"**Expression and Characterization of the Histidine-rich Protein, Hpn: Potential for Nickel Storage in *Helicobacter pylori***"

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Supplementary Material

Figure S1 Analysis of purified recombinant Hpn protein by non-denaturing (native) polyacrylamide gel electrophoresis.

Figure S2 Titration curve for the reaction of Ni^{2+} (as $NiSO_4$) with apo-Hpn. Increase in intensity at 286 nm is plotted against the ratio [Ni]/[Hpn] (*r*).

Figure S3. Confirmation of deletion of the hpn gene in H. pylori 26695 by colony PCR, using primers specific for the *hpn* gene and upstream region on chromosome.

Table S1 Comparison of Hpn biophysical properties with those of other histidine-rich proteins.

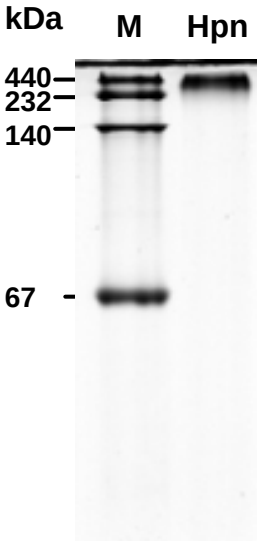

Figure S1 Non-denaturing (native) 10% polyacrylamide gel of purified recombinant Hpn protein, indicating the molecular size of the multimeric Hpn protein complex. M: native molecular weight protein markers; Hpn: Hpn protein.

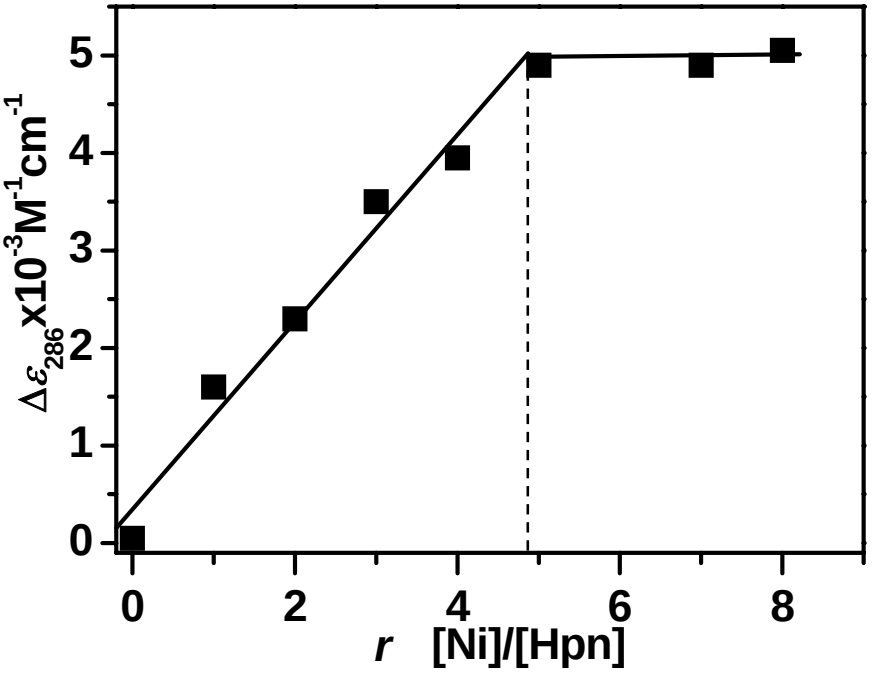

Figure S2. Titration curve for the reaction of Ni^{2+} (as $NiSO_4$) with apo-Hpn. Increase in intensity at 286 nm is plotted against the ratio [Ni]/[Hpn] (*r*).

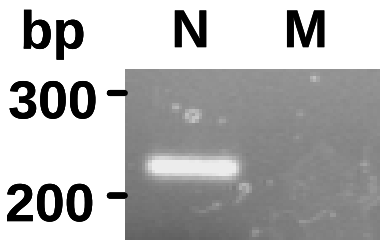

Figure S3. Confirmation of deletion of the hpn gene in *H. pylori* 26695. Colony PCR was performed on individual wild-type and *hpn*-deletion mutant clones. N: wild-type *H. pylori* 26695, M: *hpn*-deletion mutant.

Table S1 Comparison of Hpn with other histidine-rich proteins[a]

Protein	Source	Predicted molecular mass	Apparent molecular mass[b]	Percentage of His in protein	Ref.
		kDa	*kDa*	*%*	
CooJ	*R. rubrum*	12.5	32.6, 39.0, 54.3	16.5	(32)
EP	*M. edulis*	28	52.6	11.1	(42)
HRC	*Sarcoplasmic Reticulum*	165	~1200	12.7	(43)
IGPD	*Filobasidiella neoformans*	22	549[c]	5.9	(37)
UreE	*K. aerogenes*	17	35	8.9	(44)
UreE	*P. miribilis*	17	36	7.5	(45)
HypB	*B. japonicum*	32	78	7.9	(46)
Hpn	*H. pylori*	7	>500, 55, 34, 26, 20, 14, 7	47	This work

[a] All of the histidine rich proteins listed here are putatively multimeric.

[b] As determined by gel filtration chromatography, unless otherwise specified.

[c] Determined with analytical ultracentrifugation in the presence of one molar equivalent Mn^{2+}.