Induction of VEGFA and Snail-1 by meningitic *Escherichia coli* mediates disruption of the blood-brain barrier

Supplementary Materials

**Supplementary Figure S1:** The inhibition of VEGFR on meningitic *E. coli* invasion of the hBMEC. VEGFR inhibitor motesanib significantly decreased meningitic *E. coli* PCN033 invasion of hBMEC in a dose-dependent manner.

**Supplementary Figure S2:** The induction of proinflammatory factors by non-meningitic *E. coli* strain HB101. The transcriptional changes of cytokines and chemokines in hBMEC upon HB101 infection.
**Supplementary Figure S3:** *E. coli* K1 strain RS218 induction of VEGFA and Snail-1 requires the activation of MAPK-ERK1/2 signaling. Effects of the MAPK signaling inhibitors on the *E. coli* K1 strain RS218-induced upregulation of VEGFA and Snail-1. (A) The upregulation of VEGFA could be significantly inhibited by the treatment of U0126 (selective inhibitor of ERK1/2) and SB202190 (selective inhibitor of p38). (B) The upregulation of Snail-1 could be significantly blocked by U0126. Both upregulation of VEGFA and Snail-1 were not affected by the treatment of SP600125 (specific inhibitor of JNK).

**Supplementary Figure S4:** Regulation of VEGFA and Snail-1 by non-meningitic *E. coli* strain HB101. (A) Real-time PCR analysis of the VEGFA transcription in hBMEC in response to HB101 infection at the MOI of 10. GAPDH was used as the internal reference for the normalization. (B) Determination of the secretory VEGFA in the hBMEC culture supernatant in response to HB101 by ELISA. (C) Real-time PCR analysis of Snail-1 transcription in hBMEC in response to HB101 challenge.
**Supplementary Table S1: Primers used for the real-time PCR in this study**

| Primers | Nucleotide sequence (5’-3’) | Amplified fragments |
|---------|-----------------------------|---------------------|
| P1      | TGTGGAAGAGGATGAAGATGAAGA    | Human ZO-1          |
| P2      | GGTTGGAAGATGTCTGGTGTC       |                     |
| P3      | ACAAGCCACAAGATTACAAG        | Human β-catenin     |
| P4      | ATCAGCAGTCTCATCCAA          |                     |
| P5      | TTAACCTCGCCTGTGGAT          | Human Occludin      |
| P6      | TGTGTAGCTGTCTCATAGTG        |                     |
| P7      | CGCCTCTCCTGCCACAACAT        | Human Claudin-5     |
| P8      | CCAGCACCGAGTCGTACACTT       |                     |
| P9      | TGCCCTGCTCTCCTACCT          |                     |
| P10     | GACATCCATGAACCTCACCACCTT    | Human VEGFA         |
| P11     | ACTCGGATGTGAAGAGATAC        | Human Snail-1       |
| P12     | AGACTCTTTGGTGCTTTGT         |                     |
| P13     | CCTTCGGTCAATGGCTCTTCT       | Human IL-6          |
| P14     | GAGGTGAGTGGCTGTCTGTG        |                     |
| P15     | AGGATATGGAGCAACAAGT         | Human IL-1β         |
| P16     | GCAGGACAGGTACAGATT          |                     |
| P17     | AATGGCGTGAGCGTGAGA           | Human TNF-α         |
| P18     | TGGCACAGAGAGGGAGGAGGC       |                     |
| P19     | ATAGCAAGCCACCTCATT          |                     |
| P20     | GCTTCTTGGGACACTTG           | Human MCP-1         |
| P21     | AGTGTGAAGGTGAAGGTCC         |                     |
| P22     | CTTTCTGCCATCTCTTGAG         |                     |
| P23     | TGCTGTCTCTCTCTCTAGTA        |                     |
| P24     | TGTGGCTATGTACTTGCGTTTGG     | Human GRO-α         |
| P25     | TGCCCTCTCGACCAACACT         |                     |
| P26     | CGCCTGTCTCACCACCTCTC        | Human GAPDH         |
| P27     | GCCCTCCTCCTCTGATACCTCCCTC  |                     |
| P28     | CTGGCAGTGTACTCACTCTTTC      | Mouse ZO-1          |
| P29     | AGCCACAGATTACAAGAAGA        |                     |
| P30     | CCAATGTCCAGTCCAAAGA         | Mouse β-catenin     |
| P31     | ATGGAGGCTATGGCTATGG         |                     |
| P32     | GGAAGCGAGATGAAAGGAGA        | Mouse Occludin      |
| P33     | AAGGGCCTCATGGGCTGTGGTG      |                     |
| P34     | CTGGCTGTGGGCTGAGCTGGGA      | Mouse Claudin-5     |
| P35     | CCATTCTCTGCTCCACT           |                     |
| P36     | TGGCAGTGATCTCTTCCACA        | Mouse Snail-1       |
| P37     | CACTGCGCATCTCTCTCCTCCC      |                     |
| P38     | CAATAGTGATGACCTGGCCGT       | Mouse β-actin       |