Using Rapid Culture Systems to Make Pathogen Based Mastitis Treatment Decisions on Farms

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Outline

- Selective treatment of clinical mastitis cases
  - Rational for targeted antimicrobial therapy
  - Application and accuracy of on-farm culture systems to guide clinical mastitis treatment decisions
  - Study results: using rapid culture systems to selectively treat clinical mastitis cases
Mastitis remains a challenge...

- Mastitis continues to be the most costly infectious disease on dairies
  (Riekerink et al., 2007; Sargeant et al., 1998; Erskine et al., 1988; Fetrow et al., 2010; Erskine et al., 2003)

- Majority of antibiotic use on dairies related to mastitis prevention or treatment
  (Pol and Ruegg, 2007)

- Goal: promote judicious/prudent drug use while maximizing treatment success:
  - Use antibiotics only when necessary
  - Properly target therapy based on bacterial cause
Strategic decision-making

- Need to make strategic decisions to maintain excellent milk quality and high production, reduce antibiotic use and risk of residue

Diagram:

- Find infected cows
  - Treat
    - Don’t treat
      - Segregate
      - Dry off the quarter
      - Cull
  - Lactational treatment
  - Dry treatment
History + SCC + Culture

- Best practice = make individual cow treatment or management decisions based on cow history and culture results

- Some cases of mastitis do not require antibiotic therapy in order to cure
- Some cases of mastitis will not cure regardless
- Goal: reserve antibiotics for clinical mastitis cases that would benefit from treatment
## Cause of Clinical Mastitis in Lactating Cows

<table>
<thead>
<tr>
<th>References</th>
<th>Etiology (% of samples)</th>
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### References
- Riekerink et al., 2007
- Sargeant et al., 1998
- Erskine et al., 1988

### Etiology
- **NG**: No growth (9-40%)
- **Gram-Pos**: 15 – 70%
- **Gram-Neg**: 8 – 44%
## Cause of Clinical Mastitis in Lactating Cows

<table>
<thead>
<tr>
<th>References</th>
<th>NG</th>
<th>CNS</th>
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**No growth** (9.40%) **Gram-Pos** (15 – 70%) **Gram-Neg** (8 – 44%)

Do all mastitis cases require treatment with intramammary antibiotics?
Culture Negative Results

- “No Growths” = 20 – 40% of milk from clinical cases will not grow bacteria
  
  ✓ False negatives (rare):
  
  ◆ Not sensitive enough to detect at low shedding?
  ◆ Bacteria present does not grow in culture media or incubation environment (e.g. *Mycoplasma*)

  ✓ True negatives (most):
  
  ◆ By the time the sample was taken the cow had already cleared the bacteria (possible/likely for *E. coli*)
  ◆ Not a real infection

- No intramammary treatment
Gram-negative Infections

- To treat or not to treat?
  - High spontaneous cure rate for mild/moderate Gram-negative mastitis
    (Guterbock et al., 1993; Morin et al., 1998; Wilson et al., 1999)
  - vs
  - Some studies show higher bacterial cure rates with treatment, or with extended duration of treatment, but no difference in clinical cure rates or early lactation performance
    (Schukken et al., JDSci 2011. 94; Schukken et al., JDSci 2013. 96)

- Factors to consider
  - E. coli vs. Klebsiella?
    - E. coli expect up to 90-95% spontaneous cure rate (Schukken et al., 2004; Döpfer et al., 1999)
    - Klebsiella expect 35-40% spontaneous cure (Pinzón-Sánchez et al., 2011)
  - Any indication of chronicity?

- Suggestion:
  - No intramammary treatment for first cases of Gram-negative infections
  - Consider treating repeated cases
Gram-positive Infections

- Generally treat with intramammary antibiotics:
  - Coagulase negative Staph (CNS)
  - Non-ag Streps (Environmental Streps. E.g. *Streptococcus uberis*)
  - *Streptococcus agalactiae*

- *Staphylococcus aureus*?
  - Low cure rates (20-30%)
  - Degree of tissue invasion, some antibiotic resistance
  - Consider other factors
    (e.g. SCC and mastitis history)
  - Consider other options if chronic
    *(Pinzón-Sánchez et al., 2011)*
Other pathogens

- Mycoplasma, Prototheca, and yeast do not respond to intramammary antibiotics.

