JAK2-STAT3 Signaling Implicated in the Regulation of Hepatic TPO Production

Written by Maria Vinall

The Ashwell-Morell receptor (AMR) on the hepatocyte binds and removes desialylated platelets as they circulate and age in the blood. After binding to the AMR, the desialylated platelets induce hepatic expression of thrombopoietin (TPO) mRNA, thereby regulating platelet production. Renata Grozovsky, PhD, Brigham and Women's Hospital, Boston, Massachusetts, USA, presented data from a study [Grozovsky R et al. *Nat Medicine.* 2015] showing the TPO expression by way of the AMR is further regulated by Janus kinase 2 (JAK2) and signal transducer and activator of transcription 3 (STAT3) protein signaling in vivo and in vitro.

In 1994, 4 studies identified ligands that significantly increased platelet levels [Bartley TD et al. *Cell.* 1994; de Sauvage FJ et al. *Nature.* 1994; Kuter DJ et al. *Proc Natl Acad Sci USA*. 1994; Lok S et al. *Nature.* 1994]. The substance was subsequently identified as TPO, a glycoprotein hormone produced by the liver and kidney that in humans is encoded by the TPO gene. TPO regulates platelet production, supporting the survival, proliferation, and differentiation of platelet precursors and bone marrow megakaryocytes. The complete understanding of this pathway remained elusive until Dr Grozovsky's research.

In the in vitro treatment, desialylated platelets were shown to stimulate TPO production in human hepatoma cells (HepG2). The data indicate that following platelet desialylation, platelet ingestion by the AMR, TPO mRNA and the TPO protein significantly increase within 6 hours ($P < .01$) [Grozovsky R et al. *Nat Medicine* 2015].

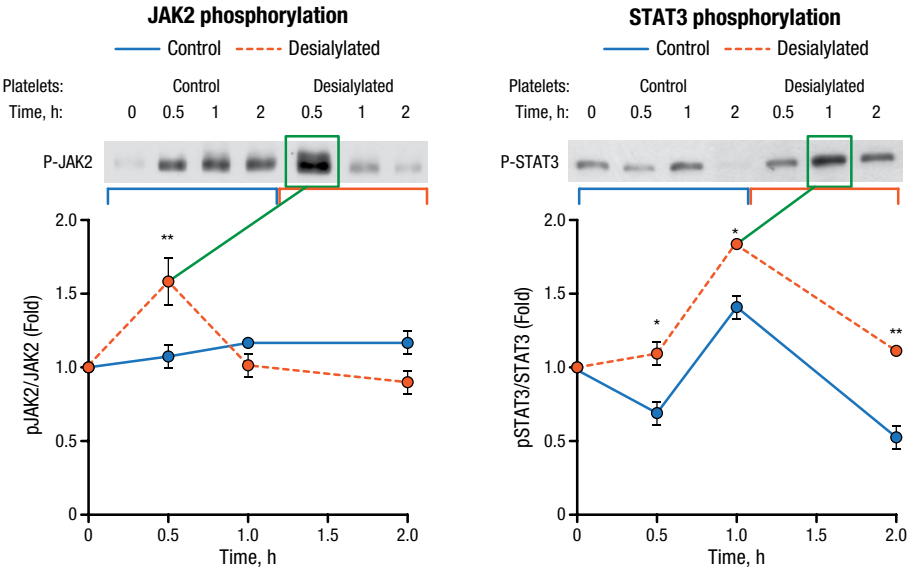
Desialylated human platelets were also shown to activate JAK2-STAT3 signaling in HepG2 cells (Figure 1). Phospho-STAT3 was translocated into the nucleus and TPO protein expression increased as well. Both of the latter were prevented by pharmacological JAK inhibition.

Peer-Reviewed Highlights From the

American Society of Hematology 56th Annual Meeting & Exposition

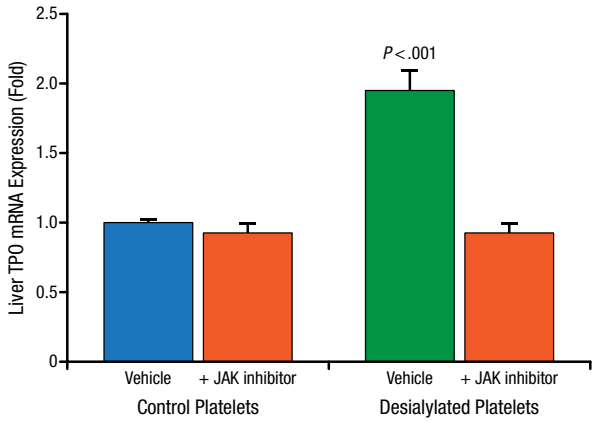
December 6–9, 2014
San Francisco, CA, USA

Figure 1. Desialylated Human Platelets Stimulate JAK2 and STAT3 Phosphorylation in HepG2 Cells



JACK2, Janus kinase 2; STAT3, activator of transcription 3.
*$P < .05$, **$P < .01$
Reproduced with permission from R Grozovsky, PhD.

