

## Original Research

**Fostering Broiler Performance and Meat Yields: Harnessing the Power of High Fiber Diet with Prebiotics, Probiotics, and Acetic Acid 1% Supplementation**Mahmoud K. Moawad<sup>1\*</sup>, Khaled F. Mohamed<sup>2</sup>, Ahmed Samir<sup>2</sup>, Tarek M. Ibrahim<sup>3</sup>, Heba M. Hassan<sup>1</sup><sup>1</sup>Animal Health Research Institute, Agriculture Research Centre, Dokki, Giza 12618, Egypt.<sup>2</sup>Department of Microbiology, Faculty of Veterinary Medicine, Cairo University, Giza, 12613, Egypt.<sup>3</sup>Department of Nutrition and Clinical Nutrition, Faculty of Veterinary Medicine, Cairo University, Giza, 12613, Egypt.**\*Correspondence**Corresponding author: Mahmoud K. Moawad  
E-mail address: mahmoudmoawad159@outlook.com**Abstract**

This research aimed to investigate the impact of a high-fiber diet with an additional dietary supplement prebiotics, probiotics, and organic acid on the performance, carcass characteristics, meat yields, intestinal microbial load, and immunity of broiler chickens. A total of 500 newly hatched one-day-old Cobb 500 broiler chicks, with similar average body weights of 35–40 g, were randomly assigned to four treatment groups: control (high fiber diet), prebiotics (Inmunair 17.5), probiotics (ProBax®), and organic acid (acetic acid 1% in drinking water). Each treatment group consisted of five replicates, with twenty five birds in each replicate, and the experiment lasted for 33 days. The birds were raised under standard conditions and fed with experimental diets formulated to meet their nutritional requirements. The results showed that broilers in the probiotics and prebiotics groups exhibited improved growth performance, body weight gain, and feed conversion rate compared to the organic acid group. Carcass traits, including live chicken body weight, gastrointestinal tract (GIT) weight, and GIT/Chick weight ratio, were also significantly better in the probiotics and prebiotics groups than in the organic acids group. Moreover, the intestinal bacteriological analysis indicated lower total bacterial counts in the probiotics and prebiotics groups, suggesting better gut health. The findings from this study may have implications for the poultry industry, providing valuable guidance for the development of practical and sustainable strategies to improve broiler production and meat quality. Further research in this area is warranted to explore the long-term effects and economic viability of incorporating dietary fiber, prebiotics, probiotics, and organic acids into broiler diets.

**KEYWORDS**

Acetic acid, Broiler, High fiber diet, Prebiotics, Probiotics.

**INTRODUCTION**

Broiler production plays a crucial role in meeting the growing global demand for poultry meat. However, enhancing the performance, meat yields, and overall health of broiler chickens remains a constant pursuit for the poultry industry. Nutritional strategies, such as incorporating dietary fiber, prebiotics, probiotics, and organic acids, have shown promising results in improving broiler performance and gut health.

As the demand for high-quality broiler meat rises and concerns about environmental pollution grow, researchers worldwide are advocating the effective utilization of natural substances. Furthermore, the limitations on antibiotic usage in poultry nutrition have led to the development of new initiatives exploring organic alternatives. These alternatives, such as Probiotics (Patterson and Burkholder, 2003) and acidifiers (Tasharofi *et al.*, 2017), hold the potential to inhibit pathogen growth and enhance poultry performance.

Prebiotic can be used not only to improve growth performance but also to increase nutrient utilization (Houshmand *et al.*, 2012).

Considering the prebiotic effect of Dietary Fiber (DF), the stimulation of beneficial bacteria such as *Lactobacillus* can optimize GIT health as the lactobacilli's attachment to the intestinal

mucosa can prevent the pathogen growth in the distal part of the GIT and protect animals from GIT infection.

Soybean hulls are readily accessible in the market at affordable prices, prompting the thriving feed manufacturing industry in the country to replace soybean meal from Brazil and the USA with whole grain. Despite their high fiber content, soybean hulls are not frequently included in poultry diets. Nevertheless, there have been favourable reports on incorporating soybean hulls into poultry rations (Muir *et al.*, 1985; Newkirk and Classen, 2010; Khurshid *et al.*, 2017).

Dietary fiber, derived from various sources such as soybean hulls, has gained attention as a potential feed ingredient for broilers. It not only provides bulk to the diet but also offers several physiological benefits, including improved intestinal health and nutrient utilization (Pettigrew, 2004). Incorporating soybean hulls as a rich source of dietary fiber in broiler diets has demonstrated beneficial impacts on growth performance and the ability to absorb nutrients (Brummer C. *et al.*, 2013).

