Dear Editor, We appreciate the reviewers suggestion for the improvement of the quality of this manuscript. The manuscript has been revised according to the suggestions and comments of the reviewers. Below is a point by point rebuttal to suggestions.

Review report 1

In this manuscript, Clement Meseko et al. reported the highly pathogenic H5Nx influenza viruses infection in Nigeria from 2021 to 2022. The globally circulating H5 viruses since 2019 have produced a huge devastation to poultry industry. The authors here detected 97 H5 viruses and characterized their genetic evolution. The study will contributed to our understanding the highly pathogenic H5 viruses infection in West Africa, however, I have several concerns that should be addressed by the authors.

1. Line 119, specific primers were used to identify the HA and NA subtype, which should be listed in the text.

The specific primers are previously described and published in :

Spackman, E.; Senne, D.A.; Myers, T.J.; Bulaga, L.L.; Garber, L.P.; Perdue, M.L.; Lohman, K.; Daum, L.T.; Suarez, D.L. Development of a Real-Time Reverse Transcriptase PCR Assay for Type A Influenza Virus and the Avian H5 and H7 Hemagglutinin Subtypes. J. Clin. Microbiol. 2002, 40, 3256–3260, doi:10.1128/JCM.40.9.3256-3260.2002.

Slomka, M.J.; Pavlidis, T.; Banks, J.; Shell, W.; McNally, A.; Essen, S.; Brown, I.H. Validated H5 Eurasian Real-Time Reverse Transcriptase-Polymerase Chain Reaction and Its Application in H5N1 Outbreaks in 2005-2006. Avian Dis. 2007, 51, 373–377, doi:10.1637/7664-060906R1.1.

Monne, I.; Ormelli, S.; Salviato, A.; De Battisti, C.; Bettini, F.; Salomoni, A.; Drago, A.; Zecchin, B.; Capua, I.; Cattoli, G. Development and Validation of a One-Step Real-Time PCR Assay for Simultaneous Detection of Subtype H5, H7, and H9 Avian Influenza Viruses. J. Clin. Microbiol. 2008, 46, 1769–1773, doi:10.1128/JCM.02204-07.

Hoffmann, B.; Hoffmann, D.; Henritzi, D.; Beer, M.; Harder, T.C. Riems Influenza a Typing Array (RITA): An RT-QPCR-Based Low Density Array for Subtyping Avian and Mammalian Influenza a Viruses. Sci. Rep. 2016, 6, doi:10.1038/SREP27211.

Panzarin, V.; Marciano, S.; Fortin, A.; Brian, I.; D'amico, V.; Gobbo, F.; Bonfante, F.; Palumbo, E.; Sakoda, Y.; Le, K.T.; et al. Redesign and Validation of a Real-Time RT-PCR to Improve Surveillance for Avian Influenza Viruses of the H9 Subtype. Viruses 2022, 14, doi:10.3390/V14061263.

These papers were cited in the manuscript.

2. Although, the outbreaks have been indicated by the map, I suggested the authors listed the detailed information of these outbreaks, including birds (chickens or ducks), infection numbers, subtype of the viruses, time and regions in figure 1 or in a new table. By the way, I can not open suppl table1.

This was previously provided in Supplementary. Table S1 and now updated and re-attached in the format that can easily be accessed

3. The detailed information of the 97 should be shown in the text by a table, including virus name, host, collection date, collection sites, subtype, accession numbers. The requested data were reported in Table S1; however, due to a formatting error, they were not available for consultation. The problem has now been solved and all information are now accessible. We apologize for any inconvenience.

4. Were the 97 viruses isolated from the clinical samples? If not, how many viruses were sequenced directly from the samples and how many viruses were sequenced from the isolates? This information has now been included in Table S1.

5. I strongly recommend the authors to update the phylogenetic tree of HA, and added representative viruses that detected from Asia, Europe, America to reveal the whole evolutionary trend of H5 viruses in the world. We addressed this comment accordingly. As this tree is too large to be included in the main text we provided it as supplementary Figure S10.

6. The phylogenetic trees of NA and the six internal genes should be provided in the manuscript. Thank you for pointing this out. Required phylogenetic trees are now provided as supplementary files (S1-S9).

7. The amino acids mutations at position 30 and 215 in M1 protein have be proved to affect the pathogenicity of viruses in mammals, and should be added in the table (PMID: 19117585, PMID: 34908445).

Thank you for pointing this out. The text was modified accordingly and Table 1 updated.

8. In the discussion part, the strategies against the highly pathogenic H5 viruses in Nigeria and China should be discussed. Because China successfully avoid the large scale infection of H5 in this 2019-2023 wave by using the vaccination strategy (PMID: 36458831, PMID: 34757542).

Nigeria also successfully contained H5N1 clade 2.2 in 2006-2008 through the biosecurity approach. Options for AIV vaccination is a Federal government policy and has not yet been adopted in Nigeria. This was mentioned in the manuscript in this context "Given the pattern of introduction and spread of IAV in Nigeria, the importance of pre-emptive bio-surveillance of wild migratory birds at major wetlands and Live Bird Markets, combined with stringent biosecurity measures cannot be overemphasised. Future discussion on additional control measure such as vaccination may be an appropriate tool in some circumstances, but its use may sometimes have negative impacts. Therefore, the government must first carry out risk/benefit assessment of the situation"

Thank you
Clement Meseko
(for authors)