*Knowledge of the pathogen is important for ensuring appropriate and targeted use of intramammary antibiotics.*
Culture data

- Gram-negatives + No-Growths = 50-80% of cases - may not need intramammary antibiotics (Roberson, 2003)

- If we only treat Gram-positive infections, we can reduce intramammary antibiotic use by ≥ 50%

- Highly variable between dairies and across seasons
How do we get culture data?

1. Commercial Diagnostic Laboratory (university or other)

2. On-farm or Vet Clinic culture
Diagnostic Laboratory Culture

- Accurate, reliable identification of any bacterial species:
  - Use culture plus biochemical or molecular tests to confirm exact bacterial species
  - Can identify non-bacterial organisms: Yeast, Prototheca

- Disadvantages:
  - Delay to obtain results
  - Cost
  - Inconvenience of shipping samples
On-Farm Milk Culture (OFC)  
(or local vet clinic)

- Aseptically collected milk samples are plated, incubated and interpreted on-farm

- Selective culture media is used to help identify bacteria without aid of additional testing
On-Farm Culture

- Fast, inexpensive way to categorize the cause of mastitis into treatment categories, for example:
  - No-Growth = No Treat
  - Gram-negative = No Treat (with exceptions)
  - Gram-positive = Treat (with exceptions)

- Limitations – must have time, space, personnel, training
Examples of On-Farm Culture Systems

- 3M Petrifilm System

Rapid Staph aureus plate

Coliform Count plate
Examples of On-Farm Culture Systems

- MN Easy™ Culture System (University of Minnesota)

Bi-Plate, $1.80

Tri-Plate, $3.00
Growth on Factor Media (bright red) = Gram-positive bacteria
   - Ex) Staph or Strep species, Bacillus, Corynebacteria, etc.

Growth on MacConkey Media (pink, translucent) = Gram-negative bacteria
   - Ex) E.coli, Klebsiella, Enterobacter, Serratia, etc.
Growth on **MTKT** Media (dark red) = Strep or Strep-like bacteria
- Ex) Streptococcus, Aerococcus, Enterococcus, etc.

Growth on **MacConkey** Media (pink, translucent) = Gram-negative bacteria
- Ex) E.coli, Klebsiella, Enterobacter, Serratia, etc.

Growth on **Factor** Media (bright red) = Gram-positive bacteria
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Outline

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Example: Targeted therapy based on bacterial cause of mastitis

Detect clinical mastitis

On-farm Culture
Example: Targeted therapy based on bacterial cause of mastitis

On-farm Culture result

No Growth

- NO Intramammary antibiotic tx
- Supportive care and systemic antibiotics if systemic signs
Example: Targeted therapy based on bacterial cause of mastitis

No Growth

- NO Intramammary antibiotic tx
- Supportive care and systemic antibiotics if systemic signs

On-farm Culture result

Gram-negative
- *E. coli*
- *Klebsiella*
Example: Targeted therapy based on bacterial cause of mastitis

- **No Growth**
  - NO Intramammary antibiotic tx
  - Supportive care and systemic antibiotics if systemic signs

- **Gram-negative**
  - *E. coli*
  - *Klebsiella*

- **Gram-positive**
  - *Strep.*, CNS
  - *Staph aureus*

On-farm Culture result
How accurate is OFC?

Few tests are 100% accurate!
Accuracy is good if interpreted at general levels (Gram positive, Gram-negative, No growth)

No growth

Gram-negative

Gram-positive

80-84% accurate if you are trying to identify Gram-positive infections for the purpose of treating with intramammary antibiotics

(Royster et al., JDS 2014)
Accuracy is also OK if using Tri-plate to identify Staphs vs Streps, and *Staph aureus*

- Environmental Strep.
- Staph Species (CNS)
- *Staph aureus*

* Verify with secondary test before culling
  (moderate Positive Predictive Value = 55-80%)
  (Royster et al., JDS 2014)
Test performance is not good at species levels: Don’t try to speciate organisms

Is this *E. coli*, or *Klebsiella* spp.? ❌
It is a Gram-negative

Is this *Strep. uberis*, or *Strep. dysgalactiae*? ❌
It is a Gram-positive and a Strep.

