

# EFFECT OF FERMENTED COW MILK AND SOYBEAN MILK WITH PROBIOTIC SUPPLEMENTATION ON BLOOD HEMATOLOGY OF BROILERS

Novi Mayasari<sup>1\*</sup>, Lovita Adriani<sup>1</sup>

<sup>1</sup>Faculty of Animal Husbandry, University Padjadjaran, Bandung, Indonesia

## Abstract

Probiotic supplementation could reduce pathogenic bacteria population, improved health and performances. The aim of this study was to determine effect of probiotic supplementation in different media (cow milk vs. soybean milk) in the diet on blood hematology of broilers. A total of 100 d-old Broiler, were obtained from a local hatchery (Cipacing hatchery, Jatiningor, Indonesia) and randomly assigned to 4 dietary treatments, each with 5 replicates, 5 chickens per replicate. The 4 diet treatments were the basal diet (no supplement/T0), basal diet + probiotic in cow's milk (T1), basal diet + probiotic in soybean milk (T2), basal diet + probiotic in cow's milk and soybean milk (ratio 1:1) (T3). Probiotic contains *Streptococcus thermophilus*, *Lactobacillus bulgaricus*, and *Lactobacillus acidophilus*. Fermented cow milk or soybean milk were given based on 1.25% of body weight. The diet treatment was given throughout the 35 d experimental period. Result showed the addition probiotic in media cow's milk and soybean milk in the diet (T3) decreased white blood cell count of broiler compared with T0, T1 or T2. There were no differences on Hb, PCV or RBC among treatments. In conclusion, the addition of probiotic in media cow's milk and soybean milk in the diet resulted in better fermentation of *S. thermophilus* due to availability of lactose (sugar milk) and active compound of isoflavones. Higher number of good probiotic bacteria better hematology profile of broiler.

**Key words:** probiotic, cow milk, soybean milk, hematology, broiler

## INTRODUCTION

Broiler are vulnerable to pathogenic microorganisms which leads to increase incidence of diseases like *Chronic Respiratory Diseases*, thus low performances [5]. Dietary strategies such as antibiotic use and various feed supplements were introduced to reduce pathogenic microorganism and to minimize the incidence of diseases. In last decades, antimicrobial compounds have been used in the poultry industry by reducing pathogenic microorganism to improve health status and performance of broiler [6]. However, recent study found that by using growth stimulating feed may threat to human health [17]. Restrictions on the use of antibiotics as a growth-stimulating feed supplements imposed in many countries. In Indonesian government had suggested the limitation use

of growth promotor and antibiotic to anticipate bad effect of antibiotic use for livestock and human.

Alternative feed supplements include enzymes; probiotic, organic acids, as well as various prebiotic and phytogetic preparations are available to poultry to replace growth promoting feed and antibiotic [7]. Probiotic is a product that contains vital microorganisms with enough number to have an ability to change a number of microorganisms inside the host which alters hygienic imported trails in the host [14]. A study has shown that the addition of probiotics to the diet of broiler might be acts as antimicrobial, anticarcinogenic, antiallergic, immune stimulating actions, improves the absorption of minerals, protects from diarrhea and optimizes nutrient digestion processes [8].

Cow milk is a common media for probiotic to grow. There are many benefit of cow milk and its' fermented for human health. Milk of the cows contains huge nutrients like protein, lipid, vitamin, mineral and important

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\*Corresponding author: novi.mayasari@unpad.ac.id  
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carbohydrate such as lactose. Bacteria can use many different sources of carbohydrates, some of the most common being; glucose, fructose, pentose and lactose. Live bacteria are often referred to as lactic acid bacteria. Most of these lactic acid bacteria use glucose as their primary energy source, an exception being *Streptococcus thermophilus* which uses lactose preferentially.

Soybean milk is also one of good media for bacteria to lives. Soybean milk and its fermented is an option healthy drink for vegan. Soybean milk not only contains carbohydrates, protein, mineral and vitamin but also contain active compound like isoflavones. Isoflavones have potential functions as antioxidants, enhanced immune function and detoxification [4]. Several studies have shown that feeding broilers with isoflavones increased weight gain, feed intake, meat quality [10], feed conversion ratio and breast muscle rate [11].

