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Research Article

EVALUATION OF DRY MATTER DIGESTIBILITY AND ORGANIC MATTER OF IN VITRO UNSATURATED FATTY ACID BASED RATION OF RUMINANT

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A B S T R A C T

This study aims to evaluate digestibility in feed ruminants based on unsaturated fatty acids *in vitro*. This research used cow rumen fluid, forage substrates and concentrates with a ratio of 60:40. The measured variables were digestibility values including: dry matter digestibility, and organic matter *in vitro*. The experimental design used was Completely Randomized Design consisting of 4 treatments consisting of: (R0) feed without soybean oil, (R1): feed with 2.5% soybean oil; (R2): feed with 5% soybean oil; (R3): feed with soybean oil 7.5% in 100% dry matter (BK). While for experiment using *cakalang* fish oil was consisted of (R0) feed without fish oil, (R1): feed with fish oil 2.5%; (R2): feed with fish oil 5%; (R3): feed with fish oil 7.5% in 100% dry matter (BK). Each treatment was performed four replications. The results showed that both soybean oil and *cakalang* fish oil had significant effect (P<0.05) on the digestibility of dry matter and organic matter. The conclusion in this research is as many as 2.5% of both soybean oil and fish oil could be used in ruminant diets because it is able to maintain good digestibility of dry matter and organic matter. Digestibility value of 2.5% use of soybean oil and *cakalang* fish oil gives the digestibility value such as without adding oil.

INTRODUCTION

Feed is one of important factors that contribute to achieve optimal production of a livestock. Feed should be put into top priority of livestock management because 60-70% of production costs are determined by the main feed. Feed for ruminant could be raw forage. As we know that feeding fresh forage is limited to meet the needs of livestock, in addition to the forage is still a major source of methane gas producing on ruminant.

Global issues that always place livestock as the cause of methane emissions in the atmosphere are a challenge for livestock experts. It would be necessary to explore for alternative feed that could suppress emissions of greenhouse gases in the atmosphere at the same time does not interfere with the digestibility of the feed value. Sondakh *et al.* (2015, 2017) mentions that the fat-containing feed may interfere methanogen activity. Disruption of methanogen activity resulted in reduced production of methane gas in the rumen. Feed containing fats other than as a source of energy for animal life, can reduce the production of methane gas, but on the other hand can disrupt microbial life. Based on the facts mention before it could be necessary to design in ruminant feed the use of fat for effective microbial life.

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The use of unsaturated fatty acids in the diet can provide benefits for livestock because these fatty acids can oxidize electrons to reduce methane production in the rumen. Unsaturated fatty acids are also able to maintain the stability of propionic acid in the rumen, so that this acid can be maximized in the chemical process for the determination of meat quality. Use unsaturated fatty acid could be interesting to investigate considering the process of fat metabolism in the rumen occurs through a process of hydrolysis and hydrogenation. The use of fatty acid, such as soybean oil and tuna oil become the focus in this study in order to determine the value of dry matter and *in vitro* organic matter.

MATERIALS AND METHODS

Examination of *in vitro* digestibility conducted at the Laboratory of Biochemistry, Facuty of Animal Science UGM. This study used rumen fluid as a source of microorganisms derived from adult ruminants. The research material consists of soyabean and tuna oil. These oils were obtained through extracts of soya bean and tuna by ether extraction method. Forage feed used is elephant grass and concentrate with a ratio of 60: 40%. Other materials used are *in vitro* testing solution, fermenters, syringe and water bath.

A completely randomized design was used for data analysis. This study used two experiments as follows: experiment using soybean oil and experiment using fish oil. The treatment of these two experiments was (R0) feed without soybean oil, (R1): feed with soybean

oil 2.5%; (R2): feed plus 5% soybean oil; (R3): feed plus 7.5% soybean oil in 100% dry matter (DM). Meanwhile, the use of tuna fish oil consists of (R0) without fish oil; (R1): 2.5% of fish oil; (R2): 5% of fish oil; (R3): 7.5% of fish oil in 100% dry matter (BK). Each treatment was performed four replications.

