HOW THE UK'S SYNCHROTRON HAS BEEN HELPING SHED LIGHT ON COVID-19

Isabelle Boscaro-Clarke
Diamond Light Source

Over the past few months, scientific research on the SARS-CoV-2 virus which causes the disease COVID-19, has been a global priority. During this time, Diamond Light Source, the UK's national synchrotron, has collaborated tirelessly with the international and national scientific community to understand more about the virus and how we can combat it. Diamond are looking at the fundamental interactions of the virus, from which it is hoped new therapies can be developed. But it is also studying how existing drugs, that have already been tested and approved for other diseases, can be repurposed and used to treat patients. The array of specialised facilities at Diamond allow for many different techniques to be used, from looking at the structure of the virus and fitting drugs into it, like a tiny jigsaw puzzle, to taking direct images of the virus without its infectious component, so we can see how it interacts with drugs.

During the entire lockdown period it was not 'business as usual', as the synchrotron was solely devoted to COVID-19 research. However, as lockdown has eased services have now been expanded and include other Life Sciences research and programmes on other infectious diseases and illnesses that research teams need access to the facility to progress with a view to ramp-up operations in the physical sciences from Summer 2020 onwards.

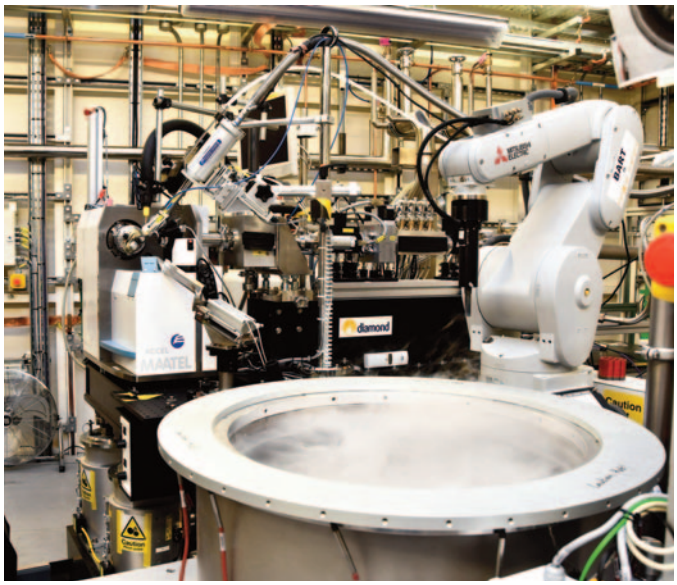
In addition to this, to facilitate the fastest dissemination of research at this critical time, it has not followed the usual research pathway of waiting for the publication of papers about research in peer-reviewed journals before releasing data. Through scientific collaboration, Diamond and its users have made (and are continuing to make,) data and results available in real time. Their goal is to share information as rapidly as possible to help inform the scientific community and wider public.

This article summarises the work and achievements to date of Diamond and its various international collaborators in the global challenge to find solutions to the worldwide pandemic:

WHY IS DIAMOND SO VALUABLE TO THE GLOBAL EFFORTS?

Diamond is one of the most advanced scientific facilities in the world. Funded by the UK Government through the Science and Technology Facilities Council (STFC) and by the Wellcome Trust, it provides the industrial and academic user communities with access to state-of-the-art analytical tools to enable world-changing science.

Shaped like a huge ring, the synchrotron works like a super-sized microscope. It accelerates electrons to near light speeds, to produce a light 10 billion times brighter than the Sun. This is then directed off into 33 laboratories known as 'beamlines'. This very intense light, predominantly in the X-ray region, (10 billion times brighter than the sun) can then be used to study minute matter such as atoms and molecules.



Beamline I04-1 Experimental Hutch - Sample changer and sample environment 2 – *Copyright of Diamond Light Source Ltd.*

SO WHAT HAS THE UK'S SYNCHROTRON BEEN DOING TO ADVANCE COVID-19 RESEARCH?

