Distinct clustering of symptomatic burden amongst myeloproliferative neoplasm patients: retrospective assessment in 1470 patients

Holly L. Geyer,1 Robyn M. Emanuel,1 Amylou C. Dueck,2 Jean-Jacques Kiladjian,3 Zhijian Xiao,4,5 Stefanie Slot,6 Sonja Zweegman,6 Federico Sackmann,7 Ana Kerguelen Fuentes,8 Dolores Hernández-Maraver,8 Konstanze Döhner,9 Claire N. Harrison,10 Deepti Radia,10 Pablo Muxi,11 Carlos Besses,12 Francisco Cervantes,13 Peter L. Johansson,14 Bjorn Andreasson,14 Alessandro Rambaldi,15 Tiziano Barbui,15 Alessandro M. Vannucchi,16 Francesco Passamonti,17 Jan Samuelsson,18 Gunnar Birgegard,19 and Ruben A. Mesa20

1Division of Hospital Internal Medicine, Mayo Clinic, Scottsdale, AZ; 2Section of Biostatistics, Mayo Clinic, Scottsdale, AZ; 3Clinical Investigation Center, Hospital Saint-Louis, Paris, France; 4MDS and MPN Centre, Institute of Hematology and Blood Diseases Hospital, Chinese Academy of Medical Sciences and Peking Union Medical College, Tianjin, China; 5State Key Laboratory of Experimental Hematology, Institute of Hematology and Blood Diseases Hospital, Chinese Academy of Medical Sciences & Peking Union Medical College, Tianjin, China; 6Department of Hematology, VU University Medical Center, Amsterdam, Netherlands; 7Fundaleu, Buenos Aires, Argentina; 8Department of Haematology, University Hospital La Paz, Madrid, Spain; 9Department of Internal Medicine III, University Hospital of Ulm, Germany; 10Dept. of Haematology, Guy's and St.
Thomas NHS Foundation Trust, London, United Kingdom; 11Unidad de Hematología, Hospital Británico, Montevideo, Uruguay; 12Hematology Department, Hospital del Mar, Barcelona, Spain; 13Hematology Department, Hospital Clínico, IDIBAPS, University of Barcelona, Spain; 14Internal Medicine, NU Hospital Organization, Uddevalla, Sweden; 15Unit of Hematology, Azienda Ospedaliera Papa Giovanni XXIII, Bergamo, Italy; 16Hematology Division of Hematology, Ospedale di Circolo, Varese, Italy; 17Department of Hematology, Fondazione IRCCS Policlinico San Matteo, University of Pavia, Pavia, Italy; 18Department of Internal Medicine, Stockholm South Hospital, Stockholm, Sweden; 19Dept. of Hematology, University Hospital, Uppsala, Sweden; 20Department of Hematology/Oncology, Mayo Clinic, Scottsdale, AZ.

Keywords: Quality of Life, Myeloproliferative neoplasms, polycythemia vera, essential thrombocytemia, myelofibrosis, symptom clusters

H.L.G and R.M.E contributed equally to this study.

Presented at the 2013 European Hematology Association meeting (Stockholm, Sweden)
Key Points:

1. Distinct clusters exist within polycythemia vera, essential thrombocythemia and myelofibrosis.

2. Clusters are not direct surrogates for current prognostic scores.
ABSTRACT

Symptom burden in myeloproliferative neoplasms (MPNs) is heterogeneous even amongst patients within the same MPN diagnosis. Utilizing cluster analysis from prospectively gathered symptom burden data in 1470 international patients with essential thrombocythemia (ET), polycythemia vera (PV), or myelofibrosis (MF), we assessed for the presence of clusters and relationship to disease features and prognosis. In myelofibrosis (4 clusters identified), clusters significantly differed by DIPSS risk (p<0.001), leukopenia (p=0.009), thrombocytopenia (p<0.001) and spleen size (p=0.02). Although an association existed between cluster and DIPSS risk, high symptom burden was noted in some low and intermediate-1 risk MF patients. In PV (5 clusters identified), total symptom score increased across clusters (p<0.001) but clusters did not significantly differ by PV risk or the risk assessment variable of age. Amongst ET patients (5 clusters identified), clusters differed by gender (p=0.04), anemia (p=0.01), and prior hemorrhage (p=0.047). Total symptom score increased across clusters (p<0.001) but clusters did not significantly differ by IPSET risk including the risk assessment variables. Significant symptom heterogeneity exists within each MPN subtype sometimes independent of disease features or prognosis.
BACKGROUND:

The Philadelphia-chromosome negative myeloproliferative neoplasms (MPNs), including essential thrombocythemia (ET), polycythemia vera (PV) and myelofibrosis (MF) originate from malignant transformation of oligoclonal and polyclonal myeloid-derived hematopoietic stem cells. Progression to MF (including post-PV MF, post-ET MF or primary myelofibrosis) leads to disease burdens that may include cytopenias, splenomegaly and/or constitutional symptoms which in aggregate may significantly compromise quality of life.

