SELECTED OPPORTUNITIES IN RARE BLOOD DISEASE

METHODS AND PHARMACEUTICAL COMPOSITIONS FOR THE TREATMENT OF THROMBOSIS IN PATIENTS SUFFERING FROM A MYELOPROLIFERATIVE NEOPLASM (BIO 17069)

BIO17069 - METHODS AND PHARMACEUTICAL COMPOSITIONS FOR THE TREATMENT OF THROMBOSIS IN PATIENTS SUFFERING FROM A MYELOPROLIFERATIVE NEOPLASM

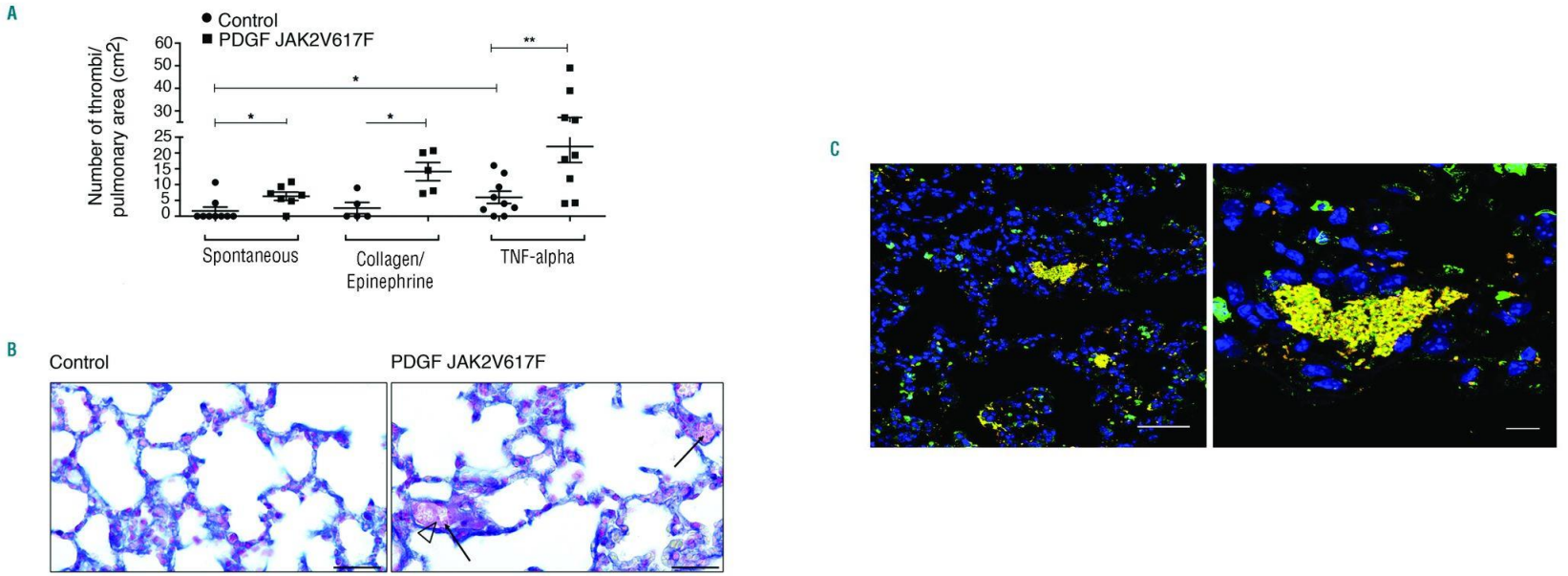
Product factsheet

In vivo PoC

- **Target:**
  - P-selectin.
- **Product:**
  - P-selectin antagonist.
- **Application:**
  - Thrombosis in in patients with JAK2V617F myeloproliferative neoplasms.
- **Technology:**
  - Antibody, small molecules.
- **Rational / POC:**
  - Thrombosis is the main cause of morbidity and mortality in patients with $JAK2^{V617F}$ positive myeloproliferative neoplasms (MPN);
  - Recent studies reported the presence of 10 $JAK2^{V617F}$ in endothelial cells in some MPN patients;
  - $JAK2^{V617F}$-expressing endothelial cells promote thrombosis through induction of endothelial P-selectin expression (*in vivo* model of mice with endothelial-specific JAK2V617F expression);
  - P-selectin inhibition (blocking antibody or hydroxyurea through direct reduction of endothelial P-selectin expression) is sufficient to reduce the increased of thrombosis in mice.
