Genetics Otago Tech Week 2023

Annual Symposium & Workshops

14th - 16th November

St David Lecture Theatre Complex

With thanks to our sponsors

Schedule at a Glance

Tuesday, 14th November	Wednesday, 15th November	Thursday, 16th November
Workshops Block I 9:00 am – 12:00 pm	Workshops Block III 9:00 am – 12:00 pm	Symposium Opening 9:00 am
Morning Tea Available 10:00 am – 11:00 am	Morning Tea Available 10:00 am – 11:00 am	Symposium Session I 9:10 am – 10:20 am
Break 12:00 pm – 1:00 pm	Break 12:00 pm – 1:00 pm	SciCom Presentations (SciCom Award voting opens) 10:20 am – 10:30 am
Poster Judging Session I 12:30 pm – 1:00 pm	Poster Judging Session II 12:30 pm – 1:00 pm	Morning Tea (SciCom Award Voting Closes) 10:30 am – 11:00 am
Workshops Block II 12:00 pm – 5:00 pm	Afternoon Tea Available 2:00 pm – 3:00 pm	Symposium Session II 11:00 am – 12:30 pm
Afternoon Tea Available 2:00 pm – 3:00 pm	Retirement Special Session 4:15 pm – 5:30 pm	Lunch 12:30 pm – 1:30 pm
	Drinks and Nibbles 5:30 pm -7:00 pm	Poster Judging Session III 1:00 pm – 1:30 pm
		Symposium Session III 1:30 am – 3:00 pm
		Afternoon Tea 3:00 pm – 3:30 pm
		Symposium Session IV 3:30 am – 4:40 pm
		Awards Presentation 4:45 pm

Workshop Descriptions

Workshops Block I

This workshop block will hold concurrent sessions for the Oxford Nanopore Technology Workshop Part One and the Science Communication Workshop.

Oxford Nanopore Technology Workshop Part One – G09 Teaching Lab, Biochemistry Building
Time: 9:00 am – 12:15 pm

9:00 am – 9:15 am	Welcome and Introduction	Augustine Chen and Rob Day *Hub Leaders, ONT Hub*
	Research exemplars utilising different ONT kits (15min talks)	
9:15 am – 9:30 am	Could direct RNA sequencing be used to detect RNA-protein interactions?	Anh Thu Phan
9:30 am – 9:45 am	Limits of detection in isoform sequencing	Amie Siemonek
9:45 am – 10:00 am	Adaptive sampling to unravel the secrets of the human genome: The untold tricks	Lamia Khaled
10:00 am – 10:15 am	Optimizing multilocus multiplex long read amplicon sequencing for discovering alleles of flowering time genes	Ayodele Fakoya
10:30 am - 10:45 am	Morning Tea Break	
	Research exemplars (continued)	
10:45 am – 11:00 am	Promethion-powered sequencing of native species to drive novel aquacultural opportunities	Marc Bailie
11:00 am – 11:15 am	Using rolling circle amplification and Nanopore sequencing to improve accessibility in non-invasive cancer diagnostics	Michael Dunnet
	Practical Workshop	
11:30 am - 12:15 pm	Flow cell loading	Augustine Chen

Science Communication Workshop – St David Seminar Room A
Time: 9:00 am – 11:00 am
Zoom Link: https://otago.zoom.us/j/94565970757?pwd=TVB3WnhCcTM5ZTRBK014Qk1VTUUzZz09
Meeting ID: 945 6597 0757, Password: 851598

This session will be facilitated by Nic Rawlence and Ellie Rowley and will cover building a social media profile, science outreach, marketing your publications to get coverage and writing opinion pieces. We'll then get people to write a pitch for The Conversation and then take them through how to write an opinion piece.

Participants will require a laptop to participate fully in this workshop.

Workshops Block II

This workshop block will hold concurrent sessions for the Oxford Nanopore Technology Workshop Part Two and the Teacher Development Workshop.

Oxford Nanopore Technology Workshop Part One – St David Seminar Room A
Time: 1:00 pm – 5:00 pm

	Computer-based Presentations and Workshop	
1:00 pm - 1:30 pm	Nanopore Update and Transcriptomics Validation Presentation	Warren Bach *Senior Strategic Account Manager ANZ,* *Oxford Nanopore Technologies*
1:30 pm - 2:00 pm	Assay Options: Direct RNA or cDNA	Simon Dunbar *Field Applications Scientist* *Oxford Nanopore Technologies* Rebecca Chesterfield *Technical Applications Scientist* *Oxford Nanopore Technologies*
2:00 pm - 2:30 pm	EPI2ME Labs: Introduction to Platform	
2:30 pm - 3:00 pm	Afternoon Tea Break	
3:00 pm - 3:30 pm	General Considerations: a roadmap from wet lab to analysis	Simon Dunbar *Field Applications Scientist* *Oxford Nanopore Technologies* Rebecca Chesterfield *Technical Applications Scientist* *Oxford Nanopore Technologies*
3:30 pm - 5:00 pm	Computer lab hands-on experience Epi2Me GUI (using demo data) wf-transcriptomes (command line)	

Participants will require a laptop to participate fully in this workshop.

Teacher Development Workshop – G09 Teaching Lab, Biochemistry Building
Time: 1:00 pm – 4:00 pm

This session will be facilitated by Emma Wade and Rebecca Oliver and will cover the use of bioinformatics and gel electrophoresis as tools to investigate the cause of rare genetic diseases. This session will use our Genetics on the GO kits, so that the participants can easily transfer the exercises back to their classrooms.

Workshops Block III

This workshop block will hold concurrent sessions for the CRISPR Workshop and the Ethical, Legal and Societal Considerations (ELSC) Hub Discussion.

CRISPR Workshop– St David Seminar Room A
Time: 9:00 am – 12:00 pm

Time	Title	Speaker
9:00 am – 10:00 am	An End-to-End Solution for CRISPR Genome Editing Research	**Joe Frangipane** *CRISPR Field Application Scientist* *Integrated DNA Technologies (IDT)*
10:00 am – 10:30 am	The power of CRISPR editing to study disease	**Jennifer Hollywood** *Postdoctoral Research Fellow* *University of Auckland*
10:30 am – 11:00 am	CRISPR-based diagnostics aided by machine learning	**Benjamin Duran-Vinet** *PhD student* *University of Otago*
Short Student Presentations		
11:00 am – 12:00 pm	Developing a gene drive blueprint for invasive wasps, how could we, and should we?	**Josh Gilligan** *Department of Biochemistry*
	Exploiting genetic interactions to design therapeutic regimens for Mycobacterium tuberculosis	**Cassie Chapman** *Department of Microbiology*
	Male germline development in plants: developing CRISPR/Cas13 as a tool	**Baeli Spedding-Devereux** *Department of Biochemistry*
	Repurposing the type I-D CRISPR-Cas system as a programmable gene silencing tool	**Rakesh Banerjee** *Department of Pathology*
	Identification of lncRNAs involved in paclitaxel sensitisation in triple negative breast cancer	**Kaitlyn Tippett** *Department of Biochemistry*

Participants will require a laptop to participate fully in this workshop.

An End-to-End Solution for CRISPR Genome Editing Research

Joe Frangipane, Ph.D.
CRISPR Field Application Scientist, Integrated DNA Technologies

CRISPR-Cas gene editing is a robust system for precise disruption of genes and knock-in of novel genetic information. This method has broad applications in functional genomics research, synthetic biology, and translational medicine. Important considerations to maximize the success of CRISPR-based gene editing include the source and quality of guide RNA and nuclease, method of reagent delivery, design and stability of guide RNA and donor template, and nuclease fidelity. Reliable methods for simultaneous optimization of the gene editing systems while preventing genotoxic and cytotoxic effects are critical needs for the gene editing community. This seminar will present complete workflows for maximizing CRISPR-based genome editing in a variety of cell lines and primary cells while mitigating off-target editing and maintaining genome stability. Topics will include:

- End-to-end workflows for CRISPR genome editing, from gRNA design through NGS-based analysis of on- and-off target editing outcomes.

- Ribonucleoprotein-based delivery of CRISPR reagents to maximize efficiency, flexibility and convenience in genome editing experiments.
- Expanded offerings in gRNA libraries and novel innovations in Cas-based nucleases.
- Methods and reagents for improved knock-in rates via homology-dependent repair.
- The latest tools from IDT for gRNA and donor template design, as well as for CRISPR validation.

ELSC Discussion– St David Seminar Room D
Time: 9:00 am – 11:00 am
Zoom Link: https://otago.zoom.us/j/91353856437?pwd=aEF3Y0pxMWlsaXBrRmx6dVBETFJZUT09
Meeting ID: 913 5385 6437, Password: 065103

This session will be facilitated by Sara Filoche and Josephine Johnson who will be engaging attendees in a discussion of the National Academies of Sciences, Engineering, and Medicine report 'Using Population Descriptors in Genetics and Genomics Research'.

This is the link to the NASEM news release, which also includes links to the full report as well as various short documents and resources
https://www.nationalacademies.org/news/2023/03/researchers-need-to-rethink-and-justify-how-and-why-race-ethnicity-and-ancestry-labels-are-used-in-genetics-and-genomics-research-says-new-report

Workshops Block IV

eDNA Workshop– St David Seminar Room A
Time: 1:00 pm – 4:00 pm

This workshop will be facilitated by Eddy Dowle and will cover Statistical analysis of eDNA data. Using an example dataset attendees will go through some standard approaches used to explore eDNA data. All analysis will be done in R.

Participants will require a laptop with the latest version of R installed to participate fully in this workshop.

Retirement Special Session

This session will be held in the St David Lecture Theatre and foyer.
Time: 4:15 pm – 7:00 pm
Zoom Link: https://otago.zoom.us/j/8323949924?pwd=NitZS0VkYXRaN0RiL0Z4N2lDTGdHZz09
Meeting ID: 832 394 9924, Password: 750757

This is a special session to celebrate the career of Professor Iain Lamont as he moves into retirement.

