MEDICAL ADVANCES WITHOUT ANIMALS

THE MAWA TRUST

REPLACING ANIMALS IN MEDICAL RESEARCH

MAWA RESEARCH PORTFOLIO: 2019-2020-2021

In addition to the Medical Advances Without Animals Trust's ongoing funding initiatives, MAWA is delighted with two significant developments which have enhanced and expanded the core work of the Trust:

* In 2019, MAWA and the **Australian National University** (ANU), which ranks very highly internationally, established the **Replacing Animals in Medical Research Fund (RAMR**) at the ANU to further develop research strength in alternatives to the use of animals in medical research**.** In 2020 RAMR awarded four research grants, an equipment grant, a scholarship and an international travel bursary to launch the RAMR Fund. In 2021 RAMR awarded another four research grants, a fellowship and two scholarships. Details of these awards are available in a separate document on MAWA's website and can be requested from MAWA's office.

* In late 2018, MAWA formed a research partnership with the charitable arm of **Australian Ethical (AE)**, a major fund management company that specializes in environmentally and socially responsible investments to protect people, animals and the planet. With MAWA, the **Australian Ethical Foundation (AEF)** is co-sponsoring medical research projects at the University of Sydney (USyd), one of Australia's top universities, and research into treatments for COVID-19 and other respiratory diseases at the highly esteemed Walter and Eliza Hall Institute of Medical Research (WEHI), Australia's oldest and largest medical research institute. AEF/MAWA awards are listed below, followed by MAWA's ongoing awards of grants, fellowships, scholarships, bursaries, sponsorships, prizes and internships.

AEF/MAWA CO-SPONSORED RESEARCH & DEVELOPMENT PROJECTS

Prof Wojciech Chrzanowski and Dr Sally Kim, Sydney Nano Institute and Sydney School of Pharmacy, Faculty of Health and Medicine, University of Sydney, NSW and National Heart and Lung Institute, Imperial College London, UK.

Breathe Easy - Development of the next generation treatment for chronic obstructive pulmonary disease (COPD) using a COPD-on-chip model to replace the use of animal models in lung disease and injury research including COVID-19.

Combining data from human physiology-mimicking organ-on-chip models with artificial intelligence is providing new opportunities for discoveries in biomedicine. To test the safety of new therapies, it is essential to create not only physiology-mimicking models but also disease-mimicking models. This project focuses on the development and validation of lung-on-chip models that mimic COPD to investigate the effects of stem cells and stem cell-derived extracellular vesicles on lung structure and functionality, and for testing the efficacy of new generation therapeutics to regenerate lungs damaged by disease thereby replacing the use of animal models.

A/Prof Marie-Liesse Asselin-Labat, Prof Marc Pellegrini, Dr Clare Weeden and Dr Cody Allison, Personalised Oncology Division and Division of Infectious Diseases and Immune Defence, Walter and Eliza Hall Institute of Medical Research (WEHI), VIC.

Assessing the repurposing of clinical compounds in lung organoids as an alternative to the use of animal models for treatment of COVID-19 infection.

Combining the expertise of lung biologists with infectious disease experts, this project is evaluating the efficacy of drugs for the treatment of COVID-19 by working with three-dimensional cultures of healthy lung cells called human lung organoids. Organoids are 3D culture systems that mimic the structure of the human lung, therefore obviating the need for animal models. This research is expected to lead to the rapid identification of drugs for testing as treatments for COVID-19 and other respiratory diseases in clinical trials.

AEF/MAWA CO-SPONSORED RESEARCH & DEVELOPMENT PROJECTS

Prof Wojciech Chrzanowski and Dr Yiwei Wang, Sydney Nano Institute and Sydney School of Pharmacy, Faculty of Health and Medicine, University of Sydney and Concord Clinical School and ANZAC Research Institute, Concord Repatriation General Hospital, NSW.

Skin-on-chip and skin injury-on-chip: Microfluidic platform for nanotoxicity testing, drug discovery and precision biology to replace the traditional use of animals in biomedical research and testing.

Testing the safety of nanomaterials by creating not only physiology-mimicking models but also injury-mimicking models. This project is developing two subclasses of skin models: A physiological healthy skin model-on-chip that mimics healthy skin; and a pathophysiological injury model-on-chip that mimics skin injuries, to investigate the effects of sunscreen-derived nanoparticles and chemicals present in cosmetics on skin structure and functionality and for the testing of wound healing. These models obviate the need for the traditional use of animals and will be vital for the testing of transdermal penetration of nanoparticles and their subsequent ability to travel into the blood stream.

AEF/MAWA CO-SPONSORED EQUIPMENT GRANT

A/Prof Marie-Liesse Asselin-Labat, Prof Marc Pellegrini, Dr Clare Weeden and Dr Cody Allison, Personalised Oncology Division and Division of Infectious Diseases and Immune Defence, Walter and Eliza Hall Institute of Medical Research (WEHI), VIC.

Funding toward the purchase of a Biorad Zoe Fluorescent Cell Imager. This specialized microscope will enable daily imaging of the organoids in 3D to follow their evolution over time in culture, a capability that was not previously available to the researchers.