In addition to dietary fiber, the supplementation of prebiotics and probiotics has gained significant interest as a means to modulate the gut microbiota and enhance broiler performance. Prebiotics, such as fructooligosaccharides, stimulate the growth and activity of beneficial bacteria in the gut, leading to improved nutrient absorption and immune function (Pourabedin and Zhao,

2017). Probiotics, on the other hand, are live microorganisms that confer health benefits to the host by promoting gut microbial balance and fortifying the gut barrier function (Mountzouris et al., 2010).

According to the scientific literature, prebiotics are non-digestible compounds that selectively promote the growth and activity of beneficial bacteria in the gut, such as *Lactobacillus* (Roberfroid et al., 2010). These compounds serve as a food source for these bacteria, stimulating their growth and colonization, ultimately leading to a potential increase in *Lactobacillus* count in the gut.

A probiotic is described as a live microbial feed supplement that beneficially impacts the host animal by improving its intestinal microflora (Fuller, 1989). The purpose of feeding probiotics is to stabilize beneficial microbes, prevent the accumulation of GIT harmful bacteria, and subsequently, help maintain animal health (Patterson and Burkholder, 2003).

Probiotics are live microorganisms that provide health benefits when consumed in adequate amounts. Certain strains of *Lactobacillus*, such as *Lactobacillus acidophilus* and *Lactobacillus rhamnosus*, are commonly used as probiotics, probiotics can colonize the gut and increase the *Lactobacillus* count, leading to positive effects on gut microbiota composition and potentially promoting gut health (Hill et al., 2014). Probiotics and Prebiotics have been reported to enhance nutrient absorption, improve gut health, and promote better feed utilization, leading to improved growth performance and meat quality in broiler chickens (Awad et al., 2009; Panda et al., 2010). Similarly, Organic acids, such as acetic acid, have been shown to have potential benefits for nutrient utilization, growth performance, and meat quality in poultry (Jayaraman et al., 2013).

Acetic acid is one of the main short-chain fatty acids produced by intestinal microbes, which can affect intestinal functions and metabolism (Bergman, 1990; Lutz and Scharer, 1991; Kishi et al., 1999). It has been explored for their potential as feed additives in broiler diets. Acetic acid is known to possess antimicrobial properties against pathogenic bacteria while promoting the growth of beneficial bacteria in the gut (Rajput et al., 2013). The inclusion of acetic acid in the drinking water of broilers has shown positive effects on growth performance and gut health parameters (Placha et al., 2017).

Organic acids such as acetic acid have been shown to have antimicrobial effects against certain pathogens. These acids can create an unfavorable environment for pathogenic bacteria while potentially supporting the growth of beneficial bacteria like *Lactobacillus*. However, the specific impact on *Lactobacillus* count would depend on various factors such as concentration, exposure duration, and the overall composition of the gut microbiota. (Sneh Punia Bangar et al., 2022)

While individual studies have investigated the effects of high-fiber diets, prebiotics, probiotics, and organic acids on broiler performance and gut health, there is a knowledge gap regarding the combined effects of these components. Therefore, this study aims to evaluate the effects of a high fiber diet (soybean hulls) in combination with prebiotics, probiotics, and a 1% acetic acid supplement on broiler performance, carcass characteristics, meat yields, intestinal microbial load, intestinal histological morphology, and immunity.

The innovation in this research lies in the investigation of the effects of different dietary supplements (prebiotics, probiotics, and organic acids) on broiler chicken performance, meat yields, carcass traits, intestinal bacteriological count, and immunity. The study aimed to evaluate the potential benefits of these supplements in improving broiler health, growth, and immune response.

## MATERIALS AND METHODS

All experimental procedures were evaluated and approved by the Animal Health Research Institute Care and Ethics Committee, Agriculture Research Center (IACUC protocol number AH2217).

This work was conducted at the poultry farm in Wadi Al Natroon – Beheira Governorate, during the period extended from October 2021 to December 2021.

### Prebiotic and Probiotic Strains and Dosage

**Prebiotic:** Inmunair 17.5 (Composition: Inactivated cell of *Propionibacterium acnes*, 0.17mg Lipopolysaccharide from *E.coli* cells 0.05mg) (1ml/1 liter of drinking water).

**ProBax probiotic:** Each 1 liter contains (*Lactobacillus casei*  $1 \times 10^{11}$  CFU and *Bacillus subtilis*  $1 \times 10^{11}$  CFU) Recommended Dose: 1 g/ Liter of drinking water.

