PJN

ISSN 1680-5194

PAKISTAN JOURNAL OF NUTRITION

ANSInet

308 Lasani Town, Sargodha Road, Faisalabad - Pakistan

Mob: +92 300 3008585, Fax: +92 41 8815544

E-mail: editorpjn@gmail.com

Pakistan Journal of Nutrition 10 (7): 625-630, 2011
ISSN 1680-5194

The Effects of Using Variety of by Products Palm Industry on Ration Towards the Characteristics Rumen Fluid of Ettawa Goat According to *In vitro* Analysis

Arief[1], Novirman Jamarun[1] and M. Winugroho[2]
[1]Faculty of Animal Science, Andalas University, Padang, West Sumatera, Indonesia
[2]Center for Animal Research, Ciawi, Bogor, Indonesia

Abstract: Palm manufacture industry has produced many kinds of by products which can potentially be a feedstuffs such as Palm Kernel Cake (PKC), Palm Oil Sludge (POS) and Palm Press Fiber (PPF). From the whole result of production, only 40% of products from palm which can be used for human's consumption, while 60% is the by product which can potentially be feedstuffs. In 2008, the area of palm plantation in Indonesia reaches 7.3 millions of Ha with the production reaches 19.2 millions of ton and on 2009, the area of palm reaches 7.9 million of Ha with the production of 19.4 millions of ton (PTPN, 2009). From those productions above, the potential of by product of palm manufacture industry which will be produced and used as feedstuffs is really big. The aim of the research was conducted rumen's fluid characteristics of ettawa goat given ration by paoducts of palm industry. The matter used on this experiment was by products of palm industry which consists of PKC, POS and PPF which can be formulated in a complete ration with different composition in forage 60% and 40% concentrate. Substrate of complete ration which will be evaluated in *in-vitro* analysis, was made in the form of flour and the making of medium liquid which consist of macro mineral, micro mineral, resazurin, buffer and Reducing Agent (RA) was pointed to the method of Theodorou and Brooks (1990). The measured variable is the characteristic of rumen's fluid including total production of gas, pH, rumen's bacteria, NH_3 and VFA throughout 48 h of incubation. The result of experiment showed that the use of by products of palm industry formulated in a complete ration produce the characteristic of rumen's liquid which still complete the standard, concerned from pH (6.44), total gas production (97.00 ml), the amount of bacteria colony (5.02×10^9 cfu/ml), NH_3 (74 mg/l) and VFA (81.68 mM). From the whole parameter above, it can be concluded that the by product of palm industry can be used as feedstuffs and have a big potential to replace substance of conventional ration which is normally used.

Key words: By products, palm industry, rumen fluid, ettawa goat, *in-vitro* analysis

INTRODUCTION

One of new resource which is very potential to be feedstuffs is by product of palm manufacture industry which consist of Palm Kernel Cake (PKC), Palm Oil Sludge (POS) and Palm Fiber (PF) (Kamaruddin, 2006; Mathius, 2008). The total amount of by product can reach 60% from total production/Ha. If 1 Ha of palm produced 16 tons of bunch press and every ton of TBS (bunches) produced 294 kg of sludge, 35 kg of palm kernel cake and 180 kg of fiber, then the total production of the three by products for each Ha is 1.132 kg of POS, 514 kg of palm kernel cake and 2.681 kg of PPF.

These three by products produced by palm industry above is qualified as ruminant feedstuffs because the three substances meet the criteria as non conventional feedstuffs, such as having a lot of availability, having no compete with human's need and lower price. Besides that, the three side products of palm manufacture industry will keep increasing as well as the increasing of the wide area for palm plantation (20%/year, Luthan, 2008) so the effort was needed to continually optimize that resource and give additional amount for palm plantation and the development of animal husbandry.

Indonesia was the second biggest country after Malaysia for the area of palm plantation. With the wide area of palm plantation in Indonesia which reached 6.07 million Ha and palm production of 16 million tons (Luthan, 2008), so the amount of by product of palm manufacture which will be produced, was the big source for feedstuffs and will be the biggest problem if it wasn't handled properly. One of the way to solve the problem is by benefitted the livestock, especially the ruminant livestock as a life machine to produce a quality side products.

