#### **Young Scholar Presentation**

Effects of monoglycerides on intestinal morphology and immune responses of weanling pigs experimentally infected with a pathogenic *E. coli* 

Sangwoo Park<sup>1</sup>, Shuhan Sun<sup>1</sup>, Kwangwook Kim<sup>1</sup>, Adebayo O. Sokale<sup>2</sup>, Adriana Barri<sup>3</sup>, and Yanhong Liu<sup>1</sup>

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**Presentation #136** 





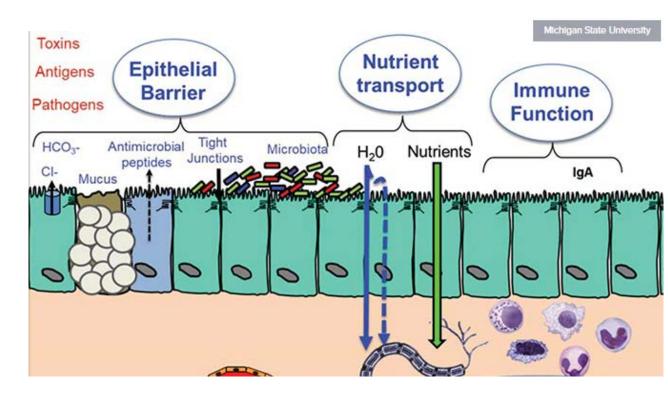
- Gut health and post-weaning diarrhea in pigs
- Monoglycerides
- Experimental design
- Results and conclusions



# **Gut health of weaning pigs**

#### Intestinal functions

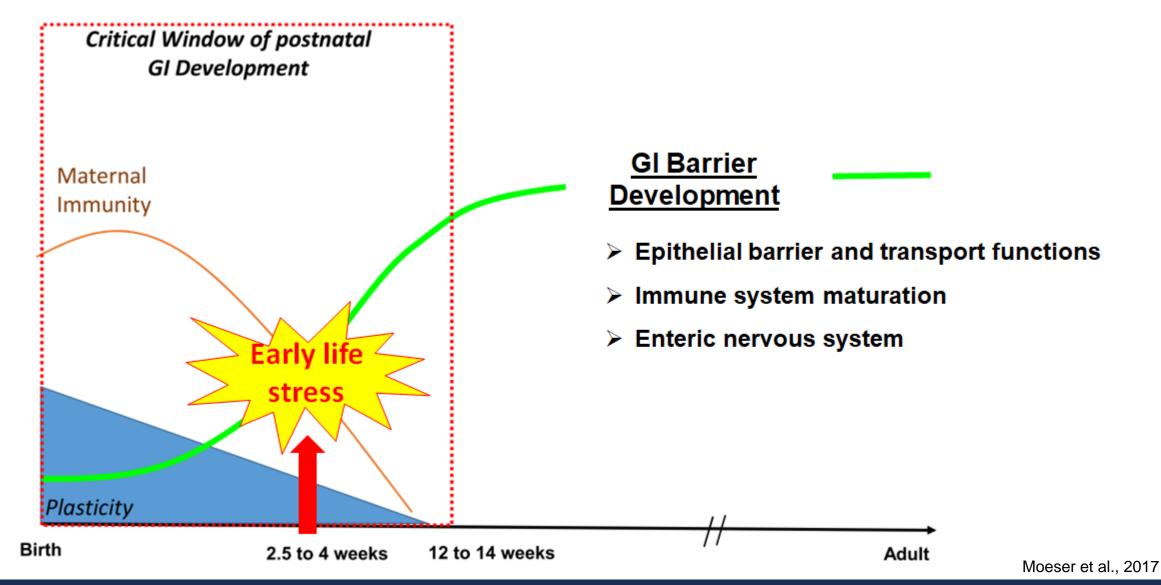
- Digest and absorb nutrients
- Protect the host
- Growth and health of pigs
- Early-life stress
  - Harmful impacts on gut health



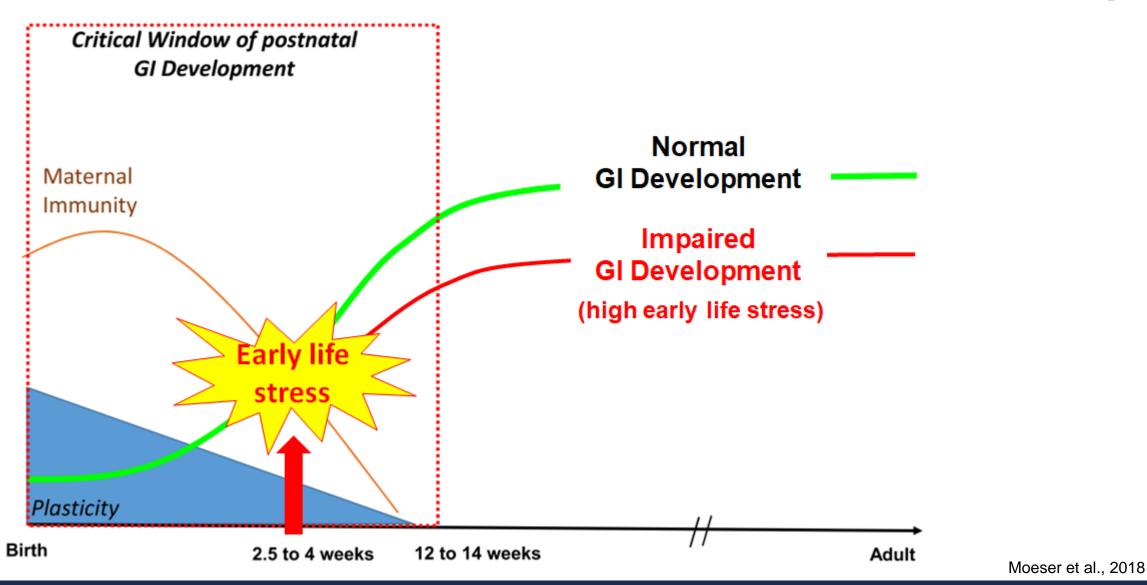
Moeser et al., 2018



#### **Gastrointestinal (GI) function development in pig**



#### **Gastrointestinal (GI) function development in pig**



# Weaning stress

- Multifactorial issue
  - > Nutritional, physiological, and environmental challenges
- Stressor-induced changes
  - Reduced appetite
  - Induced intestinal dysfunctions
  - Increased exposure and risk to pathogens
  - Post-weaning diarrhea (PWD)



