Ruxolitinib versus best available therapy for ET intolerant or resistant to hydroxycarbamide in a randomised trial

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Title page

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Short title: Ruxolitinib *vs* BAT in HC resistant/intolerant ET

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Key Points

- Ruxolitinib showed no significant improvement for attainment of either CR or PR over BAT within the first year of therapy in high-risk ET
- Ruxolitinib significantly improved some disease-related symptoms but rates of thrombosis, hemorrhage or transformation were not different

Abstract

Treatments for high-risk essential thrombocythemia (ET) address thrombocytosis, disease-related symptoms, as well as risks of thrombosis, hemorrhage, transformation to myelofibrosis and leukemia. Patients resistant/intolerant to hydroxycarbamide (HC) have a poor outlook. MAJIC (ISRCTN61925716) is a randomized phase II trial of ruxolitinib (JAK1/2 inhibitor) vs Best Available Therapy (BAT) in ET and polycythemia vera (PV) patients resistant or intolerant to HC. Here findings of MAJIC-ET are reported, where the modified intention-to-treat population included 58 & 52 patients randomized to receive ruxolitinib or BAT respectively. There was no evidence of improvement in complete response within 1 year reported in 27 (46.6%) patients treated with ruxolitinib *vs* 23 (44.2%) with BAT (*P*=.40). At 2 years rates of thrombosis, hemorrhage and transformation were not significantly different, however some disease-related symptoms improved in patients receiving ruxolitinib relative to BAT. Molecular responses were uncommon; there were two complete molecular responses (CMR) and one partial molecular response (PMR) in *CALR* positive ruxolitinib-treated patients. Transformation to myelofibrosis occurred in one CMR patient, presumably due to the emergence of a different clone raising questions about the relevance of CMR in ET patients. Grade 3&4 anemia occurred in 19% & 0% of ruxolitinib vs 0% (both grades) BAT arm, grade 3&4 thrombocytopenia in 5.2% & 1.7% of ruxolitinib vs 0% (both grades) of BAT treated patients. Rates of discontinuation or treatment switching did not differ between the two trial arms. The MAJIC-ET trial suggests that ruxolitinib is not superior to current second-line treatments for ET.

Introduction

Essential thrombocythemia (ET) is a chronic myeloproliferative neoplasm (MPN) characterized by thrombocytosis. Patients are at higher risk of thrombosis and hemorrhage. They also have disease-related symptoms, which are difficult to manage with standard therapies. Therapeutic approaches address risks of thrombosis and hemorrhage, without increasing transformation into myelofibrosis (PET-MF) or acute myeloid leukemia (AML) [1-3]. Low-dose aspirin with hydroxycarbamide (HC) is recommended first-line therapy in high-risk patients, supported by data from randomized trials[3,4]. Approximately 20% of ET patients become HC-intolerant or resistant; patients with resistance appear to be at increased risk of disease transformation and reduced overall survival[5]. No prospective trial data exists to guide management of ET patients who are HC-resistant or intolerant; treatment options are limited, and several second-line treatment options are associated with increased risk of disease transformation[2,3,6].

The discovery of the Janus kinase (*JAK2*V617F) mutation provided the first genetic marker of the malignant clone in MPN[7-9]. Furthermore, other key driver mutations associated with ET, affecting thrombopoietin receptor *MPL* and calreticulin (*CALR*) also lead to increased JAK2 signaling[14]. The JAK1/2 inhibitor, ruxolitinib, was effective in reducing spleen volume, controlling blood counts and improving symptoms in MF and PV patients[15-17]. Ruxolitinib treatment may also result in a survival advantage for patients with MF[18,19]. A previous non-randomized study in 39 ET patients, resistant or intolerant to HC, demonstrated that ruxolitinib lowered both platelet and white cell counts and the most effective starting dose was 25mg bd[20].

We conducted a randomized, phase II trial to evaluate the activity and safety of ruxolitinib *vs* Best Available Therapy (BAT) in two different patient populations (ET and PV): A randomized study of best Available therapy versus JAK inhibition in patients with high-risk PV or ET who are resistant or intolerant to HydroxyCabamide (MAJIC). The study utilized an efficient framework of a basket trial design, permitting the separate evaluation of two study populations. Here we present safety and efficacy data for the ET population, so-called MAJIC-ET.

Patients and Methods

Trial design

An independent, parallel, open-label, randomized controlled trial of ruxolitinib *vs* BAT was implemented (Supplemental Figure S1). Patients aged ≥18 with high-risk ET or PV, who met modified criteria for intolerance or resistance to HC[21] (Supplemental Table S1), were recruited. MAJIC-PV arm is on-going. High-risk ET was defined by standard criteria (Supplemental Table S2). Patients were stratified by *JAK2*V617F status and randomized 1:1 to receive either ruxolitinib (starting dose 25mg twice daily (bd) or 20mg bd, if baseline platelets were 100-200×10^9/L) or BAT. Inclusion and exclusion criteria are presented (Supplemental Table S3). The trial was registered at www.isrctn.com (ISRCTN61925716) and reviewed by an independent research ethics committee. All participants gave written informed consent in accordance with the Declaration of Helsinki. Trial data was analyzed by statisticians at the Cancer Research UK Clinical Trials Unit (University of Birmingham) and Quality of Life (QoL) analysis performed by statisticians at the Mayo Clinic, Scottsdale, Phoenix. Ruxolitinib was provided free of charge by Novartis. All authors had access to primary clinical trials data and approved the final version of manuscript.

Outcome measures

The primary outcome measure was achievement of Complete Response (CR) as defined by European Leukemia Net (ELN) criteria within 1 year of treatment[22]. CR in ET patients was defined by achieving all of the following criteria: platelet count ≤400×10^9/L; normal spleen size on imaging; white blood cell count ≤10×10^9/L. Secondary outcomes included Partial Response (PR) per ELN criteria within 1 year of treatment, duration of response (both CR and PR) and overall response (i.e. CR&PR), toxicity profile of ruxolitinib based on National Cancer Institute Common Terminology Criteria for Adverse Events (NCI CTCAE) version 4, dose intensity, histological response, molecular response; hemorrhagic and thromboembolic events, disease transformations, QoL and disease symptom burden, overall and progression free survival. The safety population included all patients who received at least one dose of protocol treatment. Hemorrhagic and thrombotic events were centrally reviewed. QoL and symptom assessment questionnaires: 10-item Myeloproliferative Neoplasm Symptom Assessment Form (MPN-10) Total Symptom Score (TSS)[23], EQ-5D[24] and M.D. Anderson Symptom Inventory (MDASI)[25] were completed at baseline (pre-treatment, 7 consecutive days for the MPN-10 and once for the other questionnaires), 2 and 4 months post randomization and continued 4 monthly whilst on trial. Overall symptom response was defined as at least a 50% reduction in TSS from baseline (average of the 7 baseline days with at least 4 of 7 days scored) at any post-baseline time point up to Month 12.

