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Forum

# Highly Pathogenic Avian H7N9 Influenza Viruses: Recent Challenges

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**Novel highly pathogenic avian influenza (HPAI) H7N9 viruses of the fifth epidemic wave infect humans and poultry. Recently, HPAI H7N9 viruses have evolved into different subtypes and genotypes, exhibited heightened virulence in mammals, and extended their host range, thereby posing a potential threat to public health and the poultry industry.**

## The Emergence of HPAI H7N9 Infecting Poultry and Humans

Initially, the avian influenza A(H7N9) virus that emerged in early 2013 in China had low pathogenicity to chickens but could replicate and spread efficiently among them. Previous studies have shown that low pathogenic avian influenza (LPAI) viruses can become highly pathogenic via the insertion of multiple basic amino acids at the cleavage site of the hemagglutinin (HA) protein, which was the case for the H5 and H7 subtype avian influenza viruses (AIVs). In July 2016, a novel HPAI H7N9 variant possessing multiple basic amino acids at the cleavage site of the HA protein emerged [1] (Figure 1) and had caused 32 human infections by November 2018[i] as well as 27 outbreaks in poultry farms in 13 provinces of China[ii], raising serious concerns about its epidemic and pandemic potential.

## Evolution and Diversity of HPAI H7N9 Viruses

Recent studies by Shi *et al.* and Qi *et al.* [1–3] unveiled the evolution of HPAI H7N9 viruses. They found that the novel HPAI H7N9 variants from both poultry and humans clustered together and formed a distinct cluster in both the hemagglutinin and neuraminidase phylogenetic trees, indicating a single origin. Of note, the hemagglutinin mutants may have arisen from two different H7N9 viruses, one of which subsequently reassorted with others to form two more genotypes [3]. A more recent analysis of the HPAI H7N9 viruses showed that the viruses evolved rapidly and had reassorted with other AIV subtypes within a few months after its emergence in early 2017, forming nine H7 genotypes, including a new 'G9' genotype – a novel HPAI H7N2 strain that was generated by the reassortment between the HPAI H7N9 viruses and other unknown duck viruses in unvaccinated domestic ducks [2] (Figure 1). Additionally, HPAI H7N9 and H7N2 viruses detected in provinces other than Guangdong are descendants of the earlier Guangdong HPAI H7N9 viruses, rather than mutated derivatives of the local LPAI H7N9 viruses. The internal genes of the novel HPAI H7N9 variants, however, are scattered across the phylogenetic tree and formed several clusters but are closely related to the LPAI H7N9/H9N2 strains circulating locally among humans and chickens in the past waves. These observations suggest that these HPAI H7N9 variants have undergone complex reassortment with LPAI H7N9/H9N2 strains, and that other subtypes of AIVs are cocirculating in poultry and forming multiple genotypes.

## Replication, Pathogenicity, and Transmissibility of HPAI H7N9 Viruses in Mammals

To date, although no sustained transmission of HPAI H7N9 viruses among humans has been documented, further adaptation of these AIVs to humans may result in transmissible viruses with pandemic potential. Therefore, timely and comprehensive evaluation of the replication, pathogenicity, and transmissibility of HPAI H7N9 viruses in mammals is critical to the understanding of their evolutionary characteristics and determinants for the emergence of potential pandemic strains. Recent studies have shown that the HPAI H7N9 viruses isolated from humans could have different consequences for mammals compared to those isolated from poultry [1,3,4] (Figure 1). In the mouse model, human strains (A/Guangdong/17SF003/2016-like virus) were lethal and more virulent than LPAI H7N9 viruses [1,4]. In ferrets, A/Guangdong/17SF003/2016-like viruses not only killed the animals but also transmitted through respiratory droplets with comparable efficiency to LPAI H7N9 viruses [4]. Replication of the viruses in extrapulmonary tissues, including in brains, was detected in mice and ferrets. In contrast, a chicken strain (A/chicken/Guangdong/SD008/2017) was not lethal to mice or ferrets nor transmitted through ferrets [3]. Therefore, human-isolated HPAI H7N9 viruses were more pathogenic in mice and more transmissible in ferrets than those isolated from poultry. However, when bearing the mammalian-adapted 627K or 701N mutation in its polymerase basic 2 (PB2) protein, the A/chicken/Guangdong/SD008/2017 virus became highly lethal in mice and efficiently transmissible in ferrets [3], which may be due to the avian-isolated HPAI H7N9 with such mutations were easily recovered [1,3]. In the recent study of Shi *et al.* [2], all 18 tested HPAI H7 subtype strains of avian-isolated viruses (including the

