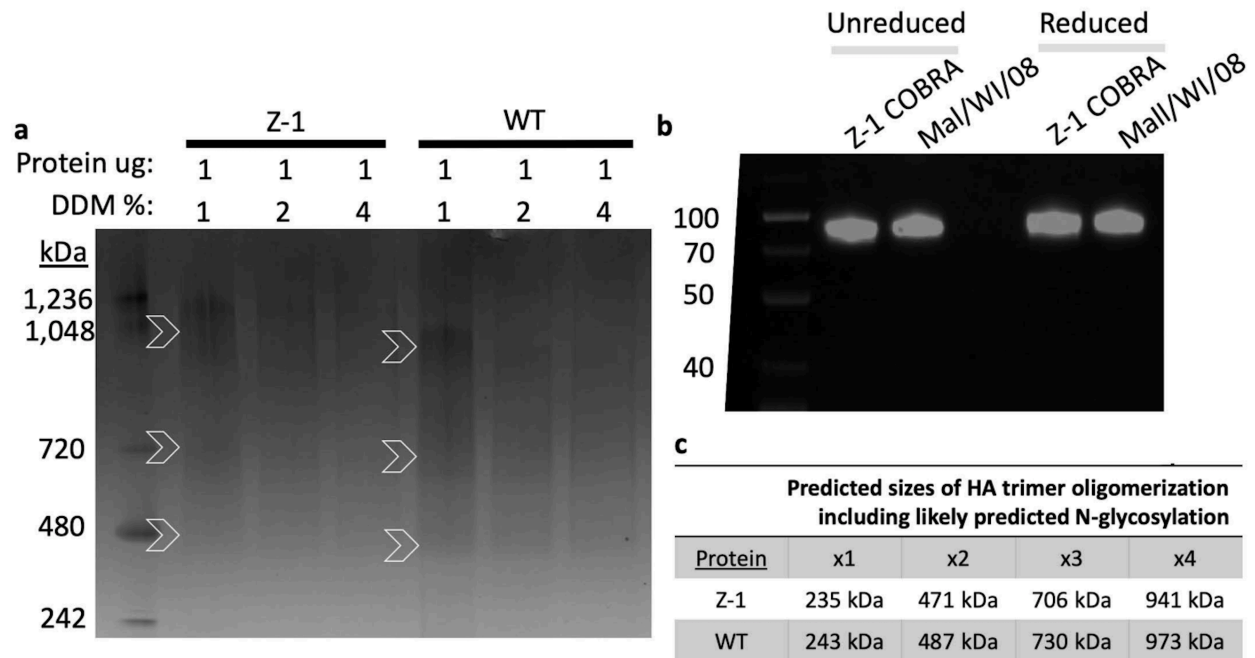


## Supplemental Figures



**Figure S1: Soluble recombinant COBRA and wildtype H2 protein maintains trimerized conformation and form higher order oligomers.** (a) BN-PAGE analysis of Z-1 COBRA NA (Z-1) and A(H2N2)/Mallard/Wisconsin/2008 (WT) soluble protein. Protein conglomerates are indicated with an arrow. Increasing detergent concentration (DDM = dodecyl maltoside) does not resolve the higher-order oligomerization. (b) Anti-His western blot analysis of BN-PAGE. Protein observed at the higher order kDa are confirmed to be H2 recombinant protein by the presence of polyhistidine tag and a dose dependent decrease in loaded protein. (c) Observed estimated protein sizes correspond with predicted sizes of an HA trimer forming higher order oligomers.

Site A										
Strain	115	118	119	120	121	132	133	136	137	139
Z1	K	P	K	D	R	R	A	V	S	N
Z3	K	P	K	D	R	R	A	V	S	N
Z5	K	P	K	D	R	R	A	V	S	N
Z7	K	P	R	D	Q	R	A	V	S	N
Mall/Neth/01	K	P	K	D	R	R	A	V	S	N
Chk/Pots/84	K	P	K	D	R	Q	G	V	F	N
Chk/PA/04	K	A	R	N	R	Q	A	I	Y	G
J/57	K	P	K	D	R	R	A	V	S	N
T/64	K	P	K	D	R	M	A	V	S	K
Sw/MO/06	K	P	R	D	Q	R	A	V	S	N
Mall/WI/08	K	P	R	D	Q	R	A	V	S	N
Mall/MN/08	R	P	R	D	Q	R	A	V	S	N