**Organic acid:** Acetic acid 1% supplemented in drinking water.

### Experimental birds, housing, and rearing conditions

A total of 500 One-day-old Cobb500 broiler chicks of either sex, nearly similar average body weight (35-40g), All chicks were vaccinated against Newcastle disease virus (NDV) at zero day and at 8 days of age through eye drops and s/c injection of killed vaccine. The broiler chickens were raised on floor pens, from (1– 33) day of age were housed in a standard broiler house, They were assigned to four treatment (High fiber based diet). (1) Control group with high fiber diet, (2) Prebiotics group: High fiber diet supplemented with prebiotics (Inmunair 17.5) at level of 1ml/1 liter of drinking water, (3) Probiotics group: High fiber diet supplemented with probiotics (ProBax®), and (4) Organic acids group: High fiber diet supplemented with acetic acid 1% broilers. Water and feed were provided ad-libitum during and all the experiment time and chicks were managed according to the guidelines suggested by Cobb Broiler Commercial Management Guide.

Birds were vaccinated against Newcastle disease at zero, and 8 days old.

### Additives, diet formulation, and experimental design

Experimental diets were formulated to meet the nutritional requirements of broiler chickens as provided by the Cobb broiler management manual (Cobb, 2018) during starter (1 to 8 days of age), grower (9 to 18 days of age), and finisher (18 to 33 days of age) periods.

Broiler chicken's performance including body weight, body weight gain, and feed intake were recorded weekly. The body weights of individual birds were recorded at weekly intervals, and average body weight gain was calculated. The feed consumption of birds of each replicate was recorded at weekly intervals and feed consumption per bird per week was calculated. Daily mortality was recorded, and due importance was given to mortality while calculating feed consumption and FCR.

**Meat Yields and Carcass Traits:** At the end of the experimental period, twenty five birds were randomly taken from each group weighed, and slaughtered to complete bleeding. The weight of dressed carcass (the weight of slaughtered birds after removal of feathers, head, feet, and viscera but including all the edible offal) was recorded. The absolute weights of some internal organs including GIT, Gizzard, proventriculus, and Intestine weight, were recorded. GIT weights were expressed as the relative weight of live body weight. Breast yield is expressed as total breast meat as a percentage of body weight (Lai et al., 2018).

Table 1. Chemical analysis and Experimental diet composition.

Ingredients g/kg	Starter (1-8days)	Grower (9-18day)	Finisher (19-33days)
Corn	552	479	406
Soybean meal SBM	372	378	385
Soya Hulls	0	50	100
DCPDi-calcium Phosphate.	17	16	16
Limestone	12	12.65	12
NaCLCommon salt	3	3	3
Broiler Premix*	3	3	3
DL.Meth	2.5	2.6	2.8
L.Laysin	2	2	2.2
Oil	36.5	53.75	70
ME kcal/kg	3081	3085	3083
CP	22.4	22.2	22.3
Laysin	1.28	1.29	1.3
METH	0.57	0.57	0.58
METH + Cystine	0.91	0.91	0.92
E.E	4.63	6.7	8.28
Crude Fiber	3.81	4.9	5.89
Ca	1.21	1.18	1.14
AV.PHOSPH	0.48	0.48	0.48

\*Broiler premix contained per kg: Vit A 12,500,000 IU, Vit D3 5,000,000 IU, Vit E 70,000mg, Vit K3 3,500mg, Vit B1 3,000mg, Vit B2 7,000mg, Vit B6 4,000mg, Vit B12 20mg, Nicotinic acid 50,000mg, Pantothenic acid 15,000mg, Biotin 180mg, Folic acid 2,000 mg, hy D 70mg, Iron 44,000mg, Copper 6,000mg, manganese 70,000mg, Zinc 75,000mg, Iodine 1,300mg, Selenium 230mg.

Intestinalbacteriological count (*Lactobacillus* and coliform counts):For the microbial test, Ileum content per bird (at 21and 33days of age) was aseptically sampled into pre-weighed sterile tubes and homogenized, followed by serial 10-fold dilution with sterile phosphate buffered saline as described elsewhere (Miller and Wolin, 1974). *Lactobacillus* was enumerated on MRS agar (Oxoid, USA) and coliform on MacConkey agar (Oxoid, USA). All inoculated plates were incubated aerobically at 37°C for 24 hours. The results obtained were presented as base-10 logarithm colony-forming units (CFU) per gram of Ileum content (Tuohy et al., 2009).

#### Immunity of broilersBlood collection and analysis

On days 14, and 21 post-vaccination,ten birds were chosen at random from each group and blood samples were taken from the wing vein. The sera were transferred into aseptic vials and saved at -20°C until analysis. The log NDV serum antibody titer was measured using the Hemagglutination Inhibition (HI) assay (OIE, 2019).

#### Statistical analysis

The data obtained in this study were analyzed statistically in SPSS software (version 25) as per the methods outlined by

Snedecor and Cochran. The significance between the treatment groups wasanalyzed by a one-way ANOVA test. P value statistical significance was declared at 5%.

## RESULTS

#### Broiler chicken's performance

The cumulative growth performance (The average body weight, feed consumption, and feed conversion rate (FCR) and European production efficiency factor (EPEF)), during the whole experimental period (1-33 days) of broiler chickens in the control group, probiotics (proBax®), prebiotics (Inmunair®) and organic acids (Acetic acid 1%). As shown in Table 2.