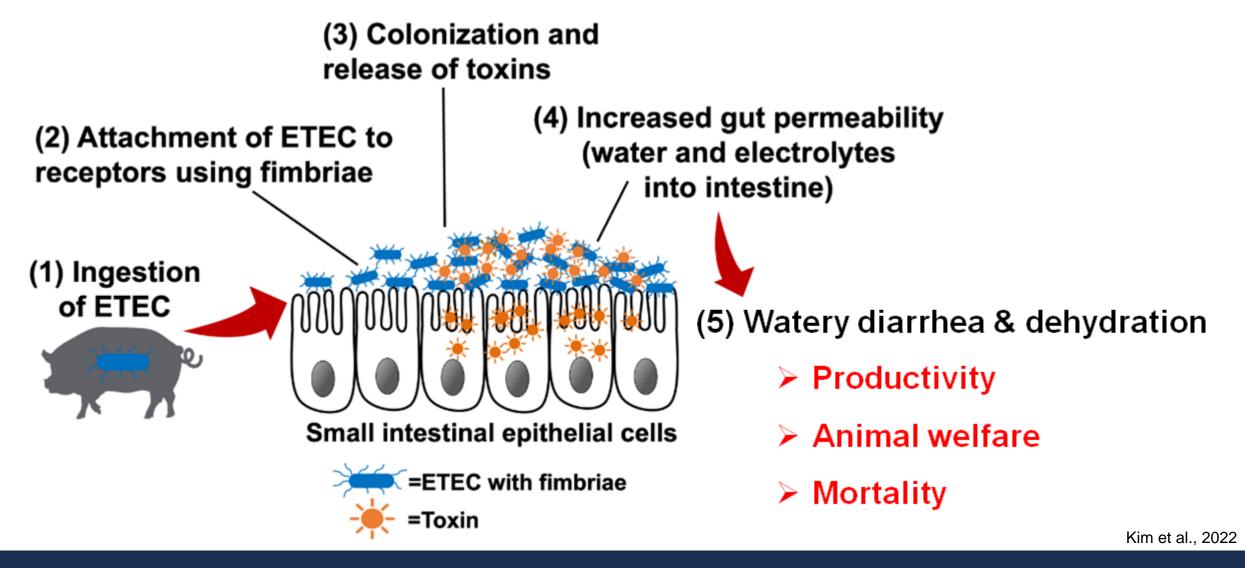
# **Post-weaning diarrhea (PWD)**

- Gastrointestinal disease
  - Economic losses
- Enterotoxigenic Escherichia coli (ETEC)
  - ➢ F4 (K88) and F18 E. coli
  - Enterotoxins
    - ✓ Secretory diarrhea





### Post-weaning E. coli diarrhea

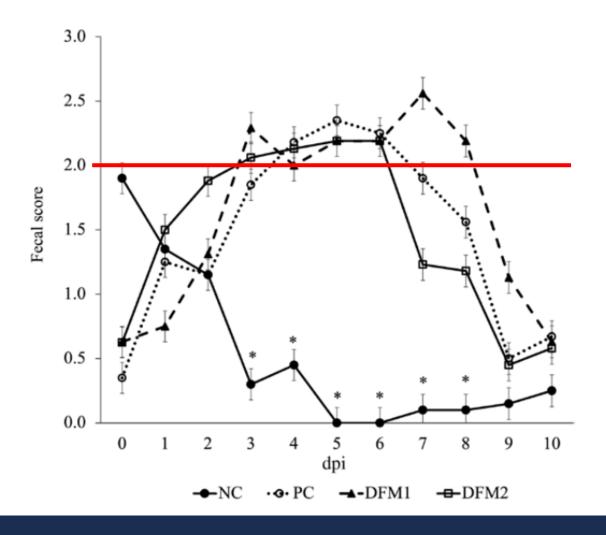




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doi:10.1093/jas/skaa113 Advance Access publication April 17, 2020 Received: 22 October 2019 and Accepted: 16 April 2020 Non Ruminant Nutrition

#### **E. coli challenge and diarrhea**



#### NON RUMINANT NUTRITION

Effects of an F18 enterotoxigenic Escherichia coli challenge on growth performance, immunological status, and gastrointestinal structure of weaned pigs and the potential protective effect of direct-fed microbial blends

Spenser L. Becker, `Qingyun Li, `Eric R. Burrough,† Danielle Kenne,† Orhan Sahin,† Stacie A. Gould,` and John F. Patience<sup>\*,1</sup>

**Figure 1.** Effects of treatment on the daily fecal score of pigs challenged with F18 ETEC. NC (n = 10); PC (n = 9); DFM1 = PC + direct-fed microbial 1 (n = 8; three strains of Bacillus amyloliquefaciens; 7.5 × 10<sup>5</sup> cfu/g of feed); DFM2 = PC + direct-fed microbial 2 (n = 7; two strains of B. amyloliquefaciens and one strain of Bacillus subtilis; 1.5 × 10<sup>5</sup> cfu/g of feed). Supplementation rates were based on manufacturer's recommendations (Danisco Animal Nutrition). P (NC vs. PC; day postinoculation (dpi) 3) < 0.001, P (PC vs. DFM1, DFM2; dpi 3) > 0.10, P (all treatments; dpi 10) > 0.10.