Diamond's research is centred on drug targets for COVID-19. The focus is mainly on the virus spikes, the receptor binding and the main protease. One specific part of its work, in conjunction with the University of Oxford, has been to look at these spikes on the outside of the virus.

Together, with a group at a hospital in Taiwan, they identified antibodies from a convalescent patient that could create a real potential for a drug target. This finding was valuable because it came from real patients who have the virus. Research about llamas' antibodies and SARS antibody CR3022 provided insights too. More on this to follow.

Further, the level of research automation offered by Diamond's synchrotron facility opened up ultra-high throughput screening methods. For example, the XChem platform's fragment screening capabilities enabled the interaction of many chemical fragments with potential drug targets within the virus to be analysed in great detail in a short amount of time.

To put fragment screening into context, it is one of the newest and most exciting techniques that offers scientists, who design new medicines, something unique: a glimpse into the future. With fragment screening, scientists can get away with testing only a few hundred tiny chemicals, what they call 'fragment compounds', to see if any of them might be the foundation for what could eventually become a new drug.

Fragment research is a recent and burgeoning field and can help produce drugs quicker, cheaper and smarter than before – critical at a time like this when finding new therapies is the focus of the scientific community. Diamond allows structural biologists to screen up to 500 structures daily. This is transforming the use of information for drug development, as information can be produced at a rate that can feed directly into chemical synthesis.

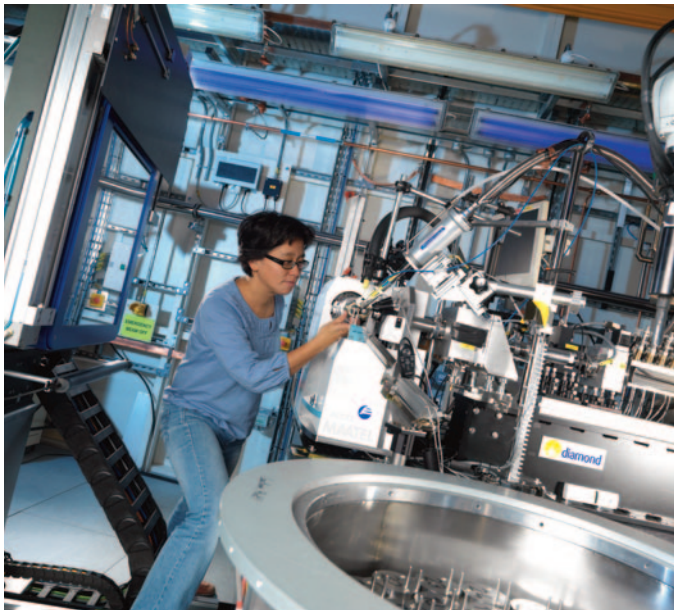
GLOBAL COMMITMENT, COLLABORATION, RESEARCH & RESULTS

As part of this acceleration, Diamond mobilised a concerted effort to collaborate on a global scale. This has involved seven researchers at Diamond (Walsh and von Delft groups), researchers from the Weizmann (London group) and Exscientia Ltd, among many others. It also established links with Public Health England (Carroll group), Shanghai (Rao group), Beijing (Wang group) and the University of Oxford (Owens group).

