


**The Effect of Continuous Low-level Feeding of Aureomycin on Dairy Cows**  
 CE Polan, GA McLaren, AH Rakes... - 1962 - West Virginia University, Agricultural ...


**The effect of continuous feeding of chlortetracycline on lactating dairy cows.**  
 CE Polan - 1960 - West Virginia University

**Biohydrogenation of unsaturated fatty acids by rumen microorganisms.**  
 CE Polan - 1963 - North Carolina State College.

## ROLE OF BACTERIA IN RUMINANT NUTRITION





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## Probiotics and the health of newborn calves?


Bob James  
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## The newborn calf




- Calf is sterile at birth
- Early microbial colonization - rather haphazard?
- Risk of colibacillosis? Early colonization by E. coli.
- Can dosing calf with “good”bacteria” colonize intestine and prevent establishment of E. coli.



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## Probiotic product from Salem, VA?

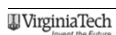
- Lactobacilli grown on wheat bran
- Dose calf with probiotic in shipped in calves housed at Vet Science
- Challenge with enteropathogenic strain of E. coli.



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## Problems with probiotics – according to W. E. C. Moore – VPI Anerobe Lab

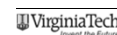
- Most intestinal organisms are anaerobes not aerobes
- Microflora is a mixed culture of organisms.
- Likelihood of single species or few species to successfully populate is nil
- Intestine is rapidly populated by organisms at birth.



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## Source of meaningful probiotic?

- Source of organisms for probiotic?
  - Older milk-fed calf?
- Duodenal cannulated
- Collect and mix with whey solution
  - Intestinal fluid is of low palatability.

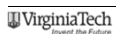


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## Influence of “probiotic” on resistance to enteropathogenic E. coli challenge

James et al, 1976

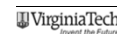
- Calves entered our facility @ ~ 3 hour of age
- Colostrum fed @ ~ 5 hours of age
  - No E.coli challenge
  - E. coli challenge @ 12 hours
  - E. coli challenge @ 24 hours
  - All with or without “probiotic” inoculum of 200 ml of duodenal fluid from milk-fed calf.
    - Whey solution as carrier



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## Results

- Calves receiving probiotic inoculum had less diarrhea and higher ADG for 1<sup>st</sup> 14 days.



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**Serum gamma globulin – g/100 ml @ 24 h**

Inoculum	O	12 h challenge	24 h challenge	Means
No inoculum	1.1	0.94	1.68	1.24
Inoculum	.5	0.49	.59	.53
Means	.80	.72	1.14	.89



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**Follow up study**

James et al., (1978)

- Three Treatments
  - Colostrum @ 2.5 hours of age
  - Colostrum and inoculum (200 ml duodenal fluid) concurrently @ 2.3 hours of age
  - Inoculum followed three h later by colostrum @ 5.6 hours of age
  - No E. coli challenge



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**Mean total protein and gamma globulin (g/dl) @ 24 hours of age.**

Treatment	Total serum protein	Gamma globulin
No inoculum	5.97+/-0.94	1.05+/- .36
Colostrum and inoculum concurrent	6.07+/-0.64	1.08+/- .41
Delayed colostrum	5.22+/- .59	.76+/- .31

Delayed colostrum calves had lower protein and gamma globulin



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**Cause of apparent depression in Ig absorption?**

- Live bacteria?
- Cell walls of bacteria (endotoxins)?
- Carrier for the inoculum – dried whey?



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## Intensive study

James et al, (1981)



- Utilized “gut loops” in newborn calves ~ 8.6 hours at beginning of treatments
- Constructed beginning 1.8 M anterior to ileocecal junction
- Each calf received all treatments
- Incubated for 4 hours
- Inject loops with <sup>125</sup>I – gamma globulin in electrolyte solution incubated for 75 min.



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## Treatments

- Live bacteria – .3ml duodenal fluid – 9 ml anaerobic rumen fluid glucose cellobiose broth (RGC). **Anaerobic culture**
- Autoclaved culture
- Sterile RGC broth



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## Treatment means for uptake, bacterial no. in tissue and inoculum

Uptake of gamma globulin <sup>a</sup>			Bacterial growth in tissue <sup>b</sup>		
Live bacteria	Sterile broth	Autoclaved bacteria	Live bacteria	Sterile broth	Autoclaved bacteria
2.09 +/- 1.15	3.18 +/- 1.50	3.56 +/- 1.49	594 +/- 316	86 +/- 82	220

<sup>a</sup> milligrams gamma globulin internalized/g tissue  
<sup>b</sup> Bacteria (X10<sup>6</sup>)  
 Uptake reduced only with live bacteria culture – (P<.05)  
 # organisms significantly related to uptake



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## Three Q's of colostrum management

- Quickness – feed as soon as possible with a goal of less than 6 hours
- Quantity – 4 liters in 1<sup>st</sup> 12 hours
- Quality - >50g/liter
- Not colostrum again!



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## Add another letter to the list?