	Retirement Special Session – Main St David Lecture Theatre	
4:15 pm – 4:30 pm	Welcome	Professor Peter Dearden *HOD, Department of Biochemistry* Associate Professor Megan Wilson *Director, Genetics Teaching Programme*
4:15 pm – 5:30 pm	Keynote	Professor Iain Lamont *Department of Biochemistry*
5:30 pm – 7:00 pm	Drinks and nibbles in the St David Lecture Theatre foyer	

Some nibbles will be provided and a bar will be open for drinks purchase (EFTPOS only, no cash).

Symposium

The main Symposium will be held in the St David Lecture Theatre.
Time: 9:00 am – 5:00 pm
Zoom Link: https://otago.zoom.us/j/8323949924?pwd=NitZS0VkYXRaN0RiL0Z4N2lDTGdHZz09
Meeting ID: 832 394 9924, Password: 750757

Symposium		
9:00 am - 9:10 am	Welcome and Opening	Peter Williamson *Rautaki Hononga / Kaitakawaenga (Māori Strategic Framework Facilitator)* Associate Professor Louise Bicknell *Co-Director Genetics Otago*
Session One		Chair:
9:10 am – 9:30 am	Shiny new toys: Knowledge structures and (in)equity in access to molecular technologies (Via Zoom)	Associate Professor Sara Filoche *Department for Obstetrics, Gynaecology and Women's Health, UOW*
9:30 am – 9:50 am	Modifiers of *BRCA1*-associated breast cancer risk and potential preventative strategies (Via Zoom)	Dr George Wiggins *Department of Pathology and Biomedical Science, UOC*
9:50 am – 10:05 am	Using a zebrafish stem cell model to understand the molecular basis of cohesinopathies	Anastasia Labudina *Department of Pathology*
10:05 am – 10:20 am	Influence of SNP rs1800795 on macrophage polarisation, colitis, and response to emerging therapies	Jildou van der Werf *Department of Pathology*
SciCom Award Presentations		Chair: Louise Bicknell
10:20 am – 10:30 am	The genetic code of the brushtail possum – *Video* A genetic analysis on the causes of truffle-like morphology-what makes a fungus decide to go round – *3MT Presentation* 8-Cell Clusters: Germline Progenitors in Honeybees- *Presentation*	Associate Professor Tim Hore et al., Finn Dobbie, Georgia Cullen
10:30 am – 11:00 am Break (SciCom Award Judging Open)		
Session Two		Chair: Caroline Stokes
11:00 am – 12:00 pm	He ira atua, he ira taiao, he ira tangata (Via Zoom)	Associate Professor Karyn Paringatai | Ngāti Porou *Te Temu | School of Māori, Pacific and Indigenous Studies*
12:00 pm – 12:15 pm	Single-cell RNA-Seq Reveals Candidate Synergistic Treatments for the Chemoprevention of Hereditary Diffuse Gastric Cancer	Kieran Redpath *Department of Biochemistry*
12:15 pm – 12:30 pm	Identification of lncRNAs involved in paclitaxel sensitisation in triple negative breast Cancer	Kaitlyn Tippett *Department of Biochemistry*
12:30 pm – 1:30 pm Lunch (poster judging)		

Session Three		Chair: Cassie Chapman
1:30 pm – 2:30 pm	Plant reproduction: the key to crop domestication	Associate Professor Lynette Brownfield *Department of Biochemistry*
2:30 pm – 2:45 pm	From islands to infectomes: the interplay between host taxonomy and environment on microbial diversity among birds across remote islands	Rebecca Grimwood *Department of Microbiology and Immunology*
2:45 pm – 3:00 pm	Bacteriophages suppress CRISPR–Cas immunity using RNA-based anti-CRISPRs	David Mayo Muñoz *Department of Microbiology and Immunology*
3:00 pm – 3:30 pm Break		
Session Four		Chair: Brooke Whitelaw
3:30 pm – 3:50 pm	Molecular mechanisms underlying resilience to climate change in the green lipped mussel (kuku)	Nathan Kenny | Te Ātiawa, Ngāi Tahu *Department of Biochemistry*
3:50 pm – 4:10 pm	Genomics Aotearoa - past, present and future	Professor Mik Black *Co-Director Genomics Aotearoa*
4:10 pm – 4:25 pm	Unravelling the Secrets of Chordate Whole-Body Regeneration using single-cell and ATAC sequencing	Associate Professor Megan Wilson *Department of Anatomy*
4:25 pm – 4:40 pm	CORSAIR-Dx: A CRISPR-Cas13a-based approach for environmental DNA biomonitoring of invasive species from water	Benjamín Durán-Vinet *Department of Anatomy*
4:45 pm – 5:00 pm	Awards Presentation	Louise Bicknell *Co-Director Genetics Otago*
5:00 pm Close		

Other Information

Catering and Sponsor Displays

The St David Lecture Theatre Foyer (Visitors Centre end) will be the location of all catering. This includes the catering supplied for workshops held in the G09 Teaching Lab (Biochemistry Building). Please note that lunch is not catered on the 14th or 15th of November and attendees will be required to source their own lunch during the break times. Sponsor displays will also be set up in this area, please take some time during the breaks to engage with our sponsors, who have kept this event free for you.

Posters

Posters will be on display in the St David Lecture Theatre Foyer (Visitors Centre end) for the duration of the event. Posters can be hung from 8:00 am on the 14th of November. If you are presenting a poster, you will be notified as to which of the three judging sessions you will be participating in. Please check carefully that you attend on the correct day, and are at your poster for the duration of the session.

Poster Judging Session I
Time: 12:30 pm – 1:00 pm, Tuesday 14th November
This judging session will include posters 1 – 6

Poster Judging Session II
Time: 12:30 pm – 1:00 pm, Wednesday 15th November
This judging session will include posters 7 – 12

Poster Judging Session III
Time: 1:00 pm – 1:30 pm, Thursday 16th November
This judging session will include posters 13– 18

Poster prizes will be awarded to the best Student and ECR posters as determined by the judges. These awards will be presented at the close of the Symposium, please try to attend this.

Awards

Awards will be presented by Genetics Otago Co-Director, Louise Bicknell at the conclusion of the Symposium. Awards to be presented are:

- The 2023 Genetics Otago Award
- Outstanding Peer Mentor Award
- Student Supervisor Award
- Student and ECR Publication Awards
- Student and ECR Poster Awards
- Science Communication Award (popular vote)

The Science Communication Award will be determined by popular vote. All entries will be presented during session one of the Symposium from 10:20 am on the 16th of November. Voting for this award is via Google form (https://forms.gle/XqvLcwZPvTtKvtk76) and will be open from 10:20 am – 11:00 am.

Abstracts
Keynote Speakers

Understanding a bacterial gene regulatory pathway – the importance of emerging technologies

Iain Lamont
Department of Biochemistry, University of Otago, Dunedin

Pseudomonas aeruginosa is an extremely problematic bacterial pathogen that causes a wide range of difficult-to-treat infections. During infection, the bacteria must obtain nutrients to be able to grow. A key nutrient, because of its low availability, is iron. To obtain iron the bacteria secrete iron-chelating compounds that scavenge iron from the infected host and deliver it into the bacterial cells. In this talk I will outline work carried out over a number of years that identified and characterised a sophisticated signal transduction pathway used by the bacteria to regulate production of iron-scavenging molecules. The talk will emphasise how the incorporation of new technologies enabled major steps forward in understanding of this biological system.

He ira atua, he ira taiao, he ira tangata

Karyn Paringatai,
Te Temu School of Māori, Pacific and Indigenous Studies, University of Otago, Dunedin

This presentation will look at the ongoing influence of atua Māori on people and place through an investigation of various narratives as precursors of genetic research as it is commonly viewed today. The University of Otago has an aspiration to be more responsive to iwi, hapū and whānau through greater collaborative research partnerships and a more deliberate incorporation of mātauranga Māori in to our curriculum. This presentation will also provide examples of how incremental, but meaningful, changes can make this happen.

Plant reproduction: the key to crop domestication

Lynette Brownfield
Department of Biochemistry, University of Otago, Dunedin

Domestication has transformed wild plants into the high-performing, high-yielding crop plants of today. Notably, only a small fraction of flowering plants have undergone this transformation. Given the need for agriculture to become more sustainable and adapt to a changing environment, there is interest in further plant domestication. In this talk I will discuss reproductive strategy as a key characteristic for plant domestication and breeding. Unlike many plants, the world's main crops plants can both inbreed, fixing beneficial or purging deleterious alleles, and outcross, introducing new genetic material. This ability to inbreeding and outcrossing is believed to have been key in domestication, and also underlies current breeding strategies, but is not possible in many plants. Understanding and overcoming the mechanisms that prevent plants from both inbreeding and outcrossing, is therefore a critical step in expanding the range of crop plants and improving the performance of others. In this talk I will describe our work on barriers to inbreeding in two plants that are critical to the New Zealand economy but are still poorly domesticated; ryegrass and kiwifruit.

Shiny new toys: Knowledge structures and (in)equity in access to molecular technologies

Sara Filoche[1], *Fiona Cram*[3], *Peter Stone*[2], *Sondra Bacharach*[4], *Angela Bear*[5], *Christina Buchanan*[6], *and Kevin Dew*[7]

[1]*Department of Obstetrics, Gynaecology and Women's Health and Department of Pathology and Molecular Medicine, University of Otago, Wellington*

[2]*Department of Obstetrics and Gynaecology, University of Auckland, Auckland*

[3]*Katoa Ltd, Auckland*

[4]*School of History, Philosophy, Political Science and International Relations, Victoria University, Wellington*

[5]*Christchurch Obstetric Associates, Christchurch*

[6]*Faculty of Medical and Health Sciences, The University of Auckland*

[7]*School of Social and Cultural Studies, Victoria University, Wellington*

Advances in molecular technologies have the potential to help remedy health inequities through earlier detection and prevention; if, however, their delivery and uptake (and therefore any benefits associated with such testing) are not more carefully considered, there is a very real risk that existing inequities in access and use will be further exacerbated. We argue this risk relates to the way that information and knowledge about the technology is both acquired and shared, or not, between health practitioners and their patients. Our work falls under the umbrella of Fricker's notion that harm can be done to another person in their capacity as a knower. When someone is not perceived as having the capacity to understand information by another, this affects how, what, and if information is shared between them – in other words, they experience informational prejudice. We take the position that both practitioners and patients are vulnerable to epistemic injustice (and thus experience informational prejudice) owing to prevalent negative stereotypes and certain structural features of contemporary healthcare practice. Our journey to this position comes from our experiences in our respective fields of indigenous and health research, epistemology and medical sociology – where, in reference to new molecular technology, we have frequently heard comments such as "oh well *they* won't understand" or "I don't have time to explain *it* to *them*". Where the *they* and *them* can be in reference to either and/or both certain practitioners and patients. Such biases and position are bound to have an impact on meeting our health care ideal of enabling people to make an informed choice in their decision to undergo testing or not. Using prenatal screening for chromosomal conditions we explore this further through a combination of approaches to better understand how knowledge structures operate information dissemination.

