



**6<sup>th</sup> International Seminar of Indonesian Society for Microbiology  
and  
11<sup>th</sup> Congress of Indonesian Society for Microbiology**

Pontianak, West Kalimantan, October 16 - 19 2013

NO. ISBN 978-602-8355-37-7

# **PROCEEDING**

**6<sup>th</sup> International Seminar of Indonesian  
Society for Microbiology**

**THE UNLIMITED USE OF MICROBIAL RESOURCES**

**Pontianak, West Kalimantan  
October 16-19, 2013**



**INDONESIAN SOCIETY FOR MICROBIOLOGY**

Proceeding

**The Unlimeted Used of Microbial Resources**  
6<sup>th</sup> International Seminar of Indonesia Society for Microbiology

Editor:  
Saeri Sagiman  
Sutarman Gafur  
Zakiatulyaqin

Badan Penerbit Universitas Tanjungpura

Proceeding

## ***The Unlimited Used of Microbial Resources***

6<sup>th</sup> International Seminar of Indonesia Society for Microbiology

Editor:

Saeri Sagiman  
Sutarman Gafur  
Zakiatulyaqin

Publikasi

"Untan Press" Anggota IKAPI No. 004/KLB/03  
Jl. Prof. Dr. Hadari Nawawi  
Telp. (0561) 739630, Fax (0561) 739637

Cetakan I, Januari 2014

Desain Cover : Nuki B. Nugroho

ISBN No. 978-602-8351-37-7

**Hak Cipta dilindungi oleh Undang-undang**

Dilarang Keras memperbanyak isi buku ini, sebagian atau keseluruhan dengan fotocopy, cetak dsb, tanpa iin tertulis dari penerbit

ANGGOTA IKAPI NO. 004/KLB/03

## Preface

As the Organizing Committee of The 6 International Seminar of Indonesian Society for Microbiology, 2013, in Aston Hotel Pontianak, we proudly present this Seminar Proceeding to the participants and to the society members. This proceeding is an important parts of dissemination process of experimental results from diverse expertise in Indonesian Society for Microbiology. Dissemination of experimetal results need to be done continuously so that the process of knowledge transfer could be accelerated.

The topic of the seminar is: Unlimited use of Microbia. There were 114 papers presented and 38 posters, however, there is only 32 of selected papers to be published in this proceeding. We greatly appreciate to all the participants who contributing for the seminar to be a great success. This seminar is an annual routine agenda of Indonesian Society for Microbiology. The objective were to disscus the current issues on microbiology, to innitiate knowledge transfer, and to trigger research collaboration and publications among the society members.

We, the Organizing Committee, deeply appologize if there is any mistake or any inconvenient we have made not only during seminar activities but also in this proceeding publication. We hope this proceeding will be very useful for you all. Finally, special thank for the auditors who have made this proceeding successfully published.

