Avian Mycoplasma

Naola Ferguson-Noel, DVM, MAM, PhD
If Georgia were a country, it would be the 7th largest in Broiler Production

(1,000 metric tons forecast for 2014)

<table>
<thead>
<tr>
<th>Country</th>
<th>Metric Tons</th>
</tr>
</thead>
<tbody>
<tr>
<td>United States</td>
<td>17,276</td>
</tr>
<tr>
<td>China</td>
<td>12,700</td>
</tr>
<tr>
<td>Brazil</td>
<td>12,678</td>
</tr>
<tr>
<td>India</td>
<td>3,725</td>
</tr>
<tr>
<td>Russia</td>
<td>3,100</td>
</tr>
<tr>
<td>Mexico</td>
<td>3,060</td>
</tr>
<tr>
<td><strong>Georgia</strong></td>
<td><strong>2,484</strong></td>
</tr>
<tr>
<td>Argentina</td>
<td>2,080</td>
</tr>
<tr>
<td>Turkey</td>
<td>1,810</td>
</tr>
<tr>
<td>Indonesia</td>
<td>1,600</td>
</tr>
</tbody>
</table>

Prepared by: Georgia Poultry Federation  
Source: USDA/FAS  
Updated: July 2014
ON AN AVERAGE DAY
GEORGIA PRODUCES

29.3 MILLION POUNDS OF CHICKEN

6.9 MILLION TABLE EGGS

5.5 MILLION HATCHING EGGS
Poultry -- The Largest Segment of Georgia Agriculture

Percent Total by Commodity

- Crops: 44%
- Poultry: 47%
- Livestock & Dairy: 9%

* Crops include row crops, vegetables, fruits, nuts, nursery/greenhouse, hay and turfgrass

Prepared by: Georgia Poultry Federation
Source: University of Georgia, 2011 Farm Gate Value Report
Updated: July 2014
### Production of poultry meat and eggs, leading nations, 2001

<table>
<thead>
<tr>
<th>Nation</th>
<th>Poultry meat</th>
<th>Chicken meat</th>
<th>Turkey meat</th>
<th>Duck meat</th>
<th>Goose meat</th>
<th>Primary eggs³</th>
</tr>
</thead>
<tbody>
<tr>
<td>WORLD</td>
<td>70,358,813</td>
<td>60,258,645</td>
<td>5,085,889</td>
<td>2,936,687</td>
<td>2,058,969</td>
<td>56,594,078</td>
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<tr>
<td>United States</td>
<td>16,747,600</td>
<td>14,210,000</td>
<td>2,485,000</td>
<td>52,600</td>
<td>Neg.</td>
<td>5,080,000</td>
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<tr>
<td>China</td>
<td>13,286,850</td>
<td>9,401,030</td>
<td>1,990</td>
<td>2,009,980</td>
<td>1,873,850</td>
<td>23,354,520</td>
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<tr>
<td>European Union (15)</td>
<td>8,852,099</td>
<td>6,632,852</td>
<td>1,860,960</td>
<td>343,112</td>
<td>14,075</td>
<td>5,303,441</td>
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<tr>
<td>Brazil</td>
<td>6,394,850</td>
<td>6,222,700</td>
<td>165,000</td>
<td>7,150</td>
<td>Neg.</td>
<td>1,582,700</td>
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<tr>
<td>France</td>
<td>2,077,100</td>
<td>1,100,000</td>
<td>735,000</td>
<td>235,000</td>
<td>6,000</td>
<td>1,047,000</td>
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<tr>
<td>Mexico</td>
<td>1,945,038</td>
<td>1,897,546</td>
<td>27,242</td>
<td>20,250</td>
<td>Neg.</td>
<td>1,881,645</td>
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<tr>
<td>United Kingdom</td>
<td>1,561,700</td>
<td>1,257,500</td>
<td>256,000</td>
<td>45,800</td>
<td>2,400</td>
<td>644,751</td>
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<tr>
<td>Thailand</td>
<td>1,366,500</td>
<td>1,260,000</td>
<td>Neg.</td>
<td>105,000</td>
<td>1,500</td>
<td>810,000</td>
</tr>
<tr>
<td>Japan</td>
<td>1,180,012</td>
<td>1,180,000</td>
<td>12</td>
<td>Neg.</td>
<td>Neg.</td>
<td>2,526,000</td>
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<tr>
<td>Italy</td>
<td>1,156,000</td>
<td>816,000</td>
<td>340,000</td>
<td>Neg.</td>
<td>Neg.</td>
<td>707,000</td>
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<tr>
<td>Canada</td>
<td>1,092,300</td>
<td>943,000</td>
<td>141,000</td>
<td>7,400</td>
<td>900</td>
<td>362,800</td>
</tr>
<tr>
<td>Spain</td>
<td>1,034,000</td>
<td>1,012,000</td>
<td>22,000</td>
<td>Neg.</td>
<td>Neg.</td>
<td>563,700</td>
</tr>
</tbody>
</table>

