

Supplementary data

Table S1. Protein identification by LC-nanoESI-MS/MS.

Band	Protein name (species)	Acc. No.	MS/mps*	Sequence coverage
1	Integration Host Factor Protein alpha (<i>Escherichia coli</i>)	2IIE_A	701/22	54%
2	HU protein beta (<i>Escherichia coli</i>)	NP_286182	656/11	65%

* MS/mps: Mowse Score/matched peptides.

Supplementary Figure Legends

Figure S1. A His-pulldown assay found interactions between DMP12 and two DNA binding proteins. When total extracted *E.coli* proteins were used as prey, the N-terminal His₁₀-tagged DMP12 bait pulled down two DNA binding proteins. LC-nano ESI-MS/MS identified these proteins as HU beta (HUB) and IHF alpha (Table S1 for protein details).

Figure S2. BS3 Cross-linking assays only confirmed the interaction between DMP12 and *Neisseria* HU. Protein-protein interactions produced a clear band only in the DMP12/HU reaction. Conversely there is no evidence for DMP12-IHF cross-linking because the faint bands in the DMP12-IHF columns are not significantly stronger than the ~ 38 kDa band in the IHF-only control. White asterisks indicate the shifted, cross-linked bands.

Figure S3. Sedimentation velocity (SV) analysis of DMP12/*Neisseria* HU complex by analytical ultracentrifugation. For sedimentation velocity (SV) analysis, all samples were diluted to a suitable concentration (OD 280 absorption between 0.1~0.8) using 20 mM Tris pH 7.4 buffer and 5 mM MgCl₂. All analytical ultracentrifugation analyses were performed at 45000 rpm using a 4-hole AnTi60 rotor at 20 °C in a Beckman Optima XL-1 AUC equipped with absorbance optics (OD 280 nm). Data

were analyzed using the $c(s)$ distribution of the Lamm equation solutions calculated by the program SEDFIT (<http://www.analyticalultracentrifugation.com>). The SEDFIT parameters were: buffer density 0.9988 g/ml; buffer viscosity 0.01069 poise; the protein partial specific volume was 0.73. The HYDROPRO program (20) was used to calculate the theoretical sedimentation coefficient (S value) from the proposed binding model and from the PDB coordinate files for the DMP12 and *Anabaena* HU.

(A) 10 μM N-terminal His₁₀ tagged DMP12. Only the monomeric form could be found. The S value from the SV data (2.01) is a good match to the theoretical S value of N-terminal His₁₀ tagged DMP12 monomer, which is given by HYDROPRO as 2.0.

(B) 100 μM C-terminal His₆ tagged *Neisseria* HU protein. Only the dimeric form could be found. The S value from the SV data (2.11) is close to the theoretical S value of HU dimer, given by HYDROPRO as 1.9.

(C) 10 μM N-terminal His₁₀ tagged DMP12 and 10 μM C-terminal His₆ tagged *Neisseria* HU protein. The observed S value (3.02) could only have resulted from the DMP12 monomer binding to the HU dimer form (HYDROPRO: 3.19). A full comparison of the theoretical and observed S values is given in Table 2.

Figure S4. The DMP12 structure binds a magnesium ion. The magnesium ion that interacts with the Asp13 of the DMP12 protein may come from the magnesium

acetate in the reservoir. This finding suggests that DMP12 may have a divalent metal ion binding activity. However, we also found that the presence or absence of magnesium did not make very much difference in our subsequent studies on HU-DMP12 interaction. More work will therefore be needed to determine the functional roles of this metal binding activity, if any.

Figure S5. EMSA results from different concentrations of *Neisseria* HU and 2.5 nM plasmid DNA substrate. *Neisseria* HU protein produced band shift of the plasmid DNA in a dose-dependent manner.

Figure S6. DMP12's ability to protect *Neisseria* HU protein from trypsin digestion is dose-dependent. The increasing molar ratios of DMP12 to 100 μ M *Neisseria* HU protein are 1:1, 2:1, 4:1, respectively. His-pulldown was used to purify the un-cleaved C-terminal tagged *Neisseria* HU protein.

Figure S7. The expression of DMP12 increased the growth rate of *E. coli*. (A) The cell density was monitored at OD₆₀₀ after the addition of 1mM IPTG. Data were obtained from three replicate experiments. The different baseline OD₆₀₀ values at 0 hour suggest that *E.coli* transformed with the DMP12 plasmid grows more slowly

than *E.coli* transformed with the empty pET21 plasmid. The reason for this is unknown. (B) Recombinant DMP12 expression in the soluble fraction extracted from the *E.coli* cells. The temperature used in this assay was 37 °C.

