## **Supplementary Information**

# Diaphanous-related formin mDia2 regulates beta2 integrins to control hematopoietic stem and progenitor cell engraftment

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## **Supplementary Figures**



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#### Supplementary Figure 1. Loss of mDia2 doesn't affect the compositions of HSPCs at steady

**state** (a) Quantitative real time PCR analyses of the mRNA levels of mouse *Diap3* in the indicated lineages. 18S ribosomal RNA was used as an internal control. The experiment was repeated in triplicate. (b) Highly enriched *Diap3* expression in HSPCs obtained from Gene Expression Commons database <sup>1</sup>. (c) Representative flow cytometry plots for the gating of different HSPC populations in the bone marrow lineage negative cells from mDia2<sup>fl/fl</sup> Vav-Cre and mDia2<sup>fl/fl</sup> control mice at 3-month old. (d) Quantitative analyses of the percentage and absolute cell number of indicated HSPC populations in c. (e) Representative flow cytometry plots of cell cycle profiles of LSK cells from the indicated mice as in c. (f) Quantitative analyses cell cycle profiles of the LK and LSK cells from the bone marrow of indicated mice at 3-month old. *n*=4 mice in mDia2<sup>fl/fl</sup> group, *n*=6 mice in mDia2<sup>fl/fl</sup> Vav-Cre group in d and f. Data is presented as mean ± SEM. \**p* < 0.05, \*\**p* < 0.01, \*\*\**p* < 0.001, \*\*\*\* *p*<0.0001. Two-tailed unpaired Student's t test was used to generate the *p* values.



#### Supplementary Figure 2. Defects of competitive engraftment ability in mDia2 deficient HSPCs

(a) The percentages of bone marrow HSPCs in wild type recipient mice 10 months after transplantation with 2 x 10<sup>6</sup> control or mDia2<sup>fl/fl</sup>Vav-Cre BMMCs. Left panel, n=8 mice in mDia2<sup>fl/fl</sup> group, *n*=4 mice in mDia2<sup>fl/fl</sup> Vav-Cre group. Rght panel, *n*=4 mice in mDia2<sup>fl/fl</sup> group, *n*=2 mice in mDia2<sup>fl/fl</sup> Vav-Cre group. (b) Cell cycle profile of LSK cells from a. n=7 mice in mDia2<sup>fl/fl</sup> group, n=3mice in mDia2<sup>fl/fl</sup> Vav-Cre group. (c) The percentages of bone marrow HSPCs in wild type recipient mice 10 months after transplantation with 2 x  $10^6$  control or mDia2<sup>fl/fl</sup>Mx1-Cre BMMCs. (d) The absolute number of the indicated HSPCs in c. n=5 mice in mDia2<sup>fl/fl</sup> group, n=3 mice in mDia2<sup>fl/fl</sup> Vav-Cre group for c and d. (e) Experimental design of competitive bone marrow transplantation with plpC treated mDia2<sup>fl/fl</sup> and mDia2<sup>fl/fl</sup> Mx-Cre mice. (f) Chimerism studies in the peripheral blood 1.5 months after transplantation as in e. (g) Quantitative analyses of the indicated lineages in e. n=9 mice in each group. (h) Experimental design of secondary competitive transplantation using 2×10<sup>6</sup> BMMCs from the indicated recipients after primary transplantation with an equal number of wild-type CD45.1 BMMCs. cBMT represents competitive bone marrow transplantation. (i) Peripheral blood chimerism analyses were performed 1 month after competitive transplantation in h. n=3 mice in each group. Error bars represent the SEM of the mean. \*p < 0.05, \*\*p < 0.01, \*\*\*p < 0.001, \*\*\*\*p < 0.0001. Twotailed unpaired student's t test was used to generate the p values.



#### Supplementary Figure 3. Loss of mDia2 doesn't affect HSPC localization to the BM

**vasculature** (**a**) Quantitative analyses of the expression levels of CXCR4 by flow cytometry in the bone marrow LK and LSK cells from the indicated mice. n=6 in each group. (**b**) Quantitative analyses of the percentage of apoptotic cells in bone marrow LK and LSK cells from the indicated mice. n=3 in each group. (**c**) Relative proliferation rate of the bone marrow c-kit+ HSPCs from the indicated mice during ex vivo expansion. n=6 in mDia2<sup>fl/fl</sup> group, n=4 in mDia2<sup>fl/fl</sup> Vav-Cre group. (**d**) Experimental design of the homing assay.  $2x10^6$  BMMCs from plpC treated mDia2<sup>fl/fl</sup> or mDia2<sup>fl/fl</sup> Mx-Cre mice mixed with an equal number of CD45.1+ BMMCs from the wild type counterparts were injected into lethally irradiated wild type recipient mice. After 24 or 48 hours, BMMCs were obtained from the recipients for chimerism studies by flow cytometry. (**e**) Representative flow cytometry plots as described in d at 24 and 48 hours. (**f**) Quantitative analyses of e. n=3 in each group. All the error bars represent the SEM of the mean.



#### Supplementary Figure 4. Loss of mDia2 compromises HSPC's trans-endothelial migration (a)

Representative images of HSPC association with the vessels. Arrows point to the transplanted HSPCs. Dashed white lines outline the vessels. (**b**) *In vivo* imaging of fluorescently labeled lineage negative donor cells in the femur of the wild type mice 4 hours after non-irradiated competitive transplantation with equal number  $(9x10^5)$  of non-fluorescently labeled CD45.1+ cells. Red, CD31+ endothelial cells. Green, CD45.2+ donor cells. Dashed white lines outline the vessels. The relative position of the indicated donor cells to the outlined CD31+ lining sinusoids or arteries were quantified (right). N=39 cells from 6 random fields in mDia2<sup>fl/fl</sup> group. N=30 cells from 5 random fields in mDia2<sup>fl/fl</sup> Vav-Cre group. (**c**) Same as b except the spleen was analyzed. N=30 cells from 3 random fields in mDia2<sup>fl/fl</sup> or mDia2<sup>fl/fl</sup> vav-Cre CD45.2+ BMMCs were mixed with equal number of wild-type CD45.1+ competitive BMMCs and transplanted into lethally irradiated wild type mice (CD45.1+, *n*=5 mice in each group) through intrafemoral route. Bone marrow chimerism studies were performed 4 weeks after transplantation using flow cytometric analyses. Error bars represent the SEM of the mean. \**p* < 0.05, \*\**p* < 0.01. Two-tailed unpaired student's t test was used to generate the p values.



Supplementary Figure 5. mDia2-SRF signaling is involved in the regulation of HSPC engraftment (a) Immunofluorescence analyses of MAL and Iamin B1 in c-Kit+ HSPCs from the indicated mice incubated with or without FBS for 15 minutes. Repeated three times with similar results. (b) Relative mRNA expression levels of *ItgaM* and *Itgb2* in c-Kit+ HSPCs from the indicated mice incubated with or without FBS for 15 minutes. (c) Same as b except the indicated genes were analyzed. The experiments in b and c were performed in triplicate. (d-g) UCSC browser track showing the expression (green, RNA-Seq), DNA methylation (red), H3K36me3 peaks (dark blue), and H3K4me3 for transcription start sites (TSS, pink) of actively transcribed regions of the indicated genes in young (4 months) and old (24 months) HSCs. Data were obtained from aging HSC Epigenome <sup>2</sup>. Blue shaded regions represent the genomic regions of *ItgaM* and *Itgb2*. Error bars represent the SEM of the mean. \**p* < 0.05, \*\**p* < 0.01, \*\*\**p* < 0.001, \*\*\*\**p* < 0.0001. Two-tailed unpaired student's t test was used to generate the p values.



#### Supplementary Figure 6. Targeting SRF intronic binding sites in ItgaM loucs via CRISPR-Cas9

(a) Luciferase activity assay with reporter construct containing *ItgaM* intronic regions co-transfected with increased amount of SRF full length (SRF-FL), exon 5 deficient isoform (SRFΔ5), or constitutively active SRF-VP16. Experiment was performed in duplicate. (b) Same ChIP assay was performed as in Fig. 4h except Acta2 was analyzed by real-time PCR in guadruplicate. Error bars represent the SEM of the mean. (c) Schematic diagram showing the effect of (GGGGS)×3 spacer in luciferase reporter assay-based screening of sgRNA targeting of the intronic regions. The details were described in Fig. 6a and methods. (d) Quantitative analyses of the activities of recombinant luciferase harboring intronic DNA-encoded epitope without (upper) or with (lower) the (GGGGS)×3 spacer. sqLuc2P targeting luciferase gene was served as positive control. (GGGGS)×3 spacer largely reduced the inhibition of luciferase activity by intronic DNA encoded epitope. Experiments were performed in duplicate except the upper panel in triplicate. Repeated twice with similar results. (e-g) Determination of in vivo genome editing within ItgaM intron 2 region containing SRF binding element (CArG-like motif). The genomic DNA was isolated from the peripheral blood of the recipient mice and the target regions shown in the upper panel of e were PCR-amplified. The PCR products were cloned and subjected to sequencing to confirm the indels and estimate the editing frequencies. sgLuc2P, negative control, 100% (3/3) clones contain intact wide type sequence (lower panel in e); sgITGAM Intron 2 T7, 20% (1/5) clones contain a 7-bp nucleotide deletion occurred in the terminal region of CArG-like motif; 40% (2/5) clones contain a 361-bp deletion. 40% (2/5) clones contain a 193-bp deletion combined with a 19-bp insertion. They all lack the CArG-like motif (f); sqITGAM Intron 2 T26, 42.9% (3/7) clones contain a 644-bp deletion. 57.1% (4/7) clones contain a 439-bp deletion. Both lack the CArG-like motif (g).



**Supplementary Figure 7. Expression of SRF and mDia2 variants** (a) Ex vivo cultured c-Kit+ HSPCs transduced with indicated genes were examined for CD11a expression by flow cytometry. (b) Same as a except apoptosis was examined. *n*=2 mice in each group for a, b. (c) Stemness (c-kit<sup>high</sup>/Sca1<sup>high</sup>) was examined in ex vivo cultured c-Kit+ HSPCs. *n*=3 mice in each group. (d) Transduction efficiency of the indicated genes detected by flow cytometry of human CD4 co-expressed from the vector. *n*=3 mice in each group. (e) Quantitative analyses of the mRNA levels of SRF from indicated cells determined by quantitative RT-PCR. Experiment was performed in triplicate. (f) Transduction efficiency of the indicated constructs quantified by human CD4 expression through flow cytometry. *n*=5 mice in each group. Error bars represent the SEM of the mean. \**p* < 0.05, \*\**p* < 0.01, \*\*\**p* < 0.001, \*\*\*\**p* < 0.0001. Two-tailed unpaired student's t test was used to generate the p values.



**Supplementary Figure 8. Flow cytometric gating strategies** (a) Flow cytometric gating of LT-HSC,ST-HSC,MPP and SLAM-LSK for Fig. 1a, 5g and Supplementary Figure 1c-d, 2a, 2c-d. (b) Gating strategy for stem cell quiescence for Fig. 1d, 1e and Supplementary Figure 2b. (c-d) Gating routines for peripheral blood, bone marrow, spleen and HSPCs in competitive bone marrow transplantation as shown in Fig. 2b-c, 5e-g, 6h-i and Supplementary Figure 2h-i.

