Suggested Modification of Janus Associated Kinase 2-Tree Algorithm for the Detection of Janus Associated Kinase 2 V617F-Positive Polycythemia Rubra Vera Patients in Pakistani Population

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Abstract:
OBJECTIVE: The Janus associated kinase-2 (JAK2 mutation V617F and exon 12) is detected in most polycythemia vera (PV) patients. It can easily be picked up by real-time polymerase chain reaction (PCR). This technique is easy to perform and is very sensitive requiring minimally invasive techniques. The decision when to order JAK2 mutation test by PCR is important to curb its wastage. To save precious laboratory resources, a peripheral blood finding-based algorithm adjusted to hemoglobin levels of patients (PV) in Pakistan has been purposed, which is a modification of algorithm recently developed by Mahe et al. for the rationalization of JAK2 analysis.

METHODS: To assist with the screening of patients being considered for JAK mutation for PV patients in the Pakistani population, we modified clinical decision rule “JAK2-tree” as modified JAK2-tree based on patients’ full blood count.

RESULTS: We tested both classical and modified JAK2-tree algorithms on two independent data sets, one an unselected population-based sample comprising 51 individuals and other on historical clinical laboratory referral set comprising 51 JAK2-positive cases of PV. Sensitivity for both the algorithms was calculated and compared.

CONCLUSION: Our work supports a “modified” decision-tree-based screening approach for Pakistani population to optimize the selection of patients most appropriate for JAK2 V617F testing.

Keywords: Janus associated kinase 2-tree, modified, Pakistani population

Introduction
The criteria published by the World Health Organization (WHO) for the diagnosis of polycythemia vera (PV) have never been evaluated in the Pakistani population so far.[1] The 2016 WHO criteria for the diagnosis of PV include three major criteria and one minor criterion. Major criteria include hemoglobin (Hb) levels of 16.5 g/dl in men and 16 g/dl in women, or other evidence of increased red cell volume or hematocrit (Hct),[2] and presence of JAK2 V617F or JAK2 exon 12 mutation and bone marrow biopsy, showing hypercellularity for age with trilineage growth (panmyelosis) with prominent erythroid, granulocytic, and megakaryocytic proliferation.[3] Minor criterion includes serum erythropoietin (EPO) levels below the reference range for normal. The JAK2

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gene at chromosome 9p24 encodes a tyrosine kinase involved in cytokine receptor signaling through the STAT pathway. When constitutively activated, most often by way of the JAK2 Val 617 Phe (V617F) mutation, the result is an over-responsive kinase to even low-level cytokine stimulation (including EPO). The JAK2 V617F and JAK2 mutations (exon 12) are present in almost every PV patients.[4,5] The clinical indication to order JAK2 mutation by polymerase chain reaction has been under debate nowadays. Usually, the decision to proceed with JAK2 testing is based on the demonstration of an increase in Hb or Hct. To assist screening of patients being considered for JAK2 testing, Mahe et al. developed a clinical decision rule, “JAK2-tree,” which can be easily applied to basic full blood count (FBC) parameters, such as Hb, platelet, and white blood cell counts (total leukocyte count [TLC]). However, JAK2-tree may be inadequate for diagnosing “early-stage” PV in our local population as there is a major difference in normal reference ranges for Hb in our and Western population.[6] We retrospectively evaluated the accuracy of both JAK2-tree and modified JAK2-tree in diagnosing PV, especially in “early-stage” patients in the Pakistani population [Figure 1].[6]

**Methods**

To assist with the screening of patients being considered for JAK mutation for PV patients in Pakistani population, we modified clinical decision rule “JAK2-tree” as modified JAK2-tree based on patients’ FBC. We retrospectively analyzed data of 51 randomly selected JAK2 V617-positive patients with similar number of individuals from normal population. We calculated their mean values for their Hb (g/l) levels, TLC, and platelet count. Based on the data obtained, we proposed a modified JAK2-tree algorithm where cutoff for Hb in males was 16 g/dl instead of 16.5 g/dl, and for females, it was 15.5 g/dl instead of 16 g/dl. We calculated the sensitivity of both classical and modified JAK2 on data sets of both normal and JAK2 population using IBM Statistical Package for the Social Sciences for windows, version 24.0 (IBM Corp., Armonk, N.Y., USA).

**Results**

We tested classical and modified JAK2-tree on two independent data sets, one historical clinical laboratory referral set of 51 individuals [Table 1] and the other referral set comprising 51 JAK2-positive cases of PV [Table 2]. We found sensitivities for classical and modified JAK2 to be 11.7% and 23.5%, respectively, for historical clinical laboratory referral set against classical JAK2-tree, which was 70.5% and 88.2% positivity for known cases of JAK2. Thus, modified JAK2-tree if applied to our historical laboratory referral dataset would have reduced JAK2 V617F testing volume over the period of evaluation by 25%.

**Discussion**

The modified JAK2-tree algorithm could be used as a simple screening tool to select patients most appropriate for JAK2 V617F mutation testing to diagnose PV. This algorithm can easily be applied using the routine FBC parameters. Our study has shown it to have a positive predictive value of 95% and negative predictive value of 96% for the detection of JAK2-positive patients. The JAK2-tree algorithm developed by Mahe et al. demonstrated a sensitivity of 94% and a negative predictive value of 92% for the prediction of a JAK2 V617F mutation in their population, which are similar to our study. It is highly recommended that only modified JAK2-tree may be applied to in our population to rationalize the use of JAK2 testing. Further studies are however required to support our findings. The modified JAK2-tree for PV is not designed to be used in predicting thrombosis at unusual sites or to provide molecular confirmation of a previous clinical diagnosis of an myeloproliferative neoplasm or to influence therapy.[7-9]

**Conclusion**

Clinical rules should be applied to expensive laboratory tests to reduce costs and unnecessary wastage.[10,11] Simple clinical decision rules such as “modified JAK2-tree for PV” can assist in appropriate test ordering and detecting underdiagnosed cases in our population. Further studies are suggested in this regard.

**Ethics approval**

The study was approved by CMH Lahore Ethical Committee.

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Conflicts of interest
There are no conflicts of interest.

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