

# MERCK ANIMAL HEALTH

## TECHNICAL SERVICES BULLETIN

INNOVAX®-ILT:

### *Full Dose Ensures Complete Protection*

The broiler market is highly cost-competitive, forcing live production managers to make decisions intended to reduce production cost, even in the face of a severe disease like infectious laryngotracheitis (ILT). A common cost reduction strategy is to use a partial dose of vaccine for the short-lived broilers.

A study conducted at the Poultry Diagnostic and Research Center by Guillermo Zavala, et al. demonstrates the potential hidden cost of using a fractional dose: reduced protection. The study was conducted using full dose, 1/2 dose and 1/4 dose of Innovax-ILT, with a full dose PFU count of 6460. The birds were vaccinated at one day of age by subcutaneous injection, and they were challenged at 25 days of age using virulent ILT virus isolated from a Georgia field case associated with high mortality (ILTV PDRC 63140 titer  $10^{3.5}$  TCID<sub>50</sub>/dose).

Note: 25 days of age is early for a laboratory challenge study, which would normally be conducted at 35 days of age. This study tests the early limits of immunity development.

#### Conclusions

- A clear statistical difference was observed in protection against clinical signs at 5 days post-challenge, with more severe clinical signs observed in groups vaccinated with ¼ or ½ dose of Innovax®-ILT compared to those vaccinated with a full dose.
- The virulent ILTV load in the trachea was also significantly higher in groups vaccinated with ¼ or ½ dose compared to those vaccinated with a full dose.
- The rHVT-ILT vaccine virus load in the spleen at 4, 7 and 14 days was progressively higher for each dose level: ¼ dose, ½ dose and full dose, indicating that vaccine virus replication was greatest for the birds vaccinated with full dose at each time point.
- Immunity against virulent ILTV challenge is developed by exposure of the broiler immune system to sufficient ILT antigen to induce a strong immune response. The higher replication rate of a full dose of vaccine leads to a more complete immunity by 25 days of age as measured by clinical signs and by virulent ILT virus load in the trachea at 5-days post challenge. The reduced clinical signs and virus load may be essential to prevent the spread of virulent ILT in a field outbreak involving multiple houses or farms.
  - Merck Animal Health recommends the use of a full dose of Innovax-ILT to ensure maximum protection with the earliest possible onset of protective immunity.

#### STUDY OBSERVATIONS:

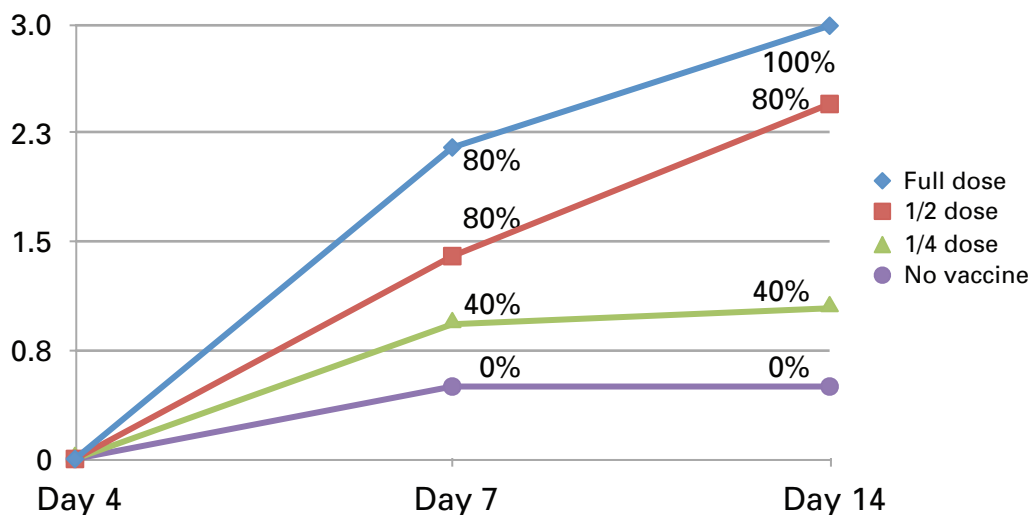
Several parameters were measured: rHVT-ILT virus quantification in the spleen at 4, 7 and 14 days of age (Figure 1), clinical signs at 5 days post-challenge (Figure 2), and virulent ILTV in the trachea at 5 days post-challenge (Figure 3). Virus quantification was based upon quantitative polymerase chain reaction (qPCR) methodology.



### rHVT-ILT Vaccine DNA Load

Quantitative real-time PCR in a duplex assay was used to determine the rHVT-ILT DNA load in the spleen for 14-days post-vaccination. The percentage of HVT-positive samples was also recorded. Results are summarized in Figure 1.