³Table eggs of all species, including chicken, duck, and goose.

Neg.: Negligible production
http://poultryhealthtoday.com/poultry-veterinarians-the-latest-focus-of-popular-youtube-series/?utm_source=PHT+eBlast+List&utm_campaign=ab73f81402-Samantha_Pohl_announcement8_20_2015&utm_medium=email&utm_term=0_5ac605299a-ab73f81402-261557785

https://www.farmaid.org/issues/industrial-agriculture/a-look-at-the-poultry-industry-how-does-chicken-get-on-your-plate/
Pedigrees

GGPs

GPs

Parents/Breeders

Broilers

1 ♂ + 10 ♀

3.5 – 5 years

28,000,000 broiler progeny
Output of a Pedigree Family

- Pedigree family = 1♂ + 10♀
- Pedigree to broiler = 3.5 - 5 years
- Up to 28,000,000 broiler progeny
- (28KK) x (2.0 Kg) x (69% yield):
  - 39,744 T of poultry meat
INTEGRATED BROILER COMPANY

Primary Breeder

Grain Procurement

Storage

Feed milling

Delivery

Chick Delivery

Hatching eggs

Breeders

Hatchery

Broilers

Process Plant

Further Process

Packaging

Transport

Retail

Chick Delivery

Cornish hen

Small bird

Roaster

Process Plant

Further Process

Packaging

Transport

Retail

Blood/Feather meal

Poultry meal

Poultry fat

Rendering

Waste water management
Structure of a Broiler Integration

Integrated Broiler Company

- Feed Mill (Complex 1)
  - Broiler Breeders
  - Hatchery
  - Broiler production (1 – 2 million broilers/week)
  - Processing Plant

- Feed Mill (Complex 2)
  - Broiler Breeders
  - Hatchery
  - Broiler production (1 – 2 million broilers/week)
  - Processing Plant

- Feed Mill (Complex 3)
  - Broiler Breeders
  - Hatchery
  - Broiler production (1 – 2 million broilers/week)
  - Processing Plant
Mycoplasma gallisepticum and 
M. synoviae

- Bacteria
  - Mollicutes
    - No cell wall
    - Smallest free-living organisms
    - Smallest genome of any free-living organism

- pathogenic and economically significant avian mycoplasma
Why is Control Important?

- Clinical disease – pathogenic strains
The World Organisation for Animal Health (OIE)

- OIE-Listed diseases
- Manual of Diagnostic Tests and Vaccines for Terrestrial Animals
NPIP

- National Poultry Improvement Plan (NPIP)
  - cooperative Federal-State-Industry program
  - use new diagnostic technology to effectively improve poultry and poultry products
  - egg-transmitted, hatchery-disseminated poultry diseases
  - provides certification that poultry and poultry products are disease free
- Pullorum-Typhoid (P-T)
- Mycoplasma gallisepticum (MG)
- Mycoplasma Synoviae
- Mycoplasma Meleagridis
- Salmonella Enteritidis
- Avian Influenza (AI)
NPIP

The official tests for *M. gallisepticum*, *M. meleagrisidis*, and *M. synoviae* shall be

- the serum plate agglutination test,
- the tube agglutination test,
- the hemagglutination inhibition (HI) test,
- the microhemagglutination inhibition test,
- the enzyme-linked immunosorbent assay (ELISA) test,
- a polymerase chain reaction (PCR)-based test, or a combination of two or more of these tests.

