

NPIP 44th Biennial Conference June 26-28, 2018 Franklin, TN

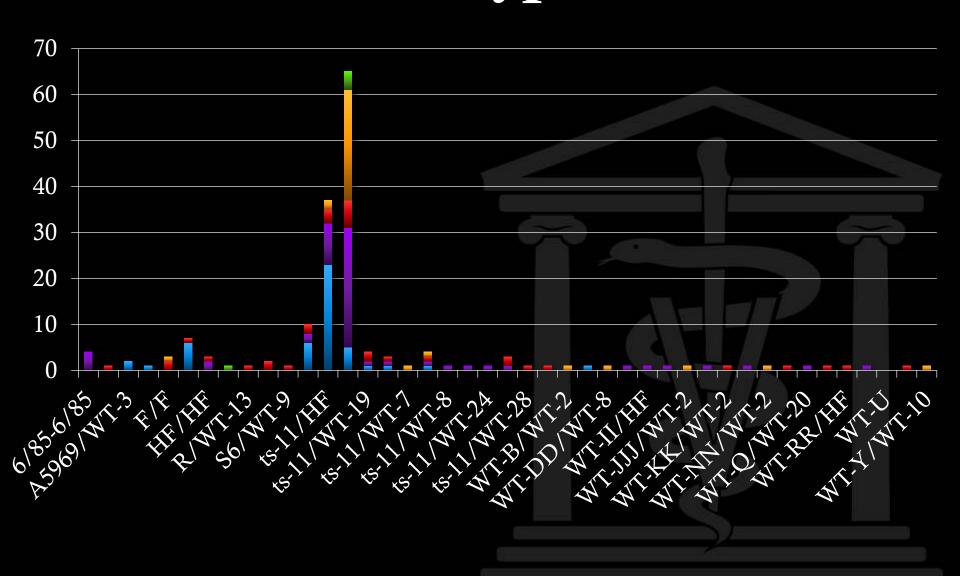
Naola Ferguson-Noel, DVM, MAM, PhD

# **Current Picture**

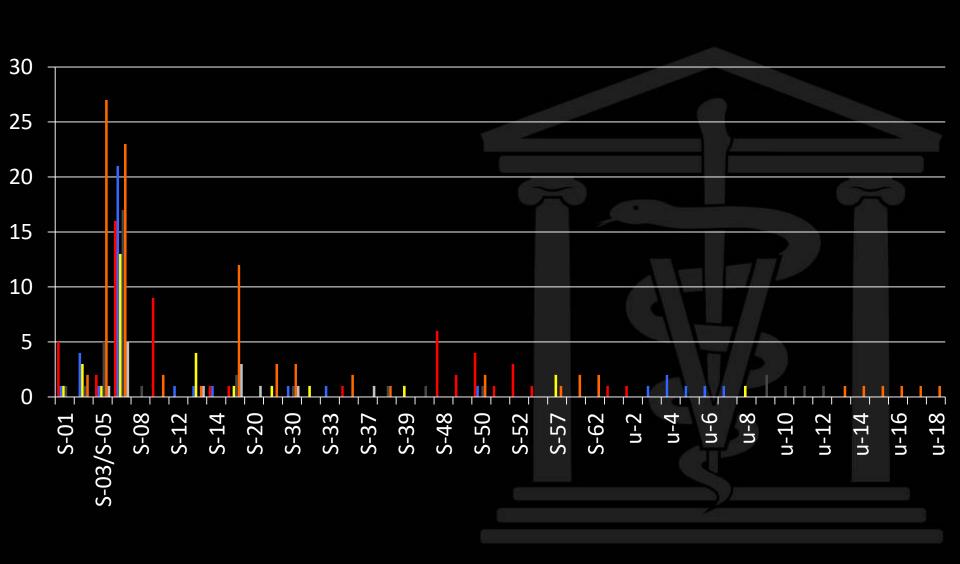
- MG
  - Low prevalence
  - High significance
- MS
  - Higher prevalence
  - Lower significance