Site B										
Strain	151	154	158	160	183	184	188	192	195	
Z1	K	S	V	K	E	A	T	N	T	
Z3	K	S	I	K	E	A	T	N	T	
Z5	K	S	V	K	E	T	T	N	T	
Z7	K	S	I	K	D	A	T	N	T	
Mall/Neth/01	K	S	V	K	E	A	A	N	T	
Chk/Pots/84	K	S	V	K	E	T	T	N	A	
Chk/PA/04	K	S	V	R	E	N	A	N	T	
J/57	K	S	V	K	E	T	T	N	T	
T/64	E	T	V	K	E	A	A	K	T	
Sw/MO/06	K	S	I	K	D	A	T	N	T	
Mall/WI/08	K	S	I	K	D	A	T	N	T	
Mall/MN/08	K	S	I	K	D	A	T	N	T	

Site C						
Strain	43	45	269	276	293	308
Z1	K	N	T	K	I	E
Z3	K	N	T	K	I	E
Z5	K	N	T	K	I	E
Z7	R	S	T	K	I	E
Mall/Neth/01	K	N	T	K	I	E
Chk/Pots/84	K	N	T	K	I	E
Chk/PA/04	K	N	T	K	I	E
J/57	K	N	T	K	V	E
T/64	K	N	T	K	V	E
Sw/MO/06	R	S	I	K	I	D
Mall/WI/08	R	S	T	R	I	D
Mall/MN/08	K	S	T	R	I	D

Site E					
Strain	72	80	86	87	88
Z1	S	M	R	N	G
Z3	S	M	R	N	G
Z5	S	M	R	D	G
Z7	S	V	V	N	G
Mall/Neth/01	T	M	R	N	G
Chk/Pots/84	T	M	R	N	G
Chk/PA/04	T	I	R	N	G
J/57	S	M	R	D	G
T/64	R	M	R	V	S
Sw/MO/06	S	V	V	N	G
Mall/WI/08	S	V	V	N	G
Mall/MN/08	S	V	V	N	G

Site F		
Strain	30	31
Z1	K	D
Z3	Q	D
Z5	K	D
Z7	K	D
Mall/Neth/01	Q	D
Chk/Pots/84	Q	D
Chk/PA/04	Q	D
J/57	K	D
T/64	K	D
Sw/MO/06	K	N
Mall/WI/08	K	D
Mall/MN/08	K	D

Site D																	
Strain	88	114	171	177	195	203	209	214	217	221	223	234	235	237	247		
Z1	G	V	M	V	T	T	I	T	K	Q	G	G	D	M	W	T	I
Z3	G	V	M	V	T	T	I	T	K	Q	G	G	D	I	W	T	I
Z5	G	V	M	V	T	T	T	T	K	Q	G	G	D	M	W	T	I
Z7	G	V	M	I	T	T	I	T	K	Q	G	G	E	T	W	V	I
Mall/Neth/01	S	V	M	V	T	T	V	T	K	Q	G	G	D	M	L	T	I
Chk/Pots/84	G	V	M	V	A	T	L	T	E	Q	G	G	D	I	W	T	I
Chk/PA/04	G	V	M	I	T	K	V	T	K	Q	G	G	D	M	L	T	I
J/57	G	V	M	V	T	T	T	T	K	L	G	G	D	M	W	T	I
T/64	S	V	M	V	T	T	I	A	K	L	G	S	D	M	W	T	V
Sw/MO/06	G	V	M	I	T	T	I	T	K	L	G	G	E	T	W	V	I
Mall/WI/08	G	V	M	V	T	T	T	T	K	Q	G	G	E	T	W	V	I
Mall/MN/08	G	I	I	V	T	T	I	T	R	Q	N	G	E	T	W	V	I

**Figure S2: Amino acid diversity in antigenic sites of WT and COBRA H2 HA sequences.**

The amino acid differences for the WT and COBRA H2 HA sequences in the six H2 HA antigenic sites are shown in the six tables. Amino acids are numbered based upon H3 numbering system. Only amino acid positions with differences are shown. All of the other amino acids in the antigenic sites are the same for all of the H2 HA sequences used in this study.