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# Biochemistry News

The newsletter of the Department of Biochemistry at the University of Otago

editor: Bronwyn Carlisle

December 2012

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## View from the Corner

Hard to believe that it is December already. The tomorrows seem to have 'leapt' rather than "crept in this petty pace from day to day". But no matter, it has been an eventful year and now is a good time to look back at some of the key events in terms of our staff, students and department.

This year we welcomed Catherine Day and last year Iain Lamont to their roles as Professors of Biochemistry, almost doubling our Professorial staff. We also welcomed Wayne Patrick to our staff. He has joined us from Massey Albany with his wife and fellow researcher, Monica Gerth, and will be carrying out work on protein evolution and design, which is a great addition to our themes of research. Wayne brought with him a coveted Rutherford Discovery Fellowship to help in the development of his research programme. Wayne, along with Anita Dunbier, Lynette Brownfield and Liz Ledgerwood represent four recently appointed academic staff that position the department well for future growth.

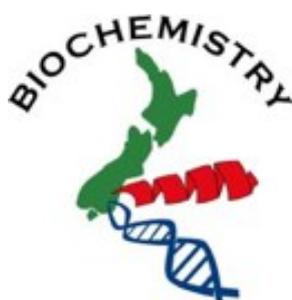
In a related development, we learned last month that our own Peter Mace, has been awarded a Rutherford Discovery Fellowship that will allow him to return to the department in mid-year 2013. Peter was a doctoral student with John and Sue Cutfield and a postdoctoral fellow with Catherine Day during his time at Otago. He is now finishing a successful stint at The Burnham Institute in San Diego carrying out research on proteins that promote chemotherapy resistance in breast cancer. Upon his return, Peter will be setting up a research program studying the structural basis of cell signaling in human disease.



This year we have had two notable retirements that deserve special mention. Sue Cutfield is stepping down as a Senior Research Fellow after spending 31 years at Otago. Sue brought enormous scientific talent and knowledge to her work on protein structure and function. Sue and her husband John Cutfield formed a formidable team and together they established protein crystallography in the South Island. Sue's scientific lineage is remarkable as she was the last graduate student of one of the world's most talented crystallographers, Nobel Laureate Dorothy Hodgkin. Sue's stint with Dorothy, in a group that had a notably Kiwi flavour, including Guy Dodson, Ted and Heather Baker, as well as her husband John, occurred just as the field was forming, and ensured that New Zealand was a party to all the amazing developments that occurred in structural biology since its inception. We will miss Sue greatly, but expect to call on her often for her expertise in protein crystal growth and structure solution.

cont'd over ...

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Just next door to the Cutfield lab is housed Warren Tate's group, which includes his long-time friend and collaborator Liz Poole. Liz has also announced that she is retiring from her position as a Senior Research Fellow at the end of this year. Liz has been with the department for 20 years apart from a brief sojourn in the Deputy Vice-Chancellors office. Liz was involved in several key projects in the group, most recently studies on PEG10, the first human protein found to undergo frameshifting. She and Warren together mentored many outstanding students, helping them tackle their honours and postgraduate degrees, leaving quite a legacy of talented scientific progeny.

Students throughout our department had an encouraging year in 2012. Our enrollments were up in second and third year, while holding steady in first year. During the year we will be graduating 34 students with a Bachelors or a Bachelors with honours degree this year. In terms of post graduates we have had 3 students complete their Masters this year and 12 Doctoral completions. Many of these students were co-authors on manuscripts in 2012.

Papers and grants are always welcome in the department and so far this year our staff members have been named as authors on 52 papers, including several appearing in quite high impact journals. We had some notable grant successes as well with Fast Start Marsdens to Lynette Brownfield and Anita Dunbier and a Standard Marsden to Richard MacKnight. In the Health Research Council round, three of our staff, Stephanie Hughes, Tony Merriman and Mik Black were named co-investigators on successful grants and all are receiving support from these budgets. Sally McCormick received a Heart

Foundation Grant, and Liz Ledgerwood and Sigurd Wilbanks received Lottery Health Grants. Several of our staff received local and Otago based support. In these tough economic times, we have done quite well.

Next year will be my third year chairing the BMS panel of the Marsden Fund and the third year that the fund will not see any increase in funding. Because of this, the success rate for most panels is hovering around 8%, when for sustainable science it needs to be at least twice that rate. More than ever we need to find a way to communicate the importance of the Marsden Fund to the New Zealand government. Many different arguments have been made, some centering on return on investment, both short and long-term. From my perspective one of the most compelling arguments is to stress how the Marsden fund helps us train our students to participate in a world that is increasingly dependent on good science for advancement.

We attract good students from NZ and around the world by having great universities that carry out internationally recognized research. If the cuts in basic science continue, research will continue to suffer and eventually the best programs will move or cease to exist. Without world-class researchers, the universities will suffer and students will be less well trained. Many of our best students may choose to study elsewhere. I hope that New Zealand will address this science funding dilemma in a way that preserves the investigator driven science that drives world class research. That is the real scientific challenge we face.

*Kent Krause*



### **Department Addition**

Lynette Brownfield's baby, James Luke McCurdy, was born at 8.55 pm Sunday 25 Nov. He weighs 3.37 kg (7.7 lb) and is 52 cm long. Rather cute too!

I should add that Lynette's husband Thys also had something to do with this.

## Recent Publications

New since last newsletter as found by literature search, please let me know if I've missed any out.

### Xiaowei Sylvia Chen and Chris M Brown

Computational identification of new structured cis-regulatory elements in the 3'-untranslated region of human protein coding genes.

