Results: In SLE patients respect to HC, plasma IL-1 β levels were unmodified whereas IL-6 was higher, resulting significantly increased in SLE-S. Monocytes isolated from SLE patients released lower quantities of IL-1 β after stimulation with BzATP, whereas the release of both IL-6 and TNF- α was significantly augmented in SLE-NS respect to both HC and SLE-S subjects after all types of stimulation. RT-PCR showed reduced P2X7R and augmented NLRP3 mRNA expression in SLE patients. Accordingly, P2X7R activity was significantly reduced in all SLE patients and did not appear to be influenced by a chloroquine pre-treatment.

Conclusion: In SLE patients, compared to HC subjects, we found reduced P2X7R mRNA expression, increased NLRP3 mRNA, as a possible compensating mechanism, and correspondingly, significantly lower BzATP-induced intracellular Ca²⁺ increase, without an apparent influence by chloroquine, one of the drugs most diffusely used for SLE treatment. The *in vitro* IL-1 β release was reduced, whereas plasma IL-1 β was unaltered, indicating an alternative source, other than monocytes, of this cytokine. Conversely, IL-6 and TNF- α levels were increased *in vitro*, and IL-6 was present in plasma at higher levels. The possible consequences of reduced P2X7R, mainly on cytokines network deregulation and lymphocyte proliferation, will be further investigated as well as the role of IL-6 and TNF- α as possible therapeutic targets.

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THU0017 IN VITRO MECHANISTIC STUDIES DEMONSTRATE FILGOTINIB ACTIVITY THAT HAS POTENTIAL IMPLICATIONS FOR DIFFERENTIATION AMONG JAK INHIBITORS

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Background: Inhibition of the Janus kinase-signal transducers and activators of transcription (JAK-STAT) pathway has demonstrated efficacy in immune-mediated diseases and has been identified as a therapeutic target for the treatment of rheumatoid arthritis (RA). Differences in JAK inhibitor specificity for JAK1, JAK2, JAK3, and TYK2 may influence their safety profiles, but the mechanism is not known. Selective JAK1 inhibition by filgotinib (FIL) may modulate a subset of proinflammatory cytokines associated with RA pathogenesis and improve the risk-benefit profile by minimizing other non-JAK1-related adverse events. JAK2 inhibition is associated with cytopenias, while JAK3 inhibition has been associated with increased risk for opportunistic infections (eg, tuberculosis and herpes zoster) and chronic low-grade inflammation. In clinical trials, FIL did not negatively impact hemoglobin, LDL/HDL ratios, or natural killer (NK) cell counts.¹⁻³

Objectives: To compare the *in vitro* profile of JAK inhibitors with different JAK selectivity profiles, for effects on erythroid progenitor cell expansion, NK cell proliferation, and liver X receptor (LXR) agonist-induced cholesterol ester transfer protein (CETP) expression, an enzyme responsible for the conversion of HDL to LDL.

Methods: JAK inhibitors (FIL, FIL metabolite [GS-829845], baricitinib [BARI], tofacitinib [TOFA], and upadacitinib [UPA]) were evaluated *in vitro* in human cell-based assays: growth of erythroid progenitors from human cord blood CD34⁺ cells using a HemaTox™ liquid expansion assay, IL-15-induced NK cell proliferation, and LXR agonist-induced CETP expression in the hepatic cell line (HepG2). Using IC₅₀s generated from these

assays and the reported human plasma concentrations of the JAK inhibitors from clinical studies,⁴⁻⁶ we calculated the target coverage for each compound at clinically relevant doses. The activity of FIL in humans was based on a PK-PD modeling algorithm⁷ of FIL + GS-829845.

Results: *In vitro* assay results are described in the table. Based on these results, human exposure data, and modeled PK-PD relationships, FIL 100 mg and FIL 200 mg result in lower calculated cellular inhibition than the other JAK inhibitors at clinical exposures. Notably, FIL 100 mg and FIL 200 mg, but not the other inhibitors, are calculated to reduce CETP expression by 17% and 27%, respectively, while BARI, TOFA, and UPA are not expected to alter CETP levels.

Abstract THU0017 Table 1. IC₅₀ ± SD in *in vitro* assays (nM, unless otherwise noted).

Assay	FIL	GS-829845	BARI	TOFA	UPA
Early erythroid progenitors	1960 ±137	19300 ±1730	38.6±2.9	210 ±15.2	42 ±2.9
Mature erythroid progenitors	1140 ±112	10600 ±1270	25.7±2.9	110 ±10	24.5 ±2.75
NK cell proliferation	314.8 ±53	9697 ±8100	6.6±1.9	12.2 ±2.1	4.1 ±1.7
Inhibition of LXR agonist-induced CETP expression	15.3 ±7.1 μ M	19.4 ±4.2 μ M	>1 μ M weak induction ^a	No effect	No effect

^a Weak stimulation of LXR agonist-induced CETP expression.

Conclusion: JAK1 selectivity of FIL and GS-829845 resulted in less inhibition of erythroid progenitor expansion and NK cell proliferation compared with BARI, TOFA, and UPA. FIL also reduced LXR agonist-induced CETP expression, while the other inhibitors did not alter these levels. These results provide a potential mechanistic link to the observed reduction of CETP concentration and activity following FIL treatment, and the observed reduction in LDL:HDL in RA patients.⁸

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THU0018 ANTI-GALECTIN-9 ANTIBODY AS A NOVEL TREATMENT OPTION IN RHEUMATOID ARTHRITIS TARGETING PATHOGENIC FIBROBLAST-LIKE SYNOVIOCYTES

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Background: Fibroblasts-like synovocytes (FLS) present in the stromal environment are key effector cells in the persistence of synovial inflammation and joint damage in rheumatoid arthritis (RA). A particular subset of FLS characterized as Thy-1⁺Podoplanin⁺CD34⁺ is significantly expanded in RA patients and has been described to be disease-associated¹. Currently, no treatment targeting the stromal environment in RA is available².

Objectives: To identify a novel treatment option targeting the stromal environment in RA to modify the disease-associated subset of fibroblasts.

Methods: An *in vitro* model was established with FLS derived from synovial fluid mononuclear cells (SFMC) from RA and osteoarthritis (OA) patients (n=6). FLS between passage 2-5 were used for further analyses. Untreated FLS in cultures were analyzed by flow cytometry for expression