For physicians, assessing and treating the symptom burden represents one of the most challenging features of managing MPNs due to the paucity of effective treatment regimens and marked heterogeneity between patient presentations. In 2007, our group published the results of a cross-sectional survey of 1,179 MPN patients which helped assess and quantify that these patients do indeed suffer from excessive fatigue compared to age-matched controls and that symptoms compromise social functioning, physical activity, independence in daily tasks, and global quality of life. Additionally, this survey found that traditional therapies including hydroxyurea, interferon, thalidomide, corticosteroids, androgens, and erythropoietin analogues were suboptimal in reducing symptoms.
To assist with assessment of MPN symptom burden, we first created the Myelofibrosis Symptom Assessment Form (MF-SAF) by utilizing the symptoms endorsed by patients in our original online survey, and then expanded it to include an additional seven symptoms relevant to ET and PV. This expanded measure, entitled the MPN Symptom Assessment Form (MPN-SAF), was validated among an international sample of MPN patients and subsequently translated into 11 languages. In order to ease survey administration for serial use in assessment of therapeutic interventions, the MPN-SAF total symptom score (MPN-SAF TSS) was created as an abbreviated 10-item measure containing only the most representative and pertinent MPN symptoms to be given in conjunction with the Brief Fatigue Inventory (BFI) as a convenient assessment of symptom burden.

Subsequent investigations utilizing these patient-reported outcomes (PROs) have identified significant symptomatic heterogeneity between MPN subtypes and their correlations to disease duration and symptomatic burden. Our more recent study included an in-depth evaluation of 17 independent symptoms present among an MPN population and found that the prevalence and severity of symptoms varied widely between MPN subtypes (range in prevalence 17% - 59% for ET, 18% - 68% for PV, and 29% - 77% for MF) with much intra- and inter-patient symptom variability. To date there have been no investigations into whether the heterogeneity observed within MPN subtypes represents unique symptom clusters. This study represents the first investigation of the existence and inter-relations between MPN symptom clusters via prospectively gathered
information on disease symptoms and features. We additionally sought to identify the spectrum and relationship of such clusters in association with both disease features as well as estimated prognosis.

METHODS:

Survey development and collection: Patient and physician-reported demographics, MPN disease features and symptom burden data were collected from an international cohort in a manner as was previously published.³ Patients were recruited from academic, government, and private medical centers internationally during a routine office visit. Physicians were queried on available laboratory and clinical data including hemoglobin level, platelet count, white blood cell count, absolute neutrophil count, percent blasts and spleen size. This study has been approved by our Institutional Review Board and designated the reference number 09-008764. This study was conducted in accordance with the Declaration of Helsinki.

Symptom burden assessment completed by the patient at a single time point included the BFI and MPN-SAF, which addressed key disease features of fatigue, early satiety, abdominal pain and discomfort, inactivity, headaches, concentration, dizziness, extremity tingling, insomnia, sexual difficulties, mood changes, cough, night sweats, pruritus, bone pain and fever. Items were scored on a 0 (absent) to 10 (worst-imaginable) scale. MPN-SAF TSS items included
“worst fatigue” from the BFI plus concentration, early satiety, inactivity, night sweats, itching, bone pain, abdominal discomfort, weight-loss and fever. For individuals completing at least 6 of the 10 MPN-SAF TSS items, the survey was scored by multiplying the average score across items by 10 to achieve a 0-100 scaled score. Survey translations were developed using a PRO translation method\(^7\) by teams of four international collaborators. Available translations of the survey included Chinese, Danish, Dutch, English (United States and United Kingdom), French, German, Italian, Japanese, Spanish, Swedish and Portuguese.