Messenger ribonucleic acids (RNAs) contain a large number of cis-regulatory RNA elements that function in many types of post-transcriptional regulation. These cis-regulatory elements are often characterized by conserved structures and/or sequences. Although some classes are well known, given the wide range of RNA-interacting proteins in eukaryotes, it is likely that many new classes of cis-regulatory elements are yet to be discovered. An approach to this is to use computational methods that have the advantage of analysing genomic data, particularly comparative data on a large scale. In this study, a set of structural discovery algorithms was applied followed by support vector machine (SVM) classification. We trained a new classification model (CisRNA-SVM) on a set of known structured cis-regulatory elements from 3'-untranslated regions (UTRs) and successfully distinguished these and groups of cis-regulatory elements not been strained on from control genomic and shuffled sequences. The new method outperformed previous methods in classification of cis-regulatory RNA elements. This model was then used to predict new elements from cross-species conserved regions of human 3'-UTRs. Clustering of these elements identified new classes of potential cis-regulatory elements. The model, training and testing sets and novel human predictions are available at: <http://mRNA.otago.ac.nz/CisRNA-SVM>.

*Nucleic Acids Research*, 2012

### C.J. Pemberton, M Siriwardena, T Kleffmann, P Ruygrok, S C Palmer, T.G. Yandle, and A.M. Richards

First identification of circulating prepro-A-type natriuretic peptide (preproANP) signal peptide fragments in humans: Initial assessment as cardiovascular biomarkers.

*Clinical Chemistry*, 2012 vol. 58 (4) pp. 757-767

### R W Bentley, D A Keown, R B Gearry, V.A. Cameron, J Keenan, R L Roberts, and A S Day

Vitamin D receptor polymorphisms in colorectal cancer in New Zealand: An association study.

*New Zealand Medical Journal*, 2012 vol. 125 (1356) pp. 47-51

### G E Evans, J A Martínez-Conejero, G T M Phillipson, C Simón, L A McNoe, P.H. Sykes, J A Horcajadas, E Y N Lam, C.G. Print, I L Sin, and J.J. Evans

Gene and protein expression signature of endometrial glandular and stromal compartments during the window of implantation.

*Fertility and sterility*, 2012 vol. 97 (6) pp. 1365-1373.e2

### Robert D Fagerlund, Poh Ling Ooi, and Sigurd M Wilbanks

Soluble expression and purification of tumor suppressor WT1 and its zinc finger domain.

Full length murine WT1 and its zinc finger domain were separately inserted into Escherichia coli expression vectors with various fusion tags on either terminus by Gateway technology (Invitrogen) and expression of soluble protein was assessed. Fusion proteins including the four zinc finger domains of WT1 were used to optimize expression and purification conditions and to characterize WT1:DNA interactions in the absence of WT1:WT1 interactions. Zinc finger protein for in vitro characterization was prepared by IMAC purification of WT1 residues 321-443 with a thioredoxin-hexahistidine N-terminal fusion, followed by 3C protease cleavage to liberate the zinc fingers and cation exchange chromatography to isolate the zinc fingers and reduce the level of the truncated forms. Titration of zinc finger domain with a binding site from the PDGFA promoter gave a  $K(d)$  of  $100 \pm 30 \text{ nM}$  for the -KTS isoform and  $130 \pm 40 \text{ nM}$  for +KTS for the isoform. The zinc finger domain was also co-crystallized with a double-stranded DNA oligonucleotide, yielding crystals that diffract to  $5.5 \text{ \AA}$ . Using protocols established for the zinc finger domain, we expressed soluble full-length WT1 with an N-terminal thioredoxin domain and purified the fusion protein by IMAC. In electro-mobility shift assays, purified full-length WT1 bound double-stranded oligonucleotides containing known WT1 binding sites, but not control oligonucleotides. Two molecules of WT1 bind an oligonucleotide presenting the full PDGFA promoter, demonstrating that active full-length WT1 can be produced in E. coli and used to investigate WT1 dimerization in complex with DNA in vitro.

*Protein Expression and Purification*, 2012

**C E Caldon, C M Sergio, J Kang, A Muthukaruppan, M N Boersma, A Stone, J Barraclough, C S Lee, M A Black, L.D. Miller, J M Gee, R I Nicholson, R L Sutherland, C.G. Print, and E A Musgrove**

Cyclin E2 overexpression is associated with endocrine resistance but not insensitivity to CDK2 inhibition in human breast cancer cells.

*Molecular Cancer Therapeutics*, 2012 vol. 11 (7) pp. 1488-1499

**M Wilsher, L Voight, D Milne, M Teh, N Good, J Kolbe, M Williams, K Pui, T Merriman, K Sidhu, and N Dalbeth**

Prevalence of airway and parenchymal abnormalities in newly diagnosed rheumatoid arthritis.

*Respiratory Medicine*, 2012

**Reagan M Jarvis, Stephanie M Hughes, and Elizabeth C Ledgerwood**

Peroxiredoxin 1 functions as a signal peroxidase to receive, transduce, and transmit peroxide signals in mammalian cells.

Hydrogen peroxide is widely viewed as the main second messenger in redox signaling, and it has been proposed that deactivation of the antioxidant peroxiredoxin (Prdx) enzymes allows free peroxide to accumulate and directly oxidize target proteins (the floodgate model). We assessed the role of cytosolic Prdx's 1 and 2 in peroxide-induced activation of the apoptosis signaling kinase 1 (ASK1)/p38 signaling pathway, in which oxidation of ASK1 is required for phosphorylation of p38. In response to peroxide, Prdx1 catalyzed oxidation of ASK1 to a disulfide-linked multimer, and this occurred via transient formation of a Prdx1-ASK1 mixed disulfide intermediate. Oxidation of ASK1 and phosphorylation of p38 were inhibited by knockdown of Prdx1, but also by overexpression of Prdx2. This suggests that these two cytosolic Prdx's have distinct roles in the cellular peroxide response and compete for available peroxide substrate. These data imply that Prdx1 can function as a peroxide receptor in response to extracellular H<sub>2</sub>O<sub>2</sub>, receiving the peroxide signal and transducing it into a disulfide bond that is subsequently transmitted to the substrate, ASK1, resulting in p38 phosphorylation. Interestingly, in response to peroxide, Prdx1 and Prdx3 transiently formed reducible higher molecular weight complexes, suggesting that multiple proteins are targets for Prdx-mediated oxidation via a disulfide-exchange mechanism. This model of active peroxide signal distribution via disulfide exchange is consistent with Prdx function in yeast and explains how peroxides may trigger specific disulfide bond formation in mammalian cells.