Dr. Ir. Sutarman Gafur, M.Sc.  
Chairman of Organizing Committee

## LIST OF CONTENT

	Title	Page
<b>PREFACE</b>		i
<b>LIST OF CONTENT</b>		ii
<b>ORAL PRESENTATION</b>		
	<b>Amplification Of Fusion Protein Gene Of Newcastle Disease Virus And Matrix Gene Of Avian Influenza Virus From Field Specimens By Multiplex Rt-Pcr Method.....</b>	<b>1</b>
	Aris Haryanto, Sri Handayani Irianingsih, Verawati, David Kristiawan, Bibiana Krisanti, Dini Wahyu Yudianingtyas	
	<b>Isolation Of Antibiotics Resistant Bact Eria From Soil Sample Of Hospitals In Purwokerto .....</b>	<b>12</b>
	Binar Asrining Dhiani, Dinar Aprilian Suwandi, Lisna Yulianis, And Retno Wahyuningrum	
	<b>Methanol Extract Of Kesum (<i>Polygonum minus</i> Huds) Leaves Antiseptic Gel Formulation .....</b>	<b>18</b>
	Famella Yulistia Pramita, Isnindar, Siti Nani Nurbaeti	
	<b>Comparison Of Antibacterial Efectivity From Kesum (<i>Polygonum minus</i> Huds.) Methanol Extract Againts Methanol Fraction .....</b>	<b>30</b>
	Siti Nani Nurbaeti, Rafika Sari, Liza Pratiwi	
	<b>Resistance Induction Of Oil Palm Clonalto <i>Ganoderma boninense</i> Using Endophytic Microbes.....</b>	<b>37</b>
	Hana Christine S, Wisnu Adi W, Rika Fithri Nurani B, Andriessa P, Elizabeth Caroline S, Helena Patricia M Ahmad Wahyu S, Giono, Nurita Toruan-Mathius, Tony Liwang	
	<b>Quality Of Agricultural By Products Fermented By <i>Trichoderma harzianum</i>.....</b>	<b>47</b>
	Nurhayati and Nelwida	
	<b>Diversity Of <i>Pythiaceae</i> Isolated From Durian Fields In Indonesia .....</b>	<b>54</b>
	Panca Jarot Santoso, Adi Pancoro, Sony Suhandono, and I Nyoman Pugeg Aryantha	
	<b>Formulation Of Lengkuas (<i>Alpinia Galanga</i> Linn) Rhizome Extract Gel As Antifungal With Carbopol Base .....</b>	<b>63</b>
	Yuni Eka Wati, Wintari Taurina Isnindar	
	<b>Phosphate Solubilizing And Sulfur Oxidizing Microbes Isolated From Andisols Dieng, Central Java, Indonesia: Their Potency As Local Microbes For Biofertilizer Inoculum. ....</b>	<b>71</b>
	Sudadi, Widyatmani Sih Dewi, Ika Ernawati and Linda Habsari Efendi Putri	
	<b>Molasses As Potential Carbon Source For Marine Yeast Growth. ....</b>	<b>81</b>
	Sukoso dan Juniar Adhani A	
	<b>In Vitro Test Of Probiotic Strain Of Seven Species Of Fungi Which Isolated From Gastrointestinal Tract Of Indigenous Chicken .....</b>	<b>88</b>
	T. Yudiarti , V. D. Yunianto B.I', R. Murwani, E. Kusdiyantini	

# Effect of Feed Supplement Containing Probiotic *Bacillus* Plus Vitamins and Minerals to Performance and Internal Organ Development of Broiler

Yudiarti T, I. Isroli, E. Widiastuti and S. Sugiharto

*Faculty of Animal and Agricultural Sciences, Diponegoro University  
Semarang, Central Java, Indonesia*

## Abstract

The aim of the study was to investigate the effect of feed supplement containing probiotic *Bacillus* plus vitamins and minerals to performance and internal organ development of broiler. Two hundred and eighty eight 1-day-old broiler chicks were randomly allotted in four groups, i.e., control birds (birds receiving diet without supplement) and birds receiving supplement either 0.1%, 0.5% or 1% from total diets. The supplement (probiotic *Bacillus* plus vitamins and minerals) contained 12.10 log cfu/g mixture of probiotic *Bacillus*, 0.100 mg vit A, 0.018 mg vit D<sub>3</sub>, 0.100 mg vit E, 1200 mg Ca, 750 mg P, 0.08 mg Mg, 0.006 mg Co, 0.045 mg Cu, 0.015 mg Se, 0.180 mg S, 0.010 mg Zn, 0.060 mg KCl, 0.030 mg I, 0.060 mg Fe and 0.100 mg Mn. The chicks were reared for 14 days, and internal organs was collected at day 12 of the experiment. Feed conversion ratio (FCR) was lower ( $P \leq 0.05$ ) in birds fed diet containing 0.5% supplement as compared to control. Body weight and accumulative feed intake of birds at day 14 was not different ( $P > 0.05$ ) among the treatment groups. At day 12, dietary supplementation (0.1% or 1% from total diet) increased ( $P \leq 0.05$ ) the relative weight of ileum of broiler chicks. The conclusion was dietary supplementation with probiotic *Bacillus* plus vitamins and minerals was improved development of ileum and feed efficiency of broiler.

