J. of Animal and Poultry Production, Mansoura Univ., Vol. 14 (9): 91 - 97, 2023

Journal of Animal and Poultry Production

Journal homepage & Available online at: www.jappmu.journals.ekb.eg

Effect of Probiotic Supplementation on Nutrients Digestibility and Intestinal Histomorphology of Growing Rabbits

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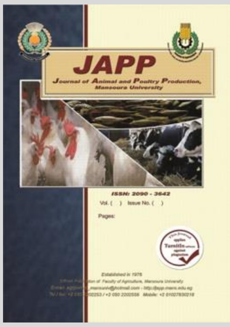

ABSTRACT

This study aimed to investigate how adding probiotics to the diet of growing rabbits affects nutrient digestion and intestinal histomorphology. The experiment involved fifteen New Zealand White rabbits, which were 12 weeks old and weighed an average of 2185 ± 7.99 grams. The rabbits were randomly assigned to five groups and were fed basal diet as a control diet and four supplemented groups with varying levels of probiotics (0.05, 0.1, 0.15, and 0.2% of dry matter). Nutrient digestibility coefficients were determined by analyzing feed and fecal samples, and the study examined the rabbits' intestinal morphology. The results showed that the digestibility of dry matter and organic matter increased with higher probiotic levels, while the digestibility of crude fiber decreased. Digestibility of crude protein (DCP) and ether extract (DEE) varied with probiotic levels. As probiotic levels increased, digestible energy per kilogram of feed decreased, and daily intake of digestible energy (DEI) showed a downward trend. The ratio of DEI to DCPI varied among the probiotic levels, with the highest value observed at the lowest probiotic level. The probiotic supplementation influenced intestinal histomorphology, with potential improvements in villus height, crypt depth, and mucosal thickness. These alterations in the intestinal region positively affected the nutrient absorption. Overall, the study suggests that dietary probiotic supplementation at levels of 0.05% and 0.1% in growing rabbits can enhance nutrient digestibility and influence the structural characteristics of the intestinal mucosa, potentially improving gastrointestinal health.

Keywords: Probiotic, nutrients digestibility, intestinal histomorphology, New Zealand White rabbits

INTRODUCTION

Probiotics are beneficial bacteria that, when ingested, can confer health benefits to the host by promoting a balanced gut microbiota. Feeding probiotics to animals, including rabbits, has been found to have a positive impact on the morphology, structure, and functions of the intestinal mucosa, particularly the villi and crypts. These beneficial bacteria can enhance villus height and density, promote the development and intestinal epithelial cells differentiation, improve the production of mucus and tight junction proteins. The villi and crypts are specialized structures within the mucosa that play crucial roles in nutrient absorption and maintaining intestinal health.

Numerous studies have shown that probiotics can modulate the composition and activity of the gut microbiota, leading to improvements in gastrointestinal health. When administered to rabbits, probiotics can colonize the gut and interact with the existing microbial community, promoting a more diverse and balanced microbiota. One of the key benefits of probiotics is their ability to enhance the structure of the intestinal mucosa. Research has demonstrated that probiotic supplementation can increase the height and density of villi in rabbits. Villi are finger-like projections that extend into the intestinal lumen and are covered by enterocytes and mucus-secreting cells known as goblet cells, whereas crypts are invaginations of the epithelium surrounding the villi (Marshman et al. 2002). By enhancing villus height and density, probiotics can improve the available surface area for nutrient absorption (Chand et al., 2016), thereby enhancing the rabbit's ability to extract nutrients from their diet.

Changes in the morphology of the intestine can serve as indicators of gut health. Rapid modifications to the mucosa layer can occur due to stress factors, as the proximity of the digesta content and the mucosal surface facilitates such alterations (Csernus and Czeglédi 2020). In this regard, as noted by Yason et al. (1987), increased crypt depth and reduced villus length may clear the toxins' presence. The health of the small intestinal surface is closely linked to the absorption of efficient nutrient. Consequently, alterations in the intestinal region, characterized by decreased crypt depth, thicker mucosa, and increased villus length can have a positive impact on nutrient absorption (Shang et al., 2015).

Samal and Behura (2015) stated that the prebiotics administration to animals has also been observed to have beneficial effects on the morphology, structure, and functions of the intestinal mucosa, specifically the villi and crypts. These improvements contribute to an increase in nutrient absorption into the bloodstream and overall nutrient utilization from the feed.

