Supplementary Information for

Parallel Evolution of Leukemic Clones in Myeloproliferative Neoplasms

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The PDF file includes:

Figures S1 to S8
Tables S1 to S2

Other Supplementary Materials for this manuscript include the following:

Data S1

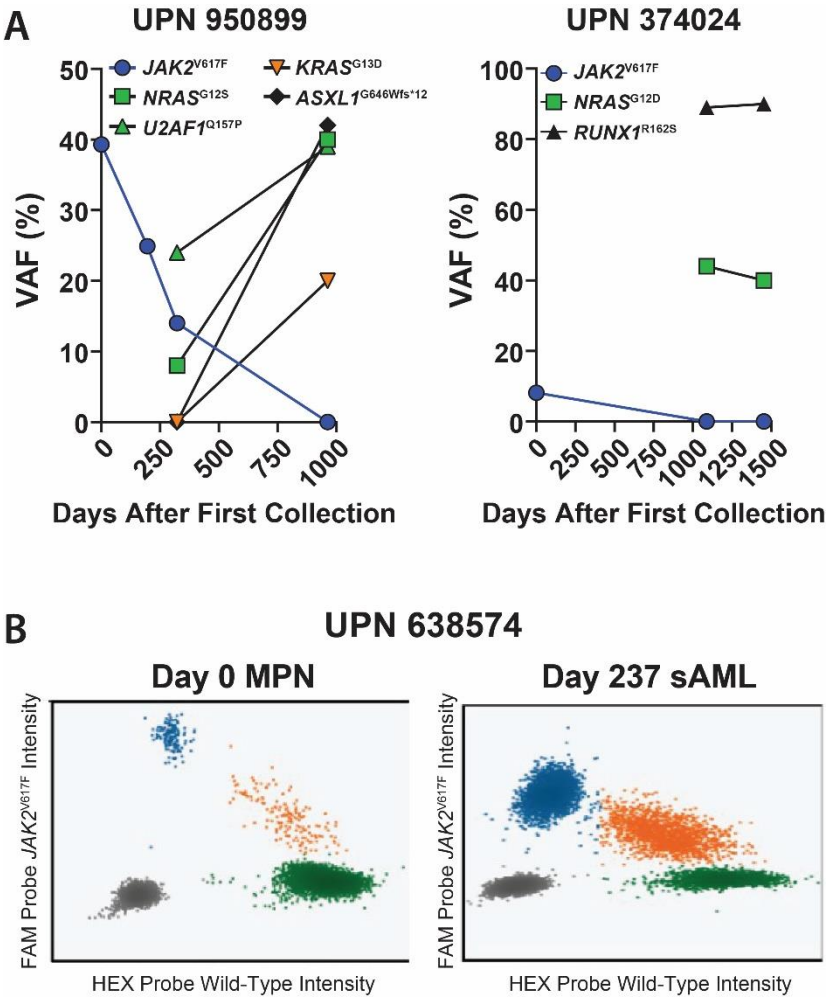

Figure S1.

(A) Variant allele frequencies (VAF) of mutations identified from clinical sequencing for indicated patients. **(B)** ddPCR quantification of *JAK2*^{V617F} mutant burden at MPN and sAML collection timepoints for UPN:638574.

