ZDOCKpro for Protein-Protein Docking

Learn how you can better understand the mechanism of action of therapeutically interesting proteins using ZDOCKpro to dock two or more proteins in silico.
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On cover: An approximately 200-residue interleukin-1 receptor that has been docked to a 21-residue peptide (yellow)
using ZDOCKpro is compared to the complex crystal structure 1g0y (blue). Superimposition of the best docked complex
to the crystal structure gives an RMSD of 0.61 Å over 138 residues based on the alpha carbon inter-face residues.
Protein-protein interactions regulate virtually every cellular process, and understanding more about them can be a first step toward learning more about disease and its potential treatment. One way to study protein-protein interactions is through protein-protein docking—the computational determination of the structure of protein-protein complexes or assemblies from individual protein structures.

ZDOCKpro is a novel protein-protein docking program that is based on the ZDOCK and RDOCK programs developed at Boston University by Professor Zhiping Weng. It is a useful tool for protein modelers and structural biologists who want to perform protein-protein docking, as well as for bioinformaticians who study protein pathways and computational chemists who examine protein or peptide ligand docking.

The information on the following pages discusses protein-protein interaction and docking in more detail and introduces you to ZDOCKpro through a sample workflow, application notes, and references.

"The protein-protein docking problem has fascinated biophysicists and computational biologists for over two decades because of its biological importance: a complex structure reveals all structural details of the molecular interaction. It continues to receive considerable attention in the post-genomic era. Structural genomics aims to determine the 3D structure of every protein in the cell including protein-protein complexes, and docking and refinement algorithms like ZDOCK combined with RDOCK can be invaluable in the detailed investigation of the protein-protein interactions, as well as in the design of novel pharmaceuticals. In fact, ZDOCK/RDOCK is one of the best performing algorithms in the community-wide blind test CAPRI (Critical Assessment of Prediction of Interactions)."

Professor Zhiping Weng, Boston University
(developer of ZDOCK and RDOCK)
Protein-Protein Interactions

Protein-protein interactions are significant because biological activity depends on the specific recognition of proteins, and most proteins interact with other proteins in a cell. Protein-protein complexes can involve the interaction of enzyme/inhibitor, antibody/antigen, hormone/hormone receptor, and protein kinase/substrate. Many of such complexes play an important role in antibody reactions and in the signal transduction pathways involved in cancer and human development. As such, a better understanding of protein-protein complexes and how they work together can increase your understanding of interactions that are important to the development and treatment of disease.

The table below defines the significance of some key protein-protein complexes found within humans.

<table>
<thead>
<tr>
<th>Example of Protein-Protein Complexes</th>
<th>Significance</th>
</tr>
</thead>
<tbody>
<tr>
<td>• Thrombin/hirudin blood</td>
<td>Blood coagulation</td>
</tr>
<tr>
<td>• Virus uracil-DNA glycosylase/inhibitor</td>
<td>Biodefense, viral and bacterial inhibition</td>
</tr>
<tr>
<td>• HIV-1 NEF/tyrosine kinase SH3 domain</td>
<td></td>
</tr>
<tr>
<td>• Vh single-domain antibody/lysozyme</td>
<td></td>
</tr>
<tr>
<td>• Antibody/VP6 virus coat protein</td>
<td></td>
</tr>
<tr>
<td>• T-Cell receptor (beta-chain)/exotoxin A1</td>
<td></td>
</tr>
<tr>
<td>• Transducin chimera (of G-protein)/G-protein</td>
<td>Signal transduction, cancer</td>
</tr>
<tr>
<td>• RAS activating domain/RAS</td>
<td></td>
</tr>
<tr>
<td>• Human growth hormone/receptor</td>
<td>Human development, cancer</td>
</tr>
<tr>
<td>• CDK2 cyclin-dependent kinase 2/cyclin</td>
<td></td>
</tr>
</tbody>
</table>

