# Chemistry

IN NEW ZEALAND

ISSN 0110-5566

Volume 72, No.1, January 2008

## *Featuring*

A Cleaner and Greener New Zealand Thanks to 2,4,5-T, Science, and Silicones

Biomedicals from Bone

Do We Expect Too Much? Reflection on Chemistry Content in Higher Education

Fighting Food Fraud with Science

MALDI-TOF Mass Spectrometry of Cyanobacteria: a Global Approach to the Discovery of Novel Secondary Metabolites

The 2007 Nobel Prize in Chemistry

*2008 International Year of Planet Earth*

Published on behalf of the New Zealand Institute of Chemistry in January, April, July and October each year.

**The New Zealand Institute of Chemistry Incorporated**

PO Box 39-112
Harewood
CHRISTCHURCH
Phone: +64 3 359 7275
Fax: +64 3 359 7248
Email: NZIC.Office@nzic.org.nz

**Managing Editors and Publishers**
Fiona Summerfield and Rebecca Hurrell
PO Box 39-112
Harewood
Christchurch
Phone: +64 3 980 4809
Email: fiona@thinkdifferent.co.nz

**Scientific Editor**
Professor B. Halton, FRSNZ, Hon, FNZIC
School of Chemical and Physical Sciences
Victoria University
PO Box 600
Wellington
Phone: +64 4 463 5954
Fax: +64 4 463 5241
Email: brian.halton@vuw.ac.nz

**Advertising Sales**
Fiona Summerfield
128a Halswell Road
Hillmorton
Christchurch
Phone: +64 3 980 4809
Mobile: 021 075 4917
Email: fiona@thinkdifferent.co.nz

**Disclaimer**
The views and opinions expressed in Chemistry in New Zealand are those of the individual authors and are not necessarily those of the publisher, the Editorial Board or the New Zealand Institute of Chemistry. Whilst the publisher has taken every precaution to ensure the total accuracy of material contained in Chemistry in New Zealand, no responsibility for errors or omissions will be accepted.

NZIC Chemistry IN NEW ZEALAND

Volume 72, No.1, January 2008

## Articles and Features



## Other Columns

# New Zealand Institute of Chemistry

***supporting chemical sciences***

## January News

2008 is the International Year of Planet Earth as designated by the UN General Assembly

The aim of the year is to demonstrate new and exciting ways in which earth sciences can help future generations meet the challenges involved in ensuring a safer and more prosperous world. To this end *Chemistry in New Zealand* intends publishing one article in each issue this year that has relevant to this topic – for this issues see: *A Cleaner & Greener NZ Thanks to 2,4,5-T, Science, and Silicones* by J A Zabkiewicz.

### RSNZ Fewllowships

Prof ***Sally Brooker*** (Otago University and NZIC Branch Delegate) was the sole chemist among the thirteen elected to Fellowship of the Royal Society of New Zealand on Nov 7. Congratulations from Council and the Branches.

### The 2007 Nobel Prizes

Commencing on October 8, the 2007 prizes were announced in their usual sequence. A brief synopsis of the science awards is provided below.

***The physiology medicine prize*** (Oct. 8) was awarded jointly to ***Mario R. Capecchi*** (University of Utah), ***Sir Martin J. Evans*** (Cardiff University), and ***Oliver Smithies*** (University of North Carolina) for their discoveries of *principles for introducing specific gene modifications in mice by the use of embryonic stem cells*. The Laureates made a series of ground-breaking discoveries concerning embryonic stem cells and DNA recombination in mammals with discoveries that led to the creation of the immensely powerful technology known as *gene targeting in mice*. It is now applied to virtually all areas of biomedicine – from basic research to the development of new therapies.

Gene targeting is often used to inactivate single genes. Such gene knockout experiments have elucidated the roles of numerous genes in embryonic development, adult physiology, aging and disease. To date, more than ten thousand mouse genes, *ca.* 50% of the genes in the mammalian genome, have been knocked out. Ongoing international efforts should make *knockout mice* for all genes available in the near future.

Gene targeting allows the production of almost any type of DNA modification in the mouse genome, thus allowing scientists to establish the roles of individual genes in health and disease. Gene targeting has already produced more than 500 different mouse models of human disorders, including cardiovascular and neuro-degenerative diseases, diabetes and cancer.

***The physics prize*** (Oct. 9) went to ***Albert Fert*** (Université Paris-Sud, France) and ***Peter Grünberg*** (Forschungszentrum Jülich, Germany) for the discovery of *Giant Magnetoresistance*. It recognises their work that led to the technology used for data reading on hard disks. It is thanks to this them that hard disks have been miniaturized so radically in recent years, as illustrated by the sensitive read-out heads for compact hard disks in laptops and some music players.

In 1988 Frenchman Fert and German Grünberg each independently discovered a totally new physical effect – *Giant Magnetoresistance* (GMR). The GMR effect was discovered thanks to new techniques developed during the 1970s to produce very thin layers of different materials. To work, GMR needs structures consisting of layers that are only a few atoms thick and for this reason it is one of the first real applications of *nanotechnology*. The very weak magnetic changes that give rise to major differences in electrical resistance in a GMR system provided the perfect tool for reading data from hard disks when information registered magnetically has to be converted to electric current. Other researchers and engineers subsequently used the effect to provide the read-out heads. The first of these was launched in 1977 and the most recent read-out techniques of today are further developments of GMR.

***The prize in chemistry*** (Oct. 10) was awarded to ***Gerhard Ertl*** (Fritz-Haber-Institut der Max-Planck-Gesellschaft, Berlin) for his studies of *chemical processes on solid surfaces*. His groundbreaking studies are industrially important and help the understanding of why iron rusts, how fuel cells function, and how the catalysts in our cars work – more detail is provided elsewhere in this issue.

***The Nobel Peace Prize*** (Oct. 12) was shared, in two equal parts, between the ***Intergovernmental Panel on Climate Change (IPCC)*** and ***Albert Arnold (Al) Gore Jr*** for their efforts to build up and disseminate greater knowledge about man-made climate change, and to lay the foundations for the measures that are needed to counteract such change. This award has not been without criticism, notably by those who are of the view that $CO_2$ levels have had no impact but also those who feel that the recipients have done little for global peace.

### 2007 New Zealand Association of Science Awards

The Council of NZAS announced it suite of awards for 2007 during November last as:

#### *Marsden Medal*

The Marsden Medal for outstanding contribution to the cause or profession of science went to Prof Ailsa Goulding, at the University of Otago, Dunedin for her sustained leadership and personal contribution to research

on bone density, osteoporosis, and the role of obesity and nutrition in children's health.

### *Shorland Medal*

The Shorland Medal for research was awarded to Dr ***Robin Mitchell*** (HortResearch) for making an outstanding personal lifetime contribution in using the skills of a chemist to answer an important biological and commercial problem - how pathogenic bacteria cause harm to plants. Robin was recipient of the 1978 NZIC Easterfield medal.

### *Research Medal*

The Research Medal, awarded to a young scientist for outstanding fundamental or applied research in the physical, natural, or social sciences went to Wellington NZIC Branch Secretary A/Prof ***Kathryn McGrath*** (Victoria University) for her outstanding research over the last three years that spans the disciplines of physical chemistry and soft-matter physics.

### *Science Communicator Award*

The Science Communicator Award went to curator of invertebrate zoology at the Canterbury Museum, Dr Simon Pollard.

## Council News

### *Postcodes*

***Fiona Summerfield***, one of the *Chemistry in New Zealand* managers, has been contracted to update the database with the new postcodes. These are required from July 2008. It is expected that the costs will be recouped in cheaper bulk mail rates.

### Institute Awards and Elections

The annual NZIC prizes have been awarded as follows:

**Easterfield Medal** to Dr ***Jadranka Travas-Sejdic*** (Auckland University) for her work on conducting polymers and their applications; she is also a member of the University's Polymer Electronics Research Centre (PERC).

**Chemical Research** (formerly HortResearch): Prof ***Andrew Brodie*** and A/Prof ***Eric Ainscough*** (Manawatu) for their work in the field of coordination chemistry. They have been longstanding collaborators at Massey, working in a range of coordination and organometallic chemistry areas, most recently on novel ligands derived from phosphazene-derived scaffolds.

**Industrial & Applied Chemistry** (formerly Nufarm) Professor ***Jim Johnston*** (Wellington). Jim has made significant contributions across a wide range of industrial and applied chemistry areas, most recently using gold nanoparticles as novel colorants in fashion textiles.

**The Denis Hogan Chemical Education Award**: ***Adrian Jull*** (Manawatu) Adrian is a chemistry tutor at Massey and has undertaken significant outreach activities involving high school students.

Sponsorship for the prizes for academic research and industrial and applied chemistry has been lost. Council is following up possible new options but any suggestions for sponsorship should be directed to the NZIC office.

Council elected to Fellowship in September last Prof ***Peter J Derrick*** (Manawatu) and Dr ***Tony Woolhouse*** (Wellington). Peter recently moved from the Warwick (UK) to become Massey University's Head - Institute of Fundamental Sciences while Tony is an organic chemist with IRL, and has a longstanding association with the Wellington Branch.

### Branch grants

Branches that submit a financial statement and budget before the first Council meeting of the year will be paid one half of their previous year's grant as an interim measure. The full grants (as recommended by the Treasurer and approved by Council) will only be paid out when ***all*** Branch budgets are available.

### Joining fee

The joining fee arrangement since Jan. 1 now require new members to pay the full fee up to 31 July of each year. From 1 August to the end of the year a half fee is payable. All members will then be invoiced for the full subscription in the following year

### Branch subsidies to students

Council urges Branches not to subsidise travel or conference fees to students who have less than 6 months membership and are financial (paid-up members) at the time of any consideration for subsidy.

### Specialist group

Moves are taking place to set up an environmental green and industrial chemistry specialist group. If you interested in being included, advise the NZIC Office.

### *Clever Kiwis* stamps

NZIC now owns a first day cover with the 'clever kiwi' stamps which includes the 'Spreadable Butter' $1.00 stamp highlighted in the last issue.

### Chemistry in NZ

It is a pleasure to announce that Dr ***Peter Hodder***, formerly of the Waikato Branch but now a resident consultant in Wellington, has joined the editorial team as assistant editor. Peter worked as a scientific editor with DSIR prior to a university appointment. He has already cut his teeth on the Nobel article and the Branch News that follow in this issue.

Council has decided to archive the *Journal* from 1936 to provide an easily accessible resource for members and a quotation from Datacap is to be considered.

### NZIC Chemical Education Trust

The Chemical Education Trust distributed $4100 by way of twelve grants last October, $705 more than in 2006. Twenty applications were received but only 12 could be funded with preference given to applications from schools that had not received grants in the last 2 years. Most of the Trust's income is from interest earned on capital investments, although donations remain an important source. The Trustees are most grateful to those NZIC members who have generously supported chemistry teaching this way. Members are reminded that donations over $5 qualify for tax exemption.

It was disturbing to see many of the applications made for very basic equipment such as burettes, pipettes, *etc.*, that one would expect a school to supply. This clearly demonstrates just how poorly equipped many schools are and it reflects how science is regarded by many in our community.

Andrew Brodie, Convener

| Grant | Item | School |
|---|---|---|
| $400 | pH/mV/ temp meter | Wanganui Girls' College |
| $400 | burettes | Waiheke High School |
| $400 | volumetric flasks | Katikati College |
| $300 | burettes | Kaiapoi High School |
| $400 | Diji pipettes | Mangere College |
| $250 | burette clamps | Cashmere High School |
| $400 | hand held pH meters | St Mary's College |
| $400 | sensors for data logger | Tararua College |
| $250 | science diagrams software | Wanganui High School |
| $350 | safety glasses | St Joseph's Māori Girls College |
| $250 | molecular models | Bayfield High School |
| $300 | molecular models & glassware | Rangitikei College |

## BRANCH NEWS

### *AUCKLAND*

***Ken Seal*** – NZIC President in 1972 – died in Auckland on October 29th, 2007. He was 84 years of age. Born in England, he came to NZ in 1952 and had a distinguished career with Ceramco. An obituary will appear in the April issue.

The Branch AGM was held on October 18. The new chair is Dr ***Jadranka Travas-Sejdik***. She gained her PhD from the University of Auckland and then worked in industry [Pacific Lithium (NZ) Ltd.] before returning to the University. These have evolved to her appointment as Senior Lecturer in Chemistry and Director of the Polymer Electronics Research Centre. She is also a Principal Investigator with the MacDiarmid Institute for Advanced Materials and Nanotechnology. Jadranka's research focuses on the development of biosensors using conducting polymers and quantum dots, and on new applications for conducting polymers. As she is on Research and Study leave until early 2008, ***Brent Copp*** (retiring chair) will continue until her return. ***Gordon Miskelly*** continues as Secretary/ Treasurer, ***Gordon Rewcastle*** retains Council delegate, and ***David Salter*** continues as Chemical Education representative. The committee members are ***Marija Gizdavic-Nikolidis, Patricia Shaw, Robert Bennett,*** and ***Geoff Beresford***. ***Corrina Thompson*** (a past-president of the students' Chemistry Club) is the new student representative.

The AGM was followed by a very interesting presentation by Dr ***Keith Bedford*** (ESR Ltd.) entitled *Chemistry, Drugs, and Clandestine Laboratories*. This talk discussed the evolving nature of the NZ drug scene, and the current scientific and legal issues associated with control of illicit drugs in this country.

***Peter Schwerdtfeger*** (Massey, Auckland) has received $720,000 from the Marsden Fund for theoretical work on chemistry under extreme conditions.

#### *University of Auckland*

The Department congratulated Prof ***Margaret Brimble*** (*Using molecules from metal enriched mines for new medicines*) and Dr ***Andrew Dingley*** (*Characterizing the molecular and structural mechanism of antimicrobial pore-forming toxins*) on obtaining Marsden grants in the 2007 round. Chemistry students featured in the Faculty of Science student poster competition with ***Annalisa Durdle*** and ***Hannah Kelly*** (both Forensic Science) receiving 2nd and 3rd prizes, respectively, while ***Mandy Herbst*** (Wine Science), ***Lynley Crawford, Tanya Kjallman,*** and ***Sandra Baur*** also received commendations.

### *CANTERBURY*

Just when it seemed that the NZIC *Trivia and Truffles* event couldn't get any better, it did! A record 23 teams enjoyed an evening of hilarity and tense competition pondering over questions that sometimes had a less than tenuous link to chemistry. Contestants came from ESR, AgResearch, Crop and Food, Chemistry, Biochemistry, Management, and the College of Science. Previous stars at this event got *done-in* by the music questions. The results were: 1st, *It takes Alkynes* (***Alan Downward, Reuben Jane*** and ***Neroli Ayling***); 2nd, *The Peacocks* (***Paul Wilson, Sam Edwards*** and ***Daniel Milligan***), and 3rd equal, *The Inhibitorz* (***Scott Walker, Hemi Cumming*** and ***Leonardo Negron***) with *Nanotrivieomics* (***Andy Muscroft-Taylor, Sophie Walker*** and ***Genevieve Evans***).

The best team name went to *The Importance of Being Nernst* with *It takes Alkynes* a close runner up. An amusing answer, which won a spot prize in response to the question *Which Australian creature could inject a nasty toxin via spurs tucked away on its back legs* was *Most of them with the exception, perhaps, of some sheep.*

The October meeting was a visit to the Canterbury Innovation Incubator. A small but enthusiastic group enjoyed a very enlightening session during which they heard how the Incubator could help them get a product to the market place. Two businesses in the Incubator were visited.

Double congratulations to ***Rebecca Hurrell*** and ***Andy Muscroft-Taylor*** on their recent marriage and her successful application for a position in the University of Canterbury Liaison office. Rebecca, the Canterbury Branch Delegate and joint manager of *Chemistry in NZ*, also maintains the NZIC website and is an active member of the Chemical Education specialist group. Through her position in the Science Outreach office she has worked extremely hard in promoting chemistry in schools. She has been instrumental in setting up the *Chemteach* website. How much she will be able to continue these activities in her new role in Liaison remains to be seen.

#### *University of Canterbury*

A welcome barbeque for new staff members Dr ***Paul Kruger***, Dr ***Sally Gaw***, and Dr ***Vladimir Golovko*** was

hosted by the current members of the Inorganic staff. In late October two Visiting Erskine Fellows, ***Evamarie Hey-Hawkins***, and ***Justin Gooding*** and his wife ***Kat***, departed after contributing a great deal both academically and socially to the life of the Department. ***Jonathan Morris***, previously an academic in the Department and now at the University of Adelaide, recently visited. Jono was in town for ***Sarah Lundy's*** PhD examination and Becs and Andy's wedding.

Recently completing their PhD studies were ***Janna Nikkel*** (Andrew Abell) - *The design, synthesis and biological assay of cysteine protease specific inhibitor*, ***Sarah Hickford*** (Murray Munro/John Blunt) - *Studies in the Chemistry of Marine Natural Products*, and ***Sarah Lundy*** (Jonathan Morris) - *Synthetic Approaches to the Bicyclic Core of TEO3.1, Hamigerone and Embellistatin*. Sarah is now the Equipment Manager/Postdoctoral Fellow for the *Biomolecular Interaction Centre* at Canterbury. ***Tyler Chen***, recently completed his MSc degree (Jim Coxon and Andrew Abell supervisors), and is now a PhD student. He will be working with Jim and Emily Parker, with Andrew as a co-supervisor-at-a-distance.

***Moriah Sandy***, a second year doctoral student at the University of California, Santa Barbara, has been working with Profs ***John Blunt*** and ***Murray Munro***. She will spend three months here on a fellowship from the NSF East Asian Pacific Summer Institute program. ***Storm Uru***, a recent student, successfully defended his world lightweight single scull 1500 m crown at the recent under 23 World Championships.

Congratulations to ***Abby (Wanting) Jiao***, who was awarded a 2007/8 UC Summer Scholarship. Abby worked with Emily Parker and Jim Coxon on *Modelling the Interaction of Calpastatin with Calpain for the Development of Modulators for Traumatic Brain Injury*. ***Amy Zhang*** was awarded the HRC scholarship, which provides one Summer Studentship to a UC student to work in a health related field. Amy will be working with Emily Parker.

## *MANAWATU*

The Manawatu Branch congratulates the Branch Chairman ***Peter Derrick*** on his election to NZIC Fellowship.

A number of members of the Manawatu Branch were involved in judging entries in the 33rd Fonterra Manawatu S & T Fair in August. The Branch NZIC prize ($150) was awarded to ***Rudi Smith*** (Palmerston North Boys' High) for his entry *Where there's muck there's brass*. The Massey University Chemistry Prize ($100) was awarded to ***David Welman*** (Palmerston North Boys' High) for his entry *Taking out the caffeine – it's no tea party.*

David Welman, Massey University Chemistry Prize winner.

The Branch held a mini-symposium on *Organic Chemistry in the Manawatu*, once again sponsored by Barry Scott from Merck. ***Serena Smalley*** (IFS, Massey) did an excellent job as mini-symposium secretary. This event highlighted the diverse nature of the Branch, with presentations from ***Ghislaine Cousins*** (NZP), ***Samir Dar Gupta*** (LASRA), ***Justin Bendall*** (Fonterra), ***Lucy Lesperance*** (Crop and Food), ***David Harding, Gareth Rowlands, Ashton Partridge*** and ***Vyacheslav Filichev*** (IFS, Massey) and ***Jaspreet Singh*** (Riddet Institute, Massey).

***Grant Boston*** has joined the NZ Pork Industry Board as Research and Innovation Manager.

### *Massey University*

The 2007 RSNZ Hatherton Award for the best scientific paper by a PhD student at any NZ university in physical, earth, and mathematical and information sciences has been awarded to ***Celia Webby***, formerly of Massey and now of Oxford University. In 2005, working with Emily Parker, Celia published a paper in the *Journal of Molecular Biology* (**2005**, *354*, 927-39) on the structure and function of an enzyme from the *Mycobacterium tuberculosis*.

Recently appointed Head of IFS, ***Peter Derrick***, has been awarded the Thermo Fisher Scientific Award by the RSC for his research on both the fundamental and applied aspects of mass spectrometry, including applications to biochemistry and medicine. ***Andrew Brodie*** was presented with a NZ S & T Medal in Wellington in December. The award recognizes Andrew's significant contribution over an extended period to the promotion of science, in particular through his far-reaching activities at the secondary-tertiary interface. ***Richard Haverkamp*** has been promoted to Professor of Nanotechnology.

A Massey University Technicians Award has been given to ***Gareth Rowlands*** to study alternative forms of chirality in the development of novel organocatalysts. The award will finance a technician salary for 2 years. University postdoctoral fellowships have been awarded to ***Paul Plieger*** for *Metal Salt Extract*, and ***Shane Telfer*** and ***Mark Waterland*** for *Connecting the Quantum Dots*.

***Vyacheslav Filichev*** has obtained

Marsden fast start funding for his project, *Synthetic Probes Yielding Stable and Selective DNZ Triplexes for Gene Visualisation*. ***Mee-Kyung Ahn*** has successfully completed her PhD under the supervision of Emily Parker and is now employed in the Institute of Molecular BioSciences with ***Gill Norris***.

## OTAGO

It is with sadness we record the death of Otago HON FELLOW Emeritus Professor ***James Roper Robinson*** on 28 September 2007, at the age of 93 years. Prof Robinson was appointed Professor and Head of the Department of Physiology in Dunedin in 1961, and retired at the end of 1979 after an illustrious career in renal and cell physiology and with a reputation as a superb medical lecturer. He steered the Department of Physiology through its first major growth phase in the 1960s and early 1970s. His contribution to research was recognised when he was made a Fellow of the Royal Society of NZ in 1963. He will be remembered as an eloquent speaker and writer, who facilitated the careers of his staff and students and welcomed them into his home. His funeral was held on Monday 1 October in Dunedin.

Another fantastic evening of *Chemistry Quizzing* was held in late September. Some 210 high school students from Dunedin, Balclutha, Gore, Invercargill, and Tuatapere attended. After five rounds of questions, the winning teams (in order) were: ***Phantorium*** (Logan Park High), ***We are better than you*** (Kavanagh College), and ***The TM Team*** (Taieri College). The winners of the ever-popular Chemical Haiku competition were ***Hey hey ho ho we love bio*** (James Hargest High). Some old, but working pH meters were donated to some of the participating schools. All in all a great night, thoroughly enjoyed by the students and the large number of teachers who also attended. The quiz was again generously supported by UBS, Poppas Pizzas, Otago Museum, Otago NZIC, and the Chemistry Department.

Dr David McMorran presents the *Bunsen Burner of Wisdom* to the winners *Phantorium* (Logan Park High)

### *Chemistry Department*

***Henrik Kjaergaard*** has returned form his year-long sabbatical with Prof. Jorgensen's quantum theory group in Denmark. Henrik, awarded a research professorship from the Arhus University research foundation, is one of four external members of the newly established Lundbeck Foundation Center for Theoretical Chemistry headed by Jorgensen. The centre funded a 4-month stay for PhD student ***Anna Garden***. Apart from research, Anna completed two papers in theoretical chemistry and now reigns as local expert on coupled cluster theory. Henrik gave an invited talk *Atmospheric photolysis of sulfuric acid* at the 3rd Asian Pacific Conference on Theoretical and Computational Chemistry (Beijing). Anna Garden and ***Jo Lane*** attended the 7th Australian Conference on Vibrational Spectroscopy in Wollongong, presenting an oral paper *Estimating the OH-stretching linewidths in water dimers* and a poster *Fluorosulfonic acid and chlorosulfonic acid: possible candidates for overtone induced photodissociation*, respectively. ***Daryl Howard*** has spent last September in Boulder, Colorado recording vibrational overtone spectra of SH-containing compounds using the FT-IR and cavity ring-down spectrometer in Prof. Vaida's laboratory.

## WAIKATO

The President's address from ***Jan Wikaira*** in October was a fascinating account of the history of crystallography in New Zealand, with the audience enjoying spotting the infamous faces from their past and present.

### *University of Waikato*

***Richard Coll*** and ***Neil Taylor*** (University of New England, NSW) have now published their book *Science Education in Context: An International Examination of the Influence of Context on Curricula Development and Implementation*. The genesis of this book was Richard's and Neil's experiences of teaching science in developing countries. Through their international contacts they ended up with 30 chapters covering science education in 26 countries. Analysis of the stories points to major issues in science curriculum development and it seems many resources have been wasted over the years. On a more positive note, the book provides some useful suggestions to improve the process of curriculum development.

***Michèle Prinsep*** attended the 5th Euroconference on Marine Natural Products (Ischia, Italy) and gave a talk on her group's recent alkaloid research. She also visited researchers in Singapore on the way back. ***Bill Henderson*** is on study leave from January until September 2008, a welcome relief after a six year period as Chairperson of the Chemistry Department. He will remain based at Waikato with short visits to Singapore and the UK.

A number of students are working in the Department over the summer. ***Rachael Linklater*** and ***Nicky Cameron*** (Bill Henderson) on chemistry of metal sulfide systems, ***Greer Tanner-Dempsey*** and ***Aimi Finnegan-Ramanui*** (Merilyn Manley-Harris) on detection of honey in imported medicines and food products, and the nature of charcoal pigment used for Ta Moko, respectively. ***Megan Grainger*** is to set up HPLC input for the ICPMS, an extremely busy instrument in the mass spectrometry facility. This will permit ion speciation of, for example, arsenic in environmental samples. ***Simon Williams*** will be analysing wool grease samples for a commercial project and also acting as a teaching assistant for a course on the operation of the Biacore SPR instrument.

