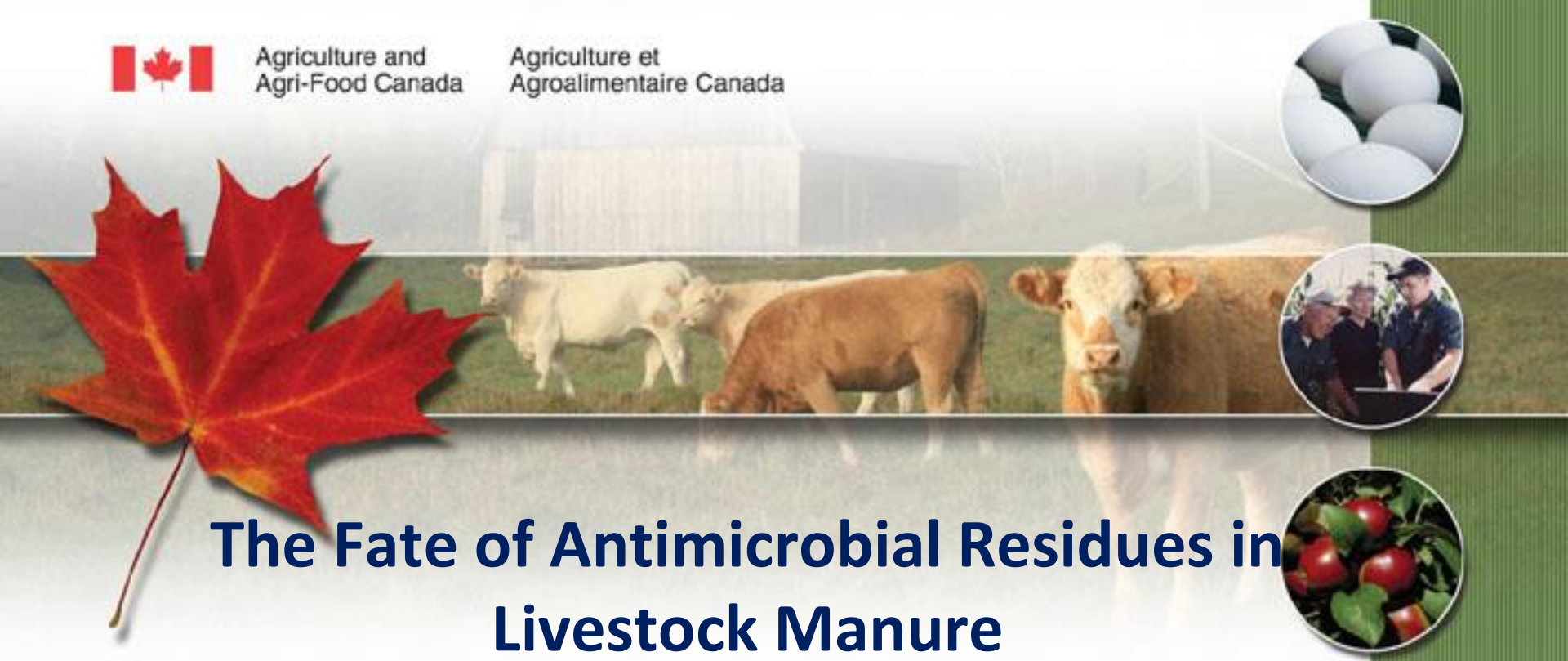

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The Fate of Antimicrobial Residues in Livestock Manure

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Manure Management Update
January 19, 2015, Lethbridge, Alberta

Canada

Use of Veterinary Antimicrobials

- Animal production
  - Therapeutic (disease prevention, control & treatment)
  - Sub-therapeutic (growth promotion & improve feed efficiency)
  - Added in feed or water
  - Available as prescription and over-the-counter

- About 70% are administered to promote growth, improve feed efficiency, and disease prevention

Use of Veterinary Antimicrobials

- Antimicrobials used to treat livestock are also used to treat humans
- Antimicrobial use on animals is ≈4 times the humans

Antimicrobial	Animals Treated	Uses in human medicine
Chlortetracycline Oxytetracycline	Cattle, swine, sheep, chicken (broilers), turkey	Pneumonia, respiratory, urinary, intestinal and skin infections; Lyme disease, smallpox, anthrax, malaria, Rocky Mountain spotted fever, typhus, venereal disease, Chlamydia.
Tylosin	Cattle, swine, sheep, chicken (broilers and layers), turkey	Pneumonia, bronchitis, diphtheria, Legionnaires' disease, whooping cough, rheumatic fever, venereal disease; ulcers; ear, lung, intestinal, urinary and skin infections.

