

# STAKEHOLDERS

La Trobe University



CSIRO



Swinburne University of Technology



University of Melbourne



Monash University



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## DIRECTOR'S REPORT

The Australian Research Council (ARC) Centre of Excellence for Coherent X-ray Science is a multidisciplinary research centre encompassing physicists, cell biologists, structural biologists and physical chemists. We are dedicated to the idea of working across disciplinary boundaries and tackling the most important scientific problems at the intersection points of these disciplines. The ARC has made a major commitment to this project, as has the member universities, the CSIRO and the Victorian State Government through its STI initiative. Our goal is to not only make some major contributions but also to demonstrate how effective a first-rate, interdisciplinary team can be, and so pioneer a new way of doing science in Australia. I have personally been delighted by the quality of the team, and the spirit of sharing and collaboration that has emerged. We are in for an exciting time.

It is widely held that science in the 21st century will depend on scientists with different backgrounds working together on complex problems. An exemplar of this type of work is the discovery of the double helix more than fifty years ago by a combination of theory, modelling and X-ray crystallography experiments. A major problem in the 21st century is the determination of the structure of proteins that cannot be crystallised – the methods of crystallography cannot be applied. The mission of CXS is to contribute to the solution of this problem. It represents an unashamedly ambitious goal, but we have assembled an excellent and committed multidisciplinary team that has just the right blend of skills to achieve it.

The first six months of the centre have been mainly one for establishment and communication. We have had some exceptionally stimulating meetings and have started the preliminary work for some of our projects. We have also learned that the CSIRO part of the centre have been granted funding from the CSIRO "Emerging Science Initiative" enabling us to work in a highly interactive and cooperative framework.

The CXS hiring process is now well underway and I think we are poised to have some exciting and scientifically productive years ahead of us, and, as will be seen in this report, the first six months have already seen steady progress. I look forward to seeing this project build over the coming years.

# FINANCIAL STATUS

Income was received in January 2006 from the ARC and collaborating institutions to the total of \$900,000. As a result there was no expenditure in 2005.

CSIRO Emerging Science Initiative funding, of approximately \$2M pa for two years, was approved and will support those areas of the CoE that were not funded by the ARC.

| FINANCIAL REPORT         |                         |                  |
|--------------------------|-------------------------|------------------|
| Current Reporting Period |                         | \$               |
| Carry Forward            |                         | Nil              |
| STI Allocated Funds      |                         | 1,000,000        |
| Other Funds              | ARC                     | 201,771          |
|                          | Uni Contribution        | 90,000           |
| <b>Total Income</b>      |                         | <b>1,291,771</b> |
| Expenditure              | Administration Salaries | 18,790           |
|                          | Consumables             | 5,625            |
|                          | Research Staff Salaries | 158,781          |
|                          | Marketing               | 4,875            |
|                          | Meetings                | 2,079            |
|                          | Visitors Program        | 4,875            |
| <b>Total Expenditure</b> |                         | <b>195,025</b>   |
|                          |                         |                  |
| <b>Balance</b>           |                         | <b>1,096,746</b> |



# MISSION

To be the world leader in the development of non-crystallographic techniques for the determination of membrane protein structures.

# AIMS

The ARC Centre of Excellence for Coherent X-ray Science (CXS) brings together leading Australian researchers in the fields of X-ray physics, the design and use of synchrotron radiation sources, and the preparation, manipulation and characterisation of biological samples.

Its aim is to open a new frontier in biotechnology; the non-crystallographic structure determination of membrane proteins. These proteins mediate the activity of pharmaceuticals in human medical therapies. Their structures, however, are still mostly unknown because they do not form suitable crystals for analysis using the conventional crystallographic techniques that have driven almost all progress in structural biology. A breakthrough in this area would revolutionise rational drug design through the insight that would be gained into the functionalities of these proteins, and would have far-reaching consequences throughout the pharmaceutical industry.

The research of CXS is driven by our access to existing third-generation synchrotron light sources and to the Australian Synchrotron, which will become operational in 2007. We are also exploring the application to imaging problems of short wavelength high-harmonic generation sources and free-electron X-ray lasers that are under development worldwide. When combined with non-crystallographic diffractive imaging techniques, the brightness and intensity of these sources in femto-second pulses offers the possibility of taking structural snapshots of biomolecules. We are exploring fundamental issues in the use of these new light sources, including the nature of the interaction between intense coherent X-rays and electronic matter, the efficiency of diffraction processes in these highly coupled light-matter systems, the detection of the scattered light, the preparation and handling of suitable biological samples, the management of radiation damage throughout the interaction and the design of algorithms to extract structural information from diffraction data.

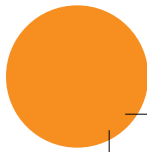
It is an ambitious inter-disciplinary program of research.

