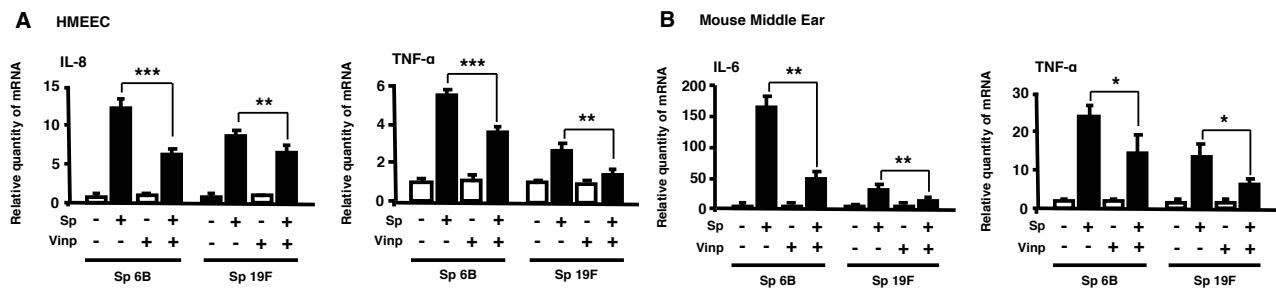## Supplemental Data

Vinpocetine suppresses *Streptococcus pneumoniae*-induced inflammation via inhibition of ERK1 by CYLD

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Supplemental Figure 1

Supplemental Figure 1. Vinpocetine suppresses *S. pneumoniae* 6B, 19F-induced inflammatory response *in vitro* and *in vivo*.

(**A**) Human middle ear epithelial cells HMEEC were pre-treated with vinpocetine (10 μM) for 1 h, followed by stimulation with *S. pneumoniae* 6B, 19F for 6 h. Relative quantity of IL-8 and TNF-α mRNA expression was measured by real-time Q-PCR analysis. (**B**) Mice were pre-treated with vinpocetine (10 mg/kg, i.p.) for 2 h and inoculated transtympanically with *S. pneumoniae* 6B, 19F (5×10^6 CFU per mouse) for 9 h. IL-6 and TNF-α mRNA expression in the middle ear of mice was measured by real-time Q-PCR analysis. Data are mean ± SD (*n* = 3). *p<0.05, **p<0.01, ***p<0.001*. Statistical analysis was performed using Student's *t*-test. Data are representative of three or more independent experiments. Sp, *S. pneumoniae*. Vinp, vinpocetine.

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Supplemental Table 1. Primers for real-time quantitative RT-PCR (Q-PCR) analysis

Primer name	Forward (5'-3')	Reverse (5'-3')
human IL-1β	AAACAGATGAAGTGCTCCTTCCAGG	CTTGTCCATGGCCACAACAACTGAC
human IL-6	AAATTCGGTACATCCTCGACGGCA	AGTGCCTCTTTGCTGCTTTCACAC
human IL-8	TCCTGATTTCTGCAAGCTCTG	GTCCACTCTCAATCACTCTCAG
human TNF-α	CCCAGGCAGTCAGATCATCTT	AGCTGCCCCTCAGCTTGA
human CYLD	GTTGACTCACAACCACAATCC	GTGAAGAACGGTCAAAGTCTGTAG
human Cyclophilin A	CGGGTCCTGGCATCTTGT	GCAGATGAAAAACTGGGAACCA
mouse IL-1β	CAACCAACAAGTGATATTCTCCATG	GATCCACACTCTCCAGCTGCA
mouse IL-6	GAGGATACCACTCCCAACAGACC	AAGTGCATCATCGTTGTTCATACA
mouse MIP-2	CCTGCCAAGGGTTGACTTCA	TTCTGTCTGGGCGCAGTG
mouse TNF-α	CATCTTCTCAAAATTCGAGTGACAA	TGGGAGTAGACAAGGTACAACCC
mouse CYLD	CCTGGCTTTTCTTTGACAGC	AAGAGTGCAGGTCCTCCAGA
mouse GAPDH	ACCCAGAAGACTGTGGATGG	GGATGCAGGGATGATGTTCT