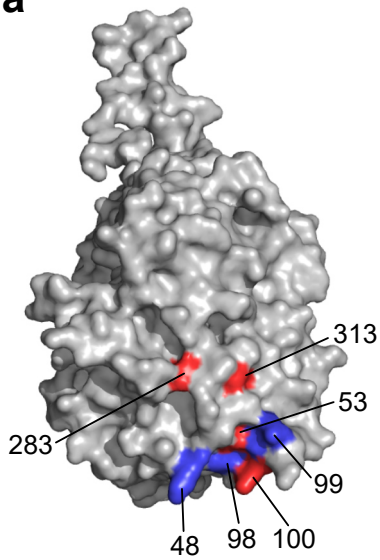
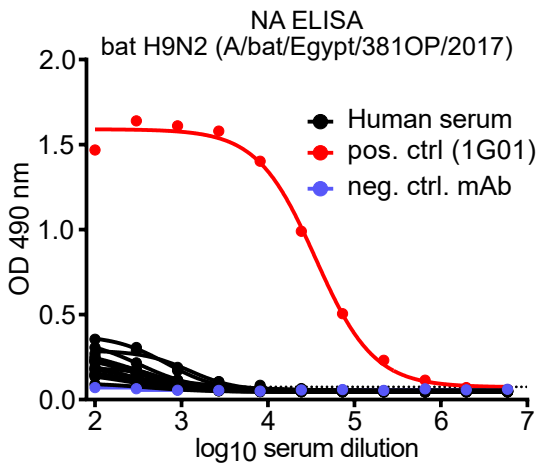
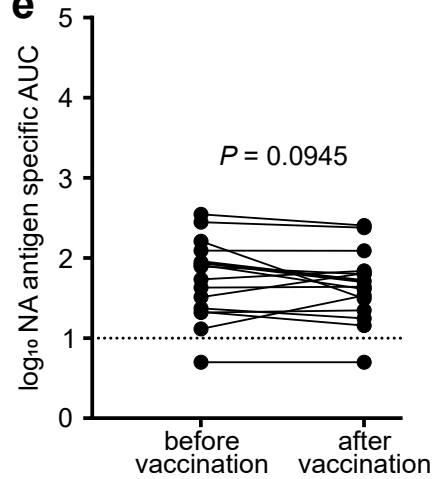
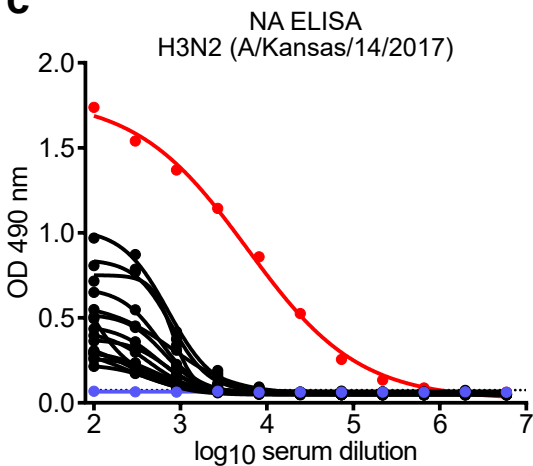
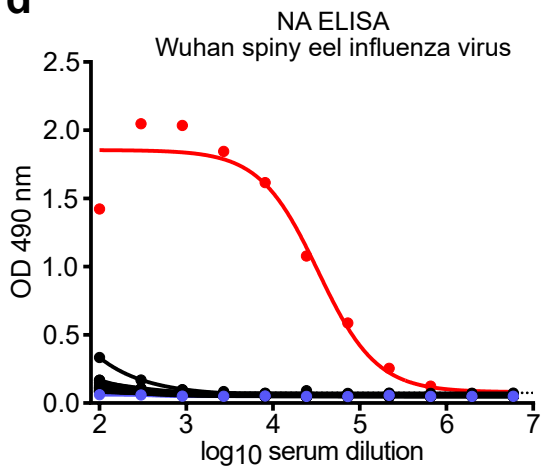


**Supplementary Fig. 1: Bat H9N2 replicates efficiently in turkey but not chicken hatchlings.** **a** Turkey hatchlings (n= 13) were oro-nasally inoculated with bat H9N2 and viral shedding was monitored by RT-qPCR analysis of oro-pharyngeal swab samples. Asterisk indicates successful virus isolation from oral swab material from one turkey tested at 5 dpi. Dashed line indicates detection limit. **b** Serum antibody titers of turkey hatchlings before and after bat H9N2 infection were determined by an IAV NP-specific ELISA. Dashed lines indicates threshold between 45% and 50% inhibition. Mean antibody titers are indicated. **c** Neutralizing antibody titers against bat H9N2. **d** A group of chicken hatchlings (n= 13) was oro-nasally inoculated with bat H9N2 and viral shedding was monitored by RT-qPCR analysis of oro-pharyngeal swab samples. Dashed line indicates detection limit. **e** Serum antibody titers of chicken hatchlings at 21 dpi with bat H9N2 were determined by an IAV NP-specific ELISA. Dashed lines indicate threshold area between 45% and 50% inhibition. Mean antibody titers are indicated. **f** 15 ferrets were directly inoculated with bat H9N2. At 1 dpi, three naïve contact ferrets were housed together with all donor animals in interconnected cage units to allow direct exposure of all ferrets and to study bat H9N2 transmission. **g** Nasal swabs containing shed infectious bat H9N2 virus taken from bat H9N2-infected (n= 15) and contact (n= 3) ferrets at the indicated time points were titrated by endpoint titration on MDCK cells. Dashed line indicates the detection limit. Data are mean  $\pm$  SD.

**a**

48	K	K	Q	K	K
53	E	D	E	E	E
98	R	R	K	R	R
99	R	R	K	R	R
100	I/V	I/V	R	R	R
283	P	L	L	L	L
313	Y	V	F	F	F
	1918-NP	pdmH1N1-NP	Belzig-NP	KAN-1-NP	bat H9N2-NP

**b****e****c****d**

**Supplementary Fig. 2: No cross-reactive antibodies to bat N2 among individuals vaccinated against seasonal influenza.** **a** Known MxA escape mutations in NP from the 1918 and the 2009 pdmH1N1 strains are highlighted in red and the resistance patch of the Eurasian avian-like swine isolate Belzig is shown in blue. Note that the avian-adapted IAV KAN-1 and bat H9N2 do not harbor any of the known MxA-resistance amino acid residues. The NP model was created with PyMol based on the available crystal structure (PDB code: 2Q06). **b** Reactivity of sera from 15 healthy adults taken before the 2022/23 seasonal influenza vaccination to recombinant N2 from bat H9N2. **c,d** Reactivity of the same sera against the recombinant N2 of a recent seasonal H3N2 strain (**c**) and the recombinant NA of the Wuhan spiny eel influenza virus (**d**). **e** Pre- and post- seasonal vaccination reactivity of sera from 15 healthy adults who received the 2022/23 seasonal influenza virus vaccine against recombinant N2 from the bat H9N2 virus. Reactivity was quantified as area under the curve (AUC). A paired *t*-test was used to determine statistical differences. For **b**, **c** and **d**, mAb 1G01 was used as positive control, an irrelevant human mAbs was used as negative control.