From these three by products of palm manufacture industry above, palm kernel cake had the highest quality of by product and would be feedstuffs because of its high biologic value. The high biologic value was marked by digestibility energy and dry matter which nearly the same with coconut meal, soybean meal and peanut meal and even higher than wheat brain and fish meal.

PKC had been used widely on livestock diets as protein replacement to replace conventional protein resource on

Corresponding Author: Arief, Faculty of Animal Science, Andalas University, Padang, West Sumatera, Indonesia

the different level (Onwudike, 1996). Loh *et al.* (2002) reported that the give of 15-25% BIS gives no significant difference against growth, consumption and livestock's weight. Despite the content of protein in palm kernel cake which is lower than another protein source of diets, the quality of protein was relatively better. Some researcher found out that palm kernel cake can be used almost 30% in ration of dairy cattle lactation which can fulfill the need of energy and the protein of cattle (Orunmuyi *et al.*, 2006).

The other by products of palm industry (PPF and POS) had also been researched quite a lot as rations. POS had a high content fiber protein and can be used as livestock ration but its low content of energy reflected its low quality other than PKC. As well as PPF, where its utilization as feedstuffs was constrained by the low fiber protein content and the high content of rough fiber, especially the component of lignin and cellulose fiber.

The research about the utilization of these three by products of palm industry, formulated in a complete ration of feedstuffs, was still limited. The research was more done by utilized that by product singly in rations. Whereas these three products were a single unit of by product derived from palm factory which can be utilized simultaneously by integrating the livestock.

Despite that it can prevent from bad effect of some by products which still got a nature of waste that can bother the environment around the factory, this research will also gives a double benefits. First is the integration between animal and palm industry will be built by utilized the by product derived from palm factory and second is the economical profit which can be shared by both side (factory and breeder around the factory).

MATERIALS AND METHODS

On this research, some by products of palm manufacture industry (Palm Kernel Cake - PKC, Palm Press Fiber - PPF and Palm Oil Sludge - POS) were formulated in a complete ration which afterwards will be continued by *in-vitro* analysis. The aim of this research was to get the best formulation from the complete ration which consist of many by products of palm industry determined by digestion and rumen's liquid characteristic.

This research used Completely Randomized Design (CRD) with the treatment of 4 kinds of ration formulations consist of palm manufacture by products with 5 replications. The composition and the content of nutrient as feedstuff for ration's composer and treatment ration can be seen in Table 1.

The data obtained was analyzed using analysis of varian (anava) followed by Steel and Torre (1991), whereas the different between each treatment was tested with Duncan's Multiple Range Test (DMRT).

The measured parameter was rumen's liquid characteristic including the production of total gas, pH, bacteria colony, VFA content on rumen's liquid measured by gas chromatography and vapour distillation, NH_3 content measured by using Conway method and ration's digestibility with NH_3.

Table 1: The composition and nutrient content of *in-vitro* trial ration (%)

	Formulation of treatment ration (%)			
Feedstuffs	A	B	C	D
Rhougage	60.00	60.00	60.00	60.00
PKC	10.00	12.00	14.00	16.00
POS	8.00	6.00	4.00	2.00
PPF	2.00	2.00	2.00	2.00
Polard	2.00	2.00	2.00	2.00
Coconut meal	9.20	9.20	9.20	9.20
Corn	8.00	8.00	8.00	8.00
Urea	0.40	0.40	0.40	0.40
Salt	0.20	0.20	0.20	0.20
Mineral	0.20	0.20	0.20	0.20
Percent (%)	100.00	100.00	100.00	100.00
Nutrition content				
Dry matter (%)	48.28	48.31	48.34	48.37
Crude protein (%)	11.68	11.79	11.86	12.00
Crude fiber (%)	26.66	26.98	27.31	27.64
Crude fat (%)	4.33	4.01	3.69	3.37
TDN (%)	67.10	67.48	67.89	68.29

The procedure of *in-vitro* analysis: The rumen's liquid used was from the Ettawa goat where the research was conducted in Ruminant Laboratory Center for Animal Research (BPT), Ciawi Bogor. The sample research was 4 complete ration formulations which made in form of flour, then as much as 1 gram was taken and put into *in-vitro* bottles. Inside the *in-vitro* bottles, there will be added medium solution consist of micro mineral, macro mineral, resazurin, buffer, reducing agent and distillation water, which will be given CO_2 after.