Challenges of Examining Protein-Protein Interactions

Historically, the examination of protein-protein interactions has been a challenge for a number of reasons, including:

- the lack of readily available, high-resolution x-ray crystallographic or NMR protein structures, such as within the Protein Databank (PDB)
- the failure of some protein-protein complexes to crystallize
- the cost and speed of sampling the conformational space of large systems
- the complexity of the process due to the occurrence of conformational changes upon complex formation
- an imperfect understanding of thermodynamics
- heterogeneities in the interactions
- difficulties in locating the interaction site because of the large, flat surface between proteins
ZDOCKpro helps solve some of the traditional problems associated with studying protein-protein interactions by providing a computational means for determining the 3D structure of protein-protein complexes or assemblies starting with the 3D structures of a protein receptor and a protein or peptide ligand. A key advantage to ZDOCKpro is that it lets you rapidly dock and refine two proteins to form an assembly without prior knowledge of the interface residues.

ZDOCKpro contains two main subprograms, ZDOCK and RDOCK. ZDOCK is a fast, rigid-body, initial stage protein-protein docking algorithm that applies ligand rotation and a pair-wise shape complementarity method that takes advantage of fast Fourier transformation. RDOCK is based on the CHARMM® simulations program and is used for further refinement of complexes generated by ZDOCK. RDOCK also ranks the docked structures based on CHARMM electrostatic interaction energy and ACE desolvation energy.

The integration of ZDOCKpro into Discovery Studio®, a comprehensive suite of modeling and simulation solutions, makes it easy for you to prepare the protein, set up experiments, and analyze the results. Within the Discovery Studio research environment, you will be able to create workflows, combining protein docking with multiple other solutions, to take your data from sequence to well-defined, docked complex. A myriad of tools are available for sequence analysis, homology modeling, structure refinement, simulations, and much more!

Evaluation of ZDOCK and RDOCK at the 2004 CAPRI Challenge

ZDOCK and RDOCK were top performers at the 2004 CAPRI Challenge—an international, blind evaluation of protein-protein docking programs. When used together as a prediction model, the ZDOCK/RDOCK algorithms ranked second out of 26 participating groups’ protein-protein docking algorithms. Figure 1 illustrates ZDOCK/RDOCK results from a docking experiment in which ZDOCK/RDOCK ranked first. In the figure, SAG1 (protein ligand) is docked to an antibody (Ab) Fab fragment (receptor). The ligand in the crystal structure complex is shown in red, the predicted docked ligand structures in blue, and the Ab Fab fragment receptor in purple. The ZDOCK/RDOCK prediction model, which utilized a blocking strategy, had 87% correct contacts and a 1.1 Å interface RMSD.

Linux Support

A complete range of Accelrys’ life science modeling and simulation tools, including Discovery Studio and ZDOCKpro, are supported on IBM workstations and laptops running Linux operating systems. Make the most of your time and money by taking advantage of the stability, speed, scalability, graphics capabilities, and cost of this powerful operating system running on IBM hardware—the platform of choice for Linux.
Sample ZDOCKpro Workflow

Predicting a Protein-Protein Complex from Two Unbound Proteins

The following workflow summarizes the basic steps you would run through using ZDOCKpro in Discovery Studio to predict the conformation of a protein-protein complex using two protein structures. The two proteins can be homology models or experimental structures (e.g. from X-ray crystallography) and can be unbound or bound. Compared to bound proteins, unbound proteins provide a challenge since they are not in their complex form. The workflow highlights some of the key functionalities of ZDOCKpro, such as blocking, that allow you to attain high-quality docked complexes for further visualization and analysis. It also suggests how you can use additional computational tools to gain further insight about your docked complexes.

Getting started

Always read the literature about the individual proteins to be docked, such as literature concerning site-directed mutagenesis. This can greatly influence the accuracy and speed of docking by letting you incorporate information about which residues may or may not be involved in the binding of the proteins.

Preparing the proteins

Correct missing residues or atoms.