The research variables measured were dry matter and organic matter (AOAC, 2005)

Research Procedure

The in vitro fermentation procedure uses a 2-level modification method according to Tilley and Terry (Van Soest, 1994).

The digestion process of fermentation samples of 2 g (DM) are inserted into the bottle. Then 30 ml of McDougall buffer solution and 15 ml of rumen fluid were added and sealed with rubber. CO_2 gas was flowed to ensure the anaerobic state. Incubation for 72 hours at a temperature of 39 ^{0} C in the incubator was performed. After reaching fermentation, the fermentation was terminated immediately by adding saturated $HgCl_2$.

The hydrolysis digestion procedure. As much as 40 mL of 0.2% pepsin solution was added in 0.1% HCl to the test vial. Incubated anaerobically at a temperature of 39^{0}C for 72 hours was then performed. After that it was centrifuged at 2,500 rpm for 15 minutes to separate the supernatant from the sediment. The remainder of the undigested sample was separated by filtration using Whatman no 41 paper with the help of a vacuum pump. The rest of the filtrating was then been oven at a temperature of 60^{0}C for 24 hours. Final step was weighing and analysis of dry matter, organic matter.
The digestibility is calculated as follows:

$$\text{KN}\ (\%) = \frac{(\text{A} - (\text{B} - \text{C}))}{\text{A}}\ x\ 100\%$$

where:
KN = Nutrient Digestibility (%)
A = Nutrient before incubation
B = Nutrient left over after *in vitro* (g)
C = Blank: residual material after *in vitro* without the sample (g)

Data analysis

Data obtained from all st variables either supplementation of soy oil and fish oil were analyzed using a completely randomized design with 4 treatments and 4 replications. If there is a difference between treatment then contras orthogonal test will be performed.

RESULTS

The average of digestibility of dry matter and organic mater using supplement of soybean and fish oil can be seen in Table 1 and Table 2. Dry matter digestibility in Table 1 show that there is a significant difference between treatments. In the sense that the use of soybean oil as a substrate supplementation on forage and conc ate effect on the dry matter digestibility. There is a decrease in dry matter digestibility when feed substrate is added with 7.5% soybean oil in 100% dry matter. While the addition of up to 5% has not significant difference. The addition of 7.5% soybean oil on the substrate forage and concentrate (60:40) led to a decrease in the digestibility of 17.28% compared with the substrate feed without soybean oil.

Table 1 Average of dry matter and organic matter digestibility that given soybean oil in ruminal fermentation *in vitro*

Digestibility	Treatment			
	R0	R1	R2	R3
Dry matter (%)	71,49^{b}	68.79^{ab}	66.57^{ab}	59.13^{a}
Organic matter (%)	57.6^{c}	54.18^{bc}	53.64^{b}	47.43^{a}

Note:
Superscript at the same rows shows signifcantly (P<0,05)
(R0) feed without soybean oil, (R1): feed with soybean oil 2.5%; (R2): feed with soybean oil 5%; (R3): feed with soybean oil 7.5%.

Table 2 Average of dry matter and organic matter digestibility that given *cakalang* fish oil in ruminal fermentation *in vitro*

Digestibility	Treatment			
	R0	R1	R2	R3
Dry matter (%)	70.37^{c}	63.25^{abc}	61.13^{ab}	57.49^{a}
Organic matter (%)	61.96^{c}	56.67^{abc}	56.21^{ab}	49.27^{a}

Note:
Superscript at the same rows shows signifcantly (P<0,05)
(R0) feed without *cakalang* fish oil, (R1): feed with *cakalang* fish oil 2.5%; (R2): feed with *cakalang* fish oil 5%; (R3): feed with *cakalang* fish oil 7.5%.

