

7th Annual ANZSCDB Melbourne Cell and Developmental Biology Symposium

5th November 2014

Alfred Medical Research and Education Precinct (AMREP), 75 Commercial Rd, Melbourne 3004

8:00 – 9:00 **Registration**
Foyer, Alfred Medical Research and Education Precinct (AMREP)

Poster Setup
Seminar Room, AMREP

9:00 – 9:15 **Welcome & Introduction**
Lecture Theatre, AMREP

ORAL PRESENTATIONS - SESSION 1

9:15 – 9:35 **Hannah Vanyai**
Identifying targets of the histone acetyltransferase, MOZ, in palate development

9:35 – 9:55 **Harley Owens**
The role of the Grainyhead-like transcription factor family in craniofacial development

9:55 – 10:40 **Plenary 1: Prof. Jane Visvader**
Joint Division Head, Stem Cells and Cancer, WEHI
Title: "Getting abreast of the mammary differentiation hierarchy and cancer"

10:40 – 11:10 **Morning Tea and Posters**
Seminar Room and Foyer, AMREP

SESSION 2

11:10 – 11:30 **Celia Vandestadt**
Defining cellular mechanisms initiating regeneration in the zebrafish CNS

11:30 – 11:50 **Ivan Gladwyn-Ng**
Bacurd2 is a novel interacting partner to Rnd2 which controls distinct phases of radial migration within the developing mammalian cerebral cortex

11:50 – 12:10 **Francesca Froldi**
Drosophila transcription factor Nerfin-1 prevents reversion of neurons into neural stem cells

12:10 – 12:30 **Jeremy Ng**
Environmental cues direct fate specification during zebrafish retinal regeneration

12:30 – 12:50 **James Godwin**
Defining the role of innate immunity and nerve signalling in adult salamander limb regeneration

12:50 – 2:00 **Lunch and Poster session**
Seminar Room and Foyer, AMREP

SESSION 3

- 2:00 – 2:20 **Prusothman Yoganantharajah**
Bisphenol A causes increased lipid deposition in a zebrafish model of obesity
- 2:20 – 2:40 **Katherine Lange**
Using comparative gene ontology analyses to identify mechanisms of early stem cell differentiation
- 2:40 – 3:25 **Plenary 2: Prof. Emma Whitelaw**
Head of Department of Genetics, La Trobe University
Title: "Epigenetics: An update"
- 3:25 – 4:00 **Afternoon Tea and Poster session**
Seminar Room and Foyer, AMREP