Step 1: Run ZDOCK to generate initial docked complexes and select the option to block residues that are not in the binding site.

- Dock a multiple chain receptor to a protein or peptide ligand.
- (Optional step) Block residues that are known to not be in the interface.
- Change the angular step size of the ligand for finer sampling (6 or 15 degree search).
- Vary the number of ZDOCK complexes produced for ranking and comparison (maximum 54,000).

Step 2: Filter the complexes based on residues known to be in the interface.

- (Optional step) Filter your ZDOCK conformations when specified residue(s) are known to be in the binding interface within a specific distance cutoff.

Step 3: Run RDOCK to minimize and rank the complexes.

- RDOCK optimizes the ZDOCK complexes by running CHARMm optimization.
  - Two stages of CHARMm minimization create the complex PDB structures with optimized coordinates.
  - Based on the complex structure of each optimization, ACE energies are calculated using the deltaG program and the total RDOCK score is calculated from the ACE energy and CHARMm electrostatic energy.

ZDOCKpro for Protein-Protein Docking
RDOCK output includes the refined receptor and ligand complexes ranked according to their ZDOCK score.

**Step 4: Rank and visualize RDOCK results.**
- Tabulate results (scores) for each docked complex.
- Rank the complexes based on the RDOCK scoring function.
- Visualize the receptor and ligand complexes.

**Analyzing the results**
When visualizing the docked complexes, note that the top ranked complexes show the ligand clustering in one area around the receptor. This indicates the most likely location of the ligand docked to the receptor. If you have an experimental complex structure available, compare this complex structure to your predicted complexes. Re-run ZDOCKpro, varying the parameters (e.g. search degree) and then compare the results with the previous run.
Since the most important factor influencing successful docking is a correct interface, we recommend that researchers use all available information (e.g. literature and results from complementary tools) to confirm the computational docking results.

**Using complementary tools to analyze and confirm the results**
Complementary computational tools (e.g. Discovery Studio®) can complement, as well as confirm, your ZDOCKpro results.
- Use Evolutionary Trace to confirm interface residues and thus determine residues for the blocking and filtering steps.
- Perform a Profiles-3D Verify analysis to identify hydrophobic patches on the surface and to guide you to the probable interface. Hydrophobic interfaces are common in homomeric complexes.
- If the protein is flexible (e.g. it contains a flexible loop or hinge region), try performing several ZDOCKpro runs using different beginning structures with varying conformations (e.g. use MODELER for loop modeling and CHARMM minimization for larger scale conformational motions).
- Perform loop refinement using Looper, which is a CHARMM-based protocol that optimizes the conformation of a contiguous segment (i.e., a loop) of a protein structure. It is based on systematic conformational sampling of the loop backbone and CHARMM energy minimization.
- Optimize protein side-chains based on systematic searching of side-chain conformations and CHARMM energy minimization using the ChiRotor protocol.
- Probe the surface of the protein using small chemical fragments that mimic the ligand and potentially bind to a specific region on the protein surface. This may guide the orientation of the docked complex (e.g. LUDI or MCSS).
- Compare surface characteristics between the proteins, for example with electrostatics using DelPhi.

Further details can be obtained by attending our protein-protein docking half-day workshop. Details at [www.accelrys.com/services/training/life/outlines.html](http://www.accelrys.com/services/training/life/outlines.html)
Example 1: Using ZDOCKpro to Study the G-Protein Regulating System
Tim Glennon & Dana Haley-Vicente—Accelrys Inc., 10188 Telesis Court, San Diego, CA 92121, USA
(For full details, see the poster at http://www.accelrys.com/reference/cases/studies/zdock_evolutionary_trace.pdf)