Probiotics play a role in regulating the functions of the intestinal mucosa. However, the intestinal mucosa is the innermost layer of the intestinal wall, responsible for absorbing nutrients and providing a barrier against harmful substances. Probiotics can enhance the production of mucus, which forms a protective layer over the intestinal epithelium. This mucus layer helps prevent the attachment and invasion of harmful bacteria, thereby reducing the risk of infections (Žikić et al. 2008). Probiotics can also strengthen the integrity of the intestinal barrier by promoting the production of tight junction proteins, which maintain the tight seal between epithelial cells, preventing the leakage of harmful substances into the bloodstream.

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DOI: 10.21608/jappmu.2023.231959.1089

Therefore, this study aimed to estimate the effects of dietary symbiotic supplementation containing the probiotic of S. cerevisiae and prebiotics of β-glucan and MOS on the nutrient's digestibility and intestinal histomorphology of the New Zealand White rabbits.

MATERIALS AND METHODS

The study was running at the experimental station located in El-Serw, Agriculture Research Center, Ministry of Agriculture, Egypt. The current research was aimed to study the influence of diets without or with different levels of probiotic on the nutrient digestibility and intestinal histology of growing rabbits.

Experimental animals and management

A total of fifteen healthy crossbred New Zealand White growing rabbits, aged 12 weeks, weighing 2185 ± 7.99 g, were selected for the experiment. They were individually housed in steel cages with dimensions of 50 cm in length, 50 cm in width, and 45 cm in height. These cages were equipped with automatic nip drinkers for providing fresh, clean water and manual feeders. The rabbits had ad libitum access to water and feed. Rabbits were randomly assigned to five similar groups consisting of three rabbits and was housed in a separate cage, resulting in three replications for each group.

The composition of the basal diet followed the suggestions of De Blas and Mateos (2020). The first pelleted diet (R1) served as the control diet and consisted of 31.4% alfalfa hay, 24.2% barley grain, 27.0% wheat bran, 12.25% soybean meal (SBM), 2.0% molasses, 0.95% limestone, 1.60% dicalcium phosphate, 0.30% sodium chloride, and 0.30% minerals-vitamins mixture. The probiotic additives in the pelleted experimental diets were (0.05, 0.1, 0.15 and 0.2% for R2, R3, R4 and R5, respectively).

Nutrient digestibility

Samples of feed and feces were collected to determine the apparent nutrient digestibility coefficients. The rabbits' feces were collected daily before their morning feeding for a week. These feces were stored and kept at -20 °C. Subsequently, they were dried, milled, and prepared for further chemical analyses. Chemical analysis of the feed and fecal samples included measurements of dry matter (DM), crude fiber (CF), crude protein (CP), ethereal extract (EE), and ash, following the AOAC (2019) guidelines.

The nitrogen-free extract (NFE) was calculated using the formula: "NFE=(organic matter-(EE+CP+CF))". Digestion coefficients for feeding values and all nutrients were calculated for each experimental group, with digestible crude protein and total digestible nutrients calculated according to the NRC (2001) guidelines, while digestible energy (DE) was determined using the equation:

DE (kcal/ kg DM)= (5.28× DCP) + (4.20 × DCF)+ (9.51× DEE)+ (4.20×DNFE), as described by Schiemann et al. (1972).

Intestinal morphology

In terms of intestinal morphology, at 13 weeks of age, the rabbits were euthanized, and their small intestines were carefully removed and dissected into 2 cm-long segments. The segments were washed in saline and fixed in a 10% formalin solution for 24 hours. Prepared samples were stained with hematoxylin and eosin following Bancroft et al. (2012) protocols. By using a light microscope (Olympus, Tokyo, Japan), five sections per rabbit were examined.

Statistical analysis

General Linear Model Program of SAS (2000) was used to analyze the data. The Duncan's Test (Duncan, 1955) was used to determine means differences among treatments. By using the following mathematical model, the studied traits were subjected to factorial analysis of variance for the different rabbit groups:

$$Y_{ij} = \mu + T_i + e_{ij},$$

Where Yij represents the observation of the tested factor, μ is the overall mean, Ti represents the effect of probiotic supplementation level and eij represents the experimental random error.