We have had a number of interesting and varied Departmental talks from visitors recently. Canterbury Erskine fellow ***Eva Hey-Hawkins*** spoke on *Bridging Organometallic and Coordination Chemistry*, while ***Neil Edmonds*** (Auckland University) gave a talk on *Development of Polymer Science Research at Tamaki Campus*; our own ***Alan Langdon*** told us about

*Some Chemical and Electrochemical Aspects of Water and Waste*. ***Brent Copp*** (Auckland University) gave a talk on *Amino Acid-Derived Marine Metabolites: Adventures in Biologically Active Alkaloids* and ***Shane Telfer*** (Massey University) told us about *Helical Motifs in Coordination and Supramolecular Chemistry*.

*ChemQuest 2007*, the annual quiz for Year 12 students run by the Chemistry Department was held on October 17. This was a fun-packed night of chemical questions in the following four categories: *Periodic Puzzlers, Sensing the Senses, The Wide World of Chemistry* and *Demon Demos*. A total of 52 teams from 18 schools entered this year, with students coming from all around the greater Waikato region to compete for the James and Wells trophy, medals, and cash prizes. After each of the four rounds of questions, it was *Teachers' Turn* with four teachers per round competing for a small prize for themselves. It was a most enjoyable night for contestants, presenters and spectators and the following prizes were awarded: 1st, St Paul Collegiate, ***Kool Kids Klub*** (Craig McConnochie, Marc Calderwood, Matthew Fisher); 2nd, The Church College of NZ, ***Team Levi***: (Levi Ensing, Ririwai Fox, Tia-may Hadfield ); 3rd, Tauranga Boys College, ***TBC Fuz*** (Bryden Church, Jonathon Mitchell, Travis Scott); 4th, Rotorua Boys High School, ***Sulphur Rocks*** (Kyle Harford, Nikhil Balakrishnan, Tom Kelly); 5th, Fairfield College, ***The Eagles*** (Erica Burns, Erica Prentice, Nikki Graziotti). For the Teachers' Turn, winners were: ***Lisa Janek*** (Fairfield College) and ***Leon Rutter-smith*** (Waihi College).

The quiz was presented by ***Richard Coll*** and ***Michèle Prinsep***, with ***Bill Henderson*** running the *Demon Demos* and ***Lyndsay Main*** acting as chief judge. Numerous other people contributed to the success of the occasion including staff and students of the Chemistry Department and School of Science and Engineering. ***David Mackaskill*** from James and Wells and ***Gina Wallace*** and ***Leisle Noble*** from Hill Laboratories (the former two being Waikato graduates) presented the prizes. The sponsors of the quiz, whose contributions are gratefully acknowledged, were Chemistry Department and School of Science and Engineering, Waikato University, Hill Laboratories, and James and Wells (Intellectual Property Lawyers).

### *NIWA*

***Michael Ahrens'*** most challenging and rewarding new thrill in the last few months has been without doubt the birth of his first son Gabriel, and the joys (and chores) of fatherhood. Switching back to science, his latest research focus has been environmental risk assessment for new and emerging chemicals of concern, including surfactants, plastic additives (flame retardants, plasticizers), various biocides and pharmaceutical and personal care products. He is currently collaborating with several colleagues at NIWA and overseas on detecting these compounds in environmental samples. Of particular interest are endocrine disrupting chemicals, which are bioactive at very low concentrations.

***Bob Wilcock*** is working on land-water interactions in agricultural landscapes, $N_2O$ and $CH_4$ emissions from waterways and the importance of ion-pair formation in lake chemistry models. ***Craig Depree*** has been busy with FRST proposal writing – re-bidding the existing antifouling research programme. A new research programme began in October that aims to develop novel marine antifouling materials incorporating photocatalytic $TiO_2$ nanoparticles. The FRST project involves researchers from IRL Ltd. and James Cook University, and includes industry partners Altex Coatings Ltd. and Resene Paints. Craig has also been digging up streets in conjunction with the Christchurch City Council and Fulton Hogan for a Land Transport NZ project looking at recycling coal-tar contaminated streets. ***Kay Vopel*** is studying the effects of three phosphorus inactivating agents on the biogeochemistry of Rotorua lake sediment, using laboratory microsensor techniques that enable sub-millimetre resolution. The agents are applied to reduce the release of phosphorus from the sediment into the water column that can drive phytoplankton blooms. The research is providing information on the effects of these agents on transport and reaction of other important redox active solutes, such as oxygen and hydrogen sulfide. ***Greg Olsen*** is currently reviewing final equipment options for the purchase of a Membrane Inlet Mass Spectrometer (MIMS) simultaneously measuring the concentrations of major and trace dissolved gases, *e.g.* $O_2$, Ar, $N_2$, $CH_4$, $CO_2$, $H_2S$, over short timescales (<1 min). ***Mike Stewart*** recently attended a Conference in Faro, Portugal on *Pheromones and Welfare Indicators in Fish* and presented some of the research currently being undertaken at NIWA. While in Europe he also visited Dr Mark Viant (University of Birmingham), an expert in Environmental Metabolomics.

## *WELLINGTON*

The Branch congratulates Prof ***Jim Johnston*** on receiving the ***Industrial and Applied award*** from the Institute and A/Prof ***Kate McGrath*** on gaining the *NZAS Research Medal.*

The September meeting saw some 40 members attend the President's visit to the Branch. Her lecture *New Zealand and New Zealanders' role in advancing single crystal structure analysis* was preceded by the presentation of Fellowship certificates to the eight recently elected members of the Branch. Late September saw the second student function for the year – this time a showing of the movie *An Inconvenient Truth*. With free ice-cream prior to the film, a good number attended.

The October meeting was a presentation by ***Paul Kilmartin*** (Auckland University) on *unlocking the aroma chemistry of NZ Sauvignon Blanc wine*. The large audience was treated to some fascinating results from the chemical, sensory, and viticultural studies on the fruity and green aromas of one of the country's popular export wines.

The November AGM returned incumbent Chairman (***Richard Tilley***), Secretary (***Kate McGrath***), Treasurer (***Suzanne Boniface***) and Delegate (***Brian Halton***) unopposed. Fellowship certificates were presented to ***Sarah Russell*** and ***Tony Woolhouse*** and the latter then addressed the

*Continued on page 28...*

Feature

# A Cleaner and Greener New Zealand Thanks to 2,4,5-T, Science and Silicones

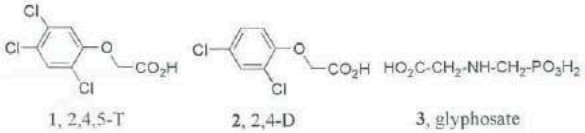
J A Zabkiewicz

Plant Protection Chemistry NZ, PO Box 6282, Rotorua (e-mail: *JAZ@ppcnz.co.nz*)

## Historical background

Those of you old enough to remember NZ being deluged with 2,4,5-T, **1** (one of a series of plant growth hormone herbicides that includes 2,4-D, **2**) thought it was the environmentalists who put a stop to this herbicide. Well, not really! It was a combination of a new herbicide and a new spray surfactant (adjuvant) that finally turned the tables on a product that had little market competition until then. And NZ science, with a touch of serendipity, played a major role in turning a *she'll be right* approach into a successful technology which has been adapted in various ways for a wide range of agrichemical products and crop situations.

Hill country covered in woody weeds was a huge physical and economic problem to the booming forestry sector in the 1970s and 1980s, as forests were planted on land which had degenerated from pasture to scrub weeds. Non-chemical methods of weed control - no, it's not a new idea - were practiced then, but involved burn-offs that frequently got out of control and cast a pall of smoke for weeks on end in places. Alternatively, huge machinery rumbled up and down the hillsides or rolled huge drums down them to crush the scrub weeds...so that they could be burned more safely. Farmers were no better off; they could stock their paddocks with goats which ate everything in sight, but then they had to have secure fencing as it took four to five years to make sure all the woody scrub weeds were controlled - but this was impossible with a plant like bracken fern. So herbicides were the first choice, and with aerial application any terrain could be treated. The herbicide 2,4,5-T was used extensively on gorse, broom, and native scrub weeds. But there was a problem (leaving aside the Agent Orange aspects and spray drift issues) for, despite a variety of product formulations and apparently good kills of gorse, the plants re-grew within a few months. This problem was recognised and a research programme involving the author was initiated in 1974 to specifically look at better methods of woody weed control at the former FRI in Rotorua. Over the next few years studies showed that herbicides that acted through the roots were impractical for mature plants, though excellent at controlling regenerating seedlings. Attention focussed on foliar uptake options for the mature plants and radiolabeled herbicides demonstrated large differences in uptake into plant foliage, as well as poor translocation by some, including **1**. In effect it was a good contact herbicide but far from ideal for woody weed control. Something new was needed.




1, 2,4,5-T 2, 2,4-D 3, glyphosate

That new herbicide was glyphosate (**3**, sold by Monsanto as Roundup® in the early 1980s) but although it was quite effective against annual broadleaf and grass weeds, when applied to the vigorous woody and rhizomatous weeds like gorse, bracken and broom, or perennials like clover, it failed. It was also very expensive, making it economically and biologically unattractive in the NZ situation. However, it had one huge advantage, it could translocate very well within plants and kill not just the foliage, but right down to the roots.

## Plant Biology and Physicochemical Interactions

It was well known that by adding different oils or surfactants (adjuvants) to the pesticide product you could improve spray efficacy. The reason for this is that the cuticle in a leaf presents a highly lipophilic surface to the external environment. The cuticular layer covering plant leaves is complex, but in general consists of a superficial wax layer (epicuticular wax), then a bilayer cuticular membrane (cuticle layer), the outermost layer (cuticle proper) composed of soluble (cuticular waxes) and polymerised lipids (cutin), and the innermost of polysaccharides which may contain high proportions of pectin (Fig. 1). The conundrum is that a lipophilic compound will readily absorb into the lipophilic cuticle, but will not readily translocate in the polar, ionic phloem or sap.[1] In the case of **1**, with an octanol/water (ow) partition ratio of log $K_{ow}$ >3, it was readily absorbed but poorly translocated.[2] In contrast, **3** has log $K_{ow}$ = – 4.5; it can be readily translocated but poorly absorbed. In the case of gorse not only has the spine (leaf) a very thick cuticle covered in wax,[3] but also it is covered in hairs (trichomes) that prevent droplets getting to the actual leaf surface (Fig. 2). So it was no surprise to discover that only 7% of glyphosate was actually being absorbed from commercial formulations[4] and field efficacy was also low.

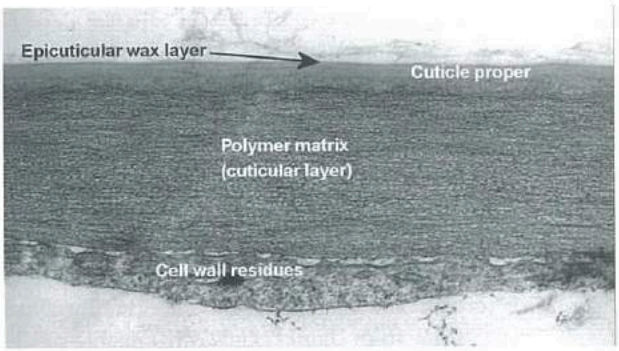



*Fig. 1.* Cross sectional representation of plant cuticle structure.

Uptake into leaves can occur in two ways, slow diffusion through the cuticle or physical flow through stomata (Fig. 3) that exist to absorb $CO_2$ for photosynthesis and expel

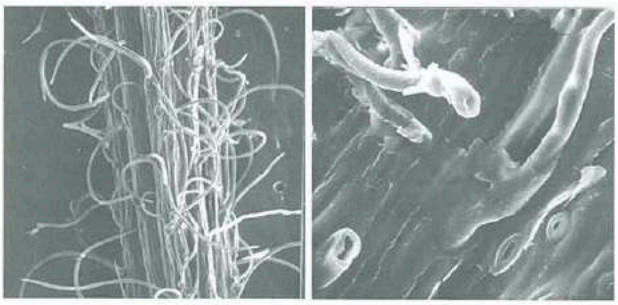


*Fig. 2.* SEM images of young gorse spine (left, x35) and close up of needle surface (right, x262) showing trichomes (hairs) and dense wax layer on needle surface.

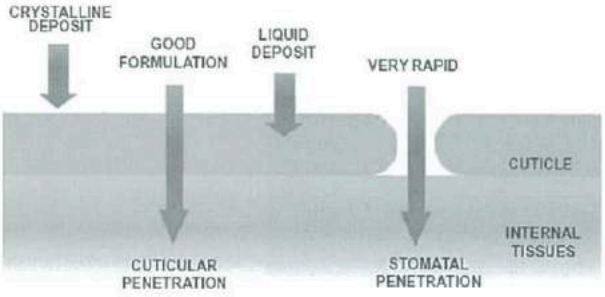



*Fig. 3.* Representation of xenobiotic uptake through leaf cuticle.

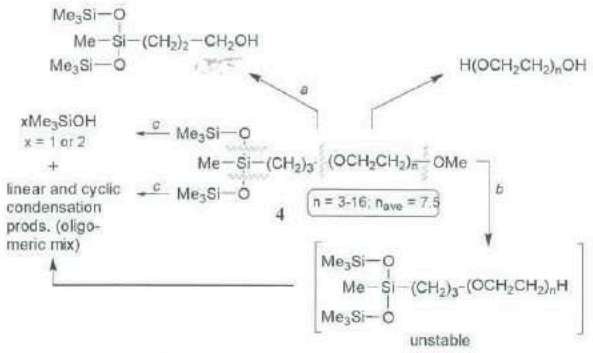
oxygen and water during respiration. Studies had shown[5] that the geometry and size of stomata determined whether liquids having a low enough surface tension could flow into the sub-stomatal chambers and hence into the leaf mesophyll. Water solutions with a surface tension of 76 mN/m were not able to flow into stomata for obvious biological reasons, and neither could Roundup solutions of **3**, so this pathway was unavailable. The advent of a material discovered fortuitously by the author (from an assortment of surfactants discarded by another researcher in Australia) provided a completely new and novel surfactant structure that had the ability to bring surface tensions down to levels never before achieved with agrichemical spray solutions.

The material was an organosilicone, an oligomeric mixture of trisiloxy polyethoxylate monomethyl ethers depicted by **4** (with $n_{ave} = 7.5$) (Scheme 1). Very dilute solutions of this trisiloxane surfactant, later marketed as Silwet L-77® (L-77) or Pulse®, were capable of wetting even Teflon surfaces, and could spread on a leaf surface 50 to 100 times more than other surfactant solutions. Solutions of **4** had surface tensions of 22 mN/m, a value well below the theoretical threshold required to infiltrate leaf stomata;[5] EtOH and $Me_2CO$ have surface tensions of 22.3 and 23.5 mN/m, respectively. Proof that this happened was provided by measuring the percentage uptake of $^{14}$C-glyphosate, with different concentrations of **4** over a period of time. Whereas normal cuticular diffusion is a slow process that requires several hours for substantial amounts of xenobiotic to be taken up into the leaf,[6] solutions with more than 0.3% **4** showed 80%+ uptake within a few minutes.[7] When the combination of Roundup® and L-77 formulations of **4** was tested on mature gorse plants, complete kill was obtained with one third the rate of the parent product formulation, which itself at full rate could only achieve two thirds mortality.[8]

The *real life* benefits of such technology are that glyphosate is used at about one quarter to one third of the rate per ha that would be needed without smart formulations. A recent survey[9] found that *ca.* 350 tonnes of glyphosate are being used annually. Forestry alone accounts for 144 tonnes of this and had the rates not been reduced, this would have exceeded 400 tonnes; national use would have been around 1000 tonnes p.a. Hence in the 20 years since smart formulations were introduced there has been a ***reduced input*** of around 5,000 tonnes in forestry and over 13,000 tonnes nationally. Prices of glyphosate products have dropped over that period but it is clear that national savings to NZ users are more than a billion dollars.

As usual, the explanation for these beneficial properties came after their discovery. The reason for such a low surface tension of a water solution of the organosilicone lies in the structure, size, and orientation of the surfactant molecules in the solution. Surfactants have a hydrophilic and lipophilic end in each molecule; the hydrophilic end associates with the water and the lipophile forms a tightly packed arrangement on the surface - essentially a monolayer surface film. So, in a water droplet, the lipophilic silicone moiety covers the surface thus having the initial interaction with the waxy leaf surface. In the process, some of the surfactant is *stripped* from the solution and lays a layer down on the wax, presenting the hydrophilic part for water to associate with and spread over. This process is fast, much faster than with conventional organic surfactants and the principles involved have been the subject of many theoretical studies.[10] It accounts for the fact that surfactant concentrations need to be well above the critical micelle concentration, so that there is excess surfactant to replace the material adsorbed into the cuticle. Evidence that treated areas retain some residual surfactant on the surface comes from the fact that if a droplet of water is placed in that region, it will spread and give a lower contact angle, in contrast to its behaviour on an untreated leaf surface. This again has biological implications as chemotactic reactions by insects and pathogens can be disrupted by *surface treatment* of foliage - but that's another story.

Another property of organosilicones such as **4** is that they are somewhat pH unstable. It is a drawback for long term *in-can* formulations and accounts for their use only as tank-mix adjuvants. It has a major environmental plus. Although stable at neutral pH for many weeks either in solution or in the presence of solid substrates, **4** can degrade within minutes in low or high pH environments. Extensive studies at Waikato University[11] have identified the degradation pathways. The potential sites for cleavage of the trisiloxane surfactant are illustrated in Scheme 1 and each generates more polar products. The Si-O bond is that most easily cleaved within the polymer and leads to silanols that are then subject to condensation and analogous depolymerisation to give a variety of linear and cyclic silanols and siloxanes. The cyclic derivatives are water soluble and known to be harmless to a range of natural organisms.




***Scheme 1.*** Potential cleavage sites (*a-c*) in the abiotic degradation of **4**.

One of the major biological advantages of these organosilicone surfactants is that they do not show any phytotoxicity to the plant and they can reduce contact phytotoxicity with non-herbicidal products. Thus they are used also with insecticides, fungicides, and plant growth regulators on field or fruit crops. Their enhanced spreading properties mean that instead of pesticide residues being deposited in discrete spots, they are spread more evenly over a surface. Such even coverage gives much better protection against pests or diseases, but at the same time it can also result in faster degradation of pesticide residues because a much larger surface area and a thinner film of pesticide molecules is involved. This is a very important consideration for food crops.

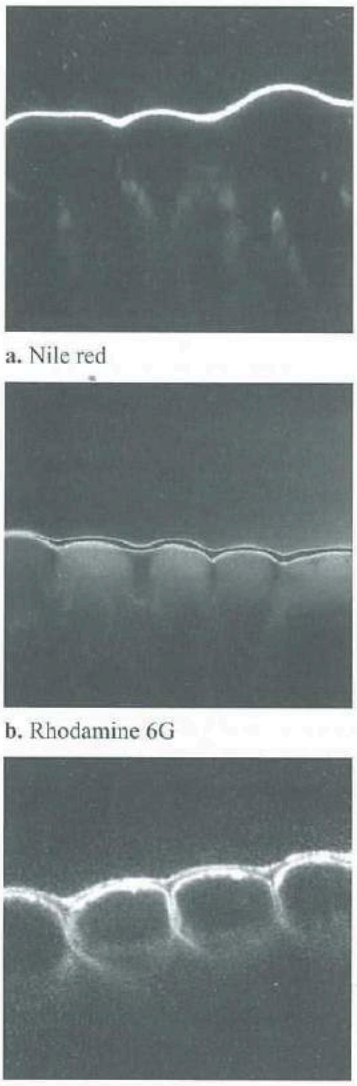
In subsequent years further work has led to the development of organosilicone blends that combine good spreading but no stomatal infiltration. Taking advantage of their ability to wet and stick to *hard to wet* surfaces, these new adjuvants are being applied to many horticultural crops where there may be large differences among leaf and fruit surfaces.[12] A further outcome is that with the correct formulation and application, spray volumes can be reduced. This allows for faster crop treatment, better use of expensive machinery and, most of all the ability to spray under the right conditions and avoid off-site drift. Such *concentrate* sprays are retained better, penetrate crop canopies more, and provide better target coverage than do standard spray applications. One such adjuvant product developed by a NZ company has been commercialized globally, so not only are there savings due to the technology but tangible revenues as well.

## Plant Cuticles and Cuticular Uptake

So what is the biology and chemistry behind these applications of formulation technology?

The predominant foliar uptake mechanism is by passive diffusion through the cuticle. The relative thickness of the cuticle layer vs the epidermal cell wall varies enormously among species. The cuticle has been shown to have a weak acid ion exchange capability and a high affinity for calcium ions,[13] as well as containing phenolic constituents[14] and reactive epoxy groups.[15] This variability illustrates the inhomogeneous nature of the cuticle and largely accounts, to date, for the failure to describe comprehensively the uptake mechanism of xenobiotics through the cuticle.

The process of xenobiotic uptake through leaf cuticles is an area of active study by Plant Protection Chemistry NZ ($PPC_{NZ}$) and other groups. An illustration is given by Fig. 4 using confocal laser scanning microscopy (CLSM) and fluorescent probes of different lipophilicity. It has been shown that the uptake and movement (in the presence of a surfactant) through the cuticle proper and into the epidermal cells varies greatly. The most lipophilic compound (Fig. 4a) is held completely in the cuticle; one that is less lipophilic diffuses evenly into the epidermal cells (Fig. 4b), while the most hydrophilic compound migrates through the epidermal cell walls (Fig. 4c).



**a.** Nile red

**b.** Rhodamine 6G

**c.** Fluorescein

***Fig. 4.*** Visualisation of movement through a leaf cuticle by fluorescent dyes of different lipophilicity from confocal laser scanning microscopy; all treatments 0.05% dye concentration in presence of 0.2% surfactant after 24 h into bean leaf.

The diffusion of substances through the cuticle is described by Fick's first law, where the flux is the amount of solute that diffuses through a unit area per unit of time, viz.:

(mass/area) × time.

It is proportional to the concentration gradient and the diffusion coefficient of the xenobiotic.[16] Researchers in Germany[17] have defined the principal factors affecting uptake rates as *solute mobility* (which is affected by temperature,

solute molar volumes, and cuticular wax composition), *tortuosity*, and *driving force*. Tortuosity is the length of the diffusion path through the *limiting skin* in the cuticle, where the limiting skin represents only a proportion of the cuticle thickness, not its entirety, and it is influenced by the size and orientation of the cuticular wax crystals. The driving force is affected by the starting and continuing concentration of active ingredient in the *solution* on the cuticle surface, in the cuticular layers, and in the epidermal cell wall. Overall, and in simple terms:

uptake = solute mobility × cuticle tortuosity × driving force.

The German studies were performed with isolated cuticles *in vitro*, using an artificial infinite concentration system; spread area was ignored, and only plants that had astomatous upper leaf surfaces were used, mainly from non-commercial species. Studies by $PPC_{NZ}$ with intact plants of many types (using leaf surfaces with or without stomata) showed that the influence of droplet spread area was highly significant *in vivo*.[18] Although two different formulations may contain the same concentrations of chemicals, *if the adjuvants are different or at different concentrations, then the residual droplet spread area will be different*. After droplet dry-down, the spray residue will be spread over different areas and the mass per unit area will vary.[19] These latter studies have shown that this solution residue or *initial dose* (ID) can be related to the mass uptake of xenobiotics.[18] This relationship has been validated with a wide range of formulations and plants that represent typical field rates and formulations.[20] An illustration of such a relationship is given in Fig. 5 for 2,4-D acid **2** in the presence of two quite different adjuvants into three plant species; very similar trends are seen for each species. Uptake per unit area at 24 h can be represented by the relationship:

$$\text{Mass Uptake} = a[\text{ID}]^b$$

where *a* and *b* are constants specific to the active on these species.

Total mass uptake can be determined from:

$$\text{Mass Uptake}_{(\text{nmole})} = a[\text{ID}]^b \times A.$$

where A is the droplet spread area. The mass uptake relationship has also been used to establish the relative importance of species, active ingredient and its concentration, and surfactant, to uptake.[21] It was found that the concentration of active ingredient increases in importance with increasing lipophilicity, but that surfactant concentration is less important as the active ingredient lipophilicity increases. The relationship between the active ingredient concentration and the species is more important for the most polar compound, while the interaction of surfactant and species increases in importance as the lipophilicity of the active ingredient increases.

These modelling approaches are markedly simpler than the original German methods and they can be applied to any model or operational system and to all plant species. Furthermore, by using a quantitative molecular basis,

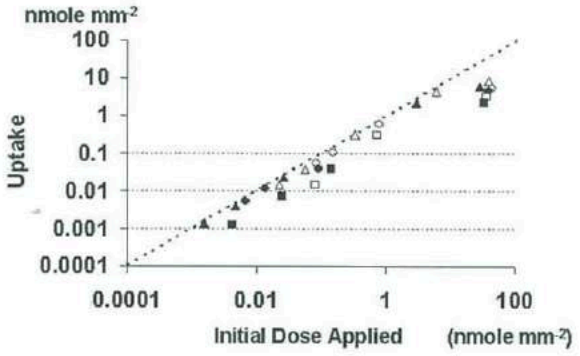



***Fig. 5.*** Mass uptake of 2 (2,4-D) in the presence of polytriethylene glycol monododecyl ether (Δ, ○, □) and trisiloxane **4** ($n_{ave}$ = 7.5) (▲, ●, ■) into *Chenopodium album, Hedera helix*, and *Stephanotis floribunda*, respectively; (---) is maximum uptake representing 100% uptake over the initial dose range.

dosages can be used to interpret interactions with endogenous plant constituents or structures from a biomolecular viewpoint. However, a much better understanding of plant leaf cuticular structures, as well as structure-activity relationships with adjuvants, is still required for a successful quantitative model of the uptake of the active ingredient. Specifically, there is a need to provide a numerical indicator (tortuosity factor) for plant cuticles that can then be incorporated into models of uptake so as to reflect species or leaf developmental differences. There have been electron microscopic studies of plant cuticles leading to a classification but the studies are qualitative only. It is known that cuticle thickness is an inappropriate input as, at times, thin cuticles can prevent uptake more than thick ones; thus cuticular structure is of prime importance.

