



▶ Allan Wilson Centre lecture series

The 2009 Allan Wilson Centre lecture series was off to a great start in February when audiences packed venues in Dunedin, Christchurch, Wellington, Palmerston North and Auckland to hear Professor Mike Morwood of the University of New England in Australia present "Hobbits in context: the life, times and death of Homo floresiensis."

If you missed the first lecture in the series, Moorwood was a guest on Radio New Zealand's Saturday Morning with Kim Hill on 14 February; the interview is available on www.radionz.co.nz.

The lecture series continues during the week of 23 March when the Allan Wilson Centre's own Professor Lisa Matisoo-Smith delivers "Great Moments in Human History: the Settlement of the Pacific".

One of the Centre's principal investigators, Matisoo-Smith is a biological anthropologist at the University of Auckland. Her research focuses on applying genetic tools to questions regarding Pacific prehistory, including the identification of population origins, migration pathways and prehistoric interactions.

"The settlement of the Pacific region was one of the most extraordinary feats in human history," says Matisoo-Smith. "It involved one of the earliest population migrations out of Africa (more than 40,000 years ago) and the last major human population expansion and it resulted in the settlement of the Polynesian Triangle."

Her talk will address questions such as: where did the various Pacific peoples come from and how and when did they settle this vast region? When did they stop exploring and who did they interact with? How did they adapt to and modify the various environments they encountered? How do we explain and understand the resulting variation we see in

Pacific populations today?

Matisoo-Smith, who has recently joined National Geographic's Genographic Project as the principal investigator for the Pacific Island region (see page two), will be presenting the latest archaeological and genetic data for tracing human population movements across the Pacific. She will focus on her most recent research in Papua New Guinea and South America and the implications of recent findings for understanding this most fascinating chapter in human history.

The third and final lecture in the series, "In Darwin's Footsteps: Bipedalism, Tools and Foraging in the Early Stages of Human Evolution", will be delivered by Professor Jack Harris in April. Harris, a professor of Anthropology at Rutgers University in New Jersey, is also a research associate with the National Museums of Kenya and the coordinator of Kenyan Field Schools in Paleoanthropology, Wildlife Ecology and Conservation.

The earliest stages of human origins are the focus of Harris's work. He is particularly interested in the time between 2.5 million and 1.5 million years ago – the time that has evidence of the earliest manufacture and use of stone tools, the incorporation of meat into the diet, the emergence of the genus Homo and the ranging patterns of our earliest ancestors to Eurasia.

Professor Harris is also involved in collaborative interdisciplinary studies with Primatologists. This collaborative work looks at the behaviours of modern chimps and monkeys, particularly their use of stone tools in foraging and food sourcing as models for the behaviours of the earliest stone tool using hominids.

For dates, times and venues, please see www.allanwilsoncentre.ac.nz or contact Susan Adams on s.i.adams@massey.ac.nz

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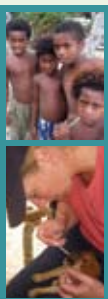
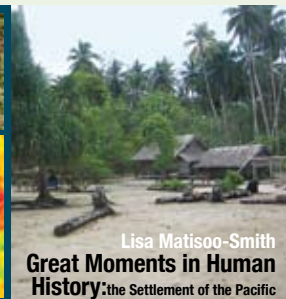
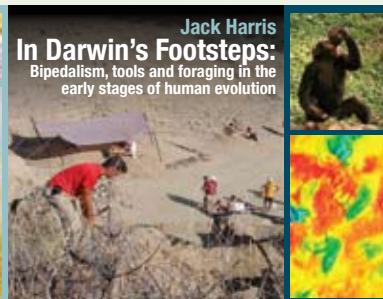
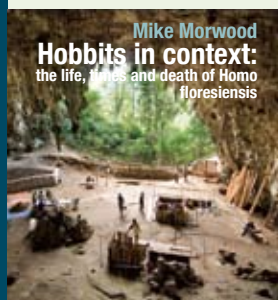
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Lisa Matisoo-Smith named principal investigator for Genographic Project

In August 2008, Allan Wilson Centre principal investigator Professor Lisa Matisoo-Smith joined an elite group of 11 international scientists when she was named principal investigator for National Geographic's Genographic Project.

