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by Fm Suhartati

Increasing of Conjugated Linoleic Acid of Dairy Milk with Additional Rice Meal Fermented and Soybean Oil

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Abstract

An experiment has been carried out to produce milk containing conjugated linoleic acid in high concentrations. The study was carried out experiments in vivo, using a Latin Square Design 6x6x6, with factorial 2x3 treatment. The first factor (A) was a type of diet, consisting of feed used for cows in group of dairy farmers Margo Mulyo, Kemutug village, district Baturaden, Banyumas (a1), and concentrate containing 30% rice bran + 20% tapioca by product which was fermented using Saccharomyces cerevisiae, +3% soybean oil (a2). As a second factor (B) were the time of milking, that are before grass feeding (b1), 2 hours after grass feeding (b2), and 4 hours after grass feeding (b3), thus there were 6 treatments. As the column were 6 sampling periods and as the row were 6 cows. The variables measured were fat, and conjugated linoleic acid of milk. The results indicated that concentrate containing 30% rice bran + 20% tapioca by product which was fermented using Saccharomyces cerevisiae + 3% soybean oil increasing 88.89% of conjugated linoleic acid and time of milking 4 hours after grass feeding increasing 30.56% of conjugated linoleic acid compare time of milking that are before grass feeding and increasing 34.29% if compare time of milking 2 hours after gras feeding. Based on the results of this study concluded that the concentrate containing 30% rice bran + 20% tapioca by product which is fermented using Saccharomyces cerevisiae + 3% soybean oil was the best feed formula, and cows were milked four hours after grass feeding was the best milking time.

Keywords: conjugated lenoleic acid, milking interval, soybean oil.

Introduction

The high content of fat and cholesterol in the body, the occurrence of atherosclerosis and cancer are a problem for many people, as a result of increasing incomes and change of diets. These problems can be overcome by consuming foods that contain conjugated linoleic acid (CLA).

CLA is an essential nutrient, present in ruminant products, including milk, as anticarcinogenic (Donovan *et al*., 2000), to prevent mammary gland tumors (Garcia *et al*., 2000) antiatherogenic (Scollan *et al*., 2006), lean body mass promotans and anti-diabetic (Gulati *et al*., 2000), anti-oxidative, cholesterol-depressing and promoting growth, reducing atherosclerosis, and lowering adipose fat (Gillis, 2004), increasing High Density Lipoprotein (HDL) blood plasma (Choi *et al*., 2006). CLA is synthesized either in the rumen as a result of incomplete bio-hydrogenation of linoleic acid (Noci *et al*., 2005), as well as on tissues through the Δ9-desaturation vaccenic acid (VA; 18:1 trans-11) (Corl *et al*., 2001). Based on the benefits that have been described, it's indicating that the CLA is required for human health.

Nutritional strategies to enrich CLA production in ruminant products can be achieved by increasing the supply of C18: 2 (linoleic acid) in the reticulo-rumen. The acid can be obtained from the molds, which are bioactive and contain linoleic acid. Results of the experiment made by Suhartati *et al.*, (2005) showed that the pollard fermentation using Saccharomyces cerevisiae was able to increase linoleic acid, but the results have not been significant. Pollard is an energy source that can be replaced by other feedstuffs that is easy to be obtained, i.e. rice bran and tapioca byproduct. To get the linoleic acid in sufficient quantity, we still need to add a compound that is able to supply a source of linoleic acid, i.e. vegetable oils (Flowers *et al.*, 2007) or grain oils (Pavan and Duckett, 2007). Soybean oil as grain oil, is a good source of unsaturated fatty acids (Kahrizi *et al.*, 2007), which is able to increase the content of linoleic acid in the substrate, which will be hydrogenated by rumen bacteria and will produce intermediate compounds, i.e. CLA which is needed by humans.

Materials and Methods

The materials for research were six cows, grass, concentrates and soybean oil. This study was conducted with experimental methods in vivo, using a Latin Square 6 x 6 x 6. The treatment is arranged in factorial 2 x 3. The first factor (A) was type of diet, which consisted of the diet used for cows in the Margo Mulyo Group (a1) and concentrates containing 30% rice bran + 20% tapioca byproduct which were fermented using Saccharomyces cerevisiae, plus 3% soybean oil (a2). Nutrient content of grass and concentrates containing 30% rice bran + 20% tapioca byproduct fermented using Saccharomyces cerevisiae are listed in Table 1. As a second factor (B) was milking time after grass feeding, before grass feeding (b1), 2 hours after grass feeding (b2) and 4 hours (b3) after grass feeding. As the columns were 6 periods of sampling and the line included 6 cows. The variables measured were fat content, which was measured using Lacto Scan and CLA of milk, which was measured using Chromatography Gas. Data obtained was analyzed using analysis of variance, Directly Significant Different test and to find the response an Orthogonal polynomials was performed (Steel and Torrie, 1993)

