Experimental and field results on the immunity induced by a rHVT-HA vector vaccine against H5N1 and other H5 type Highly Pathogenic Avian Influenza Viruses.

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Abstract
Vaccination against H5N1 Highly Pathogenic Avian Influenza (HPAIV) is one of the possible means available for affordably countries to control the disease once it has become endemic. Efficacy of vaccination against AI relies exclusively, but not necessarily, on the capacity of the vaccine to induce immunity against the targeted virus (which is prone to undergo antigenic variations) as well as its capacity to overcome interference with maternal immunity transmitted by dams to their progeny. This is a problem in intermittent circulating strains that are recognized as the most suitable in such settings. In the case of the European AIV H5N1, a single vector vaccine (rHVT-HA) is available that has been used in several European countries.

Introduction
The main issues associated with vaccination against Avian Influenza are: Antigenic stability of the challenging virus, efficacy of classical vaccinated chickens, Interference of Miller-Burnett Antibodies (MDA) with classical vaccinated vaccines, preventing early vaccination at the hatchery. 

Vaccination of high virus challenge at the flock level, which can hardly be achieved when vaccination is done at the farm [25].

The rHVT-HA vector vaccine (VetInfluenza A) is a recombinant vaccine constructed from the PIC-23 strain of HVT inside the genome of which, in a region recognized as non essential for the growth of the virus, a DNA sequence encoding for the HA of a highly pathogenic virus HPAIV (H5N1/IN/2006) has been introduced together with the challenge virus in order to express the HA protein. For safety reasons, the HPAIV strain of the HA gene has been modified to a D5a strain. This vaccine has been officially licensed by USDA in the USA and in 2012, from there in different countries. Since then, to complement information included in the original registration file, many scientific investigations, controlled trials and field studies have been conducted to evaluate knowledge regarding its characteristics and performance, so that prescription and use of this vaccine can rely on broader information. Besides, field experience on large scales has allowed us to enrich our experience. The objective of this poster is to present a brief overview of the data from various experiments conducted so far, and some of the conclusions to be drawn regarding the practical usage and monitoring of performance.

Vaccination vs. HPAIV

Inactivated influenza vaccines are used as the routine strategy for vaccination of chickens against H5N1 AI. However, a more rapid and efficient approach to control outbreaks of highly pathogenic influenza is to use vaccines administered subcutaneously. A recent report indicated that this strategy was feasible for the control of H5N1 AI in chickens, with the vaccine being used to control outbreaks of highly pathogenic influenza in chickens. The authors also reported that the vaccine was effective in preventing infection in chickens exposed to the highly pathogenic influenza virus. The vaccine was also effective in preventing mortality and morbidity in chickens exposed to the highly pathogenic influenza virus. The results of this study demonstrated the potential of using vaccines as an effective strategy for control of highly pathogenic influenza in chickens.

Immune response to vaccination and challenge (see figure 1)

After vaccination of SPF chickens, this rHVT-HA vaccine induces antibodies (Abs) clearly detectable by HI test, as soon as 3 weeks post vaccination in the majority of the tested birds, with titers reaching 6 to 8 log2 if HA antigen is used. This antibody response keeps on increasing until at 8 weeks of age and then reaches a plateau with titer values of 9 to 15 log2. In commercial chickens provided with passive immunity against HVT and AI (H5N1 or H5N2), under similar conditions of testing, antibody response is slower in the rise to titers and reaches lower HI titer values at all time points. Experiments have shown that a high level of protection is reached before homologous HI antibodies are detected in all chickens. When the vaccine is given to the vaccine but homologous to the challenge virus, the detected level of antibody response in chickens is generally lower but has a predictive value regarding protection. It is thus concluded that other components of the immune response are induced by the vaccine, and in particular the cellular and mucous immune system plays an important role (1, 4, 6).

