

Materials and Methods

miR-155 expression

miR-155 expression was detected according to the manufacturer's instructions using the miScript PCR System (QIAGEN, Valencia, CA) which comprises the miScript Reverse Transcription Kit, miScript SYBR Green PCR Kit, and miScript Primer Assay. To detect miR-155 expression in BMDMs, WT BMDMs (4×10^6 cells) were seeded into 6-well plates and treated with serum-free DMEM or LLC or B16-F10 cell conditioned medium. Total RNAs were extracted and cDNAs were prepared. miR-155 expression was detected by real-time PCR.

Table 1. Sequences of the primers used for Quantitative Real-Time PCR

Gene	Forward (5'-----3')	Reverse (5'-----3')
MMP2	ACACTGGGACCTGTCACTCC	GCGAAGAACACAGCCTTCTC
MMP9	CATTCGCGTGGATAAGGAGT	ACCTGGTTCACCTCATGGTC
PIGF	TGACCTGGCTGTGTATCTGC	AAAACGTTTCCAGCACCAAC
Arg1	TCACCTGAGCTTTGATGTCG	CACCTCCTCTGCTGTCTTCC
HIF-1α	TCAAGTCAGCAACGTGGAAG	TATCGAGGCTGTGTGCGACTG
Bcl-6	CCTGAGGGAAGGCAATATCA	CGGCTGTTCAGGAACCTTTC
c-Maf	AAGGAGGAGGTGATCCGACT	TCTCCTGCTTGAGGTGGTCT
SOCS1	TTAACCCGGTACTCCGTGAC	GAGGTCTCCAGCCAGAAGTG
18S	CGCGGTTCTATTTTGTGGT	AGTCGGCATCGTTTATGGTC

Figure legends

Supplementary Fig. S1. miR-155 expression in spleen of WT and miR-155^{-/-} chimeric mice. Quantitative real-time PCR was performed to detect miR-155 expression in spleen of WT and miR-155^{-/-} chimeric mice. Data are represented as the mean \pm SEM of 8 mice. * $p < 0.05$ by Student's *t* test.

Supplementary Fig. S2. Effects on bone marrow miR-155 deficiency on bone marrow cell mobilization in tumor-bearing mice. Flow cytometric analysis of bone

marrow cells from WT and miR-155^{-/-} chimeric mice using antibodies for specific surface markers. Data are presented as the mean ± SEM of 8 mice.

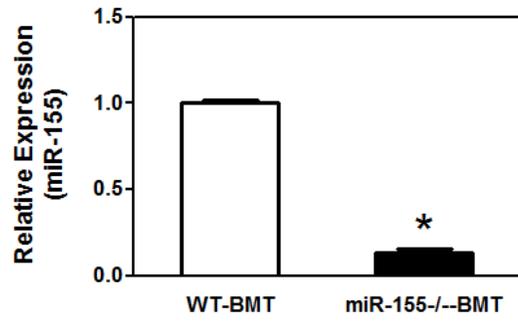
Supplementary Fig. S3. The effects of bone marrow miR-155 deficiency on the leukocyte subpopulations in spleen of tumor-bearing mice. **A.** The percentage of leukocytes in spleen from WT and miR-155^{-/-} chimeric mice was analyzed by flow cytometry using antibodies for specific surface markers. **B.** The absolute numbers of leukocytes in spleen were calculated and shown. Data were presented as the mean ± SEM of 8 mice. **p*<0.05 by Student's *t* test.

Supplementary Fig. S4. The effects on bone marrow miR-155 deficiency on the expression of tumor-derived factors. MMP2, MMP9 and PIGF mRNA levels were measured by quantitative real-time PCR in tumor tissues of WT and miR-155^{-/-} chimeric mice 14 days after LLC inoculation. Data were presented as the mean ± SEM of 8 mice. **p*<0.05 by Student's *t* test.

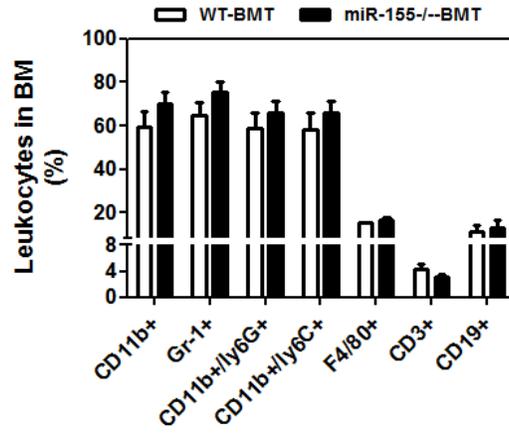
Supplementary Fig. 5. A, Expression level of miR-155 in LLC and B16-F10 cells. Quantitative real-time PCR was performed to measure the expression of miR-155 in the lung tissue of WT mice, and cultured LLC and B16-F10 cells. Data are presented as the mean ± SEM of three replicates. **, *p*<0.001 by Student's *t* test. **B.** Quantitative real-time PCR analysis of miR-155 levels in WT macrophages incubated with serum-free medium (SFM), LLC cell conditioned medium (LLC-CM) and B16-F10 cell conditioned medium (B16-CM) for 24 h. Data are presented as the mean ± SEM of three replicates.

