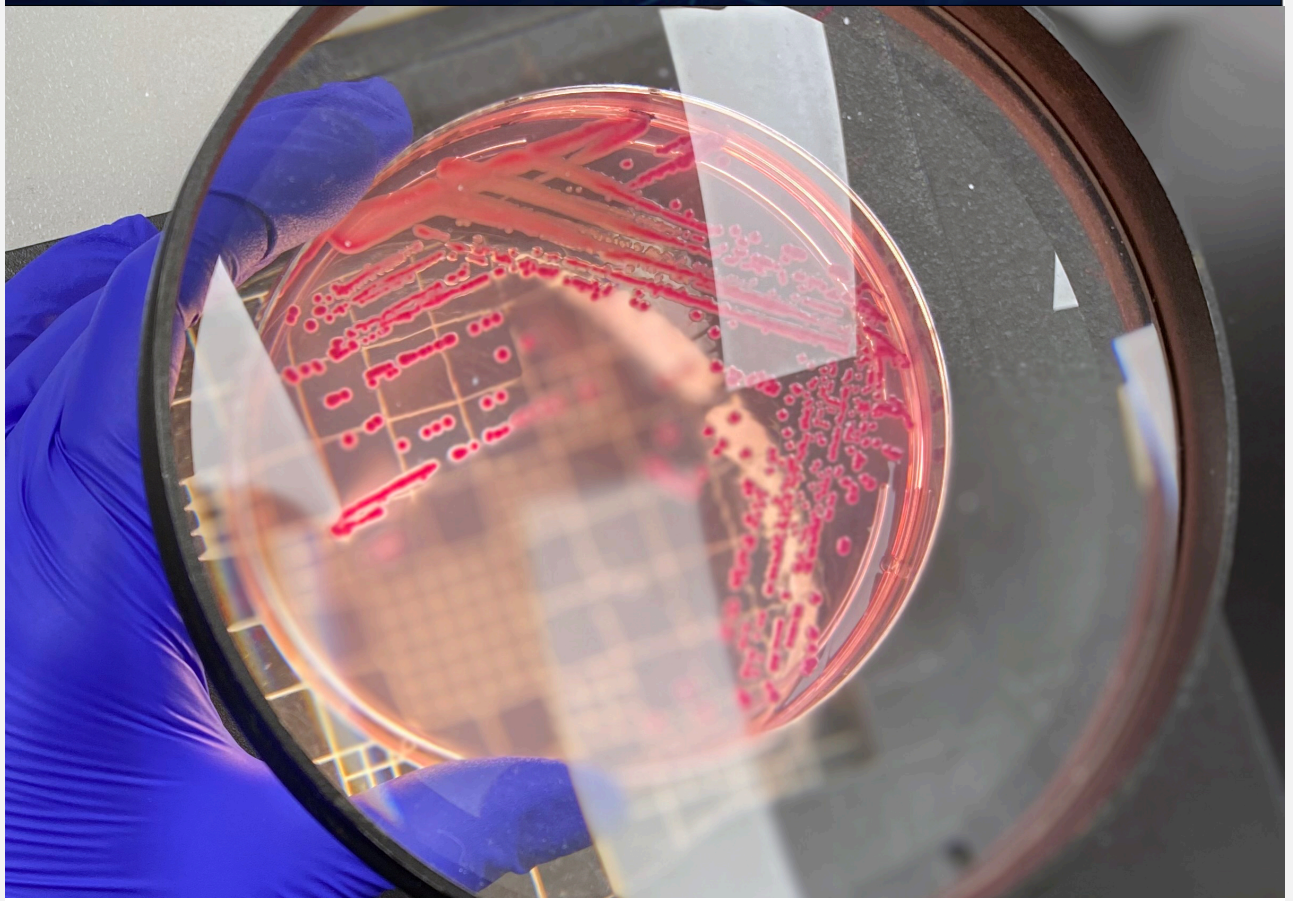
PATHKINEX UPDATE



PathKinex™ Update:

From Worms to Livestock: Impact of *Bacillus* in ETEC Challenge Models

Enterotoxigenic *E. coli* (ETEC) is a common pathogen that can lead to mortality and reduced growth performance across all livestock species and is also a major causative agent of post-weaning diarrhea in nursery pigs.[1,2] ProVent® ECL is a multi-strain direct-fed microbial (DFM) designed to promote health during challenge situations like those caused by ETEC through various mechanisms, including immune modulation, quorum sensing interference, competitive exclusion, and antimicrobial activity. Recently, teams at United Animal Health and Microbial Discovery Group have developed two models to test the potential of ProVent® ECL in direct ETEC challenge scenarios: the first involving use of *Caenorhabditis elegans* as an indicator organism and the second involving an oral challenge in nursery pigs.

C. elegans ETEC Model

C. elegans, pictured in Figure 1, is valued in disease and infection modeling due to its conserved biological pathways, genetic tractability, rapid life cycle, and transparency, which allow direct observation of disease processes and host-pathogen interactions.[3] Compared to *in vitro* testing alone, *C. elegans* models help provide greater confidence in strain benefits, which can accelerate decision making prior to the initiation of more expensive and complex animal trials.

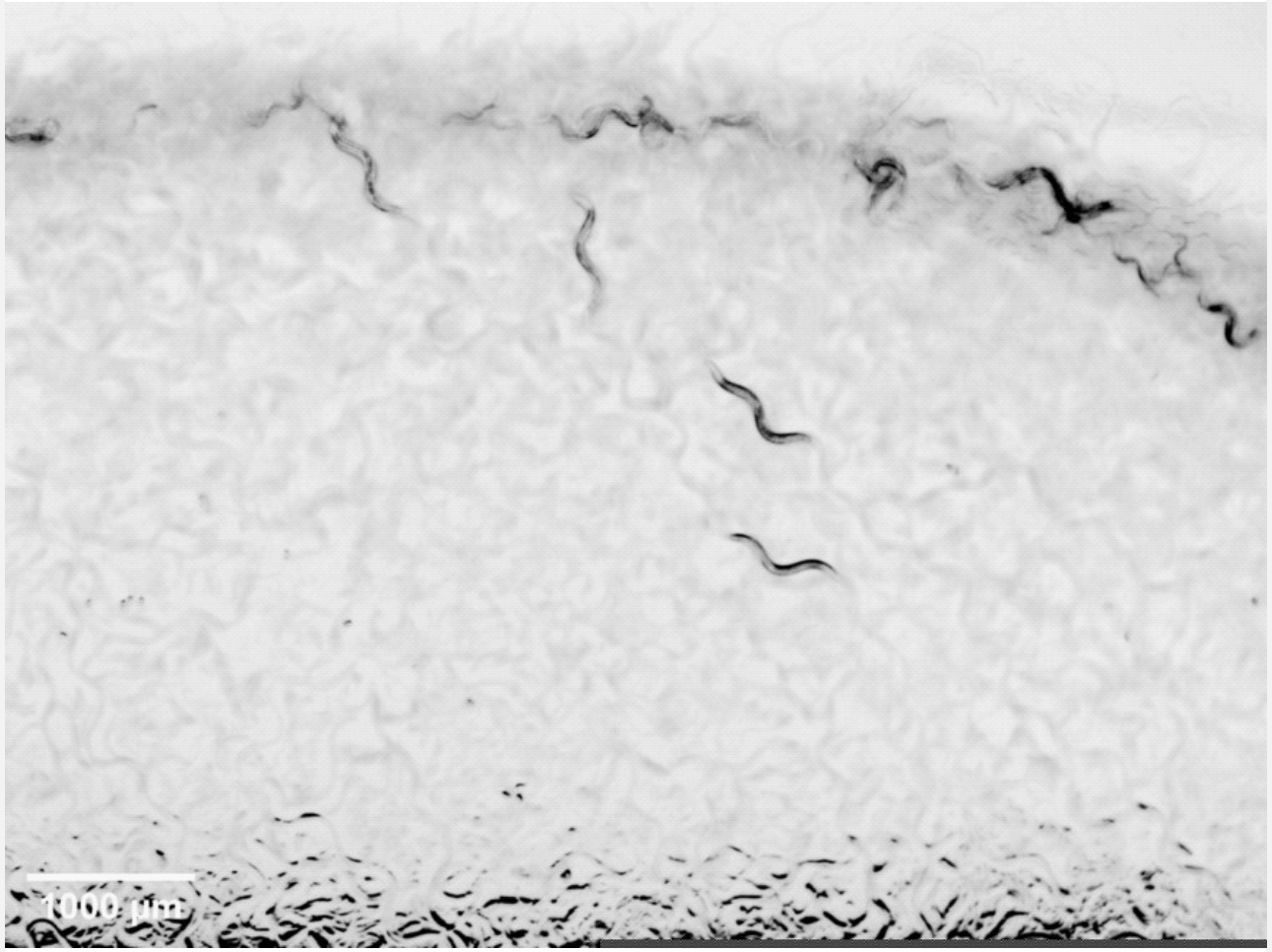


Figure 1. Video of *C. elegans* moving across an agar plate. (Video credit: N. Sepulveda)

To assess *C. elegans* performance during an ETEC challenge, worms were fed ETEC for three days. The worms were then transferred to the same ETEC challenge (control) or to ProVent® ECL. Worms were assayed for colonization of ETEC by homogenizing and plating to assess the quantities of ETEC in the gastrointestinal tract, and for performance through a swimming motility assessment. Swimming is a measure of fitness reflecting overall health and performance in response to treatments.[4] In this model:

- Worms fed ProVent® ECL harbored no detectable *E. coli* (Figure 2A, $P<0.05$).
- Worms fed ProVent® ECL exhibited 15%–30% improvement in swimming performance over the ETEC controls (Figure 2B).

Overall, the *C. elegans* ETEC challenge model revealed ProVent® ECL's ability to mitigate negative effects of ETEC disease through reduced pathogen colonization, which, in turn, improved performance.

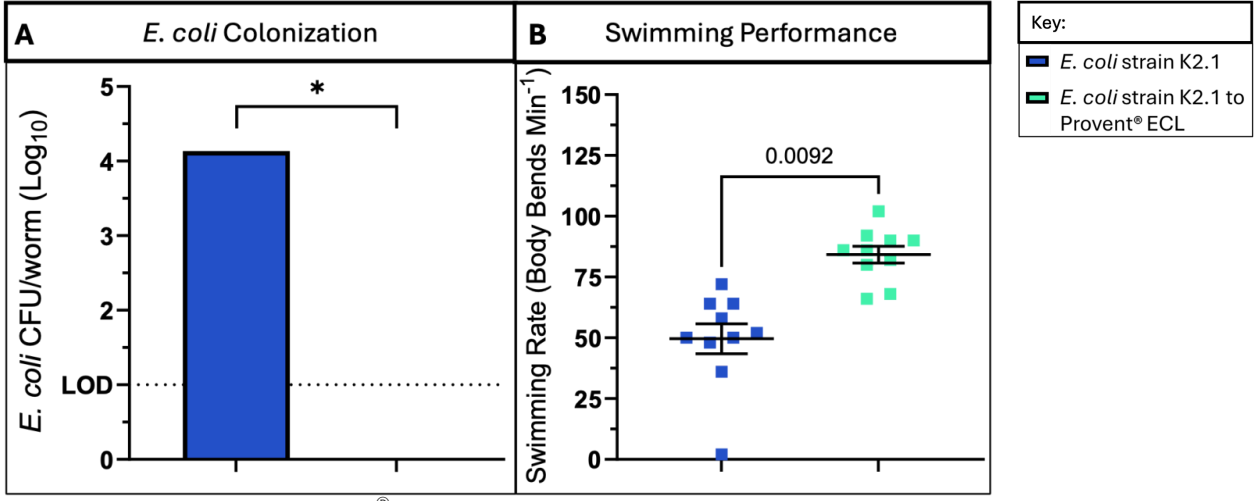


Figure 2. The impact of ProVent® ECL on colonization and swimming performance in a *C. elegans* ETEC challenge model. One-way ANOVA and a significance of P<0.05 determined differences by treatment. **A.** *E. coli* colonization per worm. LOD represents the limit of assay detection. **B.** Worm swimming rate.