## Supplementary Tables

## Supplementary Table 1. Primer sequences for cloning, RT-PCR, qPCR and ChIP assays

ORF cloning into MICD4     SRF-F   AAA 6GATCC CCCCC     ATGTTACCGAGCCAAGCTGGGGCCGC   of mouse SRF ORF; BamH1 and EcoRI restriction     SRF-R   GGG GAATTC     SRF-R   GGG GAATTC     SRF-R   GGG GAATTC     CATTCACTCTTGGTCTTGGTGCTGGGGCGCC     Forward and reverse PCR primers containing Hpal     ATGGCTTCTACCTATCCTTATG     HA-SRF VP16-R   AAA 6CGGCCGC     TTACTACCCACCGTAC     Luciferase constructs cloning     CD14-Intron 6-R   ACCTAACTGGTGCACACATGGCG     COTTACTCTTAGCTTACGCTACCACATGGCG     COTTACTGTGTGTAAGCTTAACGTGAAGC     Comward orientation cloning by In-Fusion. 15bp     CD11b-Intron 2-R     TCTATGGTCTAAGCTTACACATGGCT     COTTACGTCTTGG     GCTACCTTCTG     GCTACCTTCTG     GCTTCCTTC     GCTTCCTTC     GCTTCCTTC     GCTTCCTTC     GCTCGTTGTG     GCTCCTTCT     GCTTCCTTC     GCTTCGTTGG     GCTTCGTTGG     GCTTCCTTC     GCTAGCTCGCCAGCTAGCACATGGCT     COTTAGTGTTAAGCTCAACATGGCT	Name	Sequence (5' - 3')	Purposes/Comments		
SRF-F   AAA GGATCC GCCGCC   Forward and reverse primers for PCR amplification ATGTTACCCAGCCAGCTGGGGCCCC     SRF-R   GGG GAATTC   sites are highlighted in gray     HA-SRF VP16-F   GGG GTTAAC GCCACC   Forward and reverse PCR primers containing Hpal Ant GCCCCC     HA-SRF VP16-R   AAA GCGCCCGCGTAC   Forward and reverse PCR primers containing Hpal Ant GCCCCCC     Luciferase constructs cloning   Constructs cloning   Forward and reverse primers for 730bp <i>ItgB2</i> intron A GCCACGTGGCTAGCACATGGACG     CD18-Intron 6-F   ACCTGACCTGGCTAGCACATGGACG   Forward and reverse primers for 730bp <i>ItgB2</i> intron A GCCACGTGGCTAGCCACATGGACC     CD18-Intron 6-F   ACCTGACCTGGCTAGCACATGGCG   Forward and reverse primers for 730bp <i>ItgB2</i> intron A GCTGCTGCTGC     CD18-Intron 6-F   ACCTGACCTGGCTAGCACATTGGC   Forward and reverse primers for 730bp <i>ItgB2</i> intron A GCTGCGCTGCCTAGCACACATGGCT     CD11b-Intron 2-F   ACCTGACCTGGCTAGCACATTGGC   Forward and reverse primers for 730bp <i>ItgB2</i> intron a GCTGCCTC     CD18 Intron 6-   ACCTGACCTGGCTAGCTACACATGGCT GCACGTGGCCACATGGGTGCA   Sorward and reverse primers for 730bp <i>ItgB2</i> intron a reverse orientation cloning by In-Fusion. 15bp GCTTCTTATGGCTGAGCTCACATGGTGCA     CD18 Intron 6-   ACCTGACCTGCTAGCACATGGGTGCA   Sorward and reverse primers for 746bp <i>ItgB2</i> intron A reverse orientation cloning by In-Fusion. 15bp GCTTCTTAGGCTGAGCTCACATGGTGCA	ORF cloning into MICD4				
ATGTTACC6AGCCAGCTGGGGCCCC   of mouse SRF ORF; BamH and EcoRI restriction     SRF-R   GG GAATTC   sites are highlighted in gray     TCATTCACTCTTGGTGCTGTGGGTGGC   Forward and reverse PCR primers containing Hpal     ATGGCTTCTAGCTATCCTTATG   and Notl restriction sites (gray).     HA-SRF VP16-R   AAA GCGGCCCGC     TTACTACCCACCGTAC   Forward and reverse PCR primers containing Hpal     AGCAACTG   forward orientation cloning by In-Fusion. 15bp     CD18-Intron 6-R   ACCTGACCTCGCTACCACATTGGC     CATCGTCTGTG   Forward onientation cloning by In-Fusion. 15bp     CD11b-Intron 2-R   ACCTGACCTCGCTACCACATTGGC     COT1b-Intron 2-R   ACCTGACCTCGCTACCACATGGCTC     CD11b-Intron 2-R   ACCTGACCTCGCTACCACATGGCT     CD11b-Intron 2-R   CTACGTGTCTAGCTACACATGGCT     CD11b-Intron 2-R   CCTGACCTCGCTACCACATGGGTCCA     CD11b Intron 2-R   ACCTGAGCTCGCTACCACATGGGTCCA     CD11b Intron 2-R   CCTACGTCTCTGTG     CACCTGAGCTCGCTAGCACATTGGCT   Forward and reverse primers for 730bp <i>ItgB2</i> intron     CD11b Intron 2-R   CCTAGCTCGCTACCACATGGGTCCA     CD11b Intron 2-R   CCTAGCTCGCTACCACATGGGTCAC     COTTAGTGTCTAAGCTCACACATGGGTCACACATGGCT	SRF-F	AAA GGATCC GCCGCC	Forward and reverse primers for PCR amplification		
SRF-R   GGG GAATTC   sites are highlighted in gray     TCATTCACTCTTGGTGCTGTGGGGGC   Forward and reverse PCR primers containing Hpal     AAASRF VP16-F   GGG GTTAAC GCCACC   Forward and reverse PCR primers containing Hpal     AAASRF VP16-F   AAA GCGCCGC   Forward and reverse PCR primers containing Hpal     Luciferase constructs coloring   CO18-Intron 6-F   ACCTGAGCTCGCTAGCACATGGGAGC     C018-Intron 6-F   TCTAGTGTCTAAGCTTAAAGTGGAAGC   complementary overhangs are highlighted in gray.     C011b-Intron 2-F   ACCTGAGCTGCGCTAGCACATTGGC   Forward and reverse primers for 746bp <i>ltgdM</i> intron     C011b-Intron 2-R   TCTAGTGTCTAGCCTACACATTGGC   complementary overhangs are highlighted in gray.     C011b-Intron 2-R   ACCTGAGCTCGCTAGCACATTGGCT   complementary overhangs are highlighted in gray.     C011b-Intron 2-R   CTAGTGTCTAGCTACACATGGCT   comard and reverse primers for 730bp <i>ltgB</i> 2 intron     CCTACGTCTTCT   sequencing for indels verification.   Sorget region PCR, TA-cloning and     C011b Intron 6-   ACCTGAGCTCGCTAGTAAAGTGGAGC   forward and reverse primers for 730bp <i>ltgB</i> 2 intron     RCF   CCATGCTCTTC   sequencing for indels verification.   Sorget region PCR, TA-cloning and     RCF   CCATGAGCTCG		ATGTTACCGAGCCAAGCTGGGGCCGC	of mouse SRF ORF; BamHI and EcoRI restriction		
TCATTCACTCTTGGTGGTGGGTGGC   HA-SRF VP16-F GGG GTTAAC GCCACC ATGGCTTCTAGCTATCCTTATG Forward and reverse PCR primers containing Hpal and Notl restriction sites (gray).   HA-SRF VP16-R AAA GCGGCCGC TTAACCACCCCGTAC Forward and reverse primers for 730bp <i>ltgβ2</i> intron AGCAAGTG   CD18-Intron 6-F ACCTGAGCTCACGTAGCACATGGGTGC AGCAGTG Forward and reverse primers for 730bp <i>ltgβ2</i> intron AGCAAGTG   CD11b-Intron 2-F ACCTGAGCTCGCTAGCGACAATTGGC COT1b-Intron 2-F Forward and reverse primers for 740bp <i>ltggM</i> intron TTCTTATGGTCC   CD11b-Intron 2-F ACCTGAGCTCGCTAGCAAACTGGCT GCTTCCTTC SigtigaM Intron 2 target region PCR, TA-cloning and sequencing for indels verification.   CD18 Intron 6 ACCTGAGCTCGCTAGTAAAGTGGACA GCTTCCTTC Forward and reverse primers for 730bp <i>ltgg2</i> intron 6 reverse orientation cloning by In-Fusion. 15bp CD18 Intron 2   CD18 Intron 0 ACCTGAGCTCGCTAGTCAAACATGGCT GCAGTG Forward and reverse primer for 730bp <i>ltgg2</i> intron 6 reverse orientation cloning by In-Fusion. 15bp CD11b Intron 2   CCTAGTGTCTAAGCTCGACACATGGGTGCA COMPlementary overhangs are highlighted in gray. TCTTATGGTCC Forward and reverse primers for 746bp <i>ltgdM</i> intron 2 reverse orientation cloning by In-Fusion. 15bp CD11b Intron 2   CCTAGCTTCCTTC CCTAGCGCTCGCTAGTCAACATGGCTG Forward and reverse primers for site-direct Mutagenesis and deletion CCTCTCTTC CCTTAGCGCTAGCTAACATGCGCTAGTACACATGGCT Forward and reverse primers for site-direct Mutagenesis of <i>ltgdM</i> intron 2 SRE site. ACTCTAGCGCTCAGGAAGGAAGTAGCGTAG C	SRF-R	GGG GAATTC	sites are highlighted in gray		
HA-SRF VP16-F GGG GTTAAC GCCACC Forward and reverse PCR primers containing Hpal   ATGGCTTCTAGCTATCCTTATG and Notl restriction sites (gray).   HA-SRF VP16-R AA GCGGCCGC   TTACTACCCACCGTAC Luciferase constructs cloining   CD18-Intron 6-F ACCTGAGCTCGCTAGCACATGGGAC   FORWard and reverse primers for 730bp <i>ltgB</i> 2 intron 6 forward orientation cloning by In-Fusion. 15bp   CD11b-Intron 2-F ACCTGAGCTCGCTAGCAACATGGCC   CD11b-Intron 2-R CTAGCTGCAGCTAGCAACATGGCC   COT1b-Intron 2-R CCTGAGCTCGCTAGCAACATGGCC   CD11b-Intron 2-R CCTGAGCTCGCTAGCAACATGGCC   CD18 Intron 6- ACCTGAGCTCGCTAGCAACATGGCTAG   CD18 Intron 6- CCTGAGCTCGCTAGCAACATGGCTCA   CD18 Intron 6- CCTGAGCTCGCTAGCAACATGGCTCA   CD18 Intron 6- CCTGAGCTCGCTAGCAACATGGCTC   CCTAGCTCTCC StraygaM Intron growerhangs are highlighted in gray.   CCTAGCTCGCTAGCAACATGGCTC Forward and reverse primers for 730bp <i>ltgB</i> 2 intron   RCF CCTAGCTCGCTAGCAACATGGCTC   CD18 Intron 2 ACCTGAGCTCGCACATGGCACA   CD19 Intron 2 TCTAGTGCTCAACCATGGCTC   CCTAGCTTCCCGCTAGCACATGGCACA Complementary overhangs are highlighted in gray.   CCTAG		TCATTCACTCTTGGTGCTGTGGGTGGC			
ATGGCTTCTAGCTATCCTTATG and Notl restriction sites (gray).   HA-SRF VP16-R AAA GCGGCGCGC   TTACTACCCACCGTAC Intron CTACCCACCGTAC   Luciferase constructs cloning Forward and reverse primers for 730bp <i>llgβ2</i> intron 6 forward orientation cloning by In-Fusion. 15bp   CD18-Intron 6-R ACCTGAGCTCGCTAGCGACAATGGC Forward and reverse primers for 746bp <i>llgdM</i> intron 1TCTTATGGTCC   CD11b-Intron 2-F ACCTGAGCTCGCTAGCGACAATGGC Forward and reverse primers for 746bp <i>llgdM</i> intron 1TCTTATGGTCC   CD11b-Intron 2-R TCTAGTGTCTAAGCTTCAAACATGGCT Somplementary overhangs are highlighted in gray. gltgaM Intron 2 target region PCR, TA-cloning and CCTTCCTTC   CD18 Intron 6 ACCTGAGCTCGCTAGTAAAGTGGAAG   RCF CCTAGTGTCTAAGCTCACATGGGTGCA   CD11b Intron 2 ACCTGAGCTCGCTAGTAAACATGGCT   CRR GCAAGTG   CD11b Intron 2 ACCTGAGCTCGCTAGTCAAACATGGCT   CRR GCAAGTG   CD11b Intron 2 ACCTGAGCTCGCACATGGCACATTGGCT   CD11b Intron 2 ACCTGAGCTCCGCTAGTCAAACATGGCT   CD11b Intron 2 ACCTGAGCTCGCACATTGCACATTGGCT   CD11b Intron 2 CCTAGTGTCTAAGCTCGCACATTGGCT   CD11b Intron 2 TCTAGTGTCTAAGCTCGCACATTGGCT   CD11b Intron 2 TCTAGTGTCTAAGCTCGCACATTGGCT   CD11b Intron 2	HA-SRF VP16-F	GGG GTTAAC GCCACC	Forward and reverse PCR primers containing Hpal		
HA-SRF VP16-R AAA GCGGCCGC   TTACTACCCACCGTAC TTACTACCCACCGTAC   Luciferase constructs cloning CD18-Intron 6-F ACCTGACCTCGCTACCACATGGGTGC Forward and reverse primers for 730bp <i>ltgβ2</i> intron AGCAAGTG forward orientation cloning by In-Fusion. 15bp   CD11b-Intron 2-F ACCTGACCTCGCTACGCACATTGGC Forward and reverse primers for 746bp <i>ltgaM</i> intron 2 forward orientation cloning by In-Fusion. 15bp   CD11b-Intron 2-F ACCTGACCTCGCTACGACACTGGCT Forward and reverse primers for 746bp <i>ltgaM</i> intron 2 forward orientation cloning by In-Fusion. 15bp   CD11b-Intron 2-F ACCTGACCTCGCTAGCGACAATTGGC GCTTCTTC Forward and reverse primers for 730bp <i>ltgβ2</i> intron 6 reverse orientation cloning by In-Fusion. 15bp   CD18 Intron 6- CCATCGTCTGTGG ACCTGACCTCGCTAGTAAGTGGAGG Complementary overhangs are highlighted in gray. GCAAGTG Forward and reverse primers for 730bp <i>ltgβ2</i> intron 6 reverse orientation cloning by In-Fusion. 15bp   CD11b Intron 2 CCAAGGTG CortagGTCCAAAGTGGCTCACACATGGCT Forward and reverse primers for 746bp <i>ltgaM</i> intron 720bp <i>ltgf22</i> intron 6 reverse orientation cloning by In-Fusion. 15bp   CD11b Intron 2 TCTAGTGTCTAAGCTCCAACATGGCT Forward and reverse primers for site-direct mutant-F   CD11b-DA TCCCATCGTCTTTGTAATAATCCCTG Forward and reverse primers for site-direct mutant-F CCTTAGTAGCAGCAGATATACTACATACATAC   CD11b-AA TCCCATTCCTGTGTGAGAGGAGA		ATGGCTTCTAGCTATCCTTATG	and Notl restriction sites (gray).		
IntractacccAccGTAC     Luciferase constructs cloning     CD18-Intron 6-F   ACCTGAGCTGCCTAGCACATGGGTGC     Forward orientation cloning by In-Fusion. 15bp     CD18-Intron 6-R   TCTAGTGTCTAAGCTTAAGGTGAAGC     Complementary overhangs are highlighted in gray. CATCGTCTTGG   Convard orientation cloning by In-Fusion. 15bp     CD11b-Intron 2-F   ACCTGAGCTCCACGGACAATTGGC   Forward and reverse primers for 746bp <i>ItgaM</i> intron TCTTATGGTCC     CD11b-Intron 2-R   TCTAGTGTCTAAGCTTCAAACATGGGCT GCTTCCTTC   Sorward orientation cloning by In-Fusion. 15bp     CD18 Intron 6-   ACCTGAGCTCGCTAGTAAAGTGGAAG   Forward and reverse primers for 730bp <i>ItgB2</i> intron sigligaM Intron 2 target region PCR, TA-cloning and ACCTGAGCTCGCTAGTAAAGTGGACG     CD18 Intron 6-   Intron 6   ICTAGTGTCTAAGCTCACATGGGTGCA GCAAGTG   complementary overhangs are highlighted in gray.     CD11b Intron 2   ACCTGAGCTCGCTAGTCAAACATGGGT   Forward and reverse primer for 746bp <i>ItgaM</i> intron 2 reverse orientation cloning by In-Fusion. 15bp     CD11b Intron 2   TCTAGTGTCTAAGCTCGACAATTGGCT   complementary overhangs are highlighted in gray.     CC11D AGCTGCTAGCCTGCACACATTGGCT   complementary overhangs are highlighted in gray.     CD11b Intron 2   TCTAGTGTCTAAGCTCGACAATTGGCT   complementary overhangs are highlighted in gray.     CD11	HA-SRF VP16-R	AAA GCGGCCGC			
Luciferase constructs cloning     CD18-Intron 6-F   ACCTGAGCTGCCTAGCACATGGGTGC   Forward and reverse primers for 730bp <i>ltgB2</i> intron forward orientation cloning by In-Fusion. 15bp     CD18-Intron 6-R   TCTAGTGTCTAACCTTAAAGTGGAAGC   complementary overhangs are highlighted in gray. CATCGTCGTGG     CD11b-Intron 2-F   ACCTGAGCTCGCTAGCGACAATTGGC TCTTATGGTCC   Forward and reverse primers for 746bp <i>ltgaM</i> intron 2 forward orientation cloning by In-Fusion. 15bp     CD11b-Intron 2-R   TCTAAGGTCTAACCTTCAACATGGCT GCTTCCTTC   complementary overhangs are highlighted in gray. sqltgaM Intron 2 target region PCR, TA-cloning and sequencing for indels verification.     CD18 Intron 6-   ACCTGAGCTCGCTAGTAACATGGGCAG CCACGTGCTAGGCTAG		TTACTACCCACCGTAC			
