An Investigation into the Effect of Probiotic Supplementation

on the Prevalence and Severity of Cryptosporidium Infections in Young Calves

Cryptosporidiosis is a major cause of morbidity and economic loss in the livestock industry, particularly in neonatal calves. This study evaluated the impact of probiotic supplementation on Cryptosporidium infections in 50 calves over a 6-week period on a commercial dairy farm. Calves were divided into treatment and control groups, with probiotics administered according to manufacturer guidelines. Faecal testing, scour scoring, body weight and temperature monitoring were used to assess infection severity.

Probiotic supplementation significantly reduced Cryptosporidium prevalence and improved clinical outcomes, including higher milk consumption (morning: Estimate = 0.327 L, SE = 0.046, $p < 0.0001$; evening: Estimate = 0.185 L, SE = 0.045, $p < 0.0001$), improved temperature regulation (Estimate = −0.36°C, SE = 0.09, $p = 0.0052$) and lower scour severity (Estimate = −1.83, $z = -8.25$, $p < 0.001$). Effects varied with calf age, with significant temperature differences at 7–12 days, reflecting infection dynamics.

These findings suggest that probiotics offer a practical approach to reducing Cryptosporidium infections and improving calf health, with potential benefits for livestock productivity and farm sustainability. Further work should explore underlying mechanisms and long-term outcomes.

Cryptosporidiosis, caused by Cryptosporidium parvum, is a major health challenge in neonatal calves, leading to diarrhoea, dehydration, weight loss and mortality. These outcomes compromise both animal welfare and the economic viability of livestock production, with reduced growth rates causing substantial financial losses. Given the zoonotic potential of Cryptosporidium, effective control measures are essential for safeguarding public and animal health.

Drug-based treatments are limited by efficacy concerns and the risk of resistance, driving the need for alternative strategies. Probiotics have emerged as a promising option, offering potential benefits for intestinal health without contributing to antimicrobial resistance.

Rationale for the Study

Probiotics, live microorganisms that confer health benefits to the host, can enhance gut microbiota balance, strengthen mucosal barriers and produce antimicrobial metabolites such as short-chain fatty acids and bacteriocins. These mechanisms may help reduce Cryptosporidium infection and disease severity in calves.

Although probiotics have been widely studied in livestock, evidence for their role in managing Cryptosporidium under commercial conditions remains limited. This study addresses this gap by evaluating the efficacy of probiotic supplementation in reducing infection prevalence and severity in young calves.

CRYPTOSPORIDIUM AND PROBIOTICS IN CALVES

Cryptosporidium in Calves

Cryptosporidium parvum, a protozoan parasite, is a major cause of neonatal calf diarrhoea (Tzipori, 1983). Infection begins when ingested oocysts release sporozoites that colonise intestinal epithelial cells, resulting in nutrient malabsorption, diarrhoea, weight loss, dehydration and, in severe cases, death (Costa *et al.*, 2011; Berchtold, 2009). Young calves, with their immature immune systems, are particularly susceptible during the first weeks of life (Niine *et al.*, 2018). Beyond clinical illness, Cryptosporidiosis reduces productivity, increases veterinary costs and poses a zoonotic risk, especially for children and immunocompromised individuals (Pumipuntu & Piratae, 2018).

Probiotics and Gastrointestinal Health

Probiotics are live microorganisms that, when administered in adequate amounts, confer health benefits on the host (Kechagia *et al.*, 2013). Common strains include Lactobacillus, Bifidobacterium and Saccharomyces (Fijan, 2014). Their actions include competing with pathogens for adhesion sites, strengthening the intestinal barrier, producing antimicrobial metabolites and modulating immune responses (Bermudez-Brito *et al.*, 2012). These properties underpin their therapeutic potential against gastrointestinal pathogens, including protozoa. In livestock, probiotics have been associated with improved growth, feed efficiency and reduced diarrhoea in swine and poultry (Kabir, 2009; Liao & Nyachoti, 2017). Evidence in cattle is less extensive, but early studies suggest benefits for digestion and disease resistance (Lambo *et al.*, 2021).