Figure S8. Structural comparison of HI1450 and DMP12 (A), and multiple sequence alignment of HI1450 (B) and DMP12 (C) homologs in different species.

Supporting figures

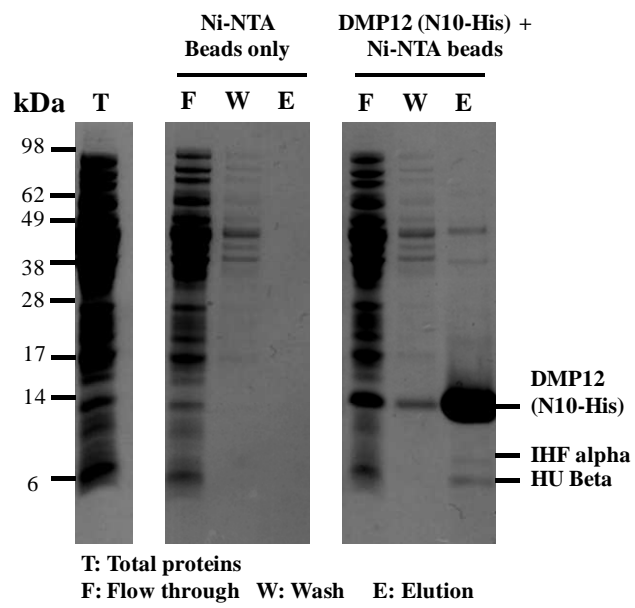


Figure S1

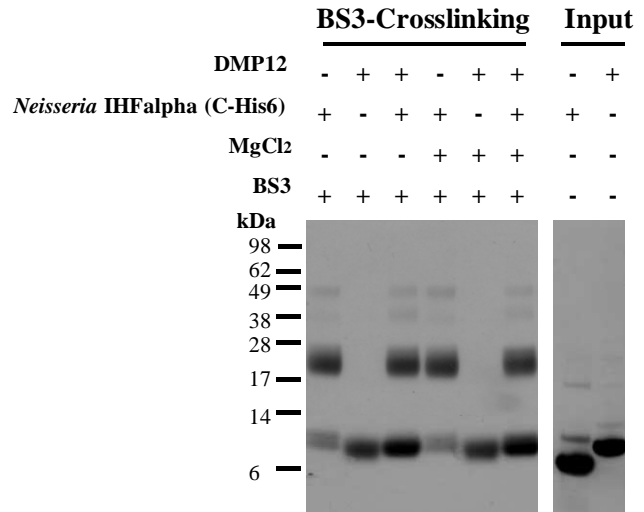
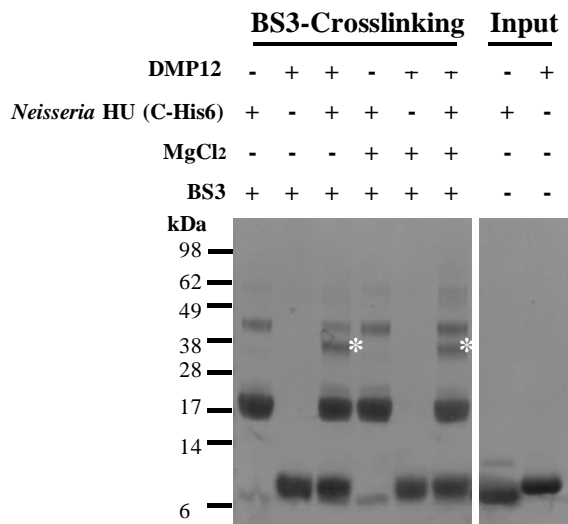


Figure S2

Figure S3 (A)