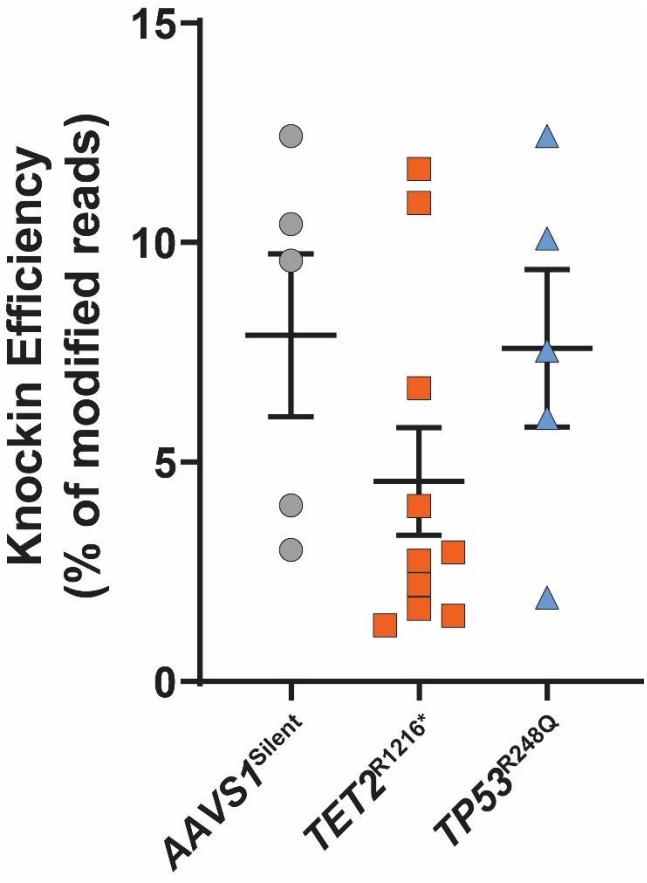

Figure S2.
Starting CRISPR/Cas9 knock-in efficiencies of indicated mutations in cord blood $CD34^+$ cells determined by next-generation sequencing.

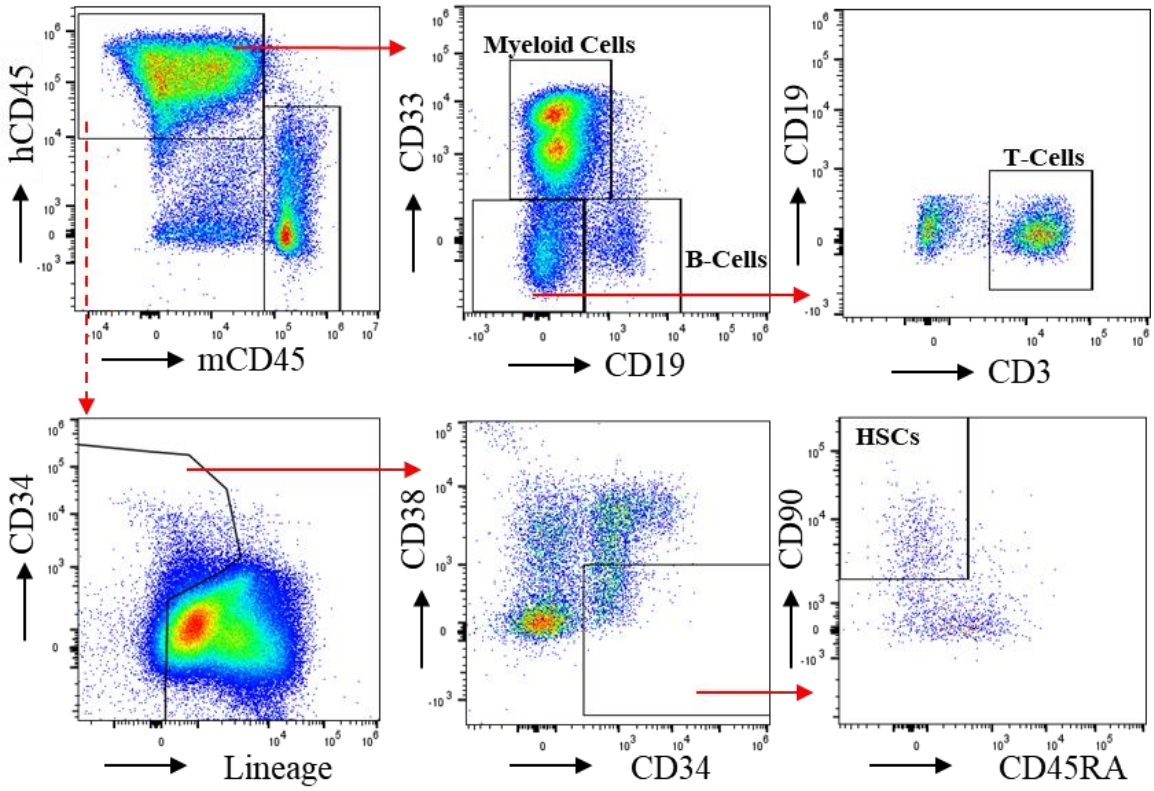

Figure S3.
Representative flow cytometry gating scheme of live singlets to identify human cell engraftment, blood cell lineage and HSC populations in PDX experiments.