Probiotics and Cryptosporidium

Several investigations have assessed probiotics against Cryptosporidium. In a large field trial, Bacillus subtilis supplementation reduced oocyst shedding in Holstein calves, while synbiotic treatments improved weight gain (Niine *et al.*, 2018). In vitro, Lactobacillus supernatants reduced C. parvum oocyst viability by up to 81%, suggesting direct antiparasitic effects (Berrilli *et al.*, 2012). However, some field trials found no effect under high environmental parasite pressure, highlighting variability in outcomes (Gaber *et al.*, 2022).

MECHANISMS OF PROBIOTIC ACTION AGAINST CRYPTOSPORIDIUM

Probiotics May Mitigate Infection Through:

- **Competition for Adhesion Sites:** Occupying intestinal surfaces reduces parasite attachment (Oliveira & Widmer, 2018).
- **Production of Antimicrobial Substances:** Organic acids and bacteriocins create an unfavourable environment for parasite survival (Sharifi-Rad *et al.*, 2020).
- **Immune Modulation:** Probiotics interact with gut-associated lymphoid tissue, enhancing cytokine production and promoting a balanced immune response (Yan & Polk, 2011).

AIM AND OBJECTIVES

Aim

To evaluate the effect of probiotic supplementation on the prevalence and severity of Cryptosporidium infections in calves under commercial farm conditions.

METHODS

Study Design and Location

The study was conducted at Roadhead Farm, Quathquan, Biggar (ML12 6ND) over 6 weeks (26 June–7 August 2023). Fifty calves were enrolled and systematically allocated to treatment (n = 25) and control (n = 25) groups using a pen-stratified sequential approach to ensure balanced housing conditions.

Housing and Management

Calves were housed indoors in pens of five, with treatment and control groups blocked by pen position. Pens were disinfected prior to entry, bedded daily with straw and maintained under uniform environmental conditions (temperature, humidity and ventilation). Sanitation practices included footbaths, regular cleaning of equipment and use of disinfectants to minimise cross-contamination.

Treatments

The treatment group received 30 ml of Precision Microbes® calf probiotic mixed in milk once daily (AM) for the first 14 days of life. The control group received no supplementation. Supportive care protocols were in place: electrolyte solution (Avital) was administered for dehydration and Meloxidyl (2 ml) for severe cases requiring pain relief.

Data Collection

Health monitoring was conducted daily, including visual assessment for diarrhoea and scour scoring (Alltech faecal scoring guide). Faecal samples were collected every second day, stored in sterile containers and tested on-site using Actus rapid detection kits for Cryptosporidium, Rotavirus, E. coli, and Salmonella. Results were compared against ELISA performance standards.

Body weights were recorded at birth and daily thereafter using a calibrated livestock scale. Body temperatures were taken daily at noon using a non-contact infrared thermometer.

Ethical Approval

All procedures complied with animal welfare regulations and were approved by the SRUC AWERB (BOR 2023-017 DAI A00, 7 June 2023). Veterinary care was provided as required.

Data Analysis

Analyses were conducted in R. Data distributions were assessed using histograms and QQ plots. Cumulative link mixed models (CLMM) were applied to scour scores and Cryptosporidium prevalence. Linear mixed-effects models (LMEM) evaluated temperature, milk intake and weight gain, with age as a random factor. Fixed effects were tested for significance and post hoc comparisons identified age-specific treatment effects.

RESULTS

Prevalence of Infection

Faecal testing confirmed Cryptosporidium as the predominant pathogen. CLMM analysis showed a significant reduction in prevalence in the probiotic group compared to controls (Estimate = −1.85, z = −3.93, $p < 0.001$). No effects were observed for breed, sex, or pen. Random effects indicated considerable variability between calves and across ages, highlighting individual differences in susceptibility.