- “C” for cleanliness



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## Microbial risks associated with feeding colostrum



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## Review Ig absorption in the calf

### Ig Absorption

- Large MW macromolecules absorbed from Jejunum and proximal ileum
- 1<sup>st</sup> 2 – 24 hours of life
- Variable efficiency
- Little selectivity in uptake
- Some selectivity in transfer through serosal side

Calf epithelium ~ 24 hours

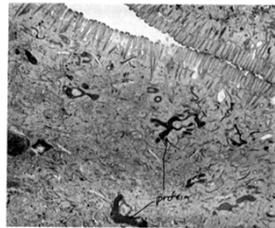


FIG. 4. 80 x. Micrograph of calf epithelium from a calf which had received colostrum prior to 6 h. Cells were sectioned longitudinally. Note arrangement of cellular granules near to the apical surface, typical of the cells responsible for IgG transfer.

Corley et al. 1977

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## Cessation of Ig absorption

- Cessation termed closure
- Differs by species
- In the calf
  - Not as diet dependent
  - Very variable onset – 6 – 24 hours
  - Precipitous decline in absorptive efficiency

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### Closure - One step process where uptake and transfer ceases

- Theories
  - Development of gastric and enzymatic function
  - Reduction in permeability of villus epithelial cells
  - Replacement by generation of cells incapable of pinocytosis

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### Replacement by generation of cells incapable of absorption.

- Micropinocytotic IgG transfer by newborn calf enterocytes
  - Existence of a receptor mediated transport system?
  - Relationship between apoptosis in cessation of Ig transfer

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### Apoptosis and IgG absorption in goats

Castro-Alonso et al. (2008)

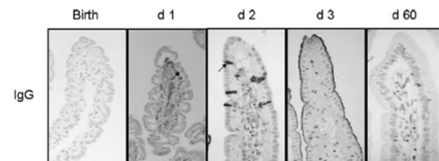
- 10 new born kids
- Fed colostrum – 2,000 mg IgG/kg body weight – 2X @ 2 and 14 h
- Sacrificed - birth – 60 d of age.
- Assessed for apoptotic cells and stained for IgG.



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### IgG staining in duodenum of goat kids harvested at .....

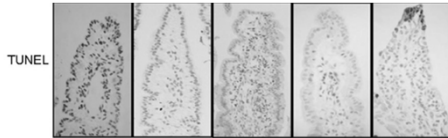


Birth - no IgG immunoreactivity  
 Day 1 – IgG internalized  
 Day 2 + IgG was bound to enterocytes but minimal internalization  
 Day 60 – No staining for IgG

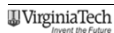
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**Terminal deoxynucleotidyl transferase nick end labeling (TUNEL) – identifies apoptotic cells**



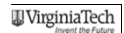
Birth – TUNEL staining at center of villus  
 Day 1 – staining is significantly reduced after day 1  
 Day 60 – only cells at villus terminus are staining



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**Conclusions**

- Relationship of IgG absorption with apoptosis in the intestine?
- Apoptosis is influenced by composition of colonizing intestinal bacteria in pigs (Willing and Van Kessel, 2007)
- How to delay apoptosis?



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Feeding heat-treated colostrum or unheated colostrum with two different bacterial concentrations to neonatal calves - Elizonda-Salazar and Heinrichs, 2009

- **Treatments**
  - Colostrum frozen - low bacteria
  - Colostrum pasteurized – 60°C / 30 min. then frozen
  - Colostrum stored at 20°C for 24 h then frozen
  - First feeding 3.8 liters / 68g IgG/liter - 1.5 – 2 h after birth with esophageal feeder.

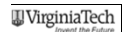


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**IgG (g/L) and SPC of colostrum Serum protein and IgG @24 h**

Item	Low bacteria	High bacteria	Heat – treated
IgG (g/L)	69.55	69.55	66.17
SPC x10 <sup>9</sup> /ml	9.332	40.738	.645
Serum protein @ 24h (g/L)	57	56.2	62.5
IgG @ 24 h (g/L)	20.2	20.1	26.7
AEA of IgG @ 24 h (%)	35.4	32.4	43.9

Heat treating improved AEA and IgG at 24 h.  
 High bacteria load was relatively low / less than 100,000 goal (McGuirk)



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## Role for microbial colonization and Ig absorption?

- Rate of intestinal cell production in the crypts
- Migration of cells up the villus and desquamation from the tips influenced by microbial colonization?
- Microbes may occupy binding sites on the apical plasma membrane.

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## Intestinal microflora and the absorptive surface

- Colostrum deprived calf receiving E. coli O55
- Exfoliation of microvilli
- Intracellular penetration of epithelial cells

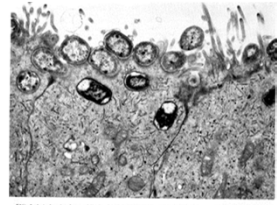


FIG. 3. Apical cells of several intestinal epithelial cells from an E. coli O55 calf which had received no colostrum. The microvilli were largely exfoliated at the apical E. coli attachment E. coli were also within the apical crypts (approximately 10,000x).

