Expression of JAK2 V617F Mutation in Chronic Myeloproliferative Disorders

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Abstract

JAK2 is a non receptor tyrosine kinase that plays a major role in myeloid development by transducing signals from diverse cytokines and growth factor receptors, a mutation in this Janus kinase interferes with the negative regulation of JAK2 and could account for the hypersensitivity of myeloid cells from MPD patients to growth factors.

JAK2 V617F mutation may play a very important role not only in the development of MPD but also in phenotypic presentation of the disease, as well as it may be useful for the diagnosis, management and follow up of the disease as the mutation increases on relapses and decreases denoting response to treatment hence it may be also used as a predictive test for the treatment outcome in CMPDs patients.

Key Words:

JAK2 V617F, MPD

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List of abbreviations

Abbreviation	The Full term
ABL	Abelson strain of murine leukemia virus
ALL	Acute Lymphoblastic Leukemia
ALCL	Anaplastic Large Cell Lymphoma
AML	Acute Myeloblastic Leukemia
ATP	Adenosine Triphosphate
BCR	Breakpoint Cluster Region
BHQ	Black Hole Quenchers
BM	Bone Marrow
CBC	Complete Blood Count
CD	Cluster of Differentiation
CEL	Chronic Eosinophilic Leukemia
CEL/NOC	Chronic Eosinophilic Leukemia Not Otherwise Classified
CI	Confidence Interval
CIMF	Chronic Idiopathic Myelofibrosis
CLL	Chronic Lymphocytic Leukemia
CMPD	Chronic Myeloproliferative Disorder
CML	Chronic Myelogenous Leukemia
CNL	Chronic Neutrophilic Leukemia
cDNA	Complementary Deoxyribonucleic Acid
del	Deletion
DNA	Deoxyribonucleic Acid
DVT	Deep Venous Thrombosis
EDTA	Ethylene Diamine Tetra-Acetic acid
EEC	Endogenous Erythroid Colony
EPO	Erythropoietin
EPOR	Erythropoietin Receptor
ET	Essential Thrombocythemia
F	Phenylalanine
FERM	Band our-point-one , Ezerin , Radixin , Moesin
FGFR-1	Fibroblast Growth Factor Receptor – 1
FISH	Fluorescence In Situ Hybridization
FLT3	Fms-Like Tyrosine kinase-3
FRET	Fluorescence Resonance Energy Transfer
G	Guanine
G-CSF	Granulocyte Colony Stimulating Factor
G-CSFR	Granulocyte Colony Stimulating Factor Receptor
GH	Growth Hormone
GM-CSF	Granulocyte Monocyte Colony Stimulating Factor
GM-CSFR	Granulocyte Monocyte Colony Stimulating Factor Receptor
Hb	Hemoglobin
Hct	Hematocrit
HES	Hypereosinophilic Syndrome
HSC	Hematopoietic Stem Cell
HS	Highly Significant

Abbreviation	The Full term
HTLV-1	Human T Cell Lymphotropic Virus-1
IL	Interleukin
IMF	Idiopathic Myelofibrosis
JAK	Janus Kinase
JH	Janus Homology
JMML	Juvenile Myelomonocytic Leukemia
LAP	Leucocyte Alkaline Phosphatase
LDH	Lactate Dehydrogenase
LIF	Leukemia Inhibitory Factor
LOH	Loss of Heterozygosity
LPS	Lipopolysaccharides
MAPK	Mitogen Activated Protein Kinase
Mbp	Major Break Point
MCD	Mast Cell Disease
MDS	Myelodysplastic Syndrome
MGB	Minor Groove Binder
Mg	Magnesium
mg	Milligram
MMM	Myelofibrosis with Myeloid Metaplasia
Mmol	Millimole
Mn	Manganese
MPD	Myeloproliferative Disorder
MPN	Myeloproliferative Neoplasm
mRNA	Messenger Ribonucleic Acid
Nk	Natural Killer
NS	Non Significant
OR	Odds Ratio
PCR	Polymerase Chain Reaction
PCM1	Pericentriolar Material - 1
PDGFRA	Platelet Derived Growth Factor Receptor A
PDGFRB	Platelet Derived Growth Factor Receptor B
PDGF	Platelet Derived Growth Factor
Ph	Philadelphia
PIAS	Protein Inhibitors of Activated STAT family
PI3K	Pathways Including phophoinositide 3 Kinase
Plts	Platelets
PMBL	Primary Mediastinal B Cell Lymphoma
PNA	Peptide Nucleic Acid
PRV-1	Polycythemia Rubra Vera
PV	Polycythemia Vera
PVSG	Polycythemia Vera Study Group
P-value	Probability Value
RA	Refractory Anemia
RAS	Murine Sarcoma Virus
RCM	Red Cell Mass
RT	Reverse Transcriptase
S	Significant

Abbreviation	The Full term
SCF	Stem Cell Factor
SCID	Severe Combined Immunodeficiency
SHP	SRC Homology Phosphatase
SLIM	STAT Interacting LIM proteins
SNP	Single Nucleotide Polymorphism
SOCS	Suppressor Of Cytokine Signalling proteins
STAT	Signal Transducers and Activators of Transcription
SUMO	Small Ubiquitin -Like Modifier
t	Translocation
T	Thymine
Tm	Melting Time
TPO	Thrombopoietin
TPOR	Thrombopoietin Receptor
TYK	Tyrosine Kinase
µL	Micro Liter
V	Valine
WBCs	White Blood Cells
WT	Wild Type
WHO	World Health Organization
X-CIP	X-Chromosome Inactivation Pattern