RESULTS AND DISCUSSION

The impact of varying probiotic levels on the digestibility of nutrients and feeding values in experimental diets are presented in Table (1). The probiotic levels, denoted as R1, R2, R3, R4, and R5, ranged from 0.00% to 0.20%. There was a slight decrease in dry matter (DM) intake as the probiotic levels increased. In terms of nutrient digestibility, there were no differences in the dry matter digestibility (DMD) across the different probiotic levels. However, organic matter (OM) digestibility slightly increased with higher probiotic levels.

Table 1. Effect of feeding experimental diets with or without probiotic levels on nutrient digestibility and feeding values.

Items	Probiotic levels, %					SEM
	R1	R2	R3	R4	R5	
	0.00	0.05	0.10	0.15	0.20	
DM intake (g/h/d)	182.74	178.91	177.75	172.52	176.45	12.138
Nutrient digestibility (%):						
DM	61.79^{ab}	64.30^{a}	61.68^{ab}	57.67^{bc}	54.74^{c}	1.254
OM	64.50^{ab}	67.52^{a}	64.65^{ab}	60.49b^{c}	57.21^{c}	0.998
CP	58.11	60.00	67.71	64.89	65.70	2.256
CF	39.88	36.57	31.04	32.80	30.62	6.079
EE	47.06^{ab}	54.41^{a}	44.47^{ab}	33.08^{b}	47.09^{ab}	3.795
NFE	81.32^{ab}	89.14^{a}	79.86^{ab}	73.68^{bc}	66.72^{c}	2.217
Digested nutrients and feeding value as DM (%):						
DCP	10.68^{b}	11.02^{ab}	12.73^{a}	12.07^{ab}	12.02^{ab}	0.415
DCF	6.09	5.58	4.63	5.14	4.65	0.929
DEE	1.703^{b}	3.220^{a}	1.697^{b}	0.907^{c}	1.440^{bc}	0.143
DNFE	46.12^{ab}	49.85^{a}	45.44^{ab}	41.93^{bc}	38.33^{c}	1.255
TDN	64.58^{b}	69.68^{a}	64.50^{b}	60.05^{c}	56.44^{d}	0.593
DCPI (g/d)	19.55	19.79	22.61	20.84	21.40	1.934
TDNI (g/d)	118.02	124.59	114.63	103.61	99.64	7.768
DE kcal/kg	2265.3^{b}	2417.1^{a}	2186.3^{b}	2049.4^{c}	1882.3^{d}	24.921
DEI kcal/d	413.8	432.0	388.5	353.6	331.6	25.261
DEI/DCPI	21.26^{a}	21.97^{a}	17.20^{b}	16.99^{b}	15.80^{b}	0.752

[a, b, c, d] **Within a row, values with different superscript letters mean there were significant differences (*p*<0.05).**

The digestibility of crude protein did not follow a consistent pattern among the probiotic levels. On the other hand, the crude fiber digestibility decreased as the probiotic levels increased. The digestibility of ether extract (EE) varied, with the highest value observed at the intermediate probiotic level. Additionally, the digestibility of nitrogen-free extract (NFE) decreased as the probiotic levels increased.

Regarding the digested nutrients and feeding value as a percentage of DM (Table 2), the digestible crude protein (DCP) increased with higher probiotic levels, while the digestible crude fiber (DCF) did not show significant differences across the probiotic levels. The digestible ether extract (DEE) decreased as the probiotic levels increased, and the digestible nitrogen-free extract (DNFE) decreased with higher probiotic levels. Furthermore, the total digestible nutrients (TDN) decreased as the probiotic levels increased.

The digestible crude protein intake (DCPI) did not exhibit a consistent pattern among the probiotic levels (Table 1). The total digestible nutrient intake (TDNI) decreased as the probiotic levels increased (table 1. The digestible energy content (DE) per kilogram of feed decreased as the probiotic levels increased, and the digestible energy intake (DEI) per day showed a decreasing trend with higher probiotic levels. The ratio of DEI to DCPI varied among the probiotic levels, with the highest value observed at the lowest probiotic level (Figure 1).

The digestion rates of ether extract (DEE%) and nitrogen-free extract (DNFE%) were significantly higher ($p < 0.05$) with the addition of probiotics at a concentration of 0.05% in the diet, as shown in Table (2). The digested energy DE (kcal/kg) was also significantly higher ($p < 0.05$) in the diet supplemented with 0.05% probiotics (2417.1 kcal/kg) compared to the other diets, as indicated in Table (2). Furthermore, the DEI/DCPI values were higher ($p < 0.05$) in the control diet rabbits or that fed diet supplemented with 0.05% probiotics (21.26 and 21.97, respectively) compared to the other diets, as presented in Table (1) and Figure (1).