*Free Radical Biology And Medicine*, 2012

**Jacob J Lamb, Julian J Eaton-Rye, and Martin F Hohmann-Marriott**

An LED-based fluorometer for chlorophyll quantification in the laboratory and in the field.

The chlorophyll content is an important experimental parameter in agronomy and plant biology research. In this report, we explore the feasibility of determining total concentration of extracts containing chlorophyll a and chlorophyll b by chlorophyll fluorescence. We found that an excitation at 457 nm results in the same integrated fluorescence emission for a molecule of chlorophyll a and a molecule of chlorophyll b. The fluorescence yield induced by 457 nm is therefore proportional to total molar chlorophyll concentration. Based on this observation, we designed an instrument to determine total chlorophyll concentrations. A single light emitting diode (LED) is used to excite chlorophyll extracts. After passing through a long-pass filter, the fluorescence emission is assessed by a photodiode. We demonstrate that this instrument facilitates the determination of total chlorophyll concentrations. We further extended the functionality of the instrument by including LEDs emitting at 435 and 470 nm wavelengths, thereby preferentially exciting chlorophyll a and chlorophyll b. This instrument can be used to determine chlorophyll a and chlorophyll b concentrations in a variety of organisms containing different ratios of chlorophylls. Monte-Carlo simulations are in agreement with experimental data such that a precise determination of chlorophyll concentrations in carotenoid-containing biological samples containing a concentration of less than 5 nmol/mL total chlorophyll can be achieved.

*Photosynthesis Research*, 2012

**J P S Chua, E J S Wallace, J A Yardley, E J Duncan, P K Dearden, and T C Summerfield**

Gene expression indicates a zone of heterocyst differentiation within the thallus of the cyanolichen *Pseudocyphellaria crocata*.

*New Phytologist*, 2012

**P O'Sullivan, K Sharples, M Dalphin, P Davidson, P Gilling, L Cambridge, J Harvey, T Toro, N Giles, C Luxmanan, C F Alves, H-S Yoon, V Hinder, J Masters, A Kennedy-Smith, T Beaven, and P J Guilford**

A multigene urine test for the detection and stratification of bladder cancer in patients presenting with hematuria.

*Journal of Urology*, 2012 vol. 188 (3) pp. 741-747

**Megan J Wilson and Peter K Dearden**

Pair-Rule Gene Orthologues Have Unexpected Maternal Roles in the Honeybee (*Apis mellifera*).

Pair-rule genes are a class of segmentation genes first identified in *Drosophila melanogaster*. In *Drosophila*, these genes act to translate non-periodic information produced by the overlapping patterns of gap gene expression into patterns of gene expression in every other segment. While pair-rule genes are, for the most part, conserved in metazoans, their function in pair-rule patterning is not. Many of these genes do, however, regulate segmentation in arthropods and do so with dual-segment periodicity. Here we examine the expression and function of honeybee orthologues of *Drosophila* pair-rule genes. Knockdown of the expression of these genes leads to extensive patterning defects, implying that they act in early patterning, as well as segmentation in honeybee embryos. We show that these pair-rule gene orthologues indeed regulate the expression of honeybee maternal and gap genes implying roles in maternal patterning of the honeybee embryo.

PLoS ONE, 2012 vol. 7 (9) p. e46490

**J L Ludgate, G Le Mée, R Fukuzawa, E J Rodger, R J Weeks, A E Reeve, and I M Morison**

Global demethylation in loss of imprinting subtype of wilms tumor.

*Genes, Chromosomes and Cancer*, 2012

**Minh Ha, Alaa El-Din Bekhit, Alan Carne, and David L Hopkins**

Characterisation of kiwifruit and asparagus enzyme extracts, and their activities toward meat proteins.

Two plant enzyme extracts from kiwifruit and asparagus were evaluated for their ability to hydrolyse commercially available substrates and proteins present in both beef connective tissue and topside myofibrillar extracts. The results show significant differences in protease activity depending on the assay used. Protease assays with connective tissue and meat myofibrillar extracts provide a more realistic evaluation of the potential of the enzymes for application in meat tenderization. Overall, the kiwifruit protease extract was found to be more effective at hydrolysing myofibrillar and collagen proteins than the asparagus protease extract. The two protease extracts appeared to target meat myofibrillar and collagen proteins differently, suggesting the potential of a synergistic effect of these proteases in improving the tenderness of specific cuts of meat, based on their intrinsic protein composition.

*Food Chemistry*, 2013 vol. 136 (2) pp. 989-998

**Philip C Robinson, Tony R Merriman, Peter Herbison, and John Highton**

Hospital admissions associated with gout and their co-morbidities in New Zealand and England 1999-2009.