The US\$40m five-year project, which is funded by *National Geographic*, IBM and the Waitt Family Foundation, was launched in April 2005 and aims to collect and analyse DNA from hundreds of thousands of people around the world, including at least 100,000 indigenous and traditional people.

According to Genographic, "the core of the project is the collection of genetic samples from collaborations with indigenous and traditional populations, whose DNA contains key genetic markers that have remained relatively unaltered over hundreds of generations, making these peoples' genetic histories reliable indicators of ancient migratory patterns." The resulting data, says Genographic, will hopefully map world migratory patterns dating back some 150,000 years and will fill in the huge gaps in our knowledge of humankind's migratory history.

Principal investigators of the project have been selected to visit remote regions of the world. Matisoo-Smith, whose work focuses on Pacific migration, was asked to join the project to help further its work with Pacific Island communities.

The project is led by population geneticist Spencer Wells, the author of *The Journey of Man: A Genetic Odyssey* and *Deep Ancestry: Inside the Genographic Project*.
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▶ Pre-doc internships offered to Māori and indigenous students

The Allan Wilson Centre and Ngā Pae o te Māramatanga, New Zealand's Māori Centre of Research Excellence, have established a joint pre-doctoral internship programme for Māori and indigenous students who are interested in pursuing doctoral degrees within the fields of research of the Allan Wilson Centre.

The programme, which begins in 2009 and has funding for at least three years, will provide an introductory research experience in areas of study such as molecular ecology, evolution, genetics and associated mathematical and computing applications.

"Different cultural perspectives offer new scientific perspectives" says Dr Kristina Ramstad, the Centre's coordinator for the project. "We think this programme is a great opportunity to increase participation of Māori and indigenous students and communities in studies of evolution and ecology and, at the same time, broaden the Allan Wilson Centre's capacity to do science that incorporates Māori and indigenous knowledge and experience."

The two CoREs brought their complementary skills together and designed a programme that is both culturally appropriate and scientifically challenging. Each student will have a cultural advisor from Ngā Pae o te Māramatanga and a scientific advisor from the Allan Wilson Centre.

"The sciences, and particularly leading-edge science such as that done by the Allan Wilson

Centre and other CoREs, are areas where Māori, Pacific and indigenous peoples are especially under-represented in university study," says Professor Michael Walker, joint director of Ngā Pae o te Māramatanga. "We believe the opportunity to fast-track young people from these groups to the leading edge through collaboration with the Allan Wilson Centre is exactly the kind of rapid change through research and advanced training that the CoREs were set up to support. We are delighted to be able to work with the Allan Wilson Centre on this initiative."

Up to ten internships will be offered each year. Each will cover ten weeks of work (which need not be continuous) and can be taken at any time of the year. While ideal candidates need to show a genuine interest in exploring a research career in one of the Centre's research areas, they are not required to have previous experience or degrees in the specialties. Students can be upper-level undergraduates, post-graduates or enrolled in a Master's programme, but they cannot have a PhD or be enrolled in a PhD programme.
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the ROYAL
SOCIETY of
NEW ZEALAND
TE APĀRANGI

▶ Pete Lockhart

... chosen as Leonard Cockayne Memorial Lecturer

The Royal Society of New Zealand has chosen Professor Pete Lockhart for the 2009 Leonard Cockayne Memorial Lecture tour. An Allan Wilson Centre principal investigator and expert in New Zealand alpine plants, Lockhart will speak about recent discoveries in evolutionary biology and describe how new sequencing technologies are being used to further the understanding of the nature – and future – of New Zealand plant species.

While fossil records of plants and their pollen have long been recognised as a kind of black box recorder for their evolutionary history, the interpretation of those records has only recently been corroborated through modern techniques that allow scientists to read the stories in the plants' DNA.