Modifiers of *BRCA1*-associated breast cancer risk and Potential Preventative Strategies

George Wiggins

Department of Pathology and Biomedical Science, University of Otago, Christchurch

Women carrying pathogenic *BRCA1* variants have approximately 72% lifetime risk of developing breast cancer. However, the age of onset of breast cancer can vary significantly between *BRCA1* pathogenic variant carriers suggesting additional factors, including genetic, influence disease penetrance. Through an international study with the Consortium of Investigators of Modifiers of *BRCA1/2* (CIMBA), we found that 1) *BRCA1* variant type (e.g. frameshift and deletions) and 2) copy number variants (at other loci) modify the risk of breast and ovarian cancer. Compared to pathogenic single nucleotide variants (SNVs), large genomic rearrangements in *BRCA1* are associated with

greater risk (OR=1.21, 95% CI 1.09-1.35) of breast cancer. An emerging process termed transcriptional adaptation has been described as a potential mechanism of genetic compensation, whereby the deleterious effect of genetic variants are tempered. Transcriptional adaptation is the ability of mutation containing RNA to modulate the expression of compensatory genes (including the wild-type allele). We also identified deletion overlapping *SULT1A1* that reduced the risk of developing breast cancer in women carrier high-risk *BRCA1* variants (RR=0.73, 95% CI 0.59-0.91). Using MCF-7 cells with and without a pathogenic *BRCA1* variant, we are investigating 1) whether transcriptional adaptation may modify *BRCA1* associated risk of cancer; and 2) the potential of SULT1A1 inhibition as a risk-reduction therapy for women carrying a *BRCA1* pathogenic variant. Our *in vitro* analyses have shown that the knockdown of *SULT1A1* reduces proliferation and increases DNA damage in pathogenic variant carrying cells compared to isogenic wild-type cells. These results will provide a better understanding into the mechanisms that underlie the risk of developing *BRCA1*-associated breast cancer and insight into potential prevention.

Molecular mechanisms underlying resilience to climate change in the green lipped mussel (kuku)

Nathan Kenny
Department of Biochemistry, University of Otago, Dunedin

Molluscs such as the green-lipped mussel or kuku (*Perna canaliculus*) are vital to our ecosystems and taonga (treasured species) of cultural and economic importance. Climate changes, including temperature extremes and ocean acidification, threaten this species, but some individuals are more resilient to these issues than others. The source of this resilience is unknown, but differences in early development are well implicated, and a degree of heritability has been noted for this trait. It is thought that slight heterochronic shifts and changes in gene expression allow differences in phenotype, conferring some individuals with an ability to survive in these conditions. Using cutting-edge approaches, we aim to pinpoint the key differences exhibited by resilient kuku. We have assembled the genome of this species to an excellent standard, in partnership with mana moana as kaitiaki. We are now leveraging this resource to determine the molecular mechanisms underlying resilience, using a range of approaches including single-cell RNA sequencing (SPLiT-seq), RNA *in situ* hybridization chain reaction, long read mapping and miRNA sequencing to approach this problem from a number of angles.

Genomics Aotearoa - past, present and future

Mik Black
Genomics Aotearoa and Department of Biochemistry, University of Otago, Dunedin

Genomics Aotearoa (GA) is a national collaboration involving ten universities and crown research institutes, led by the University of Otago, and funded by the Ministry of Business, Innovation and Employment to deliver infrastructure to support and grow genomics research in Aotearoa New Zealand. For the past five years, GA has been "building by doing" - developing and supporting genomics infrastructure by funding exemplar projects across our three Research Theme areas: environment, health, and primary production, as well as delivering bioinformatics and computational training to the genomics community, developing a national genomic data repository to support Māori data sovereignty, and building capability and capacity through an Indigenous Genomics Platform which funds Māori-led research projects. This presentation will provide an overview of GA's ongoing

work across these areas, as well as discussing our plans for the future as we head into the final year of our current funding cycle.

Using a zebrafish stem cell model to understand the molecular basis of cohesinopathies

AA Labudina[1], G Gimenez[1], M Meier[1], S Ketharnathan[2], J Antony[1], JA Horsfield[1].
[1]*Department of Pathology, Dunedin School of Medicine, University of Otago, Dunedin.*
[2]*Children's Hospital of Eastern Ontario (CHEO), Ottawa, Canada.*

Cohesinopathies are multisystem disorders caused by germline mutations in cohesin subunit genes. Cohesin has roles in both cell division and gene expression. Cohesin comprises four core subunits: Smc1a, Smc3, Rad21 and Stag1 or 2, which form a ring-shaped complex. Mutations in genes encoding Smc1a, Smc3 and Rad21 are homozygous lethal because they disrupt formation of the cohesin ring and block cell division. Mutations in *stag* genes are viable in zebrafish because the encoded proteins can functionally substitute for each other in the cohesin complex. Here we take advantage of zebrafish carrying mutations in *rad21* and *stag2* to better understand the molecular pathology of cohesinopathies using an embryonic tailbud model. Neuromesodermal progenitors (NMPs) are bipotent tailbud cells that differentiate into neuroectoderm or mesoderm under the control of Wnt signalling. We performed RNA-sequencing of wild type, *stag2* and *rad21* mutant tailbuds to investigate the consequences for NMP differentiation. Transcription dysregulation in *rad21* mutants reflects cell cycle disruption, and *rad21* mutants cannot induce mesoderm due to *sox2* and *tbxta* dysregulation in NMPs. In contrast, *stag2* mutants compensate for the lack of mesodermal induction by upregulating Wnt signalling and transdifferentiation of notochord progenitors, resulting in a narrower notochord. External stimulation of Wnt signalling rescues transcriptional signatures in *stag2*, but not in *rad21* mutants. Our results show that developmental outcomes depend on which cohesin subunit is mutated, implying that individual cohesin subunits modulate how cohesin functions in development.

Influence of SNP rs1800795 on macrophage polarisation, colitis, and response to emerging therapies

van der Werf, Jildou[1], Chin, Chu Vin (Alice)[1], Dunbier, Anita[2], Fleming, Nicholas[1]
[1]*Department of Pathology, University of Otago*
[2]*Department of Biochemistry, University of Otago*

Variation in immune system function can aid progression of colorectal cancer (CRC) and inflammatory bowel disease (IBD). Likewise, although appropriate drug treatments can severely improve life quality, the responsiveness amongst individuals varies greatly. Current research is focused on targeted immune therapy drugs, including antagonists to interleukin 6 (IL-6), a cytokine that contributes to functioning of macrophages (Mφ). The pro-inflammatory (M1) state of Mφ is associated with IBD, whilst the anti-inflammatory (M2) state promotes cancer progression. A single nucleotide polymorphism (SNP) rs1800795 in the IL6 gene is responsible for a significant proportion of variance in our IL-6 expression, and we propose it alters the onset of IBD and CRC by altering Mφ polarisation, and in turn, influencing our responsiveness to drugs targeting IL-6 trans-signalling. *In vitro* polarisation of human monocytes with the SNP showed higher M1 marker genes for the CC, whilst GG had higher M2 marker genes. Differentiated bone marrow derived Mφ from mice genetically modified for the SNP show similar results. When inducing acute colitis in these mice, CC animals developed more severe IBD symptoms, together with a greater weight loss, increased incidence of bleeding, and higher levels of IL-6 expressed. Remarkably, these mice responded

better to IL-6 trans-signalling blockade, when compared to their GG siblings, substantiating the SNP as a predictor for responsiveness to IBD treatment. The SNP may provide a reliable biomarker for personalising and facilitating the use of emerging drug treatments.

Single-cell RNA-Seq Reveals Candidate Synergistic Treatments for the Chemoprevention of Hereditary Diffuse Gastric Cancer

Kieran Redpath[1,2], Conor Vaessen[1,2], Lyvianne Decourtye-Espiard[1,2], Emily Schulpen[1,2], Tanis Godwin[1,2], Nicola Bougen-Zhukov[1,2], Kate McElroy[1,2], Mik Black[1,2] & Parry Guilford[1,2]
[1]*Centre for Translational Cancer Research, University of Otago*
[2]*Department of Biochemistry, University of Otago*

Gastric cancer is the third most lethal and fifth most common cancer worldwide. Diffuse gastric cancer (DGC) often develops due to inherited or somatic mutation of the tumour suppressor gene E-cadherin (*CDH1*), which is found in some Māori families in New Zealand. Germline inactivation of *CDH1* causes hereditary DGC (HDGC) with up to 70% penetrance. Dasatinib, an ATP-competitive tyrosine kinase inhibitor, has recently been identified as a potential therapeutic candidate for the chemoprevention of HDGC. Dasatinib targets pathways that are upregulated in *CDH1*-null cells, preferentially damaging the tumour. To improve this targeted therapy, we aim to identify synergistic partners of dasatinib – those compounds which provide a greater than additive effect when combined with dasatinib. Transcriptional changes in response to treatment can reveal drug targets (genes and pathways) that may be involved in dasatinib resistance (i.e., that are expressed in the remaining tumour cells after treatment). The higher resolution view afforded by single-cell mRNA-Seq can also reveal more subtle changes when compared to traditional "bulk" sequencing data. To this end, we treated a *Cdh1*/*Trp*53 knockout HDGC mouse model with dasatinib. Mouse stomachs were then extracted for single-cell isolation (using the 10X Genomics Gene Expression Flex kit) and mRNA sequencing (on an Illumina NextSeq 2000 with a P3 chip) for comparison to control (untreated) mice. Clustering and annotation of reads on a per cell basis revealed gastric epithelial cell types (where *CDH1-/-* HDGC develops) found across all samples: foveolar, pyloric gland/neck, and parietal cells. Differential expression analysis between grouped epithelial cell types in drugged and untreated samples revealed over 400 genes differentially regulated in response to dasatinib. Reactome pathway enrichment analysis revealed a number of clear signatures of activity (groups of Reactome pathways with related functions) that were altered between treatment and control groups. Drugs that target these signatures are likely to provide synergy with dasatinib in HDGC, likely by countering emerging dasatinib-resistant cell populations. Signatures of particular note are RHO GTPase signalling (consistent with preliminary analyses in cell line models) and innate immune response, supporting the use of broad-spectrum RHO inhibitors and immunotherapies alongside dasatinib in the treatment and prevention of HDGC.

