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Retrospective analysis of patients with chronic myeloproliferative neoplasms: a single-center experience

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SUMMARY

Chronic myeloproliferative diseases (CMPD) are clonal diseases that may cause hemostatic and thrombotic abnormalities. They progress to acute leukemia and are characterized by increases in the number of mature and immature cells in the peripheral blood as a result of uncontrolled proliferation of one or more than one type of myeloerythroid cells in the bone marrow. This study aimed to determine demographic features, incidence of Janus kinase 2 (JAK2) mutations, disease characteristics, and treatment strategies in patients diagnosed with CMPD.

A total of 100 patients with CMPD [essential thrombocytosis (ET), n = 52; primary myelofibrosis (PMF), n = 31; and polycythemia vera (PV), n = 17] having JAK mutations admitted to outpatient clinics of Hematology Department of Sakarya University Faculty of Medicine between February 2006 and February 2013 were included with the diagnosis of BCR/ABL (The ABL gene from chromosome 9 joins to the BCR gene on chromosome 22, to form the BCR-ABL fusion gene)-negative CMPD based on the 2008 World Health Organization criteria. Age, gender, family history, secondary cancer, bleeding, history of thrombosis, whole blood cell counts, and presence of hepatomegaly, splenomegaly, and other symptoms and signs at the time of diagnosis were evaluated. Besides, a history of thrombosis and hemorrhage was assessed. The presence of JAK mutations in DNA samples was analyzed using real-time polymerase chain reaction.

Age and gender distribution, family history, previous incidents of bleeding, thrombosis, secondary cancer, blood hemoglobin, lactate dehydrogenase values, platelet and white blood cell counts, constitutional symptoms, minor neurologic symptoms, and presence of hepatomegaly and splenomegaly at the time of diagnosis were assessed. The incidence of JAK2 mutations was highest among patients with PMF (70.9%), followed by patients with PV (70.6%) and ET (51.9%), in this study.

The incidence of JAK2 mutations has offered a different perspective in BCR/ABL- negative patients with CMPD and served as an acceptable diagnostic factor. The present study had a small sample size. Hence, large-sample studies should be conducted to confirm the relationship between this mutation and CMPD.

Key words: Essential thrombocytosis, myelofibrosis, polycythemia vera

INTRODUCTION

Chronic myeloproliferative diseases (CMPD) are clonal diseases that may cause hemostatic and thrombotic abnormalities. They progress to acute leukemia and are characterized by increases in the number of mature and immature cells in the peripheral blood as a result of uncontrolled proliferation of one or more than one type of myeloerythroid cells in the bone marrow. Polycythemia vera (PV), essential thrombocytosis (ET), and primary myelofibrosis (PMF) are included in this patient group (1). These diseases have common clinical and biological characteristics and may be transformed into one another. In PV, essentially persistent erythrocytosis is seen. In ET, typical increases in megakaryocyte and platelet counts

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are observed. In idiopathic myelofibrosis, bone marrow fibrosis is more prominent (2). In ET, PV and PMF are BCR/ABL-negative CMPDs. The effects of a few genetic mutations have been reported recently in BCR/ABL-negative CMPDs (2).

The information about the pathogenesis of BCR/ABL-negative chronic CMPD has increased with the discovery of Janus kinase 2 (JAK2) exon 14 mutation in the year 2015, and promising developments in the follow-up and treatment of the patients have been achieved (3). In almost all patients with PV and nearly half of the patients with ET and PMF have a JAK2V617F mutation (4). Based on these findings, the JAK2V617F mutation was a major criterion for PV and PMF and a minor criterion for ET according to the 2008 World Health Organization (WHO) criteria (5). The effects of JAK2 exon 12, myeloproliferative leukemia viral oncogene (MPL), ten-eleven translocation 2 (TET2), and additional sex comb-like 1 mutations on CMPD are not as obvious as that of JAK2V617F mutation (6).

This study aimed to determine demographic features, incidence of JAK2 mutations, disease characteristics, and treatment strategies in patients diagnosed with CMPD.

MATERIALS AND METHODS

Study design

The approval of the local institutional review board was obtained before the study. Among all patients who consulted outpatient clinics of Hematology Department of Sakarya University Faculty of Medicine between March 2012 and March 2014, 100 patients (ET, $n = 52$; PMF, $n = 31$; and PV, $n = 17$) diagnosed with BCR/ABL-negative CMPD based on 2008 WHO criteria and having JAK mutations. Data of the patients obtained from their files were retrospectively evaluated.

Outcome parameters

Age, gender, family history, secondary cancer, bleeding, history of thrombosis, whole blood cell counts, and presence of hepatomegaly, splenomegaly, and other symptoms and signs at the time of diagnosis were evaluated. Besides, a history of thrombosis and hemorrhage was assessed. The presence of JAK mutations in DNA samples obtained from the peripheral blood of the patients was analyzed using previously indicated methods and real-time polymerase chain reaction (7).

