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The 2nd International Seminar

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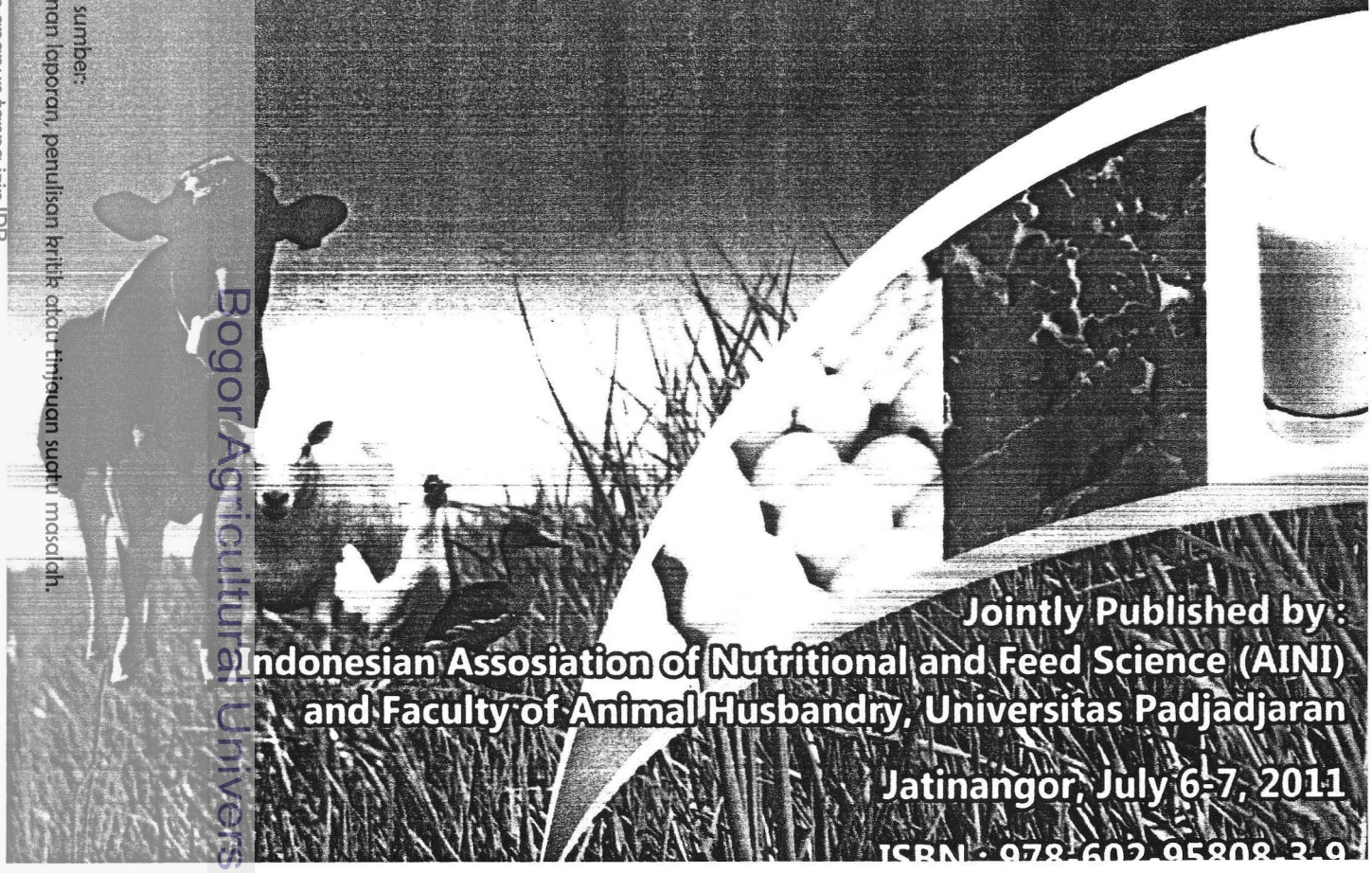