Supplementary Fig. 6. miR-155 deficiency MΦ promoted migration of B16-F10 cells. **A,** Transwell assay of B16-F10 cells migration towards WT and miR-155^{-/-} MΦ treated with B16-CM. Data were presented as the mean ± SEM of 20 fields/group. *, *p*<0.05 by Student's *t* test. **B,** Representative fluorescence images of migrated B16-F10 cells. DAPI was used to stain the nuclei. Magnification, 40 x.

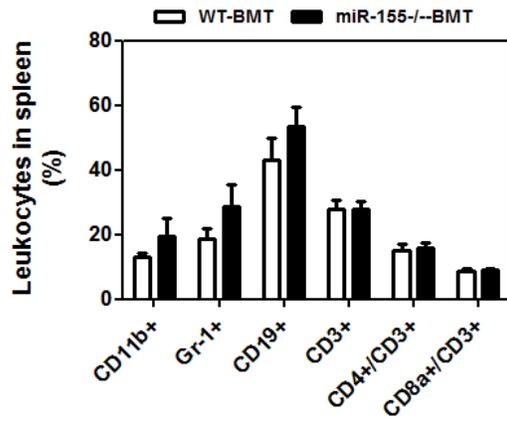
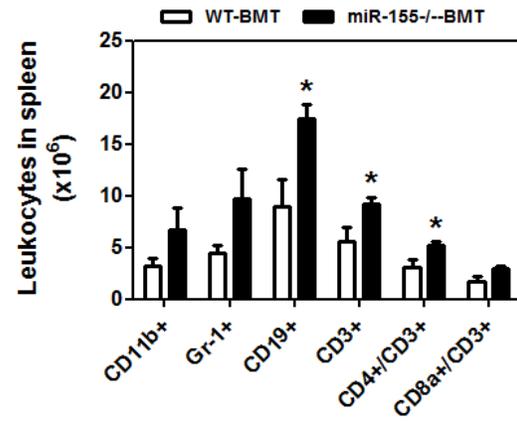
Supplementary Fig. 7. A. Transfection of mir155 inhibitor resulted in high expression level of its target genes. LLC cells were transfected with Negative control (con inhibitor) and mir155 inhibitor for 48 h and Quantitative real-time PCR was performed to measure the expression levels of HIF-1 α , Bcl-6, c-Maf and SOCS1. Data are presented as the mean \pm SEM of three replicates. **, $p < 0.001$ by Student's t test. **B.** [^3H] Thymidine incorporation assay of B16-F10 proliferation. B16-F1- cells were cocultured with WT M Φ or miR-155-/- M Φ for 48 h. [^3H] Thymidine (1 $\mu\text{Ci}/\text{well}$) was added and further incubated for 4 h. Cultures were harvested and thymidine incorporation was measured by scintillation counting. Data are expressed as CPM (mean \pm SEM) of six cultures. Three independent experiments were performed. *, $p < 0.001$ by Student's t test.