RESULTS

Age and gender distribution, family history, previous incidents of bleeding, thrombosis, secondary cancer, blood hemoglobin, lactate dehydrogenase (LDH) values, platelet and white blood cell (WBC) counts, constitutional symptoms, minor neurologic symptoms, and presence of hepatomegaly and splenomegaly at the time of diagnosis are shown in Table 1.

The distribution of the results of mutational analyses of the patients according to their diseases is shown in Table 2. The incidence of JAK2 mutations was highest among patients with PMF (70.9%), followed by patients with PV (70.6%) and ET (51.9%), in this study.

The distribution of the treatments received by the patients and their prognoses is shown in Table 3.

DISCUSSION

This novel study evaluated the incidence of JAK2 mutations in patients with CMPD in Sakarya, Turkey. The incidence rate of these mutations was 70.9, 70.6, and 51.9% in patients with PMF, PV, and ET, respectively. The results of the present study were in accordance with those in the literature data and the results of other studies performed in Turkey (8, 9).

The role of dysregulated tyrosine kinase, the BCR/ABL oncogene product, in the etiopathogenesis of CMPD has been acknowledged and suggested the possible role of genetic mutations in the pathogenesis of other CMPDs. In 2005, JAK2V617F mutations were detected in patients with BCR/ABL-negative CMPD in four different studies and changed the diagnostic criteria for these patients (5). More than 500 kinases that phosphorylate serine, threonine, and tyrosine residues have been detected in humans. JAK2 is one of the four different cytoplasmic tyrosine kinases (10). More than one member of the JAK family participates in many signal conduction pathways. However, only the JAK2 gene is involved in the case of growth factors, such as erythropoietin and thrombopoietin (11). JAK2 gene is localized on the short arm of chromosome 9 (12). In JAK2V617F somatic mutation, phenylalanine replaces valine at the 617th position of the pseudokinase domain of JAK2 gene (13). This newly developed mutation makes hematopoietic precursor cells extremely sensitive

Table 1: Demographic characteristics, clinical symptoms, and findings of the patients at the time of diagnosis.

		CMPN *(n = 100)*	ET *(n = 52)*	PMF *(n = 31)*	PV *(n = 17)*
Age at diagnosis, years (range)		59.07 ± 12.48 (33–86)	57.37 ± 12.86 (33–86)	64.26 ± 12.18 (41–84)	54.82 ± 8.79 (45–76)
Sex, male/female		49/51	21/31	16/15	12/5
Family history, *n*		4/100	1/52	1/31	2/17
Thrombosis at/before diagnosis, *n*	Arterial thrombosis	14/100	8/52	3/31	3/17
	Venous thrombosis	6/100	3/52	2/31	1/17
	Arterial and venous thrombosis	3/100	0	1/31	2/17
Bleeding at diagnosis, *n*	Nasal bleeding	10/100	6/52	3/31	1/17
	Gastrointestinal system bleeding	9/100	3/52	3/31	3/17
	Gingival bleeding	4/100	1/52	2/31	1/17
	Spleen bleeding	1/100	1/52	0	0
History of secondary malignancies		0	0	0	0
White blood cell count, x 10^9/L, mean (range)		12.63 (3.1–37.5)	11.87 (5.6–34.8)	13.65 (3.1–37.5)	13.12 (3.7–25.0)
Platelet count, x 10^9/L, mean (range)		752 (87–2295)	924 (248–1670)	546 (87–2295)	600 (210–1100)
Hemoglobin level, g/L, mean (range)		13.6 (6.0–21.5)	12.7 (6.0–17.7)	12.8 (8.3–21.5)	17.9 (11.9–21.0)
LDH levels, U/L, mean (range)		358 (137–1085)	298 (170–673)	477 (137–1085)	322 (165–610)
Constitutional symptoms, *n*		27/100	11/52	11/31	5/17
Minor neurologic symptoms, *n*		52/100	24/52	19/31	9/17
Pruritus, *n*		43/100	20/52	14/31	9/17
Splenomegaly, *n*		43/100	14/52	19/31	10/17
Hepatomegaly, *n*		24/100	8/52	10/31	6/17
Cardiovascular risk factors, *n*		82/100	41/52	28/31	13/17
Diabetes mellitus, *n*		22/100	14/52	6/31	2/17
Hypertension, n		55/100	28/52	16/31	11/17
Obesity, n		35/100	21/52	10/31	4/17
Dyslipidemia, n		21/100	14/52	3/31	4/17
Smoking history, n		46/100	17/52	19/31	10/17
Alcohol, n		5/100	1/52	2/31	2/17

CMPN: Chronic myeloproliferative neoplasm, ET: Essential thrombocythemia; LDH: Lactate dehydrogenase; NA: Not available, PMF: Primary myelofibrosis; PV: Polycythemia vera.

Table 2: Distribution of mutational analysis of the patients among diseased states.