The *in-vitro* bottles then will be kept on coolant room during 24 h, warmed in water bath about ±15 min and put into rumen's liquid of Ettawa goat as much as 10 ml, added with the reduction liquid and kept in water bath during 48 h. The measuring of gas production was done every 6 h and after the incubation of 48 h, the measuring was done towards the research parameter such as pH, total bacteria colony, NH_3 content and VFA.

The pH measuring was done by using pH-meter which had been calibrated using standard pH and the count of the total bacteria colony was done by using the procedure of Oghimoto and Imai (1980) where the calculation of NH_3 content was done by using the method of Conway and Chromatography Gas (CG) to measure VFA content.

RESULTS

Total gas production: The total production of gas from each formulation of treatment ration is presented in Table 2.

From Table 2, it can be observed that the total gas production during 48 h of incubation was revolved between 93.30-99.90 ml. The average of gas production in first 6th h incubation is 32.92% from total gas production.

Table 2: Total gas production from each formulation of treatment ration (ml)

Treatment ration	Gas production: First 6th h incubation: Total gas	Percentage (%)	Total gas production (48 h)
A	29.60	27.17	95.70
B	38.87	38.91	99.90
C	29.40	31.51	93.30
D	33.07	34.09	97.00

Table 3: The acidity (pH) from each ration's formulation treatment

Replication	Treatment ration: A	B	C	D
1	6.60	6.44	6.42	6.44
2	6.60	6.44	6.49	6.45
3	6.72	6.46	6.44	6.42
4	6.60	6.50	6.47	6.45
5	6.62	6.42	6.42	6.46
Average	6.62	6.45	6.45	6.44

Table 4: The influence of treatment to rumen's bacteria ($x10^9$ cfu/ml)

Replication	Rumen's bacteria of treatment ration ($x10^9$ cfu/ml): A	B	C	D
1	1.60	3.20	4.70	5.10
2	2.90	2.80	4.10	5.10
3	0.80	5.30	4.50	5.00
4	2.70	2.60	4.30	5.90
5	2.20	3.30	4.60	4.00
Average	2.04^c	3.64^b	4.44^{ab}	5.02^a

Difference superscript showed significant difference ($p<0.05$)

pH of rumen's Liquid: The acidity (pH) of rumen's liquid from each ration's formulation is presented in Table 3. The statistic analysis showed that ration's formulation didn't produce significant difference ($p>0.05$) towards pH of rumen's liquid and the pH produced was in normal turn. The average of pH rumen's fluid is about 6.44-6.62 (Table 3).

Rumen's bacteria: The result of research about rumen's bacteria is presented in Table 4.
The result of statistic analysis showed that the amount of bacteria colony on ration's treatment showed significant difference ($p<0.05$) and D treatment got higher amount of bacteria colony compared to other treatments. The average amount of rumen's bacteria colony from the research result was about 2.04-5.02 x 10^9 cfu/ml which was a general amount of rumen's microbe obtained from ruminant livestock (MC Donald *et al*., 1990).

NH_3-N: The result of the research about the availability of N-NH_3 on rumen's liquid is presented in Table 5.

Table 5: The availability of N-NH_3 on rumen's liquid from each formulation of ration's treatment

Treatment ration	Availability of N-NH_3 rumen fluid (mg/L)
A	66.22
B	68.08
C	68.92
D	74.00

Table 6: The concentration of VFA of rumen's liquid on research's result

Treatment ration	VFA concentration (mM)
A	51.83^c
B	59.79^{bc}
C	70.36^{ab}
D	81.68^a

Difference superscript showed significant difference ($p<0.05$)

Table 7: Ration's digestibility and NH_3

Treatment ration	N-NH_3 concentration (mM)	Dry matter digestibility (%)
A	66.22	42.30
B	68.08	56.90
C	68.92	54.00
D	74.00	62.30

The statistic analysis showed that ration's formulation didn't produce significant difference ($p>0.05$) towards NH_3-N of rumen's liquid and the NH_3-N produced was in normal turn. The average of NH_3-N rumen's fluid about 66.22-74.00 mg/L (Table 5).