Hillyer et al. (1997) emphasized the importance of pelleted rabbit diets, which consist of a combination of carefully selected ingredients to meet the specific nutrient requirements of rabbits. The typical components of the basal diet include grain by-products and grain, alfalfa hay, protein supplements, minerals, and vitamins. Gidenne (1992) and De Blas et al. (1995) demonstrated that these nutritional components were suitable for formulating diets for growing rabbits. They reported that the dietary levels of crude protein (CP) ranged from 16.3% to 19.8%, crude fiber (CF) from 14% to 18%, and ether extract (EE) at 5.5%. However, the dietary fiber level has an impact on the other nutrient digestibility in the rabbits' diet (Gidenne and Garcia, 2006).

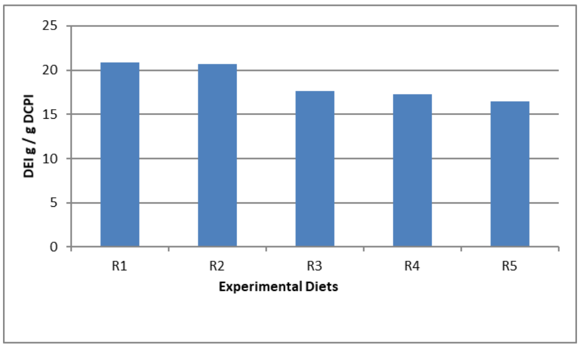


Fig. 1. Effect of feeding experimental diets on growing rabbits

Different probiotic levels yielded different outcomes, indicating the importance of selecting an appropriate probiotic level for optimizing nutrient utilization and intestinal health in the studied context. Hence, the appropriate amount of crude protein in a diet relies on its digestibility and digestible energy content. It has been recommended to maintain a ratio of 23.5 Kcal DE per gram of digestible protein or 10 grams of DP (digestible protein) per Mj (megajoule) of DE, as this ratio has been shown to optimize growth rate and reduce mortality. The inclusion of organic acids in the feed reduces its buffering capacity and speeds up the process by which stomach contents reach the ideal pH of approximately 3. This, in turn, improves the efficiency of the proteolytic enzyme pepsin. These findings have been supported by studies conducted by Blanchard and Wright (2004) and Davidson et al. (2001).

The results of the representative photomicrograph of the intestine for different treatment groups are displayed in Table (2). Feeding rabbits with diets R1, R2, or R3 resulted in significantly longer villus compared to diets R4 or R5 ($p < 0.05$). Similarly, feeding on diet R1 led to an increase in villus width compared to diets R2, R3, R4, or R5 ($p < 0.05$). The submucosa thickness value in rabbits fed diet R5 was higher ($p < 0.05$) compared to the other diets, while the muscular thickness values in rabbits fed diets R2, R3, or R4 were decreased ($p < 0.05$) compared to diets R1 or R5.

Table 2. Effect of feeding experimental rations without or with probiotic levels on Intestinal Histomorphology:

Items	Probiotic levels, %					SEM
	R1	R2	R3	R4	R5	
	0.00	0.05	0.10	0.15	0.20	
Villus height	323.93^{ab}	397.09^{a}	397.08^{a}	264.3^{b}	267.15^{b}	28.4476
Villus width	165.95^{a}	57.53^{b}	59.28^{b}	65.60^{b}	91.09^{b}	9.0911
Sub mucosa	31.65^{bc}	38.30^{bc}	43.33^{ab}	23.96^{c}	57.72^{a}	3.9480
Muscular	46.30^{a}	28.46^{b}	25.14^{b}	22.99^{b}	58.44^{a}	2.9897

$^{a, b, c}$ **Within a row, values with different superscript letters mean there were significant differences (*p*<0.05).**

Assessing the structure of the intestines, particularly factors such as villus height and the ratio of villous height to crypt depth, is crucial in evaluating digestive well-being (Shamoto and Yamauchi, 2000). Researchers Parker et al. (2017) observed that longer villi are linked to active cell growth, which can promote enzyme secretion, improve digestion, and enhance the absorption of nutrients by expanding the surface area available for absorption (Awad et al., 2009).