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The 2nd International Seminar
"Feed Safety for Healty Food"

Keynote Speaker :

Director General of Animal Husbandry and Animal Health

Main Speakers :

Prof. Fr. Jurgen Zentek (Berlin, German)

Prof. Abdul Razak Alimon (Malaysia)

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FOREWORD

We thank the Almighty Allah, the Most Gracious and the Most Merciful that the proceedings of the 2nd International Seminar, the 8th Biannual Meeting and 3rd Congress and Workshop of AINI with the theme "Feed Safety for Healthy Food" organized by Indonesian Association of Nutrition and Feed Science, Faculty of Animal Husbandry, Universitas Padjadjaran on 6 - 7 July 2011 have been completed.

These activities were to collect variety of scientific information with the purpose to collect scientific information about feed for a healthy food, to produce a draft policy on a national feed system and to make a scientific forum for Academics, Researchers, Practitioners of animal husbandry, Health and Policy makers. Scientific papers that were presented either in oral or poster stated in the proceedings.

Thanks go to all those who have provided both moral support or material so that this seminar can be carried out and the proceeding can be issued.

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EFFECTS OF SUPLEMENTAL ORGANIC CHROMIUM AND FUNGI *Ganoderma lucidum* ON MILK PRODUCTION AND IMMUNE RESPONSE IN LACTATING COWS

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ABSTRACT

Chromium (Cr) is an essential miconutrient for humans and animals required for carbohydrate, lipid, protein and nucleic acid metabolism, regulation hormonal and immune function. *Ganoderma lucidum*, an fungus, has bioactive compounds (polisaccharides) wich function for enhancing immune response and it can also be used to synthesize organic Cr. The aim of this experiment was to determine the effect of organic Cr and *Ganoderma lucidum* suplementation on milk production and immune respon of lactating cows. Fifteen lactating cows grouped and allocated in the five treatments in randomize block design, fed a basal diet composed of 60% grass and 40% concentrate. Suplementation on basal diet as treatment were : A= basal diet unsuplemented (control), B= 3 ppm inorganic Cr $CrCl_3$, C= 3 ppm organic Cr (fermentation product of fibrus feed with *Ganoderma lucidum*), D= . *Ganoderma lucidum* (5 g/ 50 kg live weight) and E= Organic Cr + *Ganoderma lucidum*. The result showed that there is no significant differences on dry matter and organic matter intake, and milk production, but Cr intake and in milk increased with organic Cr supplementation and its combination with *G. lucidum*. Suplementation of *Ganoderma lucidum* and its combination with organic Cr tend to reduce somatic sel count, mastitis skor and coloni of microorganism in milk yield. It can be concluded that *Ganoderma lucidum* and organic Cr in combination form with *Ganoderma lucidum* can improved immune response of lactating cows.

Key words: organic chromium, *Ganoderma lucidum*, milk, immune response, lactating cows.

INTRODUCTION

Milk production during lactation is affected by several factors eg. the number of functional mammary epithelial cells (degree of mammary glands development) at the beginning of lactation (Anderson, 1985),.and infection diseases. Mastitis is one of the infection diseases which can reduces milk production and milk quality, and is a frequent reason for culling cows (Bath *et al.* 1985). Subclinical mastitis is the most prevalent. It

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occures 20-30 times more frequently than the clinical forms and cause far greater losses, although it cannot be recognized unless appliying particular methods of examination (Gravert, 1987). Cow's milk infected from mastitis when milk content high somatic cell numbers that are exceeded 400 000 /ml (Sudarwanto, 2008) or exceed 500 000 /ml (Gravert, 1987) and is founded pathogen microorganism. Bacteria that cause mastitis are *Staphlococcus aureus*, *Streptococcus agalactiae*, *Str.dysgalactiae*, *Str.uberis* (Gravert 1987).