(Royster et al., JDS 2014)
Selective treatment of clinical mastitis cases

- Rational for targeted antimicrobial therapy
- Application and accuracy of on-farm culture systems to guide clinical mastitis treatment decisions
- Study results: using rapid culture systems to selectively treat clinical mastitis cases
Outcomes of studies using OFC to treat clinical mastitis

Efficacy of the Selective Treatment of Clinical Mastitis During Lactation Based on On-Farm Culture Results

Lago et al. (2011) Journal of Dairy Science

- 8 herds: MN, WI, ON (150 – 1,800 cows)
- 449 mild or moderate clinical mastitis cases
Study Objectives:

- Evaluate the effect of the selective treatment of clinical mastitis based on on-farm culture results on:
  - Antibiotic use
  - Short-term udder health outcomes (cures)
  - Longer-term outcomes (lactation SCC, milk yield, relapses, culling)
Methods: Enrollment Process

Enrollment Day

Positive-Control Group

Enrollment Process

Culture-Based Group

No Growth

Mixed Infection

Gram-pos

Next Day

Gram-neg

No Growth
### Methods: Follow-Up

<table>
<thead>
<tr>
<th>Enrollment (day 0)</th>
<th>Grade 1 or 2 Clinical Mastitis</th>
<th>Day 14</th>
<th>Day 21</th>
<th>Day 0 to 12 months</th>
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<tbody>
<tr>
<td>Sample 1</td>
<td>Cefa-Lak® IMM</td>
<td>Sample 2</td>
<td>Sample 3</td>
<td>Follow-up</td>
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<tr>
<td>Positive Control Group</td>
<td>Culture in lab</td>
<td>Treat quarter immediately after milk sample collected</td>
<td>Culture in lab</td>
<td>Culture in lab</td>
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<tr>
<td>Culture Based Group</td>
<td>Culture on Farm</td>
<td>Treat quarter only if on-farm culture shows Gram-pos IMI</td>
<td>Culture in lab</td>
<td>- Farm records: ♦ Clinical Mastitis</td>
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<tr>
<td></td>
<td>Culture in lab</td>
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<td>- DHIA records: ♦ SCC</td>
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<td></td>
<td>♦ Milk production</td>
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<td></td>
<td></td>
<td></td>
<td></td>
<td>♦ Culling/death events</td>
</tr>
</tbody>
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Results: short term outcomes

- Significant reduction in antibiotic use:
  - Positive - Control = 100% of cases treated
  - Culture - Based = 44% of cases treated

- Less extended / secondary treatment:
  - Positive - Control = 36%
  - Culture - Based = 19%

- Tendency for a reduction in days out of the tank:
  - Positive - Control = 5.9 days
  - Culture - Based = 5.2 days
Results: long-term outcomes

- No differences between treatment programs for:
  - Days to Clinical Cure
  - Bacteriological Cure
  - Clinical Mastitis Recurrence
  - Somatic Cell Count
  - Milk Production
  - Culling or Death

- No negative effect from waiting 24 hours to initiate therapy
Results - Bacteriological Cure by Etiology: No Effect of Treatment Program

Tx Program * Etiology

P = 0.41

Bacteriological Cure Risk

- Coliforms
- Streps
- CNS
- S. aureus
- Other

Positive-Control  Culture-Based
Summary

- We should target intramammary antimicrobial therapy after considering cow factors and cause of infection.
- Rapid culture systems, set up on farm or in local vet clinic, can guide clinical mastitis treatment decision.
- Culture-guided decisions can result in 50% reduced antibiotic usage & less discarded milk without changing the probability of treatment success.
Thank you!

Questions?