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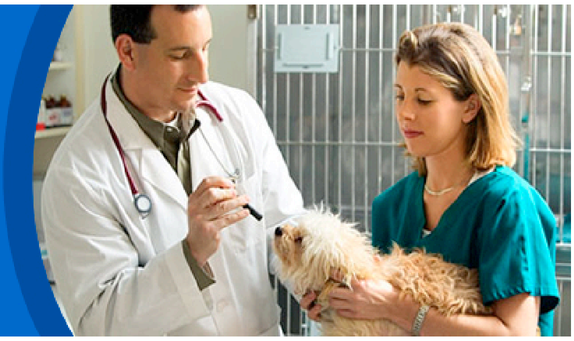

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Effect of supplementation of combination of phenolic aromatic polymer and modified lignin on gut health in broilers

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Abstract
An experiment was conducted to study the effect of phenolic aromatic polymer and modified lignin on gut health in broilers. Six treatment groups with three replicates of ten birds each for 42 days. T_1 was fed with control diet based on BIS (2007) [3], T_2 fed with reformulated diet with 2% reduction in metabolizable energy and crude protein of control diet, T_3 and T_4 supplemented with 0.2% phenolic aromatic polymer and modified lignin in control and reformulated diets, respectively and T_5 and T6 supplemented with 0.3% phenolic aromatic polymer and modified lignin in control and reformulated diets, respectively. The results showed, birds supplemented with 0.2 and 0.3% phenolic aromatic polymer and modified lignin in control diet showed significant improvement in gut health compared to control and reformulated diets fed groups. It was concluded that supplementing phenolic aromatic polymer and modified lignin improved gut health in broilers.

Keywords: Phenolic aromatic polymer, Modified lignin, gut morphology, gut microbial load, broilers

Introduction

The poultry industry is a rapidly advancing component of global animal agriculture, contributing to food security, protein supply, and rural income generation. In India, it has become a major driver of economic growth and nutrition, with poultry numbers rising by 17% between 2012 and 2019 (20th Livestock Census, 2019) [8]. Broiler production relies on fast-growing strains that are prone to stress and disease, traditionally managed through antibiotic growth promoters (AGPs). However, the emergence of antimicrobial resistance and concerns over residues have prompted restrictions on AGP use and increased interest in natural alternatives.

Polyphenols, widely distributed in fruits, vegetables, cereals, tea, and wine, represent a promising class of bioactive compounds for poultry nutrition. They are broadly classified into flavonoids and non-flavonoids with diverse biological activities including antioxidant, anti-inflammatory, immunomodulatory and antimicrobial effects (Scalbert *et al*., 2005) [10]. Although their bioavailability is limited due to extensive metabolism, polyphenols act by scavenging free radicals, modulating enzyme systems and influencing gut microbiota (Manach *et al*., 2004) [7]. They also regulate immune responses and disrupt microbial growth through mechanisms such as membrane destabilization and enzyme inhibition (Christaki *et al*., 2020) [5]. These multifunctional properties highlight their potential as natural feed additives to support health, performance and sustainability in poultry production aligning with the global shift toward antibiotic-free systems.

Previous studies have reported variable effects of dietary polyphenols and lignin derivatives on broiler performance. Dietary supplementation of phenolic aromatic polymer and modified lignin positively influenced gut morphology and microbial populations in broilers. Baurhoo *et al*. (2009) [2] reported that 1.25% alcell lignin increased jejunal villus height, goblet cell numbers, and cecal *Lactobacillus* counts, likely by suppressing pathogenic *Clostridium*. Viveros *et al*. (2011) [13] observed that grape pomace improved villus height-to-crypt depth ratio and *Enterococcus* population, while grape seed extract reduced villus height and crypt depth.

Makivic *et al*. (2018) [6] reported that lignocellulose (0.4–0.6%) increased villi length, crypt depth, and beneficial bacteria while reducing *Escherichia coli* count. Sozcu (2019) [12] found that processed lignocellulose enhanced villi length, reduced crypt depth, and supported beneficial gut microbiota, indicating improved gut maturation.

