## Rational drug design from toxins - how rational can one get?

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Developing drugs from natural products such as toxins has a great potential but the complexity of the problem has been hampering progress. Combination of experimental methods with accurate simulations of protein-peptide complexes could help to improve this situation. The two main computational problems are construction of accurate models of complexes, and prediction of reliable binding free energies. The former can be achieved using a high-end docking program such as HADDOCK, followed by refinement *via* molecular dynamics (MD) simulations. For the latter one can use umbrella sampling MD simulations to construct the potential of mean force of a ligand, from which its binding free energy can be calculated. We have tested this strategy in a potassium channel-charybdotoxin complex, where the complex structure is known from NMR, and shown that accurate complex structure and binding free energy can be obtained (Chen & Kuyucak, 2011). We have next considered binding of ShK toxin to Kv1 channels. ShK toxin binds to Kv1.3 channels with very high affinity, and for this reason it is being developed as an immunosuppressant drug. However, it also binds to Kv1.1 with similar affinity, and it is essential to find analogues of ShK with increased selectivity for Kv1.3. Obviously accurate models of Kv1-ShK complex will be very useful in this search. We have developed such models for Kv1.1, 1.2 and 1.3 channels in complex with ShK, which are validated by comparing with available mutagenesis data and binding free energies (reproduced within the chemical accuracy of 1 kcal/mol for all three channels) (Rashid & Kuyucak, 2012). Several hundred analogues of ShK have been developed so far to increase its selectivity for Kv1.3 over Kv1.1 (Chi *et al.*, 2012). We have used one of the promising candidates (ShK-K-amide) as a test case, and again reproduced the changes in the binding free energies accurately. The molecular-level explanation of how selectivity arises in ShK-K-amide will be useful in future searches for more selective analogues. The complex structures of ShK obtained by Rashid & Kuyucak have also indicated several mutations on ShK (*e.g.* K18A) that could enhance its selectivity for Kv1.3 over Kv1.1. Free energy perturbation calculations performed on A18-ShK yield about 2 kcal/mol reduction in its affinity to Kv1.1 relative to Kv1.3, confirming this expectation. These results show that, using the present computational methods, one can describe the protein-peptide toxin interactions fairly accurately, which opens the way for rational drug design from toxins and other peptides.

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