# OVERVIEW

The activities of CXS are complex and interdisciplinary. Our member institutions, University of Melbourne, La Trobe University, Monash University, Swinburne University of Technology and CSIRO, are all situated in Melbourne, with each institution hosting nodes of CXS led by a Group Leader who is also a member of the Executive Committee.

The scientific direction of CXS is coordinated by the Director, the Group Leaders, and an International Advisory Committee comprised of distinguished international leaders in associated fields.

CXS is administered by a Chief of Operations and two Administrative Assistants who are responsible for the financial management and for facilitating the outreach of CXS to the wider community. The Centre is committed to the widespread dissemination of its activities to traditional scientific journals and meetings, to directing engagements with academic and research institutions and with selected pharmaceutical, biotechnology and optical sciences industries.



# PROGRAMS

## Biological Sciences Program

The "Holy Grail" of structural biology is the determination of the molecular structure of a biomolecule without first obtaining a high-quality three-dimensional crystalline sample of the material for X-ray crystallographic analysis. X-ray crystallography has provided the basis for most of the advances in structural biology over the past eighty years through the amplification of diffracted signal afforded by a periodic crystalline structure. While this form of structural analysis is now regarded as a routine technology, it is the formation of suitable crystals of these biological molecules that presents the greatest obstacle to sustained progress. The most elusive of all crystalline samples are also amongst the most significant from a pharmacological perspective: the membrane proteins. These tend to form either nanocrystals or two-dimensional quasi-periodic structures. These systems are of such biological significance that substantial investments of resources in the development of special structural characterisation techniques are warranted.

Apart from the production and handling of suitable membrane protein samples, the Biological Sciences Program will produce and process contrast-enhanced samples for cellular imaging. Some progress has already been made in this direction by applying coherent diffractive imaging techniques to the imaging of malaria-transfected blood cells. The immediate aim is to obtain a more detailed picture of functionalities within cells by direct observation using X-ray diffractive imaging than is currently available from simple models. This is an essential step along the road to biomolecular imaging.

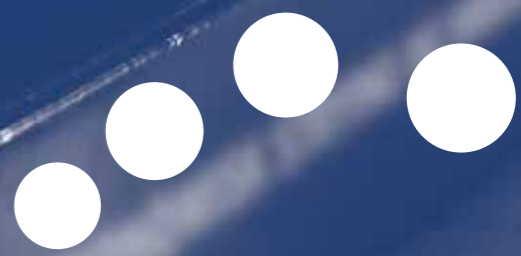
## Theory and Modelling Program

The imaging methods being developed by this group all depend on the existence of a unique relationship between a diffraction pattern and an over-sampled periodic diffracting object. The program has found success in the past year in developing techniques adapted to imaging using beams of X-rays focused by zone-plates, and iterative phase retrieval algorithms derived from Fresnel propagators.

All methods of this type use whatever a priori information that is available about the diffracting object to assist in obtaining a representation of its image from diffracted intensity data. Such information includes its spatial extent (support), its material composition, the phase and intensity structure of the illuminating source, and the geometrical parameters of the experiment. With this must also be handled sources of systematic and random error, such as sources of noise, detector response functions and errors in determining the geometrical parameters on which the reconstruction algorithms critically depend. Two general approaches are being explored in the

algorithm development program. The first of these are based on numerical propagation of the electromagnetic information between planes that are subject to known constraints. To this are currently being added a second layer of statistical methods, in which Bayesian hypothesis testing is used to assess the information content in the reconstruction on the basis of models of the diffracting object and the sources of error. This second line of attack will involve the detailed modelling of the experiment, including treatments of the partial coherence of the source, the detector response function, the characteristics of the focusing zone-plate optics and the influence of scatter from intermediate apertures in the system. As well as these physical effects, detailed electronic models of the scattering materials are to be used to formulate tests against which to measure the likely accuracy of the reconstruction to the experimental data.

In preparation for the availability of highly coherent X-ray sources from high harmonic generation or free-electron lasers, preliminary work has begun on modelling the interaction between molecules and intense electromagnetic fields. This is very much frontier territory in theoretical research because of the need to obtain a detailed description of the time-dependent response of a complex system to a rapidly varying external perturbation that is sufficiently strong to cause secondary effects such as ionisation and the cleavage of bonds. Since it is envisaged that the interaction between a molecule and such a source will occur over the lifetime of a pulse lasting of order a femto-second, the possibility exists that it will be possible to extract structural information from the diffracted intensities resulting from these encounters. Success in this part of the program will require a fundamental reappraisal of methods currently used in coherent diffractive imaging because of the non-linear response of the target to the illumination.

