ISSN 1978-3000

Improved Nutrient Contents of *Durio zibethinus* Murr Peel Powder Fermented with *Pleurotus ostreatus* and Its Addition in PUFA- Concentrate

E. Sulistyowati[1], I. Badarina[1], H. Sucianti[1], R. Hartono[1], and S. Mujiharjo[2]
[1]Animal Science Dept., Fac. of Agriculture, University of Bengkulu
[2]Agriculture Technology Dept., Fac. of Agriculture, University of Bengkulu
Jl. Raya W.R. Supratman, Bengkulu, INDONESIA 38371
Email: ensulistyowati@yahoo.com

ABSTRACT

The objective of this research was to evaluate nutrient contents of *Durio zibethinus* Murr peel powder fermented with *Pleurotus ostreatus* and its addition in PUFA- concentrate. There were two experiments have been conducted. First, fermentation process with experimental design of completely randomized treatments, F2: 2 week fermentation, F4: 4 week fermentation, F6: 6 week fermentation, and F8: 8 week fermentation with 4 replications. Second, incorporate the F8 Durio into PUFA- concentrate. Variables evaluated were in first experiment were in vitro VFA, GE, NDF and ADF. In second experiment were nutrient contents and fatty acid profile. Results showed that Fermentation of *Durio zibethinus* Murr peel powder fermented with *Pleurotus ostreatus* for 4-6 weeks seems to improve NDF, ADF, and GE ; while, fermentation for 8 weeks and incorporated into PUFA-concentrate could improve crude protein, ether extract and NDF, and lower ADF. However, it decreased fatty acid profile of SCFA, MedCFA, LongCFA, MUFA, PUFA, unsaturated, saturated, P/S, and U/S.

Key words: *Durio zibethinus* Murr peel powder, *P. ostreatus*, nutrients, fatty acid

INTRODUCTION

Availability of feed is always as a concern for livestock farming. Durian or *Durio zibethinus* Murr is one of harvested fruits that its byproduct could be utilized as one of feed ingredients for ruminants. Its potency in Indonesia is huge, while, in Bengkulu Province is around 5040.8 ton/year. Its waste of Durio peel is about 60-70% (Suhaidi, 2004), equals to 3276.52 ton/year or Durio peel powder or 491.5 ton/year or 1346.6 kg/day.

This byproduct could be used as potential feed; however, it needs treatment, such as fermentation using white rod fungi (*Pleurotus ostreatus*). This fungi grows well in any high fiber waste product media (Sangadji *et al*., 2008). This white rod fungi that is able to degrade lignin binds much more extensively than others, as it produces extracellular lignolitic ezyme, such as laccases, lignin peroxidases and Mn peroxidases (Hatakka, 1994). This fungi also improved crude protein of fermented Durio in 0 to 8 weeks, from 4.31 to 5.69%, respectively; while crude fiber decreased as reported by Suciyanti *et al*. (2015). Hartono *et al.* (2015) reported that *in vitro* organic matter digestibility (IVOMD) is significantly decreased in 2 weeks to 8 weeks of fermentation, respectively. The same manner is also found in *in*

vitro organic matter digestibility (IVOMD). On the other hand, polyunsaturated fatty acid (PUFA)-concentrate with some supplements that has been developed (Sulistyowati *et al*., 2013, 2014, and 2015) could be used as the base diet for incorporation of *Durio zibethinus* Murr peel powder fermented with *Pleurotus ostreatus.*

MATERIALS AND METHOD

Preparation of PUFA- Concentrate Containing Fermented *Durio zibethinus* Murr Peel Powder

There were three steps in this preparation. First, fermentation of *Durio zibethinus* Murr with *Pleurotus ostreatus* adapted from the mehod of Badarina *et al.* (2014). It was started from cleaning, chopping, drying, grinding, mixing the Durio peel with substrates (rice brand, gips, and CaCO3) in baglog, sterilizing, innoculating with the fungi then incubated for 2, 4, 6, and 8 weeks. Then, it was conducted Proxymate analysis as reported by Suciyanti *et al.* (2015) and analyzed for *in vitro* dry matter digestion (IVDMD) and *in vitro* organic matter digestion (IVOMD) as reported by Hartono *et al.* (2015). The results of fermented Durio showed that there was an increase in protein content from 4.73% to 6.40%; and, decreased fiber content from 47.84 to 45.45% in no fermentation and 8 week fermentation, respectively.