- **Patent and publication:**
  - PCT/EP2018/056333: METHODS AND PHARMACEUTICAL COMPOSITIONS FOR THE TREATMENT OF THROMBOSIS IN PATIENTS SUFFERING FROM A MYELOPROLIFERATIVE NEOPLASM.
  - Guy A, Gourdou-Latyszenok V, Le Lay N, Peghaire C, Kilani B, Dias JV, Duplaa C, Renault MA, Denis C, Villeval JL, Boulaftali Y, Jandrot-Perrus M, Couffinhal T, James C. *Vascular endothelial cell expression of JAK2V617F is sufficient to promote a pro-thrombotic state due to increased P-selectin expression*. Haematologica. 2019 Jan;104(1):70-81.

Proof of concept

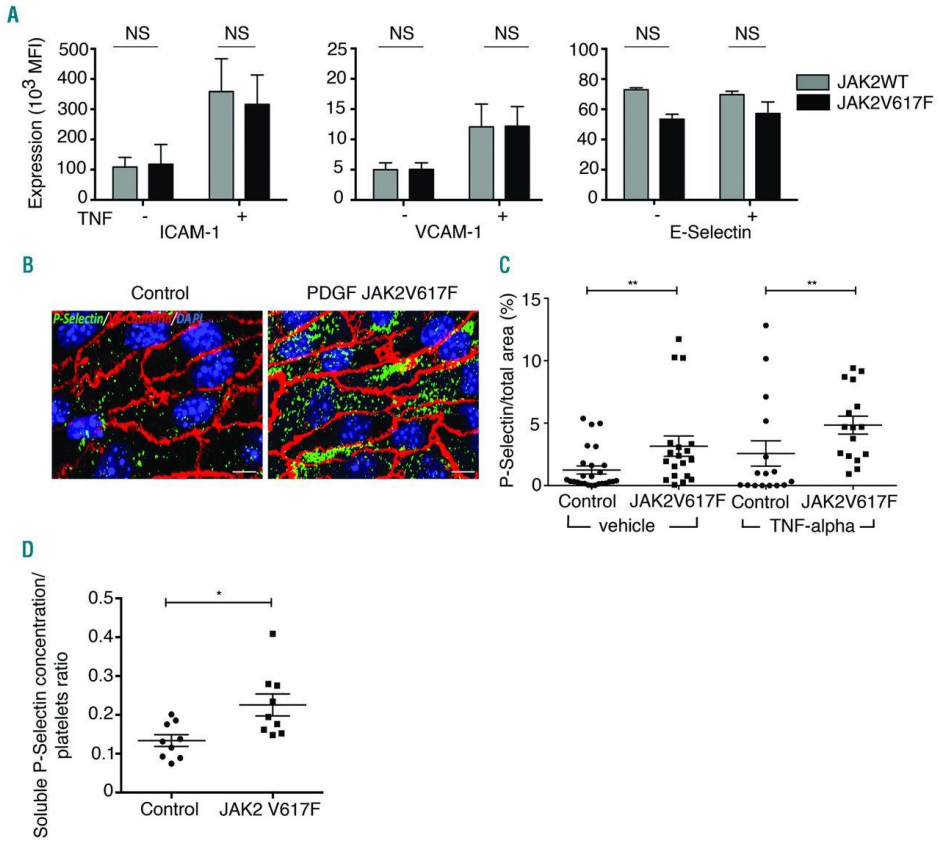
The presence of JAK2^{V617F} in endothelial cells promotes thrombus formation



(A) In *Pdgfb-iCreERT2;JAK2*$^{V617F/WT}$ mice, thrombus formation occurs spontaneously and is increased after weak platelet activation by low-dose collagen plus epinephrine, or injection of tumor necrosis factor (TNF)-alpha. (B) Carstairs staining of pulmonary thrombi in control mice (left) and *Pdgfb-iCreERT2;JAK2*$^{V617F/WT}$ mice (right) injected with TNF-alpha. Black arrows indicate thrombi. The clear arrow head indicates fibrin deposition. (C) Representative image of a thrombus formed by neutrophils (green) and platelets (yellow) in *Pdgfb-iCreERT2;JAK2*$^{V617F/WT}$ mice.