**Prognostic Scoring:** Prognostic groups for MF were calculated using the Dynamic International Prognostic Scoring System (DIPSS)\(^8\) which utilizes the factors of age (>65), blasts present in peripheral blood (>1%), hemoglobin (<10 g/dL), leukocyte count (>25x10^9/L), and the presence of constitutional symptoms (including weight loss >10%, night sweats or fevers). This scale can be used at any time during the course of MF to estimate the risk of progression and separates patients into low, intermediate-1, intermediate-2 and high-risk categories. For PV, prognostic scoring utilized a model developed by Tefferi and colleagues which includes the variables of age (over 70 or 60-69) and leukocyte count greater than or equal to 15x10^9/L to indicate low, intermediate-1, intermediate-2 or high risk.\(^9\) The International Prognostic Score for ET (IPSET)\(^10\) was assessed among ET patients and included the variables of age (over 60 years), leukocyte count (greater than or equal to 11x10^9/L), and history of thrombosis to indicate low, medium or high-risk of progression. For the purposes
of this study anemia, thrombocytopenia and leukopenia were defined as hemoglobin under 11 g/dL, platelet count under 150x10^9/L and leukocyte count under 3.5x10^9/L.

Statistical analysis: Cluster analysis was performed using 19 individual symptom items (the worst fatigue item of the BFI and the 18 individual symptom items of the MPN-SAF) within each disease group independently. The number of clusters was based on consideration of r-squared (see Figure A2) and semi-partial r-squared while maintaining individual cluster sizes to be at least 5% of the sample in hierarchical clustering using Ward linkage. In order to be included in the cluster analysis, patients needed to complete all 19 questions in the survey. Thus, 1,470 out of 1,885 patients in the database were included. Final cluster assignment within disease was based on the nonhierarchical k-means method using the number of clusters identified via the previous hierarchical clustering procedure. Split-half validation was used to examine internal validity of the final cluster results. In split-half validation within each disease group, cluster assignment is performed in a randomly selected pair of split-half subsamples, and the Euclidean distance between each subject’s symptom scores and the mean symptom scores for that patient's cluster is compared between split-half subsamples using a Kruskal-Wallis test (the size of the difference is described using the Hodges-Lehmann [HL] estimator). A small, non-significant difference between split-half subsamples would indicate adequate internal validity. In all disease groups, split-half validation indicated adequate internal validity (MF: HL=-0.02, 95% confidence interval [CI] -0.8-0.7, p=0.95; ET: HL=0.06, 95% CI -0.5-
Comparisons between symptom clusters were based on analysis of variance (ANOVA) F-tests or t-tests for continuous variables and chi-squared tests for categorical variables. Lastly, MPN-SAF TSS was compared across risk groups using ANOVA F-tests.

RESULTS:

**ET Symptom Clusters**

*Subject Demographic and Disease Characteristics:*

Analysis included data from 622 prospectively enrolled persons with ET (Chinese 149, French 174, German 20, Italian 65, Dutch 72, English 29, Spanish 67, Swedish 46). Participants were of typical age (median 58, range 15-90) for the disease. The group was predominantly female (63.2%; Table A1).

*Cluster Overview:*

Among all participants, five symptom clusters were identified. Distribution of age and gender, along with language, laboratory abnormality (anemia), and prior hemorrhage differed significantly across clusters (p<0.05, Table 1 and A1). MPN-SAF TSS also significantly differed across clusters (P<0.001, Figure 1A). A heatmap (visual representation of individual symptom levels across patients ordered by cluster) is presented in Figure 2A (also see Figure A2 for mean symptom scores by cluster). IPSET risk scores did not significantly differ across
clusters (p=0.43) including the IPSET variable of prior thrombosis (p=0.24). No chronological trends were noted among clusters (MPN duration, p=0.89).

Cluster Descriptions:

Cluster 1: Mild ET (n=247, 40%). The largest cluster, these patients had the lowest individual MPN-SAF symptom scores and the lowest MPN-SAF TSS. Mean symptom scores were all ≤1.1 other than fatigue (mean 2.0). The majority of patients (59.4%) had disease duration less than 3 years and most patients were Low (38.2%) or Intermediate (45.9%) risk. Anemia was uncommon (21.2%).

Cluster 2: Moderate-I ET (n=97, 16%). This cluster was typified by the predominant complaint of sexual dysfunction (mean 7.6) out of proportion to other symptoms (all means ≤2.6 other than fatigue, mean 3.3). These patients had the second-to-lowest overall MPN-SAF TSS. The majority of patients (62.7%) had disease duration less than 3 years. Subjects in this group were predominantly female (61.7%) and had Intermediate risk (54.9%). This cluster had the highest prevalence of anemia (36.3%). Patients also had the lowest rate of prior thrombosis (17.5%).