As genetic alterations are introduced in the cells to study infection with SARS-CoV2, the fluorescence capability of this microscope will permit monitoring of the direct effect of genetic changes on the altered cells. The use of this microscope will dramatically accelerate researchers' understanding in the formation of lung organoids and the impact SARS-CoV2 has on lung cells and will advance the development of animal-free methods and technologies.

GRANTS, FELLOWSHIPS, SCHOLARSHIPS, BURSARIES, SPONSORSHIPS & PRIZES AWARDED BY MAWA

In addition to previous MAWA awards to scientists and scholars at universities and research institutions throughout Australia who are continuing their MAWA funded research and student projects, further MAWA grants, fellowships, scholarships, bursaries, sponsorships and prizes were awarded in 2019, 2020 and 2021.

MAWA RESEARCH & DEVELOPMENT GRANTS

Prof Ted Maddess, Dr Faren Sabeti, Dr Bhim Rai, Prof Chris Nolan and A/Prof Rohan Essex, Eccles Institute for Neuroscience, John Curtin School of Medical Research and Diabetes Research Laboratory, Australian National University, Ophthalmology Department, Canberra Hospital and Optometry and Vision Science, University of Canberra, ACT.

Combining non-invasive measures of central retinal physiology, and microvascular structure and function in humans to eliminate eye experiments on large mammals (non-human primates and cats).

Using human volunteers, this project will develop novel non-contact, non-invasive means of assessing the structure and function of the fovea and surrounding macula of living human eyes affected by diabetes (the leading cause of blindness in the Western world) and other retinal diseases like macular degeneration. This will obviate the need to use animals for invasive procedures like microscopy of retinal tissue samples, or the use of electrodes for functional neural testing of the eyes of large mammals that are chosen for experiments due to the similarity of their maculas to human maculas.

Dr Charles Cox, Dr Yixiao Zhang and Dr Zijing Zhou, Mechanosensory Biophysics Laboratory, Molecular Cardiology and Biophysics Division, Victor Chang Cardiac Research Institute, NSW and Shanghai Institute of Organic Chemistry, Chinese Academy of Sciences, CHINA.

Functional and structural characterization of a novel anti-malarial target using animal origin free purification techniques.

MAWA RESEARCH & DEVELOPMENT GRANTS

Characterizing a new molecule identified in the malaria parasite to understand its structure and molecular function in order to develop novel rationally designed anti-malarial therapeutics. To study complex ion channels, purification using human cells and foetal bovine serum is usually required; however, this research will use both bacterial expression and associated purification methods for truncated versions of the malarial ion channel and full-length purification will be carried out using an immortalized (no live insects) insect cell line in combination with a new commercial animal-origin free growth media completely replacing the use of foetal bovine serum.

Dr Alex Combes, Prof Nikolic-Paterson, Dr Julie Moreau, Ms Anna Nunez-Nescolarde and Ms Rachel Lam, Monash Biomedicine Discovery Institute, Department of Anatomy and Developmental Biology, Monash University, and Department of Nephrology, Monash Medical Centre, VIC.

Assessing human kidney organoids as a replacement for mouse models of renal fibrosis.

Developing human kidney organoids to replace animal models of renal fibrosis as well as all animal products used in stem cell culture and visualisation of experimental results. Renal fibrosis is a form of tissue scarring from prolonged injury and inflammation resulting in a decline in kidney function. This project will test the capacity of organoids to model kidney injury and replicate the early sequence of events that initiates a fibrotic response in humans. This human kidney organoid model is expected to provide a new platform to accelerate the development of antifibrotic therapies to retain renal function in patients suffering from chronic kidney disease and to replace animal models.

Dr Joseph Brock, A/Prof Audrey Fahrer and Ms Jeeeun (Alice) Shin, Division of Biomedical Science and Biochemistry, Research School of Biology, Australian National University, ACT.

Replacing animal derived antibodies with synthetic nanobodies using yeast surface display in the fields of structural biology, biotechnology and medicine.

Replacing the use of animals by providing a viable alternative to the production of monoclonal antibodies for experimental use by the medical research community. Antibodies are proteins produced by our immune systems which enable us to recognise and fight off foreign invaders such as viruses. Synthetic nanobodies are not only functionally equivalent to their animal derived counterparts, they are also superior due to their small size, stability and amenability to alteration via standard molecular biology tools. Common applications include their use in medical imaging and disease diagnosis, important steps in new drug discovery and design.

Dr Neville Ng, A/Prof Lezanne Ooi, Mr Simon Mansour, Ms Michelle Newbery, A/Prof Mirella Dottori and Prof Ronald Sluyter, School of Chemistry and Molecular Bioscience, Faculty of Science, Medicine and Health, Molecular Horizons and the Illawarra Health and Medical Research Institute, University of Wollongong, NSW.

Rapid and cost-effective patient-derived transdifferentiation and assay methods to replace animal models and products in neurological disorder studies and therapeutic development research.

