ALLAN WILSON CENTRE

MARSDEN FUND WINNERS

Colliding genomes, the adaptability of small populations, flight-loss in insects, and the evolution of cancer are all research projects winning grants from the prestigious Marsden Fund in the 2014 funding round. The four projects by Allan Wilson Centre Investigators will each receive $808,000 in funding.

These four projects were among 101 selected from more than 1200 applicants from New Zealand universities, Crown Research Institutes and independent research organisations.

When genomes collide: how hybrid species respond to genome shock

Allopolyploidy, the making of a new hybrid or fusion species from different parent species, has greatly influenced evolutionary history. Modern day examples include ligers and tigons (the offspring of lions and tigers), mules (the offspring of horses and donkeys), and even the humble hybrid cotton plant – which produces the raw material used to make cotton clothing.

However, to combine different genes, proteins and chromosomes and yet still flourish, new allopolyploid species must survive 'genome shock'–the molecular incompatibilities that occur when different genomes find themselves in the same cell. Typically some genes are lost and there is genomic restructuring, as well as other molecular changes. One of the problems is that we do not know whether these changes are adaptive responses or just passive consequences of genome merger.

Murray Cox is an Associate Professor in computational biology at Massey University. He is an Allan Wilson Centre for Molecular Ecology and Evolution Associate Investigator and also an inaugural Rutherford Fellow of the Royal Society of New Zealand.

An Allan Wilson Centre Associate Investigator from Massey University, Associate Professor Murray Cox, seeks to build on an earlier pilot study, which identified several commonalities in the response to genome shock across very different hybrids. He says further research had been limited by technical hurdles, but this project will develop new tools to overcome these limitations.

"It is increasingly clear that not all allopolyploids have fitness benefits and many actually show reduced reproductive success. The major impediment to understanding why some allopolyploids are ecologically successful, yet others are not, has been our limited understanding of the responses that occur when genomes merge," says Murray.

He notes that furthering our understanding of how fusion species react to genomic shock and being able to characterise these responses within a comparative framework, such as whether various responses are reproducible and whether they have adaptive or functional significance, will have major implications for plant and animal breeding – especially for economically important plant fusion species, such as wheat, and New Zealand's own rich collection of natural and agricultural allopolyploids.

Murray's research group will be focusing on a group of fungi, Epichloë endophytes, that includes many fusion species and forms economically-important beneficial symbioses with agricultural pasture grasses such as perennial ryegrass, the main pasture grass in New Zealand. The fungus benefits by gaining a place to live, derives nutrients from the host, and uses the host's seed dispersal as its own means of dissemination. In return, the plant acquires increased tolerance to drought and makes itself an unpleasant meal for many insect species.

"In our recently published pilot study on *Epichloë*, we examined RNA, the instruction sheets needed to build proteins. Since cells contain two versions of RNA, and consequently two versions of every protein, we wondered how hybrid species keep their cellular machinery working.

We found that the hybrid produces both versions at a level found in either one parent or the other, but not some intermediate state. This means that one version of most proteins often does not get made. Determining which version gets switched off follows a complex, but regular, pattern.

The only other hybrid species in which a similar analysis has been performed is cotton. Surprisingly, the complex patterns that describe which RNA version is switched on and off in the *Epichloë* endophyte are almost identical to the patterns found in cotton.

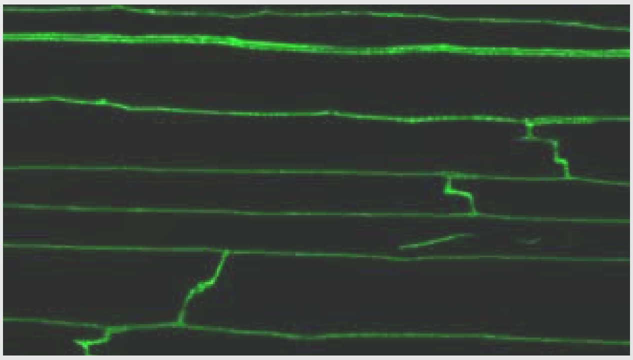
This was unexpected as the two species are radically different, from different kingdoms, with very different challenges to their survival.