**Keywords:** broiler, standard commercial feed, performance, probiotic, mineral, vitamin

## Introduction

In Indonesia, broiler farmers mostly use commercial feeds produced by feed mill industries. Commercial feeds have generally been formulated to meet the standard nutrient requirements of broilers. However, feed handling on the farm such as storage conditions and length of time in farm feed bins may adversely change the composition of feeds especially vitamins and minerals (Aviagen 2014). This may to some extent explain why deficiency diseases (particularly vitamin and mineral deficiencies) and the retarded growth rate still often found in broilers fed standard commercial feeds (Islam et al 2004; Rahman et al 2012; Harun-Ar-Rashid et al 2015). For broiler chicks, brooding period (the first two weeks of the broiler's life) is the critical stage since this period accounts a third of broiler's life (Müller 2013). Indeed, vitamin and mineral deficiencies during the brooding period may have pronounced effect as it depress the later performance of broilers (Aviagen 2014). To deal with such deficiency diseases (especially during brooding period), broiler farmers randomly supplement vitamins-mineral premix to the feeds. The farmers also supplement growth promoters to have improved growth performance and better feed efficiency of broilers (Rahman et al 2012). It has been recognized that with holding birds from feed for 24 h after hatching resulted not only in retarded growth and delayed development of the intestine, but also compromised immune responses of broiler chicks (Bhanja et al 2010). Due to logistics and transportation, feed with holding for more than 24 h however often occurs in commercial broiler production in Indonesia (Sugiharto and Lauridsen 2016). Indeed, most of broiler feeds available in the

market nowadays do not have enough components/nutrients for compensating the post-hatch feed with holding-induced growth retardation and immune suppression (Sanda et al 2015). Extra supplementation of commercial broiler diets with antibiotic growth promoters (AGPs) may be applied to enhance growth rate and immune responses of broiler chickens (Lee et al 2012; Manafi 2015). However, the global concern about antibiotic resistance both in broilers and human (as consumers) have encouraged nutritionists to reduce or substitute AGPs with natural growth promoters and immune system booster. Of the alternative to AGPs, probiotics have attracted great interest as it may improve growth performance and modulate immune system of broilers (Sugiharto 2016). Taken together, the present study aimed to investigate the effect of dietary supplement containing probiotic *Bacillus* plus vitamins and minerals on the performance, and internal organ development of broilers post brooding.

### **Materials and methods**

Two hundred and eighty eight Lohmann (MB-202) 1-day-old broiler chicks (average body weight =  $45.9 \pm 0.50$  g) were used in the current study. They were withheld from feed and water for 24 h post-hatch to mimic the real condition of broiler chicks handling in Indonesia. In an open-sided naturally ventilated broiler house, the chicks were randomly allotted to four experimental groups of 72 chicks each and 6 replicates of 12 chicks. The groups included control birds (birds receiving diet without probiotic *Bacillus* plus vitamins and minerals) and birds receiving probiotic *Bacillus* plus vitamins and minerals either 0.1%, 0.5% or 1% from total diets. For the entire study period (14 days), the chicks were fed standard commercial diet containing 13% moisture, 21-23% crude protein, 5% crude fat, 5% crude fiber and 7% crude ash (data were obtained from the company). The diets and water were offered *ad libitum* for the entire of experiment. The supplements were added at the expense of the diets. The supplement (probiotic *Bacillus* plus vitamins and minerals) contained  $12.10 \log$  cfu/g mixture of *Bacillus* probiotics (i.e., *Bacillus cereus* strain SIIA\_Pb\_E3, *Bacillus licheniformis* strain FJAT-29133, *Bacillus megaterium* strain F4-2-27 and *Bacillus* sp. 11CM31Y12). In addition to that, the supplement contained 0.100 mg vit A, 0.018 mg vit D<sub>3</sub>, 0.100 mg vit E, 1200 mg Ca, 750 mg P, 0.08 mg Mg, 0.006 mg Co, 0.045 mg Cu, 0.015 mg Se, 0.180 mg S, 0.010 mg Zn, 0.060 mg KCl, 0.030 mg I, 0.060 mg Fe and 0.100 mg Mn. Body weight, accumulative feed intake and feed conversion ratio (FCR) of chicks were recorded at day 14 of the experiment. At day 12, the chicks were then sacrificed and, immediately, the internal organs were taken out and weighed. Data were analysed according to a completely randomized design by ANOVA using the General Linear Models Procedure in SAS (SAS Inst. Inc., Cary, NC, USA). Pen was regarded as the experimental unit during the analysis. Significant differences among experimental groups were later analysed using Duncan's multiple-range test. A significant level of  $P \leq 0.05$  was implemented.