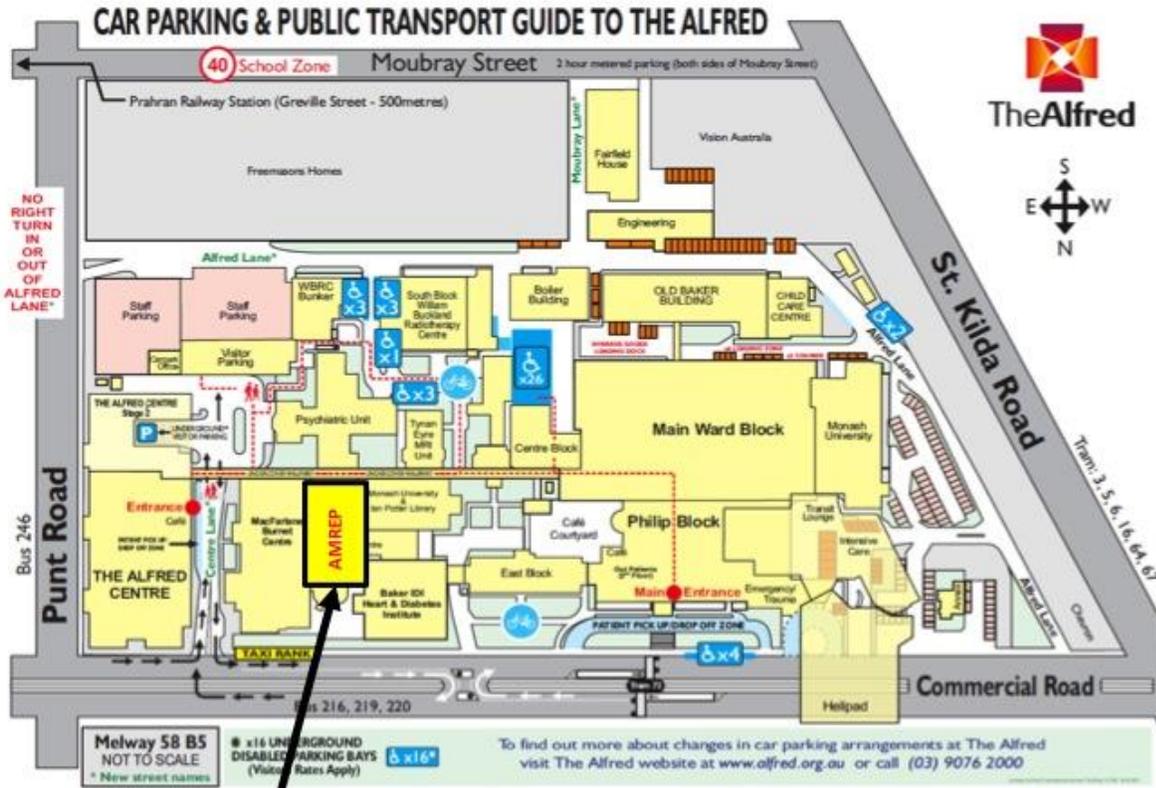
SESSION 4

- 4:00 – 4:20 **Joffrey Degoutin**
Identification of new growth regulators modulating the Hippo pathway
- 4:20 – 4:40 **Jesus Fernandez-Casanova**
The murine miR-196 cluster is essential in patterning and development of the vertebral column
- 4:40 – 5:00 **Kieran Short**
An integrated cell, tissue and whole organ profile of kidney morphogenesis
- 5:00 – 5:20 **Announcement of Prizes and Close**
- 5:30 – 6:30 **Drinks and Finger Food**
Foyer, AMREP

With Thanks to our Sponsors:



VENUE



AMREP Education Centre and Lecture Theatre
75 Commercial Rd Melbourne 3004
(Entry from Commercial Rd)

Abstracts – Oral Presentations

SESSION 1

9:15 – 9:35 **Hannah Vanyai**

Identifying targets of the histone acetyltransferase, MOZ, in palate development

Monocytic leukaemia zinc finger protein (MOZ/MYST3/KAT6A) is a member of the MYST family of histone acetyltransferases. Histone acetylation is an essential element of the regulation of gene transcription. We have shown that MOZ is required for the acetylation of lysine 9 of histone 3 (H3K9) at Hox gene loci as well as at Tbx1, a gene important in the pathogenesis of the human congenital condition DiGeorge syndrome. Moz mutant mice phenocopy DiGeorge syndrome, displaying cardiovascular defects, thymic abnormalities and cleft palate. We hypothesised that MOZ regulates genes in addition to Tbx1 during in palate development. Using a candidate gene approach as well as expression profiling by RNA sequencing, we have identified new targets of MOZ required for correct patterning of the palate. We have used conventional developmental biology approaches and well as Next Generation Sequencing techniques to expand our understanding of the pathways regulated by MOZ in this context.

9:35 – 9:55 **Harley Owens**

The role of the Grainyhead-like transcription factor family in craniofacial development

Anomalies in craniofacial development, such as oral clefts, are one of the most common of all birth defects. Because of the multifactorial mode of development of the craniofacial skeleton, pre- natal diagnosis and treatment is difficult, thus highlighting a need for comprehensive knowledge of the molecular and genetic interactions that regulate craniofacial development. The *Grainyhead-like (Grhl)* family of transcription factors, particularly *Grhl2*, have been recently described as novel regulators of development of the craniofacial skeleton. In this study, we have shown that loss of *Grhl2* leads to defective maxillary fusion and maintenance of the first pharyngeal arch (PA1), and, through tissue-specific *Cre*-mediated deletion, that this factor is an important component of mandibular morphogenesis in the pharyngeal endoderm, as well as maintenance of jaw architecture in the pharyngeal ectoderm. Furthermore, epistasis experiments have revealed a potential common pathway for *Grhl2* and *IRF6*, but not *Grhl3*, in the context of palatal shelf elevation. We have shown that *Grhl2* is essential in craniofacial development, and contend that further characterisation of this gene is an important step towards the generation of novel therapeutic approaches that can prevent craniofacial aberrations.

