JAK2 haplotype

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What is a haplotype?

- Series of inherited single nucleotide polymorphisms along a gene or chromosomal region

- Eight haplotypes account for 94% of JAK2 alleles in UK population

- Haplotypes conserved in different populations (HapMap)
**Background: the 46/1 JAK2 haplotype**

- The 46/1 JAK2 haplotype predisposes JAK2-mutated MPN (PV, ET, PMF) in individuals of European ancestry (Cross, Kralovics, Levine)

- JAK2 mutations arise preferentially on 46/1 allele

- Odds ratio = 3.7; 46/1 accounts for ~50% of the population attributable risk of developing an MPN
Genetic predisposition to MPNs: subtle effect of 46/1

**Mendelian disease: MPN families**
- Incidence V617F positive MPN: 4 / 100,000 per annum
- Incidence in individuals with 46/1: 3 / 100,000 per annum
- Incidence in individuals without 46/1: 1 / 100,000 per annum

**JAK2 46/1 GWAS variants**

*Penetrance* of allele frequency:
- Very rare
- Rare
- Uncommon
- Common

High
Intermediate
Low
Very low

Allele frequency
Why does 46/1 predispose to JAK2 mutations?

- **Hypermutability**: there is a specific mutational mechanism by which JAK2 mutations preferentially arises on the 46/1 haplotype

- **Fertile ground**: mutation rate on 46/1 and non-46/1 haplotypes is the same but the probability of MPN development is higher if mutation arises on 46/1
Multiple acquisition of JAK2 V617F

Polyclonal stem cell pool

- del13q
- Jak2V617F
- 9pUPD
- del20q

100% peripheral neutrophils

GGCC = 46/1

Olcaydu et al. Nature Genetics 2009
Genome-wide association defines more than 30 distinct susceptibility loci for Crohn’s disease

Jeffrey C Barrett1, Sarah Hansoul2, Dan L Nicolae3, Judy H Cho4, Richard H Duerr5,6, John D Rioux7,8, Steven R Brant9,10, Mark S Silverberg11, Kent D Taylor12, M Michael Barmada6, Alain Bitton13, Themistocles Dassopoulos9, Lisa Wu Datta9, Todd Green6, Anne M Griffith14, Emily O Kistner15, Michael T Murtha4, Miguel D Regueiro6, Jerome I Rotter12, I Philip Schumm15, A Hillary Steinhart11, Stephan R Targan13, Rammik J Xavier16, the NIDDK IBD Genetics Consortium33, Cécile Libioule2, Cynthia Sander2, Mark Lathrop7, Jacques Belaiche16, Olivier Dewit19, Ivo Gut17, Simon Heath17, Debby Laukens39, Myriam Nin2, Paul Rutgeerts21, André Van Gossum22, Diana Zelenika17, Denis Franchimont22, Jean-Pierre Hugot23, Martine de Vos20, Severine Vermeire21, Edouard Louis18, the Belgian-French IBD Consortium39, the Wellcome Trust Case Control Consortium33, 34, Lon R Cardon1, Carl A Anderson1, Hazel Drummond24, Elaine Nimmo24, Tariq Ahmad25, Natalie J Prescott26, Clive M Omnie26, Sheila A Fisher26, Jonathan Marchini27, Jilur Ghori26, Suzannah Bumpstead28, Rhiannon Gwilliam28, Mark Tremelling29, Panos Deloukas28, John Mansfield30, Derek Jewell31, Jack Satsangi24, Christopher G Mathew26, Miles Parkes28, Michel Georges3 & Mark J Daly32

Several risk factors for Crohn’s disease have been identified in recent genome-wide association studies. To advance gene discovery further, we combined data from three studies on Crohn’s disease (a total of 3,230 cases and 4,829 controls) and carried out replication in 3,664 independent cases with a mixture of population-based and family-based controls. The results strongly confirm 11 previously reported loci and provide genome-wide significant evidence for 21 additional loci, including the regions containing STAT3, JAK2, IKO, TSLG, CDO1, CDKAL1 and TRIG. The expanded molecular understanding of the basis of this disease offers promise for informed therapeutic development.


46/1 predisposes to Crohn’s disease

Zhang et al., British Medical Bulletin 2008
Is 46/1 associated with variation in haemopoietic colony numbers in healthy controls?

rs12340895:
- G allele = 46/1
- C allele = not 46/1
Does 46/1 influence hematological parameters or clinical course?

