1.4.3. Additional comments on *Gazing into the crystal ball* (D. M. Himmel)

I wish to expand briefly on Eddy's and Michael's comments to entertain the question of how macromolecular crystallography and structural biology might change the future. New detector technologies, such as shutterless detector systems that can collect data in continuous rotation mode (see, for example, Miyoshi *et al.*, 2008), are now being developed and deployed at synchrotrons. These detectors will improve the quality of data collection and integration. It is possible that the emergence of ever more rapid and more sensitive X-ray detectors, along with new X-ray technologies, will lead to multiple-wavelength rapidly pulsed X-ray beams that are synchronized by wavelength with the detector. At each crystal rotation position, the beam will rapidly run through each of the several tuned wavelengths and record a diffraction pattern for each wavelength. This sort of setup can be used to collect MAD data from a single data pass through the Ewald sphere. In cases where crystals are particularly sensitive to X-ray damage, this approach, along with the ability to detect anomalous signals from ever-smaller elements in the Periodic Table, may make MAD more competitive with molecular replacement as the method of choice for structure determination. Molecular replacement itself will become more powerful as it is combined with *ab initio* phasing and computational chemistry methods. In the future, due to other emerging technologies such as described in Section 1.4.4 below, whether or not one can crystallize a macromolecule may no longer limit one's ability to determine a structure by X-ray diffraction. Here, I will focus on two additional points: (1) how X-ray crystallography and allied fields (such as NMR and EM) have already started to transform the physical sciences, and (2) how they might transform the overall human experience as well.

In the mid-twentieth century, the physical sciences to a considerable extent were divided into separate fiefdoms that often competed with each other rather than fostered a collaborative spirit. Physics, chemistry, virology and biochemistry, to name a few, each attempted to stand on its own, each using its own independent preferred jargon and preferred explanations for scientific questions of the day. By contrast, what we see happening today is a convergence of all these once-separate fields to understand first principles right down to the molecular level and beyond. More than that, macromolecular crystallography and allied fields are playing a substantial role in catalysing this convergence. The determination and subsequent analysis of a macromolecular structure (such as a protein in complex with nucleic acid, co-factors and/or small-molecule ligands) of necessity culminates from the application of physics, mathematics and chemistry to begin with, followed by various biological sciences to understand the context of a molecular structure. But for physicists, chemists and biologists to speak to each other productively, they increasingly must share the same jargon and learn each other's disciplines. Sir Isaac Newton once wrote, 'If I have seen further it is by standing on the shoulders of giants.' The giants upon which we stand today in crystallography were the leaders of those separate fields that today find their synthesis in the determination and understanding of molecular structure. It is reasonable to assume that this unifying spirit will continue and will foster greater breakthroughs in structure determination and understanding the properties of molecules in the real world.

In addition to bringing various disciplines of science together, macromolecular crystallography and allied fields are likely to revolutionize the way people live in the future. Richard Feynman was reported to have said, 'What I cannot create, I do not understand.' Chapters 3.1, 3.2, and 4.3 of this volume describe the harnessing of gene expression and protein engineering to further the aims of the X-ray crystallographic experiment. Macromolecular engineering, however, does not stop there. Recent years have seen a rising interest in the field of nanotechnology, which, according to some definitions, aims to design machines and technologies that operate on a scale of about 100 nm or smaller (Farokhzad & Langer, 2009). These technologies generally encompass applications of our understanding of chemistry and physics. As this field matures, it will encounter many of the problems on the molecular level that have already been solved by the machines and devices that exist in biological systems, such as enzymes, molecular motors and structural proteins. Proofs of concept have recently been described in which components of biological molecules were redesigned for applications outside their usual environment (Goel & Vogel, 2008; van den Heuvel & Dekker, 2007; Lewis *et al.*, 2011), or in which principles learned from structural biology have been applied to the construction of completely artificial molecular devices (Ceroni *et al.*, 2010). The discoveries of structural biology may well light the way to the nanotechnologies of the future. These technologies, by operating at the molecular level like never before, will enable the manufacture of superior fabrics and materials, improve medical diagnostics, and revolutionize electronics and photonics in a whole host of devices, from computers and robotics to communication and the efficient harnessing of unconventional energy sources like light. Chemical catalysis will be performed in industry with such tight control that there will be far fewer unwanted side reactions than is commonplace today, so that, for example, medicines and other materials can be manufactured cheaply to an exceptionally high level of purity. Drug-delivery vehicles will be developed that precisely target a cell type, tissue type (Farokhzad & Langer, 2009) or even a pathogen. Some of these molecular vehicles will be modelled on methods used by viruses such as influenza, HIV or even bacteriophages, using a harpoon or plasma membrane fusion strategy, and will release their cargo in response to a chemical or other trigger. Others will employ active transport, in which the therapeutic agent will be guided to its target by remote control and carry diagnostics modules for the ride. The development of these nanotechnologies will both benefit from nanotechnological spin-offs of structural studies as well as require X-ray crystallography and other structural techniques to aid in the analysis of the nanomachines.

To peer at an image of individual molecules at atomic or near-atomic resolution, X-ray crystallography has served to provide the missing lens needed to focus that image. New and maturing fields described in the following sections will supply other ways to provide that missing lens, or, indeed, to obtain the initial phase information needed to determine a higher-resolution X-ray crystal structure.

1.4.4. *Gazing into the crystal ball* – the X-ray free-electron laser (J. C. H. Spence)

The recent invention of the pulsed hard X-ray (free-electron) laser (XFEL) is certain to impact structural biology, particularly in the areas of protein nanocrystal analysis (Chapman *et al.*, 2011), single-particle imaging (Siebert *et al.*, 2011), time-resolved crystallography and solution scattering (see the forthcoming reviews in *Reports on Progress in Physics* by Spence and Chapman). Current hard-X-ray machines provide about 10^{12}