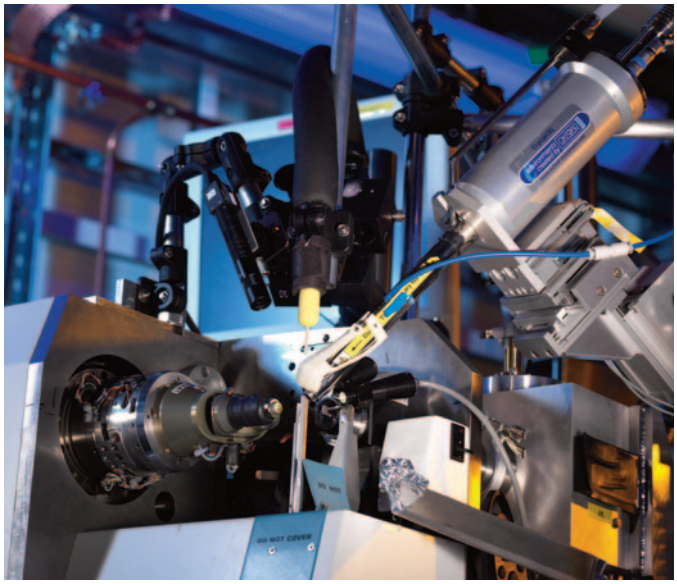
Many of the scientists on the seven beamlines kept them running all through lockdown, often with little sleep, as part of a genuine commitment to form part of global efforts to find solutions.

EXSCIENTIA, CALIBR AND OXFORD UNIVERSITY

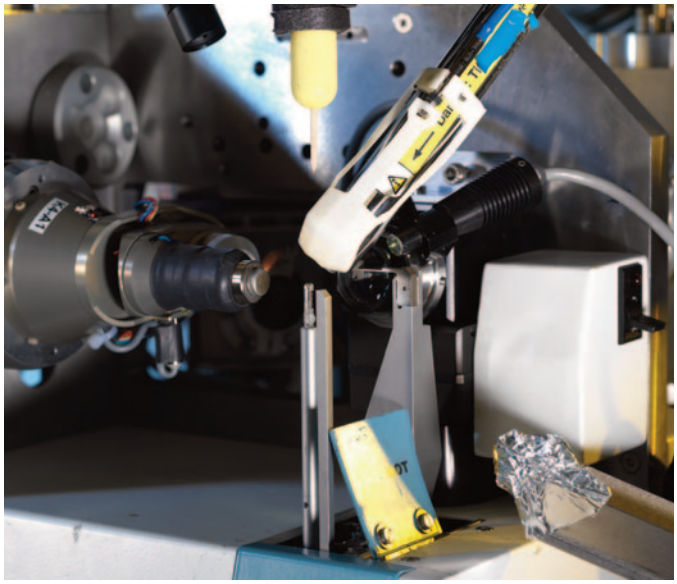
Starting in March, Diamond minimised the travel and number of people on site and suspended other user operations so that only COVID-19 research took place at the facility during the UK lockdown period. Importantly, Diamond also announced a joint initiative with Exscientia, the leading artificial intelligence (AI) driven drug discovery company and Calibr, the drug development division of Scripps Research, to search



Alice Douangamath, Senior Beamline Scientist I04-1 – *Copyright of Diamond Light Source Ltd.*



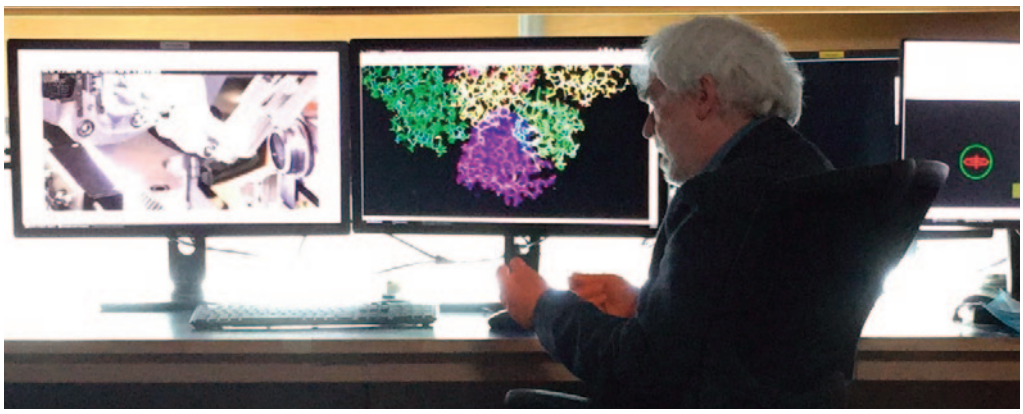
Beamline I04-1 Sample Environment – *Copyright of Diamond Light Source Ltd*