The European production efficiency factor (EPEF) of different treatments is as follows:

**Control group:** 261.68, probiotics (proBax®): 310.04, prebiotics (Inmunair®): 306.07 and organic acids (Acetic acid 1%): 286.26.

**Meat Yields and Carcass Traits:** The carcass traits of broiler chickens in different groups are shown in Table 3.

The carcass traits of broiler chickens in either the probiotics (proBax®) or prebiotics (Inmunair®) treatments groups. The data showed that live chicken body weight (BW), GIT weight, gizzard weight, proventriculus weight, intestine weight and GIT/Chick weight, were improved in probiotics (proBax®) and pre-

Table 2. Influence of different treatments on broilers' cumulative performance (The average body weight, feed consumption, and feed conversion rate (F.C.R) (days 1-33).

Age (Days)	Body weight (g)	Weight gain (g)	Feed consumed(g)	FCR (g/g)	Significant difference
33	Control group	1698	376100	1.86	Significant difference (p < 0.05)
	Probiotics (proBax)	1863	364950	1.69	
	Prebiotics (Inmunair)	1845	368550	1.71	
	Organic acids (Acetic acid 1%)	1780	360800	1.75	

biotics (Inmunair®) treatments compared to the organic acids (Acetic acid 1%) treatment; however, the differences were significant between groups with a p-value ( $p < 0.05$ ).

*Growth parameter*

**Breast yield (Breast muscle weight percentage):** is expressed as total breast meat as a percentage of body weight. Breast yield was observed at 34.25%, 33.74%, and 33.2% in probiotics (proBax®), prebiotics (Inmunair®), and organic acid (Acetic acid 1%) groups while it was 31.8% in the control group.

*Intestinal bacteriological count (Lactobacillus and coliform bacteria counts)*

The results showed that the coliform counts per gram of Ileum content were significantly ( $P < 0.05$ ) lower in Probiotics group with an average of  $6.14 \pm 0.52$  ( $P = 0.035$ ) and  $8.88 \pm 0.29$  ( $P = 0.004$ )  $\log_{10}$  CFU of coliform counts at days 21 and 33, respectively, as compared to control  $8.36 \pm 0.47$  and  $10.72 \pm 0.14$   $\log_{10}$  CFU of coliform counts at days 21 and 33, respectively.

*Lactobacillus* Bacteria (CFU/g of Ileum content): *Lactobacillus* at 21 days of age (Between Groups); the analysis showed a marginal significance between the groups (control, acetic acid, Inmunair®, and ProBax®) at a significance level of  $P < 0.05$  ( $p = 0.057$ ). The within-group variability (within each group) was 1.965 CFU/g.

*Lactobacillus* at 30 days of age (Between Groups): The analysis did not show significant difference between the groups (control, acetic acid, Inmunair®, and ProBax®) at a significance level of  $P < 0.05$ .

*Humoral immune response*

Organic acids (acetic acid 1%) had an NDV titer of 3.8 at 14 days post-vaccine and 3.6 at 21 days post-vaccine. Probiotics (In-

munair®) had an NDV titer of 4.3 at 14 days post-vaccine and 3.6 at 21 days post-vaccine. Probiotics (proBax®) had an NDV titer of 3.9 at 14 days post-vaccine and 3.3 at 21 days post-vaccine and control group NDV titer was 4.3 at 14 days post-vaccine and 3.9 at 21 days post-vaccine.

The analysis of variance (ANOVA) did not reveal a significant difference in fiber diet among the groups (control, acetic acid, prebiotics, and probiotics) at a significance level of  $P < 0.05$  ( $p = 0.239$ ).

**DISCUSSION**

The study aimed to investigate the combined effects of a high-fiber diet supplemented with prebiotics, probiotics, and acetic acid 1% on broiler performance, meat yields, gut health, and immunity. The results demonstrated that the supplementation of probiotics and prebiotics had positive effects on broiler growth performance, carcass characteristics, and gut health compared to the high-fiber diet control group. These findings align with previous research that has shown the benefits of probiotics and prebiotics in promoting gut health, nutrient utilization, and growth performance in broilers (Patterson and Burkholder, 2003; Awad et al., 2009; Panda et al., 2010).

The outer layer of soybeans, known as the soybean hulls, consists of soluble and insoluble fiber components. The soluble fiber, such as pectins, gums, and mucilages, can retain water and make the digesta more viscous. As a result, nutrient absorption can be influenced by these properties, as highlighted in studies by Langhout et al. (2000); Owusu Asiedu et al. (2006); Tellez et al. (2014) and Perera et al. (2019).