Secondly were *in vitro* total VFA analysis, fiber fraction analysis, and gross energy (GE) determination. Thirdly, based on the results earlier, which was part of this reasearch, it was decided to incorporate the 8 week fermented Durio into concentrate containing PUFA for as much 10%, as a modification to the level of coffee husk fermentation by *P. ostreatus* (Badarina *et al*., 2014). The PUFA-concentrate compositions were 30% rice hulls, 30% roasted corn grain, 25% roasted soybean meal, 4% palm oil, 0.5% urea, 0.5% mix minerals, supplemented with 0.5% yeast, and 1.5% *Curcuma xanthorrhiza* Roxb, as modified from Sulistyowati *et al*. (2015).

Nutrient Analysis

Nutrient analyses of dry matter (DM), organic matter (OM), crude protein (CP), crude fiber (CF), and ether extract (EE) were determined according to AOAC (1990). While, NFE was calculated as 100% - (moisture + ash + EE + CP + CF). Gross energy (GE) was generated from samples using Bomb Calorymeter. The contents of NDF and ADF were analyzed using the method of Goering and Van Soest (1970).

Volatile fatty acid (VFA) analysis and *in vitro* dry matter digestibility (IVDMD) and *in vitro* organic matter digestibility (IVOMD) were prepared as if that of in hidrolytic post ruminal fermentation after two timed 24 hours using the method of Tilley and Terry (1963) as reported by Sulistyowati *et al.* (2014). Using gas chromatography (GC) Shimadzu 2010 series, fatty acid methyl esters (FAME) were obtained There are several calculations, total short chain fatty

acid (C4- C10), medium chain fatty acid (C12-C16), long chain fatty acid (C>16), mono unsaturated, poly unsaturated, saturated, unsaturated, ratio of PUFA/saturated, and unsaturated/saturated fatty acid (Schmidely *et al*., 2005).

Treatments and Experimental Design

This experiment was part of a research that has been splitted to several parameters as reported by Suciyanti *et al*. (2015) and Hartono *et al*. (2015), which then modified by combining with other feed ingredients to formulate a previously evaluated PUFA- concentrate (Sulistyowati *et al*., 2013, 2014, 2015). Experimental design in this research was Completely randomized design with four treatments of fermented Durio peel powder: F2; 2 weeks fermentation, F4: 4 weeks fermentation, F6: 6 weeks fermentation, and F8: 8 weeks fermentation and 4 replications.

Data were analyzed for variance, any significant differences were further analyzed using Duncan Multiple Range Test (DMRT) as described by Lentner and Bishop (1986). Based on nutrient content from the previous results as reported by Suciyanti *et al*. (2015), especially crude protein that was higher (6.4%) than the ones of shorter fermentation duration or even the control one, the fermented Durio peel powder of 8 weeks was selected to be incorporated into PUFA- concentrate which then compared to PUFA-concentrate containing non fermented Durio peel powder. These samples were then analyzed for their nutrient and fatty acid contents. The data were analyzed descriptively.

RESULTS AND DISCUSSION

VFA and GE Contents of *Durio zibethinus* Murr Peel Fermented with *P. ostreatus*

In vitro VFA and gross energy contents of *Durio zibethinus* Murr peel fermented with *Pleurotus ostreatus* are presented on Table 1. It showed that the longer fermentation (8 weeks) resulted in higher VFA but lower GE significantly compared to that of 2 week fermentation. These increasing VFA levels (from 105.37 mM in 2 week fermentation to 147.73 mm in 8 week fermentation) are in the same manner as the N-NH3 levels along with the longer duration of fermentation. Hartono *et al*. (2015) reported that the N-NH3 contents of fermented *Durio zibethinus* Murr peel with *P. ostreatus* increased from 6.94 mM (2 week fermentation) to 9.89 mM (8 week fermentation).

Table 1. *In vitro* VFA and gross energy content of *Durio zibethinus* Murr peel fermented with *Pleurotus ostreatus*

Variables	F2	F4	F6	F8	SEM	P
Total VFA, mM	105.37	136.84	91.40	147.73	26.35	NS
GE, cal/g	3.67[a]	3.57[b]	3.56[b]	3.59[b]	0.050	**

Note: F2: 2 weeks fermentation; F4: 4 weeks fermentation; F6: 6 weeks fermentation; F8: 8 weeks fermentation. NS: non significant; ** (P<0.01), very significant.

Other results showed that fermentation of coffee husk with *P. ostreatus* lowered VFA levels (173.79 mM in control decreased to 138.71 mM in 6% ration conataining this material) as reported by Badarina *et al.* (2014). The decreasing VFA of fermented coffee husk was also found by Xu *et al.* (2007).

