

Supplementary Fig. 1 Comparisons of human H7N9 mAbs *in vitro*. **a** ELISA binding curves of the indicated mAbs to H7N9 HA1 from A/Shanghai/2/2013, A/Guangdong/17SF003/2016, and A/Hong Kong/125/2017. **b** ELISA binding curves of the indicated mAbs to H7N9 HA based on A/Shanghai/2/2013 and H7N7 HA based on A/Netherlands/219/2003. **c** Neutralization curves of the indicated mAbs against the 2013 and 2017 H7N9 pseudo viruses in MDCK cells. Data shown are mean \pm SEM, with source data provided in a Source Data file. Similar results have been independently reproduced at least once.

Heavy Chain V-gene

	-----FR1-----	CDR H1	-----FR2-----	CDR H2	-----FR3-----				
	10	20	30	40	50	60	70	80	90
IGHV4-59	QVQLQESGPGLVKPS	ETLSLTCTVSGGSIS	SYYSWIRQPPGKGLEW	IGIYYSGSTN	YNPSLKS	RVTISVDT	SKNQFSLKLS	SVTAADTAVYYC	
H7.HK1	QVQLQESGPGLVKPS	ETLSLTCSVSGGSIN	SY ^Y WTWIRQPPGKLEW	VGYI ^Y HS ^G ST ^S	YNPSLKS	RITISVAP	SKNH ^F SL ^E L ^T	SMTAADTAVYYCAR	
H7.HK2	QVQLQ ^G SGPGL ^L LRP	SETLSLTCSVSGVSI ^N	SY ^Y WSW ^V RQPPGKALEW	IGIYY ^G SN ^T	YNPSLE	SRVTISVDR	SKNQFSLK ^M T	SVTAADTARYFCAR	
IGHV7-4-1	QVQLVQSGSELKPKG	ASVKVSCASGYTFT	SYAMN ^V VRQAPGQGLEW	MGWINTNTGNPTYAQ	GFTGRFV	FLDTSVSTAYL	QICSLKAEDTAVYYC		
H7.HK3	QVQLVQSGSELK ^R PGASV	KVSC ^R ASGYTFT	SY ^T IN ^V WVRQAPGQGLEW	MGWINT ^S TG	DPTYAQ	GFTGRFV	FLDTSVSTAYL ^E I ^S R	LKAEDTAVYYCAR	
IGHV4-61	QVQLQESGPGLVKPS	ETLSLTCTVSGGSVSSGS	YYSWIRQPPGKGLEW	IGIYYSGSTN	YNPSLKS	RVTISVDT	SKNQFSLKLS	SVTAADTAVYYC	
H7.HK4	QVQLQESGPGLVKPS	ETLSLTCTVSGGSV ^R SA ^S Y	AW ^S WIRQPPGKLEW	IGD ^I YYSG ^T TN	YNPSLKS	RVTL ^S VD ^T A ^K N ^R	^R F ^S L ^R L ^R	SVTAADTAVY ^H CAR	