Materials and Methods

A total of 180 day-old commercial broiler chicks were procured from Venkateshwara Hatcheries Pvt. Ltd. (India) for the present investigation, and modified lignin was obtained from Phytaxis SA, Rueschlikon, Switzerland. Upon arrival, chicks were weighed and randomly distributed into six experimental groups, each comprising three replicates of 10 chicks.

The control group (T_1) was fed with basal diet formulated according to the Bureau of Indian Standards (BIS, 2007) [3] guidelines, while group T_2 received a reformulated diet with 2% reduction in metabolizable energy (ME) and crude protein (CP). Experimental groups T_3 and T_4 were supplemented with 0.2% phenolic aromatic polymer and modified lignin in both basal and reformulated diets, respectively. Similarly, groups T_5 and T6 were supplemented with 0.3% phenolic aromatic polymer and modified lignin in both basal and reformulated diets, respectively.

Birds were reared under standard management conditions in a deep litter system up to six weeks of age. Feed and water were provided ad libitum, and all chicks were vaccinated according to the recommended schedule. The study protocol was approved by the Institutional Animal Ethics Committee, KVAFSU, Bidar, Karnataka.

At the end of the experiment, two birds per replicate from T_1 to T6 were slaughtered, and small intestinal contents were collected aseptically for microbial analysis. *Escherichia coli* was enumerated on MacConkey agar, and *Lactobacillus spp*. on brain heart infusion agar using the spread and pour plate methods, respectively (Postgate, 1969) [9]. Ten-fold serial dilutions were performed, and results were expressed as log CFU/g of intestinal content (Weir, 1990) [14].

Tissue samples from the duodenum, jejunum, and ileocaecocolic junction were collected on day 42, rinsed in buffered saline, and fixed in 10% neutral buffered formalin for histomorphological analysis of villus height and crypt depth. Data were analyzed using CRD with one-way ANOVA following Snedecor and Cochran (1989) [11] in SPSS 30, and mean differences were tested by Tukey's Range Test at ($p<$ 0.05).

Results

Gut morphology

The results of supplementation of phenolic aromatic polymer and modified lignin on intestinal villi height and crypt depth in broilers are presented in Table 1. Statistical analysis revealed a significant difference ($p\leq$ 0.05) among the treatment groups at the end of experiment. The treatment groups with T_3 and T_5 showed significantly ($p\leq$ 0.05) higher villi height and crypt depth of duodenum, jejunum and ileocecal junction than the groups T_1, T_2, T_4, and T6. No differences were observed among T_1, T_2, T_4, and T6 and between T_3 and T_5.

Table 1: Effect of supplementing phenolic aromatic polymer and modified lignin on intestinal villi height and crypt depth (µm) (Mean±SE) in broilers.

Experimental group	Duodenal villi height	Duodenal crypt depth	Jejunal villi height	Jejunal crypt depth	Ileocaecocolic junction villi height	Ileocaecocolic junction crypt depth
T_1	1169.50±15.37^a	171.83±14.19^a	909.50±15.37^a	164.83±14.19^a	662.50±15.37^a	157.83±14.19^a
T_2	1175.00±10.30^a	162.17±16.53^a	915.00±10.30^a	155.17±16.53^a	668.00±10.30^a	148.17±16.53^a
T_3	1434.50±12.30^b	224.17±2.02^b	1124.50±23.83^b	222.83±7.95^b	920.83±12.58^b	216.00±7.22^b
T_4	1188.00 ±17.86^a	173.17±9.00^a	928.00±22.93^a	171.33±9.37^a	671.00±15.50^a	161.67±8.66^a
T_5	1449.00±15.09^b	225.33±6.09^b	1155.67±9.94^b	222.00±4.07^b	942.00±15.09^b	213.00±2.93^b
T6	1186.83±8.10^a	176.17±6.36^a	926.83±8.10^a	171.17±11.85^a	679.83±8.1^a	156.83±11.46^a

$^{a, b}$ Means in the same column with no common superscript differ significantly ($p\leq$ 0.05)

