

SUPPLEMENTAL MATERIAL

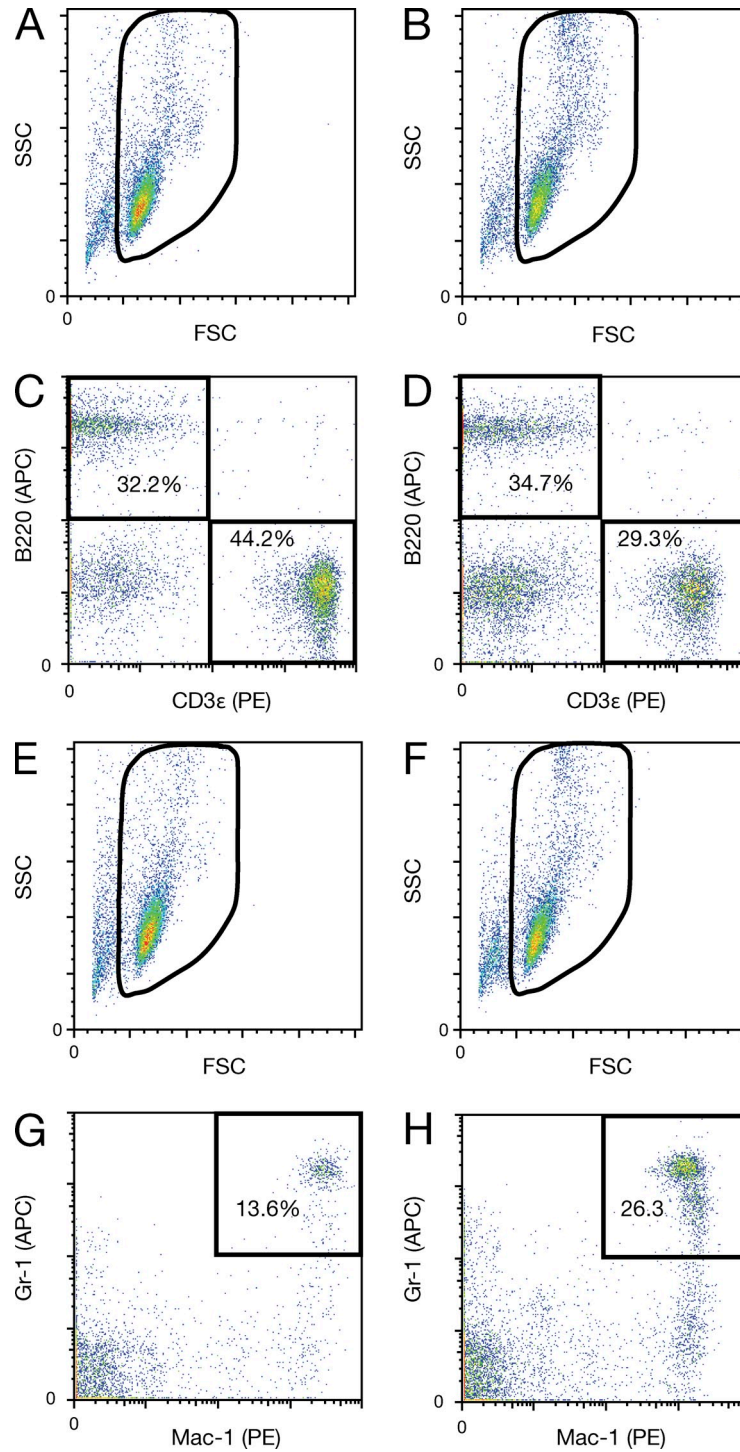
Jutzi et al., <http://www.jem.org/cgi/content/full/jem.20120521/DC1>

Figure S1. Quantification of peripheral blood leukocytes (granulocytes and B and T cells) in PB of transplanted mice. (A–H) Gating strategies. (A, C, E, and G) Representative mouse expressing WT NF-E2. (B, D, F, and H) Representative mouse expressing the NF-E2-262aa truncation. (A and B) Forward scatter (FSC) and sideward scatter (SSC) plots. Live cells are gated. (C and D) B and T cells defined by B220 and CD3 ϵ positivity, respectively. (E and F) Forward scatter (FSC) and sideward scatter (SSC) plots. Live cells are gated. (G and H) Granulocytic cells defined by Gr-1 and Mac-1 double positivity.