As stated above, plant cuticles are complex, with layers of epicuticular waxes, embedded cuticular waxes, and a polymeric cutin (or cutan) skeleton. The waxes are solid at ambient temperatures but there are more or less *plastic* regions in both the waxes and the cutin which have been termed *amorphous* or *crystalline*. Solid state NMR now provides a measurement of cuticular wax or cuticle matrix that is *amorphous* or *crystalline*. Moreover, the proportion of *crystalline* to *amorphous* wax could provide a means of quantifying the *tortuosity* factor used in diffusion mechanism equations; this is the current approach used in a joint effort between Scion and $PPC_{NZ}$ staff. Cuticles isolated from a range of plant species and analysed by $^{13}C$ solid state NMR techniques[22] show differences in their cross polarization, magic angle spinning (CPMAS) spectra. The appearance and measurement of the $^{13}C$ NMR signals account for carbon atoms at different sites of the alkyl chains that form the cuticle structures or their cuticular waxes. For example, fruit cuticles have very similar spectra and *crystalline/amorphous* (*c/a*) ratios. However, the holly cuticle shows a very different spectrum but similar *c/a* ratios to the fruit cuticles (Fig. 6), while ivy is different to all of these. It appears that species can be placed in groups based on their *c/a* ratios and spectral character, and this has considerable taxonomic as well as structural significance. Cuticles were previously characterised visu-

ally by means of their transmission electron microscopy images and grouped into six categories.[23] The species analysed by CPMAS NMR can be grouped by their *c/a* ratios and these groupings coincide very well with the microscopic categorisation.

Removal of cuticular waxes by solvent extraction caused some alterations to spectra and *c/a* ratios. Measurement of the *c/a* ratios showed that in each case there was a small but definite increase in amorphous character. Cuticles were also analysed after soaking 24 h in surfactant solutions. As the concentration of surfactant increased, so the amorphous proportion increased. These studies, while preliminary, appear very promising for the characterisation of isolated cuticles and the interaction with solutions and surfactants of the polymeric structure and the cuticular waxes.

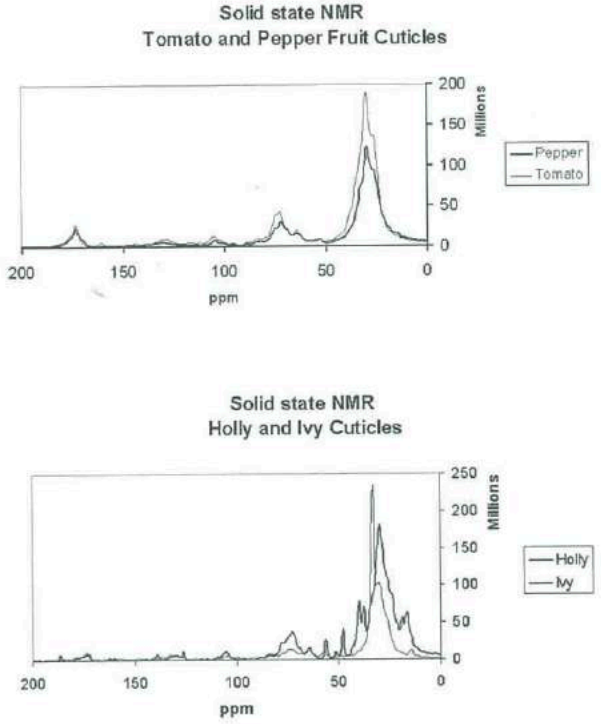



*Fig. 6.* $^{13}C$ CPMAS solid state NMR of isolated cuticles from tomato and sweet pepper fruit and holly (*Ilex paraguanensis*) and ivy (*Hedera helix*) leaves showing similarities and differences among species and sample types.

## Conclusions

Arising from a real-life problem growing on the NZ hillsides, whose resolution has been an operational, environmental, and economic success, studies associated with this very practical problem have crossed traditional boundaries between biology, chemistry, plant science, and pesticide efficacy. They involve solution rheology, solution dynamics, liquid-solid phase interactions, plant biology, and plant morphology. Consideration must also be given to the liquid and solid phase structures, both for the spray solution and for the composite biological membranes that they interact with at the cellular or nano scale. A genuine journey from macro to micro technologies, involving physical, analytical and organic chemistries and using fundamental principles which are being incorporated into practical models.

**Acknowledgements**

I would like to acknowledge the many staff of the former Forest Research Institute, now Scion, as well as Plant Protection Chemistry NZ (which arose from within the FRI), who have been or are associated with various parts of the work spanning three decades. The work has been funded by the FRS&T and several overseas and NZ companies.

## References

1. McCloskey, W. B. *Effect of surfactants and adjuvants on post-emergence herbicide efficacy* – see *http://ag.arizona.edu/crop/presentations/2003/mccloskey092403.pdf*
2. Zabkiewicz, J. A.; Gaskin, R. E. *Proc. 32nd NZ Weed Pest Control Conf.* **1979**, *32*, 315-318.
3. Zabkiewicz, J. A.; Gaskin, R. E. *New Phytologist* **1978**, *81*, 367-373.
4. Zabkiewicz, J. A.; Gaskin, R. E.; Balneaves, J. M. In *Brit. Crop Prot. Council Monogr.* No. 28, 1985, 127-134.
5. Schönherr, J. Bukovac, M. J. *Plant Physiol.* **1972**, *49*, 813-819.
6. Stevens, P. J. G.; Zabkiewicz, J. A. *Proc. Eur. Weed Res. Soc. Symp.* **1988**, 145-150; Stevens, P. J. G.; Gaskin, R. E.; Hong, S. O.; Zabkiewicz, J. A. *Adjuvants for Agrichemicals* CRC Press: Boca Raton, 1992, 385-398.
7. Stevens, P. J. G.; Gaskin, R. E.; Hong, S.O.; Zabkiewicz, J. A. *Pestic Sci.* **1991**, *33*, 371-382.
8. Balneaves, J. M.; Gaskin, R. E.; Zabkiewicz, J. A. *Ann. Appl. Biol.* **1993**, *122*, 531-536.
9. Manktelow, D.; Stevens, P.; Walker, J.; Gurnsey, S. *et al.* Project SMF 4193 Report, Ministry for the Environment 2005.
10. Ananthapadmanabhan, K. P.; Goddard, E. D.; Chandar, P. *Colloids and Surfaces,* **1990.** *44*, 281-297.
11. Bonnington, L. S. Ph.D Thesis, Waikato University, 2001; Bonnington, L. S.; Zabkiewicz, J. A.; Henderson, W. *Separation and detection* In *Comprehensive Analytical Chemistry*, Knepper, T.P.; Barceló, D. and de Voogt P. (Eds.), Elsevier Vol. 40, 2003, 627-653.
12. Gaskin, R. E.; Elliott, G. S.; Munro, J. P. *Proc. Brit. Crop Prot. Council (Pests and Diseases) Conf.* Brighton, 2000, 687-692; Gaskin, R. E.; Manktelow, D. W.; Elliott, G. S. *NZ Plant Prot.*. **2002**, *55*, 154-158; Gaskin, R. E.; Manktelow, D. W.; Skinner, S. J. *NZ Plant Prot.*. **2004**, *57*, 266-270.
13. Schönherr, J.; Huber, R. *Plant Physiol.* **1977**, *59*, 145-150; Hall, G. J.; Hart, C. A.; Jones, C. A. *Pest Manag. Sci.* **2000**, *56*, 351-358.
14. Liakopoulos, G.; Stavrianakou, S.; Karabourniotis, G. *Ann. Bot.* **2001**, *87*, 641-648.
15. Riederer, M.; Schönherr, J. *Arch. Environ. Contam. Toxicol.* **1986**, *15*, 97-105.
16. Price, C. E. *The Plant Cuticle*, Academic Press: London, 1982, 237-252.
17. Schönherr, J.; Baur, P.; Buchholz, A. *9th IUPAC Congress*, RSC: Cambridge 1999, 100-119.
18. Forster, W. A.; Zabkiewicz, J. A.; Riederer, M. *Pest. Manage. Sci.* **2004**, *60*, 1105–1113.
19. Zabkiewicz, J. A. *Crop Prot.* **2007**, *26*, 312-319.
20. Forster, W. A.; Zabkiewicz, J. A.; Riederer, M. *Pest. Manage. Sci.* **2006**, *62*, 664–672.
21. Forster, W. A.; Zabkiewicz, J. A.; Kimberley, M. O.; Riederer, M. *Trans. Am. Soc. Agric. Biol. Eng.* **2006**, *49*, 25-31.
22. Stark, R. E.; Tian, S. *Biology of the Plant Cuticle*, Blackwell: Oxford, 2006, 126-144.
23. Holloway, P. J. *The Plant Cuticle*, Academic Press: London 1982, 1-44.

# Biomedicals from Bone

Michael R. Mucalo[a] and Andrew J. Worth[b]

[a]Chemistry Department, Waikato University, Private Bag 3105, Hamilton 3240 and [b]Institute of Veterinary, Animal and Biomedical Sciences, Massey University, Private Bag 11222, Palmerston North (email: *m.mucalo@waikato.ac.nz*)

## Introduction

The realm of biomaterials, under which biomedical materials can be categorised, has a broad definition base and recognises materials that are synthesized or naturally sourced. Biomaterials are normally those that come into contact with live tissue and physiological fluids. They have applications as prostheses to replace lost function of joints or to replace bone tissue, for diagnosing medical conditions, as a form of therapy, or as a storage unit.[1] The diversity and scope of biomaterials science research, and especially its application to the improvement of trauma, disease, and congenital defects in the human condition, are making this branch of science increasingly dominant and topical in many countries. An exciting aspect is that such research is interdisciplinary. The varied problems of the human condition that biomaterials research addresses occupy the efforts not only of medical doctors who act as the end users of such technology, but also those of chemists, physicists, engineers, and biologists in creating the technological advances. Chemistry, in particular, plays a major role in such research, after all it is the foundation stone on which biomaterials polymer science and biomedical scaffold materials are built.

The replacement of any bone due to disease or trauma needs an implant. This implant can fall under one of three categories: autograft (fresh, living bone tissue harvested from somewhere else on the patient's body such as the hip), allograft (living bone tissue donated by other individuals that is sourced from a *bone bank*, or xenograft (a synthetic bone substitute that lacks any living component but which could potentially act as a *scaffold* to support cells, *etc.* While autografts represent the *gold standard* of bone implants in terms of minimisation of rejection issues, the pain of bone harvesting and the limitations of how much to harvest are disadvantageous. Allografts, alternative bone replacement materials, can have body rejection issues that couple with risks of disease transmission and paucity of material available for implants. The rationale, therefore, for development of xenograft materials is to reduce reliance on autograft and allograft bone. Within the area of biomedical materials, the quest for suitable materials that act as *osteoconductive* xenograft scaffolds *viz.* ones capable of supporting new bone deposition and its proliferation, has been an avid subject globally. The keenness of interest in xenograft implantation materials in general has been further accentuated by the recently emerging area of *Tissue Engineering* which, according to Langer and Vacanti[2] (the pioneering scientists in the field) *applies the principles of biology and engineering to the development of functional substitutes for damaged tissue.* In contrast to conventional xenograft scaffold research, the *scaffold* referred to in Tissue Engineering is effectively a biodegradable (usually polymeric) three dimensional device. It serves as a cell transplant vehicle for bringing about formation of the structural and functional tissue units by the cells that have been transplanted.

## Bone and its Chemical and Morphological Characteristics

Bone is a living interdigitated (or interlayered) composite of collagen protein and calcium phosphate platelets, the main mineral phase of which is carbonated *calcium hydroxyapatite*. Calcium hydroxyapatite, stoichiometrically $Ca_{10}(PO_4)_6(OH)_2$, is the *hydroxy end group member* of the complex apatite family and it has a more complex chemistry than the related fluorapatite and chlorapatite compounds. When prepared by precipitation from aqueous solution, the Ca:P ratio varies from 1.50-1.66 rather than being reproducibly 1.67, the value expected from its stoichiometry.[3] Structures categorised as being part of the apatite family have had their generic descriptor coined from the Greek word apato - deceit.[4] They have the characteristic and interesting property of substitutional lability in that the cationic and anionic components of the lattice structure, *e.g.* the $-PO_4$, -OH and $Ca_{2+}$, can be partially replaced with others when in solutions containing exchangeable ions. When lattice substitutions occur in calcium hydroxyapatite, the physical properties of the solids, *i.e.* their solubility and crystal morphology, can change markedly from the state prior to the lattice substitutions. Biomineralization processes lead to bone deposition in the body and they occur in a complex physiological fluid. The composition of the mineralized apatitic portion of bone is not stoichiometrically pure but rather it exists as a carbonated calcium hydroxyapatite doped with various other inorganic elements and/or organic ions. It was proposed in 1958 by Neuman *et al.*[5] that the mean composition of *bone* was $[Ca_9(H_3O)_2(PO_4)_6][Ca, Mg_{0.3}, Na_{0.3}, CO_3, citrate_{0.3}]$. A more recent report[6] has stated that its chemical composition can be approximately given by $Ca_{8.3}\square_{0.7}(PO_4)_{4.3}(HPO_4$ and $CO_3)_{1.7}$ (2OH and $CO_3)_{0.15}\square_{1.7}$ where $\square$ can equate to a lattice vacancy. In reality, it is difficult to give an exact composition of bone as it varies with species, the age of the vertebrate, and the location of bone in the body. Bone also has a specific architecture consisting of *cortical* bone, the harder outer layer of bone, and *cancellous* bone, which is described as being the softer, spongier porous portion residing inside the bone. The cancellous architecture is created by deposition and resorption processes that occur during bone formation as a result of the actions of bone tissue-associated cells such as osteoblasts, osteoclasts and osteoblasts.[7] The overall porous architecture of bone that acts as a hard tissue support for the cellular or *parenchymal* (living) component of bone is characterised by its interconnected porous channels, known as the *trabecular network*, that allow the transport of blood

through this living tissue.[8]

Any hard tissue replacement material for bone must thus attempt to replicate the typical bone architecture and be composed preferably of calcium phosphate materials that are not only biocompatible with the body but also able to be *remodelled* interfacially. This then allows a) new bone-apatite to be co-deposited with collagen after a process of dissolution-re-precipitation of the bone-implant interface and, b) new bone tissue to penetrate the implant at the bone-implant interface to provide a securely bonded bone-implant interface that effectively lodges the bone in its position. The so-termed *bioactivity* of the bone replacement material is important as such materials allow a direct chemical bond (without any so-called intervening fibrous tissue) to occur between natural bone tissue and the implant. Calcium hydroxyapatites have this property. This contrasts directly with the so-called *bioinert* (or biotolerant) materials represented by *e.g.* hard ceramics such as alumina, zirconia, stainless steel, and titanium. These *bond* to bone tissue purely through an intervening fibrous tissue layer of varying thickness which separates natural bone tissue from the implant.[8] Given these needed attributes for bone tissue replacement, cancellous bovine bone arguably is the ideal, almost ready-to-use material. After processing (see below), this bone already possesses the desired mineral composition and the necessary architecture to allow bone modelling at its interface as well as tissue in-growth.

Our main focus on producing xenograft bone replacement materials has been to use the plentiful and relatively low cost animal bone from our large cattle herds. This can be done in NZ because of the strict auditing/tracking and MAF/biosecurity procedures that show the country, historically, not to have had any serious, notifiable diseases afflicting its herds and especially *Bovine Spongiform Encephalopathy* (BSE). It has been allegedly that consumption of meat affected with this disease led to human contraction of the fatal brain-wasting disease known as *variant-Creuztfeldt-Jacob disease* (*v*-CJD) in the UK. The continued BSE-free status of NZ (and Australia) allows bone material for biomedical applications to be sourced from the country's mainstream cattle herds rather than from expensive controlled herds, *viz.* specially selected and certifiably BSE-free, that have to be used overseas for bone-sourced biomedical materials such as Bio-Oss®.[9,10] From the NZ perspective, this has the potential to produce a cheaper, high value biomedical commodity out of a traditionally low value material currently used for fertiliser or disposed of into the environment.

## Processing Bovine Bone into Xenograft Cubes or Powders

The work performed at MIRINZ (NZ Meat Industry Research Institute) and Waikato's Chemistry Department in generating xenograft materials and powders from bovine bone has been described previously and the serious reader is directed to the literature.[10,11]

### *Xenografts*

For the preparation of xenografts, it is necessary to cut cubes of cancellous bone from the *condyle* portion of the bovine femur bone using a sharp band saw. The condyle is the rounded part of a bone (here, the femoral bone of a bovine that supports its bulk) that fits into the socket of another bone to form a joint. The reason bovine condyles are suited to forming xenograft cubes lies in the relatively large size of the condyles. There is enough cancellous bone in a typical (mature) bovine condyle to produce three to four cubes of materials *ca.* 25 $mm^3$ each, by cutting.[10] Other species of animals common to NZ agriculture such as sheep, deer, or even ostriches have femoral condyles which are either too small (sheep and deer) to allow the cutting or are overly spongy with a high fat content (ostrich). Bone from other parts of the bovine skeleton, such as the rib can be processed, but it is more useful for forming re-precipitated hydroxyapatite powders by acid dissolution processes rather than as xenografts.

In cutting the cubes, only frozen femoral condyles from supermarket abattoirs are used so as to have minimal sealing of the bone pores by frictional heat-induced collagen-to-gelatin transformation during cutting. The bone cubes are then boiled in water (conventional pressure cooker for 6 h and *ca.* 15 psi) to remove the bulk of the blood and fat present in the cubes. Initial work[10,11] at Waikato involved pressure cooking of the bones followed by a 16 h soak in NaOH, water rinse, and microwave heating of the bones (in water) to bp (to assist in fat removal). The bone cubes were then refluxed in AcOMe, which has a high affinity for fat, and then vigorous shaken or blown with compressed air to remove excess liquid prior to final drying. Cubes that contain fat are yellowed in appearance (Fig. 1). Deproteination of the defatted cubes was the next processing step and this was achieved by immersing the defatted cubes in simple oxidising agents (NaOCl or $H_2O_2$). This removes the bulk of the collagen protein which, when interdigitated with carbonated hydroxyapatite, gives the bovine bone a considerable degree of hardness. Bone containing fat is yellow, defatted bone less so, but that with the protein removed has a whitened chalky consistency (Fig. 1). The last corresponds to carbonated hydroxyapatite with a significant loss in mechanical strength.[12] This attribute means that it can be shaped for the desired implant by using a knife, scissors or a trephine (Fig. 1 shows a shaped implant).

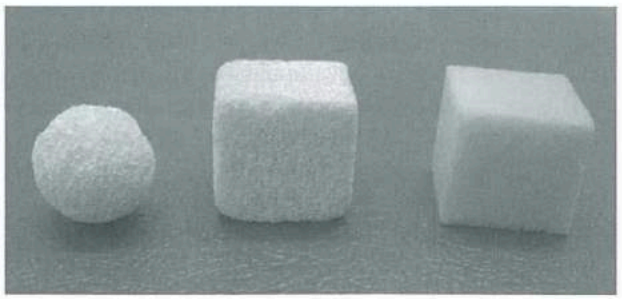


***Fig. 1.*** Bovine cancellous bone specimens. R to L: bovine cancellous bone cube prior to reflux with MeOAc showing the yellow colouration due to fat, defatted and deproteinated bovine cancellous bone as a bleached and chalky cube, and a shaped piece of defatted and deproteinated bone.

In later research carried out by Mucalo and Foster[13] and continued currently by Laird, Mucalo and Dias,[14] the pressure cooked bone was not subjected to the time consuming solvent-assisted defatting and bleach-assisted deproteination procedures. Instead it was placed in alumina crucibles and sintered at 1000 °C in a muffler furnace for several hours to burn off organic matter and leave the brittle white mineral shell of the bone intact with its porous architecture (Fig. 2). Under these conditions, the bone mineral transforms from partially crystalline carbonated hydroxyapatite into crystalline hydroxyapatite.

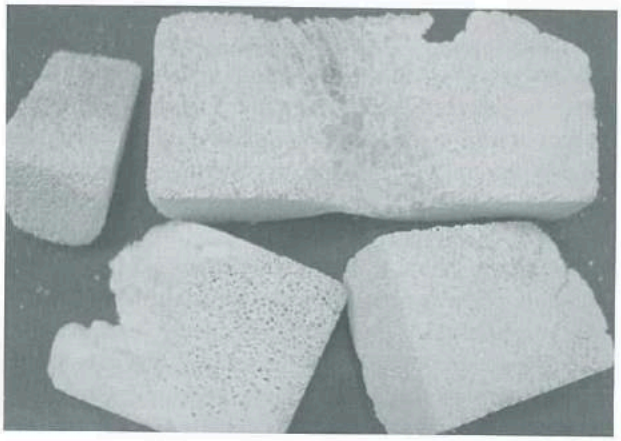


***Fig.2.*** Bovine cancellous bone after sintering at 1000°C for 3 h.

### *Powders*

A variety of methods have been used at Waikato to produce powders from bone.[11] In previous studies milled animal bone powder was produced directly by crushing raw bone from a variety of available animal types, *e.g.* rib bones, sheep, bovine, corvine bone, *etc.*, in a hydraulic press at 100 psi and then pressure cooking for 4 h to remove tissue and fat. After drying the bone chips were ground further in a hammer mill to particle sizes <2 mm diam. Further processing, such as AcOMe reflux (to remove further traces of fat), NaOH treatment, or more commonly acid-dissolution/re-precipitation (using NaOH) was then possible.

Although the acid-dissolution/re-precipitation methods produced powders, the residual fat and protein by-products arising from using raw bone as a starting material produced many problems, even when an intervening pressure cooking step was used. For example, performing the acid digestions in $HNO_3$ led to orange colouration of the resultant hydroxyapatite powders due to so-called xanthoproteic reactions,[13] which arise from interactions of the protein residues in collagen with the $HNO_3$. Even HCl digestions (in which xanthoproteic reactions are absent) of the milled bone powders led to opaqueness, most likely due to suspended collagen or fat. To remedy this, research by Mucalo and Foster[13] involving cleaner acid digestion/re-precipitation of already sintered bone pieces was performed. This minimized problematic bone matrix-associated organic impurities by the burn off of these. Cleaner, white powders were obtained, especially from the $HNO_3$ dilutions, leaving only the washing out of $NO_3^-$ from the powders after re-precipitation in the subsequent cleanup process. Subsequently, the process developed[14] was employed to provide kg-scale re-precipitated hydroxyapatite powders for plasma spraying. Here, the powders were passed through an Ar plasma under conditions where the re-precipitated hydroxyapatite particles become partially molten and can be impacted onto stainless steel or titanium metal surfaces to form a *plasma spray coating* (another biomedical type application).[15] Such coatings render metallic surfaces such as titanium or stainless steel more bioactive and give them the ability to bond more strongly to natural bone tissue through the mechanochemically bonded hydroxyapatite layer; an example of such a coating is shown in Fig. 3. The heterogeneous and porous nature of the plasma-sprayed hydroxyapatite coating not only improves the bioactivity of traditionally bioinert stainless steel or titanium substrates but may also provide a means of tissue in-growth so improving the bone-coating bonded interface.

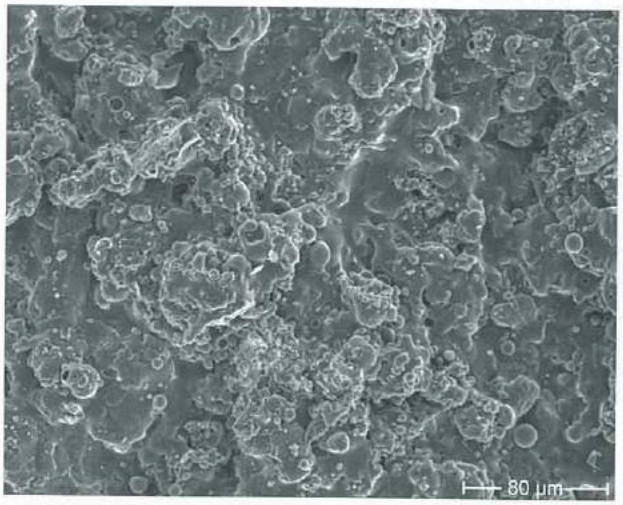



***Fig. 3.*** An SEM micrograph of plasma sprayed calcium hydroxyapatite coating on a titanium plate. The feedstock powder for this coating was produced by David Foster by re-precipitation from an acid digest of sintered NZ bovine bone.