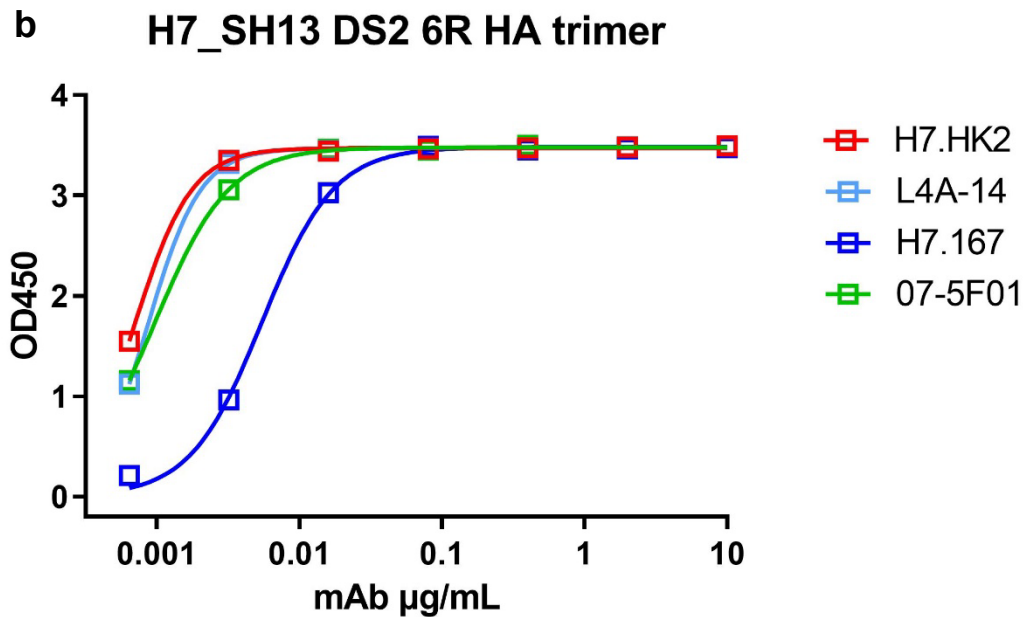
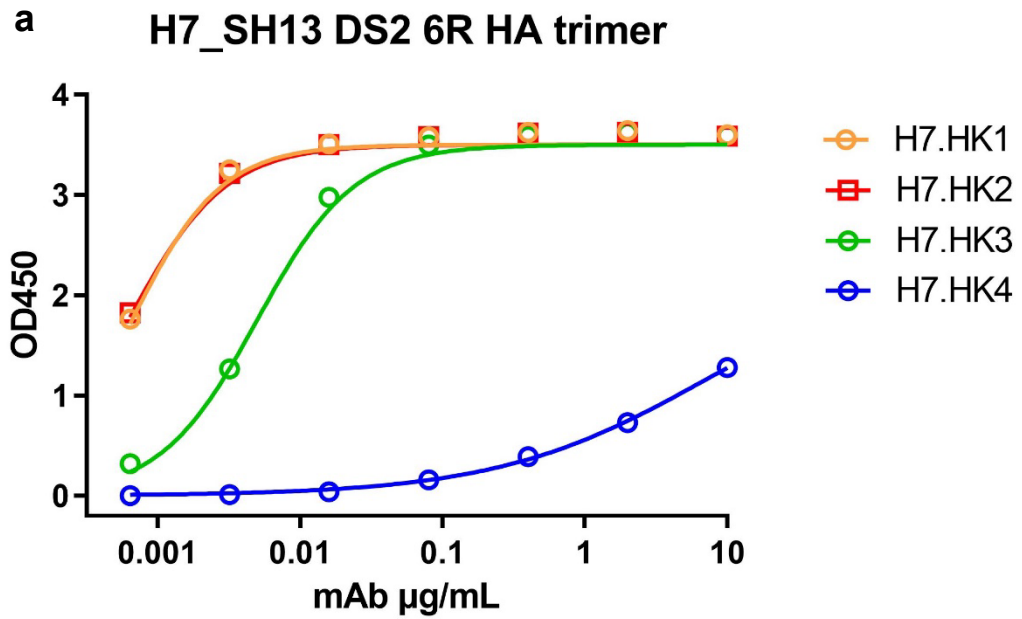
Light Chain V-gene