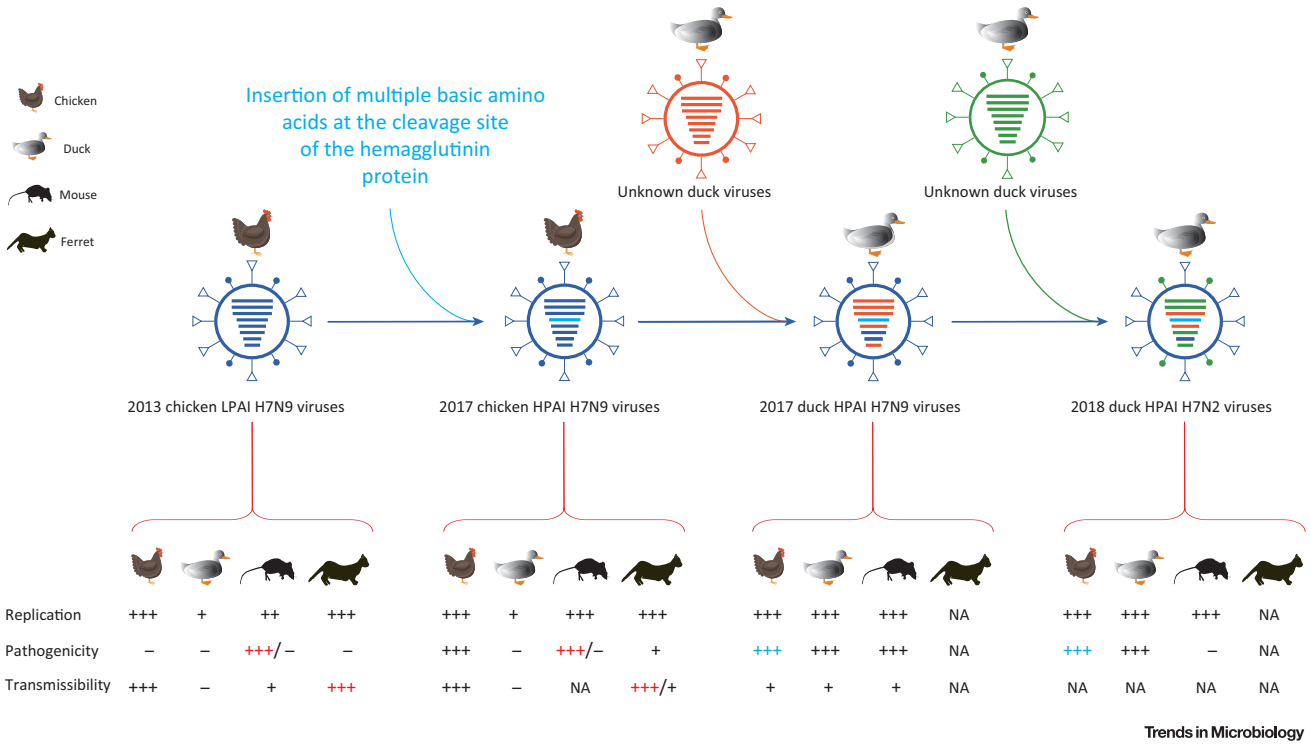


Figure 1. Brief Outline of the Evolution, Replication, Pathogenicity, and Transmissibility of Highly Pathogenic Avian Influenza (HPAI) H7N9 Virus in China. LPAI, low pathogenic avian influenza virus. '+++' (red, strains isolated from human cases or strains isolated from poultry bearing the 627K or 701N mutation in its polymerase basic 2 protein; light blue, presumed pathogenicity but unknown due to the limited data available at present) indicates efficient ability of replication, pathogenicity, or transmissibility of the viruses in animals; '++' indicates moderate; '+' indicates low; and '–' indicates that the viruses have no such capability. NA indicates no available data.

