

INTERNATIONAL UNION OF CRYSTALLOGRAPHY

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# IUCr Newsletter

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Original article text and available illustrations

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## LETTER FROM THE PRESIDENT

# Contributing to science and society

Sven Lidin



It has been an extraordinary spring and although the world is slowly opening up, we are far from normality and I expect that the world has been permanently changed from our shared experience of the COVID-19 pandemic. We have converted to online teaching and meeting, and we have a much better understanding today of what that entails, in particular the weakness of digital socializing. We can meet online and efficiently make decisions, but these meetings are seldom sources of energy.

I hope we will find that we can travel less and still maintain cohesion in our international community, but I am convinced that we cannot do without physical meetings altogether. In the post-corona age we may be better equipped to strike a balance between necessary travel and online meetings.

The triennial Congress and General Assembly of the IUCr has been postponed for a year and we are very lucky that the Prague organizers were able to book a slot for the same time period in 2021. This required some reorganization of other meetings and we are most grateful for the cooperation of everyone involved.

The importance of a fact-based world view has become painfully clear during a period when the political systems throughout the world have been under pressure in new and unexpected ways. How to act during a world pandemic is a very complex question, but the importance of facts as the basis for political decisions is clearer than ever and our science contributes to this in important ways.

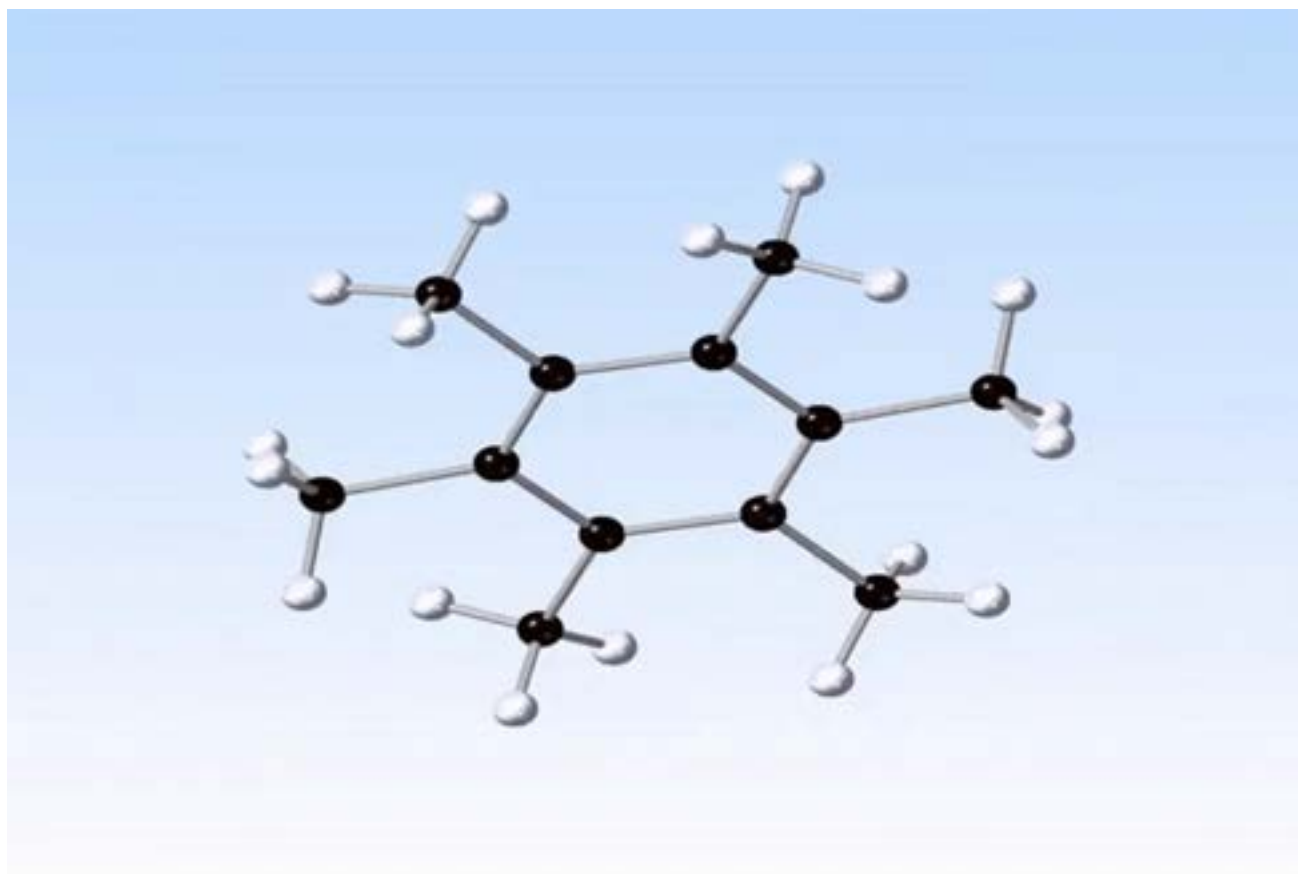
The last issue of the *IUCr Newsletter* contained a [Letter to the Editor](#) in which Marv Hackert related the fantastic story about how the structure of the spike protein of the new coronavirus was solved in record

time, starting from just the sequence – a wonderful benchmark of methodology and an excellent example of what we can contribute to science and society.

## EDITORIAL

# Editorial

Mike Glazer



I guess that as I write, most of us are still under lockdown or at least being very careful to avoid exposure from the SARS CoV-2 virus. The COVID crisis continues without an end in sight; maybe a search for a vaccine or a treatment, at least, will be found. But we can expect to be living with this virus for a very long time. It is important, however, to note that if a vaccine is to be developed it will be largely thanks to the efforts of many crystallographers working in macromolecular studies around the world, for it is our subject that helps to elucidate the structure of SARS-CoV-2 and its various associated proteins and enzymes. To illustrate the importance of crystallography in this field, we are publishing in this issue of the *IUCr Newsletter* a [series of excellent articles](#) on developments in the coronavirus research using advanced sources, thanks to our editors Tiffany L. Kinnibrugh and Abel Moreno.

On another topic, suppose I ask you: who discovered X-rays? Of course, you will be shouting ROENTGEN 1895! However, I have seen over many years that great discoveries are often not quite as they seem and are not necessarily the work of a single individual. Generally, there are other persons involved, some even making earlier discoveries, persons who are largely forgotten or ignored. The story of Roentgen's accidental discovery is well documented and widely known. However, one must ask what is meant by "discovery"? Is it a well defined or a fuzzy notion?

Roentgen's discovery of X-rays crowned research into cathode rays in which many people had been involved and contributed. Some of them were hot on the track, some were less successful or wrong in their ideas, but not only successful experiments can inspire a discovery.

A good example of the difficulty in pinning down who did what is offered by the career and work of a Ukrainian-born electrical engineer, Ivan (Johann) Puluj, and we publish [here](#) a study of his life and works, written especially for us by an historian of science, Emilie Těšínská. It is clear that some 15 years before Roentgen's work, Puluj had been researching cathode ray tubes and had invented his so-called Puluj lamp, in which an inserted screen coated with calcium sulfide glowed brightly. Some references on the internet even suggest that he had given one of his lamps to Roentgen. So, it is very likely that Puluj had been producing X-rays earlier but probably failed to realise the existence of these mysterious rays. Certainly, within months after Roentgen's announcement in 1895, Puluj was using his apparatus to take X-ray images of whole bodies of dead animals, and bone structures of superb quality, even exceeding those of Roentgen. Here Puluj surely profited from his deep knowledge and his practical work in electrical engineering. So, what do you think? Who "discovered" X-rays?

The IUCr has been in existence now for around 73 years, serving the crystallography community through its many journals and other publications, by providing grants, sponsoring students and lecturers, visiting professors, activity in Africa, and many, many more activities. Of course, you will be aware of the high quality of our journals. These are supported by highly dedicated staff at our Chester (UK) office, for whom we are always grateful. However, all this costs money, and no more importantly now given the global financial crisis that we are certainly about to suffer. In this issue of the *Newsletter*, the Convenor of the Finance Committee, Malcolm Cooper, [explains the ins and outs of this critical IUCr Committee](#). Wrong decisions by this Committee could have profound implications for the prospects of the IUCr.

As an example of the international nature of crystallography, Elena Boldyreva and colleagues [report on a successful scientific student conference](#) held in April of this year at the Novosibirsk State University. This is the kind of conference – online, global and interdisciplinary – that is now becoming commonplace. It is wonderful that the development of software tools such as Zoom, or Teams is enabling students and scientists to get together while sitting in their own homes. As another example, recently, I was asked by Sandro Scandolo at the ICTP (International Centre for Theoretical Physics, Trieste) to give a virtual plenary lecture entitled "Why Crystallography?" as part of an online course called Solid State Physics in Quarantine. It was an odd feeling to be sitting at home and talking to an audience of several hundreds around different parts of the world.

Sometimes it is worth recalling just how difficult it used to be to work in crystallography, especially as these days we have such fantastic automatic, computer-controlled systems. Jon Cooper [describes one of the first commercial diffractometers in the UK](#). This so-called linear diffractometer was used to elucidate the structure of lysozyme, the first enzyme structure to be determined. So, the linear diffractometer was a significant advance, although I remember that it was a fearful beast that was difficult to use. It is worth recalling that after W. H. Bragg built his ionisation spectrometer, the forerunner to the modern diffractometer, almost all X-ray diffraction was carried out using film techniques. I recall how, by using film methods, we had to become proficient in orienting crystals and finding our way in reciprocal space: crystallographers today are spared from having to do this.

Following on from this, the fantastic efficiencies of modern diffractometers have meant that huge numbers of crystal structures are being determined, and many are not published. The whole question of publishing this material is raised by Bill Clegg [here](#). It is a tricky problem to solve. Bill asks controversial questions: Why publish? Why not publish?

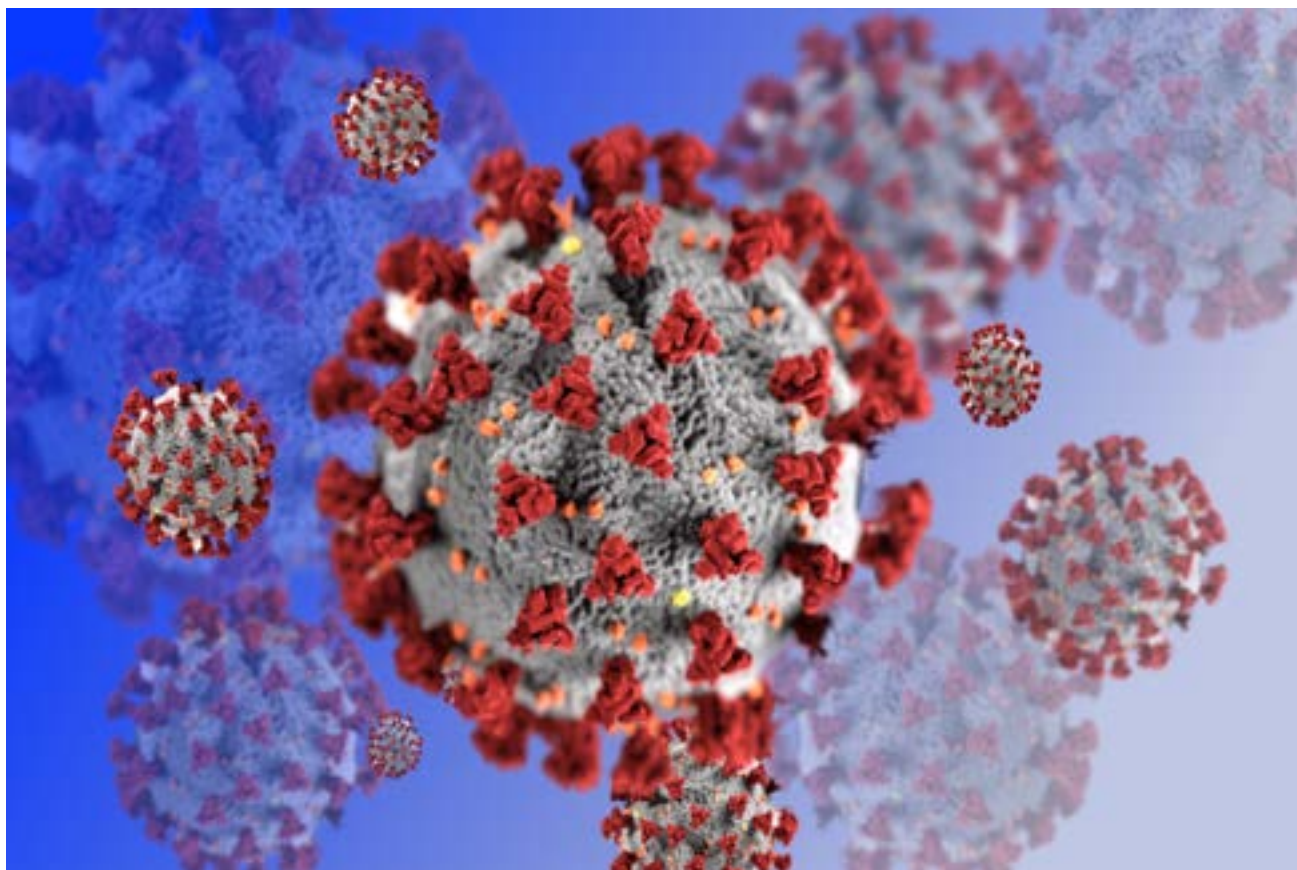
Finally, in the last *Newsletter* we published a list of [Biographical Memoirs of the Royal Society](#) where you can read the life histories of many famous scientists with a crystallographic connection. Recently, the Royal Society has added one on [J. F. Nye](#). He worked in many areas of science, including studies of ice and of dislocations in crystals. Many of you will be familiar with his classic textbook *Physical Properties of Crystals*.

Stay safe!

## RESEARCH NEWS

# An introduction to crystallography research on SARS-COV-2 (COVID-19) via X-ray diffraction at synchrotrons around the world

Tiffany L. KinnibrughAbel Moreno



Since the Middle Ages, human beings have faced different pandemics caused by both bacteria and viruses. The last two terrible pandemics happened over an interval of approximately 100 years: first, the Spanish influenza pandemic (1918–1920) was one of the deadliest humans have ever had to face, and second, the ongoing SARS-CoV-2, not as deadly as the previous pandemic, but highly expansive and contagious. Spanish influenza killed, at the time, more than 40 million people around the world. This might have been due to the lack of the many facilities in biotechnology to develop vaccines [1] – something that we do not lack nowadays. More recently, the severe acute respiratory syndromes SARS, MERS (both caused by coronaviruses) and swine flu (caused by an influenza virus), among others, have caused the deaths of millions of people around the world [2]. In the present case, the RNA virus called SARS-CoV-2 has spread at a high speed, infecting people from nearly all countries in the world in a very short time [3]. During the last few months, this virus has not only killed people of different ages (though mostly adults), but it has also affected us socially and economically by changing the world through measures such as lockdowns, self-isolation and social distancing, causing unprecedented global disruption of a magnitude never imagined before [4]. However, this is probably the first time that the world is working together to beat a common enemy. Scientists from all over the world are working

against the clock to develop a vaccine or drugs against this virus based on structural research (X-ray and cryo-EM) and molecular biology techniques [5–7]. The development of drugs uses the knowledge already collected from other severe acute respiratory syndromes, AIDS and diseases such as malaria. This knowledge was based on structural data and 3D structures of proteins already solved. Additionally, the efforts of medical doctors, molecular biologists, biochemists, microbiologists, computational scientists and immunologists are immediately focused on knowing the particular characteristics of this virus. Chemical and biological approaches are aimed at sorting out this problem in as short a time as possible. In the Americas as well as in many other countries, some novel initiatives are focused on understanding the virus structure and its mechanism of action in different ways. All these initiatives are concentrating on getting the opportunity to develop some kind of drug against this virus or even a new vaccine.

Currently, the entire world is dealing with the search for a cure for the SARS-CoV-2 coronavirus pandemic [8–10]. In some countries, this virus has been apparently less aggressive than in others, leading people to speculate about the possibility of the virus being artificially made [11]. However, after some serious and careful investigations, most researchers do not accept this speculation, thus taking the line that this RNA virus evolved naturally. The term coronavirus comes from the existence of the spike protein (S), which is a 210–230 kDa glycoprotein with 23 potential N-glycosylation sites [10]. This glycoprotein binds to angiotensin-converting enzyme 2 (ACE2). These enzymes are receptors located in human cells, in the membrane protein surrounding the cell and in the respiratory tract [12, 13]. In humans, the SARS-CoV-2 strains typically prefer terminal NeuAc $\alpha$ 2-6Gal epitopes [neuraminic acid type and glycosidic linkage ( $\alpha$ 2-3 or  $\alpha$ 2-6), which are richly expressed along the upper respiratory tract]. The process is influenced by fucosylation. Afterwards, the immune system starts the detection response based on chemical recognition *via* specialized antigen-presenting cells. This, in a few words, is the pathway that this virus uses to get into the human body *via* the upper respiratory tract. This pathway also shows how the virus can be destroyed by a strong immune system (given all the favorable conditions in healthy patients). On the other hand, there are some clinical reports that suggest that patients with chronic medical conditions such as cardiovascular disease, diabetes and obesity are more likely to be infected by this virus [14]. Currently, there are no markers that predict the virus susceptibility and reinfection. Some compelling clinical reports have established that the ABO blood types, which are carbohydrate epitopes located on the surface of human cells, might play an important role in this chemical recognition. In Mexico, a few groups have been interested in this issue, and some initiatives are being supported by the National Council for Science and Technology (CONACYT). It is important to mention that during the SARS-CoV pandemic from 2003 to 2004, and possibly the present pandemic in 2020 as well, the most infected individuals were those with groups A and B, whereas those with group AB were only slightly affected. However, group O individuals seemed to be relatively resistant to this infection [15]. The interactions of different types of fucoses (specific for each ABO blood type) with the glycoprotein S and the ACE2 receptors could provide important insight into producing tailored drugs based on this stereo-specific interaction or chemical selectivity [16, 17]. There is even a hypothesis describing the seasonality and selective trends in viral acute respiratory tract infections. The effect of temperature causing a kind of seasonality is related to changes in temperature, assuming that low temperatures or host chilling activate dormant virions [18].

At the beginning of the pandemic, scientists focused on structure elucidation of macromolecules using powerful techniques, including X-ray crystallography. This is the most powerful technique for macromolecular structure determination, reaching quasi-atomic resolution in the most favorable cases, and without *a priori* limitations on the size and complexity of the molecules being studied [19, 20]. Use is made of the free-electron laser (FEL) with micro- to nano-sized crystals, which generates extremely intense X-ray pulses of tens of femtoseconds duration with nine-to-ten orders of magnitude higher peak brilliance than third-generation synchrotrons [21–23]. In addition, cryo-EM is used with near-atomic resolution for biomolecular structures [24, 25]. Many research facilities during the pandemic faced working under reduced operations, limiting research to COVID-19 only. There has been some progress on COVID-19, with the possibility of a viable vaccine being produced by the end of this year by

either private companies or even individual countries under the umbrella of the World Health Organization. The cost of the vaccine will not be comparable with the damage caused by this virus to the less-protected people in some societies. The following articles describe the research efforts of scientists from around the world in conjunction with synchrotron facilities.

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## RESEARCH NEWS

# Lighting the way for Australian researchers: COVID-19 research at the Australian Synchrotron

Rachel M. Williamson Alan Riboldi-Tunnicliffe

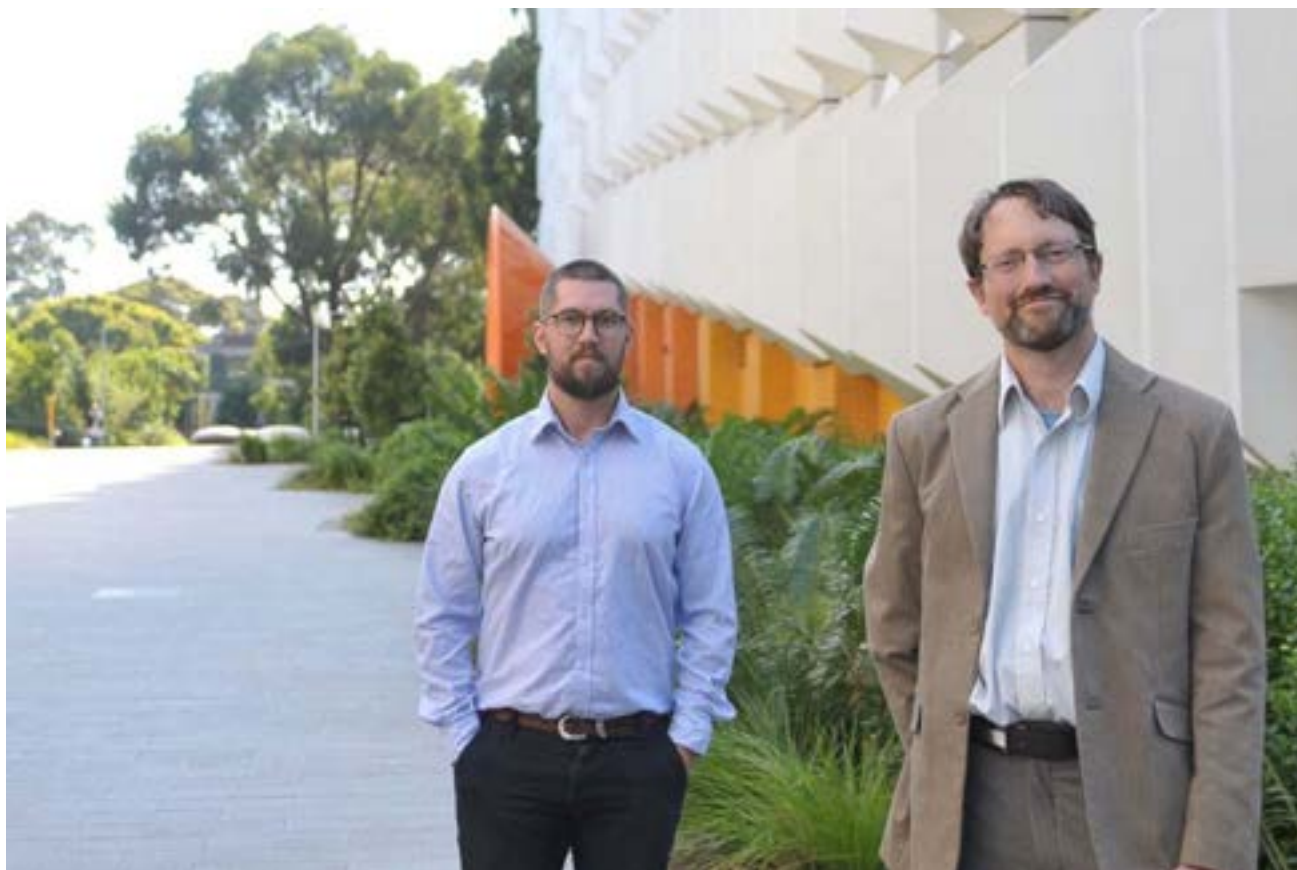


Figure 1. Ben Gully and Dene Littler at the Biomedicine Discovery Institute, Monash University.

As Australians returned to work from their summer holidays it became rapidly apparent that 2020 was going to be a year like no other. Faint reports of a strange new pneumonia in Wuhan, China, that had murmured in the global background while Australia endured the unprecedented summer bushfire season suddenly became a conflagration of their own. Australia identified its first case of confirmed COVID-19 on 25 January 2020, with events moving rapidly from that point onwards. Following Australian Government guidelines, the Australian Synchrotron moved to essential operations mode on Monday 23 March, cancelling all user experiments except for COVID-19-related research.

Like our sister synchrotrons around the world, we understood the vital importance of maintaining access to synchrotron radiation in driving structural-biology-related research into SARS-CoV-2 and made an early decision to reach out and support the efforts of our user community working in this field. In early March 2020 ANSTO/Australian Synchrotron offered a special rapid access mode to the MX2 Macromolecular Crystallography Beamline [1] for users undertaking COVID-19 research. This special rapid access mode would consist of short mini-shifts of beam time (a minimum of 2 hours), available on an “as-needed basis”. Proposal submission, review and user access processes would be fast-tracked. Our user community responded to the new special access mode with alacrity and agility, in many cases

rapidly transitioning into a new field of research. We have been delighted with the varied and productive nature of the research.

The Australian Synchrotron's user community ordinarily works across diverse fields of biology; this enabled many varied approaches to be undertaken with which to better understand SARS-CoV-2 and COVID-19 development, both *in vitro* and at a structural level, including research on both proteins from the SARS-CoV-2 virus alone and interactions between host and viral proteins.

The first SARS-CoV-2-related protein structure solved at the ANSTO/Australian Synchrotron was quickly published, and the resulting structure was made available immediately in the Protein Data Bank (PDB entry 6W9Q) by Dene Littler and colleagues from Monash University (Fig. 1) (Identification of a putative peptide-binding site at the dimer interface of the SARS-CoV-2 Nsp9 RNA-replicase. Littler, D. R., Gully, B. S. Colson, R. N. Riboldi-Tunnicliffe, A. & Rossjohn, J., to be published).

Dr Littler, working within the [laboratory of Jamie Rossjohn](#), has focused on the parts of the virus produced inside human cells (the non-structural proteins; Nsps). These act to trick our body into becoming viral factories, and their enzymatic nature results in reduced genetic drift as new viral strains arise. Dr Littler is particularly interested in the molecular details of the less well understood Nsps, determining how they function in the context of infection and whether their action is amenable to small-molecule inhibition.

No approved drugs have yet been designed that specifically target coronaviruses but pharmaceutical companies around the world have begun assessing how pre-existing compounds might be productively repurposed for this role. The most promising targets to date are generally agreed as being the Nsp12 RNA-polymerase and the Nsp5 protease. As RNA viruses are prone to genetic drift, combination therapies can be required to target simultaneously different aspects of the virus. Dr Littler and his co-workers have obtained some interesting laboratory results for Nsp9, a small conserved viral protein that was thought to be involved in RNA binding, although it is unclear how it does so. They have identified a potential protein-binding site within Nsp9 and are working to better understand its significance (Fig. 2). The initial structure report has been published via *bioRxiv* [3].

## SARS-CoV-2 RNA genome

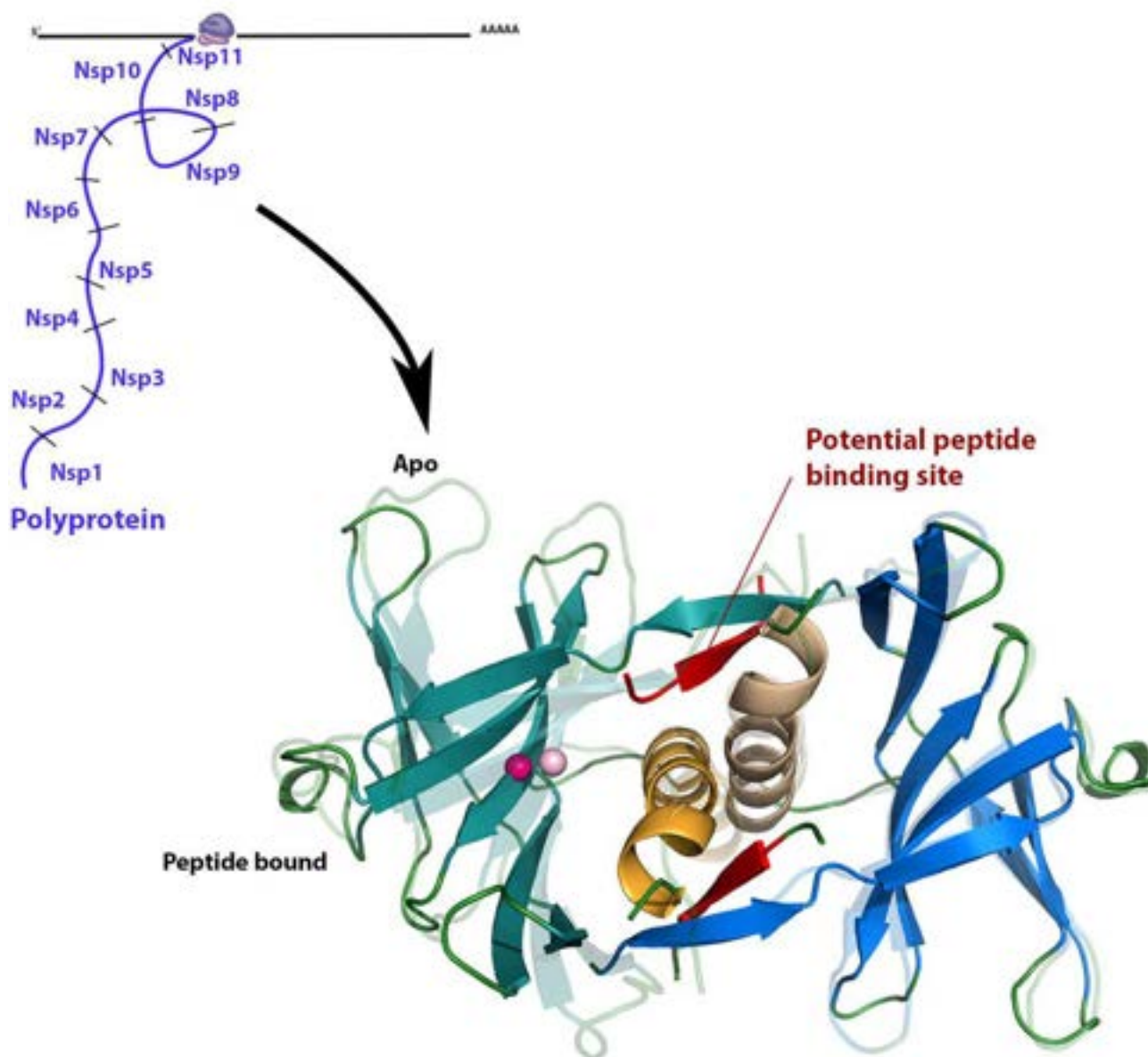


Figure 2. The structure of Nsp9 [2].

The [laboratory of Joel MacKay](#) has a proven track record in the research area of protein-protein interaction, and in particular the roles of these interactions in gene expression. A recent publication [4] revealed an important protein-protein interaction in cells infected with SARS-CoV-2.

To execute their program of replication, viruses hijack parts of the cellular machinery of their hosts. A prominent example of this phenomenon is the use that many viruses make of the host transcriptional apparatus to integrate and transcribe their genome. A recent analysis of the protein-protein interactome of the SARS-CoV-2 virus [4] has demonstrated that the viral envelope (E) protein can directly interact with the human proteins BRD2 and BRD4 (members of the BET family of bromodomain proteins). These bromodomains recognize acetylated lysines on histones and other proteins and are already promising therapeutic targets for the treatment of multiple conditions. These lead molecules available to date have moderate affinities and no paralogue specificity between BRD2, BRD4 and the related BRD3 and BRDT proteins.

In collaboration with Louise Walport from the Crick Institute in London and Hiroaki Suga in Tokyo, the MacKay laboratory is using an mRNA display of genetically reprogrammed cyclic peptides to design BET protein inhibitors with higher affinity and paralogue specificity. Their results to date demonstrate that

cyclic peptides can be generated which display sub-nanomolar affinities with specificities that are much higher than has previously been observed [5]. They are currently exploring the structural and binding diversity of the highly diverse set of peptides that have been selected in their screens. X-ray crystallography has been a central tool in this exploration process, and Karishma Patel from the McKay Group (Fig. 3) has used the Australian Synchrotron to determine the structures of several of these peptides in complex with their bromodomain targets.



Figure 3. Joel McKay and Karishma Patel at the University of Sydney.

The emergence of the SARS-CoV-2 virus in late 2019 provided a rare and unique opportunity for [Stephanie Gras and her group at Monash University](#) to expand their existing viral and structural immunology research interests and begin investigating the immune response towards an entirely novel virus. As a Viral and Structural Immunology group, Professor Stephanie Gras and her co-workers are focused on understanding T-cell responses towards different viruses through a structural lens. During a viral infection, viral peptides are presented from the immune system by proteins called human leukocyte antigens (HLA). These peptide-HLA (pHLA) complexes are displayed on the cell surface (like a flag) to be recognized by passing T cells. These T cells recognize the pHLA complex through their surface T-cell receptor (TCR), and the resulting immune response ensues. This process is complex, and Professor Gras knows from her previous work that understanding the presentation of peptides by HLA molecules and their interactions with T cells provides a wealth of knowledge about the resulting immune response. The ability to “see” the peptide presentation and recognition by the TCR using structural biology plays a significant role in understanding these interactions and is made possible utilizing X-ray crystallography at facilities such as the Australian Synchrotron. The expertise of the Gras group makes them ideally placed to understand the ability of the immune system to present various SARS-CoV-2 peptides (shared or not with SARS-CoV virus), and their ability to interact with the TCR. Understanding the ability and limitations of HLA molecules at presenting various SARS-CoV-2 derived peptides, their recognition by TCR and their impact on the immune response might provide valuable clues to aid in the design of effective therapeutics or a potential vaccine. Professor Gras’s group has solved the first structure of a SARS-CoV-2

peptide presented by an HLA molecule, and this will help them to better understand the T-cell response and participate in global efforts towards improving protective immunity against SARS-CoV-2.

The Commonwealth Science and Industry Research Organisation (CSIRO) is one of Australia's premier research facilities and, as such, has been heavily involved in [COVID-19 research](#) since the beginning of the outbreak. One area of CSIRO's research involving structural biology is being undertaken by Tom Peat and Alex Caputo. It is well established that viruses require host cellular functions in order to replicate and continue the infective process. As such, antiviral intervention can be targeted to either viral macromolecules (proteins or RNA/DNA) or specific host-cell proteins required for the viral life cycle. The latter strategy has advantages with respect to a larger barrier for drug resistance and the ability to target a broader range of viruses. Drs Peat and Caputo's colleagues at CSIRO have determined a set of host cellular factors required for Hendra virus replication [6, 7]. This work has now been expanded to show that some of these host proteins are required for the replication of viruses beyond the paramyxoviridae family. Blocking one of these host proteins, human fibrillarin [8–11], an S-adenosyl-L-methionine (SAM) dependent nucleolar methyltransferase, has been shown to decrease the replication of a range of clinically important paramyxoviruses, including the highly pathogenic Hendra and Nipah viruses by over 1000-fold, to the extent that virus replication is almost undetectable [10]. Drs Peat and Caputo performed a fragment screen by surface plasmon resonance (SPR) and crystallography to develop small-molecule inhibitors as antivirals. They discovered fragments with differing binding modalities and determined the crystal structures of several of these using the macromolecular crystallography beamlines at the Australian Synchrotron (Fig. 4). They are now in the process of building these fragments into drug-like compounds and determining whether these will be effective antiviral compounds for SARS-CoV-2.

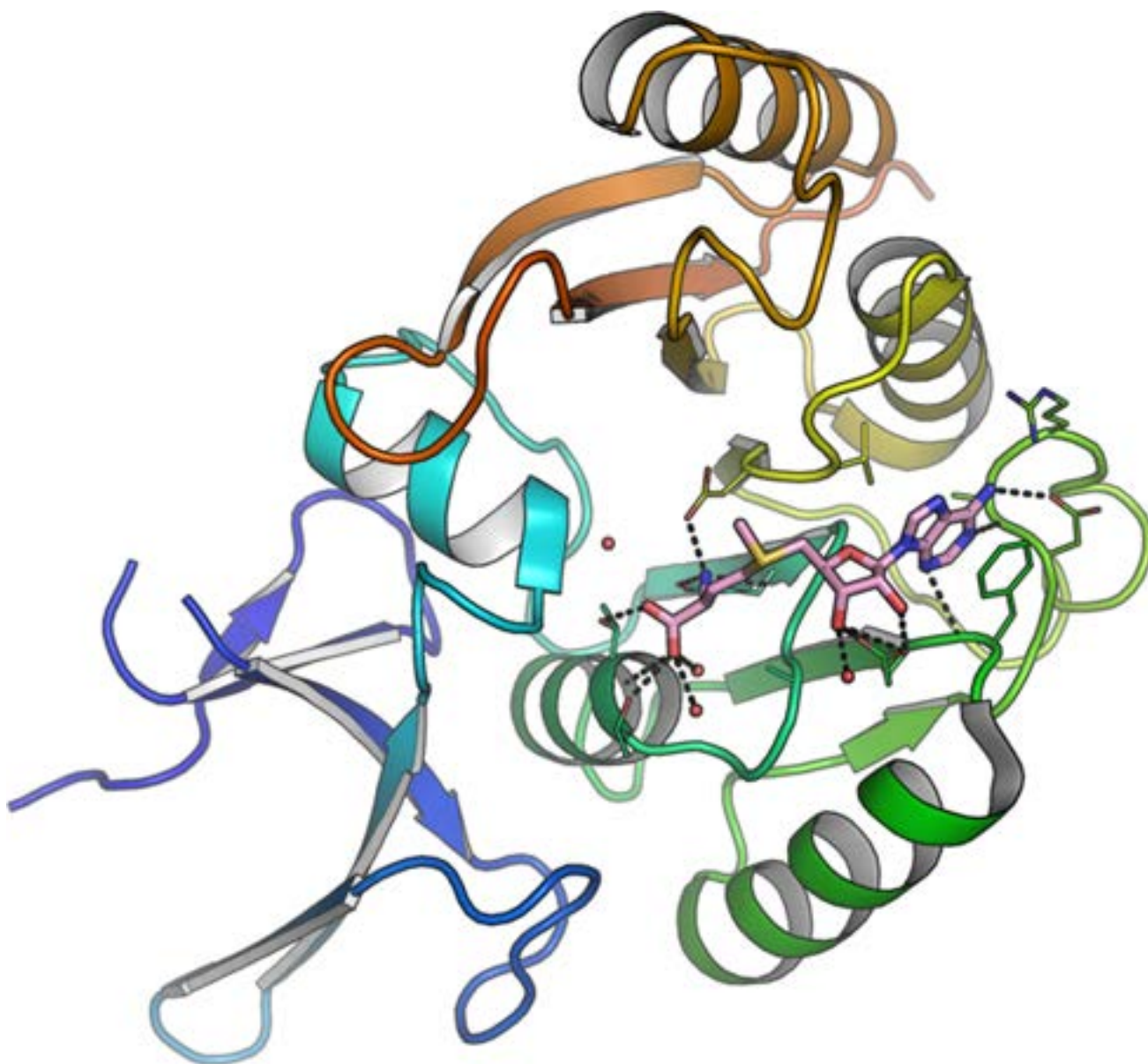


Figure 4. A 2.0 Å structure of fibrillar protein with the cofactor SAM (pink) bound in the active site determined at the MX2 beamline at the Australian Synchrotron.

The work of Associate Professor Waihong Tham's group at The Walter and Eliza Hall Institute (WEHI) aims to disrupt viral entry into host cells using nanobodies. Nanobodies are camelid-derived single-domain antibodies and represent the smallest naturally derived antigen-binding fragment (approximately 15 kDa in size). Their small size, along with high antigen-binding affinity, solubility, increased stability and ease of recombinant production (compared with traditional antibodies), make them attractive as potential therapeutics. These properties also allow the potential for the development of inhaled bio-therapies.

Entry of the SARS-CoV-2 virus is mediated by the interaction between the virus spike protein and its human receptor, angiotensin-converting enzyme 2 (ACE2). The high-affinity interaction with ACE2 is mediated by the receptor-binding domain (RBD) of the spike protein. To generate nanobodies, alpacas are immunized with virus spike and RBD proteins: the nanobody repertoire from isolated plasma cells is cloned into high-diversity phage display libraries, which are then used to select for antigen-specific nanobodies. The WEHI team will screen for nanobodies that inhibit the interaction of virus spike proteins with human ACE2, hence stopping virus entry and preventing SARS-CoV-2 infection.

Philip Pym and Melanie Dietrich are using the MX2 beamline of the Australian Synchrotron to determine the structure of the lead inhibitory nanobodies in complex with the RBD of SARS-CoV-2 to define key

immune epitopes and to understand the structural mechanisms of inhibition. These structures will provide invaluable information that will allow further development of antibody therapies against SARS-CoV-2.

## Conclusion

The ANSTO/Australian Synchrotron is committed to delivering research efforts into the SARS-CoV-2 virus, with our teams helping to address the impacts of COVID-19 via our national and landmark research platforms. One of the beamlines actively involved in SARS-CoV-2 and COVID-19 research, the Microfocus Crystallography beamline (MX2), has been used by several research teams to determine the structures of several proteins from the SARS-CoV-2 virus and/or related to our body's response to COVID-19. The members of the scientific team employed by the ANSTO/Australian Synchrotron are highly experienced scientists with a diverse set of interests (from hands-on crystallography through to improving experimental techniques/outcomes using beamline equipment modification/improvement). The team consists of eight scientists who have rapidly responded to the immediate needs of our scientific community, as well as understanding the importance positive media has to play, informing the general public of the work being undertaken both within Australia and the world in general. All team members are proud to be playing our vital part in one of the greatest public health and societal challenges of our time.

Access requests for researchers undertaking SARS-CoV-2/COVID-19 can be submitted via the special COVID-19 access round (<https://portal.synchrotron.org.au>), selecting MX1: Macromolecular Crystallography [12] as the equipment set ID and then Special Rapid Access COVID-19 (for the round).

For commercial (paid) access to undertake research at the Australian Synchrotron please contact Dr David Cookson ([cooksond@ansto.gov.au](mailto:cooksond@ansto.gov.au)).

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## RESEARCH NEWS

# Supporting COVID-19 research at the NSLS-II

Jean Jakoncic, Alex Soares, Dale Kreitler, Dean Hidas, Lin Yang, Desigan Kumaran, Martin R. Fuchs, Sean McSweeney, Wuxian Shi

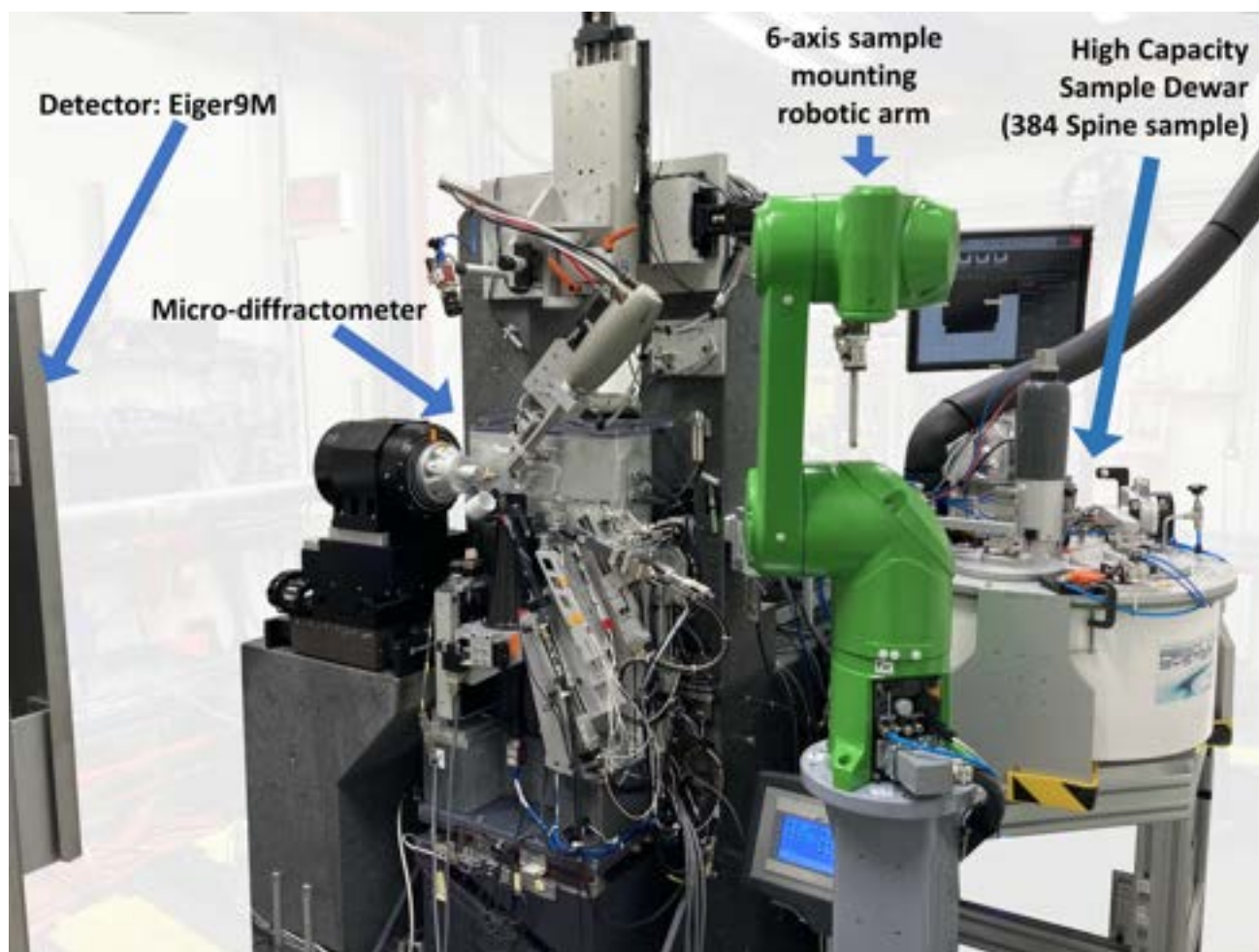


Figure 1. Endstation of one of the MX beamlines (AMX, 17-ID-1).

