

## 10. Facilities

### Computing

#### R. K. Bryan

#### Central Servers

The following systems provide central file serving and backup services which are available to all systems on the laboratory network:

- A high-availability Linux fileserver cluster, comprising:
  - two Compaq Proliant ML370 G2 systems, running Suse Linux and Mission Critical Linux Convolo.
  - Jetstor III Raid array - six Seagate 180GB disks, 900GB useable space, dual Ultra160 SCSI interface. This may be served by either system.
  - Compaq MLS5026 SuperDLT robot. 100GB/tape. Data transfer speed up to 70Gb/hr. Magazine capacity 25 tapes.
  - GForce During the year, a Jetstor SATA416 array was added for Mark Sansom's group. This array holds 16 400GB Maxtor disks, configured as RAID level 6 plus a hot spare, giving 5.2TB of useable space. RAID 6 provides an extra parity disk compared with RAID 5, so two disks may fail in this array before there is a loss of data.
- The ongoing problems with the 1.2TB GForce RI Raid array seem to have been overcome by hosting it on a Compaq Alpha workstation running Compaq Tru64 Unix, instead of an Intel/Linux system.
- A VMS cluster consisting of an AlphaServer 1200 and AlphaStation 500, providing access to about 800GB of RAID disk storage, with a TL891 DLT tape robot. The storage is served to the rest of the laboratory systems using various network protocols (VMScluster, NFS, SMB and AppleTalk). This also runs network services for mail, POP, IMAP2, IMAP4, DHCP and the laboratory web server. The inherent security of the VMS architecture gives greater protection against potential external disruption to these services.
- A 4-processor Compaq AlphaServer 4100 running Compaq Unix, which serves about 50GB of storage, and acts as the NIS master node.
- In addition, two Beowulf PC clusters provide a high-performance computing resource. The first, installed in 2001 for Mark Sansom's group, consists of 32 dual 750 MHz PIII nodes, and the second, a joint resource for the Sansom, Noble and Garman groups which was installed in 2003, consists of 67 dual 2.4GHz PIV nodes, and also 1.8TB of local RAID storage.
- Two further clusters have been added.
  - One in Mark Sansom's group, and located in the Biochemistry Tower, comprising 30 nodes with dual 2.2GHz Dual-Core Opteron processors.
  - A small cluster in Jasper van Thor's group, comprising 7 nodes with dual 2GHz Opteron processors (in the LMB computer room).

The air conditioning unit in the computer room was replaced in May. This unit was part of the original building installation, had become increasingly unreliable and was no longer maintainable. A considerable amount of work was required to remove the old floor-standing unit and associated ducting. The replacements were modern wall-mounted units, resulting in a useful increase of floor space for computer equipment. The work took approximately one month to perform, and during that time the PC clusters had to be shut down as sufficient power and air-conditioning were not available elsewhere, the disk servers were moved to other suitable locations, and the network switches and firewall were kept running in a protected corner.

## **Laboratory Network**

The laboratory network continues to be based on a 100Mbit/sec Fast Ethernet network, plus a Gigabit switch in the computer room which provides a higher-speed connection to several of the central systems. This network is connected via a Firewall (Intel PC running OpenBSD Unix and pf packet filter) to the University 10GB backbone ethernet, which provides access both to other units within the University and to the external Internet connection.

The firewall software was upgraded to OpenBSD version 3.7, which includes several new operating system security features, as well as additional firewalling features. Before the upgrade this system had been running continuously for 462 days, demonstrating the reliability of this architecture.

As a reaction to frequent break-in attempts using ssh, we restricted access using ssh from external location to just two of our systems, except for certain "trusted" locations, such as the ESRF. Hopefully, this will reduce the chances of a break-in by repeated, automated, attempts to guess usernames and passwords, and also make it easier to monitor the accessible systems. We expect to introduce shortly further measures to block automatically external hosts which make multiple ssh connections in a short space of time.

A further 'decant' of Mark Sansom's group to the Biochemistry tower took place in August 2005. Since part of the group had already been in the Biochemistry building since 2003, the required configuration for the firewall and other network services, such as NIS and NFS, was already in place for them to access the laboratory servers transparently. The main task was to assign new network address in cooperation with the Biochemistry Network Manager. In the light of the experience gained in the previous move, this was mostly set up in advance so a desktop system could be moved to its new location at a time convenient for its owner and simply plugged straight into the network without further reconfiguration.