novel H7N2 virus) with different motifs in their HA cleavage sites could replicate in the turbinates and lungs of the mice, although only about one quarter of the strains were detected in the brains or the spleens of the mice with low viral titers. There was a wide range of fatality rates in the mice, from 0% to 100%. Of more concern is that an evolved HPAI H7N9 strain (A/Chicken/Hunan/S1220/2017) was more than 1000-fold more lethal in mice than the initial HPAI strain (A/Chicken/Guanxi/SD098/2017), although these two viruses belong to the same genotype and have the same motif in their HA cleavage sites. It is clear now that some HPAI H7N9 viruses have become virulent in mice after circulating in poultry for a few months after emergence. However, the underlying mechanism of the increased virulence of these viruses remains to be explored in further studies.

## Host Range Extension of H7N9 HPAI Viruses

The LPAI H7N9 viruses isolated in 2013 were mainly transmitted in chickens and were not able to replicate efficiently in ducks [5,6] (Figure 1). Similarly, the emerged HPAI H7N9 viruses also spread quickly in chickens, despite the high fatality rate [1,3]. Analogous to the low replicability of LPAI H7N9 viruses in ducks, a pertinent question is whether these HPAI H7N9 viruses can replicate efficiently in ducks. In their assessment of the replicability of both human- and avian-isolated HPAI H7N9 strains in Peking ducks, Qi *et al.* [1] found that 20–60% of the ducks challenged with either one chicken-isolated virus (A/Chicken/Heyuan/16876/2016) or two A/Guangdong/17SF003/2016-like viruses (A/Guangdong/Th005/2017 and A/Guangdong/Th008/2017) showed viral shedding at the different sampling time points during the 2 weeks following observation and seroconversion at the end, although none showed clinical signs. This observation implies that some HPAI H7N9 strains may have already adapted to ducks by acquiring genes from duck influenza viruses. This extension of host range is further supported by the efficient replication of HPAI H7N9 (A/Duck/Fujian/SD208/2017) and H7N2 (A/Duck/Fujian/SE0195/2018) viruses isolated from ducks, and these two strains were lethal in ducks [2] (Figure 1).

## Concluding Remarks

The rapid evolution, increased virulence, and efficient transmissibility of HPAI H7N9

viruses in mammalian models, together with their extended host range, have heightened their pandemic potential, posing an imminent threat to public health and the poultry industry. Despite the success of the mass immunization of poultry with the H5/H7 bivalent poultry vaccine in stopping the spread of the H7N9 virus among both chicken and humans during 2017 and 2018 [2,7–9], several areas need to be addressed, including: (i) whether the new reassortants of H7N9 viruses that could not be protected against by the H5/H7 bivalent vaccine will emerge sooner or later in unvaccinated naïve birds, a problem seen in historical poultry mass vaccination programs against HPAI H5N1 viruses in China [10]; (ii) whether the emerging HPAI H7N9 and H7N2 viruses will widely transmit in ducks, other poultry species, or wild waterfowl; and (iii) whether the novel HPAI H7N9 and H7N2 viruses will become adapted to human hosts or even sustain human-to-human transmission. Hence, it is time to consider vaccinating ducks in future since the H5/H7 bivalent vaccine provides good protection against different H7 viruses in both chickens and ducks, although high-level vaccination coverage in ducks is challenging to achieve. Meanwhile, continued active surveillance still needs to be strengthened for early warning of the evolution and spread of these viruses. Moreover, any newly detected H7N9 and other H7 subtype viruses should be systematically evaluated for their potential ability to adapt to human hosts, and whether the current H5/H7 vaccine can protect against the new viruses.

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## Resources

[i]www.fao.org/ag/againfo/programmes/en/empres/h7n9/situation_update.html

[ii]www.oie.int/en/animal-health-in-the-world/update-on-avian-influenza

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