Guts and Bugs
The Good, the Bad, the Ugly

Jenn Rowntree, DVM

Healthy guts = healthy calves

- **Pathogens impact the small intestine’s normal microbiota and barrier function**
  - Bugs talk!? Pathogen and host-microbe interactions in the gastrointestinal tract (GIT)

- **What can we do about it?**
  - Optimize management practices that contribute to GIT environment

- **How does this affect my bottom line?**
  - Diarrhea responsible for 56% of pre-weaned heifer deaths (NAHMS 2014)
  - Reduced 1st lact 305-ME, actual milk, fat, and protein (Heinrichs and Heinrichs, 2011)
  - Increased age at first calving (Heinrichs, 2005)
Gut microbiome

• Naturally-occurring organisms in the GIT
• Common microbiota predominating in healthy cattle:
  • Firmicutes, Bacteroides, Proteobacteria (Bickhart & Weimer 2018)
  • Composition and functions vary with age and different gut locations (Malmuthuge et al., 2014)
  • Affected by physiological and health status (Bickhart & Weimer 2018)

Why is a healthy gut microbiome important?

• Critical role in metabolism, immune response, and GIT regulation (Guarner, 2006)
  • Modulate development of intestinal epithelium and mucosal layer (Sharma et al., 1995) plus immune system (Mebius 2013)
  • Maximize nutrient absorption to promote efficient growth
• Gut microbiota impacts overall health
  • Sustain immune responses that detect, prevent, and eliminate bad bugs while tolerating good bugs (Bischoff, 2011)
  • Maintain homeostasis to prevent invasion of bad bugs
Mucosal immune system

- Immune responses that occur in tissues exposed to external environment
  - Physical and chemical components
  - First week of a calf’s life critical to development (Liang et al., 2014)

- Requires a healthy gut microbiome
  - For development, maturation, and homeostasis (Hooper et al., 2012)

- Critical to eliminating pathogens
Components of mucosal immune system

- Physical barriers – prevent invasion of bad bugs from entering GIT tissue
  - Mucus layer – trap microbiota
  - Epithelium – single layer of cells connected by tight junctions (Ulluwishewa et al., 2011)
    - Transport through cells and tight junctions to absorb molecules
    - Tight junctions – primary regulator of intestinal barrier function
  - Chemical barriers – limit growth of bad bugs (Johansson et al., 2011)
    - IgA – produced by mucosal immune system
    - Protect good bugs in mucosal layer (Gutzeit et al., 2014)
    - Clear bad bugs and maintain homeostasis
    - Antimicrobial peptides – can ID and kill pathogens
  - Pattern-recognition receptors
    - Maintain integrity of intestinal barrier (Ulluwishewa et al., 2011)
    - Help immune system learn to ID bad bugs
Microbial endocrinology

- AKA – how good and bad bugs respond to signals from other gut bugs and their host
- Norepinephrine – signal molecule between host and microbiota
  - Hormone released in response to stress
  - Intermediary for both good and bad gut bugs
  - Potential to effect changes in growth and metabolism of various microbes

Damage to microbiome and mucosal immune system due to:

- Stress
- Antibiotics
- Invasive and opportunistic pathogens
Stress impacts the good gut bugs

- **Good bugs**
  - Key regulators of gut-brain axis (Foster et al., 2017)
  - **Alters microbiota (Lyte 2016)**
  - **Study in mice by Bailey et al., 2011:**
    - Decreased abundance of *Bacteroides*
    - Increased relative abundance of *Clostridium* species

Stress impacts the bad gut bugs

- **Norepinephrine released at high concentrations =>**
  - Acts on bacteria (ex. O157:H7 E. coli) to promote movement towards intestinal surface, enhance growth and virulence (Green et al., 2004, Lyte and Ernst 1992)
- **Release of signal factors from injured GIT nerves**
  - Microbial population changes from mostly gram-positive good bugs to a single gram negative species (ex. E. coli) (Lyte and Bailey, 1997)
**How do antibiotics impact the good bugs?**

