crystals

MDPI

Article

Remote Access Revolution: Chemical Crystallographers Enter a New Era at Diamond Light Source Beamline I19

Natalie T. Johnson, Paul G. Waddell, William Clegg and Michael R. Probert *

Chemistry, School of Natural and Environmental Sciences, Newcastle University, Bedson Building, Newcastle upon Tyne NE1 7RU, UK; n.johnson5@ncl.ac.uk (N.T.J.); paul.waddell@ncl.ac.uk (P.G.W.); bill.clegg@ncl.ac.uk (W.C.)

* Correspondence: michael.probert@ncl.ac.uk; Tel.: +44-191-208-6641

Academic Editor: Helmut Cölfen
Received: 6 November 2017; Accepted: 30 November 2017; Published: 5 December 2017

Abstract: Since the inception of the use of synchrotron radiation in the structural characterisation of crystalline materials by single-crystal diffraction in the late 20th century, the field has undergone an explosion of technological developments. These cover all aspects of the experiments performed, from the construction of the storage rings and insertion devices, to the end user functionalities in the experimental hutches. Developments in automation have most frequently been driven by the macromolecular crystallography community. The drive towards greater access to ever-brighter X-ray sources has benefited the entire field. Herein, we detail the revolution that is now occurring within the chemical crystallography community, utilising many of the tools developed by their more biologically oriented colleagues, along with specialised functionalities that are tailored to the small-molecule world. We discuss the benefits of utilising the advanced features of Diamond Light Source beamline I19 in the newly developed remote access mode and the step-change in productivity that can be established as a result.

Keywords: remote access; synchrotron radiation; chemical crystallography; automation; data handling; methodology; enhanced productivity

1. Introduction

Single-crystal X-ray diffraction remains the gold standard of analytical techniques, with an ever-expanding solid-state landscape being scrutinised via this method. With the increase in complexity of the chemical entities being studied, the size of individual crystals in samples is now often found to be outside the capabilities of the home laboratory. The increase in X-ray flux densities from modern home sources, rotating anodes [1], micro-focus tubes [2], multi-layer optics [3], and finally, liquid anodes [4] has somewhat alleviated this problem. However, an increasing number of samples require far more intense incident radiation to provide satisfactory results in the form of structural models of the crystalline state. This radiation can be found only at advanced central facilities. Many of these facilities, particularly at third-generation synchrotron radiation sources, have been optimised for specific experimental procedures such as high-throughput macromolecular crystallography (MMX) analysis or extreme in-situ condition generation. Diamond Light Source (DLS), the central synchrotron facility in the United Kingdom and used by international research groups, is similarly equipped, but it additionally has one oversubscribed beamline dedicated to small-molecule crystallography (SMX), the undulator-driven I19, which has recently been subject to a major hardware and software upgrade [5]. This beamline, initially inspired by the pioneering work at station 9.8 of the UK Synchrotron Radiation Source (SRS) at Daresbury Laboratories in the 1990s [6,7], has several features that are specific to the scientific questions being posed, and are detailed in the companion report by Allan et al. in this journal special issue [5]. The beamline has also recently been adapted to enable operation from a remote location, removing the need for user groups to be present on-site during scheduled experiment time. This remote operation has become increasingly the modus operandi for MMX users in recent years, but has so far been largely neglected by the SMX community. The reasons for this are varied, but are often grounded in the differences in the experimental procedures between the MMX and SMX user groups, particularly in the resolution requirements for a successful, publishable structure to be obtained from the experiment. SMX crystals are typically much more durable than the MMX equivalents (while at the same time being subject to a wider range of chemical and environmental sensitivities), in that the samples can usually withstand the far greater exposure to high flux densities provided by modern synchrotron beamlines. This durability allows the samples to be irradiated for rather longer periods of time in a variety of angular positions, often enabling extended analysis of diffraction space and higher resolution data to be recorded from one crystal without major radiation damage. Additionally the generally higher quality of SMX crystals, with far lower mosaicity, than protein crystals means the diffraction patterns contain a combination of very sharp intense reflections with weaker signals, thus requiring a different approach to exposure rates in view of modern detector capabilities. The experimental parameters for SMX crystals have, until very recently, dictated data collection times at beamline I19 of the order of 40–60 min using a conventional CCD detector and kappa-geometry diffractometer. This timeframe meant that the impact of on-site manual sample loading and repeated experimental hutch entries upon the scheduled beamtime was not significant. The improved instrumentation now available at I19 has caused a step change in the expectations of beamtime usage. Data collection times have been reduced to approximately 5–15 min of X-ray exposure for full atomic resolution of typical crystalline samples—in addition to the (necessarily somewhat variable) time required to screen individual crystals by optical centring and examination of the initial data images. Under these revised circumstances, the previous mode of operation represents a major time overhead for sample manipulation and exchange, so it is no longer fit for purpose and a change to protocols similar to those employed in the MMX world is required for efficient and cost-effective operation. The new working protocols are discussed in detail herein, from a user perspective, with additional commentary on variations of data processing approaches.