Fate of Antimicrobials in Feed and Manure

Fate of Antimicrobials in Manure

- Manure management options
  - Windrow composting, stockpiling, storage in lagoons (liquid)
  - Land application of manure as fertilizer (raw or composted)
  - Antimicrobials may be degraded

Windrow composting

Manure stockpiling

Land application of manure

Fate of Veterinary Antimicrobials in Solid Manure

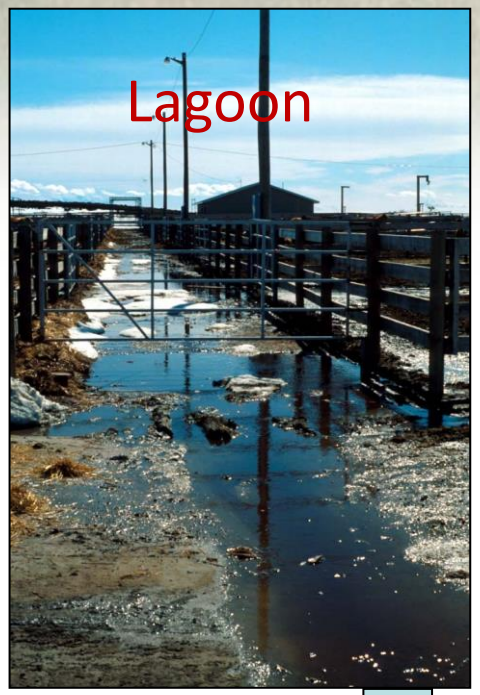
1 km

>10 km

High manure loadings

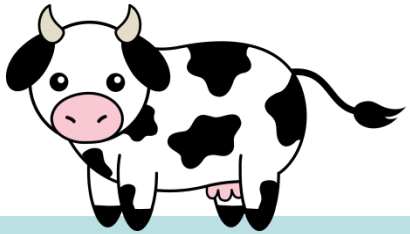
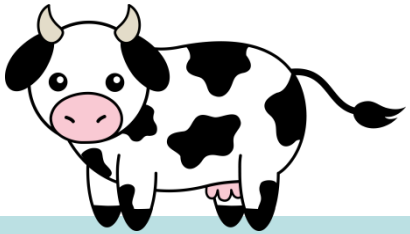
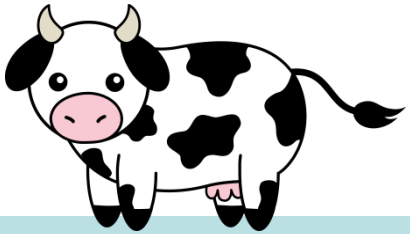
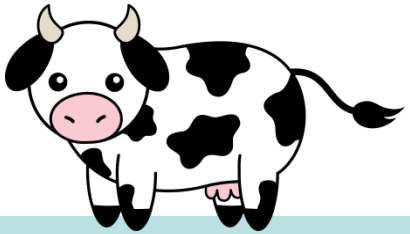
Low manure loadings

Fate of Antimicrobials in water



Study Design

- Feedlot based studies were carried out in research facility at the Agriculture and Agri-Food Canada Research Centre, Lethbridge
- Beef cattle were administered antimicrobials via feed