- UK PT-1 cohort (n=751, of which 404 were V617F positive)
- rs12340895 (46/1 tag SNP)

- No association between 46/1 and clinical features or outcome
  - Age, gender, duration of disease, Hb, wcc, platelets, epo, spleen size
  - survival, arterial thrombosis, venous thrombosis, major hemorrhage or transformation to MF, MDS or acute leukemia

- No association with clinical phenotype in ET or PMF (Tefferi et al. 2010; Guglielmelli et al., 2010 in press)
Does 46/1 influence hematological parameters or clinical course?

**JAK2 germline genetic variation affects disease susceptibility in primary myelofibrosis regardless of V617F mutational status: nullizygosity for the JAK2 46/1 haplotype is associated with inferior survival**

A Tefferi1, TL Lash3, MM Patnaik3, CM Finke3, K Hussein3, WJ Hogan1, MA Elliott1, MR Litzow1, CA Hanson2 and A Pardanani1

1Division of Hematology, Department of Medicine, Mayo Clinic, Rochester, MN, USA and 2Division of Hematopathology, Department of Laboratory Medicine, Mayo Clinic, Rochester, MN, USA

Figure 2  Survival in 130 patients with primary myelofibrosis stratified according to the JAK2 46/1 haplotype single-nucleotide polymorphism (SNP) genotype (rs12343867).

Tefferi et al., Leukemia. 2010 Jan;24(1):105-9.
### Does 46/1 predispose to other mutations in MPN?

<table>
<thead>
<tr>
<th>category</th>
<th>Number of cases</th>
<th>Number of 46/1 alleles</th>
<th>Number of non-46/1 alleles</th>
<th>Frequency of 46/1</th>
<th>P value (vs WTCCC controls)</th>
<th>OR (95% CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td>All PT-1 cases</td>
<td>751</td>
<td>511</td>
<td>991</td>
<td>0.341</td>
<td>1.7e-9</td>
<td>1.52 (1.34-1.74)</td>
</tr>
<tr>
<td>JAK2 V617F positive</td>
<td>404</td>
<td>302</td>
<td>506</td>
<td>0.374</td>
<td>3.9e-11</td>
<td>1.76 (1.49-2.07)</td>
</tr>
<tr>
<td>MPL exon 10 mutation positive*</td>
<td>32</td>
<td>23</td>
<td>41</td>
<td>0.359</td>
<td>0.060</td>
<td>1.60 (0.99-2.75)</td>
</tr>
<tr>
<td>JAK2 V617F, MPL exon 10 negative</td>
<td>318</td>
<td>189</td>
<td>447</td>
<td>0.300</td>
<td>0.025</td>
<td>1.24 (1.03-1.50)</td>
</tr>
<tr>
<td>WTCCC controls</td>
<td>1492</td>
<td>757</td>
<td>2227</td>
<td>0.252</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>

* Three MPL mutant cases also tested positive for V617F
Does 46/1 predispose to other mutations in MPN?

- Genotyped rs12340895 in 176 additional MPL mutated cases
  - all negative for V617F JAK2 by ARMS (sensitivity 1-2%)
  - W515L (n=110), W515K (n=58), W515v (n=4), S505N (n=4)
  - UK (n=56); Germany (n=69); Italy (n=27); Greece (n=24)

<table>
<thead>
<tr>
<th>Control cohorts</th>
<th>number</th>
<th>f(46/1)</th>
</tr>
</thead>
<tbody>
<tr>
<td>UK (WTCCC)</td>
<td>n=1496</td>
<td>0.252</td>
</tr>
<tr>
<td>Germany (KORA)</td>
<td>n=1814</td>
<td>0.280</td>
</tr>
<tr>
<td>Italy (InChianti)</td>
<td>n=1230</td>
<td>0.281</td>
</tr>
<tr>
<td>Greece</td>
<td>n=108</td>
<td>0.255</td>
</tr>
</tbody>
</table>
46/1 and MPL mutations: odds ratio meta-analysis

(No excess of 46/1 seen in 22 MPL-mutated cases; Patnaik et al., Leukemia 2010)
Genotyping results using the 46/1 tag SNP rs10118930 in MPL mutated cases and controls

