**I prefer:**

□ ORAL presentation

☑ POSTER presentation

**The emergence of a disease caused by a** **mosquito origin Cluster 3.2 Tembusu virus in chickens in China**

Zejun Li1\*, Dawei Yan1, Xuesong Li1,

\*lead presenter

1lizejun@shvri.ac.cn, Shanghai Veterinary Research Institute, Chinese Academy of Agricultural Sciences, P. R. China

**Abstract:**

In 2021, a chicken Tembusu virus (TMUV) caused outbreaks of a disease characterized by retarded growth and egg production decline in chickens in China. Two TMUV strains SD2021 and GX2021 were isolated from the diseased chickens and phylogenetic analysis of the E gene nucleotide sequence revealed that the chicken TMUV SD2021 and GX2021 were most close to mosquito origin TMUV in Cluster 3.2, which was distinct from the prevalent duck TMUVs in Cluster 2. The TMUV SD2021 caused growth retardation and neurological symptoms in chickens through both intranasal and intramuscular infection routes, but has no direct-contact transmissibility among chickens. The findings of this study highlight the emergence of a disease caused by a chicken adapted TMUV in China.

**Background/Objective:**

Tembusu virus (TMUV) belongs to the Ntaya virus group in the genus Flavivirus of family Flaviviridae. Since 2010, duck TMUVs (DTMUVs) caused outbreaks mainly in ducks characterized by severe drop in egg production of laying ducks, encephalitis and growth-retardation of young ducks, and spread widely in China, Malaysia and Thailand. In 2021, an infectious disease, characterized by retarded growth and egg production decline, outbroken in chickens, in Shandong and Guangxi, China.

**Methods:**

The pathogen was determined as TMUV by RT-PCR detection. In the present study, two TMUV SD2021 and GX2021 strains were isolated from the diseased chickens, and the phylogenetic based on E genes was analyzed. To evaluate the pathogenesis and transmission of the TMUV in chickens, six 3-week-old chickens were infected with 103.5 TCID50 of SD2021 through i.n. or i.m. route, respectively. Tissues were titrated and sera antibody were tested by using a blocking ELISA described previously. To evaluate the efficacy of licensed DTMUV vaccine FX2010-180P against the chicken TMUV SD2021, three chickens were vaccinated i.m. with 104.0 TCID50 of the vaccine. The antibody titers (percent of inhibition value) of immunized chickens were tested every one week post immunization. The vaccinated chickens were challenged with the TMUV SD2021 at 3 wpi, three chickens in each group were euthanized at 4 days post challenge. Tissue samples were collected and titrated.

**Results:**

In the present study, the TMUV SD2021 and GX2021 isolated from diseased chickens were most closely related to mosquito origin TMUV, but not the duck origin TMUV, which suggested that the novel TMUV SD2021 and GX2021 were not evolved from the prevalent DTMUVs. The chicken TMUV SD2021 and GX2021 caused retarded growth in the chicken embryos. TMUV SD2021 infected and caused viremia in chickens, and some infected chickens showed neurological symptoms. And the SD2021 replicated in multiple organs of inoculated chickens by either i.n. or i.m. inoculation. Whereas, the contact-transmission studies revealed the TMUV SD2021 was not transmitted among chickens by direct-contact, and the virus was not detected in lungs of infected chickens, which were quite different with that of Cluster 2 DTMUV that has contact- and aerosol-transmissibility in ducks. All the results indicated that the SD2021 infected chicken by both i.n. and i.m. inoculation, but has no contact-transmissibility among chickens, which suggested that the chicken TMUV might be spread by mosquito-bird-mosquito cycles instead of direct-contact transmission. The licensed live-attenuated TMUV FX2010-180P vaccine is able to provide an effective protection against the novel chicken TMUV in chickens.

**Conclusion:**

This study demonstrated that the novel chicken TMUV SD2021 and GX2021 of subCluster 3.2 has been adapted to chickens to some extent, but has no direct-contact transmissibility in chickens.Our findings highlighted the emergence of the novel TMUV infection in chickens in China.

**Keywords:** Tembusu virus, mosquito-origin, chicken, pathogenicity, transmission