"In 1910," says Lockhart, "Leonard Cockayne published *New Zealand Plants and Their Story* and in it he lamented: 'Perhaps... could they speak, we might learn...' Little did he know that the plants could speak through their DNA. I think Cockayne, to whom this DNA voice was silent, would be amazed at what we can learn from studying the genes and genomes of living plants."

The triennial lecture tour was instituted in 1964 and is sponsored by the Cockayne Memorial Fund. The fund, established to commemorate the life and work of botanist Leonard Cockayne, encourages botanical research in New Zealand.

Lockhart will present his lecture at Royal Society branch meetings around New Zealand in June and July 2009. For a schedule, go to www.allanwilsoncentre.ac.nz and look under "upcoming events". p.j.lockhart@massey.ac.nz

New blog for biology teachers

Hot on the heels of BioEd (read more about the conference on the back page of this newsletter) the Allan Wilson Centre is launching a teachers' blog on its website.

"The blog is aimed primarily at biology teachers as a place where they can discuss things such as ideas on how to teach evolution in the classroom," says Allan Wilson Centre executive director Susan Adams. "BioEd," says Adams, "built a lot of momentum for the teachers, who discovered how much they could learn from each other. The blog gives them an avenue to have ongoing discussions about teaching methods and to share resources and ideas for teaching biology in general and evolution in particular."

The blog will not be moderated, says Adams, so potentially it may spin off into other areas of interest to biology teachers.

Olympians in training

In the last issue of Pheno (September 2008), we highlighted the New Zealand team that brought home three bronze and one silver medal from Mumbai in July at the 2008 International Biology Olympiad. Workshops get underway from 14-23 April to train and select the team that will represent New Zealand in the 2009 games to be held in Japan in July.

"Top biology students from around New Zealand will spend a week in intense biology study," says Dr Heather Meikle, secretary of the New Zealand Biology Olympiad committee. "They will sit exams, develop their practical biology skills and increase their knowledge of biological theory. At the end of the week, the team that will travel to Japan will be chosen."

The Allan Wilson Centre is a sponsor of the Biology Olympians and some Allan Wilson Centre principal investigators will be on the training programme. For more information, please contact Heather Meikle at hmeikle@inspire.net.nz.

New Zealand gets a say

Susan Adams, the executive director of the Allan Wilson Centre, has been named Secretary of the Commission for Biology Education (CBE) of the International Union of Biological Sciences (IUBS).

The IUBS is a non-governmental, non-profit organisation which was established in 1919 with the objectives of promoting the study of biological sciences; initiating, facilitating and coordinating research and other scientific activities necessitating international, interdisciplinary cooperation; ensuring the discussion and dissemination of the results of cooperative research (particularly in connection with IUBS scientific programmes); and supporting the organisation of international conferences and assisting in the publication of their reports. The CBE, says Adams, is influential in steering biology education and identifying priorities. For example, she says, last year's BioEd conference was focused on sustainability; this year's was on Darwin.

"As secretary," says Adams, "I am the New Zealand representative to the CBE – a position which affords me the opportunity to contribute and comment on what's happening in the world of biology education."

In her role with the CBE, Adams will help conveners of the next BioEd conference which will probably be held between June and August of 2011.

▶ Holland gets another gong

Dr Barbara Holland, an associate investigator at the Allan Wilson Centre, was awarded the New Zealand Mathematical Society Early Career Award for 2008 at the Society's December meeting. Holland, who is based at Massey University's Palmerston North campus, was cited for her "groundbreaking" and "pioneering" work in evolutionary biology.

Holland's research focuses on the study of evolutionary relatedness among various groups of organisms. An evolutionary tree, or a phylogenetic tree, maps the evolutionary interrelationships among various species.

"I'd like to understand more about evolutionary trees and the so-called tree of life," she says. "Is it really a tree? Or is there so much intermingling between species that it doesn't resemble a tree at all?"

