EFFECT OF DIETARY LEVELS OF NEUTRAL DETERGENT FIBER (NDF) ON *IN VITRO* ORGANIC MATTER AND NDF DIGESTIBILITY WITH RUMEN FLUID OF BEEF CATTLE AS AN INOCULUM SOURCE

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ABSTRACT

This experiment aimed to evaluate the effect of dietary NDF levels on *in vitro* feed digestion for further *in vivo* and performance studies. It was arranged according to the complete randomized design with 6 treatments and 3 replications. The treatments were 35, 41, 47, 53, 59 and 65% NDF corresponding to NDF35, NDF41, NDF47, NDF53, NDF59 and NDF65 treatment. The basal substrates (mixture) for the experiment included elephant grass (*Pennisetum purpureum*), *Operculia turpethum*, rice straw, broken rice, soybean extraction meal and urea with the fixed crude protein of 14.6 % (DM basis) for all the treatments. The *in vitro* DM and NDF digestibility were observed from 0 to 72 h by the method of Goering and Van Soest.

Results showed that organic matter digestibility (OMD) was significantly different ($P<0.05$) among the treatments at 24, 48 and 72 hincubation. At 72 h the *in vitro* OMD values of NDF35 (85.1%) and NDF41 (82.7%) treatments were significantly higher ($P<0.05$) than those of other treatments. The data also demonstrated that increasing the NDF levels in diets from 35 to 65% gradually reduced NDF digestibility (NDFD). The linear relationship between NDF levels (%) of the treatments and the NDFD was found with the function $y = -0.274x + 69.5$ and $R^2 = 0.683$, while this between NDF levels (%) and the *in vitro* OMD was the function $y=-0.576x + 105$ and $R^2 = 0.954$. In conclusion, increasing the NDF level from 35.0% to 65.0% in the mixture gradually decreased *in vitro* OM and NDF digestibility, and the reasonable treatments from 47 to 59% NDF could be selected for the coming *in vivo* and performance studies.

Keywords: *NDF levels, OMD digestion, mixture, rumen microbes, ruminants.*

INTRODUCTION

In Vietnam beef cattle are popularly fed by local roughage sources due to the low cost and availability, while theycan beutilized well by ruminants to convert into energy by rumen microorganisms. The neutral detergent fiber (NDF) is considered as an indicator of quality assessment of ruminant feed (Mertens, 2014). The NDF content offiber feedsis often very variable (Danh Mo and Nguyen Van Thu, 2008) and increasing amount of NDF in the diet was reduced the nutrient digestibility in ruminants (Vu Chi Cuong et al., 2009 and Danh Mo, 2009), however more utilization of fiber feeds by the advantage of rumen function could be increasingly benefited in environment and economy by the ruminant producers. Rahman et al. (2009) reported that when the digestibility of NDF in diets improved, the live weight gain of fattening cattle was increased. Moreover, Arelovich et al. (2008) also indicated that NDF intake had a closed relationship with dry matter intake and net energy for growth. In Vietnam, however, studies on NDF levels in diets of beef cattle have been still limited. Therefore, the objective of this study was to find a reasonable dietary NDF levels based on *in vitro* OM and NDF digestibilities for further studies on beef cattle to improve the producers' benefits.

MATERIALS AND METHODS

Location and time

The experiment was conducted from March 2019 to May 2019 at the E205 laboratory, Department of Animal Husbandry, College of Agriculture of Can Tho University.

Experimental design, feeds and feeding

This experiment was arranged in completely randomized design with 6 treatments and 3 replicates. The treatments were NDF35, NDF41, NDF47, NDF53, NDF59 and NDF65 corresponding to 35, 41, 47, 53, 59 and 65% NDF in the substrates, which were based on the studies of Arelovich et al. (2008); Konka et al. (2015) and Filho et al. (2016). Ingredients and chemical composition of treatments was showed in Table 1.