Establishing a platform of cost-effective animal-product-free methods for direct conversion of human patient biopsy samples to cell types relevant to neurological disease, and rapid animal-free downstream analysis to greatly accelerate routine disease, therapeutic and toxicity studies. This will reduce processing times from several months to several weeks. It is expected that the promotion of these methods to the broader research community will overcome past resistance to animal-free innovations that were hitherto unable to be adopted due to greater expense or complexity.

Dr Guillermo Gomez, Prof Stuart Pitson, Dr Lisa Ebert, Prof Claudine Bonder, A/Prof Michael Samuel, Tissue Architecture and Organ Function Laboratory, Molecular Signalling Laboratory, Translational Oncology Laboratory, Vascular Biology Laboratory and Tumour Microenvironment Laboratory, Centre for Cancer Biology, South Australia Pathology and University of South Australia, SA.

Bio-fabrication and Organoid Screening Facility for Precision Neuro-oncology in SA: Enabling "animal-free" pre-clinical drug screenings using a clinically relevant human model to replace animal models and to provide a more personalised treatment for brain cancer patients.

Creating a platform to replace the use of animals for human-disease modelling and eliminating the use of animal derived hydrogels (eg Matrigel) for tissue explant culture. Patient-derived tumour organoids and tumour explants culturedfrom resected tumour tissue now constitute better model systems to study the response of tumour cells to

different treatments and provide meaningful clinical results within a relevant time frame. Specific software for image analysis and quantitation of drug responses will be engineered to fully automate reporting for superior and more personalised treatments for brain cancer patients.

A/Prof Michael O'Connor, Dr Rachel Shparberg and Ms Sophia Moriam, Regenerative Medicine Laboratory, School of Medicine and Translational Health Research Institute, Western Sydney University, NSW.

Use of human micro-lenses as an animal-replacement assay for drug development.

This project addresses critical cell culture barriers that currently limit application of human micro-lenses to commercial anti-cataract drug screening. Three-dimensional human tissues generated from pluripotent stem cells hold significant potential for replacing the use of animals (mice, rabbits and dogs) in human drug development. For this potential to be realised, laboratory-scale stem cell models must be scaled-up to meet the operational needs of drug development companies.

Dr Sam Henderson, Prof Andrea Yool and Prof Boris Martinac, Faculty of Health and Medical Sciences, University of Adelaide, SA and Mechanobiology Laboratory, Victor Chang Cardiac Research Institute, NSW.

Replacing frog oocytes and animal antibodies in aquaporin research: Developing an animal-free analysis platform to assess human Aquaporin-1 channels involved in cancer cell migration.

This project will develop novel, animal-free methods to better investigate the function and regulation of a human water and ion channel (AQP1) which is involved in cancer metastasis. This new model is a powerful approach that surpasses animal-based methodology in terms of anticipated reproducibility, accuracy and detail, making a significant difference in the lives of people with cancers. Genetically engineered yeast cells will be harnessed to synthesise human AQP1 protein, which will be validated using animal-free recombinant antibodies, thus obviating the use of frog oocytes and animal antibodies. These methods could be used to investigate other classes of human membrane proteins in the future.

Dr Matthew Baker, Dr Shelley Wickham, Dr Michael Booth and Prof Hagan Bayley, School of Biotechnology and Biomolecular Science, University of New South Wales, School of Chemistry, University of Sydney, NSW and Chemistry Research Laboratory, University of Oxford, UK.

Total Recall: scalable, controllable synthetic tissues to replace animal models for use in membrane protein research.

Building platforms for tissue modelling that replace animal research in physiology and pathology and allow researchers to do 'bottom-up' synthetic biology instead of animal experiments. Taking these methods to the next level to create tissue-scale replacements for animal models where not only membrane proteins can be interrogated, but also at the systems level where tissue-level complexes can be tested in *in vitro* systems. This project will produce data showing how effective the model system is in mimicking complex multicellular tissue for research on ion channels and drugs that target these channels.

Dr Shaun Gregory, A/Prof Vincent Pellegrino, Dr Andrew Stephens, Dr Tim Byrne and Ms Rezan Al-Jafary, Cardio-Respiratory Engineering and Technology Laboratory, Mechanical and Aerospace Engineering, Monash University and Baker Heart and Diabetes Institute, Alfred Hospital, VIC.

Replacing porcine and ovine experiments with an advanced in-vitro patient simulator for training of extracorporeal membrane oxygenation implantation to support patients suffering from cardiovascular and respiratory diseases including COVID-19.

Developing high quality laboratory-based, non-animal training tools for the implantation of extracorporeal membrane oxygenation (ECMO). Using novel combinations of synthetic materials to create life-like tissue-mimicking models that look, feel, and work just like body tissue (eg skin, fat, muscle, blood vessels). Researchers are creating artificial blood circuits and incorporating these circuits and tissue models into patient manikins to create full patient simulators for ECMO implantation training. These manikins will be made available globally to support the elimination of animals (eg pigs and sheep) used for ECMO training.

A/Prof Brian Fry and Dr Christina Zdenek, School of Biological Sciences, Faculty of Science, University of Queensland, QLD.

MAWA RESEARCH & DEVELOPMENT GRANTS

Replacing the use of animals for the study of the alpha-5 subunit of nicotinic acetylcholine receptors and to probe the drug potential of novel ligands for colitis and anti-smoking drugs.

