PHILOSOPHICAL TRANSACTIONS OF THE ROYAL SOCIETY B BIOLOGICAL SCIENCES

The birth of a new field

John C. H. Spence and Henry N. Chapman

Phil. Trans. R. Soc. B 2014 **369**, 20130309, published 9 June 2014

PHILOSOPHICAL TRANSACTIONS OF THE ROYAL SOCIETY B

rstb.royalsocietypublishing.org

The birth of a new field†

John C. H. Spence[1] and Henry N. Chapman[2]

[1]Department of Physics, Arizona State University, Tempe, AZ 85282, USA
[2]Department of Physics, University of Hamburg, Luruper Chaussee 149, Hamburg 22607, Germany

Introduction

Cite this article: Spence JCH, Chapman HN. 2014 The birth of a new field. *Phil. Trans. R. Soc. B* **369**: 20130309.
http://dx.doi.org/10.1098/rstb.2013.0309

One contribution of 27 to a Discussion Meeting Issue 'Biology with free-electron X-ray lasers'.

Subject Areas:
biophysics, structural biology

Keywords:
Louise Johnson, Introduction, X-ray lasers

Author for correspondence:
John C. H. Spence
e-mail: spence@asu.edu

†Dedicated to Dame Professor Louise Johnson, FRS 1940–2012.

In September 2011, a proposal was submitted to the Royal Society to hold two, 2-day workshops on the application of free-electron X-ray lasers to biology by Dame Professor Louise Johnson FRS, Henry Chapman and John Spence. The first 2 days, at the Royal Society in London, were to be devoted to biological applications, and the second 2 days following, at Chicheley Hall, on instrumentation development and data analysis methods. The conference was the first in an annual series to be supported on this theme by the new US National Science Foundation's Science and Technology Center on Biology with X-ray lasers.

We were therefore most grateful to the Royal Society when it agreed to financially support these workshops, which began on 14 October 2013. About 80 scientists attended each workshop, coming from many countries, at which half-hour talks were given by 15 leaders in the field at each workshop, together with poster sessions. The speakers were then invited to submit papers for this special issue of *Philosophical Transactions*, in which the resulting 26 papers are published.

It is often said that 'to see something new, you must do something new', and the invention of the X-ray free-electron laser (XFEL) (outlined briefly by Altarelli & Mancuso [1] in this issue) is a fine example of how a technological breakthrough (in this case, pulsed lasing for hard X-rays) can give scientists new eyes. Because radiation damage has historically imposed a near-fundamental limitation on the quality and resolution of images in nearly all forms of biological microscopy, the first suggestion that sufficiently brief X-ray pulses could outrun damage (to provide a useful image before the sample is destroyed) in 1982 was of considerable importance. Detailed simulations (see Neutze's [2] contribution for this background) confirmed this, and the effect was first demonstrated experimentally in 2006 (see [3]), so that over the past decade progress has been rapid. The opportunity this capability provides to image biomolecules at room temperature (without freezing to avoid damage) in their native environment, and to explore the domain of much faster time resolution was clear from the beginning. The construction of the world's first hard X-ray laser, the Linac Coherent Light Source by the U.S. Department of Energy at the Stanford Linear Accelerator Center (SLAC) in California was completed in 2009, allowing many of us at this workshop to obtain the first results for biology in that year. The machine, in a 2-mile-long tunnel near Stanford, generates 120 pulses of hard or soft X-rays per second, containing about 1×10^{12} photons per 10 fs pulse, and, using purpose-built detectors, allows the diffraction pattern from each pulse to be read-out and saved. Broadly, three types of experiments were first attempted—those in which hydrated protein nanocrystals were sprayed across the pulsed beam (serial femtosecond nanocrystallography, SFX), those in which the hard X-ray beam of micrometre dimensions traverses many biomolecules in a liquid jet (fast solution scattering, FSS—see contributions by Haldrup [4], Mendez *et al*. [5] and Pande *et al*. [6]), and single particle (SP) imaging, in which a beam of submicrometre dimensions scatters from an SP such as a virus [7–9]. Before long many other experimental arrangements had also been tried during this exciting first 4 years, including fixed samples scanned across the beam for the study of two-dimensional membrane protein crystals [10], time-resolved SFX [11] (see also Moffat [12]), and new types of sample delivery devices, such as those based on the lipid cubic phase [13,14] and on electrospraying [15].