Volatile Fatty Acid (VFA): The concentration of Volatile Fatty Acid (VFA) on rumen's liquid is presented Table 6. The statistic analysis showed that the treatment gave significant difference ($p<0.05$) towards the VFA concentration on rumen's liquid where D treatment was real ($p>0.05$) higher than other treatments.

Dry matter digestibility and NH_3: The ration's digestibility and its relation to NH_3 concentration on rumen's liquid is presented in Table 7.
From Table 7, it can be concluded that concentration of NH_3 on rumen's liquid gave a positive influence towards the dry matter's digestibility and organic matter of treatment rations.

DISCUSSION

Total gas production: This result (Table 1) was not too different from one obtained by Suharyono and Widiawati (2007) who got the total production of gas as much as 101 ml by given multinutrient supplement ration towards rumen's ecosystem. This means that the ration given consists of high enough protein and low rough fiber. The high content of rough fiber usually produced a lot of gas so that much ration energy got castaway (unutilized) especially NH_4 gas which caused green house effect related to global warming. Added by Baker (1999) that the gas produced throughout the fermentation was the image of how big the ration's energy that can't be utilized was and disappeared through the air. Between all of the ruminant livestock, cow produced almost 73% CH_4 during the fermentation of ration process and was the highest compared to other kinds of ruminant livestock (US Environment Protection Agency, 1994).

Reversely, some researcher obtained a high gas production as much as 167 ml (Suharyono and Widiawati, 2007), through the give of rice straw and concentrate.

pH of rumen's liquid: Table 3 shows that pH rumen's fluid was in normal turn. This means that the rumen's microbe activity in the ration's digestion process wasn't disturbed and the rumen's microbe can act well. The ration's digestion process would be disturbed if the pH of rumen's liquid was below 6 (Chanjula *et al*., 2004). Furthermore, it was explained that with pH 5 and 6, the rumen's microbe activity to digest ration would be blocked, even stopped according to Orskov and Ryle (1990) that pH less than 6.2 would block rumen's microbe growth significantly.

The pH value obtained from the research above showed that rumen's pH was on the good category for rumen's microbe activity where the average of normal pH for rumen was revolve around 6-7 (France and Siddon, 1993) whereas the ideal pH for fiber digestion was 6.4-6.8. The appropriate pH could help the bacteria colonization in plant's cell wall and push the activity of cellulose bacteria.

These results is not different with research of Sugoro *et al*. (2005) which obtained pH rumen fluid 6.35-6.56 in *in-vitro* analysis and Uhl *et al*. (2006) which obtained pH rumen fluid about 6.15-6.85 while the livestock fed with catalytic supplement's ration.

Rumen's bacteria: The result of statistic analysis on rumen's bacteria (Table 4) showed that the amount of bacteria colony on ration's treatment showed significant difference ($p<0.05$) and D treatment got higher amount of bacteria colony compared to other treatments. The average amount of rumen's bacteria colony from the research result was about 2.04-5.02 x 10^9 cfu/ml which was a general amount of rumen's microbe obtained from ruminant livestock (MC Donald *et al*., 1995). The performance of this high amount for rumen's microbe was related to ideal condition for rumen's pH for the activity of cellulolytic bacteria in rumen.

The performance of this high amount of microbe also related to the availability of NH_3 rumen's liquid which was above 50 mg/l so that it won't be the barrier factor in the growth of rumen's microbe. The normal amount of rumen's microbe was needed so that ration's digestion would run normally (Preston and Leng, 1987; Mc Donald *et al*., 1995). Nasrullah *et al*. (2002) added that enough content of protein in ration was needed to support the appropriate supply of ammonia for the life of rumen's microbe. The low protein in ration could lead to insufficient availability of ammonia for the growth and development of rumen's microbe.

NH_3-N: Relatively, it can be concluded that the availability of N-NH_3 on rumen's liquid (Table 5) was about 66.22-74.00 mM. which was the normal condition of NH_3 availability on rumen's liquid and it was above the minimum concentration of NH_3 needed for growth and optimum bacteria's activity that was about 50 mg/L (Preston and Lang, 1987). Furthermore, it will be explained that rhe maximum limit of NH_3 concentration for normal growth of microbe was as much as 150 mg/L. More amount of NH_3 on rumen's liquid showed that the protein ration was easily degradated in rumen. The increase of ammonia's availability would give nitrogen's balance and good energy needed by rumen's microbe to grow.