\*Fecal score: 0 = solid; 1 = semi-solid; 2 = semi-liquid; 3 = liquid (≥ 2 was considered diarrhea)

Journal of Animal Science, 2022, 100, 1–14 https://doi.org/10.1093/jas/skac353 Advance access publication 22 October 2022 Non Ruminant Nutrition

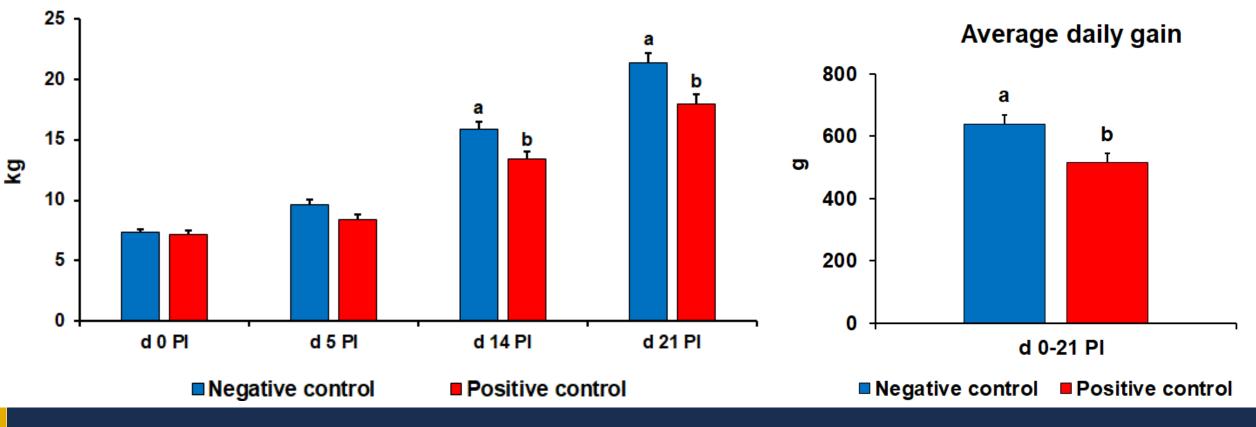


#### **E. coli challenge and performance**

Body weight

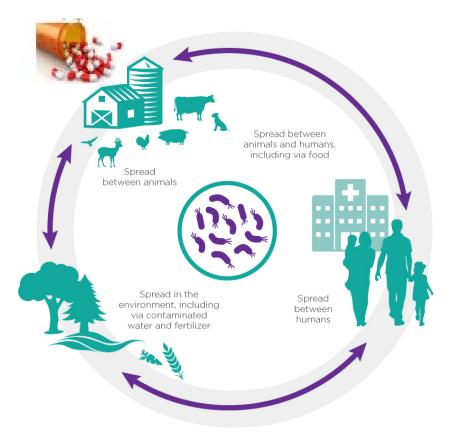
Dietary supplementation of botanical blends enhanced performance and disease resistance of weaned pigs experimentally infected with enterotoxigenic *Escherichia coli* F18

Braden T. Wong,<sup>†</sup> Sangwoo Park,<sup>†,1</sup> Lauren Kovanda,<sup>†</sup> Yijie He,<sup>†</sup> Kwangwook Kim,<sup>†,®</sup> Shiyu Xu,<sup>†</sup> Christopher Lingga,<sup>†</sup> Monika Hejna,<sup>†,‡</sup> Emma Wall,<sup>II,2</sup> Ravichandran Sripathy,<sup>II</sup> Xunde Li,<sup>§</sup> and Yanhong Liu<sup>†,3</sup>



### Antibiotics and pharmacological dose ZnO

- Prevent and treat PWD
  - Antimicrobial effects
  - Nutrients availability
- Public health risk and concern
  - Antimicrobial resistance
  - Environmental transmission
    - Prohibition of AGPs (Jan 2017, FDA)
    - Prohibition of pharmacological dose ZnO (June 2022, EU)



Public Health Agency of Canada, 2017



Ester bond

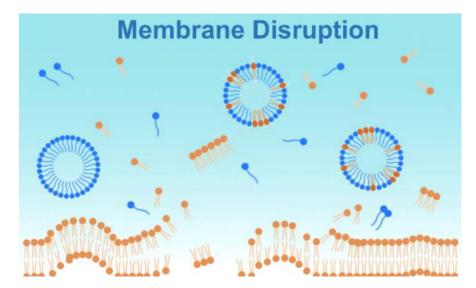
# **Monoglycerides (MGs)**

#### Glycerol linked to fatty acid (esterification)

Short chain and medium chain fatty acids

#### Natural compound

- Used in food processing and production
- Amphiphilic nature
  - > Hydrophobic & hydrophilic
  - Antimicrobial activity

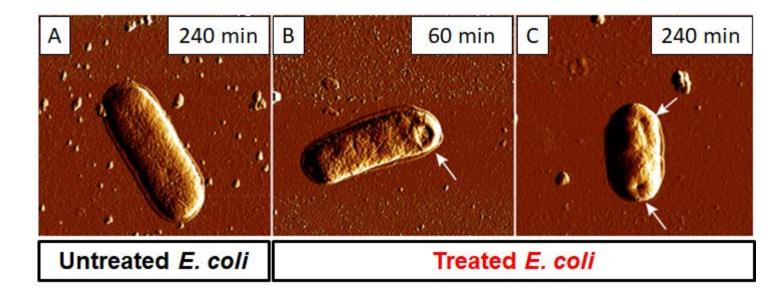


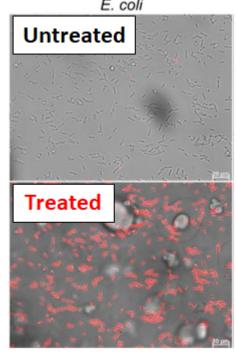
Jackman et al., 2022



# Antimicrobial effects of MGs (in vitro)

#### Incorporate into the lipid membrane of microorganisms and change the permeability





**Propidium iodide staining** 

Hyldgaard et al., 2012; Joshua et al., 2020



#### Ester bond

R

HO

# **Additional benefits of MGs**

#### Strong covalent bond

	Organic acid	Monoglycerides
Antimicrobial	$\checkmark$	$\checkmark$
Non-corrosive	-	$\checkmark$
Non-volatile	-	$\checkmark$
Heat stable	-	$\checkmark$
Neutral taste odor	-	$\checkmark$
pH-independent	-	$\checkmark$

