University of Otago
Te Whare Wananga o Otago

Centre for Gene Research

NEWSLETTER

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Editorial

A little later than planned and apologies for the delay in circulating this newsletter. However, the delay has given us the opportunity to develop our plans for a Centre for Gene Research Retreat next month - the weekend of 17 - 19 June at Orokonui. Many members will recall the very successful retreat held at the end of 1991. Many will also remember the inclement weather. I can assure you that despite the time of year, this year's retreat will be no colder and probably quite a bit warmer than the last one. We have booked accommodation for the nights of Friday, 17, and Saturday, 18 June and some people may wish to join us for the technical sessions as well. Please see the section on the retreat for the draft programme and registration form. These must be in by 7 June so as the catering arrangements can be organised.

In the last Newsletter, I suggested that we should be sorting out the archives of the Centre and requested members to submit lists of papers published since 1990 so that they could be included in the Centre records. To date, two - yes two - people have done so. Even I doubt that these two people are the only members who are publishing! Please send us a list of your appropriate publication - preferably on disk (either PC or Mac) and we will return your disk.

Guest Editorials. This, I hope, will be the last Editorial I write for the Newsletter for some time. This is because I am sure out there somewhere are people who would like to have their say about the operation of the Centre, the University or just science in general. Please volunteer these - we are not restricted to just one Editorial per issue. Next issue, by the way, will be aimed for late July. Please keep the copy rolling in. I have yet to be inundated by it!

Murray Grigor

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News from the DNA Sequencer

Over the last 3 months the sequencing service has been working at full capacity. With its connection to Email the service can now mail text files of DNA sequence information directly to your computer (assuming it is also on network). The Email address is:

deirdre.dobson@stonebow.otago.ac.nz

This should save a great deal of disk handling although the raw data needed for SeqEd analysis must still be transfered by disk.

Please note the tip about template preparation from Sue Galloway in this issue of the newsletter.

Allan Crawford

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Interest Groups Several groups operate round the campus for people with specific interests. Frequently these cross departmental boundaries. If you are the organiser of one of these and would like to have it opened up to other members of the Centre, please let **Brian Monk** (Experimental Oral Biology) know. We will then be able to advertise your meetings in the Newsletter.

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Computer Corner

The idea for Computer Corner has been borrowed shamelessly from Jenny Barna and TIBS where a similar column has been running for some months. The purpose of this column is much the same as that in TIBS; to try and help make a little more sense of computers and computing for those who would like to use them but find them difficult to understand. However, given our somewhat smaller readership, this Computer Corner will be directed towards local computing affairs. To this end I would very much appreciate hearing from people what they would like to read about in this column.

The Internet

The Internet is something that has gained much popular attention recently (I even saw a mention of it in the magazine "More"). The Internet is something that is very hard to pin down. Like a University, it is more an idea than a thing, but clearly it has physical manifestations. Physically,

the Internet is a loosely connected network of computers which are able to talk to and find each other in a very flexible manner. The result of this loose connection is that a number tools can work across the network to connect to other machines and exchange information.

Email

The first of these tools I will discuss, and perhaps the most used, is email or electronic mail. This is a sort of a cross between a telephone conversation and letter. A piece of mail can be sent to a friend or an acquaintance on the other side of the world in just a few minutes, but there is no requirement for them to be actually there to receive the message. Like mail, one needs an address, and while at first site this appears complicated, there is a logic to it. My email address on the Biochemistry Department DECStation is <craigm@sanger.otago.ac.nz>, (where the < and >, by convention, are not part of the address). This address can be broken into parts. The craigm@ indicates the username (craigm) and the @ divides the username from the name of the machine. The rest of the address is an ascending hierarchy of nodes where each part of the address is separated by a stop (.). In ascending order, the address of the DECStation is sanger (the local machine name), otago (the university's address), ac (indicating the academic network, as distinct from commercial or CRI parts of the network), nz (the country name). Addresses in the USA have no country domain and typically end in edu or com.