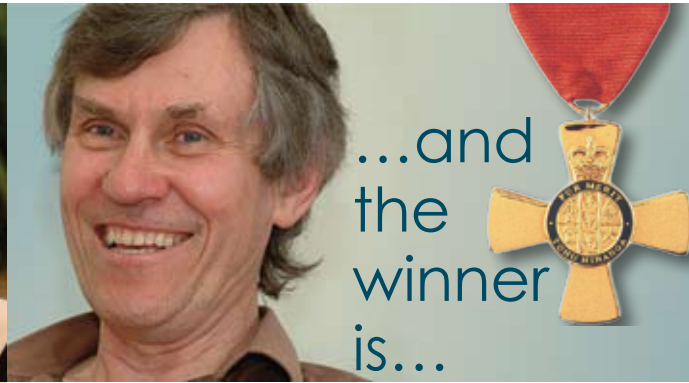
Holland didn't always think she'd be working in evolution. When she finished her undergraduate degree, she figured she'd apply mathematics to the problems faced by business. But that proved dull compared to the wealth of interesting questions posed by evolutionary biologists.

"Biology is a treasure trove of information just trying to be understood. New tools – like the Solexa 2 DNA analyser – produce terrifying amounts of data! Things that really excite me include figuring out how to use that data to get information across genomes and then understanding what may end up being much more complex patterns of evolution than we previously thought."

Such complicated work ends up addressing what at first glance seem to be simple questions. "Does our concept of species really make sense? For example, in animals," she says, "it may, but for plants, it doesn't. So then, we must ask: 'What are species? How do they behave?'"

The Early Career Award is just the latest in a sweep of honours bestowed on Holland. In 2006, she was awarded a Massey Early Career Medal and, in 2005, she won the Royal Society of New Zealand Hamilton Memorial Prize. Also in 2005, she was awarded a young investigator award by the Society for Molecular Biology and Evolution and a Foundation for Research, Science and Technology Bridge to Employment grant for which she received a full Marsden grant as the project's principal investigator.

When she isn't collecting prizes or solving the challenges of evolutionary biology, Holland enjoys riding her mountain bike but admits, "I'm still a complete chicken."



It's been a busy time for Professor Mike Hendy. In November the Allan Wilson Centre principal investigator was awarded the Royal Society of New Zealand's Science and Technology Medal; in December, he received the New Zealand Mathematical Society's Research Award and to wrap up the year, he was named an Officer of the for services to mathematical biology.

Throughout the awards season, Hendy was acknowledged for how his innovative mathematical approach to molecular ecology and evolution has led to the acceptance that evolution is testable. Hendy's work in mathematical biology focuses on modelling natural and biological processes using applied mathematical techniques and tools. The Centre's founding co-director, he has attracted international attention for more than 40 years and his quantitative methodology now forms an integral part of the phylogenetic software packages used worldwide.

Meanwhile, Allan Wilson Centre PhD student Mareike Fischer was awarded the Aitken Prize for the best contributed talk by a student at the annual New Zealand mathematics colloquium in December. Fischer, who is a student at the University of Canterbury, is supervised by Professor Mike Steel and was honoured for her talk "Curious properties of Maximum Parsimony in estimating evolutionary trees and ancestral sequence states".

The annual award, given by the New Zealand Mathematical Society is named for New Zealand-born mathematician Alexander Craig Aitken.

Kiwi feather cloak poster brings home top prize

Katie Hartnup, an Allan Wilson Centre PhD student at the Institute of Natural Sciences at Massey University's Albany campus, was awarded first prize in the student poster competition at a Charles Darwin symposium held at the University of Auckland on 12 February 2009 – the 200th anniversary of Charles Darwin's birthday.

Hartnup's poster "Historical DNA Analysis of Māori Feather Cloaks: Kahu Kiwi" is an introduction to her research, which is supervised by Allan Wilson Centre investigators Craig Millar and Leon Huynen. Hartnup's work was featured in the June 2007 and March 2008 issues of the Allan Wilson Centre newsletter, available on www.allanwilsoncentre.ac.nz

RESEARCH



▶ How distant is the drumming?