The another important factor that influences milk production is the availability of nutrients as precursors of milk components in the mammary epithelial cells (Collier, 1985), Chromium is established as an essential trace element for man and laborarory animals (NRC, 1997). Trivalent chromium is essential to normal carbohydrate, protein and lipid metabolism, hormonal regulation and immune function. The physiological function of chromium is as an integral component of biologically active chromium or Glucose Tolerance Factor (GTF), which function as cofactor of the vital hormone insulin (Anderson, 1998; Vincent, 2000). Chromium is biologically active as part of chromodulim (Vincent, 1999). Chromium may be present in the form of inorganic compounds or organic complexes (Pechova & Pavlata, 2007). Inorganic chromium is very poorly absorbed (0.4-2.0%). It must be converted to an organic complex to enable the physiological functioning of chromium. Conversion of inorganic chromium (eg. chromium chloride) in the liver or kidney to the bioactive form may be slow (Chang & Mowat, 1992). Organic chromium can be synthesize by using a fungi *Ganoderma lucidum* that can incorporates into their cell (Yang *et. al.*, 2006).

Ganoderma lucidum (Lingzhi in Chinese, Mannentake or Sachitake or Reshi in Japanese, and Youngzhi in Korea) is a species of the class Basidomycetes which belongs to the family Poliporaceae (or Ganodermataceae) of the order Aphyllophorales (Chang & Miles, 2004). Commonly known as a wood decaying fungus, it causes white root of a wide variety of trees. Because of its perceived health benefits, its fruiting body has gained wide popularity in recent years as a dietary supplement (Chang & Miles, 2004). *Ganoderma lucidum* is a unique mushroom species and has been used as a tonic and drug for more than 2000 years. It is very famous in China and other countries of Asia for its extensive physiological effect. The immune system is a physiological system, thereby protect the host against pathogens.

Ganoderma lucidum has been shown to posses potent antioxidant in multiple research studies with little or no side effect (Yen & Wu, 1999; You & Lin, 2003). The major compounds with pharmacology activities appear to be triterpens, polysaccharides, bioactive proteins and nucleic acids which function as immunomodulator (Lin & Zang, 2005; Gao *et. al.*, 2005); antioxidant (Chen *et al.*, 2005; Sun *et. al.*, 2004; Zao *et.al.*, 2004). *Ganoderma lucidum* can also be used as carrier to synthesize organic chromium. However, little information is available about the effect of organic chromium and *G. lucidum* on milk production and immune response in lactating cows especially for subclinical mastitis. Therefore, the objectives of this study were to investigate the effect of supplemental organic chromium and *G. lucidum* on milk production, mastitis test and number of bacteria in milk.

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MATERIALS AND METHODS

Animals and Diets

Fifteen lactating cows in late lactation were randomly allocated from experimental block based on milk production. Animals fed a basal diet, consisted of 60% grass and 40% concentrate and supplemented with chromium (organic Cr and inorganic Cr), fruiting bodies of *G. lucidum* and combination of organic chromium.and *G. lucidum*. Supplementation on basal diet as treatment were : A= basal diet (unsupplemented), B= 3 ppm inorganic Cr $CrCl_3$, C= 3 ppm organic Cr (fermentation product of fibrus feed with *Ganoderma lucidum*), D= . *Ganoderma lucidum* (5 g/50 kg live weight) and E= organic Cr + *Ganoderma lucidum*. This experiment was conducted in two periods: preliminary period (3 weeks) and collecting period (5 weeks). The animals were fed twice a day according to body weight and milk production, based feeding scale (NRC, 2001), whereas water was available *ad libitum*. Feed, blood and milk samples were collected in collecting period. Nutrient intake, milk yield, chromium content in milk and blood glucose were observed.

Blood and Milk Samples

Blood samples were collected from each cow before preliminary period and at the end of collecting period for glucose analysis. Blood samples were kept on ice box immediately until analysis. Milk samples were collected periodically once a week from each cow for eigh weeks. The milk samples were also kept on ice box immediately until analysis.