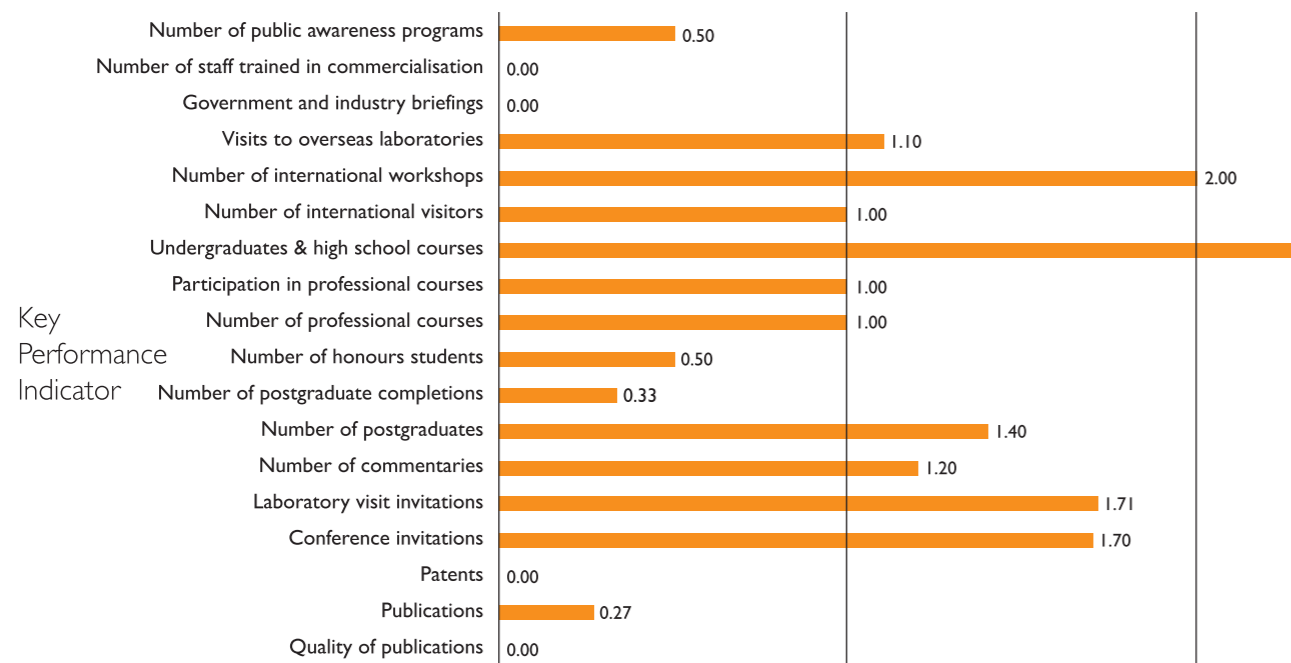


### Performance Indicator Data

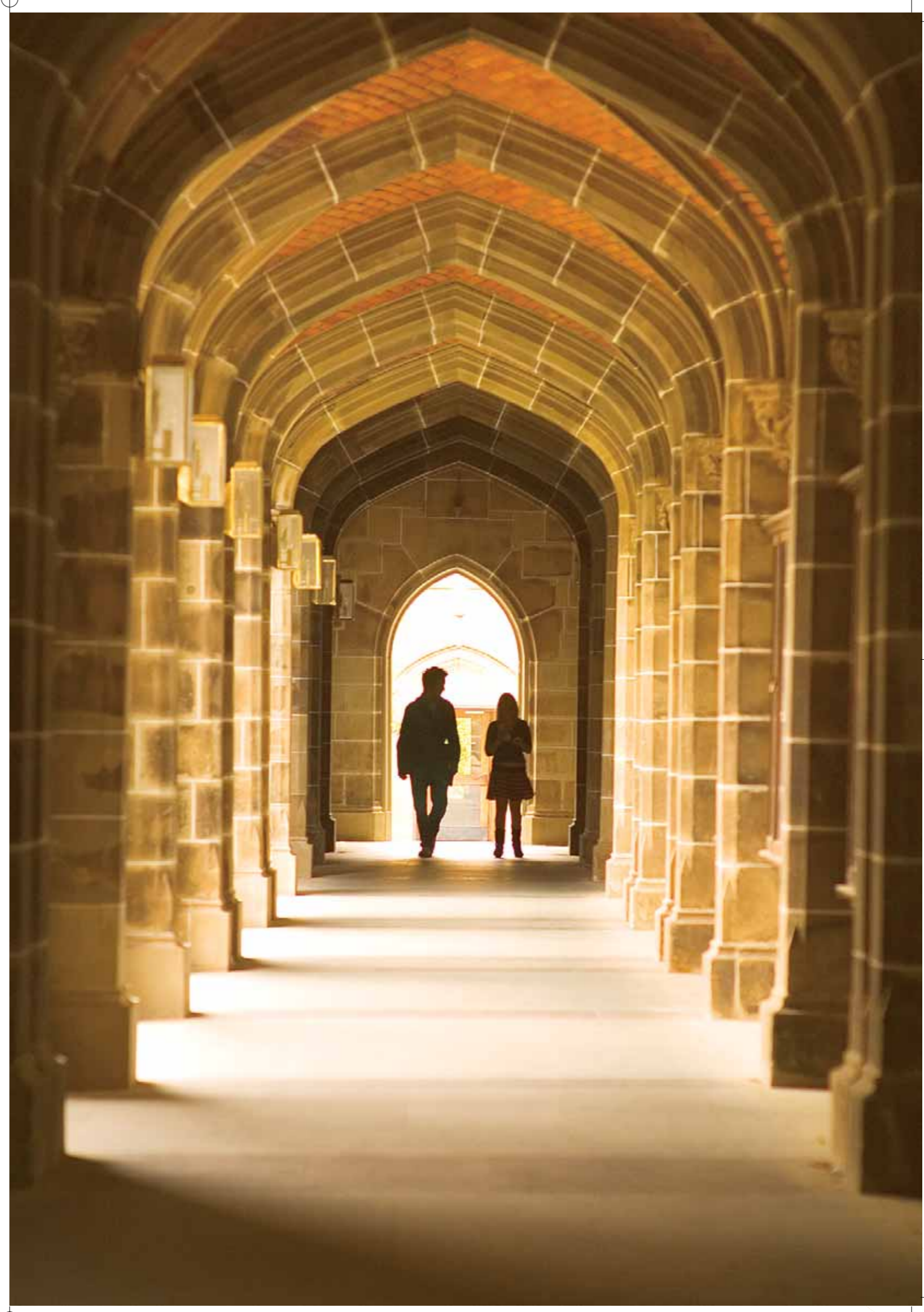
See plot below. These parameters are current as at April 2006 and so extend beyond the 2005 Calendar Year reporting period. This report also includes papers accepted for publication, but not yet published, and only considers those that acknowledge support from the centre.

