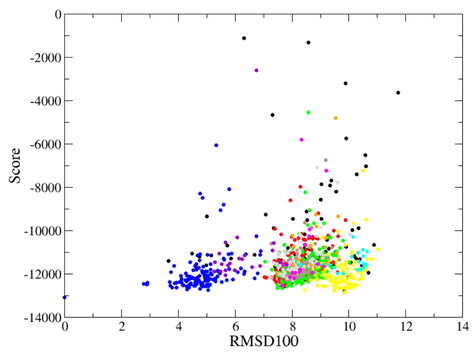


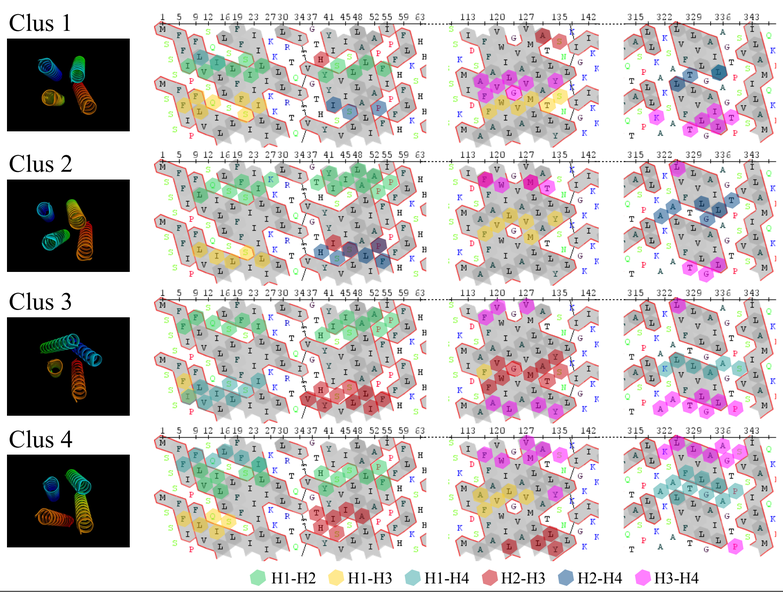
3. BlaR

BlaR has 4 TMs (7-27, 36-56, 115-135, 319–339) whose topology has been validate experimentally (Hardt 1997). For the BCL prediction, the model 781 has the lowest score (-13080). The clustering show 4 clusters that represents 71% of the sampled structures with a middle RMSD lower than 3.5 Å. These are then good models for the BlaR structure.



clusters	Model N°	Nb (/1000)	Middle RMSD
Clus 1	142	248	3.31 Å
Clus 2	891	227	3.46 Å
Clus 3	593	125	3.13 Å
Clus 4	248	108	3.31 Å