# MG Types



# MS Sequence Types



# No. of positive submissions (no vaccinated flocks)



### **Check Tests**

 Panel of convalescent chicken sera against *M. synoviae* and *M. gallisepticum*

 Panel of tracheal swabs and purified DNA from M. synoviae and M. gallisepticum infected chickens

## **Antisera Check Tests**

- 85 kits (29 Known, 56 Blind)
  - -10 MG+
  - 10 MS+
  - 10 negative

Pass = Score of >80%

### **Antisera Check Tests Results**

#### Avian Mycoplasma PDRC Check Tests 2017 Results

Lab Name: Poultry Laboratory

Invoice No. 2003

1) SEROLOGY (Blind Kit): **PASS** Score = 97%

ELISA – BioChek MGMS Combo (Score = 93%) HI – NVSL (Score = 100%)

#### Comments:

Two MS+ samples (B14 and B19) were false negative on ELISA (MS HI positive). Some other labs got similar results on ELISA with these samples.

Summary of All Participants (n=85):

Range of Scores = 87-100%

# PASS = 85

# FAIL ( $\leq 80\%$ ) = 0

### **Antisera Check Tests Results**

Range of Scores = 87-100%

$$\# PASS = 85 (100\%)$$

# FAIL (
$$< 80\%$$
) = 0

# **PCR Check Tests**

- 67 Kits
  - -49 "Blind"
  - 18 "Known"

Pass = Score of >80%

# **PCR Check Tests**

- 5 swabs pools (5 swabs each)
  - 1 negative
  - 1 MG+
  - 1 MS+
  - 1 MG and MS+ (1 weak)
- 5 DNA samples
  - 1 negative
  - 1 MG+
  - 1 MS+
  - 1 MG and MS+ (1 weak)

# PCR Check Tests

• Strong positive Ct = 20 - 27

Weak positive Ct = 30 - 35

## PCR Check Tests Results

### Swabs:

Range of Scores = 80 - 100%

# PASS = 66 (99%)

# FAIL = 1

## **PCR Check Tests Results**

### DNA:

Range of Scores = 70 - 100%

# PASS = 64 (96%)

# FAIL = 3

## **PCR Check Test Results**

#### Avian Mycoplasma PDRC Check Tests 2017 Results

Lab Name: Vet Diagnostic Lab

Invoice No. 2003

2) PCR (Blind Kit):

Swabs: FAIL Score = 80%

DNA: PASS Score = 90%

Blind Panel	Expected Results		% Participants with correct result (Mean Ct)	
Swab label	MG	MS	MG	MS
17-06	Negative	Negative	100% ( - )	100% ( - )
17-07	Negative	Positive	100% ( - )	100% (33.3)
17-08	Positive	Negative	100% (29.2)	99% (-)
17-09	Positive	Positive	100% (30.2)	99% (31.0)
17-10	Negative	Weak Positive	99% ( - )	96% (32.5)
DNA tube				
17-06	Negative	Negative	100% (-)	100% ( - )
17-07	Negative	Positive	96% ( - )	99% (24.7)
17-08	Positive	Negative	93% (23.0)	100% ( - )
17-09	Positive	Positive	99% (28.8)	97% (26.0)
17-10	Negative	Weak Positive	99% (-)	99% (31.0)

Positive expected Ct = 21 - 35 Weak Positive expected Ct = >28

#### Comments:

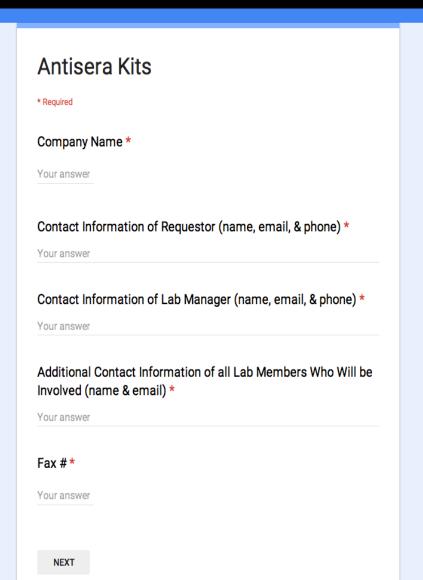
- Incorrect results in submission highlighted
- Two MS+ swab pools (17-09 and 17-10) were false negative on MS PCR.
- MS Ct value from swab (17-07) was higher than other labs (39 vs. 33.3 average)
- One DNA sample (17-07) was false positive on MG PCR.