Different bacteria with gram positive or negative requires different pH. It is hypothesized that combination of cow milk and soybean milk fermented by probiotic *Streptococcus thermophilus*, *Lactobacillus bulgaricus*, and *Lactobacillus acidophilus* will provide better condition for bacteria compared with only single media.

It is little known whether supplementation of probiotic with different media (fermented cow milk or soybean milk) in the diet still give best result on health of broilers indicated by blood hematology. The effect of probiotic supplementation in different media (fermented cow's milk vs. fermented soybean milk) in the diet on some hematological parameters of 35 days old broiler chickens were studies.

## MATERIAL AND METHOD

A total of 100-d old Broiler was raised during 35 days. The treatment started from 0-d until the broiler's age 35 days. We used basal diet from commercial product. The basal diet was formulated to meet the nutrient requirements of broiler. Energy metabolism and protein in the ratio is 3025-3125 Kcal/kg and 21.5-23.8% respectively. Feed and water were provided adlibitum. The birds were reared in floor pens.

Blood samples (approximately 3 mL) from 5 birds per treatments were collected

from the jugular vein into heparinized tubes for estimation of whole blood red blood cells (RBC), packed cell volume (PCV), hemoglobin (Hb) and white blood cells (WBC). PCV% was determined by using micro-hematocrite method according to the method of (Archer, 1965). Hemoglobin (Hb) concentration was determined by Drabkin's reagent. Count of RBC and WBC was determines by using hemocytometer.

Analysis of variance (ANOVA) using PROC MIXED SAS 9.3 was used to analyze the effect of supplementation of fermented cow milk and soybean milk with probiotics on some hematological parameters of broilers such as Hb: hemoglobin; PVC: Packed volume cell; RBC: red blood cell; and WBC: white blood cell. The four dietary treatments were : (1) T0: basal diet (no supplement), T1: basal diet+ fermented cow milk with *Streptococcus thermophilus*, *Lactobacillus bulgaricus*, and *Lactobacillus acidophilus*, T2: basal diet + fermented soybean milk with *S. thermophilus*, *L. bulgaricus*, and *L. acidophilus*, T3: basal diet + 50% fermented cow milk and 50% fermented soybean milk with *S. thermophilus*, *L. bulgaricus*, and *L. acidophilus*.

## RESULTS AND DISCUSSIONS

PCV percentage, RBC count, hemoglobin and WBC were observed in the present study. The WBC were lower in birds fed diet with probiotic in fermented cow milk and soybean milk compared with birds fed the control/basal diet, birds fed diet with probiotic in cow milk or birds fed diet with probiotic in soybean milk (Table 1 and Figure 1).

The lower WBC count in the birds fed probiotics in combination media of fermented cow milk and soybean milk may be due to the carbohydrate in cow milk; lactose (milk sugar). It is known that the fermentation of the bacteria like *Streptococcus thermophilus* which need lactose for their live had better condition compared with probiotic in fermented soybean milk. Soybean milk had no lactose in it. Acid bacteria like *Streptococcus thermophilus* one of live bacteria which use carbohydrates especially lactose as a source of energy for growth, replication and metabolic processes.

Table 1 Effect of supplementation of fermented cow milk and soybean milk with probiotics on some hematological parameters of 35 d old broilers (means±standard error)

Treatments <sup>1</sup>	Hematological parameters <sup>2</sup>			
	Hb (g/dL)	PVC (%)	RBC (x10 <sup>6</sup> )	WBC (x10 <sup>6</sup> )
Control (T0)	11.9±0.65	30.0±1.11	2.5±0.07	24.2±0.11 <sup>a</sup>
T1	11.9±0.97	29.5±2.39	2.5±0.02	20.5±0.07 <sup>b</sup>
T2	12.7±1.13	31.3±2.71	2.7±0.02	19.8±0.15 <sup>c</sup>
T3	12.3±0.06	30.2±1.09	2.5±0.01	18.2±0.11 <sup>c</sup>

<sup>a, b, c</sup>Mean values on the same column not sharing a superscript are significantly different ( $p < 0.05$ )