## Spectroscopic and Microscopic Characterisation of the Xenografts and Powders Derived from Animal Bone

A wide range of spectroscopic, microscopic and other physical or mechanical testing techniques can be used to follow the chemical and physical changes that occur in the bone materials as they are processed for biomedical purposes. Thus, X-ray diffraction, solid state NMR and IR spectroscopy, atomic absorption spectroscopy (AAS), inductively coupled plasma optical emission spectrometry (ICP-OES), X-ray photoelectron spectrometry (XPS), scanning electron microscopy/energy dispersive X-ray analysis (SEM/EDXA), differential scanning calorimetry (DSC) and mechanical testing techniques (aimed at measuring the bulk modulus and yield stress values of the bone) have been used in our studies to characterise the bone matrices as a function of processing.[10-12]

Generally, IR, solid state magic angle spinning (MAS) $^{13}C$ NMR, and (to a lesser extent) XPS showed that the main changes during boiling/defatting/deproteination that led to the xenograft bone were due to the removal of fat and protein. As mentioned above, the remaining mineral residue retaining the original porous architecture of the

bone was hydroxycarbonate apatite.[10] This was confirmed by an IR spectrum of the crushed bone (Fig. 4) with peaks characteristic of carbonate at 1451 and 872 $cm^{-1}$ in addition to the apatitic phosphate-associated vibrational modes at 1036, 605 and 564 $cm^{-1}$, respectively. However, when the defatting and bleach-assisted deproteination are replaced by sintering the boiled bone at 1000°C, the bone mineral remaining is no longer the partially crystalline carbonated hydroxyapatite apatite; transformation to crystalline calcium hydroxyapatite occurs (Fig. 5), as evidenced by weak peaks at *ca.* 1450 $cm^{-1}$ that indicate the carbonate stems from surface interactions between atmospheric $CO_2$ and CaO present within the sintered bone. SEM micrographs of boiled/defatted/bleach-deproteinated bone specimens showed the successful retention of the macroscopic structural detail of porous, cancellous bone along with the needed interconnected porosity channels for successful integration of the implant *in vivo*. For acid-digested/re-precipitated powders derived from bone, IR spectra show features typical of poorly crystalline calcium hydroxyapatite; remaining features are usually due to carbonates substituted into the calcium hydroxyapatite lattice structure. These latter carbonates can arise by $CO_2$ contamination during the alkali-induced re-precipitation.

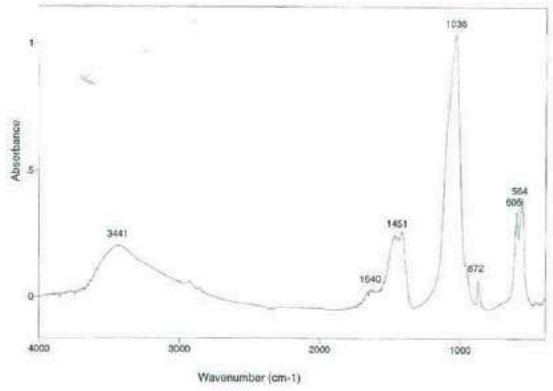



*Fig. 4.* FTIR spectrum (KBr disk) of ground bovine cancellous bone after subjection to boiling/defatting and deproteination.

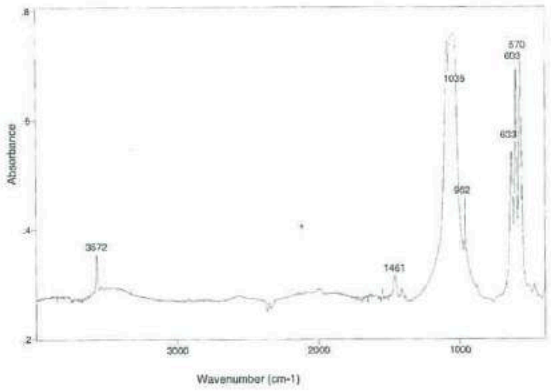



*Fig. 5.* FTIR spectrum (KBr disk) of ground bovine cancellous bone after sintering at 1000°C for 3 h.of the calcium hydroxyapatite from the acid digests.

Material strength tests predictably showed that prolonged boiling (6 h) followed by the deproteination hypochlorite treatment to give the chalky bone had a deleterious effect on the overall mechanical strength of the bovine bones tested.[12] This is desired because the xenograft material that results is easily shaped. It is important, however, not to over-process otherwise attempts at shaping can result in complete collapse of the xenograft due to extreme brittleness.

## *In vivo* Study of the Implanted Boiled/ Defatted/Deproteinated Bovine Bone in a Sheep Model

The success and safety of an implant acting as a bone substitute can only be demonstrated properly through a series of *in vitro* and subsequently *in vivo* testing procedures. *In vitro* testing involves subjecting the xenograft to a series of tests that evaluate its biocompatibility/irritation prior to placement in a living organism. One such widely used test is the hen's egg test-chorioallantoic membrane (HET-CAM) test. This is described as a biocompatibility test intermediate to classical *in vitro* and *in vivo* test protocols.[16] The test assesses the irritation potential of a particular substance by applying it directly to the highly vascularized chorioallantoic membrane of a developing chicken embryo that is <10 days old. Under these conditions there is no visibly noticeable embryo, rather a beating heart in the middle of a network of vasculature (Fig. 6) which would develop over time into a full sized chick embryo given the correct incubation conditions. A scoring system[16] evaluates the potential for tissue irritation, *e.g.* haemorrhaging by contact of the material with the blood vessels. Other tests that evaluate the material's biocompatibility for physiological environments involve the responses of cells (from specially grown cell lines) to contact with the implant materials.[17]

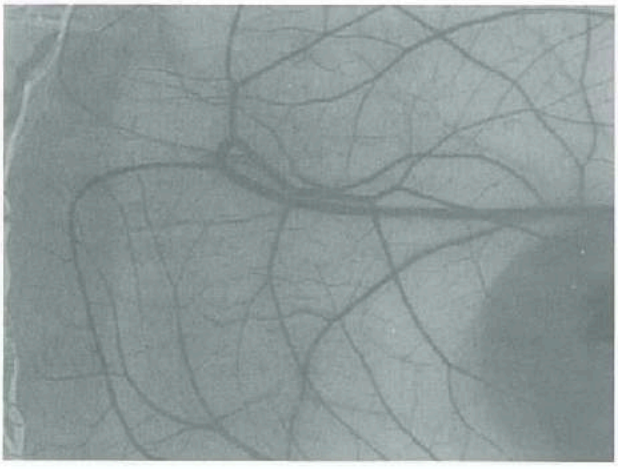


*Fig. 6.* Hen egg chorioallantoic membrane (<10 days old) used to test materials for biocompatibility with living tissues. The dark image at right is the embryo chick heart. Typical test protocols note the response of the vasculature to the materials. Photo courtesy of Dr Kavitha Babu, AgResearch, Ruakura.

The boiling/defatting/deproteination process of bovine bone was presumed sufficient to render it immunologically inert for *in vitro* testing. One must note that fat removal is important because its presence in an implant can make it *antigenic*, *viz.*cause infection once surgically inserted.[18] However, the existing wide body of clinical knowledge

relating to *in vivo* bovine-derived materials in living organisms argued for a surgical protocol for *in vivo* implantation of the bovine bone xenografts. This was developed by Warrick Bruce and Geoffery Horne (Massey University and Wellington School of Medicine, respectively) and approved by the Massey Animal Ethics Council. Surgeries were carried out at Massey's Veterinary Teaching Hospital and the Wellington School of Medicine, and the Waikato bone cubes were implanted in an ovine femoral defect model to evaluate their efficacy as an osteoconductive bone graft; the details of this work have been described in the biomedical literature.[19] The surgical protocol involved the excising of an autograft bone sample from the sheep's femur so at to create a defect for the processed bovine *xenoimplant*. The excised autograft bone tissue was placed as a control in a defect created in the opposite femur of the same test animal. Twelve mature ewes with weights 55-70 kg were used in the study so as to avoid the normal growth processes of younger animals and minimize impact upon xenoimplant incorporation. The trephine implement removed 8 mm O.D. cylindrical cores of the xenoimplant from the (boiled/defatted/deproteinated) Waikato bone and this was sterilised (γ-radiation) prior to implantation. The deposition of new bone tissue around the implants was monitored at 15-17, 30-32, and 56 days after surgery using fluorochrome label dyes that had been intravenously introduced at certain time periods after the surgical implantation. The sheep were humanely euthanized ten weeks after surgery and the distal femurs harvested. De-muscled bone sections X-rayed and then non-decalcified thin sections embedded in resin for fluorescence microscopy study. Overall, the study supported bovine bone as osteoconductive in the ovine model used and the fluorescent labelling showed that new bone material grew over the implant. Furthermore, the xenoimplant performed similarly or *better* to the autograft in osteoconductivity. This unexpected observation has been ascribed tentatively to the open, porous nature of the bovine xenoimplant after trephining. This contrasts to the polished and sealed surface of the autograft which would have delayed tissue in-growth. In particular, the study has confirmed that boiled/defatted/deproteinated bovine bone is workable, easily shaped, and compatible to surgical procedure.

Further surgical work at Massey[20] implanted boiled/defatted/deproteinated bovine bone in a defective paw of a family's pet dog. The void in the dog's paw bone was cleaned out and replaced with a grafting mixture of autogenous cancellous bone and the Waikato xenoimplant. This procedure has as its ultimate aim not simply reducing dependence on the quantity of harvested autogenous bone needed but to use the osteoinductive properties of the autogenous bone for stimulation and propagation of bone growth and lead to bone growth in the osteoconductive xenoimplant portion of the graft.

The dog's paw was X-rayed post-operatively at 1, 2, 6, and 10 months and showed over this time overall densification of bone in the region of the void. The dog itself was sound throughout this period and is still believed to be alive today at time of writing. Thus the bone developed at Waikato has immediate use as a xenograft material for veterinary purposes.

## Current and Future Activities

Work at Massey University on implantations of the Waikato bovine bone in dogs continues while collaboration with Otago is on the development of a sintered bovine bone material with the strength lost by collagen removal partially restored. Biocompatibility testing of these materials using the HET-CAM, as well as separate trials employing a specific cell line, viz. L929 (to assess whether cells proliferate on these materials) would have been conducted by the time this article has gone to press. Eventually, an *in-vivo* implantation trial in a suitable animal model (likely sheep) will also be considered on the basis of the initial biocompatibility testing results.

## Conclusions

It has been shown that NZ-sourced bovine bone provides a useful bone replacement material for veterinary applications and holds strong promise in the future for human applications. Thus value has been added to what was formerly a low value resource in agriculture. Hopefully it will create a new, specialist niche export market for this country.

### Acknowledgements

The work described in this paper could not have been carried out without the efforts, supervisory involvement or consultancy services of a number of people situated at the University of Waikato, MIRINZ, Massey University, Wellington School of Medicine, IRL Ltd, (Auckland), and the Chemnitz Technical University,(Germany). Financial assistance from the Waikato Medical Research Foundation, the Maurice and Phyllis Paykel Trust and the PGSF is gratefully acknowledged. The plasma spraying studies (Germany) were enabled through the generous funding assistance of the Deutscher Akademischer Austausch Dienst (DAAD). The efforts of former Waikato MSc students, Glenn Johnson and David Foster, are also acknowledged.

## References

1. Bruck, S.D. *The International Journal of Artificial Organs* **1978**, *1(2)*, 63-66.
2. Langer, R.; Vacanti, J.P. *Science* **1993**, *260*, 920-925.
3. Elliott, J.C. *Structure and Chemistry of the Apatites and other Calcium Orthophosphates*, Elsevier Science: B.V. 1994.
4. Aoki, H. *Medical Applications of Hydroxyapatite*, Ishiyaku EuroAmerica, Inc.: Tokyo, 1994.
5. Neuman, W.F.; Neuman, M.W. *The Chemical Dynamics of Bone Mineral*, Chicago University Press: 1958.
6. Rey, C. *Calcium Phosphates in Biological and Industrial Systems*, Kluwer Academic: Boston 1998.
7. Simkiss, K. *Bone and Biomineralisation*, Edward Arnold: London 1975.
8. Hench, L.L.; Wilson, J. *An Introduction to Bioceramics*, World Scientific: Singapore 1993; Krajewski, A.; Ravaglioli, A. *Integrated Biomaterials Science*, Kluwer Academic/Plenum Press: New York 2002.
9. Details on Bio-Oss® can be found at *http://www.osteohealth.com*
10. Johnson, G.S.; Mucalo, M.R.; Lorier M.A. *J. Mat. Sci.: Mat. Med.* **2000**, *11*, 427-441.
11. Johnson, G.S.; Mucalo, M.R.; Lorier, M.A.; Gieland, U.; Mucha, H.

Article

*J. Mat. Sci.: Mat. Med.* **2000**, *11*, 725-741.

12. Anderson, I.A.; Mucalo, M.R.; Johnson, G.S.; Lorier, M.A. *J. Mat. Sci.: Mat. Med.* **2000**, *11*, 743-749.
13. Mucalo, M.R.; Foster, D.L. *Croatica Chem. Acta* **2004**, *77*(3), 509-517.
14. Laird, D.F.; Mucalo, M.; Dias, G. *Austral. Soc. Biomat. Conf.* Mt Eliza, Melbourne, 2007.
15. Mucalo, M.R.; Foster, D.L.; Wielage, B.; Steinhaeuser, S.; Mucha, H.; Knighton, D.; Kirby, J. *J. Appl. Biomat. Biomech.* **2004**, *2*, 96-104.
16. Luepke, N.P.; Kemper, F.H. *Food Chem. Toxicol.* **1986**, *24(6-7)*, 495-496.
17. Suzuki, T.; Yamamoto, T.; Toriyama, M.; Nishizawa, K.; Yokogawa, Y.; Mucalo, M.R.; Kawamoto, Y.; Nagata, F.; Kameyama, T. *J.Biomed. Mat. Res.* **1997**, *34*, 507-517.
18. Frayssinet, P.; Asimus, E.; Autefage, A.; Fages, J. *J. Mat. Sci.: Mat. Med.* **1995**, *6*, 473-478.
19. Worth, A.; Mucalo, M.; Horne, G.; Bruce, W.; Burbidge, H. *Clin. Oral Impl. Res.* **2005**, *16*, 379-386.
20. Worth, A.J.; Thompson, K.G.; Owen, M.C.; Mucalo, M.R.; Firth, E.C. *NZ Vet. J.* **2007**, *55(3)*, 143-148.

# Do We Expect Too Much? Reflection on Chemistry Content in Higher Education†

Richard K. Coll

University of Waikato (e-mail: *r.coll@waikato.ac.nz*)

† A publication from the NZIC Chemical Education Special Interest Group

## Learning Science

Education research in the 1970s, like other related areas, was dominated by quantitative work[1] during an era for which *social sciences* sought to draw upon the successful *scientific* approach typically used in the physical sciences (in particular) to investigate teaching and learning.[1,2] So if we felt a cohort of students did not understand some concept, we tried to find out whether or not a different teaching approach could *fix* their misconceptions.[3] But how to do this? Well, drawing on a scientific approach, we would divide the class or classes up, teach one cohort the same way we always had, and the other cohort in our new way, and evaluate any differences in conceptual understanding using, *e.g.* a standardized topic test. Differences would be examined for statistical significance of evidence that our new approach to teaching had worked. And this is the way much research was done at the time. Control of variables, randomized sampling, and so on, were all embedded in such an approach to educational research.

At about this time, however, key research – some of it NZ-based – suggested teaching and learning was rather more complex. Investigation into how students arrive at their own views of scientific concepts, focused on student misconceptions, or alternative conceptions, viz. students' views that are at variance with the accepted scientific viewpoint. Perhaps it is not that surprising that students harbour misconceptions for abstract concepts such as the kinetic theory, electricity, and force. But some student views of more common concepts are less easily understood and it is likely that they are influenced by other factors such as cultural background. There are some unusual examples reported in the literature. For example, one study of misconceptions of Papua-New Guinean students found some to believe that pregnancy occurs when a spirit child enters a woman rather than as a result of sexual intercourse.[4] A Caribbean-based study found that some students believed that hair would grow more rapidly if it was cut during the full moon.[5] Other researchers have suggested that some student misconceptions may arise as a result of the learning process itself.[6] These studies might seem curious or odd but, overall, such studies suggested that factors other than the school environment and the teaching processes used were also influential in student learning. There are now huge bibliographies of student alternative conceptions complied, some with several thousand studies detailed.[7]

What is perhaps of more concern is the *remarkable tenacity of many student misconceptions*. Students in many cases seem unwilling to give up their prior beliefs even after instruction.[8-10] Similarly, early research by Osborne and colleagues[10] suggested that even very able students, *i.e.* those who passed exams with high marks, did not actually understand fundamental scientific concepts in ways we would desire.

What might be the overall origins of such problems, and what might we do about it? Let me consider this by looking at what I think is a key factor; high, *perhaps unrealistic*, expectations of our students.

## Learning Chemistry in Higher Education

As mentioned above, considerable concern has been expressed in the literature about the high incidence, and remarkable tenacity, of common student misconceptions. The vast bulk of this research is concerned with school students, but similar issues are reported also for students of advanced chemistry from the higher education sector. Some higher education research reports give a real sense of frustration experienced by teachers or lecturers as they struggle to deal with student misconceptions.[11-16] While there are a number of concepts that students traditionally find difficult such as aspects of physical chemistry, like thermodynamics and electrochemistry,[11] researchers seem more concerned at the prevalence of student misconceptions for *even very simple concepts*[12,15,16] For example Herron[16] comments that for his first-year chemistry students *fewer than 50% of the students seemed to comprehend that it was $Cl^-$ that was in table salt and not $Cl_2$ or that*

*there was a difference between the two* (see p.146).

There is a general feeling expressed in the literature that such student misconceptions are related to prior learning experiences (or lack thereof!), although some authors suggest that it may be more related to the students' level of cognitive development.[15-17] One key factor I suggest may be *the large amount of factual material that students are expected to memorise* when developing understanding of a complex body of knowledge like chemistry.[14,18]

A brief review of course material for any one of many chemistry courses shows that we expect students to memorise a large amount of material, and often at the same time demand advanced problem-solving skills. An abridged course outline for third-year analytical chemistry I once taught is given as Fig. 1.

**Advanced Analytical Chemistry**

Isotope chemistry. Methods of detection, measurement, safe handling and application of isotopes to Chemistry, Biology and the Earth Sciences. Stable isotopes. Interfacing of analytical instruments with computers. Experimental design. Statistical treatment of data. Quality control. Chromatography. Atomic absorption spectroscopy. Mass spectrometry. X-ray diffraction. Electrochemical analysis.

*Fig 1.* Topics for advanced analytical chemistry course

High-level understanding of other related disciplines typically is also presumed, particularly at advanced levels of study. For example, in advanced postgraduate level structural chemistry courses we require advanced mathematical ability and a thorough understanding of many advanced physics concepts.

Consider the following extract from a small portion of one lecture on single-crystal X-ray diffraction delivered to an advanced level chemistry class (Fig. 2).

The interaction of X-rays with the planes of a crystalline lattice is dependent upon the position of the individual atoms, or more correctly elements of electron density, present in or close to the crystal planes. Assuming discrete (i.e. atomic) scattering sources, the problem becomes one of the superposition of waves of different amplitudes and phases. Thus upon interaction with a given set of crystal planes a wave of total amplitude F, has X and Y components,

$$X = \sum f_j \cos \delta_j \text{ and, } Y = \sum f_j \sin \delta j$$

where $f_j$ is the atomic scattering factor for the $j^{th}$ atom, and $\delta_j$ is the phase for $j^{th}$ atom.

The modulus of the scattered X-ray beam is given by,

$$|F| = \sqrt{(X^2 + Y^2)},$$

and the phase is given by the arctangent of the ratio of the Y and X components *i.e.*,

$$\alpha = \arctan(Y/X).$$

The periodic nature of the unit cell restricts the allowed values for $\delta_j$ such that,

$$\delta_j = 2\pi(hx_j + ky_j + lz_j)$$

where h, k, and l are the Miller Indices for a given set of crystal planes, and $x_j$, $y_j$, and $z_j$ are the atomic co-ordinates for the $j^{th}$ atom expressed as fractions of the unit cell lengths.

We can write,

$$A_{hkl} = \sum f_j \cos 2\pi(hx_j + ky_j + lz_j) \text{ and,}$$

$$B_{hkl} = \sum f_j \sin 2\pi(hx_j + ky_j + lz_j).$$

Thus the total phase and amplitude for the wave becomes,

$$\alpha = \arctan (B/A) \text{ and,}$$

$$|F_{hkl}| = \sqrt{(A^2 + B^2)}.$$

It is convenient to express the above using complex numbers as,

$$F_{hkl} = A + iB.$$

The complex quantity $F_{hkl}$ is known as the structure factor.

Since $\exp(ix) = \cos(x) + i\sin(x)$, the structure factor can be written as a complex exponential term,

$$F_{hkl} = \sum f_j \exp[2\pi i\theta],$$

where $\theta$ is $hx + ky + lz$.

Assuming infinitesimally small elements of electron density rather than discrete atomic scattering sources, we express $F_{hkl}$ as an integral rather than a summation thus,

$$F_{hkl} = \int_v \rho(xyz) \exp 2\pi i\theta . dv,$$

where $\rho(xyz)$ is the electron density at point xyz.

Fourier transformation yields an expression for $\rho(xyz)$ in terms of the structure factor $F_{hkl}$,

$$\rho(xyz) = \sum_h \sum_k \sum_l F_{hkl} \exp(-2\pi i\theta).$$

From this expression we can calculate an electron density map for the entire contents of the unit cell and this will reveal regions of high electron density corresponding to atomic positions giving the molecular structure for the material under study. In principle the Fourier series should be evaluated for all values of *hkl* from $-\infty$ to $+\infty$. Bravis lattice restrictions and symmetry constraints do not allow this, however, and the result is small ripples in the calculated electron density map, particularly around the heavy atom positions.

*Fig. 2.* Portion of an advanced level lecture on X-ray crystallography.

Even a cursory examination of this brief portion of just one lecture clearly shows how much we expect from our students. We expect knowledge and expertise in trigonometry, differential and integral calculus, complex number theory, wave theory, atomic theory, electricity and magnetism, symmetry, and so on. This list is by no means exhaustive but it is immediately evident that we assume a remarkable in-depth knowledge of a number of highly abstract concepts.

There was a widely-held view amongst departmental staff in the institution where I worked when I taught such topics that this is exactly as it should be. The usual ratio-

nale of this is that it is important for us to maintain high academic standards in order to ensure the integrity and high reputation of our degree programs. In addition, it is viewed that such knowledge and skills are important for students engaging in postgraduate studies or research.

If we accept that such expectations are reasonable, the question remains as to whether we actually achieve the understanding and problem-solving skills we seek with our present instructional strategies? Despite reservations occasionally expressed about the students' abilities, once students reach the final year of the degree program they almost inevitably graduate. Hence, we are in effect indicating by virtue of their graduation that, in general, our students do meet these expectations. A simple illustration suggests that this may not be the case.

## Third-Year Chemistry Student Understanding of Atomic Structure

A representative sample of third-year chemistry students at one institution was briefly surveyed on their understanding of a concept that most of the teaching staff would consider very simple, namely fundamental atomic structure. By comparison with the X-ray analysis of Fig. 2, the concept of atomic structure as presented here is almost trivial.

The students were asked to sketch an appropriate representation for the electronic structure of the hydrogen atom and the carbon atom (Fig. 3). It is important to bear in mind that this exercise was carried out with a group of students that had graduated with a BSc at the end of the year this activity was conducted. Furthermore, many of these students had already been awarded good grades for previous chemistry courses, *e.g.* A (80-85%) and A+ (85-90%). Despite this, the incidence of student misconception was high. Only two respondents gave an answer that could be considered consistent with the currently held scientific view. The naiveté of the answers was quite remarkable. It seems that most of these students (*ca.* 70% of respondents) still think of atomic structure in terms of the Bohr model while some gave answers that were difficult to attribute to any recognisable model of atomic structure. The results presented here are far from rigorous, but they indicate of a lack of understanding of a fundamental and comparatively simple scientific concept. Since the Bohr model is not taught in first-year chemistry, it seems likely that this model of the atom represents prior knowledge that our senior students are bringing to the classroom.[19]

**ATOMIC STRUCTURE**

In the space below, please draw a sketch of what you understand the to be an appropriate representation for the electronic structure for the:

**Hydrogen Atom:**

**Carbon Atom:**

*Fig. 3.* Atomic structure questionnaire for third-year chemistry students.

## Content - Do We Want Depth or Coverage?

The latter half of the last century was characterised by enormous advances in science and technology that resulted in the demand for a more highly-skilled work-force. This demand has led to a large increase in students numbers studying science in high school and tertiary institutions,[20-22] and to a focus on more applied courses and vocationally-oriented degree programs.[21] Whilst this is shift may be appropriate, it does present some difficulties. For example, Buntting and co-workers, suggest that up to 50% of the intake of first-year science students lack understanding of key underpinning concepts.[22] Further difficulty lies in the enormous number of applied science topics now available, and teaching staff are faced with the difficult task of deciding what topics to include in their courses.

Many lecturers are uneasy about leaving out topics that they see as interesting and relevant to students, and there is a tendency to want to include as many topics as possible. However, research into learning and instruction suggests that it may be more beneficial to teach a few topics in depth, instead of trying to give a superficial coverage of a large number of different topics.[8] Moreover, it provides a deeper insight into how students acquire concept-knowledge and reasoning skills as suggested by Eylon and Linn.[8]

The argument here is that students need to develop their own concepts, see how to link new concepts with their existing concepts, and develop their own strategies for higher level activities such as problem-solving.[22] This, it is suggested, is problematic if they are overloaded with factual material, or encounter too much material at once. There are a number of factors that educators need to take into account during instruction,[12] namely content, organization and presentation of material, the student's level of cognitive development, and the students' level of prior knowledge.

