Zootechnical performance and pathogen control in broilers production using microencapsulated probiotics

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Abstract: The high genetic potential and substantial feed intake capacity of broiler chickens make them particularly susceptible to intestinal disturbances, which can impair nutrient absorption and increase the availability of resources for opportunistic bacteria. Addressing these challenges, this study evaluated the impact of Fortcell Feed (FF), a multi-strain microencapsulated probiotic, on intestinal health related to pathognes control and production parameters in broilers under commercial farm conditions. Chickens were divided into two groups: one receiving FF and another on a control diet (without FF). Fecal samples were analyzed for pathogens such as *Salmonella enterica*, *Escherichia coli*, and *Clostridium perfringens*, while production metrics, including feed conversion ratio (FCR), weight gain, and mortality, were recorded. FF significantly reduced pathogen abundance, notably eliminating *Salmonella* and reducing de *Staphylococcus* spp*, Pseudomonas* spp*, Klebsiella* spp *y E. Coli*. levels. Broilers fed with FF exhibited better FCR (1.54 vs. 1.60 in controls), higher daily weight gain, and a 48% reduction in mortality, enhancing overall efficiency as measured by the European Efficiency Production Factor. These findings highlight FF's ability to support intestinal integrity, reduce reliance on antibiotics, and promote sustainable poultry production. The study concludes that FF is a valuable additive for improving broiler health and productivity while aligning with global trends toward antibiotic-free farming.

Keywords: Probiotics, Broiler chickens, Intestinal health, Feed efficiency, Poultry production, Pathogens

INTRODUCTION

Broiler chickens, with their high genetic potential and substantial feed intake capacity, are particularly vulnerable to intestinal disorders that compromise digestion, nutrient absorption, and intestinal barrier function. These disruptions not only impact the birds' health but also increase the risk of infections by opportunistic bacteria, challenging the efficiency and sustainability of poultry production systems. Maintaining intestinal integrity—a vital factor for nutrient utilization and disease resistance—is a priority in modern poultry farming. Probiotics have emerged as a promising solution, offering a potential alternative to antibiotic growth promoters while addressing issues of antibiotic resistance.

Extensive research highlights the benefits of probiotics in enhancing growth performance, gut health, and immunity in poultry. Jha et al. (2020) demonstrated that probiotics could modulate intestinal microbiota, stimulate the immune system, and improve nutrient utilization, contributing to enhanced growth performance and reduced environmental waste. Mazanko et al. (2022) showed that supplementation with *Bacillus subtilis* improved broilers' growth performance and immune modulation, highlighting the strain-specific benefits of probiotics. Similarly, Mahfuz et al. (2017) reported that probiotics enhance growth rates, modulate gut flora, and strengthen immune responses. While these findings emphasize the potential of probiotics, more evidence on these results in specific environments as those of commercial farms are necessary (Revista Alfa, 2020; Dialnet, 2017).

This study addresses these gaps by evaluating Fortcell Feed, a multi-strain microencapsulated probiotic, under commercial farm conditions. Specifically, the research examines its effects on feed conversion ratios, weight gain, mortality rates, and pathogen control in broilers. The findings provide evidence of its benefits, contributing to sustainable poultry farming practices and reducing reliance on antibiotics, thereby aligning with global trends toward more sustainable and efficient livestock production.

MATERIALS AND METHODS

Commercial farm procedure

The trial was conducted at a broiler production farm located in San Félix, Medellín, utilizing broilers of Ross 308 genetics. Four batches of one day old chicks were housed in floor pens covering a total area of 1,605 square meters, with an average initial weight of 44 g per chick. Three batches were fed a control diet (T1) with authorized antibiotic growth promoters but without the probiotic consortium, while the fourth batch, was supplemented with the probiotic consortium (Fortcell feed avicultura) added in the feed (the same as the control diet, including the antibiotic growth promoters) at 500 g/ton of feed (T2).

The study aimed to compare the performance of broilers fed isocaloric and isoproteic diets, differing only in the inclusion of the probiotic consortium. Key production parameters were analyzed at the end of the production process, including stocking density, initial and final bird counts, meat production, feed consumption and wastage, average slaughter age, daily weight gain, carcass rejections (weight and percentage), mortality rates, average body weight, feed conversion ratio (FCR), and the European Production Efficiency Factor (EPEF). The initial and final weights of the birds were recorded.