We are generally tracking well against the KPIs and we are particularly pleased to note that we have exceeded our targets for postgraduate students.

### Fraction Achieved



Achievement of Key Performance Indicators for 2005 expressed as a ratio of achievement to target. Some of these, such as publications and invitations to speak, are figures projected forward for 2006. The quality of publication KPI was not assessed for this graph.



# PERFORMANCE MEASURES

CXS entered 2005 with the negotiations of our collaborative agreement, the setting up of the Advisory Board and Executive Committee, and other preparatory work. Through these efforts we believe we have created a stable foundation for the achievement of our goals.

After the interview with the ARC for the CXS on March 8, and the announcement of funding on June 15, we are pleased to report that some time for centre-inspired science was made; however progress toward commercialisation and technology transfer has not moved forward as we are in the preliminary stages of set up, with committed funding not being received until January 2006. Gains toward this objective will commence in 2006.

## Progress towards Research Targets

CXS has held a series of meetings with the Executive Committee and Advisory Board discussing scientific objectives and forging relationships. There is a great deal of excitement and a large number of ideas being generated.

Our researchers have developed samples for the investigation of cellular architecture and acquired some X-ray diffraction data from them (see Figure 1). We are working on the reconstruction of this data and have identified issues with the staining of the samples that we are now endeavouring to solve.

We have developed methods for the high-resolution characterisation of the structure of a coherent wave produced by a zone plate.

We have also begun the identification and tender process for the purchase of the Ultrashort-Pulse, High-Pulse Energy Laser System, and other high-cost equipment.

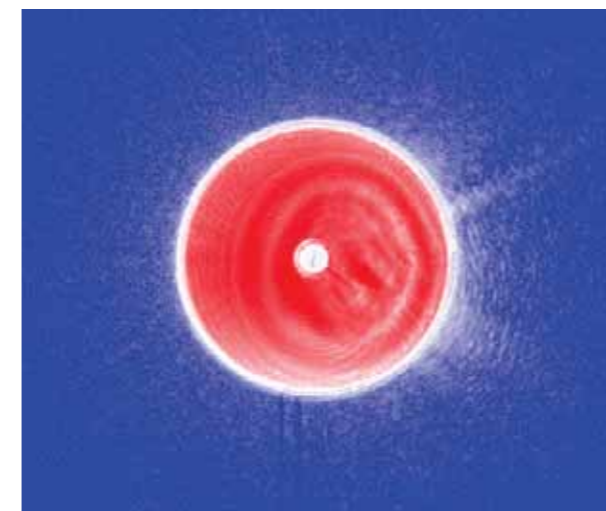


Figure 1: Diffraction data from a malaria parasite infected blood cell acquired by the experimental x-ray physics group

## Education and Training

Seven post-graduate students and seven honours students were recruited in 2005/early 2006. Four summer vacation studentships were offered; three to undergraduate students and one to a Year 10 student from Leongatha, Victoria. A Masters of Science student, Mr. Maik Fröhlich, from Fachhochschule Jena University of Applied Sciences, Germany, undertook a 5 month internship at the University of Melbourne, from October 31st, with Associate Professor Trevor Smith.