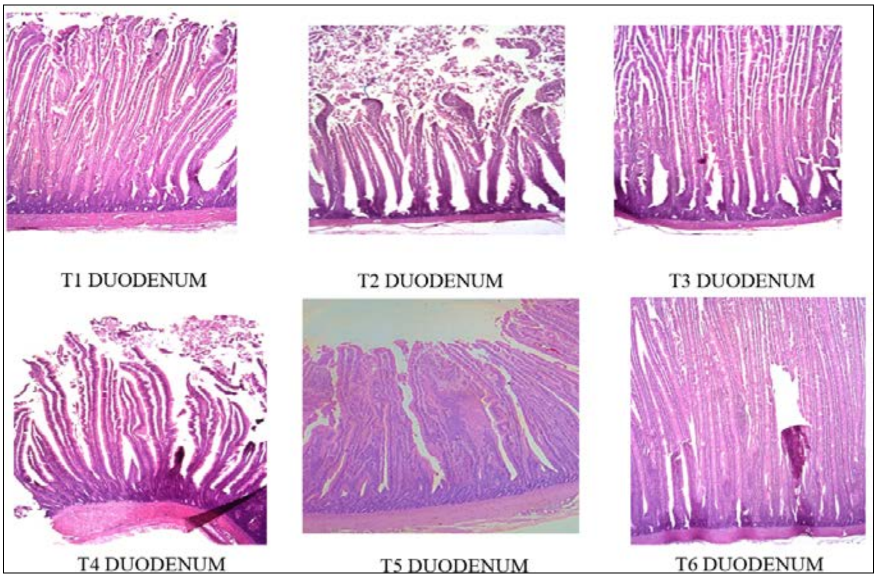

Plate 1: Section of duodenum from 42nd day old broilers fed with diets supplemented with phenolic aromatic polymer and modified lignin on duodenum villi height and crypt depth

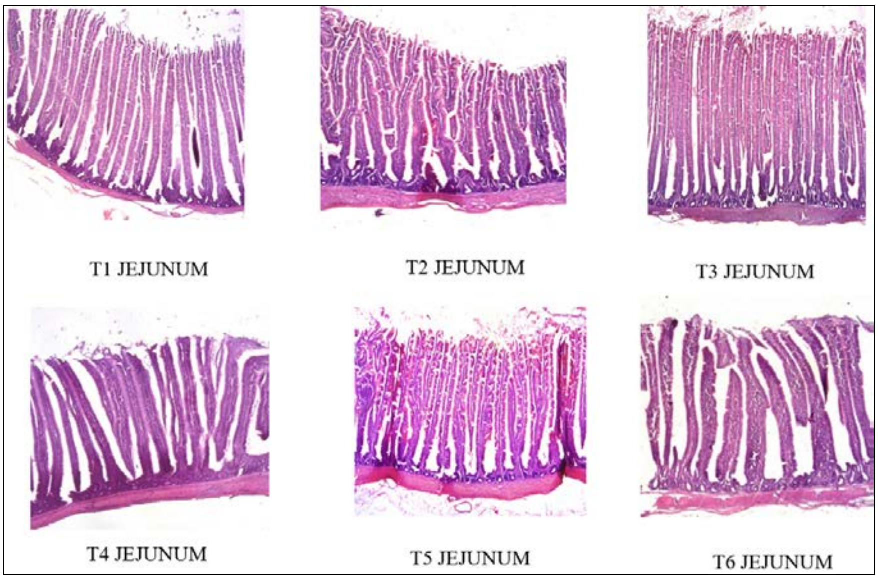

Plate 2: Section of jejunum from 42nd day old broilers fed with diets supplemented with phenolic aromatic polymer and modified lignin on jejunum villi height and crypt depth