**Objectives.** To describe the national demographics, co-morbidities and mortality of admissions associated with gout in New Zealand (NZ) from 1999 to 2009 and compare this with English gout admission data from the same period. **Methods.** The characteristics of all admissions due to or complicated by gout in NZ from 1999 to 2009 were analysed. These findings were then compared with the wider NZ population and the English National Health Service (NHS) gout admission rates from 1999 to 2009. **Results.** There were 10 241 admissions due to gout (group A) and 34 318 admissions complicated by gout (group B) in NZ from 1999 to 2009. There were 32 741 admissions due to gout in England over the same period. Gout admissions rose at 5.5% per year in NZ and at 7.2% per year in England over the study period. NZ gout patients admitted to hospital were more likely to be Māori or a Pacific Islander and had 3-7 co-morbidities. Multiple admissions were common with 1479 NZ gout patients admitted more than once. Co-morbidities varied between the NZ groups A and B: hypertension (19-39%), renal disease (16-27%) and diabetes mellitus (20-27%) were common. Heart failure (27.6%) and cardiovascular disease (39.1%) were common in those who had gout complicating their hospital admission. This group also had poorer survival compared with those admitted primarily for gout. **Conclusion.** This is the first study to describe the epidemiology of admissions associated with gout across an entire country. Admissions are rising in both countries studied and those admitted in NZ have a high rate of co-morbidity and re-admission.

*Rheumatology* (Oxford, England), 2012

**D J Smith, G J Anderson, I L Lamont, P Masel, S C Bell, and D W Reid**

Accurate assessment of systemic iron status in cystic fibrosis will avoid the hazards of inappropriate iron supplementation.

*Journal of cystic fibrosis : official journal of the European Cystic Fibrosis Society*, 2012

**G F Kelso, A Maroz, H M Cochemé, A Logan, T A Prime, A V Peskin, C.C. Winterbourn, A M James, M F Ross, S Brooker, C M Porteous, R F Anderson, M.P. Murphy, and R.A.J. Smith**

A mitochondria-targeted macrocyclic Mn(II) superoxide dismutase mimetic.

*Chemistry and Biology*, 2012 vol. 19 (10) pp. 1237-1246

**Harold S Bernhardt**

The RNA world hypothesis: the worst theory of the early evolution of life (except for all the others)a.

ABSTRACT: The problems associated with the RNA world hypothesis are well known. In the following I discuss some of these difficulties, some of the alternative hypotheses that have been proposed, and some of the problems with these alternative models. From a biosynthetic - as well as, arguably, evolutionary - perspective, DNA is a modified RNA, and so the chicken-and-egg dilemma of "which came first?" boils down to a choice between RNA and protein. This is not just a question of cause and effect, but also one of statistical likelihood, as the chance of two such different types of macromolecule arising simultaneously would appear unlikely. The RNA world hypothesis is an example of a 'top down' (or should it be 'present back?') approach to early evolution: how can we simplify modern biological systems to give a plausible evolutionary pathway that preserves continuity of function? The discovery that RNA possesses catalytic ability provides a potential solution: a single macromolecule could have originally carried out both replication and catalysis. RNA - which constitutes the genome of RNA viruses, and catalyzes peptide synthesis on the ribosome - could have been both the chicken and the egg! However, the following objections have been raised to the RNA world hypothesis: (i) RNA is too complex a molecule to have arisen prebiotically; (ii) RNA is inherently unstable; (iii) catalysis is a relatively rare property of long RNA sequences only; and (iv) the catalytic repertoire of RNA is too limited. I will offer some possible responses to these objections in the light of work by our and other labs. Finally, I will critically discuss an alternative theory to the RNA world hypothesis known as 'proteins first', which holds that proteins either preceded RNA in evolution, or - at the very least - that proteins and RNA coevolved. I will argue that, while theoretically possible, such a hypothesis is probably unprovable, and that the RNA world hypothesis, although far from perfect or complete, is the best we currently have to help understand the

backstory to contemporary biology. REVIEWERS: This article was reviewed by Eugene Koonin, Anthony Poole and Michael Yarus (nominated by Laura Landweber).

*Biology direct*, 2012 vol. 7 p. 23

**Karen Knapp, Matthias Zebisch, Jan Pippel, Ali El-Tayeb, Christa E Müller, and Norbert Sträter**

Crystal Structure of the Human Ecto-5'-Nucleotidase (CD73): Insights into the Regulation of Purinergic Signaling.

In vertebrates ecto-5'-nucleotidase (e5NT) catalyzes the hydrolysis of extracellular AMP to adenosine and represents the major control point for extracellular adenosine levels. Due to its pivotal role for activation of P1 adenosine receptors, e5NT has emerged as an appealing drug target for treatment of inflammation, chronic pain, hypoxia, and cancer. Crystal structures of the dimeric human e5NT reveal an extensive 114° conformational switch between the open and closed forms of the enzyme. The dimerization interface is formed by the C-terminal domains and exhibits interchain motions of up to 13°. Complex structures with adenosine and AMPCP indicate that structural control of the domain movement determines the selectivity for monophosphate nucleotides. Binding modes of nucleotide-derived and flavonoid-based compounds complexed to the C-terminal domain in the open form reveal an additional binding pocket of ~210 Å<sup>3</sup> that might be explored to design more potent inhibitors.

*Structure* (London, England : 1993), 2012

**A Chatterjee, E J Rodger, P A Stockwell, R J Weeks, and I M Morison**

Technical considerations for reduced representation bisulfite sequencing with multiplexed libraries.

*Journal of Biomedicine and Biotechnology*, 2012 vol. 2012



Mel Kennedy had a big birthday party in the Reading Room this year.

The birthday wasn't a terribly significant one as far as I can tell, but the party also marked Mel's moving into the reception area, which followed from a rearrangement of printing facilities in the Department.

