Project Title: The role of altered monocyte activity in the long-term potential of peritoneal dialysis as a therapy

Acronym: ROAMinPD

Summary

Worldwide, >250,000 individuals rely on peritoneal dialysis (PD) as a major modality of renal replacement therapy for treating end-stage renal failure. Despite treatment advance in the past three decades, PD-related peritonitis remains one of the major causes for treatment failure as well as patient mortality. Despite the general acceptance that monocyte/macrophages are a frontline component of peritoneal host defense to infection, the immunobiology of this key cell population remains poorly understood.

Summary of Project Objectives

The aim of our project was to:

1. To complete a contemporary systematic and detailed study of the impact of peritoneal dialysis on the monocyte/macrophage system, including the identification of corresponding subsets between mouse and human.
2. To assess the impact of this altered cell biology on functional responses of PD monocytic cells to infectious challenge, the regulation of immunity and tissue damage.

Description of work performed and results so far

In this study, we applied contemporary immunological techniques to perform a full phenotypic and functional characterisation of distinct peritoneal monocytic subsets through the course of PD therapy, from one week after PD catheter implantation to 6 months after the dialysis commencement. Firstly, we have performed a detailed immunophenotyping as well as expression profiles of relevant surface markers on peritoneal monocytic cells from PD patients at different stages by advanced multi-colour flow cytometry. Our findings indicated human peritoneal monocyte/macrophages comprise several discrete subpopulations, which cannot be simply classified into a single conventional population, i.e. ‘$CD14^+$ cells’. Individual subsets had differential marker profiles representing various stages of macrophage activation, differentiation and maturation. More importantly, the numbers and the distributions of distinct macrophage subsets were altered by the dialysis process. We have observed, along the course of PD therapy, the decrease in total number of

peritoneal macrophages, though their proportion within total peritoneal leukocytes increase considerably. Additionally, our data suggested circulating blood monocytes are recruited into peritoneal cavity, where they became more mature. However, this process has been perturbed by continuous dialysis intervention as peritoneal macrophages resemble blood monocytic phenotype.

Secondly, we have conducted a variety of functional assays to better characterize the distinct peritoneal macrophage subsets we have identified. The functional analysis included *ex vivo* phagocytosis capacity assay, respiratory burst assay to detect reactive oxygen species generation upon microbial challenges, intracellular cytokine production (e.g. TNF-α, IL-1β, IL-6) in response to exogenous stimuli, and *in vitro* antigen presentation/T cell stimulation assay. The results, in general, revealed that human peritoneal macrophages from PD patients retain a wide range of functional capabilities. However, differential functional profiles have been noted among the distinct subpopulations, i.e. some with relatively poor phagocytic properties, but with superior antigen presentation ability (more “dendritic cell-like”). This functional diversity suggested individual monocytic subsets may play different roles upon the host-pathogen interaction and in the process of inflammation/resolution within the peritoneal cavity.

Final results and potential impact

Our research work has clearly demonstrated that the dialysis process perturbs the tightly regulated homeostatic environment in normal human peritoneal cavity, and also change the phenotypes and function of peritoneal monocyte/macrophage population. This alteration would not only modify the host immune response to bacterial infection, but also might have a link to long-term peritoneal membrane injury. Overall, the major contribution of this study is to advance our knowledge of macrophage biology in the human peritoneal cavity, particularly in patients undergoing PD. Our novel findings from this research project have made a significant step forward, not only in understanding the alterations to monocyte/macrophage biology in the peritoneal cavity of the dialysis patient, but also into helping to understand how the immune system responds to infection, a key clinical risk associated with PD. The results have been influential in securing substantial additional research funds to continue and expand our work in this area. It is hoped that collectively these studies will provide prognostic and mechanistic data that will be of benefit to PD patients and the long-term potential of PD as a therapy in chronic kidney disease.