Sample size justification and statistical analysis

Sample size calculations were based upon rates from a previous phase II study[20] using an one-sided normal test without continuity correction and unpooled variance. CR rate for controls was estimated at 30%. A clinically significant improvement was considered to be 20%. Thus, assuming CR rates in the control and treatment group were 30% and 50%, 55 patients were required in each arm to detect a clinically significant difference of 20% with 82% statistical power at 10% level of significance. As this is a randomized screening trial to evaluate a direct, but nondefinitive comparison, with the aim of screening for promising signal of activity in ruxolitinib, a relaxed one-sided significance level of 10% was utilized[26]. Allowing for a 5% drop out rate, 116 patients were required.

P<.10 was considered significant for the primary outcome. For other analyses, two-sided tests were used and a *P*<.05 was considered significant. Number and proportion of patients reported for categorical variables by treatment group and overall. Descriptive statistics (number of patients, mean, standard deviation (SD), median, Interquartile Range (IQR)) reported for continuous variables by treatment group and overall. Time-to-event outcomes analyzed using the method of Kaplan and Meier and differences in survival time determined using a Cox's model with adjustment for *JAK2*V617F status per baseline data. Sensitivity analysis adjusting the Cox models for hemoglobin and disease duration were performed and reported where treatment effect differed. Normal Z-tests were used to assess difference in proportions. Univariate and multivariate logistic regression were fitted models to assess effect of baseline measures on primary outcome, transformations and toxicity. Apart from the primary outcome, additional hypotheses testing were exploratory and non pre-specified. All summaries and statistical analyses for efficacy were primarily carried out on a modified intention to treat (mITT) basis, including patients analyzed according to their randomized treatment allocation, starting treatment within one year of randomization and with at least one response. Summary statistics for safety variables were based on the safety population, which included patients according to the treatment they actually received and who received >1 doses of treatment. Statistical analyses were performed using Stata version 14.2.

QoL and symptom data were analyzed in mITT population using SAS version 4. Symptom response rate was compared using chi-squared test, maximum percentage reduction from baseline during the first 12 months using a Wilcoxon rank-sum test. Symptom response and percentage reduction at post-baseline time points (or at most recent assessment if no symptom data at the given time point were provided) were compared using chi-squared and Wilcoxon rank-sum tests. Comparisons of mean scores longitudinally employed a linear mixed model for each outcome (from month 2 assessment) using all available data. In addition each model included a continuous covariate for baseline value of outcome and used planned month of assessment as categorical time value with compound covariance structure.

Treatment and assessments

Ruxolitinib was initiated based on baseline platelet count. BAT was assigned according to physician's choice but had to be an active agent, change of and combination of BAT therapies was permitted with the aim of achieving a CR. No crossover of BAT to ruxolitinib was permitted. Low-dose aspirin

(75mg od) was advised unless contraindicated. Protocol-specified dose reductions for ruxolitinib were in place and patients allowed to re-escalate if toxicity had resolved. Lowest permitted dose of ruxolitinib was 5mg once-daily. Hematological response was assessed 2 weekly for 3 months, then 6 weekly in order to determine the primary outcome of CR during year 1 (cut-off week 54). Ultrasound was performed at baseline and centrally reviewed. If splenomegaly was present at baseline repeat ultrasound showing resolution was required for CR. Ruxolitinib continued beyond 1 year if CR or PR were maintained. Those discontinuing ruxolitinib moved to BAT arm for follow-up. Patients who transformed to PET-MF, myelodysplastic syndrome (MDS) or AML discontinued the trial but were followed for survival.

Assays for *JAK2*V617F, *CALR* and *MPL* mutation allele burden was quantified using next generation sequencing as previously described[27]. An analysis of histological features is currently being performed and this data is not being presented as part of this manuscript.

Results

Patient characteristics

116 patients were recruited in 31 UK centers between September 2012-February 2015, median follow up of 2.61 years (range: 0.23 – 4.12). In total 110 were eligible for the mITT analysis, comprising 58 (52%) and 52 (48%) patients in the ruxolitinib and BAT arms respectively. Median age of patients was 64.2 years with 44 (40%) males and 66 (60%) female patients, overall 28/110 (25.4%) were resistant to HC, 57/110 (51.8%) intolerant or 25/110 (22.7%) both. Baseline characteristics were balanced (Table 1), except for the ruxolitinib arm with longer disease-duration and lower hemoglobin. Six patients were excluded from mITT analysis: 4 withdrew without treatment (2 did not wish to be on BAT arm, one ineligible, one had transformed to PET-MF) and 2 did not start treatment within one year from randomization. All CALR indels and MPL mutations are provided in Supplemental Table S4.

Trial Treatment

For patients receiving ruxolitinib, the mean dose intensity of ruxolitinib during year one was 19mg bd (Figure 1). The most common BAT therapies utilized at least once included HC in 37/52 (71.1%), anagrelide in 25/52 (48.1%) and interferon in 21/52 (40.4%) patients.

Patient disposition

Patient disposition at the time of analysis (2 year follow up) is shown in Figure 2. Thirty BAT patients (57.7%) switched their initially assigned therapy at least once and there were 86 switches across the BAT group. In total, 45 patients (49.5%) discontinued treatment, with 40 discontinuations occurring within the first treatment year. Thirty-five patients (60.3%) receiving ruxolitinib and 10 patients (19.2%) receiving BAT discontinued treatment. The main reasons for discontinuation in the ruxolitinib arm were loss of response (11/35 (31.4%)) and transformation (9/35 (25.7%)). The main reasons for discontinuation in the BAT arm were transformation (3/10 (30%]) and death (2/10 (20%)). Discontinuations and therapy switches are shown in Table 2.

Efficacy analysis

For patients meeting the criteria for mITT analysis the primary outcome (CR) was achieved in 27 (46.5%) of the patients in the ruxolitinib arm *vs* 23 (44.2%) in the BAT arm (Unadjusted *P*=.40, adjusted for *JAK2*V617F status *P*=.40) with a difference of proportions -2.3% between BAT and ruxolitinb (80% CI: -15%, 10%). PR occurred in 27 (46.5%) patients in the ruxolitinib arm and 27 (51.9%) patients in the BAT arm. Time to first response (CR or PR) between the two arms was significantly different (*P*=.01) with BAT patients taking longer. Duration of CR appeared shorter for ruxolitinib patients (borderline significant difference, *P*=.05; adjustment for hemoglobin and disease duration rendered this insignificant, *P*=.2). There was no evidence of a difference in duration of overall response between ruxolitinib and BAT. Overall survival (OS) and progression free survival (PFS) at 1 year were similar (OS: .98 (95%CI .86, .99) for BAT and .98 (95%CI 0.88, 0.99) for ruxolitinib patients, PFS: .96 (95%CI .85, .99) for BAT and .93 (95%CI .81, .97) for ruxolitinib patients). In multivariable analyses performed to assess baseline factors influencing CR (modelled for: treatment received, HC resistance/intolerance, white cell count, platelets, hemoglobin & *JAK2/CALR* status) no factor was shown to be significant and did not change the treatment effect (Supplemental Table S5).

