



**Karolinska
Institutet**

Institutionen för kvinnors och barns hälsa

Multiple roles of HMGB1 in clinical and experimental arthritides

AKADEMISK AVHANDLING

som för avläggande av medicine doktorsexamen vid Karolinska Institutet offentligen försvaras på svenska språket i Föreläsningssalen, CMM L8:00, Karolinska Universitetssjukhuset, Solna, Stockholm

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av

Hanna Schierbeck

Huvudhandledare:

Professor Ulf Andersson
Karolinska Institutet
Institutionen för kvinnors och barns hälsa
Enheden för pediatrik

Bihandledare:

Docent Helena Erlandsson Harris
Karolinska Institutet
Institutionen för medicin, Solna
Enheden för reumatologi

Fakultetsopponent:

Professor Hans Carlsten
Göteborgs Universitet
Institutionen för medicin
Enheden för reumatologi och
inflammationsforskning

Betygsnämnd:

Docent Mikael Karlsson
Karolinska Institutet
Institutionen för medicin, Solna
Enheden för translationell immunologi

Docent Johan Rönnelid

Uppsala Universitet
Institutionen för immunologi, genetik och
patologi
Enheden för klinisk immunologi

Docent Jonas Wetterö

Linköpings Universitet
Institutionen för klinisk och experimentell
medicin
Enheden för reumatologi

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SUMMARY

Inflammation can be infectious and/or sterile depending on the initiating event. The proinflammatory mediator High mobility group box protein 1 (HMGB1) is a nuclear protein released from cells during both sterile and infectious inflammation and once extracellular, initiates and potentiates inflammation by inducing cytokine production and by recruiting inflammatory cells.

Autoimmune diseases are characterised by chronic sterile inflammation leading to tissue destruction. HMGB1 has been implicated in the pathogenesis of several autoimmune diseases including rheumatoid arthritis (RA), systemic lupus erythematosus, multiple sclerosis and myositis. The involvement of HMGB1 in arthritis has been shown by overexpression of HMGB1 in RA synovial tissue and synovial fluid, by beneficial outcome of therapeutic HMGB1-blockade in several experimental arthritis models and by the induction of arthritis by intra-articular injection of recombinant HMGB1 into mice.

In this thesis work I set out to investigate the potential role of HMGB1 in juvenile idiopathic arthritis (JIA), to further delineate mechanisms by which HMGB1 can contribute to arthritis pathogenesis and to study the means by which HMGB1 activity can be suppressed.

I could report for the first time that HMGB1 levels were increased in synovial fluid as compared to plasma during JIA. HMGB1 levels in synovial fluid did not correlate to disease duration. In contrast, the recorded levels of IL-8 and S100 proteins were higher in synovial fluid during early phases of disease. This indicates a change in the inflammatory phenotype during the progression of JIA. High HMGB1 levels in synovial fluid correlated with early JIA onset, suggesting differences in immunopathogenesis between patient groups.

I have also demonstrated that HMGB1 may form complexes with the exogenous TLR ligand LPS or the endogenous inflammatory mediators IL-1 α and IL-1 β , respectively. Compared to each mediator alone such complexes stimulated synovial fibroblasts from arthritis patients to enhanced production of cytokines and tissue degrading enzymes. This enhancement is mediated via the reciprocal receptor for each HMGB1-partner molecule. Since all the studied mediators are present in arthritic joint during inflammation, this is a potential mechanism through which HMGB1 enhances ongoing inflammation and destruction during rheumatic diseases.

Finally, I have demonstrated that the proinflammatory activity of HMGB1 can be therapeutically targeted, either by inhibiting its active release by clinically approved anti-rheumatic drugs or by neutralization with a HMGB1-specific monoclonal antibody. Extracellular secretion of HMGB1 from LPS+IFN- γ stimulated human primary monocytes was inhibited by dexamethasone, chloroquine and gold sodium thiomalate *in vitro* as recorded using an ELISPOT assay. Therapeutic administration of an HMGB1-specific HMGB1 monoclonal antibody ameliorated arthritis in two separate experimental models.

In conclusion, my thesis work has added to the growing evidence that HMGB1 is involved in the pathogenesis of arthritis, has revealed a potential mechanism for its proinflammatory function and has demonstrated a means by which HMGB1-mediated activities can be counteracted.