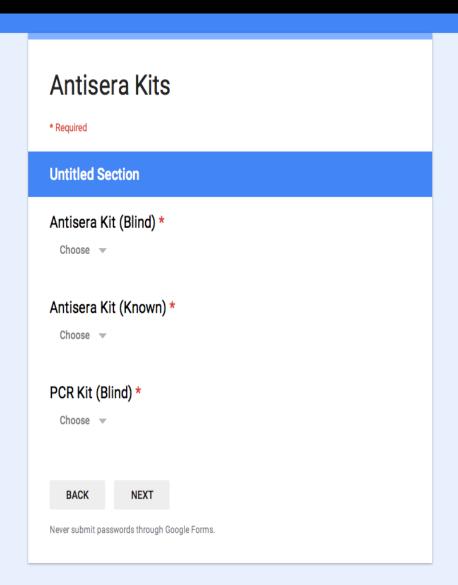
#### Summary of All Participants:

Swabs (n= 67): Range of Scores = 80-100% # PASS = 66 # FAIL (≤ 80%) = 1 DNA (n= 67): Range of Scores = 70-100% # PASS = 64 # FAIL (\$ 80%) = 3



# Next panel –September 2018

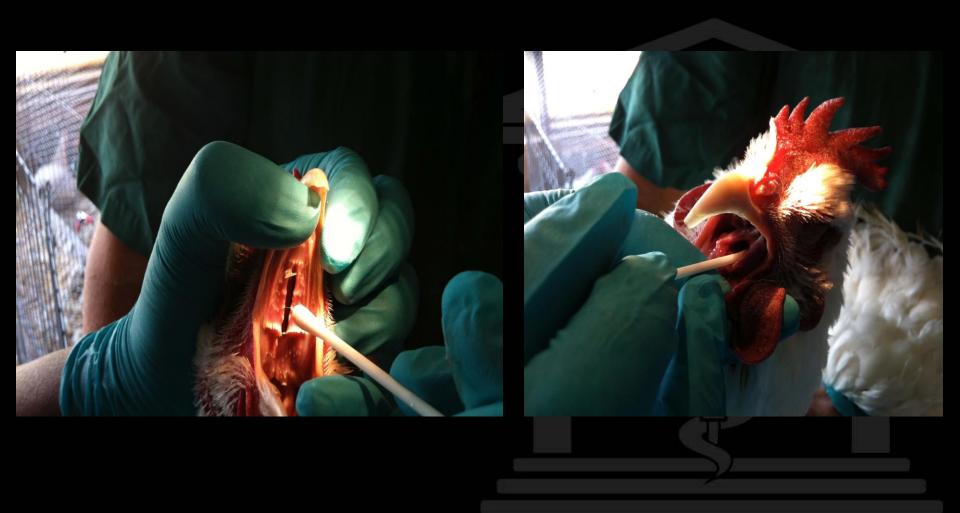




### **PCR**

- Swab material
- Moistened vs Dry swabs
- Temperature and time of storage
- Transport/Prep media