Cluster 3: Moderate-II ET (n=155, 25%). This cluster was characterized by high MPN-SAF symptom scores related to fatigue (mean 5.4) and insomnia (mean 3.5). The majority of patients (55.4%) had disease duration less than 3 years. Patients were primarily of Low (40.8%) or Intermediate (45%) risk. This
cluster was least likely to have a laboratory abnormality (23.1%) including anemia (19.8%) and leukopenia (1.7%).

**Cluster 4: High-I ET (n=55, 9%).** Cluster 4 has the highest level of fatigue (mean 7.0) along with a high level of complaints of sexual dysfunction (mean 6.8). This cluster also had the highest percentage of patients in the High IPSET risk score category (24.2%) among ET clusters. The majority of patients (52.6%) had disease duration of 3 years or more. This cluster also had the second highest MPN-SAF TSS. This cluster also had the highest level of anemia (33.3%) and greatest spleen size (mean 1.2 cm below the left costal margin).

**Cluster 5: High-II ET (n=68, 11%).** This profile demonstrated considerable symptomatology in virtually all individual MPN symptoms (all means ≥3.5 other than fever, mean 1.2, and weight loss, mean 2.6). The majority of patients (55.2%) had disease duration less than 3 years. Patients were represented equally between Low (40.7%) and Intermediate-1 (40.7%) IPSET risk score categories. This cluster was likely to have a laboratory abnormality (38.2%) including leukopenia (9.1%) and thrombocytopenia (5.5%), along with prior thrombosis (27.7%), prior hemorrhage (11.9%) and need for red blood cell transfusions (2.9%).
PV Symptom Clusters

Subject Demographic and Disease Characteristics:

Data from 519 patients was prospectively collected and combined (Chinese 87, French 131, German 25, Italian 49, Dutch 119, English 27, Spanish 42, Swedish 39). Age (median 62, range 22-91) and gender (56.9% male) were typical of the disease. Overall, 28% of patients had the presence of at least one lab abnormality (Table A2) including anemia (21%), leukopenia (4%) and thrombocytopenia (8%).

Cluster Overview:

Five clusters were identified. Distribution of gender, language, laboratory abnormalities, spleen size, and leukopenia varied significantly across clusters (p<0.05, Table 1 and A2). MPN-SAF TSS varied significantly among clusters (p<0.001; Figure 1B). A heatmap showing variability of symptoms across clusters is presented in Figure 2B (also see Figure A2 for mean symptom scores by cluster). No significant association was noted between PV risk group and clusters (p=0.87). No association was noted between disease duration and clusters (p=0.73).

Cluster Descriptions:

Cluster 1: Mild PV (n=253, 49%). The largest cluster (n=253), individual symptom scores remained relatively low (all means ≤1.5 except fatigue, mean
2.5). The majority (56%) of patients had disease duration of 3 years or more. Subjects were mostly male (58.9%). Most patients were in the Low (36.5%) or Intermediate-1 (34%) risk category. This cluster also had the lowest number of patients with a laboratory abnormality (21.3%) including anemia (15.8%) and leukopenia (1.5%). These patients also had the smallest spleen size (mean 1.7 cm) and the lowest rate of prior hemorrhage (4.3%) of all clusters.

Cluster 2: Moderate-1 PV (n=118, 23%). This cluster was typified by the predominant complaint of sexual dysfunction (mean 7.2) following by fatigue (mean 4.7) and insomnia (mean 3.6) with the second to lowest mean MPN-SAF TSS. The majority of patients (62.5%) had disease duration of 3 years or more. Subjects in this cluster were predominantly male (65.8%) and were equitably dispersed among PV risk categories of Low (32.6%), Intermediate-1 (34.9%) and Intermediate-2 (30.2%) risk. This cluster had the largest percentage of patients with a laboratory abnormality (41.9%) including anemia (29.1%), thrombocytopenia (11.6%) and leukopenia (10.5%). They also had the highest percentage of patients with a prior splenectomy (1.6%).

Cluster 3: Moderate-2 PV (n=62, 12%). This cluster was characterized by high MPN-SAF scores in cytokine–related symptoms including fatigue (mean 6.9) and pruritus (mean 6.9). The majority of patients (68.4%) had a disease duration of 3 years or more. Patients in this cluster were of equal gender distribution (48.4% male) and the majority of patients were in the Low PV risk category
(36.8%). The cluster had the second to lowest percentage of patients with a laboratory abnormality (25.6%) including anemia (18.4%) and leukopenia (2.6%). These patients also had the highest rate of prior hemorrhage (14.8%) amongst the clusters.