The National Synchrotron Light Source II (NSLS-II) at Brookhaven National Laboratory (BNL), Upton, NY, is the latest and brightest synchrotron facility in operation in the US. As of May 2020, it remains in a special user-operation mode to support exclusively COVID-19 research. All DOE BES Synchrotron facilities have re-scheduled their planned maintenance shutdowns to provide continuous access to X-ray beams for life sciences experiments in the US. As a result, the NSLS-II will remain in extended operation until mid-June 2020. The NSLS-II supports research from 29 operating beamlines and, under the current special circumstances, maintains normal operation for three dedicated beamlines.

Two macromolecular crystallography (MX) beamlines and one dedicated life-science X-ray scattering beamline accept Rapid Access proposals for COVID-19 related experiments. To maintain continuous safe operation, all experiments are performed remotely with minimum staff at the beamline to provide support.

The highly Automated and Frontier MX beamlines (AMX and FMX, respectively) (Fuchs *et al.*, 2016) offer remote access and automated data collection, providing state-of-the-art capabilities in terms of X-ray beam (size and flux) and rapid screening for potential crystals as small as 1  $\mu\text{m}$  (Fig. 1). Most of the efforts at the MX beamlines are either to support or implement research intended to determine the structures of proteins from the Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2). The major targets have been the spike protein and the main protease, which happen to be studied by most groups worldwide.

Operating the three beamlines safely and at full performance, with minimal staff observing social distancing, and without the possibility for users to come on site requires several changes. All users conduct their experiments remotely through a *NoMachine* session. Users that access the beamlines for the first time have dedicated training sessions one week before their actual beam time, to ensure their connection performance and help them make full use of the beamlines' capabilities and beam specifications. During a user experiment, only one staff member is present at the beamline, with engineering and technical support on call if needed. In daily walkthroughs and regular test experiments, we confirm that all systems are running optimally and establish the beamline performance. Newly developed monitoring systems and remotely accessible websites developed by the accelerator division of the NSLS-II greatly facilitate supporting and observing systems from outside the laboratory.

In order to further accelerate early discoveries of potential diffracting conditions from either microcrystals or poorly diffracting samples that are typically produced in the very early stages of drug-discovery projects, we have adapted one of our existing data collection protocols to deliver improved results to highlight possible hits. In short, the pipeline evaluates the fast raster data in an initial spot-finder run and attempts to index frames with a number of reflections above a threshold. Successfully indexed frames with the smallest cell dimensions above a threshold are highlighted. Planned updates include further tuning and graphical presentation of the scan results.

NSLS-II started minimum safe operation mode, where only the two MX beamlines were in operation in March of 2020 and soon authorized the SAXS beamline to also enter operation under the same conditions (COVID-19-related work). So far we have supported 20 experimental visits from several proprietary groups and started in-house research as well.

## In-house/collaboration research

### Spike protein: membrane fusion

Structure-guided development of viral fusion inhibitors: after the S protein receptor-binding domain (RBD) has bound to the angiotensin-converting enzyme 2 (ACE-2) receptor, the heptad repeat 1 (HR-1) and 2 (HR-2) domains interact with each other to form a six-helix bundle (Tang *et al.*, 2020: 6HB; Fig. 2). This conformational rearrangement brings the viral envelope into proximity with the host-cell membrane and facilitates membrane fusion. This type I fusion mechanism, employed by other viruses such as HIV, Ebola and RSV, presents a viable target for the treatment or prevention of infection by SARS-CoV-2 (Tang *et al.*, 2020). Strategies that aim to exploit this type of fusion mechanism mainly involve the development of synthetic variants of the S protein HR-2 domain (or equivalent), which bind to the HR-1 domain (or equivalent) trimer and disrupt the formation of the 6HB (Tang *et al.*, 2020). These fusion inhibitory HR-2 domain variants are typically large polypeptides in excess of 3 kD which form an extensive interface with a protein surface; therefore, structural information plays an integral role in expediting the development process.

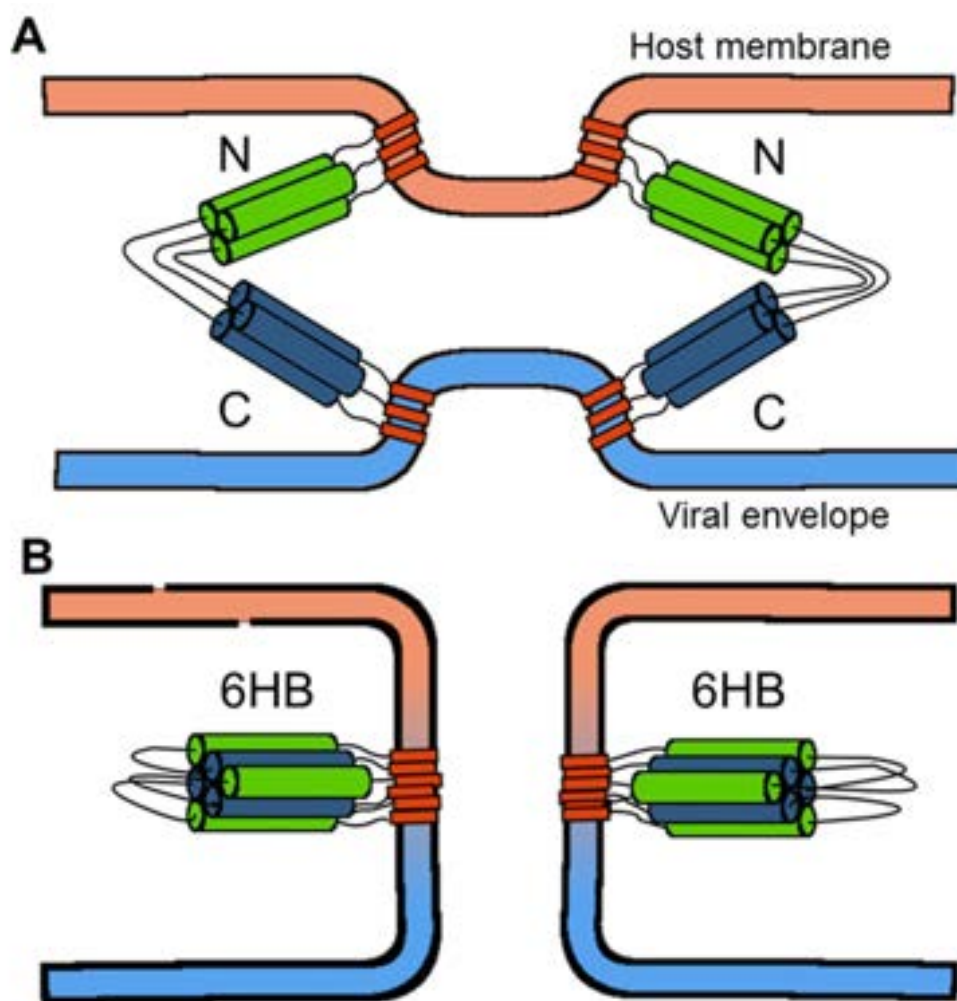


Figure 2. (a) The prefusion hairpin intermediate in which the N-terminal HR-1 domain has inserted into the host cell membrane. (b) Formation of the 6-helix bundle (6HB) drives membrane fusion between the host cell membrane and the viral envelope.

Recently, Lu *et al.* published the structure of the SARS-CoV-2 6HB (PDB entry 6LXT) in addition to describing a fusion inhibitor peptide with broad-spectrum activity against multiple human coronaviruses, laying the groundwork for the development of SARS-CoV-2 fusion inhibitors derived from the S protein HR-2 domain (Lu *et al.*, 2020) (Fig. 3). Currently, work is under way at NSLS-II to provide structural support in a collaborative effort with molecular dynamics (MD) and chemists at academic institutions to generate peptide fusion inhibitors with improved inhibitory activity and/or pharmacokinetic properties. The in-house crystallization laboratory, adjacent to the MX beamlines, has allowed for the immediate screening of potential lead conditions, and the microbeams and raster processing pipelines have been essential for the rapid identification and indexing of microcrystals from initial sparse matrix hits.

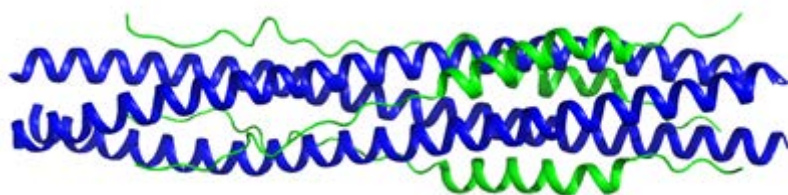


Figure 3. Structure of the SARS-CoV-2 6-helix bundle (6HB) in the post-fusion conformation (PDB entry 6LXT, Lu *et al.*, 2020).

## Spike protein receptor-binding domain

The SARS-CoV-2 spike protein induces cellular infection *via* a multi-step process, with different steps carried out by different domains. The spike protein is the only extracellular COVID-19 therapeutic target. The spike protein RBD is responsible for initial recognition between the spike protein and the human ACE-2. This makes RBD an attractive target for antiviral drug development. The RBD domain has been biochemically characterized, and crystal structures are available for RBD bound to monoclonal antibodies (PDB entries 6YLA, 6W41) and bound to ACE-2 (PDB entries 6LZG, 6M17). Low-resolution cryo-EM structures are available for the full-length spike protein trimer in the closed conformation (PDB entry 6VXX), in the open conformation (PDB entry 6VYB) and in a mixed state (PDB entry 6VSB). At the time of writing, there is no crystallographic structure known of the RBD trimer.

To effectively use fragment screening as a search tool for potential COVID-19 antivirals, we are searching for crystallization conditions for RBD unbound to antibodies or ACE-2. We are attempting crystallization trials for three RBD constructs. We have begun crystallization trials for the full-length 239 amino acid RBD domain. Additional crystallization trials are ongoing for a reduced-length 193aa construct with an N-terminal truncation, and for a 131aa construct with both N- and C-terminal truncations (the shortened constructs were designed to improve crystallization characteristics such as stability and solubility). Once reliable crystallization conditions are established, we will use structure-based fragment screening as a technique to identify potential antivirals which target SARS-CoV-2 RBD.

## Main protease

The SARS-CoV-2 genome consists of ~30,000 nucleotides, and its replicase gene encodes two overlapping polyproteins, pp1a and pp1ab, which are required for viral replication and transcription (Zhou *et al.*, 2020; Wu *et al.*, 2020). These two polyproteins undergo extensive proteolytic processing largely by the main protease ( $M^{\text{pro}}$ ), also referred to as the 3-chymotrypsin-like cysteine protease ( $3CL^{\text{pro}}$ ).  $M^{\text{pro}}$  has a Cys-His catalytic dyad at the active site and cleaves 11 sites in the p1 position of pp1a and pp1ab (Shimamoto *et al.*, 2015). This cleavage process produces a mature protein that anchors the replication/transcription complex and releases the mature non-structural proteins (NSPs) (Hsu *et al.*, 2005; Barretto *et al.*, 2005). Inhibition of  $M^{\text{pro}}$  prevents the production of mature proteins required for further replication, which makes it an attractive drug target.

The structure of  $M^{\text{pro}}$  with its mechanism-based inhibitor has been reported (Jin *et al.*, 2020), and this study gave us a starting point to develop more potent inhibitors against the  $M^{\text{pro}}$  activity. Using a structure-based drug design (SBDD) approach, we screened the FDA-approved drug database and selected potential antiviral and pathogen-specific hits. We are screening the selected drugs for inhibition, and the positive candidates will be subjected to co-crystal structure determination. In addition, we have determined the higher-resolution crystal structure of  $M^{\text{pro}}$  in a native conformation (Fig. 4). In the crystal structure, the active site of  $M^{\text{pro}}$  is open to the solvent region, and it is in a favorable form for the soaking and co-crystallization experiments. Our goal is to derive the structure-activity relationship (SAR) to identify drug modifications that improve potency and efficacy through multiple rounds of design, synthesis and *in vitro/in vivo* testing.

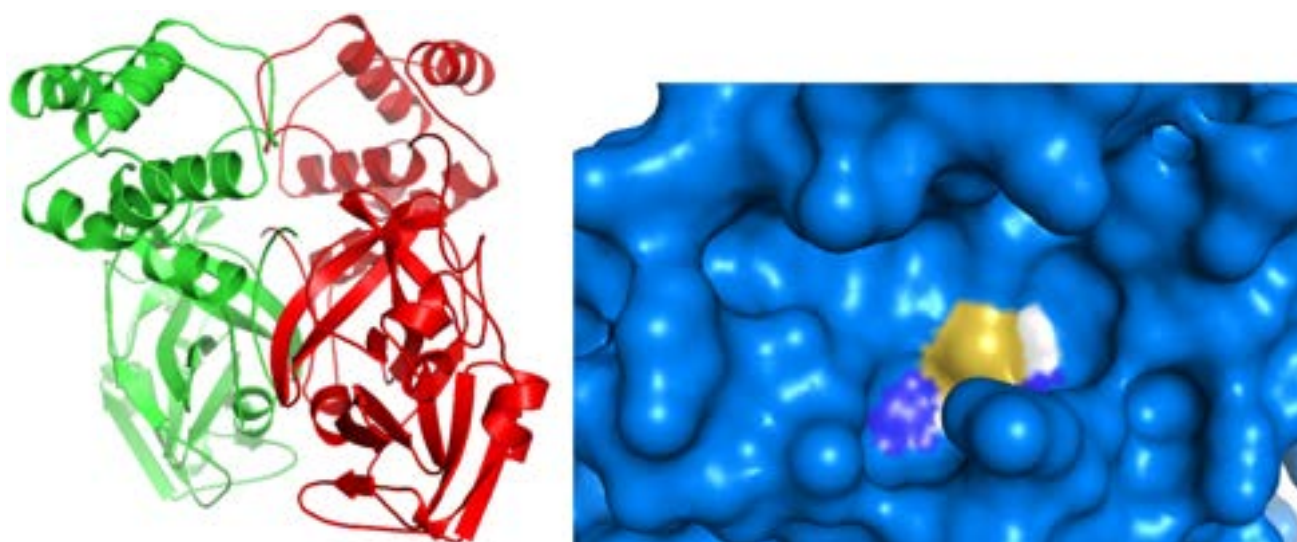


Figure 4. Crystal structure of M<sup>pro</sup> in the native conformation. (Left) Ribbon representation of M<sup>pro</sup> homodimer. (Right) Surface representation of M<sup>pro</sup> at the active-site pocket. The catalytic Cys145 and His163 are shown in yellow and dark-blue patches, respectively.

## Research carried out by pharmaceutical users

NSLS-II has hosted several companies that are researching treatments for COVID-19, with more companies planning to use the beamlines as early as possible. These companies are requesting repeated access. New protocols have been established to dedicate all resources to COVID-19-related research, to fast track approval of new COVID-19 projects and to ensure that data can be safely obtained from COVID-19-related samples. The approved projects tested a wide variety of strategies such as antiviral drugs that interfere with the viral infection and drugs that boost the immune system before infection and heal injuries post-infection. To date, more than 1000 samples have been screened, 40% of them have provided useful data (processed files).

The metadata from proprietary users indicate a significant change in the usage pattern during the COVID-19 crisis. For example, companies are bringing more samples related to the early stages of research, and fewer samples from mature projects (preliminary research has a high proportion of samples rejected during diffraction screening). Proprietary users are applying for beam time to test microcrystals in the size range 2–5  $\mu\text{m}$  from difficult-to-crystallize targets or from initial crystallization screenings. This change in usage pattern confirms that our new protocols for expedited approval are encouraging proprietary users to take advantage of the unique micro-focusing and automation capabilities of NSLS-II at the earliest possible stage in their drug development work flow.

## Research carried out at the bioSAXS beamline

There is a demand for solution scattering at the LiX beamline to support COVID-19 research as well. Solution scattering provides low-resolution structural information when the molecule or molecular complex cannot be crystallized or needs to be studied under near-physiological conditions. So far, the LiX beamline has scheduled two user experiments through COVID-19 Rapid Access proposals. More experiments, including those in support of the in-house research described above, are under discussion. In addition to the capabilities for high-throughput measurements and in-line size-exclusion chromatography, the ability to collect data up to wide scattering angles ( $q \sim 3.2 \text{ \AA}^{-1}$ ) (Yang *et al.*, 2020) is of particular interest to these users, to detect molecular binding (Fischetti *et al.*, 2004).

## Research carried out using computer modeling on the NSLS-II/CBMS cluster

Additionally, CBMS and NSLS-II computing resources, when not in use for ongoing data collection at the two MX beamlines, are made available using the SLURM *Workload Manager* system to other groups from NSLS-II. These roughly 700 cores are currently part of a large collaborative project to target the entire proteome of SARS-CoV-2 to identify small molecules with positive binding.

Using physics-based and artificial intelligence (AI)-driven virtual screening models, and AI-enhanced MD to further assess binding, the most promising molecule and SARS-CoV-2 protein complexes will be investigated experimentally, including structural biology studies. Nodes are used every night, every weekend and every day that the beamline is not operating.

We particularly thank the NSLS-II accelerator division staff members who maintain the continuous operation of the storage ring to support user and staff research and develop new monitoring systems and control systems for remote operation. We also thank the ICT group from NSLS-II who continuously maintain stable operation of the file system and our small dedicated cluster.

We appreciate that in times like these, operating the light source safely and supporting our work is a difficult undertaking, and we are very thankful for the continued support by the technical, engineering and operations groups.

We thank the Diamond Light Source for sending the plasmid for the main protease and for sharing protocols for expression, purification and crystallization.

Work on the M<sup>pro</sup> is a collaboration with John Shanklin from the Biology Department at BNL, NY. The computing work is led by Hubertus Van Dam at the Computing Science Initiative, BNL, NY.

The work on the spike protein membrane fusion is a collaboration with Victor Outlaw and Sam Gellman from the University of Wisconsin-Madison and with Matteo Porotto and Anne Moscona from Columbia University.

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## RESEARCH NEWS

# SSRL joins global fight against COVID-19

Michael SoltisAina E. Cohen

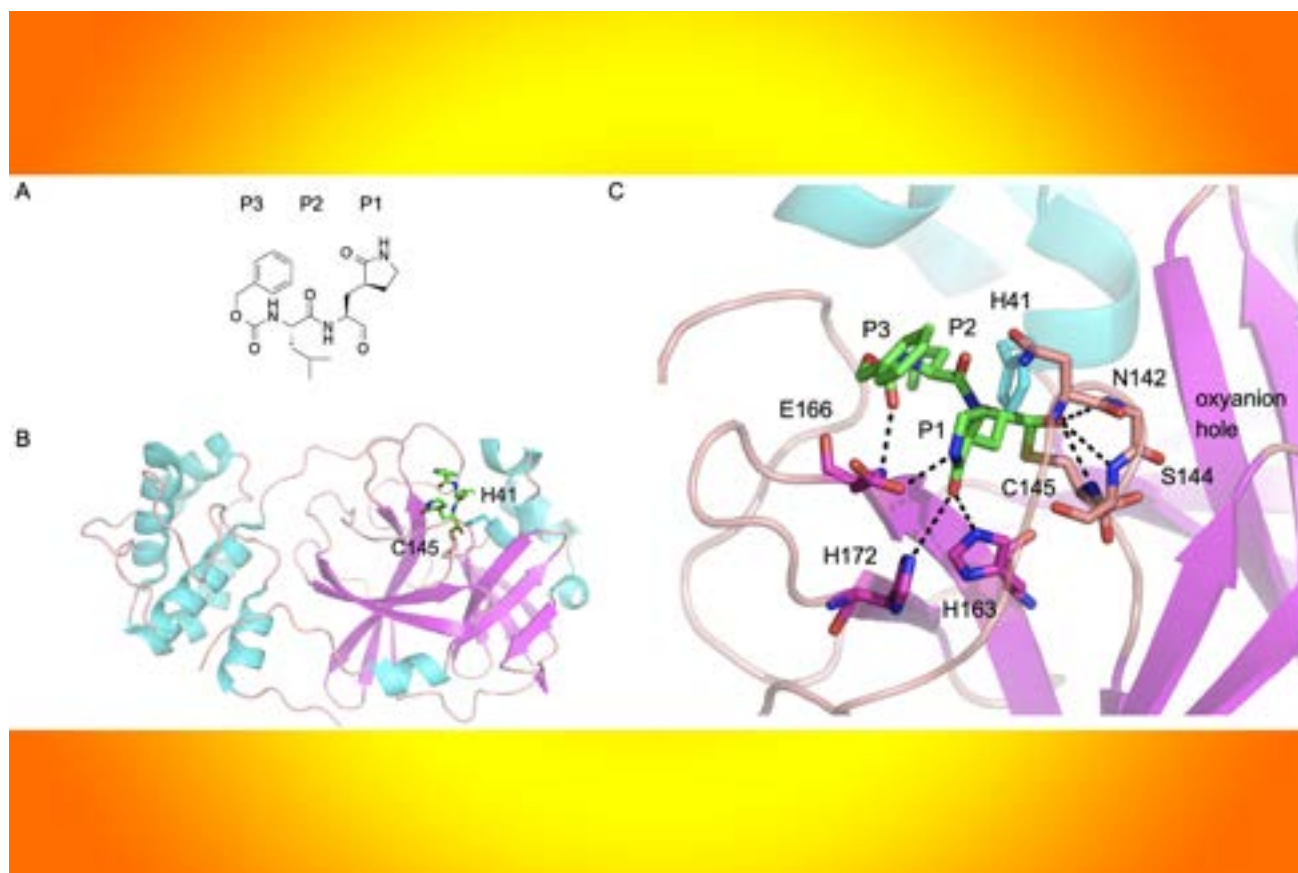


Figure 1. GC373 binds in the active-site pocket of SARS-CoV-2 M<sup>pro</sup> (3CL<sup>pro</sup>). A) The feline drug GC373 is a peptidyl inhibitor; the P1, P2 and P3 residues are indicated. B), C) GC373 forms a covalent bond with the catalytic residue Cys145. The oxyanion is stabilized by backbone hydrogen bonds with G143, S144 and C145. Hydrogen bonds are established with the P1 and P3 residues of GC373 and side chains of H163, E166 and H172 as well as the backbone of residue E166. SARS-CoV-2 M<sup>pro</sup> is represented in cartoon form with the inhibitor in green. PDB entry 6WTJ (Khan, M. B., Arutyunova, E., Young, H. S. & Lemieux, M. J., to be published).

As part of the international response to the COVID-19 pandemic, the Stanford Synchrotron Radiation Lightsource (SSRL) at the U.S. Department of Energy's (DOE) SLAC National Accelerator Laboratory has re-opened some of its beamline facilities. They will be used to obtain fundamental information on the atomic structure of macromolecules related to the SARS-CoV-2 virus and how it interacts with potential drug treatments and vaccines.

While the March 16 San Francisco Bay Area's shelter-in-place order temporarily shut down all but essential operations at SLAC, SSRL is operating a subset of its Structural Molecular Biology resource synchrotron experimental stations. This includes three macromolecular crystallography (MC) beamlines, a small-angle X-ray scattering (SAXS) beamline and several cryo-EM instruments to perform COVID-19-related research.

Researchers from academic institutions in the USA and Canada have been using the MC X-ray beamlines to study the atomic structures of multiple SARS-CoV-2 targets. They include groups from Stanford

University, University of California in San Francisco, The Scripps Research Institute, Thomas Jefferson University and the University of Alberta.

This work includes antibody complexes of the spike proteins that reside on the surface of SARS-CoV-2. Spike proteins dock onto receptor molecules on the surface of cells in the lungs, heart or intestines and allow entry of the virus into the host cells where the virus is replicated.

Several other research projects are employing structure-based approaches to design antiviral drugs targeting RNA-dependent RNA polymerase and drugs that treat the disease by modulating or blocking host proteases that are central to virus replication.

Just recently, the Lemieux group (University of Alberta) solved several structures of coronavirus drugs bound at the active-site pocket of the main protease of SARS-CoV-2 (Fig. 1), effectively blocking virus replication [1]. This may be a strong drug candidate for the treatment of human coronavirus infections because it has already been successful in treating SARS-CoV in animals.

The MC facilities are fully operated by the external investigators in remote-access mode, while supported by a minimal number of on-site SSRL staff, in accordance with physical distancing rules. SSRL was a pioneer in the field of automation, robotics and remote access [2–5], and is in an ideal position to operate under the conditions of a pandemic. The Stanford Auto-Mounter (SAM) robot mounts samples, stored in liquid nitrogen, into the experimental station (Fig. 2). The MC beamlines are typically used *via* remote access (about 95%), and the MC-developed control software and GUI, *Blu-Ice*, allows for real-time experimental control and collaboration all over the world [5]. See <https://www6.slac.stanford.edu/news/2020-04-16-slac-joins-global-fight-against-COVID-19.aspx>.

In the video shown above, a robot mounts samples, stored in liquid nitrogen, into an experimental station for X-ray crystallography at SLAC's synchrotron, SSRL (Aina Cohen, Silvia Russi, SLAC National Accelerator Laboratory).

"We're able to do this research and, at the same time, follow strict physical distancing protocols because our experimental stations are highly automated and require only minimal staff on site," said Aina Cohen, who co-leads SSRL's Structural Molecular Biology Division. "Researchers can send us frozen samples to be mounted by robots into the experimental stations. The remote researchers then use our software and video tools to control the robots, view their samples and control all the details of their experiment from their home laboratory, or even from their homes."

Studies of a conserved coronavirus enzyme, at physiological temperatures associated with infection in humans, were also conducted by the Fraser group (UCSF). Understanding how these changes affect the structure and interactions of this enzyme, which has been shown to promote virulence in the coronavirus family, may provide key insight leading to the development of antiviral therapeutics.

Cohen added, "We are excited to be soon expanding our remote program to offer the ability to safely ship and collect data at ambient temperature. This will significantly improve the efficiency of these types of experiments and the quality of the CoV-related structures determined at physiologically relevant temperatures."

Another method used at SSRL for COVID-19-related research is SAXS. This method is used to probe macromolecules and, in particular, macromolecular assemblies and complexes in a solution environment. It not only determines the general shape and solution structure of the molecule or molecular complex or assembly but also can be applied to investigate interaction potentials and structural changes due to ligand binding.

Similar to the MC facilities at SSRL, the instrument and some of the major sample environments at the SAXS beamline 4-2 are highly automated [6] and can be operated remotely *via* a customized version of the *Blu-Ice* control system. Thus, the most frequently performed experiments at the beamline can be carried out by users at their home institutions with only minimal help from on-site staff members.

In one of the current projects, Thomas Weiss, who oversees the research at this experimental station (Fig. 2), is collaborating with Gerard Wong's laboratory and other scientists at the University of California, Los Angeles, as well as researchers at the University of California, San Diego, to look at the interactions of selected pieces of the viral proteome that interact with the machinery of the human immune system. Insights into these interactions could help scientists understand why SARS-CoV-2 is so infectious and causes so much inflammation – a principal reason for why it is so deadly.



Figure 2. SAXS experimental station at SSRL used for COVID-19 research (Ivan Rajkovic, SSRL).

Researchers at two National Institutes of Health (NIH)-funded centres – the Stanford-SLAC Cryo-EM Center and the National Center for Macromolecular Imaging – are pursuing answers to very similar questions with cryo-EM, which uses electron beams to study the novel coronavirus and how its molecular components interact with drugs or antibodies. One advantage of this technique is that it produces atomic resolution images of biomolecules in their frozen hydrated state, with no need for crystallization.

“One of the projects in full swing is a collaboration with Professor Rhiju Das at the Stanford Medical School that is aimed at designing a drug to keep viral RNA from carrying out its normal function,” said Wah Chiu, director of the NIH centres. This work involves developing synthetic RNA molecules that mimic stretches of the viral RNA, determining their 3D structures with cryo-EM and using that information to build molecules that can bind and deactivate the RNA.

In addition, Chiu and his colleagues have teamed up with several groups around the world that are preparing virus-like particles, viral proteins and RNA-protein complexes for cryo-EM analysis, as well as virus-infected cells for tomographic studies of what exactly the virus does once it enters host cells.

SLAC is also planning to bring experimental stations at its Linac Coherent Light Source (LCLS) back online for COVID-19-related research. One of the brightest X-ray sources on Earth, this X-ray free-electron laser will complement other techniques – looking, for example, at hard-to-crystallize viral particles at room temperature to see how the virus attacks host cells and interacts with potential drugs under conditions that are close to those in the human body.

*The SSRL Structural Molecular Biology Program is supported by the DOE Office of Biological and Environmental Research, and by the NIH and the National Institute of General Medical Sciences (including P41GM103393).*

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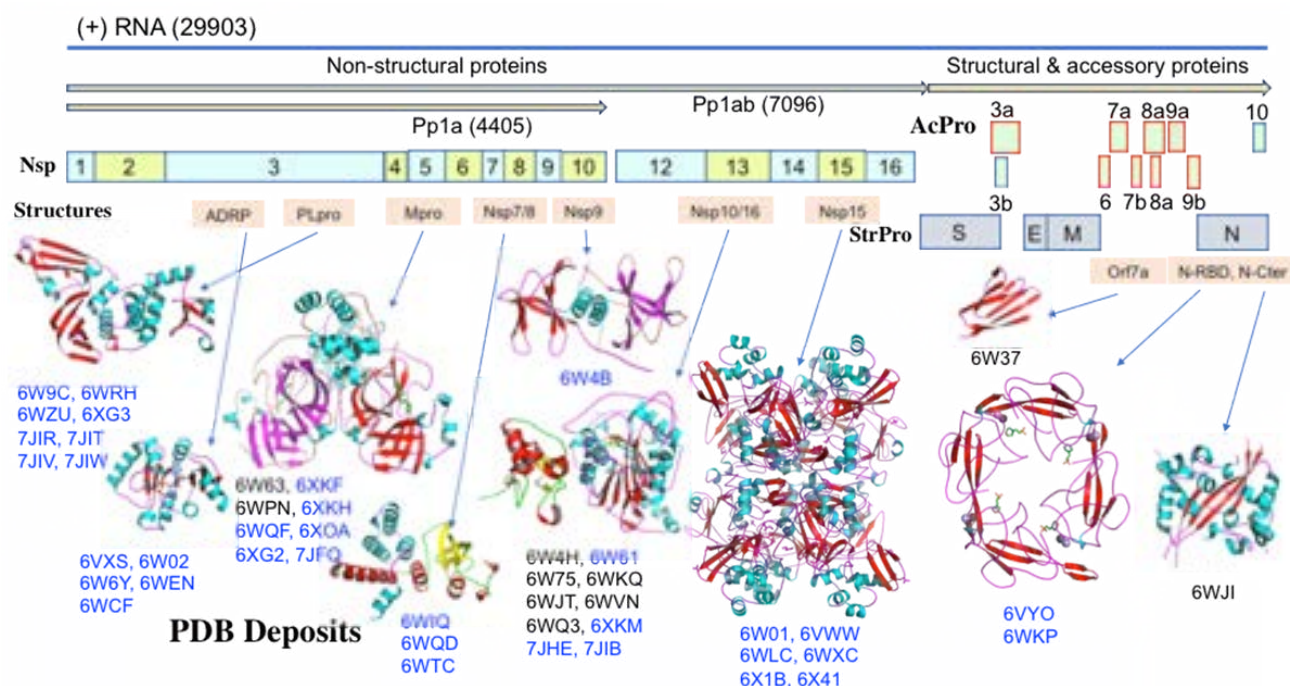
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## RESEARCH NEWS

# Structural biology of SARS-CoV-2 proteins

Andrzej Joachimiak Karolina Michalska



SARS-CoV-2 genome coverage by CSGID structures.

## Background

At the end of 2019 in the Wuhan province of China, a new virus emerged and spread rapidly, infecting thousands. The Severe Acute Respiratory Syndrome coronavirus 2 (SARS-CoV-2) responsible for Coronavirus Disease 2019 (COVID-19) is rapidly propagating around the world. The virus spreads very efficiently and sustainably between people in close contact, and it is now found in over 180 countries and on all continents, and there seems to be no stop to its dispersal with new centers emerging in different parts the world. There is no existing vaccine or proven drug treatment to prevent infections and stop virus proliferation. Detailed information about SARS-CoV-2 proteins, their functions and structures, is urgently needed to support the development of effective measures. A number of research groups around the world have initiated studies to determine structures of key proteins from the new virus and bring them into the public domain. The advances have been much faster than in the past, and they can lead the way to new treatments.

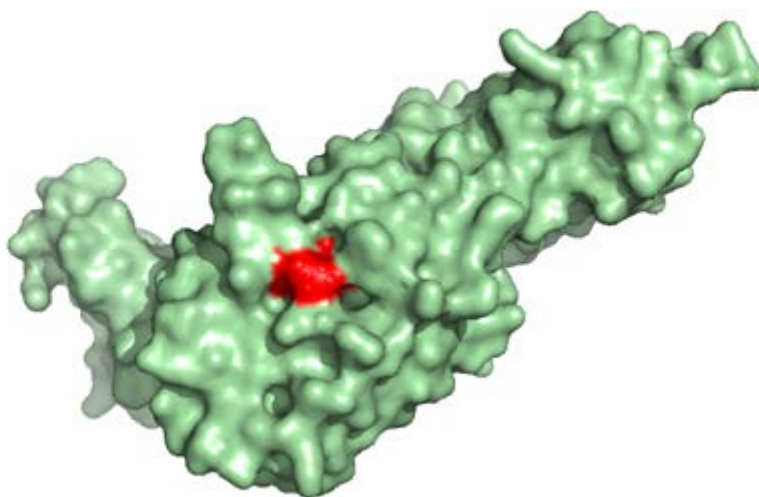
For over 100 years, since the Spanish flu pandemics, humanity has not experienced a major, global infectious disease. But a lot has changed since then. The human population grows and infringes on the environment, and people travel globally; the CDC estimates every year approximately one billion people cross borders, and pathogens move with them. In the 21st century, the world has experienced a number of viral outbreaks that threatened large human populations. These include Severe Acute Respiratory Syndrome [SARS 2002–2003 (Luk *et al.*, 2019)], Middle East Respiratory Syndrome [MERS 2015 (Memish *et al.*, 2020)], Ebola Virus Disease [EVD 2013–2014 (Cenciarelli *et al.*, 2015)] and Zika Virus Disease [ZVD 2016 (Nugent *et al.*, 2017)] to mention a recent few. Only for some of these diseases do effective antibodies and dedicated small-molecule drugs exist. However, these outbreaks were contained well before they spread worldwide. This has not been the case with COVID-19.

## SARS-CoV-2

The emergence of SARS-CoV-2, an etiologic agent responsible for the current outbreak of COVID-19, is different. At the end of 2019, there were increased numbers of pneumonia reports in central China which rapidly spread to other provinces. The agent associated with COVID-19 was quickly identified as another coronavirus, termed SARS-CoV-2, likely of zoonotic origin. In the five months since it was discovered, the disease has dispersed uncontrollably, often through large superspreading events, causing a pandemic. Governments and health organizations put in place various strategies and countermeasures that work with varying degrees of success. The challenges are enormous, millions of people are being quarantined and the epidemic is impacting the world economy. It is clear that the current approach to develop effective drugs for emerging diseases is not working very well and we need a long-term strategy in place to deliver several treatment options against viruses and other pathogens.

Coronaviruses have been known for a long time; the first coronavirus of human origin, B814, was described in 1966 (Tyrrell & Bynoe, 1966). These viruses often have zoonotic origin and have been isolated from civets, camels, alpacas, rodents and pigs, with bats being recognized as a major reservoir species. The viruses can jump into humans and cause a variety of diseases with a range of outcomes, from a mild cold to pneumonia to deadly SARS. Such animal-to-people followed by human-to-human transmissions led to the SARS and MERS outbreaks, but they were well controlled at the time.

Similar to other coronaviruses, the genomic information of SARS-CoV-2 is stored in a single (+)sense RNA molecule. The genome codes for several Orfs encoding 15 non-structural (Nsps), 4 structural and 9–10 accessory proteins. The majority of the genome is dedicated to the replicase gene encompassed within large, slightly overlapping Orf1a and Orf1b. These Orfs are translated into two polyproteins, pp1a (4405 amino acids) and pp1ab (7096 amino acids) (Cui *et al.*, 2019). The production of pp1ab requires a –1 ribosomal frameshift upstream of the Orf1a termination codon to extend translation of the pp1a. The resulting polypeptides are cleaved by two viral proteases to generate Nsps (Báez-Santos *et al.*, 2015). The main protease M<sup>pro</sup>, also referred to as 3C-like protease (3CL<sup>pro</sup>), has a chymotrypsin-like fold and is encoded by Nsp5. The other proteolytic enzyme is a papain-like protease (PL<sup>pro</sup>) and is encoded within Nsp3. Nsp3 is the largest protein produced by coronaviruses and it has several domains. In addition to PL<sup>pro</sup>, it contains an ubiquitin-like domain (Ubl1), acidic HVR domain, ADP-ribose-1-phosphatase (ADRP, also called Mac1, MacroD) and transmembrane region (TM). Viral Nsps, together with RNA, form a large membrane-bound assembly known as a replication and transcription complex. Although the functions of many Nsps have been directly associated with viral replication, the roles of some proteins, even with known enzymatic activities, remain poorly characterized. Understanding the structure, function and interactions of the RNA-synthesizing machinery of coronaviruses with host macromolecules is the key to rationalizing the development of improved control strategies.



Structure of SARS-CoV-2 PL<sup>pro</sup> protease determined at 1.8 Å resolution. The active site residues C111 and H272 are shown in red.

The worldwide spread of SARS-CoV-2 has raised questions as to how the virus achieved such high transmission rates. Unfortunately, despite many years of research, there is no approved vaccine, antibody or antiviral therapy for treatment of any of the SARS and MERS viruses.

In January 2020, the genome of SARS-CoV-2 became available to researchers around the world. The reaction of the scientific community was instantaneous. Within a few days, many molecular and structural biology laboratories redirected their efforts towards a single goal: producing proteins from SARS-CoV-2, characterizing their functions and solving protein structures to aid structure-based drug discovery. At the molecular level, the SARS-CoV-2 proteins are generally similar to their equivalents from the closely related SARS-CoV, and less like MERS-CoV proteins. However, the level of similarity varies significantly between proteins in SARS-CoV-2–SARS-CoV comparisons from 46% to 100% identity. These variances must be, at least in part, responsible for different host–pathogen interactions and epidemiological outcomes for these two closely related viruses (Wu *et al.*, 2020). During the SARS 2003–2004 outbreak, major international effort was launched which was coordinated by the *International Research Network on Structural Genomics of SARS-CoV* (Canard *et al.*, 2008). Within four years, 17 structures of SARS-CoV Nsps, structural and accessory proteins were determined in the US, Europe and China, covering 45% of the SARS-CoV proteome and 53% of its soluble proteins. These structures were determined by X-ray crystallography and NMR, and it became very clear that X-ray light sources made major contributions. This effort was highly relevant to attacking SARS-CoV-2, as it helped to decipher the SARS-CoV-2 ~30 kbs genome and design constructs for protein expression and structural studies. The Adam Godzik lab at UCR quickly created a [resource](#) that compares genome coverage for SARS and SARS-CoV-2. But how long would it take to generate the data urgently needed by the scientific community to find effective treatments against the global plague infecting millions by April 2020? It turned out that it did not take very long because of the past 15 years of technological advances. There were several important developments that have helped to speed up the science. Light sources, particularly the Advanced Photon Source (APS) in the US and the Diamond Light Source in the UK were very well set up for rapid-access single-crystal X-ray crystallography (Douangamath *et al.*, 2020). X-ray crystallography has made tremendous progress in the past 20 years. Synthetic gene technology with sequences codon-optimized for expression of soluble proteins in *E. coli* was a commercially available resource. Many plasmid-based expression systems were available for expression and co-expression of interactive proteins. Rapid purification and crystallization protocols were established (Kim *et al.*, 2011). The cryo-EM revolution provided a complementary technique to attack proteins and complexes that are difficult to crystallize (Walls *et al.*, 2020).

And the race began; it will not be a sprint, but rather a marathon because of the significant challenges. A massive therapeutic effort has been put together towards vaccine development and repurposing FDA-approved drugs, with somewhat limited success. The blood from virus-infected survivors is being used to treat the most severe cases with some success, and a number of potential treatments are in initial trials. However, it is expected that the virus and its global reservoir will evolve, making some treatments obsolete and leading to the emergence of new outbreaks.

## Research

Structural characterization of viral proteins is essential to develop vaccines, antibodies and small-molecule drugs. From the beginning, there were several important players that contributed to swift progress, and these contributors follow one of the most critical rules that all data are shared with the scientific community prior to publication, allowing science to accelerate. At the NIH/NIAID-funded Center for Structural Genomics of Infectious Diseases (CSGID), we redirected our efforts in January 2020 and focused on structural characterization of as many SARS-CoV-2 macromolecules as possible. The Center is a consortium of Northwestern University (Karla Satchell, CSGID co-director), the University of Chicago operating at Argonne National Laboratory (UChicago/ANL) (Andrzej Joachimiak), the School of Medicine at Washington University (Daved Fremont), the University of California Riverside School of Medicine

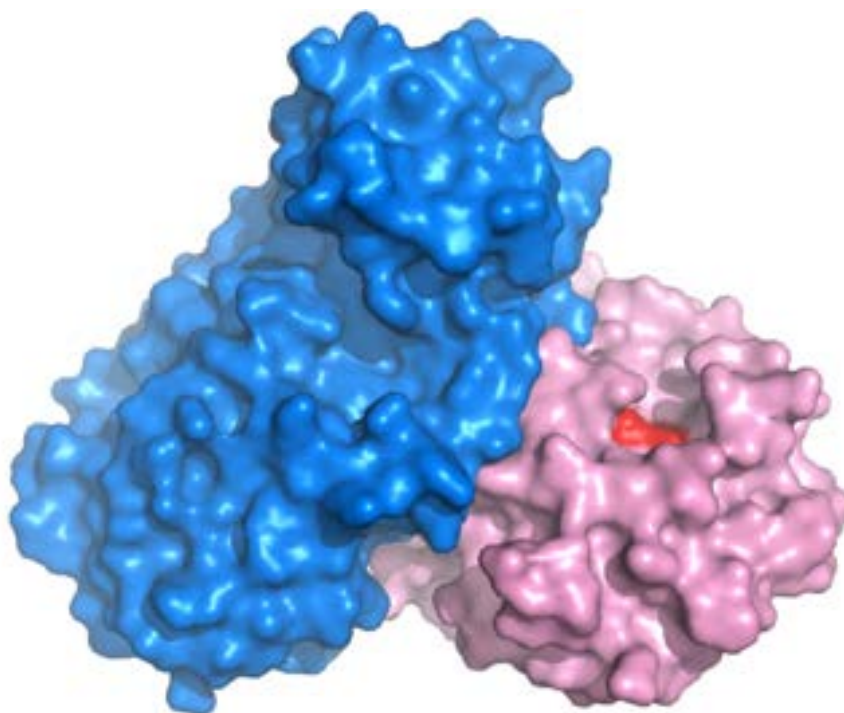
(Adam Godzik), Purdue University (Andy Mesecar and Richard Kuhn), the University of Virginia (Wladek Minor), UT Southwestern Medical Center at Dallas (Zbyszek Otwinowski and Dominika Borek) and the University of Calgary (Alexei Savchenko), all of which contributed to the current efforts. Some members of the consortium had previous experience with SARS-CoV proteins. Our high-throughput protein production and structure-determination pipeline have allowed us to produce multiple samples for crystallographic and cryo-EM work and to support other scientific endeavors. Most of these involve multiple collaborations aimed at enhancing the impact of SARS-CoV-2 research by addressing small-molecule screening, vaccine and antibody development, and basic molecular biology of the coronavirus.

The first structure reported for CoV-2 was the main protease ( $M^{\text{pro}}$ ) in complex with the inhibitor N3. The structure was determined by the Zihe Rao group in Beijing [Protein Data Bank (PDB) entry 6LU7, released on 6 February 2020] and later published in *Nature* (Jin *et al.*, 2020). By the middle of February, the CSGID was well advanced in the purification and crystallization of viral proteins, and at the UChicago/ANL CSGID site, we determined the second CoV-2 structure. The Nsp15 endoribonuclease was deposited on 20 February 2020 (PDB entry 6W01, released on 4 March 2020) and published in *Protein Science* (Kim *et al.*, 2020). In the following five months, the CSGID determined and made available to the scientific community 45 structures of 11 SARS-CoV-2 proteins (Table 1), including 35 determined in our laboratory at UChicago/ANL.

**Table 1. List of CSGID structures from SARS-CoV-2**

|                                                             |                                                |
|-------------------------------------------------------------|------------------------------------------------|
| Nsp3 Papain-like protease (PL <sup>pro</sup> )              | 6W9C, 6WRH, 6WZU, 6XG3, 7JIR, 7JIT, 7JIV, 7JIW |
| Nsp3 ADRP/MacroD                                            | 6VXS, 6W02, 6W6Y, 6WEN, 6WCF                   |
| Nsp5 Main protease (M <sup>pro</sup> , 3CL <sup>pro</sup> ) | 6W63, 6WPN, 6WQF, 6XG2, 6XKF, 6XKH, 6XOA, 7JFQ |
| Nsp7/Nsp8 Primase complex                                   | 6WIQ, 6WQD, 6WTC                               |
| Nsp9 RNA/DNA binding                                        | 6W4B                                           |
| Nsp10/Nsp16 2'-O-MT2'-O-ribose methyltransferase complex    | 6W4H, 6W61, 6W75, 6WKQ, 6WJT, 6WVN, 3WQ3       |
| Nsp15, EndoU, U-specific endoribonuclease                   | 6W01, 6VWW, 6WLC, 6WXC, 6X1B, 6X41             |
| Orf7a                                                       | 6W37, 6XKM, 7JHE, 7JIB                         |
| N-protein RNA binding domain                                | 6VYO, 6WKP                                     |
| N-protein C-terminal domain                                 | 6WJI                                           |

The important drug targets include high-resolution structures of PL<sup>pro</sup> and M<sup>pro</sup> proteases participating in the polyprotein processing. The three PL<sup>pro</sup> structures from the UChicago/ANL site correspond to the WT enzyme in two different crystal forms and the active-site cysteine mutant (C111S) determined at 1.6 Å resolution. The additional room-temperature (RT) structure of PL<sup>pro</sup> is in refinement. Two M<sup>pro</sup> structures from the Mesecar laboratory in Purdue include complexes with inhibitors (PDB entries 6W63, 6WPN). The third structure of M<sup>pro</sup> was determined at RT (PDB entry 6WQF) in collaboration with Andrey Kovalevsky's team at ORNL to advance computational modelling (the publication has been accepted by *Nature Communications*). Rolf Hilgenfeld at the University of Lübeck, Germany, who has studied SARS CoV proteins for years, provided important high-resolution structures of M<sup>pro</sup> complexes with potential inhibitors and developed the lead compound of the main protease (Zhang *et al.*, 2020).



