Triple-negativity identifies a subgroup of patients with better overall survival in essential thrombocythemia

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Abstract

Essential thrombocythemia as defined by WHO in 2016, is a Philadelphia-negative chronic myeloproliferative neoplasm showing a better prognosis than polycythemia vera and myelofibrosis. In a variable percentage, patients with essential thrombocythemia show none of the known driver-gene mutations that may occur on JAK2, CALR and MPL genes. Such patients are classified as triple-negative and their clinical features and prognosis have not been described with precision yet.
In this study we evaluated some of the characteristics of this population by comparing them with those of patients with driver-gene mutated ET.
Data from 266 consecutive essential thrombocythemia patients were analysed. Triple-negative patients had a significantly lower symptom load and a lower frequency of splenomegaly at diagnosis. The rate of thrombosis resulted to be equal in the two subgroups. Overall survival was slightly better in the triple-negative group of patients.

Introduction

Philadelphia-negative chronic myeloproliferative neoplasms (MPNs) were initially identified in 1951 as myeloproliferative disorders (1), although only in 2000 these diseases were definitely classified as malignant neoplasms, according to the 3rd edition of the International Classification of Diseases for Oncology (2). The V617F mutation of the JAK2 gene was discovered in 2005 (3) and subsequently mutations of the thrombopoetin receptor gene (MPL) (4,5), mutations of the JAK2 exon-12 (6) and, finally, mutations of the calreticulin gene (CALR) (7) were demonstrated to be strictly associated with MPN pathogenesis, respectively in 2006 in 2007 and in 2013. These recent discoveries have given an important impulse to the knowledge of the MPNs molecular mechanisms. In 2008, the World Health Organization (WHO) redefined the myeloproliferative disorders as myeloproliferative neoplasms (8) and in 2016 made a further revision of the case definition for Essential Thrombocythemia (ET), Polycythemia Vera and Myelofibrosis, introducing the distinction between pre-fibrotic myelofibrosis and overt myelofibrosis (9).

Among the three major myeloproliferative diseases classified according to the WHO, ET is the less lethal and more benign, with a 5-year survival rate of 85-91%; it occurs frequently in the female sex (10,11), with a median age at diagnosis of 67 years and in the USA. ET is classified as a rare disease, with an estimated incidence of 1.1-2.0 cases per 100,000 persons per year and a prevalence of 24 -58 per 100,000 persons, in 2016 (12). Some authors have shown a significant reduction in age at diagnosis (13,14). Clinical manifestations in essential thrombocythemia are similar to those seen in other myeloproliferative diseases and sometimes can be particularly severe. Some authors have conducted a survey of 1179 patients and have shown that 70% of patients report constitutional symptoms and splenomegaly-related symptoms. The most frequent symptom is various-grade asthenia, reported by the 81% of the patients (11).

However, the main features of ET are the marked thrombocytosis on the blood count and the increased thrombotic risk. In fact, between 1297 patients with ET, 17.8% have reported a thrombus prior to or at the time of ET diagnosis (15). Thrombotic risk is currently estimated applying the so called IPSET-t score, which provides the distinction into 4 risk categories: very-low, low, intermediate or high risk (16). Acetylsalicylic acid alone is provided for patients in very low or low risk, while cytoreductive therapy (hydroxyurea, busulfan or interferon) is indicated for high risk, and to be taken into consideration, but not mandatory, for intermediate risk patients (17).

As regards the mutational status, the V617F JAK2, CALR and MPL mutations (driver mutations) are harboured by the pathological cell population in the 50-60%, 15-30% and 1-5% of the ET cases, respectively. In the remaining percentage of patients with ET, none of the driver mutations can be found and these patients are thus classified as “triple-negative” (TN). In the past, some authors have highlighted peculiar characteristics in this population (18). Starting from these evidences, we aimed to describe the clinical characteristics of the TN-ET patients followed at our center and to determine whether they have a better prognosis than the driver-mutated.

