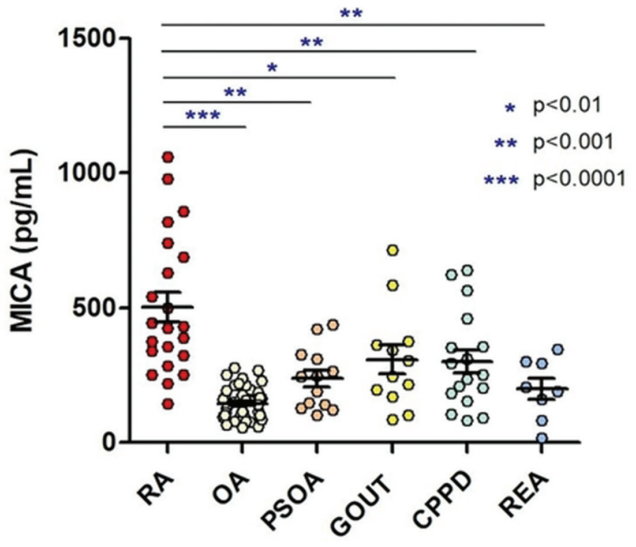
Pyrophosphate Deposition Disease (CPPD), 8 Reactive Arthritis (REA) and 45 Osteoarthritis patients. Gout and CPPD diseases were confirmed by the presence of crystals in SF. Clinical data were collected. The concentration of soluble MIC-A (sMIC-A), interleukin (IL)−1, IL-6 and IL-8 was measured by ELISA. Murine Rae-1, H60 and Mult1 transcripts were quantified by real-time quantitative PCR (RT-qPCR) in 3 models of joint inflammation: Serum Transfer Arthritis (STA), Collagen-induced arthritis (CIA) and Collagenase-Induced Osteoarthritis (CIOA).

Results: Significantly higher levels of sMIC-A were found in the synovial fluid of RA patients in comparison with all others diseases (p<0.001, figure n.1). sMIC-A levels were correlated to white blood cell counts and levels of inflammatory cytokines IL-1, IL-6 and IL-8. Similarly, higher expression levels of Rae-1, H60 and Mult1 were found in chronic arthritis mouse models in comparison with osteoarthritic mice.



Abstract AB0069 – Figure 1

Conclusions: Our data identifies synovial sMIC-A as an important player in rheumatoid arthritis compared to other rheumatic diseases and osteoarthritis. Investigations in mouse models are in agreement with this finding

REFERENCES:

[1] Bahram, Seiamak, et al. A second lineage of mammalian major histocompatibility complex class I genes. Proceedings of the National Academy of Sciences 1994;91(14):6259–6263.

[2] Groh, Veronika, et al. Costimulation of CD8αβ T cells by NKG2D via engagement by MIC induced on virus-infected cells. Nature immunology 2001;2(3):255.

[3] Spear, Paul, et al. NKG2D ligands as therapeutic targets. Cancer Immunity Archive 2013;13(2):8.

[4] Carapito, Raphael, Seiamak, Bahram. Genetics, genomics, and evolutionary biology of NKG2D ligands. Immunological reviews 2015;267(1):88–116.

Disclosure of Interest: None declared
DOI: 10.1136/annrheumdis-2018-eular.5169

AB0070 ADIPOKINES AND CYTOKINES IN THE PATHOGENESIS OF PSORIASIS AND METABOLIC DISORDERS

M. Khobeysh[1], K. Sysoev[2], E. Sokolovskiy[1], S. Lapin[3]. *[1]Department of Dermatology and Veneral Diseases, Academician I.P. Pavlov First St. Petersburg State Medical University; [2]Pavlov Institute of Physiology, RAS; [3]Academician I.P. Pavlov First St. Petersburg State Medical University, St. Petersburg, Russian Federation*

Background: Psoriasis is a systemic immune-associated disease with a specific comorbidity. The manifestations of the metabolic syndrome in this category of patients are changes that develop on systemic immune-associated inflammatory psoriatic process background and contribute to the progression of chronic inflammation.

Objectives: The aim of the study was determination of the main adipokines and cytokines content in the serum of peripheral blood, severity and activity of the disease in patients with psoriasis; clinical and laboratory evaluation of metabolic disorders. The interconnection between the production of adipokines and cytokines in psoriasis was analysed depending on the severity and activity of the psoriatic process and the nature of metabolic disorders.