Table 1. Ingredient and chemical composition of treatments (%)

Item	Ingredient	NDF35	NDF41	NDF47	NDF53	NDF59	NDF65
Ingredient composition (%DM)	Elep. grass at 15 days	4.88	-	-	4.84	-	-
	Elep. grass at 25 days	-	24.8	-	-	-	-
	Elep. grass at 65 days	-	-	42.0	56.4	74.7	87.9
	O. turpethum(50cm)	39.1	27.3	24.9	7.95	-	-
	O. turpethum(301-350cm)	30.2	21.6	5.14	3.00	1.49	1.49
	Rice straw	6.09	6.57	8.08	8.05	8.03	6.08
	Broken rice	14.7	11.7	10.7	6.79	2.51	1.06
	Soybean extr. meal	4.87	7.76	8.72	12.6	12.5	1.34
	Urea	0.22	0.23	0.40	0.45	0.80	2.14
	Total	100	100	100	100	100	100
Chemical composition	DM	92.8	93.1	93.2	93.5	93.8	94.4
	OM	94.7	94.5	94.2	94.0	93.9	93.3
	CP	14.6	14.6	14.6	14.6	14.6	14.6
	EE	3.40	2.84	2.50	2.13	1.91	1.96
	CF	20.2	22.3	24.9	26.6	29.9	33.4
	NDF	35.0	41.0	47.0	53.0	59.0	65.0
	ADF	29.4	31.1	33.5	35.4	38.5	42.4
	NFE	57.0	55.4	53.3	52.0	49.8	49.6
	ME, Kcal	2.358	2.323	2.273	2.241	2.182	2.045

Note: DM: dry matter, OM: organic matter, CP: crude protein, CF: crude fiber, EE: ether extracted, NDF: neutral detergent fiber, ADF: acid detergent fiber, NFE: nitrogen free extract, ME: metabolizable energy. NDF35, NDF41, NDF47, NDF53, NDF59 and NDF65 contained neutral detergent fiber at 35, 41, 47, 53, 59 and 65 based on dry matter

Elephant grass *(Pennisetum purpureum),Operculia turpethum*, rice straw and concentrate feed (including broken rice and soybean extraction meal) were mixed with different ratio according the treatments.The concentrate was formulated from broken rice and soybean extraction meal and the basal substrates (mixture) as the treatments, which had the same level of 14.6% CP (DM basis) and urea was used to fix CP level in different treatments.

To meet the requirement of the treatment NDF levels, elephant grass at the age of 15, 25 and 65 days; and different length of *Operculia turpethum* vines were used to formulate (Table 1). While *Operculia turpethum* vines were collected with the length from the top of 50cm and the length from 301cm to 350cm, which were shown in Figures 1 and 2. In Table 1 also indicated that from the treatment NDF47 to NDF67 were more comfortable to formulate the diets due to the availability of the ingredients than the NDF35 and NDF41 treatments.

Figure 1. *Operculia turpethum* vines 50cm

Figure 2. *Operculia turpethum* vines 301-350cm

Measurements taken

Chemical composition. Including dry matter (DM), organic matter (OM), crude protein (CP), crude fiber (CF) and ether extracted (EE) which were determined following the Association of Official Analytical Chemists methods (AOAC, 1990). While neutral detergent fiber (NDF) and acid detergent fibers (ADF) were determined according to Van Soest et al. (1991) and metabolizable energy (ME) was estimated by the formula of Abate and Mayer (1997), forages: ME (MJ/kgDM) = 20,27 – 0,1431CF – 0,1110NFE – 0,2200Ash and concentrates: ME (MJ/kgDM) = − 4,80 + 0,6004CF – $0,0640CF^2$ + $0,0015CF^3$ + 1,1572NFE – $0,0236NFE^2$ + $0,00014NFE^3$.

*In vitro digestibility of OM and NDF.*Determining at 0, 12, 24, 48 and 72 hours based on the description of Goering and Van Soest (1970) and modified by Nguyen Van Thu and Peter Udén (2003). They were calculated by following formulas:

$OMD\% = 100- [(W_{dried}-W_{burned})/(P_{sample}\times DM\times OM)]\times 100$ (1)

$NDFD\% = 100- [(W_{dried}-W_{burned})/(W_{sample}\times DM\times NDF)]\times 100$ (2)

(W_{dried}: sample weight after drying, W_{burned}: sample weight after burning, W_{sample}: sample weight)

Statistical analysis

The data were calculated by using the Excel software and then analyzed variance by using the ANOVA of General Linear Model (GLM) of Minitab Reference Manual Release 16.1 (Minitab, 2010). When the paired comparison of two treatments evaluated the Tukey test of the Minitab was used.