These shared patterns suggest that there are universal rules that determine how gene expression in hybrid species controls the two sets of protein machinery. If there are universal rules, research on one hybrid could tell us how gene expression functions in a completely different hybrid species – which would be useful given they make up a large part of the agricultural sector, and are increasingly being recognised in nature.

We'll also be extending our research to look at three-parent fungal allopolyploids and how they deal with genome merger."

Murray says that the new research project will feed directly into applied research programmes, including AgResearch's 'third generation' commercial endophyte products - a programme that seeks to bring new agricultural species to market, including *Epichloë* endophytes with a broader repertoire of chemicals to better protect crops against insect damage.

A transmission electron microscope image emphasizing the close symbiotic relationship between the fungal endophyte and its grass host. A single fungal cell (dark circle in center) is growing at the junction of three plant cells (light spaces). Credit: Prof Murray Cox, Massey University.



A light microscope image showing cells of a genetically modified fungal endophyte (fluorescent green lines) growing between cells of the grass host (dark spaces) in the laboratory. Credit: Prof Murray Cox, Massey University.

Dr Anna Santure is a lecturer and researcher at the University of Auckland's School of Biological Sciences. She re-joined the Allan Wilson Centre in 2014 as an Affiliate Investigator when she returned to New Zealand after several years working in the UK. Anna has a background in mathematics and genetics, and was previously a member of the AWC as a University of Otago PhD student from 2002–2006.

Credit: Dr Anna Santure, University of Auckland.

Predicting the adaptive potential of small populations: New Zealand hihi

Predicting how populations will adapt to environmental challenges, such as climate change, is crucial, especially for many small populations of endangered New Zealand species. But knowing how and whether a population will adapt requires knowledge of both the complex genetic basis of traits linked to survival and reproduction, and the selection pressures acting on the population.

An Allan Wilson Centre Affiliate Investigator from the University of Auckland, Dr Anna Santure, along with colleagues Dr John Ewen and Dr Patricia Brekke from the Zoological Society of London, will be studying a reintroduced population of the endangered New Zealand native bird, the hihi or stitchbird (*Notiomystis cincta*), on Tiritiri Matangi Island. She says her research team aims to understand how the history of these birds has determined the genetic diversity we see today and predict how the population might respond to future selection pressures.

"Since reintroduction the wild hihi population on Tiritiri Matangi Island has been well studied by the Hihi Recovery Group, co-chaired by Dr Ewen, with individual measurements of body size and other morphological traits, annual adult survival and annual reproductive traits including laying date, clutch size, number of clutches, nestling and fledgling survival and offspring recruitment. In addition, almost the entire population has had blood samples taken which we can extract DNA from. This offers us a unique opportunity to determine both the genetic basis of traits important for survival and reproduction, and the selection on these traits."

"Happily, the island also has a weather station that records temperature, wind, humidity and rainfall, which we can use to understand the selection pressures the population is under."

Previous research has shown that higher than normal temperatures late in the breeding season can cause the death of hihi chicks. Anna says that with rising temperatures the island, and other reserves in the same northern zone, may become unsuitable for hihi unless the populations can adapt to a warmer and drier climate.

"As part of the research we will identify hihi with high predicted fitness for breeding programmes and for founding new populations, maximising the evolutionary potential of these new populations – to the best of my knowledge, this will be the first time anywhere in the world genomics has been applied to the adaptive management of an endangered species. We expect the research will help us understand the evolutionary potential of other threatened populations, as well as broaden our biological understanding of how evolutionary processes are determined by genetic traits in wild populations. This should allow us to produce guidelines for the use of genomics to manage other endangered species," says Anna.

Dr Anna Santure, an Allan Wilson Centre Affiliate Investigator, will be studying the adaptive potential of New Zealand hihi.

Credit: John Ewen.

Jon Waters is Professor of Zoology at Otago University and a Principal Investigator with the Allan Wilson Centre. He specialises in the analysis of genetic variation in spatial and evolutionary contexts, with a particular interest in how biological communities respond to environmental change (such as climate fluctuations), and also the impact of historical contingency (such as extinction events) in ecology and evolution.

