Supplemental Information

Cancer-secreted exosomal miR-1468-5p promotes

tumor immune escape via the immunosuppressive

reprogramming of lymphatic vessels

Chenfei Zhou, Wenfei Wei, Jing Ma, Yang Yang, Luojiao Liang, Yanmei Zhang, Zici Wang, Xiaojing Chen, Lei Huang, Wei Wang, and Sha Wu
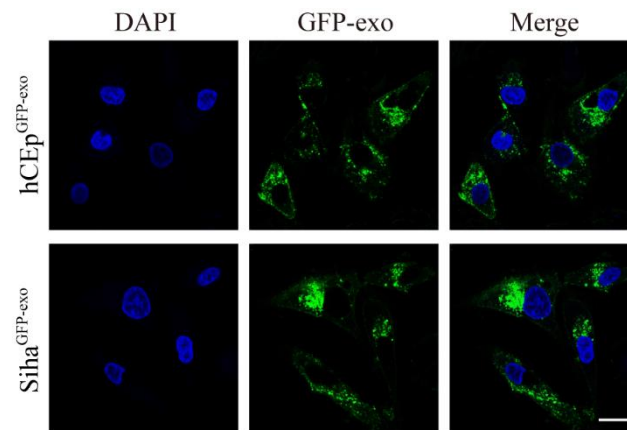
Supplemental figures and legends

Figure S1

Supplementary Figure S1. Siha and hCEp successfully were transfected with lentiviral-CD63-GFP, related to Fig. 3. Representative micrographs of Siha and hCEp transfected with lentiviral-CD63-GFP.
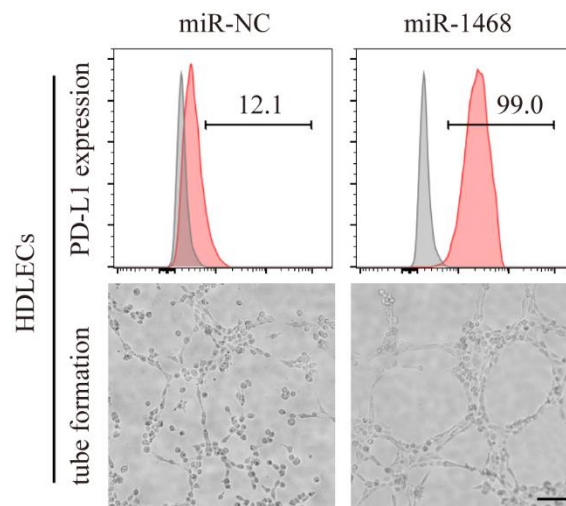

Figure S2

Supplementary Figure S2. miR-1468-5p mimics contributed to PD-L1 expression and tube formation of HDLECs, related to Fig. 4. Flow cytometry analysis of PD-L1 expression and tube formation assay in HDLECs treated with miR-1468-5p mimics
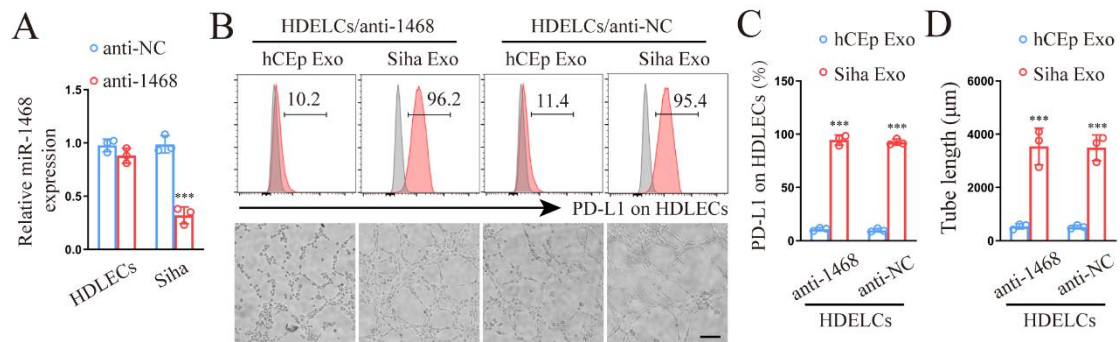
11   or NC. Scale bar, 10 µm.