2. Remote Access in Development

The prerequisites for full remote access experimental control were not all present in the initial design, construction, and operational mode of DLS beamline I19; some have been introduced gradually since 2008, and others have been put in place through the 2016 major upgrade. With a view to future plans and developments, we have been involved in preparing for remote access in recent years, in close collaboration with the beamline scientists.

Cryogenic diffraction data collection has been available as an option from the beginning of I19 user experiments, and has been largely regarded as a routine and standard process by most users. The introduction of robotic sample exchange came later, followed by cryogenic sample storage in a robot-accessible liquid nitrogen dewar in the experimental hutch. At this point, it became possible to pre-mount batches of individual crystals, immediately store them in pucks in liquid nitrogen, and have confidence that the mounting would remain mechanically secure and protected from the atmosphere until X-ray investigation. This meant it was possible, well before the beamline upgrade, to practise, optimise, and routinely introduce these procedures, which themselves generated small but significant savings of time at the beamline.

The next step was to transfer the crystal cryo-mounting exercise from Diamond to the home laboratory, using equipment similar to that in use on MMX beamlines but in a variant design compatible with the existing I19 robot and sample dewar. This has the advantage that, once samples are initially screened using in-house diffractometers and a decision made that they require synchrotron radiation, suitable crystals can immediately be selected, mounted, and cryogenically stored, ready for the next allocation of beamtime, each new batch of samples being added to the puck(s) and shipping dewar. The full set of pre-mounted samples was then either transported to Diamond at the time of the beamtime visit along with the user team, or sent in advance by courier; the latter approach provided a useful test of the shipping procedures and protocols, as well as a demonstration that the equipment was being used appropriately and potential problems such as ice formation on the samples was avoided—first attempts were not all successful, and led to refinements in the methods.

Following the beamline upgrade, and after a first visit of the full normal team of users to gain familiarity with the new operations, we undertook two experiments in which some team members travelled to Diamond, with pre-mounted samples and/or sending them by courier in advance, while others remained in Newcastle for the first remote access trials. Although some local intervention (by users, the beamline staff, and software engineers) was required at first to deal with minor problems in the procedures, it quickly proved possible to conduct complete experiments from robotic crystal mounting through alignment and data collection to dismounting entirely by remote access. Late 2017 saw the first completely remote access operation, in which the samples were pre-mounted and shipped by courier, and the entire user team remained in the home laboratory. Diffraction data were converted to other formats as desired by automated scripts running on computing clusters at Diamond, and were downloaded by standard data transfer protocols as each experiment was completed.

3. Remote Access in Practice

The remote access protocols can essentially be broken down into three discrete consecutive operations, following an outline broadly similar to standard experiments completed in-house: sample loading and transportation (which occurs in advance of the scheduled beamtime); data collection (operated entirely by remote access, covering all the processes familiar to users working on site except for bulk sample loading); data processing and interpretation (which can begin in parallel with data collection and continues afterwards). These operations are described in turn herein, with the advantages of the new protocols being detailed in terms of both efficiency and scientific endeavour.