### **Results and Discussion**

#### ***Performance and internal organs of broiler chicks***

It has been known that optimal growth performance during brooding period is critical, as it determines the final body weight of broiler chickens. Several factors have been recognized to adversely affect the performance of broilers during brooding period, one of which is the delayed access to feed and water (due to logistics and long transport from hatchery to commercial farms) (Müller 2013). In the present study, the body weight of broiler chicks at day 14 was lower as compared to the standard body weight of Lohmann broiler chicks at the same age (Aviagen 2007). This seemed to be the effect of feed and water with holding during the first 24 h applied to chicks in the present study. Indeed, the body weight as well as feed intake of birds at day 14 was not different ( $P > 0.05$ ) among the treatment groups. Compared to control, however, FCR was lower ( $P \leq 0.05$ ) in birds fed diet containing 0.5%

supplement (Table 1). This result suggested that dietary supplementation with probiotic *Bacillus* plus vitamins and minerals was beneficial in improving the feed efficiency of birds fed standard commercial diets. With regard to probiotics, recent study showed that such additive was beneficial for the improved nutrient metabolism and utilization by broiler chicks (Sugiharto 2016; Zheng et al 2016). Concomitantly, vitamins and minerals are essential for optimal feed utilization and metabolism in poultry (Islam et al 2004). Taken together, supplementation of standard commercial diets with probiotic *Bacillus* plus vitamins and minerals may therefore be beneficial for the improved metabolism and thus feed utilization and efficiency of birds (Islam et al 2004; Rahman et al 2012; Dhama et al 2014).

**Table 1.** Performances of broiler chicks<sup>†</sup>

Items	Dietary treatments				SE	Pvalue
	Control	Pro-0.1	Pro-0.5	Pro-1.0		
Body weight(g/bird)	318	324	329	310	8.72	0.44
Feed intake(g/bird)	403	395	386	392	14.7	0.88
FCR	1.47 <sup>a</sup>	1.42 <sup>ab</sup>	1.36 <sup>b</sup>	1.48 <sup>a</sup>	0.02	<0.01

<sup>†</sup>Data were collected at day 14

Control= birds receiving diet without supplementation; Pro-0.1= birds receiving 0.1% probiotic *Bacillus* plus vitamins and minerals; Pro-0.5= birds receiving 0.5% probiotic *Bacillus* plus vitamins and minerals; Pro-1.0= birds receiving 1% probiotic *Bacillus* plus vitamins and minerals; SE= standard error

<sup>a,b</sup>Values with different letters within the same row were significantly different

Result in the present study showed that supplementation of standard commercial diets with 0.1% and 1% probiotic *Bacillus* plus vitamins and minerals increased ( $P \leq 0.05$ ) the relative weight of ileum of broiler chicks at day 12. Concomitant with this, Shabani et al (2012) reported that feeding probiotic (protexin) was able to increase the relative weight of ileum of broiler chickens. With regard to other internal organs, the current treatments, however, had no significant impact (Table 2). The mechanism through which probiotics increased the relative weight of ileum in the current study was not clear, as the effect of probiotics on the internal organ weight in broilers is equivocal. Indeed, Olnood et al (2015) found no difference in the relative weight of internal organs (liver, spleen, bursa, duodenum, jejunum, ileum) of broilers when feeding probiotic *Lactobacillus johnsonii*. In addition to probiotic effect, minerals and vitamins may also contribute to the enhanced weight of ileum of broiler chicks in the present study. Islam et al (2004) suggested that supplementation of standard commercial diets with mineral-vitamin premix may enhance digestion, absorption and metabolism, and thus improve the intestinal development of broilers.

**Table 2.** Internal organs of broiler chicks<sup>†</sup>

Items (% live body weight)	Dietary treatments				SE	Pvalue
	Control	Pro-0.1	Pro-0.5	Pro-1.0		
Heart	0.82	0.79	0.77	0.89	0.04	0.12
Liver	2.99	2.75	2.84	3.13	0.11	0.09
Spleen	0.08	0.07	0.09	0.09	0.01	0.36
Bursa of Fabricius	0.23	0.23	0.26	0.29	0.03	0.39
Duodenum	1.57	1.59	1.58	1.58	0.08	0.99
Jejunum	2.49	2.53	2.54	2.61	0.12	0.91
Ileum	1.38 <sup>a</sup>	1.69 <sup>b</sup>	1.52 <sup>ab</sup>	1.76 <sup>b</sup>	0.09	0.03

<sup>†</sup>Data were collected at day 12

Control= birds receiving diet without supplementation; Pro-0.1= birds receiving 0.1%

---

*probiotic Bacillus plus vitamins and minerals; Pro-0.5= birds receiving 0.5% probiotic Bacillus plus vitamins and minerals; Pro-1.0= birds receiving 1% probiotic Bacillus plus vitamins and minerals; SE= standard error*

*<sup>a,b</sup>Values with different letters within the same row were significantly different*

In conclusion, dietary supplementation with probiotic *Bacillus* plus vitamins and minerals was beneficial for the improved development of ileum and feed efficiency of broiler during brooding period.

