

Supplemental materials:

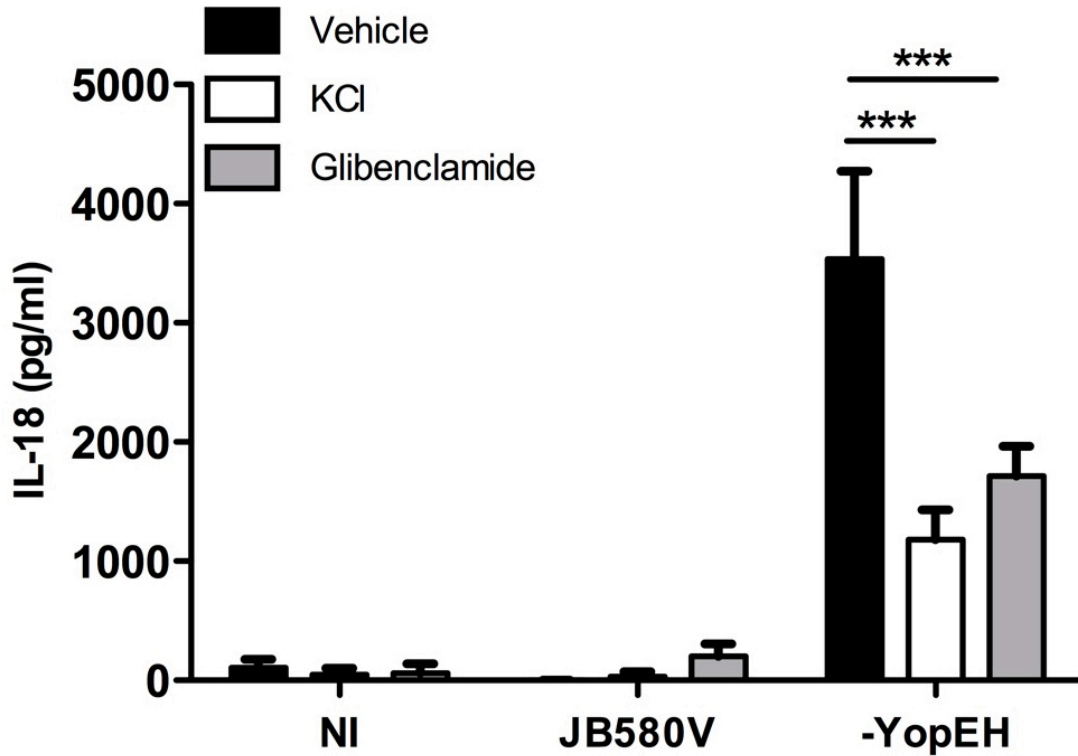


Figure S1. IL-18 secretion is dependent on K⁺ efflux. (Related to Figure 2) Caco-2 cells were treated with 50 μ M glibenclamide, 130mM KCl, or DMSO for 30 min before infection with wild type or *yopEH* mutant *Y. enterocolitica*. Cell culture supernatants were collected post infection and analyzed for secreted IL-18 by ELISA. The data represent the mean \pm SD of triplicate samples from two independent experiments. *p < 0.001 versus vehicle.