Beamline I04-1 Sample Environment 2 – *Copyright of Diamond Light Source Ltd.*

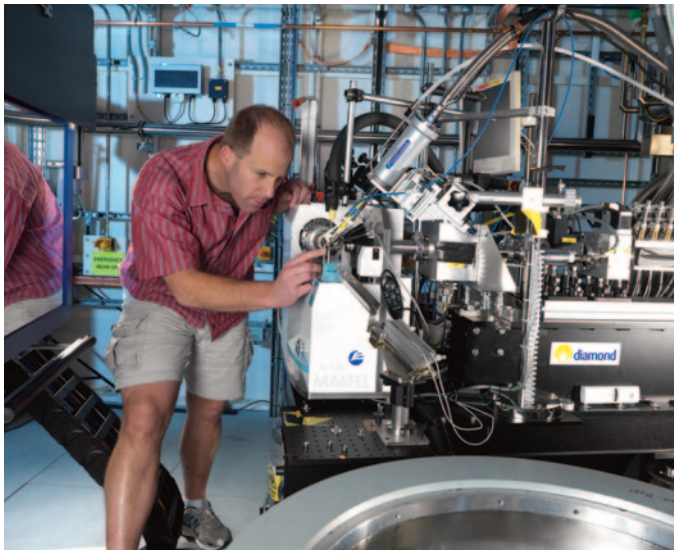
for clinically approved drugs that could be viable clinical drug candidates for the rapid treatment of COVID-19.

Through using Diamond's research on COVID-19 at its facilities, Exscientia screened nearly every known approved and investigational drug - 15,000 clinical drug molecules - against COVID-19 drug targets to search for rapid treatments. Additionally, Diamond and Oxford University have worked together since January to develop methods to produce the viral proteins for drug screening and structural analysis. This is providing atomic levels of detail in understanding how anti-viral drugs can work against the virus. Dr Martin Redhead, Head of Quantitative Pharmacology at Exscientia, carried out the analysis and commented at the time: *"Given the ever-expanding scale and rapid speed at which COVID-19 is spreading, one of the most urgent needs right now is to find ways to discover an existing drug we can repurpose to treat the virus, at speed and at scale. The Scripps Research collection allows us to screen nearly every drug that has been tested in human clinical trials, against a number of virus drug targets."*



Professor David Stuart & I03 Beamline - Director of Life Sciences at Diamond Light Source – *Copyright of Diamond Light Source Ltd.*

Prof. Dave Stuart FRS is Director of Life Sciences at Diamond Light Source and Joint Head of Structural Biology at the Department of Clinical Medicine at the University of Oxford. He is co-ordinating the efforts between Diamond Light Source and Oxford University along with colleagues, Dr Martin Walsh and Dr Jonathan Grimes. He added: *"The drugs we are testing have either been approved by the FDA for other diseases or have been extensively tested for human safety. By being able to repurpose existing molecules, we can save a lot of time in the drug discovery process, meaning a faster route to clinical trials, and potentially to treatment for patients."*



Frank von Delft, Principal Beamline Scientist I04-1 *Copyright of Diamond Light Source Ltd.*

THE COVID MOONSHOT INITIATIVE: DRIVING PROGRESS COLLABORATION

In April, Diamond Light Source, PostEra Inc. and an international group of scientists from academia and industry teamed up to form a new, ground-breaking non-profit initiative – the COVID Moonshot. The name is derived from its unprecedented aim to develop a clinically effective antiviral more rapidly than ever before; by crowdsourcing designs of new inhibitors from chemists around the world, mining the rich "fragment" data measured at Diamond in record time during March and April. All this data will be released in real time and in the open to enable worldwide collaboration and rapid progress.