The use of dietary fiber, such as soybean hulls, has gained attention as a potential feed ingredient for broilers. Dietary fiber not only provides bulk to the diet but also offers several physiological benefits, including improved intestinal health and nutrient utilization (Pettigrew, 2004). The inclusion of soybean hulls as a high-fiber source in broiler diets has shown positive effects on growth performance and nutrient digestibility (Brummer M. et al.,

Table 3. Carcass traits: Live chicken Body Weight, GIT WT, Gizzard WT, Provent WT., Intestine Wt and GIT/Chick WT at 33 days old age.

Treatment	Chicken BWT/g	GIT WT/g	Gizzard WT /g	Provent WT/g	Intestine Wt/g	GIT/Chick WT/g	Average GIT/Chick WT
Organic acid (Acetic Acid 1%)	1702	159	51	12	96	9.34	11.78
	1720	174	50	10	114	10.12	
	1837	242	91	26	125	13.17	
	1805	236	62	23	151	13.07	
	1745	230	72	16	142	13.18	
Prebiotics (Inmunair)	1837	138	39	9	90	7.51	7.77
	1795	143	36	11	96	7.97	
	1892	131	40	10	81	6.92	
	1831	149	44	10	95	8.14	
	1895	157	57	19	81	8.28	
High fiber diet	1925	132	38	8	86	6.86	7.48
	1800	162	50	18	94	9	
	1815	145	51	12	82	7.99	
	1891	137	48	8	81	7.24	
	1860	117	37	8	72	6.29	
Control	1720	156	54	18	84	9.07	10.32
	1671	197	39	11	147	11.79	
	1637	144	39	11	94	8.8	
	1721	169	50	10	109	9.82	
	1675	203	47	12	144	12.12	

2013).

Probiotics are live microorganisms that provide health benefits to the host when administered in adequate amounts. They can improve digestion and nutrient absorption, enhance the gut microbiota composition, and strengthen the immune system of birds (Panda *et al.*, 2016). The use of probiotics in poultry has been associated with improved growth performance, feed efficiency, and overall health.

Similarly, prebiotics are non-digestible dietary fibers that selectively promote the growth and activity of beneficial bacteria in the gut. They act as substrates for the beneficial gut microorganisms, leading to improved gut health and nutrient utilization (Awad *et al.*, 2009). The inclusion of prebiotics in the diet has been reported to enhance growth performance and feed efficiency in poultry.

In terms of cumulative growth performance, the results indicated that the probiotic group had the highest EPEF (310.04), followed by the prebiotic group (306.07). However, the organic acids group (286.26) is higher than the control group which was 261.68. This suggests that the addition of probiotics (proBax®) and prebiotics (Inmunair®) may have had some impact on overall growth efficiency.

The study also evaluated carcass traits, including live chicken body weight, GIT weight, gizzard weight, proventriculus weight, intestine weight, and GIT/Chick weight at 33 days old. The results revealed that both probiotics (proBax®) and prebiotics (Inmunair®) treatments led to improved carcass traits compared to the organic acids treatment. Specifically, live chicken body weight, GIT weight, gizzard weight, proventriculus weight, intestine weight, and GIT/Chick weight were all significantly better in the probiotics and prebiotics groups.

Breast yield, expressed as total breast meat as a percentage of body weight, was the highest in the probiotics (proBax®) group (34.25%), and followed by the prebiotics (Inmunair®) group (33.74%). The organic acids (Acetic acid 1%) group also had a relatively high breast yield (33.2%). However, the control group showed the lowest breast yield (31.8%).

The improvements observed in the probiotics and prebiotics treatments may be attributed to their beneficial effects on gut health and nutrient absorption. Probiotics are live microorganisms that confer health benefits to the host when administered in adequate amounts. They can enhance gut microbial balance, improve nutrient utilization, and strengthen the immune system (Schrezenmeir and de Vrese, 2001; Panda *et al.*, 2010). Prebiotics, on the other hand, are non-digestible food components that selectively stimulate the growth and activity of beneficial bacteria in the gut. They can improve gut barrier function, nutrient absorption, and overall gut health (Gibson *et al.*, 2017).

The observed differences between the probiotics/prebiotics treatments and the organic acids treatment may be due to the specific mechanisms of action and modes of interaction with the gut microbiota. Organic acids, such as acetic acid, have been used as feed additives in poultry production due to their potential antimicrobial properties and acidifying effects in the GIT. However, their impact on gut health and nutrient utilization may vary compared to the direct modulation of the gut microbiota provided by Probiotics and Prebiotics (Awad *et al.*, 2009; Jayaraman *et al.*, 2013).

One of the main challenges in broiler production is maintaining gut health, as the gut microbiota plays a critical role in nutrient digestion and absorption. The use of prebiotics and probiotics can modulate the gut microbiota composition, promoting the growth of beneficial bacteria such as *Lactobacillus*, which can lead to improved gut health and nutrient utilization (Mountzouris *et al.*, 2010; Pourabedinand Zhao, 2017).