These durations of fermentation showed effects that there were improvements in ruminally *in vitro* of *Durio zibethinus* Murr peel powder. These VFA and N-NH3 performances are within normal ranges of 80- 160 mm and 4- 12 mM, respectively (Sutardi, 1977).

However, these VFA levels showed opposite results of significantly ($P<0.05$) decreasing GE contents between 2 week and 8 week fermentation; eventhough quantitatively was very small amount (0.08 cal/g). While for the 4 and 6 weeks were remained the same as it was in 8 weeks of fermentation. Therefore, it could be stated that fermentation of Durio with *P. ostreatus* could be prolonged for up to 8 weeks as it was supported with higher VFA levels (105.37 mM in 2 weeks and 147.73 mM in 8 weeks).

Fractions of Fiber of *Durio zibethinus* Murr Peel Powder Fermented with *P. ostreatus*

In overall, fractions of fiber of all four treated fermented *Durio zibethinus* Murr peel powder fermented with *P. ostreatus* were not significantly different; however, the neutral detergent fiber (NDF) acid detergent fiber (ADF) tend to increase with the longer (in 2 and 8 weeks) the fermentation, as presented in Table 2. On the other hand, these fractions are decreased in 4 weeks of fermentation, then increased in 6 and 8 weeks of fermentation.

Table 2. Fractions of fiber of *Durio zibethinus* Murr peel fermented with *P. ostreatus*

Variables	F2	F4	F6	F8	SEM	P
NDF	75.49	71.29	75.13	77.19	2.49	NS
Hemicelulose	8.04	8.30	6.76	9.17	1.00	NS
ADF	64.84	63.17	68.43	67.93	2.51	NS

Note: F2: 2 weeks fermentation; F4: 4 weeks fermentation; F6: 6 weeks fermentation; F8: 8 weeks fermentation.
NS: non significant

The improved NDF is an indicator of the loosen fiber due to the work of *P. ostreaatus*; while, at the same time the ADF is increasing with the longer fermentation in 6 and 8 weeks maybe to the the higher fiber content of lignin binding of the michelium of *P. ostreaatus* itself. Suggesting that with 6 weeks of fermentation of *Durio zibethinus* Murr peel powder fermented with *P. ostreatus* is the optimal time in improving fiber quality (75.13% NDF and 68.43% ADF). Compared to other results of concentrate containing polyunsaturated fatty acid (PUFA- concentrate) that showed the NDF reduced (72.09% to 68.82%) with proceed time (2 to 6 weeks) of storage, respectively; while the ADF increased (24.78% to 30.76%) significantly with the longer the storage (2 to 4 weeks),

respectively as reported by Sulistyowati *et al*., 2015). Based on these two results, it is suggested that for ration containing high fraction of fiber, it needs fermentation with fungi for 6 weeks; while, for ration containing concentrate, lower fiber content, could be stored for the length of 2-6 weeks.

Nutrient contents of PUFA- concentrate containing *Durio zibethinus* Murr peel powder fermented with *Pleurotus ostreatus*

Nutrients of PUFA- concentrates of non fermented and 8 week fermented PUFA- concentrate containing *Durio zibethinus* Murr peel powder fermented with *Pleurotus ostreatus* are presented in Table 3. Moisture, dry matter and ash contents of these two rations are quantitatively unchanged. However, organic matter, crude protein, ether extract and fiber fractions are improved. These nutrients are incresed quantitaively; while the NDF and ADF are decreased in fermented ration. These improved nutrients suggested that fermentation with *P. osetreatus* works well in breakdown lignin binding in Durio peel powder so that some nutrients available for the growth of the fungi which then improved the overall quality of of its nutrients. While, gross energy is remained relatively the same.

Table 3. Nutrient contents of PUFA- concentrate containing *Durio zibethinus* Murr peel powder fermented with *Pleurotus ostreatus*

Variables	F0-PUFA	F8-PUFA	SEM
Moisture, %	6.32	6.35	0.02
Dry matter, %	93.69	93.66	0.02
Ash, %	7.94	7.44	0.35
Organic matter, %	85.75	86.22	0.33
Crude protein, %	13.17	15.65	1.75
Ether extract, %	11.74	15.30	2.52
Crude fiber, %	7.87	1.40	4.57
NFE, %	52.43	46.88	3.93
NDF, %	41.90	27.71	10.03
ADF, %	14.21	13.98	0.16
Gross energy, cal/g	4.42	4.53	0.07

Note: F0- PUFA: PUFA- Concentrate containing non fermented Durio; F8- PUFA: PUFA- Concentrate containing 8 weeks fermented Durio.