	CMPN *(n = 100)*	ET *(n = 52)*	PMF *(n = 31)*	PV *(n = 17)*
BCR/ABL mutation, (positive/negative)	0/100	0/52	0/31	0/17
JAK mutation, (positive/negative)	61/39 (61%)	27/25 (51.9%)	22/9 (70.9%)	12/5 (70.6%)
Mutational analysis		52	30	17

CMPN: Chronic myeloproliferative neoplasm, ET: Essential thrombocythemia, PMF: Primary myelofibrosis, PV: Polycythemia vera.

Table 3: Distribution of the treatment and prognoses of the patients.

		ET (*n*= 52)	PMF *(n = 31)*	PV *(n = 17)*
First cytoreductive therapy (hydroxyurea)		47/52	31/31	16/17
Second cytoreductive therapy (thromboreductin)		46/52	2/31	1/17
Antiplatelet therapy		51/52	30/31	17/17
Uricolysis		46/52	30/31	17/17
Androgens		0	0	0
Steroids		1	0	0
Erythrocyte transfusion		11	17	0
Splenectomy		0	0	0
Secondary myelofibrosis	1/4	0	0	1
	2/4	6	0	3
	3/4	0	10	0
	4/4	0	20	0
Leukemic transformation		0	0	0
Stem cell transplantation		0	1	0
Alive		51	29	16

ET: Essential thrombocythemia, PMF: Primary myelofibrosis, PV: Polycythemia vera.

to growth factors (14). In the WHO-revised criteria, the presence of JAK2V617F mutation is included as the diagnostic criteria for PV, ET, and PMF. The estimated incidence rate of this mutation is 95% in PV and 50%–60% in PMF (15). Recent studies have demonstrated that complications, especially thrombotic events, are more frequently seen in patients diagnosed with CMPD having JAK2V617F mutation (16).

In patients with ET, nearly 40%–60% of the patients are expected to have JAK2V617F mutation (8, 17). MPL and TET2 mutations were detected in JAK2V617F-negative patients with ET, though at a lower frequency (18). This mutation was reported in a large case series performed by Campbell et al. (53% of 776 patients with ET), Carabbio et al. (57% of 867 patients with ET) and Karkucak et al. (42% of 78 patients with ET) in Turkey (8, 19). In parallel with these data, the JAK2 mutation was seen in 27 of 62 patients with ET in this study (51.9%). Antonioli et al. investigated the impact of JAK2V617F in patients with ET on clinical characteristics and detected a correlation between increased hemoglobin and WBC counts at admission and faster conversion to leukemic state (20). Toyama et al. detected a significant correlation between the presence of this mutation and higher WBC counts and incidence of thrombosis in patients with ET (21). In another study on 106 patients with ET, the presence of this mutation was found to be correlated with increased hematocrit levels, higher WBC counts, and lower platelet counts (22). In the present study, higher platelet counts and increased frequency of platelet thrombosis and bleeding were detected in patients with ET.

PMF has a complex physiopathology. It is the most rarely seen disease among CMPDs. PMF is a clonal, multipotent, hematopoietic progenitor cell disorder with unknown etiology (23). Detection of specific molecular lesions as a JAK2V617F mutation is important for the diagnosis, treatment, and prognosis of these patients with PMF (23). Phase II studies are being performed on the treatment of patients with PMF using JAK2 inhibitors (CEP 701) (24). The JAK2V617F mutation is seen in nearly 50% of patients with PMF, although the frequency of these mutations in patients with PMF is lower than expected. The incidence rates of JAK2V617F mutation in patients diagnosed with PMF were also reported by various authors (Barosi et al., 63.4% of 304 patients; Levine et al., 39% of patients) (25-27). In the present study, the JAK2 mutation was detected in 70.9% of patients with PMF.

Detection of JAK2V617F mutation in 90%–95% of patients with PV ensured its inclusion in WHO 2008 guideline as one of the major criteria for PV (5). A JAK2K539L mutation was detected on

exon 12, which is another JAK2 domain, in most patients with PV where this mutation found to be negative (28). These data indicate that JAK2 mutations have been found in almost all patients with PV (29). The JAK2 mutation was detected in the present study, in accordance with literature findings in 70.6% of patients with PV. Hematocrit values and incidence of splenomegaly were reported in patients with PV having a JAK2V617F mutation and increased WBC counts (30). Treatment with a JAK2 blocker leads to a dramatic resolution of splenomegaly in CMPD, confirming the correlation between this mutation and splenomegaly (31).

CONCLUSIONS

The JAK2 mutation has offered a different perspective in BCR/ABL-negative patients with CMPD and served as an acceptable diagnostic factor. The incidence of this mutation among patients diagnosed with CMPD in the present study was in accordance with literature findings. However, this study had a limited sample size. Hence, large-sample studies should be conducted to confirm the relationship between this mutation and CMPD.

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