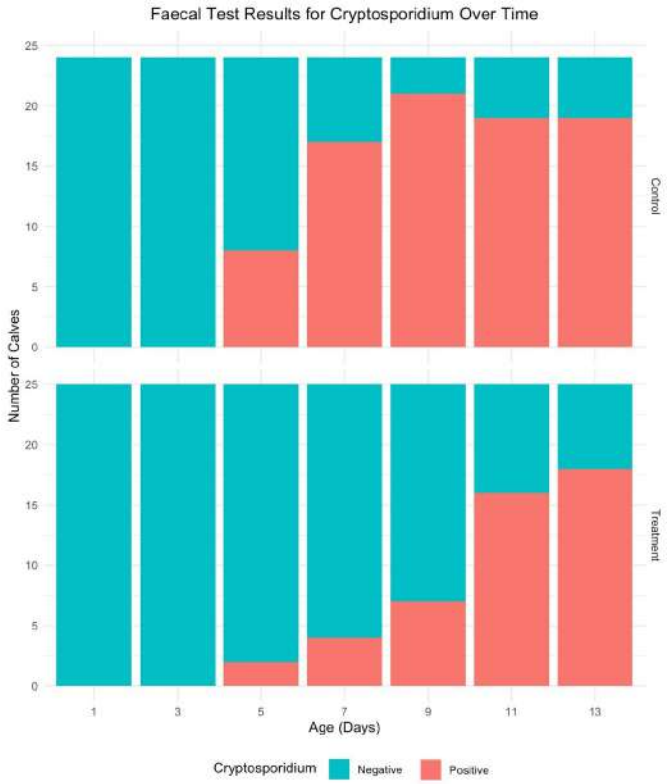

Figure 1. Cryptosporidium Faecal Test Results Over Time, with One Calf Removed from the Control Group Due to Death on Day 1

SEVERITY OF SYMPTOMS

Milk Intake

Probiotic supplementation significantly increased milk consumption. Wilcoxon tests revealed higher morning ($p < 0.0001$) and evening ($p < 0.0001$) intakes in treated calves. LMEMs confirmed this effect (morning: Estimate = 0.33 L, SE = 0.046; evening: Estimate = 0.18 L, SE = 0.045). Age-specific analysis showed the strongest differences at days 7–10, when control calves experienced marked intake reductions.

Weight Gain

Treated calves had significantly greater weight gain than controls (Estimate = 0.24 kg/day, SE = 0.060, $p < 0.001$). Differences were most evident by day 14. Breed, sex and pen effects were not significant.