SESSION 2

11:10 – 11:30 **Celia Vandestadt**

Defining cellular mechanisms initiating regeneration in the zebrafish CNS

In mammals, spinal cord (SC) injury results in a devastating loss of function due to the restricted degree of tissue repair in the central nervous system (CNS). By contrast, zebrafish have a remarkable capacity to regenerate their CNS following injury, making them an ideal model to understand the regenerative process. The regenerative response of CNS cell types following injury is beginning to be unravelled, however, the contribution of immune and vascular cells in mediating the regenerative response remain unclear. We have developed a larval SC lesion assay with which we can examine CNS regeneration at the cellular level with live confocal imaging *in vivo* to investigate the contribution and interaction of immune and vasculature-derived cells with glia and neurons during the regenerative process. We show first that in agreement with other injury models, glial regeneration precedes the reestablishment of neuronal cell types. Second, while macrophages and neutrophils are recruited rapidly following injury, they display a lack of spatial interactions with CNS cell types, including a lack of phagocytosis. Third, we show that glial bridging occurs prior to vascular regeneration. Finally, using the *cloche*^{-/-} mutant we reveal that neither the acute immune response nor vasculature is involved in initiating SC regeneration. Collectively our results indicate that vasculature and immune cells do not appear to be required for the initiation of glial or neuronal regeneration following SC injury in larval zebrafish.

11:30 – 11:50 **Ivan Gladwyn-Ng**

Bacurd2 is a novel interacting partner to Rnd2 which controls distinct phases of radial migration within the developing mammalian cerebral cortex

Within the developing mammalian cerebral cortex, newborn excitatory neurons engage in radial migration as they leave their birthplace in the germinal ventricular zone and reach their final location before undergoing terminal differentiation. We have previously reported that the atypical RhoA GTPase Rnd2 promotes the radial migration of newborn cortical projection neurons within the embryonic cerebral cortex (REF), but its downstream signalling pathways are not well understood. In this study, we identify Bacurd2 (a member of the BTB-domain containing adaptor for Cul3-mediated RhoA degradation) as a novel interacting partner to Rnd2 which promotes radial migration within the mouse cerebral cortex during embryonic and post-natal development. We find that Bacurd2 binds Rnd2 at its C-terminus, and this interaction is critical to its role in cell migration. To investigate how the interaction between Bacurd2 and Rnd2 might be important for cell migration within the embryonic cortex, we engineered a Bacurd2:Rnd2 chimeric construct and discovered that the migration-defect of Rnd2shRNA-treated cells could be corrected by co-delivery of this construct. Our cellular analysis further reveals that Bacurd2-Rnd2 signalling is critical for coordinating the multipolar-to-bipolar transition of neurons within the intermediate zone, as well as their radial migration within the cortical plate. Therefore, our results identify Bacurd2 as a critical player during cerebral cortical development which guides the proper positioning of newborn neurons through its interaction with Rnd2.

11:50 – 12:10 **Francesca Foldi**

Drosophila transcription factor Nerfin-1 prevents reversion of neurons into neural stem cells

Cellular dedifferentiation is the regression of a cell from a specialized state to a more pluripotent state and is implicated in cancer. However, the transcriptional network that prevents differentiated cells from reacquiring stem cell fate is so far unclear. *Drosophila* neuroblasts are a model for the regulation of stem cell self-renewal and differentiation. We show that the *Drosophila* zinc finger transcription factor Nervous fingers 1 (Nerfin-1) locks neurons into differentiation, preventing their reversion into neuroblasts. Following Prospero-dependent neuronal specification in the Ganglion Mother Cell (GMC), Nerfin-1-specific transcriptional program maintains differentiation in the postmitotic neurons. The loss of Nerfin-1 causes reversion to multipotency, and results in tumours in several neural lineages. Both the onset and rate of neuronal dedifferentiation in Nerfin-1 mutant are dependent on Myc and Tor-mediated cellular growth. In addition, Nerfin-1 is also necessary and sufficient to promote neuroblast differentiation at the end of neurogenesis. RNA-sequencing and Chromatin Immunoprecipitation (ChIP) analysis show that Nerfin-1 administers its function in neurons and neuroblasts by repression of self-renewing- and activation of differentiation-specific genes. Our findings support the model of bidirectional inter-convertibility between neural stem cells and their postmitotic progeny, and highlight the importance of Nerfin-1 regulated transcriptional program in neuronal maintenance.

