Antibiotics Resistance and Antibiotic Use in Beef Cattle Production

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ANTIMICROBIAL RESISTANCE
Global Report on surveillance 2014

The report is the most comprehensive picture to date, with data provided by 114 countries.

Looking at 7 common bacteria that cause serious diseases from bloodstream infections to gonorrhoea.

High levels of resistance found in all regions of the world.

Significant gaps exist in tracking of antibiotic resistance.

Source: WHO, 2014
THE RESISTANCE MOVEMENT
Carbapenem-resistant Enterobacteriaceae have been on the move since at least 1996.


2. 2003: KPC-positive bacteria are found spreading rapidly through hospitals across New York City. By 2007, 21% of Klebsiella in the city carry the resistance gene.

3. 2005: KPC-positive bacteria make their way from New York to several other countries, including Israel. From Israel, the bacteria travel to Italy, Colombia, the United Kingdom and Sweden.

4. 2008: Doctors in Sweden find a new carbapenem-resistance gene, NDM. Traced back to India, NDM-positive bacteria have moved quickly.

Nature doi:10.1038/nature.2014.15135
Microbes are part of the natural world

Deep sea vents
On our skin and in our digestive tract
In the digestive tract of cattle
In our food
Resistance genes are ubiquitous and ancient

Lechuguilla Cave, New Mexico
Region isolated for 4M years

Bhuller et al., 2012

D’Costa et al., 2011
Resistance genes are ubiquitous and ancient

Chait et al., 2012

Schmieder and Edwards, 2012
Void in antibiotic development pipeline

Adapted from Silver 2011 (1) with permission of the American Society of Microbiology Journals Department.
Antibiotic use in Canada

Number of superbug infections in 2012 from 57 Canadian hospitals

- **Clostridium difficile**
- Methicillin resistant Staphylococcus aureus (MRSA)
- Vancomycin resistant Enterococci
- Carbapenem-resistant Enterobacteriaceae

Source: AMMI, 2013
Therapeutic Uses

Treat
Animals diagnosed with an illness

Control
The spread of illness in a herd or flock

Prevent
Illness in healthy animals when exposure is likely

Growth
Balance good/bad bacteria for improved nutrition

Healthy animals
Animals with illness
ANTIMICROBIAL RESISTANCE AND USE IN CANADA
A Federal Framework for Action

Public Health Agency of Canada, Health Canada, Canadian Food Inspection Agency, Canadian Institutes of Health Research and Agriculture and Agri-Food Canada

• Surveillance
• Stewardship
• Innovation

Health Canada, 2014
Canadian Animal Health Institute

April 11, 2014 - Agreed to phase out the usage of medically important antibiotics for growth promotion and supports increased veterinary oversight in antimicrobial use

• Announced on April 11 2014 by Canadian Animal Health Institute (CAHI) companies

• Restricts medically important antibiotic usage in food animals to specific disease challenges under veterinarian direction

• Expected to be implemented over the next three years possibly through the Regulatory Cooperation Council
Importance in treating serious human infections

**CATEGORY**

- **I**: Preferred option for serious infections and limited or no treatment alternatives available
- **II**: Preferred option for serious infections, alternatives available
- **III**: Not a preferred option for serious infections, alternatives available
- **IV**: Not a preferred option, not used in human medicine
Antibiotics approved for animal feed, ranked by importance to humans

<table>
<thead>
<tr>
<th>SWINE FEED</th>
<th>POUlTRY FEED</th>
<th>CATTLE FEED</th>
</tr>
</thead>
<tbody>
<tr>
<td>Antibiotic</td>
<td>Antibiotic</td>
<td>Antibiotic</td>
</tr>
<tr>
<td>Virgiamycin</td>
<td>Virginiamycin</td>
<td>Tylosin phosphate</td>
</tr>
<tr>
<td>Lincomycin</td>
<td>Erythromycin thiocyanate</td>
<td>Oxytetracycline hydrochloride and Neomycin sulphate</td>
</tr>
<tr>
<td>Tilmicosin</td>
<td>Hygromycin B</td>
<td>Bacitracins</td>
</tr>
<tr>
<td>Tylosin phosphate</td>
<td>Penicillin from Procaine penicillin</td>
<td>Chlortetracycline</td>
</tr>
<tr>
<td>Chlortetracycline, sulfamethazine and procaine penicillin</td>
<td>Zinc bacitracin and Procaine penicillin</td>
<td>Sulfamethazine</td>
</tr>
<tr>
<td>Lincomycin and Spectinomycin</td>
<td>Penicillin</td>
<td>Bacitracins</td>
</tr>
<tr>
<td>Tylosin phosphate and Sulfamethazine</td>
<td>Bacitracins</td>
<td>Chlortetracycline</td>
</tr>
<tr>
<td>Zinc bacitracin and procaine penicillin</td>
<td>Chlortetracycline</td>
<td>Oxytetracycline</td>
</tr>
</tbody>
</table>
Ranked by importance to human medicine (I-IV)

