

Effects of *Bacillus subtilis* probiotics on growth performance, diarrhea, and fecal β -hemolytic coliforms of weaned pigs experimentally infected with an enterotoxigenic *Escherichia coli*

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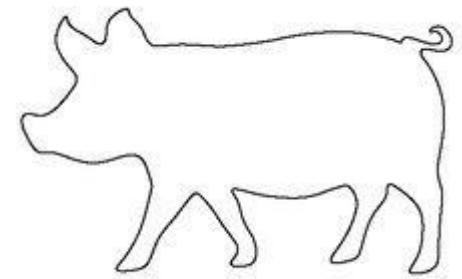
Introduction

☐ Weaning stress

- Separation from the sow and littermates
- Different physical environment
- Sow milk to solid food

☐ Intestinal structure and function

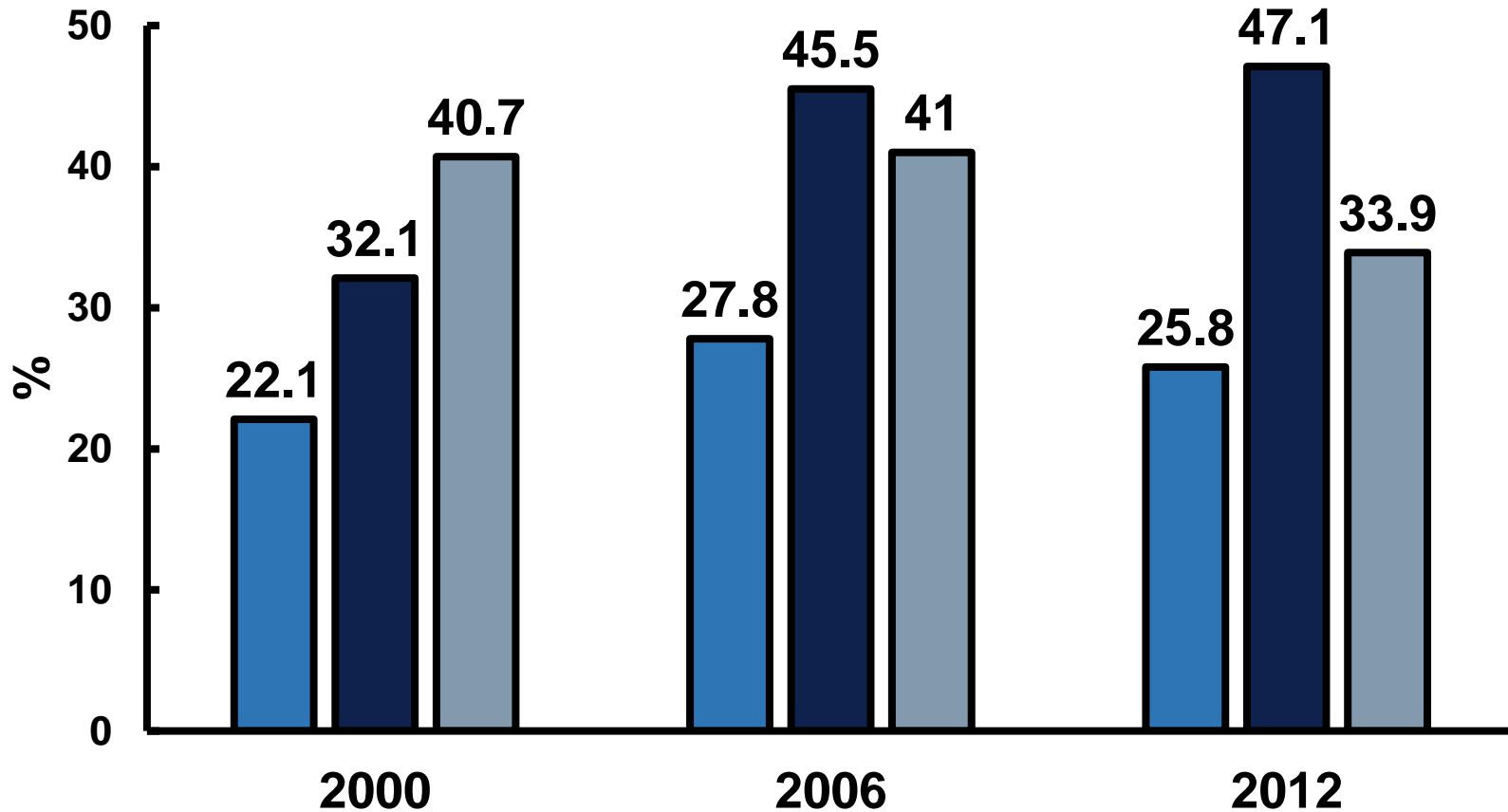
☐ **Decreased** growth performance



(Pluske et al., 1997)

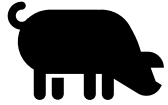
Post-weaning diarrhea by *E. coli* in US

■ Small (< 2000) ■ Medium (2,000 to 4,999) ■ Large (>5000)

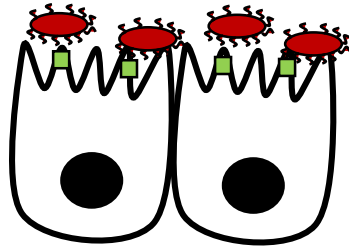


(USDA NAHMS studies, 2000; 2006; 2012)

Pathogenesis of *E.coli*

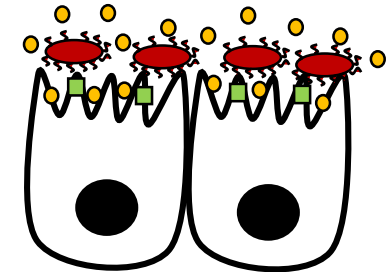
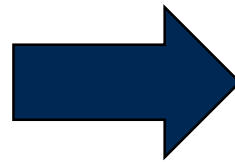