Nursery F18 ETEC Oral Challenge Model

An *E. coli* challenge trial was conducted in nursery pigs to evaluate the effectiveness of ProVent® ECL in reducing mortality from ETEC. Piglets were fed dietary treatments (Table 1) over a 28-day period, and a high dose of *E. coli*—greater than would typically be found on the farm—was administered orally on Day 14. At the conclusion of the trial (Day 28), all pigs were euthanized, ileum samples were collected, digesta was removed, and the mucosal surface was swabbed for analysis.

Table 1. Dietary treatments utilized in a swine ETEC oral challenge model.

Treatment Key	Treatment Description
A	Negative control
B	Negative control and 1 lb ProVent® ECL per finished ton
C	Negative control and 3 lbs ProVent® ECL per finished ton
D	Negative control and ZnO (internal control)

In this trial, the higher inclusion of ProVent® ECL (3 lbs per finished ton) delayed the onset of infection post-challenge and improved survival compared to the other treatments (Figure 3, P<0.07). ProVent® ECL at 1 lb was likely not enough

to overcome the high dose of ETEC, while the 3 lb inclusion showed the potential of ProVent® ECL in this severe model. Additionally, the *F18 E. coli* gene was not detected in any pigs from the 3 lb ProVent® ECL treatment in Day 28 ileum samples (Figure 4, P=0.061).

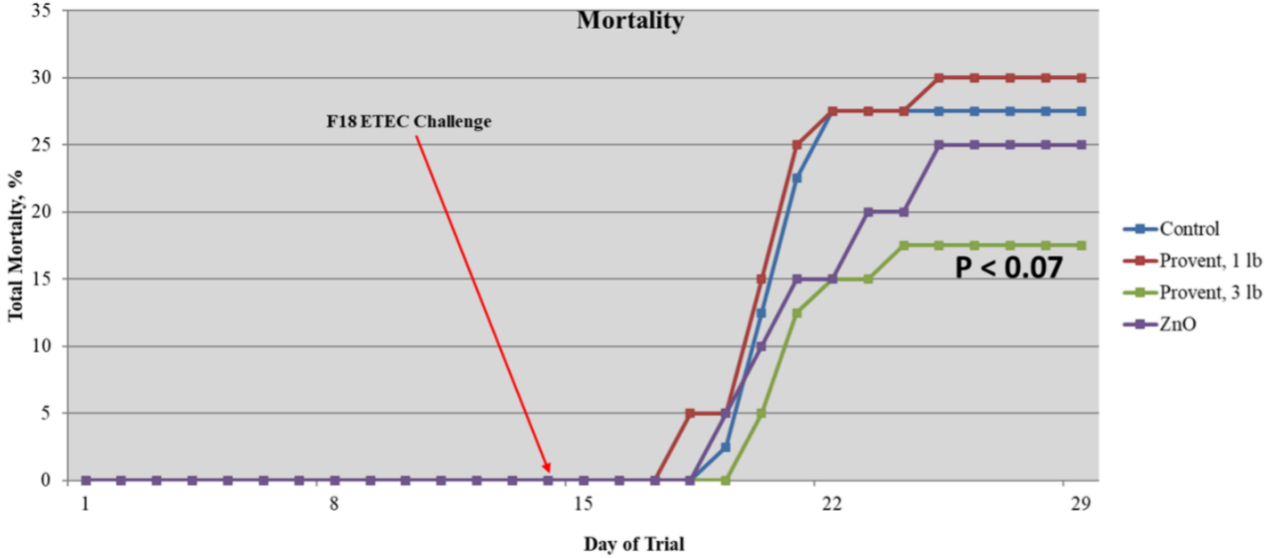


Figure 3. Percent mortality by treatment recorded throughout the trial from Day 0 through Day 28.