Patients and Methods

We conducted a retrospective analysis on 266 consecutive ET patients referring to our centre from January 1997 to January 2021. As per definition of triple negativity, in the absence of data for non-driver mutations that may define myeloid clonality, all the included cases of TN-ET were diagnosed requiring the exclusion of secondary causes of thrombocytosis as minor criterion, according to the 2016 WHO diagnostic criteria for myeloid neoplasms (8). The median follow-up time is 51.5 months. For each of these patients the absence of the three driver genes mutations has been determined.

At diagnosis, MPN-10 score (19) was calculated to evaluate the symptoms load, presence of splenomegaly, frequency of venous and arterial thromboses or other cardiovascular events, eventual evolution into myelofibrosis and/or leukemia. The number of deaths and survival data of these patients were recorded, comparing these characteristics in triple negative patients versus patients

with known mutations on the driver genes. Comparison between frequencies was carried out with the chi-square method, comparison between medians with the Kruskal-Wallis test. Survival was calculated with the Kaplan and Meyer method and the comparison between survival data was carried out with the log-rank test.

Results

Of the 266 patients included in the study, 192 harboured the V617F mutation of the JAK2 gene (71.8%), 26 patients carried a pathogenic mutation of CALR (9.77%) and 4 patients were mutated on MPL (1.50 %). On the other hand, 45 patients show none of the above mentioned mutations on the three MPNs driver genes and are thus considered triple-negative MPNs (TN-MPN) (16.92%).
In the examined population, TN-MPN patients have a median age of 52.3 years (range 14.3 - 89.7), while driver-mutated patients are significantly older, with a median age of 66.6 years (range 18.02 - 90.71) ($p = 0.00028$). Regarding gender in TN patients, there is a slight prevalence of the female sex (34/45, 75.7%) between TN patients, while driver-mutated patients are female in 146/221 cases (60.1%). The difference does not achieve strong significance, but is to be considered as a trend p.0.21.
Data from MPN-10 score at diagnosis is available for 148 patients, of which 116 with driver mutations and 32 TN-MPN. The median score is 19 points (range 8 - 23) in patients with mutations and 13 in TN-MPN patients (range 4 - 19) ($p = 0.001$). The frequency of thrombotic and cardiovascular events appears to be similar in the two groups of patients with 12/45 episodes in TN (26.66%) and 59/221 (26.70%) in patients with mutation. At diagnosis, splenomegaly is more frequently described in TN-MPNs (6.7% versus 15.8% in driver-mutated patients) with a p-value of 0.051.
Evolution into myelofibrosis was diagnosed in 5 patients in the observation period, with further evolution into acute leukemia in 2 cases. Of those, only one case of MF evolution occurred in the TN group. Overall, 28 deaths occurred in patients with mutations on the driver genes (12.7%), while only 3 deaths occurred in TN patients (6.7%) ($p = 0.2$). Survival curves are slightly more favourable for the TN-MPN group than driver-mutated ($p = 0.1$), as shown in Figure 1.