- IV: 29.24%
- III: 42.33%
- II: 15.35%
- II/III: 13.05%
- I: 0.03%

Source: CIPARS report, 2008
Ranked by importance to human medicine (I-IV)

I  
26.26%

II  
57.58%

III  
16.16%

Source: CIPARS report, 2008
Antibiotic classes (cellular target)

- **Translation (protein synthesis)**
  - 50s inhibitors:
    - erythromycin
    - chloramphenicol
    - clindamycin
    - lincomycin
  - 30s inhibitors:
    - tetracycline
    - streptomycin
    - spectinomycin
    - kanamycin
    - gentamycin

- **DNA replication**
  - nalidixic acid
  - quinolones

- **DNA synthesis**
  - metronidazole

- **Cytoplasmic membrane**
  - polymyxins

- **Cell wall synthesis & integrity**
  - bacitracin
  - carbapenems
  - cephalosporins
  - D-cycloserine
  - monobactams
  - penicillins
  - vancomycin

- **DNA metabolism**
  - trimethoprim
  - sulfonamide

- **Transcription**
  - rifampicin

- **Translation (protein synthesis)**
  - 50s inhibitors:
    - erythromycin
    - chloramphenicol
    - clindamycin
    - lincomycin
  - 30s inhibitors:
    - tetracycline
    - streptomycin
    - spectinomycin
    - kanamycin
    - gentamycin

- **Folic acid metabolism**
  - trimethoprim
  - sulfonamide

- **DNA replication**
  - nalidixic acid
  - quinolones
Mechanisms of antibiotic resistance

Adapted from Levy & Marshall, 2004
EU withdrawal of nontherapeutic antibiotics in food animal production

- European ban on tet, pen & strep for growth promotion
- 1970
- 1980
- 1990
- Sweden bans AGPs
- Denmark bans routine prophylactics
- 2000
- Specific antibiotic bans in Norway, Germany, Netherlands, Denmark, Sweden & EU
- EU ban on all AGPs
- 2010
POSITIVE outcomes of removing subtherapeutic antibiotics

Johnsen et al., 2009. Lancet Infect Dis 9: 357-364
Cattle drug sales in Denmark since 2001

Source: BeefResearch.ca
Experimental timeline

DAY 1
ARRIVAL AT FEEDLOT

DAY 315
SLAUGHTER

BACKGROUNDING

FINISHING

= Feeding of subtherapeutic levels of antimicrobial agents

= Sampling date
Design

• 150 feedlot steers fed finishing diet
• 4 antimicrobial treatments (3 pens per treatment)
  – Control (no antimicrobials)
  – A44 (chlortetracycline, 44 ppm)
  – AS700 (chlortetracycline + sulfamethazine, 44 ppm each)
  – T11 (tylosin, 11 ppm)

• Within each treatment, feces from each pen were pooled and formed into duplicate artificial fecal pats
Prevalence of shedding: **tetracycline** resistant *E. coli*

![Graph showing the proportion of steers shedding tetracycline resistant E. coli under different treatments.](image-url)
Prevalence of steers shedding *ampicillin* resistant *E. coli*

![Graph showing the prevalence of steers shedding *ampicillin* resistant *E. coli* over time, with different treatments represented by different lines.](image-url)
Experimental Design

**TREATMENT**
- Tylosin (Tylan™) in-feed 11ppm, housed in individual pens in separate wings, n = 10
- Tulathromycin (Draxxin™) 2.5 mg/kg BW, n = 10
- Tilmicosin (Micotil™) 10 mg/kg BW, n = 10
- Control, n = 10

**DURATION**
- Systemic treatment
- In-feed treatment
- Barley (70% silage; 25% grain) + 5% supplement
- Faecal & Nasal sample collection
  - Day 0          *  7          *  14          *  21          *  28