12:10 – 12:30 **Jeremy Ng**

Environmental cues direct fate specification during zebrafish retinal regeneration

In response to injury in vertebrate retinas, different types of growth, cell signalling factors and secreted peptides are produced to drive regeneration. Some of these extrinsic factors are now being tested in animal models to improve regeneration. However, the relative contribution of extrinsic factors and their influences on different aspects of regeneration such as on fate specification is largely unknown. It is furthermore unclear, if cell types are regenerated according to the same developmental birth order, or if only the degenerated neuron types are specifically regenerated. Using 2 injury models in the highly regenerative zebrafish retina, I have conducted a comparative study on fate specification to understand the interactions between environmental cues and the downstream regenerative response. Preliminary data suggests that an injury that ablates only inhibitory neurons will generate larger proportions of inhibitory neurons in comparison to an injury model that ablates all cell types, where all cell types are generated in equal proportions as in development. This suggests that regeneration, at least in the retina, follows a different mechanism than during development and the environment can direct fate specification during regeneration. Thus as a potential therapy for retinal diseases, transplanting unspecified progenitors can selectively generate only the missing neurons.

12:30 – 12:50 **James Godwin**

Defining the role of innate immunity and nerve signalling in adult salamander limb regeneration

In response to injury, adult mammals have limited potential for regeneration. In contrast, adult salamanders display a remarkable capacity for faithful repair by regenerating complex structures such as amputated limbs or damaged hearts, brains and spinal cords. Regeneration of a new limb is accomplished through the formation of a mound of progenitor cells (blastema) that arises after amputation only in the presence of damaged nerve. The blastemal progenitor cells then undergo differentiation and patterning into a new limb. Using the axolotl (*Ambystoma mexicanum*) as a model to study vertebrate regeneration, we have previously demonstrated a temporally defined requirement for innate immune cells (macrophages) during the limb regeneration process and identified nerve dependent signals capable of rescuing regeneration in the absence of nerve. We established FACS based protocols to isolate specific cell populations during limb regeneration using gene expression analysis to profile the regeneration specific response. These studies provide the foundation to identify and investigate novel genetic programs promoting scar-free wound healing or regeneration and define important aspects of the neuro-immunological axis of regeneration.

SESSION 3

2:00 – 2:20 **Prusothman Yoganantharajah**

Bisphenol A causes increased lipid deposition in a zebrafish model of obesity

Bisphenol A, a chemical in plastics receives scrutiny because it is an estrogenic endocrine disrupting chemical (EDC). BPA exposure at low or even short term doses causes a variety of health complications and diseases. Many countries have stopped using BPA in the manufacturing of food containers, especially for infant products. The Plastics industry has proposed to substitute BPA with Bisphenol S and Bisphenol AF. This project aimed to determine if physiologically relevant doses of BPA, BPS and BPAF can cause increased lipid deposition and increase the chances of developing obesity. Exposure to BPA, BPS BPAF during zebrafish embryogenesis increases lipid deposition, and also induces the activation of the classical estrogen pathway; however this is not responsible for the increase in lipid deposition. Bisphenol exposure also increased levels of, LPL, cebp α and LFABP, demonstrating these increases maybe due to increased levels of triglycerides, long chain fatty acids and elevated numbers of pre-adipocytes. BPA, BPS and BPAF exposure during the early stages of zebrafish embryogenesis has an effect on lipidogenesis and adipogenesis and this increases an individual's susceptibility towards developing obesity.

2:20 – 2:40 **Katherine Lange**

Using comparative gene ontology analyses to identify mechanisms of early stem cell differentiation

Successful application of stem cells for therapeutic purposes relies on understanding the molecular signals governing the balance between pluripotency and differentiation. Traditional views of lineage commitment advocate progressive restrictions in cell potency along one of the three primary tissue lineages driven by key lineage-specific transcription factors. However, these views have been challenged by recent studies suggesting multi-potent progenitor stages (Tzouanacou et al. 2009), a pluripotency spectrum and priming for differentiation (Pera and Tam 2010). This research examines the role of one such early differentiation marker, Mixl1, implicated in regulation of mesoderm and endoderm differentiation and/or morphogenesis (Robb et al. 2000; Hart et al. 2002). Using a comparative global transcriptional analysis, of mouse ES cells with an inducible Mixl1-transgene, and Mixl1-null mouse embryos and embryoid bodies, we have identified a novel role for Mixl1 in epithelial-to-mesenchyme transition (EMT) and cytoskeletal rearrangement to initiate endoderm expansion during gastrulation. These results highlight the importance of cell movement and changing molecular signals over time encountered by a differentiating cell during normal embryonic development, and suggest considering using specific changes in external signals for more specific directed cell differentiation in vitro.