<sup>1</sup>Hb: haemoglobin; PVC: packed volume cell; RBC: red blood cell; and WBC: white blood cell

<sup>2</sup>T0: basal diet (no supplement), T1: basal diet+ fermented cow milk with *Streptococcus thermophilus*, *Lactobacillus bulgaricus*, and *Lactobacillus acidophilus*, T2: basal diet + fermented soybean milk with *S. thermophilus*, *L. bulgaricus*, and *L. acidophilus*, T3: basal diet + 50% fermented cow milk and 50% fermented soybean milk with *S. thermophilus*, *L. bulgaricus*, and *L. acidophilus*.

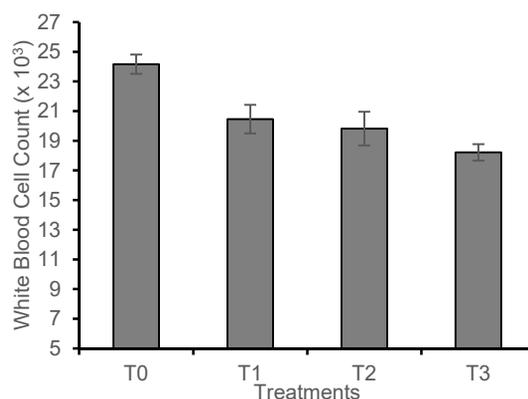


Fig. 1 Effect of supplementation of fermented cow milk and soybean milk with probiotics on white blood cell count of 35 d old broilers (means±standard error). T0: basal diet (no supplement), T1: basal diet+ fermented cow milk with *Streptococcus thermophilus*, *Lactobacillus bulgaricus*, and *Lactobacillus acidophilus*, T2: basal diet + fermented soybean milk with *S. thermophilus*, *L. bulgaricus*, and *L. acidophilus*, T3: basal diet + 50% fermented cow milk and 50% fermented soybean milk with *S. thermophilus*, *L. bulgaricus*, and *L. acidophilus*

Low WBC count found in birds fed probiotics in combination media of fermented cow milk and soybean milk may also be due to the genistein (GEN) as component of isoflavones in soybean milk. Previous study found that dietary GEN in the diet of broiler enhanced immune function by increasing antigen processing and presentation, macrophage activation, B lymphocyte, NK cell and helper T cell proliferation, and CD4<sup>+</sup> T lymphocyte differentiation. Moreover, dietary GEN also increased IgM and IgG concentrations, antibody titers, and antioxidant capacity [18].

It is assumed that isoflavones in soybean milk affect colonies of the probiotic and the WBC count in Broiler.

In the present study, there were no significant different on Hb, PVC and RBC among treatments. In line with previous study that the addition of probiotic to broiler diet had no significant effects on RBC count [15] and Hb concentration [1]. However, in other study reported that supplementation of probiotic to the broiler diet significantly increased Hb concentration [2]. Hb acts as a transporter of oxygen from the lungs to the tissues and as a transporter of carbon dioxide

from the tissues back to the lungs. This function depends on the molecular structure of hemoglobin, which contains four heme groups, each with a central iron molecule [13]. Due to the specific diet with low iron content for several weeks, animal might develop anemia, which has a negative impact on growth and feed conversion ratio [9, 12]. PCV is a variable related to the number of RBC in an animal. By definition, PCV is the ratio of the RBC to the volume of whole blood (which contains also WBC and plasma). PCV can be influenced by environmental challenges, such as dietary probiotic. Both higher and lower PCV values might be good indicators of an ongoing disease process. Broiler with diseases like diarrhea might experience excessive fluid losses that lead to higher PCV values. By contrast, lower PCV can be used for the diagnosis of anemia or other health problems [16]. It is assumed that there is no negative effect of dietary probiotic supplementation in fermented cow milk or soybean milk in broiler diet on some hematology profile.

## CONCLUSIONS

In conclusion, dietary supplementation of fermented cow milk and soybean milk with probiotics in the broiler diet could have positive effects on some hematological parameters of broilers (less inflammation indicated by low white blood cell count). This could have positive effects on performance thereby improving the physiological, health and metabolic activities of broiler.

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