Troubleshooting a Marek’s Disease Outbreak

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Marek’s Disease

• Marek’s Disease was first reported in 1917 by Dr. Jozsef Marek in Budapest, Hungary

• Three serotypes described

• Causes malignant tumors in the chicken’s skin and internal organs
Marek’s Disease

- The field virus tissue tropism and severity of the lesions may vary
- Causes mortality & condemnations at the processing plant
- Causes immunosuppression
Marek’s Disease Virus Serotypes

<table>
<thead>
<tr>
<th>Serotype 1</th>
<th>Serotype 2</th>
<th>Serotype 3</th>
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<tbody>
<tr>
<td><strong>Oncogenic strains</strong></td>
<td>SB1</td>
<td>HVT</td>
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<tr>
<td>- GA</td>
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<td>- MD11</td>
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<td>- MD5</td>
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<tr>
<td>- TK</td>
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<td>- RB1B</td>
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<tr>
<td>- etc.</td>
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<tr>
<td><strong>Low Pathogenicity</strong></td>
<td>301B</td>
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<tr>
<td>strains</td>
<td></td>
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<tr>
<td>- CVI988 (Rispens)</td>
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<td>- Clone C</td>
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<td>- CR-6</td>
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</table>
Marek’s Disease

• The virus replicates to the fully infectious form in the feather follicles

• The route of infection is the respiratory system by inhaling infectious viral particles

• Tumors (lesions) occur due to the malignant transformation of the lymphocytes
Marek’s Disease

- The first wave of virus infection targets the bursa and thymus
- The infection may produce immunosuppression in a transient or permanent form
- The disease may show various clinical signs
Marek’s Disease

- Very virulent strains (vvMDV+) have been reported in a number of countries around the world and have affected broilers, breeders and commercial layers

- Great economic impact

- Marek’s can be prevented by implementing a vaccination program
Clinical Signs
Marek’s Disease in Broiler Breeders
Marek’s Disease in Broiler Breeders

Paralysis & depression
Paralysis

Marek’s Disease in Broiler Breeders
Paralysis

Marek’s Disease in Broiler Breeders
Leg tumors (Alabama red leg)

Marek’s Disease in Broiler Breeders
Skin Tumors

Marek’s Disease in Broiler Breeders
Spleen Tumors

Marek’s Disease in Broiler Breeders
Intestinal Tumors

Marek’s Disease in Broiler Breeders
Marek’s Disease in Broilers
Marek’s Disease in Broilers

Leg tumors (Alabama red leg)
Marek’s Disease in Broilers

Leg tumors
Marek’s Disease in Broilers

Skin Tumors
Marek’s Disease in Broilers

Liver Tumors
Marek’s Disease in Broilers

Liver Tumors with necrotic centers
Marek’s Disease in Broilers

Control Bursa  MDV-Infected bursa
Marek’s Disease in Commercial Layers
Marek’s Disease in Commercial Layers

Liver Tumors in Layers
Marek’s Disease

The only known methods of preventing Marek’s Disease are:

• Vaccination
• Good flock management
• Cleaning, disinfection and biosecurity
Marek’s Vaccination Failure

MD outbreaks may appear in vaccinated flocks because:

• High field challenge by very virulent Marek’s disease virus strains (vvMDV or vvMDV+)
• Interval vaccination – Challenge
• Presence of immunosuppressive agents
• Inadequate vaccination programs
• Poor vaccine preparation and administration
Marek’s Vaccination Failure

• High Field challenge by Very Virulent Marek’s Disease Virus (vvMDV or vvMDV+) Strains
  
  - If the field challenge is too high, it can overcome the vaccine immunity and produce an outbreak even in older birds
  
  - Previous reports indicate the incidence of Marek’s outbreak in layers and broiler breeders over 40 weeks of age
Marek’s Vaccination Failure

• The Presence of other immunosuppressive agents may interfere with the development of protection against Marek’s disease
  • Infectious Bursal Disease Virus (IBDV)
  • Chicken Anemia Virus (CIAV)
  • Avian Leukosis subtype J
  • Mycotoxins
Marek’s Vaccination Failure

• Interval Vaccination – Challenge

• Good cleaning and disinfection should delay the exposure of baby chicks to field challenge

• This delay of the exposure will allow the proper development of vaccine-provided protection

• Day old chicks should be placed in an environment as clean as possible
Interval Vaccination-Challenge
Chart 1. Percent Total Affected (Lesions Plus Mortality) 2 Through 8 Weeks After Challenge With MDV T.K. (Respiratory Route Using a Shedder System) at One Day of Age or When 5 Days Old

Montiel, 1995
Marek’s Vaccination Failure

• Inadequate Vaccination Programs
  • Vaccination programs must be designed according to the type of bird and field challenge
  • Re-vaccination may be considered under high challenge conditions
### Most commonly Used MD Vaccination Programs

<table>
<thead>
<tr>
<th>Program</th>
<th>Broilers</th>
<th>Layers</th>
<th>Broiler Breeders</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>HVT</td>
<td>HVT+Rispens</td>
<td>HVT+Rispens</td>
</tr>
<tr>
<td>2</td>
<td>HVT+SB1</td>
<td>HVT+Rispens +SB1</td>
<td>Rispens</td>
</tr>
<tr>
<td>3</td>
<td>Rispens &amp; Combinations</td>
<td>HVT+SB1</td>
<td>HVT+SB1</td>
</tr>
</tbody>
</table>
Commonly Used MD Vaccination Programs

- The best program in meat-type birds is the HVT+Rispens combination.
- More recent publications suggest that the best program for commercial layers is the HVT+Rispens+SB1 combination.
Percent Protection in Commercial Female Broilers Challenged with the MDV TK Isolate* by intra-abdominal injection 5 days post vaccination