DNA extraction and bioinformatics for pathogen Analysis

Fecal samples were collected from broilers subjected to the two treatments (T1 and T2) in the commercial farm setting. The DNA from the samples was extracted using the NORGEN Stool DNA Isolation Kit following the manufacturer's protocol. DNA samples were quantified using the NanoDrop 2000/2000c spectrophotometer (Thermo Scientific). DNA sequencing was performed by preparing a library with the Nextera XT DNA Library Preparation Kit (Illumina), using a total DNA input of 1 ng. The DNA libraries were purified using AMPure magnetic beads (Beckman Coulter) and quantified using the Qubit 4 fluorometer and the Qubit dsDNA HS Assay Kit. Subsequently, the libraries were sequenced on the Illumina NGS platform targeting the 16S rRNA gene.

The bioinformatic analysis was conducted using CosmosID-HUB, a high-performance k-mer-based data mining algorithm. The pre-computation phase generated a phylogenetic tree of microbes along with variable-length k-mer fingerprint sets uniquely associated with specific branches and leaves of the tree. The sample computation phase searched short-read sequences or contigs from preliminary de novo assemblies against these fingerprint sets, enabling sensitive and highly accurate detection and taxonomic classification of microbial NGS reads.

The resulting data was analyzed to evaluate the abundance of several pathogens including *Escherichia coli* (*Escherichia-Shigella*), *Salmonella enterica*, *Clostridium perfringens*, *Staphylococcus* spp., *Pseudomonas* spp., and *Klebsiella* spp. This analysis provided insights into the impact of Fortcell Feed on the intestinal microbiota and its potential for pathogen control.

RESULTS AND DISCUSSION

Zootechnical performance

Table 1 presents a comparison of key zootechnical parameters for broiler chickens under the two dietary treatments: T1 and T2. It can be seen that over several parameters, a difference arises between both treatments. Some key differences that can be seen are: Higher daily weight gain in T2, lower mortality rates in T2 compared to T1, improved feed conversion in T2 compared to T1 and a superior productivity index in T2 compared to T1.

Table 1. Zootechnical parameters of the chickens in different treatments

Treatment	Chickens housed	Chickens remaining	Chicken meat (Kg)	Feed consumption (g)	Average age at slaughter (d)	Daily weight gain (g/d)	Rejected weight (Kg)	% Rejections	Mortality	Mortality%	Average weight (Kg)	Feed conversion	Productivity index
1	23.415	22.495	52.874	81.360	39,8	59,1	757,1	1,43%	920	3,93%	2,350	1,54	99
1	23.401	22.043	56.899	92.120	40,5	63,7	510	0,90%	1.358	5,80%	2,581	1.62	98
1	22.999	21.163	46.850	76.520	39,3	56,3	436	0,93%	1.836	7,98%	2,214	1.63	83
2	17.850	17.300	43.063	66.400	39,1	63,7	230	0,53%	550	3,08%	2,489	1,54	105

These key differences can be better appreciated in figure 1 that compares the aforementioned key parameters. (including error bars for the triplicates of T1). As it can be observed, for the daily weight gain the T2 group exhibited a daily weight gain of 63.7 grams, compared to an average of 59.7 grams in the T1 group. Statistical analysis yielded a p-value of 0.0899, suggesting a trend towards significance but not reaching conventional thresholds. The difference of approximately 4 grams suggests that the microencapsulated probiotics promote faster growth rates. In the case of mortality percentage, it was significantly lower in T2 (3.08%) compared to T1, where mortality averaged 5.90%. This corresponds to a 48% reduction in mortality rates with T2. The p-value of 0.0071 confirms this as a statistically significant difference, highlighting the potential health benefits of the probiotic. Fo the parameter Feed conversion, the T2 group was more efficient, with a ratio of 1.54 compared to 1.60 in T1. This represents a 5.92% improvement, consistent with reports of enhanced feed utilization in probiotic-supplemented diets. The p-value of 0.0668 indicates marginal significance, suggesting this trend warrants further exploration. Finally, for the productivity index: The productivity index was substantially higher in T2 (105) versus T1 (93.33). This difference is statistically significant, as evidenced by a p-value of 0.0006, emphasizing the cumulative benefits of improved growth rates, feed efficiency, and reduced mortality.