At the same time, the highly coherent nature of the XFEL has resulted in entirely new kinds of data, providing both opportunities and challenges in data analysis. It was immediately realized that the high spatial coherence provided

new approaches to the phase problem, for both SPs (Loh [7], Ourmazd and co-workers [8], Martin [9], Schwander *et al*. [16]) and nanocrystals (Millane & Chen [17], Kirian *et al*. [18], Spence *et al*. [19] and Barty *et al*. [20]). Atomic resolution has so far been obtained from unknown structures only using nanocrystals (and near-atomic resolution in the FSS when small changes in a known structure are studied), so that a crucial area for development of the BioXFEL field is the development of new methods for growing nanocrystals (Kupitz *et al*. [21], Caffrey *et al*. [22], Gallat *et al*. [23] and Stevenson *et al*. [24]). When the study of the evolving damage processes (reviewed by Chapman *et al*. [3]), diffraction physics (White [25]), simultaneous emission spectroscopy (Kern *et al*. [15]) and detector development (Denes [26]) is added to this list of subfields, it will be seen that structure and dynamics in biology with XFELs is an extremely rich interdisciplinary field, now undergoing rapid innovation and creative development.

With the first 'new biology' results recently published (see papers of Liu *et al*. [14], Gallat *et al*. [23]), and the first high-resolution time-resolved solution-scattering results from proteins about to appear (Neutze [2]), it appears that the confidence of the founders of this new field of structural and dynamic biology was not misplaced, and the US Department of Energy's gamble in building the first XFEL will pay off scientifically in many fields. New XFELs are now under construction in several countries. In addition to the second XFEL now operating in Japan, new machines will start soon in Hamburg, Germany, and Villigen, Switzerland. We look forward to the time when the use of XFELs in biology will be as common as that of synchrotrons, with the added benefits of improved time resolution for structural dynamics, coherence, ability to study proteins which are difficult to crystallize with the possibility for rapid high-throughput structure determination, and radiation damage amelioration.

Louise Johnson's pioneering contributions to structural biology are well-known to all its practitioners, and will be documented in detail in the Royal Society's obituary notices. Her book with Sir Tom Blundell FRS, is an indispensable reference in universities and at many beamlines. Her support for this field, at a time when it had many sceptics, was typical of her adventurous approach to science. She took part in some of the first experiments at the FLASH soft X-ray FEL where she was very satisfied with 'flying' her samples across the beam, and was a strong advocate for building an FEL in the UK. As a young student in the earliest days of protein crystallography, she worked with David Phillips KBE, FRS and others to solve the first enzyme structure, lysozyme, in 1965 (then only the third protein to be solved). It was that structure which first demonstrated, around the time of her death, that the SFX method could, indeed, demonstrate atomic resolution. Her death was an immense loss to this new field, and we will always be grateful for her help in founding it.

The excitement of developing a new field, being first to see new effects, the design of novel instrumentation and the long-term prospects for imaging molecular machines at work emerges strikingly from all these papers. As in the early days of the development of the scanning tunnelling microscope, the discovery of a new signal is generating an enormously creative initial period of invention, ferment and stimulus, in which many ideas are being tried out, only some of which will mature to become useful techniques for biologists. As is clear from these papers, the birth of a new field is the most exciting time to be involved in research.