RESULTS AND DISCUSSION

Chemical composition of feeds

Chemical composition of the feeds as ingredients was shown in Table 2. CP and ME content of elephant grass at 15 days were 11.5% and 2,311 kcal/ kg DM, respectively. While at 25 and 65 days they were 9.85% and 2,141 kcal/kgDM and 8.34% and 2,076 kcal/kgDM, respectively. Their NDF contents were 57.4, 64.6 and 68.4%, respectively. The results agreed with those of Vu Chi Cuong et al. (2010) found in elephant grass at 35 to 50 days being 68.1 and 74.1%, respectively).

Table 2. Chemical composition (%) of feeds used in the experiment

Feed, %DM	DM	OM	CP	EE	NFE	CF	NDF	ADF	Hemi	ME, kcal/DM
Elep. grass at 15 days	94.3	95.7	11.5	2.73	62.6	18.9	57.4	35.3	22.1	2,311
Elep.grass at 25 days	94.8	95.5	9.85	1.66	53.1	30.9	64.6	40.7	23.9	2,141
Elep. Grass at 65 days	94.5	95.5	8.34	1.99	50.0	35.2	68.4	44.2	24.2	2,076
O. turpethum(50cm)	93.0	95.8	18.1	4.07	50.5	22.7	35.5	33.3	2.20	2,770
*O. turpethum (*301-350 cm	93.5	96.5	9.19	4.18	56.2	27.0	42.1	36.8	5.30	2,247
Rice straw	94.1	95.8	4.99	1.94	56.3	32.6	64.8	46.9	17.9	2,015
Broken rice	90.6	90.0	8.33	1.57	79.5	0.60	6.23	1.37	4.86	2,187
Soybean Ext. meal	90.3	89.9	42.4	1.34	41.3	4.85	15.6	9.58	6.02	3,049
Urea	100	-	288	-	-	-	-	-	-	-

The chemical compositions of 2 types of *Operculia turpethum* vines (50cm length and from 301-305cm used in the present study were presented in Table 2. The results were similar to those reported by Nguyen Thi Vinh Chau and Nguyen Van Thu (2014) in which *Operculia turpethum* vines with 1.5m length had CP and NDF content being 14.2 and 39.0%, respectively. NDF content of rice straw in this experiment was 64.8% and it was consistent with the report of Nguyen Van Thu and Nguyen Thi Kim Dong (2011) being 64.9%.

In vitro **organic matter digestibility (OMD)**

The *in vitro* organic matter digestibility of treatments over incubation times was showed in Table 3.

Table 3. *In vitro* OMD values (%) of treatments over incubation times

Incubation time (h)	NDF35	NDF41	NDF47	NDF53	NDF59	NDF65	P	SE
0	62.9^{a}	55.1^{b}	52.7^{b}	41.2^{c}	37.6^{c}	31.7^{d}	0.001	0.904
12	74.1^{a}	69.2^{b}	64.0^{c}	59.1^{d}	52.3^{e}	48.5^{f}	0.001	0.773
24	79.2^{a}	76.4^{a}	68.0^{b}	64.8^{c}	63.2^{c}	58.0^{d}	0.001	0.596
48	85.8^{a}	81.0^{b}	73.5^{c}	70.7^{d}	68.0^{e}	64.3^{f}	0.001	0.462
72	85.1^{a}	82.7^{a}	76.2^{b}	73.8^{bc}	71.4^{c}	68.2^{d}	0.001	0.578

Note: $^{a, b, c, d, e, f}$ values with different superscript letters within one row are significantly different at the level of 5%.