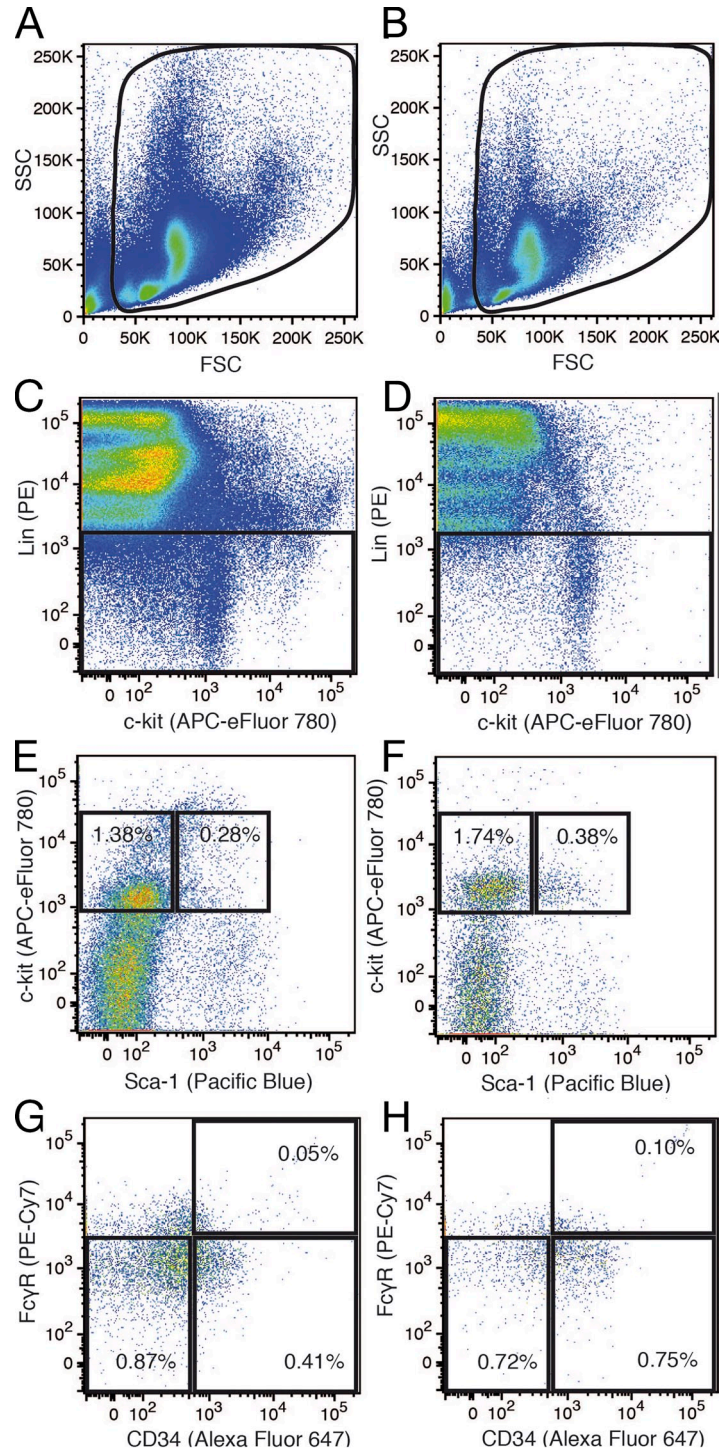


Figure S2. Quantification of hematopoietic stem and precursor cells (KL, KSL, CMP, GMP, and MEP) in BM of transplanted mice. (A–H) Gating strategies. Positively staining cells as well as negative cells were determined by fluorescence-minus-one (FMO) stainings, as previously published (Kaufmann et al., 2012). (A, C, E, and G) Representative mouse expressing WT NF-E2. (B, D, F, and H) Representative mouse expressing the NF-E2-262aa truncation. (A and B) Forward scatter (FSC) and sideward scatter (SSC) plots. Live cells are gated. (C and D) Lineage-negative cells. (E and F) Kit-positive lineage-negative (KL) cells and kit-positive sca-positive lineage-negative (KSL) cells are shown. (G and H) CMP, GMP, and MEP cells are defined by Passequé et al. (2005).