This project will develop a high-throughput, economical, animal-free way of testing in neuronal medical research that allows for robust investigations into human nerve channels. Researchers will develop a specific assay to study human nicotinic acetylcholine receptors, a type of nerve receptor involved in critical human physiological pathways and deleteriously affected by a myriad of diseases and health conditions. These innovative methods will be used to test the human-health effects of changes in the alpha-5 nicotinic acetylcholine receptor, a target of great medical interest, and to determine the therapeutic potential of novel ligands that selectively bind to this therapeutically important receptor.

Dr Antony McNamee, Prof Geoff Tansley, Dr Michael Simmonds and Dr Jo Pauls, Biorheology Research Laboratory, Menzies Health Institute Queensland, School of Engineering and Built Environment, Griffith University and Innovative Cardiovascular Engineering and Technology Laboratory, Critical Care Research Group, The Prince Charles Hospital, QLD.

Eliminating animal blood products from the development of artificial organs: building evidence for an optimal international standard.

Artificial organs for circulatory support are lifesaving systems that support or replace the function of failing organs in critically ill patients. However, increasing evidence implicates sub-optimal system design as the likely cause of blood damage. Development processes of artificial organs are limited by international standards that recommend the use of *bovine* blood for predicting success of *human* life-support systems. Due to inherent biological differences, it is likely that current bovine recommendations have resulted in the development of nonrepresentative models of blood-device compatibility. Thus, the aim of this project is to gather the necessary data to propose a substantial revision to the international standards that currently recommend the use of animal products in place of human products.

Dr Guillermo Gomez, Dr Mariana Oksdath Mansilla, Dr Dario Arrua, Prof Ernst Wolvetang, A/Prof Leonie Quinn, A/Prof Oliver Sieber and Prof Bryan Day, Tissue Architecture and Organ Function Laboratory, Centre for Cancer Biology, Future Industries Institute, University of South Australia, SA, Stem Cell Engineering Laboratory, UQ Centre in Stem Cell Ageing and Regenerative Engineering, Australian Institute for Bioengineering and Nanotechnology, University of Queensland, QLD, Brain Cancer Model and Therapeutics Group, John Curtin School of Medical Research, Australian National University, ACT, Personalised Oncology Division, Walter and Eliza Institute of Medical Research, VIC, and Sid Faithfull Brain Cancer Laboratory, Queensland Institute for Medical Research, QLD.

A national effort for large-scale pre-clinical test of combination therapy for glioblastoma using brain organoids and the entire FDA-approved drug catalogue: A new platform to replace animal models in brain cancer research.

Brain Cancer research has been fundamentally performed using animal xenograft models on which patient derived cells are injected into the brains of mice. To develop new and better models of human brain disease there is an urgent need to replace the use of animals for brain cancer research. By injecting patient derived brain cancer cells into human brain organoids in combination with innovative culture media conditions for brain tissue, a new system has been developed to provide an ideal model to study how brain cancer cells grow and develop within the human brain. This approach provides an ideal solution to eliminate the use of animals for brain cancer research.

Prof Ben Corry and Dr Amanda Buyan, Transport Proteins and Computational Biophysics Group, Research School of Biology, Australian National University, ACT.

Developing sodium channel inhibitors for the treatment of chronic pain and epilepsy, replacing the need for in-vivo and animal testing.

Local anaesthetics and many anti-epileptics work by blocking nerve impulses in different parts of the body. However, both of these types of medication can cause side effects because they have multiple targets in the body - not just the desired target. The aim of this project was to understand how to make these medications more target specific to reduce side effects and to design drugs for treating chronic pain. Computer simulations provide a better understanding of how these drugs work, replacing animal testing that causes immense suffering because the main target of this research is new pain medications, so animal testing usually involves intentionally inflicting pain upon the experimental animals.

MAWA RESEARCH & DEVELOPMENT GRANTS

A/Prof Cedric Bardy and Dr Zarina Greenberg, Mind and Brain Theme, South Australian Health and Medical Research Institute (SAHMRI), Centre for Cancer Biology and Royal Adelaide Hospital, SA.

Using advanced RNA sequencing technologies for profiling primary glioblastoma brain tumours to identify therapeutic targets in relapsed patients as a replacement for xenograft animal models.

In this project researchers used advanced technologies as an alternative approach to animal models to study the composition and profile of glioblastoma, the most common and aggressive brain cancer. Different cells within the tumour of a patient were identified that show genetic susceptibility to treatments. This was performed as part of a clinical trial of a new cell therapy targeting a particular subset of tumour cells. Machine learning was used for all the data generated in this study to predict patient outcomes and survival, and to identify the measures most predictive of patient outcomes. These measures have shed light on how the diversity of cell types within a tumour hinders current treatments and possible susceptibilities of cell populations.

Prof Stefan Broer and Dr Stephen Fairweather, Membrane Physiology and Epithelial Transporters Group, Research School of Biology, Australian National University, ACT.

Development of a new membrane protein expression system to replace the use of Xenopus laevis in novel drug development for the treatment of type II diabetes mellitus and glutamine-dependent carcinomas.