Scour Score

Probiotic supplementation reduced scour severity (Estimate =

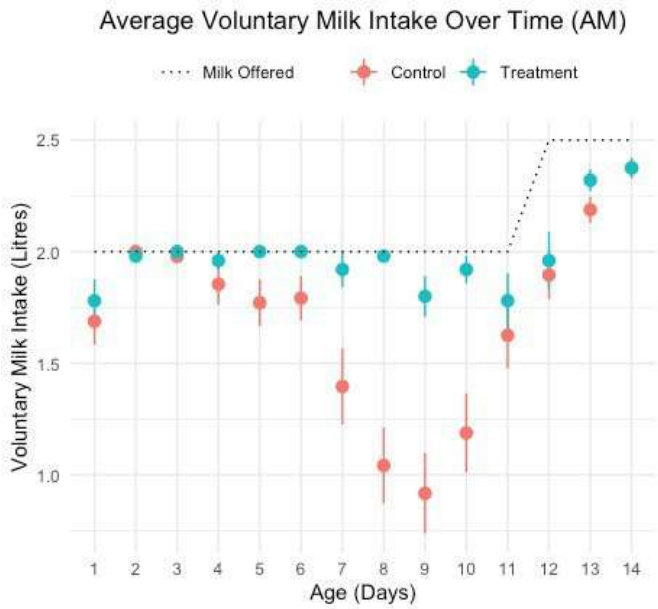

Figure 2. Average Morning Voluntary Milk Consumption Over Time

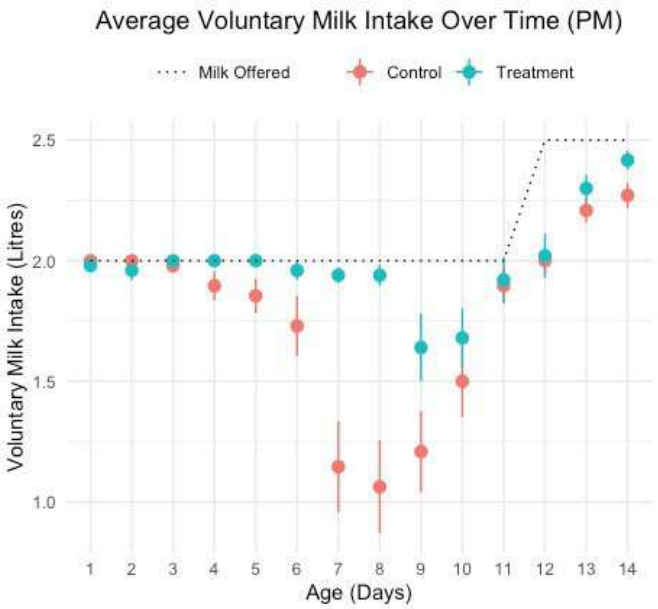

Figure 3. Average Evening Voluntary Milk Consumption Over Time

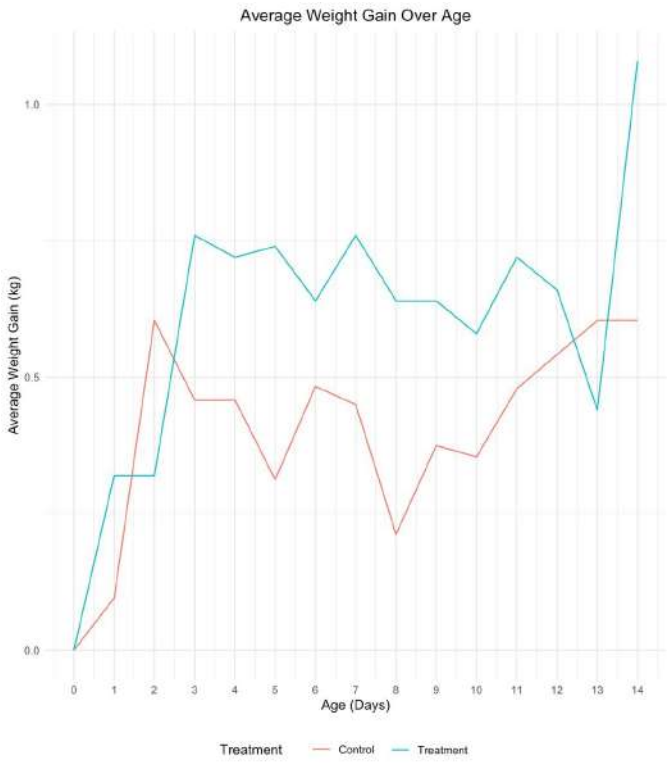

Figure 4. Average Daily Weight Gain of Treatment and Control Group

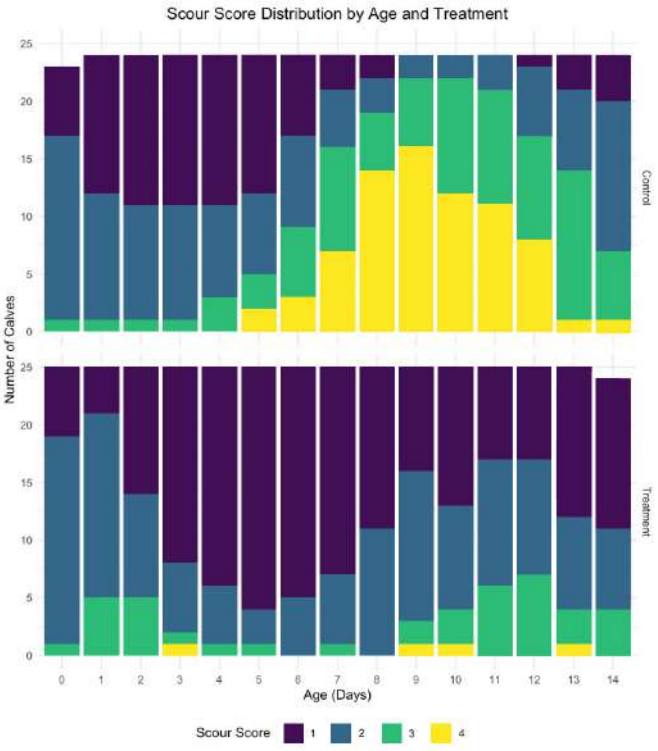

Figure 5. Scour Score Distribution over time, showing a significant increase in scour score in the control group from ages 4–14.