The instructional strategies suggested by Eylon and Linn[8] are based on teaching by a more learner-centered or constructivist teaching approach. Interestingly, other educators have reported that less content is covered when teaching by a constructivist approach,[23] which fits in with reducing our emphasis on content coverage.[22]

The view that teaching institutions should teach less material and instead focus on developing greater learning skills is gaining increasing attention at tertiary teaching institutions in this country.[22,24] It also has been suggested that the increasing ease of access to sources of information such as the Internet means that fewer educators should place emphasis on the mere provision of factual material, and greater emphasis on higher-level cognitive skills.[22,24]

The overall focus for us, as chemistry teachers, should be to have clear aims and objectives for individual courses and degree programs. In other words, what we need is a clear picture of what knowledge and skills we want our students to possess upon graduation, and what instructional strategies we need to implement in order to achieve those aims. Research into learning and instruction sug-

Article

# Fighting Food Fraud with Science

Bea Perks

Deputy Editor, Chemistry World, Royal Society of Chemistry, Thomas Graham House, Science Park, Milton Road, Cambridge CB4 0WF, UK

Text reproduced from *Chemistry World*, **2007**, *4*(9), 48-52 by permission of The Royal Society of Chemistry (http://www.rsc.org/chemistryworld/Issues/2007/September/FightingFoodFraudWithScience.asp).

Does your extra virgin olive oil come from a Tuscan grove? Is your cappuccino made from the finest Arabica beans? Bea Perks meets some of the scientists subjecting our food's credentials to forensic examination.

Gone are the days of reading the word, 'olive oil' in a recipe, and trotting down to the pharmacist for a very tiny bottle of 'Olive Oil BP'. Today, supermarket shelves heave under the weight of a bewildering choice of olive oils - from a fruity Ornellaia ('marvellous for salads') to a light Ligurian ('ideal for deep-fried zucchini flowers'... yes, really). And it's not just olive oil. There was a time when people bought, simply 'beef' or 'chicken'. Now you can choose where your desired animal lived, or what it ate (mountain reared, corn fed). And so it continues - where did the cow live that produced the milk used to make the cheese?

'The big problem', says food scientist Peter Berry Ottaway, a consultant at the UK Institute of Food Science and Technology, 'is that any high value ingredient has a risk of being adulterated.' So how are the fakes wheedled out from the genuine - free-range, mountain-reared, organic - articles?

Ottaway's work is a stark illustration of the lengths scientists must go to in order to prove, or disprove, a product's authenticity. He once travelled to the Arctic Circle in search of cod and the plankton they eat - to determine if cod livers were the single source of a particular brand of cod-liver oil. 'It's very expensive and there's a huge temptation to cut it with rapeseed oil,' he explains.

The fatty acid profiles for the plankton, cod, and rapeseed weren't far apart, he recalls. But the picture changed when he looked at sterols. The sterols found in the Arctic plankton were the same as the sterols in the cod, but were not the same as the sterols in rape seeds. When he analysed the cod liver oil using HPLC he spotted 'there was something fishy - or not quite so fishy' going on.

The alarm bells had rung in the first place because the amount of cod liver oil leaving the processing plant was miraculously greater than the amount of cod liver entering the plant. The oil turned out to be about 12 or 13 % rapeseed oil, says Ottaway.

## The Perfect Crime

One of the greatest problems facing this branch of criminal investigation - food forensics - is that consumers can't always tell when they're being defrauded. The product might taste similar, will probably look similar, will cost about the same, and will not necessarily do the consumer any harm (apart from financially). For the same reason it's difficult to say quite how widespread the problem is. John Spink. Director of the Packaging for Food and Product Protection Initiative at Michigan State University, US, estimates the cost to the global food industry at $49 billion (£25 billion). 'In the UK, the Food Standards Agency (FSA) estimates the level of fraud at 10%, equating to around $7 billion,' he says.

Distinguishing pure from adulterated cod liver oil would certainly take an expert consumer, and even that might be a doddle compared with distinguishing pure from adulterated bee propolis. Bee propolis, for those not familiar with the stock of their local health food store, is a by-product of honey production. It is a wax-like substance that bees collect from buds, and then use as a sort of cement for hive maintenance. When not blocking out drafts in beehives, propolis is a popular food supplement or ointment ingredient. It is reputed to alleviate a long list of ailments including inflammation, viral diseases, ulcers, burns - the list goes on.

But the genuine article is not easy to identify. Propolis can be more or less any colour from green to red, and it is expensive - 25 mL of a solution of unspecified concentration costs about £10 - making it a fraudster's dream.

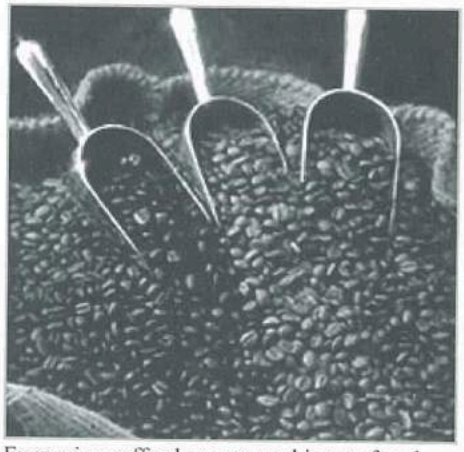
Bees have even been known to make fraudulent propolis themselves, sometimes exploiting window putty in place of plant bulbs. Forensic analysis of propolis has shown that the genuine article - not even meddled with by the bees themselves - contains distinct proportions of particular flavonoids (plant metabolites): a discovery that could help nail genuine fraudsters, and improve propolis quality overall by flagging up the window putty varieties.

## Technological Revolution

Fraudsters have capitalised on the variety and popularity of

***Left:*** The Manufacture of Oil, drawn and engraved by J. Amman in the Sixteenth Century (http://upload.wikimedia.org/wikipedia/commons/8/88/The_Manufacture_of_Oil_drawn_and_engraved_byJ Amman_in_the_Sixteenth_Century.png) and ***right:*** Italian olive oil: (http://upload.wikimedia.org/wikipedia/commons/e/e9/Italian_olive_oil_2007.jpg)

health products. One study instigated at the behest of Dutch trade officials discovered that a third of products purporting to contain pure aloe vera (a plant sap used to treat a wide variety of medical complaints) had been adulterated. The evidence for this particular study came from what was then a new chapter in food forensics: nuclear magnetic resonance (NMR).



Expensive coffee beans are subject to fraud

Aloe vera comprises three main components: glucose; malic acid; and the polysaccharide acemannan, which is composed of a long chain of mannose monomers. On average, each mannose monomer ring has one acetate group attached to one of three available positions, explains German food scientist Berndt Diehl, who discovered that the NMR profile of these different acetate groups represented an exact fingerprint for aloe vera. 'Manipulation of this signal is practically impossible,' Diehl wrote in a report of his findings back in 1998.

Today, NMR is just one of a long-list of food forensics techniques you might expect to find in a CSI -style forensics lab.

## Fingerprinting Food

Earlier this year, Italian researchers reported their findings using the polymerase chain reaction (PCR) to study mozzarella. PCR is a molecular biology technique used to replicate and amplify a small fragment of DNA. In this case, it was used to detect and quantify rogue cow DNA in mozzarella labelled as being made from pure water-buffalo milk (R M Lopparelli *et al. J. Agric. Food Chem.* **2007**, *55*, 3429).

Buffalo mozzarella is a highly sought-after Italian product certified by the European Protected Designation of Origin (PDO). Mozzarella can be made from cow's milk, but it wouldn't get PDO certification and wouldn't cost anywhere near as much either to make or indeed to buy. This has tempted food fraudsters to slip at least some cow's milk into the mix.

Barbara Cardazzo and colleagues at the University of Padua analysed 64 commercially available 'buffalo' mozzarellas by real-time PCR, and found that most of the samples were contaminated with cow's milk. The researchers say that their PCR technique, looking for a cow milk-specific gene, is a marked improvement on the current control method, isoelectrofocusing of milk proteins (separating the proteins according to their net charge by passing them through a gel). This can generate inaccurate results if the cheese has been subjected to high heats, *e.g.* when the milk is pasteurised.

But PCR also needs to be applied with care, warn researchers in the UK. Silvia Doveri and colleagues at the National Institute of Agricultural Botany in Cambridge showed that the genetic profile of olive oil isn't necessarily the same as that of the olive fruit. It could be a serious problem for anyone trying to tell if their fruity Ornellaia extra virgin olive oil (about £10 for 500 mL) really did come from the 2000 olive trees on the Ornellaia estate in Tuscany.

A host of certification systems, like the PDO enjoyed by buffalo mozzarella, exist to prove the worth of olive oils - from PDO, to protected geographical indication (PGI) and traditional speciality guaranteed (TSG). They are important awards recognised by the European Union referring to the quality of the olive oils. 'Before such awards are given, regulations imply detailed rules on the [olive] varieties to use, the geographical area of production, and the methods of oil extraction,' write Doveri and colleagues in a report of their findings (S Doveri *et al. J. Agric. Food Chem.* **2006**, *54*, 9221). 'As these labels reflect quality products they are awarded to command price premiums.'

## Plant Paternity Testing

Chemical analyses per se are not sufficient to verify olive oil authenticity, except in cases of adulteration with other vegetable oils, notes Doveri. So DNA markers - unique, short sequences of DNA that can be used to identify olive cultivars - are increasingly being applied to solve provenance issues.

There is a significant drawback to this, she says, because whole olive fruits are crushed in the milling process. The stone inside each olive fruit is an embryo, and has almost certainly been fertilised with pollen from another cultivar. 'Questions about paternal DNA on the genetic profiles need to be addressed before DNA markers can be used with confidence,' she notes.

Doveri says her group was the first to compare the DNA in leaves, olives and oil from a single olive tree. What they showed was that DNA in a leaf from the olive tree didn't match DNA in the oil. It's not an insurmountable problem, she says, because certified oils that are grown in specific regions may well have a limited, specific, number of possible pollinators - in which case such analysis might further support an oil's authenticity. But future analysis might be safer if restricted to specifically maternal markers - such as those found in mitochondrial DNA (which is only inherited from the mother).

## Black or White?

Another classic example of an expensive food product vulnerable to fraud is coffee, one of the most important food commodities in world trade, according to Gregory Tucker at the University of Nottingham, UK. The commercial coffee trade consists almost entirely of Arabica and Robusta coffee varieties, with Arabica considered the highest quality and, naturally, the most expensive. Arabica beans cost two

### In short

- Food fraud is a growing problem that costs the UK food industry hundreds of millions of pounds every year
- Foods and supplements including cod liver oil, aloe vera, mozzarella and coffee have been found to be adulterated
- A variety of chemical techniques are available to food forensic scientists, including NMR and genetic analysis - similar to the DNA fingerprinting used in criminal cases

to three times as much as Robusta beans and constitute more than 70% of the world's coffee production - so ensuring that inferior Robusta beans don't get into Arabica production chain is essential.

Most current methods to discriminate between Arabica and Robusta coffees fall under the analytical/instrumental heading, says Tucker. Pure varieties are distinguished according to profiles of analytes such as sterols, fatty acids, and total amino acids. Mixtures are characterised using Fourier transform infrared spectroscopy. The beans contain different amounts of the two main coffee compounds - chlorogenic acid and caffeine - which have distinctive infrared spectra.

DNA-based analysis is new to coffee authentication, but Tucker and colleagues say their work on PCR analysis and lab-on-a-chip capillary electrophoresis offers a quick and straightforward method suited to routine coffee analysis.

Tucker's team used PCR coupled with restriction fragment length polymorphism (RFLP), where amplified DNA is cut at specific sites along its sequence - using so-called restriction endonucleases - in order to determine, in this instance, particular coffee varieties. PCR-RFLP is a classic forensic technique, equally at home in murder cases and paternity testing as it is in food authentication. It is an essential component of DNA fingerprinting.

Tucker's method combined PCR-RFLP with capillary electrophoresis to separate individual genes and quantify adulteration in green (unroasted) coffee beans. A genetic marker in chloroplast DNA, which is maternally inherited like mitochondrial DNA, was found to differentiate Arabica from Robusta varieties.

## The Forensic Approach

Most of the above methods are targeted procedures, only applicable to one commodity and/or one type of fraud. But a pan-European effort is underway to develop more generic procedures for tracing and verifying food, by scientists within the Trace consortium, a €19 M (£13 M) European Commission initiative. 'The international team of scientists within the project are developing *food mapping* procedures that will allow provenance claims to be more easily checked,' says Paul Brereton from the UK Department for Environment, Food and Rural Affairs' (Defra's) Central Science Laboratory in York, the co-ordinator of Trace. They are attempting to link key parameters in food with those found in the local environment. By studying the climate and geology, the scientists aim to predict what profile of parameters should be expected in a food of given provenance. It is then relatively easy to check if the actual profile of the food matches with that predicted.

Alongside food mapping, Trace is also producing spectroscopic and biological fingerprinting methods that can be used to verify food. The increased power of data capture and interpretation techniques developed in recent years allows atypical samples to be rapidly identified. This is ideal for a food verification system as it allows a more forensic approach to authenticating food. 'Rather than us having to know what type of fraud is taking place, we can now quickly look for differences between products and identify what is causing those differences...', says Adrian Charlton, head of the NMR fingerprinting team at CSL and a key researcher in Trace.

Ten years ago there was much less media interest in food authenticity. But nowadays there's a much more discerning consumer - who makes their purchases based on what Brereton calls 'quality attributes'. These might be attributes that the consumer cares about deeply, but they can't always identify unaided, such as: provenance; production (GM, organic, free range); ethical issues (animal welfare, fair trade); and sustainability (food miles).

Brereton's team recently developed an analytical method to identify corn-fed chicken - chickens that command a higher price as a result of their relatively luxurious diet. The method exploits the differences between the biosynthetic pathways that exist between maize (C4 pathway) and temperate cereals such as wheat and barley (C3 pathway). C3 and C4 plants provide markedly different $^{13}C/^{12}C$ ratios when measured using stable isotope ratio mass spectrometry. Comparison with a database of results from chickens fed differing maize diets provides an objective means of confirming that a chicken was fed on corn (maize).

The CSL has worked in this area for 20 years: '15 years ago our main focus was in developing methods to prevent the [European] Commission getting defrauded through aspects of the Common Agricultural Policy – sugar mountains and wine lakes and all that – whereas the consumer is the focus now,' says Brereton.

Working out whether consumers are being taken for a ride looks set to focus national and international authorities and relevant experts for years to come. The FSA alone currently spends approximately £1 million each year on its food authenticity research programme.

'History has taught us that wherever there is a large price difference between two similar products and no objective means of checking is available, fraud can take place,' he stresses.

So you might think that a professional food detective would religiously scan the supermarket shelves for reputable labels before buying. 'No, in terms of food shopping, I tend to be a sceptic,' says Brereton. 'I buy based on what tastes good rather than what's on the label.'

## Acknowledgements

We are gratefull to Mark Peplow (Editor, *Chemistry World*) for facilitating reproduction of the text and Martin Howard, ESPRESSOWORKZ LTD., Mt Eden, Auckland (www.espressoworkz.co.nz) for providing the coffee beans artwork.

## Further Reading

1. Trace project: www.trace.eu.org/index.php Central Science Laboratory: www.csl.gov.uk.
2. Food Standards Agency: www.food.gov.uk.
3. Institute of Food Science and Technology: www.ifst.org.

# MALDI-TOF Mass spectrometry of Cyanobacteria: a Global Approach to the Discovery of Novel Secondary Metabolites

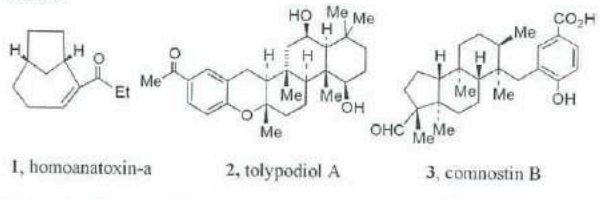
Jonathan Puddick and Michèle R. Prinsep

Department of Chemistry, University of Waikato, Private Bag 3105, Hamilton 3240
(e-mail: *m.prinsep@waikato.ac.nz*)

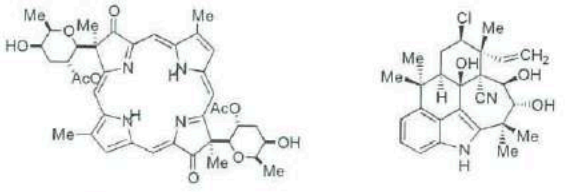
Cyanobacteria (blue-green algae) are a group of ancient prokaryotic organisms dating back between three and four billion years.[1] They have been attributed with oxygenating the earth's atmosphere[2] but, since the anthropogenic euthrophication of lakes, ponds and oceans, they have become synonymous with water hygiene issues.[3] This is due to the alteration of the nutrient composition of their habitat to one which is optimal for growth (or blooms). Cyanobacterial blooms may simply cause foul tastes and odours,[4] but they can also lead to the production of toxic secondary metabolites poisonous to humans and animals upon ingestion.[5] NZ has yet to suffer a human fatality, but the deaths of several dogs in Wellington was attributed to homoanatoxin-a **1** (Chart 1) from a *Phormidium* species.[6]

Although toxins are the most highly publicized cyanobacterial secondary metabolites, a vast array of compounds are produced which range in size, structure, and bioactivity. Terrestrial cyanobacteria have yielded diterpenes such as the anti-inflammatory tolypodiol **2**,[7] and the antimicrobial comnostins[8] such as comnostin B **3**, in addition to other unusual metabolites including tolyporphin A **4** (a porphyrin-like compound with multi-drug resistance reversal properties)[9] and the ambiguenes, *e.g.* ambiguene F **5**, which are antifungal chlorinated alkaloids (Chart 1).[10]

*Chart 1*



1, homoanatoxin-a 2, tolypodiol A 3, comnostin B



4, tolyporphin A 5, ambiguene F

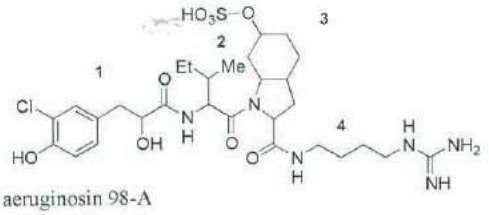
The major class of secondary metabolites produced by cyanobacteria is that of the oligopeptides, which are synthesised by non-ribosomal peptide synthetases.[11] These can be divided into six families depending on their structural characteristics,[12] namely the aeruginosins, the microginins, the anabaenopeptins, the cyanopeptolins, the microcystins, and the microviridins, as exemplified by metabolites **6-11** of Chart 2.

In the past, oligopeptides have been detected via enzyme-linked immunosorbent assays, enzyme inhibition assays, or according to their toxicity.[13] These assays have focused on obtaining quantitative data on the metabolites present, therefore the potential of these methods as discovery tools is limited. Analysis by high performance liquid chromatography (HPLC) is hindered by a lack of commercially available standards[14] so that time is wasted isolating known metabolites. Bioactivity-directed isolation has proved to be very effective in the past8 but again limits the researcher to detecting molecules possessing a certain activity. More powerful still is liquid chromatography-mass spectrometry (LC-MS). Here, one can separate the components in a complex mixture and obtain their relative molecular masses. This allows one to assess the potential novelty of a compound according to both mass and elution time prior to large-scale purification and characterization. Most LC-MS instruments allow for tandem MS that enables structural clues to be deduced and the identity of known molecules to be confirmed.[15] However, separation by HPLC involves costly and time consuming sample preparation and, due to long run times, high throughput can be cumbersome. Analysis of cyanobacterial extracts by matrix assisted laser desorption ionization-time of flight (MALDI-TOF) MS can provide comparable data to those from LC-MS but with far simpler sample preparation.

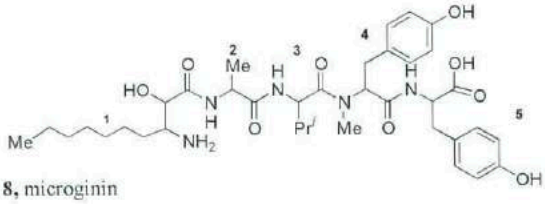
MALDI-TOF produces ions from laser irradiation of a sample co-crystallized with a matrix; the laser energy is absorbed and passed to the analyte molecules. This method of ionization predominantly produces singly protonated ions to *ca.* $m/z = 5000$, a range which encompasses the oligopeptides. Thus complex mixtures can be analyzed from a minute amount of sample without prior separation, and the relative molecular mass of each component present deduced from the protonated molecular ions.[16] Cyanobacterial extracts are assessed simply from mixing with the matrix, application of the mix to a target, and spectral recording.

The advantages described above make MALDI-TOF screening of cyanobacterial extracts particularly useful in the discovery of novel secondary metabolites. Due to the high sensitivity, low sample volumes, and speed of analysis, environmental samples can be assessed for the presence of novel compounds prior to culturing. Even single cyanobacterial colonies can be analysed by suspending them directly in matrix solution.[17] Novel compounds are easily detected using this method by comparing the component masses recorded with those in an appropriate database. If the mass spectrometer is also equipped for the analysis of post source decay (PSD) species, it is then analogous to LC-MS with tandem MS, and the presence of known compounds can be confirmed from the masses of the fragment ions produced.

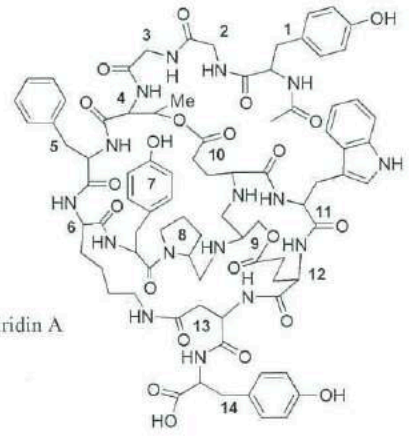
*Chart 2*



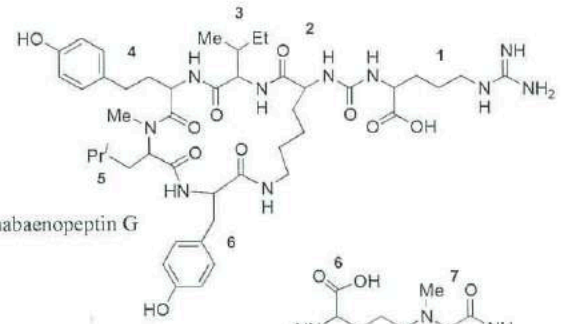
6, aeruginosin 98-A



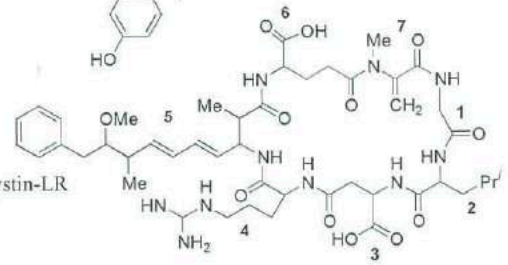
8, microginin



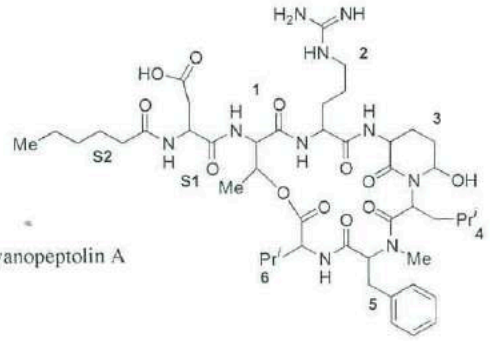
10, microviridin A



7, anabaenopeptin G



9, microcystin-LR



11, cyanopeptolin A

Since cyanobacterial oligopeptides are already well characterized, PSD allows for partial characterization of any novel compounds discovered. Often the subclass of oligopeptide present can be deduced by the presence of diagnostic fragment ions in the spectrum, e.g. the presence of a $m/z$ = 135 ion ($PhCH_2CHOMe^{+}$) is characteristic of 2*S*,3*S*,8*S*,9*S*-3-amino-9-methoxy-2,6,8-trimethyl-10-phenyldeca-4*E*,6*E*-dienoic acid (Adda, **12**), the unique amino acid found in microcystins.[16] The low mass daughter ions indicate the amino acids present in the molecule, while those at higher mass can indicate how the amino acids are joined together.

Oligopeptide characterization by MALDI-TOF MS has been undertaken successfully in Germany. Using the approach described above, von Dohren and co-workers were able to characterize a range of oligopeptides including aeruginosins, microginins, anabaenopeptins, and cyanopeptolins, whilst assessing the oligopeptide diversity of different cyanobacteria. They deduced structures for anabaenopeptin G, **7** (Chart 2) and anabaenopeptin 820 from analysis of the PSD fragments.[12,18,19]

The anabaenopeptins are cyclic peptides containing six amino acids. Each contains a D-lysine unit that has an ureido bond to a carbonyl that is linked to a side-chain amino acid. The D-lysine also forms a secondary peptide bond which encloses the ring. The CO-linked side-chain and the ring amino acids vary as does their degree of amino methylation.[11] There are 21 published structures of anabaenopeptins and these are listed in Table 1. The different compounds have varied biological activities including relaxation of norepinephrine-induced contraction,[20] protein phosphatase inhibition, and protease inhibition.[21-24] Like the German workers, we too have been able to deduce most of the structure of a new metabolite using MALDI-TOF MS, namely anabaenopeptin 906, **13**.

An environmental sample containing cyanobacteria was collected from a North Island lake. The MALDI-TOF MS (Fig. 1) showed the presence of several known compounds as well as an unknown metabolite with $m/z$ = 907. The PSD spectrum of this $m/z$ = 907 ion is shown as Fig. 2 and the loss of 200 Da is clear. This is diagnostic for an anabaenopeptin possessing an arginine side-chain. The low mass/charge species indicate the presence of arginine (Arg; $m/z$ = 70), lysine (Lys; $m/z$ = 70, 84), isoleucine (Ile; $m/z$ = 86), methylleucine (MeLeu; $m/z$ = 100), and methylhomotyrosine (MeHTyr; $m/z$ = 107, 164). Thus, five of the six amino acids present in the anabaenopeptins are identified, with the missing mass/charge entity correlating with that of a phenylalanine (Phe) residue. This also matches well with anabaenopeptin G, **7** in that its mass is only 2 Da higher than the new **13**; it corresponds to the loss of a hydroxyl group from the tyrosine in position 5, and an additional amino methylation on HTyr in position 3.

