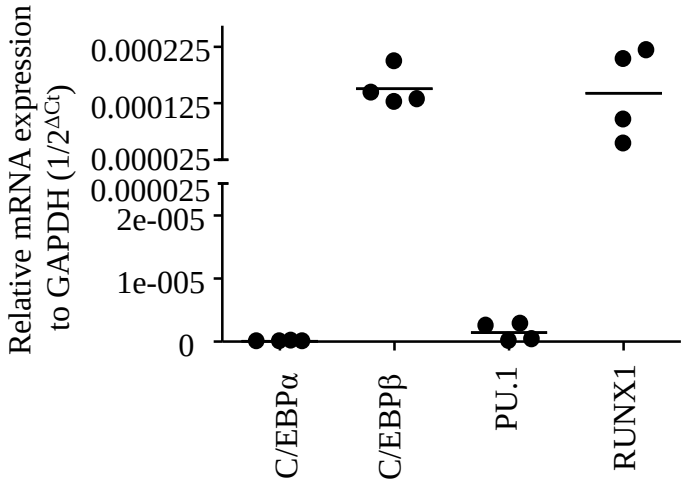
Supplementary Table 1. List of sequences of qPCR primers, guide RNAs, CRISPR validation primers, 3C analysis primers and restriction enzyme digestion efficiency primer.

Experiment	Target	Primer Sequence
qPCR/ddPCR	GAPDH	F: GCATTGTGGAAGGGCTCATG R: TTGCTGTTGAAGTCGCAGGAG
	M-CSFR	F: GAGGTGTCTGTGGGTGACAG R: CCTCTGAGGCCTTTACACGG
	IL-1β	F: GTGGACCTTCCAGGATGAGG R: GCTTGGGATCCACACTCTCC
	577	F: TGGTCTTAGGTGACCCCTGT R: TAGCTGCTTCAGACACCTGTG
	318	F: GCTGTACCTCCATGGTTTGTG R: AGCAATGAAAATAATTTTAGGGTGT
	-78	F: GTTGTGAAATCAGTTAACCCAAG R: GAGACTTACTTGCACAAGGAAG
	-321	F: ACTTAAAATGTACAGCTAACCCA R: GGAGAAGCTTGATGGGAATTC
	-534	F: CCTGACCCACACAAGGAAGT R: ATGTGCGGAACAAAGGTAGG
	-1251	F: TACTGCCTGCATCCATCTGC R: GGGAGCTCTTCTTGCTTGGA
	-2258	F: ATGTTGTGCAACTTGCCTGC R: AGGAGGTTTGTCTGGGAGGA
	-2860	F: ATGAGAGGGAAAGAACAGACCC R: GCTAAGCAATGACTGTCCTCA
	-3236	F: ACTTGGGGAGGAAAGGATGT R: ATGAGGAGCAAGCCAGTGAG
	-4152	F: AGTGCATGTTCCAACGTCAA R: GACCATCAAGAACAGCAGCA
	-5370	F: CTAGTCCCAGGGAGTTCTGC R: AGGGTTAGGCGCTATGGTCT
	-6212	F: CTATGGCCTATGGCTTCTGC R: TTTTGCCACATGGCTGATAA
	-7182	F: ACAGTCTCGCCACAGAAAGAA R: CCATCAAAAGGACAACTGCAT
	-7951	F: AATCACGAACAGACGACCATC

		R: GCCTCCCTATCTCCCTACCTT
	-8453	F: AGTAGTACCAGAGCCCCATGT R: GCTTCCCTTTGCATCTAGCA
	-9013	F: GGGTTTAAGGGTCTGGTCTTG R: CAGAAAGCTGGGAATTGGAG
	-9236	F: CATCAACCAGGATTGGACGTG R: GCACTGGGGATCCTATTAACC
	-9486	F: CGGGGAAGTGGCTGATAGTA R: TCAGGCTTCCTTCAGTGGAT
	-10368	F: ATGGAGCCCATCCCAGAG R: AGTTACCAGCAGGGCCACTC
	-10841	F: AGCCGGAGCTAAAATGGAGAC R: CCACCACCCCAAGGACTTATC
	-11743	F: AGACATTGCCCTCCAGATCC R: CTGGGGAAAAGATGGGCAAC
	-13252	F: CGCTTATGTTGGGAATTTGG R: TCACAGAAGCAGGCAAGATG
	-14256	F: CCCAGGAAAGTGACGTTGTT R: GACCTTGCTTCCACTCTTGC
	-15232	F: GGCCCAGGGAGTAGCTCTAT R: TGGAGGGGCTGAGAGTTCTA
CRISPR guide RNA construction sequences	-11843	F: CACCGAAATGCCAGCGCCCTGAGAG R: AAACCTCTCAGGGCGCTGGCATTTC
	-10833	F: CACCGACTACAACGCAGTAGTCAGT R: AAACACTGACTACTGCGTTGTAGTC
	-9824	F: CACCGGGACTTTCCAAATGAGTCGT R: AAACACGACTCATTTGGAAAGTCCC
3C analysis	TaqMan probe	5’-JOE/TCGTTCACCACC/ZEN/ TTTGCACTGTGCAAC/BkFQ-3’
	Constant primer	TGCTCATGAACAGGCAGATG
	Test primer #1	TTGTCTGGGAGGATTTGGAG
	Test primer #2	TCTGTAGGCAAGCCTGT
	Test primer #3	GATGCAAGTACCATGGGATG
	Test primer #4	AAAGGAAAGTGGTGTGTTTGTG
	Test primer #5	GCTGGTGGTTCTGGGTTCTA
	Test primer #6	AGGGCAACTTTGTGCAGATG