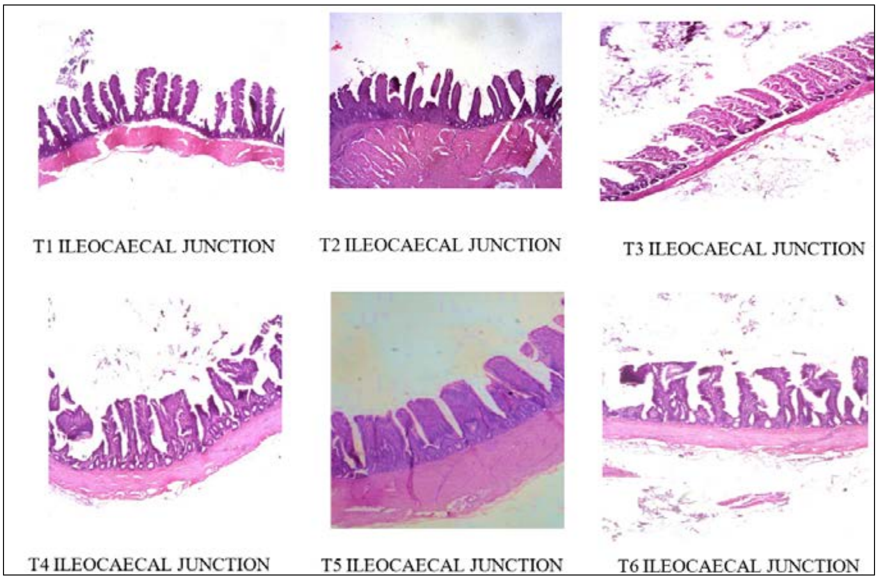

Plate 3: Section of Illeocaecocolic junction from 42nd day old broilers fed with diets supplemented with phenolic aromatic polymer and modified lignin on Illeocaecocal junction villi height and crypt depth

Gut microbial count

The results of supplementation of phenolic aromatic polymer and modified lignin on gut microbial load in broilers are presented in Table 2. Statistical analysis revealed significant differences ($P \leq 0.05$) were observed among treatments. T_3 and T_5 showed lower *E. coli* count and higher *Lactobacillus* counts compared to T_1, T_2, T_4, and T6, while T_1, T_4, and T6 had lower *E. coli* count and higher *Lactobacillus* counts than T_2. No significant differences were noted between T_3 and T_5 and among T_1, T_4, and T6.

Table 2: Effect of supplementing phenolic aromatic polymer and modified lignin on gut microbial load (log_{10} CFU / g) (Mean±SE) in broilers.

Experimental group	E.coli (log_{10} CFU / g)	Lactobacillus (log_{10} CFU / g)
T_1	6.741 ± 0.046^b	6.723 ± 0.049^b
T_2	7.183 ± 0.022^c	6.454 ± 0.076^a
T_3	6.390 ± 0.086^a	7.132 ± 0.054^c
T_4	6.800 ± 0.105^b	6.750 ± 0.040^b
T_5	6.353 ± 0.035^a	7.211 ± 0.014^c
T6	6.741 ± 0.103^b	6.712 ± 0.07^b

$^{a, b}$ Means in the same column with no common superscript differ significantly ($p \leq 0.05$)

Discussion

Phenolic aromatic polymer and modified lignin significantly ($p \leq 0.05$) improved intestinal morphology and gut microbial load. The present study is in agreement with Baurhoo *et al.* (2009) [2] who reported increase in villus height and crypt depth and increase in beneficial microbes such as *Lactobacillus* and *Bifidobacterium*, while decrease in *E. coli* count due to selective inhibition of pathogens and favorable conditions for fiber fermentation. Conversely, Viveros *et al*. (2011) [13] who reported grape seed extract supplementation decreased villus height and crypt depth and no significant changes in gut microbial counts with grape seed extract supplementation indicating potential mild adverse effects of certain phenolic compounds.

Conclusion

Based on the results, it was concluded that the addition of 0.2 and 0.3% modified lignin to both the basal and reformulated diets improved gut health. However the beneficial effect of addition of 0.2 and 0.3% phenolic aromatic polymer and modified lignin in reformulated diet was similar to like that of control diet. It was concluded that the addition of 0.2% phenolic aromatic polymer and modified lignin in reformulated diet proved beneficial in improving gut health in broilers.

Conflict of Interest

Not available

Financial Support

Not available

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