Methods: Serum levels of adipokines (C-peptide, ghrelin, insulin, glucagon, leptin, visfatin, resistin, GIP, GLP-1 and PAI-1) and cytokines (IL-1β, IL-1ra, IL-2, IL-4, IL-5, IL-6, IL-7, IL-8, IL-9, IL-10, IL- 12 (p70), IL-13, IL-15, IL-17, eotaxin, FGF-2, G-CSF, GM-CSF, IFN-γ, IP-10, MCP-1, MIP-1α, MIP-1β, PDGF-BB, RANTES, TNF-α and VEGF) were measured in 36 patients with moderate and severe psoriasis, psoriatic arthritis. There was control group of 15 basically healthy persons. Clinical and laboratory evaluation of metabolic disorders (BMI, dyslipidemia, carbohydrate metabolism disorders) and cardiovascular diseases was performed for all examined patients. The duration, severity and the amount of body surface area involved in psoriasis were evaluated in all patients using recommended indices (BSA, PASI).

Results: Patients with psoriasis showed an increase in the production of glucagon, leptin, visfatin, GLP-1 (p<1,0E-03) and a decrease in the level of C-peptide, insulin, GIP, PAI-1, resistin (p<1,0E-06) compared with the control group. The difference of ghrelin concentrations in both group was not statistically significant. Patients with psoriasis showed an increase in the level of IL-1β, IL-1ra, IL-2, IL-4, IL-5, IL-6, IL-8, IL-9, IL-10, IL-12 (p70), IL-13, IL-15, IL-17, eotaxin, FGF-2, GM-CSF, IFN-γ, IP-10, MCP-1, MIP-1β and TNF-α compared to control group (p<0, 01). Positive correlations between the level of adipokines and cytokines were revealed.

Conclusions: The obtained data allow us to define adipokines as mediators between immune and endocrine systems. The imbalance between the proinflammatory and anti-inflammatory effects of adipokines observed in psoriasis demonstrates lipid metabolism dysfunction as one of the possible provoking factors of chronic inflammation determining the severity of the underlying disease. According to the results of the study, the following biological adipokines and cytokines should be classified as early biological markers of severity of the psoriatic immune-associated inflammatory process, with all its comorbid risks: GLP-1, glucagon, leptin, wisfatin, IL-1ra, IL-2, IL-4 IL-5, IL-6, IL-7, IL-8, IL-12, IL-13, IL-17, eotaxin, FGF-2, GM-CSF, IFN-γ, IP-10, MCP-1, MIP-1β, TNF-α. The levels of adipokines and cytokines are probably the earliest biological markers in patients with metabolic syndrome and psoriasis, the control of adipokines and cytokines level can be used to optimise therapy.

Disclosure of Interest: None declared
DOI: 10.1136/annrheumdis-2018-eular.5684

AB0071 EFFECTS OF CHONDROITIN SULPHATE AND GLUCOSAMINE ON INFLAMMATORY CYTOKINES IN MACROPHAGES

M.-F. Hsueh[1], E. Montell[2], V.B. Kraus[1]. *[1]Molecular Physiology, Duke University, Durham, USA; [2]Pre-Clinical RandD Area, Bioibérica, S.A.U., Barcelona, Spain*

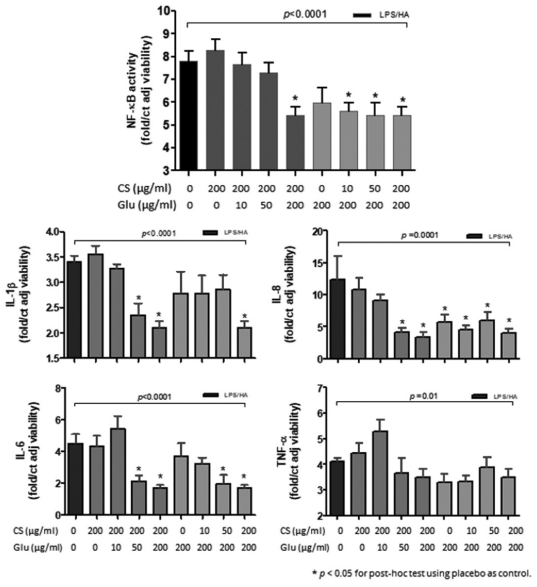
Background: The combination of pathogen-associated molecular patterns (PAMPs), such as lipopolysaccharide (LPS), and endogenous danger-associated molecular patterns (DAMPs), such as hyaluronan (HA) fragments, are known to be able to induce an inflammatory response from macrophages characterised by the release of pro-inflammatory cytokines[1]. We have previously shown that pharmaceutical grade chondroitin sulphate (CS), commonly used in the symptomatic treatment of osteoarthritis (OA), can attenuate the inflammatory response.