−1.83, z = −8.25, p < 0.001). Random effects confirmed variability between calves and across ages, but treatment consistently lowered scour scores relative to controls.

Temperature

Treated calves exhibited more stable body temperatures, averaging 0.36°C lower than controls (Estimate = −0.36, SE = 0.09, p = 0.005). Post hoc tests identified significant treatment effects at days 7–12, coinciding with peak infection in controls.

DISCUSSION

Disease Prevalence

Probiotics significantly reduced the prevalence of Cryptosporidium infection, supporting earlier in vitro findings that certain Lactobacillus strains impair oocyst viability (Foster *et al.*, 2003). This suggests that probiotics may modulate the gut environment to limit parasite establishment, with potential benefits for animal health and farm economics. Although breed and sex had no measurable influence, variability between individuals and across ages highlights the heterogeneous nature of infection dynamics. Seasonal studies and stricter biosecurity (e.g., individual housing) would help confirm these findings and minimise cross-contamination risks.

Nutritional Performance

Treated calves showed higher milk intake and significantly greater weight gain. Reduced intake in control calves coincided with peak infection, indicating that probiotics

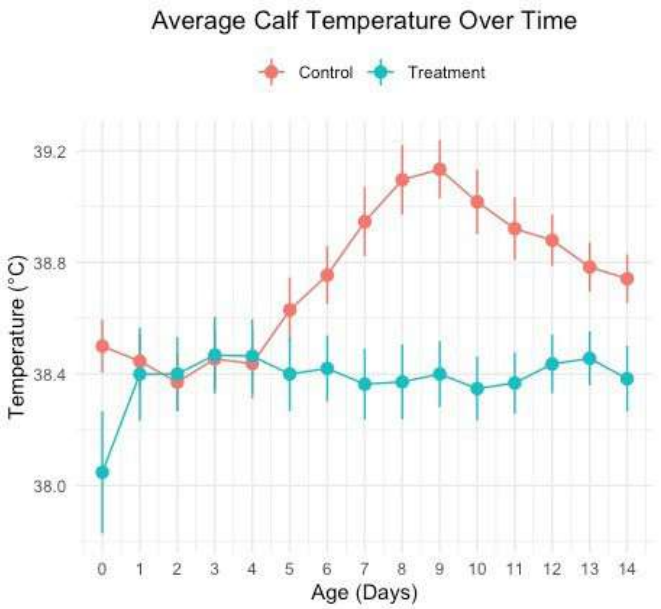

Figure 6. Average Calf Temperature Over Time

helped preserve appetite and nutrient absorption during disease challenge. Enhanced weight gain is consistent with improved gastrointestinal integrity, possibly via protection of villi from parasite-induced damage (Maradana *et al*., 2023). These results align with previous work showing benefits of synbiotic supplementation in heifers, though our findings demonstrate clear gains from probiotics alone.

Clinical Symptoms

Scour severity was substantially reduced in treated calves, consistent with the hypothesis that probiotics strengthen mucosal barriers, suppress pathogens and improve microbiota balance. Temperature regulation was also improved, with treated calves maintaining lower, more stable body temperatures during peak infection periods. Together, these outcomes suggest that probiotics mitigate the physiological stress of Cryptosporidium infection.

Study Considerations and Future Directions

While results were robust across breeds and sexes, unmeasured environmental or management factors (e.g., pen conditions) may have influenced outcomes. Broader trials across seasons and farm systems are needed to confirm efficacy. Further work should also clarify strain-specific effects, optimal dosing and long-term benefits for growth, productivity and resilience.

Conclusion

This study demonstrates that probiotic supplementation significantly reduces the prevalence and severity of Cryptosporidium infections in young calves. Treated calves showed lower infection rates, reduced scouring, improved temperature regulation, greater milk intake and enhanced weight gain compared to controls.

