

ADVANCED RESEARCH TECHNOLOGY LABORATORIES



*Dr John Barlow
Manager
Australian Genome
Research Facility
Melbourne Division*



*Dr Frank Battye
Head
Flow Cytometry
Laboratory*



*Dr Anthony Kyne
Head
Computing and
Bioinformatics Services*



*Dr Frank Köntgen,
Head
Embryonal Stem Cell
Laboratory*



*Ms Judy Smith
Head
DNA Sequencing
Laboratory*

OVERVIEW

The Institute aims to provide state-of-art technologies and equipment for all its researchers. In this digital age these items are extremely costly, in some instances in excess of \$1 million, and are rapidly obsolete as new machines with greater data-handling capacity are developed. Costs are beyond the capacity of any individual scientist or laboratory.

By centralising these sophisticated technologies, all scientists at the Institute can gain access. Expert advice and customisation to particular experimental needs is provided by the scientist-in-charge. Access is also provided to external collaborators on a cost-recovery basis.

AUSTRALIAN GENOME RESEARCH FACILITY

The year ending June 1999 marks the first full financial year of operation of the Australian Genome Research Facility. Without doubt the highlight of this year was the official opening of the AGRF by the Minister for Health, the Hon. Dr. Michael Wooldridge. The opening occurred simultaneously in Melbourne and Brisbane by an ambitious video link between the two cities. Further details of this momentous occasion can be found in the Director's Report.

The primary focus of the AGRF (Melbourne) is microsatellite analysis. Microsatellites are small regions of DNA that can be used to characterise an individual's genetic make-up. By measuring the size of microsatellite DNA researchers can identify genetic regions that are associated with disease. Ultimately, this technology can be used to find actual gene variants that cause complex human diseases. This year we have measured more than 1 million microsatellites for researchers both within the WEHI and throughout the world who are investigating conditions such as hypertension, endometriosis, diabetes, Paget's disease, schizophrenia, and myocardial infarction. In addition, because genomics technology is applicable across the entire biological spectrum we have also worked with DNA samples from sheep, cattle and a variety of plant species.

The second function of the AGRF is to identify mutations in genes that have already been identified and which are known to be associated with a particular disease. In collaboration with Professor Dick Cotton from the Mutation Research Centre at St. Vincent's Hospital we have modified an established laboratory mutation

detection technique for high-throughput analysis. The technique, invented by Professor Cotton, uses specific chemicals that cut DNA at a mutation point. The DNA fragments are then easily detected by electrophoresis. This year we have focussed on detecting mutations in two known breast cancer genes, BrCa I and BrCa II, and on a bank of genes known to be associated with dementia. We expect to be able to offer these two gene screens as aids to diagnosis of these diseases within the coming twelve months.

The AGRF is keeping up-to-date with significant investment in microassay technology, and purchasing an ABI 3700 micro-Capillary sequencing machine.



DNA SEQUENCING LABORATORY (DSL)

The DNA Sequencing Laboratory has once again experienced a large growth in throughput over the past year. The laboratory is now handling approximately 3250 samples per month up from 2330 last year. Well over 25 million bases have been sequenced since the lab was established late in 1996.

Two innovations have allowed the DSL to handle such growth:

- an accounting database has been created,
- an online submission form has automated samplesheet preparation.

The DNA Sequencing Laboratory has become widely regarded as one of the most efficient and effective in Australia. Because of this increased demand current technology is inadequate. We had to decide whether to make a major investment in new equipment or to merge this laboratory, with the AGRF.

By mutual agreement the DSL is being transferred into the AGRF from July 1999.

COMPUTING AND BIOINFORMATICS SERVICES

The bioinformatics services of the Institute continue to grow with the increased pace of the human genome program and other activities. This is reflected in the higher growth of the DNA and Protein sequence databases (by 85% and 28% respectively) and the demand for more specialised genome based databases, particularly in the area of malaria research.