	-----FR1-----	CDR L1	-----FR2-----	CDR L2	-----FR3-----				
	10	20	30	40	50	60	70	80	88
IGKV2-28	DIVMTQSP ^L SLPVT	PGEPASISCRSSQ	SLLSHNGYNYLDWY	LQKPGQSPQLLI	YLGSNRASGV	PDFRFS	SGSGSGTDF	TLKISRVEAEDVGVYYC	
H7.HK1	DIVMTQSP ^V SLPVT	PGEPASIS ^C NSSQ	SLLSH ^N GYA ^L	LDWY ^L QKPGQSP ^K LM ^I YL	GLNRA ^F	GVPDFRFS	SGSGSGTDF	TLKISRVEAEDVGVYYC	
H7.HK2	DIVMTQSP ^L SLPVT	PGEPASISCRS ^N QSL	QHSN ^G YV ^L	LDWY ^R QKPGQSP ^H LLI	YLG ^F NRA ^S	GVPDFRFS	QHSN ^G YV ^L	TLKISRVEAEDVGVYYC	
IGKV1-5	DIQMTQSPSTLSASV	GDRVITICRASQSI	SSWLA	WYQQKPGKAPKLLI	YDASSLES	GVPSRFS	SGSGSGTEFTLT	TISSLQPDFATYYC	
H7.HK3	DIQMTQSPSTLSASV	GDRVITICRASQSI	SSWLA	WYQQKPGKAPKLLI	YKASSLES	GVPSRFS	SGSGSGTEFTLT	TISSLQPDFATYYC	
IGKV1-16	DIQMTQSPSSLSASV	GDRVITICRASQGI	SNYLA	WFQQKPGKAPKSLI	YAASSLQ	SGVPSRFS	SGSGSGTDF	TLTISSLQPDFATYYC	
H7.HK4	DIQMTQSPSSLSASV	GDRVITICRASQGI	RNYLA	WFQQKPGQ ^A PKSLI ^F	FAASSL ^H T	GVPSRFS	SGSGSGTDF	TLTISSLQPDFATYYC	

CDR3

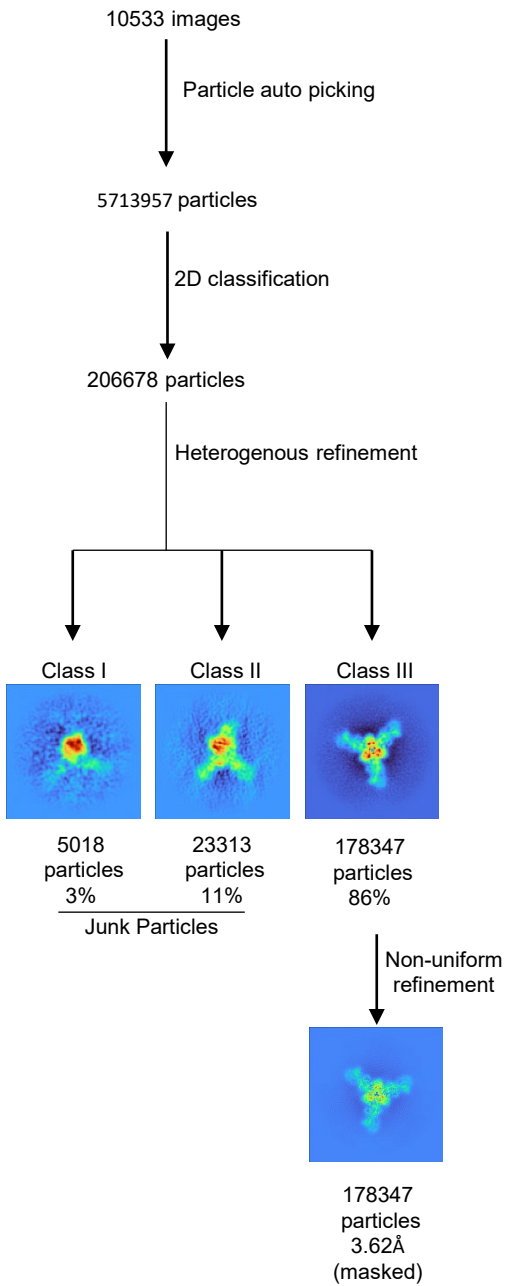
	CDR H3	----FR4----	CDR L3	----FR4----
	95 100ABC	103 110	90 98 106	
H7.HK1	LG ^G H ^G D ^Y G ^S D ^Y	WGQGT ^L V ^T V ^S S	MQALQ ^T P ^F T ^F G ^P G ^T R ^V DIK	
H7.HK2	QGI ^F G ^D Y ^G S ^D Y	WGPG ^T L ^V T ^V S ^S	MQGLQ ^T P ^F T ^F G ^P G ^T T ^V DLK	
H7.HK3	AFGLT ^V VRG ^G IVG ^V WGQGT ^T V ^T V ^S S		QQYNSYSQ ^T FGQ ^G T ^K VEIK	
H7.HK4	ERYYYGSSGDFD ^Y	WGQGT ^L V ^T V ^S S	QHYN ^S Y ^P PT ^F GQ ^G T ^K LEIK	