CD18-Intron 6-F ACCTGAGCTCGCTAGCACATGGGTGC Forward and reverse primers for 730bp <i>ItgB2</i> intron 6 forward orientation cloning by In-Fusion. 15bp   CD18-Intron 6-R TCTAGTGTCTAAGCTTAAAGTGGAAGC complementary overhangs are highlighted in gray.   CD11b-Intron 2-F ACCTGAGCTCGCTAGCGACAATTGGC Forward and reverse primers for 746bp <i>ItgaM</i> intron 2 forward orientation cloning by In-Fusion. 15bp   CD11b-Intron 2-R TCTAGTGTCTAAGCTTCAAACATGGCT complementary overhangs are highlighted in gray.   GCTCCTTC SqltgaM Intron 2 target region PCR, TA-cloning and sequencing for indels verification. fbp ItgB2 intron 6   CCATCGTCTGTG 6 reverse orientation cloning by In-Fusion. 15bp forward and reverse primers for 746bp <i>ItgB2</i> intron 6   CD18 Intron 6 ACCTGAGCTCGCTAGTCAAACTGGCTGCA forward and reverse primers for 746bp <i>ItgB2</i> intron 6   RCF CCAAGGT complementary overhangs are highlighted in gray.   RCR GCTAGGTCGCTAGTCAAACATGGCT forward and reverse primers for 746bp <i>ItgB4</i> intron 2   RCF GCTCCTTC 2 reverse orientation cloning by In-Fusion. 15bp   CD11b Intron 2 ACCTGAGCTCGCTAGTCAAACATGGCT forward and reverse primers for site-direct   mutagenesis and deletion CortTCATTAGCTCAACACATGTAGTATGTAGCT forward and reverse primers for site-direct   Mutagenesis and deletion <td< td=""><td>Luciferase constru</td><td>icts cloning</td><td></td></td<>	Luciferase constru	icts cloning			
AGCAAGTG   6 forward orientation cloning by In-Fusion. 15bp     CD18-Intron 6-R   ACCTGACCTCAGCTAGCGCAACTGGC   complementary overhangs are highlighted in gray.     CD11b-Intron 2-F   ACCTGACCTCGCTAGCCAACTGGC   Forward and reverse primers for 746bp <i>ItgaM</i> intron     CD11b-Intron 2-R   TCAGTGTCCAACCTGCAACATGGCT   complementary overhangs are highlighted in gray.     CD11b-Intron 2-R   TCAGTGTCCAACCTGCAACAGGCT   complementary overhangs are highlighted in gray.     CD11b-Intron 2-R   TCAGTGTCCAACACTGGCAC   forward and reverse primers for 730bp <i>ItgB2</i> intron     CD18 Intron 6-   ACCTGAGCTCGCTAGTCAAACATGGCAC   Forward and reverse primers for 730bp <i>ItgB2</i> intron     RCF   CCATGGTCTAAGCTCAACATGGGTGCA   complementary overhangs are highlighted in gray.     RCR   GCAAGTG   complementary overhangs are highlighted in gray.     RCR   GCTTCCTTC   2 reverse orientation cloning by In-Fusion. 15bp     CD11b Intron 2   TCTAGTGTCAAGCTCCAACATGGCT   Forward and reverse primers for 746bp <i>ItgM</i> intron     RCR   TCTTATGGTCC   2 reverse orientation cloning by In-Fusion. 15bp     CD11b Intron 2   TCAGTGTCTATGCGACAATTGGCT   Forward and reverse primers for site-direct     Mutagenesis and deletion   ACCTGAGCACGTATAGTACTACATA	CD18-Intron 6-F	ACCTGAGCTCGCTAGCACATGGGTGC	Forward and reverse primers for 730bp <i>ltg</i> $\beta$ 2 intron		
CD18-Intron 6-R TCTAGTGTCTAAGCTTAAAGTGGAAGC complementary overhangs are highlighted in gray.   CD11b-Intron 2-F ACCTGAGCTCGCTAGCGACAATTGGC Forward and reverse primers for 746bp <i>ItgaM</i> intron   CD11b-Intron 2-F ACCTGAGCTCGCTAGCGAACAGTGGCC complementary overhangs are highlighted in gray.   CD11b-Intron 2-F ACCTGAGCTCGCTAGGAAAGTGGAAG Forward and reverse primers for 730bp <i>ItgB2</i> intron   CD18 Intron 6 ACCTGAGCTCGCTAGTAAAGTGGAAG Forward and reverse primers for 730bp <i>ItgB2</i> intron   CD18 Intron 6 ACCTGAGCTCGCTAGTCAAACATGGGTGCA complementary overhangs are highlighted in gray.   CD18 Intron 7 ACCTGAGCTCGCTAGTCAAACATGGCT Forward and reverse primers for 746bp <i>ItgaM</i> intron   RCR GCTTCCTTC complementary overhangs are highlighted in gray.   CD11b Intron 2 ACCTGAGCTCGCTAGTCAAACATGGCT complementary overhangs are highlighted in gray.   RCR GCTTCCTTC complementary overhangs are highlighted in gray.   RCR TCTAGTGTCTAAGCTCGACAATTGGCT complementary overhangs are highlighted in gray.   RCR TCTAGTGTCTAAGCTCAACATGGCT complementary overhangs are highlighted in gray.   RCR TCTAGTGTCTAAGCTCAACATGCTF forward and reverse primers for site-direct   Mutagenesis and deletion Complementary overhangs are		AGCAAGTG	6 forward orientation cloning by In-Fusion. 15bp		
CATCGTCGTGTG CD11b-Intron 2-F ACCTGAGCTGCGACGACATTGGC TCTTATGGTCC CD11b-Intron 2-R TCTAGTGTCTAAGCTTCAAACATGGCT GCTTCCTTC GCTTCCTTC CD11b-Intron 6- ACCTGAGCTCGCTAGTAAGCTGGAG CD18 Intron 6- ACCTGAGCTCGCTAGTAAGGTGGAG GCAAGTG CD18 Intron 7 ACCTGAGCTCGCTAGTCAAACATGGGTGCA CD18 Intron 7 ACCTGAGCTCGCTAGTCAAACATGGGTGCA CD18 Intron 7 ACCTGAGCTCGCTAGTCAAACATGGGTGCA CD11b Intron 2 ACCTGAGCTCGCTAGTCAAACATGGGTGCA CD11b Intron 2 ACCTGAGCTCGCTAGTCAAACATGGGTGCA CD11b Intron 2 ACCTGAGCTCGCTAGTCAAACATGGGTGCA CD11b Intron 2 ACCTGAGCTCGCTAGTCAAACATGGGTGCA CD11b Intron 2 ACCTGAGCTCGCTAGTCAAACATGGGTGCA CD11b Intron 2 ACCTGAGCTCGCTAGTCAAACATGGGT CD11b Intron 2 ACCTGAGCTCGCTAGTCAAACATGGGT CD11b Intron 2 CCTTCCTTC CTTAGTGTCTAAGCTCGCACAATTGGCT CD11b Intron 2 CCTTCCTGTTTTGTAAACTGCGCACAATTGGCT CD11b-AA CTCTTATGGTCC Mutagenesis and deletion CD11b-AA CTCTTAATAGACACGTATACTACTACTACTACM Mutant in this study. CD11b-AA CTCTTAATAGAGCAGCTATACTACATACTACTACM Mutant in this study. CD11b-AA CTCTTAATAGAGCAGCTATACTACATACTACM Mutant in this study. CD11b-AA CTCTCAAGGCTGCGCAGATAGTGTGTAGT Mutant in this study. CD11b-AA CTCTCCAAGGCTGGAGAGTACGTAGT Mutant in this study. CD11b-AAT CCTCTCAAGGCTATACTACGTACTACTACTACM Mutant in this study. CD11b-AAT CCTCCAAGGCTCGGAAGTAGTAGTGTAGT Mutant in this study. CD11b-AAT CCTCCAAGGCTCAGGAAGTACGTAGT Forward and reverse primers for site-direct mutant-F ATAGCTGTCTTATTAAGAGCAGCTATACTACGTACTT CTCMAAGGCAAGTGGA CD11b-AT CCCTGAGAGTTAGTACACGTAGTGCTAGT Mutated nucleotides are highlighted in gray. CD11b-AAT CCCTGAGAGTCAGGAAAGTAGCTAGTGG ACAGGGATTATTACAAACAGGAAGTGGA ACACGGGATTATACAAACAGGAAGTAGTGG CD18-MF CTCTTCATAGCTGCTGGTGGGAGGTGTTTGC CAACAGGCTAGGCAGGAGGTGTTGC CD18-MR CAACAGGGCATGGAAAGGAAAGGAAAGGTAGCTAM CD18-MR CAACAGGGCATGGAAAGGAAAAGGCTAMACGCC CD18-MR CAACAGGGCATGGAAAGGAAAAGGCAAAAGGCTAMACCC CD18-MR CAACAGGGCATGGAAAGGAAAAGGCAAAAGGCAAAAGGCTAMACCC CD18-MR CAACAGGGCATGGAAAGGAAAAGGCAAAAGGCAAAAGGCTAMACCC CD18-MR CAACAGGGCATGGAAAGGAAAAGGAAAAGGCTAMACCC CD18-MR	CD18-Intron 6-R	TCTAGTGTCTAAGCTTAAAGTGGAAGC	complementary overhangs are highlighted in gray.		
CD11b-Intron 2-F ACCTGAGCTGAGCAGCATTGGC Forward and reverse primers for 746bp //ga// intron 2 forward orientation cloning by In-Fusion. 15bp CD11b-Intron 2-R TCTAGTGTCTAAGCTTCAAACATGGCT GCTTCCTTC GCTTCCTTC CD11b Intron 6- ACCTGAGCTCGCTAGTAAAGTGGAAG Forward and reverse primers for 730bp //gβ2 intron 6 reverse orientation cloning by In-Fusion. 15bp CD18 Intron 6- RCF CCATCGTCTGTG CD11b Intron 2 ACCTGAGCTCGCTAGTAAAGTGGAAG Forward and reverse primers for 730bp //gβ2 intron 6 reverse orientation cloning by In-Fusion. 15bp CD18 Intron 6 RCF CCAAGTG CD11b Intron 2 ACCTGAGCTCGCTAGTCACACATGGGT CD11b Intron 2 ACCTGAGCTCGCTAGTCAAACATGGCT CD11b Intron 2 TCTAGTGTCTAAGCTCGACAATTGGCT CD11b Intron 2 TCTAGTGTCTAAGCTCGACAATTGGCT CD11b Intron 2 TCTAGTGTCTAAGCTCGACAATTGGCT CD11b Intron 2 TCTAGTGTCTAAGCTCGACAATTGGCT CD11b-AA Mutagenesis and deletion CD11b-AA CTCTTAATAGAGAAAAGTATGTAGTATAG Mutagenesis of <i>llgaM</i> intron 2 SRE site. CTGTCTTATTAAGAG CD11b-AA CTCTTAATAGAGAGCTATACTACCTGF Forward and reverse primers for site-direct Mutagenesis of <i>llgaM</i> intron 2 SRE site. CTGTCTTATTAAGAG CD11b-AA Mutant-R CTTTAATAGAGAGCTATACTACATACG Mutated nucleotides are highlighted in gray. CD11b-AA CTCTTAATAGACAGCTATACTACATACA Mutated nucleotides are highlighted in gray. CD11b-AA CTCTTAATAGACAGCTATACTACCTACTAC Mutated nucleotides are highlighted in gray. CD11b-AA CTCTAATAGACAGCTATACTACCTACTACTACA Mutated nucleotides are highlighted in gray. CD11b-AA CTC CTCAAGGCTTGAGAAAAGTACGTAGT Mutated nucleotides are highlighted in gray. CD11b-ATC CTCAAGGCTCTAG CTCTAATAGCAGCTATACTACGTACTTAC Mutated nucleotides are highlighted in gray. CTCTTATAGGTGTCTTATTACAAAAAGTACGTAG Mutated nucleotides are highlighted in gray. CTTTTTAAGGTGCTGGGAGGTGTTTGC AACCGGGATTATTACAAAAAAAATACTCTTTT Mutated nucleotides are highlighted in gray. CTTTCTTGGCTGGAGGAGGTGTTGC CAACAGCGTGTGTGAGAAAAAGTACGTAG Mutated nucleotides are highlighted in gray. CAACAGGAATTGTAGCTGCTGCTGGCAGGTGTTGC CAACAGGCATGGAGGAAGAAAGAGTAGTAGTAG CAACAGGAGTGTGTG CAACAGGCACTGGCAGGAGGAGGTGTTTGC AACCACAGGTCTGCTGAAAAAAGAGTAGTGAG A		CATCGTCTGTG			
ITCTTATGGTCC   2 forward orientation cloning by In-Fusion. 15bp     CD11b-Intron 2-R TCTAGTGTCAAACTTGGCT   complementary overhangs are highlighted in gray. sglugal Intron 2 target region PCR, TA-cloning and sequencing for indels verification.     CD18 Intron 6-   ACCTGACCTCGCTAGTAAAGTGGAAG   Forward and reverse primers for 730bp <i>Itgβ2</i> intron 6 reverse orientation cloning by In-Fusion. 15bp     CD18 Intron 6   ICTAGTGTCTAAGCTCACATGGGTGCA   Forward and reverse primers for 730bp <i>Itgβ2</i> intron 6 reverse orientation cloning by In-Fusion. 15bp     CD11b Intron 7   ACCTGAGCTCGCTAGTCAAACATGGCT   Forward and reverse primer for 746bp <i>ItgaM</i> intron 2 reverse orientation cloning by In-Fusion. 15bp     CD11b Intron 2   ACCTGAGTCCACATGGACAATTGGCT   Forward and reverse primer for 746bp <i>ItgaM</i> intron 2 reverse orientation cloning by In-Fusion. 15bp     CD11b Intron 2   TCTAGTGTCTAAGCTCGACAATTGGCT   Forward and reverse primers for site-direct     Mutagenesis and deletion   TCCTACTCTGTTTTTGTAATAATCCTG   Forward and reverse primers for site-direct     Mutath-F   ACGAGCAGAGAGAGTAGCTAACATACA AACAGGAAGGAGGAGGAGTACGTACTACATACA   Mutated nucleotides are highlighted in gray.     CD11b-AA   TCTCTCAAGCTGACAAAGGTAACGTACTACTAC Mutated nucleotides are highlighted in gray.   TCTTATATAAGACAGCTAACTACCTCC     CD11b-AC   CTCTCAAGGCTCAACAAAGTACGTACTACTAC TAC   Mutated nucleotides are highlighted in gray.	CD11b-Intron 2-F	ACCTGAGCTCGCTAGCGACAATTGGC	Forward and reverse primers for 746bp <i>ItgαM</i> intron		
CD11b-Intron 2-R ICTAGIGICTAAGCTTCAAACATGGCT complementary overhangs are highlighted in gray.   SQ18 Intron 6- ACCTGAGCTCGCTAGTAAAGTGGAAG Forward and reverse primers for 730bp <i>Itgβ2</i> intron 6 reverse orientation cloning by In-Fusion. 15bp   CD18 Intron 6- CCATCGTCGTAGCTCACATGGGTGCA complementary overhangs are highlighted in gray.   RCR GCAAGTG complementary overhangs are highlighted in gray.   RCR GCTAGTGTCTAAGCTCACATGGGTGCA complementary overhangs are highlighted in gray.   RCR GCTAGTGTCTAAGCTCACATGGCT forward and reverse primer for 746bp <i>ItgaA</i> intron 2 reverse orientation cloning by In-Fusion. 15bp   CD11b Intron 2 RCTGAGGTCTCAAGCTCGACAATTGGCT complementary overhangs are highlighted in gray.   RCR TCTAGTGTCTAAGCTCGACAATTGGCT complementary overhangs are highlighted in gray.   RCR TCTAGTGTCTAAGCTCGACAATTGGCT complementary overhangs are highlighted in gray.   RCR TCTAGTGTCTAAGCTCACAATGGACT forward and reverse primers for site-direct   Mutagenesis and deletion ccattrocatroca		TTCTTATGGTCC	2 forward orientation cloning by In-Fusion. 15bp		
GCTTCCTTC   signal intron 2 target region PCR, 1A-cloning and sequencing for indels verification.     CD18 Intron 6   ACCTGAGCTCGCTAGTAAAGTGGAAG   Forward and reverse primers for 730bp //tgβ2 intron 6 reverse orientation cloning by In-Fusion. 15bp     CD18 Intron 6   TCTAGTGTCTAAGCTCACATGGGTGCA   complementary overhangs are highlighted in gray.     CD11b Intron 2   ACCTGAGCTCGCTAGTCAAACATGGCT   Forward and reverse primer for 746bp //tga// intron     RCF   GCTTCCTTC   2 reverse orientation cloning by In-Fusion. 15bp     CD11b Intron 2   ACCTGAGGTCGACAATTGGCT   complementary overhangs are highlighted in gray.     CD11b Intron 2   TCTAGTGTCTAAGCTCGACAATTGGCT   complementary overhangs are highlighted in gray.     CD11b-AA   TCCACTTCCTGTTTTGTAATAATCCTCG   Forward and reverse primers for site-direct     Mutagenesis and deletion   CTGTCTTATTAAGAG   Mutated nucleotides are highlighted in gray.     CD11b-AA   TCTCAATTCTCAGGGATTACTACATACA   Mutated nucleotides are highlighted in gray.     CD11b-AA   CTGAGCCTTGAGAGGAAGTACGTAGT   Forward and reverse primers for site-direct     Mutant-F   ATAGCGTCTTATTAAGAGGGAAGTACGTAGT   Forward and reverse primers for site-direct     Mutant-F   ATAGCTGTCTTATTA   Mutated nucleotides are highlighted in gray.	CD11b-Intron 2-R	TCTAGTGTCTAAGCTTCAAACATGGCT	complementary overhangs are highlighted in gray.		