The G-protein regulating system includes regulator of G-protein signaling (RGS) proteins and the alpha subunit of G-proteins. The RGS family of proteins are involved in accelerating GTP hydrolysis of the G-protein alpha subunit, which leads to rapid recovery of signaling transduction cascades that are involved in controlling vision, cardiac function, and many aspects of neuroendocrine signaling. To analyze and validate this system, we used ZDOCKpro for protein-protein docking and a complementary tool, Evolutionary Trace (ET, Lichtarge et al. 1996), for protein family analysis. We aimed to validate the binding of the unbound ligand, RGS4 (pdb code 1ezt, 205 residues), to the unbound receptor, Guanine Nucleotide-Binding Protein G-(iα) (1git, 353 residues). Results were then compared to the crystal structure complex, 1agr, of the two proteins, 1agr, for validation.

ZDOCKpro analysis alone revealed a good hit that ranked second out of the top 500 refined and re-ranked docked complexes. ET identified a cluster of residues in the RGS domain that includes the RGS-Gαi binding interface. These residues were included in a second round of ZDOCKpro analysis that was used to refine (e.g. filtering before running RDOCK) and re-rank the initial results. After including the ET analysis data, the second hit became the first hit and many false positive hits were removed, indicating protein-protein docking results improve with additional data, such as that which comes from experimental data (e.g. mutagenesis data) or computational methods (e.g. ET data). Larger improvements using ET and ZDOCKpro are seen with more complex systems (data not shown here). Figure 2 illustrates the best-ranked, ZDOCKpro-predicted, docked complex superimposed (based on the receptors, Gαi) with the 1agr crystal structure complex. The RMSD of the interface residues is 4.3Å.

Figure 2: Best-ranked, ZDOCKpro-predicted, docked complex superimposed (based on the receptors, Gαi) with the 1agr crystal structure complex. Predicted ZDOCKpro complex: RGS (1ezt) ligand is shown in purple and the G-alpha (1git) receptor in light blue. Crystal structure complex (1agr): ligand is shown in orange and receptor in green.
Example 2: Can We Use Homology Models in Protein-Protein Docking?

Studying protein-protein interactions is complicated by the fact that the PDB does not contain an abundance of protein-protein complexes and protein-protein interaction databases tend to contain more in vitro and sequence information rather than structure data. Thus, we pose the question: Can we use homology models in protein-protein docking? And, if we can, when can they be used and what are important factors to consider?

To address these questions, acetylcholinesterase (AChE) complexed with fasciculin2 (Fas) (pdb code 1mah) was chosen as the model system. Based on the sequence of 1mah, we built four different models based on four different PDB templates. Align123 (in Insight II) was used for generation of the sequence alignments and MODELER was used for building the homology models. However, only three of the four templates used resulted in good models. One template had structural differences in the binding interface that caused it to produce a model with a loop pointing directly into the binding site, which thereby prevented good docking results.

The homology models were then used as receptor proteins in ZDOCKpro docking. After initial docking with ZDOCK, the 2000 best poses of near native structures were refined and re-ranked with RDOCK. The ZDOCKpro scores were then evaluated for the top five re-ranked complexes, and then visually clustered, taking the centers of mass of each ligand and visualizing these for the top structures versus the rest of the ligand poses. Figure 3 shows results of one homology model (blue ribbon) built from the template 1b41 that had 87.8% sequence identity with 1mah. All the spheres in the figure represent the center of mass for each ligand position as calculated by ZDOCKpro. The larger green spheres represent the center of mass of the top five re-ranked ligand poses; the small white spheres correspond to the rest of the top 2000 poses, and the orange ribbon schematic is the original ligand position relative to the receptor. Note the green spheres of the top five ligand poses lie in the same location as the original ligand, indicating that these ligand poses are in the right location. These results have shown that using homology models in protein-protein docking is feasible. Similar results were shown using 1dx4 as the template, which has a 59.3% sequence identity. Additionally, the CAPRI Challenge results (not shown here) also provide examples of the successful use of homology models in ZDOCKpro docking.