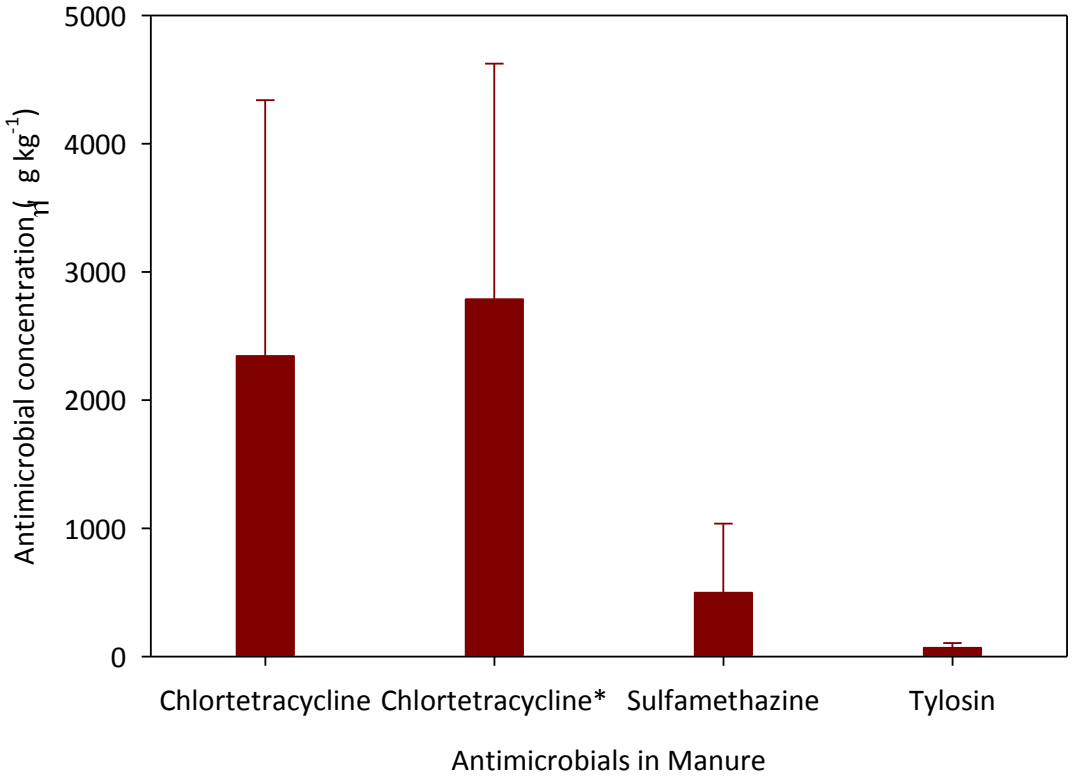





CTC	CTCSMZ	TYL	Control
Chlortetracycline @ 44 mg per kg feed	Chlortetracycline + sulfamethazine @ 44 mg kg^{-1} feed each	Tylosin @ 11 mg kg^{-1} feed	Feed only