All users on sanger have access to email and in case their email address will be their username (the name that you user to login to sanger) followed by <@sanger.otago.ac.nz> which is often shown as <login@sanger.otago.ac.nz>. The other thing about email you need to know is how to find the postbox. There are a number of programs on sanger that can be used to send mail but I believe that only two of them are fit for human consumption. For the new user who is not very familiar with Unix and sanger, try pine; the more confident user may prefer to use elm. To use these, type <pine> or <elm> at the prompt and follow the instructions. I do not propose to tell you how to use these mailers here but the Computing Services Centre (CSC) have manuals and help can be had on sanger by typing <man elm> or <man pine>. Those users who have a readily available Macintosh connected to the ethernet may prefer to use a program called Eudora which is graphically oriented and quite powerful. However, this is not so useful for multiple users of the same Macintosh and you will not have the same control over who can read your mail. The CSC have a free manual on how to use Eudora and how you can go about getting an address.

Next time I plan to discuss some of the other tools that are available to users over the Internet,in particular ftp, gopher, tin, telnet and the World Wide Web. If anybody has other suggestions about what should be in this column, I would be happy to hear from you.

Craig Marshall

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Technical Tips

An alternative plasmid miniprep method for automatic sequencer

Some people have been finding that the Magic/Wizard miniprep method has not been working too well in providing good plasmid for the ABI sequencer lately, although it has worked well in the past, and is still working for regular plasmid minipreps if you don't need sequence from them. Here's a quick alternative method that seems to work fine (from Hort Research via the AgResearch lab) Modified from Feliciello and Chinali, Analytical Biochemistry (1993) 212, 394-401

* centrifuge ~3ml O/N culture in 1.5 ml microfuge tube
* wash with 750 µl STE (8% sucrose, 50 mM EDTA, 50 mM Tris pH 8.0)
*resuspend in 185 µl ice-cold TE containing 50 mM glucose
* leave on ice 3-5 min
* add 350 µl lysing soln (0.1 M NaOH, 1% SDS), mix and leave on ice 3-5 min
* add 560 µl ice-cold neutralizing soln (2 M potassium acetate, 1 M acetic acid), shake and leave on ice 5-10 min
* centrifuge full speed in microfuge
* remove 1ml supernatant and add it to 500 µl isopropanol, invert a few times and leave on bench about a minute
* centrifuge full speed and resuspend pellet in 250 µl TE with 10 µg RNAse
* incubate at room temp for 15 min
* add 225 µl 88% isopropanol/0.2 M potassium acetate
* centrifuge, dry pellet and resuspend in 20-50 µl TE

So if the magic has gone out of your

minipreps try this method for a spell. We wish you less bubble, bubble, toil and trouble.

Sue Galloway

(Sue and Murray Broom are looking forward to any tips that you have developed and would like to share with us via this column. Both are in the AgResearch Molecular Biology Unit and can be contacted there.)

Kit comment:

Jean Fleming has provided this report on her experiences with **mRNA isolation.**

Before contemplating buying a kit for mRNA isolation, you should think about how many extractions you will be doing. Many of the kits available are adequate for 2-5 preparations, which make them very expensive if you are contemplating dozens of extractions. I was in this situation in 1989, where I needed a method of mRNA isolation where one person could extract 6 tissue samples in parallel, which would yield RNA good enough for northern blotting in one day. Initially I used Invitrogen's FastTrack™ kits. These proved to be good, but very expensive. Penny Greenwood (Wallaceville) and I devised a method based on the Invitrogen kit, which gave good yields (10-50 µg poly A^+-enriched RNA/g tissue extracted) and was of a quality that was more than adequate for northern blotting, RNase protection or primer extension. This method is published in Fleming *et al.*, J. Molec. Endocrinol. **9**: 207-211 (1992) and I still use it for tissue samples of 0.1-0.5 g. Copies of the "in lab" protocol are available from me.

I have also tried the Micro-FastTrack™ kits from Invitrogen, for extracting samples of bone marrow cells. I had several complete failures with these kits and resorted to using a simple total RNA extraction, using homogenisation in guanidinium isothiocyanate and an acid-phenol-chloroform extraction. After a recent experience with a free sample of BRL's Trizol (Life Technologies), I would now recommend this product for total RNA extraction from small samples.

