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MEETING ABSTRACT

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STAT1-deficient mice develop a B-cell malignancy reminiscent of JAK1/2-inhibition-associated B-cell lymhomas in MPN patients

Andrea HOELBL-KOVACIC[1,#], Edit PORPACZY[2,#], Sabrina TRIPOLT[1,#], Bettina GISSLINGER[2], Zsuzsanna BAGÓ-HORVÁTH[1,3], Emilio CASANOVA-HEVIA[4,5], Thomas DECKER[6], Sabine FAJMANN[1], Daniela A. FUX[1], Sinan GÜLTEKIN[2], Gerwin HELLER[7], Harald HERKNER[8], Thomas KOLBE[9,10], Eva M. PUTZ[1], Christoph KORNAUTH[3], Mathias MÜLLER[11], Michaela PRCHAL-MURPHY[1], Ana-Iris SCHIEFER[3], Christine SCHNECKENLEITHNER[1], Cathrin SKRABS[2], Wolfgang R. SPERR[2,12], Birgit STROBL[11], Peter VALENT[2,12], Robert KRALOVICS[13,16], Leonhard MÜLLAUER[3], Ingrid SIMONITSCH-KLUPP[3], Emmanuelle CLAPPIER[14,15], Emmanuel RAFFOUX[14,15], Jean-Jacques KILADJIAN[14,15], Maria-Theresa KRAUTH[2], Philipp B. STABER[2], Georg GREINER[16], Gregor HÖRMANN[12,16,17], Ulrich JÄGER[2,12,§], Heinz GISSLINGER[2,§] and Veronika SEXL[1,§,*] (*[#]contributed equally; [§]shared last authorship)*

[1]Institute of Pharmacology and Toxicology, Department for Biomedical Sciences, University of Veterinary Medicine, Vienna, Austria; [2]Division of Hematology and Hemostaseology, Department of Internal Medicine I, Comprehensive Cancer Center, Medical University of Vienna, Austria; [3]Clinical Institute of Pathology, Medical University of Vienna, Austria; [4]Ludwig Boltzmann Institute for Cancer Research (LBI-CR), Vienna, Austria; [5]Institute of Physiology, Centre of Physiology and Pharmacology, Comprehensive Cancer Centre, Medical University of Vienna, Austria; [6]Max F. Perutz Laboratories (MFPL), University of Vienna, Austria; [7]Clinical Division of Oncology, Department of Medicine I, Comprehensive Cancer Center, Medical University of Vienna, Austria; [8]Department of Emergency Medicine, Medical University of Vienna, Austria; [9]Biomodels Austria, University of Veterinary Medicine, Vienna, Austria; [10]IFA Tulln, University of Natural Resources and Life Sciences, Tulln, Austria; [11]Institute of Animal Breeding and Genetics, Department of Biomedical Sciences, University of Veterinary Medicine, Vienna, Austria; [12]Ludwig Boltzmann Cluster Oncology, Medical University of Vienna, Austria; [13]CeMM Research Center for Molecular Medicine, Austrian Academy of Sciences, Vienna, Austria; [14]Hôpital Saint-Louis, AP-HP, Centre d'Investigations Cliniques, Paris, France; [15]Université Paris-VII Denis Diderot, Paris, France; [16]Department of Laboratory Medicine, Medical University of Vienna, Austria; [17]MLL Munich Leukemia Laboratory, Munich, Germany

Background: The highly conserved JAK/STAT signaling pathway regulates proliferation, differentiation, apoptosis and immune responses. Several malignancies are associated with constitutive activation of STAT family members. Activating mutations in STAT3 drive the development of diffuse large B-cell lymphomas. The STAT3 counter-player STAT1 is generally considered a tumour suppressor. We observed that the loss of STAT1 provokes spontaneous haematopoietic tumours in mice.

Methods: Spontaneous hematopoietic tumours were analysed by FACS. The co-existence of a myeloid hyperplasia (MH) and a malignant B-cell disease was assessed via transplantations, myeloid cell depletion and all-*trans* retinoic acid (ATRA) treatment *in vivo*. High-purity sorting of individual haematopoietic cell lineages followed by transplantations were performed to identify the leukemia-initiating cell. Clonality of B cells was assessed by Southern blotting and PCRs for DJ rearrangements. Expression of STAT1-dependent target genes as well as general hallmark genes for B-cell lymphomas were analysed by qPCR. Two independent human patient cohorts were monitored for co-occurrence of myeloproliferative neoplasms (MPN) and B-cell lymphoma. Transcriptional profiles of human and murine patients were compared via RNA sequencing.

Results: STAT1-deficient mice develop an MH, which initially masks a malignant B-cell disease. Upon transplantation, malignant B cells arise and cause a fatal disease. The malignant B cells can be maintained *in vitro*. Transcriptional profiling reveals an up-regulation of *c-Myc*, *Bcl-2*, *SpiB*, *Mef2B*, *Card11* and *Cd274* (*PD-L1*) and down-regulation of *Socs-1*, *Cdkn2a*, *B2m* and *Prdm1*—alterations found in aggressive human B-cell lymphoma. The malignant B cells are already present in the *Stat1*$^{-/-}$ mouse during MPN. Elimination of the myeloid pool via ATRA freed these malignant B cells and allowed them to expand. We observed a similar switch from MPN to aggressive B-cell lymphoma in a subset of human patients upon inhibition of Janus kinase 1/2 (JAK1/2). The inhibition of JAK1/2 eliminates myeloid cells, but appeared to cause a fatal aggressive B-cell lymphoma later on. To identify the global frequency of this adverse effect, 626 MPN patients (557 with conventional, 59 with JAK1/2 inhibitor treatment) from Vienna and 929 (872 *vs.* 57) from Paris were monitored. In the cohort of 626 patients, B-cell lymphomas evolved in 5.8% upon JAK1/2 inhibition compared to 0.36% with conventional treatment (16-fold increased risk). A similar increase was observed in the independent cohort of 929 MPN patients. Comparison of transcriptional profiles identified 213 genes with overlapping expression patterns in murine and human patients. As in MH^{+} *Stat1*$^{-/-}$ mice, a significant proportion of MPN patients who developed an aggressive B-cell lymphoma harboured clonal B cells, which already existed during MPN.

Discussion: We conclude that JAK/STAT1 pathway inhibition in MPN is associated with an elevated frequency of aggressive B-cell lymphomas. Detection of a pre-existing B-cell clone will identify individuals at risk.

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*Corresponding author e-mail: *veronika.sexl@vetmeduni.ac.at*