Sequential Infection of Poultry with Low and then High Pathogenicity H7N7 Avian Influenza Viruses: Investigating Factors Contributing to Disease Outcome

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Introduction

Factors driving the evolution of high pathogenicity avian influenza viruses (HPAIVs) from low pathogenicity viruses (LPAIVs) remain undefined. In July 2015, a disease investigation prompted by increased mortality on a UK commercial layer chicken farm, identified an incursion of H7N7 LPAIV that circulated within birds on the premises shortly before mutating to a HPAIV. HPAIV induced mortality in a subset of chickens, but the majority were protected, potentially due to prior exposure to LPAIV. Whilst genetic evidence for both H7N7 LPAIV and HPAIV were detected during the outbreak, only the HPAIV (A/chicken/England/26352/2015, H7N7-HP) was successfully isolated [1].

Aims

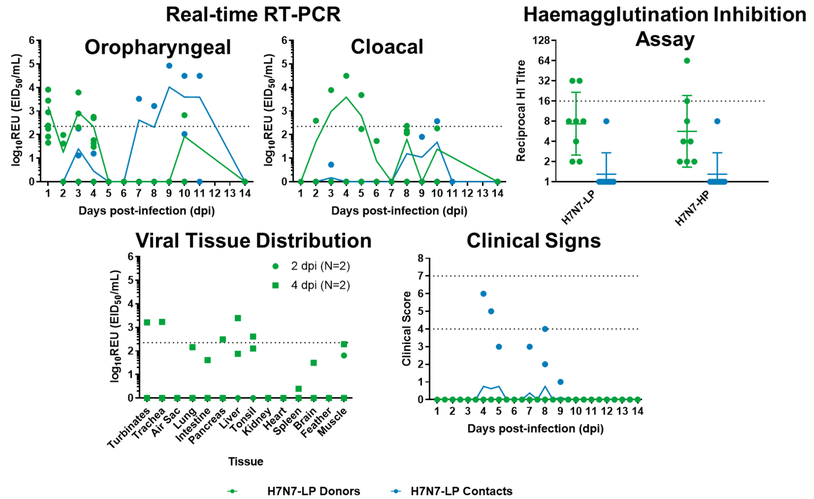
To model the sequential events that occurred during the 2015 H7N7 HPAIV chicken outbreak in the UK and thereby gain a further understanding of the effects of prior LPAIV infection upon a latter homosubtypic HPAIV infection.

Results

1. A/mallard/Netherlands/19/2015 (H7N7-LP) is an appropriate H7N7 LPAIV surrogate in the absence of viable UK LPAIV from the outbreak as shown by high sequence identity to H7N7-HP

Percentage Amino Acid Identity to H7N7-HP (%)										
Influenza A Protein	PB2	PB1	PA	HA	NP	NA	M1	M2	NS1	NS2
H7N7-LP	98.8	99.2	98.2	99.8	99.2	99.2	99.2	99.0	99.1	97.5

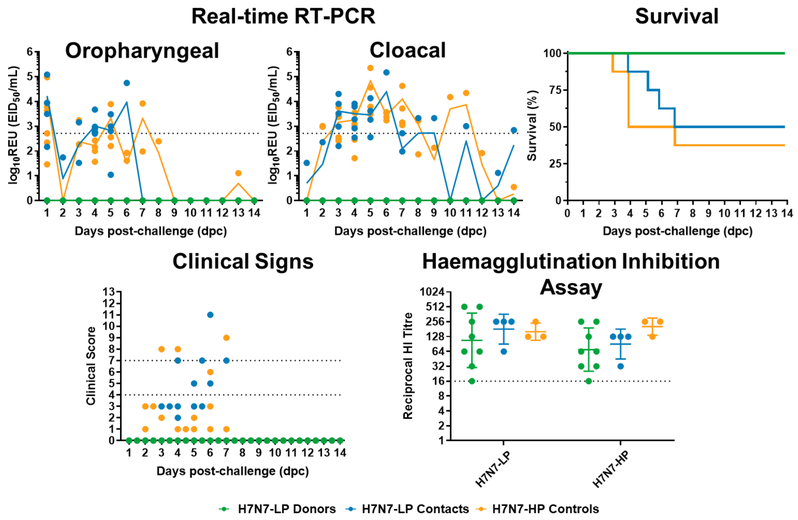
2. H7N7-LP infects chickens and transmits to naïve contacts