Structure of SARS-CoV-2 main protease M<sup>pro</sup> dimer determined at room temperature. The active site residues H41 and C145 are shown in red.

At UChicago/ANL, structures of several other SARS-CoV-2 proteins have been determined as well, with an average resolution of 1.95 Å. Multiple structures were determined for Nsp15 endoribonuclease, assumed

to play a role in the evasion of host immunity. These include complexes with nucleotides and Tipiracil, an inhibitor from the list of FDA-approved drugs (publication in preparation). Five structures of Nsp3 domain ADRP/MacroD have been determined, including an *apo* form and in the presence of adenosine 5'-diphosphate ribose (ADPr) and AMP, as well as an atomic-resolution structure of the complex with MES buffer (PDB entry 6WCF) determined at 1.07 Å (Michalska *et al.*, 2020), providing detailed information about the binding site and interaction with the ligand. The function of this domain is to bind ADP-ribose or poly(ADP-ribose) and remove ADPr attached to human proteins or RNA in response to viral infection and thus may be involved in de-MARylation and de-PARylation (Li *et al.*, 2016; Ecker *et al.*, 2017; Munnur *et al.*, 2019). There are also three structures of the Nsp7/Nsp8 primase complex determined which show interactions and flexibility of Nsp8. The structure of Nsp9 shows a tight dimer (PDB entry 6W4B). Nsp9 from SARS and MERS was shown to interact with RNA and DNA. Nsp9 belongs to a smaller member of the replicase complex which is a crucial cofactor contributing to the emerging "Nsp interactome".

We have also determined structures of the N nucleocapsid protein and N-terminal RNA-binding domain (PDB entries 6VYO, 6WKP) at UChicago/ANL, and the C-terminal domain at Northwestern University laboratory (PDB entry 6WJI) (publication in preparation). The N nucleocapsid protein covers the entire virus genomic RNA in the virion but may also play other functions as it seems to interact with Nsp3. Comparison of the monomer with other homologs shows quite large conformational differences, suggesting that, despite high-sequence identity, this domain has built-in conformational flexibility required for binding RNA. Seven structures of Nsp10/Nsp16 methyltransferase complex have been determined at Northwestern University and UChicago/ANL. These structures describe the Nsp10/Nsp16 assembly crystallized under different conditions, as well as its complexes with S-adenosylmethionine and 7-methyl-GpppA dinucleotide (Rosas-Lemus *et al.*, 2020). We also determined one structure for an accessory protein. The Daved Fremont laboratory solved the structure of Orf7a (PDB entry 6W37). The exact function of this small protein, as for the majority of accessory proteins, is unknown.

The APS light source has become a central facility in the US for SARS-CoV-2 projects. Overall, the CSGID and other users of the APS have deposited 45 crystal structures for 11 different proteins. Many structures involve interactions with ligands and inhibitors. The CSGID structures are released to the scientific community prior to publication, and materials are available at the [NIH/NIAID BEI resource](#).

Our approach has taken us in a different direction (though very complementary) to the efforts at the Diamond Light Source in the UK. There the focus has been on determining many structures of one essential enzyme using a large-scale screen of electrophile and non-covalent fragments through a combined mass spectrometry and X-ray approach. The teams of Frank von Delft and Martin Walsh have been very successful in delivering over 110 structures of M<sup>Pro</sup> with small ligands (Douangamath *et al.*, 2020). They have identified 71 hits that span the active site and 3 hits at the dimer interface. These structures reveal potential routes for rapid development of more potent inhibitors through merging covalent and non-covalent fragment hits. We believe that this approach can provide useful structural and reactivity information for structure-based drug discovery and can serve as starting points for antivirals targeting specific enzymes. Therefore, we have established collaboration with both teams at Diamond on screening Nsp15 and PL<sup>Pro</sup> targets.

However, one crucial protein of SARS-CoV-2 thus far has eluded crystallization efforts, RNA-dependent RNA polymerase Nsp12. This is the target of one of the most promising drug candidates: remdesivir. Here, cryo-EM has become the key tool for structural information (Hillen *et al.*, 2020; Yin *et al.*, 2020). Additional cryo-EM structures reported for full-length human ACE2, in the presence of a neutral amino acid transporter B<sup>0</sup>AT1, with or without the receptor-binding domain (RBD) of the surface spike glycoprotein (S protein) of SARS-CoV-2 [PDB entry 6M17 (Yan *et al.*, 2020)] and for the spike ectodomain structure [PDB entries 6VYB, 6VXX (Walls *et al.*, 2020)], as well as structures of RBD with the SARS-CoV specific antibodies (Tian *et al.*, 2020; Wrapp *et al.*, 2020). Some complexes with antibodies have been determined by X-ray crystallography [PDB entry 6W41 (Yuan *et al.*, 2020)] or a combination of cryo-EM and X-ray crystallography.

## Outlook

Most importantly, X-ray crystallography has done exceptionally well in this important challenge and has rapidly contributed atomic-resolution data essential to the structure-based development of treatments for COVID-19. Since February 2020, high-resolution X-ray structures have been deposited in addition to 18 cryo-EM structures and 1 NMR structure. Our efforts have provided high-resolution insight into the SARS-CoV-2 virus which can be utilized by other researchers. For example, these structures have been used by the Rick Stevens and Arvind Ramanathan groups at the Argonne National Laboratory and others for *in silico* drug discovery using advanced computational approaches such as [DeepDriveMD](#).

In parallel with experimental work, a number of resources have been developed for COVID-19 that combine sequence (<https://github.com/Knowledge-Graph-Hub/kg-COVID-19/wiki>) and structural-quality evaluation data (<https://COVID-19.bioreproducibility.org/>). These sites provide important information about the genomes, connectivities, interactions and the structure-quality assessment. This work is ongoing and we hope for significant progress soon.

## Acknowledgements

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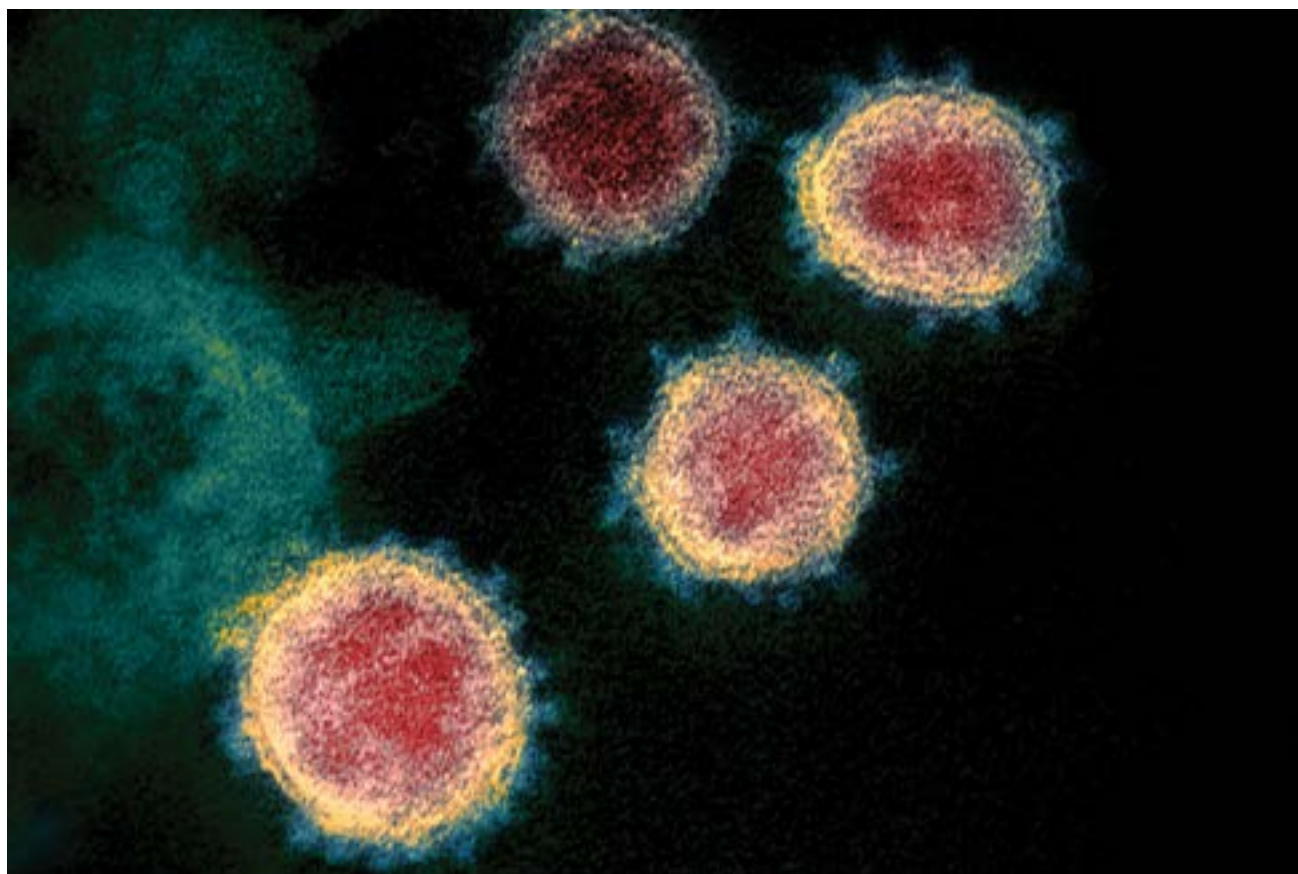
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## EDITORIAL

# COVID-19 and cryo-EM

Sriram Subramaniam



The last few years have seen spectacular growth in the widespread adoption of cryoelectron microscopy (cryo-EM). With the advent of direct electron detectors, the number of depositions in the [EM Data Bank](#) (EMDB) has increased steadily at a rate of ~30% every year since 2014, crossing the 10 000 mark in early 2020.

What may not be widely appreciated is that this extraordinary rate of growth in cryo-EM is not new. The number of depositions in the EM data repository increased on average by ~40% each year between 2004 and 2014, quickly expanding beyond the 52 entries listed in 2004. Besides the sheer increase in numbers of structures being determined using cryo-EM methods, an important aspect to this growth is the application of cryo-EM to determine structures of heterogeneous and dynamic assemblies. Protein complexes of this kind will most likely continue to remain largely intractable to crystallographic approaches that require the generation of ordered two- or three-dimensional crystals.

The rapid progress in the application of cryo-EM methods to study SARS-CoV-2 proteins provides a convincing demonstration of the power of cryo-EM in the arsenal of structural biology. Within a month or so of the availability of the gene sequence, Wrapp *et al.* (2020) and Walls *et al.* (2020) were able to obtain the first structures of the soluble part of the trimeric SARS-CoV-2 spike protein. Nearly a dozen groups worldwide have extended these studies in short order to decipher many conformational variants of the spike protein, including structures of native spikes displayed on intact viral membranes. These are major achievements that are also a testament to the speed, quality and biomedical relevance of present day cryo-EM methods. The structures provide important insights into the complexity of spike

architecture, especially in the conformational spread of the domain that binds the human ACE2 receptor. These studies, in combination with emerging structural information on the binding sites of various antibodies on the S-protein are fundamentally relevant to the design of effective therapeutics and vaccines against COVID-19.

Before January 2020, of the 84 coronavirus cryo-EM structures deposited in the repository, 82 were of trimeric spike proteins. In contrast, of the 34 entries related to SARS-CoV-2 that have been deposited in 2020, only ~50% are of spike proteins, with the rest coming from entries such as the ORF3a membrane protein, ACE2 protein in complex with the membrane protein BOAT1 and RNA polymerase complexes in different conformational states [Fig. 1(a)]. What is noteworthy about these advances is that protein complexes of this kind are often flexible and conformationally heterogeneous, making them challenging to study by X-ray crystallography. The advent of ‘single particle’ methods such as cryo-EM and XFELs offer the exciting prospect to transcend the description of proteins in terms of static structures, and instead derive conformational landscapes that may provide a much better way to understand their biological function (Ourmazd, 2019). We are now publishing an increasing number of important papers in this area in **IUCrJ**, and are particularly keen to welcome papers in **IUCrJ** and other IUCr journals that increase understanding of SARS-CoV-2.

The realization of the potential impact of cryo-EM led to the nucleation of several national and regional facilities in different countries to nurture the growth of the cryoEM field (Subramaniam, 2019). While these facilities continue to play a key role, we are now seeing growth in the cryo-EM field that is fueled by the increased availability of high-end cryo-EM instrumentation within individual institutions. For this trend to continue, it is critical that the cost of cryo-EM instrumentation necessary for high-resolution structure determination decreases significantly. Funders of biomedical research need to encourage manufacturers to drive costs down to allow the technology to be accessed more readily by the larger biological community.

The critical role played by the EMDB over the last two decades in the dissemination and impact of cryo-EM cannot be underestimated. Established at a time far before the wider recognition of the cryo-EM revolution, the access provided by the repository of structures to both specialists and nonspecialists has catalyzed the growth of the field enormously. The recent launch of **EMPIAR** as an archive where authors are able to deposit raw cryo-EM data has enabled crowdsourcing of data analysis. I enthusiastically endorse their appeal to the structural biology community to deposit COVID-19-related raw cryo-EM data in EMPIAR to enable more in-depth analysis following initial publication of the data given the rapid growth in our collective knowledge of this pandemic.

While cryo-EM techniques are being used to glean atomic level information on SARS-CoV-2, it is worth reflecting on the historical context of electron microscopy in the study of these types of viruses. In the 1960s, a young technician named June Almeida made seminal observations in the course of electron microscopic investigations of viruses. Almeida, who trained initially in Canada and subsequently worked at a hospital in London, showed that antibodies derived from infected individuals could be used to coat viruses, thereby enabling their detection in the electron microscope using heavy-atom stains to generate contrast. Among her many contributions to the imaging of viruses such as Rubella and hepatitis B in an electron microscope, one that stood out was the observation of a new kind of virus that she and her colleagues labeled as a ‘coronavirus’ in recognition of the viral surface proteins, which appeared like a crown or ‘corona’ on the membrane.

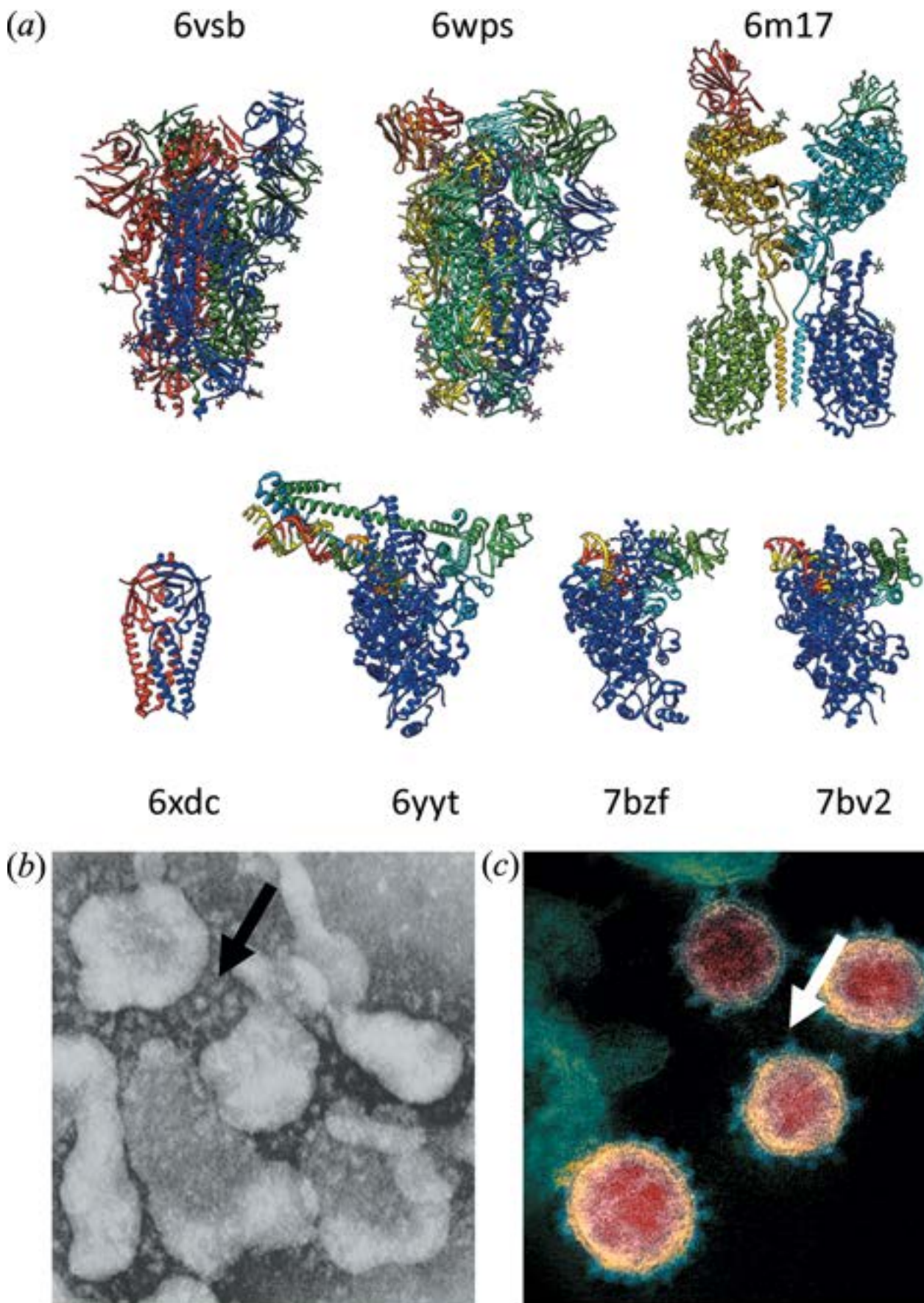


Figure 1. (a) A selection of recently reported cryo-EM structures of SARS-CoV-2 proteins including spike proteins, RNA polymerase complexes and membrane proteins in the viral envelope. The PDB identifiers of the structures are indicated. (b) Negatively stained transmission electron microscopic image (Almeida & Tyrrell, 1967) of a novel virus that was named a 'coronavirus' by June Almeida in 1966. (c) Colorized transmission electron microscope image of negatively stained infectious

SARS-CoV-2 particles recorded at NIAID's Rocky Mountain Laboratories (RML) in Hamilton, Montana. The arrows in panels (b) and (c) point to the spike proteins on the surface of the virus particles.

The resemblance of June Almeida's image from 1966 [Fig. 1(b)] of a sample referred to as B814 and the iconic recent image of the SARS-2 coronavirus [Fig. 1(c)] produced by scientists at the NIAID laboratories in Montana is remarkable. Looking ahead, a major goal of structural virology will be to bridge the gap between the knowledge of atomic resolution structures of individual viral components and the critical macromolecular interactions that define their assembly along with RNA into an intact, infectious virus. These kinds of integrative structural studies, combined with biological analyses of the processes involved in viral replication will be essential to understand the molecular mechanisms by which SARS-CoV-2 enters cells to cause its devastating effects on human health.

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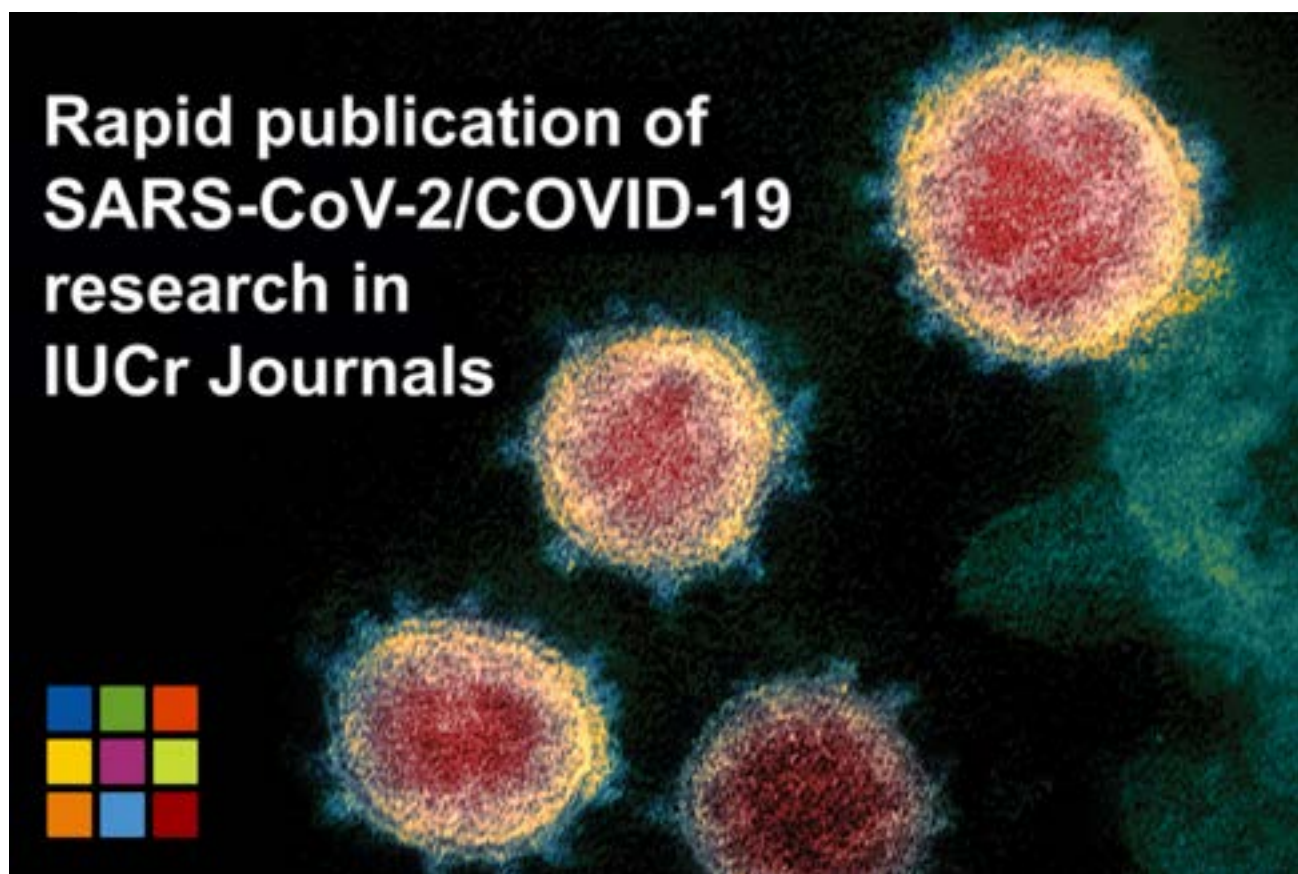
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## IUCr JOURNALS NEWS

# Rapid publication of SARS-CoV-2/COVID-19 research in IUCr Journals



The following IUCr journals are now taking part in a [rapid review system](#) for COVID-19 research:

- *Acta Crystallographica Section D, Structural Biology*
- *Acta Crystallographica Section F, Structural Biology Communications*
- **IUCrj**

We have put together a dedicated team of editors and reviewers who are ready to peer review your article on the structural biology of SARS-CoV-2. Upon acceptance, publication in our journals will be expedited.

Structure determinations using any structural biology technique - including crystallography, SAS, NMR, cryo-EM and/or other techniques - are welcomed. Articles describing new methods or software developments for COVID-19 research are also encouraged.

We expect articles to be published [open access](#) to make your research freely available to as wide an audience as possible. Fees may be covered by transformative deals or discounted rates are available by contacting the [Editorial Office](#).

To take advantage of our rapid review system, submit your article to one of our journals using the buttons below.

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SUBMIT



SUBMIT



SUBMIT

## IUCR JOURNALS NEWS

## Three new Co-editors appointed to *Acta Crystallographica* Section D, Structural Biology



The Editors of *Acta Cryst. D*, Randy Read and Elspeth Garman, have welcomed three new Co-editors to the editorial board, saying "They enhance the already impressive range of expertise in our current board, and they help to reflect the diversity of our subject coverage, authors and readers".

Kristina Djinović-Carugo is Head of the Department of Structural and Computational Biology in the Max Perutz labs in Vienna, Austria. With a major research interest in understanding the structural biology of complex systems such as the muscle Z-disk and the actin-based cytoskeleton, as well as of metalloproteins, she has developed expertise in integrative structural biology techniques that combine a variety of methods.

Ana González is the BioMAX beamline manager at the MAX IV synchrotron facility in Lund, Sweden. She brings 30 years of experience at a variety of synchrotrons around the world (Daresbury SRS, ESRF, EMBL Hamburg and SSRL before moving to Sweden) to the journal, with expertise in synchrotron science, radiation damage, optimal data collection for different phasing experiments, and the use of XFEL sources.

K. R. Vinothkumar is based at the National Centre for Biological Sciences of the Tata Institute of Fundamental Research in Bangalore, India. Prior to this, he developed expertise in cryo-EM techniques working with Werner Kühlbrandt at the MPI in Frankfurt, Germany, and with Richard Henderson at the MRC-LMB in Cambridge, UK. His current research on membrane proteins and macromolecular complexes

employs a combination of cryo-EM and X-ray diffraction, relevant to the growing number of cryo-EM papers submitted to *Acta Cryst. D* and other IUCr journals.

## IUCR JOURNALS NEWS

# Journal of Synchrotron Radiation: Open call for new Main Editor to join leadership team



The *Journal of Synchrotron Radiation* (*JSR*) is one of the 10 leading journals published by the IUCr. In this context, *JSR* provides comprehensive coverage of the entire field of synchrotron radiation and free-electron laser (FEL) research including instrumentation, theory, computing and scientific applications in areas such as biology, nanoscience and materials science. While presently a hybrid journal with both fully open-access papers and papers accessible on a traditional subscription basis, it is planned that *JSR* will become a 100% open-access research journal in the near future.

At this time, we seek a new Main Editor of *JSR*. The successful candidate will work with the existing *JSR* Main Editors, the entire *JSR* Editorial Board, its Managing Editor and staff in the IUCr Editorial Office in Chester, UK, as well as the IUCr Journals Editor-in-Chief, to take *JSR* through this important transition. The successful candidate will help provide dynamic leadership of *JSR* through a smooth transition to open access while maintaining its leading position in the publication of synchrotron- and FEL-based research across all relevant fields.

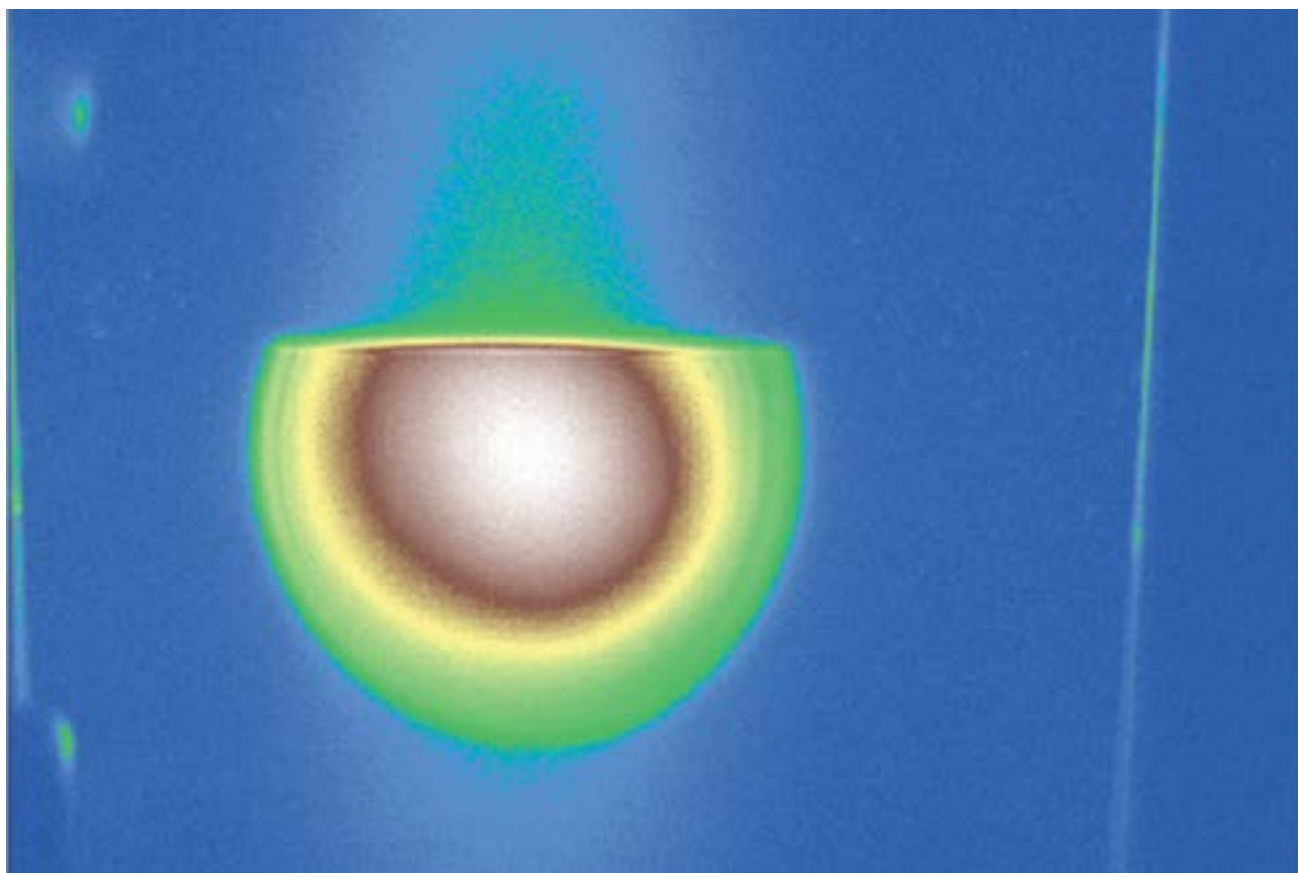
In addition to sharing in the editorial workload for papers submitted to *JSR*, the Main Editors provide leadership for the journal's Editorial Board, scientific development and welfare. Working with the *JSR* Managing Editor and staff in Chester, the recently appointed IUCr Journals Commissioning Editors, and the Editor-in-Chief, the *JSR* Main Editors recruit new Co-editors, commission Lead and Feature Articles, as well as plan or commission Special Issues, including the recruitment of Special Issue Guest Editors. In all these aspects, please note that the IUCr is committed to improving gender and geographical balance

while maintaining the highest scientific standards, and everyone associated with IUCr Journals is expected to respect these guidelines.

Appointment criteria for IUCr Journals Main Editors are given [here](#) but some flexibility will be applied in appointing the best possible candidate at this time for what is anticipated to be a key role for pioneering the future of *JSR*, and IUCr journals as a whole.

Please write in confidence **by 31 August 2020** to the Editor-in-Chief ([eic@iucr.org](mailto:eic@iucr.org)) and Executive Managing Editor ([med@iucr.org](mailto:med@iucr.org)), enclosing your CV, a list of your 20–25 most important publications, your vision for the future development of the *JSR* in 500 words, and a full list of publications.

## Two new virtual special issues of Journal of Synchrotron Radiation



### PhotonDiag2018 Workshop special issue

The *Journal of Synchrotron Radiation*'s first virtual special issue contains some highlights of the PhotonDiag2018 Workshop on FEL Photon Diagnostics, Instrumentation, and Beamlines Design, which took place in Hamburg, Germany, in September 2018. The workshop, the fourth in the PhotonDiag series, focused on advances in X-ray optics and photon diagnostics, one of the most dynamically evolving topics in the X-ray science world.

Guest-edited by Daniele Cocco, Jan Grünert, Elke Ploenjes, Kai Tiedtke and Marco Zangrando, these [virtual proceedings](#) start with the articles focused on diagnostics to study the spectral distribution, the wavefront, the intensity, the profile, the polarization and the temporal distribution. The combined use of photon diagnostics and electron diagnostics to study machine performance follows, and the third part deals with more conventional X-ray optics, from metrology over adaptive focusing mirrors to crystal monochromators. The proceedings close with one crucial component to most experiments and diagnostics, *i.e.* the X-ray detectors.

### X-ray Free-Electron Lasers special issue

The last five years have seen a remarkable explosion in both the availability and capabilities of X-ray free-electron lasers (XFELs). Considering this rapid progress, it was considered timely for another special FEL-related issue in the *Journal of Synchrotron Radiation*, following the [first one](#) published in May 2015.

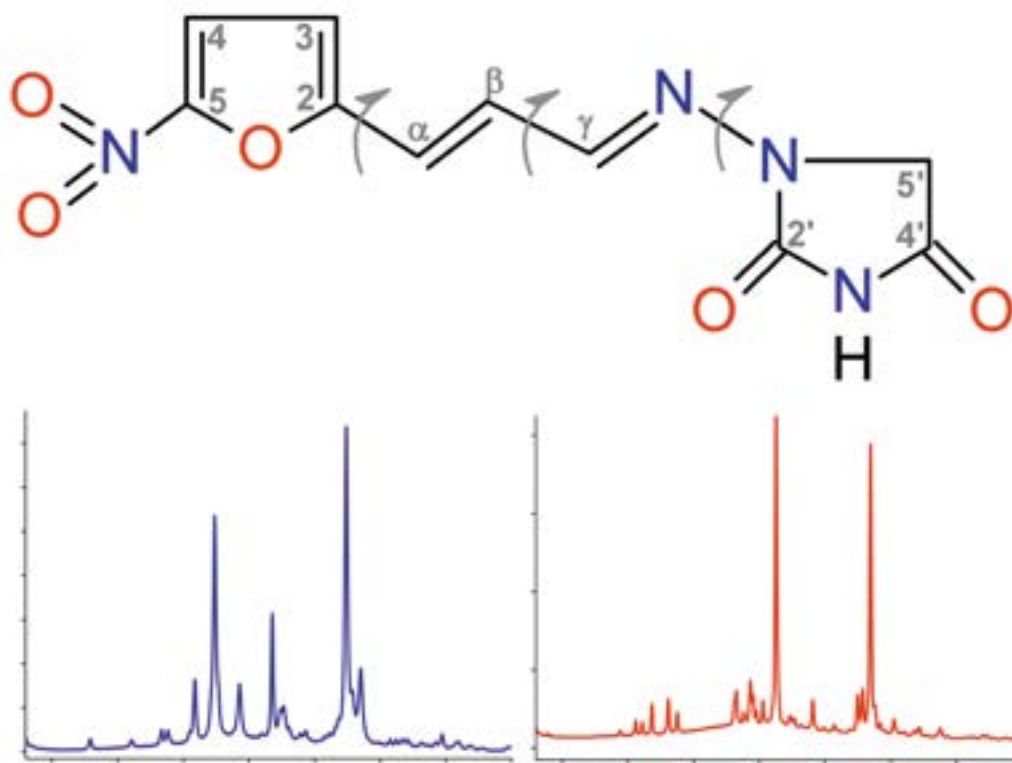
The current [virtual edition](#), guest-edited by P. H. Fuoss, I. Schlichting, T. Tschentscher and M. Yabashi, provides information to guide both X-ray experts and new users.

The issue features articles describing a variety of sources and facilities, including FLASH, LCLS, SACLA, SwissFEL, PAL and EuXFEL, that give insight into the current status, development plans and scientific directions at these facilities. Papers focused on individual instruments provide detailed information into new experimental capabilities along with recent results highlighting the scientific opportunities being developed at XFELs. Overviews of the design and performance of important subsystems, for example laser systems for time-resolved measurements, give critical information to plan successful XFEL experiments. In particular, a number of papers focus on the infrastructure requirements and challenges at the European XFEL, the first of the next generation of megahertz repetition rate XFEL producing hard ( $\leq 1$  Å wavelength) X-rays. Finally, new X-ray scientific methods are constantly being ported to, and developed at, FEL sources. This edition highlights a few of these capabilities including spectroscopy techniques.

## COMMENTARY

# Combining forces: complementary techniques brought together to determine tricky crystal structures

Graeme M. Day



In a recent issue of *Acta Crystallographica Section B*, Dudek *et al.* (2020) report the determination of two crystal structures of the pharmaceutical molecule furazidin, whose existence has been reported in patents, but whose structures have to date been uncharacterized. The work shows how multiple techniques – powder X-ray diffraction, nuclear magnetic resonance (NMR) spectroscopy and computational crystal structure prediction (CSP) – are sometimes required to determine crystal structures of organic molecules when direct determination from diffraction data is not possible, and demonstrates the complementarity of information provided by the different methods.

Crystallization of furazidin in this work produced microcrystalline powders and no single crystals that could be used for crystal structure determination. This is a common problem that can frustrate attempts to understand the structure of a material. However, developments in applying NMR techniques to solids, along with quantum mechanical methods that can provide accurate predicted NMR parameters from putative structures, can offer an alternative to diffraction-based structure solution. This approach of determining a crystal structure by constructing or identifying a structural model that reproduces the observed NMR spectra is termed NMR crystallography. A challenging stage in applying NMR crystallography is the generation of realistic structural models to which quantum mechanical predictions of NMR parameters can be applied.

This is where CSP has entered the field of NMR crystallography; computational methods for CSP have been developed to predict, from nothing more than the chemical diagram, the likely crystal packings of a molecule. While CSP is often discussed as an *ab initio* method, requiring no input of experimental data, these methods can also be combined with experiment, providing the missing ingredient in NMR crystallography determination of structures. The process seems simple: apply CSP to predict possible crystal structures, perform calculations to predict the NMR parameters for the best predicted structures and compare these with the measured NMR spectra. However, CSP for molecules of the size and flexibility of furazidin is challenging, particularly when the experimental information shows that there are two symmetrically unrelated molecules in one of the polymorphs. CSP is relied on in this method to locate all possible structures, which correspond to local energy minima on the energy surface defined by all degrees of freedom of the crystal structure. Molecular flexibility and multiple independent molecules add to these degrees of freedom and lead to very large numbers of energy minima, often numbering in the hundreds or thousands within the usual energetic range of polymorphism. This creates a computational challenge in itself – the location of *all* possible structures – and creates a second problem for NMR crystallography: within a large set of computer-generated crystal structures, one can find false-positive agreement with the measured NMR spectra. These are structures where calculated chemical shifts seem to be close enough to experiment (within the errors of the quantum mechanical methods used to predict the NMR), but where this agreement is fortuitous, and the structure is incorrect.

The complexity of CSP is reduced in this work by identifying constraints from the experimental data to the degrees of freedom that need to be explored. Diffraction data and NMR are able to provide complementary views of the structure; NMR gives local structural information, reporting on molecular geometry, close contacts and nearest neighbour interactions, while the most readily extracted information from diffraction is the long-range periodicity of the molecular arrangement. Powder X-ray diffraction was used to narrow the list of possible space-group symmetries that needed to be considered during structure prediction, while NMR provided information to narrow the set of molecular conformations, which further reduced the search space. This led to a tractable CSP problem.

The issue of false positives was addressed by comparing calculated and measured NMR parameters for multiple nuclei – here, both  $^1\text{H}$  and  $^{13}\text{C}$ . While incorrect structures sometimes reproduced either the measured  $^1\text{H}$  or  $^{13}\text{C}$  chemical shifts, only one structure for each polymorph was found to reproduce both. This is an important message from this study: not to rely solely on  $^1\text{H}$  or  $^{13}\text{C}$  data because of the risk that agreement with experiment is spurious.

The report by Dudek and coworkers of integrating these techniques – diffraction, NMR and structure prediction – to determine crystal structures that could not have been determined using a single method is an important example that nicely illustrates the process and demonstrates the importance of careful validation.

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This article was originally published in *Acta Cryst.* (2020). **B76**, 294–295.

## COMMENTARY

# Shock-synthesized quasicrystals

Péter Németh

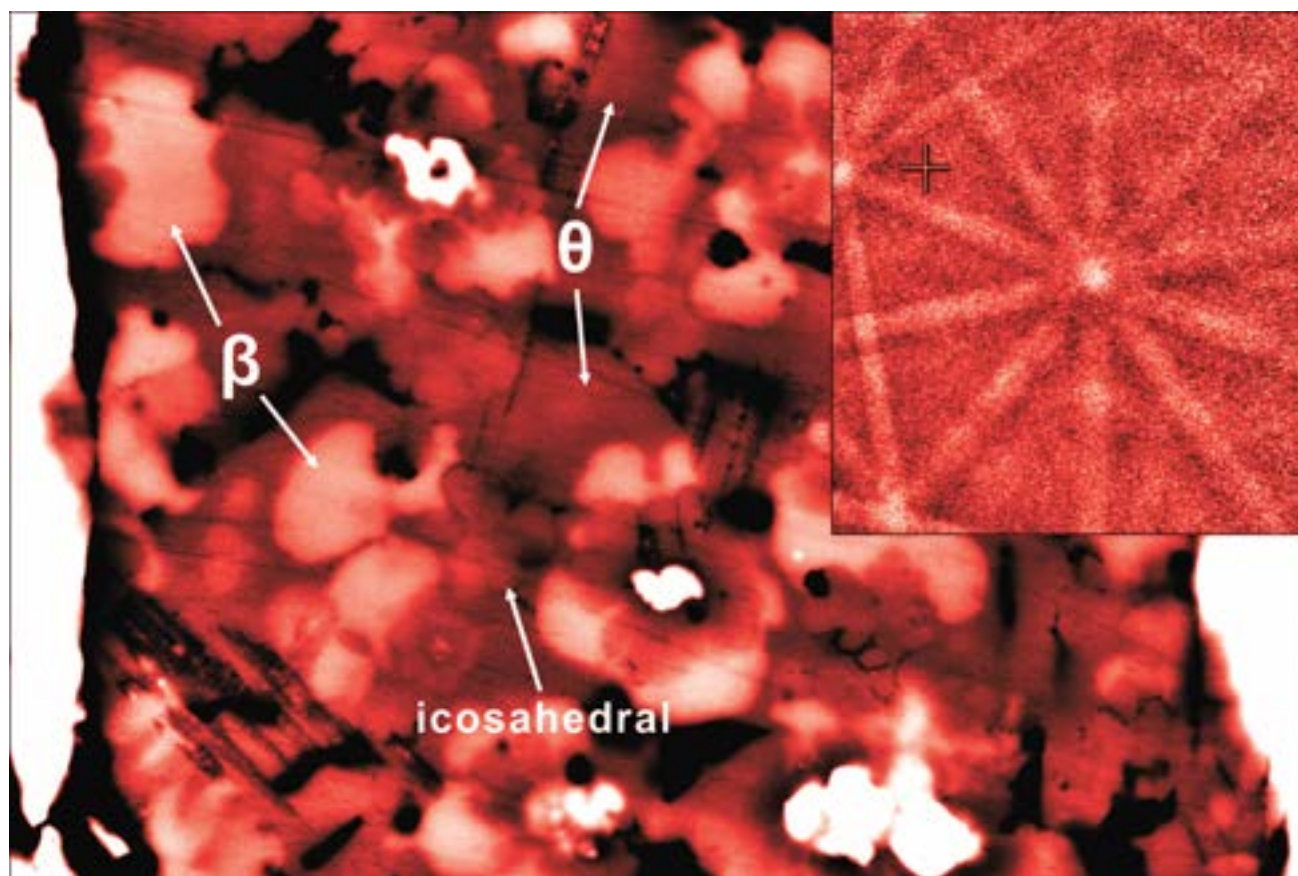


Figure 1. The  $\text{Al}_{62}\text{Cu}_{31}\text{Fe}_7$  icosahedral quasicrystal revealing fivefold rotation and its coexisting Cu–Al phase assemblage of stolperite ( $\beta$ ,  $\text{AlCu}$ ) + khatyrkite ( $\theta$ ,  $\text{Al}_2\text{Cu}$ ) from a laboratory-shocked sample. Adapted from Hu *et al.* (2020).

Impact-processing of planetary materials and laboratory-shock experiments are of great interest. During these extreme processes a hypervelocity shock wave generates extremely high pressure and high temperature for exceptionally short times (micro- and nanoseconds) and provides favourable conditions for the formation of peculiar structures. In a recent **IUCrj** article, Hu *et al.* (2020) report the shock synthesis of an unusual material, the icosahedral  $\text{Al}_{62}\text{Cu}_{31}\text{Fe}_7$  quasicrystal (QC), which was previously identified in the Khatyrka meteorite (Bindi *et al.*, 2016).

