International Research Journal of Advanced Engineering and Science
ISSN (Online): 2455-9024

Supplementation Mannan-Rich Fraction (MRF) and / or Combination with Probiotic-Enhanced Water Acidifier on Dietary Male Broiler at 28 on Intestinal Micro Flora and Ratio of *Lactobacillus* / *Coliform*

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***Abstract*—** *According to newest regulation PERMENTAN/14/16/2017 Indonesia has banned the use of antibiotics on poultry. The using feed additive is one method to improve the quality of feed. The research purpose to carry out the possible effect of mannan-rich fraction (MRF) and/ or combination with probiotic enhanced water as natural growth promoters (NGPs) on intestinal micro flora and ratio of lactobacillus / coliform. A total of 160 broilers (males) 1-d-old Arbor Acres broiler were divided among 4 dietary treatments with 4 replicate cages per treatments and 10 males broiler per cage assigning experimental units to treatments randomly. The chicken will sacrificed at starter periods (28days) (6 each chicken will sacrificed) with amount total 24 broiler sacrificed each periods. The first experiment method was used completely randomized design with 4 treatments and 4 replicates. The treatments used for research were dietary with T0 (control), T1 (basal feed + MRF (Actigen™) 800g/ 400g / 200 g), T2 (Drinking water + 2 ml / L Combination feed additive (Acid-Pak 4-way®)), T3 (basal feed + MRF (Actigen™) 800g/ 400g / 200 g+ Drinking water 2 ml / L Combination feed additive (Acid-Pak 4-way®)). The parameters observed were intestinal micro flora and ratio of lactobacillus / coliform The data would be statistically analyzed using the analyses of variance using one-way anova with Completely Randomized Design (CRD) in Minitab® 16th edition. The differences between the means of groups were separated by Duncan Multiple Range Test ($P < 0.05$). To sum up the addition of mannan-riched fraction and combination with probiotic enhanced liquid acidifier has not significantly different ($P > 0.05$) intestinal micro flora and ratio of lactobacillus / coliform.*

***Keywords*—** *Broiler, mannan-rich fraction, natural growth promoters, prebiotic.*

I. INTRODUCTION

Poultry serves an essential role in supporting the availability of cheap animal protein sources that are easy to obtain in Indonesia. This condition is reflected on how the demand on the poultry market increased nationally. For example, in 2017, broilers increased by about 6.82% compared to the population in 2016 (1.6 billion heads). However, the poultry business is mostly attached to the use of antibiotics as a growth promoter. Although, the use of antibiotics growth promoters (AGPs) as a feed additive in animal feed has lasted more than 40 years in Europe. Lately, the use of antibiotic compounds has decreased and even in several European countries has suppress the use of antibiotics growth promoters (AGPs) as non-nutritive values feed additives in animal feed. The limitation of using antimicrobial began in 1997 when the European Union (EU) banned using *avoparcin* for growth promoters to the livestock Maron *et al.* [1]. The chase continued, until the last phase when the EU-wide ban on AGPs in animal feed (poultry) took effect since 2006.

However, the antibiotics used are still prevalent in the poultry farm business. Due to the reason that key of the quality meat is from the feed; the farmer used the antibiotics as a feed additive; while the combination used are acidifier, hormone, prebiotics, probiotics even the symbiotic to increasing the nutritive value. Different substances often referred to as natural growth promoters (NGPs) are supposed to achieve high consumer acceptance since they do not usually pose any risk regarding bacterial resistance.

The amount of the pathogen bacteria is decreasing due to the condition that inhibits the growth, while the absorption of the nutrient for the internal organ also increase. The activator of the prebiotics increases the number of intake for the growth of the internal organ of chicken Maron *et al*. [1]. Therefore, the objective of this experiment was carried out to determine the possible effect of mannan-rich fraction (MRF) and/ or combination with probiotic enhanced water as natural growth promoters (NGPs) on intestinal micro flora and ratio of lactobacillus / coliform

II. MATERIALS AND METHODS

A. Location of Study

This study was carried out at the poultry research station, Department of Animal Science, National Pingtung University of Science and Technology (NPUST), Pingtung, Taiwan.

B. Animals and Study Design

A total of 160 broilers (males) 1-d-old Arbor Acres broiler were divided among 4 dietary treatments with 4 replicate cages per treatments and 10 males broiler per cage assigning experimental units to treatments randomly. The chicken will sacrificed at starter periods (28days) (6 each chicken will sacrificed) with amount total 24 broiler sacrificed each periods. The treatments used for research were dietary with T0 (control), T1 (basal feed + MRF (Actigen™) 800g/ 400g / 200 g), T2 (Drinking water + 2 ml / L Combination feed additive (Acid-Pak 4-way®)), T3 (basal feed + MRF (Actigen™) 800g/ 400g / 200 g+ Drinking water 2 ml / L Combination

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International Research Journal of Advanced Engineering and Science
ISSN (Online): 2455-9024

feed additive (Acid-Pak 4-way®)). The basal diet content were showed in the table 1.