A public seminar entitled, *The Marriage of Physics & Biology: Will DNA be an only child?* was delivered by Keith Nugent on September 6th at the Institute for Advanced Studies at La Trobe University with over 100 attendees, and at Swinburne University of Technology on October 28th. Additional educational seminars were delivered including a seminar entitled *Einstein's Science* by Dr Andrew Peele and an undergraduate seminar on the Centre's work to Physics students by Keith Nugent on October 7th. Keith Nugent also presented an in-service lecture for teachers on November 11th held at the University of Melbourne and gave a related seminar at the NSSLRC synchrotron facility on December 12th.

## Industry and Community Linkages

We have consolidated an existing relationship with the Advanced Photon Source under the General User Program and are conducting a continuing campaign of experiments at this facility. We have been invited to participate in the Guest Investigator Program at the Spring8 synchrotron in Japan, and following the offer of dedicated beamtime access at the Taiwanese National Synchrotron Radiation Research Centre, we conducted an exploratory experiment in December 2005 to investigate how well that facility can serve our needs.

Discussions with Professor Dick Wettenhall with respect to interactions with Bio21 Institute have been initiated.

On November 7th representatives of the CoE met with members of the Smorgon Family to discuss how their investment has helped the centre.

The work of the CoE was presented by various members at University seminars and at national and international conferences, including the Australian Society for Biophysics, ACT, September 2005; the CSIRO Emerging Sciences Initiative for Synchrotron Sciences Workshop, CSIRO, CMIT, Clayton, (12/04/2005); the First International Workshop on Optical Probes for Molecular and Cellular Imaging, Sonoma, CA, USA, December, 2005; and the 2nd Italian-Australian Workshop in Imaging and Spectroscopy, Trieste, Italy, (09/02/2005).



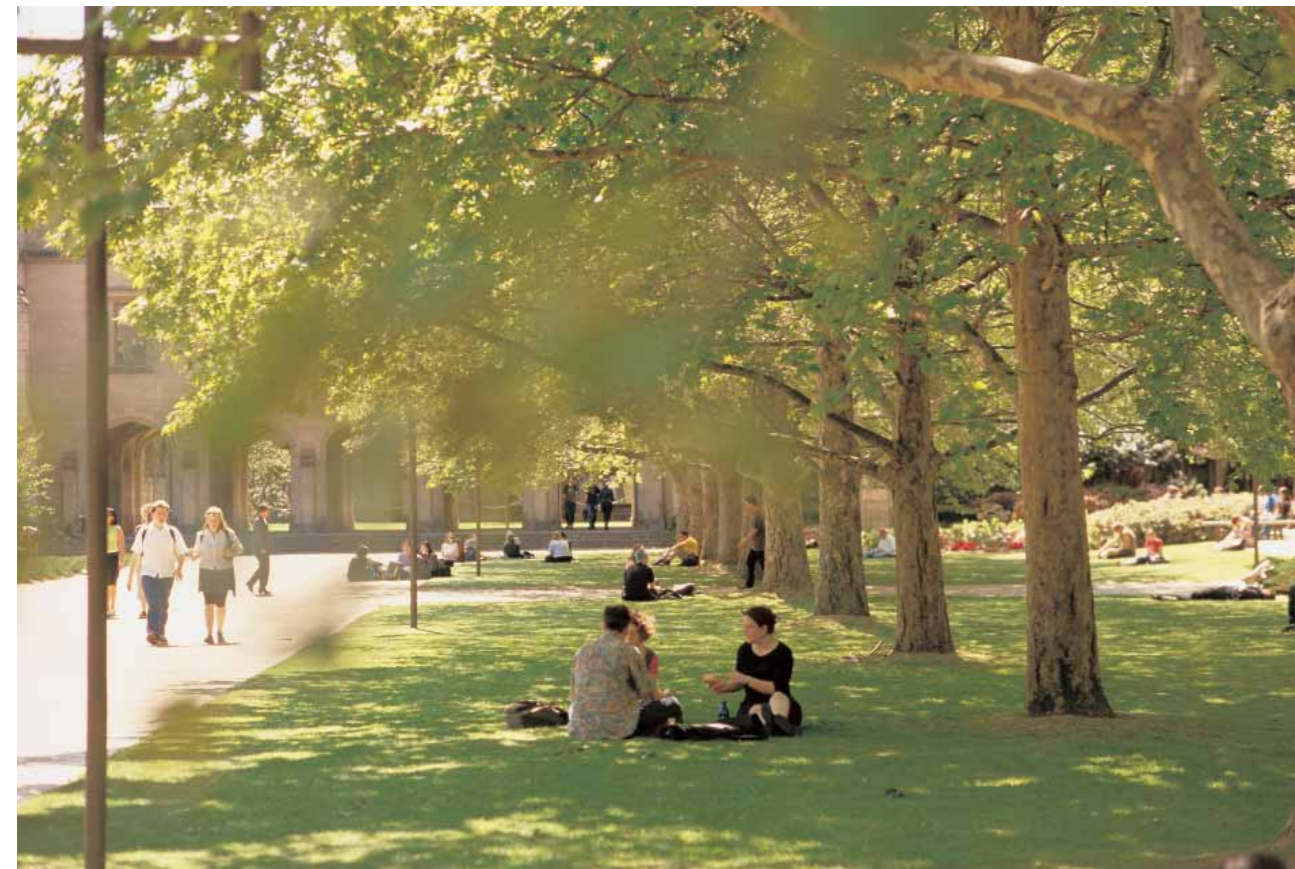
# PUBLICATIONS AND MEDIA COVERAGE

The following papers were accepted for publication:

- *Diffractive Imaging of Highly Focused X-ray Fields*; H.M Quiney, A.G. Peele, Z.Cai, D.Paterson, K.A. Nugent, published in *Nature Physics*/Vol. 2/ February 2006
- *X-ray Optics Imaging at arm's length*; C. Jacobsen, published in *Nature Physics*/Vol. 2/ February 2006
- *Phase-space reconstruction of focused x-ray fields*; C. Tran, A.Mancuso, B.Dhal, K.Nugent, published in Vol.23, No. 7/July 2006/*Journal of the Optical Society of America*

A number of commentaries about the centre and its achievements have been published including the following articles:

- *X-Ray Eyes*, Campus Chatter, The Age, July 18th 2005
- *Major Developments in Coherent X-ray Science*, La Trobe Bulletin, July 22nd, 2005
- *New centre to look at approaches to imaging proteins*, Australia Biotechnology News August 8th, 2005



## Experimental Methods Program

This program is developing the techniques that will be used in X-ray imaging experiments, building on our recent successes using curved beam illumination to aid image recovery. The fabrication of test samples within CXS forms an important part of this development work, since it will allow us to investigate critical aspects of the phase retrieval problem. These include the design of the experiment, the sensitivity of the phase recovery algorithms to noise, partial coherence, radiation damage and systematic effects such as internal translation of the beam or the various optical components.

Since the duty cycle for synchrotron experiments is so long, and because many of the features of diffractive imaging using X-rays can be simulated using visible sources, CXS is currently installing an experimental laser research laboratory. By a suitable scaling of the relevant variables and the careful selection of scattering targets it is possible to simulate almost all of the features of an X-ray experiment using relatively inexpensive components and visible coherent light sources in a laboratory.

The Experimental Methods Program will work closely with the Theory and Modelling Program to put in place effective methods of image recovery in advance of the commissioning of the Australian Synchrotron facility in 2007. This will allow CXS to hit the ground running when this national resource becomes available.

## Short Wavelength Laser Source Program

One of the long-term possibilities being investigated by CXS is the use of X-ray free-electron laser sources in imaging applications. Such facilities are under development in both Germany and the USA. While we expect to be able to gain at least limited access to XFEL sources when they become available, there are currently no plans for similar resources to be built in Australia. They are hugely expensive facilities involving the construction of linear accelerators more than one kilometre in length.

In the meantime, the short-wavelength laser source groups based at Swinburne University of Technology and the University of Melbourne are developing a tabletop source of highly coherent vacuum-ultra-violet or soft X-rays using high harmonic generation techniques. An intense pulse of visible or UV wavelength light is allowed to interact with an atomic plasma to form a highly coupled radiation-matter system. In the de-excitation process, pulses are created corresponding to multi-photon absorption by the plasma followed by the coherent emission of a photon whose wavelength is perhaps fifty times shorter than that of the incident laser.

The Centre is engaged in the development of these compact short-wavelength sources to investigate alternatives to the use of XFELs in coherent diffractive imaging. The significance of this research goes far beyond just the production of the source, since their use in any process involving the interaction of the highly coherent intense pulse produced by high-harmonic generation raises fundamental issues concerning the modelling of the electronic processes involved. Investigation of the nature of these non-linear physical processes in laser-molecule interactions forms an integral part of the activities of the Short Wavelength Laser Source Program.

## Detector and Beamline End-Station Development Program

The technical expertise of the members of the Detector and Beamline End-Station Development Program will be employed to develop experimental facilities at the Australian Synchrotron. These resources are essential to realising much of the research that is to be conducted by the other programs.

This program will develop detectors that are optimised to the experimental conditions prevailing in our X-ray diffraction experiments and will address issues such as sensitivity, response non-linearity, latent memory and readout fidelity.

In order to be able to complete the scientific goals of CXS, this program will undertake to design and assemble the instrumentation for a vacuum beamline end-station at the Australian Synchrotron suitable for routine diffractive imaging of nanoscale particles.

## Structure Determination Methods Program

One of the main aims of the Structure Determination Methods Program is the development of novel experimental techniques and data analysis methods for extracting structural information from 2-dimensional crystals, especially relating to the determination of the structure of integral membrane proteins (IMPs).