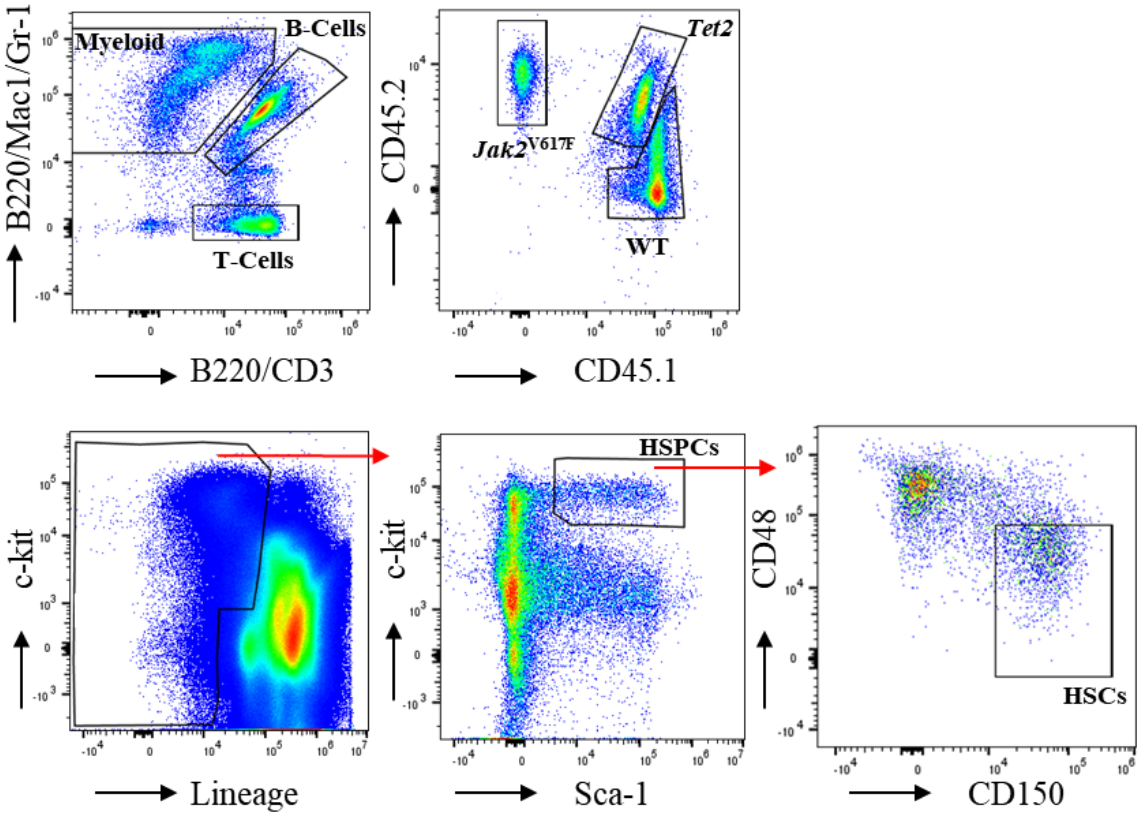

Figure S4.
Representative flow cytometry gating scheme of live singlets to identify (top row) mouse peripheral blood cell lineage distribution and donor-derived chimerism and (bottom row) discrimination of mouse HSPC and HSC populations in BM of recipient mice.