Table 1. The nutrient content of grass and concentrates

Feedstuff	DM (%)	Based on Dry Matter (DM)				
		Crude Protein	Extract Ether	Crude Fibre	Ash	Nitrogen Free Extract
Grass	12.80	9.27	2.84	33.58	11.96	42.35
Concentrates containing fermented rice bran and tapioca byproduct	70.37	13.88	6.26	12.29	4.37	59.06
Control diet	86.27	10.04	15.83	19.69	8.98	45.46

Description: Analysis at Nutrition and Feed Science Laboratory, Animal Science Faculty of Jenderal Soedirman University (2010)

Results and Discussion

Milk fat

Milk fat content ranged from 2.85% to 4.45% (Table 2). Based on the analysis of variance, diets and the interaction between diet and milking time had no significant effect (P> 0.05) on milk fat content, while the milking time had significant effect (P <0.01) on milk fat content.

Based on HSD test, the milk fat content before grass feeding was lower than 2 and 4 h after grass feeding. The orthogonal polynomial test showed that milking time had quadrater responses (P <0.01) on the fat content of milk by the equation: Y = 3.04 + 1.04 x - 0.18 x2; coefficient of determination (R2) = 0.57; P (2.89; 4.54) (Figure 1)

Table 2. milk fat and conjugated Linoleic Acids content of dairy milk (%)

Treatment	Milk Fat (%)*	CLA (% Fat)**
a_1b_1	3.23 ± 0.514	0.24 ± 0.02
a_1b_2	4.45 ± 0.504	0.27 ± 0.02
a_1b_3	4.14 ± 0.365	0.32 ± 0.10
a_2b_1	2.85 ± 0.507	0.48 ± 0.05
a_2b_2	4.30 ± 0.711	0.44 ± 0.02
a_2b_3	4.33 ± 0.615	0.62 ± 0.13

Description:* Analysis at Koperasi Pesat Karang Kemiri, Purwokerto (2010)
** Analysis at Kimia Terpadu Laboratory, IPB Bogor (2010)

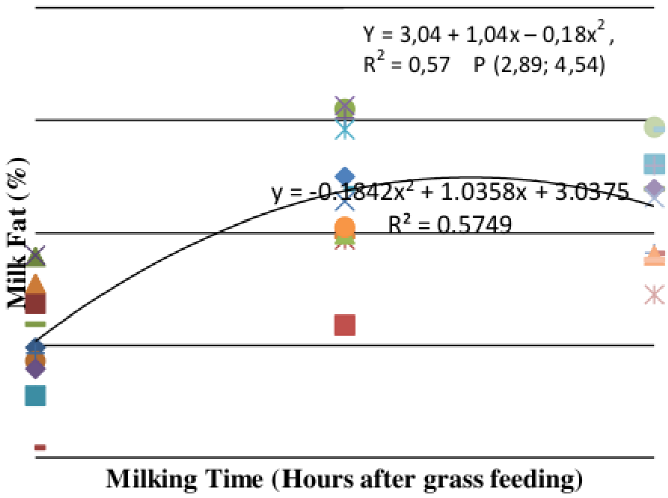

Figure 1. Effect of milking time on milk fat content of dairy milk

Conjugated Linoleic Acids (CLA)

Data from Table 2 showed that Conjugated Linoleic Acids (CLA) ranged from 0.24% to 0.62%. Based on the analysis of variance there was no interaction effect between types of feed and milking time (P> 0.05) but both (the type of feed or milking time) had highly significant effect (P <0.01) on milk CLA content of milk. CLA milk feed treatment (0.51 ± 0.11%) increased by 88.89% when compared with the control feed (0.27 ± 0.07%) (P <0.01). Orthogonal polynomial test showed that milking time gives a linear response (P <0.01) on the CLA content of dairy milk by the equation Y = 0.32 + 0.026 x; coefficient of determination (r2) = 0.10 (Figure 2).

The highest fat content was achieved about three hours after grass feeding. These results indicated that for milk fat synthesis was required within 3 hours after grass feeding, although between 3 and 4 hours after grass feeding had no significant differences of effect (P>0.05). Fat content of dairy products was influenced by fermentative digestion in the rumen. According to Putra (2006) digestion of feed in fermentation, both dry matter (DM) and organic matter (OM), degraded the higher when the fermentation process took place. Based on the research results of Putra (2006) the highest Volatile Fatty Acid (VFA) production at 3 to 4.5 hours of incubation was achieved. VFA at 3 hours of incubation increased as a result of microbial growth that had begun to increase with a more stable ecological condition, thus feed degrading activity also increased. During the digestion process in the rumen, fermented grass produced volatile fatty acids consisting of acetic acid, propionate and butyrate. Based on research done by Vlaeminck *et al*., (2006), acetic acid is the highest VFA (66%), followed by propionate (21%) and the least is butyrate (13%). The proportion of VFA partially was influenced by the type of feed. Feedstuffs containing high crude fiber will produce acetic acid with a percentage more. From table 1, grass is the feed material with a high content of crude fiber, which is 33.58% (Table 1). Acetic acid is a precursor of milk fat, thus after the cows are fed grass, milk fat content will increase, and the more time available for the synthesis of milk fat content is also growing.