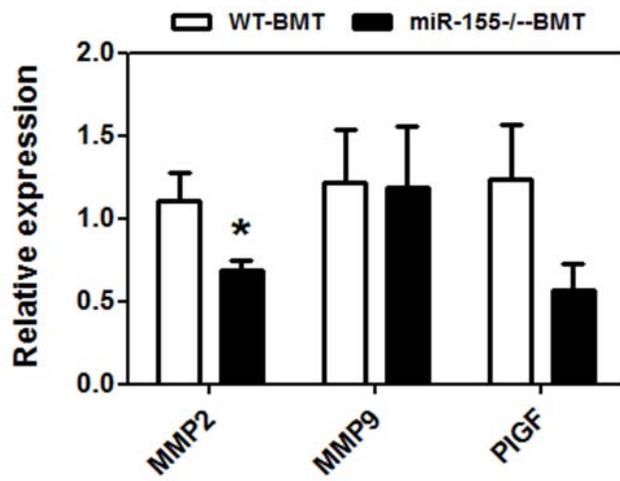


Suppl. Fig. 1

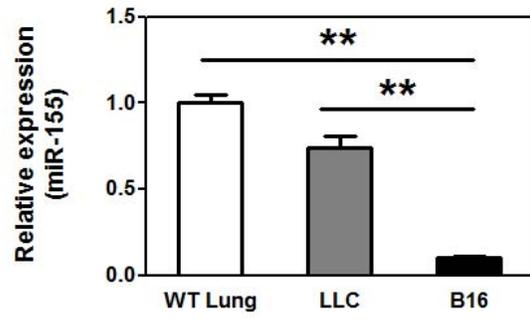
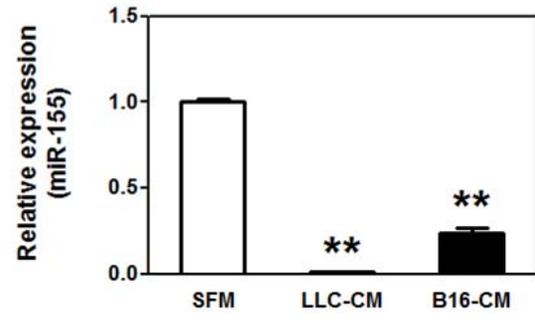


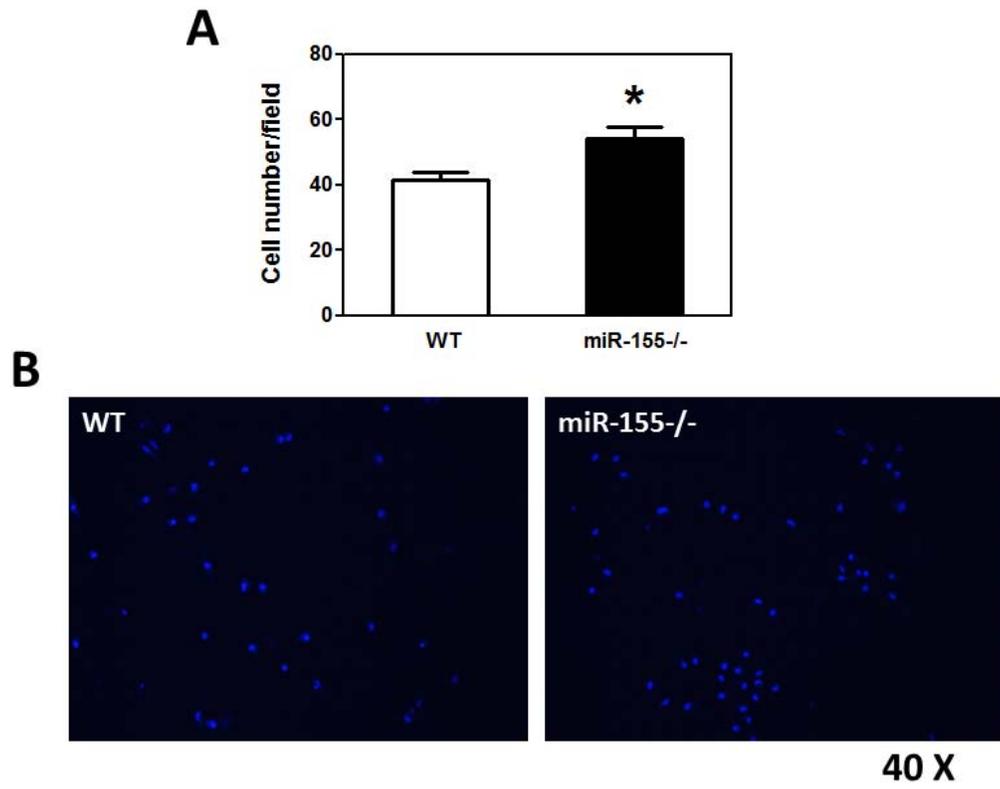
Suppl. Fig. 2

A**B****Suppl. Fig. 3**

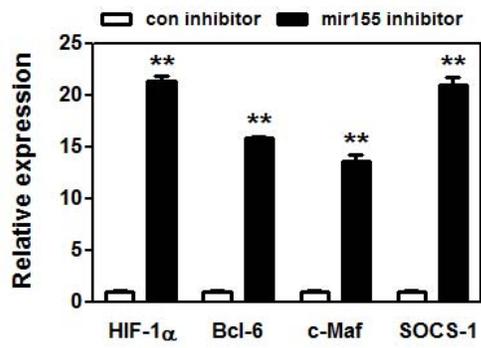
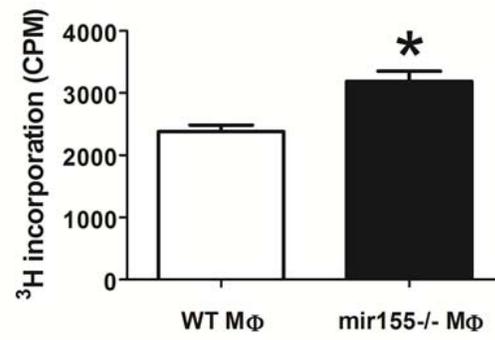


Suppl. Fig. 4

A**B****Suppl. Fig. 5**



Suppl. Fig. 6

A**B****Suppl. Fig. 7**