Use it or lose it: flight-loss in alpine insects

No where is the 'use it or lose it' principle more true than in evolution. In *Origin of the Species*, Charles Darwin recognised the trend commenting that, "Rudimentary, atrophied, or aborted organs, or parts in this strange condition, bearing the stamp of inutility, are extremely common throughout nature."

Among the many other observations of redundant morphology he noted, "Nothing can be plainer than that wings are formed for flight, yet in how many insects do we see wings so reduced in size as to be utterly incapable of flight, and not rarely lying under wing-cases, firmly soldered together!"

Over 150 years later we know this 'evolution in reverse' characterises biology at scales ranging from the molecular to gross morphology and drives much of the world's biodiversity, and yet we still know little about the genetic mechanisms behind the process.

An Allan Wilson Centre Principal Investigator, Professor Jon Waters from the University of Otago, will be attempting to unravel the genetic basis of flight-loss in New Zealand's own alpine insects. He says that our alpine habitats support a host of recently-evolved flightless insect lineages, providing a unique system for understanding how animals respond rapidly to new conditions.

"Our extraordinary diversity of wing-reduced alpine stoneflies, in particular, represents a natural laboratory for understanding how species form. At least 25 of New Zealand's 120 endemic *Plecoptera* species currently exhibit wing reduction, with five of the 11 *gripopterygid* genera being completely alpine and wingless.

"In addition, widespread genera such as *Zelandoperla* and *Zelandobius* have numerous wingless or wing-reduced alpine populations, implying numerous independent losses of flight. There is a clear association between altitude and wing reduction, with lowland populations dominated by fully-winged phenotypes, versus wingless or wing-reduced populations at high altitude."

Using next-generation genomic and transcriptomic techniques, Jon's research team will hunt for 'speciation genes'–the genetic changes that cause the loss of flight in high-altitude populations.

"We will also undertake comparative studies of winged and wingless species to help understand the biological consequences of flight loss."

Jon suggests that these rapid changes may underpin diversification: "While these genetic processes could be seen as evidence of evolution's power to destroy rather than create, such repeated transitions can potentially underpin high rates of diversification."

Jon explains that the loss of dispersal ability can drastically alter a taxon's evolutionary trajectory, by driving reproductive isolation, geographic isolation, and rapid molecular divergence, "As part of the study we will be test the hypothesis that wingless populations experience relatively rapid rates of genetic divergence and cladogenesis [the formation of a new group of organisms by evolutionary divergence] relative to flighted populations."

The research also proposes to test the hypothesis that repeated wing-reduction events in stoneflies may be mediated through a small handful of genes and that these speciation genes will show evidence of convergent evolution across wingless lineages. "If we can unravel the genetic changes, that is, identify the speciation gene or genes promoting these morphological switches, this will be the key to understanding evolution."

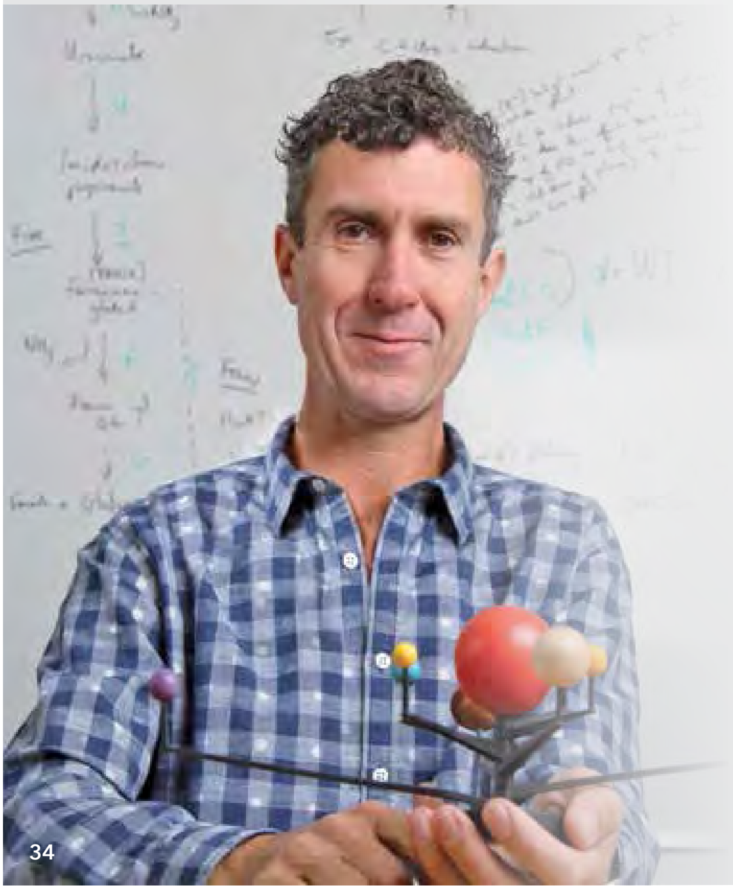
Professor Jon Waters, an Allan Wilson Centre Principal Investigator, will be studying the genetic basis of flight-loss in alpine insects. Credit: University of Otago.