Ingestion of *E.coli*



Attachment of the *E. coli* to microvilli

Production/Economic loss



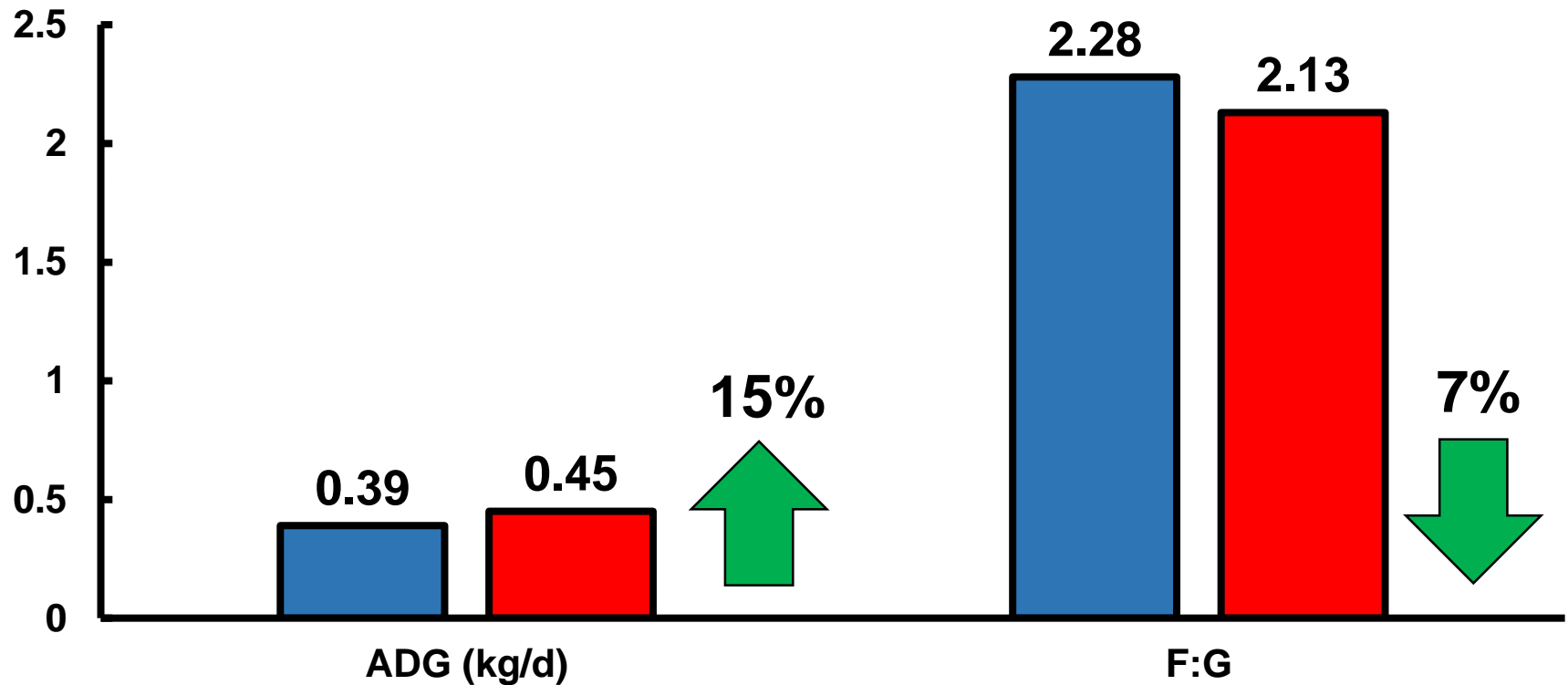
Production of enterotoxins

Diarrhea
Death



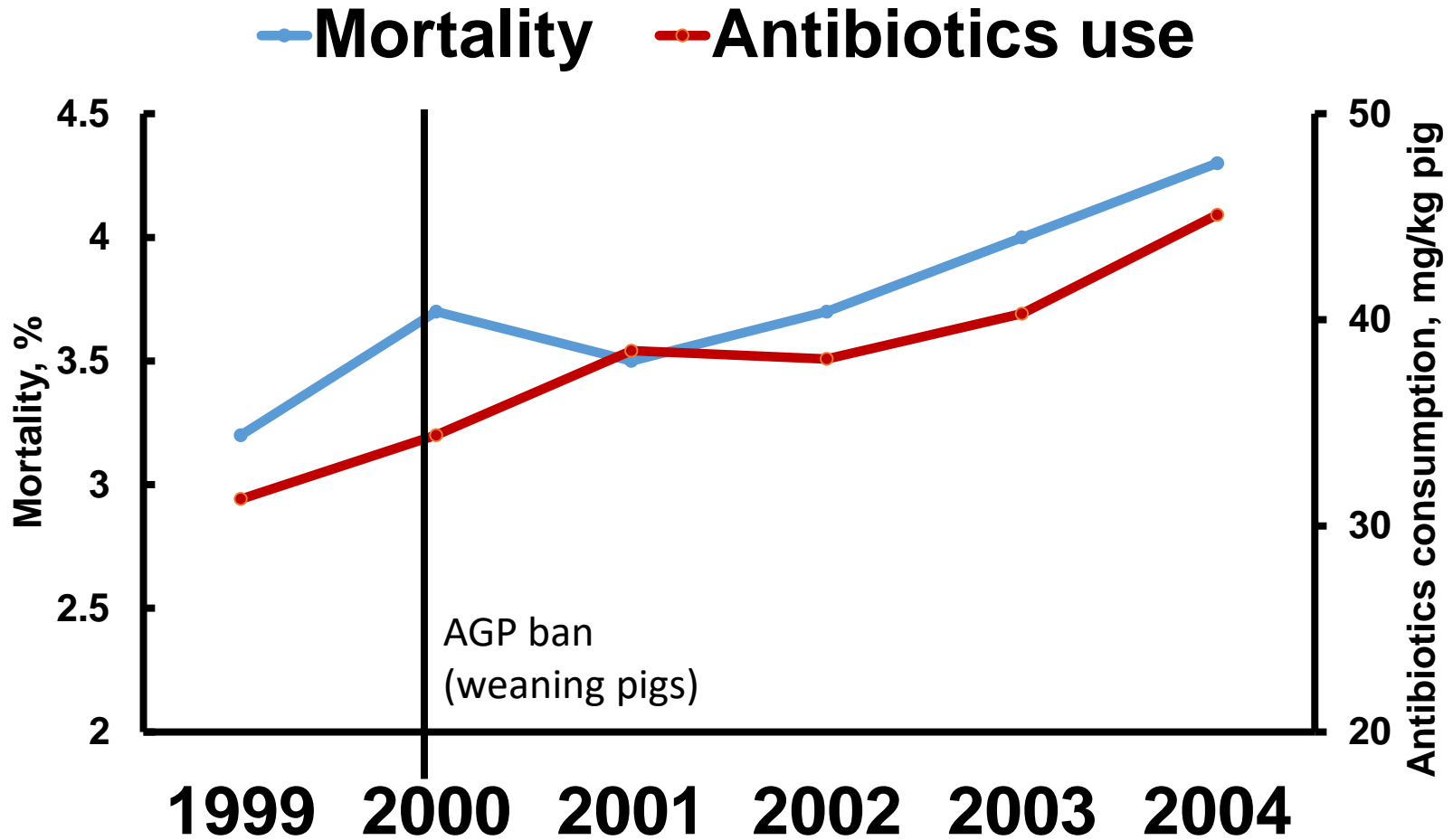
Antibiotics growth promoter

■ CON ■ ABX



(Cromwell, 2002)

AGP ban in Denmark



(Aarestrup et al., 2009)

Antibiotics alternatives - probiotics

- ❑ Live microorganisms confer a health benefit
- ❑ Modes of action
 - Competition for adhesion sites
