Coccidiosis in captive northern bobwhites

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Captive rearing of quail is big business

- Birds raised in captive facilities → released as adults
  - 20-30 million/yr in U.S.

- High bird density results in efficient pathogen transmission
  - Blackhead (*Histomonas*)
  - Ulcerative colitis (*Clostridium colini*)
  - Coccidiosis (*Eimeria* spp.)
What are coccidia and why do they cause disease?

- Microscopic protozoal parasites
- Environmentally resistant stage
- Intestinal cells infected in host
  - Very prolific replication
    - 1 ingested oocyst can produce > 50,000 oocysts/bird
- Can cause large mortality and morbidity events
Three previously described *Eimeria* species in bobwhites

- *Eimeria dispersa*
  - Able to infect wild turkeys, ruffed grouse & quail

- *Eimeria colini*

- *Eimeria lettyae*
  - Most pathogenic (Ruff and Wilkins, 1987)
Our research questions

1) What is the prevalence and geographical distribution of coccidia spp. from captive bobwhite farms?

2) Are there drug resistant strains of coccidia from farms using anticoccidials to treat and prevent coccidiosis?
Captive bobwhite litter samples originated from 12 states

- 31 samples collected from 12 states
- Flock age: 2 weeks → adult
- Propagated in bobwhites → Xenodiagnosis
Construction of PCR specific primers to differentiate bobwhite *Eimeria* spp.

- Use genus wide *Eimeria* spp. primers BSEF and BSER (Schnitzler *et al.*, 1999. Avian Pathology)
  - Amplified (ITS-1) region of rRNA
    - ITS region are non-coding
    - Useful for species differentiation and intraspecies phylogenetic analysis (Su *et al.*, 2003. Veterinary Parasitology)
Species specific primers developed and tested for specificity

• Construct primers to nucleotide sequences conserved among *Eimeria* spp. within group, but different than other groups

• Forward and reverse primers constructed – similar Tm

• C or G nucleotides at 3’ ends

• Avoided hairpins & dimers

NOBW02 C1  AATTATAAATTGTGTGTATTGTCACACCCATGGAGCAAACCGTA
NOBW02 C5  ....TAA....T.CG..-------------..AGA...A........................-T...CG........
NOBW03 C3  ...GCCCATTCAACGTTTTCACG. ........G........------A.........T...G......
NOBW05 C3  ...GCCCATTCAACGTTTTCACG. ........G........------A.........T...G......
NOBW06 C1  ...GCCCATTCAACGTTTTCACG. ........G........------A.........T...G......
NOBW02 C5  ...GCCCATTCAACGTTTTCACG. ........G........------A.........T...G......
Developed PCR primers were specific to corresponding plasmid insert for each group

100 bp ladder  Primer group 1  Primer group 2  Primer group 3

Amplicons: 280-320 bp
Annealing temp: 45-48 C
All farms contained at least one species of coccidia

Frequency of detection of various *Eimeria* spp.

<table>
<thead>
<tr>
<th>Species</th>
<th>Number of samples</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Eimeria colini</em></td>
<td>29</td>
</tr>
<tr>
<td><em>Eimeria dispersa</em></td>
<td>22</td>
</tr>
<tr>
<td><em>Eimeria lettyae</em></td>
<td>20</td>
</tr>
</tbody>
</table>
Majority of farms had at least two *Eimeria* spp. present.

Frequency of number of *Eimeria* spp. detected from samples

<table>
<thead>
<tr>
<th>Number of <em>Eimeria</em> spp.</th>
<th>Number of samples</th>
</tr>
</thead>
<tbody>
<tr>
<td>3</td>
<td>13</td>
</tr>
<tr>
<td>2</td>
<td>14</td>
</tr>
<tr>
<td>1</td>
<td>4</td>
</tr>
</tbody>
</table>
Discussion: survey results

• First known survey of captive bobwhite farms
  – 100% of farms contained coccidia
  – 27 (87.1%) samples had at least two species
  – No associations with geographical location or flock age

• Research needed to understand the effects on wild quail
The efficacy of anticoccidial products against *Eimeria* spp. in northern bobwhites
Examination of resistance in field isolates

- Anticoccidial trials performed similar to those of domestic poultry
  - 1 pen 10 birds each
  - 6 to 10 field isolates used per compound
    - Monensin (90 ppm)
    - Salinomycin (55 ppm)
- Used percent weight gain of birds as index of drug efficacy
  - Compared to uninfected controls
Resistance to monensin and salinomycin observed in multiple isolates

![Percent Gain Monensin](chart1)

![Percent Gain Salinomycin](chart2)
Summary of anticoccidial study

• At least half of the tested isolates were resistant to at least one of the anticoccidial compounds

• Demonstrates that captive quail farming is selecting for resistant strains of bobwhite coccidia
  – Potential contamination of environment with resistant strains
How does this research relate to conservation

• Propagation of captive bobwhites are frequently seen as a “fix” for declining wild quail populations
  – Leads to less emphasis on habitat conservation

• Our research demonstrates the significant disease ramifications of captive quail propagation
  – Use information in our efforts to promote habitat conservation for quail restoration
Acknowledgements

• Larry McDougald
• Buffy Howerth
• Robert Beckstead
• Michael Yabsely
• Numerous wildlife biologists