Chemical Analysis

Dry matter and organic matter analysis of feed were carried out accorging to proximate analysis (AOAC, 1990). Analysis of chromium content in feed and milk were started with wet ashing and were determined by atomic absorption spectrometry (AAS). Sub clinical mastitis test was determined by IPB-1 method and by counting somatic cell count (SSC). The number of microorganisms present in milk samples was counted by using total plate count (TPC) with Kock method (Sudarwanto, 2008).

Statistical Analysis

Statistical analysis were performed with general linear model (GLM) procedure of statistical analysis system (SAS, 1997). Analysis of variance was performed by ANOVA procedure. Significant difference between means were determined by Duncan's multiple range test.

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RESULTS AND DISCUSSION

Environmental Temperature

Average of air temperature was 22.85 ± 0.41^{0}C in the morning and 32.62^{0}C in the middle of day. Respiration of cows was 30.6 ± 4.7 time/minute in the morning and 44.9 ± 6.3 time minute in the afternoon. The air temperature during experiment was higher than thermoneutral for dairy cattle (18-20^{0}C). The air temperature at the middle of day have coused the cows was heat stress because air temperature in the middle of day was higher than critical maximum temperature (25^{0}C – 26^{0}C) (Yousef, 1985; Jones & Stallings, 2008). Therefore, the cows have increased their respiration up to 44.9 ± 6.3 time/minute.

Dry Matter Intake

There were no significant difference on dry matter and organic matter intake by both chromium supplememtation: organic chromium and inorganic chromium, *G. lucidum* supplementation, and organic chromium + *G. lucidum* supplementation (Table 1).

Table 1. Effect of supplemental organic chromium and *Ganoderma lucidum* on dry matter and organic matter intake and blood glucose level in lactating cows

Parameter	Treatment				
	Control	Inorganic Cr	Organic Cr	Gano derma	Organic Cr + Ganoderma
Dry matter intake (kg/day))	12.16±0.91	11.86±1.08	11.49±0.71	12.02±0.92	12.67±1.51
Organic matter intake (kg/day)	9.21±0.74	9.03±0.80	8.81±0.64	9.21±0.66	9.55±1.06
Blood glucose (mg/dl)	52.5±0.28	55.58±8.24	58.13±4.46	64.12±3.86	63.58±2/10

Dry matter and organic matter intake of cows fed inorganic and organic chromium were 11.86 ± 1.08 kg/day and 11.49 ± o.71 kg/day respectively, whereas control was 12.16 ± 0.91 kg/day. Suplementation of *G. lucidum* and combination organic chromium with *G. lucidum* were also not significant different on dry matter and organic matter intake. This is consistent with Subiyatno *et. al.* (1996), who also founded that dry matter intake were not affected by chromium supplementation when fed to lactating cows. There were also no differences among treatment for dry matter intake when chromium was supplemented to cattle during the growing period.and stress condition (Chang & Mowat, 1992. In contrast, Al-Saidy *et al.*, (2004) founded that chromium increases dry matter intake when each cow fed 4 g of chromium yeast a day during 4 months lactation.

Blood Glucose

The concentration of blood glucose was not different among treatment. Supplementation of chromium or *G. lucidum* and its combination could not change the blood glucose concentration. Each animal could reach a normal level of blood glucose

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for ruminant (52 mg/dl) (Larson, 1974). However, there were indication that supplementation of organic chromium and *G. lucidum* tend to increase with a blood glucose level 58.13 mg/dl and 64.12 mg/dl respectively. Chromium is needed as a cofactor for insulin in moving glucose from circulation into peripheral tissue (Anderson,1998).

Milk Production

There was no difference in milk yield after treatment for 8 weeks experiment. However, milk production was numerically increased when cows supplemented with organic chromium. Milk production of cows supplemented with organic chromium was 92.99% and this value is the highest.