 - Direct antagonism
 - Lactic acids – lowering pH
 - Modulation of immune system
 - Anti/pro-inflammatory cytokines
 - Immune cells population

(Kenny et al., 2011)

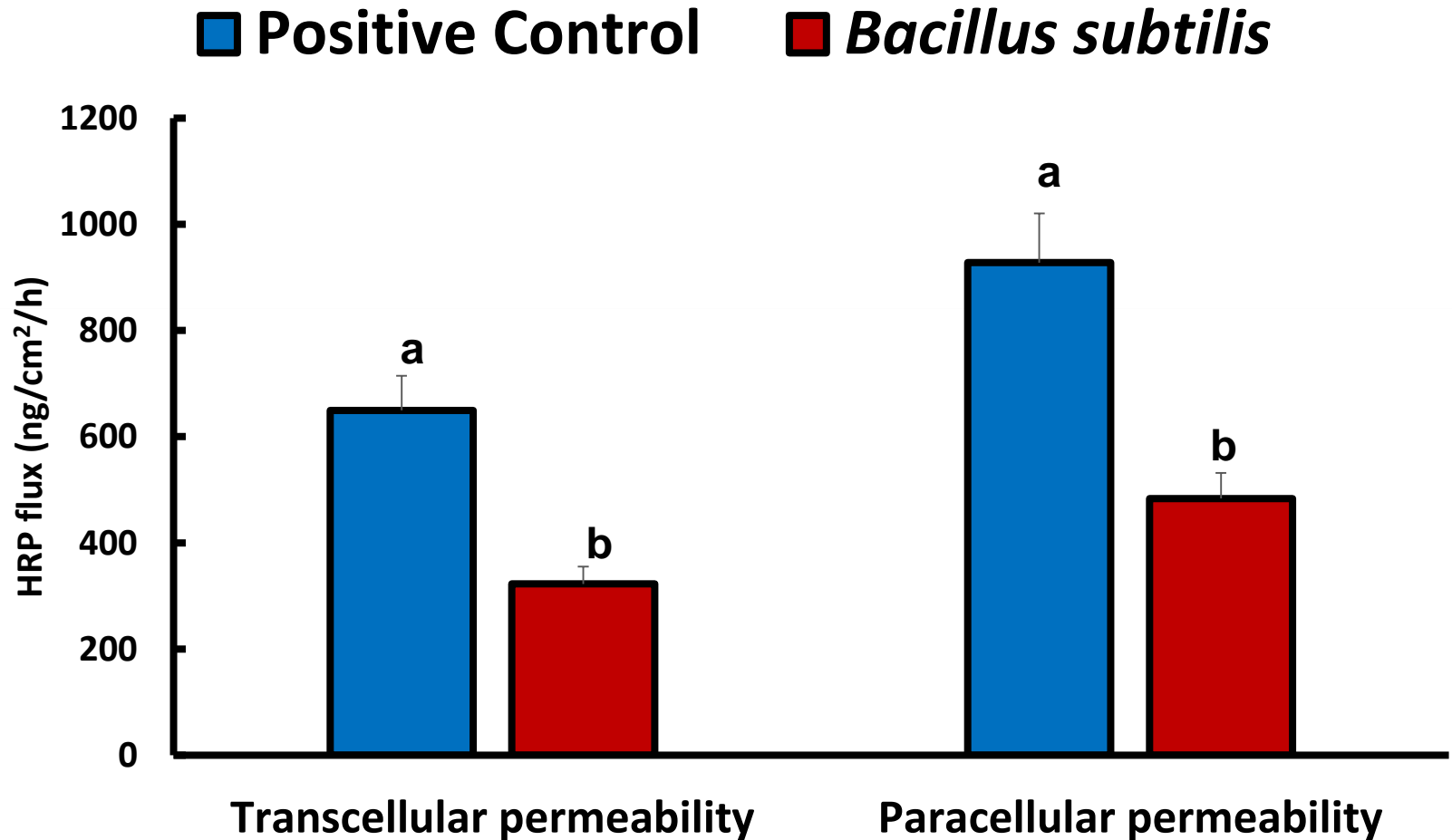
Bacillus subtilis

- Spore-forming *Bacillus spp.*
 - Resistance to harsh environment
 - Long term storage

- Favorable results
 - Reduced incidence of diarrhea
 - Improved intestinal epithelial barrier integrity

(*Bhandari et al., 2008; Yang et al., 2016*)