These findings highlight probiotics as a practical, cost-effective strategy for improving calf health and mitigating the impact of cryptosporidiosis in commercial dairy systems. Consideration of age-related responses and individual variability will be important for optimising use in practice. Further research on breed, sex, dosing protocols and long-term outcomes is warranted to strengthen application across diverse livestock settings.

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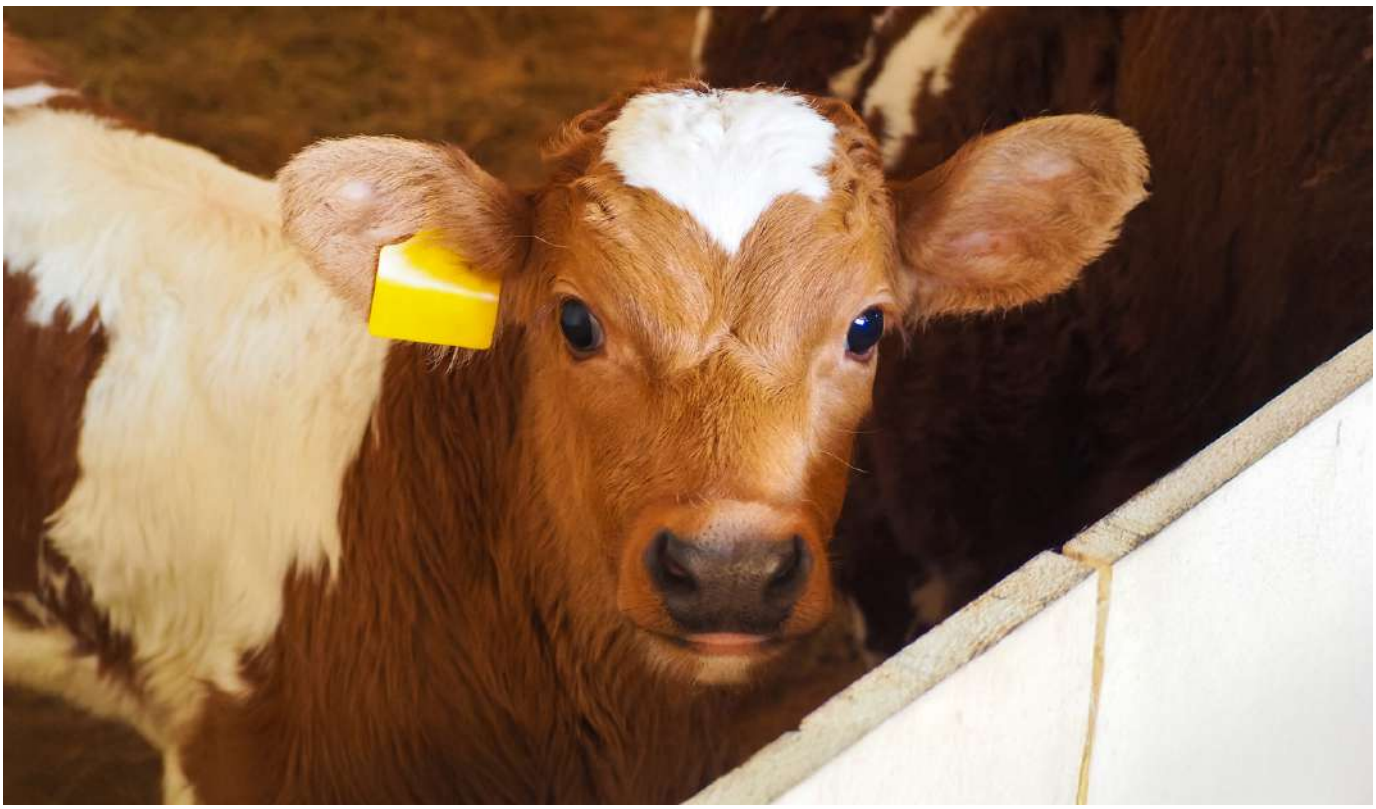

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