Thrombosis, hemorrhage and disease transformation

After 2 years of follow up transformation to PET-MF occurred in 8 ruxolitinib vs. 5 BAT treated patients. Transformation to AML was seen in 1 patient who received ruxolitinib. Transformation-free probability was not significantly different between the two arms (*P*=.29; Supplemental Figure S2A). Concerning thrombosis and hemorrhage, following central review, 10 patients (17.2%) on the ruxolitinib arm experienced 11 thrombotic events compared to 3 patients (5.8%) on the BAT arm experiencing 5 events. Hemorrhagic events were 1 (1.7%) *vs* 5 (8.9%) for ruxolitinib and BAT patients respectively (Table 3). Concerning thrombosis-free probability, the differences were borderline but not statistically significant (*P* =.09; Supplemental Figure S2B). Hemorrhage was less frequent for patients treated with ruxolitinib, however this difference was also not significant (*P* =.14; Supplemental Figure S2C). Since all of these events are considered clinically relevant we performed an analysis of transformation, thrombosis and hemorrhage as a composite endpoint; there was no evidence of a difference (*P* =.35; Supplemental Figure S2D). Most thrombotic and hemorrhagic events occurred in patients in CR or PR (Supplemental Figure S3). In a multivariate analysis of factors influencing transformation to PET-MF, this event only occurred in patients with baseline WBC <10×10^9/L (Supplemental Table S6).

Molecular responses (MR)

The mean baseline allele burdens for *JAK2*V617F, *CALR* or *MPL* mutation positive patients are displayed in Table 1. At 12 months, or the last available sample during year 1, the overall mean allele burden had not changed significantly for any mutation in either treatment arm. However, 1 complete molecular response (CMR) and 1 partial molecular response (PMR) per ELN criteria were seen for *JAK2*V617F positive patients on the ruxolitinib arm and 2 CMRs and 1 PMR for *CALR* positive patients on ruxolitinib compared to 0 CMRs/PMRs for patients with these mutations receiving BAT. A *JAK2*V617F positive patient who achieved a PMR on ruxolitinib also had resolution of a cytogenetic abnormality at one year. There was no pattern of MR or progression with complete or partial hematological response or transformation, but 1 *CALR* positive patient who transformed to PET-MF had a CMR.

Impact on ET Related Disease Symptom Burden

Among 110 patients in the mITT cohort, 85 completed the baseline and at least one post-baseline questionnaire (ruxolitinib N=47, BAT N=38). While overall symptom response rate during the first 12 months did not significantly differ between arms (ruxolitinib 12/42 (29%) vs BAT 6/31 (19%), *P*=.37), maximum percentage TSS reduction at any point during the first 12 months of treatment was significantly greater for ruxolitinib compared to BAT (median reduction 32% vs 0%, *P*=.03, Figure 3A). Symptom response was rapid in the ruxolitinib arm (8/42 (19%) at 2 months) as compared to BAT (1/31 (3%) at 2 months, *P* =.04). Longitudinally, mean TSS (*P*=.03) and the individual symptom of pruritus (*P*=.01) were significantly lower for ruxolitinib vs BAT (Figure 3B and 3C), with trends observed for improved concentration (*P*=.05), lower anxiety/depression (EQ5D *P*=.09), and higher ability to perform usual activities (EQ5D *P*=.09) on the ruxolitinib arm compared to BAT.

Safety

All safety analysis was conducted on the safety population: 115 patients (57 BAT, 58 ruxolitinib). A total of 128 Grade 3/4 events occurred in 89 patients on the trial (Supplemental Table S7). Hematological toxicities (36/128) and metabolism/nutrition disorders (17/128 – 10 relating to hyponatremia) were the most common. Grade 3 or 4 anemia occurred in 12/58 (21%) of ruxolitinib patients vs 0/57 (0%) in the BAT patients (*P*<.005), grade 3 or 4 thrombocytopenia in 2/58 (3.4%) of ruxolitinib vs 0/57 (0%) of BAT patients (*P*=.32), and grade 3 (only) infections occurred in 9/58 (15.5%) of patients in the ruxolitinib arm compared to 2/57 (3.5%) (grade 3 and 4) in the BAT arm (*P*=.03). Overall 2 patients discontinued ruxolitinib for anemia; there were no discontinuations related to thrombocytopenia. Blood counts during the trial according to treatment arm are shown in

Supplemental Figure S4, demonstrating equivalent control of leucocytes and platelets but lower hemoglobin from week 4. An unplanned multivariate model (modelled for hemoglobin (≥ 100g/dl) and *JAK2/CALR* status) demonstrated that baseline hemoglobin (≥100g/dl) was significant in predicting the occurrence of anemia or thrombocytopenia (OR=.17, 95% CI=.04, .72, *P*=.01) (Supplemental Table S8).

There were 5 patient deaths in the ruxolitinib arm and 2 in the BAT arm, none were considered treatment related. The deaths in the BAT arm were due to multiple organ failure and cerebral hemorrhage. In the ruxolitinib arm, deaths were due to carcinomatosis combined with esophageal cancer, bowel infarction due to adhesions, acute left ventricular failure, ischemic cardiomyopathy and sepsis combined with pancreatic cancer.

Discussion

ET is often regarded as the most indolent of the Philadelphia negative MPNs, treatments offer improvements in blood counts and reduction in risk of thrombosis and hemorrhage with a lack of certainty regarding effects upon transformation to PET-MF and AML[2,3]. Criteria for resistance or intolerance to therapy with HC were originally developed to guide clinicians when to initiate second-line therapies; however, there is now evidence that HC resistant patients have a poor outlook[28]. In addition, disease-related symptom burden is increasingly recognized as an important disease feature, causing significant morbidity with few effective treatments[2-4]. In previous studies patients with MF gained a survival benefit with ruxolitinib, which also reduced spleen size and symptoms compared to BAT[18]. In the RESPONSE study in HC resistant/intolerant PV patients there was a suggestion of lower rates of thrombosis in patients receiving ruxolitinib compared with BAT, as well as better control of blood counts, spleen size and symptoms[17].

