Abstract

Metabolomic Insights into the Pharmacological and Genetic Inhibition of Cyclooxygenase-2

William T. E. Briggs

The cyclooxygenase (COX)-2 inhibitors, or “coxibs,” are excellent anti-inflammatory agents, but their reputation has been tarnished by the adverse cardiovascular (CV) events, including heart failure (HF), with which they are associated. Whilst the risk of HF represents the greatest adverse CV event signal seen with these compounds, it is also perhaps the least well understood and has often been explained away as a consequence of the thrombotic risk with which the coxibs are also associated. One recent hypothesis, put forward by Ahmetaj-Shala et al., suggests that asymmetric dimethylarginine (ADMA) may serve as a mechanistic bridge between COX-2 inhibition and HF. However, the ADMA-COX-2 hypothesis was developed based on findings in a constitutive mouse model of COX-2 knock-out (KO), which is compromised by severe developmental cardio-renal pathology, and pharmacological studies which may not accurately reflect coxib use in clinical practice.

Various studies have explored the metabolic changes induced by coxib treatment. However, these studies have been limited in scope and have tended to focus on specific pathways or certain tissues/bio-fluids. This has left large regions of the metabolome, in the context of coxib-treatment, unexplored. Given that metabolic remodelling is a key feature of HF, changes in these metabolites may hold the key to understanding the pathogenesis of coxib-induced HF.

L-Carnitine shuttles activated long-chain fatty acids (FAs) across the inner mitochondrial membrane to the mitochondrial matrix, where they are oxidised by β-oxidation. This is especially important in the heart, which derives the majority of its energy from the metabolism of FAs. Changes in carnitine metabolism are also seen in HF. It is therefore biologically plausible that derangements in carnitine metabolism may contribute to the pathogenesis of coxib-induced HF.

This thesis employs a combination of targeted and untargeted metabolomic techniques, stable isotope labelling and quantitative reverse transcription polymerase chain reaction (RT-qPCR) to i) profile the metabolic changes induced by celecoxib and rofecoxib, in the mouse; ii) specifically interrogate the effect of celecoxib, rofecoxib and global COX-2 gene deletion on carnitine synthesis, metabolism and shuttling, and iii) explore the advantages and disadvantages of the inducible post-natal global (IPNG) COX-2 KO (COX-$2^{-/-}$) mouse, an alternative to the constitutive COX-$2^{-/-}$ mouse used by Ahmetaj-Shala et al.

The results of this thesis demonstrate that i) celecoxib and rofecoxib have similar metabolomic consequences in the mouse; ii) carnitine metabolism may be affected by celecoxib, rofecoxib and dietary composition, via a peroxisome proliferator-activated receptor-alpha (PPAR-α) mediated effect on hepatic carnitine synthesis and iii) the IPNG COX-$2^{-/-}$ mouse neither exhibits the severe developmental cardio-renal pathology nor the altered ADMA metabolism observed in the constitutive COX-$2^{-/-}$ mouse. These findings contradict those of Ahmetaj-Shala et al., oppose the ADMA-COX-2 hypothesis and highlight a potential role for carnitine metabolism and diet in coxib induced HF.