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Figure S3

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14   **Supplementary Figure S3. Cancer cells reprogrammed HDLECs by transferring**

15   **exosomal miR-1468-5p to HDLECs, related to Fig. 4.** (A) qRT-PCR analysis of miR-

16   1468-5p expression in HDLECs and Siha both stably transfected with lentiviral anti-

17   1468-5p or NC. (B) Flow cytometry analysis of PD-L1 expression and tube formation

18   assay in HDLECs/anti-1468-5p or HDLECs/anti-NC in the presence of hCEp-exo or

19   Siha-exo. Scale bar, 10 µm. (C and D) Quantification of PD-L1 expression (C) and tube

20   formation (D) in HDLECs/anti-1468-5p or HDLECs/anti-NC in the presence of hCEp-

21   exo or Siha-exo. Error bars represent the mean ± SD of three independent experiments.

22   ***, P<0.001.

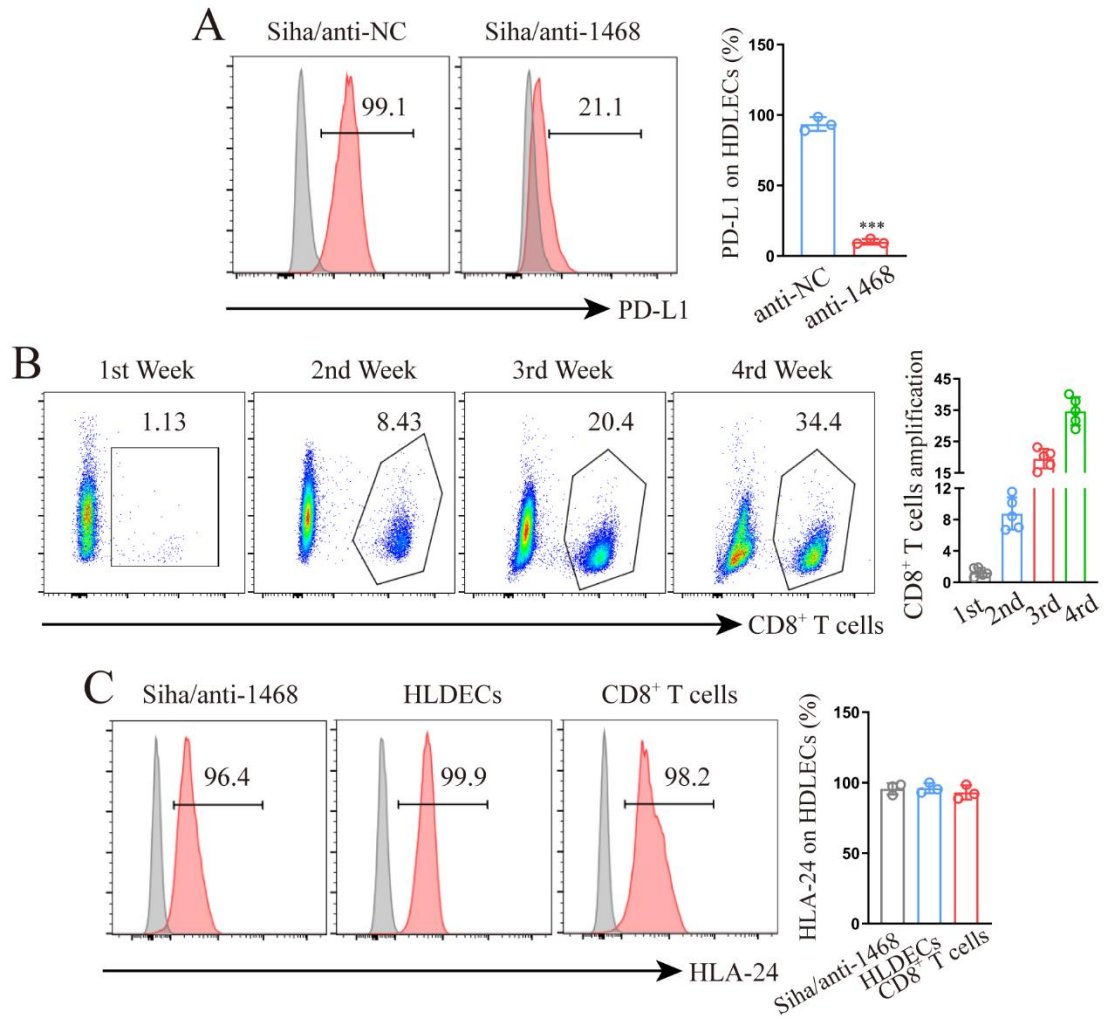23

Figure S4