QCs are a unique type of material, which are characterized by sharp diffraction features and possess symmetry elements that apparently are inconsistent with the conventional rules of crystallography. In particular, the first  $\text{Al}_{86}\text{Mn}_{14}$  alloy QC to be recognized has icosahedral symmetry featuring among other things fivefold rotation axes (Shechtman *et al.*, 1984). The appearance of these unconventional rotations triggered exciting discussions right from the first report [reviewed in Prodan *et al.* (2017) and <https://paulingblog.wordpress.com/tag/quasicrystals/>]. On the one hand, Shechtman and colleagues presented QCs as a new type of crystalline material, in which atoms are repeated according to a quasiperiodic translation (Lifshitz, 2003). The interpretation of the QCs' structure was also approached with the so-called Amman tiling, the 3D equivalent of the 2D Penrose tiling (Lord *et al.*, 2000). On the other hand, Pauling insisted that no crystallographic rules were violated and considered QCs as multiply twinned cubic structures (e.g. Pauling, 1985). Although it turned out that Pauling's interpretation did not

hold, recently Prodan *et al.* (2017) reported that QCs could in fact be interpreted as multiply twinned structures, considering the unit cell of the primitive golden rhombohedra, and demonstrated that ‘tiling’ and ‘multiple twinning’ are fully compatible with each other. However, this interpretation requires the occurrence of equal size twin domains, which is uncommon in nature. In either way, the crystallographic identity of QCs seems to be still debated.

Since their discoveries, over a hundred QCs have been described from syntheses. In contrast, only a few studies report QCs from natural materials and, in fact, all have been associated with the Khatyrka meteorite. So far three new QC types have been described from this meteorite: the i-phase I with composition of  $\text{Al}_{63}\text{Cu}_{24}\text{Fe}_{13}$ , officially named icosahedrite (Bindi *et al.*, 2009); the d-phase with composition of  $\text{Al}_{71}\text{Ni}_{24}\text{Fe}_5$ , referred to as decagonite (Bindi *et al.*, 2015); and the i-phase II with  $\text{Al}_{62}\text{Cu}_{31}\text{Fe}_7$  composition (Bindi *et al.*, 2016). However, this list perhaps can be extended with additional extraterrestrial phases (personal communication with Luca Bindi).

In either way, to date three natural QCs are known from the Khatyrka meteorite. In meteorite research the mineral assemblage can aid understanding of the formation condition of an associated phase; however, the proper interpretation demands a laboratory-controlled synthesis. Hu *et al.* apply recent advances in the Graded Density Impactor fabrication technique (Kelly *et al.*, 2019) to shock-synthesize i-phase II. The authors use a special Al-Cu-W disk in an Fe-bearing 304 stainless-steel target chamber and perform two experiments corresponding to an estimated peak shock pressure of 20–30 GPa for 800 ns and 30–35 GPa for 600 ns, respectively. The careful sample characterization, including the state-of-the-art electron backscattered diffraction method, reveals the first experiment (Fig. 1) resulted in coexisting intermetallic phases of i-phase II, stolperite ( $\beta$ , AlCu) + khatyrkite ( $\theta$ ,  $\text{Al}_2\text{Cu}$ ), which perfectly matches with the assemblage found in the Khatyrka meteorite. The second experiment shows the formation of a rather Fe-rich quinary i-phase ( $\text{Al}_{68.6}\text{Fe}_{14.5}\text{Cu}_{11.2}\text{Cr}_4\text{Ni}_{1.8}$ ), together with stolperite ( $\beta$ , AlCu) and hollisterite ( $\lambda$ ,  $\text{Al}_{13}\text{Fe}_4$ ).

A key finding of Hu *et al.* is the shock-synthesized i-phase II, because it represents the first example of a quasicrystal composition discovered in nature prior to being synthesized in the laboratory. Although i-phase I (icosahedrite) together with the d-phase (decagonite) were previously produced by Al-alloy quenching experiments and reported to be thermodynamically stable with a very narrow range of composition (Tsai *et al.*, 1987), such laboratory conditions were likely inconsistent with those experienced by the Khatyrka meteorite. Furthermore, the previous Al-alloy quenching experiments could not explain the stability of i-phase II and its relationship to i-phase I. Now the authors’ findings provide unique insights into the intricate details of the Al-Cu-Fe phase diagram. In particular, they suggest the thermodynamically stable  $\text{Al}_{63}\text{Cu}_{24}\text{Fe}_{13}$  phase could separate into two disconnected fields under shock pressure above 20 GPa, leading to the co-existence of Fe-rich (i-phase I) and Fe-poor (i-phase II) QCs consistent with the previous findings of the Khatyrka meteorite (Bindi *et al.*, 2016).

QC formation requires controlled syntheses including rapid quenching, during which unusual crystal nucleation and growth can be favoured. The new findings reported by Hu *et al.* demonstrate that shock-recovery experiments provide a novel strategy for preparing and exploring this exciting material group.

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This article was originally published in [IUCrJ \(2020\). 7, 368–369](#).

## COMMENTARY

# Making sense of vacancy correlations with single-crystal diffuse scattering data

Matthew Krogstad

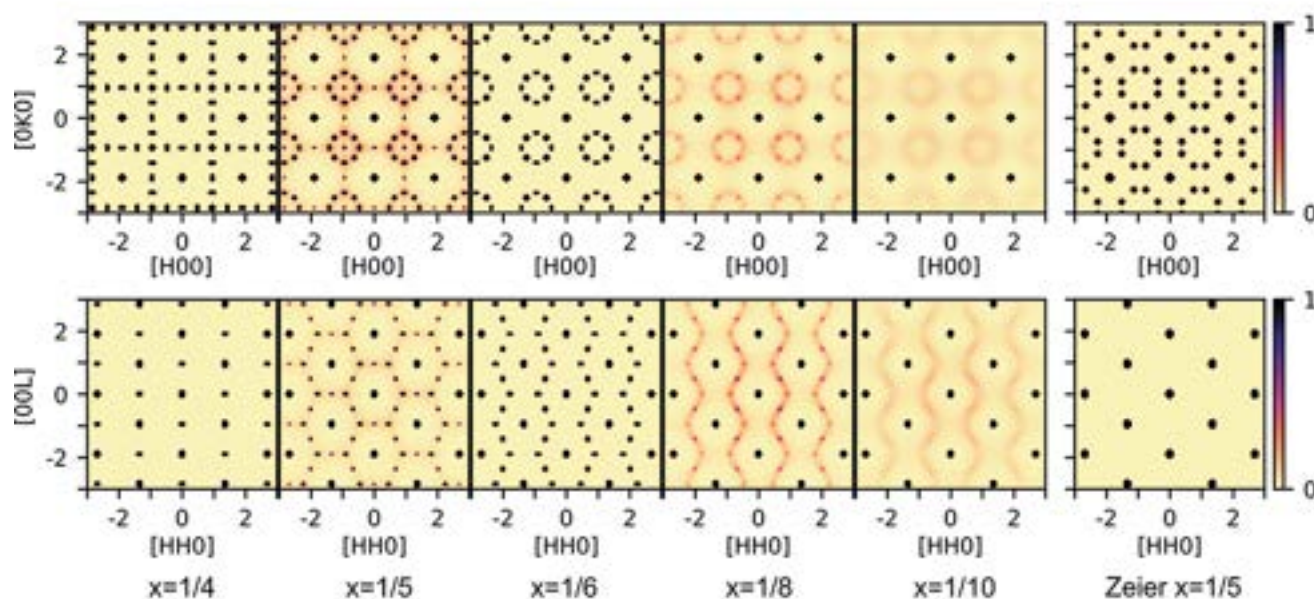


Figure 1. Scattering data from simulated crystals of  $\text{Nb}_{1-x}\text{CoSb}$  from Roth *et al.* (2020). For  $x = 1/4$ , an ordered state is produced where there are no nearest-neighbor vacancies, producing a commensurate set of superstructure peaks, and for  $x = 1/6$ , next-nearest neighbors can be completely avoided as well, producing eight-point rings when appropriately twinned. For concentrations with  $1/4 > x > 1/6$ , a disordered mixture of the two states is shown to minimize energy compared to proposed ordered states and reproduces observed scattering in this range. For concentrations with  $x < 1/6$ , disordered states produce broad rings and waves.

Local ordering of atoms within a crystalline structure is increasingly recognized as essential to understanding the properties of a variety of interesting materials. This is shown by the success of pair distribution function (PDF) analysis of scattering data from powder samples to investigate departures from perfectly crystalline order (Billinge & Kanatzidis, 2004; Egami & Billinge, 2012). Diffuse scattering from single crystals can also be a powerful probe of local order, potentially providing key information that would be difficult to obtain from PDF methods; however, difficulties in both measurement and interpretation of data have too often restricted their use by a larger community. In this issue of **IUCrJ**, Roth *et al.* (2020) demonstrate how simple concepts can be applied to better understand single-crystal diffuse scattering from half-Heusler systems.

Recent developments in instrumentation and computing have made the measurement of single-crystal diffuse scattering much easier than it was even ten years ago (Ye *et al.*, 2018; Keen *et al.*, 2015).

Valuable data can also be obtained with lab-based X-ray sources and electron sources, which are able to produce high-quality data in a carefully chosen set of planes (Izquierdo *et al.*, 2011; Welberry & Goossens, 2016). However, the analysis of such data can still seem a daunting task, especially when compared to the powder-based PDF data. A key step for proposed solutions is comparison of experimental scattering data with those from simulated structures, but it can be difficult to connect diffuse scattering from simulated structures to specific structural motifs.

Roth *et al.* (2020) have produced a compelling analysis of diffuse scattering from half-Heusler materials  $\text{Nb}_{1-x}\text{CoSb}$ . These have proven challenging to understand via crystallographic techniques; while crystallographic solutions can confirm the concentration of vacancies on the Nb sites, the distribution of these vacancies is more difficult to discern. Electron diffraction data collected by Xia *et al.* (2018, 2019) show clear diffuse scattering patterns over a range of concentrations, implying that vacancies are spatially correlated locally without being ordered over the entire structure. Simulations based on density functional theory (DFT) have reproduced the scattering for certain concentrations but cannot be generalized over the full range of vacancy concentrations and provide limited insight into the systematics of local structures in these systems.

Roth *et al.* (2020) approach these data starting with the simple concept that Nb site vacancies will tend to avoid one another. Simulated crystals are then generated by a Monte Carlo procedure which allows vacancies to move and penalizes vacancies being nearest-neighbors and next-nearest neighbors via a simple Ising-like model. Diffuse scattering from these models is rapidly calculated using the program *SCATTY* (Paddison, 2019). While the number of individual states produced by these models is quite large, the only free parameters are the vacancy concentration, the ratio between energies associated with nearest and next-nearest neighbors, and the temperature.

For a single ratio of energies, these models impressively reproduce published electron diffraction data over a range of vacancy concentrations (see Fig. 1). Distinct ordered ground states are reproduced at  $x = 1/4$  and  $x = 1/6$ , with intermediate compositions showing short-range combinations of the two. For concentrations with  $x < 1/6$ , broader wave-like scattering is seen. This entire range of compositions can be modeled and explained with self-avoiding vacancies.

These results provide a significant advance in the knowledge of the structure of these materials as well as a great example of how short-range ordering can be understood, modeled and described. Roth *et al.* (2020) have provided a clear picture of occupational correlations in half-Heusler compounds. With single-crystal diffuse scattering data becoming easier to obtain, the use of simple models of short-range substitutional correlations is a key method for understanding such data.

## Acknowledgements

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## IUCr ACTIVITIES

# The IUCr's Finance Committee

Malcolm Cooper



## Background

What is the background against which the IUCr's Finance Committee (FC) operates? Well, the IUCr is an international 'not for profit' organisation registered in Switzerland but with its offices and all its employed staff, amounting to about 25 people, working in Chester in the UK. It exists to serve crystallographers from around the world by bringing our community closer together, providing a home for its publications, supporting scientists (especially young scientists) and delivering other forms of scientific outreach. The IUCr is not subject to UK tax on any of its trading profits, and Swiss tax is not levied as long as the commercial value of the time freely donated by Union members to IUCr work matches the revenue generated by the sales of the Union's journals, which, thankfully, the Swiss authorities currently accept that it does. In order to preserve its tax-free status within Great Britain, meetings of the FC and the Executive Committee (EC) have to take place outside the UK, and all investment portfolios have to be 'offshore'.

Thus the finances of the Union are far from straightforward, and it is no surprise that the IUCr needs an FC with long-term membership and a long-term memory (!) to advise each successive EC, as the EC's composition changes significantly every three years. One exception to that rule is the EC post of General Secretary and Treasurer (GST), which has a tenure of nine years precisely because the holder definitely has to master the intricacies of the finances. My tenure on the FC as its Convenor is actually much longer than that: I joined the FC over two decades ago and still fulfil that role.

## Multicurrency complications

A complication arising from the international nature of the Union is that, inevitably, several currencies are involved. For example, most of the income is in US dollars, and most expenditure is in pounds sterling, and yet, until recently, the accounts were expected to be presented in Swiss francs (CHF). Investments are usually spread across both sterling- and US dollar-denominated funds. Recently it has become possible to present the accounts in US dollars rather than in CHF but, nonetheless, currency fluctuations can still create mayhem and lead to exchange-rate losses and gains on the balance sheet, which are difficult to understand and interpret.

Broadly speaking, the Swiss franc has appreciated steadily over decades against most other major currencies, and the pound sterling has depreciated markedly and fluctuated noticeably. Wherever practicable in this article, I will quote values in US dollars, but the Swiss franc and the US dollar are conveniently currently of very similar value. Counter-intuitively, the fall in the value of sterling that we have seen over the last decade against a broad range of currencies is not bad news for the Union's finances. This is because most of our expenses are the salaries of the Chester office personnel, and these are in pounds sterling. In contrast, most of the income is made up of journal subscriptions, which are mainly denominated in US dollars.

Actually, the only income we have directly in Swiss francs (CHF) is the annual dues from the 52 adhering bodies who pay in multiples of 1,000 CHF per annum depending on the number of votes they hold in the General Assembly. Historically this round number unit has not been changed with inflation, and the total yearly subscription income now represents just about 5% (say approximately 175,000 US dollars) of the annual turnover of the Union, which is, in round figures, approximately 3,500,000 US dollars. The dues would not be enough by themselves to run a Union Secretariat, let alone a publishing arm, and so it is essential that the IUCr has profitable products such as its portfolio of journals to provide another much larger income stream.

## Tasks

In years gone by, one major task of the FC was to fix the annual price rises of the journals in order to maintain a sufficient gap between the income and expenditure to fund all the Union's activities. This would, of course, include the obvious IUCr 'good works' like the support of young scientists – as recommended by the IUCr's Calendar Sub-committee – at meetings organised by the crystallographic community worldwide, ventures such as 'Crystallography in Africa' and other outreach activities, such as 'OpenLabs'. All these initiatives are reported and commented upon from time to time in the *Newsletter* and on the IUCr webpages. In addition to funding these activities, it is necessary to amass sufficient resources to invest in updating the journal publication process to keep the IUCr in the frontline of publishing. Looking back, this was especially important when journals transitioned to becoming 'electronic', because a large investment in research and development was necessary. It was successfully achieved using our own investment reserves.

The necessary across-the-board excess of income over expenditure on journals historically was referred to as 'Bob's gap' after one of my predecessors as Convenor – Bob Diamond. Then, when Sine Larsen was the GST (1996–2005), we refined the approach. It was decided that the prices of individual journals should be more closely related to their specific costs of production, including all aspects of technical editing etc. Thereby the pricing of each journal represents the work involved in its production. More recently, we have tended to tie our annual price rises to those of our agents, Wiley, who sell subscriptions to our journals and many others to individual organisations and, more and more, to consortia of buyers who expect a significant discount due to 'bulk buying'.

Increasingly we have had to decide about the open-access charges that should be levied on authors to provide articles freely open to all readers. Open-access income is now a very significant element of the journals' revenue, and it is rising year on year as more authors and funding agencies opt to have

research papers published free for all to view. There is a natural desire to keep these charges as low as possible – to help serve our crystallographic community – but, at the same time, that wish has to be tensioned against the need to optimise income in order to provide a surplus that can be used to fund the IUCr's work.

## Membership

So, who is involved in the FC? Historically the Convenor used to be a member of the EC who happened to be based in the UK and therefore could liaise easily with the Chester office (this was in the days before the universality of email) and who was familiar with UK employment laws and practices, which is still an important consideration. The choice of the term 'Convenor' rather than 'Chair' implies that the role is to bring together a group of experts to discuss the finances and make their recommendations to the EC. In the run-up to the 1996 IUCr Congress in Seattle, Mike Hart, then at Manchester University, was the Convenor. When he moved from the UK to become the Director of the National Synchrotron Light Source in the USA, I stepped into the role as an independent, UK-based successor.

The composition of the FC, which meets twice a year, is somewhat flexible (the present incumbents' names are given below in parentheses). Firstly, given the heavy financial reliance on the functioning of the journals, it is no surprise that the journals' Editor-in-Chief (Andrew Allen) is a key member, as is the Executive Managing Editor (Peter Strickland) on an *ex-officio* basis. Of course, the Executive Secretary (Alex Ashcroft) must be there to service the meeting and contribute to it from a comprehensive knowledge of the IUCr's activities and finances. These days we also regularly invite the Executive Outreach Officer (Michele Zema) to be present as it's his job to spend some of our money! From the EC, the GST (Luc Van Meervelt) is an unquestionably essential member of the committee, as is the current President of the IUCr (Sven Lidin). Hanna Dabkowska, currently EC Vice-President, is another member of the FC with a keen interest in our finances. In addition, we are lucky to have a well-known crystallographer with commercial experience (Mike Glazer) on the committee. Finally, our current investment advisor is Narasinga Rao, who has also been guiding the ACA's investment portfolio for many years.



Attendees at the March 2019 FC meeting (left to right): Andrew Allen, Malcolm Cooper, Luc Van Meervelt, Hanna Dabkowska, Sven Lidin, Peter Strickland, Alex Ashcroft, Mike Glazer and Michele Zema. Not pictured: Narasinga Rao.

## Investments

The IUCr has investment reserves of a similar magnitude to its annual turnover. In fact, if push came to shove and we had to survive on our assets alone and at short notice, our current funds would cover about 10 months of operation. These reserves are certainly needed as a minimum 'rainy day' provision. For example, if the COVID-19 pandemic causes a severe lack of submitted papers or a reduction in the number of purchasers of the journals, these funds would hopefully allow us to ride out the ensuing storm. On a more positive note, the investments are there to finance journal developments, such as the 'electronification' of all the journals over the last two decades, and the launch of new journals, such as the *Journal of Synchrotron Radiation* in 1994 and more recently **IUCrJ** in 2014. New publications swallow money at an alarming rate in their early years: it can cost more than half a million US dollars before a major new journal begins to show an operating profit. That money must come out of our investment reserves, with the hope that one day the cash flow can be reversed, and then the journal's profits can be channelled back into investments for future needs. In our experience, most of a decade is needed to establish a journal to the stage where it is financially profitable.

Another big-ticket item, which was a landmark in the history of the Union, was the International Year of Crystallography (IYCr2014). This undoubtedly was a big help in raising the profile of crystallography within the wider scientific community. However, the activities before, during and after IYCr2014 did cost the IUCr a lot of money. Between 2012 and 2016, the IUCr's assets decreased by approximately 2 million US dollars, in no small part due to the International Year. In 2017 the FC was faced with some tough choices to safeguard the immediate future of the Union and recommended several cost-cutting measures, which were adopted. Thankfully, consequently, the situation has slowly but surely improved, but the FC is maintaining its vigilance.

All this necessary emphasis on investments means that we are subject to the vagaries of the stock markets and become nervous followers of fluctuations of the FTSE (UK) and the Dow Jones (USA) indices. The role of our financial guru is to get the best advice possible for an organisation that is interested in conservative investments: ones at the low-risk end of the spectrum. We do not seek high-risk investments that could make spectacular gains – or spectacular losses – but 'safe' ones that should return some modest capital appreciation and perhaps deliver a dividend. Before COVID-19 reared its ugly head, the IUCr assets stood at some 3.7 million US dollars, of which, in round terms, about 35% was held in sterling, 40% in dollar holdings and almost 25% in a CHF account. The US dollar has held up well as the international currency of last resort, and the Swiss franc continues to appreciate as another safe haven. The UK stock market took a bit of a hit after the arrival of COVID-19 but the Dow Jones has shown more resilience. Overall, at the time of writing, our assets have not been badly affected by the global pandemic.

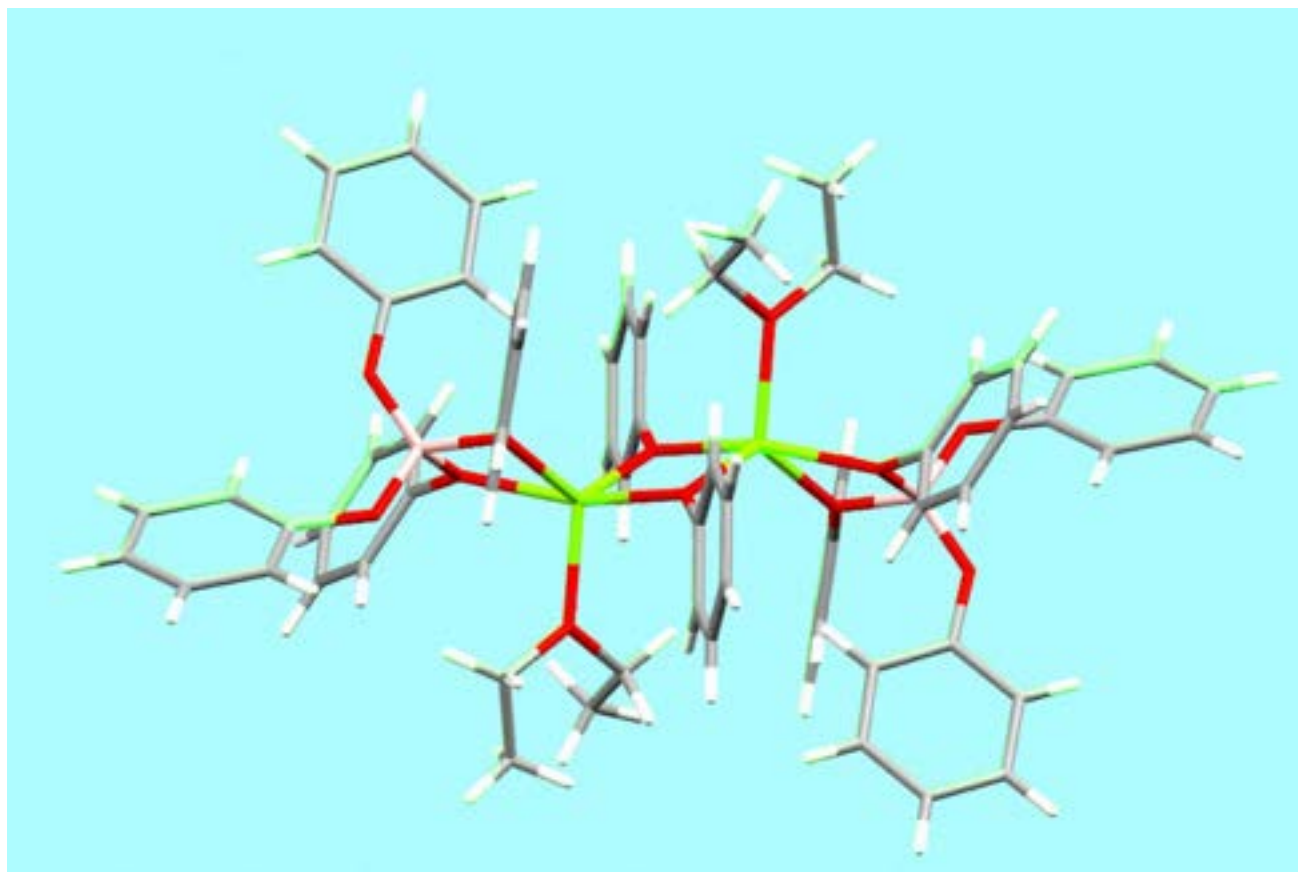
## The future

What does the IUCr's financial future look like after COVID-19? Well, on the positive side, there are a large number of crystallographers working from home right now, hopefully writing up a backlog of research papers destined for one of our journals! On the negative side, there may be libraries being directed by their administrations to trim their journal subscriptions for 2021, and we might be in the firing line there. After a number of years with worrying negative balances, the 2019 financial out-turn was a modest but very satisfying operating profit of around 10% of turnover, and 2020 might possibly be similar. Clearly, open access will become more and more important financially, but beyond that, it would be unwise to speculate. The future is somewhat uncertain, given the rapidly evolving state of the publishing industry, and we may yet be grateful for our 'rainy day' reserves.

## FEATURE ARTICLE

# Why do so many crystal structures remain unpublished?

William Clegg



In describing some uses of the Cambridge Structural Database (CSD) in research and teaching in a recent *IUCr Newsletter* (Clegg, 2020), I mentioned the general agreement that this and other crystallographic databases undoubtedly contain only a minority of crystal structures that have been successfully solved and refined.

The 2010 British Crystallographic Association (BCA) Spring Meeting included a session devoted to issues of publication of small-molecule crystal structures. As part of that session, I gave an invited lecture with the title “Reasons good and bad for not publishing crystal structures,” which was followed by a lively discussion. This article is adapted from the material of that lecture, with appropriate updates. Much has changed in ten years!

The continued development of high-throughput equipment for diffraction measurements, of ever more brilliant X-ray sources, and of sophisticated and increasingly automatic software for structure solution and refinement in the intervening years has surely added enormously to the wealth of unpublished results of good technical quality. At the same time, opportunities and facilities for formal publication have seen no major advances on a comparable scale, and reasons for not publishing have probably multiplied through competing pressures on time and other resources.

Here I pose and seek to address a number of related questions: Why publish at all? Why *do* structures not get published? Why *should* structures not get published? What do we mean by publication anyway?

The views expressed are, of course, personal and are based on decades of experience in a rapidly changing environment; they are intended, as at the BCA meeting, to provoke reaction and discussion.

## Why publish at all?

It is right to ask such fundamental questions of motive, especially when publication significantly consumes time and other resources. Several positive answers can readily be given.

1. **To justify public funding:** in recent years this has become a definite formal policy of research funders, who often ask for at least outline publication plans when they are considering grant applications, combined with a requirement of some kind of open-access status of resulting publications, for which an earmarked proportion of the funding may be provided. It should also be considered a moral imperative – public finance demands public access to the results of the research.
2. **To further careers:** this raises the follow-up question, whose careers? Research usually involves both established personnel and those who are at an early stage or still in formal training, and publication, preferably with high profile and impact, is important at all stages of career development.
3. **For reputation:** not only personal but also institutional. There is no doubt that publication approaches and practices are strongly influenced by official research assessment exercises, by all sorts of metrics, and by overused performance league tables.
4. **To contribute to knowledge:** this is, of course, the fundamental purpose of research in general. Clearly, some knowledge is more useful and more important than others, but there is a good argument in crystallography for extending our knowledge and understanding of structures as widely as possible to enable the recognition of patterns and notable exceptions to them. The failure to bring “less interesting” results into the public domain is a form of bias in the available complete set of data.
5. **To avoid unnecessary duplicate work:** there must be many structures that have not been published because they were not the expected or desired result. In some cases, these are recovered starting materials, unwanted by-products, or the products of “failed” reactions, but nevertheless are previously unreported structures. Having these available, at least in an accessible database if not in a journal publication, can save time and effort by other researchers if a matching unit cell is recognised in the first stages of a data collection.

Worthy as these motives are, they are not sufficiently strong to prevent a growing mountain of unpublished structures accumulated in research laboratories across the world. The factors militating against publication are many and forceful.

## Why do structures not get published?

Here are a number of reasons – and excuses – that might be given in response. Their validity is itself a matter for debate.

1. **There are too many results to be able to publish them all.** In the days of photographic data collection and then of serial diffractometers measuring one reflection at a time, when it took days or weeks to determine a crystal structure, it was possible for publication to keep up with the output of results, but it began to fall behind as data collection (Clegg, 1981) and computing facilities became faster. With the widespread introduction of area detectors for routine use in the 1990s, the flood of new structures overwhelmed the channel of publications, and large reservoirs of results began to build up. Now we have various criteria and priorities to decide which structures make it through to publication and which are left behind, many of them not based on the intrinsic crystallographic merit of the individual structures.
2. **There is too much time and effort involved in publication.** It can certainly take far longer to generate a manuscript, or even just the crystallographic component of one, than to carry out a complete

structure determination. This is true despite the enormous electronic advances in publishing in recent decades, significantly aided by the virtually universal adoption of CIF as an archiving and dissemination medium for crystal structures alongside powerful validation, analysis and graphical tools.

**3. Publication is not my responsibility.** This depends on the status of crystallographers and the terms and conditions under which they operate, ranging from independent research leaders in their own right to managers and staff of an externally funded technical service. It is tied up with potentially contentious issues, such as the question of who is the primary “owner” (in terms of intellectual property rights and of accountability) of a crystal structure as a piece of information. Who actually chooses whether, and how, and when and where, any particular structure is submitted for publication?

**4. Other people involved (chemists, research group leaders) have moved, retired or died.** Is relevant information and expertise to generate a suitable publication still available? This may be combined with or have similar impacts to the next two reasons.

**5. Funding for this work has ended.**

**6. We are no longer interested in this topic.** Priorities, funding opportunities and fashions do change in scientific research, for both good and bad reasons. In some cases, a series of crystal structures may be generated as part of a mainly synthetic PhD project, a primary aim being to obtain definitive confirmation of the chemical identity of the compounds or some specific structural feature such as conformation or absolute configuration. Once the results are included in a successfully defended thesis, and the student takes up another post, this particular project is left behind, and any good intention to convert the results into publications may get buried in other priorities and new projects.

**7. The result is not interesting, or not what we wanted.** In particular, it may just not be important enough for possible inclusion in performance-related assessments, such as in the UK Research Excellence Framework or similar exercises elsewhere. Unexpected results can, of course, turn out to be particularly exciting and lead to publications in high-profile journals, but many are regarded as disappointing and even a waste of time and resources. As one example of this contrast, my own involvement in characterising polyoxometalates has revealed some unexpected condensed products with structural complexity (Errington *et al.*, 2007). But reactions in this field can often lead to the thermodynamic sink of the  $[M_6O_{19}]^{2-}$  cluster anion ( $M = \text{Mo}, \text{W}$ ; Fig. 1). We have encountered many salts of these anions with various organic cations, including numerous polymorphs and different solvates, only some of which have previously been reported. These structures are never going to be considered important enough for a publication.

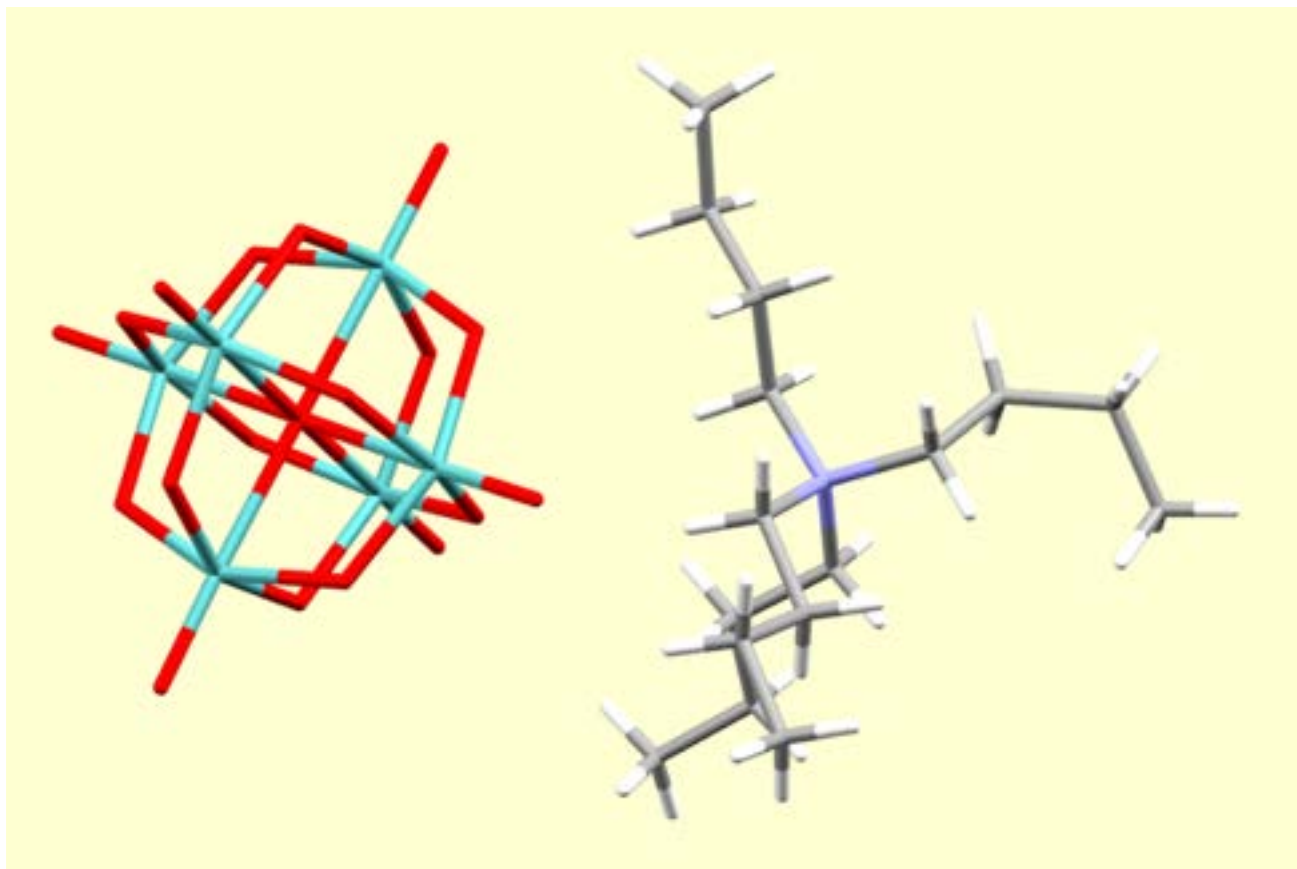


Figure 1. The anion (on a twofold rotation axis) and cation of one unpublished polymorph of  $(n\text{-Bu}_4\text{N})_2[\text{Mo}_6\text{O}_{19}]$ .

**8. There are further experiments to be done for a full publication.** This may be combined with or be an alternative version of the next one.

**9. We do not wish to reveal these results (yet, if ever).** Avoiding a “salami-slicing” approach to the publication of individual results that would be much better in a combined authoritative account of an entire project is commendable; but I have known cases in which the “further experiments” take many years and are never successfully completed, or are simply overtaken by new priorities. A more acceptable reason for delay is the preparation and filing of a patent for which the structure is part of the evidence, so its premature appearance in the public domain must be avoided. Most crystallographic work carried out in the chemical or pharmaceutical industry is never published; outsourcing of commercial work to other structural service providers is usually covered by non-disclosure agreements, with the full intellectual property rights explicitly assigned to the funders.

**10. The result is not of good quality (at least, not yet).** Crystal structures vary considerably in their overall quality, precision and reliability. Some of the factors that can adversely affect the result include very small crystal size, poor crystal quality, disorder, twinning and radiation damage. In some cases, improvements can be achieved through recrystallisation, use of a more intense X-ray source and/or a more sensitive detector, or the preparation of a different derivative or solvate, among other approaches. As an example, in a study of the impact of different substituents on the cage geometry of the *ortho*-dicarbaborane  $\text{C}_2\text{B}_{10}\text{H}_{12}$ , especially the effect on bond lengths, varying the substituent at one C atom while leaving the other C-H unsubstituted leads to unresolvable disorder of the C-H and four B-H positions; the result is a statistically averaged geometry with no useful outcome. Attachment of a methyl or (better) phenyl group to the second C atom prevents this disorder and gives clear results for a series of substituents at the first C atom (Fig. 2), with a well-defined substantial effect on the C-C bond length (Coult *et al.*, 1992; Boyd *et al.*, 2004; Fox, MacBride *et al.*, 2009; Fox, Peace *et al.*, 2009).

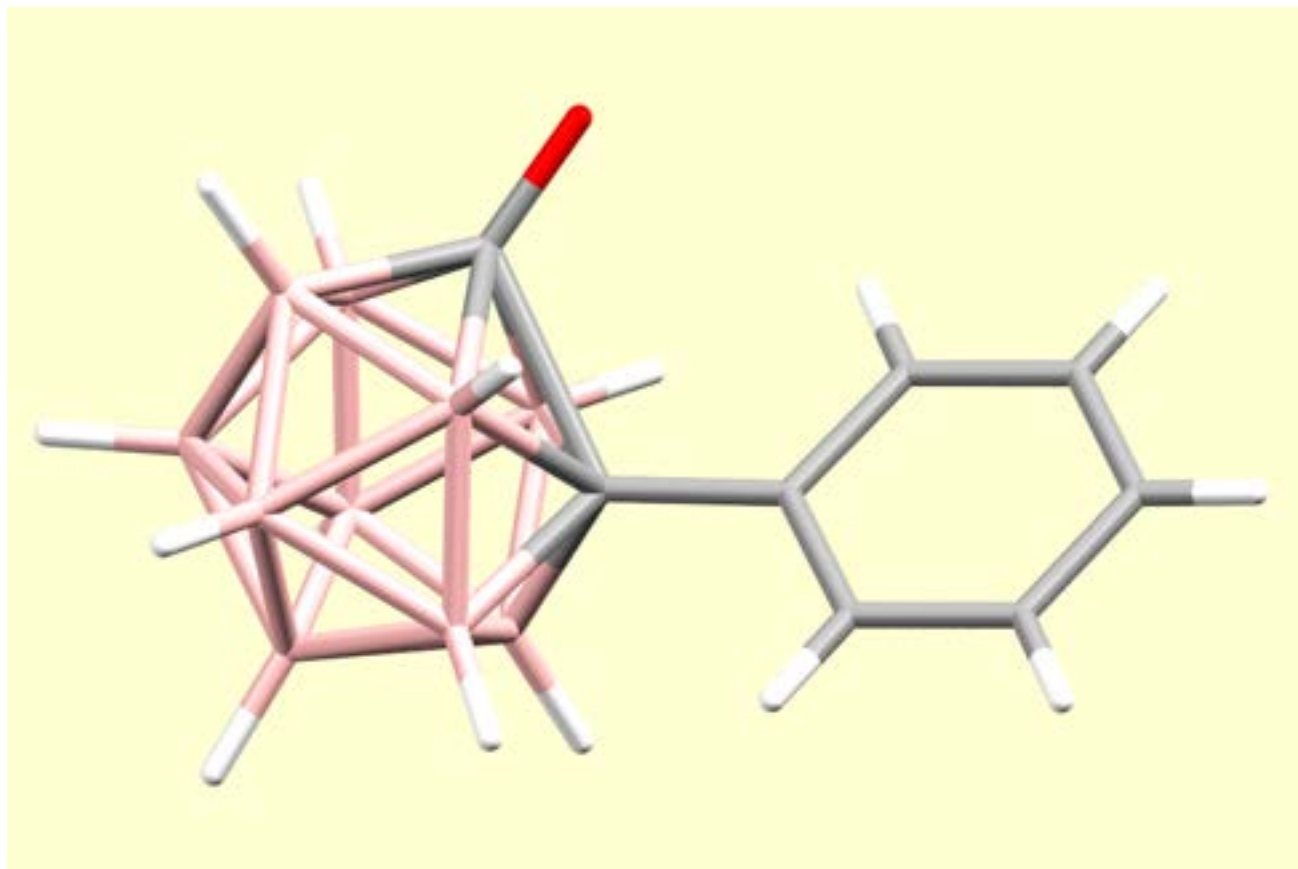


Figure 2. The anion  $o\text{-PhCB}_{10}\text{H}_{10}\text{CO}^-$  with an exceptionally long C-C bond (from Boyd *et al.*, 2004).

11. **We tried, but the paper was rejected.** This rejection may not be because of the crystallography component, though some chemical journals do have a regrettable attitude that denigrates a manuscript judged to be “only crystallography”. Sometimes criticism is made by identifiably crystallographic referees. Constructive criticism is, of course, the proper responsibility of referees, and it often leads to improvement and correction. However, I take issue with what I categorise as three particular types of poor crystallographic referees:

- (a) **the sloppy**, whose review of the manuscript appears to be superficial, offering no significant comment and, in some cases, failing to notice obvious faults that consequently appear in publications;
- (b) **the robotic**, who merely cite *PLATON/checkCIF* alerts (Spek, 2020) and incorrectly call these “errors” that need to be corrected, basing a recommendation of rejection purely on these with no consideration of their possible causes or author responses to them;
- (c) **the opinionated**, who seem to consider that their personally preferred approach to the palette of available refinement tools (particularly the use of restraints) is the only acceptable one and who demand a revision carried out in their particular way.

## Why should structures not get published?

While there are journal referees who do a poor job, most are surely conscientious and impartial. Their role is essential to ensure the accurate and reliable reporting of valuable structural results. They, and editors, serve as a defence against the publication of unsuitable results. Crystallographers themselves, however, have the primary responsibility of judging their own results, as to how appropriate they are for release in the public domain, and in what way, for use by others. Some structures should simply not be published, as they do not serve the purposes outlined in the first section of this article. What features can make a structure unsuitable for publication?

1. **It is of poor quality.** Quality is a relative property on a continuous scale, not a binary characteristic, so this requires judgement against relevant criteria. It depends on the purpose of the experiment in the first place, and on the way in which the results are to be used in the context of the overall work. The “quality” of a structure may also not be uniform in all its features. For example, a badly disordered solvent molecule or counterion that has not been well modelled, despite best attempts, does not usually detract from the value of the main part of the structure (though it does generally mean a reduced precision overall). On the other hand, a poorly defined H atom could be a key feature of the structure and severely limit important conclusions about the chemistry.

2. **It is not fit for purpose.** What is the point of the experimentally determined structure? A lower quality can be tolerated for a result intended to confirm the identity of a product from among a number of possibilities, provided the answer is unambiguous, than for a study of detailed molecular geometry in which numerical standard uncertainties need to be minimised if sensible conclusions are to be drawn. Whatever the purpose, however, there is no excuse for sloppy work; the goal should always be the best possible structural result, with all stages of experiment and computation geared to this.

3. **It is incorrect.** Various experimental failings, lack of appropriate corrections for known systematic errors and effects, and poor use of software can lead to inappropriate structural models and misinterpretation of the results. Possible errors include wrongly assigned atom types, missing or misplaced H atoms, and a failure to recognise symmetry-related connections in a polymeric structure. I was particularly amused, while also being horrified, by one example from my work as a founding Section Editor of *Acta Crystallographica Section E* in the early 2000s. It would be unethical to give details, but it became clear to me on close inspection that the authors had incorrectly identified F atoms as OH groups and a nitro group as a carboxylic acid (Fig. 3), leading to some unacceptable geometry for supposed hydrogen bonds; correct atom assignments resulted in a structure with markedly lower refinement residuals that was actually already known. Correspondence with the principal author, who would not accept these findings, was protracted until I recommended that he submit the paper to *Nature*, because the reported structure, if correct, would be unusual enough to make the front cover! I preserve to this day the comment sent to me that “I fear you are suffering from a disillumination of the mind”!

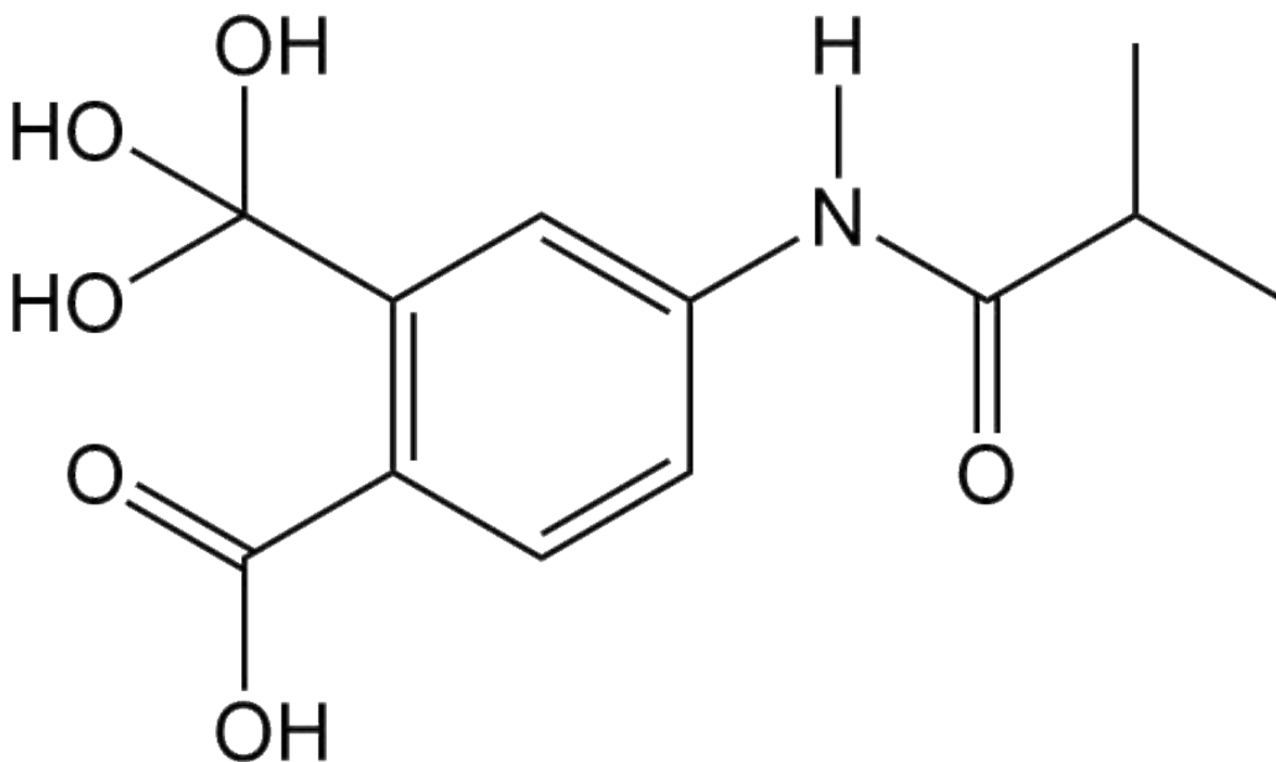


Figure 3. A proposed but manifestly incorrect chemical structure.