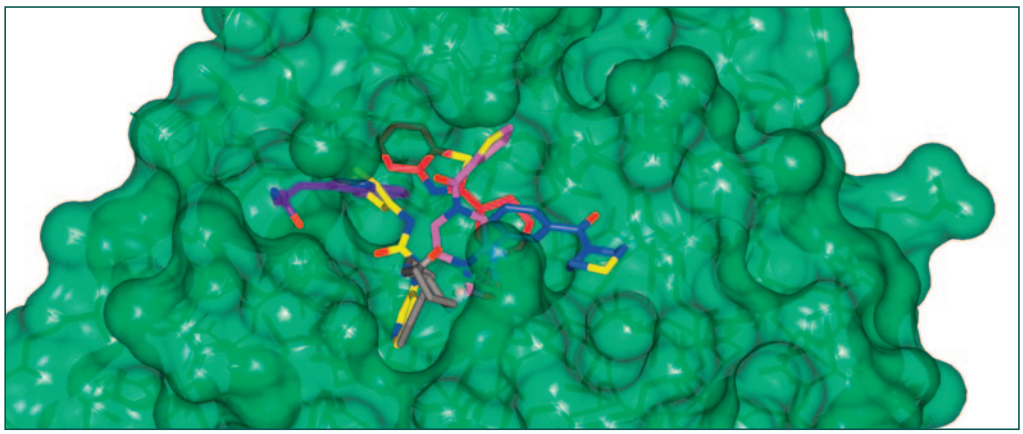
This collaboration of international scientists is working together to find new ways of tackling the COVID-19 challenge and have created a clear design-to-clinic strategy and timeline. Chemists were called on to come up with new molecules and to have practical input towards the global efforts to combat COVID-19. Researchers could submit their designs to PostEra, who were running machine learning algorithms in the background to triage suggestions and generate synthesis plans to enable a rapid turnaround. Promising compounds were then synthesized and tested for antiviral activity and toxicity.

Prof Frank von Delft, Science Leader of the XChem laboratory at Diamond commented: *"A key part of the strategy is to pursue only molecules that are easy to produce – because if we do find something, large quantities of it will be needed very quickly. If a safe, easy-to-make, effective antiviral compound exists, then we need to know, and fast. We have been humbled by the readiness of so many organisations and companies to contribute effectively pro bono, particularly Enamine in the Ukraine; and then of course, by the huge intellectual contribution from the scientific community."*

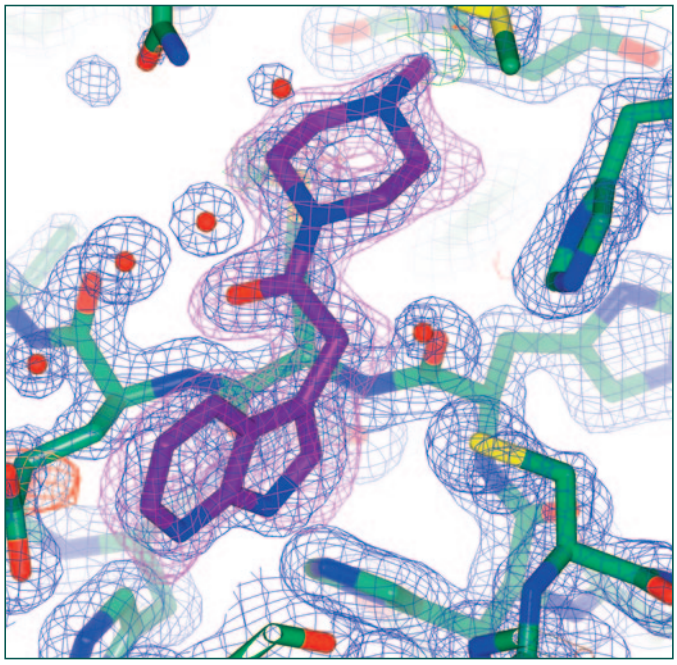
This new approach has already paid dividends. Following just two rounds of designs, the team received over 3500 molecular design contributions and the first compounds have been tested. A paper is close to submission and they have already identified at least one serious therapeutic possibility at Oxford.