# **Swabbing for PCR**



# Tracheal



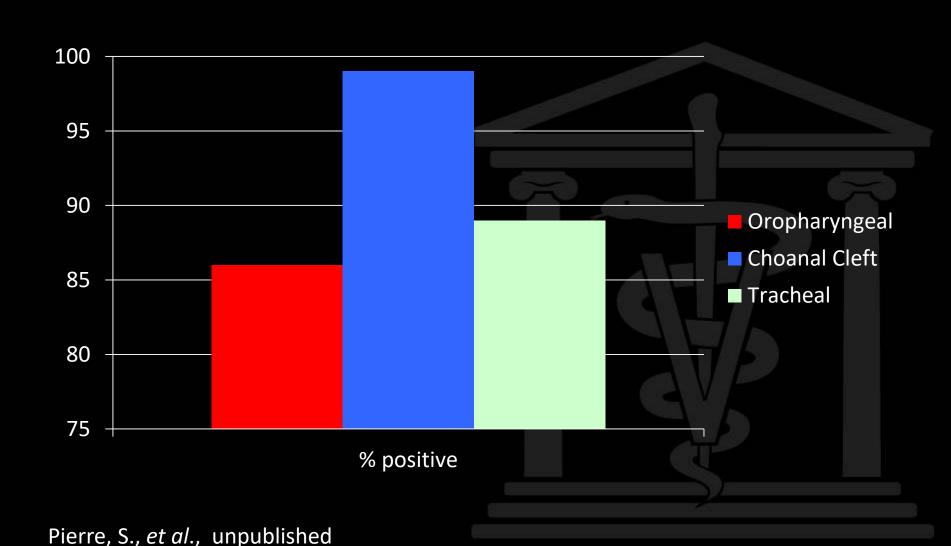
# Choanal



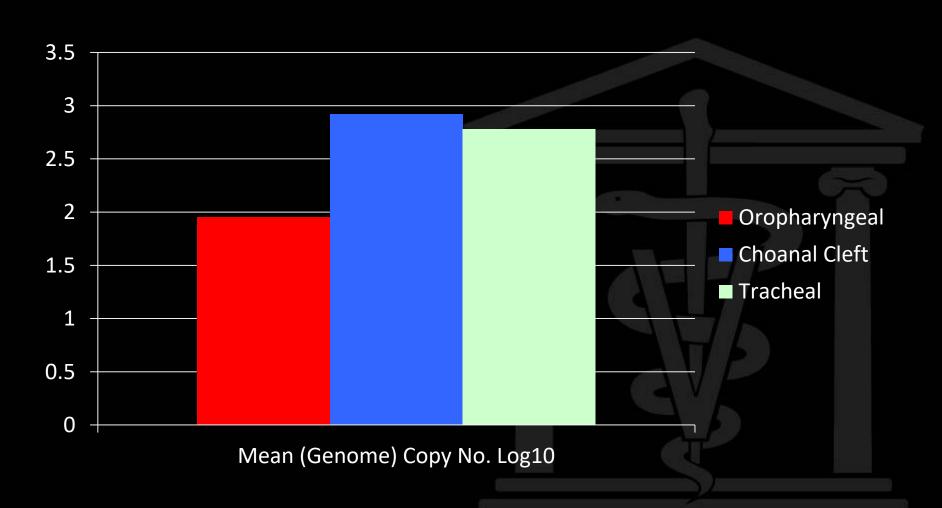
# Oropharyngeal



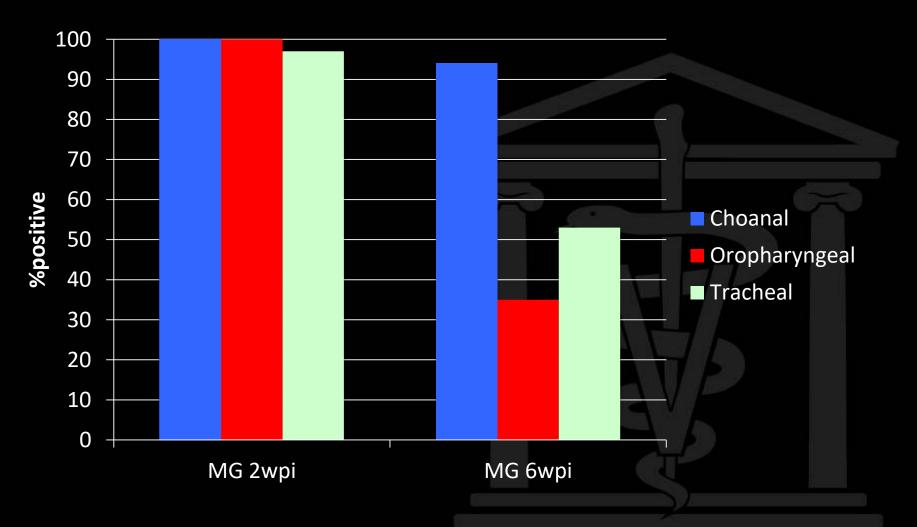
# Swabbing for MG and MS Detection by Real-Time PCR