Supplementary Fig. 2 H7.HK mAb sequences. Protein sequences of the heavy and light chain variable regions of H7.HK mAbs are aligned to the putative germline V-genes at top, with amino acid substitutions in red, and in magenta for substitutions shared between the clonally related mAbs H7.HK1 and H7.HK2. Spaces are added to maintain alignment; framework regions (FR) and complementarity-determining regions (CDRs) are indicated based on the Kabat numbering and nomenclature. Highlighted in yellow are the mAb residues (paratopes of H7.HK1 and H7.HK2) contacting the H7 antigen. The putative N-linked glycosylation sites on the light chain CDR L1 of H7.HK1 and H7.HK2 and the heavy chain CDR H2 of H7.HK3 are underlined.

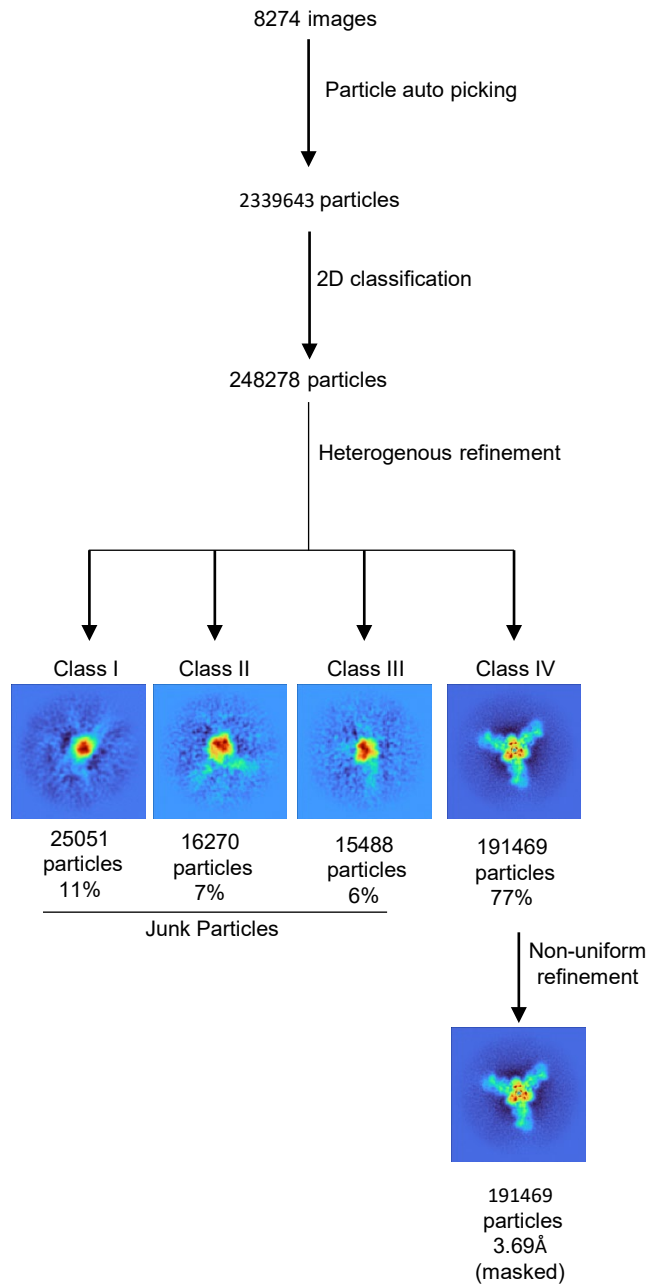


Supplementary Fig. 3 Human H7N9 mAbs binding to a soluble, disulfide-stabilized, fully cleaved H7 HA trimer H7 SH13 DS2 6R. **a** ELISA binding curves of the H7.HK mAbs to H7 SH13 DS2 6R HA trimer. **b** ELISA binding curves of previously reported mAbs to H7 SH13 DS2 6R HA trimer; the H7.HK2 mAb was included for comparison. Source data are provided in the Source Data file. Similar results have been independently reproduced at least once.