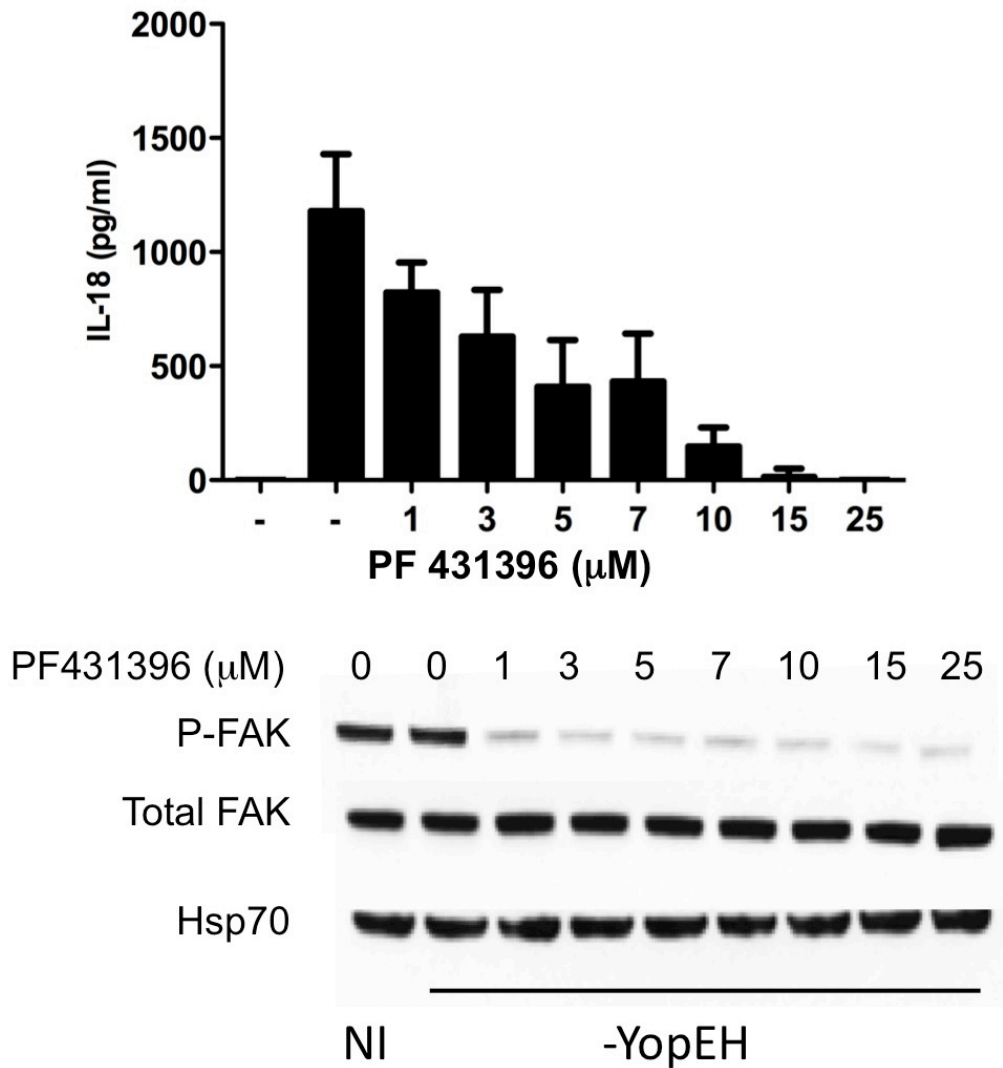


Figure S2. IL-18 secretion is dependent on the activity of FAK. (Related to Figure 5) Caco-2 cells were pre-treated with increasing concentrations of the FAK inhibitor PF-431396 or DMSO (0 μ M) for 1h and then infected with indicated bacterial strains at an MOI of 10. After 8 h, ELISA determined the concentration of IL-18 in the conditioned media. Cells were also lysed and subjected to immunoblot analysis of phospho-FAK, total FAK, and the loading control Hsp70. Data represents mean \pm SD of two independent experiments. *p < 0.001.

Table S I: Bacterial strains for infection studies

Strain	Description	Source or Reference
JB580V	Wild-type <i>Y. enterocolitica</i> 8081v - <i>yenR</i> (r ⁻ m ⁺)	(Kinder et al., 1993)
JB580C	JB580V cured of virulence plasmid pYVe8081; lacks Ysc/Yop T3SS	(Pepe and Miller, 1993)
-YopP	JB580V <i>yopP</i>	(Bose et al., 2012)
-YopE	JB580V <i>yopE</i>	This work
-YopH	JB580V <i>yopH</i>	This work
-YopEH	JB580V <i>yopEH</i>	This work
-YopQ	JB580V <i>yopQ</i>	This work
-Inv	JB580V <i>inv</i>	gift from Virginia Miller (UNC Chapel Hill)
JB580C -Inv	JB580C <i>inv</i>	This work
-YopEH -Inv	JB580V <i>yopEH inv</i>	This work
STm	<i>Salmonella enterica</i> subsp. <i>enterica</i> (ex Kauffmann and Edwards) Le Minor and Popoff serovar Typhimurium	(ATCC® 14028™)
<i>E. coli</i> Inv	HB101 (pMV101) <i>E. coli</i> expressing <i>Y. enterocolitica</i> Invasin	(Pepe and Miller, 1990); gift from Virginia Miller (UNC Chapel Hill)
<i>E. coli</i> vector	HB101 pMV101	(Pepe and Miller, 1990);
<i>E. coli</i> InvD760A	<i>E. coli</i> expressing <i>inv</i> with D760A	This work
<i>E. coli</i> InvD809A	<i>E. coli</i> expressing <i>inv</i> with D809A	This work

Table S II: Primers used in this study:

Oligo sequence 5'-3'	
YopE- Upstream	YopE- Downstream
Forward: <u>GTCGACGATGAGGTTTGTAGCCGT</u> TC	Forward: <u>TTAATTAAGTGATATGGTGACTAGTCC</u> TGC
Reverse: <u>TTAATTA</u> ACTTAGTGGGAAAATAG CCGG	Reverse: CCCTGCCTCTTACTCTACTTC
YopH- Upstream	YopH-Downstream
Forward: <u>GTCGACGATAATACCCCCAGGTTG</u> A	Forward: <u>TTAATTAAGGACAAGGGCGACCATTA</u> TT
Reverse: <u>TTAATTA</u> GGTGAGCCGTGTATTTTCAT	Reverse: CTGGCACCTATCAATGAGAAGG
YopQ- Upstream	YopQ- Downstream
Forward: <u>GTCGACGATGCAGAACTGGCGAAT</u> AC	Forward: <u>TTAATTA</u> ACTACTCTCAATGAGCTTCC CATG
Reverse: <u>TTAATTA</u> GAGATCTTCCGGCATACT GAC	Reverse: GTAGTGTCCGTGAATACTGTCC
Primers used for RT-PCR	
Oligo sequence 5'-3'	

GAPDH
Forward: GTCAGTGGTGGACCTGACCT
Reverse: AGGGGTCTACATGGCAACTG
Caspase-1
Forward: ATCCGTTCCATGGGTGAAGGTACA
Reverse: CAAATGCCTCCAGCTCTGTA
NLRP3
Forward: TTCTTTCTGTTTGCTGAGTTTTTG
Reverse: TTCCTGGCATATCACAGTGG
Primers used for invasin site-directed mutagenesis
Oligo sequence 5'-3'
Inv D760A
Forward: GCCAGCGCACTGCCTTTACTGCGCTGATC
Reverse: GATCAGCGCAGTAAAGGCAGTGCCTGGC
Inv D809A
Forward : GACCAGTACAGCCTTTGTCACCTATGG
Reverse: CCATAGTGACAAAGGCTGTACTGGTC