In this project researchers developed an alternative set of approaches to discover novel pharmaceuticals by looking at the molecules and their drug-like properties to overcome side-effects and toxicity often not picked up in animal-based pre-clinical screening, and to make drug discovery much faster and cheaper. In order to optimise molecular approaches there is a need to continually search for the cleanest, most chemically isolated approaches. This technique of cell-free target synthesis is one such technique that can replace invasive animal models at the initial discovery phase and can remove the need for later phase animal trials.

MAWA EQUIPMENT GRANTS

Dr Guillermo Gomez, Prof Stuart Pitson, Dr Lisa Ebert, Prof Claudine Bonder, A/Prof Michael Samuel, Tissue Architecture and Organ Function Laboratory, Molecular Signalling Laboratory, Translational Oncology Laboratory, Vascular Biology Laboratory and Tumour Microenvironment Laboratory, Centre for Cancer Biology, South Australia Pathology and University of South Australia, SA.

Funding towards the purchase of BioAssemblyBot and BioStorageBot robotic solutions, recently developed by Advanced Solutions Life Sciences. BioAssemblyBot is an intelligent robot used by life scientists to build biological structures - mimicking a human arm to bioprint, pick up biology, and complete complex assays. BioStorageBot is an automated conveyance and controlled storage system for biological specimens.

This equipment provides the scalability and automation capabilities that can be combined with recent developments in culturing patient-derived glioblastoma explants (GBE) to maintain the cellular and genetic composition of the primary tumour. The new equipment will also allow the establishment of automated pipelines for testing and optimization of treatments for brain cancer as well as other cancers including colorectal cancer, breast cancer and pancreatic cancer, and will be the first of its kind in Australia.

MAWA POST-DOCTORAL RESEARCH FELLOWSHIPS

Dr Charles Cox as MAWA VCCRI Fellow, Mechanosensory Biophysics Laboratory, Molecular Cardiology and Biophysics Division, Victor Chang Cardiac Research Institute, NSW.

Dr Cox is studying complex ion channels to characterize a new molecule identified in the malaria parasite to understand its structure and molecular function to develop novel rationally designed anti-malarial therapeutics. This research will use animal-origin-free purification techniques and a new non-animal growth media completely replacing the use of foetal bovine serum traditionally used in these studies.

Dr Bhim Rai as MAWA ANU Fellow, The Maddess Group, Diagnostics for Eye Disease, Eccles Institute for Neuroscience, John Curtin School of Medical Research and Diabetes Research Laboratory, Australian National University, ACT.

MAWA POST-DOCTORAL RESEARCH FELLOWSHIPS

Dr Rai is contributing to the development of a non-invasive visual field testing machine based on pupillography to replace the use of large animals such as non-human primates and cats in invasive vision experiments. Current research studies include vitreoretinal diseases such as age-related macular degeneration, diabetic retinopathy, macular oedema, diabetic peripheral neuropathy, glaucoma, multiple sclerosis, Alzheimer's disease and concussion injuries.

Dr Rachel Shparberg as MAWA WSU Fellow, Regenerative Medicine Laboratory, School of Medicine and Translational Health Research Institute, Western Sydney University, NSW.

Dr Shparberg is using human micro-lenses as an animal-replacement assay for anti-cataract drug development. Human pluripotent stem cells possess the ability to produce large quantities of normal or diseased human tissue in the laboratory, providing an opportunity to replace animals (mice, rabbits and dogs) in preclinical drug development. For this potential to be realised, specific drug screening and toxicology assays will be developed that fulfil the industrial-scale needs of drug development companies.

Dr Antony McNamee as MAWA Griffith Fellow, Disability and Rehabilitation, Menzies Health Institute Queensland, Griffith University, QLD.

Dr McNamee's aim is to demonstrate that human blood, rather than bovine blood, must be used for artificial organ haemocompatibility testing to improve the outcomes and quality of life of patients receiving artificial organ therapies. Dr McNamee and colleagues are gathering the necessary data to propose a substantial revision to current international standards in that future devices must be designed, tested, and optimised for humans because bovine blood is a poor model of human tissue and should not be used as a surrogate.

Dr Marina Oksdath Mansilla as MAWA UniSA Fellow, Tissue Architecture and Organ Function Laboratory, Centre for Cancer Biology, University of South Australia, SA.

Dr Oksdath Mansilla's expertise in tissue culture and material sciences enabled her to contribute to the development of a new brain organoid platform using a combination of therapies to efficiently target the different type of cells within patients' tumours. This approach eliminates the need for animal xenograft models for brain cancer research and the need for animal-derived culture media in which to grow brain tissue. This animal-free model of brain cancer much more accurately reflects the physiology and genetics of the human brain.

Dr Amanda Buyan as MAWA ANU Fellow, Transport Proteins and Computational Biophysics Group, Research School of Biology, Australian National University, ACT.

Dr Buyan worked extensively on computer simulations of drug interactions with sodium channels making medications more target specific to reduce side effects and to increase their therapeutic potential. The aim of this project was to understand how current local anaesthetics work at a molecular level to provide a pathway for developing more targeted medications for the treatment of chronic pain and epilepsy, and for replacing animal testing which involves intentionally inflicting pain on the experimental animals, with computational studies.