SESSION 4

4:00 – 4:20 **Joffrey Degoutin**

Identification of new growth regulators modulating the Hippo pathway

Control of organ size is a key biological process during development of organisms and it is likely that crucial tissue growth regulators are yet to be discovered. We performed a screen in *Drosophila melanogaster* to find new genes that act on the Hippo growth control pathway. We focused on uncharacterised genes conserved throughout evolution. Out of 500 tested genes, we identified 17 candidates that strongly regulate growth. One particular gene, that we named *wen*, is able to regulate tissue growth through a regulation of the transcriptional Hippo pathway co-activator Yorkie.

4:20 – 4:40 **Jesus Fernandez-Casanova**

The murine miR-196 cluster is essential in patterning and development of the vertebral column

The total number of vertebrae, and the identity of each segment, are highly reproducible within a given vertebrate species. Here we show that the miR-196 family of microRNAs are essential in defining the total vertebral number in mouse as well as shaping correct regionalisation of the vertebral column. Using an extensive allelic series of mouse knockouts, we show that the three miR-196 family members (miR-196a1, miR-196a2 and miR-196b) act redundantly to restrict axis length, in part via modulation of Wnt activity. Independent of this, miR-196a2 and miR-196b act in a dose-dependent manner to control the number of rib-bearing vertebrae and positioning of the sacrum. We reveal unappreciated complexity in miR-196 regulation of Hox cluster expression dynamics. Loss of miR-196 leads to a collective upregulation of numerous trunk Hox target genes with a concomitant delay in posterior Hox gene activation, genes which are proposed to signal the end of axis extension and whose delayed activation would support regionalised thoracic expansion. By feeding in to multiple genetic networks controlling vertebrae formation and patterning, miR-196 is critical player defining morphological output.

4:40 – 5:00 **Kieran Short**

An integrated cell, tissue and whole organ profile of kidney morphogenesis

While cell, tissue and even organism level analyses of morphogenesis are feasible in invertebrates, the size, opacity and complexity of mammalian organs has impeded systematic analyses of developmental processes critical to organ function. Here, we integrate optical projection tomography, single-cell resolution confocal and quantitative image analysis to comprehensively document mouse kidney organogenesis across time. This reveals a previously unappreciated structurally stereotypic organ architecture undergoing a temporally non-uniform process of development with respect to rates of cellular proliferation, dominant morphogenetic processes and spatial relationships between key cellular compartments. The existence of such distinct phases predicts temporal sensitivity to genetic/environmental insults, potentially enhancing our understanding of the mechanism of developmental anomalies. This approach facilitates quantitative analysis of even subtle perturbations to kidney development and is also applicable to other organ systems.

Poster Presentations:

- 1 - Sultan Alasmari *"Neutrophil extracellular trap (NET) formation by zebrafish neutrophils in vitro and in vivo"*
- 2 - Mitra Amiri *"Investigation of cellular source and molecular factors that drive remyelination after spinal cord injury (SCI) in zebrafish"*
- 3 - Molly Buntine *"Investigating the role of tryptophan metabolism in ASD-associated behaviours using Drosophila melanogaster"*
- 4 - Marina Carpinelli *"Ets1^{+/-};Fli1^{+/-} mice model aspects of Jacobsen's Syndrome"*
- 5 - Denny Cottle *"p53 activity contributes to psoriasis-like features in murine skin"*
- 6 - Alissa de Novais Freire *"Understanding the mechanisms associated with cardiomyopathies in Nkx2-5 mouse models"*
- 7 - Ana Beatriz Delavia Thomasi *"Designing tools for exacerbating dystrophic phenotypes in zebrafish"*
- 8 - Lucas Dent *"Regulation of the Hippo growth control pathway by a RhoGEF and ArfGAP"*
- 9 - Stefanie Dudczig *"Characterization of secretagogin positive neurons in the zebrafish retina"*
- 10 - Ophelia Ehrlich *"Laminin as a therapy for muscle regeneration"*
- 11 - Farrah El-Saafin *"TAF8 restricts P53 mediated apoptosis"*
- 12 - Timo Friedrich *"Detailed analysis of neural regeneration through Dynamic imaging in vivo"*
- 13 - Nurul Fuad *"Microfluidic Embryo Sorting Technology – Towards in flow analysis, sorting and dispensing of individual vertebrate embryos"*
- 14 - Liana Goodings *"Necessity of Nr4a2 in dopaminergic vs. non-dopaminergic fates in the retina"*
- 15 - Michelle Henstridge *"Localised control of Torso receptor tyrosine kinase activation in Drosophila terminal patterning"*
- 16 - Caitlin Hennessy *"Identification of new genetic regulators of wing disc eversion EMT"*
- 17 - Alexei Ilinykh *"Characterisation of IGF-1 signaling in the adult mouse heart"*
- 18 - Natasha Jansz *"Characterising the molecular mechanisms of the epigenetic modifier Smchd1"*
- 19 - Cristina Keightley *"Differential requirement for Zbtb11 in myeloid lineages in basal versus emergency myelopoiesis"*
- 20 - Kynan Lawlor *"Investigating the contribution of dermal cell populations to the basal keratinocyte microenvironment"*
- 21 - Benjamin Lindsey *"Molecular regulation of adult-born neurons in sensory niches of the zebrafish brain with exposure to a chemosensory-induced novelty assay"*
- 22 - Viola Lobert *"Determining the role of the phosphatases PHLPP1 and PHLPP2 in development and cancer"*
- 23 - Jan Manent *"Investigating the tumour suppressive properties of the autophagy pathway in Ras-driven epithelial tumorigenesis"*
- 24 - Wouter Masselink *"Evolutionary emergence of limbs: a unifying hypothesis as evidenced by infiltration of somitic mesoderm into the Apical Ectodermal Ridge"*