**Cluster 4: High-I PV (n=59, 11%).** Cluster 4 is characterized by a higher proportion of abdominal-related symptoms including abdominal discomfort (mean 4.6), abdominal pain (mean 4.2) and early satiety (mean 5.1) in addition to high fatigue (mean 7.0) and insomnia (mean 5.5). Unlike Clusters 1, 2 and 3, the majority of these patients had disease duration less than 3 years (61.1%). Patients were of equal gender distribution (49.2% male) and the majority of patients were in the Low (47.1%) or Intermediate-2 (32.4%) PV risk category. A high percentage of patients had anemia (29.4%). This cluster demonstrated the lowest proportion of patients with prior thrombosis (19.3%) and the highest proportion requiring red blood cell transfusions (3.4%).

**Cluster 5: High-II PV (n=27, 5%).** This profile was characterized by high levels of all individual MPN-SAF symptoms (all means ≥5.4) with the exception of fever (mean 2.6) and weight loss (mean 3.1) and is notable for the highest mean MPN-SAF TSS of all PV clusters. The majority of patients (60.0%) had disease duration of 3 years or more. The smallest cluster (n=27), patients were predominantly female (63%) and were comprised of PV risk category
Intermediate-1 (45.5%). No patients had thrombocytopenia but the cluster did demonstrate the highest rate of prior thrombosis (34.8%). The rate of anemia was 27.3%.

**MF Symptom Clusters**

**Subject Demographic and Disease Characteristics:**

Data from 329 prospectively enrolled persons with MF was collected (Chinese 102, French 54, German 19, Italian 22, Dutch 45, English 51, Spanish 29, Swedish 7) including 223 PMF, 67 post-ET MF and 39 post-PV MF patients. Participants were of typical age (median 60, range 26-87) and gender for MPN disease (52.6% male).

**Cluster Overview:**

Among all participants, four symptom clusters were identified. Among clusters, disease features including leukopenia, thrombocytopenia and enlarged spleen varied significantly among clusters (p<0.05, Table 1 and A3). MPN-SAF TSS varied significantly among clusters (p<0.001; Figure 1C). Heatmap showing variability of symptoms across clusters is presented in Figure 2C (also see Figure A2 for mean symptom scores by cluster). DIPSS risk was associated with cluster assignment (p=0.001) with the proportion of patients with Intermediate-2 or High-risk classification increasing from 20.5% in the first cluster to 66.7% in the last cluster (when clusters are arranged by mean MPN-SAF TSS). Longer MF
duration trended towards higher cluster as well (p=0.06), though association between MPN duration and cluster was not statistically significant (p=0.15).

**Cluster Descriptions:**

**Cluster 1: Mild MF (n=150, 46% [69% PMF, 20% post-ET MF, 11% post-PV MF])**. The largest of all MF clusters, Cluster 1 was characterized by fatigue-dominant complaints (mean 3.1) in the setting of the lowest overall MPN-SAF TSS, the shortest MF disease duration (61.3% under 3 years) and the highest proportion of males (58.7%). The majority of patients in this cluster were DIPSS Intermediate-1 risk (46.2%), followed by Low risk (33.3%). Individuals in this group had the lowest prevalence of a laboratory abnormality (64.6% total; anemia, 55.6%; thrombocytopenia, 19.8%). Additionally, these individuals had the lowest proportion of clinical deficiencies including splenomegaly (mean 6.0 cm below the left costal margin), need for red blood cell transfusions (20.4%), prior thrombosis (8.8%) and prior hemorrhage (4.7%). Interestingly, individuals in this group were most likely to have had a prior splenectomy (5.8%).

**Cluster 2: Moderate-I MF (n=105, 32% [65% PMF, 20% post-ET MF, 15% post-PV MF])**. Cluster 2 was the second largest cluster. Patients had the longest MF disease duration of all clusters (mean 6.7 years) with 50% having disease duration under 3 years. The majority of patients in this cluster were DIPSS Intermediate-1 risk (54.2%), followed by Intermediate-2 risk (23.7%). Subjects had a moderate rate of lab abnormalities (67.4%; anemia, 56.2%; thrombocytopenia 34.5%) but high severity of fatigue (mean 6.1), sexual
dysfunction (mean 6.0), sad mood (mean 4.1), and inactivity (mean 4.0). Transfusion dependency was present in 21% of the sample and this cluster also had the largest spleen size (mean 8.7 cm below the left costal margin).