About 150 host addresses are registered on the Laboratory network, which include a wide variety of systems:- the central servers as described above; a large number of desktop systems, including Intel systems running either Windows or Linux, and Apple Macintosh computers; personal laptop computers; and a number of systems dedicated to specific tasks, such as control of the X-ray Area Detectors and EM Image acquisition

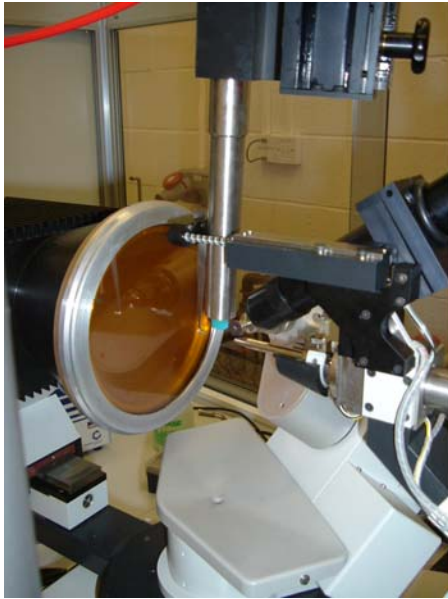
## **X-ray Facilities**

### **Drs. Ed Lowe and Elspeth Garman**

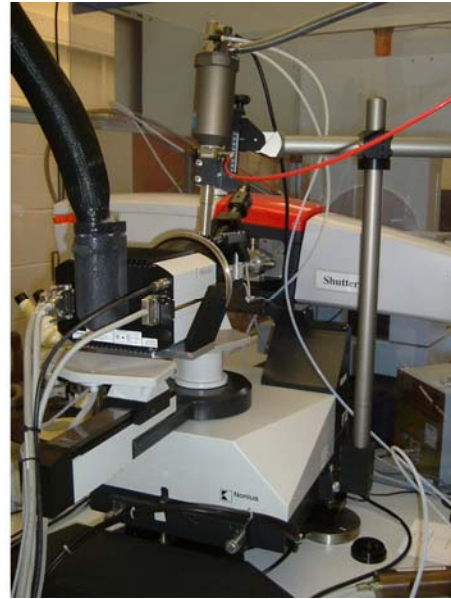
This year we have commissioned and further developed the new X-ray facility centred around our Wellcome Trust funded 2.7 kW Bruker MicroStar X-ray generator with Montel optics on both sides, that was installed in Room T3 on 8/6/04.

The Dunn School of Pathology decommissioned their Rigaku RU300 generator and X-ray equipment in November 2004, and kindly donated their Mar345 Imaging plate detector (plus their 600 series Oxford Cryostream and blue Osmic mirrors) to us. We have installed the detector on the right side of the MicroStar. This has allowed us to leave the Rigaku RU200H facility in T5 complete, thus reducing pressure on the new equipment. This has considerably simplified its commissioning, enabling us to introduce it into full user status in a stress free manner. The necessary radiation safety testing was complete on 18/11/04, and the right hand side then became a user facility. An extra 6cm flight path recently installed between the Montel mirrors and the Mar345 slit box has placed the mirror focus (i.e. the focal point of the X-ray beam) after the Mar slits and 40mm before the crystal position, resulting in a smaller beam at the crystal position.

During the first year of operation, we have had very few technical difficulties with the MicroStar, and those we have had (a noisy bearing [Jan 05] and a malfunctioning shutter [June 05]) have been swiftly rectified by the Bruker engineers, who also carried out a scheduled annual anode rebuild in July 05.

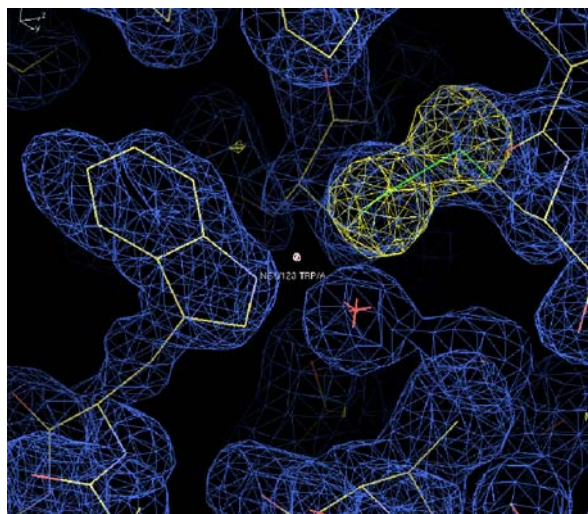


**Figure 1a.** Bruker Platinum 135 CCD Detector with 4-circle KAPPA goniometer mounted on the left hand port of the MicroStar generator



**Figure 1b.** Looking east across the new X-ray facility. The Mar345 imaging plate detector can be seen mounted on the far side.

In early May, Bruker installed a Platinum 135 CCD Detector with 4-circle KAPPA goniometer (see Figure 1a and b). This was commissioned successfully in June and July, with 2 days of in-house training being provided by a Bruker applications scientist. The control and analysis software is crystallographically advanced and easy to use. The instrument is already proving very useful, two new structures having been solved on it in its first 2 months of operation. Professor Martin Noble has collected data from small crystals of mouse CD44 in complex with an oligosaccharide (diamond shaped plates with face edges of  $75\mu\text{m}$  and an approximate thickness of  $15\mu\text{m}$ ) which revealed an important conformational change difficult to observe at a synchrotron source, possibly due to radiation damage. We are currently exploring the potential for carrying out sulphur-SAD phasing using it. The structure of hen egg white lysozyme (HEWL) has been successfully phased using sulphur-SAD data collected using this system and experimental phases are shown in Figure 2. Further sulphur-SAD experiments are planned.