Sequencing tor indels verification.     Sequencing tor indels verification.     RCF   CCATCGTCGTGG     RCF   CCATCGTCTGTG     Forward and reverse primers for 730bp <i>Itgβ2</i> intron     GCAAGTG   Forward and reverse primers for 730bp <i>Itgβ2</i> intron     CD11b Intron 2   ACCTGAGCTCGCTAGTCAAACATGGCT     GCAAGTG   Complementary overhangs are highlighted in gray.     CD11b Intron 2   ACCTGAGCTCGCTAGTCAAACATGGCT     GCAAGTG   Complementary overhangs are highlighted in gray.     CD11b Intron 2   TCTAGTGTCTAAGCTCGACAATTGGCT     GCAATTGAGACAAAGTATGTAGTCAAACAGGCT   Forward and reverse primers for site-direct     Mutagenesis and deletion   mutant.F     CD11b-AA   TCCACTTCCTGTTTTTGTAATAATCCCTG     CD11b-AA   TCCACTTCAGGCATACTACACACAGCAAAAGTAGTAG     Mutated nucleotides are highlighted in gray.   CD11b-AA     CTCTTAATAAGACAGCTATACTACACACAGAAA   AAACAGGAAGTGGA     CD11b-AA   CTCTTAATAAGACAGCAAGTACGTAGT     CD11b-AA   CTCTTAATAAGACAGCAAGTACGTAGT     CD11b-C   CTGAGCCTTGAGAGAGAACAGTACGTAGT     Mutated nucleotides are highlighted in gray.   CD11b-C     CTGAGCTGTGCAG   M		GCTTCCTTC	sgltgaM Intron 2 target region PCR, TA-cloning and		
CD18 Intron 6- ACCTGATCGTCGTG Forward and reverse primers for 730bp //g/2 intron   RCF CCATGGTCTGTG 6 reverse orientation cloning by In-Fusion. 15bp   CD18 Intron 6 CCTAGTCCTCAGCTAGTCAAACATGGCT Forward and reverse primer for 746bp //ga/l intron   RCF GCAAGTG 2 reverse orientation cloning by In-Fusion. 15bp   CD11b Intron 2 ACCTGAGCTCGCTAGTCAAACATGGCT Forward and reverse primer for 746bp //ga/l intron   RCF GCAAGTG 2 reverse orientation cloning by In-Fusion. 15bp   CD11b Intron 2 TCTAGTGTCTAAGCTCGACAATTGGCT Forward and reverse primers for site-direct   Mutagenesis and deletion CCACTTCGTCTTGTTTTGTAATAATCCCTG Forward and reverse primers for site-direct   Mutant-F AGAATTGAGAAAAGTATGTAGTAATAG Mutagenesis of //ga/l intron 2 SRE site. AA mutant in this study.   CD11b-AA CTCTTAATAAGACAGCTATACTACATACA Mutated nucleotides are highlighted in gray.   mutant-R CTGAGCCTTGAGAGAAGTAGTAGTAGT Forward and reverse primers for site-direct   mutant-F ATAGCTGTCTTATA Mutated nucleotides are highlighted in gray.   CD11b-TC TCAAGCTGTCTAAGAAAAGTACGTAGT Forward and reverse primers for site-direct   mutant-F ATAGCTGTCTTATA Mutated nucleotides are highlighted in gray.			sequencing for indels verification.		
RCF   CCATCGTCTGTGTG   Prevense of leftation cloning by In-Poston. 13bp     CD18 Intron 6   CCAAGTG   complementary overhangs are highlighted in gray.     RCR   GCAAGTG   complementary overhangs are highlighted in gray.     RCF   GCTTCCTTC   2 reverse orientation cloning by In-Fusion. 15bp     CD11b Intron 2   TCTAGTGTCTAAGCTCGACAATTGGCT   complementary overhangs are highlighted in gray.     RCR   TCTTATGGTCC   2 reverse orientation cloning by In-Fusion. 15bp     CD11b Intron 2   TCTAGTGTCTAAGCTCGACAATTGGCT   complementary overhangs are highlighted in gray.     RCR   TCTTATGGTCC   2 reverse orientation cloning by In-Fusion. 15bp     Mutagenesis and deletion   CCACCTCTGTTTTGTAATGTCCTG   complementary overhangs are highlighted in gray.     CD11b-AA   TCCACTTCCTGTTTTGTAATGTAGTAGTAGT   mutagenesis of <i>ItgaM</i> intron 2 SRE site.     CD11b-AA   CTCTTAATAGACAGCTATACTACTACTACTACT   Mutated nucleotides are highlighted in gray.     CD11b-TC   TAATGAGAAAAGTGTGAGAAAAGTACGTAGT   Forward and reverse primers for site-direct     mutant-R   ATTGAGCTCTAACTACCTACCTACTACT   Mutated nucleotides are highlighted in gray.     CD11b-TC   TAATAGAGAGAAAAGTAGTAGTAGT   Forward and reverse primers f	CD18 Intron 6-	ACCIGAGCICGCIAGIAAAGIGGAAG	Forward and reverse primers for 730bp $IIgB2$ intron		
CD18 Int016 ICTAGTORICAGAGETCAACATGGCA Complementarity overhalitys are highlighted in gray.   RCR GCAAGTG   CD11b Intron 2 ACCTGAGCTCGCTAGTCAAACATGGCT   FORM GCTTCCTTC   CD11b Intron 2 TCTAGTGTCTAAGCTCGACAATTGGCT   CONDEnsion Construction   CD11b Intron 2 TCTAGTGTCTAAGCTCGACAATTGGCT   CONDEnsion Complementary overhangs are highlighted in gray.   CD11b-AA TCCACTTCCTGTTTTGTAATAATCCCTG   Mutagenesis and deletion CTGTTATTAGGAG   CD11b-AA TCCACTTCCTGTTTTGTAATAATCCCTG   Mutant-F AGAATTGAGAAAAGTATGTAGTATAG   Mutagenesis and deletion Mutated nucleotides are highlighted in gray.   CD11b-AA TCCACTTCAAGACAGCTATACTACATACA   Mutated nucleotides are highlighted in gray. TTTTCCAAAGCAGCTATACTACGTACTTC   Mutated nucleotides are highlighted in gray. TCTCTCAAGGCTCAG   Mutated nucleotides are highlighted in gray. TCTCCAAGGCTCAGCAAAAAGTACGTAG   CD11b-TC TAATAAGACAGCTATACTACGTACTTC TC mutant in this study.   CD11b-AATC CCTGAGAATTGAGAAAAAGTACGTAG Forward and reverse primers for site-direct   mutant-F TATAGCTGTCTTATTA Mutated nucleotides are highlighted in gray.	RUF CD19 Intron 6		o reverse orientation cioning by in-rusion. Topp		
NCK   BCARGIN     CD11b Intron 2   ACCTGAGCTCGCTAGTCAAACATGGCT     F   GCTTCCTTC     PCD11b Intron 2   TCTAGTGTCTAAGCTCGACAATTGGCT     CD11b Intron 2   TCTAGTGTCTAAGCTCGACAATTGGCT     Mutagenesis and deletion   CCACTTCCTGTTTTGTAATAATCCCTG     CD11b-AA   TCCACTTCCTGTTTTGTAATAATCCCTG     Mutagenesis and deletion   CCACTTCAGGCACAATAGTAGTAAGTACTACGTAGTATAG     CD11b-AA   TCCTAATAAGACAGCTATACTACATACA     Mutaten trip   AGAATTGAGAAAAAGTATGTAGTAACTACCATAC     Mutate   CTGTCTTATTAAGAGCAGCATATCTACATACA     Mutated nucleotides are highlighted in gray.   CTGTCTTAATAAGACAGCGAAGTACGTAGT     Mutatet   CTGTAGGCCTTGAGAGGAAGTACGTAGT     CD11b-AA   CTGTCAAGCCTTACTACCTACGTACT     CD11b-TC   CTGAGCCTTGAGAGGAAGTACGTAGT     Mutatet   AAACAGGAAGTGGA     CD11b-TC   TAATAAGACAGCTATACTACGTACTTCT     Mutatet   TC <mutat in="" study.<="" td="" this="">     CD11b-AATC   CCCTGAGAATTGAGAAAAAGTACGTAG     Mutatet   CCCTGAGAATTGAGAAAAAGTACGTAG     Mutatet   CCCTGAGAATTGAGAAAAAGACAGCGAGAGTGGGAGTAGTAG     CD11b-AATC   CCCTGAGGAATTGAGAGAAAAGGTAGGAGAGTGG</mutat>			complementary overhangs are nighlighted in gray.		
CD11b Intol 012 ACCT CACCT C 2 reverse orientation cloning by In-Fusion. 15bp   CD11b Intron 2 TCTAGTGTCTAAGCTCGACAATTGGCT 2 reverse orientation cloning by In-Fusion. 15bp   CD11b Intron 2 TCTAGTGTCTAAGCTCGACAATTGGCT complementary overhangs are highlighted in gray.   Mutagenesis and deletion CCACTTCCTGTTTTGTAATAATCCCTG forward and reverse primers for site-direct   Mutagenesis and deletion CCACTTCTGAGACAAAAGTATGTAGTAATAG mutagenesis of <i>ItgaM</i> intron 2 SRE site.   CD11b-AA CTCTTAATAAGACAGCTATACTACATAC Mutagenesis of <i>ItgaM</i> intron 2 SRE site.   Mutant-R CTTTCTCAAGGCAAGGAA ACACGGAAGTGGA   CD11b-TC CTAAGCTGTCTTATTA mutagenesis of <i>ItgaM</i> intron 2 SRE site.   Mutated nucleotides are highlighted in gray. CCTTCAAGGCTGCAG Mutated nucleotides are highlighted in gray.   CD11b-TC TAATAAGACAGCTAACTACGTACTTC TC mutant in this study. mutant-R   CD11b-AATC CCCTGAGACTACTACGTACGTAGCTAG Mutated nucleotides are highlighted in gray.   CD11b-AATC GCTATACTACGTACTTTTTTCTCAATTCT Mutated nucleotides are highlighted in gray.   CD11b-AATC GCTATACTACGTACTTTTTTCTCAATTCT Mutated nucleotides are highlighted in gray.   CD11b-AATC GCTATACTACGTACGTAGGAAGAGAGTGG AATC mutant in th	CD11h Introp 2		Eanward and reverse primer for 746bp //cc// introp		
Notified Tig Defined Tig 2 reverse orientation coming by hish disort. 150p   Provide the disort of the disort of the disort of the disort of the disort. 150p Complementation coming by hish disort. 150p   RCR TCTATGGTCC Complementary overhangs are highlighted in gray.   Mutagenesis and deletion Complementary overhangs are highlighted in gray.   CD11b-AA TCCACTTCCTGTTTTGTAATAATCCCTG   Mutant-F AGAATTGAGAAAAGTATGTAGTATAG   Mutant-R CTCTTAATAAGACAGCATATCTACAACAC   Mutant-R TTTTCTCAATTCTCAGGGATTATTACA   Mutant-F ATAGCTGTCTTATTA   Mutant-F ATAGCTGTCTTATTA   Mutant-F ATAGCTGTCTTATTA   Mutant-F CTCCAAGGCTCAG   Mutant-R CTCTCAAGGCTCAG   Mutant-R CTCTCAAGGCTCAG   Mutant-R CTCTCAAGGCTCAG   Mutant-F TAAGCTGTCTTATTAAGAACAGTAGTAGTAG   Mutant-R CCTCAAGGCTCAG   Mutant-F TAAGCGTGTCTTATTAAGAAAAGTACGTAGT   Mutant-R CCTCCAAGGCTCAG   Mutant-R CCTCAGGAAATTGAGAAAAGTACGTAGT   CD11b-AATC CCCTGAGAATTGAGAAAAAGTACGTAGT   Mutant-R CAGAGGATCATTTTAAGAGACGAGCTAAGTACGTAGT		ACCIGAGCICGCIAGICAAACAIGGCI	2 reverse orientation cloning by In-Eusion 15bp		
CD11b-Intentally Overhality overhality.   CD11b-AA CCCCTGAGAAAAGGTAGGAAGTACGTAAC Mutated nucleotides are highlighted in gray.   CD11b-AATC CCCTGAGAATTGAGAAAAAGTACGTAG Mutated nucleotides are highlighted in gray.   CD11b-AATC CCCTGAGAATTGAGAAAAAGTACGTAGG Mutated nucleotides are highlighted in gray.   CD11b-AATC CCTTACTACGTACTTTTTCCAATTCT Mutated nucleotides are highlighted in gray.   CD11b-AATC GCTATACTACGTAGGAGAGGAGGGGAGTGTTGC AATC mutant in this study.   CD11b-AATC <td>CD11b Introp 2</td> <td></td> <td>complementary overbands are highlighted in gray</td>	CD11b Introp 2		complementary overbands are highlighted in gray		
Mutagenesis and deletion   CD11b-AA TCCACTTCCTGTTTTGTAATAATCCCTG   mutant-F AGAATTGAGAAAAAGTATGTAGTATAG   Mutagenesis of <i>ItgaM</i> intron 2 SRE site.   CTGTTTATAAGAG AA mutant in this study.   CD11b-AA CTCTTAATAAGACAGCTATACTACATAC   Mutagenesis of <i>ItgaM</i> intron 2 SRE site. AA mutant in this study.   CD11b-AA CTCTTAATAAGACAGCTATACTACATAC   Mutated nucleotides are highlighted in gray. Mutated nucleotides are highlighted in gray.   CD11b-TC CTGAGCCTTGAGAGGAAGTACGTAGT Forward and reverse primers for site-direct   mutant-F ATAGCTGTCTTATTA mutagenesis of <i>ItgaM</i> intron 2 SRE site.   CD11b-TC TAATAAGACAGCTAACGTACGTACTTC TC mutant in this study.   Mutated nucleotides are highlighted in gray. Mutated nucleotides are highlighted in gray.   CD11b-AATC CCCTGAGAATTGAGAAAAGTACGTAG Mutated nucleotides are highlighted in gray.   CD11b-AATC GCTATACTACGTACTTTTTACAAAACAGGAAGTGG AATC mutant in this study.   CD11b-AATC GCTATACTACGTACTTTTTCCAATTCT Mutated nucleotides are highlighted in gray.   CD11b-AATC GCTATACTACGTACTTTTTCCAATTCT Mutated nucleotides are highlighted in gray.   CD11b-AATC GCTATACTACGTAGTCTTTTTCCAATTCT <t< td=""><td>RCR</td><td>TCTTATGGTCC</td><td>complementary overhangs are highlighted in gray.</td></t<>	RCR	TCTTATGGTCC	complementary overhangs are highlighted in gray.		
CD11b-AATCCACTTCCTGTTTTGTAATAATCCCTG Forward and reverse primers for site-direct mutant-FAGAATTGAGAAAAAGTATGTAGTATAG CTGTCTTATTAAGAGmutagenesis of <i>ItgaM</i> intron 2 SRE site. AA mutant in this study.CD11b-AACTCTTAATAAGACAGCTATACTACATAC Mutant-RCD11b-TCCTGAGCCTTGAGAGGAAGTACGTAGT AAACAGGGAAGTGGACD11b-TCCTGAGCCTTGAGAGGAAGTACGTAGT Mutant-Fmutant-FATAGCTGTCTTATTA ATAGCTGTCTTATTACD11b-TCTAATAAGACAGCTATACTACCTACT TAATAAGACAGCTATACTACCAGCCD11b-TCTAATAAGACAGCTATACTACCTACCTACT Mutant-RCTCTCAAGGCTCAGMutated nucleotides are highlighted in gray.CD11b-AATCCCCTGAGAATTGAGAAAAAGTACGTAG TATAGCTGTCTTATTAAGAGCTGAGCACD11b-AATCCCCTGAGAATTGAGAAAAAGTACGTAG TATAGCTGTCTTATTAAGAGCTGAGCAMutated nucleotides are highlighted in gray.CD11b-AATCGCTATACTACGTACTTTTTCTCAATTCT Mutated nucleotides are highlighted in gray.CD11b-AATCGCTATACTACGAGGAGGGGTGTTTGC AGAGATCCD11b-AATCGCTATACTACGAGGAGAGAGGAGTAGGAAGAGGA AGAGATCCD11b-AATCGCTATACTACGTGGGGGGGGTGTTGCC AGAGATCCD11b-AATCGCTATACTACGTGGGGGGAGGTGTTGCC AGAGATCCD11b-AATCGCTATACTACGTGGGGAGGAGAGAGGAGAGGAGAGAGGAGAGAGTA AGAGATCCD11b-AATCGCTATACTACGTGGGGGGAGGAGAAGAGGAGAGAGGAGAGAGA	Mutagenesis and	deletion			
mutant-FAGAATTGAGAAAAAGTATGTAGTATGT AGAATTGAGAAAAGTATGTAGTATGTAGTATAG CTGTCTTATTAAGAGmutagenesis of <i>ltgaM</i> intron 2 SRE site. AA mutant in this study.CD11b-AACTCTTAATAAGACAGCTATACTACATAC AAACAGGAAGTGGAMutated nucleotides are highlighted in gray.CD11b-TCCTGAGCCTTGAGAGGGAAGTACGTAGT AAACAGGAAGTGGAForward and reverse primers for site-direct mutagenesis of <i>ltgaM</i> intron 2 SRE site.CD11b-TCCTGAGCCTTGAGAGGGAAGTACGTAGT ATAGCTGTCTTATTAForward and reverse primers for site-direct mutagenesis of <i>ltgaM</i> intron 2 SRE site.CD11b-TCTAATAAGACAGCTATACTACGTACTTC TAATAAGACAGCTCAGGMutated nucleotides are highlighted in gray.CD11b-AATCCCCTGAGAATTGAGAAAAAGTACGTAG CTCForward and reverse primers for site-direct mutagenesis of <i>ltgaM</i> intron 2 SRE site.CD11b-AATCCCCTGAGAATTGAGAAAAAGTACGTAG TATAGCTGTCTTATTAAGAGCTGAGCA CTCMutated nucleotides are highlighted in gray.CD11b-AATCGCTATACTACGTACTTTTTCCAATTCT AATAGCTGTCTTATTACAAACAGGAAGTGG AGAGATCMutated nucleotides are highlighted in gray.CD11b-MFCTTCTTTTGTGGCTGGGAGGTGTTTGC AGGGATTATTACAAAACAGGAAGTGG AGAGATCForward and reverse primers for site-direct mutagenesis of <i>ltgβ2</i> intron 6 SRE site in luciferase construct.CD18-MRCAACACAGTCATGAAAAGGAAAAGGAAAAGAGTA TTTTTAAAATGCAAACACCMutated nucleotides are highlighted in gray.	CD11b-AA	TCCACTTCCTGTTTTGTAATAATCCCTG	Forward and reverse primers for site-direct		
