Institutionen för Medicin, Enheten för Reumatologi

Characterization of the PGE$_2$ Pathway in Arthritis and Inflammation: mPGES-1 as a Therapeutic Target

AKADEMISK AVHANDLING
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ABSTRACT

The inducible prostaglandin (PG) E$_2$ pathway is defined by the concerted activities of the enzymes cyclooxygenase (COX)-2 and microsomal prostaglandin E synthase (mPGES)-1 in producing PGE$_2$. PGE$_2$ has pro-inflammatory and immunomodulatory functions and is involved in an array of diseases with an autoimmune and chronic inflammatory component, including rheumatoid arthritis (RA). In RA, mPGES-1 and COX-2 are up-regulated in the inflamed synovium of patients. COX inhibitors like non-steroidal anti-inflammatory drugs (NSAIDs) and selective COX-2 inhibitors (COXibs) relieve inflammation and pain in RA, but their respective GI tract and cardiovascular side effects precluding long-term use, which is essential in chronic inflammation. Moreover, other effective RA therapies like TNF blockade and B-cell depletion therapy leave the inducible PGE$_2$ pathway unaffected, suggesting that targeting this pathway could have additional benefits in a combinatorial approach. mPGES-1 is currently investigated as a target that could dissociate the anti-inflammatory benefits of COX inhibitors from their detrimental side effects. However, most inhibitors of human mPGES-1 activity generated so far fail to inhibit the murine enzyme ortholog, complicating characterization of mPGES-1 inhibition in vivo in relevant disease models.

The first objective of this thesis was to extend the characterization of the PGE$_2$ pathway in arthritis. The second objective was to investigate mPGES-1 as a therapeutic target. The latter was accomplished in part through the development and evaluation of pharmacological inhibitors active on both human and murine mPGES-1.

To fulfill the first objective, we characterized a new mechanism for the induction of PGE$_2$ production in RA synovial fibroblasts (RASFs), whereby complexes of the alarmin high mobility group box protein-1 (HMGB1) and the cytokine IL-1β cause an up-regulation of COX-2 and mPGES-1 expression. We also characterized 15-prostaglandin dehydrogenase (15-PGDH) to co-localize with mPGES-1 and the COX enzymes in the synovium of RA patients, suggesting a concerted activity of the anabolic and catabolic parts of the PGE$_2$ cascade in the joint. In the same patients, we determined that methotrexate therapy did not interfere with the expression of COXs, mPGES-1 or 15-PGDH, adding that therapy to the list of approaches leaving the PGE$_2$ pathway unaffected. Lastly, we could not confirm an association between the expression of COXs, mPGES-1 and 15-PGDH and quantitative pain assessment or arthritis development in arthralgic individuals at risk of developing arthritis or in early arthritis patients. In line with the second objective, we developed compounds II and III, which inhibit PGE$_2$ synthesis in vitro in different human and murine cell assays and in vivo in the air pouch model of acute inflammation. Compound II also reduced edema in the rat adjuvant-induced arthritis model. When the PG profile elicited by mPGES-1 inhibition with compound III was compared to that resulting from mPGES-1 gene deletion in the air pouch model, different results were observed suggesting the two modes of inhibition might not have the exact same outcome. While inhibition of PGE$_2$ synthesis with both compound II and III did not result in the shunting of PGH$_2$ to other prostanoids, a shunt to thromboxane (TX) B$_2$ was observed in the mPGES-1 knockout mouse. We also used the mPGES-1 knockout mouse to investigate the impact of mPGES-1 gene deletion on the eicosanoid and fatty acid profiles in inflammation. We discovered that it resulted in macrophages producing more 15-deoxy-Δ$^{12,14}$ PGJ$_2$ and the spleen containing more eicosadienoic acid (EDA). This suggests mPGES-1 inhibition could not only inhibit the synthesis of the pro-inflammatory PGE$_2$, but also cause the up-regulation of anti-inflammatory pathways.

In conclusion, this thesis further advances the knowledge about the PGE$_2$ synthesis cascade in arthritis and describes two new mPGES-1 inhibitors with an in vivo activity in native rodent models of disease. The latter constitute new valuable tools for the study of mPGES-1 in whichever pathology it has an involvement.