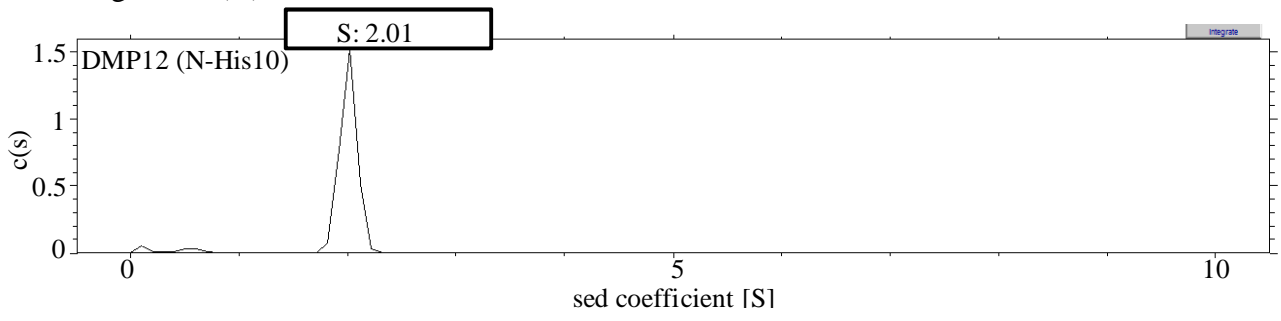


Figure S3 (B)

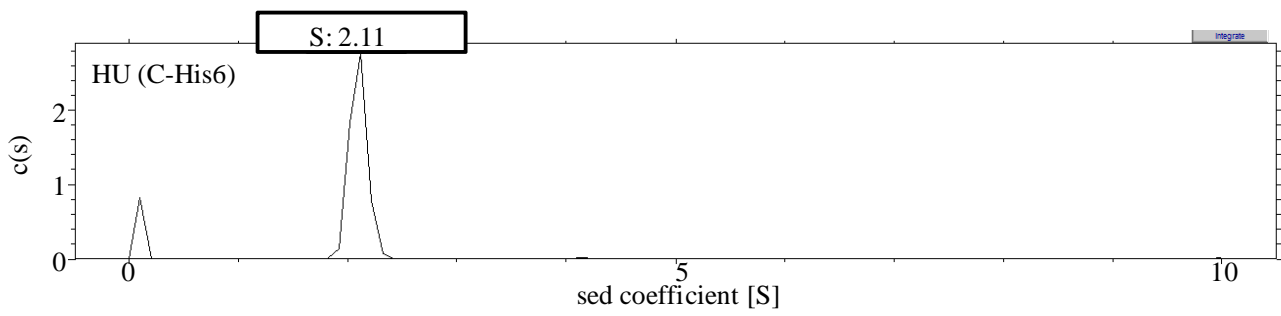


Figure S3 (C)