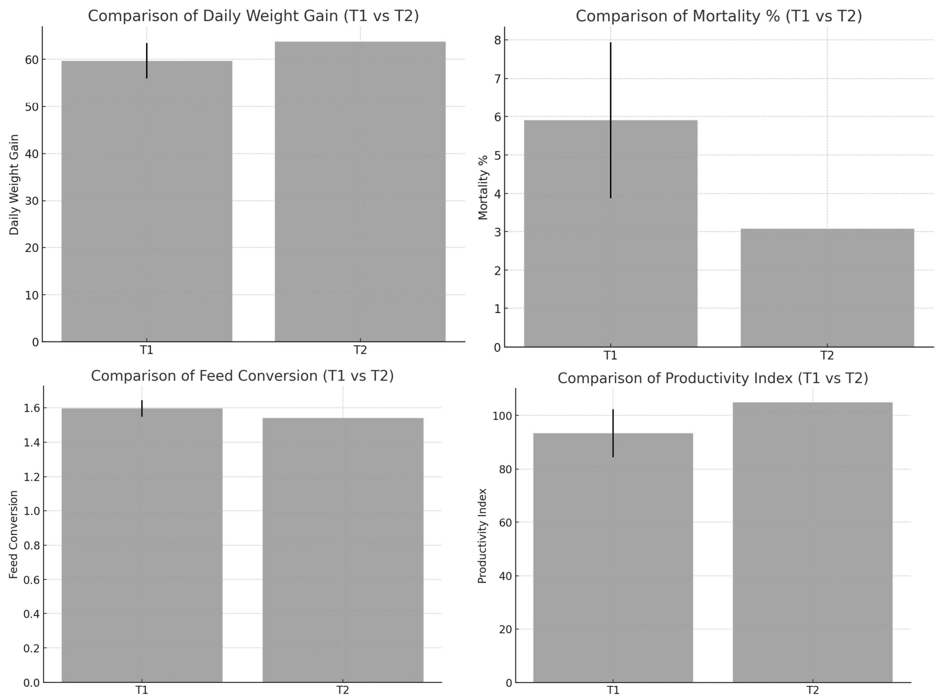

Figure 1. Comparison of key parameters for the two treatments. Error bars represent a value of standard deviation over and under the average value (average ± standard deviation)

According to the results, it can be said that the inclusion of Fortcell Feed Avicultura, a microencapsulated multi-strain probiotic, significantly enhanced the performance of broiler chickens compared to the basal diet without additives. These findings corroborate previous studies reporting the beneficial effects of probiotics on growth performance, feed conversion, and health outcomes.

The higher daily weight gains with the probiotics and more efficient feed conversion are improvements that are economically significant, as they shorten production cycles and reduce feed costs. Such results align with prior research demonstrating that probiotics optimize nutrient absorption and digestion by improving gut microbiota composition. On the other hand, the reduction in mortality and rejection rates with probiotics indicates better overall health, likely due to the immunomodulatory effects of the probiotic. Finally, the higher productivity index observed with probiotics underscores the cumulative impact of improved growth, higher efficiency, and better health. This metric reflects the overall efficiency of the production system and confirms the additive's value in optimizing broiler production.

Pathogens control

In the literature, it can be found several species of bacteria that have been reported as pathogens for broilers. Among the more reported ones are *Escherichia coli* (*Escherichia-Shigella*), *Salmonella enterica*, *Clostridium perfringens*, *Staphylococcus* spp., *Pseudomonas* spp. y *Klebsiella* spp

After the DNA sequencing and bioinformatics analysis, it was observed that *Clostridium perfringens* was absent in both groups of treatments. The comparison of relative abundances of other pathogens in the samples of the two treatments groups are shown in figure 2.