*Challenge dose: 1:100 dilution of TK seedstock
Marek’s Vaccination Failure

• Vaccine preparation and administration

  • Must follow the manufacturer’s instructions

  • Improper vaccine preparation and administration techniques may seriously compromise the vaccine titer.
Marek’s Vaccination Failure

- Improvement in vaccine preparation and administration has contributed to the decrease in the incidence of Marek’s disease in a number of operations around the world.
Preparation for Vaccine Mixing
Mix vaccine in a clean, low-traffic area.
Keep Frozen Vaccines Properly Identified.
Liquid Nitrogen Safety

Protective Clothing Required
Routinely monitor the level of liquid nitrogen in the tank.
Vaccine Handling
Check Frozen Vaccines Ampules

- Marek’s vaccines ampules may be stored inverted in the liquid nitrogen tank. When checking ampules, make sure you follow the quality assurance indicator before thawing.
Quality Assurance Indicator

(Product in the frozen state)

Acceptable  Acceptable  Unacceptable
Store Diluent at

*Room Temperature*
Examine Each Bag for Contamination.
Examine Each Bag for Contamination.

Good diluent  Contaminated
Fill thaw-bath with distilled water.

Do not add bleach or sanitizer.
Ampules may be thawed in water at 27-37 °C (80-98.6 F).

Most Hatcheries keep thaw-bath at 80 °F (27 °C).

Thaw no more than what is needed for immediate use.
Preparation Checklist

• Mixing Room
  • clean, low traffic

• Liquid Nitrogen
  • safety considerations, adequate levels of liquid nitrogen, no Newcastle contamination

• Diluent
  • room temperature, no contamination

• Thaw-bath
  • Fresh distilled water, 80 – 96 °F
Mixing the Vaccine
Always mix the vaccine following the instructions on the vaccine insert
Clean and Prepare the Working Area
Wash hands thoroughly before mixing.
Adding Antibiotics

- Vaccines are not licensed for combination with these products.
- Some antibiotics can significantly reduce the Marek’s vaccine titer
Calculating the Number of Ampules Needed
Determining the Number of Ampules Needed

0.05 ml/egg

1200 ml bag ÷ 0.05 ml per chick = 24,000 eggs per bag

1 ampule = 2000 doses (check ampule)

At full dose, you would need 12 ampules (24,000 ÷ 2000) of the 2000 dose vaccine or 6 ampules of the 4000 dose vaccine
Thaw ampules by gently swirling the ampules in the thaw-bath.
Never Leave Ampules Unattended
Once an ampule has thawed, never refreeze the ampule

Ampules that have been thawed and re-frozen can lose up to 97% of its PFU.
Use paper towels to dry hands and ampules.

Cloth towels can become contaminated.
Merial’s Ampule Breaker
Steps in Vaccine Mixing

1* dye added 5 minutes prior vaccine mixing
Steps in Vaccine Mixing

1. Disinfect the port of diluent bag
2. Take some diluent in a sterile syringe
3. Extract vaccine from ampule with the diluent-filled syringe
4. Gently inject the vaccine into the diluent bag
5. Take some diluent into the syringe again and rinse the ampule
6. Mix the bag gently to distribute the vaccine evenly
By rinsing the **ampules & ampule tips**, additional virus can be recovered.
Marek's Vaccine Titer Percent Difference in rinsed ampoules compared with a non-rinsed control

-6%  
+12.8

Titer

None  Ampule only  Ampule + cap
Complete Process From Point A to Point B as Soon as Possible

Point A
Take vaccine out of liquid nitrogen tank

Point B
Inject the vaccine in the diluent bag
Mix the bag gently to distribute the vaccine evenly.
The prepared vaccine should be used within 1 to 2 hours from the time of preparation.
Place the bag on ice for transport to the *in ovo* machine.
An ice pack should be used to keep the vaccine cool.
Proper disposal of biohazard material is critical.
An alternate mixer should be trained and allowed to mix the vaccine at least once a week.
Vaccine Mixing Checklist

- Hands washed, mixing area cleaned
- Dye or antibiotic added more than 5 minutes before vaccine
- Number of ampules per bag calculated properly
- Vaccine thawed properly

- Hands and ampules dried before opening the ampule
- Vaccine carefully added to bag
- Ampules and tips rinsed
- Bag gently squeezed to evenly mix the vaccine
- Alternate mixer evaluated
Vaccine Administration
Subcutaneous vaccination at one day of age
Subcutaneous vaccination at one day of age
Inspecting Subcutaneous Vaccination

• Conduct routine random checking of vaccinated chicks
• The minimum level of accuracy (properly vaccinated chicks) must be at least 97%
• If less than 97% accurate, the procedures need to be reviewed to detect where the problem is and implement correctives
In ovo Injection
Proper Procedures for *in ovo* Vaccination
Proper maintenance and QC is critical for in ovo vaccination.

Always adhere to scheduled maintenance of your vaccination equipment.

Proper equipment sterilization and cleaning is critical.

Properly maintained equipment will always be more dependable.
Prepare fresh sanitizing solutions daily.
Loading the Eggs
Run The Check Plate After Each Buggy to Make Sure There Are No Plugged Needles
Check Eggs For Contamination
Summary

- Vaccination, in conjunction with the following initiatives, will greatly reduce the incidence of Marek's disease in the field:
  - Good farm cleaning and disinfection
  - Proper reception practices
  - Adequate downtime between flocks
  - Accurate vaccination program
Summary (cntd.)

- Vaccination programs tailored to the type of bird and field situation
- Good vaccine preparation and administration practices
References


References


References