Derivation and Characterisation of induced pluripotent stem cell lines from patients with X-Linked Alport syndrome – a model for examining mechanisms and therapies

Dr. Judy Savige, University of Melbourne (Australia)
Dr. Sharon Ricardo, Monash University (Australia)
Awarded 2013

The aim of this research project is to determine how nonsense and missense mutations cause disease in X-linked Alport syndrome using podocyte cell lines derived from induced pluripotent stem (iPS) cells from affected Individuals. Dr. Savige and Dr. Ricardo will apply their established iPS technology to generate kidney podocytes from Alport patient-derived iPS cells for use in disease modelling, and drug development and screening. Reprogramming of adult cells to generate cell lines represents a major advance in the development of disease models. These cells will help in understanding disease pathogenesis, screening new treatments, and offering possibilities for regenerative medicine. iPS cells are immature and can be maintained in culture indefinitely. Dr. Savige and Dr. Ricardo have reported a differentiation protocol to efficiently generate kidney podocytes from human iPS cells and propose here to generate iPS cells with podocyte characteristics derived from patients with Alport syndrome. Alport syndrome results from a podocyte genetic mutation, therefore *in vitro* kidney disease modelling based on iPS-derived podocytes provides a powerful tool in which the cellular defects underlying Alport syndrome can be investigated and used for screening therapeutic compounds. Understanding these mechanisms is the first step in developing novel disease-modifying treatments in order to provide benefits for the community from research discovery.

Dr. Savige and Dr. Ricardo hypothesize that any increase in the amount of the collagen IV a3a4a5 network in Alport-derived kidney cells has a beneficial effect on glomerular basement membrane (GBM) structure and integrity, and delays the onset of renal failure.

Specific Aims and Objectives are:

1. To generate iPS cells from patients with X-linked Alport syndrome and non-Alport normals and to characterise these cell lines for podocyte features.

2. To determine the effects of Alport mutations on collagen IV a1-a6 mRNA and protein levels in the iPS-derived podocytes. To confirm the same effect on mRNA levels in the urine from the corresponding patients. To determine the baseline effect of missense mutations on the ‘unfolded protein response’ and endoplasmic reticulum (ER) stress, apoptosis and autophagy, and of nonsense mutations on ‘nonsense-mediated decay’.

3. To investigate the effect of treatments targeting nonsense and missense mutations in Alport derived cells on the levels of collagen IV a1-a6 chain mRNA and chain synthesis. Potential therapies include angiotensin converting enzyme (ACE) inhibitors in addition to protein synthesis inhibitors in cell lines from patients with nonsense mutations, and of chemical chaperones in cell lines from patients with missense mutations. These experiments will also determine whether these treatments have a deleterious effect on the cells by increasing the unfolded protein response and ER stress, or apoptosis and autophagy.