Most people assume that the tools of evolutionary analysis are applied only to living organisms. But visiting intern Martin Scholz was interested in tracing the evolutionary history of traditional African drum rhythms.

Scholz, a graduate student from the University of Applied Sciences in Weihenstephen, Germany, spent four months in 2008 working with Allan Wilson Centre principal investigators Allen Rodrigo and David Bryant.

While modern evolutionary research often relies on genetic analysis, the fitness and chance to pass on genetics also depends on an organism's social and cultural surroundings, explains Scholz. Consequently culture, especially music, can also be a good measure for the interpretation of affinity among human populations. Scholz, who plays the drums himself, compared traditional African rhythms using several distance measurements combined with new ideas of comparing single beat-line rhythms. He then compared the distances between rhythms to the geographical distances between regions, and found that there is a statistically significant affinity between geographical distance and rhythm distance.

"In a sense, Martin was able to construct a very simple evolutionary distance based on the homology of pauses and beats across different rhythms," says Rodrigo. "Using these distances to build trees provides a means of visualising how these rhythms are related on a geographic scale."

The algorithm, Scholz says, introduces new ways of comparing single beat line rhythms and these ideas could also be applied to studies of melody comparisons in the future.

Scholz returned to Germany at the end of 2008, where he is completing his studies and preparing his research results for publication.



▶ Congratulations Dr Jon Waters

At the end of 2008 Dr Jon Waters, an Allan Wilson Centre associate investigator, was promoted to associate professor at the University of Otago. Waters' research interests include molecular evolution and biogeography. He pioneered the use of river capture as a means of calibrating molecular clocks and recently initiated a novel direction in marine phylogeography using DNA techniques to test the evolutionary and ecological importance of kelp-rafting in the marine environment. His post-graduate students are conducting conservation genetics research in yellow-eyed penguins, bull-kelp, takahe, New Zealand freshwater fishes, trematode parasites and southern stoneflies.

It's ok, we're not cousins by blood.

Yeah right.®



Tui



▶ Rethinking marriage taboos

Being able to use a beer ad to illustrate a scientific article is a feat about which Allan Wilson Centre principal investigator Hamish Spencer is quite thrilled.

One of Spencer's current projects examines how marine intertidal snails – not known for their great swimming ability – manage to be widely spread around the Southern Hemisphere. But that's not the story that fitted a Tui advertisement so nicely.

In December 2008, Spencer – a specialist in statistics and population genetics based at the University of Otago – teamed up with long-time collaborator Diane Paul (an emeritus professor from the University of Massachusetts and a research associate at Harvard University) to take a look at what has been tagged 'The Cousin Marriage Controversy'. The historical perspective, which is freely available through on-line journal *Public Library of Science Biology* (PLoS Biology), opines that laws that prevent cousins from marrying are outdated.

"It's not really an important question to most people," says Spencer. "But in New Zealand you most certainly can marry your first cousin as you can everywhere in the western world with the exception of 31 of the United States, as well as a few Asian countries including mainland China and Korea."

Still, the practice is highly stigmatised – which explains the Tui billboard ("It's OK, we're not cousins by blood. Yeah right."). The popular explanation is that offspring of such unions are at a higher genetic risk than other children, but Spencer and Paul (who Spencer describes as one of the world's leading genetic historians) pointed out that this risk is about the same as that to children born to women over the age of 40, a risk we as a society consider acceptable.

Spencer's interests frequently find applications at the interplay between genetics and society and, a self-described 'gadfly', he applies his interests as a tool to examine contemporary issues in medicine and medical genetics.

