AB0078 SYSTEMIC AND LOCAL IL-17A AND MIR-223 LEVELS IN RHEUMATOID ARTHRITIS PATIENTS

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Background: IL-17 is a proinflammatory cytokine, which overproduction promotes the autoimmune reaction in rheumatoid arthritis (RA). Recent studies have shown that IL-17 production in lymphocytes or its function could be regulated by miR-223 by targeting Roquin ubiquitin ligase or its receptors.1–4

Objectives: To examine a possible correlation between the expression levels of miR-223 and IL-17A in peripheral blood in patients with active RA. There was no significant difference between the effects of alfacalcidol and calcitriol, except decreased TGF-β production (p=0.535) after calcitriol supplementation. Methylprednisolone supplementation leads to a significant reduction of the IL-6 (p=0.002), IL-17 (p=0.001), TNF-α (p=0.002) and IL-6 (p=0.04), and induce more intense anti-inflammatory cytokine production IL-4 (p=0.0001), IFN-γ (p=0.05), TGF-β (p<0.0001) and IL-10 (p=0.09), in PBMC cultures of patients with active RA. There was no significant difference between the effects of alfacalcidol and calcitriol, except decreased TGF-β production (p=0.535) after calcitriol supplementation. Methylprednisolone supplementation leads to a significant reduction of the IL-6 (p=0.002), IL-17 (p=0.001), TNF-α (p=0.002) and IL-6 (p=0.017) production, while increased production of IL-21, IL-10, IFN-γ didn’t reach statistical significance. Cotreatment with alfacalcidol and methylprednisolone leads to additional, more significant reduction of IL-17 (p=0.0001) and TNF-α (p=0.0001) in PBMCs of patients with active disease.

Conclusions: Alfacalcidol, in vitro, showed a significant immunomodulatory effect through the specific inhibition of Th1–cytokine production, while Th2 cell response was enhanced – “Th2 switch”. Our results demonstrate that alfacalcidol has significant additive effects on glucocorticoid-mediated inhibition of Th1 cytokine production when combined with methylprednisolone. These findings demonstrate the potential use of alfacalcidol as an immunosuppressive agent when combined with corticosteroids in Th1, but not Th2, immune response.

REFERENCES:

Disclosure of Interest: None declared

AB0080 DIFFERENTIAL EFFECTS OF TR14 VERSUS DICLOFENAC ON PRO-RESOLVING LIPID MEDIATORS REVEALED BY RNASEQ

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Background: Anti-inflammatory agents are used widely in treating numerous pain and inflammatory conditions. With a focus on the specialised pro-resolving mediators (SPM) pathways in cutaneous wound repair in mice, the therapeutic activities of TR14 (Traumeel®), a multicomponent/multitarget natural product, and diclofenac (NSAID), a non-selective cyclooxygenase (COX) inhibitor were compared. COX inhibitors can block the synthesis of prostaglandins and thromboxanes from arachidonic acid, and can have important effects on the synthesis of resolvin and protectins produced by similar enzymes from eicosapentanoic and docosahexanoic acid substrates. Differential effects were identified via transcriptome analysis (RNaseq).

Objectives: To compare the transcriptomic changes after administration of TR14 or diclofenac in a mouse cutaneous wound healing model, with particular emphasis on the SPM pathways, which include resolvin and protectins.

Methods: After abrasive wounding, the wounds were treated with topical TR14 (34 mg/ml) in combination with subcutaneous TR14 injections (9.5 mg/ml), or with subcutaneous TR14 injections only, or topical diclofenac at clinically relevant doses (2 mg/ml). Skin samples were analysed for RNA transcript profiling by RNaseq at specific times (12 hour, 24 hour, 36 hour, 72 hour, 96 hour, 120 hour, 192 hour) after injury. Differentially expressed genes (DEGs) were computed at each time point between diclofenac vs control or TR14 vs control, using EdgeR.

Results: At early time points (12–36 hour), TR14-treated wounds, and to a lesser extent diclofenac-treated wounds, showed marked induction of 3-lipoxygenase expression with concomitant decrease in the expression of 5-lipoxygenase.
Lipocalin-2 (LCN2) has become increasingly relevant as a potential clinical biomarker of rheumatic diseases. The biological role of LCN2 in the adaptive immunity is less understood. It has been shown that LCN2 can induce immune tolerance by upregulation of human leukocyte antigen G (HLA-G) expression and by expansion of T-regulatory cells. LCN2-deficient mice have been found to be more susceptible to induction of autoimmunity. Systemic lupus erythematosus (SLE) is a disease associated with loss of tolerance. Pregnancy complications seen in SLE are also regarded as a consequence of immune dysregulation. LCN2 might therefore play a role as an immune modulator in SLE pregnancies.

Objectives: The study objective was to obtain a better understanding of immune regulation in pregnant women with SLE. In this study, we analysed serum LCN2 and clinical parameters in women with RA, SLE and healthy controls during pregnancy and postpartum.

Methods: The Norwegian National Advisory Unit on Pregnancy and Rheumatic Diseases collects serum samples in a biobank from women with inflammatory rheumatic diseases. Samples were obtained before pregnancy, in each trimester and 6 weeks, 6 months and 12 months postpartum from pregnant women with SLE (n=28), RA (n=34) and healthy pregnant controls (n=19). A sandwich ELISA was used to measure LCN2 in the serum samples. The biobank database was linked to RevNatus, a Norwegian quality registry collecting comprehensive clinical data about these women.

Results: Our cohort of pregnant women with SLE and RA had low disease activity throughout pregnancy and 67%–95% used medication (table 1). LCN2 levels in serum samples from women with SLE were found significantly lower compared to women with RA and healthy controls at all time-points (p<0.05) (graph).

<table>
<thead>
<tr>
<th>Disease activity</th>
<th>SLE patients</th>
<th>RA patients</th>
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<tbody>
<tr>
<td></td>
<td>1st trimester</td>
<td>2nd trimester</td>
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<tr>
<td>LAI-P Mean (range)</td>
<td>0.04 (0–0.25)</td>
<td>0.02 (0–0.16)</td>
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<tr>
<td>DASS28 Mean (range)</td>
<td>2.34 (1.79–7.91)</td>
<td>2.57 (1.42–1.79)</td>
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Conclusions: Pregnant women with SLE had lower levels of LCN2 compared to pregnant women with RA and healthy controls. Our cohort of women had well-controlled disease, making it likely that our findings represent inherent biological differences rather than effects of disease activity. Low LCN2 levels can be a possible mechanism for loss of tolerance seen in SLE patients during pregnancy.