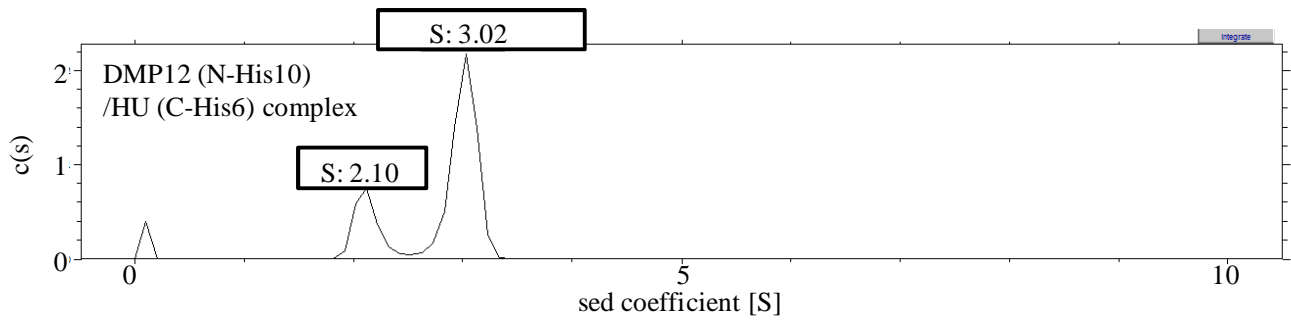


Figure S3

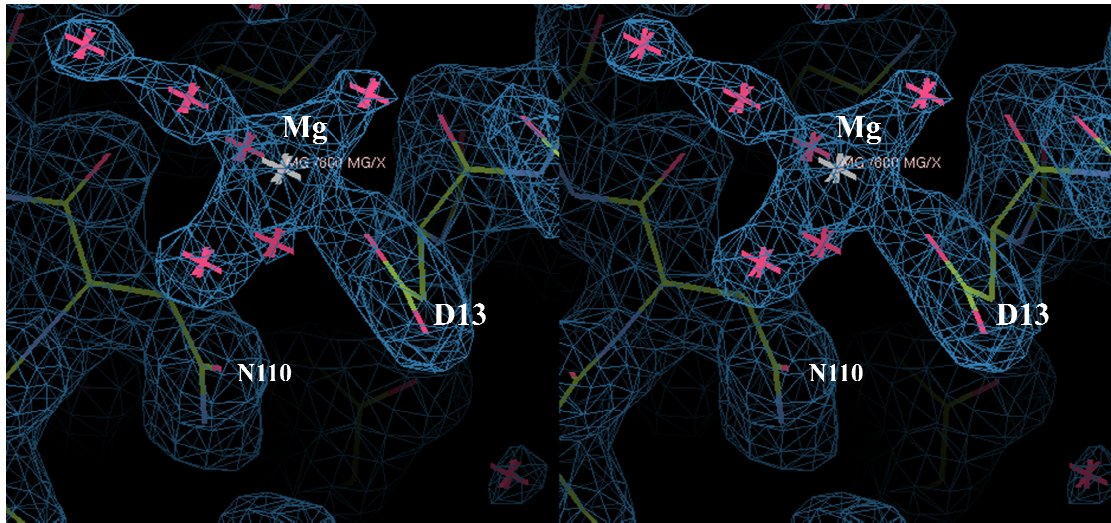
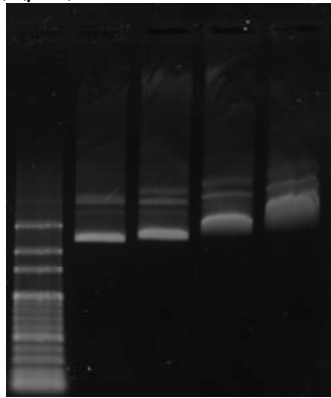


Figure S4

Neisseria HU (C-His6) (μM) 0 1 2 4



DNA substrate concentration: 2.5 nM

Figure S5

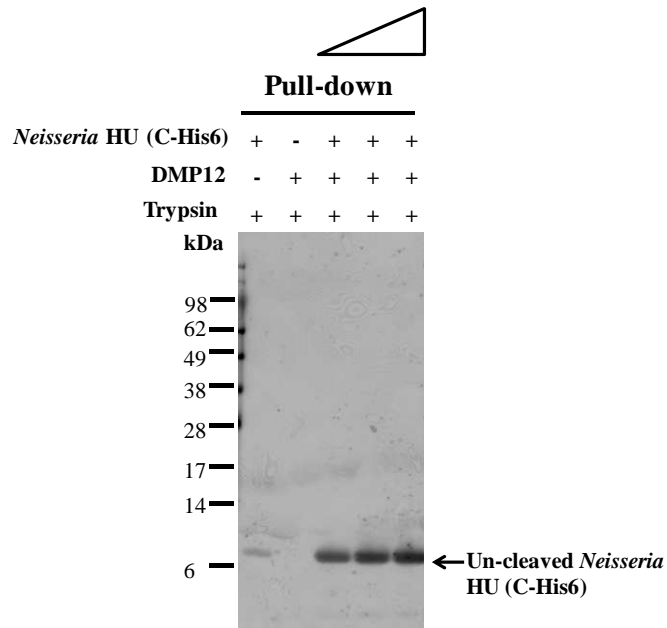


Figure S6

Figure S7 (A)