Identification of lncRNAs involved in paclitaxel sensitisation in triple negative breast cancer

Kaitlyn M. Tippett[1] and Sarah D. Diermeier[1,2].
[1]*Department of Biochemistry, University of Otago, Dunedin*
[2]*Amaroq Therapeutics, Dunedin*

Triple-negative breast cancer (TNBC) is an aggressive subtype of breast cancer, making up 15-20% of breast cancer cases. It is characterised by being oestrogen receptor (ER), progesterone receptor (PR) and human epidermal growth factor 2 (HER2) negative. TNBC is

a highly heterogeneous disease, resulting in the standard treatment regimen for TNBC patients being combinations of chemotherapy, radiation therapy and surgery. Developing resistance to chemotherapies is a major concern for TNBC patients and is the main cause of of treatment failure, particularly in metastatic disease. Identifying novel therapeutic targets to help sensitise cancerous cells to chemotherapies may be able to overcome resistance and improve patient outcome. Long non-coding RNAs (lncRNAs) are defined as transcripts longer than 200 nt and lacking a significant open reading frame. LncRNAs have huge potential to be used as cancer therapeutic targets. They are commonly dysregulated in cancer and have been shown to have roles in tumorigenesis and drug resistance in numerous cancers, including breast cancer. All these features make lncRNAs desirable to investigate their role in chemotherapy sensitisation which is poorly understood in TNBC. We are particularly interested in targeting lncRNAs to enhance chemotherapeutic performance, to prevent chemoresistance arising. Here, we used a CRISPR screen to identify lncRNAs that can sensitise TNBC cells to paclitaxel. Time points were collected at 0, 6, 14 and 20 cell doublings for paclitaxel and vehicle control groups. Future experiments include validation of the identified sensitiser lncRNAs using CRISPRi loss-of-function models and analysis of patient data to assess their clinical significance in TNBC patients.

From islands to infectomes: the interplay between host taxonomy and environment on microbial diversity among birds across remote islands

R. M. Grimwood[1], J. Welch[2], G. Taylor[2], T. Emmitt[2], T. Thurley[2], T. Makan[2], L. Lim[3], J. Dubrulle[1], K. McInnes[2], E. C. Holmes[3], J. L. Geoghegan[1,4]
[1]*Department of Microbiology and Immunology, University of Otago, Dunedin,*
[2]*Department of Conservation/Te Papa Atawhai*
[3]*University of Sydney, Sydney, NSW, Australia*
[4]*Institute of Environmental Science and Research, Wellington*

Accelerating biodiversity loss necessitates urgent pathogen monitoring in vulnerable species. With a third of New Zealand's avifauna considered at risk of extinction, a greater understanding of factors influencing pathogen transmission in these island ecosystems is needed. We used metatranscriptomics to study the infectomes of seven bird species, including the critically endangered black robin (*Petroica traversi*), on the remote Chatham Islands archipelago, New Zealand. Virome characterisation unveiled 21 likely novel avian viruses across nine viral families. Black robins harboured viruses from the *Flaviviridae*, *Herpesviridae*, and *Picornavirales*, while introduced starlings (*Sturnus vulgaris*) and migratory seabirds (Procellariiformes) carried viruses from six additional viral families. We identified cross-species transmission of a novel *passeri*-like virus between native (black robins and grey-backed petrels) and introduced (starlings) birds, broadening our understanding of the role of introduced and migratory birds as potential vectors of disease that may impact threatened island-endemic species. Additionally, we detected bacterial genera, apicomplexan parasites as well as a novel megrivirus linked to disease outbreaks in other native New Zealand birds. Notably, host taxonomy (order) and sampling location (island) significantly influenced differences in avian microbiomes, underlining the wider interplay between host ecology, microbial diversity and transmission dynamics.

Bacteriophages suppress CRISPR–Cas immunity using RNA-based anti-CRISPRs

Sarah Camara-Wilpert[1], David Mayo-Muñoz[2,3,4], Jakob Russel[1], Robert D. Fagerlund[2,3,4,5], Jonas S. Madsen[1], Peter C. Fineran[2,3,4,5], Søren J. Sørensen[1] & Rafael Pinilla-Redondo[1,2]
[1]*Section of Microbiology, University of Copenhagen, Copenhagen, Denmark*

[2]Department of Microbiology and Immunology, University of Otago, Dunedin
[3]Genetics Otago, University of Otago, Dunedin
[4]Maurice Wilkins Centre for Molecular Biodiscovery, University of Otago, Dunedin
[5]Bioprotection Aotearoa, University of Otago, Dunedin, New Zealand

Many bacteria use CRISPR–Cas systems to combat mobile genetic elements, such as bacteriophages and plasmids. In turn, these invasive elements have evolved anti- CRISPR proteins to block host immunity. Here we unveil a distinct type of CRISPR– Cas Inhibition strategy that is based on small non-coding RNA anti-CRISPRs (Racrs). Racrs mimic the repeats found in CRISPR arrays and are encoded in viral genomes as solitary repeat units. We show that a prophage-encoded Racr strongly inhibits the type I-F CRISPR–Cas system by interacting specifically with Cas6f and Cas7f, resulting in the formation of an aberrant Cas subcomplex. We identified Racr candidates for almost all CRISPR–Cas types encoded by a diverse range of viruses and plasmids, often in the genetic context of other anti-CRISPR genes. Functional testing of nine candidates spanning the two CRISPR–Cas classes confirmed their strong immune inhibitory function. Our results demonstrate that molecular mimicry of CRISPR repeats is a widespread anti-CRISPR strategy, which opens the door to potential biotechnological applications.

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Unravelling the Secrets of Chordate Whole-Body Regeneration using single-cell and ATAC sequencing

Berivan Temiz, Michael Meier and Megan J. Wilson.
Department of Anatomy, University of Otago.

Botrylloides diegensis, a marine chordate, displays a remarkable ability for whole-body regeneration (WBR) by regenerating an entire body system from its vascular network. Our research encompasses gene and pathway characterisation, transcriptome profiling, and haematological analyses during various stages of regeneration. Recently, we have used single-cell RNA sequencing (sc-RNA-seq) of mature colonies and multiple WBR stages and ATAC-sequencing to identify gene regulatory elements to gain new insights into this chordate model of WBR. Notably, we observed the emergence of large transient cell populations exclusively during the early stages of WBR. Although lacking distinct highly expressed markers, sub-clustering revealed shared molecular signatures with committed cell clusters, suggesting orchestrated differentiation processes. We identified *SoxC* as a pivotal stem cell marker, exhibiting robust expression within aggregates of stem-like cells, regeneration vesicle-forming cells, and cells initiating organogenesis. Our cell trajectory analyses consistently depict a trajectory from *SoxC*+ cell populations through transient states towards more specialized cell lineages. Our findings collectively highlight the remarkable plasticity inherent in *B. diegensis* WBR.

CORSAIR-Dx: A CRISPR-Cas13a-based approach for environmental DNA biomonitoring of invasive species from water

Benjamín Durán-Vinet[1], Gert-Jan Jeunen[2], Jo-Ann L. Stanton[1], Anastasija Zaiko[3], Xavier Pochon[4,5], Neil Gemmell [1,].*
[1]Department of Anatomy, School of Biomedical Sciences, University of Otago, Dunedin
[2]Department of Marine Sciences, University of Otago, Dunedin
[3]Sequench Ltd, Nelson
[4]Biosecurity Group, Cawthron Institute, Nelson

[5]*Institute of Marine Science, University of Auckland, Auckland*

The demand for cost-effective, portable, highly specific tools to monitor invasive species in terrestrial and marine ecosystems using environmental DNA (eDNA) is increasing. Molecular-based approaches that have proven popular include quantitative polymerase chain reaction (qPCR). However, PCR-based approaches are prone to environmental inhibitors, contamination, and are less portable. Novel methods using the CRISPR-Cas toolbox (Clustered regularly interspaced palindromic repeats/CRISPR-associated proteins) coupled with isothermal pre-amplification methods (e.g., recombinase polymerase amplification) have unveiled new possibilities for environmental biomonitoring in terms of portability and quality. Therefore, we have focused on piloting the use of the bacterium *Leptotrichia wadeii* Cas13a (LwaCas13a) system and ADAPT (Activity-informed Design with All-inclusive Patrolling of Targets) machine learning to facilitate the design of highly active diagnostic and species-specific CRISPR RNA (crRNAs) sets for biomonitoring applications using cytochrome c oxidase subunit I (COI) gene. Specifically, we aimed to detect the invasive Mediterranean fanworm (*Sabella spallanzanii*) and Asian seaweed (*Undaria pinnatifida*) as proof of concept. Our preliminary findings indicate the necessity for specialised modelling to forecast the LwaCas13a diagnostic rate performance for environmental deployment accurately. Nevertheless, the current model provides a comprehensive and rapid pipeline to create highly active crRNA sets for eDNA biomonitoring. This fusion of CRISPR-based diagnostics and machine learning for environmental applications has been named CORSAIR: CRISPR-Cas-based biomonitoring of environmental biological threats. This proof-of-concept may represent an innovative step towards robust, swift, cost-effective, and species-specific environmental biomonitoring.