The higher mass fragments provide the sequence of the ring amino acids in **13**. Thus, the $m/z$ = 275 fragment shows that the Phe is joined to the MeLeu and the $m/z$ = 449 fragment shows that the Phe is also attached to the Lys, thus placing it in either position 3 or 6. The $m/z$ = 466 fragment can then be used to show that MeHTyr is attached to MeLeu as Phe is already attached to both Lys and MeLeu. This gives a final sequence of Ile-MeHTyr-MeLeu-Phe, and supports the presence of an Ile-MeHTyr fragment at $m/z$ = 305.

***Table 1.*** Amino acid sequence of the known Anabaenopeptinsa

| Compound | Mr (Da) | 1 | 3 | 4 | 5 | 6 | Ref. |
|---|---|---|---|---|---|---|---|
| Anabaenopeptin A | 843 | Tyr | Val | HTyr | MeAla | Phe | 20 |
| Anabaenopeptin B | 836 | Arg | Val | HTyr | MeAla | Phe | 20 |
| Anabaenopeptin C | 808 | Lys | Val | HTyr | MeAla | Phe | 25 |
| Anabaenopeptin D | 827 | Phe | Val | HTyr | MeAla | Phe | 25 |
| Anabaenopeptin E | 850 | Arg | Val | MeHTyr | MeAla | Phe | 26 |
| Anabaenopeptin F | 850 | Arg | Ile | HTyr | MeAla | Phe | 26 |
| Anabaenopeptin G | 908 | Arg | Ile | HTyr | MeLeu | Tyr | 18 |
| Anabaenopeptin G* | 929 | Tyr | Ile | HTyr | MeHTyr | Ile | 23 |
| Anabaenopeptin H | 922 | Arg | Ile | HTyr | MeTyr | Ile | 23 |
| Anabaenopeptin I | 759 | Ile | Val | HTyr | MeAla | Leu | 22 |
| Anabaenopeptin J | 793 | Ile | Val | HTyr | MeAla | Phe | 22 |
| Anabaenopeptin T | 865 | Ile | Val | HTyr | MeHTyr | Ile | 24 |
| Anabaenopeptin KT864 | 864 | HArg | Ile | HTyr | MeAla | Phe | 27 |
| Anabaenopeptin 820 | 820 | Arg | Val | HPhe | MeAla | Phe | 12 |
| Ferintoic Acid A | 866 | Trp | Val | HTyr | MeAla | Phe | 28 |
| Ferintoic Acid B | 880 | Trp | Ile | HTyr | MeAla | Phe | 28 |
| Nodulapeptin A | 929 | Ile | Met($O_2$) | HPhe | MeHTyr | Ser(Ac) | 29 |
| Nodulapeptin B | 913 | Ile | Met(O) | HPhe | MeHTyr | Ser(Ac) | 29 |
| Oscillamide B | 868 | Arg | Met | HTyr | MeAla | Phe | 21 |
| Oscillamide C | 956 | Arg | Ile | HTyr | MeHTyr | Phe | 21 |
| Oscillamide Y | 857 | Tyr | Ile | HTyr | MeAla | Phe | 21 |

[a]Numbering for anabaenopeptins amino acids is as for 7 of Chart 2;, D-Lys is omitted as it is always present in position 2 in the known anabaenopeptins.

***Table 2.*** Fragment ions of anabaenopeptin 906 observed by PSD.

| m/z | Sequence |
|---|---|
| 70 | Arg/Lys-related ion |
| 84 | Lys-Immonium ion |
| 86 | Ile-Immonium ion |
| 100 | MeLeu-Immonium ion |
| 107 | Tyr-side chain |
| 112 | Arg-Immonium ion |
| 129 | Arg-Immonium ion |
| 164 | MeHTyr-Immonium ion |
| 175 | Arg + 2H |
| 201 | CO + Arg |
| 275 | MeLeu + Phe + H |
| 305 | Ile + MeHTyr + H |
| 449 | Arg + CO + Lys + Phe - CO + 2H |
| 466 | MeHTyr + MeLeu + Phe + H |
| 579 | Ile + MeHTyr + MeLeu + Phe + H |
| 594 | Lys + Phe + MeLeu + MeHTyr + 2H |
| 603 | Arg + CO + Lys + Phe + MeLeu + H |
| 707 | Lys + Ile + MeHTyr + MeLeu + Phe +2H |
| 907 | M + H |

None of the fragments observed confirm the order in which the amino acids are present in the ring and whether Ile or Phe is located at position 3. The structure proposed as **13** has been constructed according to the sequences of presently known anabaenopeptins, where Phe is commonly seen at position 6 and an aromatic amino acid, such as HTyr, is always at position 4. This illustrates the limitation in characterizing secondary cyanobacterial metabolites by MALDI-TOF MS as, at times, the complete structure cannot be elucidated and stereochemistry can never be deduced. Ultimately, full characterization of these novel compounds requires purification and NMR spectroscopic investigations.

The screening of cyanobacterial extracts for oligopeptides by MALDI-TOF MS is a very powerful technique that can lead to the discovery of new compounds. It is simple, quick and inexpensive. Its use requires only a minute amount of sample that gives a rapid assessment of the presence or absence of novel metabolites.

## Acknowledgements

The contribution of Susanna Wood (Cawthron Institute, Nelson) is gratefully acknowledged.

## References

1. Schopf, W. J.; Packer, B. M. *Science* **1987**, *237*, 70-72.

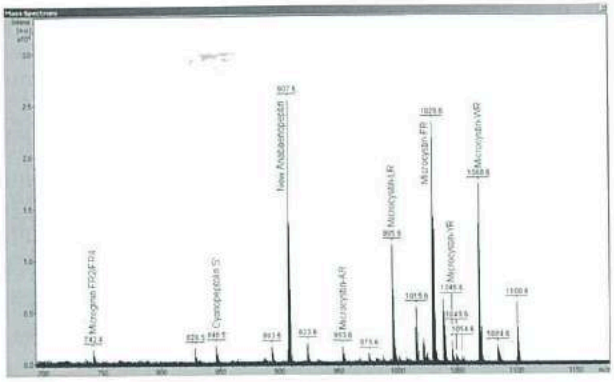


***Fig. 1.*** MALDI-TOF MS of a NZ lake sample.

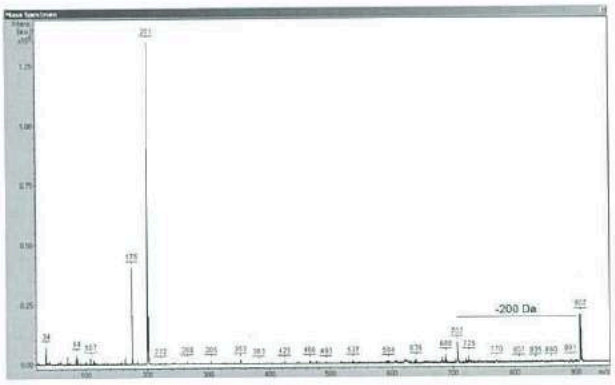



***Fig. 2.*** The MALDI-TOF PSD spectrum of $m/z$ = 907 from Fig. 1.


2. Whitton, B. A.; Potts, M. *The ecology of cyanobacteria, their diversity in time and space* Kluwer: Dordrecht, Netherlands, 2000.
3. Chorus, I.; Bartram, J. *Toxic cyanobacteria in water: a guide to their public health consequences, monitoring and management* Spon: London, 1999.
4. Jones, G. J.; Korth, W. *Water Sci. Technol.* **1995**, *31*, 145–151.
5. Codd, G. A.; Morrison, L. F.; Metcalf, J. S. *Toxicol. Appl. Pharmacol.* **2005**, *203*, 264-272; Azevedo, S. M.; Carmichael, W. W.; Jochimsen, E. M.; Rinehart, K. L.; *et al. Toxicology* **2002**, *181-182*, 441-446.
6. Wood, S. A.; Selwood, A. I.; Rueckert, A.; Holland, P. T.; *et al. Toxicon.* **2007**, *50*, 292-301.
7. Prinsep, M. R.; Thomson, R. A.; West, M. L.; Wylie, B. L. *J. Nat. Prod.* **1996**, *59*, 786-788.
8. Jaki, B.; Orjala, J.; Heilmann, J.; Linden, A.; *et al. J. Nat. Prod.* **2000**, *63*, 339-343.
9. Prinsep, M. R.; Caplan, F. R.; Moore, R. E.; Patterson, G. M.; Smith, C. D. *J. Am. Chem. Soc.* **1992**, *114*, 385-387.
10. Smitka, T. A.; Bonjouklian, R.; Doolin, L.; Jones, N. D.; *et al. J. Org. Chem.* **1992**, *57*, 857-861.
11. Welker, M.; von Dohren, H. *FEMS Microbiol. Rev.* **2006**, *30*, 530-563.
12. Welker, M.; Brunke, M.; Preussel, K.; Lippert, I.; von Dohren, H. *Microbiol.* **2004**, *150*, 1785-1796.
13. Xu, L. H.; Lam, P. K. S.; Chen, J. P.; Xu, J. M.; *et al. Chemosphere* **2000**, *41*, 53-58; Domingos, P.; Rubim, T. K.; Molica, R. J. R.; Azevedo, S. M. F. O.; Carmichael, W. W. *Environ Toxicol.* **1999**, *14*, 31-35.
14. Mountford, D. O.; Holland, P.; Sprosen, J. *Toxicon.* **2005**, *45*, 199-200.
15. Jemal, M. *Biomed. Chromatogr.* **2000**, *14*, 422-429.
16. Welker, M.; Fastner, J.; Erhard, M.; von Dohren H. *Environ. Toxicol.* **2002**, *17*, 367-374
17. Welker, M.; Marsalek, B.; Sejnohova, L.; von Dohren, H. *Peptides* **2006**, *27*, 2090-2103.
18. Erhard, M.; von Dohren, H.; Jungblut, P. *Rapid Commun. Mass Spectrom.* **1999**, *13*, 337-343.
19. Erhard, M.; von Dohren, H.; Jungblut, P. *Nat. Biotechnol.* **1997**, *15*, 906-909.
20. Harada, K.; Fujii, K.; Shimada, T.; Suzuki, M.; *et al. Tetrahedron Lett.* **1995**, *36*, 1511-1514.
21. Sanō, T.; Usui, T.; Ueda, K.; Osada, H.; Kaya, K. *J. Nat. Prod.* **2001**, *64*, 1052-1055.
22. Murakami, M.; Suzuki, S.; Itou, Y.; Kodani, S.; Ishida, K. *J. Nat. Prod.* **2000**, *63*, 1280-1282.
23. Itou, Y.; Suzuki, S.; Ishida, K.; Murakami, M. *Bioorg. Med. Chem. Lett.* **1999**, *9*, 1243-1246.
24. Kodani, S.; Suzuki, S.; Ishida, K.; Murakami, M. *FEMS Microbiol. Lett.* **1999**, *178*, 343-348.
25. Fujii, K.; Harada, K.; Suzuki, M. *Occurrence of novel cyclic peptides together with microcystins from toxic cyanobacteria,* Anabaena *species* In: *Harmful and Toxic Algal Blooms* Yasumoto, T.; Oshima, Y.; Fukuyo, Y. (Eds.) Intergovernmental Oceanographic Commission of UNESCO Paris, 1996, 559-562.
26. Shin, H. J.; Matsuda, H.; Murakami, M.; Yamaguchi, K. *J. Nat. Prod.* **1997**, *60*, 139-141.
27. Beresovsky, D.; Hadas, O.; Livne, A.; Sukenik, A.; *et al. Isr. J. Chem.* **2006**, *46*, 79–87.
28. Williams, D. E.; Craig, M.; Holmes, C. F. B; Andersen, R. J. *J. Nat. Prod.* **1996**, *59*, 570-575.
29. Fujii, K.; Sivonen, K.; Adachi, K.; Noguchi, K.; *et al. Tetrahedron Lett.* **1997**, *38*, 5525-5528.


*... continued from page 7*

Branch on *The Rise and Fall of IRL BioPharm: the good, the bad and the ugly - a personal view.*

### *Victoria University*

Recent visitors to the School have included alumnus Dr ***Paul Kilmartin*** (Auckland University) who spoke on *new applications for conducting polymers*, providing an overview of the recent work at the Polymer Electronics Research Centre at the University.

Dr ***Kathryn McGrath*** attended the 2007 International Soft Matter Conference (Aachen, Germany) in October. She gave an oral presentation *Dynamic Processes in Emulsion* and a poster *Cell Membrane Dynamics during Exocytosis in Gonadotrophs* based on the work of her former MSc student ***Pascale Savigny***.

***Emma Turner*** and ***Shivali Gulab*** successfully defended their PhD researches (*The Design and Synthesis of Hybrid Peloruside A-Laulimalide Analogues* and *An Aldol Approach Towards the Synthesis of Peloruside A and Analogues Thereof*) in the week of 8 October. Their studies were performed under the joint supervision of ***Joanne Harvey*** and ***Paul Teesdale-Spittle***; John Hoberg had supervised Shivali prior to his departure.

# The 2007 Nobel Prize in Chemistry

The Chemistry Laureate for 2007 was Gerhard Ertl, an Emeritus Professor and retired director of the Max-Planck Fritz Haber Institute in Berlin. The award recognises his successes in providing detailed descriptions of how chemical reactions take place on surfaces, studies that have laid the foundation of modern surface chemistry. He is awarded the prize for showing how reliable results can be obtained for such chemical processes.

Ertl, a German by birth, gained his PhD in physical chemistry in 1965 from the Technical University in Munich. In 1986 he succeeded Heinz Gerischer as director of the Department of Physical Chemistry of the Haber Institute and was appointed Scientific Fellow. His research focuses on structure and chemical reactions at solid surfaces. He has received more than 60 awards for his work, the latest being the Nobel Prize.

## Introduction

Despite the stereotypical image of the chemist holding a test tube in which a number of chemicals have been mixed to produce a new compound, we know that much more information is needed to understand how a chemical reaction actually occurs. The branch of chemistry concerned with reactions on solid surfaces – *surface chemistry* - demands advanced dust-free laboratories and sophisticated electronic instrumentation, coupled with advanced methodology and great precision. It is neither straightforward nor cheap! But surface reactions play such a vital role in both chemical industry and natural systems that they demand to be studied. Knowledge of surface chemistry can help explain such diverse processes as why iron rusts, how artificial fertilizers are produced, how the catalyst in a car's exhaust pipe works, and why chemical reactions on the surfaces of ice crystals in the stratosphere are causing the $O_3$ layer in the atmosphere to deteriorate. Knowledge about chemical reactions on surfaces helps to produce renewable fuels more efficiently and create new materials for electronics.

## Surface chemistry: a brief history

Chemical processes at surfaces and interfaces have a long history. One half of the 1912 Nobel Prize was awarded to P. Sabatier for *his method of hydrogenating organic compounds in the presence of finely disintegrated metals whereby the progress of organic chemistry has been greatly advanced in recent years*. It was later realized that the crucial molecular event is the adsorption of $H_2$ molecules on the metal surface, where they are dissociated into the constituent atoms. Refined, the method remains a standard procedure for hydrogenation of organic molecules. Heterogeneous catalysis was also central to the award of the Nobel Prize to Fritz Haber in 1918 *for the synthesis of ammonia from its elements*. Despite technical improvements, the same basic concept is used in today's process. In 1932 Irving Langmuir was awarded the prize for *discoveries and investigations in surface chemistry*, in which he made a range of seminal contributions relevant to both heterogeneous catalysis and to processes at the air-water interface.

After Langmuir, there was little progress in the study of chemical processes at surfaces because two major difficulties had to be overcome. Firstly, it was, and still is, notoriously difficult to prepare surfaces of controlled composition and morphology. Secondly, there were few experimental techniques that enabled the direct monitoring of molecular events at the surfaces. Instead, the researcher had to rely on measuring the chemical composition in the gas phase outside the surface. Inferences can be made about molecular surface events from such studies, but the information is uncertain. A transformation of the whole field was triggered by the emergence of semiconductor technology during the 1950s and 60s, when methods for handling surfaces under high vacuum conditions were developed. Furthermore, a number of new methods of studying surfaces under high vacuum conditions emerged. These developments led to the establishment of *surface science*, a research discipline that has attracted scientists with backgrounds in condensed matter physics, physical chemistry and chemical engineering. By the end of the 1960s a number of scientists had come to realize that useful tools for studying molecular processes at surfaces had become available. They hoped that these tools would continue to improve so as to enable really detailed chemical studies of reactions at surfaces to be undertaken.

Precisely because surfaces are so very chemically active, it is difficult to keep them clean enough to study a specific reaction - one of the reasons that precision combined with a high vacuum system is essential for success. In air, any surface is immediately covered by molecules from the gases present. Ertl displayed a unique understanding of how to make use of different experimental technologies, and he incorporated new technologies in his palette in order to produce as complete a picture as possible of the reaction under investigation. Apart from generating important knowledge about specific reactions, he constructed,

above all, a methodology that other researchers have been able to apply to completely different surface reactions. Initially, Ertl studied the behaviour of H2 on metal surfaces and his studies of fundamental molecular processes at the gas-solid interface were particularly thorough.

When a small molecule hits a solid surface from a gas phase there are two possible outcomes. The molecule may simply bounce back or it can be adsorbed. It is the latter case that raises the most interesting possibilities. The interaction with the atoms of the surface can be so strong that the molecule dissociates into its constituent groups or atoms. The molecule can also react directly with surface groups and change the chemical properties of the surface. A third possibility is that the adsorbed molecule encounters another previously adsorbed one and there is a binary chemical reaction on the surface.

Very important practical situations exist where these scenarios are the key chemical events; heterogeneous catalysis has been central to the chemical industry for more than a century. Since 1913, agriculture has been supplied with fertilizers rich in nitrogen, produced by the Haber-Bosch process in which $N_2$ gas is converted to $NH_3$ using an iron-based catalyst. These days, every car has a catalyst system that converts (toxic) CO and hydrocarbons to $CO_2$ in the exhaust gases; the catalyst also adsorbs the nitrous gases present reducing their quantity in the vehicle's emissions. Currently, large resources are devoted to the development of efficient fuel cells using $H_2$ as a standard vehicle fuel, where surface reactions between electrodes and $H_2$ are critical. Corrosion is caused by chemical reactions at surfaces; it is a major problem in everyday life and in sophisticated industrial contexts such as nuclear power plants and aircraft. Damage by corrosion may be reduced by adjusting the composition of the surface so that it is protected by an oxide layer formed in air. Thin semiconductor layers are produced by chemical vapor deposition in large quantities in the microelectronics industry. Chemical processes at surfaces are, therefore, central not only to a wide range of highly significant practical and economic applications of chemistry but also to the basic chemical research needed to unravel the details.

Our theoretical description of chemical reactions conceptually provides the simplest case for the formation of a molecule in the gas phase where the reacting species is affected only by encounter with its reaction partner. However, in most practical applications, reactions occur in more complex environments where the reacting species are constantly exchanging energy and momentum with other neighbouring molecules. For example, in a solution, the environment is disordered and dynamic and any description typically relies on considering the effect of the environment through its average properties. The gas-solid interface provides an example of an environment that is intermediate between the relative simplicity of the gas phase and the molecular complexity of the liquid phase. At the surface of a solid an adsorbed molecule can exchange energy and momentum with the surface material, but in the most ideal cases this support has long-range order. The consequence is that the interaction between molecule and support is much more regular, and this allows both more precise experiments and more detailed theoretical descriptions. Thus, the study of chemical reactions on surfaces provides one route towards a deeper understanding of reactions in condensed phases in general.

## Ertl's contributions to surface chemistry

Sabatier's work left a long-standing question of how $H_2$ is organized on metals like Pd, Pt and Ni. This question is relevant not only for understanding the hydrogenation of organic molecules, but also how hydrogen gas is used or produced at metal electrodes in many electrochemical processes. By combining experimental studies using low energy electron diffraction (LEED) with measurements of desorption, and also using modeling, Ertl was able to provide a quantitative description of how hydrogen is exposed on the metal surfaces.[1] This was highly relevant to the then current discussion of catalytic mechanisms. Ertl not only gave answers to a number of that had been posed for a long time, but also demonstrated how one could utilize the LEED method in combination with other experimental approaches. The most relevant chemical questions clearly needed more than one method. His approach to science is that when new opportunities appeared he revisits fundamental problems that he had analyzed previously. Thus, his latest publication on $H_2$ adsorption on a metal surface concerns the vibrational spectrum.[2]

The next long-standing and industrially important problem that Ertl attacked concerned the molecular mechanism of the catalytic formation of NH3 in the Haber-Bosch process (Eq. 1). Ertl's contribution was in providing detailed knowledge about how this process works. But above all, this study provides an example of systematic methodology applied to surface chemistry problems. In this way he has established an experimental school of thought for the entire discipline.

$$N_2 + 3H_2 \longrightarrow 2NH_3 \quad \text{..... (Eq. 1)}$$

In order to obtain a suitable thermodynamic driving force for the Haber-Bosch process (Eq. 1), industrially it is performed under high pressure. The commonly used catalyst consists of Fe particles with added KOH on a support of alumina and silica. Owing to its economic importance, numerous investigations had been made by the time Ertl initiated his studies in the mid-1970s. Although it was understood from kinetic studies that the rate-limiting step was the chemisorption of $N_2$, the underlying mechanism and the nature of the reactive species were unclear. Alternative mechanisms had been suggested, based on either atomic or molecular nitrogen, but it was impossible to discriminate between these on the basis of kinetic data alone. Equipped with the tools of surface science Ertl took the opportunity to investigate aspects of the reaction in model systems, however far from the realities of the Haber-Bosch process these seemed.

Ertl had previously studied $H_2$ on metal surfaces and it was straight-forward for him to show that, on the Fe of the Haber-Bosch process, the behavior of $N_2$ was qualitatively similar.[3] He measured the concentration of nitrogen atoms on the iron surface while simultaneously adding *hydrogen* to the system. He saw that as he added more $H_2$, the concentrations of N-atoms on the surface diminished.

Ertl concluded that nitrogen atoms on the surface disappear as they react with hydrogen molecules. This showed that the first step in the Haber-Bosch-reaction takes place between hydrogen molecules and nitrogen atoms. If the reaction had taken place between molecular hydrogen and molecular nitrogen, atomic nitrogen would still form on the surface, but it would remain unperturbed by the amount of hydrogen added.

In the then current literature, the most controversial issue was whether nitrogen would dissociate on the surface. The N-N triple bond is one of the strongest known and it appeared counterintuitive for interaction with the surface to be sufficiently strong to cleave $N_2$ into atoms. Ertl showed that atomic nitrogen was, in fact, present on clean iron surfaces,[4] and he deduced a detailed structural model for the iron-nitrogen structure on the surface.[5] Moreover, it was possible to characterize the kinetics of the nitrogen adsorption in detail.[6] The formation of atomic nitrogen occurs with a low activation energy but with a very small pre-exponential factor making the process slow. Ertl also discovered that although the activation energy was different for different crystal planes, the reaction proceeds on all of the three major crystal planes, (111), (110) and (100). Furthermore, the energy barrier increases with increasing surface coverage so that the kinetic difference between the crystal planes decreases.

Initially it was far from obvious that these model studies applied to the molecular events in the industrial Haber-Bosch process. To demonstrate the applicability, Ertl and Thiele[7] analyzed the surface composition of a commercial catalyst using Auger Electron Spectroscopy (AES). They found that under ambient conditions the surface had a complex composition but, under the reducing conditions of the process, iron and potassium dominate at the surface. Through a characterization of adsorption energies, it was concluded that it is only the adsorbed atomic nitrogen that remains on the surface when the reaction chamber is emptied after a catalytic cycle at high pressures. By using AES to analyze how surface nitrogen coverage varied with $H_2$ pressure during the reaction, the high-pressure data were shown consistent with those for model measurements at low pressures. Furthermore, there was consistency between the observed rates of the elementary processes and the macroscopically measured kinetics. These studies, bridging what is called the *pressure gap*, were crucial in gaining acceptance of the *surface science approach* to catalysis by a community struggling with the realities of industrial processes involving heterogeneous catalysis.

