for developing, validating, and benchmarking computational screening, docking, and binding affinity prediction, including both curating crystal structures and experimental binding affinities (see NIH NIGMS RFA-GM-08-008). NIST has recently become interested in developing and curating such data as well. Most importantly, there is a much larger wealth of data in proprietary pharmaceutical databases that no longer has significant intellectual property value, and a system for releasing such data to the broader community would be immensely valuable for development of improved drug design methods.

Another problem in the development of free-energy techniques is that most large-scale validations and comparisons of methodologies have been retrospective. There are relatively few opportunities to participate in large-scale prospective trials, as confidence of experimentalists in quantitative predictions made by computer is usually not high enough to motivate the high-quality experiments that can validate the computational results.

The quality of techniques for protein structure prediction has increased since the introduction of CASP (Critical Assessment of Structure Prediction) in 1994. Despite some criticisms about some aspects of the program,[232] it is considered to have played an important role in developing computational structure prediction. Other successful ongoing prospective computational challenges have been CAPRI (Critical Assessment of PRedicted Interactions) for protein/protein complexes,[233] the Industrial Fluid Properties Simulation Collective (IFPSC) (at http://www.fluidproperties.org), the Cambridge Crystallographic Data Centre's blind tests of small-molecule crystal structure prediction,[234] and the McMasters high-throughput screening competition.[235] It is clear that providing an opportunity for truly blind predictions of chemical and biological properties structure has been beneficial for the computational methodology community. There has therefore naturally been an interest in developing a similar true prospective trial for prediction of ligand binding affinities.

One such attempt called CATFEE (Critical Assessment of Techniques for Free Energy Evaluation) was attempted in 2000 but failed because the experimental data never became available.[236] A more recent attempt, called SAMPL (Statistical Assessment of the Modeling of Proteins and Ligands), run by OpenEye Software, was conducted in late 2007 to early 2008, with two protein targets (urokinase, with data contributed by Abbott, and JNK3 kinase, with data contributed by Vertex) and sixty-three ligand binding points determined by IC_{50}s, but with the same assay for each target. The competition consisted of virtual screening against decoys, pose prediction of known actives, and prediction of binding affinity from crystal structures. Although the summary of the results is still in preparation, by almost all measures they were somewhat discouraging, with correlations to predictions using various physically based methods significantly worse than 1 kcal/mol root-mean-square error. Interestingly, the best method was a less computationally demanding approximation to MM-PBSA that essentially ignored the entropy contribution[237] but even this method was very unreliable. The initial SAMPL generated significant participation and interest and is very likely to continue.

In the foreseeable future, fully atomistic free-energy calculations may be most important not solely for reliable predictions of binding affinity, but for a wealth of additional atomistic information such as probabilities of occupation of binding pose and water structure in the binding site that are impossible to gather from either experiment or more approximate methods. Free-energy calculations may also be of use in the future for the computation of octanol/water partition coefficients of molecules that are difficult to predict by standard rule-based algorithms like CLOGP or for even more direct membrane permeability simulations. Calculating the free energy, and thus stability, of different tautomers represents another important application of fully physical simulations. Questions of ligand-binding specificity can frequently be seen as a multivariate optimization problem, with binding to the intended target maximized, while binding to the alternative targets is minimized.

For further information, readers are encouraged to read a number of reviews on the subject of free-energy calculations published more recently,[8–11,13,238–240] useful textbooks,[15–17,231] as well as older reviews and books that may provide more historical perspective.[32,241,242]

A number of the reviews on the subject of free-energy calculations in ligand binding since the late 1980s conclude that free-energy calculations of ligand binding have finally overcome the problems and false starts of the past and that the time for free energies in pharmaceutical industry is nearly here or has already arrived. We will not make nearly as strong a claim here. An extensive survey of the latest results is somewhat mixed, and it is not clear that these methods will necessarily be an important part of the pharmaceutical work flow anytime in the near future. In some cases, computational simulations may be approaching the level of accuracy that they can provide some additional utility in some aspects of lead optimization, but the accuracy and speed of the methods presented in this chapter must both be improved drastically. Many computational chemists working in industry that were questioned by the authors thought that rigorous free-energy methods may eventually become a routine part of drug discovery methods but perhaps not for another twenty years.

It does, however, appear that improvements in computational power and methodologies have made it possible to compute increasingly reliable relative and absolute binding affinities, albeit with significant computational and human effort. Continuing improvements in techniques will make physics-based simulations more and more attractive, leading to improved simulation tools and eventually to a vital place in the pharmaceutical work flow.