# SCHERING-PLOUGH ANIMAL HEALTH technical services bulletin

## In-ovo IBDV Vaccination of Maternal Antibody-Positive Broiler Chicks With Univax-BD®

## Part I: Impact of in-ovo IBDV Vaccination on the Bursa

yperimmunization of broiler breeders against infectious bursal disease virus (IBDV) is a common practice among US broiler integrators. Intensive breeder vaccination programs are designed to maximize maternal antibody titers of the progeny, protecting broiler flocks from the immunosuppressive effects of early subclinical IBDV infection. Integrators, however, have questioned the effectiveness of day-ofage or *in-ovo* IBDV vaccination in hightitered progeny; many have abandoned the

#### practice based on the supposition that maternal antibodies would neutralize vaccine virus before it could be beneficial.

A study presented by Dr. John McCarty at the 2003 American Veterinary Medical Association-American Association of Avian Pathologists annual convention demonstrated the effects of *in-ovo* IBDV vaccination on the bursas of broilers with high maternal antibody titers.

#### **Study Design**

For the study, 70 eggs with maternal antibodies to IBDV were obtained from a commercial broiler integrator and 70 specificpathogen-free (SPF) eggs were used as antibody-free controls. Both groups of eggs were injected at 18 days of incubation with a full dose of an intermediate classic strain IBDV vaccine (Univax-BD\*, Schering-Plough Corporation). The eggs were hatched and the hatchlings were grown in isolation units through 20 days of age.

Five birds were sacrificed from each group daily beginning 1 day post-hatch. Bursas were harvested and submitted either for histological examination or for IDEXX reverse transcriptase/polymerase chain reactionrestriction fragment length polymorphism (RT/PCR-RFLP) assay. The assay determines the presence or absence of vaccine virus in the tissue. Where bursal size permitted, half of each bursa was submitted for histological examination and half for RT/PCR-RFLP.

## **Key Points**

Transient vaccine virus was detected in the bursa of *in*-ovo vaccinated chicks with high maternal antibody titers.

Histopathology demonstrated transient variation in the follicle size of bursas from vaccinates between 8 and 16 days of age. Follicle size returned to normal by 16 days of age.

Univax-BD<sup>®</sup> vaccine administered *in-ovo* successfully reached and had an impact on the bursa despite the presence of high maternal antibody levels. Table 1: PCR virusdetection in maternalantibody-positive vs.maternal antibody-negative broilersfollowing *in*-ovo IBDVvaccination with anintermediate classic(molecular group 4)IBDV vaccine.

Test completed by IDEXX

| Flock Status   | Tissue | Age | Sample #   | RT/PCR             | Subtype           | BstN       | Pattern     | 1   |     | Mbc<br>102 | I Patte کا 1  | irn<br>1 220 |
|----------------|--------|-----|------------|--------------------|-------------------|------------|-------------|-----|-----|------------|---------------|--------------|
|                | adkı   |     |            | Vesuits            | 'n                | 7/1 607 00 | -<br>+<br>- |     | 004 | 604        | 77 700        | 477 +        |
| I. Ab Negative | Bursa  | р   | 12411205-1 | Negative           | N/A               |            |             |     |     |            |               |              |
| 2. Ab Positive | Bursa  | p   | 24  205-2  | Negative           | N/A               |            |             |     |     |            |               |              |
| 3. Ab Negative | Bursa  | 5d  | 12411205-3 | Positive           | Molecular Grp. 4  | BAND BAND  | BAND        | BAN |     |            | 3AND          | BAND         |
| 4. Ab Positive | Bursa  | 5d  | 12411205-4 | Positive           | Molecular Grp. 4  | BAND BAND  | BAND        | BAN |     |            | <b>BAND</b>   | BAND         |
| 5. Ab Negative | Bursa  | ΡĹ  | 12411205-5 | Positive           | Molecular Grp. 4  | BAND BAND  | BAND        | BAN |     |            | <u> 3</u> AND | BAND         |
| 6. Ab Positive | Bursa  | ΡĹ  | 24  205-6  | Positive           | Molecular Grp. 4  | BAND BAND  | BAND        | BAN |     |            | 3AND          | BAND         |
| 7.Ab Negative  | Bursa  | P6  | 12411205-7 | Positive           | Molecular Grp. 4  | BAND BAND  | BAND        | BAN |     |            | 3AND          | BAND         |
| 8. Ab Positive | Bursa  | P6  | 12411205-8 | very weak Positive | Unable to do RFLP |            |             |     |     |            |               |              |