3.1. Sample Loading and Transportation

The traditional mounting methods developed in recent years, using magnetic bases fitted with Kapton micro-mounts for crystals, are fully transferable to the remote access mode of operation. A small amount of additional equipment for the loading and transport of materials is required; this may be owned by the user group (which is our preference), or it can be borrowed from the beamline, and information from the beamline staff should be sought well in advance of any scheduled time. After mounting, using appropriate inert-oil media and manipulations that depend on the properties of the samples with respect to possible chemical reaction or solvent loss on exposure to the atmosphere, the crystals are immediately frozen in liquid nitrogen in a Unipuck [8] holder that is compatible with the robotic sample handler installed at beamline I19. This part of the protocol differs from the approach taken by the MMX community where, due to the hydration content of their samples, crystals are usually immersed in a cryo-protectant before flash-cooling. This additional step is not required, due to the relative robustness and different properties of SMX samples. Once frozen, samples are protected from atmospheric conditions by vitrification of the mounting oil and by the liquid nitrogen; they are essentially stored in an inert environment that is suitable even for samples of extreme air-sensitivity—we frequently work with highly reactive organometallic species. Unipucks are designed to hold 16 samples per puck (see Figure 1); these, once filled, are transported to DLS in a specially designed dry shipping dewar that can hold up to 7 pucks (i.e., a total of 112 crystals) and can maintain an inert atmosphere at liquid nitrogen temperature for weeks if unopened. In practice, we have found that it is advisable to mount a minimum of 2 crystals for each sample submitted for study (i.e., 56 samples per shipping dewar), in order to allow for occasional problems with the mounting.

Figure 1. Puck loaded with 16 single crystal samples; image taken after synchrotron experiment and subsequent warming to room temperature.

There are significant advantages to dry-shipping individually mounted crystals rather than taking bulk samples to the synchrotron. Samples loaded in the home laboratory benefit from users' access to familiar experimental arrangements and standard operational tools, which generally leads to sample loading efficiency and reduced problems of degradation of material through solvent loss or exposure to oxygen- or moisture-containing environments. The pre-mounting also takes place without the time pressure associated with the limited experimental time allocated at the central facility. Additionally, vastly less material is transported (individual crystals for synchrotron study have dimensions usually measured in microns), and this is done in fully sealed units using established protocols agreed with the shipping couriers rather than transporting bulk samples, possibly in mother liquor, where all COSHH implications are at best estimated from the constituent components. Thus, the risk of public exposure to unknown chemical substances is significantly reduced, particularly if this means that chemical samples of unknown hazards are not carried on public transport.

The proposed protocol, however, should not be considered a panacea for samples, as there remains a small subset of crystalline materials for which it is not appropriate. These tend to be the samples that have a catastrophic phase transition between room temperature and 77 K, or those for which the proposed experiment is to investigate irreversible temperature-dependent properties.

3.2. Data Collection

On arrival at DLS, samples are handled by the beamline staff or the Experimental Hall Coordinators (EHCs); such procedures have been routine for years on MMX beamlines. The samples are transferred at the appropriate time from the dry shipper, after filling with liquid nitrogen, to the cryogenic dewar inside Experiment Hutch 1 (EH1). They can be selected in turn from this dewar and mounted on the goniometer by the robotic sample handler (see Figure 2), and the only personnel intervention requiring hutch entry is the initial setting up.