4. **It is fraudulent.** Fortunately, as far as we are aware, this is rare in reported crystal structures. Sadly, there was a spate of fraudulent structures based on manipulated diffraction data in *Acta Crystallographica Section E* around 2007 (Harrison *et al.*, 2010), perpetrated by a small number of dishonest authors. It was uncovered by careful use of validation software, which has subsequently been enhanced to provide better detection of such fraud and other problems and is routinely used by IUCr journals and many others. Several other science journals, some very prestigious, were prompted by the up-front honesty of the *Acta* editors to investigate, and found serious fraud in some of their own publications. The nature of crystal structure results and their relationship to the original experimental data (deposition of which is demanded by IUCr journals) makes this kind of abuse much harder to cover up than in many other areas of science, where deliberate fraud can be difficult to detect.

## What do you mean by publication anyway?

The word “publication” covers a range of ways in which scientific research results are placed in the public domain. The conventional publication used to be, principally, a physical manuscript submitted to, peer-reviewed for, and accepted by a publisher for inclusion in a printed journal issue (or a book), with a recognised literature reference. It could also be used for articles published without a peer-review step. The term has been broadened over the years by the appearance of hybrid print/electronic and electronic-only journals and by the distinction between subscription-paid and open-access publication, with various levels of “open access” having a range of embargo and release stages. Other forms of presence in the public domain, with different degrees of effective accessibility, include conference presentations and abstracts, doctoral theses usually lodged in an institutional library, institutional and organisational electronic document repositories, databases that may be freely accessible or available on a subscription basis, and personal or corporate websites.

Important designations and properties that have been introduced over the years for publications, associated with different perceived degrees of status and value, include acceptance into indexing

services such as [Chemical Abstracts](#) (a division of the American Chemical Society) or [Web of Science](#) (Clarivate Analytics PLC, formerly *ISI Web of Knowledge*), and the assignment of Digital Object Identifiers (DOI). On the basis of these and other criteria such as journal impact factors, different kinds of publication, and different publications within these kinds, are generally considered to have different degrees of worth in uses such as personal CVs and institutional research assessment submissions.

Whatever form of publication we choose, the dissemination of our crystal structure results is an important part of the research work overall. What, may we ask, is the point of doing research at all unless we show and explain to others what we have done?

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## MEETING REPORT (IUCR SUPPORTED)

# To.Sca.Lake 3.0: Total Scattering for Nanotechnology 2019

Norberto Masciocchi



To.Sca.Lake 3.0: Total Scattering for Nanotechnology on the Como Lake was the third edition of a biennial series of Summer Schools that are organized within the initiatives of the Lake Como School of Advanced Studies and are fully dedicated to the foremost characterization methods, based on scattering techniques (from X-rays to visible light), developed for the multiple (atomic and nanometre) scale investigation of engineered nanomaterials.

The Workshop, which took place in the Villa del Grumello (Como, Italy; pictured above), lasted five days (27–31 May 2019) and included lessons, demos and *hands-on* tutorials. It was attended by 55 young researchers, internationally renowned teachers and commercial sponsors, all participating in a very active manner in the different sessions. The wide international attendance, with students from four different continents, made for a rich cultural and scientific exchange.

Foreign teachers (Hans Beat Bürgi, Simon Billinge, Jan Skov Pedersen, Antonio Cervellino, Martin Schmidt, Arnt Kern, Maksym Kovalenko and Detlef Beckers) and Italian experts in scattering techniques (Norberto Masciocchi, Antonella Guagliardi, Ruggero Frison and Fabio Ferri) presented, within the first three days, experimental techniques and high-level scientific results, including both theoretical and experimental aspects [small-angle X-ray scattering, total scattering in direct (PDF) and reciprocal (DSE) spaces, and static and dynamic light scattering]. The help of young researchers (Federica Bertolotti and Maxwell Terban) enabled the set up of a few parallel sessions of data analysis with *PDFGui*, *DebUsSy* and

*WLSQ\_SAXS*, modeling computer programs developed in recent years by some of the speakers. A few hours were also dedicated to oral contributions by young participants, and a permanent poster session was organized and duly attended. Finally, in the last day, Professor Ferri organized a live-demo experiment with the dynamic light scattering technique on colloidal suspensions of Pb(II)S, CdSe and CsPbBr<sub>3</sub>, highly performing materials in a variety of technological fields.

Organizing this Workshop was possible thanks to the funds raised from registration fees, funds provided by the Dipartimento di Scienza e Alta Tecnologia of the Università dell'Insubria and by the Lake Como School, sponsorship from national and international scientific bodies (IUCr, ICDD, AIC, ECA and INSTM) and commercial sponsors. The financial support generously provided by the IUCr was used to support the participation of five young attendees (Marcelo Malagutti, Brazil; Afshin Pendashteh, Iran; Kamal, India; Jelena Bjielic, Croatia; and Martin Alonso Garro Gonzàles, Costa Rica).

## MEETING REPORT (IUCR SUPPORTED)

# ECM32 Satellite Meeting: Mathematical and Theoretical Crystallographic Workshop

Massimo Nespolo Bernd Souvignier Berthold Stöger



Ahead of the ECM32 meeting, hosted by the University of Vienna, Austria, numerous satellite workshops were held at the TU (Technical University) Wien. Most took place in the electrical engineering building with shared coffee and lunch breaks. Within this scope, the IUCr Commission on Mathematical and Theoretical Crystallography (MaThCryst) organized a 2.5 day satellite workshop, from 17 to 19 August 2020, with a special focus on graph theory and modular structures.

Fifteen participants from diverse backgrounds (chemistry, mathematics and crystallography) attended the workshop. The participants hailed from six countries: Austria, Germany, Israel, Japan, Russia and the UK.

On the first day, fundamentals of graph theory and their applications to crystal structure analysis were presented by Bernd Souvignier (Radboud University Nijmegen, Netherlands). Special focus was placed on the analysis of periodic nets. These are crucial to describe the connectivity of atoms or groups in crystalline materials. In the morning session of the second day, Massimo Nespolo (Université de Lorraine, Nancy, France) introduced the application of space groupoids to describe modular structures in a general way. In modular structures, which are ubiquitous in all classes of materials, operations are applied to subspaces of crystal space. These substructures can be non-periodic blocks or sub-periodic units (rods or layers). As the operations relating two modules need not apply to the whole crystal, their composition forms a groupoid instead of a group. In the afternoon Berthold Stöger (Technical University Vienna,

Austria) focused on the concrete case of order-disorder (OD) structures. These are modular structures composed of layers that are arranged in such a way that there is only one geometrical way of placing adjacent layers. Thus, an infinity of different stacking arrangements, which are nevertheless locally equivalent, can exist, leading to challenging problems such as twinning or disorder. The sessions on graph theory and modular structures comprised approximately two thirds lecture and one third exercises, including discussion.



In the morning session of the last day, the session on OD structures was completed and two oral talks of ca 30 minutes were given by two attendees. Shelomo I. Ben-Abraham (Beer-Sheba, Israel) presented *Multidimensional crossed cube tilings*. Vitaliy Kurlin (Liverpool, UK) in his talk *How to correctly sample unit cells in computer simulations of crystal structures* proposed using Wigner-Seitz cells instead of reduced cells to provide a unique and continuous mapping of lattice metrics. The talks resulted in a lively discussion. The workshop closed during lunch of 19 August with a poster presentation by three attendees.

On the evening of the second day the participants of all workshops met at a typical Viennese “Heuriger” to enjoy local food and wine. This social evening provided a unique networking opportunity.

Four young participants (two male and two female) from three countries (UK, Germany and Russia) applied for travel assistance, which was generously provided by the IUCr.

The feedback was generally positive. Unifying participants from such diverse backgrounds was challenging, but ultimately satisfying. The organizers and participants published six papers in the [May 2020 special issue of \*Crystal Research and Technology\* on \*Mathematical Crystallography 2019\*](#). Owing to the positive experience, the organizers plan to repeat workshops on mathematical and theoretical crystallography at upcoming ECMs.

## MEETING REPORT (IUCR SUPPORTED)

# 17th European Conference on Solid State Chemistry

Olivier Mentré



The European Conference on Solid State Chemistry (ECSSC) is a biennial high-level international conference with a long history, ECSSC1 having taken place in Strasbourg, France, in 1978. It is dedicated to all aspects of the synthesis, design and evolution of solid functional materials with extended structures and interesting chemical or physical properties.

**ECSSC17** took place from 1 to 4 September 2019 at the MACC of the University of Lille, France. It was organized by a local committee composed of members belonging to the FST, the University of Artois, the University of Picardy Jules Vernes, the ENSCL and the CNRS in partnership with the Solid State Chemistry Division of EUCHEMS (European Chemical Society) and the Solid State Chemistry Division of the SCF (Société Française de Chimie). This conference brought together around 230 participants from about 30 European, Asian, African and American countries. In addition, the scientific program included 12 invited speakers, 12 keynote speakers and a plenary talk by [Robert Cava](#) (Princeton University, NJ, USA) on "New Materials with Interesting Magnetic and Electronic Properties".



The opening session was dedicated to the [International Year of the Periodic Table 2019](#) with specific related talks. The main topics of the conference were Solid State Chemistry, Materials, Energy and Information Storage with the following themes:

- Functional Materials and Design
- Nanomaterials and Thin Films
- Hybrids, Micro/Mesoporous Materials
- Advances in Solid State Materials Synthesis/Materials Design
- Crystalline and Disordered Compounds, Glasses
- Nitride, Chalcogenides and Mixed Anion Compounds
- Electrochemistry, Energy
- Quantum Materials
- Advanced Characterization Techniques
- Mixed Solid State Chemistry

This was a specialized conference, a place for communication and exchange of innovative results between world experts in the field of solid-state chemistry and materials. Students and researchers found information and contacts to renew their research and launch collaborations. In addition, the participating exhibitors provided a clear vision of the technological advances of the dedicated equipment. The IUCr supported the attendance of students from Germany (2), Russia (2), Spain (1), Sweden (1) and UK (4).

## MEETING REPORT (IUCR SUPPORTED)

# Fourth International Workshop on X-ray Crystallography in Structural Biology

Maqsood Ahmed

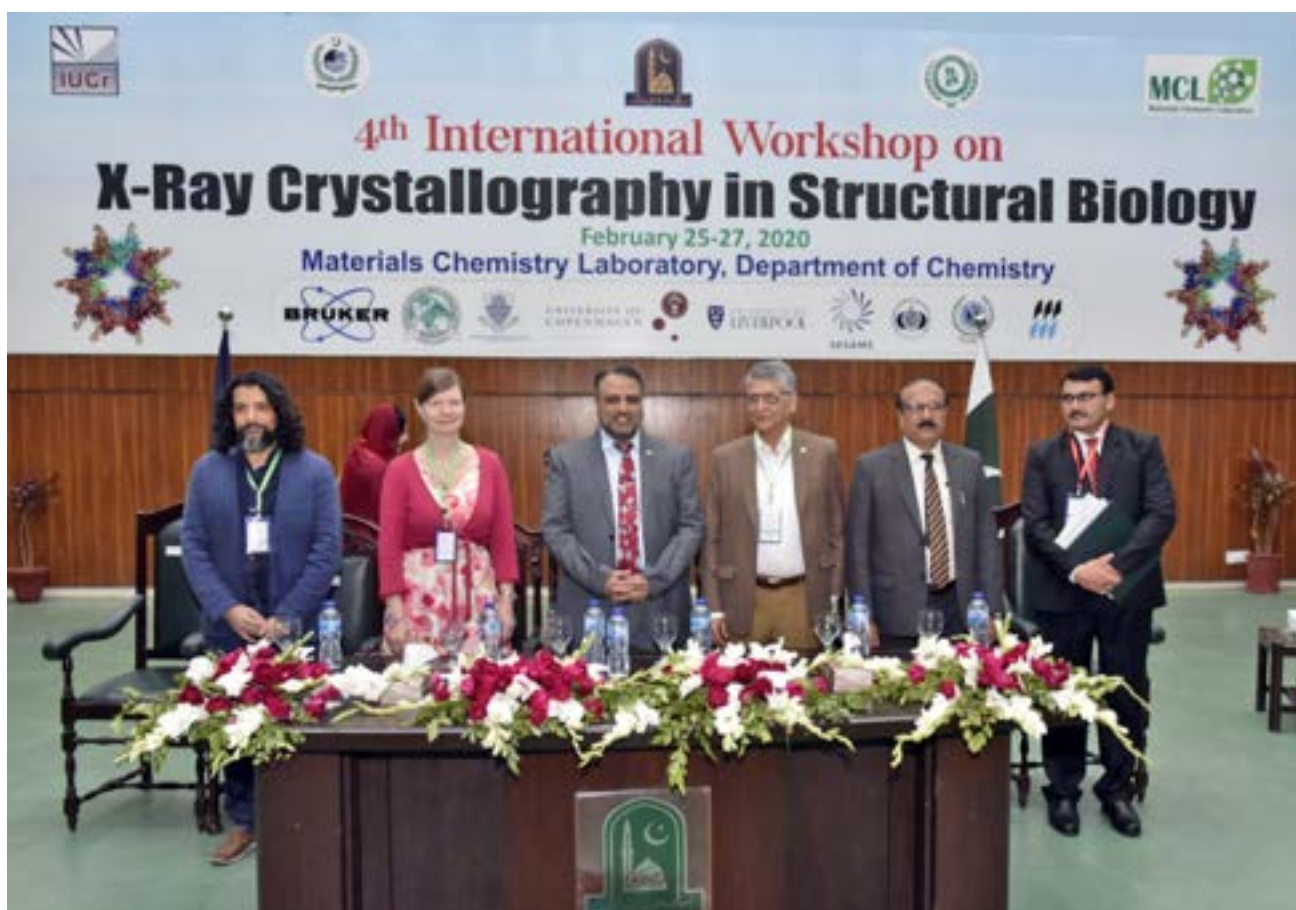


The Fourth International Workshop on X-ray Crystallography in Structural Biology was held at the Islamia University of Bahawalpur, Pakistan, from 25 to 27 February 2020. The first meeting in this series was held at the National Institute for Biotechnology and Genetic Engineering (NIBGE), Faisalabad, in November 2015; the second at FC College University Lahore in October 2016; and the third at the International Centre for Chemical and Biological Sciences (ICCBS), University of Karachi, in 2018. The aim of these workshops is to build capacity in the area of structural biology, which has had a very late start in Pakistan but is now gaining ground. As with the previous meetings, the fourth workshop was sponsored by the IUCr. Additional financial support was provided by the Pakistan Science Foundation (PSF), the Higher Education Commission of Pakistan. The Islamia University of Bahawalpur provided complete logistical support. Bruker Corporation and Douglas Instruments provided significant sponsorship.



Group photo.

Bahawalpur was chosen as the venue for the fourth workshop for multiple reasons. It has a unique landscape with the Cholistan Desert on one side and agricultural fields on the other. The city is famous for its palaces, which were built by the Nawabs of Bahawalpur. It is located almost in the centre between Karachi and Islamabad. The city has three universities. It is easily accessible from other large cities like Multan, Lahore and Faisalabad through a network of motorways and also has its own airport. And above all, the Islamia University of Bahawalpur has recently established a Materials Chemistry Laboratory that hosts a dual-source single-crystal X-ray diffractometer and a powder X-ray diffractometer. This lab is now regularly publishing research on small molecules and seriously wanted to undertake structural biology.



Opening ceremony.

To provide high-quality hands-on training with a high student-to-teacher ratio, renowned structural biologists from abroad and from within Pakistan were invited. Samar Hasnain (University of Liverpool UK), Elspeth Garman (University of Oxford, UK), Richard Garratt (University of Sao Paulo, Brazil; sponsored by the IUCr) and Osman Asghar Mirza (University of Copenhagen, Denmark) carefully designed courses over the span of three days. They were assisted by national experts.

There were around 70 participants of which more than half were female. The participants were mainly Master's or PhD students. The IUCr sponsored full economy class travel and lodging for 3 nights for 40 young scientists from across Pakistan. This support from the IUCr to young scientists proved crucial for the success of the workshop as the most serious participants from the disciplines of chemistry, physics, biotechnology, medicine and agriculture were able to participate.

Each day started with one or two comprehensive lectures. In his plenary lectures, Professor Hasnain gave an extensive overview of hot topics in structural biology, which the audience found very inspiring, and Professor Garratt nicely explained the basic concepts of symmetry and diffraction. The participants were divided into three groups and parallel sessions were organized. The participants received hands-on training in all aspects of protein structure determination including expression, purification, crystallization and data collection on the newly installed Bruker D8 Venture diffractometer equipped with an Oxford Cryosystems Cobra cryosystem. Moaz-ur-Rehman, Ahmed Akram, Muhammad Imran and Amir Shehzad conducted tutorials on expression and purification of proteins. Professor Garman and Maqsood Ahmed provided extensive hands-on training on harvesting the crystals under an Olympus SZX10 polarizing microscope equipped with a high-resolution DP74 camera. The students learnt how to mount the crystals on the goniometer head and visualize the diffraction. Lengthy sessions on data processing and model building using *CCP4* and *COOT* were organized on the second and third days. Professor Garman prepared the course material on *CCP4*, and Dr Mirza, Malik Muhammad Shoaib and Saima Rasheed conducted the tutorials on refinement and model building.



Hands-on training. Left: protein expression and purification; right: crystal hunting and the single-crystal X-ray diffractometer.

On the final day of the workshop, the participants were required to take a written examination, which focused on the activities of the three days. Most of the participants fared very well. Prizes provided by Bruker were distributed among the top five scorers.

Each day the participants were served with lunch and dinner. An elegant gala dinner was hosted on the evening of the second day on the lawns of the beautiful Noor Mahal, where the participants admired the light show.

The opening and closing ceremonies were presided over by the Vice Chancellor of the Islamia University of Bahawalpur who took a keen interest in the event and offered his full support to efforts to promote structural biology in Bahawalpur.

A one-day sightseeing trip was organized on the next day of the workshop. The participants visited the historical Sadiq Garh Palace and the Derawar Fort in the Cholistan Desert. The participants also had a guided tour of the Biodiversity Park of the Islamia University of Bahawalpur situated in the Cholistan Desert.



Elspeth Garman and Richard Garratt enjoying a camel ride at Derawar Fort.

The event gained extensive coverage in newspapers and [on television](#). Additional photos can be seen [here](#).

## MEETING REPORT

# A scientific student conference at Novosibirsk State University: interdisciplinary, international, online

Anna Gaydamaka Adam Michalchuk Elena Boldyreva

zoom

МНСК-2020: ФизМетоды и ХТТ, 14-16 апреля · Shared screen with speaker view

**Материалы и методы**

**Природные полисахариды:**

**Хитозан**  
буфер: pH 5,4  
Ионная сила 0,15M

**Сульфатированный арабиногалактан**

**Анионный краситель:**

**Эозин**

**Используемые методы:**

- Распределение частиц по размерам и заряда-потенциал: метод динамического рассеяния света
- метод электрофоретического рассеяния света (Zetasizer Nano ZS; Malvern Instruments Ltd., Великобритания).
- SEM-изображения частиц: сканирующая электронная микроскопия (S-5500; Hitachi, Япония).

Science has always surpassed borders, and those imposed by the global COVID-19 lockdown are no different. Around the world, science continues, with conferences and meetings transforming to digital platforms. Inspired by these global actions, a session on "Physical Methods in Solid State Chemistry" of the [58th International Scientific Students Conference \(ISSC\)](#) was carried out as a live digital conference from 10 to 13 April 2020. This conference was hosted by Novosibirsk State University (NSU), Russia.

Breaking decades of tradition, the NSU Chairs of "Physical Methods in Natural Sciences and Materials Sciences" (Department of Physics) and of "Solid State Chemistry" (Department of Natural Sciences) decided to host a joint symposium at the ISSC. This tradition-breaking decision reflects the early success of our new interdepartmental MSc program "[Advanced Techniques in Solid State Sciences](#)", which actively cultivates interdisciplinary discussions between chemists, physicists, geologists and materials scientists. The enthusiastic discussions during the joint ISSC session were a clear testament to the importance and strengths of such a program.

The virtual, interdisciplinary session of the ISSC comprised a total of 33 speakers. This included talks from NSU, Novosibirsk State Technical University, Tomsk State University, Tomsk Polytechnic University, Siberian Federal University (Krasnoyarsk), S. Amanzholov East Kazakhstan State University (Kazakhstan)

and even the University of Oxford (UK). NSU was represented by students from across three departments: Physics, Natural Sciences, and Mechanics and Mathematics. Speakers came from a range of levels, starting from first-year BSc students to final-year MSc students. Moreover, NSU MSc students from India and China also participated in the conference. Additional to the impressive list of speakers, conference participants tuned in from across Russia and Europe. Registered participants included those from eight scientific institutes of the Siberian Branch of the Russian Academy of Sciences [1], nanomaterials manufacturer OCSiAl (Novosibirsk), the Kurchatov Institute (Moscow) and members from the ESRF (Grenoble, France).

Demonstrating the strength and reach obtainable through virtual conferences, a quarter of the participants at this ISSC session were not actually present in Novosibirsk during the conference. These participants "attended" the conference without moving from their home city.

The session's expert panel comprised a highly multidisciplinary team of researchers ranging from mathematicians, physicists and chemists to geologists and mineralogists. This breadth of expertise on the panel was truly necessary, owing to the exceptional level of talks presented by all speakers across a significant range of disciplines. Discussion between participants (speakers, panelists and attendees) was outstanding. Frequently, questions had to be cut short to allow progression of the session. The best speakers were awarded prizes.

Many talks were associated with the use of synchrotron radiation (SR). This included discussion on the development of new elements for the generation of SR, X-ray lenses and new sample holder designs. Presentations regarding new approaches for synchrotron data reduction were also given. Participants heard much about the significant strengths of SR for investigating the structure, properties and synthesis of compounds, minerals, and composite and low-dimensional nanomaterials. In addition, and complimentary to SR, the significant role of other analytical methods in investigating the solid state was made clear. This included discussion of X-ray diffraction and spectroscopy, solid-state NMR, electron microscopy, EELS, electrochemical methods and computational methods (molecular dynamics).

As aptly highlighted by one of the session speakers, physicists actively develop instruments that chemists use, while chemists set the problems for physicists, providing them with objects for studies. This interrelation epitomizes the intertwining nature of the sciences and highlights the importance of interdisciplinary interactions as often and early in scientific careers as possible. One can expand this idea – physicists and mathematicians develop techniques, hardware and software that is further used by, for example, chemists, materials scientists, geologists and biologists. This interaction was very visible during the conference. We plan to use a similar format in addition to the traditional one in the future to increase the geographical spread of participants and nonresident plenary speakers.

zoom

МНСК-2020: ФизМетоды и ХТТ, 14-16 апреля - Shared screen with speaker view

Department of Physics

UNIVERSITY OF OXFORD

Introduction

Crystalline structure of inorganic-organic hybrid lead perovskite  
(D. Li, et al.: *RSC Adv.*, 2016, **6**, 89356-89366)

Nanowire laser.  
Scale bar 2  $\mu\text{m}$   
(S. Eaton, et. al.: *PNAS*, 2016, 1600789113)

Perovskite solar cell (Saulle Technologies)

2

## Feedback from speakers, experts and other participants

### Sveta Scharaya (second-year Bachelor student, Department of Natural Sciences, NSU)

I was very happy to participate in this conference as an attendee. Despite the online format, there was a friendly working climate. I liked mostly the talk about the design of novel crown esters.

### Maksim Sukhov (fourth-year student, Department of Natural Sciences, NSU, Vorozhtsov Institute of Organic Chemistry; Winner Third Place Oral Presentation Prize)

I was thrilled that, despite all, the conference went ahead in its innovative design. The talks were very interesting, the atmosphere was friendly, everything was well organized. Of course, I am happy to have won a prize recognizing my efforts and commitment to my work!

### Amit Kumar Pal (first year, Department of Natural Sciences, Master program “Materials Science”, Lavrentiev Institute of Hydrodynamics)

My participation in ISSC-2020 was a great experience. Using the platform of this conference, I came to know about new scientific research performed by young researchers and scientists. The conference allowed me to interact and discuss my research work with fellow participants. It helped to improve my communication and presentation skills. I appreciate the valuable comments from colleagues and experts and will use their insights to direct my future research work.

**Maria Atamanova [second-year Master student, Institute of Engineering Physics and Radioelectronics, Siberian Federal University (Krasnoyarsk); Winner Second Place Oral Presentation Prize]**

I would like to thank the organizing committee for the conference. The level of organization pleasantly surprised me. I was anxious about some technical points during discussions because of the online format, but everything was great! The explanations provided about the online format and connection were easy to understand. As a speaker, I just had to worry about my talk, not about technical questions. I have only positive feelings about the conference. I was very happy to attend it and hope to meet everybody in the future, in person.

**Andrey Trebushinin (first year of Master program “Physical methods for the condensed state studies”, Physics Department, NSU, Budker Institute of Nuclear Physics; Winner Second Place Oral Presentation Prize)**

I am working on optimizing the parameters of X-ray optics for a new SR facility “SKIF”, which is being constructed in Novosibirsk. I worried about the topic of my talk. I was not sure if I could explain my work to chemists, geologists and materials scientists. Other students presented the results of the synthesis of new nanomaterials and of structural studies by X-ray diffraction and spectroscopy. Everything presented by these speakers differed from my own presentation: topics, approaches and even terminology. But then I recognized that these researchers are my future colleagues at SKIF. I am developing the very machine that they will use, and therefore recognized the importance of making my talk understandable for a wider audience. I was amazed by the relevance of the questions I was asked after my talk and the high professional level of discussion. These discussions were very valuable for my work, both with respect to our designing of the efficiency of SR generation and to the optimization of the conditions available for future researchers. I thank the organizers, speakers and the audience for such a pleasant scientific discussion. I am looking forward to meeting everyone again next year, in a face-to-face format!

**Nikolay Kahidze (first year of Master program, Physical-technical Department of Tomsk State University; Winner Third Place Oral Presentation Prize)**

I am very glad that the conference went ahead and was carried out very successfully. I would like to emphasize the high level of the presentations as well as the expertise of the organizers and coordinators. I also got useful experience in online presentations and learnt how to use Zoom. I think next time it would be great to motivate participants to switch on their web-camera. Many thanks again!

**Elena Boldyreva (professor of the Chair of Solid State Chemistry, Department of Natural Sciences of NSU, Boreskov Institute of Catalysis; co-chair of the conference)**

I was very happy to see my long-term dream come true – we could organize this student conference not as a local event of the single chair like a defense repetition, but as a true scientific meeting in which experts from various fields come together and discuss the latest scientific achievements. It was pleasant to see that our efforts to harmonize education combining the three main modules (“Solid-state structure and properties – Solid-state transformations and practical implementation – Physical methods in solid-state research”) have started to yield fruit. Many students have already worked at SR facilities either in Russia (Budker Institute of Nuclear Physics, Novosibirsk; Kurchatov Institute, Moscow) or abroad (ESRF, BESSY, Petra, DESY), and also at the European XFEL.

**Sergey Tsybulya (professor, Head of the Chair of Physical Methods of Investigation of Solid State, Physics Department, NSU, Boreskov Institute of Catalysis; co-chair of the conference)**

Chemistry, mineralogy and materials sciences suggest problems for physicists to solve. The solution of these problems requires the development of new experimental and mathematical instruments. Students should see the possibility of applying their skills and knowledge in other research fields from the very beginning of their education. This should motivate them in their studies. This is extremely important. The joint session has shown that this is today a reality. Not only students but also the expert panel members learnt much that is new from the interdisciplinary research projects presented and discussed at the session. We all were equal participants in the event.

**Vladislav Komarov (senior lecturer of the Chair of Inorganic Chemistry, Department of Natural Sciences, NSU, Nikolaev Institute of Inorganic Chemistry)**

Organizing the student conference in the online format was a challenging and very successful experience. The strong student talks and the multifaceted questions from the expert panel and audience made this conference a great success, in line with the best professional scientific conferences. I only missed live conversations during discussion breaks ... but I hope in the future, technologies will enable them!

**Boris Zakharov (associate professor of the Chair of Solid-State Chemistry, Department of Natural Sciences, NSU, Boreskov Institute of Catalysis)**

In my opinion, the united session was a very successful experience. It showed what true interdisciplinary studies are and how science done by mathematicians, physicists and chemists is interrelated. Scientists have a common goal – getting knowledge about the universe. I was pleasantly surprised by the very high level of the contributions; sometimes I had the feeling that I was attending a “real, professional” high-level international scientific conference. The fact that I took part in such an event even during the lockdown regime was certainly a pleasant bonus.

**Anna Gaydamaka (assistant to the Solid-State Chemistry Chair of the Department of Natural Sciences, NSU, Boreskov Institute of Catalysis)**

This year I attended the ISSC for the first time not as a speaker, but as an attendee. I enjoyed the idea of the joint sessions because it is a chance to become familiar with the work of students from other chairs, departments and even cities. Usually, we are limited by our chairs and narrow topics. Interdisciplinarity is a modern world trend; it is often impossible to divide studies into “physical” and “chemical”. I was impressed by the talks of the students who develop new instruments and software. Some of the students, aged 22–23 years, already have significant experience in taking part in “Mega-science” projects. They work together with experienced senior researchers. Interestingly, the students are not scared to choose difficult problems and new methods. For example, some go into protein crystallography. The well-known paradigm of solid-state chemistry “Composition – Structure – Processing – Performance” can be complemented by another one from physics, “Phenomenon – Method – Instrument”, in order to define the novel interdisciplinary scientific approach.

**Ella Moroz (leading researcher, Boreskov Institute of Catalysis)**

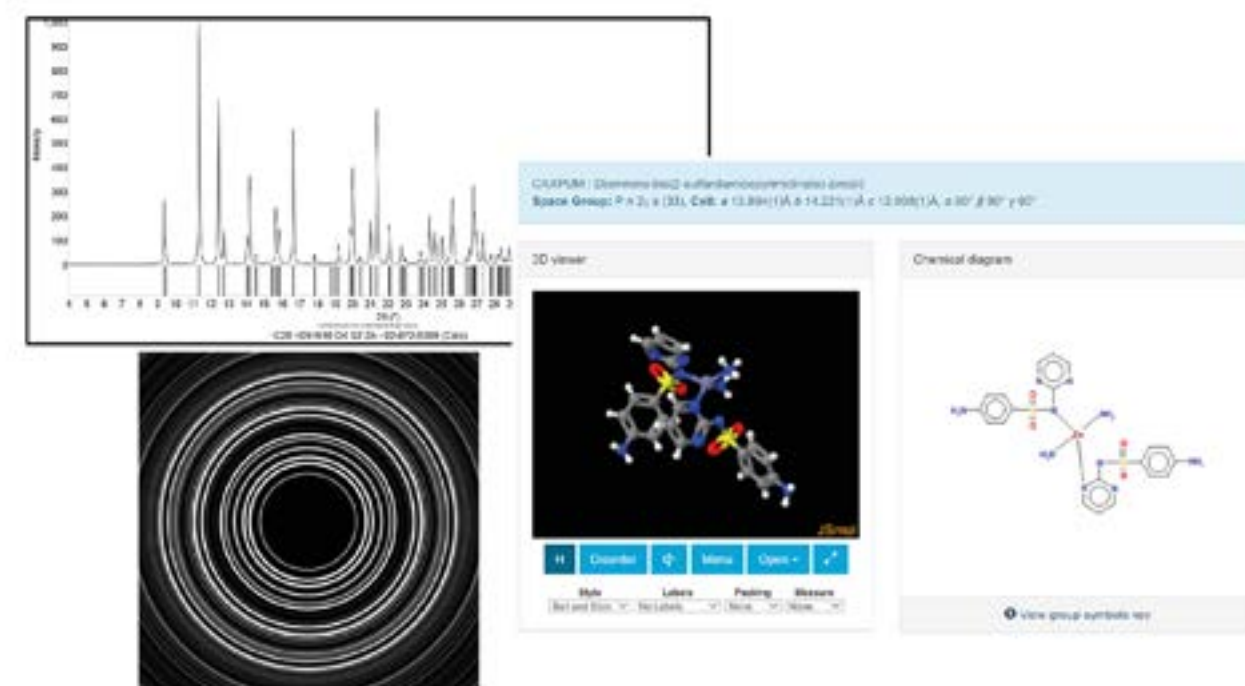
This event is very important, especially nowadays when we have to limit our face-to-face communications. I am grateful to the co-chairs, S. Tsybulya and E. Boldyreva, who did their best to organize this conference. The rich scientific program allowed me to become familiar with the studies of our young colleagues in our country and abroad. I learnt how they master novel instrumental methods and how they are developing them. I learnt a considerable amount about new scientific developments. I

liked the friendly spirit, the interest of all the audience; everybody had the opportunity to ask questions. But most of all, I enjoyed the “children”. The speakers answered the questions of the expert panel members and other participants with much confidence. One could see the positive influence of their supervisors. The most important thing – everybody was enthusiastic about their studies. It’s great! It is most important for their future scientific career. In February, I took part in the Winter School of the NSU “Meeting the Big challenges”, which was organized jointly by the Chairs of Solid State Chemistry and of Physical Methods of Studying the Solid State. I was jealous of the “scholars”. Great lectures from leading experts from Russia and Europe, good conditions, food, and social and cultural program. I understood that our science has a future!

[1] Nikolaev Institute of Inorganic Chemistry, Boreskov Institute of Catalysis, Budker Institute of Nuclear Physics, Lavrentiev Institute of Hydrodynamics, Sobolev Institute of Geology and Mineralogy, Vorozhtsov Institute of Organic Chemistry, Institute of Solid State Chemistry and Mechanochemistry, Voevodsky Institute of Kinetics and Combustion.

## NOTICE

# Data partnership to drive greater opportunities in pharma and materials development



The ICDD CCDC partnership will see the power of powder and single-crystal data combined.

Two of the largest chemical data repositories have announced a new partnership that will allow greater utility and insights in the development of new drugs and materials.

The [International Centre for Diffraction Data](#) (ICDD) publishes the PDF<sup>®</sup> (Powder Diffraction File) and has managed the curation, publication and distribution of X-ray powder diffraction data for 75 years. The [Cambridge Crystallographic Data Centre](#) (CCDC) curates and maintains the CSD (Cambridge Structural Database) – the world's repository for small-molecule organic and metal-organic crystal structures. Both non-profit organisations have a strong focus on data quality, with entries edited and verified manually to ensure accuracy and standardized format.

The partnership will see the ICDD calculate and publish diffraction patterns, including atomic coordinates, from structures found in the CSD. By combining powder and single-crystal data in this manner, the ability to match samples to known structures will be enhanced – enabling, for example, rapid analysis in the pharmaceutical industry from active pharmaceutical ingredient discovery through drug delivery formulation.

*"This collaboration between our two organizations is a win for the scientific community. Materials characterization is enhanced by this symbiotic relationship, particularly in the areas of phase*

*identification and quantitative analysis. The goal is straightforward, to get more data into the hands of more analysts to help them get the right answer."* Tom Blanton, Executive Director ICDD.

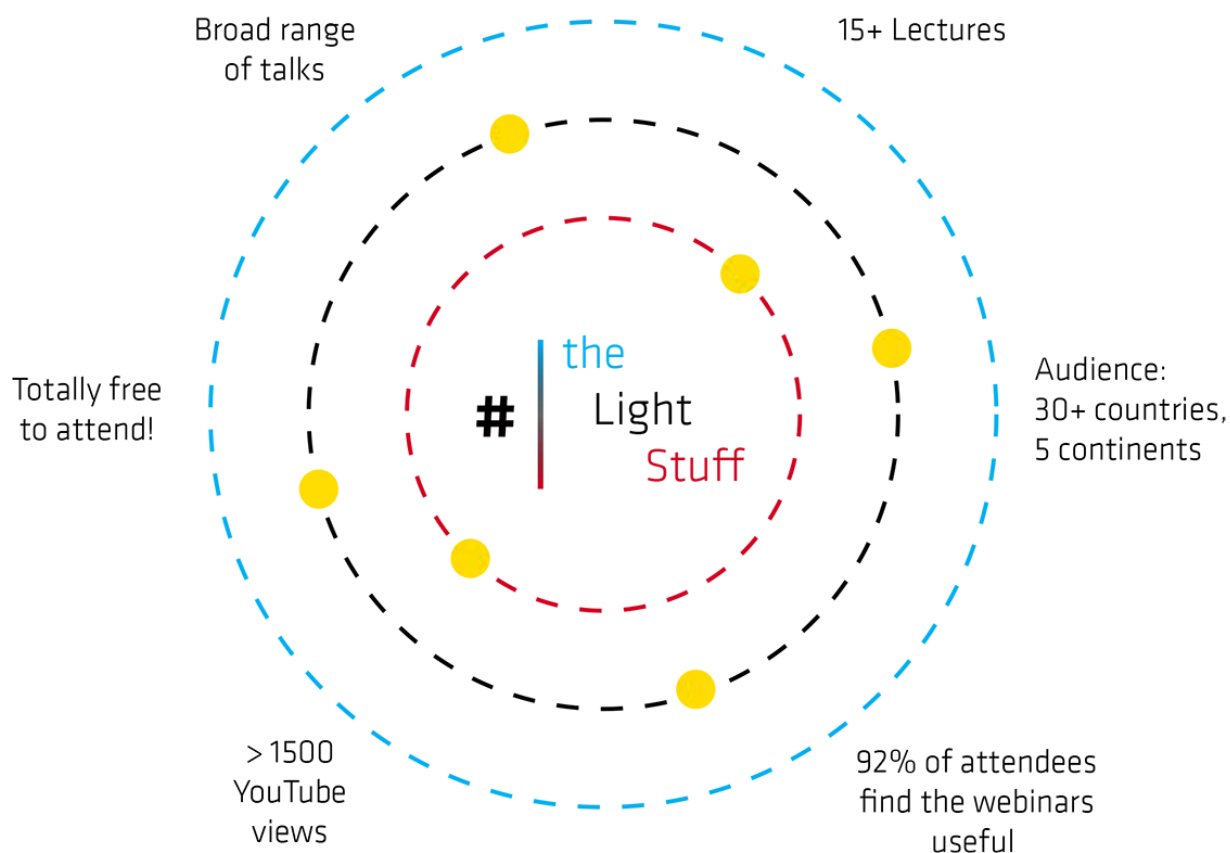
*"CCDC and ICDD have a common aim of collating, enhancing and making high-quality structural chemistry data available to scientists across the world. I am delighted that this collaboration helps extend our commitment to the FAIR data principles increasing the findability, accessibility, interoperability and reusability of this important dataset."* Juergen Harter, CEO CCDC.

As discoveries and developments made using or even exclusively by informatics-based approaches continue to rise, this partnership could open further opportunities for innovative research in the ever-changing pharmaceutical and organic materials landscape.

Please address enquiries to Sophie Bryant, Marketing Manager, CCDC ([sbryant@ccdc.cam.ac.uk](mailto:sbryant@ccdc.cam.ac.uk)), or Helen McDonnell, Marketing Manager, ICDD ([mcdonnell@icdd.com](mailto:mcdonnell@icdd.com)).

## NOTICE

# #theLightStuff Lecture Series



In March 2019, the ongoing #theLightStuff lecture series was established to highlight materials science with an emphasis on the application of diffraction and scattering. Given the current global pandemic, coupled with the increasing awareness of the impact of academic travel on the climate crisis and inclusivity, the online format was established by the team consisting of Brian R. Pauw, Jenni Haberland, Glen J. Smales and Claire Murray from Bundesanstalt für Materialforschung und -prüfung (BAM) in Berlin, Germany and Diamond Light Source in the UK.

The lecture series aims to provide a platform for early-career scientists as well as more established scientists to highlight their work in an accessible way for all members of the community during the lockdowns, regardless of their geographical location. To date, over 450 people have attended these lectures live and the YouTube video recordings collectively have racked up over 1,500 views, which, when combined, are the equivalent of a small IUCr Congress! Every lecture presents a unique angle on a particular technique and/or application of diffraction and scattering and have all been well received by attendees. The audience is comprised largely of PhD students and postdocs, with many attendees using scattering and diffraction methods for their own research. All lectures are recorded (with the permission of the presenter) and are available for you to watch [here](#). We'd love for you to join us for the next lecture, which, as always, is announced on our [website](#).

## HISTORY OF CRYSTALLOGRAPHY

# Johann Puluj (1845–1918): his career and the “invisible cathode rays”

Emilie Těšínská



X-ray pictures of a hand taken by J. Puluj: negative on the left, positive on the right. (From the Archives of the Czech Technical University in Prague, J. Puluj collection, published with permission.)

## 1. Introduction

This article commemorates the physicist and electrical engineer of Ukrainian origin Johann Puluj (1845–1918), who completed his professional career and life as a Professor of the German Technical University in Prague over the period 1884–1918. It was here, where, at the beginning of 1896, he learned the news about the discovery of X-rays by W. C. Röntgen. This article focuses on the professional career of J. Puluj and his work in Prague, mainly based on written sources in the Prague archives and in the Archives of the University of Vienna. For a comprehensive view of the life and work of J. Puluj, reference has been made to the existing biography [1].

## 2. Family background, studies and beginning of his scientific career

Johann Puluj was born on 2 February 1845 in Grzymałów (Hrimailiv/Гримайлів) in the then Galicia, which was part of the Austrian monarchy (today Ukraine). He was born into an Orthodox landowners' family, and was the oldest of several children.



Figure 1. Puluj's native village, Grzymałów, on a postcard from 1900. [Klomm, S. (1937). *Monografia antropogeograficzna miasta Grzymałów.*]

After graduating from the Tarnopol gymnasium, Puluj continued his studies at the University of Vienna (*k. k. Universität in Wien*), at first in the Evangelical-Theological Faculty (1866–1869) and then in the Philosophical Faculty, where, in the school year 1872/1873, he achieved the approbation for teaching mathematics and physics at gymnasiums with German as a teaching language [2]. Consequently, he began to work in the Physics Department (*Physikalisches Cabinet*) of the University of Vienna, led by Professor Viktor von Lang. Puluj started his study of the dependence of internal friction in gases on temperature there; he summed up the results in two essays, which were presented by Professor Lang on 5 February and 23 July 1874 to the mathematics-science class (department) of the Academy of Sciences of Vienna (*die Kaiserliche Akademie der Wissenschaften in Wien*) to be published [3].

### 3. Assistant at the Naval Academy in Fiume

From 1 October 1874, Puluj took up the vacant position of assistant to the Chair of Physics and Mechanics at the Naval Academy (*k. k. Marine-Akademie*) in the then Fiume (today Rijeka).

The Chair was held by Professor Emil Stahlberger. A public selection procedure was announced for the position of an assistant. The annual stipend was 600 guildens plus an apartment in kind (or an annual supplement of 120 guildens for housing). The position was occupied for a period of two years (with the possibility of extension to four years). Applications had to be submitted no later than by 10 September 1874 to the Austrian Ministry of War, at its naval section (*Reichs-Kriegs-Ministerium, Marine-Section*); university graduation in physics and scientific activity had to have been documented. The selection procedure notice pointed out that in case of candidates with the same qualifications, those who would be able to prove their ability to substitute also for other disciplines, namely descriptive and practical geometry, would be preferred. The naval administration also offered reimbursement of the costs of moving the newly appointed assistant from his former place of work [4].

Puluj was recommended to the position by the Philosophical Faculty of the University of Vienna. Immediately after his arrival, in the winter semester of 1874, he was charged with physics lectures on heat, and in the summer semester, after E. Stahlberger had died unexpectedly on 3 May 1875, with

substituting for all the teaching of physics and mechanics [5]. During his work at the Naval Academy, Puluj designed a simple device to demonstrate and measure the mechanical equivalent of heat. He described the principle of the instrument and the results of measurements made with it in two reports sent from Fiume to the Academy of Sciences. At the meetings of the mathematics-science class on 15 April and 10 June 1875, Professor Lang presented them again. The instrument consisted of two main parts, one calorimetric and one dynamometric, which were connected to a simple rotary machine (which at that time was one of the recommended basic pieces of equipment of physics departments of secondary schools). The heat generated by the mechanical friction between the casings mutually inserted in cast-iron cones, one of which was fixed to the rotary machine and the other rotated together with it, was measured. One measurement reportedly only took 30 to 60 seconds. The mechanical heat equivalent values determined by Puluj using this instrument conformed to the value determined by J. Joule and were remarkable for their achieved accuracy [6].