When considering such topics, he says, it is important to not only look at the scientific evidence available, but the historical perspective in which the laws or attitudes arose. For example, while first cousins tying the knot might be taboo in many families, for centuries it was acceptable; in fact, Charles Darwin did just that when he married Emma Wedgwood. But many of the laws in the US, he says, were passed after the country's Civil War as some states began to intrude into what had previously been considered private aspects of life (e.g., the minimum age people could marry).

"Unlike the situation in Britain and much of Europe, cousin marriage in the US was associated not with the aristocracy and upper middle class but with much easier targets: immigrants and the rural poor," Spencer and Paul wrote in the paper.

While the cousin marriage debate may seem as outdated as the laws, examining it may help illuminate the way society considers modern medical controversies. Spencer, who has also written about the history (and failing) of eugenics, uses his work in population genetics to ask the broadest of questions, including: what are the things that we think are important in society? The applications of his work often arise when genetics and society mingle and when we consider such things as: what kinds of things should people be tested for? Who should have access to the data generated by medical tests? Is it fair to allow insurance companies access to a person's medical test results? How do we manage pre-natal testing and genetic diagnosis of embryos?

Along with his research into snails, molluscs and bull kelp, Spencer is also interested in developing mathematical models that will help explain how genes can affect offspring without being passed on. "A female, for example, may have a particular gene that affects the way she nurtures her offspring," he says. "She won't necessarily have passed on that gene to the offspring, but the success of her offspring will benefit from her having it." h.spencer@otago.ac.nz



Kelp holds clue to Ice Age

On the day Pheno contacted Ceridwen Fraser to talk about her work, the Allan Wilson Centre PhD student was collecting samples along the Dunedin coast.

“Some very exciting kelp washed up yesterday,” she explained later in the day.

Fraser’s work is centred on the genetic diversity of southern bull kelp, *Durvillaea Antarctica*, but the samples she usually collects aren’t often so convenient to her University of Otago laboratory. In a paper she recently published in the Proceedings of the National Academy of Sciences (PNAS), data was derived from bull kelp specimens collected from many of the sub-Antarctic islands including Marion and Gough (accessed from South Africa), the Falklands (accessed from South America) and Campbell, Auckland, the Snares and Antipodes (accessed from New Zealand). She’s also collected samples from along the coast of Chile and, of course, New Zealand. It’s meant a lot of time at sea and physically, it’s gruelling work.

The kelp grows in extremely rough surf and to get samples, she wears a wet suit, carries an axe and whacks the stuff off at its base.

The time available to get samples doesn’t always correspond to nice weather and flat water. With only three weeks on Gough Island and unrelenting four- to seven-metre surf, for instance, Fraser had the added stress of spending a lot of time and money getting to an island and perhaps not being able to collect any samples. But getting into the water was terrifying, too, she laughs.

“I feel like I’ve grown up a lot in the last couple of years,” she says. “It’s been an unforgettable, life-changing experience, but it’s been exhausting, both emotionally and physically.”

Along the way, she has had some unusual experiences. On the trip to Marion Island, for example, the boat needed to divert its course to rescue survivors from a sunken yacht; on the trip to Gough Island, there was a stabbing.

But through it all, she collected the specimens and when she analysed the data was able to draw some surprising conclusions.

“We found that there is a lot of genetic diversity of the bull kelp further north, but next to no diversity further south. What this suggests,” says Fraser, “is that the southern locations were only recently colonised by the species.”

The implication that follows, then, is that the sea ice during the last Ice Age was more extensive than previously thought and during the last Ice Age, kelp would have been physically knocked off by the ice rubbing against the shore. When the ice receded (after the Ice Age), the kelp could come back in and colonise. In the kelp DNA, Fraser found evidence of sea ice wiping kelp off islands further north than other studies ever estimated the ice had reached.

In places like New Zealand and Chile – that are well out of the ice zone – there is a lot of genetic diversity from one collection site to another. This suggests, says Fraser, that the kelp has been there for a long time, enabling genetic diversity to occur. If the kelp had been in the sub-Antarctic islands for a long time, Fraser says she would expect that same level of diversity. However, because it’s all genetically identical, she can assume that it all came from a single source and hasn’t had time to mutate.