References

1. Altarelli M, Mancuso AP. 2014 Structural biology at the European X-ray free-electron laser facility. *Phil. Trans. R. Soc. B* **369**, 20130311. (doi:10.1098/rstb.2013.0311)
2. Neutze R. 2014 Opportunities and challenges for time-resolved studies of protein structural dynamics at X-ray free-electron lasers. *Phil. Trans. R. Soc. B* **369**, 20130318. (doi:10.1098/rstb.2013.0318)
3. Chapman HN, Caleman C, Timneanu N. 2014 Diffraction before destruction. *Phil. Trans. R. Soc. B* **369**, 20130313. (doi:10.1098/rstb.2013.0313)
4. Haldrup K. 2014 Singular value decomposition as a tool for background corrections in time-resolved XFEL scattering data. *Phil. Trans. R. Soc. B* **369**, 20130336. (doi:10.1098/rstb.2013.0336)
5. Mendez D *et al*. 2014 Observation of correlated X-ray scattering at atomic resolution. *Phil. Trans. R. Soc. B* **369**, 20130315. (doi:10.1098/rstb.2013.0315)
6. Pande K, Schwander P, Schmidt M, Saldin DK. 2014 Deducing fast electron density changes in randomly orientated uncrystallized biomolecules in a pump–probe experiment. *Phil. Trans. R. Soc. B* **369**, 20130332. (doi:10.1098/rstb.2013.0332)
7. Loh ND. 2014 A minimal view of single-particle imaging with X-ray lasers. *Phil. Trans. R. Soc. B* **369**, 20130328. (doi:10.1098/rstb.2013.0328)
8. Hosseinizadeh A, Schwander P, Dashti A, Fung R, D'Souza RM, Ourmazd A. 2014 High-resolution structure of viruses from random diffraction snapshots. *Phil. Trans. R. Soc. B* **369**, 20130326. (doi:10.1098/rstb.2013.0326)
9. Martin AV. 2014 The correlation of single-particle diffraction patterns as a continuous function of particle orientation. *Phil. Trans. R. Soc. B* **369**, 20130329. (doi:10.1098/rstb.2013.0329)
10. Pedrini B *et al*. 2014 7 Å resolution in protein two-dimensional-crystal X-ray diffraction at Linac Coherent Light Source. *Phil. Trans. R. Soc. B* **369**, 20130500. (doi:10.1098/rstb.2013.0500)
11. Tran R *et al*. 2014 The Mn_4Ca photosynthetic water-oxidation catalyst studied by simultaneous X-ray spectroscopy and crystallography using an X-ray free-electron laser. *Phil. Trans. R. Soc. B* **369**, 20130324. (doi:10.1098/rstb.2013.0324)
12. Moffat K. 2014 Time-resolved crystallography and protein design: signalling photoreceptors and optogenetics. *Phil. Trans. R. Soc. B* **369**, 20130568. (doi:10.1089/rstb.2013.0568)
13. Weierstall U. 2014 Liquid sample delivery techniques for serial femtosecond crystallography. *Phil. Trans. R. Soc. B* **369**, 20130337. (doi:10.1098/rstb.2013.0337)
14. Liu W, Wacker D, Wang C, Abola E, Cherezov V. 2014 Femtosecond crystallography of membrane proteins in the lipidic cubic phase. *Phil. Trans. R. Soc. B* **369**, 20130314. (doi:10.1098/rstb.2013.0314)
15. Kern J *et al*. 2014 Methods development for diffraction and spectroscopy studies of metalloenzymes at X-ray free-electron lasers. *Phil. Trans. R. Soc. B* **369**, 20130590. (doi:10.1098/rstb.2013.0590)
16. Schwander P, Fung R, Ourmazd A. 2014 Conformations of macromolecules and their complexes from heterogeneous datasets. *Phil. Trans. R. Soc. B* **369**, 20130567. (doi:10.1098/rstb.2013.0567)
17. Millane RP, Chen JPJ. 2014 Aspects of direct phasing in femtosecond nanocrystallography. *Phil. Trans. R. Soc. B* **369**, 20130498. (doi:10.1098/rstb.2013.0498)
18. Kirian RA, Bean RJ, Beyerlein KR, Yefanov OM, White TA, Barty A, Chapman HN. 2014 Phasing coherently illuminated nanocrystals bounded by

partial unit cells. *Phil. Trans. R. Soc. B* **369**, 20130331. (doi:10.1098/rstb.2013.0331)

19. Spence JCH, Zatsepin NA, Li C. 2014 Coherent convergent-beam time-resolved X-ray diffraction. *Phil. Trans. R. Soc. B* **369**, 20130325. (doi:10.1098/rstb.2013.0325)
20. Yefanov O, Gati C, Bourenkov G, Kirian RA, White TA, Spence JCH, Chapman HN, Barty A. 2014 Mapping the continuous reciprocal space intensity distribution of X-ray serial crystallography. *Phil. Trans. R. Soc. B* **369**, 20130333. (doi:10.1098/rstb.2013.0333)
21. Kupitz C, Grotjohann I, Conrad CE, Roy-Chowdhury S, Fromme R, Fromme P. 2014 Microcrystallization techniques for serial femtosecond crystallography using photosystem II from *Thermosynechococcus elongatus* as a model system. *Phil. Trans. R. Soc. B* **369**, 20130316. (doi:10.1098/rstb.2013.0316)
22. Caffrey M, Li D, Howe N, Shah STA. 2014 'Hit and run' serial femtosecond crystallography of a membrane kinase in the lipid cubic phase. *Phil. Trans. R. Soc. B* **369**, 20130621. (doi:10.1098/rstb.2013.0621)
23. Gallat F-X *et al*. 2014 *In vivo* crystallography at X-ray free-electron lasers: the next generation of structural biology? *Phil. Trans. R. Soc. B* **369**, 20130497. (doi:10.1098/rstb.2013.0497)
24. Stevenson HP, DePonte DP, Makhov AM, Conway JF, Zeldin OB, Boutet S, Calero G, Cohen AE. 2014 Transmission electron microscopy as a tool for nanocrystal characterization pre- and post-injector. *Phil. Trans. R. Soc. B* **369**, 20130322. (doi:10.1098/rstb.2013.0322)
25. White TA. 2014 Post-refinement method for snapshot serial crystallography. *Phil. Trans. R. Soc. B* **369**, 20130330. (doi:10.1098/rstb.2013.0330)
26. Denes P. 2014 Two-dimensional imaging detectors for structural biology with X-ray lasers. *Phil. Trans. R. Soc. B* **369**, 20130334. (doi:10.1098/rstb.2013.0334)