The current results showed that the height of the villi in the intestines varied among the probiotic levels, with the highest values observed at R2 and R3, and the lowest values at R4 and R5. The width of the villi showed a significant decrease at R2 compared to the other probiotic levels. The thickness of the submucosa varied among the probiotic levels, with the highest value observed at R5. The thickness of the muscular layer showed variations among the probiotic levels, with the highest values observed at R1 and R5.

The findings of this study are consistent with previous research on the effects of probiotics on intestinal morphology in rabbits and weaning pigs. The increased height of duodenal villi in rabbits fed a diet supplemented with probiotics supports the idea that this supplementation can enhance nutrient absorption and improve feed conversion ratio. Similar positive effects on villi height have been observed with the use of mannan-oligosaccharides (MOS) as an alternative to antibiotics. However, growing rabbits that were fed a diet supplemented with 0.0%, 0.05%, and 0.10% probiotics showed a significant increase in duodenal villous heights. These findings are consistent with previous research that examined the effects of probiotic supplementation, as by Liu et al. (2019) and Bassiony et al. (2021) on intestinal morphology in growing rabbits and by Galeano et al. (2016) and Wang et al. (2019) on weaning pigs. In relation to this matter, Mourao et al. (2006) showed an elevation in the villi height when mannan-oligosaccharides were added as antibiotics alternative. This positive effect on villi can enhance nutrient absorption in the intestine, potentially leading to an improved feed conversion ratio. As partial support for these findings, Mourao et al. (2006) demonstrated an increase in villi height from 1.0 to 1.5 g/kg of MOS, without an increase in fecal recovery coefficient.

Other studies have also reported that MOS supplementation increases the length of villi in the ileum, potentially due to improvements in the intestinal environment. This enhanced development of intestinal villi can contribute to higher feed utilization and improved growth performance in animals. Multiple studies stated that the length of villi in the ileum increased by MOS supplementation, could be due to improvements in the intestinal environment (Pinheiro et al., 2004; Mourao et al., 2006). The enhanced development of intestinal villi can provide an explanation for the higher feed utilization observed in rabbits supplemented with MOS 1.0% (Bovera et al., 2010).

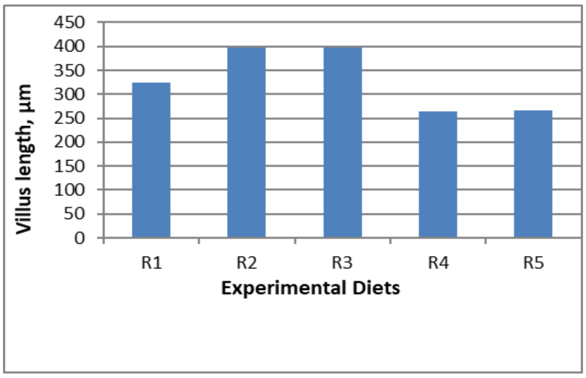


Fig 2. Effect of feeding experimental diets villus length

It is worth noting that prebiotics, which are non-digestible dietary components that promote the growth of beneficial bacteria in the gut, have also been observed to have beneficial effects on the structure, morphology, and functions of the intestinal mucosa. The combination of probiotics and prebiotics, known as symbiotics, may provide even greater benefits in terms of nutrient digestibility and intestinal health. Hassanin et al. (2015) investigated the impact of synbiotic dietary supplementation on duodenal morphology, specifically villus width, height, and duodenal gland length. They found that dietary synbiotic supplementation influenced these histomorphological measurements of gland length and duodenal villi. Feed additives significantly increased the lengths of villi and glands in all treated groups when compared to the control.