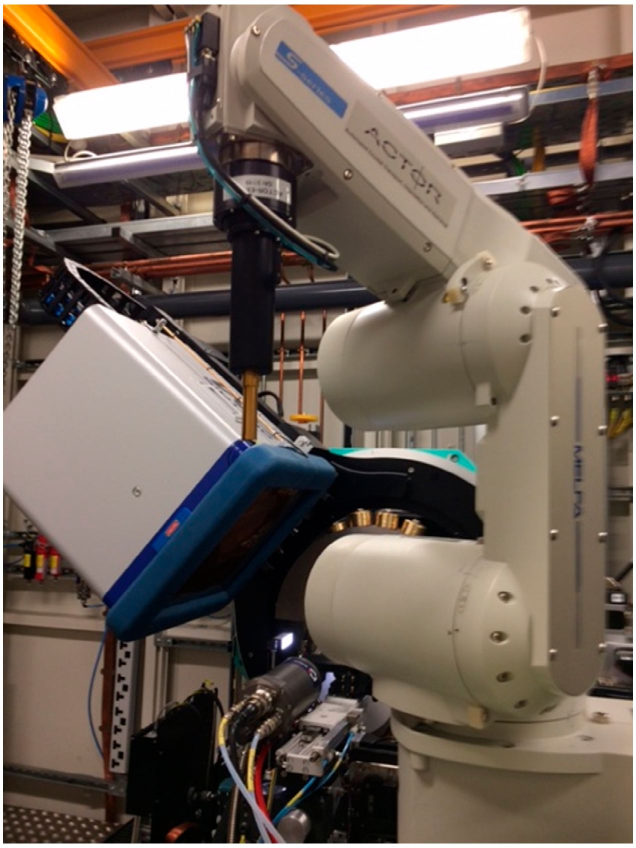

Figure 2. The ACTOR robot mounting device in place on beamline I19, DLS; the Pilatus detector, part of the goniometer, and the Cryostream sample cooler can be seen in the background.

The current dewar within EH1 can hold 5 pucks at any given time; therefore, in the extreme case of each puck containing a single crystal for a different sample, without any duplicates, 80 different samples can be examined before the beam has to be interrupted for re-entry of the hutch. This process significantly increases the usable experimental time in any given shift allocation, due to reduced overheads per diffraction experiment, and it is also operationally much less tiring and stressful for users.

The parameters for data collection on beamline I19 under standard operating conditions are well understood, and are described in the paper of Allan et al. [5], as well as in beamline user manuals. The remote operation of the beamline from the users' home laboratory (or indeed from their home) should present no noticeable differences from the arrangement whereby the user is present in person in the beamline Control Room. The connection protocols to allow remote access are described in full on the DLS website, but consist mainly of making a connection to the beamline using the NOMACHINE [9] remote desktop software. The only major difference to the user experience from an operational perspective is that the 'Baton', designating which computer currently holds the control directives for the instruments, must be passed explicitly from the beamline to the user.

Due to the desktop real-estate usage of the beamline software, it is advisable to connect from a workstation that has multiple large displays (see Figure 3). Additionally, due to the bandwidth requirements of a large desktop with many components, it is important to conduct the remote access from a workstation with a high-speed internet connection; however, in the authors' experience, problems with connection loss have never significantly affected data collections and merely require the re-establishment of the link.