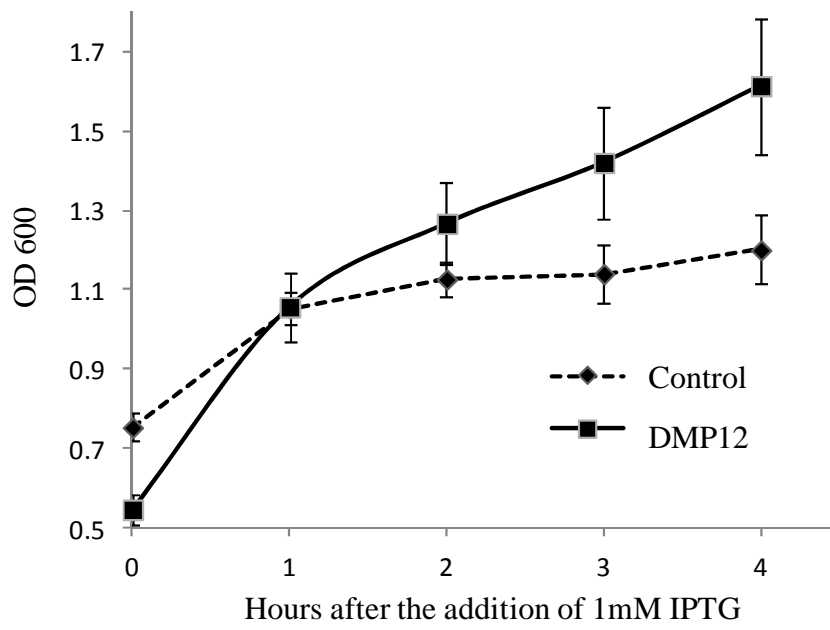


Figure S7 (B)

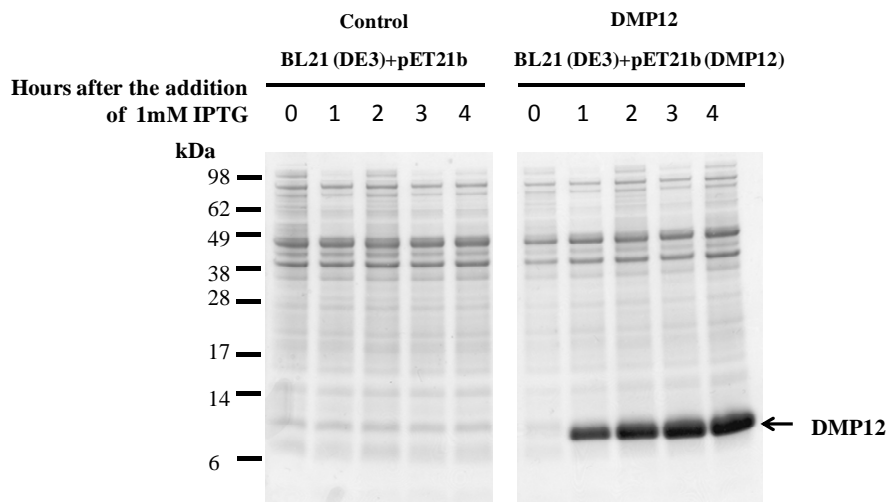


Figure S7

Figure S8 (A)

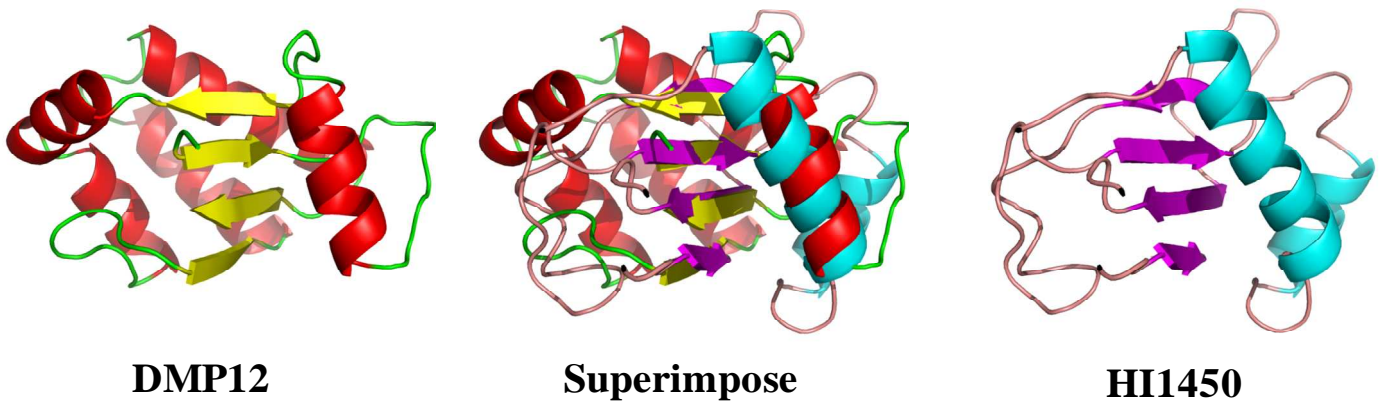


Figure S8 (B)

