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**Evolution and biological characteristics of the circulated H8N4 avian influenza viruses**

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**Abstract:**

**Background/Objective:** The circulating avian influenza viruses in wild birds have a high possibility of spillover into domestic birds or mammals at the wild bird-domestic bird or bird-mammal interface. H8N4 viruses primarily circulate in migratory wild waterfowl and have rarely been identified in domestic birds.

**Methods:** In this study, data statistical analysis, isolation and identification of viruses, genetic evolution analysis, and host adaptability study were used.

**Results:** In this study, we summarized the spatial and temporal distribution of global H8 viruses, specified their natural reservoirs, and performed detailed evolutionary analysis on the dominant H8N4 viruses. Here, we also reported a novel H8N4 virus isolated from a Eurasian coot sample from a wetland in eastern China in 2022. Animal infection studies indicated that the wild bird-originated H8N4 virus can replicate and transmit efficiently in ducks but has not adapted to chickens. Additionally, this naturally isolated H8N4 virus can replicate in mice without prior adaptation.

**Conclusion:** These results indicate that H8 viruses exist mainly in the wild duck reservoir and pose a high infection risk to domestic ducks. Therefore, the active surveillance of influenza viruses at the wild and domestic waterfowl interface will contribute to monitoring the circulation of these viruses.

**Keywords:** avian influenza viruses; H8N4; wild birds