Sulistyowati *et al.* (2015) reported that PUFA- concentrate supplemented with yeast and *C. xanthorrhiza* Roxb (PCM) seemed to remain stable in moisture, dry matter, ash, and organic matter composition during the storage of 2, 4, and 6 weeks. Other study showed that basal diet for dairy cows in which 56g XP yeast incorporated, was having 31% NDF and 20.4% ADF (Hristov *et al.* 2010). Compared to our data, the NDF and ADF are lower.

Fatty acid contents of PUFA- concentrate containing *Durio zibethinus* Murr peel fermented with *Pleurotus ostreatus*

Fatty acid profile of PUFA-concentrate containing *Durio zibethinus* Murr peel powder fermented with *Pleurotus ostreatus* is presented in Table 4. Fat content of PUFA- concentrate with fermented Durio is higher (2.32%) than that of non fermented one. However, total fatty is lower in PUFA- concentrate with fermented Durio is lower (2.91%). All variables of fatty acid (SCFA, MedCFA, LongCFA, MUFA, PUFA, unsaturated, saturated, U/S, P/S) in PUFA- concentrate containing *Durio zibethinus* Murr peel powder fermented with *Pleurotus ostreatus* is lowered compared to that of PUFA- concentrate containing non fermented Durio. This fatty acid profile is on the opposite of the fatty acid content of PUFA- concentrate with supplementation of either yeast, curcuma or mixture of both that are higher in all variable of fatty acids in 2 weeks of storage (Sulistyowati *et al*., 2015).

Table 4. Fatty acid contents of PUFA- concentrate containing *Durio zibethinus* Murr peel powderbfermented with *Pleurotus ostreatus*

Variables	Pecentage (% w/w)	
	F0-PUFA	F8-PUFA
Fat Content	13.37	15.69
Fatty Acid (% of fat)		
Caprilic Acid, C8:0	n.d	0.09
Capric Acid, C10:0	n.d	0.06
Lauric Acid, C12:0	0.05	0.51
Myristic Acid,C14:0	0.22	0.40
Palmitic acid, C16:0	10.58	11.83
Palmitoleic Acid, C16:1	0.10	0.09
Heptadecanoic Acid, C17:1	0.05	0.05
Cis-10- Heptadecanoic Acid, C17:1	0.03	0.02
Stearic Acid, C18:0	1.75	1.81
Elaidic Acid, C18:1n9t	0.03	0.02
Oleic Acid, C18:1n9c	25.10	25.04
Linoleic Acid, C18:2n6c	30.21	26.44
Arachidic Acid, C20:0	0.20	0.20
y- Linolenic Acid, C18:3n6	0.02	0.02
Cis- 11- Eicosenoic Acid, C20:1	2.70	2.28
Cis- 11. 14- Eicosedienoic Acid, C20:2	0.07	0.05
Fatty Acid Total	71.09	68.90
Total SCFA (C4- C10)	nd	0.15
Total MedCFA (C12- C16)	10.68	11.92
Total LongCFA (C>C16)	60.16	55.93
Total MUFA	28.01	27.5
Total PUFA(P)	30.30	26.51
Total Saturated fat (S)	12.78	14.89
Total Unsaturated (U)	58.31	54.01
Ratio - U/S	4.56	3.63
Ratio- P/S	2.37	1.78

Note: F0- PUFA: PUFA- Concentrate containing non fermented Durio; F8- PUFA: PUFA- Concentrate containing 8 weeks fermented Durio.

The ratios of P/S were 2.37 and 1.78 of PUFA- concentrate containing non fermented Durio peel and containing fermented Durio peel, respectively are in contrast to that of the diets containing saturated fat and unsaturated fat as reported by Harvatine and Allen (2006). Therefore, the type of fat supplemented in concentrate would determine its type of fatty acid. There is interdependency of different PUFA, any other vegetable oil as supplement would respond differently in its fatty acid content (Chilliard *et al.*, 2006).

CONCLUSION

Fermentation of *Durio zibethinus* Murr peel powder fermented with *Pleurotus ostreatus* for 4-6 weeks seems to improve NDF, ADF, and GE ; while, fermentation for 8 weeks and incorporated into PUFA-concentrate could improve crude protein, ether extract and NDF, and lower ADF. However, it decreased fatty acid profile of SCFA, MedCFA, LongCFA, MUFA, PUFA, unsaturated, saturated, P/S, and U/S.

REFERENCES

Badarina, I., D. Evvyernie, T. Toharmat, and E.N. Herliyana. 2014. Rumen fermentability and *in vitro* digestibility of diet supplemented with coffee husk fermented with *Pleurotus ostreatus*. (In Indonesian). Jurnal Sain Peternakan Indonesia 9 (2): 102-109.