*Scheme 1*

$$H_2 \rightleftharpoons 2H_{ad}$$

$$N_2 \rightleftharpoons N_{2,ad} \rightleftharpoons 2N_s$$

$$N_s + H_{ad} \rightleftharpoons NH_{ad}$$

$$NH_{ad} + H_{ad} \rightleftharpoons NH_{2,ad}$$

$$NH_{2,ad} + H_{ad} \rightleftharpoons NH_{3,ad}$$

$$NH_{3,ad} \rightleftharpoons NH_3$$

Having identified that the dissociation of $N_2$ into atoms was slow, and having demonstrated that the model systems were relevant for to the Haber-Bosch process, it was comparatively easy to show that the mechanism was that of Scheme 1. Although this had been suggested previously, Ertl not only confirmed its correctness but also gleaned the energetic details of the individual steps, the later ones starting from $NH_3$ and monitoring the steps backwards (which is favoured at low pressures). Adsorption of $NH_3$ on Fe involves an energy gain of < 75 kJ/mol, small enough to ensure complete desorption at typical process conditions ($T \geq 400$ °C). According to Scheme 1, the adsorbed $NH_3$ can dissociate on the surface. The presence of $NH_2$ could not be quantified spectroscopically but, by co-adsorbing $NH_3$ and $D_2$, Ertl was able to infer the dissociation and recombination rates for the reaction:

$$NH_{3,ad} \rightleftharpoons NH_{2,ad} + H_{ad}/D_{ad}$$

NH is present in quantities large enough for observation using methods like ultraviolet photoelectron spectroscopy, secondary ion mass spectroscopy, and high resolution electron energy loss spectroscopy.[8] From these measurements, it then became possible to formulate the mechanism of Scheme 1 in energy terms.[9]

Despite this success, one essential feature of the industrial process remained to be explained. Empirically, it had been found that the presence of $K^+$ ions in the catalyst improved the rate of the catalytic cycle. Ertl had found that the potassium remained on the surface of the catalyst under process conditions. Since $N_2$ cleavage is rate-limiting, the potassium must influence this reaction step. It was then found that in the presence of potassium ions $N_2$ is adsorbed more readily on the surface and the adsorption energy increases by 10-15 kJ/mol; this is attributable to potassium donating electrons to neighbouring Fe atoms.[10]

Ertl's investigations serve as a model of how sophisticated experimental methods can be used to study a phenomenon of the utmost practical relevance. He began by identifying the crucial features of the reaction in the industrial context, demonstrated the relevance of model studies, and then identified a number of elementary steps that became the targets of focused studies. The steps were characterized from structural, energetic, and kinetic points of view using state-of-the-art methodology that involved the use of many different techniques with highly sophisticated equipment. For each question there is, at any given point in time, an optimal method. It is clear that, throughout his career Ertl's ambition has been to use that method.

Ertl not only clarified the molecular events of the Haber-Bosch process, but he also demonstrated what it takes to unravel mechanisms of catalytic processes in general. This has had a lasting influence on the field of heterogeneous catalysis.

In the Haber-Bosch process, the observed macroscopic kinetics of $NH_3$ production are related to the kinetics of the individual steps of the reaction observed under idealized conditions. For some heterogeneously catalyzed reactions it had been found earlier that the macroscopic kinetics indicated an oscillatory rate, a clear sign of non-linear dynamic behaviour. Challenged by such observations, Ertl also made an in-depth study of another *classical* catalytic reaction - the oxidation of CO by $O_2$ on Pt. This reaction is

important to the catalytic converter in a car's exhaust system. The crucial questions *What is the mechanism behind the non-linear kinetics?* and *What other phenomena can be inferred in addition to the kinetic oscillations?* led to this reaction illustrating a range of phenomena typical of non-linear kinetic reactions. Ertl showed that the rates of different steps in the reaction vary over time. Some steps oscillate between different rates, and the reaction proceeds differently depending on the coverage of the platinum surface. Sometimes these variations lead to a chaotic course of events so that the reaction is not reversible and, as a consequence, becomes much more difficult to study than the Haber-Bosch process.

A series of imaginative studies[11] led Ertl to the microscopic causes of the observed non-linear behaviour. Again, he demonstrated how the full spectrum of surface physics/chemistry methods can be combined to yield a comprehensive understanding of important and complex catalytic processes. High pressure *in situ* methods, FTIR, and X-ray diffraction gave information on the state of the catalyst itself. These methods are generally much less precise than high vacuum techniques, but they gave invaluable corroborating information and helped close the pressure gap. In the study of sensitive oscillatory reactions on surfaces, the energy input must be controlled and minimized, and this is an added constraint. Thus, the use of AES, although powerful for the Haber-Bosch studies, is not feasible. Instead, low energy methods such as LEED were employed to directly monitor structural changes, and photoemission electron microscopy (PEEM) to monitor the local work function with high spatial resolution. These studies enabled Ertl to demonstrate that his methodology applies not only to systems where the kinetics are dominated by a single rate-limiting step, as for the Haber-Bosch process, but also to systems where non-linear dynamics prevail.

### Ertl's lasting contribution to the understanding of surface chemistry

The 2007 Laureate, Gerhard Ertl, was one of the first to understand the potential of the new technology and he laid the methodological foundations for an entire field of research. The great reliability of Ertl's results can be attributed to his meticulous precision combined with an outstanding capacity to refine problems. He painstakingly and systematically searched for the best experimental techniques to investigate each separate question.

His methodology sets a standard for how chemical processes on surfaces can be studied and elucidated.

### References


1. Conrad, H.; Ertl, G.; Latta, E.E. *Surface Sci.* **1974**, *41*, 435; Christmann, K.; Schober, O.; Ertl, G.; Neumann, M. *J. Chem. Phys.* **1974**, *60*, 4528; Christmann K.; Ertl, G.; Pignet, T. *Surface Sci.* **1976**, *54*, 365; Christmann, K.; Behm, R. J.; Ertl, G.; Van Hove, M.A.; Weinberg, W. H. *J. Chem. Phys.* **1979**, *70*, 4168.
2. Badescu, S.C.; Salo, P.; Ala-Nissila, T.; Ying, S. C.; *et al. Phys. Rev. Lett.* **2002**, *88*, 136101; Badescu, S.C.; Jacobi, K.; Wang, Y.; Bedürftig, K.; *et al. Phys. Rev. B* **2003**, *68*, 205401.
3. Bozso, F.; Ertl, G.; Grunze, M.; Weiss, M. *Appl. Surface Sci.* **1977**, *1*, 103.
4. Bozso, F.; Ertl, G.; Grunze., M. *J. Catalysis* **1977**, *49*, 18; Ertl, G.; Huber, M.; Lee, S.B.; Paál, Z.; Weiss, M. *Appl. Surface Sci.* **1981**, *8*, 373.
5. Imbihl, R.; Behm, R.J.; Ertl, G.; Moritz, W. *Surface Sci.* **1982**, *123*, 129.
6. Ertl, G.; Lee, S.B.; Weiss, M. *Surface Sci.* **1982**, *114*, 515.
7. Ertl, G.; Thiele, N. *Appl. Surface Sci.* **1979**, *3*, 99.
8. Weiss, M.; Ertl, G.; Nitschke, F. *Appl. Surface Sci.* **1979**, *2*, 614; Drechsler, M.; Hoinkes, H.; Kaarmann, H.; Wilsch, H.; Ertl, G. *Appl. Surface Sci.* **1979**, *3*, 217.
9. Ertl, G. *J. Vac. Sci. Tech.* **1983**, *A1*, 1247.
10. Ertl, G.; Weiss, M.; Lee, S.B. *Chem. Phys. Lett.* **1979**, *60*, 391.
11. Behm, R.J.; Thiel, P.A.; Norton, P.R.; Ertl, G. *J. Chem. Phys.* **1983**, *78*, 7437; Cox, M.P.; Ertl, G.; Imbihl, R. *Phys. Rev. Lett.* **1985**, *54*, 1725; Imbihl, R.; Cox, M.P.; Ertl, G.; Mueller, H.; Brenig, W. *J. Chem. Phys.* **1985**, *83*, 1578; Imbihl, R.; Cox, M.P.; Ertl, G. *J. Chem. Phys.* **1986**, *84*, 3519; Eiswirth, M.; Moller, P.; Wetzl, K.; Imbihl, R.; Ertl, G. *J. Chem. Phys.* **1989**, *90*, 510; Jakubith, S.; Rotermund, H.H.; Engel, W.; von Oertzen, A.; Ertl, G. *Phys. Rev. Lett.* **1990**, *65*, 3013; Kim, M.; Bertram, M.; Pollmann, M.; von Oertzen, A.; *et al. Science* **2001**, *292*, 1357; Beta, C.; Moula, M.G.; Mikhailov, A.S.; Rotermund, H.H.; Ertl, G. *Phys. Rev. Lett.* **2004**, *93*, 188302.


Compiled by Brian Halton and Peter Hodder from material freely available from the Nobel Foundation. Further details may be obtained from: http://nobelprize.org

## Chemistry Behind the News

### Drugs and Toys

Two children being admitted to hospital caused a popular children's toy to be pulled from shop shelves. They became ill from swallowing beads that were part of the toy.

The toy is a craft kit made up of multicoloured beads that when sprayed with water, stick to each other so they can be used to make pictures and other items.

1,5-pentanediol is used in the toys' manufacture, but it appeared this had been substituted with 1,4-butanediol.

In the liver, 1,4-butanediol ($C_4H_{10}O_2$) is broken down by alcohol deyhydrogenase and aldehyde dehydrogenase into metabolic products including gamma-hydroxybutyrate (GHB). GHB is abused as a recreational drug. It is also found naturally in the brain where it is thought to be a neuromodulator. GHB also seems to affect dopamine levels in the brain.

1,4-butanediol is mostly used in the manufacture of polyurethanes such as surface coatings, foam and adhesives.

The chemical that was meant to be used in manufacture was pentamethylene glycol or 1.5-pentanediol ($OHCH_2(CH_2)_3CH_2OH$). It is a water miscible liquid that is used as a hydraulic fluid as well as in the manufacture of polyester and polyurethane resins.

# Significant Changes To The US Patent System

By Blair Hesp and Jarrod Ward

***STOP PRESS: The USPTO rule changes discussed below were intended to come into force on 1 November 2007. However, they are now the subject of an 11th hour interim injunction against the USPTO granted in a US District Court on 31 October 2007. Although the new rules are not currently in force, the intention of the USPTO is to apply these rules retrospectively to any application under examination if, and when, the injunction is lifted. Therefore, new applicants should be aware of the following issues when planning a US patent strategy.***

The US is the home of the world's largest economy, and many inventors consider the grant of a US patent to be a significant milestone on the road to protecting and commercialising an invention. Consequently, the large number of patent applications filed by applicants from the US, and other countries, has created a heavy examination workload for the United States Patent and Trademark Office (USPTO). In response, the USPTO has attempted to introduce several new rules in order to accelerate patent examination.

### *Number of Claims*

Firstly, the USPTO prefers new inventions to be encompassed within a single, succinct patent application comprising a maximum of 25 claims. If this limit is exceeded, the USPTO would then require the applicant to restrict the number of claims during examination, or to justify the number of claims in an "examination support document".

In addition, the USPTO has proposed further rule changes in an attempt to stop applicants circumventing the new claim limit by filing several applications directed to different embodiments of the same general invention. In particular, the USPTO will seek details of any related US application if the related application has a common inventor, a common owner, and was filed within two months of the new application.

Furthermore, if the disclosure of a new application is substantially the same as any other US patent application or granted US patent, then the USPTO will reject the new application on the presumption that the claims of each application are not distinct. However, this presumption would be able to be rebutted by demonstrating that the claims of the new application are patentably distinct when compared to the claims of the earlier application.

### *Continuation Applications*

It is common practice in the US, especially in the pharmaceutical field, to file "continuation applications" before a parent application is granted. These applications have the same invention description and priority date as the parent application, but different claims. For example, a continuation application may be directed to a preferred embodiment of an invention which was described, but not claimed, in the original application.

In a further attempt to reduce the number of applications awaiting examination, the USPTO has attempted to restrict the number of continuation applications filed from any parent application, or family of applications, to no more than two. However, this limit would not include applications which have been divided out from a parent application in response to a unity of invention objection (i.e. when an application is deemed to be directed to more than one invention).

The USPTO believes that these rule changes will reduce examination times, while promoting innovation and improving patent quality. While it is yet to be seen how effective these measures will be if, and when, they are implemented, it is clear that any US patent applicant must now operate with a heightened level of caution. In particular, contingencies for all of the above-mentioned factors should be incorporated into any US patent strategy.

A reminder: if you have any queries regarding patents, or indeed any form of intellectual property, please direct them to:

Patent Proze
Baldwins
PO Box 852, Wellington

Email: email@baldwins.com

Blair Hesp and Jarrod Ward of Baldwins specialise in chemistry and biotechnology patents. Blair joined Baldwins in 2006, and has a PhD in pharmacology from the University of Otago as well as a NZDipBus with a management focus. Jarrod joined Baldwins in 2007, and has submitted a PhD thesis in chemistry to the University of Auckland. Blair and Jarrod are currently studying towards law degrees and registration as patent attorneys.

Comment

# Comment on Climate Change Mythconceptions

Vincent R. Gray

75 Silverstream Rd, Crofton Downs, Wellington (e-mail: *vinmary.gray@paradise.net.nz*)

The article by D. S. Mackie and K. A. Hunter in the last issue of this *Journal*[1] revived the traditional game of Aunt Sally as a means of scientific discussion. The authors assembled what they claimed to represent as *Common Arguments* supposedly made by *Climate Change Deniers*. No actual *denier* was identified, and the replies did not display a very deep knowledge of Climate Science. Some of those *Common Arguments* are correct whatever the authors may say.

**Common Argument 5:** ***Temperature measurements by satellite and radiosonde balloons show no significant warming***

There was no significant warming shown by both of these measurements from 1979 to 1997 and from 2001 to 2007 when both periods experienced large greenhouse gas increases. This author[2] has refuted completely the claim that *new datasets have been developed that do not show such discrepancies (between surface and lower atmosphere records)* quoted from the web-published CSSP Report. It might be remarked that the latter period, 2001-2007, also shows no significant warming in the surface record either.

**Common Argument 6:** ***Computer models of the climate are worthless***

No computer model has ever been *validated* as this needs a rigorous process to be carried out by computer engineers and requires successful future prediction. The IPCC know this is true since they only claim that the models are *projections*. The IPCC uses the *gut feeling* of self-styled *experts* as a substitute for validation. Their levels of *likelihood* with their spurious *probabilities* have no scientific basis. No model has ever been shown to be successful in future prediction.

The obvious example is the fact that the *globe* has not been *warming* for the past eight years, however you measure it. This proves without any doubt that the models are worthless.

**Arguments not Answered**

***1. The data and procedures used to provide evidence for* global warming *are scientifically and mathematically unsound***

The *Mean Annual Global Temperature Record*, which figures so largely in IPCC thinking, has been obtained without using any actual measured average temperature from any place on the earth's surface. So in order to provide a long sequence for their record, these authors were stuck with continual use of the procedure adopted by the first meteorological stations in 1850. This involves a once daily measurement from a maximum and minimum temperature thermometer. The mean of these two is taken to be a *daily average*, but, as any statistician will tell you, this is not true. The bias (usually positive) is variable, impossible to estimate, and easily can exceed the supposed *global warming* of the past century.

The samples are grossly unrepresentative of the earth's surface - it is like judging the results of an election from polling only one suburb of one city. The sites chosen are mainly near towns and subject to urban heating.

Attempts to *correct* for some of these errors have shown that there has been no *global warming* in the USA or China for the past 100 years. Many better kept local long-term records confirm this, including those for Christchurch from 1900 to the present. Comparisons of *proxy* temperature data from the past with the recent human-influenced surface record merely emphasize the recent influence of urban heating.

***2. Atmospheric carbon dioxide concentration is not* well-mixed *but variable***

Measurement of [$CO_2$] in the atmosphere is manipulated by the IPCC in an attempt to conceal its variability. Some 90,000 previous measurements[3] have been suppressed and unwanted figures are not published by the proponents. The reason for this is that the formula for calculating radiative effect of the gas is logarithmic, so it gives a higher figure when applied to *constant* [$CO_2$] than for a variable sample.

***3. Levels of uncertainty***

Uncertainty levels for climate quantities are often unknown and, even when these are supplied, they are questionable. Much of the Mackie-Hunter article uses line graphs of highly uncertain quantities to make a number of points. The uncertainty levels of these plots and of their Table 1 are not even mentioned.

The question as to whether ancient atmospheric [$CO_2$], measured with huge uncertainty in only one place, increased before or after a rise in temperature is the subject of much controversy and this cannot be decided from such uncertain data.

**Conclusion**

Scientists do not go in for consensus, but for facts and evidence. I would encourage the readers to make up their own minds, not just by trying to find the *flaws* in only one side of the argument, as a game of Aunt Sally, but by an impartial assessment of all the evidence.

**References**

1. Mackie, D.S.; Hunter, K. A. *Chem. in NZ* **2007**, *71*, 72-78
2. Gray, V R. *Energy Environ.* **2006**, *17*, 707-714.
3. Beck, E-G. *Energy Environ.* **2007**, *18*, 239-281.

# New Way to Test Sulfur

AgResearch has developed a new sulfur soil test.

Dr Gordon Rajendram developed the test as part of a PhD project. The new test makes scientific analysis easier and gives farmers accurate information to decide how much sulfur fertiliser their soil needs.

Sulfur is very important for good plant growth because it is used in the production of amino acids that make up plant proteins. A sulfur deficiency gives plants yellow leaves and they grow poorly.

Currently there are two soil tests that measure easily extractable organic sulfur and sulfate-S ($SO_4$). These tests are measured on two different instruments and then easily extractable organic sulfur is calculated using a difference technique.

The new test is easier to measure using a single instrument and gives more reliable results. Hill Laboratories is carrying out the new test.

# Response

Doug S. Mackie and Keith A. Hunter

Department of Chemistry, University of Otago, PO Box 56, Dunedin (e-mail: *dmackie@chemistry.otago.ac.nz*)

When informed that a scientific objection had been made to our article and asked if we wished to respond we were naturally keen to engage in meaningful debate and we are grateful to the editor for this opportunity. However, having now read the criticisms, we feel they are so lacking in clarity and substance that it would be uncharitable to respond in detail. Instead, we have confined ourselves to a few observations of points that puzzled us and will doubtless puzzle readers.

The common arguments we cite came from a list entitled *Ten facts about global warming THEY don't want you to know*.[1] For several months a link to these *Ten facts* appeared at the top of the must read list at the website of the New Zealand Climate Science Coalition (NZCSC) – a group of which Gray is a founding and current member. After we pointed out some errors in this list of arguments to NZCSC, the list was altered in accordance with the suggestions of the NZCSC science advisor – in our view giving the list a stamp of approval (and demonstrable input) by NZCSC. The NZCSC amendments were cosmetic and in no way altered the substance of the claims. We will be glad to provide copies of the list and relevant correspondence should anyone want this from us. We will also be glad to provide copies of press releases and popular articles written by NZCSC members that were, until recently, hosted by their website but which were removed only days after we began to critically examine them – as was the link to the *10 facts* list.

Gray makes reference to articles he has produced for *Energy and Environment* and we must confess that we have not read his oeuvre there. *Energy and Environment* is a journal not listed by Thompson-ISI and cannot, therefore, be assessed in terms of its quality. It is interesting, however, that only critics of mainstream climate science seem to publish in it, which speaks volumes for its reputation amongst the majority of climate scientists. The commentary by *Environmental Science and Technology*[2] about *Energy and Environment* is especially informative.

We are puzzled that Gray can assert that ground temperature measurements are invariably biased and cannot be relied upon, but only a few lines later uses precisely the same type of data for Christchurch to *show* that no global warming has occurred.

It is very sad to see that Gray reverts to the classic *conspiracy theory* concerning suppression of evidence by the IPCC. It is ludicrous to suggest that the many hundreds of respectable climate scientists around the world involved with IPCC would stoop to this, and much less credible that such a conspiracy could actually be organized and kept secret from all but a few. Gray references to the work of E.G Beck, also published in *Energy and Environment*. This article presents a graph summarizing historical $CO_2$ measurements (not considered reliable by IPCC scientists) which purports to show, *inter alia*, that the concentration of $CO_2$ in the atmosphere changed from 310 ppm in 1936 to 472 ppm in 1943, and then back down to 320 ppm in 1950. This represents a net change, in both directions of ~5.1 x $10^{16}$ g C per year, approximately 15 times the fossil fuel emissions for 2004 (7.4 x $10^{15}$ g C). The global carbon cycle simply does not, and indeed cannot, work like this. To believe that it can, shows a willful disregard for the basic principles of chemistry, physics and biology, and we doubt that the debate will be furthered by reference to such egregiously flawed work.

We conclude by stating what is obvious to us as scientists. Human activities such as agriculture, deforestation, and fossil fuel combustion have seriously disturbed the atmospheric inventories of greenhouse gases, particularly $CO_2$, over the last 250 years. Atmospheric $CO_2$ is intimately linked to the regulation of the Earth's climate, and this has been so for millions of years. Therefore, we should be worried about the future. The almost unilateral action of world governments to address this problem has not been undertaken lightly, and has come about because of an enormous body of scientific evidence and opinion. To suggest that the latter is nothing but a falsehood borne out of conspiracy belongs only in *Alice in Wonderland*.

**References**

1. See: http://www.numberwatch.co.uk/ten_facts_about_global_warming.htm
2. See: http://pubs.acs.org/subscribe/journals/esthag-w/2005/aug/policy/pt_skeptics.html .

The websites were checked at the time of going to press (October 18, 2007).

# Finding Answers in Food

Dr Lai Yeap Foo from Industrial Research Limited in Wellington received an award from the cranberry growers' co-operative, Ocean Spray for his contribution to international molecular nutrition and food science.

He found what mechanism was responsible for the cranberries' ability to inhibit urinary tract infections. Since his research the cranberry product business has grown 150%.

Cranberries are one of only three species of fruit native to North America and New Zealand is now growing its own cranberries on the West Coast of the South Island.

Cranberries are one of the top ten selling botanicals in the USA today but indigenous North Americans also used them medicinally.

Research is continuing to be done in the area of cranberries and urinary tract infections. In April 2007, the positive results of a pilot study on concentrated cranberry extract preventing recurrent urinary tract infections in women was published in the journal, Phytomedicine.

Dr Lai has also developed an anti-inflammatory product from passionfruit skins that can lower blood pressure for patients suffering from hypertension.

In the future he is hoping to further develop a grapefruit skin wound healing product.

# Letter to the Editor

**Re: *Climate change mythconceptions*** (*This Journal*, **2007**, *71*, 72-78)

Further to this article and those global processes that result in temperature change and [$CO_2$] in the atmosphere –what I call *the mechanisms* involved. I think the issue of global temperature increase is unquestioned. What is at issue is the way the [$CO_2$] is increasing and I suggest that the following ***policy brief*** summarises the issues key to any discussion.

**There are two crucial *facts* of science:**

1. Human activity has increased and intensified global $CO_2$ production.
2. Global systems adjust slowly to *excess* $CO_2$ and human activity has eroded some of the absorption capacity.

**Other scientific issues:**

3. Baseline for $CO_2$ data is with no human activity; this is not realistic.
4. Global systems will adapt to increased $CO_2$, but it will take centuries.
5. Humanity will adapt to global systems and produce less $CO_2$, but overall levels will be higher than the current baseline being used which is in absence of human activity.
6. $CO_2$ will increase as ppm in atmosphere over next 50 years (minimum).
7. Global temperature/climate change will increase/continue over next century (minimum).
8. Climate events will become more erratic and severe.

**Key stuff is political:**

9. There is no global consensus on action.
10. Global consensus is unlikely in foreseeable future.
11. Effective restriction of $CO_2$ is likely 10 to 15 years away.
12. There will be large costs geographically, socially, and economically.

**Global conclusion:**

13. Global warming is the perfect hammer needed to forge global social/political consensus. If so, costs are worth it in relation to long term social/political benefits to humanity, and the loss of life and costs likely to be much less than many of the other alternatives for forging the consensus.

**New Zealand conclusion:**

14. NZ is not able to lead global consensus without serious adverse impact and economic risk.
15. NZ needs take prudent steps manageable within current economic circumstances.
16. NZ forward planning needs bring to account the implications of this policy brief.
17. NZ should be a *fast follower*.

Graham R. Little (*grl@xtra.co.nz*)

# Dates of Note

This year marks the 100th anniversary of the α-particle scattering experiment devised by Ernest ***Rutherford*** with Hans ***Geiger*** and E. ***Marsden*** that led to the discovery of the atomic nucleus in 1911.

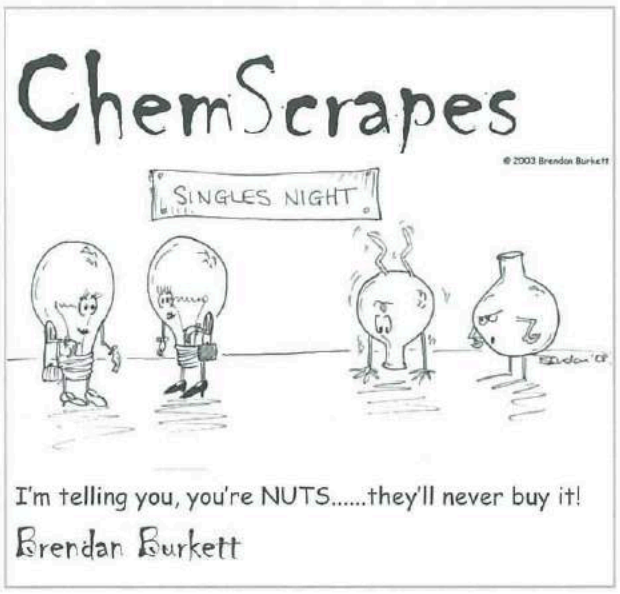
February 1 is the 35th anniversary of ***Hewlett-Packard***'s introduction of the hand-held calculator. The HP 35 retailed for $US395.

On February 29, 1936, Nature carried Niels ***Bohr***'s Neutron capture and nuclear constitution often referred to as the bowl of balls explanation for the effect of bombarding particles on a nucleus (*Nature*, **1936**, *137*, 344-348).

March 2 is the 125th anniversary of the birth of Walter Norman ***Haworth***, recognised for his projection formulae, while the 5th marks the 200th anniversary of the birth of Petrus Jocobus ***Kipp***, inventor of the apparatus for hydrogen sulfide generation named after him.

Some 340 years ago (March 10, 1668) German-Dutch chemist Johann Rudolf Glauber, the German father of chemistry, died. He prepared HCl from NaCl and $H_2SO_4$ and pointed out the virtues of the residue, $Na_2SO_4$ - *sal mirabile*, better known as Glauber's salt.