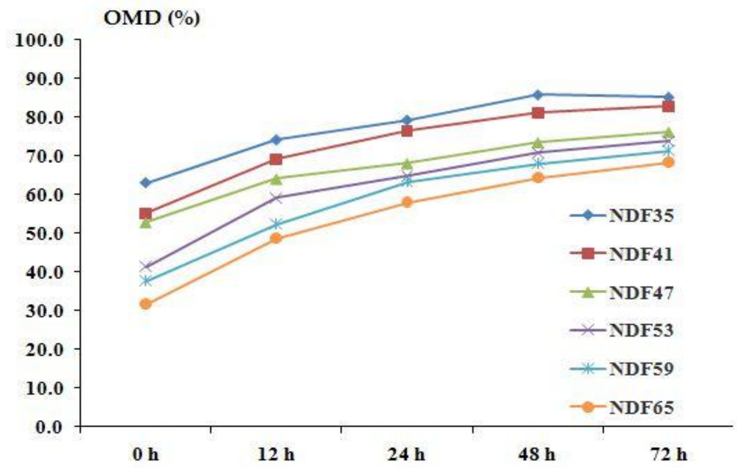

Figure 3. *In vitro* OM digestibility of treatments over incubation times

In Table 3 and Fig. 3 showed that the OMD values over incubation times were significantly different (P<0.01) among the treatments and they were gradually reduced from the NDF35 to NDF65 treatment with the highest values for the NDF35 treatment and the lowest values for the NDF65 treatment over incubation times. Results of the present study agreed with those of the previous trial of when NDF in diet increased from 64.6% to 76.6% in *in vitro* experiment using Rye grass and concentrate feed reported by Mpemba et al. (2018) that the OMD had reduced from 33.3 to 41.9% (76.6 – 64.6% NDF in experiment). Particularly at 72 h the accumulated OMD values were significantly higher for the NDF35 (85.1%) and NDF41 (82.7%) compared toother treatments, while these were 76.2, 73.8 and 71.4% for the NDF47, NDF53 and NDF59 treatment.

Relationship between NDF levels and OM digestibility of the treatments at 72 h

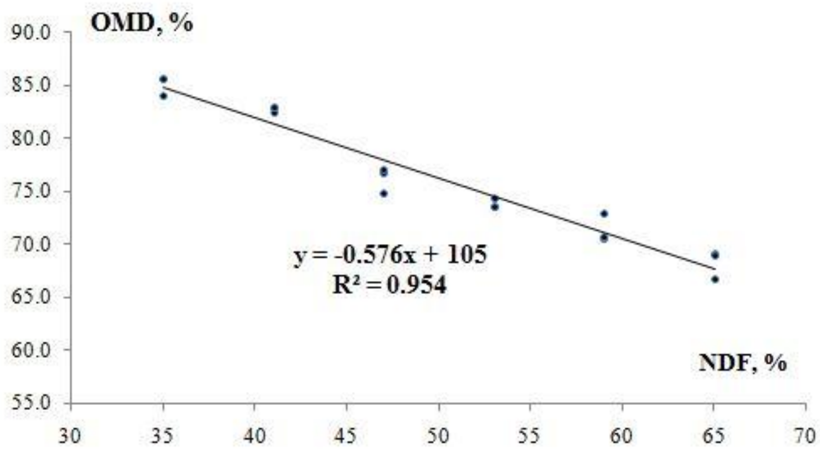

Figure 4. Relationship between NDF levels and OM digestibility

In Figure 4 confirmed that there was a close linear relationship between NDF (%) and the *in vitro* OMD with the function y=-0.576x + 105 and R^2 = 0.954 and when increasing the NDF (%) in the mixture the OMD was gradually reduced. Thus the reduction of OMD indicated that there was an great effect of the NDF increase (%) on the *in vitro* digestion for other nutrients of the mixture such as CP, EE, NFE, etc.

In vitro neutral detergent fiber digestibility (NDFD)

The NDF digestibility of different treatments over time were shown in the Table 4 and Fig.5

Table 4. *In vitro* NDFD values (%) of treatments over incubation times (%)

Periods (hour)	NDF35	NDF41	NDF47	NDF53	NDF59	NDF65	P	SE
0	0.28	0.10	1.09	0.08	0.86	1.07	0.995	2.126
12	29.9	29.1	28.0	27.7	26.0	24.2	0.091	1.325
24	45.6^{a}	43.7^{ab}	41.6^{abc}	39.6^{bcd}	37.8^{cd}	36.1^{d}	0.001	1.118
48	61.5^{a}	56.3^{b}	49.1^{c}	48.7^{c}	48.3^{c}	47.0^{c}	0.000	0.835
72	60.3^{a}	59.8^{a}	54.5^{b}	54.3^{b}	53.8^{b}	52.4^{b}	0.001	1.074

Note: $^{a, b, c, d}$ values with different superscript letters within one row are significantly different at the level of 5%.