Table S1. Clinical characteristics of the MPN MDS, sAML, and CMML patients studied

Diagnosis	Number of patients	Gender m/f	Mean age at diagnosis (range)	Mean duration of disease (range)	Hemoglobin (at sample)	WBC (at sample)	Platelets (at sample)
			<i>yr</i>	<i>yr</i>			
ET	120	55/65	49.7 (17.3–82.3)	4.9 (0.0–21.5)	14.1 (8.6–33.9)	8.1 (2.5–31.4)	671 (149–2,393)
PV	144	71/73	55.5 (16.7–78.4)	5.3 (0.0–29.3)	14.6 (7.9–20.3)	15.2 (1.8–209.0)	513 (40–2,055)
PMF	192	102/90	56.9 (21.3–86.4)	3.5 (0.0–20.5)	10.7 (1.2–17.2)	15.1 (0.8–102.1)	292 (99–593)
MPN-U	49	23/26	71.0 (34.3–87)	n.d.	11.0 (5.8–20.0)	43.8 (5.8–185)	271 (12–1,027)
MDS	57	23/34	57 (23.7–82.7) ^a	3.0 (0.2–13.9) ^b	5.5 (2.1–8.1) ^c	4.7 (0.8–37.2) ^d	117 (9–517) ^e
sAML (post MPN)	39	21/18	57.0 (42.2–76.0)	7.0 (0.0–20.9)	9.7 (6.9–15.5)	39.2 (1.5–182.4)	83 (11–362)
CMML	67	41/26	71.2 (23.9–85.5)	n.d.	11.2 (6.6–16.5)	22.9 (2.6–91.4)	131 (14–711)

All patients were diagnosed according to the WHO criteria (Swerdlow et al., 2008). Complete blood count was taken at the time the blood sample was obtained. Abbreviations used and data displayed: MPN-U, MPN-unclassified; MDS, myelodysplastic syndrome; sAML, secondary AML; CMML, chronic myelomonocytic leukemia; WBC, WBC count.

^aData missing in 3 cases.

^bData missing in 18 cases.

^cData missing in 2 cases.

^dData missing in 2 cases.

^eData missing in 3 cases.

Table S2. Clinical features of MPN patients carrying NF-E2 mutations

UPN	Diagnosis	Age at diagnosis	Duration of disease at sample	Hemoglobin at sample	WBC at sample	Platelet count at sample	Status
			<i>yr</i>	<i>g/dl</i>	<i>10⁹/liter</i>	<i>10⁹/liter</i>	
202	PV	47.0	15.2	12.8	13.4	421	Alive
209	PV	49.1	10.1	13.4	18.9	412	Deceased ^a
241	Post PV-MF	44.2	20	9.4	6.4	156	Alive
409	PV	69.6	11.6	12.6	5.0	458	Alive
442	Post PV-MF	53.7	0	9.3	6.8	240	Alive
532	PMF	59.9	9.2	12.0	9.9	99.0	Alive
980	Post ET-MF	73.2	9.0	11.3	3.8	593	Alive
2,836	Post ET-MF	81.7	0	13.3	7.2	702	Deceased ^b

^aCause of death: sepsis following ERCP (endoscopic retrograde cholangiopancreatography).

^bCause of death unknown.

Table S3. *NF-E2* variants detected in MPN patients and healthy controls

UPN	Diagnosis	Variant cDNA NM_006163.1	Variant protein	Genetics
245	PMF	c.1094C>G	p.R365P	n.d.
311	PV	c.631C>T	p.P211S	Constitutive
401	ET	c.359T>C	p.L120P	n.d.
405	PV	c.677A>G	p.D226G	n.d.
445	Post PV-MF	c.1094C>G	p.R365P	Constitutive
713	PV	c.1093C>T	p.R365W	n.d.
727	PMF	c.1094C>G	p.R365P	Constitutive
806	PV	c.913C>T	p.R305W	n.d.
945	PMF	c.1094C>G	p.R365P	Constitutive
1,136	PV	c.1094C>G	p.R365P	n.d.
1,455	ET	c.758T>A	p.L253Q	n.d.
2,618	PV	c.1094C>G	p.R365P	n.d.
	Healthy control	c.1094C>G	p.R365P	n.d.
	Healthy control	c.C1093T	p.R365W	n.d.

n.d., not determined.