The MAJIC trial was designed to compare ruxolitinib with BAT in patients with HC intolerance/ resistance in two populations, MAJIC-ET and MAJIC-PV. Both trial populations are fully recruited and here we report the findings of MAJIC-ET trial. The patients recruited into MAJIC-ET displayed characteristics that were well-balanced between the two arms with the exception of baseline hemoglobin and prior disease duration. Distribution of driver mutations *JAK2*V617F, *MPL* exon 10, *CALR* mutations were as expected. Our patients had a long disease duration (up to 31 years) some of whom had received multiple therapy lines with up to nine prior therapies. Some features of advanced disease, for example splenomegaly and leukocytosis, were present at baseline however transformation to PET-MF was excluded at trial entry. Diagnostic criteria have been controversial in ET and those in use at trial centers were: British Committee for Standards in Hematology (BCSH n=18); WHO 2001/2008 (n=10) and both combined (n=3). The BCSH and WHO criteria were recently shown to perform equally well[29].

Usual therapy choice in the second-line setting for ET would be anagrelide or interferon, however in order to perform a "real-life comparison" we allowed investigator choice. Overall the majority, 79% (41/52), of BAT patients received one or both agents before or during the study. On-study BAT included in addition busulfan ^{32}P and HC; several international guidelines recommend busulfan or ^{32}P for older patients.

Proportions of patients reaching CR within one year were similar: 27 (46.5%) in the ruxolitinib arm *vs* 23 (44.2%) for BAT, with similar PR rates. Time to any first response (CR or PR) was significantly faster for patients treated with ruxolitinib (*P*=.01). A particularly interesting finding, as patients in CR who were randomized to receive ruxolitinib had to change therapy and potentially lost any pre-existing response yet managed to attain CR faster than BAT patients who may not have changed therapy thus only needing to maintain response. In addition, BAT patients were also allowed to combine or to switch therapies and frequently did so. Importantly the duration of CR appeared shorter for ruxolitinib patients with a marginally significant value, while the duration of overall response (CR and PR) was not different between both arms. We confirm that HC resistant/intolerant ET patients have a high-risk of thrombosis, hemorrhage and transformation to PET-MF; event rates here being higher than reported in the non-resistant/intolerant patients e.g. PT-1 or ANAHYDRET studies[30,31]. However, overall thrombosis, hemorrhage or transformation considered separately or together as a composite endpoint were not statistically different between the ruxolitinib and BAT. Furthermore, in a *post hoc* unplanned analysis for factor influencing transformation to PET-MF, only a leucocyte count $<10 \times 10^9$/L was significant.

Studies have reported that post-randomization exclusions of patients in randomized trials may affect trial results[32], with some raising concerns that the investigated therapy might be favored[33,34]. However, in the MAJIC-ET trial, if we were to conduct a pure intention-to-treat (ITT) analysis, this would require imputation of missing response data for 6 BAT patients. Missing data imputation may bias estimates of treatment effects[35]. A commonly used technique is nonresponder imputation, which will attribute all 6 BAT patients as not achieving CR within a year. This will result in a less conservative ITT analysis of 23/58 CR (BAT) vs 27/58 CR, with p-value of .22 compared to the mITT analysis (p=.4). Our primary findings of no evidence of superiority of ruxolitinib were however consistent using either mITT or ITT analysis.

Molecular responses were uncommon in the first year of the trial, as described previously[20]. However, ruxolitinib was associated with two CMR and one PMR in a *CALR* positive patient; this has not previously been reported. Transformation to PET-MF in one *CALR* positive patient, who achieved a CMR, presumably occurred due to the emergence of a different clone, consistent with patients reported with *JAK2*V617F positive chronic phase developing *JAK2*V617F negative AML[36], and raises questions about the relevance and value of CMR in patients with ET.

Patterns of adverse events with ruxolitinib were similar to those already reported, most prevalent events related to hematological, nutritional and metabolic events. Infections were also more common with ruxolitinib therapy. There was no suggestion of imbalance between the two arms of MAJIC-ET for non-melanoma skin cancer as was previously noted in the RESPONSE trial[17]. Treatment discontinuation occurred more frequently for patients treated with ruxolitinib, with 35 patients discontinuing treatment compared to 9 discontinuations in the BAT arm. However, 30 BAT patients switched their initially assigned BAT treatment for various reasons, which indicates a similar rate of treatment ineffectiveness or intolerance. For the first time, we show baseline anemia predicted for treatment emergent anemia and thrombocytopenia.

Patients with ET have a high burden of symptoms, which have been consistently reported to affect their quality of life[37]. The symptom response rate, defined as a 50% reduction in TSS, during the first 12 months did not significantly differ between the two arms. However, maximum percentage TSS reduction during the first 12 months of treatment was significantly greater for ruxolitinib compared to BAT and was more rapid in the ruxolitinib arm. Longitudinally, mean TSS and individual symptom of pruritus were significantly lower for ruxolitinib, with trends observed for improved concentration, lower anxiety/depression and higher ability to perform usual activities for ruxolitinib arm compared to BAT indicating a novel and important benefit to ET patients of ruxolitinib therapy.

Limitations of our trial include that the trial reflected “real life practice” in use of diagnostic criteria and selection of BAT therapies. The majority of our centers used either BCSH (n=18) or WHO (n=10) or both (n=3) diagnostic criteria thus perhaps illustrating non-standardized diagnostic processes, however this is a second line study and BCSH/WHO criteria both perform equally well[29] in addition transformation was excluded at study entry. Guidelines recommend anagrelide or IFN as second line therapy for ET, many BAT patients had already been treated with these drugs (25 received interferon, 7 anagrelide and 7 both agents) before study entry, and overall 79% received them before or during the study. The use of HC as a BAT and frequent switching of BAT therapies in 30 BAT patients also reflect real-life constraints and limited treatment options for ET patients with resistance/intolerance to HC and highlight the need for newer therapies in this field.

In conclusion, the MAJIC-ET trial suggests that ruxolitinib does not have improved treatment efficacy compared to BAT for most clinically relevant events. Symptom responses were superior with ruxolitinib therapy but there was no difference in this study for control of blood counts or other relevant endpoints such as transformation, thrombosis or hemorrhage.

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Authorship Contributions

CH, AJM, MFM and CY designed the MAJIC trial. CH, SF, CY and AH wrote and reviewed the protocol. CY, AP, AH and CH devised the statistical analysis plan. JE, MW, FC, JC, NP, SK and SA recruited patients. AP conducted the statistical analysis, with statistical support from CY and AH. AD, RS and RM conducted the QoL analyses. CH, AM, AP, CY, AH, MFM, RM and AD interpreted the results. CH, SJF, AP, EG and CY drafted the manuscript and all authors reviewed the final version. CH is the guarantor.