Cluster 3: Moderate-II MF (n=53, 16% [64% PMF, 25% post-ET MF, 11% post-PV MF]). The majority of patients (70.6%) had MF disease duration of 3 years or more with a mean duration of 4.8 years. This cluster had the highest percentage of Intermediate-1 risk patients (63.6%) of all clusters. Under 5% of Cluster 3 patients were Low or High risk. Cytopenias were intermediate (68.4%; anemia 65.8%; thrombocytopenia 27%). Patients in this cluster had many cognitive and nighttime-related complaints including sexual difficulties (mean 6.4), night sweats (mean 6.1), insomnia (mean 5.4) and concentration problems (mean 5.6). It also had the highest relative proportion of post-ET MF cases among the clusters. Additionally, individuals in this cluster had the lowest rate of leukopenia (7.9%), the second smallest spleen size (mean 7 cm) and the second lowest rate of prior thrombosis (9.6%), prior hemorrhage (7.8%) and requirement for red blood cell transfusions (21.2%). No patients had a history of splenectomy.

Cluster 4: High MF (n=21; 6% [81% PMF, 14% post-ET MF, 5% post-PV MF]). Cluster 4 was the most symptomatic cohort with the highest proportion of subjects with PMF. Mean MF disease duration was higher than most clusters (6.5 years), but less than that observed in Cluster 2. This cluster had the highest percentage of DIPSS High risk patients (33.3%) and no patients were Low risk. This cluster had lower levels of end-organ complaints including abdominal pain
(mean 4.8), cough (mean 5.7), and headaches (mean 4.1) compared to other symptoms. Symptoms including fatigue (mean 8.0), sexual difficulties (mean 8.9), sad mood (mean 7.8) and insomnia (mean 8.0) were predominant. Subjects also had the highest prevalence of prior thrombosis (28.6%), prior hemorrhage (14.3%) and transfusions (42.9%). Additionally, this cohort had the largest prevalence of lab abnormalities (76.5%) with frequent anemia (70.6%), thrombocytopenia (70.6%) and leukopenia (41.2%). No subjects had a history of prior splenectomy.

DISCUSSION

Myeloproliferative neoplasms including myelofibrosis, polycythemia vera and essential thrombocythemia can be grouped into specific profiles with similar symptomology, physical and laboratory findings. Our previous studies have indicated that symptom burden among MPN patients is severe and heterogeneous within disease subtypes despite active treatment with standard therapies. However, the question of whether the heterogeneity observed in each MPN subtype was attributable to the existence of unique symptom clusters remained unknown. This study represents the first examination of these differences in symptom burden by cluster analysis.

As previously described, significant heterogeneity was observed both between and within MPN subtypes and likely originates from variances in
biological activity of disease, genetic mutations and cultural influences present within the MPN population. In addition to JAK2\textsuperscript{V617F} which occurs at a frequency between 55\% and 90\% in MPNs, mutations including MPL,\textsuperscript{13} EZH2,\textsuperscript{14} ASXL1,\textsuperscript{15} IDH1/2,\textsuperscript{16} TET2,\textsuperscript{17} CBL,\textsuperscript{18} IKZF1\textsuperscript{19} and p53\textsuperscript{20} have been identified as contributing to cellular deregulation. Such mutations have been found to affect hematopoietic response and cytokine signaling which likely results in many of the variances observed in laboratory and physical exam findings. In myelofibrosis, association existed between clusters and DIPSS risk groups. However, significant symptom burden was also detected within Low and Intermediate-1 risk disease suggesting that the phenotypic heterogeneity observed within MPNs is not solely a surrogate for disease prognosis as currently assessed by MPN prognostic scores and that stages within each MPN subtype may exist as a combination of assessed symptomatic burden and prognosis. The absence of linear chronologic progression between symptoms and risk scores within ET and PV further support the existence of biological subsets within MPN subtypes and suggest that current risk scores cannot be applied as surrogates of disease severity.

Analysis of laboratory abnormalities in MF revealed a direct association with symptom severity suggesting that the roles anemia, leukopenia and thrombocytopenia play in MPN symptom development should not be disregarded. Anemia may result in increased fatigue and thrombocytopenia may reflect platelet sequestration within the spleen which may secondarily result in abdominal pain/discomfort, early satiety and weight loss. Leukopenia may cause
increased risk of infection and subsequent fatigue and fevers. The associations between symptoms and laboratory abnormalities in PV and ET were less robust. This can be explained by the decreased prevalence and severity of lab abnormalities in comparison to MF. Additionally, this study included post-PV and post-ET MF within the MF analysis which represent later and more symptomatic stages of ET and PV. The correlation between decreased splenomegaly and minimal cytopenias in MF and PV Cluster 1 is consistent with previous findings that splenic sequestration can lead to anemia and thrombocytopenia.