## News from Around the Department

### Cancer Genetics

Our news comes with it's Paua undies on the outside! Combine some awesome t-shirts, a whanau of genetic mutants, 30 runners and 30 supporters for the Moro Marathon, some Paua Rangers (complete with canine mascot) at Mud, Sweat and Tears (soon to be seen again at the Rail-Trail Duathlon), Bradley Wiggins, a unicycle, a touch team, a cricket team and an indoor netball team and the Centre for Translational Cancer Research (ctcr.otago.ac.nz) have taken the ODT and Sky TV by storm!



The notorious leader of the Murdoch's Miscreant Mos was hesitant to allow his image to be published – however two of the much maligned but madly motivated, mesmerisingly mustachioed Mo-vember participants were



captured on film showing their malice and machismo! A standing mo-ivation to all those making marathon efforts to raise awareness of men's physical and mental health issues!

Suji has planned her escape from Dunedin's cold weather, it involves marriage and PhD submission – the oral will be the final task before she boards a plane to warmer climes. Best of luck from all of us, Suji.

We've gained more fun folk in the lab over the last few months: Lux has been saved from plants, George has already had a syndrome named after him, Rob Day has moved al they way from NZGL and we've managed to tempt back 2 of our 4th years – Chris has signed up for summer with us, continuing to fight valiant battles against R (pronounced Arrgh!) and Henry is due to set up a fencing business come February. Big waves and wishes of success to Tanya & Valerie who are off to face the wide world. Our final strawberries on the pavlova are new recruit Andrew and returning apprentice Briar who are here for the summer. Anita's faststart grant is going to be bringing some wee furry white (honorary) lab members too!



**\*\*Shameless plug:** If anyone would like a breathable, wicking t-shirt (presented with support from the ever awesome Lab Supply, Millennium Science, Raylab and Ngaio), tell Tanis by Dec 14th and we'll see you well dressed!\*\*

Laboratory for Cold Adaptation

A peripatetic few months for many lab members. Anna and Noelia returned to parts European to visit family for the boreal summer and to escape the balmy Dunedin winter. Abhishek et al. attended the Protein Society conference in Sand Diego, James visited Rotorua and Melanie spent some time in Auckland in between oyster analyses. I went to Christchurch and to Wellington.

Other changes in the lab include Lincoln handing in his thesis and Stephen closing in on a final draft of his, and several papers progressing through drafts to submission. Acceptance will prompt suitable celebrations.

We also have some well-established lab animals: James's *Tenebrio* colony goes from strength to strength with the invaluable help of Sarah (beetle sorting) and Gary (who built proper tops for the cages). We also now have cultures of *Artemia* who will be giving their all in the pursuit of a structure for their haemoglobin: A project that started more than 30 years ago.



Brendon Lee has started a Summer Studentship looking for ice active proteins in a winter active grass grub beetle (from my garden) and has some very encouraging preliminary results. I'm not sure that will help in limiting their winter depredations but there is a certain frisson in turning them into beetle soup.

Craig

Maternity, Migration, Marsdens and Medicago

Macknight and Brownfield Lab personnel have had several successes and conference visits of late. Rowan Herridge aced his PhD oral and has since made the necessary final corrections. The hard bound copies are looking particularly swish. Rowan is one of three Lab 308 PhDs to graduate in December, along with Lux and Simon Jackson from JER lab. In the mean time, Rowan is writing papers and preparing necessary paperwork to become a desirable alien in the USA - he's got a post-doc at the prestigious Cold Spring Harbor Laboratory near New York (of Watson, Delbruck and Luria fame), in the lab of Rob Martienssen. All the very best, Rowan.

Richard was successful in the latest Marsden round, gaining \$910k over three years for *Molecular understanding of flowering time regulation in legumes*, which includes some exciting genome wide association studies to unravel the why different wild *Medicago* populations flower at different times. Lynette is due to give birth to her first baby (to be named JASON?) (see p 2) is now on maternity leave, which may or may not give her a little bit of time to ponder her impending discoveries with her Marsden FastStart, \$345k over three years for *The role of asymmetric division in male germ line specification in flowering plants*. Ben Peters, Lynette's Hons student from this year, has enjoyed working with JASON so much that he's staying on for a summer studentship in the lab. WenHann has completed his Hons thesis for Plant Biotechnology and Julianne a Genetics PGDip. All three are looking to stay on for additional study next year. We welcome summer student Elsie Jacobson to the lab, who is doing *Medicago* flowering time and assisting with *Arabidopsis* seed development, sprouting from Rowan's PhD work. We also farewell Jane Campbell, who is retiring in December, and thank her for all her work in the lab and plant room - we will all miss her.

Richard, Manda and Jared have been to various conferences in September. Manda attended the Roche user group meeting in Taupo and enjoyed some North Island scenery. Richard was at the International Conference for Agricultural Biotechnology in Rotorua. A particular highlight for Richard was being seated nearby some hardcore anti-GE types at the conference dinner. Jared has also been busy this half of the year as he looks towards completion of his Master's Degree. Jared recently attended the ComBio conference in Adelaide, notably winning the most prestigious award available to poster-presenters of plant-science-related work in New Zealand - The NZSPB student poster prize (an honour shared by a few past Macknight-labbers; you are in an exclusive club now, Jared). Jared was also lucky enough to meet (read: schmooze) with some big names in the field from around the world, so the conference was a big success for him, and hopefully the first of many.

Rowan and Jared

## Day Lab

People often say every beginning leads to an end. I feel like this can't be the whole truth because nature doesn't work that way. We know this from studying metabolic pathways. With some pathways, we know what comes in and what goes out, but a lot of pathways are cyclical (like the famous citric acid cycle). So where's the start? Where's the end? There's no start or an end. So doesn't it mean that the pathway is pointless because there's no specific "goals" to achieve? Well ... on the contrary! They give us useful chemicals with reducing power that can ultimately sustain our existence just by maintaining the flux of intermediates through the pathways time and again. And before you ask ... No, I haven't been smoking anything. Let's just say that I've had a lot of time thinking about life, the nature of existence, the quantum nature of the universe and great pastas while writing my thesis (sadly, the answer is not always 42!!! D: ).