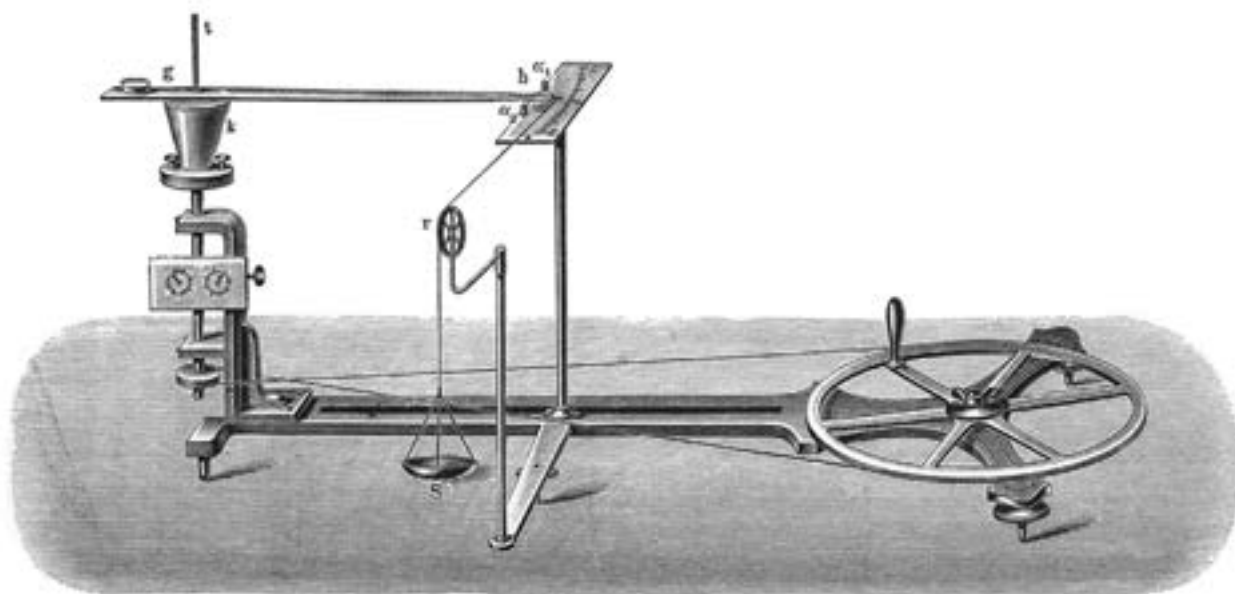


Figure 2. Puluj's apparatus for determining the mechanical equivalent of heat.

Puluj also sent a statement on the instrument to the German physics journal *Annalen der Physik* [7]. In 1878, the device was awarded a silver medal at the General International Exhibition in Paris. In his curriculum vitae written in 1911, Puluj stated that the original device was at that time located in the Physics Institute of the German University (*k. k. deutsche Karl Ferdinands-Universität*) in Prague. A copy made according to this original under his supervision was given to the *Deutsches Museum von Meisterwerken der Naturwissenschaft und Technik* in Munich [8].

#### 4. Doctorate at the University of Strasbourg

For the professorship at the Naval Academy that became vacant after the death of E. Stahlberger, a public selection procedure was declared with a deadline for filing applications by the end of July 1875. A commencing salary of 1,600 guildens was offered (with the possibility of salary promotion up to 2,800 guildens) as well as a service apartment or housing supplement. Qualification for (and experience in) teaching physics at secondary schools with German as the teaching language was required; knowledge of English or French was desirable and was considered an advantage [9]. The professorship was granted to Peter Salcher, Doctor of Philosophy and formerly a Professor at a secondary school (*Oberrealschule*) in Trieste, who was three years younger than Puluj [10].

Salcher took up the professorship at the Naval Academy at the beginning of the school year 1875/1876. Thus, Puluj applied to the Austrian Ministry of Culture and Education (*k. k. Ministerial für Cultus und Unterricht*) for a scholarship for further education at the University of Strasbourg. The scholarship was granted to him in November 1875, subject to his statement that he intended to study for his habilitation and to work for six years at an Austrian university [11]. He left the assistant position at the Naval Academy in Fiume before the end of the year [12].

The *Kaiser-Wilhelm Universität* in Strasbourg was a newly built Imperial-German University in Alsace, which had been annexed to Germany. Puluj started his doctoral dissertation at the Physics Institute, headed by Professor August Kundt. Under the guidance of Kundt and using the instrument Kundt made at his disposal, he continued to study the dependence of internal friction in gases on temperature.

The then dynamic gas theory indicated that the internal friction in gases did not depend on pressure and was proportional to the square root of the absolute temperature. Pressure independence had been experimentally verified (including by measurements by A. Kundt and E. Warburg over a wide range of pressures), but the results of the temperature dependence differed both from the theory and from that of various authors. Puluj carried out measurements of the temperature dependence for room air, carbon dioxide and hydrogen. From 3 January to 2 February 1876, he carried out measurements of 149 samples; he usually did his experiments in the evening or early morning to avoid the disturbing vibrations in the room during daily operation at the institute.

Puluj summed up his results in a treatise dated in Strasbourg in April 1876, which he sent then to the Academy of Sciences in Vienna for publication; it was presented at the meeting of the mathematics-science class on 5 May 1876 [13]. In the treatise, presented as his doctoral dissertation at the University of Strasbourg, Puluj concluded that the internal friction does not depend on the temperature in the same way with all gases. He also mentioned and acknowledged the help of the Professor at the Naval Academy in Fiume, Budisavljević, who carried out calculations for him of the logarithmic decrements using a mechanical calculating machine (*Arithmometer*). At the end of May 1876, Puluj was awarded the title of “Doctor of Natural Philosophy” in Strasbourg. At that time, he was 31 years old. It is worth mentioning that he worked at the University in Strasbourg almost at the same time as W. C. Röntgen (who was only less than two months younger but at that time already a Professor) [14].

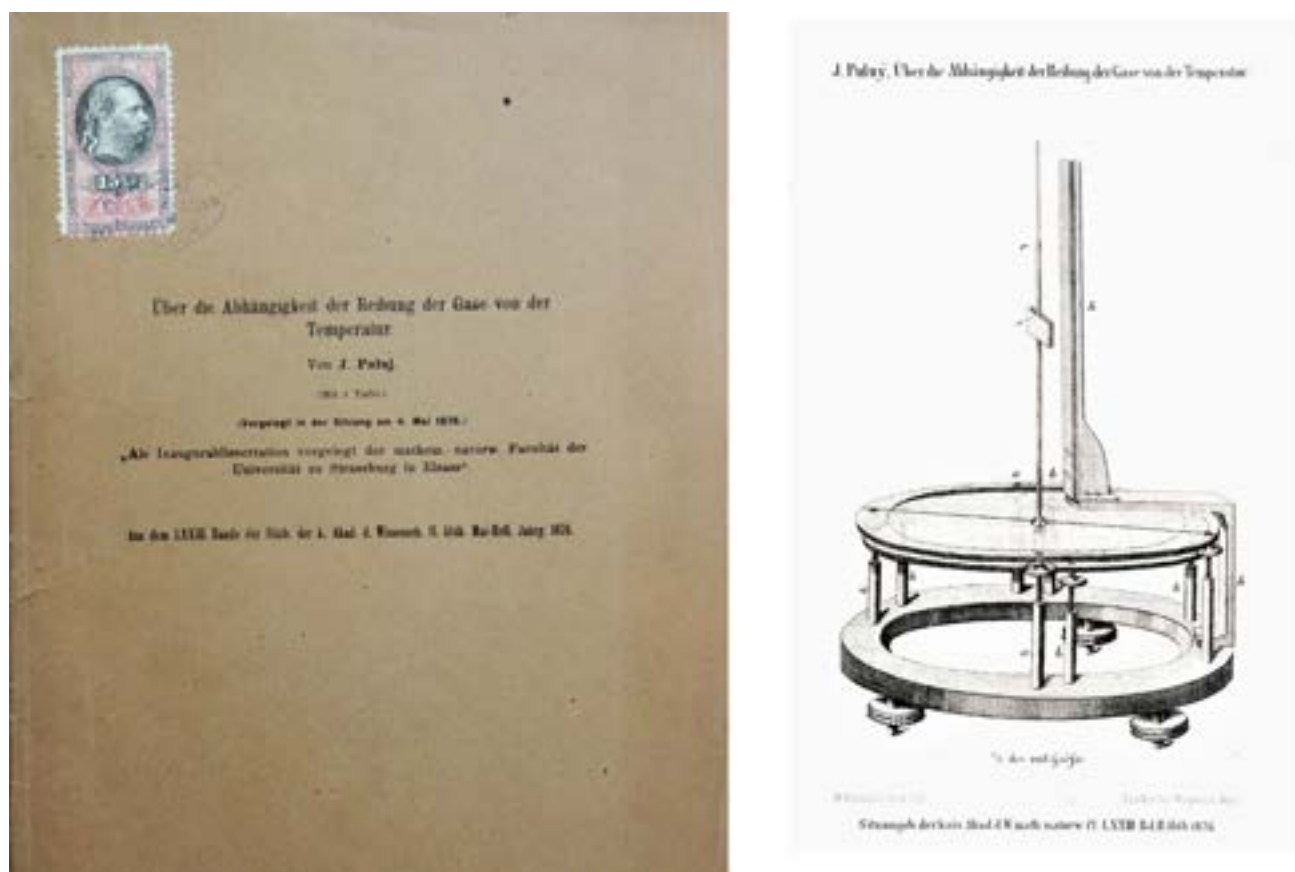


Figure 3. Puluj's treatise presented as his doctoral dissertation at the University of Strasbourg in May 1876. The title page is shown on the left and the apparatus he used for the measurements on the right. (From the Archives of the University of Vienna, collection PH PA 2041/Puluj, J., published with permission.)

## 5. Return to the University of Vienna, the position of assistant and habilitation

After completing his doctorate, Puluj returned to Vienna and on 15 June 1876, he applied for his habilitation [*i.e.* for permission to lecture as a Privatdozent (unsalaried senior lecturer)] in physics at the Philosophical Faculty of the University of Vienna. In the application he stated that he wanted to give lectures on theoretical mechanics, acoustics, optics, magnetism and electricity; in particular, he wanted to offer to the students a well-conceived repetitorium (*i.e.* a revision course) of lectures “of his highly esteemed teacher, Director Stefan” [15]. He outlined in more detail a program of lectures on theoretical optics that he intended to hold in the next semester, highlighting as the main topics the emission and wave theory of light, interference, bending, polarization and double refraction [16].

A commission consisting of Professors J. Stefan, J. Loschmidt, L. Boltzmann and V. v. Lang was established to assess Puluj's application. The Commission (or more precisely, its three members Stefan, Boltzmann and Lang) met on 24 June 1876. It unanimously recommended that Puluj be allowed for further habilitation procedure, *i.e.* for a colloquium and a lecture on probation. On the same day, their opinion was submitted to the Professorial Collegium (board) of the Philosophical Faculty. Further habilitation steps, however, were delayed by the requirement to supplement Puluj's application with the original doctoral diploma and its nostrification (*i.e.* recognition as equivalent to a doctoral diploma from an Austro-Hungarian university); at least a two-year interval between the doctorate and the habilitation was also required. The habilitation colloquium was held on 7 July 1877. Meanwhile, at the suggestion of Professor Lang, Puluj was appointed an assistant to the Physics Department (from October 1876), and he supplemented his habilitation application with some more scientific publications. The habilitation lecture on probation followed on 11 July 1877. Puluj offered three topics: 1. On the radiometer, 2. On the diffusion of gases, 3. On the mechanical theory of heat. The commission opted for the third one. Both the

colloquium and the lecture given by Puluj were recognized as fully sufficient. This successful habilitation procedure was then reported by the Dean of the Faculty to the Ministry of Culture and Education, which by a decision of 6 September 1877 agreed to both the nostrification of Puluj's doctoral diploma and his admission as a Privatdozent of physics at the University of Vienna [17].

After returning to the University of Vienna, Puluj continued his studies of internal friction in gases, vapours and gas mixtures, diffusion of gases. From these topics, he then went on to study phenomena in low-pressure gases in discharged (vacuum) tubes; in the period 1877–1882, he presented to the mathematics-science class of the Academy of Sciences in Vienna four communications on radiometers and four communications on “radiant electrode matter” [18].

Puluj rejected the hypothesis of a fourth state of aggregation of matter, pronounced by the British physicist and chemist Sir William Crookes in his lecture “On Radiant Matter” at the Annual Meeting of the British Association for the Advancement of Science in Sheffield on 22 August 1879, to explain phenomena in highly evacuated cathode tubes (e.g. the creation of a dark space near the cathode). Puluj called the Crookes hypothesis “transcendental” with an inclination to spiritism. With reference to works of the German physicist Johann W. Hittorf on conducting electricity in gases, Puluj offered a more natural (materialistic and mechanical) explanation based on the idea of the “radiant matter of electrodes,” i.e. of physical particles mechanically released from the electrodes that are charged with negative electricity, moving in a straight line at high speed, and together with the residual molecules of the gas mediating the conduction of electricity between the electrodes of the discharge tube [19]. This was before the discovery of the electron as the primary bearer of negative electricity. The lecture by W. Crookes also awoke a prompt and lively interest of physicists in the Czech lands. In 1880 a paper entitled (in Czech) “On the Fourth Aggregate State or Radiant Matter” appeared in the *Journal of Czech Mathematicians and Physicists*. The paper was written by the assistant of physics at the Czech Technical University František Čecháč “using the material that W. Crookes had sent to him most willingly at a kind intercession of the Professor of Physics at the Czech Technical University in Prague K. V. Zenger” [20].

Puluj supported his idea and interpretation of phenomena in vacuum tubes by a series of experiments, to which he alone designed several sophisticated “vacuum apparatuses” in the Physics Department of the University of Vienna. Their production was taken over by the company F. O. R. Goetze of Leipzig. Puluj mentioned the cooperation with this company in his letter to Professor Lang on 25 July 1879 (written in Krieglach, Styria). In the letter, he wrote that the mechanic Robert Goetze from Leipzig had just made a radiometer with a cylindrical glass casing according to his own (Puluj) design; the price of one piece was 12 Marks. Puluj ordered one piece and asked Professor Lang whether he was also interested in buying a specimen for his radiometric collections [21].

Puluj's vacuum apparatuses were introduced in 1881 at the first international electrical exhibition (*Exposition internationale d'Électricité*) in Paris. In particular, the Puluj cathode tube with a fluorescent screen placed between the cathode and an anode that emitted very intense light aroused the interest of both experts and natural science lovers. Puluj was awarded a diploma for his participation in the exhibition. Some of the devices he presented there remain in the *Conservatoire National des Arts et Métiers* in Paris.

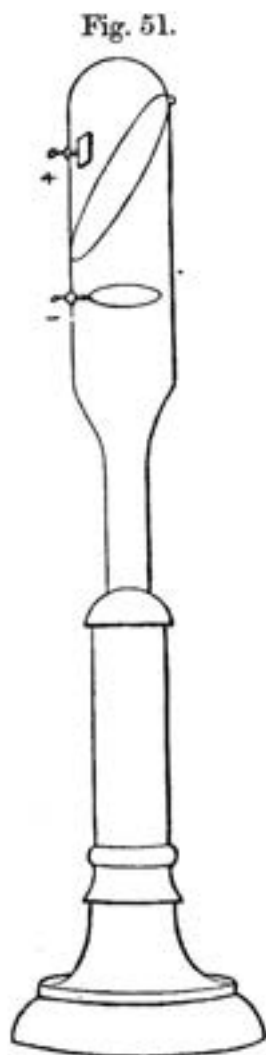


Figure 4. The Puluj phosphorescent lamp. It consisted of a simple glass tube having in the center an obliquely placed elliptical plate of mica covered with calcium sulfide. Below the mica plate was a disc of aluminium of the same diameter as the section of the tube, which served as the cathode (-). Above the mica plate was the much smaller anode (+). The light of the phosphorescent lamp was sufficiently brilliant to light a room, and it was possible to read even at some distance from it [Puluj, I. (1889). *Radiant electrode matter and the so-called Fourth State*, I. c., pp. 293–294, Fig. 51].

In 1882, Puluj received a grant from the Academy of Sciences in Vienna to continue his work on discharges in dilute gases [22]. The strong interest he encountered in Paris prompted him to publish a collected edition of his papers on the radiant matter of electrodes [23]. The dispute with the opinions of such an authority as W. Crookes subsequently became an impulse to translate this edition into English. The translation, according to the text modified by the author concerning new knowledge, was published in 1889 in the series *Physical Memoirs, selected and translated from foreign sources*, which began to be published by the Physical Society of London. In the same volume, a translation of three papers by J. W. Hittorf on the conduction of electricity in gases was published, to which Puluj referred in particular. Translations into English were done by the British physicist, chemist and astronomer Warren De la Rue, who was also interested in the issue of electrical discharges in low-pressure gases; he died before the translation was published [24].

The Puluj collected edition was published in English during his time working in Prague. Related to this was an appended chapter (dated in Prague in February 1888), in which Puluj drew attention to an article by the Professor of General and Analytical Chemistry at the German Technical University in Prague, Wilhelm F. Gintl, of January 1880, where Gintl also critically commented on the Crookes' concept of radiant matter and which had previously escaped Puluj's attention [25].

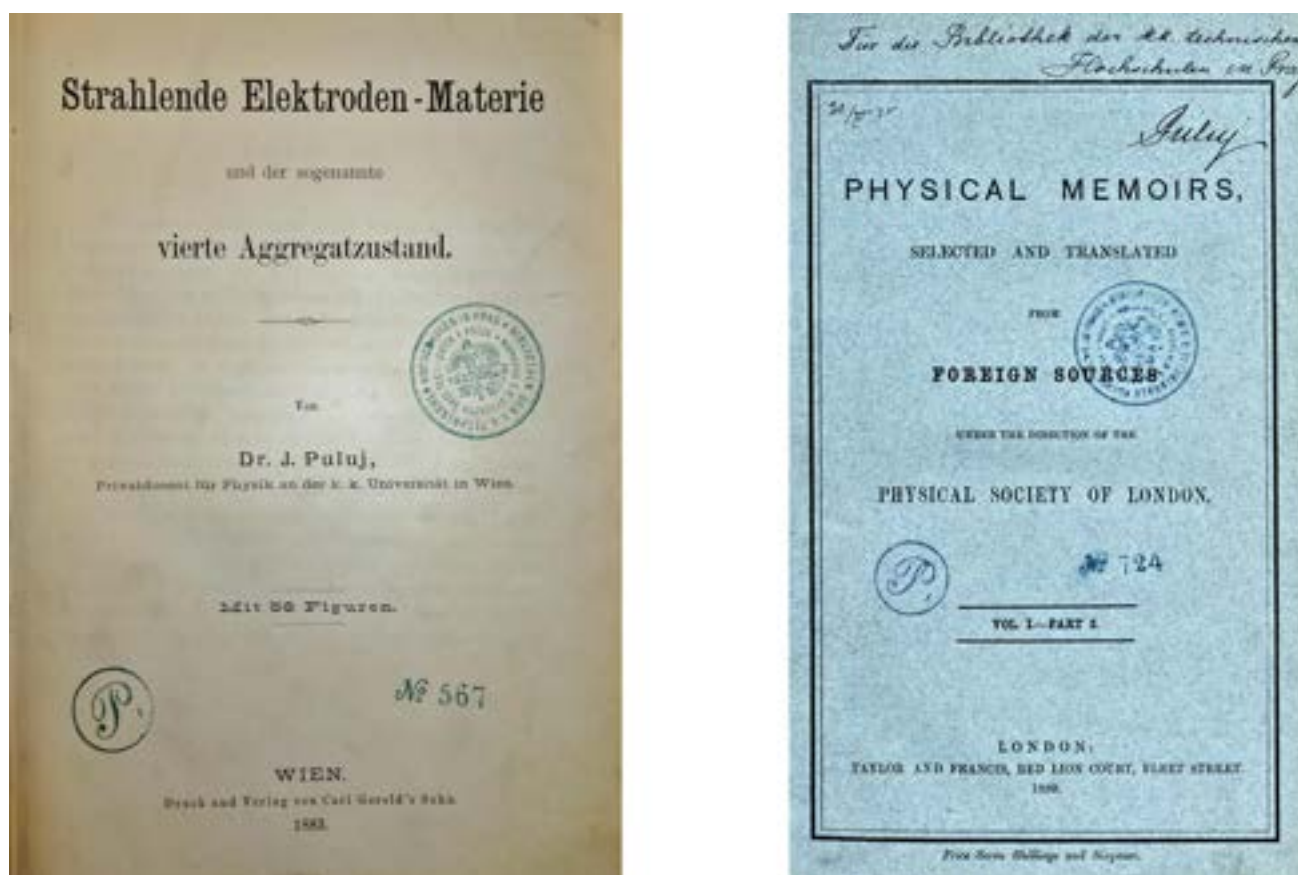


Figure 5. Puluj's treatise on "radiant electrode matter": the 1883 edition in German is on the left and a copy of the *Physical Memoirs* of 1889 (with the translation of the treatise into English) dedicated by Puluj to the Library of Prague Technical Universities on the right. (National Library of Technology in Prague, Rare Books and Special Collections, published with permission.)

## 6. Practical work in the electrical engineering industry

The appointment of Puluj as assistant at the Physics Department of the University of Vienna was extended twice for another two years (which was the permitted maximum), until the end of September 1882. In the suggestions for extending his assistant service, Professor Lang stated with words of praise that Puluj had performed his assistant duties with enthusiasm, published numerous scientific papers and was a skilled experimenter [26].

Working at the University of Vienna in the circle of leading physicists of that time was stimulating for Puluj's career. Yet, his financial situation was uncertain and not great. His assistant salary amounted to 700 guildens a year, a sum that was not easy to live on in Vienna at that time. The income from his lectures as Privatdozent was dependent on the number of enrolled students, which was usually less than ten, and the enrolment fees were usually not even enough to cover the costs of preparing the lectures [27].

Puluj repeatedly turned to the Ministry of Culture and Education with the request for a "fair financial reward" for his teaching and scientific activities at the university. Both Professor Lang and the Professorial Collegium warmly supported his application. But ministerial support was sparse and little. Puluj's financial situation would have been improved by his appointment as a secondary school teacher

in a university city where he could continue his lectures and scientific work. However, Puluj applied for such a position without success [28].

Though the substance of electricity was still unknown (electricity being still taken to be a fluidum), electricity was already successfully applied in technical practice. In 1882, after six years as an assistant and with the knowledge he had acquired, Puluj went into electrical engineering practice. From 1882 to 1883, he was employed as technical manager for the production of Geisler tubes at the Vienna office of the firm Ganz & Co. in Budapest; the company participated in installing the electric lighting at the electrical exhibition held in Vienna in 1883. Then, by the end of 1883, Puluj became a technical consultant to the Austrian arms factory (*Waffenfabrik-Aktiengesellschaft*) in Steyr; the factory chief executive, Joseph Werndel, decided to organize an electrical exhibition in Steyr in 1884 and commissioned Puluj with the task of building and managing a new bulb factory.

Puluj did well in his work in the electrical industry. Yet his relations with Director Werndel were strained. Therefore, an offer coming from Prague was taken by Puluj as an opportunity to resolve his troubles.

In Steyr, Puluj became acquainted with František Křižík, the leading Czech electrical engineer and businessman, who invented an automatically regulated arc lamp in 1880. Křižík later stated in his Memoirs that, in Steyr in 1884, he learned from Puluj “to blow bulbs,” which he then tried to introduce at his electrical plant in Karlín (today a district of Prague). He also acted for some time as an intermediary in some difficult communication between Werndel and Puluj [29].

## 7. Professor at the German Technical University in Prague

### 7.1 Called up to Prague

By the supreme decision of the Emperor Franz Joseph of 8 August 1884 “the Privatdozent at the University of Vienna and Head of Electrical Engineering company Werndel in Steyr, Dr. Johann Puluj” was appointed as an ordinary (full) professor at the Chair of Experimental and Technical Physics in the German Technical University in Prague (*k. k. deutsche technische Hochschule in Prag*, hereafter DTH in Prague), as of the beginning of the school year 1884/1885 [30]. By that time, he had been released from service at Werndel.

I have not been able to find documents explaining the circumstances by which Puluj was called up to the DTH of Prague. However, the matter was clearly under discussion before the end of 1883. The decisive moment was undoubtedly Puluj's expertise and his willingness to include the teaching of electrical engineering within a single Professorship of Physics. An independent Chair of Electrical Engineering had not been set up at the DTH yet (for economic and budgetary reasons and the lack of teaching and working space); Puluj could hardly anticipate that such an interim state would last for the next 16 years! The Philosophical Faculty of the University of Vienna fully supported the proposal to appoint Puluj as a professor at the DTH [31]. Before taking up his new post, on 2 October 1884, Puluj married 21-year-old Katharina, born Stozicky, in Vienna; the marriage produced five (surviving) children, who were born in Prague [32].



Figure 6. Johann Puluj [Stark, F. (1906). *Die k. k. deutsche technische Hochschule in Prag 1806–1906. Festschrift zur Hundertjahrfeier*, Prag, p. 180] and his wife, Katharina Puluj, born Stozicky. [National Archives in Prague, collection "PŘ Praha II, všeobecná spisovna, 1941–1950", box 9230, sign. P 4129/2, Puluj Katharina b. 1863 (passport No. 23 666 issued on 29. 7. 1924), published with permission.]

Having been appointed professor at the DTH of Prague crowned Puluj's earlier attempts to obtain a civil service post that would allow him to continue his scientific work. He accepted the assignment of duties formulated as a representation of the nominal field (*i.e.* experimental and technical physics) and also in the field of electrical engineering, fully corresponding to the needs of the relevant school, as recognition of his scientific and technical activities to date. His starting salary at the DTH (*i.e.* 2,000 guildens plus an efficiency bonus of 480 guildens per year) was lower than the salary he was paid as a consultant and plant manager at Werndel (*i.e.* a fixed salary of 3,000 guildens per year plus a share in the company profit and an apartment in kind) [33]. However, the final provision in the new post (which occurred in 1889) presumed a salary promotion and entitlement to a retirement pension.

## 7.2 Taking up the professorship

At the DTH of Prague, Puluj succeeded Professor Adalbert von Waltenhofen (1828–1914), who was by the supreme decision of 1 August 1883 (after 14 years at the DTH) called up to the Technical University of Vienna to extend the teaching of electrical engineering there [34]. In the school year 1883/1884, the vacated Chair was taken over by the assistant Wilhelm Paukert (who then obtained a scholarship for further education in electrical engineering in Germany) [35].

Puluj took on pedagogical and scientific-organizational duties at the DTH with enthusiasm and support from the Minister of Culture and Education, S. Conrad von Eybesfeld. As Puluj stated in his later biography, thanks to the minister's liberality, a new electrical laboratory and engine room were furnished in the main building of the DTH in Husova Street in Prague already in 1884/1885. In addition, all the rooms of the existing Physics Department (*Physikalisches Institut*) were adapted, and another workshop

for fine mechanics was furnished, in addition to the existing workshop for gross mechanical works, and positions for one mechanic and one servant were established [36].



Figure 7. The old DTH building in Husova Street, Prague (*Die k. k. deutsche technische Hochschule in Prag 1806–1906. Festschrift zur Hundertjahrfeier*, Prag: DTH 1906, s. p.).

Teaching physics was a part of the compulsory curriculum of virtually all the fields of study in the first two years. This included a lecture on general (experimental) physics and a lecture on technical physics supplemented with practical seminars. Electrical engineering, which Professor Waltenhofen began to teach at the DTH, was incorporated into technical physics. However, Puluj introduced separate lectures and seminars in electrical engineering starting from the school year 1885/1886; they were intended for students of higher classes, especially in the field of machine building (mechanical engineering). At first, these lectures were held by Puluj without a requirement for any remuneration. Starting from the school year 1888/1889, he was remunerated for them with 1,000 guildens a year [37].

Rudolf Suchomel became the first assistant to Professor Puluj at the DTH; he held the post from October 1884 for two school years. The position of assistant to the Chair of Experimental and Technical Physics was later changed to the higher position of a constructor (a design engineer). It was not easy to get an expert with the necessary skills to hold the position. The high demands of Puluj were well met by the Doctor of Philosophy Johann Sahulka, who became Puluj's assistant in Prague from 1889 to 1890. Sahulka (1857–1927) studied mathematics and physics at the University of Vienna, and for two years, he also attended lectures in electrical engineering at the Technical University of Vienna. At the DTH, he then passed an electrical engineering examination in 1890 (with Puluj) and in May 1891 applied for a habilitation in electrical engineering there; during the habilitation procedure, on the proposal of Puluj, the subject of the Sahulka habilitation was specified as “*venia legendi* for alternating currents and their application.” In 1894 Sahulka returned to Vienna, and in 1903 he became Professor of Electrical Engineering at the Technical University of Vienna [38].

### 7.3 Project for a modern institute for physics and electrical engineering

In 1895 Puluj proposed to build a modern institute for physics and electrical engineering at the DTH in Prague. In connection with this and with a grant of 300 gulden from the Ministry of Culture and Education, he went on a study trip to Switzerland and Germany in the summer of 1895. In his report on the trip (submitted to the Ministry on 27 May 1896), he stated that foreign technical universities with magnificent electro-engineering institutes outperformed Austrian technical schools by a full decade [39].

Nevertheless, Puluj did not live to experience a new modern institute for physics and electrical engineering at the DTH, despite his repeated suggestions, urgencies and persuasive arguments (which were commonly recognized as legitimate).

It was at the time of his planning of the new institute, and after returning from the study trip to Switzerland and Germany, that Puluj learned, in early 1896, of the discovery of X-rays by W. C. Röntgen, which was bitter news for him. We shall come back to Puluj's response to Röntgen's discovery in part 8 at the end of the article.

### 7.4 Establishing a Chair of Electrical Engineering

Requirements for the teaching of electrical engineering increased, both in the amount of the curriculum and in the interest of students. This was related to the development of the field and its growing economic importance. Ensuring quality teaching of electrical engineering within a single physics Chair and its confined space in the main building of the DTH in Prague was becoming unbearable. After 16 years, at the threshold of the new millennium, Puluj objected to this situation.

In his letter dated 12 March 1900 and sent directly to the Ministry of Culture and Education, Puluj called for the division of his professorship into a separate professorship for experimental physics and the one for electrical engineering. He noted that his teaching commitment at the DTH at that time included two two-semester courses in physics (general physics and technical physics, totaling 7 hours of lectures per week) and two two-semester courses in electrical engineering (totalling 4 hours of lectures and 3 hours of practical seminars per week); he had taught both disciplines with enormous effort and regardless of his health; he was not able to bear this burden any longer than by the end of the summer semester 1900 [40].

In a personal audience given to Puluj, the Minister of Culture and Education, Wilhelm von Hartel, promised to consider the request. The Professorial Collegium of the DTH fully supported Puluj's demands. Based on the proposal of a commission set up for the case, the collegium recommended: 1) to systematize the Chair of Electrical Engineering as an ordinary (full) professorship and entrust it to Professor J. Puluj, 2) that steps be taken to fill the new Chair of Experimental Physics also by an ordinary professor. The most complicated question, namely where to locate the new Chair of Electrical Engineering, however, was not decided for the moment.

The Ministry of Culture and Education and the Ministry of Finance assessed the presented proposals in terms of financial costs. The division of the Chair of Experimental and Technical Physics at the DTH was agreed upon, but the implementation was postponed until 1901 [41]. Having been assured by the Ministry of Culture and Education that the division of the existing Chair of Physics at the DTH would take place from the summer semester of 1901, Puluj assented to the request for teaching of both physics and electrical engineering for another semester. However, at the end of the winter semester 1900/1901, by a letter to the Rectorate of the DTH of 12 February 1901, the Ministry again postponed the division of the Chair to the beginning of the school year 1901/1902. The problem arose, namely around the location of the Chair of Electrical Engineering and the associated costs [42].

Puluj responded with a new urgent letter to the Ministry dated 3 March 1901. This time he categorically rejected further representation of both subjects. He wrote that his mental and physical exhaustion was

exacerbated by a complicated influenza illness he had suffered in the meantime. He insisted on the division of the Chair of Experimental and Technical Physics from the coming summer semester of 1901. On a question that the Ministry had addressed to him, Puluj expressed his willingness to take over the new Chair of Electrical Engineering but he set his own conditions. If they could not be fulfilled, he intended to keep the Professorship of Physics, with the existing staff. In this case, he recommended that the new electrical engineering Chair was occupied by a full professor “with the necessary qualifications” and that at least one constructor, one laboratory technician and a corresponding subvention were provided to him.

There was evident interest on the part of the Professorial Collegium and the Rectorate of the DTH that Puluj, as a highly renowned expert in electrical engineering, should take over the new Professorship of Electrical Engineering; filling a Chair of Physics with a good specialist seemed easier. The requirements stated by Puluj were generally justified. However, resolving the issue before the beginning of the semester was not possible for an advanced date. For the summer semester of 1901, a temporary solution had to be undertaken. Puluj was willing only to teach physics in that semester. As it was necessary to provide the obligatory teaching of electrical engineering for students of machine building too, Puluj suggested that the constructor at the chair of physics, Robert Mayer, should be appointed as a substitute for the lectures, and possibly to supervise practical seminars. He offered to share a physics auditorium with him. Puluj said that Mayer was an excellent graduate of the DTH (in the machine-building field). He had also studied in London for a year but had only been involved in electrical engineering for one year (since his appointment as a constructor). For the time being, however, he did not have the qualifications to provide full electrical-engineering education. Therefore, this only was an emergency solution for the summer semester of 1901, which was not to continue in the next school year [43].

The division of the Chair of Experimental and Technical Physics at the DTH was made on 1 October 1901 based on a decision of the Ministry of Culture and Education of 2 July of the same year. The decision stipulated that one of the two newly created independent Chairs, on which the current Professor, Puluj, would decide, would be systematized as an ordinary professorship, and the other one would be an extraordinary professorship. The Ministry pointed out that there were no financial resources available to systematize a second ordinary professorship. The former positions of the constructor and mechanic were transferred to the Chair of Electrical Engineering. For the Chair of (Experimental) Physics, a systematized assistant post with the annual salary of 1,400 K was established from 1 October 1901. The Chair of Electrical Engineering was awarded an annual subvention of 2,000 K. Lectures, and practical seminars on electrical engineering were supposed to be held as so far in the premises of the physics Chair in the main building in Husova Street; when drawing up the school timetable, however, care had to be taken to ensure that physics and electrical engineering did not interfere with each other [44].

Puluj decided on the Chair of Electrical Engineering (even though not all his requirements were met). Therefore, the Chair of Physics became an extraordinary professorship. Until the filling of this vacant position, Josef Geitler von Armingen, a Privatdozent with the title (not salary) of extraordinary professor at the German University of Prague, was entrusted as a substitute for physics lecturing at the DTH.

## 7.5 Filling the Chair of Physics

A commission headed by Puluj was entrusted with the task of preparing a proposal for filling the extraordinary physics professorship at the DTH on 17 July 1901. At its first meeting on 7 January 1902, the commission asked Puluj to look for suitable candidates. On 8 April 1902, Puluj presented the professional curricula of five candidates. The commission then drew up a proposal of the usual “triennial,” which was submitted to the Professorial Collegium on 23 June 1902 and with their approval passed to the Ministry on 6 November 1902 by the Rector of the DTH.

In the first place, two “local” candidates were proposed *ex aequo* (as equal in power): the above-mentioned 31-year-old Josef Geitler von Armingen (a native of Prague) and 37-year-old Josef Tuma, at that time Privatdozent and adjunct of the Physics Institute of the German Technical University in Brno (Moravia). Stefan Meyer, a Privatdozent and assistant of the Physics Institute of the University of Vienna, was nominated in second place. Two other *ex aequo* candidates were nominated in third place: Anton Lampa, a Privatdozent and assistant of the Physics Institute of the University of Vienna, and Eugen von Schweidler, a Privatdozent and assistant of the Physical Chemistry Institute of the University of Vienna. The Ministry decided on J. Tuma. His advantages included higher age and richer professional experience, and also the order in which Tuma and Geitler were previously proposed to the Professorship of Technical Physics at the German Technical University in Brno (Tuma was second and Geitler the third candidate for that professorship) [45].

The final filling of both Chairs was made by the supreme decision of 18 October 1902: Puluj was appointed ordinary Professor of Electrical Engineering and Tuma as extraordinary Professor of Physics. Each of their teaching duties was defined as a proper representation of the field in the theoretical and practical direction to the extent corresponding to the requirements of the particular technical university, subject to possible later modification [46]. Puluj's salary remained unchanged. Tuma was assessed a commencing salary of 3,600 K plus an efficiency bonus of 840 K per year, and took up the new post on 1 November 1902; five years later he was appointed ordinary professor. J. Geitler continued to work at the German University of Prague until September 1906, when he was appointed ordinary Professor of Physics at the University of Czernowitz in Bukovina (now Ukraine).

## 7.6 Professor of Electrical Engineering

As a Professor of Electrical Engineering, Puluj held a two-semester lecture, “General Electrical Engineering,” at the DTH of Prague, which was obligatory for students of machine building and was completed with practical seminars in a laboratory. Also, “on his own initiative” and until 1912/1913 without any remuneration requirements, he gave optional one-semester lectures on selected chapters of electrical engineering; these lectures were intended for students of the field of machine building who had attended the compulsory lecture and were interested in a more in-depth education in electrical engineering because they intended to work in the electrical engineering industry.

In particular, in the school year 1911/1912 Puluj gave optional lectures: “Selected chapters from AC electrical engineering: theory and calculation of transformers and three-phase motors” (1 hour per week in the winter semester), “Special electrical engineering: power plants for lighting and power transmission” (2 hours per week in the summer semester) and “Introduction to electrical engineering machines and devices” (1 hour of lectures and practical seminars in the summer semester).

Puluj had to supervise by himself all the practical seminars because only unqualified persons still applied for the position of constructor (whom he had to train first and who, usually after one or two years, would leave for a better-paid position in industry) [47]. Furthermore, Robert Mayer did not stay as constructor at the DTH for very long. From the beginning of the school year 1903/1904 he took up the post of a teacher of electrical subjects at the German Technical College (*k. k. deutsche Staats-Gewerbeschule*) in Brno; the commencing payment offered for this post was 2,800 K plus an efficiency bonus of 600 K, whereas as the constructor at the DTH, Mayer drew an annual remuneration of 2,400 K [48].

The proposal for the establishment of the second electrical engineering Chair at the DTH, which was to form the basis for the establishment of an Electrical Engineering Department with an independent second state examination, was submitted to the Professorial Collegium in 1906. Particular negotiations on this issue in the Collegium and with the Ministry, however, did not start until 1916, when it was related to Puluj's retirement. The establishment of the Electrical Engineering Department subordinate together with the Department for Machine Building to the same dean's office (*Abteilung für*

*Maschinenbau und Elektrotechnik*) and the Electrical Engineering Institute only became a reality with Puluj's successor.

### 7.7. Puluj's last assistant at the DTH in Prague

Puluj's last assistant (paid from the resources arranged for a constructor) was Ing. Johann Wegscheider, who held this post after 1 December 1912 [49]. Just like Mayer, he graduated from the machine-building department at the DTH. He continued to work as an assistant under the guidance of Puluj's successor [50]. During World War I, when he was called up to the army, the student Johann Fukke stood in for him [51].

### 7.8 Retirement

In the school year 1914/1915 (on 2 February 1915), Puluj reached the age of 70, which was – at that time – the age limit for holding a university teaching office. At a meeting of the Professorial Collegium on 18 January 1915, the decision was made to ask him to hold his teaching office for one more honorary school year. The Rector, J. Tuma, however, disagreed and attached his separate *votum* to the proposal presented to the Ministry. He argued that Puluj's successor should be provided with the opportunity to get involved as soon as possible in the establishment of the Electrical Engineering Department and the Electrical Engineering Institute. While the Ministry listened to this argument, it nevertheless complied with the proposal to extend Puluj's mandate at the DTH in Prague – given his merits “and not to endanger his health.”



Figure 8. House in Prague-Smíchov, now Presl (at Puluj's time Fibich) Street No. 1268, the last residential address of Puluj and his family. The plaque remembering Puluj was placed on the house in 1930. (Photo credit: E. Těšínská, January 2020.)

Puluj retired on 30 September 1916. In this context, upon the supreme decision of 28 October 1916, he was bestowed *Komturkreuz des Franz-Joseph-Ordens* [52]. He was assessed a pension of 11,840 K, which, however, he did not enjoy for long. He did not live to the age of 73, nor to the end of the World

War and the collapse of Austria-Hungary. He died on 31 January 1918. He and his wife (who died on 8 August 1945, after World War II) are buried in the cemetery in Prague's Smíchov, Malvazinky.



Figure 9. Grave of Puluj and his wife at the cemetery in Prague's Smíchov, Malvazinky. (Photo credit: E. Těšínská, January 2020.)

Puluj's successor at the Chair of Electrical Engineering at the DTH of Prague became Friedrich Niethammer (1874–1947), formerly Professor of Electrical Engineering at the German Technical University (*k. k. deutsche Franz-Joseph-Technische Hochschule*) in Brno. He took up the position from 1 March 1917 [53].

## 7.9 Career posts and social honours

During his work at the DTH in Prague, Puluj held responsible academic positions. He worked in expert committees and associations, and as an expert, he continued to cooperate with industry. In the academic year 1888/1889, he held the office of Rector of the DTH. He was a member of the examination committee for civil engineers and architects, and chairman of the scholarship committee for outstanding academic achievement. He was a sworn expert in Electrical Engineering at the Commercial Court for Bohemia. He participated in the construction of power plants in Bohemia, Moravia and elsewhere. As an invited expert, he also endorsed the project of the Prague municipal power plant with a single AC central power station. He was an honorary member of the *Elektrotechnischer Verein* in Vienna. He was awarded the title of Court Councillor (*Hofrat*) and a gold medal at the Jubilee Exhibition (*Kaiser Franz-Josef Jubiläums-Ausstellung*) in Prague in 1908.

## 8. Puluj's response to the discovery of X-rays

The sensational news of Röntgen's discovery of X-rays also reached Prague in early January 1896. On 7 January, for example, the German newspaper *Bohemia* reported on it by reprinting a piece of news from the Vienna newspaper *Die Presse* [54]. The author of the news was the Austrian physicist Ernst Lecher, son of the editor of the newspaper *Die Presse* and from 1 October 1895, Professor of Physics at the German University in Prague (where he succeeded Ernest Mach). Lecher learned about the discovery of X-rays by Röntgen in Vienna on 5 January from the professor of the University of Vienna, Franz Exner, his former teacher [55]. Three weeks later, on 24 January, Ernst Lecher delivered a lecture, "On the new discovery of Professor Röntgen (including demonstrations)" to the Prague Association of German Medical Doctors. Incidentally, it was Lecher who succeeded Puluj as Professor Lang's assistant at the University of Vienna in 1882. However, there were also other communication channels, through which people at the Prague universities could learn about Röntgen's discovery. Their response to the discovery was similar to elsewhere in the world. In the physics and electrical engineering departments of both the Czech and German Prague universities, Röntgen's experiments were immediately repeated, and X-rays, their origin and properties, subjected to further investigation. X-ray images of various objects were produced in cooperation with experts or amateurs in the field of photography. A possibility to use X-rays in medicine was the subject of special interest (though initially also with scepticism). These first experiments were reported at meetings of learned societies and professional associations; apart from that, lectures and demonstrations were also organized for the general public [56].

On 17 January 1896, the Professor of Electrical Engineering at the Czech Technical University in Prague, Karel Domalíp, reported, probably as the first one in Prague, on his experiments with X-rays at a meeting of the mathematical-science class of the Czech Academy of Emperor Franz Joseph for Sciences, Literature and Art. He presented an X-ray picture of a human hand there, which he took at the Institute of Physics and Electro-Engineering of the Czech Technical University on 11 January [57]. It was allegedly the third object photographed "under the cathode light according to Professor Röntgen's method" at that institute using a Crookes tube with a platinum cross inside, connected to the anode, and a side cathode in the shape of a concave mirror (the first two photographed objects were a thermoelectric pile and a metallic ornament) [58]. The picture was photographically processed (he developed a negative and produced a positive) by the Privatdozent of Technical Chemistry (from 1899 the Professor of Fermentation Chemistry and Photography) at the Czech Technical University in Prague, Karel Kruis [59]. This successful picture was subsequently published in the Czech popular science journal *Živa* [60] and then also reproduced in other magazines.