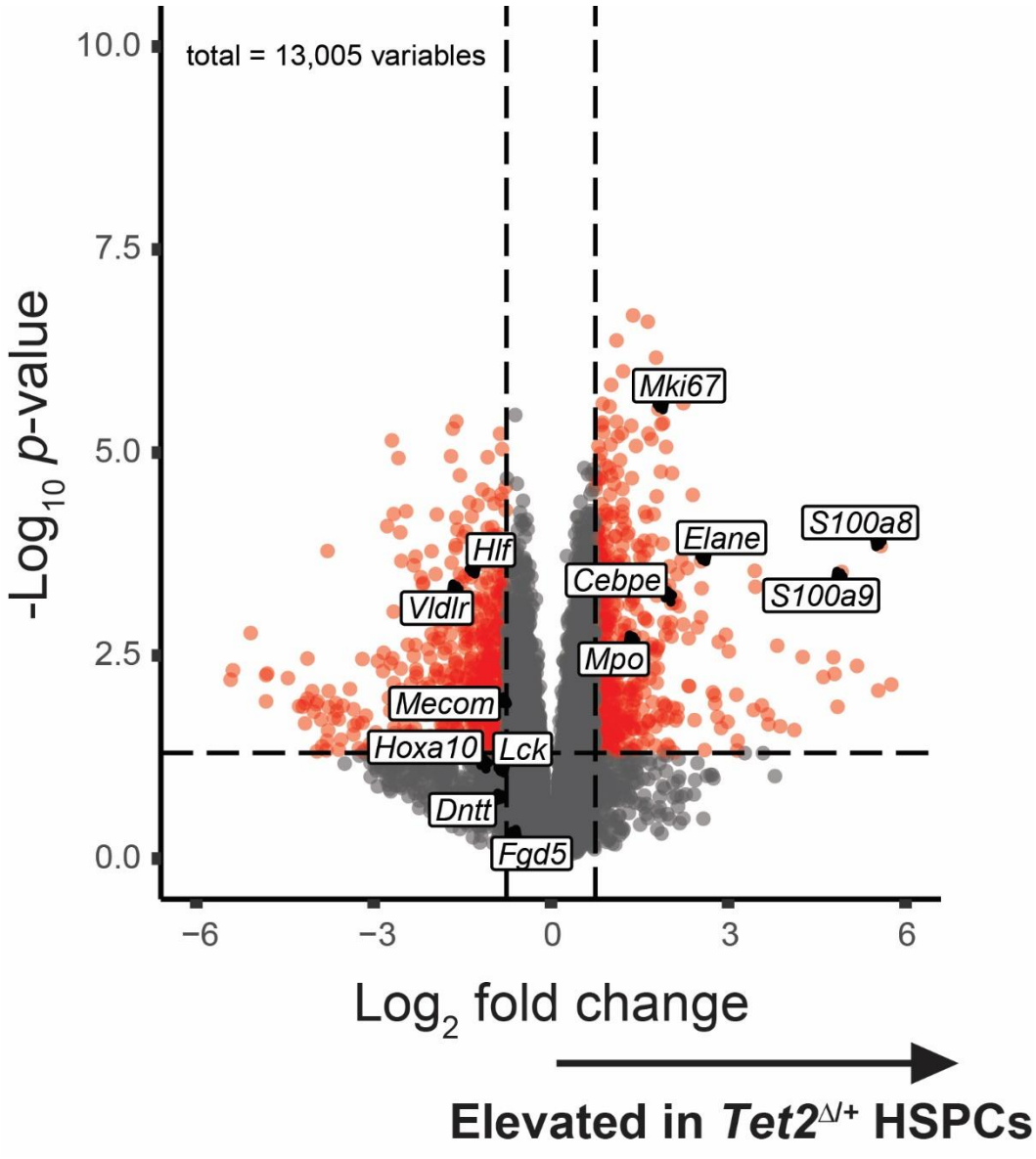

Figure S5.

Volcano plot depicting differentially expressed genes between *Tet2*$^{\Delta/+}$ HSPCs from a *Jak2*^{V617F} environment compared to WT HSPCs from a *Jak2*^{V617F} environment.