- Commercial feedlots (Acme and Nanton, Alberta)
- Irrigation canal supply water

Antimicrobials in Manure from Feedlot Pens

- All three antimicrobial were detected in the manure over 3-year period

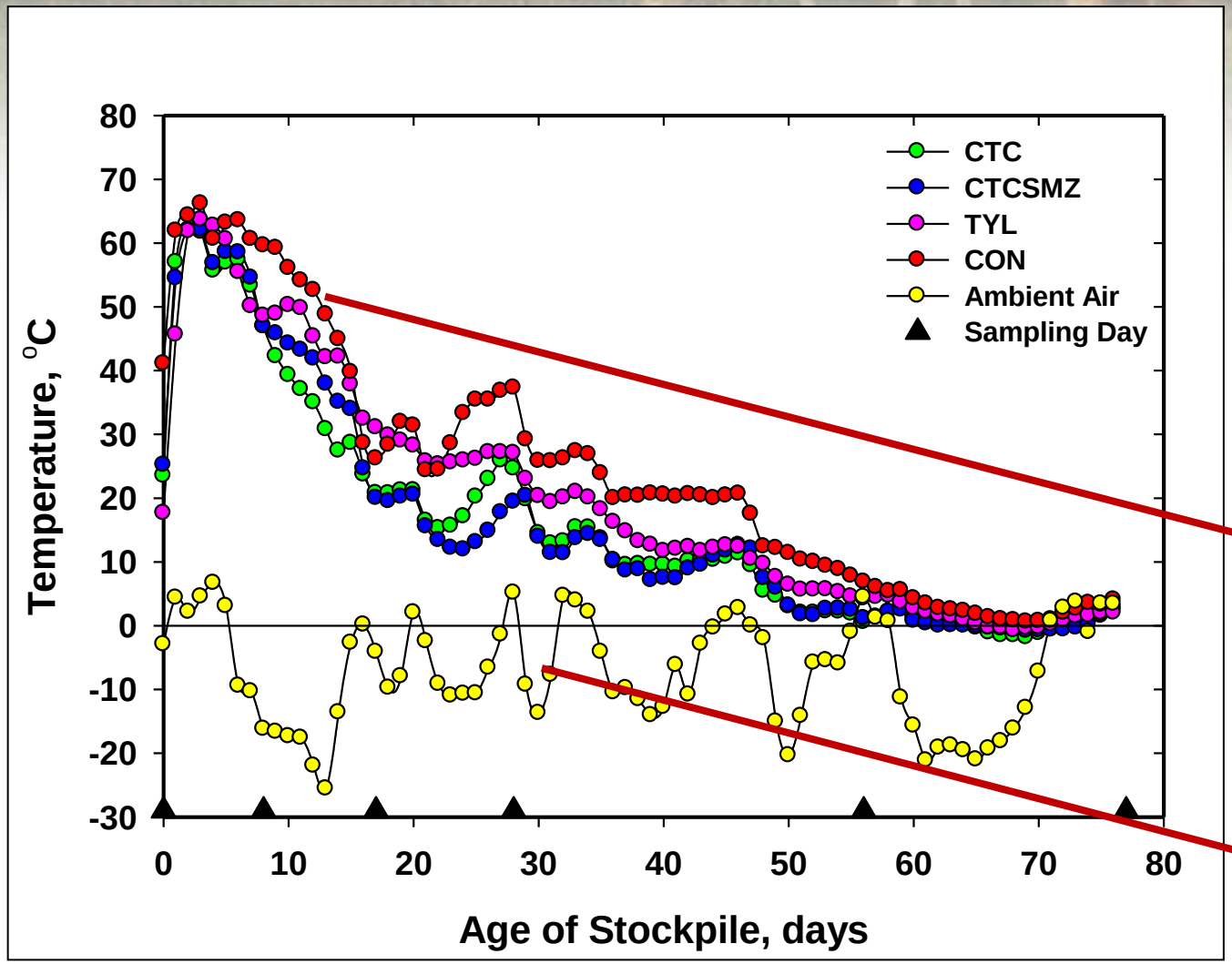


*** Chlortetracycline in CTCSMZ**

Degradation of Antimicrobials during stockpiling

- Study objective: To determine the degradation of antimicrobials during manure stockpiling
- Feedlot pens cleaned and stockpiles set up
- 4 treatments (CTC, CTCSMZ, TYL and Control) x 2 replicates = 8 stockpiles
- Temperature measured within the stockpiles
- Manure sampled at Days 0, 8, 17, 28, 56, 77, and 140
- Manure samples were analysed for antimicrobial concentration

Temperature within the stockpile



Manure temperature: 0.4 °C to 65.4 °C

Air temperature: 6.3 °C to -24 °C

Degradation of Antimicrobials: Stockpiles vs Windrows

Antimicrobial	Half-lives* (days)	
	Stockpiles	Windrows
Chlortetracycline	2 to 6	15 to 21
Sulfamethazine	21	27
Tylosin	5	32

* Half-life is the time period for an antimicrobials to decrease to 50 % of initial concentration – measure of persistence. Shorter half-life means antimicrobial disappears rapidly.

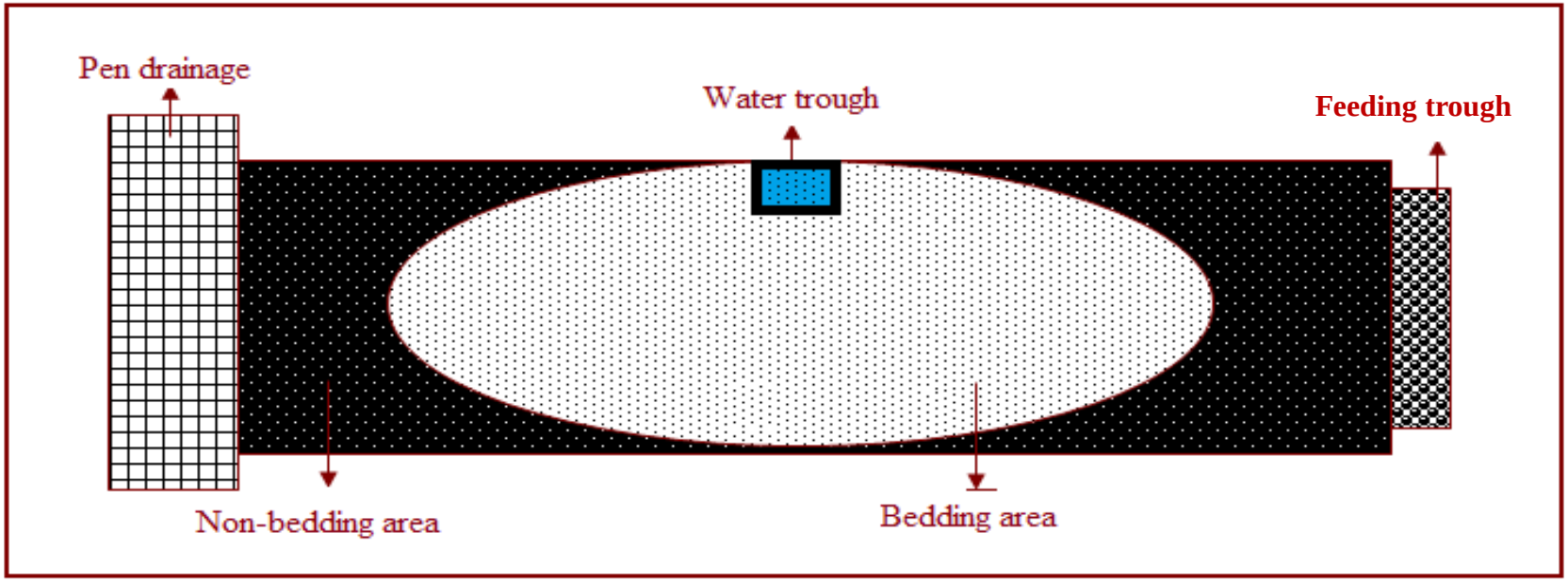
% of initial concentrations remaining after Day 77

Antimicrobial	Day 0	Day 77	Remaining
	µg kg^{-1}	µg kg^{-1}	%
Chlortetracycline	4892 to 5568	26 to 32	0.5 to 0.7
Sulfamethazine	4432	47	1.1
Tylosin	76	18	23.7

Loss of Antimicrobials in Rainfall Runoff: Feedlot Pens

- To quantify the concentrations of antimicrobials (chlortetracycline, sulfamethazine, tylosin) in the simulated rainfall runoff from two locations (bedding and non-bedding areas) in feedlot pens
- To quantify the amount of each antimicrobial that would potentially be transported in runoff from a feedlot pen to the adjacent catch basin