- **Reduce total bacterial populations** (Ubeda et al., 2010)
  - Decrease beneficial bacteria (Xie et al., 2013)
  - Precipitate *Salmonella* infections
- **Increase antibiotic resistance in opportunistic bugs** (Ubeda et al., 2010)
  - Ex. *E. coli, Enterococcus*, (Xie et al., 2013)
- **Cause microbial imbalance in GIT**

**Antibiotics and the microbiome**

- **Avoid use of oral antibiotics for treatment of scours** (Constable 2004)
  - Current label Rx options not consistently effective
  - Goal for scours treatment:
    - Control growth of *E. coli* in small intestine
    - Minimize damage to beneficial gut microflora
  - Diarrhea, normal appetite, no fever – monitor, administer electrolytes
  - Diarrhea, no appetite, fever – administer broad-spectrum antibiotic, electrolytes
    - Ex – ceftiofur, amoxicillin or ampicillin
    - NOT enrofloxacin
What about waste milk?

- Maynou et al., 2017 – calves fed waste milk vs. milk replacer:
  - Whole waste milk contained B-lactam residue =>
    - Higher number of antibiotic-resistant *E. coli* bacteria in feces
      - To enrofloxacin, florfenicol, and antibiotics with B-lactamase genes
    - Higher prevalence of colistin-resistant *Pasteurella multocida* in nasal swabs
  - Antibiotic resistance changes with age
  - How will this impact effectiveness of antibiotics in sick calves?
    - More research needed

Bad gut bugs

Invaders and commensals that overstay their welcome

- Common perpetrators:
  - *Clostridium perfringens*
  - Rotavirus and coronavirus
  - *Cryptosporidium parvum*
  - Coccidia

https://www.proactiveherapymb.com/dysbiosis.html
**Clostridium perfringens**

- **Gram positive, anaerobic bacteria**
  - Found in environment, GIT microflora
  - Multiple types

- **Toxins cause damage**
  - Presence of bacteria ≠ disease

- **Risk factors for toxin production:**
  - Large quantities of soluble carbs and/or protein
  - Presence of signaling molecules – ex. norepinephrine, epinephrine
    - Epinephrine increases growth rate and decreases infective dose (Cooper 1946)

  Roos et al., 2015

---

**Rotavirus**

- **Non-enveloped virus**
  - No outer shell = resilient

- **Transmission:**
  - Older calves and adult cows serve as carriers

- **Diarrhea at 7-14 days of age**
  - Lasts 3-7 days – calves shed millions of viral particles/gram of feces
  - 50-100% of calves affected, varying death rates
  - Often coexists with other pathogens

- **Damage limited to small intestines**
  - Decreased absorption and digestion of nutrients
Coronavirus

- **Enveloped virus**
  - Outer shell easily damaged
- **Fecal-oral transmission**
- **Diarrhea at 7-10 days of age**
  - Lasts ~1 week
  - Severe – death rate >50% when combined with other bad bugs
- **Mass destruction to SI and LI**
  - Reduced ability to absorb electrolytes
  - +/- IV fluids, systemic antibiotics

Cryptosporidium parvum

- **Protozoan**
  - Extremely hardy, persists for months
- **Transmission:**
  - Fecal-oral – infective dose <100 oocysts
  - ZOONOTIC
- **Diarrhea in calves 5-28 days old**
  - Calves shed millions of oocysts per gram of feces
- **Destroys host cells along entire GIT**
  - Cell death and damage predispose calf to other infections – ex. *E. coli*, viruses, *Salmonella*
Response to cryptosporidium infection

The Ugly Gut Bugs

Quorum-sensing culprits
- E. coli, Salmonella, Klebsiella (Curtis et al., 2014, Moreira et al. 2016)
- Quorum-sensing proteins
  - Involved in virulence factors, bacterial growth, and colony density (Lyte et al., 2018)
  - Release of NE enhances ability to infect host:
    - Improves E. coli 0157:H7 attachment to SI and fluid secretion (Vlisidou et al., 2014)
    - Increases replication of Salmonella typhimurium in GIT (Pullinger et al., 2010)
    - Bacteria adhere to intestinal lining => use NE to call other bacteria (Pasupuleti et al., 2014)
What can we do about it?