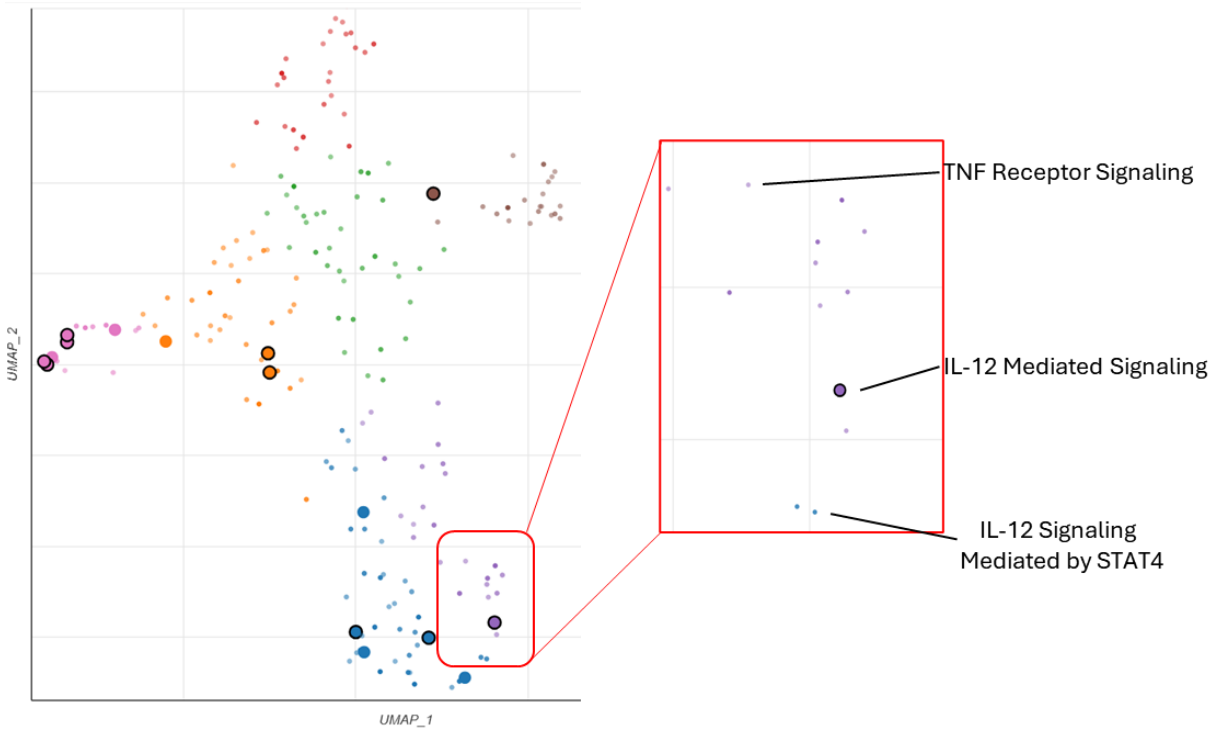

Figure S6.

UMAP visualization of relationships between enriched cancer-related pathways determined by input list of differentially expressed genes between *Tet2*$^{\Delta/+}$ HSPCs cells from a *Jak2*^{V617F} environment compared to WT HSPCs form a *Jak2*^{V617F} environment. Terms with more similar gene sets are positioned closer together. The darker and larger the point, the more significantly enriched the term.