Figure 3: ZDOCKpro docking results run on a homology model built from the PDB sequence of 1b41.
Development of the ZDOCK and RDOCK Methodology


The development of scoring functions is of great importance to protein docking. Here we present a new scoring function for the initial stage of unbound docking. It combines our recently developed pairwise shape complementarity with desolvation and electrostatics. We compare this scoring function with three other functions on a large benchmark of 49 nonredundant test cases and show its superior performance, especially for the antibody-antigen category of test cases. For 44 test cases (90% of the benchmark), we can retain at least one near-native structure within the top 2000 predictions at the 6° rotational sampling density, with an average of 52 near-native structures per test case. The remaining five difficult test cases can be explained by a combination of poor binding affinity, large backbone conformational changes, and our algorithm's strong tendency for identifying large concave binding pockets. All four scoring functions have been integrated into our Fast Fourier Transform based docking algorithm ZDOCK, which is freely available to academic users at http://zlab.bu.edu/dock. (© 2003 Wiley-Liss, Inc.)


We present a simple and effective algorithm RDOCK for refining unbound predictions generated by a rigid-body docking algorithm ZDOCK, which has been developed earlier by our group. The main component of RDOCK is a three-stage energy minimization scheme, followed by the evaluation of electrostatic and desolvation energies. Ionic side chains are kept neutral in the first two stages of minimization, and reverted to their full charge states in the last stage of brief minimization. Without side chain conformational search or filtering/clustering of resulting structures, RDOCK represents the simplest approach toward refining unbound docking predictions. Despite its simplicity, RDOCK makes substantial improvement upon the top predictions by ZDOCK with all three scoring functions and the improvement is observed across all three categories of test cases in a large benchmark of 49 non-redundant unbound test cases. RDOCK makes the most powerful combination with ZDOCK2.1, which uses pairwise shape complementarity as the scoring function. Collectively, they rank a near-native structure as the number-one prediction for 18 test cases (37% of the benchmark), and within the top 4 predictions for 24 test cases (49% of the benchmark). To various degrees, funnel-like energy landscapes are observed for these 24 test cases. To the best of our knowledge, this is the first report of binding funnels starting from global searches for a broad range of test cases. These results are particularly exciting, given that we have not used any biological information that is specific to individual test cases and the whole process is entirely automated. Among three categories of test cases, the best results are seen for enzyme/inhibitor, with a near-native structure ranked as the number-one prediction for 48% test cases, and within the top 10 predictions for 78% test cases. RDOCK is freely available to academic users at http://zlab.bu.edu/dock. (© 2003 Wiley-Liss, Inc.)


A comprehensive docking study was performed on 27 distinct protein-protein complexes. For 13 test systems, docking was performed with the unbound X-ray structures of both the receptor and the ligand. For the remaining systems, the unbound X-ray structure of only molecule was available; therefore the bound structure for the other molecule was used. Our method optimizes desolvation, shape complementarity, and electrostatics using a Fast Fourier Transform algorithm. A global search in the rotational and translational space without any knowledge of the binding sites was performed for all proteins except nine antibodies recognizing antigens. For these antibodies,
we docked their well-characterized binding site - the complementarity-determining region defined without information of the antigen - to the entire surface of the antigen. For 24 systems, we were able to find near-native ligand orientations (interface C. root mean square deviation less than 2.5 Å from the crystal complex) among the top 2,000 choices. For three systems, our algorithm could identify the correct complex structure unambiguously. For 13 other complexes, we either ranked a near-native structure in the top 20 or obtained 20 or more near-native structures in the top 2,000 or both. The key feature of our algorithm is the use of target functions that are highly tolerant to conformational changes upon binding. If combined with a post-processing method, our algorithm may provide a general solution to the unbound docking problem. Our program, called ZDOCK, is freely available to academic users (http://zlab.bu.edu/rong/dock/). (© 2002 Wiley-Liss, Inc.)