According to Erwanto *et al*. (1993), NH_3 concentration on rumen's liquid also determined the efficiency of microbe's protein synthesis which finally would influence the result of organic ration's fermentation formed of Volatile Fatty Acid (VFA) which was the main energy source for ruminant livestock. Also added by Winugroho and Maryati (1999) that the concentration of NH_3 beyond 12 mM, the conversion process of NH_3 to N would be disturbed and if NH_3 was lower than 4 MM (low condition of ration's protein), the degradation process would also be disturbed. Whereas according to Satter and Slytter (1994) ammonia concentration needed to optimum growth for rumen's microbe in *in-vitro* analysis was 3.57 mM. If NH_3 concentration lower than 1.41-2.83, this condition will not support optimum growth for rumen's microbe (Haryanto *et al*., 2005).

Volatile Fatty Acid (VFA): The statistic analysis showed that the treatment gave significant difference ($p<0.05$) towards the VFA concentration on rumen's liquid where D treatment was real ($p>0.05$) higher than other treatments. The high concentration of VFA on D treatment was caused by the increase of fermentation due to the increase of rumen's microbe. This result obtained was also in line with the increase of NH_3 availability on rumen's liquid so that microbe could grow well and work, with the final result of VFA's availability which was the energy source for microbe.

The average concentration of VFA according to research result for those four treatment was 51.83-81.68 mM. According to Mc Donald *et al*. (2002) that the normal amount of VFA concentration on optimal rumen's liquid for microbe's growth was 80-160 Mm, while according to Preston and Leng (1987) that the minimum amount of VFA on rumen's liquid for microbial survival was 50 mM.

Dry matter digestibility and NH_3: The increasing concentration of NH_3 on rumen's liquid gave a positive influence towards the dry matter's digestibility and organic matter of treatment rations. This was related to the fulfillment of NH_3 needs for protein synthesis by microbe in rumen. Tillam *et al.* (1991) said that ration with enough content of protein would supply nitrogen like NH_3 microorganism and energy source which enough for rumen's microbe that will help the organic digestion for normal running. Soeharyono *et al.* (2007) obtained the same result with this research through the use fo ration supplement UMMB. Also added that the increase of VFA and ammonia would increase the digestibility of dry matter and organic matter on ration.

Sutardi (1980) found that VFA produced by rumen's fluid on ration added with Urea Multinutrient Molasses Block (UMMB) and Supplement Nutrient Ration (SNR) produced VFA rumen's fluid about 44.41-92.33 which approve that the concentration was still below the optimum average for protein's microbe synthesis. This also showed low degradable protein and crude fiber ration because the ration contains high fiber. High concentration of VFA in ration D is in line with high microbe content in D ration. This also showed the more amount of rumen's micrpbe, the higher amount of VFA production will be.

Conclusion: From the description above, it can be concluded that,

- The use of by products of palm industry in complete ration gives no influence on rumen's liquid characteristic of Ettawa goat according to *in-vitro* analysis
- The formulation of complete ration with the percentage of the use of more palm kernel cake gives a better result reviewed from gas production and rumen's liquid characteristic.

ACKNOWLEDGEMENT

The authors are very gratefull to Directorate General of Higher Education Department of National Education Republic of Indonesia that funded this experiment by Hibah Kompetitif Penelitian Strategis Nasional Project, year 2010.

REFFERENCES

Ahafemule, F., O. Dan and J.A. Ibeawuchi, 2009. Variation in weekly milk yeld and composition of muturu, N'dama and white fulani cows in early lactation. J. Anim. Vet. Adv., 7: 469-474.

Baker, S.K., 1999. Rumen methanogens and inhibition of methanogenesis. Aust. J. Agric. Res., 50: 1293-1298.