Proof of concept

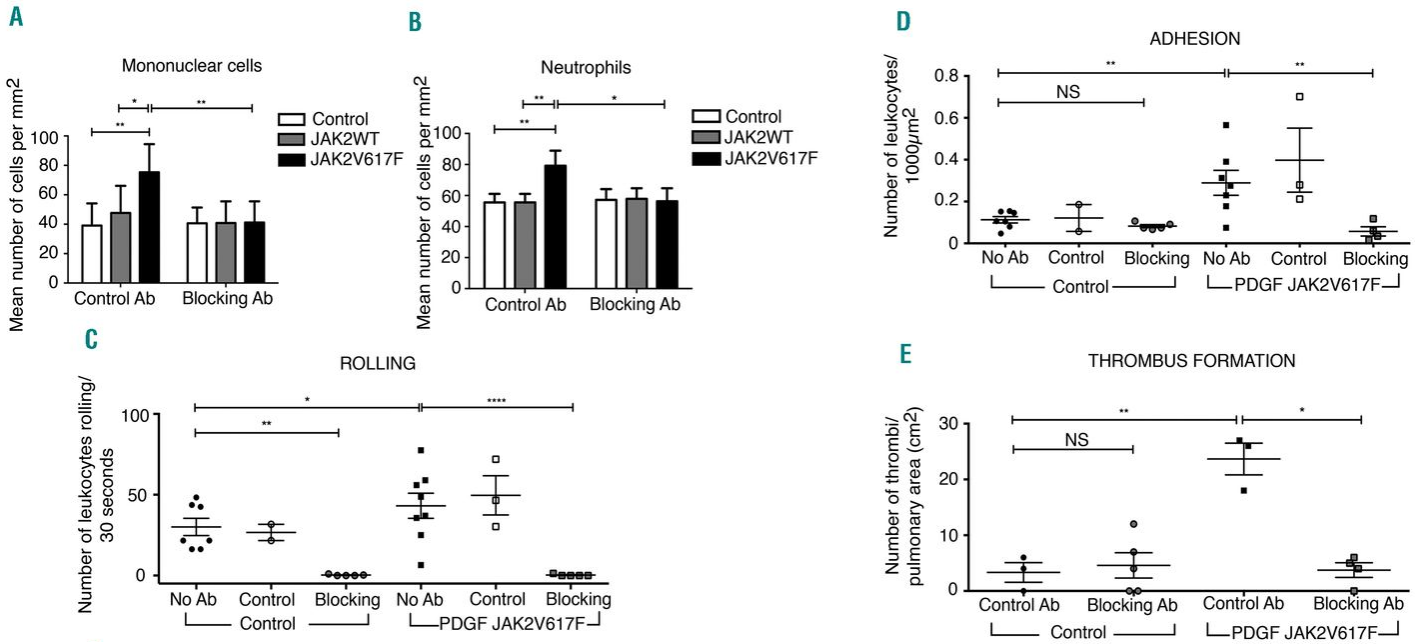
The presence of JAK2^{V617F} in endothelial cells promotes thrombus formation



(A) There was no modification of cell surface expression of the adhesion molecules, ICAM-1, VCAM-1, and E-selectin on JAK2^{V617F} human umbilical vein endothelial cells (HUVEC). (B) Representative images of P-selectin staining (green) in carotid endothelial cells. Nuclei are stained with DAPI (blue) and VE-cadherin (red). (C) Cell surface expression of mouse P-selectin is increased in carotid endothelial cells from *Pdgfb-iCreERT2;JAK2$^{V617F/WT}$* mice whether or not they received tumor necrosis factor (TNF)-alpha. Each dot represents one image (4 images per mouse). (D) The ratio between soluble P-selectin concentration and platelet count is significantly increased in *Pdgfb-iCreERT2;JAK2$^{V617F/WT}$* mice.

