Use of Serology to Solve Clinical Case Studies & Interpreting Poultry Baselines

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Agenda

1. Uses of Serology
2. Developing a Flock Health Monitoring Program
3. Interpreting Data from IDEXX ELISA testing
4. Epidemiologic Uses of Serology
5. 10 Principles of Serologic testing
6. Conclusions
Standard Diagnostic/Monitoring Tools

- Clinical and postmortem examination
- Serology
- Bacteriology and Mycology
- Virology
- Histopathology
- Molecular detection

Serology: Common Applications

- Disease monitoring/surveillance
- Diagnosis of Field infections
- Epidemiology Studies
- Vaccine Administration and Efficacy monitoring
Disease Monitoring/Surveillance

- Common diseases/pathogens:
  - Newcastle (NDV)
  - Infectious Bronchitis (IBV)
  - Bursal Disease (IBD)
  - Reovirus (REO)
  - Avian Encephalomyelitis (AE)

- These diseases are frequently monitored via watching the mean titer value over time

- Diseases of great concern:
  - Mycoplasma gallisepticum (MG)
  - Mycoplasma Synoviae (MS)
  - Avian Influenza (AI)

- These are typically given a flock-wide positive/negative value

- Purposes
  - Assess field challenge
  - Timely response to disease
  - Monitor management changes

What is a Baseline?

- A baseline is a range of titer values which is set based on previous experience to define average expected titers in flocks with known production parameters
- Typically this includes a low, medium, and high value. If there is an excursion high or low, this is more easily noted with a baseline to use for reference
- It is important that the baselines be specific to:
  - Bird types
  - Bird ages
  - Vaccine schedule
  - Vaccine type
  - Vaccine program
  - Geography
  - Seasonality
Benefits of Using a Baseline

- Compare new data with historical information previously archived
- Provides useful information for anticipating outbreaks, evaluating trends, and comparing vaccination programs
- Elevated titer values can suggest infection early in disease process
- Low titer values may reflect improper vaccine administration or immunosuppression
- Excellent way to monitor management changes and establish epidemiological trends
- Quickly noting changes in the birds can reduce production losses associated with disease
- Helps monitor return on investment of biosecurity programs

Why Titer Values are Established

Vaccination of Adult Breeder or Layer Birds

- Vaccinating is expensive and time consuming
- Timing of testing and vaccination is important to make sure the bird’s immunity does not wane to dangerous levels before revaccination
- Monitoring vaccine titers helps prevent field infections and production losses
- Revaccinating early can be wasteful and ineffective
Why Titer Values are Established, continued

Assuring robust immunity in breeding birds
• It is important that parent stock have strong antibody levels to allow for adequate passage of MDA to their chicks

• The presence of antibodies indicates that the birds have been exposed to field challenge or had contact with an antigen

Disease detection
• A humoral response can be detected as early as 2 weeks post vaccination. When using live vaccines, we recommend sampling 4 weeks post vaccination and 6-8 weeks post vaccination when monitoring an inactivated vaccine. Having a baseline will help to distinguish this normal vaccine response from a disease challenge response.

• Rapid detection of field challenge responses and subsequent biosecurity adjustments can prevent further spreading of the disease

Basics of Developing an Effective Monitoring Program
Key Points in Designing a Monitoring Program

• Select correct serological test
  - Validation of the assay (testing for performance of the test)
  - Sensitivity
    - Differentiation of infected and vaccinated animals (DIVA), field virus, variants
  - Specificity, cross-reactions
  - Repeatability

• Keep in mind sample size and sample timing
  - Make sure you are examining a statistically significant number of samples from each flock
  - Compromising on sample size may cause unreliable and misleading results
  - The more variable you expect the titer to be, the more birds you should sample per flock

• Interpret the results
  - Use baselines to identify trends and monitor changes in your protocols over time

Baseline ELISA Serology (IBV)
Expect Baseline Variation

<table>
<thead>
<tr>
<th>BROILERS (Processing ELISA Titers)</th>
<th>NORTH GA GMT (FLOCKS)</th>
<th>SOUTH GA GMT (FLOCKS)</th>
</tr>
</thead>
<tbody>
<tr>
<td>NVD</td>
<td>339 (190)</td>
<td>427 (87)</td>
</tr>
<tr>
<td>IBV</td>
<td>2158 (191)</td>
<td>903 (87)</td>
</tr>
<tr>
<td>IBDV</td>
<td>7668 (120)</td>
<td>5435 (87)</td>
</tr>
<tr>
<td>REO</td>
<td>707 (206)</td>
<td>649 (6)</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>BREEDERS (20-30 Weeks of Age)</th>
<th>NORTH GA GMT (FLOCKS)</th>
<th>SOUTH GA GMT (FLOCKS)</th>
</tr>
</thead>
<tbody>
<tr>
<td>NVD</td>
<td>3467 (177)</td>
<td>1762 (72)</td>
</tr>
<tr>
<td>IBV</td>
<td>6058 (176)</td>
<td>4863 (70)</td>
</tr>
<tr>
<td>IBDV</td>
<td>11866 (144)</td>
<td>8609 (71)</td>
</tr>
<tr>
<td>REO</td>
<td>5509 (206)</td>
<td>5350 (38)</td>
</tr>
</tbody>
</table>