Figure 2. JAK Inhibitors Block TPO mRNA Increase in Wild Type Mice



JAK, Janus kinase; TPO, thrombopoietin.

Reproduced with permission from R Grozovsky, PhD.

In the in vivo treatment, AMR-deficient mice had increased platelet count, survival, and loss of sialic acid compared with wild type mice showing that removal of desialylated platelets by the AMR regulated in vivo platelet survival.

In livers isolated from Asgr2-null mice, TPO mRNA expression decreased by 40%, while TPO mRNA expression in livers from mice lacking sialyltransferase (St3gal4-null) was increased by 30%, compared with wild type mice. In St3gal4-null mice, desialylated platelet clearance is increased and specifically mediated by the AMR. This indicates that desialylated platelet uptake by the AMR regulates liver TPO levels.

When desialylated platelets isolated from St3gal4-null or Asgr2-null mice are infused into wild type mice, hepatic TPO mRNA levels increase as early as 12 hours after infusion, while desialylated platelets infused into Asgr2-null mice have no effect on TPO mRNA synthesis. Further, treatment with JAK inhibitors block the desialylated platelet stimulated TPO mRNA increase in wild type mice (Figure 2).

The accumulated data show that circulating desialylated platelets by way of the AMR and JAK2-STAT3 signaling stimulated TPO production, providing a physiological feedback mechanism to regulate plasma TPO levels and platelet production in vivo and in vitro. The complete understanding of this feedback mechanism illuminates the pathophysiology of platelet diseases, such as thrombocythemia and immune thrombocytopenia. This feedback has clinical implications, however, as the administration of JAK1/2 inhibitors can cause thrombocytopenia in some groups of patients.

AALL0434 Treatment Therapy Has Excellent Outcomes in Children With T-ALL

Written by Maria Vinall

Children with T-cell acute lymphoblastic leukemia (T-ALL) have excellent outcomes after treatment with AALL0434, a standard 4-drug induction therapy. End-induction minimal residual disease (MRD) was more important in predicting treatment response than the early thymic precursor (ETP) immunophenotype, according to the results from the Combination Chemotherapy in Treating Young Patients With Newly Diagnosed T-Cell Acute Lymphoblastic Leukemia or T-Cell Lymphoblastic Lymphoma [NCT00408005] trial presented by Brent L. Wood, MD, PhD, University of Washington, Seattle, Washington, USA.

The phase 3 trial enrolled 1895 patients (aged 1 to 30 years) with T-ALL. The prognostic impact of MRD was measured by 8- and 9-color flow cytometry in the peripheral blood at day 8 and in bone marrow at end of induction (day 29), and end of consolidation was compared with prognostic ability of ETP status assessed at diagnosis. AALL0434 therapy consisted of prednisone for 28 days, vincristine weekly for 4 weeks, pegaspargase on day 4, and daunorubicin weekly for 4 weeks. An initial subset of cases confirmed that near ETP meets ETP criteria except for elevated CD5 B-cells.

An initial subset of thymocytes, ETP, retains stem-cell-like features, as reviewed by Coustan-Smith and colleagues [*Lancet Oncol.* 2009], who helped to define criteria for standardization in the AALL0434 study. The presence of ETP cells in patients with T-ALL confers a poor prognosis with use of standard intensive chemotherapy. Thus, early recognition is believed essential for the development of an effective clinical management strategy.

In the AALL0434 study, ETP, near ETP, and not ETP were noted in 11.3%, 17.0%, and 71.6% of patients, respectively. At day 29, MRD < 0.01% was significantly more evident in the group without ETP compared with those with ETP or near ETP ($P < .0001$). Induction failure, defined as > 35% blasts by morphology in bone marrow at day 29, was proportionally more evident in the group with ETP (45.5%) and near ETP (48.1%) compared with not ETP (11.2%). However, percentage outcomes for 4-year event-free survival (EFS) and overall survival (OS) were similar regardless of the subtype (Table 1).

MRD is an important predictor of relapse in children with T-ALL. An MRD day 29 risk < 0.1% was considered low risk, < 1.0% intermediate, and ≥ 1.0% high. An MRD D29 < 0.01% to < 10.0% was associated with a higher