

# 18. Poultry vector vaccines: innovative serological assays for vaccination monitoring and DIVA testing for H5 avian Influenza A

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Influenza viruses belong to the family Orthomyxoviridae. There are four types of influenza viruses: A, B, C and D, which are defined by the nature of their internal nucleocapsid antigen. Type A is the most conserved genus and can be further divided into subtypes based on their Hemagglutinin and Neuraminidase antigens. Some subtypes containing H5 or H7 are associated with highly pathogenic forms of the disease and high rate of mortality. A current H5 HPAI lineage has been circulating worldwide since 2004 and has been responsible for important poultry losses. To control poultry disease, vaccination is more and more used, especially with recombinant vaccine technology. In the last 5 years, successive waves of H5 Influenza in Europe pushed the health authorities to review their vaccination strategy concerning this virus. Given the need for rapid and reliable serological tools for monitoring of vaccination, IDvet has developed unique indirect ELISAs: one, based on H5 recombinant protein, for the monitoring of recombinant vaccines, and one, based on NP protein, for DIVA strategy (differentiated Infected from Vaccinated Animals). Layer and broiler flocks vaccinated with different technology of vaccines (H5 RNA, r-HVT-AI(H5) or inactivated sub-unit AIV-H5 vaccines) were tested. Antibody titers for H5 were evaluated using the H5 iELISA. Samples were also tested with the NP iELISA to monitor field challenge. For each tested flock, the following parameters were measured: mean titers, minimum, maximum, and CV%. All samples with titers higher than 732 for H5 iELISA, and higher than 668 for NP iELISA, were considered positive. All the flocks vaccinated with H5 vaccines were found positive with the H5 iELISA. Some of them were also found positive with the nucleoprotein iELISA. Therefore, the positivity of the H5 iELISA, belonging to negative flocks with the NP iELISA, demonstrated the detection of seroconversion induced by vaccines. The positivity of the NP ELISA in some flocks highlighted the presence of challenge with one HxNy strain. The H5 indirect ELISA presented is the only quantitative test for the specific detection of H5 antibodies which allows for H5 vaccination monitoring. The NP indirect ELISA is an excellent tool for the detection of wild virus in populations vaccinated with recombinant H5 vaccine.

Keywords: vector vaccines; vaccination monitoring; innovative ELISAs