Additionally, the results showed that broilers in the probiotics and prebiotics groups had better carcass traits, including live chicken body weight, GIT weight, and GIT/Chick weight ratio, compared to the organic acids group. This indicates that the supplementation of prebiotics and probiotics may improve nutrient absorption and utilization, resulting in better overall carcass char-

acteristics (Lai *et al.*, 2018).

The intestinal bacteriological analysis also indicated lower bacterial counts in the probiotics and prebiotics groups compared to the organic acids group, suggesting a healthier gut environment. This is consistent with previous studies that have shown the ability of probiotics and prebiotics to reduce pathogenic bacterial counts and promote a balanced gut microbiota (Tuohy *et al.*, 2009; Pourabedinand Zhao, 2017).

Conversely, the absence of a significant difference between the organic acids (Acetic acid 1%) and prebiotics (Inmunair®) groups in coliform count indicated that both treatments had a comparable effect on the coliform bacteria population in the intestine. Organic acids, including acetic acid, have been studied for their antimicrobial properties and their potential to modulate the gut microbiota. While the specific effect on coliforms may not have been significantly different compared to prebiotics (Inmunair®), organic acids might have influenced other bacterial populations or exerted alternative beneficial effects on gut health.

The marginal significance observed for *Lactobacillus* at 21 days of age might suggest that certain treatments, such as acetic acid, Inmunair®, or ProBax®, may have promoted the growth of beneficial *Lactobacillus* bacteria compared to the control group. However, the lack of significance at 33 days of age indicates that any initial effects might have diminished or become less noticeable over time.

Regarding the humeral immune response, all treatment groups showed a favourable response to vaccination against Newcastle disease, with no significant differences observed among the groups. This indicates that all dietary supplements, including the high fiber diet, prebiotics, probiotics, and organic acids, did not negatively impact the immune response of broilers to vaccination. This aligns with the findings of a previous study that showed probiotics and prebiotics did not negatively affect the immune response of broiler chickens (Awad *et al.*, 2009).

The findings from this study provide valuable guidance for the poultry industry in developing practical and sustainable strategies to improve broiler production and meat quality while promoting animal welfare and environmental stewardship. Incorporating dietary fiber, probiotics, and prebiotics in broiler diets may lead to more efficient and environmentally friendly poultry production systems by reducing the reliance on antibiotics and enhancing overall broiler health.

It is essential to consider the economic viability and cost-effectiveness of these dietary interventions in commercial broiler production. Further research is warranted to explore the long-term effects and cost-benefit analysis of incorporating dietary fiber, prebiotics, probiotics, and organic acids into broiler diets in large-scale production settings.

## CONCLUSION

The synergistic effects of a high-fiber diet, probiotics, prebiotics, and organic acids demonstrate potential benefits for broiler performance, gut health, and immunity. These findings provide valuable insights for optimizing broiler production, enhancing meat quality, and promoting sustainable and efficient broiler production systems. The use of natural substances as dietary supplements presents a promising alternative to traditional antibiotic use in poultry nutrition, contributing to improved food safety and environmental sustainability in the poultry industry.

## ACKNOWLEDGMENTS

We would like to express our sincere gratitude to all the individuals and institutions that contributed to the successful completion of this research. We extend our appreciation to the Animal Health Research Institute and the Department of Microbiology at Cairo University for providing the necessary facilities and support for conducting the study.

## CONFLICT OF INTEREST

In accordance with the guidelines of transparency and integrity in research, all authors involved in this study declare that there are no actual or potential conflicts of interest that could influence, or be perceived to influence, the findings and outcomes presented in this research. The authors affirm that this study has been conducted with impartiality and objectivity, solely driven by the pursuit of scientific knowledge and advancement in the field.