Selection of poly A^+ RNA from total RNA gets difficult with smaller samples and the risk of "losing everything" becomes substantial. One method which I have used recently involves filling a sterile push column (I used an emptied Nuc-Trap column for labelled probe purification; no, not used!) with oligo dT cellulose equilibrated in 0.5 M NaCl in TE, and basically following the column purification method. I passed total RNA (~800 µg, in 0.5 M NaCl) over the column three times, then washed the column with 5 ml 0.5 M NaCl in TE, followed by 5 ml 0.2 M NaCl in TE. The poly A^+ RNA was eluted with 2 or 3 0.5 ml aliquots of TE and ethanol precipitated. Each sample took roughly 30 min and the yields were 10-30 µg/g tissue extracted, or 1-2% of the total RNA. The results looked reasonable on electrophoresis. To regenerate the column between samples (I did 3) the column was washed with 2 ml 0.2 M NaOH, followed by 10 ml of 0.5 M NaCl in TE, until the pH was neutral again.

Ian Ross adds:

To reiterate Jean's comment that..."the risk of losing everything becomes substantial"...,in my experience the most successful isolations are those with the least handling. We in the Grigor/Tate lab routinely use the method of Chomczynski and Sacchi (Anal. Biochem. **162**, 156-9 (1987)) using homemade reagents; this protocol is available to anyone. I've tried the Promega paramagnetic oligo-dT kit and in our applications found highly variable yields due to vagaries in the magnetic properties.

Below is a quick summary of the range of kits available for mRNA isolation and costs.
(Source; kit name; number of preps; cost)

Promega; PolyAT tract Sys III; 15 (x 100mg), $288

Promega;Sys 1000; 4 (x1 g); $600

Invitrogen; Fast Track; 6; $520
Invitrogen; Micro Fast Track; 20; price n/a

Life Tech; Trizol; 100 ml; $210

Stratagene; Poly A Quik; 4 columns(=12 samples); $780

Bresatec; Ultraspec-II; 50; $500
Bresatec; On Track; 4-5 mg; $540

Integrated Sci; Scigen; 25; $450

Ian Ross edits this section. He can be contacted at 479 7841 or ianr@sanger.otago.ac.nz. If you have

suggestions for kits for our next newsletter please contact him.

Meeting reports

The joint annual conference of the **New Zealand Society for Biochemistry and Molecular Biology and the New Zealand Microbiology Society**, entitled **"Molecules to Microbes II"** took place from May 9 -12 this year at the University of Auckland. A large number attended the conference which was kicked off by a vineyard and wine tour, and the relaxed and social atmosphere prevailed throughout the three day meeting. With concurrent sessions for biochemists and molecular biologists, there was something for everyone in the diversity of topics covered by the programme. Now that molecular biology and microbiology are beginning to share a common frontier, some conference goers had difficulty choosing between sessions.

The major highlights of the conference were the talks given by the plenary speakers, which exhibited tremendous quality and depth in a format that could be interpreted and appreciated by everyone present. Professor Ian Orme from the Colorado State University gave an excellent review of the history of tuberculosis, molecular mechanisms of immune response, and the grave problems we face today with multi-drug resistant strains of this increasingly dangerous pathogen. Paul Reynolds from Hort+Research in Palmerston North gave an impressive account of the extensive ground covered by his research group over the last five years, highlighting the use of immunomolecular techniques in their studies of the biology of root nodule symbiosis. Finally, Professor Bryce Kendrick from the University of Waterloo, Canada, gave a truly inspiring lecture entitled "Why Classify?" - a thousand and one really good reasons why we need more taxonomists, not the least of which included saving the Earth from certain doom!

Also worth a mention is the extremely high quality of the student presentations in the Oral Paper competitions and in the poster sessions of the conference. The very high quality research and impressive presentation effort exhibited in those sessions perhaps made them the most worthwhile of all.

Lastly, the academic and social successes of the conference would not have been possible without the support of the sponsors, who were major contributors in every way (excepting their oversight in not picking me to win the case of wine!).