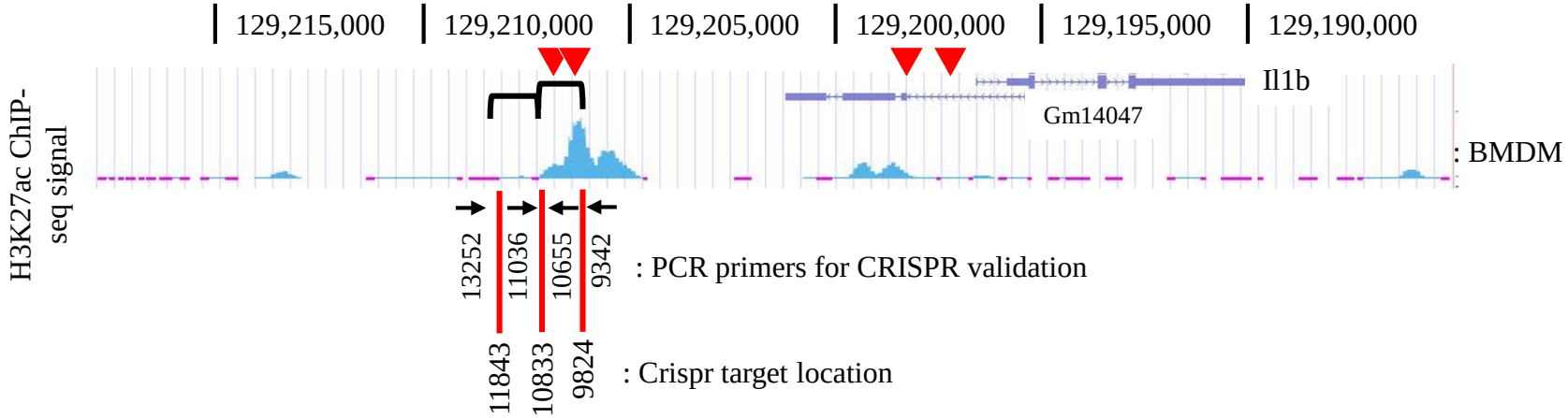
	Test primer #7	CCATCTCCTCACTCCCTTCC
	Test primer #8	GCCATCAAAAGGACAACTGC
	Test primer #9	CGACCATCAATGAGACCAAA
	Test primer #10	CTCTCCAGCACCCGTGAAT
	Test primer #11	AGACCAGACCCTTAAACCCT
	Test primer #12	TTCCGATTCACTTCCTCACC
	Test primer #13	TGCGTTGTAGTTGAAGCTGT
	Test primer #14	CTAACCCCTTCCAACACCT
	Test primer #15	GCTTACTCTGACTGCTTGCC
	Test primer #16	GTGTTCTCAGGCTGCCTTTC
	Undigested DNA examining primers	F: CCTGACCCACACAAGGAAGT R: ATGTGCGGAACAAAGGTAGG
	Digested DNA examining primers	F: TGCTCATGAACAGGCAGATG R: TATCCCTTTTCCAGGTCTCC



Supplemental Fig.1S. Expression level of C/EBPα, C/EBPβ, PU.1, RUNX1 in B16-BL6 melanoma cells were analyzed by RT-qPCR using Power SYBR Green PCR Master Mix and their expression level were compared to GAPDH housekeeping gene expression. Data are expressed as means and ±S.D. (n=4). Primers used were: C/EBP-α (forward , 5`-GAAGTCGGTGGACAAGAACA-3` and reverse 5`-AAGGAGCTCTC AGGCAGCTG-3`), C/EBP-β (forward ,5`-CAAGCTGAGCGACGAGTACA-3` and reverse 5`-AGCTGCTCCACCTTCTTCT G-3`), PU.1 (forward ,5`-CTGGAACAGATGCACGTCC-3` and reverse 5`-CTGGTACAGGCGAATCTTTTTC-3`), RUNX1 (forward ,5`-GCCAAGGATTTGCTCTTGAAG-3` and reverse 5`-TTGGATCTTTGGGGTACAGC-3`).