Proof of concept

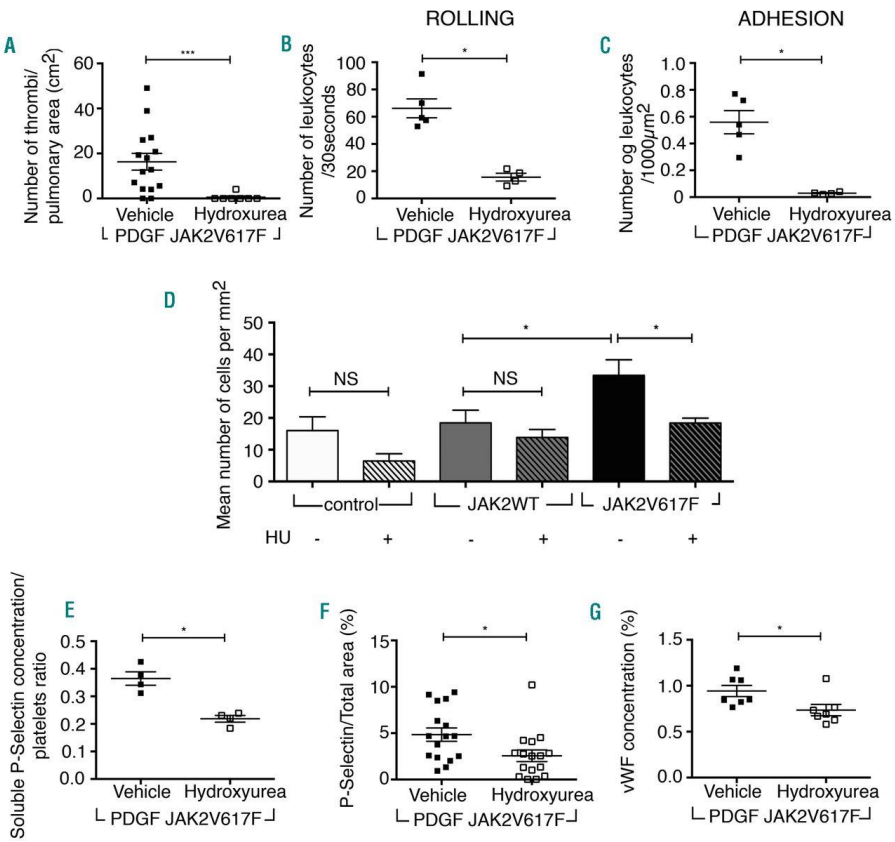
Increased endothelial P-selectin expression is responsible for the pro-adhesive phenotype of JAK2^{V617F}-expressing endothelial cells



(A,B) Under static conditions, increased adhesion of (A) normal mononuclear cells and (B) neutrophils on JAK2^{V617F} human umbilical vein endothelial cells is reversed in the presence of a P-selectin blocking antibody (Ab). In *Pdgfb-iCreERT2;JAK2$^{V617F/WT}$* mice, increased (C) rolling and (D) adhesion of leukocytes is abolished in the presence of a P-selectin blocking antibody. (E) Increased thrombus formation in *Pdgfb-iCreERT2; JAK2$^{V617F/WT}$* mice is abrogated in the presence of a P-selectin blocking antibody.

Proof of concept

Treatment with hydroxyurea decreases the pro-thrombotic and pro-adhesive phenotype of JAK^{2V617F}-expressing endothelial cells