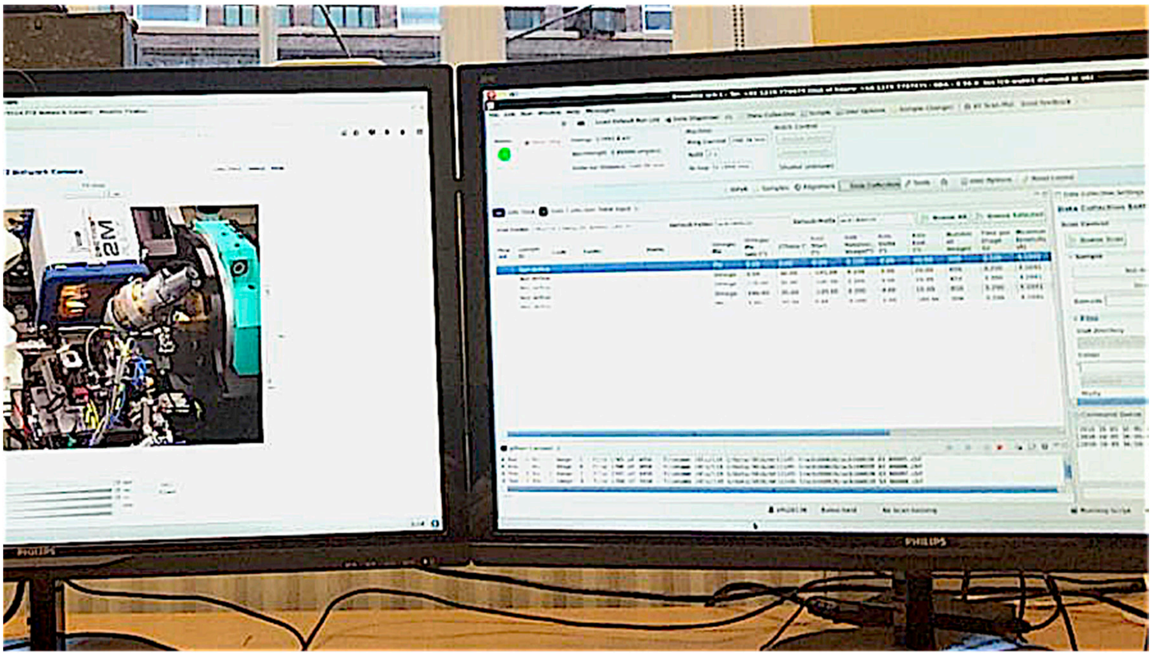

Figure 3. Remote desktop connection to beamline I19, DLS from Newcastle University, UK; visible here are a webcam image of the goniometer and detector, and a data collection in process.

During our development of these procedures since the upgrade of the beamline, we have identified particular bottlenecks and limitations in the remote access protocols, and these have been swiftly investigated and satisfactorily dealt with by beamline and software staff at Diamond. Most of the problems have been associated with graphical displays, such as the viewing of individual diffraction images and the use of webcams inside the hutch. Data collection itself makes little demand on the connection bandwidth, as the raw data are initially stored on site using Diamond's fast internal network, and only a brief running commentary is output to the user's screen.

Due to the well-debugged protocols previously developed by the MMX community, for this style of beamline operation, the user is afforded a near-seamless experience similar to any previous visit under the older system operation. One significant difference does present itself due to the increased efficency of use of the beamline and that is the rate at which data are recorded, and therefore the rate at which concurrent analysis can occur. The authors have found that, if operating in the most optimised configuration, it is possible to perform only a crude level of analysis of the data concurrently with data collection during the allocated beamtime. This is significantly helped by the automated data processing pipeline that has been built into the standard beamline operation protocols as part of the recent upgrade. However, most users will be well aware that samples are studied at DLS either due to a very specific scientific question being investigated or because samples do not provide sufficient diffraction intensity with home laboratory instrumentation. In both of these cases, it is not uncommon for data treatment to require additional expert user effort and initiative, reducing the chances of success of the automated routines.

3.3. Data Processing and Interpretation

With the upgrade to the instrumentation and the advances in the rate at which data can be collected, the data processing now has the potential to be the rate-limiting factor in the determination of structures from SMX synchrotron data, and therefore, new approaches have to be considered. The first of these is the aforementioned automatic data-handling pipeline, although the success of this varies significantly depending on the samples being analysed, particularly when issues such as crystal twinning (or other multiple-component samples) and major structural disorder are encountered.

A more fundamental aspect of general data processing and interpretation is the likelihood of success, and the extraction of optimum processed data from any raw dataset is somewhat dependent on the familiarity of the user with the processing software; this is particularly true for samples that have provided probematic data. The automated data pipeline handles routine cases very well, but usually struggles with increasing non-standard issues with the data. These situations are where user experience is key to the final success of the experiment. Therefore, it is important that data can always be handled in a way that is consistent with the users' experience already established outside the synchrotron environment.