### **Acknowledgements**

The study was funded by Directorate of Research and Community Service, the Ministry of Research, Technology and Higher Education of the Republic of Indonesia through Penelitian Unggulan Perguruan Tinggi.

### **References**

**Aviagen 2007** Lohmann meat broiler stock performance objectives. [www.aviagen.com](http://www.aviagen.com)

**Aviagen 2014** Ross broiler management handbook. [www.aviagen.com](http://www.aviagen.com)

**Bhanja S K, Devi C A, Panda A K and Sunder G S 2010.** Effect of post-hatch nutrient intubation on performance, intestinal growth, meat yield and immune response in broiler chickens. *Asian Australasian Journal of Animal Sciences*. 23: 515-520.

**Dhama K, Tiwari R, Khan R U, Chakraborty S, Gopi M, Karthik K, Saminathan M, Desingu P A and Sunkara L T 2014.** Growth promoters and novel feed additives improving poultry production and health, bioactive principles and beneficial applications: the trends and advances-a review. *International Journal of Pharmacology*10: 129-159.

**Harun-Ar-RashidM, AhmadN, AminM R and Mohammad LalmoddinMollah 2015** Effects of selected vitamins and minerals on growth rate and hematological parameters in broilers. *Asian Journal of Medical and Biological Research*. 1, 487-494.

**Islam M S, Bhuiyan M E R, Begum M I A, Miah M A and Myenuddin 2004** Effect of vitamin-mineral premix supplementation on body weight and certain haemato-biochemical values in broiler chickens. *Bangladesh Journal of Veterinary Medicine*. 2: 45-48.

**Lee K W, Lillehoj H S, Lee S H, Jang S I, Park M S, Bautista D A and Ritter G D 2012** Effect of dietary antimicrobials on immune status in broiler chickens. *Asian Australasian Journal of Animal Sciences*. 3: 382-392.

**Manafi M 2015.** Comparison study of a natural non-antibiotic growth promoter and a commercial probiotic on growth performance, immune response and biochemical parameters of broiler chicks. *Journal of Poultry Science*. 52: 274-281.

**Müller H 2013** Focus on optimal starting conditions for day-old chicks. *Lohmann Information*. 48: 34-37.

- Olnood C G, Beski S S M, Iji P A and Choct M 2015** Delivery routes for probiotics: Effects on broiler performance, intestinal morphology and gut microflora. *Animal Nutrition*. 1: 192-202.
- Rahman M A, Parvin M S, Sarker R R and Islam M T 2012** Effects of growth promoter and multivitamin-mineral premix supplementation on body weight gain in broiler chickens. *Journal of Bangladesh Agricultural University*. 10: 245-248.
- Sanda M E, Ezeibe M C O and Anene B M 2015** Effects of vitamins A, C and E and selenium on immune response of broilers to Newcastle disease (ND) vaccine. *IOSR Journal of Agriculture and Veterinary Science*. 8: 13-15.
- Shabani R, Nosrati M, Javandel F and Kioumars H 2012** The effect of probiotics on carcass and internal organs of broilers. *Annals of Biological Research*. 3: 5475-5477.
- Sugiharto S 2016** Role of nutraceuticals in gut health and growth performance of poultry. *Journal of the Saudi Society of Agricultural Sciences*. 15: 99-111.
- Sugiharto S and Lauridsen C 2016** Dietary *Chlorella* supplementation effect on immune responses and growth performances of broiler chickens exposed to post hatch holding time. *Livestock Research for Rural Development*.  
<http://www.lrrd.org/lrrd28/7/sugi28119.html>
- Zheng A, Luo J, Meng K, Li J, Bryden W L, Chang W, Zhang S, Wang L X N, Liu G and Yao B 2016** Probiotic (*Enterococcus faecium*) induced responses of the hepatic proteome improves metabolic efficiency of broiler chickens (*Gallus gallus*). *BMC Genomics*. 17: 89. Doi: 10.1186/s12864-016-2371-5