The Day lab has been learning about the cyclical nature of things first hand. A couple of new members have just joined us. A few months ago, we welcomed Martina who flew over from northern Italy to start a PhD project. Recently, Adam from Canada landed on the land of the long white cloud with his family to take a postdoc position. Unfortunately for him, there are no real Hobbits around here to take pictures of (or ... is there?! D: ).

Sadly, for one of us, it's time to venture into the unknown (into a dark abyss of horror called "Melbourne" mwahuahuahahaha!!! just kidding ... : p It's not that bad. There's a lot of food there : ). For Gabby, the time has come for her to take her life to another level. A PhD project is now on her mind and I heard the guys over there are pretty keen on having her in their lab. We wish her good luck! And there's bound to be interesting things happening in the next few years! Yayy!!

We also welcome Jess who joined us not long ago to start a summer project. Nowadays, the main thing on her plate is cloning (No! Not cloning!!!). Don't worry, it'll be worth it in the long run. Gabby's been awesome with showing her what to do and where to find things.

Josh has been chasing a package containing samples for NMR but somehow they got stuck in an Aussies' immigration depot somewhere. Fingers crossed ... In the mean time, he has done a lot of thinking and got his data together to write a paper. Good luck for both!

Since I've been sitting in our little office for a while, I noticed that Mat and Yoshio always ate relatively healthy and balanced lunches (Is this what married life is like?). Only Georgina accompanied me munching them cookies. And here I am thinking about doughnuts again while I'm writing this ... Anyway, for many of us, there will generally be a larger amount of food towards the end of December. Enjoy responsibly and have a great rest of the year! Remember, what goes around comes around, and the end of something always marks the beginning of something else. Perhaps, this is a good time to think about how we're going to do things differently for the better next year. Take care.

Rhesa (for Lab 223)

## Krause Lab



Two new summer students have joined the Krause lab. Welcome to Galen Correy and Emily Davis! Emily is from Napier, Hawkes Bay. She is currently a Biochemistry major and has just finished 3<sup>rd</sup> year at Uni. Her summer project is to work on cloning and expressing alanine racemase and glutamate racemase from *Acinetobacter baumannii* with the assistance of senior graduate student Emma Scalletti. Emily loves to travel when not studying.

Our second summer student Galen Correy is from Hobart, Tasmania and is studying Chemistry and Philosophy at the Australian National University in Canberra. He is visiting Otago on a Dean's prestigious summer scholarship. For his project he is working on determining the three-dimensional structure of Apobec 3G, and is working with the assistance of our experienced postdoctoral fellow, Karen Knapp. Apart from study, Galen enjoys hiking and biking.



Michele Krause

Dearden Lab

Hollah.

The last few months have gone by with enormous speed, and the time is rapidly approaching when Christmas carols are again **tolerated** in the lab. The big conferences for the year have been and gone. Peter, Liz, Megan and Ajay attended the European Evolution and Development meeting in Lisbon. Everyone gave awesome talks in Lisbon, and we all had a great time and ate too much meat. Peter and Liz also managed to fit in a trip to visit collaborators in Oxford and Cambridge (Liz stayed in the bedroom of George Darwin (son of Charles)). Liz then went to a conference in Brazil where she was an invited speaker (and again ate too much meat at a Churrasco!).

But more importantly the Dearden Lab has experienced some changes of late, changes and milestones worthy of much celebration!

In September Rosannah passed her PhD viva with minor corrections, inducing impromptu whiskey drinking in the reading room and a hilarious lab dinner that night. Sarah passed hers in the same manner in November and both shall be graduating at the ceremony in December. Collective lab hearts are breaking at the thought of their departure.

Speaking of departure and breaking hearts, Frano, our Bioc honours student has finished all his exams and returned to North Otago for stimulating potato picking before heading off on a world tour. Like all children of the Dearden lab, he is missed already.

Meaghan filed her minor corrections for her MSc and printed her thesis for hard binding along with Sarah and Rosannah. Collectively keeping the bindery in business. Meaghan too will be graduating in the December ceremony.

And in non-departure, out genetics honours student for the year, Julie, was brilliant enough to gain a summer studentship and is remaining amongst our hallowed halls for the summer. So delayed goodbyes in this regard, but another to add to the graduation ceremony in December.

Our second summer student, Mackenzie, has joined us in a bit of summer preparation for her Biomedsci honours year next year and already seems to be fitting into the lab very well indeed. We have also had Anat arrive from Israel, who is spending a couple of months in the lab working on the Aphid project. Again in the arrivals lounge, we have Jodi from the prep room with us for the summer working with Cridge, who, incidentally, has settled in like he never left.

Lucy-Amy, our fly postdoc, gave birth to a beautiful wee girl called Josie in early October and we are looking forward to coffee dates with our youngest lab minion. Unfortunately (or otherwise) due to maternity leave she is unable to attend this year's NRCGD Symposium (In *Palmy...*) which Megan, Ajay, Sarah and Liz are all to be giving oral presentations at. Peter, whilst chairing a session has this year been spared a speaking spot, but to make up for it is in Melbourne all this week.

