Supplemental Tables and Figures

Supplemental Table 1. List of baseline NDM including genomic position, nucleotide change, protein consequence, mutation type and variant allele frequency (VAF).

ID	Gene	Genomic position	Nucleotide change	Protein consequence	Mutation type	VAF (%)
35	*TET2*	4:106157960	c.2861G>A	p.Trp954Xaa	SNV; nonsense	52.39
35	*TET2*	4:106164860	c.3729_3733delACT	p.Tyr1245GlyfsTer21	Frameshift del	6.18
35	*TET2*	4:106193999	c.4462delA	p.Asn1489MetfsXaa82	Frameshift del	17.74
54	*TET2*	4:106155920	c.822delC	p.Asn275IlefsXaa18	Frameshift del	38.55
55	*TET2*	4:106197236	c.5570_5571delCT	p.Pro1857ArgfsXaa17	Frameshift del	7.06
106	*TET2*	4:106155301	c.202G>T	p.Gly68Xaa	SNV; nonsense	37
108	*TET2*	4:106164939	c.3803+4A>T		Frameshift ins	28.86
128	*TET2*	4:106156540	c.1441C>T	p.Gln481Xaa	SNV; nonsense	41.35
131	*TET2*	4:106180796	c.3824G>T	p.Gly1275Val	SNV; missense	33.92
148	*TET2*	4:106162586	c.3500G>A	p.Arg1167Lys	SNV; missense	28.4
180	*TET2*	4:106196446	c.4781del	p.Pro1594LeufsXaa2	Frameshift del	16.61
184	*TET2*	4:106156365	c.1270del	p.Ser424AlafsXaa3	Frameshift del	5.47
213	*TET2*	4:106180829	c.3857C>T	p.Ser1286Phe	SNV; missense	34.35
213	*TET2*	4:106180854	c.3882C>G	p.Tyr1294TXaa	SNV; nonsense	40.71
215	*TET2*	4:106156812	c.1716dup	p.His573SerfsTer10	Frameshift ins	17.79
35	*SF3B1*	2:198266834	c.2098A>G	p.Lys700Glu	SNV; missense	46.58
73	*SF3B1*	2:198267483	c.1874G>T	p.Arg625Leu	SNV; missense	28.77
87	*SF3B1*	2:198267359	c.1998G>C	p.Lys666Asn	SNV; missense	43.62
103	*SF3B1*	2:198267359	c.1998G>C	p.Lys666Asn	SNV; missense	41.33
151	*SF3B1*	2:198267359	c.1998G>C	p.Lys666Asn	SNV; missense	40.38
193	*SF3B1*	2:198267359	c.1998G>C	p.Lys666Asn	SNV; missense	19.56
123	*SF3B1*	2:198267360	c.1997A>G	p.Lys666Arg	SNV; missense	17.2
27	*TP53*	17:7578406	c.524G>A	p.Arg175His	SNV; missense	19.8
38	*TP53*	17:7577545	c.736A>G	p.Met246Val	SNV; missense	38.46
118	*TP53*	17:7577578	c.703A>G	p.Asn235Asp	SNV; missense	5.09
118	*TP53*	17:7577568	c.713G>A	p.Cys238Tyr	SNV; missense	13.01
131	*TP53*	17:7577556	c.725G>A	p.Cys242Tyr	SNV; missense	16.98
158	*TP53*	17:7577157	c.783-2A>C		Splice site	8.79
180	*TP53*	17:7578495	c.433_435del	p.Leu145del	Frameshift del	19.3
184	*TP53*	17:7577120	c.818G>A	p.Arg273His	SNV; missense	11.06
25	*ASXL1*	20:31022442	c.1934dup	p.Gly646TrpfsXaa12	Frameshift ins	28.64
115	*ASXL1*	20:31022442	c.1934dup	p.Gly646TrpfsXaa12	Frameshift ins	22.7
125	*ASXL1*	20:31022442	c.1934dup	p.Gly646TrpfsXaa12	Frameshift ins	27.72
130	*ASXL1*	20:31023408	c.2893C>T	p.Arg965Xaa	SNV; missense	9.03
196	*ASXL1*	20:31022793	c.2278C>T	p.Gln760Xaa	SNV; nonsense	38.79
61	*DNMT3A*	2:25457242	c.2645G>A	p.Arg882His	SNV; missense	15.69
128	*DNMT3A*	2:25457242	c.2645G>A	p.Arg882His	SNV; missense	37.12
198	*DNMT3A*	2:25457278	c.2603_2609del	p.Phe868SerfsXaa11	Frameshift del	23.97
57	*IDH2*	15:90631934	c.419G>A	p.Arg140Gln	SNV; missense	46.03
193	*IDH2*	15:90631934	c.419G>A	p.Arg140Gln	SNV; missense	19.22
101	*ZRSR2*	X:15821921	c.312+2T>A		Frameshift ins	10.98
104	*ZRSR2*	X:15838370	c.868C>T	p.Arg290Xaa	SNV; nonsense	92.73
61	*ETV6*	12:12006434	c.403del	p.His135ThrfsXaa74	Frameshift del	16.02
196	*PHF6*	X:133511706	c.59_60insT	p.Lys21Xaa	Frameshift ins	74.96
106	*SRSF2*	17:74732959	c.286G>A	p.Pro95Leu	SNV; missense	35.29
213	*EZH2*	7:148514322	c.1402T>G	p.Cys468Gly	SNV; missense	20.77
99	*CSF3R*	1:36932076	c.2474G>A	p.Gly825Glu	SNV; missense	39.63