- 25 - Sachini Meegahakumbura *"gumballs: a novel regulator of transit-amplifying germ cell development in the Drosophila testis"*
- 26 - Ivana Mirkovic *"Functional characterisation of SCUBE2 in zebrafish embryonic muscle development"*
- 27 - Thabatta Nakamura *"Deciphering epistatic interactions between splicing regulators and signalling pathways during zebrafish neuronal development"*
- 28 - Phong Nguyen *"Examining zebrafish muscle stem cell clonal behaviors during homeostasis"*
- 29 - Vahid Pazhakh *"Modelling antifungal treatments in zebrafish embryos infected with Penicillium marneffeii"*
- 30 - Marta Portela Esteban *"Lgl regulates Notch signaling via endocytosis in the developing Drosophila eye"*
- 31 - Jeanette Rientjes/Arianna Nenci *"Monash Gene Targeting Facility: Targeting Animals for Success"*
- 32 - Ain Roesley *"Control of cell proliferation by Brahma chromatin-remodelling complex"*
- 33 - Avnika Ruparelia *"Zebrafish models of BAG3 myofibrillar myopathy suggests a toxic gain of function leading to BAG3 insufficiency"*
- 34 - Olivier Serralbo *"Migrating cells mediate long-range WNT signaling"*
- 35 - Tamar Sztal *"Zebrafish models for ACTA1 nemaline myopathy reveals a spectrum of nemaline bodies and identifies the cause of muscle weakness"*
- 36 - Markus Tondl *"Establishing and optimising a zebrafish ChIP protocol to study cis-regulatory modules in cardiogenesis"*
- 37 - Gloria Ursino *"Fetal inhibition of inflammation improves disease phenotypes in harlequin Ichthyosis"*
- 38 - Raquel Vaz *"Identification of candidate genes for nemaline myopathy"*
- 39 - Emma Watson *"MCI-1 is essential for endothelial cells in developmental angiogenesis"*
- 40 - Kevin Watt *"The Hippo pathway effector YAP is a critical regulator of skeletal muscle fibre size"*
- 41 - Ben Williams *"Not so minor after all; Emerging roles for minor class splicing in development, homeostasis and cancer"*
- 42 - Christina Woelwer *"A chemical screening approach identifies a novel role for CDK9 in erythroid enucleation"*
- 43 - Alasdair Wood *"Zebrafish models of FKRP and FKTN deficiency"*
- 44 - Jennifer Yang *"Characterization of testis- and ovary-specific long non-coding RNAs"*
- 45 - Hon Yan Kelvin Yip *"Characterization of the niche regulators that control the formation and maintenance of colon crypts"*
- 46 - Mo Zhao *"The expression and function of myomesin genes in zebrafish"*
- 47 - Feng Zhu *"A high-throughput Lab-on-a-Chip interface for zebrafish embryo biotests in drug discovery and ecotoxicology"*

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