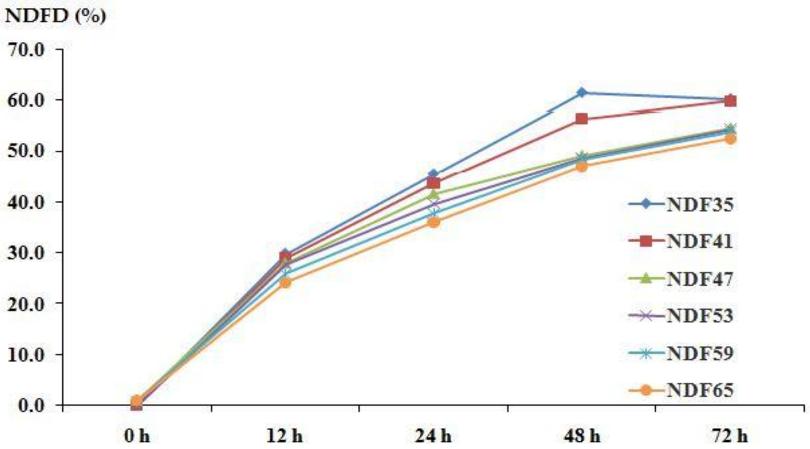

Figure 5. *In vitro* NDF digestibility of treatments over incubation times

There were not significantly different (P>0.05) in of NDFD among the treatments at 0 and 12 hour (Table 4). From 24 to 72 h The NDFD values were significantly different among treatments (P<0.05) and at 72 h the accumulated NDFD values (%) of NDF35 (60.3) and NDF41(59.8) were significantly higher than the rest of treatments and the NDFD of NDF47, NDF53, NDF59 and NDF65 were similar (P>0.05), despite there was a slightly gradual reduction (Table 4 and Fig. 5). Marid et al. (2002) also found that diets contained low NDF; the NDFD values were increased slowly from 48 to 72 hour.

Relationship between NDF levels and NDF digestibility of the treatments at 72 h

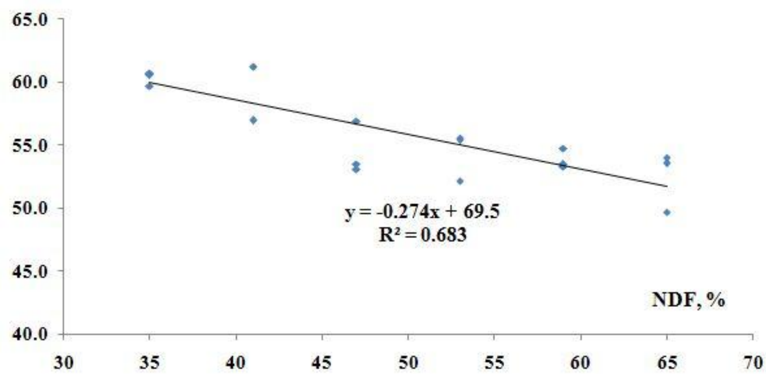

Figure 6. Relationship between NDF level (%) and NDFD

The linear relationship between NDF levels of the treatments and the NDFD was found with the function y = -0.274x + 69.5, however the R^2 = 0.683 was much lower than that of the OMD (R^2 = 0.954) in the present study. Thus this indicated that there was a limittation of only the NDF level of treatment effects on the NDFD.

Harper and McNeill (2015) reported that low NDF content in beef diet may be difficult to achieve in the tropic and subtropics due to restriction on nutritive value and genetics of lower beef growth, higher NDF content in the diet can be considered more feasible target in tropical cattle system. According to Nguyen Binh Truong and Nguyen Van Thu (2019) found that NDF content of diets from 49.7 to 57% with 10.7-27.8% concentrate supplementation for beef cattle production raised by local producers in An Giang province. It was also indicated that formulating NDF35, NDF41, NDF47, NDF53, NDF59 and NDF65 diets following the costs (with the prices in 2020) were 1.181; 1.198; 1.162; 1.061 and 785 VND/kg, respectively. Thus based on the results of OMD and NDFD, availability of feed resources, the dietary costs and more fiber feeds utilized by beef cattle, the NDF treatments from 47 to 59% could be reasonable for further studies to find out the optimum NDF levels of the beef cattle diets.

CONCLUSIONS

It was concluded that increasing the NDF level from 35.0% to 65.0% in the mixture gradually decreased *in vitro* organic matter and neutral detergent fiber digestibility; however there was a closer linear relationship between NDF levels and *in vitro* OM digestibility. The reasonable treatments from 47 to 59% NDF could be selected for *in vivo* and performance studies.

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