The addition of soybean oil from 2.5% to 5% may decrease the value of dry matter, although in statistical analysis show that there is no significant difference. Similarly, with the digestibility of organ materials, the addition of soybean oil has significant effect on the dig ibility of organic matter. The addition of soybean o causes a decrease in the digestibility of organic matter. The decrease in the digestibility of organic matter occurs. When the substrate is added with 5% soybean oil, a decrease of 6.8% compared with no soybean oil will occur. When the soybean oil increased to 7.5%, the value of organic matter digestibility decreased by 17.88%.

The dry matter digestibility value in Table 2 shows that the use of fish oil on forage substrates and concentrates has a significant effect. In the sense that the use of fish oil as supplementation on feed substrate forage and ncentrates will effect on dry matter digestibility. There was a decrease in dry matter digestibility when the feed substrate was added with 5% and 7.5% fish oil. While an increase up to 2.5% has no significant difference. The addition of 5% fish oil to forage substrates and concentrates (60:40) caused a decrease in digestibility by 13.13%. If the fish oil is increased up to 7.5%, the dry matter decreases much as 18.30% compared with no addition of fish oil. The addition of fish oil causes a decrease also to the digestibility of organic matter. Lowering an organic matter digestibility values occurs when the substrate added with tuna fish oil as many as 5% - 13 %. While the addition of 2.5% of fish oil did not cause the decrease in the digestibility of organic matter. Addition of 5% of tuna oil causes organic matter digestibility decreased about 9.28%, then if tuna oil is increased up to 7.5% causing decrease of organic material digestibility up to 12.69%.

DISCUSSION

Effects of decreased feed digestibility

The digestibility of dry matter in ruminants indicates high nutrients digested by microbes and digestive enzymes in the rumen. According Riswandi *et al.,* (2015), increasing in a percentage of dry matter digestibility of feed ingredients will increase the quality of the feed material. High-value digestibility reflects the size of a particular nutrient

contribution to livestock, while feeds with low digestibility indicate that the feed is less able to supply nutrients for basic living or for livestock production purposes (Yusmadi, 2008). Organic matter digestibility showed amounts of nutrients like lipids, carbohydrates, and proteins that can be digested by livestock. Digestibility of nutrients in ruminants is not dependent on the protein quality of the feed but the crude fiber content and activity of rumen microorganisms, mainly celulolytic bacteria. The decrease in digestibility shown in the administration of 5% - 7% both soybean oil and fish oil is due to the influence of the use of oil. Oil is a substrate that can wrap food substances, so the food cannot be digested by microbial rumen (Sobhy and Samir, 2010). The decline rumen microbial caused by oil has been studied by Mao *et al.* (2010). Rumen bacteria, especially methanogenesis bacterial undergoes reduction of substrate containing vegetable oil. Mapato *et al.* (2012) and Lunsin *et al.* (2012) suggest that supplementation of sunflower oil and rice bran oil as much as 60 g / kg of dry matter consumption causes decreased dry matter, organic matter and NDF in lactating dairy cows. This was confirmed by Wanapat *et al.* (2011) that sunflower oil as a source of unsaturated fatty acids can reduce the consumption of dry matter. However, according to Toral *et al.* (2009) the changes that occur in rumen fermentation depend on the given basal feed. As stated also by Homem *et al.* (2010) that the digestibility depends on the source of fat supplemented to the feed. Another results was purposed by Pamquist and Griinari (2006) who mention that some fatty acids are toxic in the rumen microbial biohydrogenation process, because it contains medium chain fatty acids and long-chain polyunsaturated fatty acids. That why there is a decrease of 7.5% digestibility both by fish oil and soybean oil. The content of this fatty acid, medium chain fatty acids and polyunsaturated fatty acids it is mostly found in fish oil and soybean oil.

CONCLUSION

As many as 2.5% of both soy bean oil and fish oil could be used in ruminant diets because it is able to maintain good digestibility of dry matter and organic matter. Digestibility value of 2.5% use of soybean oil and *cakalang* fish oil gives the digestibility value such as without adding oil.

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