The HI test or the microhemagglutination inhibition test shall be used to confirm the positive results of other serological tests. HI titers of 1:40 or more may be interpreted as suspicious, and final judgment must be based on further samplings and/or culture of reactors.
Avian Mycoplasma Diagnosis

- Serology - SPA, HI and ELISA
- PCR – conventional and real-time
- Culture
- Bioassay
Gene Targeted Sequencing (GTS)

- Compare to database
  - >200 sequences
- Assign type
  - WT#BB/WT#4
  - S-1, S-2, S-3
- No culture required
- Good reproducibility
- Reference database
- Can be combined with diagnostic PCR
Approaches to Control

- Keep it out
  - Surveillance
  - Eradication

- Live with it
  - Medication
  - Vaccines
Mycoplasma Surveillance Tests

- Sensitive
  - early detection
- High throughput
- Economical
- Tolerate some false positives
- SPA, ELISA, (real-time PCR)
Sources of Infection

- Egg transmission
  - Rate of transmission unpredictable
Vertical transmission

"F" STRAIN

Fig. 1. Egg transmission of F-strain-vaccinated chickens (black bar) and unvaccinated controls (white bar).

Sources of Infection

- **Egg transmission**
  - Rate of transmission unpredictable

- **Horizontal transmission**
  - Direct or indirect contact with infected birds
  - Biological carriers
  - Mechanical carriers – dust, droplets, feathers, equipment, rodents
  - Aerosol transmission possible over short distances
Mycoplasma on GA farms: Prevention and Containment

Mycoplasma infections (MG or MS) are important because positive flocks can transmit the organism to the progeny (usually broilers), possibly making them sick. Mycoplasma positive breeder flocks may also show signs of disease, affecting their productivity; hatching eggs exports are disturbed. The flocks lose their "clean" NPIP classification and may have to be sold early.

My birds tested positive: how did they get it?

THE MOST COMMON WAY TO GET MYCOPLASMA IN THE FIELD: IT WALKS IN, USUALLY ON TWO LEGS!
DIRECT CONTACT OF THE FARM OR GROWER with:
Persons, vehicles, equipment that have been in contact with non-commercial poultry, commercial layers, positive farms;
hired help, contractors, crews, family members, other visitors are by far the most common means of infecting a flock!

A FARM CAN ALSO GET MYCOPLASMA FROM MIGRATION (less likely) from a Mycoplasma positive source or farm through rodents or insects.

A FARM CAN ALSO GET CONTAMINATED THROUGH THE AIR or wild birds (least likely) MG and MS are sensitive to heat. They do not survive well outside the birds. However, they will withstand freezing. MG may also come from wild birds (ex. house finches).

How do I keep Mycoplasma from spreading from my farm to others?

• Do not lend equipment out
• Practice enhanced rodent, insect control before load out.
• After load out, make sure all birds are disposed of promptly and properly.
  • Keep house empty for a week before removing the litter; Wash and disinfect houses.
  (Mycoplasma are sensitive to disinfectants).
• In GA, MG infected birds have to be treated before transportation to decrease shed.

MOST IMPORTANTLY: How do I keep my birds CLEAN in the first place?

• Avoid contact with other birds
• Dispose of all dead birds promptly and properly
• Make sure your visitors and hired help have not been in contact with high risk poultry; No pets in houses
• Use dedicated equipment and vehicles on the farm
• Practice entry biosecurity at all times (no exceptions): Shower (on some farms), change of clothing or disposable coveralls, use dedicated footwear or disposable boots and hairnets, use foot pans. THIS IS VERY EFFECTIVE IN PREVENTING ENTRY!
• Bird proof houses, practice rodent and insect control