**ANALYSIS**
- Enterococci
  - Ery^R genes PCR
  - PFGE
  - Enterococci isolation on BEA & BEA+Ery
- M. haemolytica
  - Co-isolated species
  - Ery^R genes PCR
  - 16S rRNA PCR for species identification
  - PFGE
- Disc diffusion

Experimental Design

Enterococci

Species identification by pyrosequencing

M. haemolytica

Co-isolated species

Ery^R genes PCR

16S rRNA PCR for species identification

PFGE

Disc diffusion

Experimental Design

Enterococci

Species identification by pyrosequencing

M. haemolytica

Co-isolated species

Ery^R genes PCR

16S rRNA PCR for species identification

PFGE

Disc diffusion
Erythromycin resistance

- Pasteurella multocida: 25%
- Staphylococcus spp.: 20%
- Mannheimia haemolytica: 13%
- Escherichia / Shigella: 8%
- Acinetobacter: 9%
- Bacillus licheniformis: 7%
- Bacillus claussi / Bacillus spp.: 4%
- Staphylococcus chromogenes: 5%
- Unique: 3%
- Bacillus pumilus: 2%
- Macrococcus caseolyticus: 1%
The species profiles of bovine isolates differ from clinical isolates of human origin.

- **Bovine isolates**
  - **E. hirea**: 98%
  - **E. casselilavus**: 2%
  - Total: 130 isolates

- **Human clinical isolates**
  - **E. faecalis**: 80%
  - **E. faecium**: 20%
  - Total: 51 isolates

$n$: Number of isolates pyro-sequenced
Proportion of Ery-resistant bovine enterococci over the study period, with d 0 samples collected prior to treatment. **Compared to the control group, treated groups were 76 times more likely (P < 0.05) to have erythromycin resistant enterococci.**
Enumeration data

Average proportion of tyl resistance (%)

Day

Control
Tylosin

Average proportion of ery resistance (%)

Day

Control
Tylosin
Distribution of enterococcus species over sampling days and between treatments

<table>
<thead>
<tr>
<th>Species prevalence (%)</th>
<th>0</th>
<th>14</th>
<th>84</th>
<th>112</th>
<th>224</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control E. hirae</td>
<td></td>
<td></td>
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<tr>
<td>Tylosin E. faecium</td>
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<tr>
<td>Control E. faecium</td>
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<tr>
<td>Tylosin E. durans</td>
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<tr>
<td>Control E. casseliflavus</td>
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<td></td>
</tr>
<tr>
<td>Tylosin E. faecalis</td>
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</tbody>
</table>

Legend:
- E. hirae
- E. faecium
- E. durans
- E. casseliflavus
- E. faecalis
Resistance in *E. coli* from beef processing continuum

Adapted from: Schmidt et al., 2014 AEM 81:713-725
Mobile genetic elements

- Plasmids
- Integrated plasmids
- Genomic islands
- Prophages
- Integrons
- Integrative conjugative elements
- Conjugative transposons
- Transposons

IS elements
Resistance is encoded by diverse soil phyla.

Functional selections of 18 soil libraries yield diverse ARGs.

Effects of manure on the abundances of culturable soil bacteria.

Udikovic-Kolic N et al. PNAS 2014;111:15202-15207
I Chip Culturing the Unculturable.
The structure of teixobactin and the predicted biosynthetic gene cluster.

Time-dependent killing of *S. aureus* by teixobactin.
Teixobactin is efficacious against MRSA and *S. pneumoniae*

MRSA - septicemia

MRSA – muscle infection

*Streptococcus pneumoniae* – lung challenge

The SCIENCE of antimicrobial usage in livestock production

**RESEARCH**
Study the kinds & flow of resistance (genes) and zoonotic potential of microbes

**MANAGEMENT**
Consider livestock production practices to reduce use and potential impact on human medicine

**SURVEILLANCE**
Monitor the emergence of resistance (genes) and potential for transfer between livestock – the environment – and consumer products

**DEVELOPMENT OF ALTERNATIVES**
Design/test therapeutic approaches against veterinary or zoonotic diseases
Take Home Point

• Microbes are masters of adaptation

Oct. 15, 2012 - Health Canada Recalls Antibacterial hand soap
– *Pseudomonas aeruginosa*
Beef AMR / AMU funding
Thank you

Questions?