Eight donor White Leghorn chickens (H7N7-LP Donors) were infected with 10^6 EID_{50} H7N7-LP at three weeks-of-age and then co-housed with eight naïve contact chickens (H7N7-LP Contacts). from 1 dpi.

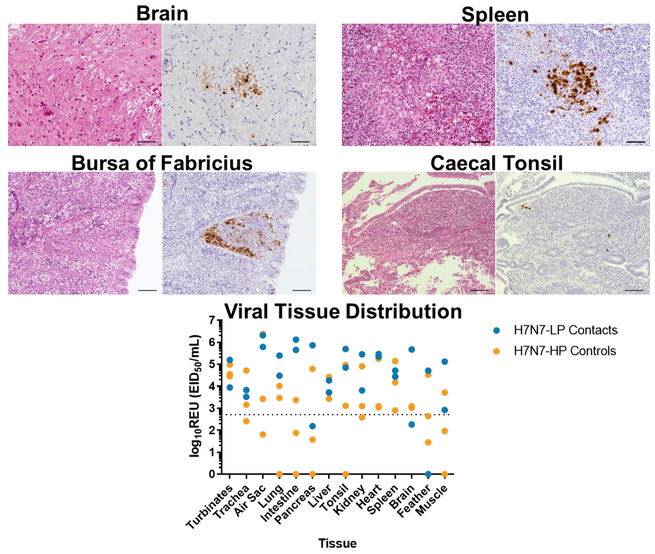
- All H7N7-LP Donors shed viral RNA [2,3] and seroconverted at 10 dpi, whilst only 50% of the H7N7-LP Contacts shed viral RNA and 37.5% seroconverted after H7N7-LP infection.
- Additional H7N7-LP Donors euthanised at 4 dpi showed viral RNA was restricted to the tissues of the respiratory tract.
- Clinical signs including huddling, ruffled feathers, conjunctivitis and periocular oedema were only observed in the H7N7-LP Contacts.

3. H7N7-HP causes high mortality in naïve controls and H7N7-LP Contacts, but does not infect H7N7-LP Donors



At 14 dpi after H7N7-LP infection, all 16 H7N7-LP Donors and Contacts, along with an additional eight naïve chickens (H7N7-HP Controls), were challenged with 10^6 EID_{50} H7N7-HP.

- All H7N7-HP Controls, 87.5% of H7N7-LP Contacts and none of the H7N7-LP Donors shed viral RNA after challenge with H7N7-HP.
- 62.5% of the H7N7-HP Controls and 50% of the H7N7-LP Contacts succumbed to infection with clinical signs including: huddling, ruffled feathers, cyanosis of the combs or wattles, loss of balance, closed eyes and dropped wings.
- All surviving chickens seroconverted to both the H7N7-LP and H7N7-HP viruses at 14 dpc.



- Immunohistochemical [4] and real-time RT-PCR analysis of tissues collected during the H7N7-HP challenge demonstrated systemic viral distribution as well as necrosis in the brain, spleen, bursa of Fabricius and caecal tonsil of the H7N7-HP Controls and H7N7-LP Contacts.

Conclusions

- A novel experimental approach was employed to investigate the putative sequence of events that occurred during the UK H7N7 HPAIV outbreak in 2015.
- H7N7-LP was able to infect and transmit, albeit at low levels between naïve chickens.
- H7N7-HP caused high mortality in naïve chickens, but prior H7N7-LP infection afforded protection against H7N7-HP.
- This study provides further insight into the field observations from outbreaks where there is initial LPAIV incursion, followed by mutation to the corresponding HPAIV and emphasises the importance of surveillance for notifiable LPAIVs.

References

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Acknowledgements

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