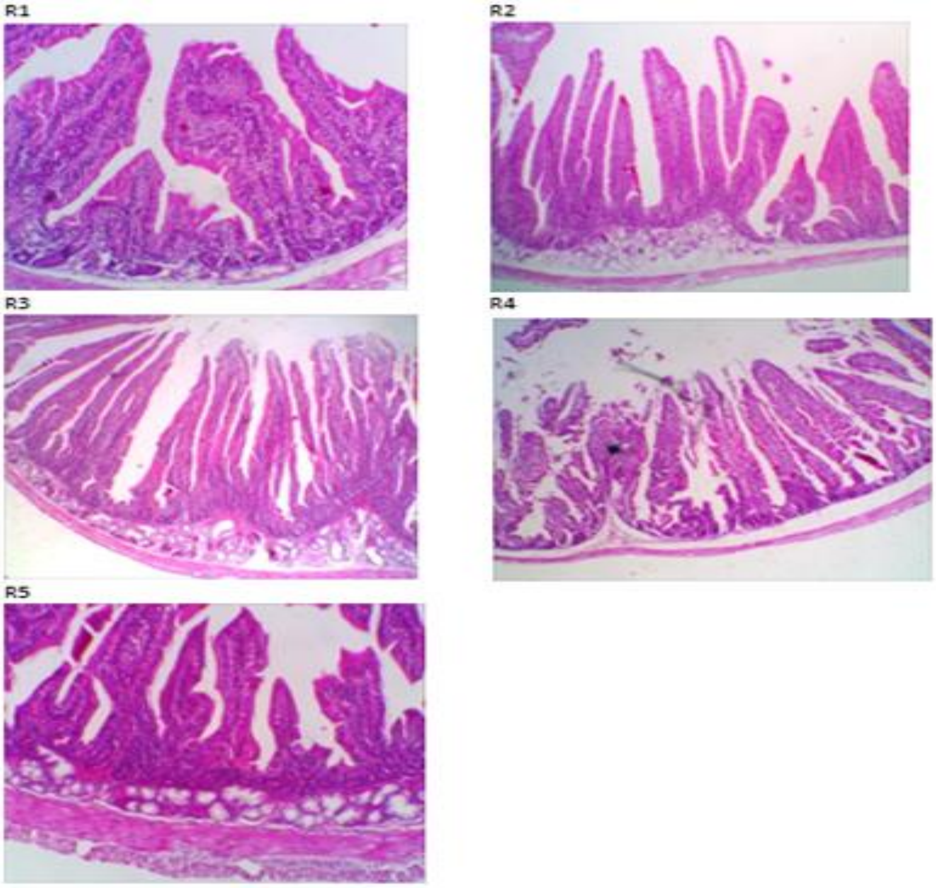
Furthermore, the improved growth and feed efficiency observed in rabbits given probiotics can be attributed to the enhanced intestinal morphology, including increased villus height, and a more balanced intestinal microbial environment, which promotes better digestion and absorption of nutrients. Bassiony et al. (2021) reported that rabbits fed probiotics exhibited enhanced growth performance, which was linked to improvements in the structure of the intestinal mucosa, leading to increased nutrient absorption (Samli et al., 2007). The results presented for the DEI/DCP ratio indicate that feeding rabbits with diets R1, R2, or R3 resulted in higher values compared to diets R4 and R5.

Reduced villi length can have detrimental effects on nutrient absorption, leading to increased secretion within the gastrointestinal tract and subsequently resulting in decreased performance (Xu et al., 2003). On the other hand, higher villus height and a greater ratio of villus height to crypt depth are directly linked to an improved turnover of epithelial cells (Fan et al., 1997). Additionally, increased villus height indicates the activation of villus function (Langhout et al., 1999; Shamoto and Yamauchi, 2000). Consequently, it can be inferred that the function of intestinal villi is stimulated when dietary probiotics and synbiotics are administered. This implies that the presence of probiotics and synbiotics in the diet may promote the growth and development of intestinal villi, enhancing nutrient absorption and overall digestive health.

Furthermore, probiotics have been observed to positively influence the morphology of the intestinal mucosa. They can promote the development and differentiation of intestinal epithelial cells, which line the mucosa and form the villi and crypts. This enhanced cell proliferation and differentiation contribute to the maintenance of a healthy intestinal epithelium, ensuring efficient nutrient absorption and providing a protective barrier against pathogens.

Multiple mechanisms have been suggested to explain this intestinal architecture phenomenon (Lao et al., 2020). Mechanisms such as the production of short-chain fatty acids (SCFAs) through fermentation and enhanced activity of digestive enzymes in the gut contribute to the improved growth and turnover rate of the intestinal mucosa observed with prebiotic supplementation. However, Samal and Behura (2015) stated that one of the mechanisms involves short-chain fatty acids (SCFAs) produced through fermentation, which promote cell proliferation in the small intestines and large intestines, leading to increased villi height and crypt depth, thereby facilitating nutrient absorption. Additionally, Yang et al. (2005) reported that the enhanced activity of digestive enzymes in the gut could be a contributing factor to the improved growth and turnover rate of the intestinal mucosa in animals receiving prebiotics.