Objectives: To evaluate the synergistic effects of CS and glucosamine (GLU) in combination on LPS/HA mediated inflammatory responses of an immortalised human macrophage cell line (THP-1) and primary synovial fluid cells.

Methods: THP-1 monocyte cells were grown and differentiated into mature macrophages by the addition of 200 nM of phorbol 12-myristate 13-acetate (PMA) as previously described[1]. Cells were cultured with a physiologically achievable range of concentrations of CS and GLU (0, 10, 50, 200 μg/ml of each, Bioibérica, S.A. U.) for 6 hours, then primed with physiologically relevant concentrations of LPS (10 ng/ml) (n=12/group). After 24 hours, cell culture media were replaced with serum free Opti-MEM supplemented with the previously mentioned concentrations of CS and GLU, LPS, and 10 μg/ml HA fragments (ultra-low molecular weight, Lifecore). After a further 24 hours, supernatants were harvested for NF-κB activity and pro-inflammatory cytokine (IL-1β, IL-6, IFN-γ, and TNF-α) assessment. Cell viability was measured using PrestoBlue reagent. The human knee

primary synovial fluid cells were collected at the time of joint replacement and cultured with CS (200 μg/ml) and GLU (200 μg/ml), singly or in combination, with the addition of LPS and HA (n=2/group). After normalisation for cell viability, all results were expressed as fold change from the negative control (media only). One-way ANOVA with Dunnett's post-hoc test was performed using GraphPad Prism.

Results: CS and GLU in combination (200 μg/ml of each) significantly reduced NF-κB activity by 70% compared to the positive control group (LPS/HA only). Although CS (200 μg/ml) alone did not reduce NF-κB activity, the addition of the lower concentration of CS (10–50 μg/ml) to GLU (200 μg/ml) significantly reduced NF-κB activity compared with GLU (200 μg/ml) alone. Addition of lower concentrations of GLU (10–50 μg/ml) to CS (200 μg/ml) modestly reduced NF-κB activity (Fig 1). Similar trends were observed in secreted pro-inflammatory cytokines (IL-1β, IL-6, IFN-γ, and TNF-α); namely, CS and GLU in combination significantly attenuated the LPS/HA mediated pro-inflammatory responses ($p<0.05$) (Fig 1). Although, a diverse range of inflammatory responses to the LPS/HA activation was observed, constitutive pro-inflammatory cytokine production by primary synovial fluid cells was reduced by the combination of CS and GLU.



Abstract AB0071 – Figure 1

*p<0.05 for post-hoc test using placebo as control.

Conclusions: Inflammatory reactions of THP-1 macrophages, induced by physiologically relevant concentrations of LPS and HA fragments, were suppressed synergistically by the combination of physiologically achievable concentrations of CS and GLU. A similar trend was observed in primary human synovial cells but further investigations are required. These data could explain, at least in part, the clinical efficacy of CS and GLU in combination observed in OA patients.

REFERENCE:

[1] Stabler TV. Osteoarthritis Cartilage 2017;25(1):166–174.

Disclosure of Interest: M.-F. Hsueh Grant/research support from: Bioibérica, S. A.U., E. Montell Employee of: Bioibérica, S.A.U., V. Kraus Grant/research support from: Bioibérica, S.A.U.