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1. Upstream open reading frames in the Plantae kingdom

Alexandra Gibbon[1], Richard Macknigh[1], and Chun Shen Lim[1]
[1]*Department of Biochemistry, University of Otago, Dunedin*

Producing proteins comes at an energy cost and changes in both the internal and external environment require organisms to make rapid and efficient alterations to their proteome. Upstream open reading frames (uORFs) provide a system allowing organisms to regulate translation of key proteins, putting translation of mRNA 'on hold', until the protein is required. uORFs are present in the 5' UTR and regulate translation of the downstream main open reading frame (mORF). In plants this can provide a fast response to changing conditions. Ribosome stalling followed by re-initiation or leaky scanning by the PIC (pre-initiation complex) leads to decreased translation of the mORF. A change in conditions can then lead to increased mORF translation via either leaky scanning, in which the PIC recognises the mORF start as opposed to the uORF, or re-initiation in which a partially disassociated ribosomal complex re-associates and translation continues. Many molecular features such initiation and termination codon contexts, RNA structure and other motifs present in the 5' UTR are thought to contribute to uORF regulation. Microtubule associated stress protein one (MASP1) and CBL-interacting kinase 6 (CIPK6) are both implicated in stress response in *Arabidopsis*. Under normal conditions MASP1 protein levels are low, whereas under drought stress high protein levels are observed. There are similar observations of CIPK6. It has been proposed that these changes in protein translation are due to uORFs present in the 5' UTR of both genes. However, the mechanism of action of these uORFs is yet to be formally characterised. This project aims to analyse and characterise these uORFs as well as investigating general mechanisms of uORF regulation and uORF features across the plant kingdom.

2. Understanding Active DNA Demethylation Specificity and Transcriptional Consequences in Stem Cells

Cassandra R. Glanfield[1], Claudia I. Davies[1], Mirunalini Ravichandran[2,3], Dominik Rafalski[4], Oscar Ortega-Recalde[1], Andrew H. Wang[1], Matthias Bochtler[4,], Tomasz P. Jurkowski[3,5,*] & Timothy A. Hore[1,*]*
[1]*University of Otago, Department of Anatomy, Dunedin*
[2]*University of California, Department of Anatomy, San Francisco, USA*
[3]*University of Stuttgart, Department of Biochemistry, Stuttgart, Germany*
[4]*International Institute of Molecular and Cell Biology in Warsaw (IIMCB), Warsaw, Poland*
[5]*Cardiff University, School of Biosciences, Cardiff, Wales*

The presence and regulation of cytosine methylation across the genome is essential for vertebrate development and lineage commitment of cells. A key player in the regulation of DNA methylation is the Ten-Eleven-Translocation (TET) hydroxylases which catalyse the active removal of DNA methylation and enables developmental potency both *in vivo* and during the generation of induced pluripotent stem cells. However, the mechanism by which TET is targeted to specific sites of DNA methylation, and the resulting transcriptional consequences, remained unclear. Our recent work with collaborators has shown that the catalytic domain of mammalian TET enzymes favours specific CpGs containing hexamers with up to 250-fold preference *in vitro*. To test this inherent sequence specificity of TET, the

TET catalytic activity was rescued by doxycycline-induced overexpression in a TET-triple knockout cell line. The demethylation kinetics were calculated for individual CG-containing hexamer sites using whole-genome bisulfite sequencing (WGBS) data and validated by APOBEC-coupled epigenetic sequencing (ACE-seq). TET was shown to have a strong preference for specific CpG sites based on the flanking dinucleotides, occurring in a strand-specific manner. These favoured sites represent DNA sites bound by methylation-sensitive immediate-early genes (IEG) such as MYC and JUN/FOS. Alternately, sites least favoured by TET include octamer-binding transcription factor 4 (OCT4). While these favourable and unfavourable TET-targeted sites highlight the functional importance of CpG sites being able to lose methylation quickly or to hold onto methylation, respectively, we are yet to demonstrate immediate transcriptional effects. Ongoing experimental work using RNA-sequencing is testing this hypothesis.

3. Male-specific DNA aging and building the Androgen Clock

Sugrue, V.J.[1], Zoller, J.A.[2], Narayan, P.[3], Lu, A.T.[4], Ortega-Recalde, O.J.[1], Grant, M.J.[3], Bawden, C.S.[5], Rudiger, S.R.[5], Haghani, A.[4], Bond, D.M.[1], Hore, R.R.[6], Sears, K.E.[7], Wang, N.[8], Yang, X.W.[8,9], Snell, R.G.[3], Campbell, R.[1], Garratt, M.[1], Horvath, S.[4], Hore, T.A.[1]*.*

[1]Department of Anatomy, University of Otago, Dunedin
[2]Department of Biostatistics, Fielding School of Public Health, UCLA, Los Angeles, CA, USA
[3]Applied Translational Genetics Group, School of Biological Sciences, Centre for Brain Research, The University of Auckland, Auckland
[4]Department of Human Genetics, David Geffen School of Medicine, UCLA, Los Angeles, CA, USA
[5]Livestock and Farming Systems, South Australian Research and Development Institute, Roseworthy, SA, Australia
[6]Blackstone Hill Station, Becks, Omakau
[7]Department of Ecology and Evolutionary Biology, UCLA, Los Angeles, CA, USA
[8]Center for Neurobehavioral Genetics, Semel Institute for Neuroscience and Human Behavior, UCLA, Los Angeles, CA, USA;
[9]Department of Psychiatry and Biobehavioral Sciences, David Geffen School of Medicine at UCLA, Los Angeles, CA, USA.

In mammals, females generally live longer than males, significantly so for sheep[1]. Nevertheless, the mechanisms underpinning sex-dependent longevity are currently unclear. Epigenetic clocks are powerful biological biomarkers capable of precisely estimating chronological age and identifying novel factors influencing the aging rate using only DNA methylation data[2]. We developed the first epigenetic clock for domesticated sheep (Ovis aries), capable of predicting chronological age with a median absolute error of 5.1 months using DNA methylation data at 185 CpG sites throughout the genome[3]. We have discovered that castrated male sheep have a decelerated aging rate compared to intact males, mediated at least in part by the removal of androgens. Furthermore, we identified several androgensensitive CpG dinucleotides that become progressively hypomethylated with age in intact males, but remain stable in castrated males and females. Comparable sex-specific methylation differences in MKLN1 also exist in bat skin and a range of mouse tissues that have high androgen receptor expression, indicating it may drive androgen-dependent hypomethylation in divergent mammalian species. In characterising these sites, we identify biologically plausible mechanisms explaining how androgens drive male-accelerated aging in sheep and other mammals. Using a single androgen-sensitive CpG site in MKLN1, we have developed a rapid and effective tool to measure the period of androgen exposure in sheep and mice, with median absolute errors of 4.3 and 1.4 months respectively. We term this predictor the Androgen Clock and show its ‘tick’ can be accelerated beyond that in normal male mice by supplementing females with dihydrotestosterone. The Androgen Clock provides a rare

opportunity to warp the 'ticking rate' of an epigenetic time predictor in a way that does not affect cellular viability, and could also aid in diagnosis of conditions characterised by longterm elevated androgens, such as polycystic ovary syndrome (PCOS).

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4. A genetic analysis of the causes of truffle-like morphology *-what makes a fungus go round*

Finn LW Dobbie[1,2], David A Orlovich[3], Chris M Brown[1, 2]
[1]Department of Biochemistry, University of Otago, Dunedin
[2]Genetics Otago, University of Otago, Dunedin
[3]Department of Botany, University of Otago, Dunedin

Truffle-like fungi are one of the worlds most well-known mysteries. We know that they are all over the world, they came from mushrooms but live for much longer, they have a distinctive round appearance and have multiple unique secondary metabolites. What we don't know is how these changes came to be. By investigating the changes from a mushroom to a truffle-like fungus I hope to identify genes and developmental pathways that will aid both our understanding of fungal development as a whole, as well as identify useful genes/pathways for the mushroom industry to use.

5. Genotype-first approach to identify associations between *CDH1* germline variants and cancer phenotypes: a multicentre study by the European Reference Network on Genetic Tumour Risk Syndromes