Corley et al. , 1977

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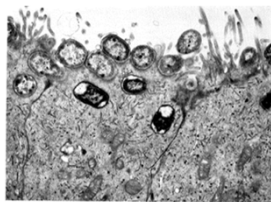


FIG. 2. Apical cells of several intestinal epithelial cells from an E. coli O55 calf which had received no colostrum. The microvilli were largely exfoliated at the apical E. coli attachment E. coli were also within the apical crypts (approximately 10,000x).

Colostrum deprived

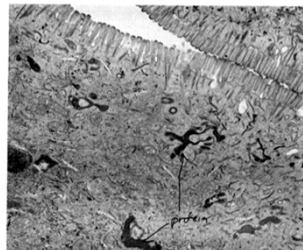


FIG. 4. Apical cells from a calf which had received colostrum prior to E. coli were exfoliated only. Thick aggregations of colostrum proteins were in the apical tubular system of the cells (approximately 10,000x).

Colostrum fed calf

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## How is colostrum managed on many dairies?



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## How is colostrum managed on dairies?



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## Field studies of colostrum quality

- Swan et al., 2007 –
  - 12 Minnesota and Wisconsin dairies
  - Median TPC –  $6.15 \times 10^8$
  - Varied from  $7 \times 10^7$  -  $> 10^9$
  - 93% of samples over goal of 100,000 TPC
- Poulsen et al., 2002
  - 82% of samples over 100,000 TPC

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## Critical control points for colostrum contamination



udder



Collection bucket

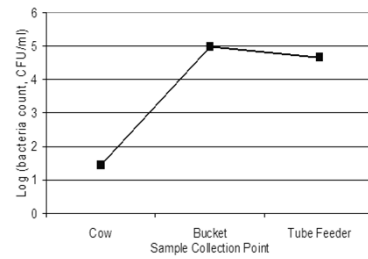


Esophageal feeder

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## Critical control points Stewart et al, 2005

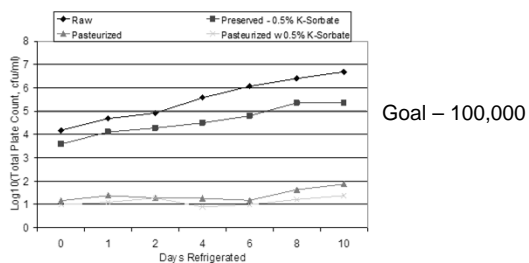


Samples from cow, bucket and tube feeder  
64% of samples collected within 20 min of harvest were  
<100,000 TPC

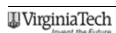
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### Growth during storage



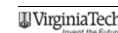
Goal – 100,000



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### Batch pasteurization of colostrum

- 60° – 60 minutes
- Batch pasteurization
- Higher temperatures = cottage cheese



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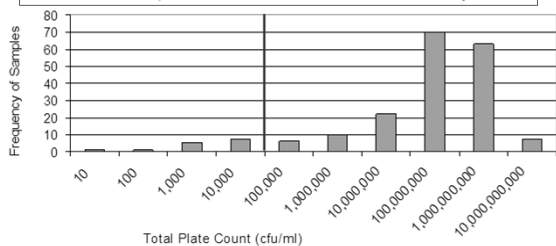
### Total Bacteria Counts in Minnesota Colostrum

(Swan et al. 2007. JDSci. 90)

Median TPC = 615 million cfu/ml (73 to 104 billion)

93% of samples > 100,000 cfu/ml TPC

“We are feeding ‘fat-laden’ manure” Rob Trembley, 2006



From - S. Godden



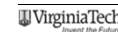
### Raw vs. pasteurized colostrum

Johnson et al., 2007

Parameter	Raw	Pasteurized
IgG – mg/ml	72.6	67.3
Total plate count	46,000	872

Colostrum	Serum protein	IgG concentration	AEA %
Raw	5.9 g/dl	18.1 mg/ml	35.6
Pasteurized	6.3g/dl	22.34 mg/ml	26.1

Correct table in proceedings



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## Recent UMN Field Study

M. Donahue, S. Godden

- 1,000 calves / 6 herds
  - ½ fed raw and ½ fed heat-treated colostrum
- Colostrum total plate count and serum IgG – negative
- Colostrum IgG concentration – positive effect
- Heat treatment – positive – independent of Total plate count

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## 30 years later - lots of common sense



Did some "West Virginia" rub off?  
I hope so!!!!

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## Common Sense and Colostrum

- Clean cows – infection and sanitation
- Don't pool colostrum
- Milk fresh cows first and process colostrum immediately.
- If refrigerating – cool rapidly in ice bath
- Clean buckets, bottles and esophageal feeders
- If herd size and herd infection status warrants – pasteurize or use replacer
- Avoid use of probiotics for 24 hours

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## Thanks, Carl!



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