Bacillus subtilis reduce gut permeability



Objectives

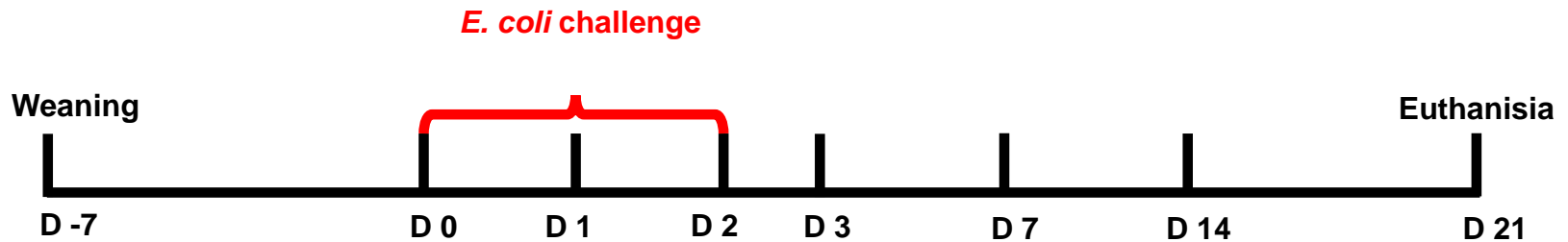
- To investigate the effects of supplementation of *Bacillus spp.* to weaned pigs experimentally infected with an enterotoxigenic F-18 *E. coli*
 - Growth performance
 - Systemic immunity
 - Intestinal health

Materials and Methods

- ❑ 48 pigs: 21-d of age, BW = 6.17 ± 0.36 kg
- ❑ Individual pens
- ❑ 4 dietary treatments
 - Negative Control (**NC**)
 - Positive Control (**PC**)
 - CON + Carbadox (50 mg/kg*) (**AGP**)
 - CON + *Bacillus subtilis* (500 mg/kg*) (**PRO**)
- ❑ 12 replicates/treatment

*500 mg/kg = 1×10^9 cfu/kg diet

Materials and Methods



- Genotyping: F18 receptor
- Oral inoculation of F18 *E. coli*
 - LT, STb, SLT-2
 - 10^{10} cfu/dose with 3 doses
- Fecal samples
 - β -hemolytic coliforms

Materials and Methods

Growth performance

- Body weight
- Average daily gain (ADG)
- Average daily feed intake (ADFI)
- Feed to gain ratio (FG)

Daily diarrhea scores

jejunum and ileum

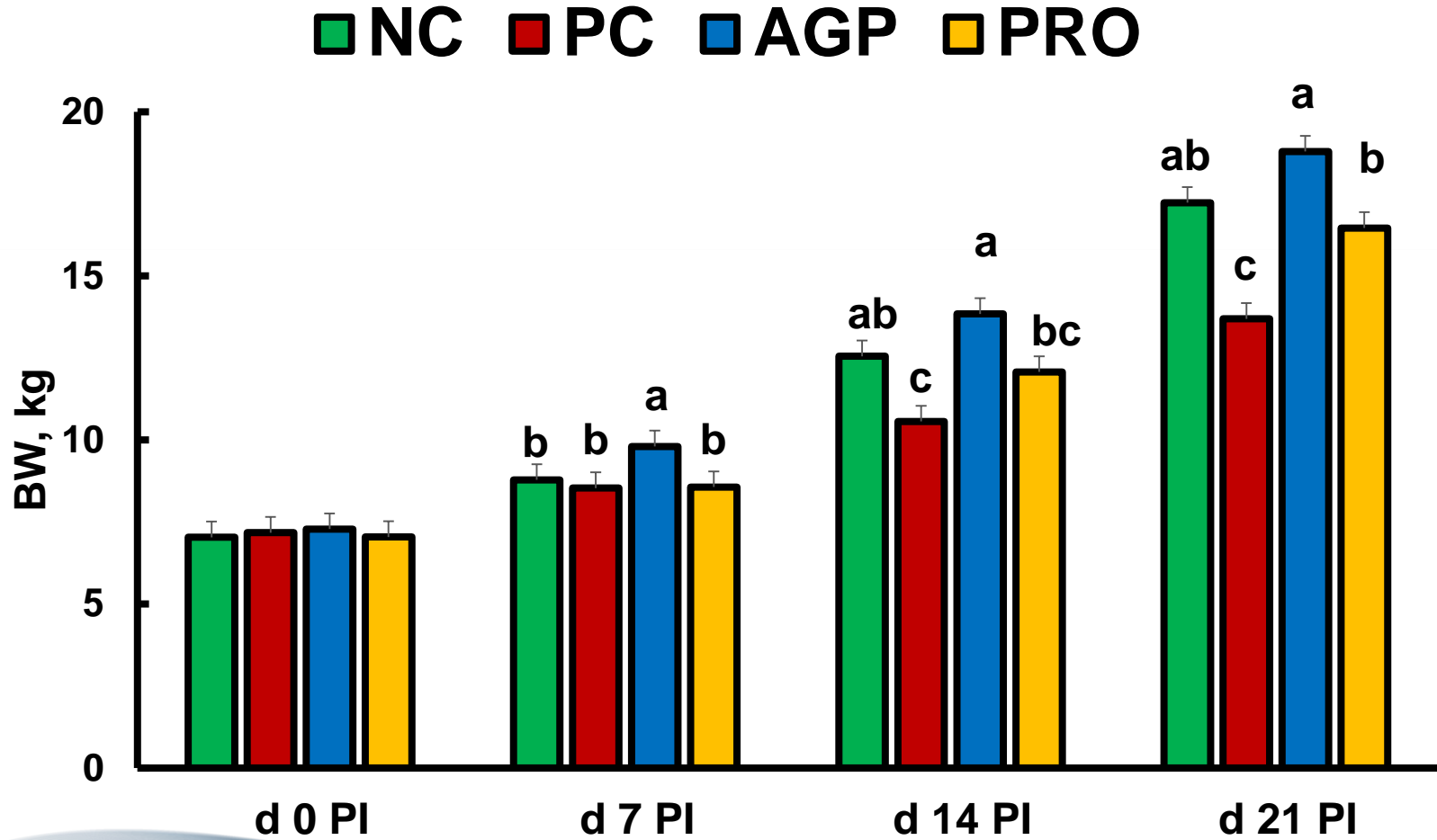
- Jejunal tight junction protein mRNA expression
- Ileal pro-inflammatory cytokine mRNA expression