- 4 treatments (CTC, CTCSMZ, TYL and Control)
- Three pens per treatment
- 2 locations (randomly selected) per pen (bedding pack and pen floor)



Schematic diagram of a beef cattle pen in Lethbridge Research Farm at Agriculture and Agri-Food Canada, Lethbridge, Alberta

Rainfall Simulation Setup

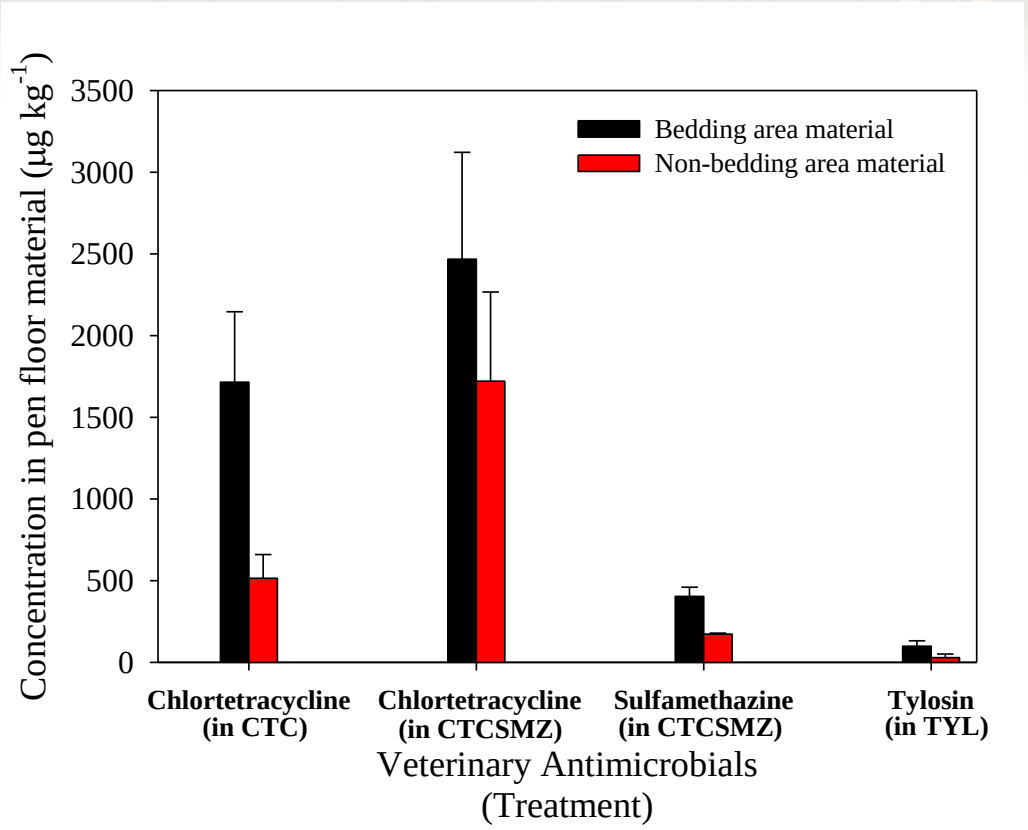
Photograph showing one of the beef cattle pens in the Lethbridge Research Centre Research Feedlot used in this study. Guelph rainfall simulator seen in the in-set.

Materials & Methods: In Feedlot Pens

- Sample Collection
  - Rainfall runoff
    - Fourteen 1-L samples of runoff
    - Additional runoff after rain was stopped
    - Samples 1, 2, 6, 10, 14 (individual samples)
    - Samples 3, 4, 5, 7, 8, 9, 11, 12, and 13 (composite sample)
- Manure samples
  - Composite pen floor and bedding manure (adjacent to rainfall simulator before rainfall was started)

Antimicrobial in Pen Floor Material

- Concentrations of all three antimicrobials were higher in the bedding than non-bedding material.



Concentrations of antimicrobials in bedding and non-bedding area material.

Antimicrobials Mass transfer in Runoff

Antimicrobial in Treatment	Runoff Export Coefficient (REC) ($mg\ m^{-2}\ min^{-1}$)
Chlortetracycline in CTC	1.31
Chlortetracycline in CTCSMZ	2.47
Sulfamethazine in CTCSMZ	1.62
Tylosin in TYL	0.18

- Runoff export coefficients (REC) were derived by dividing the total mass of antimicrobial transported by the time taken to collect runoff from start of rainfall.
- REC is a measure of the mass of each antimicrobial transported in the runoff per unit area per unit time.

Conclusions

- Mass of antimicrobials transported in rainfall runoff:

Antimicrobial	Loss from pens
Chlortetracycline	13 to 36 g (2.4 to 6.7%)
Sulfamethazine	19 g (3.6%)
Tylosin	2 g (1.5%)

- Our study indicates that the runoff from feedlot pens can be a source of veterinary antimicrobials and reiterates the importance of well-maintained catch basins for retaining runoff from intensive feedlot operations.