Shared via the Georgia Poultry Lab website
Georgia Poultry Laboratory Website (USA)

- http://www.gapoultrylab.org/
Understanding Variations in Titers

- Caused by normal biological variation
  - Geographic
  - Current field challenges
  - Management conditions

- Caused by vaccine failure

- Caused by human or instrument errors
  - Improper sample collection, storage or handling
  - Improper ELISA technique
  - Instrument malfunctions

- Poor sample management may lead to errant or inaccurate results

- Use correct ELISA techniques to ensure reproducibility and reliability
Interpreting the Data from your IDEXX ELISA

Baseline Example: IBV

IDEXX IBV Antibody ELISA Sample Baseline

- High Titers
- Medium Titers
- Low Titers

Test With Confidence™

IDEXX Poultry Diagnostics
### Example: IDEXX Baselines

<table>
<thead>
<tr>
<th>Test</th>
<th>Vaccine Type</th>
<th>Mean Titer (weeks after vaccination)</th>
<th>Mean Titer Day-Old Chicks</th>
</tr>
</thead>
<tbody>
<tr>
<td>AE</td>
<td>Live</td>
<td>3000–4000 (5–8 weeks)</td>
<td>—</td>
</tr>
<tr>
<td>IBV</td>
<td>Live</td>
<td>1000–4000 (3–5 weeks)</td>
<td>—</td>
</tr>
<tr>
<td></td>
<td>Inactivated</td>
<td>5000–16000 (5–8 weeks)</td>
<td>2000–6000</td>
</tr>
<tr>
<td>IBD</td>
<td>Live</td>
<td>1000–4000 (3–5 weeks)</td>
<td>—</td>
</tr>
<tr>
<td></td>
<td>Inactivated</td>
<td>4000–20000 (5–8 weeks)</td>
<td>3000–10000</td>
</tr>
<tr>
<td>NDV</td>
<td>Live</td>
<td>1000–4000 (3–5 weeks)</td>
<td>—</td>
</tr>
<tr>
<td></td>
<td>Inactivated</td>
<td>10000–32000 (5–8 weeks)</td>
<td>5000–15000</td>
</tr>
<tr>
<td>REO</td>
<td>Live</td>
<td>2000–4000 (3–5 weeks)</td>
<td>—</td>
</tr>
<tr>
<td></td>
<td>Inactivated</td>
<td>4000–10000 (5–8 weeks)</td>
<td>3000–9000</td>
</tr>
</tbody>
</table>

Based on two live priming and one inactivated boosting at 16–18 weeks, 1:500 dilution.

Titers may vary according to type of bird, vaccine type, vaccination program, etc.

### ELISA Serology vs. Condemnations

Company “C”

**OCTOBER 2010**

**DECEMBER 2010**

- **IBV GMT**
- **AIRSAC CONDEMN (%)**

Broiler Flocks
Baseline IBV HI Serology
Company “E”

Epidemiological Studies
Company A IBD-XR Titers vs GA Average
Company A: Red, GA Average: Blue
Breeder Baseline Examples

Titer Groups

- A titer group is a numeric value assigned to a titer value range, used to simplify interpretation.

- Remember that by grouping titers, you lose sensitivity to changes in your individual data points.
  - Example, titers of 8000 and 9999 may fall in the same titer group, while 10,000 falls into a different group.

- Baseline tracking is better accomplished by tracking actual titers.
Tracking Values for a Baseline

**In xChekPlus® Software**
- Data generated by the lab can be evaluated in several ways using the IDEXX software
- Eliminates errors in recording or data transfer
- Built in Baseline and Multiple case reporting features can be employed

**In Microsoft Excel®**
- A master workbook can be generated and then data can be added from each run
- Consider different tabs for various tests, and one master worksheet to compare tests performed at the same time
- Excel allows the generation of bar graphs and line graphs

Serology: Basic Principles

1. Use rapid, convenient, specific & economical methods
2. Build in-house databases (baseline)
3. Use regional baseline comparisons
4. Consider seasonal baseline comparisons
5. Assign serology its relative diagnostic value
Serology: Basic Principles

6. Avoid single time point evaluations
7. Comply with official regulations
8. Do not assume protection or exposure based just on serology
9. Know the local epidemiology
10. Apply qualitative and/or quantitative serology appropriately

Conclusions

• Make sure the data you are recording is an accurate interpretation of the bird’s status
• The more information about the flock you can incorporate into your baseline, the more valuable your results will be
• Utilize the features provided in xChekPlus® software to help you generate your baselines
• Use resources available to benchmark your titers to those in your region
• Make sure the information you are collecting is the information needed to make management decisions that impact the economic performance of your company/customer
Questions?