CTGTCTTATTAAGAGAA mutant in this study.CD11b-AACTCTTAATAAGACAGCTATACTACATACmutant-RTTTTTCTCAATTCTCAGGGGATTATTACA AAACAGGAAGTGGACD11b-TCCTGAGCCTTGAGAGGAAGTACGTAGT AAACAGGAAGTGCAACD11b-TCCTGAGCCTTGAGAGGAAGTACGTAGT AAACAGGCAGCTATATTACCD11b-TCTAATAAGACAGCTATACTACGTACTTC TCCD11b-TCTAATAAGACAGCTATACTACGTACTTC TCTCAAGGCTCAGCD11b-TCTAATAAGACAGCTATACTACGTACTTC TCTCAAGGCTCAGCD11b-AATCCCCTGAGAATTGAGAAAAAGTACGTAG TATAGCTGTCTTATTAAGAGCTGAGCA CTCCD11b-AATCCCCTGAGAATTGAGAAAAAGTACGTAG TATAGCTGTCTTATTAAGAGCTGAGCA CTCCD11b-AATCGCTATACTACGTACTTTTTCTCAATTCT CAGGGATTATTACAAAACAGGAAGGAGGAGGTG AGAGATCCD18-MFCTTCTTTTGTGGCTGGGAGGTGTTTGC CAGAGCTGTGTGTGTGTGTGCD18-MRCAACACAGTCATGAAAAGGAAAAGGAGAA CACACAGTCATGAAAAGGAAAAGGAAGAGTA Mutated nucleotides are highlighted in gray.CD18-MRCAACACAGTCATGAAAAGGAAAAGAGAGTA CAGAGCTATGAAAAGAAAGAAAGAGAGTA Mutated nucleotides are highlighted in gray.	mutant-F	AGAATTGAGAAAAAGTATGTAGTATAG	mutagenesis of <i>ItagM</i> intron 2 SRE site.		
CD11b-AA mutant-RCTCTTAATAAGACAGCTATACTACATAC TTTTCTCAATTCTCAGGGATTATTACA AAACAGGAAGTGGAMutated nucleotides are highlighted in gray.CD11b-TCCTGAGCCTTGAGAGGAAGTACGTAGT ATAGCTGTCTTATTAForward and reverse primers for site-direct mutagenesis of <i>ItgaM</i> intron 2 SRE site.CD11b-TCTAATAAGACAGCTATACTACGTACTC TAATAAGACAGCTCAGForward and reverse primers for site-direct mutagenesis of <i>ItgaM</i> intron 2 SRE site.CD11b-TCTAATAAGACAGCTATACTACGTACTC TCCAAGGCTCAGMutated nucleotides are highlighted in gray.CD11b-AATCCCCTGAGAATTGAGAAAAAGTACGTAG TATAGCTGTCTTATTAAGAGCTGAGCA CTCForward and reverse primers for site-direct mutagenesis of <i>ItgaM</i> intron 2 SRE site. AATC mutant in this study.CD11b-AATCGCTATACTACGTACTTTTTCCAATTCT AGGGATTATTACAAAACAGGAAGGG AGAGATCMutated nucleotides are highlighted in gray.CD11b-AATCGCTATACTACGTACTTTTTCCAATTCT AGGGATTATTACAAAACAGGAAGTGG AGAGATCMutated nucleotides are highlighted in gray.CD18-MFCTTCTTTTGTGGCTGGGAGGGGGGTGTTTGC CAGCACAGTCATGAAAAAAAAAAAAAACTCTTTTT CTTTCATGACTGTGTGForward and reverse primers for site-direct mutagenesis of <i>Itgβ2</i> intron 6 SRE site in luciferase construct.CD18-MRCAACACAGTCATGAAAGGAAAAGGAAAAGAGTA TTTTTTGTATTTAAAATGCAAACACCMutated nucleotides are highlighted in gray.		CTGTCTTATTAAGAG	AA mutant in this study.		
mutant-RTTTTTCTCAATTCTCAGGGATTATTACA AAACAGGAAGTGGACD11b-TCCTGAGCCTTGAGAGGAAGTACGTAGT ATAGCTGTCTTATTAForward and reverse primers for site-direct mutagenesis of <i>ltgαM</i> intron 2 SRE site.CD11b-TCTAATAAGACAGCTATACTACGTACTTC TCTCAAGGCTCAGTC mutant in this study. Mutated nucleotides are highlighted in gray.CD11b-AATCCCCTGAGAATTGAGAAAAAGTACGTAG TATAGCTGTCTTATTAAGAGCTGAGCAForward and reverse primers for site-direct mutagenesis of <i>ltgαM</i> intron 2 SRE site.CD11b-AATCCCCTGAGAATTGAGAAAAAGTACGTAG TATAGCTGTCTTATTAAGAGCTGAGCA CTCForward and reverse primers for site-direct mutagenesis of <i>ltgαM</i> intron 2 SRE site. AATC mutant in this study.CD11b-AATCGCTATACTACGTACTTTTTCTCAATTCT CAGGGATTATTACAAAACAGGAAGTGG AGAGATCMutated nucleotides are highlighted in gray.CD18-MFCTTCTTTTGTGGGCTGGGAGGTGTTTGC ACACACAGTCATGAAAGAAAGAACACCForward and reverse primers for site-direct mutagenesis of <i>ltgβ2</i> intron 6 SRE site in luciferase construct.CD18-MRCAACACAGTCATGAAAGGAAAAGAGAGTA TTTTTGTATTTTAAAATGCAAACACCMutated nucleotides are highlighted in gray.	CD11b-AA	CTCTTAATAAGACAGCTATACTACATAC	Mutated nucleotides are highlighted in gray.		
AAACAGGAAGTGGACD11b-TCCTGAGCCTTGAGAGGAAGTACGTAGT ATAGCTGTCTTATTAForward and reverse primers for site-direct mutagenesis of <i>ltgαM</i> intron 2 SRE site.CD11b-TCTAATAAGACAGCTATACTACGTACTTC TCTCAAGGCTCAGTC mutant in this study. Mutated nucleotides are highlighted in gray.CD11b-AATCCCCTGAGAATTGAGAAAAAGTACGTAG CCTTAAGCTGTCTTATTAAGAGCTGAGCA CTCForward and reverse primers for site-direct mutagenesis of <i>ltgαM</i> intron 2 SRE site. AATC mutant in this study.CD11b-AATCCCCTGAGAATTGAGAAAAAGTACGTAG CTCForward and reverse primers for site-direct mutagenesis of <i>ltgαM</i> intron 2 SRE site. AATC mutant in this study.CD11b-AATCGCTATACTACGTACTTTTTCTCAATTCT CAGGGATTATTACAAAACAGGAAGTGG AGAGATCMutated nucleotides are highlighted in gray.CD18-MFCTTCTTTTGTGGCTGGGAGGTGTTTGC ACACACAGTCATGAAAGGAAAAGGAAAGGTA TTTTAAAATACAAAAAGGAAAAGGAAAGGTA TTTTTAAAATGCAAAAGGAAAGGAAAAGAGTA TTTTTTTTTTTTTTAAAATGCAAAAGGAAAAGAGTA TTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTT	mutant-R	TTTTTCTCAATTCTCAGGGATTATTACA			
CD11b-TCCTGAGCCTTGAGAGGAAGTACGTAGT ATAGCTGTCTTATTAForward and reverse primers for site-direct mutagenesis of <i>ItgaM</i> intron 2 SRE site.CD11b-TCTAATAAGACAGCTATACTACGTACTTC TATAAGACAGCTCAGTC mutant in this study. Mutated nucleotides are highlighted in gray.CD11b-AATCCCCTGAGAATTGAGAAAAAGTACGTAG TATAGCTGTCTTATTAAGAGCTGAGCA CTCForward and reverse primers for site-direct mutagenesis of <i>ItgaM</i> intron 2 SRE site. AATC mutant in this study.CD11b-AATCCCTGAGAATTGAGAAAAAGTACGTAG TATAGCTGTCTTATTAAGAGCTGAGCA CTCForward and reverse primers for site-direct mutagenesis of <i>ItgaM</i> intron 2 SRE site. AATC mutant in this study.CD11b-AATCGCTATACTACGTACTTTTTCTCAATTCT CAGGGATTATTACAAAACAGGAAGTGG AGAGATCMutated nucleotides are highlighted in gray.CD18-MFCTTCTTTTTGTGGCTGGGAGGTGTTTGC CAACACAGTCATGAAAGAAAAAAAAAAAAAAAAAAAAAA		AAACAGGAAGTGGA			
mutant-FATAGCTGTCTTATTAmutagenesis of <i>ltgaM</i> intron 2 SRE site.CD11b-TCTAATAAGACAGCTATACTACGTACTTCTC mutant in this study.mutant-RCTCTCAAGGCTCAGMutated nucleotides are highlighted in gray.CD11b-AATCCCCTGAGAATTGAGAAAAAGTACGTAGForward and reverse primers for site-directmutant-FTATAGCTGTCTTATTAAGAGCTGAGCAmutagenesis of <i>ltgaM</i> intron 2 SRE site.CD11b-AATCGCTATACTACGTACTTTTTCTCAATTCTMutated nucleotides are highlighted in gray.CD11b-AATCGCTATACTACGTACTTTTTCTCAATTCTMutated nucleotides are highlighted in gray.CD11b-AATCGCTATACTACGTGGGAGGTGTTTGCForward and reverse primers for site-directmutant-RCAGGGATTATTACAAAACAGGAAGTGG AGAGATCMutated nucleotides are highlighted in gray.CD18-MFCTTCTTTTGTGGCTGGGAGGTGTTTGC CTTCATGACTGTGTTGForward and reverse primers for site-directCD18-MRCAACACAGTCATGAAAGGAAAAGAGTA TTTTTTGTATTTTAAAATGCAAACACCMutated nucleotides are highlighted in gray.	CD11b-TC	CTGAGCCTTGAGAGGAAGTACGTAGT	Forward and reverse primers for site-direct		
CD11b-TC mutant-RTAATAAGACAGCTATACTACGTACTTC CTCTCAAGGCTCAGTC mutant in this study. Mutated nucleotides are highlighted in gray.CD11b-AATC mutant-FCCCTGAGAATTGAGAAAAAGTACGTAG TATAGCTGTCTTATTAAGAGCTGAGCA CTCForward and reverse primers for site-direct mutagenesis of <i>ItgαM</i> intron 2 SRE site. AATC mutant in this study.CD11b-AATC CD11b-AATCGCTATACTACGTACTTTTTCTCAATTCT CAGGGATTATTACAAAACAGGAAGTGG AGAGATCMutated nucleotides are highlighted in gray.CD18-MFCTTCTTTTGTGGCTGGGAGGTGTTTGC ATTTTAAAATACAAAAAAAAAACTCTTTT CCTTTCATGACTGTGTTGForward and reverse primers for site-direct mutagenesis of <i>Itgβ2</i> intron 6 SRE site in luciferase construct.CD18-MRCAACACAGTCATGAAAGGAAAAGAGATA TTTTTTGTATTTTAAAATGCAAACACCMutated nucleotides are highlighted in gray.	mutant-F	ATAGCTGTCTTATTA	mutagenesis of <i>ItgαM</i> intron 2 SRE site.		
mutant-RCTCTCAAGGCTCAGMutated nucleotides are highlighted in gray.CD11b-AATCCCCTGAGAATTGAGAAAAAGTACGTAGForward and reverse primers for site-directmutant-FTATAGCTGTCTTATTAAGAGCTGAGCA CTCmutagenesis of <i>ItgaM</i> intron 2 SRE site. AATC mutant in this study.CD11b-AATCGCTATACTACGTACTTTTTCTCAATTCT Mutated nucleotides are highlighted in gray.CD11b-AATCGCTATACTACGTACTTTTTCTCAATTCT CAGGGATTATTACAAAACAGGAAGTGG AGAGATCMutated nucleotides are highlighted in gray.CD18-MFCTTCTTTTGTGGCTGGGAGGTGTTTGC ATTTAAAATACAAAAAAAATACTCTTTT CCTTTCATGACTGTGTTGForward and reverse primers for site-direct mutagenesis of <i>Itgβ2</i> intron 6 SRE site in luciferase construct.CD18-MRCAACACAGTCATGAAAGGAAAAGAGTA TTTTTTGTATTTTAAAATGCAAACACCMutated nucleotides are highlighted in gray.	CD11b-TC	TAATAAGACAGCTATACTACGTACTTC	TC mutant in this study.		
CD11b-AATC CCCTGAGAATTGAGAAAAAGTACGTAG Forward and reverse primers for site-direct   mutant-F TATAGCTGTCTTATTAAGAGCTGAGCA mutagenesis of <i>ItgaM</i> intron 2 SRE site.   CD11b-AATC GCTATACTACGTACTTTTTCTCAATTCT Mutated nucleotides are highlighted in gray.   CD11b-AATC GCTATACTACGTACTTTTTCTCAATTCT Mutated nucleotides are highlighted in gray.   mutant-R CAGGGATTATTACAAAACAGGAAGTGG AATC mutant in this study.   CD18-MF CTTCTTTTGTGGCTGGGAGGTGTTTGC Forward and reverse primers for site-direct   CD18-MR CAACACAGTCATGAAAGGAAAAGAGTA mutagenesis of <i>Itgβ2</i> intron 6 SRE site in luciferase construct.   CD18-MR CAACACAGTCATGAAAGGAAAAGAGTA Mutated nucleotides are highlighted in gray.	mutant-R	CTCTCAAGGCTCAG	Mutated nucleotides are highlighted in gray.		
mutant-F TATAGCTGTCTTATTAAGAGCTGAGCA CTC mutagenesis of <i>ItgαM</i> intron 2 SRE site.   CD11b-AATC GCTATACTACGTACTTTTTCTCAATTCT Mutated nucleotides are highlighted in gray.   mutant-R CAGGGATTATTACAAAACAGGAAGTGG AGAGATC Mutated nucleotides are highlighted in gray.   CD18-MF CTTCTTTTGTGGCTGGGAGGTGTTTGC ATTTTAAAATACAAAAAAATACTCTTTT CCTTTCATGACTGTGTTG Forward and reverse primers for site-direct mutagenesis of <i>Itgβ2</i> intron 6 SRE site in luciferase construct.   CD18-MR CAACACAGTCATGAAAGGAAAAGAGTA TTTTTTGTATTTTAAAATGCAAACACC Mutated nucleotides are highlighted in gray.	CD11b-AATC	CCCTGAGAATTGAGAAAAAGTACGTAG	Forward and reverse primers for site-direct		
CTC AATC mutant in this study.   CD11b-AATC GCTATACTACGTACTTTTTCTCAATTCT Mutated nucleotides are highlighted in gray.   mutant-R CAGGGATTATTACAAAACAGGAAGTGG Mutated nucleotides are highlighted in gray.   CD18-MF CTTCTTTTGTGGCTGGGAGGTGTTTGC Forward and reverse primers for site-direct   CD18-MF CTTCTTTGACTGTGTTG Forward and reverse primers for site-direct   CD18-MR CAACACAGTCATGAAAGGAAAAGAGTA mutagenesis of <i>Itgβ2</i> intron 6 SRE site in luciferase   CD18-MR CAACACAGTCATGAAAGGAAAAGAGTA Mutated nucleotides are highlighted in gray.	mutant-F	TATAGCTGTCTTATTAAGAGCTGAGCA	mutagenesis of <i>ItgαM</i> intron 2 SRE site.		
CD11b-AATC GCTATACTACGTACTTTTTCTCAATTCT Mutated nucleotides are highlighted in gray.   mutant-R CAGGGATTATTACAAAACAGGAAGTGG   AGAGATC AGAGATC   CD18-MF CTTCTTTTGTGGCTGGGAGGTGTTTGC   ATTTTAAAATACAAAAAAATACTCTTTT mutagenesis of <i>Itgβ2</i> intron 6 SRE site in luciferase construct.   CD18-MR CAACACAGTCATGAAAGGAAAAGAGTA   TTTTTTTTTTTTTTTTAAAATGCAAAAGGAAAAGAGTA Mutated nucleotides are highlighted in gray.		СТС	AATC mutant in this study.		
mutant-R CAGGGATTATTACAAAACAGGAAGTGG AGAGATC   CD18-MF CTTCTTTTGTGGCTGGGAGGTGTTTGC ATTTTAAAATACAAAAAATACTCTTTT CCTTTCATGACTGTGTTG Forward and reverse primers for site-direct mutagenesis of <i>Itgβ2</i> intron 6 SRE site in luciferase construct.   CD18-MR CAACACAGTCATGAAAGGAAAAGAGTA TTTTTTGTATTTTAAAATGCAAAGGAAAAGAGTA TTTTTTTGTATTTTAAAATGCAAACACC	CD11b-AATC	GCTATACTACGTACTTTTTCTCAATTCT	Mutated nucleotides are highlighted in gray.		
AGAGATC AGAGATC   CD18-MF CTTCTTTTGTGGCTGGGAGGTGTTTGC Forward and reverse primers for site-direct   ATTTTAAAATACAAAAAAATACTCTTTT mutagenesis of <i>Itgβ2</i> intron 6 SRE site in luciferase   CCTTTCATGACTGTGTTG construct.   CD18-MR CAACACAGTCATGAAAGGAAAAGAGTA Mutated nucleotides are highlighted in gray.   TTTTTTTTTTTTTTTTTAAAATGCAAACACC TTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTTT	mutant-R				
CD18-MF CTTCTTTGTGGCTGGGAGGTGTTGC Forward and reverse primers for site-direct   ATTTTAAAATACAAAAAATACTCTTTT mutagenesis of <i>ltgβ2</i> intron 6 SRE site in luciferase   CCTTTCATGACTGTGTTG construct.   CD18-MR CAACACAGTCATGAAAGGAAAAGAGAGTA   TTTTTTTTTTTTTTTAAAATGCAAACACC Mutated nucleotides are highlighted in gray.		AGAGAIC	Forward and several animary for site divest		
CCTTTCATGACTGTGTTG construct. CD18-MR CAACACAGTCATGAAAGGAAAGGGAAAGGGAAAGGGTA Mutated nucleotides are highlighted in gray. TTTTTTTGTATTTTAAAATGCAAACACC	CD18-MF		Forward and reverse primers for site-direct		
CD18-MR CAACACAGTCATGAAAGGAAAAGAGTA Mutated nucleotides are highlighted in gray.			nucligenesis of <i>hgp2</i> infron 6 SRE site in luciferase		
TITTITTGTATTTAAAATGCAAAAGAGTAMMulaled hucleolides are highlighled in gray.			uunsuuuu. Mutated nucleotides are bigblighted in grov		
			indialed nucleolides are nighilghled in gray.		
TCCCAGCCACAAAAGAAG		TCCCAGCCACAAAAGAAG			