Conflict of Interest Disclosures

CH has participated in advisory boards for Novartis, CTI, Baxaltra and Celgene; speakers bureau for Novartis, CTI, Baxaltra, Shire, Gilead, INCYTE; received honoraria from Novartis, Shire CTI, Gilead, Baxaltra, INCYTE and received research funding and travel, accommodation and expenses from Novartis. AM has participated in advisory boards for Novartis, CTI and Baxaltra; received honoraria from Novartis, Gilead, Shire and Baxaltra and also received research funding and travel, accommodation and expenses from Novartis. FC and JC have received travel, accommodation and expenses from Novartis. SK has participated in advisory boards for Novartis; received honoraria from Novartis, Shire and Gilead and received travel accommodation and expense from Novartis and Celgene. SA received honoraria from Novartis and participated in advisory boards for Novartis. AH has participated in advisory boards for Novartis and received speakers bureau from Gilead. NCPC has participated in advisory boards for Novartis, received honoraria from Novartis and research support from Novartis. RM has consulted for Novartis, Ariad, and Galena; received research funding from Incyte, Gilead, CTI, NS Pharma, Celgene, and Promedior. MFM has participated in advisory boards for Novartis and Gilead; received honoraria from Novartis, Shire and Celgene and received travel, accommodation and expenses from Novartis. The remaining authors declare no competing financial interests.

References

1. Cortelazzo S, Viero P, Finazzi G, D'Emilio A, Rodeghiero F, Barbui T. Incidence and risk factors for thrombotic complications in a historical cohort of 100 patients with essential thrombocythemia. *Journal of clinical oncology : official journal of the American Society of Clinical Oncology*. 1990;8(3):556-562.

2. Alimam S, Wilkins BS, Harrison CN. How we diagnose and treat essential thrombocythaemia. *Br J Haematol*. 2015;171(3):306-321.

3. Rumi E, Cazzola M. How we treat essential thrombocythemia. *Blood*. 2016.

4. Barbui T, Barosi G, Birgegard G, et al. Philadelphia-negative classical myeloproliferative neoplasms: critical concepts and management recommendations from European LeukemiaNet. *Journal of clinical oncology : official journal of the American Society of Clinical Oncology*. 2011;29(6):761-770.

5. Hernandez-Boluda JC, Alvarez-Larran A, Gomez M, et al. Clinical evaluation of the European LeukaemiaNet criteria for clinicohaematological response and resistance/intolerance to hydroxycarbamide in essential thrombocythaemia. *Br J Haematol*. 2011;152(1):81-88.

6. Sever M, Newberry KJ, Verstovsek S. Therapeutic options for patients with polycythemia vera and essential thrombocythemia refractory/resistant to hydroxyurea. *Leuk Lymphoma*. 2014;55(12):2685-2690.

7. James C, Ugo V, Le Couedic JP, et al. A unique clonal JAK2 mutation leading to constitutive signalling causes polycythaemia vera. *Nature*. 2005;434(7037):1144-1148.

8. Baxter EJ, Scott LM, Campbell PJ, et al. Acquired mutation of the tyrosine kinase JAK2 in human myeloproliferative disorders. *Lancet*. 2005;365(9464):1054-1061.

9. Levine RL, Wadleigh M, Cools J, et al. Activating mutation in the tyrosine kinase JAK2 in polycythemia vera, essential thrombocythemia, and myeloid metaplasia with myelofibrosis. *Cancer cell*. 2005;7(4):387-397.

10. Cazzola M, Kralovics R. From Janus kinase 2 to calreticulin: the clinically relevant genomic landscape of myeloproliferative neoplasms. *Blood*. 2014;123(24):3714-3719.

11. Dahabreh IJ, Zoi K, Giannouli S, Zoi C, Loukopoulos D, Voulgarelis M. Is JAK2 V617F mutation more than a diagnostic index? A meta-analysis of clinical outcomes in essential thrombocythemia. *Leuk Res*. 2009;33(1):67-73.

12. Carobbio A, Finazzi G, Antonioli E, et al. JAK2V617F allele burden and thrombosis: a direct comparison in essential thrombocythemia and polycythemia vera. *Exp Hematol*. 2009;37(9):1016-1021.

13. De Stefano V, Za T, Rossi E, et al. Increased risk of recurrent thrombosis in patients with essential thrombocythemia carrying the homozygous JAK2 V617F mutation. *Annals of Hematology*. 2010;89(2):141-146.

14. Araki M, Yang Y, Masubuchi N, et al. Activation of the thrombopoietin receptor by mutant calreticulin in CALR-mutant myeloproliferative neoplasms. *Blood*. 2016;127(10):1307-1316.

15. Verstovsek S, Mesa RA, Gotlib J, et al. A double-blind, placebo-controlled trial of ruxolitinib for myelofibrosis. *N Engl J Med*. 2012;366(9):799-807.

16. Harrison C, Kiladjian JJ, Al-Ali HK, et al. JAK inhibition with ruxolitinib versus best available therapy for myelofibrosis. *The New England journal of medicine*. 2012;366(9):787-798.

17. Vannucchi AM, Kiladjian JJ, Griesshammer M, et al. Ruxolitinib versus standard therapy for the treatment of polycythemia vera. *N Engl J Med*. 2015;372(5):426-435.

18. Harrison CN, Vannucchi AM, Kiladjian JJ, et al. Long-term findings from COMFORT-II, a phase 3 study of ruxolitinib vs best available therapy for myelofibrosis. *Leukemia*. 2017;31(3):775.

19. Verstovsek S, Mesa RA, Gotlib J, et al. Long-term treatment with ruxolitinib for patients with myelofibrosis: 5-year update from the randomized, double-blind, placebo-controlled, phase 3 COMFORT-I trial. *J Hematol Oncol*. 2017;10(1):55.

20. Verstovsek S, Passamonti F, Rambaldi A, et al. Long-Term Results from a Phase II Open-Label Study of Ruxolitinib in Patients with Essential Thrombocythemia Refractory to or Intolerant of Hydroxyurea. *Blood*. 2014;124(21):1847-1847.