Statistical analysis

- ❑ Mixed Procedure of SAS
- ❑ Randomized complete block (BW x Sex)
- ❑ Fixed effect: diet
- ❑ Random effect: block
- ❑ Significance at $P \leq 0.05$ and tendency at $P \leq 0.10$

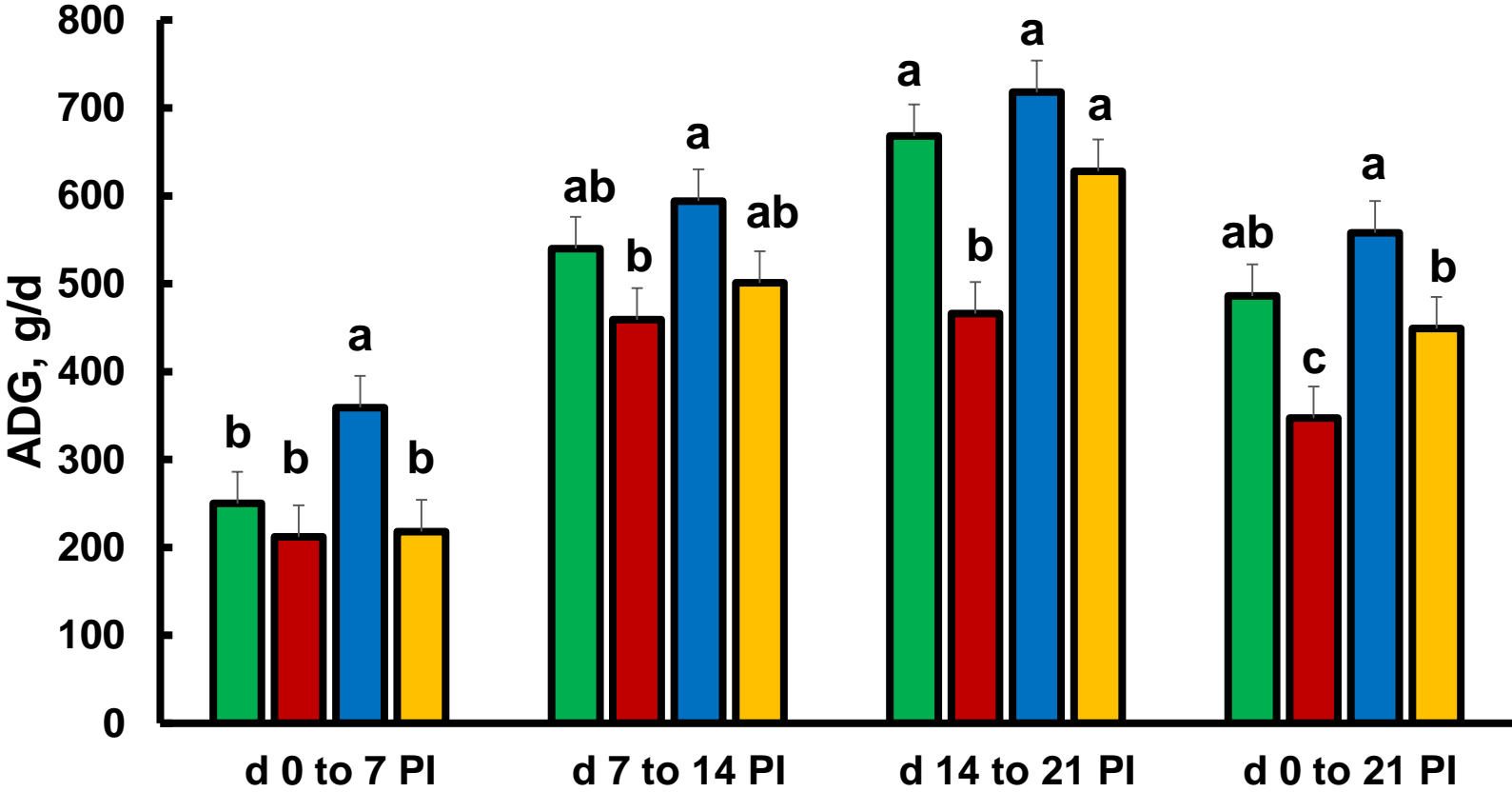
Results

Body weights



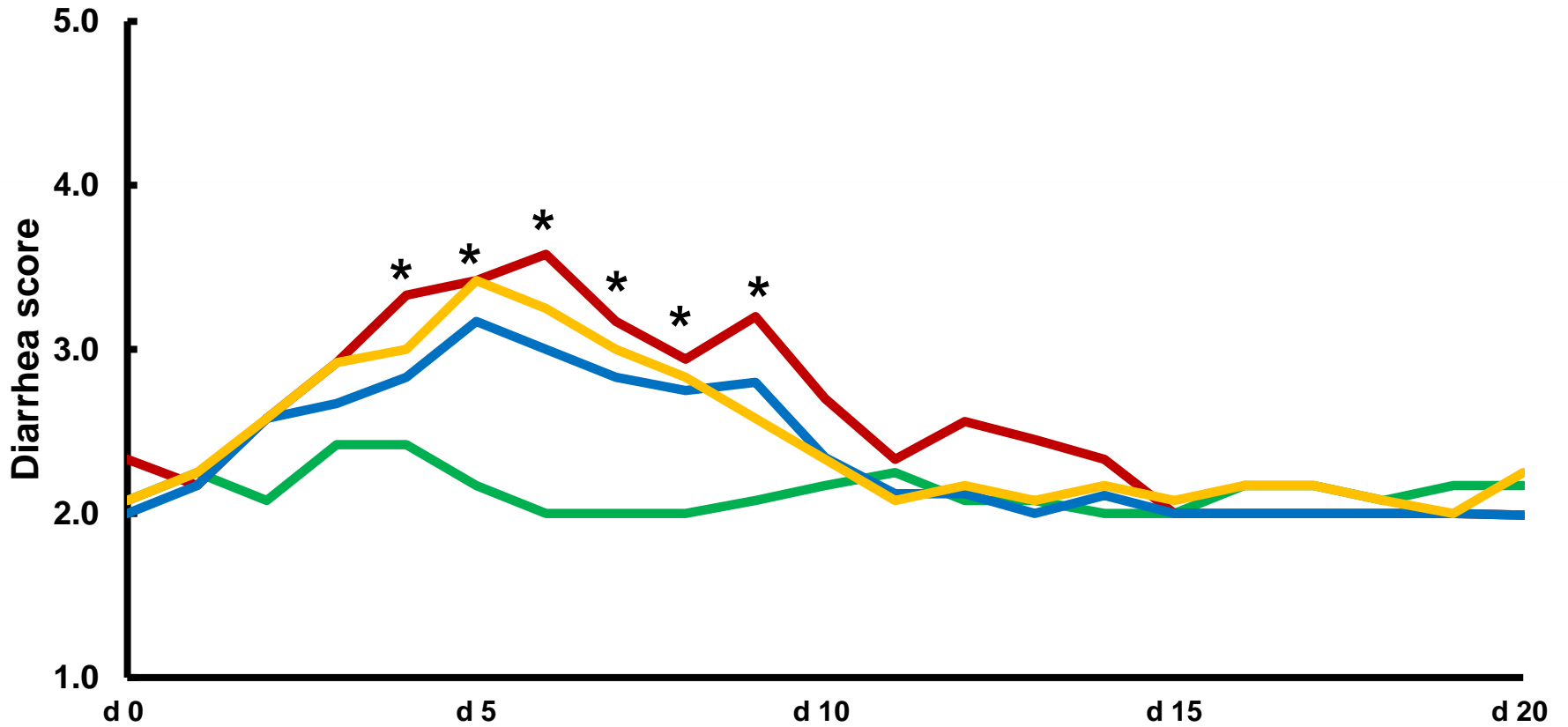
Average daily gain (ADG)