Dr Zarina Greenberg as MAWA SAHMRI Fellow, Mind and Brain Theme, South Australian Health and Medical Research Institute (SAHMRI),

Dr Greenberg used advanced RNA sequencing technologies for profiling primary glioblastoma brain tumours to identify therapeutic targets in relapsed patients as a replacement for xenograft animal models. This cutting-edge technology was used to study the composition and profile of glioblastoma to identify different cells that showed genetic susceptibility to treatments to improve patient outcomes. The project revealed how the diversity of cell type within a tumour hinders current treatments and identified possible susceptibilities of cell populations.

Dr Stephen Fairweather as MAWA ANU Fellow, Membrane Physiology and Epithelial Transporters Group, Research School of Biology, Australian National University, ACT.

Dr Fairweather has contributed to the development of an alternative set of approaches, including a new membrane protein expression system, to remove the dependence on invasive animal derived models such as *X.laevis* oocytes to discover novel pharmaceuticals for the treatment of type II diabetes and glutamine-dependent carcinomas. This technique of cell-free target synthesis is expected to replace animal models at the initial discovery phase and remove the need for later phase animal trials, making drug discovery much faster and cheaper.

MAWA DOCTORAL INTERNSHIPS

Ms Nicole Wheatley, School of Chemistry and Molecular Biosciences, University of Queensland, QLD.

Ms Wheatley contributed to MAWA's core work as an intern and represented MAWA at various forums while completing her doctoral studies. Her interactions with MAWA-supported researchers beyond her discipline and potential MAWA partners, as well as her participation in a series of webinars, expanded her knowledge of alternatives and understanding of the extent to which cutting edge fundamental research can be undertaken without animals.

MAWA DOCTORAL RESEARCH SCHOLARSHIPS/FUNDING SUPPORT

Ms Jeeeun (Alice) Shin, Division of Biomedical Science and Biochemistry, Research School of Biology, Australian National University, ACT.

Ms Shin is contributing to the replacement of animal derived antibodies with synthetic nanobodies which have been engineered to be smaller, more stable and more easily modified. They are, therefore, more useful and versatile for use in the fields of structural biology, biotechnology and therapeutic research and medical procedures including medical imaging and disease diagnosis, important steps in drug discovery and design.

Ms Ana Nunez-Nescolarde, Monash Biomedicine Discovery Institute, Department of Anatomy and Developmental Biology, Monash University, VIC

Ms Nunez-Nescolarde is developing human kidney organoids to replace animal models of renal fibrosis as well as the range of animal products used in stem cell culture and the visualisation of experimental results. This project will test the capacity of human-derived organoids to model kidney injury and replicate the early sequence of events that initiate a fibrotic response in humans.

Mr Simon Maksour, School of Chemistry and Molecular Bioscience, Faculty of Science, University of Wollongong, NSW.

Mr Maksour is one of two PhD students contributing to the establishment of a platform of cost-effective animal-product-free methods for direct conversion of human patient biopsy samples to cell types relevant to neurological disease, and rapid animal-free downstream analysis. It is expected that the promotion of these methods to the broader research community will overcome past resistance to animal-free innovations that were hitherto unable to be adopted due to greater expense or complexity.

Ms Michelle Newbery School of Chemistry and Molecular Bioscience, Faculty of Science, University of Wollongong, NSW.

Ms Newbery is one of two PhD students contributing to the establishment of a platform of cost-effective animal-product-free methods for direct conversion of human patient biopsy samples to cell types relevant to neurological disease, and rapid animal-free downstream analysis. It is expected that the promotion of these methods to the broader research community will overcome past resistance to animal-free innovations that were hitherto unable to be adopted due to greater expense or complexity.

Ms Thanh Huyen Phan, Sydney Nano Institute and Sydney School of Pharmacy, Faculty of Health and Medicine, University of Sydney, NSW.

Ms Huyen Phan is combining data from human physiology-mimicking organ-on-chip models with artificial intelligence for the development of new treatments for lung disease and injury including COVID-19. The development and validation of lung-on-chip models is expected to replace the use of animal models for testing the efficacy of new generation therapeutics to regenerate lungs damaged by disease and injury.

Ms Rezan Al-Jafary, Cardio-Respiratory Engineering and Technology Laboratory, Mechanical and Aerospace Engineering, Monash University, VIC.

Ms Al-Jafary is contributing to the creation of life-like tissue-mimicking patient manikins to create advanced patient simulators used for training in medical implantation to support patients suffering from cardiovascular and respiratory diseases, including COVID-19. These manikins will be made available globally to replace the use of pigs and sheep that are traditionally used for this training.

MAWA DOCTORAL RESEARCH SCHOLARSHIPS/FUNDING SUPPORT

Mr Anton Nathanson, Graduate School of Biomedical Engineering, Faculty of Engineering, University of New South Wales, NSW.

Mr Nathanson is using multibeam scanning electron microscopy, machine learning and geospatial information systems for the development of diagnostics for degenerative diseases of the bone and brain to replace large animal models. These methods will create new early detection diagnostic technologies to shift away from predominately animal-based discovery in degenerative research, particularly osteoarthritis and Alzheimer's Disease.