Jules Horsfield

15th Scientific Meeting of the International Society of Hypertension (ISH) Melbourne, March 20-24, 1994

The first paper to use molecular biology techniques to search for the genetic basis of hypertension was published in Science only five years ago when John Rapp showed the renin gene of the Dahl salt-sensitive rat cosegregated with increased blood pressure in an cross between inbred salt-sensitive (S) and salt-resistant (R) rats. ISH conferences are huge affairs covering all aspects of hypertension research, but the area we were most interesed in, the genetic basis of hypertension and cardiovascular disease related to hypertension, is the fastest growing area of hypertension research. This area was well represented at the Melbourne conference and at a preliminary one-day conference 17 March in Sydney devoted solely to the genetics of hypertension in rats and man. It would be impossible to cover all the topics of interest even restricting the subject area to genetics and slighting other areas of great interest, so this report will only cover highlights.

At the Sydney genetics conference Ted Kurtz of San Francisco reported that, paradoxically, the renin allele of the S rat cosegregated with **decreased** blood pressure in a congenic strain with the S renin allele transferred onto an R background. Brian Morris, the organizer of the Sydney conference, presented results showing an allele of the ACE gene in humans disposes toward early death. Some reseachers were dubious about this, but I think that in a few years we may see that these results are indeed correct and that a version of the ACE gene will underlie abnormal growth in the human vasculature causing effects similar to the cardiomyopathies resulting from lesions of the gene for myosin b chain. A Montreal group presented results in both Sydney and Melbourne showing that the increased DNA synthesis exhibited *in vivo* and *in vitro* by SHR VSMC is accompanied and balanced by increased apoptosis.

For a number of years it has been recognized that exagerated growth of the vasculature can, in and of itself, contribute to hypertension. But now there seems to be a controversy

brewing as to whether increased blood pressure alone causes the cardiovascular hypertrophy which accompanies hypertension or whether there can be cardiovascular hypertrophy independent of increased blood pressure. Our oral presentation was very well received in some quarters probably because the results we have found in the New Zealand genetically hypertensive rat directly support the latter view. In male F_2 rats from our GH X Brown Norway (BN) cross the GH allele of the gene for angiotensin converting enzyme (ACE) strongly cosegregates with increased left ventricular mass but not blood pressure whereas the GH allele of renin cosegregates with **decreased** heart mass also independently of blood pressure in males. Stephen Harrap of Melbourne presented preliminary evidence that the gene for nerve growth factor may independently contribute to cardiovascular hypertrophy in the SHR model.

Currently there are at least two large groups mapping the rat genome in search for the genes underlying cardiovascular disease. Howard Jacob's Boston Massachusetts General Hospital group reported a gene on rat chromosome 1 is linked to a component of renal failure independent of hypertension. Hypertension is well-known to cause renal failure as one of its complications. A Brazilian collaborator reported on a number of genes cosegregating with hypertention in a cross between the SHR and BN.

The pace of work in the genetics of hypertension is currently very fast and furious and therefore a bit overwhelming. I found much of intense interest at the conferecene and was able to met old friends, new people and collaborators with whom I'd been in contact only by E-mail previously. Anybody who wishes may borrow the weighty tome of closely printed abstracts for their perusal.

Jean Harris

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Grants:

Successful FRST applicants this year include from the Centre:

Glenn Buchan	$110,000
Diana Hill	101,000
Clive Ronson	66,000
Murray Grigor	95,000
Patrick Sullivan	88,000
John Harris	100,000
Kevin Farnden	110,000

New Zealand Wool Board, $75,000 to Grant Montgomery (AgResearch) over three years to study genetic markers for wool traits.
MAF Policy, $60,000 to Murray Grigor over three years to study casein synthesis in the possum mammary gland.

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Visitors

Moshe Tal, a professor in Biochemistry from Israel has been visiting Warren Tate at Otago for the last 6 months. Just before his departure last week he agreed to give us this view of modern science in Israel plus a little of the history of his institution:

Higher education in Israel; something about the present and a little about the past

In addition to a highly developed agriculture and industry which has been in exiistence for many years in Israel, there have recently been advancements in two new high-tech fields: biology involving genetic engineering and biotechnology, and engineering mainly software and hardware technologies.