# Swabbing for MG and MS Detection by Real-Time PCR



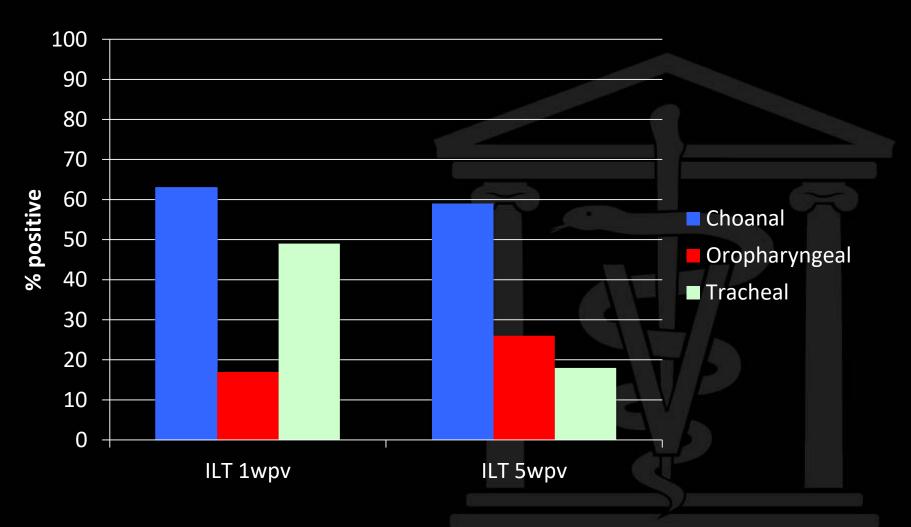
# **Swabbing for MG Detection by Real-Time PCR**



Jude, R., et al., unpublished

This project was supported by Agriculture and Food Research Initiative Competitive Grant no. (2015-68004-23131) from the USDA National Institute of Food and Agriculture.

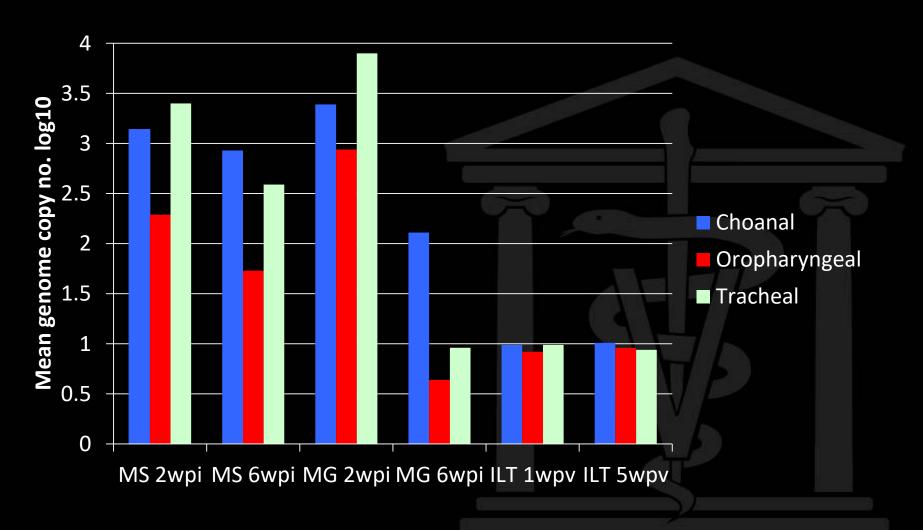
# **Swabbing for ILT Detection by Real-Time PCR**



Jude, R., et al., unpublished

This project was supported by Agriculture and Food Research Initiative Competitive Grant no. (2015-68004-23131) from the USDA National Institute of Food and Agriculture.

# Swabbing for MG, MS & ILT Detection by Real-Time PCR



# Thank you

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