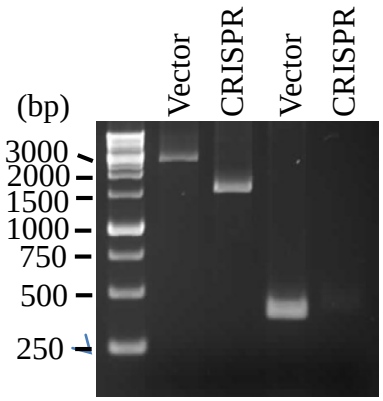
UCSC Genome: chr2: 129,218,460-129,185,345

▼ Putative PU.1 binding site (AG(or A)AGGAAGTG)



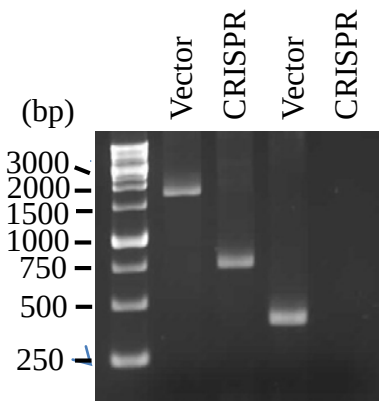
(A) eRNA deleted (Δ11843-10833)

Primer set: [13252(FW) & 10655(RV)] [11036(FW) & 10655(RV)]

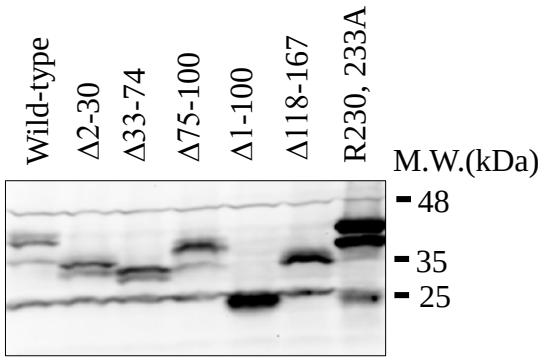


(B) PU.1 binding site deleted (Δ10833-9824)

Primer set: [11036(FW) & 9342(RV)] [11036(FW) & 10655(RV)]



Supplemental Fig.2S. Genetic deletion of the putative enhancer regions by a CRISPR/Cas9 system in B16-BL6 cells. Visual representation of the relative locations of CRISPR/Cas9-mediated cleavage sites and primers used to validate the deletion of the putative enhancer region in B16-BL6 cells (top panel). (A-B) Validation for Crispr-Δ eRNA (Δ11843-10833) and Crispr-Δ PU.1 (Δ10833-9824). Crispr-control, Crispr-Δ eRNA, and Crispr-Δ PU.1 cells were cultured and harvested, and genomic DNAs were prepared as described in "Materials and Methods". (A) 400 ng genomic DNAs which were prepared from Crispr-control, Crispr-Δ eRNA cells, were used for PCR as the templates. PCR were then carried out using two primer sets [13252 forward (5`- CGCTTATGTTGGGAATTTGG-3`) and 10655 reverse (5`-CCATTATCCATGCCTTGCTT-3`), 11036 forward (5`-GAGAGGAGAGCCTGGGAGTT-3`) and 10655 reverse (5`-CCATTATCCATGCCTTGCTT-3`)] for confirming deletion. The PCR products were analyzed on the agarose gel and visualized as described in "Materials and Methods". (B) Genomic DNAs were prepared from Crispr-control, Crispr-Δ PU.1 cells, and PCR were carried out as above (A) using following primers [11036 forward (5`-GAGAGGAGAGCCTGGGAGTT-3`) and 9342 reverse (5`-TCAGGCTTCCTTCAGTGGAT )primers, 11036 forward (5`-GAGAGGAGAGCCTGGGAGTT-3`) and 10655 reverse (5`-CCATTATCCATGCCTTGCTT-3`)]. The PCR products were analyzed and visualized as described in "Materials and Methods" and above (A).



Supplemental Fig.3S. B16-BL6 cells were transfected with MIG-PU.1 (wild type), MIG-PU.1 (Δ2-30), MIG-PU.1 (Δ33-74), MIG-PU.1 (Δ75-100), MIG-PU.1 (Δ1-100), MIG-PU.1 (Δ118-167) and DNA binding inactive PU.1 [pLVX-human PU.1 (R232, 235A)] plasmids for 48 hours, and cell lysates were prepared as described in "Materials and Methods". Expression of PU.1 proteins (wild type and truncated) were analyzed by immunoblotting using antibody raised from C-terminus of PU.1 protein. Representative immunoblot from three independent transfection is shown.