Chen R. & Weng Z., “A Novel Shape Complementarity Scoring Function For Protein-Protein Docking,” Proteins, 2003, 51, 397–408. Shape complementarity is the most basic ingredient of the scoring functions for protein-protein docking. Most grid-based docking algorithms use the total number of grid points at the binding interface to quantify shape complementarity. We have developed a novel Pairwise Shape Complementarity (PSC) function that is conceptually simple and rapid to compute. The favorable component of PSC is the total number of atom pairs between the receptor and the ligand within a distance cutoff. When applied to a benchmark of 49 test cases, PSC consistently ranks near-native structures higher and produces more near-native structures than the traditional grid-based function, and the improvement was seen across all prediction levels and in all categories of the benchmark. Without any post-processing or biological information about the binding site except the complementarity-determining region of antibodies, PSC predicts the complex structure correctly for 6 test cases, and ranks at least one near-native structure in the top 20 predictions for 18 test cases. Our docking program ZDOCK has been parallelized and the average computing time is 4 minutes using sixteen IBM SP3 processors. Both ZDOCK and the benchmark are freely available to academic users (http://zlab.bu.edu/rong/dock/). (© 2003 Wiley-Liss, Inc.)

Evaluation of ZDOCK and RDOCK at CAPRI
We present an evaluation of the results of our ZDOCK and RDOCK algorithms in Rounds 3, 4, and 5 of the protein docking challenge CAPRI. ZDOCK is a Fast Fourier Transform (FFT)-based, initial-stage rigid-body docking algorithm, and RDOCK is an energy minimization algorithm for refining and reranking ZDOCK results. Of the 9 targets for which we submitted predictions, we attained at least acceptable accuracy for 7, at least medium accuracy for 6, and high accuracy for 3. These results are evidence that ZDOCK in combination with RDOCK is capable of making accurate predictions on a diverse set of protein complexes. (© 2005 Wiley-Liss, Inc.)

The CAPRI Challenge is a blind test of protein-protein-docking algorithms that predict the complex structure from the crystal structures of the interacting proteins. We participated in both rounds of this blind test and submitted predictions for all seven targets, relying mainly on our Fast Fourier Transform based algorithm ZDOCK that combines shape complementarity, desolvation, and electrostatics. Our group made good predictions for three targets and had at least some success with three others. Implications of the treatment of prior biological information as well as contributions of manual inspection to docking predictions are also discussed. (© 2003 Wiley-Liss, Inc.)
Publications Citing use of ZDOCK/RDOCK


Computational methods are employed to simulate interaction of scorpion toxin ScyTx in complex with the small conductance calcium-activated potassium channel rsk2. All of available 25 structures of ScyTx in the Protein Data Bank determined by NMR were considered for improving performance of rigid protein docking of ZDOCK. Four main binding modes were found among a large number of predicted complexes by using clustering analysis, screening with expert knowledge, energy minimization, and molecular dynamics simulations. The quality and validity of the resulting complexes were further evaluated by molecular dynamics simulations with the generalized Born solvation model and by calculation of relative binding free energies with the molecular mechanics Poisson-Boltzmann surface area (MM-PBSA) in the AMBER 7 suit of programs. The complex formed by the 22nd structure of the ScyTx and rsk2 channel was identified as the most favorable complex by using a combination of computational methods; which contain further introduction of flexibility without restraining residue side chain. From the resulted spatial structure of the ScyTx and rsk2 channel, ScyTx associates the mouth of the rsk2 channel with α-helix rather than β-sheet. Structural analysis first revealed that Arg13 played a novel and vital role of blocking the pore of the rsk2 channel, whose role is remarkably different from that of highly homologous scorpion toxin POS. Between the interfaces in the ScyTx-rsk2 complex, strong electrostatic interaction and hydrogen bonds exist between Arg13 of ScyTx and Gly-Tyr-Gly-Asp sequential residues located in the four symmetrical chains of the pore region. Simultaneously, five hydrogen bonds between Arg6 of ScyTx and Asp341(C), Val366(A), and Pro367(C), and electrostatic interaction between Arg6 of ScyTx and Asp364(B) and Asp341(C) are also found by structural analysis. In addition, His31 located at the C-terminal of ScyTx is surrounded by Val342(A), Asp364(A), Met365(A), Pro367(B), and Asn366(B) within a contact distance of 4.0 Å. These simulation results are in good agreement with experimental data and can effectively explain the binding phenomena between ScyTx and the potassium channel at the level of molecular spatial structure. The consistency between results of molecular modeling and experimental data strongly suggests that our spatial structure model of the ScyTx-rsk2 complex is reasonable. Therefore, molecular docking combined with molecular dynamics simulations followed by molecular mechanics Poisson-Boltzmann surface area analysis is an attractive approach for modeling scorpion toxin-potassium channel complexes a priori for further biological studies. (© 2004 The Biophysical Society)


