Trace Amounts of Antibiotic is Detrimental to the Health of Weaned Pigs

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Challenges in swine industry

In-feed antibiotics & Risk

Research: Part 1 & 2
Life cycle of a market pig

16 to 17 weeks (115 to 120d)
6 to 8 weeks (42 to 56 d)
3 months, 3 weeks and 3 days (114 d)
3 weeks (21 d)
Weaning stress

- Environmental changes
- Abrupt transition of diet
- Transportation stress
- Increased exposure to pathogens

Weaning stress
Gastrointestinal (GI) tract development during weanling

Critical Window of postnatal GI Development

- Epithelial barrier and transport functions
- Immune system maturation
- Enteric nervous system

Maternal Immunity

Plasticity

Birth

2.5 to 4 weeks

12 to 14 weeks

Adult

Moeser et al., 2017
Post-weaning diarrhea in pigs

• One of the most serious threats for the swine industry

• Usually associated with proliferation of enterotoxigenic *E. coli* (ETEC)
  ✓ F4 (K88)
  ✓ F18
Post-weaning ETEC diarrhea morbidity

% of sites (US)

- 24 % (2000)
- 31.8 % (2006)
- 32.4 % (2012)

E. coli diarrhea

Swine (Livestock) production technologies

- Genetics
- Reproduction
- Health
- Nutrition
  - Macro-nutrients
  - Micro-nutrients
  - Non-nutrient feed additives
Antibiotics use in livestock

- Antimicrobial substances active against bacteria
- Disease prevention
- Disease treatment
- Growth promotion

Efficacy of antibiotics as growth promoters for weaned pigs (7-25 kg)

Zimmerman, 1986

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Antibiotics as growth promoter in animal diets poses risk

- Emergence of antibiotic resistance
- Banned in the European Union since 2006
- Also banned in the United States since 2017
- Alternatives to antibiotic are highly demanded

https://fairfarmsnow.org
Trace levels of antibiotics: A global health hazard

- Manure
- Surface water
- Soil
- Air
- Dust
- Farm environment

Harbarth et al, 2015
Adverse effects of trace levels of antibiotics

➢ Toxicity
➢ Mutagenicity
➢ Carcinogenicity
➢ Hypersensitivity
➢ Antibiotic resistance

Young animals are more sensitive!

Delay the growth & recovery from diseases
Central Idea & Objective

To investigate the potential detrimental effects of trace amounts of antibiotics on weaned pigs experimentally infected with a pathogenic *E. coli*
Part 1) Trace amounts of antibiotic exacerbated diarrhea and systemic inflammation of weaned pigs infected with a pathogenic *E. coli*

Published; *Journal of Animal Science* (2021)

- **Pathogenic F18 *E. coli* challenge (LT, STb, SLT-2)**; oral inoculation, $10^{10}$ cfu/dose with 3 doses

### NON RUMINANT NUTRITION

Trace amounts of antibiotic exacerbated diarrhea and systemic inflammation of weaned pigs infected with a pathogenic *Escherichia coli*

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**UNIC number:** 2018-0387-0451-02-06

**Abstract**

The experiment was conducted to investigate the effects of trace amounts of antibiotic on growth performance, diarrhea, systemic immunity, and intestinal levels of weaned pigs experimentally infected with an enteropathogenic *Escherichia coli*. Weaned pigs ($n = 34$, initial body weight (BW) 9.4 kg) were individually housed in fiberglass metabolism cages and randomly divided into one of three dietary treatments: control diet (CON) and two additional diets supplemented with 10 or 0.5 mg/kg carbadox to the control base diet (CFM or PBS, respectively). The experiment started at 24 days old and continued until day 14 after the weaning. The pigs were randomly assigned to one of the dietary treatments: control diet (CON), trace amounts of antibiotic (LOW), and recommended dose of antibiotic (REC). Weaning, peak infection period, and recovery period: days 1-7, days 7-11, and days 11-14, respectively. The pigs were challenged at day 7 PI, whereas the weanling pigs were euthanized at the end of the experiment to collect the jejunal and ileal mucosa and homogenates samples for gene expression and histological examination, respectively. The pigs in the CON received only the control diet and the pigs in the LOW and REC groups received all three diets. Weaning, peak infection period, and recovery period: days 1-7, days 7-11, and days 11-14, respectively. The pigs were challenged at day 7 PI, whereas the weanling pigs were euthanized at the end of the experiment to collect the jejunal and ileal mucosa and homogenates samples for gene expression and histological examination, respectively. The pigs in the CON received only the control diet and the pigs in the LOW and REC groups received all three diets.