J. Garcia-Pelaez[1,2,3,4,5], R. Barbosa-Matos[#1,2,4,5,6], S. Lobo[#1,2,4,5,6], A. Dias[#1,2,4], L. Garrido[7], S. Castedo[1,2,3,4,5,7], S. Sousa[1,2,4], H. Pinheiro[1,2,4,8], L. Sousa[1,2,4,9], R. Monteiro[1,2], J. J. Maqueda[1,2,10], S. Fernandes[3], F. Carneiro[1,2,3,4,5,7], N. Pinto[1,2,4,11], C. Lemos[1,4,6,12], C. Pinto[4,13], M. R Teixeira[4,5,6,13], S. Aretz[5,14,15], S. Bajalica-Lagercrantz[5,16,17,18], J. Balmaña[5,19], A. Blatnik[5,20], P. R Benusiglio[21], M. Blanluet[22], V. Bours[5,23,24], H. Brems[25], J. Brunet[5,26], D. Calistri[27], G. Capellá[5,28,29], S. Carrera[30], C. Colas[5,22], K. Dahan[31], R. de Putter[5,32], C. Desseignés[21], E. Domínguez-Garrido[33], C. Egas[34], D. G. Evans[35,36], D. Feret[31], E. Fewings[37], R.C. Fitzgerald[38], F. Coulet[21], M. Garcia-Barcina[39], M. Genuardi[5,40,41], L. Golmard[22], K. Hackmann[42,43,44,45], H. Hanson[46], E. Holinski-Feder[5,47,48], R. Hüneburg[5,15,49], M. Krajc[5,20], K. Lagerstedt-Robinson[5,16,17], C. Lázaro[5,28,29], M. J. L. Ligtenberg[5,50,51,52], C. Martínez-Bouzas[53], S. Merino[39], G. Michils[25], S. Novaković[54], A. Patiño-García[55], G. N. Ranzani[56], E. Schröck[5,42,43,44,45,57], I. Silva[58], C. Silveira[58], J. L. Soto[59], I. Spier[5,14,15], V. Steinke-Lange[5,47,48], G. Tedaldi[27], M. I. Tejada[53], E. R. Woodward[35,36], M. Tischkowitz[37], N. Hoogerbrugge[5,50,52], C. Oliveira[1,2,3,4,5]
[#]Equal contribution; [1]Instituto de Investigação e Inovação em Saúde (i3S), University of Porto, Porto, Portugal; [2]Instituto de Patologia e Imunologia Molecular da Universidade do Porto, Porto, Portugal; [3]Faculty of Medicine, University of Porto, Porto, Portugal; [4]Porto Comprehensive Cancer Center Raquel Seruca, Porto, Portugal; [5]European Reference Network on Genetic Tumour Risk Syndromes (ERN GENTURIS); [6]Institute of Biomedical Sciences Abel Salazar, University of Porto, Porto, Portugal; [7]Centro Hospitalar Universitário São João, Porto, Portugal; [8]Serviço de Medicina Interna, Centro Hospitalar Tâmega e Sousa, Penafiel, Portugal; [9]Escola de Economia e Gestão, Universidade do Minho, Braga, Portugal; [10]Bioinf2Bio, Porto, Portugal; [11]Center of Mathematics, University of Porto, Porto, Portugal; [12]Institute for Molecular and Cell Biology, University of Porto, Porto, Portugal; [13]Department of Laboratory Genetics, Portuguese Oncology Institute of Porto, Porto, Portugal; [14]Institute of Human Genetics, Medical Faculty, University of Bonn, Bonn, Germany; [15]National Center for Hereditary Tumor Syndromes, University Hospital Bonn, Bonn, Germany; [16]Department of Oncology Pathology, Karolinska Institutet, Stockholm, Sweden; [17]Department of Clinical Genetics, Cancer Genetic Unit, Karolinska University Hospital Solna, Stockholm, Sweden; [18]Cancer Theme, Karolinska University Hospital Solna, Stockholm, Sweden; [19]Hospital Vall d'Hebron, Universitat Autonoma de Barcelona, Barcelona, Spain; [20]Department of Clinical Cancer Genetics, Institute of Oncology Ljubljana, Ljubljana, Slovenia; [21]Medical Genetics Department, Pitié-Salpêtrière Hospital, AP-HP and Sorbonne University, Paris, France; [22]Service de Génétique Oncologique, Institut Curie, Paris, France; [23]Laboratory of Human Genetics, GIGA Institute, University of Liège, Liège, Belgium; [24]Center of Genetics, University Hospital,

Liège, Belgium; [25]Department of Human Genetics, University of Leuven, Leuven, Belgium; [26]Hereditary Cancer Programme, Catalan Institute of Oncology, Bellvitge Institute for Biomedical Research and Girona Biomedical Research Institute, Barcelona-Girona, Spain; [27]Laboratorio di Bioscienze, IRCCS Istituto Romagnolo per lo Studio dei Tumori (IRST) "Dino Amadori", Meldola, Italy; [28] Hereditary Cancer Program, Catalan Institute of Oncology, Bellvitge Institute for Biomedical Research, Barcelona, Spain; [29]Centro de Investigación Biomédica en Red de Cáncer (CIBERONC), Madrid, Spain; [30]Oncology Service, Biocruces Bizkaia Health Research Institute, Cruces University Hospital, Cruces, Barakaldo, Bizkaia, Spain; [31]Center of Human Genetics, IPG, Gosselies, Belgium; [32]Clinical Genetics Department, University Hospital of Ghent, Ghent, Belgium; [33]Molecular Diagnostics Laboratory, Fundación Rioja Salud, Logroño, Spain; [34]CNC—Center for Neuroscience and Cell Biology, University of Coimbra, Coimbra, Portugal; [35]Division of Evolution and Genomic Sciences, University of Manchester, Manchester, UK; [36]Manchester Centre for Genomic Medicine, Manchester, UK; [37]Department of Medical Genetics, National Institute for Health Research Cambridge Biomedical Research Centre, University of Cambridge, Cambridge, UK; [38]Early Cancer Institute, University of Cambridge, Cambridge, UK; [39]Genetics Unit, Biocruces Bizkaia Health Research Institute, Basurto University Hospital, Bilbao, Bizkaia, Spain; [40]Sezione di Medicina Genomica, Dipartimento di Scienze della Vita e Salute Pubblica, Università Cattolica del Sacro Cuore, Rome, Italy; [41]UOC Genetica Medica, Dipartimento di Scienze di Laboratorio e Infettivologiche, Fondazione Policlinico Universitario A Gemelli IRCCS, Rome, Italy; [42]Institute for Clinical Genetics, University Hospital Carl Gustav Carus and Faculty of Medicine, Technische Universität Dresden, Dresden, Germany; [43]National Center for Tumor Diseases, Dresden, Germany: German Cancer Research Center, Heidelberg, Germany; [44]Helmholtz-Zentrum Dresden Rossendorf, Dresden, Germany; [45]German Cancer Consortium, Dresden, Germany; [46]SouthWest Thames Regional Genetics Service, St George's University Hospitals NHS Foundation Trust, London, UK; [47]Medizinische Klinik und Poliklinik IV, Klinikum der Universität München, Munich, Germany; [48]Medizinisch Genetisches Zentrum, Munich, Germany; [49]Department of Internal Medicine I, University Hospital Bonn, Bonn, Germany; [50]Department of Human Genetics, Radboud University Medical Center, Nijmegen, Netherlands; [51]Department of Pathology, Radboud University Medical Center, Nijmegen, Netherlands; [52]Radboud Institute of Molecular Life Sciences, Radboud University Medical Center, Nijmegen, Netherlands; [53]Genetics Service, Biocruces Bizkaia Health Research Institute, Cruces University Hospital, Cruces-Barakaldo, Bizkaia, Spain; [54]Department of Molecular Diagnostics, Institute of Oncology Ljubljana, Ljubljana, Slovenia; [55]Unidad de Medicina Genómica y Pediatría, Clínica Universidad de Navarra, Programa de Tumores Sólidos, Centro de Investigación Médica Aplicada, Instituto de Investigación Sanitaria de Navarra, Pamplona, Navarra, Spain; [56]Department of Biology and Biotechnology, University of Pavia, Pavia, Italy; [57]Max Planck Institute of Molecular Cell Biology and Genetics, Dresden, Germany; [58]GenoMed—Diagnósticos de Medicina Molecular, Lisbon, Portugal; [59]Molecular Genetics Laboratory, Elche University Hospital, Elche, Spain

Hereditary Diffuse Gastric Cancer (HDGC) syndrome is caused by germline *CDH1* Pathogenic/Likely Pathogenic variants (PV/LPV). Rare *CDH1* missense variants are frequently classified as variants of unknown significance (VUS). Surveillance/prophylactic surgery is life-saving in asymptomatic PV/LPV carriers, but clinical management remains challenging in carriers lacking clinical criteria. We present the largest genotype-phenotype analysis in *CDH1* rare-variant carriers and their relatives to study *CDH1*- associated spectrum and optimize clinical management. 1971 phenotypes from 854 carriers of 398 *CDH1* rare-variants and 1021 relatives from 29 institutions and 10 ERN-GENTURIS countries were analyzed. Variants were classified with *CDH1* ACMG-AMP guidelines. Genotype–phenotype associations were analyzed by Student's t test, Kruskal-Wallis, $\chi 2$ and multivariable logistic regression models. Equivalence test, Youden index, ROC and Z test were used to assess performance of HDGC clinical criteria sets. Lobular Breast cancer-LBC and Diffuse Gastric cancer-DGC had the greatest positive association with the presence of truncating-PV/LPVs (OR=12.39 [95% CI 2.66–57.74], p=0.0014; OR=8.00 [2.18–29.39], p=0.0017, respectively), as opposed to missense-VUS. *CDH1*-PV/LPVs occurred in 136/182 (75%) families fulfilling 2015 HDGC-clinical criteria, and in 40/672 (6%) families lacking criteria. Amongst the latter 40, 18 presented LBC but did not fulfill recent 2020 clinical criteria. Three new LBC-centred criteria improved testing sensitivity while maintaining high specificity. The probability to find a *CDH1*-PV/LPV in patients fulfilling the LBC-expanded criteria, compared with the 2020 criteria, increased significantly (AUC 0.92 vs 0.88; p=0.0004). This study supports association of *CDH1* truncating-PV/LPVs, but not missense-VUS, with HDGC specific phenotypes and supports widening HDGC-clinical criteria through the expansion of LBC-centred criteria.

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7. Genetic protection against cancer informed by Mendelian conditions.

Emily Nielsen Dandoroff Louise Bicknell and Mik Black
Department of Biochemistry, University of Otago, Dunedin

Height is a well-established risk factor for cancer development: each 10cm increase in height correlates to a ~10% increase in cancer risk. There does not appear to be a simple explanation for this association, reflecting the genetic complexities of both height and cancer. Mendelian (single gene) syndromes have tremendous power to potentially better understand such complex associations. The relationships between Mendelian overgrowth syndromes and cancer have been studied extensively, with the majority conferring increased cancer risk. Conversely, we know far less about growth-restricting syndromes and cancer risk, however, one pituitary dwarfism, Laron syndrome, was recently identified as having a protective effect against cancer development. My research aims to investigate whether variants associated with other growth-restricting syndromes are also protective against cancer. To do this, I am using a large genetic and phenotypic dataset, the UK Biobank, undertaking bioinformatic analysis to investigate potential associations between variants in genes associated with growth-restricting syndromes, and cancer risk. I have optimised specific tools to predict whether variants in these genes are likely to be deleterious, with the goal to compare incidence rates in a wide range of cancers. I am also investigating the effects of knocking down short stature and microcephaly genes on cancer cell proliferation rates. Through this research, we hope to understand more broadly the protective role variants in these disease-linked genes might play, enabling more precise risk prediction in an era of personalised medicine.