Introduction

The myeloproliferative disorders (MPDs) comprise a spectrum of chronic haematologic diseases. They are a heterogeneous group of diseases, characterised by increased numbers of differentiated blood cells and are believed to arise in a multipotential haemopoietic progenitor. They include polycythaemia vera (PV), essential thrombocythemia (ET), primary idiopathic myelofibrosis (IMF) (the classic MPDs), as well as chronic eosinophilic leukaemia/hypereosinophilic syndrome (CEL/HES), systemic mastocytosis and chronic myeloid leukemia (CML) ***(Campbell and Green, 2006)***[16]**.**

Polycythaemia Vera (PV) and essential thrombocythemia (ET) share several features: a hypercellular marrow with overproduction and predominance of one lineage; hypersensitivity to cytokines such as erythropoietin (EPO); presence of extramedullary haemopoiesis; progression of a significant proportion of cases to myelofibrosis and a relatively low propensity for evolution to acute leukemia in the absence of the use of leukaemogenic cytoreductive treatment. The role of the BCR-ABL tyrosine kinase in CML pathophysiology has now been established for some years and more recently it has become apparent that aberrant tyrosine kinase activity is also associated with other MPDs ***(De Keersmaecker and Cools, 2006)***[23]**.**

Diagnosis of the MPDs is difficult due to lack of diagnostic markers. Recently, the acquisition of a mutation in the Janus kinase 2 (JAK2) gene by haemopoietic cells has been described as a genetic defect underlying myeloproliferative disorders. The mutation leads to constitutive activation of JAK2, a tyrosine kinase involved in cytokine receptor signalling ***(Poodt et al., 2006)***[110]**.** So the impact of the JAK2

V617F mutation on the cytokine signalling pathways suggests that it plays an important role in the pathogenesis of MPDs and represents a major breakthrough in molecular understanding of the myeloproliferative disorders (MPDs) that may have significant implications for diagnosis and treatment ***(Skoda.,2007)***[132]**.**

The JAK family of tyrosine kinases includes JAKs 1–3 and tyrosine kinase 2 (TYK2). JAKs are expressed ubiquitously in all cells with the exception of JAK3, which is found only in haematopoietic cells ***(Skoda, 2007)***[132]**.** The most important structural domain of the JAK molecules is the enzymatic kinase domain (JH1), which phosphorylates tyrosines on target proteins. The pseudokinase domain (JH2) has no enzymatic activity and is thought to inhibit the kinase domain, while the FERM domain is important in regulating binding of the JAK proteins to cytokine receptors ***(Khwaja. , 2006)***[53]**.**

JAK2 has a critical role in mediating the signalling pathways of thrombopoietin (TPO), erythropoietin (EPO) and other cytokines involved in haemopoiesis. JAK2 is activated by the binding of these ligands to cytokine receptors ***(Larsen et al., 2007)***[68]**.** Activation of cytokine receptors by JAK2 also stimulates several other signalling pathways mediated by adaptor proteins, including Ras, mitogen-activated protein kinases, phosphoinositide 3 kinases, protein kinase B and phospholipase C. Together, these events alter the proliferation, differentiation and survival of haematopoietic cells ***(Royer et al., 2005)***[120].

The V617F somatic mutation in the Janus kinase 2 (JAK2) gene, which causes the substitution of phenylalanine for valine at amino acid position 617 (V617F), has recently been found in the majority of patients with polycythaemia (PV) and in many with essential thrombocythemia

(ET) or idiopathic myelofibrosis ***(Scott et al., 2007)***[127]**.** This gene encodes a cytoplasmic tyrosine kinase. The mutation, which occurs in the JAK homology 2 (JH2) negative regulatory domain increases JAK2 kinase activity and causes cytokine-independent growth of cell lines and cultured bone marrow cells. Mutant JAK2 transfected into murine bone marrow cells produces PV-like phenotypes including erythrocytosis and subsequent myelofibrosis in recipient animals, suggesting a causal role for the mutation ***(Wernig et al., 2006)***[171]**.**

The main functional consequence of the JAK2 V617F tyrosine kinase mutation is an increased sensitivity of cytokine receptors to incoming signals. The activating effect of the mutation is such that the JAK2 complex becomes partially independent of the signal ***(Kralovics et al., 2005)***[58]**.**

Based on the current understanding of JAK2 V617F, the onset of MPD is marked by the acquisition of the somatic JAK2 V617F mutation in the heterozygous state or a mutation in an as-yet-unknown gene. Following acquisition of the JAK2 V617F mutation, mitotic recombination can subsequently lead to the mutation occurring in the homozygous state. By allowing duplication of the mutated JAK2 gene and elimination of wild-type JAK2, mitotic recombination could explain observations of a more aggressive disease course in patients with homozygous JAK2 V617F ***(Vannuchhi et al, 2006)***[162]**.** The majority of the MPDs patients were heterozygous for V617F, however, many were hemizygous (one mutant and deleted wild-type gene), or homozygous (two mutant genes) ***(Ma et al., 2006)***[82]**.**

Although the JAK2 V617F mutation is sufficient to cause features resembling PV in animal models, the situation in human MPD is