“This research has shown that sea ice could have been more extensive. But we’ve based that on one species of kelp, so it’s really hard to draw big conclusions,” she says. “Other studies, including genetic research of a whole range of species, and geologic studies, need to be conducted in these regions, and I’d like to continue the work on other species.”

The results of Fraser’s research that were reported in the PNAS not only found their way to the *Otago Daily News* and *The Press* but hit the wire through the Singapore office of Reuters, and landed in publications including the *Arab Times*, the *Economic Times*, the *Al Watan Daily*, the Australian science magazine *Cosmos* and online science network *Redorbit*.

“It’s been nice to have the media attention,” she says. “It makes me feel like I’m doing something worthwhile that people care about.”

Fraser, who plans finish her PhD by May, hopes to secure funding that would allow her to continue the project. “We’ve answered some questions with this work but in the process have brought up some new ones,” she says. “That’s part of what makes science exciting.” ceridwen.fraser@gmail.com

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bioEd success story

February 12th wasn't just the 200th anniversary of Darwin's birth. It was also the first day of BioEd – Evolution in Action.

The first of seven coordinated international events under the umbrella "Darwin 200 Symposia" BioEd was held at the Christchurch Convention Centre. BioEd opened with a birthday party for the man himself (televised on TV3 news), ran for three days and attracted 160 people from 25 countries.

The Darwin 200 Symposia, which celebrate the birth of Charles Darwin, are being run under the auspices of the International Union of Biological Sciences (IUBS), Commission on Biological Education (CBE) and the United Nations Educational Scientific and Cultural Organization (UNESCO).

"The conference was on biological education," says Professor Pete Lockhart, conference convenor, "and an important focus for many of the sessions was on the teaching of evolution in the classroom. It was a great conference that brought people from vastly different disciplines together in an informal, friendly setting."

Dr Heather Meikle, a biology teacher at Palmerston North Girls High School, agrees.

"We had world leaders in evolutionary biology sitting down and eating lunch with science teachers from the smallest schools in New Zealand. It was wonderful – they told us great stories and the camaraderie was fantastic."

"For teachers," says teacher Ally Jerram, "it represented a rare chance for contact with scientists working in evolution around the globe and for the scientists, an opportunity to see how teachers use the research data that they generate."

Highlights from the scientific sessions included talk about the evolution of medieval manuscripts by Cambridge University Professor Chris Howe and a discussion on French teachers' and students' conceptions on evolution by a group of scientists visiting from the University of Lyon, in France. Outside the scientific sessions, two plays had their premier performance as part of the conference: Collapsing Creation on the first night and Unnatural Selection on the last. Collapsing Creation was a professional production sponsored by the Allan Wilson Centre while Unnatural Selection was written by Bernard Beckett, a high school teacher and former Allan Wilson Centre Royal Society Teacher Fellow, and performed by his students.

CONFERENCE

The Allan Wilson Centre will be hosting the 11th annual Australasian Research Management Society (ARMS) conference

Date: 16 to 18
September 2009

Location:
Christchurch
Convention Centre,
Christchurch,
New Zealand

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SCIENCE AT THE BEACH

Participants from 10 countries came to Kaikoura in February for the 13th annual New Zealand phylogenetics conference. Hosted by the Allan Wilson Centre, the meeting was held at the University of Canterbury's marine laboratory and brought 50 mathematicians and evolutionary biologists together for five days. The conference dealt with a wide range of evolutionary topics from purely mathematical results through to applications, such as the evolution of song in North Island saddleback birds and the use of phylogenetics in the thoroughbred horse industry. Highlights included the keynote talk "Hydrothermal vents and the origin of life", delivered by Professor Bill Martin of the University of Duesseldorf in Germany and "Darwin Rocks", a computer program written by Professor Daniel Huson from Tuebingen University, also in Germany, in which random tunes evolve as the audience selects ones they like the sound of.