Dr Christina Limantoro, Sydney Nano Institute and Sydney School of Pharmacy, Faculty of Health and Medicine, University of Sydney, NSW.

Dr Limantoro has been awarded her PhD for work on the development of antimicrobial nanoparticles for wound healing applications to replace the traditional use of animals in this field of biomedical research and testing. She contributed to the development of skin-on-chip and injury-on-chip models to test both toxicity and efficacy of the nanoparticles to aid wound healing and to inhibit bacterial growth.

MAWA MASTERS RESEARCH SCHOLARSHIPS

Ms Isabelle Harris, Faculty of Medicine, University of Melbourne and Department of Neurology, St Vincent's Hospital, VIC.

Ms Harris investigated epilepsy using a combination of mathematical modelling and voluntary human data including imaging techniques as a viable replacement for animal models. The diagnosis and treatment of epilepsy is highly patient specific, therefore it is necessary to study the relationship between a patient's brain structure (connectivity) and dysfunction (seizure dynamics) and how particular network connectivities influence seizure initiation and termination.

Ms Qingyu (Sarah) Lei, Sydney Nano Institute and Sydney School of Pharmacy, Faculty of Health and Medicine, University of Sydney, NSW.

Ms Lei contributed to the establishment of skin-on chip and skin injury-on-chip models - a microfluidic platform for nanotoxicity, drug discovery, and precision biology to replace the use of animals in this field of research. These models will underpin fundamental studies of nanoparticle penetration through the skin and their impact on skin function to establish the safety of nanoparticles used in sunscreens, cosmetics and other topical formulations.

MAWA HONOURS RESEARCH SCHOLARSHIPS

Ms Laura Shuttleworth, Nitsche Group, Research School of Chemistry, College of Sciences, Australian National University, ACT.

Ms Shuttleworth is contributing to the development of new anti-viral drug candidates to inhibit the replication of SARS-CoV-2 without the use of animal models and animal-derived antibodies to assess toxicity and effectiveness of conventional drugs in pre-clinical studies. Her project uses synthetic organic chemistry, computational models and X-ray crystallography to replace microbial systems using animal products for producing large libraries of peptide-based inhibitors of random sequences.

Ms Yosha Pathmaperuma, Division of Biomedical Science and Biochemistry, Research School of Biology, Australian National University, ACT.

Ms Pathmaperuma is contributing to the replacement of animal derived antibodies used by the medical research community with synthetic nanobodies using yeast surface display. These synthetic nanobodies are superior due to their small size, stability and amenability to alteration via standard molecular biology tools. They can be used in the fields of structural biology, biotechnology and medicine for disease diagnosis and drug discovery and design.

Mr Jonathan King, Saliba Group - Physiology and Biochemistry of the Malaria Parasite, Research School of Biology, College of Sciences, Australian National University, ACT.

Mr King has contributed to the replacement of mouse models of human tuberculosis infections which are extensively used in TB research despite a number of recognised limitations, by culturing *M. tuberculosis in vitro* and by replacing the use of bovine serum albumin which is traditionally used in the growth medium. This work is assisting with developing novel shortened TB treatments to overcome compliance problems and increasing drug resistance.

MAWA INTERNATIONAL CONFERENCE GRANTS

Prof Wojciech Chrzanowski, Sydney Nano Institute and Sydney School of Pharmacy, Faculty of Health and Medicine, University of Sydney, NSW.

10th International Conference on Materials for Advanced Technologies Conference (ICMAT2019), Marina Bay Sands, Singapore.

ChinaNANO International Conference on Nanoscience and Technology, Beijing, China

11th World Congress on Alternatives and Animal Use in the Life Sciences (WC11), Virtual, Maastricht, Netherlands.

Dr Andre Peterson, Department of Electrical & Electronic Engineering, University of Melbourne and Department of Neurology, St Vincent's Hospital, VIC.

11th World Congress on Alternatives and Animal Use in the Life Sciences (WC11), Virtual, Maastricht, Netherlands.

Dr Guillermo Gomez, Tissue Architecture and Organ Function Laboratory, Centre for Cancer Biology, South Australia Pathology and University of South Australia, SA.

11th World Congress on Alternatives and Animal Use in the Life Sciences (WC11), Virtual, Maastricht, Netherlands.

Dr Antony McNamee, Disability and Rehabilitation, Menzies Health Institute Queensland, Griffith University, QLD.

11th World Congress on Alternatives and Animal Use in the Life Sciences (WC11), Virtual, Maastricht, Netherlands.

Ms Isabelle Harris, Faculty of Medicine, University of Melbourne and Department of Neurology, St Vincent's Hospital, VIC.

11th World Congress on Alternatives and Animal Use in the Life Sciences (WC11), Virtual, Maastricht, Netherlands.

Ms Michaela Vranic-Peters, Faculty of Medicine, University of Melbourne and Department of Neurology, St Vincent's Hospital, VIC.

11th World Congress on Alternatives and Animal Use in the Life Sciences (WC11), Virtual, Maastricht, Netherlands.