**Figure 1: rHVT-ILT vaccine DNA load in the spleen (mean  $\log_{10}\Delta\Delta Ct$  values) and percent of samples positive for HVT in the spleen.**



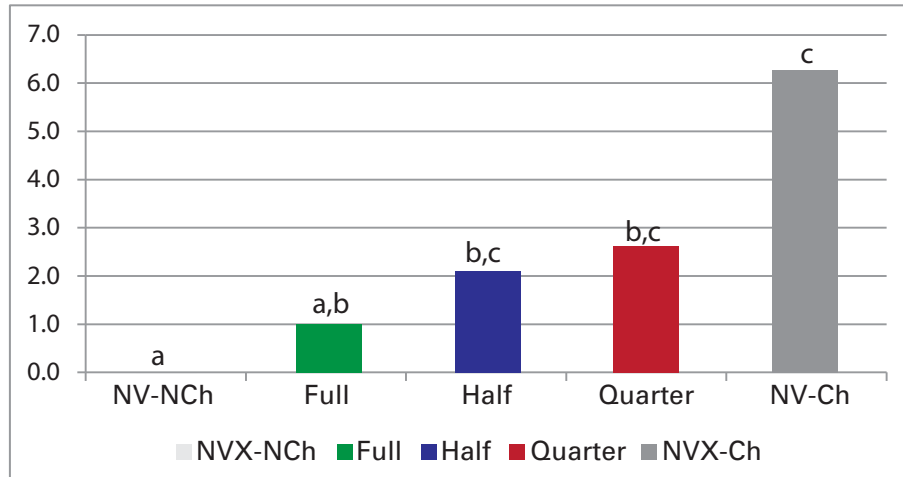
The development of immunity against virulent ILTV challenge depends upon exposure of the immune system to ILT proteins coded by the genetic material inserted into the rHVT-ILT virus. Higher virus load means more protein to stimulate the immune response. The full vaccination dose reaches higher overall virus load by 14 days of age and the multiplication occurs at a faster rate than the fractional doses. Only the full dose group reached 100% HVT-positive samples by 14 days, the half dose and quarter dose groups were only 80% and 60% positive, respectively.

### Clinical Signs

The birds were challenged at 25 days of age, and clinical signs were scored at 5 days post-challenge according to the severity of depression, conjunctivitis and respiratory signs using a scoring system of 0 (normal), 1 (mild), 2 (moderate) or 3 (severe). Any deaths automatically received a score of 9. The challenge strain was PDRC isolate 63140 at  $10^{3.5}$  TCID<sub>50</sub>/bird. This is a rather high challenge dose in a young bird to highlight the effects of fractional dosing. Results are summarized in Figure 2.

There was no significant difference between the birds vaccinated with a full dose of Innovax-ILT and the non-vaccinated, non-challenged controls (superscript "a"). The birds that received a fractional dose were not significantly different than the non-vaccinated, challenged controls (superscript "c").

**Figure 2: Clinical Signs Score at 5 days following 25-day challenge with virulent ILTV**

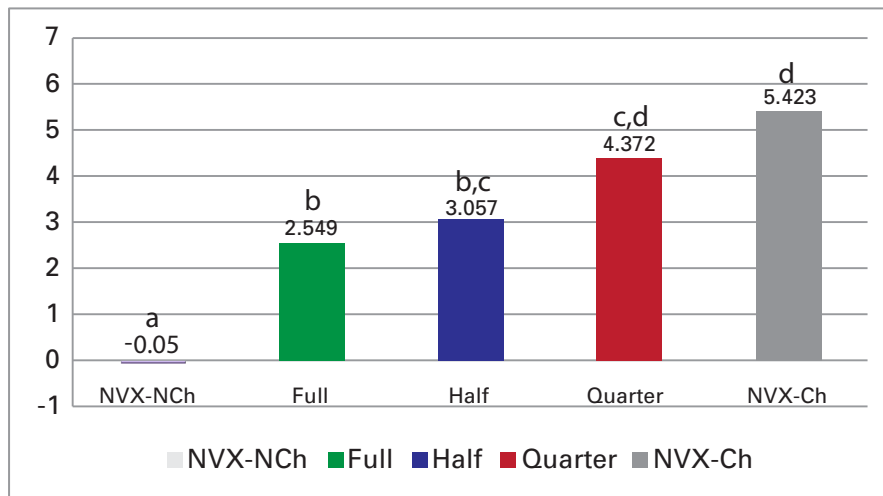


<sup>a,,b,c</sup> Bars without a common superscript differ significantly (P<0.05)

### Virulent ILT virus present in the trachea at 5 days post-challenge

Quantitative real-time PCR in a duplex assay was used to determine the virulent ILT virus load in the trachea at 5 days post-challenge. The virus load was significantly reduced in the birds receiving a full dose of Innovax-ILT, while those receiving ¼ dose had a virus load that was not significantly different from a non-vaccinated, challenged bird. Results are summarized in Figure 3.

**Figure 3: Virulent ILTV DNA in the trachea at 5 days post-challenge ( $\log_{10}\Delta\Delta\text{CT}$  values, represented by numbers above the colored bars)**



<sup>A,B,C</sup> Bars without a common superscript differ significantly (P<0.001)

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