Supplemental Table 2. Baseline clinical characteristics in patients with and without non-MPN driver mutations (NDM).

Differences were analyzed using the chi-squared test for categorical variables or fisher's exact test if there was a value less than 5 in any cells of the contingency table and non-parametric Mann-Whitney U test for continuous variables (i.e. age, disease duration, blood counts). MPN=myeloproliferative; patients=patients; n=number; BAT=best available therapy; Hb=hemoglobin; HC=hydroxycarbamide; Hct=hematocrit; NPM=non-MPN (myeloproliferative neoplasm) driver mutation; P32=radioactive phosphorus; Plt: platelet count; RUX=ruxolitinib; y=years; TN=triple negative; WBC=white blood cell count.

Baseline Clinical Characteristics		Patients with NDM n (%)	Patients without NDM n (%)	*P*
All patients (n=110)		33 (30)	77 (70)	
Median age in years (range)		71 (44 – 91)	64 (35 – 85)	0.0001
Gender	**Female** **Male**	17 (51.5) 16 (48.5)	49 (63.6) 28 (36.4)	0.234
HC-Resistant **HC-Intolerant**		19 (57.6) 14 (42.4)	34 (44.2) 43 (55.8)	0.197
Treatment	**BAT** **RUX**	17 (51.5) 16 (48.5)	35 (45.5) 42 (54.5)	0.560
Disease Duration at TE (y)		6.6 (0.8 – 31)	7.7 (0.4 – 25.9)	0.699
No. of Prior Therapy Lines	**<3** **≥3**	27 (81.8) 6 (18.2)	52 (67.5) 25 (32.5)	0.127
Interferon **Anagrelide**		2 (6.1) 15 (45.5)	16 (20.8) 38 (50.6)	0.089 0.706
Busulfan/P32/Pipobroman		3 (9.1)	9 (11.7)	1.0
Previous Thrombosis **Previous Hemorrhage**		7 (21.2) 2 (6.1)	28 (36.4) 4 (5.2)	0.118 0.855
Baseline palpable spleen		3 (9.1)	7 (9,1)	1.0
Baseline blood counts (median, range)	**WBC (x 10^9/l)** **Hb (g/l)** **Hct (%)** **Plt (x10^9/l)**	5.8 (1.7 – 15.2) 115 (90 – 147) 36 (28 - 45) 517 (166 – 1406)	6.1 (2.6 – 29.8) 125 (87 – 160) 38 (27 – 49) 530 (89 – 1139)	0.917 0.01 0.207 0.927
Driver mutation status	***JAK2*V617F** ***CALR*** ***MPL*** **TN**	19 (57.6) 7 (21.2) 3 (9.1) 4 (12.1)	36 (46.8) 26 (33.8) 2 (2.6) 13 (16.9)	0.237
***JAK2*V617F allele burden ≥50%**		5/19 (26.3)	6/36 (16.7)	0.395
***CALR* allele burden ≥50%**		0/7 (0)	2/26 (7.7)	1.0

Supplemental Table 3. Logistic regression predicting the influence of non-MPN driver mutations (NDM) on clinical outcomes. All models were adjusted for *JAK2*V617F mutation status and treatment type since patients were stratified by these at trial entry. Further adjusted analysis was performed for outcomes with significant ($p<0.05$) odd ratios (OR) on initial analysis to include age, TE hemoglobin level and platelet counts denoted indicated with "(adj)" next to OR. The presence of a HMR mutation significantly increased the odds of a transformation event, p=0.015. Hemorrhagic events outcomes were associated specifically with the presence of SF mutations. Platelet counts closest to the hemorrhagic event were normal in 3 of these SF-mutated patients and reduced (*ZRSR2*- mutated; 54 x 10^9/l) and elevated (*SF3B1*-mutated, 504 x 10^9/l) in one patient each. adj=adjusted; AML=acute myeloid leukemia; CHR=complete hematological response; CI=confidence interval; ELN; European LeukemiaNet; HMR=high molecular risk mutation (SF and/or *TP53* mutations); MF=myelofibrosis; n_E =number of events; non-MPN (myeloproliferative neoplasm) driver mutation; OR=odds ratio; RUX=ruxolitinib; SF= splicing factor mutation (*SF3B1, ZRSR2, SRSF2)*; TE=trial entry; UV=univariate. *Driver mutation allele burden ³50% and gender also included in final model for transformation.