#### Easy to handle and use

FRAmelco, 2017





#### Effects of a mixture of monoglycerides on weaned pigs experimentally infected with a pathogenic Escherichia coli (E. coli) F18

- ✓ Growth performance
- ✓ Diarrhea
- ✓ Intestinal health
- ✓ Immune responses



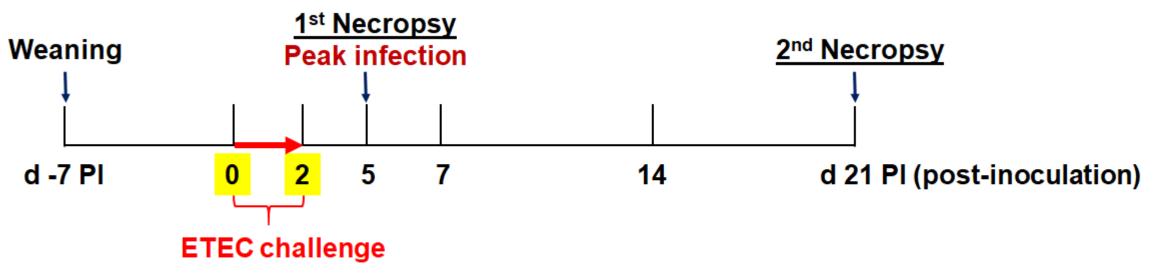
# Animals & experimental design

#### ✤ Animals

- $\geq$  60 weaned pigs (initial BW = 6.5 ± 0.74 kg; 21 d old)
  - ✓ Individual house (15 replications/treatment)
- ✤ 4 dietary treatments
  - Corn-soybean meal-based diet (Control)
  - Control + 0.3% monoglycerides
  - Control + 3000 ppm zinc oxide (ZnO)
  - Control + 50 mg/kg of antibiotic (carbadox)
- 2-phase feeding (2 weeks/phase; overall 4 weeks)



### **Timeline and data collection (I)**

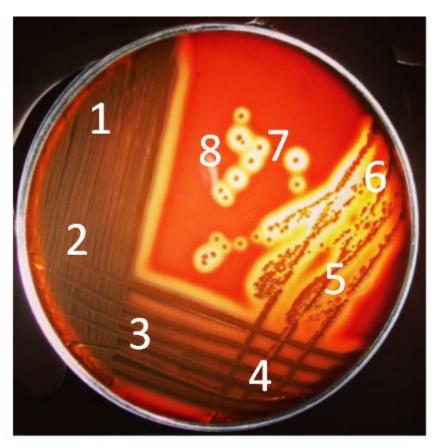


All pigs were orally inoculated with pathogenic *E. coli* F18 (10<sup>10</sup> CFU/dose)

- Daily fecal scores
  - $\succ$  Score 1 to 5 = firm feces to watery diarrhea
- Percentage of β-hemolytic coliforms in feces
- Growth performance (ADG, ADFI, and G:F)



# <u>β-hemolytic coliforms</u> (feces)

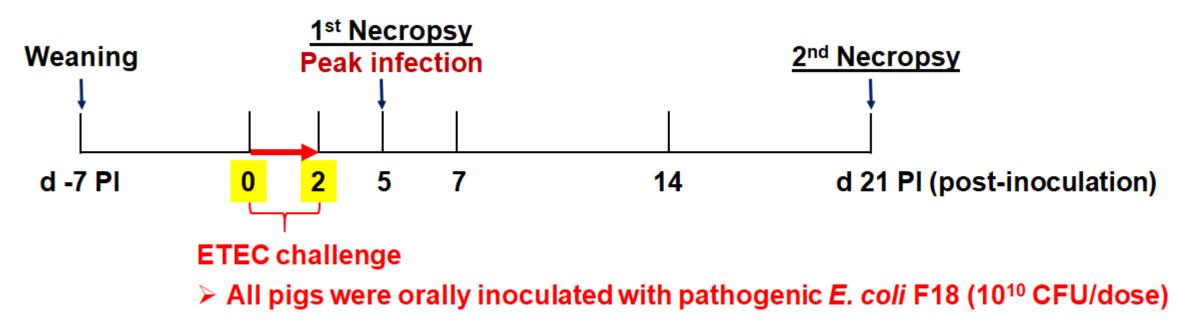


Columbia blood agar (β-hemolytic coliforms)



MacConkey agar (Lactose-fermenting bacteria)

### **Timeline and data collection (II)**



Serum acute phase protein level (d 0, 2, 5, and 21 PI)

C-reactive protein and haptoglobin

✤ Intestinal morphology and immune-related gene expression
➢ d 5 PI (6 pigs/treatment) and d 21 PI (9 pigs/treatment)