CD18-Deletion-F	TGGCTGGGAGGTGTTTAATCTAGTTTC AGGTCTGAAGAGGGTTGCAAAGACAT	Forward and reverse primers for 52bp deletion of ΠtgαM intron 2 region harboring SRE site.
CD18- Deletion-R	GACCTGAAACTAGATTAAACACCTCCC AGCCACAAAAGAAGGGACCAAGGGAC	
	ATCCA	
Quantitative PCR		
Acta2	F: GGCACCACTGAACCCTAAGG R: ACAATACCAGTTGTACGTCCAGA	qRT-PCR of <i>Acta2</i> , in Exon 4 and 5/6 <sup>3</sup> .
Krt17		aPT PCP of $KPT_{17}$ both in Evon $4^4$
	R: CGGCCATCACGCCACAGTTT	
Flna	F: GAGTTCGGCATTTGGACTAGG	gRT-PCR of <i>Flna</i> , in Exon 41and 42 <sup>3</sup> .
	R: GGGCTATCAGGTATGTGCTCC	
Fhl2	F: GATCGGCACTGGCATGAAG R: AGCAAAGGGCTTGTCCACC	qRT-PCR of <i>FHL2</i> , both in Exon 3 <sup>4</sup> .
Myh9		aRT-PCR of $Myh9$ both in Exon $14^3$
iviyi10	R: CTTGGGCTTCTGGAACTTGG	
Actb	F: GGCTGTATTCCCCTCCATCG	qRT-PCR of <i>Actb</i> , in Exon 2 and 3 <sup>3</sup> .
	R: CCAGTIGGTAACAATGCCATGT	
Actg1	F: CGTCCACCGCAAATGCTTC R: TGCCAGGGCAAATTGATACT	qRT-PCR of <i>Actg1</i> , both in Exon 6°.
Srf	F: GCAAGGCGCTGATTCAGAC	gRT-PCR of Srf, both in Exon 2 <sup>3</sup> .
	R: TCAGATTCCGACACCTGGTAG	
Egr3	F: AGCCCAATCCGGAACTCTCTT	qRT-PCR of <i>Egr3</i> , both in Exon 2 <sup>6</sup> .
	R: GGAAGGAGAGTCGAAAGCGAA	
Egr1	F: ATTGATGTCTCCGCTGCAGATC	qRT-PCR of <i>Egr1</i> , both in Exon 1 <sup>6</sup> .
Fosb		qRI-PCR of <i>Fosb</i> , designed by Primer3 (version
<u>а Гаа</u>		0.4.0), III EXOI 3 and 4.
<i>c-гоs</i>	R: GCTCCAAGGATGGCTTGGGCTC	qRI-PCR of Egr1, both in Exon 4°.
Coro1c	F: CGCAGAGCGTGCTTATTCG	gRT-PCR of Coro1c intron-containing pre-mRNA,
	R: TGCCAACCATTTCCAAAACTAA	both in Intron 1 <sup>7</sup> .
Jun B	F: ACAAGGTGAAGACACTCAAGGCT	qRT-PCR of <i>JunB</i> , both in Exon 1 <sup>6</sup> .
	R: ATGACCTTCTGCTTGAGCTGC	
Egr2	F: GAGCAAATGATGACCGCCAA	qRT-PCR of <i>Egr2</i> , both in Exon 1 <sup>6</sup> .
	R: TGTCAGGCAGCTGGTGCATAA	
Vcl	F: AGCCCAGATGCTTCAGTCAGA	qRT-PCR of <i>Vcl</i> , both in Exon 3 <sup>3</sup>
Nr4a1		qRI-PCR of <i>Nr4a1</i> , both in Exon 2'.
Tuftelin		aRT-PCR of <i>Tuffelin</i> both in Exon 10 <sup>4</sup>
T uncom		
Tagin		gRT-PCR of <i>Tagln</i> both in Exon 4-5 <sup>3</sup>
ragin	R: GTGAAGTCCCTCTTATGCTCCT	
ltga1	F: CAGTGGAGGACATGTTTGGAT	gRT-PCR of Itga1, designed by Primer3 (version
0	R: TCTCTCTCTCCCAACTGGACA	0.4.0), in Exon 2 and 3.
ltga2	F: GCAACTGGCTACTGGTTGGT	gRT-PCR of Itga2, designed by Primer3 (version
U	R: AGCTTTTCACAGGTGGCAGT	0.4.0), in Exon 2/3 and 3.
ltga4	F: GGGCTTGTGAACCCAACTTC	qRT-PCR of <i>Itga4</i> , both in Exon 22 <sup>5</sup> .
	R: TGCATGTTTCTGGCTCGTTTT	
ltga5	F: TTCTCCGTGGAGTTTTACCG	qRT-PCR of Itga5, designed by Primer3 (version
	R: TAGACAGCACCACCTTGCAG	0.4.0), in Exon 1 and 2.
ItgaL	F: CCCGCTTGGTCGGTTTG	qRT-PCR of <i>ItgaL</i> , both in Exon 14⁵.
	R: CAGTCAGCCTATCCCCATTGA	
ltgaM	F: CTGAACATCCCATGACCTTCC	qRT-PCR of <i>ItgaM</i> , in Exon 2 and 4 <sup>8</sup> .
	R: GCCCAAGGACATATTCACAGC	