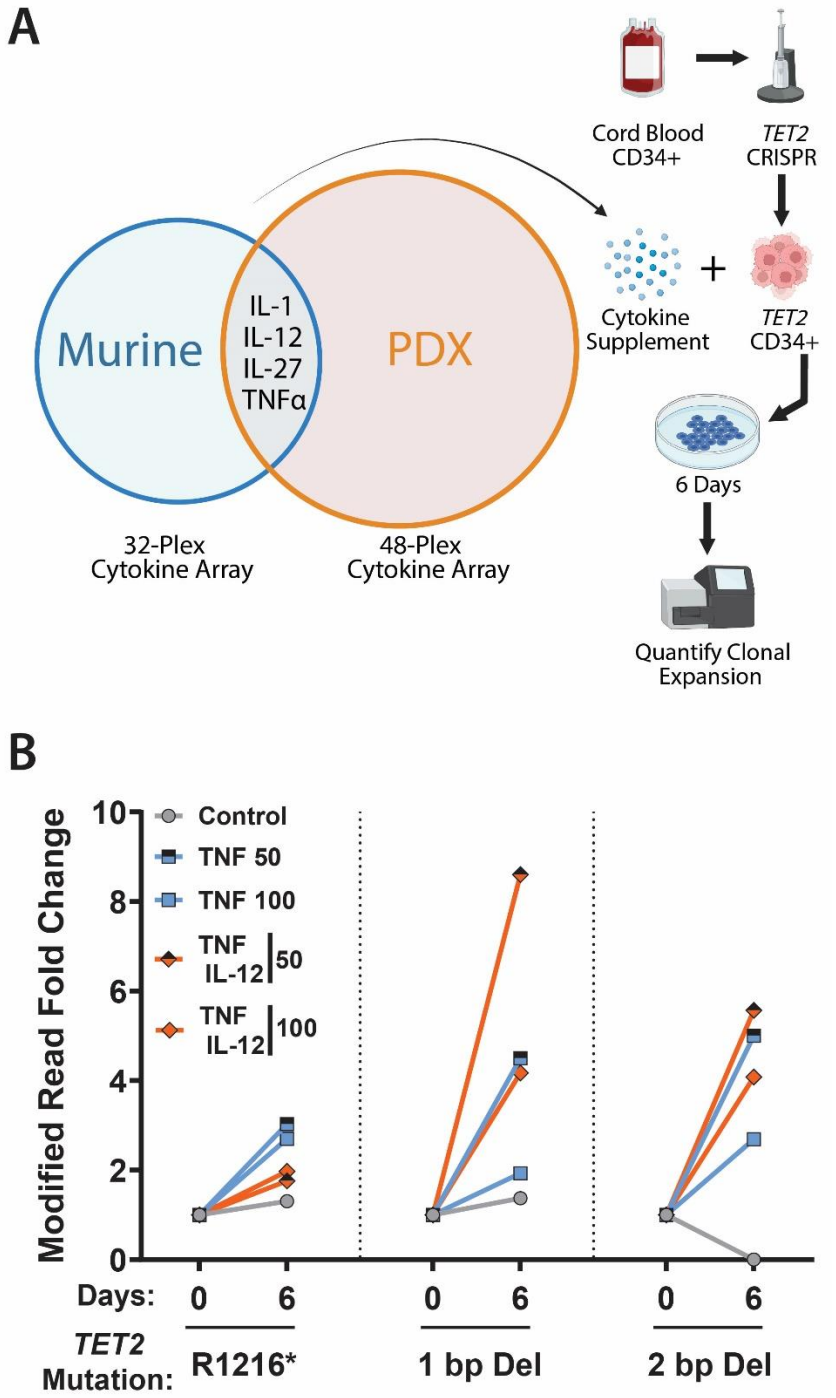

Figure S7.
(A) Schematic illustrating workflow for testing cytokines identified to correlate with *TET2*-mutant clone expansion from *in vivo* studies. **(B)** VAF quantification of tracked CRISPR-engineered *TET2* mutations in CB-derived CD34+ cells in media supplemented with indicated cytokines.