Table 2. Effect of Suplemental organic chromium and Fungi *Ganoderma lucidum* on milk production, chromium intake and chromium in milk in lactating cows

Parameter	Treatment				
	Control	Inorganic Cr	Organic Cr	Gano derma	Organic Cr+ Ganoderma
Milk yield before treatment (kg/day)	6.34±3.49	5.82±3.89	4.28±2.08	5.99±3.79	6.84±4.48
Milk yield after treatment (kg/day)	5.52±2.01	5.11±2.10	3.98±1.76	5.34±3.01	6.14±4.40
Milk production (%)	87.07	87.8	92.99	89.15	89.77
Chromium:					
Cr intake (mg/day)	114.1±8.5[a]	146.8±13.4[b]	142.2±8.8[b]	112.7±8.6[a]	156.8±19.7[c]
Cr in milk (ppm)	0.39±0.07[a]	0.54±0.19[b]	1.03±0.09[c]	0.53±0.07[a]	0.85±0.35[c]

[a-c] Mean in same raw having different letter are significantly different (P<0.01).

Chromium in Milk

Chromium in milk and Cr intake in lactating cows supplemented with chromium fed basal ration which content 9.39 ppm chromium was shown in Table 2. Chromium intake increased significantly (P<0.01) when animals supplemented with chromium. Chromium in milk also significantly different (P<0.01). Organic chromium was more readily absorbed so its value in milk is the highest (1.03 ± 0.09 ppm).

Immune Response of Lactating Cows to Mastitis

Supplementation of *G. lucidum* and its combination with organic chromium have decreased amount of somatic cell count. 3 ppm organic chromium in combination with *G. lucidum* (5 g/50 kg live weight) decreased somatic cell count up to 300 cell/ml, which was lower than standard value of mastitis (400 000 cell/ml) (Table 3). Mastitis score also be low when cows supplemented with combination of organic chromium and *G. lucidum*. The number of microorganism in milk also decreased up to < 1 estimated. This indicated that cows is not anymore in subclinical mastitis condition.

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Table 3. Mastitis test and number of bacteria in milk of lactating cows supplemented wit h organic chromium and *G. lucidum*

Parameter	Treatment				
	Control	Inorganic Cr	Organic Cr	Ganoderma	Organic Cr+ Ganoderma
Somatic cell count (SCC) x 10^5 cell/ml					
Before treatment	15.2±10.8	31.7±36.7	6±3.8	9.1±6.5	24.3±36.5
4 weeks	14±16.6	5.9±4.1	15.2±16.4	9.6±13.2	10.1±15.5
6 weeks	28.3±13	17.1±20.4	42.7±54.5	5.3±1.4	35.9±37.8
8 weeks	8±9.8	6.1±1.1	12.7±9.3	5.3±2.2	3.1±3.1
Mastitis test with IPB-1 method (subclinical mastitis score : 0, 1, 2, 3)					
Before treatment	1.3±0.6	2±1	0.7±0.6	1±1	1±1.7
4 weeks	1.3±0.6	1±0	1.3±0.6	1±1	0.7±1.1
6 weeks	1±0	1±0	1.3±0.6	0.7±0.6	1.3±1.5
8 weeks	0.7±1.1	1±0	1.7±0.6	0.7±0.6	0.3±0.7
Number of Bacteria (Koch method) x 10^4 CFU/ml					
Before treatment	7.4±11.7	0.1±0.06	3.2±2.3	1.9±1.8	0.8±1.07
4 weeks	0.48±0.16	0.83±0.49	0.73±0.32	0.42±0.18	1±0.28
6 weeks	1.03±0.63	1.11±0.69	5.38±6.66	2.06±1.39	0.44±0.27
8 weeks	3.13±2.26	2.13±0.56	5.932.97	1.07	<100 est

CONCLUSION

It can be concluded that supplementation of *Ganoderma lucidum* 5 g/50 kg live weight and 3 ppm organic Cr in combination form with *Ganoderma lucidum* can improved immune response of lactating cows to prevented subclinical mastitis.

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