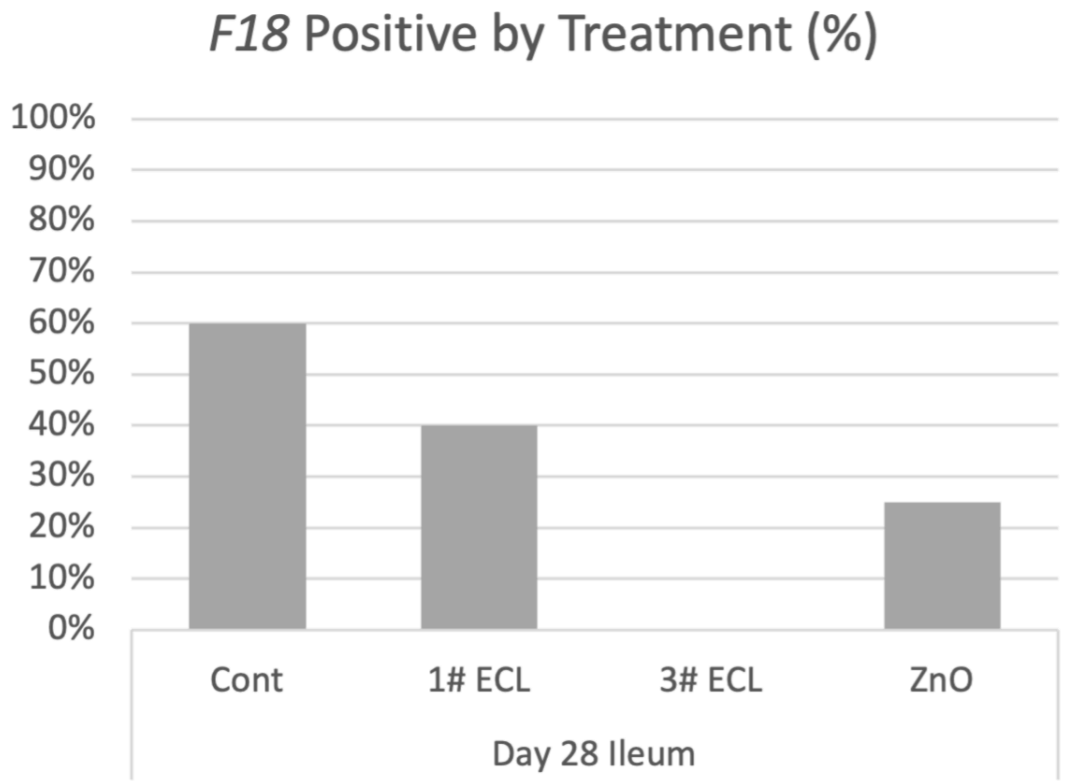


Figure 4. Percent of samples *F18* positive by treatment in ileum samples on Day 28.

Conclusion

Both the *C. elegans* and swine nursery ETEC challenge models demonstrate the strong ability of ProVent® ECL to mitigate the effects of enterotoxigenic *E. coli*. In *C. elegans* studies, ProVent® ECL treatment led to reduced pathogen colonization and improved swimming performance. Similarly, during a severe *E. coli* challenge in nursery pigs, ProVent® ECL at 3 lbs per ton reduced ETEC quantities by qPCR to undetectable levels and improved piglet survival. While ProVent® ECL demonstrates efficacy against ETEC challenge alone, integrating it with complementary health strategies may further optimize performance and protection on the farm.

Discussion Questions

What strategies (vaccines, antibiotics, hygiene protocols, etc.) are you currently using to prevent or control ETEC infections?

Respond to MDG

About the Author

Kaley Pederson is a Microbiologist III at Microbial Discovery Group. She is focused on further understanding swine host-microbe interactions through the application of large-scale microbial surveillance platforms, as well as characterization of novel microbial strains.

References:

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