Loss of Antimicrobials in runoff from manure-amended croplands

Loss of Antimicrobials in rainfall runoff: Manure-amended croplands

- Antimicrobials are transported in rainfall runoff from manure-amended croplands

Antimicrobial Concentration in Runoff (% of applied)		
Antimicrobial	**Surface application**	**Soil incorporated**
Chlortetracycline	8.7-9.5	1.8-3.4
Sulfamethazine	6.5	4.6
Tylosin	0.6	0.5

- Antimicrobial losses were greater when surface applied compared to soil incorporated

Antimicrobial Concentrations: Commercial Feedlot Catch Basins

Antimicrobial concentrations at Acme and Nanton sites		
Antimicrobial	**Acme (ng/L)**	**Nanton (ng/L)**
Chlortetracycline	20	31
Sulfamethazine	1	5
Tylosin	5	56
Monensin	6	310
Lincomycin	69	7
Tetracycline	38	107

Antimicrobial Concentrations: Irrigation Water from Canals in Alberta

Antimicrobials concentrations* in irrigation water in Alberta				
Antimicrobial	**June (ng L^{-1})**	**July (ng L^{-1})**	**August (ng L^{-1})**	**September (ng L^{-1})**
Chlortetracycline	41	30	28	38
Sulfamethazine	1	1	-	2
Tylosin	1	2	1	18
Monensin	-	1	4	2
Lincomycin	2	2	1	2
Tetracycline	57	77	70	85
Erythromycin	5	3	1	5

* Concentrations are average value of 24 sites

Antimicrobial Concentrations: Safe levels in animal tissue/food products

Maximum residue limits for antimicrobials foods (Health Canada, 2014)				
Antimicrobial	**Meat ($\mu g\ kg^{-1}$)**	**Kidney ($\mu g\ kg^{-1}$)**	**Milk ($\mu g\ L^{-1}$)**	**Eggs ($\mu g\ kg^{-1}$)**
Chlortetracycline	200	1200	100	400
Sulfamethazine	100	100	10	-
Tylosin	200	200	-	-
Monensin	50	50	10	-
Lincomycin	100	-	-	-
Tetracycline	200	1200	100	-
Erythromycin	100	100	50	-

Conclusions

Via feed @ 11 to 44 mg kg^{-1}

~ 80% is excreted in feces and urine

Detected in pen floor manure (@ µg kg^{-1})

Transported in runoff (CTC @ 13 to 36 g per pen)

Degrade in stockpiled manure (CTC: 2 to 6 days)

Degrade in windrows (CTC: 15 to 21 days)

Transported in runoff (CTC @ 1100 µg L^{-1})

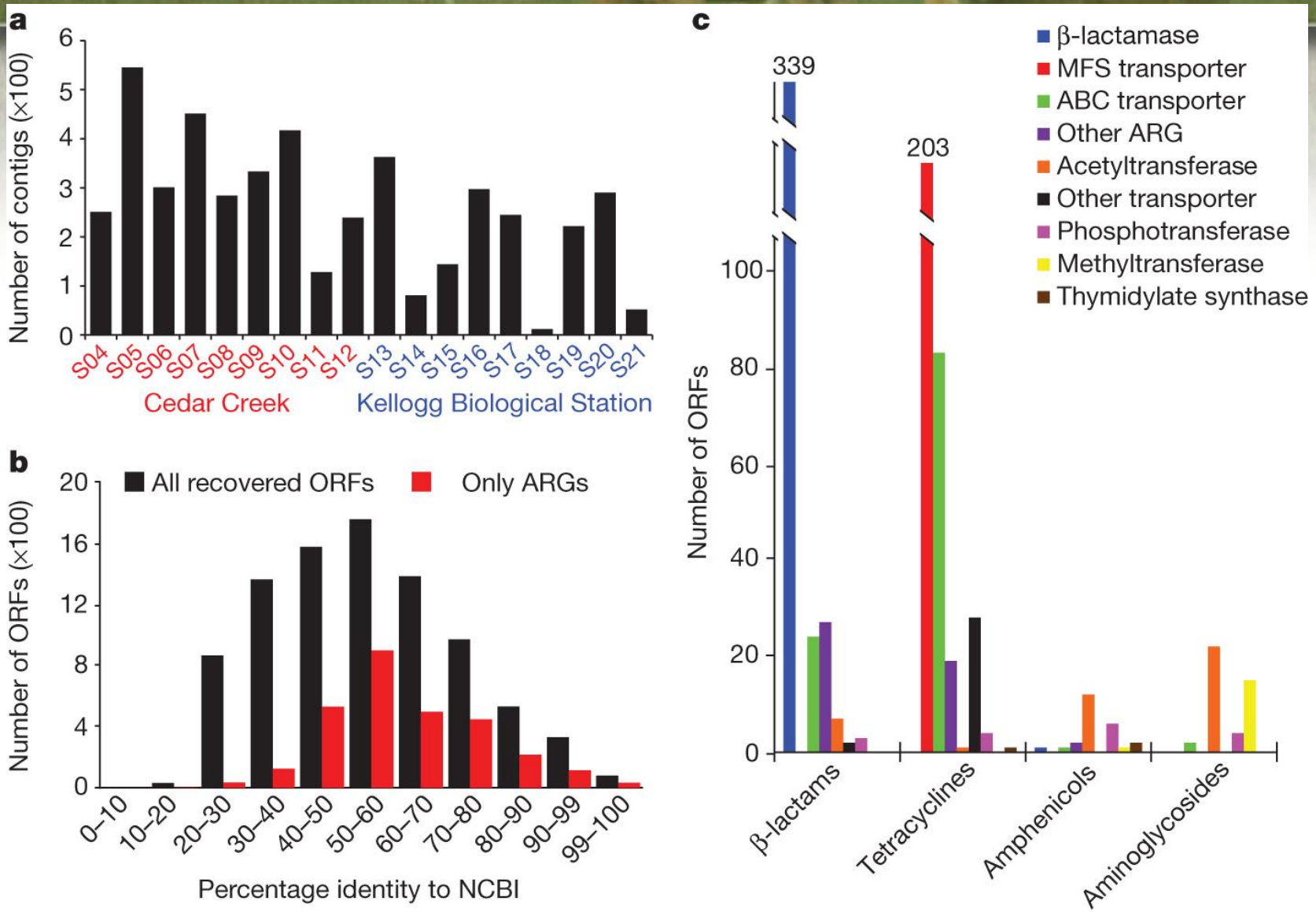
Transported in runoff from croplands (CTC @ 17 to 43 µg L^{-1})