■ NC ■ PC ■ AGP ■ PRO



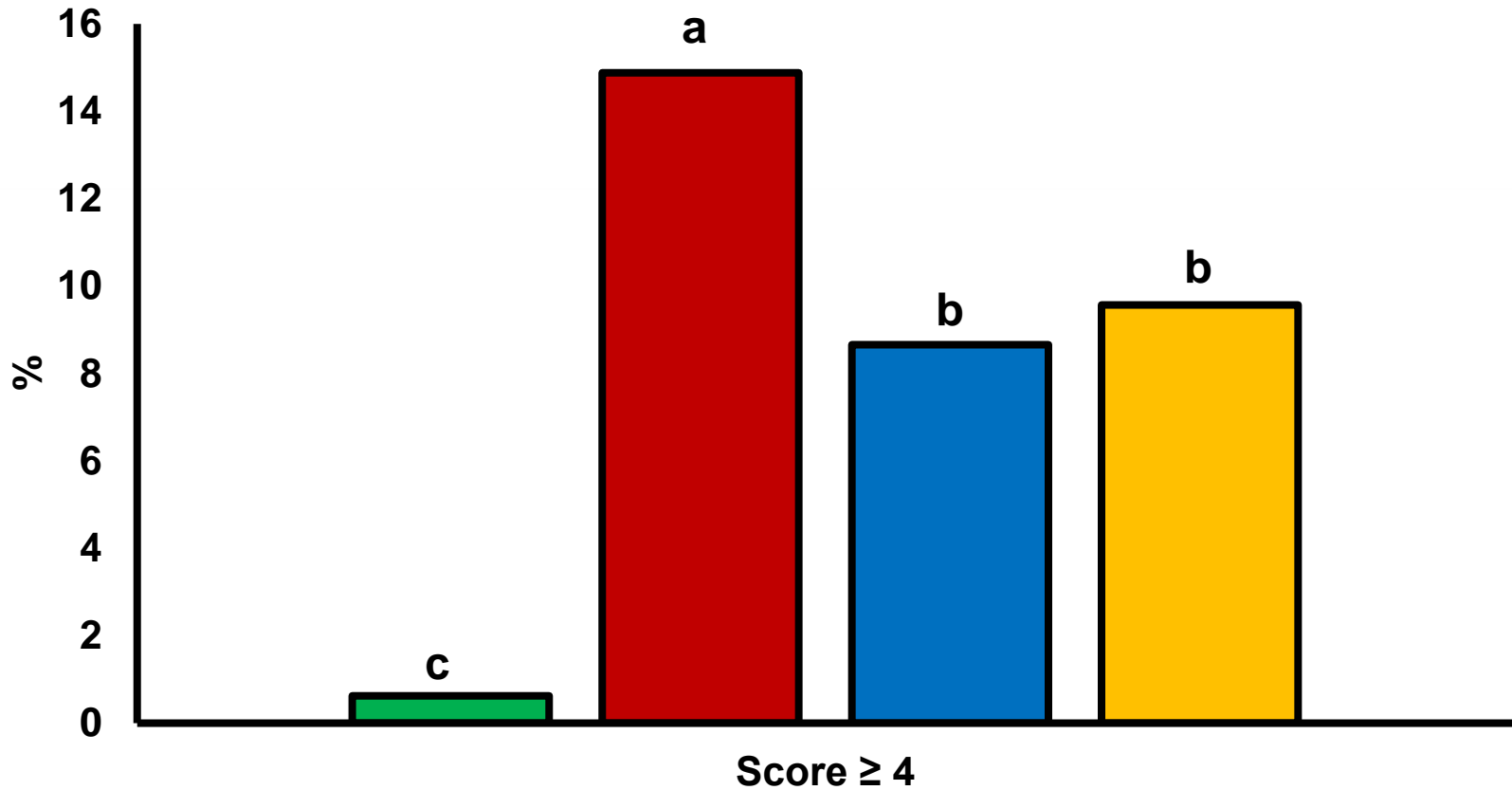
Daily diarrhea score

—NC —PC —AGP —PRO



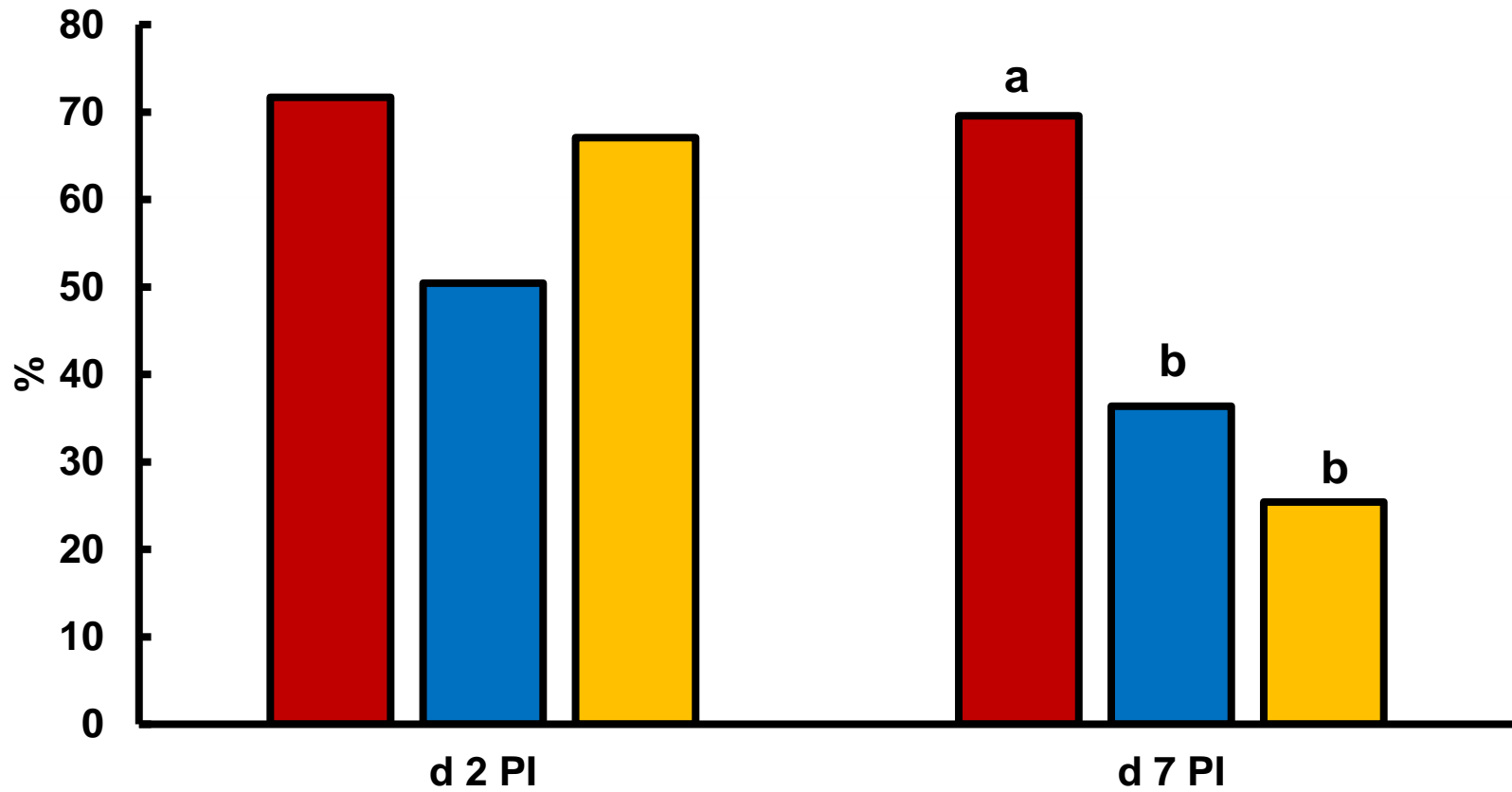
Frequency of diarrhea (d0 to d20)