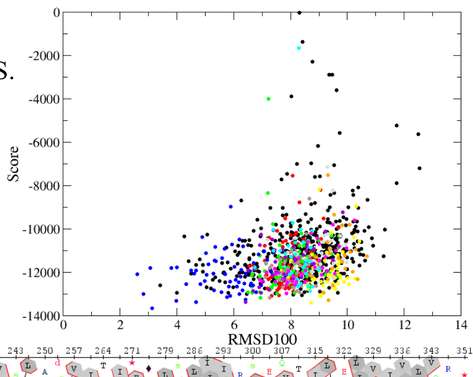


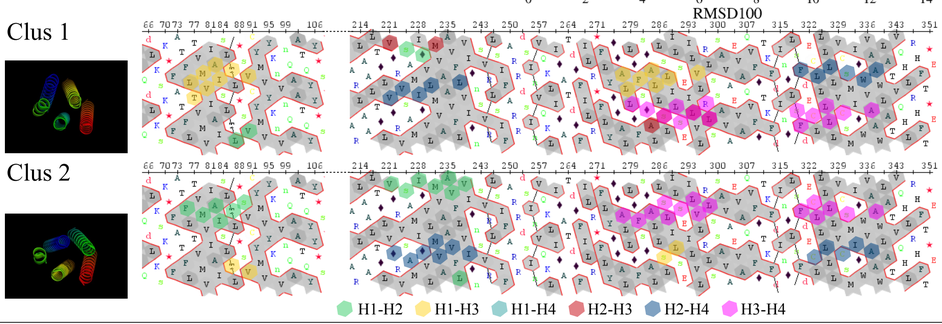


4. FtsX

FtsX has 4 potential TMs (75-95, 224–244, 276-296, 323-343). For the BCL prediction, the model 825 has the lowest score (-14026). The clustering show 2 clusters with more than 100 structures and a middle RMSD greater than 4 Å. From the scatter plot, we can assume that the native structure has been sampled but it is not possible from the clustering to define a good model. Tuning the TMs definition could give better results, instead of using SPOCTOPUS.

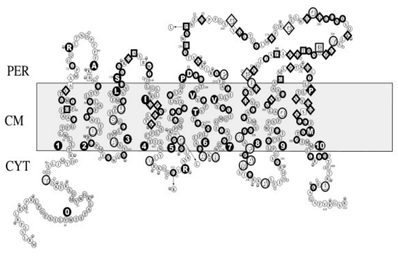
clusters	Model N°	Nb (/1000)	Middle RMSD
Clus 1	226	106	4.06 Å
Clus 2	39	101	4.07 Å
Clus 3	670	61	4.15 Å
Clus 4	82	56	3.92 Å

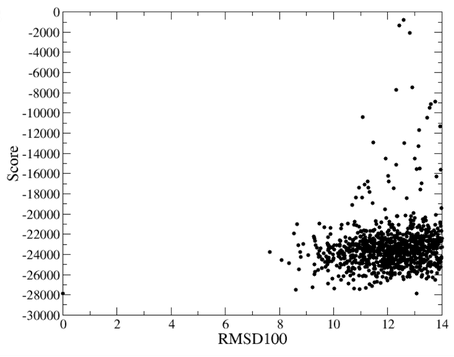




5. FtsW

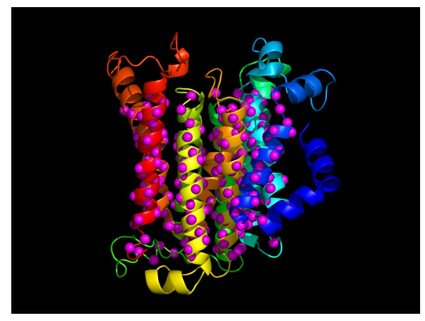
FtsW has 10 TMs (47-66, 86-104, 110-129, 141-162, 177-195, 200-216, 220-239, 307-324, 343-364, 374-394) whose topology has been validate experimentally (Lara 2002). For the BCL prediction, the model 118 has the lowest score (-27885) but the RMSD scatter plot show that the sampled structures are to distant from each other and no clusters can be defined.

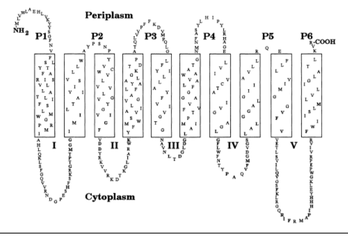




6. MraY

MraY has 10 TMs (22-42, 72-92, 97-117, 132-152, 170-190, 199-219, 236-256, 264-284, 286-306, 338-358) whose topology has been validate experimentally (Bouhss 1999). The structure of MraY from *Aquifex aeolicus* has been resolved in 2013 by Chung *et al*. (4J72). This protein has 48% identity with the MraY from *E. coli*. To model this protein, I-Tasser has been used. This software built 3D models based on multiple-threading alignments and iterative template fragment assembly simulations. It has been ranked as No 1 server for protein structure prediction during the last CASP experiments. The experimental topology is represented by the magenta spheres.





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