1. Promote diverse gut microbiome
   - Robust mucosal immune system
   - Focus on nutrition

2. Limit stress
   - And exposure of GIT to stress-related hormones

3. Reduce exposure to pathogens
   - Implement biosecurity protocols
   - Even if exposure is inevitable

1. Focus on nutrition: start with colostrum

   • Provide clean, high-quality colostrum ASAP
     - 4 quarts in 4 hours
     - Calves need a source of glucose
   
   • Helps beneficial bacteria colonize SI (Malmuthuge et al., 2015)
     - Single feeding of heat-treated colostrum soon after birth (<12 hours) promoted colonization with *Bifidobacterium* and reduced colonization with *E. coli*
     - Natural prebiotic = help the good bugs beat the bad bugs to the SI
Colostrum during disease challenge

Study by Chamorro et al., 2017: 2 treatment groups of 100 calves

- Control group – no colostrum supplement
- Treatment group – 150g colostrum supplement powder twice daily for first 14 days of life

• Results
  - Mean body weight, ADG at weaning not significantly different among treatment groups
  - Reduced antibiotic therapy in treatment group (18.8%) vs. control group (76.5%)
  - Reduced disease in treatment group

• On-farm:
  - Freeze good quality colostrum (BRIX >20) in ice-cube trays
  - Feed 1 cube per calf per feeding for 1st 14 days of life

1. Focus on nutrition: milk

• Quality
  - Clean and free of bad bugs
  - Osmolality similar to cow’s milk
  - Fed at 12-14% total solids

• Quantity
  - Must meet energy requirements for growth
  - 100lb calf requires 5.7 lbs (2/3 gallon) of whole milk for maintenance per day (Drackley, 2008)
  - Enough to support immune function, temperature extremes
1. Focus on nutrition: quality calf starter

Early introduction to high quality calf starter:

• **Promotes rumen development and facilitates early weaning**
  - Diverse microbial community (Malmuthuge et al., 2013)
  - Improved growth rates

• **Increased ability to fight disease**
  - Earlier expression of antimicrobial defense molecules that help ID and kill pathogens (Malmuthuge 2015)
  - Influences GIT barrier function and immune responses

1. Focus on nutrition: probiotics

**Probiotic** – a source of **live**, viable good bugs or yeast

• Interact with microflora, GIT epithelium, immune cells

• **Bacillus subtilis** – increased ADG, feed efficiency, decreased weaning age by 7 days (Sun et al., 2010)

• **Saccharomyces cerevisiae** – decreases susceptibility of calves to GI infections
  - Decrease # of days with diarrhea in calves with failure of passive transfer (Galvao et al., 2005)
  - Feeding with grain decreases incidence of diarrhea and death rate in calves <70 days of age (Magalhães et al., 2008)

• **Potential problems**
2. Limit stress to calves

- Minimize pain associated with procedures whenever possible
- Avoid simultaneous stressors – dehorning, vaccines, moves
- Gradual weaning
- Temperature control – avoid heat or cold stress (40-70°F ideal)
- Dry, well-bedded environment
- Adequate ventilation, volume and area per calf
- Fly control

3. Minimize exposure to pathogens

- Biosecurity and biocontainment practices can reduce risk of pathogen transmission
  - Effective cleaning and disinfecting protocols
  - Manage animal movement
- Infection and disease result of:
  I. Innate resistance of host animal
  II. Infectious dose received
  III. Virulence of particular strain
Conclusion

Maximize resistance of animal and reduce pathogen exposure

- Promote healthy gut microbiome to increase GIT immune system
  - Colostrum, starter intake
  - Dry cow vaccines
  - Optimal facilities and ventilation

- Reduce exposure to triggers that break down GIT barrier function
  - Stress
  - Poor hygiene
  - Pathogen load

Citations

Citations

- Ubeda et al., 2010. Vancomycin-resistant Enterococcus domination of intestinal microbiota is enabled by antibiotic treatment in mice and precedes bloodstream infection in humans. J. Clin. Invest. 120:4332-4341.