DOI: 10.1136/annrheumdis-2018-eular.2954

AB0072 EVALUATION OF ANTI-INFLAMMATORY EFFECTS OF NAPROXEN SODIUM ON HUMAN OSTEOARTHRITIS SYNOVIAL CELLS

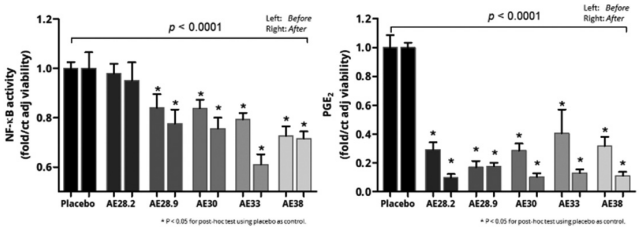
M.-F. Hsueh, V.B. Kraus. *Molecular Physiology, Duke University, Durham, USA*

Background: Inflammation is increasingly recognised as an essential factor in the pathogenesis and progression of osteoarthritis (OA). Etarfolatide imaging for activated macrophage quantification in knee joints confirms a high prevalence (~70%–80%) of joint inflammation in association with OA[1]. Naproxen sodium is a non-steroidal anti-inflammatory drug that is widely available over-the-counter (OTC) and has been shown to be effective in different pain models including OA[2, 3]. Naproxen pre-treatment reduces inflammatory responses of chondrocytes. However, the effects of naproxen sodium on human OA synovial cells are not well known.

Objectives: Evaluate the ability of naproxen sodium to block the inflammatory responses and to reduce the activated inflammatory responses of a human monocyte cell line and primary human synovial fluid (SF) cells *in vitro*.

Methods: The immortalised human monocytic cell line, THP-1, was grown and differentiated into mature macrophages using phorbol 12-myristate 13-acetate as described previously[4]. Mature macrophages were treated with various concentrations of naproxen sodium two hours before or 24 hours after inducing an inflammatory reaction using lipopolysaccharide (LPS) and hyaluronan (HA) fragments (n=8/group). After a further 24 hours, the cell culture supernatants were assessed for NF-κB activity, pro-inflammatory cytokines (IL-1β, IL-6, IFN-γ, and TNF-α), and prostaglandin E_2 (PGE_2). Cell viability was assessed using PrestoBlue reagent. Primary human SF cells were collected at the time of knee joint replacement for OA and treated 24 hours with naproxen sodium with and without the addition of LPS/HA (n=2/group). All results were expressed as fold change from the negative control (media only) after normalisation for cell viability. One-way ANOVA with Dunnett's post-hoc tests were done using GraphPad Prism.

Results: Compared to the placebo group, NF-κB activity of THP-1 cells was significantly reduced by as little as 28.9 mg/L naproxen sodium (corresponding to a trough plasma concentration achieved by a daily oral dose of 55 mg naproxen sodium) when added before or after the activation by LPS/HA (84% and 78% NF-κB activity reduction, respectively) (Fig1). When cells were treated before the activation with 33 mg/L naproxen sodium (corresponding to the concentration achieved by a 220 mg daily OTC dose), NF-κB activity was reduced 79% and IL-6 secretion was reduced by 77%. Cyclooxygenase enzyme activity, represented by PGE_2 production, was reduced to basal levels by as little as 28.2 mg/L naproxen sodium ($p<0.05$) when cells were treated either before or after the activation. Primary human SF cells treated with LPS/HA showed a striking increase in cytokine secretion ranging from 50-fold (IL-8) to 600-fold (IL-6). Cytokine production was reduced by naproxen sodium but a rebound phenomenon was observed for the highest concentration of 55/mg/L, which may indicate the cell stress response.



Abstract AB0072 – Figure 1

*p<0.05 for post-hoc test using placebo as control.
*p<0.05 for post-hoc test using placebo as control.

Conclusions: Naproxen sodium at low dose can both prevent and reduce inflammatory responses of a human monocytic cell line and primary human SF cells *in vitro*. These results highlight the potent activity of the OTC dose of naproxen sodium to dramatically reduce PGE_2, NF-κB activity and cytokine production.

REFERENCES:

[1] Kraus VB. Osteoarthritis Cartilage 2016;24(9):1613–21.
[2] Golden HE. Am J Ther 2004;11(2):85–94.
[3] Schiff M. J Rheumatol 2004;31(7):1373–83.
[4] Stabler TV. Osteoarthritis Cartilage 2017;25(1):166–174.

Disclosure of Interest: M.-F. Hsueh Grant/research support from: Bayer HealthCare, V. Kraus Grant/research support from: Bayer HealthCare

DOI: 10.1136/annrheumdis-2018-eular.3337