The program includes the fabrication of 2-D arrays by novel methods. It also includes the exploitation of grazing incidence X-ray diffraction (GIXD) methods to obtain high sensitivity structural information for thin samples. A powerful aid to the development of these techniques is simulation software being developed in order to help better understand the likely nature of diffraction data on such systems, including the influence of structural disorder. Methods of data analysis proposed for implementation include Bayesian and MaxEnt methods that seek to incorporate both data and a priori information.

Initial system being studied is Purple Membrane, a naturally occurring 2-d crystal. This is a classical membrane protein system that has been much studied by high resolution cryo-electron-microscopy and so valuable comparisons can be made about the efficacy of both electron and X-ray methods.

A long-term aim of the program is to develop novel instruments and techniques for the structure determination of integral membrane proteins, which might be implemented at the Australian Synchrotron.

Supported by:



Australian Government  
Australian Research Council



Australian Synchrotron  
lighting the path to innovation

# MEMBERSHIP

## Professional Staff

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# GOVERNANCE

## Centre Advisory Board

The Advisory Board will meet annually with the first meeting scheduled for April 2006, to coincide with a workshop of all centre participants.

We will form two sub-committees of the Advisory Board: A scientific sub-committee and an IP sub-committee. The membership of these will be finalised during the April meeting.

### Members are:

Professor John McKenzie, Deputy Vice Chancellor (Research), University of Melbourne (Chair)  
dean@science.unimelb.edu.au

Professor Erich Weigold, Deputy Vice Chancellor (Research), La Trobe University (or nominee)  
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Bruce Whan, Chairman of INNOVIC (Victorian Innovation Centre Ltd) and Director, Swinburne Knowledge  
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## Executive Committee

The Executive Committee meets every two months and has been doing so since July 2005.

