



Figure 8.1. Hierarchy of QM methods used in drug design.

production techniques, interest in novel applications of x-ray crystallography in a high-throughput fashion have also been growing. The use of high-throughput crystallography in fragment-based drug discovery, for example, has been explored.<sup>5-7</sup>

For these high-throughput approaches to be practical for drug discovery, structure determination must be rapid enough to provide timely feedback to the design team; meanwhile, the structure of the complex, especially the ligand, must be determined accurately enough to provide adequate reliability on the subsequent hypotheses made about the observed binding interactions. However, it is often overlooked that protein crystallography operates at a resolution that is lower than that observed for small molecules, which gives rise to poor data-to-parameter ratios in protein structure refinements. In particular, the amount of x-ray

diffraction data observed is usually not sufficient to determine the coordinates, occupancies, and temperature factors for all the atoms. It can be shown that at a resolution of 2Å, the data-to-parameter ratio is slightly better than 2, while if the resolution drops to 2.7Å, a resolution that would not be uncommon for high-throughput crystallography, this ratio is less than 1. The issue of poor data-to-parameter ratios is dealt with in the energetically restrained refinement (EREF) formalism by introducing energy restraints to complement the x-ray data:<sup>8</sup>

$$E_{\text{total}} = E_{\text{chem}} + w_{\text{x-ray}} E_{\text{x-ray}}, \quad (8.1)$$

where  $E_{\text{total}}$  is the function minimized during the refinement,  $E_{\text{chem}}$  is the energy function conventionally approximated with MM,  $E_{\text{x-ray}}$  is the x-ray target function, and  $w_{\text{x-ray}}$  is the weight that balances the contributions from