21. Barosi G, Besses C, Birgegard G, et al. A unified definition of clinical resistance/intolerance to hydroxyurea in essential thrombocythemia: results of a consensus process by an international working group. *Leukemia*. 2007;21(2):277-280.
22. Barosi G, Birgegard G, Finazzi G, et al. Response criteria for essential thrombocythemia and polycythemia vera: result of a European LeukemiaNet consensus conference. *Blood*. 2009;113(20):4829-4833.
23. Scherber R, Dueck A, Kiladjian J-J, et al. The Myleloproliferative Neoplasm Symptom Assessment Form (MPN-SAF) Derived Total Symptom Score (TSS): An International Trial of 1433 Patients with Myeloproliferative Neoplasms (MPNs). *ASH Annual Meeting Abstracts*. 2011;118(21):3839-.
24. EuroQol G. EuroQol--a new facility for the measurement of health-related quality of life. *Health Policy*. 1990;16(3):199-208.
25. Cleeland CS, Mendoza TR, Wang XS, et al. Assessing symptom distress in cancer patients: the M.D. Anderson Symptom Inventory. *Cancer*. 2000;89(7):1634-1646.
26. Rubinstein LV, Korn EL, Freidlin B, Hunsberger S, Ivy SP, Smith MA. Design issues of randomized phase II trials and a proposal for phase II screening trials. *J Clin Oncol*. 2005;23(28):7199-7206.
27. Jones AV, Ward D, Lyon M, et al. Evaluation of methods to detect CALR mutations in myeloproliferative neoplasms. *Leuk Res*. 2015;39(1):82-87.
28. Hernandez-Boluda JC, Alvarez-Larran A, Gomez M, et al. Clinical evaluation of the European LeukaemiaNet criteria for clinicohaematological response and resistance/intolerance to hydroxycarbamide in essential thrombocythaemia. *British journal of haematology*. 2011;152(1):81-88.
29. Gisslinger H, Jeryczynski G, Gisslinger B, et al. Clinical impact of bone marrow morphology for the diagnosis of essential thrombocythemia: comparison between the BCSH and the WHO criteria. *Leukemia*. 2017;31(3):774-775.
30. Harrison CN, Campbell PJ, Buck G, et al. Hydroxyurea compared with anagrelide in high-risk essential thrombocythemia. *The New England journal of medicine*. 2005;353(1):33-45.
31. Gisslinger H, Gotic M, Holowiecki J, et al. Anagrelide compared to hydroxyurea in WHO-classified essential thrombocythemia: the ANAHYDRET Study, a randomized controlled trial. *Blood*. 2013.
32. Montedori A, Bonacini MI, Casazza G, et al. Modified versus standard intention-to-treat reporting: are there differences in methodological quality, sponsorship, and findings in randomized trials? A cross-sectional study. *Trials*. 2011;12:58.
33. Melander H, Ahlqvist-Rastad J, Meijer G, Beermann B. Evidence b(i)ased medicine--selective reporting from studies sponsored by pharmaceutical industry: review of studies in new drug applications. *BMJ*. 2003;326(7400):1171-1173.
34. Tierney JF, Stewart LA. Investigating patient exclusion bias in meta-analysis. *Int J Epidemiol*. 2005;34(1):79-87.
35. EMA. Guideline on missing data in confrmatory clinical trials. In: (EMA) EMA ed. London; 2010.
36. Campbell PJ, Baxter EJ, Beer PA, et al. Mutation of JAK2 in the myeloproliferative disorders: timing, clonality studies, cytogenetic associations, and role in leukemic transformation. *Blood*. 2006;108(10):3548-3555.
37. Mesa R, Miller CB, Thyne M, et al. Myeloproliferative neoplasms (MPNs) have a significant impact on patients' overall health and productivity: the MPN Landmark survey. *BMC Cancer*. 2016;16:167.

Figures

Figure 1. Dose of Ruxolitinib received throughout study

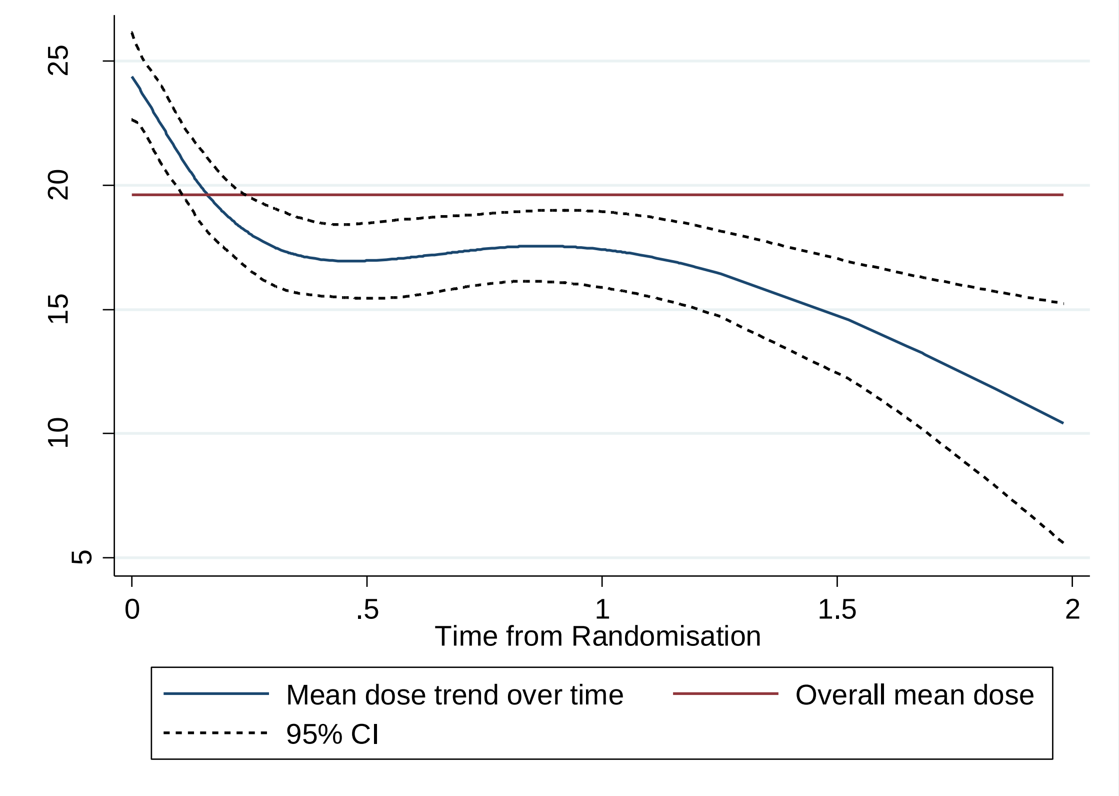
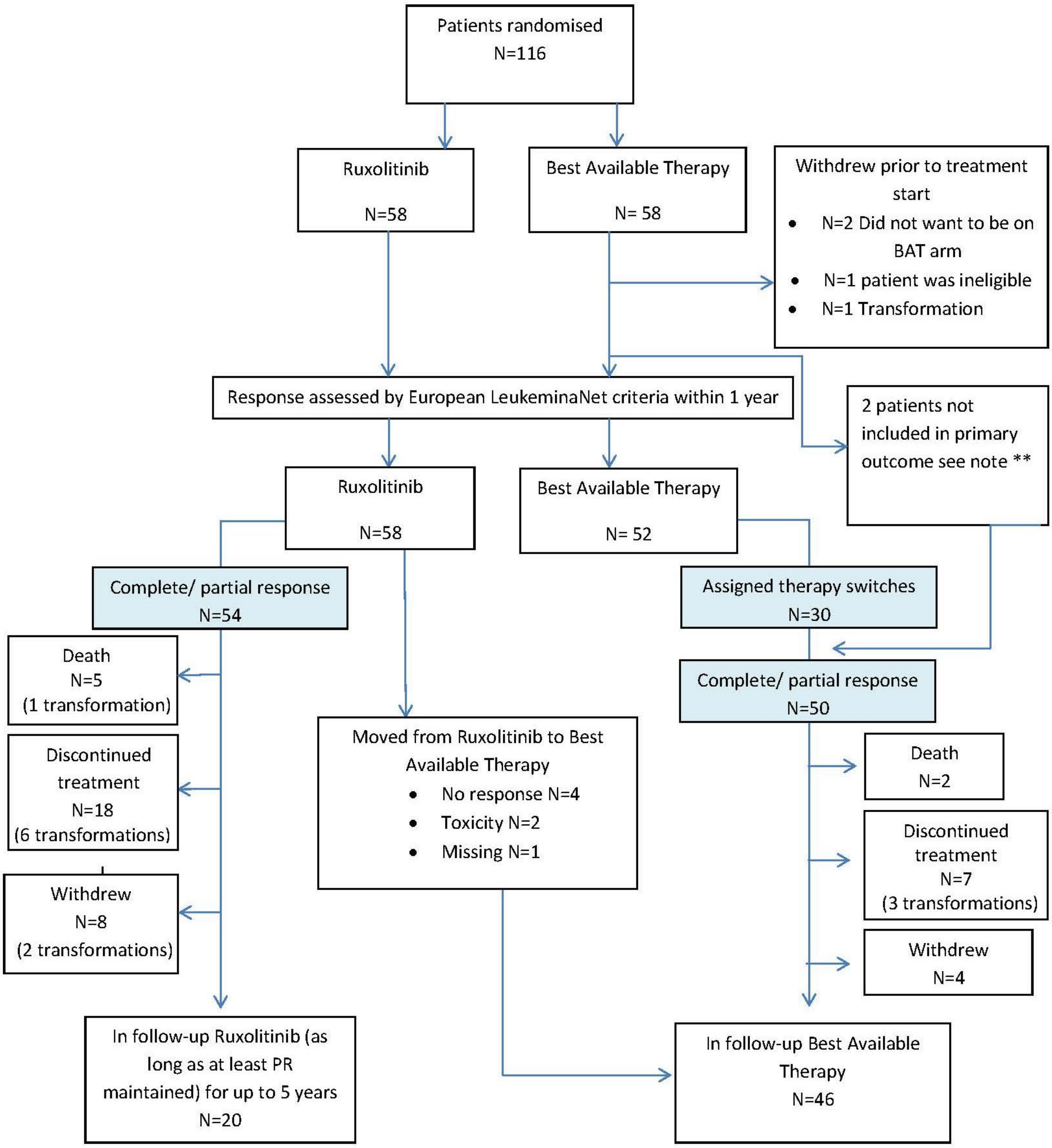