The unit was involved in maintaining the continuity of GDB (Human Genome Database) as it moved from Johns Hopkins University to the Toronto Hospital for Sick Children following the discontinuance of funding from the US Department of Energy, and the subsequent revival of the database.

Other achievements have been to replace the original desktop acquisitions of the last decade, continue replacing backbone segments with 100MB fibre links, maintain a computer network security policy, and to upgrade the computer based control systems of the lecture theatre to permit audio/video broadcasts and transmissions of major events.

The unit continues to monitor possible effects of the year 2000 on the Institute's work. Patches for all systems are installed as they become available. All but 2 systems are now year 2000 compliant. The Institute is relying on the integrity of compliance statements from vendors and suppliers of systems and services.

EMBRYONAL STEM CELL LABORATORY

In the past year, the Embryonal Stem Cell (ES) Laboratory has undergone staff changes which has seen extra blastocyst injectionists trained and additional staff taught the skills and theory relating to gene knock-out technology. The facility has continued a supportive role for the WEHI and wider Australian community, collaborating with two Divisions and nine other Institutes in the past year.

Cre/Lox mediated site-directed gene excision technology is now in routine use. We have generated two conditionally-targeted mouse strains utilising in vivo excision with Cre transgenic mice.

Research within the facility has included germline transmission of two ES cell line strains with a serum-free culture system. The results are being written for publication. In collaboration with Professor Jim McCluskey of Melbourne University, we have used lipofection to introduce yeast artificial chromosomes (YACs) into ES cells to produce transgenic mice.

With the help of Computing Services, a new record keeping database has been developed to enable tracking of projects, detailed project summaries for collaborating scientists, and maintain quality control.

FLOW CYTOMETRY LABORATORY

A remarkable diversity of research applications has characterised the services provided by the Cytometry Laboratory. This extends to both arms of the laboratory's operations; flow cytometry and confocal microscopy.

In flow cytometry, a number of groups have made use of the extended capabilities of our newest cell sorter. The Immunology Division has performed several 3 and 4-way sorting operations in which three or four phenotypically identified sub-populations of dendritic cells have been simultaneously isolated. Many groups have benefited from the 4-fold increase in sorting speed that, on the one hand, has allowed the sorting of rare cells (for example transfected cells identified by fluorescent protein genetic markers) and, on the other, has enabled the purification of less rare cells in far greater numbers than was hitherto possible. A similarly disparate group of analytical operations has been seen, from multi-colour immunofluorescence, through the combination of immunofluorescence with genetic markers to time course studies of intracellular calcium flux.

A revised scheme for management of the large volume of computer data generated by flow cytometry has led to the setting up by the Computer Services group of a new file server and deployment to Institute Macintosh and Windows computers of

the Cytometry Laboratory's own flow cytometry data analysis software.

Confocal microscopy is being re-discovered by increasing numbers of Institute scientists as a powerful analytical tool in areas no less diverse than those seen in flow cytometry. The Development and Neurobiology group has analysed the cell markers associated with differentiation of neural stem cells. Members of the Infection and Immunity Division have used the technique for visualising surface antigens of Leishmania parasites and Leishmania infected mammalian cells. Making full use of the ability of the confocal microscope to optically "slice" a sample, investigators from the Molecular Genetics of Cancer Division have studied the localisation and co-localisation within cells of cell death markers.

Some proportion of our cytometry services has continued to be offered to colleagues in neighbouring institutions, although this has had to be curtailed somewhat in the face of the year's steeply increasing demand from WEHI scientists. This year, flow cytometry analyses have increased by 10% and cell sorting by 42%. Confocal microscopy usage has increased by a remarkable 4-fold.