### Members are:

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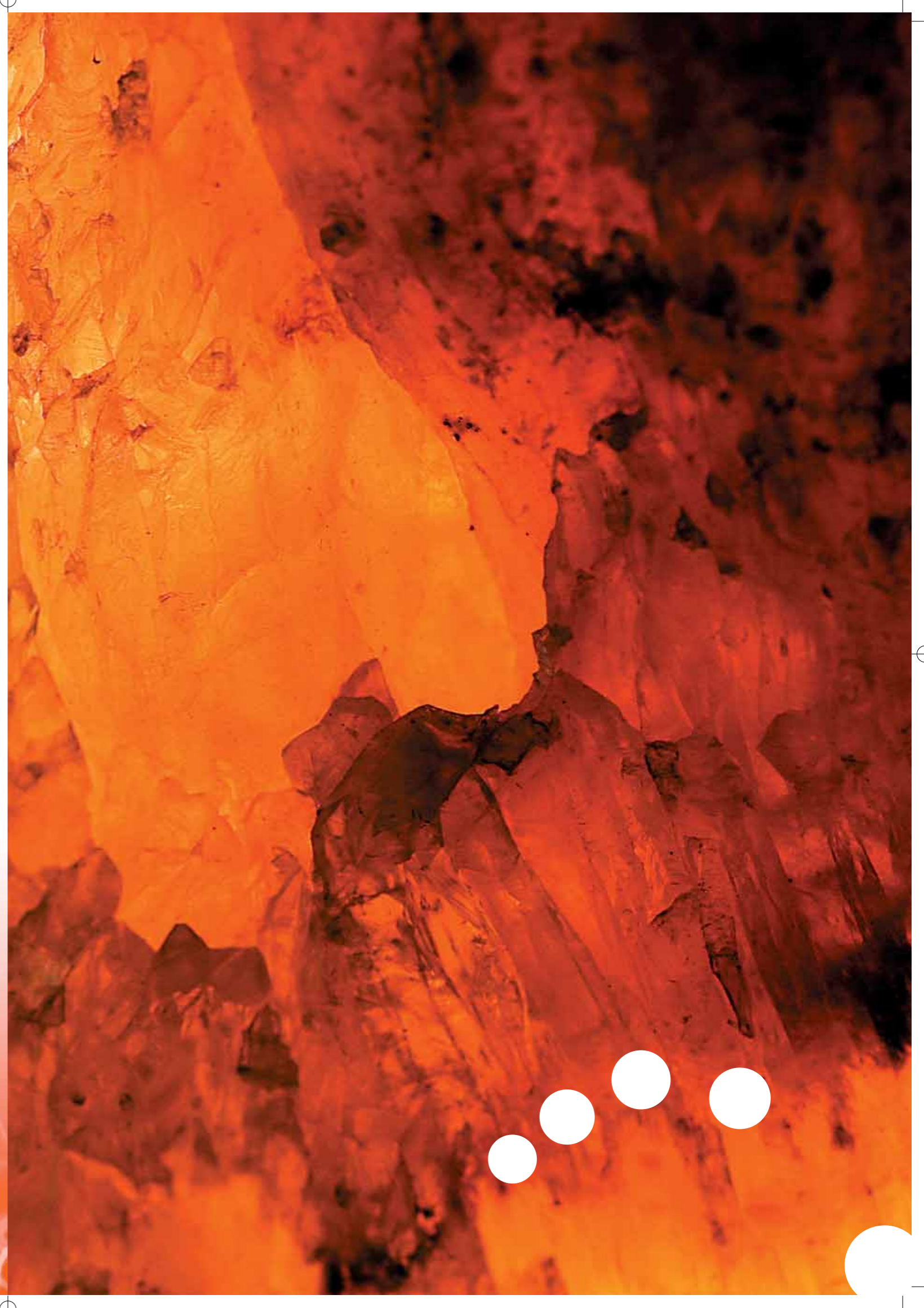
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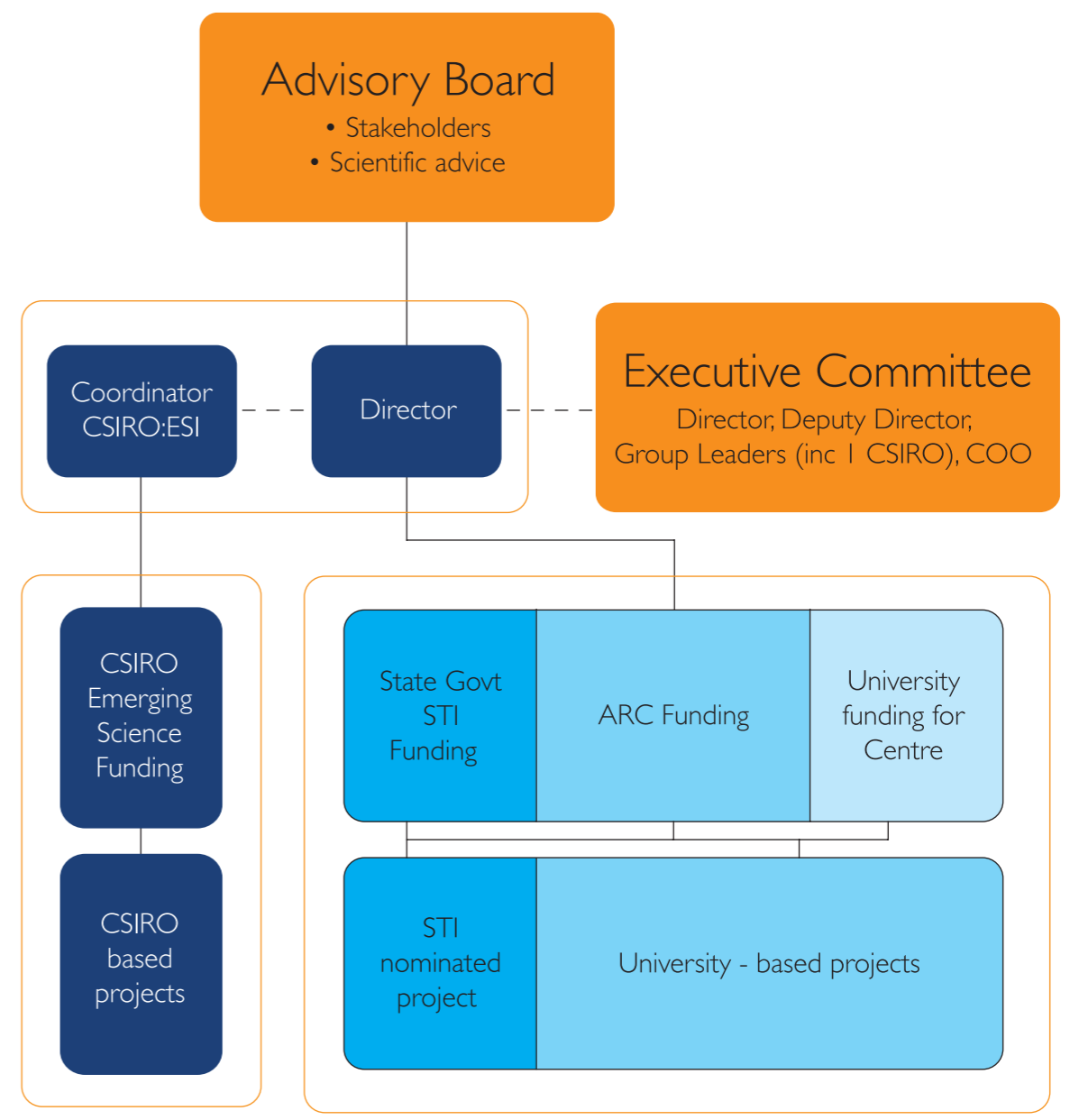
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Professor Robert Lewis, X-ray and Synchrotron Physics, Monash University  
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# MANAGEMENT STRUCTURE



*CXS organisational structure. Note the parallel Emerging Science Initiative stream. This is independently funded and is managed via a system using shared goals and collaborations. There is no transfer of money between CSIRO and the university sector.*