After challenge, a clear rise in heterologous HI antibody titers is observed. The rise in heterologous HI antibody titre is more limited than in homologous HI antibodies, and can hardly be systematically attributed to challenge virus replication. A similar rise in intensity is also observed in vaccinated but non challenged chickens, as a continuation of the natural antibody response development. HA titre increase of the non- or low anti-HA antibody vaccine group is to be considered as a "full virus", do confirm this statement. In some experiments, only few vaccinated and challenged chickens turned positive in these assays when used, indicating no or little replication of the challenge virus (9, 10). The use of such a test in rHVT-HA vaccinated chickens allows for a GISD monitoring and detection of excretion and evolution of antibodies (10).

Immune response to vaccination and challenge (see table 1)

2 criteria are considered as important regarding protection: clinical signs (morbidity and mortality) and shedding of the challenge virus after inoculation.

Table 1: compilation of result only collected through various experiments conducted with rHVT-HA (VetInfluenza A) vaccine. Please refer to corresponding publications for more details.

Onset of Immunity (O01)

The type of chicken i.e. breeders versus layers: O01 is earlier in broilers.

"The presence of MDA against AV: AV response earlier in MDA-> chicks. chickens, especially free range, may have been vaccinated with other different vaccines exclusively from vaccine replication and 2 to 3 weeks are necessary to reach satisfactory levels, although earlier resistance to challenge has not been observed.

In chickens with MDA against AV, protection also comes from passive immunity which is actually protects against challenges induced by vaccination cumulates with passive protection and overall protection is increased. For this reason, in endemic countries, vaccination of breeders against AI complies and does not contraindicate vaccination of broilers and pullets.

Duration of Immunity (O01)

Deaths regarding efficacy of vaccine to long lived chickens against very late challenge are not available yet. However, the following elements are suggesting of a long lasting protection:

"HVT is known to persist forever in infected animals, so that expression of the HA inserted inside the rHVT-HA vaccine is likely to persist as a corresponding residual protection."

Protection against mortality was good, very varying from 60 to 100%, including against challenge with H5N1 strains of different clades, or different pathogenic (H5N1, H5N2) HAs without any differences regarding homology of the HA genes. This allowed concluding to "cross-clade" protection of the same type, which was the one described.

Reduction of shedding considering the percentage of shedding as well as the amount of viruses was always detected with different intensities, believed that the different experimental conditions as well as the different techniques used to detect and quantify excretion (RT-PCR of different types, antigen detection ELISA, various species tested by antigen detection ELISA), as well as the percentages of shedding 2 or 3 days post challenge are given as an illustration, although probably comparable with excretion noted in the field.

Target species

Experiments have demonstrated replication of rHVT-HA, expression of the HA antigen and induction of protection in chickens of the SPF, broiler and layer types, as well as in turkeys (14). Replication has also been evidenced in waterfowl of the Goose, Muscovy ducks and Mallard ducks types. Surprisingly, very rapid replication and no protection were observed in Pekin ducks (12). Replication was also observed in quails and pheasants but not in pigeons (Ceva registration dossier).

Conclusion

In 2012, this rHVT-HA vector vaccine against Avian Influenza due to H5 AI viruses was first licensed in the USA and since then commonly commercially available in various countries where the disease is endemic (Egypt, Bangladesh, Malaysia), but, for local administrative reasons, not yet all.

Protection induced by this vaccine, following a single injection to day-old chicks at the hatchery, has been tested in various locations, by independent researchers, under various experimental and field conditions against AIV H5N1. This vaccine has been used in many different countries, and a wide range of vaccine types has been used. The vaccine has been shown to be effective in preventing infection in chickens exposed to the highly pathogenic influenza virus. The vaccine was also effective in preventing mortality and morbidity in chickens exposed to the highly pathogenic influenza virus. The results of this study demonstrated the potential of using vaccines as an effective strategy for control of highly pathogenic influenza in chickens.

Because of all these characteristics as well as because of the possibility of a semi-automatic injection in the hatchery, which is the only practical way to reach a high percentage of vaccine coverage (up to 98%), it is believed that this rHVT-HA vaccine is an extremely powerful tool to vaccine against Avian Influenza caused by H5 type AI viruses. Because of its superb antigenic antigens, its use does not interfere with a serological detection (IDHA) or serology-based vaccine.

References
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