The professional manpower needed for these developments is obtained from our seven universities as well as from those Israelis who have studied abroad. Many major international companies have established research and development centres in Israel: Intel, Microsoft, IBM and National semiconductors. A reason for this is the fact that a number of Israelis have reached top positions in these companies in the US and for various reasons decided to return to Israel. It should also be mentioned, however, that there are quite a few important and very successful high-tech Israeli companies like Elscint, Saitex, Indigo etc.

It is relevant to say something about the beginning of the higher education in Palestine in the period at the beginning of the 20th century. The first academic institution founded was the Technion - now called Technion-Israel Institute of Technology. It was built in Haifa before the first world war but was first opened for studies in the mid 20's. The subjects that were taught were: electrical, mechanical, chemical and civil engineering. Only in the mid 50's were the following science departments opened: mathematics, physics,

chemistry, biology and medicine. Now the Technion is a full-scale university.
Just before the Technion was opened a very heated debate took place which involved the question of language for lectures. This was not a simple problem at all, since the use of Hebrew as a daily language had only started at the end of the 19th century. Needless to say, our language had been kept through the history for many generations but mainly for religious purposes. It was quite obvious that the vocabulary in scientific and technological areas was very poor and not suitable for the purpose of teaching in an academic institution. The debate was therefore, whether or not to use Hebrew, with the assumption that additional vocabulary would be developed or another European language would be chosen. The language that was the candidate for this purpose was German. It is obvious that if this story is told to anybody, including our students, none believe it. At the beginning of the 20th century, German was the most important language in central Europe, moreover, most of the teachers who joined the Technion came from German universities in Germany, Vienna or Prague. Finally it was decided to use Hebrew, which fact accelerated its revival.

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Centre for Gene Research Retreat:
When: 17 - 19 June
Where: Orokonui (near Waitati)
Why: To build up communications and share technology and ideas amongst our members and have a good time
We have put together a programme based on different aspects of modern technology. Each section of the technical programme will contain brief introductions from people doing hands-on work with the technique in question outlining what they are doing and why they chose that particular technique, followed by plenty of discussion time. Come along with your questions in mind.
The topics chosen are:

1. PCR (RT-PCR, PCR cloning, PCR sequencing)
2. Bacterial and yeast expression systems
3. Insect and mammalian expression systems
   (all on Saturday morning)
4. Detection of expression (in situ hybridization, immunohistochemistry)
5. DNA sequence determination analysis and intrepretation.
   (Sunday morning).

As well we will have a mixer on the Friday evening, a session on the future of science funding in New Zealand and its effect on gene research (Saturday afternoon), an after-dinner talk from **Jim Watson** (Auckland) on Molecular Biology in 1999, and a session on cooperation within the Centre (Sunday morning).
The Retreat will finish with lunch on Sunday.

How much:
Accomodation - $18 per night
Registration - $30 ($20 for students)
Sunday session only $10

Please note that registration costs include meals and that people attending will be rostered to assist in the preparation of the meals. You will also need to bring your own sleeping bag and pillow case - but the building will be heated.

Deadline for registations: 7 June - please use form on the back of Newsletter. (Additional forms are available from Murray Grigor or Allan Crawford.)

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Queenstown meeting
People intending to go to this meeting between 14 and 19 August are reminded that the closing date for registrations and abstracts is **17 June**. Kevin Farnden has forms for both if you need them. If you are going, please have a whinge to the organisers about holding the meeting in our term time. This week is the last week of our term and many of our teaching staff find they cannot attend because of teaching commitments.