KEEP MYCOPLASMA OUT!!!
# Survival of MG on Various Substances

<table>
<thead>
<tr>
<th>Material</th>
<th>Days</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cotton</td>
<td>4</td>
</tr>
<tr>
<td>Rubber</td>
<td>2</td>
</tr>
<tr>
<td>Straw</td>
<td>2</td>
</tr>
<tr>
<td>Shavings</td>
<td>8 hours</td>
</tr>
<tr>
<td>Wood</td>
<td>1 day</td>
</tr>
<tr>
<td>Feed</td>
<td>4 hours</td>
</tr>
<tr>
<td>Feathers</td>
<td>4</td>
</tr>
<tr>
<td>Hair</td>
<td>3 days</td>
</tr>
<tr>
<td>Ear</td>
<td>4 hours</td>
</tr>
<tr>
<td>Nose</td>
<td>1 day</td>
</tr>
<tr>
<td>Skin</td>
<td>&lt; 4 hours</td>
</tr>
<tr>
<td>Buffer</td>
<td>1 day</td>
</tr>
</tbody>
</table>

Antibiotics

- Will not eliminate infection
- Antibiotic resistance may develop
<table>
<thead>
<tr>
<th>Genotype</th>
<th>Date Isolated</th>
<th>Company</th>
<th>MIC tylosin (µg/mL)</th>
<th>Sensitivity to tylosin</th>
<th>MIC tetracycline (µg/mL)</th>
<th>Sensitivity to tetracycline</th>
</tr>
</thead>
<tbody>
<tr>
<td>S-56</td>
<td>Jun-12</td>
<td>1</td>
<td>1</td>
<td>S</td>
<td>0.5</td>
<td>S</td>
</tr>
<tr>
<td>S-56</td>
<td>Jun-12</td>
<td>1</td>
<td>1</td>
<td>S</td>
<td>0.5</td>
<td>S</td>
</tr>
<tr>
<td>S-48</td>
<td>Feb-13</td>
<td>2</td>
<td>0.0625</td>
<td>I</td>
<td>0.25</td>
<td>S</td>
</tr>
<tr>
<td>S-56</td>
<td>May-14</td>
<td>5</td>
<td>2</td>
<td>S</td>
<td>0.25</td>
<td>S</td>
</tr>
<tr>
<td>S-56</td>
<td>May-14</td>
<td>5</td>
<td>1</td>
<td>S</td>
<td>0.25</td>
<td>S</td>
</tr>
<tr>
<td>S-56</td>
<td>May-14</td>
<td>5</td>
<td>1</td>
<td>S</td>
<td>0.25</td>
<td>S</td>
</tr>
<tr>
<td>S-56</td>
<td>Jun-14</td>
<td>5</td>
<td>1</td>
<td>S</td>
<td>0.25</td>
<td>S</td>
</tr>
<tr>
<td>S-56</td>
<td>Jun-14</td>
<td>4</td>
<td>1</td>
<td>S</td>
<td>0.5</td>
<td>S</td>
</tr>
<tr>
<td>S-56</td>
<td>Jun-14</td>
<td>3</td>
<td>1</td>
<td>S</td>
<td>0.25</td>
<td>S</td>
</tr>
<tr>
<td>S-56</td>
<td>Jun-14</td>
<td>3</td>
<td>1</td>
<td>S</td>
<td>0.25</td>
<td>S</td>
</tr>
<tr>
<td>S-56</td>
<td>Jun-14</td>
<td>5</td>
<td>1</td>
<td>S</td>
<td>0.25</td>
<td>S</td>
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<tr>
<td>S-56</td>
<td>Jun-14</td>
<td>5</td>
<td>4</td>
<td>R</td>
<td>0.375</td>
<td>S</td>
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<tr>
<td>S-56</td>
<td>Jun-14</td>
<td>5</td>
<td>1</td>
<td>S</td>
<td>0.25</td>
<td>S</td>
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<tr>
<td>S-56</td>
<td>Oct-14</td>
<td>5</td>
<td>1</td>
<td>S</td>
<td>0.25</td>
<td>S</td>
</tr>
<tr>
<td>S-56</td>
<td>Feb-15</td>
<td>2 (Farm A)</td>
<td>2</td>
<td>I</td>
<td>0.1</td>
<td>S</td>
</tr>
<tr>
<td>S-56</td>
<td>Feb-15</td>
<td>2 (Farm B)</td>
<td>1</td>
<td>I</td>
<td>0.1</td>
<td>S</td>
</tr>
<tr>
<td>S-56</td>
<td>Feb-15</td>
<td>2 (Farm C)</td>
<td>1</td>
<td>S</td>
<td>0.1</td>
<td>S</td>
</tr>
<tr>
<td>S-56</td>
<td>Mar-15</td>
<td>2 (Farm D)</td>
<td>2</td>
<td>S</td>
<td>0.5</td>
<td>S</td>
</tr>
<tr>
<td>S-56</td>
<td>Mar-15</td>
<td>2 (Farm E)</td>
<td>4</td>
<td>I</td>
<td>0.2</td>
<td>S</td>
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<tr>
<td>S-56</td>
<td>Mar-15</td>
<td>2 (Farm F)</td>
<td>2</td>
<td>R</td>
<td>0.5</td>
<td>S</td>
</tr>
</tbody>
</table>