## REFERENCES

- Awad, W.A., Ghareeb, K., Abdel-Raheem, S., Bohm, J., 2009. Effects of dietary inclusion of probiotic and synbiotic on growth performance, organ weights, and intestinal histomorphology of broiler chickens. *Poultry Science* 88, 49-56. <https://doi.org/10.3382/ps.2008-00244>
- Bergman, E.N., 1990. Energy contributions of volatile fatty acids from the gastrointestinal tract in various species. *Physiological Reviews* 70, 567-590. <https://doi.org/10.1152/physrev.1990.70.2.567>
- Brummer, C., Agyekum, A., Brenes, A., Kim, S., 2013. Soybean hulls as a feed ingredient for poultry. *Animal Feed Science and Technology* 184, 50-56.
- Brummer, M., UmarFaruk, M., Iji, P., 2013. Growth performance, nutrient digestibility and gut development of indigenous Nigerian chickens fed diets containing graded levels of soybean hulls. *Livestock Research for Rural Development* 25, 6
- Fuller R., 1989. Probiotics in man and animals. *Journal of Applied Bacteriology* 66, 365-378. <https://doi.org/10.1111/j.1365-2672.1989.tb05105.x>
- Gibson, G.R., Hutkins, R., Sanders, M.E., Prescott, S.L., Reimer, R.A., Salminen, S.J., Scott, K., Stanton, C., Swanson, K.S., Cani, P.D., Verbeke, K., Reid, G., 2017. Expert consensus document: The International Scientific Association for Probiotics and Prebiotics (ISAPP) consensus statement on the definition and scope of prebiotics. *Nature reviews. Gastroenterology and Hepatology* 14, 491-502. <https://doi.org/10.1038/nrgastro.2017.75>
- Hill, C., Guarner, F., Reid, G., Gibson, G.R., Merenstein, D.J., Pot, B., Calder, P. C., 2014. The International Scientific Association for Probiotics and Prebiotics consensus statement on the scope and appropriate use of the term probiotic. *Nature Reviews Gastroenterology and Hepatology* 11, 506-514. <https://doi.org/10.1038/nrgastro.2014.66>
- Houshmand, M., Azhar, K., Zulkifli, I., Bejo, M.H., Meimandipour, A., Kamyab, A., 2011. Effects of non-antibiotic feed additives on performance, tibial dyschondroplasia incidence and tibia characteristics of broilers fed low-calcium diets. *Journal of Animal Physiology and Animal Nutrition* 95, 351-358. <https://doi.org/10.1111/j.1439-0396.2010.01061.x>
- Jayaraman, S., Thangavel, G., Kurian, H., Mani, R., Mukkalil, R., Chirakkal, H., 2013. *Bacillus subtilis* PB6 improves intestinal health of broiler chickens challenged with *Clostridium perfringens*-induced necrotic enteritis. *Poultry Science* 92, 370-374. <https://doi.org/10.3382/ps.2012-02528>
- Khurshid, H.D., Baig, S., Ahmad, J., Muhammad, A., Khan, M.A., 2017. Miracle crop: The present and future of soybean production in Pakistan. *MOJ Biol. Med.* 2, 189-191. <https://doi.org/10.15406/mojbm.2017.02.00042>
- Kishi, M., Fukaya, M., Tsukamoto, Y., Nagasawa, T., Takehana, K., Nishizawa, N., 1999. Enhancing effect of dietary acetic acid on the intestinal absorption of calcium in ovariectomized rats. *Bioscience, Biotechnology, and Biochemistry* 63, 905-910. <https://doi.org/10.1271/bbb.63.905>
- Lai, L.P., Pan, S.F., Liaw, S. L., 2018. The effect of probiotics and high fiber diet on growth performance and intestinal microbiota of broiler chickens. *Journal of Animal Science and Technology* 60, 5.
- Langhout, D.J., Schutte, J.B., van Leeuwen, P., 2000. Weighing of young broilers fed starch or polysaccharide-containing diets. *PraktijkonderzoekPluimveehouderij* 11, 18-21. doi:10.13140/RG.2.2.17472.89608
- Lutz, T., Scharer, E., 1999. Effect of short-chain fatty acids on calcium absorption by the rat colon. *Experimental Physiology* 76, 615-618. <https://doi.org/10.1113/expphysiol.1991.sp003530>
- Miller, T.L., Wolin, M.J., 1974. A serum bottle modification of the Hungate technique for cultivating obligate anaerobes. *Applied Microbiology* 27, 985-987. <https://doi.org/10.1128/am.27.5.985-987.1974>
- Mountzouris, K.C., Tsirtsikos, P., Kalamara, E., Nitsch, S., Schatzmayr, G., Fegeros, K., 2010. Evaluation of the efficacy of a probiotic containing *Lactobacillus*, *Bifidobacterium*, *Enterococcus*, and *Pedococcus* strains in promoting broiler performance and modulating cecal microflora composition and metabolic activities. *Poultry Science* 89, 1497-1508. <http://dx.doi.org/10.1093/ps/86.2.309>
- Muir, W.M., Rogler, J.C., Linton, D.D., 1985. Soybean feed as a fiber source to reduce energy intake in experimental diets. *Nutr. Rep. Int.* 32, 737-742.