Staff List

Australian Genome Research Facility

John Barlow, BSc MSc PhD *Mon*, Divisional Manager
Glenn D'Cunha, BSc(Hons) *LaT* PhD *Melb* (from 3/98)
Kelly Ewen, BSc(Hons) *Mon* PhD *LaT*
Garry Myers, BSc (Hons) *Northern Territory* PhD
Syd (from 10/98)
Karl Poetter, PhD *Ohio*
Dianne Arnold, BSc *Melb*, Genotyping Assistant
Malcolm Barker, BSc(Hons) *Manchester*, Information
Technology Services Manager
Timothy Barlow, BSc *Mon*
Nancy De Filippis, BSc (Hons) DipEd *Melb*
Lisa Dube, BSc(Hons) *Victoria* (from 5/99)
Marie Everest, (from 10/98)
Margareta Go, BMedBiolSc *Hogeschool van Utrecht*
Daniel Lucas, BSc(Hons) *Mon* (from 2/99)
Phuong Thuy Nguyen, (from 2/99)
Paige Stevenson, BSc *LaT*
Wayne Ward, BSc(Hons) *LaT*

Computing and Bioinformatics Services

Anthony P Kyne, MSc PhD *Melb*, Head
Vladimir Brusic, MSc *Belgrade* DipCompSci *LaT* (to 7/98)
Janice Coventry, BSc DipEd *Melb*
Miffy Edwards, BA *Mon*
Robert Flegg, MSc PhD *Melb*
Andrew Fleming, BSc PostGradDipCompSci *LaT*
Keith Satterley, BSc DipEd *Melb* DipCompSci *LaT*
Ryan Saunders BA *LaT*
Marina Spaulding, BSc *Tas*
Jim Sun, BSc *Beijing* DipComp MSc *Melb*

DNA Sequencing Laboratory

Judy Smith, BSc(Hons) MSc *Qld*
Melanie O'Keefe, BSc(Hons) *Melb*

ES Cell Laboratory

Frank Köntgen, PhD *Albert-Ludwig*, Head
Louise Barnett, BAppSc *RMIT*
Jodie De Winter, (from 2/99)
Helen Dillon (to 12/98)
Anita Steptoe, BSc(Hons) *UWA* (from 2/99)
Michelle Swift, BAppSc *RMIT*

Flow Cytometry

Frank L Battye, BSc(Hons) PhD *LaT*, Head
Dora Kaminaris, BSc *Mon*
Viki Lapatis
Jennie Parker, BSc(Hons) *Melb*

Staff Notes

Arrivals

Glenn D'Cunha, Genotyping Officer from Quantum
Scientific
Garry Myers, Research Officer from the Menzies School
of Health Research, Darwin

Major Invited Lectures and Exchanges

John Barlow

Invited Lecture, Baker Institute seminars, 10/98
Invited Lecture, Florey Institute seminars, 10/98
Invited Lecture, AMRAD, 10/98
Human Genome Meeting, HGM '99, Brisbane, 3/99
Invited Lecture, Department of Biochemistry, Melbourne
University, 4/99
International Federation of Clinical Chemistry, Florence,
Italy, 6/99, *Poster*

Kelly Ewen

Invited Lecture, Murdoch Institute, 7/98
Boden Conference, Thredbo, 2/99
European Society for Human Genetics, Geneva,
Switzerland, 5/99, *Poster*

Karl Poetter

Cancer Gene Analysis and Mutation detection
Workshop, Melbourne, 7/98.
Invited Lecture, Science 2000, Melbourne, 9/98
Invited Lecture, Mutation Interest Group, Melbourne,
11/98
Invited Lecture, Hereditary Eye Disease Group,
Melbourne, 11/98
Invited Lecture, Department of Microbiology, LaTrobe
University, 5/99



New staff and students: from left, Glenn D'Cunha, Daniel Lucas, Lisa Dube, Marie Everest, and Garry Myers.

Acknowledgement

The Australian Genome Research Facility thanks the Federal Department of Industry, Science and Tourism for the support to establish this facility, and the Department of Health and Community Services in Victoria and The Royal Melbourne Hospital for the provision of space.