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For Sale:
Thermal cycler: Perkin Elmer Cetus. AgResearch Lab. Due to our switch to 96-well format for pcr this cycler is now suplus to quirements. In good working order, Any reasonable offers considered.
Contact Allan Crawford (479 7663 or crawforda@agresearch.cri.nz)

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New faces

Jamie Day (Assistant Lecturer in Biochemistry) writes:
My training was very much in plant physiology at the University of Adelaide. For my PhD I examined the control of flowering in two native Australian pant species. This work included extracting and measuring cytokinins using RIA and GC-MS. Changes in cytokinin and carbohydrate concentration were correlated with changes in morphology of the flower during flower development. In 1992 I arrived in New Zealand to take up a post-doctoral position in the Plant Science Group at Auckland University. My work examined the branching pattern of juvenile and adult forms of *Elaeocarpus hookerianus* (pokaka) a native New Zealand tree. This work was basically descriptive; however, I again extracted cytokinins in an attempt to relate cytokinin concentration to changes in branch pattern during ontogeny. I am now employed as an assistant lecturer in the Biochemistry Department at Otago University. This appointment will be short-lived as I have now accepted a FoRST post-doctoral position to work in Kevin Farnden's laboratory. My research will continue from my previous post-doctoral work, examining genetic control of branching patterns in plants.

Julian Eaton-Rye (Lecturer in Biochemistry) writes:
My undergraduate studies were in Botany at the University of Manchester, U.K. I completed a Ph.D. in Plant Physiology in 1987 at the University of Illinois at Urbana-Champaign, U.S.A. My Ph.D. was directed at using chlorophyll fluorescence as a probe to follow electron transport during quinol formation in the photosystem II (PS II) protein complex. Subsequently I went to the National Institute for Basic Biology in Japan to study protein-protein interactions in PS II and I then did a second post-doc at The Center for Early Events in Photosynthesis at Arizona State University. In Arizona I used the cyanobacterium *Synechocystis* 6803 as a model system to introduce oligonucleotide-directed mutations to study structure/function relationships of the PSII core antenna. Before joining the Department of Biochemistry this April I spent a year at Brookhaven National Laboratory on Long Island, New York, working on the phosphorylation of the chlorophyll-binding antenna proteins of PS II. Currently I am looking for research students with an interest in applying molecular genetics and protein chemistry to study photosynthesis. Should you or someone you know be interested please feel free to contact me (Tel: (03) 479 7685).

Christine Piotte (Post-doctoral Fellow in Biochemistry) writes :
I completed my PhD at the National Institute for Research in Agronomy (INRA) in the south of France, my native country, enjoying the sunny weather and working hard on the molecular typing of some phytoparasitic nematodes (genus *Meloidogyne*). I have isolated a few useful tools from repeated DNA (satellite DNA, non-transcribed ribosomic DNA) and managed to set up a field-like experiment based on the hybridization of squashed individual nematodes. I chose my post doc in order to extend my skills in molecular biology and biochemistry and, I must admit, to travel a lot. New Zealand welcomed me through Murray Grigor's group. I am now working on the mysteries of possum milk. The goal of this project funded by FRST is to understand key features about lactation in the brush-tailed possum (*Trichosurus vulpecula*) to enable potential targets for the long term control of this feral marsupial to be identified. I am especially interested in the regulation of the expression of whey proteins. Outside the lab, I take the time to enjoy New Zealand which is an attractive and amazing country.

Welcome back from Study Leave for Committee member, Charles Beresford

Charles writes: I was fortunate to spend nine months as the guest of Robin Carrell (formerly of Christchurch) in the Department of Haematology and Medical Research Council Centre at the Addenbrooke's site in Cambridge. A return to full time bench work was a major shock to the system and very hard on the feet! Regarded by the post-Docs as the "geriatric research assistant", I had a thoroughly enjoyable time amplifying globin genes and directly sequencing the products. A much less tedious way of investigating haemoglobinopathies than tryptic digestion and peptide chromatography! Results with haemoglobin Sherwood Forest served to confirm a new allosteric mechanism for oxygen binding involving movement of chloride ions in the central core of the molecule, much to the delight of Max Perutz.

Robin Carrell's team is making good progress with antithrombin structural/functional relationships, having crystallized the molecule and erected an intriguing hypothesis regarding the structure of the reactive loop and its possible role in loop-sheet polymerisation - a mechanism which may be common to all of the serine protease inhibitor family of molecules.

Visiting scholar status at Trinity College meant some good dinners and better conversation and the University Graduate Club proved a haven from the tourist-crowded pavements of central Cambridge.

