Supplemental Figure Legends:

Supplemental Figure 1: Schematic showing the structure of the C/EBPα mRNA. The C/EBPα gene is intronless and generates two isoforms as a result of the differential utilization of alternate translation start codons. The resultant p42kD (full length) and p30kD (truncated) C/EBPα proteins differ from each other at the N-terminus. Translational control of C/EBPα isoform expression occurs via a conserved cis-regulatory uORF (upstream open reading frame) in the 5'UTR (untranslated region) that is out of frame with the coding region of C/EBPα and is thought to be responsive to the activities of the translation initiation factors eIF4E and eIF2. When levels of these factors are high, the out-of-frame uORF (red AUG) is translated, but termination of its translation very close to the translational start site (AUG) for p42 is thought to prevent re-initiation at the p42 AUG. Instead, ribosomes continue to scan and reinitiate at a downstream AUG, resulting in the expression of C/EBPα p30. In contrast, under basal conditions, when levels of the initiation factors are relatively low, most ribosomes do not initiate translation at the uORF but instead initiate translation at the p42 AUG by a process involving "leaky ribosome scanning", resulting in translation of the full length C/EBPα p42 isoform. The translation initiation AUG codon of a putative shorter C/EBPα isoform shown in Figure 4C is also indicated (see dashed red arrow). TEI, II and III are activation domains, bzip: basic-leucine zipper DNA binding domain.

Supplemental Figure 2: Expression levels of C/EBPα p30 and p42 in HEK293 cells. A. Expression plasmids for C/EBPα p30 and p42 were transfected into HEK293 cells. 24 hours post- transfection cell lysates were prepared and subjected to western blot analysis. The blot was probed sequentially for C/EBPα N-terminal and C-terminal recognizing antibodies and β-actin. Note: C/EBPαp30 was recognized only when the C/EBPα C-terminus antibody was used but not with the N-terminal antibody. C/EBPαp42 was recognized by both antibodies.

Supplemental Figure 3: Transient transfection analysis of a C/EBP-Luciferase reporter (2x) with expression plasmids for C/EBPα p42 and C/EBPα p30 in HEK293T cells. Transient transfection analysis was conducted using a C/EBP-reporter plasmid (2x) harboring tandem C/EBP sites and C/EBPα expression plasmids. HEK293T cells were transiently cotransfected with 2x and expression plasmids for C/EBPα p42 (wild-type) alone or with increasing concentration of C/EBPα p30 (truncated). Normalized luciferase (to that of a cotransfected β- galactosidase expression plasmid) values have been represented as a fold change over the enzyme activity of 2x reporter plasmid alone (equal to 1). The figure represents normalized mean+/- s.e. obtained from an experiment performed in triplicate. Increasing concentrations of the p30 isoform significantly reduced the transactivation activity of the wildtype isoform.

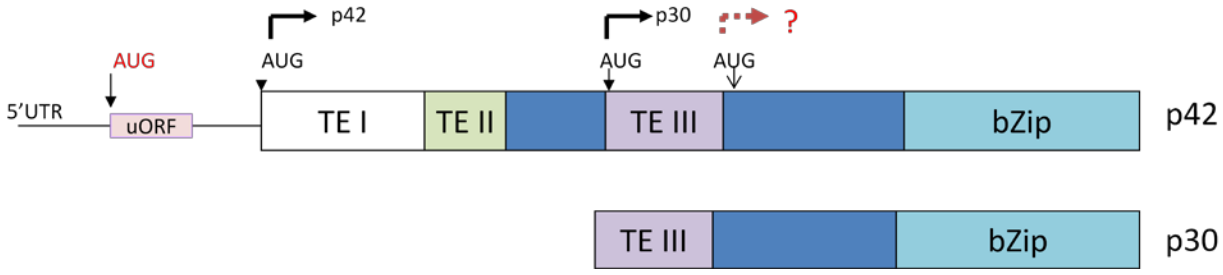
Supplemental Figure 4: Expression levels of GM-CSFR and G-CSFR in MNPM1$^{+/+}$ and $^{+/-}$ cells.
Expression levels of GM-CSFR and G-CSFR were measured using real time PCR analysis using cDNA derived from total RNA from uninduced and 48 hour GM-CSF-induced MNPM1$^{+/+}$ and MNPM1$^{+/-}$ cell lines. Expression levels were normalized to that of β actin, calculated by the delta-delta Ct method and represented as a fold change over the expression in NPM1$^{+/+}$ cells. The expression of each gene was measured in triplicate and S.E. values have been indicated.
While expression levels of the GM-CSFR remained unchanged between the two cell lines, levels of the G-CSFR were reduced in NPM$^{+/-}$ +GM-CSF treated cells.