8. Single-cell RNA sequencing reveals synthetic lethal targets for chemoprevention of hereditary diffuse gastric cancer.

M Hughes[1], T Brew[1], T Godwin[1], M Black[1] & P Guilford[1]

[1] Centre for Translational Cancer Research (Te Aho Matatū), Cancer Genetics Laboratory, Department of Biochemistry, University of Otago, Dunedin

CDH1 is a tumour suppressor gene encoding E-cadherin, a protein responsible for cell-cell adhesion, polarity, and differentiation in epithelial tissues. Heterozygous germline mutations in CDH1 are a well-established cause of hereditary diffuse gastric cancer (HDGC). HDGC confers up to an 80% lifetime risk of diffuse gastric cancer, as well as up to a 60% lifetime risk of lobular breast cancer; for which there are limited, and highly invasive, preventative measures. Genes which have a synthetic lethal (SL) relationship with CDH1 could be promising drug targets for chemoprevention and treatment of HDGC, as well as treatment of tumours with CDH1 loss. Promising SL candidates are screened in a unique murine gastric organoid model of HDGC, derived from mice with a cre-lox inducible CDH1 knockout system. Using single cell RNA sequencing (scRNA-seq) of non-cancerous organoids, with and without CDH1 expression; we have identified 19 genes which appear to have an SL relationship with CDH1 and could be involved in tumorigenesis of HDGC. Pathways involved in immune and stromal interactions are overrepresented in this gene set, highlighting the importance of these interactions in tumorigenesis. A subset of these genes have been targeted using five existing drugs in an isogenic pair of MCF10A cell lines, with and without CDH1 expression. Three of these drugs demonstrate a synthetic lethal effect in CDH1 null cells compared to CDH1 wild type cells. Work to validate these SL interactions in our organoid model of HDGC is currently ongoing, with further investigations to confirm downregulation of target genes to follow. Drugs validated to have an SL effect in the organoid model may be viable chemopreventative or treatment options for individuals with HDGC.

9. How STAG proteins influence cohesins role in cell fate determination

Lynch D. M., Meier, M., Labudina A., Antony J., and Horsfield J. A.

Department of Pathology, Dunedin School of Medicine, University of Otago

Cohesin is a multi-protein complex comprising of core proteins; RAD21, SMC1A and SMC3, and either STAG1 or STAG2. During mitosis cohesin is responsible for sister chromatid cohesion. In interphase cohesin is involved in three-dimensional regulation of chromatin, to ensure correct gene expression in different cell types. Cohesin is essential for normal development and germline mutations in the cohesin complex cause an array of developmental disorders, named cohesinopathies. During interphase cohesin-STAG1 and cohesin-STAG2 occupy different gene regulatory roles, cohesin-STAG1 is usually located at chromatin interaction boundaries whereas cohesin-STAG2 is associated with small scale loops that control tissue specific gene expression. Single *STAG* mutations don't affect sister chromatid cohesion which allows us to study the gene regulatory role of cohesin without affecting its mitotic role. Using the zebrafish as a model organism, and the zebrafish tailbud as a model for stem cell differentiation, we study the effects of cohesin mutations on developmental cell fate determination. In this ongoing project we use *in-situ* techniques to investigate the contribution tailbud stem cells to growing structures of the zebrafish embryo in a *stag1* mutant background. RNA-seq data has highlighted that BMP signaling genes and endothelial cell markers are dysregulated in *stag1* mutants. Follow up experiments are being carried out to confirm whether the developmental endothelial lineage is affected when cohesin-Stag1 is lost.

10. Evaluating the impacts of climate change at a cellular resolution through comparative single cell sequencing of kuku (green-lipped mussels)

Mary K N Hawkes[1], Peter K Dearden[1], Neil J Gemmell[2], Nathan J Kenny[1]
[1] *Department of Biochemistry Te Tari Matū Koiora, University of Otago, Dunedin*
[2] *Department of Anatomy Te Tari Kikokiko, University of Otago, Dunedin, Aotearoa*

The kuku (green-lipped mussel, Perna canaliculus) is a New Zealand-endemic species. It is the most valuable species for New Zealand's aquaculture industry, is considered a taonga (treasure) due to its cultural importance, and is a keystone species in coastal ecosystems. This species' contributions to New Zealand's well-being are under threat as it feels the effects of climate change, with both adult and larval kuku experiencing mass mortality events. Fortunately, not all kuku are affected equally, with some showing resilience to heat and ocean acidification stresses. The basis of this phenomena is currently poorly understood and early developmental processes have been implicated. Understanding and contrasting development at fine detail will therefore provide vital data for understanding resilience and mitigating the deleterious effects of climate change in this species. To understand these fundamental biological processes, we are utilizing the SPLiTseq method of single cell RNA sequencing, after cell dissociation using the ACME approach. ACME simultaneously fixes and dissociates cells, preserving a biologically relevant transcriptional state, while SPLiTseq uses combinatorial barcoding to offer low-cost single cell sequencing without specialized equipment. The combination of these techniques means developmental atlases can be created for the understudied non-model organisms that climate change threatens the most. Furthermore, we are able to contrast samples taken from different genetic backgrounds and environmental conditions. Alongside more traditional methods (bulk RNAseq, HCR imaging), this will allow us to discern the cellular, transcriptomic and developmental changes that organisms will use to endure and adapt to climate change, now and into the future.

11. Molecular characterisation of novel *de novo* variants in *ELAVL2*

Meghan R. Mulligan[1], Louise S. Bicknell[1] and the ELAVL2 clinical consortium
[1]Rare Disorder Genetics Laboratory, Department of Biochemistry, University of Otago, Dunedin

Neurodevelopment is a highly complex process that requires tight regulation. Disruption can result in neurodevelopmental disorders (NDDs) such as autism spectrum disorder, developmental delay, and epilepsy. We have collated an international cohort of nine patients who all have de novo variants in the gene ELAV-*like* 2 (*ELAVL2*) and present with similar neurodevelopmental phenotypes. ELAVL2 is an RNA-binding protein implicated in the regulation of RNA metabolism, particularly in the brain, and has not yet been associated with NDDs. This project aims to determine whether variants in *ELAVL2* are causing a novel genetic disorder and will explore the exact function of ELAVL2 in the cell and how this may be disrupted in our patients. Five truncating variants cause a null allele, confirming that haploinsufficiency for ELAVL2 is the likely disease mechanism. However, the three missense variants and a terminal exon frameshift (predicted to cause a truncated protein) require further molecular investigation. We hypothesise that these variants are also acting in a haploinsufficient manner, by disrupting the levels or function of ELAVL2. To determine the pathogenicity of the missense variants, a cycloheximide chase assay was performed to assess protein stability. Results indicate that one of the missense variants (p.Ala333Glu) and the terminal exon frameshift decrease protein stability, supporting our haploinsufficiency hypothesis and that these two variants are pathogenic. The remaining two variants are currently being tested for ELAVL2 dimer formation and RNA binding ability. Recently, a further four missense variants have been identified and are undergoing molecular characterisation. Our results support that variants in *ELAVL2* cause a novel neurodevelopmental disorder, with future RIP-seq and RNA-seq experiments providing insight into the specific targets and processes that are likely being disrupted.

12. Exploring the diverse viromes of New Zealand's bats

Stephanie J. Waller[1], Pablo Tortosa[2,3], Tertia Thurley[4], Colin O'Donnell[4], Rebecca Jackson[4], Gillian Dennis[4], Rebecca M. Grimwood[1], Edward C. Holmes[5], Kate McInnes[4], Jemma L. Geoghegan[1,6].
[1]Department of Microbiology and Immunology, University of Otago, Dunedin
[2]UMR PIMIT Processus Infectieux en Milieu Insulaire Tropical, Université de La Réunion, CNRS 9192, INSERM 1187, IRD 249, Plateforme de recherche CYROI, 2 rue Maxime Rivière, 97490 Ste Clotilde, France.
[3]Department of Zoology, University of Otago, P.O. Box 56, Dunedin 9054
[4]Department of Conservation, P.O. Box 10420, Wellington 6143
[5]Sydney Institute for Infectious Diseases, School of Life and Environmental Sciences and School of Medical Sciences, The University of Sydney, Sydney, NSW 2006, Australia.
[6]Institute of Environmental Science and Research, Wellington

The lesser short-tailed bat (*Mystacina tuberculata*) and the long-tailed bat (*Chalinolobus tuberculatus*) are Aotearoa New Zealand's only native terrestrial mammals. Both species are thought to have arrived in New Zealand from Australia. The long-tailed bat migrated an estimated two million years ago to New Zealand and is closely related to other bat species found in Australia today. The lesser short-tailed bat, in contrast, is the only extant species within the family Mystacinidae and is estimated to have been living in isolation in New Zealand for the past 16-18 million years. Throughout this long period of isolation, the lesser short-tailed bat has become one of the most terrestrial and highly concentrated tree roosting bats in the world despite their vulnerable status. In this study, we aimed to uncover the

viromes of New Zealand's bats to determine whether viruses have jumped between these species in the past two million years and whether they harbour viruses of importance to public health. We performed a metatranscriptomic analysis of guano samples from eight locations across New Zealand. We found an increase in viral richness among long-tailed bats compared to lesser short-tailed bats. That is, we identified bat viruses spanning seven different viral families infecting long-tailed bats while we were unable to identify bat-specific viruses in the lesser short-tailed bats, although in both species we identified an abundance of dietary- and environmental-associated viruses. Further, we identified an *Alphacoronavirus* in long-tailed bat guano that had previously been identified in bat guano from lesser short-tailed bats, suggesting that this virus has jumped the species barrier since the former species migrated to New Zealand. Overall, this study has uncovered many novel viruses harboured by New Zealand's only native terrestrial mammals, adding to our knowledge of the diversity of bat viromes generally.