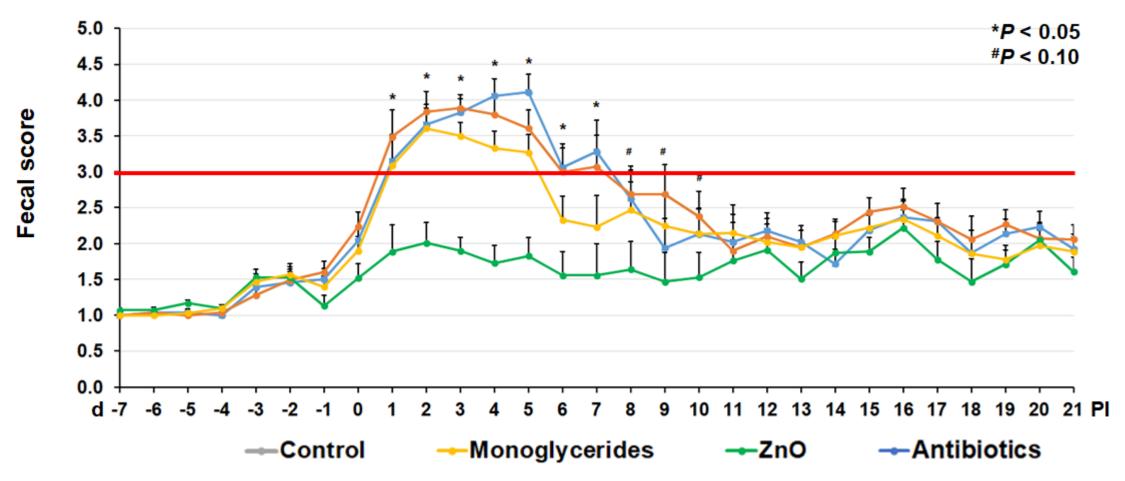
# **Statistical analysis**

#### PROC MIXED of SAS

- Randomized complete block design (block: BW)
- Experimental unit: pig
- Fixed effect: dietary treatment
- Chi-square test
  - Frequency of diarrhea

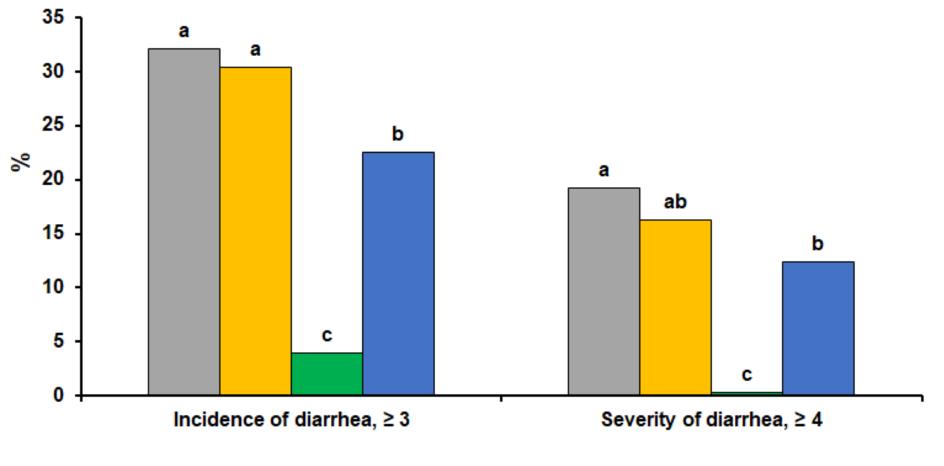


### **Daily fecal score**



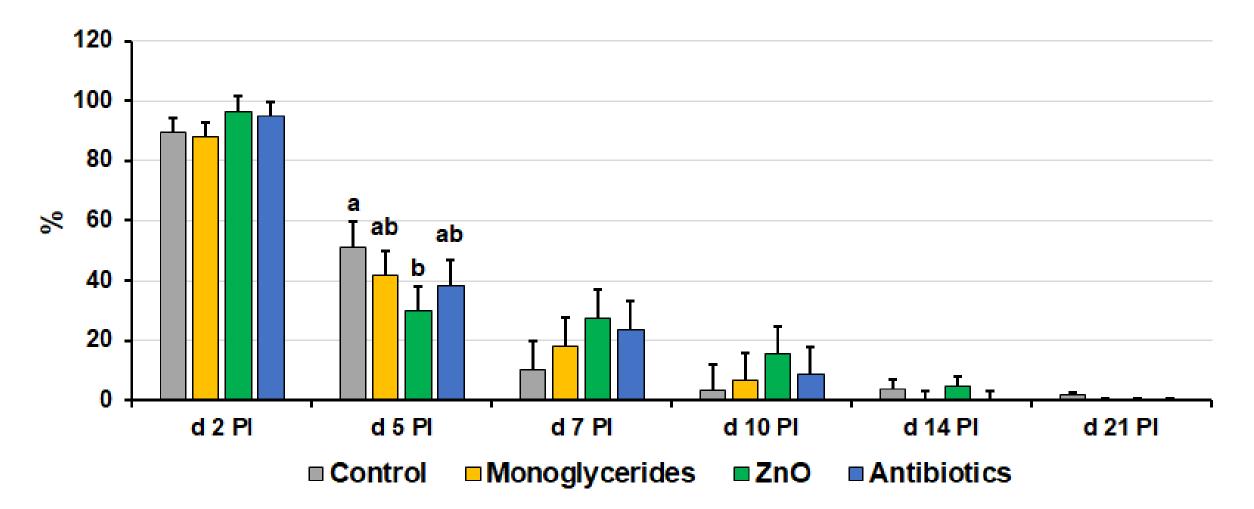
\*Fecal score = 1, firm feces; 2, moist feces; 3, mild diarrhea; 4, severe diarrhea; 5, watery diarrhea

# Frequency of diarrhea (overall)



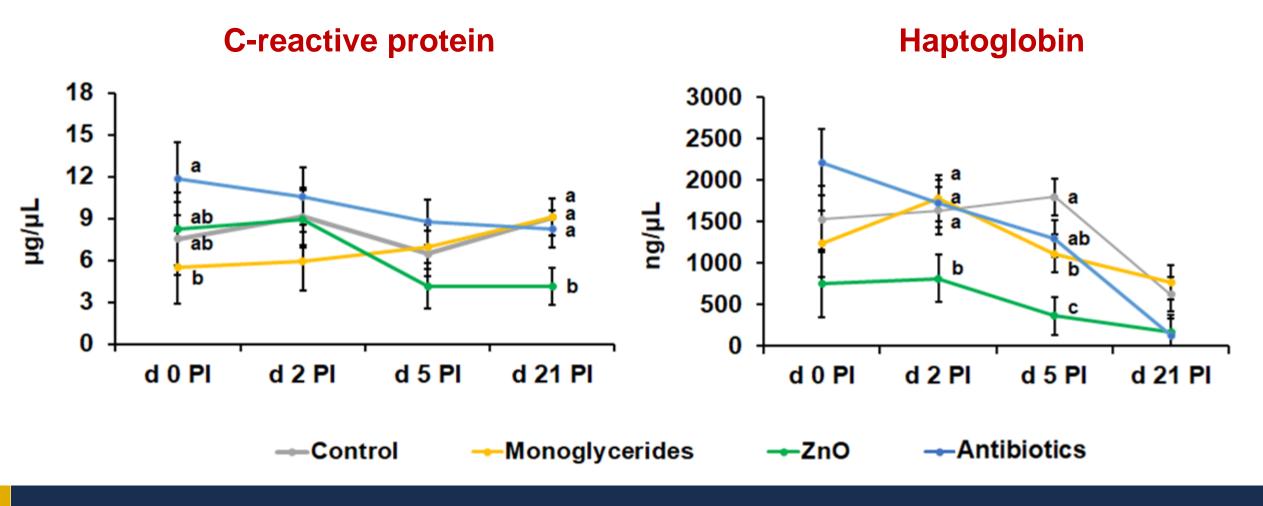
■ Control ■ Monoglycerides ■ ZnO ■ Antibiotics