Supplemental Figure 5: Adding an NPM1 expression plasmid back into NPM1 haploinsufficient cells restores neutrophil specific gene expression. MNPM1$^{+/-}$ cells were nucleofected with empty vector or an NPM1 expression plasmid. 48 hours post-transfection, total RNA was isolated, converted to cDNA and subjected to qRT-PCR analysis using gene specific oligomers. Expression levels have been normalized to that of β-actin and calculated using the delta-delta Ct method. Fold change in expression

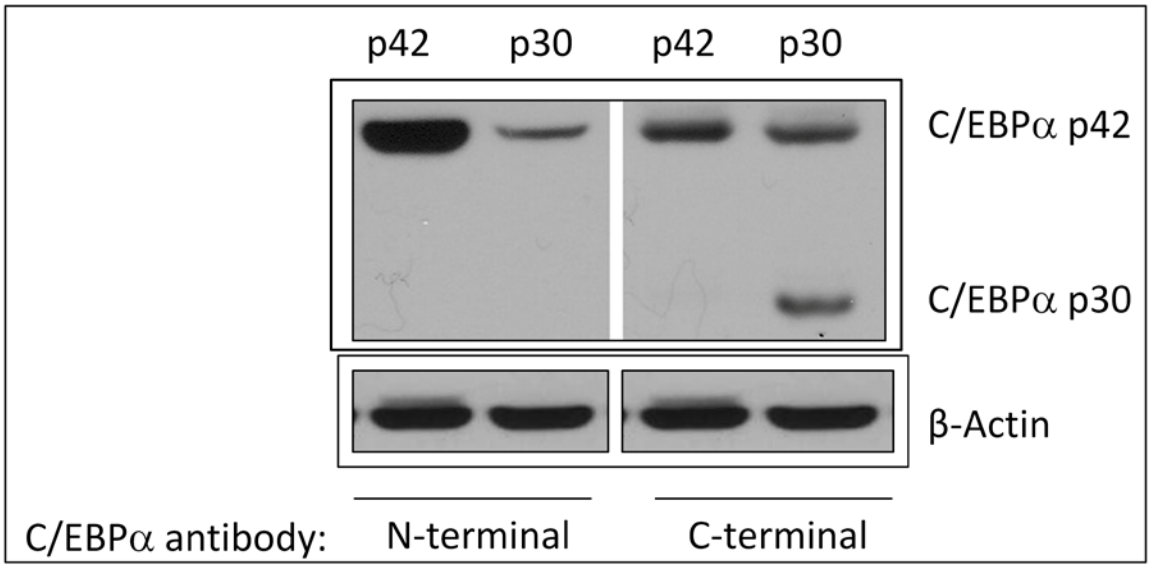
has been calculated for each gene by assuming its expression in vector transfected cells is 1. Note: that NPM1 reexpression restores expression of the same myeloid genes as C/EBPαp42 (Figure 5).

Supplemental Table 1: Sequences of Oligonucleotides used in q-RT-PCR analysis.