13. Identifying Superheroes in Neurodegenerative Disease

Caroline Stokes, Indranil Basak, Warren Tate, Stephanie Hughes
Department of Biochemistry, University of Otago, Dunedin

CLN5 Batten disease is a monogenic lysosomal storage disease. Symptoms include cognitive & motor decline, loss of vision & epilepsy. Variability in clinical presentation is indicative that the broader genetic background may be affecting the phenotypic outcome of the patient. "Superhero genes" or genetic modifiers that compensate in part for deleterious effects of the mutated gene may be present. A whole-genome CRISPRi screen for such "superhero genes" in a human neuronal model of CLN5 Batten disease is being used to identify protective modifiers & potential targets/pathways for therapeutic intervention. Initial steps have included characterising the novel iPSC-derived human neuronal model of CLN5 Batten disease, assessing lysosomal activity in the cells, & confirming the integrity of the commercial CRIPSRi library. The integrity & status of the iPSC cell line following CRISPRi knockdown of CLN5 was confirmed through immunocytochemistry for pluripotency markers (Nanog, Oct4, Ssea4). CLN5 loss was confirmed by RT-qPCR, & western blotting, which revealed no detectable CLN5 protein in knockdown cell lines. A shift in lysosomal acidity, which is characteristic of CLN5 Batten disease state, was confirmed in the CLN5i cells by FACS experiments. The next steps are to use a lentiviral CRISPRi library to transduce stable CLN5i iPSCs, which will be differentiated into neurons. Cells with significantly increased/decreased lysosome acidity will be isolated by FACS. Study of genetic modifiers in monogenic disorders such as Batten disease provides a pipeline for future studies in more common neurodegenerative diseases.

14. Exploring a role for Histone H4 in the cell: Impact of missense variants

Naik N. and Bicknell. L
Department of Biochemistry, University of Otago, Dunedin

The dynamic reorganisation of the chromatin network is a critical mechanism through which gene expression is regulated during development, with disruptions of the mechanism being associated with occurrence of developmental disorders. Histones are proteins associated with the chromatin, playing an important role in chromatin dynamics, influencing gene expression and genome integrity. Recent research and clinical data have shone light on the involvement of histone H4, specifically the importance of post translational modifications (PTMs) of H4,

in development. Mutation of one of the H4 genes, cause the amino acid residue, Lysine-91 to be replaced by glutamic acid disrupting the PTM ubiquitination leading to a neurodevelopmental and growth disorder. Our aim here is to characterise the disruption of PTM ubiquitination of K91 as well as check if disruption of other conserved PTMs of histone H4 can be a risk factor for development. We have established stable HEK293 cell lines engineered to have an inducible expression of our mutant H4 protein using the Flp-In™ T-REx™ system. By inducing expression with tetracycline, we will check for the impact of mutant H4 on genome integrity, RNA polymerase progression, gene expression, chromatin accessibility and overall change in PTM patterns.

15. A second hotspot for pathogenic exon-skipping variants in *CDC45*

Mischa S G Ruegg[1], Bridget Fellows[1], Louise S Bicknell[1], and *CDC45* clinical collaborators.
[1]Rare Disorder Genetics Laboratory, Department of Biochemistry, University of Otago, Dunedin

Biallelic pathogenic variants in *CDC45* cause craniosynostosis and reduced stature, which can often manifest as Meier-Gorlin syndrome with craniosynostosis. Identified variants act through a hypomorphic loss of function mechanism, to reduce CDC45 activity and impact DNA replication initiation. In addition to missense and premature termination variants, several pathogenic synonymous variants have been identified, most of which cause increased exon skipping of exon 4 which encodes an essential part of α/β domain I. Here we have identified a second cohort of families segregating *CDC45* variants, where patients have craniosynostosis and a reduction in height, alongside common facial dysmorphism, including thin eyebrows. In this cohort, skipping of exon 15 is a consequence from two different variants, including a recurrent synonymous variant that is enriched in individuals of East Asian ancestry. Other identified variants are predicted to alter key intramolecular forces in α/β domain I, or cause intron retention within the 3′UTR. Our cohort and functional data confirm exon skipping is a relatively common pathogenic mechanism in *CDC45*, and highlights the need for alternative splicing events, such as exon skipping, to be especially considered in variants of lower priority.

16. What's in that poo? Metagenomic screening of kurī palaeofaeces from Whenua Hou

Meriam van Os[1], Brooke Tucker[2], Atholl Anderson[2], Michael Knapp[1], Catherine Collins[1], Karen Greig[2]
[1]Department of Anatomy, University of Otago, Dunedin
[2]Archaeology Programme, University of Otago, Dunedin

Palaeofaeces are preserved ancient faeces that provide a unique timestamp of the past. Not only can they inform us about the nuclear and mitochondrial DNA of the defecator, but also about its microbiome, diet, health and disease, and the environment in which it lived. While palaeofaeces are not unusual in archaeological sites throughout Aotearoa New Zealand, their full potential has yet to be explored, especially in terms of the genetic information they can yield. This poster presents the initial results of metagenomic shotgun analyses of ten kurī (the extinct Māori dog) palaeofaeces, excavated in 2019 from the archaeological site of Sealers Bay Camp (D48/5) on Codfish Island/Whenua Hou (Foveaux Strait). The site was inhabited during two distinct periods, with an initial Māori occupation phase (thirteenth to the fifteenth century AD), and a nineteenth-century bicultural village, formed in the early stages of Māori-Pākehā cultural engagement in Foveaux Strait. Significantly, kurī were present during both periods. This poster focuses on the metagenomic findings from the palaeofaeces, placing this data within its broader archaeological context.

17. Unveiling the Hidden Role of Histone H4 in Chromatin Dynamics and its Significance in Neurodevelopmental Disorders

Rosie Sullivan, Nihar Naik, Karen Knapp, Louise Bicknell
Rare Disorder Genetics Laboratory, Department of Biochemistry, University of Otago, Dunedin

Chromatin, a fundamental structural component, consists of nucleosomes, each comprising a DNA double-stranded fibre wrapped around a histone octamer. This intricate organization underpins vital cellular processes, including DNA replication, transcription, and repair across all eukaryotes. Dynamic chromatin reorganization plays a crucial role in the regulation of gene expression during development, with its disruption is strongly linked to neurodevelopmental disorders. While chromatin regulators have been studied, very little is known about the role and functions of histones in brain development. Human histone H4 is encoded by fourteen canonical histone H4 genes, each distinct at the nucleotide level while encoding a conserved protein. Our study focuses on an international cohort of approximately 40 patients with de novo missense variants in six of the 14 H4 genes (*H4C3, H4C4, H4C5, H4C6, H4C9,* and *H4C11*). Individuals present Tessadori-Bicknell-van Haaften neurodevelopmental syndrome characterised by poor overall growth, profound global developmental delay with absent speech, and characteristic dysmorphic facial features, including hypertelorism, abnormal nose, and wide mouth. The exact mechanism by which variant histone H4 is leading to this disorder is unclear. We hypothesise these patient variants are altering chromatin dynamics, disrupting brain development. Stable Hek 293 cell lines expressing WT or patient variant H4 have been made and are being used in a variety of cellular assays to better understand the consequences of these mutations on cellular functioning.

18. Genomic Strategies for Avian Survival: Unravelling Rat Population Dynamics to Protect New Zealand's (Aotearoa) Feathered Taonga

Brooke L. Whitelaw, Maren Wellenreuther, James C. Russell, Catherine J. Collins
Department of Anatomy, University of Otago, Dunedin

The introduction of Norway (*Rattus norvegicus*) and ship rats (*Rattus rattus*) to Aotearoa by European vessels in the late 1700s devastated native vertebrate and invertebrate populations. These opportunistic invasive mammals threaten borrowing seabirds and forest taonga (treasured by NZ Māori) bird species. Current methods of rodent elimination, primarily poison and traps, scale poorly on the mainland, where refuge populations facilitate survivors and reinvasion. To implement a successful eradication strategy, a greater understanding of population dynamics, structure and the movement of individuals is required. Previous work examining the d-loop region of *R. rattus* and *R. norvegicus* mitochondrial genomes identified putative invasion events with either English or Chinese origins and established. It established haplotype diversity across the Aotearoa mainland and surrounding islands. We present an in-depth examination of *R. rattus* and *R. norvegicus* mitochondrial genomes sampled from across Aotearoa, providing a greater context of population dynamics for both species. This work will be complemented, in the future, by an accompanying whole genome sequencing data set, which is currently undergoing investigation. In the future, this work will be complemented by an accompanying whole genome sequencing data set currently being investigated. Additionally, we extracted and examined the *Vkorc1* gene for mutations, which

has been found to confer anticoagulant resistance rendering some toxins ineffective for rodent control.

19. Formalin-fixed paraffin-embedded (FFPE) samples help to investigate potential wildlife disease

Miller, Allison[1]; Baker, Cindy[2], Brosnahan, Cara[3], Dowle, Edwina[1], Gemmell, Neil[1], Geoghegan, Jemma[5], Kitson, Jane[4], Pande, Anjali[3]
[1]*Department of Anatomy, University of Otago, Dunedin*
[2]*National Institute of Water and Atmospheric Research Limited*
[3]*Animal Health Laboratory, Ministry for Primary Industries*
[4]*Kitson Consulting Ltd*
5 Department of Microbiology and Immunology, University of Otago, Dunedin

Infectious diseases impact numerous organisms. Knowledge of host-pathogen interactions and host responses to infection is crucial for conservation and management. Obtaining this knowledge quickly is made increasingly possible by a variety of genomic approaches, yet, for many species the bottleneck to understanding this, remains access to appropriate samples and data. Lack of sample availability has also limited our understanding of how pathogens and the immune responses of hosts change over time. Archival materials may provide a way to explore pathogen emergence and host responses over multiple—possibly hundreds—of years. Here, we tested whether formalin-fixed paraffin-embedded (FFPE) tissue samples could be used to understand an unknown pathology, lamprey reddening syndrome (LRS), affecting a wild fish species (pouched lamprey, *Geotria australis*). Our differential expression analyses of dermal tissues from four unaffected lampreys and eight affected lampreys collected in 2012 alluded to potential agents associated with LRS. Interestingly, the pathways associated with viral infections were overrepresented in affected versus unaffected lamprey. Gene ontology analyses of the affected and non-affected lampreys also provided new insights into the largely understudied immune responses of lampreys. Our work confirms that FFPE samples can be used to infer information about the transcriptional responses of a wild fish species affected by unknown historical pathologies/ syndromes. In addition, the use of FFPE samples for transcriptomics offers many opportunities to investigate the genomic responses of wild populations to a variety of environmental changes. We conclude with a discussion about how to best sample and utilize these unique archival resources for future wildlife transcriptomic studies.