150 years ago, on March 30, the first US patent for a combination lead pencil and eraser was issued to Hyman L. ***Lipman*** (No. 19,783) while on 6 April 1938, Roy J. ***Plunkett*** discovered **Teflon** at DuPont.

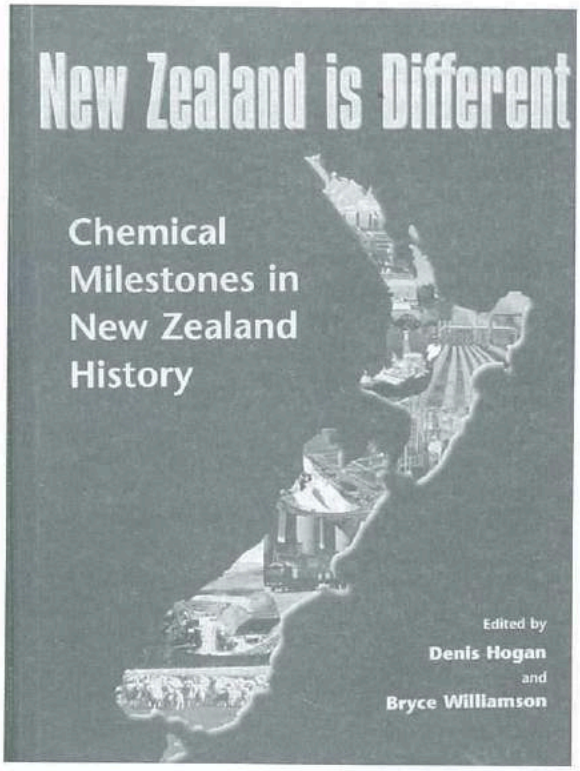


# New Zealand is Different: Chemical Milestones in New Zealand History

This NZIC publication, edited by the late Denis Hogan and Bryce Williamson was published by Clerestoy Press, (Christchurch) in 1999. It was subsequently distributed free to every secondary school in NZ through funding from the Stout Trust and is now out of print. Council is keen to gauge interest in a possible reprint. If you are interested in purchasing a copy please e-mail us at: *NZIC.office@nzic.org.nz* The decision to reprint will be made in conjunction with the publishers and will depend upon a number of factors.

Council is also convinced that there are number of stories akin to those in this volume that are still to be brought to the attention of people with an interest in things chemical and also to the general public. To this end **New Zealand is Different Vol. 2** is being considered. If you have ideas of suitable stories, know of potential authors for a suggested essay, or are prepared to write an essay yourself please e-mail the NZIC office: *NZIC.office@nzic.org.nz*

We reprint below an abridged and edited version of the 2000 review (Kennedy, M. *This Journal*, **2000**, *64*, 15-16).




The NZIC is to be congratulated in commissioning this splendid chemical history. It contains elements of social history and political and commercial skullduggery, twists and turns typical of the best detective stories, all interwoven with true-life tales about some of NZs outstanding chemists - and written in a style that can easily be understood by the lay as well as the scientific reader. For the 26 essays in *New Zealand is Different* are written by people who not only understand their research (and were often at the centre of the work they describe) but who know how to communicate that work with great clarity and flair.

The opening essay points out those NZs chemical problems, originating as they do from the country's unique geology and geography are unlike most. Politicians and administrators, dubious of home-grown expertise, demanded reassurance from overseas experts who too often come, give advice, take their fee and depart. The NZ chemists and chemical engineers have been left to discover and solve the real problems.

In the early days, most NZ research was done in Government institutions like the DSIR. Hogan traces the evolution of such institutions from the days of the Colonial Museum through to the creation of the CRIs in 1991. Since then, and perhaps because of the market-driven forces, it has become fashionable to disparage the achievements of the DSIR. He suggests it is too soon to tell whether the new system will produce better science for NZ. However, one doubts whether the achievements recorded herein would be possible under the present system of funding. Essayists Earle, Davey and Wright benefited from Sir Ernest Marsden (Head, DSIR) establishing the Defence Scientific Corps by gaining research overseas experience via a doctoral degree before returning to NZ.

Each essay is appropriately referenced and contains enough information to whet the appetite. Abstruse technical detail is minimal and limited to *information boxes* that can be omitted on first reading. The essays stand alone but have common themes and cross-linkages. Walker [known to many readers as theaffable vegetable gardener on the (earlier) Maggie Barry show] gives an erudite account of the impact of rye-grass and clover-based pastures on the economy, and the importance of superphosphate as a fertilizer. Superphosphate production, the first major chemical process industry, had handling difficulties with the raw materials as described by Higgins.

Wright traces 50 years of research carried out on climate controlled facial eczema. Clare relates bush-sickness/North Island soil cobalt deficiency to effects in grazing animals. The solution to this problem more than paid for all the agricultural research up to that time. Letham's account of cytokinins and their influence on plant development typifies the ability of NZ scientists to carry out high-quality research under poor conditions with limited resources. Clinton describes work on developing atomic absorption spectroscopy so necessary in these studies.

The Wright, Clare and Letham essays are fascinating tales of detection. So too is Walker's account of what caused NZ wool cargoes to ignite spontaneously during shipping. It provides an example of good chemistry underpinning an important industry. Fenby tells how NZ developments of cyanidation led to resurgence in the gold industry; while Kennerley shows how chemistry made important contributions to the science of concrete-making. Davey describes the chemists and biochemists of the MIRNZ efforts to establish why Canterbury lamb was tough when it reached the UK; Robertson introduces us to the cheese making system developed by the NZDRI that has mechanized the making of world cheeses; Foster *et al.*, trace the rise and fall of the NZ ceramic industry.

Difficulties in chemistry commercialization are evidenced in Leary's essay on the work of DSIR Chemistry Division and Ian Miller's story of seaweed industry and, also in Cambie's NZ plant natural products account. Stonyer gives a tragi-comedy on attempts to make terpineol economically from by-product turpentine; one couldn't help but wonder what might have happened had TVL and AC Nottingham combined forces. The success of small-scale fish-liver oil production during wartime for

the nation's infants is described by Mattingley.

Since 1964, the growth of chemical and allied industries has been large by world standards and industrial chemists and chemical engineers provide essays dealing with many of these. A chemical engineering viewpoint on the lactose industry by Wood is particularly interesting as is Earle's model for university-industry collaboration through NZ Pharmaceuticals.

Collaboration led to NZs dominance in the understanding and utilization of geothermal power - Ellis shows how. The DSIR zeolite team under Parker is largely responsible for NZs success in the conversion of natural gas to petrol by the untried Mobil process. Marshall and Wylde describe steel-making from NZ iron sands as a classic case-study. Similarly, the solar salt industry at Lake Grassmere is told by Pollard. Lastly, NZs great industrial success in establishing a pulp and paper industry based on *Pinus radiata*, is told by former NZFP chemist and managing director, Mackney.

What a collection of plots and authors. Appearing in the cast, from time to time, are politicians like Ward, Seddon, Sullivan, Douglas, Birch, and Muldoon. Too often, they showed a lack of confidence in the ability and ingenuity of NZ scientists and engineers, and blind faith in the overseas expert. One way to help this change would be to make *New Zealand is Different* compulsory reading for every MP, and Government Department Head so that Government's wish to add value to NZ exports might more easily be achieved. Finally, readers of *Chemistry in New Zealand* will find it a snip at $30.

Abridged by the Editor

# Book Review: Handbook of Australasian Edible Oils

Editor: Charmian O'Connor. Managing Editor: Laurence Eyres ISBN 978-0-473-12283-6, 297 pages

With widespread comment and discussion in the media on food related health matters such as obesity, low density lipoproteins vs. high density lipoproteins, saturated fats vs. unsaturated fats, *trans*-fatty acids vs. *cis*-fatty acids, omega-3 fatty acids, 'natural' and modified fats and oils, etc. this publication from the Oils & Fats Specialist Group of the New Zealand Institute of Chemistry is very timely. It contains 14 chapters written by 25 people with expertise in their particular fields. Listing the titles of the chapters is the best way to illustrate this and convey the material covered:

**1.** Methods of analysis – a brief introduction; **2.** Avocado oil and other niche culinary oils in NZ; **3.** The development of the NZ extra virgin olive oil industry; **4.** Setting quality standards for Australasian olive oils; **5.** Marine oils in Australasia; **6.** A survey of omega-3-fatty acids in common NZ seafood; **7.** Marine oil production for nutraceutical use; 8. Lipids and cancer; **9.** Nutracceutical oils derived from plants; **10.** Fat in the diet of children; **11.** International evidence supporting addition of plant sterols and stanols to functional foods; **12.** Processing oils and fats in NZ; **13.** The edible oil industry in Australia; **14.** Deep-frying mediums in NZ.

A twelve page Table of Contents listing the headings of every section in each chapter adequately also fulfils the role of an index, and allows a reader to quickly find a topic of interest. Abbreviations (e.g. PUFA) are widely used through out the book, but an excellent alphabetical list prior to the first chapter allows the reader to find the meaning easily. Every chapter has an extensive list of references to the primary literature.

This book will be invaluable to a wide range of people, both professional and lay. In view of the public interest in many of the topics I would hope all public libraries purchase a copy. Health and food journalists, political spokespersons on health and food should certainly have ready access to it. It will be invaluable for students in the areas of food technology, health and medicine. The composition and properties of all the different fats and oils used in making products for a wide range of food manufacturers, and of the finished products these manufacturers produce and we find in our supermarkets, are given clearly in many tables throughout the book. The manufacture of butter, a major item in our diet, is not covered in this book as it so familiar to most people, but comparisons of its composition and properties with products made from other animal and vegetable fats and oils are given.

Those interested or concerned about the pros and cons of what they should and should not eat will find authoritative information on which to make decisions.

Reading this book reminded me of what I experienced as an "academic chemist" on receiving articles for *Chemical Processes in New Zealand*: how each industry has its own jargon, much of which is familiar to lay people, but the meaning of which is not always obvious to someone outside that industry, be they a chemist, technologist, cook etc. So I was so pleased to find that familiar terms used in chapters 12 and 13, covering the processing of raw materials to produce the range of edible products we find on supermarket shelves, were clearly explained as they were introduced. There were occasions where I felt a brief glossary of some terms would have been useful to readers with limited chemical knowledge, especially on the numbering used in unsaturated fatty acids. However a recent previous publication of the same group, *A New Zealand Lipid Manual*, also edited by Charmian O'Connor, and subtitled *Readings and introduction to the science, technology, analysis and nutrition of oils and fats*, 136 pages, is available (from Ruth Eyres – see below) and provides a basic introduction to this field.

The title *Handbook* is apt as it will serve as a comprehensive reference book for those working and researching in the food and health industries. However it is not just a reference book. Individual chapters are easy to read and cover many topics that should interest consumers.

The Oil and Fats Group should be congratulated in producing this invaluable source of information of interest to a broad range of people.

The cost including GST and postage is $80 and the book can be ordered by post (Ruth Eyres, PO Box 25-499, St Heliers, Auckland), by fax (09 528-7032) or e-mail *eyresy@internet.co.nz*

*Reviewed by John Packer*

# Conference Calendar

**26th Australian Colloid & Surface Science Student Conference, Warrnambool, Victoria, Australia, 4-8 February 2008**

Further details available at the website: *http://home.iprimus.com.au/jaymez/stucon2008/index.html*

**Fifth International Meeting on Photodynamics, Havana, Cuba, 4-8 February 2008**

**New Zealand Trace Elements Group Conference 2008, University of Waikato, Hamilton, New Zealand, 13-15 February 2008**

Further details available at the website: *www.tracenz.org*

**ICAM2008, International Conference on Advanced Materials, Kottayam, India, 18-21 February 2008**

Further details available at the website: *www.materialschem.org/*

**International Symposium on Biothermodynamics, Frankfurt, Germany 21-22 February 2008**

Further details available at the website: *http://events.dechema.de/Biothermodynamics_2008*

**ICONN 2008, 2008 International Conference on Nanoscience and Nanotechnology, Melbourne, Australia, 25-29 February 2008**

Further details available at the website: *www.ausnano.net/iconn2008/*

**Heterocyclic and Heteroatom Chemistry Conference, Heavier heterocycles and heteroatom chemistry, Cancun, Mexico, 25-29 February 2008**

Further details available at the website: *www.zingconferences.com/*

**Pittcon 2008 Conference and Expo; 59th Pittsburgh Conference on Analytical Chemistry and Applied Spectroscopy, New Orleans, Louisiana, USA 1-7 March 2008**

Further details available at the website: *www.pittcon.org/*

**Chem. 05, Green and Sustainable Chemistry for Developing Countries, Cairo, Egypt, 3-6 March 2008**

Further details available at the website: *http://chem05.cu.edu.eg/*

**ASCC Conference 2008, Annual conference of the Australian Society of Cosmetic Chemists, Gold Coast, Queensland, Australia, 6-9 March 2008,**

**Organic Process Research and development, the original process chemistry conference, Dublin, Ireland, 11-14 March 2008**

**10th Young Scientists conference on Chemistry, Rostock, Germany, 27-29 March 2008**

Further details are available at the website: *www.jcf-fruehjahrssymposium.de/2008/*

**WFC10, 10th World Filtration Congress, Leipzig, Germany, 14-18 April 2008,**

Further details available at the website: *www.wfc10.com/*

**ICC2008, International Catalysis Conference, Tehran, Iran, 28-30 April 2008**

Further details at the website: *http://icc2008.sbu.ac.ir/*

**WHEC 2008, 17th World Hydrogen Energy Conference, Brisbane, Australia, 15-19 June 2008**

Further details available at the website: *www.whec2008.com/*

**Dalton Discussion 11: The Renaissance of Main Group Chemistry, Berkeley, California, United States, 23-25 June 2008.**

Further details available at the website: *www.rsc.org/ConferencesAndEvents/RSCConferences/DD11/index.asp*

**III International Conference on Colloid Chemistry and Physicochemical mechanics, Moscow, Russia, 24-28 June 2008**

Further details at the website: *www.icc2008.ru/en/*

**Gordon Research Conference on Polymer Physics, Newport, United States, 29 June - 4 July 2008,**

Further details available at the website: *www.grc.org/*

**Drug Discovery & Development, Couran Cove Island Resort, Queensland, Australia, 13-17 July 2008**

Further details available at the website: *www.3datthecove.org/*

**BOSSXI, 11th Belgian Organic Synthesis Symposium, Ghent, Belgium, 13-18 July 2008,**

Further details available at the website: *www.boss11.org*

**19th IUPAC Conference on Physical Organic Chemistry, Santiago de Compostela, Spain 13-18 July 2008**

Further details available at the website: *www.icpoc2008.org/*

**XXII IUPAC Symposium on Photochemistry, Goteborg, Sweden 28 July - 1 August 2008**

Further details available at the website: *http://photoscience.la.asu.edu/Goteborg2008/*

**5th SETAC World Congress, The Society of Environmental Toxicology and Chemistry, Sydney, Australia, 3-7 August 2008**

Further details available at the website: *www.setac2008.com/*

**XXth International Symposium on Medicinal Chemistry, Vienna, Austria, 31 August - 4 September 2008**

Further details available at the website: *www.ismc2008.org*

**PSA2008, Particulate Systems Analysis 2008, Stratford Upon Avon, United Kingdom, 2-4 September 2008**

Further details available at the website: *www.psa2008.co.uk/*

**Praha 2008, The 20th International Conference on High Resolution Molecular Spectroscopy, 2-6 September 2008**

Further details available at the website: www.chem.uni-wuppertal.de/conference/

**23rd European Colloquium on Heterocyclic Chemistry, Antwerp, Belgium, 9-13 September 2008.**

Further details available at the website: *www.echc08.org/*

**RACI Organic 08, Wrest Point, Hobart, Tasmania, Australia, 7-12 December 2008**

Further details available at the website: *www.organic08.org/*

# Grants and Scholarships

### International Conference Fund

This fund is provided by the Minister of Research, Science and Technology and administered by the Royal Society of New Zealand. It is funding to assist organizations and institutions to host major international conferences in New Zealand. There is no closing date for applications.

For further information see the website: *www.rsnz.org/funding/int_conf/*

### Conference Assistance Programme

This programme provides assistance for bidding to host an international conference in New Zealand. This can include discounted airfares to travel to present bid, help to write bid documents and accompanying marketing material as well as other assistance.

For further details see the website: *www.conventionsnz.com/cap.aspx*

### Technology in Industry Fellowships

These fellowships enable completion of R & D projects in companies. Funding provides a stipend of up to $25,000 depending on the level of postgraduate study, $5,000 host fee to tertiary institute and $1,000 accommodation and travel allowance

For further details, criteria and an application form see the website: *www.frst.govt.nz/Fellowships/Tif.cfm*

### Growth Services Fund

This fund is intended to assist new initiatives that will have a significant impact on a business that will lead to substantial and sustained growth. Co-funding of up to 50% is offered.

For further details see the website: *www.nzte.govt.nz/section/11964.aspx#acc*

### Royal Society of New Zealand Travel Grants

This provides for $1,000 to assist students, undertaking full-time PhD study in science, to attend their first overseas scientific conference (excluding any conferences they have been to in Australia). Closing date for applications is 1 March 2008.

For further details and an application form see the website: *www.rsnz.org/awards/travel/*

### L B Wood Travelling Scholarship

This scholarship is to supplement postgraduate study in Britain. It is for not more than three years and has an annual value of $3000. Closing date for applications is 1 March 2008.

For further details, criteria and an application form see the website: *www.nzvcc.ac.nz*

### Shirtcliffe Fellowship

This fellowship is to assist graduate students of outstanding ability and character to continue their studies. The Fellowship is for up to three years at a value of $5,000 per year. Closing date for applications is 1 March 2008.

For further details, criteria and an application form see the website: *www.nzvcc.ac.nz*

### William Georgetti Scholarship

This scholarship is to help encourage postgraduate study and research, usually in New Zealand. It is usually for three years and provides up to $20,000 per year. Closing date for applications is 1 March 2008.

For further details, criteria and an application form see the website: *www.nzvcc.ac.nz*

### Meat & Wool New Zealand Postgraduate Scholarships

These scholarships are for high achieving students to help complete a postgraduate qualification that will support the sustainable development of the meat and wool industry in New Zealand. Successful Masters applicants receive a stipend up to $30,000 in total, while successful doctoral applicants will receive $25,000 per year for up to three years. Applications close 14 March 2008.

For further details and criteria see the website: *www.meatandwoolnz.com*

### Bank of New Zealand Scholarships

These scholarships are available to Masters, PhD or postgraduate diploma students. The scholarship is $4000 for one year. Closing date for applications is 1 April 2008.

For further details, criteria and an application form see the website: *www.bnz.co.nz*

### New Zealand Science and Technology Postdoctoral Fellowship Scheme

These fellowships are for those with a doctoral degree. Funding is $58,000 per annum for up to three years and up to $29,000 per annum for research related costs. Closing date for applications is 3 April 2008.

For further details see the website: *www.frst.govt.nz/fellowships/PostDocs.cfm*

### Maori Education Trust Professions Scholarship

This scholarship is available to Maori students undertaking research at post-graduate level in a profession where very few Maori are represented. It is valued at $5000. Closing date for applications, April 2008.

For further details, criteria and an application form see the website: *www.maorieducation.org.nz/sch/post_grad.html*

### Horticulture New Zealand Scholarship

This scholarship is for postgraduate students with a special interest in the fruit or vegetable industry. The scholarship is for one year and up to $10,000. Applications close 10 March 2008.

For further details, criteria and an application form see the website: www.hortnz.co.nz/communications/pdfs/Schol-Brochure.pdf

### New Zealand Postgraduate Study Abroad Award

This award is available to postgraduate students enrolled in either Doctoral or Master's degree programmes at a New Zealand institution, whose research would benefit from up to six months of study or research overseas. The value of the award is up to $10,000 depending on the proposed project. Closing date for application is 1 May 2008.

For further details see the website: *www.newzealandeducated.com/int/en/institutions_courses/scholarships/outgoing/new_zealand_postgraduate_study_abroad_award*

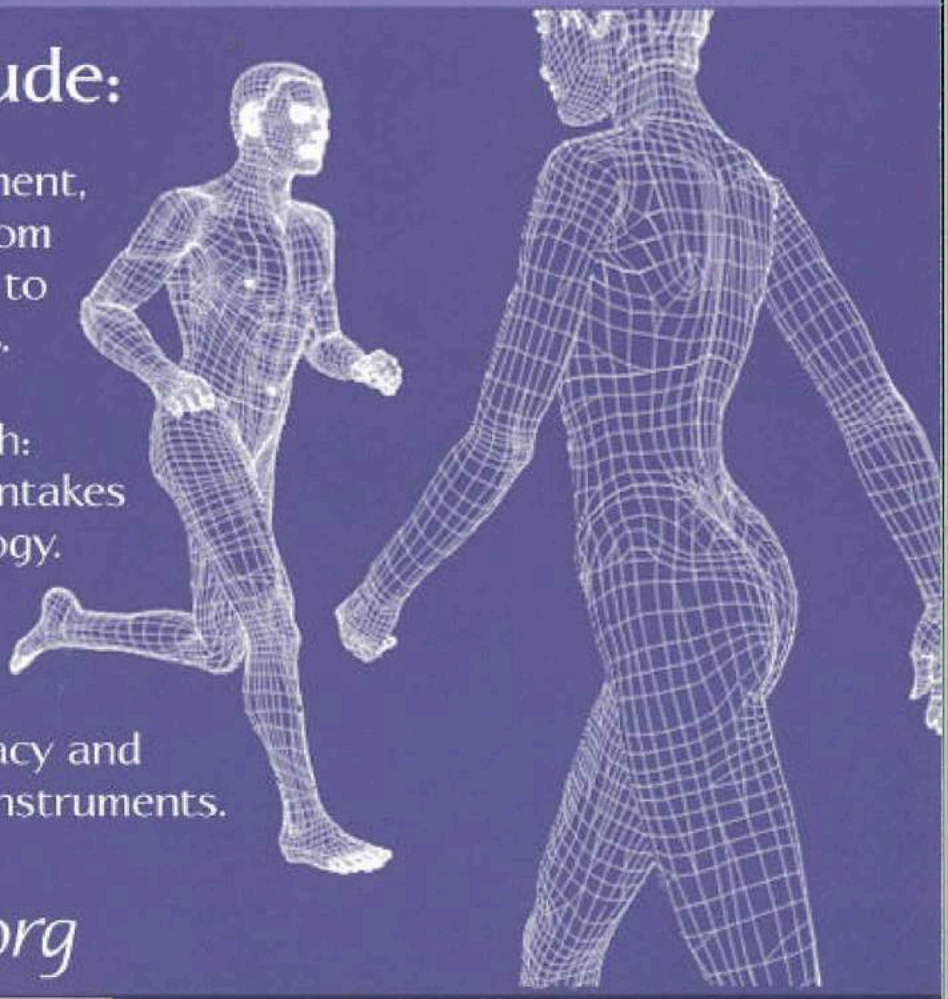
# New Zealand Trace Elements Group
# Conference 2008

## Topics will include:

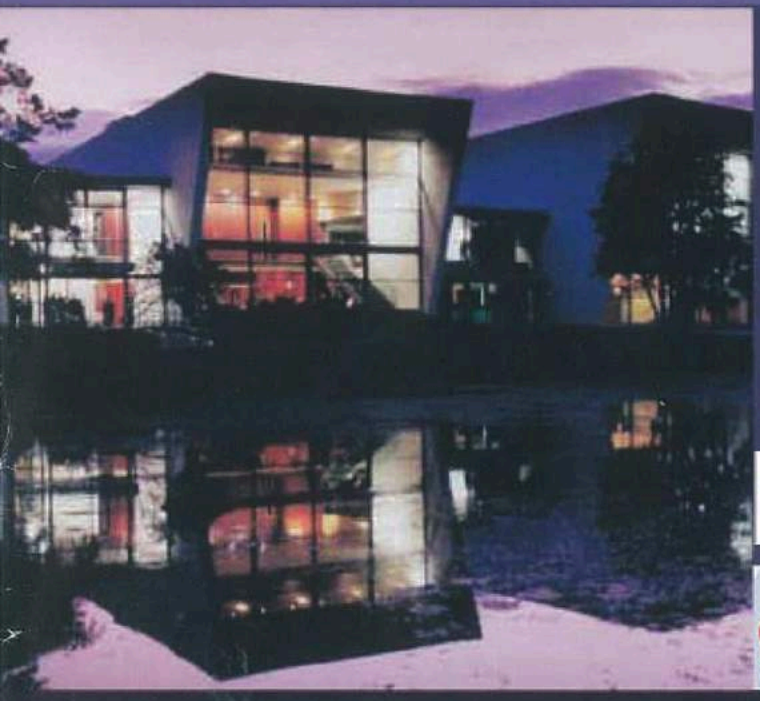
Trace elements in the environment, agriculture and horticulture: from natural geochemical processes to modern anthropogenic impacts.

Trace elements in human health: essential, non-essential, from intakes and biochemistry to epidemiology.

Analytical techniques for measuring trace elements: from ensuring analytical accuracy and precision to state-of-the-art instruments.



Web: *www.tracenz.org*



## Venue:

WEL Energy Trust
Academy of Performing Arts
University of Waikato
Hamilton, New Zealand

tracenz

Hill Laboratories
A WORLD LEADER IN ANALYTICAL SERVICES

NZIC

# Diary dates: 13-15 Feb 2008

New Zealand Institute of Chemistry

The Secretariat and central office of the NZIC is based in Christchurch

*Contact*

Richard Rendle

Honorary General Secretary

PO Box 39 112

Harewood

Christchurch

Phone: +64 3 359 7275

Fax: +64 3 359 7248

Email: NZIC.Office@nzic.org.nz

www.nzic.org.nz