Detected in catch basins (CTC @ 20 to 30 ng L^{-1})

Detected in irrigation water (CTC @ 30 to 40 ng L^{-1})

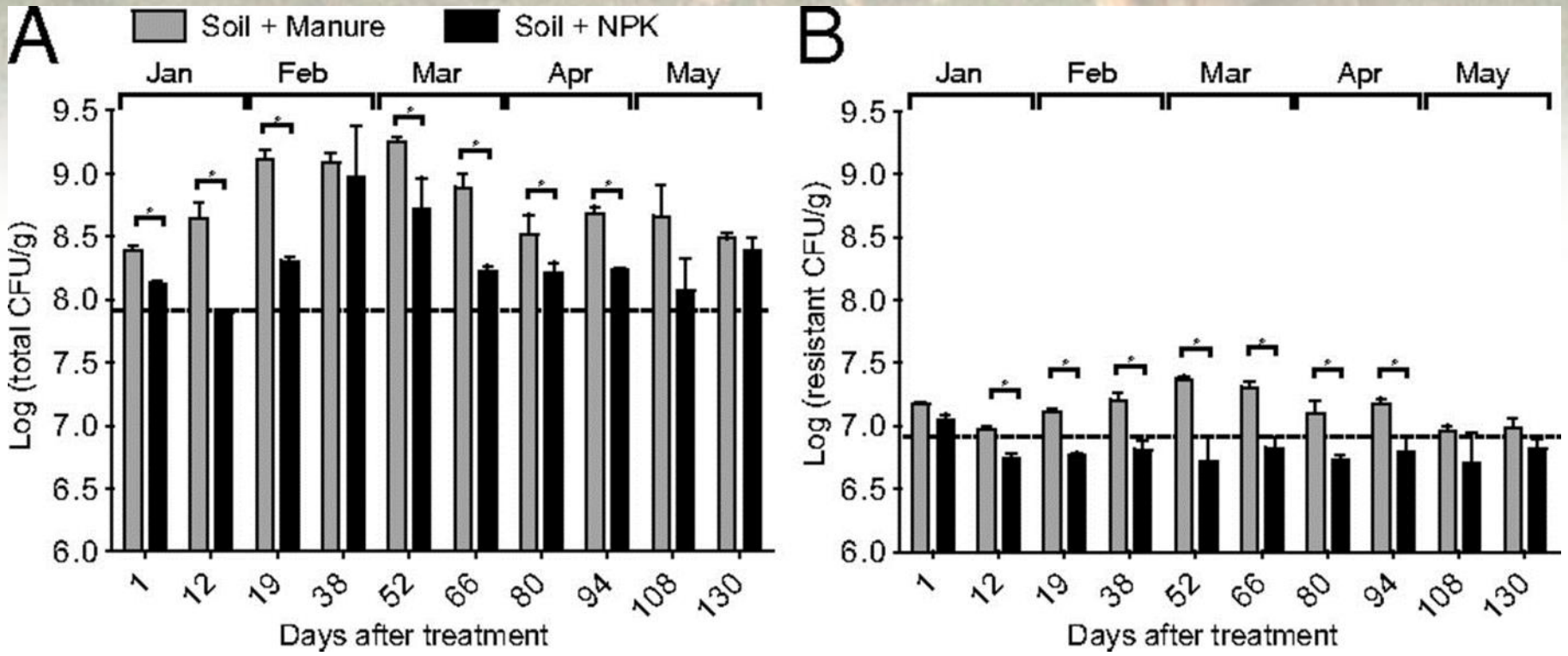
Maximum residue limits (CTC @ 100 to 1200 µg kg^{-1})

Functional selections of 18 soil libraries yield diverse ARGs.