| Maternal Antibody-Positive |                               | Maternal Antibody-Negative   |               |                               |   |
|----------------------------|-------------------------------|--|---------------|-------------------------------|---|
| Age<br>(Days)              | Histologic<br>Bursal<br>Score | Diagnosis  | Age<br>(Days) | Histologic<br>Bursal<br>Score | Diagnosis   |
| 2                          | 1.2                           | Normal   | 2             | 4.0                           | Acute follicle necrosis consistent with IBDV                      |
| 4                          | 1.0                           | Normal   | 4             | 4.0                           | Severe variation in follicle size                                 |
| 6                          | 1.0                           | Normal   | 6             | 4.0                           | Acute follicle necrosis<br>consistent with IBDV                   |
| 7                          | 3.4                           | Severe variation in follicle size  | 7             | not done                      |   |
| 8                          | 3.2                           | Moderate to severe<br>variation in follicle size<br>consistent with IBDV | 8             | 3.5                           | Severe variation in follicle size                                 |
| 9                          | 2.2                           | Moderate variation in follicle size                                      | 9             | not done                      |   |
| 10                         | 2.2                           | Moderate variation in follicle size                                      | 10            | 3.6                           | Sections show chronic<br>IBDV infection with<br>some regeneration |
|                            | 1.8                           | Mild variation in follicle size  | No furt       | her samples s                 | ubmitted  |
| 12                         | 1.6                           | Moderate variation in follicle size                                      |               |                               |   |
| 13                         | 1.8                           | Mild variation in follicle size  |               |                               |   |
| 4                          | 1.4                           | Mild variation in follicle size  |               |                               |   |
| 15                         | 1.4                           | Sections normal with<br>I stressed bird                                  |               |                               |   |
| 16                         | 1.6                           | Sections normal with<br>I stressed bird                                  |               |                               |   |
| 17                         | 1.2                           | Sections normal for this age   |               |                               |   |
| 18                         | 1.0                           | Sections normal for this age   |               |                               |   |
| 19                         | 1.0                           | Sections normal for this age   |               |                               |   |

Table 2: Bursal histopathology in maternal antibody-positive vs. maternal antibodynegative broilers following *in*-ovo IBDV vaccination.

Smaller bursas were submitted in entirety for either histological examination or RT/PCR-RFLP on alternate days. Samples were collected from five birds daily from day 1 through day 20 post-hatch.

Bursas submitted for histological examination were scored according to follicle size and appearance. A score of 1 to 2 was considered normal, a score of 3 to 4 was considered unhealthy while a score between 2 and 3 was considered suspect.

#### **Results**

IBDV was detected in both the antibodynegative and antibody-positive groups by IDEXX RT/PCR-RFLP. Virus was detected in the antibody-positive birds beginning at 3 days of age and was detectable through 9 days of age. The test results showed a weak positive IBDV at 9 days of age but was negative by 11 days of age.

Virus was detected in the antibody-negative birds beginning at 5 days of age and was still strongly positive at 9 days of age. No further RT/PCR-RFLP testing was completed after 9 days of age (see Table 1, page 2).

In both cases, the virus detected in the bursas matched the molecular pattern of the vaccine virus (molecular group 4).

Histologically, bursas from antibody-positive birds demonstrated variation in follicle size (score 3.2) beginning at 8 days of age. By 16 days of age, follicles had returned to normal (score 1.6).

Bursas from antibody-negative birds showed acute follicle necrosis consistent with IBDV infection by 2 days of age (score 4.0). At 10 days of age, the bursas showed signs of chronic IBDV infection with some regeneration (score 3.6; see Table 2, page 3).

#### **Conclusion and Discussion**

This study demonstrates that there are some differences in the way that maternal antibody-positive and antibody-negative chicks respond to *in-ovo* IBDV vaccination, but that maternal antibody-positive chicks do respond to *in-ovo* IBDV vaccination with Univax-BD. Despite the presence of maternal antibodies, transient vaccine virus was detected in the bursa of vaccinated chicks from days 3 through 9 followed by histological changes in the bursa follicle size from days 8 through 16. The follicles recovered completely by 16 days of age.

The maternal antibody-positive chicks experienced milder histological changes in the bursa compared to antibody-negative controls.

In Part 2 of this technical bulletin series, the impact of *in-ovo* IBDV vaccination with Univax-BD on the *immunity* of maternal antibody-positive broilers is investigated by sequential challenge with classic IBDV and the Delaware E variant of IBDV.

