Immunogen <sup>a</sup>	Description	Peptide	Dose used (μg)	Immunizations tested by ELISA <sup>b</sup>
f8-5	WT phage	-	10	1 <sup>st</sup> , 3 <sup>rd</sup> , 5 <sup>th</sup>
Δ3	Truncated pIII N- terminus	-	10	1 <sup>st</sup> , 3 <sup>rd</sup> , 5 <sup>th</sup>
Δ8	Altered pVIII ID residues	-	10	1 <sup>st</sup> , 3 <sup>rd</sup> , 5 <sup>th</sup>
Δ8Δ3	pIII truncation and pVIII alteration	-	10	1 <sup>st</sup> , 3 <sup>rd</sup> , 5 <sup>th</sup>
f8-5/MD10rec	-	MD10	10	$2^{nd}$ , $4^{th}$
∆3/MD10rec	-	MD10	10	2 <sup>nd</sup> , 4 <sup>th</sup>
f8-5/MD10conj		MD10	2	2 <sup>nd</sup> , 4 <sup>th</sup>
∆3/MD10conj		MD10	2	2 <sup>nd</sup> , 4 <sup>th</sup>
f8-5/4E10, <sup>A</sup> rec	-	4E10, <sup>A</sup>	10	2 <sup>nd</sup> , 4 <sup>th</sup>
∆3/4E10 <sub>L</sub> <sup>A</sup> rec	-	4E10 <sup>-A</sup>	10	2 <sup>nd</sup> , 4 <sup>th</sup>
f8-5/4E10, <sup>B</sup> conj		4E10, <sup>B</sup>	2	2 <sup>nd</sup> , 4 <sup>th</sup>
∆3/4E10 <sub>L</sub> <sup>₿</sup> conj		4E10 <sup>⁻</sup>	2	2 <sup>nd</sup> , 4 <sup>th</sup>
f8-5/SPDP	-	-	2	2 <sup>nd</sup> , 4 <sup>th</sup>
∆3/SPDP	-	-	2	2 <sup>nd</sup> , 4 <sup>th</sup>

 Table S1. Immunogen composition and immunization schedule.

<sup>a</sup> All immunogens were administered in 100  $\mu$ l PBS, *via* subcutaneous route, without adjuvant, every two weeks. Suffix "rec" indicates that the peptide was recombinantly fused to pVIII and "conj" indicates that a synthetic version of the peptide was chemically conjugated to the phage coat.

<sup>b</sup> Bleeds were taken two weeks after each immunization.

- Supplemental Figure S1 Average half-maximal Ab titers against each phage variant (f8-5,  $\Delta$ 3,  $\Delta$ 8, and  $\Delta$ 8 $\Delta$ 3) produced by mice immunized five times with a single variant (f8-5,  $\Delta$ 3,  $\Delta$ 8, or  $\Delta$ 8 $\Delta$ 3). Error bars show standard error of the arithmetic mean. Note that the Y-axis is log scaled.
- Supplemental Figure S2 Serum Ab reactivity with MD10 peptide and *S. flexneri* LPS after immunization with phage displaying recombinant MD10 peptide, phage treated with SPDP crosslinker alone, or unmodified phage. (A) Average anti-MD10 peptide and anti-*S. flexneri* LPS ELISA OD<sub>405-490</sub> signals at 1:50 serum dilutions after two and four immunizations with f8-5/MD10rec or Δ3/MD10rec recombinant phage. (B) Average anti-phage (f8-5 or Δ3), anti-SPDP, and anti-*S. flexneri* LPS ELISA OD<sub>405-490</sub> signals at 1:50 serum dilutions with f8-5 phage, Δ3 phage, f8-5/SPDP phage or Δ3/SPDP phage. Error bars show standard error of the arithmetic mean. All anti-phage signals, and possibly the highest anti-LPS signals, are at saturation.
- Supplemental Figure S3 Titration ELISA of MAbs SYA/J6 or 4E10 on: (A) f8-5 and Δ3 phage displaying recombinant MD10 peptide; (B) MD10 synthetic peptide conjugated to f8-5 and Δ3 phage via the SPDP crosslinker; (C) f8-5 and Δ3 phage displaying recombinant 4E10<sup>A</sup> peptide; (D) 4E10<sup>B</sup> synthetic peptide conjugated to f8-5 and Δ3 phage via the SPDP crosslinker.
- Supplemental Figure S4 SDS-PAGE analysis of (A) MD10 synthetic peptide conjugated to phage *via* the f8-5 and Δ3 SPDP crosslinker or (B) 4E10<sup>B</sup><sub>L</sub> synthetic peptide conjugated to f8-5 and Δ3 phage *via* the SPDP crosslinker. (A) Lane 1: f8-5, lane 2: Δ3, lane 3: f8-5/MD10conj, lane 4: Δ3/MD10conj, lane 5: Δ3/SPDP-Cys, lane 6: f8-5/MD10 no SPDP crosslinker control; (B) Lane 1: f8-5, lane 2: Δ3, lane 3: f8-5/4E10<sup>B</sup><sub>L</sub> conj, lane 4: Δ3/4E10<sup>B</sup><sub>L</sub> conj, lane 5: f8-5/4E10<sup>B</sup><sub>L</sub> no SPDP crosslinker control, lane 6: Δ3/SPDP-Cys.



Supplemental Figure S1





□ SA + bio-MD10 peptide

**(B)** 1.2 1 0.8 OD<sub>405-490</sub> 0.6 0.4 0.2 0 ELISA antigen: f8-5 LPS Δ3 LPS f8-5 SPDP LPS Δ3 SPDP LPS Immunizing f8-5/SPDP f8-5 Δ3 Δ3/SPDP Phage:

Supplemental Figure S2





Supplemental Figure S3



MD10 Phage Conjugate

4E10<sup>B</sup> Phage Conjugate