Figure 2. Trial consort diagram at 2 year follow up



Death was recorded as such regardless of prior discontinuation or withdrawal. Patients were clustered as withdrawals regardless of prior discontinuations.

**Note: 1 BAT patient started treatment 1 year after randomisation. 1 other BAT patient did not have a recorded response within 1 year. Both have been excluded from primary analysis, but included in follow-up.

Figure 3. Changes in ET Related Symptom Burden during year 1 of the MAJIC-ET trial

Figure 3A.

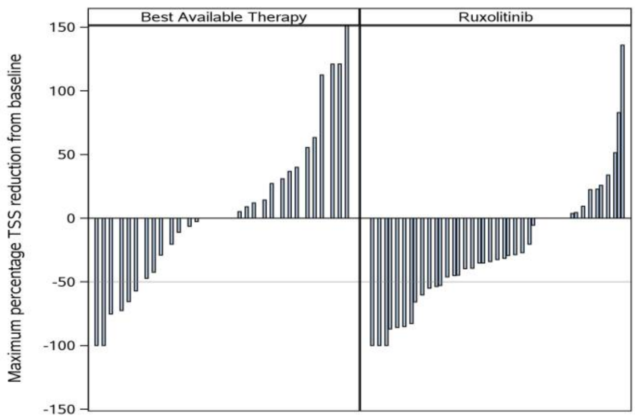


Figure 3B.

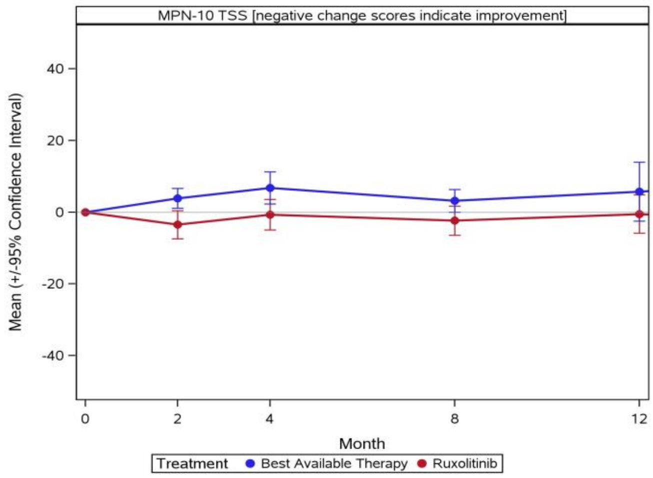


Figure 3C.