# **β-hemolytic coliforms (feces)**

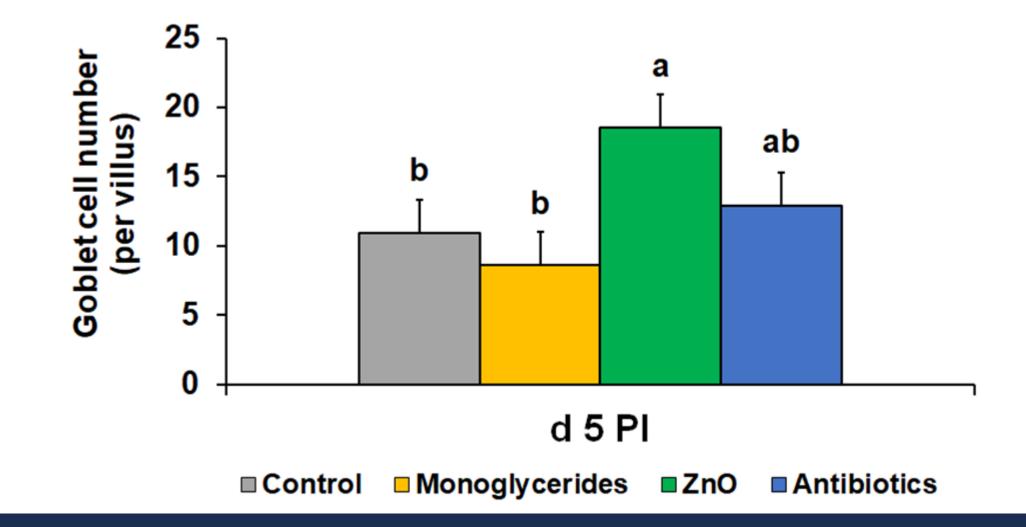




### Acute phase proteins (serum)



### **Duodenum goblet cell number**

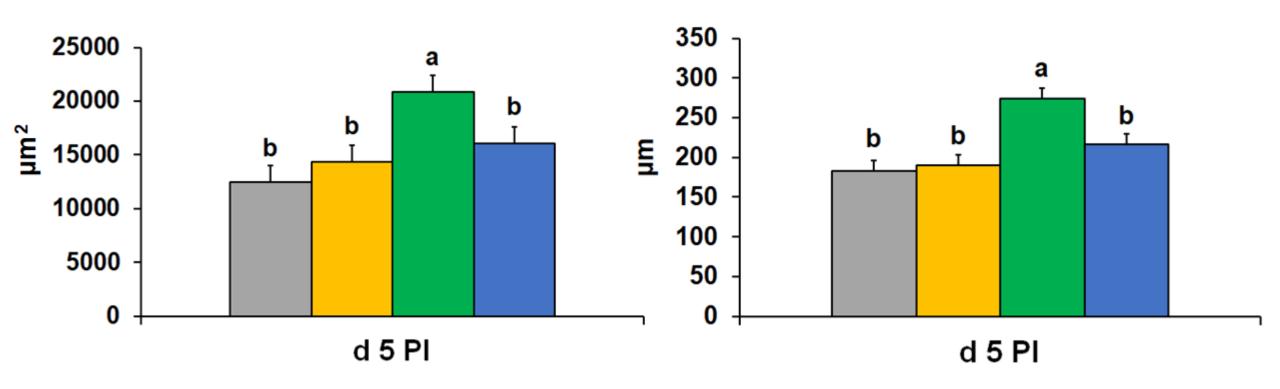




### **Duodenum villi area and height**

Villi area

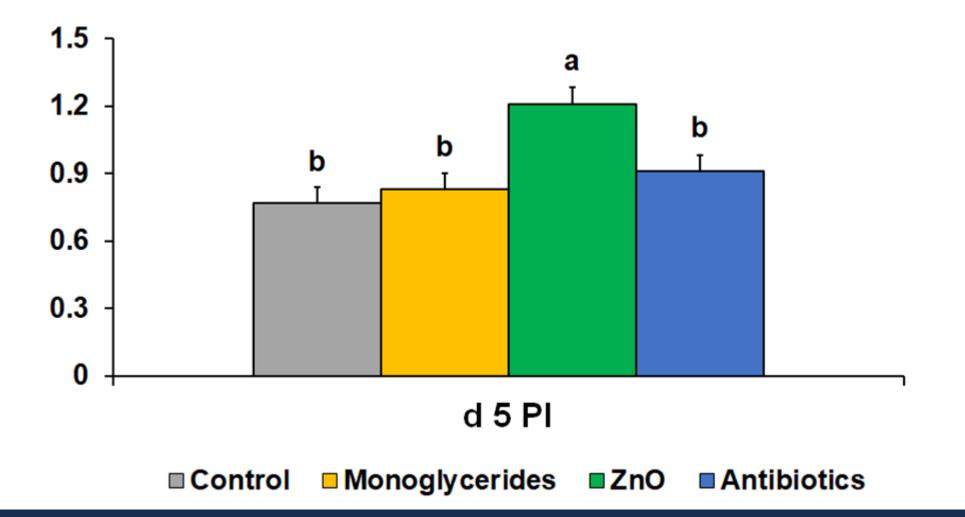
Villi height



■ Control ■ Monoglycerides ■ ZnO ■ Antibiotics



### **Duodenum VH:CD**

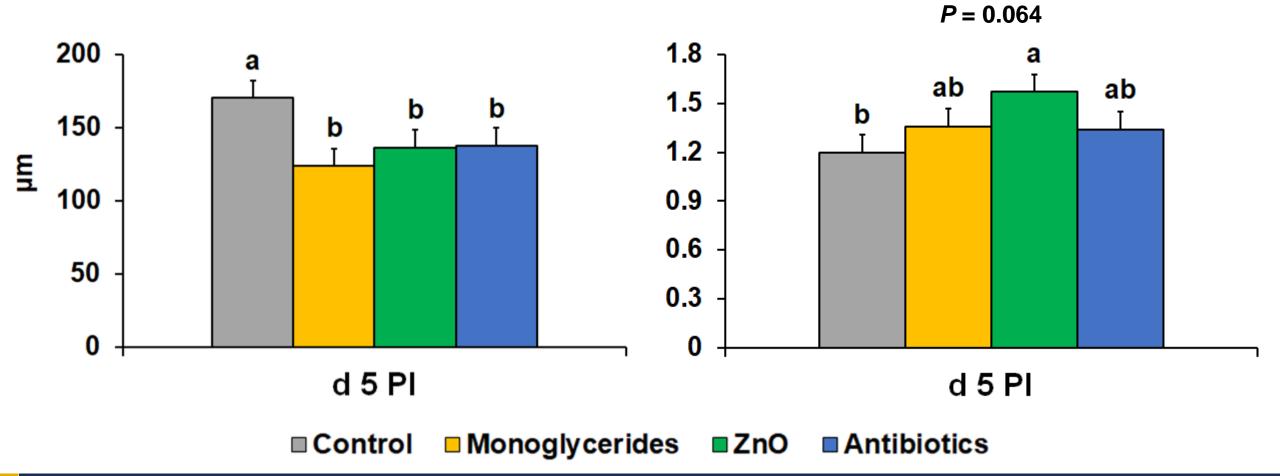




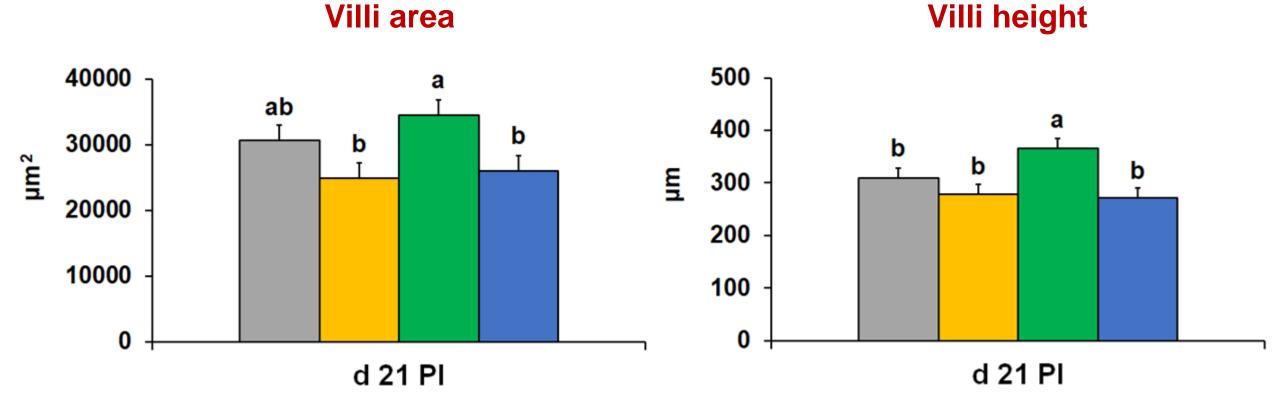
# **Ileum CD & VH:CD**

Crypt depth