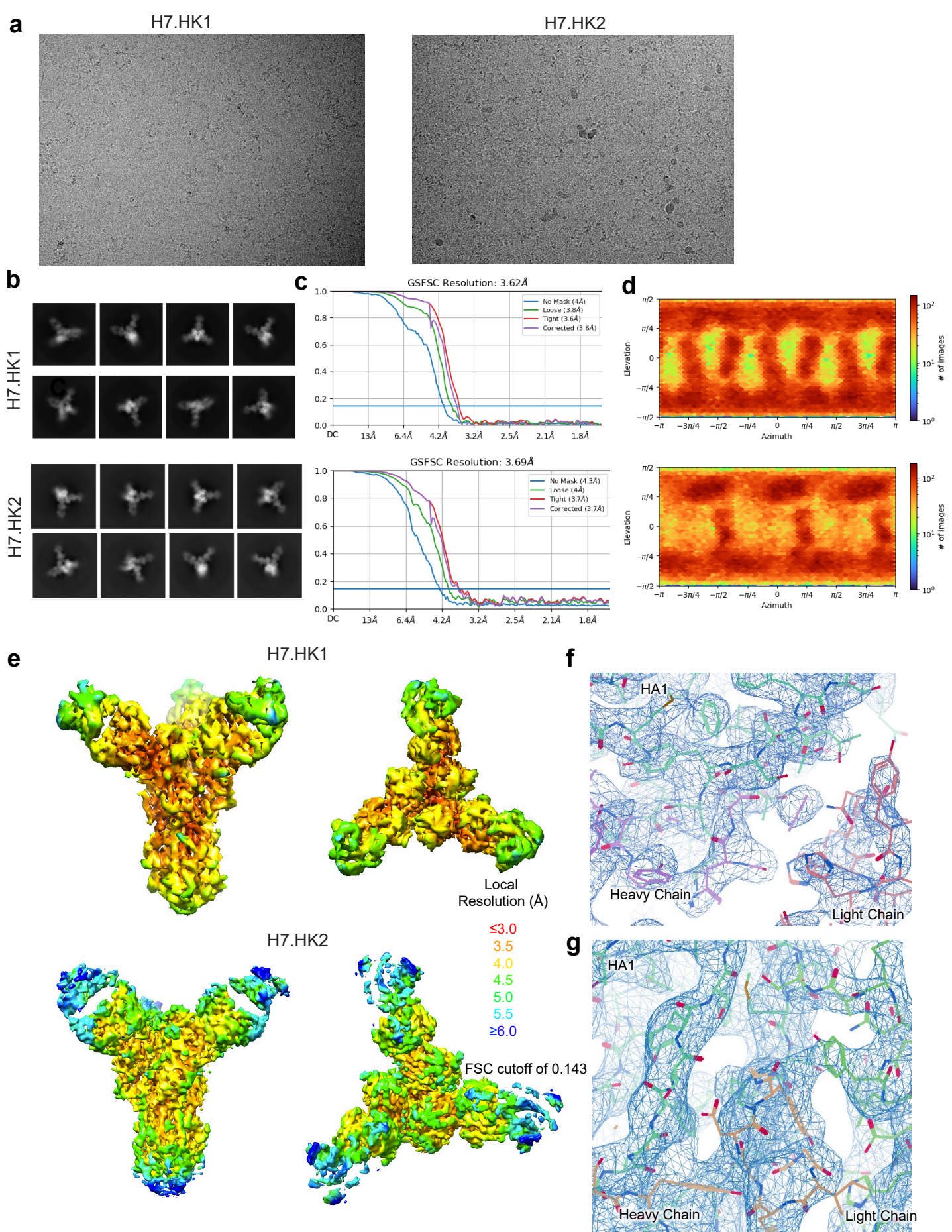
H7.HK1 Data Processing



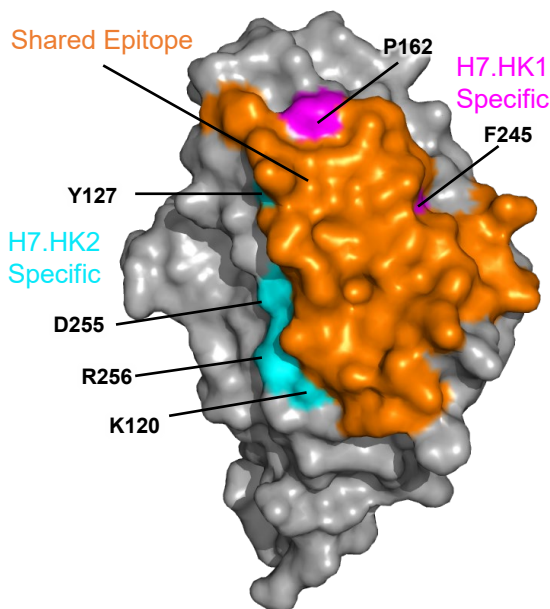
H7.HK2 Data Processing



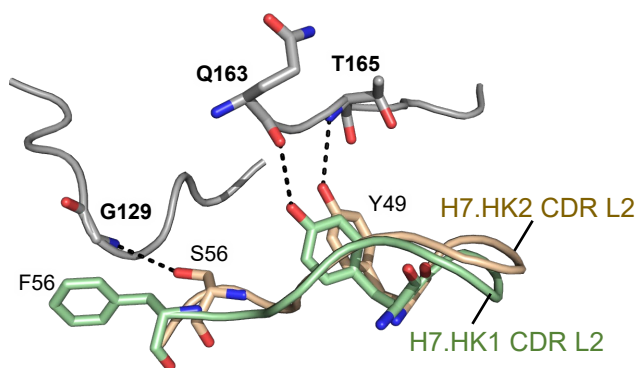
Supplementary Fig. 4 Cryo-EM data processing for H7.HK1 and H7.HK2 in complex with H7 SH13 DS2 6R HA trimer.