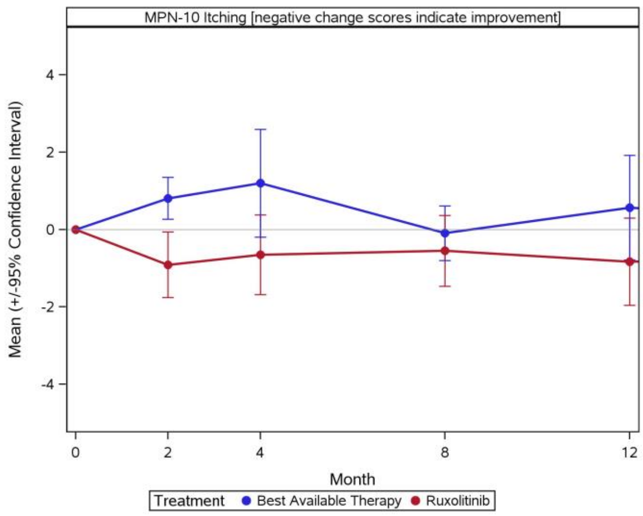


Figure Legends

Figure 1

Figure illustrating doses of ruxolitinib throughout the MAJIC-ET trial

Figure 2

Trial consort diagram at 2 year follow up

Figure 3

Changes in ET Related Symptom Burden during year 1 of the MAJIC-ET trial

Figure 3A shows a waterfall plot of maximum percentage change in the MPN SAF TSS score, dotted line indicates 50% reduction in TSS

Figure 3B shows mean MPN-SAF TSS throughout the first year of the trial there was a consistent trend for reduction for ruxolitinib

Figure 3C shows the mean MPN-SAF score for itching during the first 12 months of the MAJIC-ET trial

Tables

Table 1. Baseline characteristics by treatment

	Best Available Therapy (52)	Ruxolitinib (58)	Overall (110)
Age			
Mean (sd) [Range]	65.6 (13.5) [37.2, 85.4]	62.9 (12.3) [34.5, 90.5]	64.2 (12.9)[34.5, 90.5]
Gender, n (%)			
Female	30 (57.7)	36 (62.1)	66 (60.0)
Male	22 (42.3)	22 (37.9)	44 (40.0)
Mutation status, n (%)			
*JAK2*V617F Positive	26 (50.0)	28 (48.3)	54 (49.1)
CALR mutation positive	14 (26.9)	20 (34.5)	34 (30.9)
MPL mutation positive	3 (5.8)	3 (5.2)	6 (5.5)
Triple negative	7 (13.5)	6(10.3)	13(11.8)
Not run	2 (3.8)	1 (1.7)	3 (2.7)
HC Resistant or Intolerant*, n (%)			
Resistant	25 (48.1)	28 (48.3)	53 (48.2)
Intolerant	27 (51.9)	30 (51.7)	57 (51.8)
Time from diagnosis to randomization, years**			
Mean (sd) [Range]	6.9 (5.8) [.4, 23.6]	10.4 (6.7) [.7, 31.2]	8.8 (6.5)[.4, 31.2]
Hemoglobin, g/L**			
Mean (sd) [Range]	126 (17) [90.0, 160.0]	119 (17) [87.0, 152.0]	122 (17) [87.0, 160.0]
Platelet count x 10^9 / L			
Mean (sd) [Range]	573.0 (227.1) [166.0, 1406.0]	545.4 (215.3) [89.0, 1139.0]	558.4 (220.4) [89.0, 1406.0]
WBC count x 10^9 / L			
Mean (sd) [Range]	6.8 (2.7) [2.8, 15.2]	7.5 (4.8) [1.7, 29.8]	7.2 (3.9) [1.7, 29.8]
Hematocrit			
Mean (sd) [Range]	0.4 (0.1) [0.3, 0.5]	0.4 (0.1) [0.3, 0.5]	0.4 (0.1) [0.3, 0.5]
Spleen size			
Enlarged	9	14	23
Normal	38	37	75
Splenectomy	2	3	5
Missing	3	4	7
Number of Previous Therapies, n (%)			
1	15(28.8)	14 (24.1)	28 (26.4)
2	20 (38.5)	24 (41.4)	44 (40.0)
3	8 (15.4)	12 (20.7)	20 (18.2)
4	5 (9.6)	5 (8.6)	10 (9.1)
5	2 (3.8)	2 (3.4)	4 (3.6)
6	2 (3.8)	0 (.0)	2 (1.8)
9	0 (.0)	1 (1.7)	1 (.9)
Total number of previous therapies by treatment, number (%) [¥]			
Hydroxycarbamide	59 (52.2)	70 (58.8)	129 (55.6)
Anagrelide	29 (25.7)	31 (26.1)	60 (25.9)
Interferon	7 (6.2)	11 (9.2)	18 (7.8)
Pegylated Interferon	2 (1.8)	2 (1.7)	4 (1.7)
Busulfan	8 (7.1)	1 (.8)	9 (3.9)
32P	3 (2.7)	1 (.8)	4 (1.7)
Pipobroman	1 (.9)	1 (.8)	2 (.9)
Fedratinib	1 (.9)	1 (.8)	2 (.9)
Vorinostat	2 (1.8)	0 (.0)	2 (.9)
Thalidomide	0 (.0)	1 (.8)	1 (.4)
Missing	1 (.9)	0 (.0)	1 (.4)

HC Hydroxycarbamide; WBC white blood cell; *25 patients were both resistant and intolerant. These patients have been included as resistant; **Time from diagnosis to randomization and baseline hemoglobin were different between the two treatment arms; [¥] Patients were allowed to receive multiple therapies, therefore total number of therapies in each category might exceed number of patients

Table 2. **Overview of assigned therapy switches and discontinuations per treatment arm**

		Ruxolitinib	BAT	Total
Assigned therapy switches				
Patients that switched BAT therapy at least once		N/A	30	30
Total number of times BAT therapy was switched		N/A	86	86
Discontinuations				
Transformation		9	3	12
Loss of response		11	0	11
Lack of efficacy		5	1	6
Toxicity	Anemia	2	0	2
	Other	3	1	4
Other		3	3	6
Death		1	2	3
Withdrawal of consent		1	0	1
Total		*35*	*10*	*45*

Table 3. Thrombotic and hemorrhagic events

		BAT			Ruxolitinib			Total
		Grade 1&2	Grade 3&4	Grade 5	Grade 1&2	Grade 3&4	Grade 5	
Hemorrhagic events								
Hematuria		1	0	0	0	0	0	1
Intracranial hemorrhage		0	0	1	0	0	0	1
Oral hemorrhage		1	0	0	1	0	0	2
Rectal hemorrhage		1	1	0	0	0	0	2
Total		*3*	*1*	*1*	*1*	*0*	*0*	*6*
Thrombotic events ᵻ								
Chest pain - cardiac		0	1	0	0	0	0	1
Myocardial infarction		0	0	0	0	2	0	2
Cerebrovascular ischemia		1	0	0	0	0	0	1
Retinal vascular disorder		1	0	0	1	0	0	2
Thromboembolic events	PE	0	0	0	0	3*	0	3
	DVT	0	0	0	1	1	0	2
	Calf vein DVT	0	0	0	0	1	0	1
Transient ischemic attacks		2	0	0	2	0	0	4
Total		*4*	*1*	*0*	*4*	*7*	*0*	*16*

PE Pulmonary Embolism; DVT Deep Vein Thrombosis

ᵻ The death of a ruxolitinib treated patient due to ischemic cardiomyopathy occurred more than 30 days past treatment and is therefore not recorded as an event
* 1 patient experienced PE and DVT at the same time, but was counted in the PE category