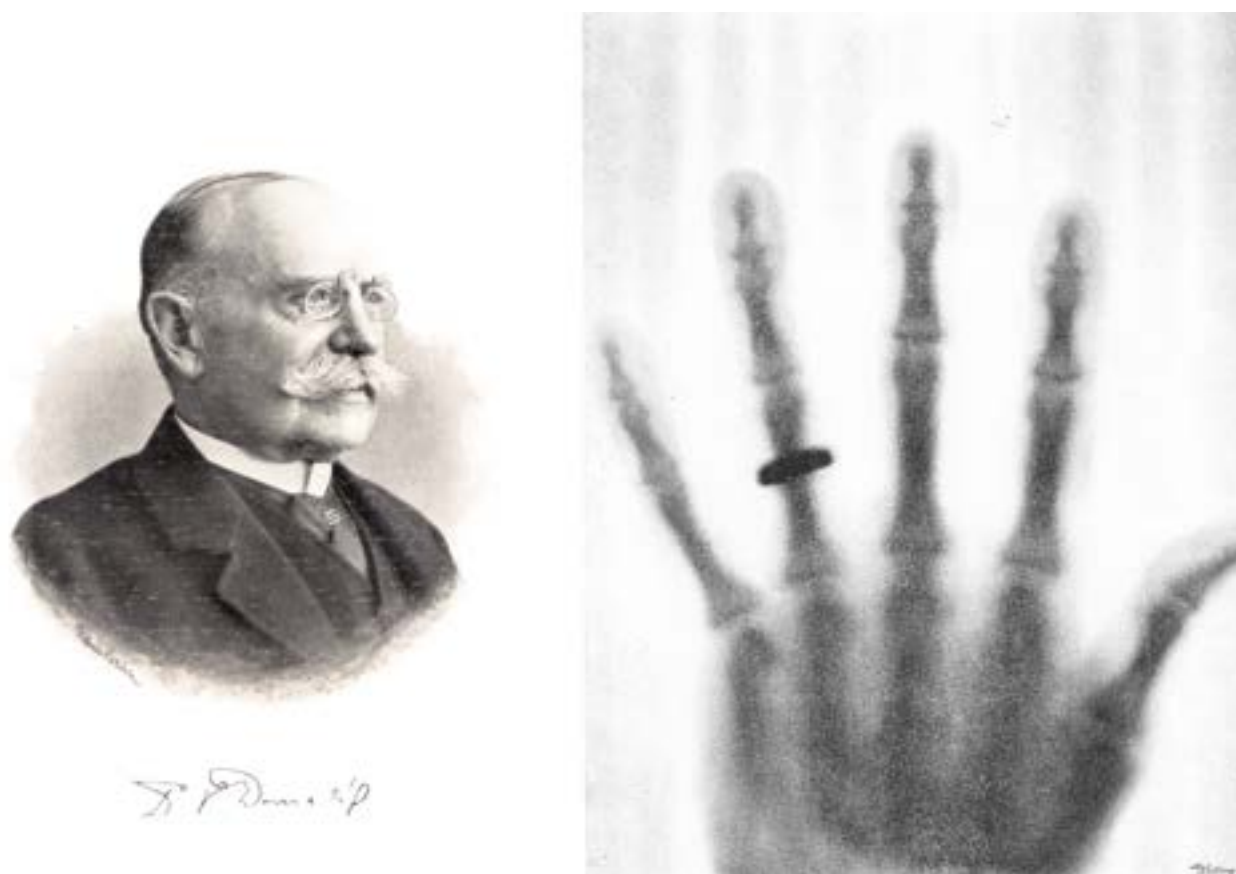


Figure 10. Karel Domalíp (1846–1909), Professor of Electrical Engineering at the Czech Technical University in Prague (on the left) [*Časopis pro pěstování matematiky a fyziky*, 1910, Vol. 39, s. p.] and the X-ray picture of a hand made by K. Domalíp in cooperation with the chemist K. Kruis on 11 January 1896 [*Živa, časopis přírodnický*, Vol. 4, 1896, s. p.].

K. Domalíp (1846–1909) was a professional counterpart of Puluj at the Czech Technical University in Prague. He had studied mathematics and physics at the Philosophical Faculty of the University in Prague (at that time not yet divided into Czech and German), where he was one of the students of E. Mach. After his studies, in 1871–1876, he worked as an assistant to A. von Waltenhofen at the DTH in Prague, where he completed his habilitation for theoretical physics in 1875. In 1884 he made his habilitation for electrical engineering at the Czech Technical University in Prague, where, in 1891, he was appointed extraordinary professor (and in 1893 ordinary professor). In the meantime, in the years 1875–1891, he was employed as a secondary school teacher [61].

Puluj was somewhat disappointed about the news of Röntgen's discovery of X-rays, which overshadowed all previously made pioneering works on cathode rays, namely his own valuable contributions. Based on the archives being at my disposal, I am not able to document Puluj's early experiments and alleged X-ray pictures he had made (unknowingly) with his lamp before Röntgen's discovery. For this interesting, but sensible question, I prefer to refer to the existing biographical literature and their sources for the moment [62]. I limit myself here just to Puluj's immediate response to Röntgen's discovery.

During January–February 1896, in the Physics Department at the DTH, Puluj verified the emergence of and the photography by "invisible cathode rays" using his old lamp with a fluorescent screen. He achieved very successful images. Consequently, he focused on the location of X-ray formation in the cathode tube and on the origin of the X-rays. He reported on the results of his experiments, as before, to the Academy of Sciences of Vienna.

At the meeting of the mathematics-science class of the Vienna Academy on 6 February 1896, it was recorded that Professor J. Puluj sent eight photographic reproductions of pictures he had made in the DTH Physics Department in Prague with his vacuum apparatuses. In the accompanying commentary, Puluj stated that he had been encouraged to do so by a lively interest that was aroused in the widest

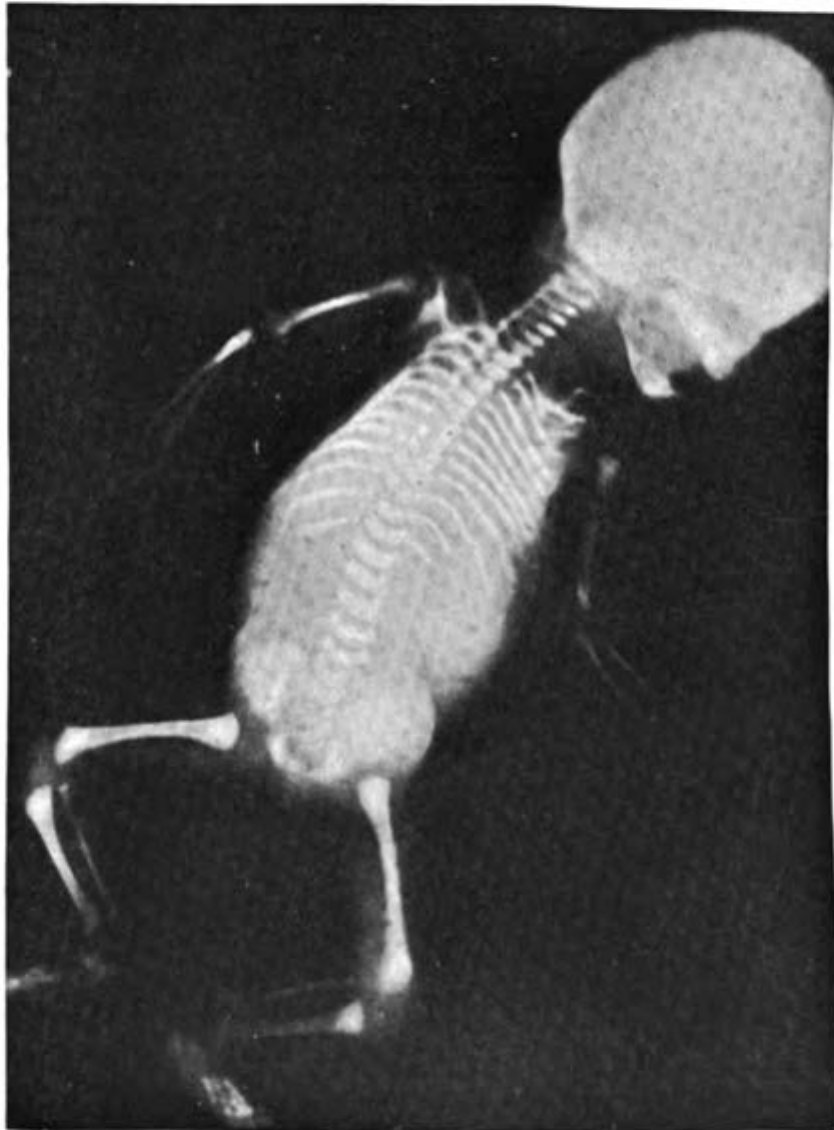
circles by the photographic effects of the invisible cathode rays discovered by Professor Röntgen. The pictures he presented, he continued, showed a "not insignificant" progress over other reproductions known to him and showed what use this method of photography would find in surgical practice (see photos at the start of this article). He added that on 2 February, he managed to photograph a two-month-dead baby in this way; the reproduction to be sent later to the Academy clearly showed not only the details of the chest but also the entire spine up to the head; the newborn head could not be displayed due to the small size of the photo plate. He added that the head of an adult human, however, could not be displayed with the radiation provided by the current cathode tubes [63].

On 8 February 1896, just six weeks after Röntgen presented his discovery of X-rays, Puluj published his own findings in the French journal *La Nature* in Paris.

In a report from the following meeting of the mathematics-science class of the Vienna Academy on 13 February 1896, "another remarkable reproduction of a cathode-ray tube picture taken in the DTH Physics Department in Prague led by Puluj" was recorded, namely a photograph of a dead nine-day-old baby. In the same session, a communication by Puluj on the origin of X-rays and their photographic effects was accepted for publication. In it, based on his own experiments, Puluj stated to have confirmed Röntgen's assumption that X-rays came from the glass wall of the discharge tube, which was exposed to visible cathode radiation and which phosphoresced. He further confirmed that a screen covered with calcium sulfide in his lamp (which he constructed 15 years before) not only shined strongly (as described earlier) but also emitted very intense invisible radiation; he stated that with this lamp, clear images of small objects could be recorded on the photographic plate after just two seconds of exposure. Puluj further suggested that all substances that were intensely shining in cathode rays had a strong emission capability even for X-rays. He described several new modifications of his lamp, which he made especially for photography with the invisible cathode rays. He discussed in more detail several clinically interesting cases that he photographed with the new method: a tuberculous hand, a hand with a revolver bullet embedded in the upper bone of the middle finger, the head of a young man with a 6-mm revolver bullet inside for years (yet, even the 1.5-hour exposure failed to locate the bullet), a leg with a small iron tip in the thigh, a dead newborn baby, a healing broken arm of a 13-year-old boy, a hand of a two- and a four-year-old child and a full body picture of a newborn baby. Puluj pointed out the problem with displaying small metal objects in the human or animal body resulting from the divergence of X-rays of the cathode tube, but he was trying to solve this problem. As a success, he mentioned a picture of a "shot guinea pig", in which it was possible to depict clearly small metal pellets and a bullet. Regarding the way X-rays are formed, Puluj stated his opinion that the conversion of visible cathode radiation to invisible radiation took place through material bodies. Particles mechanically plucked from the cathode, which transferred negative static electricity, balanced their electrical charges when they struck glass walls or other solid bodies, causing molecules and their *ether* shells to shake. Each point of the glass wall or screen on which the cathode radiation fell became the starting point of the *ether* waves, which propagated (depending on the type of oscillation and the duration of oscillation) either as visible or as invisible radiation [64].

THE PHOTOGRAM.

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HUMAN SKELETON.

*Radiographed by Professor Puluj.*

Figure 11. A full body picture of a newborn baby made by J. Puluj in 1896 [*The Photogram*, 1896, Vol. 3, No. 28 (April), Special supplement, p. 104. Accessible [here](#)].

In a supplementary communication, which was adopted for publication at the meeting of the mathematics and science class of the Vienna Academy on 5 March 1896, Puluj described further direct experiments that had confirmed the correctness of the assumption that in Puluj's lamp new radiation was created on the phosphorescent mica screen (namely on the side covered with the phosphorescent substance) rather than at the glass wall or the cathode [65].

Upon making pictures of clinically interesting cases, Puluj cooperated with German university clinics and hospitals in Prague (with Professor G. Pick from the internal clinic, Professor A. Wölfler from the surgical clinic and the pathological anatomist H. Chiari). A short report on the pictures taken by Puluj using his "vacuum apparatuses" was published as early as 31 January 1896 by the German newspaper *Prager Tagblatt* [66]. In the Czech magazine *Světlozor*, in the 31 January 1896 issue, a reproduction of the above-mentioned picture of a "shot guinea pig" was published. The label stated that the photo was taken using Puluj's lamp by amateur photographers (K.) Paspá and (? E.) Šafařík; on the left front leg of the animal, there were well recognizable "bird" pellets, and behind the right foot, a bullet from a Flobert gun [67].

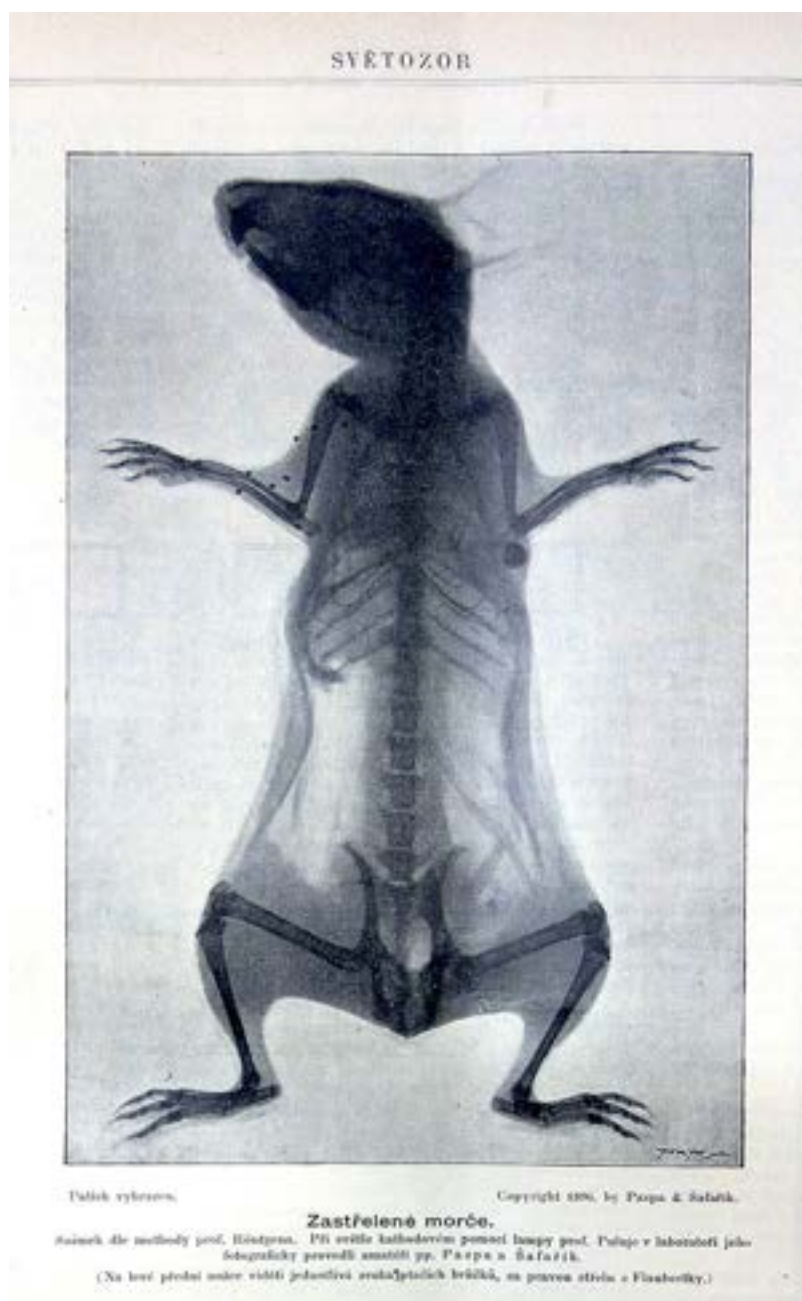


Figure 12. Picture of a shot guinea pig made with "cathode light of Puluj's lamp" in Puluj's laboratory by photographers Paspá and Šafařík. Pellets can be seen in the left front leg of the animal and a bullet behind its right front leg (*Světlozor*, 1896, Vol. 30, No. 14, p. 143).

On the evening of Saturday 14 February 1896, Puluj gave a lecture on "invisible cathode rays" for the public. A detailed report on the lecture was published on 18 February 1896 in the German newspaper *Bohemia*. The lecture took place in the physics auditorium in the main building of the DTH in Husova Street. It was widely visited, and "there were even numerous ladies among those present." It lasted two hours and ended with lively applause from the audience. The financial proceeds from the lecture were donated by Puluj to a fund for supporting poor students of electrical engineering.

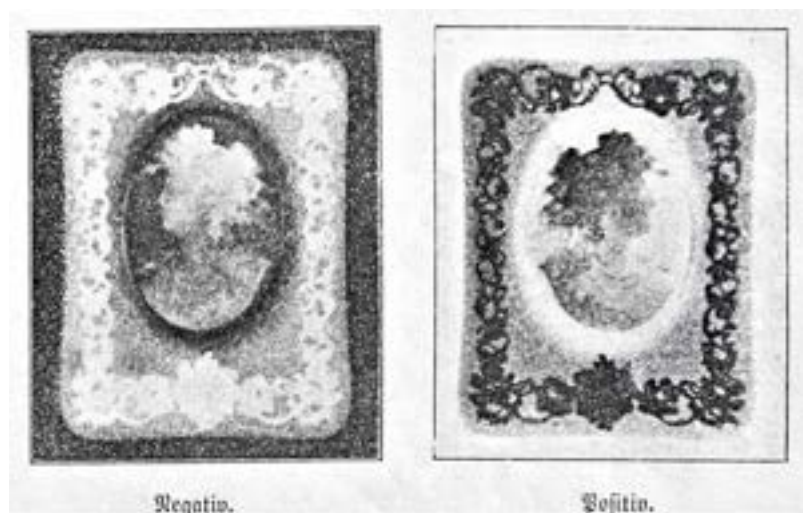


Figure 13. A cameo with a nacre frame, an X-radiograph made by Puluj in Prague in 1896 (?): negative on the left and positive on the right [published by Lanner, A. (1897). "Die Entdeckung der Röntgenstrahlen," *Jugendheimat*, Vol. 11, p. 141. (Originals preserved in the Archives of the National Museum in Prague, J. Puluj collection.)

As the newspaper report states, Puluj focused at the beginning of the lecture on the work on the cathode rays that preceded Röntgen's discovery (that he called a lucky coincidence). He mentioned above all his own experiments performed in the early 1880s by instruments of his own design. During the lecture, Puluj demonstrated how to photograph using "invisible cathode rays". In addition, he showed his previously (referring most probably to 1896) taken images that required more prolonged exposure: skeletons of animals, a child's broken forearm and a skeleton of a newborn baby. Copies of the images were hung on the walls of the auditorium so that those present could better view them. The report stated that all the photographs taken by Puluj using "invisible cathode rays" were photographically processed (and made in multiple copies) by the photographer H. Eckert. Eckert, who had graduated from the Prague Polytechnic School, was a reputable photographer, a member of the Photographic Society in Vienna, and he also collaborated, e.g. on photo documentation of F. Křížík's businesses [68].



Figure 14. Interior of Puluj's Physics laboratory at the DTH in 1896 (?). Various cathode tubes can be seen on the upper shelf of the cupboard. Over the desk, Puluj's X-ray pictures are exhibited. [Archives of the Czech Technical University in Prague, J. Puluj collection, published with permission.]

Prior to the public lecture at the DTH, Puluj promised to give a lecture on cathode rays with demonstrations also in the local German Science and Medical Association „Lotos“ (*Deutscher naturwissenschaftlich-medizinischer Verein für Böhmen „Lotos“*) [69]. Puluj became a regular member of this association immediately after his arrival in Prague in 1884; by the end of 1899, his wife also became a (supporting) member of the association. The association had an interdisciplinary character. Its active core consisted of teachers of Prague's German universities; E. Mach also was a member and its representative when he was Professor in Prague in 1867–1895. Occasional lecturing was one of the basic duties of the regular members. Puluj gave his first lecture there in 1886 “on two instruments designed for determining the mechanical equivalent of heat and thermal conductivity of mica leaf” [70]. Puluj gave the promised lecture at the Lotos association on 14 March 1896. The lecture entitled „*Ueber Kathodenstrahlen und ihre Transformation in die von Röntgen entdeckten unsichtbaren Strahlen*” was announced a day before in the newspaper *Prager Tagblatt*. The lecture again took place in the physics auditorium in the main building of the DTH in Husova Street. With respect to the limited capacity of the auditorium, it was only reserved for members of the association [71]. It is worth mentioning that on the same day, within a series of popular lectures organized by the Lotos association outside Prague, Joseph

Geitler, at that time E. Lecher's assistant in the Physics Institute of the German University of Prague, lectured on X-rays in Plzeň in West Bohemia [72].

Puluj returned to his routine work at the DTH, in which electrical engineering had already outweighed physics. The discovery of X-rays by Röntgen and the several weeks of his own intensive work on X-rays undoubtedly contributed to his increasing physical and mental exhaustion. At about the same time when W. C. Röntgen was awarded the first Nobel Prize in Physics in 1901, J. Puluj was bestowed the new Chair of Electrotechnical Engineering at the DTH (without, however, any increase in his salary).

## Acknowledgements

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  14. W. C. Röntgen (1845–1923) assumed the post of A. Kundt's assistant at the University of Strasbourg on 1 April 1872, and within a year he made his habilitation for physics there. From 1 May 1875, Röntgen was appointed professor of mathematics and physics at the Academy of Agriculture (*Landwirtschaftliche Akademie*) in Hohenheim by Stuttgart. He came back to the University in Strasbourg in autumn 1876 where he succeeded Emil Warburg in the Professorship of Physics. Cf. Fölsing, A. (1995). *Wilhelm Conrad Röntgen. Aufbruch ins Innere der Materie*, pp. 333–334. München u. Wien: Carl Hanser Verlag (Zeittafel).
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  19. In his second communication on the "radiant matter of electrodes", J. Puluj summarised his views as follows (quoted in German): "Nach meiner Ansicht bestehen die von der negativen Elektrode in gasverdünnten Raume ausgehenden Strahlen nicht aus elektrischen Teilchen des zurückgebliebenen Gases, das nach Crookes in einem eigenthümlichen 'übergasigen' Zustand, dem vierten Aggregatzustand, sich befinden soll, sondern aus Körpertheilchen, welche durch den elektrischen Strom von der negativen Elektrode losgerissen, mit statischer negativen Elektrizität geladen und mit sehr grosser Geschwindigkeit in den gasverdünnten Raum fortgeschleudert werden. Damit kein Zweifel über die Natur der Kathodenstrahlen obwalte, nenne ich sie 'strahlende Elektrodenmaterie'". Cf. Puluj, J. (1881). "Strahlende Elektrodenmaterie. II. Abhandlung." *Sitzungsberichte d. AdW*, Vol. 82, p. 402.
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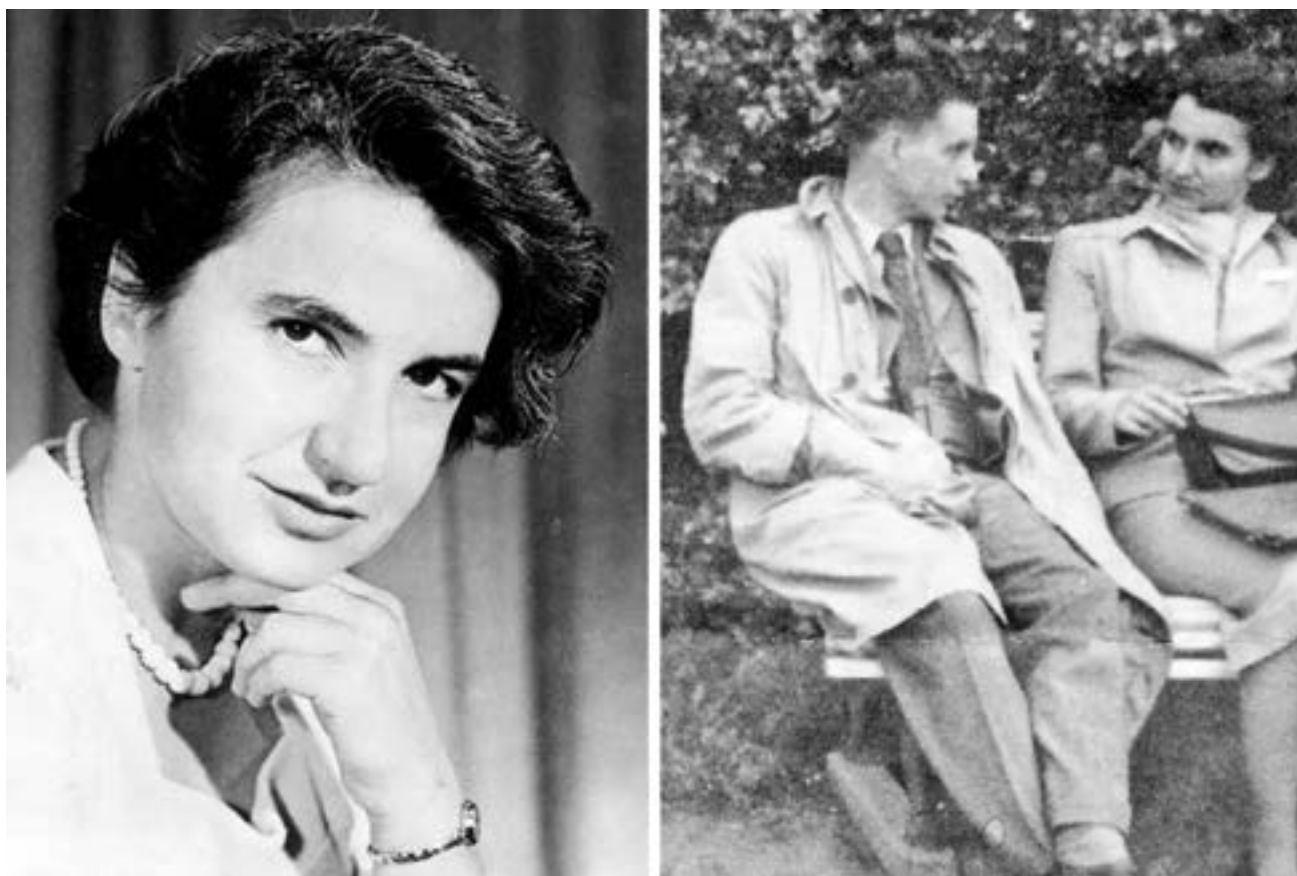
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## HISTORY OF CRYSTALLOGRAPHY

# Rosalind Franklin 1920-1958

Elspeth F. Garman



Rosalind Franklin. Left: photo credit Pictorial Press Ltd / Alamy Stock Photo. Right: with Durward Cruickshank on a visit to Uppsala Castle during the Second IUCr Congress in Stockholm, Sweden, in 1951; contributed to the IUCr photo gallery by the estate of D. W. J. Cruickshank.

This year marks the 100th anniversary of the birth on 25th July 1920 of Rosalind Franklin, whose pivotal contribution to the discovery of the structure of DNA has been increasingly recognised since her untimely death from ovarian cancer at the age of 37 in 1958. There is now a general consensus that, if she had lived longer, she would have 'deserved' to be among the three awardees of the 1962 Nobel Prize in Physiology or Medicine. Each Nobel Prize can be bestowed on a maximum of three people and is never awarded posthumously. The Prize went to Francis Crick, James Watson, and Maurice Wilkins 'for their discoveries concerning the molecular structure of nucleic acids and its significance for information transfer in living material'. However, the debate still rages on as to whether, given the climate at the time, she would have been included instead of one of the three awardees.

A realisation of the vital importance of Rosalind Franklin's work has only gradually gained traction. Very belatedly, her name is becoming much better known and the public are being disabused of the veracity of the scurrilous comments about her in the famous book, *The Double Helix*, an account written by James Watson of the DNA structure discovery. It was not until 1999 that Watson finally said that '... the Franklin photograph was the key event ...' i.e. he at last admitted that Photo 51 was absolutely essential information for construction of the model.

So what was Photo 51 and why was it so important? I will try to give a brief and distilled account below. Much has already been written on the subject and the interested reader is referred to more detailed descriptions listed at the end of this piece.

Rosalind Franklin (RF) was born in London to Muriel and Ellis Franklin. Her brother, David, was one year older, and she had two younger brothers (Colin and Roland) and a younger sister, Jenifer. Rosalind showed early signs of wanting to challenge herself: her Aunt Mamie Bentwich, on holiday with the Franklin family on the Cornish coast in 1926, wrote to her husband that 'Rosalind is alarmingly clever – she spends all her time doing arithmetic for pleasure and invariably gets her sums right'. She attended boarding school at Bexhill-on-sea from age 9 to 11, and then in January 1932 went as a daygirl to St Paul's Girls' School, where according to the 'High Mistress', Miss Ethel Strudwick, 'every girl is being prepared for a career. The High Mistress considers that no woman has a right to exist who does not live a useful life ... look beyond marriage as your goal'. Rosalind shone at both sport and in her studies, and her mother wrote: 'All her life, Rosalind knew exactly where she was going, and at 16 she took science as her subject'.

In the summer of 1938, Rosalind visited Paris and so began her lifelong love of France and all things French, including the people. She perfected her spoken French and developed a passion for hiking and mountains. On her return, from 1938 to 1941 she went to Newnham College, Cambridge University, to study Natural Sciences with a focus on Chemistry, at which she excelled. She thought nothing of riding her bike home to London at the end of term, a distance of 65 miles! In the 1st year exams ('Prelims') she came 2nd in the whole year group. She studied 'Crystal Physics' in the autumn of her second year, and became familiar with crystallographic space groups and various crystal forms. Her final year research project went well and was supervised by Fred Dainton, a photo- and polymer chemist. However, she did not actually 'graduate' until 1948 since degrees were not awarded to women by Cambridge until 1947, whereas in Oxford they did so from 1921 onwards.

Newnham gave Rosalind a 4th year research scholarship, and from 1941 to 1942 her supervisor was Ronald Norrish who was 'bad-tempered' and given to 'autocratic treatment of juniors'. She was housed in a claustrophobic small dark workspace and allocated an impossible project on the polymerisation of formic acid and acetaldehyde. She did not enjoy the experience and did not respect her supervisor, who said that 'confrontation when cornered was her tactic'. Generally it was known that she 'didn't suffer fools gladly'.

In 1942 she registered for a Cambridge PhD but worked as an 'Assistant Research Officer' in Kingston for the British Coal Utilisation Research Association (BCURA) on the permeability and shrinkage of coals to gas (helium) and in water as a function of temperature. Activated charcoal was used in gas masks to very good effect in WW1, saving thousands of lives, and an understanding of the effect of gas on different coal types could be applied to make more effective masks and also had other industrial applications. During this period of WWII, RF acted as an air-raid warden. Her 1945 PhD thesis was entitled *The physical chemistry of solid organic colloids with special reference to coal and related materials* and in 1946 she published her first (of 37) peer-reviewed papers [*Thermal expansion of coals and carbonised coals*, Bangham & Franklin (1946)], putting forward the hypothesis of 'molecular sieves' to explain the variation in porosity of various types of coal.

For her first postdoctoral position she happily returned to her beloved France and worked from 1947 to 1951 in Paris, studying the crystallography of coal and graphite under Jacques Mering (who had been trained at the Royal Institution by W. H. Bragg) at the Laboratoire Central des Services Chimiques de l'Etat (a Government Laboratory). She was paid £5/week and in turn paid £3/week rent, she became 'unEnglished' and she felt that at work 'women engaged as equals'. She carried out powder diffraction on amorphous solids with monochromatic X-rays and identified the carbons that turned into graphite when heated to 3000 °C ('graphitising carbon') and those that did not ('non-graphitising carbon', a rigid finely porous mass). She worked closely with Vittorio Luzzati from Argentina, becoming an expert in the

crystallography of such samples. Five papers were published in 1948 on various aspects of her coal studies and her first letter to *Nature* came out in 1950, *On the influence of bonding electrons on the scattering of X-rays by carbon*.

At the beginning of 1951 she moved back to England, taking up a three-year Turner and Newall Fellowship at King's College, London to work under John Randall on proteins in solution and changes in their structure when they are heated or dehydrated, causing them to denature. She was concerned about her lack of knowledge in the new research field: 'I am, of course, most ignorant about all things biological, but I imagine most X-ray people start that way' (as an ex-nuclear physicist, I can sympathise with this view!). However, just before she arrived, Randall suddenly changed her project to the investigation of some DNA fibres that Maurice Wilkins, also working at King's with Randall, had obtained in May 1950 from Rudolf Signer in Berne. Randall wrote to her that '... This means that as far as the experimental X-ray effort is concerned there will be at the moment only yourself and Gosling ...'. Unfortunately, Randall neglected to tell Wilkins about this new arrangement, even though Wilkins and Ray Gosling (a research student supervised by Wilkins) had already obtained good X-ray diffraction patterns from the DNA fibres. This set the scene for difficulties between Wilkins and RF which quickly escalated over the first six months of 1951 while RF was building new equipment to control the humidity of the DNA fibres using hydrogen gas bubbling through different salts.

The situation between RF and Wilkins reached such an impasse that, in October 1951, Randall directed RF and Gosling to work on the A (dehydrated) form of DNA using the fibres from Signer and the best X-ray camera, and Wilkins to work on the B (hydrated) form with some other fibres that did not crystallize well. Photo 51 was taken by RF and Gosling in May 1952 using a micro-camera, and was the clearest photo yet obtained, but it was of the B form (92% humidity) on which they were not supposed to be working, so it was put away. The X-ray generator was a prototype fine-focus device built at Birkbeck by Werner Ehrenberg and Walter Spear which had been given to Wilkins and Gosling, but was then used solely by RF and Gosling.

By January 1953, Gosling wanted to finish his thesis and he showed Photo 51 to Wilkins, who in turn, unknown to RF, showed it to James Watson (JW) when he visited King's from Cambridge. JW was working there with Francis Crick (FC) on building a model of DNA with newly obtained permission from W. L. Bragg, the head of the Cavendish Laboratory. Bragg had previously banned them from pursuing further DNA modelling following an embarrassing incorrect model (helical with the bases on the outside) that they had trumpeted the year before. From the 1938 X-ray diffraction work in Leeds of William Astbury and the crystallographer Florence Bell on DNA from calf thymus, it was known that the DNA structure repeated every 27 Å and that the bases were stacked 3.4 Å apart and lay flat, but they did not discover that it formed a helix. Wilkins had requested a physicist colleague at King's, Alex Stokes, to calculate the diffraction pattern arising from a helical structure, and as a result Stokes was the first to show that DNA was probably helical. However, it was not known how many chains constituted the helix. Wilkins also told JW the RF experimental helical parameter values: a 34.4 Å repeat with the bases stacked 3.4 Å apart. Meanwhile, RF found that someone had tampered with her laboratory notebooks, and her longing to leave King's intensified.



Photo 51. Courtesy Ava Helen and Linus Pauling Papers, Oregon State University Libraries.

On the 9th February 1953, FC and JW were shown RF's December 1952 MRC Review Committee report by Max Perutz. This report was not marked confidential, but the results in it were unpublished. It gave the space group of the B form DNA as face-centred monoclinic (C2), and specified the unit-cell dimensions and angles. FC realised that this space group gave a vital clue as to the structure, because it meant that the DNA looked the same both ways up, so that it must consist of two antiparallel helical chains, not three chains as some had postulated. Erwin Chargaff had already found that in DNA the number of adenines (A) + guanines (G) was equal to the number of thymines (T) + cytosines (C), and also that the numbers of A and T were the same, as well as the numbers of G and C. Another critical piece of the puzzle was solved when Jerry Donohue, a postdoc who shared an office with the model builders and was watching them at work, suggested that the bases were the keto and not the enol forms that they had been trying to use.

By 7th March 1953, FC and JW had built a model that seemed to fit with all the known information. Each purine (A,G) was paired with a pyrimidine (T,C) across the inside of the double helix formed by two antiparallel carbon-phosphate backbones. Wilkins went to see it on 12th March and told everyone at King's about it on his return. RF was about to leave King's where she was miserable and felt she could no longer work in the same environment as Wilkins. She and Gosling had already sent off two papers on the structure of the A form and had almost finished one on the B form, of which she was very near to having the structure. How much RF ever knew about which of her results were shown to whom and when they were shown, remains a matter of current debate.

In mid-March 1953, funded by the Agricultural Research Council (ARC) as a senior scientist, RF moved to Birkbeck College where John D. Bernal ('Sage') provided a supportive and happy environment for her

new research group working on virus structure. Her office was on the 5th floor of a bombdamaged house: 'I swapped a palace for a slum'. The X-ray laboratory was in the basement and leaked, so that an umbrella was required during experiments! Here she worked on RNA and on Tobacco Mosaic Virus (TMV), the first virus to be discovered in 1892. Bernal thought the world of her and supported/protected her, calling her a 'brilliant experimentalist' and writing after her death: 'As a scientist, Miss Franklin was distinguished by extreme clarity and perfection in everything she undertook. Her photographs are among the most beautiful X-ray photographs of any substance ever taken'. Aaron Klug, a future Nobel Prize winner (Chemistry in 1962) who had come from South Africa to do his PhD at Cambridge and then won a Fellowship to work under Bernal, met Rosalind and transferred to study viruses in collaboration with her. By 1955 her group consisted of three postgraduate students: James Watt, John Finch and Ken Holmes, and also Don Caspar (who first coined the phrase 'structural biology') from the USA on a Fellowship. From interpreting fibre diffraction patterns, and using multiple isomorphous replacement methods, RF hypothesised that all TMVs were the same length and had a hollow core with the RNA deeply embedded in protein units and coiled inside (published in *Nature* in 1955). Together, her group determined the first virus structure, showing that indeed the 50 MDa TMV had a diameter of 150 Å with the RNA coiled round a hollow inside. The first model had been made using 288 bicycle handle bar grips! However, Norman Pirie, a plant virus physiologist at the Rothamstead Experimental Station in Hertfordshire, fundamentally disagreed with this result, and as a result stopped sending her virus material on which to work, so she and Klug then had to prepare their own viruses.

The structure allowed the TMV infection process to be understood, and a famous model of the virus was displayed at the 1958 World Trade Fair (Expo1958) in Brussels. To aid the highly calculation-intensive interpretation of the X-ray diffraction patterns, a 'computer' was employed: she was called Mrs Cratchby! Results on pea streak, potato, turnip, tomato and cucumber viruses were reported in seven papers in 1956 and six in 1957. The group then expanded their focus from plant viruses and started work on the Polio virus.

During her time at Birkbeck, RF went on a couple of two month-long tours of America (1954 and 1956) and she thoroughly enjoyed the recognition and respect she was given there. It was on the second of these that she experienced pains in her abdomen which were the first sign of the illness that would cut her life so short.

Rosalind has received much belated posthumous recognition that sadly she did not live to witness, with at least 39 buildings or projects named after her, including the 2019 European Space Agency's ExoMars rover. Newnham College, Cambridge have named a student residential building, there is a blue plaque on the house in London where she lived from 1951 to 1958, in 2000 King's College named their new Dental Education Centre the Franklin-Wilkins Building (a painfully ironic coupling of names), and in 2018 the Rosalind Franklin Institute was launched at the Harwell Campus in Oxfordshire as an autonomous medical research centre under the joint venture of 10 universities, and funded by UK Research and Innovation. Beyond the UK, among others there is the Rosalind Franklin University of Medicine and Science in Chicago.

There has been much controversy regarding Franklin's contribution to the unravelling of the structure of DNA. A balanced account was given in *Nature* by Aaron Klug in 1968: *Dr Klug discusses Dr Franklin's contribution to the discovery of the structure of DNA in the light of accounts given by Professor Watson in his book The Double Helix and by Dr Hamilton in a recent article in Nature.*

In 2017, under the Planning Act of 1990, Historic England listed her tomb as of 'special architectural or historic interest', with the official description (which sums up the her scientific impact very well): 'the tomb commemorates the life and achievements of Rosalind Franklin, a scientist of exceptional distinction, whose pioneering work helped lay the foundations of molecular biology; Franklin's X-ray observation of DNA contributed to the discovery of its helical structure'.

Notably, last year the University of Portsmouth announced that on 2nd September it was changing the name of its James Watson Halls to Rosalind Franklin Halls. Perhaps this act shows in microcosm the growing appreciation of the impact of Franklin's life and work, somewhat redressing the balance in the previous mis-allocation of credit.

*Further reading: Rosalind Franklin, The Dark Lady of DNA* by Brenda Maddox (2002); *My Sister Rosalind Franklin* by Jenifer Glynn (2012).

## Acknowledgements

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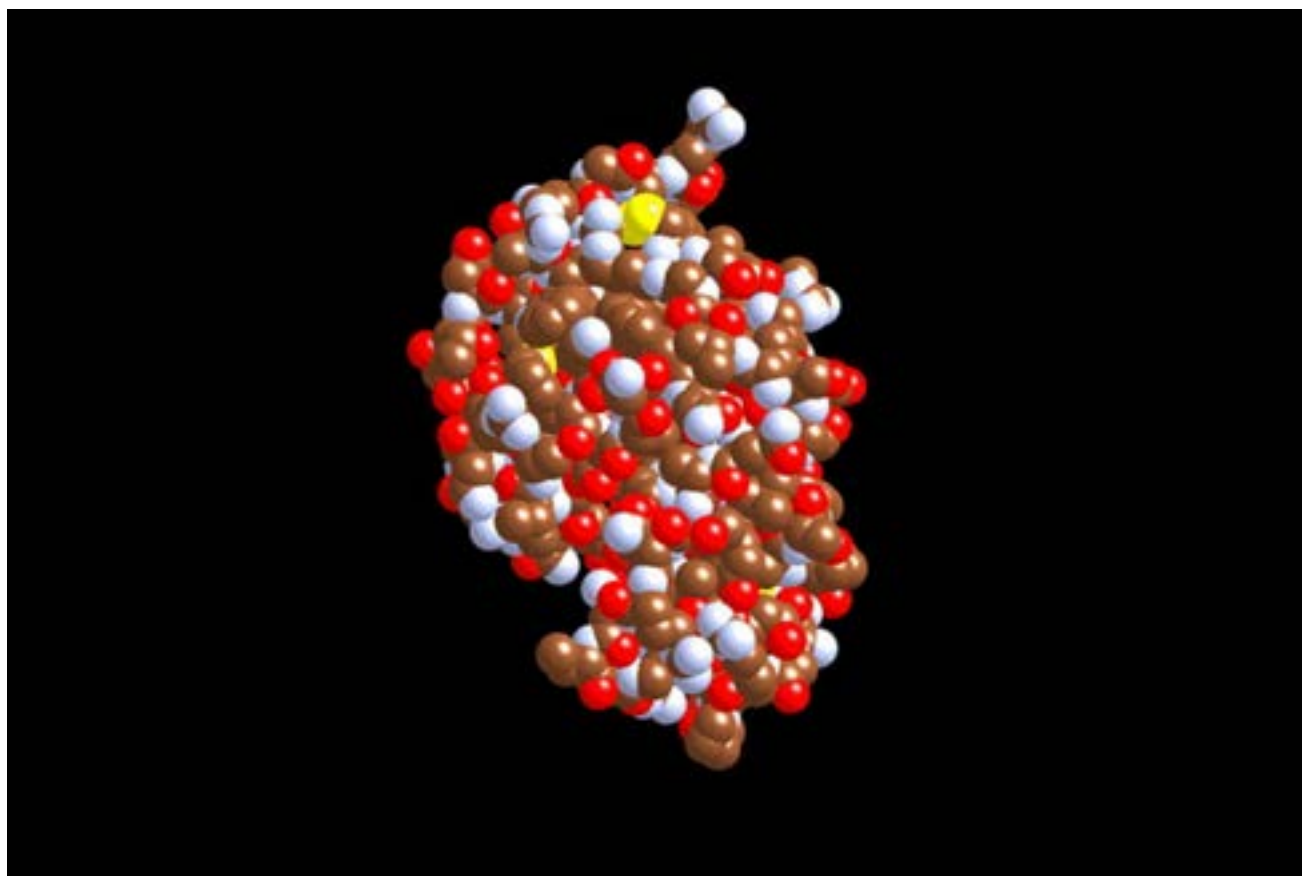
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## HISTORY OF CRYSTALLOGRAPHY

# Squaring the blue four-circle

Jon Cooper



From the 1960s to the 1980s, X-ray data collection in many laboratories in the UK relied on the Hilger & Watts diffractometer, which some may well remember using, fondly, or perhaps not. Collecting macromolecular data often took several months, during which power dips and other electrical instabilities, cooling-system floods, crystal slippage, radiation decay and capillary breakage, as well as helium-gas and paper-tape shortages, all took their toll on your sanity. In the days before automatic indexing, usually the hardest part was determining the orientation matrix. This involved pre-aligning the crystal using a precession camera, transferring it to the diffractometer and then locating a number of strong reflections, usually axial, orthogonal and of known index. Arguably, it was good training for coronavirus stress. As these machines have long since gone to the great resting place for diffractometers, it seemed interesting to catalogue their history, and last year I committed some details to a [Wikipedia page](#) and a [blog](#) (referenced therein), which have many more details and historical documents than would fit here.

Hilger & Watts was based in several locations in the London, UK, area but diffractometer production centred on the Camden Town factory, which was the largest employer in the area. The company had a long history, dating back to the middle of the 19th century, specialising in the production of scientific instruments (mostly optical), many of which were commissioned in large numbers for military purposes during both world wars. They also supplied space-flight instruments to NASA.

The first diffractometer in the UK (or the world even!) was actually the ionisation spectrometer of W. H. Bragg in 1912–1913, but in the intervening years, X-ray data were mainly collected by film techniques

before the advent of computers and more modern instrumentation. The first X-ray diffraction instrument to be produced by Hilger & Watts was known as the linear diffractometer. This had been developed by Uli Arndt and David Phillips [1] at the Royal Institution (RI) in London (see [here](#) for a full description). In those pre-internet days, the setting angles for each reflection had to be calculated using the [EDSAC computer](#) in Cambridge. To overcome this inconvenience, the team set about developing an analogue method for determining these angles involving protractors and a scale drawing of the crystal lattice. Quoting from Uli Arndt's autobiography (Arndt, 2006) [2], "David remarked to me one Friday what a pity it was that the rotations of the protractors could not be linked directly to those of the crystal shafts". Arndt then describes how the following weekend, he borrowed a Meccano set from the son of a sailing friend to try out possible linkages for just such an automatic diffractometer. Having established the feasibility from such humble beginnings, the subsequent design and prototyping took another two years. This was complicated by the fact that Arndt had to spend a year on sabbatical in Wisconsin, communicating with the RI team by transatlantic mail. Following his return to London, a prototype of the machine was exhibited at the Physical Society. This was eventually manufactured under licence by Hilger & Watts, who went on to sell around 100 of these instruments worldwide, as the Y190 model (Fig. 1), in the late 1950s to the early 1960s. In 1963 (a year of some biological significance for the author), the RI team adapted the linear diffractometer so that it could effectively measure three reflections at a time. This proved to be crucial for the first structure determination of an enzyme, lysozyme, which followed two years later.