Chanjula, P., M. Wanapat, C. Wackhrapakorn and P. Rowlinson, 2004. Effecs synchronizing starch sources and protein (NPN) in rumen feed intake, rumen microbial fermentation, nutrient utilization and performance of lactating dairy cows. Departement of Technology, Prince of Songkla University Thailand. Asean Aust. J. Anim. Sci., 17: 1400-1410.

Erwanto, T. Sutardi, D. Sastradipraja and M.A. Nur, 1993. Effects of ammoniatic zeolite on metabolic parameter of rumen microbes. J. Trop. Agric., 5: 5-6.

France, J. and R.C. Siddon, 1993. Volatil Fatty Acid Production in Forbers France, J editor Quantitative Aspect, Ruminant Digestion and Metabolism, CAB Int, Wellingford, UK.

Haryanto, B., Supriyati, A. Thalib and S.N. Jarmani, 2005. Peningkatan nilai hayati jerami padi melalui bio proses fermentatatif dan penambahan zinc organic. Proseding Seminar Nasional Peternakan dan Veteriner Bogor, 12-13 September 2005. Puslitbangnak Peternakan, Bogor.

Kamaruddin, A., 2006. Masalah dan Solusi Pakan ternak Ruminansia di Indonesia. Pidato Pengukuhan Sebagai guru Besar Tetap dalam ilmu Nutrisi dan Makanan Ternak Fakultas peternakan Universitas Andalas padang.

Loh, T.C., H.L. Foo, B.K. Tan and Z.A. Zealand, 2002. Effect of palm kernel cake on performance and blood lipid in rats. Asian-Austr. J. Anim. Sci., 15: 1165-1169.

Luthan, F., 2008. Meningkatkan Kesejahteraan dan Ketahanan Pangan Hewani Nasional dengan Akselerasi Pengembangan Sapi Potong. Orasi Ilmiah yang Disampaikan dalam rangka Lustrum IX Fakultas Peternakan Universitas Andalas Padang, Oktober 2008.

Mathius, I.W., 2003. Perkebunan kelapa sawit dapat menjadi basis pengembangan sapi potong. Warta penelitian dan pengembangan pertania, 25: 1-4.

Mathius, I.W., D. Sitompul, B.P. Manurung and Azmi, 2004. Produk samping tanaman dan pengolahan buah kelapa sawit sebagai bahan dasar pakan komplit untuk sapi. Pros. Lokakarya Nasional Sistem Integrasi Kelapa sawit dan Sapi. Departemen Pertanian. Propinsi Bengkulu.

Mathius, I.W., 2008. Pengembangan sapi potong berbasis industri kelapa sawit. Pengembangan inovasi pertania, 1: 206-224, Pusat Penelitian dan Pengembangan Peternakan, Bogor.

Mc Donald, M. P., R. A. Edward and J. F. D. Greenhalgh. 1990. Animal Nutrition, 4th ed, Longman Inc, New York.

Mc Donald, P., R.A. Edward and J.F.D. Greenhalge, 1995. Animal Nutrition, 5th Edn., Longman Group Ltd, London.

Mc Donald, P., R.A. Edward, J.F.D. Greenhalge and C.A. Morgan, 2002. Animal Nutrition, 6th Edn., Longman Science and Tekhnology, New York.

Nasrullah, M., Nilmi, R. Akashi and O. Kawanuna, 2002. Nutritive evaluation of forage plant grown in South Sulawesi, Indonesia. J. Anim. Sci., 16: 693-701.

Oghimoto, K. and S. Imai, 1980. Atlas of Rumen Microbiology. Japan Scientific Societies Press, Tokio.

Onwudike, O.C., 1996. Palm kernel meal as a feed of poultry I. Composition of palm kernel meal and acailability of its amino acid to chick. Anim. Feed Sci. Technol., 16: 179-186.

Orskov, E.R. and M. Ryle, 1990. Energy Nutrition in Ruminants. Elsevier Applied Science. London.

Orunmuyi, M.G.S., Bawa, F.D. Adeyinka, O.M. Daudu and I.A. Adeyinka, 2006. Effect of Graded Levels of Palm Kernel Cake on Performance of Grower Rabbit. National Animal procuction, Research Institut, Ahmadu Bello University Shika, Zaria, Nigeria.

Preston, T.R. and R.A. Leng, 1987. Matching Ruminant Production System with Available Resources in the Tropics. Penambul Books, Armidale.