Cluster 2 in both ET and PV was dominated by significant complaints of sexual dysfunction. Uniquely, these two clusters were expressed in both eastern and western language groups and differed between males and females (PV=Dutch, M>F; ET=Chinese, F>M). These differences may originate from the prevalence of each gender surveyed within the MPN subgroup (M>F in PV, F>M in ET). The pathophysiology behind sexual complaints is likely related to both microvascular and macrovascular pathology, along with elevated cytokines that inhibit autoregulatory function inherent to reproductive processes. Intriguingly, Cluster 2 in ET had the lowest history of prior thrombosis suggesting that thrombocytosis and thrombosis may play a lesser role in the development of sexual dysfunction than historically speculated. Differences in the prevalence of specific languages between clusters was also observed (insignificant for MF, p<0.001 for ET/PV clusters). It remains to be determined if the increased expression of certain symptoms within language groups reflects cultural values or
phenotypic variations of the same disease process as a result of geographical segregation.

Limitations to this study include the possibility of inappropriate categorization of early MF patients into the ET and PV cohorts. As noted in Tables A2 and A3, a subset of PV and ET patients demonstrated hemoglobin levels under 11 g/dL and/or transfusion dependency. The anemia in these populations may stem from variety of sources including other medical comorbidities and do not necessarily infer miscategorization. Given the large number of patients and international nature of the study, the primary authors did not personally review bone marrow histology and individual contributing institutions were required to follow standard-of-care diagnostic techniques in determining formal diagnosis. As previously described, the majority of ET and PV patients were receiving concurrent therapy for their MPN disorders, likely contributing to anemia development. No statistical differences were noted between ET and PV clusters when analyzing treatment history. Additionally, the number of ET and PV patients with anemia far exceed the number expected to transform to myelofibrosis from the time of bone marrow biopsy to study initiation. Patient characteristics (for all combined clusters) within each MPN category including patient demographics, exam findings and MPN history are consistent with the known literature on these topics further supporting the limited impact of miscategorization. This study is also notable for having a limited number of patients within MF Cluster 4 and PV Cluster 5. As we identified, the MF
symptom clusters correlated with DIPSS risk scores and thus the low number of patients in MF Cluster 4 may be a reflection of disease severity and higher mortality rate hens limiting population size. The low number of patients in the highly symptomatic PV cluster 5 is congruent with current literature suggesting a limited number of PV patients within the community who describe a high symptom burden.

Within the past decade, investigations of pharmacotherapies capable of ameliorating MPN symptoms have uncovered a variety of compounds demonstrating efficacy in improving symptoms, quality of life and overall prognosis. JAK2 inhibitors are amongst those therapies that have been approved for the treatment of MPN related symptoms. Ruxolitinib was the JAK1/JAK2 inhibitor first to reach FDA approval in November, 2011 after its efficacy was demonstrated in two randomized controlled studies, The Controlled Myelofibrosis Study with Oral JAK Inhibitor Treatment Trials (COMFORT-I and COMFORT-II). In both investigations, ruxolitinib was compared against placebo (COMFORT-I)\textsuperscript{21} or best available therapy (COMFORT-II)\textsuperscript{22} in Intermediate and High-risk MF and demonstrated efficacy in reducing splenic size, improving symptoms and imparting survival advantage. Similarly, evaluation of ruxolitinib in PV has shown significant reduction in splenic length, leukocytosis and JAK2 burden.\textsuperscript{23} In ET, improvements in thrombocytosis and splenomegaly were noted.\textsuperscript{23} Other JAK2 inhibitors including CYT387,\textsuperscript{24} pactritinib (SB1518),\textsuperscript{25} SAR302503 (TG101348)\textsuperscript{26} and CEP701 (Lestaurtinib)\textsuperscript{27} have documented
similar results. Ruxolitinib is currently approved only for MF patients with Intermediate and High-risk disease. However, the results of this study demonstrate that even Low-risk MF populations may experience significant symptom burden and that these symptoms do not necessarily follow linear chronological progression across DIPSS risk categories. Results were similarly seen within ET and PV populations. Given this information, broader therapeutic application of these and other novel agents to lower risk MPN populations is a subject deserving of ongoing investigation.