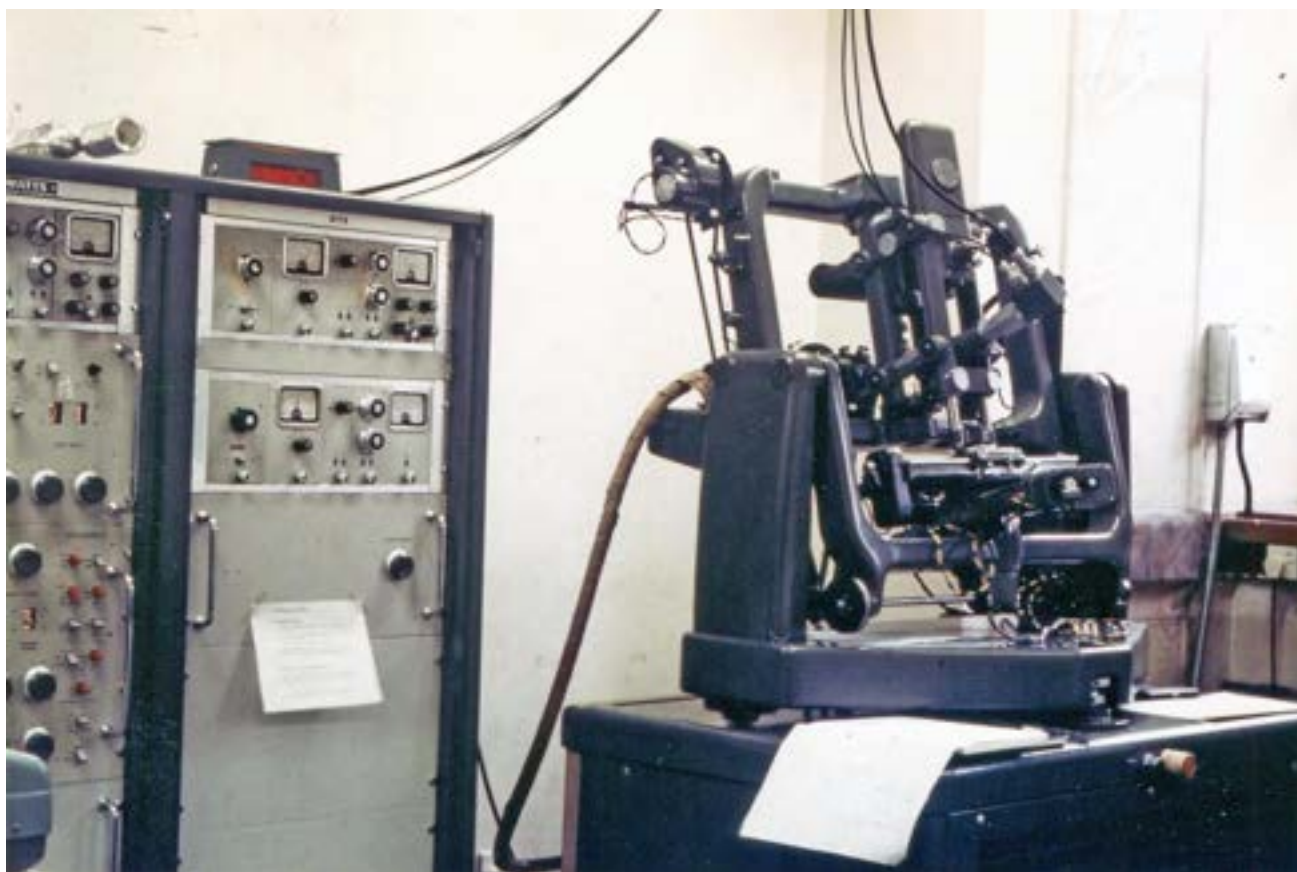


Figure 1. A Hilger & Watts Y190 linear X-ray diffractometer.

By the late 1950s it was clear that the instrument of the future would be the four-circle diffractometer. Developments in computer control of machine tools would mean that, before long, each diffractometer would have its own digital computer for controlling data collection, and the built-in analogue device was to become a thing of the past. Around this time, Arndt had realised that the relatively well-funded field of neutron diffraction had not invested significantly in the development of automatic diffractometers. This led to a collaboration with B. T. M. (Terry) Willis at the Atomic Energy Research Establishment, Harwell, where "there was a large staff of engineers... who were somewhat under-employed". Their considerable

instrument-design skills led to the rapid development of the world's first automated four-circle diffractometer, which, again, was manufactured under licence by Hilger & Watts, initially as the Y230 model, but later in greater numbers as the Y290 (Figs. 2–4). The very early machines used a Ferranti computer for controlling the instrument and “gave a lead of at least two years over any other similar instrument on the market”. However, there were reliability issues with the computer control, and this led Hilger & Watts to develop improved electronics in partnership with the University of Manchester, where both David B. G. Edwards (Computer Science) and Owen S. Mills (Chemistry) were involved. On their advice, the cheaper and more compact DEC PDP-8 (right-hand side of Fig. 3) became the computer of choice, and the resulting improved models of the diffractometer were given an unforgettable pale blue colour. One of the production engineers claimed that the electronic research department in Camden had PDP-8 No. 4 from DEC, so Hilger & Watts was likely to have been one of DEC's very first PDP-8 customers. The book *Single Crystal Diffractometry* (Arndt & Willis, 1966) [3] has photographs and other excellent diagrams showing the construction of both diffractometers, and is available online. For commercial production of the Y290, Hilger & Watts would go on to win two Queen's Awards to Industry, the first for Services to Export in 1966 and the second for Technological Achievement in 1968.

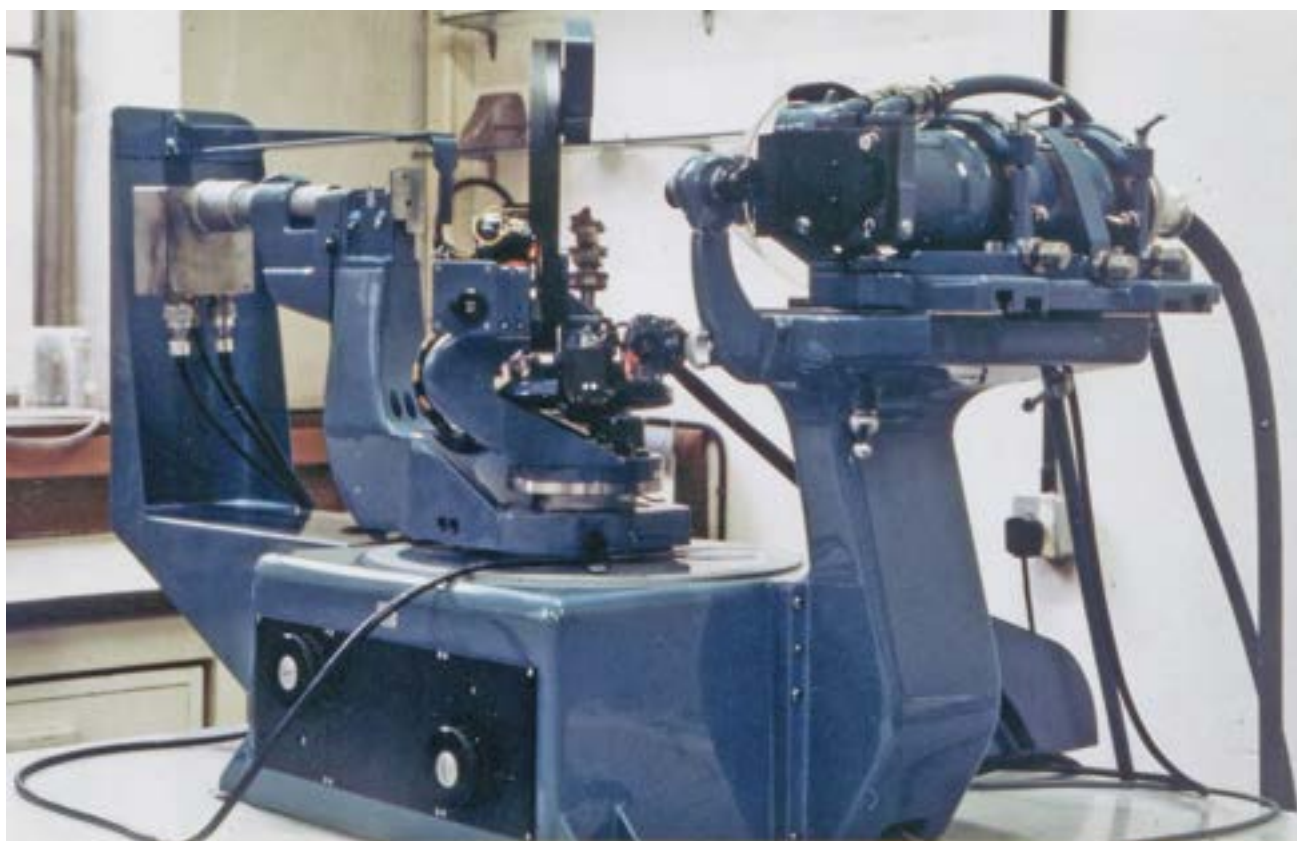


Figure 2. The unmistakably elegant style of the Hilger & Watts Y290 four-circle diffractometer.

The Y290 diffractometer had an optical motor-positioning system based on moiré fringes that were recorded by photocells – sometimes the user would find that after checking the centring of their crystal under a bright light and forgetting to turn it off, the diffractometer motors would then completely lose their settings and the resulting data would be useless. The X-ray data were usually written to paper tape by teletype and, by the mid-1980s, users were struggling to find computer facilities that could still read their tapes for downstream analysis. In the 1970s, David Phillips, by now at the University of Oxford, developed an interesting five-circle version of the machine that could measure up to five X-ray reflections in each scan *via* the addition of a tiltable linear array of five counters to the detector arm.

Some recollections from a former Hilger & Watts engineer, Derek Coggrave: “I started work there around 1962/63. The reason for employing me was that the production of the linear diffractometer was due to start, and I would be assigned to test and install it. The linear diffractometer didn't last long because it

was superseded by the four-circle geometry. On my first visit to Japan to install a four-circle there was no instruction manual for the instrument. The customer complained, and the agent became very agitated. I was there for about seven weeks, and so wrote a manual by hand for the user in the evenings and weekends. The agent had the thing typed, and the drawings copied, and this was given to the customer before I left. Later this was then used to produce the official version. It is amazing that in just over 50 years we have advanced from the PDP-8 with 4K of memory and costing around £8K (my house only cost £5.7K at the time)."



Figure 3. The control electronics for a Y290 diffractometer, including the PDP-8 minicomputer.

"There was an interesting letter in the *Financial Times* newspaper recently about the RI where David Phillips was mentioned. That struck a note with me because I went there several times to install and service a four-circle for Professor Phillips. After a couple of years, the instrument was moved to Oxford when he moved there. It was around 1965/6 when I went to the RI with Len Wood for the installation. One difficulty was the hydraulic lifts and getting heavy items to the upper floors. The transformer, which weighed half a ton, made the lift sink as soon as the first two wheels reached the lift floor. A toss of a coin determined who stayed in the lift, and with help, the transformer was run into the lift as fast as we could push it. As my colleague and the transformer sank from view, we climbed the stairs and waited. Thankfully, after a few minutes, he, the lift and transformer reappeared."

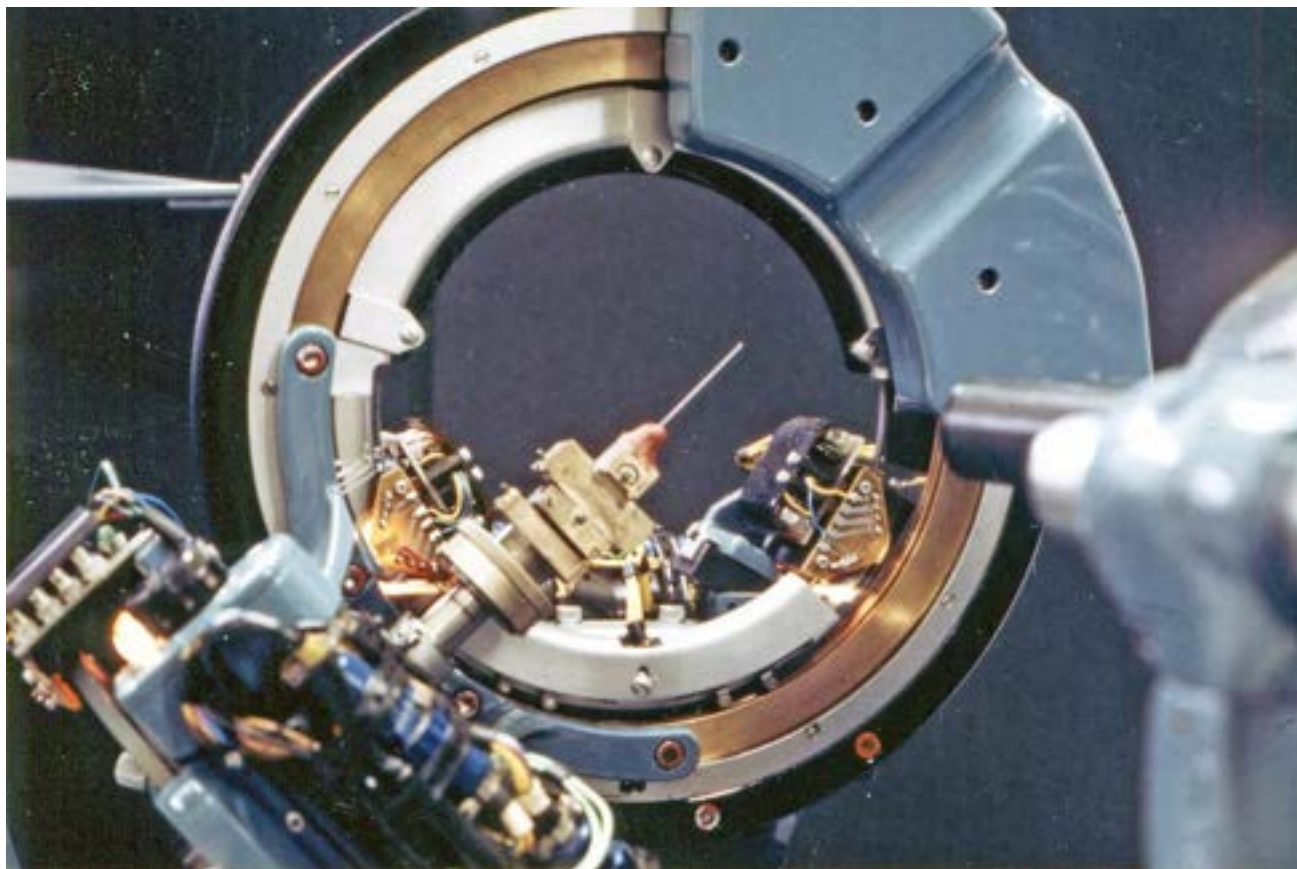


Figure 4. Close-up of the crystal on a Y290 showing the distinctive chi-circle of the Eulerian cradle.

"Later, another colleague servicing the instruments worked until around 10 pm. Turning the lab lights off, he found the rest of the building in darkness. He groped his way to the front door – locked and bolted. Daunted by a night alone in the RI, he climbed upwards, looking for help. Seeing light around the edge of a door, he knocked loudly, expecting a porter. However, the flat was occupied by Sir Lawrence Bragg [the director of the RI] who appeared in his dressing-gown, produced a set of keys and allowed my colleague's exit."

"Prior to that, probably around 1962/63, I had quite often visited the department in Oxford run by Dorothy Hodgkin. I installed her linear diffractometer with Len Woods. We were always encouraged to entertain the customer, so Len and myself took Dorothy to the Royal Oxford Hotel for lunch a couple of times. We were quite right wing in our politics, and she was very left, although we didn't know it at the time. However, she was always extremely polite and very gracious. In comparison, on one occasion, when we were in the RI, the telephone in the lab rang, so I answered it. Before I could ask who was calling, Dr Arndt rushed in yelling at me for picking the receiver up. Professor Phillips, on the other hand, was always very polite but was inclined to ask rather quizzical and penetrating questions, testing whether one was up to the job. But, he was always quite content provided things were done to his satisfaction."

Some comments from Lindsay Sawyer (Edinburgh): "The pale blue Y290 four-circle was their Mark 2 version. Physics in Edinburgh had a Mark 1 (same colour as the linear diffractometer), which relied on a huge box of Ferranti electronics and had gratings which were too coarse for positioning the circles. Positioning to 1/10 of a fringe was unreliable at the time, and most of the folk in the lab became expert at keeping the thing going to finish data collection. There was also a peg-legged night watchman who was trained to do the night shift. The Edinburgh (ex-RI) linear diffractometer had three counters, so if you had an orthogonal space group (like me) you could collect three layers at the same time. Eddie Komorowski collected 6 Å triclinic data on it, one layer at a time! The output of the linear diffractometer was 5-hole paper tape, on the four-circle, 8-hole. I may still have some rolls somewhere. Happy days!"

More from Derek Coggrave: “In July 1968 Hilger & Watts was taken over by Rank Precision Industries – Rank had a large cash flow from their vast Xerox copier production. However, selling scientific instruments tends to be a one-off process. Another issue was that Hilger & Watts had growing problems of its own. Many of the managerial positions were occupied by long-serving employees who had spent decades engaged in the production of optical instruments and had no knowledge of electronics or the science involved. Secondly, the catalogue of instruments it manufactured was, as far as I remember, around 90. Many only sold a few each year: no doubt uneconomic. The main housing of these devices was often a casting, and it was always difficult to reach the adjustment mechanisms. I can remember using dental mirrors, torches, dental probes to hook springs, and right-angled screwdrivers.”

Rank chose to move their Camden operation to Thanet in Kent, to an iconic 1960s factory that was large enough to locate all their functions on one site. “The prime purpose at Hilger & Watts was the design and building of scientific instruments for a limited market, which because of technological developments, was subject to sudden change and required a process of constant redesign and upgrades. A lot of investment was required in personnel and equipment, which presumably was not forthcoming, so the company fell apart. Rank lost patience. Oxford Instruments, which at the time was a competitor of Hilger & Watts, has thrived until today. So, I believe the opportunity was there, but the management skills and the will to succeed was wanting.” Thus, over time almost everything was sold off or wound down, although the successful company Hilger Crystals still operates at the Thanet site today.

It would be interesting to hear from anyone with further recollections of these instruments.

## Acknowledgement

I am very grateful to Dave Watkin (University of Oxford) for the provision of the photographs.

## Notes and references

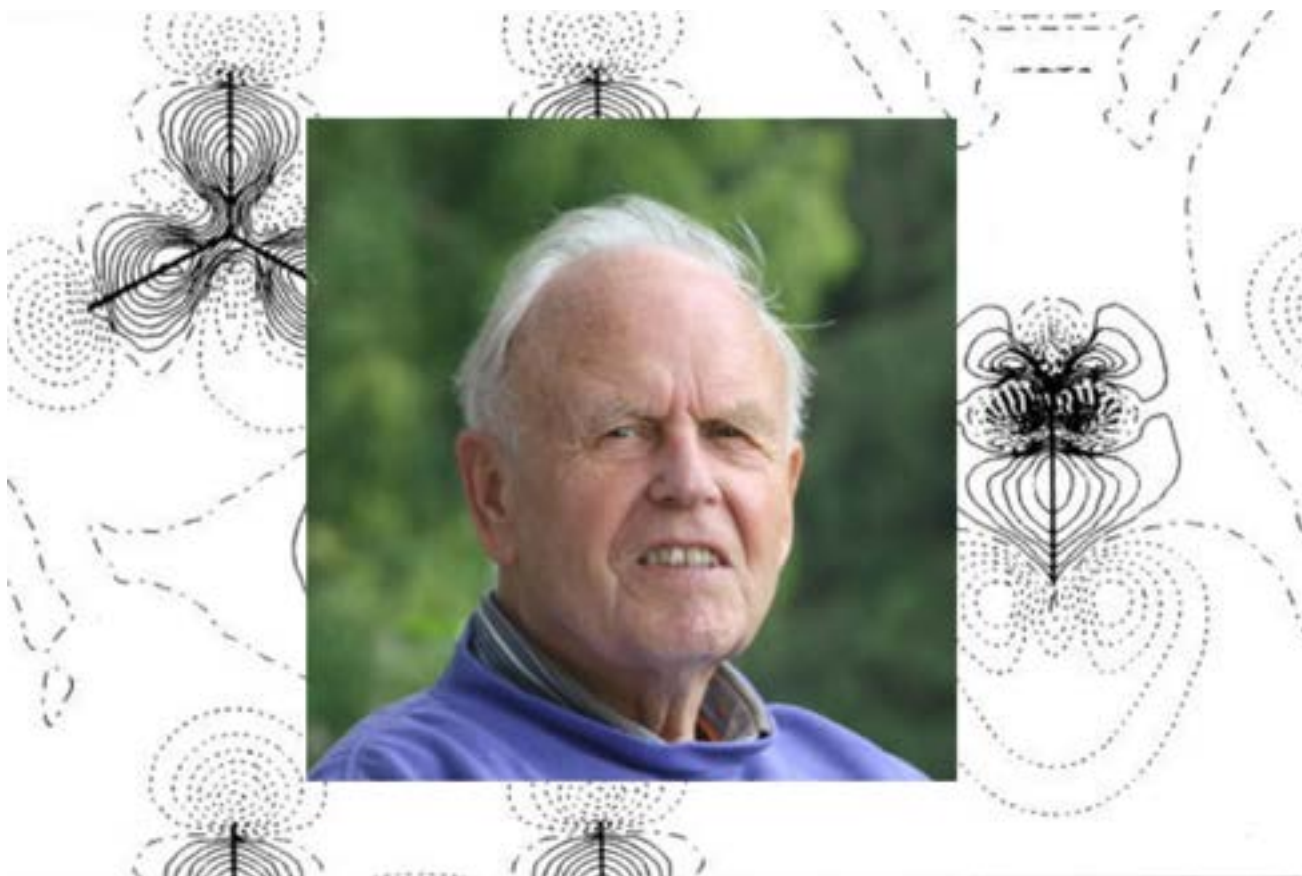
- [1] You can read about [Uli Arndt](#) and [David Phillips](#) in the *Biographical Memoirs of the Royal Society*.
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## OBITUARY

**Dirk Feil (1933–2020)**

Grant MossBarbara Moss



Dick Feil (photo credit: M. Luizink) against a background of an *Acta Cryst. B* figure. Dick was Chair of the IUCr Commission on Charge, Spin and Momentum Densities (now Quantum Crystallography) from 1993 to 1996, having been a member since 1987.

Dirk (Dick) Feil passed away on 22 April 2020 at the age of 87. Dick Feil was born in Zeist, The Netherlands, in 1933. Married in 1958, Dick and his young wife Annet set out to see something of the world. In 1959, despite misgivings about the apartheid regime, Dick took up a lectureship at Cape Town University in South Africa, which itself was opposed to apartheid. The experience was, as Dick recalled, formative. Dick Feil received his PhD in 1961 under Professor J. M. Bijvoet.

In 1964 the family returned to Enschede where, three months after the creation of the TH Twente, Dick assumed the position of lecturer in physical chemistry. Two years later he was appointed professor in chemical physics. Thus began an association that was to span 55 years. In 1998 Dick was appointed Professor Emeritus, the same year in which he retired. Post retirement Dick maintained an active interest and participation in university life.

Dick's research largely focused on the interaction between quantum chemistry and crystallography. He loved the beauty of theoretical formulations, Maxwells' equations being a particular delight. He was one of the early group of crystallographers to use experimental X-ray diffraction data to determine charge density and electrostatic potentials, and to compare them with theoretical calculations.

Dick loved to teach. A gifted and enthusiastic teacher he taught and mentored many students over a long period. His lab was always welcoming to visitors, readily accepting them as colleagues and oftentimes providing access to computing and other resources. He was actively involved in the IUCr, the Dutch Crystallographic Society (NKV) and in the inception of the ECA in 1972. He was also an excellent speaker and gave many memorable talks at various conferences and meetings.

Beyond his scientific and lecturing duties Dick was integral to the creation of the Committee for International Cooperation and Assistance Twente (CICA-Twente), resulting in projects in Bandung, Nigeria and Zambia. In 1975 he initiated a group dedicated to development studies, the Department of Developmental Studies (VOK).

Outside university life, Dick was a keen camper and sailor, vacations often spent with family on the many lakes and waterways that span The Netherlands. Active in his local community, Dick was a passionate advocate for the environment and equality.

Dick was resolute in his conviction that dialogue and exchange of ideas were essential to understanding and overcoming differences, a conviction that he applied equally to both his scientific work and his championing of social issues.

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## OBITUARY

# Jean-Luc Ferrer (1964-2020)

Juan C. Fontecilla-CampsG rard Bricogne



## Biography

Jean-Luc Ferrer studied for his engineering degree from the prestigious Ecole Centrale Paris in 1987. After obtaining a Dipl me d'Etudes Approfondies (DEA) in Physical Chemistry from Universit  Paris VI, Jean-Luc Ferrer undertook a doctoral thesis in physics on the dynamics of the spectrum of a free-electron laser at the Bruy res-le-ch tel research centre of the CEA (Commissariat   l'Energie Atomique). This work enabled him to obtain a doctorate degree from Universit  Paris XI in 1990 (Iracane & Ferrer, 1991). At the end of the same year, Jean-Luc was recruited by the Direction des Science du Vivant (DSV) at the CEA in Grenoble and joined the Laboratoire de Cristallographie et Cristallog nese des Prot ines (LCCP). This laboratory, directed by one of us (JCF-C), had just been created by Michel Suscillon, Director of DSV, as part of the Protein 2000 program, to boost the development of structural biology in France. This initiative had been taken following the decision, in January 1989, to build the European Synchrotron Radiation Facility (ESRF) in Grenoble, that became operational in 1994. Once at the LCCP, Jean-Luc joined Michel Roth's team, whose mission was to build the Collaborative Research Group (CRG) French beamline BM02-D2AM. This beamline, 50% dedicated to protein crystallography, was especially designed to support X-ray multi- and single-wavelength anomalous dispersion (MAD and SAD) data collection (Ferrer *et al.*, 1998).

In 1992, the LCCP joined the Institut de Biologie Structurale (IBS), which resulted from the merging of the CEA's Protein 2000 program with its CNRS counterpart IMABIO. From the very start, Jean-Luc distinguished himself by his interest and skills in both the technical and the scientific aspects, sometimes

very complex, of beamline construction, and of the determination of protein crystal structures. Concerning the latter, he maintained a very fruitful collaboration with Professor Joseph P. Noel at the Salk Institute (La Jolla, USA), where he spent sabbatical leave in 1996 and 2005. Jean-Luc's work concentrated mainly on the structural biology of phenylpropanoid synthesis in plants: he co-authored 20 papers with Professor Noel on this and related subjects, that were published in prestigious international journals (Ferrer *et al.*, 1999).

After Michel Roth's retirement in 2001, Jean-Luc became head of the new CRG French beamline for protein crystallography BM30A-FIP (Ferrer, 2001). Initially intended as a national platform, the FIP line became over the years an essential tool for both the French and the international communities of protein crystallography. As before, but now with newly acquired responsibilities, Jean-Luc displayed a great deal of creativity, persistence and dynamism in exploiting the advantages of synchrotron radiation for protein crystallography. For example, he was able to anticipate the automation of crystallography beamlines and helped develop, at the ESRF, the first crystal sample loader based on a robotic arm. He also had the excellent idea of using the same robotic arm directly as a goniometer, which, for the first time, allowed X-ray data collection directly from crystals still in the drops of a crystallization plate. This ingenious development paved the way for the *in situ* crystallographic approach to ligand screening for drug design (Emamzadah *et al.*, 2009; le Maire *et al.*, 2011). In 2009, he ensured the technological transfer of these different innovations by founding the start-up biotechnology company NatX-ray.

Upon the restructuring of the IBS in 2011, Jean-Luc became head of the Synchrotron Group (GSY). As part of the program of the ESRF aimed at creating a new extremely bright X-ray source (EBS), Jean-Luc was able to secure funding towards the rebuilding and upgrading of the FIP beamline in order to continue providing the best possible service to the community. He followed this beamline upgrade project to its end, which will become BM07-FIP2 next autumn when the ESRF reopens.

## Achievements and legacy

Putting Jean-Luc Ferrer's achievements and legacy in a proper perspective requires winding the clock back two decades to the turn of the millennium, when structural genomics (SG) projects were being proposed and set into motion.

SG's aim of harvesting 'low-hanging fruit' (*i.e.* solving on a massive scale novel structures from easily grown crystals of a good size giving good diffraction) gave a tremendous boost to macromolecular crystallography (MX) infrastructure in general by underwriting coordinated, wide-ranging technological developments and automation efforts (including the necessary invention, standardization and commercialization of new equipment and consumables) that arguably would not have taken place without it, and that laid the technological foundations for the large-scale use of MX in structure-based drug discovery (SBDD). These efforts converged on the operational paradigm of a highly automated workflow structured around the notion of 'one complete dataset from one big crystal per loop in a big beam' that has underpinned much of high-throughput MX for SBDD to this day.

Prior to these initiatives, the numerous successive steps of a typical MX structure determination workflow were carried out by human operators, whose interventions involving decisions or computational tasks were supported by *interactive* software tools aimed at providing the investigator with the fullest possible exposure to the scientific context and the specific data pertaining to each structure. Indeed, the prevailing standard at the time was that of *interactivity through user-friendly GUIs* (*i.e.* in effect 'getting the human into the loop as directly and in as immersive a manner as possible'), in striking contrast to the imperative of complete automation (*i.e.* 'getting the human *out of* the loop' as comprehensively as possible) that was being promoted by SG and SBDD.

It is in this context that the French FIP beamline at the ESRF took up the challenge of providing its users with the facilities needed for supporting SG projects. By its very definition, SG involved the *de novo* determination by experimental (MAD) phasing of large numbers of structures, and the FIP team worked

creatively towards the systematic automation of all steps involved in both the alignment of the beamline and its operation in production mode (Roth *et al.*, 2002). Jean-Luc Ferrer took the lead in linking existing programs for the sequence of steps involved downstream of data collection itself into a very early instance of what would nowadays be called an auto-processing pipeline directly coupled to a beamline (Ferrer, 2001). The combined resource described by Roth *et al.* (2002) and Ferrer (2001) was at the time second to none in terms of automated MX. The team maintained its lead by next designing and deploying the CATS robotic sample changer (Ohana *et al.*, 2004) combining a commercial six-axis Stäubli robot with an original design for a combination of 'grips' and 'flipping grips' that enabled the robot to deliver crystals held in both screw-cap and magnetic base vials while keeping them in liquid nitrogen during the entire operation, thus minimizing ice formation and crystal damage. The design of these tools was patented, and the whole technology was transferred to the IRELEC company that went on to sell the CATS sample changer to numerous beamlines around the world (Jacquamet *et al.*, 2009).

In the first wave of automation to which the FIP developments just reviewed belong, the introduction of robots took place in a stepwise manner to provide an exact replacement for the very same actions that were previously performed by the human operator who was being systematically displaced. This was done without any kind of 'refactoring' (*i.e.* redistribution of functionalities) between those now robotized steps.

A major stumbling block remained in the automated workflows that resulted from this piecewise robotization, *viz.* the persistent need for human intervention (and hence an obligatory breakpoint in automation) in the harvesting of crystals out of their growth and/or soaking media and their conversion to diffraction-ready samples suitable for shipping to a synchrotron beamline, as well as in the subsequent centring in the X-ray beam of the crystal(s) in each sample to enable the collection of diffraction data to proceed. The early anticipation of these impediments had led McPherson to propose the idea of *in situ* crystallography as early as 2000 in a paper that was truly remarkable for its foresight (McPherson, 2000), and to demonstrate the feasibility of crystal detection by diffraction and limited data collection with a manually assembled crystallization plate on an in-house X-ray source.

Fresh from their experience in harnessing the power of the Stäubli robot to create the CATS sample changer, the FIP team were able to come up with a versatile practical implementation of McPherson's idea by a brilliant piece of 'thinking out of the box': while the other automated sample changers available at the time (Muchmore *et al.*, 2000; Cohen *et al.*, 2002) were based on four-axis robots, the availability of two extra axes on the Stäubli robot, combined with the use of an additional specific plate gripper, made it possible to program the robot arm in such a way as to endow it with the extra functionalities of both a centring stage and a goniometer axis. This then made it possible to screen for diffraction-quality crystals in crystallization drops by directly placing the 96-well crystallization plates themselves in an intentionally broadened synchrotron X-ray beam at room temperature; then using the advanced level of automation already available on the FIP beamline to scan the drops while recording their scattering pattern, looking for Bragg spots to detect whether they contained diffracting crystals; and finally performing a limited-range ( $\sim 30^\circ$ ) data collection by rotating a confirmed crystal in the beam.

This refactoring of functionalities shifted the role of robots in the automation process beyond the mere pointwise replacement of human interventions in the traditional human workflow. In the developers' own words (Jacquamet *et al.*, 2004) *this new system completely changes the design of the experimental setup by merging functions that were previously considered to be distinct*. This radical innovation was hailed as a 'seminal experiment' by the Diamond group (Aller *et al.*, 2015) who have since built a whole beamline (VMX-i) dedicated to *in situ* crystallography (Sanchez-Weatherby *et al.*, 2019). It also triggered the development of numerous new crystallization plates designed specifically to minimize the X-ray background and increase the rotation angle achievable in such *in situ* data collection experiments. Last but not least, this work marked a return to considering the potential of room-temperature diffraction experiments, with the increased speed of radiation damage being compensated by the avoidance of the complications and hazards of cryo-protection and by the use of multiple crystals.

Subsequent further elaborations of advanced modes of use of the Stäubli robot arm produced the G-Rob (for Goniometer Robot) (Heidari Khajepour *et al.*, 2013a,b), while the replacement of its sixth axis by an air-bearing goniometer axis to reduce the size of its sphere of confusion led to the RoboDiff (Nurizzo *et al.*, 2016), a key constituent of the ESRF MASSIF-1 fully automated beamline (Bowler *et al.*, 2015). Original developments based on the G-Rob continued at the FIP beamline with the design of a versatile Visualization Bench (Heidari Khajepour *et al.*, 2013a) and with the robotic harvesting of crystals it made possible (Heidari Khajepour *et al.*, 2013b), establishing a seamless connection between harvesting and data collection through a Crystal Listing capability that circumvented the need to subsequently centre each crystal in the conventional way. In a separate line of developments, the needs of SBDD for a human-free, fully automated means of exposing crystals to ligands prior to high-throughput compound screening by *in situ* diffraction were considered in a collaboration with a group in Montpellier, which led to the further innovation of 'dry soaking' (le Maire *et al.*, 2011; Gelin *et al.*, 2015). The technological know-how embodied in the G-Rob was patented in 2008 by the French CEA, with Jean-Luc Ferrer and his prominent FIP team member Lilian Jacquamet (deceased 2007) as joint inventors, and served as the basis for the creation in 2009 of the NatX-ray start-up company by Jean-Luc himself, Nathalie Larive and Xavier Vernède.

During the past few years, Jean-Luc and his team returned to the need to provide access to the capabilities of the FIP beamline to users located anywhere in the world in the most flexible and integrated manner possible, and were front runners in doing so through a web-based user interface. *WIFIP* was released in September 2015 and made FIP one of the very first beamlines offering the full remote control of an MX experiment through an interface based entirely on web services (Sallaz-Damaz & Ferrer, 2017).

It is worth noting that the totally different context of structural biology's pursuit of X-ray structure determination for ever more challenging targets (*i.e.* high-hanging fruit) led to parallel developments along the seemingly very different track of micro-crystallography, with its distinct emphasis on micro-beams, rapid sample micro-positioners and next-generation X-ray sources. These developments, however, shared the same requirement for automated handling of small and fragile crystals as those that led to *in situ* crystallography and for refactoring the diffraction experiment away from its traditional 'one complete dataset from one crystal' paradigm. This common concern resulted in a substantial degree of convergence between these distinct tracks: ultimately, the exciting recent development of serial crystallographic experiments on micro-crystals held on fixed-target micro-fabricated sample grids and shifted at great speeds by high-accuracy micro-positioners (see *e.g.* Roedig *et al.*, 2016; Cohen *et al.*, 2014; Baxter *et al.*, 2016; Owen *et al.*, 2017) can be seen as an extrapolation and miniaturization of the original experiment by the FIP group [see Fig. 7 in Jacquamet *et al.* (2004)], adapted to micro-crystals and to next-generation X-ray sources.

In summary, the last two decades have seen strongly innovative and distinguished contributions from Jean-Luc Ferrer and his FIP team. In spite of the modest resources available to him, he produced ground-breaking and highly influential advances in the technologies now being deployed on an unprecedented scale to search for drugs against SARS-CoV-2 replication enzymes and thus limit the worldwide impact of COVID-19. His success emanated from his wholehearted commitment to structural biology, from his daring initiatives based on the strong engineering background he put to the service of that commitment and, last but not least, from his determination to disseminate his innovations through entrepreneurship, undeterred by the bureaucratic complexities surrounding technology transfer procedures from public research organizations into the private sector. It is a great loss to the MX community that he passed away so prematurely.

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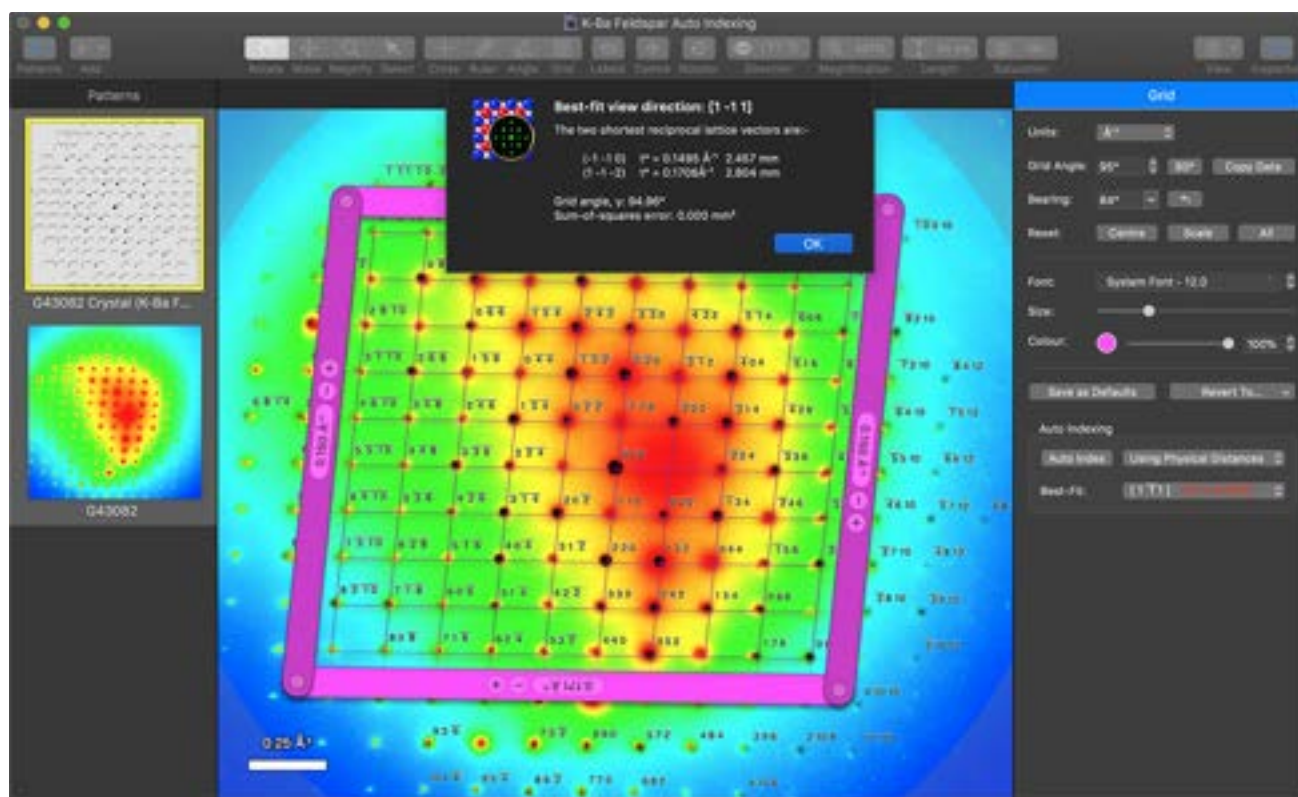
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## NEW PRODUCTS

# SingleCrystal 4: real-time multi-phase diffraction simulation



Using *SingleCrystal 4*'s auto-indexing feature to determine the orientation of an observed electron diffraction image.

CrystalMaker Software Ltd has announced the release of a new version of their *SingleCrystal* program: a tool for understanding the diffraction properties of crystalline materials.

Designed for physicists, chemists, material scientists and geologists, *SingleCrystal 4* can simulate the results of laboratory diffraction experiments – in real time, and from the comfort of the user's laptop. Observed electron diffraction images can also be analysed and their crystal orientations calculated, saving hours of painstaking measurement and analysis.

*SingleCrystal 4* features a brand new design: the source code was completely rewritten to take advantage of the latest 64-bit multi-core operating systems with high-resolution graphics and multi-touch control. A new interface delivers a streamlined easy-to-use workflow and offers real-time adjustment of experimental and sample parameters.

A key feature of *SingleCrystal 4* is the ability to simulate multi-phase diffraction – to replicate real-world effects such as 'twinning', topotactic relationships, crystal coatings and impurities – as well as showing 'Kikuchi lines' in transmission electron microscope patterns and 'powder rings' from nanocrystalline samples.

Whilst *SingleCrystal 4* can be used as a standalone program – to create new simulations from scratch, or import crystal data files from other programs – it also works seamlessly with *CrystalMaker X*, allowing users to edit crystal structures at the atomic level (in *CrystalMaker*) and immediately see how this affects the diffraction properties: an ideal teaching tool.

*SingleCrystal 4* is available as true native 64-bit Mac and Windows applications. The Mac version works with Catalina, offering Retina graphics, 'Dark Mode', multi-touch, haptic feedback and a dedicated Touch Bar interface for MacBook Pro. The Windows version is designed to run on the latest generation of Windows PCs and tablets, with high-DPI displays and touch-sensitive control.

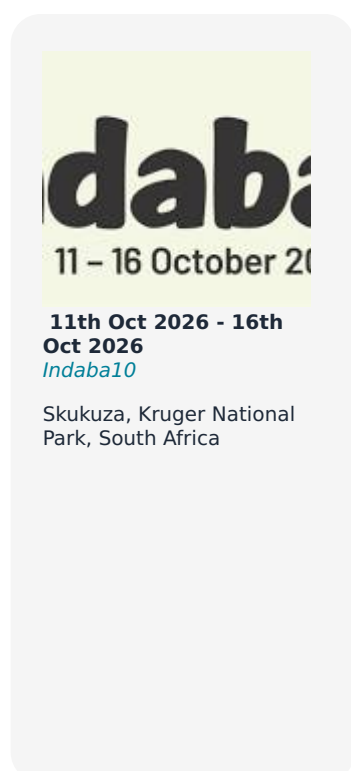
The software is sold directly by [CrystalMaker Software Ltd](#). Multi-user, education and student licensing options are available.

## IUCR SUPPORTED EVENTS

# IUCr supported meetings

During this period of meeting cancellations and postponements because of international health concerns, the IUCr will make best efforts to indicate the status of advertised meetings whenever we receive updates. Please refer in all cases to any website maintained by the meeting organizers for definitive information.

These forthcoming meetings are supported by grants from the IUCr Calendar Committee.





**5th Dec 2026 - 10th Dec 2026**

*African Materials Research Society (AMRS 2026)*

Nairobi, Kenya

## MEETINGS CALENDAR

## Meetings calendar

During this period of meeting cancellations and postponements because of international health concerns, the IUCr will make best efforts to indicate the status of advertised meetings whenever we receive updates. Please refer in all cases to any website maintained by the meeting organizers for definitive information.

This is a list of meetings of interest to crystallographers occurring within the next twelve months, as recorded in the IUCr Calendar of Events. Please let us know of any that are missing by [completing this form](#) or sending an email to [forthcoming.meetings@iucr.org](mailto:forthcoming.meetings@iucr.org).

|                                                                                     |                                                                                                                                    |
|-------------------------------------------------------------------------------------|------------------------------------------------------------------------------------------------------------------------------------|
|    | <b>Indaba10</b> South Africa 11th Oct 2026 - 16th Oct 2026                                                                         |
|    | <b>Synchrotron Radiation: Science, Knowledge, Innovations, Future (SKIF-2026)</b> Russian Federation 18th Oct 2026 - 23rd Oct 2026 |
|  | <b>CCP-EM/Diamond Icknield Workshop</b> United Kingdom 9th Nov 2026 - 13th Nov 2026                                                |
|  | <b>Instruct-ERIC hands-on workshop on Advanced cryo-FIB-SEM methods</b> Germany 9th Nov 2026 - 13th Nov 2026                       |
|  | <b>Crystal36-AXAA 2026 Meeting</b> Australia 26th Nov 2026 - 2nd Dec 2026                                                          |
|  | <b>African Materials Research Society (AMRS 2026)</b> Kenya 5th Dec 2026 - 10th Dec 2026                                           |
|  | <b>Joint Polish-German Crystallographic Meeting 2027</b> Germany 15th Feb 2027 - 18th Feb 2027                                     |