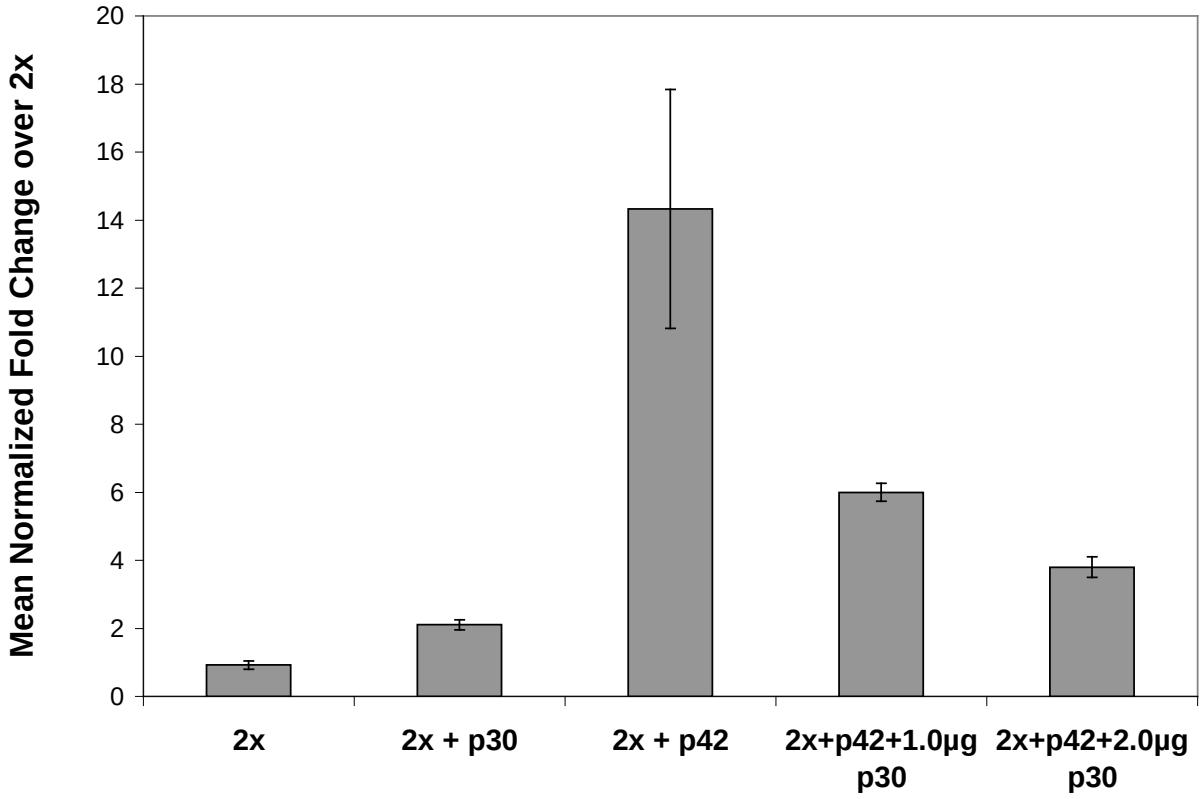
Supplemental Figure 1



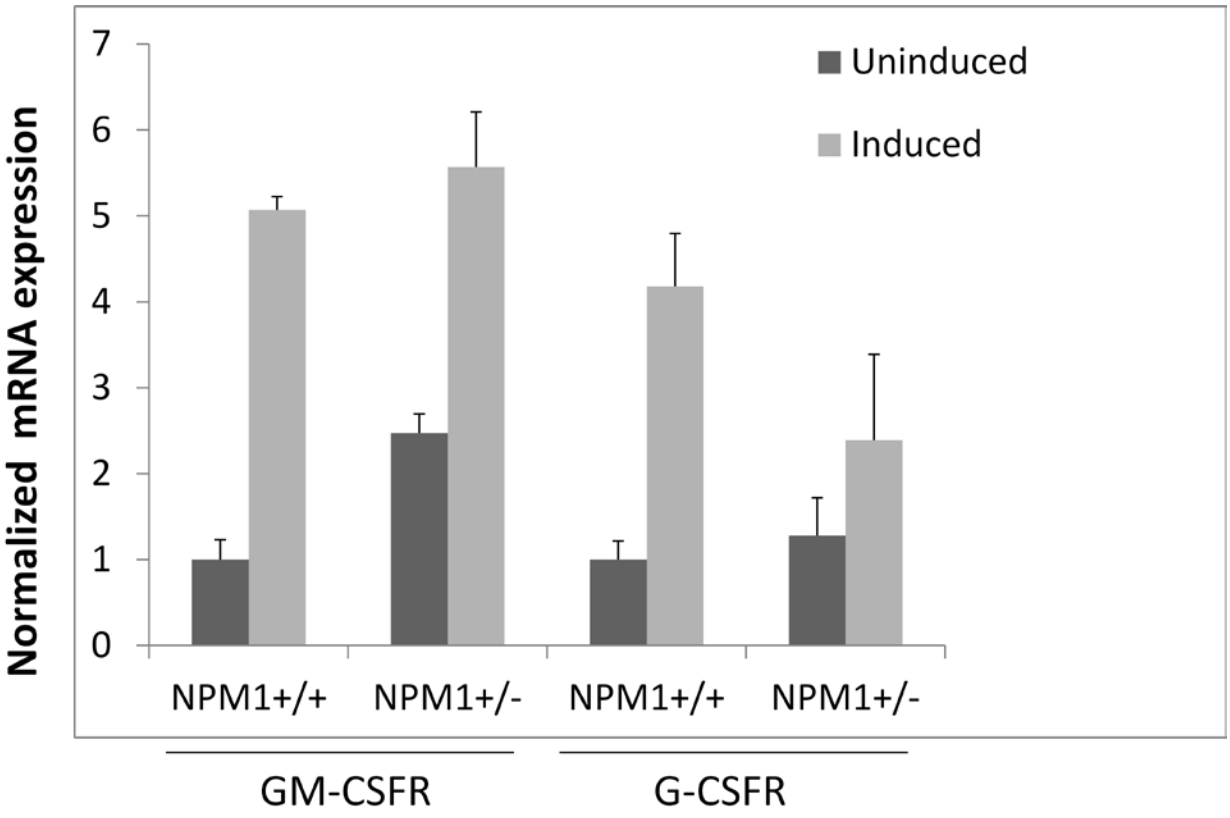
Supplemental Figure 2



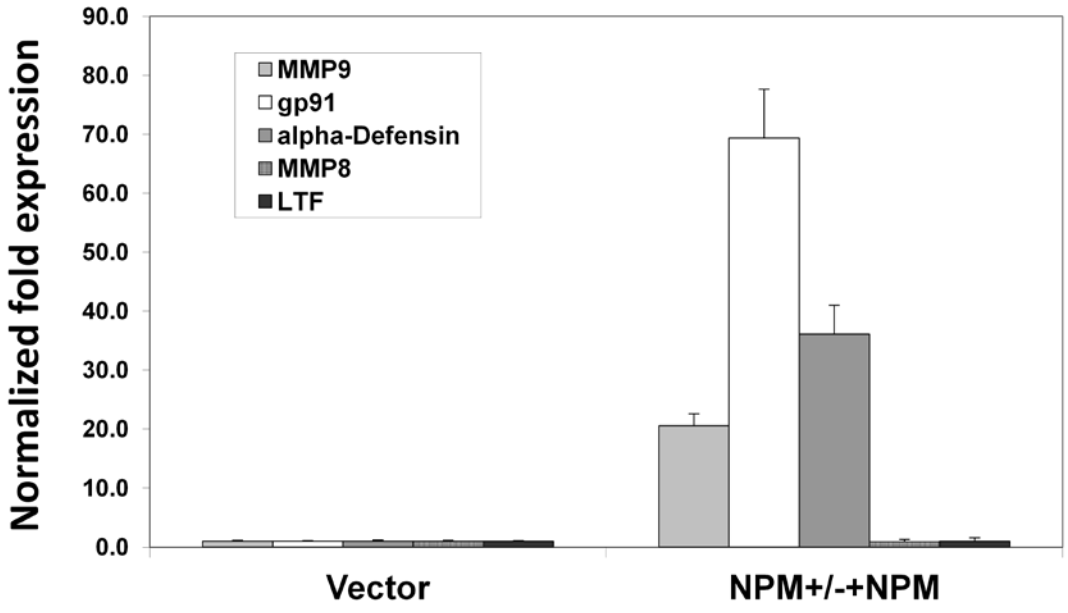
Supplemental Figure 3:



Supplemental Figure 4:



Supplemental Figure 5



Supplemental Table 1

Name	Sense	Anti-sense
NPM	5′ GGGGCTTTGAAATTACACCA 3′	5′ GAGCAGATCGCTTTCCAGAC 3′
c/EBPα	5′TGGACAAGAACAGCAACGAG3′	5′CCTTGACCAAGGAGCTCTCA3′
c/EBPα	5′GAGTCGGCCGACTTC3′	5′AAGGCGGCCGGGTCGATGT3′
c/EBPε	5′ TCTCCGCCTACATCGAGTCT 3′	5′ CCTATCTGGGCCAAATGTGT 3′
LACTOFERRIN	5′ GGAGCCTTGAGGTGTCTGAG 3′	5′ CCAGGTGGCACTCCTTGTAT 3′
DEFENSIN	5′ CCAGGTCCAGGCTGATTCTA 3′	5′ CCTTTCTGCAGGTTCCATTC 3′
MMP9	5′ CATTCGCGTGGATAAGGAGT 3′	5′ ACCTGGTTCACCTCATGGTC 3′
MMP8	5′ AACGGTCTTCAGGCTGCTTA 3′	5′ AATGGCTTGGACACTCCTTG 3′
gp91Phox	5′ GACTGCGGAGAGTTTGGAAG 3′	5′ GGTGATGACCACCTTTTGCT 3′
GMCSFR	5′CACCGCGTCCTGTAACTCTT3′	5′GCACCTTGACCTTGTGACCT3′
GCSFR	5′GAGCTGTGGACACATCGAGA3	5′AGGAAGGCCTGGGTGTAGTT3′
β-ACTIN	5′ GTGGGCCGCTCTAGGCACCA 3′	5′CGGTTGGCCTTAGGGTTCAGGGGGG 3′

Supplemental Table 1