In conclusion, our cluster analysis suggests that heterogeneic phenotypes exist within MPN subgroups and that these clusters are not direct surrogates for prognostic scores. Significant symptomatic burden may be observed in patients with Low and Intermediate-risk scores. Recognition of such subsets may impact choice, timing and goals of therapy as well as underline the role for serial assessment of symptom burden in a clinical setting.
Acknowledgements:

This article was produced through collaborative efforts by the MPN International Quality of Life Study Group. Study group participants are listed in Appendix 1. Additional contributors to the authorship of this article are listed in Appendix 2. We would also like to thank our French collaborators for their contributions to this project (listed in Appendix 3).

Contribution: RAM designed and supervised the study. HLG, RME, ACD and RAM interpreted results and drafted the paper. ACD performed statistical analysis. All other authors were instrumental in data acquisition and article review.

Conflict of Interest Disclosure: The authors declare no competing financial interests

Corresponding Author Contact Information: Ruben A. Mesa, MD, Mayo Clinic, 13400 E Shea Blvd. Scottsdale, AZ 85259; email: mesa.ruben@mayo.edu.
REFERENCES:


Table 1. Associations between cluster and patient disease characteristics by MPN subtype.

<table>
<thead>
<tr>
<th></th>
<th>P value^§</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>ET</td>
</tr>
<tr>
<td>Age</td>
<td>0.01</td>
</tr>
<tr>
<td>Gender</td>
<td>0.04</td>
</tr>
<tr>
<td>Language</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Risk Score^*</td>
<td>0.43</td>
</tr>
<tr>
<td>MPN duration (years)</td>
<td>0.89</td>
</tr>
<tr>
<td>Lab abnormality</td>
<td>0.04</td>
</tr>
<tr>
<td>Prior hemorrhage</td>
<td>0.047</td>
</tr>
<tr>
<td>Prior thrombosis</td>
<td>0.24</td>
</tr>
<tr>
<td>Prior splenectomy</td>
<td>0.44</td>
</tr>
<tr>
<td>Spleen size†</td>
<td>0.76</td>
</tr>
<tr>
<td>Palpable spleen</td>
<td>0.75</td>
</tr>
<tr>
<td>Required red blood cell transfusion</td>
<td>0.47</td>
</tr>
<tr>
<td>Receiving treatment‡</td>
<td>0.21</td>
</tr>
<tr>
<td>Anemia, hemoglobin under 11 g/dL</td>
<td>0.01</td>
</tr>
<tr>
<td>Leukopenia, leukocyte count under 3.5x10^9/L</td>
<td>0.14</td>
</tr>
<tr>
<td>Thrombocytopenia, platelet count under 150x10^9/L</td>
<td>0.28</td>
</tr>
</tbody>
</table>

MPN indicates myeloproliferative neoplasm; ET, essential thrombocythemia; PV, polycythemia vera; and MF, myelofibrosis.

^*Risk scores as assessed by IPSET^10 (ET), Tefferi 2012^9 (PV) or DIPSS^8 (MF).

† Spleen size determined by physical exam with mean size being represented by the number of cm below the left costal margin.

‡ Treatment versus no treatment/aspirin only.

^§See Tables A1-A3 in appendix for point estimates and measures of variability for each stated p-values.
Figure Legend

**Figure 1. Distribution of MPN-SAF TSS by cluster:** (A) Essential thrombocytemia; (B) Polycythemia vera; (C) Myelofibrosis.

**Figure 2. Symptom heatmaps.** (A) Essential thrombocytemia; (B) Polycythemia vera; (C) Myelofibrosis.
Distinct clustering of symptomatic burden amongst myeloproliferative neoplasm patients: retrospective assessment in 1470 patients


Information about reproducing this article in parts or in its entirety may be found online at: http://www.bloodjournal.org/site/misc/rights.xhtml#repub_requests

Information about ordering reprints may be found online at: http://www.bloodjournal.org/site/misc/rights.xhtml#reprints

Information about subscriptions and ASH membership may be found online at: http://www.bloodjournal.org/site/subscriptions/index.xhtml

Advance online articles have been peer reviewed and accepted for publication but have not yet appeared in the paper journal (edited, typeset versions may be posted when available prior to final publication). Advance online articles are citable and establish publication priority; they are indexed by PubMed from initial publication. Citations to Advance online articles must include digital object identifier (DOIs) and date of initial publication.