■ NC ■ PC ■ AGP ■ PRO

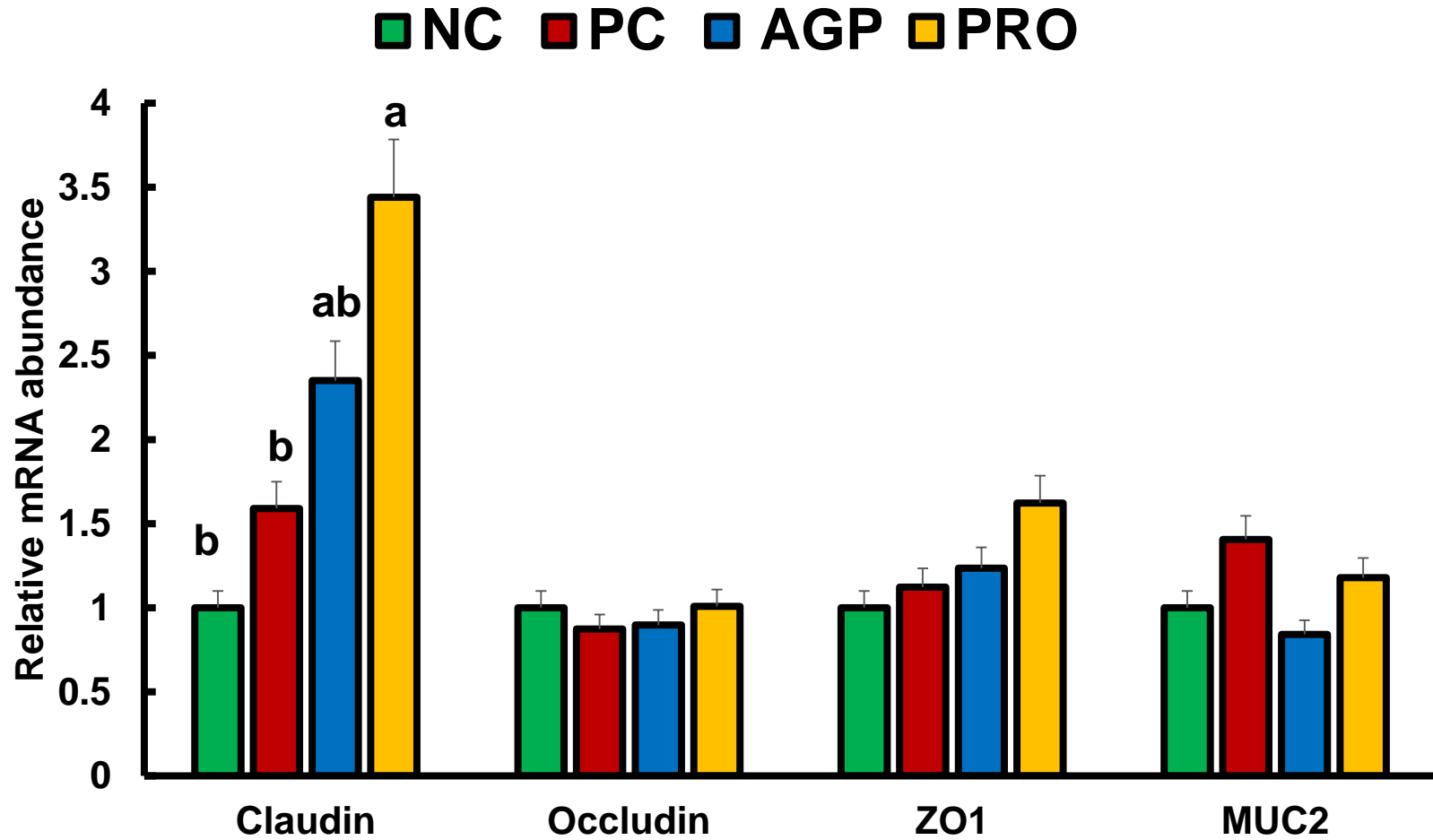


β - hemolytic coliforms

■ PC ■ AGP ■ PRO

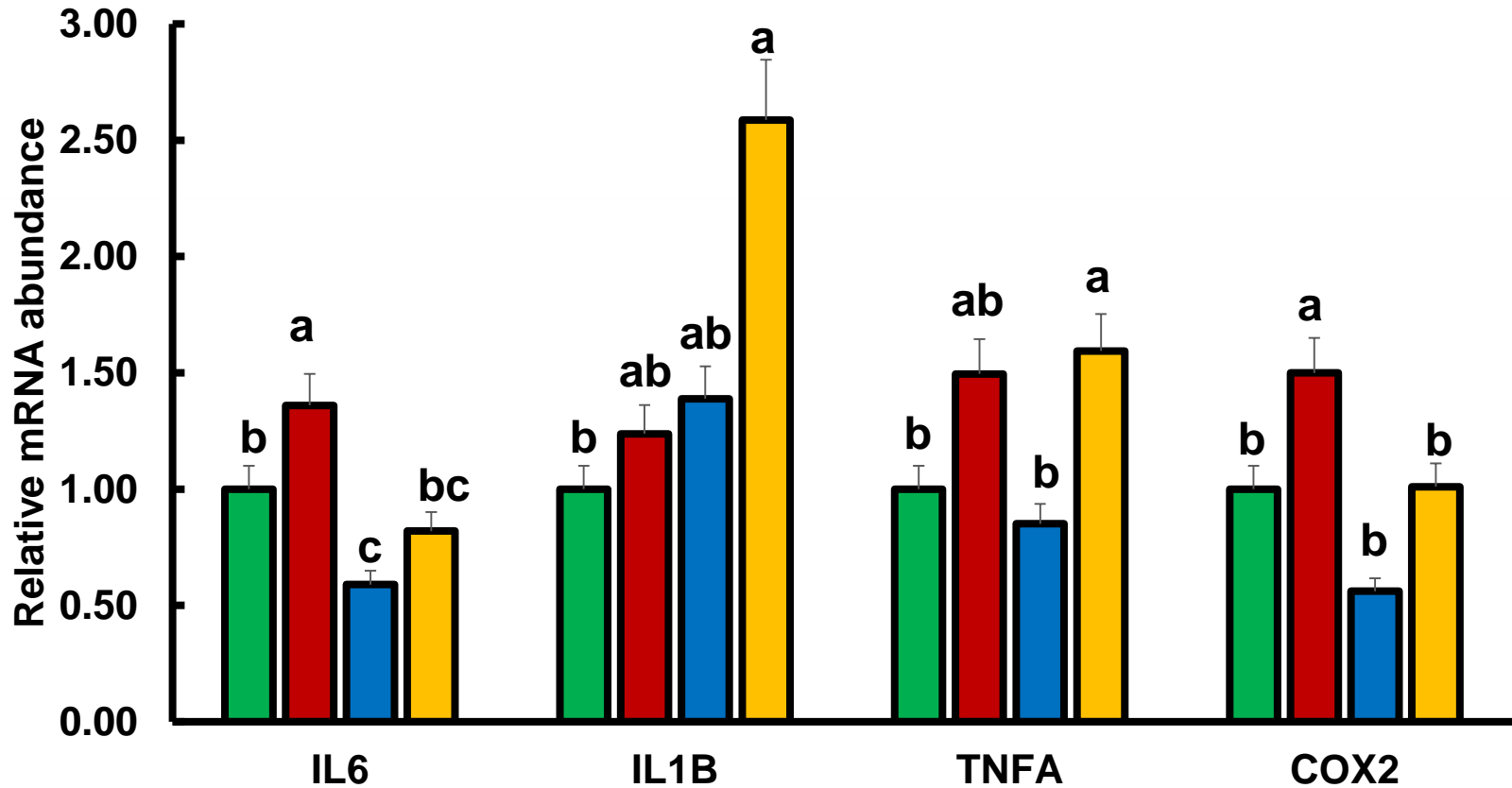


Tight junction protein in jejunum



Proinflammatory cytokines in ileum

■ NC ■ PC ■ AGP ■ PRO



Summary

	AGP	PRO
BW	↑	↑
ADG	↑	↑
Feed efficiency (overall)	↑	↑
Diarrhea score	↓	↓
Frequency	↓	↓
β - hemolytic coliforms	↓	↓
Claudin	NS	↑
IL6	↓	↓
COX2	↓	↓
IL1B	NS	NS

Conclusions

□ *Bacillus subtilis* and carbodox supplementation to *E. coli* challenged weaned pigs reduced:

- Diarrhea score and frequency

and improved:

- Growth performance
- Intestinal health

Acknowledgement

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Thank you!