To aid the processing of data for users, diffraction images can be imported into different processing packages (whether public-domain or proprietary) or converted into file formats that are consistent with the users' previous experience and preference. Conversion routines [10], including all conversions of goniometer setting angles, allowing processing with commercially available software, have been made available, both by us and the beamline staff, on beamline I19. The conversion of the images can be spawned to centralised computer services within DLS and occurs at a rate that is consistent with the data collection times for 'standard' experiments, avoiding a backlog of unconverted images.

The combination of automated and different available algorithms allows users to have a large degree of confidence in the data collections on each sample as they are proceeding. In essence, use of remote access, rather than on-site operation, makes no difference to the quality of data obtained at the beamline. As is usually the case with single-crystal samples taken to the synchrotron facility, data quality varies considerably; the best data give statistics and structural results similar to those obtained in the home laboratory for more strongly diffracting samples.

As these developments have taken place since the 2016 upgrade of the beamline, and our first completely remote-access experiment was very recent, few structural results derived from this work have yet been published, though a number of manuscripts are currently in preparation, or have been submitted for publication. The use of cryo-mounting, courier shipping, and partial remote-access operations featured in some recently reported work [11,12], but were not specifically identified as such in the publications, nor is there any reason why this should be evident by inspection of the results.

4. Consequences of Remote Access

The move from manual, on-site operation of beamline I19 can seem quite daunting to the inexperienced user, but significant advantages are gained for all parties concerned. The immediate benefit to users is the reduction/elimination in travel times to and from the facilities, often saving many hours and frequently days per annum. The efficiency of use of the beamline increases dramatically as soon as users adopt the full robotic mounting procedures with the additional gains that the chance of error of operation is reduced, by limiting the required access to the Experimental Hutch. In the ever-tightening economic conditions of the science landscape, DLS benefits from reduced costs of operation, not only in expenses for support of accommodation and travel, but also in diminished operational overheads in user support, management and security. Further increases in efficiency of the beamline operation are planned through the introduction in the future of shorter shift allocations (few user groups can fill 24 h of continuous beamtime with the greatly increased data collection speed introduced by the beamline upgrade), the ability to pass the operating baton between consecutively scheduled users and between individual users in user group consortia ('Block Allocation Groups' in Diamond terminology) who may be at geographically distant institutions, and similar situations that can be truly effective only if remote access protocols are adopted by an increasing user base.

With the viability and reliability of remote access to I19 for what may be regarded as 'standard' single-wavelength cryogenic ambient-pressure data collection procedures now fully demonstrated and assured following substantial preparatory developments and a completely successful extended set of data collections, it is expected that the remote access approach will be documented and promoted as increasingly the normal mode of operation, and will lead to a significant improvement in the effective exploitation of this major and popular crystallographic facility. In principle, similar procedures can be introduced at other central facilities.

Remote access operation and automation of experimental and computational procedures for chemical crystallography single-crystal diffraction beamlines such as I19, at Diamond Light Source, still have potential for further enhancement. Modifications to protocols to enhance efficiencies in the procedures, already developed and in use in the MMX field, are planned. While there is some small usage of MMX beamlines by 'small molecule' crystallographers (indeed, we began our own development and exploitation of synchrotron facilities by such use of one of Daresbury Laboratory's MMX beamlines, station 9.6, in the 1990s), their setup is not ideal for atomic-resolution data collection of relatively small structures. Access to higher Bragg angles is usually restricted or non-existent with fixed detectors. Data completeness can be compromised, particularly for low-symmetry space groups, on MMX instruments that utilise diffractometers with a single rotation axis. Additionally they make use of routines for the automation of optical centring, diffraction screening, and data collection strategy that are based on the typical properties of MMX crystals, which are quite different from those of most 'small molecule' structures. What we describe here is, however, a significant step on the way, making remote access a realistic operation for the first time for chemical crystallography beamlines, and demonstrating the desirability of continuing the evolution towards full unsupervised automation.