ltgb1		qRT-PCR of <i>Itgb1</i> , designed by Primer3 (version
ltgb2		qRI-PCR of <i>Itgb2</i> , both in Exon 6°.
	R: TIGCCGACCICIGICIGAAAC	
RI-PCR		
SRF RT-PCR-F		Forward and reverse primers for RT-PCR detecting murine SRF truncations.
SRF RT-PCR-R	CAGGAACACCTGAGGGACAC	
aBlock DNA		
Itob2/aM CDS	ggtaaagccaccatgaAGTACAAAGTCAGCA	Oligo sequence containing potential sgRNA target
	GTTGCCGGGACTGTATCCAGTCGGGG	sites for both <i>Itab2</i> and <i>ItaaM</i> protein coding
	CCTGGCTGTTCCTGGTGCCAGAAGCT	regions was shown in uppercase.
	GTCACTGCTGCTTGCCCTAGCTGGACT	
	GTTCTTCCTGGGATCTGcACGGGCACA	
	GCTGCTGCTGAAGGGTTGTCCAGCCG	
	ATGATATCATGGACCCCAGGAGCATC	
	GCTAATCCTGAGTTCGACCAACGGGG	
	GCAACGGAAACAGCCCCATGACCTTC	
	CAAGAGAATGCAAAAGGCTTTGGACA	
	GAGTGTGGTCCAGCTTGGCGGACCAG	
	TGTGGTTGTTGCAGCCCCCAGGAGG	
	CAAAGGCTGTTAACCAGACAGGtgaagat	
	gccaaaaacattaagaagggcccagcgccattctacc	
ItgaM intron 2	ggtaaagccaccatgATCCGTACATACAGAA	Oligo sequence containing potential sgRNA target
	TAAGAGACCGGGGGCTATACTACATACT	sites covering SRE in <i>ItgaM</i> intron 2 region was
	TCCTCTCAAGGCTCAGGGATTATTACA	shown in uppercase, and underlined sequence
	ACAGCCTCTGTGACTGCAGACACCAG	encoded (GSSSS)×3 spacer.
	CAC <u>GGTGGAGGTGGAAGTGGAGGTG</u>	
	<u>GAGGTTCTGGAGGCGGGGGTAGC</u> gaa	
	gatgccaaaaacattaagaagggcccagcgccattct	
	acc	
15bp-F	ggtaaagccaccatg	PCR primers for gBlock amplification to introduce
15bp-R	ggtagaatggcgctg	15bp overhang for In-Fusion reactions.
sgRNA sequence		
sgLuc2P		
sgltgb2_	F: CACCg ATCCTGAGTTCGACCAACGG	sgRNA targeting Itgb2 Exon4 with high efficiency,
Т6	R: AAAC CCGTTGGTCGAACTCAGGAT	used in all subsequent experiments.
sgltgb2 T13	F: CACCg	sgRNA targeting Itgb2 Exon3 with minimal
000	AGTCGGGGCCTGGCTGTTCC	suppression effect.
	R: AAAC GGAACAGCCAGGCCCCGACT	
	С	
sgltgaM T4	F: CACCg	sgRNA targeting ItgaM Exon2 with high efficiency,
	CAGAGTGTGGTCCAGCTTGG	used in all subsequent experiments.
	R: AAAC CCAAGCTGGACCACACTCTG	
	С	
sgltgaM_ T12	F: CACCg TAACAGCCTTTGCCTCCTGG	sgRNA targeting ItgaM Exon3 with lower efficiency.
	R: AAAC CCAGGAGGCAAAGGCTGTTA	
	С	
sgItgaM_ intron	F: CACCgATACTACATACTTCCTCTCA	sgRNA targeting ItgaM intron 2 SRF binding site.
2_T7	R: AAACTGAGAGGAAGTATGTAGTATC	
sgItgaM_intron	F: CACCgATACTTCCTCTCAAGGCTCA	sgRNA targeting ItgaM intron 2 SRF binding site.
2_T26	R:	
	AAACTGAGCCTTGAGAGGAAGTATC	
ChIP qRT-PCR pi	rimer sequence	
ChIP-ItgaM	F: GGCTGTCATCCATTGCTTCT	qRT-PCR primers for ChIP assay detecting SRF
-	R: ACCCTCAGCGAATCTCTCAA	binding in ItgaM