In a feeding experiment conducted by Shim (2005) using pigs supplemented with fructo-oligosaccharides at a concentration of 0.25%, a significant elongation of villi by up to 24% was observed, along with a notable increase in the ratio of villi-height to crypt-depth of the small intestines. Similarly, in studies involving broilers supplemented with inulin or fructo-oligosaccharides, an improvement in nutrient absorption capacity was obtained, accompanied by increased villi height, crypt depth and the number of mucus-secreting cells (Yusrizal and Chen 2003; Žikić et al., 2008). These findings further support the positive impact of prebiotic supplementation on the intestinal mucosa of animals.



Representative photomicrograph of intestine from different treatment groups.

R1: control group showing normally arranged intestinal villi with few edemas separated mucosal layers from sub mucosa. The muscular is layers with few aggregations of inflammatory cells in between the crypt, and with short villus length.

R2: showing long intestine villus with normally arranged intestinal villi and normal goblet cell.

R3: showing normally arranged intestinal villi with normal goblet cell and long villus length.

R4: showing normally arranged intestinal villi with few aggregations of inflammatory cells in between the crypt.

R5: showing intestine group with multifocal, mild inflammatory cells, and observed increase numbers villus width.

Overall, the results obtained demonstrate that probiotic supplementation can affect intestinal histomorphology, with variations observed in villus height, width, submucosa thickness, and muscular layer thickness among different probiotic levels. These changes in intestinal morphology have implications for nutrient absorption, feed utilization, and overall animal performance. The combination of probiotics and prebiotics (synbiotics) may supply even greater benefits in the form of intestinal health, as a result improved nutrient digestibility.

CONCLUSION

In summary, the findings indicate that incorporating probiotics at levels of 0.05% and 0.1% in the experimental diets influenced nutrient digestibility and feeding values. Furthermore, supplementation with probiotics at these levels was observed to have positive effects on the intestinal mucosa' morphology, structure, and functions, particularly the crypts and villi. By promoting the health of the intestinal mucosa, probiotics contribute to improved nutrient absorption, a balanced gut microbiota, and overall gastrointestinal well-being in rabbits.

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تأثير إضافة البروبيوتيك على معاملات الهضم والتركيب الهستولوجى للامعاء الدقيقة فى الأرانب النامية

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الملخص

أجريت هذه التجربة على خمسة عشرة من الارانب النيوزيلاندي البيضاء في عمر 13 أسبوعًا وبمتوسط وزان 2185 ± 7,99 جم، وتم توزيع الأرانب إلى خمس مجموعات بالتساوي. كانت العليقة التجريبية الاولى عليقة كنترول وتم إضافة البروبيوتيك بمستويات (0.05 و0.1 و0.15 و0.2 %) الى العليقة التجريبية لتكوين العلائق التجريبية للمجموعات الأربعة على التوالي. وكانت أهم النتائج المتحصل عليها هي زيادة معامل هضم مستخلص الاثير والمستخلص الخالى من الازوت معنويا عند مستوى 0.05٪ من البروبيوتيك، وكذلك زادت القيمة الغذائية معنويا للمركبات الغذائية المهضومة (69,68٪) مقارنة بالمجموعات الأخرى. زادت ايضا قيم كل من البروتين الخام المهضوم ومستخلص الاثير المهضوم والمستخلص الخالى من الازوت المهضوم معنويا على مستوى إضافة 0.05٪ من البروبيوتيك، كما زادت الطاقة المهضومة للعليقة معنويا (2417,1 kcal/ kg) مقارنة بالعلائق الأخرى. أظهرت النتائج كذلك انه حدث تحسن للاستفادة من الطاقة المهضومة الى البروتين المهضوم DEI / DCP عند التغذية على 0.05% و0.1% (21,26 و 21,97 على التوالي) مقارنة بالعلائق الأخرى. زاد طول الخملات معنويا بالتغذية على العليقة التجريبية مع اضافة البروبيوتك حتى تركيز 0.1%، وكان سمك العضلات أقل في المجموعات المضاف لها بروبيوتك حتى تركيز 0.15% ، وكان عرض الخملة أعلى في المجموعة الكنترول عند المقارنة بالمجموعات الاخرى، بينما كان سمك الغشاء المخاطي أعلى عند التغذية على اعلى مستوى بروبيوتك 0.2% مقارنة بالمجموعات الأخرى. وبناء على النتائج المتحصل عليها فإن إضافة نسبة 0.05 أو 0.1% من البروبيوتك إلى العليقة في الأرانب أدى الى تحسن فى نمو الامعاء الدقيقة مما أدى الى تحسن فى عمليات الهضم والامتصاص.