Ms Thanh Huyen Phan, Sydney Nano Institute and Sydney School of Pharmacy, Faculty of Health and Medicine, University of Sydney, NSW.

Asian Pacific Societies for Extracellular Vesicles (APSEV) Inaugural Meeting and Korean Society for Extracellular Vesicles (KSEV) Annual Meeting, Jeju, Republic of Korea.

17th International Conference on Biomedical Engineering (ICBME), National University of Singapore, Singapore.

International Society of Extracellular Vesicles (ISEV) Virtual Conference, Mount Royal, New Jersey, USA.

Ms Sarah Lei, School of Pharmacy, Faculty of Health and Medicine, University of Sydney, NSW.

American Association of Pharmaceutical Scientists Conference (AAPS2019), San Antonia, Texas, USA.

MAWA INTERNATIONAL TRAVEL GRANTS

Dr Amy Li and Dr Sean Lal, Sydney Heart Bank and Department of Anatomy and Histology, Sydney School of Medical Sciences, University of Sydney, NSW

Drs Li and Lal travelled internationally to visit various BioBanks to observe their collection, tissue processing and storage protocols and to collaborate worldwide to ascertain the best methods for harvesting, preserving and maintaining high quality human heart tissue for human models of cardiovascular disease.

MAWA DOMESTIC CONFERENCE GRANTS

Dr Christina Limantoro, Sydney Nano Institute and Sydney School of Pharmacy, Faculty of Health and Medicine, University of Sydney, NSW.

Safe by Design - Sustainable Nanotechnology Symposium, Sydney, NSW.

Dr Stephen Fairweather, Membrane Physiology and Epithelial Transporters Group, Research School of Biology, Australian National University, ACT.

Australian and New Zealand Council for the Care of Animals in Research and Teaching (ANZCCART) Conference, Canberra, ACT.

Dr Amanda Buyan, Transport Proteins and Computational Biophysics Group, Research School of Biology, Australian National University, ACT.

Australian and New Zealand Council for the Care of Animals in Research and Teaching (ANZCCART) Conference, Canberra, ACT.

Ms Thanh Huyen Phan, Sydney Nano Institute and Sydney School of Pharmacy, Faculty of Health and Medicine, University of Sydney, NSW.

28th Australian Society for Medical Research (ASMR) Virtual Conference, Sydney, NSW.

Ms Sarah Lei, School of Pharmacy, Faculty of Health and Medicine, University of Sydney, NSW.

Pharmacy Postgraduate Research Showcase, Sydney, NSW

MAWA SPONSORSHIPS OF CONFERENCES

Joint Australian Physiological Society (AuPS) and The Australian Society for Biophysics (ASB) Annual Scientific Meeting, Canberra, ACT.

Sponsorship of Alternatives to the Use of Animals in Medical Research Symposium, International Conference Travel Grant and Student Prizes.

Molecular Modelling Australasia Conference (MM2021), Hybrid Conference, Brisbane, QLD.

Conference Sponsorship and Student Prizes

MAWA SPONSORSHIPS OF INTERNATIONAL SPEAKERS

Prof Eduardo Perozo, Biochemistry and Molecular Biology, Institute for Biophysical Dynamics, University of Chicago, USA.

International Keynote Speaker for the Joint Australian Physiological Society (AusPS) and the Australian Society for Biophysics (ASB) Annual Scientific Meeting, Symposium: Alternative Approaches to the Use of Animals in Physiology and Biophysics, Canberra, ACT.

MAWA CONFERENCE PRIZES

MAWA Student Prizes for the joint Australian Physiological Society (AusPS) and Australian Society for Biophysics (ASB) Annual Scientific Meeting, Canberra, ACT and Molecular Modellers Australasia Conference, Brisbane, QLD.

MAWA AMBASSADORS

Dr Antony McNamee, MAWA Fellow from Griffith University, **Dr Emelie Flood,** Post-Doctoral Researcher from RMIT University**, Ms Jade Vennetti,** RAMR Junior Fellow from the Australian National University and **Ms Nicole Wheatley** from the University of Queensland were invited to take on MAWA Ambassadorial roles and to join MAWA's growing network of ambassadorial representatives.

MAWA's Ambassadors represented the Trust and presented at a variety of events throughout 2019, 2020 and 2021, including university careers nights, a range of conferences, seminars, meetings and symposiums, plus events organised by other not-for-profit foundations, funding bodies, charities, and both Australian and state government departments.

THE MAWA TRUST

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MAWA ANU Fellows: Dr Rong Chen; Dr Michael Thomas; Dr Stephen Fairweather; Dr Amanda Buyan **MAWA ANU JCSMR Fellow:** Dr Bhim Rai
MAWA UMelb Fellow: Dr Andre Peterson **MAWA UMelb CERA Fellow:** Dr Raymond Wong **MAWA USyd Fellow:** Dr Belal Chami
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MAWA UniSA Fellow: Dr Mariana Oksdath Mansilla **MAWA WSU Fellow:** Dr Rachel Shparberg
MAWA SAHMRI Fellow: Dr Zarina Greenberg **MAWA Griffith Fellow:** Dr Antony McNamee

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