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Assessment of pathogenicity and tissue distribution of infectious bronchitis virus strains (Italy 02 genotype) isolated from Moroccan broiler chickens

Khadija Khataby[1, 2], **Faouzi Kichou**[3], **Chafiqa Loutfi**[2] and **My Mustapha Ennaji**[1]
[1]University Hassan II Mohammedia, Morocco
[2]Society Biopharma, Morocco
[3]Agronomic and Veterinary Institute Hassan II, Morocco

Avian infectious bronchitis (IB) is one of the most important viral diseases of poultry, affecting chickens of all ages and causing major economic losses in poultry flocks. The aim of this study is to evaluate pathogenicity and tissue distribution of Moroccan Italy 02 genotype of infectious bronchitis virus (IBV). 40 one-day-old specific pathogen free chickens were divided randomly into four groups. Group-1, 2 and 3 were inoculated intra oculo-nasally with 103.5 EID50 of virus and group 4 was kept as control. Chickens in each group were monitored for 14 days post-infection (dpi). Chickens in all infected groups showed severe respiratory signs, which most of them have been reproduced on 2 dpi, with varying times of appearance and disappearance. The infected birds appeared lethargic, reluctant to move with specific respiratory signs and macroscopic lesions. The specific histological lesions developed in all infected birds confirm the ability of the three tested strains to induce severe respiratory disease. The results at 14 dpi also revealed that all strains were able to induce serological response. Virus re-isolation from infected organs and amplification of the viral RNA by real-time PCR proved the presence of the virus in lung and trachea of infected chicks. Neither re-isolation nor significant viral RNA detection were detected in the kidney. These results demonstrated that the three strains Italy 02 genotype emerging in Moroccan poultry farms have a wide distribution for respiratory system without kidney damage and without causing mortality.

khadija.khataby@edu.uca.ac.ma

Plaque formation by Newcastle virus strain V4 on cell culture and characterization with RT-PCR

Mohammad Javad Mehrabanpour[1], **Sobhani S**[2] and **Mehrabanpour D**[2]
[1]Razi Vaccine and Serum Research Institute, Iran
[2]Islamic Azad University, Iran

Cloned vaccines are nowadays used in many countries. One of the ways for cloning a virus is propagation of the virus on cell culture to obtain discrete different plaques in order to study their morphology and genetics. In this study monolayer Madin-Darby Canine Kidney (MDCK) cell cultures were prepared by standard method. Various dilutions of the viruses were inoculated into monolayer MDCK cell cultures that were supplemented with magnesium sulfate and trypsin and over laid with agar medium. The viruses could reproduce on these cells and caused cytopathic effect and plaques. At 10^{-6} virus dilution, 6 various shape and size discrete plaques were obtained and inoculated into alantoic fluid, 9-11 days embryonated eggs. After 48 hours, the allantoic fluids contain plaques were harvested and their RNA extracted. Cleavage site of fusion protein with RT-PCR test was performed and the PCR products were purified and sequenced. The sequences of nucleotides and amino acids for each plaque were compared with those of the registered strain at gene bank as well as with each other. Molecular studies showed that all plaques are lentogenic strain of Newcastle disease virus and has about 97% to 99% homology with the strain V4 in the gene bank. The aim of this study is produce clear plaque by V4 strain of NDV on MDCK cell line and studies the molecular variations among them.

Mehrabanpourj@yahoo.com