Supplementary Figure S4. Flow cytometry analysis of PD-L1 and HLA-A24 expression, related to Fig. 5. (A) Flow cytometry analysis of PD-L1 expression on Siha/anti-NC and Siha/anti-1468-5p. (B) Flow cytometry analysis of CD8$^+$ T cells amplification *in vivo* at indicated time point. (C) Flow cytometry analysis of HLA-A24 expression on Siha/anti-1468-5p, HDLECs, and CD8$^+$ T cells. Error bars represent the mean $\pm$ SD of three independent experiments. ***, P<0.001.
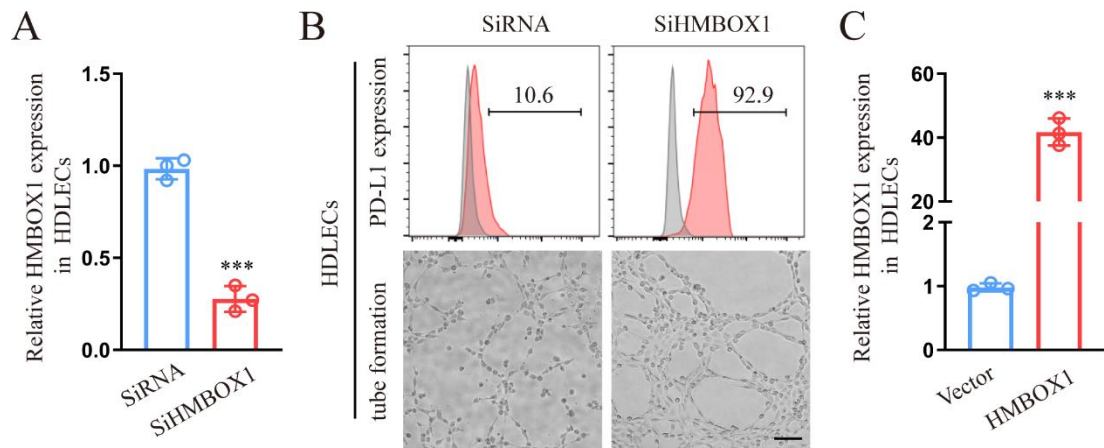
Figure S5

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33 **Supplementary Figure S5. miR-1468-5p directly targets HMBOX1 in HDLECs**

34 **reprogramming, related to Fig. 6.** (A) qRT-PCR analysis of HMBOX1 expression in

35 HDLECs transfected with SiHMBOX1 or SiRNA. (B) Flow cytometry analysis of PD-

36 L1 and tube formation assay in HDLECs treated with SiHMBOX1 or SiRNA. Scale

37 bar, 10 μm. (C) qRT-PCR analysis of HMBOX1 expression in HDLECs transfected

38 with HMBOX1 or vector. Error bars represent the mean $\pm$ SD of three independent
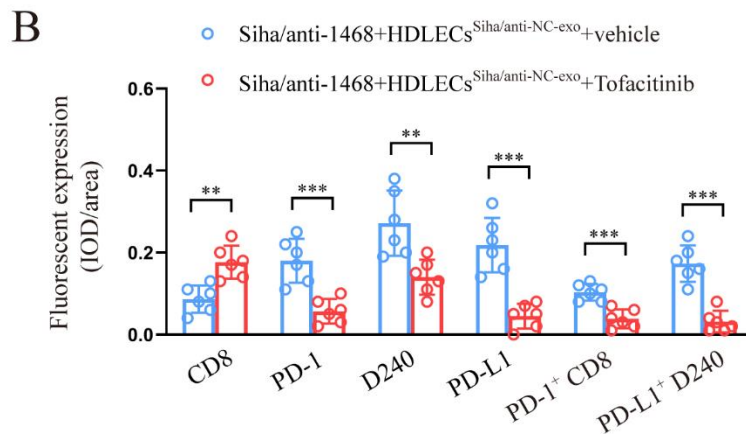
39 experiments. ***, $P<0.001$.