Outcome (n_E)	OR (UV)	OR 95% CI	*P*
1-year CR (ELN) (n_E=50)	0.72	0.3 – 1.6	0.43
1-year post hoc platelet response (n_E =55)	0.5	0.2 – 1.2	0.12
1-year Overall symptom score response (³50% reduction) (n_E =12)	0.71	0.2 – 2.6	0.61
Transformations (MF n_E =13, AML n_E =1)	3.8 (NDM) (adj)* 14.4 (HMR) (adj)*	0.7– 21.5 1.7-122.8	0.129 0.015
Thrombotic event (n_E =21)	0.75	0.2 – 2.3	0.62
Hemorrhagic event (n_E =10)	2.1 (NDM)(adj) 18.9 (SF)(adj)	0.5 – 10 2.2 - 161	0.34 0.007
Death (n_E =13)	2.14	0.6– 2.3	0.21
Stopping RUX treatment (n_E =40)	0.38 (adj)	0.7 – 1.9	0.25

Supplemental Table 4. List of new follow-up NDM including genomic position, nucleotide change, protein consequence, mutation type and variant allele frequency (VAF).

ID	Gene	Genomic position	Nucleotide change	Protein consequence	Mutation type	VAF (%)
55	*TET2*	4:106158481	c.3382dup	p.Tyr1128LeufsXaa2	Frameshift ins	16.68
55	*TET2*	4:106162529	c.3443A>G	p.Tyr1148Cys	SNV; missense	17.29
56	*TET2*	4:106157914	c.2815C>T	p.Gln939Xaa	SNV; nonsense	7.63
118	*TET2*	4:106190831	c.4109G>A	p.Gly1370Glu	SNV; missense	6.15
180	*TET2*	4:106155238	c.141_150del	p.Val48ThrfsXaa16	Frameshift del	6.79
203	*TET2*	4:106164080	c.3590A>G	p.Lys1197Arg	SNV; missense	5.97
55	*ASXL1*	20:31023440	c.2925T>A	p.Cys975Xaa	SNV; nonsense	13.61
55	*ASXL1*	20:31022442	c.1934dup	p.Gly646TrpfsXaa12	Frameshift ins	17.73
67	*ASXL1*	20:31022442	c.1934dup	p.Gly646TrpfsXaa12	Frameshift ins	9.75
76	*ASXL1*	20:31022903	c.2388G>A	p.Trp796Xaa	SNV; nonsense	23.25
89	*TP53*	17:7578206	c.643A>G	p.Ser215Gly	SNV; missense	14.5
128	*TP53*	17:7577568	c.713G>A	p.Cys238Tyr	SNV; missense	6.67
128	*TP53*	17:7579515	c.169_172del	p.Asp57GlnfsXaa65	Frameshift del	7.43
81	*SF3B1*	2:198267359	c.1998G>C	p.Lys666Asn	SNV; missense	5.77
118	*SF3B1*	2:198267359	c.1998G>C	p.Lys666Asn	SNV; missense	5.44
114	*SETBP1*	18:42531907	c.2602G>A	p.Asp868Asn	SNV; missense	36.44
128	*U2AF1*	21:44524456	c.101C>T	p.Ser34Phe	SNV; missense	8.5

Supplemental Figure 1. Longitudinal mutational analyses (A) Waterfall plot of driver mutation change in VAF for each patient at 12months; median change 15.3% (0-400%) and (B) driver mutation change in VAF from 12m to latest time point; median 21.6% (0-389%). (C) BAT-treated patient achieving a JAK2 V617F CMR at 12 months; VAF 22 to 0%. Subsequently, they had a loss of CMR at 44 months with JAK2 V617F VAF 4% coinciding emergence of low level TET2 mutation. (D) RUX-treated patient achieving a CALR PMR at 12 months; VAF 65 to 9%. Subsequently, they had a loss of PMR with a CALR VAF 44% at 60 months and antecedent to this, an ASXL1 mutation emerged at 56 months. Notably, this patient switched from RUX at 20 months due to toxicity. BAT=best available therapy; CMR=complete molecular response; VAF=variant allele frequency; RUX=ruxolitinib; PMR=partial molecular response; TE=trial entry; n=number.

Supplemental Figure 1.