PT Perkebunan Nasional (PTPN), 2009. Luas Perkebunan Sawit Nasional. Http://www.datacon.co.id. Diakses 2 Maret 2009 jam 14.15 WIB.

Rossi, E. and N. Jamarun, 1997. Pengaruh penggunaan serat sawit dan bungkil inti sawit dalam ransum terhadap daya cerna bahan kering, protein kasar dan retensi nitrogen domba lokal. Jurnal Peternakan dan Lingkungan Fakultas Peternakan Universitas Andalas Padang.

Satter, L.D. and L.L. Slytter, 1994. Effects of ammonia concentration on rumen microbial protein production *in vitro*. Br. J. Nutr., 22: 199-208.

Sinurat, A.P., T. Purwadaria, I.W. Mathius, D.M. Sitompul and B.P. Manurung, 2004. Integrasi Sapi-Sawit, Usaha Pemenuhan Gizi Sapi dari Produk Samping Kelapa Sawit. Proceeding Seminar Integration Plan-Animal System. Center for Animal Research Agriculture Technology Study Board Bali dan Casren.

Suharyono and Y. Widiawati, 2007. ekosistim rumen sapi PO yang diberi pakan Supplemen Multinutrien (SPM) dalam Prosiding Seminar Nasional Teknologi Peternakan dan Veteriner, Bogor, 2007.

Soeharyono, Arni, W.H. Sasongko and L. Andini, 2007. Substitusi Hijauan Sorgum dengan Supplemen Pakan UMMB Tekhnologi Hsil Fermentasi secara *In-vitro*. Proseeding Seminar Nasional Peternakan dan Veterriner, Bogor, 2007.

Steel, R.G.D. and J.H. Torre, 1991. Principple and procedure of Statistics, a Biometrical Approach. 2nd Edn., Mc Graw Hill Int Books Co. Singapura.

Sugoro, I., I. Gobel and Lelananingtyas, 2005. Pengaruh probiotik khamir terhadap fermentasi dalam cairan rumen secara *in-vitro*. Proseding Seminar Nasional Peternakan dan Veteriner. Bogor 12-13 Sept 2005 Puslitbangnak, Bogor.

Sutardi, T., 1980. Landasan Ilmu Nutrisi Jilid I, Departemen Ilmu Makanan ternak Fakultas Peternakan Institut Pertanian Bogor.

Tillam, A.D., H. Hartadi and S. Reksohadiprojp, 1991. Ilmu Makanan Ternak Umum. Gajahmada University Press, Jokjakarta.

Theodorou, M.K. and A.E. Brooks, 1990. Evaluation of a New Laboratory Procedue for Estimating the Fermentation Kinetics of Tropical Feeds. AFRC Institute for Grassland and Envirounment Research, Plas Gogerdan, Aberyswyth, Dyfed, Australian, December 5-21.

Uhl, H.T., A. Parakkasi and B. Haryanto, 2006. Pengaruh supplementasi katalitik terhadap karakteristik dan populasi mikroba rumen domba. Media Peternakan, Jurnal Ilmu Pengetahuan Peternakan dan Tekhnologi Peternakan, 29: 20-26.

US EPA (Envorounment Protection Agency), 1994. International Antropogenic Methane Emission, Estimate for 1990. EPA 230-R93-010 (Office of Police, Palnning and Evaluation, Washington, DC).

Wan Zahari, M., O.B. Hassan, H.K. Wong and J.B. Liang, 2003. Utilization of frond-based diets for beef cattle production in Malaysia. Asian-Aust. J. Anim Sci., 16: 625-634.

Winugroho, M. and Maryati, 1999. Kecernaan daun kelapa sawit sebagai pakan ternak ruminansia. Laporan APBN 1998/1999. Balai Penelitian Ternak Puslitbang Peternakan Bogor.

Winugroho, M. and Y. Widiawati, 2003. Candida utilis sebagai pengganti Saccharomyces cereviceae pendamping bioplus untuk meningkatkan produktivitas ternak. Pros. Seminar Nasional Petrnakan dan Veteriner Bogor, 29-30 September 2003. Puslitbang Peternakan Bogor.