Structures of macromolecular complexes are necessary for a mechanistic description of biochemical and cellular processes. They can be solved by experimental methods, such as X-ray crystallography, NMR spectroscopy and electron microscopy, as well as by computational protein structure prediction, docking and bioinformatics. Recent advances and applications of these methods emphasize the need for hybrid approaches that combine a variety of data to achieve better efficiency, accuracy, resolution and completeness. (© 2004 Elsevier Science)


Virtually every biological process involves protein–protein contact but relatively few protein–protein complexes have been solved by X-ray crystallography. As more individual protein structures become available, computational methods are likely to play increasingly important roles.
in defining these interactions. Tubulin folding and dimer formation are complex processes requiring a variety of protein cofactors. One of these is cofactor A, which interacts with β-tubulin prior to assembly of the α-tubulin–β-tubulin heterodimer. In the yeast Saccharomyces cerevisiae, β-tubulin is encoded by TUB2 and cofactor A by RBL2. We have used computational docking and site-directed mutagenesis to generate a model of the Rbl2–Tub2 complex from the solved structures of these two proteins. Residues in the N termini and the loops of the Rbl2 homodimer appear to mediate binding to β-tubulin. These interact with β-tubulin residues in the region that contains helices H9 and H10. Rbl2 and α-tubulin share overlapping binding sites on the β-tubulin molecule providing a structural explanation for the mutually exclusive binding of Rbl2 and α-tubulin to β-tubulin. (© 2004 Elsevier Ltd)


A model structure of the Hsc70/auxilin complex has been constructed to gain insight into interprotein substrate transfer and ATP hydrolysis induced conformational changes in the multidomain Hsc70 structure. The Hsc70(auxilin system, which is a member of the Hsp70/Hsp40 chaperone system family, uncoats clathrin-coated vesicles in an ATP hydrolysis-driven process. Incorporating previous results from NMR and mutant binding studies, the auxilin J-domain was docked into the Hsc70 ATPase domain lower cleft using rigid backbone/flexible side chain molecular dynamics, and the Hsc70 substrate binding domain was docked by a similar procedure. For comparison, J-domain and substrate binding domain docking sites were obtained by the rigid-body docking programs DOT and ZDOCK, filtered and ranked by the program ClusPro, and relaxed using the same rigid backbone/flexible side chain dynamics. The substrate binding domain sites were assessed in terms of conserved surface complementarity and feasibility in the context of substrate transfer, both for auxilin and another Hsp40 protein, Hsc20. This assessment favors placement of the substrate binding domain near D152 on the ATPase domain surface adjacent to the J-domain invariant HPD segment, with the Hsc70 interdomain linker in the lower cleft. Examining Hsc70 interdomain energetics, we propose that long-range electrostatic interactions, perhaps due to a difference in the pKa values of bound ATP and ADP, could play a major role in the structural change induced by ATP hydrolysis. Interdomain electrostatic interactions also appear to play a role in stimulation of ATPase activity due to J-domain binding and substrate binding by Hsc70. (Copyright © 2004 The Protein Society)