### Control diet (CON)

- Trace amounts of antibiotic (LOW)
- Control diet + 0.5 mg/kg Carbadox
- Control diet + 50 mg/kg Carbadox

### Pathogen F18 *E. coli* challenge

- Oral inoculation, $10^{10}$ cfu/dose with 3 doses

### E. coli challenged

-Recommended dose of antibiotic (REC)

Key words: *weanling, diarrhea, enteropathogenic *Escherichia coli*, gut health, immunity, weaned pigs
Trace amounts of antibiotic reduced growth performance

Average daily gain

CON  TRA  REC

d -7 to 0  d 0 to 5 PI  d 5 to 11 PI

Gain:Feed

CON  TRA  REC

d -7 to 0  d 0 to 5 PI  d 5 to 11 PI

TRAt = 0.5 mg/kg Carbadox

RECs = 50 mg/kg Carbadox

PI=post-inoculation
Trace amounts of antibiotic exacerbated the severity of diarrhea

**Diarrhea score**

- **CON**
- **TRA**
- **REC**

**Frequency of diarrhea ≥ 4**

**TRA** = 0.5 mg/kg Carbadox

**REC** = 50 mg/kg Carbadox

**PI** = post-inoculation

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Trace amounts of antibiotic worsened the *E. coli* infection

**β-hemolytic coliforms in feces**

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<thead>
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<th>CON</th>
<th>TRA</th>
<th>REC</th>
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<tbody>
<tr>
<td>d 2 PI</td>
<td>a</td>
<td>b</td>
<td>c</td>
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<tr>
<td>d 5 PI</td>
<td>a</td>
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<tr>
<td>d 8 PI</td>
<td>a</td>
<td>b</td>
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</tr>
<tr>
<td>d 11 PI</td>
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**Bacterial translocation in mesenteric lymph node**

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<tbody>
<tr>
<td>d 5 PI</td>
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<tr>
<td>d 11 PI</td>
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**TRACHE** = 0.5 mg/kg Carbadox

**REC** = 50 mg/kg Carbadox

**PI** = post-inoculation
Trace amounts of antibiotic elevated systemic inflammatory markers

**TNF-α**

**C-reactive protein**

**Haptoglobin**

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Part 2) Trace amounts of antibiotic altered metabolomic and microbial profiles of weaned pigs infected with a pathogenic *E. coli*

Gresse et al. (2017); Metabolomic Technologies Inc.
Partial Least Squares Discriminant Analysis (PLS-DA) 2D score plot of the metabolites in serum showed separated clusters between trace amounts of antibiotic and label-recommended dose of antibiotic groups
Significantly changed pathways in serum between trace amounts of antibiotic and label-recommended dose of antibiotic groups

- Citrate cycle (TCA cycle)
- Arginine biosynthesis
- Alanine, aspartate, and glutamate metabolism
- Glycolysis / Gluconeogenesis
- Arginine biosynthesis
- Aminoacyl-tRNA biosynthesis
- Alanine, aspartate, and glutamate metabolism
- D-Glutamine and D-glutamate metabolism
Partial Least Squares Discriminant Analysis (PLS-DA) 2D score plot of the metabolites in colon digesta showed separated clusters between trace amounts of antibiotic and label-recommended dose of antibiotic groups.
Significantly changed pathways in colon digesta between trace amounts of antibiotic and label-recommended dose of antibiotic groups

**d 5 PI**
- Starch and sucrose metabolism
- Arginine biosynthesis
- Arginine and proline metabolism
- Purine metabolism

**d 11 PI**
- Aminoacyl-tRNA biosynthesis
- Arginine biosynthesis
- Arginine and proline metabolism
- Glutathione metabolism
- Glyoxylate and dicarboxylate metabolism
- Alanine, aspartate, and glutamate metabolism
- Pentose and glucuronate interconversions

Relative abundance of Firmicutes family in colon digesta of pigs fed diets supplemented with different dose of antibiotic on d 5 and 11 post-inoculation
Trace amounts of antibiotic have shown detrimental effects on growth performance and disease resistance of pigs challenged with ETEC F18, potentially by exacerbating systemic inflammation and altering metabolic and microbial profiles.

On-going and future research
- RNA sequencing
- Correlation analysis
- Targeted-metabolomics
- Metagenomics
Acknowledgements

Comparative Animal Nutrition & Physiology Laboratory
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Research project No. W4002
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