Chilliard, Y., J. Rouel, A. Ferlay, L. Bernard, P. Gaborit, K. Raynal-Ljutovac, A. Lauret, and C. Leroux. 2006. Optimising goat milk and cheese fatty acid composition. Pages 281- 282. In Improving the Fat Content of Foods. C. Williams and J. Buttriss, ed. Woodhead Publishing Limited, Cambridge, UK.

Goering, H.K. and P.J. Van Soest. 1970. Forage fiber analysis. Agricultural Handbook No: 379. USDA. Agricultural Research Service. Washington DC, pp. 1-9.

Hartono, R., Y. Fenita, dan E. Sulistyowati. *In vitro* dry matter digestibility, organic matter digestibility, and N-NH3 production of *Durio zibethinus* fermented with white rod fungi (*Pleurotus ostreatus*) incubated in different time. (In Indonesian). Jurnal Sain Peternakan Indonesia 10 (2): 87- 94.

Harvatine, K.J. and M.S. Allen. 2006. Effects of fatty acid supplements on milk yield and energy balance of lactating dairy cows. J Dairy Sci. 89: 1081- 1091.

Hatakka, A. 1994. Lignin-modifying Enzymes from Selected White-rot Fungi. Production and Role in Lignin Degradation. FEMS Microbiol. Rev. 13: 125-135.

Hristov, A.N., G.Varga, T. Cassidy, M. Long, K. Heyler, S.K.R. Karnati, B. Corl, C.J. Hovde, and I.Yoon.

2010. Effect of *Saccharomyces cerevisiae* fermentation product on ruminal fermentation and nutrient utilization in dairy cows. J Dairy Sci. 93 :682–692.

Lentner, M. and T. Bishop. 1986. Experimental Design and Analysis. Valley Book Co. VA

Sangadji, I., A. Parakkasi, K. G. Wiryawan, B. Haryanto. 2008. Perubahan Nilai Nutrisi Ampas Sagu Selam pada Fase Pertumbuhan Jamur Tiram Putih. Jurnal Ilmu Ternak. 8 (1): 31-34.

Schmidely, P., P. Morand- Fehr, and D. Sauvant. 2005. Influence of extruded soybeans with or without bicarbonate on milk performance and fatty acid composition on goat milk. J Dairy Sci. 88: 757- 765.

Suciyanti, H., E. Sulistyowati, dan Y. Fenita. Nutrient evaluation of *Durio zibethinus* fermented with white rod fungi (*Pleurotus ostreatus*) incubated in different time. (In Indonesian). Jurnal Sain Peternakan Indonesia. 10 (2): 77-86.

Suhaidi, I. 2004. Pemanfaatan Limbah Biji Durian sebagai Bahan Pakan Ternak Ayam Pedaging. Thesis. Program Pascasarjana. Universitas Sumatera Utara. Medan. http://www.repository.usu.ac.id/bitstream/123456789/6603/1/05000574.pdf. Diakses tanggal 10 Desember 2014.

Sulistyowati, E., A. Sudarman, K. G. Wiryawan, and T. Toharmat. 2013. Quality of milk fatty acid during late lactation in dairy goat fed on PUFA-diet supplemented with yeast and *C. xanthorrhiza* Roxb. J. of the Indonesian Trop. Anim. Agric. 38 (4): 247- 256.

Sulistyowati, E., A. Sudarman, K. G. Wiryawan, and T. Toharmat. 2014. *In vitro* goat fermentation of PUFA-diet supplemented with yeast and *Curcuma xanthorrhiza* Roxb. Media Peternakan. 37(3) : 175-181.

Sulistyowati, E., A. Sudarman, K. G. Wiryawan, and T. Toharmat. 2015. The nutritive values of PUFA-concentrate supplemented with yeast and *Curcuma xanthorrhiza* Roxb stored in several weeks. J. of the Indonesian Trop. Anim. Agric. 40 (1): 11-22.

Sutardi, T. 1977. Ikhtisar Ruminologi. Bahan Penataran. Kursus Peternakan Sapi Perah Kayu Ambon, Lembang. BPPLP- Ditjen Peternakan- FAO.

Tilley, J.M.A. and R.A. Terry. 1963. A two stage technique for the *in vivo* digestion of forage crops. J. of the British Grassland Society. 18: 104-111.

Xu, C.C., Y. Cai, J.G. Zhang, and M. Ogawa. 2007. Fermentation quality and nutritive value of total mixed ration silage containing coffee ground at ten or twenty percent of dry matter. J. Anim. Sci. 85: 1024-1029.