TABLE I. The basal diet content used in study

Feed Nutrient	Content
Dry matter (%)	87.00
Moisture (%)	13.00
Ash (%)	9.00
Crude Protein (%)	20.36
Fat (%)	6.00
Crude fibre (%)	3.00
Ca	1.00 ± 0.25
P	0.70±0.25
Copper (ppm)	30
Zinc (ppm)	120

C. Statistical Analysis

The data would be statistically analyzed using the analyses of variance using one-way anova with Completely Randomized Design (CRD) in Minitab® 16th edition. The differences between the means of groups were separated by Duncan Multiple Range Test ($P < 0.05$).

III. Result and Discussion

A. Intestinal Microflora

The broilers sacrificed at 28 days old. The part intestinal were cut six cm and the digesta were analyze at agar plate that already preparation. There was no difference at the beginning on the intestinal microflora, but at 28 days the amount of coliform better than control male broilers (3.35 vs. 2.70 log cfu/g, DM; $P < 0.05$) on lactobacillus. However, There was no diff erence in the intestinal microflora of broiler chickens both jejunum and ileum parts. According to Venema and Carmo [2] the probiotics-enhanced liquid acidifier used an alternative to increase the number of *Lactobacillus* and Bifidobacteria in gastrointestinal tract (jejunum and ileal). The colon is to incorporate prebiotics into animal feed to stimulate the resident microbiota to proliferate. The levels of beneficial bacteria such as *bifidobacteria* and lactobacilli resident in the host Charalampopoulus and Rastall [3].

According to Leeson and Summers [4] the term 'competitive exclusion' is often used synonymously with probiotics. It is assumed that the probiotic will have a competitive advantage over any inherent pathogen, and either replace it, or prevent its colonization. Lactic acid from Lactobacilli and other species is an example of such a product. Probiotic organisms may also stimulate mucosal immunity. While undefined mixtures of bacteria, usually derived from cecal contents of healthy broiler Leeson and Summers [4]

TABLE II. Effect of mannan-riched fraction and probiotics enhanced liquid acidifier on *lactobacillus* of male broilers

Treatment	*Lactobacillus* (28d)
T0	2.70^{b}
T1	3.51^{a}
T2	3.54^{a}
T3	3.35^{b}
SEM	0.38

TABLE III. Effect of mannan-riched fraction and probiotics enhanced liquid acidifier on *coliform* of male broilers

Treatment	*Coliform* (28d)
T0	4.66^{a}
T1	1.80^{b}
T2	2.44^{ab}
T3	1.72^{b}
SEM	1.43

TABLE IV. Effect of mannan-riched fraction and probiotics enhanced liquid acidifier on *enterococcus* of male broilers

Treatment	*Enterococcus* (28d)
T0	0.41^{ab}
T1	2.08^{a}
T2	0.06^{b}
T3	0.18^{ab}
SEM	0.05

B. Ratio Lactobacillus / Coliform

At the end of the periods (28 days old), while the level of mannan-riched fraction and probiotics-enhanced liquid acidifier better more than control (T2 and T3) on coliform in the ileum parts (3.22; 6.56 vs. 15.91 (female) and 12.93 (male) log cfu/g, DM; $P < 0.05$). The mannan-riched fraction and probiotics-enhanced liquid acidifier treatment group help to reduce the amount of harmful bacteria. The hypothesis the amount bacteria had optimum work at 28 days and continued to reduce the negative bacteria compared than control in the body of broiler. Di Giola and Biavati [5] the condition happens due to unstable microflora in the day old chicks. The pathogens bacteria, e.g., enterococci still dominated the crop, duodenum, and ileum while coliforms dominate the caecum during the seven days. Spring *et al* [6] the caecum is the site for bacterial fermentation of nondigestible carbohydrate and the main place for the development of pathogens area. Microbial communities vary considerably by location along the gastrointestinal tract, e.g., crop, gizzard, duodenum, and ileum share similar bacteria dominated by Lactobacillus. The lactobacilli will dominate the crop, duodenum, and ileum while anaerobic bacteria still dominate the caeca. Rodjan *et al* [7] at 28 days the positive microbial community starts to stabilize indicated feed consumption increase and the microbial stable on bacterial fermentation.

TABLE V. Effect of mannan-riched fraction and probiotics enhanced liquid acidifier on *lactobacillus / coliform* of male broilers

Treatment	*Lactobacillus* / *Coliform* (28d)
T0	0.57
T1	1.95
T2	1.45
T3	1.94
SEM	0.44

IV. Conclusion

The amount of micro flora were optimized at 28 days, both male and female broiler the amount of *Lactobacilli* were better compared than control eventhough did not share significant different. The amount of *Coliform* also reduced at 28 days. After that period it can't help to reduce harmful bacteria.

Acknowledgement

The authors are grateful to the corporation which has facilitated Actigen™ (Alltech Inc., Nicholasville, Kentucky:

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USA) for the learning process in Taiwan

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