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Figure S6

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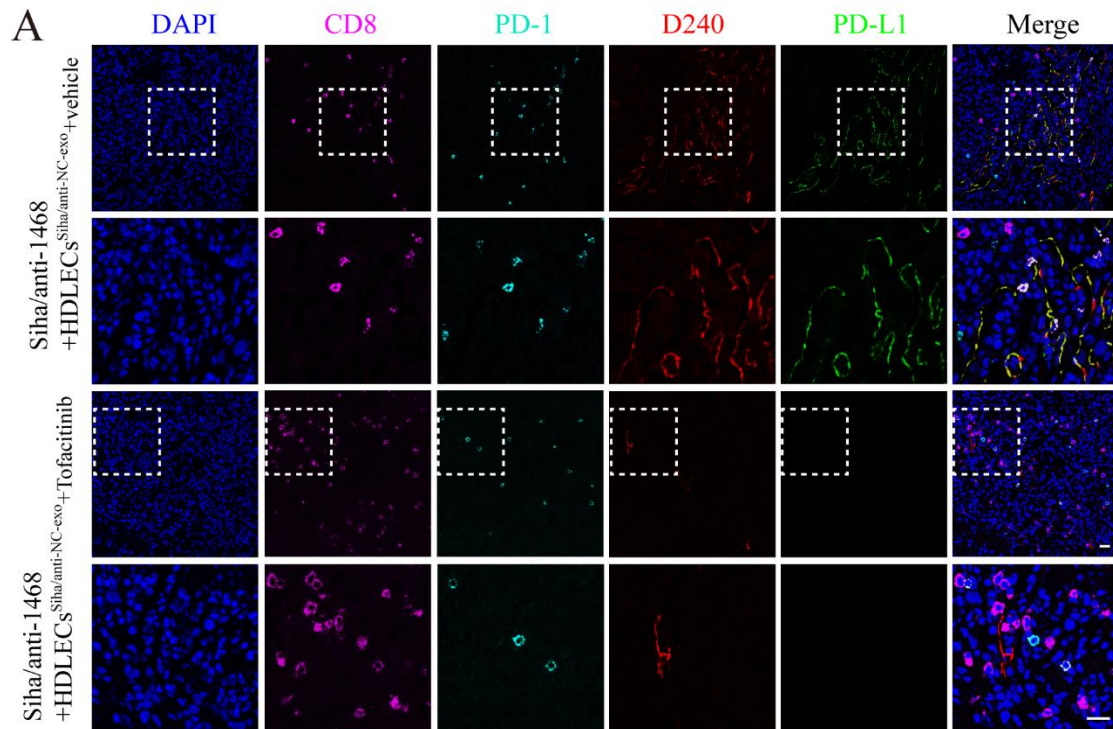
Supplementary Fig S6. JAK/STAT inhibitor neutralizes the effect of exosomal

miR-1468-5p-induced CD8$^+$ T cell immunosuppression *in vivo*, related to Fig. 7.

(A) Representative micrographs of CD8, PD-1, lymphatics, and PD-L1

immunofluorescence staining in sections of xenograft tumors. Scale Bar: 20 μm. (B)

Quantification of CD8, PD-1, lymphatics, PD-L1, PD-1$^+$ CD8, and PD-L1$^+$ lymphatics

expression in indicated groups. Error bars represent the mean ± SD of three independent

48   experiments. **, P<0.01; ***, P<0.001.

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50   **Supplementary Tables**
51   **Table S1. Detailed primer sequences in the study**

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	Forward	Reverse
MCMBP	CTGCCCAGCAATACTTCCCTT	TTCTGCCACGTTATGAGGTTG
ZNF501	AACTTTCCGCAAACAAGCACA	TCCCACATCCAACACACTCATA
HMBOX1	CACCAACTCGCTACCATGCAA	TCCGATTGGCAAGAAAGGCTT
PRRT2	TTCTGTCTGAGAGTGTAGGGG	CAGGCTACCTCGGGGAGAT
SV2A	TATTCCCGAAGATCGTACTCCC	GCCCTCAGTAGCATCACTGG
MLH3	TCTCTCACTCATGCACCCTTC	TCGGGAACATACGTCTTTGGT
LCORL	GAGGAGCTATCATCTCAGGGC	TGAGCAACTAGGGGAATGTTAGG
GAPDH	CCATCAATGACCCCTTCATTGACC	GAAGGCCATGCCAGTGAGCTTCC
SiARID2	GGAGAUGGUUCUCAUUUAATT	UUAAAUGAGAACCAUCUCCTT
SiRNA	GCAGUUGUUGCACAGGUAATT	UUACCUGUGCAACAACUGCTT
HBS1	CTTCCAGGAGAGAAGCCGTCT	GCTGTAATTTGCACACGTGTTTGTG
HBS2	CTCGCACGACAATCGGAATCAC	CAGATTGGGAGCTCTGGACCT
HBS3	CCAGCCTCAGTTTCTTCCGC	CGCCCTGCTGTCCCGAA
HBS4	TCTGAGAGCTGTCTGTAGAGGGC	CTACTGCTCCGTCAGGCATC

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