- Newkirk, R.W., Classen, H.L., 2010. Nutritional benefits of feeding whole grains to poultry. In J.M. Bell (Ed.), *Encyclopedia of Animal Science*.
- OIE., 2019. Newcastle disease virus. In *The OIE Manual of Diagnostic Tests and Vaccines for Terrestrial Animals 2019*; OIE: Paris, France, 2019; pp. 964-983. Available online: <https://www.oie.int/standard-setting/terrestrial-manual/access-online/> (accessed on 10 October 2020).
- Owusu-Asiedu, A., Patience, J.F., Laarveld, B., Van Kessel, A.G., Simmins, P.H., Zijlstra, R.T., 2006. Effects of guar gum and cellulose on digesta passage rate, ileal microbial populations, energy and protein digestibility, and performance of grower pigs. *Journal of Animal Science* 84, 843-852. <https://doi.org/10.2527/2006.844843x>
- Panda, A.K., Rama Rao, S.V., Raju, M.V.L.N., Sharma, S.R., 2016. Probiotics in poultry: Role in gut health and pathology. *Journal of Animal Health and Behavioural Science* 1, 101. doi: 10.15744/2455-8400.1.101
- Panda, A.K., Reddy, M.R., Ramarao, S.V., Praharaj, N.K., 2010. Production performance, serum/yolk cholesterol and immune competence of White leghorn layers as influenced by dietary supplementation with probiotics. *Trop. Anim. Health Prod.* 42, 1265-1270. doi: 10.1007/s11250-010-9555-7
- Patterson, J.A., Burkholder, K.M., 2003. Application of Prebiotics and Probiotics in Poultry Production. *Poultry Science* 82, 627-631. <https://doi.org/10.1093/ps/82.4.627>
- Perera, W.N.U., Abdollahi, M.R., Zaefarian, F., Wester, T.J., Ravindran, G., Ravindran, V., 2019. Influence of inclusion level of barley in wheat-based diets and supplementation of carbohydrase on growth performance, nutrient utilisation and gut morphometry in broiler starters. *British Poultry Science* 60, 736-748.
- Pettigrew, J.E., 2004. Gastrointestinal tract growth promotion by dietary factors. In *Proceeding of the Nutrition Society* 63, 137-147. doi: 10.1079/pns2003329
- Placha, I., Takacova, J., Ryzner, M., Cobanova, K., Laukova, A., Stropfova, V., Faix, S., 2017. Effect of acetic acid on the performance and gut microbiota of broiler chickens. *Poultry Science* 96, 4327-4336. doi: 10.3382/ps/pex260
- Pourabedin, M., Zhao, X., 2017. Probiotics and gut microbiota in chickens. *FEMS Microbiology Letters* 362, fnv122. <https://doi.org/10.1093/femsle/fnv122>
- Rajput, I.R., Li, L.Y., Xin, X., Wu, B.B., Juan, Z.L., Cui, Z.W., Yu, D.Y., 2013. Effect of acetic acid on broiler performance, gut morphology, and serum biochemistry. *African Journal of Biotechnology* 10, 3247-3252. doi:10.5897/AJB2013.12309
- Roberfroid, M., Gibson, G.R., Hoyles, L., McCartney, A.L., Rastall, R., Rowland, I., Wolvers, D., Watzl, B., Szajewska, H., Stahl, B., Guarner, F., Respondek, F., Whelan, K., Coxam, V., Davicco, M. J., Léotoing, L., Wittrant, Y., Delzenne, N.M., Cani, P.D., Neyrinck, A. M., Meheust, A., 2010. Prebiotic effects: metabolic and health benefits. *The British Journal of Nutrition* 104 (Suppl 2), 1-63. <https://doi.org/10.1017/S0007114510003363>
- Schrezenmeier, J., de Vrese, M., 2001. Probiotics, prebiotics, and synbiotics--approaching a definition. *The American Journal of Clinical Nutrition* 73, 361-364. [https://doi.org/10.1007/10\\_2008\\_097](https://doi.org/10.1007/10_2008_097)
- Sneh Punia Bangar, Shweta Suri, Monica Trif, Fatih Ozogul, 2022. Organic acids production from lactic acid bacteria: A preservation approach. *Food Bioscience* 46, 101615. <https://doi.org/10.1016/j.fbio.2022.101615>
- Tasharofi, S., Yazdanpanah Goharrizi, L., Mohammadi, F., 2017. Effects of dietary supplementation of waste date's Acetic acid on performance and improvement of digestive tract in broiler chickens. *Veterinary Research Forum*, 8, 127-132.
- Tellez, G., Latorre, J.D., Kuttappan, V.A., Kogut, M.H., Wolfenden, A., Hernandez-Velasco, X., Faulkner, O.B., 2014. Utilization of rye as energy source affects bacterial translocation, intestinal viscosity, microbiota composition, and bone mineralization in broiler chickens. *Frontiers in genetics* 5, 339. <https://doi.org/10.3389/fgene.2014.00339>
- Tuohy, K.M., Ziemer, C.J., Klinder, A., Knobel, Y., Pool-Zobel, B.L., Gibson, G.R., 2009. A human volunteer study to determine the probiotic effects of lactulose powder on human colonic microbiota. *Microb. Ecol. Health Dis.* 14, 165-173. <https://doi.org/10.1080/089106002320644357>