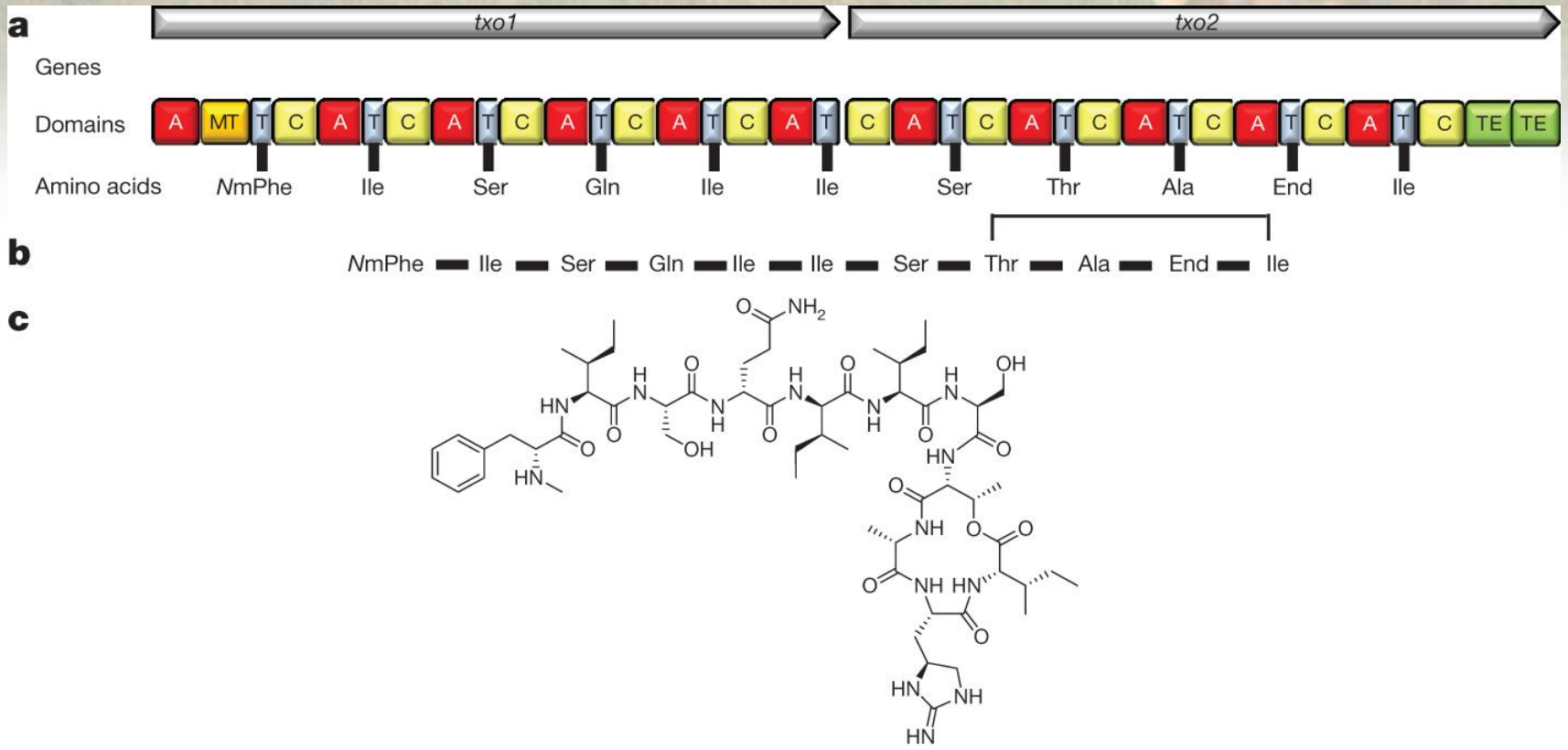
KJ Forsberg *et al. Nature* **000**, 1-5 (2014) doi:10.1038/nature13377

Effects of manure on the abundances of culturable soil bacteria.



Udikovic-Kolic N et al. PNAS 2014;111:15202-15207

The structure of teixobactin and the predicted biosynthetic gene cluster.



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Acknowledgements

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Thank you!

Growing Forward 2

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