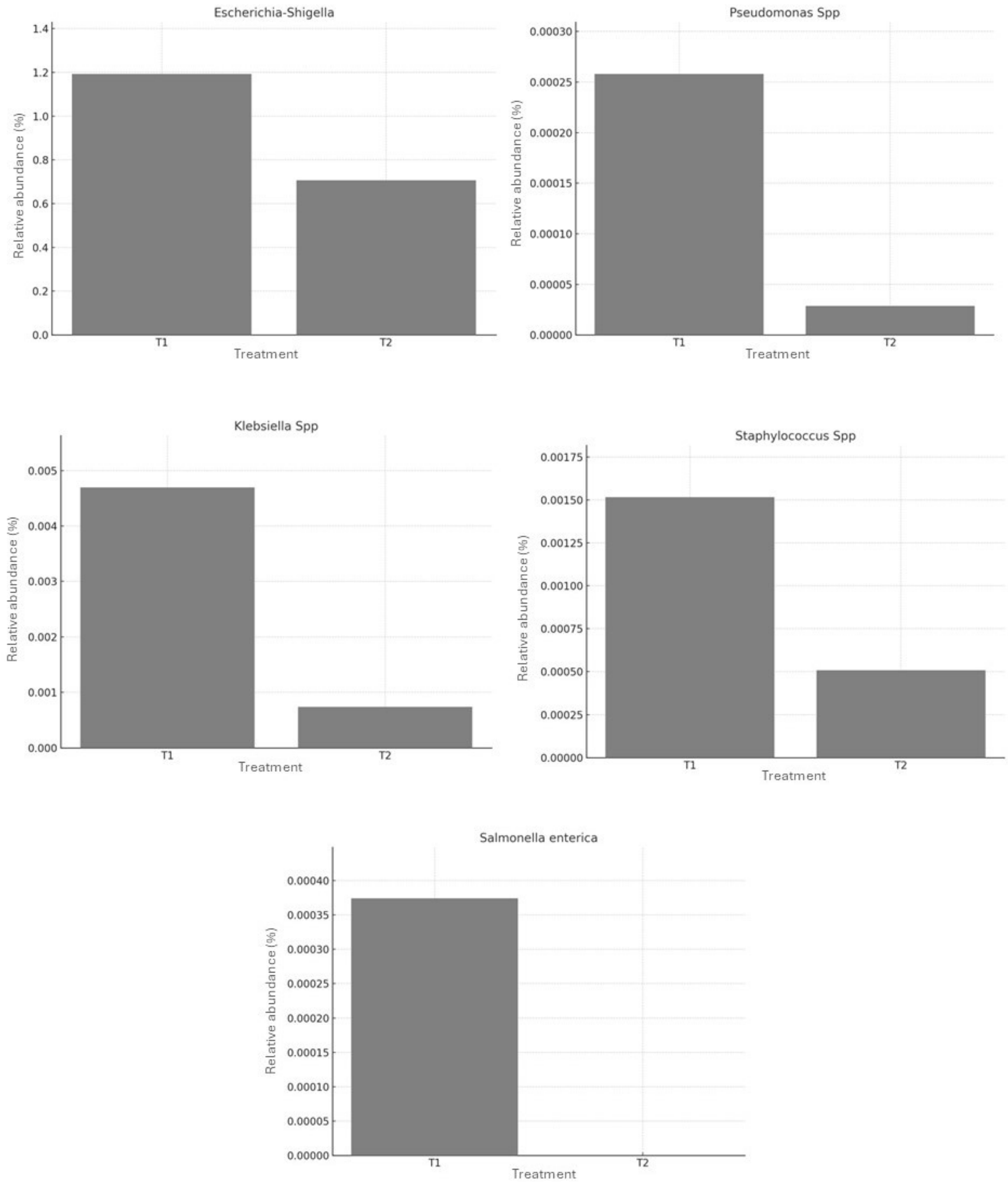

Figure 2. Comparison of the relative abundances of different pathogens present in the two treatments groups.

The results depicted in figure 2 provide insights into the impact the microencapsulated probiotics on the abundance of various pathogenic genera in the microbiota of broiler chickens. In the case of *Escherichia-Shigella* its relative abundance showed no statistically significant difference (p = 0.864) between T1 and T2. While the bar chart suggested a slight reduction in T2, this reduction can be a trend but cannot be confirmed as biologically meaningful without further evidence. Remarkably, *Salmonella enterica* was only found in group T1 (with a relative abundance of 0.00035). For *Staphylococcus* spp., the relative abundance was significantly lower in T2 (p

= 0.048). This supports the effectiveness of FF in reducing opportunistic pathogens associated with infections such as dermatitis, arthritis, and respiratory issues. A reduction of over half of its abundance in T2-fed chickens highlights the potential of probiotics for improving bird health and welfare. In the case of *Pseudomonas* spp. It was almost undetected in T2. The practical absence of this genus in probiotic-treated chickens reflects a strong inhibitory effect, aligning with its role in reducing the prevalence of opportunistic infections that can affect respiratory and systemic health. Finally, although *Klebsiella* spp. abundance was reduced in T2, no statistically significant difference was detected (p = 0.145). As a pathogen linked to systemic infections and antimicrobial resistance, the observed reduction could have important implications for health management, even if further studies are required to confirm statistical significance.