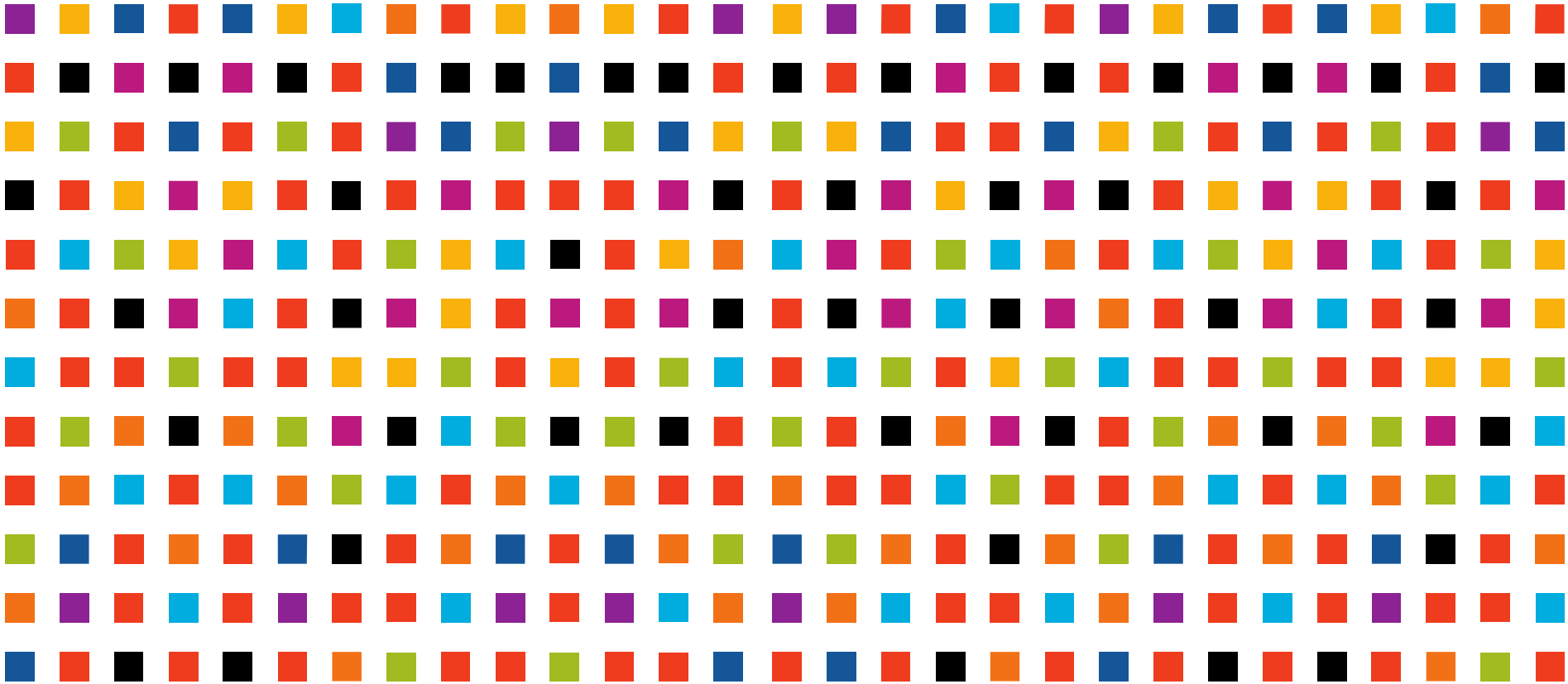
(A) Treatment with hydroxyurea for 15 days decreases tumor necrosis factor-induced thrombosis in the lungs of *Pdgfb-iCreERT2;JAK2$^{V617F/WT}$* mice. Treatment with hydroxyurea decreases (B) rolling and (C) adhesion of leukocytes on mesenteric venules in *Pdgfb-iCreERT2;JAK2$^{V617F/WT}$* mice treated with tumor necrosis factor. (D) Pre-treatment of JAK2^{V617F} HUVEC with hydroxurea (HU) decreases static adhesion of neutrophils. (E) Treatment with hydroxyurea for 15 days led to a decrease of the ratio of soluble P-selectin: number of platelets in plasma of *Pdgfb-iCreERT2;JAK2$^{V617F/WT}$* mice treated with tumor necrosis factor. (F) Hydroxyurea decreases the expression of P-selectin at the surface of carotid JAK2^{V617F} endothelial cells. (G) Treatment of JAK2^{V617F} HUVEC with hydroxyurea decreases secretion of von Willebrand factor (vWF)..

Epidemiology

Myeloproliferative Neoplasms

- **Rare Blood Cancers**
- **Overproduction of white or red blood cells, or platelets**
- **Create blood flow problems**
- **Prevalence*:**
  - Polycythemia Vera (PV): 1/3 000
  - Essential Thrombocythemia (ET): 1/3 000
  - Myelofibrosis: 1/100 000

*Source: Orphanet (https://www.orpha.net/consor/cgi-bin/index.php?lng=EN)



AYMERIC.EMPEREUR@INSERM-TRANSFERT.FR

Inserm Transfert - Paris Biopark
7 Rue Watt - 75013 Paris
Tel: +33 1 55 03 01 00 / Fax: +33 55 03 01 60
www.inserm-transfert.fr