Detection of Janus kinase 2 (JAK2V617F) in Sudanese Patients with polycythemia in Khartoum State, Sudan

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Abstract

Background & Aims

Erythrocytosis is form of hematological diseases, the main members of which are polycythemia vera (PV) , familial polycythemia and idiopathic erythrocytosis. The molecular pathogenesis of these disorders is unknown, but tyrosine kinases have been implicated in several related disorders. We investigated the presence of the cytoplasmic tyrosine kinase JAK2 in patients with polycythemia and the hematological difference between these types in Sudanese patients.

Methods

We measure the full blood counts and obtained DNA samples from patients with erythrocytosis. The *JAK2V617F mutation* was detected from peripheral-blood samples. Allele-specific PCR were undertaken on subgroups of patients who satisfied the WHO criteria of Polycythemia vera.

Results

A single point mutation (V617F) was identified in JAK2 in 31 (54.4%) of 57 patients with polycythemia and there are no significant differences of hemoglobin, Hematocrit, and neutrophil between patients with the JAK2 mutation and who without the mutation. Polycythemia Vera patients who are not detected for the mutation and patients with Idiopathic erythrocytosis showing no difference except in Platelet's Count .

Interpretation

A single mutation of JAK2V617F was noted in more than half of patients with erythrocytosis. If the person is negative for JAK2V617F mutations, the person may still have a PV. The person could have a JAK2 exon 12 mutation which was not detected during research.

Rational

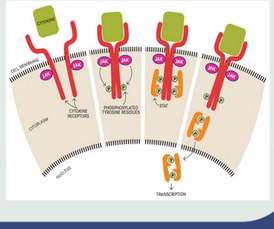
Many polycythemia's Sudanese patients who are currently on phlebotomy therapy without known what the actually causes of Raised Hemoglobin levels (PV, Secondary, Apparent polycythemia or idiopathic erythrocytosis). Patients with PV frequently demonstrate a complex of hematological disorder and hemoagulation disorders like thrombosis and paradoxical hemorrhage due increase in blood viscosity which may lead to cerebral and myocardial infarction. In the longer term, these disorders can develop secondary myelofibrosis or transform into acute myelogenous leukemia (AML) or also can triggering mechanism in the development of the DIC syndrome. So our studies determined the important to use JAK2 mutation (V167F) detection as a necessary investigation in patients who have high level of hemoglobin and Packed cell volume according to WHO criteria especially who high leukocyte and platelets count as a diagnostic test for patients with polycythemia in Sudan to use other treatment with veinsection and know the frequency of JAK2 mutation in our population in Sudan.

Polycythrmia and Jak2 Mutation

Erythrocytosisis defined as an increase in red cells (or blood hemoglobin) per unit volume. Polycythemia (also called absolute erythrocytosis) indicates an absolute increase in red cell mass and reflects an increase in red blood cell (RBC) production. Since the red cell count given on the complete blood count (CBC) reflects a ratio of red cells per unit volume, it is possible to have an increase in the RBC count (or hemoglobin or hematocrit) due to either an increase in the red cell mass (polycythemia) or a decrease in the plasma volume (relative erythrocytosis). Polycythemia can be a response to some other condition causing hypoxemia (secondary polycythemia), can result from inappropriate erythropoietin secretion due to renal disease or some neoplasms, or can represent an autonomous neoplastic condition (polycythemia Vera). Polycythemia Vera (sometimes called polycythemia rubra Vera) is a chronic myeloproliferative disorder

In 1992, JAK2 was mapped on the short arm of chromosome 9p24 by Pritchard and his colleagues; it has 140 kb spanning 25 exons to form 1132 amino acid JAK2 protein.

In 2005, four separate groups using different modes of discovery and different measurement techniques reported the presence of a novel somatic point mutation in the conserved autoinhibitory pseudokinase domain of the gene coding for the JAK2 protein in patients with classic MPNs. The mutation was noted to cause a valine-to-phenylalanine substitution at amino acid position 617 (JAK2V617F). Loss of JAK2 autoinhibition caused by JAK2V617F results in constitutive activation of the kinase and in recruitment and phosphorylation of substrate molecules including signal transducers and activators of transcript (STAT) proteins (JAK-Stat signaling) Figure 1-1
In 2007, JAK2 gene exon 12 mutations are specific to JAK2V617f negative PV and were first described. JAK2 exon 12 mutations include in-frame deletions, point mutations and duplications, mostly affecting seven highly conserved amino-acid residues (F537–E543). The clinical course of these patients seems to be similar to that of patients with JAK2V617F-positive.



Result and Disscution

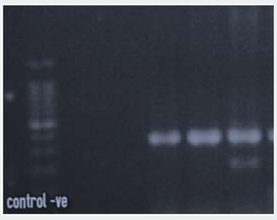
Whole blood samples of polycythemia's patients that were collected from Hospital's blood banks for veinsection therapy or who already diagnostic with polycythemia were examined using cells counter machines (Sysmex kx21) to detect hematological parameters (Hemoglobin, Hematocrit, platelets, Neutrophil). All samples that characterized the Polycythemia Vera criteria (WHO criteria) will be tested by Polymerase chain reaction for JAK2V617F mutation

The majority of the Patients who included in this study were with Polycythemia Vera criteria (WHO criteria) (68.55%), followed by Smokers (26.15%). Other Idiopathic erythrocytosis showed the lowest values (5.3%).

The JAK2 mutation was detected by the qualitative method in 31/57 (54.4%) patients.

The samples for this study were received from patients who were referred for investigation of one or more of the following: high hemoglobin level, high platelet count, neutrophilia, or hematopathologic findings suggestive of Polycythemia. A total of 130 cases were evaluated Cases classified By the World Health Organization criteria as one of the following: PV (89 cases) and idiopathic erythrocytosis (34 cases smokers and 7 non-smokers). There is no clinically significant difference between tow group in hematological department except in platelets count which were gave a significant difference (P=0.009). However, in 2009 other study show there is no significant difference in all Hematological parameters between polycythemia Vera and idiopathic erythrocytosis

At present, finding the *JAK2* mutation is diagnostic for MPD, given the high specificity of the mutation for clonal myeloid diseases. The observation in this study shows that the V617F mutation alteration is seen in 54.4% of patients who satisfied the WHO criteria for polycythemia Vera Patients. Our results showed close percentage with other research in other country rather than Sudan; 69.6% Parma-Italy 2009, 26% Cambridge-UK 2005, 47.1% China 2014



		N	P value
Hb g/dl	Positive	31	0.733
	Negative	26	
HCT %	Positive	31	**0.05**
	Negative	26	
PLTs	Positive	31	0.15
	Negative	26	
Neutrophil	Positive	31	0.2
	Negative	26	

Conclusion

While the *JAK2* mutation is already a diagnostic value, it is reasonable to consider that in the near future, it will also play a huge role in the choice of treatment and monitoring of erythrocytosis by given the development of drugs inhibiting the JAK2 pathway

JAK2V617F mutation was reported in a large proportion of Sudanese patients; this should be considered when investigations of patients with erythrocytosis are carried out, and JAK2V617F inhibitors should be implemented for those with the mutation.

Polycythemia's patients with JAK2 V617F negative should tested for JAK2 Exon12 Mutation.

Sequence analysis for patients with positive V617F mutation which will show the mutant peak and determined the frequency of the heterozygous and homozygous allele in Sudan

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