Anyone looking for good rental acommodation for central Cambridge and the Addenbrooke's site should contact me as we were particularly fortunate in the place we found in the village of Trumpington.

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An important development on campus this year has been the establishment of the **Developmental Biology Unit.** Here **John Harris** brings us up to date with what is both planned and already happening.

The Developmental Biology Unit is a new collaboration between the University and AgResearch, similar to the Molecular Biology Unit in the department of Biochemistry. It will be housed in the second floor and penthouse of the Wellcome Research Institute and these areas are about to be renovated, with an (optomistic?) completion date of the end of June. University research personnel involved in the new unit come from the departments of Anatomy, Pharmacology and Physiology. Some, including Phil Sheard and John Harris and their staff and students will move their offices to the Wellcome Institute penthouse, while others, including Ian McLennan, Marilyn Duxson and Jean Fleming will continue to be based in their existing space. The unit is under the joint direction of Jimmy Suttie from AgResearch, and John Harris and George Petersen from the university.

The new laboratory space is being designed as special purpose rooms to be shared by different people. With the aid of generous gifts from the Telford Trust and from Medical School bequest funds, a suite of five rooms will be provided with filtered clean air and will include a class 2 biohazard protected room for retroviral and human tissue culture preparations, a room for RNA work, a room for PCR work, a room for DNA preparation and a communal tissue culture room which will house Cathy Allen and David Green's hybridomas as well as other tissue cultures. Other communal facilities include a room housing fluorescence microscopes and SAMBA image analysis computer, a light microscope histology suite and new upgraded housing for the FACS scan equipment.

The major gene research component in the collaborative research is the use of retroviruses to insert lineage markers into cells. Chunyi Li will use these to study growth of deer antlers, and the Harris group is using them to study lineages in skeletal muscle development. Other collaborative work includes Janice Bolter's studies of muscle proteases and their role in development, and Phil Sheard's work on the innervation of deer antlers.

The new laboratory space is designed to be accessible to members of the Medical School research community who need facilities for molecular biology or tissue culture studies, and for this reason the space has not been organized into personal laboratories or offices. People using the facilities of the unit are expected to retain office space elsewhere, and we hope that the downgrading of laboratory space to office space that has occurred in the Wellcome Institute in recent years will soon be reversed.

Research Profile: Genetics

MEREDITH LONGLEY

DR RUSSELL POULTER

The genetics group has a wide range of activities covering bacteria, fungi, plants and animals. Despite this diversity there are common techniques which help to create a cohesive group.

Genetics of *Candida albicans*

Russell Poulter, Margi Butler, Ewan Plant, Tim Goodwin.

The group was involved in the original development of parasexual genetics in this imperfect yeast and the demonstration that it was diploid. Current interests include genetic mapping, including comparative mapping and comparative sequence analysis. Of particular present emphasis is the analysis of the retrotransposons of *Candida albicans*. The group has isolated the first autonomously active *C. albicans* retroelement and completed the sequence of this 6426 bp structure, termed TCa2.

NICHOLAS HOLTON

DR MARGI BUTLER

It is now hoped to extend the analysis of this functional retrotransposon in various ways including a study of the virus like particles. In parallel, attempts are being made to understand the defective, retrotransposon-like, element TCa1.

Molecular genetics of *Bacillus popilliae*

Meredith Longley.

B. popilliae is an obligate pathogen of the New Zealand grass-grub *Costelytra zealandica*. The organism has considerable potential as a biological control agent if only ..... The bacterium is very (very) difficult to culture *in vitro* and, although it sporulates *in vivo* (in larva) it has so far evaded all attempts to induce it to sporulate *in vitro*. An attempt is being made to develop the molecular genetics of this bacterium using a 5.5 kb plasmid found in several New Zealand strains of the bacterium and isolated at Otago. Methods of isolating the plasmid (difficult) have been developed and the structure has been partially cloned and sequenced. There appear to be several lethal genes present on the plasmid which makes cloning a tricky operation. Despite these problems, rapid progress continues (joke).