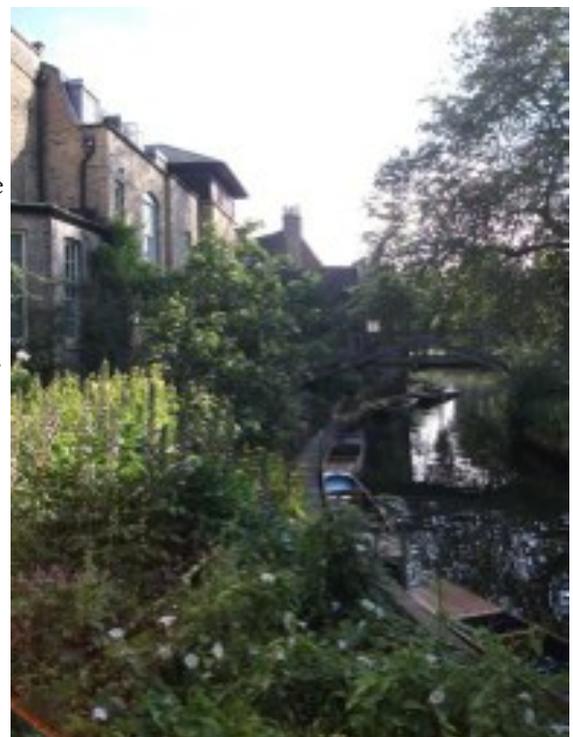
The bee season is once again back up and running, with frantic collections and dissections and so on occurring on a weekly basis. Everyone loves bees (except the people who love aphids, or flies....). (And except those who have carparks under hives - Ed.)



*Mmmmeat - Brazilian BBQ, or Churrasco*



*Jerónimos Monastery in Lisbon*



*Darwin College, Cambridge. Will be very familiar to a certain new member of staff.*

**Liz** has recovered from her semester 2 teaching and is now busy dreaming up new grant ideas and getting papers in shape for submission. The lab farewelled Moira with a pot luck dinner. Moira is now working with Biochem alumni Aleks Filipovska, Ollie Rackham and Stefan Davies in Perth.

**Lily Ong** has survived her first winter in New Zealand and is looking forward to **warm summer days**. Now, she is gearing up for more flow cytometry analysis in 2013.



**Aziz's** sanity, surprisingly, is still intact after finishing his fourth year. He is still recovering from the trauma of the countless immunoblots he has done over this period. He is currently lost about his future (nothing unusual) and is exploring various options that would allow him to retire by 35. He hopes Lionel Messi wins the Ballon D'Or award next year. 'Kristina' Ronaldo is going down!!

**Carolyn Porteous** is sad to be finishing working for Rob Smith who will be retiring at the end of 2012. She has enjoyed working with Rob and Mike Murphy and will get to catch up with Mike in December when he is here. She will then enjoy an extended holiday but be back in the new year.



Merry Christmas!

**Emily Sutton** has joined the lab for the summer after receiving a **Dean's Prestigious Summer Student award**. She is working on the cytochrome c project and is becoming proficient at setting up 96 well plate assays to measure the peroxidase activity of cytochrome c and handling the data that it generates.

*We wish everyone a safe and happy Christmas break*



**Tracy Josephs** has had an eventful year submitting her PhD thesis, sitting an oral exam, presenting a seminar and has now been given the all clear to **graduate** in December. She will be back in the Ledgerwood lab next year until the end of February at which stage she wants to leave the country to start a post doc.

**In other news...** Gill Hughes, Gemma Dickson, Carolyn and Tracy met in secret to develop their artistic skills and produce some **colourful, creative cupcakes**.



Introducing E<sup>3</sup>:

The Laboratory for Enzyme Engineering and Evolution

Strange things are afoot in Lab 114. The peace has been shattered by the arrival of Wayne Patrick, Monica Gerth and Matteo Ferla from Massey Albany, together with summer student Jordan Minnell.

Monica and Wayne arrived in September, in a small car packed with substrates, chemicals, tips, tubes, stirrers, rockers, vortex mixers, a thermal cycler...and a couple of clean shirts. Then began the Sisyphean task of cleaning and organising the new lab. Key discoveries included John and Sue's enormous Sephadex

collection, a packet of J.H. Munktell's Unrivalled Genuine Swedish Filtering Paper (Recommended By The Most Clever Chemists), and a bottle of SnCl<sub>2</sub>·2H<sub>2</sub>O that was received in January, 1961. There's a free drink at Happy Hour for anyone who can describe a biochemistry experiment that would require the latter!



Speaking of Happy Hour, Matteo arrived in time for the great Departmental Table Tennis Championship. With the Bradbeer Cup up for grabs, the E<sup>3</sup> Lab excelled itself. Wayne saw off Tyler McInnes in the final, Monica retired undefeated (after Julian failed to front for their second-round match), and Matteo was handily beaten by everyone, thus winning the coveted jar of pickled onions for Worst Player in the Tournament. Though the photographic evidence suggests that he may have thrown his last match, just to get the pickled onions.

Meanwhile, Monica's summer student, Jordan, has the honour of being the E<sup>3</sup> Lab's first ever Otago student. Jordan has brought his youthful enthusiasm and an uncanny knack for cloning chemoreceptor genes. Wayne and Monica suspect that Matteo is pleased he has another student to talk to, too.



So – the E<sup>3</sup> Lab is rolling. We even have a logo (above), best described by its designer, Matilda Newton, thus: “An enzyme riding a velociraptor is a universal symbol of strong science.” The lab survived its first MPI inspection (“No worries...it's only 10,148 GMOs in our freezer...”). Matteo survived his first committee meeting, and he has big plans to characterise and evolve multi-functional enzymes over the summer. Jordan will have pesticide-sniffing bacteria in no time. Monica and Wayne are so confident in their abilities, that they're off to walk the Rees-Dart next week!