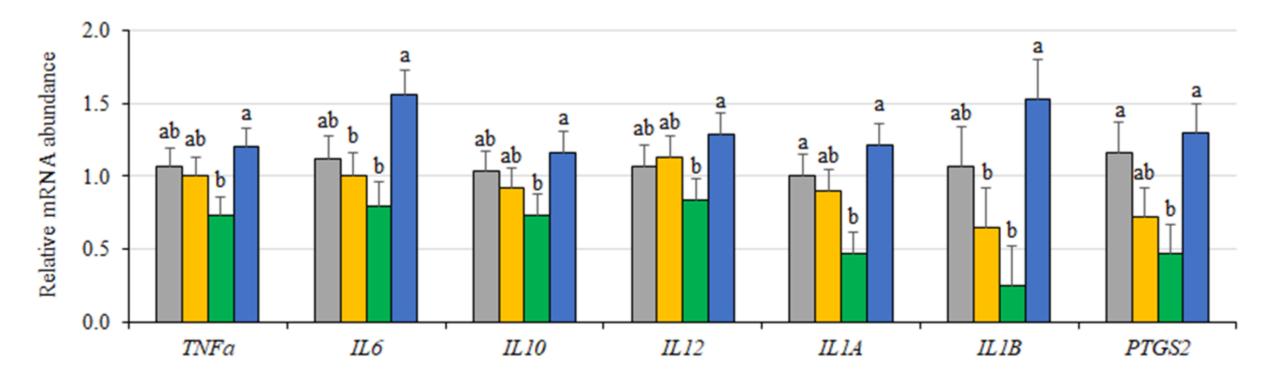


### **Duodenum villi area and height (d 21 PI)**



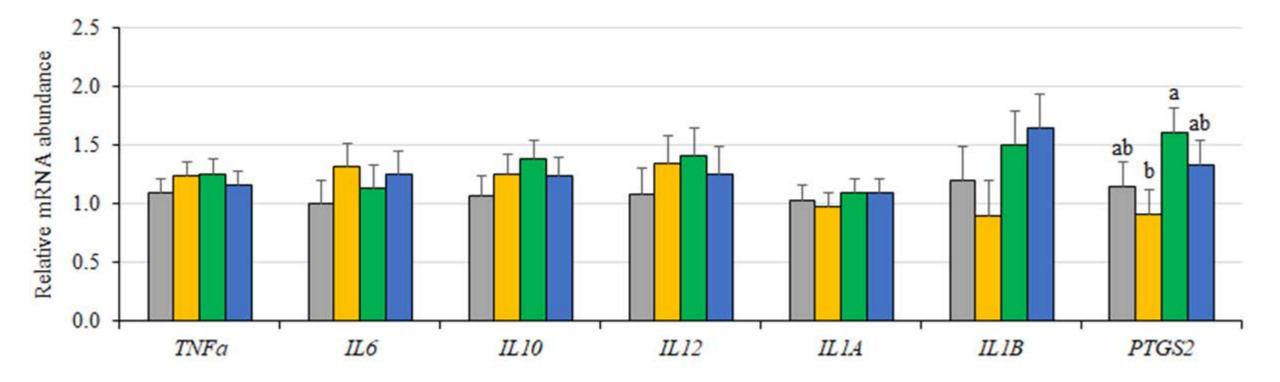
■ Control ■ Monoglycerides ■ ZnO ■ Antibiotics

### **Ileum mucosa gene expression (d 5 PI)**



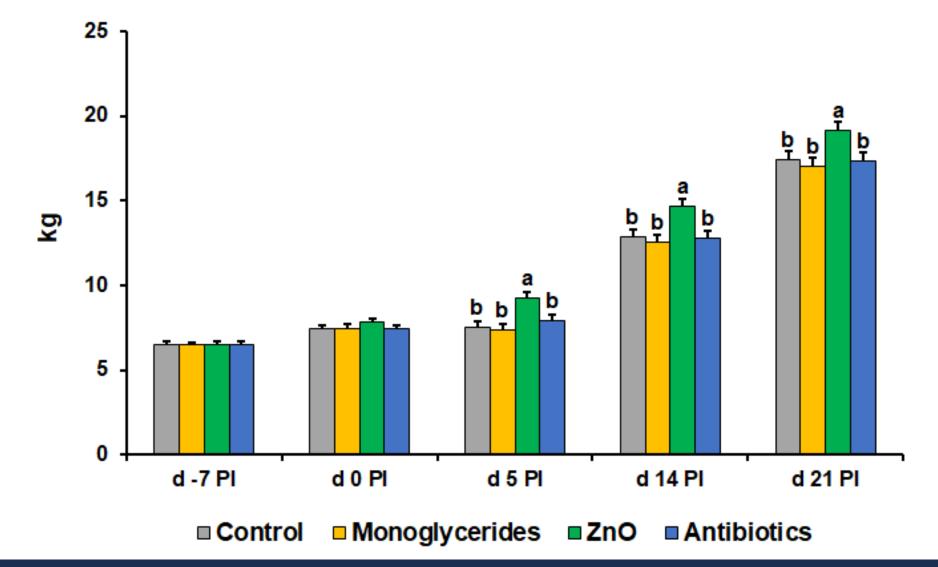
■ Control ■ Monoglycerides ■ ZnO ■ Antibiotics

### **Ileum mucosa gene expression (d 21 PI)**

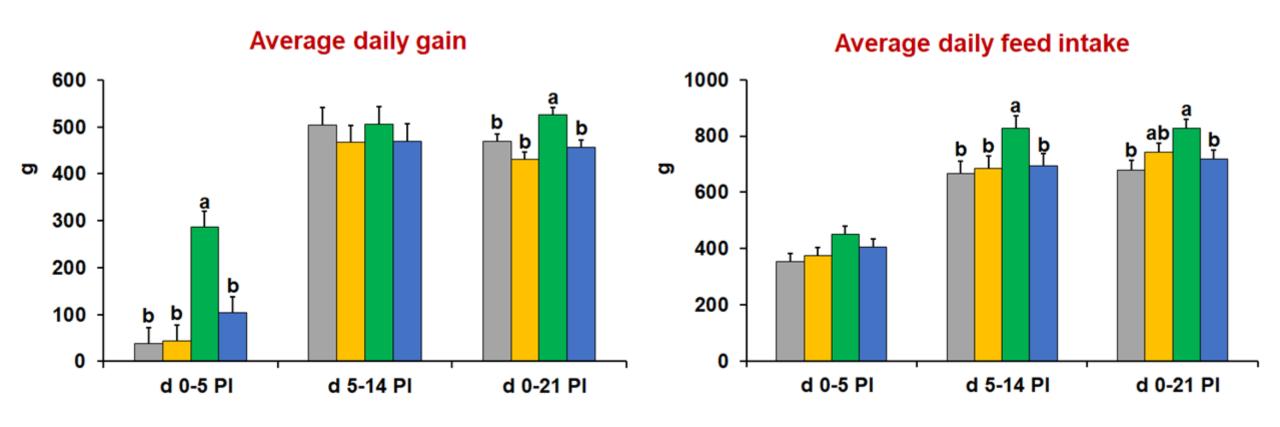


Control Monoglycerides ZnO Antibiotics

# **Body weight**



### **Growth performance**



■ Control ■ Monoglycerides ■ ZnO ■ Antibiotics

- Monoglycerides supplementation
  - Reduce the diarrhea severity
  - > Have a positive effect on the intestinal morphology
  - > Modify the intestinal and systemic inflammation
  - of weaned pigs infected with ETEC F18



# **Acknowledgements**

- Comparative Animal Nutrition & Physiology Laboratory
- BASF Corporation



https://animalnutr-ansci.faculty.ucdavis.edu/







### **Greatly appreciate your attention!**