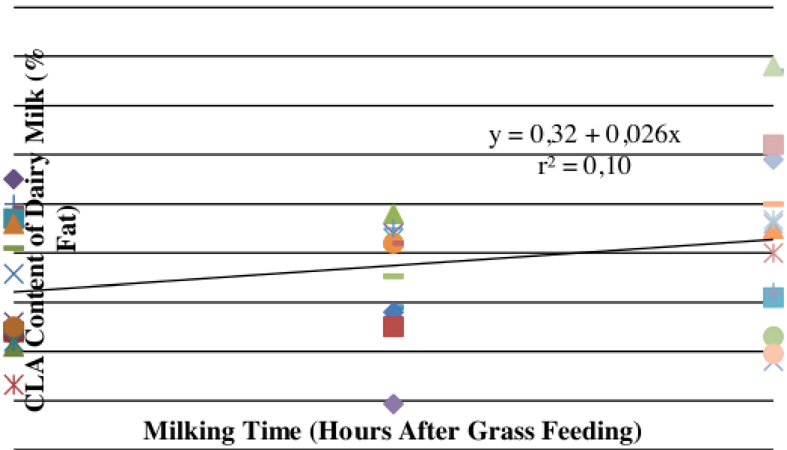

Figure 2. Effect of milking time on CLA content of dairy milk

Dietary treatment that contain fermented rice meal and tapioca byproduct added with 3% soybean oil could increase 88,89% CLA. Soybean oil has a large proportion of C18:2 (51% wt/wt), the precursor of CLA, and has been an effective dietary supplement for increasing the CLA content in Milk (Dhiman *et al*., 2000), (this outlook was supported by results of the analysis made at the Laboratorium of Kimia Terpadu IPB, namely that soybean oil contained 48.16% linoleic acid). Kelly *et al*., (1998) showed that CLA concentration in milk fat could be increased by dietary PUFA supplementation, especially oils rich in linoleic acid. The cis -9, trans-11 CLA isomer derived directly from C18:2 isomerization during ruminal biohydrogenation, and is the major CLA isomer found in animal derived foods (Huang *et al*., 2008). Besides that, the oil also serves as a defaunating agent, so the population of protozoa in the rumen could be decreased. Protozoa are predators of bacteria so that the declining population of protozoa may enhance the growth of bacteria (Muhtarudin and Liman, 2006), including group A bacteria that can hydrogenate linoleic acid and α-linolenic acid in the rumen, as the end product is trans-11 C18: 1. Group A bacteria are part of the Butyrivibrio fibrisolvens group, an ill-defined taxon that includes the genera Butyrivibrio and Pseudobutyrivibrio (Kim *et al*., 2008). In the rumen, linoleic acid that had undergone an isomeration become Cis-9, trans-11 CLA, and was then hydrogenated by bacteria group A into the trans-11 C18: 1. The hydrogenation of Cis-9, trans-11 CLA to trans-11 C18: 1 is faster than the hydrogenation of trans-11 C18: 1 to stearic acid (made by the group B bacteria), thus the accumulation of trans-11 C18: 1 in the rumen made it more available to be absorbed into tissues, including the mammary tissue. Lactation cows have the ability to synthesize the endogenous CLA in the mammary tissue, namely de-saturated trans-11 C18: 1 experienced by the Δ9-desaturase to cis-9, trans-11 C18: 2 (CLA) (Bauman *et al*., 1999) which is the main CLA isomer in ruminant fat (75-90% of total CLA) and mainly derived from endogenous synthesis via Δ9-desaturase in the mammary gland (Lock and Bauman, 2004). Increased rumen production of trans-11 C18:1 resulted in increased synthesis of cis-9, trans- 11C18:2 catalyzed by Δ9- de-saturated in the tissues of ruminants (Whitlock *et al*., 2006).

Conclusion

The concentrate containing 30% rice bran + 20% tapioca byproduct which is fermented with *Saccharomyces cerevisiae* + 3% soybean oil was the best feed formula, and milking four hours after grass feeding was the best milking time.

Acknowledgements

We would like to express a huge gratitude to Project I-MHERE Batch III Sub-Component B.1. of the Jenderal Soedirman University in Purwokerto who have funded this research.

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