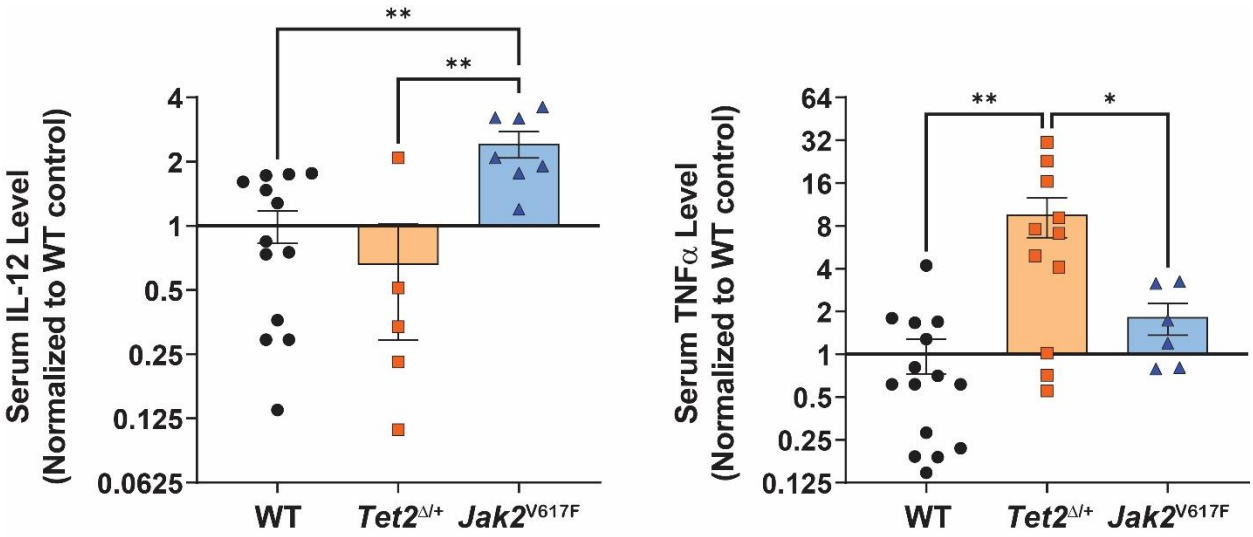

Figure S8.
Serum cytokine levels of IL-12 and TNFα from donor WT, *Tet2*$^{\Delta/+}$ and *Jak2*^{V617F} mice normalized to average levels of WT mice.

Figure	UPN	Age	Sex	Dx	*JAK2*^{V617F} VAF	Other Variants	Treatments
1	950899	73	M	MF	25%	None Reported	Ruxolitinib + Aspirin
1	950899	75	M	sAML	ND	U2AF1, NRAS, KRAS	Transfusion
1	374024	75	M	PV	8%	None Reported	Aspirin
1	374024	79	M	sAML	ND	NRAS	Aranesp
1	638574	59	M	MF	Positive	None Reported	N/A
1	638574	60	M	sAML	ND	RUNX1, PHF6	N/A
1	101811	78	F	PV	40%	DNMT3A, PPM1D	Hydrea + Aspirin + Intermittent phlebotomy
1	867898	64	F	MF	40%	None Reported	
2 / 4	117987	38	M	PV	5%	None Reported	Aspirin + Intermittent phlebotomy
2 / 4	867898	64	F	MF	40%	None Reported	Momelotinib + Aspirin
2, 4, 5	702759	26	F	PV	2%	None Reported	Aspirin
2 / 4	172431	76	F	MF	86%	BCOR	Ruxolitinib + Retacrit
2 / 4	899567	58	F	PV	88%	None Reported	Hydrea + Aspirin + Intermittent phlebotomy
2 / 4	497757	55	F	MF	47%	None Reported	Ruxolitinib + Aspirin
2 / 4	603873	49	M	PV	5%	None Reported	Aspirin + Intermittent phlebotomy
2 / 4	523915	73	M	MF	10%	ASXL1, CBL, EZH2, SETBP1	Ruxolitinib

Table S1.

Clinical characteristics from patient samples used throughout the study. All MPN samples contained a *JAK2*^{V617F} driver mutation.

N/A = data not available.

ASXL1	*ERG*	*KDM6A*	*NRAS*	*SMC1A*
ATM	*ETV6*	*KIT*	*PHF6*	*SMC3*
BCOR	*EZH2*	*KMT2A*	*PPM1D*	*STAG2*
BRAF	*FLT3*	*KRAS*	*PTEN*	*STAT3*
CALR	*GATA2*	*MPL*	*PTPN11*	*TET2*
CBL	*GNAS*	*MYC*	*RAD21*	*TP53*
CHEK2	*IDH1*	*MYD88*	*RUNX1*	*U2AF1*
CSF3R	*IDH2*	*NF1*	*SETBP1*	*WT1*
DNMT3A	*JAK2*	*NPM1*	*SF3B1*	*ZRSR2*

Table S2.
MissionBio Tapestri Myeloid Panel targeting 45 genes with 312 amplicons over approximately 65kb of target space.

Data S1. (separate file)

Gene expression analysis of 1) *Tet2*$^{\Delta/+}$ HSPCs isolated from a *Jak2*^{V617F} background vs. *Tet2*$^{\Delta/+}$ HSPCs isolated from a WT background, 2) *Tet2*$^{\Delta/+}$ HSPCs isolated from a *Jak2*^{V617F} background vs. WT HSPCs isolated from a *Jak2*^{V617F} background, and 3) WT HSPCs isolated from a *Jak2*^{V617F} background vs. WT HSPCs isolated from a WT background.