THE ROLE OF OPEN SCIENCE AND NEW RESEARCH PORTALS

Aside from COVID Moonshot, Diamond reaffirmed its commitment to COVID-19 research by agreeing to act as a collaborator in an Open Science initiative, led by Wellcome and UKRI, when a consortium of scientists launched the COVID-19 Protein Portal in May. Crucially, the portal enables UK scientists to access protein reagents for critical research relating to SARS-CoV-2. Protein reagents are provided free of charge by a consortium of leading protein production laboratories.



Surface representation of SARS CoV-2 Mpro protein with fragment hits from XChem platform bound in active site (Green) – *Copyright of Diamond Light Source Ltd.*



Modelling of fragment hits into electron density maps obtained from X-ray diffraction CoV-2 Mpro protein crystals 1 – *Copyright of Diamond Light Source Ltd.*

ENGINEERED LLAMA ANTIBODIES NEUTRALISE COVID-19 VIRUS

In July scientists discovered that antibodies derived from llamas neutralise the SARS-CoV-2 virus in lab tests. Researchers included Diamond Light Source, the Rosalind Franklin Institute, Oxford University and Public Health England. They hoped the antibodies – known as nanobodies due to their small size – could eventually be developed as a treatment for patients with severe COVID-19. The peer reviewed findings were published in Nature Structural & Molecular Biology.

Professor James Naismith, Director of The Rosalind Franklin Institute and Professor of Structural Biology at Oxford University said: *"These nanobodies have the potential to be used in a similar way to convalescent serum, effectively stopping progression of the virus in patients who are ill. We were able to combine one of the nanobodies with a human antibody and show the combination was even more powerful than either alone. Combinations are particularly useful since the virus must change multiple things at the same time to escape; this is very hard for the virus to do. The nanobodies also have potential as a powerful diagnostic."*

Dave Stuart added: *"The electron microscopy structures showed us that three nanobodies can bind to the virus spike, essentially covering up the portions that the virus uses to enter human cells."*

SARS ANTIBODY CR3022 ALSO NEUTRALISES SARS-COV-2

New research points out that targeting spike proteins could provide an effective treatment for SARS-Cov-2. This will provide further insights to teams trying to develop vaccines. The research indicates that the SARS antibody CR3022 can neutralise SARS-CoV-2 by destroying prominent spike proteins. Conducted by a Diamond team, the research discovered that the CR3022 antibody may have therapeutic potential alone or in combination with other antibodies.

Dave Stuart explained: *"This new research demonstrates that CR3022 works in an entirely different way. Using the high-resolution Cryo-electron microscopes at Diamond's Electron Bio-Imaging Centre, the team were able to watch the antibodies in action against the virus and to determine their method of attack. It destabilises the spike protein entirely, neutralising the virus. CR3022,,therefore, could work on its own, or in tandem with receptor blocking antibodies."*

"This finding is particularly important in this antibody jigsaw puzzle because the most common way to screen antibodies for effectiveness against SARS-CoV-2 doesn't check for this alternative method of attack. Further screening could identify more useful antibodies that had previously been classed as ineffective. These results could lead to antibodies that can be used to treat SARS-CoV-2. As human antibodies arise within the body, they are relatively safe. Drug companies can replicate antibodies quickly in the quantities that would be needed."

As the search for drug targets continue, great strides have been made at Diamond Light Source in researching, testing and the analysis required to facilitate progress that will benefit the world. Diamond Light Source has played a critical role in working on a wide range of COVID-19 related projects that teach us more about the virus and how we can combat it - and will continue to do so in the future. □