Paul Rainey FRSNZ is Distinguished Professor of Evolutionary Genetics at Massey University's New Zealand Institute for Advanced Study and a Principal Investigator in the Allan Wilson Centre. He is also a Scientific Member of the Max Planck Society and External Director at the Max Planck Institute for Evolutionary Biology in Plön, Germany, and currently holds a Chaire Blaise Pascal at the École Supérieure de Physique et de Chimie Industrielles de la Ville de Paris.

Lineage selection and the evolution of cancer

Cancer is a problem of evolution; its origins are intimately and inseparably connected to the origins of multicellular life on Earth. The building blocks of the first multicellular organisms were single cells capable of evolving as independent units. In order to exploit new ecological opportunities single cells formed simple cellular collectives. For such collectives to persist the benefits from community existence had to exceed the costs to individual members; however, the very existence of such collectives created opportunity for cell types that do not contribute toward collective functionality, but nonetheless take advantage of benefits derived from being part of the collective.

The unchecked growth of these free-loading 'cheating' types threatens the integrity of the collective and marks the earliest manifestation of cancer. So, from the most primordial stages in the evolution of multicellular organisms there emerged conflicts between the reproductive interests of collectives and their constituent cells. Conflicts between different levels of biological organisation remain a persistent challenge to the viability and integrity of extant multicellular organisms, including humans.



Building on a five-year research project into the evolution of multicellularity, published in November 2014 in the science journal *Nature*, Allan Wilson Centre Principal Investigator, Professor Paul Rainey, from the New Zealand Institute for Advanced Study, and his team will attempt to use simple multicellular organisms that evolved in real-time evolution experiments to perform a combined empirical and theoretical analysis of the earliest events underpinning the evolution of cancer and the mechanisms that suppress it.

"When cells cooperate as a community to produce a multicellular organism, sometimes a single cell will forget it's part of a collective. It acquires a gene mutation that makes it multiply or gives it longevity, giving it an advantage over other cells – it's natural selection favouring the individual rather than the collective. These abnormal cells might acquire more mutations in more genes, allowing them to run amok – this uncontrolled growth is cancer to the collective.

"In our previous experiments using single bacterial cells, we were able to capture the emergence of simple multicellular life, replete with a soma/germ distinction, from single-celled organisms in real-time. These new organisms were made up of a single tissue for acquiring oxygen, but this tissue also generated cells that are the seeds of future generations: a reproductive division of labour. Intriguingly, the cells that serve as a germ line were derived from 'cheating' cells whose destructive effects were tamed by integration into a life cycle that allowed groups to reproduce."

Paul says the life cycle turns out to be a spectacular gift to evolution. "Rather than working directly on cells, evolution was able to work on a developmental programme that eventually merged cells into a single organism. When this happened groups began to prosper with the once free-living cells coming to work for the good of the whole.

"The emergence of these primordial life cycles holds the key to understanding some of biology's most profound problems: the origins of multicellularity; the origins of soma/germ differentiation, of reproduction, of development – even the origins of cancer."