<table>
<thead>
<tr>
<th>category</th>
<th>Number of cases/controls</th>
<th>Number of 46/1 alleles</th>
<th>Number of non-46/1 alleles</th>
<th>Frequency of 46/1</th>
<th>P value</th>
<th>OR (95% CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td>UK cases</td>
<td>56</td>
<td>38</td>
<td>78</td>
<td>0.339</td>
<td>0.164</td>
<td>1.41 (0.92-2.13)</td>
</tr>
<tr>
<td>UK (WTCCC) controls</td>
<td>1498</td>
<td>799</td>
<td>2197</td>
<td>0.267</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>German cases</td>
<td>69</td>
<td>49</td>
<td>89</td>
<td>0.355</td>
<td>0.189</td>
<td>1.27 (0.87-1.83)</td>
</tr>
<tr>
<td>German (KORA) controls</td>
<td>1814</td>
<td>1099</td>
<td>2529</td>
<td>0.303</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Greek cases</td>
<td>24</td>
<td>22</td>
<td>26</td>
<td>0.458</td>
<td>0.008</td>
<td>2.42 (1.20-4.82)</td>
</tr>
<tr>
<td>Greek healthy controls</td>
<td>110</td>
<td>57</td>
<td>163</td>
<td>0.259</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Italian cases</td>
<td>27</td>
<td>23</td>
<td>31</td>
<td>0.426</td>
<td>0.022</td>
<td>1.90 (1.05-3.39)</td>
</tr>
<tr>
<td>Italian (InCHIANTI) controls</td>
<td>1209</td>
<td>679</td>
<td>1739</td>
<td>0.281</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Total cases</td>
<td>176</td>
<td>132</td>
<td>220</td>
<td>0.375</td>
<td>0.0003</td>
<td>1.51 (1.20-1.90)</td>
</tr>
</tbody>
</table>
Predisposition to JAK2 and MPL mutations

EPOR

TPOR (MPL)
Are MPL mutations associated with MPL haplotype?

• WTCCC controls: LDMapper and PHASE analysis
• 88% of MPL alleles in UK accounted for by 4 haplotypes
• Captured by 3 tag SNPs

• No difference in tag SNP frequency between cases and controls:

<table>
<thead>
<tr>
<th>SNP</th>
<th>MPL mutated cases N=139</th>
<th>WTCCC controls N=1500</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>rs1199038</td>
<td>0.391</td>
<td>0.388</td>
<td>1.0</td>
</tr>
<tr>
<td>rs11210838</td>
<td>0.141</td>
<td>0.147</td>
<td>1.0</td>
</tr>
<tr>
<td>rs839757</td>
<td>0.344</td>
<td>0.341</td>
<td>0.9</td>
</tr>
</tbody>
</table>
Is the association between 46/1 and JAK2 mutations unique?

A sequence variant at 4p16.3 confers susceptibility to urinary bladder cancer

Kiemeney et al., Nat Genet. 2010

- T allele of rs798766 significantly more frequent in Ta tumors with FGFR3 mutations (OR = 2.81, 95% CI 1.21–6.51, P = 0.016, n=90).
Why does JAK2 46/1 predispose to MPN?

- No difference in expression levels between 46/1 and non-46/1 (in peripheral blood leukocytes)
- No difference in JAK2 exon and UTR sequence between 46/1 and non-46/1
- No difference in JAK2 splicing between 46/1 and non 46/1
<table>
<thead>
<tr>
<th>Hypermutability</th>
<th>Fertile Ground</th>
</tr>
</thead>
<tbody>
<tr>
<td>Excess of 46/1 in JAK2 mutated MPN</td>
<td>✓</td>
</tr>
<tr>
<td>Multiple acquisition of JAK2 mutations</td>
<td>✓</td>
</tr>
</tbody>
</table>
Fertile ground hypothesis suggests incomplete penetrance of JAK2 mutations?

Brief report

JAK2<sup>V617F</sup>: prevalence in a large Chinese hospital population

Xuesong Xu,1,2 Qi Zhang,1 Jian Luo,1 Shu Xing,1,3 Qingshan Li,1 Sanford B. Krantz,4 Xueqi Fu,1 and Zhizhuang Joe Zhao1,3

1Edmond H. Fischer Signal Transduction Laboratory, College of Life Sciences, Jilin University, Changchun, China; 2Clinical Laboratory, China Japan Union Hospital, Jilin University, Changchun, China; 3Department of Pathology, University of Oklahoma Health Sciences Center, Oklahoma City; 4Hematology/Oncology Division, Department of Medicine, Vanderbilt-Ingram Cancer Center, Vanderbilt University, Nashville, TN

Blood. 2007;109:339-42

Is the JAK2<sup>V617F</sup> mutation detectable in healthy volunteers?

Christophe Martinaud,1 Patrick Brisou1 and Marie-Joelle Mozziconacci2*

Am J Hematol. 2010;85:287-8
Acknowledgements

David Oscier
Philip Beer
Peter Campbell
Linda Scott
Tony Green
Melanie Percy
Claire Harrison

Andreas Reiter
Andreas Hochhaus
Francisco Cervantes
Katerina Zoi
Heike Pahl
Holger Cario
Susanne Schnittger
Alessandro Vannucchi
Paola Guglielmelli

Richard Silver

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