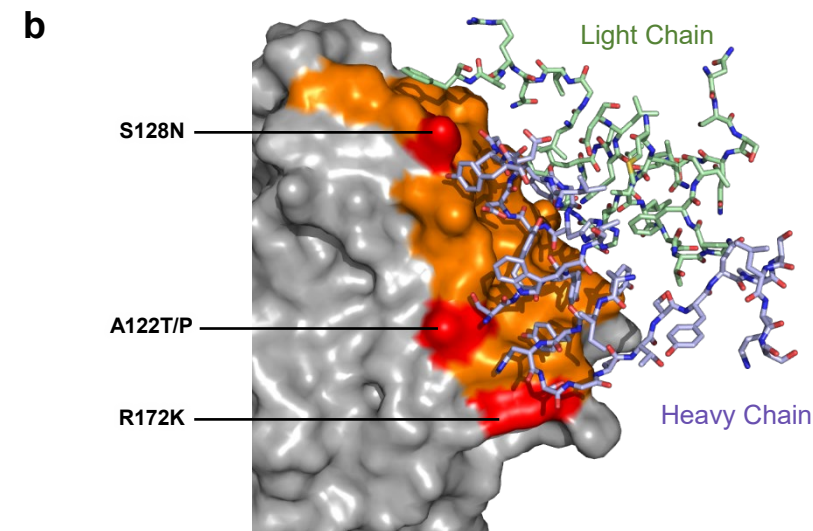
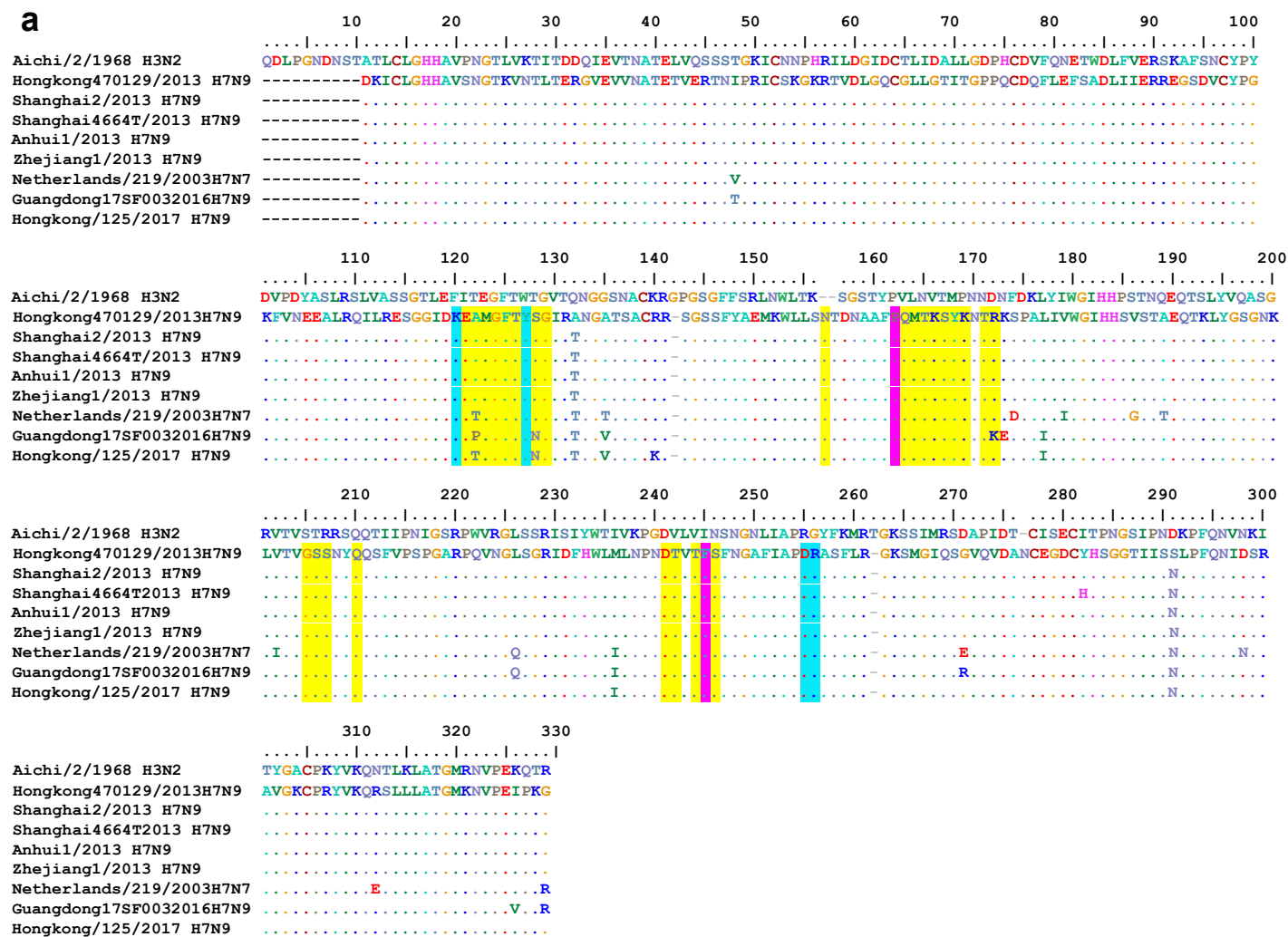
Supplementary Fig. 5 Cryo-EM details of H7.HK1 and H7.HK2 in complex with H7 SH13 DS2 6R HA trimer. **a** Representative micrograph of H7.HK1 and H7.HK2. **b** Representative 2D class averages of H7.HK1 and H7.HK2. **c** The gold-standard Fourier Shell Correlation (FSC) resulted in a resolution of 3.62 Å for the overall map of H7.HK1 and 3.69 Å for the overall map of H7.HK2. Non-uniform refinement with C3 symmetry was used for both reconstructions. **d** The orientations of all particles used in the final refinement are shown as a heatmap. **e** The local resolution of the final overall map is shown contoured at 0.0989 for both structures. Resolution estimation was generated through cryoSPARC using an FSC cutoff of 0.143. **f** Representative density is shown for the interface of H7.HK1 heavy chain, light chain, and H7 HA. **g** Representative density is shown for the interface of H7.HK2 heavy chain, light chain, and H7 HA.