ChIP-	F: CTGGGAGGTGTTTGCATTTT	qRT-PCR primers for ChIP assay detecting SRF
ltgb2	R: CAATCCAGTGCCAAGGAAGT	binding in Itgb2
ChIP-LINE1	F: AAACGAGGAGTTGGTTCTTTGAG	qRT-PCR primers of negative control for ChIP
	R: TTTGTCCCTGTGCCCTTTAGTGA	assay
ChIP-Acta2	F: GAGGCCTGGGTCTCTTCCA	qRT-PCR primers for ChIP assay detecting
	R: GCTGAGCTGCCTCCTGTTTC	SRF/MAL binding in Acta2

## Supplementary Table 2. Antibodies used for FACS and immunofluorescence staining

Antibodies for flow cytometric a	ssay	
Antibody	Catalog number	Supplier or comments
APC-CD11b(human)	ICRF44, Cat# 301310	BioLegend, 1:100 dilution
APC-CD11b(mouse)	M1/70, Cat# 17-0112-82	eBioscience, 1:100 dilution
APC-CD18	C71/16, Cat# 562828	BD Pharmingen, 1:100 dilution
APC-CD11a	M17/4, Cat# 101119	BioLegend, 1:100 dilution
BV421-CD18	M18/2, Cat# 744597	BD OptiBuild, 1:100 dilution
V500-CD11b	M1/70, Cat# 562127	BD Horizon, 1:100 dilution
Biotin-c-Kit	2B8, Cat# 13-1171-85	eBioscience, 1:100 dilution
PE-CD11b	M1/70, Cat# 553311	BD Pharmingen, 1:100 dilution
PE-Cy7-Gr1	RB6-8C5, Cat# 108416	BioLegend, 1:100 dilution
Pacific Blue-B220	RA3-6B2, Cat# 103227	BioLegend, 1:100 dilution
APC-eFluor780-CD3e	17A2, Cat# 47-0032-82	eBioscience, 1:100 dilution
APC/Fire 750-CD3e	17A2, Cat# 100248	BioLegend, 1:100 dilution
FITC-CD45.2	104, Cat# 109806	BioLegend, 1:100 dilution
V500-CD45.2	104, Cat# 562130	BD Horizon, 1:100 dilution
APC-CD45.1	A20, Cat# 110714	BioLegend, 1:100 dilution
PE-Sca1(Ly-6A/E)	D7, Cat# 108108	BioLegend, 1:100 dilution
PE-Cy7-CD117(c-Kit)	2B8, Cat# 105814	BioLegend, 1:100 dilution
APC-CD135	A2F10, Cat# 135310	BioLegend, 1:100 dilution
BV421-CD34	RAM34, Cat# 562608	BD Horizon, 1:100 dilution
PerCP-Cy5.5-CD16/CD32	93, Cat# 101323	BioLegend, 1:100 dilution
Pacific Blue-CD117 (c-Kit)	2B8, Cat# 105820	BioLegend, 1:100 dilution
APC-CD150 (SLAM)	TC15-12F12.2, Cat# 115910	BioLegend, 1:100 dilution
APC-eFlour780-CD48	HM48-1, Cat# 47-0481-82	eBioscience, 1:100 dilution
PerCP-Cy5.5-CD127(IL-7Ra)	A7R34, Cat# 135022	BioLegend, 1:100 dilution
APC-CD184 (CXCR4)	2B11, Cat# 17-9991-80	eBioscience, 1:100 dilution
APC-CD117(c-Kit)	2B8, Cat# 17-1171-82	eBioscience, 1:100 dilution
Antibodies for immunofluoresce	ence staining	
Alexa Fluor 594 Phalloidin	Cat# A12381	Thermo Fisher Scientific, 165 nM
Alexa Fluor 647 anti-Tubulin-α	Cat# 627908	BioLegend, 1:300 dilution
Anti-MAL	Cat# sc-390324	Santa Cruz Biotechnology, 1:200 dilution
anti-Lamin B1	Cat# ab16048	Abcam, 1:200 dilution
Alex Fluor 594 goat anti-mouse	Cat# A-11005	Thermo Fisher Scientific, 1:400 dilution
lgG		
Alex Fluor 647 donkey anti-	Cat# A-31573	Thermo Fisher Scientific, 1:400 dilution
rabbit IgG		
Antibodies for other use		
PE-CD31	Cat# 12-0311-82	Thermo Fisher Scientific, 3µg/ml
anti-SRF	2C5, Cat# 61386	Active Motif, 3-5µg per ChIP
anti-MAL	Cat# sc-390324	Santa Cruz Biotechnology,3-5µg per ChIP
anti-GFP	B2, Cat# sc-9996	Santa Cruz Biotechnology, 3-5µg per ChIP

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