S ≤ 1 / I = 2 / R > 4  
S ≤ 4 / I = 8 / R > 16

X. Gong & Ferguson-Noel, N., Unpublished (manuscript in preparation)
Tylosin Treatments Less Effective Over Time

A. Kiers & Ferguson-Noel, N., Unpublished (manuscript in preparation)
Vaccines

• Inactivated oil-emulsion bacterins
• Recombinant MG Vaccine
• Live vaccines
  • F Strain
  • ts-11
  • 6/85
Reasons to Vaccinate

- Prevent clinical disease
- Reduce egg transmission
- Avoid cost of medication
- Eradicate virulent field strains
Displacement

- Depends on ability of vaccine to prevent secondary infection with wild type
Displacement - R-strain experiment

Copy No. Log10

0 WPC  2 WPC  4 WPC  8 WPC

None
6/85
ts-11
F-strain
K-strain
MG – GA Broiler-type

![Bar chart showing MG - GA Broiler-type from 2006 to 2010. The chart compares the years 2006, 2007, 2008, 2009, and 2010. The categories 'ts-11/ts-11' and 'ts-11/HF' are represented by different colors. The chart indicates a significant increase in MG in 2008.](#)
Genotyping South African MG Isolates

Genotyping South African MG Isolates

MG epidemiology in Jordan

- Relatively large and developing chicken industry
- No national plan to control MG infection
- Farmers depend on vaccination and/or chemotherapy
- 70% of broiler flocks that suffered from respiratory disease were positive for MG by ELISA

Ferguson WVPA 2015
MG epidemiology in Jordan

• 2004-2005 vs 2007-2008 (n=24)

• None of the flocks were vaccinated

MG epidemiology in Jordan

- 2004-2005 vs 2007-2008 (n=24)
- None of the flocks were vaccinated
- 21 isolates indistinguishable from F-strain

MG epidemiology in Jordan

- 2004-2005 vs 2007-2008 (n=24)
- None of the flocks were vaccinated
- 21 isolates indistinguishable from F-strain
- The most common vaccine used in Jordan in the early 2000’s was F-strain
- In later years, F-strain availability was very limited

Jordan F-Strain – Increased Virulence in Broilers

Air Sac Lesion Scores

- **F-strain**
- **K6330-7**
- **R strain**
- **None**

![Bar graph showing air sac lesion scores for different strains at 10 and 28 days post challenge (dpc).](image-url)
MG Outbreak in NE GA and ts-11 Vaccination

Outbreak begins

Vaccination begins

1st Incident - A

2nd - A

3rd - B

4th - C

5th - D

6th - D

Nov. 2006

Summer 2007

July 2008

Jan. 2009

Oct. 2009

Jan. 2010

Feb. 2010

April 2010

FergusonWVPA2015
Increased Virulence and Vertical Transmission

- **Pathogenicity Trial**

- **Vertical Transmission Trial**

MG Types

Thank you

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