		10	20	30	40	50
HI1450 (<i>Haemophilus influenzae</i>)	1	-MTTEIKKLD	PDTAIDIA	YDIFLEM	AGENLDP	ADILLFNLQFEE
HI1450 homolog (<i>Pasteurella multocida</i>)	1	--MTEITKLD	PDVAIDL	AYDIFLE	MAPHLDP	ADILLFNLQFEE
HI1450 homolog (<i>Vibrio splendidus</i>)	1	-MTEANDL	MSYDDA	IDTAYD	I FLEM	APDNLEPAD
HI1450 homolog (<i>Vibrio cholerae</i>)	1	----MAEL	ISIDDT	IDTAYD	I FLEM	APDNLEPAD
HI1450 homolog (<i>Escherichia coli</i>)	1	MDMDL	NHRLTE	DETELE	QAYDIF	LELAADN
Consensus		:	*:::	*:::	*:::	*:::
		60	70	80	90	100
HI1450 (<i>Haemophilus influenzae</i>)	50	VETADDW	EEI	IGVLI	DPEEY	AEVWV
HI1450 homolog (<i>Pasteurella multocida</i>)	49	VETADDW	EEM	IGVLI	DPEEY	AEVWV
HI1450 homolog (<i>Vibrio splendidus</i>)	50	VETGDDW	VEH	VGF	VDKEI	IYAEV
HI1450 homolog (<i>Vibrio cholerae</i>)	47	VDVGDDW	DDQ	VGF	VDKEI	IYAEV
HI1450 homolog (<i>Escherichia coli</i>)	51	FEP	AEDWQ	EHVD	FDLNP	DFFAE
Consensus	32	:	:::	:::	:::	:::
		110				
HI1450 (<i>Haemophilus influenzae</i>)	99	REFHVI	WKK	-		
HI1450 homolog (<i>Pasteurella multocida</i>)	98	REFHVV	WKK	-		
HI1450 homolog (<i>Vibrio splendidus</i>)	100	KFCH	ILW	KRD		
HI1450 homolog (<i>Vibrio cholerae</i>)	97	KFCH	MLW	KRD		
HI1450 homolog (<i>Escherichia coli</i>)	101	KLCH	I	WRE	-	
Consensus	61	:	:::	:::		

Figure S8 (C)

		10	20	30	40	50
DMP12 (<i>Neisseria meningitidis</i> MC58)	1	-MNEH	NLLIF	CLKDN	VSI	SEYTEM
conserved hypothetical protein (<i>Neisseria gonorrhoeae</i> PID332)	1	-MNEH	NLLIF	CLKDN	VSI	SEYTEM
hypothetical protein NgonD_00327 (<i>Neisseria gonorrhoeae</i> DG118)	1	-MNEH	NLLIF	CLKDN	VSI	SEYTEM
hypothetical protein (<i>Neisseria lactamica</i> 020-06)	1	-MNEH	NLLIF	CLKDN	VSI	SEYTEM
hypothetical protein E9Q_09985 (<i>Moraxella catarrhalis</i> BCI)	1	MSN	TRLYP	VFCLE	RNI	EINDL
Consensus		*	:	:::	:::	:::
		60	70	80	90	100
DMP12 (<i>Neisseria meningitidis</i> MC58)	50	RGLW	G	LVSEI	T	DNWLF
conserved hypothetical protein (<i>Neisseria gonorrhoeae</i> PID332)	50	RGLW	G	LVSEI	T	DNWLF
hypothetical protein NgonD_00327 (<i>Neisseria gonorrhoeae</i> DG118)	50	RGLW	G	LVSEI	T	DNWLF
hypothetical protein (<i>Neisseria lactamica</i> 020-06)	50	CGLW	G	LVSEI	T	DNWLF
hypothetical protein E9Q_09985 (<i>Moraxella catarrhalis</i> BCI)	51	TGLW	G	I	SEET	DNWLF
Consensus		*	:::	:::	:::	:::
		110				
DMP12 (<i>Neisseria meningitidis</i> MC58)	100	I	I	HVLE	YAI	KNEK
conserved hypothetical protein (<i>Neisseria gonorrhoeae</i> PID332)	100	I	I	HVLE	YAI	KNEK
hypothetical protein NgonD_00327 (<i>Neisseria gonorrhoeae</i> DG118)	100	I	I	HVLE	YAI	KNEK
hypothetical protein (<i>Neisseria lactamica</i> 020-06)	100	I	I	HVLE	YAI	KNEK
hypothetical protein E9Q_09985 (<i>Moraxella catarrhalis</i> BCI)	101	I	L	F	L	DYAI
Consensus	63	*	:::	:::	:::	:::

Figure S8