Acknowledgments: The authors are indebted to the beamline staff at I19 for their continued support of our drive towards the now fully operational remote access state of the beamline. We are grateful for the support of the software support team at DLS, who have worked to incorporate additional features into the control software to ease the transition to remote access, and to the senior management of DLS for their encouragement and enthusiasm for this project. Finally, the authors thank their Durham University partners in the North-East England Chemistry BAG for sharing this journey of development, and DLS for continued access to beamline I19 in pursuit of important structural science before, during and since the remote access implementation.

Author Contributions: Michael R. Probert and William Clegg have been responsible for obtaining synchrotron beamtime at Diamond Light Source I19 for these and other experiments. Michael R. Probert and Paul G. Waddell have optimised the sample mounting and shipping procedures. Natalie T. Johnson has written diffraction image format conversion software. All authors have carried out the data collection experiments described here, both on site and by remote access and additionally contributed to the written document and preparation of publication material.

Conflicts of Interest: The authors declare no conflict of interest.

References

1. Longley, W.; Miller, R. A simple rotating anode X-ray generator. *Rev. Sci. Instrum.* **1975**, *46*, 30–32. [CrossRef]
2. Incoatec GmbH. The High-End Incoatec Microfocus X-ray Source—Unique Efficiency and Brightness. Available online: https://www.incoatec.de/ (accessed on November 2017).
3. Schuster, M.; Gobel, H. Calculation of improvement to HRXRD system throughput using curved graded multilayers. *J. Phys. D Appl. Phys.* **1995**, *28*, A270–A275. [CrossRef]
4. Hemberg, O.; Otendal, M.; Hertz, H.M. Liquid-metal-jet anode electron-impact X-ray source. *Appl. Phys. Lett.* **2003**, *83*, 1483–1485. [CrossRef]
5. Allan, D.R.; Nowell, H.; Barnett, S.A.; Warren, M.R.; Wilcox, A.; Christensen, J.; Saunders, L.K.; Peach, A.; Hooper, M.T.; Zaja, L.; et al. A Novel Dual Air-Bearing Fixed-χ Diffractometer for Small-Molecule Single-Crystal X-ray Diffraction on Beamline I19 at Diamond Light Source. *Crystals* **2017**, *7*, 336. [CrossRef]
6. Cernik, R.J.; Clegg, W.; Catlow, C.R.A.; Bushnell-Wye, G.; Flaherty, J.V.; Greaves, G.N.; Burrows, I.; Taylor, D.J.; Teat, S.J.; Hamichi, M. A new high-flux chemical and materials crystallography station at the SRS Daresbury. 1. Design, construction and test results. *J. Synchrotron Radiat.* **1997**, *4*, 279–286. [CrossRef] [PubMed]
7. Clegg, W. Synchrotron chemical crystallography. *J. Chem. Soc. Dalton Trans* **2000**, 3223–3232. [CrossRef]
8. MiTeGen, LLC. Universal Pucks and Tools. Available online: https://www.mitegen.com/product/universal-pucks-and-tools/ (accessed on November 2017).
9. NoMachine. Available online: https://www.nomachine.com/ (accessed on November 2017).
10. Johnson, N.T.; Probert, M.R. cbf_to_sfrm.py (Alpha Version). 2016. Available online: https://github.com/nu-xtal-tools/cbf_to_sfrm (accessed on November 2017).

11. Lamming, G.; Kolokotroni, J.; Harrison, T.; Penfold, T.J.; Clegg, W.; Waddell, P.G.; Probert, M.R.; Houlton, A. Structural diversity and argentophilic interactions in one-dimensional silver-based coordination polymers. *Cryst. Growth Des.* **2017**, *17*, 5753–5763. [CrossRef]
12. Maddock, L.C.H.; Nixon, T.; Kennedy, A.R.; Probert, M.R.; Clegg, W.; Hevia, E. Utilising sodium-mediated ferration for regioselective functionalization of fluoroarenes via C–H and C–F bond activations. *Angew. Chem. Int. Ed.* **2017**. [CrossRef]