## JFC Heading for Retirement

With my old lab now under new management (Wayne and Monica) and teaching over for the year I am able to turn my attention to more scholarly matters, mainly writing up those last few papers before I retire next year. Sue retired back in August and to mark the occasion we took off to Europe for some 'light adventures', namely sailing in the Saronic Gulf (Greece) and cycling in Umbria (Italy). As Sue is a qualified skipper we have chartered yachts in the Med previously and this time we got ourselves a nice new 37 footer for a week's cruising amongst some of the Greek islands. With a different



mooring every night we sampled fine scenery, pleasant breezes, archaeological sites and great food - thoroughly earned of course after "arduous" days filled with sailing, swimming and expeditions ashore. The cycling holiday was a little more energetic as most of the historic Umbrian towns and villages are located on hillsides. Perugia, Assisi, Spello, Trevi, Spoleto, Montefalco, Bevagna, Ripa are names that roll off the tongue like the smooth dark wine of the region. Umbria is less well known than Tuscany but it has a special charm with lovely countryside patchworked with olive groves, vineyards and small farms; great cycle touring territory and great cuisine (olive oil, truffles, prosciutto...).



Preparing for retirement by working two days a week is rather pleasant. It is also a very satisfying platform from which to view the successes of former students whom you once supervised. Wayne Patrick is one of these and now another, Peter Mace, who is also the recipient of a Rutherford Discovery Fellowship, will join the department next year. A third, Ben Berks who is a Professor of Biochemistry at Oxford University, has recently visited the university as a guest of the NZ Microbiological Society and gave several talks. And I mustn't forget Craig and Bronwyn whose influence lies deep within our department, and Yoshio who is master of all things protein. Ah yes, my fiendish plan to infiltrate the entire department is working very well!

John

JER Chronicles

2012 - What a year it's been, with such memorable occasions such as the London Olympics, the Queen's diamond jubilee, the 100<sup>th</sup> anniversary of the Titanic sinking, the US elections, the first TV series of The Ridges and the devastating end to the Twilight movie saga!! Whoa, quite a list, and it's all been happening in Lab JER too!

Firstly, massive congratulations to both Lux Selvanesan and Simon Jackson who have submitted their PhD theses and passed their oral examinations, graduating in December!! A brilliant effort. Hard on their heels is Peter Mabbit who hopes to submit his PhD thesis at the end of November. Jake Lamb has finished his Masters thesis and is awaiting his final mark, then will decide where to go from there. As well as writing his thesis, he also had time to write a paper, and maybe has another one in the pipeline! No mucking around. Max Waack has also completed his MSc in Plant Biophysics with Julian and the thesis is being bound as I write. As for the rest of the lab, Simon Cabout is typing frantically and wanting his Masters thesis to be completed yesterday. Ryan Hill is doing the same on his PhD thesis, although has the odd experiment he is continuing with.

Potentially there could be a mass exodus from the JER lab this summer!

Back at the bench, Asher Dale is doing what Asher does best – we aren't sure what that is exactly, but he's sticking around for a bit longer. Tim Crawford is continuing with his PhD project, and flits between Botany and Biochemistry leaving a trail of destruction behind him. He is about to attend the NZMS conference held here at the University Of Otago. We have welcomed two new students into the lab. Harvinder Singh has begun his PhD with us investigating mutations in Psb27 and Jack Hervey is working as a summer student hoping to put his-tags on two Photosystem II protein assembly factors for us.

Julian has had a busy year attending conferences, teaching, doing Acting HOD stuff and getting whacked on the head requiring stitches and freaking out Air NZ passengers. He tells us he is planning on spending the summer writing - with papers and a book chapter to contend with, as well as preparing grant applications. Busy, busy!

So, yes, the year is closing and the decorations are slowly going up around the city. Santa will be in town soon, and without this turning into a Tui ad too much, this calls for the lab to don their best attire and attempt to go out for a civil and sophisticated, well mannered night out ... Yeah, right!

This year Lab 308 are embarking on the Monarch for a one-hour cruise on our lovely harbour, taking in the views and scenery while enjoying some canapés and beverages on board. After this we wander around to Customhouse for the remainder of our meal and exchange our secret Santa gifts... OK, you know us, in reality we are running a sweepstake to see who is gently pushed/trips into the harbour, and secret Santa is actually quite a nerve-wracking experience!! No doubt news of the evening will filter through the department in due course.

We all wish the rest of the Department a very happy holiday and hope you have a well earned, relaxing break. Don't eat too much and remember to slip, slop, slap on that sunscreen ... if the sun actually decides to appear.

See you in 2013!

As you were ...

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Cheeseburger Pie Recipe (as mentioned in last newsletter)

2 sheets pastry	Line pie dish with pastry.
500g mince beef	
5 onions	Dice 1 onion. Cook mince, onion, gherkin, thyme and garlic until brown. Mix in mustard, chilli powder and 1/2 cup of cheese. Remove from pan and cool.
1 garlic clove, minced	
1 tsp thyme	Chop 4 onions into rings, cook with butter and sugar until soft.
1 Tbsp dijon mustard	
1/2 tsp chilli powder	Put mince into pie dish, cover with cheese, add onion rings, cover with more cheese.
2 cups cheese, grated	
3 Tbsp butter	Whisk eggs, milk and cayenne pepper together. Pour over pie. Coat with cheese, salt and pepper.
2 tsp sugar	
4 eggs	Bake at 230C for 15mins. Reduce heat to 180C and bake for a further 20mins.
1 cup milk	
cayenne pepper	<i>Thanks heaps to Tanya Flynn for satisfying my curiosity as to this culinary delight. It actually does sound yummy -Ed</i>
gherkins	

## Ready-made moustaches

The Cancer Lab kindly provided these cut-out moustaches for those who are unable or unwilling to go to the effort of growing their own for Movember. They also suggested not publishing them when they heard the newsletter wasn't coming out until December, but they are JUST TOO GOOD. Stash them away for next year people.



Fu Manchu



Dali



Groucho Marx



Tom Selleck



Boulangier



Handle bar



Ron Jeremy



Joseph Stalin