a**b**

	<u>H7.HK1</u> : <u>H7</u>	<u>H7.HK2</u> : <u>H7</u>
Heavy Chain		Y52 : Y168
H Bonds	Y52 : E121	Y53 : E121
	R94 : G124	R94 : G124
	G99 : S167	G99 : S167
	D100 : T126	D100 : T126
	Y100a : T165	Y100a : T165
	Y100a : S167	Y100a : S167
	S100c : T126	S100c : T126
Heavy Chain Salt Bridge	H53 : E121	
Light Chain		
H Bonds	Y49 : Q163	Y49 : T165
		S56 : G129

c

Supplementary Fig. 6 Comparison of H7.HK1 and H7.HK2 binding epitopes to H7. **a** Differences in the epitopes of H7.HK1 and H7.HK2. Majority of surface contacts are conserved, shown in orange. H7.HK1 specific surfaces are shown in magenta, and H7.HK2 specific surfaces are shown in cyan. **b** Hydrogen bonds and salt bridges formed by H7.HK1 and H7.HK2 with H7. **c** Differences in CDR L2 binding to H7 by H7.HK1 and H7.HK2 as a result of F56S substitution in H7.HK2. S56 forms an additional hydrogen bond with G129 of H7. Additionally, position of Y49 is shifted so that it forms a hydrogen bond with T165 for H7.HK2 instead of Q163 for H7.HK1.



Supplementary Fig. 7 Antigenic drift of H7 HA1 in 2016-2017. **a** The A/Aichi/2/1968 H3N2 HA1 protein sequence is shown at top to indicate the H3 numbering of HA1. The H7 HA1 sequences from the indicated viral isolates are aligned to the 2013 Hong Kong H7N9 autologous isolate, with identical amino acids shown in dots. "-" depicts gap. Highlighted in yellow are the H7 contact residues (epitope) with both mAbs H7.HK1 and H7.HK2. H7.HK1 specific epitopes are in magenta; H7.HK2 specific epitopes are in cyan. **b** Surface presentation of the H7 HA1 domain highlighting the epitopes (orange) of mAbs H7.HK1 and H7.HK2, with three mutations in red that appeared in the 2016-2017 viral isolates of H7N9. The sticks are interacting CDRs of mAb H7.HK1 heavy and light chains.

Supplementary Table 1 Cryo-EM data collection, refinement, and validation statistics for H7 SH13 DS2 6R HA in complex with H7.HK1 and H7.HK2 Fabs.

	H7 SH13 DS2 6R H7.HK1 (EMD-41422) (PDB: 8TNL)	H7 SH13 DS2 6R H7.HK2 (EMD-41441) (PDB: 8TOA)
Data collection and processing		
Magnification	105000	105000
Voltage (kV)	300	300
e-/Å ²	58	58
Defocus range (µm)	0.8-2	0.8-2
Pixel size (Å)	0.83	0.83
Symmetry imposed	C3	C3
Initial particle images (no.)	5713957	2339643
Final particle images (no.)	178347	191469
Map resolution (Å)	3.62	3.69
FSC threshold	0.143	0.143
Refinement		
Initial model used (PDB code)	6IDD	8TNL
Model resolution (Å)	3.62	3.69
FSC threshold	0.143	0.143
Model composition		
Non-hydrogen atoms	16487	15570
Protein residues	2112	2109
Ligands	7	11
<i>B</i> factors (Å ²)		
Protein	39	58
Glycans	58	48
R.m.s. deviations		
Bond lengths (Å)	0.005	0.007
Bond angles (°)	1.121	1.231
Validation		
MolProbity score	1.65	2.23
Clashscore	5.45	12.08
Poor rotamers (%)	0.06	1.62
Ramachandran plot		
Favored (%)	94.86	92.30
Allowed (%)	5.14	7.41
Disallowed (%)	0.0	0.29