KAMERON GEEVES

TIMOTHY GOODWIN

Responses of *Nicotiana* spp to plant growth hormones

Nick Holton.

Nicotiana spp have been widely used in plant molecular genetics for a variety of reasons including their ease of transformation. The interspecific cross *N. langsdorfii* x *N. glauca* has been of particular interest because growth of the hybrid plant tissue is hormone independent. In addition the hybrid is of interest because it develops spontaneous tumours. It has been suggested that this unusual behaviour may be due to the presence of genes derived from the bacterium *Agrobacterium rhizogenes* which have been integrated into the genome of *N. glauca* and related species. The present research is concentrated on testing this hypothesis and analysing the *A. rhizogenes* derived Ng*rol*C gene. The evolution of the integrated Ng*rol*C gene is also being studied.

MARSH YEOMAN

EWAN PLANT

Genetics of *Brachydanio rerio*

Marsh Yeoman, Kameron Geeves.

The small tropical fresh-water fish *Brachydanio rerio* (Zebra fish) is becoming widely used as a model for vertebrate differentiation and development. This is, in part, due to the ease with which it can be genetically analysed and modified by transformation. Research is concentrated on evaluating various reporter genes in transformation studies and on the development of a homologous recombination system.

Centre for Gene Research Membership Database

This is now available to anyone who wants it and can be obtained in one a number of ways - none of which involve paper. It has been prepared using ClarisWorks and has been saved as: (a) a ClarisWorks database, (b) a SYLK file (Mac or PC) and (c) a text file. Both (b) and (c) can be opened into any spreadsheet like Excel. To get a copy either send Murray Grigor a blank disk and specify which format you want, or, if you have a Mac on the AppleShare network, access the **Drop Folder** on his computer using the instructions below that Craig Marshall has prepared. Alternatively, you can email him a request and he will send you a text file that can be transferred onto your desk-top computer and accessed into any spreadsheet programme.

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Next Newsletter

The next Newsletter is scheduled for late July. I shall be looking for copy for it from the beginning of July. Here again you can send me files by email or use the Drop Folder on my Mac.

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Drop Boxes and Drop Folders

To assist with compiling items for the newsletter we have set up Macintosh "drop boxes". This means that any user with a Macintosh who is on the University network can place copy directly on the editor's machine. The instructions for setting up and using drop boxes are in the April 1994 edition of the the Campus Lan, and I have included the instructions for using them below. There are two drop boxes relevant to the Centre for Gene Research; both on the machine called "Grigor's Macintosh" in the Biochemistry Apple Talk Zone. One is called "Drop Folder" which is the place to put copy for the newsletter, and the other is called "Database Folder" which contains copies of the CGR database.

Using Drop Boxes

These instructions are lifted from Campus Lan. If you do not have this, a copy of the instructions can be had from Craig Marshall, Rm 132, Biochemistry Department.

1. Launch the Chooser (under the File menu)
2. Click on the Appleshare icon.
3. Click on the zone the computer you want to connect to is on.
4. Find the computer's name in the list on the right and double-click it.
5. Connect as a Guest
6. Find the Drop Folder and double click on it.
7. Close the Chooser

The icon for the drop box folder will be on your desktop, just drop onto it any files you want the other person to have.

To get to the database, go through the same steps but select the Database Folder. Open this folder and copy the database file you want onto your own hard drive. You can then open it using whichever application you wish. Note that you cannot alter the any of files in the actual Database Folder.

To make the whole procedure a lot easier:

1. Go through the steps above so that the icon for the shared folder (drop box) is on your desktop.
2. Click on the folder icon.
3. Choose Make alias from the File menu
4. Put the alias somewhere handy

To use a drop box that has been set up with an alias—drag the file onto the the alias. You can make an alias for each folder or user you commonly share files with and just drag the item to the appropriate alias.

This will only work for users who are on the University Network; so for those users who are not connected up to the network or who are at another site, it is worth thinking of using email to make your submissions. Murray Grigor's email address is grigor@sanger.otago.ac.nz. You may have to work out how to get your priceless prose into your email account, but it is probably easier and better to send it by email than send paper copy and suffer transcription errors.

Craig Marshall

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