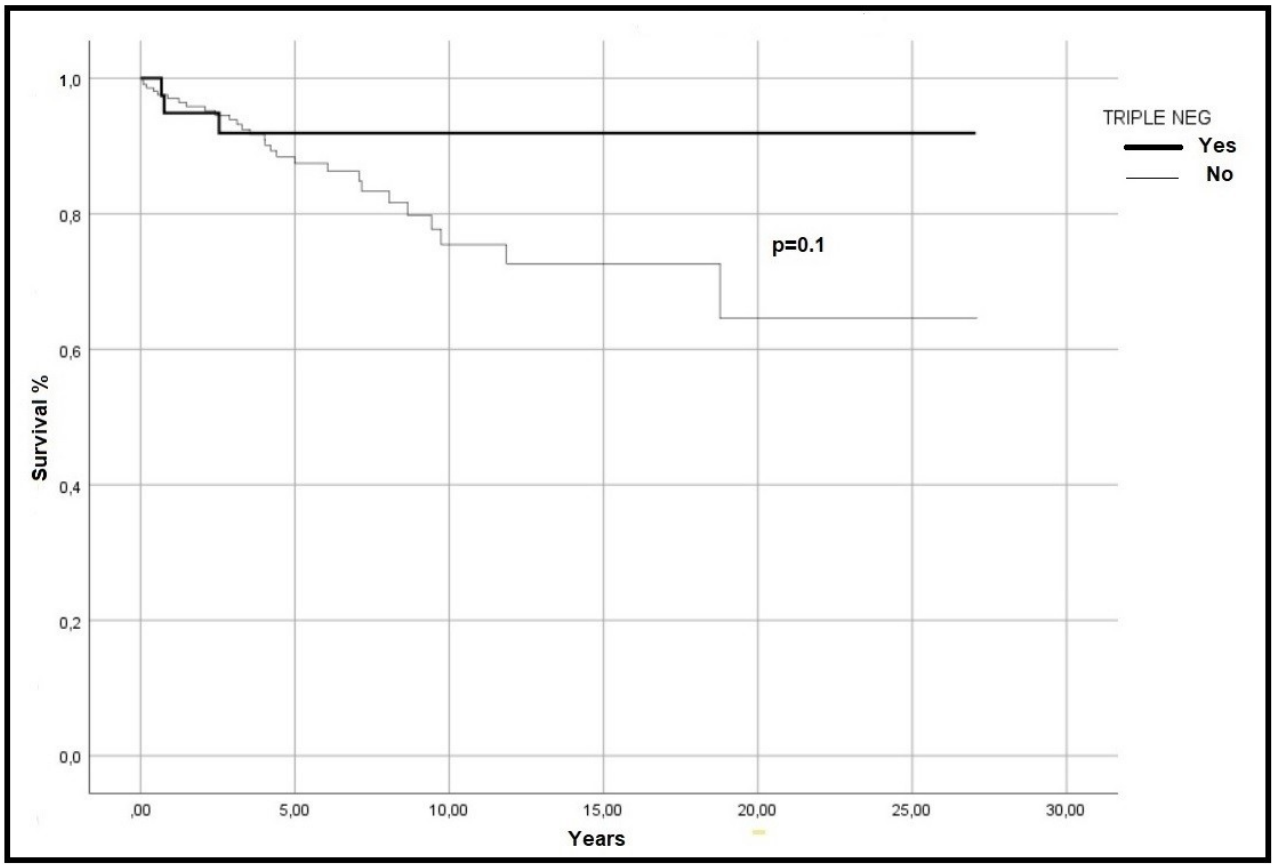

Figure 1. Overall survival in 266 ET patients. The difference between the TN and the driver-gene mutated patients is significant with a p-value of 0.1.

Discussion and Conclusions:

The existing literature reports that the expected survival of ET patient accounts for about 20 years (20). In our experience TN patients show a survival of 24.5 years, higher than that found in patients with mutations affecting one of the three driver genes (21.7 years). Our finding confirms previous experiences that have already reported a slightly better survival for TN ET patients (18, 21). One of the possible reasons that can explain this better outcome is the lower age at diagnosis in TN cases.
Patients with TN ET show a significantly lower symptom load, when it is evaluated by the MPN-10 score, along with a lower frequency of splenomegaly at diagnosis in TN patients. Splenomegaly is present in about 15% in patients with ET and appears to correlate with a worse prognosis (22).
Thromboses occurred without significant differences in triple negative and in driver genes mutated patients in our study, in contrast with literature (21).
A weakness of our study is the lack of information regarding other myeloid-related genes asset, as non-driver mutations were not evaluated in our study. Literature do not strongly report the association between the presence of non-driver mutations hitting genes like TET2, SH2B3, and ASXL1 and thrombotic events or survival. Moreover, some atypical JAK2 CALR and MPL mutations have been identified in some TN patients (23, 24).
Nowadays, there is not much information about the clinical and molecular characteristics of patients with TN-ET and the clinical history of large cohorts of these patients has not been described in detail. Overall, our findings, even within the limits of a mono-institutional study, give hints for a better prognosis in patients with TN-ET.
The hypothesis that TN-ET patients embody a population with better prognosis and different clinical features than driver-gene mutated ET is interesting and may have a clinical impact in the management of these patients, but it has to be supported by large perspective observational studies.

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