**Figure 2:** Experimental electron density map from HEWL derived from anomalous sulphur data collected on the Bruker Proteum system in T3. The shown in blue is contoured at  $1\sigma$ . The anomalous Fourier map shown in yellow is contoured at  $6\sigma$ .

The radiation safety requirements required to run the two sides of the generator as independent user facilities with separate failsafe circuits were fulfilled and the system was successfully inspected in September.

As noted last year, measurements carried out by Robin Owen using a photodiode calibrated at the ESRF indicate a photon flux at 80% maximum power (40kV / 55mA) of  $7.24 \times 10^8$  photons  $s^{-1}$ , almost 3-fold higher than the  $2.5 \times 10^8$  photons  $s^{-1}$  measured for the RU200H in T5 operating at maximum power. The attached Montel multilayer optics produce a beam of cross section  $150 \times 150 \mu m^2$  at the sample. This small beam size in combination with the higher flux has enabled us to look at much smaller crystals in-house which we are obtaining from robot crystallisation trials. This has resulted in several projects to make faster progress, since small crystals can be tested for diffraction in house as soon as they appear, and optimisation be carried out before synchrotron trips, whereas previously we had to wait to do the first diffraction tests for the next synchrotron visit.

The RU200H generator ('Myrtle') with a Mar345 imaging plate detector and Osmic Optics in T5 ran very smoothly this year, and was heavily used during the commissioning of the new generator. The anode annual rebuild was carried out by us on September 5<sup>th</sup> without incident. Users included numerous members of the LMB, Dr. Ben Berks group and OCMS, and Dr. Arzhang Ardavan's correlated electron systems group from the Clarendon Laboratory collaborating with Robin Owen and Elspeth Garman to investigate the effects of radiation damage on organic superconductors, the results of which have been submitted to Phys Rev Letters.

## **Report on crystallisation facility.**

### **Marc Morgan and Jenny Gibson**

The Oxford LMB crystallisation facility currently has over 50 users and this number is growing due to the acquisition of new technologies designed to improve result throughput and scoring. In late 2002 the LMB purchased a TECAN liquid handling platform with the aim of fully automating the setup of protein crystallisation trials. It was at first necessary to reconfigure the liquid handling system for crystallographic use. Since then the TECAN Genesis 150 has been operating effectively in its new role. We will briefly outline the set-up, level and mode of use. In October 2003, the LMB also acquired a high definition Leica microscope with an automated Prior stage in order to keep pace with the results produced by the robot.



The reconfigured TECAN robot has a unique set-up that allows it to reproducibly pipette a variety of crystallisation screens. The liquid handling arm has a total of 8 tips: 4 standard volume tips, which pipette volumes between 250  $\mu$ l and 0.5  $\mu$ l, and 4 low volume tips which pipette volumes between 1 and 0.25  $\mu$ l. The standard tips are powered by 250  $\mu$ l capacity syringes, and the low volume tips are powered by smaller 50  $\mu$ l NPS (Nano-Pipetting System) syringes.

The original design of the 50  $\mu$ l syringes was identical to that of the larger 250  $\mu$ l syringes: a glass chamber fitted with a Teflon capped aluminium syringe plunger. However, after several months of use, substantial wear and tear on the 50  $\mu$ l syringes was noticed, which in turn significantly impaired the accuracy and reproducibility of low-volume pipetting. In consultation with TECAN the glass-clad aluminium plungers were replaced with more durable ceramic plungers enclosed in a plastic syringe body. These have been a great success and have certainly proved flexible enough to cope with the demands placed on the system.

A number of new crystallisation plates have recently been configured on the robot so that users can choose which plate format they would like to use. Although the Greiner 96 flat bottomed well plates are still the plates of choice for most users, those working with membrane proteins can now use a variety of plates with round or conical bottomed wells. The shape of the well in these plates ensures that a drop containing detergent does not distend over the well surface.

The automated Leica microscope is now fully operational. Users of the microscope can either scan an entire 96 well crystallisation plate, or select an individual well for imaging. Picture quality and the speed of data processing are both highly satisfactory, with the system able to generate 96 high definition images in under 5 minutes. The lens depth of focus is good enough to see the majority of crystalline particulates at the low magnifications required for plate scanning, but also performs excellently at the high magnifications required for more in-depth analysis. It has also been effective in overcoming problems of imaging at very low exposures necessary for viewing light-sensitive proteins. Planned future developments for the microscope include an image/crystallisation database for cataloguing interesting crystallisation characteristics. We are also continually testing new crystallisation plates for improved picture quality, although the images obtained with the current plates are highly satisfactory.



The crystallisation facility boasts an ever-growing number of users, who are now profiting from the new technologies recently installed. The addition of the robot has increased not only throughput, but also output. The full impact of the microscope will become apparent once the crystallisation database is operational.