The observed reductions in pathogenic genera with the inclusion of microencapsulated probiotics align with its proposed role in enhancing gut health and controlling enteropathogens in broiler chickens. The significant reduction in *Staphylococcus* spp. and the absence of *Pseudomonas* spp. when adding the microencapsulated probiotics suggest that Fortcell feed avicultura effectively modulates the microbiome, creating an environment less favorable for opportunistic pathogens. This microbiota shift supports better intestinal integrity and nutrient absorption, improving overall health and productivity. While reductions in *Escherichia-Shigella* and *Klebsiella* spp. were observed, their lack of statistical significance indicates variability that warrants further investigation. However, the practical absence of *Pseudomonas* spp. and the significant decrease in *Staphylococcus* spp. abundance underscore Fortcell feed avicultura potential as a targeted intervention for managing gut pathogens. The control of pathogens like *Pseudomonas* spp. and *Staphylococcus* spp. reduces the risk of infections that compromise animal welfare, such as respiratory diseases and septicemia. By lowering pathogen load, the microencapsulated probiotics also reduce stress and immune challenges, contributing to improved productivity and well-being. Finally, The ability of the microencapsulated probiotics to lower pathogen abundance aligns with global efforts to reduce antibiotic use in livestock production. This positions Fortcell Feed as a strategic solution for producers aiming to meet regulatory requirements for antibiotic-free systems while maintaining food safety and productivity.

This study demonstrates that Fortcell Feed avicultura effectively modulates the gut microbiota of broiler chickens, reducing opportunistic pathogens and enhancing intestinal health. These findings support its role as a valuable tool in antibiotic-free poultry production, promoting food safety and improving production efficiency. It is recommended that Fortcell Feed avicultura should be incorporated into broiler chicken diets to improve gut health, reduce pathogen loads, and enhance overall performance. Also, to establish regular microbiota monitoring programs to assess pathogen prevalence and the long-term effects of Fortcell Feed avicultura supplementation.

Implications of Microencapsulation Technology

The microencapsulation of Fortcell Feed ensures the stability and functionality of the probiotic throughout the digestive tract, enhancing its ability to mitigate the growth of pathogenic bacteria and promote gut health. This technology can therefore be essential for the successful application of probiotics, ensuring their effective delivery to the intestine. Additionally, this technology allows probiotics to function synergistically with antibiotics, as demonstrated by the superior performance of the T2 group compared to T1. This suggests that probiotics can complement the use of antibiotics by reducing the required dose and mitigating concerns related to antibiotic resistance.

CONCLUSIONS

The inclusion of Fortcell Feed in broiler diets significantly enhances productive performance of commercial farms, positioning it as a viable alternative to improve productivity and profitability in the poultry industry. When combined in a standard commercial diet, Fortcell Feed demonstrated superior zootechnical outcomes compared to the exclusive use of the standard diet (containing antibiotics). The treatment combining the standard diet (with antibiotics) and Fortcell Feed avicultura (T2) achieved better feed conversion ratios, higher daily weight gain, and reduced mortality, confirming that the microencapsulation technology effectively preserves probiotic viability, allowing positive impacts on intestinal health and animal performance. These results validate Fortcell Feed as an effective tool for controlling enteropathogens, by reducing the presence of several microbial genera such as *Klebsiella* spp., *Salmonella* spp., *Pseudomonas* spp., *Eschericia* y *Staphylococcus* spp. Such reduction allows the optimization of feed efficiency, and a reduction of economic losses due to health issues in broilers. Additionally, the continuous use of Fortcell Feed enhances food safety and minimizes the reliance on antibiotics, benefiting both producers and consumers. These findings emphasize the importance of adopting Fortcell Feed in intensive poultry production systems while encouraging regular microbiome health monitoring and exploring its application in other animal species to maximize its potential benefits.

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